Review article

Vascular calcification

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Abstract: *Calcification is a process that results to calcium deposition in non-bone tissue. The present study focuses on the molecular mechanisms and the clinical significance of vascular calcification in accordance with atherosclerosis and diabetes mellitus. The clinical significance of calcification in atherogenesis should be estimated in terms of its ability to affect the plaque stability. In diabetes, calcification seems to be an independent predictor of cardiovascular mortality correlated with the complications of diabetic patients, initially those concerning the autonomic system. These two pathological conditions seem to have important similarities and differences in their molecular pathways. The identification of hydroxyapatite and the detection of all kinds of bone-tissue cells in the wall of the calcified arteries reveal that calcification, either in atherosclerosis or in diabetes, differs from the dystrophic deposition of calcium minerals, which is observed during chronic inflammation and tissue necrosis. Vascular calcification uses all the mechanisms of embryonic osteogenesis, concerning the chondrogenic or osteogenic differentiation. Hippokratia 2006; 10(2): 60-67*

Key words: *calcification, atherosclerosis, diabetes, cardiovascular disease*

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Introduction

The term calcification includes a group of eterogenic processes that result to calcium deposition in non-bone tissues. A clear-cut distinction among the different types of calcification is: a) Metastatic calcification: occurs when concentrations of calcium phosphate in extracellular fluid exceed the saturation point, b) Dystrophic calcification, meaning the replacement or transition of injured, degenerated and necrotic tissue by mineral deposit, c) Ectopic calcification, which means the transdifferentiation of mesenchymal cells into bone tissue. Vascular wall calcification seems to be the most common form of ectopic calcification[1].

Histopathologically, calcification of cardiovascular system can be organized into four histoanatomic variants. 1) Calcification of atherosclerosis which develops in the intima layer of the arterial wall, 2) Medial artery calcification of diabetes mellitus and end-stage renal disease, 3) Cardiac valve calcification and 4) Vascular calcifylaxis, a special form of vascular calcification, as a result of chronic renal disease, iatrogenic hyperphosphatemia, tumor lysis and rabdomyolysis[2].

The present study focuses on the clinical significance of vascular calcification and the molecular mechanisms of calcification, which are associated with atherosclerosis and diabetes mellitus. These two pathological conditions, seem to have important similarities and differences in their molecular pathways, which are stimulated or suppressed by a huge number of hormones, cytokines and proteinic molecules, indicating that calcification is a very interesting process, with a high biological and clinical significance.

Calcification of the intima layer - Atherosclerosis

Atherosclerosis is characterized by chronic inflammation, as a result of dyslipidemia and other well-established risk factors. It begins very early, during childhood, by the formation of lipid strikes in the intima layer of arterial wall, which evolve to atherosclerotic plaques. According to epidemiological studies, despite the high prevalence of atherosclerosis in adults, most of them, never show clinical manifestation and they die from other causes. The answer to this paradox is given by studies, which indicate that the surfeit of cardiovascular manifestations, such as myocardial infarction, are caused by the erosion of atherosclerotic plaque. Thus, what is clinical significant is the composition of the plaque and not its size[3]. Factors associated with increased risk of plaque rupture are the increased size of the lipid core, the thin fibrous cap, the small number of smooth muscle cells, the presence of inflammatory cells and the increased proteolytic activity[4].

The process of calcification can begin at any time, during the development of atherosclerosis. The specific kind of intima layer impairment that triggers the initiation of calcification is not well known. However, it seems that calcification has a linear correlation with the duration of atherosclerosis. That's why the presence of calcification is associated with the severity of atherosclerosis[5] (Figure 1).

The clinical significance of calcification should be estimated in terms of its ability to affect the plaque stability. This fact is not disambiguated in the present literature. It is possible though, that the risk of plaque rupture, which is associated with calcification is "dose-dependent". According to current mechanistic models, the acting strain at the arterial wall is higher at the points where the stiffness changes. The calcified atherosclerotic plaques are 4-5 times stiffer than the non-calcified, making interfaces between calcified and

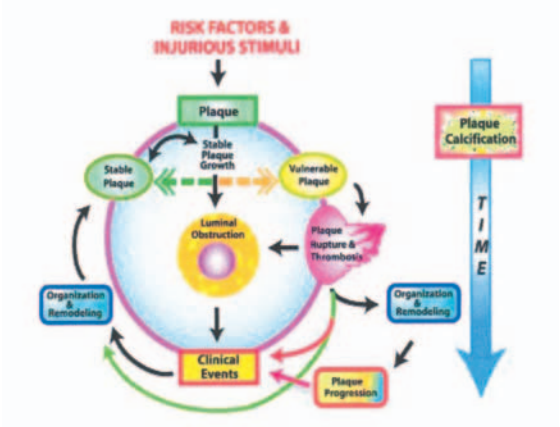

Figure 1. ***The process of calcification can begin at any time, during the development of atherosclerosis. It seems that it has a linear correlation with the duration of atherosclerosis. (Doherty T M, et al[5]).***

non-calcified tissue more vulnerable to rupture. Figure 2 shows that as the concentration of calcium increases, the interface arias augment until a peak, after which they decrease, implicating that the excessive calcification may prevent the plaque rupture[6]. This hypothesis however, has not been clinically confirmed.

