Endothelium-dependent acetylcholine-induced vasodilatory response of saphenous vein grafts

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Abstract

Introduction: It has been suggested that endothelial dysfunction is critical to saphenous vein graft (SVG) occlusion.

Aim: To evaluate in vivo endothelium-mediated vasoreactivity in angiographically non-stenotic SVG.

Methods: The group consisted of 31 patients (pts), aged 58.7±8.7 years, 54±38 months after coronary bypass surgery. In each patient one angiographically normal SVG was selected for the study. Endothelium-dependent vasoreactivity was investigated with acetylcholine (Ach) 50 µg intragraft infusion. Graft diameter changes were measured by quantitative computer angiography (QCA).

Results: In 17 (54.8%) pts there was a significant reduction in graft diameter following Ach infusion, from 3.8±0.7 to 3.2±0.7 mm (p=0.0001), whereas in 4 (12.9%) pts there were no diameter changes (3.2±0.7 mm). In the 10 remaining pts (32.3%) we found graft dilatation from 3.5±0.5 mm to 3.9±0.5 mm (p=0.0002). In multivariate linear regression analysis, SVG dilatation positively correlated with a low ratio of graft/artery diameter (p <0.002), high HDL-cholesterol level (p <0.001) and absence of hypertension (p <0.03), and negatively correlated with postoperative myocardial infarction (p <0.01).

Conclusions: Endothelium-dependent vasodilatory response to Ach is present in one third of old SVG. Dilative response to Ach 50 is better preserved in SVG with smaller difference between graft/grafted artery diameters. Adequate matching of the graft/grafted artery diameters probably preserves the endothelium-dependent dilative response of the graft.

Key words: saphenous vein grafts, endothelial function, vasomotor tone

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Introduction

The obstruction of saphenous vein grafts (SVG) is the main cause of recurrence of angina and myocardial infarction (MI) in patients after coronary artery bypass grafting (CABG) [1, 2]. It is still unclear whether SVG possess endothelium-dependent activity and vasomotor properties. Published data are sparse and the results of experimental and clinical studies are inconsistent [3, 4].

The aim of the study was to evaluate in vivo endothelium-dependent reactivity in the region supplied by non-stenotic SVG. The intention of the authors was also to create an easily performed physiological test to access not only morphology but also the function of the graft.

Methods

Patients

The study group consisted of 31 patients (25 men, 6 women, mean age 58.7±8.2 years) referred for coronary angiography due to recurrence of angina. The time from CABG surgery to repeated coronary angiography was 54.0±37.8 months (range 6-144 months). Table I presents clinical details of the study group. We studied 31 saphenous vein grafts: 16 to the left anterior

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descending, 10 to the circumflex, 3 to the diagonal and 2 to the right coronary artery.

Study protocol

The protocol was accepted by the District Ethics Committee and all patients gave informed consent to participate in the study. In each patient one angiographically non-stenotic SVG supplying coronary artery free of significant lesions distally to graft anastomosis was selected for the study. Patients who had received long-acting nitrates during the last 17 hours were excluded from the study. Endothelium-dependent reactivity of the graft and supplied artery were examined using acetylcholine (Ach) (Acetylcholinum, Dispersa), which was administered through the catheter to the ostium of the graft as a 50 μg infusion (time of infusion 30 seconds), similarly to the method reported by Yasue et al. [5]. As shown by Ludmer et al., in normal coronary arteries Ach exerts a vasodilatory action (mediated by the endothelium and nitric oxide), but, paradoxically, it causes spasm of atherosclerotic arteries [6]. In the present study, changes in the diameter of the graft and grafted artery were assessed in relation to the respective initial values using quantitative coronary angiography (QCA) (edge detection, Hicor-TOP 1.5) and a non-ionic contrast medium (Ultravist, Schering), 30 seconds after the end of an Ach infusion. The graft diameter was measured at four distant sites and the mean of these measurements was used for further calculations.

Biochemical parameters which could potentially influence the patency of grafts were also measured. These included lipid levels, lp(a), fibrinogen, platelets, antithrombin III, lipid-lowering medication, and time after surgery.

Statistical analysis

All data are reported as mean values ±SD or as percentages. Student's paired t-test was used for analysis of vessel diameter changes before and after acetylcholine, ANOVA with Dunnett's tests for comparison of mean values between three groups. Multivariate stepwise linear regression analysis was used to define significant determinants of graft diameter changes.

Results

Different vascular reactions in the supplied region after infusion of 50 μg Ach into the graft were observed.

