Use of endoparasitic helminths as tags in delineating stocks of American plaice (*Hippoglossoides platessoides*) from the southern Gulf of St. Lawrence and Cape Breton Shelf

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Abstract—Endoparasitic helminths were inventoried in 483 American plaice (*Hippoglossoides platessoides*) collected from the southern Gulf of St. Lawrence, NAFO (North Atlantic Fisheries Organization) division 4T, and Cape Breton Shelf (NAFO subdivision 4Vn) in September 2004 and May 2003, respectively. Forward stepwise discriminant function analysis (DFA) of the 4T samples indicated that abundances of the acanthocephalans *Echinorhynchus gadi* and *Corynosoma strumosum* were significant in the classification of plaice to western or eastern 4T. Cross validation yielded a correct classification rate of 79% overall, thereby supporting the findings of earlier mark-recapture studies which have indicated that 4T plaice comprise two discrete stocks: a western and an eastern stock. Further analyses including 4Vn samples, however, indicated that endoparasitic helminths may have little value as tags in the classification of plaice overwintering in Laurentian Channel waters of the Cabot Strait and Cape Breton Shelf, where mixing of 4T and 4Vn fish may occur.

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American plaice (*Hippoglossoides platessoides*) are found in Northwest Atlantic waters along the continental shelf and upper continental slope from west Greenland to Rhode Island, favoring intermediate depths (90–250 m), cold waters (<0–1.5°C), and fine sand or mud bottom (Scott and Scott, 1988). Amercian plaice has ranked second in importance to Atlantic cod (*Gadus morhua*) among groundfish landed in the southern Gulf of St. Lawrence (North Atlantic Fisheries Organization [NAFO] division 4T) over the past four decades, but commercial landings, which had ranged from 4907 to 11,780 t between 1965 and 1992 (Morin et al.[1]), fell to 401 t by 2004 (Fisheries and Oceans Canada[2]). Commercial and research survey data show that declines in abundance of southern Gulf plaice since 1991 have occurred primarily in western 4T between the Gaspé Peninsula and the Magdalen Islands (see Fig. 1), and abundances east of the Magdalens have remained stable. The eastern and western 4T stocks monitored in recent assessments are consistent with the meristically indistinguishable "Miscou-Magdalen" and "Cape Breton" groups which Powles (1965) delineated through mark-recapture experiments. Powles (1965) suggested that the meristic uniformity of these two groups may be a consequence of larval drift because mature fish from the respective groups seldom mix on their summer feeding grounds. More recently, Stott et al. (1992) found no evidence of population subdivision of Southern Gulf plaice in respect to allozyme variation and restriction fragment length polymorphisms in mitochondrial DNA.

As evident from commercial catch records and tag returns, 4T plaice, with the exception of some immature fish that remain in the shoals year round, migrate from summer feeding grounds on the Magdalen Shallows to deeper waters of the Laurentian Channel in winter (Powles, 1965). The winter distribution of plaice in the Laurentian Channel is continuous from the Gaspé to the Cabot Strait (Clay, 1991) and eastward along the Cape Breton Shelf (NAFO subdivision

[1] Morin, R., I. Forest, and G. Poirier. 2001. Status of NAFO Division 4T American plaice, February 2001. Canadian Science Advisory Secretariat Research Document. 2001/023, 70 p. Fisheries and Oceans Canada, Science Branch, Marine Fish Division, Gulf Region, P.O. Box 5030, Moncton, New Brunswick, Canada.

[2] Fisheries and Oceans Canada. 2005. American plaice in the southern Gulf of St. Lawrence (Division 4T). Fisheries and Oceans Canadian Science Advisory Secretariat Science. Advisory Report. 2005/008, 5 p. Maritime Provinces, Regional Advisory Process, Fisheries and Oceans Canada, P.O. Box 1006, Dartmouth, Nova Scotia, Canada.