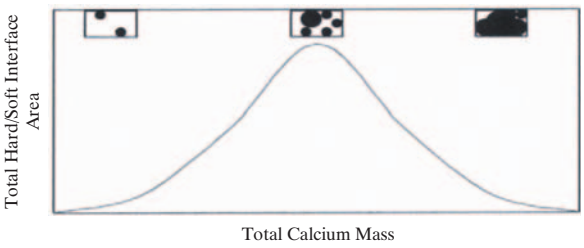

Figure 2. ***The concentration of calcium increases, the interface arias augment until a peak, after which they decrease as calcified plaques begin to coalese. (Doherty T M, et al[5]).***

Modern imaging methods, such as Electron Beam Computed tomography (EBCT) or Helical CT are used for the calculation of artery wall calcification - index. With EBCT serial images are obtained for the purposes of detecting coronary artery calcification and defining the calcium score. Recently, calcium score calculated by EBCT is used more often in the screening of asymptomatic people to access those of high risk for developing cardiovascular heart disease (CHD)[7]. The EBCT prognostic value is still an issue of studies and meta-analyses, with controversial results[8-18]. Consensus of the American Heart Association and the American College of Cardiology (AHA/ACC) in 2000, recognizes the great sensitivity and the negative prognostic value of EBCT, in the early diagnosis of cardio-vascular complications[19]. More data about the prognostic value of EBCT are expected after the termination of studies, such as MESA (Multiethnic Study of Atherosclerosis).

It is interesting, though, that histopathological analysis of coronary arteries showed that 51% of arteries with 50-80% stenosis and 36% of arteries with stenosis >80% were not calcified. The atherosclerotic plaques may increase and obstruct the entire lumen of the artery, without any sign of calcification. These findings eliminate the value of EBCT and the calcium score in estimating the size of atherosclerotic plaque and the grade of the arterial stenosis[20].

Media layer calcification - Monckerberg's sclerosis

Media layer calcification can co-exist or appear independently of the presence of atherosclerotic calcification. Usually is detected in the aorta and in arteries with decreased risk of atherosclerosis, such as gastric arteries, thyroid arteries and arteries of the mammary gland, while it is more rare in the coronary arteries. Media layer calcification usually develops concentric, giving radiographically the image of the "railway tracks" (Figure 3).

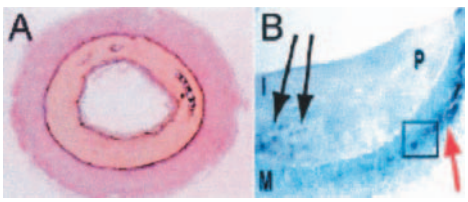

Figure 3. ***Histopathological examples of human calcification. A. Arteries may exhibit concentric media layer calcification, giving the image of the "railway track". B. Calcification in the media (red arrow) and in the intimal plaque (black arrow). (Doherty T M, et al[5]).***

Although media layer calcification is detected in young adults without metabolic diseases, it seems that it is common in older people and it has a positive linear correlation with age[3]. However, high frequency of media layer calcification exists at generalized metabolic or electrolyte disorders, especially at hypervitaminosis D, end-stage renal disease and diabetes mellitus[6].

In diabetes, calcification of media layer seems to be an index, which is dependent on the duration, the complications and the grade of glycemic control[3,5], whereas it seems to be an independent predictor of cardiovascular mortality in the diabetic patients. Ishimura et all have shown the significance of glycemic control reflected by the level of HbA1C, for the development of calcification, calculating that for every 1% increase of HbA1C, there was 2.1 fold increase in the risk of vascular calcification[21]. Other investigators report that calcification of peripheral arteries is associated with the grade of glycemic control in type II diabetes, while coronary artery calcification is associated with the glycemic control in type I diabetes[6].

Media layer calcification seems to have a strong correlation with the complications of diabetes, initially those concerning the autonomic system. In addition, an independent correlation of calcification with non-

diabetic neuropathies may exist. For example, Goebl and Fuesl showed that patients undergone bilateral sympathectomy developed, media layer calcification 6-8 years later, even though most of them were non-diabetics. Diabetic patients appeared to have more extensive calcification from non-diabetics, but the differences between two groups were not statistically significant. Thus, sympathectomy may also provoke media layer calcification independently of the presence of diabetes.

