Title: Blood Collection in Fish

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1.0 OBJECTIVE
The objective of this Standard Operating Procedure is to describe a method of blood collection in fish.

2.0 HEALTH AND SAFETY
All personnel must be enrolled in the Clemson University Medical Surveillance Program. Used needles, syringes, and/or tubes must placed into a sharps container for disposal.

3.0 PERSONNEL/TRAINING/RESPONSIBILITIES
Any employee familiar with the equipment, techniques and trained in this and referenced SOPs may perform this procedure. Prior to being assigned full responsibility for performing this procedure, personnel must have demonstrated proficiency in the use of the technique in a closely supervised environment.

4.0 GUIDELINES
Up to 10% of the blood volume of a fish can be taken per week. Blood collection should not exceed 20% of the blood volume per month. Anything above 20% of the blood volume should not be collected except when being conducted as a terminal blood collection under anesthesia followed by euthanasia. Fish are estimated to have a total blood volume of conservatively 2% of the body weight. Therefore the blood collected in a non-terminal collection should not exceed 2ml per kilogram.

5.0 PROCEDURE
1. Fish to be bled are anesthetized in water containing buffered (pH 7.0-7.5) MS-222 at about 100 mg/liter.
2. The area where the needle is to enter is swabbed with Betadine or equivalent antiseptic, prior to blood collection.
3. For serial sampling, blood should be collected from the hemal arch with a vacutainer tube, or appropriately sized needle and syringe.
4. If the collection is to be terminal, a cardiac puncture is appropriate for some larger species, and collection via a tail cut for smaller species.
5. Immediately after the blood is collected the fish is placed into a buffered (pH 7.0-7.5) MS-222 solution of 1 to 1.5 gr/liter of water for fifteen minutes to ensure death. As an alternative to emersion, a euthanasia strength solution may be applied directly onto the gill plate by dropper or spray.

6.0 QUALITY CONTROL CHECKS AND ACCEPTANCE CRITERIA
All procedures are subject to review by the Quality Assurance Unit.