

## Article

# Seasonal Trends in Water Retention of Atlantic Sea Cucumber (*Cucumaria frondosa*): A Modeling Approach

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**Abstract:** Sea cucumbers are widely consumed as a delicacy or in eastern medicine across many Asian countries. Due to the depletion of traditional stocks, new species are increasingly harvested, including the Atlantic sea cucumber (*Cucumaria frondosa*), the most abundant, cold-water species found in the North Atlantic. This species is harvested in NAFO subdivision 3Ps off the south coast of Newfoundland and Labrador, Canada. As part of their respiration, stress response, and locomotion, sea cucumbers draw and retain oxygenated water within their body cavity, resulting in significant water content at landing. Historically, Fisheries and Oceans Canada (DFO) have applied a 23% deduction to the landed weight to account for this water retention. To validate this deduction, the authors conducted experiments across thirteen sampling events in 2019 and 2020. Randomized samples were collected during offloading and were categorized into three sizes of bin—small ( $x \leq 150$  g), medium ( $150 \text{ g} < x \leq 250$  g), and large ( $x > 250$  g)—and water loss was measured. Water loss was analyzed in relation to multiple factors, including processor, unloading method, year, license, month, fishing area, hold location, size, and processing method. Key findings included the following: (a) sea cucumbers typically contained more than 23% free water; (b) large and medium-sized specimens, which dominated landings, retained more free water; (c) water loss was highest for the samples collected from the top of the hold; (d) the unloading method influenced free water retention, as did the processing method used to cut the sea cucumbers; (e) license, processor, and fishing area had strong collinearity with other factors or were not found to be statistically significant; and (f) water loss appeared higher in 2020 than 2019, largely due to the increased use of vacuum transfer methods. Based on these findings, DFO revised the water retention allowance to 34%.

**Keywords:** *Cucumaria frondosa*; sea cucumber; water loss; drain weight; processing

**Key Contribution:** This research quantified and modelled the water-holding capacity of landed Atlantic sea cucumbers as a percentage of body weight. The findings demonstrated that the historic deduction of 23% underestimated actual water content, leading to an industry-wide adjustment to 34% and ensuring more accurate weight assessments and regulatory practices.



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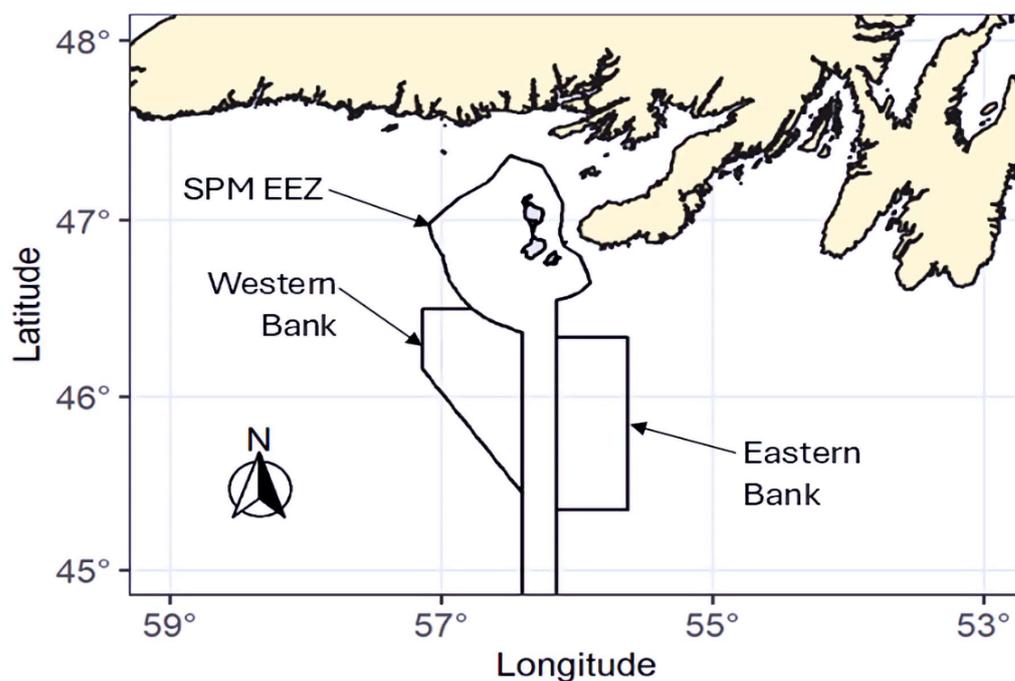
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## 1. Introduction

Harvested since the third century B.C. [1], sea cucumbers are presently consumed in countries such as China, Korea, Japan, Malaysia, Indonesia and Russia [2]. Rich in protein, vitamins, essential amino and fatty acids, and mineral content while low in fat and cholesterol [3], sea cucumbers are also valued in traditional eastern medicine for their

purported anticancer, anti-hypertensive, anti-angiogenic, anti-inflammatory, antidiabetic, anti-coagulation, antimicrobial, antioxidation, and anti-osteoclastogenic properties [2,4].

There are more than 70 species of sea cucumbers that are commercially harvested [5]. Overfishing of traditional stocks [6,7] has resulted in the development of new sea cucumber fisheries in Chile, Mexico, Egypt, USA and Canada, and the introduction of several new species to the market [8]. One example, the Atlantic sea cucumber (*Cucumaria frondosa*), is the most abundant cold-water species found in the North Atlantic [2]. *C. frondosa* is native to the waters of Newfoundland and Labrador, Canada. Harvesting began in the North Atlantic Fisheries Organization (NAFO) subdivision 3Ps as an emerging fishery in 2003 and transitioned to a commercial fishery in 2012 [9]. Figure 1 depicts the two main concentrations of sea cucumber in the Canadian portion of the St. Pierre Bank; one lies northwest (Western Bank) and one lies southeast (Eastern Bank) of the French Exclusive Economic Zone (EEZ) [9]. The market for these products has been growing steadily [10]; landed volumes in Newfoundland and Labrador have increased from 454 MT in 2003 to 6084 MT in 2022, with a landed value of CAD 9.73 M [2,11]. The primary commercial products produced in Newfoundland and Labrador are frozen or dried, eviscerated and cleaned body walls.



**Figure 1.** Map of the Eastern Bank and Western Bank sea cucumber fishing areas in Newfoundland and Labrador. Map data from the Global Administrative Areas (GADM) database (<http://gadm.org/> (accessed on 23 June 2022)). St. Pierre and Miquelon (SPM) Exclusive Economic Zone (EEZ) data from the Marine Regions database (<https://www.marineregions.org/gazetteer.php?p=details&id=8494> (accessed on 23 June 2022)). Figure generated using ggplot2 v3.3.6 [12].

To facilitate respiration, sea cucumbers ingest large volumes of oxygenated water into their body cavity through their anus [13]; enabling them to transition from a limp to a rigid state. Additionally, by taking in seawater, they increase their buoyancy and move along the ocean floor using the ocean current [14]. When confronted with danger or after harvesting stress, sea cucumbers contract their body, retract their tentacles into their mouth, close their mouth and anus, and hold the water in their cavity [7].

Some sea cucumber species lose up to 30% of their cavity water in the first 60 min after removal from their aquatic environment [15]. In the case of *C. frondosa*, 48 h after harvesting, the cavity water content can still be very high, ranging 20–80% [16], making

it challenging to identify the expected body wall yield from hail weights. If handled correctly after harvesting and during storage in the vessel, sea cucumber should remain rigid; Gianasi, Hamel & Mercier [17] have reported that the optimal cooling medium for transporting *C. frondosa* consists of iced seawater. When improperly handled, critically weak or dead animals were observed to be limp with their tentacles distended and have off odors. Consequently, free water or drained weight serves as an important indicator of *C. frondosa* health [18], and both harvesters and processors must carefully balance quality and yield.

Water loss assessments conducted by commercial *C. frondosa* fishery proponents have not been documented in the public literature. Historically, Fisheries and Oceans Canada (DFO) deducted 23% of the landed weight as water weight [19]. However, local processors' proprietary data indicated that this deduction underestimated the free water content, prompting a request for an independent, standardized study. This study comprehensively quantified the free water held by these animals to better explain the relationship between live weight, product yield, and quality. The investigation also examined the impact of harvest location, seasonality, and handling methods on water loss and yield. This was accomplished by performing comprehensive water loss assessments which are reported within. Furthermore, the rationale behind this research was twofold: to optimize sea cucumber production and enhance the final product quality. The anticipated outcomes include more accurate sea cucumber quota allocations, a consistent method for assessing the free-water content in whole specimens, and overall improvements in the quality of the raw product supply.

## 2. Materials and Methods

### 2.1. Sample Collection and Preparation

Five samplings were collected and analyzed during the 2019 deployment and eight were collected and analyzed during the 2020 deployment. Highlights of each sample are presented in Table 1. The equipment used for each sampling is itemized in Appendix A.1.

**Table 1.** Overview of sample collection details.

Date	Offloading Location	Fishing Area	Hail Weight (lbs.) <sup>1</sup>	Offloading Method
22 August 2019	OCI, St. Lawrence	Western Bank	60,000	Vacuum
30 August 2019	OCI, St. Lawrence	Eastern Bank	90,000	Vacuum
21 September 2019	QuinSea, Fortune	Western Bank	50,000	Bucket
27 September 2019	OCI, Fortune	Eastern Bank	90,000	Bucket
11 October 2019	OCI, St. Lawrence	Eastern Bank	55,000	Vacuum
22 July 2020	OCI, St. Lawrence	Eastern Bank	63,000	Vacuum
29 July 2020	QuinSea, Fortune	Western Bank	96,000	Bucket
11 August 2020	QuinSea, Fortune	Western Bank	95,000	Bucket
3 September 2020	Clearwater Grand Bank	Eastern Bank	45,000	Vacuum
17 September 2020	Green, Burin	Western Bank	55,000	Vacuum
29 September 2020	Fogo, Riverhead	Eastern Bank	60,000	Vacuum
7 October 2020	Clearwater Grand Bank	Eastern Bank	60,000	Vacuum
14 October 2020	OCI, St. Lawrence	Eastern Bank	33,000	Vacuum

<sup>1</sup> Newfoundland and Labrador sea cucumber harvesters typically report hail weights in lbs.

