

Sources of Variation in Growth of the European Eel (*Anguilla anguilla*) Estimated from Otoliths

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Tetracycline labelling of otoliths was used for studying the growth of the European eel (*Anguilla anguilla*), which has strong individual variability. Two groups of eels were injected with tetracycline and released into a natural pond (Camargue, south of France) in spring 1989 and in autumn 1989. The marginal growth of otoliths between marking (tetracycline mark) and capture (otolith margin) was measured for all fish sampled until spring 1990. Comparisons of otolith growth rates were made according to fish size, age, sex, and growth period (ANCOVA). The results showed a great variability. After 15 mo of growth, the mean growth in length (back-calculated) was 6.6 cm. The three main factors tested, age, sex, and seasonal growth period, had a significant effect on otolith growth and therefore on somatic growth, since the relation between fish length and that of the otolith is highly significant ($r^2 = 0.802$). There was also a very significant effect of size at marking on otolith growth. It is difficult to rank qualitative importance of these various factors, even though there are no interactions among them. The best absolute growth in Mediterranean lagoons could be attained by 2-yr-old female fish growing in the summer months.

Le marquage à la tétracycline des otolithes a été utilisé pour étudier la croissance de l'anguille européenne (*Anguilla anguilla*) qui présente une forte variabilité individuelle. On a injecté de la tétracycline à deux groupes d'anguilles qui ont été relâchées dans un étang naturel (Camargue, sud de la France) au printemps de 1989 et à l'automne de 1989. La croissance marginale des otolithes entre le marquage (marque à la tétracycline) et la capture (bord de l'otolithe) a été mesurée chez tous les poissons échantillonnés jusqu'au printemps de 1990. On a comparé le taux de croissance des otolithes selon la taille du poisson, l'âge, le sexe et la période de croissance (ANCOVA). Les résultats étaient très variables. Après une croissance de 15 mo, la croissance moyenne en longueur (rétro-calcul) était de 6,6 cm. Les trois principaux facteurs vérifiés, l'âge, le sexe et la période de croissance saisonnière, avaient un effet important sur la croissance des otolithes, et donc sur la croissance somatique, puisque le rapport entre la longueur du poisson et celle de l'otolithe est très significative ($r^2 = 0,802$). Il y avait également un effet très important de la taille au moment du marquage sur la croissance des otolithes. Il est difficile de classer l'importance relative de ces différents facteurs même s'il n'y a pas d'interactions entre eux. C'est chez les femelles âgées de 2 ans en croissance pendant la période estivale que l'on pourrait observer la meilleure croissance absolue dans les bassins méditerranéens.

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Exact estimates of the individual growth of fish in their natural environment can only be made by individual marking. This type of manipulation implies a minimum of four steps: capture of fish, marking, release into a given environment, and recapture. Marking can be external (tags, tattooing, etc.) or internal (magnetic rods, injection of fluorescent markers, various stress actions, etc.) or a combination of the two. Three main constraints are involved in marking experiments: (1) the marking must not affect subsequent growth, (2) the individual marks must remain visible for the entire length of the experiment, and (3) sufficient fish must be recaptured to obtain a good estimate of growth.

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Internal markers of the fluoromarker type (e.g., tetracyclines) have been used for a long time to mark calcified tissues during their mineralisation (Van Coillie 1967; Weber and Rigway 1967; Meunier 1974; Meunier and Boivin 1974). These markers have often been used in experiments to validate the timing of deposition of growth marks on calcified tissues to determine the growth cycle (Beamish and McFarlane 1983, 1987), but they have seldom been used to study the variability in the growth of individual fish. If marking experiments can provide high rates of recapture, vital marking of calcified tissues, e.g., otoliths, can constitute a database for studying individual growth. Where growth is proportional to growth of the calcified piece (otolith), back-calculation can allow growth in length to be estimated. Results of back-calculation must, however, always be interpreted with care because the mathematical methods are

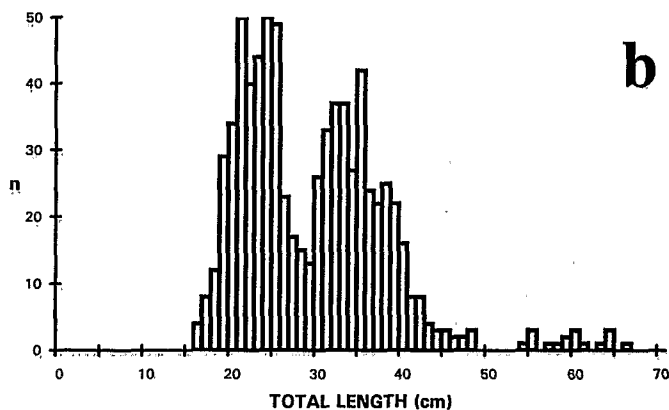
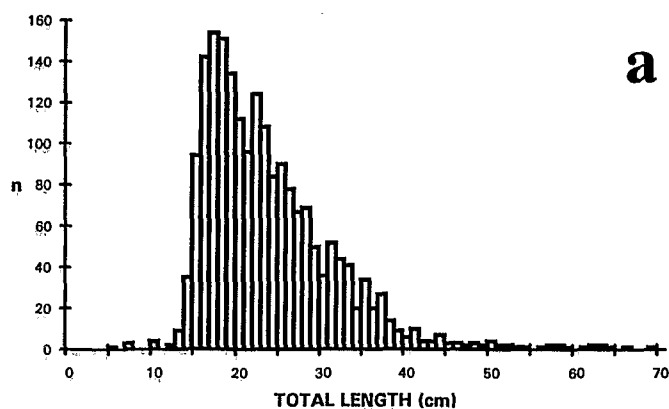


FIG. 1. Size distributions of eels at marking, released in Les Garcines pond. (a) Group 1 marked in March 1989; (b) group 2 marked in October 1989.

varied (Francis 1990) and the possibilities of bias due to the modelling are not insignificant (Small and Taylor 1987; Campana 1990; Ricker 1992).