Graft response

In 17 (54.8%) pts (group I) we found graft diameter reduction from the mean 3.8±0.7 to 3.2±0.7 mm (–14.5%) p=0.0001 (Figure 1). In 4 (12.9%) pts (group II)

Table I. Study group clinical characteristics (n=31)

Unstable angina	–	7 pts	(22.6%)
MI after CABG	–	8 pts	(25.7%)
Diabetes	–	3 pts	(9.7%)
Hypertension	–	12 pts	(28.7%)
LVEF	–	43.9±16%	
Total cholesterol	–	220±31 mg%	
LDL cholesterol	–	122±24 mg%	
HDL cholesterol	–	48±13 mg%	
Lipoprotein (a)	–	38.1±41 mg%	
Medical treatment (vasoactive drugs)			
β-blockers	–	22 pts	(71%)
Calcium-blockers	–	11 pts	(35%)
ACE inhibitors	–	16 pts	(51%)

Abbreviations: MI - myocardial infarction, CABG - coronary artery bypass grafting, LVEF - left ventricular ejection fraction, ACE - angiotensin-converting enzyme

Table II. Age of grafts in different types of response to acetylcholine 50 μg

Response	n (%)	mean graft age (months)	p value
Contraction - gr. I	17 (55)	50.6±35	p <0.05 gr I *vs* gr II
No response - gr. II	4 (13)	103.2±36	
Dilatation - gr. III	10 (32)	37.1±20	p <0.05 gr II *vs* gr III

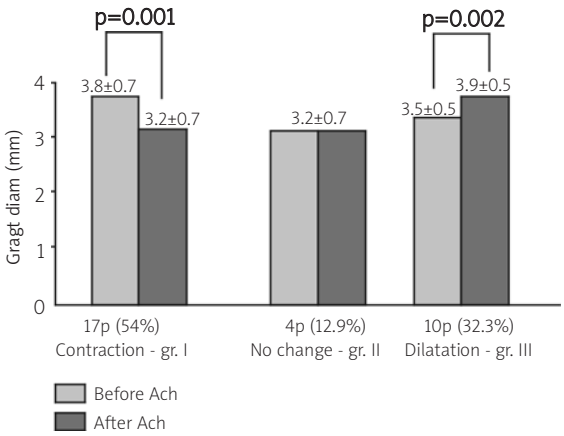

Figure 1. Graft diameter response (acetylcholine 50 μg)

Table III. Saphenous vein graft dilative response modifiers (multivariate stepwise linear regression analysis)

	Regression coefficient	Standard error	p value
Graft/Artery diameter ratio	–22.5	5.83	0.002
Hypertension	–11.86	4.82	0.03
HDL–cholesterol	0.67	0.16	0.001
MI after CABG	–14.8	4.66	0.01

Total cholesterol, LDL-cholesterol, platelets, fibrinogen, lipid-lowering medication, antithrombin III and time after bypass surgery did not contribute to the regression model

there were no graft diameter changes (3.2±0.7mm), whereas in 10 (32.2%) pts we found graft dilatation from the mean 3.5±0.5 to 3.9±0.5 mm (+8.6%), p=0.0002. Time from CABG surgery to angiography is presented in Table II. There was no significant difference in the age of contracting and dilating grafts. We found that stiff grafts (group II) were significantly older than both groups of responding grafts (p <0.05).

In a multivariate linear regression analysis we tested which clinical and biochemical factors from our database are modifiers of the graft dilative response (Table III). We found that SVG dilatation was positively correlated with a low ratio of graft/artery diameter (p <0.002), increased HDL-cholesterol level (p <0.001), absence of hypertension (p <0.03) and absence of MI in the postoperative period (p <0.01).

Native coronary artery response

The response of native coronary arteries downstream to the graft anastomosis was practically uniform. In 29 (93.5%) pts investigated arteries constricted with nearly 50% diameter reduction from 1.2±0.5 to 1.1±0.5 mg (p=0.0001). Only in 2 pts did we find Ach induced dilatation of the supplied artery. The reaction of the graft was also dilatation in these 2 cases.

Discussion

The long-term function of neoendothelium, vasoactive properties and natural history of SVG degeneration have not been sufficiently investigated. Usually these grafts are considered to be passive vessels (stiff conduits) connecting the aorta to a coronary artery [1, 2]. In studies of Hanet et al., and Mulcahy et al., neither Ach nor nitroglycerin produced significant changes in the diameter of venous or arterial grafts [7, 8]. In contrast, Berglund et al. in an intravascular ultrasound study reported an endothelium-independent vasodilatory effect of nitroglycerin in SVG [9]. Previous studies indicated that due to mechanical damage, ischaemia, abrupt exposure to high arterial pressure and pulsative flow, the endothelium-dependent action of Ach in SVG is lost early after surgery and later does not recover [4, 10, 11]. On the other hand, Ku et al. examined *in vitro* old (7 months – 12 years) SVG harvested during reoperation and found that Ach causes relaxation of these vessels [12]. In 1998, Nishioka et al. for the first time reported NO activity *in vivo* in blood samples obtained from the distal portion of SVG, which was similar to the activity obtained from internal mammary grafts [13]. In their study low-dose Ach (5 µg) stimulation did not increase SVG NO activity; nor did it change grafted artery diameter. In contrast, in mammary grafts, after similar Ach stimulation, they reported a 1.8±1.07 fold increase in NO activity and vasodilation of the grafted artery.

