

**Abstract.**— A laboratory study showed that when small male snow crabs are tagged with the anchor of a t-bar tag inserted into the dorsal musculature, tag retention and survival through the first molt are excellent; when the tag anchor is situated in the basal leg musculature, animals die while molting. When recaptures were obtained from larger, field-tagged animals, it was noted that some animals which had molted had been tagged in the leg musculature. Dissections were performed on 43 animals which molted and 89 animals which did not molt to determine tag anchor placement. Four general locations were noted: dorsal musculature, basal leg musculature, loose in the body cavity, and attached internally to carapace. Relative tag retention/survival associated with molting was estimated for the different tag anchor locations by comparing the proportions of tags in each location among animals which molted and among those which did not. Animals with anchors in the leg musculature appeared to survive and retain the tag through a molt as well as those tagged in the dorsal musculature and those with the tag attached to the inside of the carapace. Animals with anchors loose in the body cavity appeared to have worse tag retention/survival than those tagged in the dorsal musculature. The hypothesis that tag placement does not affect retention/survival through molt was tested by fitting hierarchical loglinear models and testing for a significant interaction between molt status (i.e., did or did not molt) and tag anchor location. No statistically significant effect was found, but it still seems prudent to try to place tag anchors into the dorsal musculature.

# Effect of Tag Anchor Location on Retention/Survival through Molt in Male Snow Crabs *Chionoecetes opilio*

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A tagging study was initiated in Conception Bay, Newfoundland, Canada, to determine the growth in size of male snow crabs *Chionoecetes opilio* at the time of molting. The t-bar tag (Floy Tag Mfg. Co., Inc., Seattle, WA 98105) was selected because preliminary studies had shown that the tag can be retained through a molt (McBride 1982, Taylor 1982). Early tag return rates were lower than expected, so a study was initiated to evaluate the performance of the tag when applied to animals held in captivity (Hurley et al. 1990). The laboratory study, conducted on animals ranging in size from 60 to 83 mm CW (carapace width), demonstrated that survival and retention of tags through a molt was excellent, provided the tag anchor was inserted into the dorsal musculature (Fig. 1). However, when the tag anchor was inserted into the basal leg musculature, the animals died during the molt. Tissue necrosis associated with *Pseudomonas* sp. bacteria was frequently noted around the tag anchor regardless of where it was located in the body.

For the Conception Bay study, an effort was made to carefully insert the tag anchors into the dorsal musculature. It was therefore surprising when some animals that had molted were recaptured with the anchor in the basal leg musculature. Also, some crabs had tags with the anchor loose in the body cavity, while in others it was attached to the carapace. Since the animals examined in the field

study were larger (82–120 mm CW at tagging) than the ones used in the laboratory study, it was hypothesized that placement of the tag anchor may not be as critical for large animals as for smaller ones.

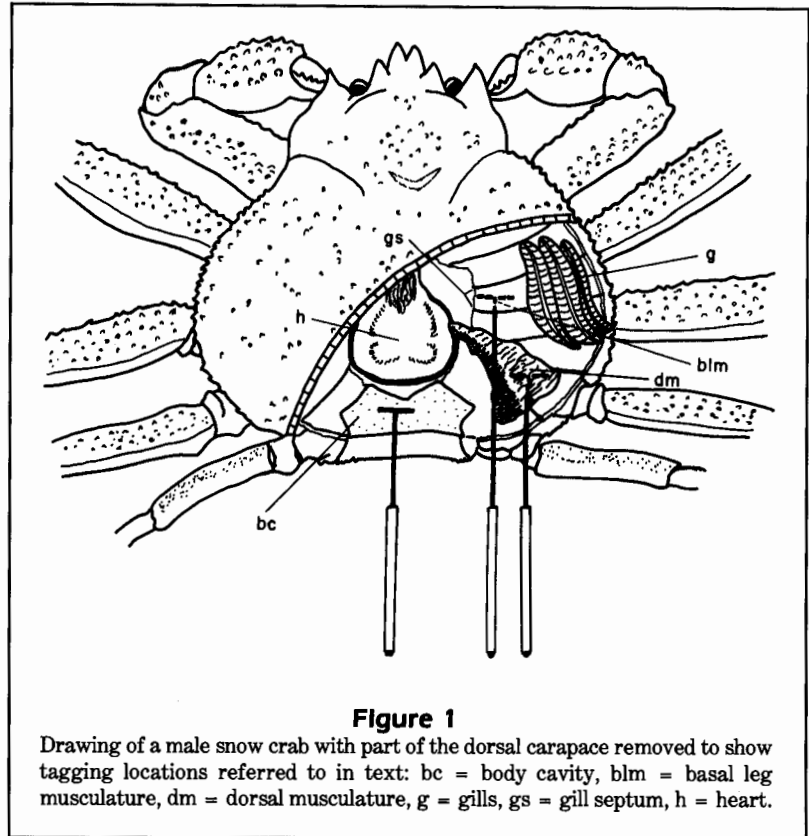
In this paper, we present the results of dissecting 132 recaptured animals to determine location of the tag anchor within the body. We develop analytical procedures to estimate the relative rate of tag retention and survival at the time of molting for different anchor locations. We test the hypothesis that retention/survival is the same for all locations using hierarchical loglinear models.

