Journal of Fish Biology (2000) **57,** 571–588
doi:10.1006/jfbi.2000.1334, available online at http://www.idealibrary.com on IDEAL®

Seasonal changes of thyroid hormones in field-collected Atlantic cod in relation to condition indices, water temperature and photoperiod

L. A. Comeau*¶, S. E. Campana†, J. M. Hanson‡ and G. A. Chouinard‡

**Department of Biology, Dalhousie University, Halifax, Nova Scotia B3H 4J1, Canada; †Department of Fisheries and Oceans, Marine Fish Division, Bedford Institute of Oceanography, P.O. Box 1006, Dartmouth, Nova Scotia B2Y 4A2, Canada and ‡Department of Fisheries and Oceans, Gulf Fisheries Centre, P.O. Box 5030, Moncton, New Brunswick E1C 9B6, Canada*

(*Received 23 November 1999, Accepted 7 April 2000*)

Serum T_4 and T_3 in wild Atlantic cod *Gadus morhua* ranged from 1 to 12 ng ml^{-1} and from 2 to 27 ng ml^{-1} respectively over a 3-year period. In general, the concentrations increased from summer (T_3) or early autumn (T_4) to maxima in mid-winter and declined abruptly during spring. The T_4/T_3 monthly means were lowest in summer and highest in winter. The seasonal patterns of thyroid hormones were weakly correlated with changes in water temperature. However, both T_4 and T_3 co-varied simultaneously with photoperiod. In addition, T_3 was correlated with the hepatosomatic index and condition factor during summer and autumn. It is suggested that the seasonal changes in the release of T_4 from the thyroid were photoperiod-driven, and that the course of T_3 was regulated by the metabolic state of the fish during the somatic growth period.

Key words: *Gadus morhua*; thyroid hormones; liver; condition; temperature; photoperiod.

INTRODUCTION

Although the teleostean thyroid has been researched quite extensively over the past decades, current understanding of its various functions is based almost entirely on freshwater and diadromous species. In these taxa, thyroid hormones promote growth (Donaldson *et al.*, 1979; Higgs *et al.*, 1982) as in the case of higher vertebrates (Gorbman *et al.*, 1983), induce smolting (Boeuf, 1993), and stimulate early gonadal development (Cyr & Eales, 1996). They may also trigger and sustain upriver and seaward migrations (Godin *et al.*, 1974; Grau *et al.*, 1981; Castonguay *et al.*, 1990; Høgåsen & Prunet, 1997).

In marine teleosts, thyroid hormones accelerate larval development (Brown & Kim, 1995; Tanaka *et al.*, 1995) and initiate metamorphosis in pleuronectiformes (Inui & Miwa, 1985; Inui *et al.*, 1994). However, apart from their role in early ontogeny, little is known about the function of these hormones in marine species, and more precisely about their cyclical patterns in adult marine fishes. The few sporadic reports lead to the conclusion that the seasonal changes in thyroid

¶Author to whom correspondence should be addressed. Tel.: 506 783 8844; fax: 506 783 3509; email: lcomeau@is2.dal.ca

0022–1112/00/090571+18 $35.00/0

secretions differ markedly from one species to another. For instance, the concentrations of plasma L-thyroxine (T_4) increased to a single peak during spring in the winter flounder *Pseudopleuronectes americanus* Walbaum) (Eales & Fletcher, 1982), whereas the plaice *Pleuronectes platessa* L., presented a bimodal series with T_4 peaks occurring in both summer and winter (Osborn & Simpson, 1978). In contrast, the Atlantic cod *Gadus morhua* L. showed no obvious T_4 cycle in the laboratory (Cyr *et al.*, 1998), although its thyroid follicular cell height cycled clearly in the wild (Woodhead, 1959).

The reasons behind the contradictory accounts of thyroidal cycling are unknown. It could be argued that thyroidal changes are species or population dependent, being sensitive to patterns of food intake and diet composition (Mackenzie *et al.*, 1998), to physical attributes of the habitat (Leatherland, 1982; Leloup & De Luze, 1985), and to the timing of reproductive events (Cyr & Eales, 1996). On the other hand, the thyroidal discrepancies may simply reflect the different study conditions (laboratory *v.* field) or the different capture techniques in field investigations (SCUBA *v.* fishing gear). Some of the divergence between the reports might also reflect the use of different criteria (histological index *v.* blood hormones) for the assessment of thyroidal status (Eales & Brown, 1993).

In an attempt to help clarify and understand better thyroid cycling in temperate marine fishes, thyroid hormones were studied in field-collected Atlantic cod. The seasonal changes of circulating levels of T_4 and its extrathyroidally-produced derivative 3,5,3′-triiodo-L-thyronine (T_3) were documented over a 3-year period. An attempt is made to identify the environmental and physiological variable(s) that might regulate thyroidal changes, including food intake (stomach fullness), condition, water temperature and photoperiod. Thyroidal patterns are compared with previous work on cod and other marine teleosts and the functional significance of the thyroid cycles in cod is discussed.

MATERIALS AND METHODS

SAMPLING AREA

Atlantic cod were sampled from the southern Gulf of St Lawrence population on a quasi-monthly schedule between July 1995 and June 1998. The exact sampling area varied according to the seasonal migrations of the cod (Fig. 1). During the summer–autumn period, samples were collected in traditional fishing areas inside the southern Gulf of St Lawrence. During the winter and spring, samples were collected in the Cabot Strait area, where southern Gulf of St Lawrence cod overwinter (Paloheimo & Kohler, 1968; Campana *et al.*, 1999). Most samples were collected as part of scientific surveys or sentinel fishery expeditions. Mobile gear vessels were employed to catch cod, except off the western coast of Cape Breton Island where baited-longlines were used occasionally. A total of 108 sites was visited during 43 trips. The time interval between individual trips ranged from 1 to 2 weeks during the summer and autumn and from 1 to 2 months during the winter and spring.

SAMPLES ATTRIBUTES AND ENVIRONMENTAL DATA

The number of fish collected each month ranged from nine to 177 and both sexes were generally well represented (Table I). Except for the June 1998 sample, the average fork length of cod varied from *c.* 49 to 55 cm. Following the collection of blood (see protocol below), the cod were kept on ice and dissected generally within 48 h; otherwise, they were stored frozen at -20° C and dissected after having been partially thawed. The dissections were carried out to determine sex and to measure recent food intake,

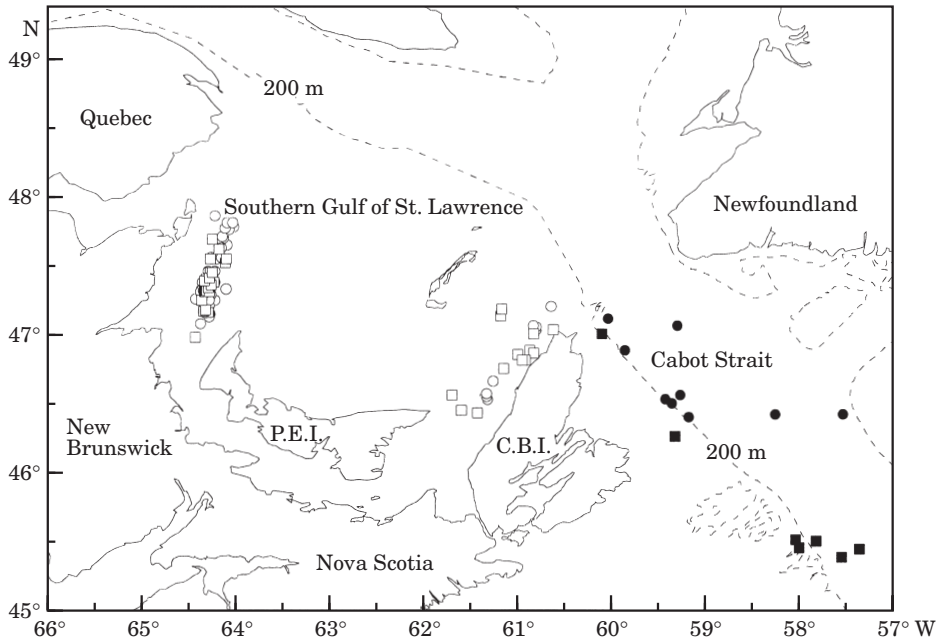

FIG. 1. Map of the southern Gulf of St Lawrence area showing capture locations of cod samples during the 1995–1998 study period. Samples were collected in winter (●), spring (■), summer (○), and autumn (□). The dashed lines show the 200 m depth contour. C.B.I., Cape Breton Island; P.E.I., Prince Edward Island.

hepatosomatic index (I_H), and condition factor. Food intake was reported as the stomach fullness index, representing the weight of the stomach contents as a percentage of the carcass weight (Schwalme & Chouinard, 1999). The I_H and condition factor were calculated using somatic weight as in Lambert & Dutil (1997).