For the diabetic patients media layer calcification is an indepented predictor of cardiovascular complications. Lehto et all showed that calcification is an independent predictor of the total mortality, caused by cardiovascular complications and coronary artery disease. Everhart et all showed that non-diabetic patients with calcification of media layer had the same mortality rates with patients without calcification. Thus, media layer calcification probably is associated with higher cardiovascular risk in diabetic patients, but not in non-diabetics[3]. However, we have to underlie that all the studies are based on radiographic methods (x-ray films), which are characterized of high specificity, low sensitivity, and thus unavailability of distinguishing between media layer calcification and calcification of atherosclerosis. Thus, calculation of cardiovascular risk due to either atherosclerosis or media layer calcification seperately, in case they co-exist, is not easy.

Calcification or Ossification?

In 1840, Virchow, originally, noticed histopathologically the existence of bone tissue in the arterial wall. However, much later, in 1980, Schmid et all published in "Atherosclerosis" the identification of the hydroxyapatite crystal, in the wall of calcified arteries, indicating the suspicion, that the calcification is not a simple precipitation of calcium minerals, but it is a more complex process[21].

In 1933, Bostrom et all, reported the existence, in vitro, of pluripotent arterial cells defining them as calcifying vascular cells (CVCs), which were immunologically distinct from other arterial cells. CVCs were localized in areas of high expression of bone-related proteins, including BMP-2, a potent osteogenic agent, and also could form mineralized nodules in cell cultures under certain conditions. Later, Steitz (2002) and Nishizawa (2003), demonstrated, in vitro, that mural smooth muscle cells could express bone tissue proteins. Where as, Demer and colleagues (2003) showed that CVCs were a subset of vascular smooth muscle cells (VSMCs). Approximately 10-30 % of mural VSMCs are CVCs[2].

The identification of hydroxyapatite and the detection of all kinds of cells of bone-tissue in the wall of the calcified arteries (osteoblasts, osteoclasts, chondrocytes) demonstrated that the process of calcification, either in atherosclerosis or in media layer in diabetes, differs from the dystrophic deposition of calcium minerals, which is observed during chronic inflammation and tissue necrosis[5]. Vascular calcification uses all the mechanisms of embryonic osteogenesis, concerning the formation of either long bones (chondrogenic differentiation) or hymenogenic bones (osteogenic differentiation)[6] (Figure 4).

Molecular mechanisms of ossification

In vertebrals, the growth of bone tissue take place through 2 different pathways: chondrogenic ossification and osteogenic differentiation. The whole process is

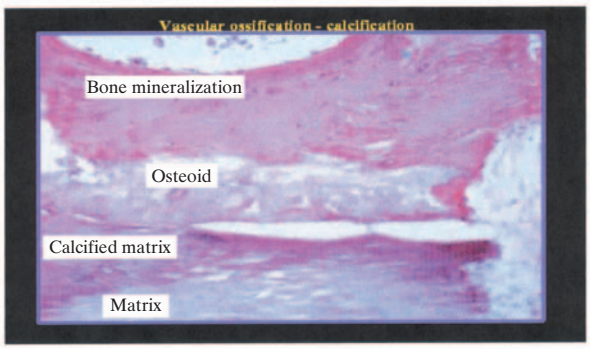

Figure 4. ***Transitional stages in vascular calcification recapitulate embryonic endochondral ossification, including an accellular matrix (matrix), amorphous mineralized matrix (calcified matrix), remodeling (osteoid), and following the formation of the complete bone tissue (bone mineralization).*** **(Hayden M R, et al[22]).**

under control of specific genetic programs, which regulate the expression of a huge number of specified molecules. Among them, the Bone Morphogenetic Protein in the calcified areas of the arteries, plays a basic role[3]. The BMP-2 signaling leads to the differentiation for either the bone formation or the artery calcification.

The multipotent progenitor mesenchymal cells of the artery wall are capable to differentiate into chondroblasts, osteoblasts, lipocytes or myoblasts (Figure 5). The direction of the differentiation depends on the expression of specific metagraphic agents.

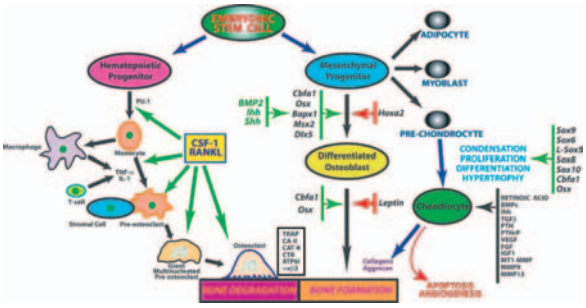

Figure 5. ***Differentiation of the progenitor stem cells to chondroblasts, osteoblasts and osteoclasts. (Doherty T M, et al[3]).***

Chondrogenic or endochondral ossification is direct dependent on the expression of metagraphic factor Cbfa-1. On the other hand, the molecular pathways of hymenogenic ossification are activated by the BMP-2

expression. Which of these mechanisms will be activated, depends on the stimulant, which triggers the whole process.