Individual marking of European eels (*Anguilla anguilla*) during their continental life stage is especially difficult (see Nielsen 1988 for a review). Almost all marking techniques have deleterious effects on this fish, and marking is generally only carried out on groups (e.g., by tattooing). This is undoubtedly due to the morphology of this species and its benthic life style which prevent the use of external markers. In addition, these markers have a considerable effect on subsequent growth as has been shown by marking experiments in natural environments: e.g., Berg (1986) showed that jaw tags led to reduced individual growth, and Tulonen (1989) found that eels marked with Carlin-type tags practically ceased growing.

All the data in the literature on the growth of the European eel in various habitats show extreme qualitative and quantitative individual variability (Vøllestad 1989, 1992; Fontenelle 1991). This variability has been assessed to be great within any one population: in the estuarine environment (Fernández-Delgado et al. 1989), in freshwater habitats (Moriarty 1983; Vøllestad and Jonsson 1988; Poole 1991), in lake environments (Paulovits and Biró 1986; Nagiec and Bahnsawy 1990), and in fish farms (Egusa 1979; Kuhlmann 1979; Wickins 1985). Differences in growth among individuals are not restricted to the European species but have also been found in other species (*Anguilla rostrata*, Hansen and

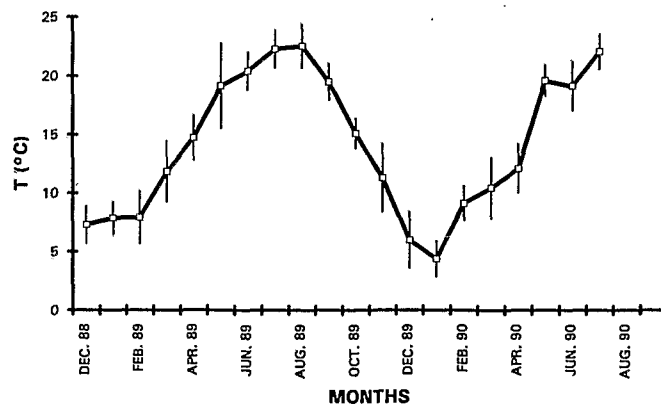


FIG. 2. Monthly means and standard deviations of the daily bottom temperatures in Les Garcines pond during the experiment.

Eversole 1984; *Anguilla mossambica*, McEwan and Hecht 1984; *Anguilla australis*, Chisnall and Hayes 1991). Variability in the growth of the eel has usually been detected either directly in fish farms or indirectly by age studies within populations, but age is a variable that is difficult to assess from otoliths without ambiguity (Michaud et al. 1988; Vøllestad et al. 1988) because of problems of interpretation. Thus, observed variability in length at a given age could be due to errors introduced by the method used for age determination (Vøllestad 1985; Fontenelle 1991). A better understanding of the biotic and abiotic factors leading to this variability would undoubtedly enable more effective management of the exploited stages of this species throughout European inland waters and would also allow aquacultural constraints to be optimised. The sources of variation in eel growth in the natural environment are still to be explored. The main sources of individual variations certainly include age, sex, and seasonality in growth (a function of temperature). It is, however, essential to determine which of the various factors has the preponderant influence before defining the best management criteria.

For the European eel, a marking experiment was carried out with the aim of studying sources of variability in individual growth in a natural environment representative of a category of biotopes colonized by certain populations: the Mediterranean waterbodies of the south of France. Otoliths were marked with tetracycline, this internal mark being an indicator visible later upon recapture at various dates, for growth comparison. The effects of factors such as age, sex, and the growth period were tested.

Materials and Methods

Marking and Sampling

Two groups of eels originating from lagoons in the south of France were marked, in March 1989 (group 1) and in October 1989 (group 2), and then released into a natural freshwater pond (Les Garcines, Camargue), at densities of 30 kg·ha⁻¹ (1973 individuals) and 20 kg·ha⁻¹ (749 individuals), respectively. The eels in the two groups were of various sizes, ranging from 6 to 69 cm at the time of marking (Fig. 1). Marking consisted of (1) internal marking by intraperitoneal injection of tetracycline (75 mg·kg live weight⁻¹) and (2) external marking by ablation of part of the pectoral fin to distinguish between groups and by ventral

