2019 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 2008). 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 2019. During this period annual spawning escapements have averaged 10,365 (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 sockeye escapement in 2019 was 6,382 (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.

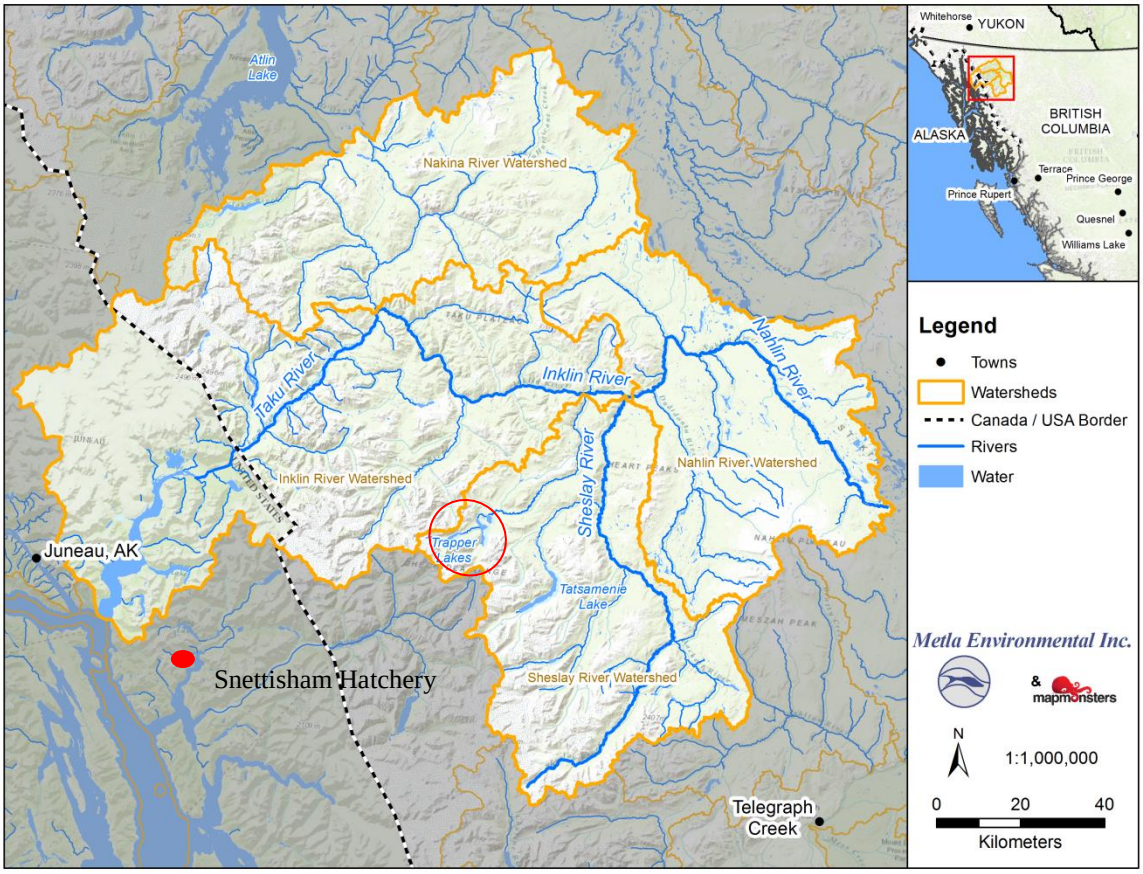

Figure 1. Location of Little Trapper Lake in Taku River watershed.

2.0 OBJECTIVES

The objectives of the 2019 Little Trapper Lake egg take were the following:

1. Capture enough female Sockeye broodstock to obtain 500,000 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

Little Trapper Lake is remote and accessible by air only. Required supplies and materials were 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 16 and continued through to August 20. 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 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 4.2 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 portable tent structure 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 2019 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 28 years. There are no virus free water sources practically obtainable at Little Trapper Lake.

3.4 Egg Transport

Two egg take flights were conducted from Little Trapper Lake 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 444 sockeye (170 males and 274 females) was captured and placed in the net pen. This represented 7% of the total escapement (6,382) into Little Trapper Lake. Broodstock capture occurred over 5 days from August 16 through August 20. The fish were held over a period of approximately 21 days. Total female and male pre-spawn mortality was 7 (2.5%) and 10 (5.8%) respectively (Table 1).

4.2 Spawning and Fertilization

The spawning and egg transport summary is listed in Table 1. A total of 130 females and 88 males were spawned. The estimated fecundity was 3,300 resulting in an estimated 429, 000 green eggs delivered to the hatchery. Green egg to 100 CTU survival as determined by Snettisham Hatchery was 71.5 % (Appendix 2).

4.3 Egg Transport

Both egg transport flights were conducted using fixed wing aircraft. The eggs from both egg takes were delivered to the hatchery the same day they were taken.