In atherosclerosis, oxidized LDL provokes the expression of Cbfa-1 and the initiation of the chondrogenic ossification, which begins from the lipid core of the atherogenic plaque and elaborates asymmetrically[2]. In diabetes mellitus, both the direct reaction of glucose and the increased production of the glucose products are responsible for the diversion of mesenchymal cells gene expression, resulting in the expression of BMP-2 and differentiation of mesenchymal cells to osteoblasts, process similar to osteogenic ossification. The whole process results in the medial layer calcification, with concentric distribution. (Figure 6A, 6B).

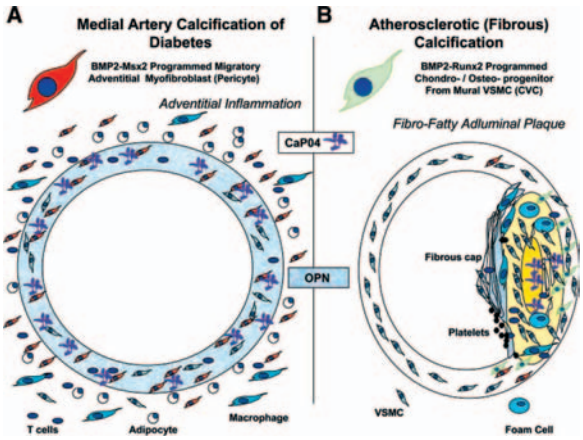

Figure 6. ***Vascular calcification of diabetes (left) and atherosclerosis (right). (Vattikuti R, et al[5]).***

On the other hand the atherosclerotic lesions rich in monocytes and macrophages consist a great source of pro-osteoclasts. The maturation of pro-osteoclasts to osteoclasts demands the presence of two specific cytokines: Colony Stimulating Factor-1 of monocytes (CSFM-1) and Receptor Activator of Nuclear Ligand (RANKL), which have been detected in the atherosclerotic plaque[6]. The formation and the activity of osteoclasts is also affected by a large number of cytokines (including TNF-a), indicating that the signaling pathways which are associated with the formation and the reabsorption of bone are interlinked with those of the immune system[5].

Molecular regulators of calcification

In table 1, there is a list of molecules known to participate in the development and function of osteoblasts, osteoclasts and chondrocytes, and their chromosomal locations in human.

Matrix Gla Protein

Almost a decade ago, tissue calcification was thought to be a passive precipitation associated with areas of advanced tissue degeneration or necrosis and also with an unavailability of the local inhibitor agents to prevent it[5]. Several proteins known to be expressed in arteries modulate precipitation of calcium salts in the extracellular fluid, notably proteins that contain unusual glutamine residues with an additional carboxy group added in an enzymatic reaction catalyzed by γ-carboxylase. Glutamine residues carboxylated at the γ-position are called Gla residues. Proteins containing Gla residues are known as Gla proteins and Gla residues are an important functional feature of both arterial calcification and homeostasis.

MGP is a protein that contains five Gla residues, each one representing a strong affinity with Calcium phosphate (hydroxyapatite)[1]. It has been shown to be present in association with atherosclerotic plaques, which seems to be paradox from a first point of view. However, the inhibiting ability of MGP depends on the degree of MGP- [gamma]-carboxylation and the ratio of concentrations of the 2 molecules[6]. Lack of the function (from insufficient- [gamma]-carboxylation) rather than the amount of MGP, may be the factor that increases risk of calcification. γ-Carboxylase exists in many tissues, including arteries. Both expression and enzyme activity of γ-carboxylase is inhibited at atherosclerotic plaques, causing, by a negative feedback mechanism, the production of MGP, which is uncarboxylated and inactive.

Enzyme γ-carboxylase is vitamin k-dependent[6]. This feature explains the development of calcification during therapy with warfarin and products.

Warfarin anticoagulator activity takes place through the inhibition of γ- carboxylation of Gla residues of homeostatic factors, averting the γ-carboxylation of MGP protein[6].

From the study of MGP knock out mice is concluded that MGP acts not only directly on calcium mineral formation but also regulates the cell differentiation. This evidence indicates that MGP inhibits mesenchymal cell differentiation to the osteogenic lineage, by suppressing the osteogenic factor BMP-2.