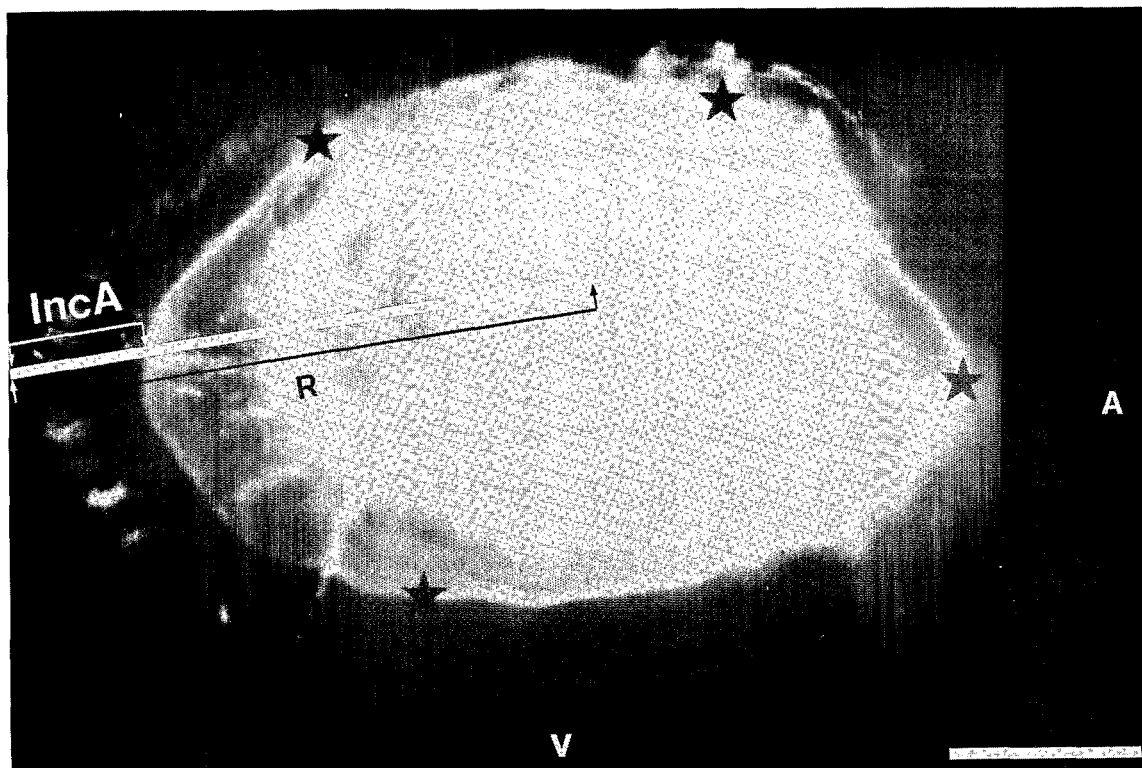


FIG. 3. Tetracycline-labelled otolith observed under reflected light, a dark background, and incident ultraviolet light. This view shows the measurements on the otolith: R , radius of maximum growth on the anteroposterior axis; $IncA$, absolute increment between the tetracycline mark (stars) and the margin. A = anterior; V = ventral. Scale bar = 0.3 mm.

TABLE 1. Number of individuals marked and recaptured at each date in Les Garcines pond.

	Group 1	Group 2
Marking in March 1989	1973 (G1)	
Intermediary captures (April to September 1989)	38	
Captures in October 1989	37 (G1O)	
Marking in October 1989		749 (G2)
Intermediary captures (September 1989 to May 1990)	51	12
Captures in June 1990	602 (G1J)	302 (G2J)

tattooing with alcian blue (subcutaneous injection). Les Garcines pond (2.1 ha) was a closed system throughout the experiment, with managed inputs and outputs of water and a known initial population of fish. Samples of fish were collected at monthly intervals from the first release and a more intensive fishing effort was made before the second release. At the end of the experiment in June 1990, the pond was drained and the remaining pool was treated with rotenone to capture all remaining fish. The numbers of fish marked and recaptured are given in Table 1. The water temperature in the pond was recorded daily throughout the experiment (Fig. 2).

Individual Examination

At each sampling date, the eels were measured (total length (TL), centimetres), weighed (grams), sexed macroscopically (undifferentiated: U, male: M, female: F), and their otoliths (sagitta) extracted. A total of 1004 individuals were recaptured, including 37 from group 1 in October 1989 and 602 from

group 1 and 302 from group 2 in June 1990 (Table 1). All the otoliths were examined whole in reflected light against a black background (cleared in a bath of rosemary essential oil), with the distal face uppermost, under an epifluorescent microscope (Olympus BH, HBO 100-W lamp providing UVB at 490 nm). The tetracycline mark was located by its yellow-green fluorescence under the UVB excitation. Two measurements (millimetres) were made on the otolith (Fig. 3): R , the otolith radius along its anteroposterior axis measured from the nucleus centre to the posterior margin, and $IncA$, the absolute otolith increment between marking and capture, i.e., the distance between the inner margin of the tetracycline mark and the otolith margin along the radius defined above. The relative otolith increment, $IncR$, is the ratio of $IncA$ over R ($IncR = IncA/R$). The age (A , years) of each individual in continental water was calculated by counting the number of opaque rings on the otolith from the nucleus outwards; it had previously been shown that a broad opaque zone and a broad hyaline were laid down each year in French Mediterranean lagoons (Panfili et al. 1992).

Analysis

Several variables are available for comparing individual growth: IncA, IncR, age (A), sex (U, M, F), and the various growth periods (G1O, G1J, G2J). G1O is the growth period for group 1 between March 89 and October 89, i.e., 7 mo including spring and summer. G1J is the growth period for group 1 between March 89 and June 90, i.e., 15 mo including an entire annual cycle. G2J is the growth period for group 2 between October 89 and June 90, i.e., 8 mo including autumn, winter, and the start of spring.