Our study clearly demonstrates the various functional status and reactions of the vascular bed supplied by SVG. The present study reports in vivo the presence of Ach-induced vasodilation of SVG in 10 (32.3%) of 31 pts and confirms the results of in vitro experiments of Ku et al. [12]. It is an important finding that endothelium-dependent NO activity and vasodilatory capacity may recover in some SVG and be present as late as 3-5 years after surgery. As NO inhibits platelet aggregation and leukocyte adhesion and protects endothelium from superoxide radicals, such grafts may be functionally privileged [13]. The strongest modifier of the graft dilative response in multivariate analysis was the ratio of graft/grafted artery diameter (p=0.002). We think that a large disproportion between graft and grafted artery diameters increases sheer stress in the graft and impairs its vasoactive properties.

Another interesting observation in the present study is that grafts not *responding* to Ach tended to be older than those with vosoactive capacity. It is tempting to speculate that in some cases after 7-8 years thickening and fibrosis of media and intima may be so advanced that neither dilative nor constricting stimulation could change the graft diameters. Probably because of the *overlapping* of the age of dilating and contracting grafts in multivariate analysis, the age of the graft was not found to be an independent predictor of the graft dilative response.

Limitations of the study

The first limitation is that investigated grafts supplied different vessels and therefore areas of myocardium. In contrast to many previous studies patients remained on vasoactive drugs during the study because our intention was to maintain and investigate the true basal vasomotor tone of the graft. The dose of

Ach is often controversial in flow studies. Initially we administered consecutive incremental doses of 25 and 50 µg. The dose of 25 µg did not cause any significant changes of vessel diameter. The dose of 50 µg was chosen in order to simplify the methodology and avoid false results. This dose was also used by other authors [8, 11]. Saphenous vein grafts and the supplied artery are functionally united and in such a type of study in the cath lab it is difficult to separate the reaction of the graft from the influence of downstream area reactions. However, in our study we observed the same reaction of the grafted artery in nearly all patients but different types of response of the grafts, which encourages us to consider graft diameter change as an independent reaction.

Conclusions

Endothelium-dependent vasodilatory response to Ach is present in one third of SVG. Dilative response to Ach 50 µg is better preserved in SVG with a small difference between graft and grafted artery diameters. Adequate matching of the graft's size to the size of the grafted artery is probably an underestimated factor which preserves the endothelium-dependent dilative response of the graft.

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Zależna od śródbłonka odpowiedź zapalna żylnych pomostów aortalno-wieńcowych spowodowana acetylocholiną

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Streszczenie

Wstęp: Dotychczas opublikowano niewiele prac oceniających funkcję śródbłonka żylnych pomostów aortalno-wieńcowych (SVG), chociaż powszechnie uznaje się, że ma ona decydujący wpływ na czas ich funkcjonowania.

Cel: Ocena *in vivo* odległej, zależnej od środbłonka wazoreaktywności angiograficznie prawidłowych SVG.

Metoda: Badaną grupę stanowiło 31 chorych w wieku średnio 58,7±8,7 lat, skierowanych na badania hemodynamiczne z powodu nawrotu dolegliwości wieńcowych średnio 54±38 mies. po operacji tętnic wieńcowych. U każdego chorego badaniu poddano jeden SVG. Zależną od śródbłonka wazoreaktywność pomostu badano za pomocą oceny, metodą angiografii ilościowej, zmiany średnicy SVG po bezpośrednim podaniu do pomostu 50 µg acetylocholiny (Ach) w bolusie.

Wyniki: U 17 chorych (54,8%) po podaniu Ach stwierdzono zwężenie światła SVG ze średnio 3,8±0,7 mm do 3,2±0,7 mm (p=0,0001), a u 4 chorych (12,9%) średnica SVG nie uległa zmianie (3,2±0,7 mm). U 10 chorych (32,3%) pod wpływem Ach doszło do istotnego poszerzenia światła pomostu z 3,5±0,5 mm do 3,9±0,5 mm, (p=0,0002). W analizie wieloczynnikowej regresji liniowej poszerzenie się SVG pod wpływem Ach było skorelowane z mniejszą dysproporcją pomiędzy średnicą SVG a średnicą pomostowanej tętnicy (p <0,002), wyższym poziomem cholesterolu HDL (p <0,001), brakiem nadciśnienia tętniczego (p <0,03) i przebytego zawału serca (p <0,001).

Wnioski: Zależna od śródbłonka, rozkurczowa reakcja na Ach występuje w 1/3 żylnych SVG. Właściwy dobór średnicy pomostu żylnego, tak aby był zbliżony do średnicy pomostowanej tętnicy, jest prawdopodobnie niedocenianym czynnikiem wpływającym na dłuższe zachowanie zależnej od śródbłonka reakcji rozkurczowej SVG.

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