2007 GBT Monitoring Program Protocol for Juvenile Salmonids

Fish will be examined externally for signs of gas bubble trauma (GBT). The unpaired fins, and eyes will be examined for the presence of bubbles and the area covered with bubbles will be quantified. Monitoring of migrating juvenile salmonids will be conducted at Lower Granite, Little Goose, Lower Monumental, Rock Island, McNary, and Bonneville dams. The goal of the juvenile salmonid examinations is to determine the relative extent to which the migrating juvenile salmonids passing the dam or sampling location have been exposed to harmful levels of total dissolved gas based upon the prevalence and severity of GBT induced bubbles on the fish. The data will be reported to the fisheries management entities, the water quality agencies of Washington and Oregon, and will be made available to other interested parties through Fish Passage Center weekly reports and daily postings to the FPC web site during the season.

Method of fish examination for GBT

Fish will be examined using a variable magnification (6X to 40X) dissecting scope. Unpaired fins, and eyes will be examined for the presence of bubbles. Fish to be examined will be netted off the separator (or removed from bypass or other sampling apparatus at Rock Island and Bonneville dams) and anesthetized. A specially designed tray, that allows fish to be continually anesthetized during the GBT examination, will be used to hold the fish. Fins on the left side of the fish will be examined for signs of GBT and then both eyes will be examined for signs of GBT. The eye with the highest % of bubbles will be used for ranking, using the same ranking as for fins.

The fish exam will begin with the unpaired fins and then the eyes will be examined and data recorded based on the percent area of the fin or eye covered with bubbles. A minimum magnification for these examinations will be 10X. The area covered will be estimated using the examiners best judgement. A visual technique for estimating the area of the fin covered by bubbles is illustrated in Figure 1.

A rank will be assigned based upon the percent area of the fin or eye covered with bubbles. A rank 0 is assigned if no bubbles occur; rank 1 will be assigned if 1 to 5 percent of the fin or eye is covered with bubbles; rank 2 is assigned for 6 to 25 percent area covered; rank 3 for 26 to 50 percent area covered; and rank 4 for greater than 50 percent area covered. The rank reported for the eyes will be the highest rank for either eye. When the percent area covered is near the boundary for two ranks (e.g. at or near 25 percent) then the higher rank will be assigned. A summary of ranks is listed in Table 1 in the data entry section below.

Other information will be collected and recorded for each fish examined: species; time of examination; fork length (mm); origin (hatchery, wild, or unknown), and comments regarding tags and fish condition as deemed relevant by the examiner. A sample data sheet is attached.
Figure 1. Conceptual drawing depicting the estimation of area of a fin occluded. The fin on the left is what might actually be viewed on a fish, and the fin on the right shows the fin are divided in areas approximating 25% of fin area and occlusion grouped to estimate actual percent area covered.

Figure 2. Conceptual drawing depicting the estimation of area of an eye occluded.
**Sample Size**

The number of juvenile salmonids to be examined at each site each day will be adequate to detect signs of GBT. The target sample of 100 juvenile chinook and/or steelhead is a daily minimum and is based on the availability of fish at each monitoring site. This number is sufficient to detect signs of GBT that would indicate potential mortality in the population. Based on calculations developed by USGS - Biological Resources Division, a sample size of 100 fish should be able to detect within ±6% the incidence of fish in a population showing signs of GBT based on a population where 10% of the fish had signs. We consider this level of precision and subsequently the sample size, optimal for the monitoring program.

**Method of Collection**

Fish to be examined for GBT will be collected at the separator at transportation sites and by the standard collection methods at Rock Island and Bonneville dams. Fish will be netted one at a time and placed in a dark bucket (not white) filled with anesthetic water. At transportation sites, no more than 10 fish per examiner will be netted off the separator at a time, so that all fish can be examined within 15 minutes of netting. Fish netted off the separator will be placed in a bucket containing a solution of 30mg/l MS-222 and if necessary 30mg/l sodium bicarbonate buffer (see method of anesthetizing below).

**Handling PIT-tagged fish at Transportation Sites**

At Lower Granite, Little Goose, Lower Monumental, McNary and Bonneville dams, fish that are netted off the separator for GBT exams will be scanned for the presence of PIT-tags. As soon as the fish can be handled (after anesthetization) it will be scanned for a PIT-tag. If a tag is encountered, a tag file will be created for that date and the information sent to PTAGIS. The PIT-tagged fish will be placed in a recovery bucket and returned to the separator as soon as possible.

**Method of Anesthetizing fish for GBT examination**

Each site will have five 5-gal plastic buckets. Three buckets will be used for holding fish and two will be used for the gill irrigation system while fish are being held in the examination tray. Prior to examination fish will be held in 30mg/l MS-222 anesthetic solution (buffered if necessary depending upon the pH change in the water when anesthetic is added). Once all fish are anesthetized they will be moved to an 80mg/l solution of MS-222 just prior to examination to fully anesthetize them. During the examination a solution of 30mg/l MS-222 will be washed over the gills of the fish to keep fish under anesthetic for the entire examination. The fish will be held in an examination tray during the examination. The tray will be modified to hold a siphon tube that will carry anesthetic water over the animal's gills. The anesthetic water will drain out of the tray into another bucket via a drain tube. After the examination fish will be placed in a recovery bucket of fresh water containing an air stone. The recovery bucket will have a lid and the air stone will vigorously pump air into the bucket.

**Handling of Fish After Examination at Transportation Sites**

At transportation sites, after the fish have been examined for GBT, the fish will be placed in the location where other SMP fish are placed after examination: fish should be placed in a recovery tank or a holding tank. Ultimately these fish will be transported, as are fish that are
examined by SMP for condition information. These GBT examined fish will be counted as fish sampled at 100% sample rate. Daily totals of the number sampled by species and origin will be reported to the FPC so that this information can be included in their daily sampling reports.