The bottom water temperatures at the time of capture were recorded by securing data loggers (Minilog, Vemco Ltd, Canada) to the fishing gear or by performing CTD casts. The Minilogs had been calibrated at the factory with an estimated accuracy of 0·2° C. In January 1996, bottom temperatures were not recorded for stations where blood was collected. Therefore, the temperatures measured at neighbouring stations (<10 km) were used, having similar depths and visited during the same cruise. Photoperiod data for the 15th of each month were obtained from the U.S. Naval Observatory (Astronomical Applications Department).

THYROID HORMONES

Blood samples were retrieved within *c*. 20 min after the fish were brought on board. To reduce sampling time, the aorta was cut rapidly and blood was allowed to flow directly into 7 ml Vacutainer vials. The vials were capped and refrigerated for 12–24 h. Then the serum fraction was transferred into 2·5 ml cryovial tubes and stored at − 72° C until the thyroid hormones were measured by radioimmunoassay (RIA).

Total (protein bound and protein unbound) T_4 and total T_3 were measured using a combined T_4–T_3 RIA (Omeljianuk *et al*., 1984) and the reagents employed by Cyr *et al*. (1998). Barbital buffer (50 mM sodium barbital, 10 mM barbital, pH 8·6) was used for the T_4 segment of the RIA and phosphate buffer (100 mM Na_2HPO_4, 3 mM Na_2EDTA, pH 7·4) for the T_3 segment. T_4 and T_3 (Sigma Chemicals, St Louis, MO) were diluted serially into 0·1 N NaOH to obtain working standards. ^{125}I-T_4 and ^{125}I-T_3 tracers (New England Nuclear, Boston, MA, specific activity 44–46 MBq μg^{-1}) were diluted in 0·1 N

TABLE I. Sampling dates, average fork lengths and number of fish included in the assessment of thyroid hormones

Year	Month	Length cm		Number of fish		
		Mean	(S.E.)	Male	Female	Total
1995	Jul.	48·9	(1·4)	11	7	18
	Aug.	55·4	(0·6)	6	11	17
	Sep.	52·2	(0·7)	14	18	32
1996	Jan.	51·0	(0·8)	23	19	42
	Feb.					
	Mar.	54·8	(2·3)	3	6	9
	Apr.					
	May	55·0	(1·7)	12	9	21
	Jun.	53·3	(1·0)	26	37	63
	Jul.	51·8	(1·0)	27	33	60
	Aug.	54·7	(0·8)	43	43	86
	Sep.	51·3	(0·6)	36	41	77
	Oct.	54·2	(0·8)	27	43	70
	Nov.	49·1	(0·7)	23	39	62
	Dec.					
1997	Jan.	52·2	(1·2)	22	17	39
	Feb.					
	Mar.	52·0	(1·1)	18	31	49
	Apr.					
	May	49·5	(1·5)	15	15	30
	Jun.	53·4	(0·7)	48	49	97
	Jul.	54·3	(0·6)	33	64	97
	Aug.	55·0	(0·5)	54	63	117
	Sep.	54·1	(0·8)	20	17	37
	Oct.	53·1	(0·5)	34	30	64
	Nov.	50·4	(0·8)	8	19	27
1998	Feb.	na	na	na	na	9
	Jun.	68·6	(2·9)	8	12	20

NaOH until activity was lowered to about 10 000 cpm per 50 µl of solution. T_4 and T_3 antisera (Sigma Chemicals, St Louis, MO) were diluted in the fitting buffers until binding with the null standard was at 50–60%.

A total of 1143 samples was analysed in duplicate with RIA. The first step in the procedure consisted of introducing 50 µl of each tracer onto G-25 Quik-Sep columns (Isolab Inc., Akron, OH) previously equilibrated in 0·1 NaOH. This was followed by the introduction of standards (50 µl of T_4 and 50 µl of T_3) or serum samples (100 µl). Then columns were swirled and the bottom caps removed. After the tracers and standards (or samples) had drained completely into the Sephadex columns, 2 ml of barbital buffer was added to each column and the eluates containing free iodide (^{125}I contamination) were allowed to run to waste. [^{125}I contamination was quantified and corrected for by collecting the eluates from the non-specific binding (NSB) columns.] Then T_4 antiserum (1·0 ml) was pipetted into the columns and the bottom and top caps were replaced to prevent evaporation. After a 4-h incubation at room temperature, 2 ml of barbital buffer was added and the eluates were collected into glass test tubes (12 × 75 mm) for the measurement of the ^{125}I-T_4 fractions. Columns were moved over new glass tubes for the second phase of the procedure, where T_3 antiserum (0·5 ml) was added to each column and the caps were replaced. At the end of an overnight incubation, 2 ml of phosphate buffer was pipetted into the columns and the eluates were collected for the determination

of the ^{125}I-T_3 fractions. Non-specific binding was determined on separate columns that received ^{125}I-T_4 and T_3 antiserum only, or ^{125}I-T_3 and T_4 antiserum only. Radioactivity was measured using a Beckman Gamma 5500 Counting System. The dilution of pooled serum samples yielded T_4 and T_3 levels close to the theoretical values, and the dilution curves of samples were parallel to the dilution curves of standards. Intra- and interassay coefficients of variation were <10%.

STATISTICS

Monthly means of thyroid hormones varied in parallel for both genders, with the male-female coefficients of determination being 0·71 for T_4 and 0·90 for T_3. Therefore the average of the male and female monthly means were plotted against time and, in some figures, trend lines were fitted with three-point centred moving averages. However, before running the three-point average function, missing months (eight out of a 30-month series) were replaced with computed values (linear interpolation of two neighbouring values). The trend lines that were generated with this procedure revealed strong cyclic changes over the study period, and autocorrelation function analysis confirmed that the series were highly autocorrelated and thus unsuitable for regression analysis. Therefore, correlation analysis was used to quantify the degree to which the variables varied together. No interpolated values were included in the correlation analyses; the analyses were performed on measured data (monthly means) only. A correlation was considered significant when the correlation coefficient (r) was greater than twice its standard error.

In addition, cross-correlation function (CCF) analysis was used to examine whether thyroid series lagged behind environmental or physiological changes, a situation that might be expected if the concentration levels of circulating hormones changed more slowly than those in the regulatory variable. Because CCF analysis is appropriate only for observations at equally spaced time intervals, missing monthly means were replaced with linearly interpolated values. CCF analyses were performed on continuous time series covering the period July 1995 to November 1997, with the exception of those involving condition indices, which were limited to the period June 1996 to November 1997 because it was judged that too many data points were missing during the winter and spring of 1996. The results of each CCF analysis were inspected to determine the lag (0–6 months) at which the maximum correlation coefficient occurred between the two variables. Then the previously interpolated values were removed and the two variables correlated at the fitting lag. Therefore, although the identification of the lag time was based on both measured and interpolated values, the correlation coefficients themselves were computed using measured data only.

