2017 LITTLE TRAPPER LAKE

SOCKEYE EGG TAKE

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1.0 INTRODUCTION

The sockeye egg take project at Little Trapper Lake was initiated in 2016 as part of a program to establish a self-sustained anadromous sockeye population in Trapper Lake. It was proposed that Little Trapper Lake sockeye stock would be used as the donor stock to re-colonize Trapper Lake with anadromous Sockeye salmon. The objectives of the Trapper Lake sockeye enhancement program are contained in previous feasibility and risk assessment reports (Mercer 2005, 2006). Little Trapper Lake sockeye were previously used as a donor stock for a sockeye enhancement project at Trapper Lake from 1990 –1994 (PSC 1998). The project is funded by the Northern Fund of the Pacific Salmon Commission (PSC). Technical direction and the Trapper Lake sockeye enhancement program objectives are determined by the Enhancement Sub-Committee of the Transboundary Technical Committee (TTC) of the PSC.

1.1 Description of the System

Little Trapper Lake is located in the Taku River system at the headwaters of Kowatua Creek, a tributary of the Inklin River (Figure 1). The Taku River drainage encompasses approximately 45,000 sq. km. of which 97% is in Canada. The Taku River empties into Taku Inlet in Southeast Alaska.

1.2 Little Trapper Lake Sockeye Stocks

Fisheries and Oceans Canada has contracted the operation of a sockeye enumeration weir at the outlet of Little Trapper Lake from 1983 through 2017. During this period, annual spawning escapements have averaged 11,177 (range 2,158 – 31,227). It is estimated that the Little Trapper sockeye stocks contribute 11% annually (1986-2014 average) to the total Taku River sockeye escapement (TTC 2015). The Little Trapper Lake escapement in 2017 was 6,552 (Table 2).

The Little Trapper Lake origin sockeye begin their migration up the Taku River in mid- June and continue through to early September, with peak numbers passing through the Little Trapper Lake weir during the first week in August. Mean travel time from the Taku Inlet to Little Trapper Lake is approximately 30 days. The condition of returning sockeye at the weir site ranges from bright to sexually mature, however the majority of fish entering the lake exhibit evidence of sexual maturation.

The Little Trapper Lake sockeye stock spawns in the inlet stream connecting Little Trapper and Trapper lakes. Approximately 90% of the spawning occurs within 1 km of the inlet into Little Trapper Lake.

Spawning occurs from mid August through September with peak spawning typically during the last week in August and first week in September.

