

Age, Growth and Distribution of the Antarctic Fish *Chaenocephalus Aceratus* Based on Otoliths

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Abstract: The *Chaenocephalus aceratus* were sampled in summers between 1979 and 1990. Their otoliths show pattern of daily microincrements as otoliths of similar species—*Pseudochaenichthys georgianus* and fishes both temperate and tropical waters. Changes of the microincrements growth pattern and otolith shape are described in relation to larval, hatching and metamorphosis stages have similar patterns for *Ps. georgianus* and *Champscephalus gunnari*, reflecting similar habitats in their early life. Width of larvae-postlarvae daily increments are: $(1.0-1.6) \times 10^{-3}$ SSI, $(1.8-2.8) \times 10^{-3}$ SGI and $(1.5-2.4) \times 10^{-3}$ ANI. They were search and chosen automatically from density profiles of otolith tissues. Tissues were prepared by new saving time and materials way—one time up to 40 ones per microscopic glass. Age of fish estimated from microincrements was alike to that inferred from the body length distribution, otoliths mass (age (years) = $140.82 \text{ OM (g)} + 0.8546$) and otolith shape changes—large in length and surface on medial plane. Those parameters create age groups of close neighbours. They have different environments. *C. aceratus* due to its adaptation to cold water, attain rapidly large body according to a growth equation: $L_t = 75.1 \times (1 - e^{-0.26(t - 0.51)})$. Among large fishes there were only females.

Key words: *Chaenocephalus aceratus*, otolith shape, age of icefish, Antarctic fish.

1. Introduction

Age determination of individual fishes can provide basic information about fish ecology such as population structures or changes in population growth due to environmental changes. The problems of ageing Antarctic fish *Channichthyidae* are commonly known [1-4], they have not scales and their bones undergo constant and large reduction [5]. In published papers, it deals with age of *C. aceratus* based on growth of otolith in some stated age up to 12 years. However, North [6] found that otoliths of icefish have sequences of 4 marks—complete seasonal marks per year (not as 2 marks per year). Since that, all age estimates based on yearly marks should have half a year less. Kompowski [7] former age reader for this species, estimate the year increments from number of hills (personal information).

The main aim of this study was obtaining data on *C. aceratus* age from otoliths and catch observation and comparing them to similar species. The changes in otolith shape were linked to life stages.

C. aceratus is a cold adapted fish living exclusively in Antarctic waters on the shelf of Bouvier, South Georgia, South Sandwich, South Orkneys, South Shetlands and Palmer Archipelago in range of depth from 5 m to more than 770 m. Its different life history stages live at different mean depths and distance from the shore, as their diet varied for them [8], and their time development are likely to be broadly synchronized with the life cycles of their food species. So, that young hatch and develop during period elevated production [9]. At South Georgia, egg (4.4 mm in diameter) laid demersal inshore (to 240 m depth) in late summer from February or March to May and at Elephant I. from May to June. Larvae (after 1-2 month development in egg [8, 10]) hatch at about 11-17 mm TL during four months in winter from June up to spring—November [10]. Hatched larvae create

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large shoals within 5 km of the coast of the island in sublittoral waters (less than 40 m depth) [6]. They survive winter (large yolk 50% of body mass) by feeding on the overwintering stages of neritic copepods. After winter (in spring in September and summer), numerous fish larvae feed on immature copepods (abundance which were linked with phytoplankton high level productions in summer: from November to February). Larvae have a better muscle arrangement for cruise swimming than the adults, in which slow muscles are only a minor component of the trunk muscles [9, 10]. The larva *C. aceratus*—15-40 mm SL, grow fast with growth rate about 0.11-0.16 mm SL per day and after 6-7 months in January, could be as 8 cm postlarvae. Postlarval and juveniles' forms, in Jan., 18 cm TL (21 g) were lived in the pelagic zone often with krill [6, 10, 11]. Inshore juvenile in Jan., 26-38 cm TL (90-350 g) feed mostly on decapods shrimps and fish [8]. The adults 45 cm TL (641 g) usually occupy the deepest waters [10] further from the shore fed mainly upon mysids and fish [11]. The feeding behaviours as well as habitat varied during their life cycle. Seashore, which they display a high level of nocturnal activity, linked to as much of the benthos, is also more active at night. Offshore feeding intensity of adults is probably highest during the day as was found for *N. rossi* by Linkowski and Rembiszewski [12] with assumption of Burchett and other [8].

2. Material and Methods

Samples of *C. aceratus* were collected during summer's cruises (Dec.-Feb.) in the vicinity of the Antarctic Peninsula in 1979 and in the South Georgia area in 1986 and 1990 (Fig. 1).

The ship investigations, stations and haul jobs were described in Fig. 2 [13-15].

Measurements of *C. aceratus* like in the case of other consumption species included: total length (cm), weight (g), sex, maturity, stomach contents and otolith subtraction (Figs. 3 and 4).

The larvae of *C. aceratus* and other species were

found in the commercially travelled net.

The ages were determined from daily increments counted in otoliths (Figs. 5-7) and from the ontogeny changes seen in otolith morphometry synchronizing with changes of the environment conditions (Fig. 6).

Daily increments of otolith linked with daily changes of fish activity in feeding intensity (Fig. 8).



Fig. 1 Cumberland bay of south Georgia.



Fig. 2 Subsamples were taken from the catch into bascets, larvae were found on the deck.

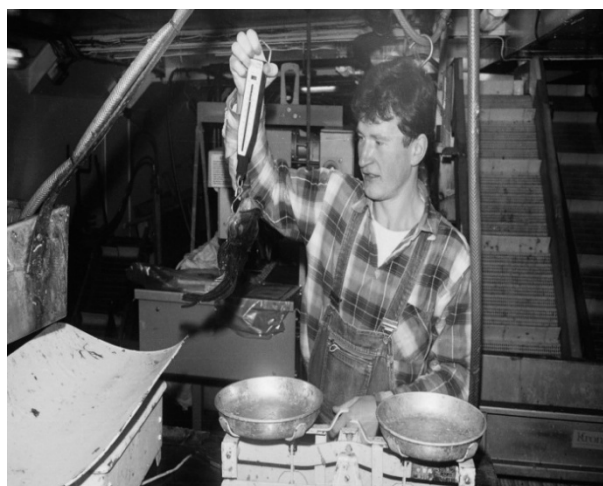


Fig. 3 Part fish data were collected personally by author (during 2 cruises).



Fig. 4 *Champocephalus aceratus*, 58 cm TL, during collecting data: length, weight, sex and maturity.

