Fish Flavour Impairment Study

May, 1996

Prepared for:

Suncor Inc.

Prepared by:

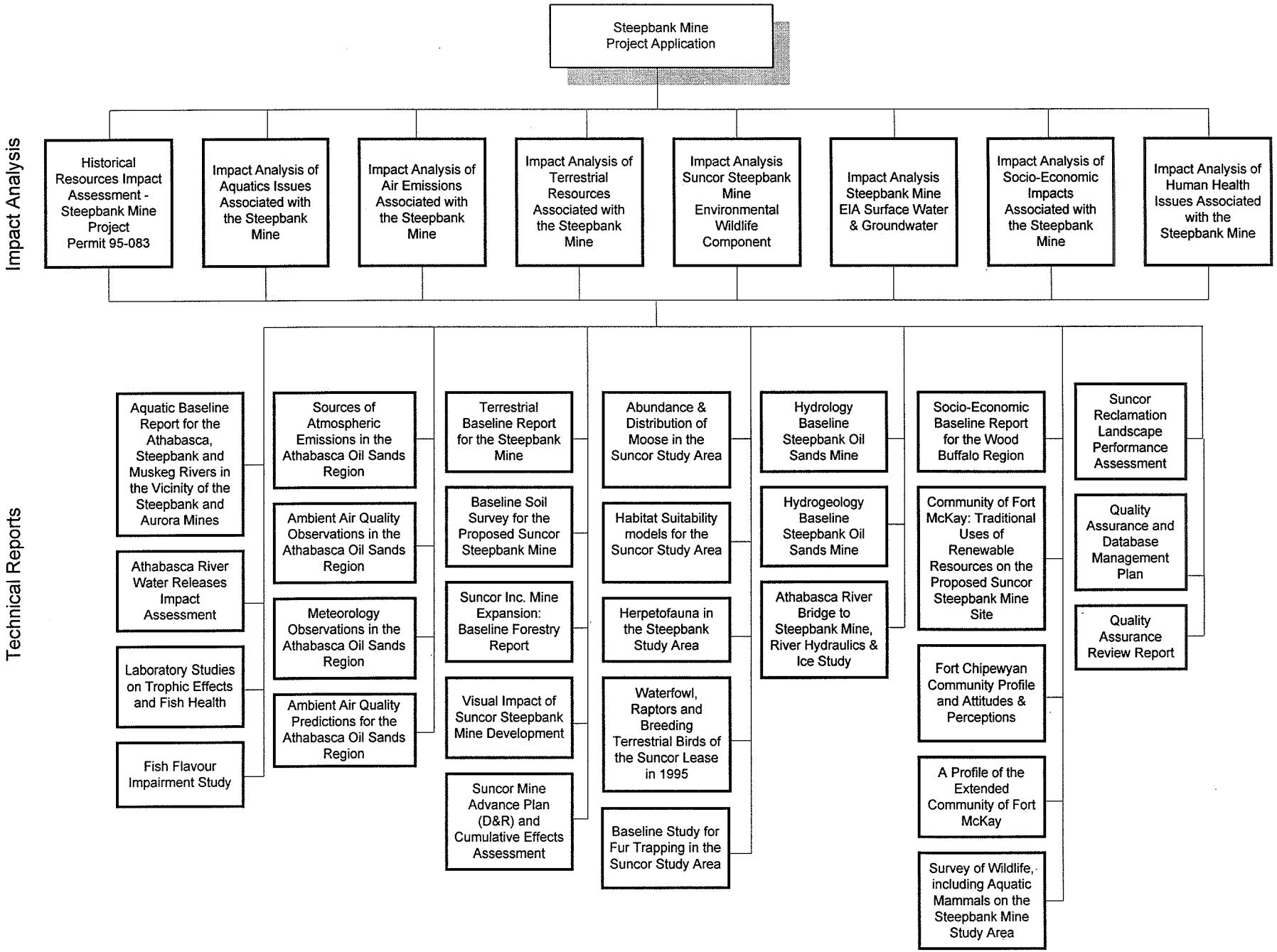
Golder Associates

952-2307

This report is one of a series of reports prepared for Suncor Inc. Oil Sands Group for the Environmental Impact Assessment for the development and operation of the Steepbank Mine, north of Fort McMurray, Alberta. These reports provided information and analysis in support of Suncor's application to the Alberta Energy Utilities Board and Alberta Environmental Protection to develop and operate the Steepbank Mine, and associated reclamation of the current mine (Lease 86/17) with Consolidated Tailings technology.

For further information, please contact:

Manager, Regulatory Affairs
Suncor Oil Sands Group
P.O. Box 4001
Fort McMurray, AB
T9H 3E3



Reports Prepared for the Steepbank Mine Environmental Assessment

r:\1995\2307\repprep5.040\rpt-sch.vsd

1.0 INTRODUCTION

Suncor Inc., Oil Sands Group (Suncor) is planning to expand their oil sands facilities to the west side of the Athabasca River, north of Fort McMurray, Alberta. As part of the environmental impact Assessment (EIA) for this expansion a combined laboratory and field investigation was conducted to evaluate the potential for fish flavour impairment (tainting) from various types of water releases associated with Suncor's present and future operations.

Rainbow trout were exposed to different water regimes (0.5% Tar Island Dyke water, 0.5% Refinery Effluent Water, Athabasca River Water) for 10 days in the laboratory plus caged fish were also held for 10 days in the Athabasca River upstream of industrial oil sands operations (i.e., Suncor and Syncrude Canada Ltd.). Fillets from these fish were then submitted to a taste-test panel to determine the relationship between exposure of the fish to different water regimes and the flavour of the fillets.

2.0 METHOD

The general design and operation of the exposure system is based on the method Standard Practice for Evaluating an Effluent for Flavour Impairment to Fish Flesh, 1989 (Annual Book of ASTM Standards, Vol. 11.04, D 3696-89). A copy of this standard practice is attached (Appendix I).

The fish were exposed in 600 L tanks containing roughly 400 L of test solution. The inflow was directed along the inside of the top of the tank to assist circulation of the water in a clockwise fashion. A submersible pump located on the tank bottom near the tank wall was also used to generate a circular flow. The outlet on the tank bottom was connected to an external stand pipe. The height of the stand pipe was used to adjust the tank volume. A schematic is attached along with photographs of the experimental setup (Figures 1, 2, 3, and 4).

Each tank could be fitted with a Ranco 1 hp chiller unit for additional temperature control. A chiller was only required for the Athabasca River water sample. The other two samples were diluted with laboratory water that was at the test temperature.

The Tar Island Dyke water and refinery effluent were diluted with laboratory dilution water before entering the tank. Sample flows were regulated with constant flow pumps. Flow rates were measured with a calibrated flow meter and adjusted daily if required. Valves fitted to the dilution water and sample lines offered fine flow adjustments (Figure 1).

The fish were obtained from a supplier certified as disease free (Bob Allen's Trout Farm, Calgary, AB) and weighed between 200 to 300 g. Roughly 200 fish were transported to the laboratory on October 4, 1995 and allowed to acclimate for ten days prior to the exposures. A number of fish were taken from the laboratory to Fort McMurray on 4 October, 1995 for the field exposures.

At test initiation, 30 to 40 fish were placed into each of three tanks. The treatments were, Athabasca River Water, 0.5% Tar Island Dyke water, and 0.5% refinery effluent. The Tar Island Dyke and refinery effluent samples were collected on October 27, 1995 and received at the test facility on the same day. The water samples for the Athabasca River exposure were collected on 29 September, 27 October, and 3 November, 1995. Flows were set at 1.4 L/min or roughly 2 m^3 per day (three to four tank volumes per day). Over 20 m^3 of Athabasca River water was required for the ten day exposure. The water was collected and shipped in 6 m^3 plastic water tanks on a flatbed trailer. The samples were received on the day of collection. Sample handling and transport containers are illustrated in Figure 5.

Conductance, dissolved oxygen, temperature, pH and ammonium were measured daily. Temperatures were regulated between 14 to 16 °C. The fish were fed a ration equal to roughly 3 to 5% body weight per day throughout the test. The tanks were also cleaned daily.

At the end of the exposure period, the fish were removed and examined individually. Lengths, weights, sex, external and internal condition were recorded and scale samples removed. The laboratory control fish were sacrificed prior to test initiation.

3.0 RESULTS

3.1 Laboratory Exposures

The test on the Athabasca River water was initiated on October 26, 1995 and terminated on November 5, 1995. The refinery effluent and Tar Island Dyke water exposures were started on October 28, 1995 and terminated on November 7, 1995.

General water quality conditions during the exposure period are summarized in Tables 1a, 1b, and 1c. No mortality or signs of stress and abnormal behaviour were noted in any of the tanks over the ten day exposure. There were also no discernible differences in the appearances of the fish amongst all treatments (both internal and external examinations). The data collected on the fish are presented in Appendix II. The carcasses were frozen, and then packed in dry ice and sent to Diversified Laboratories Ltd. on November 9, 1995 and received on November 10, 1995 (See Appendix III for shipping documentation).

A sample of laboratory dilution water was submitted for analyses of selected water quality parameters. These data are summarized in Table 2.

3.2 Flavour Impairment Assessment

The fish tainting evaluation was conducted by Diversified Laboratories Ltd. (Appendix IV). Samples of fish were submitted to a tasting panel to determine if there was a difference in the taste and if there was a taste preference. A double triangle difference test was used to determine differences in taste. This test was two-part and only samples that were correctly identified in both tests were used to assess taste differences. The results of the difference test are presented in Appendix IV. Fish exposed to 0.5% Tar Island Dyke water and 0.5% refinery effluent water were found to taste different than fish exposed to Athabasca River water either in the field or in the lab. In addition, fish exposed to Athabasca River water in the lab tasted different that control fish.

The fish samples were also ranked for overall preference in both Test 1 and Test 2. Only samples from fish exposed to 0.5% refinery effluent water were rejected. Hence, tainting was evident in the trout exposed to 0.5% wastewater (diluted with laboratory water), but not in dyke drainage water from Tar Island Dyke or Athabasca River water (Appendix IV).

4.0 REFERENCES

Standard Practice for Evaluating an Effluent for Flavor Impairment to Fish Flesh, 1989. Annual Book of ASTM Standards, Vol. 11.04, D 3696-89.

TABLES

Table 1a - Water Quality Monitoring Data, Athabasca River

DAY	pH	Conductance (uS/cm)	Dissoved Oxygen (mg/L)	Temperature (^{0}C)	Ammonium (mg-n/L)
0	7.4	399	5.0	15	0.5
1	8.0	301	7.7	13	1.0
2	7.9	294	7.3	14	1.3
3	7.9	281	7.9	14	1.5
4	7.6	278	6.8	14	1.2
5	7.9	276	8.1	13	1.4
6	7.9	275	8.2	14	1.0
7	7.9	278	8.3	13	1.0
8	7.9	275	8.1	14	1.1
9	7.9	283	8.0	13	1.1
10	7.8	292	7.4	13	1.4

Table 1b - Water Quality Monitoring Data, Tar Island Dyke

	pH	Conductance (uS/cm)	Dissoved Oxygen (mg/L)	Temperature (^{0}C)	Ammonium (mg-n/L)
0	7.9	392	7.6	14	1.0
1	7.9	390	7.2	15	1.4
2	7.8	392	7.7	15	1.2
3	7.9	411	7.8	14	2.0
4	7.9	406	7.7	14	1.0
5	7.9	406	7.7	14	0.9
6	7.9	405	7.8	13	0.9
7	7.9	401	7.7	13	0.9
8	7.9	400	7.1	14	1.2
9	7.8	414	6.9	14	1.7
10	7.9	403	7.7	13	0.7

Table 1c - Water Quality Monitoring Data, Refinery Effluent

	pH	Conductance (uS/cm)	Dissoved Oxygen (mg/L)	Temperature (^{0}C)	Ammonium (mg-n/L)
0	7.9	394	7.6	14	1.0
1	7.9	405	7.5	15	1.4
2	7.9	398	7.8	15	1.2
3	7.9	410	7.8	14	1.5
4	7.9	405	7.8	14	1.0
5	7.9	406	7.8	14	0.8
6	7.9	403	7.9	13	1.0
7	7.9	402	8	13	0.9
8	7.9	409	7.4	14	1.2
9	7.8	412	7.3	14	1.5
10	8.0	407	8.3	13	0.6

Table 2 - Chemical Analyses of Laboratory Dilution Water

PARAMETER	VALUE
Total Organic Carbon (mg/L)	2
Dissolved Oxygen (mg/L)	Saturated
Biological Oxygen Demand (BOD; mg/L)	<2
Chemical Oxygen Demand (COD; mg/L)	<1
pH	7.5
Hardness (as $CaCO_3$; mg/L)	199
Total Dissolved Solids (mg/L)	216
Conductance (uS/cm)	371

FIGURES

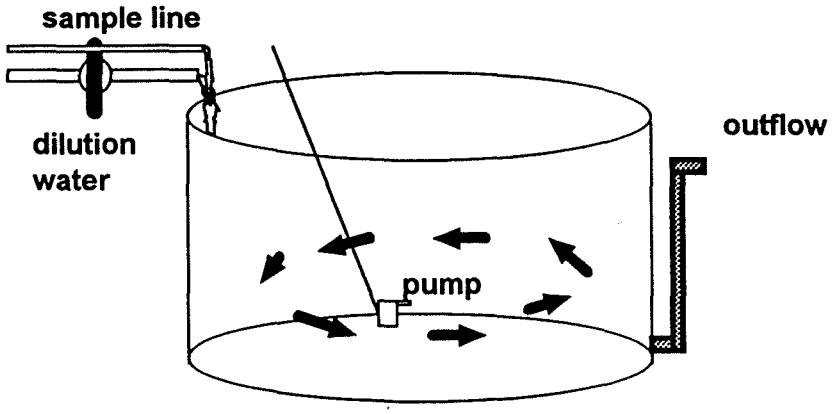


Figure 1. Schematic of Exposure Tank

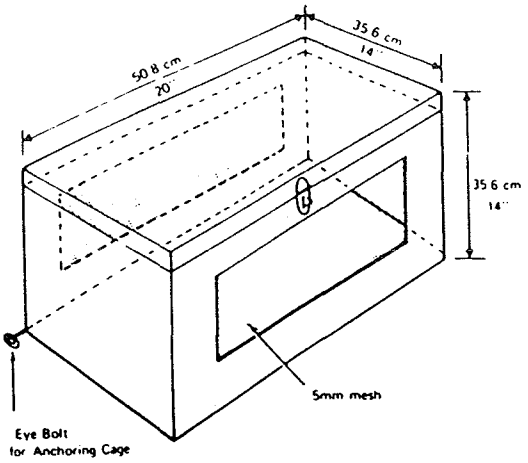


FIG. 1 Fish Cage

be used. Reference (1, 2)[5] and Practice E 729 describe suitable systems that are or can be modified for effluents.

6.2.2 *Compressed Air* (oil free).

6.2.3 *Aquaria*—Aquaria should be large enough to provide an adequate volume of water and sufficient room for the fish, such as 35 by 35 by 50 cm or larger.

NOTE 1: **Caution**—Testing apparatus that comes in contact with the water in which the fish are exposed should not contain any substance that could be leached out. Glass, No. 316 stainless steel, and perfluorocarbon plastics should be used whenever possible. Other plastics that contain no leachable plasticizers may also be used. However, substances that are known to absorb organics should be avoided. Rubber, copper, brass, zinc, and lead should not come in contact with the water or effluents to which the fish are exposed.

