



# Scanning Electron Microscopy Study of the Elemental Composition of Endothelium in Arterial Atherosclerosis

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## Abstract

A previously unknown existing fact has been established – high boron content in arterial endothelium (22.14%) in 17 autopsy preparations of various stages of atherosclerosis using scanning electron microscopy. Among 22 elements of the endothelium, a high content of carbon, nitrogen, oxygen has been revealed, the compounds of which with boron exhibit unique properties, possibly explaining the strength, density, hardness, elasticity, shiny smooth layer, and high fluidity of the endothelium. Boron oxide in water releases heat, forms an acid that dissolves in lipids and alcohol, penetrates into cells, and damages them. It is likely that boric acid disrupts the integrity of the endothelium and promotes the “passage” of chemical elements and the formation of atheroma. This assumption helps to reconsider the pathogenesis of arterial atherosclerosis, and possibly its treatment. The compound of boron with hydrogen, their varieties are a source of high energy used as rocket fuel. A hypothesis is proposed that the compound of boron with hydrogen and oxygen is a source of human energy, which has important scientific and practical significance and requires in-depth study. In the calcified stage of atherosclerosis, the content of aluminum, phosphorus and calcium increases 10 times compared to the initial stage.

**Keywords:** Atherosclerosis; Endothelium; Artery; Scanning electron microscope; Element; Boron; Oxygen; Carbon; Nitrogen.

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## 1. Introduction

Cardiovascular diseases (CVD), particularly atherosclerotic diseases, are a major cause of morbidity and mortality, affecting more than 523 million people worldwide. More than 20.5 million people die from this pathology annually, with arterial atherosclerosis being the leading cause. Half of these deaths are due to coronary heart disease (CHD), and another quarter to ischemic stroke.<sup>[1,2]</sup>

The arterial endothelium plays a major role in the formation of atherosclerosis, is a single-layer cellular layer that lines the inner surface of all blood vessels and provides a barrier between the intra- and extravascular spaces, controls vascular permeability, determines the vascular tone of the underlying smooth muscles and plays an important role in the

inflammatory response,<sup>[3]</sup> and also controls blood fluidity, platelet adhesion and aggregation, leukocyte activation, adhesion and transmigration.<sup>[4]</sup> It is well known that everything that is present in the atherosclerotic plaque has passed through the endothelial walls.<sup>[5]</sup>

### 1.1 Elemental composition of the artery in atherosclerosis

Most publications report the results of studying the elemental composition of the artery in atherosclerosis. A number of authors report that using Raman spectroscopy it is possible to accurately determine quantitatively the chemical composition of the atherosclerotic artery - cholesterol, cholesterol esters, triglycerides and phospholipids, calcium salts.<sup>[6-10]</sup>

The elemental composition of the atherosclerotic human popliteal artery was studied using the proton-induced X-ray emission (PIXE) method and the concentrations of Cl, K, Ca, Fe, Cu, Zn, Br and Pb were determined, as well as their localization in different parts of the arterial walls. The highest average concentrations of Cl, K, Zn and Br were found in the shells of the medium.<sup>[11]</sup> In 14 subjects, the concentration and

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localization of P, S, Cl, K, Ca, Fe, Cu, Zn, and Br were measured in the popliteal arteries using the PIXE and micro-PIXE methods. The presence of PO<sub>4</sub>(3-) and CO<sub>3</sub>(2-) groups was assessed using the IR method. The amount of P and Ca increased with age, approaching 9 and 20% in places, and mineral deposits were found in the middle layer. However, data on the content of the above elements in the endothelium were not presented.<sup>[12]</sup> Elevated iron and zinc are usually combined with microcalcifications. Rich calcium deposits resemble amorphous phosphate formations rather than pure hydroxyapatite.<sup>[13]</sup> A number of researchers have suggested the need to assess the physiology of the coronary artery and metabolic bone diseases for “stiffness, fibrosis, calcification”.<sup>[14]</sup> Proton microprobing of the carotid artery in mice has shown that the atherosclerotic process is associated with locally accumulated calcium, iron, zinc, calcium-rich deposits resemble amorphous calcium phosphate.<sup>[15,16]</sup> As can be seen in the presented studies of determining the chemical elements of the arterial wall, clinical and anatomical changes depending on the stage of atherosclerotic lesion of the artery have not been studied.

## 1.2 Elemental composition of atherosclerotic plaque

SEM-EDX analysis showed an abundance of phosphorus and calcium in quantities sufficient to form calcium phosphates, and a markedly reduced sodium level. The most abundant mineral in atheromatous plaque is hydroxyapatite (Ca<sub>10</sub>(PO<sub>4</sub>)<sub>6</sub>(OH)<sub>2</sub>), on which cholesterol crystals and lipid cores are deposited, stratifying the plaque into layers reflecting different stages of its formation. The difference in calcium and sodium concentrations between arteries with and without atheromas may indicate an important relationship in the pathophysiological development of calcium deposits.<sup>[9]</sup> Both large and small mineral deposits have been isolated from atherosclerotic plaque and have been shown to have essentially the same chemical composition: calcium apatite (71%), carbonate (9%), and contain a relatively high percentage of protein (15%). However, scanning electron microscopy revealed that the deposits were heterogeneous, consisting of five distinct structures: (1) isolated and conglomerates of smooth-surfaced spheres composed of spherical layers; (2) spheres composed of spindle-shaped, radially arranged particles; (3) fibers forming networks and bundles, which sometimes included spherical particles; (4) irregularly shaped particles with fuzzy surfaces, and (5) flat plates with smooth surfaces.<sup>[17]</sup> Other authors report on the elemental composition of atheroma. The results of morphology, immunohistochemistry, transmission electron microscopy, and energy-dispersive X-ray microanalysis of

