Distinguishing trophic and habitat partitioning among sympatric populations of the estuarine fish *Osmerus mordax* Mitchill

F. LECOMTE AND J. J. DODSON*

Département de biologie, Université Laval, Pavillon Vachon, Québec, G1K 7P4, Canada

Sympatric populations of rainbow smelt *Osmerus mordax* of the St Lawrence middle estuary exhibited distinct morphologies suitable for exploiting either pelagic or benthic habitats in the absence of consistent morphological features related to specific diets. The magnitude of morphological differentiation was elevated and constant over 3 years even though statistically significant differences were observed among samples within populations. Both populations were spatially segregated in the estuary but some population mixing occurred at low densities of rainbow smelt. Diet analysis revealed that both populations opportunistically feed on large macrozooplankton. An amended version of Schoener's *D* was developed and used to compare the magnitude of niche overlap. It revealed that the diet of the middle estuary rainbow smelt differed according to the area where samples were collected but no differences existed between populations. The exploitation of ecologically distinct habitats appeared to be effective in maintaining morphologically distinct sympatric populations within an estuarine setting in the absence of diet differentiation. © 2005 The Fisheries Society of the British Isles

Key words: diet; habitat segregation; morphological differentiation; resource partitioning; sympatric populations.

INTRODUCTION

For sympatric populations, resource partitioning involves the exploitation of distinct niches to reduce competition through the evolution of resource-based polymorphisms (Skúlason & Smith, 1995; Smith & Skúlason, 1996). Correlating morphology and resource utilization represents the first step in illustrating the existence of resource polymorphism among species pairs (Schluter, 1995). In post-glacial species pairs, the nature of the polymorphism is diverse, but it appears that feeding polymorphisms are the most common (Smith & Skúlason, 1996; Robinson & Schluter, 2000). Typically, benthivorous (*e.g.* low gill raker count) *v.* planktivorous morphotypes (*e.g.* high gill raker count) are observed. In most cases, this dichotomy is paralleled by habitat-specific distributions (benthic and pelagic) and differential growth patterns (normal and dwarf).

*Author to whom correspondence should be addressed. Tel.: +1 418 656 3102; fax: +1 418 656 2043; email: julian.dodson@bio.ulaval.ca

Although correlating morphology with resource utilisation appears a relatively simple task, observed diet differences may be caused by either truly selective behaviour (for prey and habitat) or simply by the availability of prey in distinct areas. Furthermore, selection may be more important during periods of low productivity. Hence, adequate spatial and temporal coverage is mandatory to yield evidence sufficiently robust to demonstrate the relative importance of habitat segregation independent of prey selection. The perspective that spatial segregation might evolve independently of trophic segregation should not be discarded *a priori* as a mechanism leading to the sympatric coexistence of distinct morphotypes.

Smelts (genus *Osmerus* spp.) are small pelagic species found in both freshwater and marine systems throughout the northern hemisphere (Nellbring, 1989). The rainbow smelt *Osmerus mordax* (Mitchill) is native to north-eastern North America (Scott & Crossman, 1974; Scott & Scott, 1988). Two glacial races are recognized, representing an Acadian and an Atlantic lineage, (Baby *et al.*, 1991; Bernatchez, 1997) but they receive no specific taxonomic recognition at this time. These two races came into secondary contact *c.* 8000 years ago in the St Lawrence middle estuary (Bernatchez, 1997). Today, two genetically distinct sympatric populations are found in the middle estuary (Bernatchez & Martin, 1996). Population genetics studies have revealed a significant amount of past and contemporary gene flow between the sympatric populations implying some level of hybridization between the two historical lineages (Bernatchez & Martin, 1996; Bernatchez, 1997; Lecomte & Dodson, 2004; unpubl. data). Mt DNA lineage frequency within the two contemporary populations shows that they are dominated by either one of the two ancestral lineages. The north shore population (NSP) is composed of 80·1% Atlantic lineage while the south shore population (SSP) is composed of 79·3% Acadian lineage (Bernatchez & Martin, 1996; Lecomte & Dodson, 2004). Both early life history constraints and adult resource partitioning contribute to the genetic isolation of this sympatric pair. Lecomte & Dodson (2004) showed that the two populations spawn in distinct habitats (SSP spawning in tributaries and NSP spawning in the St Lawrence River itself) and larvae are found in population-specific nursery habitats (SSP in south shore shoals and NSP in middle estuary channels). In addition, adult populations are well differentiated morphologically (Lecomte & Dodson, 2004), and morphological characteristics correlate to the spatial distribution of the two populations among distinct habitats. The superficially 'pelagic' NSP is found in deep channels of the north shore whereas the superficially 'benthic' SSP inhabits the south shore shoals (Lecomte & Dodson, 2004). The resulting phenotype-environment association, however, does not appear to involve diet differentiation as both populations consume the same prey. Such an observation is inconsistent with many previous studies showing that habitat-specific distributions are normally associated with differences in diet.

These observations raise two concerns about resource partitioning studies, one related to ecosystem setting and one related to methodological approach. Firstly, estuarine ecosystems are very different from freshwater lakes where most studies of resource polymorphisms are conducted. Estuaries are far more productive and their physical settings far more heterogeneous than in north temperate lakes. The nature of resource partitioning may therefore be more

varied in such heterogeneous ecosystems. Secondly, a generalized shortcoming of many resource partitioning studies, as introduced above, is the failure to clearly distinguish between the partitioning of food and the partitioning of habitat. This is largely associated with limited spatio-temporal coverage of patterns of distribution and diet. Opportunistic feeding across a highly heterogeneous prey field could be erroneously interpreted as selective feeding in specific habitat types. In addition, traditional methods used to test for niche overlap, such as Schoener's D (Schoener, 1968), fix arbitrary values to delineate distinct or overlapping feeding niches with no estimate of variance and hence no estimate of statistical validity (Feinsinger *et al.*, 1981; Bacheler *et al.*, 2004).

The objective of this study was to confirm the hypothesis that the sympatric rainbow smelt populations of the St Lawrence estuary partition habitat rather than food resources. To do so, procedures were developed to distinguish between habitat and dietary resource partitioning: 1) the temporal stability was established of the morphological differentiation, documented by Lecomte & Dodson (2004), originally based on only 1 year of sampling, 2) the spatial and temporal scales of population distribution were increased to confirm the segregation of the two populations throughout the estuary and throughout an entire growth season, 3) this extended spatial-temporal sampling was exploited to analyse *in situ* population specific diets and 4) to circumvent the shortcomings of traditional methods of estimating niche overlap, a new method of analysis permitting the statistical comparison of similarity (or lack thereof) in diets was developed.

MATERIALS AND METHODS

STUDY AREA

The St Lawrence (Fig. 1) is one of the largest river systems in the world in terms of length (3 260 km), freshwater outflow (*c.* 400 km^3 year^{-1}) and the surface area of its drainage basin (1 610 000 km^2) that holds over 25% of the world's fresh water (Vincent & Dodson, 1999). The St Lawrence middle estuary, where fresh water mixes with salt water, stretches 180 km and is 17 km wide on average (Fig. 1). This region is characterized by important tidal energy (up to 6 m tidal amplitude at Québec City) freshwater discharge (12 380 m^3 s^{-1} on average at Québec City) and saltwater intrusion. In the first 50–60 km of the saltwater-freshwater transition zone, an estuarine turbidity maximum (ETM) develops and includes most of the salinity gradient between fresh water and the marine region of the estuary (Fig. 1). Downstream of the ETM, the environmental gradients (temperature and salinity) are less steep and species composition is more marine in nature (Dodson *et al.*, 1989). The circulation within the middle estuary is greatly affected by tidal movements and freshwater outflow. Other important physical processes are also involved in the circulation (*i.e.* Coriolis force, vertical mixing, bathymetry and two-layer circulation). The saline intrusion occurs mainly along the north shore while most of the freshwater outflow occurs along the south shore (Fig. 1). The middle estuary has highly heterogeneous bathymetry; the north shore is typified by deep channels starting near the shore while large bays and shoals are found along the south shore (Fig. 1).

