An Interdisciplinary Assessment of Winter Flounder Stock Structure

Gregory R. DeCelles and Steven X. Cadrin NOAA/UMass Cooperative Marine Education and Research Program School for Marine Science and Technology

Stock structure and management of winter flounder (Pseudopleuronectes americanus) was evaluated throughout its geographic range in the northwest Atlantic. Information on genetics, morphology, meristics, larval dispersal, life history traits, applied mark experiments and environmental signals was reviewed. Winter flounder in U.S. waters are currently managed as three separate units; Georges Bank, Gulf of Maine and Southern New England/Mid-Atlantic. Estuarine spawning, which likely plays an important role in reproductive isolation and population structure, appears to be obligate in southern New England, non-existent on Georges Bank and variable in the Gulf of Maine. Despite evidence for reproductively isolated estuarine groups, information from tagging, meristic analysis, and life history studies suggest extensive mixing, thereby supporting the current U.S. management regimen. In Canadian waters, winter flounder are managed as three units: western Scotian Shelf (NAFO Div. 4X), eastern Scotian Shelf (NAFO Div. 4VW), and the southern Gulf of St. Lawrence (NAFO Div. 4T). Genetic analysis and parasite markers indicate that these Canadian management units are distinct. However. examination of inshore and offshore winter flounder within division 4X suggests little interchange occurs between these groups. Several separate stocks probably exist within the 4T management area as well. Stock assessment and fishery management would likely benefit from stock composition analysis of mixed-stock fisheries of both U.S. and Canadian fishery resources.

Keywords: winter flounder, multidisciplinary stock identification

Contact author: Greg R. DeCelles, NOAA/UMass CMER Program, School for Marine Science and Technology, 838 South Rodney French Boulevard, New Bedford, MA 02744-1221 U.S.A. [tel: +1 508 910 6394 email: gdecelles@umassd.edu]

Introduction

Stock assessment and management require the recognition of self-sustaining groups within a species. However, stock identification, stock delineation and stock composition analysis of mixed-stock fisheries are needed for many fishery resources. Stock identification involves interdisciplinary analysis of life history information, genetics, geographic variation of phenotypic traits, movement and environmental signals (Cadrin, et al. 2005). Advances in several of these disciplines warrant reanalysis and re-evaluation of stock structure as new information arises (Begg and Waldman, 1999).

The objective of this paper is to synthesize information on the stock structure of winter flounder (*Pseudopleuronectes americanus*) across its geographic range. We review the benchmark case studies of winter flounder stock structure from a variety of backgrounds and disciplines. In addition, we consider the appropriateness of current management protocols in both the United States and Canada based on our review of the literature. Finally, we offer suggestions for further research to address some of the uncertainty that remains concerning the assignment of winter flounder stock units.

Early research on winter flounder was focused primarily on movement analysis, life history rates and meristic analysis. Over time, more disciplines were employed for winter flounder stock identification, and researchers began to use genetic analysis, parasitic characters and hydrodynamic modeling to investigate this species. Currently, newer methods such as otolith chemical analysis and telemetry tagging are being used to better understand the stock structure of winter flounder

Management of Winter Flounder in the United States and Canada

Winter flounder are found in coastal waters of the Northwest Atlantic from North Carolina to Newfoundland (Bigelow and Schroeder, 1953; Collette and Klein-MacPhee, 2002; Pereira et al., 2002). Winter flounder in U.S. waters are currently managed as three separate stocks; Gulf of Maine, Georges Bank and Southern New England/Mid-Atlantic (Figure 1; NEFSC 2003). The Georges Bank stock is defined as statistical areas 522, 525, 551, 552 and 562. The SNE/MA stock comprises statistical areas 521, 526, 533-538, 611 and 639. The Gulf of Maine stock includes the waters from Cape Cod Bay northward to the Canadian border, which comprises statistical areas 511-515.

Winter flounder in Canadian waters are managed as three separate resources (Figure 2): (1) Browns Bank, St. Marys Bay and Bay of Fundy winter flounder are managed concurrently in NAFO Div. 4X; (2) winter flounder from the Scotian Shelf and points eastward are managed together in NAFO Div. 4VW; and (3) winter flounder are managed as one unit in the Southern Gulf of St. Lawrence in NAFO Div 4T.

Review of Stock Identification Information

Life History Traits

Larval dispersal- Winter flounder form relatively distinct estuarine or coastal spawning units throughout its geographic range. While this species lays demersal eggs, the larvae are pelagic and remain in the water column for a period up to 60 days (Bigelow and Schroeder, 1953). Therefore, the amount of larval interchange between adjacent spawning areas plays an important role in determining the amount of localized population structure that exists. Pearcy (1962) sampled in Mystic River, Connecticut and found that the majority of winter flounder larvae were found in the bottom of the water column. He postulated that since net transport in the bottom of these estuaries was landward, estuarine hydrodynamics should retain flounder larvae. Further, Pearcy concluded that nursery and breeding grounds should be closely linked, based on the non-dispersive nature of early life stages.

Chant et al. (2000) modeled the hydrodynamics of Little Egg Inlet in New Jersey, an important nursery area for larval winter flounder, and determined that strong tidal currents create a horizontally advective flow pattern which deliver flounder larvae into inlets where they settle. Based on this finding, they postulated that currents may promote the retention of winter flounder larvae within their natal estuaries. This was supported by additional beam trawling surveys (N=48), which found higher abundances of larvae inside the inlets, compared to the adjacent channel.