5.0 Discussion

The objectives of the 2019 egg take at Little Trapper Lake were not fully achieved in that the number of eggs collected was approximately 71,000 less than the goal of 500,000. After the second fish sort on September 6 the decision was made by the project manager to take the eggs as soon as possible rather than wait until sufficient ripe broodstock were available to meet the 500,000 egg target. This decision was

primarily based on the poor egg quality observed during egg take #1 on September 4. It was observed the broodstock were not holding well and that waiting longer would result in lower egg quality and increased holding mortality. In addition, the weather was conducive to fixed wing air transport on September 6 but was forecast to deteriorate for several days thereafter. It was decided by the project manager that the number of eggs delivered to the hatchery was sufficiently near the egg target to not warrant a third small (approximately 20 females) egg take.

Overall the egg quality of the 2019 Little Trapper Lake egg take (71.5% survival to 100 CTU's) was lower than the egg quality encountered in 2017 (75%) and 2016 (85%). A total of 7 spawned and fertilized females were subsequently discarded following the water hardening process. This was due to the high number of dead eggs observed. It is not known why the egg quality was less than average in 2019. In general it was observed the fish did not hold as well in 2019 compared to past years as evidenced by the higher than average holding mortality and the general poor physical condition of the held broodstock. The broodstock exhibited more dermal abrasion, fin wear and fungus than was observed in previous egg takes at this site. The cause of this is not known as the procedures used in 2019 were similar to those used in previous years.

It is suggested that if annual egg targets continue at the levels of 500,000 or more that additional net pens be employed to reduce the holding densities. If sockeye escapements allow a larger number of broodstock could also be captured which would reduce the holding period required. Holding more broodstock would reduce the length of time required to obtain sufficient ripe fish to meet the egg targets.

Table 1. Little Trapper Lake 2019 egg take summary.

Egg Take No.	1	2
Broodstock Sort Date	02-Sep	Sep-06
Egg Take Date	4-Sep-19	6-Sep-19
Date Eggs Delivered	4-Sep-19	6-Sep-19
# Females Spawned	72	58
# Females Spawned and Discarded	4	3
Estimated Fecundity	3,300	3,300
# Eggs Delivered	237,600	191,400
Cum. Eggs delivered	237,600	429,000
Cum. Female Prespawn Mort.	1	7
# IHNV Samples	0	28
Water Temp.	11.5	11.5
Air Temp. (min)	14	12
Transport Method	Cessna 185	Cessna 185

No. Females Spawned	130		No. Males Held in Pens	170
No. Males Spawned	88		No. Females Held in Pens	274
Average fecundity (estimated)	3,300		No. Females Released Unspawned	86
Estimated # Eggs To Hatchery	429,000		No. Males Released Unspawned	54
% Survival to 100 CTU's	71.5%		Male Prespawn Mortality	6.2%
Total weir count	6,342		Female Prespawn Mortality	3.1%
Total female escapement at weir (un-weighted estimate)	4,531		Number Females removed for broodstock plus holding mortality	141