It would be preferable to compare the absolute otolith increments (IncA). However, it is essential beforehand to test whether age (A) or size (TLm) of the individual at the time of marking has an influence on this variable. If A or TLm does have a significant influence on IncA, which is probable in view of the deceleration of growth with age or size, relative individual increments (IncR) must be compared. To this end, TLm was back-calculated from the length at recapture (TL) and the otolith radius at the time of marking (R-IncA). The regression between the length of the fish and that of the otolith, necessary for back-calculation, was linear and highly significant ($p < 0.01$):

$$(1) \quad TL = 1.163 + 26.336 \cdot R \quad (n = 908 \quad r^2 = 0.802).$$

The back-calculation formula used that of Francis (1990), based on the assumption that the length of fish and the growth of the otolith are proportional over time:

$$(2) \quad TLm = [(1.163 + 26.336 \cdot (R - IncA)) / (1.163 + 26.336 \cdot R)] \cdot TL.$$

A one-way analysis of variance (ANOVA, Scheffe range test, Statgraphics®) was carried out on IncA to test the effects of age (A) and length at the time of marking (TLm). For this ANOVA, the TLm values were grouped into 5-cm size classes and the analysis was carried out for each time period (G1O, G1J, G2J). Initial tests showed significant differences in IncA between the various age ($F_{(5,573)} = 14.72$, $p < 0.01$, for G1J) and TLm classes ($F_{(8,578)} = 21.99$, $p < 0.01$, for G1J). Moreover, a two-way ANOVA (sex and period) conducted on the length of eels at recapture (TL) showed a very significant effect of sex on TL ($F_{(2,957)} = 478.1$, $p < 0.01$), but no influence of growth period ($F_{(2,957)} = 1.6$, $p > 0.05$). The size at marking therefore had a significant effect on absolute otolith increment. To make growth comparisons between the sexes, it is therefore necessary to examine the relative otolith increment (IncR). The analysis was then conducted on the IncR values.

An analysis of covariance (ANCOVA) (multifactor ANOVA, Scheffe range test, Statgraphics®) was carried out on IncR as a function of age (A, from 1 to 3 yr), sex (U, M, F), and period of growth (G1O, G1J, G2J), with TLm as covariate. Age classes older than 3 yr had to be eliminated from this analysis because they were not represented in all growth periods. The effect of individual maturation (change to the "silver eel" stage) could not be taken into account because of the very low number of silver eels recaptured (1% for G1J and 4% for G2J). Beforehand, a test for normality of distributions and homogeneity of variances was undertaken. This showed that the distributions were more similar to a Poisson type and that the variances were slightly heterogeneous for G2J, so the ANCOVA was performed on the transformed variable, $\log(IncR + 1)$, for which the distributions were normal and the variances homogeneous. The

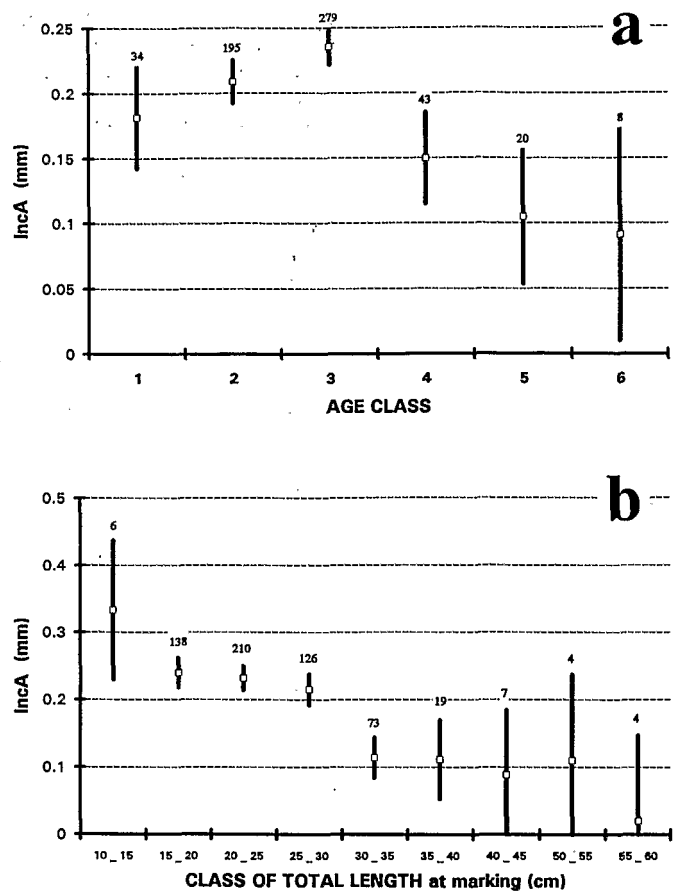


FIG. 4. Means and confidence intervals (95%) of the absolute otolith increment (IncA) for each class (G1J). Numbers above each class correspond to sample sizes. (a) Age classes; (b) total length at marking (TLm) classes.

relative importance of the various factors having a significant effect on IncR was assessed from an analysis of the centred residuals derived from the ANCOVA. To do this, the residual variances and the distributions of the residuals were examined.

Results

Absolute Growth

The absolute growth of otoliths in relation to age, sex, or growth period was extremely variable in Les Garcines pond, as shown by the high values for the coefficients of variation (CV) of IncA (Table 2). In October 89 (G1O), after 7 mo of growth, the length increment varied between 1.7 and 10.1 cm, with a mean of about 5 cm, sexually undifferentiated individuals and 2-yr-old fish having the greatest growth (Table 2). After 15 mo of growth (G1J), extreme values for length increments were from 0 to 18 cm, with a mean of 6.6 cm, and females and 3-yr-old fish had the best growth (Table 2). The growth of the otolith did slow with age (Fig. 4a) and there was a clear decrease in growth as a function of size at marking (Fig. 4b). For the G1J group, the percentage of individuals with zero growth was low: 0.4% of undifferentiated fish, 2.3% of males, and 6.5% of females, with no growth occurring particularly among the larger individuals (but not specially silver eels). Finally, the group that remained for 8 mo in the pond, over winter (G2J),

TABLE 2. Values of IncA (mm) for each class of the two factors age and sex and for each period. SD = standard deviation; CV = coefficient of variation; IncL = back-calculated length increment (cm) from mean of IncA.