#### 2.1.1. Sample Collection from the Vessel

Samples were collected from fishing vessels using the following procedure.

1. The following information was collected from the processor or unloading facility supervisor and recorded the following prior to sampling:
  - a. vessel information;
  - b. fishing area;

- c. hail weight.
2. Using the hail weight, the load was divided into top, middle and bottom based on storage position in the boat's hold.
3. Unloading was executed using one of the following two methods, depending on the facility:
  - a. Unloading staff shoveled product into a bucket which was hoisted out of the hold and loaded into a tote (bucket method).
  - b. Sea water was pumped into the hold and product was transferred over a drain screen to the tote using a vacuum pump (vacuum transfer).
4. As the vessel was unloaded, the following was undertaken:
  - a. Two 2000 L or three 1000 L insulated totes were randomly selected and identified using Number Generator Random, v 2.1 [20] from each of the top, middle, and bottom sections of the hold.
  - b. Once filled, these totes were segregated and used for sample collection.
  - c. No water was added to the process.
5. The Transcell scale was tared for the weight of the Stacknest pans and ~40 kg of sea cucumber samples were randomly transferred from each segregated tote into each pan.
  - a. Each pan was weighed and tagged to identify the collection time, weight, and sampling location.
  - b. In total, 18 pans were collected, six from each of the bottom, middle and top of the hold.
6. The remaining product in each tote was returned for processing.

#### 2.1.2. Subsample Collection for Evaluation

Subsamples were collected for evaluation using the following procedure.

1. The Ohaus and Western scales were tared for the weight of the bus pans and the Transcell scale was tared for the weight of the StackNest pans.
2. Beginning with the six StackNest pans collected from the top layer, sea cucumbers were randomly selected one at a time from each StackNest pan, weighed, and sorted based on landed round weight ( $x \leq 150$  g = small,  $150 < x \leq 250$  g = medium, and  $x > 250$  g = large). These size bands were selected based on the range of available animal sizes and were used throughout the project.
3. An amount of 12 sea cucumbers, 40 each of small, medium and large size, were collected from each layer for water loss testing.
4. The sampling time, net weight of the sample, and sample weighing time (to estimate the time duration between sampling and weighing) were recorded on the data collection form (Supplementary Materials).
5. Once sufficient samples were collected, the expelled water remaining in the StackNest pan was weighed and recorded on the data collection form.
6. Each bus pan was tagged with the size, location, and time the sample was collected.
7. Approximately 230 random individuals were weighed for each of the top, middle, and bottom layers at each landing to estimate a size distribution for the project.
8. The remaining sea cucumbers were returned to the processor for processing.
9. This procedure was repeated for the middle and bottom layers of sea cucumber.

### 2.1.3. Quantifying Water Loss in Samples

To calculate water loss, samples were weighed at 0, 10, 20 and 30 min after cutting using the following procedure.

1. The Ohaus and Western scales were tared for the weight of the bus pans.
2. Beginning with the small sea cucumbers collected from the top layer of the hold, we undertook the following:
  - a. A set of five animals was selected at random.
  - b. Two sets were taken at a time, one for the longitudinal cut and one for the flower cut (Figure 2).



**Figure 2.** Sea cucumber cutting methods. **(Left)** longitudinal cut: the body wall is cut along its length between two muscle bands. **(Right)** flower cut: the body wall is cut transversely just behind the aquapharyngeal bulb (flower).

3. Each set of five individuals was weighed and the weight recorded.
4. Each individual was cut, the water sack broken, and placed back in the bus pan.
  - a. In the case of flower cut samples, the trimmings were saved.
5. To calculate the initial water loss, each set of five animals was weighed again immediately after cutting and the weight recorded.
  - a. In the case of flower cut samples, the trimmings were weighed for each set and the weight was recorded.
6. Each set was placed on the lump roe drain screen to drain.
7. Steps 1–6 were repeated for each set of five individuals in the sample group (e.g., top layer, small animals).
8. The water remaining in the bus pan was weighed and recorded.
9. The weight of each of the eight sample sets was measured after draining for 10, 20 and 30 min and recorded.
10. This procedure was repeated for each of the nine sample groups.

A total of 1440 observations were recorded during the 2019 program and 2200 observations for the 2020 program. These are broken down in Table 2.

**Table 2.** Water loss data point collection.

Description	2019	2020
Trips	5	8
Hold locations	3	3
Sea cucumber sizes	3	3
Cut methods	2	2
Sets	4	4
Water loss times	4	4
Total data points	1440	2200 <sup>1</sup>

<sup>1</sup> No middle location was sampled on the 29 September 2020 trip due to time constraints. Two small samples are missing from the 19 October 2020 set due to the size distribution of animals harvested during that trip.

## 2.2. Data Analysis

The estimated water-holding loss as a percentage of sample weight for each sample (e.g., small animals from the top section of the hold) was calculated using Equation (1).

$$L = 100 (\sum d_j) (\sum w_j)^{-1} + 100 D (\sum W_{2k})^{-1} \quad (1)$$

where  $L$  = total water loss for each sample set (%),  $d_j$  = weight of water lost in each StackNest pan (kg),  $w_j$  = weight of product in each StackNest pan (kg),  $j$  = StackNest pan number (1–6),  $D$  = total weight of lost water measured in bus pans (kg),  $W_{2k}$  = total weight of each set of 5 sea cucumbers (kg), and  $k$  = bus pan number (1–9).

The total weight for each sample set assumed that water loss while in storage was proportional to the weight of the set and was estimated using Equation (2).

$$W_T = W_i + 0.01 W_i L \quad (2)$$

where  $W_T$  = total weight for each sample (kg),  $W_i$  = initial weight recorded for each sample (kg), and  $L$  = total water loss while being held for each sample set (%).

## 2.3. Trim Loss

As discussed in Section 2.1.3, the weight of material trimmed from the samples prepared using the flower cut method was recorded for each sample set of five sea cucumbers. As this material was not included in the water loss study, it was removed to calculate the round weight using Equation (3). The round weight was used as the starting weight for each water loss test and trim weight was added back in to obtain the total weight.

$$W_R = W_T - W_t \quad (3)$$

where  $W_T$  = total weight for each sample (kg),  $W_t$  = trim weight for each sample set (kg), and  $W_R$  = round weight for each sample set (kg). In the case of longitudinally cut sea cucumbers,  $W_R = W_T$ .

## 2.4. Total Water Loss for Each Sample

Each set of five animals was cut using one of the two cutting methods, before being reweighed and recorded. Each set was then placed on the lump roe drain screen, reweighed and recorded at  $t = 10, 20$  and  $30$  min. Water loss (kg) was calculated using Equation (4) and water loss (% of wet weight) using Equation (5).

$$FW_{dt} = W_R - W_{dt} \quad (4)$$

where  $FW_{dt}$  = water loss measured at  $dt$  (kg),  $W_R$  = round weight for each sample set (kg),  $W_{dt}$  = weight for each sample set (kg) measured at  $dt$ , and  $dt$  = drain time (0, 10, 20, 30 min).

$$FWp_{dt} = 100 FW_{dt} W_T^{-1} \quad (5)$$

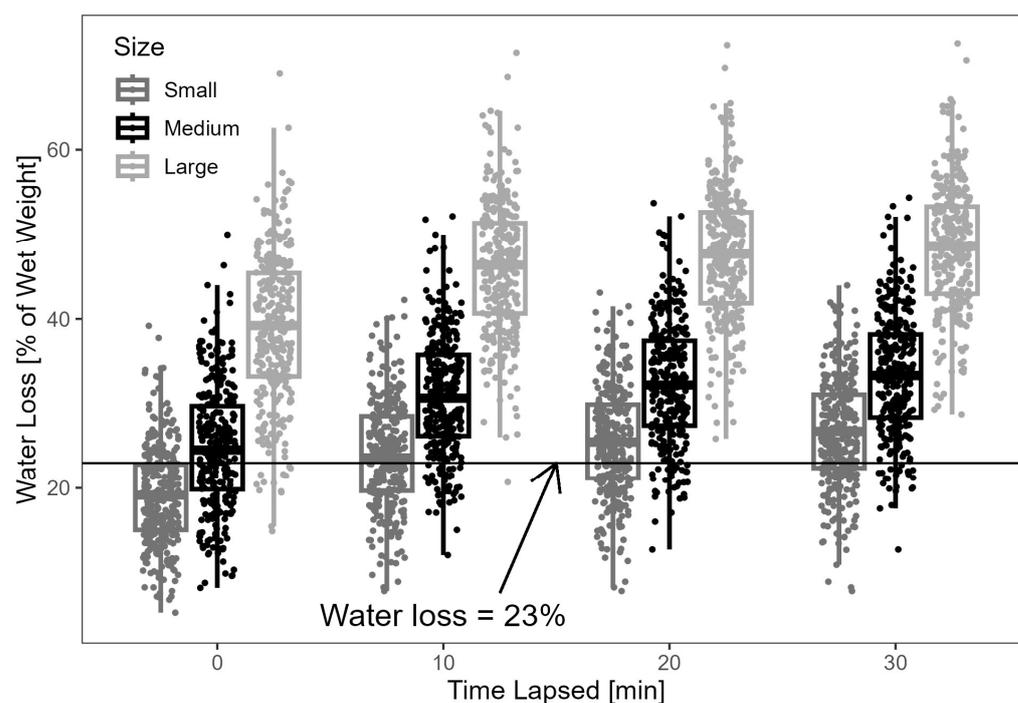
where  $FWp_{dt}$  = water loss (% of wet weight) measured at  $dt$ ,  $FW_{dt}$  = water loss (kg) measured at  $dt$ ,  $W_T$  = total weight for each sample set (kg), and  $dt$  = drain time (0, 10, 20, 30 min).