229 carotid plaques show that calcification is present in 77.3% of cases.<sup>[18]</sup> The researchers, in elemental and mineralogical analysis, claim that phosphate deposition occurs in the affected tissues associated with magnesium inclusion, which probably leads to a transition to the crystalline phase.<sup>[19]</sup> The study of the composition of non-calcified plaques was assessed using dual-energy CT (DECT). The mass percentage of elements with higher atomic numbers, such as F, Na, Mg, S, Si, P, Cl, K and Ca, was found to be insignificant, higher in vulnerable plaques compared to stable plaques. A significantly increased mass percentage of nitrogen was also found in stable plaques.<sup>[20]</sup> With proton and electron microprobe, infrared spectroscopy and X-ray diffraction, histochemistry, calcification was observed only in the atheroma, while in the media after 2-10 days others.<sup>[21]</sup> A number of researchers report the capabilities of dual-energy computed tomography (CT) for quantitative assessment of the chemical composition of the plaque in terms of water, lipid, protein and calcium content.<sup>[22,23]</sup> The results of quantitative assessment of the chemical composition of plaques by spectroscopic analysis of laser-induced fluorescence (LF) are presented by other authors.<sup>[24]</sup> Several researchers have shown the advantages of using <sup>1</sup>H NMR techniques for quantitative imaging of lipid components of human atheroma in vivo, as well as for studying their chemical and physical properties, especially in human hard arterial plaques.<sup>[25]</sup>

## 1.3 Elemental composition of the endothelium

It should be emphasized that publications devoted to the elemental composition of the intima are rare. In the studied atherosclerotic popliteal artery of humans, the elemental composition of the sections of Ca and Fe distribution varied: sometimes these elements predominated in the intima, while in other cases the highest concentrations were observed in the middle or adventitia of the shell.<sup>[11]</sup>

Other researchers argue that calcification is a pathological thickening of the intima (from <0.5 to 15 μm). As it progresses, it becomes larger (>15 μm - 1 mm in diameter), fragmented (>1 mm) and, ultimately, leaf-shaped (>3 mm). In this case, the calcium content as an element is not determined.<sup>[26]</sup>

In numerous scientific studies, the role of cholesterol (C<sub>27</sub>H<sub>46</sub>O), simple and complex lipids (CH<sub>3</sub>(CH<sub>2</sub>)<sub>n</sub>COOH), triglyceride (C<sub>55</sub>H<sub>98</sub>O<sub>6</sub>), phospholipid C<sub>81</sub>H<sub>158</sub>O<sub>17</sub>P<sub>2</sub>, high-density lipoproteins (HDL), low-density lipoproteins (LDL), very high-density lipoproteins (VLDL) and other compounds dominates in determining the pathogenesis of atherosclerosis. It is accepted that atheroma consists of a composite of carbon atoms, oxygen, hydrogen in different proportions and a small amount of calcium, nitrogen, phosphorus, while other

elements are absent. The main function of HDL is the transport of cholesterol from peripheral tissues to the liver, which plays a role in the biodistribution of lipids.<sup>[27]</sup> HDL are known for their antiatherogenic and anti-inflammatory properties due to the absorption and return of cholesterol stored in the foam cells of atherosclerotic plaques to the liver. Thus, reducing the size of the plaque and the inflammation associated with it.<sup>[28,29]</sup>

Therefore, determining the elemental composition of the endothelium is extremely important and sheds new knowledge and light on the understanding of the pathogenesis, morphology and clinical picture of arterial atherosclerosis.

The purpose of this study was to determine the chemical elemental composition of the endothelium in atherosclerotically affected arteries at various stages of its development.

We conducted a study of publications based on Scopus, EndNote, Google Scholar, Web of Science, PubMed with the keywords "chemical elements of the arterial endothelium", and 24 publications were identified that were closest to the topic.

The focus of the study was a literature review of the study of the elemental composition of the arterial wall as a whole, the atherosclerotic plaque and the endothelial wall itself and to conduct our own study of the elemental composition of the endothelium.

Thus, it can be stated that the above publications on the elemental composition concern either atherosclerosis or atheroma, and the elemental composition of the endothelium has not been studied at present. In this regard, the qualitative

and quantitative analysis of elements in the endothelium and the study of their dynamics depending on the clinical stage of atherosclerosis are of not only scientific interest, but also clinical.

**2. Material and methods of the study**

The study was conducted on a Quanta 200i 3D scanning electron microscope (FEI Company, USA), which is equipped with an energy-dispersive X-ray analysis system (EDAX). The range of elements determined is B÷U. Energy resolution is 132 eV (Mn K $\alpha$ ). The sample modules were heated to +1000 °C.

The objects of the study were anatomical materials - arteries of those who died at various stages of atherosclerosis, where the cause of death was myocardial infarction, stroke, patients who underwent stenting, aortic aneurysms, and others. The preparations were removed by pathologists and preserved in a formalin solution. Before the study, the arteries were cleared of fatty tissue from the outside, cut into thin layers transversely and dried.