In an earlier study, Crawford and Carey (1985) identified two probable spawning sites in a Rhode Island estuary. Using two-dimensional hydrodynamic models, they hypothesized that larvae originating from these locations would be retained within the estuary. Ichthyoplankton surveys revealed that larvae were retained within the estuary, at locations similar to those predicted by the hydrodynamic model. These studies suggest that estuarine spawning sites maximize larval retention, which in turn promotes localized population structure between inshore populations.

Life History Traits- Population parameters such as growth, abundance, maturity and recruitment can be used to distinguish among discrete stocks of fish because these parameters are phenotypic expressions of the interaction between genotypic and environmental influences (Begg, 2005). Several studies have indicated that Georges Bank winter flounder have significantly greater growth rates than inshore stocks and merit treatment as a separate stock. Lux (1973) examined scale annuli patterns (N=510) and found that growth rates for Georges Bank winter flounder. Building on the findings of Lux, Howe and Coates (1975), tagged 12,151 winter flounder in Massachusetts coastal waters, on Nantucket and Martha's Vineyard shoals, as well as on Georges Bank from 1960 to 1965. Growth rates were calculated based on 4,440 tag returns, and were found to be significantly different between each stock unit. Winter flounder from Georges Bank had the fastest growth rate, flounder north of Cape Cod (GNE/MA stock) had intermediate growth rates (Figures 3 and 4). Using aged

scale samples (n=3,035), Witherell and Burnett (1993) also found that winter flounder from the SNE/MA stock have faster growth rates than individuals from the Gulf of Maine stock (Figures 5 and 6).

Butts and Litvak (2007) tested the genetic basis for the faster growth exhibited by Georges Bank winter flounder. Female winter flounder from Passamaquoddy Bay, NB, were fertilized with sperm of males collected from Georges Bank and Passamaquoddy Bay. Larvae were raised under identical laboratory conditions for a period of 28 days. The growth of both larval groups was compared using a mixed-model nested ANOVA with respect to head depth, eye length, standard length and jaw length. Larvae sired by Georges Bank males exhibited significantly faster growth during certain stages of development than larvae sired by males from Passamaquoddy Bay when reared in a common environment, suggesting a genetic basis for the difference.

A clinal gradient of maturity at age has been observed for winter flounder with maturity occurring at older ages for individuals in northerly latitudes (Collette and Klein-MacPhee, 2002; Table 1). O'Brien et al. (1993) estimated age and length at 50% maturity ($A_{50\%}$ and $L_{50\%}$, respectively) for the management units of winter flounder in the United States. For Georges Bank winter flounder $A_{50\%}$ was estimated at 1.9 years for both sexes and $L_{50\%}$ of 25.6cm for males and 24.9cm for females. For SNE/MA winter flounder, $A_{50\%}$ and $L_{50\%}$ was estimated at 3.3 years and 29cm for males and 3.0 years and 27.6cm for females. For the Gulf of Maine fish $A_{50\%}$ and $L_{50\%}$ was estimated to be 3.3 years and 27.6cm for males and 3.5 years and 29.7cm for females.

Perlmutter (1947) found earlier maturity schedules than reported by O'Brien et al. (1993) for SNE/MA winter flounder. In the coastal waters of New York, average maturity occurred at ages 2 and 3 for males and females respectively, when individuals were between 20-25cm total length (Perlmutter, 1947). Witherell and Burnett (1993) reported maturity schedules similar to those found by O'Brien (1993). For SNE/MA fish, $A_{50\%}$ and $L_{50\%}$ was found to be 3.0 years and 28.3cm for females, and 3.1 years and 28 cm for males. Witherell and Burnett (1993) estimated $A_{50\%}$ and $L_{50\%}$ to be 3.3 years and 28.7 cm for females and 3.3 years and 27.2cm for males in the Gulf of Maine. The slowest maturity schedules have been observed in Newfoundland with average maturity occurring at age 6 and 25 cm for females and age 7 and 21 cm for males (Kennedy and Steele, 1971).

Differences in the timing and location of spawning events are useful stock identification criterion because they can lead to reproductive isolation among stocks by reducing gene flow (Bailey et al., 1999). Spawning has been shown to occur earlier in southerly latitudes for winter flounder (Table 2). Winter flounder spawning events in the SNE/MA area have been observed from December until May, and the timing of spawning varies with latitude. Lobell (1939) observed that spawning occurred in Long Island Sound, NY between December and May, although more recent observation (Monteleone, 1992) found that spawning in this area occurred in March and April. In Narragansett Bay, RI, spawning takes place from January through March (Buckley et al., 1991), and in Connecticut from February until April (Pearcy, 1962). Spawning typically occurs later in

the Gulf of Maine stock and has been observed from late January to May in Plymouth Harbor, MA (Scherer, 1984) and from March to May in Boothbay Harbor, ME (Bigelow and Schroeder, 1953). In Newfoundland, peak spawning has been observed from May to early June (Kennedy and Steele, 1971).

Morphology

Meristics- Several studies have used fin ray counts to differentiate between stocks of winter flounder. Geographic variations in fin ray counts between different stocks of fish suggest that there is little interchange between these groups, and reproductive isolation is likely. Values attained for meristic characters are the products of interactions between the genetics of an individual and its environment (Waldman, 2005). Perlmutter (1947) sampled 1,047 winter flounder from Georges Bank, the Gulf of Maine (Cape Cod Bay to Nova Scotia) and the southern New England/Mid-Atlantic area (New York and New Jersey). Fish taken from Georges Bank had significantly higher counts of anal, dorsal and pectoral fin rays than fish sampled at inshore locations. Fish collected from the Gulf of Maine (N=266) and the Southern New England/Mid-Atlantic area (N=703) showed significant differences in the pectoral fin ray counts, but no significant differences were observed when anal and dorsal ray counts were analyzed.