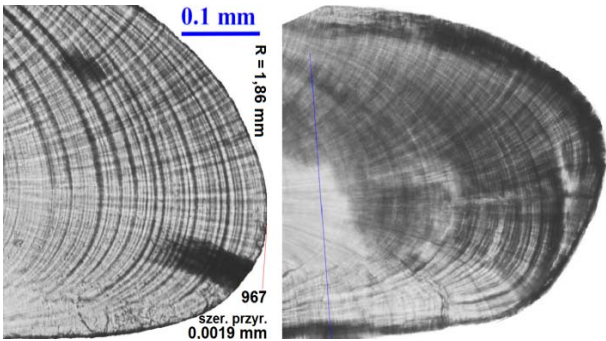


Fig. 5 Daily increments from the otolith dorsal edge, 16 discontinues in dorsal edge from old fish.

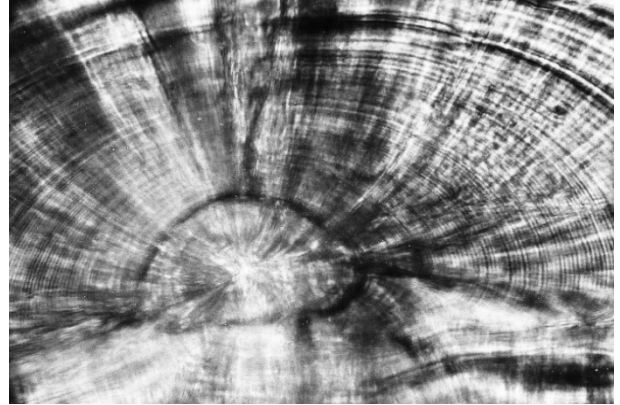


Fig. 6 Daily increments, central primordium CP and larval nucleus LN in the central part of otolith.



Fig. 7 Daily increments from the middle part of otolith dorsal radius.

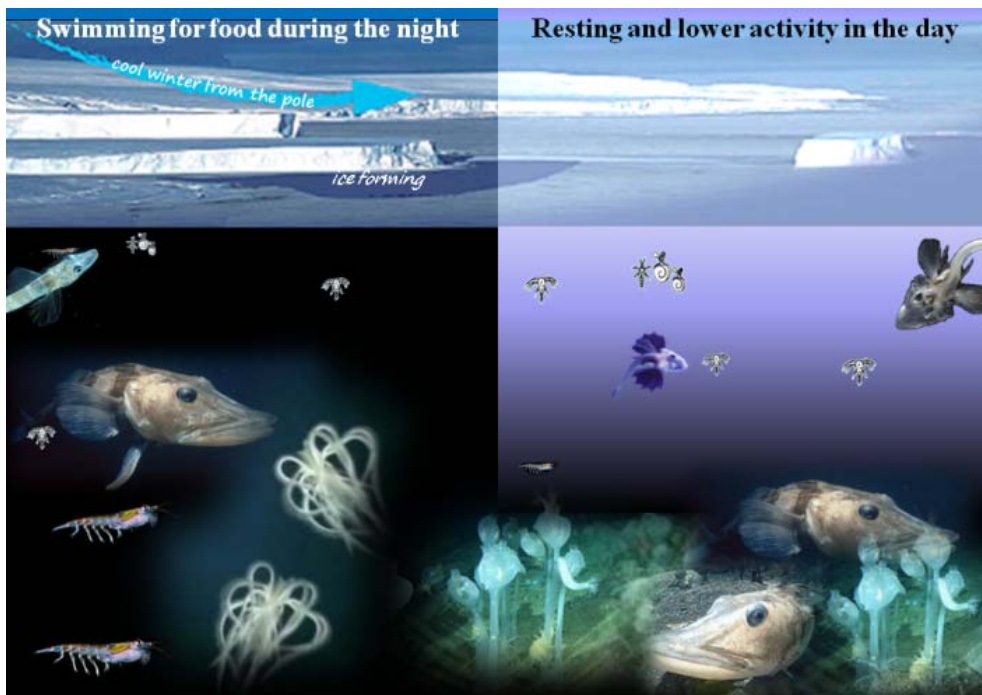


Fig. 8 For seashore *Champocephalus aceratus* preying in the night daily cycle result from locomotor activities.

Otolith—each sagittal pair prior to age reading in the laboratory was cleaned with a clorox (5.25% sodium hypochlorite), washed with water, dried, weighted (accuracy ± 0.001 mg) and measured. Selected otoliths were arranged along piece of wood and embedded in epoxy resin and cut through the nucleus mostly along the transverse plane (Fig. 9) (few larval on median sagittal plane (Fig. 10)) with a saw.

They were grounded on both sides using carborundum paper (No. 400-800) and polished with a 1 μ m diamond compound by hand and grinding machine under water to the thickness of 0.1-0.15 mm (Fig. 11).

Sagittal transverse sections obtained in this way were mounted in Eukitt (microscopic mounting medium) directly on microscopic glass, about 40 otoliths slides per glass (Fig. 12).

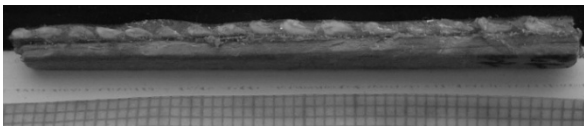


Fig. 9 Otolith along bar of wood in epoxy resin.

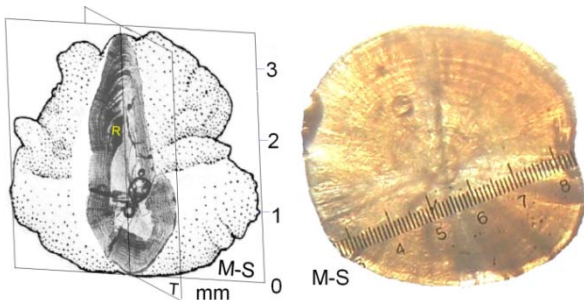


Fig. 10 Transverse (T) and median sagittal (M-S) planes.



Fig. 11 Sagittal transverse slides arranged along bar wood.

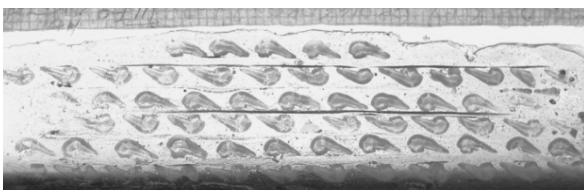


Fig. 12 They were arranged in 40 slides per glass.