6.3 *Equipment Required for Taste Evaluation*:

6.3.1 *Oven*, capable of 190 ± 5°C (electric or gas).

6.3.2 *Plates, glasses*, and *disposal cups*.

6.3.3 *Metal Foil*.

6.3.4 *Cookie Sheets*.

7. Dilution Water

7.1 The minimal water quality criteria for any flavor impairment testing are that the fish will survive in it and remain healthy for the acclimation period and during the tainting tests. The more general acceptable criteria are that first instar (newly hatched) daphnids will survive in the dilution water for 48 h without food (see Guide E 729). The water must be free of taint producing materials. Also sample water for chemical analysis in accordance with Specification D 1192 and Practices D 3370. Some suggested chemical analyses are designations Methods D 2579, Method D 1252, Method D 3250, Method D 1293, Methods D 1126, Methods D 1125, and Methods D 1888.

7.2 *Fresh or Frozen Unsweetened Lemon Juice*, 1+32 dilution or weaker.

8. Safety Hazards

8.1 Do not wash fish that are being cleaned in the field with effluent or the dilution water (river, lake, etc.). Use paper towels to wipe the fish clean. Do not taste fish that are dead in the exposure tank or show any signs of toxic effects, as they may be toxic to the taster or possible tissue deterioration may influence the test results.

8.2 Minimize personal contact with the effluent or dilutions of the effluent as it is always possible that some hazardous material, bacterial, or viral pathogen may be present. Thoroughly clean hands, clothing, and equipment after contact.

8.3 Follow local water safety laws and practices in field studies. Check with local enforcement agencies, since these laws vary from one area to another. When wading in water, wear boots or chest high waders. Wear a life vest or preserver when wading in deep water or in a boat.

8.4 A current food handler's certificate may be required by local law for the cleaning, handling, and preparation of fish samples.

9. Test Specimen (Fish)

9.1 Any edible fish available in sufficient numbers is an acceptable test species. Cultured fish, such as rainbow trout, *Salmo gairdneri* Richardson, and bluegill, *Lepomis macrochirus* Rafinesque, are two freshwater species that have commonly been used. Fish should be large enough to provide a fair-sized fillet. Trout 200 to 300 mm or bluegill 150 to 200 mm are of sufficient size.

9.2 Prior to testing, hold fish in a flow-through water system of similar water quality to that of the experimental exposure for at least 10 days. Maintain a sufficient flow in and out of the holding tankage to provide dissolved oxygen of at least 60 % saturation and to flush out fish excretory products. Holding temperature should be ± 2°C of the exposure temperature.

9.3 Feed the stock fish and those being exposed, if practical, but it *must* be recognized that materials may be bioaccumulated from the food and also cause flavor impairment.

9.4 Conducting fish taste tainting studies may require a permit of some type; therefore, notify the local conservation department or enforcement agency.

10. Procedure

10.1 *Field Studies—Using Caged Fish:*

10.1.1 A common field technique of evaluating fish flesh tainting can be accomplished by placing fish in cages at various locations relative to the outfall. If trout are exposed in small wire mesh cages, include minnows as food. In a river or stream, place cages upstream of the effluent, at the outfall, and at sites downstream. The number of cages placed above and below an effluent and the distances of sites from the river effluent vary depending upon the hydrology of the river. In a lake or large river, the wind velocity and direction and other factors may also affect the effluent concentration to which the fish are exposed. Placement of the cages as to area of study and depth is at the discretion of the investigator. An exposure of 10 days is accepted as adequate.

10.1.2 Place at least one cage of fish as a control in the water upstream from the outfall or away from influence of the effluent.

10.1.3 The number of fish per cage is dependent upon the size of the test species and the number of taste panel

[5] The boldface numbers in parentheses refer to the references appended to this standard.

members. Plan on at least a 10-g fish portion per taster per exposure concentration.

10.2 *Laboratory Studies:*

10.2.1 Pump a representative sample (Practices D 3370) of effluent from the discharge or storage containers through a flow-through system capable of providing a series of effluent dilutions. Include a dilution water control in the test series. Place fish in each dilution and control. See 10.1.3 for suggested number of fish per concentration. Feed the fish once a day throughout the exposure.

10.2.2 Allow sufficient flow through the test aquaria to maintain a dissolved oxygen concentration of at least 60 % saturation. Do not aerate aquaria because the flavor test material may be volatilized from the test water.

10.2.3 Maintain the test temperature of the aquaria at ± 2°C of the average receiving water temperature outside the effluent mixing zone. If a test temperature other than that of the receiving water is used, report the reason and temperature.

10.3 The uptake of materials by fish from water varies from one material to another. Because of this variation, there is not one exposure period that will cover all situations. However, it is recommended the exposure period be 10 days.

10.4 After exposure, remove, kill, fillet, wrap, and refrigerate the fish. If the flavor impairment evaluation is performed within 2 days (48 h), it is desirable to keep the fish refrigerated (or iced) rather than freezing. Freezing is believed by some to alter the texture and taste of the fillet. However, if the flavor evaluation is delayed for more than 2 days, freezing the fish samples is the best method of preservation.

NOTE 2—Take special care to avoid contamination of the fish fillet with oil, gasoline, detergent, etc., because these materials impart flavor.

10.5 On the day of the flavor evaluation, cut fish fillets (if frozen, thaw first) into several portions (at least 10 g) and double wrap in metal foil with the same side out (shiny or dull). This may affect the cooking time. Randomly code each packet of fish to identify each individual fish, area of fish from which the fillet was taken, and the dilution or cage to which the fish was exposed. Provide enough packets of control fish for known and unknown controls, plus a few extra.

10.6 Place the encoded foil packets on a flat cookie sheet and bake at 190°C for 20 to 30 min. Small portions of fish may require less cooking time; therefore, include a few extra packets of control fish to pull at earlier time intervals to check if the fish are properly baked. Overcooking dries out the flesh, seriously damaging the taste quality.

10.6.1 An alternative method for cooking the portions is to wrap each portion in a suitable polyethylene or poly(vinyl chloride) film or bag and then cook in a microwave oven for 5 min. Since various microwave ovens may vary in cooking efficiency, several extra portions should be available to check cooking times.

10.7 *Flavor Impairment Panel:*

10.7.1 A flavor panel can be selected from associates at work, a university, or another testing group to taste the fish fillets. The panel may be comprised of experienced or inexperienced fish tasters, but not a mixture of both. Inexperienced tasters would more likely represent the typical consumer. The smallest detectable concentration will probably vary depending upon whether experienced or inexperienced tasters are used. The normal flavor test panel should have at least 10 members. Flavor panels with fewer than 10 members are likely to produce results with no significant difference between the exposed fish and hidden control fish. Members of the flavor panel should be free from head colds or chest colds, and be free from allergic reactions to hayfever, etc., in order to perform satisfactorily as members of the flavor panel. Any physical disturbance that could affect the flavor panel at the time of taste testing should be cause for preventing a member from active participation at the time of testing.

10.7.2 After the flavor panel members or consulting groups have been selected, inform them of the basic nature and purpose of the test. It may be advisable to initiate an "Informed Consent Agreement" between the flavor panel members and the sponsoring organization.

10.7.3 Record the following information on the rating sheet (see Fig. X1.1 of the Appendix):

1	Name of tester
2	Date
3	Species of fish tested
4	Sample number of code
5	Rating
6	Area for comments

10.7.4 After baking, rate the coded fillet samples by comparing each sample with the known control:

0	Same as or better than known control
1	Slight flavor impairment
2	Moderate flavor impairment
3	Strong flavor impairment
4	Extreme flavor impairment

Before any portion is tasted and between fish samples, rinse the mouth with dilute unsweetened lemon juice (1+32). This helps to prevent carry over of flavor from one sample to another.

10.7.5 Prepare the panel to start as soon as the fish are baked. The taste evaluation is best accomplished with as little distraction as possible. Isolate panel members whenever possible while tasting. Prior to the evaluation, review the tasting and rating procedure with the panel.

10.7.6 Always identify at least one control fillet.

10.7.7 Before tasting begins, note the odor of the fish. If any odor is detected, taste samples in an order of increasing odor.

10.7.8 Taste the known control first. Chew and spit out the cooked fish. Rate each fish before proceeding to the next sample. The panel member may refer back to the control as necessary.

10.7.9 For more detailed information on the principle of sensory evaluation and testing methods, consult two ASTM manuals (3) (4).

11. Calculation or Interpretation of Results

11.1 Tabulate the results of the flavor evaluation panel, similar to that shown in the appendix. Once the data are tabulated, it can be easily seen if flavor impairment is detected. If the flavor impairment data are treated statistically, the method of analysis used should reflect the fact that data are often very consistent at high concentrations and for the hidden control, and less consistent in between (nonnormal, unequal variance). Another restriction in

choosing a statistical procedure is that the ratings from each concentration are not independent because the same tasters evaluate each concentration. With these restrictions, a simple method of statistical analysis that may be used is Wilcoxon's modification of Friedman's nonparametric analysis of variance for matched samples (5). This test minimizes the inherent approximation in Wilcoxon's matched-pairs sign-ranked test if a large number of ties are present, and requires only simple hand calculations. An example of some sample data is provided in the appendix (see Tables X1.1 to X1.4).

12. Report

12.1 A suggested listing of the data to be included in the report may be found in the appendix.

APPENDIX

(Nonmandatory Information)

X1. EXAMPLE OF WILCOXON'S MODIFIED NONPARAMETRIC ANOVA

X1.1 Rank data within each taster, assigning average rank for ties.

X1.2 Calculate the absolute differences between the rank sum for the control and the rank sum of the other concentrations.

X1.3 Compare the calculated results with Tables X1.1 and X1.2 where: p = the number of effluent concentrations and n = the number of tasters.

X1.4 If the calculated result is equal to or greater than the tabled (critical difference) result, then a significant difference exists.

TABLE X1.1 Critical Differences for Two-Way Classification: Comparing Several Treatments with a Control

n = 3(1)25 and p = 3(1)10
P = 0.01 (one-sided)

n	$p = 3$	$p = 4$	$p = 5$	$p = 6$	$p = 7$	$p = 8$	$p = 9$	$p = 10$
3	6	8	11	13	15	18	20	22
4	7	10	12	15	18	20	23	26
5	8	11	14	17	20	23	26	29
6	9	12	15	18	22	25	28	31
7	10	13	16	20	23	27	30	34
8	10	14	18	21	25	29	33	36
9	11	15	19	23	26	30	35	39
10	11	15	20	24	28	32	36	41
11	12	16	21	25	29	34	38	43
12	13	17	21	26	31	35	40	44
13	13	18	22	27	32	37	41	46
14	14	18	23	28	33	38	43	48
15	14	19	24	29	34	39	45	50
16	14	20	25	30	35	41	46	51
17	15	20	26	31	36	42	47	53
18	15	21	26	32	37	43	49	54
19	16	21	27	33	38	44	50	56
20	16	22	28	34	39	45	51	57
21	17	22	28	34	40	47	53	59
22	17	23	29	35	41	48	54	60
23	17	23	30	36	42	49	55	62
24	18	24	30	37	43	50	56	63
25	18	24	31	38	44	51	58	64

TABLE X1.2 Critical Differences for Two-Way Classification: Comparing Several Treatments with a Control

n = 3(1)25 and p = 3(1)10
P = 0.05 (one-sided)

n	$p = 3$	$p = 4$	$p = 5$	$p = 6$	$p = 7$	$p = 8$	$p = 9$	$p = 10$
3	5	7	8	10	12	14	16	18
4	5	8	10	12	14	16	18	21
5	6	8	11	13	16	18	21	23
6	7	9	12	14	17	20	23	25
7	7	10	13	16	19	21	24	27
8	8	11	14	17	20	23	26	29
9	8	11	14	18	21	24	28	31
10	9	12	15	19	22	26	29	33
11	9	12	16	20	23	27	31	34
12	9	13	17	20	24	28	32	36
13	10	14	17	21	25	29	33	37
14	10	14	18	22	26	30	34	39
15	11	15	19	23	27	31	36	40
16	11	15	19	24	28	32	37	41
17	11	16	20	24	29	33	38	43
18	12	16	20	25	30	34	39	44
19	12	16	21	26	30	35	40	45
20	12	17	22	26	31	36	41	46
21	12	17	22	27	32	37	42	47
22	13	18	23	28	33	38	43	49
23	13	18	23	28	34	39	44	50
24	13	18	24	29	34	40	45	51
25	14	19	24	30	35	41	46	52

TABLE X1.3 Modified from Wastewater Exposure, (7 Days Using Rainbow Trout 01-16-76) [A]

Taster	Percent Effluent					
	0	12.5	25	33	50	100
Fish Tainting Data						
A	0	0	0	1	0	4
B	0	0	0	0	1	3
C	1	2	0	3	1	4
D	0	0	1	1	1	3
E	0	3	3	3	3	3
F	0	0	1	2	2	1
G	0	0	2	2	3	3
H	1	0	0	1	3	4
Ranked Data						
A	2.5	2.5	2.5	5.0	2.5	6.0
B	2.5	2.5	2.5	2.5	5.0	6.0
C	2.5	4.0	1.0	5.0	2.5	6.0
D	1.5	1.5	4.0	4.0	4.0	6.0
E	1.0	4.0	4.0	4.0	4.0	4.0
F	1.5	1.5	3.5	5.5	5.5	3.5
G	1.5	1.5	3.5	3.5	5.5	5.5
H	3.5	1.5	1.5	3.5	5.0	6.0
	16.5	19.0	22.5	33.0	34.0	43.0
Δ From Control		2.5	6.0	16.5	17.0	26.5

[A] There are six concentrations and eight tasters. For $p = 6$ and $n = 8$, the critical difference is 17 (5 % level) or 21 (1 % level). Therefore, the 50 and 100 % concentrations are different at $p \leq 0.05$ and the 100% concentration different at $p \leq 0.01$ The other concentrations are not different at $p \leq 0.05$.

TABLE X1.4 Summary of Data to be Included in the Final Report

Principal investigator
Laboratory
Dates of samples, exposure, and taste evaluation
Chemical or physical properties of dilution water
Characteristics of stream, river, lake or effluent plus dilution water
(a) pH
(b) Total oxygen demand
(c) Chemical oxygen demand
(d) Total dissolved solids
(e) Suspended solids
(f) Dissolved oxygen
(g) Total organic carbon
(h) Conductivity
Species name, weight, length, number of fish exposed to each concentration, source.
Description of exposure system and length of exposure
Effects on fish
Flavor Evaluation.
(a) number of panel members
(b) relative experience level of each panel member, for example:
frequent tasting experience
some tasting experience
no previous tasting experience
(c) summary of raw data
Statistical method used to evaluate data.

FIG. X1.1 Sample Flavor Impairment Rating Sheet

Name _____

Date _____

Species of fish _____

Rating Code

0 Same or better than known control
1 Slight flavor impairment
2 Definite impairment
3 Bad
4 Repulsive

Sample	Rating	Comments
Known Control		

REFERENCES

(1) Shumway, D. L. and Palensky, J. R., "Impairment of the Flavor of Fish by Water Pollutants," Ecological Research Series No. EPA-R3-73-010 U.S. Environmental Protection Agency, Washington, D.C. 1973, p. 80.

(2) Mount, D. I. and Brungs, W. A., "A Simplified Dosing Apparatus for Fish Toxicological Studies," Water Res. Vol 1, 1967, pp. 21-29.

(3) *Basic Principles of Sensory Evaluation, ASTM STP 433*, ASTM 1968.

(4) *Manual on Sensory Testing Methods, ASTM STP 434*, ASTM 1968.

