Hatchery Management Plan Supporting Kootenai River White Sturgeon Restoration

Prepared by the Kootenai Tribe of Idaho

Dr. Shawn P. Young, PhD and Nathan R. Jensen

Editors: Meridian Environmental, Inc.

Jeannie Heltzel and Robyn Rice

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The Kootenai Tribe of Idaho (KTOI) would like to recognize the efforts of all those who have played key roles in advancing the restoration of the Kootenai River White Sturgeon population. KTOI would like to acknowledge our partners, Idaho Department of Fish and Game (IDFG), British Columbia Ministry of Forests (BC Ministry), Montana Fish Wildlife and Parks (MFWP), U.S. Fish and Wildlife Service (USFWS), U.S. Corps of Engineers (USACE), and the Bonneville Power Administration (BPA). This work, KTOI *Project 198806400*, is funded by the Bonneville Power Administration as part of the Northwest Power and Conservation Council's (NPCC) Columbia Basin Fish and Wildlife Program.

This document is an update of the Monitoring and Evaluation Plan for Kootenai River White Sturgeon produced during the NPCC Step 1-3 Master Plan process, which was required to secure funding for the Twin Rivers Tribal White Sturgeon and Burbot Hatchery. The Kootenai Tribe of Idaho would like to acknowledge the previous version of this document was based on the monitoring and evaluation (M&E) plans created for the Chief Joseph Dam Hatchery Program by the Confederated Tribes of the Colville Reservation and their consultant, D.J. Warren & Associates Inc. (Colville Tribes 2009a, 2009b). We believe their approach, which provided a framework for implementing and evaluating hatchery and field monitoring and a process for multi-agency cooperation and management, is well suited to the Kootenai River Native Fish Conservation Aquaculture Program (KRNFCAP) and has greatly helped to ensure the program's successful implementation.

Cover photo: Releasing Kootenai River White Sturgeon sac-fry near Crossport, Idaho into the Kootenai River mainstem (rkm 256).

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The goals of the Kootenai River Native Fish Conservation Aquaculture Program (KRNFCAP or Program) are to prevent extinction of the endangered White Sturgeon (*Acipenser transmontanus*) population and reintroduce Burbot (*Lota lota maculosa*), both of which are culturally significant resources to the Kootenai Tribe of Idaho, related First Nations, and the citizens of the Kootenai Basin. The Sturgeon program goals are to rebuild a healthy age-class structure using conservation aquaculture techniques with wild, native broodstock, and preserve the existing gene pool by spawning, rearing, and releasing fish that survive, grow, and mature. The next generation of Kootenai Sturgeon will be predominantly hatchery-origin. These hatchery origin fish are expected to supply the reproductive potential to restore a self-sustaining population that will also support cultural and/or recreational fisheries. To date, the program has successfully warded off extinction and rebuilt a healthy age-class structure by spawning >450 wild adults (~150 females crossed with ~300 males) and creating >350 unique families including >30 year-classes since 1990. The annual spawning plan captures approximately 80% (since 2012) of the allelic variation from each wild spawning class; and overall has captured an estimated 96% of the genetic diversity of the wild population (Schreier and Van Eenennaam 2019).

Since 1990, the program has released >300,000 juveniles throughout the Lower Kootenai River and Kootenay Lake Recovery Area in Montana, Idaho, and British Columbia, with the majority released into the Meander Reach downstream of Bonners Ferry, Idaho, and into Kootenay Lake, British Columbia. Post-release monitoring and evaluation (M&E) of hatchery fish is primarily conducted by Idaho Department of Fish and Game (IDFG) *Project 198806500*, British Columbia Ministry of Forests (BC Ministry) subcontracted by IDFG *Project 198806500* and KTOI *Project 198806400*, and Montana Fish Wildlife and Parks (MFWP) *Project 200600800* within their respective jurisdictions. Age-1 juvenile releases have resulted in a river/lake abundance of 12,000-15,000 hatchery-reared Sturgeon, now ages ≤30 years old (Dinsmore et al. 2015; Hardy et al. 2020), with the first sexually mature hatchery-origin males observed in 2020 (KTOI unpublished data, IDFG unpublished data). Evaluation of post-release recaptures determined that 56% of all families released were represented in the population through 2014 (Schreier et al. 2015). The Twin Rivers Tribal Sturgeon and Burbot Hatchery (Hatchery 2, “H2” or “TR”), which began operations in 2015, combined with the Kootenai Tribal Sturgeon Hatchery (Hatchery 1, “H1” or “KT”) now incorporates twice as many wild adult spawners per year as prior to 2015, which is of paramount importance as wild adult abundance declines. By doubling the number of unique families created annually, the program increases the probability that genetic diversity is maintained while also increasing behavioral and phenotypic diversity. The ability to create more unique families or “family groups” is important for the implementation of a genetic Parental Based Tagging (PBT) program.

The Sturgeon program is addressing two major issues detected in recent years: 1) apparent declines in post-release survival and growth of hatchery-origin Sturgeon, and 2) spontaneous autopolyploidy. M&E results indicate post-release survival of hatchery Sturgeon during their first year in the river from age-1 to age-2 has declined as hatchery

juvenile/sub-adult abundance has increased (Dinsmore et al. 2015). Further, recapture data indicates highly variable individual growth rates (Ross et al. 2015; Stephenson and Evans 2015; Hardy et al. 2020). These findings may suggest the Sturgeon carrying-capacity of the altered ecosystem has been reached, and density-dependent effects are being observed. Since the Sturgeon program was initiated in 1990, the ecosystem has endured an additional 30 years of human-induced ultra-oligotrophic conditions and variable hydro-operations resulting from Libby Dam operations. The hatchery Sturgeon population structure will be managed to balance abundance, growth, and niche space to maintain a diverse standing-stock, which will continue to require adding future year-classes as recruitment failure persists. KTOI has agreed with co-managers to reduce annual year-class releases to 500 per family group of age-1 juveniles per female spawned (e.g., 10 females spawned = 5,000 juveniles released) and to implement a removal program in the Idaho portion of the river to remove spontaneous autopolyploids, portions of over-represented year-classes, and individuals exhibiting abnormal growth or physical condition.

Removal of spontaneous autopolyploids will be the top priority of in-river removal due to the long-term effects of spontaneous autopolyploids breeding with normal ploidy fish. The condition was discovered in Kootenai Sturgeon during routine genetics monitoring of the 2011 year-class (Schreier et al. 2013); and has since been identified in most Columbia Basin White Sturgeon sub-populations, other White Sturgeon populations, commercial aquaculture operations, and conservation aquaculture programs, at various levels of occurrence (Schreier et al. 2021). A more detailed summary concerning spontaneous autopolyploidy in White Sturgeon is presented in Schreier et al. 2021. The number of abnormal ploidy Sturgeon released before 2012 is unknown, but it is known that these hatchery fish survived and are maturing. Since 2012, management of the issue has evolved as investigators explored potential causes of spontaneous autopolyploidy (Gille et al. 2015; Van Eenennaam et al. 2020) and developed accurate genetic testing methods (Fiske et al. 2019, 2022, 2023).

Along with removal and reduced release numbers, additional habitat improvement actions are needed to improve ecosystem productivity in the Meander Reach, both in-channel and off-channel. Such actions are proposed through KTOI's *Project 200200200* and *200201100*. The Meander Reach, a focal habitat for releases, supports most of both hatchery Sturgeon and Burbot; in addition, some fish released in other locations have emigrated to the reach (Stephenson et al. 2013; Hardy et al. 2015; Ross et al. 2018; Hardy et al. 2020). Both Sturgeon and Burbot spawn within this reach (Ross et al. 2018; Hardy et al. 2020). Historically, the Kootenai Valley Floodplain supported abundant fish populations, and remains a preferred habitat even in its current degraded condition. Expanding in-channel nutrient additions to the Braided Reach as proposed by KTOI *Project 199404900* and to the Meander Reach as proposed by KTOI *Project 198806400* would connect the current Canyon Reach and Kootenay Lake nutrient additions currently implemented by *Project 199404900* creating a continuum of enhanced conditions through critical Sturgeon habitat and the recovery area as whole. Adding in-channel structure, restoring off-channel habitats and connections, and re-evaluating spring flow management (timing and magnitude) are other actions needed in synergy to address recruitment failure and provide suitable habitat for all

life stages, including hatchery fish that must survive, grow, and mature to support the next generations.

The KRNFCAP will continue to rebuild the Sturgeon population in a manner that (1) maintains genetic diversity, population structure, and abundance until natural recruitment is restored to a magnitude that is self-sustaining, and (2) maintains adult abundance above minimum adult spawning targets to support cultural and recreational harvest (KTOI 2007; KTOI 2010; KTOI 2012; KTOI 2012a; KTOI 2012b; KTOI 2018a; KTOI 2019a; USFWS 2019). As of 2020, the long-term goal of restoring natural recruitment to self-sustaining levels that can also support fisheries has not been achieved. Empirical evidence, such as annual captures of only a few naturally produced larvae (Ross et al. 2018; Hardy et al. 2020), indicate recruitment failure persists. Several factors will dictate if and when natural recruitment may be restored to self-sustaining levels, including time to maturity of hatchery-origin Sturgeon; a declining wild adult population; persistence of wild adults spawning in the sand and silt dominated Meander Reach; hydro-operations variability; poor ecosystem productivity; the magnitude of habitat restoration needed; contaminants leaching in from upriver coal mines in southeastern British Columbia and other anthropogenic activities.

This Hatchery Management Plan for Kootenai River White Sturgeon will provide management guidance for the program. The plan will be updated annually with the most recent RM&E data, an update of any changes in the aquaculture program, and will reflect decisions made during the annual adaptive management processes, which includes an Annual Program Review (APR) workshop attended by all co-managing agencies. Future APR workshops will be hosted by co-managing agencies and KTOI on a rotating basis.

1.0 INTRODUCTION

The goals of the Kootenai River Native Fish Conservation Aquaculture Program (KRNFCAP) are to prevent extinction of the endangered White Sturgeon (*Acipenser transmontanus*) population and reintroduce Burbot (*Lota lota maculosa*), both of which are culturally significant resources to the Kootenai Tribe of Idaho (KTOI or Tribe), related First Nations, and the citizens of the Kootenai Basin; to rebuild a healthy age class structure using conservation aquaculture techniques; and to preserve the existing gene pool by spawning, rearing, and releasing fish that survive, grow, and mature (KVRI 2005; KTOI 2007; KTOI 2010; KTOI 2012; KTOI 2012c; KTOI 2018b; KTOI 2019b).

KRNFCAP (*Project 198806400*) was initiated in 1989 to assist with recovery of Federally endangered Kootenai River White Sturgeon (designated in 1994). The 2019 Revised Recovery Plan for the Kootenai River Distinct Population Segment of White Sturgeon (USFWS 2019) lists two specific recovery actions pertaining to *Project 198806400*. Recovery Action 1.1 recommends continuing the conservation aquaculture program; and Recovery Action 1.2 recommends continuing to adaptively manage the conservation aquaculture program as implemented by KTOI's KRNFCAP.

KTOI operates two conservation aquaculture facilities, Kootenai Tribal Sturgeon Hatchery constructed in 1989 (*Project 198806400)* and Twin Rivers Tribal White Sturgeon and Burbot Hatchery constructed during 2014 (*Project 200902400*; now operated under *Project 198806400)* to restore Kootenai River White Sturgeon and Burbot. The KRNFCAP is an excellent example of how fish population restoration via conservation aquaculture programs may achieve objectives through collaborative adaptive management guided by research, monitoring, and evaluation (RM&E) results.

Post-release monitoring and evaluation (M&E) of hatchery fish is primarily conducted by Idaho Department of Fish and Game (IDFG) *Project 198806500*, British Columbia Ministry of Forests (BC Ministry) subcontracted by IDFG *Project 198806500* and KTOI *Project 198806400*, and Montana Fish Wildlife and Parks (MFWP) *Project 200600800* within their respective jurisdictions.

The above-mentioned projects collectively with other KTOI projects, *199404900 Kootenai River Ecosystem Restoration*, *200200200 Restore Natural Recruitment of Kootenai River White Sturgeon*, and *200201100 Kootenai River Operational Loss Assessment* demonstrate how habitat restoration in combination with conservation aquaculture may achieve significant ecosystem-scale goals. The future program will strive to achieve these goals while also providing experimental releases of early life stages to investigate the altered habitat dynamics that perpetuate recruitment failure of Kootenai Sturgeon and Burbot.

Overall, the KRNFCAP has successfully met interim biological objectives set forth in planning documents for hatchery fish (KVRI 2005; KTOI 2007; KTOI 2010; KTOI 2012; KTOI 2012a;

KTOI 2012b; KTOI 2012c; KTOI 2018a; KTOI 2018b; KTOI 2019a; KTOI 2019b; USFWS 2019). In doing so, KTOI has recently re-evaluated and updated several long-standing biological and implementation objectives in collaboration with working-group members representing co-managing agencies through co-hosted Annual Program Reviews (APRs) for both Sturgeon and Burbot.

KTOI's goals extend beyond merely avoiding extinction to recovering Kootenai Sturgeon as a viable component of a functional ecosystem. Sturgeon recovery is also essential to the provision and maintenance of federal trust responsibility and mitigation obligations for the negative effects of federal hydropower development in the Kootenai River system. An effective aquaculture program is part of KTOI's comprehensive ecosystem restoration effort that also includes: 1) nutrient addition to increase biological productivity and food availability; 2) aquatic, riparian, and adjacent terrestrial habitat restoration; and 3) rebuilding of other depleted native species (e.g., Kokanee *O. nerka* and Burbot *L. Lota*).

The purpose of this Hatchery Management Plan for Kootenai River White Sturgeon is to provide a framework that summarizes program goals and objectives, describes the conservation aquaculture program components, introduces what is being monitored and evaluated and details how all the working parts of the program are adaptively managed. This plan is designed to provide management guidance updated annually with the most recent RM&E data, a description of any changes in the aquaculture program, and a discussion of decisions made during the annual adaptive management processes. Following this introduction, Section 2 provides an overview of the Kootenai River White Sturgeon population status and recovery goals. Section 3 describes future program management and aquaculture program objectives; provides Status and Trends data for the program (hatchery and post-release); and summarizes the program's Key Assumptions, Decision Guidelines, and Biological Objectives. Section 4 describes the program's RM&E activities, focusing primarily on in-hatchery RM&E. Finally, Section 5 describes the programs Adaptive Management process, which includes an Annual Program Review attended by all co-managing agencies.

In addition to the core co-managing agencies (KTOI, IDFG, MFWP, BC Ministry), this plan includes significant contributions from Tribal Governments; Federal action agencies (Bonneville Power Administration (BPA), US Army Corps of Engineers (USACE)); Federal regulatory agency personnel (US Fish and Wildlife Service (USFWS)); Canadian Federal and Provincial Governments (Fisheries and Oceans Canada (DFO), and local community stakeholders (Kootenai Valley Resource Initiative (KVRI).

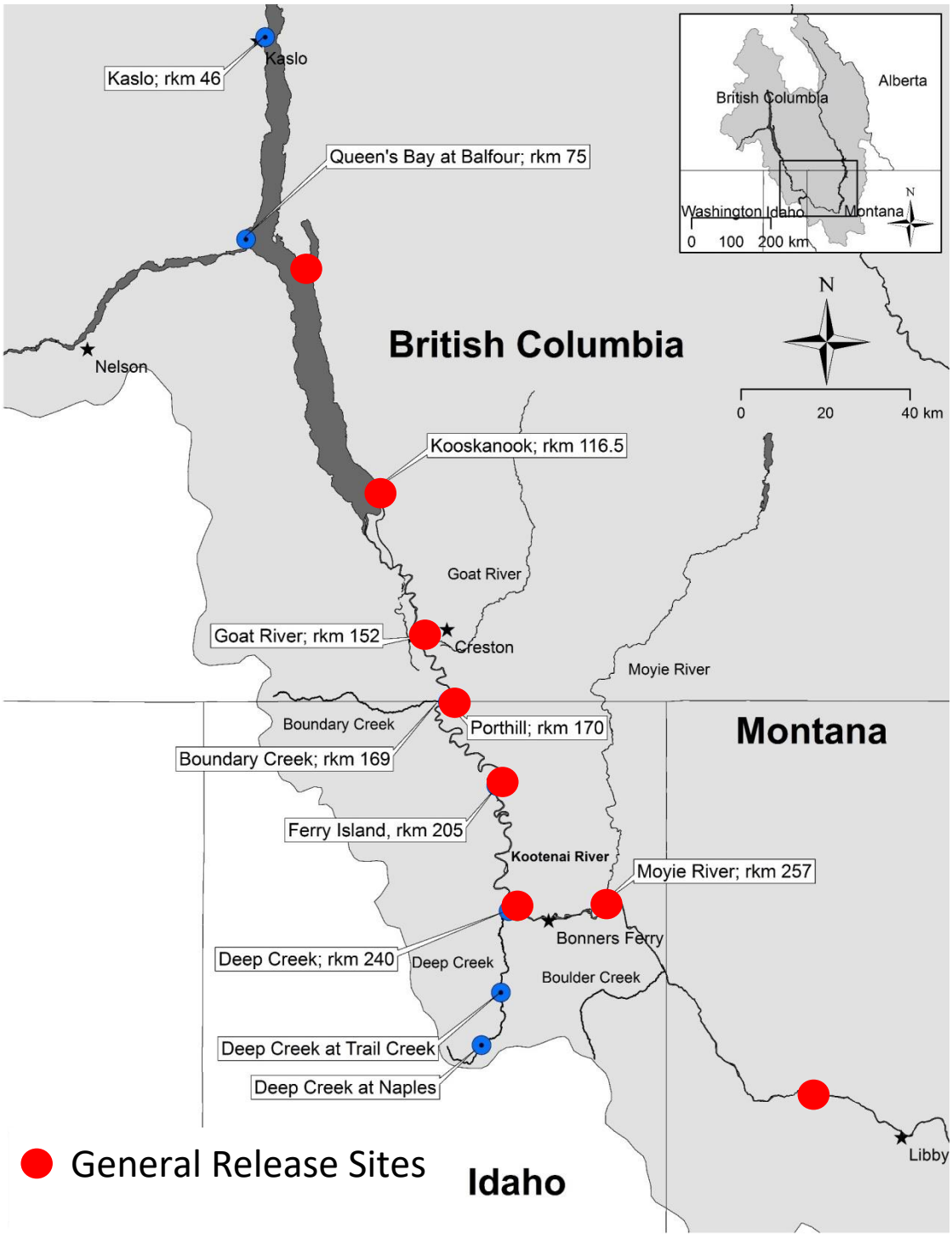

Figure 1. Kootenai River Basin project area (courtesy TJ Ross, IDFG).

2.0 RECOVERY GOALS AND PLANNING OBJECTIVES

Past and planned Sturgeon Program activities address long-term recovery goals and interim objectives that chart a pathway to recovery. Recovery goals and hatchery objectives have evolved over the past three decades based on new information and experience. This plan is an adaptive response to developments and new information collected since the Kootenai River White Sturgeon Recovery Plan was adopted in 1999 (USFWS 1999) and revised in 2019 (USFWS 2019). This plan reflects the goals and restoration strategies proposed by KTOI and mutually agreed upon by co-managers (KTOI 2018a, 2019a); and then as described in the Revised Recovery Plan (USFWS 2019) and KTOI Project 198806400 proposal for the NPCC Independent Scientific Review Panel (ISRP) review of resident fish and Sturgeon programs (KTOI 2020).

2.1 PROGRAM HISTORY

Kootenai River White Sturgeon are in a very difficult situation. Their physical and biological ecosystem has been severely altered by decades of human activity. Self-sustaining natural recruitment has failed for over 50 years (Paragamian et al. 2005; Ross et al. 2018; Hardy et al. 2020). The wild population now consists of an ever-dwindling cohort of large, old fish. The most recent modeling indicates the remaining wild adult population is larger than the previous 2014 estimate consisting of approximately 1,700 fish (Ross et al. 2018; Hardy et al. 2020). Population monitoring also indicates that natural recruitment is still at low levels insufficient to sustain the population. High levels of mortality are still occurring during early life stages (Ross et al. 2018; Hardy et al. 2020). Twenty-plus years of research and experimentation has determined that recruitment is failing in the incubation and early larval stages because spawning currently occurs in an area of unsuitable habitat. If not for their longevity, Sturgeon would have followed Kootenai Burbot and Kokanee into extirpation years ago.

