

The role of 20-HETE in myogenic responses of the afferent arterioles and tubuloglomerular feedback

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Abstract

Previous studies have indicated that 20-hydroxyeicosatetraenoic acid (20-HETE) modulates renal and cerebral vascular tone in large arteries through inhibition of K_{Ca} channel activity but its role in the myogenic response in the afferent arteriole and TGF is unknown. In the present study isolated Af-Art with the attached glomerulus was micro-dissected and perfused from rabbit and mouse kidneys to elicit a myogenic response. Isolated Af-Art and distal tubule with the attached glomerulus was micro-dissected and double perfused from a rabbit for TGF measurements. The luminal diameter decreased by 9.18 ± 0.48% (from 11.91 ± 0.5 mm to 10.95 ± 0.5 mm, n=8) on mice and 8.84 ± 1.31% (from 17.00 ± 0.39 mm to 15.44 ± 0.39 mm, n=5) on rabbit when the perfusion pressure was increased from 60 mmHg to 120 mmHg. Administration of a 20-HETE synthesis inhibitor, HET0016 (1 μM), or a selective 20-HETE antagonist, WIT002 (10 μM) completely blocked the myogenic response. In other experiments, increasing NaCl concentration from 10 to 80 mM in the tubular perfusate to the macula densa constricted the afferent arteriole from 18.2 ± 2.0 μm to 12.2 ± 1.5 μm (TGF=6.0 ± 1.4 μm, n=3). Administration of 20 hete antagonist (Wit002) prevent the TGF mediated constriction. Addition of 20-HETE to the perfused Af-Art restored the vasoconstrictor response to either elevation in perfusion pressure or changes in Na concentration at the MD in the presence of HET0016. These studies indicate a key role of 20-HETE in modulating the responsiveness of the afferent arteriole to TGF and myogenic response and may help explain how deficiencies in the renal formation of 20-HETE promote the development of glomerular disease in salt sensitive forms of hypertension.

HYPOTHESIS

20-HETE modulates the responsiveness of the afferent arteriole to both TGF and myogenic response by blocking K_{Ca} channels.

