Resorption of scales in Atlantic salmon (*Salmo salar*) during its anadromous migration: a quantitative study

by

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Abstract. – Resorption processes in the scales of Atlantic salmon, during its anadromous migration are known since 1935, but have never been quantified. In fact, the ellipsoid form of the ascending salmon scale resorbs into an irregular highly deformed shape in spawning salmon. The aim of this study is to quantify the changes of both anterior and posterior scale areas using image analysis techniques. Salmon scales were sampled taking into account the sex and sea age of fish at two stages of migration, *viz* at the beginning of the upstream migration in rivers and at the spawning areas. Analyses confirmed that scales are highly resorbed during the anadromous migration and that the resorption is caused by the osteoclastic activity. Morphometric analyses showed that the scale posterior area eroded significantly more intensely than the anterior one. The reason behind the higher erosion in the posterior surface is probably the fact that this area is closer to the circulatory system. Whatever the sex, scales eroded because they ensure a source of minerals used during sexual maturation. However, scales of males eroded significantly more than those of females. Therefore, scale quantification could well be used to discriminate males from females especially after spawning. The time spent at sea seemed to play a minor role on scale modifications during the upstream migration in females. In the light of these quantifications, the use of eroded scales could constitute an accurate tool to improve the knowledge and the management of wild populations.

Received: 3 Dec. 2012
Accepted: 29 Apr. 2013
Editor: J.Y. Sire

Key words
Salmonidae
Salmo salar
Brittany River
Anadromous migration
Scales
Resorption
Image analysis

Résumé. – Résorption des écailles du saumon Atlantique (*Salmo salar*) au cours de sa migration anadrome : étude quantitative.

Les processus de résorption des écailles du saumon Atlantique, au cours de la migration anadrome, sont connus depuis 1935, mais n'ont jamais été quantifiés. En fait, la forme ovale de l'écaille du saumon en phase de croissance se résorbe en une forme plus ou moins asymétrique chez le saumon en fin de reproduction. Une étude quantitative de l'évolution des différentes parties de l'écaille, antérieure et postérieure, a été réalisée en utilisant des techniques d'analyse d'images. Les écailles du saumon ont été échantillonnées en tenant compte du sexe du poisson, de l'âge en mer et de deux étapes de leur migration, à savoir au début de la migration dans les rivières puis dans les zones de frai. Les analyses ont confirmé que les écailles sont fortement résorbées au cours de la migration anadrome et que la résorption est provoquée par l'activité des ostéoclastes. Les analyses morphométriques ont montré que la partie postérieure de l'écaille est significativement plus érodée que la partie antérieure. Cette érosion plus importante est probablement due au fait que le champ postérieur recouvert de l'écaille est plus proche du système circulatoire. L'érosion des écailles garantit une source de minéraux utilisés au cours de la maturation sexuelle chez les deux sexes. Cependant les écailles des saumons mâles sont significativement plus érodées que celles des femelles. Par conséquent, la morphométrie des écailles pourrait bien être utilisée pour distinguer les mâles des femelles surtout après la période de la ponte. Le temps passé en mer semble jouer un rôle mineur sur les modifications des écailles lors de la migration anadrome chez les femelles. À la lumière des résultats de ces quantifications, l'utilisation des écailles pourrait constituer un outil précis pour améliorer la connaissance et la gestion des populations sauvages.

Since the studies of Crichton (1935) and Van Someren (1937), it was known that the scales of Atlantic salmon (*Salmo salar* Linnaeus, 1758) are submitted to resorption processes during the anadromous migration of the adults before and during spawning. In fact, the ellipsoid form of the ascending salmon scale resorbs into to an irregular highly deformed shape in spawning salmon. The shape of the resulting scale, brings into mind what local fishermen call "the form of a dead man's head", in males only. It was also shown that scale resorption is closely related not only to sexual maturation but also to the energy used during the upstream migration while the animals are in a fasting stage. This resorption process results from the activities of specialised cells, the osteoclasts, and could be put into evidence by specific histochemical techniques (Persson *et al*., 1995, 1997), or by using transmission electronic microscopy (Persson *et al*., 1999). The salmon that survive the stress caused by the activities of migration and spawning return to the sea (Fontaine, 1948; Fontaine *et al*., 1950). The mean postspawning survival varies strongly according to the stock; it ranges

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between 10 and 80% for some populations of Newfoundland, New Brunswick and Labrador (Cunjak *et al.*, 1998). Survival falls to 1% for the French populations (Baglinière and Porcher, 1994) and presently to 0.6% in some French stocks (Lower-Normandy and Brittany) (Baglinière, unpubl. data). Moreover, survival is generally 4.3 times higher in females than in males (Baglinière, unpubl. data). When returning to the sea, salmon scales regenerate a part of the eroded areas, but indelible morphologic traces of the resorption process remain clearly visible (Crichton, 1935; Van Someren, 1937; Shaerer, 1992). Indeed, a spawning mark, defined as a continuous concentric line, appears on the scales and their reading allows the identification of the fish as a multispawner (Baglinière, 1985; Ombredane and Baglinière, 1992; Shaerer, 1992).

Outside of the period of sexual maturation, Atlantic salmon does not show a pronounced sexual dimorphism (Maisse and Baglinière, 1986; Maisse *et al.*, 1988; Flemming, 1996). Therefore, for determining the sex outside of the spawning season in live fish several methods have been investigated. The first one is the serodiagnostic method based on the immunological identification of vitellogenin (Le Bail and Breton, 1981; Baglinière *et al.*, 1981). This method is of constraining use but quite reliable. The second one is a morphological method based on the measure of the relative lower jawbone length (Maisse and Baglinière, 1986; Maisse *et al.*, 1988; Prévost *et al.*, 1991). These two methods can be used as early as the beginning of July for returning fish. It is generally well known that the scale resorption is significantly higher in males than in females (Baglinière, 1985). If this affirmation is justified, it would be certainly possible to separate both sexes by a simple examination of scale morphology. However, to our knowledge, no quantitative study of the scale resorption has been carried out during the anadromous migration of salmon to test this hypothesis.

