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Northern River Basins Study

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by Gordon Court D. A. Westworth and Associates Ltd.

## NORTHERN RIVER BASINS STUDY PROJECT REPORT NO. 4 COLLECTION OF YOUNG-OF-THE-YEAR MERGANSERS WAPITI AND ATHABASCA RIVERS AUGUST, 1992

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### **PREFACE:**

The Northern River Basins Study was initiated through the "Canada-Alberta-Northwest Territories Agreement Respecting the Peace-Athabasca-Slave River Basin Study, Phase II - Technical Studies" which was signed September 27, 1991. The purpose of the Study is to understand and characterize the cumulative effects of development on the water and aquatic environment of the Study Area by coordinating with existing programs and undertaking appropriate new technical studies.

This publication reports the method and findings of particular work conducted as part of the Northern River Basins Study. As such, the work was governed by a specific terms of reference and is expected to contribute information about the Study Area within the context of the overall study as described by the Study Final Report. This report has been reviewed by the Study Science Advisory Committee in regards to scientific content and has been approved by the Study Board of Directors for public release.

It is explicit in the objectives of the Study to report the results of technical work regularly to the public. This objective is served by distributing project reports to an extensive network of libraries, agencies, organizations and interested individuals and by granting universal permission to reproduce the material.

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	(Peter Melnychuk, Co-Chair) $\frac{29/3/93}{(Date)}$

### COLLECTION OF YOUNG-OF-THE-YEAR MERGANSERS WAPITI AND ATHABASCA RIVERS AUGUST, 1992

### STUDY PERSPECTIVE

Environmental toxicologists now recognize the value of waterbirds, particularly piscivorous (fish eating) for useful "monitors" forms. as measuring environmental contamination. Since fish-eating birds and man share a common food source, any pollution that might affect water birds has the potential to directly affect the welfare of local human populations.

### **Related Study Questions**

11) Have the riparian vegetation, riparian wildlife and domestic livestock in the river basins been affected by exposure to organochlorines or other toxic compounds?

Within the Northern River Basins Study area, the Common Merganser appeared to be a desirable waterbird species for monitoring contaminant movement through the food chain and accumulation. Their young feed almost exclusively on aquatic foods with minimal influence from external basin sources of contamination. Contaminants of particular interest to this study are predominately associated with pulp mill effluent. This report describes the efforts to collect young-ofthe-year common mergansers from above and below three pulp and paper mill sites within the Wapiti and Athabasca River systems. The low merganser population failed to provide sufficient samples to satisfy the original study plans, and therefore, the survey was prematurely terminated to avoid unnecessary expense. Sufficient bird specimens were collected from the Weyerhaeuser Canada pulp and paper mill site on the Wapiti River and were submitted for contaminant analysis.

Future sampling will be dependent on the analytical findings. In the meantime, the Canadian Wildlife Service continues to investigate the utility of sampling bald eagles and other wildlife. Waterfowl from the Peace-Athabasca Delta are to be collected in the fall of 1993 as part of a national contaminant survey. Results of that investigation combined with the work done on mergansers and mink under the Northern River Basins Study will provide future investigators with better direction on what species to collect and when.

## Acknowledgements

The author gratefully acknowledges the assistance of several people during the course of the study. Special thanks are extended to Mr. M. Wayland, Canadian Wildlife Service, Saskatoon, Saskatchewan for his assistance in the field and his editorial comments. The capable assistance of Mr. P. Hvenegaard in jetboat operation and field logistics is also acknowledged.

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# INTRODUCTION

In recent years, there has been increasing interest in the development of forests in Alberta, Saskatchewan, and Manitoba for pulp and paper. Pulp mills now operate along most rivers in northern Alberta and more are planned to begin operation within the next decade. This development will have wide-ranging effects on wildlife not only in the boreal forest, but within drainages downstream of pulp mill effluents. Of particular concern is the potential for contamination of river systems by potentially-harmful environmental pollutants such as dioxins. In Canada, there exists a consensus among the general public that levels of these chemicals should be monitored within ecosystems.

Over the last few decades toxicologists have come to recognize the value of waterbirds, particularly piscivorous forms, as vehicles through which pollutants in the environment may be monitored (Mineau et al. 1984, Fox and Weseloh 1987). Some fish-eating species are capable of bio-accumulating lipid-soluble pollutants in their body tissues to levels millions of times that of the ambient concentration in local water bodies. For example, individual Herring Gulls (*Larus argentatus*) from the Great Lakes bio-concentrated levels of polychlorinated bi-phenyls (PCBs) and DDE (the primary metabolite of DDT) in their eggs to 25,000,000 times that in lake water and over 20 times that of the average levels in local fish (Norstrom *et al.* 1978). Since fish-eating birds and man share a common food source, any pollution that might affect water birds has the potential to directly affect the welfare of local human populations.

Within the Northern River Basins Study Area, the Common Merganser (*Mergus merganser*) appears to represent an ideal species through which to monitor potential pollutants that might originate from the pulp and paper industry. The Canadian Wildlife Service, in cooperation with D.A. Westworth and Associates Ltd., evaluated the suitability of this species for such work by conducting a census and collection of young-of-the-year mergansers near three mill sites on two drainages in northern Alberta. The following report summarizes the success of these collec-

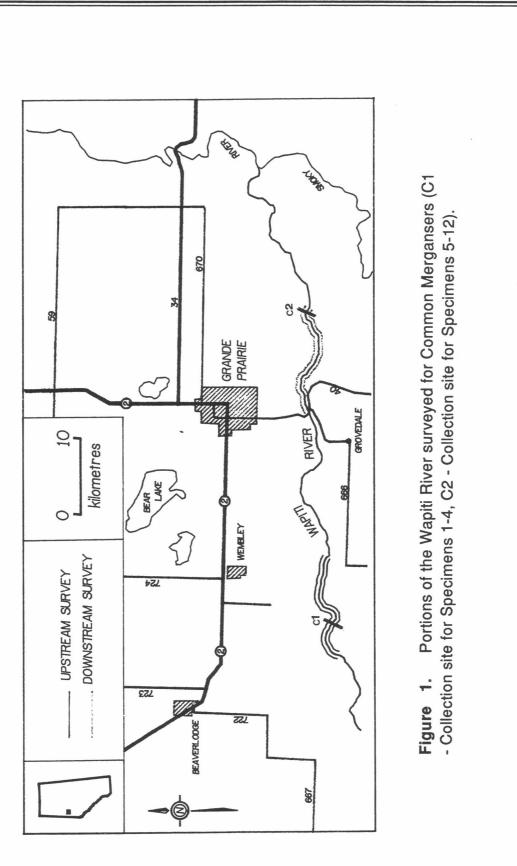
tions and details the range of samples taken from birds collected for contaminant analyses.

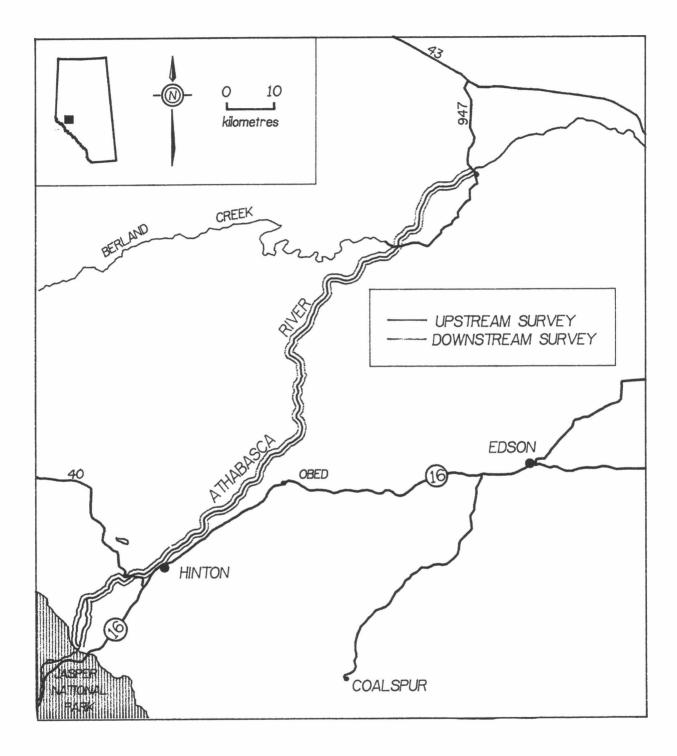
# METHODS

A field program, designed to survey and collect mergansers over 6 stretches of river (upstream and downstream of 3 mill sites) using a jetboat, was conducted between August 4 and August 12, 1992. Site 1, which included stretches of the Wapiti River, was comprised of an upstream section extending 15.5 km upstream of Pipestone Creek, and a downstream section 15.6 km downstream of the Grovedale Bridge (Highway 40), near Grande Prairie (Figure 1). Site 2 was located on the upper Athabasca River with an upstream section extending 33.5 km upstream of the Hinton town site to Brule Lake, and a downstream section 121.1 km downstream of Hinton to the Highway 947 Bridge (Figure 2). Site 3 was located on the lower Athabasca River. The upstream section consisted of 59.4 km of river upstream of the Athabasca town site, and a 138.6 km section downstream of Athabasca (Figure 3).