RESULTS

Isolated Rabbit Renal vessels CYP (cytochrome P-450) enzyme assay

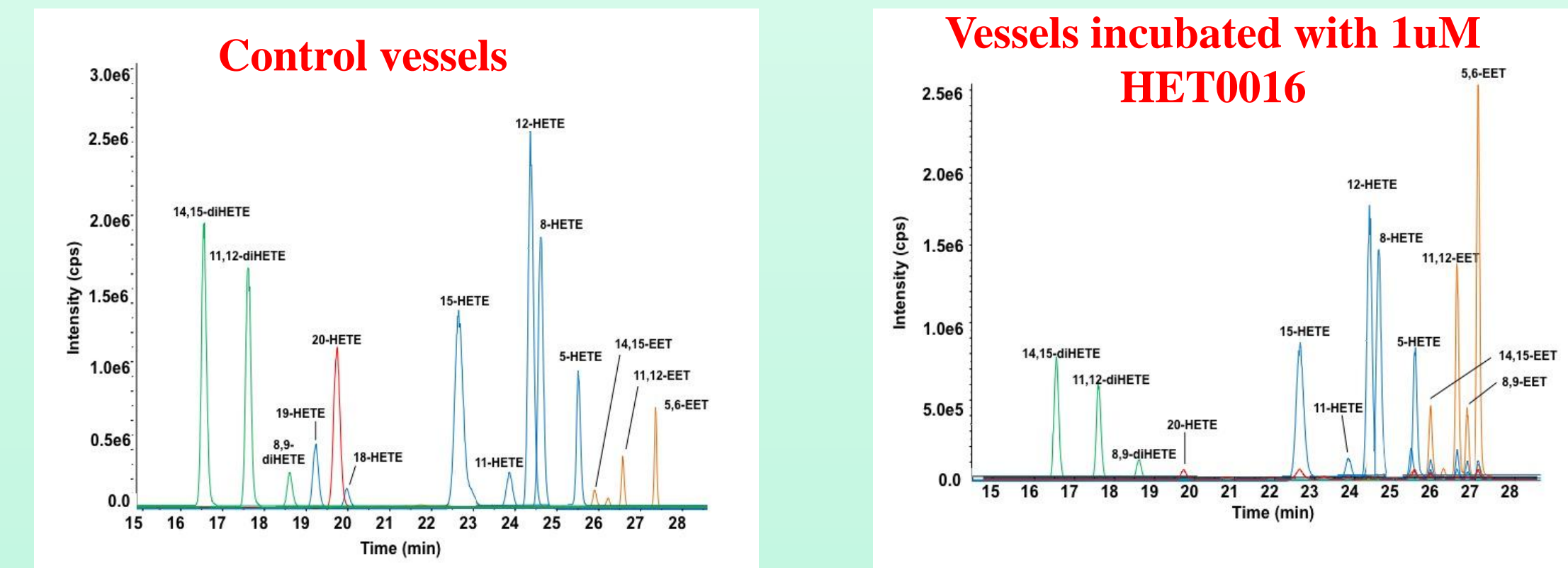


Figure 1. Representative LCMS/MS chromatograms of CYP450 metabolites of AA produced by rabbit preglomerular microvessels incubated with a saturating concentration of AA (40 μM) for 30 min.