Polished section to change side was released from Eukitt with chloroform or xylene. To reveal incremental patterns, they were etched for 1-8 min with EDTA. This was on both sides for *C. aceratus* because their microincrements were visible in light microscope only when the both sides of otolith slides were etched. They were cleaned with water and dried. The etched surface was pressed into acetone soaked acetate sheets paper to produce acetate replicas. Etched otolith surfaces and their acetate replicas were coated with a gold to the nano-thickness using an electric arc current at an angle of 45° in a vacuum: 9.4×10^{-8} Tr. Coated otolith sections and their acetate replicas glued on microscopic glasses were mounted on stubs (Fig. 13).

Coated daily increments were examined by scanning microscope (SEM) (Fig. 14).

Growth pattern of microincrements were automatically recorded as an intensity of transmitted light along growth radii and processed by the programs for measurements of images (Fig. 15).

After optimization of the contrast, extraction and resetting the background, to the density measurements

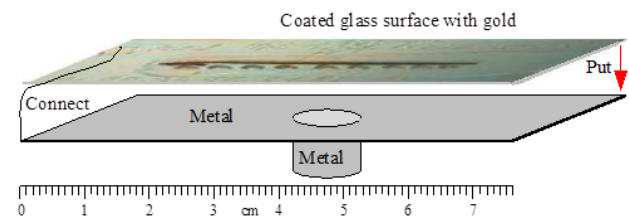


Fig. 13 Coated otolith and acetate replicas glued on glasses were mounted on SEM stubs.

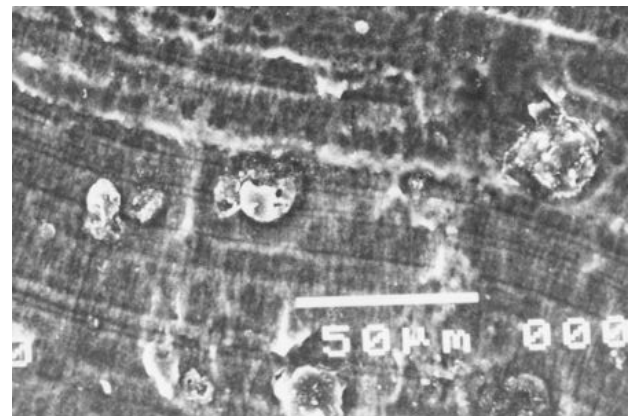


Fig. 14 Coated acetate replicas of daily increments (SEM).

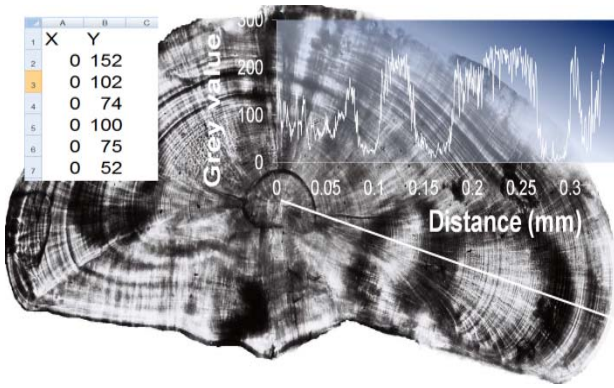


Fig. 15 Daily increment were recorded as intensity of transmitted light along growth radii (white line) and processed by programs (graphic line of from recording and their numerical data).

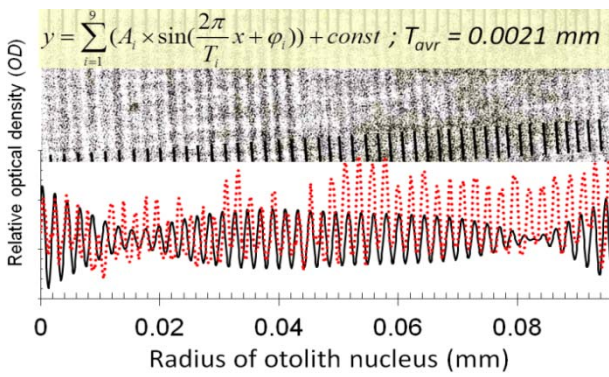


Fig. 16 Fitting sine wave to density profile and choose daily component.

a sine wave was adjusts from which component of daily increments was selected (Fig. 16).

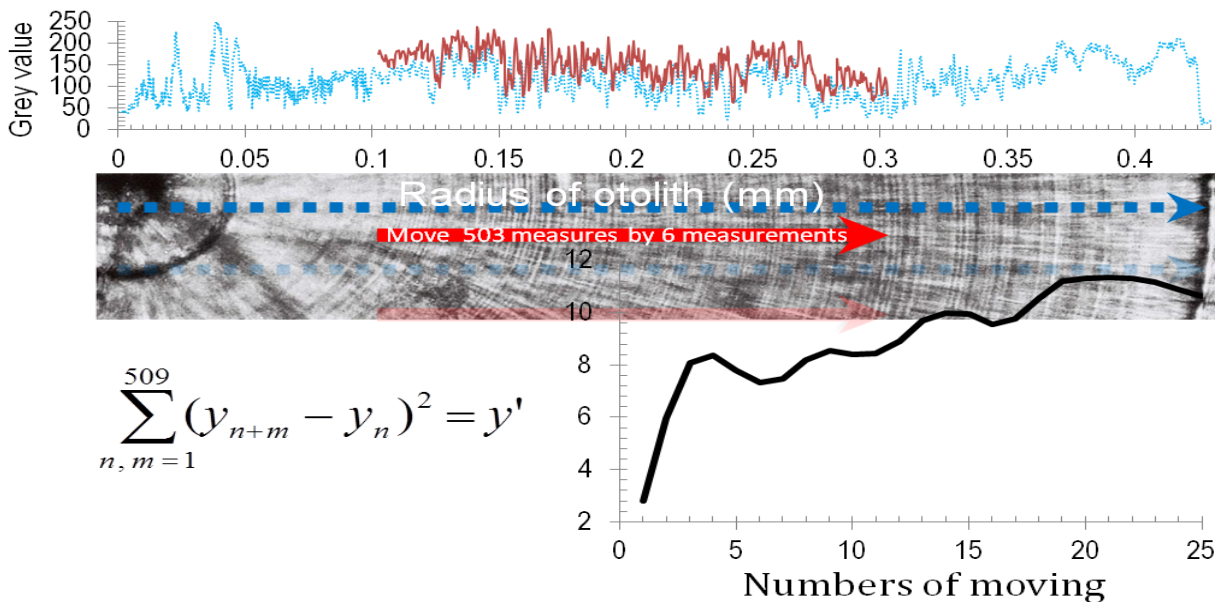


Fig. 17 X_n: 503 duplicated measurements (0.2 mm) and moved by 6 measures = 0.0024 mm gave the first drop of squared difference.

