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Developments in restenosis

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Abstract

Restenosis is a major complication leading to the failure of vascular procedures, including surgery, angioplasty and stenting. Major efforts including over 100 clinical trials have been made to overcome this complication, with little success to date. Issues relating to trial rationale, design, measurement and biology are addressed in this review.

Introduction

The failure of initially successful vascular procedures is one of the most vexing problems in cardiovascular medicine. Roughly 25-40% of such procedures fail within a year after an apparent success, and this rate is remarkably consistent for all vascular procedures, surgery, angioplasty, laser angioplasty, stent and stent graft; and in all vascular territories studied; coronary, renal, iliac, femoropopliteal carotid, and upper limb arteriovenous dialysis fistulas.

Failure is due to re-narrowing of the lumen restored by vascular reconstruction, or restenosis. Such narrowing can be thought of as comprising three principal components;

1. initial constriction; vasoconstriction in vein grafts, and recoil after stretch with balloon angioplasty
2. intimal hyperplasia; smooth muscle cell appearance, proliferation and matrix formation in the intimal part of the operated vessel
3. remodelling; poorly understood mechanisms which can constrict or widen the hyperplastic vessel wall.

The relative importance of these restenotic mechanisms is thought to vary widely in different clinical situations. For example, use of a stent is thought to abolish the initial recoil constriction after balloon dilatation.

There have been over 140 randomised clinical trials of therapies to prevent human restenosis, chiefly after coronary artery angioplasty or stent. There may be a similar number of unpublished trials, distinguished only by the flood of acronyms used up and made unavailable to future investigators. The results of the overwhelming majority of restenosis trials are negative. There have been a few positive trial results, but none of these agents has progressed into widespread clinical use for various reasons, most commonly that an initial trial with small numbers looked promising, only for hopes to be dashed by the negative result of the larger, more controlled follow-up trial. Another curious feature of positive restenosis trials is that

the treatment group demonstrated a stable, expected restenosis rate of 25-40%, but the comparison control group developed higher than expected rates of restenosis, so that the improvement seen, although statistically impressive, was more apparent than real.

The range of negative trials include almost any cardiovascularly active agent that can be imagined. This illustrates both the clinical importance of the problem, as companies continue to be willing to invest money in this field, and also the magnitude of the failure of the empirical approach.

Why are trials mostly negative?

All the restenosis trials proceed along conventional lines. Activity of the agent is demonstrated in animal models of intimal hyperplasia after vascular injury, a human protocol is set up, with limitations of toxicity and routes of administration and the trial then proceeds. But why are there so many negative trials, if the approach is unexceptionable?

Animal results translate badly into the clinical restenosis scenario. Vascular injury and healing is part of normal pathophysiology and is likely to be conserved, in an evolutionary sense, between mammals, possibly between all vertebrates. Results in pig, mouse or rat should not be wildly variant from man. However, most animal models are based on injury of normal animal arteries, particularly balloon injury of the rat carotid, and this does not reflect the human situation, where an atherosclerotic artery is the target, or a normal vein used as a bypass graft. Injury of a normal animal artery rarely produces a reaction in the wall which develops into a significant restenosis of the arterial lumen. It is likely that extrapolation of effects on normal vascular healing in animals to human restenosis may well have been a hopeful, rather than an evidence-based step. Development of atherosclerosis-prone animal models for testing vascular injury models is a step in the right direction.

There has always been a question over the possibilities of both false negative and false positive errors in restenosis trials. This results from the dominance of coronary angioplasty in these clinical studies, and the technique of quantitative coronary angiography (QCA) used as the chief measurement. The definition of restenosis according to QCA measurements has been established as a 50% loss of the initial gain in minimum luminal

diameter. This definition is flawed, as substantial narrowing of a large diameter gain may not result in a significant decrease in blood flow, whereas even minor narrowing may produce major blood flow diminution where the initial gain was small. It also cannot apply to non-angioplasty situations, such as bypass grafts. Simple measurement of minimum luminal diameter has attempted to address these shortcomings; however, this measurement has drawbacks of its own.