HET0016 impairs 20-HETE formation in isolated rabbit vessels

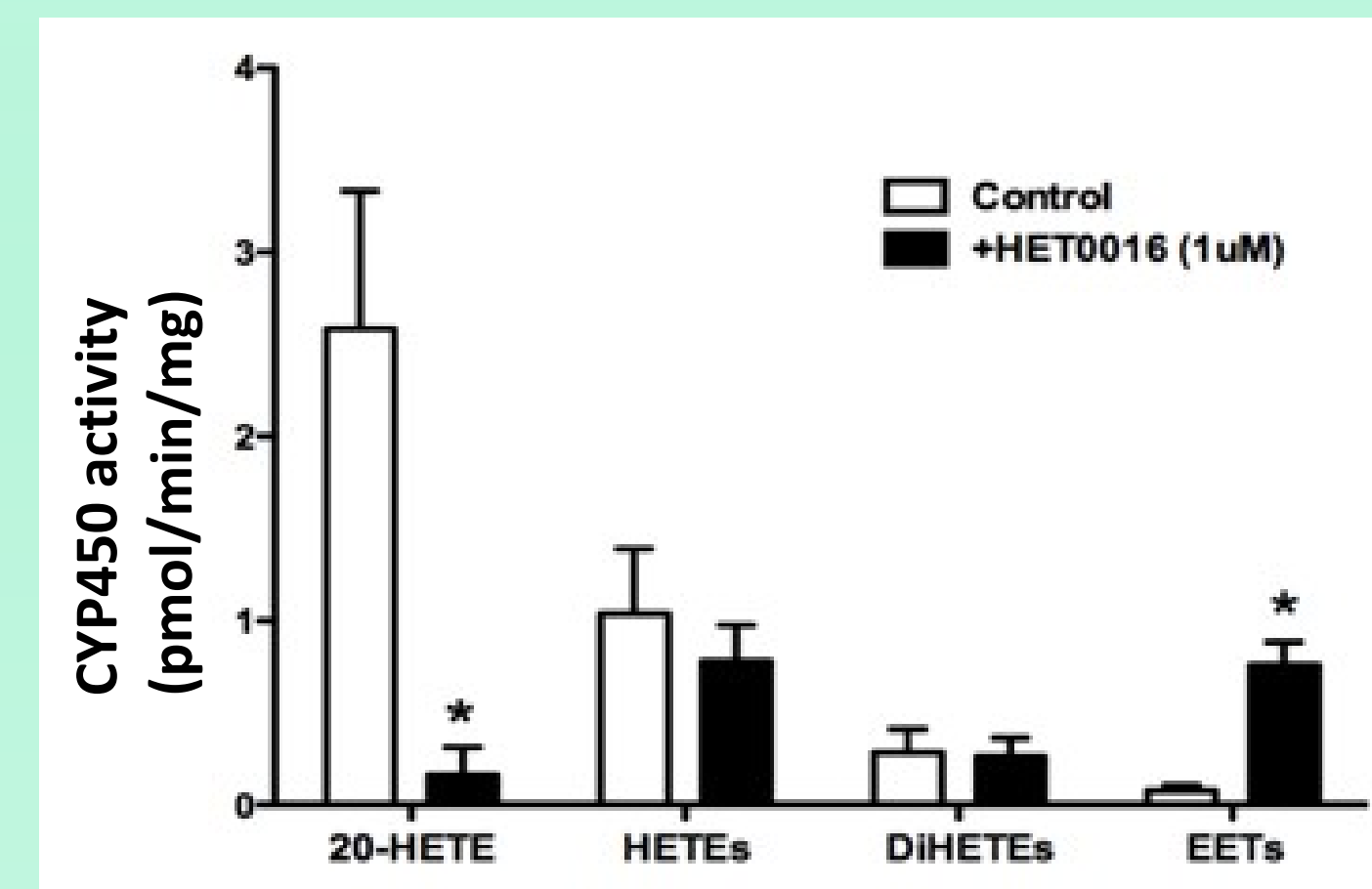


Figure 2. Effects of HET0016 (1μM) on the CYP450-depaendent metabolism of AA to 20-HETE, EETs, DiHETEs, and HETEs by rabbit preglomerular microvessels.

Effects of HET0016 and a 20-HETE agonists on myogenic response of rabbit and mouse afferent arteriole.