KTOI's conservation aquaculture program was initiated in 1988 as a low-capital, experimental facility designed to assess gamete viability and explore the feasibility of Sturgeon aquaculture, which was then in its infancy in North America. Kootenai River White Sturgeon assessment and conservation efforts under the NPCC Fish and Wildlife Program began in 1989 and are ongoing under a multi-agency cooperative effort (KTOI 2007; KTOI 2012; KTOI 2018a; KTOI 2019a; Ross et al. 2015; Stephenson and Evans 2015; Hardy et al. 2016). The Distinct Population Segment (DPS) of Kootenai River White Sturgeon was listed as endangered by USFWS in 1994. Subsequently, the Kootenai River White Sturgeon Recovery Team was formed in 1994 to develop and help coordinate implementation of a Recovery Plan (Duke et al. 1999). Program objectives and facilites have evolved over time in response to demonstrated successes, lessons learned through implementation of the program, and the growing awareness of the Program's significance to Sturgeon conservation. In 2013, an APR workshop was initiated by the KTOI to further assist co-manager decisions/agreements to utilize conservation aquaculture and guide research, monitoring, and evaluation activities (KTOI 2012). In the Canadian portion of the Kootenai

drainage, Sturgeon were Species at Risk Act (SARA) listed as Endangered in 2006 and are addressed in the Recovery Strategy (DFO 2014). As of 2021, the APRs were the main adaptive management forum as the Kootenai River White Sturgeon Recovery Team has not convened for several years.

Similar to other non-recruiting populations, management actions are focused on: (1) hatchery supplementation as an interim measure and (2) habitat restoration to re-establish natural recruitment. Since 1990, >300,000 juvenile Sturgeon have been released by KTOI's conservation aquaculture program, including both Tribal hatcheries and the fail-safe Kootenay Hatchery in British Columbia (2015 was final year for Kootenay Hatchery releases). Annual production goals at KTOI's Tribal facilities are shown in Table 1.

Table 1. Annual production goals at KTOI Sturgeon facilities.

	Tribal Sturgeon Hatchery (H1)	Twin Rivers Hatchery (H2)	Total
Broodstock number	Up to 20*	Up to 40*	Up to 60*
Families	Up to 20	Up to 20	Up to 40
Family groups (maternal)	5-10 (if both hatcheries are operational);10-20 (if all spawning occurs at KT hatchery)	5-10 (if both hatcheries are operational); 10-20 (if all spawning occurs at TR hatchery)	10-20
Total releases per year	2,500-5,000	2,500-5,000	≤10,000

* Up to 20 female and 40 male broodstock annually per USFWS Section 10 permit. Actual numbers of broodstock vary annually by facility; Males are not typically taken to a hatchery. Sperm is collected in the field and transported to the hatcheries for spawning activities.

The current estimate of the standing hatchery-reared population of juveniles and sub-adults is 12,000–15,000 (Dinsmore et al. 2015; Hardy et al. 2020). The first mature hatchery origin male transported to a KTOI hatchery was captured during 2021. KTOI has now incorporated multiple mature, flowing hatchery origins males into program activities. A total of four ripe hatchery males were incorporated in 2022 (ages of ripe males ranged 17-25 years old). The number of hatchery-origin adults in the population is expected to increase into the foreseeable future. Meanwhile, experiments with flow augmentation and modification of spawning substrate have been attempted to restore natural recruitment. Recent studies of flow, river substrate, and sediment transport suggest lack of suitable spawning substrate is likely the factor limiting recruitment and indicate that an aggressive habitat enhancement program could reverse recruitment failure (Paragamian et al. 2009; KTOI 2009; McDonald et al. 2010). However, an interesting and enigmatic pattern of adult spawning behavior has been a major determinant in deciding future restoration activities. Adult Sturgeon show high fidelity year after year to spawning sites near Bonners Ferry, ID, that have not supported adequate recruitment for decades, regardless of flow and habitat actions.

Adults have full access to plentiful upstream habitat deemed 'classical' or 'critical' White Sturgeon spawning habitat; yet the wild adults rarely move upstream into these habitats (Ross et al. 2015; Hardy et al. 2016; Hardy et al. 2020). Translocation of reproductively

ready adult Sturgeon from the downriver spawning sites to upstream areas with cobble and boulder substrate was attempted, but most translocated adults rapidly left the release location. Although some eggs have been collected at the release location, it was uncertain whether they had been fertilized (Rust 2011). Another strategy employed was releasing free embryos from 2008-2012 (Table 5). The results of this action has been inconclusive due to the lack of immediate recaptures and the absence of PBT to assess survival of these embryos at older ages (Rust and Wakkinen 2011).

Other recovery strategies implemented for Kootenai River Sturgeon include major river channel rehabilitation; spawning substrate enhancement; floodplain and off-channel habitat reconnections and restoration; and nutrient enhancement (*KTOI Projects 200200200, 200201100, and 199404900*), with more planned.

The program has achieved, at least for the time being, the immediate objectives of producing consistent annual juvenile age classes and forestalling demographic extinction. Post-release monitoring conducted by IDFG, BC Ministry, and MFWP has determined that substantial numbers of hatchery-reared fish have successfully adapted to current river conditions. If enough hatchery-produced fish continue to survive, grow, and mature, the hatchery program will have bought time to implement the large-scale ecosystem improvements necessary to restore natural production and long-term population sustainability.

The hallmark of the Kootenai River White Sturgeon recovery effort has been its experimental adaptive approach to address substantial uncertainties and ever-changing habitat dynamics. The program has evolved in response to new data and information, changing demands, and ever-changing habitat dynamics. New information and evaluations have characterized the Sturgeon conservation and recovery effort in the face of very large uncertainty regarding limiting factors and effective remedies. Hatchery facility requirements have also changed over the course of the recovery effort. We have every reason to expect this pattern to continue for the duration of the recovery effort. The additional facility at Twin Rivers along with upgrades of the Kootenai Tribal Sturgeon Hatchery provide flexibility in space, systems, and water supplies necessary to continue to implement this program in an adaptive and effective manner.

To address Sturgeon recovery goals and fill the demographic and genetic gaps left by the absence of natural reproduction, hatchery-reared Kootenai Sturgeon have been spawned from wild adult broodstock annually since 1990. Since 1992, the Kootenai Tribe's Kootenai Sturgeon aquaculture program has released >300,000 hatchery-reared juvenile Sturgeon into the Kootenai River basin (Table 2A). During 1995-2014, 2,000 to 40,000 juveniles ranging from age-0 to age-4 (mainly age-1) were released annually. Year classes were genetically represented by as many as 18 families (1 female crossed with 1 male) until 2015 when the addition of Twin Rivers Hatchery allowed for an increase up to 40 families per year class. Given the increase in capacity and numbers of adults incorporated into each year class KTOI now tracks Family Groups (3-5 males crossed with each female) rather than Families (one to one cross) because although female ova aliquots are fertilized individually,

they are pooled into one large group post fertilization due to facility space limitations (Table 2).

Table 2. Summary of facility and post-release survivorship contributions to hatchery spawning, releases, and post-release survivorship of Kootenai White Sturgeon from 1992-present.

(A)

Years / Year Classes	Hatchery					
	KTOI – Tribal Sturgeon*		BC**		KTOI – Twin Rivers*	
	Released/ Families/Groups	Mean Wt (g)	Released/ Families/Groups	Mean Wt (g)	Released/ Families/Groups	Mean Wt (g)
1992 - 1998 / 5	2,630 / NA	47 – 863				
1999 - 2003 / 5	19,857 / 50	33 – 294	27,927 / 25	44 - 99		
2004 - 2006 / 3	65,363 / 30	10	37,107 / 15	10		
2007 - 2014 / 8	44,839 / 80	32 – 76	55,647 / 40	56 - 101		
2015 - 2018 / 4	***17,916 / 70	74 (6 - 125)			***41,853 / 108	46 (20 - 125)
2019 - 2020 / 1					2162 / 8	56 (54 -58)
2021 – 2022 / 2	1,365 / 8	140			1,481 / 8	164
2022 – 2023 /						

*Did not cull fish from year classes 1992 - 2018.
**Per agreements, culled fish < 20g some years.
***Beginning in 2015, half of each family group reared at each KTOI facility after spawning.

(B)

Years / Year Classes	Hatchery					
	KTOI – Tribal Sturgeon*		BC**		KTOI – Twin Rivers*	
	Season Released	First Year Survival	Season Released	First Year Survival	Season Released	First Year Survival
1992 - 1998 / 5	Variable	0.70 – 0.88				
1999 - 2003 / 5	Variable	0.13 – 0.43	Variable	0.57 – 0.61		
2004 - 2006 / 3	Fall & Spring	0.008 – 0.12	Fall & Spring	0.11 – 0.22		
2007 - 2014 / 8	Fall	0.004 – 0.11	Spring	0.05 – 0.21		
2015 - 2018 / 4	Fall	< 0.10			Spring	~0.10
≥2019 / >1	Spring	unknown			Spring	unknown

*Hatchery-specific survival related to size and season of release (Dinsmore et al. 2015; Hardy et al. 2020).

The aquaculture program has undergone five major iterations in the number of juveniles released (Table 2A), age/size at release (Table 2A), and season released (Table 2B). Releases from 1992-1998 were largely experimental and consisted of small year classes of variable ages and sizes. During 1999-2003, average annual releases increased to ~10,000 age-1 and age-2 juveniles as a second facility, for back-up, was added to the program. Another experimental iteration implemented by KTOI and the co-managers focused on releasing a high number of smaller, age-0 and age-1 juveniles during 2004-2006. Average annual releases increased to ~34,000 juveniles with a mean weight of only ~10 grams.

Following the 2004-2006 experiment of releasing high numbers of smaller/younger juveniles, the program returned to the strategy used during 1999-2003. Average annual releases were ~12,500 age-1 juveniles; however, the KTOI Tribal Sturgeon Hatchery and BC Kootenay Trout Hatchery released fish during different seasons. This was due to the hatcheries using different temperature regimes for fish rearing, resulting in disparate size-at-age of juveniles. Also, the two hatcheries reared different numbers of families, which later confounded estimation of post-release genetic contributions (Schreier et al. 2015) and is a contributing factor for overrepresented year-classes.

Size-at-release and season-of-release significantly affect survival of age-1 hatchery-origin juveniles (Beamesderfer et al. 2014a; Dinsmore et al. 2015; Hardy et al. 2020). Fish reared on an accelerated temperature regime at the BC Hatchery were released as larger, age-1 juveniles in the spring, whereas fish reared on the ambient temperature regime at the KTOI Tribal Sturgeon hatchery were released the following fall/winter. This resulted in significant differences in survival and growth between the two hatcheries (Table 2B); in addition, the BC hatchery only reared 5-6 families annually compared to 10-12 at the KTOI Tribal Hatchery. The knowledge gained from the BC hatchery is now used to adaptively manage the KTOI program's rearing strategies.

Beginning with the 2015 year-class, the BC Trout Hatchery was no longer used for the Sturgeon program; all juveniles are now reared at the two KTOI Tribal hatcheries. Currently, all fish are reared on heated water in winter (Kootenai River water is heated in winter at Tribal Hatchery 1 and ground water at Tribal Hatchery 2-Twin Rivers) to accelerate growth. Accelerating growth allows for all hatchery sturgeon from both facilities to be released in spring. Spring releases and increased size at the time of release has been shown to positively affect survival (Dinsmore et al. 2015; Hardy et al. 2020).

2.2 KTOI GOAL

KTOI's recovery goal for Kootenai River White Sturgeon is:

To ensure the persistence and viability of a naturally-reproducing population as an essential element of an adequately functional ecosystem and a resource supporting traditional beneficial uses.

Note: The KTOI conservation aquaculture program is also designed to address Tribal trust responsibilities.

2.3 2019 RECOVERY PLAN OBJECTIVES

The Kootenai River White Sturgeon Recovery Plan (USFWS 1999) identified objectives and criteria reflecting the best available information at that time. The Revised Recovery Plan (USFWS 2019) updated the recovery objectives, strategies, and criteria for downlisting and de-listing. The Revised Recovery Plan identifies a long-term goal of down-listing and delisting Kootenai White Sturgeon when the population becomes self-sustaining. The plan notes that recovery will not be complete until there is survival to sexual maturity, which may take 25 to 30 years for males and females to mature (IDFG 2020 unpublished data; Appendix A). Note: the first mature hatchery-origin males were captured in 2020; were identified to the 1995 year class; thus, were 25 years old at time of capture. Sperm expression and motility confirmed sexual maturation of these males (n=2).

The Revised Recovery Plan also describes key elements of the recovery strategy. These include: 1) continuing to implement the conservation aquaculture program, 2) developing and implementing a long-term strategy for Libby Dam flow and temperature management to benefit Sturgeon, 3) continuing to implement nutrient addition programs, 4) restoring and enhancing Kootenai Sturgeon habitat, 5) continued RM&E, and 6) continued public outreach and education. The Revised Recovery Plan suggests that down-listing and de-listing would be appropriate when the following criteria are met:

Downlisting Criterion – Kootenai Sturgeon demonstrate consistent natural in-river production of juveniles, with production of wild age-3 juveniles occurring at an annual average of at least 700 individuals over 10 consecutive years. Production of 700 or more wild age-3 juveniles occurs in at least 3 of the 10 years, ensuring the annual average is not the result of an anomalous single-year event.

Delisting Criterion – The number of Kootenai Sturgeon wild recruits (offspring that survive to sexual maturity at 25 years of age) added to the adult (25 years or older) population annually averages at least 250 individuals per year over 10 years. In addition, the population includes at least 10,000 wild juveniles aged from 3 to 24 years.

Hatchery-origin fish do not count toward recovery goals under the ESA; only naturally recruited progeny count toward the downlisting and delisting criteria. Thus, given the next generation of Kootenai White Sturgeon are and will continue to be predominantly hatchery-origin, population recovery criteria is not likely to be satisfied for a minimum of 25 years into the future.

Field surveys and genetic analyses show that post-release, hatchery-origin Kootenai Sturgeon are surviving at levels sufficient to contribute to the future spawning adult population. The Revised Recovery Plan notes that due to the continued lack of in-river recruitment among Kootenai Sturgeon, continuing the conservation aquaculture program is vital to the recovery of Kootenai Sturgeon.

2.4 POPULATION OBJECTIVES

In addition to the recovery objectives listed in Section 2.3, the program has the following population objectives:

Abundance

- Sustain a hatchery juvenile and sub-adult (ages 3-24) abundance of 12,000-15,000 in order to achieve and sustain an adult abundance of 8,000 25+ year-old adults (hatchery and/or wild) through 2045 (KTOI 2007; KTOI 2010; KTOI 2012a; KTOI 2012b; KTOI 2018a; KTOI 2019a; USFWS 2019). For more background about the 8,000 adult recruitment abundance goal, please review KTOI 2020.

Population Structure

- Natural recruitment sufficient to support the desired adult population size.
- Hatchery Sturgeon generation(s) possess health and condition to reach sexual maturity providing sufficient reproductive potential to jump start and/or sustain natural recruitment.
- Stable or increasing trends in juvenile and adult numbers.
- Representative and stable size and age structure.

Distribution

- Distribution and use of habitats throughout the historical range.
- Breadth of distribution such that population is not vulnerable to any single human-caused catastrophic event (e.g., train derailments; chemical spills; etc).

Genetic Diversity

- Stable genetic diversity (based on common and rare allele frequencies).
- Effective population sizes adequate to allow for normal genetic and evolutionary processes.

Harvest

- Population numbers (consistent with above) adequate to support traditional uses, subsistence harvests, and a recreational fishery.

Selective Removal

The co-managers have a selective removal agreement with the following three objectives:

1) Remove spontaneous autopolyploids that survived post-release; the autopolyploid condition was not known about until 2011.
2) Remove a portion of over-represented year-classes, particularly 2004-2006.
3) Consider removal of individuals exhibiting poor growth and/or poor physical condition.

2.5 HABITAT OBJECTIVES

Changes to the Kootenai River ecosystem extend from physical habitat and ecological function loss to primary and secondary system productivity, nutrient availability, and possible contaminant dynamics (Northcote 1973; Ashley et al. 1997; Ashley et al. 1999; Anders et al. 2002; Schindler et al. 2011; KTOI 2020, 2021). Some factors such as harvest, levee construction, hydro development and altered environmental water quality factors are implicated; population collapse likely resulted from the combined impacts of these multiple factors. However, levee construction was completed decades before persistent natural recruitment failure; and the fisheries moratorium in place since 1983 has not resulted in any reversal of recruitment failure. Habitat alterations associated with Libby Dam construction and operations have had the most detrimental effects on the status of the Sturgeon population. The effects of Libby Dam combined with other factors discussed above, especially levee construction and diking have led to widespread loss of intermittent floodplain connectivity dynamics.

Forty years of research and experimentation has determined that recruitment continues to fail in the incubation and early larval stages because spawning continues in an area dominated by clay, clay rubble, and sand substrates, where embryos are smothered by fine sediment. It remains unknown why adult sturgeon choose to spawn in such unfavorable habitat. Whether spawning sites were historically suitable or fish previously spawned in more favorable locations is unclear. More importantly, we do not know what actions will restore post-spawn natural recruitment. All attempts to date to restore natural recruitment including modifications to flow in the Kootenai River have failed. Predation by other fish species (e.g., suckers (*Catostomus* spp.), peamouth chubs (*Mylocheilus caurinus*), etc.) may also be contributing to early life stage losses.

Growth rates of hatchery-origin Sturgeon in recent years appear to be lower than 20 years ago as shown by mean fork length at age of recapture (Figure 2; Hardy et al. 2020). Hardy et al. (2020) showed differences in fish reared on ambient river water as compared to those reared on warmer water temperatures in winter months (i.e., accelerated growth rearing strategy) at KTOI hatcheries. This was also apparent with fish reared on warmer water temperatures at the FFSBC facility (located near Fort Steele, BC); particularly the 2004–2006-year classes reared at this facility.

Fish captured in the mainstem Kootenai River and reared on ambient Kootenai River water have declined in length-at-capture over time, yet those captured in Kootenay Lake did not exhibit this reduction. Conversely, those fish captured in the river and the lake that were

reared on accelerated temperatures both exhibited little difference in growth. Hardy et al. (2020) also highlighted that fish recaptured in the lake were larger in fork length than those recaptured in the river. In addition, an evaluation of individual year class recaptures (e.g., 2009) suggested that those reared on ambient river temperatures have exhibited reduced growth over time. The trend of reduced fork length over time is also evident in age-9 hatchery-origin Sturgeon recaptured in the Kootenai River from 2000-2018 (Figure 4). There has not been an equivalent study evaluating growth rates of the wild adult population over time. In addition, the population size has leveled off despite increased numbers of hatchery releases in recent years (Figure 5). We speculate that this may be an indication the in-river population is fully utilizing available habitats and resources or showing carrying capacity density effects. In response to this, KTOI and co-managers agreed to initiate a removal program and reduce releases to 500 juveniles per female spawned starting in 2021. As of now, the overarching removal plan had not been finalized and agreed upon by co-managers; however, SA blood screening of juvenile hatchery origin fish will begin in 2023 to examine the prevalence of SA in the river/lake juvenile population.

Objectives for the Kootenai River White Sturgeon program related to habitat include:

1) Restore the conditions needed to allow for successful early life stage survival.

2) Hatchery Sturgeon removal (and limited hatchery releases) to begin addressing potential density dependence issues.

3) Initiate additional actions to improve ecosystem productivity in the Meander Reach (see KTOI *Project 198806400* 2020 ISRP project proposal for more detail).