RESULTS

Thyroid hormones were examined in relation to the size of fish sampled. For each sampling cruise, the concentrations of thyroid hormones in individuals were regressed against their fork length. Significant although generally weak relationships were found in only 7% of cruises for T_4 and 16% for T_3. Moreover, the relationships did not appear at a particular season during the year and the slopes were manifested in both directions, indicating that hormone levels were not influenced by size of fish.

Serum T_4 levels ranged from *c.* 1 to 12 ng ml^{-1} (monthly means), whereas serum T_3 varied from *c.* 2 to 27 ng ml^{-1}. Both hormones varied similarly across seasons, with a single peak occurring annually (Fig. 2). In general, the hormones increased from summer (T_3) or early autumn (T_4) to maxima in mid-winter and declined abruptly during spring. The complete T_4 and T_3 series were introduced into a CCF analysis to determine the extent to which the two hormones

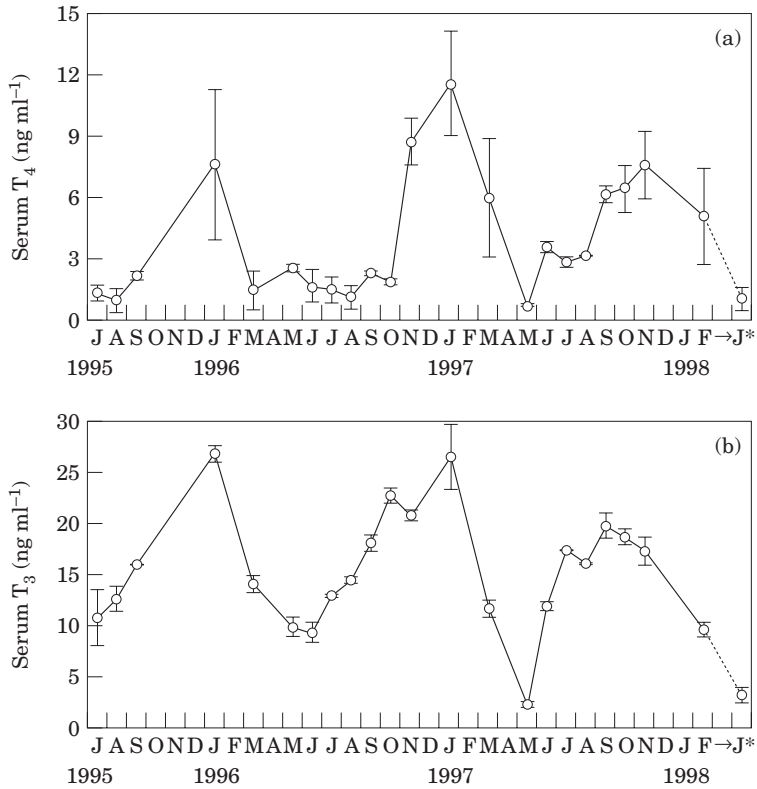

FIG. 2. Seasonal changes in serum L-thyroxine (T_4) (a) and triiodo-L-thyronine (T_3) (b) levels in southern Gulf of St Lawrence cod. Data points represent averages of male and female monthly means. Error bars show S.E., J*, June 1998.

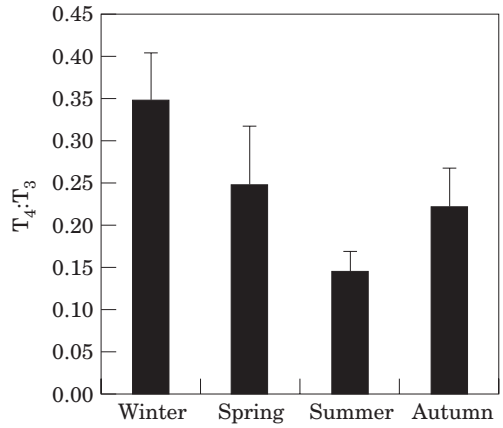

FIG. 3. Seasonal averages of T_4 : T_3 molar ratios for southern Gulf of St Lawrence cod. The averages are based on monthly means. Error bars show S.E.

co-varied. The peak correlation coefficient appeared when the T_3 cycle was set ahead of the T_4 cycle by 1 month. However, after removing the July–September period from the series, the two hormones co-varied simultaneously, indicating

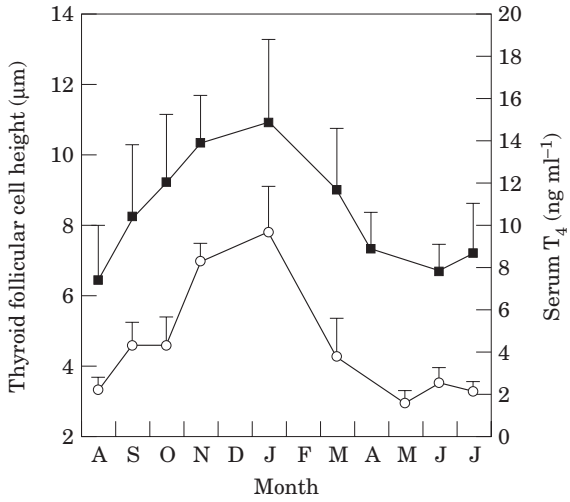

FIG. 4. Seasonal changes in thyroid follicular cell height in Barents Sea cod (August 1956 to July 1957—data from Woodhead, 1959) (■) and serum T_4 in the southern Gulf of St Lawrence cod (○). T_4 represents average of 1996 and 1997 series. Error bars represent S.E.

that the T_3 lead over T_4 was limited to the summer period. The T_4/T_3 monthly means were lowest in summer (Fig. 3).

The above seasonal changes are strikingly similar to the seasonal changes in the thyroid cell height reported for the Barents Sea cod (Woodhead, 1959) (Fig. 4). A CCF analysis indicated that the T_4 and histological index co-vary simultaneously. Moreover, a correlation analysis showed that the coefficient of determination between T_4 and the histological index was high ($r^2=0{\cdot}84$). However, the extrathyroidally produced T_3, does not co-vary as well with the histological index, the coefficient of determination being 0·58.

Thyroidal cycles, in relation to bottom water temperature, photoperiod, and condition, were considered next. In simple correlation analyses, T_4 was weakly correlated ($r^2=0{\cdot}28$) with temperature whereas T_3 was not correlated with temperature (Table II). Moreover, a visual examination of the time series suggested that T_4, and particularly T_3, preceded the temperature cycle [Fig. 5(a,b)]. Such a lead by thyroid hormones is counter-intuitive to a temperature control mechanism. So CCF analyses were used to examine the possibility that the thyroid hormones lagged behind temperature. The hormones were strongly correlated with temperature only when a four to five month lag was applied (Table II). Also, the correlations at these lags were negative. In contrast, thyroid hormones and the thyroid histological index (data taken from Woodhead, 1959) were correlated simultaneously and positively with night-length ($r^2=0{\cdot}58-0{\cdot}92$) [Fig. 5(c,d)]. CCF analyses failed to improve the correlations with night-length. As regards condition indices, T_4 and T_3 were correlated negatively with the stomach fullness index [Fig. 6(a,b)]. A positive and strong correlation was found when T_4 lagged behind the stomach fullness index by five months [Fig. 6(a)], or the condition factor by one month [Fig. 6(e)]. T_3 in turn co-varied simultaneously and positively with the I_H [$r^2=0{\cdot}43$, Fig. 6(d)] and condition factor [$r^2=0{\cdot}52$, Fig. 6(f)], although only during the June to October period.