We have studied 22 elements (B, C, N, O, F, Na, Mg, Br, Al, Si, P, S, Pb, Cl, K, Ca, I, Ba, Mn, Fe, Cu, Zn) of the endothelium (intima) of atherosclerotically affected arteries of 17 autopsy preparations, including those in the initial (6) and calcified stages (11). The coverage of the study object was 20  $\mu$ m. The results of each study were recorded as a photo indicating the coverage area (a), the ratio of 22 elements in percent of mass and atom in total taken as 100% (c) and a diagram (d). (Fig. 1)

All obtained data were processed by a special program of

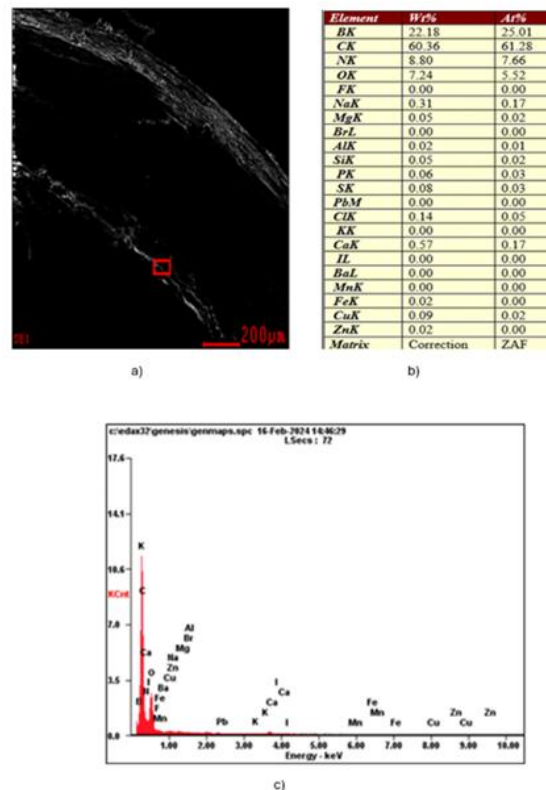


Fig. 1: (a) Coverage area, (b) mass and atom of elements in percentage, (c) diagram.

the Quanta 200i 3D scanning electron microscope, recorded and stored on a PC, and then subjected to comparative statistical analysis by elements for each stage of atherosclerosis.

### 3. Results and discussion

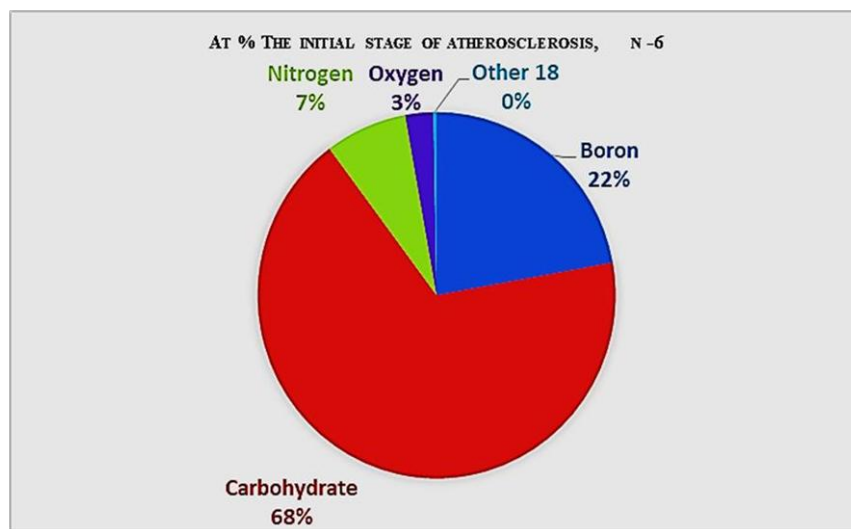
#### 3.1 In the stage of primary atherosclerosis (TYPE I)

The structure of the elemental composition of the endothelium (Table 1), the ratio of chemical elements in percentage (Fig. 2) and the highest values of elements (B, C, N, O) (Fig. 2) are presented.

Interesting data were obtained that among all the 22 elements studied, 4 elements had a large specific gravity (99.71%), in particular boron - 22.14% (CI: 20.05-25.81),

**Table 1:** Structure of the elemental composition of the endothelium of atherosclerosis in the initial stage.

Elements	At %/ number of studies 6						Confidence interval (CI)
	1	2	3	4	5	6	
<i>B</i>	22,09	22,20	20,05	20,55	25,81	22,18	22,14(CI 20,05-25,81)
<i>C</i>	65,01	69,95	69,60	70,66	63,77	66,88	67,64(CI 63,77-70,66)
<i>N</i>	8,09	5,73	8,34	5,71	7,70	8,84	7,4(CI 5,71-8,84)
<i>O</i>	4,36	1,90	1,75	2,76	2,62	1,84	2,53(CI 1,84-4,36)
<i>F</i>	0,00	0,00	0,00	0,00	0,00	0,00	0,00
<i>Na</i>	0,22	0,04	0,02	0,02	0,04	0,06	0,06
<i>Mg</i>	0,02	0,00	0,01	0,01	0,00	0,01	0,05
<i>Br</i>	0,00	0,00	0,00	0,01	0,00	0,00	0,01
<i>Al</i>	0,01	0,01	0,03	0,05	0,00	0,04	0,02
<i>Si</i>	0,01	0,01	0,03	0,03	0,01	0,02	0,02
<i>P</i>	0,05	0,05	0,01	0,02	0,00	0,01	0,02
<i>S</i>	0,05	0,01	0,04	0,03	0,01	0,01	0,02
<i>Pb</i>	0,00	0,00	0,00	0,00	0,00	0,00	0,00
<i>Cl</i>	0,01	0,02	0,00	0,01	0,00	0,01	0,05
<i>K</i>	0,01	0,00	0,00	0,01	0,01	0,02	0,05
<i>Ca</i>	0,03	0,07	0,04	0,05	0,01	0,06	0,04
<i>I</i>	0,00	0,00	0,01	0,01	0,00	0,00	0,02
<i>Ba</i>	0,00	0,00	0,00	0,00	0,00	0,00	0,00
<i>Mn</i>	0,00	0,00	0,00	0,01	0,00	0,00	0,01
<i>Fe</i>	0,00	0,00	0,02	0,01	0,01	0,00	0,01
<i>Cu</i>	0,02	0,00	0,02	0,02	0,01	0,00	0,01
<i>Zn</i>	0,03	0,01	0,01	0,03	0,00	0,00	0,01



**Fig. 2:** The ratio of chemical elements in percentage in the group of the initial stage of atherosclerosis.

carbohydrate - 67.64% (CI: 63.77-70.66), nitrogen - 7.4% (CI: 5.71-8.84) and oxygen - 2.53% (CI: 1.84-4.36), and the remaining 15 elements in total amounted to - 0.29%, while fluorine, lead, barium (F, Pb, Ba) were not detected in all samples of the preparations (Fig. 3, 4).