	<i>n</i>	Mean	SD	CV (%)	Incl
G1O					
Age					
1	7	0.1486	0.0773	52.0	5.1
2	15	0.1587	0.0850	53.6	5.3
3	4	0.1300	0.0841	64.7	4.6
Sex					
U	14	0.1771	0.0811	45.8	5.8
M	5	0.1160	0.0792	68.3	4.2
F	17	0.1235	0.0864	69.9	4.4
G1J					
Age					
1	34	0.1815	0.0701	38.7	5.9
2	195	0.2087	0.0921	44.1	6.7
3	279	0.2355	0.0974	41.4	7.4
4	43	0.1498	0.1277	85.3	5.1
5	20	0.1050	0.1225	116.7	3.9
6	8	0.0912	0.0608	66.6	3.6
Sex					
U	251	0.2052	0.0842	41.0	6.6
M	211	0.1906	0.1038	54.5	6.2
F	134	0.2401	0.1287	53.6	7.5
G2J					
Age					
1	119	0.0610	0.0453	74.2	2.8
2	110	0.0453	0.0604	133.4	2.4
3	33	0.0673	0.1167	173.5	2.9
4	10	0.0250	0.0556	222.5	1.8
Sex					
U	151	0.0704	0.0613	86.9	3.0
M	97	0.0274	0.0373	136.1	1.9
F	43	0.0532	0.0923	173.4	2.6
Age, undifferentiated					
1	145	0.0929	0.0735	79.1	3.6
2	157	0.1827	0.0968	53.0	6.0
3	94	0.2151	0.0882	41.0	6.8
Age, male					
1	7	0.0314	0.0261	83.0	2.0
2	123	0.1129	0.1217	107.7	4.1
3	135	0.1894	0.1038	54.8	6.1
4	27	0.0807	0.0690	85.4	3.3
5	11	0.0473	0.0559	118.2	2.4
Age, female					
1	8	0.0975	0.0767	78.6	3.7
2	40	0.1362	0.1022	75.0	4.7
3	87	0.2605	0.1333	51.2	8.0
4	25	0.1772	0.1567	88.4	5.8
5	9	0.1467	0.1646	112.2	5.0
6	5	0.1040	0.0767	73.7	3.9

showed the lowest growth of 2.7 cm on average, with the greatest length increment among the undifferentiated and 3-yr-old fish (Table 2). Variability in growth was great for G2J, with a CV usually greater than 100% (Table 2).

Irrespective of the growth period, growth was always greater for individuals belonging to the younger age classes (1, 2, or 3 yr) and lower for males (Table 2). In general, variability (CV) was greater in older individuals, and undifferentiated individuals always had the lowest variability

(Table 2). Variability in growth was greater for the short growth periods (G1O and G2J) than for the longer period (G1J). In the latter group, the CV was low for the first three age classes and for the different sex categories.

The ranking of the periods in terms of growth was the same for each sex category (Table 2): growth was lowest for G2J (8 mo, including winter), highest for G1J (15 mo), and intermediate for G1O (7 mo, including summer). In each sex category, age class 3 had the highest otolith incre-

TABLE 3. ANCOVA on the effect of age (1–3 yr), sex, and period on the relative increment of otoliths (IncR) after transformation (covariate = total length at marking, TLm). SS = sum of squares; df = degrees of freedom; *effect significant ($p < 0.01$).

Source of variation	SS	df	Mean square	F-ratio	Significance level
Covariate					
TLm	0.49943	1	0.49943	1000.000	0.001*
Main effects	0.40191	6	0.06698	165.300	0.001
Age	0.03156	2	0.01578	38.941	0.001*
Sex	0.05075	2	0.02537	62.613	0.001*
Period	0.05694	2	0.02847	70.259	0.001*
Two-factor interactions	0.00670	12	5.58×10^{-4}	1.379	0.170
Age vs. sex	0.00353	5	7.05×10^{-4}	1.740	0.123
Age vs. period	0.00233	5	4.66×10^{-4}	1.151	0.332
Sex vs. period	0.00063	4	1.58×10^{-4}	0.391	0.815
Residual	0.31122	768	4.05×10^{-4}		
Total	1.21926	787			

ment and the lowest variability (Table 2). Variability in growth was lowest for all sex categories in the longest growth period (Table 2).

Factor Effects on Growth

ANCOVA on IncR (transformed as $\log[\text{IncR} + 1]$) revealed a significant effect of age, sex, and growth period ($p < 0.01$; Table 3). The covariable TLm also had a highly significant effect ($p < 0.01$) on relative otolith growth (Table 3). In addition, there were no significant first-order interactions ($p > 0.05$) between the various factors taken as pairs (Table 3).

Otolith growth is therefore influenced by the age, body length, and sex of individuals. The age classes are clearly distinct for each of these factors (Table 4): for the first three age classes, growth increases with age. There is, however, a clear reduction in growth as from age class 3 in the G1J group, in which the number of age classes goes up to six (Fig. 4a). Undifferentiated fish and females form a homogeneous group in the factor sex and grow faster than males (Table 4). The period of growth also has an influence (Table 4): for the same time interval, growth is notably greater if the period includes the summer months than if it includes winter or even the start of spring. An entire annual growth cycle tends to reduce variability within age and sex classes (Table 2).