The aim of the present study is to quantify the scale resorption in adult salmon trapped in the Scorff River in the south of Brittany (France). This study was achieved during the adult upstream migration from the river entry up to spawning area and throughout and/or after spawning. The present study was carried out by separating the two sexes as early as possible and the two main marine components of adult population: grilse (one sea winter 1SW) and spring salmon (multi sea winter MSW).

MATERIAL AND METHODS

Fish

Salmon were sampled from a southern Brittany river, the Scorff (03°24'07"W; 47°50'06''N), a coastal river 75 km long with a catchment area of 480 km^2. Its estuary length is about 15 km. A trapping station (up- and downstream), which operates continuously, is located in "Moulin des Princes" at the limit of tidal influence. The salmon within river distribution and spawning area range over 50 km upstream the trap. The biological characteristics (sex, sea age and migration stage) of 94 sampled salmon are presented in table I. Salmon were trapped at the Moulin des Princes station, from April to the end of July or caught at night by electrofishing on the spawning grounds during the reproduction season (end of November-end of December). They were measured (fork length) and 4-6 scales were removed for estimating age on the left hand side of the fish, 3-6 rows above the lateral line and on a line extending from the anterior edge of the anal fin to the posterior edge of the dorsal fin (Baglinière, 1985; Ombredane and Baglinière, 1992). A total of 441 scales were analysed excluding regenerated scales.

Morphometry

The scales were dried and conserved between two glass slides. Their image was digitized with an "AGFA SNAP-

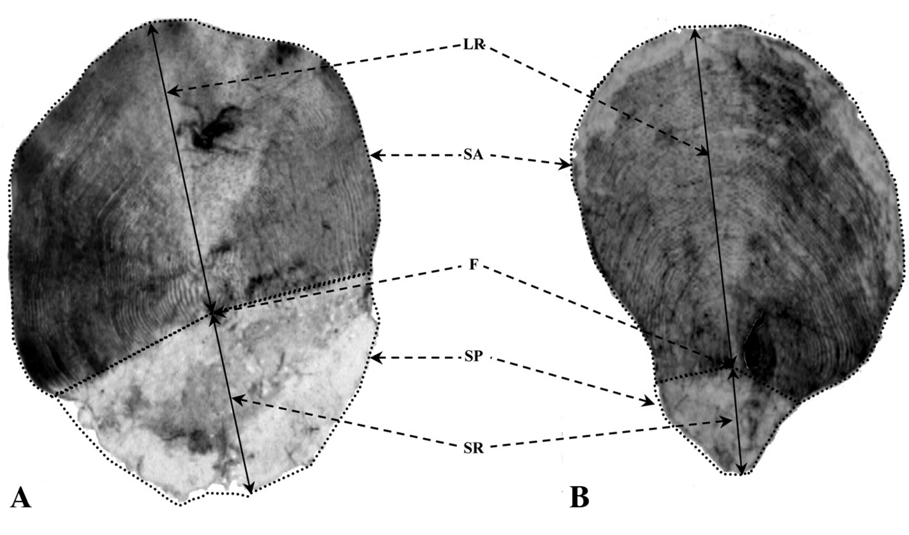

Figure 1. - Measurements taken on salmon scales. **A**: Scale of ascending spring salmon; **B**: Scale of spawning spring salmon. F: Focus; LR: long radius; SA: surface of the anterior field of the scale; SP: surface of the posterior field of the scale; SR: small radius.

Table I. - The sample of Atlantic salmon caught in the Scorff River (Brittany).

Adult stage	Ascending fish		Spawning fish		Total
Sea age	Grilse	Spring salmon	Grilse	Spring salmon	
Males	11	9	7	12	39
Females	11	14	11	19	55
Total	45		49		94

SCAN 600" scanner and then analysed by the NIH Image 1.61/fat software. Scale analyses included the following measurements (Fig. 1): surface of the anterior field of the scale or overlapped field (**SA**); surface of the posterior field of the scale or overlapping field (**SP**); total surface (**ST**) obtained by adding the two surfaces: **SA + SP**; main (or "long") radius (**LR**) corresponding to the maximal length of overlapped field (distance focus-anterior scale margin); and small radius (**SR**), corresponding to the maximal length of overlapping field (distance focus-posterior scale margin).

In order to analyse the temporal variation of these parameters, different ratios were calculated: **RE1**: total area of the scale related to the square fork length: **ST / FL^2 (%)**; **RE2**: surface of the overlapped field related to the total surface of the scale: **SA / ST(%)**; **RE3**: small radius related to the long radius: **SR / LR (%)**.

These ratios were standardized by a transformation to arcsines (Sokal and Rohlf, 1981) and tested by ANOVA (Statview 4.02) in order to analyse the influence of sex, sea age and adult stage (ascending fish sampled during upstream migration and spawning fish sampled during spawning season). For the purpose of visualizing the distribution of the different salmon groups, a Principal Component Analysis (PCA) was carried out on the experimental data as a function of quantitative and qualitative variables, using the XLStat software (Addinsoft SARL, New York, NY).

Morphology

Some scales were cleaned with a diluted solution of hypochlorite in order to remove the cellular remains, dehydrated in ethanol of increasing concentration, dried and settled upon a metallic support. Their external side was metallised with a "gold-platinum" layer and observed with a JEOL-SEM-35 electron microscope to examine the state of scale surface.

RESULTS

The scales of the Atlantic salmon are cycloid elasmoid scales with two areas: The anterior overlapped field and the posterior overlapping field. They are oval shaped, thin, transparent and composed of a basal plate of lamellar tissue (see Meunier, 1987, for further details), which is overlaid by a well-mineralized superficial layer (= external layer) (Zylberberg *et al*., 1992). The focus of the scale is clearly decentred and driven backward (Fig. 1). At low magnification the scale surface clearly showed that material is absorbed from the margin of the scale and also relatively frequently on the median area (Fig. 1).