(5) Wilcoxon, F. and Wilcox, R. A., "Some Rapid Approximate Statistical Procedures," Lederle Laboratories, Pearle River, N.Y., 1964

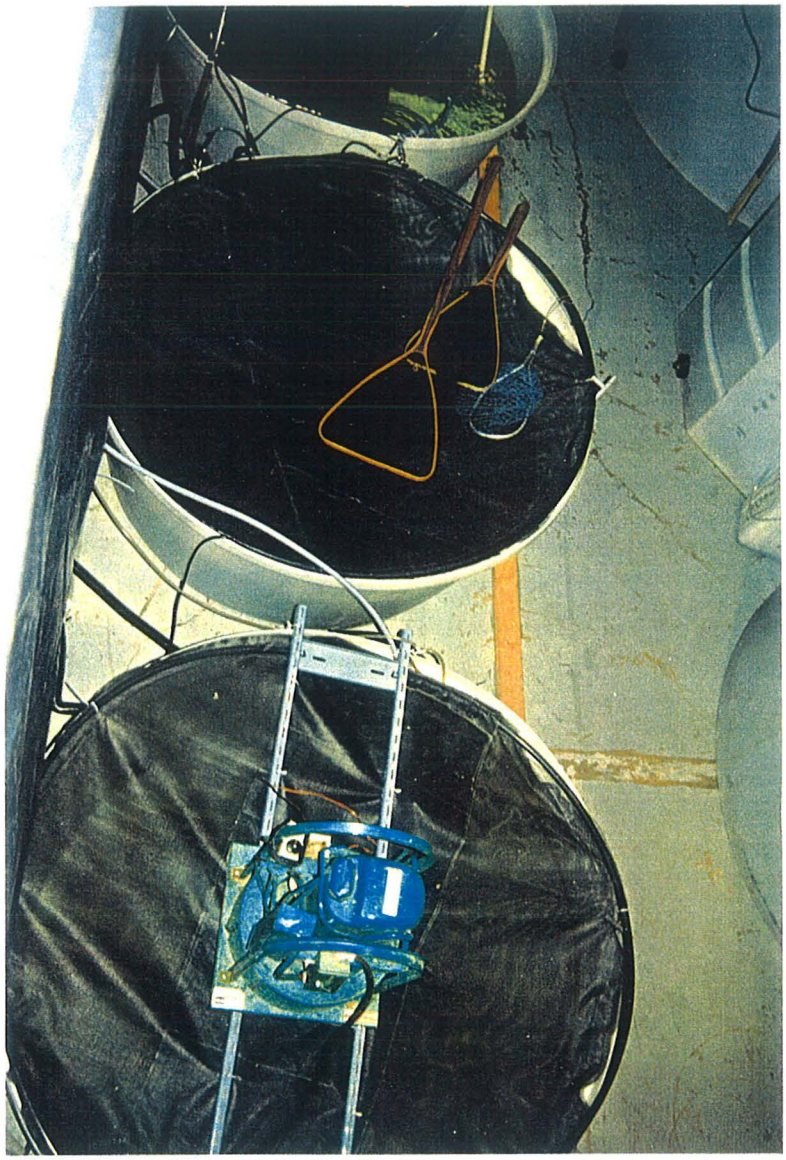


Figure 2. Overhead Photograph of Exposure and Sample Holding Tanks

(top to bottom, tar island dyke water, refinery effluent, and Athabasca River Water; the sample holding tanks are on the right hand side of the photograph)

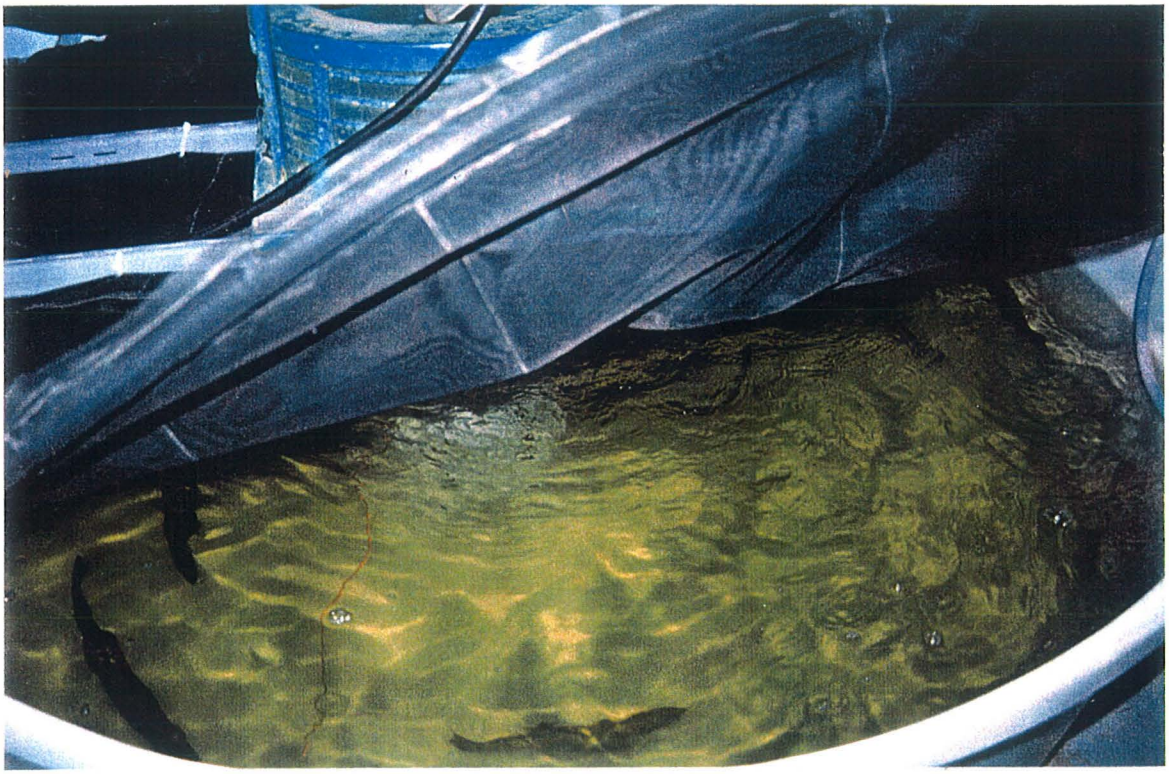


Figure 3. Photograph of Fish in Athabasca River Water

Figure 4. Photograph of Fish in 0.5% Tar Island Dyke Water

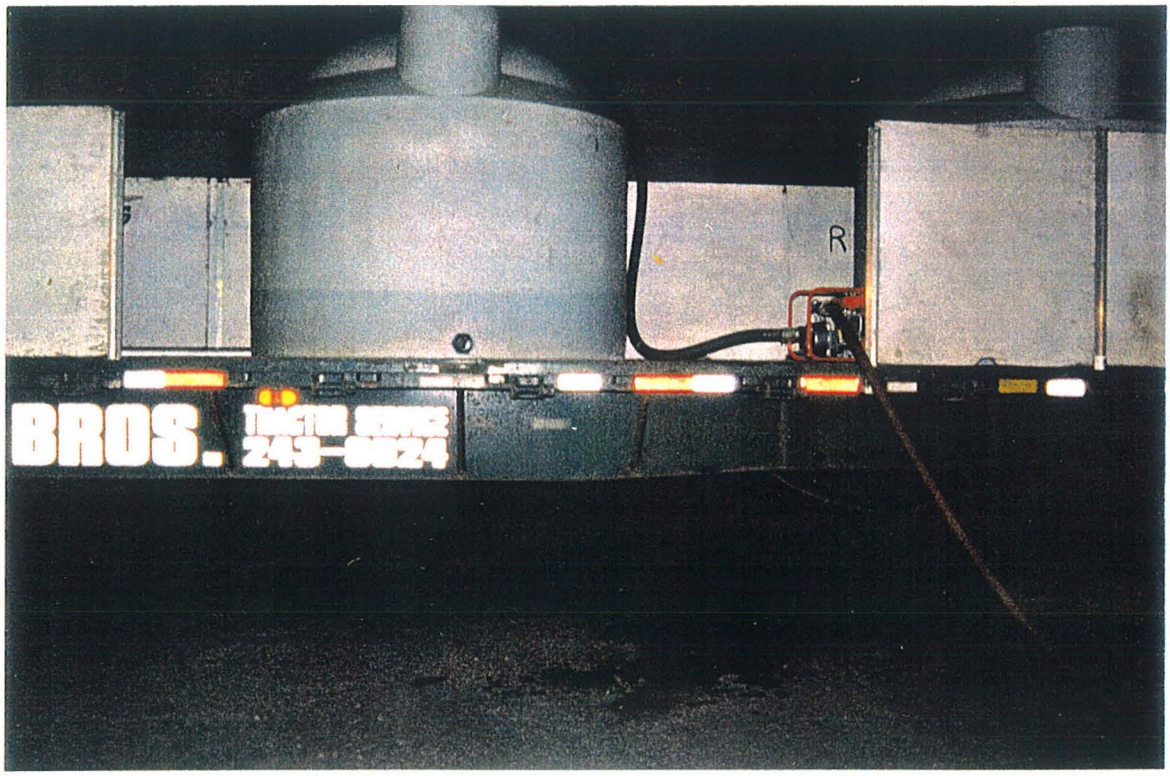


Figure 5. Photograph of Sample Transport and Handling Procedures

APPENDICES

APPENDIX I

Standard Practice for Evaluating an Effluent for Flavour Impairment to Fish Flesh, 1989

Designation: D 3696 - 89

Standard Practice for Evaluating an Effluent for Flavor Impairment to Fish Flesh[1]

This standard is issued under the fixed designation D 3696; the number immediately following the designation indicates the year of original adoption or, in the case of revision, the year of last revision. A number in parentheses indicates the year of last reapproval. A superscript epsilon (ε) indicates an editorial change since the last revision or reapproval.

1. Scope

1.1 This practice covers the potential of an effluent to cause flavor impairment of fish flesh. Caged fish can be exposed either in the effluent, at various sites relative to the discharge, or to dilutions of the effluent in a laboratory. Depending upon the uptake rate of the chemicals into the flesh from the discharge, from 1 day to several weeks may be required for a detectable off flavor. However, an exposure of 10 days is usually adequate. This practice is applicable to respective fish in fresh or salt water.

1.2 *This standard may involve hazardous materials, operations, and equipment. This standard does not purport to address all of the safety problems associated with its use. It is the responsibility of the user of this standard to establish appropriate safety and health practices and determine the applicability of regulatory limitations prior to use.* Specific hazard statements are given in Section 8.

2. Referenced Documents

2.1 *ASTM Standards:*

D 1125 Test Methods for Electrical Conductivity and Resistivity of Water[2]

D 1126 Test Methods for Hardness in Water[2]

D 1129 Terminology Relating to Water[2]

D 1192 Specification for Equipment for Sampling Water and Steam[2]

D 1252 Test Method for Chemical Oxygen Demand (Dichromate Oxygen Demand) of Water[2]

D 1293 Test Methods for pH of Water[2]

D 1888 Test Methods for Particulate and Dissolved Matter, Solids, or Residue in Water[2]

D 2579 Test Methods for Total and Organic Carbon in Water[3]

D 3250 Test Method for Total Oxygen Demand in Water[3]

D 3370 Practices for Sampling Water[2]

E 729 Guide for Conducting Acute Toxicity Tests With Fishes, Macroinvertebrates, and Amphibians[4]

3. Summary of Practice

3.1 Fish are exposed to an effluent or dilutions of an effluent either in the field or in the laboratory. After an exposure sufficient to allow tainting, fish are removed, cleaned and eviscerated, double-wrapped in metal foil, and refrigerated. Each fish sample including control fish, is encoded for identification. Fish are baked at 190°C for 20 to 30 min, then tasted by the testing panel. Flavor results are evaluated statistically to detect flavor impairment.

4. Significance and Use

4.1 This procedure, although subjective, may detect qualitative contamination of a fishery resource. Enforcement agencies usually recognize as a water quality standard that no substance shall be discharged into water that imparts an undesirable flavor to fish.

4.2 This practice does not eliminate all bias.

5. Terminology

5.1 *Definitions:*

5.1.1 For definitions of terms used in this practice, refer to Terminology D 1129.

5.2 *Description of Terms Specific to This Standard:*

5.2.1 *flavor impairment*—a detectable flavor deterioration between a test and control sample. Flavor tainting, off flavor, and undesirable flavor are considered synonyms.

6. Apparatus

6.1 *Field Study:*

6.1.1 *Cages*—Cages should be large enough to allow free swimming of the fish. The wire mesh or holes used to provide water circulation into and out of the cage should be small enough to retain small minnows, yet large enough to allow free passage of stream drift organisms. A 5-mm screen has proven satisfactory. See Fig. 1 for a typical fish exposure cage.

6.1.2 *Nets*—Nets should be pocketed to retain fish. Use soft nets to prevent abrasion of the test fish during handling. Thoroughly wash new nets to remove any textile finish present.

6.1.3 *Chest Waders or Hip Boots.*

6.1.4 *Boat*—Depending upon the nature of the study, especially when large rivers or lakes are being investigated, a boat is a necessary piece of equipment. The type of boat necessary must be matched to the type and size of the water body.

6.1.5 *Life Preservers*—There shall be one for each person.

6.1.6 *Holding Cages or Tankage*—Fish should be held for at least 10 days prior to testing.

6.2 *Laboratory Study:*

6.2.1 *Flow-Through System*—Many metering systems can

---

[1] This practice is under the jurisdiction of ASTM Committee E-47 on Biological Effects and Environmental Fate and is the direct responsibility of Subcommittee E47.01 on Aquatic Toxicology.

Current edition approved April 28, 1989. Published June 1989. Originally published as D 3696 - 78. Last previous edition D 3696 - 78 (1984)[ε1].

[2] *Annual Book of ASTM Standards*, Vol 11.01.

[3] *Annual Book of ASTM Standards*, Vol 11.02.

[4] *Annual Book of ASTM Standards*, Vol 11.04.

APPENDIX II

Fish Data

FISH DATA

The physical data and observations made on each fish are attached. The are four sets of data corresponding to the laboratory controls, Athabasca River Water, Tar Island Dyke water, and the refinery effluent. Each fish submitted to Diversified Laboratories Ltd., was weighed, measured, and examined. Scale samples were removed and archived.

HYDROQUAL LABORATORIES LTD.
FISH SAMPLE RECORD

PROJECT NO.	9522345	PERSONNEL	TB/SG/DM/JR/JH	TASK	0.5% RE	LOCATION	HydroQual Laboratories Ltd.