Lux et al. (1970) and Pierce and Howe (1977) followed up on the work of Perlmutter by conducting extensive meristic studies of winter flounder. Lux et al. (1970) concluded that winter flounder in U.S. coastal waters form three distinct stocks. Fish from Georges Bank (N=121) had the most anal and dorsal fin rays and were the most distinct from the other stocks. Samples taken from the Gulf of Maine (N=224) had the fewest fin rays, while samples from Southern New England/Mid-Atlantic (N=388) had intermediate fin ray counts. Pierce and Howe (1977) sampled young of the year winter flounder at 23 locations along the Massachusetts coast over a three year period. They were unable to detect significant differences in fin ray counts at an estuarine level. Although localized population structure has been indicated by both genetic and tagging studies (Crivello et al. 2004; Phelan 1992), the Pierce and Howe (1977) meristic data suggest that individual estuaries do not necessarily produce distinct stocks. However, significant differences were observed between fish sampled from the Gulf of Maine and from Southern New England/Mid-Atlantic area. Based on meristic characteristics, winter flounder in U.S. coastal waters form at least three distinct units.

Environmental signals

Patterns of parasitic infestation- Parasites can be useful tools in stock identification studies. If a fish becomes infected with a parasite that has a known endemic range, it can be inferred that the fish was within that area within the life span of the parasite (MacKenzie and Abaunza, 2005). When groups of fish have unique parasitic characters, limited interchange between those groups can be inferred. Scott (1982) examined parasitological differences between winter flounder in the southern Gulf of St. Lawrence (NAFO area 4T) and the Scotian Shelf (NAFO area 4W). Significant geographic variation was noted between the two areas for three parasite species; *Derogenes varicus, Fellodistomum furcigerum* and *Lecithaster gibbosus*. Scott (1982) concluded that based on these parasitological characteristics, winter flounder in the Gulf of St. Lawrence and

those on the Scotian Shelf constitute separate stocks. These findings were expanded upon by McClelland et al. (2005) who examined 189 adult winter flounder from four geographic regions: St. Marys Bay (50); Georges Bank (50); Browns Bank (41); and Sable Island Bank (48). Seven parasite species were examined, which included five species of digeneans and two species of larval nematodes. Individual fish could be identified to their sampling site with an 84% overall classification accuracy using a discriminant function analysis. Parasite characteristics provided evidence that the Georges Bank stock was distinct from groups of winter flounder in adjacent Canadian coastal waters.

Biochemical analysis- Similar to parasitic infestation, chemical contaminants can serve as acquired marks and be used to infer isolation or mixing among groups. Carr et al. (1991) collected winter flounder over two summers at Deer Island (Boston Harbor) and off Plymouth, MA. High concentrations of harmful organic contaminants are found in the sediment of Boston Harbor, which receives outflow from industrial activities. The Plymouth Harbor site is relatively pristine and was used as a control. Carr et al. (1991) measured several biochemical parameters for each group of fish and found that about 50% of the fish collected in Boston Harbor had apparent apoptotic hepatic lesions (AAHPC), while AAHPC was not detected in any of the fish collected from Plymouth Harbor. Other biochemical parameters (amino acid concentrations, glycogen levels) were also shown to differ significantly between the two sites. While this study was not conducted for stock identification purposes, these biomarkers have potential application for this purpose. Given the significant differences in chemical contamination between the two groups of sampled fish, it can be inferred that little or no interchange occurs between the two populations, despite their geographic proximity.

Genetic analysis

Microsatellite Analysis- Microsatellite analysis is one of the primary tools used by genetic researchers for stock identification. Microsatellites have very high levels of genetic variation that can be detected at individual loci, can be analyzed relatively easily and there are a large number of loci that can be screened (Wirgin and Waldman, 2005). Microsatellite studies of winter flounder in Canadian waters (McClelland et al. 2005) revealed the existence of at least four distinct stocks. Mature winter flounder (N=186) were sampled from four geographic locations; Georges Bank, Browns Bank, Sable Island Bank and St. Marys Bank. Fish from these areas were evaluated for genetic differences based on an analysis of four microsatellite loci. Significant genetic differences were observed between the four sampling locations. The Georges Bank sample was found to be the most genetically distinct, while the Browns Bank and Saint Marys samples had the least genetic dissimilarity. Fish were classified to their original group with 86-96% accuracy using a discriminant function analysis.

Microsatellite analysis has also proven useful for studying genetic differences at the estuarine level for this species. Crivello et al. (2004) collected 536 winter flounder larvae from three spawning areas (Niantic, Thames and Westbrook rivers) in Long Island Sound, NY using a bongo net from late February until April of 2001. Larvae from each group were then analyzed for genetic differences by measuring allele frequencies and

base pair lengths at six microsatellite loci. Significant genetic differences were observed between the three groups, suggesting localized stock structure. Of the 18 tests conducted (six loci at three sampling locations), 13 were found to deviate from the expected Hardy-Weinberg equilibrium. In addition, these differences were geographically based, with the greatest amount of genetic differences observed between the two most distant groups, and the least amount of difference between the two closest groups.