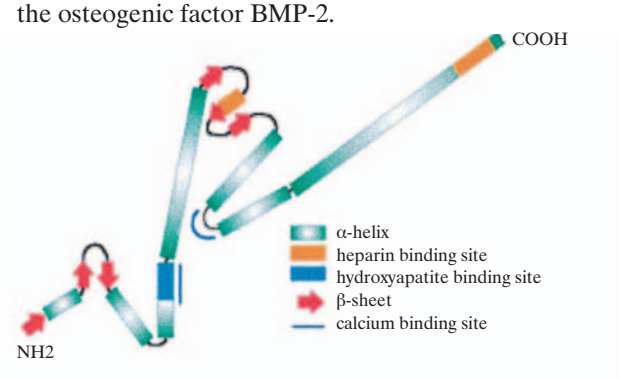

Figure 7. ***The molecule of osteopontin (www.cmb.lu.se/ctb/photos/OPN2.jpg).***

Osteopontin (OPN)

Osteopontin (OPN) is another Bone Matrix Protein, which is not found in normal arteries but is indeed expressed in atherosclerotic plaques and colocalizes with calcified plaque regions[5]. This hydrophilic,

phosphorylated acidic glycoprotein of 298 amino acids, binds calcium phosphate and cell surface integrin receptors. It is like cytokines, produced by the macrophage[2]. OPN is expressed from the energized tissue macrophage and also from the endothelial and the SMCs of the calcified arteries[6]. Other sites of OPN production seem to be kidney, placenta and nervous system.

OPN production reflects an osmotic, inflammatory or stress metabolic response, cellular transformation or osteogenic differentiation. Elevated blood glucose in diabetes stimulates factor 1 and Fos, which upregulates OPN expression of the VSMCs[2]. The main molecular stimulators of OPN production are TNF-a, interleukin 1, interferon-γ and endotoxines[22].

OPN inhibits calcification through a variety of ways: 1) inhibits the development of apatite crystals through self-aggregation and adhension of the later, 2) stimulates bone resorption by decreasing the cytosolic calcium of osteoclasts, which leads to their activation. Steitz et all suggest that OPN promotes resorption of ectopic calcification by creating an acidic enviroment via the expression of carbonic anydrase II[6], 3) it inhibits directly BMP-2-Msx2 dependent calcification.

Biologically active OPN exists in a minimum of three forms: 1) a matrix-bound form, 2) a circulating soluble

Table 1. ***Molecular determinants of calcification.***

Common name	Gene name	Locus
Alkaline Phosphatase	ALPL	1p36.1-p34
Bone Gla Protein(Osteocalcin)	BGP	1q25-q31
Bone Sialoprotein II	BSP	4q21-25
Calcitonine Receptor	CALCR	7q21.3
Carbonic anhydrase II	CA2	8q22
Cathepsin K	CTSK	1q21
Chordin	CHRD	3q27
Core-binding factor α-1	Cbfa-1	6p21
Core-binding factor -β	CBF-β	16q22.1
Klotho	KL	13q12
Matrix Gla Protein	MGP	12p13.1-p12.3
Noggin	NOG	17q22
Osteonectin	ON	5q31.3-q32
Osteopontin	OPN	4q21-q25
Tartrate-resistant acid phosphatase	ACP5	19p13.3p13.1
Osterix	OSX	12q12
Bone Morphogenetic Protein -2	BMP-2	20p12
Bone Morphogenetic Protein -4	BMP-4	14q22-q23
Bone Morphogenetic Protein Receptor -IA	BMPR-IA	10q22.3
Bone Morphogenetic Protein Receptor -IB	BMPR-IB	4q22-q24
Bone Morphogenetic Protein Receptor II	BMPR-II	2q33-q34
Mothers against decapentaplegic homolog 6	SMAD5	15q22-q23
Colony-Stimulating Factor -1	CSF-1(M-CSF)	1p21-p13
Colony-Stimulating Factor -1 Receptor	CSFR-1	5p33.2-33.3
Osteoprotegerin	OPG	8q24
Receptor Activator of Nuclear Factor κB1	RANK	18q22.1
Receptor Activator of Nuclear Factor κB1 Ligand	RANKL	13q14
Tumor Necrosis Factor -α	TNFα	6p21.3
Tumor Necrosis Factor -α Receptor	TNFR1	12p13.2
TNF receptor-associated factor 6	TRAF6	11p11.2
Interferon -β	INFB	9p21
Parathyroid hormone-related peptide	PTHrP	12p12.1-p11.2
Transforming Growth Factor -β	TGFB1	19p13.1
Parathyroid Hormone	PTH	11p15.3-15.1
Leptin	LEP	7q31.3
Leptin Receptor	LEPR	1p31
Low density lipoprotein Receptor-related Protein 5	LRP5	11q13.4
Matrix Metaloproteinase-3	MMP3	11q22.3
Angiotensin-Converting Enzyme 1	ACE1	17q23
Angiotensin-Converting Enzyme 2	ACE2	Xp22

form, 3) a secreted soluble form. Some data indicate the ability of OPN to inhibit calcification depends on the phosphorylation of its molecule. Current research will give answers to this hypothesis[2].