### 3. Results

The raw data collected during the 13 site visits are provided in the Supplementary Data folder. The analyses of water loss were conducted at 0, 10, 20 and 30 min after cutting.

#### 3.1. Preliminary Water Loss Analysis

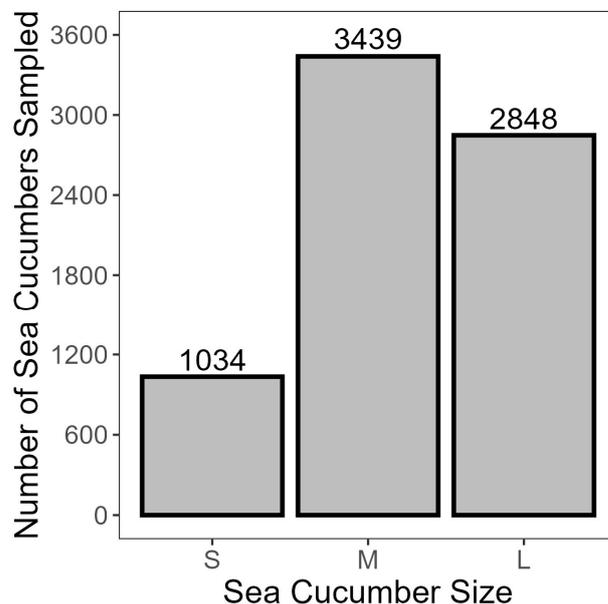
Water loss data were plotted as a function of animal size and drain time (Figure 3). This figure shows that the majority of water loss data appear to be greater than 23%. It also shows that medium and large sea cucumbers lose more water, as a percentage of wet weight, than small sea cucumbers and that water loss appears to increase with drain time from 0 to 30 min.



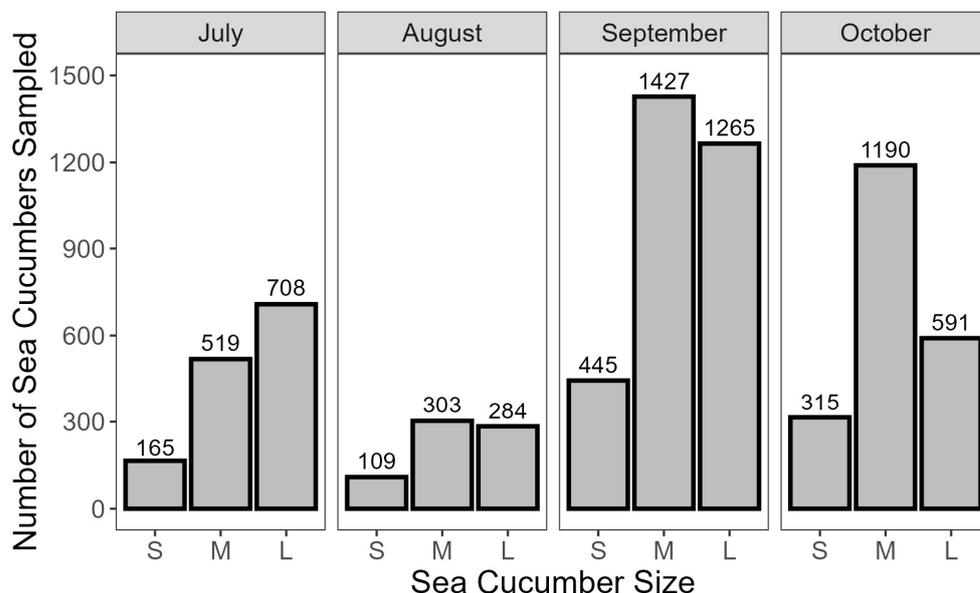
**Figure 3.** Boxplots showing water loss as a percentage of landed weight grouped by animal size (small, medium and large) and water loss time (0, 10, 20 and 30 min) recorded during the 2019 and 2020 fishing seasons. Total observations,  $n = 3640$ . Horizontal lines within each box represent the median water loss. The lower and upper edge of each box represents the first and third quartiles, respectively. The lower and upper whiskers show water loss values within the 95% confidence interval but outside of the interquartile range. Dots show individual readings. The horizontal line shows the 23% water content allowance deducted by DFO from landings during this study. Figure prepared using ggplot2 v3.3.6 [12].

Figure 4 presents the size distributions for sea cucumbers sampled between September 2019 and October 2020. This figure shows that the majority of sea cucumbers sampled (85.9%) are in the medium (47.0%) to large (38.9%) size range, further validating the belief among some industry participants that 23% water weight is a low estimate. Furthermore,

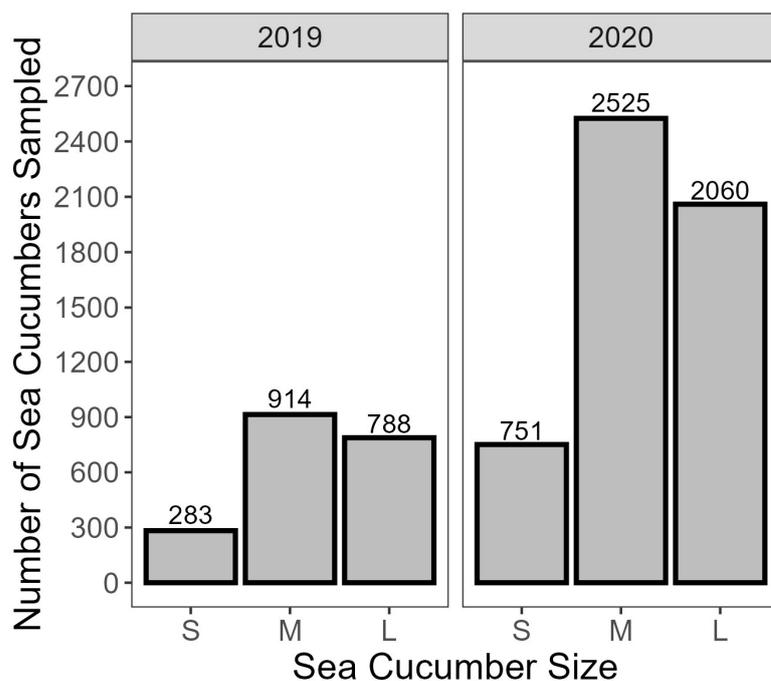
breaking the size distribution down by month (Figure 5), year (Figure 6), hold location (Figure 7) and fishing area (Figure 8) consistently shows substantially less small sea cucumber. The absence of smaller sea cucumber implies that higher water loss percentages from medium and large individuals are more prevalent.



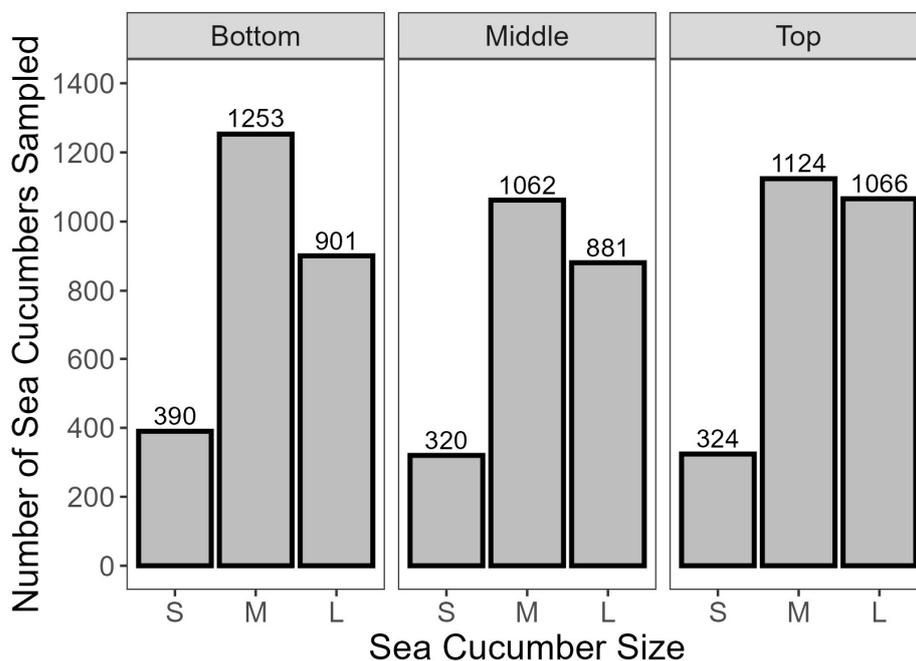
**Figure 4.** Histogram showing the sea cucumber size distribution for samplings taken between September 2019 and October 2020. Total observations,  $n = 7321$  sea cucumbers. The number placed over each bar gives the total number of samples counted for that size bin. Small (S) sea cucumbers make up 14.1% of the sea cucumbers sampled, medium (M) sea cucumbers make up 47.0% of the sea cucumbers sampled, and large (L) sea cucumbers make up 38.9% of the sea cucumber sampled. Figure prepared using ggplot2 v3.3.6 [12].