From the experiment conducted in this study, the growth periods can be summarized by the following equation:

$$G10 + G2J = G1J$$

$$(\text{March 1989 to October 1989}) + (\text{October 1989 to June 1990}) = (\text{March 1989 to June 1990}).$$

For each sex, the mean growth in G1J should therefore be equal to the sums of the mean growths in G10 and G2J. Taking the back-calculated absolute length increments (centimetres; Table 2), the values are as follows:

$$\text{Undifferentiated: } G10 + G2J (8.8 \text{ cm}) > G1J (6.6 \text{ cm})$$

$$\text{Males: } G10 + G2J (6.1 \text{ cm}) \approx G1J (6.2 \text{ cm})$$

$$\text{Females: } G10 + G2J (7.0 \text{ cm}) \approx G1J (7.5 \text{ cm}).$$

The equality of the growth periods is fairly well respected for males and females, but not for undifferentiated fish. These values were, however, calculated for all age classes.

TABLE 4. Homogeneity of the groups in terms of the various factors used in the ANCOVA on transformed IncR (Table 3). For each factor, similar letters indicate homogeneous groups.

Factor	n	Average	Homogeneous groups
Age			
1	158	0.04228	a
2	316	0.05976	b
3	314	0.07353	c
Sex			
Male	263	0.05215	a
Undifferentiated	393	0.06600	b
Female	132	0.06818	b
Period			
G2J	258	0.02423	a
G10	26	0.05734	b
G1J	504	0.08117	c

Importance of Factors

Among the various factors having an influence on otolith growth, age, sex, and growth period seem to have qualitatively similar effects, on the basis of the ANCOVA residuals (Fig. 5) and the residual standard deviations (Table 5). Among the age classes, age class 2 has the preponderant effect on growth rate whereas females are preponderant for the factor "sex" and the longest growth period (Table 5). In order of relative importance, the longest growth period (1 yr) has the strongest effect, then the "female" sex, then the "undifferentiated" sex, followed by the summer growth period and age class 2 (residual standard deviations from Table 5). If the best growth performance is wanted for eels in Les Garcines pond, 2-yr-old females should be grown for an entire annual cycle (or at least during the summer period).

Discussion

The high rate of recapture in this experiment enabled us to use marking with tetracycline to estimate the individual growth of eel otoliths. In a previous experiment, Dekker (1986) marked 3000 eels by tetracycline injection, but