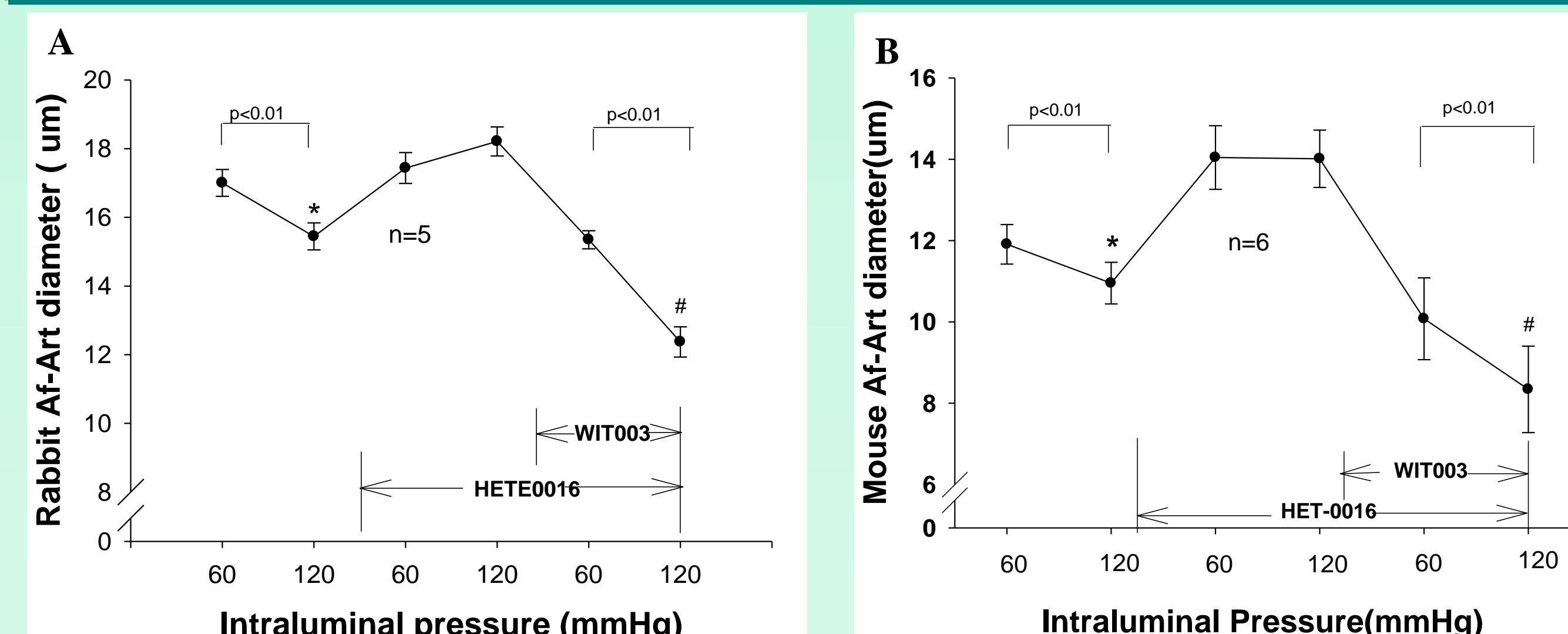


Figure 3. Administration of a 20-HETE synthesis inhibitor, HET0016 (1 μM) completely blocked the myogenic response in both rabbit (panel A) and mouse (panel B) afferent arterioles. Addition of a 20-HETE mimetic restored the vasoconstrictor response and reduced the diameter of mouse Af-Art by 23% and 19% in rabbit Af-Art.