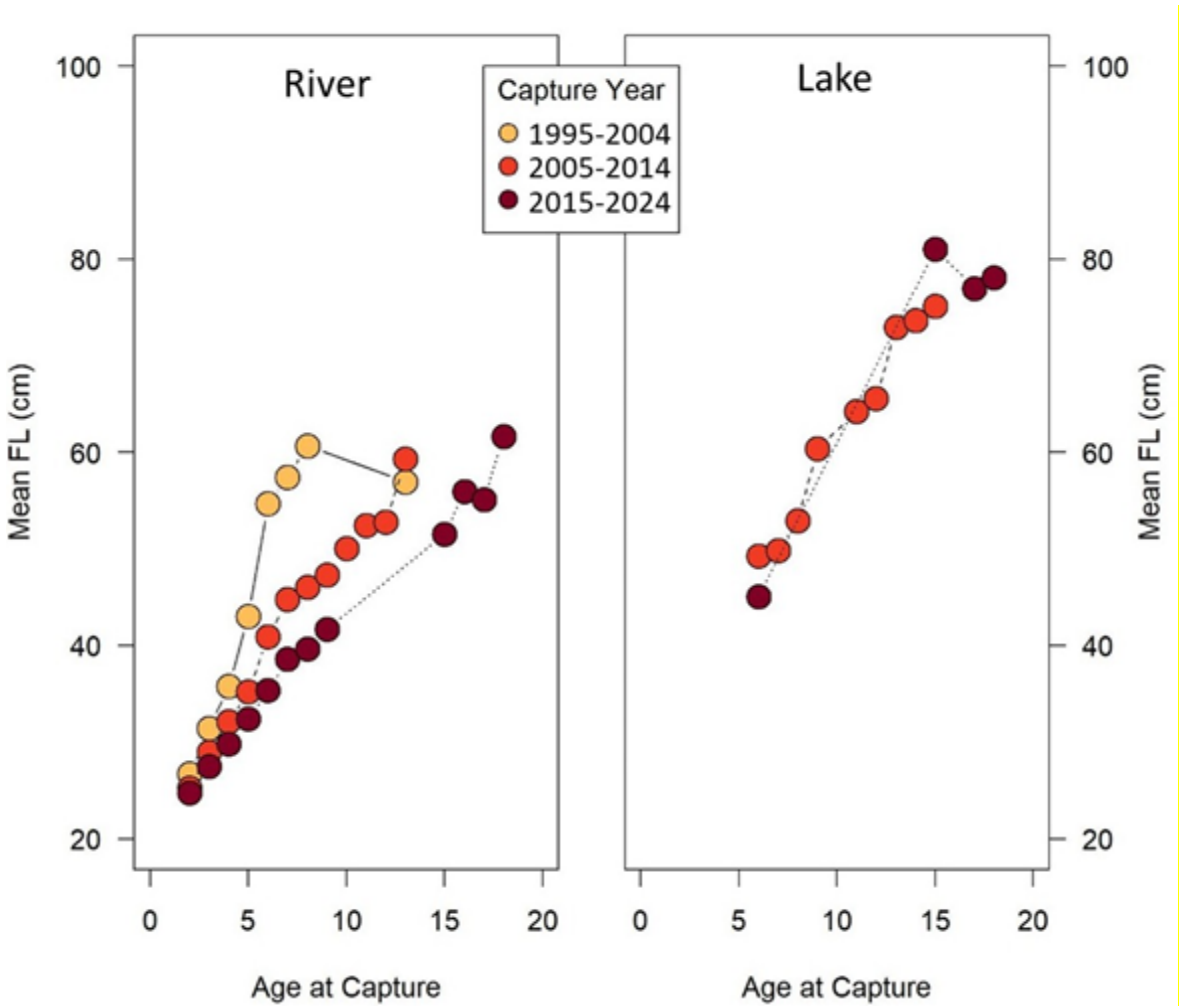

Figure 2. **Fork length (cm) of hatchery-origin Kootenai Sturgeon recaptured in the Kootenai River and Kootenay Lake from 1995-2019. The figures include fish reared on ambient river water released at age-1 (Hardy et al. 2020).**

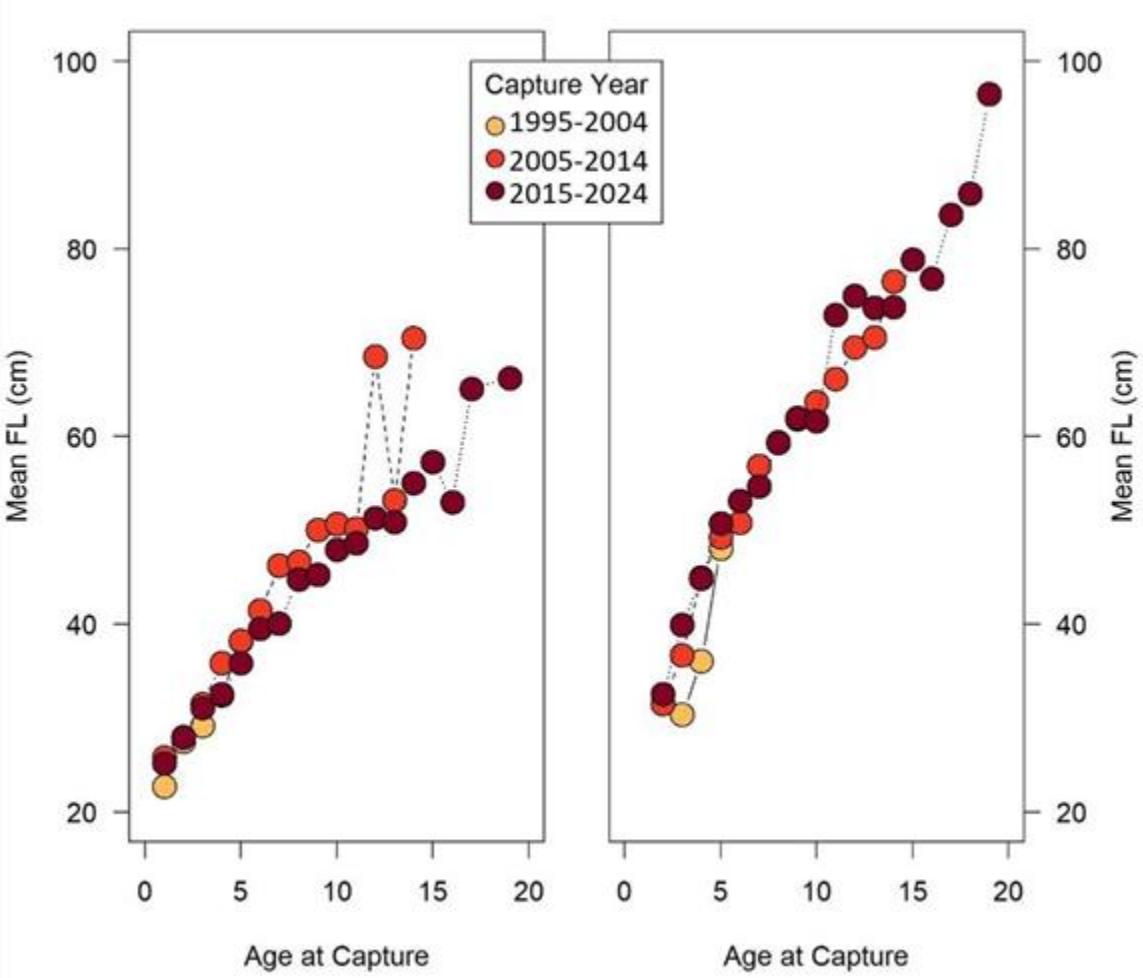

Figure 3. **Fork length (cm) of hatchery-origin Kootenai Sturgeon recaptured in the Kootenai River (left) and Kootenay Lake (right) from 1995-2019. The figures include fish reared on accelerated river water released at age-1 (Hardy et al. 2020).**

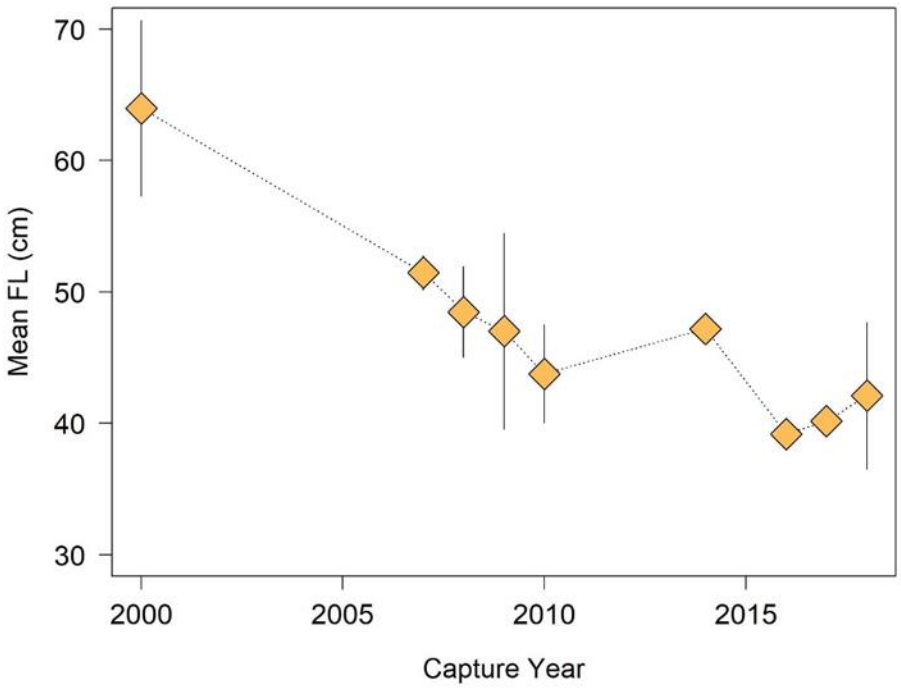

Figure 4. Fork length of age-9 hatchery Kootenai River White Sturgeon recaptured in the Kootenai River from 2000-2018. The figure represents fish reared on ambient river water and released as age-1 into the Kootenai River basin (Hardy et al. 2020).

3.0 CONSERVATION AQUACULTURE PROGRAM

The basis of conservation aquaculture is to conserve and recover imperiled fish populations, without harvest or focus on commercialized food production quotas. Conservation hatcheries are focused on producing sustainable age class structures in populations with limited or no natural recruitment, while optimizing remaining native genetic diversity. Conservation aquaculture involves incorporating local gene pools and allowing sufficient migration of genes into hatchery broodstock and progeny groups to enhance allelic representation for population viability and persistence. It requires careful selective breeding to provide sufficient diversity within a family-group, year-class or fish population. It necessitates eliminating as much artificial conditioning as possible. When successful, it provides the increased population base on which natural selection can operate. Because of its design, conservation aquaculture may reduce the commonly considered risks associated with high-density hatchery production programs, such as competitive feeding behaviors, reduced growth rates, domestication selection, and increased incidence of disease.

Table 3. Comparison of Kootenai River White Sturgeon, Burbot, and Kokanee reproductive and life history strategies (Ross et al. 2018; Hardy et al. 2020).

Reproductive and Life History Attributes	White Sturgeon (*Acipenser transmontanus*)	Burbot (*Lota lota*)	Kokanee (*Oncorhynchus nerka*)
Spawning type	Iteroparity Communal spawners Broadcast spawning	Iteroparity Communal spawners Broadcast spawners	Semelparity Paired spawners Redd-building
Individual fecundity (number of eggs per female)	100,000 – 1 million eggs	200,000/kg body weight	300 – 1,500 eggs
Generation length (Kootenai Population)	20-30 years	~ 2-10 years	4 years
Longevity (Species-wide)	> 100 years	~ 20 years	3-4 years
Age at first maturity (Kootenai Population)	20-30 years	2-3 years	3-4 years
Number of year-classes spawning together	Multiple	Multiple	1-2
Female spawning frequency	3-8 years	Annual	Once

Table 4. Life history strategies of White Sturgeon.

Life History Strategy	Importance to Conservation Aquaculture
Iteroparity	Multiple opportunities to pass gametes on to subsequent generations within a single lifespan
Overlapping generations	Increases between and among generation gene flow
Differential sex-specific age at first maturity	Reduces reproductive synchrony of male and female siblings
Differential sex-specific spawning periodicity; communal, broadcast spawning	Reduces reproductive synchrony of male and female siblings

The life history attributes (Table 3) and strategies (Table 4) of White Sturgeon are reflected in the hatchery broodstock collection and spawning plan. White Sturgeon are iteroparous; spawning multiple times during their lifetime, knowing this the hatchery program may use mature males and females as broodstock more than once on rare occasions. For production, eggs from each female are fertilized with milt from up to five males because in the wild, Sturgeon are communal, broadcast spawners, and eggs from one female may be fertilized by multiple males. Natural reproduction in viable Sturgeon populations is also

characterized by breeding individuals from many cohorts, year-classes, generations, and families. Spawning matrix design can partially incorporate this natural reproductive strategy by crossing fish of considerably different ages (sizes) and cohorts.

3.1 FUTURE PROGRAM OVERVIEW

The Sturgeon program will continue to be adaptively managed to address two major issues detected in recent years: 1) declining post-release survival and growth of hatchery-origin Sturgeon, and 2) spontaneous autopolyploidy, an abnormal ploidy level found in wild and hatchery-origin Sturgeon, with apparent occurrence higher in hatchery-origin cohorts (Schreier et al. 2021). M&E results indicate post-release survival of hatchery Sturgeon during their first year in the river from age-1 to age-2 has declined as hatchery juvenile/sub-adult abundance has increased (Dinsmore et al. 2015). Further, recapture data indicates declining and variable individual growth rates (Ross et al. 2015; Stephenson and Evans 2015; Hardy et al. 2020). These findings suggest the Sturgeon carrying-capacity of the altered ecosystem may have been reached and density-dependent effects are being observed. Since the Sturgeon program was initiated in 1990, the ecosystem has endured an additional three decades of human-induced ultra-oligotrophic conditions and variable hydro-operations resultant from Libby Dam operations. The situation now requires shaping the hatchery Sturgeon population structure to balance abundance, growth, and niche space to maintain a diverse standing-stock, which will continue to require adding future year-classes as recruitment failure persists. KTOI has agreed with co-managers to reduce annual year-class releases to 500 age-1 juveniles per family group and to implement a removal program in the river to remove spontaneous autopolyploids, portions of over-represented year-classes, and individuals exhibiting poor growth.

Removal of spontaneous autopolyploids will be the top priority of in-river removals. The condition was discovered in Kootenai Sturgeon during routine genetics monitoring of the 2011 year-class (Schreier et al. 2013). The number of abnormal ploidy Sturgeon released before 2012 is unknown, but it is likely ploidy fish survived and if so will be near maturity. Since 2012, management of the issue has evolved as investigators explored potential causes of spontaneous autopolyploidy (Gille et al. 2015; Van Eenennaam et al. 2020; Schreier et al. 2021) and developed accurate testing methods (Fiske et al. 2019, 2022, 2023). As of the 2019 APR, all broodstock candidates are verified to be 8N (normal chromosome number) using a protocol specific to KTOI before they may be used for spawning. Any 12N wild adult is released immediately back to the river and sperm samples from 12N males are not used. All family groups of juveniles produced each year are screened for spontaneous autoploidy (12N). For more detail, see Section 4.3.2.

Along with removals and reduced releases, additional habitat improvement actions are needed to improve ecosystem productivity in the Meander Reach, both in-channel and off-channel. Such actions are proposed through KTOI's *Project 200200200* and *200201100*. The Meander Reach supports the majority of both hatchery Sturgeon and Burbot because it has been a focal habitat for releases; in addition, some fish released in other locations have emigrated to the reach (Stephenson et al. 2013; Hardy et al. 2015; Ross et al. 2018; Hardy et

al. 2020). Both Sturgeon and Burbot spawn within the Meander Reach; Sturgeon at the uppermost area, and Burbot throughout (Ross et al. 2018; Hardy et al. 2020). Historically, the Kootenai Valley Floodplain supported abundant fish populations, and remains a frequently selected habitat even in its current degraded condition. Expanding in-channel nutrient additions to the Braided Reach as proposed by *Project 199404900* and to the Meander Reach as proposed by *Project 198806400* would connect the current Canyon Reach and Kootenay Lake nutrient additions currently implemented by *Project 199404900* creating a continuum of enhanced conditions through the recovery area. Adding in-channel structure, restoring off-channel habitats and connections, and re-evaluating spring flow management (timing and magnitude) are all actions needed in synergy to address recruitment failure and provide suitable habitat for all life stages, including hatchery fish that must survive, grow, and mature to perpetuate the next generations.

The KRNFCAP will continue to rebuild the Sturgeon population in a manner that (1) maintains genetic diversity, population structure, and abundance until natural recruitment is restored to a magnitude that is self-sustaining, and (2) maintains adult abundance above minimum adult spawning targets to support cultural and recreational harvest (KTOI 2007; KTOI 2010; KTOI 2012; KTOI 2012a; KTOI 2012b; KTOI 2018a; KTOI 2019a; USFWS 2019). At present, the long-term goal of restoring natural recruitment to self-sustaining levels that can also support fisheries has not been achieved. Empirical evidence, such as annual captures of only 0-3 naturally produced larvae (Ross et al. 2018; Hardy et al. 2020), indicate factors causing recruitment failure still to persist. Many factors will dictate if and when natural recruitment may be restored to self-sustaining levels; including, time to maturity of hatchery-origin Sturgeon; a declining wild adult population; persistence of wild adults spawning in the sand and silt dominated Meander Reach; hydro-operation variability; poor ecosystem productivity; the magnitude of habitat restoration needed; and contaminant leakage and subsequent food-web pollutant bioaccumulation associated with Elk River Valley coal mines and other anthropogenic sources.

3.2 PROGRAM GOALS AND OBJECTIVES

The overall short and long-term objectives of the conservation aquaculture program are listed in Box 2. In the short-term, the program's emphasis is to prevent extinction and preserve genetic diversity of the remaining wild population. In the long-term, the objectives are to ensure a viable population of mature, wild and/or hatchery-origin spawners with the potential to jump start and maintain natural recruitment if habitat actions restore appropriate conditions.

Overall Program Objectives

1. Prevent demographic extinction.
2. Establish an increasing trend and broad distribution of ages and sizes in the hatchery-origin and wild population to ensure future sustainability.

3. Preserve native genetic, phenotypic, and life history diversity by capturing and spawning the maximum allowable numbers of broodstock.
4. Provide contingencies for uncertain future availability of wild and/or hatchery-origin broodstock, and prospects for restoring natural recruitment.
5. Inform recovery strategies by using hatchery fish to identify limiting factors by releasing multiple life stages.
6. Avoid annual spawning stock limitation where too few fish might be available to capitalize on favorable natural spawning conditions in any year (or to continue to provide hatchery broodstock).
7. Minimize, to the extent possible, the time interval between the functional extinction of remaining wild adults and maturation of future hatchery fish generations.
8. Maintain an effective population size adequate to avoid genetic bottlenecks that risk loss of diversity or inbreeding depression in the next generation.
9. Prevent hatchery selection effects as much as possible that might reduce fitness/viability.
10. Minimalize spontaneous autoploidy in-hatchery and in-river.
11. Provide future harvest opportunities for tribal and non-tribal harvest/fisheries consistent with population persistence, viability, and conservation goals.

In addition to these overall program objectives, KTOI will adaptively manage hatchery production in collaboration with co-managers through Annual Program Reviews to achieve the specific goals and objectives listed below.

Population Objectives

Sustain a hatchery juvenile and sub-adult (ages 3-24) abundance of 12,000-15,000 to achieve and sustain an adult abundance of 8,000 25+ year-old adults (hatchery and/or wild) through 2045 (KTOI 2007; KTOI 2010; KTOI 2012a; KTOI 2012b; KTOI 2018a; KTOI 2019a; USFWS 2019). KRNFCAP has now released >30 year-classes resulting in a combined juvenile and sub-adult (ages 2-29) abundance of 12,000-15,000 (Dinsmore et al. 2015; Hardy et al. 2020). Consistent hatchery-origin year-class production is expected to maintain population structure as long as recruitment failure persists.