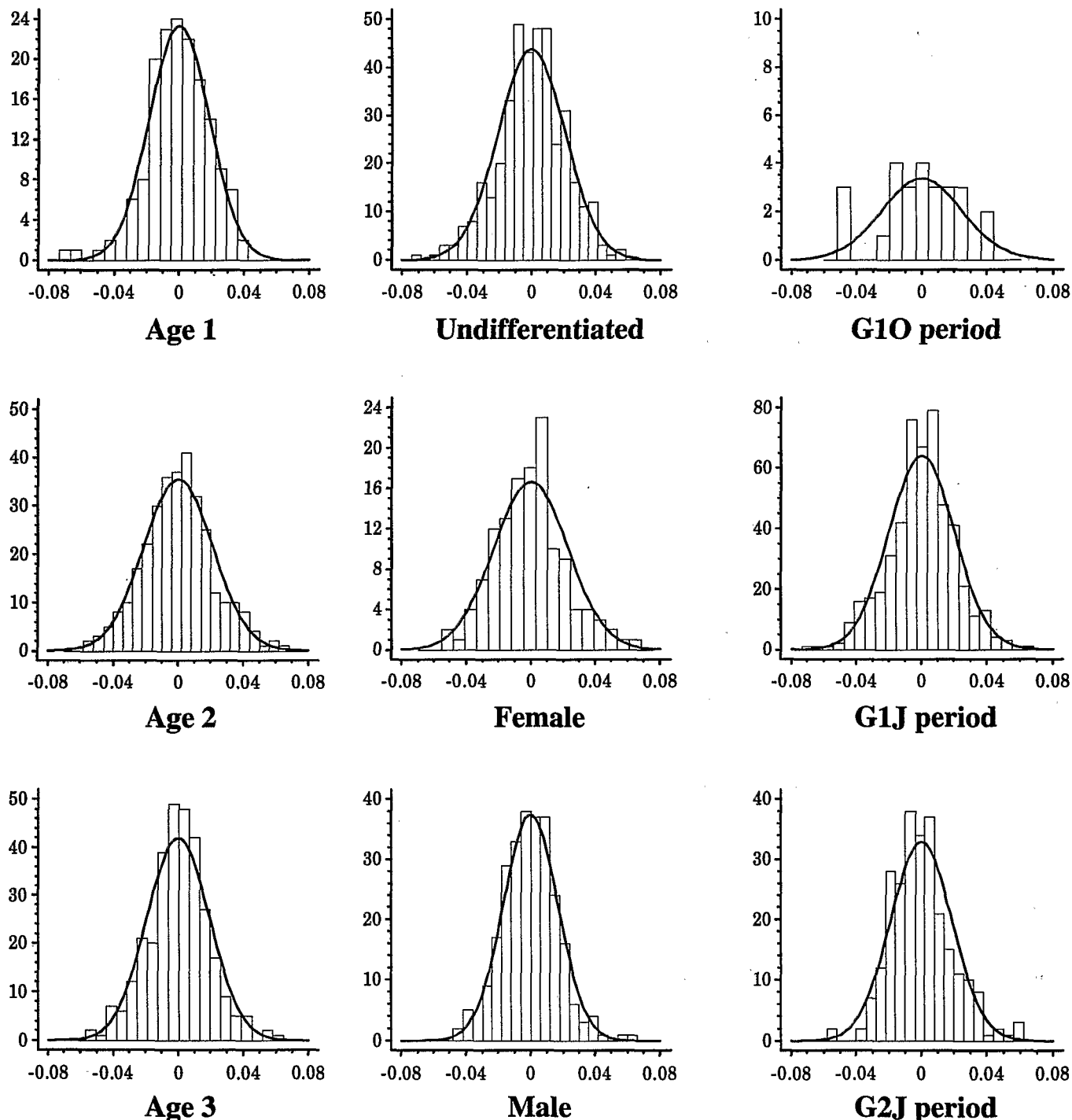


FIG. 5. Distributions (numbers of individuals) of centred residuals coming from the ANCOVA for each factor.

because of problems of recapture recovered too few individuals to be able to estimate growth. In marking experiments in which there is a high recapture rate, the main bias is that the marking itself may have an effect on subsequent growth of individuals. This bias is always difficult to detect because to do so requires an unmanipulated control group, subjected to the same conditions, but not having been manipulated beforehand. In addition, even if there is a real decrease in growth, the cause must still be determined – it could be due to the handling during marking or the inability of the fish to adapt to the new environment because of the density of

fish, food availability, etc. For example, Peters (1982) showed that dominance relationships in eels lead to stress, and it is known that high densities in pisciculture affect growth (Degani and Levanon 1983; Degani et al. 1984). The initial population of eels in Les Garcines pond was, however, low and the quantity that was then released into the pond ($50 \text{ kg}\cdot\text{ha}^{-1}$ in total) remained moderate compared with other Mediterranean lagoons (Tesch 1977; Rossi et al. 1988); the density recorded at the end of the experiment was $40.7 \text{ kg}\cdot\text{ha}^{-1}$. Finally, a comparison of the values of the condition factors (K_c) of the natural population in the pond

TABLE 5. Statistics on centred residuals coming from the ANCOVA (Table 3) for the various factors (age, sex, and period) SD = standard deviation; CI (95%) = confidence interval for mean at 95%.

Factor	<i>n</i>	Mean	SD	CI (95%)
All residuals	788	0	0.01989	$-1.3909 \times 10^{-3} \ll 1.3909 \times 10^{-3}$
Age				
1	158	0	0.01843	$-2.8972 \times 10^{-3} \ll 2.8972 \times 10^{-3}$
2	316	0	0.02133	$-2.3615 \times 10^{-3} \ll 2.3615 \times 10^{-3}$
3	314	0	0.01913	$-2.1246 \times 10^{-3} \ll 2.1246 \times 10^{-3}$
Sex				
Undifferentiated	393	0	0.02070	$-2.0530 \times 10^{-3} \ll 2.0530 \times 10^{-3}$
Female	132	0	0.02301	$-3.9621 \times 10^{-3} \ll 3.9621 \times 10^{-3}$
Male	263	0	0.01680	$-2.0405 \times 10^{-3} \ll 2.0405 \times 10^{-3}$
Period				
G10	26	0	0.02467	$-9.9669 \times 10^{-3} \ll 9.9669 \times 10^{-3}$
G1J	504	0	0.02021	$-1.7693 \times 10^{-3} \ll 1.7693 \times 10^{-3}$
G2J	258	0	0.01876	$-2.3011 \times 10^{-3} \ll 2.3011 \times 10^{-3}$

and of the marked fish showed no differences after 15 mo of growth (Panfili 1992). There appeared, therefore, to be little bias from the experimental manipulation in this study.

The overall back-calculated growth of the marked population over a cycle of slightly more than 1 yr (15 mo for G1J) was 6.6 cm, for all age classes and sizes combined. For these individuals having grown for an annual cycle, the mean growth of an age class *i* represents the growth between age *i*-1 and age *i*, and this can therefore be compared with data in the literature. The back-calculated growth in length was 6.7 cm for individuals in Les Garcines aged 2 yr (therefore, between 1 and 2 yr), 7.4 cm between 2 and 3 yr, and 5.1 cm between 3 and 4 yr. Published records of growth in length between 1 and 2 yr vary: 6.2 cm in a Spanish estuary (Arias and Drake 1985), 6.1 cm in a Portuguese lagoon (Gordo and Jorge 1991), between 4.5 and 8.4 cm in northern European rivers (Rasmussen and Therkildsen 1979; Moriarty 1983; Vøllestad and Jonsson 1988), and from 5.1 to 9.4 cm in lake environments (Berg 1985; Paulovits and Biró 1986; Nagiec and Bahnsawy 1990). The growth observed by marking in Les Garcines therefore falls within the range of growth values recorded across the whole of Europe. Our results are therefore within acceptable values and tend to show that growth was good even after marking.

There was great variability in individual otolith growth during the experimental periods, if all fish are taken into account. Age or size at marking had a pronounced influence on this growth. Although overall growth slowed with age, the differences became more pronounced after age 3 and increased in variability. A small proportion of large individuals showed no visible otolith growth even after spending 15 mo in the pond; this was not due to maturation of these individuals, since the percentage of silver eels captured remained very low. Growth was therefore rather fast in the first few years in Les Garcines. Elsewhere in Mediterranean habitats, particularly in lagoons and estuaries, and in contrast with more northerly freshwater habitats, younger fish also show rapid growth (Rossi and Villani 1980; Ardizzone and Corsi 1985; Arias and Drake 1985; Fernández-Delgado et al. 1989; Gordo and Jorge 1991).