More importantly, there is a potential for error at the very heart of the QCA process. The standard deviation for QCA measurements is in the order of 0.36 mm, in a vessel of 1–2 mm diameter.¹ However, the differences between treatment and control groups in clinical trials can be as little as 0.11 mm (in the well-known Stent Restenosis Study [STRESS], which showed an advantage of stenting over angioplasty alone),² well below the error of measurement. Currently, simple tests of statistical significance do not take into account variation due to measurement; this leads to the debate on differences being 'clinically significant' rather than 'statistically significant'. However, it can be recognised that both false positive and false negative errors can be generated where the differences detected are close to, or even less than, the measurement error. This is particularly important in trials on small numbers of patients. In a blinded trial, there should be no systematic error and the positive and negative errors should even out over a large group of patients. This simple numerical consideration may largely explain the frequent occurrence of positive results in small trials, later negated by a larger, more controlled study.

Frustration with the number of negative trials has inevitably led to erosion of the definition of restenosis. The angiopeptin trial, using a peptide somatostatin analogue to prevent restenosis after coronary angioplasty, first noted a divergence between effects on restenosis (no improvement) and on clinical events around the angioplasty (improved).³ This has led to a second definition of restenosis, defined by the absence of adverse vascular events after the procedure, chiefly myocardial infarction, recurrent angina, secondary revascularisation procedure and death. Clearly this weakens the 'pure' restenosis definition and represents a movement of goalposts. Proponents, however, would argue that this change represents greater clinical utility, and has the benefit of not requiring a second, follow-up invasive angiogram. Many trials are now collecting both angiographic and clinical data and presenting both definitions. Perhaps the ideal definition of restenosis, a decrease in blood flow over time, is just around the corner, with developments in ultrasound and MR angiography.

Probably the most contentious feature of the restenosis literature is the number of 'false dawns' previously alluded to. An initial positive trial leads to a great wave of intemperate enthusiasm, proclamations that restenosis is no longer a limitation of vascular intervention and favourable movements in stock market prices. Over a period of time, this

Table 1 Results of lovastatin studies; discrepancy between early results in hypercholesterolaemic rabbits and definitive multicentre trial in patients.

1991	"When the number of vessels restenosed was considered, only nine of 72 vessels (12.5%) restenosed in the lovastatin group, compared with 13 of 34 vessels (38.2%) in the control group ($p < 0.002$). Similarly, 10 of 80 (12.5%) PTCA sites restenosed in the lovastatin group compared with 15 of 36 (41.7%) in the control group ($p < 0.001$)." ⁴
1994	"Treatment with high-dose lovastatin initiated before coronary angioplasty does not prevent or delay the process of restenosis in the first six months after the procedure." ⁵

enthusiasm dissipates, as longer follow-up, better trials, or less selective clinical practice dispels the initially favourable impression. The agent or technique then either passes into disuse, the commercial sponsor losing interest, or it stays in use, but with far lower expectations.

There have been many false dawns; the cases involving lovastatin, heparin-bonded stents, and brachytherapy are characteristic in different ways.

Lovastatin was one of the earliest statins used for the treatment of hyperlipidaemias. Following encouraging results in reducing vascular injury lesions in hypercholesterolaemic rabbits, a small positive trial was reported, triggering a larger, multicentre study. This definitive study was resoundingly negative, providing a canonical example of the sequence described earlier in this review (Table 1). The most recent example of the sequence involves tranilast, a putative growth factor inhibitor; the definitive Prevention of Restenosis with Tranilast and its Outcomes (PRESTO) trial, which proved negative, apparently involved 11,500 patients and an expenditure of over \$100 million.

Brachytherapy, defined as the use of internally applied radiation therapy, is a large field in vascular medicine, and it is probably unfair to consider all devices, isotopes and delivery methods together as a bundle. However, these quotes from the Scripps trial, one of the earliest in the field, illustrate the changes in expectation between an initial optimism and the more sober three-year results, where the improvement in restenosis in the treatment group is dependent on a very high rate in the control group (Table 2). Much of the change from early optimism to a more guarded viewpoint came from the realisation that, although radioactive stents reduced the intimal proliferation response within the stent, there is an edge effect, where restenosis occurs at the proximal and distal stent borders, with the same clinical outcome as restenosis within the stent. Realisation that there is a late failure rate in radioactive stents, and uncommon complications such as coronary aneurysms, may further limit the role of intravascular brachytherapy, although there are technical developments in the industry to try and overcome these problems.