In the second method, density profile were moved along a copy until receive the minimum from squared differences. Displacement is the daily increment (average with close proximity) (Fig. 17).

3. Results

3.1 Daily Increments

Otolith of *C. aceratus* shows daily microincrements, manifest conspicuous growth pattern related to larval, hatching and metamorphosis—important changes in their life, following yearly sequences [16]. Developed embryonic inner ear of fish is indicated by Central Primordium (CP)—otolith primer, small ball with the radius of 0.0076-0.0125 mm, contain inner 3-7 microincrements that may reflect embryonic developments stages. Hatching that start free swimming of 1.5 cm SL yolk sac larvae in water is indicated by the hatching mark (surrounding Larval Nucleus—LN). This mark is first dark wide ring near narrow ones of 1.17 μm. LN has radius 0.037-0.066 mm from the centre (CP), contain 24-58 daily increments (Fig. 18). It has high deposition of aragonite shown by EDTA. Separating age group is from seasonal cycle of fish growth and development (Fig. 19).

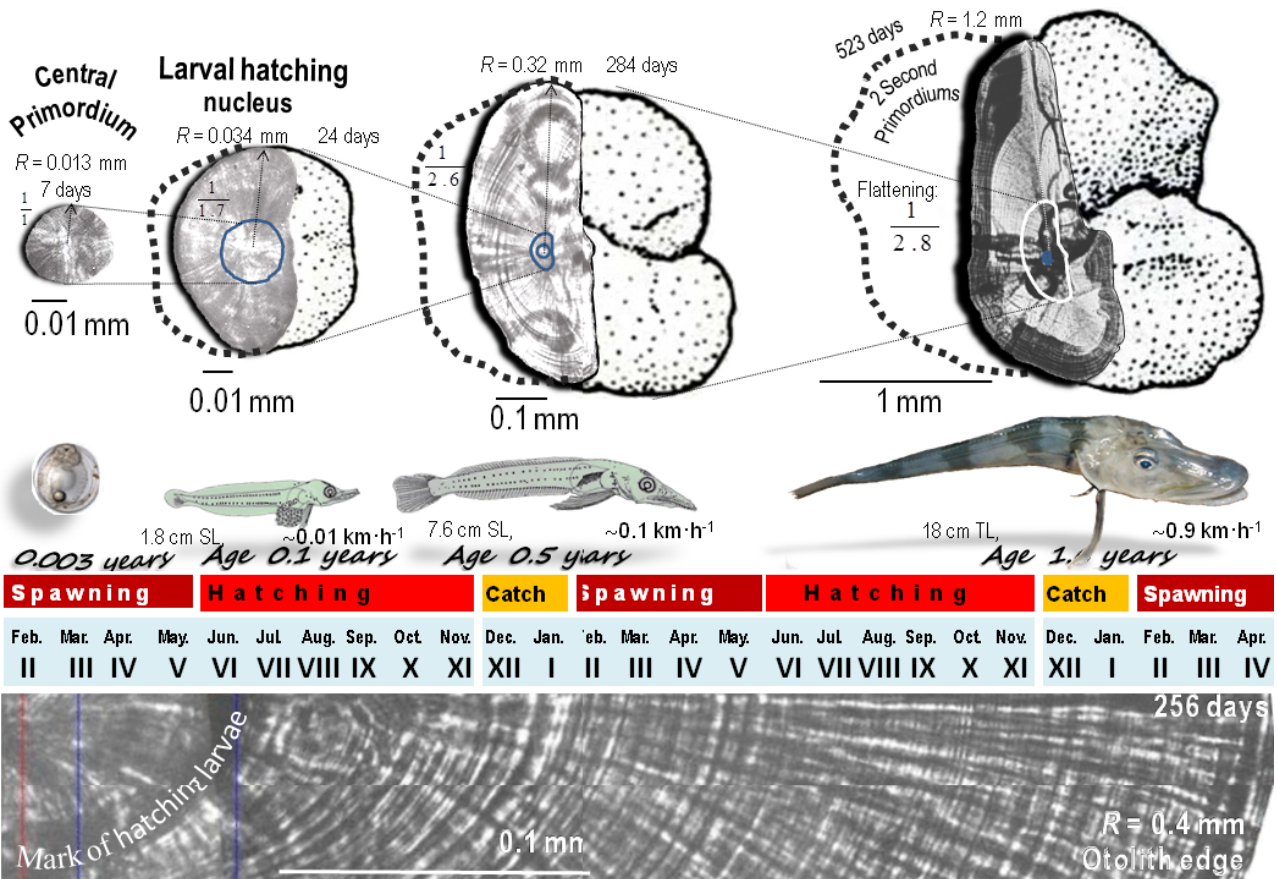


Fig. 18 In July, 1.8 cm larvae after hatch shift from the eggs to the marine water, what indicate mark in otolith having a higher content of aragonite, larger 7 cm postlarvae caught in the summer in March live in coastal waters within 5 km from shore, next year fish 18 cm shift to deeper water, what indicate two Second Primordiums (SP—second growth centres) in dorsal part of otolith, that fish perform vertical migrations for krill what indicates an increase SP with about 0.6 mm more.

Hatching of the larvae generation ends in November and further hatching run next year between June-November. Next generation appears with previous hatched larvae, which is 10-12 cm larger now. This time-space distance separate first 4 age groups which have piks into frequency of TL and of otolith size and development marks (LN, SP).

Zero age group of 4-9 cm TL post larvae have otolith 0.3 mg, with radius equal to 0.32 mm and 0.48 mm conteing 242-284 daily increments (Fig. 18). I age group of 17-23 cm TL, juvenile have about 5 mg otoliths with radius OR = 1.218 mm and with about 523 daily increments. II age group: 23-39 cm TL juvenile have 10 mg otolith, 1.81 mm OR and about 957 daily increments. III age group of 34 cm TL fish: have 15 mg otolith, large 3.94 mm OL and less than 1,450 increments.

IV and V age groups of large fish 46 cm and 55 cm TL have OM 25 mg and 35 mg, contain over 1,450 daily increments (Fig. 19).