Date y/m/d	Fish No.	Species Code	Fork Length (mm)	Weight (g)	Sex	Maturity Code	Age Material	Comments
95/11/07	0001	RNTR	270	255	M	IM	SC	
95/11/07	0002	RNTR	285	260	M	IM	SC	
95/11/07	0003	RNTR	275	270	M	IM	SC	
95/11/07	0004	RNTR	275	220	M	IM	SC	
95/11/07	0005	RNTR	275	220	M	IM	SC	
95/11/07	0006	RNTR	290	265	M	IM	SC	
95/11/07	0007	RNTR	295	260	M	IM	SC	
95/11/07	0008	RNTR	285	310	M	IM	SC	
95/11/07	0009	RNTR	275	260	M	IM	SC	
95/11/07	0010	RNTR	265	220	M	IM	SC	
95/11/07	0011	RNTR	275	240	M	IM	SC	
95/11/07	0012	RNTR	235	160	M	IM	SC	
95/11/07	0013	RNTR	280	250	M	IM	SC	
95/11/07	0014	RNTR	285	270	M	IM	SC	
95/11/07	0015	RNTR	240	160	M	IM	SC	
95/11/07	0016	RNTR	265	240	M	IM	SC	
95/11/07	0017	RNTR	280	260	M	IM	SC	
95/11/07	0018	RNTR	255	190	M	IM	SC	
95/11/07	0019	RNTR	235	160	M	IM	SC	
95/11/07	0020	RNTR	270	310	M	IM	SC	
95/11/07	0021	RNTR	220	150	M	IM	SC	
95/11/07	0022	RNTR	255	190	M	IM	SC	
95/11/07	0023	RNTR	220	130	M	IM	SC	
95/11/07	0024	RNTR	265	240	M	IM	SC	
95/11/07	0025	RNTR	275	275	M	IM	SC	
95/11/07	0026	RNTR	270	270	M	IM	SC	
95/11/07	0027	RNTR	250	225	M	IM	SC	
95/11/07	0028	RNTR	265	220	M	IM	SC	
95/11/07	0029	RNTR	210	125	M	IM	SC	
95/11/07	0030	RNTR	195	100	M	IM	SC	
95/11/07	0031	RNTR	170	75	M	IM	SC	
95/11/07	0032	RNTR	152	60	M	IM	SC	
95/11/07	0033	RNTR	175	65	M	IM	SC	
95/11/07	0034	RNTR			M	IM	SC	
95/11/07	0035	RNTR			M	IM	SC	

NOTES: RNTR, Rainbow Trout; M, Male; IM, Immature; SC, Scales

HYDROQUAL LABORATORIES LTD.
FISH SAMPLE RECORD

PROJECT NO.	9522345	PERSONNEL	SG/MG/JR/JH/TB	TASK	T O Control Fish	LOCATION	HydroQual Laboratories Ltd.

Date y/m/d	Fish No.	Species Code	Fork Length (mm)	Weight (g)	Sex	Maturity Code	Age Material	Comments
95/10/27	0001	RNTR	290	230	M	IM	SC	
95/10/27	0002	RNTR	280	245	M	IM	SC	
95/10/27	0003	RNTR	265	175	M	IM	SC	
95/10/27	0004	RNTR	290	245	M	IM	SC	
95/10/27	0005	RNTR	255	190	M	IM	SC	
95/10/27	0006	RNTR	260	170	M	IM	SC	
95/10/27	0007	RNTR	270	210	M	IM	SC	
95/10/27	0008	RNTR	250	170	M	IM	SC	
95/10/27	0009	RNTR	275	240	M	IM	SC	
95/10/27	0010	RNTR	260	195	M	IM	SC	
95/10/27	0011	RNTR	265	195	M	IM	SC	
95/10/27	0012	RNTR	260	200	M	IM	SC	
95/10/27	0013	RNTR	285	240	M	IM	SC	
95/10/27	0014	RNTR	250	160	M	IM	SC	
95/10/27	0015	RNTR	280	210	M	IM	SC	
95/10/27	0016	RNTR	305	290	M	IM	SC	
95/10/27	0017	RNTR	275	210	M	IM	SC	
95/10/27	0018	RNTR	265	190	M	IM	SC	
95/10/27	0019	RNTR	300	290	M	IM	SC	
95/10/27	0020	RNTR	270	200	M	IM	SC	
95/10/27	0021	RNTR	275	240	M	IM	SC	
95/10/27	0022	RNTR	280	205	M	IM	SC	
95/10/27	0023	RNTR	265	190	M	IM	SC	
95/10/27	0024	RNTR	285	240	M	IM	SC	
95/10/27	0025	RNTR	295	300	M	IM	SC	
95/10/27	0026	RNTR	270	210	M	IM	SC	
95/10/27	0027	RNTR	305	300	M	IM	SC	
95/10/27	0028	RNTR	250	200	M	IM	SC	
95/10/27	0029	RNTR	270	210	M	IM	SC	
95/10/27	0030	RNTR	270	220	M	IM	SC	
95/10/27	0031	RNTR	290	230	M	IM	SC	
95/10/27	0032	RNTR	295	255	M	IM	SC	
95/10/27	0033	RNTR	285	230	M	IM	SC	
95/10/27	0034	RNTR	280	235	M	IM	SC	
95/10/27	0035	RNTR	290	240	M	IM	SC	

NOTES: RNTR, Rainbow Trout; M, Male; IM, Immature; SC, Scales

HYDROQUAL LABORATORIES LTD.
FISH SAMPLE RECORD

PROJECT NO.	9522345	PERSONNEL	SG/TB/DM/GD	TASK	Athabasca River	LOCATION	HydroQual Laboratories Ltd.

Date y/m/d	Fish No.	Species Code	Fork Length (mm)	Weight (g)	Sex	Maturity Code	Age Material	Comments
95/11/05	0001	RNTR	195	80	M	IM	SC	
95/11/05	0002	RNTR	275	210	M	IM	SC	
95/11/05	0003	RNTR	320	360	M	IM	SC	
95/11/05	0004	RNTR	310	275	M	IM	SC	
95/11/05	0005	RNTR	285	230	M	IM	SC	
95/11/05	0006	RNTR	285	240	M	IM	SC	
95/11/05	0007	RNTR	270	225	M	IM	SC	
95/11/05	0008	RNTR	280	240	M	IM	SC	
95/11/05	0009	RNTR	250	160	M	IM	SC	
95/11/05	0010	RNTR	245	130	M	IM	SC	
95/11/05	0011	RNTR	275	200	M	IM	SC	
95/11/05	0012	RNTR	280	210	M	IM	SC	
95/11/05	0013	RNTR	250	160	M	IM	SC	
95/11/05	0014	RNTR	265	240	M	IM	SC	
95/11/05	0015	RNTR	265	180	M	IM	SC	
95/11/05	0016	RNTR	240	140	M	IM	SC	
95/11/05	0017	RNTR	255	150	M	IM	SC	
95/11/05	0018	RNTR	285	270	M	IM	SC	
95/11/05	0019	RNTR	260	175	M	IM	SC	
95/11/05	0020	RNTR	275	210	M	IM	SC	
95/11/05	0021	RNTR	275	220	M	IM	SC	
95/11/05	0022	RNTR	285	225	M	IM	SC	
95/11/05	0023	RNTR	305	275	M	IM	SC	
95/11/05	0024	RNTR	270	210	M	IM	SC	
95/11/05	0025	RNTR	255	220	M	IM	SC	
95/11/05	0026	RNTR	280	225	M	IM	SC	
95/11/05	0027	RNTR	265	200	M	IM	SC	
95/11/05	0028	RNTR	290	275	M	IM	SC	
95/11/05	0029	RNTR	295	290	M	IM	SC	
95/11/05	0030	RNTR	265	210	M	IM	SC	
95/11/05	0031	RNTR	220	110	M	IM	SC	
95/11/05	0032	RNTR	240	150	M	IM	SC	
95/11/05	0033	RNTR	295	300	M	IM	SC	
95/11/05	0034	RNTR	285	225	M	IM	SC	
95/11/05	0035	RNTR	285	270	M	IM	SC	

NOTES: RNTR, Rainbow Trout; M, Male; IM, Immature; SC, Scales

HYDROQUAL LABORATORIES LTD.
FISH SAMPLE RECORD

PROJECT NO.	9522345	PERSONNEL	SG/TB/DM/GD	TASK	Athabasca River	LOCATION	HydroQual Laboratories Ltd.

Date y/m/d	Fish No.	Species Code	Fork Length (mm)	Weight (g)	Sex	Maturity Code	Age Material	Comments
95/11/05	0036	RNTR	265	200	M	IM	SC	
95/11/05	0037	RNTR	260	200	M	IM	SC	
95/11/05	0038	RNTR	285	200	M	IM	SC	
95/11/05	0039	RNTR	250	150	M	IM	SC	
95/11/05	0040	RNTR	260	160	M	IM	SC	

NOTES: RNTR, Rainbow Trout; M, Male; IM, Immature; SC, Scales

HYDROQUAL LABORATORIES LTD.
FISH SAMPLE RECORD

PROJECT NO.	9522345	PERSONNEL	SG/TB/DM/JR	TASK	0.5% TID	LOCATION	HydroQual Laboratories Ltd.

Date y/m/d	Fish No.	Species Code	Fork Length (mm)	Weight (g)	Sex	Maturity Code	Age Material	Comments
95/11/07	0001	RNTR	300	310	M	IM	SC	
95/11/07	0002	RNTR	300	290	M	IM	SC	
95/11/07	0003	RNTR	245	150	M	IM	SC	
95/11/07	0004	RNTR	270	240	M	IM	SC	
95/11/07	0005	RNTR	260	225	M	IM	SC	
95/11/07	0006	RNTR	195	100	M	IM	SC	
95/11/07	0007	RNTR	260	205	M	IM	SC	
95/11/07	0008	RNTR	260	175	M	IM	SC	
95/11/07	0009	RNTR	270	225	M	IM	SC	
95/11/07	0010	RNTR	265	180	M	IM	SC	
95/11/07	0011	RNTR	245	150	M	IM	SC	
95/11/07	0012	RNTR	275	250	M	IM	SC	
95/11/07	0013	RNTR	260	160	M	IM	SC	
95/11/07	0014	RNTR	265	240	M	IM	SC	
95/11/07	0015	RNTR	240	175	M	IM	SC	
95/11/07	0016	RNTR	310	340	M	IM	SC	
95/11/07	0017	RNTR	260	190	M	IM	SC	
95/11/07	0018	RNTR	260	200	M	IM	SC	
95/11/07	0019	RNTR	295	260	M	IM	SC	
95/11/07	0020	RNTR	265	210	M	IM	SC	
95/11/07	0021	RNTR	275	210	M	IM	SC	
95/11/07	0022	RNTR	245	180	M	IM	SC	
95/11/07	0023	RNTR	290	290	M	IM	SC	
95/11/07	0024	RNTR	295	275	M	IM	SC	
95/11/07	0025	RNTR	255	220	M	IM	SC	
95/11/07	0026	RNTR	285	240	M	IM	SC	
95/11/07	0027	RNTR	295	250	M	IM	SC	
95/11/07	0028	RNTR	200	110	M	IM	SC	
95/11/07	0029	RNTR	300	320	M	IM	SC	
95/11/07	0030	RNTR	265	210	M	IM	SC	
95/11/07	0031	RNTR	290	300	M	IM	SC	
95/11/07	0032	RNTR	260	240	M	IM	SC	
95/11/07	0033	RNTR	265	200	M	IM	SC	
95/11/07	0034	RNTR	285	310	M	IM	SC	
95/11/07	0035	RNTR	245	140	M	IM	SC	

NOTES: RNTR, Rainbow Trout; M, Male; IM, Immature; SC, Scales

APPENDIX III

Shipping Documentation

HydroQual Laboratories Ltd.

#3, 6125 - 12 Street S.E. Calgary, Alberta Canada T2H 2K1
TEL: (403) 253-7121 FAX: (403) 252-9363 1-800-808-6942
FILE: 951109DL.DOC

TRANSMITTAL

DATE: November 9, 1995

FROM: J. Stephen Goudey, Ph.D., P.Biol.
General Manager

TO: Lillian Lennox
Diversified Laboratories

TEL: (416) 922-5100
FAX: (416) 922-4318

TEL: (403) 253-7121
FAX: (403) 252-9363

RE: FISH TAINTING

TREATMENT: 0.5% REFINERY EFFLUENT
NUMBER OF FISH: 33 (200-250 g)

Please fill out the data, time, and sign this transmittal upon receipt of the cooler and FAX it back to me at (403)-252-9363.

DATE RECEIVED: Nov. 10/95

TIME RECEIVED: call from airport 9:00 am.

RECEIVED BY: J Lewis at airport

L Lennox

- sample frozen solid.

HydroQual Laboratories Ltd.

#3, 6125 - 12 Street S.E. Calgary, Alberta Canada T2H 2K1
TEL: (403) 253-7121 FAX: (403) 252-9363 1-800-808-6942

FILE: 951109DL.DOC

TRANSMITTAL

DATE: November 9, 1995

FROM: J. Stephen Goudey, Ph.D., P.Biol.
General Manager

TO: Lillian Lennox
Diversified Laboratories

TEL: (416) 922-5100
FAX: (416) 922-4318

TEL: (403) 253-7121
FAX: (403) 252-9363

RE: FISH TAINTING

TREATMENT: TIME ZERO CONTROL FISH
NUMBER OF FISH: 35 (200-250 g)

Please fill out the date, time, and sign this transmittal upon receipt of the cooler and FAX it back to me at (403)-252-9363.

DATE RECEIVED: Nov 10/95

TIME RECEIVED: call from airport 9:00 am

RECEIVED BY: Joe Louis at airport
L Lennox

N - samples frozen solid

HydroQual Laboratories Ltd

#3, 6125 - 12 Street S.E. Calgary, Alberta Canada T2H 2K1
TEL: (403) 253-7121 FAX: (403) 252-9363 1-800-808-6942

FILE: 951109DL.DOC

TRANSMITTAL

DATE: November 9, 1995

FROM: J. Stephen Gouday, Ph.D., P.Biol.
General Manager

TO: Lillian Lennox
Diversified Laboratories

TEL: (416) 922-5100
FAX: (416) 922-4318

TEL: (403) 253-7121
FAX: (403) 252-9363

RE: FISH TAINTING

TREATMENT: ATHABASCA RIVER
NUMBER OF FISH: 40 (250-275 g)

Please fill out the date, time, and sign this transmittal upon receipt of the cooler and FAX it back to me at (403)-252-9363.

DATE RECEIVED: Nov 10/95

TIME RECEIVED: call from airport 9:00 am

RECEIVED BY: Joe Louis at airport

L Lennox sample frozen solid

HydroQual Laboratories Ltd.
#3, 6125 - 12 Street S.E. Calgary, Alberta Canada T2H 2K1
TEL: (403) 253-7121 FAX: (403) 252-9363 1-800-808-6942

FILE: 951109DL.DOC

TRANSMITTAL

DATE: November 9, 1995

FROM: J. Stephen Goudey, Ph.D., P.Biol.
General Manager

TO: Lillian Lennox
Diversified Laboratories

TEL: (416) 922-5100
FAX: (416) 922-4318

TEL: (403) 253-7121
FAX: (403) 252-9363

RE: FISH TAINTING

TREATMENT: 0.5% TAR ISLAND DYKE
NUMBER OF FISH: 35 (200-250 g)

Please fill out the date, time, and sign this transmittal upon receipt of the cooler and FAX it back to me at (403)-252-9363.

DATE RECEIVED: Nov 10/95

TIME RECEIVED: phone call from airport 9:00 am.