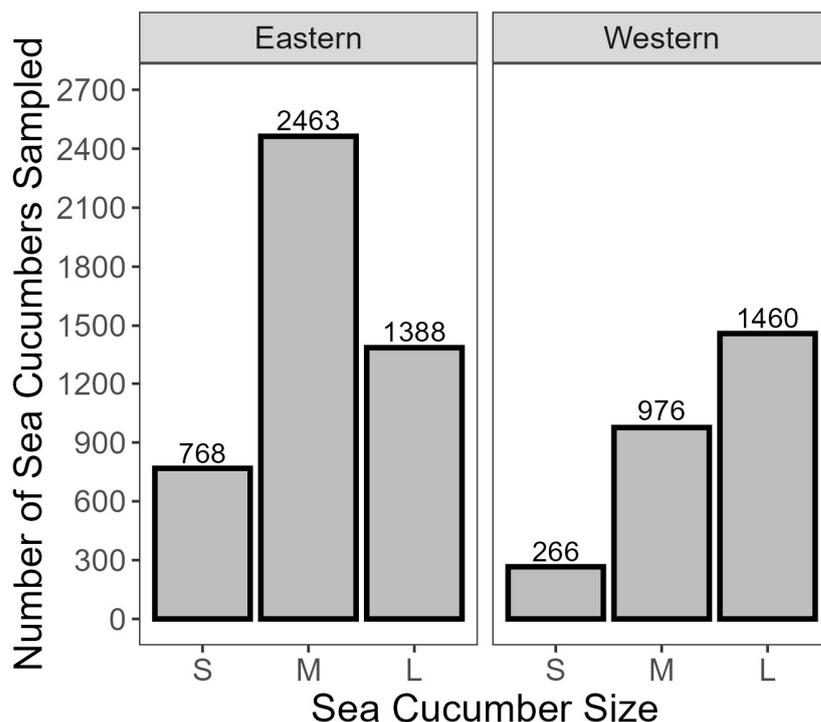
**Figure 5.** Histogram showing sea cucumber size distributions for samplings taken between September 2019 and October 2020 broken down by harvest month (July 2020, August 2020, September 2019 and 2020, October 2019 and 2020). Total observations,  $n = 7321$  sea cucumbers. The number placed over each bar gives the total number of samples counted for that size bin and month. In each sub-figure, the number of small (S) sea cucumbers is substantially less than the number of medium (M) or large (L) sea cucumbers. Figure prepared using ggplot2 v3.3.6 [12].



**Figure 6.** Histogram showing sea cucumber size distributions for samplings taken between September 2019 and October 2020, broken down by harvest year. Total observations,  $n = 7321$  sea cucumbers. The number placed over each bar gives the total number of samples counted for that size bin and year. In each sub-figure, the number of small (S) sea cucumbers is substantially less than the number of medium (M) or large (L) sea cucumbers. Figure prepared using ggplot2 v3.3.6 [12].



**Figure 7.** Histogram showing sea cucumber size distributions for samplings taken between September 2019 and October 2020, broken down by location in the vessel's fish hold. Total observations,  $n = 7321$  sea cucumbers. The number placed over each bar gives the total number of samples counted for that size bin and fish hold location. In each sub-figure, the number of small (S) sea cucumbers is substantially less than the number of medium (M) or large (L) sea cucumbers. Figure prepared using ggplot2 v3.3.6 [12].



**Figure 8.** Histogram showing sea cucumber size distributions for samplings taken between September 2019 and October 2020, broken down by fishing area. Total observations,  $n = 7321$  sea cucumbers. The number placed over each bar gives the total number of samples counted for that size bin and fishing area. In both sub-figures, the number of small (S) sea cucumbers is substantially less than the number of medium (M) or large (L) sea cucumbers. Figure prepared using ggplot2 v3.3.6 [12].

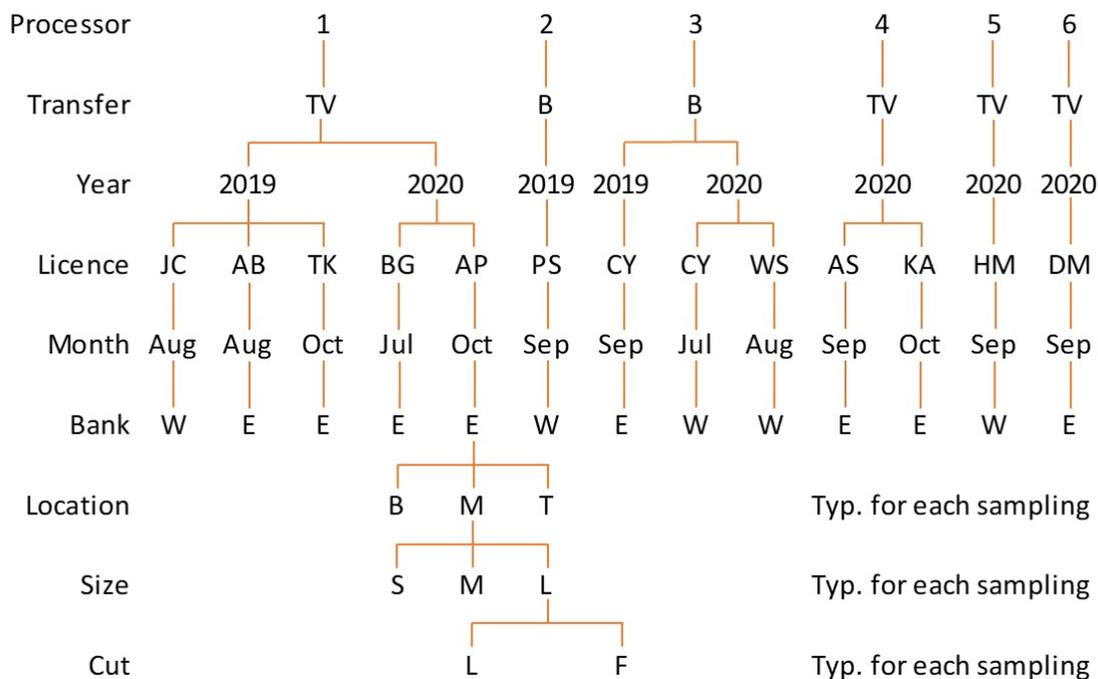
### 3.2. Statistical Analysis

The team assessed the data collected in accordance with the method proposed by Zuur & Ieno [21] using R v4.1.3 [22] along with Tidyverse v1.3.1 [23], Lubridate v1.8.0 [24], Statmod v1.4.37 [25] and MASS v7.3-58.1 [26]. The hypotheses analyzed were as follows: (A) The average total water loss for sea cucumbers exceeds 23% after 0, 10, 20, and 30 min of drain time; (B) water loss as a percentage of initial weight for large sea cucumbers is greater than for smaller sea cucumbers; (C) seasonality affects water-holding capacity, hence, water loss; and (D) water loss for boats unloaded using vacuum transfer are greater than those unloaded by bucket.

Key water loss data collected or calculated during each sampling are presented in Table 3 and their data dependency structure is presented in Figure 9.

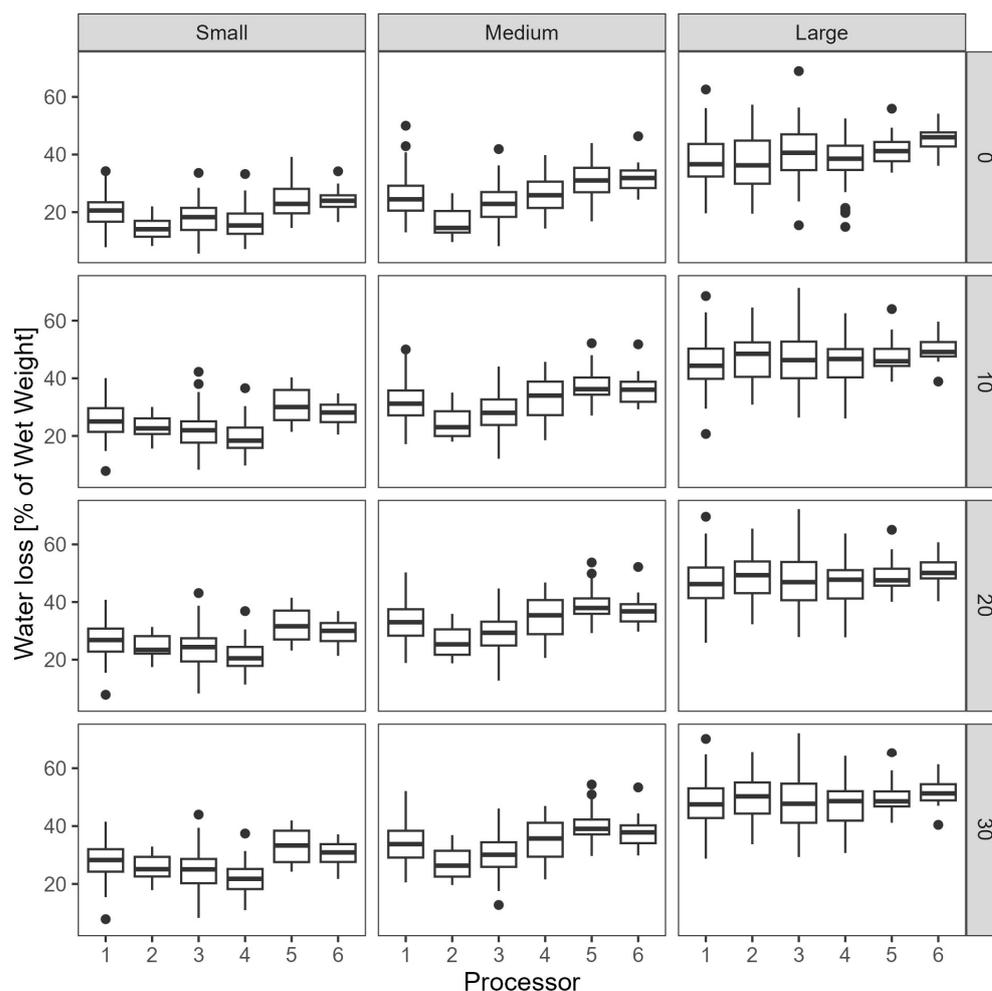
**Table 3.** Key variables collected or computed during the two-year water loss sampling program.