Daily increments resolve age problem [2, 3] show linear relationship with otolith mass (OM) well to set up age from it (Fig. 19): Age (years) = 0.8546 + 140.82 × OM (g). Equation confirms linear dependence show by otolith mass distribution and body length distribution (Fig. 19). They growth with Bertalanffy parameters: $K = 0.28$, $L_{\infty} = 75.0$, $t_0 = 0.51$. In 1990 and 1992, growth of fish remains similar (Fig. 20).

Despite the sex differences, the growth of both sexes can explain by equation. *C. aceratus* obtain about 19 cm, 27 cm, 34 cm, 45 cm and 55 cm TL in I, II, III, IV and V age group. Large fish were only females about 60 cm, 64 cm and 67 cm TL in V, VI and VII+ age group.

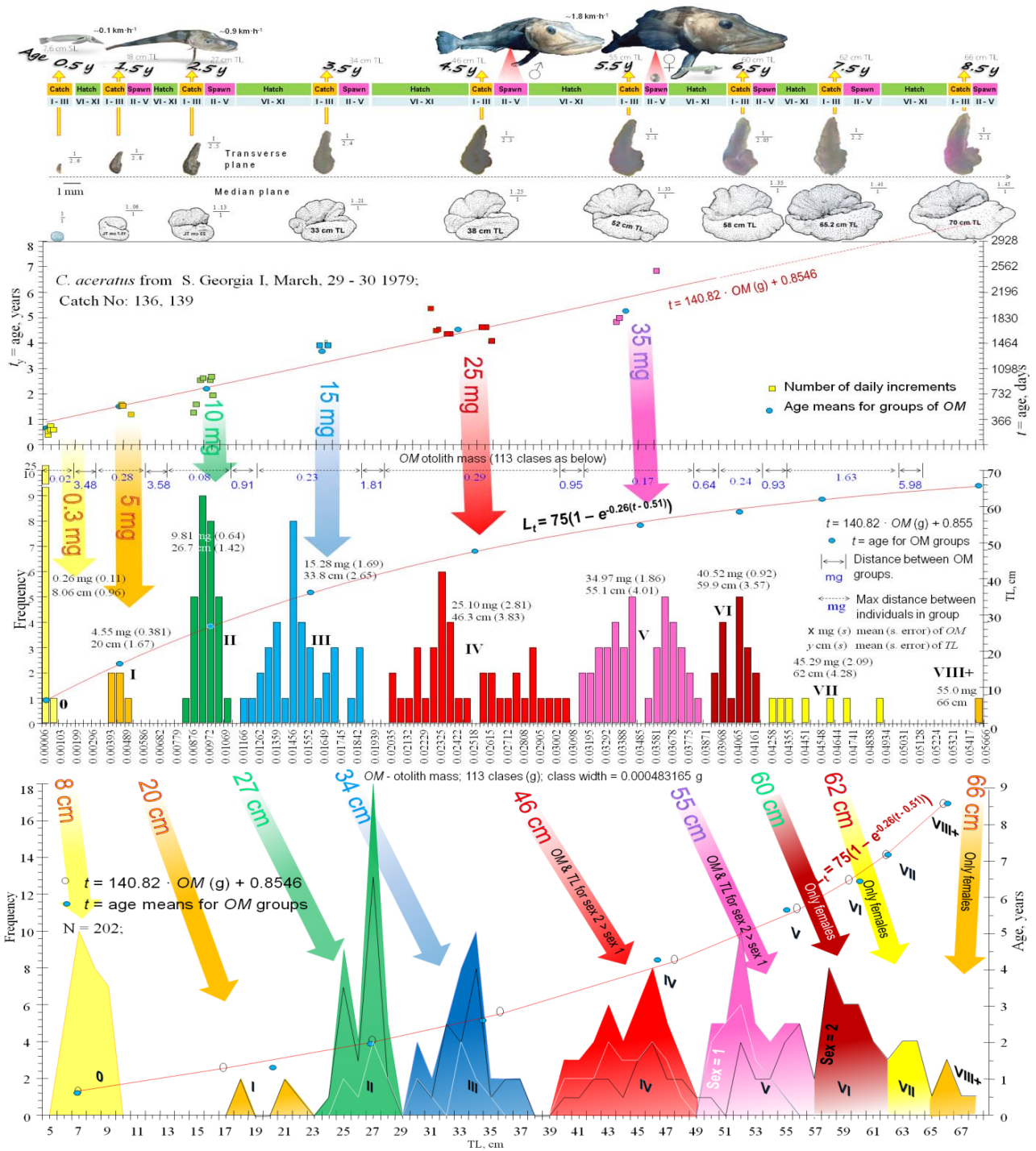


Fig. 19 Getting otolith mass (OM) of age groups and their Total Lengths. Older age groups of *C. aceratus* have larger otolith length OL than otolith height (OH), so their age get traditionally along OH may have errors, but their OM frequency show old age groups having neighbors' distances 10× large between than inside the group. ♀ are large than ♂ up to 10 cm.