RECEIVED BY: Joe Louis (picked up at airport)

J Lennox - sample frozen solid

APPENDIX IV

Results of Fish Tainting Evaluation (Diversified Research Report)

A CONFIDENTIAL
REPORT

RAINBOW TROUT
FISH TAINT ANALYSIS STUDY

Prepared for

Ms. Kym Holley
GOLDER ASSOCIATES LTD.
1011 6th Avenue S.W.
Calgary, Alberta
T2P 0W1

Diversified Research

Partners in Progress and Profitability

Prepared by

DIVERSIFIED RESEARCH
LABORATORIES LIMITED
1047 Yonge Street
Toronto, Ontario
M4W 2L2

DECEMBER 1995

Diversified Research Laboratories Limited

1047 Yonge Street
Toronto, Ontario
Canada M4W 2L2

Telephone: (416) 922-5100
Fax: (416) 922-4318
Ontario Toll Free: 1-800-387-0023

95-A-12349

December 28, 1995

RAINBOW TROUT FISH TAINT ANALYSIS STUDY

EXECUTIVE SUMMARY

OBJECTIVE

I) To determine if there is a perceivable difference between five samples of Rainbow Trout [(i) 0.5% Tar Island Dyke Water, (ii) Lab Athabasca River Water, (iii) Time Zero Control, (iv) 0.5% Refinery Effluent Water and (v) Field Athabasca River Water] using two successive (Double) Triangle Difference Tests, with the In-House Taste Panel at Diversified Research Laboratories. Difference to be determined between the following pairs of samples:

1) 0.5% Tar Island Dyke Water vs. Lab Athabasca River Water

2) 0.5% Tar Island Dyke Water vs. Time Zero Control

3) 0.5% Refinery Effluent Water vs. Lab Athabasca River Water

4) 0.5% Refinery Effluent Water vs. Time Zero Control

5) Lab Athabasca River Water vs. Time Zero Control

6) 0.5% Tar Island Dyke Water vs. Field Athabasca River Water

7) 0.5% Refinery Effluent Water vs. Field Athabasca River Water

8) Time Zero Control vs. Field Athabasca River Water

II) To determine if there is an overall preference between five samples of Rainbow Trout [(i) 0.5% Tar Island Dyke Water, (ii) Lab Athabasca River Water (iii) Time Zero Control, (iv) 0.5% Refinery Effluent Water and (v) Field Athabasca River Water] using two successive (Double) Overall Preference Rank Tests, with the In-House Taste Panel at Diversified Research Laboratories.

SUMMARY OF FINDINGS

I) <u>(Double) Triangle Difference Test</u>

The results of the (Double) Triangle Difference Test performed on five samples of Rainbow Trout are presented below. Thirty In-House Taste Panel respondents participated in each of the 8 evaluations. Each evaluation was performed twice (Test 1 and Test 2). The Combined result is based on the tabulation of only those panelists who correctly perceived a difference in <u>both</u> Test 1 and Test 2.

95-A-12349 December 28, 1995

SUMMARY OF FINDINGS (cont'd)

I) (Double) Triangle Difference Test (cont'd)

Sample	Test 1	Test 2	Combined
1) 0.5% Tar Island Dyke Water vs. Lab Athabasca River Water	Trend for Different (91% confidence level)	Not Different	Different (99% confidence level)
2) 0.5% Tar Island Dyke Water Vs. Time Zero Control	Not Different	Different (95% confidence level)	Not Different
3) 0.5% Refinery Effluent Water vs. Lab Athabasca River Water	Different (95% confidence level)	Not Different	Different (95% confidence level)
4) 0.5% Refinery Effluent Water vs. Time Zero Control	Not Different	Different (95% confidence level)	Not Different
5) Lab Athabasca River Water vs. Time Zero Control	Not Different	Different (95% confidence level)	Different (99% confidence level)
6) 0.5% Tar Island Dyke Water vs. Field Athabasca River Water	Different (95% confidence level)	Not Different	Different (95% confidence level)
7) 0.5% Refinery Effluent Water vs. Field Athabasca River Water	Different (95% confidence level)	Different (99% confidence level)	Different (99% confidence level)
8) Time Zero Control vs. Field Athabasca River Water	Trend for Different (91% confidence level)	Different (95% confidence level)	Not Different

95-A-12349 December 28, 1995

SUMMARY OF FINDINGS (cont'd)

I) (Double) Triangle Difference Test (cont'd)

In summary, some differences were found between the five samples of Rainbow Trout. Based on the Combined results (counting only those panelists who correctly identified the different sample in both Test 1 and Test 2), a difference was found between the following pairs of Rainbow Trout samples:

0.5% Tar Island Dyke Water vs Lab Athabasca River Water

0.5% Tar Island Dyke Water vs Field Athabasca River Water

0.5% Refinery Effluent Water vs Lab Athabasca River Water

0.5% Refinery Effluent Water vs Field Athabasca River Water

Lab Athabasca River Water vs Time Zero Control

II) (Double) Overall Preference Rank

Thirty In-House Taste Panel respondents participated in this evaluation. The Rainbow Trout samples were ranked for overall preference twice (Test 1 and Test 2). The results of the Overall Preference Rank tests are summarized below.

Overall Preference Rank - Summary

Sample / Test	0.5% TID Water	Lab Athabasca River Water	Time Zero Control	0.5% Refinery Effluent Water	Field Athabasca River Water
Test 1	Preferred (99% confidence level)	Neither preferred nor rejected	Preferred (95% confidence level)	Rejected (99% and 95% confidence level)	Trend for Preferred
Test 2	Neither preferred nor rejected	Neither preferred nor rejected	Preferred (99% confidence level)	Rejected (99% and 95% confidence level)	Preferred (95% confidence level)

95-A-12349 December 28, 1995

SUMMARY OF FINDINGS (cont'd)

II) (Double) Overall Preference Rank (cont'd)

Based on the results of the In-House Taste Panel, the 0.5% Refinery Effluent Water Rainbow Trout sample performed the poorest with respect to overall preference.

The Time Zero Control Rainbow Trout sample performed the best among the five samples with respect to overall preference.

The Field Athabasca River Water Rainbow Trout sample also performed well with respect to overall preference.

Approved by:

DIVERSIFIED RESEARCH
LABORATORIES LIMITED

Laura Sayles

Lillian Lennox
Sensory Evaluation & Consumer Research

Diversified Research Laboratories Limited

1047 Yonge Street
Toronto, Ontario
Canada M4W 2L2

Telephone: (416) 922-5100
Fax: (416) 922-4318
Ontario Toll Free: 1-800-387-0023

RAINBOW TROUT FISH TAINT ANALYSIS STUDY

TECHNICAL REPORT

OBJECTIVE

I) To determine if there is a perceivable difference between five samples of Rainbow Trout [(i) 0.5% Tar Island Dyke Water, (ii) Lab Athabasca River Water, (iii) Time Zero Control, (iv) 0.5% Refinery Effluent Water and (v) Field Athabasca River Water] using two successive (Double) Triangle Difference Tests, with the In-House Taste Panel at Diversified Research Laboratories. Difference to be determined between the following pairs of samples:

1) 0.5% Tar Island Dyke Water vs. Lab Athabasca River Water

2) 0.5% Tar Island Dyke Water vs. Time Zero Control

3) 0.5% Refinery Effluent Water vs. Lab Athabasca River Water

4) 0.5% Refinery Effluent Water vs. Time Zero Control

5) Lab Athabasca River Water vs. Time Zero Control

6) 0.5% Tar Island Dyke Water vs. Field Athabasca River Water

7) 0.5% Refinery Effluent Water vs. Field Athabasca River Water

8) Time Zero Control vs. Field Athabasca River Water

II) To determine if there is an overall preference between five samples of Rainbow Trout [(i) 0.5% Tar Island Dyke Water, (ii) Lab Athabasca River Water, (iii) Time Zero Control, (iv) 0.5% Refinery Effluent Water and (v) Field Athabasca River Water] using two successive (Double) Overall Preference Rank Tests, with the In-House Taste Panel at Diversified Research Laboratories.

SAMPLES

Rainbow Trout samples were received from Hydro Qual on Nov. 10, 1995 and Golder Associates on Oct. 18, 1995. The fish had been gutted and individually packaged in sealed plastic bags. The fish had been packed in coolers on dry ice during transportation. Upon receipt, the fish samples were stored frozen at -20°C and were tested within the 30 day time period limit.

SAMPLES (cont'd)

The fish samples were recorded as follows:

Rainbow Trout

Sample	Received from	Amount
1) 0.5% Tar Island Dyke	Hydro Qual	35 (200 - 250 g)
2) Lab Athabasca River	Hydro Qual	40 (250 - 275 g)
3) Time Zero Control	Hydro Qual	35 (200 - 250 g)
4) 0.5% Refinery Effluent	Hydro Qual	33 (200 - 250 g)
5) Field Athabasca River code: 001-021	Golder Associates	44 (weight not specified)

The fish samples were randomly selected for each evaluation. The codes of the specific fish used in each evaluation are recorded in Appendix B.

METHODOLOGY

In-House Taste Panel

The In-House Taste Panel at Diversified Research Laboratories is composed of Diversified Research employees who go through an orientation session where they become familiar with the basics of food tasting. The panelists attend taste panels on a regular basis and are readily able to verbalize their opinions about food products.

I) (Double) Triangle Difference Test

A (Double) Triangle Difference Test was used in this study. The Triangle Difference Test was performed in order to determine if there was a perceivable difference between two samples of Rainbow Trout. The following pairs of samples were tested:

1) 0.5% Tar Island Dyke Water vs. Lab Athabasca River Water

2) 0.5% Tar Island Dyke Water vs. Time Zero Control

METHODOLOGY (cont'd)

I) (Double) Triangle Difference Test (cont'd)

3) 0.5% Refinery Effluent Water vs. Lab Athabasca River Water

4) 0.5% Refinery Effluent Water vs. Time Zero Control

5) Lab Athabasca River Water vs. Time Zero Control

6) 0.5% Tar Island Dyke Water vs. Field Athabasca River Water

7) 0.5% Refinery Effluent Water vs. Field Athabasca River Water

8) Time Zero Control vs. Field Athabasca River Water

For each test, panelists received three samples: two of the samples were identical ("duplicate") and one was the different ("odd") sample. Panelists were asked to identify the "odd" sample.

Specifically, for this fish study, two Triangle Difference Tests were conducted in succession during one taste panel (Double Triangle Difference Test). If one sample is called "A" and the other sample is called "B", for the first test (Test 1), each panelist was presented two "A" samples and one "B" sample. Each sample was coded with a three digit number and evaluated in a fixed randomized order. For the second test (Test 2) the procedure was repeated, with each panelist receiving two "B" samples and one "A" sample.

Panelists filled out two questionnaires and did not know that the samples were the reciprocal of the previous test. The "Combined Test" result represents the number of panelists who correctly identified the "odd" sample in <u>both</u> Test 1 and Test 2.

The questionnaire and statistical treatment of the individual Triangle Difference Test ($p=1/3$) and the Combined Test ($p=1/9$) are found in Appendix A.

II) (Double) Overall Preference Rank Test

An Overall Preference Rank Test was performed in order to determine if there was a preference between the five samples of Rainbow Trout. Panelists were presented with 5 coded samples of Rainbow Trout (0.5% Tar Island Dyke, Lab Athabasca River, Time Zero Control, 0.5% Refinery Effluent and Field Athabasca River) and were asked to rank the samples in order of "most preferred" to "least preferred" (ie. ranked 1st, 2nd, 3rd, 4th and 5th). Samples were presented in a balanced randomized order.

Specifically, for this fish study, two Overall Preference Rank Tests were conducted in succession during one taste panel (Double Overall Preference Rank Test). The results of the two individual Overall Preference Rank Tests are reported as Test 1 and Test 2. The questionnaire and statistical treatment of the Overall Preference Rank Tests are presented in Appendix A.

METHODOLOGY (cont'd)

Sample Presentation

Approximately 5 mL of flaked fish was placed into a 100 mL white styrofoam cup with lid. The serving size was small, due to limited sample. All samples were identified with a set of randomly chosen three-digit numbers, for each of the three types of tests.

Taste Panel Procedure

All panelists signed a form agreeing to participate in the study. Panelists chewed and evaluated each sample and then expectorated the chewed sample into spit cups provided.

Dilute lemon rinse water (1 oz. lemon juice per 32 oz. spring water, room temperature) was provided to clear the palate before tasting each sample. White plastic forks were provided to facilitate tasting.

Panelists evaluated the samples in individual isolated sensory testing booths within the Sensory Evaluation Lab.

Sodium lights were used for the panel to mask any colour differences among the samples.

RESULTS AND DISCUSSION

Each evaluation was done twice, with the first evaluation called Test 1 and the second evaluation called Test 2.

I) (Double) Triangle Difference Test

1) 0.5% Tar Island Dyke (TID) Water vs. Lab Athabasca River Water

Thirty panelists participated in this evaluation.

The results of the 0.5% TID Water versus Lab Athabasca River Water (Double) Triangle Difference Test are presented in Table 1.

As may be observed in this table, in Test 1, there was a trend for a difference to be perceived between 0.5% TID Water and Lab Athabasca River Water Rainbow Trout samples (91% confidence level). Fourteen out of 30 panelists correctly identified the different sample in Test 1.

In Test 2, no significant difference was found between 0.5% TID Water and Lab Athabasca River Water samples of Rainbow Trout (95% confidence level). Thirteen out of the 30 panelists correctly identified the different sample in Test 2.

RESULTS AND DISCUSSION (cont'd)

I) (Double) Triangle Difference Test (cont'd)

1) 0.5% Tar Island Dyke (TID) Water vs. Lab Athabasca River Water (cont'd)

When the results of the two Triangle Difference Tests were combined, and only those panelists who correctly identified the different sample in both tests were considered, a significant difference was perceived between the 0.5% TID Water and Lab Athabasca River Rainbow Trout samples (95% confidence level). Eight out of the 30 panelists correctly identified the odd sample in both Triangle Difference Tests.

Panelists' comments about the difference perceived between 0.5% TID Water and Lab Athabasca River Water Rainbow Trout samples are presented in Table 2.

2) 0.5% Tar Island Dyke (TID) Water vs. Time Zero Control

Thirty panelists participated in this evaluation.

The results of the 0.5% TID Water versus Time Zero Control (Double) Triangle Difference Test are presented in Table 3.

As may be observed in Table 3, no significant difference was perceived between 0.5% TID Water and Time Zero Control samples in Test 1 (95% confidence level). A significant difference was perceived in Test 2 (95% confidence level). Ten out of 30 panelists correctly identified the different sample in Test 1, while 15 out of 30 panelists correctly identified the different sample in Test 2.

The Combined result, where only those panelists who correctly identified the different sample in both Test 1 and Test 2 were counted, showed no significant difference between the 0.5% TID Water and Time Zero Control Rainbow Trout samples (95% confidence level). Five out of 30 panelists correctly identified the different sample in both Triangle Difference Tests.

Panelists' comments about the difference perceived between the 0.5% TID Water and Time Zero Control Rainbow Trout samples are presented in Table 4.

3) 0.5% Refinery Effluent Water vs. Lab Athabasca River Water

Thirty panelists participated in this evaluation.

In Table 5, the results of the 0.5% Refinery Effluent Water versus Lab Athabasca River Water Rainbow Trout (Double) Triangle Difference test are presented.

As may be observed in this table, a significant difference was perceived between 0.5% Refinery Effluent Water versus Lab Athabasca River Water Rainbow Trout in Test 1, but not in Test 2 (95% confidence level). In Test 1, 15 out of the 30 panelists correctly identified the different sample; in Test 2, 13 out of the 30 panelists correctly identified the different sample.

RESULTS AND DISCUSSION (cont'd)

I) (Double) Triangle Difference Test (cont'd)

3) 0.5% Refinery Effluent Water vs. Lab Athabasca River Water (cont'd)

The Combined result, where only those panelists who correctly identified the different sample in both Test 1 and Test 2 were counted, showed a significant difference was found between 0.5% Refinery Effluent Water and Lab Athabasca River Water Rainbow Trout (95% confidence level).

Panelists' comments about the difference perceived between the 0.5% Refinery Effluent Water and the Lab Athabasca River Water Rainbow Trout are presented in Table 6.

4) 0.5% Refinery Effluent vs. Time Zero Control

Thirty panelists participated in this evaluation.