MORPHOLOGICAL DIFFERENTIATION

Samples used to assess morphological differentiation between spawning populations were obtained by collecting anadromous rainbow smelt caught during the 1997, 1998 and 1999 spawning seasons (Table I and Fig. 1). The inclusion of 3 years of sampling allowed
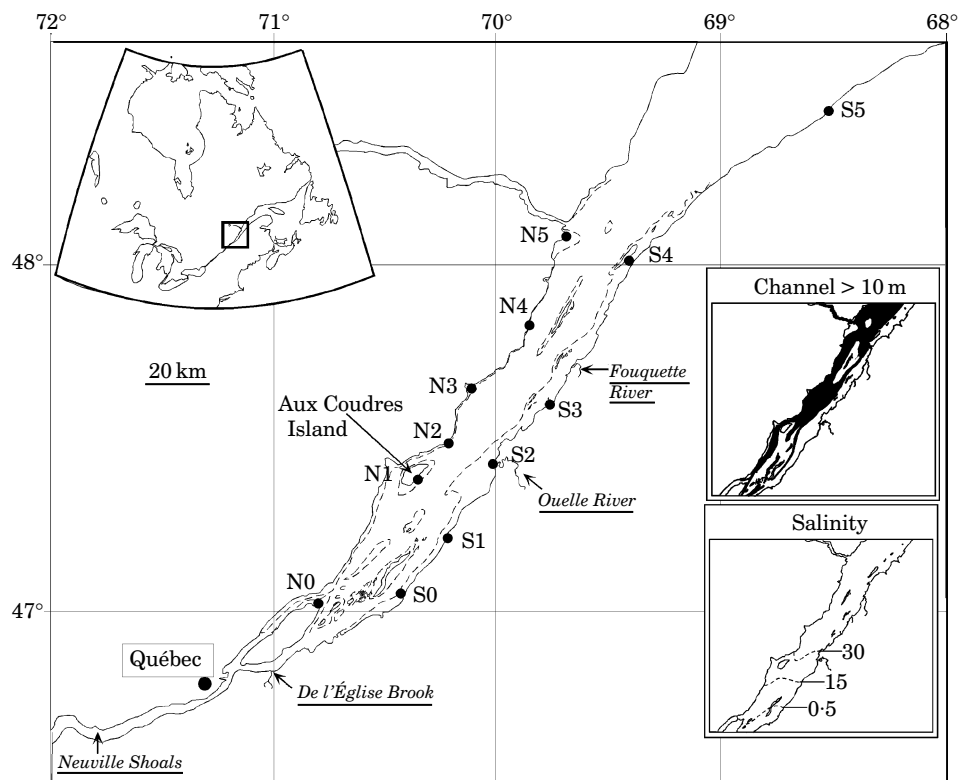
FIG. 1. Study area, location of channels, salinity gradient (isohalines at high tide) and shallow shoals (---, <2 m depth at low tide). The sampling sites are indicated by codes referring to their position. Spawning sites of the NSP are located nearby Neuville shoal and the SSP spawning tributaries are all located along the south shore and include De l'Église Brook, Ouelle River and Fouquette River.

TABLE I. Total number of rainbow smelt collected and basic descriptors as a function of population identity (north shore population, NSP; south shore population, SSP) and the site of capture. A *t*-test was used to compare mean fork length of populations (fish with gut content, GC) sampled in the four areas (region-shore)

Region	Shore	Population	Total number	Number with GC	Per cent with GC	Mean L_F (mm)	Mean L_F with GC (mm)	Mean GC dry mass (mg)
ETM	North	NSP	102	85	83·3	145·0	144·6	118·7
		SSP	10	10	100·0	148·6	148·6	91·3
	South	NSP	75	58	77·3	148·7	149·2	52·3
		SSP	53	41	77·4	155·7	153·6	115·2
Marine	North	NSP	116	45	38·8	149·7	151·4[1]	36·2
		SSP	27	14	51·9	143·5	138·5[1]	70·0
	South	NSP	24	19	79·2	150·9	150·9	116·4
		SSP	190	161	84·7	161·4	161·9	198·5

[1]*t*-test, significant at $\alpha = 0.05$, NSP is larger.

if differentiation was stable among years and not a sampling artefact to be estimated. The SSP was sampled from their three active spawning sites (de l'Église Brook 1997–1998–1999, Ouelle River 1998–1999 and Fouquette River 1997–1998–1999). A minimum of 200 fish were collected at each site to obtain rainbow smelt of every sex and cohort. Analyses of the 1998 samples were presented by Lecomte & Dodson (2004). The NSP spawn directly in the St Lawrence fluvial estuary (rather than within tributaries) and the time of spawning coincides roughly with the spring break-up of ice in the main river (Lecomte & Dodson, 2004) when sampling is restricted. As the NSP spawns before all other populations and migrates downstream following spawning, post spawning adults caught in April at Aux Coudres Island in 1997, 1998 and 1999 (N1 on Fig. 1) were used as an alternative. Genetic analysis using both mt DNA and microsatellite markers confirmed they belonged to the NSP (Lecomte & Dodson, 2004; unpubl. data).

Morphological measurements were made using an image analysis system (SigmaScan Pro 3.0; Jandel software) or an electronic calliper for measurements that exceeded system capacity. A group of 18 morphometric variables and two meristic counts were selected for their power to discriminate anadromous populations. A first group of measurements was informative about trophic ecology while remaining unaffected by belly distension. These included fork length (L_F), head length (L_H), head depth (D_H), head depth after the operculum (D_{HO}), upper jaw length (L_J), snout length (L_{Sn}), eye area (A_E), upper gill raker number (N_{UGR}), lower gill raker number (N_{LGR}), upper gill arch length (L_{UG}), lower gill arch length (L_{LG}), length of each of the three longest gill rakers (L_{GR1}, L_{GR2} and L_{GR3}) and the composite measurement of the gill raker sieve mesh (S). To increase the capacity to discriminate populations, four body measurements were added: M_{1_2} (distance between snout and dorsal fin), M_{3_4} (distance between dorsal fin and caudal fork), M_{5_6} (length of anal fin base) and M_{7_1} (distance between pectoral fin and snout).

Since the goal was to assess population admixture through the growing season, sex could not be considered as a variable since sexual dimorphism was not present for a large part of the year. The morphological differentiation was used to: 1) discriminate populations among years, 2) quantify the magnitude and constancy of morphological differentiation and 3) create a mixed-stock procedure allowing individual identification of fish collected in the estuary.

POPULATION DISCRETENESS AMONG YEARS

To quantify morphological discreteness, the confounding effect of size was controlled using principal component analysis (PCA) and a discriminant analysis (DA) was used to discriminate populations. Among the various methods available to morphologically discriminate populations (Cadrin, 2000), the PCA and DA was retained to analyse the rainbow smelt data base (Lecomte & Dodson, 2004). For each fish, all morphometric measurements were combined to calculate a size variable (factor 1 in the factorial PCA). Individual observations for each variable were then plotted against the size variable. Residuals of the regression were used in the DA. The DA was performed to evaluate the morphological discreteness of the different groupings (either shore or spawning sites). Selection of the most informative variables to use in the analysis was made with a stepwise DA technique. At each step, the most informative variable to delineate groups was selected. This was repeated until reaching a minimum threshold ($\alpha = 0.05$) under which no more variables were included in the discriminant function, thereby eliminating redundant variables. The canonical DA analysis was built upon variables selected by the stepwise DA (Lecomte & Dodson, 2004).

To assess morphological differentiation and to obtain error estimates, scores obtained from the discriminant functions were calculated and probabilities were evaluated for their association to each group. Each individual was grouped according to the highest probability obtained. If *a priori* groupings reflected weak morphological differentiation, large numbers of individuals would have been expected to be misclassified. Error was estimated as the percentage of fish attributed to the incorrect group of origin (raw estimate) and the *a posteriori* unstratified error estimate (equal priors), a corrected error estimate that considered unequal sample sizes.

MAGNITUDE AND CONSISTENCY OF MORPHOLOGICAL DIFFERENTIATION

Magnitude of the differentiation between spawning populations was assessed by univariate comparisons using size-corrected measurements [$\log_{10}(x\,L_F^{-1})$; Reist, 1985] on variables associated with feeding ecology (L_H, D_H, D_{HO}, A_E, L_J, L_{UG}, L_{LG}, L_{GR1} and S). The N_{UGR} and N_{LGR} were not corrected for size, as they did not vary with size. A raw estimate of the magnitude of morphological differentiation (Z) was given by calculating the per cent difference of mean population values considering the formula: $Z = \overline{x}_{NSP}\,\overline{x}_{SSP}^{-1} - 1$, where, $\overline{x}_{NSP}$ and $\overline{x}_{SSP}$ are the size-corrected mean population values for the variable of interest. It represents the proportion of difference considering the NSP as the reference population. Positive values are associated to variables for which the NSP exhibit relatively larger character dimensions. Significance of the differences recorded between populations was assessed by a t-test corrected for multiple comparisons (sequential Bonferoni corrections; Holm, 1975; Rice, 1989). Secondly, differences observed between samples (*i.e.* year × sites) were investigated by *Scheffé's post hoc* tests.

MIXED-STOCK AND INDIVIDUAL IDENTIFICATION

Mixed-stock analysis using PCA and DA was performed using the discriminant function obtained when contrasting NSP *v.* SSP spawners. The size factor (factor 1) and then the corresponding probability of belonging to each population were calculated for each individual to be reclassified. Individual fish were then assigned to either spawning populations according to the maximal probability. Error associated to the PCA and DA mixed-stock analysis are given by the *a posteriori* error estimate obtained when comparing spawning population (*i.e.* pure reference populations). In addition to measuring the extent of population mixing throughout the middle estuary, identification at the individual level was used to contrast the diet of the two populations *in situ*.