Each section of river was surveyed for mergansers twice, once on the outward journey and once on the homeward journey. The noise and speed of the jetboat was more than adequate to flush waterfowl from the rivers, although all side channels were scanned with field glasses and in some cases 'ground truthed' to check for birds. Broods of mergansers (flightless during the survey period) were easily recognized as they flushed at distances of up to 500 metres in front of the jetboat. In most cases, the speed of the jetboat was used to run up on broods to allow collection. The location (latitude and longitude) of the collection site was recorded for each bird.

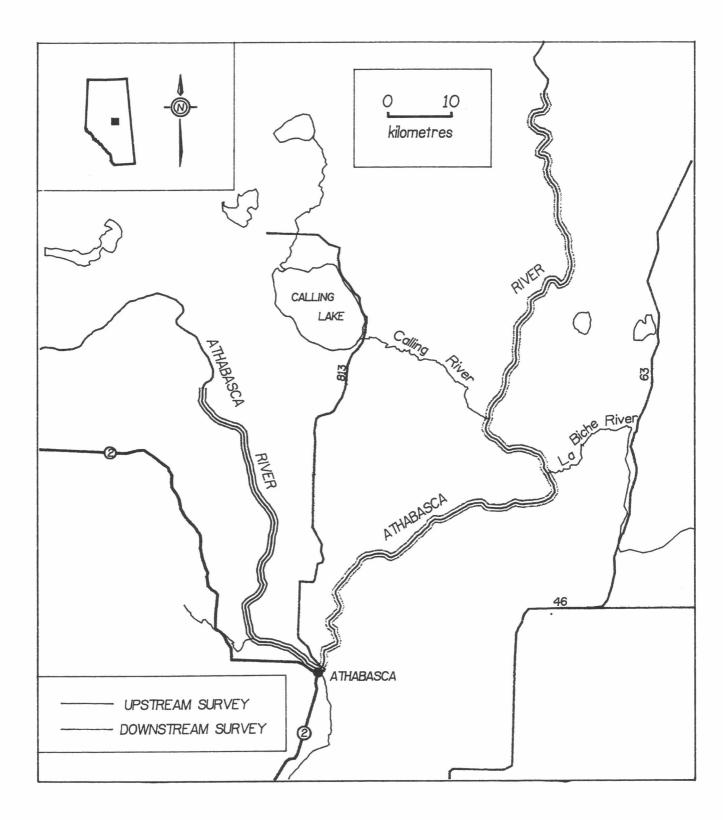
Young mergansers were shot with 12 gauge shotguns and were dissected and processed for contaminant analyses immediately after collection. Field preparation of specimens and tissues followed the methodology given in Appendix II (Trudeau 1992). Seventeen different samples were taken from each bird for analyses that will include mixed-function oxidase (MFO) induction, organochlorine residue determination (including PCBs, pesticides, and dioxins), heavy metal residue analysis, and vitamin A, porphyrin, stable isotope, and DNA integrity analyses. Fresh weights of selected tissues were taken upon dissection (Appendix I). Each bird was sexed





**Figure 2.** Portions of the Upper Athabasca River surveyed for Common Mergansers.

### COLLECTION OF YOUNG-OF-THE-YEAR MERGANSERS Northern River Basins Study



**Figure 3.** Portions of the Upper Athabasca River surveyed for Common Mergansers.

urements, including body weight, bill depth, bill length, wing length, 9th primary length, and tarsal length were recorded for each bird (Appendix I). Samples for MFO analyses, which were frozen in liquid nitrogen, were transported to the Canadian Wildlife Service, National Wildlife Research Centre (NWRC), Hull, Quebec, for analysis. Other samples including kidneys, portions of the liver, and carcasses were placed on ice in a cooler in the boat immediately following dissection, transported to a local freezer each evening, and shipped in a cooler filled with dry ice to Edmonton where they were placed in Alberta Fish and Wildlife Division freezers. From there, the samples were packed in dry ice and shipped in coolers to the Canadian Wildlife Service, Saskatoon, Saskatchewan by air. From Saskatoon, liver and kidney samples will be shipped on dry ice to the NWRC where they will be homogenized and stored until such time as they are requested by the Northern River Basins Study Office.

# **RESULTS AND DISCUSSION**

## Wapiti River

A single brood of seven merganser young was encountered on the upstream portion of the Wapiti River. Apart from the single adult attending this brood, no other mergansers were seen on this section of the river (C1, Figure 1). Four of the seven young were collected. A complete list of samples and morphometric measurements from each bird appears in Appendix I (Specimens 1-4).

A large 'gang' brood of approximately 30 young-of-the-year and molting adult mergansers was encountered on the downstream portion of the Wapiti River (C2, Figure 1). Although this group was located only 5.6 km downstream of the Procter and Gamble pulp mill effluent, 8 individuals (6 young and 2 adults) were collected from this group. It appeared prudent to sample these birds as they were in a 'heavy effluent' area located upstream of the Smoky and Wapiti River confluence (ie. not birds from that portion of the Wapiti drainage where effluent would be diluted or otherwise altered by water from another drainage). This site is also in the same general area as the SENTAR fish sampling sites, so residues in local mergansers should be of comparative value for these studies. A complete list of samples and morphometric measurements from each bird, both adults and juveniles, appears in Appendix I (Specimens 5-12). Overall, the Common Merganser appears to be a good species for monitoring pollution in this drainage.

## **Upper Athabasca River**

No merganser broods were encountered on either upstream or downstream portions of the Upper Athabasca River survey area (Figure 2). Only 5 adults were seen during this part of the survey and all were along stretches downstream of the Hinton town site. High turbidity and low productivity of this glacially-fed drainage may account for the low numbers of waterfowl in the area. From these results, particularly in light of the large segments of river surveyed, it appears that the Common Merganser is not a suitable species for monitoring pollution levels in this portion of the Athabasca drainage.

## Lower Athabasca River

No merganser broods were encountered on either upstream or downstream portions of the Lower Athabasca River survey area (Figure 3). Only one adult was seen during this part of the survey and it was observed in the stretch upstream of the Athabasca town site. Like the Upper Athabasca section, waterfowl production in this area appears to be poor. Based on these results, it appears that the Common Merganser is not a suitable species for monitoring pollution in this portion of the Athabasca drainage.

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**Appendix I.** Specimen Collection Sheets for Common Mergansers Collected on the Wapiti River, Alberta, August 5 and 6, 1992.

Date: 5/8/92	Location: Wapiti River
Latitude: <u>55°02</u> 1	ongitude: <u>119°09</u> Habitat: <u>Main Stream</u>
No. Collected: _/	Time of Collection: 1358 Method: Shot
Collector: <u>M. Waylard</u>	comments: Upstream of Pipestone Creek
Species: Ol <u>Common</u> Merganser (Juvenile)	Sex: Se
	from a single brood of 7 young.