OPN plasma levels of patients with coronary artery disease (CAD) were significantly higher than those of healthy individuals, increasing with disease severity, independently of the conventional risk factors. The above evidence implies that OPN plasma levels, could be a possible predictor of coronary artery disease[6].

Osteoprotegerin (OPG)-RANKL

Osteoprotegerin is another protein-link between bone and vascular tree. It is a member of the tumor necrosis factor- [gamma] receptor superfamily and an indirect inhibitor of osteoclastogenesis, through its ability of binding and inhibiting RANKL. RANKL is an activator agent of its receptor RANK, which triggers the maturation of pro-osteoclasts to osteoclasts[6]. Specifically, the activity of OPG seems to be determined from the ratio OPG/RANKL.

OPG and RANKL are both expressed at the unaffected arterial wall and, in vitro, at cultures of SMCs, derived from the coronary arteries. Current researche has shown a possible correlation of OPG plasma levels with some pathological conditions. In patients with stable angina, OPG levels are associated with significant coronary artery narrowing. It seems that, estrogen therapy is associated with increased OPG levels.

Treatment

Intensive observation of the atherosclerotic plaque implicate that calcification is a reversible process. On the other hand, the grade of calcification is a predictor associated with increased risk of cardiovascular incidences and diabetic complications. The reduction of vascular calcification is the central point of the work many researchers and strategies are being developed to address this shortcoming.

The main pharmaceutical agents that have been used in atherosclerosis-calcification experimental models are Ca-antagonists and statins. Ca-antagonists block the Ca-channels inhibiting the Ca-ions entrance in the intracellular fluid and are drugs with well-known anti-hypertensive action. The main representatives are verapamile, diltiazem and dihydropyridines (such as nifedipine and amlodipine). The agents of this category seem to affect some pathways of atherosclerotic calcification, like the local adhesion of collagen fibres, elastin and Ca, the macrophage infiltration of the plaque and the proliferation and migration of the SMCs[23, 24].

The use of verapamile in mice (with experimental calcification of the vascular wall, after high dosage of Vitamin D) resulted in the inhibition of calcification[25]. The INSIGHT study (Intervension as a Goal in Hypertension Treatment) showed also that nifedipine in hypertensive patients decreases the development of the coronary artery calcification[26, 27]. Flenkenstein et all compared the results of Ca-antagonists in three different experimental models of calcification: 1) Ca type (development of calcification in mice after high dosage of Vitamin D and nicotine), 2) cholesterol type (rabbit on rich lipid diet), 3) mixed type. The calcium antagonists inhibited the calcification at type 1 and 3 but they didn't affect type 2 of calcification, which is more common to human atherosclerotic model[28].

Statins are inhibitors of HMG-CoA anagogase and apart from their lipid-lowering activity, they have a pleiotropic activity, including the improvement of the endothelial function, the plaque stability, anticoagulative, antioxidative and antiinflammatory actions. In addition, recent evidence suggests that statins have prevented the osteoporotic fractures. Therapeutical use of statins seemed to provoke reduction of the vascular calcification together with a reduction of the atherosclerotic plaque volume. Studies that compared statins and Ca-antagonists in experimental models showed that statins had better affect in reducing plaque volume. Ca-antagonist, seemed to reduce calcification more effectively, when combined with statins[32].

Biphosphonates (pamidronate, etidronate, aledronate) are used in the treatment of osteoporosis, Paget disease, and hypocalcaemia of neoplastic disease. They act by interfering in the hydroxyapatite crystals of bone, reducing the rate of both osteogenesis and osteolysis.

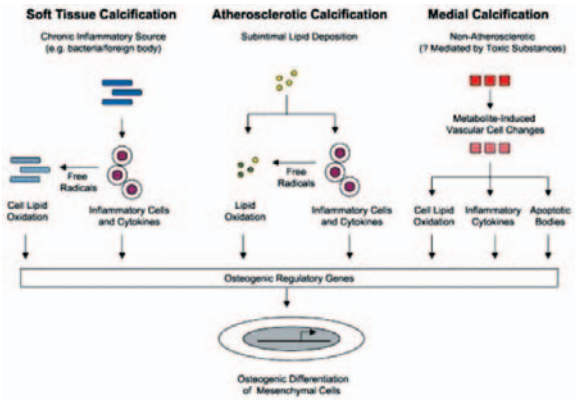

Figure 8. ***Oxidized lipids, inflammatory cells and cytokines indicate a common feature of calcification either on the soft tissues or the calcification of the media layer and atherosclerosis (Linda L Demer et al[21]).***