Table 2. Little Trapper Lake 2019 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
23-Jul	0	0	0	0	0	0	0	0	0	0	0	0	0	12	86	
24-Jul	0	0	0	0	0	0	0	0	0	0	0	0	0	12	84	9:00
25-Jul	0	0	0	0	0	0	0	0	0	0	0	0	0	12	82	9:00
26-Jul	0	0	0	0	0	0	0	0	0	0	0	0	0	12	82	9:00
27-Jul	0	0	0	0	0	0	0	0	0	0	0	0	0	12	79	9:00
28-Jul	0	0	0	0	0	0	0	0	0	0	0	0	0	12	79	9:00
29-Jul	0	0	0	0	0	0	0	0	0	0	0	0	0	12	79	9:00
30-Jul	0	0	0	0	0	0	0	0	0	0	0	0	0	13	75	9:00
31-Jul	332	332	20	20	352	352	20	0	4	4	0	0	4	13	72	9:00
01-Aug	163	495	30	50	193	545	30	0	3	7	0	0	7	13	72	9:00
02-Aug	48	543	0	50	48	593	0	0	3	10	0	0	10	12	71	9:00
03-Aug	367	910	20	70	387	980	20	0	6	16	1	1	17	12	71	9:00
04-Aug	369	1279	30	100	399	1379	30	0	1	17	0	1	18	13	71	9:00
05-Aug	106	1385	20	120	126	1505	20	0	2	19	0	1	20	13.5	71	9:00
06-Aug	663	2048	20	140	683	2188	20	0	12	31	1	2	33	13	75	9:00
07-Aug	352	2400	40	180	392	2580	40	0	4	35	0	2	37	14	79	9:00
08-Aug	204	2604	50	230	254	2834	50	0	3	38	0	2	40	13.5	80	9:00
09-Aug	157	2761	50	280	207	3041	50	0	5	43	1	3	46	13.5	78	9:00
10-Aug	41	2802	0	280	41	3082	0	0	1	44	0	3	47	13.5	75	9:00
11-Aug	230	3032	20	300	250	3332	20	0	3	47	1	4	51	13.5	74	9:00
12-Aug	150	3182	40	340	190	3522	30	0	2	49	0	4	53	13.5	71	9:00
13-Aug	191	3373	40	380	231	3753	40	0	2	51	1	5	56	14	68	9:00
14-Aug	62	3435	10	390	72	3825	10	0	0	51	0	5	56	14	70	9:00
15-Aug	103	3538	20	410	123	3948	20	0	3	54	0	5	59	14	72	9:00
16-Aug	103	3641	20	430	123	4071	20	0	3	57	0	5	62	14	74	9:00
17-Aug	139	3780	10	440	149	4220	10	0	9	66	0	5	71	13.5	76	9:00
18-Aug	121	3901	30	470	151	4371	30	0	2	68	0	5	73	13	82	9:00
19-Aug	155	4056	30	500	185	4556	30	0	2	70	1	6	76	12.5	77	9:00
20-Aug	138	4194	30	530	168	4724	30	0	2	72	0	6	78	12	71	9:00
21-Aug	103	4297	30	560	133	4857	30	0	3	75	0	6	81	12	67	9:00
22-Aug	42	4339	30	590	72	4929	30	0	1	76	0	6	82	11.5	61	9:00
23-Aug	116	4455	30	620	146	5075	30	0	4	80	0	6	86	11.5	60	9:00
24-Aug	62	4517	30	650	92	5167	30	0	3	83	0	6	89	11	60	9:00
25-Aug	32	4549	30	680	62	5229	30	0	2	85	0	6	91	11	63	9:00
26-Aug	53	4602	20	700	73	5302	20	0	3	88	0	6	94	11	66	9:00
27-Aug	206	4808	20	720	226	5528	20	0	7	95	0	6	101	11	72	9:00
28-Aug	78	4886	20	740	98	5626	20	0	2	97	0	6	103	11.5	75	9:00
29-Aug	209	5095	20	760	229	5855	20	0	2	99	0	6	105	11	74	9:00
30-Aug	175	5270	10	770	185	6040	10	0	2	101	0	6	107	11	71	9:00
31-Aug	10	5280	10	780	20	6060	10	0	1	102	0	6	108	11.5	67	9:00
01-Sep	23	5303	10	790	33	6093	10	0	0	102	0	6	108	12	66	9:00
02-Sep	10	5313	10	800	20	6113	10	0	1	103	0	6	109	11.5	66	9:00
03-Sep	54	5367	0	800	54	6167	0	0	0	103	0	6	109	11	67	9:00
04-Sep	8	5375	0	800	8	6175	0	0	0	103	0	6	109	11.5	70	9:00
05-Sep	98	5473	0	800	98	6273	0	0	3	106	0	6	112	11	69	9:00
06-Sep	36	5509	0	800	36	6309	0	0	0	106	0	6	112	11	67	9:00
07-Sep	13	5522	0	800	13	6322	0	0	1	107	0	6	113	11	66	9:00
08-Sep	20	5542	0	800	20	6342	0	0	1	108	0	6	114	10.5	65	9:00
09-Sep	30	5572	0	800	30	6372	0	0	1	109	0	6	115	11.5	64	9:00
10-Sep	3	5575	0	800	3	6375	0	0	0	109	0	6	115	11.5	62	9:00
11-Sep	1	5576	0	800	1	6376	0	0	0	109	0	6	115	11.5	61	9:00
12-Sep	6	5582	0	800	6	6382	0	0	0	109	0	6	115	11.5	61	9:00

References

Mercer 2008. Trapper Lake Sockeye Enhancement Project *Evaluation Studies 2006-2007* Unpublished report for Pacific Salmon Commission Northern Fund.

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, 2019.

EGGTAKE DATE	DATE RECEIVED	EGG LOT NUMBER	MODULE NUMBER / INCUBATOR	NUMBER OF FEMALES	EGG TEMP. AT RECEIPT (RANGE L-H)	EGG TEMP. AT RECEIPT (CORE) AVE.	INCUBATOR LOADING TEMP.	ESTIMATE FECUNDITY	ESTIMATED GREEN EGGS	100 CTU % SURVIVAL
09/04/19	09/04/19	1	IM-3 / 9	72	2.0 - 6.1	4.3	7.3	3,300	237,600	66.5%
09/06/19	09/06/19	2	IM-3 / 10	58	1.1 - 7.6	4.7	7.6	3,300	191,400	76.5%
Totals & Averages				**130**		**4.5**	**7.5**	**3,300**	**429,000**	**71.5%**