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HISTOLOGICAL CRITERIA IN OSTEOPOROSIS

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Abstract: *Osteoporosis is a first line concern in the fight with suffering and death. Considered a silent epidemic, osteoporosis and especially its complications have devastating consequences on quality of life, health and not least on health budgets. Histological study of muscle and bone in the same patient shows a clear lesion parallelism, proving, that some of the blame for the production of the fracture falls upon the muscle and bone.*

Keywords: *osteoporosis, hyaline degeneration of muscle fibre, lipid vacuoles.*

1. Introduction

When the bone strength is lower, the fracture occurs in minor injuries. The most frequent cause for a hip fracture is falling from the same level. A football player for example, suffers in a single match, 20-25 trauma injuries by falling and 40-45 trauma injuries by direct impact, without suffering fractures; a patient with osteoporosis suffers a hip fracture at the first injury by falling. This example shows the importance of the muscle mass in protection against fractures. [3, 6, 10].

Histological study of muscle and bone on the same patient shows a clear lesion parallelism, proving, that some of the blame for the production of the fracture falls upon the muscle and bone. [1, 11, 12].

We can anticipate that osteoporosis with low bone strength is not strictly related to chronological age, but especially to the degree of use of the musculoskeletal system; the more a person moves, it maintains its muscle tone through physical exercises adjusted with the biological conditions, the more its muscle tone, muscle mass, bone mass and bone strength remain within protection limits against the risk of fracture. The importance of muscle/ bone lesion parallelism and the existence of concrete evidence could change more or less the managerial aspects of osteoporosis in all sections of it (prevention, investigation, treatment, complications, and treatment of complications). [8, 9]. Bone is not an inert organ; it has an intense histological dynamic, so that after 120 days the bone remodelling is considered complete, renewing cycle of the cells functioning optimal in the first decade of life. [7]. The main problem of bone tissue is to supply the cell population with nutrients, in the conditions in which these substances can not diffuse freely through the bone. A circular disposition of cells around the central channel creates an efficient supply geometrical system, for supplying a maximum quantity of tissue with a minimum number of vessels. The muscle near the bone is the contractile, effectors organ, but not only that; the muscle near the bone becomes a circulatory pump, which, through its

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contraction, contributes to the bone vasculature. Therefore, it is particularly important, the histological analysis of the muscle not only of the bone, in osteoporosis. [2, 4, 5].

2. Method and Material

From the group of 18 patients, were prepared fragments of muscle and fragments of bone, which were examined under the optical microscope and electron microscope. For optical microscope were prepared slides from hematoxylin-eosin staining, aniline blue, Van Gieson's method.

For examination under transmission electronic microscope, the processing protocol of biological samples was the following:Fixation in 2% glutaraldehyde solution + 2% formaldehyde in 0.15 M Na cocodilat buffer, pH 7.2-7.4

b. Wash in buffer Na cocodilat 0.15 2 / 10 min. at pH 7.2-7.4
c. Post-fixation in osmium tetroxide 1% - 90 min.
d. Wash in distilled water 2 / 10 min.
e. Dehydration in solutions with increasing concentrations of alcohol.
f. Clarification in propylene oxide.
g. Impregnation in epoxy resins.
h. Inclusion and polymerization 48 h at 60 degrees C.
i. Slicing at the ultramicrotome.
j. Contrasting with uronil acetate and lead citrate.
k. Examination under an electron microscope (Philips CM 12).

3. Results and Discussions

In light of optical and electronic microscopic studies of muscle tissue and bone tissue, we can conclude that the lesion parallelism of the binomial bone - muscle is obvious.

The muscle shows significant structural changes observed both, by optical microscope, through conventional techniques (HE, Van Gieson's method), or histochemical (NADH2 - cytochrome - C - reductase, acetylcholinesterase), and by electronic microscopic exam.

Classical techniques show at the optical microscope, the constant appearance into the striated muscle fibres, of clear lipid vacuoles, different sizes, arranged either isolated or crowded in groups with variable numbers of vesicles, either in the monomial.