TABLE II. Correlation and cross-correlation function (CCF) analyses between environmental and thyroid data presented in Fig. 5, and between condition and thyroid data presented in Fig. 6. r^2=coefficient of determination. The thyroid follicular cell height data were taken from Woodhead (1959)

Variable 1	Variable 2	Correlation		CCF		
		r^2	r^a	Lag[b]	r^2	r^a
Environment						
Temperature	T_4	0·28*	+	5	0·85*	−
	T_3	0·13		4	0·59*	−
Night-length	T_4	0·59*	+			
	T_3	0·58*	+			
	Follicular cell height	0·92*	+			
Condition						
Stomach fullness	T_4	0·44*	−	5	0·70*	+
	T_3	0·31*	−			
I_H	T_4	0·03				
	T_3	0·43*[JO]	+			
Condition factor	T_4	0·30*[JO]	+	1	0·47*	+
	T_3	0·52*[JO]	+			

[a]Sign associated with correlation coefficient *r*.
[b]Number of months that thyroid cycles lagged behind environmental or condition cycles.
*Significant correlation (r>2 S.E.).
[JO]Analysis including the June to October (1995–1997) period only.

DISCUSSION

DESCRIPTION OF THYROID CYCLES

Serum T_4 in wild cod varied seasonally from *c.* 1 to 12 ng ml^{-1} (monthly means), a range which is comparable with those of other demersal species sampled after catch: T_4 varied from *c.* 2 to 16 ng ml^{-1} in the winter flounder (Eales & Fletcher, 1982), and from *c.* 3 to 13 ng ml^{-1} in the plaice (Osborn & Simpson, 1978). In contrast, levels of the extrathyroidally produced T_3 were more variable and seemingly higher than those reported for flounder and plaice. T_3 ranged from 2 to 27 ng ml^{-1} in cod, compared with *c.* 2 to 19 ng ml^{-1} in flounder (Eales & Fletcher, 1982) and 3–9 ng ml^{-1} in plaice (Osborne & Simpson, 1978). Also, unlike flounder and plaice, cod exhibited T_3 levels that were consistently higher than T_4 levels. Cyr *et al.* (1998) found similar T_4–T_3 concentration differences for laboratory male cod (females unavailable). Therefore, the study on both sexes reinforces the suggestion that cod have a remarkable capacity for synthesizing T_3.

Serum T_4 and T_3 in field-collected cod followed a consistent annual pattern with a single peak occurring in winter. A short-lasting but recurring divergence in the courses taken by the two hormones was noted: T_3 levels began rising during summer whereas T_4 levels remained low until at least September. Different results pertaining to seasonal changes of thyroid hormones were obtained under laboratory conditions (Cyr *et al.*, 1998), where male cod displayed three annual T_3 peaks (October, February, and May) and no obvious

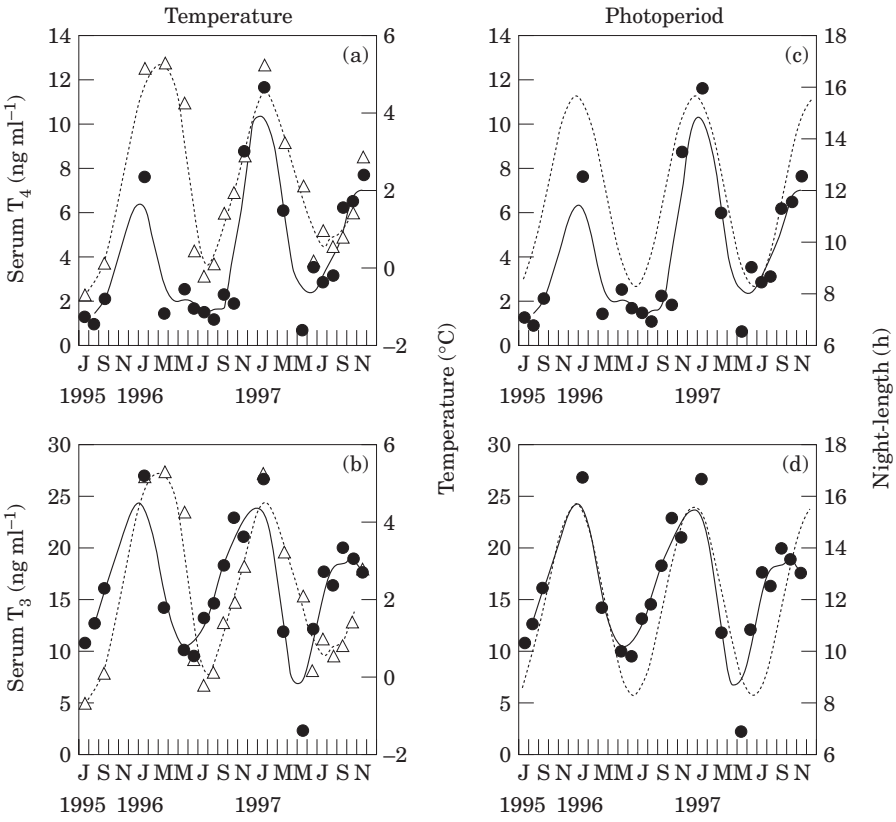

FIG. 5. Cycling of thyroid hormones in relation to bottom water temperature and night-length. ●, Monthly means of either T_4 [(a), (c)] or T_3 [(b), (d)]; △, bottom water temperature. Trend lines show three-point centred moving averages for hormones (——) and temperature (– – –) series. Night-length dotted lines (under Photoperiod) simply connect data points on 15th of each month.

T_4 cycle. The exact reasons for these contrasting results cannot be answered, although satiation feeding (Mackenzie *et al.*, 1998) or stress associated with captivity and stocking density (Laidley & Leatherland, 1988) may have distorted the natural course of thyroidal changes in laboratory cod. Similarly, it is recognized that the cod sampled at sea were stressed to some extent by capture and the sudden removal from their deep-water environment. The total time between the capture near sea bottom and the collection of blood samples was <90 min for most individuals. In comparison, significant changes in plasma T_4 levels in rainbow trout *Oncorhynchus mykiss* (Walbaum) were detected only 8 h after treatment with a handling and asphyxiation stressor (Leatherland, 1985) and 2 h following deliberate disturbance (Himick & Eales, 1990). Brown *et al.* (1978) found no changes in rainbow trout plasma T_4 30 min following physical injury; a peak in T_4 was recorded only 2 h following injury. Plasma T_3 levels were unaltered by the handling/asphyxiation stressor (Leatherland, 1985) and deliberate disturbance (Himick & Eales, 1990), and were mostly unresponsive to physical injury (Brown *et al.*, 1978). Eales & Fletcher (1982) came to similar conclusions after examining the flounder's thyroid responsiveness to a physical stressor (individuals kept upside down in a confined area for 1 h). Therefore, the