Heparin-bonded stents were used in the Stents in human coronary arteries (BENESTENT II) trial to reduce the anticoagulation regime associated

Table 2 Quotes from the Scripps trial of bradytherapy.

1997	An independent, blinded analysis performed off-site showed a 60–80% reduction in all the angiographic indexes of restenosis. Notably, the late-loss index, a sensitive measure of the proliferative response to injury, was reduced from 0.6 to 0.12 ($p < 0.01$). Thus, of more than 50 clinically tested therapeutic agents, gamma radiation with the use of iridium-192 is one of the first found to reduce the rate of restenosis after coronary angioplasty. ⁶
2000	The dichotomous restenosis rate at three-year follow-up was also significantly lower in (192)Ir patients (33% vs. 64%; $p < 0.05$). ⁷

with stenting at the time. The effect on restenosis was noted in the preliminary report, in which a very low 6% rate was reported in their protocol-finding subgroup.⁸ However, by the time the definitive report was produced, this rate had increased to 16%,⁹ while other groups have now reported rates of 22% and above.¹⁰ The authors very properly guarded against excessive initial optimism by stating "the very low rate of restenosis observed seems promising and needs confirmation in a larger randomised approach", but this did not stem the enthusiastic interpretation of their data by many other groups, until the publication of more definitive results.

Currently, rapamycin/sirolimus-bonded stents are going through the same process. Very low rates of restenosis in early trials have prompted statements attributing virtually zero restenosis,^{11,12} and a complete cure for this condition, although the data are still early. Whether this agent will prove to be the great breakthrough or yet another false dawn remains to be seen.

How is the renin-angiotensin system involved?

The renin-angiotensin system (RAS) has been implicated in restenosis for some time. Following very preliminary animal work, one of the first multi-million dollar multicentre restenosis trials^{13,14} was set up in both Europe and America, to study the effect of cilazapril on restenosis after coronary angioplasty. The familiar negative result did much to dampen enthusiasm (and funding opportunities) in the succeeding years.

More recently, the angiotensin II receptor blocker (ARB), valsartan, has been reported as favourably influencing restenosis in a small (200 patients) trial of coronary stenting. The results were reported as showing a reduction in restenosis from 38.6–19.2%.¹⁵ Again, these data need to be interpreted conservatively, in view of the concerns outlined earlier.

Experimental data in animals indicated that angiotensin-converting enzyme (ACE) and angiotensin II stimulate vascular smooth muscle cell (VSMC) proliferation. As ACE levels are partly controlled by a deletion/insertion (D/D) polymorphism on chromosome 17, a link was sought between the polymorphism and restenosis. After the first report that the D polymorphism of the

ACE gene was associated with restenosis after coronary stenting,¹⁶ further negative reports could find no such association, either after coronary stenting or coronary angioplasty.^{17–19}

This is a controversial area, but as increasing numbers of studies are reporting, the majority find no relation between ACE genotype and restenosis. Clearly a definitive meta-analysis is not far away.

In some subgroup analyses in the ACE gene studies, there did appear to be an effect of the DD homozygous polymorphism. DD patients treated with quinapril in a very small (115 patients) trial of coronary stenting appeared to have higher incidences of restenosis, contrary to expectation.²⁰ Again, this may be a false negative error, and requires conservative interpretation.

Pratt and Dzau, writing in an editorial for *Circulation* in 1996, state "one may conclude, on the basis of these studies, that the renin-angiotensin system does not appear to be a pivotal player in the complex process of restenosis".²¹ Nothing that has happened in the intervening years substantially changes this assessment. They go on to cautiously conclude "strategies aimed at blocking selective biological mediators are limited by the multiplicity of the mediators, the plurality of the cell surface receptors and intracellular signalling mechanisms." In simple terms, there are multiple redundant pathways that can mediate restenosis; targeting a single pathway may not be enough.

What advances can we hope for?