Variable	Type	Levels	Units	Values
License (L)	Factor	12		AB, AP, AS, BG, CY, DM, HM, JC, KA, PS, TK, WS
Processor (P)	Factor	6		1, 2, 3, 4, 5, 6
Bank (B)	Factor	2		Eastern, Western
Transfer (T)	Factor	2		Bucket, vacuum transfer
Month (M)	Factor	4		Oct, Sept, Aug, Jul
Year (Y)	Factor	2		2019, 2020
Location (L)	Factor	3		Bottom, middle, top
Size (S)	Factor	3		Small, medium, large
Cut (C)	Factor	2		Flower, longitudinal
Lapsed (t)	Integer		mins	10, 20, 30, 40
TLapsed (tl)	Integer		mins	84–661
Free water (FW)	Number		kg	0.034–2.786
Free water % (FWp)	Number		%	5.28–72.46



**Figure 9.** Data dependency structure for water loss experiments. This defines the interdependency between the processor or unloading site (randomly assigned integer from 1 to 6); transfer method used (bucket (B) or vacuum transfer (TV)); sampling year (2019 or 2020); license (AB, AP, AS, BG, CY, DM, HM, JC, KA, PS, TK, WS); harvest month (July, August, September, October); fishing area (Eastern or Western bank); fish hold location (bottom, middle, top); sea cucumber size (small, medium, large); and cutting method (flower cut or longitudinal cut). Location, size and cut are typical for each sampling. For clarity, location, size and cut show the typical dependency structure for one level above.

To avoid common errors, data exploration was performed in accordance with the method proposed by Zuur, Ieno & Elphick [27]. Salient findings included the following. (A) There were no obvious outliers attributable to data entry errors. (B) Data appeared to be relatively homogenous. Variances in water loss were observed when measured across processors (Figure 10); however, these were within acceptable levels [28]. (C) Water loss (as a % of wet weight) was found to be continuous, positive, and skewed to the right at all water loss time intervals (Figure 11). Therefore, the authors propose a Gamma generalized linear model (Gamma GLM) [29]. (D) All water loss measurements were positive; therefore, there was no risk of zero trouble. (E) License showed significant collinearity with year, month and processor, and was deleted from the statistical model because the study is concerned with seasonality. This was consistent with the data dependency structure (Figure 9). After retesting the revised model, the processor showed significant collinearity with year. Processor was therefore deleted because the study was more focused on the effect of seasonality. The revised model was retested, and bank showed significant collinearity with transfer and month. Bank was removed because it could be explained by the transfer method and month. One final test of the model revealed some collinearity between transfer and month. This could be explained by the lack of bucket unloading samples and ignored. (F) Possible interactions included transfer and size, transfer and month, month and year, and year and size, and these were assessed during data modelling.

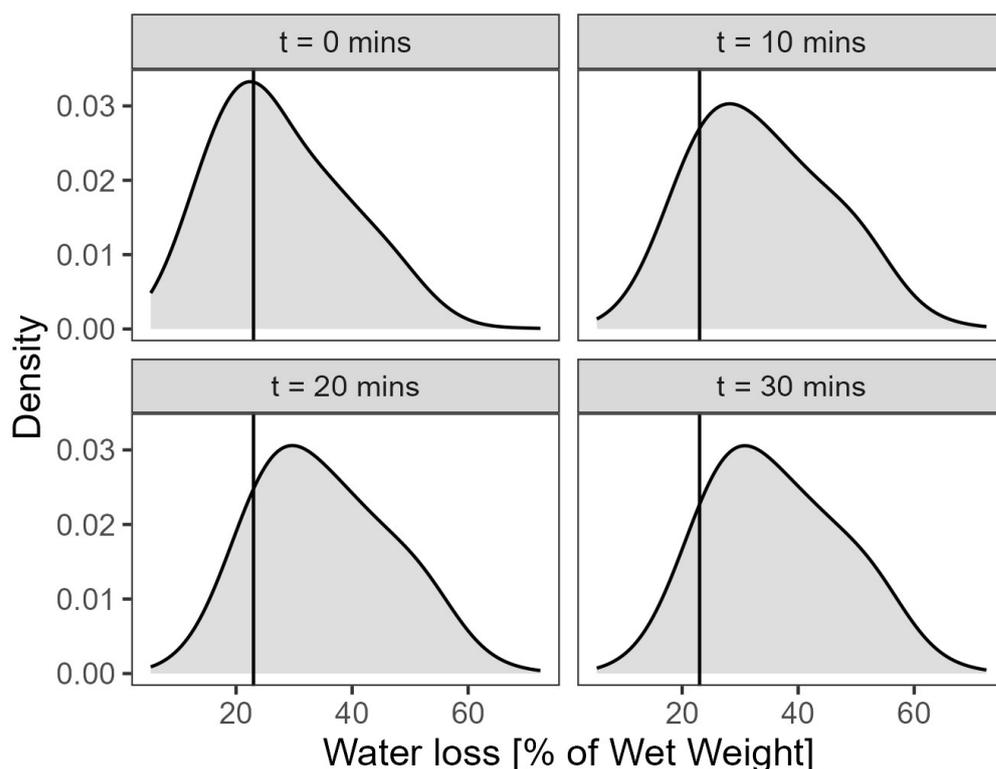


**Figure 10.** Boxplot comparing water loss against processor, faceted horizontally by size (small, medium and large) and faceted vertically by lapsed water drain time (0, 10, 20 and 30 min). Horizontal lines within each box represent the median water loss. The lower and upper edges of each box represent the first and third quartiles, respectively. The lower and upper whiskers show water loss values within the 95% confidence interval (CI) but outside of the interquartile range. Black dots show values outside the 95% CI. The ratio between the greatest and the smallest variances appears to be less than four and therefore should not impair the least-squares estimate [28]. Figure prepared using ggplot2 v3.3.6 [12].

Following data exploration, the dependency structure was assumed to be as follows: the dependent variable, percent water loss, was modeled as a function of the independent variables: lapsed (time after cutting), size, month, year, location, transfer, and cut. Percent water loss was also modeled as a function of the following interactions: transfer and size, transfer and month, month and year, and year and size. These independent variables and interactions were assessed further during the modelling process. Based on the results of data exploration, a Gamma distribution with an identity link was selected for the analysis [29] (Equation (6)).

$$\begin{aligned}
 \text{FWp} &\sim \Gamma(\kappa_i, \theta_i) \\
 E &= \mu_i = \kappa_i \theta_i \\
 \sigma^2_i &= \kappa_i \theta^2_i
 \end{aligned}
 \tag{6}$$

where FWp = water loss (% of wet weight),  $\Gamma$  = Gamma distribution,  $\kappa$  = shape factor,  $\theta$  = scale parameter,  $\mu$  = mean, and  $\sigma^2$  = variance.



**Figure 11.** Probability density figure showing the likelihood of a water reading faceted by lapsed drain time (0, 10, 20 and 30 min). The data appear to be continuous, positive, and right-skewed for each time interval. Vertical line shows 23% water loss. Figure prepared using ggplot2 v3.3.6 [12].

Given an identity link, the proposed model is presented in Equation (7).

$$\eta_i = \mu_i = \alpha + \beta_1 t_i + \beta_2 S_i + \beta_3 M_i + \beta_4 Y_i + \beta_5 L_i + \beta_6 T_i + \beta_7 C_i + \beta_8 T_i S_i + \beta_9 T_i M_i + \beta_{10} M_i Y_i + \beta_{11} Y_i S_i \quad (7)$$

where  $\eta$  = link function,  $\mu$  = expected value,  $\alpha$  = intercept,  $\beta$  = coefficients of the fitted model.

The model defined in Equation (7) was fit using R v4.1.3 [22]. The interactions month  $\times$  transfer and month  $\times$  year were not found to be statistically significant ( $p$ -value  $> 0.05$ ) using the Chi-squared test [30] and were removed from the model (Equation (8)).

$$\eta_i = \mu_i = \alpha + \beta_1 t_i + \beta_2 S_i + \beta_3 M_i + \beta_4 Y_i + \beta_5 L_i + \beta_6 T_i + \beta_7 C_i + \beta_8 T_i S_i + \beta_9 Y_i S_i \quad (8)$$

This model was fit, and the results are summarized in Table 4.

**Table 4.** Significance table for model defined using Equation (8).

	Df	Deviance	Scaled Dev	$p$ (>Chi)
<none>		176.81		
Lapsed	1	214.50	833.34	$<2.2 \times 10^{-16}$
Mon	3	182.50	125.93	$<2.2 \times 10^{-16}$
Location	2	190.69	306.97	$<2.2 \times 10^{-16}$
Cut	1	178.66	41.12	$1.43 \times 10^{-10}$
Size:Transfer	2	180.88	90.11	$<2.2 \times 10^{-16}$
Size:Year	2	178.88	45.75	$1.16 \times 10^{-10}$

To confirm that the identity link fit the data best, Equation (8) was fit using R v4.1.3 [22] with link = inverse and link = log and tested using the Bayesian information criteria (BIC) [31]. Based on BIC, link = identity fit the model best.

