

## **The Myosin Heavy Chain specific A4.1025 antibody discriminates different cardiac segments in ancient groups of gnathostomes: Morphological and evolutionary implications.**

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The pan-Myosin Heavy Chain (pan-MyHC) marker MF20 have been reported to show similar, homogeneous signal in the myocardial segments of the heart of teleosts and tetrapods. However, in an ongoing study of the myocardial structure of the dogfish (*Scyliorhinus canicula*; Chondrichthyes), we observed differential immunostaining of the cardiac segments using another pan-MyHC, the A4.1025 antibody. In order to investigate the relevance of this finding for better understanding of the morphology and evolution of the vertebrate heart, we performed immunohistochemistry, slot blot and western blot in several species of chondrichthyans, actinopterygians and mammals using the above mentioned antibodies.

In the dogfish heart, A4.1025 and MF20 specifically recognized MyHC isoforms, although with different degree of affinity. MF20 reactivity was homogeneous and high in all the myocardial segments. However, A4.1025 reactivity was heterogeneous. It was high in the sinus venosus (external layer), atrium and atrioventricular region, low in the ventricle and conus arteriosus, and null in the internal layer of the sinus venosus. A heterogeneous pattern of A4.1025 immunoreactivity was also detected in two other elasmobranchs, a holocephalan, a polypteryform and an acipenseriform. In all of these species, MF20 immunoreactivity was homogeneous. In addition, both markers showed a homogeneous immunoreactivity pattern in teleosts and mammals.

Our results indicate that in the hearts of ancient gnathostomes, in all of which a conspicuous conus arteriosus exists, one or more MyHC isoforms with low affinity for A4.1025 show segment-specific distributions. Thus, A4.1025 appears to be an appropriated marker to identify the cardiac segments and their boundaries. We propose that the segment-specific distribution of MyHC isoforms may generate a particular type of myocardial contractility associated with the presence of a conus arteriosus.

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