Date: <u>5/8/92</u>	Location: Wapiti River
Latitude: <u>55°02'</u> Lo	ongitude: 119°09' Habitat: <u>Main Stream</u>
No. Collected: 2	Time of Collection: 1358 Method:
Collector: <u>M. Wayland</u>	comments: <u>Upstream of Pipestone Creek</u>
Specimen No. <u>OZ</u> <u>Common</u> Merganser (Juvenile)	Sex: ? Weight: 950g Bill Depth: 14.7 mm Bill Length: 43.6 mm Wing Length: 134 mm Tarsal Length: 62.0 mm 9th Primary: 38 mm Bile Collection (Y/N): N Whole Liver Weight: 31.49 Left lobe wt. 13.09 Right Lobe Wt. 18.49 MFO (20 ml vial): ✓ Metals (2 ml cryovial) ✓ Weight: 0.49 Vitamin A (2 ml cryovial) ✓ Weight: 0.49 DNA Integrity (2 ml cryovial) ✓ Weight: 0.49 Reserve 1 (2 ml cryovial) ✓ Weight: 0.49 Reserve 2 (2 ml cryovial) ✓ Weight: 0.49 Reserve 3 (2 ml cryovial) ✓ Weight: 0.49 OC/PCB/Dioxins: ✓ Weight: 0.49 Time MFO into Liquid Nitro: 1448 Kidney left Wt: 4.29 Keight: 1.49 Metal Analysis: ✓ Weight: 0.49 Metal Analysis: ✓ Weight: 0.40 Metal Ana

Date: 5/8/92	Location: Wapiti River
Latitude: 55°02' L	ongitude: <u>119°08'</u> Habitat: <u>Main Stream</u>
No. Collected: 3	Time of Collection: 1740 Method: Shot
Collector: <u>M. Wayland</u>	comments: <u>Upstream of Pipestone</u> Creek
Specimen No. <u>03</u> Species: <u>Common</u> merganser (Juvenile)	Sex: $\underline{M}$ Weight: $\underline{990g}$ Bill Depth: $\underline{15.5mm}$ Bill Length: $\underline{44.0mm}$ Wing Length: $\underline{143mm}$ Tarsal Length: $\underline{63.3mm}$ 9th Primary: $\underline{43mm}$ Bile Collection (Y/N): $\underline{N}$ Whole Liver Weight: $\underline{26.4g}$ Left lobe wt. $\underline{12.5g}$ Right Lobe Wt. $\underline{13.9g}$ MFO (20 ml vial): $\checkmark$ Metals (2 ml cryovial) $\checkmark$ Weight: $\underline{0.6g}$ Vitamin A (2 ml cryovial) $\checkmark$ Weight: $\underline{0.6g}$ DNA Integrity (2 ml cryovial) $\checkmark$ Weight: $\underline{0.6g}$ Reserve 1 (2 ml cryovial) $\checkmark$ Weight: $\underline{0.6g}$ Reserve 2 (2 ml cryovial) $\checkmark$ Weight: $\underline{0.6g}$ Reserve 3 (2 ml cryovial) $\checkmark$ Weight: $\underline{0.6g}$ Reserve 4 (2 ml cryovial) $\checkmark$ Weight: $\underline{0.6g}$ Reserve 3 (2 ml cryovial) $\checkmark$ Weight: $\underline{0.6g}$ Reserve 4 (2 ml cryovial) $\checkmark$ Weight: $\underline{0.6g}$ Reserve 3 (2 ml cryovial) $\checkmark$ Weight: $\underline{0.6g}$ Reserve 4 (2 ml cryovial) $\checkmark$ Weight: $\underline{0.6g}$ Reserve 3 (2 ml cryovial) $\checkmark$ Weight: $\underline{0.6g}$ Reserve 4 (2 ml cryovial) $\checkmark$ Weight: $\underline{0.6g}$ Notallothionein: $\checkmark$ Weight: $\underline{0.6g}$ Metallothionein: $\checkmark$ Weight: $\underline{1.0g}$ Metallothionein: $\checkmark$ Weight: $\underline{1.0g}$ Metallothionein: $\checkmark$ Weight: $\underline{1.0g}$ Metal Analysis: $\checkmark$ Tibia: $\underline{N}$ Heart: $\underline{N}$ Spleen: $\underline{N}$ Adrenals/Gonads: $\underline{N}$ Thyroids/Parathyroids: $\underline{N}$ Lung: $\underline{N}$ Thymus: $\underline{N}$ Brain: $\underline{N}$ Breast Muscle for Stable Isotopes (freeze) $\underline{\vee}$ Blood on slide for hematozoa: $\underline{\vee}$
	Carcass Saved $(Y/N) \longrightarrow Y$

### COLLECTION OF YOUNG-OF-THE-YEAR MERGANSERS Northern River Basins Study

#### MERGANSER COLLECTIONS

Date: 5/8/92	Location: Wapiti River
Latitude: <u>55°02'</u> Lo	ongitude: 119°08' Habitat: Main Stream
No. Collected: 4	Time of Collection: 1741 Method: Shot
Collector: <u>M. Wayland</u>	comments: <u>Upstream of Pipestone</u> Creek
Specimen No. <u>04</u> Species: <u>Commu</u> Merganser (Juvenile)	Sex: $\underline{M}$ Weight: $\underline{950}$ Bill Depth: $\underline{14.1mm}$ Bill Length: $\underline{43.0 \text{ mm}}$ Wing Length: $\underline{140 \text{ mm}}$ Tarsal Length: $\underline{61.1 \text{ mm}}$ 9th Primary: $\underline{43 \text{ mm}}$ Bile Collection (Y/N): $\underline{N}$ Whole Liver Weight: $\underline{37.69}$ Left lobe wt. $\underline{11.89}$ Right Lobe Wt. $\underline{15.89}$ MFO (20 ml vial): $\underline{\checkmark}$ Metals (2 ml cryovial) $\underline{\checkmark}$ Weight: $\underline{0.69}$ Vitamin A (2 ml cryovial) $\underline{\checkmark}$ Weight: $\underline{1.09}$ DNA Integrity (2 ml cryovial) $\underline{\checkmark}$ Weight: $\underline{1.09}$ Reserve 1 (2 ml cryovial) $\underline{\checkmark}$ Weight: $\underline{0.69}$ Reserve 2 (2 ml cryovial) $\underline{\checkmark}$ Weight: $\underline{0.69}$ Reserve 3 (2 ml cryovial) $\underline{\checkmark}$ Weight: $\underline{0.69}$ Reserve 4 (2 ml cryovial) $\underline{\checkmark}$ Weight: $\underline{0.69}$ Reserve 3 (2 ml cryovial) $\underline{\checkmark}$ Weight: $\underline{0.69}$ Reserve 4 (2 ml cryovial) $\underline{\checkmark}$ Weight: $\underline{0.69}$ Reserve 4 (2 ml cryovial) $\underline{\checkmark}$ Weight: $\underline{0.69}$ Kidney left Wt: $\underline{4.99}$ Kidney right Wt: $\underline{5.49}$ Metalothionein: $\underline{\checkmark}$ Weight: $\underline{1.32}$ Reserve: $\underline{\checkmark}$ Weight: $\underline{1.32}$ Metal Analysis: $\underline{\checkmark}$ Tibia: $\underline{N}$ Heart: $\underline{N}$ Spleen: $\underline{N}$ Adrenals/Gonads: $\underline{N}$ Thyroids/Parathyroids: $\underline{N}$ Lung: $\underline{N}$ Thymus: $\underline{N}$ Brain: $\underline{N}$ Breast Muscle for Stable Isotopes (freeze) $\underline{Y}$ Blood on slide for hematozoa: $\underline{\curlyvee}$ Carcass Saved (Y/N) $\underline{\curlyvee}$