The initial control treatment for this model assumes that lapsed = 0 min, size = small, month = October, year = 2019, location = bottom, transfer = bucket, and cut = flower. The coefficients of each variable and their standard errors are presented in Table 5 and the 95% confidence intervals are presented in Table 6 for the fitted model. This model has an explained deviance of 65.5%.

**Table 5.** Model summary for the water loss model (Equation (8)) being fit using Gamma GLM (link = identity) using R v4.1.3 [22].

	Estimate	Std. Error	t	p (>  t )
(Intercept)	12.936	0.422	30.669	$<2 \times 10^{-16}$
Lapsed	0.277	0.009	30.010	$<2 \times 10^{-16}$
Size = medium (M)	2.980	0.405	7.364	$2.19 \times 10^{-13}$
Size = large (L)	23.201	0.614	37.802	$<2 \times 10^{-16}$
Mon = Sep	1.778	0.295	6.019	$1.93 \times 10^{-09}$
Mon = Aug	3.538	0.318	11.120	$<2 \times 10^{-16}$
Mon = Jul	1.687	0.377	4.477	$7.82 \times 10^{-06}$
Year = 2020	0.883	0.306	2.882	$3.98 \times 10^{-03}$
Location = middle	0.149	0.296	0.624	0.533
Location = top	3.927	0.255	15.421	$<2 \times 10^{-16}$
Transfer = vacuum (TV)	3.662	0.316	11.603	$<2 \times 10^{-16}$
Cut = longitudinal	1.311	0.203	6.445	$1.31 \times 10^{-10}$
Size = M × transfer = TV	2.287	0.468	4.886	$1.07 \times 10^{-06}$
Size = L × transfer = TV	−4.257	0.666	−6.395	$1.81 \times 10^{-10}$
Size = M × year = 2020	3.098	0.462	6.710	$2.25 \times 10^{-11}$
Size = L × year = 2020	1.604	0.631	2.542	0.011

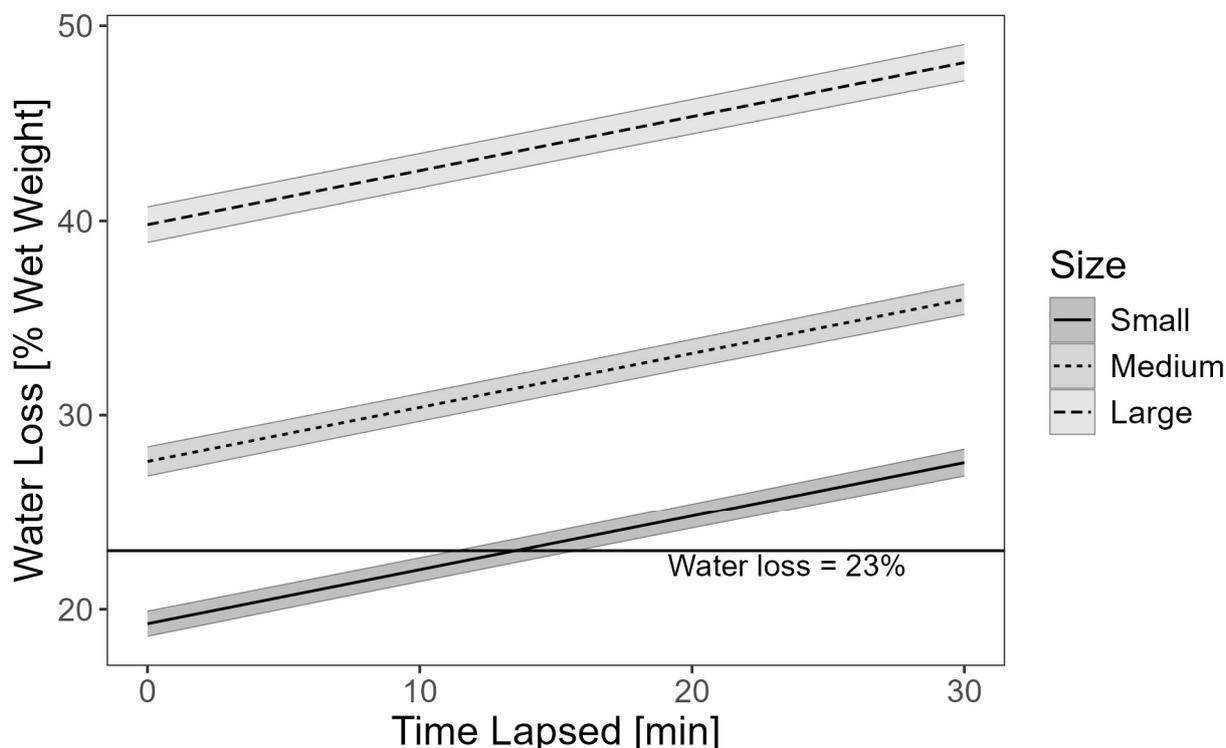
**Table 6.** Confidence intervals calculated for the water loss model (Equation (8)), fit using Gamma GLM (link = identity) using R v4.1.3 [22].

	2.5%	97.5%
(Intercept)	12.111	13.771
Lapsed	0.258	0.296
Size = medium (M)	2.176	3.789
Size = large (L)	22.008	24.415
Mon = Sep	1.200	2.355
Mon = Aug	2.916	4.160
Mon = Jul	0.946	2.432
Year = 2020	0.284	1.481
Location = middle	−0.322	0.621
Location = top	3.428	4.427
Transfer = vacuum (TV)	3.040	4.428
Cut = longitudinal	0.910	1.713
Size = M × transfer = TV	1.372	3.201
Size = L × transfer = TV	−5.569	−2.956
Size = M × year = 2020	2.197	4.000
Size = L × year = 2020	0.364	2.840

#### 4. Discussion

The effect of drain time for each size class of sea cucumber based on the model defined in Equation (8), is presented in Figure 12. This plot shows that the expected water loss for medium and large sea cucumber exceeds the current allowance of 23% at all drain times. Small sea cucumber are expected to exceed the 23% threshold after approximately 14 min of draining. However, based on sampling data from this study, small sea cucumber

account for only 14.1% of the population (Figure 4) and therefore, processors and harvesters can generally expect that the water content in landed sea cucumbers will exceed the 23% allowance.

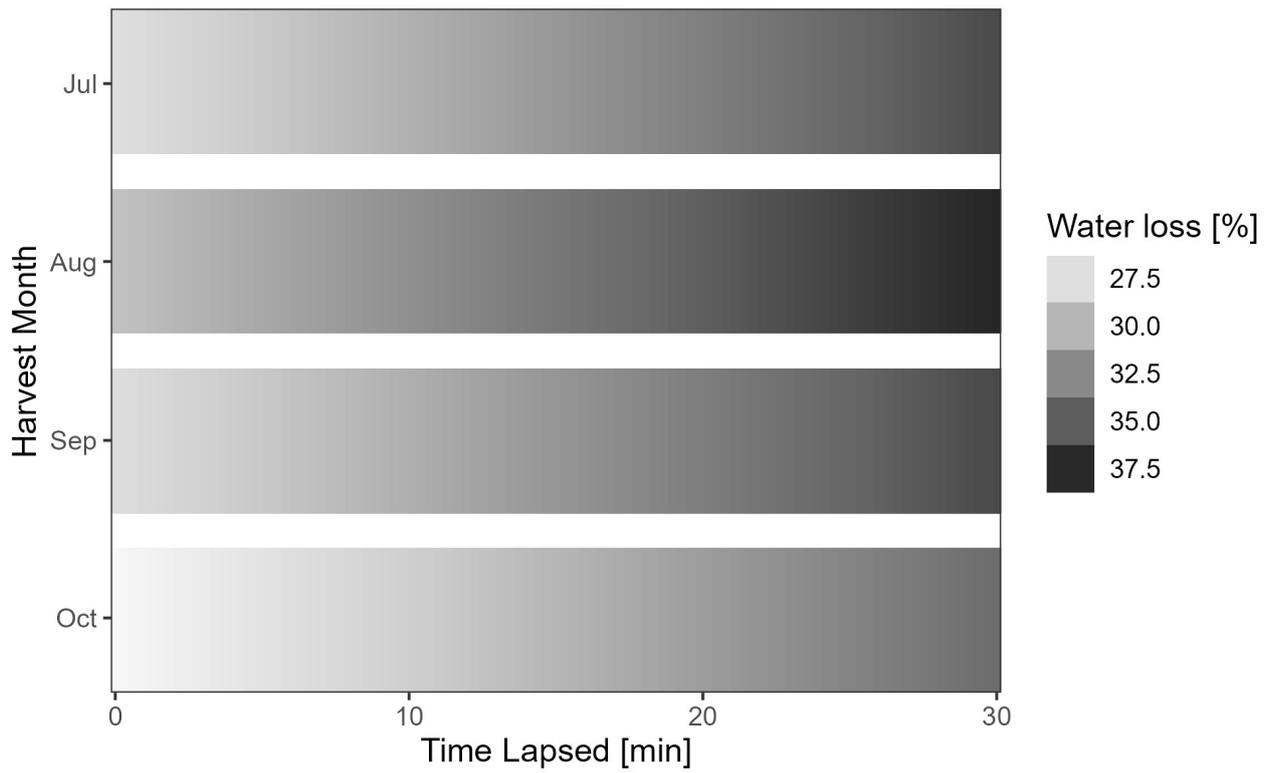


**Figure 12.** Expected water loss as a percentage of wet weight is plotted versus lapsed drain time with best-fit line and confidence bands shown for each size class. Figure produced using Visreg v2.7.0 [32].