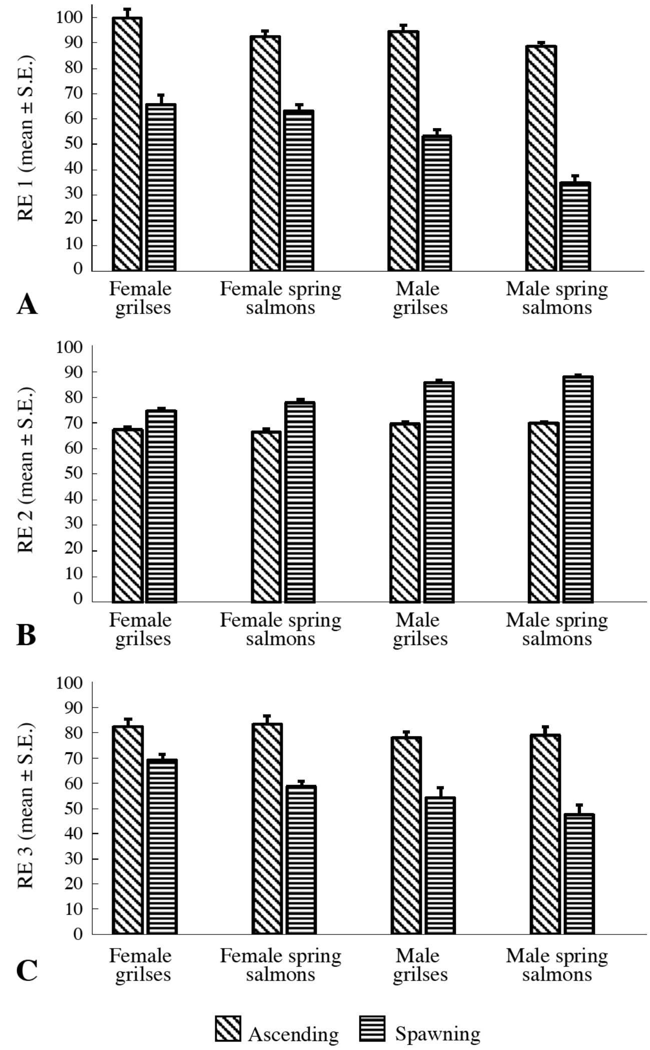

Figure 2. - Variations of salmon scale ratios according to sex, sea age (grilse or spring salmon) and migration stage (ascending or spawning). **A**: RE1 ratio (total scale surface / fork length squared). **B**: RE2 ratio (anterior field surface / total scale surface). **C**: RE3 ratio (small radius / long radius of scale). SE = standard error.

The differences between migrating and spawning fish

The RE1 ratio decreased significantly ($p < 0,001$) between the upstream and spawning (Fig. 2A). The reduction of the total scale surface illustrates an important erosion of the two layers (superficial layer and basal plate). The decreasing RE1 ratio depends significantly on sex rather than sea age ($p < 0,001$) and is less pronounced in females than in males.

Table II. - Results of variance analysis (ANOVA) of the RE1, RE2 and RE3 ratio in salmon, according to sex, migration stage and sea age. RE1: total area of the scale in relation to the square of fork length; RE2: surface of the overlapped field in relation to the total surface of the scale; RE3: the small radius in relation to the long radius.

	Sex (Male / Female)			Stage of migration (Ascending / Spawning Salmon)			Sea age (Grilse / Spring salmon)		
Variables	F (1, 433)	P	Test	F (1, 433)	P	Test	F (1, 433)	P	Test
RE1	9.9	0.0026	S	254.6	< 0.0001	S	3.2	0.08	NS
RE2	86.7	< 0.0001	S	212.2	< 0.0001	S	0.65	0.02	S
RE3	12.2	0.02	S	97.1	< 0.0001	S	4.2	0.02	S

The RE2 ratio in ascending salmon is significantly lower than in spawning salmon ($p < 0.0001$; Tab. II; Fig. 2B). This shows that the relative surface of the posterior and anterior fields decreases at the same time but not with the same speed of resorption. In the same way, the RE3 ratio decreases significantly ($p < 0.05$; Tab. II) indicating that the erosion is highly important at the overlapping field of the scale (Fig. 2C). The evolution of these two ratios during the upstream migration is always significantly influenced by sex and sea age (Tab. II). Thus, in the spawning salmon, the erosion of the overlapping field is significantly more important in males than in females, which developed a lower resorption process. Furthermore, at the spawning stage, the RE2 ratio is significantly higher in spring salmon than in grilse ($p < 0.05$) (Fig. 2B; Tab. II).

Principal component analysis of salmon scale data revealed that the adult stage was clearly separated along PC1, which explained 51.84% of variance in the data (Fig. 3). Salmon at the beginning of the migration were positively loaded along PC1. Grilse and spring salmon were further separated along PC2 (27.11% explained variance), while males and females were distributed on both sides of the axis PC3 (Fig. 4).