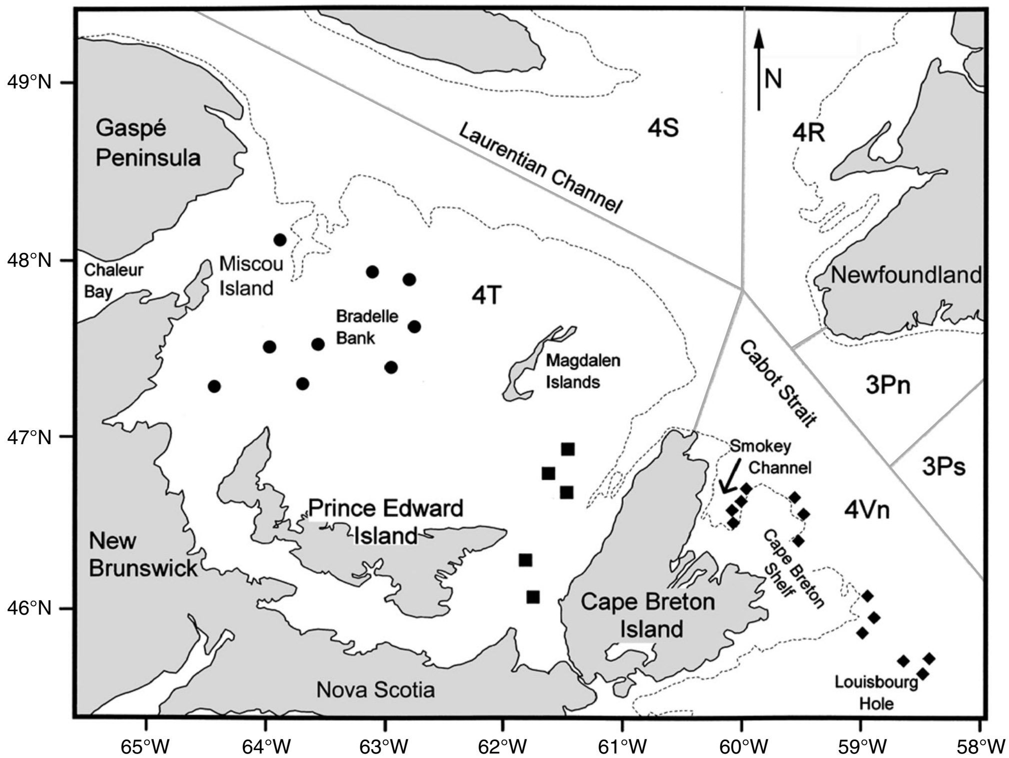

Figure 1

Map of eastern Canada indicating sites where American plaice (*Hippoglossoides platessoides*) were sampled in the southwestern (●) and southeastern (■) Gulf of St. Lawrence, and on or near the Cape Breton Shelf (♦). The depth contour (.....) is at 100 m. The pale gray lines demarcate North Atlantic Fisheries Organization divisions (4T, 4S. 4R) and subdivisions (3Pn, 3Ps, and 4Vn).

4Vn) (Swain et al., 1998). Clay (1991), noting the abundance of plaice in the Gulf portion of the Channel in January, surmised that southern Gulf plaice, unlike 4T cod, with which they are often closely associated, do not migrate through the Cabot Strait to over-winter in 4Vn. Length-at-age data from commercial landings indicate that plaice taken near the Cabot Strait late in the year are primarily from the slower growing eastern 4T (Cape Breton) stock (Tallman, 1991). A few older plaice tagged on Bradelle Bank, northwest of the Magdalen Islands, however, have been recovered in the Cabot Strait area in fall and winter (Powles, 1965). Clearly, tagging data and information on seasonal distributions from research surveys and commercial fisheries have provided little insight into the movements of 4T plaice stocks in the Laurentian Channel in winter and have not dispelled the possibility that 4T stocks may be exploited during the winter fishery within 4Vn.