## Materials and methods

Male snow crabs were tagged in Conception Bay during 1983 and 1984 using methods described by Hurley et al. (1990) and Taylor and Hoenig (1990). The t-bar tag consists of a vinyl anchor 8 mm long, 1.2 mm in diameter, attached perpendicularly to a 25 mm-long shaft, 0.5 mm in diameter, which in turn connects to a 50-mm length of Number 20 vinyl tubing printed with identification information. Tags were inserted through the posterior ecdysial suture (epimeral line) which was made visible by applying gentle upward pressure to the carapace. The location of tag insertion was on the right side of the body 2–6 mm from the coxopodite of the last walking leg (Fig. 1). Before releasing the crab, the end of the tag

was given a gentle tug. If the tag appeared loose, it was removed and the animal was discarded.

Recaptured animals were obtained from commercial fishermen and stored in a freezer. Animals were then thawed and dissected in order to determine the location within the animals of the tag anchor. The carapace was cut diagonally on either side of the protruding tag by inserting the lower blade of a pair of scissors into the epimeral line. The forward portion of the triangular piece of cut carapace was then lifted to uncover the end of the tag. Tags were classified as being in one of four positions (Table 1): (1) anchor lodged in the dorsal musculature; (2) anchor loose in the body cavity, e.g. nestled against the hepatopancreas; (3) anchor lodged in the soft, newly forming carapace under the hard carapace, or attached to the underside of the hard carapace; and (4) anchor lodged in the basal leg musculature. The extent of any necrotic tissue around the anchor was also noted (Table 2).



**Figure 1**

Drawing of a male snow crab with part of the dorsal carapace removed to show tagging locations referred to in text: bc = body cavity, blm = basal leg musculature, dm = dorsal musculature, g = gills, gs = gill septum, h = heart.

## Analysis of recapture data

The logic of our analysis is as follows. We don't know the proportion of animals with the tag anchor placed in each of the four locations, and we don't know the magnitude of any initial tag loss or mortality immediately (within one month) following release of the tagged animals. However, those recaptured animals at liberty for more than one month which did not molt provide an estimate of the proportions of animals with tag anchors in each location prior to molting. Similarly, the recaptured animals which did molt provide an estimate of the proportions after molting. Consequently, differences in the proportions reflect different tag retention and/or survival.

This analysis requires two assumptions: (1) tags in all of the tagging locations affect the timing of molting in the same way (if at all); (2) differential mortality and tag loss among tagging locations is zero both prior to and subsequent to molting (with the possible exception of the period immediately after tagging and release of animals). A number of studies have shown that molting can be a critical period for tag-induced mortality and tag shedding (Restrepo and Hoenig 1988, Hurley

**Table 1**

Recapture data for male *Chionoecetes opilio* tagged in Conception Bay, Newfoundland, given by year of tagging, molt status (molted vs. did not molt), location of tag anchor within the body, and data pooled over the 2-year period.

Recapture of animals tagged in:	Molt status	Tag locations			
		dorsal musculature	loose in body	in shell	leg musculature
1983	did not molt	7	2	8	7
	molted	4	1	4	3
1984	did not molt	29	14	11	11
	molted	16	1	6	8
Years combined	did not molt	36	16	19	18
	molted	20	2	10	11

et al. 1990). Hence, it is reasonable to presume that if animals survive the first month at liberty with their tag intact, then they are likely to remain viable until the time of molting.

Consider the recapture data for 1984 in Table 1. The relative abundance of animals with tag anchors loose in the body cavity and in the dorsal musculature, among animals which did not molt, is 14:29 or 0.48:1. Among animals which molted, the relative abundance is 1:16

**Table 2**

Incidence of necrosis around the tag anchor in male *Chionoecetes opilio* tagged in Conception Bay, Newfoundland, with t-bar tags.

