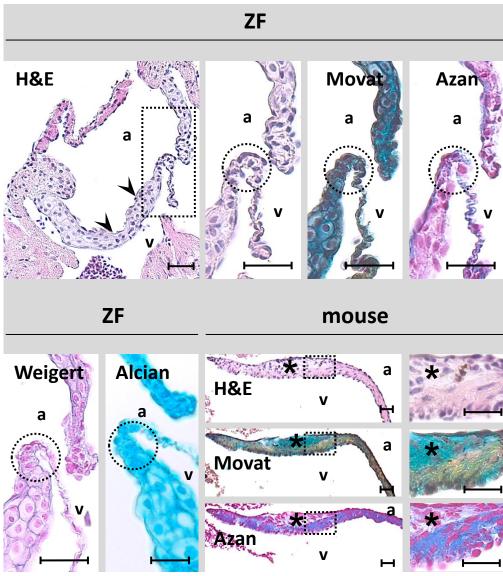
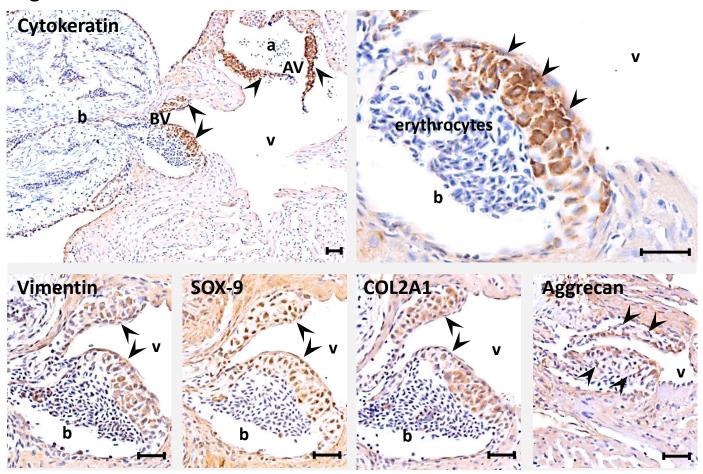


**Figure S1:** Bulbo-ventricular (BV) valves of adult (2.5-year old) ZF. Leaflets are outlined by arrow heads. Parts of the bulbus are marked by a 'b', while the ventricle is marked by a 'v'. Comparable to the findings in atrio-ventricular valves (Figure 1) large polygonal-shaped cells are found as central element in the base and mid-region of BV valves in ZF. A marginal layer (arrows) of collagen and elastic material locates at the ventricular (inflow) aspect of the BV valves. Strong Alcian blue staining demonstrates presence of a cartilage-like matrix. Squares indicate the regions magnified on the further right photographs. Scale bar equals 25  $\mu$ m in each photograph. Russel-Movat's pentachrome stain (Movat), Azan stain, Weigert's resorcin-fuchsin stain and Alcian blue stain.



**Figure S2:** Atrio-ventricular (AV) valves of young adult (1-year old) ZF and mice. Leaflets are outlined by arrow heads. Parts of the atrium are marked by an 'a', while the ventricle is marked by a 'v'. Compared to the large polygonal-shaped cells in the proteoglycan-rich base and mid-region of the AV leaflets (Figure 1) the tip region (circles) characterizes high cellularity of small sized cells. High cellularity is also seen in collagen-free, proteglycan/ glycosaminoglycan-rich myxomatous changes (asterisks) frequently found in the tips region of murine heart valves. Squares indicate the regions magnified on the further right photographs. Scale bar equals 25  $\mu$ m in each photograph. Russel-Movat's pentachrome stain, Azan stain, Weigert's resorcin-fuchsin stain and Alcian blue stain.

Fig. S3



**Figure S3:** Immunohistochemical staining of bulbo-ventricular (BV) valves in young adult (1-year old) zebrafish (ZF). Leaflets are outlined by arrow heads. Parts of the bulbus are marked by a 'b', the atrium is marked by 'a', while the ventricle is marked by a 'v'. Comparable results are obtained in the central cell sheet to those from atrio-ventricular valves (AV, Figure 2) i.e. co-expression of epithelial (cytokeratin) and mesenchymal (vimentin) markers, SOX-9 staining in the nuclei (circles) and aggrecan and type 2a1 collagen based matrix formation. COL2A1 = type 2a1 collagen. Scale bar equals 25  $\mu$ m in each photograph.