Date: 5/8/92		Location:	Wapiti' 1	River
Latitude: <u>55°05'</u> Lo	ngitude: <u>118°35'</u>	Habitat:	ide Chanr	nel
No. Collected: <u>5</u>	Time of Collection: _	1150 Me	thod: _5	not
Collector: <u>M. Wayland</u> Crambel efflu	Comments: <u>Downstrea</u>	am of Proc	ter and	
<u>Specimen No.</u> <u>O5</u> <u>Common</u> Merganser (Juvenile)	Sex: <u>M</u> Weight: Bill Length: <u>48.2 m</u> Tarsal Length: <u>58.9 m</u> Bile Collection (Y/N): Whole Liver Weight: Right Lobe Wt. <u>20.2 M</u> Metals (2 ml cryovial) Vitamin A (2 ml cryovi Porphyrin (2 ml cryovi Reserve 1 (2 ml cryovi Reserve 2 (2 ml cryovi Reserve 3 (2 ml cryovi Reserve 4 (2 ml cryovi OC/PCB/Dioxins: <u>Time MFO into Liquid 1</u> Kidney left Wt: <u>4.5</u> Mettalothionein: <u>V</u> Porphyrin: <u>V</u> Reserve: <u>V</u> Metal Analysis: <u>V</u> Breast Muscle for Stal Gut Sample (Freeze): Blood on slide for he Carcass Saved (Y/N) N.B. Specimens from a gang bro individuals. All W 5.6 km downstree	m Wing Leng 9th Prima <u>35.3 g</u> Lef g MFO (2 ) Wei ial) Wei ial) Wei ial) Weight: ial) Weight: Nitro: 12 G Kidney r Weight: Wei	th: 160 ry: 53 t lobe wt. 0 ml vial): ght: 0.99 ght: 1.10 Weight: Weight: Weight: Weight: Weight: Weight: Weight: Weight: Weight: Spleen: Parathyroid Brain: freeze) Y Vere colle uf 30 d about	$\frac{mm}{mm}$ $\frac{15 \cdot l_{q}}{V} (14 \cdot l_{q})$ $\frac{10 \cdot l_{q}}{10 \cdot l_{q}}$
-	and Grambel eff	- luent.		

Date: 6/8	192			Locati	on: _V	lapiti	River
Latitude: _	55°05' L	ongitude:	118° 35'	Habitat:	Side	cha	nnel
No. Collected	d: <u>6</u>	Time of	Collection:	1150	Metho	d: _5	ihot
Collector:	<u>m.Wayland</u> Gambel	Comments: effluent	Downst	ream of	Procter	and	
Specimen No. Species:	<u>Common</u> Merganser (Adu It)	Bill Len Tarsal L Bile Col Whole Li Right Lo Metals ( Vitamin Porphyri DNA Inte Reserve Reserve Reserve Reserve Reserve CC/PCB/D Time MFO Kidney 1 Mettalot Porphyri Reserve: Metal An Tibia: Adrenals Lung: Breast M Gut Samp	Alysis: N /Gonads:	Image: Second structure       Wing         Image: Second structure       9th I         Image: Second structure       Ming         Image: Second structure       Ming      <	Length: Primary: Left 1 FO (20 m Weight Weight Weight Weight: 125: hey righ ght: Sids/Para N Des (fre	- <u>147</u> - <u>11</u> obe wt. 1 vial) : <u>1.3c</u> eight: eight: eight: eight: eight: <u>7.59</u> <u>1.39</u> <u>0.99</u> <u>1.39</u> pleen: thyroid Brain: eze) Y	$\frac{14.7 \text{ g}}{14.7 \text{ g}} 2 \text{ g})$ $\frac{14.7 \text{ g}}{0.7 \text{ g}} 2 \text{ g})$ $\frac{9}{0.7 \text{ g}}$ $\frac{0.7 \text{ g}}{0.9 \text{ g}}$ $\frac{0.9 \text{ g}}{0.6 \text{ g}}$ $\frac{0.6 \text{ g}}{0.6 \text{ g}}$ $\frac{3.9 \text{ g}}{0.6 \text{ g}}$ $\frac{3.9 \text{ g}}{100000000000000000000000000000000000$

Location: Wapit, River
Longitude: 118° 35' Habitat: Side Channel
Time of Collection: 1150 Method: Shot
comments: <u>Downstream</u> of Procter and effluent.
Sex: $\underline{M}$ Weight: $\underline{12759}$ Bill Depth: $\underline{16.3mm}$ Bill Length: $\underline{49.4 mm}$ Wing Length: $\underline{190 mm}$ Tarsal Length: $\underline{62.3 mm}$ 9th Primary: $\underline{180 mm}$ Bile Collection (Y/N): $\underline{N}$ Whole Liver Weight: $\underline{50.1a}$ Left lobe wt. $\underline{23.69}$ Right Lobe Wt. $\underline{26.5a}$ MFO (20 ml vial): $\underline{\sim} (\sqrt{8.7g})$ Metals (2 ml cryovial) $\underline{\sim}$ Weight: $\underline{1.6g}$ Vitamin A (2 ml cryovial) $\underline{\sim}$ Weight: $\underline{1.7g}$ DNA Integrity (2 ml cryovial) $\underline{\sim}$ Weight: $\underline{1.7g}$ Reserve 1 (2 ml cryovial) $\underline{\sim}$ Weight: $\underline{1.7g}$ Reserve 2 (2 ml cryovial) $\underline{\sim}$ Weight: $\underline{0.7g}$ Reserve 3 (2 ml cryovial) $\underline{\sim}$ Weight: $\underline{0.7g}$ Time MFO into Liquid Nitro: $\underline{1303}$ Kidney left Wt: $\underline{5.5g}$ Kidney right Wt: $\underline{5.2g}$ Metal Analysis: $\underline{\sim}$ Weight: $\underline{1.4g}$ Reserve: $\underline{\sim}$ Weight: $\underline{1.4g}$ Reserve: $\underline{\sim}$ Weight: $\underline{1.4g}$ Reserve: $\underline{\sim}$ Weight: $\underline{1.4g}$ Metal Analysis: $\underline{\sim}$ Weight: $\underline{1.4g}$ Metal Kidney left Wt: $\underline{5.5g}$ Kidney right Wt: $\underline{5.2g}$ Metal Analysis: $\underline{\sim}$ Weight: $\underline{1.4g}$ Reserve: $\underline{\sim}$ Weight: $\underline{1.4g}$ Metal Analysis: $\underline{\sim}$ Weight: $\underline{1.4g}$ Metal Analysis: $\underline{\sim}$ Metal for Spleen: $\underline{N}$ Adrenals/Gonads: $\underline{N}$ Thyroids/Parathyroids: $\underline{N}$ Lung: $\underline{N}$ Thymus: $\underline{N}$ Brain: $\underline{N}$ Breast Muscle for Stable Isotopes (freeze) $\underline{\vee}$ Blood on slide for hematozoa: $\underline{\vee}$
Carcass Saved $(Y/N) $

Date: 6/8/92			Location:	Wapiti	River
Latitude: <u>55°</u> C	5' Longitude:	118°35'	Habitat: <u>Si</u>	de chann	rel
No. Collected: _	Time of C	Collection:	1150 Me	ethod:	Shot
	ayland comments: nbel effluent	Downstr	eam of Pro	octer and	(
Specimen No. O Species: Com Mer	8 Sex: Bill Leng ganser Tarsal Leng ganser Tarsal Leng Whole Liv Right Lot Metals (2 Vitamin A Porphyrin DNA Integ Reserve Reser	gth:       43.9 m         ength:       56.1 m         ength:       56.1 m         lection (Y/N)       yer Weight:         be Wt.       20.6         2 ml cryovial       4 (2 ml cryovial         A (2 ml cryovial       4 (2 ml cryovial         3 (2 ml cryovial       4 (2 ml cryovial         4 (2 ml cryovial       4 (2 ml cryovial         into Liquid       eft Wt:       5.3         hionein:          n:              A (2 ml cryovial          into Liquid       eft Wt:       5.3         hionein:	<u>32.19</u> Let <u>g</u> MFO (2 vial) <u>v</u> Wei vial) <u>v</u> Wei vial) <u>v</u> Wei vial) <u>v</u> vial) <u>v</u> vial) <u>v</u> vial) <u>v</u> vial) <u>v</u> vial) <u>v</u> weight: <u>v</u> <u>v</u> Weight: <u>v</u> Weight: <u>v</u> <u>v</u> Weight: <u>v</u> <u>v</u> Weight: <u>v</u> <u>v</u> Weight: <u>v</u> <u>v</u> <u>v</u> <u>v</u> <u>v</u> <u>v</u> <u>v</u> <u>v</u>	th: <u>169</u> ary: <u>71</u> to lobe wt. 20 ml vial) ight: <u>0.76</u> ight: <u>1.65</u> Weight: Weight: Weight: Weight: Weight: Weight: <u>1317</u> right Wt: <u>1.39</u> <u>1.139</u> <u>1.29</u> Spleen: Parathyroio Brain:	$\frac{Mm}{mm}$ $\frac{11.5g}{(16.0g)}$ $\frac{11.5g}{(16.0g)}$ $\frac{9}{9}$ $\frac{100}{9}$
	Carcass	Saved (Y/N)	<u> </u>		