The results of the study showed a fairly high content of boron (B) in the intima of the artery within 22.14 (CI 20.05-

25.81), such a percentage content was stably detected in all groups. In the available scientific and medical literature, no reports were found on the results of a study of boron as a structural element of blood vessels. At the same time, studies show that the elements that form atherosclerosis calcifications (Mg, Br, Al, SK, P, SK, Pb, Cl, K, Ca, I, Ba, Mn, Fe, Cu) were found in very small quantities.

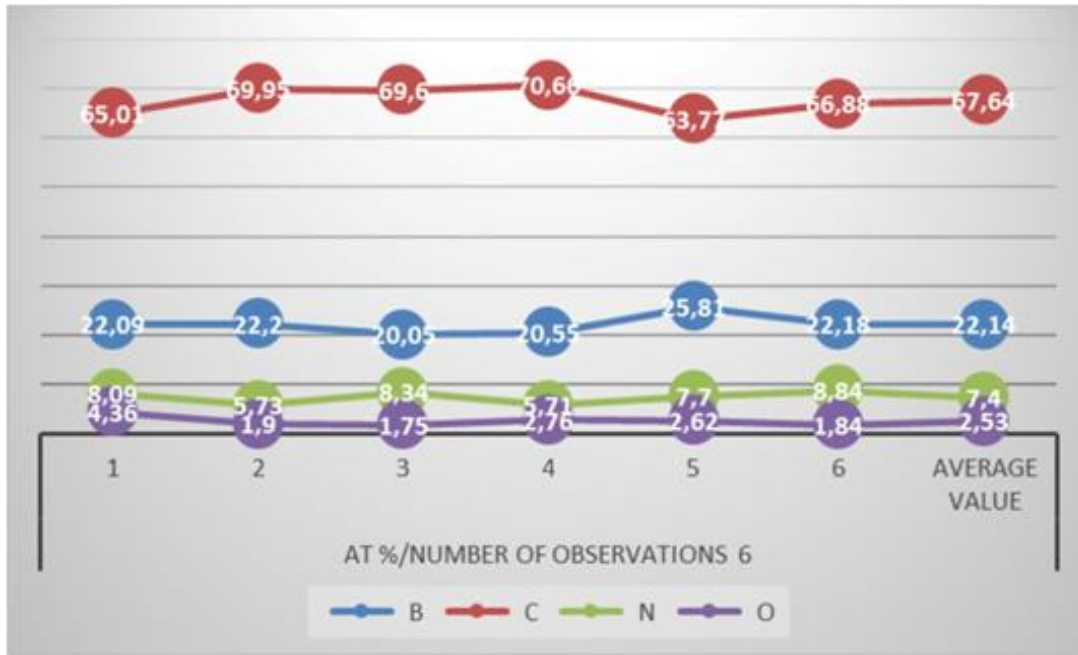


Fig. 3: The highest levels of elements (in At% - B, C, N, O) in the endothelium in the initial stage of atherosclerosis (N=6).

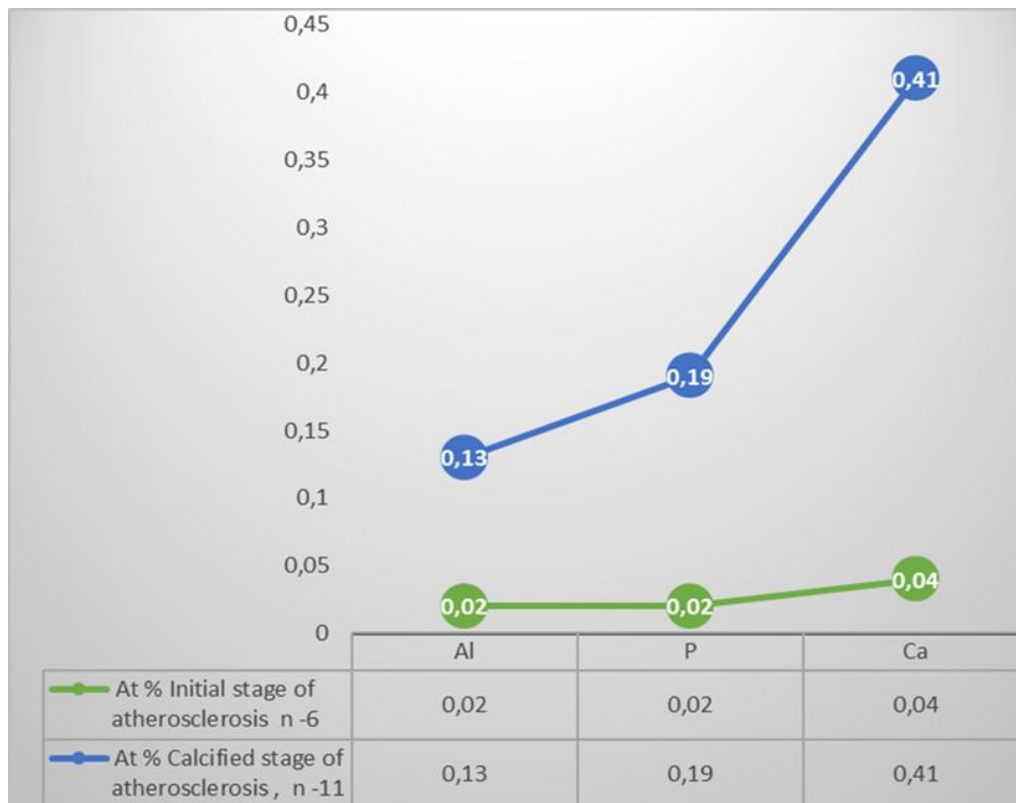


Fig. 4: The content of Al, P, Ca increases 10 times in the calcified stage of atherosclerosis compared to the initial stage.