POPULATION DISTRIBUTION AND INDIVIDUAL IDENTIFICATION

The distribution of adults of the sympatric populations in the estuary during the 1998 growth season was investigated by sampling rainbow smelt along both shores of the middle estuary (Fig. 1). Samples were collected on 16 April (just one site was accessible during spring flood), 5–12 May (only north shore sites were surveyed as the SSP was spawning in their south shore tributaries at this time), 2–13 June, 7–15 August and 22–28 September. Four of these samples were analysed by Lecomte & Dodson (2004). Samples were provided either by commercial fishermen, sport fishermen or by gillnetting (four experimental nets, two mesh sizes: 19 and 32 mm). Although no standardized captures per unit effort (CPUE) could be calculated as too many types of gear were used, an important variation in fishing success was noted. For example, some commercial fishermen caught only 12 fish during a 3 day period while individual sport fisherman could capture >50 rainbow smelt within 1 h. Thus, a qualitative estimate of relative abundance (none present, 0; <25 caught, −; 25–99 caught, +; >100 caught, +++) was determined. All samples were frozen (−20° C) until analysis.

DIET ANALYSIS

As the diet of rainbow smelt changes drastically from the larval and juvenile stage to the adult stage (Vinni *et al.*, 2004), there is a potential for biasing conclusions if diet is not compared among similar age groups. Thus, only adult-sized fish were considered in the present analysis; for the St Lawrence populations, reproduction starts at 2+ years (>100 mm L_F). A multitude of methods exist for gut content analysis (Hyslop, 1980; Cortés, 1997), illustrating the lack of consensus on a standardized method, but also showing that specific questions demand specific analyses. Here, the interest was not to analyse the diet in depth (*e.g.* prey contribution, energetic return and feeding rate) or the

trophic position of rainbow smelt (Lecomte & Dodson, 2004), but rather to assess solely if sympatric populations are partitioned among distinct feeding niches. To achieve this objective, diet was analysed using three complementary methods. First, the raw diet variables of populations were compared as a function of the zone of capture (the ETM v. downstream marine region and the north shore v. the south shore). The 'marine region' where salinity was >25 at high tide (including sites N3 to N5 and S3 to S6; Fig. 1) was considered. Secondly, Schoener's D index of niche overlap (Schoener, 1968) was used to quantify diet differentiation. Several other overlap indices exist and each possesses its own advantages (Hurlbert, 1978; Ricklefs & Lau, 1980; Slobodchikoff & Schultz, 1980; Mueller & Altenberg, 1985; Marshall & Elliott, 1997). Schoener's D index (Schoener, 1968) was selected since it is commonly used and its simplicity facilitates the interpretation of values obtained. This index is also known under several other names (*i.e.* percentage overlap) and it is periodically 'reinvented' (Hurlbert, 1978). As Schoener's D is calculated from pooled data, however, it ignores the sources of variability in diet estimates. An alternative method was thus used that considers both within- and between-group variability to statistically compare dietary overlap ('amended Schoener's, D^a').

GUT CONTENT

Guts were dissected under microscope and every item was identified to the lowest taxonomic level possible. Sixteen taxonomic categories, principally genera, were considered. Because the size distribution of gammarids was fairly large, this group was split into 'large' (*i.e.* >1·5 cm) and 'small' (*i.e.* ≤1·5 cm) categories. Occurrence, dry mass (gut contents were oven dried for 48 h at 60° C) and number were recorded individually. As rainbow smelt consumed a wide variety of prey, from small plankton to adult fishes, caution was taken in assessing the average contribution of a prey item to the population diet using either mass or number. Diet was expressed by averaging individual per cent contribution of each prey item rather than pooling raw data and dividing by the number of stomachs analysed. As such, the maximum contribution of a specific prey category found in an individual stomach to the global sample diet is n^{-1}, where n is the total number of individuals included in the sample. Averaging contributions also eliminates the need to correct for the size of the fish. Gut contents were conserved at −20° C or −80° C prior to their analysis, this method proved sufficient to preserve even small planktonic prey.

RAW ESTIMATES

Strong population-specific selection in terms of type of prey and size spectra could be identified using raw per cent occurrence ($O\%$) and per cent diet content by mass and number ($M\%$ and $N\%$ respectively). Populations were compared according to the major contributor to their diet, either in relation to the prey's biology (*i.e.* zooplankton, macrozooplankton, benthic invertebrates and fishes) or size spectra (*i.e.* limited by gill raker sieve size or by mouth gape size).

SCHOENER'S D NICHE OVERLAP

Schoener's D index of niche overlap (Schoener, 1968) compares pairs of observations, with the index value ranging from 0 (no overlap) to 1 (complete overlap). The value is obtained by summing the differences in percentage of contribution of each category used to compare diet. The following formula was used: $D = 1 - 0·5(\sum_i |p_{xi} - p_{yi}|)$, where, x_i and y_i are the frequency of item i in the diet of group x and y respectively. Arbitrarily, the value of 0·6 is used as a threshold to delineate similar (>0·6) from distinct feeding niches (<0·6; Smith & Zaret, 1982; Wallace & Ramsey, 1983). An index value of 0·6 indicates that 60% of the diet of both groups, expressed in per cent contribution of the various categories, overlaps.

AMENDED SCHOENER'S NICHE OVERLAP

The approach used by Turgeon *et al.* (1999) that considered the variability of Schoener's *D* index calculated between samples was expanded to interpret the overall mean overlap between populations. Using as an example the comparison of the shore of capture (grouping variable = shore), traditional Schoener's *D* pools all samples collected along a shore and thus eliminates intersite variability. Instead, the Schoener's *D* niche overlap was calculated between all pairs of samples that were assigned to a specific group (*i.e.* south shore or north shore). If diet was fairly homogenous within a group (*e.g.* along a shore), it would yield a high mean overlap value. If diet was not homogenous, the mean overlap value would be lowered and variability around the mean would increase. Similarly, niche overlap was calculated between all pairs of samples that came from distinct groups (*e.g.* between shores).

If the grouping variable reflects a real difference that exists in the system (*e.g.* diet is similar within shore but differs between shores), the values calculated within groups will be higher than values calculated between groups. The Mann–Whitney *U*-test was used to compare statistically the values obtained between *v.* within groups.

As samples were used to assess the variability that existed within and between groups, only sites for which sample size was sufficiently large to provide a true representation of the population's diet were considered. In this study power curves built on bootstrap resampling were used to identify those samples (combinations of population-site-date) having numbers of fish sufficient to be considered as representative. To integrate the number of prey categories present and their relative proportion in the diet, Levins' niche breadth (*B*) (Levins, 1968 from Hurlbert, 1978) was used as an estimator of the diet. This index is defined by: $B = \left(n \sum_i p_{xi}^2\right)^{-1}$, where *n* is the total number of categories and p_{xi} is the proportion of the i th category. This index is based on both variables that influence Schoener's *D*, namely the proportion and the number of categories. If all prey categories are found in equivalent proportions, the value will be maximal, indicative of a generalist. The power of this index to accurately portray diet as a function of sample size was modelled by: 1) varying, among individuals, the proportion of the various categories composing the individual diet, and 2) varying among individuals the number of categories consumed (with equal contributions of each category within an individual diet). The shape of the power curves and the s.d. resulting from 1000 bootstrap resamplings were used to determine a threshold beyond which increasing the number of observations did not change the population's diet estimate. A slope of 0·5% of the power curves was considered as the threshold where the addition of new individuals did not justify the added labour to improve diet estimates. All combinations of population-site-date were analysed and only those samples that reached the threshold slope of 0·5% were retained for subsequent analyses. Contribution of each item to the diet was calculated using both number and mass.