Effect of WIT002 on the myogenic response

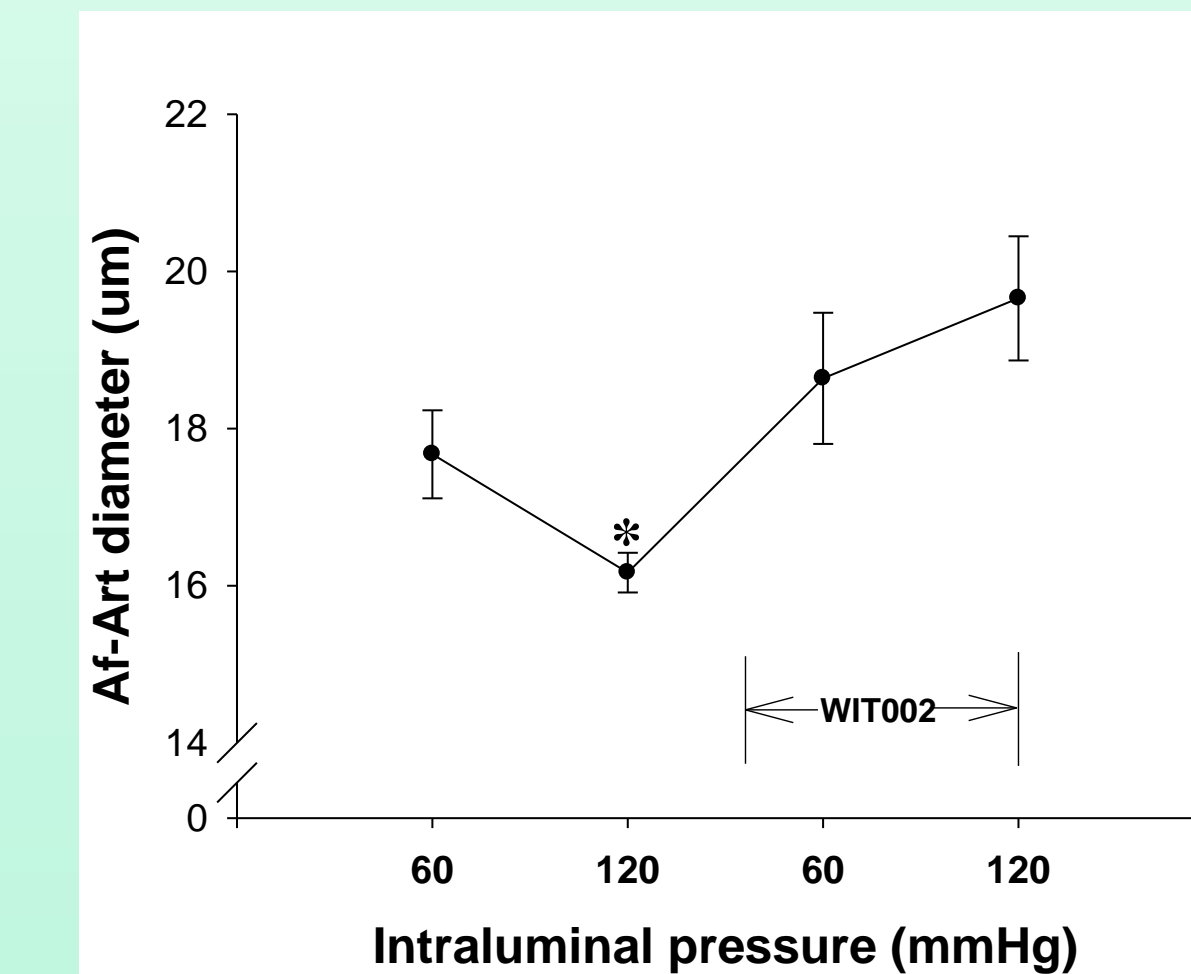


Figure 4. Administration of a selective 20-HETE antagonist, WIT002 (10 μM) completely blocked the myogenic response in rabbit Af-Art.

Effect of Iberitoxin on Myogenic Response

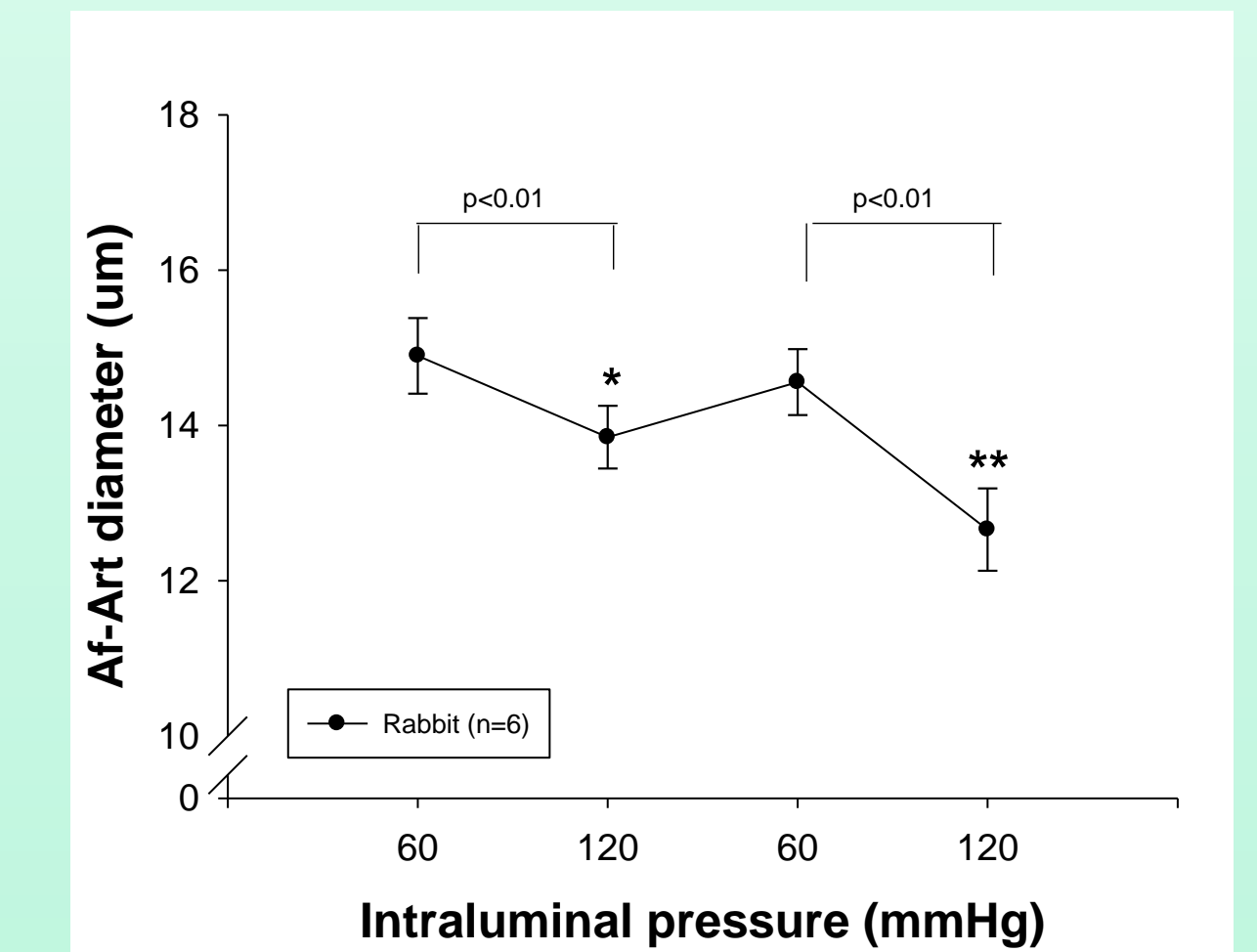


Figure 5. Blockade of K_{Ca} channels with iberitoxin (100nM) enhances myogenic response in rabbit Af-Art.