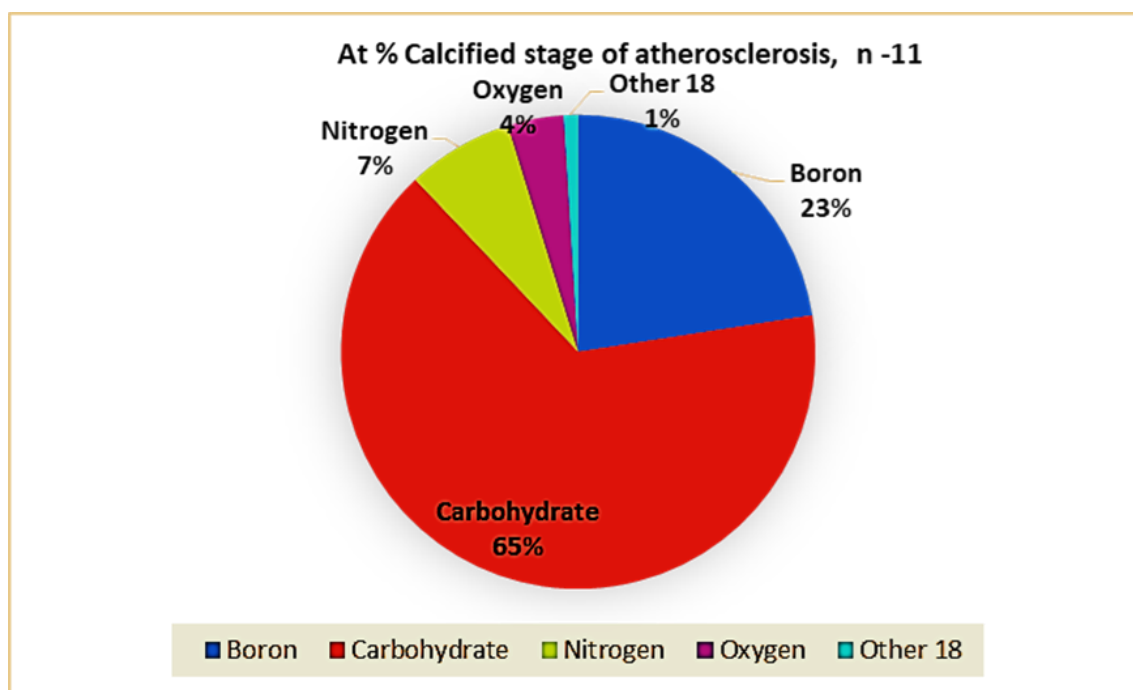
**3.2 The structure of the elemental composition of the atherosclerotic endothelium in the calcified stage**

These studies indicate a high content of four elements (boron - 22.53% (CI: 11.63-23.97), carbohydrate - 65.41% (CI: 62.43-68.55), nitrogen - 7.27% (CI: 2.32-10.02) and oxygen - 3.83% (CI: 2.11-12.97) is 99.04%, the remaining 15 elements

(F, Pb, Ba) are absent in all samples of the preparations (Table 2, Fig. 5).

**Table 2:** The structure of the elemental composition of the atherosclerotic endothelium in the calcified stage.

Elements	At %/ number of studies 11											Average value
	1*	2	3	4	5	6	7	8	9	10	11	
<i>B</i>	23,16	11,63	23,13	21,49	23,97	24,01	24,39	22,95	25,27	24,53	23,31	22,53
<i>C</i>	63,98	66,14	68,55	66,59	62,43	64,31	64,09	67,63	64,00	63,62	68,23	65,41
<i>N</i>	9,11	2,32	6,05	7,10	8,92	8,28	7,77	6,30	10,02	9,36	4,74	7,27
<i>O</i>	3,57	12,97	2,11	4,29	4,48	3,16	3,64	2,54	0,59	2,31	2,48	3,83
<i>F</i>	0,00	0,00	0,00	0,00	0,00	0,00	0,00	0,00	0,00	0,00	0,00	0,00
<i>Na</i>	0,08	0,31	0,08	0,16	0,13	0,09	0,03	0,01	0,01	0,00	0,04	0,08
<i>Mg</i>	0,01	0,06	0,00	0,02	0,01	0,01	0,01	0,02	0,01	0,01	0,02	0,02
<i>Br</i>	0,00	0,03	0,00	0,00	0,00	0,00	0,00	0,01	0,00	0,00	0,00	0,00
<i>Al</i>	0,06	0,08	0,02	0,02	0,03	0,02	0,02	0,30	0,01	0,01	0,10	0,13
<i>Si</i>	0,00	0,06	0,00	0,01	0,00	0,01	0,01	0,02	0,01	0,01	0,05	0,01
<i>P</i>	0,02	1,93	0,03	0,09	0,00	0,03	0,01	0,00	0,00	0,01	0,04	0,19
<i>S</i>	0,01	0,01	0,00	0,04	0,01	0,03	0,02	0,04	0,01	0,03	0,09	0,07
<i>Pb</i>	0,00	0,00	0,00	0,00	0,00	0,00	0,00	0,00	0,00	0,00	0,01	0,00
<i>Cl</i>	0,00	0,00	0,00	0,05	0,02	0,02	0,00	0,00	0,00	0,00	0,05	0,01
<i>K</i>	0,00	0,04	0,00	0,01	0,00	0,00	0,00	0,00	0,01	0,00	0,07	0,01
<i>Ca</i>	0,00	3,97	0,02	0,14	0,00	0,04	0,00	0,05	0,02	0,04	0,29	0,41
<i>I</i>	0,00	0,07	0,00	0,00	0,00	0,00	0,00	0,00	0,01	0,00	0,06	0,01
<i>Ba</i>	0,00	0,03	0,00	0,00	0,00	0,00	0,00	0,00	0,00	0,00	0,06	0,00
<i>Mn</i>	0,00	0,06	0,00	0,00	0,00	0,00	0,00	0,01	0,01	0,00	0,07	0,01
<i>Fe</i>	0,00	0,08	0,00	0,00	0,00	0,00	0,00	0,03	0,01	0,01	0,12	0,02
<i>Cu</i>	0,00	0,09	0,00	0,00	0,00	0,00	0,00	0,02	0,01	0,01	0,09	0,02
<i>Zn</i>	0,00	0,11	0,00	0,01	0,00	0,00	0,00	0,05	0,01	0,02	0,08	0,02