RESULTS

MORPHOLOGICAL DIFFERENTIATION

The two sympatric populations were easily distinguished morphologically and population-specific characteristics were maintained over the 3 years sampled (Figs 2 and 3). The PCA and DA analysis contrasting NSP and SSP for pooled samples (*n* = 100 for 1997, *n* = 300 for 1998 and *n* = 300 for 1999) revealed that 95·0% of all individuals were successfully reclassified to their population of origin, with an *a posteriori* error estimate of 4·1% (when considering the unequal number of observations for each population; NSP = 180 and SSP = 520). For this analysis, the discriminant function used nine morpho-metric measurements (L_F, L_J, L_{LGA}, L_{Sn}, L_H, L_{GR1}, M_{7_1}, L_{UGA} and D_{HO}) and one meristic count (N_{UGR}). When considering years independently, the *a*
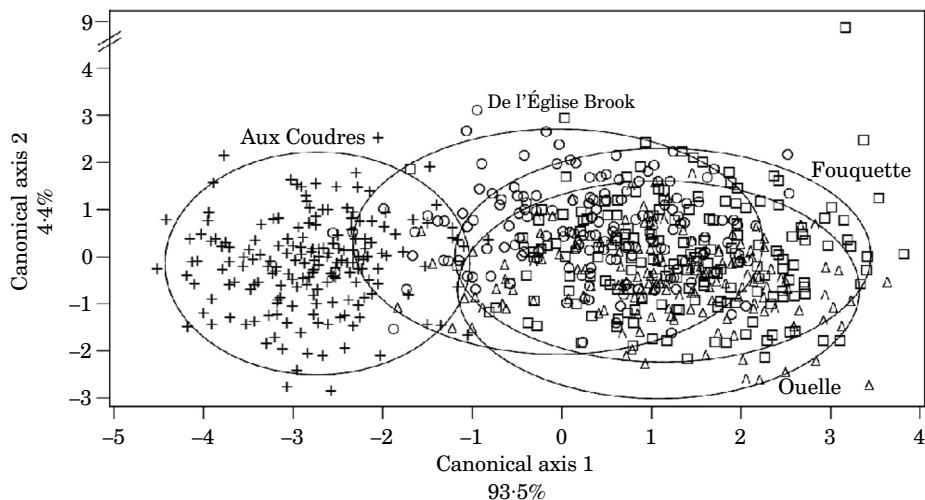
FIG. 2. Graphical representation of PCA and DA analysis contrasting spawning sites. Total $n = 700$ [Aux Coudres (+) $n = 180$; De l'Église (○) $n = 180$; Ouelle (△) $n = 150$; Fouquette (□) $n = 190$]. Per cent variation explained by canonical axis 1 and 2 are indicated.

posteriori error estimates remained similar; 1.7% for 1997 samples ($n = 100$), 3.3% for 1998 ($n = 300$) and 4.0% for 1999 ($n = 300$).

The PCA and DA contrasting sampling sites showed that the magnitude of differentiation between NSP (Aux Coudres Island) v. SSP spawning sites (De L'Église Brook, Ouelle River and Fouquette River) was important but also identified considerable overlap within SSP sites (Fig. 2). The first canonical axis separated Aux Coudres Island (NSP) from the three spawning sites belonging to the SSP sites and explained 93.5% of the variation. The second and third axes accounted for an additional 4.4 and 2.1% respectively.

MORPHOLOGICAL CHARACTERISTICS

The magnitude of morphological differentiation and the consistency between years of differences observed reinforced the idea that differentiation involved selective pressures associated with the exploitation of distinct resources (Lecomte & Dodson, 2004). The NSP exhibited features common to pelagic morphotypes such as a more profiled body (the L_H was 11.7% longer than the L_H of SSP fish, for a similar size of fish), a bigger A_E (on average $+30.8\%$ bigger), and a longer L_{Sn} (on average $+11.6\%$ longer) whereas the SSP shared opposite features typical of a benthic morphotype. The apparent benthic and pelagic dichotomy was not paralleled by benthivorous and planktivorous adaptations or differential growth patterns (Fig. 3). The NSP possessed a larger mouth (*i.e.* L_j was $+16.6\%$ longer), longer L_{GR1} ($+12.4\%$), fewer gill raker counts (-0.9% and -1.8% for N_{UGR} and N_{LGR}) and less dense S ($+13.5\%$). Since the magnitude of morphological differentiation observed between NSP and SSP was maintained over the 3 years surveyed (Fig. 3), the results reported by Lecomte & Dodson (2004) were not caused by error related to small sample sizes. Although NSP individuals were smaller on average (*i.e.* L_F at-age;
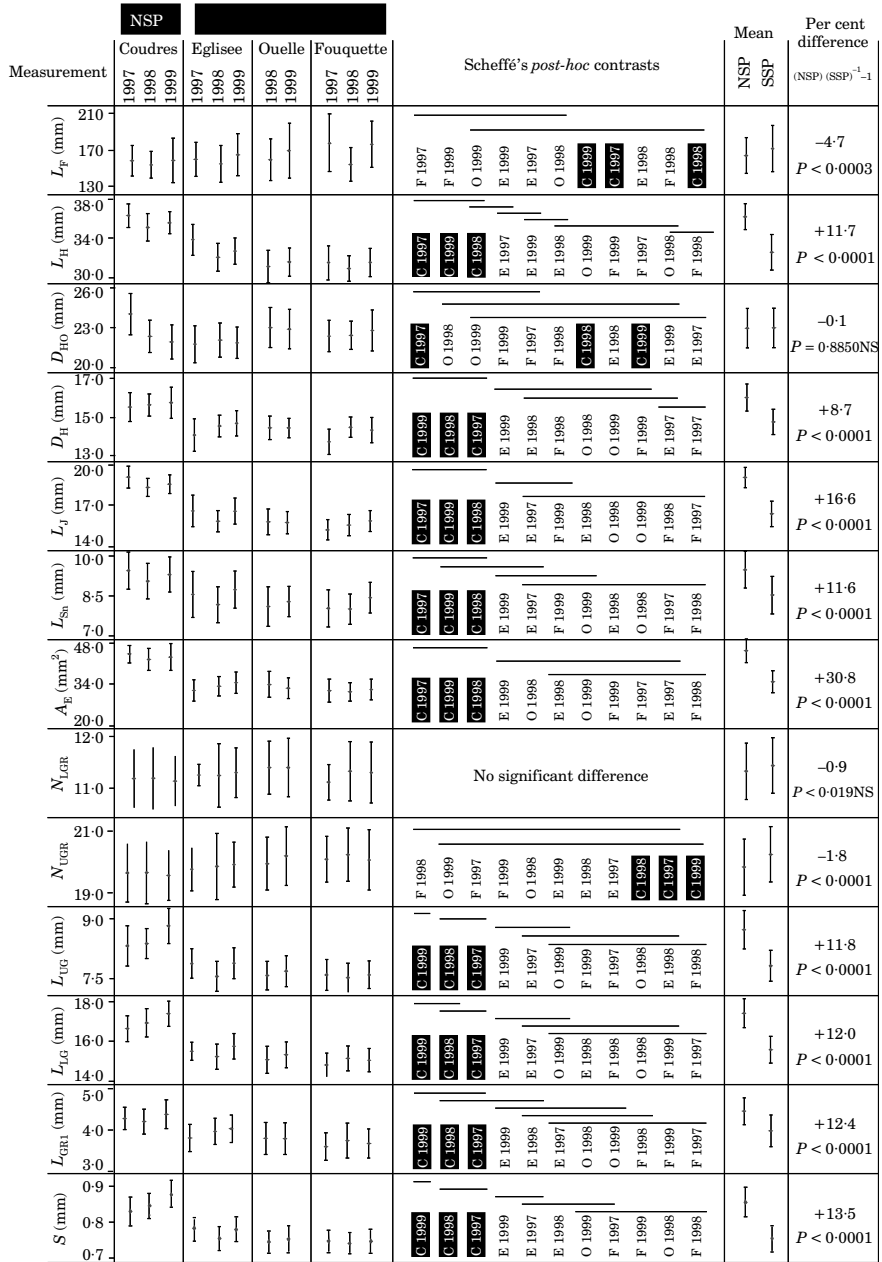
FIG. 3. Size-corrected (to 160 mm fork length, L_F) morphological measurements (L_H, head length; D_{HO}, head depth after the operculum, D_H, head depth; L_J, upper jaw length; L_{Sn}, snout length; A_E, eye area; N_{LGR}, upper gill raker number; N_{UGR}, lower gill raker number; L_{UG}, upper gill arch length; L_{LG}, lower gill arch length; L_{GR1}, longest gill raker; S, gill raker sieve mesh) of rainbow smelt (except fork length, L_F). Mean ± S.D. are indicated for each sample (site + year) and for population (NSP, north shore population; SSP, south shore population). Statistical comparisons are calculated using $\log_{10}$ size-corrected values (see text). Scheffé's *post hoc* contrasts were calculated by considering each sample (C, Aux Coudres; E, De l'Église; O, Ouelle; F, Fouquette). Means, per cent differences of population means and *t*-test were performed on pooled population. NS, not significant. Shading indicates values assigned to NSP.

unpubl.data), the size distribution of the two populations almost completely overlaped (Fig. 3). As such, differences observed after correcting for size were not an artefact associated with the comparison of distinct populations exhibiting different growth patterns (*i.e.* smaller and younger fish having a proportionally larger head).

The *post hoc* Scheffé's contrasts showed that for all size-corrected measurements (*i.e.* excluding L_F, N_{UGR} and N_{LGR}) for which the magnitude of differentiation was significant between populations, the three annual samples of the NSP clustered and exhibited either the highest or lowest values (Fig. 3). In the three cases where overlap occurred between Aux Coudres Island (NSP) and another sampling site, it always involved de l'Église Brook, the most upstream spawning site of the SSP and closest to the NSP spawning grounds. Significant differences between years were observed within populations (Fig. 3) suggesting that phenotypic plasticity was involved. This variability, however, was minor relative to the amplitude of the morphological differentiation recorded between populations and did not influence conclusions concerning the annual stability of population discrimination.