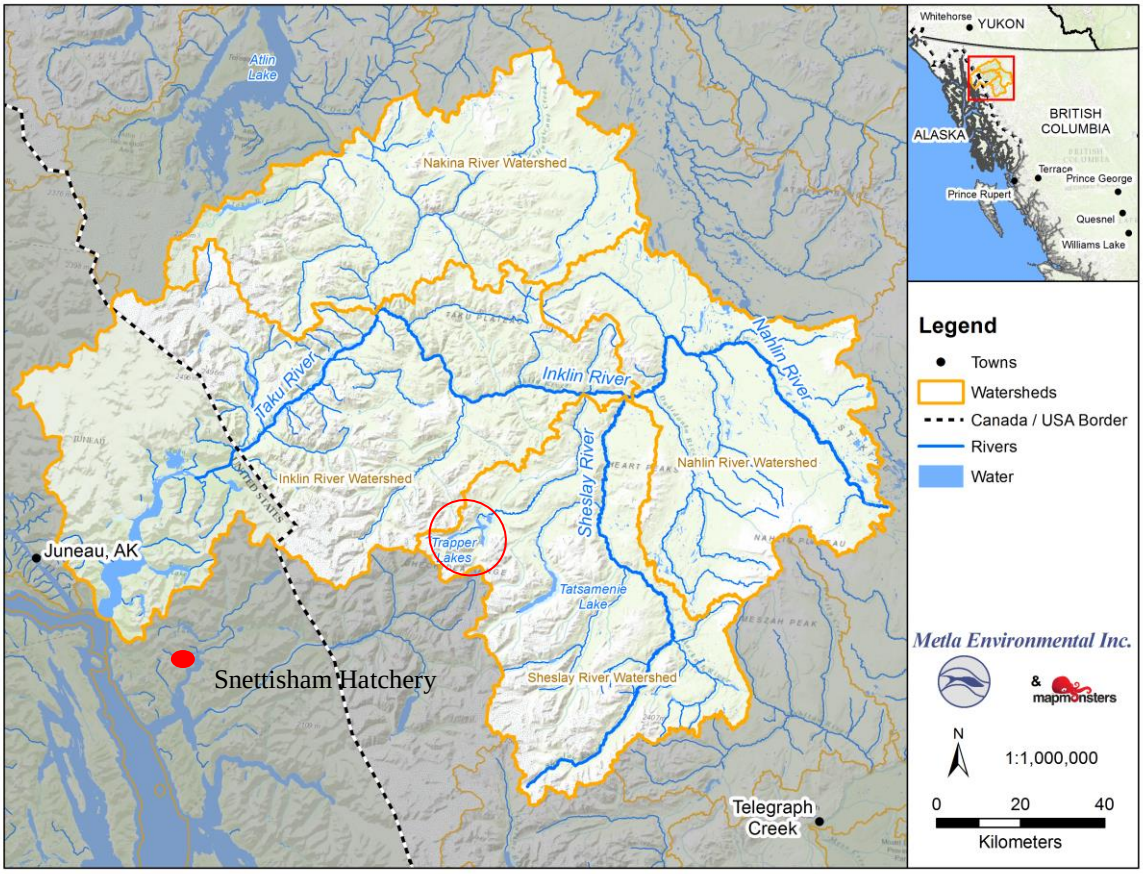

Figure 1. Location of Little Trapper Lake in Taku River watershed.

2.0 OBJECTIVES

The objectives of the 2017 Little Trapper Lake egg take were the following:

1. Capture enough female Sockeye broodstock to obtain 250,000 eggs (or 30% of the spawning escapement, whichever is less) and sufficient males for a 1:1 spawning ratio.

2. Conduct killing, spawning and fertilization procedures according to revised OIE Infectious Haematopoietic Necrosis Virus (IHNV) disinfection/avoidance protocol.

3. Transport fertilized water-hardened eggs by float plane or helicopter to the Snettisham hatchery, Alaska. The eggs are to be transported in coolers as prescribed by ADF&G protocol.

4. Maintain records detailing:
    (a) Number of males and females captured for broodstock
    (b) Number of fish retained for spawning by sex
    (c) Number of pre-spawning mortalities
    (d) Fish culture procedures
    (e) Time and method of transport
    (f) Ambient air and water temperature and egg temperature during transport.

3.0 METHODS

3.1 Mobilization

Required supplies and materials were purchased in Vancouver and Whitehorse and transported to Little Trapper Lake by float plane. Personnel, materials, and supplies were transported to the project site via the contractor's aircraft. Egg take personnel stayed at the contractor's camp at Little Trapper Lake.

3.2 Broodstock Capture, Transport, and Holding

Broodstock capture began on August 18 and continued through to September 1. The sockeye were captured in Little Trapper Lake at the mouth of the inlet stream. Capture was accomplished using a 100 m long and 8 m deep seine net. This net was set from the front of a boat and the captured sockeye crowded into shallow areas near shore. The sockeye captured were examined for their suitability as broodstock and either released in the lake or placed in a transport tub in a boat. Fish free of wounds and fungus and in advanced stages of sexual maturity were retained. The transport tub contained 200 litres of water to which was added 8 ml of "Aquacalm" (metomidate hydrochloride), resulting in a transport solution of 1 ppm. At this concentration, "Aquacalm" has a sedative effect on the transported fish that lasts up to 24 hours after application. Use of this tranquilizer facilitated ease of handling, reduced stress to the fish, and was found in previous sockeye transport/holding projects to reduce overall pre-spawn mortality. During transport the fish were supplied with supplementary oxygen by administering bottled oxygen at a rate of 0.5 l/min. via tygon tubing and a ceramic diffuser stone positioned on the bottom of the transport tub. Broodstock were

transported to a floating net pen anchored approximately 10m from the west shore and 100m from the inlet stream. Distance from the capture site to net pens was approximately 100 m. Approximately 10 - 15 fish were transported per trip.