**Fig. 5:** The ratio of chemical elements in percentage in the group of calcified stage of atherosclerosis.

### 3.3 Comparative characteristics of the elemental composition of the endothelium in the initial and calcified stages of atherosclerosis.

When comparing the indices of endothelial elements in the initial stage of atherosclerosis with the calcified stage, a

slight increase in the content of boron and oxygen, respectively, is observed (22.14% - 22.53%), (2.53-3.83), a decrease in carbon and nitrogen - (67.64%-65.41%), (7.4%-7.27%) (Fig. 6, Table 3).

At the same time, the indicators of this group are

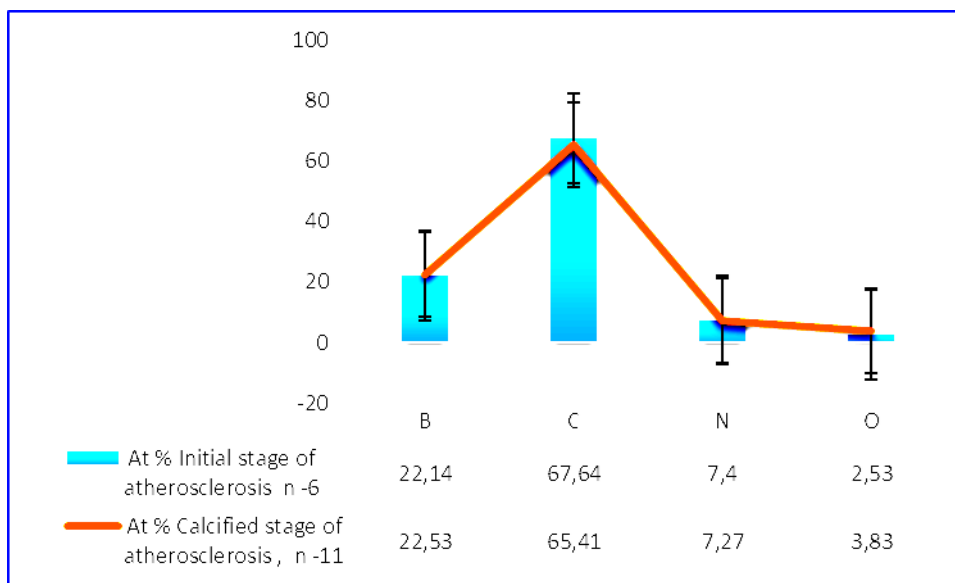


Fig. 6: The ratio of chemical elements in % in two groups of atherosclerosis.

Table 3: Comparative characteristics of the elemental composition in the dynamics of the initial and calcified stage of atherosclerosis.

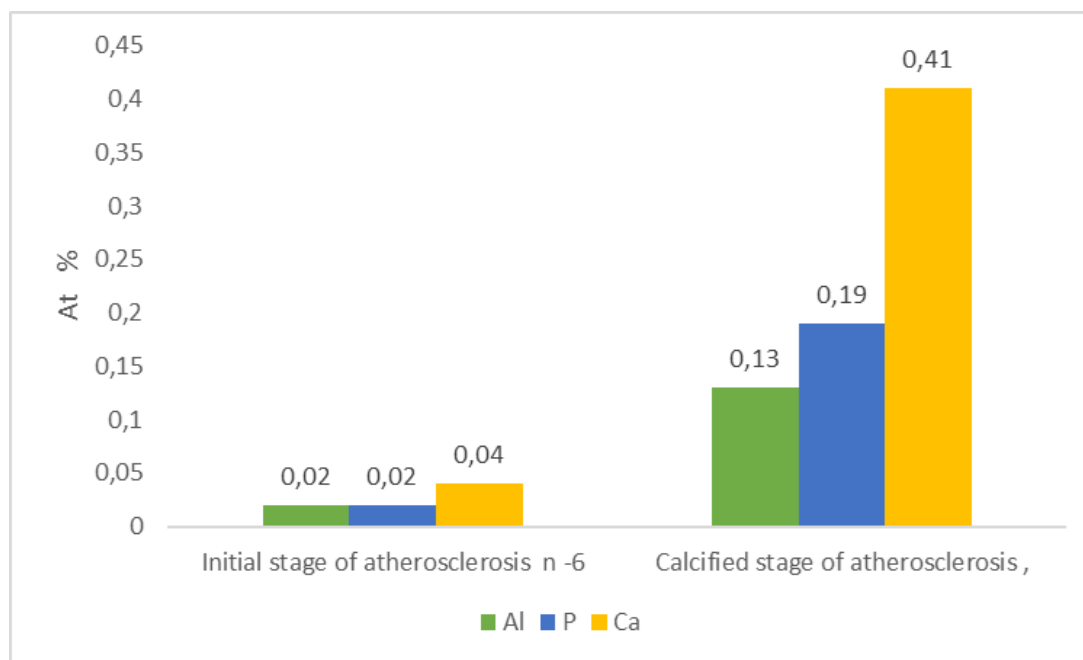
Elements	At %	
	Initial stage of atherosclerosis , n -6	Calcified stage of atherosclerosis , n -11
B	22,14 ( CI : 20,05-25,81)	22,53 ( CI: 11,63-25,27)
C	67,64 ( CI :63,77-70,66)	65,41 ( CI :62,43-68,55)
N	7,4 ( CI :5,71-8,84)	7,27 ( CI :2,32-10,2)
O	2,53 ( CI :1,84-4,36)	3,83 ( CI :0,59-12,97)
F	0,00	0,00
Na	0,06	0,08
Mg	0,05	0,02
Br	0,01	0,00
Al	0,02	0,13
Si	0,02	0,01
P	0,02	0,19
S	0,02	0,07
Pb	0,00	0,00
Cl	0,05	0,01
K	0,05	0,01
Ca	0,04	0,41
I	0,02	0,01
Ba	0,00	0,00
Mn	0,01	0,01
Fe	0,01	0,02
Cu	0,01	0,02
Zn	0,01	0,02