Data Recording and Data Entry Procedures

As each fish is examined, data will be recorded on a data sheet. The following information will be recorded as part of a record for each fish: the date of examinations; the time each examination, the site examinations were done, species, origin, fork length (mm), the rank of GBT in each unpaired fin, rank of GBT in the eyes, information on tags, and fish handled but not examined for GBT. See data sheet.

Data will be entered to the GBT data entry program and sent to FPC via e-mail or other electronic transmission technique. The following section describes the format of data that will be entered to the spreadsheets. Data will be entered into a Data Entry Program provided by FPC to the sites. Data entry must strictly follow the guidelines below so that data can be transferred to the database properly.

Definition and format of data entries:
Format codes: A = character strings; Iw = integers, where w=field width; Fw.d = Fixed, real numbers, where w=field width and d=# of decimal places.

1. Site: An acronym for the sample site name. Format = A3
   Rock Island Dam = RIS   Lower Granite Dam = LGR   Little Goose Dam = LGS
   McNary Dam = MCN   Lower Monumental = LMN   Ice Harbor = IHR

2. Date: Start date of GBT sampling entered as month-day-year (MM/DD/YY).

3. Examiner: Initials of person(s) doing actual fish exams for GBT. Format = A2

4. Time: Hour-minute (HHMM) military time, to the nearest minute for each fish examination. Format = Iw (XXXX), e.g. (5:15 for 5:15 am or 19:25 for 7:25 pm). Midnight is 2400 hours. One minute after midnight is 00:01 hours.

5. Num: Special field – Enter this number to coincide with “Num” field of the GBT data entry program GBTDEP. This can only be entered while entering data into the GBTDEP.

6. Species: (Species)
   CH = Chinook
   ST = Steelhead

7. Age: For Chinook, 1 or 0, otherwise leave blank.

8. Race: For Chinook, SP (spring) or FA (fall) for Elastomer tagged fish only.
8. **FL: (Fork Length)** Enter fork length to the nearest millimeter

9-12. **CA, AN, DO, EY: GBT Rank in Unpaired Fins and Eyes**
See section above for a description of how to do examination. Enter the number corresponding to the percent area of the fin or eye that is covered by bubbles. Rank is entered into the data sheet as 0, 1, 2, 3, or 4.

<table>
<thead>
<tr>
<th>Rank</th>
<th>Percent area covered With bubbles</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>1</td>
<td>1 to 5%</td>
</tr>
<tr>
<td>2</td>
<td>6 to 25%</td>
</tr>
<tr>
<td>3</td>
<td>26 to 50%</td>
</tr>
<tr>
<td>4</td>
<td>Greater than 50%</td>
</tr>
</tbody>
</table>

13. **Clip Code** To record clipped fins, and clipped fin combinations on marked fish.
NC = NO FIN CLIPS  AD = Adipose  RV = Right ventral only
AR = Adipose/Right ventral  LV = Left ventral only  AL = Adipose/Left ventral
NW = NO FIN CLIPS with Coded Wire Tag

14. **Mark Type:** Enter the type of external mark observed on a fish or group of fish with identical mark types/attributes. Current valid mark types are:

EL: Elastomer Tags
FB: freeze brands
FL: Floy tags
VI: Visual Implant tags

**NOTE:** Coded Wire tags and PIT tags are internal marks that are not included in this entry screen. Recaptured PIT tagged fish are reported directly to PTAGIS. They are counted as not examined and put back in the separator.

15. **Mark Location:** (i.e. Brand Location, Tag Location) Enter location code for freeze brands and elastomers

**Brand Location**
LA = Left anterior  RA = Right anterior  LD = Left Dorsal
RD = Right dorsal

**Elastomer Location**
LE = Left Eye  RE = Right Eye
16. **Color Code:** For ELASTOMER TAGS, the appropriate color code should be entered, (RE=Red, GR=Green, and BL=Blue) (Leave blank for numbered VI tags). For FREEZE BRANDS enter the brand orientation here, e.g. 12 o’clock = 1, 3 o’clock = 2, 6 o’clock = 3, 9 o’clock = 4.

17. **Exam Y/N?:** Enter N if fish was/were not examined for GBT. Leave blank if fish was examined.

18. **Num Fish:** Leave blank if fish was examined for GBT. Enter number of fish if previous field = N.

**Data Transfer Procedures**

The data will be saved as FoxPro .dbf and .cdx files, and both files will be transferred to FPC via e-mail or other available electronic medium. Files will be named according to the site and the date on which the data was collected. For example if data were collected on April 10, at Bonneville dam, the data file would be named "BO1410.dbf" and “BO1410.cdx”. The first three characters of the file name are the site designator, the next character is the month, and the next two characters are the day data was collected. File names must be 6 characters long.

**Data Reporting Procedures**

Once the faxed data is received at FPC it will be used to check the electronically transferred spreadsheet file. The final report will be generated based on the data management system that the FPC has developed.

**Quality Assurance and Quality Control**

In order to assure quality control/quality assurance several checks will be included as part of the monitoring program. At the first step in the process, fish examinations, there will be visits to the monitoring sites to assess the accuracy of the results of examinations and data recording. A supervisory fish biologist from Fish Passage Center or USGS - Biological Resources Division will visit the sites to perform QA/QC checks. The supervisor, who will also compare those examinations with the results from the on-site biotechnician's exams, will examine twenty fish. The results of these visits will be compiled in a report and be available for interested parties.