Gene expression- Hayes et al. (1991) found that winter flounder sampled from Nova Scotia, Newfoundland, Long Island Sound and the Bay of Fundy had a high gene copy number and a large tandem component for the antifreeze protein gene. Fourney et al. (1984) also found little variation in plasma antifreeze polypeptides from fish sampled from New York to Newfoundland. Winter flounder frequently encounter ice or very low temperatures in estuarine habitats, and as expected, possessed the most genes for the antifreeze protein. Gene expression was reduced in fish sampled from Georges Bank, Browns Bank, and Passamaquoddy Bay, New Brunswick, which may indicate that thee groups are not adapted to winter, estuarine habitats. In addition, Browns Bank and Georges Bank flounder had dissimilar copy numbers and tandem components, suggesting that these groups were genetically distinct, despite their close geographic proximity. However, these genetic differences need to be considered cautiously as only one fish was sampled from each location.

Movement Analysis

Applied Marks- Tagging studies can provide important insight into the stock structure of marine fish. Several comprehensive tagging studies have provided convincing evidence of natal homing in winter flounder. Homing is the tendency for individuals to return to a place formerly occupied instead of going to another similar place (Gerking, 1959).

Perlmutter (1947) tagged 10,809 winter flounder from New Jersey to Maine. This large geographic area was divided into 10 strata for analysis. 1,790 tags were recaptured and most individuals (94%) were recaptured within the stratum in which they were tagged, with limited movement observed during the spawning season. Saila (1961) tagged 952 winter flounder in a Rhode Island salt pond, of which 309 were recaptured. During the spawning months, most fish were recaptured in close proximity to their release sites, suggesting a strong tendency of individual homing to spawning sites. Phelan (1992) also found evidence for natal homing of individuals tagged within the Inner New York Bight as many individuals were recaptured in close proximity to their release location (i.e., breeding site) after over 100 days at liberty.

Analysis of seasonal movement patterns suggests that winter flounder in the SNE/MA area and in the Gulf of Maine exhibit markedly different patterns of movement. McCracken (1963) reviewed seasonal movement patterns of winter flounder and found that in the Gulf of Maine stock winter flounder undergo limited seasonal migrations. However, winter flounder in the SNE/MA area undergo more extensive migrations, typically leaving shallow bays and estuaries in the summer months as water temperatures increase above 15^oC. These findings have been corroborated by applied mark experiments. Perlmutter (1947) and Howe and Coates (1975) observed that winter

8

flounder movements in the Gulf of Maine stock were localized and confined to inshore waters. However, in the SNE/MA area individuals appeared to move more extensively during the summer months as water temperatures increased. All of these tagging studies documented a general trend for SNE/MA flounder to disperse to the south and east during the summer months (Perlmutter, 1947; Saila, 1961; Howe and Coates, 1975; Phelan, 1992). During this time, the localized inshore groups of winter flounder intermix in offshore waters, a phenomenon described by Phelan as a "dynamic assemblage".

However, evidence of contingent structure in winter flounder in the SNE/MA area has also been noted from tagging studies. Contingents are groups of fish that appear to maintain their integrity by engaging in a distinct pattern of seasonal migration not shared by other groups of fish (Clark, 1968). Phelan (1992) observed that some tagged winter flounder were caught offshore during the winter spawning months. Phelan postulated that these individuals likely did not spawn offshore, and were either late inshore spawners, or possibly did not spawn at all to conserve body mass. In addition, winter flounder have also been documented inshore year round in the SNE/MA area, suggesting that these individuals are perhaps, non-migratory (Olla et al., 1969; Wilk et al., 1977). In addition, contingent migrations may make a stock more resilient to overfishing, increase genetic diversity and cause variable susceptibility to anthropogenic impacts (Secor, 1999; Hilborn et al. 2003). Contingent migration patterns may have important implications in the stock structure of this species and merit further investigation.

Howe and Coates (1975) calculated that 1.7% of the fish that they tagged moved between the Gulf of Maine and the SNE/MA area. In addition there was little interchange (0.49%) between the SNE/MA (Nantucket Shoals) and Georges Bank. These findings suggest that the three management units of winter flounder in U.S. coastal waters are relatively discrete and reproductive isolation is likely.

McCracken (1963) observed seasonal distributions of winter flounder in several regions of Canada. In St. Marys Bay and Passamaquoddy Bay, New Brunswick (NAFO Div. 4X) winter flounder were found inshore during the winter and spring months and gradually dispersed offshore during the summer and fall as water temperatures increase above 15°C. In Northumberland Strait, Gulf of St. Lawrence (NAFO Div, 4T) flounder were also observed inshore, in shallow water during the winter and spring and were found to disperse to deeper water in the summer. However, in Pubnico Harbor, Nova Scotia (NAFO Div. 4X) winter flounder remained inshore year round as water temperatures seldom exceeded 14°C. The contrasting movement patterns of individuals from Pubnico Harbor and those from St. Marys Bay and Passamaquoddy Bay suggests that different groups of winter flounder may be present on the western Scotian Shelf.

Hanson and Courtenay (1996) found that winter flounder in the Southern Gulf of St. Lawrence began to enter the Miramichi estuary in late autumn. The majority of these fish overwintered in the estuary, where water temperatures were warmer than the Southern Gulf, and where a refuge existed from flowing ice packs. In spring as the salinity of the estuary was reduced by snow melt adult fish left the estuary and migrated to spawning grounds in coastal waters.