The groups of lipid vacuoles, the linear, are located in ectosarcoplasma in close proximity of sarcolemma. Isolated lipid vacuoles are intrasarcoplasmatic dispersed among myofibril and monofilaments.

The form of the muscle fibre is different: oval, round, rarely angular, but all having an irregular shape.

The sizes of myocytes are unequal and slightly hypertrophied, frequently hyperplastic, with aspects of hyaline degeneration. It can be noticed fibres with clear "central core" or moth-eaten muscle fibres (with moth bites "Moth - eaten" - Brooke and Engel).

In some muscle fibres it appears lipofuscin pigment.

Histochemical studied, the muscle highlights, especially through the action of NADH2 - cytochrome - C – reductase, the aspects of "moth bites"; the reaction is intense in some fibres, low in others, or with central active area, or clear lizere subsarcolemmal.

Striated muscle fibres are partially or totally surrounded by weakly positive cholinergic nerves (neuro-muscular relation, modified in deficiency).

Interstitials between the striated muscle fibres appear as different sized, in the smaller ones there is less conjunctival matrix, but, with the growing, are gradually added the capillaries, arterioles and connective cells (fibroblasts rare, rare macrophages). (Fig. 1, 2).

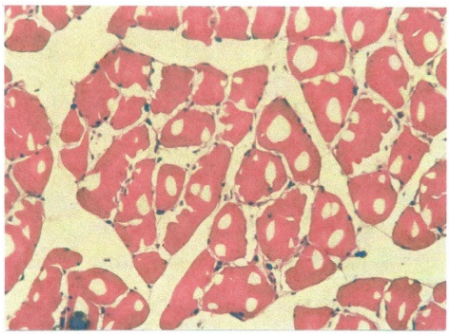

Fig. 1. *Cross section*.
Striated muscle fibres with an irregular shape (oval, round, rarely angular) with an aspect of hyaline degeneration, which presents 1-3 clear vacuoles, of different sizes, in the sarcoplasma,. Around each muscle fibre and blood capillaries are observed very rare connective cells. Col. HE/140

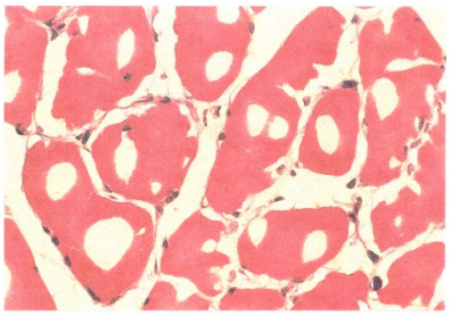

Fig. 2. *Muscle cross section*.
Larger image of an area from Fig. 1. It can be observed the clear vacuolar areas, uniform, with different sizes, inside of striated muscle fibres (mild hypertrophy of fibres, some looking like moth bites), a moderate hyalinisation, sanguine capillaries and rare connective tissue cells migrated into the interfibrilar area. In the muscle fibres sarcoplasma are present microvesicles slightly hypertrophied. At some myocytes are observed, in addition, small sarcolemmal, invagination described as the "moth bites";. CoI.HE/540

At the electronic microscope it appears frequently intrasarcoplasmatic lipid vacuoles, of different sizes, under three aspects: isolated, in groups, monomial, the last two, located in ectosarcoplasma near sarcolemma.

The mitochondria appear like balloons, with tight cristae, the organelles appear to be crowded by lipid vacuoles. Mitochondria in some muscle fibres are presented in the two extremes of their functional cycle, respectively condensation and ballooning, and it can be considered as being in good activity.

Rarely, in some muscle fibres it may be seen fragments of the Z line and the membrane H.

Triads are apparently normal shape in most muscle bundles, what shows that there appear rather pronounced deterioration that occurs in some myocytes structures having as a consequence the altered contraction of muscle fibre, fact supported by the changes in neuromuscular relationship, observed through acetylcholinesterase reaction.

Both the sarcolemma of striated muscle fibre and core nucleolema appear strongly sinuous and, from place to place, with very deep invaginations. On relatively large areas sarcolemma appears with aspects different than normal.

Interstitials between the muscle fibres and bundles contain extracellular matrix in amount proportional to their size, in the larger spaces are present capillaries, and then with enlargement, are added arterioles and connective cells.