Parasites as biological tags have often proven advantageous over more costly mark-recapture methods in studies of fish stock structure and migration (Williams et al., 1992) and are useful as markers in surveys for allocating quotas and combating illegal landings (Power et al., 2005). Parasite markers have shown potential in stock delineation of numerous demersal fish species (Marcogliese et al., 2003; MacKenzie and Abuanza, 2005; Melendy et al., 2005), including flatfish species such as Greenland halibut (*Reinhardtius hippoglossoides*) (Arthur and Albert, 1993; Boje et al., 1997), Pacific halibut (*Hippoglossus stenolepis*) (Blaylock et al., 2003), and winter flounder (*Pleuronectes americanus*) (McClelland et al., 2005). Although there have been no prior attempts to use parasite assemblages in describing the stock structure of American plaice, parasitological surveys of plaice in the Northwest Atlantic have identified a number of potential parasite tags. Scott (1975), for example, concluded that the enteric digeneans *Steringotrema ovacutum*, *Zoogonoides viviparous*, and *Fellodistomum furcigerum* might prove useful as tags for plaice stocks from the southern Gulf of St. Lawrence, Scotian Shelf, and northeastern Gulf of Maine. Zubchenko (1985) remarked that species compositions and infection parameters of 20 protozoan and metazoan parasites of plaice from the Grand Bank, Flemish Cap, and northeastern Newfoundland and Labrador are peculiar to their geographic origin. Finally, studies of the temporal and geographic distributions of larval sealworm (*Pseudoterranova decipiens*) (Nematoda) in Atlantic Canadian groundfish have revealed significant disparities in prevalence and abundance of sealworm in neighboring plaice stocks (McClelland et al., 2000; McClelland and Martell, 2001b).

The primary objective of the present study was to investigate the possibility of using parasite markers to delineate the western and eastern 4T plaice stocks, evident in earlier mark-recapture experiments (Powles, 1965). In light of potential mixing of southern Gulf and Cape Breton Shelf plaice over-wintering in the Laurentian Channel, parasite tags were also employed in an effort to examine the discreteness of 4T and 4Vn stocks.

Materials and methods

Sampling of the host

American plaice, 31 to 40 cm in total length, were collected from two locations in the southern Gulf of St. Lawrence (NAFO division 4T) and from three locations on or near the Cape Breton Shelf (NAFO subdivision 4Vn) (Fig. 1). Samples from the southern Gulf were collected from the Canadian Coast Guard Ship (CCGS) *Alfred Needler* and the CCGS *Teleost* during a Fisheries and Oceans Canada (DFO) demersal fish survey in September of 2004, and plaice from the Cape Breton Shelf were sampled from the *Alfred Needler* in May 2003 during a dedicated survey of parasites and diseases in 4Vn groundfish. Fish were caught with a Western IIA otter trawl which was towed for 30 minutes at each station. A total of 483 plaice were collected, including 137 plaice taken at depths of 43 to 88 m in the western 4T division and 95 plaice sampled at depths of 49 to 70 m in eastern 4T division. In the 4Vn survey, 128 plaice were sampled from the eastern slope of the Smokey Channel, 46 along the eastern edge of the Cape Breton Shelf, and 77 in the Louisbourg Hole. “Channel,” “Edge,” and “Hole” samples were taken at depths ranging from 77 to 124 m, 72 to 158 m, and 77 to 135 m, respectively. Samples were frozen onboard in a walk-in freezer at –17°C and transferred on landing to a –20° C walk-in freezer at the Gulf Fisheries Centre (GFC), Moncton, New Brunswick, where they were stored for future examination.

Individual fish were thawed at room temperature and measured lengthwise to the nearest centimeter. External surfaces and gills were inspected by eye for signs of trauma and other disease conditions. Viscera were examined for endoparasitic helminths with a dissecting microscope, and fillets were removed and sliced into thin sections (McClelland and Martell, 2001a) for detection of larval digeneans and nematodes. All helminth parasites were counted with the exception of *Stephanostomum baccatum* metecercariae. Because the latter were often too numerous and too widely distributed in host tissues to be counted within a reasonable time frame, only their presence was noted.