ADULT DISTRIBUTION OF THE TWO SYMPATRIC POPULATIONS

Rainbow smelt populations were generally segregated along the two shores of the estuary; NSP adults were primarily caught along the north shore while SSP adults were caught along the south shore (Fig. 4). Important mixing occurred early in the season along the north shore (May) where pre and post spawning SSP adults were caught with NSP adults. For the remainder of the season,
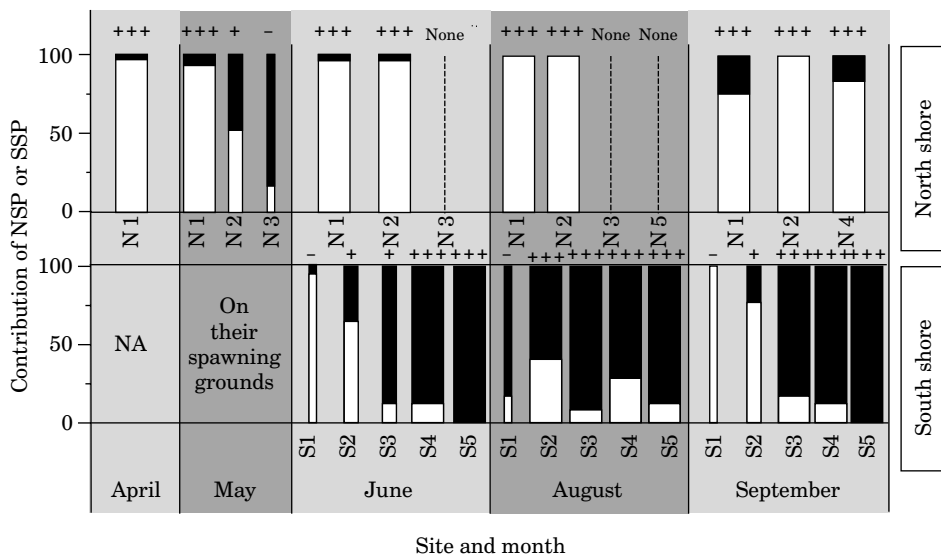

FIG. 4. Distribution of the two populations [NSP, north shore population (□); SSP, south shore population (■)] during the 1998 growth season. For site codes see Fig. 1 (S0 and N0 are omitted as no adults were ever captured). Relative abundance is indicated (+++, >100; +, 25–99; −, <25; none). NA, not available. Dashed vertical lines, no fish were captured.

mixing occurred mainly within the upstream portion of the middle estuary (roughly corresponding to the estuarine turbidity maximum). When these results were analysed as a function of relative abundance, it was clear that mixing occurred only in samples where abundance was low (Fig. 5). Considering only samples of high smelt abundance (*i.e.* +++), the mean ± S.D. percentage population mixing was $10.1 \pm 11.5\%$ on average ($n = 16$, error $= 4\%$) whereas samples characterized by low smelt abundance (*i.e.* − and +) exhibited a higher percentage of mixing between populations, $50.6 \pm 37.9\%$ on average ($n = 10$, error $= 4\%$). The mixing estimated within sites of high abundance could have been artificially inflated by the error associated with the mixed-stock analysis (*e.g.* 4%), however this could not explain the magnitude of mixing observed in samples of lower rainbow smelt abundance.

GUT CONTENTS

A total of 597 fish were dissected of which 72.5% (433), contained prey in their guts (Table I). The percentage of individuals with gut content was fairly stable among regions except for the north shore samples collected in the 'marine region' (*i.e.* downstream of the ETM) where more smelt, independent of the population, had empty stomachs. The L_F of adult rainbow smelt from NSP and SSP was similar along the same shore within regions except along the north shore within the marine section where SSP were smaller on average (Table I; *t*-test, d.f. = 141, $P = 0.016$). The average total dry mass of gut content of individuals caught along the shores of the ETM or the marine region revealed that SSP had on average more gut content than NSP smelt except along the north shore within the ETM (Table I). Although this was suggestive of reduced foraging efficiency of SSP smelt in NSP habitat, the non-parametric tests required to compare these differences could not be performed because sample sizes were too small; a great overlap existed between populations at low gut-content mass (*i.e.* the gut-content mass
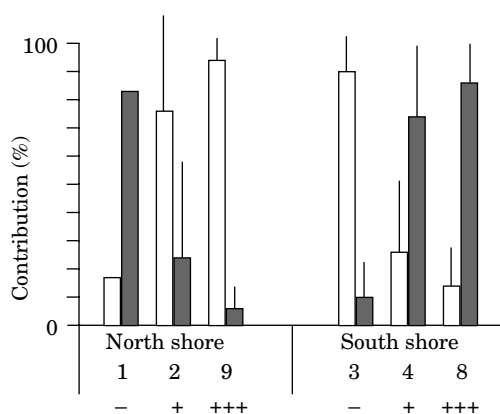

FIG. 5. Population mixing within the samples collected along both shores are compared according to their relative CPUE (−, <25 fish caught; +, 25–99 caught; +++ >100 caught). The contribution + S.D. is expressed in percentage of the two populations (□, north shore population; ■, south shore population) and numbers are the number of sites within each category.

distribution was not normal, the majority of adults exhibiting low gut-content mass) and gut content analyses exhibited high variance.

Both populations targeted large and medium prey whereas small zooplankton species were virtually absent from the diet (Table II), as previously reported by Lecomte & Dodson (2004) based on late-summer and autumn observations (*i.e.* August and October). Both populations consumed prey of various categories such as fishes, benthic invertebrates (*e.g.* nereids), large planktonic invertebrates (*e.g. Crangon*, large *Gammarus* and *Mysis*) and medium-sized (*i.e.* 0·5 to 1·5 mm) planktonic invertebrates (*e.g.* small *Gammarus* and *Neomysis*). No population, however, concentrated exclusively on one of these categories. Several species found in gut contents are normally considered as benthic (*e.g. Crangon* and *Gammarus*) but because of tidal currents in the middle estuary, they are often found throughout the water column (Dodson *et al.*, 1989). Both populations consumed fish prey, but they were more successful in capturing fishes along the south shore shoals whereas few fishes were consumed in northern sampling sites. Fishes occurred in 30% of the gut content of SSP rainbow smelt collected in the ETM along the north shore (Table II). Only 10 SSP adults with gut content, however, were captured there and all three SSP individuals who consumed fishes were found in the same sample.

SELECTION OF REPRESENTATIVE SAMPLES

Modelling the impact of variables on niche breadth estimates revealed that individual variation in the percentage of contribution of prey categories (Fig. 6) was more rapidly buffered than individual variation in the number of prey categories included in the diet (Fig. 6). With only 10 individuals (Fig. 6), the niche breadth index estimated the real population value with little bias. On the other hand (Fig. 6), the variation of the number of categories included in the population diet severely underestimated niche breadth when small numbers of fish were included. The threshold slope of 0·5% (indicated by an arrow on curves of Fig. 6) generally indicated that the curves reached a plateau. Despite such a minor slope, in one case the real niche breadth (calculated from an infinite number of observations) was underestimated by 27% (Fig. 6). This case, however, represented an ecologically improbable extreme; individual fish possessed gut content composed uniquely of one prey category, but the exact category consumed varied between fish. For St Lawrence rainbow smelt, the mean ± S.D. number of prey categories found in samples with >10 individuals was $5·8 ± 1·2$ ($n = 20$), while individuals consumed on $1·7 ± 0·9$ ($n = 433$) categories each. When considering such *in situ* observations, the threshold slope of 0·5% used to select representative samples underestimated by <10% the niche breadth.

For the following analysis, only combinations of population, site and date for which bootstrap resampling yielded niche breadth estimates that reached the minimal 0·5% slope were kept. Examples of the curves obtained for four samples are illustrated in Fig. 6. A total of 26 samples were kept for further analyses. The inter-individual variability within samples was surprisingly low; on average ± S.D. only $6·2 ± 2·3$ or $5·4 ± 2·3$ individuals were sufficient to estimate satisfactorily the niche breadth (*i.e.* slope $< 0·5\%$) considering either mass or number, respectively. Finally, the bias caused by incorrect morphological

TABLE II. Percentage of occurrence O%, percentage contribution to the diet estimated by number (N%) and mass (M%) as a function of the zone of capture (region + shore) and population (north shore population, NSP; south shore population, SSP; identified by morphology) for each prey category considered. Number of individual with gut content (GC) is shown. Shaded areas, occurrences >10%