Latitude: <u>55°05'</u> Longitude: <u>118°35'</u> Habitat: <u>Side Channel</u> No. Collected: <u>9</u> Time of Collection: <u>1150</u> Method: <u>Shot</u> <u>Collector: M.Wayland</u> comments: <u>Downshream of Procter and</u> <u>Gambel effluent</u> Specimen No. <u>09</u> Sex: <u>M</u> Weight: <u>12000</u> Bill Depth: <u>15.0mm</u> Bill Length: <u>42.1 mm</u> Wing Length: <u>156 mm</u> Merganser (Juvenile) Bile Collection (Y/N): <u>N</u> Whole Liver Weight: <u>34.29</u> Left lobe wt. <u>16.80</u> Right Lobe Wt. <u>17.49</u> MFO (20 ml vial): <u>V(12.00</u> Metals (2 ml cryovial) <u>V</u> Weight: <u>0.69</u> Porphyrin (2 ml cryovial) <u>V</u> Weight: <u>0.63</u> Porphyrin (2 ml cryovial) <u>V</u> Weight: <u>1.29</u> Reserve 2 (2 ml cryovial) <u>V</u> Weight: <u>1.29</u> Reserve 3 (2 ml cryovial) <u>V</u> Weight: <u>1.29</u> Reserve 4 (2 ml cryovial) <u>V</u> Weight: <u>1.29</u> Reserve 4 (2 ml cryovial) <u>V</u> Weight: <u>1.29</u> Reserve 4 (2 ml cryovial) <u>V</u> Weight: <u>1.29</u> Reserve 3 (2 ml cryovial) <u>V</u> Weight: <u>1.29</u> Reserve 4 (2 ml cryovial) <u>V</u> Weight: <u>1.29</u> Reserve: <u>V</u> Weight: <u>1.29</u> Metal Analysis: <u>V</u> Weight: <u>1.29</u>	Date: 6/8/92		Location:	Wapiti	River
Collector:       M:Wayland       Comments:       Dwnstream       of       Procter       and         Gambel       effluent         Specimen No.       O9       Sex:       M       Weight:       17000       Bill Depth:       150mm         Species:       Common       Bill Length:       42.1 mm       Wing Length:       156 mm         Merganser       Tarsal Length:       59.7 mm       9th Primary:       51.0 mm         Minde Liver Weight:	Latitude: <u>55°05′</u> Lon	gitude: <u>118°35'</u>	Habitat:	ide Chan	inel
Grambel effluent         Specimen No.       OP         Species:       Sex:       M         Weight:       1200g         Bill Length:       42.1 mm         Wing Length:       156 mm         Merganser       Tarsal Length:       51.0 mm         Kight Lobe Wt.       17.4g       MFO (20 ml vial):       V(12.0g)         Whole Liver Weight:       34.2g       Left lobe wt.       16.8g         Right Lobe Wt.       17.4g       MFO (20 ml vial):       V(12.0g)         Whole Liver Weight:       34.2g       Left lobe wt.       16.8g         Right Lobe Wt.       17.4g       MFO (20 ml vial):       V(12.0g)         Witamin A (2 ml cryovial)       V       Weight:       0.6g         Vitamin A (2 ml cryovial)       V       Weight:       0.6g         Porphyrin (2 ml cryovial)       V       Weight:       0.7g         Reserve 2 (2 ml cryovial)       V       Weight:       0.7g         Reserve 3 (2 ml cryovial)       V       Weight:       1.0g         Reserve 4 (2 ml cryovial)       V       Weight:       1.0g         Reserve 4 (2 ml cryovial)       V       Weight:       1.0g         Reserve 4 (2 ml cryovial)       V	No. Collected:	Time of Collection:	1150 Me	thod:	Shot
Species:       Common       Bill Length: <u>42.1 mm</u> Wing Length: <u>156 mm</u> Merganser       Tarsal Length: <u>59.3 mm</u> 9th Primary: <u>51.0 mm</u> Bile Collection (Y/N):       N         Whole Liver Weight: <u>34.29</u> Left lobe wt. <u>16.89</u> Right Lobe Wt. <u>17.49</u> MFO (20 ml vial): <u>V(12.09</u> Metals (2 ml cryovial)       V       Weight: <u>0.69</u> Vitamin A (2 ml cryovial)       V       Weight: <u>0.69</u> Porphyrin (2 ml cryovial)       V       Weight: <u>0.69</u> DNA Integrity (2 ml cryovial)       V       Weight: <u>0.49</u> Reserve 1 (2 ml cryovial)       V       Weight: <u>1.13</u> Reserve 3 (2 ml cryovial)       V       Weight: <u>1.29</u> Reserve 4 (2 ml cryovial)       V       Weight: <u>1.559</u> OC/PCB/Dioxins:       V       Weight: <u>1.59</u> Time MFO into Liquid Nitro:       1309         Kidney left Wt: <u>5.29</u> Kidney right Wt: <u>6.99</u> Mettalothionein:       V       Weight: <u>1.13</u> Porphyrin:       V       Weight: <u>1.13</u> Mettal Analysis:       V       Weight: <u>1.13</u>	collector: <u>M.Wayland</u> c Gam	comments: <u>Downstr</u> bel effluent	ream of Proc	cter and	1
Tibia:       N       Heart:       N       Spleen:       N         Adrenals/Gonads:       N       Thyroids/Parathyroids:       N         Lung:       N       Thymus:       N       Brain:       N         Breast Muscle for Stable Isotopes (freeze)       Y       Gut Sample (Freeze):       Y       Supplementation         Blood on slide for hematozoa:       Y       Y       Supplementation       Y       Supplementation	Specimen No. <u>09</u> Species: <u>Common</u>	Sex: <u>M</u> Weigh Bill Length: <u>42.1</u> Tarsal Length: <u>59.7</u> Bile Collection (Y/N Whole Liver Weight: Right Lobe Wt. <u>17.4</u> Metals (2 ml cryota Vitamin A (2 ml cryota Vitamin A (2 ml cryota DNA Integrity (2 ml Reserve 1 (2 ml cryota Reserve 2 (2 ml cryota Reserve 2 (2 ml cryota Reserve 3 (2 ml cryota Reserve 3 (2 ml cryota Reserve 4 (2 ml cryota OC/PCB/Dioxins: <u>Time MFO into Liquita</u> Kidney left Wt: <u>5.1</u> Mettalothionein: <u>Porphyrin:</u> Reserve: <u>Metal Analysis: <u>Same</u> Tibia: <u>N</u> F Adrenals/Gonads: <u>Lung: N</u> Breast Muscle for St Gut Sample (Freeze) Blood on slide for 1</u>	mm       Wing Leng         9th Prima         9th Prima         N:       N         34.29       Lef         49       MFO (2         al)       Wei         ovial)       Wei         ovial)       Wei         ovial)       Weight:         ovial)       Weight:         ovial)       Weight:         Weight:       N         Heart:       N         table       Isotopes (         :	th: <u>156</u> ry: <u>51.0</u> t lobe wt. 0 ml vial) ght: <u>0.6</u> ght: <u>0.6</u> ght: <u>0.6</u> Weight: Weight: Weight: Weight: Weight: Weight: Weight: <u>8.0</u> ight Wt: <u>1.13</u> <u>1.29</u> Spleen: Parathyroid Brain: (freeze)	$\frac{16.8g}{0.12.0g}$