Statistical analysis

Prevalence (P) and abundance (A) of individual parasite species were calculated according to the methods of Bush et al. (1997), with prevalence being the numbers of infected fish in a sample divided by the total number of fish in a sample, expressed as a percentage, and abundance being the total number of parasites recovered from a sample divided by the total number of fish in a sample. Individual fish were coded 1 (infected), or 0 (uninfected) (Li, 1964; Neter et al., 1985) for analyses of prevalence, and a log (n+1) transformation was used to bring the distributions of parasite counts closer to normality (Sokal and Rohlf, 1969; Platt, 1975). Parasite infection parameters that best distinguished between sampling locations were selected by forward stepwise discriminant function analysis (DFA). The Kappa statistic (K) was used to determine the improvement over chance of DFA (Titus et al., 1984). Misclassification rates of DFAs were calculated by employing cross validation procedures described by Arthur and Albert (1993) and Boje et al. (1997). All statistical procedures were performed with Systat for windows, vers. 7.0 (SPSS Inc., Chicago, IL). Because the survey was confined to plaice in a narrow length range (31–40 cm TL) (McClelland et al., 2000), effects of host size on parasite infection parameters were not investigated. Plaice from the edge of the Cape Breton Shelf were not included in the DFA analysis because of the small sample size (n=46) and the fact that they were collected from two widely separated groups of trawl sets (Fig. 1); the northern (n=21) and southern (n=25) portions of the sample were more distant from each other than were the samples from Smokey Channel and Louisbourg Hole.

Results

Plaice sampled from the southern Gulf of St. Lawrence (NAFO Division 4T) and the Cape Breton Shelf and vicinity (NAFO subdivision 4Vn) ranged from 31 to 40 cm in total length (TL), but the great majority fell within the lower half of this range. Mean length of fish (±standard error) was 32.34 ±0.180 cm (n=137) for plaice from the western 4T sample, and 32.31 ±0.22 cm (n=95) for the eastern 4T sample. Mean lengths of 4Vn plaice were 33.17 ±0.26 cm (n=128) and 32.88 ±0.23 cm (n=77) for Smokey Channel and Louisbourg Hole fish, respectively.

Eleven species of endoparasitic helminths were identified during routine parasitological examinations of 483 plaice (Table 1), but the larval digeneans *Otodistomum* sp., plerocercoid larvae of an unknown cestode species, and the nematode *Hysterothylacium aduncum* were observed too infrequently (<4% prevalence in all samples) to be considered useful as biological tags (see MacKenzie and Abaunza, 1998). Forward stepwise DFA of infection parameters of the remaining species revealed that abundances of the acanthocephalans *Corynosoma strumosum* and *Echinorhynchus gadi* contributed significantly to the classification of plaice from the southwestern and southeastern Gulf of St. Lawrence (K=0.57) and that *E. gadi* abundance was the more significant variable (Table 2). Cross-validation yielded an overall rate of 79% correct classification (Table 3), and the lowest rate of misclassification (15%) was found in fish from the western 4T division. Abundances of *C. strumosum* (F=9.58), the digenean *Fellodistomum* sp. (F=11.61), and the larval anisakine nematode *Pseudoterranova decipiens* (F=8.89), and prevalence of the larval digenean *Stephanostomum baccatum* (F=8.64) were significant factors in the classification of 4Vn plaice from Smokey Channel and Louisbourg Hole (K=0.40), and cross-validation resulted in an overall classification efficiency of 70% (Table 4).

Stepwise DFA of helminth infections in plaice from all four sampling locations indicated abundances of *E. gadi* (F=35.80), *C. strumosum* (F=13.89), *Fellodistomum* sp., (F=5.81), and *P. decipiens* (F=4.75) and the prevalence of *S. baccatum* (F=5.71) were significant markers (K=0.30) for the classification of plaice from the entire (4T–4Vn) survey area. The overall rate of correct classification, however, was only 48%, and 51% of the plaice from western 4T divison were misclassified as 4Vn fish (Table 5). In a final analysis of samples from the east and west coasts of Cape Breton Island (eastern 4T and Smokey Channel, respectively), *E. gadi* abundance was again the most important marker selected (F=59.10), although abundances of the larval anisakine nematodes *Anisakis simplex* (F=6.50) and *Contracaecum osculatum* (F=6.60) also contributed significantly to classification (K=0.49). Cross-validation yielded a 77% rate of overall correct classification, but, although nearly all Smokey Channel plaice were classified correctly, more than half of the plaice from eastern 4T were misclassified as Smokey Channel fish (Table 6).