Broodstock Collection and Spawning Objectives

The two KTOI hatcheries (combined) typically spawn 10-12 females per year crossed with up to 40 males to create as many distinct female-based “family groups” as possible. An average of 30-36 unique families, or 8-10 “family groups” typically survive and contribute to each year class (KTOI 2018a, KTOI 2019a). The current program limits female egg collections to 15,000 eggs per female per hatchery (~30,000 total) for production. Eggs from each female are divided into groups based on the number of males available:

Example: If five males are available eggs will be divided into sub-groups of 3,000 eggs each and each is fertilized with sperm from a different male to create five individual families. The five families from each female spawned are subsequently re-combined post-fertilization to form a "family group".

Incubation and Rearing Objectives

Family groups are kept separate throughout incubation and rearing. Each incubator is loaded with fertilized eggs from a single-family group. Family group size targets at release are 250 fish per female per hatchery; or appropriately balanced between hatcheries based upon family group survival between hatcheries to target 500 juveniles total per family group. Regardless of total egg take per spawn, each family group is split so half of each family group is reared at each KTOI hatchery. This accomplishes two objectives: 1) it ensures all family groups are represented in case of catastrophic failure at one of the hatcheries, and 2) it allows KTOI to diversify rearing strategies if needed which may affect post-release survival for each family group. Current strategies include accelerating growth by warming water in winter to maximize size-at-release, accelerating feed size transitions to drive fish onto larger sized feed faster to improve water quality with less waste feed and the season-of-release is now only Spring because it has been shown to increase post-release survival (Dinsmore et al. 2015; KTOI 2018a, KTOI 2019a).

During winter months, half of the juvenile year-class is reared at Twin Rivers Tribal Hatchery (Hatchery 2) on heated groundwater and the other half are reared at the Tribal Sturgeon Hatchery (Hatchery 1) on heated Kootenai River water for "accelerated growth". As of 2021, both hatcheries are now applying accelerated growth rearing methods exclusively. Hatchery 2 capabilities include two surface water sources (Kootenai River and Moyie River) as well as a groundwater source; Hatchery 1 only has one water source, the Kootenai River. Groundwater is the preferred water source to heat efficiently and affordably through winter because of its stable temperature and water quality. Applying accelerated growth methods using ambient river water with fluctuating temperatures has been more expensive and variable at Hatchery 1.

Thus, no more juveniles will be reared at ambient water temperatures at Hatchery 1 mimicking what a 'naturally produced' Sturgeon would experience in the river unless decision makers decide differently. The in-river population has lived in an altered thermograph for approximately 50 years and will do so into the foreseeable future. The overall long-term effects of ambient river water temperature fluctuations on Sturgeon physiology at this time appear to be negative; however, given that Libby Dam will be operating into the foreseeable future, Sturgeon will need to adapt.

Release Objectives

The program is required to mark Sturgeon prior to release. For juvenile hatchery sturgeon lateral line scute removal is used as a physical mark denoting the year spawned and used to show which hatchery the individual was reared in based on which side the scutes are removed (e.g., Right = Tribal Hatchery 1; Left = Tribal hatchery 2). PIT-tags are also used if

the individual juveniles are large enough to safely inject the tag. Release numbers of juvenile hatchery origin fish is ≤500 per spawn per year; or as agreed upon by co-managers. However, juvenile release numbers are in addition to early life stage (eggs or sac fry larvae) releases. Early life stage releases began in 2022 and may occur if genomic parentage-based tagging is used to 'mark' the fish. Read below for additional information on Parentage Based Tagging or PBT.

Prior to release, all juvenile family groups are randomly subsampled (n=30) to check chromosome bundle size or "ploidy level". A White Sturgeon typically has eight sets of chromosomes. A sturgeon with eight sets of chromosomes is 'normal' or referred to as '8N ploidy'. The N stands for one chromosome set. Abnormal ploidy levels are 10N or 12N. For the KTOI conservation aquaculture program only 8N (normal) and 12N (abnormal) ploidy is screened for because 10N ploidy detection requires specialized equipment for flow cytometry.

Any family group determined to be ≥50% 12N ploidy level is not released and the entire family group is euthanized. If a family group has <10% 12N ploidy all the ≤500 juveniles from that family group may be released. This is done in part to save costs and reduce waste associated with blood screening but also because the probability of a 12N fish from a family group with <10% surviving to maturity is very low. However, if ploidy level is determined to be between 10% and 50% the entire family group may be screened and only 8N individuals will be selected for release. Again, all fish tested and verified to have abnormal ploidy levels (12N) and all normal 8N juveniles exceeding 500 per family group will be destroyed unless it is agreed upon to use the excess individuals for other purposes (e.g., cultural uses, aging structures, contaminants toxicity tests, transport to other habitats, education, and outreach (KTOI 2019a)).

Parental-based Tagging Objectives

A more accurate Parental Based Tagging (PBT) program has been developed in collaboration with the University of California-Davis Genomics Variation Lab (Lead - Dr. Andrea Schreier, subcontracted by KTOI *Project 198806400*). Previously, Schreier et al. (2012) developed parental assignment methods that were deemed not sufficient in accuracy by some co-managers. There are unique challenges associated with PBT analysis of Kootenai Sturgeon versus other White Sturgeon populations due to their low population genetic variability, and PBT analysis of White Sturgeon in general caused by their extremely large genome, which includes eight replicates of their ~240 chromosomes (Schreier et al. 2013). Now that a Kootenai River White Sturgeon PBT protocol is completed and its accuracy verified, early life stages of Kootenai Sturgeon will be released at specific locations and times to evaluate recruitment dynamics and early life stage ecology across the recovery area. PBT programs utilize the known genetic profiles of broodstock spawned to monitor surviving progeny. Entire year classes may be included in a PBT program (similar to KTOI's Burbot program); alternatively, a smaller, targeted PBT program may include targeted broodstock and their progeny. At this time, KTOI will use Sturgeon PBT for targeted uses; and then expand to year class-wide monitoring in the future if it is feasible to do so. The data gleaned from

recaptured progeny, including physiology and habitat use data, will be used to test hypotheses about recruitment dynamics and early life history eco-physiology otherwise impossible to gather under the current conditions of little to no natural recruitment.

To use PBT as a marking technique it is critical that each parent of each released fish or group is genotyped and catalogued to create a reference list of parents to compare to. If a physically unmarked fish is encountered post-release a tissue sample can be used to compare back to the established parental reference list to determine if it's a match. Explicit family group segregation must be done in-hatchery to keep family groups unique from one another and when a group of fish is released at a size too small to physically tag additional care must be taken to only release in defined locations and discrete life-stages (e.g., egg stage or larval stage but not both).

More information on PBT is provided in Section 4.2.3.

Spontaneous Autopolyploidy Objectives

Spontaneous autopolyploidy (SA) was discovered in Kootenai Sturgeon during routine genetic monitoring of the 2011 year-class (Schreier et al. 2013). It remains unknown how many spontaneous autopolyploids were released prior to 2012. It is likely some pre-2012 year-class SA fish have survived (KTOI unpublished data). To create niche space for future year-class contributions and to remove 12N spontaneous autopolyploids, in-river removal planning and implementation will be conducted by *Projects 198806400* and *198806500* (KTOI 2019a). See Selective Removal Objectives below.

Paramount to all spawning protocols include broodstock candidate screening for 12N SA; or rather verification that all candidates are normal 8N ploidy before they are included in the spawning program. Blood collections are analyzed for SA using a Coulter Counter. Currently, all testing is completed by KTOI staff. No 12N adult Sturgeon will be included in spawning matrices; however, any 12N 'wild', unmarked, unencountered male or female adult is released back to the river unharmed. If this occurs, it will be recorded in the master database per co-manager agreement.

All juvenile family groups released will be verified ≥ 90% 8N normal ploidy level. All detections of 10N or 12N fish may be euthanized. Juveniles are tested prior to release using methods discussed in Fiske et al. (2019). All testing is completed by trained KTOI staff. KTOI staff screen adults to ensure no 12N fish are spawned again in the future. See Section 4.2.2 for details on screening protocols.

Selective Removal Objectives

Although significant program interim goals have been achieved (e.g., capturing >90% of the wild Kootenai River Sturgeon allelic/genetic variation, hatchery origin releases are now being used as broodstock), a decline in hatchery-origin sturgeon age-1 survival and variable growth rates of juvenile sturgeon are concerning (KTOI 2018a; KTOI 2019a). Co-manager M&E program data suggest this may be due to density-dependent effects; however, poor ecosystem productivity and pollution from upstream mining operations may also be

contributing to declines in survival, growth, and reproductive/recruitment success. Much more investigation is needed with regards to pollutant effects on Kootenai Basin Sturgeon; however, extensive M&E modeling of population abundance has been done; and redone, related to potential density-dependent effects that may be evident. The density-dependent hypothesis is proving to be a valuable sticking point to begin discussions about developing selective removal agreements that may be used to re-shape the standing stock of hatchery origin sturgeon. If selective removal plans are enacted than managers may be able to address density effects in the short-term but also address a long-term problem inflicted by mature hatchery origin fish with chromosome abnormalities (i.e., removal of spontaneous autopolyploids (12N)). To begin selective removal these are the main objectives:

1) Identify the prevalence of 12N hatchery Sturgeon currently in the river.
2) Record fish via PIT tag code(s) that may be candidates for removal.
3) Selectively remove 12N fish to begin to reduce density in river (these removals may also be used to investigate fish tissue pollutant concentrations). *Note, the 12N problem was not recognized until 2011; approximately two decades of unscreened releases occurred prior to 2011.*
4) Potentially remove portions of over-represented year-classes (e.g., 2004-2006).
5) Potentially remove juvenile fish exhibiting sub-optimal growth.
6) Investigate translocation as a potential alternative to culling fish selected for removal to improve growth.
7) Investigate the possibilities of creating a 10j population as an alternative to culling select fish.

Habitat Restoration Objectives

To improve age-1 survival and growth of a substantial number of hatchery fish and any wild recruits, *Project 198806400* proposes to systematically add organic matter to the Meander Reach through 2025. *Project 199404900* has been adding nutrients in the upriver Canyon Reach and into Kootenay Lake; however, the positive effects have been predominantly localized. *Project 199404900* may also initiate a second nutrient site that may benefit the Braided Reach between the Canyon and Meander Reaches. If the Braided (via *Project 199404900*) and Meander Reaches (via *Project 198806400*, *200200200*, and *200201100*) overall productivity increases through various methods, a more representative river-continuum may benefit these reaches on a biologically meaningful scale and better support hatchery and wild fish. Thus, expansion of nutrient and plant material additions into the Meander Reach will hopefully spur higher productivity of prey items for larval, juvenile, and sub-adult life stages of Sturgeon and other species. Most hatchery fish are released and/or move to the Meander Reach after release. Also, natural recruits will likely be spawned and transported to these areas during early life. *Project 199404900* has long-standing monitoring sites that *Project 198806400* will continue to monitor and will reference for treatment comparisons. In combination with the substantial habitat restoration actions

proposed by *Projects 200200200* and *200201100* and associated actions, this proposed objective may help alleviate density-dependent effects.

Since 2009, Libby Dam hydro-operations have been shaped from mid-May through late June to better support adult migration, spawning behavior, and early life stages. On an annual basis, a team of co-manager agency representatives led by USACE develop a flow plan based on Libby Dam volume and local snowpack runoff amount and timing. The plan has been experimental, as different flow shaping strategies have been implemented.

Natural Production Objectives

Please note that hatchery-origin Sturgeon do not count toward ESA delisting or down listing criteria, only naturally recruited individuals are considered for recovery plans. However, a naturally recruited offspring of hatchery-origin parents does count for ESA considerations.

To date, Sturgeon natural recruitment is negligeable. Only a small number of wild, unmarked, unencountered Sturgeon are captured annually in gill-net surveys, and few larvae are captured post-spawn (Ross et al. 2018; Hardy et al. 2020). Through 2030, if Kootenai River White Sturgeon adults (whether current wild adults, future hatchery-origin adults, or a combination) demonstrate consistent natural in-river production of juveniles, with production of wild age-3 juveniles occurring at an annual average of at least 700 individuals over 10 consecutive years. If production of 700 or more wild age-3 juveniles occurs in at least 3 of the 10 years, ensuring the annual average is not the result of an anomalous single-year event, the population will meet the Downlisting Criterion in the Revised Recovery Plan (USFWS 2019). If this is observed, the KRNFCAP will adaptively manage hatchery production in collaboration with co-managers.

Through 2045, the number of wild recruits (offspring that survive to sexual maturity at 25 years of age) added to the adult (25 years or older) population annually averages at least 250 individuals per year over 10 years. In addition, the population includes at least 10,000 wild juveniles aged from 3 to 24 years. If the population meets these objectives, it will achieve the Delisting Criterion in the Revised Recovery Plan (USFWS 2019). If this is observed, the KNFCAP will adaptively manage hatchery production in collaboration with co-managers.

3.3 STATUS AND TRENDS

Status and trends represent actual program outcomes and will be used to evaluate key assumptions and program performance (e.g., did we meet the biological objectives? do the targets need to be modified?). This information is typically reported at an APR and incorporated into this Plan, and will also be shared with the public and other management entities. This section of the Plan will include an overview of the most recent data on Kootenai Sturgeon Population Status, Broodstock Collection and Spawning, Incubation and Rearing Outcomes, Release Strategies, and Post-release Survival. Multiple agencies

contribute data on an annual basis; however, not all metrics/data updates are on the same timeline.

3.3.1 Population Status

IDFG tracks adult abundance (wild and hatchery) and age structure of the Sturgeon population in the Kootenai River. Revised estimates of the wild adult Kootenai Sturgeon abundance suggest the remaining wild adult population is larger (approximately 1,744 as of 2017) and annual survival is higher (≈ 96%) than previously reported (Figure 5; Hardy et al. 2020). In 2020, two male hatchery-reared Sturgeon; from the 1995 year-class, were captured during the spawning season at known spawning locations expressing milt. One male was used to fertilize eggs as part of KTOI's 2020 and 2021 activities (Appendix A). However, population monitoring also indicates that natural recruitment remains very low due to high levels of mortality still occurring in early life stages.

The program has achieved the immediate objectives of producing multiple juvenile age-classes and forestalling demographic extinction (Figure 6). Post-release monitoring conducted by IDFG, BC Ministry, and MFWP has demonstrated that a substantial standing stock of hatchery-reared fish (15,000-20,000 in recent years; Figure 6) has successfully adapted to current river conditions. If enough hatchery-produced fish continue to survive, grow, and mature, the hatchery program will have bought time to implement the large-scale ecosystem improvements necessary to restore natural production and long-term population sustainability. In 2019, annual hatchery release numbers were reduced to <10,000 age-1 juveniles. Previously, up to 37,000 juveniles were released annually. First year mortality averaged approximately 8-10% for the most recent 10 years of mark-recapture data (Figure 6; Hardy et al. 2020).

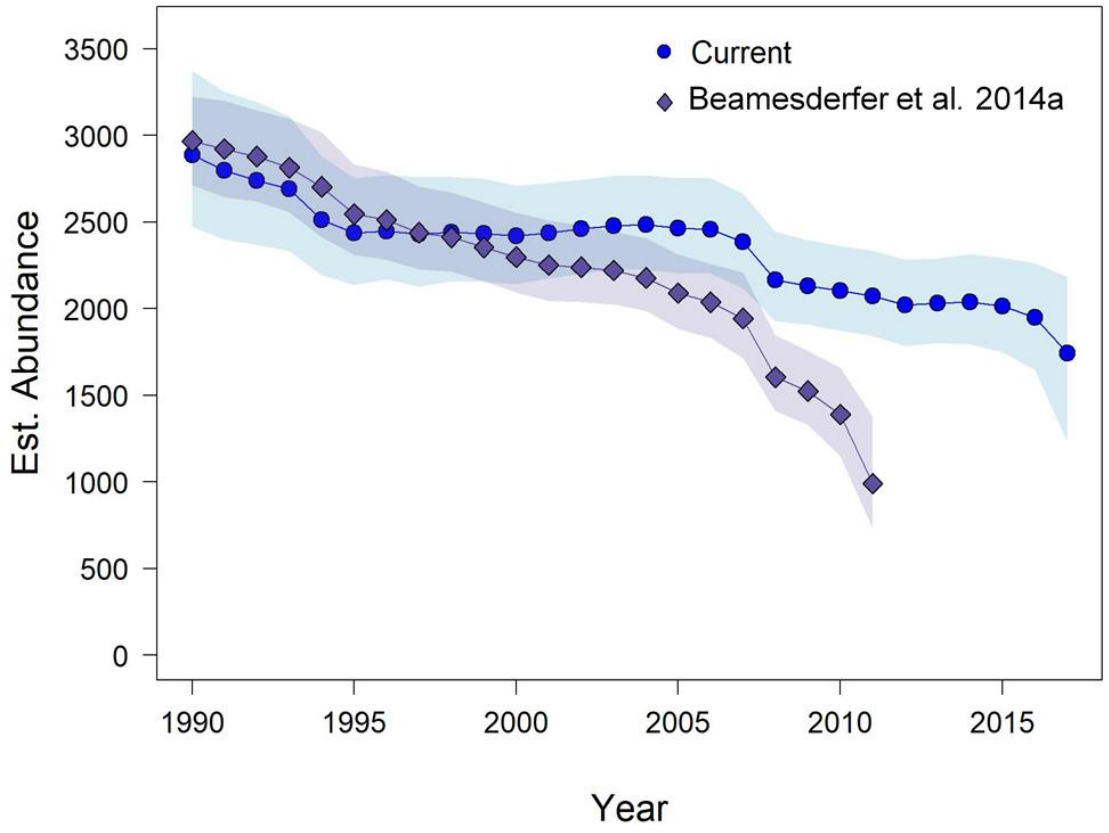

Figure 5. Comparison of current adult Kootenai Sturgeon population abundance (Hardy et al. 2020) and those reported by Beamesderfer et al. (2014). Values are the mean and 95% credible interval (Hardy et al. 2020).

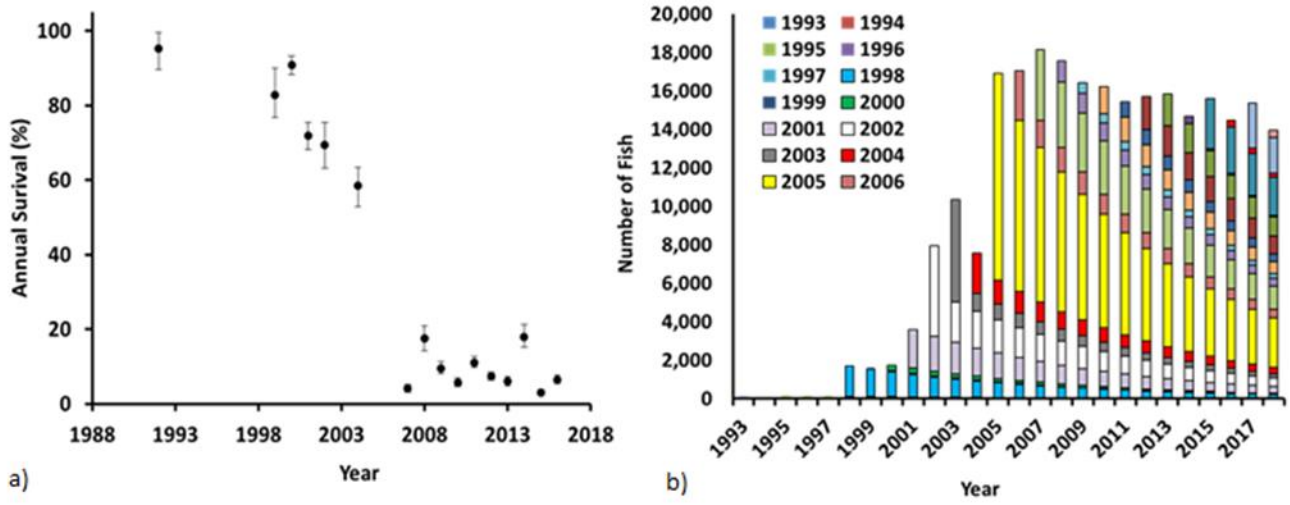

Figure 6. Age-1 annual survival (a) and abundance (b) of juvenile hatchery fish by year-class in the Kootenai River from 1993-2018 (Hardy et al. 2020).