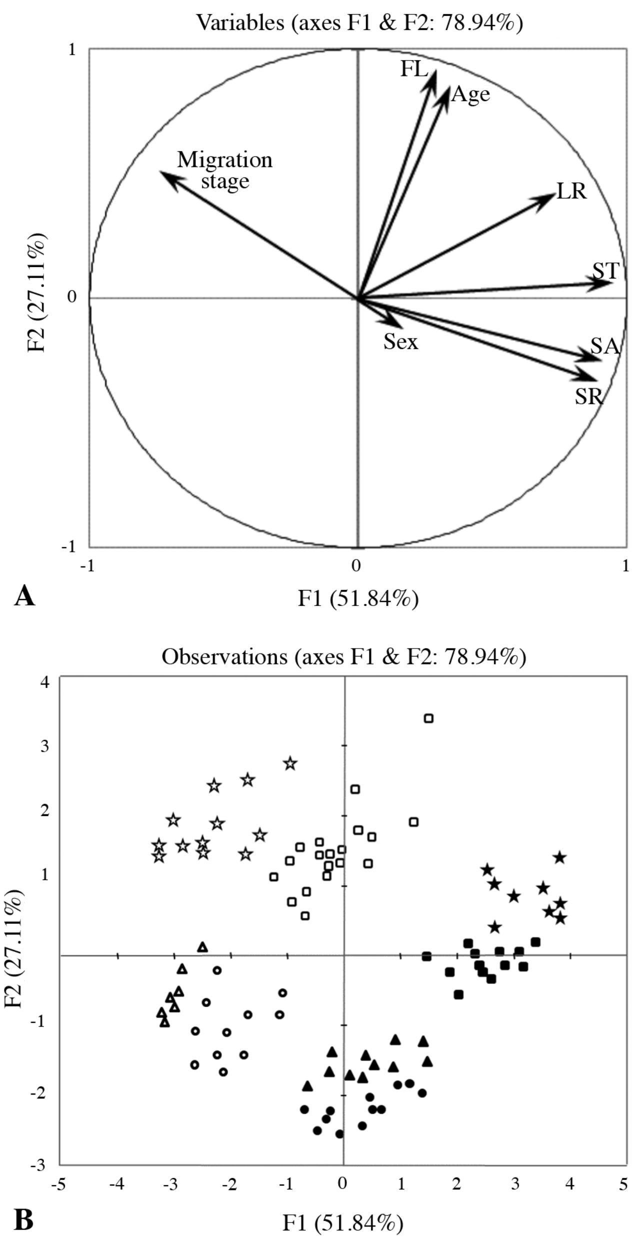

Figure 3. - Results of the principal components analysis (PCA) of the data regarding the Plots of variable loadings (**A**) and observation scores (**B**) according to the first two axes. ● Female grilse at the beginning of migration, ○ Female grilse at the spawning area, ■ Female spring salmon at the beginning of migration, □ Female spring salmon at the spawning area, ▲ Male grilse at the beginning of migration, △ Male grilse at the spawning area, ★ Male spring salmon at the beginning of migration, ☆ Male spring salmon at the spawning area. (FL = Fork length; LR = Long radius; SA = Surface of the anterior field of the scale; SR = Small radius; ST = Total surface of the scale).

Morphological characteristics

SEM (Scanning Electron Microscopy) observation of salmon scales at the final phase of upstream migration highlights the rough aspect of the scale surface; the circuli are divided up and numerous superficial alveolae can be seen. This irregular organization confirms the presence of resorbed areas in the focus region but mostly at the scale margin (Fig. 5A). These resorption areas directly result from the destruction of the external layer and part of the basal plate. According to the large numbers of alveolae on the whole scale surface (Fig. 5B), the resorption seems to result preferentially from osteoclastic activity. Scale resorption concerns

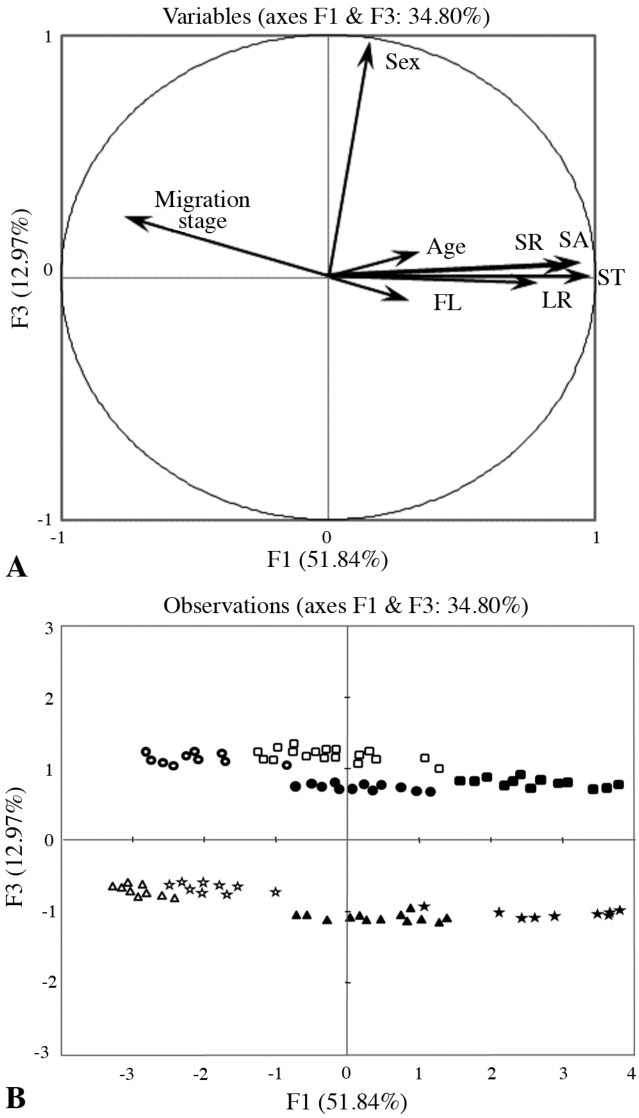

Figure 4. - Results of the principal components analysis of the data regarding the Plots of variables loadings (**A**) and observations scores (**B**), according to the first and the third axis. ● Female grilse at the beginning of migration, ○ Female grilse at the spawning area spawning area, ■ Female spring salmon at the beginning of migration, □ Female spring salmon at the spawning area, ▲ Male grilse at the beginning of migration, △ Male grilse at the spawning area, ★ Male spring salmon at the beginning of migration, ☆ Male spring salmon at the spawning area. (FL = Fork length; LR = Long radius; SA = Surface of the anterior field of the scale; SR = Small radius; ST = Total surface of the scale).

at first the external layer and then expands more or less in the upper part of the basal plate.