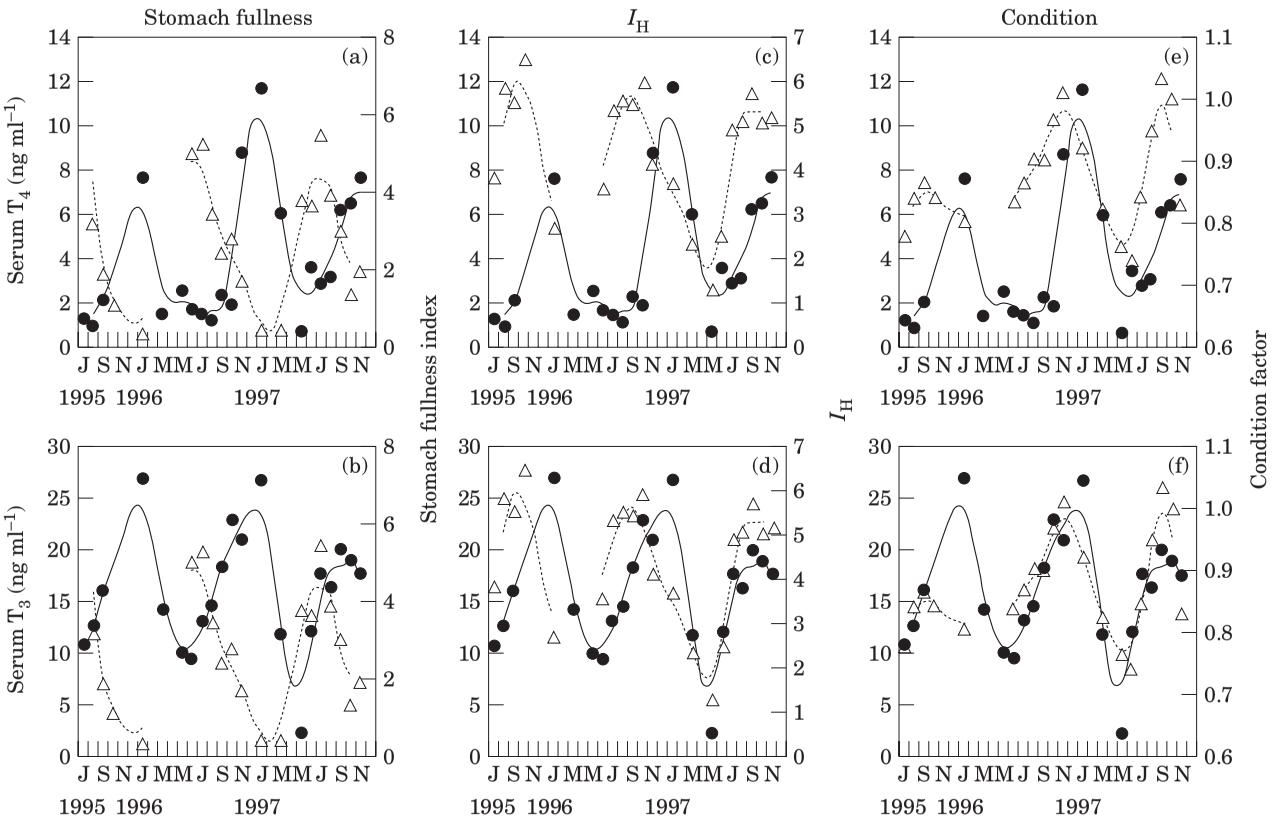

FIG. 6. Cycling of thyroid hormones in relation to the stomach fullness index, I_H, and condition factor. ●, Monthly means of either T_4 [(a), (c), (e)] or T_3 [(b), (d), (f)]; △, stomach fullness index (a), (b), I_H (c), (d), condition factor (e), (f). Trend lines show three-point centred moving averages for hormones (——), stomach fullness index (– – – under Stomach fullness), I_H (– – – under I_H), and condition factor (– – – under Condition). Dotted trend lines were not plotted for the winter of 1996 because too many data points were missing.

cycles of thyroid hormones in this study probably reflect natural changes given the relatively short period of time associated with the capture event.

The present data were correlated unexpectedly with those of another field investigation, in which the thyroid follicular cell height was monitored as a proxy for thyroid hormone production in Barents Sea cod (Woodhead, 1959). While histological indices are considered unreliable for detecting short-term changes in thyroidal status, they have been successful in detecting chronic changes in plasma T_4 in salmonids (Eales & Brown, 1993). In the present study, the correlation between the histological index and hormones was particularly strong for T_4 ($r^2=0{\cdot}84$), which is consistent with the view that the thyroid secretes primarily T_4. Therefore, the southern Gulf of St Lawrence and Barents Sea cod probably share a similar annual cycle of thyroid secretions. This result is of particular interest since the two cod populations inhabit very different oceanographic systems, temperate and sub-arctic, on different sides of the Atlantic. The parallel changes in thyroid function for both populations suggest that the thyroid is an important regulator of physiological processes in cod.

However, the seasonal course of thyroidal secretions seems to differ among the few marine fishes investigated to date. T_4 followed different annual patterns in the winter flounder (Eales & Fletcher, 1982), plaice (Osborn & Simpson, 1978), and cod (this study). Nevertheless, similarities between the marine fishes emerge when thyroidal status is judged by the $T_4 : T_3$ ratio. This ratio is highest during winter (cod, plaice) or spring (flounder), and lowest during summer (cod, plaice, flounder) (Osborn & Simpson, 1978; Eales & Fletcher, 1982; this study). In other teleosts, the $T_4 : T_3$ (or $T_3 : T_4$) ratio has been found to be a sensitive index of thyroidal changes that were not necessarily detected by T_4 or T_3 levels alone (Eales & Brown, 1993; Björnsson *et al*., 1998). The ratio is capable of signalling a shift in the equilibrium between T_4 and T_3 serum pools and, conceivably, also between T_4 and T_3 production or degradation rates. At present, the few data available suggest that the summer $T_4 : T_3$ disequilibrium is a common thyroidal feature among temperate marine teleosts. The likely environmental and physiological factors regulating the course of T_4, T_3, and $T_4 : T_3$ ratios are identified below.

CONTROL OF THYROID CYCLES

Swift (1960) suggested that the seasonal changes in thyroidal activity in cold-water teleosts are regulated primarily by water temperature. However, more recent data concerning the influence of temperature on blood circulating levels of thyroid hormones are conflicting. On the one hand, plasma levels of thyroid hormones were sensitive to temperature in starved eels *Anguilla anguilla* L. (Leloup *et al*., 1984; Leloup & De Luze, 1985) and also in trout fed specific diets (Leatherland *et al*., 1977, 1980). On the other hand, levels of thyroid hormones were not altered by temperature in trout fed common diets (Eales *et al*., 1982, 1986) or cod fed to satiation with capelin *Mallotus villosus* (Müller) (Cyr *et al*., 1998). For cod sampled in their natural habitat (this study), T_4 was weakly correlated ($r^2=0{\cdot}28$) with spatio-temporal changes in water temperature. Strong and negative correlations were found when thyroid hormones lagged temperature changes by 4–5 months, suggesting, for instance,

that peak winter temperatures resulted in reduced thyroidal status in spring or summer. These findings make it unlikely that thyroid cycles in cod were entrained by temperature.

A temperature effect on thyroidal status might be detected through the $T_4 : T_3$ ratios. Elevated water temperatures in summer were suspected of lowering of this ratio in rainbow trout (Osborn *et al.*, 1978), plaice (Osborn & Simpson, 1978), and winter flounder (Eales & Fletcher, 1982). Eales & Fletcher (1982) suggested that the lowering of the $T_4 : T_3$ ratio in summer could be attributed in part to elevated temperatures acting upon T_4 degradation rates and T_4 to T_3 conversion rates. This interpretation is consistent with laboratory work showing that temperature tends to increase T_4 degradation rates (Eales *et al.*, 1982), *in vivo* T_4 to T_3 conversion rates (Eales *et al.*, 1982), and *in vitro* hepatic T_4ORD (T_4 outer-ring deiodinase—responsible for T_4 to T_3 conversion) activity (Johnston & Eales, 1995) in the rainbow trout. Yet, in the present study, such effects of temperature were not noticeable through the seasonal changes in the $T_4 : T_3$ ratio. Although there was a significant lowering of the ratio during summer, as reported for other temperate species, cod inhabits colder waters in summer than in winter [Swain *et al.*, 1998; see also Fig. 5(a)]. In keeping with this observation, it seems unlikely that habitat temperature acting on T_4 degradation rates or T_4 to T_3 conversion rates lowered the $T_4 : T_3$ ratio during summer. Instead, cold water in summer might have lowered the $T_4 : T_3$ ratio by reducing T_4 secretion rates and possibly also T_3 clearance and tissue uptake rates. However, there is no support yet for the hypothesis that temperature regulates T_4 release in cod (Cyr *et al.*, 1998; this study), and the data as to whether the effects of temperature on T_3 clearance rates can modify T_3 levels in the blood compartment are conflicting (Leloup *et al.*, 1984; Leloup & De Luze, 1985; Eales *et al.*, 1986). In summary, there was no obvious relationship between habitat temperature and thyroid cycling (T_4, T_3, $T_4 : T_3$), and thus it is suggested that the annual thyroid cycles in wild cod are not temperature-driven.

