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EDUTORIAL

Which Mouse Model of Abdominal Aortic Aneurysm Deserves Triple A Status?

Different abdominal aortic aneurysm (AAA) mouse models have been used over the last two decades. Here, the main AAA models will be highlighted, and what has been learned from them will be discussed.

The angiotensin II (AngII) infusion model is the most widely used AAA model. AngII was given to mice susceptible to the development of atherosclerosis to accelerate disease. However, AngII also induced aortic aneurysms, dissections, and ruptures in the ascending, descending, and abdominal aorta.

This AAA model consists of a four week AngII infusion via an osmotic mini pump implanted under the skin.[1] To study drug mediated inhibition of AAA formation, usually hyperlipidaemic (apolipoprotein E or low density lipoprotein receptor deficient) male mice are used to obtain an AAA incidence of 80% in the placebo group. A beneficial effect of the intervention would lower the AAA incidence, aortic diameter, and rupture rate. If the aim is to study a genetic predisposition that may accelerate AAA development, hyperlipidaemia or male sex is not necessary. AngII infusion in wild type (mostly C57Bl6 background) mice results in an incidence of AAA of only 25%, which allows detection of enhanced AAA development in the genetically modified mice of interest. AngII can promote these aortic events independent of its blood pressure function.[2] On a cellular level, AngII induces arterial inflammation, which is the main cause of AAA formation in this model. Massive leucocyte infiltration into the aorta causes severe aortic damage by degrading the extracellular matrix (ECM) and causing smooth muscle cell (SMC) phenotypic switching to respond to injury. Using drugs or genetically modified mice (with mutations), disturbing the ECM or SMC phenotype/health will obviously influence AAA development.[3,4] In contrast to human aorta, the murine aorta consists of just a few SMC layers. Consequently, AngII induced inflammation easily causes aortic dissection and rupture, making this model attractive to study these aortic events as well.

Interestingly, the AAA progresses for as long as AngII is infused. Hence, it is feasible to inhibit *established* AAA to mimic human pathology.[5] In this so called AAA *progression* model, AngII is first administered to induce AAA formation, before intervention is implemented. These experiments take longer, are more expensive, require a surgical change of AngII mini pumps, and are less likely to show significant results on AAA growth. The first progression study was conducted with the antibiotic doxycycline as the intervention. Doxycycline is considered the gold standard to prevent aneurysm formation in any aneurysm model. However, in this study, no beneficial result on AAA progression was observed.[5] Interestingly, similar results were shown in human patients with AAA treated with doxycycline, in whom *enhanced* AAA growth was measured.[6] Thus, even though the AAA progression model is demanding, it may have more value for translation to the clinic.

The second most used AAA model is the elastase perfusion model, where part of the abdominal aorta is surgically isolated to infuse elastase locally. This 5—10 min infusion causes cleavage of aortic elastic fibres (ECM damage) and thus triggers an inflammatory response, with consequences for SMC behaviour.[7] In the first two weeks an AAA will develop, which will disappear on resolution of inflammation. This is a technically challenging model, but it is fast and feasible in any type of genetically modified mouse. Within two weeks you will have the results of the intervention, making it a suitable model for AAA prevention studies, rather than AAA progression studies. However, performing this model in genetically predisposed mice may generate novel advanced models of AAA. A simplified version of this model requires local peri-aortic elastase application, which also results in AAA formation.[8] However, to increase the compatibility of the model to human AAAs, adaptations have been made by more extensive suturing to induce haemodynamic changes.[9]

The third AAA model is the calcium chloride model, which is based on local peri-aortic application of calcium chloride for 15—20 min. Again, this causes ECM damage, SMC apoptosis, and inflammation.[10] During a period of four to six weeks an AAA develops that will also resolve thereafter. Timewise, this leaves room for delayed intervention, studying AAA resolution rather than AAA progression. This model is less difficult and feasible in any type of genetically modified mouse. An adapted version of the model was generated to enhance AAA severity by applying phosphate buffered saline after calcium chloride treatment.[11] This induces vascular calcification, which causes enhanced aortic damage and thus more inflammation.

While these are the three main aneurysm models used, there are other aneurysm models, in other animal species, which are highlighted in a review by Sénémaud *et al*.[12]

Inflammation is the driver of disease in all AAA models, and factors enhancing aortic inflammation influence AAA severity. Consequently, any compound with anti-

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inflammatory properties has been shown to prevent AAA formation/dissections in these models. Yet, in humans, chronic anti-inflammatory drugs actually enhance AAA growth,[13,14] similar to chronic doxycycline treatment.[6] This adverse effect is probably caused by the cytostatic effect that most immunosuppressive drugs and doxycycline have on SMC with long term use.

So, what have we learned from the AAA mouse models? All three models have their own benefits; however, the AngII infusion model seems to outweigh the other models because aneurysms and dissections can be studied at multiple sites throughout the aorta and AAA *progression* studies are feasible. Concerning the translation from bench to bedside, we should, perhaps, reconsider the way we interpret the murine data. In mice, drug treatments of 2–4 weeks are usually extrapolated to 3–6 years in humans, based on lifespan differences. However, inflammatory cell and SMC turnover between mice and humans are quite similar. The drugs are meant to have an effect on the inflammatory cells, while leaving the vulnerable SMC within the AAA alive. Thus, possibly (repeated) short term treatment strategies in patients could be useful, which supports SMC driven aortic repair after resolution of inflammation. Obviously, the AAA models have provided us precious information on AAA development. Yet, the value of (AAA) mouse models is optimal if we understand how to interpret them in the human setting.

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