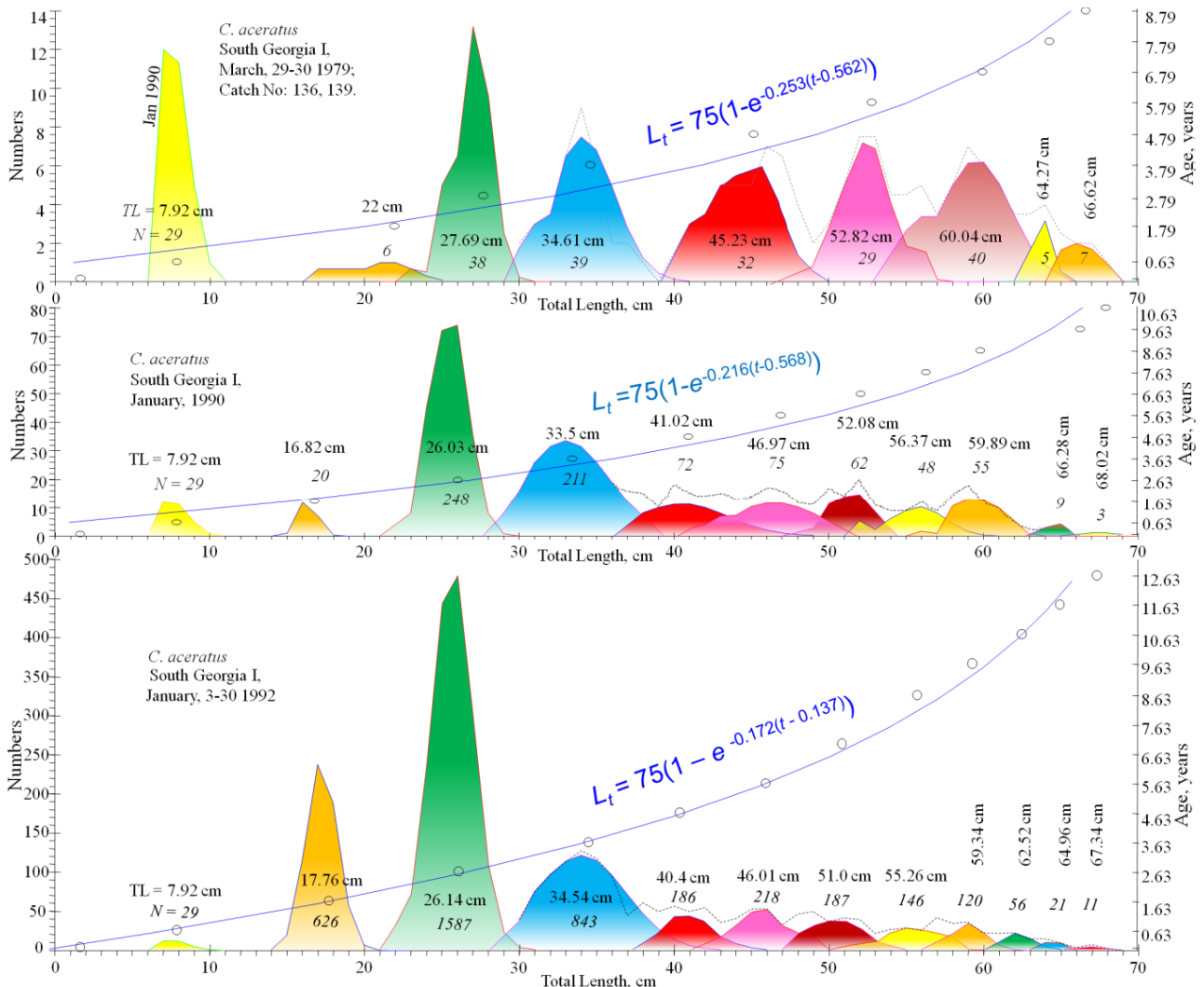


Fig. 20 Length distribution of *C. aceratus* with Bhattacharya metod, fish groups of 41 cm and 46 cm TL in January 1990 and 1992 are probably from sex differences within one age group of IV, 45 cm in reference to total length and otolith mass of fish March 1979, similar could be for next older fish groups of V, 52 cm.

Further analysis within close proximity confirmed the presence of the size, age and sex separate groups (Fig. 21).

First age TL-OM group are small females with lengths about 20 cm TL and 0.005 g of OM (Catch No. 136, ♀). Second age group are females and males with up to 30 cm TL and about 0.01 g OM. Third age group males and females have length about 35 cm, up to 40 cm and 0.015 g OM. Forth age group has about 43 cm, up to 50 cm TL and about 0.025 g OM. Fifth and older age group have males only up to 55 cm and females up to 70 cm TL. They were distinguished among 5-8 age groups by frequency of otolith mass

and TL (with additional daily increments), but measure of OM, which is very small is time consuming. Much faster and automatic is measure of otolith dimensions, give age group (Fig. 22).

Usually, the authors check age of Antarctic fish in otolith on transverse plane along growth of otolith dorsal radius. For *C. aceratus*, it work well for first 4 age group within close proximity if it is taken into account sex and catch number (Fig. 22), but it do not work for large fish because their otolith radius do not grow with the age (Fig. 22). However, area of transverse plane (TA) growth shows that otolith growth is going on for large fish, but in other dimension,