Genetic analysis of wild Kootenai River White Sturgeon broodstock and their surviving hatchery-reared progeny provides a population-level indicator of how well the program is incorporating wild population genetic attributes into the next generation; which will be predominantly hatchery-origin. Broodstock genetic variability (frequency distribution of alleles) and genetic diversity (total number of alleles) is monitored annually. All broodstock spawned and all progeny groups produced in the hatchery are analyzed using microsatellite DNA methods that have become widely used for many conservation and management applications due to their high resolution and highly variable nature (McQuown et al. 2000; Rodzen and May 2002; Rodzen et al. 2004; Drauch and May 2007, 2008, 2009).

Microsatellite analysis by Rodzen et al. (2004) found that the wild Kootenai River White Sturgeon population is approximately 25% to 50% less diverse than eight other North American White Sturgeon populations. Similar results are reported by Drauch-Schreier et al. (2011), as shown in Figure 7. This is not surprising, given that the Kootenai population is a headwater population at the edge of the species' geographic range and has likely experienced a natural loss of rare alleles since post-Pleistocene re-founding due to genetic drift. Further artificial loss of native diversity has likely occurred due to recent anthropogenic and demographic changes and associated genetic bottlenecks.

To date, >400 Kootenai River White Sturgeon adults have been genotyped by the University of California (UC) Davis Genomics Variation Lab (GVL). The annual spawning plan captures approximately 80% of the genetic diversity from the existing wild spawning classes, and overall has captured 96% of the genetic diversity of the wild population (Schreier and Van Eenennaam 2019).

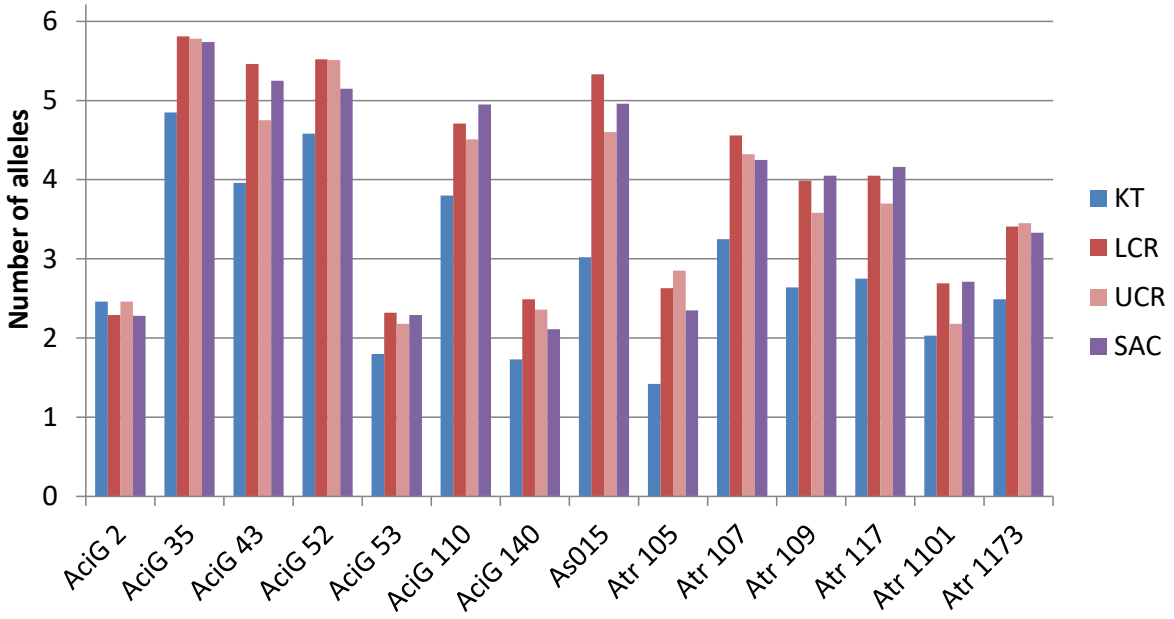

Figure 7. Number of alleles identified for 14 loci in Kootenai, Lower Columbia, Upper Columbia, and Sacramento River White Sturgeon populations (Drauch Schreier et al. 2011).

3.3.2 Broodstock Collection and Spawning

Female broodstock collection in the Kootenai River typically occurs from April through June, depending on annual water temperature and flow patterns. In-hatchery spawning typically occurs in June. Initial broodstock collection efforts are focused on females in later stages of vitellogenesis (ova development) for spawning. Female broodstock candidates with ova typically >3mm, dark colored with an apparent 'bulls-eye' are considered ripe for spawning; however, ova development is carefully monitored in the hatchery to ensure the germinal vesicle within the ova are in the correct proximity for ovulation/fertilization. In general, when an adult females' germinal vesicles within her ova are near the cell wall, she is stimulated to luteinize and release her ova with a hormone injection (Conte et al. 1988, KTOI 2016, KTOI 2021b). More specifically, ova maturation is measured by applying a Polarity Index value (PI value). Briefly, ova samples are boiled, bisected and the location of the germinal vesicle or polar body (n=10) within the ova are measured for the distance to the chorion or shell relative to the total diameter of each ova measured. If a female is determined to be ready for spawning, and appropriate quantities of milt and milt samples have been procured, the female is then injected with two doses (a priming (10%) and a resolving dose (90%)) of a Luteinizing and Releasing hormone analog (LHRHa) calculated based on the individuals body weight to stimulate ovulation. It takes approximately 36 hours after a priming dose for a female to release ova (see KTOI 2021b).

Milt (sperm) collection from male broodstock is a critical component to successful spawning and maximizing genetic contributions for each year class produced. It is not uncommon for milt collections and milt viability to be the primary limiting factors for spawning success. If no milt is available, no females will be induced to spawn. Males are seldom transported to the hatcheries, so collections are typically done under field conditions. Since the mid-1990s, milt has been successfully collected in the field from flowing males. Milt expressed from flowing males in the field can be preserved for several days. Throughout the spring broodstock sampling season, field crews monitor male ripeness to gauge male gamete availability for use in the hatchery across the spawning window from mid-May through June. All milt samples transported to the hatchery are monitored daily to evaluate sperm quality and viability (e.g., density, percent motility, motility duration).

The Kootenai program typically divides the eggs from individual females into separate batches, which are fertilized individually with milt from three to five males in order to maximize fertilization, survival and genetic contribution. A family is defined as a 1:1 female:male cross. Individual families are pooled post fertilization to create a family group. The Sturgeon protocols (KTOI 2021b) were changed in 2019 in a manner similar to the Burbot program (KTOI 2021a, 2021c), "family groups" are reared separately. A family group is defined as all offspring from one female. By doing so, the Sturgeon program can rear

unique families and/or family groups to ensure robust PBT implementation when PBT is applied.

To date, the Program has successfully rebuilt a healthy age-class structure by spawning approximately 450 wild adults (~150 females crossed with ~300 males) and creating approximately 350 unique families across ~30 year-classes during 1990-present (Table 5). The annual spawning plan captures approximately 80% of allelic variation (genetic diversity) from the existing wild spawning classes, and overall has captured 96% of the genetic diversity of the wild population (Schreier and Van Eenennaam 2019).

Broodstock and Spawning Objectives

The Kootenai Tribal hatcheries are permitted to annually spawn up to 20 females; however, 15 females are the annual target based on logistics. Females may be crossed with up to 40 males to create up to 40 unique families and comprise up to 15 distinct female-based "family groups". Although 15 distinct family groups are possible, typically 30-36 unique families, or 8-10 "family groups" survive each year and are used to contribute to year classes (KTOI 2018a, KTOI 2019a). An average of ~30,000 eggs are collected from each female for hatchery production and an additional 50-100,000 may be collected for early life stage releases. Only 8N, verified normal chromosome ploidy fish are used as broodstock.

The current program objectives are to maximize the number of broodstock and family groups used in the program. The 30+ family/year goal may not be met every year due to variable environmental conditions, success in fish capture, spawning, incubation and early life rearing.

Table 5. Summary of Sturgeon program broodstock collection, egg take, and hatching success from 1990-present.

	Males	Females			Egg takes (Thousands)			Egg-larval Survival	
Year	Brood	Held	Brood	Families	Total	Mean	Range	Mean	Range
1990	1	1	1	1	60	60	--	2%	--
1991	3	2	1	1	69	69	--	20%	--
1992	3	2	1	3	142	142	--	16%	Na
1993	2	2	1	2	86	86	--	21%	Na
1994	0	0	0	0	0	--	--	--	--
1995	4	2	2	4	143	71	71–72	28%	--
1996	2	2	1	2	62	62	--	<1%	--
1997	5	4	3	6	201	67	40–97	30%	Na
1998	6	3	3	6	217	72	60–92	28%	Na
1999	8	5	4	8	277	69	38–105	63%	40–80%
2000	11	6	6	11	306	51	17–112	73%	25–92%
2001	10	8	5	10	294	59	51–69	70%	35–86%
2002	9	6	3	9	151	50	34–62	86%	50–97%

	Males	Females			Egg takes (Thousands)			Egg-larval Survival	
Year	Brood	Held	Brood	Families	Total	Mean	Range	Mean	Range
2003	13	8	4	13	246	61	56–74	93%	85–99%
2004	11	13	5	17	369	74	60–98	81%	15–95%
2005	14	13	6	16	1,163	108	17–255	78%	15–97%
2006	15	11	7	11	79	113	54-164	88%	66-100%
2007	18	8	5	18	289	58	38-85	94%	72-98%
*2008	19	12	11	17	1,070	97	53-162	89%	60-99%
*2009	17	11	9	12	1,025	114	70-179	95%	60-99%
*2010	15	15	11	17	1,487 410	25	20-30	90%	48-96%
*2011	9	7	7	16	680 232	15	10-20	90%	80-99%
*2012	12	13	9	17	828 228	15	10-20	95%	90-99%
2013	10	12	9	17	221	13.5	10-17	85%	20-98%
2014	14	10	8	17	263	15	12-16	85%	0-99%
2015	25	16	14	27	401	20	14-27	81%	10-95%
2016	27	13	9	29	152	7.3	7-8	79%	37-98%
2017	22	11	7	29	183	7	5-8	92%	57-99%
2018	26	12	10	36	300	8.4	7-9	86%	50-99%
**2019	28	10	8	40 8	128	3.5 16*	3-4 15-19	71%	0-89%
2020	No Year Class - Research only due to COVID-19								
2021	26	16	14	13	339	24	15-45	73%	36-90%
***2022	25	11	10	8	724	90	61-128	81%	55-94%
2023	40	15	14	12	1,324	95	5-148	62%	0-97%
Totals	450	280	208	444 40**	14,149	80	0-255	0-95%	0-100%

* 2008-2012 egg takes also for co-manager yolk-sac larvae releases.
** Switch from families to family groups (#'s). A family group consists of 3-5 males crossed with each female.
***Egg takes for egg and larval releases included.

3.3.3 Incubation and Rearing Outcomes

Incubation and rearing occur at both hatcheries in designated areas. Incubation and rearing methods are described in detail in KTOI 2021b (Chapter 4 – Sturgeon Incubation and Chapter 5 – Sturgeon Rearing). In recent years, KTOI has been evaluating fertilization and egg incubation techniques that are more natural in hopes that early life physiology may benefit, and spontaneous autopolyploidy occurrence may be minimized (see Appendix A). As noted above, post-fertilization, deahheasion, each family group is split between KTOI

hatcheries; half of each family group is reared at each hatchery to provide a safeguard against catastrophic loss due to mechanical failure or some unforeseen circumstance (e.g., train derailment in 2020).

The conservation aquaculture program established family size targets of 1,000 - 1,500 yearlings from 1994 through 2003 (KTOI 2004) and 2009 to 2012. This range accommodated differences in in-hatchery survival of family groups due to variation in egg fertilization and survival to release. Family size targets were established to: 1) ensure survival of a representative number of offspring from each family to an age where they can spawn and contribute to the next generation, 2) avoid excessive contributions from any one family that might swamp population genetics, and 3) limit total population size in order to limit intra-species competition and density-dependent growth and survival limitations.

With the construction of the Twin Rivers facility, family size targets were reduced to 500 - 1,000 yearlings. More recent, beginning 2019, family/family group size targets are now 150 - 200 juveniles per family, or 400 – 500 fish per family group. This reduction was intended to balance the need for increased broodstock numbers to optimize phenotypic expression while also controling total release numbers which might trigger a negative compensatory response at higher densities.

Large family sizes may be better from a genetic and/or phenotypic diversity expression point of view because it increases the probability offspring from each family will survive to reproduce. However, co-managers are cognizant of habitat capacity limitations and the potential negative demographic responses to releasing too many fish.

3.3.4 Release Strategies

Release strategies have changed repeatedly since the program started releaseing fish in 1992. Since 1992, >320,000 hatchery-produced Kootenai White Sturgeon juveniles have been released in the Kootenai River and Kootenay Lake (Table 6). All hatchery releases prior to 1997 were considered experimental. Expanded release numbers began in 1997 after the hatchery program was identified as a critical component of the Recovery Plan.

Production was increased in 2003 after hatchery upgrades. Annual releases ranged from 3,000 to ~40,000 fish per year from 2003 to 2019. Currently, ≤500 age-1 juveniles/female are released from Tribal hatcheries given the ESA Section 10 permit limitation of 20 total females may be spawned per year. Taken together, a total of 10,000 juveniles is the most that will be released based on the co-manager agreement to limit each release group produced to 500 per female coupled with the the ESA Section 10 limitation; however, the annual female broodstock used for production is typically 8 to 12 per year.

Prior to 2005, fish were released at age-1 or older to be able to PIT-tag every hatchery fish so they could be distinguished from wild/unmarked juveniles. KTOI prefers fish to be at least 20 g for PIT-tagging. Beginning in 2005, fish were released at younger ages and smaller sizes to increase production numbers while working within the rearing limitations of

the Tribal Sturgeon Hatchery. Concerns about ongoing natural recruitment failure led KTOI and co-managers to to this agreement. At that time, fish were being released at 10-15 g as Age-0+ in fall rather than ≥30 g at Age-1+. This strategy avoided space limitations in the existing hatchery caused by rearing overlapping brood years. Minimizing time in the hatchery was also expected to minimize opportunities for hatchery selection effects and unforeseen rearing catastrophes (disease, equipment failure, etc.). This strategy was believed to be a failure until 2012-2014, when these fish started to recruit to gill net surveys in large numbers.

In most years, production was constrained by the requirement to raise all fish to sizes suitable for PIT-tag placement and retention, and to rear families separately so that family sizes could be equalized within an order of magnitude upon release. Subsequent evaluations concluded that a low population size in the next generation is a much more acute demographic and genetic risk than unequal family contributions in the following generation. Thus, some fish to small for PIT-tagging were allowed to be batch marked for several years. Batch marking of fish with scute removal patterns allows a smaller size at release while preserving a means of distinguishing hatchery-reared fish in the wild. Eliminating the PIT-tag requirement also provides flexibility to release fish at smaller sizes and ages which opened up space for more family groups in the hatchery. Upon release, smaller fish were expected to survive at similar annual rates as those observed in previous groups, although an extra year of natural mortality means that fewer fish from any release group would be expected to survive to a given age.

However, subsequent monitoring found that survival of the more recent release groups has declined from the early estimates (see Figure 3; Hardy et al. 2020). The decline was most pronounced among the smaller size fish (< 25 cm) while survival of larger hatchery fish was like previous estimates. This negative relationship between release numbers and survival suggested that density-related competition or predation may be influencing mortality of juvenile Sturgeon during their first-year post-release. However, this effect appeared limited to the first-year post-release, as indicated by the relatively stable survival rates for fish recaptured two or more years following release.

Survival of juveniles from the 2004-2006 year-classes, released at smaller sizes, was initially underestimated. Releasing younger, smaller fish delayed their susceptibility to being captured during gillnet surveys. Once the survivors from these releases were recruited to the survey gear, their survival rates were higher than anticipated. The benefit of this adaptive experiment was the identification of a second life history bottleneck concerning growth during the first year of life that may affect hatchery fish and presumably wild recruits as well. The effect of KTOI's habitat restoration measures on this first-year bottleneck will be one of the outcomes monitored. Recent M&E results have shown that the large releases in year-classes 2004-2006 are now overrepresented. As a result, fish from these year-classes may be targeted for selective removal as part of the fish restoration program.

Starting in 2019, KTOI and co-managers agreed to reduce the number of yearlings released from each family-group to 500 juveniles per group. The standard program objective is to strive to create at least 30 families, 10 family groups (e.g., 1 female x 3 males) per year. Given current age-specific survival rates, this would result in recruitment to age-25 of two fish per family, or 6-8 per family group; however, projections are extremely sensitive to small differences in estimates of annual survival. For instance, increasing the survival assumption by 3% per year triples the projected number of recruits. Decreasing the survival assumption by 3% per year reduces the estimate by two thirds, and eliminates year-class contribution. Moreover, small differences in initial annual survival have a big effect on long-term survival and the resulting age class structure due to compounding effects over many years. It should be noted that an individual's size-at-release will affect probabilities of post-release survival. Size at release has also varied widely since the first releases in 1992. The present goal of the hatchery program is to maximize size at release by incorporating 'accelerated growth' practices (e.g., heated water in winter, advanced feeding strategies, low density rearing and extensive culling of slow growing and/or abnormal fish) applied to each 500/female family groups.

Release Objectives

Releasing Age-1 and older fish has been the standard method for rebuilding the population. The program goal is to release 500 age-1 Sturgeon per family group produced each year from the two Tribal hatcheries (250/hatchery/family-group). This plan assumes 250 fish from each hatchery from each family group; however, it is likely that hatchery contributions will not be realized given the inherent annual variability that occurs with regards to environmental changes, hydro operations, and variable hatchery practices.

Table 6. (A) Numbers of hatchery-produced White Sturgeon released into the Kootenai River and Kootenay Lake in Idaho, Montana, and British Columbia, 1992-present. (B) Early life stage releases, 2008-present (KTOI data provided to co-manager database).