Two floating net pens were assembled and used during the broodstock capture and sorting operation. One pen measured 3m x 7m x 2.2 m deep, and the other measured 3m x 3m x 2.2m deep. The pens were constructed using floating frames that were made by laminating 4 cm styrofoam between 2 pieces of 5 cm x 15 cm lumber. The upper margins of the knotless nylon net pens were fastened to the pen frames using fencing staples. Polyethylene tarps covered the pens and were secured with boards along the frame rim. The pens were anchored at a depth of about 6 m in the lake. The total pen volume was 65 m^3. Broodstock holding densities were approximately 4 fish/ m^3.

3.3 Sorting, Spawning, and Fertilization

The held broodstock were sorted by towing the net pens into shallow water near shore using a boat, removing the cover, and crowding the held fish into one end of the pen. The pen was then partitioned by sliding a pole under the net and securing it to the top of the net frame. Ripe, sorted fish were transferred to the smaller net pen in preparation for spawning the following day. The pen containing the sorted broodstock was re-covered and towed out into deeper water and secured to an anchored float.

A 3.5 m x 3.5 m tarp was erected to serve as a spawning facility. Tables to hold iodophore solutions, water, coolers, and egg cups were positioned around the inside perimeter of the structure. Killing, spawning, and fertilization was conducted according to current OIE IHNV avoidance protocol as described in Appendix 1. Ripe females were retrieved from the pen, killed with a club and dipped in a 200 ppm iodophore (ovadine™) solution. The ventral area of each fish was scrubbed with a brush dipped in the iodophore. The fish were then hung on a rack and bled through a knife cut to the carotid artery. The ventral area was then dried with a paper towel. Clean paper towels were used for each fish. A few eggs were expelled prior to stripping the eggs into a 2 litre plastic container. One container was used per female, with only loose eggs retained. Hands and all related equipment were disinfected prior to handling and processing each fish.

After being killed, males were dipped in a 200 ppm iodophore bath, the ventral area scrubbed with a brush, and the fish hung on a rack. The belly and vent area were then wiped dry. A small quantity of milt was expressed to avoid potential contamination with iodophor.

After the eggs were stripped excess ovarian fluid was removed from the egg container using a disposable pipette. Prior to fertilization the eggs were rinsed in a 0.9% saline solution and the solution drained. Milt from each male was expressed into 2 separate egg containers, each containing the eggs from 1 female. The process was repeated with a second male resulting in the fertilization of each female with two males. The egg container was gently swirled to mix eggs and milt before adding approximately 200 ml of virus free water to activate the sperm. A lid was placed over the container and the container swirled for thorough mixing. The eggs, milt and water mixture was left for approximately 1 minute to ensure fertilization. The activation water was decanted and a 100 ppm iodophore solution added to achieve a ratio of approximately 4 parts iodophore solution to 1 part egg mass. The eggs in the iodophore solution were gently mixed, left for approximately 5 minutes and then drained. Each container was labeled with a consecutive number and the time, and the eggs then left to water harden in the solution for at least 30 minutes.

After water hardening the solution was decanted and the eggs poured into a cooler lined with a plastic bag containing approximately 5 litres of virus free water (Figure 3). The eggs from 15 - 18 females were pooled into each cooler. Virus free water was added to each bag covering the eggs to a depth of 40mm to 50mm. The bags were twisted closed and sealed with a tight fitting elastic band. If required, crushed ice was placed under and around the bags in each cooler to lower the egg temperature. Labels were placed in each cooler detailing time of pooling and number of females.

The fecundity of spawned females was not calculated during the 2017 egg take. Instead an estimated fecundity was used to calculate the total eggs delivered. The estimated figure was based on the approximate mean fecundity calculated during previous Little Trapper Lake egg takes (3,300) as well as subjective estimation by the project manager. Accurate fecundities including the total number of eggs delivered were determined by Snettisham Hatchery during the egg picking process (Appendix 2).

Virus free water for milt activation, ice and pooling of eggs was obtained from a stream entering into Tatsamenie Lake. Tatsamenie Lake is located 20 km south of Little Trapper Lake and the water was transported to Little Trapper Lake via float plane. This water is from a steep gradient stream that has been successfully used for an egg take at Tatsamenie Lake for the past 27 years. There are no virus free water sources practically obtainable at Little Trapper Lake.

3.4 Egg Transport

Two egg take flights were conducted to Snettisham Hatchery using the contractor's aircraft.

3.5 Demobilization

All materials and equipment remain at Little Trapper Lake. The net pen frames and spawning shed were dismantled and returned to the contractor's residence. Egg transport coolers, net pens and related equipment were dried and stored at the contractor's facility.