Kennedy and Steele (1971) observed that winter flounder in Conception Bay, Newfoundland exhibited seasonal distribution patterns that were similar those undertaken by winter flounder in the SNE/MA area. Individuals remained inshore in shallow, estuarine waters from autumn until the spring. After spawning in May and June adults migrated offshore to deeper waters to feed. Similar movement patterns were observed by Van Guelpen and Davis (1979) also in Conception Bay, Newfoundland, where storminduced turbulence or the formation of ice in shallow waters caused winter flounder to temporarily emigrate to deeper inshore waters.

In summary, tagging information suggests limited mixing occurs between the current management areas. Seasonal movement patterns also vary by geographic region. South of Cape Cod, it appears that winter flounder mix in coastal waters in summer, but home to natal estuaries to spawn in winter. In more northern habitats, residence in estuarine habitats is variable, with some groups spawning on offshore banks, others wintering in estuaries, and others occupying estuaries briefly. These different behaviors with respect to estuarine spawning may have important implications for reproductive mixing or isolation among spawning groups.

Synthesis and Conclusion

Basis for Assignment of Management Stock Units in the United States and Canada Prior to 1996, winter flounder were managed as four stock units in the U.S. waters of the northwest Atlantic: Mid-Atlantic, Southern New England, Georges Bank and Gulf of Maine. In 1996 (at the 21st Stock Assessment Workshop [SAW]), the Southern New England and Mid- Atlantic groups were combined to form a single unit (SNE/MA) for assessment purposes (Shepherd et al. 1996). The Workshop concluded that while there was evidence of localized estuarine populations present in the two areas, the fisheries in these regions are typically conducted when winter flounder populations are intermixed in coastal offshore waters. Tagging results indicated some degree of mixing between these two areas (Perlmutter 1947; Howe and Coates, 1975; Phelan, 1992), provided the primary basis for combining the SNE and MA areas. The Workshop also examined life history traits and found that growth rates were more variable within each management unit than between the two units. Finally, survey data showed that the length structure in both units was similar, and therefore, these two areas should be merged into a single management unit.

The 21st SAW also considered the stock structure of Gulf of Maine winter flounder, and concluded (based on tagging studies, primarily Howe and Coates, 1975), that there was sufficient interchange between winter flounder groups in the Gulf of Maine to manage them as a single unit (Cadrin et al. 1996). The 28th SAW considered the stock structure of Georges Bank winter flounder and concluded that based on (a) tagging data (Perlmutter 1947; Howe and Coates, 1975), (b) meristic analysis (Lux, 1973) and (c) differences in life history characteristics (Lux et al., 1970) that the winter flounder on Georges Bank should be managed as a separate entity.

Management units of winter flounder in Canadian waters are based upon geographic distribution patterns inferred from Canadian summer research vessel surveys on the Scotian Shelf (Stobo et al. 1997; DFO 1997) and in the Southern Gulf of St Lawrence (Morin et al. 2002; DFO 2005). Prior to 1994 on the Scotian Shelf (NAFO Divs. 4VWX), yellowtail flounder, witch flounder and American plaice were managed as one stock complex because a large component of the catches was landed as "unspecified flounders". During this period, winter flounder in the area were excluded from management considerations (DFO 2002a). In 1994, the Scotian Shelf management area was divided into eastern (NAFO Div. 4VW) and western components (NAFO Div. 4X), winter flounder was included in these management components, and overall TACs (for the four flatfish species combined) were established for the two regions based on catch histories (DFO 2002b). Management of the four species together under area TACs was an explicit recognition that it has been impossible to obtain reliable landings statistics for each individual species.

In the Southern Gulf of St. Lawrence (NAFO Div. 4T), winter flounder came under TAC management in 1996, although the first assessment of this stock was conducted in 1994 (DFO 2005). Several localized stock units (or partially isolated breeding populations) are thought to exist in the region based on geographic differences in resource survey abundance trends (DFO 2002c), but information on which to assess local stock units is extremely limited (Morin et al. 2002). Although the Fisheries Resource Conservation Council (FRCC) has recommended since 1998 that DFO science, managers and industry collaborate to implement local management measures, this effort has been hampered as the annual (since September 1971) groundfish survey in Div. 4T does not cover the inshore habitat of winter flounder (e.g., water depths < \sim 20 m) (Morin et al. 2002).

Critique of Assigned Stock Units

Based upon this review of the literature, it appears that management of winter flounder fisheries in U.S. waters is generally consistent with multidisciplinary information on stock structure. Evidence from tagging studies, life history studies, and meristic analysis indicates that the winter flounder in the Gulf of Maine area distinct from those in the Southern New England/Mid-Atlantic area. Tagging studies and genetic analyses in the SNE/MA area show evidence for localized population structure, and numerous local stocks may exist in this region. However, it would be very challenging to identify and manage each of these units as a discrete entity. In addition, the majority of commercial and recreational fishing effort occurs during the summer and fall months, when adult fish from each localized stock are mixed in coastal offshore waters. Stock composition analysis of the mixed population fisheries in this area would help to address questions regarding the amount of localized population structure present and the relative contribution of each local stock.

Georges Bank winter flounder exhibit the highest growth rates and fin ray counts, and exhibit little interchange with inshore stocks. Therefore, this group also merits treatment as a single transboundary resource in U.S. and Canadian waters of Georges Bank. Based on growth rates, antifreeze protein gene copy numbers, microsatellite DNA analysis and

parasite characteristics, the Georges Bank unit is also separate from adjacent winter flounder units on the Scotian Shelf.