(A)

Year class	Rearing facility	Release number		Mean total length mm (SD)	Mean weight g (SD)	Release season & year
		Tagged	Untagged			
1990	KT	14	0	457 (53)	321 (112)	Summer 1992
1991	KT	104	0	255 (17)	66 (13)	Summer 1992
1992	KT	123	0	483 (113)	549 (483)	Fall 1994
1995	KT	1,075	0	228 (27)	47 (17)	Spring 1997
1995	KT	884	0	344 (44)	148 (64)	Fall 1997
1995	KT	96	0	411 (68)	288 (138)	Summer 1998
1995	KT	25	0	582 (40)	863 (198)	Summer 1999
1998	KT	309	0	260 (42)	79 (44)	Fall 1999
1999	KT	827	0	256 (22)	71 (18)	Fall 2000
1999	KH	1,358	0	248 (33)	67 (28)	Fall 2000

Year class	Rearing facility	Release number		Mean total length mm (SD)	Mean weight g (SD)	Release season & year
		Tagged	Untagged			
1999	KT	491	0	284 (54)	108 (60)	Spring 2001
1999	KH	1,582	0	306 (40)	56 (39)	Spring 2001
1999	KH	1	0	520	980	Spring 2010
2000	KT	2,286	0	244 (39)	64 (31)	Fall 2001
2000	KH	1,654	0	240 (23)	58 (16)	Fall 2001
2000	KH	2,209	0	283 (29)	99 (30)	Spring 2002
2000	KH	30	0	365 (14)	195 (20)	Summer 2002
2000	KT	214	0	409 (54)	294 (110)	Fall 2002
2000	KT	907	0	333 (36)	193 (63)	Jan. 2003
2000	KT	10	0	558 (28)	88 (18)	Feb. 2004
2000	KT	3	0	662 (61)	425 (66)	Summer 2006
2001	KT	2,672	0	200 (38)	33 (16)	Fall 2002
2001	KH	4,469	0	227 (24)	52 (17)	Fall 2002
2001	KH	1,715	0	257 (26)	72 (24)	April 2003
2001	KT	1	0	570	750	Summer 2006
2001	KH	1	0	560	1152	Spring 2009
2002	KH	5,864	0	217 (25)	41 (14)	May 2003
2002	KT	856	0	214 (44)	42 (23)	Oct. 2003
2002	KT	0	550			Nov. 2003
2002	KT	3,852	0	215 (37)	43 (20)	Winter 2003
2002	KT	3,663	0	214 (55)	43 (27)	Winter 2003-2004
2002	KT	1	0	550	740	Summer 2006
2002	KH	3	0	523 (25)	1073 (145)	Spring 2010
2002	KH	1	0	530	1020	Spring 2012
2003	KH	9,020	0	223 (26)	49 (24)	Spring 2004
2003	KH	19	0	230 (27)	52 (19)	Sept. 2004
2003	KT	3,519	0	227 (47)	55 (32)	Late winter 2004
2003	KT	3	0	437 (27)	347 (49)	Summer 2006
2003	KT	1	0	690		Winter 2011
2004	KT	0	3,000			Fall 2004
2004	KT	0	1,275			Late wtr '04-early wtr '05
2004	KT	0	17,723			Spring 2005
2004	KH	1,238	800	196 (28)	57 (33)	Spring 2005
2004	KH	0	3,440			Spring 2005
2004	KT	0	8,637			Summer 2005
2004	KT	1	0	510	490	Winter 2007
2004	KH	5	0	452 (23)	563 (117)	Spring 2009
2005	KT	0	6,200			Fall 2005
2005	KH	14	0	299 (14)	174 (28)	Spring 2006
2005	KH	1,762	0	198 (25)	54 (22)	Spring 2006

Year class	Rearing facility	Release number		Mean total length mm (SD)	Mean weight g (SD)	Release season & year
		Tagged	Untagged			
2005	KH	0	13,665			Spring 2006
2005	KT	0	3,947			Spring 2006
2005	KT	510	0	171 (47)	27 (20)	Fall 2006
2005	KH	1	0	330	225	Spring 2009
2005	KH	4	0	400 (34)	414 (132)	Spring 2010
2005	KH	1	0	500 (42)	860 (197)	Spring 2012
2006	KH	0	6,900			Fall 2006
2006	KH	0	600	149 (11)	23 (5)	Fall 2006
2006	KT	0	6,175			Fall 2006
2006	KH	0	5,800			Spring 2007
2006	KH	1,877	1,000	182 (15)	44 (12)	Spring 2007
2006	KT	0	12,973			Spring 2007
2006	KT	4,922	0	171 (30)	22 (11)	Winter 2007
2006	KH	1	0	390	220	Spring 2010
2007	KH	2,167	0	241 (24)	92 (27)	Spring 2008
2007	KT	884	201	151 (36)	20 (10)	Fall 2008
2007	KT	7	0	455 (46)	426 (12)	Winter 2011
2008	KH	9,982	0	198 (35)	56 (19)	Spring 2009
2008	KT	3,874	883	194 (52)	32 (19)	Fall 2009
2008	KT	3	0	412 (29)	276 (74)	Winter 2011
2008	KH	1	0	430	555	Spring 2012
2009	KH	7,884	0	207 (42)	67 (22)	Spring 2010
2009	KT	5,343	808	218 (39)	45 (23)	Fall 2010
2010	KH	5,759	0	197 (25)	58 (22)	Spring 2011
2010	KT	7,785	1,825	230 (40)	56 (29)	Winter 2011
2011	KH	11,244	0	202 (20)	56 (22)	Spring 2012
2011	KT	10,280	907	244 (34)	62 (27)	Fall 2012
2012	KH	6,074	0	240 (24)	101 (31)	Spring 2013
2012	KT	132	0	265 (30)	88 (21)	Fall 2013
2012	KT	4,498	384	356 (45)	76 (38)	Spring 2014
2012	KT	111	0	555 (63)	663 (202)	Fall 2016
2012	TR	21	0			Fall 2017
2013	KH	7,502	0	217 (17)	76 (17)	Spring 2014
2013	KT	1,239	0	242 (39)	61 (31)	Fall 2014
2014	KH	5,035	0	217 (23)	66 (22)	Spring 2015
2014	KT	0	5,786		3.59	Spring 2015
2014	KT	324	0	261 (41)	67 (30)	Fall 2016
2014	KT	0	22			Fall 2016
2015	KT	0	10,381		7	Spring 2016
2015	TR	14,450	0	233 (37)	54 (28)	Spring 2016

Year class	Rearing facility	Release number		Mean total length mm (SD)	Mean weight g (SD)	Release season & year
		Tagged	Untagged			
2015	TR	0	6,350		< 20	Spring 2016
2015	TR	95	0	251 (38)	61 (30)	Fall 2016
2016	TR	1,408	0	207 (26)	33 (15)	Spring 2017
2016	TR	0	2,480		< 20	Spring 2017
2016	TR	563	0	212 (40)	40 (30)	Summer 2017
2016	TR	84	0	217 (20)	40 (21)	Fall 2017
2017	TR	3,301	0	230	57	Spring 2018
2017	TR	0	2,273		< 30	Spring 2018
2017	KT	2,749	0	250	75	Fall 2018
2017	KT	0	655		< 30	Fall 2018
2018	TR	7,891	0	267	75	Spring 2019
2018	TR	0	3,087		< 30	Spring 2019
2018	KT	4,131	0	264	82	Fall 2019
2019	TR	2,162	0	203	58	Spring 2020
2019	KT	0	0			
2020	No Year Class – Research only due to COVID-19					
2021	TR	2,488	0	257	72	Spring 2022
2021	KT	2,577	0	275	90	Spring 2022
2022	TR	1481	0	343	164	Spring 2023
2022	KT	1365	0	320	140	Spring 2023
2023	TR					Spring 2024
2023	KT					Spring 2024
Subtotal		**195,092**	**128,727**			
Total		**323,819**				

(B)

Year class	Rearing facility	*Release number		Mean larval length (mm)	Mean larval weight (g)	Release Dates
		Eggs	Pre-fed larvae			
2008-2009	*KT*		*<2,000,000*			*Spring/Summer 2008-2009 (IDFG)*
2010 - 2012	*KT*		*<3,000,000*			*Spring/Summer 2010-2012 (IDFG)*
2022	TR	74,400	219,496			June 15 to June 30, 2022
2022	KT	67,750	64,720			June 14 to July 5, 2022
2023	TR	125,191	136,122			June 1 to June 15, 2023
2023	KT	77,782	194,057			June 1 to June 15, 2023
2024	TR	TBD	TBD			Spring/Summer 2024
2024	KT	TBD	TBD			Spring/Summer 2024
Subtotal		**345,123**	**614,395**			
Total		**959,518**		***Subtotals and Total only include KTOI releases since 2022**		

*Eggs released assume 100% fertilization. Pre-fed larvae includes any larvae released between onset of hatch (7-10 days post fertilization) and anal plug release (20-22 days post fertilization). Pre-fed larvae are not exposed to commercial fish feeds. Larvae release numbers incorporate neuralation to estimate the number of *live* larvae released. All releases include all biological material (e.g., egg shells, live, dead, deformed) within each release group.

Experimental Early Life Stage Releases (Past and Future)

Monitoring programs determined post-release survival of natural Kootenai Sturgeon embryos and larvae is essentially zero (Rust and Wakkinen 2004; Hardy et al. 2020). This prompted a recommendation by the USFWS Recovery Team (KRWSRT) members to release age-0 embryos upstream from Bonners Ferry during 2008-2012 to investigate early life stage survival. The releases were categorized as a Reasonable and Prudent Alternative (RPA) in the 2006 Biological Opinion, and 2008 BiOp interpretation for Libby Dam operations (USFWS 2006, 2008). The five-year experiment was intended to evaluate: 1) post-release survival of free embryos produced at the Tribal Sturgeon Hatchery, and 2) suitability of habitat for age-0 life stages. To date, there is no evidence of survival from the 2008-2012 hatchery-origin embryo releases; except one 14-day larvae was captured as part of a side-channel incubation and early rearing experiment during the study time frame (Rust and Wakkinen 2011). Despite low success of these efforts, co-managers have agreed to allow additional early life stages from Kootenai Tribal hatcheries now that Parental Based Tagging (PBT) methods have been developed.

Kootenai River Sturgeon PBT research is progressing at UC-Davis (Schreier and Fiske 2021). During July-September 2020, KTOI staff assisted the collaboration by angling 185 hatchery-origin juveniles. KTOI staff recorded fish metrics, PIT-tag ID, and scute patterns; and collected fin tissue for genetic analyses. Fin tissue and individual fish information were provided to UC-Davis to complete a blind parentage assignment test run on these known hatchery-origin Sturgeon. Prior to 2021, parental assignment accuracy was deemed insufficient to identify hatchery versus wild individuals that would not be marked or tagged if released as early life stages. Now that PBT methods have been developed for Kootenai River White Sturgeon, they are being implemented like the Kootenai River Burbot program

(KTOI 2021a, 2021c), however on a smaller and more focused level. Only broodstock used for spawning, beginning in 2022, will be catalogued for future PBT analyses to identify post release captures of larvae by the IDFG MR&E program. In addition, any physically unmarked, untagged Sturgeon may be analyzed for parentage assignments if warranted by co-managers.

Now that the Kootenai White Sturgeon PBT program is underway, the primary objectives of the 2008-2012 may once again be explored by releasing distinct family groups of eggs and larvae at strategic times and places. As the program moves forward with annual egg and larval releases, environmental conditions (e.g., water quality, outflows from Libby Dam, etcetera) at the release sites will also be recorded. Assuming survival is detected someday in the future following the targeted early life stage releases, coupled with the records of the environmental conditions, post-hoc characterization of suitable habitat may be defined and help frame why decades of recruitment failure have occurred. Such experimental releases could also potentially provide valuable information about the spatial and temporal aspects of Sturgeon recruitment failure in other parts of the world.

Expected outcomes from PBT methods research are presented in the KTOI Burbot Hatchery Management Plan (KTOI 2021c). UC-Davis maintains the genetic catalog of KTOI Sturgeon Program broodstock.

3.4 KEY ASSUMPTIONS

The key assumptions are a set of parameters that help the co-managers make in-season and long-term restoration decisions about the hatchery program (e.g., number of fish released, life stages to be released, release locations, etc.). Generally, these assumptions are based on current aquaculture techniques, recent M&E findings, and strategic goals of the co-managing agencies. The parameters are grouped into four categories: 1) hatchery production, 2) natural production, 3) natural spawning, and 4) harvest parameters. Each parameter is defined in Table 7, and the assumed values are compared to recent observed values based on program M&E results.

Table 7. Key assumptions for hatchery production, natural production, and natural spawning.

Parameter	Definition	Tribal Sturgeon Hatchery	Twin Rivers Tribal Hatchery
Hatchery Production			
Broodstock donor source / wild, hatchery	The population from which broodstock and/or gametes are collected.	Kootenai River, wild (Note: first mature hatchery-origin adult spawned 2020)	Kootenai River, wild (Note: first mature hatchery-origin adult spawned 2020)
Broodstock percent survival	Percentage of fish used for broodstock surviving handling procedures and released back to river/lake.	99%	99%
Number of broodstock spawned	Number of broodstock spawned at each hatchery or in the field to rear progeny at the respective hatchery.	Up to 20	Up to 20
Number of broodstock (females: males) per family	Ratio of females to males used for broodstock.	≥1:2 (Note: acquisition of ripe males is a limiting factor)	≥1:2 (Note: acquisition of ripe males is a limiting factor)
Number of families/ family groups	Family: a 1:1 cross between a male and female. Family-group: a cross of one or more females with one or more males. At minimum, each female will be crossed with multiple males, but individually.	12-18 families 5-10 family groups	16-20 families 5-10 family groups
Number fertilized eggs	Total number of eggs successfully fertilized as determined under microscope.	3,000/ family 15,000/ family-group	3,000/ family 15,000/ family-group
Percent hatch	Percentage of fertilized eggs that successfully incubate and hatch.	90%	90%
Percent larval survival	Percentage of fish that survive from hatch until feeding stage.	50%	50%
Percent fry survival	Percentage of fish that survive from larvae to fry stage.	50%	50%
Percent YOY juvenile (Age 0 - 6 months) survival	Percentage of fish that survive from larval stage to 6-m juvenile.	50%	50%
Percent survival to Age-1 in hatchery	Percentage of fish that survive from 6-m juvenile to Age-1 juvenile.	50%	50%
Water source	Hatchery water source	Kootenai River	Kootenai River, Moyie River, Groundwater

Parameter	Definition	Tribal Sturgeon Hatchery	Twin Rivers Tribal Hatchery
Rearing regime		Ambient/Accelerated	Accelerated
Release timing		Fall/Spring	Spring
Age at release		Age-0+	Age-0+
Natural Production			
Percent survival, egg to Age-1	Survival from egg to Age-1 juvenile (Proxy = In-Hatchery)	5% (1.5-7.7% In-Hatchery)	
Percent Age-1+ survival in wild	Survival from Age-1 to Age-2 juvenile	~10%	
Percent Age 2+ annual survival	Survival from Age-2 to Age-3 juvenile	~70%	
Percent Age 3+ to 24 annual survivals	Survival from Age-3 to Age-24 (annual)	~93%	
Percent Age 25+ annual survival	Adults	97%	
Natural Spawning			
Estimated number of spawners	Total number wild/hatchery sexually mature adults per year	~800 wild adults/year (220 female, 490 male) First hatchery-origin males in 2020	
Number of larvae captured	Naturally recruited Sturgeon larvae captured in field surveys	<10 per year	

3.5 DECISION GUIDELINES

All available M&E data will be used to update status and trends information and key assumptions before each APR. Using new information, cooperating agencies will decide whether to proceed with current Decision Guidelines or adjust better address program goals. The cooperating agencies are collectively bound by the phrase "consensus, minus one" to determine biological objectives for the coming year. Consensus, minus one means that no single co-managing entity may prevent the majority from moving a reasonable, well-defined plan forward. Biological objectives help determine annual hatchery production and any needed adjustments to M&E activities. Table 8 displays the initial program Decision Guidelines for each phase.

Table 8. Program Decision Guidelines for Kootenai River White Sturgeon.

Variables	Initial	Current
Donor source	Kootenai River	Kootenai River

Variables	Initial	Current
Percent broodstock from Kootenai River	100%	100%
Percent Kootenai River natural origin broodstock	100%	Begin to use hatchery-origin broodstock as available
Families produced	12-18 at KT / H1 16-20 at TR / H2	12-18 at KT / H1 16-20 at TR / H2
Family groups produced	NA	5-10 at KT / H1 5-10 at TR / H2
Age-0+ YOY released	Variable	Variable
Age-1+ released	Variable	Variable, ≤500 per family group
Natural recruitment	None or not biologically meaningful	None or not biologically meaningful

3.6 BIOLOGICAL OBJECTIVES

Biological objectives for Sturgeon hatchery production will be determined by KTOI and the co-managers at the APR. Most in-season management decisions involve updating the annual hatchery production plan and release strategy; however, RM&E is also discussed to ensure appropriate evaluations to guide the program. The annual plan will change as needed, as the Sturgeon population and habitat are restored.

An example of how biological objectives for Sturgeon production were initially estimated is shown in Table 9. Table 9(a) lists initial assumptions about in-hatchery production (number of eggs collected and life-stage survival rates) to produce the initial goal of 30,000 age-1 juveniles for release into the Kootenai River. Table 6(b) shows how releasing 30,000 age-1 juveniles was initially projected to meet the program's adult abundance objective for the Lower Kootenai River based on the age-specific survival probabilities for each year-class. These in-hatchery and post-release survival assumptions were used to design and scale the Sturgeon program at Twin Rivers Hatchery and are updated annually at the APR.

Table 10 provides current assumptions about in-hatchery production and post-release survival to meet the program's current release goals and adult abundance objective. Note that each spawn is divided between each hatchery as a fail-safe. All family-groups are sub-sampled to determine ploidy level proportions within each family groups. Entire family-groups are euthanized if abnormal ploidy levels exceed 50%. Changes in rearing techniques are intended to increase juvenile fitness and subsequent in-river survival, i.e., a focus on quality vs. quantity.

Table 9. Initial Master Plan (KTOI 2012) assumptions about in-hatchery production to produce the initial goal of 30,000 Age-1 juveniles for release into the Kootenai River.

(A)

1 Family	Hatchery		
Life Stage	**Numbers**	**Survival Probability**	**Stage**
Eggs	10,000	0.9	Fertilization
Fertilized eggs	9,000	0.9	Egg-Hatch
Free embryos	8,100	0.5	Hatch-Larvae
Larvae	4,050	0.5	Larvae-Fry
6-mo juveniles	2,025	0.5	Fry-6-mo juv
Age 1 juveniles	1,013	0.5	6-mo-1 yr juv
30 Families	**Hatchery**		
Life Stage	**Numbers**	**Survival Probability**	**Stage**
Eggs	300,000	0.9	Fertilization
Fertilized eggs	270,000	0.9	Egg-Hatch
Free embryos	243,000	0.5	Hatch-Larvae
Larvae	121,500	0.5	Larvae-Fry
6-mo juveniles	60,750	0.5	Fry-6-mo juv
Age 1 juveniles	30,375	0.5	6-mo-1 yr juv

(B)

Year Class	Post-Release		
Life Stage	**Numbers**	**Survival Probability***	**Stage**
Age 1 juveniles	30,375	0.10	Age 1+ - Age 2+
Age 2+ juveniles	3,037	0.70	Age 2+ - Age 3+
Ages 3-24 (total) juveniles/subadults	2,125	0.93 (constant age 3-24)	Age 3+ - Age 24
Ages 25-75 (total) adults	1,976	0.97 (constant age 25+)	Age 25+

Table 10. Current assumptions about in-hatchery production to produce 5,000 Age-1 juveniles for release into the Kootenai River.

(A)

1 Family Group	Hatchery		
Life Stage	**Numbers**	**Survival Probability**	**Stage**
Eggs	15,000	0.9	Fertilization
Fertilized eggs	13,500	0.5	Egg-Hatch
Free embryos	6,750	0.5	Hatch-Larvae

Larvae	3,375	0.5	Larvae-Fry
6-mo juveniles	1,687	0.5	Fry-6-mo juv
Age 1 juveniles	843	0.5	6-mo-1 yr juv
10 Family Groups	**Hatchery**		
Life Stage	**Numbers**	**Survival Probability**	**Stage**
Eggs	150,000	0.9	Fertilization
Fertilized eggs	135,000	0.5	Egg-Hatch
Free embryos	67,500	0.5	Hatch-Larvae
Larvae	33,750	0.5	Larvae-Fry
6-mo juveniles	16,875	0.5	Fry-6-mo juv
Age 1 juveniles	8,437	0.5	6-mo-1 yr juv (accelerated growth release)
Age-1+ @ 18 months	4,218		12-18 months (ambient growth release)

(B)

Year Class	**Post-Release**		
Life Stage	**Numbers**	**Survival Probability***	**Stage**
Age 1 juveniles	5,000	0.10	Age 1+ - Age 2+
Age 2+ juveniles	500	0.70	Age 2+ - Age 3+
Ages 3-24 (total) juveniles/subadults	350	0.93 (constant age 3-24)	Age 3+ - Age 4+
Ages 25-75 (total) adults	325	0.97 (constant age 25+)	Age 25+

*Values from Dinsmore et al. 2015; Hardy et al. 2020

4.0 MONITORING AND EVALUATION

Conservation aquaculture for Sturgeon is a critical component of the recovery of this species within the Kootenai River subbasin of Idaho and British Columbia. All phases of the hatchery program incorporate best management practices as outlined in the hatchery operations manual initially developed by KTOI in 2015-2016 and updated in 2020-2021 (KTOI 2021b). This plan, which incorporates the APR and ISMP processes, will track program outcomes and guide future implementation.

Monitoring and evaluation activities are prioritized based on three criteria:

1. **Impact on management decisions.** Metrics needed to implement the four-step In-Season Management Procedure (ISMP) and used in the Adaptive Management

process. The ISMP steps include: 1) Update Status and Trends information, 2) Update Key Assumptions, 3) Review Decision Guidelines, and 4) Set Biological Objectives for the upcoming year.

2. **Degree of uncertainty.** Metrics with a high degree of variability due to random effects, or that vary in space and time.