DISCUSSION

Salmonid scales as well as other teleost scales are composed of mineralized tissue deposited by scleroblasts, but in contrast to mammalian bone, the cells are not incorporated in the tissues (Ouchi *et al*., 1972 ; Persson *et al*., 1995). However, osteoclastic cells (osteoclasts), which are quite similar to the multinucleated cells observed in mammalian bone, were described in several mineralized tissues of teleosts and notably in the scales (Sire *et al*., 1990). These clastic cells may frequently be mononucleated in a number of taxa (Witten and Huysseune, 2009). The observation of Howship lacunae, at both levels of the external layer and of the basal plate, shows that the osteoclasts are present in the salmon scales. The alveolae have a variable depth and their morphology is characteristic of the Howship lacunae that housed together multi-nucleated and mononucleated osteoclasts probably. Like in other salmonids, scale resorption starts by a demineralization process then rapidly continues by a proteolytic destruction of the organic matrix (Persson *et al*., 1999).

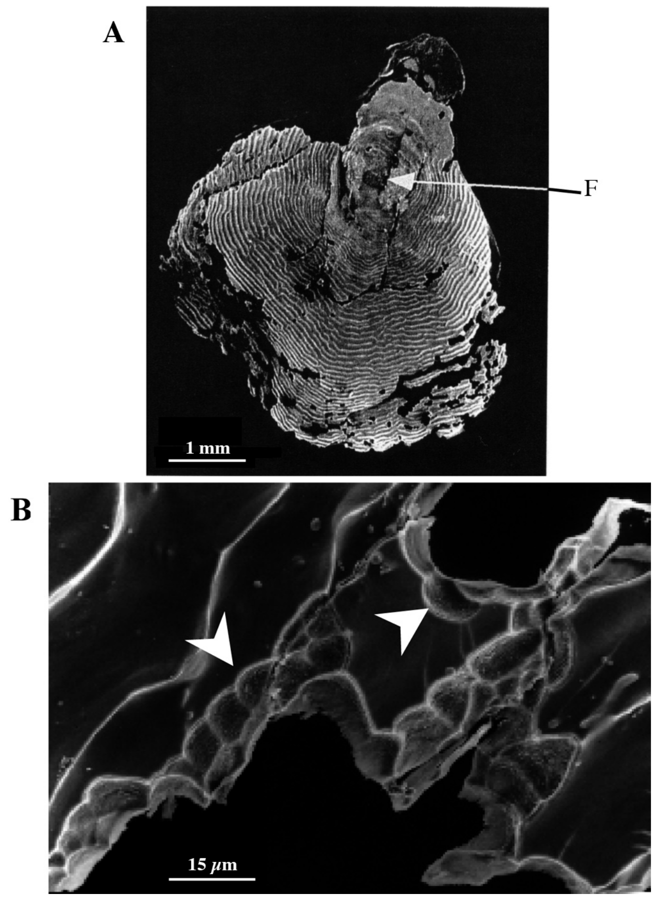

Figure 5. - Scanning electron microscopy. **A**: View of a spawning male scale (F = focus); **B**: Detail of the anterior field of the same scale showing some Howship's lacunae (arrows), which are evidence for osteoclastic resorption.

According to Simkiss (1974), in teleost fish the scales can represent an important pool of calcium (up to 20% of the total calcium weight of the organism). For many authors, only the scales would play a role as a reservoir of calcium in teleosts (Mugiya and Watabe, 1977; Carragher and Sumpter, 1991; Persson, 1997). However, recent studies have pointed to other mineral resources. Thus, the skeleton can be solicited preferentially to provide calcium and phosphorus in certain circumstances. Phosphorus is an essential element for a harmonious development and growth of the skeleton in fish (Roy *et al.*, 2002; Witten and Huysseune, 2009). In fact, according to the ecological and biological constraints both minerals (calcium and phosphorus) may act either separately or in synergy on the skeletal mineral store using various resorption processes of bone: Osteoclasy (mono- and/or multinucleated osteoclasts (Lopez, 1973; Takagi and Yamada, 1993; Persson *et al.*, 1995, 1997, 1999; Kacem *et al.*, 1998), periosteocytic osteolysis (Kacem and Meunier, 2000) or halastasy (Lopez *et al.*, 1970; Lopez, 1973; Kacem and Meunier, 2003; Sbaihi *et al.*, 2007, 2009). Beside the mineral supply scale resorption as well as vertebral bone resorption deliver more or less complex organic component that can be used in other metabolic mechanisms of the organism. For example, collagenous derivatives are useful for the synthesis of new bone for the kype in the male (Witten and Hall, 2003; Witten *et al.*, 2004; Kacem and Meunier, 2009). So, scales make up a mineral and organic reservoir that can be used in case of necessity. In salmonids, the sexual maturation requires a great quantity of calcium (Persson *et al.*, 1997). Generally, calcium mobilization is often obtained from external environment sources (food and water) and also from an increased removal of calcium from the mineralized elements (bones and scales) if there are not enough external resources (Lopez, 1973; Simkiss, 1974; Takagi and Hamada, 1993; Sbaihi *et al.*, 2007, 2009). Moreover, adult salmon do not feed from the moment they enter the river and migrate upstream in freshwater that is a poor mineral environment compared to the ocean. In these conditions, it is obvious that the organic matter and the minerals (particularly calcium) should be essentially drawn from internal sources and notably from scales.

The present quantitative study confirmed the high resorption of scales usually observed qualitatively in salmonids during the anadromous migration in males as well as in females (Crichton, 1935; Järvi and Menzies, 1936; Van Someren, 1937; Baglinière, 1985;Ouchi *et al.*, 1972; Takagi, 1990; Persson *et al.*, 1997). The resorption of the superficial layer and the basal plate of scales contributes clearly to the calcium supply. During the sexual maturation period, that takes place during upstream migration, the increasing plasmatic level of calcium, which is linked to the vitellogenesis transport and to the quantity of calcium to store into the ovocytes, induces a high demand for this mineral in females (Wallace, 1985; Carragher and Sumpter, 1991; Persson, 1997). Therefore, the scales indirectly play a crucial role in the development of the ovocytes in female salmon by transferring and then accumulating the vitellin mineral reserves, which will be used later by the embryos. In males, the calcium demand is significantly much lower (Witten and Hall, 2002).