Gulf of Maine winter flounder are the least studied of those in U.S. waters. At present, there is no research that distinguishes Gulf of Maine winter flounder from those found in inshore Canadian waters of the Bay of Fundy or the Scotian Shelf. Tagging studies that estimate the amount of interchange between flounder in U.S. and Canadian waters may resolve some of this ambiguity. Data from trawl surveys may also be examined to detect any persistent differences in life history traits between inshore flounder populations in the northern Gulf of Maine and southern New Brunswick.

Canadian winter flounder management units are assigned on the basis of abundance patterns derived from resource surveys. However, genetics and life history traits (such as growth and maturity) might also be utilized in further defining these units.

At least two stocks of winter flounder exist within the western Scotian Shelf (NAFO Div. 4X). McClelland et al. (2005) found that winter flounder on Browns Bank are distinct from those inhabiting St. Marys Bay based on microsatellite analysis and parasitic characteristics. In addition, Hayes et al. (1991) noted differences in the expression and copy number of antifreeze protein genes between Browns Bank winter flounder and those in the inshore areas of the Bay of Fundy. Currently, all winter flounder in NAFO Div. 4X are managed under a single total allowable catch (TAC) value encompassing four different flounder species.

Winter flounder on the eastern Scotian Shelf (NAFO Div. 4VW) is comprised of a relatively localized population that resides on Sable Island Bank. These fish are geographically distinct, and parasitic comparisons suggest these fish are significantly different from those in the Southern Gulf of St. Lawrence (NAFO Div. 4T) and on Browns Bank (NAFO Div. 4X). Microsatellite analysis also suggests that winter flounder on the eastern Scotian Shelf are distinct from those found elsewhere in Canadian waters.

Winter flounder in the southern Gulf of St. Lawrence stock (NAFO Div. 4T) are geographically separate from those inhabiting the eastern Scotian Shelf stock, and parasitic assemblages differ significantly between the two areas. Genetic analysis also suggests the Gulf of St. Lawrence is distinct from other stocks. Although winter flounder in NAFO Div. 4T are likely composed of a number of localized populations (which support a number of localized fisheries for lobster bait and limited human consumption markets), due to data limitations, the resource is presently managed as a single entity.

The management of winter flounder in both Canadian and U.S. waters would likely benefit from a stock composition analysis of fishery catches, especially in areas where ambiguity remains concerning stock boundaries (i.e. between the northern Gulf of Maine and southern Nova Scotia). Several disciplines including genetics, meristics, morphometrics and chemical analysis are available to address the questions that still remain. A holistic approach incorporating several of these techniques would likely provide the most useful information for fisheries management decisions.

References

- Bailey, K. M., Quinn, T. J., II, Bentzen, P., and Grant, W. S. 1999. Population structure and dynamics of walleye pollock, *Theragra chalcogramma*. Advances in Marine Biology 37: 180-255.
- Begg, G.A. 2005. Life history parameters. *IN:* Stock Identification Methods, Applications in Fishery Science. *Edited by* S.X. Cadrin, Friedland, K.D., and Waldman, J.R. Elsevier Academic Press, San Diego. pp. 119-151.
- Begg, G.A., and Waldman, J.R. 1999. An holistic approach to fish stock identification. Fisheries Research **43**: 35-44.
- Bigelow, H. B., and Schroeder, W. C. 1953. Fishes of the Gulf of Maine. U.S. Fish and Wildlife Service, Fishery Bulletin **53**: 577 pp.
- Buckley, L. J., Smigielski, A. S., Halavik, T. A., Caldarone, E. M., Burns, B. R. and Laurence, G. C. 1991. Winter flounder *Pseudopleuronectes americanus* reproductive success. II. Effects of spawning time and female size on size, composition and viability of eggs and larvae. Marine Ecology Progress Series 74: 125-135.
- Butts, I. A. E., and Litvak, M. K. 2007. Parental and stock effects on larval growth and survival to metamorphosis in winter flounder (*Pseudopleuronectes americanus*) Aquaculture, doi:10.1016/j.aquaculture.2007.04.012
- Cadrin, S.X., Friedland, K.D., and Waldman, J.R. (*Editors*). 2005. Stock Identification Methods, Applications in Fishery Science. Elsevier Academic Press, San Diego.
- Cadrin, S., Howe, A., Correia, S., Shepherd, G., Lambert, M., Gabriel, W., and Grout, D. 1996. An index-based assessment of winter flounder populations in the Gulf of Maine. Northeast Fisheries Science Center Reference Document 96-05a.
- Carr, R. S., Hillman, R.E. and Neff, J.M. 1991. Field assessment of biomarkers for winter flounder. Marine Pollution Bulletin 22: 61-67.
- Chant, R. J., Curran, M. C., Able, K.W., and Glenn, S.M. 2000. Delivery of winter flounder (*Pseudopleuronectes americanus*) larvae to settlement habitats in coves near tidal inlets. Estuarine, Coastal and Shelf Science 51: 529-541.
- Clark, J. 1968. Seasonal movements of striped bass contingents in Long Island Sound and the New York Bight. Transactions of the American Fisheries Society **97**: 320-343.
- Collette, B.B., and Klein-MacPhee, G. 2002. Fishes of the Gulf of Maine. Smithsonian Press, Washington.
- Crawford R. E., and Carey, C. G. 1985. Retention of winter flounder larvae within a Rhode Island salt pond. Estuaries **8**: 217-227.
- Crivello J. F., Danila, D. J., Lorda, E., Keser, M., and Roseman, E.F. 2004. The genetic stock structure of larval and juvenile winter flounder larvae in Connecticut waters of eastern Long Island Sound and estimations of larval entrainment. Journal of Fish Biology 65: 62-76.
- DFO (Department of Fisheries and Oceans). 1997. Southwest Nova winter flounder,

American plaice and yellowtail flounder. DFO Science Stock Status Report A3-21: 8 pp.