3. **Feasibility of monitoring.** Metrics that can be monitored efficiently (i.e., with a reasonable investment of time and resources) and effectively (i.e., with sufficient accuracy and precision).

The purpose of this section of the Plan is to:

- Identify the key metrics to be monitored.
- Identify the agency or agencies responsible for monitoring each metric.
- Explain how this information will be used to adaptively manage the Sturgeon restoration program.

4.1 POPULATION STATUS

Monitoring and evaluation metrics used to assess the status of the Kootenai River Sturgeon population are summarized in Table 11. These include metrics monitored in the field and in-hatchery by KTOI, IDFG, BC Ministry and MFWP. These metrics are used to evaluate the progress and success of the restoration program in meeting population goals stated in Section 3.2.

Table 11. Monitoring and evaluation metrics are used to assess the status of the Kootenai River Sturgeon population.

Metric	Field or In-hatchery	Agency Responsible
Total adult abundance	Field	IDFG *Project 198806500,* BC Ministry (subcontracted under KTOI *Project 198806400* and IDFG *Project 198806500*), KTOI *Project 198806400,* and MFWP *Project 200600800*
Population genetic structure	Field and In-hatchery	KTOI *Project 198806400 (subcontract to UC Davis GVL);* all samples sent to GVL for processing, KTOI pays all
Age composition of population and age-specific survival	Field	IDFG *Project 198806500*, BC Ministry (subcontracted under KTOI *Project 198806400* and IDFG *Project 198806500*) and KTOI *Project 198806400; MFWP 200600800*

Metric	Field or In-hatchery	Agency Responsible
Annual capture probability	Field	IDFG *Project 198806500*, BC Ministry (subcontracted under KTOI *Project 198806400* and IDFG *Project 198806500*)
Timing and location of spawning	Field	IDFG *Project 198806500*
Sex ratio	Field and In-hatchery	IDFG *Project 198806500*, BC Ministry (subcontracted under KTOI *Project 198806400* and IDFG *Project 198806500*) and KTOI *Project 198806400 (subcontract to UC Davis GVL); MFWP 200600800*
Annual growth, size at age	Field	IDFG *Project 198806500*, BC Ministry (subcontracted under KTOI *Project 198806400* and IDFG *Project 198806500*) and KTOI *Project 198806400; MFWP 200600800*
Fecundity and sperm motility	In-hatchery	KTOI *Project 198806400*
Egg fertilization rate	In-hatchery	KTOI *Project 198806400*
Egg hatch rate	In-hatchery	KTOI *Project 198806400*
Larval survival rate	Field and In-hatchery	In-hatchery: KTOI *Project 198806400;* Field: IDFG *Project 198806500*
Juvenile survival rate	Field and In-hatchery	IDFG *Project 198806500*, BC Ministry (subcontracted under KTOI *Project 198806400* and IDFG *Project 198806500*), KTOI *Project 198806400,* MFWP *Project 200600800*
Natural egg abundance	Field and In-hatchery	In-hatchery: KTOI *Project 198806400;* Field: IDFG *Project 198806500*
Natural larval abundance	Field	IDFG *Project 198806500*
Natural juvenile abundance	Field	IDFG *Project 198806500*, BC Ministry (subcontracted under KTOI *Project 198806400* and IDFG *Project 198806500*), and MFWP *Project 200600800*

Metric	Field or In-hatchery	Agency Responsible
Water flows and temperatures	Field and In-hatchery	KTOI *Project 198806400,* IDFG *Project 198806500* and BC Ministry (subcontracted under KTOI *Project 198806400* and IDFG *Project 198806500*), and US Army Corps of Engineers
River productivity, plankton and algal communities, N:P ratio	Field	KTOI *Projects 198806400, 199404900 200200200, and 200201100;* and BC Ministry (subcontracted under KTOI *Project 198806400* and *199404900)*
Seasonal movements and migrations	Field	IDFG *Project 198806500*, BC Ministry (subcontracted under KTOI *Project 198806400* and IDFG *Project 198806500*), and MFWP *Project 200600800*

4.2 BROODSTOCK COLLECTION

Adult Kootenai River White Sturgeon are captured by angling (KTOI) and use of setlines (IDFG) from April through June to supply the aquaculture program with broodstock. In addition to collecting broodstock for the hatchery program, sampling efforts provide the basis to estimate the status of the remnant wild population and recruitment of hatchery-origin fish to the adult population (IDFG, BC Ministry, KTOI, and MFWP capture adults).

The goals of broodstock collection are to:

- Spawn all wild, unmarked adults as possible before wild remnant population is extirpated.
- Maximize the temporal and geographic extent of annual broodstock capture to incorporate the maximum amount of genetic diversity into the annual production of families in the hatchery, and therefore, into the recipient population over time.
- Meet program demographic and genetic objectives at annual and decadal scales.
- Include hatchery-origin broodstock into the program breeding matrices.

Brood collection is focused primarily on the annual spawning component of the population, although both spawners and non-spawners are present in sampling areas. Broodstock sampling is concentrated from April-June in staging areas between river kilometer (rkm) 205-215, and from May - June at known spawning sites (rkm 229-245). Female candidates

are typically captured and transported from staging areas during April/May. Annual KTOI angling activities generally provide < 100 broodstock candidates (KTOI 2015-2019).

Below is a list of metrics used to characterize the biological and reproductive health, condition, and behavior of individual fish used as spawners in the conservation aquaculture program.

Metrics to be monitored during brood collection and holding

- Fin tissue clips for genetic samples (one clip in field, one fin clip in hatchery)
- Broodstock survival in the hatchery
- Number of broodstock and sex ratio
- Broodstock maturation
- Length, weight, age, and condition factor
- Location and timing of captures
- Ploidy - only normal ploidy (8N) Broodstock are used

4.3 GENETICS

4.3.1 Diversity

As noted above, the annual spawning plan captures approximately 80% of the annual genetic diversity from each annual wild spawning class, and overall has captured 96% of the genetic diversity of the wild population (Schreier and Van Eenennaam 2019). Evaluation of post-release recaptures determined that 56% of all families released were represented in the population through 2014 (Schreier et al. 2015). The Twin Rivers Tribal White Sturgeon and Burbot Hatchery, combined with the Kootenai Tribal Sturgeon Hatchery now incorporate twice as many wild adult spawners per year as prior to Twin Rivers Hatchery startup during 2015, which is of paramount importance as wild adult abundance declines. By doubling the number of broodstock spawned and subsequent unique family-groups created annually, the program increases the probability that genetic diversity is maintained while also increasing behavioral and phenotypic diversity. Each of the families or "family groups" produced is explicitly segregated until release which helps the program control year-class diversification and is vital to the implementation of the Kootenai River Sturgeon PBT program.

Metrics to be monitored

- Annual broodstock allelic variation (genetic diversity)

- Genetic diversity of the hatchery origin progeny released
- Long-term family contributions to hatchery Sturgeon standing stock
- Assign recaptured Sturgeon to parents supporting a PBT program (in progress)

4.3.2 Spontaneous Autopolyploidy

All White Sturgeon are polyploids (i.e., have more than one set of chromosomes from each parent). Normal ploidy White Sturgeon are octoploid, which means everyone has 4 sets of chromosomes from each parent that combine to create progeny with 8 chromosome copies (8N). Abnormal ploidy states have been observed (e.g., 10N and 12N) in White Sturgeon. Recent studies suggest that abnormal ploidy individuals may have reduced fitness, performance, and reproductive viability (Leal et al. 2021).

Spontaneous Autopolyploidy (SA) was discovered in Kootenai Sturgeon during routine genetics monitoring of the 2011 year-class (Schreier et al. 2013). The number of abnormal ploidy Sturgeon released before 2012 is not known; in addition, spontaneous autopolyploids continued to be released in small numbers as understanding of the issue developed. Some of those fish have survived (KTOI unpublished data). Since 2012, management of the issue has evolved as research investigated potential causes (Gille et al. 2015; Van Eenennaam et al. 2020) and developed accurate determination techniques (Fiske et al. 2019).

Kootenai River White Sturgeon Conservation Aquaculture Agreements (2019) for Spontaneous Autoploidy

All broodstock candidates are screened for SA. Blood collection is part of broodstock candidate workups. If no blood is collected, the sample cannot provide a ploidy determination or the adult is determined to be 12N, the adult is not used for spawning. This includes all males that staff collect milt from. All broodstock are tested using methods discussed within Fiske et al. (2019). Beckman Coulter Counter Particle Analyzers are used for all testing. All testing is completed by KTOI staff. Ploidy assignment is completed by KTOI using a protocol specific to KTOI. A 12N wild adult is released immediately, is noted in the database, and sperm samples from 12N males are discarded. This is an important action given that a 12N female was spawned in 2016. This resulted in euthanizing all four, very large families created with the female (10N validated by flow cytometry as part of Fiske et al. 2019).

The total number of age-1 juveniles released annually is limited to 500/family group. All juvenile family groups released will be > 90% 8N (normal ploidy level). All individual juvenile sturgeon that are verified 12N fish (not normal ploidy level) may be destroyed depending on co-manager agreements. All juvenile family groups are examined prior to release using methods discussed in Fiske et al. (2019). Coulter Counters are used for all testing and are calibrated annually.

KTOI pre-screens a minimum of 30 individuals per "family" (1 female x 1 male) or "family group" (1 female x 3-5 males) for SA. SA pre-screening involves a random grab of 30 fish. If a family group grab sample indicates SA occurrence exceeds 50%, KTOI will euthanize the entire family group. The pre-screen is also used to estimate how many fish need to be screened from each hatchery, determine costs/supplies needed and labor needed to complete the testing. All SA pre-screening or individual testing may occur when fish reach sufficient size (preferably 20 g or 7 months of age; although methods have been developed to test fish as small as 2.2 g at 3 months of age; however, fish <3g are too small to PIT tag which prevents individual identification (Elliston et., al. KTOI unpublished data). Ideally, all fish to be released are PIT-tagged to be able to match blood screen results to an individual after testing is complete. The timing of screening, individual data protocols and fish segregation are determined by KTOI staff, taking into consideration co-manager agreements and regulatory restrictions.

Spontaneous Autoploidy actions completed under previous Kootenai River White Sturgeon Conservation Aquaculture Agreements (2013-2019)

Due to abnormal results observed during other genetics monitoring activities (2011-2012), Schreier et al. (2013) investigated the occurrence of SA in KTOI hatchery juveniles. At that time, the Kootenai River working group agreed to release all families with < 33% SA occurrence by screening 15-30 fish per family depending on family size. Families exceeding 33% SA would had all individuals screened and abnormal ploidy fish were euthanized.

In 2014, KTOI realized that SA likely had been occurring throughout the program's history and would likely be observed in every subsequent year-class spawned in the future. At that time, using flow cytometry for routine SA testing at KTOI's Tribal hatcheries was not feasible so blood smears were used as an alternative. A pilot study was completed on a single family, and the blood smears compared to flow cytometry were found to be 100% accurate. Expansion of this study was put on hold; however, based on the preliminary result compared to flow-cytometry, blood smears were used to determine SA of juvenile Sturgeon for the following couple years. A formal study to expand SA testing methods was completed using the 2016 year-class during spring 2017 before release (Fiske et al. 2019). The results were important for four reasons: 1) a wild female was verified 12N, spawned and 10N progeny were created; 2) blood smears were found to underestimate SA occurrence in the Kootenai population; 3) blood smears were fairly accurate (family variation effects accuracy), but not at the level preferred by co-managers, and 4) Coulter Counter methods were 100% accurate and validated by flow cytometry (Fiske et al. 2019). Thus, KTOI switched to using Culter Counters and invested in the technology.

The 2016 year-class had a high occurrence of SA (10N and 12N) within several females' progeny. Over 8,000 juveniles were euthanized, and another 5,000 juveniles were tested to eliminate 12N fish. This was the worst occurrence for any year-class known on record; however, some females' progeny had 0% SA, demonstrating the variability that may be observed across brood years. Using a Coulter Counter, all broodstock candidates have since been screened to verify normal ploidy before the fish are used for spawning; from 2017 to

present. Again, blood collection/screening is standard practice for broodstock candidate workups and before 2017, broodstock were not screened.

In summary, the percent SA thresholds determining release or complete screening of each family were 33% (2013-2016 year-classes) and 20% (2017-2018 year-classes). The current protocol is < 10% 12N family-groups will be released (i.e., family groups with < 10% SA may be released without further testing beyond the initial random sub-sampling in part because the probably of survival to maturity is very low; family groups with 10-33 % will all be sorted, tested and verified 8N; family groups with ≥ 50% 12N will be euthanized).

Metrics to be monitored

- Prevalence of SA (8N or 12N) in broodstock and juveniles using Coulter Counters

4.3.3 Parentage-based Tagging (PBT)

The Sturgeon hatchery spawning plan emphasizes genetic management (KTOI 2018a; KTOI 2019a) as well as creating and maintaining "family groups" to fully utilize the power of PBT. A critical component of future M&E will incorporate PBT. It replaces the need for physical tagging (PIT tagging, etc.) of all hatchery-origin fish if done properly. PBT may be applied across entire year classes to expand its potential and the flexibility of applying results to RM&E studies. PBT may also be applied to select broodstock crosses whose progeny will be released for targeted purposes supporting specific studies. The premise is the same regardless of scope.

The first and most important step is to sample fin tissue clips from all broodstock spawned as part of PBT. Second, progeny from all crosses or spawning adult groups are kept separate during hatchery rearing. These unique "families" form "family groups" when combined and are released at specific times and places. When offspring are recaptured, a fin clip is collected, DNA is extracted and genotyped to determine parentage, which provides information on hatchery versus wild origin, year-class, and release-site survival without applying traditional tags. Another novel use of PBT is experimental release strategies of different life stages in the recovery area. By releasing specific "family groups" at key times and places and monitoring ecological conditions at those respective times and places, studies investigating habitat-related recruitment failure may be undertaken while simultaneously rebuilding the population. Such experimental releases could potentially provide valuable information about the spatial and temporal aspects of Sturgeon recruitment failure.

Kootenai White Sturgeon PBT program is being maintained in collaboration with the University of California-Davis Genomics Variation Lab (Dr. Andrea Schreier) and implemented into hatchery production plans (subcontracted by KTOI *Project 198806400*) with the intention of parental assignments to targeted early life stage (fertilized eggs, yolk-sac fry, or unfed larvae) releases. There are unique challenges associated with PBT analysis of Kootenai Sturgeon versus other White Sturgeon populations due to their low population

genetic variability, and PBT analysis of White Sturgeon in general caused by their large genome, which includes eight replicates of their ~240 chromosomes (Schreier et al. 2013). A Sturgeon PBT protocol was completed in 2021 (Fiske et al. 2023), funded by KTOI *Project 198806400* and its accuracy verified. Early life stages of Kootenai Sturgeon were first released in 2022 at specific locations and times to begin to evaluate recruitment dynamics and early life stage ecology across the recovery area. Parental Based Tagging programs utilize the known genetic profiles of selected broodstock spawned and their surviving progeny to determine individual metadata. The data gleaned from recaptured progeny, including physiology and habitat use data, may be extremely informative for testing hypotheses about recruitment dynamics and early life history eco-physiology that would otherwise be impossible to gather under the current conditions of little to no natural recruitment. Any sturgeon captured (except for eggs) during M&E activities can now be screened for hatchery-origin or if a capture does not match known hatchery crosses it may imply a natural 'wild' recruit was produced.

Metrics to be monitored

- Genetic lineage/parentage
- Post-release survival
- Gender
- Year-class
- Release location
- Post-release behavior (movement/ dispersal)
- Post-release growth
- Habitat suitability at release/capture location(s)

4.4 SPAWNING

The KTOI hatcheries (combined) typically spawn 10-12 females crossed with up to 40 males to create up to 40 unique families which comprise 10-12 distinct female-based "family groups". An average of 30-36 unique families, or 8-10 "family groups" typically survive and contribute to each year class (KTOI 2018a, KTOI 2019a). The current program goal is to collect 15,000 eggs per female for production purposes. Eggs from each female are divided into five groups (3,000 eggs each) and each is fertilized with sperm from a different male; forming five families. The five families from each female form a "family group".

KTOI is evaluating more natural methods of fish production. Inducing female ovulation without hormone injections was one objective. To date, placing a maturing female with ripe males has not induced natural ovulation in-hatchery.

Below is a list of metrics used to monitor the success of the aquaculture program in meeting the spawning objectives.

Metrics to be monitored

- Gamete viability: Female germinal vesicle breakdown (GVBD)
- Polarity index (PI) values
- Male sperm motility/viability
- Number of broodstock used for spawning matrices; parents within each family group
- Genetic contribution
- Number of eggs collected for spawning
- Number of fertilized eggs (estimated using number of eggs collected X neurulation percentage; this is used to predict hatch percentages)

4.5 INCUBATION

Incubation takes place at both KTOI hatcheries. Incubation methods are described in detail in KTOI 2021b (Chapter 4 – Sturgeon Incubation).

KTOI is evaluating more natural methods of egg incubation by incubating eggs in natural rock collected from the Kootenai River arranged in flow-through raceways as a more natural option of egg incubation. Survival is anticipated to be low using this incubation method but given the reduced release targets the program now produces (e.g., 500 juveniles/family group), release targets may still be achievable, and this method eliminates standard methods that include mechanical agitation. Briefly, natural rock substrate (disinfected with 1% sodium hypochlorite for 24 hours) from the river is arranged in a raceway and fertilized eggs are broadcast onto the disinfected substrate 1-minute post fertilization (at the point when eggs become adhesive). This study doesn't include egg deadhesion or manual stirring of eggs for long periods of time and eliminates the use of a commercial egg incubator that tumbles the eggs. All methods that agitate the eggs post-fertilization are thought to possibly promote SA. If natural substrate methods can be developed, they may help reduce incidence of SA, minimize potential hatchery effects on fish and improve growth (see Appendix A). Results dating back to 2019 are preliminary with variable success, but promising. Fungal infestation is problematic using this method and survival has been low as expected compared to standard hatchery incubation practices. Fungicide treatments are now being evaluated to determine the appropriate concentration to use that limit fungal growth while not adversely affecting sturgeon embryos. KTOI intends to continue to expand these evaluations and applications. For more detail, see KTOI 2021b Chapter 4.

Below is a list of metrics used to monitor the success of the aquaculture program in meeting the incubation objectives.

Metrics to be monitored

- Number of eggs per incubation jar/raceway
- Percent neurulation (72 hours post-fertilization)
- Percent fertilization
- Estimated hatch number (based on neurulation)
- Fungal control/removal
- Water temperature

4.6 REARING

Fish rearing takes place at two KTOI hatcheries. Rearing methods are described in detail in KTOI 2021b (Chapter 5 – Sturgeon Rearing). Between the time of egg collection and juveniles are 4 months old, each family group is split in half; half of each family group is reared at each hatchery. Standalone egg incubators and rearing systems allow for segregation of family groups throughout all phases until release. Splitting family groups between hatcheries provides a safeguard against catastrophic loss due to potential facility failure(s).

Current individual family size targets may be 100-200 juveniles, or for a family-group, 500 juveniles is the target. This range accommodates inherrant differences that occur within each year class due to variations in fertilization, growth, high-grading, health, ploidy related culling and survival to release. Family size targets were established to: 1) ensure survival of a representative number of offspring from each family to an age where they may spawn and contribute to the next generation, 2) avoid excessive contributions from any one family that might skew a year-classes genetic variability, and 3) limit total population size to limit intra-species competition and density-dependent growth and survival limitations post-release.

From 2015-2020, half of all juveniles were reared at Twin Rivers Tribal Hatchery on heated groundwater (“accelerated growth method”) to promote larger size-at-release in late spring. The other half of juveniles were reared at the Tribal Sturgeon Hatchery on ambient Kootenai River water (“ambient growth method”) until release the following fall (KTOI 2018a, KTOI 2019a). Thus, half of all juveniles from 2015-2021 (excluding Covid19 shutdown) were reared at ambient temperatures mimicking what any naturally produced Sturgeon experienced with regards to water temperature. Beginning in 2022 accelerated growth methods are now being applied at both Tribal Sturgeon Hatcheries and all juveniles are being released in Spring. In addition, early life stage releases are now part of the

conservation aquaculture program and include releases of eggs or larval sac-fry (not both to maintain the ability to discern life stage specific inquiry's) that may be temporarily reared one or both hatcheries from each family group.

