Endothelial nitric oxide synthase and vascular nitric oxide signalling in amphibians

Melissa S. Cameron, Sofie Trajanovska and John A. Donald
School of Life and Environmental Sciences, Deakin University, Geelong, Australia, 3217

In vertebrates, nitric oxide (NO) is generated by NO synthase (NOS) of which there are three isoforms. Neuronal NOS (NOS1) and inducible NOS (NOS2) are common to all vertebrates, but, endothelial NOS (NOS3) is not a ubiquitous NOS isoform in vertebrates. This is reflected in the fact that endothelial NO signalling cannot be demonstrated in amphibians and fishes, and instead, NO released from NOS1in nitrergic nerves mediates vasodilation. Here we characterise NOS3 in the amphibian, *Bufo marinus*. *B. marinus nos3* encoded a 1170 amino acid protein (BmNOS3) that showed 81% sequence identity to *X. tropicalis* NOS3. Interestingly, *B. marinus nos3* mRNA expression was found in blood vessels with and without endothelium suggesting that it is expressed in the non-endothelial component of the vessel wall, and in the brain, kidney, bladder and heart. An affinity-purified BmNOS3 antibody was generated with BmNOS3-immunoreactivity (BmNOS3-IR) observed in the kidney tubules, but not in the endothelium of renal glomeruli and systemic blood vessels. The expression of *nos3* mRNA in the non-endothelial component of the blood vessel wall raises the possibility that NOS3 may be involved in vascular regulation in amphibians, in addition to NOS1 in perivascular, nitrergic nerves.