	Region	Shore	Population	Number with GC	Fishes	Nereis spp.	Crangon septemspinosa	Themisto libellula	Megaciphanes norvegica	Gammarus spp. ≥1·5 cm stenolepis	Mysis <1·5 cm spp.	Gammarus americana	Neomysis americana	Insects	Snails	Cladocera	Calanus spp.	Eurythemora affinis	Haparticoids copepods	Fish eggs
O%	ETM	North	NSP	85	2·4	30·6	16·5	1·2	1·2	25·9	54·1	65·9	28·2	0	0	0	0	0	0	1·2
	ETM	North	SSP	10	30·0	10·0	20·0	0	0	10·0	50·0	70·0	50·0	0	0	0	0	0	0	0
	ETM	South	NSP	58	20·7	12·1	15·5	0	0	8·6	34·5	29·3	10·3	1·7	0	6·9	0	12·1	0	0
	ETM	South	SSP	41	14·6	34·1	22·0	0	0	14·6	7·3	31·7	4·9	0	2·4	0	0	17·1	0	0
	Marine	North	NSP	45	0	11·1	6·7	4·4	8·9	26·7	4·4	60·0	0	0	0	0	0	0	11·1	2·2
	Marine	North	SSP	14	0	0	7·1	0	0	21·4	21·4	92·9	0	7·1	0	0	0	0	0	0
	Marine	South	NSP	19	10·5	36·8	21·1	0	0	21·1	5·3	52·6	0	0	0	0	0	0	0	5·3
	Marine	South	SSP	161	19·3	46·6	21·7	1·2	0·6	22·4	3·1	32·9	1·2	0	1·2	0	8·1	0	0	0·6
N%	ETM	North	NSP	85	1·8	6·8	3·8	0	0	6·0	40·7	23·7	16·1	0	0	0	0	0	0	1·1
	ETM	North	SSP	10	15·7	0·9	0·7	0	0	3·6	20·7	31·0	27·3	0	0	0	0	0	0	0
	ETM	South	NSP	58	10·2	2·9	11·4	0	0	4·0	25·6	20·3	6·0	0·9	0	6·9	0	12·0	0	0
	ETM	South	SSP	41	13·0	15·1	14·6	0	0	8·2	5·7	23·3	3·3	0	0·2	0	0	16·7	0	0
	Marine	North	NSP	45	0	7·1	5·6	2·4	6·7	10·5	3·3	51·2	0	0	0	0	0	0	11·0	2·1
	Marine	North	SSP	14	0	0	3·6	0	0	2·3	7·3	86·8	0	0·1	0	0	0	0	0	0
	Marine	South	NSP	19	7·9	32·0	6·5	0	0	11·2	1·3	35·9	0	0	0	0	0	0	0	5·3
	Marine	South	SSP	161	15·2	28·3	15·3	0·3	0·2	12·0	1·9	20·9	0·8	0	0·8	0	3·8	0	0	0·6
M%	ETM	North	NSP	85	27·7	1·4	9·6	0	0	11·5	34·8	17·0	7·4	0	0	0	0	0	0	0·5
	ETM	North	SSP	10	15·5	7·9	7·2	0	0	5·2	16·4	14·6	4·0	0	0	0	0	0	0	0
	ETM	South	NSP	58	12·9	19·4	13·1	0	0	4·5	25·5	20·3	3·3	1·5	0	5·3	0	8·0	0	0
	ETM	South	SSP	41	0	4·8	19·2	0	0	8·2	5·7	20·3	0	0	0·5	0	0	10·4	0	0
	Marine	North	NSP	45	0	0	6·6	2·6	8·3	14·8	4·3	49·1	0	0	0	0	0	0	9·0	0·5
	Marine	North	SSP	14	0	0	7·0	0	0	9·5	10·8	72·6	0	0	0	0	0	0	0	0
	Marine	South	NSP	19	10·2	29·0	13·4	0	0	15·9	0·2	26·0	0	0	0	0	0	0	0	5·3
	Marine	South	SSP	161	18·0	31·2	16·5	0·7	0·5	14·0	1·8	15·3	0·7	0	0·7	0	0·6	0	0	0

identification (*e.g.* 4% misclassification error) in a sample of 25 fish implied, on average, the misclassification of only one fish.

AMENDED SCHOENER'S D^a

The comparison of the magnitude of niche overlap showed that diet of middle estuary rainbow smelt differed according to the area where samples were collected but no differences existed between populations (Table III). The
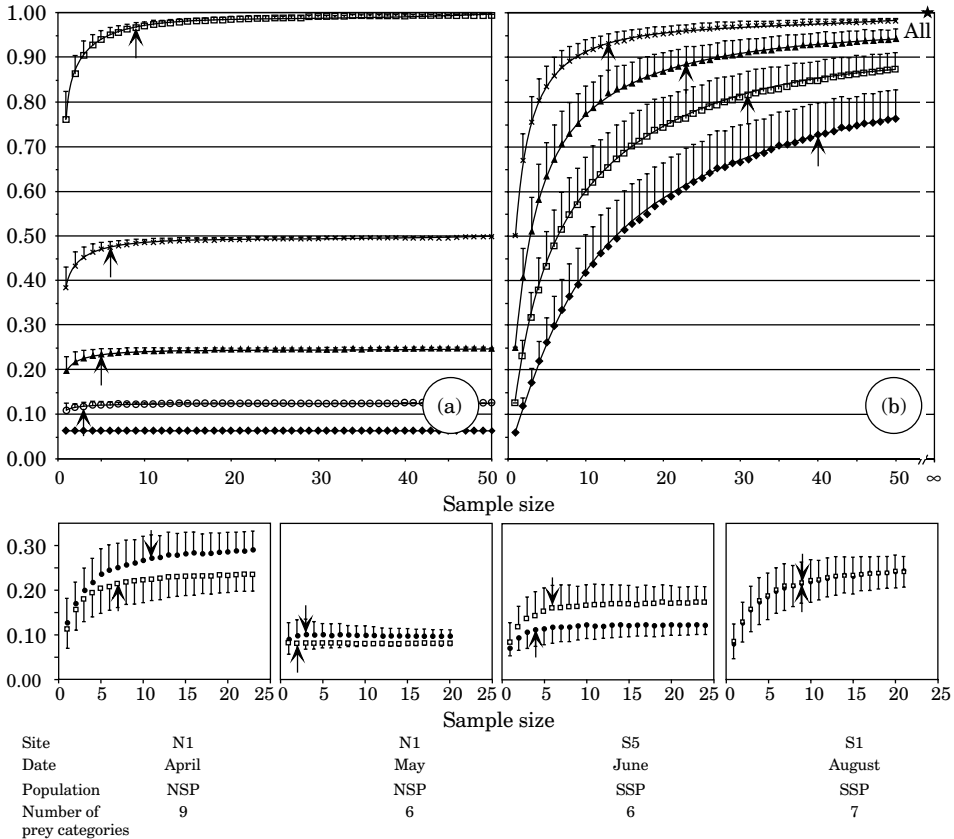

Site	N1	N1	S5	S1
Date	April	May	June	August
Population	NSP	NSP	SSP	SSP
Number of prey categories	9	6	6	7

FIG. 6. Impact of (a) number of categories and (b) the per cent contribution of categories on Levins' niche breadth. Each point represents 1000 bootstrap resamplings for various sample sizes and their corresponding S.D. (★, values for infinite sample size). ⬆, slopes reach the threshold of 0·5%. (a) Per cent contribution of individual categories varied within a fixed number of categories (*i.e.* for ▲ = 4, each individual fish consumed prey from the same four categories, but per cent contribution of each prey category differed among individuals); number ranged from one to 16 categories (◆, 1; ○, 2; ▲, 4; ×, 8; □, 16). (b) Number of prey categories consumed varied from one to 16 among a total of 16; per cent contribution of each category was fixed (*i.e.* for ▲ = 4, each individual consumed an equal proportion of four prey categories, but the four categories varied among the 16 potential prey categories from individual to individual); number of categories ranged from one to eight from a total of 16 categories (◆, 1: each category contributed 100%; □, 2: each category contributed 50%; ▲, 4: each category contributed 25%; ×, 8: each category contributed 12·5%). *In situ* observations ± S.D. are presented in the four lower panels for mass (●) and number (□) (see Fig. 1 for site). (NSP, north shore population; SSP, south shore population).