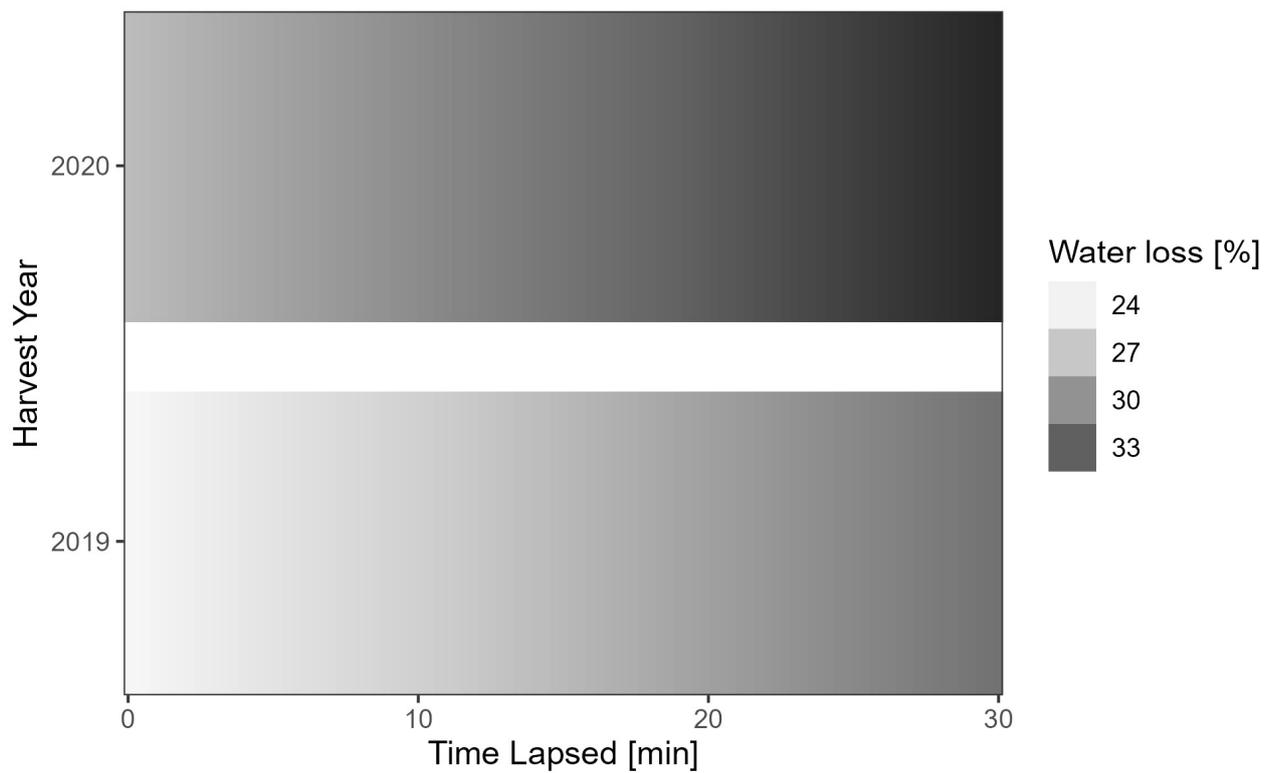
The effect of seasonality on water loss is presented in Figures 13 and 14. Based on model predictions from the data collected, sea cucumber harvested in August are expected to contain the highest percentage of water, while October samples are expected to contain the lowest. The anomaly observed in an early October sample may be attributed to factors such as sample size, maturity, sex or a combination of biological and ecological conditions [4]. Additionally, sea cucumber harvested during the 2020 fishing season are expected to have the highest percentage of water content which may be linked to the greater proportion of vacuum transfer unloading sites compared to bucket unloading sites in that year. Therefore, the authors recommend the collection and analysis of additional data to confirm the cyclicity of water content.

The effect of location within the fishing vessel's hold on water loss is presented in Figure 15. Based on model predictions from the data collected, sea cucumbers located in the top of the hold are expected to contain the highest percentage of water, while those located in the bottom are expected to contain the lowest. This makes sense based on the authors' assumptions that sea cucumber stored near the top of the hold are harvested last and are, therefore, livelier.

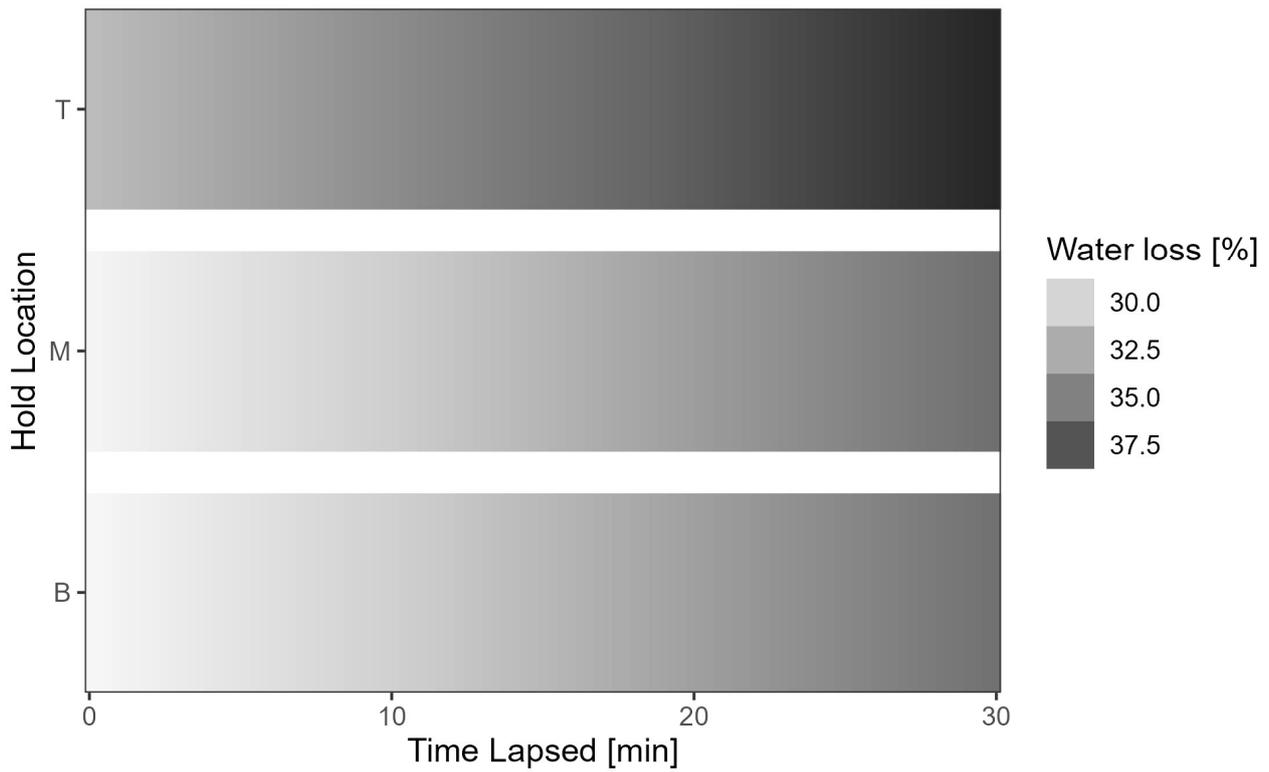
The effect of the unloading method on water loss is presented in Figure 16. Based on model predictions from the data collected, sea cucumber transferred using vacuum transfer (TV) are expected to contain higher water content than sea cucumber unloaded manually using a bucket (B). This observation is consistent with the fact that the vessel's hold is flooded with fresh seawater just prior to offloading, which is subsequently pumped out along with the product.



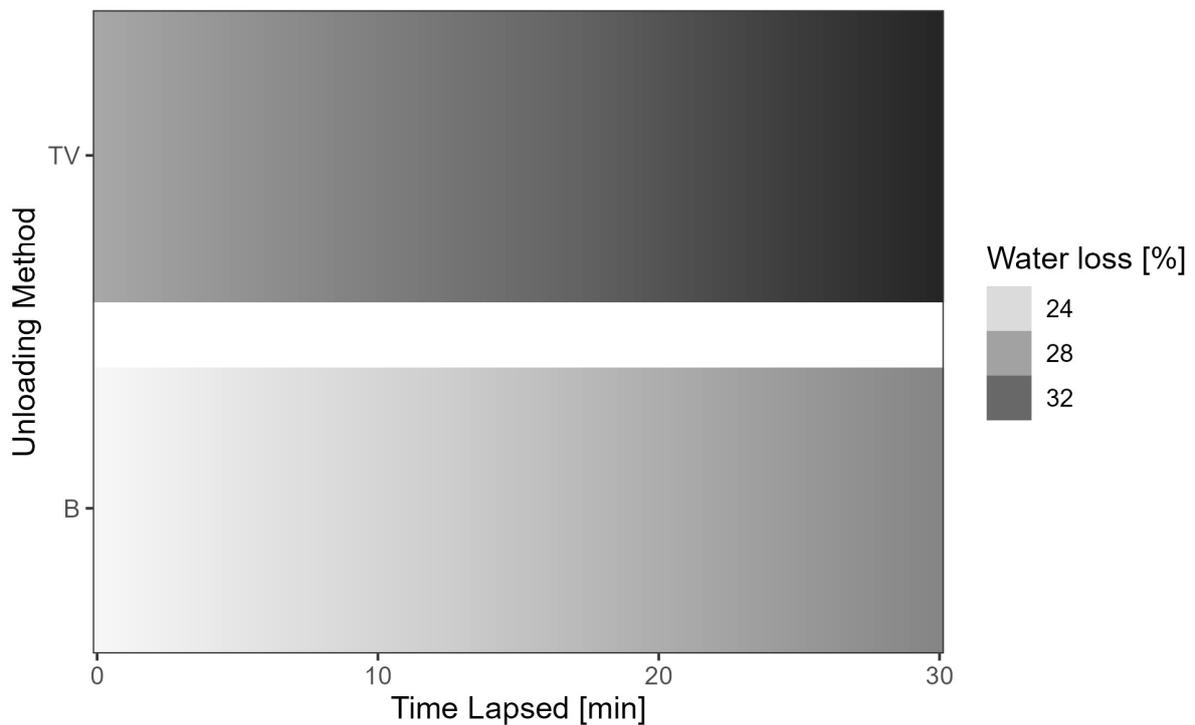
**Figure 13.** Expected water loss as a percentage of wet weight is plotted versus lapsed drain time for each month during the fishing season. Figure produced using Visreg v2.7.0 [32].