noteworthy: aluminum (0.02%-0.13%), phosphorus (0.02%-0.19%) and calcium (0.04%-0.41%) increase tenfold compared to the initial stage of atherosclerosis (Fig. 7). A slight decrease in the following elements Na, Mg, Si, Cl, K, Fe, Cu, Zn was found.

We were the first to detect using scanning electron microscopy in 17 autopsy artery preparations a reliably high content of boron 22.14% (CI 20.05-25.81) as a chemical element in the endothelial wall, as well as carbohydrate - 67.64% (CI: 63.77-70.66), nitrogen - 7.4% (CI: 5.71-8.84),

oxygen - 2.53% (CI: 1.84-4.36), which together make up (99.71%) of the total proportion of elements (Table 1). At the same time, the role of boron in the animal organism is not clear. Human muscle tissue contains  $(0.33-1) \cdot 10^{-4}\%$  boron, bone tissue  $(1.1-3.3) \cdot 10^{-4}\%$ , and blood - 0.13 mg/l. Boron is contained in vessels  $(0.13 \text{ mg/l} \cdot 4.5 \text{ liters of blood}) = 0.585 \text{ mg}$ , *i.e.* 0.5 g. A person receives 1-3 mg of boron daily with food.<sup>[30]</sup>

The results of our studies have shown that the consistently high content of boron, oxygen, nitrogen and carbon in the



**Fig. 7:** Dynamics of element indicators (Ca, P, Al) in the endothelium at the initial (N=6) and calcified stage of atherosclerosis (N=11).

studied groups are not random facts. In this regard, the logical question arises - what consequences can boron compounds with these elements lead to, which together make up 99.71% of the total share of 22 studied elements in the arterial endothelium. Despite extensive research, many aspects of the properties of boron and its compounds with other chemical elements remain shrouded in mystery and require comprehensive scientific research. Modern researchers report potential and novel applications of boron, exceptional properties of prime importance in manufacturing, in the performance aspects of numerous products, in materials science, biotechnology, medicine, in nuclear power plants as a neutron absorber, agriculture, in energy storage, space, superconductors, materials, magnetic/non-magnetic nanoparticles and medical applications including cancer therapy, in the production of ceramics, optical fibers, fiberglass, glass, *etc.*<sup>[31-35]</sup>

A review of the literature showed that the compound of boron with carbon (B4C) - boron carbide is the hardest and most chemically resistant substance, is the third hardest known substance after diamond and cubic boron nitride, for which it received the nickname "black diamond". It is used for the

production of glass for chemical glassware, bulletproof vests, the nuclear industry, for the manufacture of grinding and abrasive materials, electronics and glass and chemical industries, and high-strength ceramic products.<sup>[36,37]</sup>

A compound of boron with nitrogen (BN) is used as a high-quality abrasive material, superior to diamond in many respects, it is in demand in heavy engineering, the automotive industry, the mining industry, construction, and for high-quality optical microscopes. Boron nitride (Borazon, Elbor) does not enter into any reactions, is universal and heat-resistant, strong and durable, stable, and is used in aircraft construction.<sup>[38-40]</sup>

According to literary data, boron oxide (B2O3) easily forms a glassy state, which explains the creation of transparency, smoothness, and high fluidity of the endothelium.<sup>[41]</sup>

Thus, during visual examination of autopsy preparations, we established that arterial endothelium was shiny, dense, hard, strong, transparent, smooth with high fluidity, their preparation was very difficult. (Fig. 8-11)

This is confirmed by the results of the study, when we directly affected atherosclerotic plaques of 124 arterial vessels



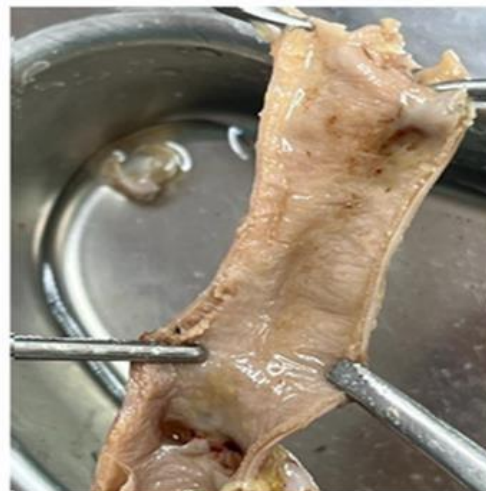
**Fig. 8:** Bifurcation of the abdominal aorta.



