

## MORPHOLOGY AND PATHOMORPHOLOGY

# Morphological Examination of Changes in Intact and Atherosclerotic Human Aortic Walls Induced by Laser Irradiation

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We performed morphological examination of intact and atherosclerotic aortic regions after laser irradiation with various characteristics (wavelength and energy). Particular attention was given to the type of damages to the vascular wall around craters formed by the laser beam. Combined irradiation with UV and visible lights produced least pronounced damages to surrounding tissues in the aortic wall.

**Key Words:** *atherosclerotic injury; aorta; laser irradiation; crater*

Previous studies showed the effect of laser irradiation on the vascular wall. However, clinical tests of laser angioplasty produced negative results [2,5]. In our opinion, damage to the vascular wall around atherosclerotic plaques (ASP) produced by laser irradiation was not completely analyzed, in particular, the regimen of irradiation was not optimized.

The aim of the present study was the search for laser parameters ensuring minimal destruction of the vascular wall around the crater. To this end we compared the effects of various lasers on intact and atherosclerotic aorta wall.

## MATERIALS AND METHODS

Samples of the aorta from men and women (50-80 years) were obtained at autopsy 6-12 h after death. We examined fragments of intact and atherosclerotic aortas. Aorta samples were irradiated with laser

light, fixed in 10% formalin, dehydrated in alcohols, and embedded in paraffin. Sections were stained with hematoxylin and eosin. We estimated the size of craters (depth, width, and volume) and the state of the vascular wall around it.

The following lasers were used: LIDA-T UV-laser ( $\lambda=0.308 \mu$ , regimens 1 and 2, Table 1), UV laser equipped with a scattering cell and providing irradiation in UV ( $\lambda=0.308$  and  $\lambda=0.355 \mu$ , regimen 3) and visible ranges ( $\lambda=0.414 \mu$ ), Nd-YAG laser ( $\lambda=1.06 \mu$ , regimen 4), and Nd-YAG laser with nonlinear crystal transmitting irradiation into the second harmonic ( $\lambda=0.53 \mu$ , regimen 5). Pulse frequency varied from 20 to 50.

## RESULTS

Laser irradiation led to the formation of funnel-like or rectangular craters with smooth edges, which was associated with complete destruction of tissues in the irradiated zone. Sometimes carbonized detritus fragments were seen at the bottom of craters after irradiation with Nd-laser. Under various regimens of

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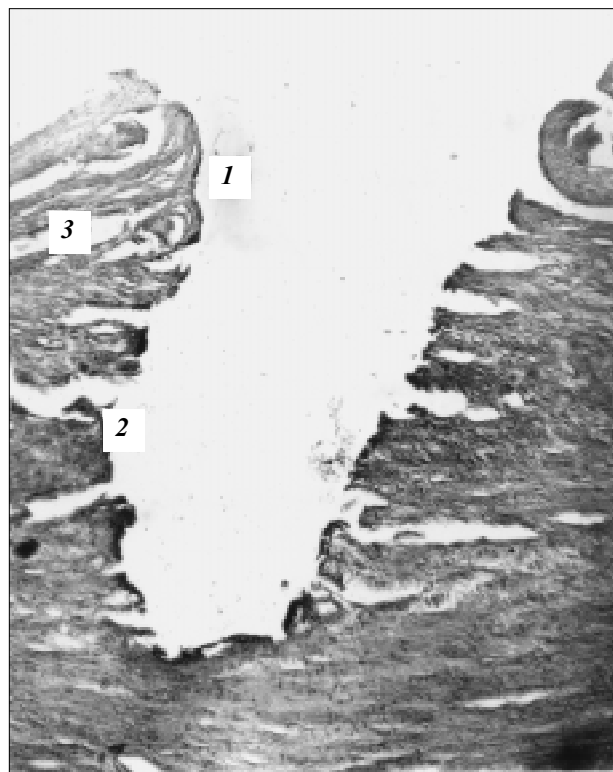
irradiation the burned zone with modified tinctorial properties was relatively small (25-30 and not more than 10  $\mu$  in intact aortic tissues and ASP, respectively). The vascular wall around the craters was loosened (Fig. 1). The depth of craters depended on the laser source and state of the vascular wall.

The effects of UV irradiation under regimen 1 depended on laser energy and diameter of the laser beam. A positive correlation was found between pulse energy and crater depth. The higher was laser energy, the greater was mechanical destruction of surrounding tissues in the aortic wall. If the diameter of laser beams decreased, relatively deep craters were formed after low-energy irradiation (Table 1). Damages to the aortic wall did not depend on the diameter of a laser beam and were insignificant at low-energy irradiation of intact tissues and ASP. These data indicate that irradiation with 20 laser pulses (energy 30 mJ/pulse, beam diameter 0.5 mm) produced craters in ASP with a depth of 1.2 mm, which was not accompanied by loosening of surrounding tissues in the aortic wall.

Twenty pulses with laser energy of 500 mJ/pulse produced a through channel in the intact aortic wall. However, this procedure was accompanied by severe thermal and mechanical damages to the vascular wall. Laser irradiation-induced changes in intact aortic regions were more pronounced than in atherosclerotic zones (Table 1). After irradiation the zone of necrosis in intact aorta was greater than in ASP (Table 1).