Date: 618192		Location:	Wapiti River
Latitude: <u>55°05'</u> Lor	ngitude: 118°35'	Habitat:	ide Channel
No. Collected: 10	Time of Collection:	1150 Me	thod: Shot
collector: <u>m.wayland</u> G	comments: <u>Downstre</u> combel effluent.	eam of Pro	octer and
Specimen No. <u>10</u> Species: <u>Common</u> Merganser (Juvenile)	Sex: <u>M</u> Weight Bill Length: <u>46.0 m</u> Tarsal Length: <u>60.1 m</u> Bile Collection (Y/N) Whole Liver Weight: Right Lobe Wt. <u>22.0</u> Metals (2 ml cryoval Vitamin A (2 ml cryoval Vitamin A (2 ml cryoval Porphyrin (2 ml cryoval DNA Integrity (2 ml c Reserve 1 (2 ml cryoval Reserve 2 (2 ml cryoval Reserve 3 (2 ml cryoval Reserve 3 (2 ml cryoval Reserve 4 (2 ml cryoval Reserve 5 (2 ml cryoval Reserve 5 (2 ml cryoval Reserve 6 (2 ml cryoval Reserve 7 (2 ml cryoval Reser	Wing Leng Ming Leng Mro (2 MFO (2	t lobe wt. $14.59$ any: <u>68 mm</u> t lobe wt. $14.59$ and vial): $\sqrt{15.29}$ and: <u>0.59</u> and: <u>1.19</u> Weight: <u>1.29</u> Weight: <u>0.59</u> Weight: <u>0.59</u> Weight: <u>0.59</u> Weight: <u>0.59</u> <u>1317</u> tight Wt: <u>5.59</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.49</u> <u>1.59</u> Spleen: <u>N</u> Brain: <u>N</u>
	Carcass Saved $(Y/N)$	Y	

Date: 618192		Location: Wapiti River
Latitude: <u>55°05′</u> Log	ngitude: 118°35'	Habitat: Side Channel
No. Collected: 11	Time of Collection:	1150 Method: Shot
collector: <u>M. Waylard</u> G	comments: <u>Downstr</u> ampel effluent.	ream of Procter and
Species: <u>ll</u> <u>Common</u> <u>merganser</u> (Juvenile)	Bill Length: <u>41.5r</u> Tarsal Length: <u>55.1</u> Bile Collection (Y/N Whole Liver Weight: Right Lobe Wt. <u>19.5</u> Metals (2 ml cryovia Vitamin A (2 ml cryo Porphyrin (2 ml cryo DNA Integrity (2 ml Reserve 1 (2 ml cryo Reserve 2 (2 ml cryo Reserve 3 (2 ml cryo Reserve 4 (2 ml cryo OC/PCB/Dioxins: Time MFO into Liquid Kidney left Wt: <u>6.7</u> Mettalothionein: <u>V</u> Porphyrin: <u>V</u> Reserve: <u>V</u> Metal Analysis: <u>V</u> Tibia: <u>N</u> H Adrenals/Gonads:	N): $N$ <u>36.86</u> Left lobe wt. <u>17.39</u> <u>56</u> MFO (20 ml vial): $\nu(5.36)$ al) $\nu$ Weight: <u>0.69</u> ovial) $\nu$ Weight: <u>0.69</u> cryovial) $/$ Weight: <u>0.69</u> ovial) $\nu$ Weight: <u>0.69</u> ovial) $\nu$ Weight: <u>0.69</u> ovial) $\nu$ Weight: <u>0.69</u> ovial) $\nu$ Weight: <u>0.79</u> $\nu$ Weight: <u>1.09</u> Weight: <u>1.00</u> Weight: <u>1.00</u> $\sqrt$

Date: 618192	Location: Wapiti River
Latitude: <u>55°05'</u> Lo	ongitude: 118°35' Habitat: Side Channel
No. Collected: 12	Time of Collection: 1150 Method: 5bot
Collector: <u>M. Waylard</u>	Comments: Downstream of Procter and Grambel effluent.
Specimen No. <u>12</u> Species: <u>Common</u> merganser (AduIt)	Sex: <u>F</u> Weight: <u>1225 g</u> Bill Depth: <u>14.8 mm</u> Bill Length: <u>45.2 mm</u> Wing Length: <u>158 mm</u>
	N.B. This individual was an adult.

**Appendix II.** Protocol for field collection and preservation of bird tissues (from Trudeau 1992).

#### INTRODUCTION

This document was prepared to give biologists, blochemists and wildlife technicians an outline of protocols to be used in the field for handling and preserving bird tissues used for studies of environmental contamination using biomarkers.

It is of prime importance that the protocols described in this document be followed. They were established and checked by people who have field and laboratory experience. The labelling and preservation procedures may seem laborious to you but it is essential that they be followed to the letter in order that "YOUR SAMPLES" arrive safely and can be identified at the laboratory. The success of your projects depends on adherence to the protocols.

The protocols described herein are for tissues and blood from birds weighing at least 600 g. If your project involves the collection of tissues and/or blood from smaller birds, you must discuss changes to the original protocols with Blomarker Lab personnel. In some cases the tissue weight can be decreased, in others it cannot, in principle, it should be possible for you to obtain all necessary supplies in your region. If you have difficulty in obtaining certain items on the required materials lists, please contact Biomarker Lab personnel. Make no substitutions without prior inquiry.

Although this document is addressed mainly to users of the CWS Biomarker Lab, it contains some details that go beyond the activities of this laboratory (eg. preservation of samples for histology studies). We have included this information for your use. Several forms which may be used as examples for developing your own forms have been annexed. If you find them suitable, they may be used without alteration.

We wish you a very successful field season.

1.1

## GENERAL RECOMMENDATIONS FOR SPECIMEN COLLECTION AND TISSUE HANDLING

All vials, jars and carcass bags should be labelled before taking them into the field. Each bird should be processed as it is trapped. Do not trap a few and hold them until you process them. This way, each bird is subjected to the same level of stress. Each bird is weighed and a blood sample is taken before it is sacrificed. Birds should be sacrificed by decapitation with a guillotine. This method permits the bird to bleed from the neck and it removes most of the blood from the tissues. Dissections should be performed in a sheltered location such as a tent or a cabin to avoid airborne contamination. Dissections should be done immediately after the sacrifice of the bird. Tissue must be put on lice as soon as it is removed and must be frozen no longer than 30 minutes after the sacrifice of the birds. To avoid metal contamination use stainless steel dissecting tools and rinse tools with distilled water between specimens, using a large acid-weshed weigh boat as working surface. Consistently take the same part of each organ for the same analysis. Do not substitute any of the recommended materials without first verifying with the Biomarker Laboratory Staff. Ensure that all blood and tissue samples are preserved at the appropriate temperature.

SECTION 1 : AVIAN BLOOD AND TISSUE COLLECTION FOR BIOCHEMICAL AND CHEMICAL ANALYSI

This section outlines protocols for collecting and storing blood and tissue from wild birds for the following analyses:

BIOCHEMICAL ANALYSIS	TISSUE
Aminolevulinic Acid Dehydratase (ALA-D)	Whole Blood
Clinical chemistry (e.g. Calcium, Phosphorus)	Plasma
DNA Integrity	Liver, Brain, Red bicod cells
Metallothionein	Kidney
Mixed-Function Oxidases (MFOs)	Liver
Neurotaxicity (eg. Blogenic Amines)	Brain
Porphyrins	Liver, Kidney
Thyroid Hormones	Plasma
Vitamin A	Liver, Plasma
CHEMICAL ANALYSIS	TISSUE
Lead	Tibia
Metals (Mercury, Cadmium)	Liver, Kidney
Organochlorines, PCBs, Dioxins and Furans	Liver
Polyaromatic hydrocarbons	Bile

## OTHERS

## Blood Hemetocrit

Carcass Handling for Tissue Bank

## PROTOCOLS FOR AVIAN BLOOD AND TISSUE COLLECTION FOR BIOCHEMICAL AND CHEMICAL ANALYSIS

### **BLOOD COLLECTION**

Collect a maximum of 10 ml of blood from the wing vein. Use a 22G needle, a Vacutainer holder needle, and the appropriate Vacutainer tube. Blood for ALA-D analysis requires Vacutainers containing powdered EDTA; blood for thyroid hormones, vitamin A and clinical analysis requires heparin-coated Vacutainers.