- DFO (Department of Fisheries and Oceans). 2002a. Updates on selected Scotian Shelf groundfish stocks in 2001. American plaice, yellowtail flounder, and winter flounder on the western Scotian Shelf (Div. 4X). DFO Science Stock Status Report A3-3: pp. 29-31.
- DFO (Department of Fisheries and Oceans). 2002b. Updates on selected Scotian Shelf groundfish stocks in 2002. American plaice, yellowtail flounder, and winter flounder on the western Scotian Shelf (Div. 4X). DFO Science Stock Status Report A3-35: pp. 24-26.
- DFO (Department of Fisheries and Oceans). 2005. Winter flounder in the southern Gulf of St. Lawrence (Div. 4T). DFO Canadian Science Advisory Secretariat Science Advisory Report 2005/015: 6 PP.
- Fourney, R. M., Hew, C. L., Joshi, S. B. and Fletcher, G. L. Comparison of antifreeze polypeptides from Newfoundland, Nova Scotia, New Brunswick and Long Island Sound. Comparative Biochemical Physiology 78B: 791-796.
- Gerking, S. D. 1959. The restricted movement of fish populations. Biological Reviews of the Cambridge Philosophical Society **34**: 221-242.
- Hanson, J. M., and Courtenay, S. C. 1996. Seasonal use of estuaries by winter flounder in the southern Gulf of St. Lawrence. Transactions of the American Fisheries Society 125: 705-718.
- Hayes, P.H., Davies. P. L., and Fletcher, G. L. 1991. Population differences in antifreeze protein gene copy number and arrangement in winter flounder. Genome 34: 174-177.
- Hilborn, R. T. P. Quinn, D. E. Schindler, and D. E. Rogers. 2003. Biocomplexity and fisheries sustainability. Proceedings of the Natural Academy of Sciences, 100: 6564-6568.
- Howe, A. B., and Coates, P. G. 1975. Winter flounder movements, growth and mortality off Massachusetts. Transactions of the American Fisheries Society 104: 13-29.
- Kennedy, V. S., and Steele, D. H. 1971. The winter flounder (*Pseudopleuronectes americanus*) in Long Pond, Conception Bay, Newfoundland. Journal of the Fisheries Research Board of Canada 28: 1153-1161.
- Lobell, M. J. 1939. A biological survey of the salt waters of Long Island, 1938. Report on certain fishes: Winter flounder (*Pseudopleuronectes americanus*). New York Conservation Department, 28th Annual Report, Supplemental Part I. pp. 63-96.
- Lux, F. E. 1973. Age and growth of the winter flounder, *Pseudopleuronectes americanus*, on Georges Bank. Fishery Bulletin **71**: 505-512.
- Lux, F. E., Peterson, A.E., and Hutton, R.F. 1970. Geographical variation in fin ray number in winter flounder, *Pseudopleuronectes americanus* (Walbaum), off Massachusetts. Transactions of the American Fisheries Society **99**: 483-488.
- MacKenzie, K., and Abaunza, P. 2005. Parasites as biological tags. *IN:* Stock Identification Methods, Applications in Fishery Science. *Edited by* S.X. Cadrin, Friedland, K.D., and Waldman, J.R. Elsevier Academic Press, San Diego. pp. 211-226.
- McCracken, F. D. 1963. Seasonal movements of the winter flounder

Pseudopleuronectes americanus (Walbaum), on the Atlantic coast. Journal of the Fisheries Research Board of Canada **20**: 551-586.