In vivo studies have shown that biphosphonates prevent ectopic vascular calcification by inhibiting not only mineralization process but the cholesterol, elastin and collagen accumulation as well. Biphosphonates and statins seem to have similar actions both in the bone and in the lipid metabolism. Amino-bi-phosphonates localize their action in the same metabolic pathway, as this of HMG-CoA reductase, inhibiting the phamesyl-biphosphonate synthetase[3]. Indeed, there is Clinical evidence suggesting that statins augment bone density, while biphosphonates reduce blood lipid levels and

prevent atherosclerosis. Cohort studies should be made in order to determine the actions of the drugs above and implicate possible new indications.

Perspectives

Aging is consistent with osteoporosis, on one hand, and with vascular calcification on the other. Study of this fact suggests that these two processes are linked to some extent. Bearing in mind the similarities in embryogenesis of bone and vascular cells, as well as their interaction during the development of the skeleton, this relationship may not be inexplicable[6].

Nevertheless bone and vessels respond, in a diametrical different way, in chronic inflammation. A possible explanation comes from the evolution theory. Bacterial cell wall is rich in lipids, which are oxidized by the immune system cells. These oxidized lipids accumulate in the tissues surrounding the infection. In chronic inflammation, when the immune system response fails, a secondary reaction of the surrounding connective tissue is initiated in order to deteriorate the spreading of the injurius stimulus. For example, a repetitive injury of the skin results in scar or callous formation. In a similar way chronic osteomyelitis causes lysis of the surrounding bone in order to eliminate the substrate on which the microbial infection is thriving[21].

In both situations, hyman's response intends to differentiate the substrate, creating a hostile environment for each given injurious stimulus. Similar responses are triggered by the chronic inflammation of atherosclerosis or diabetes in the vessel wall. Oxidized LDL may mimic the lipids of the bacterial wall. Thus, the contact of oxidized LDL with endothelium and with bones as well, may trigger a response, that leads to ectopic bone formation in the vessel wall and to bone resorption, resulting in vascular calcification and osteoporosis at the same time.

There are still many questions to be answered, concerning the pathogenesis and the clinical significance of vascular calcification. Why some atherosclerotic plaques are calcified, whether others are not, and how calcification affects the evolution of atherosclerotic disease? When should vascular calcification be treated and what would be the treatment goals?

As a conclusion, more research is needed in order to clarify the mechanisms of chronic inflammation, and the role of the immune system in vascular calcification, as well as its relationship with osteoporosis.