In males, the level of calcium accumulated in the testicles is quite reduced relatively to that accumulated in the ovaries (Persson *et al.*, 1997) suggesting that the need of calcium is lesser during the gonadic maturation of males contrary to the females. Nevertheless, in mature male adult salmon (genera *Salmo* and *Oncorhynchus*), this demand is rather linked to the growth of the kype, a pronounced hypertrophy of the jaws in the shape of “beak" (Fontaine, 1948; Fontaine *et al.*, 1950; Fleming, 1996; Witten and Hall, 2002, 2003; Witten *et al.*, 2005; Kacem and Meunier, 2009) which certainly requires large amounts of calcium. Moreover, in the Atlantic salmon, kype morphogenesis is accompanied by an important remodelling of bones and cartilages in the anterior region of the skull, essentially the ethmoidal zone (Tchernavin, 1938a; 1944), and the appearance of new teeth: the breeding teeth (Tchernavin, 1938b). This morphogenetic phenomenon is taking place at the same time as scales are resorbed. This suggests that kype osteogenesis in males is occurring at the expense of scales. However, the Pacific salmon, *Oncorhynchus masu*, shows an important resorption of the scales during the genital maturation, while it does not undergo the hypertrophy of the jaws (Ouchi *et al.*, 1972; Persson, 1997; Persson *et al.*, 1997). Thus, the resorption of scales in adult salmon males could be rather under the control of sexual hormones (testosterone, corticosteroids) (Idler, 1973; Lall and Lewis-McCrea, 2007).

The analysis of surface variation of the overlapped (anterior) and the overlapping (posterior) fields illustrate that the latter is more resorbed during the anadromous migration. This preferential resorption of the overlapping field could be explained by the presence of bloody vessels close to this part of the scale. Indeed, a rich vascular network may exist in the longitudinal sections of the skin in the loose dermis region (Whitear, 1986). The presence of osteoclasts in this area (Persson *et al.*, 1999), and the proximity of the inner part of the scale to the vascular system, would thus favour a more important resorption in the overlapping field. The overlapped field is settled deeply into the dense dermis where the circulatory system is more diffuse, and the scale surface is better protected by the surrounding fibrous tissue, which is essentially collagenous.

Otherwise, the mineral density of the elasmoid scales is significantly higher in the superficial layer than in the basal plate (Meunier, 1984). The resorption efficiency of the superficial layer is thus above that of the basal plate, which is incompletely mineralized in salmonids (Maekawa and Yamada, 1970) as in most teleost scales (Zylberberg *et al.*, 1992).

This mineral supply of scales in the calcium metabolism could be added to the mineral generated by the vertebral skeleton that is also clearly requested during the upstream migration in salmon (Meunier and Desse, 1978; Kacem and Meunier, 2000, 2003; Kacem *et al*., 1998, 2000).

The sexual dimorphism affecting scale resorption rate in spawning salmon could have important practical applications, especially in rivers where fishing is allowed in late season. Indeed, salmon angling occurs in early autumn in most of salmon rivers in Lower-Normandy and Brittany and fishermen have to declare their catches and to give fish characteristics and scale samples. The declarations rarely include sex information. Thus, the results obtained in the present study (quantification of erosion rate in scales) can provide a useful tool to determine the sex and know the eventual sexual selectivity of this fishery at this period of the year.

A second application of this quantitative approach of measurement of erosion rate in scales might allow restoring the initial form of the scale at the beginning of upstream migration. Using image analysis this restoration might allow estimating the sea age notably in spring/summer salmon as the strong erosion can remove up to two annual growth marks (Shearer, 1992).

A latter application of this study would concern the possible positive relationship between the erosion rate and the mortality rate observed after spawning. In other words, the estimation of the erosion rate in scales that vary according to sex (established results), to sea age, and to entry period in the river, might allow forecasting the survival rate of salmon after the spawning in a given stream and/a given year. To perform this application, scales from adult salmon captured and tagged during freshwater migration will be compared to scales from same fish recovered dead after spawning or during their downstream migration to sea as mended-kelts.

Acknowledgements. – We would like to thank Ms Hélène Francillon-Vieillot for her scientific advices, Mrs Nicolas Jeannot (INRA Experimental Unit Rennes) for his help to sample fish and Ms Christiane Chancogne-Weber for her kind technical help in the MEB.

REFERENCES

BAGLINIÈRE J.L., 1985. - La détermination de l’âge par scalimétrie chez le saumon atlantique (*Salmo salar* L.) dans son aire de répartition méridionale : utilisation pratique et difficultés de la méthode. *Bull. Fr. Pêche Piscic.*, 298: 69-109.

BAGLINIÈRE J.L. & PORCHER J.P., 1994. - Caractéristiques des stocks de reproducteurs et comportement lors de la migration génésique. *In :* Le Saumon atlantique, Biologie et Gestion de la Ressource (Guéguen J.C. & Prouzet P., eds), pp. 101-122. Brest: IFREMER.

BAGLINIÈRE J.L., LE BAIL P.Y. & MAISSE G., 1981. - Détection des femelles de salmonidés en vitellogenèse. 2 - Un exemple d’application : recensement dans la population de truite commune (*Salmo trutta* L.) d’une rivière de Bretagne sud (Le Scorff). *Bull. Fr. Pêche Piscic.*, 283 : 89-95.

CARRAGHER J.F. & SUMPTER P.S., 1991. - The mobilization of calcium from calcified tissues of rainbow trout (*Oncorhynchus mykiss*) induced to synthesize vitellogenin. *Comp. Biochem. Physiol.*, 99A(1-2): 169-172.

CRICHTON M.I., 1935. - Scale-absorption in salmon and sea trout. *Fish. Board Scotl. Salmon Fish.*, 4: 1-8.