Table 1

Prevalence (P) (%) and abundance (A) of parasitic helminths in American plaice (*Hippoglossoides platessoides*) sampled from the southwestern (Western 4T) and southeastern (Eastern 4T) Gulf of St. Lawrence, Smokey Channel, edge of the Cape Breton Shelf, and Louisbourg Hole in 4Vn. Parasite. n/a=not available.

	Location (n)									
	Western 4T (n=137)		Eastern 4T (n=95)		Smokey Channel (n=128)		Edge of shelf (n=46)		Louisbourg Hole (n=77)	
Species	P	A	P	A	P	A	P	A	P	A
Digenea										
Hemiuriformes										
Derogenes varicus	4	0.04 ±0.02	6	0.06 ±0.03	3	0.17 ±0.05	24	0.30 ±0.09	16	0.17 ±0.05
Otodistomum sp.[1]	0	0	0	0	1	0.01 ±0.01	2	0.02 ±0.02	0	0
Strigeiformes										
Fellodistomum sp.	29	0.89 ±0.17	26	1.3 ±0.38	45	2.6 ±0.51	19	1.3 ±0.93	17	0.46 ±0.19
Plagiorchiiformes										
Stephanostomum baccatum[1]	15	n/a	33	n/a	33	n/a	24	n/a	13	n/a
Cestoda										
Cestoidea										
Unidentified cestode [1]	3	0.03 ±0.01	1	0.02±0.02	3	0.11 ±0.09	0	0	4	0.04±0.02
Nematoda										
Ascaridida										
Anisakis simplex[1]	10	0.13 ±0.03	8	0.08 ±0.03	22	0.23 ±0.04	15	0.19 ±0.07	21	0.21±0.05
Hysterothylacium Aduncum	1	0.01 ±0.0007	4	0.04 ±0.02	2	0.02 ±0.01	0	0	3	0.04±0.03
Contracaecum osculatum[1]	26	0.64 ±0.18	15	0.31 ±0.1	39	0.74 ±0.17	9	0.11 ±0.06	34	0.66±0.17
Pseudoterranova decipiens[1]	73	1.5 ±0.12	67	1.34 ±0.13	68	1.77 ±0.18	60	1.4 ±0.32	50	0.97±0.15
Acanthocephala										
Palaeacanthocephala										
Echinorhynchus gadi	7	0.1 ±0.031	50	1.3 ±0.3	4	0.09 ±0.05	0	0	13	0.27±0.1
Corynosoma strumosum[1]	8	0.15 ±0.06	30	0.5 ±0.15	25	0.48 ±0.1	33	1.6 ±0.6	38	2.1±0.9

[1] Larval.

Discussion

Forward stepwise DFA of parasite tags has been successfully employed in describing the stock structure of various flatfish species. Arthur and Albert (1993) used five species of larval helminth to classify widely separated stocks of Greenland halibut from the Saguenay fiord, St. Lawrence estuary, Labrador coast, and Baffin Island, amd Boje et al. (1997) used six helminth species to gain insight into the stock structure of Greenland halibut from northeastern Newfoundland and Greenland. The overall correct classification rate was >99% in the former, and 77% in the latter study. Blaylock et al. (2003) used eight helminth species to describe the stock structure of Pacific halibut along the Pacific coast of North America from the Bering Sea to northern California and achieved an overall correct classification rate of 83%. In all three studies, accuracy of classification increased when the numbers of geographical categories were reduced by pooling sampling locations.

Table 2

Results of forward stepwise discriminant function analysis of samples of American plaice (*Hippoglossoides platessoides*) from the southern Gulf of St. Lawrence (North Atlantic Fisheries Organization Division 4T) showing the relative importance of abundances of two species of acanthocephalan in the classification of individual fish to western and eastern 4T stocks.

Variable	Standardized coefficient of first canonical variable	df	*F* score	Eigenvalue
Echinorhynchus gadi	0.862	230	71.48	0.46
Corynosoma strumosum	0.499	229	20.84	

Table 3

Cross-validation of American plaice (*Hippoglossoides platessoides*) from the western and eastern North Atlantic Fisheries Organization division 4T (southern Gulf of St. Lawrence) determined by discriminant function analysis of abundances of two species of acanthocephalans.