The effects of photoperiod on the thyroidal system of marine teleosts are not documented, but short day-lengths or complete darkness tend to stimulate the thyroid in freshwater and euryhaline species (Eales, 1979; Brown, 1988). In these taxa at least, short day-lengths may decrease the sensitivity of the hypothalamic-hypophyseal negative feedback mechanism, thereby enabling prolonged thyroidal responses to the thyroid-stimulating hormone (Brown & Stetson, 1985; Grau *et al.*, 1985). Strong field correlations in the present study support such a thyroid responsiveness model: the night-length was correlated positively and simultaneously with the thyroid follicular cell height ($r^2 = 0{\cdot}92$) (data taken from Woodhead, 1959) and the major thyroid secretion (T_4) ($r^2 = 0{\cdot}59$). These correlations suggest that photoperiod is the main entrainer of thyroid secretions in cod. Woodhead & Woodhead (1965) came to a similar conclusion after reviewing their histological data for Barents Sea cod. However, they suggested that the activation of the thyroid is cued to the autumn equinox and not to the rate of change of photoperiod. In agreement with this model, the present T_4 series did not show any increasing trend before September. However, once activated at the autumn equinox it seems likely that the thyroid in cod would respond to the rate of change of photoperiod, as suggested by present field correlations.

There was a positive correlation between night-length and T_3 ($r^2=0{\cdot}58$), which suggests that photoperiod might have acted also on the peripheral control of thyroidal status. As the majority of T_3 in teleosts is thought to derive from T_4ORD activity in peripheral tissues (Eales & Brown, 1993), the field correlation suggests that photoperiod acted specifically on the T_4 deiodinating pathways. Unfortunately, tissue T_4ORD activity was not monitored and so it cannot be verified that T_4ORD varied in concert with photoperiod in cod. However, Cyr *et al.* (1998) exposed cod to a naturally simulated photoperiod but constant temperature (2–4° C) and feeding regime, and reported that hepatic T_4ORD activity was about three-fold higher in February than in June. While this experiment was not designed specifically to test the effects of photoperiod, its results, together with the present night-length *v.* T_3 field correlation, raise the question as to whether photoperiod entrained an underlying rhythm of T_4ORD activity. Alternatively, it is possible T_4ORD activity was not affected by photoperiod directly, but by T_4 substrate availability. In this case, the diminishing daylength in winter would have increased T_4 levels (see above), which in turn would have enhanced T_4ORD activity and thus T_3 levels in circulation. However, a photoperiod control mechanism of T_3, either direct or indirect (*via* T_4), does not conform to the present summer results. Indeed, it appears that T_3 preceded night-length during summer, particularly in 1997 [Fig. 5(d)]. The T_3 lead over night-length, although small (about one month), implies that the T_3 producing mechanism was insensitive to photoperiod at this time of year. Also, low T_4 levels during summer are inconsistent with the idea that T_4 increased T_4ORD activity and T_3 levels in circulation. Therefore, the data indicated that photoperiod might have some influence upon T_3 production in cod, except during the summer period where the potential effects of photoperiod seemed overriden by other factors.

The thyroid cycles in cod might have been influenced by marked seasonal changes in feeding intensity. In aquaculturally important species, thyroid hormones have been related positively to food intake, as demonstrated by various experimental designs (food deprivation, refeeding, different rations) (Eales, 1988; Mackenzie *et al.*, 1998). For cod, thyroid hormones were negatively correlated with the stomach fullness index, which is remarkably inconsistent with experimental evidence. A positive correlation was found only when the T_4 cycle lagged behind the stomach fullness index by 5 months. Thus there was no evidence that the thyroidal system in cod responded instantaneously to the seasonal changes in feeding intensity. However, it seems that T_3 levels were influenced by changes in condition during summer and autumn (June to October). At this time, T_3 co-varied simultaneously with both the I_H ($r^2=0{\cdot}43$) and the condition factor ($r^2=0{\cdot}52$). The difference between the two condition indices may be related to their capacity in reflecting changes in T_4ORD activity in cod. It is possible that the condition factor, which yielded the highest coefficient of determination, reflected changes in total (all tissues) T_4ORD activity. The I_H in turn may be correlated with hepatic T_4ORD activity mostly. In keeping with these assumptions, one might conclude that the liver alone explained a substantial proportion of the variation in T_3 levels. As T_4ORD activity was not monitored, it cannot be confirmed that the recorded seasonal changes in the condition indices were accompanied by parallel changes in the

cod's potential for generating T_3. Nonetheless, it is concluded that the thyroidal trends in wild cod were in broad agreement with the emerging view that the primary control mechanism for teleostean T_3 is peripheral, with the liver playing a central role (Farbridge *et al.*, 1992; Eales & Brown, 1993; Morin *et al.*, 1993; Van der Geyten *et al.*, 1998).

Together the above correlations lead to the suggestion that the diverging courses taken by T_4 and T_3 during summer, and hence the lowering of the $T_4 : T_3$ ratios, were related to increased day-length and condition. The overall impression is that long day-lengths reduced T_4 secretion at the same time as the rising metabolic state of cod increased T_3 production. Further, because T_3 is derived from T_4 (Eales & Brown, 1993), it is possible that enhanced T_3 production during summer also contributed to the lowering of serum T_4 levels (i.e. on top of the thyroid's low T_4 release). This intrepretation differs from the one presented for the trout (Osborn *et al.*, 1978) and winter flounder (Eales & Fletcher, 1982), where temperature was identified as one likely cause for the summer disequilibrium between T_4 and T_3. Present findings are more consistent with those of Osborn & Simpson (1978), who suggested that the plaice's thyroid might be more sensitive to changes in day-length than temperature.

FUNCTIONAL ROLE OF THYROID CYCLES

Thyroid hormones are widely recognized as permissive growth inducers in aquaculturally important teleosts (mainly salmonids) (Donaldson *et al.*, 1979; Higgs *et al.*, 1979, 1982). This growth-promoting role has yet to be validated for adult marine fishes displaying marked seasonal changes in feeding intensity and growth patterns. Recent data on the seasonal changes in carcass weights for the southern Gulf of St Lawrence cod (Schwalme & Chouinard, 1999) indicate that maximal growth rates occur between June and October in this population. In comparison, serum T_4 levels in the present study started increasing in autumn only. However, T_3 levels started increasing during summer well in advance of T_4. The closer association between T_3 and maximal growth rates in cod is consistent with evidence that T_3 is more effective than T_4 in promoting fish growth (Higgs *et al.*, 1979). It is equally interesting that serum T_3 levels, in agreement with the hormone's stimulating effect on early ovarian development in trout (Cyr & Eales, 1996), started increasing immediately after spawning (early summer, Schwalme & Chouinard, 1999). Thus the promotion of early gonadal development is a second plausible reason for the increasing T_3 levels in cod during summer. In contrast, the lowering of T_3 levels in the final stages of gonadal development (spring, Schwalme & Chouinard, 1999) is consistent with the view that thyroid hormones are inessential to the latter stages of the reproductive events (Cyr & Eales, 1996). Moreover, by curtailing somatic growth, a suppressed thyroidal status in spring may allow the remaining metabolic reserves to be directed towards continued gonadal growth.