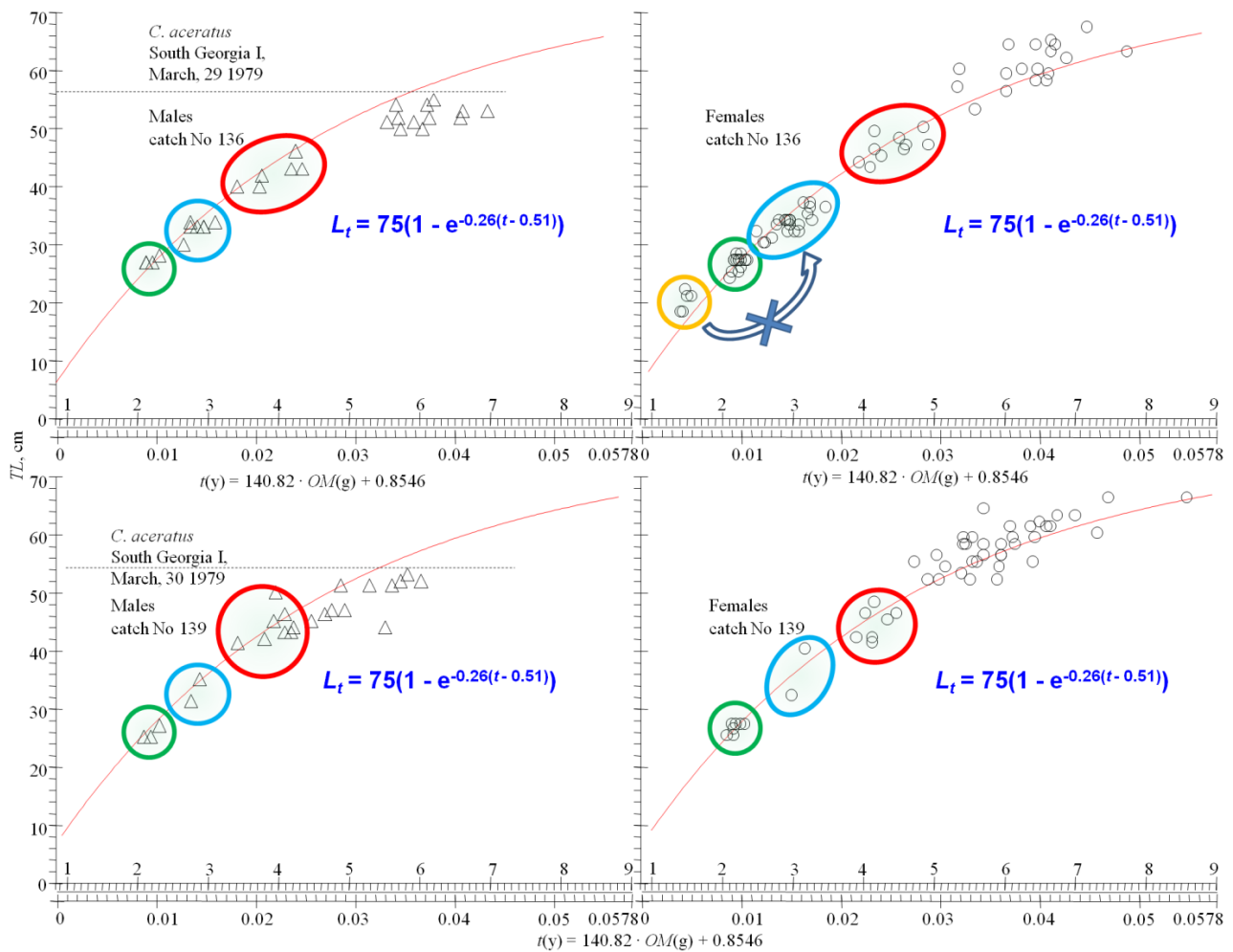


Fig. 21 Parameters of TL and OM show age groups in account of sex and side difference (The smaller group of TL–OM are only 20 cm females with otolith mass 0.005 g, next large group are 27 cm females and males with 0.01 g of OM, and the next that follow Bertalanffy equation: group 3 of 35 cm with 0.015 g of OM; group 4 of 45 cm with 0.023 g of OM; group 5 and older 52 cm females and males with 0.035 g of OM, males do not growth over 56 cm, but their OM does, females growth large in both parameters along appropriate relationship).

along otolith length. From old samples, there do not have OL—all otoliths prior to age reading, which were cut or broken on transverse planes in the past. But using transverse area the authors may predict older age of fish from equation: $Age = 0.9884 \cdot TA + 0.6318$, and next Bertalanffy growth of body parameters: $L = 75$ cm, $K = 0.2$ (Figs. 5 and 6).

Because growth radius along otolith length (R_{MAX}) for *C. aceratus* is the largest, also divide it by width of daily increment give the sample of age data to compare with age group (from closer neighbors) and for derive age—otolith parameter with TL relationship. The best is median area, next otolith length is better

than traditional otolith radius indicators of age for old fish (Fig. 23). Those have larger distances among age groups give more linear relationships (Fig. 23). Especially dorsal radius for old fish, this species is useless. This is confirmed by otolith shape factors (Fig. 24). Otolith elongations, plus depended from otolith length growth constantly with the age and total length. It is minus depended from otolith height showing inappropriate otolith radius for age estimate. This also shows dropping down circularity of otolith.

Use median area to age estimate give similar growth of fish as to ones from otolith mass (Fig. 25).

Constant growth of the otolith elongation has 2

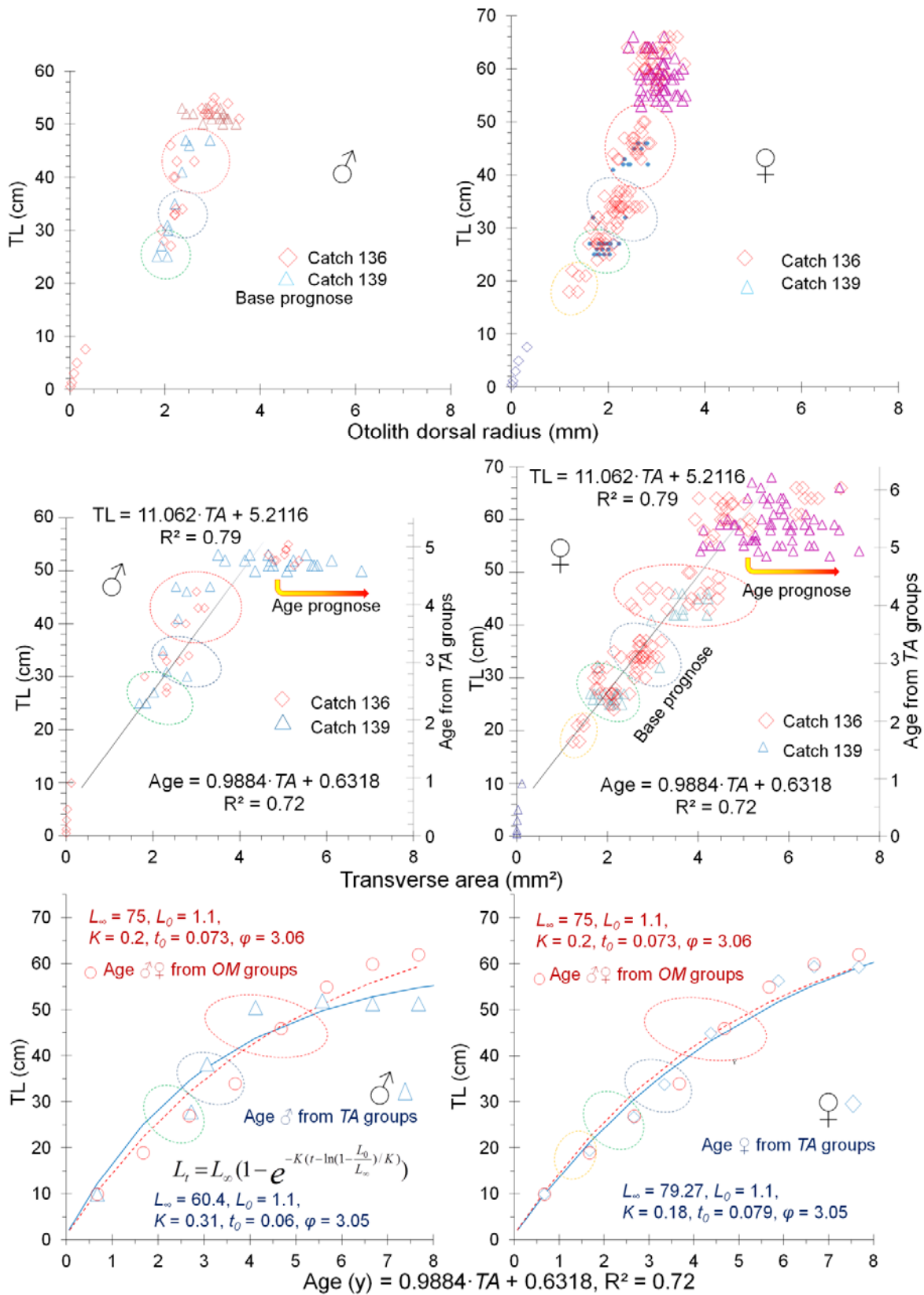


Fig. 22 TL and dorsal radius do not distinguish old age, but transverse area (TA) do, and give similar Bertalanffy growth.