Connective cells are represented by fibroblasts well equipped with organelles through active macrophages located near sarcolemma, bordered, inside the cell, by the clear vacuoles chain, or by the groups formed by it.

The presence of macrophages near these myocytes areas, brings immediately to attention the fact that in those areas tends to occur in intense activity of phages, cleanup and lipid removal. On one hand, the fibre muscle that tends to marginalize and eliminate them, on the other hand, the macrophage which destroys them through lysosomal enzymes, in large amounts in the cell.

Evidence of intense macrophage function are the frequency multivezicules corpora present in the cell cytoplasm.

Occasionally in the interfibrilar spaces can still be surprised a mast, with granular loaded cytoplasm and many, very long microvili. (Fig.3, 4).

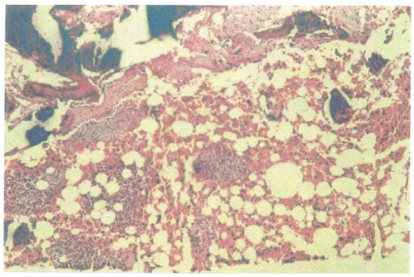

Fig. 3. *Section of the femoral head and spinal canal by bone. There can be observed fragments of bone with osteoid aspect, with disorganized slides and the bone marrow appears in intense lipid transformation.*
Col Van Gieson's method \ 110

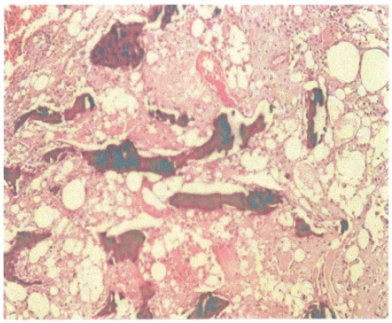

Fig. 4. *Section through the area bazipertrohanteriana. Low number of bone span with hypoplasia surrounded by bone marrow with marked lipid transformation, surrounding atrophic spinal cord, paucicelular areas. Col. Van Gieson's method \ 240*

Comparing changes in striated muscle fibre in osteoporosis with cell activities from the conjunctive interstitium, we can say that the first, have problems of structural deterioration while the seconds seem to be functionally stimulated for local defence (macrophages, mast cells) but also for local fibrosis.

Muscle fibre of the muscles in osteoporosis changes suffer metabolic, although uncharacteristic, because these changes occur in other muscle diseases, which draws attention to the fact that this bone disease should not be limited to hard tissue, but should be thought in response associative tissue muscle / bone.

Thereby, we could ask the question who is the first to blame or who has the dominant influence in the installation and progression of the disease. Also on this new idea, it will probably necessary to reconsider the drug therapy and physiotherapy applied.

Bone marrow and also medullary suffers changes: enlarged marrow spaces, enclosed with thin, simple, incomplete bays or branched bays, without cell structure. Usually, these bays are surrounded by broken broad spaces filled with fat.

The bone in these areas appears different aspects predominate in very large fatty marrow cavities, the spaces appear smaller proportion of small medullary bone marrow of normal appearance. In other holes can be found exclusively fibrous marrow, and some of them look mixed (fat - man).

There is clear distinction between trabecular bone and cortical bone. In the trabecular bone, the span areas much thinned mostly homogeneous, acellular, hypo or hipercalcificate. In cortical bone, bays look close to normal, with the presence of osteoplastic containing one, two osteocite with bone intercellular substance diminished in quantity and density. (Fig. 5, 6)

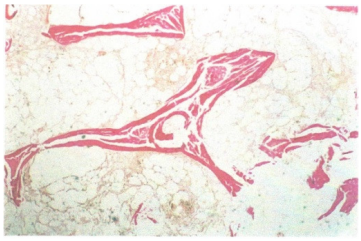

Fig. 5. *Section through the bazipertrohanteriana area. Thin bone span with hypoplasia, which limit enlarged marrow spaces filled with fat.*
Col HE \ 132