While T_3 may be involved in the stimulation of somatic and early gonadal growth, T_4 could function as an endocrinological trigger and metabolic stimulus to the *c.* 500 km (Hanson, 1996) autumn migration. Until now support for this hypothesis has been based on the cycling of the thyroid follicular cell height in migratory Barents Sea cod (Woodhead, 1959, 1975; Woodhead & Woodhead, 1965), and also on the observation of increased locomotor activity in T_4-treated

cod (Woodhead, 1970; Castonguay & Cyr, 1998). Present work has provided the first documentation of an autumnal T_4 increase inside a highly migratory cod population, adding support to the view that T_4 might be involved in the physiological mechanism triggering migration.

CONCLUSION

In conclusion, blood-circulating levels of T_4 in cod were similar to those reported for plaice (Osborn & Simpson, 1978) and winter flounder (Eales & Fletcher, 1982), although T_3 levels were comparatively elevated in cod. Both hormones followed a consistent annual pattern with a single peak occurring in winter. A comparative analysis with previous work suggests that: (1) thyroid cycling varied identically in the two cod populations and (2) at least one thyroidal feature, the summer disequilibrium between T_4 and T_3 circulating levels, may be common in temperate marine teleosts. Spatio-temporal changes in water temperature provided no satisfactory explanation for thyroidal patterns. However, it seems that the metabolic condition of cod regulated T_3 levels during summer and autumn. These results imply that the seasonal stimulation of somatic and gonadal growth by T_3 is determined ultimately by food resources, and not by variations in water temperature. Photoperiod was identified as the prime candidate for entraining the annual cycle of T_4 release from the thyroid. There are also indications that the thyroid's response to photoperiod might be triggered by the autumn equinox. Such a mechanism would lead to a synchronicity throughout the northern hemisphere of physiological and behavioural processes under the influence of T_4, such as perhaps the onset of seasonal migrations.

We thank the staff at Biorex Inc. for their technical assistance throughout this study; O. Benoit for coordinating efforts; B. Aucoin, S. Boucher, B. Branch, C. Doiron, S. Godin, B. Harper, and G. Vienneau for field assistance; skippers P.-H. Haché and F. Paulin and their crews for help with sampling; the Bedford Institute of Oceanography for provision of materials and facilities to L.A.C; P. Comeau, P. Fanning, and W. MacEachern for help with data collection at sea; D. Cyr for advice on RIA procedures; and P. Cormier, B. Irwin and J. Smith for laboratories facilities for the isotopic work.

References

Björnsson, B. T., Halldórsson, Ó., Haux, C., Norberg, B. & Brown, C. L. (1998). Photoperiod control of sexual maturation of the Atlantic halibut (*Hippoglossus hippoglossus*): plasma thyroid hormone and calcium levels. *Aquaculture* **166,** 117–140.

Boeuf, G. (1993). Salmonid smolting: a pre-adaptation to the oceanic environment. In *Fish Ecophysiology* (Rankin, J. C. & Jensen, F. B., eds), pp. 105–135. London: Chapman & Hall.

Brown, C. L. (1988). Photoperiod effects on thyroid function in fish. In *Processing of Environmental Information in Vertebrates* (Stetson, M. H., ed.), pp. 1–17. New York: Springer-Verlag.

Brown, C. L. & Kim, B. G. (1995). Combined application of cortisol and triiodothyronine in the culture of larval marine finfish. *Aquaculture* **135,** 79–86.

Brown, C. L. & Stetson, M. H. (1985). Photoperiod-dependent negative feedback effects of thyroid hormones in *Fundulus heteroclitus. General and Comparative Endocrinology* **58,** 186–191.

Brown, S., Fedoruk, K. & Eales, J. G. (1978). Physical injury due to injection or blood removal causes transitory elevations of plasma thyroxine in rainbow trout, *Salmo gairdneri. Canadian Journal of Zoology* **56,** 1998–2003.

Campana, S. E., Chouinard, G. A., Hanson, J. M. & Fréchet, A. (1999). Mixing and migration of overwintering Atlantic cod (*Gadus morhua*) stocks near the mouth of the Gulf of St Lawrence. *Canadian Journal of Fisheries and Aquatic Sciences* **56,** 1873–1881.

Castonguay, M. & Cyr, D. G. (1998). Effects of temperature on spontaneous and thyroxine-stimulated locomotor activity of Atlantic cod. *Journal of Fish Biology* **53,** 303–313.

Castonguay, M., Dutil, J.-D., Audet, C. & Miller, R. (1990). Locomotor activity and concentration of thyroid hormones in migratory and sedentary juvenile American eels. *Transactions of the American Fisheries Society* **119,** 946–956.

Cyr, D. G. & Eales, J. G. (1996). Interrelationships between thyroidal and reproductive endocrine systems in fish. *Reviews in Fish Biology and Fisheries* **6,** 165–200.

Cyr, D. G., Idler, D. R., Audet, C., McLeese, J. M. & Eales, J. G. (1998). Effects of long-term temperature acclimation on thyroid hormones deiodinase function, plasma thyroid hormone levels, growth, and reproductive status of male Atlantic cod, *Gadus morhua. General and Comparative Endocrinology* **109,** 24–36.

Donaldson, E. M., Fagerlund, U. H. M., Higgs, D. A. & McBride, J. R. (1979). Hormonal enhancement of growth. In *Fish Physiology*, Vol. 8 (Hoar, W. S. & Randall, D., eds), pp. 455–597. London: Academic Press.

Eales, J. C. (1979). Thyroid function in cyclostomes and fishes. In *Hormones and Evolution*, Vol. 1 (Barrington, E. J. W., ed.), pp. 341–436. New York: Academic Press.

Eales, J. (1988). The influence of nutritional state on thyroid function in various vertebrates. *American Zoologist* **28,** 351–362.

Eales, J. G. & Brown, S. B. (1993). Measurement and regulation of thyroidal status in teleost fish. *Reviews in Fish Biology and Fisheries* **3,** 299–347.

Eales, J. G. & Fletcher, G. L. (1982). Circannual cycles of thyroid hormones in plasma of winter flounder (*Pseudopleuronectes americanus* Walbaum). *Canadian Journal of Zoology* **60,** 304–309.

Eales, J. G., Chan, J. P., Van Der Kraak, G. J., Omeljaniuk, R. J. & Uin, L. (1982). Effects of temperature on plasma thyroxine and iodide kinetics in rainbow trout, *Salmo gairdneri. General and Comparative Endocrinology* **47,** 295–307.

Eales, J. G., Van Der Kraak, G., Chang, J. P. & Omeljaniuk, R. J. (1986). Effects of temperature on triiodothyronine plasma levels, kinetics and hepatocyte nuclear binding in rainbow trout, *Salmo gairdneri. Canadian Journal of Zoology* **12,** 2658–2664.

Farbridge, K. J., Flett, P. A. & Leatherland, J. F. (1992). Temporal effects of restricted diet and compensatory increased dietary intake on thyroid function, plasma growth hormone levels and tissue lipid reserves of rainbow trout *Oncorhynchus mykiss. Aquaculture* **104,** 157–174.

Godin, J. G., Dill, P. A. & Drury, D. E. (1974). Effects of thyroid hormones on behaviour of yearling Atlantic salmon (*Salmo salar*). *Journal of the Fisheries Research Board of Canada* **31,** 1787–1790.

Gorbman, A., Dickhoff, W. W., Vigna, S. R., Clark, N. B. & Ralph, C. L. (1983). *Comparative Endocrinology*. New York: John Wiley.

Grau, E. G., Dickhoff, W. W., Nishioka, R. S. & Folmar, L. C. (1981). Lunar phasing of the thyroxine surge preparatory to seaward migration of salmonid fish. *Science* **211,** 607–609.

Grau, E. G., Brown, C. L. & Stetson, M. H. (1985). Photoperiod regulation of thyroid responsiveness to TSH in *Fundulus heteroclitus. The Journal of Experimental Zoology* **234,** 199–205.

Hanson, J. M. (1996). Seasonal distribution of juvenile Atlantic cod in the southern Gulf of St Lawrence. *Journal of Fish Biology* **49,** 1138–1152.