If whole blood (FOR ALA-D) and plasma (FOR THYROID HORMONES, VITAMIN A AND CLINICAL ANALYSIS) are required, collect 3 ml of blood with a 7 ml Vacutainer tube containing powdered EDTA. Withdraw the Vacutainer tube (without withdrawing the needle and the holder). Insert the 7 ml heparincoated Vacutainer tube into the holder and collect about 7 ml of blood. Withdraw the Vacutainer tube and, with a gauze sponge, apply pressure on the vein where the needle was introduced, and withdraw the needle assembly (needle and holder). Keep pressure on the vein for about 30 seconds to stop the bleeding. Mix blood very gently by inversion and keep on wet ice, in the dark until ready to centrifuge (centrifugation is required for THYROID HORMONES, VITAMIN A AND CLINICAL ANALYSIS but it is not required for ALA-D ANALYSIS).

## IMPORTANT: THE BLOOD MUST BE CENTRIFUGED WITHIN 2 TO 3 HOURS AFTER ITS COLLECTION. FENDING CENTRIFUGATION, THE BLOOD MUST BE KEPT ON WET ICE, IN THE DARK.

### VIA FOR THYROID HORMONES, VITAMIN A AND CLINICAL ANALYSIS

After the sample has been taken for hematocrit measurement, centrifuge the blood in the 7 ml heparincoated Vacutainer tube at approximately 4,000 rpm for 5 minutes and transfer 5 X 0.75 ml plasma aliquots into 1 ml prelabelled cryovials (see below for collection of red blood cells for determination of DNA integrity). Label as outlined above. Freeze on dry ice or in liquid nitrogen. These samples will be kept in a -80°C freezer until THYROID HORMONES, VITAMIN A AND CLINICAL ANALYSIS.

## IMPORTANT: THE BLOOD MUST BE CENTRIFUGED WITHIN 2 TO 3 HOURS AFTER ITS COLLECTION PENDING CENTRIFUGATION THE BLOOD MUST BE KEPT ON WET ICE, IN THE DARK.

CHEMICALS AND DISPOSABLE ITEMS	QUANTITY	SUPPLIER	CATALOGUE #
Critosed sealant for capillary tubes)	1 package	Fisher Scient.	02-678-20
Cryocanes (aluminium)	2/specimen	Gibco	334878
Cryomarkers Cus	tew sets	Sarsted	95963 (blue) 95954 (black) 95955 (green) 95956 (red)
Cryosleeves (Cardboard) or (Plastic)	2/specimen	Respircare Fisher Scient.	MV97-10899 5016-0001
Cryovials 1.0 ml (Nunc)	7/specimen	Gibco	366656
Geuge sponges / CUIS	2/specimen	Pharmacy	
Gloves (latex) V CwS	few boxes	Fisher Scient.	11-394125 A/smail B/medium Ç/large
isopròpanoi	4 litres	Fisher Scient.	A962-4
Ice (wet) V CONTRACTOR	*		
Liquid nitrogen	· · · ·	·	• •
Microhematorit capillary tubes	1/specimen	Fisher Scient.	0266868 wo/heparin
Pipets (plastic) / Cw 5	2/specimen	Fisher Sclent.	13-711-7
Vacutainer holder needle (reusable) المنافع	1 package	Baxter	83023111 (for 7 mi)
Vacutainer sample needle(B-D 7220)	1/spe <b>cimen</b>	Fisher Scient.	22G: 02-885-24
Vacutainer tobe containing powdered EDTA, 7 ml (B-D 6451)	1/specimen	Baxter	B3002.23
Vacutainer tube coated with heparin, 7 ml (B-D 6480)	1/specimen	Baxter	B3014-3A
Wash bottle V CWS	2	Fisher Scient.	03-409-10D

## TABLE 1 MATERIALS REQUIRED FOR BLOOD COLLECTION

\* Quantity depends on the amount of specimen collected.

101-5
104E
110
dium:11394305 ge:1139420 <b>0</b>
589
9-84
5

\* Quantity depends on the amount of specimen collected.

### BIRD SACRIFICE

Sacrifice the bird by decapitation with a guillotine. Let the bird bleed from neck to remove most of the blood from the tissues before dissection. If the brain is required, follow the procedure below. Open the abdominal cavity by making a transverse incision low on the abdomen and pulling the skin flap over the rib cage. Care must be taken to exclude feathers and dirt from cavity. Sever the digestive tract at the oesophagus, clamp with locking forceps (Kelly forceps) and remove whole from the cavity, leaving the caudal end attached at the cloaca to avoid faecal contamination. NOTE: Although the dissecting protocol described in Section II refers to the collection of tissues for histology, you may find it useful. But for purposes other than histological and physiological examination, please use it reservedly. You may not need to go through all the steps of the dissection if you are not collecting all the tissues and organs. Also the tissue handling and storage protocols are not applicable to biochemical and chemical analysis.

REMOVAL OF THE LIVER AND GALL BLADDER

Remove the entire liver with the gall bladder without rupturing the latter. Put on ice in a weight-boat. Remove the gall bladder as outlined below.

## REMOVAL OF GALL BLADDER AND BILE CONTENTS FOR PAH ANALYSIS

To avoid contaminating the liver with bile, clamp (to close it) the gall bladder with locking forceps as close to the liver as possible and then sever above the closure, freeing the gall bladder from the liver. Avoid getting bile on the liver. Place the whole gall bladder in weigh boat, remove locking forceps and collect bile that flows into boat with a pipet. Pipet sample into a 2:0 ml Nunc cryovial and store in liquid nitrogen. In addition to the sample I.D., the label should specify the analysis required. At NWRC, these samples will be stored in à -80°C freezer.

## LIVER HANDLING

After the gall bladder has been removed and stored, record the weight of the entire liver and of the right and left lobes.

## LIVER FOR MFO ANALYSIS

The LEFT LOBE is wrapped tightly in a square piece of autoclave bag and taped. Label tape with a cryopen. Place in 20 ml plastic scintillation vials (2 vials may be required). Label the exterior of the vials with a cryopen. In addition to the sample I.D. and the date of collection, the label should specify the analysis required. Freeze in liquid nitrogen. These samples must be kept in liquid nitrogen or in a -150°C freezer until MFO ANALYSIS. (Do not use tape to label the outside of the vial. It will fall off in liquid nitrogen.)

## LIVER FOR METAL ANALYSIS

Put 1 g of the **RIGHT LOBE** (record the weight) into an acid-washed\* prelabelled 2 mi cryovial. Label the exterior of the vial with a cryopen as outlined above. In the field, freeze on dry Ice or in liquid ogen. At NWRC, these samples will be kept in a -40°C freezer until METAL ANALYSIS.

### LIVER FOR VITAMIN A, PORPHYRIN AND DNA INTEGRITY ANALYSIS

Take 3 X 1 g samples of the **RIGHT LOBE** (record the weights) and place each sample in prelabelled 2 ml cryovials. Label the exterior of the vial with a cryopen as outlined above. In the field, freeze in liquid nitrogen. At NWRC, samples for **VITAMIN A, PORPHYRIN AND DNA INTEGRITY ANALYSIS** will be stored in a - 80°C freezer.

# LIVER AS A "RESERVE" (IF POSSIBLE)

Take 4 X 1 g of the RIGHT LOBE (record the weights) and place each sample in prelabelled 2 ml cryovials. Label the exterior of the vial with a cryopen as outlined above. In the field, freeze in liquid nitrogen. At NWRC, these samples will be stored in a -150°C freezer.

### LIVER FOR OC, PCB, DIOXIN AND FURAN ANALYSIS

Put remainder of liver into a solvent-rinsed\* 50 ml glass jar. Routine OC, PCB, moisture and lipid analysis requires at least 3 g of liver (preferably 5 g). Dioxin and furan analysis requires at least 25 g of liver. Coplanar PCB analysis requires at least 5 g of liver. Moisture and lipid analysis of samples for dioxin and coplanar PCB analysis requires an additional 2 g. Often the liver of the species collected will not be large enough for analysis of dioxins, furans and coplanar PCBs on individuals (eg. the average Herring Guil liver weight is about 28 g). In such cases, dioxin, furan and coplanar PCB analysis is performed on "pooled samples". Label the exterior of the jar using temperature and moisture resistant tape. Put each jar in a Whirl-Pak bag. Label the exterior of the bag using a cryopen. Freeze on dry ice. At NWRC, these samples will be kept in a - 40°C freezer until OC, PCB, DIOXIN AND FURAN \* The vials are rinsed three times with acetorie and three times with hexane.