**Fig. 9:** Iliac artery.



**Fig. 10:** Carotid artery.



**Fig. 11:** Abdominal aorta.

with 8 lipolytic drugs and literary data.<sup>[42-44]</sup> Direct evidence of these properties of the endothelium is the maintenance of systolic blood pressure up to 150-230 mm Hg in hypertension, which, according to WHO estimates, affects 1.28 billion adults aged 30-79 years worldwide.<sup>[45]</sup> Thus, we believe that the above properties of the endothelium are associated with chemical compounds of boron with carbohydrate, nitrogen and oxygen. Boron oxide ( $B_2O_3$ ) dissolves in water with strong heating and formation of boric acid. 100 g of boron oxide added to 125 g of water brings it to a boil, soluble in hot water.<sup>[46]</sup> The compound of boron with hydrogen ( $B_xH_x$ ), as shown by the analysis of the literature, is characterized by high chemical activity and an extremely high heat of combustion, has the ability to "spontaneously ignite or explode in air". Diborane -  $B_2H_6$ , pentaborane, tetraborane, carborane (boron with hydrogen and carbon  $B_{10}H_{10}C_2H_2$ ) are used as rocket fuel and as hydrogen fuel cells for electric vehicles, since their heat of combustion ( $\sim 70$  kJ/g) is significantly higher than that of hydrocarbons (45 kJ/g).<sup>[47-52]</sup> As for the source of human hydrogen, water in blood plasma makes up to 90% of the volume of circulating human blood and a person consumes at least 1.5 liters of water daily. Based on the above, a hypothesis

is put forward that heat in the human body (and possibly warm-blooded animals) is probably produced due to reactions of a compound consisting of boron, nitrogen, hydrogen and carbon in the lumen of an artery, *i.e.* in the blood with the participation of the endothelium.

A number of researchers claim that boric acid dissolves well in lipids, water, glycerin, ether, alcohol and quickly penetrates into cells, coagulates cytoplasmic protein and leads to their death.<sup>[49]</sup> The content of boric acid ( $H_3BO_3$ ) in the blood is 7 - 100  $\mu g / l$ . and a person consumes 1-18 mg of boron daily with food, lipids in norm/pathology - total cholesterol (200/240 mg/dl), LDL cholesterol ("bad" 100/190 mg/dl), HDL cholesterol ("good" 40/60 mg/dl) and Triglycerides (150/500 mg/dl, causing hardening of the arteries.<sup>[52]</sup>

The presence of lipids, glycerol, and alcohol in the blood are well-known facts, so it is likely that the presence of boric acid in the blood, disrupting the integrity of the endothelium and creating conditions for the migration of chemical elements through the endothelium walls, contributes to the formation of atheroma in the artery wall.

Particular attention is drawn to the significant difference in

the content of boron (22.53%) and carbon (65.41%) compared to calcium (0.41%). The hardness and strength of boron carbide, which are not inferior to cubic boron nitride and diamond, and high chemical resistance are described above. Based on the results obtained, we believe that the formation and development of "calcifications" of an atherosclerotic plaque is associated with a combination of boron and carbon (B4C), and possibly nitrogen. Therefore, the above allows us to question the existing concept that such an insignificant content of calcium (0.04-0.41%), phosphorus (0.02-0.19%), aluminum (0.02-0.13%) and other elements in the endothelial wall can form calcified atherosclerotic plaques.

#### 4. Conclusion

Scanning electron microscopy of the arterial endothelium in the initial and calcified stages of atherosclerosis has revealed for the first time a new, previously unknown, unidentified and verifiable to mankind object of the material world and an objectively existing chemical element boron (B) in the wall of the arterial endothelium within 22.14-22.53 At.%.

We assume that the high fluidity of the arterial endothelium for blood, its shiny, dense, hard, strong, transparent, smooth properties are associated with chemical compounds of boron with carbohydrate, nitrogen and oxygen, which together accounted for 99.71% of the total share of 22 elements.

We state that the consistently high content of boron, nitrogen and carbon in the studied groups is not a random fact. Probably due to the reactions of a compound consisting of boron, nitrogen, hydrogen and carbon in the lumen of the artery, *i.e.* in the blood with the participation of the endothelium, heat is generated in the human body (and possibly warm-blooded animals). Based on scientific facts and research results, we hypothesize that boric acid in the blood, disrupting (or destroying) the integrity of the endothelium and creating conditions for the migration of chemical elements through the walls of the endothelium, contributes to the formation of atheroma in the arterial wall.

The results of this study suggest that the formation and development of "calcifications" of an atherosclerotic plaque is associated with a compound of boron and carbon (B4C) and cast doubt on the existing concept that such an insignificant content of calcium (0.04-0.41%), phosphorus (0.02- 0.19%), aluminum (0.02-0.13%) and other elements in the endothelial wall can form atherosclerotic plaques.

Undoubtedly, the proposed hypotheses and assumptions will cause scientific discussion, and in the future they require comprehensive, more in-depth research and evidence, the results of which have important scientific and, in the long term, practical significance.

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#### Conflict of Interest

There are no known competing financial interests or personal relationships that could affect the work described in this article.

#### Supporting Information

Not applicable.

#### CRedit Statement

**Oralbay Darmenov:** Supervision, Investigation, Data curation, Writing - Original draft. **Erbol Oralbaevich Darmenov:** Data curation, Visualization, Formal analysis. **Zhuldyz Abilseitkyzy Zhaksybai, Nazim Isaeva:** Resources, Investigation, Formal analysis. **Saken Duisenbaevich Koibakov:** Resources, Investigation. **Akhmedsadyk Dinara:** Investigation, Formal analysis.

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