TABLE III. Amended Schoener's (D^a) comparing diet overlap between populations [north shore populations (NSP) v. south shore populations (SNP)] shores (north v. south) and regions [estuarine turbidity maximum (ETM) v. marine section]. Populations were also nested with shore and region. Sample size of paired groups are compared. D^a contrasts, the number of comparisons calculated within (intra) and between (inter) groups. The values of D^a reported are the mean values obtained from the n D^a contrasts, P-values compared intra v. inter. Comparisons are based on mass and numbers of prey categories. A typical example of a confounded comparison leading to the erroneous conclusion that populations were exploiting distinct niches is shown

Comparison	n Sample size (Pair)	n D^a contrasts Intra:inter	Schoener's D^a - Mass			Schoener's D^a - Number		
			Intra	Inter	P	Intra	Inter	P
Population	14:12 NSP:SSP	157 : 168	0·375	0·349	0·245	0·355	0·318	0·162
Shore	10:16 North:south	156:169	0·383	0·339	**0·029***	0·348	0·323	0·263
Region	13:13 ETM:marine	165:160	0·385	0·339	**0·032***	0·336	0·299	**<0·001***
Population by shore								
Within north shore	8:2 NSP:SSP	16:29	0·411	0·381	0·514	0·381	0·374	0·915
Within south shore	6:10 NSP:SSP	60:60	0·404	0·348	0·122	0·362	0·352	0·282
Population by region								
Within ETM	10:3 NSP:SSP	48:30	0·351	0·320	0·528	0·343	0·286	0·295
Within marine	4:9 NSP:SSP	42:36	0·439	0·423	0·787	0·427	0·432	0·700
Population by region and shore								
Within ETM and north shore	5:1 NSP:SSP	10:5	0·413	0·271	0·142	0·370	0·375	0·807
Within ETM and south shore	5:2 NSP:SSP	11:10	0·279	0·328	0·481	0·283	0·282	0·833

Within marine and north shore	3:1 NSP:SSP	3:3	0·555	0·633	0·513	0·520	0·547	0·827
Within marine and south shore	1:8 NSP:SSP	28:8	0·454	0·522	0·323	0·451	0·495	0·447
Confounded conclusion (considering only sites where each population was the most abundant)								
NSP in ETM and north shore v. SSP in marine south shore	5:8 NSP:SSP	38:40	0·443	0·353	**0·030***	0·430	0·296	**0·002***

*, Intra values are significantly higher that inter values.

average niche overlap measured within groups was very low, indicating an important variability in the diet of rainbow smelt. In only two contrasts out of 24 did the within-group overlap exceed values of 0·5. Thus, despite the homogeneity observed between individuals within samples, there was an important variability between samples. The shore and the region of capture significantly affected the mean overlap. For all significant cases reported in Table III, the within group variability was smaller than the between group variability indicating that the factor contrasted (*e.g.* shore and region) affected the diet. Although the Mann-Whitney *U*-tests were significant, within group niche overlap values were well below the value of 0·6 usually considered as indicative of separate niches (*i.e.* the same population exploits distinct niches depending on the site of capture). When considering globally the effect of population, shore or region of capture, population identity was not the principal factor contributing to diet differentiation. There were no significant differences in the mean dietary niche overlap measured between populations and within populations. If only sites where the two populations were the most abundant (*i.e.* maximum catch per unit effort) had been considered, a significantly smaller diet overlap between populations than within populations (Table III) would have been concluded. The expanded analysis considering the various sources of variation, however, revealed that such a conclusion is confounded by variability associated with the shore and region of capture (Table III).

DISCUSSION

In a previous study that focused on various aspects of the biology of sympatric rainbow smelt (Lecomte & Dodson, 2004), adult fish were strongly spatially segregated among ecologically-distinct habitats but foraged on similar prey items. As a result of a limited number of samples and the spatio-temporal coverage, several sources of error or some systematic bias could not be ruled out *a priori*. In the present study previous results were confirmed and it was demonstrated further that: 1) spatial segregation was maintained throughout the middle estuary whereas some population mixing occurred at low abundance, 2) populations exhibited similar diets typical of opportunistic fish that targeted large prey, 3) the diet of rainbow smelt, independent of the populations considered, reflected the heterogeneous distribution of prey taxa (large variation between samples) and possibly the dynamics of schooling behaviour of rainbow smelt (little variation within samples) and finally 4) the morphology and degree of differentiation between morphotypes was stable over 3 years and thus not the fortuitous result of sampling error.

The spatial segregation of the two populations was observed to be a function of local rainbow smelt abundance. When local abundance was high, NSP individuals dominated the north shore samples whereas the SSP individuals dominated south shore samples. At low abundance, per cent population mixing was highly variable (Fig. 5). Such differences may be associated with schooling behaviour. When schools of rainbow smelt are present, local abundance is high resulting in large catches. Alternatively, when local abundance is low captures are likely to be based on solitary individuals or at least on very small aggregations. Thus, when considering schools of rainbow smelt (*i.e.* high

abundance), the populations appear to be effectively segregated in ecologically distinct habitats. The two populations, however, were mixed throughout the estuary at low abundance.

Rainbow smelt were difficult to obtain along the north shore; the Aux Coudres Island (N1 on Fig. 1) is the only location presently sustaining a locally important fishery along that shore (S. Georges, unpubl. data). In contrast, rainbow smelt were fairly common along the south shore and abundance increased downstream of the ETM. Considering the abundance and population composition of the samples (Fig. 4), there is a possibility that populations were also spatially segregated between the ETM and the downstream marine section of the middle estuary. Globally, the NSP was only abundant along the north shore, mainly within the ETM whereas the SSP was abundant along the south shore primarily downstream of the ETM. The near complete segregation observed by Lecomte & Dodson (2004) is thus explained by the fact that samples used in their study came from sites where rainbow smelt were present in high abundance.

The results of the present analysis greatly clarified the perception of the nature of trophic and habitat partitioning between the two sympatric populations. Both adult populations consumed a wide variety of predominantly large and medium-sized prey (*i.e.* >0·5 cm). Small planktonic prey were not important in the diet at any time of the year as revealed by their low contribution in terms of mass. There was important within-population variation in diet composition but little within-sample variability. Longitudinal differences in zooplankton community structure influenced rainbow smelt diets. Finally, at the local scale, both populations ate the same prey items in similar proportions.

The diet of St Lawrence middle estuary rainbow smelt appears to be typical of an opportunistic species that maximizes its energetic return through the exploitation of the largest prey available locally. Rainbow smelt are capable of feeding upon a very wide spectrum of prey as illustrated by their morphology (*e.g.* mouth gape size and gill raker sieve mesh; Fig. 3) and the *in situ* observation of the prey categories found in gut contents (Table II). The high abundance of potential prey in the system (Dodson *et al.*, 1989; Laprise & Dodson, 1994) may explain the lack of dietary differentiation between populations and the relatively high occurrence of large prey. The prey abundance could also explain the elevated proportion of fish with gut content (72·5 % on average; Table I) and the high mean dry mass of gut contents (116·7 mg dry mass on average; Table I).

Superimposing opportunistic feeding on a heterogeneous prey field will invariably yield significant differences between samples collected in distinct areas. This contributes to the differences in diet exhibited by rainbow smelt collected in the ETM *v.* the marine section (Tables II and III). Similarly, opportunistic feeding translates into low dietary niche overlap (*i.e.* distinct niche) even for within-population comparisons (Table III). Comparing only sites where each population was found at their greatest abundance will result in highly biased conclusions as several sources of variation are confounded. Population identity is not the most important factor in explaining variation in diet (see Table III). For similar studies involving a heterogeneous prey field, all sources of variation must be considered and compared.

In contrast to the large number of prey categories consumed globally by rainbow smelt, Levin's niche breadth estimates coupled with a bootstrap approach revealed an impressive within sample homogeneity in the diet. Individuals fed quite homogenously on a few prey categories within a site at a specific date, but the diet was highly diversified and varied enormously according to the site and date of capture. The shoaling behaviour of rainbow smelt may explain such an observation. The 'forage-area copying' concept (Pitcher & House, 1987; Hughes, 1997) proposes that members of a shoal will start feeding once seeing some members foraging. Thus, once locating an abundant prey aggregation, it is conceivable that all individuals of a school of smelt will start feeding and will consequently have similar gut contents.

Incorporating sources of variance to compare diet overlap allowed the significance of the calculated niche overlap indices to be calculated. The approach proposed is thus an improvement compared to the traditional Schoener's D index and its reliance on an arbitrary fixed value (*i.e.* 0·6; Smith & Zaret, 1982; Wallace & Ramsay, 1983) to delimit distinct *v.* similar diet niches. As illustrated in the present study (see Table III), the amended Schoener's (D^a) allowed the sources of variability underlying dietary variation to be identified and distinguished. To add statistical validity to the niche overlap, other authors have used a bootstrap and jackknife approach to estimate confidence intervals around the mean overlap values or conducted simulations to estimate error (Smith & Zaret, 1982; Mueller & Altenberg, 1985; Labropoulou & Eleftheriou, 1997). Although it is an improvement over the use of plain overlap indices, such approaches did not yield estimates of intra-group niche overlaps (*e.g.* population, species and area). Schoener's D^a coupled with the bootstrap approach used to select representative samples allowed full use of the sampling protocol to be made and the *in situ* variability in local conditions (shore and region) to be exploited as a means of testing alternative hypotheses. In cases where within-population niche overlap is low (see Table III), the value obtained for between-group niche overlap should be compared with within-group overlap rather than with the threshold value of 0·6. In natural systems where diet is influenced by multiple sources of variation, the use of 0·6 as the threshold separating similar from dissimilar diet may be misleading as the upper value of within-group overlap may be much smaller than 0·6, as in the present case.