### KIDNEY COLLECTION AS A "RESERVE"

Put the remainder of the RIGHT KIDNEY into a prelabelled 20 ml plastic scintillation vial. Label as outlined above. Freeze in liquid nitrogen. At NWRC, these samples will be kept in a -160°C freezer.

### KIDNEY COLLECTION FOR METAL ANALYSIS

Put the entire LEFT KIDNEY into a 20 ml prelabelled acid-washed\* plastic scintillation vial. Label as outlined above. Freeze on dry ice or in liquid nitrogen. At NWRC, these samples will be kept in a -40°C freezer until METAL ANALYSIS.

\* The vials are soaked in 10% nitric acid for 24 hrs and rinsed three times with distilled water.

### CARCASS HANDLING FOR TISSUE BANK

Double-wrap the carcass in polyethylene bags, closing up the abdominal cavity to avoid contamination or dehydration of remaining organs. Insert a label between the two bags and stable the bags. Freeze on dry ice. In the field, the carcass can be carried on wet ice until frozen but it should be shipped to NWRC on dry ice.

### GUIDELINES TO FOLLOW PRIOR TO SHIPPING SAMPLES TO NWRC BIOMARKER LABORATORY

The Laboratory Services Section is responsible for the registry of ALL samples received at NWRC that will be analyzed by the Biomarker Laboratory. You must inform the technologist responsible (see name below) AT LEAST ONE DAY IN ADVANCE of the date of arrival of your samples at NWRC to ensure that someone will take care of your shipment upon arrival. If your shipment is by air, courier or bus, the weighbill number is required so we can trace your shipment if it does not arrive when expected. It is preferable to ship your samples early in the week (Monday or Tuesday) and in the morning. Thus, if the shipment is lost, there will be plenty of time to trace it before the weekend. Each shipment must be accompanied by a covering letter which indicates the name of the Project and the Project Officer, the sample numbers and type (e.g.brain, plasma, fiver, etc.), and the analysis (e.g. ALA-D, MFOs, Porphyrins, etc.). Ensure that your samples are preserved at the appropriate temperature during shipment (see Table 4 on the following page).

## Appendix III. Terms of reference.

## NORTHERN RIVER BASINS STUDY

## SCHEDULE OF TERMS OF REFERENCE FIELD COMPONENT OF CONTAMINANTS IN MERGANSERS STUDY

## PROJECT 236:Wildlife Contaminants 1992/93 ProjectSUB-PROJECT 2363:Contaminants in Aquatic Birds

### I. PROJECT DESCRIPTION

The Northern River Basins Study (NRBS) requires the contractor to coordinate and conduct selected aspects of the field component of a study on contaminants in young-of-the-year common mergansers in cooperation with a representative of the Canadian Wildlife Service (CWS), Western and Northern Region. This study will be done on portions of the Athabasca, Peace, Wapiti, and Smoky Rivers.

## II. TERMS OF REFERENCE

### TASKS:

- 1. The contractor is to supply two people to collect, in cooperation with CWS personnel, five young-of-the-year, age Class III common mergansers (see Gollopp and Marshall, 1954, Mississippi Flyway Council Technical Section) from each of the following sites:
  - i) upstream from the Weldwood/Hinton effluent outfall on the Athabasca River, or in nearby feeder creeks and lakes;
  - ii) downstream from the Weldwood/Hinton effluent outfall on the Athabasca River;
  - iii) upstream from the P&G effluent outfall on the Wapiti River or on nearby feeder creeks and lakes;
  - iv) downstream from the P & G effluent outfall;
  - v) upstream from the Daishowa mill effluent outfall near Peace River on the Peace River;
  - vi) downstream from the Daishowa mill effluent outfall on the Peace River;
  - vii) upstream from the ALPAC mill site on the Athabasca River;
  - viii) downstream from the ALPAC mill site on the Athabasca River;
  - ix) upstream from Fort McMurray on the Athabasca River;
  - x) downstream from the SUNCOR plant on the Athabasca River.

Mergansers should not be collected within a buffer zone extending 20 km upstream from and 20 km downstream from the above-named outfalls/ townsites. Ideally, birds will be collected as close as possible to the borders of these buffer zones. However, where necessary, collections may be made up to 130 km upstream or downstream of each aforementioned outfall/townsite.

- 2. The contractor will assist the CWS representative in processing and preparing the samples.
- 3. The contractor will ensure that their representative(s) is in possession of a valid Firearm Acquisition Certificate, and in cooperation with the CWS representative, will ensure that all relevant information is obtained regarding prohibitions of discharges of firearms.
- 4. The contractor will arrange to ship pertinent tissues to the Canadian Wildlife Service, National Wildlife Research Centre (CWS- NWRC), 100 Gamelin Blvd., Hull, Quebec, K1A OH3 for tissue preparation and archiving.

### **METHODS/APPROACHES:**

- 1. Through their knowledge of the aforementioned river systems, the contractor will undertake to supply all the necessary transportation equipment for this project (including boats, motors, jetboats (if required), fuel, fuel tanks, vehicles and trailers). The contractor will also supply 10 gram, 100 gram and 1 kilogram Pesolas for weighing the birds and tissues, metric Vernier callipers for measuring the specimens, tinfoil and plastic freezer bags for storing the specimens, and labels and markers for labelling the specimens. The contractor will also have available a 12 or 20 gauge shotgun and ammunition for collections.
- 2. The contractor will determine all relevant points of access to these rivers and will advise NRBS and the CWS representative of risks/ dangers/difficulties in accessing certain zones of these rivers well in advance of the field work in order that alternative plans can be devised.
- 3. In cooperation with the CWS representative, the contractor will undertake to collect the aforementioned birds by shooting with a 12 or 20 gauge shotgun, or by other means such as drive trapping or nightlighting in situations where the use of firearms may not be permitted.
- 4. The contractor will assist the CWS representative with the weighing, aging, sexing and measuring of the carcass, removal of gut contents, livers and kidneys, and with the bagging, labelling and storage on dry ice or liquid nitrogen of all tissues.

- 5. The contractor will obtain the necessary Export permits from the Government of Alberta prior to shipping tissues and will arrange to ship the tissues on dry ice in suitable containers to CWS- NWRC, 100 Gamelin Boulevard, Hull, Quebec, K1A OH3, c/o Mr. Michael Kassera (Telephone: 819-953-8701). Mr. Kassera should be contacted by phone before the tissues are sent. The contractor will ensure that the tissues are in transit for no more than 18 hours and will enlist the services of a professional expediter to facilitate this process.
- 6. Ideally, the contractor will kill a maximum of one (1) duckling per brood at any given site. However, where brood densities are low, a kill of more than one (1) duckling per brood up to a maximum of five (5) per brood will be permitted.

### **III. REPORTING REQUIREMENTS**

1. The contractor will keep field records concerning all pertinent information for each specimen. Upon completion of the field work, the contractor will prepare a final report detailing methodology, exact collection locations, date of collections, collection method, approximate age, weight, sex, bill length, measurements of overall size (bill length, depth, wing chord length, tarsal length, keel length), gut contents present or absent, and disposition of various tissues (i.e., frozen immediately and remained frozen up to and including shipment date or remained thawed for X hours prior to freezing, or placed on liquid nitrogen immediately, etc.). The report will also indicate the date that the tissues were shipped. Copies of the field records should be submitted with the final report. One copy of the final report should be sent to the NRBS and another to the Scientific Authority (see below).

### IV. SCHEDULING

- 1. Complete itinerary and submit to CWS representative no later than July 24, 1992.
- 2. Field work August 4 August 21.
- 3. Provide final report by Sept. 11, 1992.
- V. COMPLETION DATE

September 11, 1992.

### VI. SCIENTIFIC AUTHORITY

Mark Wayland CEPA Wildlife Biologist Canadian Wildlife Service 115 Perimeter Road Saskatoon, Saskatchewan S7N OX4 Phone: (306) 975-6340 Fax: (306) 975-4089 3 1510 00135 5685