The results of the 0.5% Refinery Effluent vs. Time Zero Control Rainbow Trout (Double) Triangle Difference Test are presented in Table 7.

As may be observed in Table 7, no significant difference was found between the 0.5% Refinery Effluent and Time Zero Control samples of Rainbow Trout in Test 1 (95% confidence level). Ten out of 30 panelists correctly identified the different sample in Test 1. In Test 2, a significant difference was perceived between 0.5% Refinery Effluent and Time Zero Control Rainbow Trout samples (95% confidence level). Fifteen out of 30 panelists correctly identified the different sample.

The Combined result (only those panelists who correctly identified the different sample in both Test 1 and Test 2) showed no significant difference between 0.5% Refinery Effluent and Time Zero Control samples of Rainbow Trout (95% confidence level).

Five out of the 30 panelists correctly identified the different sample in both Triangle Difference Tests.

In Table 8, panelists' comments about the 0.5% Refinery Effluent and Time Zero Control Rainbow Trout samples are summarized.

5) Lab Athabasca River Water versus Time Zero Control

Thirty panelists participated in this evaluation.

In Table 9, the results of the Lab Athabasca River Water versus Time Zero Control Rainbow Trout (Double) Triangle Difference test are presented.

As may be viewed in this table, no difference was found in Test 1 (95% confidence level). Twelve out of 30 panelists correctly identified the different sample.

RESULTS AND DISCUSSION (cont'd)

I) (Double) Triangle Difference Test (cont'd)

5) Lab Athabasca River Water versus Time Zero Control (cont'd)

In Test 2, a difference was perceived between Lab Athabasca River Water and Time Zero Control Rainbow Trout samples (95% confidence level). Sixteen out of 30 panelists correctly perceived a difference.

The Combined result, where only those panelists who correctly identified the different sample in both tests were counted, showed a significant difference was perceived between Lab Athabasca River Water and Time Zero Control Rainbow Trout samples (99% confidence level). Nine out of the 30 panelists correctly identified the different sample in both Test 1 and Test 2.

Panelists' comments about the difference perceived between Lab Athabasca River Water and Time Zero Control Rainbow Trout samples are presented in Table 10.

6) 0.5% Tar Island Dyke Water (TID) vs. Field Athabasca River Water

Thirty panelists participated in this evaluation.

The results of the 0.5% TID Water versus Lab Athabasca River Water (Double) Triangle Difference Test are presented in Table 11.

As may be observed in this table, in Test 1, a difference was perceived between 0.5% TID Water and Field Athabasca River Water Rainbow Trout samples (95% confidence level). Fifteen out of 30 panelists correctly identified the different sample.

In Test 2, no significant difference was found between 0.5% TID Water and Field Athabasca River Water samples of Rainbow Trout (95% confidence level). Thirteen out of 30 panelists correctly identified the different sample in Test 2.

The Combined result (number of panelists who correctly identified the different sample in both Test 1 and Test 2) indicated a significant difference between 0.5% TID Water and Field Athabasca River Water Rainbow Trout samples. Seven out of 30 panelists correctly identified the different sample in both Test 1 and Test 2.

Panelists' comments about the 0.5% TID Water vs. Field Athabasca River Water Rainbow Trout samples are presented in Table 12.

RESULTS AND DISCUSSION (cont'd)

I) (Double) Triangle Difference Test (cont'd)

7) 0.5% Refinery Effluent vs. Field Athabasca River Water

Thirty panelists participated in this evaluation.

In Table 13, the results of the 0.5% Refinery Effluent vs. Field Athabasca River Water (Double) Triangle Difference Test are presented.

As may be observed in Table 13, a clear difference was perceived between these two Rainbow Trout samples. A significant difference was found between 0.5% Refinery Effluent and Field Athabasca River Water samples of Rainbow Trout in Test 1 (95% confidence level), Test 2 (99% confidence level) and in the Combined results (99% confidence level).

Panelists' comments about the 0.5% Refinery Effluent vs. the Field Athabasca River Water Rainbow Trout samples are presented in Table 14.

8) Time Zero Control vs. Field Athabasca River Water

Thirty panelists participated in this evaluation.

The results of the Time Zero Control versus Field Athabasca River Water Rainbow Trout (Double) Triangle Difference Test are presented in Table 15.

As may be observed in this table, in Test 1, there was a trend for a difference to be perceived between Time Zero Control and Field Athabasca River Water Rainbow Trout samples. Fourteen out of the 30 panelists correctly identified the different sample in Test 1.

In Test 2, a significant difference was found between Time Zero Control and Field Athabasca River Water Rainbow Trout samples. Fifteen out of 30 panelists correctly identified the different sample in Test 2.

The Combined result (counting the number of panelists who correctly identified the different sample in both Test 1 and Test 2) showed no significant difference between the Time Zero Control and Field Athabasca River Water samples. Five out of 30 panelists correctly identified the different sample in both Test 1 and Test 2.

Panelists' comments about the difference perceived between Time Zero Control and Field Athabasca River Water Rainbow Trout samples are presented in Table 16.

RESULTS AND DISCUSSION (cont'd)

II) (Double) Overall Preference Rank

Thirty panelists participated in this evaluation. The Overall Preference Rank Test was performed twice, with the first evaluation called Test 1 and the second evaluation called Test 2. The results of the (Double) Overall Preference Rank Test are presented in Tables 17 and 17-A.

As may be observed in Table 17 (Test 1), 0.5% TID Water and Time Zero Control Rainbow Trout samples were ranked overall preferred over 0.5% Refinery Effluent Rainbow Trout (99% and 95% confidence level, respectively).

There was a trend for the Field Athabasca River sample to also be ranked overall preferred over the 0.5% Refinery Effluent Rainbow Trout sample.

No other overall preferences were shown between the five Rainbow Trout samples in Test 1.

In Test 2 (Table 17-A), Time Zero Control and Field Athabasca River Water Rainbow Trout samples were ranked overall preferred over 0.5% Refinery Effluent Rainbow Trout (99% and 95% confidence level, respectively).

No other overall preferences were shown between the five samples of Rainbow Trout in Test 2.

In summary, the findings from Test 1 and Test 2 Overall Preference Rank Test indicate that the 0.5% Refinery Effluent Rainbow Trout sample performed the most poorly with respect to overall preference. Time Zero Control Rainbow Trout performed the best among the five samples, with respect to overall preference. The Field Athabasca River Rainbow Trout sample also had a good showing with respect to overall preference.

CONCLUSION

Results from the In-House Taste Panel (n=30) indicate some differences were found between the five samples of Rainbow Trout. Based on the Combined result (counting only those panelists who correctly identified the different sample in both Test 1 and 2) a difference was found between the following pairs of Rainbow Trout samples:

0.5% Tar Island Dyke Water vs Lab Athabasca River Water

0.5% Tar Island Dyke Water vs Field Athabasca River Water

0.5% Refinery Effluent Water vs Lab Athabasca River Water

0.5% Refinery Effluent Water vs Field Athabasca River Water

Lab Athabasca River Water vs Time Zero Control

CONCLUSION (cont'd)

With respect to overall preference, the 0.5% Refinery Effluent Water Rainbow Trout sample performed the poorest. The Time Zero Control Rainbow Trout sample performed the best among the five samples with repect to overall preference. The Field Athabasca River Water Rainbow Trout sample also performed well with respect to overall preference.

Approved by:

DIVERSIFIED RESEARCH
LABORATORIES LIMITED

Laura Sayles

Lillian Lennox
Sensory Evaluation & Consumer Research

Diversified Research
Laboratories Limited

95-A-12349 December 28, 1995

TABLE 1
RAINBOW TROUT

0.5% TID WATER VS. LAB ATHABASCA RIVER WATER

---

1) 0.5% TID Water vs. Lab Athabasca River Water

(Double) Triangle Difference Test

	Test 1	Test 2	Combined[1]
Total number of panelists participating	30	30	30
Number of panelists who correctly identified the "odd" sample	14^{+}	13^{+}	8**
Degree of difference perceived	1.9 (slight - moderate)	2.2 (moderate - much)	1.9 (slight - moderate)
Number of panelists incorrectly identifying the "odd" sample	16	17	22

---

[1] Of the 30 panelists participating, those panelists who correctly identified the odd sample in both Test 1 and Test 2.

$^{+}$ Not statistically significant (95% confidence level)
** Statistically significant (99% confidence level)

Critical value required for significance:	Test 1 & Test 2	(95% confidence) = 15/30 (99% confidence) = 17/30
	Combined	(95% confidence) = 7/30 (99% confidence) = 8/30

Test 1 - Actual confidence level achieved = 91%
Test 2 - Actual confidence level achieved = 83%

Diversified Research
Laboratories Limited

95-A-12349 December 28, 1995

TABLE 2
RAINBOW TROUT - PANEL #1
PANELISTS' VERBATIM COMMENTS

0.5% TAR ISLAND DYKE (TID) VS. LAB ATHABASCA RIVER

	Base	Number of Panelists Stating Test 1 (14)	Test 2 (13)
0.5% TID			
- more flavour		1	2
- strong "fishy" flavour		1	1
- fishier flavour		1	-
- more fishy / oil flavour		1	2
- musty flavour		2	1
- grassy flavour		1	-
- less flavour		-	1
- no off flavour		-	2
- firmer texture		1	3
- drier texture		2	2
Lab Athabasca River			
- blander / milder flavour		2	-
- less "fishy" tasting		3	1
- less fish oil flavour		1	1
- no grassy flavour		1	-
- no musty flavour		1	-
- "earthy" odour and flavour		-	1
- less fresh tasting		1	-
- bitter		1	-
- more moist		3	1
- softer texture		2	-
- more juicy		1	-
- mushy texture		-	2

95-A-12349 December 28, 1995

TABLE 3
RAINBOW TROUT

0.5% TID WATER VS. TIME ZERO CONTROL

---

2) 0.5% TID Water vs. Time Zero Control

(Double) Triangle Difference Test

	Test 1	Test 2	Combined[1]
Total number of panelists participating	30	30	30
Number of panelists who correctly identified the "odd" sample	10+	15*	5+
Degree of difference perceived	1.1 (slight - moderate)	1.5 (slight - moderate)	1.2 (slight - moderate)
Number of panelists incorrectly identifying the "odd" sample	20	15	25

---

[1] Of the 30 panelists participating, those panelists who correctly identified the odd sample in both Test 1 and Test 2.

+ Not statistically significant (95% confidence level)
* Statistically significant (95% confidence level)

Critical value required for significance:	Test 1 & Test 2	(95% confidence) = 15/30 (99% confidence) = 17/30
	Combined	(95% confidence) = 7/30 (99% confidence) = 8/30

95-A-12349 December 28, 1995

TABLE 4
RAINBOW TROUT - PANEL #2
PANELISTS' VERBATIM COMMENTS

0.5% TAR ISLAND DYKE (TID) VS. TIME ZERO CONTROL

	Base	Number of Panelists Stating Test 1 (10)	Test 2 (15)
0.5% TID			
- fishier tasting		2	-
- more fish flavour		1	2
- fresher fish flavour		2	-
- sweet fish flavour / sweeter		1	1
- less sour / fishy flavour		1	-
- more bland / slightly milder flavour		1	1
- chemical flavour		-	1
- petroleum flavour		1	1
- grassy flavour		-	1
- bitter, not sweet		-	1
- stronger off flavour		-	1
- swampy aftertaste		-	1
- tinny / metallic taste		1	-
- drier texture		2	1
- juicier texture		1	-
- softer texture		-	1

95-A-12349 December 28, 1995

TABLE 4 (cont'd)
RAINBOW TROUT - PANEL #2
PANELISTS' VERBATIM COMMENTS

0.5% TAR ISLAND DYKE (TID) VS. TIME ZERO CONTROL

	Base	Number of Panelists Stating Test 1 (10)	Test 2 (15)
Time Zero Control			
- less fishy flavour		1	-
- weaker flavour		1	-
- bland flavour		-	2
- fresher flavour		-	1
- no petroleum flavour		1	-
- stronger fish flavour		1	-
- fishy flavour		1	-
- more sour		1	-
- swampy tasting		1	-
- slight grassy flavour		2	-
- earthy flavour		-	1
- dirty aftertaste		1	-
- drier texture		2	-
- less chewy		1	-

95-A-12349 December 28, 1995

TABLE 5
RAINBOW TROUT

0.5% REFINERY EFFLUENT VS. LAB ATHABASCA RIVER WATER

---

3) 0.5% Refinery Effluent vs. Lab Athabasca River Water

(Double) Triangle Difference Test

	Test 1	Test 2	Combined[1]
Total number of panelists participating	30	30	30
Number of panelists who correctly identified the "odd" sample	15*	13+	7*
Degree of difference perceived	1.3 (slight - moderate)	1.2 (slight - moderate)	1.2 (slight - moderate)
Number of panelists incorrectly identifying the "odd" sample	15	17	23

---

[1] Of the 30 panelists participating, those panelists who correctly identified the odd sample in both Test 1 and Test 2.

+ Not statistically significant (95% confidence level)
* Statistically significant (95% confidence level)

Critical value required for significance:	Test 1 & Test 2	(95% confidence) = 15/30
		(99% confidence) = 17/30
	Combined	(95% confidence) = 7/30
		(99% confidence) = 8/30

95-A-12349 December 28, 1995

TABLE 6
RAINBOW TROUT - PANEL #3
PANELISTS' VERBATIM COMMENTS

0.5% REFINERY EFFLUENT VS. LAB ATHABASCA RIVER WATER

	Base	Number of Panelists Stating Test 1 (15)	Test 2 (13)
0.5% Refinery Effluent			
- stronger flavour		-	1
- stronger fishy flavour		-	1
- more flavour		1	1
- cleaner flavour		1	-
- better tasting		2	-
- less flavour		1	-
- sweet flavour		1	-
- more flavour		-	1
- muddy taste		-	1
- grassy flavour		1	-
- stronger fish oil aftertaste		-	1
- moister		-	1
- juicier		1	1
- soft		2	-
- slightly drier		-	2

95-A-12349 December 28, 1995

TABLE 6 (cont'd)
RAINBOW TROUT - PANEL #3
PANELISTS' VERBATIM COMMENTS

0.5% REFINERY EFFLUENT VS. LAB ATHABASCA RIVER WATER

	Base	Number of Panelists Stating Test 1 (15)	Test 2 (13)
Lab Athabasca River Water			
- bland		3	1
- milder flavour		1	2
- less fish flavour		1	-
- tastier		1	-
- sweeter tasting		1	-
- more flavour		2	-
- strong fish flavour		-	1
- swampy taste		1	-
- bitter		1	1
- more moist		2	1
- slightly mushier		1	-
- less juicy		1	-
- firmer		1	-
- slightly dry		-	2

95-A-12349 December 28, 1995

TABLE 7
RAINBOW TROUT

0.5% REFINERY EFFLUENT VS. TIME ZERO CONTROL

---

4) 0.5% Refinery Effluent vs. Time Zero Control

(Double) Triangle Difference Test

	Test 1	Test 2	Combined[1]
Total number of panelists participating	30	30	30
Number of panelists who correctly identified the "odd" sample	10[+]	15*	5[+]
Degree of difference perceived	1.1 (slight - moderate)	1.1 (slight - moderate)	1.2 (slight - moderate)
Number of panelists incorrectly identifying the "odd" sample	20	15	25

---

[1] Of the 30 panelists participating, those panelists who correctly identified the odd sample in both Test 1 and Test 2.