Higgs, D. A., Fagerlund, U. H. M., McBride, J. R. & Eales, J. G. (1979). Influence of orally administered L-thyroxine or 3,5,3′-triiodo-L-thyronine on growth, food consumption and food conversion efficiency of underyearling coho salmon (*Oncorhynchus kisutch*). *Canadian Journal of Zoology* **57,** 1974–1979.

Higgs, D. A., Fagerlund, U. H. A., Eales, J. G. & McBride, J. R. (1982). Application of thyroid hormones and steroid hormones as anabolic agents in fish culture. *Comparative Biochemistry and Physiology* **73B,** 143–176.

Himick, B. A. & Eales, J. G. (1990). Acute correlated changes in plasma T sub(4) and glucose in physically disturbed cannulated rainbow trout, *Oncorhynchus mykiss. Comparative Biochemistry and Physiology* **97A,** 165–167.

Høgåsen, H. R. & Prunet, P. (1997). Plasma levels of thyroxine, prolactin, and cortisol in migrating and resident wild arctic char, *Salvelinus alpinus. Canadian Journal of Fisheries and Aquatic Sciences* **54,** 2947–2954.

Inui, Y. & Miwa, S. (1985). Thyroid hormone induces metamorphosis in flounder larvae. *General and Comparative Endocrinology* **60,** 450–454.

Inui, Y., Miwa, S. & Yamano, K. (1994). Hormonal control of flounder metamorphosis. In *Perspectives in Comparative Endocrinology* (Davey, K. G., Peter, R. E. & Tobe, S. S., eds), pp. 408–411. Ottawa: National Research Council of Canada Publications.

Johnston, C. E. & Eales, J. G. (1995). Effects of acclimation and assay temperature on outer- and inner-ring thyroxine and 3,5,3′-triiodo-L-thyronine deiodination by liver microsomes of rainbow trout, *Oncorhynchus mykiss. Journal of Experimental Zoology* **272,** 426–434.

Laidley, C. W. & Leatherland, J. F. (1988). Cohort sampling, anaesthesia and stocking-density effects on plasma cortisol, thyroid hormone, metabolite and ion levels in rainbow trout, *Salmo gairdneri* Richardson. *Journal of Fish Biology* **33,** 73–88.

Lambert, Y. & Dutil, J. D. (1997). Can simple condition indices be used to monitor and quantify seasonal changes in the energy reserves of Atlantic cod (*Gadus morhua*)? *Canadian Journal of Fisheries and Aquatic Sciences* **54** (Suppl. 1), 104–112.

Leatherland, J. F. (1982). Environmental physiology of the teleostean thyroid gland: a review. *Environmental Biology of Fishes* **7,** 83–110.

Leatherland, J. F. (1985). Studies of the correlation between stress-response, osmoregulation and thyroid physiology in rainbow trout, *Salmo gairdneri* (Richardson). *Comparative Biochemistry and Physiology* **80A,** 523–531.

Leatherland, J. F., Cho, C. Y. & Slinger, S. J. (1977). Effects of diet, ambient temperature, and holding conditions on plasma thyroxine levels in rainbow trout (*Salmo gairdneri*). *Journal of the Fisheries Research Board of Canada* **34,** 677–682.

Leatherland, J. F., Cho, C. Y., Hilton, J. W. & Slinger, S. J. (1980). Further studies on the effect of diet on serum thyroid hormone concentrations and thyroid histology in rainbow trout, *Salmo gairdneri* (Pisces, Salmonidae). *Environmental Biology of Fishes* **2,** 175–179.

Leloup, J. & De Luze, A. (1985). Environmental effects of temperature and salinity on thyroid function in teleost fishes. In *The Endocrine System and the Environment* (Follett, B. K., Ishii, S. & Chandola, A., eds), pp. 23–32. Berlin: Springer-Verlag.

Leloup, J., De Luze, A. & Seugnet, I. (1984). Thyroid function and environmental temperature in eel (*Anguilla anguilla*). *General Comparative Endocrinology* **53,** 498 (abstract).

Mackenzie, D. S., VanPutte, C. M. & Leiner, K. A. (1998). Nutrient regulation of endocrine function in fish. *Aquaculture* **161,** 3–25.

Morin, P. P., Hara, T. J. & Eales, J. G. (1993). Thyroid hormone deiodination in brain, liver, gill, heart and muscle of Atlantic salmon (*Salmo salar*) during photoperiodically-induced parr-smolt transformation. I. Outer- and inner-ring thyroxine diodination. *General and Comparative Endocrinology* **90,** 142–156.

Omeljianuk, R. J., Cook, R. F. & Eales, J. G. (1984). Simultaneous measurement of thyroxine and triiodothyronine in trout plasma using a solid-phase radioimmunoassay. *Canadian Journal of Zoology* **62,** 1451–1453.

Osborn, R. H. & Simpson, T. H. (1978). Seasonal changes in thyroidal status in the plaice, *Pleuronectes platessa* L. *Journal of Fish Biology* **12,** 519–526.

Osborn, R. H., Simpson, T. H. & Youngson, A. F. (1978). Seasonal and diurnal rhythms of thyroidal status in the rainbow trout, *Salmo gairdneri* Richardson. *Journal of Fish Biology* **12,** 531–540.

Paloheimo, J. E. & Kohler, A. C. (1968). Analysis of the southern Gulf of St Lawrence cod population. *Journal of the Fisheries Research Board of Canada* **25,** 555–578.

Schwalme, K. & Chouinard, G. A. (1999). Seasonal dynamics in feeding, organ weights, and reproductive maturation of Atlantic cod (*Gadus morhua*) in the southern Gulf of St Lawrence. *ICES Journal of Marine Science* **56,** 303–319.

Swain, D. P., Chouinard, G. A., Morin, R. & Drinkwater, K. F. (1998). Seasonal variation in the habitat associations of Atlantic cod (*Gadus morhua*) and American plaice (*Hippoglossoides platessoides*) from the southern Gulf of St. Lawrence. *Canadian Journal of Fisheries and Aquatic Sciences* **55,** 2548–2561.

Swift, D. R. (1960). Cyclical activity of the thyroid gland of fish in relation to environmental changes. *Symposia of the Zoological Society of London* **2,** 17–27.

Tanaka, M., Tanangonan, J. B., Tagawa, M., DeJesus, E. G., Nishida, H., Isaka, M., Kimura, R. & Hirano, T. (1995). Development of the pituitary, thyroid and interrenal glands and applications of endocrinology to the improved rearing of marine fish larvae. *Aquaculture* **135,** 111–126.

Van der Geyten, S., Mol, K. A., Pluymers, W., Kühn, E. R. & Darras, V. M. (1998). Changes in plasma T_3 during fasting/refeeding in tilapia (*Oreochromis niloticus*) are mainly regulated through changes in hepatic type II iodothyronine deiodinase. *Fish Physiology and Biochemistry* **19,** 135–143.

Woodhead, A. D. (1959). Variations in the activity of the thyroid gland of the cod, *Gadus callarias* L., in relation to its migrations in the Barents Sea. I. Seasonal changes. *Journal of the Marine Biological Association of the UK* **38,** 407–415.

Woodhead, A. D. (1975). Endocrine physiology of fish migration. *Oceanography and Marine Biology Annual Reviews* **13,** 287–382.

Woodhead, A. D. & Woodhead, P. M. J. (1965). Seasonal changes in the physiology of the Barents Sea cod, *Gadus morhua* L., in relation to its environment. I. Endocrine changes particularly affecting migration and maturation. *International Commission for the Northwest Atlantic Fisheries Special Publication* **6,** 691–715.

Woodhead, P. M. J. (1970). An effect of thyroxine upon the swimming of cod. *Journal of the Fisheries Research Board of Canada* **27,** 2337–2338.