The rate of ASP craterization (crater depth/pulse) induced by UV irradiation for 200 nsec under regimen 2 was lower than that observed after irradiation with laser energy of 150 mJ/pulse for 60 nsec. Laser irradiation for 60 and 200 nsec produced similar effects on the intact wall (Table 1). As differentiated from regimen 1, irradiation under regimen 2 was accompanied by the formation of pronounced thermal and mechanical damages to the surrounding tissues in the vascular wall. It should be emphasized that the positive correlation between pulse frequency and crater depth was not necessarily found during irradiation under regimen 2. The first doses of laser energy caused most pronounced destruction of tissues. Despite severe damages to the vascular wall, this regimen of irradiation is widely used in devices for angioplasty [3,5].

UV irradiation under regimen 3 produced moderate effects on intact and atherosclerotic aortic walls. We did not find necrotized tissues and destructed regions around craters. After irradiation with laser energy of 60 mJ/pulse the depth of craters in ASP 2-fold surpassed that observed after UV irradiation for 60 nsec (Table 1). Increasing the



**Fig. 1.** Craterization of atherosclerotic plaque after irradiation with a XeCl UV laser ( $\lambda=0.308 \mu$ , laser energy 60 mJ/pulse, 60 nsec, beam diameter 1 mm). Hematoxylin and eosin staining ( $\times 120$ ). Evaporated substance (1), carbonization (2), and mechanical damages to the vascular wall (3). Insignificant carbonization of margins in the lower crater segment. Preserved contours of the crater and loosening of the base substance.

frequency of pulses was accompanied by a decrease in the rate of craterization (similarly to regimen 2). This regimen is most favorable for treatment of ASP (with respect to the rate of craterization and minimal mechanical and thermal damages to surrounding tissues in the aortic wall).

It should be emphasized that craters in ASP and intact aortic wall were rapidly formed after irradiation with the infrared laser under regimen 4 (Table 1). Under this regimen we did not find necrotic damages to surrounding tissues in the aortic wall. However, morphological examination revealed cavernous loosening of elastic fibers in surrounding tissues of the vascular wall. As differentiated from regimen 3, these loosening were positioned not only perpendicular to, but also along the axis of craters (above the bottom). Craters were partially filled with fragments of evaporated substances and endothelial layer. It should be emphasized that irradiation under regimen 4 resulted in the formation of small craters in calcified regions of the vascular wall.

Irradiation with green light under regimen 5 produced craters with a greater depth (compared to regimen 3, Table 1). This treatment caused

**TABLE 1.** Effects of Irradiation under Various Regimens on Intact and Atherosclerotic Aortic Walls

Irradiation regimen (source, pulse duration, beam diameter); power	Depth, $\mu$ /pulse		Necrotic zone, $\mu$		Mechanical damage, degree	
	normal	ASP	normal	ASP	normal	ASP
XeCl, 60 nsec (No. 1)						
1 mm; 250 mJ	60 $\pm$ 4	48 $\pm$ 5	30	5	III	III
150 mJ	30 $\pm$ 3	20 $\pm$ 2	20	5	II	II
60 mJ	22 $\pm$ 2	12 $\pm$ 2	10	3	II	II
0.5 mm; 300 mJ	—	64 $\pm$ 5	—	5	—	III
150 mJ	—	65 $\pm$ 4	—	5	—	II
30 mJ	—	52 $\pm$ 4	—	—	—	II
XeCl, 200 nsec, 1 mm (No. 2)						
150 mJ	30 $\pm$ 3	14 $\pm$ 2	30	10	III	III
70 mJ	28 $\pm$ 2	14 $\pm$ 2	20	5	II	II
XeCl+scattering cell, 60 nsec, 1 mm; 60 mJ (No. 3)	25 $\pm$ 3	22 $\pm$ 2	—	—	I	I
Nd-YAG, 10 nsec, 0.3 mm; 60 mJ (No. 4)	50 $\pm$ 2	54 $\pm$ 3	—	—	III	II
Nd-YAG, second harmonic, 10 nsec, 0.3 mm; 50 mJ (No. 5)	34 $\pm$ 3	34 $\pm$ 3	—	—	II	II

**Note.** Degree I: insignificant injury without laser-induced changes around craters. Degree II: considerable damages with the formation of lateral caverns and loosening. Degree III: severe damages and necrosis.

pronounced mechanical, but not thermal damages to the surrounding tissues in the vascular wall.

Loosening of elastic fibers reflects mechanical damages to the vascular wall induced by a laser beam. In our experiments, damaged zones produced by laser light were similar to those observed previously [1,4].

Irradiation with the same pulse frequency was followed by the formation of deep craters in the intact aorta and small craters in the fibrous membrane covering atheromatous tissues. In this case, the subintimal layer was “pressed” into atheromatous tissues. Irradiation of calcified zones in the vascular wall did not lead to the formation of craters.

Our results indicate that regimen 3 (irradiation with UV and visible lights) is most favorable with

respect to the rate of craterization and minimal mechanical and thermal damages to surrounding tissues in the aortic wall. This regimen of irradiation should undergo *in vivo* studies and clinical tests.

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