**Figure 14.** Expected water loss as a percentage of wet weight is plotted versus lapsed drain time for 2019 and 2020. Figure produced using Visreg v2.7.0 [32].



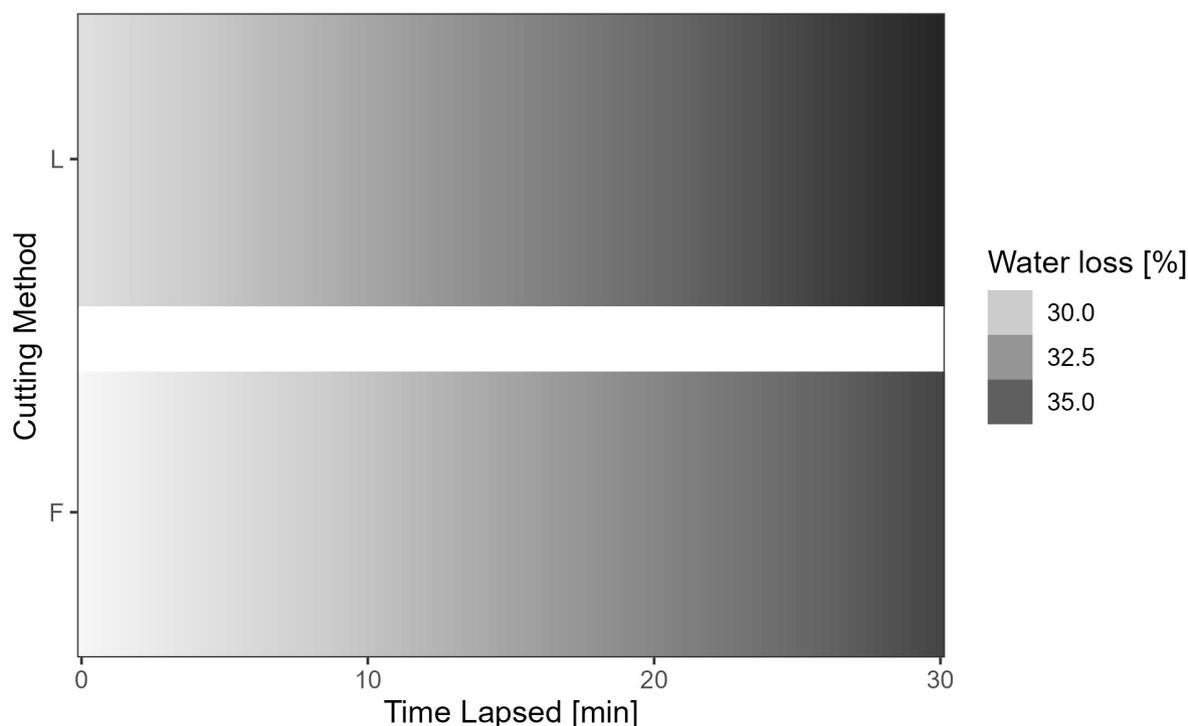
**Figure 15.** Expected water loss as a percentage of wet weight is plotted versus lapsed drain time for each location within the fishing vessel’s hold. Locations include top (T), middle (M) and bottom (B) of the hold. Figure produced using Visreg v2.7.0 [32].



**Figure 16.** Expected water loss as a percentage of wet weight is plotted versus lapsed drain time when using vacuum transfer (TV) or bucket (B) unloading. Figure produced using Visreg v2.7.0 [32].

The effect of the cutting method on water loss is presented in Figure 17. Based on model predictions from the data collected, sea cucumbers cut longitudinally (L) are expected to exhibit a higher percentage of water loss than those with only the flower (F)

removed. The authors observed that longitudinally cut sea cucumber consistently lost more water than flower cut samples as drain time increased. This trend is expected as longitudinally cut sea cucumber are fully cut open and present a larger exposed surface area allowing more water to be released.



**Figure 17.** Expected water loss as a percentage of wet weight is plotted versus lapsed drain time for flower cut (F) and longitudinally cut (L) sea cucumber. Figure produced using Visreg v2.7.0 [32].

Finally, the model was evaluated to determine the predicted average water loss for a landing as a percentage of the landed weight. Using the percentage size breakdown values presented in Figure 4 (i.e., 14.1% small, 47.0% medium and 38.9% large) and the mean percentage water loss values at each time lapsed reading (i.e.,  $t = 0, 10, 20,$  and  $30$  min) presented in Figure 12, the model predicts 31.2% water loss at  $t = 0$ , 34.1% at  $t = 10$ , 36.4% at  $t = 20$  and 39.2% at  $t = 30$  min. Therefore, for a typical landing of Atlantic sea cucumbers, one would expect more than 23% water loss.

## 5. Conclusions

This study evaluated the water-holding capacity of Atlantic sea cucumbers harvested off the south coast of Newfoundland and Labrador, Canada to assess the adequacy of the historic 23% water weight allowance established by the DFO. Based on two seasons of sampling, the findings revealed that the actual water-holding capacity exceeded the 23% deduction. Consequently, following this research and subsequent follow up, DFO increased the water-holding allowance to 34% [33].

**Supplementary Materials:** The following supporting information can be downloaded at <https://www.mdpi.com/article/10.3390/fishes10050212/s1>, DataSheet.xlsx: Data Collection Sheet and Water.csv: Water Loss Data.

**Author Contributions:** Conceptualization, P.B., W.M., J.C.G., D.D.; methodology, P.B., W.M., J.C.G., D.D.; validation, P.B., H.J.B.; formal analysis, P.B., W.M.; investigation, P.B., H.J.B., W.M.; resources, H.J.B., D.D.; data curation, P.B.; writing—original draft preparation, P.B.; writing—review and editing, P.B., W.M., H.J.B., J.C.G., D.D.; visualization, P.B.; supervision, H.J.B., D.D.; project administration,

H.J.B.; funding acquisition, P.B., H.J.B., J.C.G., W.M., D.D. All authors have read and agreed to the published version of the manuscript.

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**Institutional Review Board Statement:** Research performed in this study used sea cucumbers that were already being processed during typical commercial fishing operations, therefore no animal care protocol was required according to Canadian regulations [34]. All processing took place at registered processing facilities.

**Informed Consent Statement:** Not applicable.

**Data Availability Statement:** Data are contained within the article and Supplementary Materials.

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## Abbreviations

The following abbreviations are used in this manuscript:

BIC	Bayesian information criteria
DFO	Fisheries and Oceans Canada
EEZ	Exclusive economic zone
GADM	Global administrative areas
NAFO	North Atlantic Fisheries Organization
SPM	St. Pierre and Miquelon

## Appendix A

### Appendix A.1. Equipment

Equipment used for sample and free-water data collection included the following:

1. Western Scale digital indicator (#M2000, Western Scale Co., Ltd., Port Coquitlam, BC, Canada) with a TSS 16'' × 16'' stainless steel base (#TSS1616, The Scale Shop, St. John's, NL, Canada).
  - a. This scale has a 25 kg capacity and a 0.005 kg resolution.
  - b. This scale's calibration was certified in January 2019 and January 2020.
  - c. The calibration was checked periodically throughout each sampling season.

2. Ohaus Defender 5000 scale with digital indicator (#D52XW25WQR5, Ohaus Corp., Parsippany, NJ, USA).
  - a. This scale has a 25 kg capacity with a 0.001 kg resolution.
  - b. The scale's calibration was certified in December 2019 and July 2020.
  - c. The calibration was checked periodically throughout the 2020 sampling season.
3. Transcell digital indicator and 16" × 16" stainless steel base (#TI-500E SS, Transcell Technologies Inc., Buffalo Grove, IL, USA).
  - a. This scale has a 50 kg capacity and a 0.01 kg resolution.
  - b. This scale was certified in January 2019 and July 2020.
  - c. The calibration was checked periodically throughout each sampling season.
4. Seventy-liter StackNest closed pans (#60258 70L, IPL Inc., St-Damien de Buckland, QC, Canada).
5. Fourteen-liter Rubbermaid commercial bus pans (#FG334992GRAY, Newell Brands Co., Atlanta, GA, USA).
6. Vented drying pans (#11020, Sommer-Allibert S.A., La Défense, France).
7. Lump roe drain screens (#43186, C&W Industrial Fabrication & Marine Equipment, Bay Bulls, NL, Canada).
8. Insulated totes with capacities of 1000 and 2000 L (various brands, processor supplied).
9. Additional items included filleting knives, and Teflon tags.

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