# Abdominal Aortic Aneurysm Development in a Rat model

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## Introduction

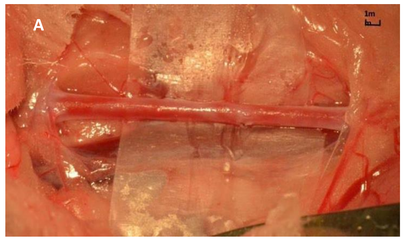
Abdominal aortic aneurysm (AAA) is a progressive chronic dilatation of the abdominal aorta which can result in eventual rupture. The etiology can vary due to factors such as genetics, trauma, infection, high blood pressure, atherosclerosis, and smoking. The disruption of the elastic matrix leading to AAA is generally considered irreversible, due to the inherent inability of adult vascular smooth muscle cells (SMCs) to produce sufficient elastin, the key protein component of elastic fibers, and their inability to organize elastin precursor molecules (tropoelastin) into mature, crosslinked fibers.

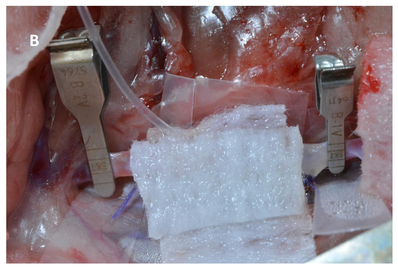
We present data here on a strategy to induce AAA, and the appearance of the vessel after one month. If the treated artery and resultant aneurysm were found to have stabilized or even strengthened, such as through inducement of an intimal hyperplasia response, it would indicate that our model is inadequate in comparison with genuine aneurysms. If our model can be shown to mimic genuine AAA, it will provide a benign platform to perfect possible interventions to forstall catastrophic aortic rupture.

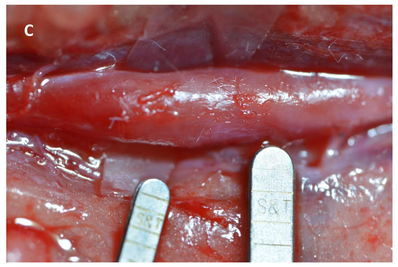
## Materials and Methods

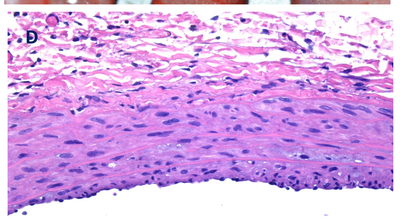
We began our studies with a 1-month treatment protocol, in which chemical and enzymatic treatments (0.5M $CaCl_2$ and 5 U/ml elastase for 30 minutes ) were employed to induce an aneurysm (defined as 50% or more increase in vessel diameter) in adult Sprague Dawley rats. The changes in aorta diameter are determined by comparing pre/post photographs using micro vascular clamps as size references. The aortae were then excised and embedded in paraffin and stained for histologic examination. We are now exploring the state of our aneurysm model at 2-months to confirm the aneurysmal size progression and arterial weakening. We hope to extend the time even further, having obtained permission from the IACUC.

## DATA




**A. Pre-treatment exposure of the abdominal aorta in a Sprague-Dawley Rat.** A sterile piece of parafilm has been placed below the vessel to prevent leakage of the solution of elastase and calcium chloride

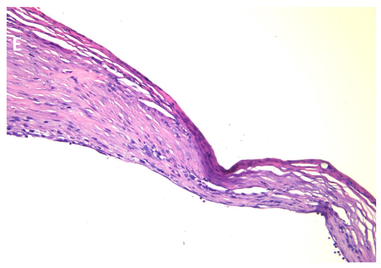
**B. Treatment of the aorta with the solution of calcium chloride and elastase.** Microvascular clamps are used to limit blood flow through the vessel during treatment. The gauze is soaked with the chemicals and kept in place for 30 minutes.

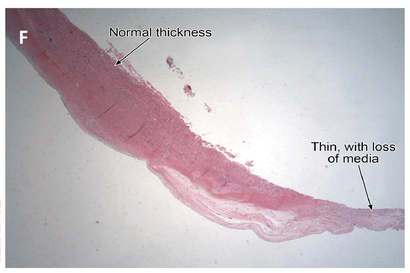
**C. The same vessel after 28 days.** General swelling of the vessel is evident where the solution had been applied. It appears to be similar to an early-stage AAA.

**D. Histology of a normal rat aorta.** This image shows H&E staining of a normal, untreated rat aorta from the same area that would have been treated.

**E. Histology of a vessel exposed to our treatment on day 28.** There is evident thinning of the arterial wall, suggesting a severely weakened vessel.

**F. Image from the literature of histology of a human AAA.** Although more studies are required for proof, there is a striking similarity to our 28-day treatment.




## Results

This study is still in progress and analysis is ongoing. Using an optimized approach, we have achieved increases in vessel diameter of greater than 60% by 1 month. To date, the treatments out to 2-months have not resulted in any ruptures or unanticipated events, but the final data have not yet been collected. Our treatments successfully produced vascular wall thinning and expansion of the lumen along with thickening of the intimal layer. Long term studies are needed.

## Discussion

No effective medical treatment exists for AAA. To date, surgical intervention is the only viable option to treat large, rupture-prone aortas. However, it has recently become technically feasible to deliver chemical or genetic payloads to the vascular walls, either by perfusion of the afflicted area or by direct delivery by contact with a balloon or stent carrying the payload in the form of viral vectors or nanoparticles.

In the next phase of this study, we will continue to examine the progression of the aneurysm out to 3 months and analyze gene expression in the aneurysm to determine whether our creation mimics a genuine aneurysm, and if so, to deliver of genetic payloads to the area using nanoparticles and/or viral vectors. These particles will contain one of several genes known to stimulate the growth of SMCs or to cause cells around the delivery area to secrete elastin, collagen or other fibrous proteins with the intent of strengthening the area and preventing a catastrophic failure. The results of the interventions will be followed by histologic and genetic analysis of the affected area at various times after treatment.

## Conclusion

Developing a method to create genuine AAA will allow us to study genetic interventions to prevent or reverse this condition in the next phase of the study.

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