It is easy to be negative in this field. In part, this might be necessary, as a counterweight to excessive optimism that is prevalent everywhere, and the attendant stock market movements that reflect the dominance of commercial concerns in this sector. Indeed, it has frequently been said that the major morbidity of restenosis is economic; most coronary interventions are performed for angina, and restenosis results chiefly in recurrent angina, and then in a second procedure, rather than in more serious outcomes. Clearly, restenosis within a distal bypass graft for limb salvage will have different implications.

Negativity does not disrespect the well-intentioned and hard-working approach of investigators in this field. It is apparent that we are all doing our best, but working within limitations, and the limitations are in the way we view the problem, particular in the equation of physiological vascular healing with pathological vascular restenosis.

Gene therapy

There have been many proposals for applying advances in human genetic manipulation to restenosis. There are some advantages; restenosis is a common and deeply studied process, there is no shortage of patients, there are established clinical trial procedures, and there is relatively easy access for transfection of the genetic agent into the vascular wall.

Many genetic therapies can arrest VSMC proliferation. Dzau's group have shown that genetic manipulation to arrest cell cycle progression in

rabbit vein grafts did not interfere with their integrity or function as bypass conduits. They have conducted studies in human lower limb bypass grafts, using pressure-mediated *ex vivo* transfection. The genetic medication is a decoy sequence which binds and inactivates a transcription factor, E2F, which controls the production of cell cycle mediators, and effectively arrests cell growth. Although their aim was to show predicted effects on the cell cycle, they have been encouraged by the progress of treated grafts, which have so far demonstrated a lower rate of clinical failure, again in an early study with very small numbers.²²

My belief is that human genetic therapy for restenosis is only useful as a proof-of-concept experiment, to demonstrate that a specific agent directed at a specific target would achieve clinically important results. I don't believe it is justifiable to use a therapy with an unknown long-term safety profile to treat a condition that would only have affected a minority of the treated population, with mostly non-fatal consequences.

Human restenosis biology

The differences between human restenosis and animal models have largely been glossed over. Even more so, the differences between human restenosis in the minority of patients and non-restenosis in the majority, have been studied at easily accessible levels, for risk factor stratification, but not much at a biological level. The studies on human restenosis which exist have revealed interesting leads which may lead to better therapeutic targets. Our group described heparin resistance in human VSMC culture from restenoses, and prospectively from bypasses which would later develop restenosis.²³ Heparin resistance is likely to be just the presenting symptom of a far-reaching phenotype alteration that is involved in pathological restenosis, as opposed to more normal vascular healing. The molecular correlates of this condition are just now being established and coming to press. McCaffrey's group described a mutation in the polyA tract of the type II receptor for TGF β in a small number of restenoses;²⁴ but our group and others have not been able to confirm this. Bennett's group have described enhanced apoptosis induced by p53 in restenosis compared with normal VSMC, and a specific defect in cyclin D degradation induced by DNA damage. They believe that this character would render restenosis cells more sensitive to radiation damage and to support the improved results of brachytherapy currently being reported.²⁵

These kinds of restenosis-specific studies are more likely to give insights which improve our understanding of the real problem, and to lessen our dependence on models of animal vascular healing, which have proved inadequate at modelling clinical restenosis.

Measurement technology

Coronary restenosis is not the only clinical area in which restenosis is important. In view of the inherent problems of QCA, there is a need for

developing other methods of studying restenosis. To this end, intravascular ultrasound (IVUS) has been increasingly used in coronary studies. It is possible that IVUS can detect effects that QCA cannot.²⁶ Larger peripheral arteries are more accessible for study, and as the diameter of the vessels is greater than coronaries, measurement error is proportionally less significant. Leg arteries can be studied by serial Duplex ultrasound and by MR angiography. Perhaps the narrow focus of commercially-sponsored trials on the coronary circulation has led to neglect of the periphery, whose characteristics may be less prone to error and more able to deliver definitive answers to the restenosis question.

Conclusions

If zero restenosis is just around the corner, as enthusiasts for the rapamycin/sirolimus eluting stents believe, then much of this article is of historical interest. If it proves to be another false dawn, then it must be concluded that history has much to teach us about the future, that the long view is more honest and less self-serving than blind optimism and faith, and that the restenosis story represents one of the most spectacular examples in science of the First World War approach, in which frontal assaults on a problem, without the requisite advances in knowledge and technology, result in repeated and costly failure.²⁷

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