[+] Not statistically significant (95% confidence level)
* Statistically significant (95% confidence level)

Critical value required for significance:

Test 1 & Test 2	(95% confidence) = 15/30 (99% confidence) = 17/30
Combined	(95% confidence) = 7/30 (99% confidence) = 8/30

95-A-12349 December 28, 1995

TABLE 8
RAINBOW TROUT - PANEL #4
PANELISTS' VERBATIM COMMENTS

0.5% REFINERY EFFLUENT VS. TIME ZERO CONTROL

	Base	Number of Panelists Stating Test 1 (10)	Test 2 (15)
0.5% Refinery Effluent			
- OK flavour		-	1
- tasteless		-	1
- less muddy flavour		-	1
- slight muddy flavour		1	-
- fresher taste		-	1
- sweet taste		-	1
- slightly sweeter flavour		-	1
- less flavour		1	-
- worse flavour		-	1
- no off flavour		-	1
- better aftertaste		-	1
- slight aftertaste		-	1
- slight bitter taste		-	1
- less bitter		1	-
- slight bitter aftertaste		1	-
- fishy aftertaste		1	-
- drier texture		1	3
- slightly less moist		1	-
- firmer meat		1	1
- moister		-	1

95-A-12349 December 28, 1995

TABLE 8 (cont'd)
RAINBOW TROUT - PANEL #4
PANELISTS' VERBATIM COMMENTS

0.5% REFINERY EFFLUENT VS. TIME ZERO CONTROL

	Base	Number of Panelists Stating Test 1 (10)	Test 2 (15)
Time Zero Control			
- better flavour		-	1
- less fishy flavour		1	-
- bland		-	2
- slightly better aftertaste		1	-
- cleaner aftertaste		1	-
- no bitter aftertaste		1	-
- stronger flavour		1	-
- dirty taste		1	1
- slight metallic flavour		-	1
- slight muddy flavour		-	1
- clay flavour		-	1
- more bitter		1	1
- slightly moister		1	-
- juicier		1	-
- softer		-	1
- less moist		-	1

95-A-12349 December 28, 1995

TABLE 9
RAINBOW TROUT

LAB ATHABASCA RIVER WATER VS. TIME ZERO CONTROL

---

5) Lab Athabasca River Water vs. Time Zero Control

(Double) Triangle Difference Test

	Test 1	Test 2	Combined[1]
Total number of panelists participating	30	30	30
Number of panelists who correctly identified the "odd" sample	12[+]	16*	9**
Degree of difference perceived	1.2 (slight - moderate)	1.6 (slight - moderate)	1.4 (slight - moderate)
Number of panelists incorrectly identifying the "odd" sample	18	14	21

---

[1] Of the 30 panelists participating, those panelists who correctly identified the odd sample in both Test 1 and Test 2.

[+] Not statistically significant (95% confidence level)
* Statistically significant (95% confidence level)
** Statistically significant (99% confidence level)

Critical value required for significance:	Test 1 & Test 2	(95% confidence) = 15/30 (99% confidence) = 17/30
	Combined	(95% confidence) = 7/30 (99% confidence) = 8/30

95-A-12349 December 28, 1995

TABLE 10
RAINBOW TROUT - PANEL #5
PANELISTS' VERBATIM COMMENTS

LAB ATHABASCA RIVER VS. TIME ZERO CONTROL

		Number of Panelists Stating	
		Test 1	Test 2
	Base	(12)	(16)
Lab Athabasca River			
- milder / blander flavour		-	2
- less flavour		-	1
- cleaner fish flavour		1	1
- less fishy taste		-	1
- no musty flavour		-	1
- no grassy flavour		-	1
- fresher fish flavour		-	1
- sweeter		1	1
- no off flavour		2	2
- less bitter fish flavour		-	1
- more initial flavour		-	1
- slight chemical taste		-	1
- firmer texture		-	1
- drier texture		-	1
- moist		-	1

95-A-12349 December 28, 1995

TABLE 10 (cont'd)
RAINBOW TROUT - PANEL #5
PANELISTS' VERBATIM COMMENTS

LAB ATHABASCA RIVER VS. TIME ZERO CONTROL

	Base	Number of Panelists Stating Test 1 (12)	Test 2 (16)
Time Zero Control			
- better flavour		1	-
- fresher flavour		1	-
- less fishy		1	-
- bland taste		-	1
- stronger flavour		2	-
- less fresh flavour		1	-
- mild grassy flavour		1	1
- grassy off flavour		-	1
- musty flavour		-	2
- earthy flavour		1	-
- swampy taste / aftertaste		1	1
- slight metallic taste		-	1
- slight chemical flavour		1	-
- dirty petroleum flavour		1	-
- slight fishy off flavour		1	-
- bitter		-	2
- oily flavour		-	1

95-A-12349 December 28, 1995

TABLE 11
RAINBOW TROUT

0.5% TID WATER VS. FIELD ATHABASCA RIVER WATER

---

6) 0.5% TID Water vs. Field Athabasca River Water

(Double) Triangle Difference Test

	Test 1	Test 2	Combined[1]
Total number of panelists participating	30	30	30
Number of panelists who correctly identified the "odd" sample	15*	13+	7*
Degree of difference perceived	1.8 (slight - moderate)	1.6 (slight - moderate)	1.8 (slight - moderate)
Number of panelists incorrectly identifying the "odd" sample	15	17	23

---

[1] Of the 30 panelists participating, those panelists who correctly identified the odd sample in both Test 1 and Test 2.

+ Not statistically significant (95% confidence level)
* Statistically significant (95% confidence level)

Critical value required for significance:	Test 1 & Test 2	(95% confidence) = 15/30
		(99% confidence) = 17/30
	Combined	(95% confidence) = 7/30
		(99% confidence) = 8/30

95-A-12349 December 28, 1995

TABLE 12
RAINBOW TROUT - PANEL #6
PANELISTS' VERBATIM COMMENTS

0.5% TAR ISLAND DYKE (TID) VS. FIELD ATHABASCA RIVER

	Base	Number of Panelists Stating Test 1 (15)	Test 2 (13)
0.5% TID			
- stronger fishy taste		2	-
- awful fishy flavour		-	1
- chemical flavour		-	1
- metallic off flavour		1	1
- metallic aftertaste		1	1
- muddy flavour		1	-
- swampy flavour		1	1
- bitter		1	-
- more flavour		-	1
- milder		-	1
- bland		-	1
- less fishy flavour		-	1
- softer		1	2
- moist		1	2
- juicier		-	1
- drier		-	1

95-A-12349 December 28, 1995

TABLE 12 (cont'd)
RAINBOW TROUT - PANEL #6
PANELISTS' VERBATIM COMMENTS

0.5% TAR ISLAND DYKE (TID) VS. FIELD ATHABASCA RIVER

	Base	Number of Panelists Stating Test 1 (15)	Test 2 (13)
Field Athabasca River			
- better flavour		1	-
- milder flavour		1	-
- bland		-	1
- less fishy flavour		1	1
- sweet tasting		1	1
- less musty flavour		1	-
- no swampy taste		1	-
- less aftertaste		1	-
- stronger fish flavour		1	1
- slightly fishier taste		1	-
- didn't taste as fresh		1	-
- more fishy aftertaste		1	-
- bad aftertaste		1	1
- drier		4	1
- more chewy		1	-
- juicier		1	-
- moister		1	-

95-A-12349 December 28, 1995

TABLE 13
RAINBOW TROUT

0.5% REFINERY EFFLUENT VS. FIELD ATHABASCA RIVER WATER

7) **0.5% Refinery Effluent vs. Field Athabasca River Water**

(Double) Triangle Difference Test

	Test 1	Test 2	Combined[1]
Total number of panelists participating	30	30	30
Number of panelists who correctly identified the "odd" sample	15*	18**	11**
Degree of difference perceived	1.3 (slight - moderate)	1.6 (slight - moderate)	1.4 (slight - moderate)
Number of panelists incorrectly identifying the "odd" sample	15	12	19

[1] Of the 30 panelists participating, those panelists who correctly identified the odd sample in both Test 1 and Test 2.

* Statistically significant (95% confidence level)
** Statistically significant (99% confidence level)

Critical value required for significance:	Test 1 & Test 2	(95% confidence) = 15/30 (99% confidence) = 17/30
	Combined	(95% confidence) = 7/30 (99% confidence) = 8/30

95-A-12349 December 28, 1995

TABLE 14
RAINBOW TROUT - PANEL #7
PANELISTS' VERBATIM COMMENTS

0.5% REFINERY EFFLUENT VS. FIELD ATHABASCA RIVER

	Base	Number of Panelists Stating Test 1 (15)	Test 2 (18)
0.5% Refinery Effluent			
- better flavour		1	1
- fresher flavour		-	1
- cleaner fish flavour		1	-
- no fishy flavour		1	-
- no off flavour		1	2
- bland / mild flavour		2	2
- weaker flavour		1	-
- sweeter taste		1	1
- strong fish flavour		-	1
- slight metallic taste		-	1
- slight grassy flavour		-	1
- less sweet tasting		-	1
- more bitter taste		-	2
- moister		1	2
- less moist		-	1
- softer		-	1
- firmer texture		-	1

95-A-12349 December 28, 1995

TABLE 14 (cont'd)
RAINBOW TROUT - PANEL #7
PANELISTS' VERBATIM COMMENTS

0.5% REFINERY EFFLUENT VS. FIELD ATHABASCA RIVER

	Base	Number of Panelists Stating Test 1 (15)	Test 2 (18)
Field Athabasca River			
- slightly / more fishy flavour		3	2
- stronger flavour		1	-
- stronger "fishy" flavour		1	1
- dirty flavour		-	1
- poor flavour		1	-
- sweet taste		1	-
- less bitter taste		1	1
- slightly more oily flavour		1	-
- has an off flavour		-	1
- stronger aftertaste		-	1
- "tinny" aftertaste		1	-
- softer		1	1
- moister		1	1
- drier		1	1
- firmer		-	1

95-A-12349 December 28, 1995

TABLE 15
RAINBOW TROUT

TIME ZERO CONTROL VS. FIELD ATHABASCA RIVER WATER

---

8) Time Zero Control vs. Field Athabasca River Water

(Double) Triangle Difference Test

	Test 1	Test 2	Combined[1]
Total number of panelists participating	30	30	30
Number of panelists who correctly identified the "odd" sample	14[+]	15*	5[+]
Degree of difference perceived	1.4 (slight - moderate)	1.2 (slight - moderate)	1.5 (slight - moderate)
Number of panelists incorrectly identifying the "odd" sample	16	15	25

---

[1] Of the 30 panelists participating, those panelists who correctly identified the odd sample in both Test 1 and Test 2.

[+] Not statistically significant (95% confidence level)
* Statistically significant (95% confidence level)

Critical value required for significance:	Test 1 & Test 2	(95% confidence) = 15/30 (99% confidence) = 17/30
	Combined	(95% confidence) = 7/30 (99% confidence) = 8/30

Test 1 - Actual confidence level achieved = 91%

95-A-12349 December 28, 1995

TABLE 16
RAINBOW TROUT - PANEL #8
PANELISTS' VERBATIM COMMENTS

TIME ZERO CONTROL VS. FIELD ATHABASCA RIVER

		Number of Panelists Stating	
		Test 1	Test 2
	Base	(14)	(15)
Time Zero Control			
- better flavour		-	1
- slightly milder		-	1
- blander flavour		1	1
- weak fish flavour		1	-
- less fishy taste		-	1
- slight fishy flavour		1	-
- sweeter		1	-
- no off flavour		-	-
- stronger fish flavour		-	1
- muddy flavour		1	1
- slightly musty flavour		-	1
- slight metallic flavour		1	1
- slight bitter taste		1	1
- slight phenolic taste		-	1
- moister		2	1
- drier		1	1

95-A-12349 December 28, 1995

TABLE 16 (cont'd)
RAINBOW TROUT - PANEL #8
PANELISTS' VERBATIM COMMENTS

TIME ZERO CONTROL VS. FIELD ATHABASCA RIVER

	Base	Number of Panelists Stating Test 1 (14)	Test 2 (15)
Field Athabasca River			
- better flavour		1	-
- sweeter flavour		2	-
- slightly fresher flavour		1	-
- no off flavour		1	-
- mild fish flavour		1	-
- less oily flavour		1	-
- stronger flavour		2	2
- fishy flavour		1	-
- slight metallic flavour		1	-
- slight bitter taste		1	-
- less sweet		-	1
- off aftertaste		-	1
- drier texture		4	-
- moisture		1	-

95-A-12349 December 28, 1995

TABLE 17
RAINBOW TROUT

OVERALL PREFERENCE RANK - Test 1

	TEST 1									
	0.5% TID Water		Lab Athabasca River Water		Time Zero Control		0.5% Refinery Effluent		Field Athabasca	
Ranked 1st	8	(27%)	5	(17%)	9	(30%)	0	(0%)	8	(27%)
Ranked 2nd	13	(43%)	4	(13%)	3	(10%)	2	(7%)	8	(27%)
Ranked 3rd	1	(3%)	6	(20%)	10	(33%)	10	(33%)	3	(10%)
Ranked 4th	1	(3%)	11	(37%)	5	(17%)	8	(27%)	5	(17%)
Ranked 5th	7	(23%)	4	(13%)	3	(10%)	10	(33%)	6	(20%)
Rank Sum Total[1] (n = 30)	76 a		95 ab		80 a		116 b		83 ab	

[1] Where ranked first = 1, ranked second = 2, ranked third = 3, ranked fourth = 4 and ranked fifth = 5; the lower the Rank Sum Total, the better the score.

Totals followed by the same letter are not statistically significant (95% confidence level).

Critical difference value required between the Rank Sum Totals for statistical significance = 34 (95% confidence level) and = 40 (99% confidence level).

95-A-12349 December 28, 1995

TABLE 17-A
RAINBOW TROUT

OVERALL PREFERENCE RANK - Test 2

	TEST 2									
	0.5% TID Water		Lab Athabasca River Water		Time Zero Control		0.5% Refinery Effluent		Field Athabasca	
Ranked 1st	4	(13%)	3	(10%)	12	(40%)	2	(7%)	9	(30%)
Ranked 2nd	7	(23%)	9	(30%)	5	(17%)	2	(7%)	7	(23%)
Ranked 3rd	4	(13%)	6	(20%)	6	(20%)	9	(30%)	5	(17%)
Ranked 4th	8	(27%)	6	(20%)	3	(10%)	6	(20%)	7	(23%)
Ranked 5th	7	(23%)	6	(20%)	4	(13%)	11	(37%)	2	(7%)
Rank Sum Total[1] (n = 30)	97 ab		93 ab		72 a		112 b		76 a	

[1] Where ranked first = 1, ranked second = 2, ranked third = 3, ranked fourth = 4 and ranked fifth = 5; the lower the Rank Sum Total, the better the score.

Totals followed by the same letter are not statistically significant (95% confidence level).

Critical difference value required between the Rank Sum Totals for statistical significance = 34 (95% confidence level) and = 40 (99% confidence level).