	Tagging location			
	Dorsal muscle	Body cavity	Shell	Leg muscle
Number with necrosis	11	4	7	7
Number without necrosis	45	14	22	25
% necrotic	20	22	24	22

or 0.06:1. Hence animals with anchors loose in the body cavity appear to have become less abundant relative to those tagged in the dorsal musculature, and this suggests lower tag retention and/or lower survival for animals with anchors loose in the body cavity. In fact, the relative retention/survival rate can be estimated from the 1984 data as

$\alpha$  = relative retention/survival (loose:dorsal)

$$\alpha = \frac{1}{\frac{16}{14}} = \frac{1 \cdot 29}{16 \cdot 14} = 0.13.$$

The estimator  $\alpha$  is the cross-product ratio frequently used in survival analysis (Fienberg 1980).

If all data were collected from animals tagged in the same year, then it would be a simple matter to use a  $\chi^2$  test to test the null hypothesis that the proportions in each location are the same for the two molt states (molted versus did not molt). However, animals were tagged in two separate years and there is at least a reasonably strong possibility that the tagging procedure varied between the years, e.g., due to developing tagging skill. This suggests that the interaction terms involving year may be significant. If interaction terms are ignored and the data from different years are pooled, then associations between variables (i.e., between molt status and tagging location) can be distorted and the apparent direction of the association can even reverse (see Fienberg 1980, chap. 3). An analysis which explicitly accounts for this possibility can be conducted using hierarchical loglinear models.

The 16 counts in Table 1 for individual years 1983 and 1984 can be envisioned as comprising a  $2 \times 2 \times 4$  contingency table with main effects (categorizations) due to year of tagging (Yr), molt status (Molt), and location of tag within the body (Loc). There are nine log-linear models that can be fitted to these data (Table 3) ranging from the model of complete independence through seven models of partial dependence to the completely saturated model (see Fienberg 1980 for a discussion). The contingency table can be collapsed over the year variable if one (or both) of the two-factor interactions involving year is not significant. The hypothesis that tagging location has no influence on retention/survival through molt can be rejected if it is necessary to have an interaction between location and molt status (Loc \* Molt) to have a good fit to the data.

Even the simplest model of main effects only (model 1) is not rejected by the likelihood ratio test ( $P = 0.2067$ , Table 3). It is still of interest to explore how strong the evidence may be that location of the tag affects survival/retention through the molt. We will therefore examine the following four models in more detail.

model 1:

$$\text{Count} = [\text{Molt}] [\text{Yr}] [\text{Loc}]$$

model 3:

$$\text{Count} = [\text{Molt}] [\text{Yr}] [\text{Loc}] [\text{Yr}] * [\text{Loc}]$$

model 4:

$$\text{Count} = [\text{Molt}] [\text{Yr}] [\text{Loc}] [\text{Molt}] * [\text{Loc}]$$

model 5:

$$\text{Count} = [\text{Molt}] [\text{Yr}] [\text{Loc}] [\text{Molt}] * [\text{Loc}] [\text{Yr}] * [\text{Loc}]$$

**Table 3**

Loglinear model analysis of tagging count data in Table 1. C is the predicted count, [Yr] represents year of tagging, [Molt] represents molt status, and [Loc] represents location of the tag anchor within the body. Asterisk (\*) indicates an interaction between variables. To make the notation more compact, only the highest-order interactions are given for each variable. For example, the model  $C = [\text{Loc}] [\text{Yr}] * [\text{Molt}]$  represents main effects for location, year, and molt, plus the interaction between year and molt. The order of presentation of the variables has no significance in the model notation.

Model	Likelihood ( $\chi^2$ )	df	P
1) $C = [\text{Yr}] [\text{Molt}] [\text{Loc}]$	13.31	10	0.2067
2) $C = [\text{Yr}] * [\text{Molt}] [\text{Loc}]$	13.30	9	0.1495
3) $C = [\text{Molt}] [\text{Yr}] * [\text{Loc}]$	7.03	7	0.4262
4) $C = [\text{Yr}] [\text{Molt}] * [\text{Loc}]$	8.11	7	0.3233
5) $C = [\text{Yr}] * [\text{Loc}] [\text{Molt}] * [\text{Loc}]$	1.82	4	0.7688
6) $C = [\text{Yr}] * [\text{Molt}] [\text{Loc}] * [\text{Molt}]$	9.09	6	0.2313
7) $C = [\text{Molt}] * [\text{Yr}] [\text{Loc}] * [\text{Yr}]$	7.01	6	0.3196
8) $C = [\text{Molt}] * [\text{Yr}] [\text{Loc}] * [\text{Yr}] [\text{Molt}] * [\text{Loc}]$	1.81	3	0.6120
9) $C = [\text{Yr}] * [\text{Molt}] * [\text{Loc}]$	0	0	1.0000

The above model descriptions are a common way of representing loglinear models. Model 1 indicates that the logarithm of the count in any cell can be predicted as the sum of the main effects for molt status, year of tagging, and location, i.e., these factors act independently. Model 3 is the same as model 1 except that an interaction term (or dependency) between year of tagging and location of anchor is also needed to predict the counts. Model 4 has a similar interpretation but the logarithm of the count depends on an interaction between anchor location and molt status. Model 5 includes the interaction between year and location and between molt status and location. Thus, models 1 and 3 imply no differential survival/tag retention among tagging locations, whereas models 4 and 5 imply that tagging location has an effect on the probability of surviving a molt with the tag still in place.