- McClelland, G., Melendy, J., Osborne, J., Reid, D., and Douglas, S. 2005. Use of parasite and genetic markers in delineating populations of winter flounder from the central and south-west Scotian Shelf and north-east Gulf of Maine. Journal of Fish Biology 66: 1082-1100.
- Monteleone, D. M. 1992. Seasonality and abundance of ichthyoplankton in Great South Bay, New York. Estuaries: **15**: 230-238.
- Morin, R., Forest, I., and Benoit, H. 2002. Status of NAFO Division 4T winter flounder, February 2002. Canadian Science Advisory Secretariat Research Document 2002/033: 56 pp.
- Northeast Fisheries Science Center. 2003. Report of the 36th Northeast Regional Stock Assessment Workshop (36th SAW): Stock Assessment Review Committee (SARC) consensus summary of assessments. Northeast Fisheries Science Center Reference Document 03-06.
- O'Brien, L. J., Burnett, J., and Mayo, R. K. 1993. Maturation of nineteen finfish species off the northeastern coast of the United States, 1985-1990. NOAA Technical Report NMFS **113**: 66p.
- Olla, B. L., Wiklaund, R. and Wilk, S. 1969. Behavior of winter flounder in a natural habitat. Transactions of the American Fisheries Society. **98**: 717-720.
- Pearcy, W. G. 1962. Ecology of an estuarine population of winter flounder, *Pseudopleuronectes americanus* (Walbaum). Parts I-IV. Bulletin of the Bingham Oceanographic Collection 18: 1-78.
- Pereira, J.J., Goldberg, R., Ziskowski, J.J., Berrien, P.L., Morse, W.W., and Johnson, D.L. 1999. Winter flounder, *Pseudopleuronectes americanus*, Life history and habitat characteristics. NOAA Technical Memorandum NMFS-NE-**138**: 39 pp.
- Perlmutter, A. 1947. The blackback flounder and its fishery in New England and New York. Bulletin of the Bingham Oceanographic Collection **11**: 1-92.
- Phelan, B. A. 1992. Winter flounder movements in New York Inner Blight. Transactions of the American Fisheries Society **121**: 777-784.
- Pierce, D. E., and Howe, A. B. 1977. A further study on winter flounder group identification off Massachusetts. Transactions of the American Fisheries Society 106: 131-139.
- Saila, S. B. 1961. A study of winter flounder movements. Limnology and Oceanography **6**: 292-298.
- Secor, D. H. 1999. Specifying divergent migrations in the concept of stock: the contingent hypothesis. Fisheries Research, **43**: 13-34.
- Scherer, M. D. 1984. The ichthyoplankton of Cape Cod Bay. *IN*: Observations on the ecology and biology of western cape Cod Bay, Massachusetts. Edited by J. D. and D. Merriman. Springer and Verlag, Berlin. pp. 151-190.
- Scott, J. S. 1982. Digenean parasite communities in flatfishes of the Scotian Shelf and southern Gulf of St. Lawrence. Canadian Journal of Zoology **60**: 2804-2811.
- Shepherd, G., Cadrin, S., Correia, S., Gabriel, W., Gibson, M., Howe, A., Howell, P., Grout, D., Lazar, N., Lambert, M., and Ling, W. 1996. Assessment of winter flounder in Southern New England and the Mid-Atlantic. Northeast Fisheries Science Center Reference Document 96-05b: 112 pp.

- Stobo, W.T., Fowler, G.M., and Smith, S.J. 1997. Status of 4X winter flounder, yellowtail flounder, and American plaice. Canadian Stock Assessment Secretariat Research Document 97/105:
- Van Guelpen, L., and C.C. Davis. 1979. Seasonal movements of the winter flounder, *Pseudopleuronectes americanus*, in two contrasting inshore locations in Newfoundland. Transactions of the American Fisheries Society **108**: 26-37.
- Waldman, J. R. 2005. Meristics. *IN:* Stock Identification Methods, Applications in Fishery Science. *Edited by* S.X. Cadrin, Friedland, K.D., and Waldman, J.R. Elsevier Academic Press, San Diego. pp. 197-210.
- Wilk, S. J., Morse, W. W., Ralph, D. E., and Azarovitz, T. R. 1977. Fishes and associated environmental data collected in New York Bight, June 1974- June 1975. NOAA Technical Report NMFS SSRF 716.
- Wirgin, I., and Waldman, J. R. 2005. Use of nuclear DNA in Stock Identification: Single-Copy and Repetitive Sequence Markers. *IN:* Stock Identification Methods, Applications in Fishery Science. *Edited by* S.X. Cadrin, Friedland, K.D., and Waldman, J.R. Elsevier Academic Press, San Diego. pp. 331-370.
- Witherell, D.B., and Burnett, J. 1993. Growth and maturation of winter flounder, *Pseudopleuronectes americanus*, in Massachusetts. Fishery Bulletin **91**: 816-820.

	age at 50)%	length at 50	% maturity	
	maturity	(v)	(cm)	,	
	maturity	(y)	(UIII)		
Location	Male	Female	male	female	Source
					Kennedy & Steele
Newfoundland	6.0	7.0	21	25	1971
North of Cape Cod	3.3	3.5	27.6	29.7	O'Brien et al. 1993
· · ·					Witherell & Burnett
North of Cape Cod	3.3	3.3	27.2	28.7	1993
Georges Bank	1.9	1.9	25.6	24.9	O'Brien et al. 1993
					Witherell & Burnett
South of Cape Cod	3.1	3.0	28	28.3	1993
South of Cape Cod	3.3	3.0	29	27.6	O'Brien et al. 1993
New York	2.0	3.0	20-25	20-25	Perlmutter 1947

Table 1. Size and age at maturity of winter flounder by geographic area.

Table 2. Spawning seasons of winter flounder by location (months during spawning season are shaded grey).

Location	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Source
Newfoundland													
Boothbay Harbor Gulf of Maine													Kennedy & Steele 1971 Bigelow & Schroeder 1953
													Scherer 1984
Narragansett													
Bay													Buckley et al. 1991
Long Island													
Sound													Monteleone 1992
Connecticut													Pearcy 1962



Figure 1. Statistical areas used to define winter flounder stocks in the coastal waters of the United States.



Figure 2. Geographical areas used to define stock boundaries for winter flounder in Canadian coastal waters.



Figure 3. Growth rates of female winter flounder calculated from tag return data (Howe and Coates, 1975).

Figure 4. Growth rates of male winter flounder calculated from tag return data. Growth rates for Gulf of Maine males were not available due to a low number of tag returns (Howe and Coates, 1975).





Figure 5. Growth rate of female winter flounder calculated from aged scale samples (Witherell and Burnett, 1993).

Figure 6. Growth rates of male winter flounder calculated from aged scale samples (Witherell and Burnett, 1993).

