# High Resolution In Vivo Cardiac MRI of Zebrafish With An Integrated Coil Flow Cell Design

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**Target Audience** Cardiac Investigators, Coil developers.

**Purpose** The zebrafish is an increasingly popular model organism in cardiovascular research. The model offers several unique advantages over traditional rodent models given the rapid generational cycle, low maintenance costs and regenerative abilities of the species. To date however imaging studies of zebrafish have been limited to early life stages (1), during which time the animal is optically transparent and internal anatomy is accessible to direct observation. Increasing pigmentation of the maturing animal's skin prevents such evaluation of the adult animal, therefore new modalities are required to assess cardiac function. The small size and aquatic habitat of the zebrafish have prevented the widespread deployment of existing non-invasive imaging techniques (such as ultrasound or Magnetic Resonance Imaging (MRI)) to this organism although there have been some previous limited successes (2, 3, 4). Subject immobilisation and biological stability during cardiac studies provide further challenges to developing MRI for zebrafish. We aimed to construct a handing and restraint system to enable the routine imaging of live zebrafish to oversome these challenges.

**Methods** A solenoid micro-imaging coil (insulated copper wire, 3 loops, coil length = 4.00mm inner diameter = 7.46mm) was embedded in a shaped chamber within a machined acrylic block (Fig. 1). Anesthetised zebrafish (MS222 125mg/l) are placed within this channel, positioned so that the heart of the specimen is within the coil. Shaped inserts and small pieces of medical swab are positioned around the fish to ensure immobilisation during scans. This chamber is then sealed with a Parafilm gasket and acrylic lid to make it water tight. Freshwater (temp. 16-20°C) containing a maintenance level dosage of MS222 (100mg/l) is then pumped through this chamber (2ml/min), providing both anaesthesia during the scan and a continuous supply of air oxygenated water. Scans were performed on a 7T Bruker Biospec MRI scanner equipped with a microimaging gradient insert (1000mT/m). The water supply for the flow cell is monitored for temperature and oxygen values using an optical inline monitoring system (FireSting O2, Pyroscience). IntraGate images TR=20ms, TE=2.8ms, 300 reps, FOV=1.00cm, Navigator slice FA=10° and 0.5mm thick. After the procedure (up to 45mins long) all fish are returned to a tank of fresh tank water where they recover successfully.

**Results** Images can be readily produced from the equipment in time scales equivalent to that of existing rodent scans (Fig. 2). All fish thus far scanned (n=13) recovered from the procedure rapidly after removal from the flow cell and showed no observable negative effects from scans.

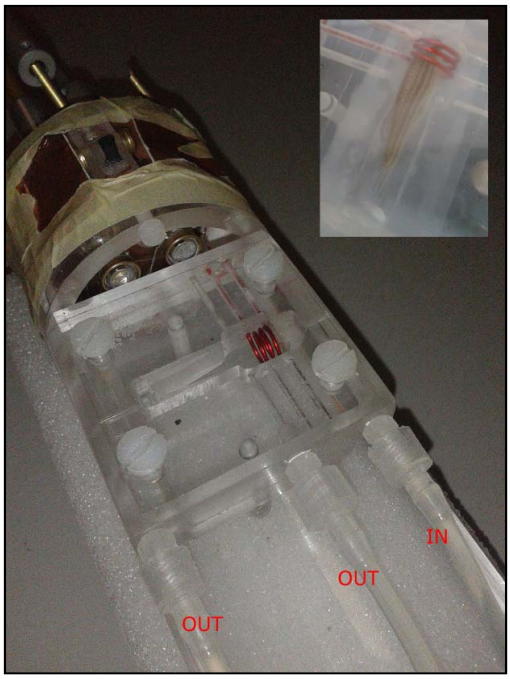


**Fig. 1** Integrated MRI flow cell and coil (red) set up. Water channels marked IN and OUT. Zebrafish in flow cell (insert).

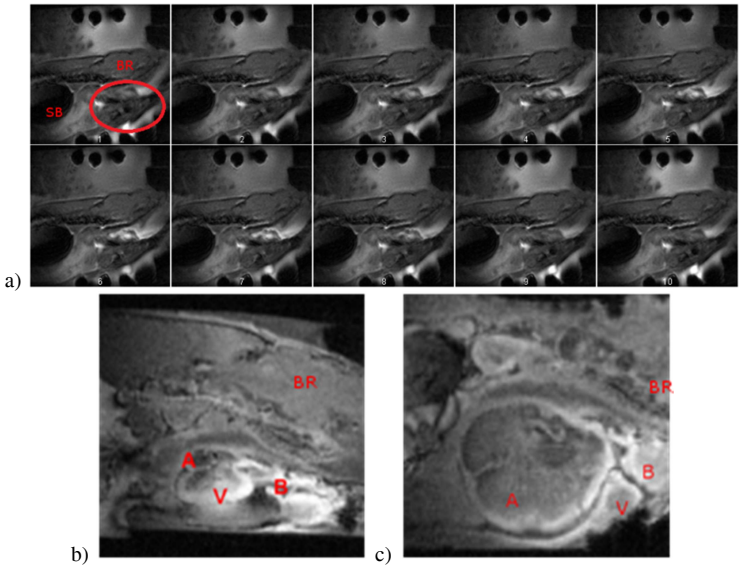


**Fig. 2 a)** *in vivo* IntraGate FLASH cardiac (circled) sag. images at different time points (50µm in plane, 200µm thick), **b) + c)** GE3D *ex vivo* cardiac sag. Images (50µm isotropic) of fixed wild type (b) and mutant (c) zebrafish. V = ventricle, A = atrium, B= bulbous arteriosus, BR = brain, SB = swim bladder. Posterior of fish to left of all images.

**Discussion** We have demonstrated a successful design for an integrated coil and handling system to overcome the unique challenges involved with scanning live zebrafish. Comprising of a solenoid-based MRI coil, embedded in a water filled flow chamber within which a zebrafish can be comfortably maintained and immobilised for the duration of an MRI experiment.

**Conclusion** Imaging the zebrafish *in vivo* with MRI is a viable prospect when equipped with the proper equipment. Handling, restraint and immobilisation necessary for this does not adversely affect the fish being scanned.

**References** 1. Denvir et al., 2008 BMC Biot. 8:21 2. Merrifield et. al., 2013, Proc. Intl. Soc. Mag. Reson. Med. 21:1387, 3 van Herck et. al., 2014, Proc. Intl. Soc. Mag. Reson. Med. 22:3890, 4. Manivannan et. al., 2014, Proc. Intl. Soc. Mag. Reson. Med. 22:2394.