4.0 RESULTS

4.1 Broodstock Capture and Holding

A total of 255 sockeye (120 males and 135 females) was captured and placed in the net pen. This represented 3.9% of the total escapement (6,552) into Little Trapper Lake. Broodstock capture occurred over 10 days from August 18 through August 27. The fish were held over a period of approximately 22 days.

Total female and male pre-spawn mortality was 0 fish (Table 1). The female and male broodstock pre-spawn mortality rate was the lowest that has been recorded for egg takes at this site. .

4.2 Spawning and Fertilization

The spawning and egg transport summary is listed in Table 1. A total of 88 females and 88 males were spawned. The estimated fecundity was 3,300 resulting in an estimated 290, 000 green eggs delivered to the hatchery. Subsequent to shocking and picking of the eggs, the measured mean fecundity determined by Snettisham Hatchery was 3,184. This resulted in an actual delivery of 280,000 green eggs (Table 1, Appendix 2). Green egg to eyed survival as determined by Snettisham Hatchery was 75.5 %. This resulted in 210,000 Little Trapper Lake origin eyed eggs incubating at Snettisham Hatchery.

4.3 Egg Transport

Both egg transport flights were conducted using fixed wing aircraft. The eggs from the first egg take were delivered the day after the egg take and from the second egg take the eggs were delivered the same day.

5.0 Discussion

The objectives of the 2017 egg take at Little Trapper Lake were achieved. Brood stock collection in 2017 started 5 days earlier than occurred in 2016. As a result of the longer holding period two egg takes were required to reduce the incidence of over ripe egg being collected. The first egg take was delayed by approximately 2 days due to poor flying weather. Scheduling of the egg take to coincide with weather conducive to flying to the hatchery resulted in the egg delivery occurring on the day following the first egg take. It is probable that the relatively lower green to eyed egg survival (75%), versus 85% in 2016, was a result of the spawning of some over ripe females in the first egg take.

The 2017 broodstock holding mortality was 0%. Low holding mortality is considered normal for this stock. The fertilization rates of the delivered eggs were lower than those reported for 2016, but the percentage of live seeded eggs (75%) was within the expected norm. It is anticipated that the Little Trapper Lake egg take project results could be repeated over the next three years in order to continue seeding Trapper Lake with anadromous Sockeye salmon.

Table 1. Little Trapper Lake 2017 egg take summary.

Egg Take No.	1	2				
Broodstock Sort Date	30-Aug	Sep-06				
Egg Take Date	1-Sep	Sep-09				
Date Eggs Delivered	02-Sep	Sep-09				
# Females Spawned	61	27				
Estimated Fecundity	3,300	3,300				
# Eggs Delivered	201,300	89,100				
Cum. Eggs delivered	201,300	290,400				
Cum. Male Prespawn Mort.	0	0				
Cum. Female Prespawn Mort.	0	0				
# BKD Samples	0	0				
# IHNV Samples	0	0				
# Otolith Samples	0	0				
Water Temp.	12.5	11.5				
Air Temp. (min)	14	12				
Transport Method	Maule	Maule				

			No. Males Held in Pens	120
No. Females Spawned	88		No. Females Held in Pens	135
No. Males Spawned	88		No. Females Released Unspawned	47
Average fecundity (estimated)	3,300		No. Males Released Unspawned	32
Estimated # Eggs To Hatchery	290,400		Male Prespawn Mortality	0.0%
% Survival to 100 CTU's			Female Prespawn Mortality	0.0%
Total weir count	6,552		Number females removed for broodstock plus holding mortality	88
Total female escapement at weir (un-weighted estimate)	3,780			

Table 2. Little Trapper Lake 2017 weir count.