CUNJAK R.A., PROWSE T.D. & PARRISH D.L., 1998. - Atlantic salmon (*Salmo salar*) in winter : “the season of parr discontent”? *Can. J. Fish. Aquat. Sci.*, 55(Suppl. 1): 161-180.

FLEMING I.A., 1996. - Reproductive strategies of Atlantic salmon: ecology and evolution. *Rev. Fish Biol. Fish.*, 6: 376-416.

FONTAINE M., 1948. - La physiologie du saumon. 1ère partie. *Ann. Stn. Cent. Hydrobiol. Appl.*, 2: 152-183.

FONTAINE M., CALLAMAND O. & VIBERT R., 1950. - La physiologie du saumon. 2e partie. *Ann. Stn. Cent. Hydrobiol. Appl.*, 3: 15-26.

IDLER D.R., 1973. Hormones in the life of the Atlantic salmon. *In:* International Atlantic Symposium (Smith M.W. & Carter W.M., eds), pp. 43-54. Fredericton: Unipress.

JÄRVI T.H. & MENZIES W. J. M., 1936. - The interpretation of the zones on the scales of salmon, sea trout and brown trout. *Rapp. P.-V. Reun.*, 47: 5-63.

KACEM A. & MEUNIER F.J., 2000. - Mise en évidence de l’ostéolyse périostéocytaire vertébrale chez le saumon atlantique *Salmo salar* L. (Salmonidae, Teleostei), au cours de sa migration anadrome. *Cybium*, 24: 105-112.

KACEM A. & MEUNIER F.J., 2003. - Halastatic demineralization in the vertebrae of Atlantic salmon, during their spawning migration. *J. Fish Biol.*, 63(5): 1122-1130.

KACEM A. & MEUNIER F.J., 2009. - Study of the transformations of the texture and the mineralization of the dentary bone in the Atlantic salmon, *Salmo salar* L., (Teleostei, Salmonidae), during genital maturation. *Cybium*, 33(1): 61-72.

KACEM A., MEUNIER F.J. & BAGLINIÈRE J.L., 1998. - A quantitative study of morphological change in the skeleton of *Salmo salar* during its anadromous migration. *J. Fish Biol.*, 53(5): 1096-1109.

KACEM A., GUSTAFSSON S. & MEUNIER F.J., 2000. - Demineralization of the vertebral skeleton in Atlantic salmon *Salmo salar* L. during spawning migration. *Comp. Biochem. Physiol.*, 125(4): 479-484.

LALL S.P. & LEWIS-MCCREA L.M., 2007. - Role of nutrients in skeletal metabolism and pathology in fish – An overview. *Aquaculture*, 267(1-4): 3-19.

LE BAIL P.Y. & BRETON B., 1981. - Rapid determination of the sex of puberal salmonid fish by technique of immunoagglutination. *Aquaculture*, 22: 367-375.

LOPEZ E., 1973. - Étude morphologique et physiologique de l’os cellulaire des poissons téléostéens. *Mém. Mus. Hist. Nat.*, 80: 1-90.

LOPEZ, E., LEE, H. S. & BAUD, C. A. 1970. - Étude histophysique de l’os d’un téléostéen, *Anguilla anguilla* L. au cours d’une hypercalcémie provoquée par la maturation expérimentale. *C. R. Acad. Sci.*, Paris, 270: 2015-2017.

MAEKAWA H. & YAMADA J. 1970. - Some histological and fine structure aspects of growing scales of the rainbow trout. *Bull. Fac. Fish. Hokkaido Univ.*, 21: 70-78.

MAISSE G. & BAGLINIÈRE J.L., 1986. - Le sexage morphologique du saumon atlantique (*Salmo salar* L.). *Bull. Fr. Pêche Piscic.*, 300: 13-18.

MAISSE G., BAGLINIÈRE J.L., LANCHY G., CARON F. & ROULEAU A. 1988. - L'identification externe du sexe chez le saumon atlantique (*Salmo salar* L.). *Can. J. Zool.,* 66(10): 2312-2315.

MEUNIER F.J., 1984. - Étude de la minéralisation de l'os chez les Téléostéens à l'aide de la microradiographie quantitative. Résultats préliminaires. *Cybium*, 8(3): 43-49.

MEUNIER F.J., 1987. - Os cellulaire, os acellulaire et tissus dérivés chez les Ostéichthyens : les phénomènes de l'acellularisation et de la perte de minéralisation. *Ann. Biol.,* 26: 201-233.

MEUNIER F.J. & DESSE G. 1978. - Interprétation histologique de la "métamorphose radiographique" des vertèbres caudales du saumon (*Salmo salar* L.) lors de sa remontée en eau douce. *Bull. Fr. Piscic.,* 271: 33-39.

MUGIYA Y. & WATABE N., 1977. - Studies on fish scale formation and resorption-II. Effect of estradiol on calcium homeostasis and skeletal tissue resorption in the Goldfish, *Carassius auratus*, and the Killifish, *Fundulus heteroclitus*. *Comp. Biochem. Physiol.*; 57A : 197-202.

OMBREDANE D. & BAGLINIÈRE J.L. 1992. - Les écailles et leurs utilisations en écologie. *In*: Tissus durs et Âge individuel des Vertébrés (Baglinière J.L., Castanet J., Conand F. & Meunier F., eds), pp. 151-192. Colloques et Séminaires ORSTOM-INRA.

OUCHI K., YAMADA J. & KOSAKA S., 1972. - On the resorption of scales and associated cells in precocious male parr of the *Masu* salmon (*Oncorhynchus masu*). *Bull. Jpn. Soc. Sci. Fish,* 38: 423-430.

PERSSON P., 1997. - Calcium regulation during sexual maturation of female Salmonids: Estradiol 17ß and calcified tissues. Ph.D. thesis, 42 p. Göteborg Univ., Sweden.