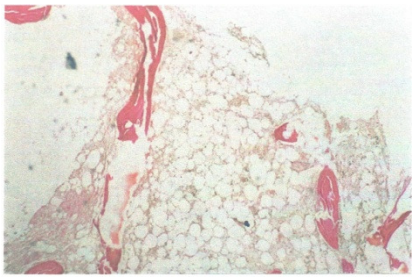

Fig. 6. *Section through the femoral. Bone marrow fat with large areas of degeneration. Fragments of bone span with extremely rare osteoplastic spaces.* CoI.HE \ 132

It is interesting the observation of bone line, bone marrow / cortical bone, which presents, strung along it cavities of different sizes and shapes, containing at least one capillary or sanguine arteriole, surrounded by connective tissue in varying amounts.

From observations made on bone, it obviously comes out also the fact that the bone marrow is in marked lipid or fibrous degradation and, in the same degree, alteration of the spongy bone span and cortical bone in the lower proportion. Bone rarefaction bone is present in the entire bone tissue, but in different degrees from one segment to another or from one bone 0area to another (epiphyseal bone marrow, cortical bone).

Lack of physical activity, decreased metabolic and energetic activity, the appearance of lipid vacuoles, fibrous degeneration, it automatically leads to a decrease of bone mass through the appearance of trabecular disorganization, broken bays, replication of cutting cones, damage and sinus capillary disorganization, growth of fibroblasts population.

These aspects lead or should lead to changes in osteoporosis management options, both in terms of prevention and treatment.

It naturally follows a simple conclusion, that the cheapest method of prophylaxis and treatment is the activity in any form, leading to a real musculoskeletal rehabilitation.

It is clear that an increase in muscle mass, an improved energy and metabolic activity leads to increased bone mass and fracture resistance.Any medical treatment in the absence of kineto-therapy it seems illusory and it should be avoided the managerial traps of drugs manufacturing companies.

We can estimate one person's osteoporosis according to how it moves daily, nutrition, habits and less by her chronological age.

Osteoporosis, in urban areas, where physical inactivity is common, is higher than in rural areas where physical activity is constant during the day and takes no account of age.

The growth of bone matrix without matrix production does not increase bone strength. Increasing muscle mass, increasing muscle vascularisation can produce an improvement in bone vascularisation, essential for osteoblast activity, which through matrix production, leads to a real increase bone strength.

Once released the harmful complication of osteoporosis, fracture, it should be treated, in order to have a fast recovery, a fast musculoskeletal recovery.

The most serious complication is the hip fracture, which should only to be treated surgically for an immediate recovery.

We can anticipate that osteoporosis with low bone strength is not strictly related to chronological age, but especially to the degree of use of the musculoskeletal system; the more a person moves, it maintains its muscle tone through physical exercises adjusted with the biological conditions, the more its muscle tone, muscle mass, bone mass and bone strength remain within protection limits against the risk of fracture. The importance of muscle/bone lesion parallelism and the existence of concrete evidence could change more or less the managerial aspects of osteoporosis in all sections of it (prevention, investigation, treatment, complications, and treatment of complications).

In terms of prevention, we believe that the best method is the musculoskeletal system sustentation, through any form of activity, calcium addition, especially food containing calcium; quitting smoking appears to be a necessity in preventing respiratory insufficiency.

The treatment of installed osteoporosis should be done shaded, because this disease should not be seen only as a bone disease.

Considering osteoporosis as an epiphenomenon within various organ failure is fair. Varying degrees of cardiac insufficiency, respiratory insufficiency, renal insufficiency (with direct determinism), and liver insufficiency all lead to musculoskeletal insufficiency.

If we consider the physical activity as the fundamental phenomenon of living forms, osteoporosis is a negative epiphenomenon of movement. The elapsed time has a biological cost for a human being, an exchange, and one of these costs is loss of muscle mass and, implicit, of bone mass.

4. Conclusions

The optical and electronic microscopic studies showed a lesion parallelism between the muscle and bone.

Mecanostatic theory tries to explain how the activation of multicellular units of work occurs, as an response to variations of effort size; it is based on the hypothesis that minimum effective effort in bone structure, should be able to trigger an adaptive response to the mechanical force.

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