The inclusion of three successive years of observations and a total of 700 individuals reinforces the conclusion of Lecomte & Dodson (2004) that populations exhibit morphologies suitable to exploit distinct habitats [pelagic (NSP) *v.* benthic (SSP)] in the absence of consistent morphological features related to specific diets. The magnitude of differentiation is elevated for several measurements, but they are mainly associated with the head morphology (*e.g.* head length, eye area and jaw length). The magnitude of differentiation is also suggestive of the selective pressures involved. In the present study, the most divergent measurements were A_E and L_J that were respectively 30·8 and 16·6% larger in NSP adults (Fig. 3). In contrast, among the seven anadromous populations surveyed by Taylor & Bentzen (1993), the maximal differences recorded between the two most divergent populations were well below the values reported in the present study; 14·9 % for A_E (using 'eye length' to estimate A_E) and only 4·7% for L_J.

Several non-mutually exclusive causes may be involved in the morphological differentiation observed here, including: 1) historical contingency (i.e. secondary contact of two morphologically distinct glacial races; Bernatchez, 1997), 2) varying environmental conditions during ontogeny (Lecomte & Dodson, 2004) and 3) selective pressures relative to foraging in distinct habitats.

The competition for limited food resources is seen as a potent mechanism favouring the divergence and coexistence of sympatric populations. The exploitation of distinct habitats exhibiting divergent ecological characteristics is equally effective in maintaining sympatric populations within an estuarine setting without diet differentiation as demonstrated in the present study. This observation raises the possibility that in other ecosystem settings, spatial segregation may precede diet segregation. The correlation between morphological adaptations and differences in diet generally leads to the conclusion that divergent selection favours the utilization of the underexploited food resource. This is generally represented by planktonic prey in lakes. As a consequence, populations segregate spatially to exploit such prey. An equally parsimonious explanation, however, is that divergent selection favours the utilization of an underexploited habitat where prey are present. In the case of relatively simple systems such as lakes, the morphological adaptations to exploit the small prey dominating the pelagic environment may have appeared after the habitat partitioning. The present study indicates that the latter is possible. Spatial partitioning may very well be the initial response to reduction of competition for any resource.

References

Baby, M.-C., Bernatchez, L. & Dodson, J. J. (1991). Genetic structure and relationships among anadromous and landlocked populations of rainbow smelt, *Osmerus mordax*, Mitchill, as revealed by mtDNA restriction analysis. *Journal of Fish Biology* **39** (Suppl. A), 61–68.

Bacheler, N. M., Neal, J. W. & Noble, R. L. (2004). Diet overlap between native bigmouth sleepers (*Gobiomorus dormitor*) and introduced predatory fishes in a Puerto Rico reservoir. *Ecology of Freshwater Fish* **13**, 111–118.

Bernatchez, L. (1997). Mitochondrial DNA analysis confirms the existence of two glacial races of rainbow smelt (*Osmerus mordax*) and their reproductive isolation in the St-Lawrence R. estuary (Québec, Canada). *Molecular Ecology* **6**, 73–83.

Bernatchez, L. & Martin, S. (1996). Mitochondrial DNA diversity in anadromous rainbow smelt, *Osmerus mordax* Mitchill: a genetic assessment of the member-vagrant hypothesis. *Canadian Journal of Fisheries and Aquatic Sciences* **53**, 424–433.

Cadrin, S. X. (2000). Advances in morphometric identification of fishery stocks. *Reviews in Fish Biology and Fisheries* **10**, 91–112.

Cortés, E. (1997). A critical review of methods of studying fish feeding based on analysis of stomach contents: Application to elasmobranch fishes. *Canadian Journal of Fisheries and Aquatic Sciences* **54,** 726–738.

Dodson, J. J., Dauvin, J.-C., Ingram, G. R. & D'Andlejan, B. (1989). Abundance of larval rainbow smelt (*Osmerus mordax*) in relation to the maximum turbidity zone and associated macroplanktonic fauna of the Middle St. Lawrence Estuary. *Estuaries* **12,** 66–81.

Feinsinger, P., Spears, E. E. & Poole, R. W. (1981). A simple measure of niche breadth. *Ecology* **62,** 27–32.

Holm, S. (1975). Sequential inversion sum tests. *Scandinavian Journal of Statistics* **2,** 1–10.

Hughes, R. N. (1997). Diet selection. In *Behavioural Ecology of Teleost Fishes* (Godin, J.-G., ed.), pp. 134–162. Oxford: Oxford University Press.

Hurlbert, S. H. (1978). The measurement of niche overlap and some relatives. *Ecology* **59,** 67–77.

Hyslop, E. J. (1980). Stomach contents analysis – a review of methods and their application. *Journal of Fish Biology* **17,** 411–429.

Labropoulou, M. & Eleftheriou, A. (1997). The foraging ecology of two pairs of congeneric demersal fish species: importance of morphological characteristics in prey selection. *Journal of Fish Biology* **50,** 324–340.

Laprise, R. & Dodson, J. J. (1994). Environmental variability as a factor controlling spatial patterns in distribution and species diversity of zooplankton in the St. Lawrence Estuary. *Marine Ecology Progress Series* **107,** 67–81.

Lecomte, F. & Dodson, J. J. (2004). Role of early life-history constraints and resource polymorphism in the segregation of sympatric populations of an estuarine fish. *Evolutionary Ecology Research* **6,** 631–658.

Levins, R. (1968). *Evolution in Changing Environments: Some Theoretical Explorations.* Princeton, NJ: Princeton University Press.

Marshall, S. & Elliott, M. (1997). A comparison of univariate and multivariate numerical and graphical techniques for determining inter- and intraspecific feeding relationships in estuarine fish. *Journal of Fish Biology* **51,** 526.

Mueller, L. D. & Altenberg, L. (1985). Statistical inference on measures of niche overlap. *Ecology* **66,** 1204–1210.

Nellbring, S. (1989). The ecology of smelts (genus *Osmerus*): A literature review. *Nordic Journal of Freshwater Research* **65,** 116–145.

Pitcher, T. J. & House, A. C. (1987). Foraging rules for group feeders: area copying depends upon food density in shoaling goldfish. *Ethology* **76,** 161–167.

Reist, J. D. (1985). An empirical evaluation of several univariate methods that adjust variation in morphometric data. *Canadian Journal of Zoology* **63,** 1429–1439.

Rice, W. R. (1989). Analysing tables of statistical tests. *Evolution* **43,** 223–225.

Ricklefs, R. E. & Lau, M. (1980). Bias and dispersion of overlap indices: results of some Monte Carlo simulations. *Ecology* **61,** 1019–1024.

Robinson, B. W. & Schluter, D. (2000). Natural selection and the evolution of adaptive genetic variation in northern freshwater fishes. In *Adaptive Genetic Variation in the Wild* (Mousseau, T. A., Sinervo, B. & Endler, J. A., eds), pp. 65–94. New York: Oxford University Press.

Schluter, D. (1995). Adaptive radiation in sticklebacks: Trade-offs in feeding performance and growth. *Ecology* **76,** 82–90.

Schoener, T. W. (1968). The *Anolis* lizards of Binimi: Resource partitioning in a complex fauna. *Ecology* **49,** 704–726.

Scott, W. B. & Crossman, E. J. (1974). *Poissons d'eau douce du Canada.* Ottawa: Ministère de l'Environnement, Service des pêches et sciences de la mer.

Scott, W. B. & Scott, M. G. (1988). *Atlantic Fishes of Canada.* Ottawa: University of Toronto Press & Minister of Fisheries and Oceans & Canadian Government Publishing Centre.

Skúlason, S. & Smith, T. B. (1995). Resource polymorphisms in vertebrates. *Trends in Ecology & Evolution* **10,** 366–370.

Slobodchikoff, C. N. & Schultz, W. C. (1980). Measures of niche overlap. *Ecology* **61,** 1051–1055.

Smith, E. P. & Zaret, T. M. (1982). Bias in estimating niche overlap. *Ecology* **63,** 1248–1253.

Smith, T. B. & Skúlason, S. (1996). Evolutionary significance of resource polymorphisms in fishes, amphibians, and birds. *Annual Review of Ecology and Systematics* **27,** 111–133.

Taylor, E. B. & Bentzen, P. (1993). Evidence for multiple origins and sympatric divergence of trophic ecotype of smelt (*Osmerus*) in northeastern North America. *Evolution* **47,** 813–832.

Turgeon, J., Estoup, A. & Bernatchez, L. (1999). Species flock in the North American Great Lakes: molecular ecology of lake Nippigon ciscos (Teleostei: Coregonidae: *Coregonus*). *Evolution* **53,** 1857–1871.

Vincent, W. F. & Dodson, J. J. (1999). The St. Lawrence River, Canada-USA: the need for an ecosystem-level understanding of large rivers. *Japanese Journal of Limnology* **60,** 29–50.

Vinni, M., Lappalainen, J., Malinen, T. & Peltonen, H. (2004). Seasonal bottlenecks in diet shifts and growth of smelt in a large eutrophic lake. *Journal of Fish Biology* **64,** 567–579. doi: 10.1046/j.1095-8649.2003.00323.x

Wallace, R. K. & Ramsey, J. S. (1983). Reliability in measuring diet overlap. *Canadian Journal of Fisheries and Aquatic Sciences* **40,** 347–351.