References

1. Noriyashu Kobayashi, Riko Kitazawa, Sakan Maeda, et al. T-138 Polymorphism of Matrix Gla Protein promoter alters it's expression but is not directly associated with atherosclerotic vascular calcification. Kobe J Med Sci 2004; 50:1-13
2. Vattikuti R, Towel DA. Osteogenic regulation of vascular calcification: an early perspective. Am J Physiol Endocrinol Metab 2004; 286:E686-E696
3. Terens M. Doherty, Lorraine A. Fitzpatrick, Daisuke Inoue, et al: Molecular, endocrine and genetic mechanisms of arterial calcification. Endocrine Reviews 2004; 25:629-627
4. Schmermund A, Erbel R. Unstable coronary plaque and its relation to coronary calcium. Circulation 2001; 104:1682-1687
5. Terens M. Doherty, Kamlesh Asotra, Lorraine A. Fitzpatrick, et al. Calcification in atherosclerosis: Bone biology and chronic inflammation at the arterial crossroads. PNAS 2003; 100:11201-11206
6. Abedin, Moeen, Tintut, et al. Vascular calcification: Mechanisms and clinical ramifications[Brief Reviews]. Arterioscler Thromb Vasc Biol 2004; 24:1161-1170
7. Redberg RF, Shaw LJ. A review of electron beam computed tomography: implications for coronary artery disease screening. Prev Cardiol 2002; 5:71-78
8. Schmermund A, Stang A, Mohlenkamp S, et al. Prognostic value of electron-beam computed tomography-derived coronary calcium scores compared with clinical parameters in patients evaluated for coronary artery disease. Prognostic value of EBCT in symptomatic patients. Z Kardiol 2004; 93:696-705
9. Schmermund A, Mohlenkamp S, Erbel R. Coronary artery calcium and its relationship to coronary artery disease. Cardiol Clin 2003; 21:521-534
10. Nasir K, Budoff M. J, Post WS, et al. Electron beam CT versus helical CT scans for assessing coronary calcification: current utility and future directions. Am Heart J 2003; 146:969-977
11. Fuseini M, Goodwin W. J, Ferris E. et al. Does electron beam computer tomography provide added value in the diagnosis of coronary artery disease? Curr Opin Cardiol 2003; 18:385-393
12. Sinitsyn V, Belkind M, Matchin Y, et al. Relationships between coronary calcification detected at electron beam computed tomography and percutaneous transluminal coronary angioplasty results in coronary artery disease patients. Eur Radiol 2003; 13: 62-67
13. Thomson LE, Hachamovitch R. Coronary artery calcium scoring using electron-beam computed tomography: where does this test fit into a clinical practice? Rev Cardiovasc Med 2002; 3:121-128
14. Raggi P. The use of electron-beam computed tomography as a tool for primary prevention. Am J Cardiol 2001; 88(7B): 28J-32J
15. Keelan P. C, Bielak L. F, Ashai K, et al. Long-term prognostic value of coronary calcification detected by electron-beam computed tomography in patients undergoing coronary angiography. Circulation 2001; 104:412-417
16. Nallamothu B. K, Saint S, Bielak L. F, et al. Electron-beam computed tomography in the diagnosis of coronary artery disease: a meta-analysis. Arch Intern Med 2001; 161:833-838
17. Haberl R, Becker A, Leber A, et al. Correlation of coronary calcification and angiographically documented stenoses in patients with suspected coronary artery disease: results of 1,764 patients. J Am Coll Cardiol 2001; 37:451-457
18. Bielak L. F, Rumberger J. A, Sheedy P. F 2nd, et al. Probabilistic model for prediction of angiographically defined obstructive coronary artery disease using electron beam computed tomography calcium score strata. Circulation 2000; 102:380-385
19. American College of Cardiology/American Heart Association. Expert consensus document on Electron-Beam Computed Tomography for the diagnosis and prognosis of coronary artery disease. Circulation 2000; 102:126-140
20. Wexler L, Brundage B, Crouse J, et al. Coronary artery calcification: Pathology, epidemiology, imaging methods,

and clinical implications: A statement for health professionals from the American Heart Association. Circulation 1996; 1175-1192
21. Demer L L. Mineral Exploration: Search for the mechanism of vascular calcification and beyond. The 2003 Jeffrey M. Hoeg award lecture. Arterioscler Thromb Vasc Biol 2003
22. Hayden M, Tyagi S C, Kolb L, et al. Vascular ossification-calcification in metabolic syndrome, type 2 diabetes mellitus, chronic kidney disease and calciphylaxis-calcific uremic arteriolopathy : The emerging role of sodium thiosulfate. Cardiov Diab 2005; 4:4
23. Motro M, Shemesh J, Grossman E. Coronary benefits of calcium antagonist therapy for patients with hypertension. Curr Opin Cardiol 2001; 16:349-355
24. Armstrong M L, Heistad D D, Lopez J A. Regression of atherosclerosis. A role for calcium antagonists. Am J Hypertens 1991; 4(7 Pt 2):503S-511S
25. Fleckenstein-Grun G, Thimm F, Frey M, et al. Progression and regression by verapamil of vitamin D3-induced calcific medial degeneration in coronary arteries of rats. J Cardiovasc Pharmacol 1995; 26:207-213
26. Ruilope L M. Long-term protection in at-risk hypertensive patients—a role for nifedipine GITS? Blood Press 2002; 11:106-109
27. Taddei S, Ghiadoni L, Salvetti A. Current treatment of patients with hypertension: therapeutic implications of INSIGHT. Drugs 2003; 63:1435-1444
28. Fleckenstein-Grun G, Thimm F, Frey M, et al. Role of calcium in arteriosclerosis-experimental evaluation of antiarteriosclerotic potencies of Ca antagonists. Study group for calcium antagonism. Basic Res Cardiol 1994; 89 (Suppl 1):145-159
29. Callister T Q, Raggi P, Cooil B, et al. Effect of HMG-CoA reductase inhibitors on coronary artery disease as assessed by electron-beam computed tomography. N Engl J Med 1998; 339:1972-1978
30. Achenbach S, Ropers D, Pohle K, et al. Influence of lipid-lowering therapy on the progression of coronary artery calcification: a prospective evaluation. Circulation 2002; 106:1077-1082
31. Raggi P. Regression of calcified coronary artery plaque assessed by electron beam computed tomography. Z Kardiol 2000; 89 (Suppl 2):135-139
32. Delsing D J, Jukema J W, van de Wiel M A, et al. Differential effects of amlodipine and atorvastatin treatment and their combination on atherosclerosis in ApoE*3-Leiden transgenic mice. J Cardiovasc Pharmacol 2003 Jul; 42:63-70