Myocardial protection by collateral vessels during experimental coronary ligation: A prospective study in a canine two-infarction model

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Summary

Previous work of this laboratory has shown that collateral flow can be increased over six weeks by a subcritical external constriction of the circumflex artery causing a 50 ± 10 % reduction of postocclusive reactive hyperemia. To investigate collateral function in acute myocardial infarction, the model was used to ligate two distant coronary branches on the ventricle simultaneously in order to compare in 8 dogs infarct size and perfusion area of the ligated vessels in control and collateralized sections. The acute collateral flow measured 7.2 ± 2.5 ml/100 g/min⁻¹ and increased to 17.3 ± 6.7 (p < 0.001) over 6 weeks. Separate analysis revealed a predominant increase of collateral flow in the epicardial layers 23.1 ± 4.7 (p < 0.01) versus 6.9 ± 2.8 (p < 0.01) in the subendocardium. Infarct size in the control area was 52.0 ± 14.7 % of the perfusion area, in the collateralized zone 19.0 ± 14.2 % (p < 0.001). Infarct size expressed as per cent of perfusion area and collateral flow in the area at risk expressed as per cent of flow of normal sections correlated: (r = 0.76; p < 0.05). Therefore, infarct size after a 6 hour coronary occlusion can be considered a function of the collateral flow over normal perfusion ratio. Localized induction of collaterals in this model caused a significant reduction of infarct size in relation to the perfusion area at risk.

Key words: collaterals, myocardial protection

Introduction

Previous work from our laboratory has shown that collateral growth can be induced in a relatively standardized manner in the dog (1). An external constrictor was placed on the circumflex artery in order to reduce post occlusive reactive hyperemia by 50 % . This intervention, after a time interval of 6 weeks, resulted in an increase of the functional collateral flow of approximately 100 % in the poststenotic myocardium as measured in previous studies (2). Post-mortem angiographic studies indicated that large epicardial collaterals developed predominantly in areas close to the apex, where large epicardial branches of LAD and circumflex artery lie in relative proximity. Interestingly it became obvious that the myocardial section, where the development of collaterals was to occur, appeared to be limited to the anatomical site of the myocardium that could be identified by acute temporary occlusion of the vessel that was constricted in the course of the experiment, whereby the lateral borders appeared to remain relatively constant. In addition, an endoepicardial gradient of the perfusion provided by collaterals in favour of the outer layers was prevalent in these experiments (3).

Based on the evidence that collateral flow could be induced in a relatively standardized way and on the fact that the development of collaterals was limited to the poststenotic
section of the canine myocardium, we felt encouraged to use a two-infarction model, as previously described by Schaper (4), in order to simultaneously induce two separate ischemic regions that were anatomically separated in the same heart muscle. In this model one infarction was induced in the area that was considered poststenotic "collateralized" myocardium, the other served as control.

If the extension of the perfusion area of an occluded vessel and the O₂-demand of the myocardium at risk are the two major determining factors of infarct size, this model should be capable of giving information on the role of the collateral flow during the development of ischemic myocardial damage. Under the provision that perfusion area, infarct size, and flow can be measured with convincing accuracy, this model should provide answers to the question of a threshold flow that might be capable of protecting the myocardium from ischemic damage under a given experimental condition, including peripheral circulatory demands and myocardial O₂-consumption.

Material and methods

Eight bastard dogs of both sexes, weighing 25-30 kg were premedicated with Piritramide and anesthetized with Pentobarbital Na, 16 mg/kg body weight, 15 min later. The anesthesia was maintained after intubation with a N₂O-O₂, 4:1 mixture under utilization of a volume cycled respirator (Engström®). Additional analgesia was provided through a continuous microinfusion of Piritramide 2-5 mg/kg/h, whenever this was necessary.

A left lateral thoracotomy was used for access. The pericardium was opened and the circumflex artery dissected free of fat and connective tissue over a distance of 1-1.5 cm distal to the LAD. A snare and an electromagnetic flowmeter were attached to the circumflex artery. The flow was measured at rest and after 40 sec of temporary occlusion. Subsequently a Teflon cuff, 0.5 cm long with an inner diameter of 2-2.5 mm was slipped over the empty, collapsed circumflex artery. Reexpansion of the artery provided secure fixation of the constrictor. Flowmeter measurements at rest and during reactive hyperemia were used to ensure normal flow at rest and to verify a reduction of post occlusive reactive hyperemia by approximately 50%. When these conditions were not met, the constrictor was exchanged accordingly.

Radioactive microspheres were injected during acute temporary occlusion in order to determine the acute collateral flow in the poststenotic area and the control flow in the perfusion area of the LAD. Subsequently the thoracotomy was closed. The animals were observed in a recovery area over a period of 24 hours and provided with pain medication as needed (Piritramide 20 mg i.m.). After one day they usually had resumed spontaneous intake of fluids and solid food and could be returned to the animal facilities. Six weeks later the animals were reanesthetized and the thoracotomy was repeated.

A proximal diagonal branch of the LAD and a distant marginal branch of the circumflex artery that did not show any superficial connecting anastomoses were selected and prepared free of connective tissue over a distance of 3-5 mm. Both branches were occluded simultaneously by ligation.

A microsphere injection was given into the left atrium 1 hour after occlusion. Six hours post occlusion the hearts were excised and the coronaries injected with a BaSO₄ solution at a pressure of 100 mm Hg. After slicing of the ventricle in 5 approximately 1-cm-wide slices, post mortem macrohistological p-NBT staining was performed (5, 6). In addition, angiographs were obtained of each slice of the myocardium. Infarct size and perfusion area were determined by planimetry and superimposition of angiograms and color photographs of the slices stained by p-NBT as described earlier (7). The cutting of the slices for counting and discriminative detection of the microspheres in order to reconstruct the topography of flow during the first and second sessions of the experiment was performed according to methods published earlier by this laboratory (8, 9).

Results

The surface of the perfusion area was determined by planimetry on the angiograms for both the "collateralized" and the control regions and was comparable in size with 17.6 ± 5.3 cm² in the control and 16.3 ± 8.9 cm² in the post stenotic area. Under the provision that the thickness of all slices that