Below is a list of metrics used to monitor the success of the aquaculture program in meeting the rearing objectives.

Metrics to be monitored during rearing

- Family group segregation and separation
- Larvae on feed: percent survival
- 4-month-old juveniles: percent survival, length, weight, condition factor
- Age-1+: percent survival, length, weight, condition factor (accelerated rearing)
- Age-1+: percent survival, length, weight, condition factor (ambient rearing)
- Ploidy testing

4.7 RELEASE STRATEGIES

Releasing eggs or larvae in June/July of the spawn year or 10-month-old juveniles in May of the following year are the release strategies. PBT methods were successfully developed at UC-Davis; funded by KTOI *Project 198806400* and is now part of the conservation aquaculture program that allows the release of eggs or larvae. PBT will now allow KTOI and co-managers to evaluate recruitment dynamics. By releasing discrete family groups and early life stages at key times and places and documenting ecological conditions at those respective times and places, studies investigating habitat-related recruitment failure may be undertaken while simultaneously rebuilding the population. Such experimental releases are expected to provide valuable information about the spatial and temporal aspects of Sturgeon recruitment failure.

Release sites include (see page 3; T.J. Ross General Release Location Map):

- Montana near Troy, MT (rkm 304)
- Moyie River Confluence (rkm 259)
- Crossport (rkm 256)
- Bonners Ferry, ID (rkm 244.5)
- Deep Creek (rkm 240)
- Nimz Ranch KTOI Tribal property, off-channel habitat, pond complex (rkm 222)

- Copeland (rkm 200)
- Porthill (rkm 170)
- Goat River, BC (rkm 144.5)
- Kootenay Lake (various; most recent release site was Crawford Bay)

Below is a list of metrics used to monitor the success of the aquaculture program in meeting the release objectives.

Metrics to be monitored

- Number released by life stage and at each location
- Habitat conditions at release sites
- Post-release behavior, growth, and condition
- Annual post-release survival (to date)
- Survival and movements as a function of:
  - Size at release
  - Location
  - Season of release

4.8 RESEARCH

- Further develop Parental Based Tagging methods
- Evaluate early life history dynamics across habitats using PBT
- Spontaneous autopolyploidy screening techniques and causes
- Post-release survival of abnormal ploidy Sturgeon
- Investigate novel natural incubation techniques
- Improve rearing techniques
- Gender determination (development of a genetic sex markers)

5.0 ADAPTIVE MANAGEMENT

Kootenai River White Sturgeon conservation and recovery efforts continue to face daunting challenges and uncertainties. We do not know when natural recruitment will be restored; how long the aging wild population will remain reproductive; whether hatchery-origin fish will spawn and recruit successfully in the wild; and to what extent future habitat productivity may change given nutrient additions and habitat restoration actions. Other factors to consider include Libby Dam operations, Kootenay Lake level management, climate change, river/lake (recovery area) carrying capacity and pollutants entering the recovery area from upstream coal mines in British Columbia.

KTOI strives to use the best available science to identify and qualify potential risks and benefits associated with the impending demographic issues. In many cases, the relative magnitudes of risks and benefits, and associated response curves and thresholds can be modeled, but are impossible to precisely predict given critical uncertainties related to climate variability and socioeconomics driving hydro-operations and other anthropogenic affects.

The Kootenai River Sturgeon program undergoes annual reviews of critical variables and metrics as part of the ISMP and APR processes described below, which provide the adaptive management foundation for the Sturgeon aquaculture components of the program and associated RM&E activities. KTOI and co-managers may adjust release numbers and modify culture techniques in response to annually updated age-specific survival rates, ages at first maturity for males and females, and spawning frequency or periodicity values for Sturgeon in the Kootenai River. Thus, this suite of direct adaptive feedback loops will continue to serve the program well, as reflected in the program's successful history.

5.1 ANNUAL PROGRAM REVIEW

Each year, decisions about broodstock management, gamete collection, production goals, release strategies, and M&E activities for the coming year, are made collectively at an Annual Program Review (APR) workshop. The purpose of the APR is to implement the four-step (In-season management procedure) ISMP described above with all stakeholders present to support information sharing, informed decision-making, and management to meet conservation objectives.

The agenda for the APR workshop follows the steps outlined in the ISMP. The APR is generally a science-driven process that results in an annual action plan. The APR participants typically include appointed representatives from the four primary co-managing agencies (KTOI, IDFG, MFWP, BCMOF), action agencies (BPA, USACE, DFO), and regulatory entities within the USFWS. The workshop and action plan agreements constitute the coordinated implementation component of the program.

Participants in the APR workshop will include appointed representatives of agencies with specific M&E and management responsibilities as well as Sturgeon experts; this group will include some members who are also part of the Kootenai River White Sturgeon Recovery Team. Information generated through the APR will be reviewed with the Kootenai River White Sturgeon Recovery Team when the team convenes.

The Kootenai River White Sturgeon APR workshop should be conducted each spring to allow time to complete the previous year's M&E results for use in planning the upcoming year, while arriving at goals for broodstock and gamete collection before the spawning period (which occurs between mid-May and mid-July for Sturgeon). A third-party facilitator helps guide the workshop to address four fundamental questions:

1) Given the information provided, what are the best estimates for the key assumptions (see ISMP Step 2, Section 5.2.2)?

2) Do the assumptions need to be changed (see ISMP Step 3, Section 5.2.3)?

3) What are the biological objectives for the coming year (see ISMP Step 4, Section 5.2.4)?

4) How can the M&E program be improved in the coming year?

The first part of the workshop features presentations of M&E results related to the key assumptions for the KTOI hatchery program. The APR also includes sessions on: (1) hatchery operations, (2) post-release survival and distribution, (3) habitat, and (4) spawning and natural recruitment. Prior to the workshop, KTOI coordinates with participating agencies and entities to ensure that the most current information will be presented and discussed at the workshop. The ISMP tool will then be populated with the most recent empirical data and analytical results to update population status and trends.

In the second part of the workshop, the management team reviews the discussions and conclusions from part one of the workshop regarding the Decision Guidelines (see ISMP Step 3). The management team includes policy and technical personnel from collaborating agencies and entities and presents conclusions and modifications to the Decision Guidelines as needed (Figure 9). The purpose of the Decision Guidelines is to assure that the long-term goals for conservation and harvest established in the conservation aquaculture program are met over time. The product of this portion of the workshop is an updated annual action plan to guide and coordinate M&E activities during the coming year(s).

For the third part of the APR workshop, the annual action plan is reviewed and updated as needed. Based on the action plan and workshop outcomes, each agency and entity involved in actively implementing the M&E Plan will review their planned hatchery and/or field M&E activities for the upcoming year. After the workshop, the facilitator provides a draft workshop summary to all workshop participants, incorporating findings, conclusions, and final decisions for review. Workshop participants will confirm (and if necessary correct) the workshop summary and the facilitator will produce and distribute a final workshop

record. The ISMP database, management tools, Decision Guidelines and other associated products will be retained along with the workshop summary for reference in subsequent APR workshops.

In addition to this formal process, the participatory agencies will continue to communicate routinely throughout the year to coordinate the month-to-month logistics of program activities and reporting requirements.

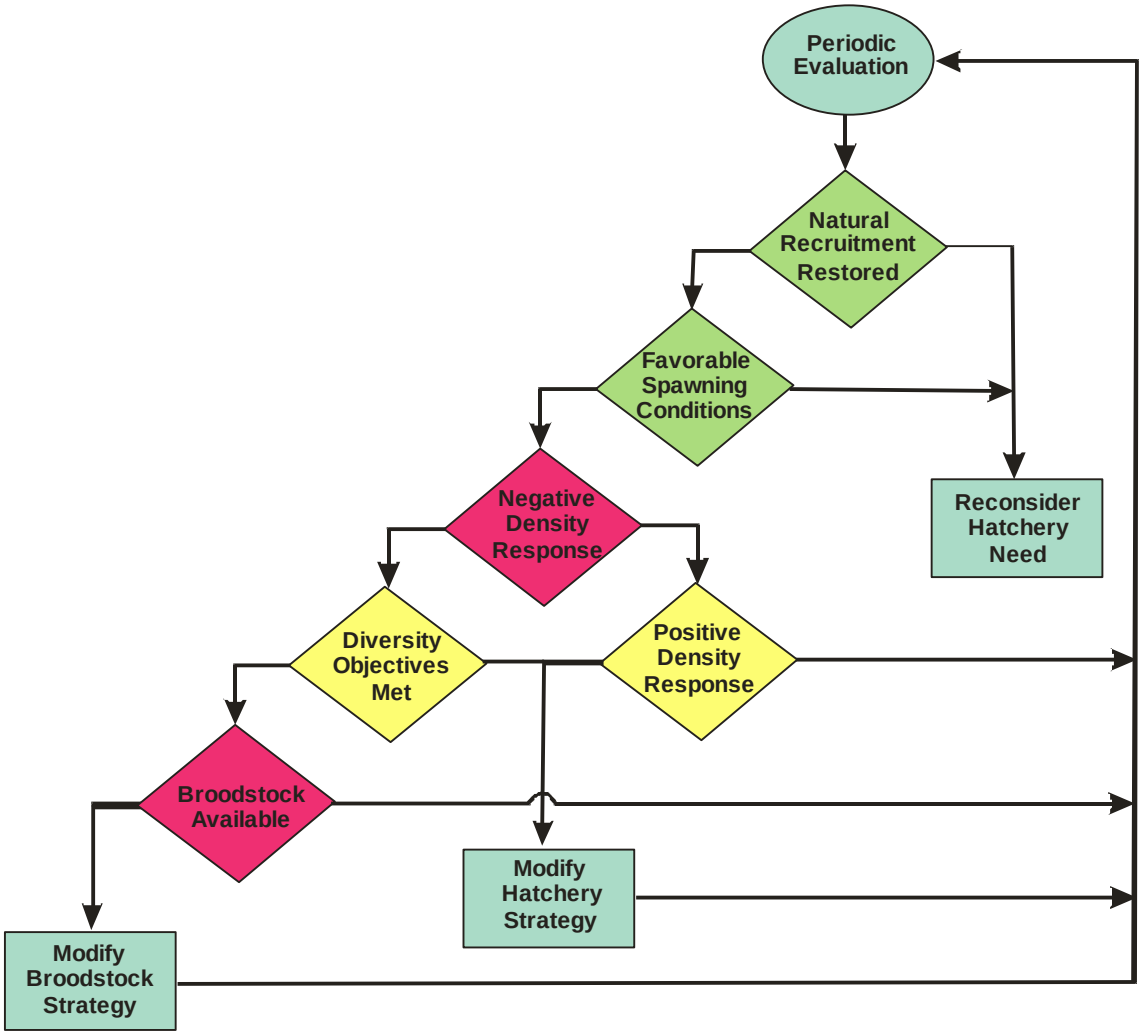

Figure 8. Monitoring and evaluation feedback loops for the Kootenai Sturgeon conservation aquaculture program.

Question 1:	Has substantial natural recruitment occurred?
Metrics:	*Number/percentage of unmarked fish in juvenile sampling program.*
Response:	*Re-evaluate appropriate level of hatchery supplementation based on frequency and magnitude of natural recruitment.*
Question 2:	Has the wild spawning distribution shifted to upstream areas of potentially more suitable spawning habitat?
Metrics:	*Telemetry data on movements of mature fish during spawning periods.*
Response:	*Re-evaluate whether wild broodstock, if limited, are best employed in the wild or the hatchery.*
Question 3:	Are wild spawner numbers adequate to continue to provide hatchery broodstock?
Metrics:	*Catch per unit effort, annual number of broodstock collected, percentage of previously-unspawned individuals in annual adult sampling program*
Response:	*Consider modification of broodstock collection program numbers, need for extended broodstock holding, or reduction in program based on cost/benefit analysis and progress toward objectives.*
Question 4:	Are broodstock numbers and mating strategies adequate to optimize genetic diversity?
Metrics:	*Effective population size based on broodstock numbers, representation of genetic types in broodstock*
Response:	*Consider increasing or decreasing annual broodstock numbers as appropriate.*
Question 5:	Has survival, growth or condition of Age-1 or older juveniles declined substantially in response to increasing density?
Metrics:	*Annual survival and growth rates estimated with mark-recapture data from juvenile monitoring program. Condition estimated from length-weight relationship. Size and age at maturation and reproductive periodicity of hatchery-origin fish.*
Response:	*Consider reductions in annual releases, changes in release distribution, changes in family sizes, rearing fish to a larger size to avoid size-specific limitations, and fish removal to reduce biomass as appropriate based on risk/benefit calculation and progress toward objectives.*
Question 6:	Has juvenile Sturgeon distribution or behavior changed substantially in response to increasing density?
Metrics:	*Catch per unit effort by area, movement data from tagged fish.*
Response:	*Weigh relative benefits of expanded distribution versus detriments of increased competition in considering program modifications.*
Question 7:	Are there other new data, information or developments that warrant consideration?
Metrics:	*Associated with habitat, nutrient and other species monitoring efforts.*
Response:	*Program refinements as appropriate.*

Figure 9. Examples of decision pathways guiding future monitoring and implementation of the Kootenai Sturgeon aquaculture program.

5.2 IN-SEASON MANAGEMENT PROCEDURE AND GOALS

The goal of the ISMP is to provide a structured decision-making framework that guide hatchery production goals, identifies M&E needs, and supports effective agency

cooperation consistent with the guidelines established each year. The Kootenai Tribe will implement the four-step ISMP (Figure 10) in cooperation with co-management agencies, research institutes, and stakeholders (as appropriate). The ISMP procedure is formalized in database(s) and a set of management tools, as well as through the APR to ensure consistency and accountability. The database will store and document data and assumptions, while management tools will use predictive models to arrive at outcomes from which decision guidelines and biological objectives may be derived. The tools document the basis for these targets and establish expectations for all performance indicators. They also will help simplify the implementation process and document the rationale for recommended annual restoration actions. KTOI's biologists and fellow co-managing agencies responsible for implementing in-season management will use these tools to prepare for the APR workshop, where analytical results will be presented and shared with all interested parties. The management tools used in the ISMP will be further refined over time through implementation of the ISMP and APR processes as new information is obtained and analyzed.

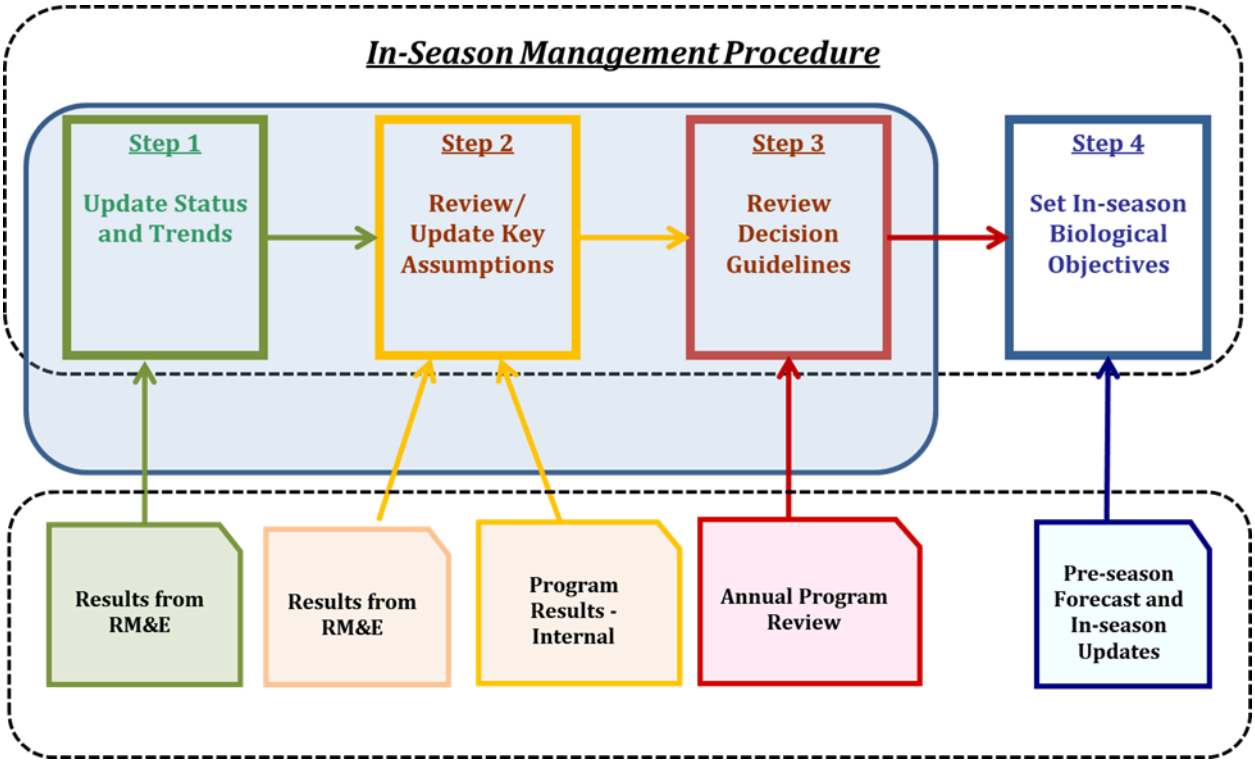

Figure 10. In-Season Management Procedure (ISMP) framework for the Kootenai Sturgeon M&E Plan.

This program has been designed to support flexible production and operations to accommodate inherent uncertainty and expected annual variability in broodstock abundance as the numbers of remaining wild Kootenai River broodstock continue to decline. This flexibility is reflected in the design and operation of the hatchery facilities and in the Decision Guidelines that determine annual hatchery production and the integration of future natural production.

5.2.1 Update Status and Trends Information

In this step, the most recent status and trends information are entered into the database for both the hatchery and natural production components of the population. The initial predictive tool focuses on hatchery production and survival of hatchery-reared Sturgeon. This tool will also be modified to integrate the contribution of any natural production into the population and program operations. This step will occur each year at the APR workshops.

5.2.2 Update Key Assumptions

The annual ISMP will integrate newly acquired data and analyses to update a set of key assumptions (see Table 4).

5.2.3 Review Decision Guidelines

Once the key assumptions and stock status have been updated, the Decision Guidelines (see Table 5) will be reviewed to determine if they need to be revised. This adaptive management step will occur at the APR workshop. Although not expected to change frequently, the Decision Guidelines may need to be periodically revised to account for 1) changes in conservation goals in the United States and Canada, 2) unsatisfactory goal achievement across the project area, 3) Libby Dam operations/habitat related issues, 4) new scientific discoveries, and 5) other changes in management or environmental conditions in the subbasin or the region.

The purpose of the Decision Guidelines is to ensure the Sturgeon hatchery program meets annual and longer-term goals for abundance and distribution. The goal of the Decision Guidelines for Kootenai River White Sturgeon is to establish a naturally reproducing population with an expanded geographic range within the Kootenai River increasingly like historical records. The Decision Guidelines are based on a set of key assumptions about our capability to accurately detect and respond to annual variation in abundance and availability of hatchery produced and natural origin spawners. This plan identifies the information needed to update and apply these guidelines and describes how data will be collected. KTOI expects to meet resource goals over time with relevant adaptive management actions, through collaboration with co-managing agencies and local entities.

5.2.4 Set Biological Objectives for the Coming Year

Biological objectives are set based on co-manager agreements following the APR process. Adult-population abundance estimates, hatchery facility capacity, research needs and broodstock collection targets are all factors that play into setting biological objectives for future years. Release strategies based on specific biological life-stages are also considered to plan for the following year while also meeting target juvenile production numbers. For example, the adult-abundance prediction models are typically updated annually and presented at the APR. All updates are entered into modeling and analysis tools and the

tool(s) then generate expected outcomes that may be used to guide hatchery production and set biological objectives for the program annually.

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Appendix A

Kootenai Tribe of Idaho
XXXX Activities Supporting Kootenai River White Sturgeon Restoration

Appendix A.

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