The difference between models 1 and 3 is that model 3 implies that the proportions of animals tagged in each location varied between the two years while model 1 implies the same proportions occurred in both years. If year has no effect on location (i.e., the interaction between year and location is unimportant), then a test of the effect of location on survival/retention through molt can be achieved by comparing models 1 and 4. But, if the proportions tagged in each location varied among the years, then the comparison that isolates the effect of tagging location on survival/retention through molt is the comparison of models 3 and 5.

The choice between two nested models can be made on the basis of a likelihood ratio test by subtracting the log-likelihoods and referring to a  $\chi^2$  table with degrees of freedom equal to the difference in degrees of freedom for the two models. The computed test statistic for comparing models 1 and 3 is

$$\chi^2_{\text{comp}} = 13.31 - 7.03 = 6.28, \quad \text{df} = 10 - 7 = 3$$

and the resulting *P*-value is 0.099. We fail to reject model 1 in favor of model 3 at the 5% level and conclude that the evidence is not strong enough to conclude that the proportions tagged in each body location varied by year. However, the test results could be considered marginally or nearly significant.

The hypothesis of interest is whether the tagging location affects the survival/retention through molt. This can be accomplished by comparing models 1 and 4,

$$\chi^2_{\text{comp}} = 13.31 - 8.11 = 5.20, \quad \text{df} = 10 - 7 = 3,$$

for which the *P*-value is 0.1577. Alternatively, we can compare models 3 and 5 and obtain virtually the same results. Thus, the statistical evidence is not strong enough to conclude that location affects survival/retention through molt at the customary 5% level. However,

in light of the small sample sizes and possible low power of the test, the results provide some evidence, albeit weak, that location may be important.

Since at least one two-factor interaction involving year is not significant in each of the models we considered, one can validly collapse the three-dimensional table over year to obtain a  $2 \times 4$  table. The estimated relative retention/survival rates,  $\alpha$ , can be computed from the pooled data (Table 1). Relative to animals tagged in the dorsal musculature, the tag retention/survival of animals with tag anchors loose in the body cavity, embedded in shell, and in the dorsal leg musculature are estimated to be 0.23, 0.95, and 1.10, respectively.

Although 18% of the dissected animals had some blackening around the anchor of the tag, only 2 animals (1.5%) had extensive areas of necrosis. All animals had been at liberty for at least a year. The proportion of animals with necrotic tissue did not appear to vary much among the different tagging locations (Table 2).

## Discussion

Although the laboratory study indicated that tagged animals die at the time of molting if the tag anchor lodges in the basal leg musculature, there was no evidence of this in the field data. We hypothesize that this is because the field-tagged animals were larger than the laboratory animals, and larger animals may tolerate a tag in the leg musculature better than smaller animals. If relative retention/survival due to anchor location depends on size of the animals, then size category should be considered as another variable in the analysis. However, in our field study only animals in a narrow range of sizes (82–120 mm CW at tagging) were examined so that there seems little point in dividing the limited number of recaptures into size classes.

Since the field data indicate that having the end of the tag loose in the body cavity reduces the chances of recovering the animal with tag intact ( $\alpha$  0.23), and the laboratory data indicate that tagging in the basal leg musculature reduces recoveries, it seems prudent to try to insert the tag anchor into the dorsal musculature. Our lack of statistically significant results does not imply that tagging location is not a significant biological factor in determining the success of a tagging program. Rather, it may simply indicate that our sample sizes were inadequate to obtain strong evidence of the importance of location. We recommend that, prior to initiating a field tagging program, a few trial animals be sacrificed to determine how consistently the tag anchor is being placed into the targeted dorsal musculature.

The loglinear model approach we used is quite general and allows any number of covariates (such as size at the time of tagging) to be properly accounted for. For those who prefer a regression approach to data analysis, the same analyses can be cast as logit models (see Fienberg 1980, chap. 6).

The laboratory study by Hurley et al. (1990) suggested that tag insertion frequently results in extensive necrosis of the tissue and carapace around the anchor. This was not seen in the field data. We speculate that the decreased levels of necrosis in the field-tagged animals may be due to the lower water temperatures in Conception Bay ( $-1.2$  to  $-0.5^{\circ}\text{C}$  in the Bay versus  $4-6^{\circ}\text{C}$  in the laboratory study).

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