Location	Western 4T	Eastern 4T	% Correct
Western 4T	117	20	85
Eastern 4T	28	67	71
Totals	145	87	79

Table 4

Cross-validation of American plaice (*Hippoglossoides platessoides*) from the Smokey Channel and Louisbourg Hole, North Atlantic Fisheries Organization subdivision 4Vn, as revealed by discriminant function analyses of the prevalence or abundance of four helminth species (prevalence of *Stephanostomum baccatum*, and abundances of *Fellodistomum* sp., *Corynosoma strumosum,* and *Pseudoterranova decipiens*).

Location	Smokey Channel	Louisbourg Hole	% Correct
Smokey Channel	86	42	67
Louisbourg Hole	20	57	74
Totals	105	99	70

Although our survey was confined to a much smaller geographic area than those surveyed in the studies above, the accuracy of classification of plaice stocks from the southern Gulf of St. Lawrence (NAFO division 4T) is remarkably high (79%) (Table 3). DFA with only two markers, namely the abundances of the acanthocephalans *E. gadi* and *C. strumosum*, indicated that southern Gulf plaice consist of discrete western and eastern stocks on their summer feeding grounds; this result supports the results of earlier mark-recapture studies (Powles, 1965). Because the acanthocephalans are acquired passively in prey, and 4T plaice feed primarily in the Magdalen Shoals, spring through fall, while fasting in winter (Swain et al., 1998), these markers may also be employed in studies of seasonal migrations and mixing of the two stocks. Analysis of infection parameters of four helminth taxa (abundances of *C. strumosum*, the digenean *Fellodistomum* sp., the larval anisakine nematode *Pseudoterranova decipiens*, and prevalence of the larval digenean *Stephanostomum baccatum*) reveals a somewhat lower degree of discreteness (70% correct classification) between plaice from Smokey Channel and Louisbourg Hole, within NAFO subdivision 4Vn .

Abundances of *Fellodistomum* sp., *P. decipiens*, *E. gadi,* and *C. strumosum*, and prevalence of *S. baccatum* are significant factors in a DFA of plaice from all four locations, although the overall accuracy of classification falls to only 48% (Table 5). Although an improvement over random classification (25%), this latter result offers little encouragement for the use of parasites tags in future investigations of movements and possible mixing of 4T and 4Vn plaice overwintering in Laurentian Channel waters of the 4Vn subdivisioin. DFA of eastern 4T and Smokey Channel (4Vn) samples similarly seems to indicate that helminth tags would have little value in studies of the winter composition of plaice stocks in the Cabot Strait area. In this analysis, with the use of *E. gadi* and the larval nematodes *Anisakis simplex* and *Contracaecum osculatum* as markers, 98% of the 4Vn plaice were classified correctly, but 52% of the plaice from southeastern 4T were misclassified as 4Vn fish. However, given the differences in infection parameters of *E. gadi* in eastern 4T (prevalence [P]=50%, abundance [A]=1.30) and Smokey Channel samples (P=4%,

Table 5

Cross-validation of American plaice (*Hippoglossoides platessoides*) from the western and eastern North Atlantic Fisheries Organization (NAFO) division 4T and from Smokey Channel and Louisbourg Hole in NAFO subdivision 4Vn, as revealed by discriminant function analyses of the prevalence or abundance of five species of parasitic helminth (prevalence of *Stephanostomum baccatum*, and abundances of *Fellodistomum* sp., *Corynosoma strumosum*, *Echinorhynchus gadi*, and *Pseudoterranova decipiens*).

Location	Western 4T	Eastern 4T	Smokey Channel	Louisbourg Hole	% Correct
Western 4T	57	10	37	33	42
Eastern 4T	14	48	15	18	51
Smokey Channel	28	5	64	31	50
Louisbourg Hole	18	8	11	40	52
Totals	117	71	127	122	48

A=0.09) (Table 1), this acanthocephalan may yet prove to be a useful marker for the detection of southeastern 4T migrants among plaice overwintering in Cabot Strait and 4Vn.