									TAGS				Total			
	Count		SAMPLED		TOTAL		TAG SCARS		RECOVERED		NOT RECOVERED		Tags	WATER		
DATE	Daily	Cumulative	Daily	Cumulative	Daily	Cumulative	Fish Inspected	Observed	Daily	Cumulative	Daily	Cumulative		Temp°C	Level	Time
21-Jul														11	77	
22-Jul														12	80	
23-Jul	0	0	0	0	0	0	0	0	0	0	0	0	0	12	80	
24-Jul	0	0	0	0	0	0	0	0	0	0	0	0	0	12	81	9:00
25-Jul	0	0	0	0	0	0	0	0	0	0	0	0	0	12	80	9:00
26-Jul	0	0	0	0	0	0	0	0	0	0	0	0	0	12	80	9:00
27-Jul	0	0	0	0	0	0	0	0	0	0	0	0	0	12	80	9:00
28-Jul	0	0	0	0	0	0	0	0	0	0	0	0	0	12	80	9:00
29-Jul	0	0	0	0	0	0	0	0	0	0	0	0	0	12	80	9:00
30-Jul	0	0	0	0	0	0	0	0	0	0	0	0	0	11.5	80	9:00
31-Jul	0	0	0	0	0	0	0	0	0	0	0	0	0	11.5	78	9:00
01-Aug	0	0	0	0	0	0	0	0	0	0	0	0	0	12	75	9:00
02-Aug	0	0	0	0	0	0	0	0	0	0	0	0	0	12	74	9:00
03-Aug	0	0	0	0	0	0	0	0	0	0	0	0	0	12	75	9:00
04-Aug	0	0	0	0	0	0	0	0	0	0	0	0	0	12.5	76	9:00
05-Aug	100	100	0	0	100	100	0	0	2	2	1	1	3	13	76	9:00
06-Aug	579	679	50	50	629	729	50	0	7	9	1	2	11	13	77	9:00
07-Aug	741	1420	60	110	801	1530	60	0	9	18	0	2	20	14	80	9:00
08-Aug	305	1725	40	150	345	1875	40	0	5	23	0	2	25	14	81	9:00
09-Aug	320	2045	40	190	360	2235	40	0	6	29	0	2	31	14	82	9:00
10-Aug	405	2450	40	230	445	2680	40	0	9	38	0	2	40	14	81	9:00
11-Aug	491	2941	50	280	541	3221	50	0	9	47	1	3	50	14	80	9:00
12-Aug	262	3203	30	310	292	3513	30	0	4	51	0	3	54	14	80	9:00
13-Aug	165	3368	30	340	195	3708	30	0	5	56	0	3	59	14	79	9:00
14-Aug	68	3436	30	370	98	3806	30	0	4	60	0	3	63	13.5	79	9:00
15-Aug	40	3476	20	390	60	3866	20	0	1	61	0	3	64	13	81	9:00
16-Aug	117	3593	40	430	157	4023	40	0	4	65	0	3	68	13	81	9:00
17-Aug	66	3659	30	460	90	4119	30	0	4	69	0	3	72	13	81	9:00
18-Aug	72	3731	30	490	102	4221	30	0	3	72	0	3	75	13	82	9:00
19-Aug	60	3791	30	520	90	4311	30	0	3	75	0	3	78	12.5	81	9:00
20-Aug	23	3814	30	550	53	4364	30	0	2	77	0	3	80	12.5	79	9:00
21-Aug	75	3889	30	580	105	4469	30	0	2	79	0	3	82	12.5	79	9;30
22-Aug	171	4060	30	610	201	4670	30	0	5	84	1	4	88	13	80	8:30
23-Aug	113	4173	30	640	143	4813	30	0	2	86	0	4	90	13	80	9:00
24-Aug	78	4251	30	670	108	4921	30	1	3	89	0	4	93	12.5	80	9:00
25-Aug	132	4383	30	700	162	5083	30	0	2	91	1	5	96	12.5	79	9:00
26-Aug	192	4575	30	730	222	5305	30	0	5	96	1	6	102	12.5	80	8:30
27-Aug	105	4680	20	750	125	5430	20	0	3	99	0	6	105	12.5	80	9:00
28-Aug	51	4731	20	770	71	5501	20	0	3	102	0	6	108	12.5	71	9:00
29-Aug	184	4915	10	780	194	5695	10	0	10	112	0	6	118	12.5	70	9:00
30-Aug	88	5003	10	790	98	5793	10	0	2	114	0	6	120	13	70	9:00
31-Aug	101	5104	0	790	101	5894	0	0	2	116	1	7	123	12.5	70	9:00
01-Sep	21	5125	0	790	21	5915	0	0	2	118	0	7	125	12.5	74.5	9:00
02-Sep	143	5268	10	800	153	6068	10	0	3	121	1	8	129	12.5	76	9:00
03-Sep	44	5312	0	800	44	6112	0	0	2	123	0	8	131	12	75	9:00
04-Sep	86	5398	0	800	86	6198	0	0	2	125	0	8	133	12	78	9:00
05-Sep	122	5520	0	800	122	6320	0	0	6	131	0	8	139	12	76	9:00
06-Sep	142	5662	0	800	142	6462	0	0	12	143	0	8	151	12	78	9:00
07-Sep	66	5728	0	800	66	6528	0	0	2	145	0	8	153	11.5	86	9:00
08-Sep	9	5737	0	800	9	6537	0	0	0	145	0	8	153	11.5	86	9:00
09-Sep	8	5745	0	800	8	6545	0	0	0	145	0	8	153	11.5	84	9:00
10-Sep	7	5752	0	800	7	6552	0	0	0	145	0	8	153	11.5	81	9:00