95-A-12349 December 28, 1995

TABLE 18
RAINBOW TROUT - PANEL #9
PANELISTS' VERBATIM COMMENTS

OVERALL PREFERENCE RANK

	Base	Number of Panelists Stating Test 1 (30)	Test 2 (30)
0.5% Tar Island Dyke (TID)			
- good appearance		1	-
- good flavour		1	1
- bland flavour		1	-
- tasteless		-	1
- not enough flavour		2	2
- least strong flavour		1	-
- not fishy		1	-
- mild fish flavour		-	1
- slightly metallic		1	1
- muddy		1	-
- grassy flavour		1	1
- slight off		1	1
- odd flavour		1	-
- bitter		3	-
- sour		1	-
- slight sweet		-	1
- slightly too fishy		1	-
- samples similar		2	2
- good texture		1	1
- moist		2	-
- tacky texture		-	1
- oily and greasy		1	-
- drier		-	2
- slightly dry		-	1

95-A-12349 December 28, 1995

TABLE 18 (cont'd)
RAINBOW TROUT - PANEL #9
PANELISTS' VERBATIM COMMENTS

OVERALL PREFERENCE RANK

	Base	Number of Panelists Stating Test 1 (30)	Test 2 (30)
Lab Athabasca River			
- good flavour		-	1
- best flavour		-	1
- too fishy		-	1
- fishy		2	2
- strongest fish flavour		-	2
- slightly fishy		1	-
- slightly strong in flavour		2	-
- unusual initial flavour		1	-
- less bitter		1	-
- slightly bitter		-	1
- bitter		-	1
- odd flavour		-	1
- off flavour		1	1
- metallic		1	1
- slightly metallic		-	1
- strong grassy flavour		1	1
- stronger aftertaste		-	1
- similar to other samples		2	2
- good texture		-	1
- moist		-	1
- moistest		2	-
- a little too mushy		1	-
- too watery / mushy		1	-
- too moist mushy		-	1
- oily and greasy		1	-

95-A-12349 December 28, 1995

TABLE 18 (cont'd)
RAINBOW TROUT - PANEL #9
PANELISTS' VERBATIM COMMENTS

OVERALL PREFERENCE RANK

	Base	Number of Panelists Stating Test 1 (30)	Test 2 (30)
Time Zero Control			
- good flavour		1	2
- bland flavour		2	3
- milder		-	1
- no / low flavour		2	2
- least strong fish flavour		-	3
- fishy		-	1
- slightly fishy		2	1
- slightly metallic		-	1
- bitter		1	1
- slightly bitter		1	1
- slight off flavour		2	1
- slight muddy flavour		-	2
- similar to other samples		2	2
- good texture		1	1
- moist		1	1
- tender		-	1
- not too wet		-	1
- slightly fatty		1	1

95-A-12349 December 28, 1995

TABLE 18 (cont'd)
RAINBOW TROUT - PANEL #9
PANELISTS' VERBATIM COMMENTS

OVERALL PREFERENCE RANK

	Base	Number of Panelists Stating Test 1 (30)	Test 2 (30)
0.5% Refinery Effluent			
- OK flavour		1	1
- very bland		1	-
- tasteless		-	1
- fishy		3	4
- very fishy		2	1
- unusual initial flavour		1	1
- odd flavour		-	1
- strong fish oil flavour		1	1
- slight off flavour		-	1
- off flavour		-	1
- slightly metallic		1	1
- strong metallic		-	1
- slightly muddy flavour		2	1
- strong grassy flavour		-	1
- bitter off flavour		2	1
- most bitter		-	1
- sour		-	1
- objectionable / awful		-	1
- similar to other samples		1	2
- moist		2	-
- too dry		1	-
- dry		-	3
- tacky texture		-	1

95-A-12349 December 28, 1995

TABLE 18 (cont'd)
RAINBOW TROUT - PANEL #9
PANELISTS' VERBATIM COMMENTS

OVERALL PREFERENCE RANK

	Base	Number of Panelists Stating Test 1 (30)	Test 2 (30)
Field Athabasca River			
- good flavour		1	1
- freshest flavour		1	-
- tastier		1	-
- very bland		1	-
- least fishy		1	-
- tasteless		1	-
- mild fish flavour		-	2
- sweet taste		1	1
- slightly too fishy flavour		1	-
- slightly fishy		-	1
- strong flavour		2	-
- strong muddy flavour		-	1
- distinct grassy flavour		1	1
- metallic flavour		1	1
- slight metallic flavour		1	-
- off flavour		1	-
- slight off flavour		-	1
- objectionable		1	-
- similar to other samples		2	2
- good firm texture		1	-
- moist		2	2
- juicy		-	1
- too watery / mushy		1	1
- oily and greasy		1	-
- too dry		1	-
- least moist		1	-

APPENDIX A

- Questionnaire

- Method sheets

DATE: _____ NAME: _____

PRODUCT: _____ CODES: _____ _____ _____

Instructions:

You have been given 3 coded samples. Two of these are identical and one is different. Choose the one that is different and place its number in the following blank.

1) I believe Sample # _____ is different from the other 2 samples.

2) In what way is this sample different? (Please describe)

3) Please indicate the degree of difference:

slight ( )

moderate ( )

much ( )

very much ( )

extreme ( )

4) Please make as many comments as possible about the samples:

NAME: _____

DATE: _____

RAINBOW TROUT

OVERALL PREFERENCE RANK

Please **RANK** the five samples presented to you in order for **OVERALL PREFERENCE**. Evaluate the samples in the following order:

_____ _____ _____ _____ _____

RANK	**SAMPLE #**
FIRST (most preferred)	_____
SECOND	_____
THIRD	_____
FOURTH	_____
FIFTH (least preferred)	_____

Comments: _____

_____

_____

_____

Thank you!

(DOUBLE) TRIANGLE DIFFERENCE TEST

METHOD

This test is performed in order to determine if there is a perceivable difference between two samples.

Each panelist receives three samples. Two of the samples are identical ("duplicate") and one is different ("odd"). Panelists are asked to choose the "odd" sample and indicate the degree of difference perceived using a five-point scale (slight - moderate - much - very much - extreme).

In this particular study two Triangle Difference Tests were conducted in succession (Double Triangle Difference Test).

For Test 1, each panelist received two "A" samples and one "B" sample. For Test 2, each panelist received the reciprocal: two "B" samples and one "A" sample. Each set had different identification codes.

The samples were randomized as follows:

Test 1 - AAB	Test 2 - BBA
ABA	BAB
BAA	ABB

These sets are distributed at random among the panelists. Samples are identified with 3-digit, randomly chosen numbers from a table of random numbers.

ANALYSIS

Triangle Difference Test

A table of critical values necessary to determine if there was a statistically significant difference perceived was used to analyze the raw sensory data (p=1/3).

The degrees of difference were converted to a numerical score where slight = 1 and extreme = 5, and the average degree of difference was calculated. It should be noted that only the ballots of panelists who correctly identified the "odd" sample were used for calculation of the degree of difference.

ASTM Manual on Sensory Testing Method STP 434 Table 5 (Triangle Difference Test). An extension of the quoted procedures has been made to include degree of difference.

Double Triangle Difference Test

Based on the number of panelists who correctly identify the "odd" sample in both successive tests (Test 1 and Test 2) and with p=1/9, the Z score is calculated to determine if there is a statistically significant perceivable difference between the samples.

$$Z = \frac{(x-1/2) - np}{\sqrt{npq}}$$

(DOUBLE) RANKING FOR OVERALL PREFERENCE

METHOD

The Sensory Taste Panel used a Ranking Test to evaluate the samples for overall preference.

The five samples were ranked for overall preference twice. The first evaluation is called Test 1 and the second evaluation is called Test 2.

The results are reported individually. There was no "Combined" data analysis applicable to this test.

Each sample was coded with a 3-digit randomly chosen number from a table of random numbers.

Presentation of the samples to the panel was randomized to avoid positional bias.

ANALYSIS

The ranking data was analyzed using the Friedman Rank Sum Method of Analysis (Newell, G.J., MacFarlane, J.D. 1987. Expanded Tables for Multiple Comparison Procedures in the Analysis of Ranked Data. Journal of Food Science 52(6): 1721-1275).

APPENDIX B

- Fish Sample Codes

Rainbow Trout - Panel #1
0.5% Tar Island Dyke Water vs. Lab Athabasca River Water

Sample Codes Used

I) <u>0.5% Tar Island Dyke Water</u>
95/11/07
9522345
1) RB Trout - 34
2) RB Trout - 14
3) RB Trout - 9
4) RB Trout - 16
5) RB Trout - 33
6) RB Trout - 12
7) RB Trout - 17
8) RB Trout - 20

II) <u>Lab Athabasca River</u>
TUB #1
95/11/05
1) RB Trout - 16
2) RB Trout - 11
3) RB Trout - 31
4) RB Trout - 28
5) RB Trout - 6
6) RB Trout - 21
7) RB Trout - 2
8) RB Trout - 18
9) RB Trout - 4

Rainbow Trout - Panel #2
0.5% Tar Island Dyke TID Water vs. Time Zero Control

Sample Codes Used

I) <u>0.5% Tar Island Dyke Water</u>
95/11/07
9522345
1) RB Trout - 18
2) RB Trout - 4
3) RB Trout - 10
4) RB Trout - 15
5) RB Trout - 13
6) RB Trout - 27
7) RB Trout - 7
8) RB Trout - 19

II) <u>Time Zero Control</u>
95/10/27
SUN 95 LDW RWTR
1) RB Trout - 13
2) RB Trout - 8
3) RB Trout - 19
4) RB Trout - 3
5) RB Trout - 12
6) RB Trout - 11
7) RB Trout - 26
8) RB Trout - 31

Rainbow Trout - Panel #3
0.5% Refinery Effluent Water vs. Lab Athabasca River Water

Sample Codes Used

I) **0.5% Refinery Effluent Water**

95/11/07
9522345
1) RB Trout - 16
2) RB Trout - 19
3) RB Trout - 10
4) RB Trout - 14
5) RB Trout - 15
6) RB Trout - 26
7) RB Trout - 12
8) RB Trout - 13

II) **Lab Athabasca River Water**

95/11/05
TUB #1
1) RB Trout - 14
2) RB Trout - 12
3) RB Trout - 7
4) RB Trout - 15
5) RB Trout - 10
6) RB Trout - 26
7) RB Trout - 29
8) RB Trout - 1
9) RB Trout - 27
10) RB Trout - 32

Rainbow Trout - Panel #4
0.5% Refinery Effluent Water vs. Time Zero Control

Sample Codes Used

I) **0.5% Refinery Effluent Water**
95/11/07
9522345
1) RB Trout - 22
2) RB Trout - 21
3) RB Trout - 5
4) RB Trout - 17
5) RB Trout - 8
6) RB Trout - 2
7) RB Trout - 25
8) RB Trout - 6

II) **Time Zero Control**
95/10/27
SUN 95 LDW RNTR
1) RB Trout - 5
2) RB Trout - 24
3) RB Trout - 2
4) RB Trout - 23
5) RB Trout - 9
6) RB Trout - 10
7) RB Trout - 4
8) RB Trout - 6

Rainbow Trout - Panel #5
Lab Athabasca River Water vs. Time Zero Control

Sample Codes Used

I) <u>Lab Athabasca River Water</u>
95/11/05
TUB #1
1) RB Trout - 19
2) RB Trout - 3
3) RB Trout - 13
4) RB Trout - 22
5) RB Trout - 20
6) RB Trout - 5
7) RB Trout - 24
8) RB Trout - 17
9) RB Trout - 37

II) <u>Time Zero Control</u>
95/10/27
SUN 95 LDW RWTR
1) RB Trout - 16
2) RB Trout - 21
3) RB Trout - 41
4) RB Trout - 18
5) RB Trout - 32
6) RB Trout - 7
7) RB Trout - 25
8) RB Trout - 17

Rainbow Trout - Panel #6
0.5% Tar Island Dyke Water vs. Field Athabasca River Water

Sample Codes Used

I) **0.5% Tar Island Dyke Water**
95/11/07
9522345
1) RB Trout - 2
2) RB Trout - 5
3) RB Trout - 8
4) RB Trout - 29
5) RB Trout - 23
6) RB Trout - 25
7) RB Trout - 1
8) RB Trout - 3

II) **Field Athabasca River Water**
Golder Associates
Oct 17 1995
Project # 952-2345
Richard Seraphin
Caged Fish Sample
Athabasca River
SUN 95 FAR CRNTR
1) RB Trout - 27
2) RB Trout - 30
3) RB Trout - 32
4) RB Trout - 29
5) RB Trout - 12
6) RB Trout - 11
7) RB Trout - 28
8) RB Trout - 20
9) RB Trout - 31

Rainbow Trout - Panel #7
0.5% Refinery Effluent Water vs. Field Athabasca River Water

Sample Codes Used

I) **0.5% Refinery Effluent Water**
95/11/07
9522345
1) RB Trout - 23
2) RB Trout - 1
3) RB Trout - 4
4) RB Trout - 20
5) RB Trout - 3
6) RB Trout - 7
7) RB Trout - 11
8) RB Trout - 24

II) **Field Athabasca River Water**
Golder Associates
Oct 17 1995
Project # 952-2345
Richard Seraphin
Caged Fish Sample
Athabasca River
SUN 95 FAR CRNTR
1) RB Trout - 6
2) RB Trout - 38
3) RB Trout - 18
4) RB Trout - 35
5) RB Trout - 16
6) RB Trout - 26
7) RB Trout - 36
8) RB Trout - 13
9) RB Trout - 15

Rainbow Trout - Panel #8
Time Zero Control vs. Field Athabasca River Water

Sample Codes Used

I) **Time Zero Control**
95/10/27
SUN 95 LDW RNTR
1) RB Trout - 34
2) RB Trout - 22
3) RB Trout - 27
4) RB Trout - 29
5) RB Trout - 35
6) RB Trout - 15
7) RB Trout - 1

II) **Field Athabasca River Water**
Golder Associates
Oct 17 1995
Project # 952-2345
Richard Seraphin
Caged Fish Sample
Athabasca River
SUN 95 FAR CRNTR
1) RB Trout - 22
2) RB Trout - 37
3) RB Trout - 14
4) RB Trout - 17
5) RB Trout - 23
6) RB Trout - 4
7) RB Trout - 19
8) RB Trout - 24
9) RB Trout - 25

Rainbow Trout - Panel #9
Overall Preference Rank Test

Sample Codes Used

I) **0.5% Tar Island Dyke Water**
95/11/07
9522345
1) RB Trout - 21
2) RB Trout - 28
3) RB Trout - 22
4) RB Trout - 26
5) RB Trout - 6
6) RB Trout - 30
7) RB Trout - 24
8) RB Trout - 32
9) RB Trout - 11
10) RB Trout - 35

II) **Lab Athabasca River Water**
95/11/05
TUB #1
1) RB Trout - 9
2) RB Trout - 23
3) RB Trout - 36
4) RB Trout - 25
5) RB Trout - 40
6) RB Trout - 33
7) RB Trout - 35
8) RB Trout - 8
9) RB Trout - 39

III) **Time Zero Control**
95/10/27
SUN 95 LDW RNTR
1) RB Trout - 33
2) RB Trout - 28
3) RB Trout - 30
4) RB Trout - 20

Rainbow Trout - Panel #9 (cont'd)
Overall Preference Rank Test

Sample Codes Used

IV) **<u>0.5% Refinery Effluent Water</u>**

95/11/07
9522345
1) RB Trout - 28
2) RB Trout - 27
3) RB Trout - 18
4) RB Trout - 9
5) RB Trout - 29
6) RB Trout - 32
7) RB Trout - 30
8) RB Trout - 33
9) RB Trout - 31

V) **<u>Field Athabasca River Water</u>**

Golder Associates
Oct 17 1995
Project # 952-2345
Richard Seraphin
Caged Fish Sample
Athabasca River
SUN 95 FAR CRNTR
1) RB Trout - 1
2) RB Trout - 2
3) RB Trout - 3
4) RB Trout - 10
5) RB Trout - 9
6) RB Trout - 8
7) RB Trout - 5
8) RB Trout - 7
9) RB Trout - 33