PERSSON P., TAKAGI Y. & BJÖRNSSON B.T., 1995. - Tartrate resistant acid phosphatase as a marker for scale resorption in rainbow trout, *Oncorhynchus mykiss*: Effects of estradiol-17β treatment and refeeding. *Fish Physiol. Biochem.,* 14(4): 329-339.

PERSSON P., JOHANNSON S.H., TAKAGI Y. & BJÖRNSSON B.T., 1997. - Estradiol 17β and nutritional status affect calcium balance, scales and bone resorption, and bone formation in rainbow trout, *Oncorhynchus mykiss*. *J. Comp. Physiol.,* 167: 468-473.

PERSSON P., BJÖRNSSON B.T. & TAKAGI Y. 1999. - Characterization of morphology and physiology actions of scales osteoclasts in rainbow trout. *J. Fish Biol.,* 54: 669-684.

PREVOST E., VAUCLIN V., BAGLINIÈRE J.L., BRANA-VIGIL F. & NICIEZA A.G., 1991. - Application d'une méthode de détermination externe du sexe chez le saumon atlantique (*Salmo salar* L.) dans les rivières des Asturies. *Bull. Fr. Pêche Piscic.,* 323: 149-159.

ROY P.K., WITTEN P.E., HALL B.K. & LALL S.P., 2002. - Effects of dietary phosphorus on bone growth and mineralisation of vertebrae in haddock (*Melanogrammus aeglefinus* L.). *Fish Physiol. Biochem.,* 27: 35-48.

SBAIHI M., KACEM A., AROUA S, BALOCHE S., ROUSSEAU K., LOPEZ E., MEUNIER F.J. & DUFOUR S., 2007. - Thyroid hormone-induced demineralization of the vertebral skeleton of the eel, *Anguilla anguilla*. *Gen. Comp. Endocrinol.*, 151: 98-107.

SBAIHI M., ROUSSEAU K., BALOCHE S., MEUNIER F., FOUCHEREAU-PERON M. & DUFOUR S., 2009. - Cortisol mobilizes mineral stores from vertebral skeleton in the European eel: an ancestral origin for glucocorticoid-induced osteoporosis? *J. Endocrinol.*, 201: 241-252.

SHEARER W.M., 1992. - Atlantic Salmon Scale Reading Guidelines. ICES Cooperative research report 188. Copenhagen.

SIMKISS K., 1974. - Calcium metabolism of fish in relation to ageing. *In*: Ageing of Fish (Bagenal T.B., ed.), pp. 1-12. England: Unwin Brothers Ltd.

SIRE J.Y., HUYSSEUNE A. & MEUNIER F.J., 1990. - Osteoclasts in teleost fish: Light and electron-microscopical observations. *Cell Tissue Res.*, 260: 85-94.

SOKAL R.R. & ROHLF F.J., 1981. - Biometry. *In*: The Principles and Practices of Statistics in Biological Research (Freeman W.H., ed), pp. 859. San Francisco.

TAKAGI Y., 1990. - Studies on the dynamics of bone and scale metabolism in teleost, with special reference to their calcium homeostasis. Ph.D. thesis. Hokkaido Univ., Hokkaido.

TAKAGI Y. & HAMADA J., 1993. - Changes in metabolism of acellular bone in Tilapia, *Oreochromis niloticus*, during deficiency and subsequent repletion of calcium. *Comp. Biochem. Physiol.,* 105A: 459-462.

TCHERNAVIN V., 1938a. - The absorption of bones in the skull of salmon during their migration to the river. *Fish. Board Scotl.: Salmon Fish.,* 6: 1-4.

TCHERNAVIN V., 1938b. - Changes in the salmon skull. *Trans. Zool. Soc. Lond.,* 24: 103-184.

TCHERNAVIN V., 1944. - The breeding characters of salmon in relation to their size. *Proc. Zool. Soc. Lond.,* 113: 206-232.

VAN SOMEREN V.D., 1937. - A preliminary investigation into the causes of scale absorption in Salmon (*Salmo salar* L.). *Fish. Board Scotl., Salmon Fish.,* 11: 1-11.

WALLACE R.A., 1985. - Vitellogenesis and oocyte growth in non mammalian vertebrates. *In*: Developmental Biology (Browder L.W., ed.) pp. 127-176. New York: Plenum Publishing Corporation.

WHITEAR M., 1986. - The skin of fishes including cyclostomes. *In*: Biology of the Integument: Vertebrates (Bereaiter-Hahn J., Matoltsy A. & Richards K.S., eds) pp. 8-38. Berlin: Springer-Verlag.

WITTEN P. E. & HALL B.K., 2002. - Differentiation and growth of kype skeletal tissues in anadromous male Atlantic Salmon (*Salmo salar*). *Int. J. Dev. Biol.,* 46: 719-730.

WITTEN P.E. & HALL B.K., 2003. - Seasonal changes in the lower jaw skeleton in male Atlantic salmon (*Salmo salar* L.): remodeling and regression of the kype after spawning. *J. Anat.,* 203: 435-450.

WITTEN P.E., HALL B.K. & HUYSSEUNE A., 2005. - Are breeding teeth in Atlantic Salmon a component of the drastic alterations of the oral facial skeleton? *Arch. Oral Biol.,* 50: 213-217.

WITTEN P.E. & HUYSSEUNE A., 2009. - A comparative view on mechanisms and functions of skeletal remodeling in teleost fish, with special emphasis on osteoclasts and their function. *Biol. Rev.,* 84: 315-346.

WITTEN P.E., ROSENTHAL H. & HALL B.K., 2004. - Mechanisms and consequences of the formation of a kype (hook) on the lower jaw of male Atlantic salmon (*Salmo salar* L.). *Mitt. Hambg. Zool. Mus. Inst.,* 101: 149-156.

ZYLBERBERG L., GÉRAUDIE J., MEUNIER F.J. & SIRE J.Y., 1992. - Biomineralization in the integumental skeleton of the living lower vertebrates. *In*: Bone (Hall B. K., ed.), Vol. 4, pp. 171-224. Boca Raton: CRC Press.