Effects of WIT002, WIT003 on TGF in the double perfused rabbit Af-Art with attached MD preparation

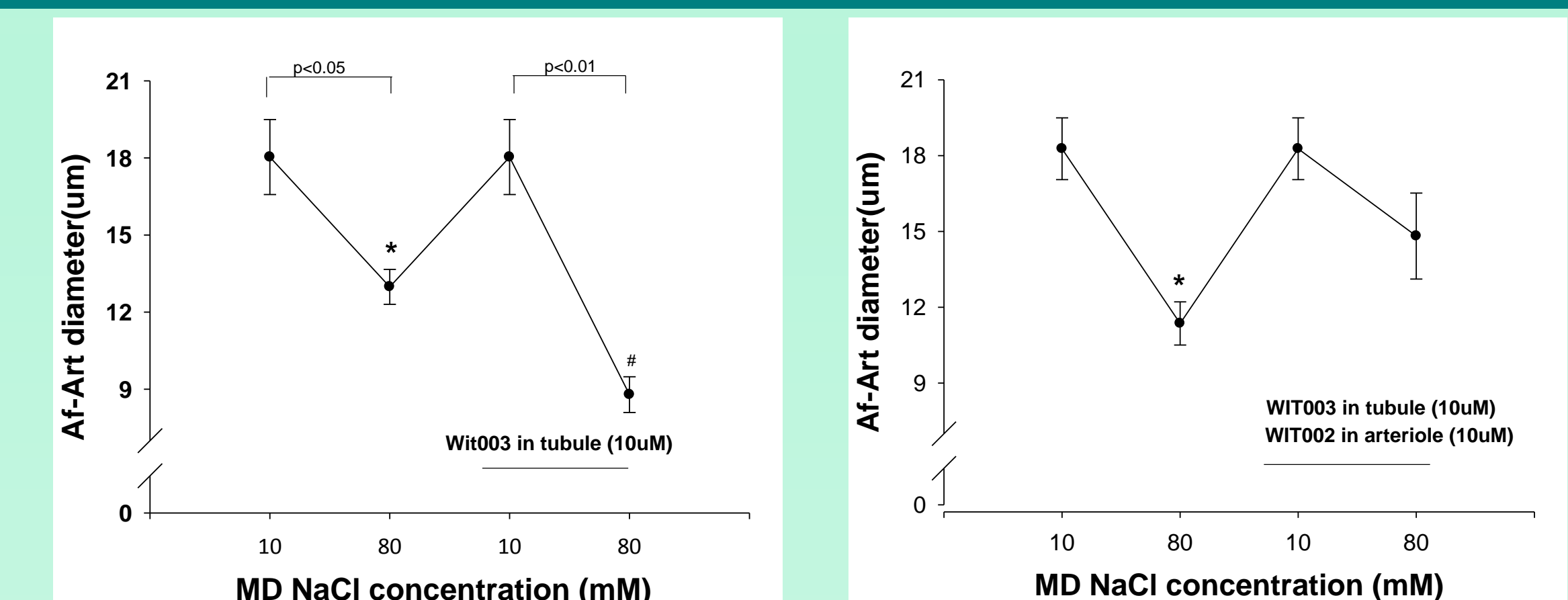


Figure 6. Increasing NaCl concentration from 10 to 80 mM in the tubular perfusate to the macula densa constricted the afferent arteriole of rabbits from 18.2 ± 2.0 μm to 12.2 ± 1.5 μm (TGF=6.0 ± 1.4 μm, n=6). Administration of 20-Hete antagonist (Wit002) prevented, while a 20-HETE agonist enhanced the TGF mediated constriction.

CONCLUSION