In this experiment, only absolute otolith growth was measured and back-calculation was used to show the significant effect of size at marking on subsequent growth of the

otolith. The use of back-calculation has, however, been the subject of controversy (Small and Taylor 1987; Campana 1990; Francis 1990). The bias produced by this method can be of three types: (1) a poor relation between fish length and otolith size, (2) variation in measurements recorded due to the preparation technique used on the calcified pieces (Panfili and Ximénès 1992), and (3) a poor calculation technique (Campana 1990; Francis 1990). Although some authors have shown uncoupling between otolith and somatic growth (Mosegaard et al. 1988; Reznick et al. 1989; Secor and Dean 1989, 1992; Secor et al. 1989; Wright et al. 1990), this has only been for very short growth periods (daily cycles). It is not certain that these differences occur at the seasonal scale. In addition, the decrease in growth of the otolith with fish size is very pronounced in Les Garcines and indirectly confirms the strong relation between fish length and that of the otolith. This relation is also confirmed by the correlation coefficient of the linear regression. Estimation of somatic growth by back-calculation from otolith growth is certainly sensitive to the method of calculation used. For this reason, our comparisons have concentrated on otolith growth.

Great variability in the growth of eels has been reported on several occasions in the literature (Vøllestad 1989, 1992; Fontenelle 1991). In our experiments over all growth periods, otolith growth was strongly influenced by age and sex class. It is remarkable that the variability within classes is great, in agreement with what has previously been reported in the literature. Growth was distinctly different between the various age and sex classes, with little overlap. In this experiment, variability in growth increased with age, but the effect of age was confounded by the passage from the undifferentiated stage to the male or female categories, which would have reduced the variance of individuals whose sexual identity became apparent but not for the others. In the latter, the variability was not the same in all age classes and could reflect the presence of successive cohorts within the same year. Differences in growth between sex categories of eels have been recorded in few cases in the natural environment (Vøllestad and Jonsson 1988; Tulonen 1989), but in pisciculture, these differences are very clear (Egusa 1979). In natural populations, a pronounced difference has often been recorded in the ages and mean lengths of males and females,

the latter being larger and older (Lee 1979; Rossi 1979; Rossi and Villani 1980; Aprahamian 1986, 1988; Vøllestad and Jonsson 1988). Two hypotheses could explain why females are larger: either females reach larger sizes because they live longer or they have faster growth rates from the start and the difference in size is due both to longevity and to growth rate. This study supports the second hypothesis because the growth of females was notably greater than that of males over an annual cycle, even if age class, which would have accentuated the difference, is disregarded. In our closed population, a bias could be caused in the growth estimates of the various sex classes, since size could only increase among the undifferentiated class, as there was no recruitment of small fish in this population whereas such recruitment did occur among males and females as a result of undifferentiated fish becoming macroscopically sexually distinguishable. This maturation from a sexually undifferentiated stage to a differentiated stage can explain the inequalities in absolute growth between the periods for the undifferentiated fish.

In this Mediterranean pond, the most favorable period for growth is the spring, and especially the summer. Otolith growth in this study was slow at the start of spring, but did occur. In contrast, it is certain that winter growth was extremely slow or nonexistent. Over a long growth period of more than one annual cycle, growth variability was reduced, but was much more pronounced for periods of 7 or 8 mo. These results agree to some extent with those of Fernández-Delgado et al. (1989) on a population in a Spanish estuary, who noted that growth was fast in May, low from June to October, and zero from November to April. The most important difference between that study and ours is the fast growth in May, a phenomenon that was not recorded in Les Garcines pond, if a comparison is made of the growth in the first (G1J) and second groups (G2J). In this pond, growth is best between the end of spring (May-June) and the start of autumn (October), but smaller scale variations cannot be distinguished within this long period.

In pisciculture, the optimal temperatures for growth vary slightly from study to study but fall between 22 and 26.5°C (26.5°C, Kuhlmann 1979; 22-23°C, Sadler 1979; 26°C, Seymour 1989 and Holmgren et al. 1991). In Les Garcines pond, the temperature at the bottom exceeded 20°C from June 89 to September 89, but never exceeded 25°C except at the surface (Fig. 2). This is the period during which growth should be optimal, judging from the growth results obtained in pisciculture. This also corresponds exactly to the best period of growth recorded by marking otoliths with tetracycline. The period during the experiments when the temperature fell below 10°C was between November 89 and March 90 (Fig. 2): this corresponds closely to the very low otolith growth of marked individuals. The best period for growth in Mediterranean lagoons is therefore the summer period.

The three main factors having an effect on growth, age, sex, and season of growth, all have qualitatively similar effects. Growth is, however, always better in the summer period and/or for females and/or for young individuals. However, as sexual dimorphism does not occur in the eel, and the future sex of undifferentiated individuals cannot be determined, and is even difficult to determine macroscopically on mature individuals (Van de Wijdeven 1990), it is impossible to select females for rearing. In aquaculture, it is

also known that males stop growing when they reach a length of about 30 cm (Van de Wijdeven 1990). The fish farmer who wants to obtain the best yield should therefore opt for the 2- to 3-yr-old age classes. But the selection of fish of this age based on their length will always pose problems because of the variability in length growth in eel.

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