Parasites that contributed to the DFAs above meet MacKenzie's (1987) criteria for biological tags. None of the helminths are known to reproduce directly on or in plaice. Each of the markers is abundant in fish from at least one of the sampling areas, and there are significant geographical variations in the infection parameters of each species within the survey area. The third criterion, that the parasite must be long-lived in the host, is clearly met by five species of larval helminth that contribute significantly to classification. Metacercariae of *S. baccatum*, the larval anisakines *A. simplex*, *C. osculatum*, and *P. decipiens*, and cystacanths of *C. strumosum*, found variously in the body cavity and musculature, are believed to survive indefinitely in the fish host and have been used as markers in other studies of flatfish stock structure (Arthur and Albert, 1993; Boje et al.,1997; Blaylock et al., 2003).

Although the life spans of enteric helminths (*Fellodistomum* sp. and *E. gadi*, herein) in marine fish remain largely unknown, it is possible that species infecting cold-water hosts may persist for several months or even years (Margolis and Boyce, 1969)—sufficient time to meet MacKenzie's (1987) criterion for tag longevity. *Echinorhynchus gadi* infecting a relict population of Atlantic cod in Lake Mogil'noye, Russia, for example, recruit in the fall and die off in late summer and early fall of the following year (Kulachkova and Timofeyeva, 1993). Similarly, *E. lageniformis* survive for about a year in the intestines of starry flounder (*Platichthys stellatus*) in the coastal waters of Oregon (Olson and Pratt, 1971). In any event, parasites need not be long lived in order to be suitable as markers, and there are numerous precedents for use of enteric helminths as markers in studies of host stock structure (Williams et al., 1992). Khan and Tuck (1995) identified *E. gadi* as an important indicator of the discreteness of Newfoundland cod stocks, and Power et al. (2005) employed abundances of enteric digeneans in classifying bogue (*Boops boops*) (Sparidae), a demersal species from Spanish fisheries.

Table 6

Cross-validation of American plaice (*Hippoglossoides platessoides*) to eastern North Atlantic Fisheries Organization (NAFO) division 4T and Smokey Channel (NAFO subdivision 4Vn) as revealed by discriminant function analyses of the abundances of three helminth species (abundances of *Echinorhynchus gadi*, *Anisakis simplex,* and *Contracaecum osculatum*).

Location	Eastern 4T	Smokey Channel	% Correct
Eastern 4T	46	49	48
Smokey Channel	3	125	98
Totals	49	174	77

Abundances of passively transmitted parasites are ultimately a function of host feeding behavior. Hence, geographical differences in intermediate-host abundance and frequency in plaice diets would manifest themselves as disparities in infection parameters of passively transmitted helminths in 4T and 4Vn plaice stocks. Digeneans, which mature in the digestive tract of marine fish, are usually acquired through the consumption of invertebrates (crustaceans, molluscs, polychaetes, and echinoderms, among other taxa) which harbor encysted metacercariae (Rohde, 2005). Brittle stars (Ophiuroidea), which are frequently exploited by both 4Tand 4Vn plaice (Powles, 1965; Minet, 1975), are host to the metacercariae of *Fellodistomum* sp. (Koie, 1980), an important influence in our DFAs involving 4Vn plaice. Larval anisakine nematodes (*A. simplex*, *C. osculatum*, and *P. decipiens*), which proved significant in stock delineation herein, are acquired through predation on the parasite's invertebrate hosts, usually crustaceans (Rohde, 2005). Although the life cycle of *A. simplex* is largely pelagic, the larvae may be transmitted to demersal fish, such as plaice, by diurnal vertical migrants (euphausiids, shrimp, etc.) which feed pelagically at night but are found near the seafloor by day (McClelland, 1990). Larval *P. decipiens* infections are acquired through consumption of various benthic invertebrates, including mysids, isopods, amphipods, decapods, and polychaetes (McClelland, 2002) The acanthocephalan *E. gadi*, which was the most significant species in the classification of 4T plaice stocks in this study, reach maturity in the intestines of dozens of species of marine fish in the North Atlantic and use gammaridean and caprellid amphipods and mysids as intermediate hosts (Marcogliese, 1994).