References

Transboundary Technical Committee (TTC). 2015. Preliminary Estimates of Transboundary River Salmon Production, Harvest and Escapement and a Review of Joint Enhancement Activities in 2015.

Appendix 1. ADF&G and OIE avoidance protocols.

STEP	ADF&G Procedure	OIE Recommended Procedure	Fall of 2016 Little Trapper Lake Procedure
Pre-fertilisation	• Collect eggs from individual females into separate disposable containers.	• Eggs separated from ovarian fluid prior to being rinsed in 0.9% saline (30-60 seconds).	• Eggs separated from ovarian fluid prior to being rinsed in 0.9% saline (30-60 seconds).
Fertilisation	• Add milt into each egg container. It is critical that the elapsed time for milt addition not exceed 30 to 45 seconds prior to mixing and/or water addition. • Activate the milt using virus free water.	• Sperm added and fertilisation allowed to proceed for 5-15 minutes. (*No description of activation process)	• Add milt into each egg container. It is critical that the elapsed time for milt addition not exceed 30 to 45 seconds prior to mixing and/or water addition. • Activate the milt using virus free water.
Post-Fertilisation	• Rinse the eggs with 100 ppm iodophore solution, discarding the rinse. • Repeat this procedure until solution is clear of organic material.	• Rinse with 0.9% saline (30-60 seconds) to remove excess sperm and other organic materials.	• Rinse with 0.9% saline (30-60 seconds) to remove excess sperm and other organic materials.
Water Hardening	• Refill egg container by forcefully adding (fall of about 12 inches) clean 100 ppm iodophore solution. • Maintain the original dark brown color for one hour by periodic addition of more iodophore solution. • After one hour, pour off the iodophore solution and rinse eggs with virus free water.	• W.H. begins with 1 minute rinse with 100 ppm iodophore. • That solution is then discarded and replaced with a fresh 100 ppm solution (1:4 egg to solution ratio) with the eggs disinfected for a further 30 minutes. • Water hardening should be finished using clean water.	• W.H. begins with 1 minute rinse with 100 ppm iodophore. • That solution is then discarded and replaced with a fresh 100 ppm solution (1:4 egg to solution ratio) with the eggs disinfected for a **further 30 minutes.** • The eggs should then be finished water hardening and transported in fresh IHNV free water.

Appendix 2. Egg receipt summary at Snettisham Hatchery for Little Trapper Lake eggs, 2017.

TRAPPER LAKE
EGGTAKE - EGG PICKING SUMMARY
BROOD YEAR 2017

MODULE IM-3
Hatch Code: TRAPPER17 4,2,3H

EGGTAKE DATE	NUMBER OF DAYS DELAYED	EGG LOT NUMBER	EGG TEMP AT RECEIPT (CORE) AVE.	100 CTU % SURVIVAL	ESTIMATED FECUNDITY	ACTUAL FECUNDITY	NUMBER OF FEMALES	ESTIMATED GREEN EGGS	ACTUAL GREEN EGGS	DEAD EGGS PICKED	LIVE EYED EGGS	GREEN TO EYED SURVIVAL	OTM DESIGNATION
09/01/17	1	Trap 1	0.4	72.1%	3,400	3,199	61	207,400	195,147	60,459	134,688	69.0%	**4,2,3H**
09/09/17	0	Trap 2	3.6	91.0%	3,300	3,150	27	89,100	85,047	9,390	75,658	89.0%	**4,2,3H**
Totals & Averages			**2.0**	**81.6%**	**3,369**	**3,184**	**88**	**296,500**	**280,194**	**69,849**	**210,345**	**75.1%**	

Base Marked Group for **TRAPPER17** = **210,345**