Among influences on digenean infection parameters in fish are the distributions and abundances of molluscan intermediate hosts, where the parasites perform one or more generations of asexual reproduction. Brittle stars, which transmit *Fellodistomum* metacercariae to plaice, become infected by feeding on cercariae which develop in bivalve mollusks (Koie, 1980). In contrast to the other helminths used as markers here, the digenean *S. baccatum*, an important component of DFAs involving 4Vn plaice, is transmitted actively, through penetration of the skin by cercariae (Wolfgang, 1955). Mollusks that host the asexual reproduction of *S. baccatum* to the cercarial stage are whelks, especially the common or waved whelk (*Buccinum undatum*), which is widely distributed and commercially exploited in eastern Canadian waters.

Infection parameters of larval helminths in 4T-4Vn plaice may also be influenced by temporal and spatial distributions of the final hosts. The digenean *S. baccatum* matures and reproduces in the intestines of large piscivorous fish such as sea raven (*Hemitripterus americanus*) and Atlantic halibut (*Hippoglossus hippoglossus*) (Wolfgang, 1955), and the anisakine nematode *A. simplex* matures and reproduces in the stomachs of cetaceans (Rohde, 2005). Adults of the anisakines *Contracaecum osculatum* and *P. decipiens*, and the acanthocephalan *C. strumosum* occur, respectively, in stomachs and intestines of seals. Surveys of various demersals in waters off Nova Scotia, Canada (Marcogliese and McClelland, 1992; McClelland et al., 2000; McClelland and Martell, 2001a&b) revealed that infection parameters of *P. decipiens* and *Corynosoma wegeneri* increased with proximity to Sable Island, site of the largest grey seal (*Halichoerus grypus*) colony in the Northwest Atlantic.

Spatial disparities in prevalences and abundances of parasitic helminths in fish may also be traced to variation of physical parameters (temperature, salinity, depth, and bottom habitat) that influence distributions of the invertebrate precursor hosts (Williams and Jones, 1994). Our 4T plaice samples were collected at relatively uniform depths (43–88 m), and the substrates in both sampling areas ranged from sandy pelite to sandy gravel and had outcroppings of sandstone bedrock (see Loring and Nota[3]). The mean near-bottom temperature for sampling stations in eastern 4T was 1.62 (0.49–3.23)°C (n=5), but only 0.21 (–0.01–0.77)°C (n=9) for stations in western 4T. Near-bottom temperatures prevalent on the Magdalen Shoals are extremely low throughout the year (Swain et al., 1998), and small variations in temperature may have dramatic effects on the developmental and transmission rates of helminth parasites, as well as on the distributions and developmental rates of their poikilothermic intermediate hosts. Hence, the fact that acanthocephalan infections in southeastern Gulf plaice were much heavier than those found in plaice from the northwestern Magdalen Shoals may, to some extent, reflect the relative warmth of waters occupied in eastern 4T.

In summary, DFAs of abundances of the acanthocephalans *E. gadi* and *C. strumosum* support the findings of earlier mark-recapture studies (Powles, 1965), which indicate the presence of distinct northwestern (Miscou-Magdalen) and southeastern (Cape Breton) plaice stocks in the southern Gulf of St. Lawrence. Moreover, both parasite markers could be employed in future studies of migration and mixing of the two 4T stocks within 4T, and infection parameters of *E. gadi* alone may prove useful for detecting the presence of southeastern 4T migrants among stocks overwintering in Laurentian Channel waters of the Cabot Strait and 4Vn. The strength of our conclusions may be mitigated, however, by the fact that 4T and 4Vn plaice were sampled only during September and May, respectively, and samples from the two areas were taken more than a year apart. Hence, the possibility of seasonal or longer term variations in infection parameters of enteric helminths, e.g., *Fellodistomum* sp. and *E. gadi*, could not be investigated. Finally, given problems inherent in stepwise procedures (Power et al., 2005), statistical procedures employed in the present study, and in similar studies, could be improved upon. Power et al. (2005), for example, adopted an "all possible subsets" approach to selection of indicator parasites used in linear and quadratic discriminant analyses and in nonparametric classification of bogue landed at Spanish fishing ports. Efforts will be made to address these shortcomings in future surveys.

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