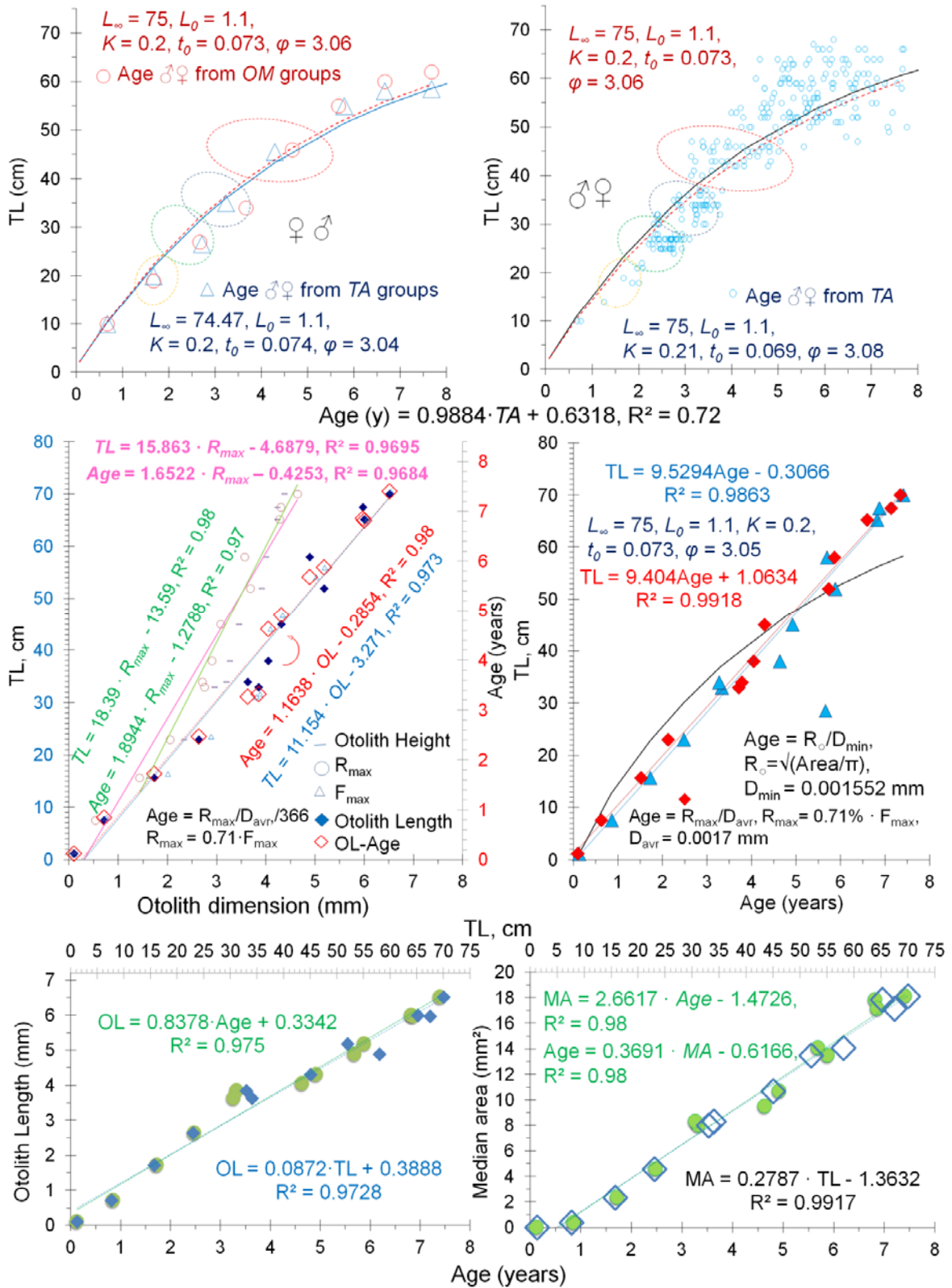


Fig. 23 Determine growth of fish from TA, age from median area (MA) and from otolith length (OL).

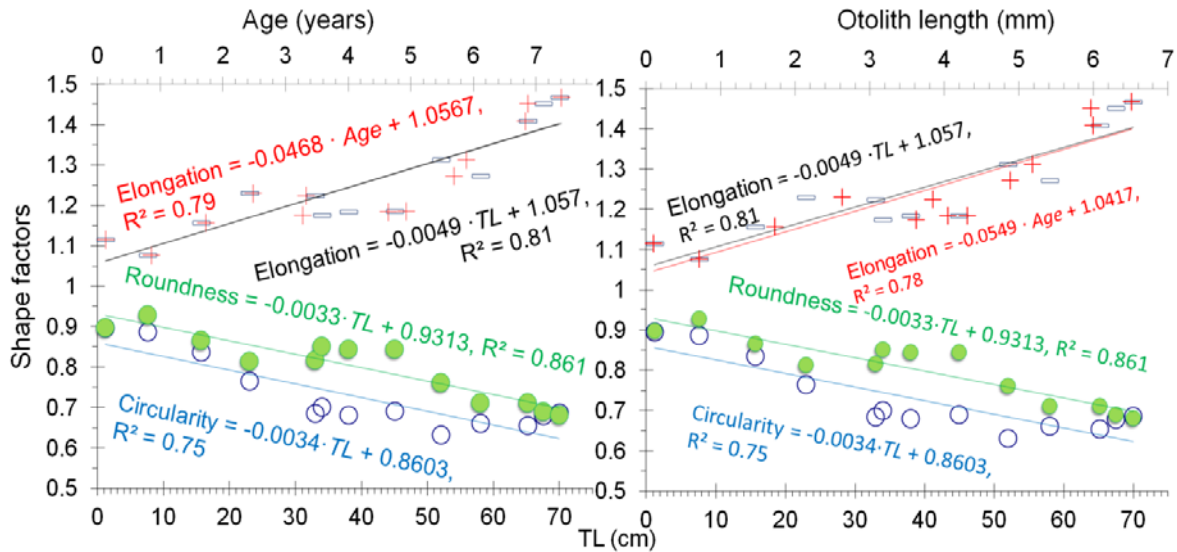


Fig. 24 Constantly growth of otolith elongation show successful use of otolith length to age estimate, and speeding of swimming with the age, dropping down circularity and roundness show growth activity of fish.

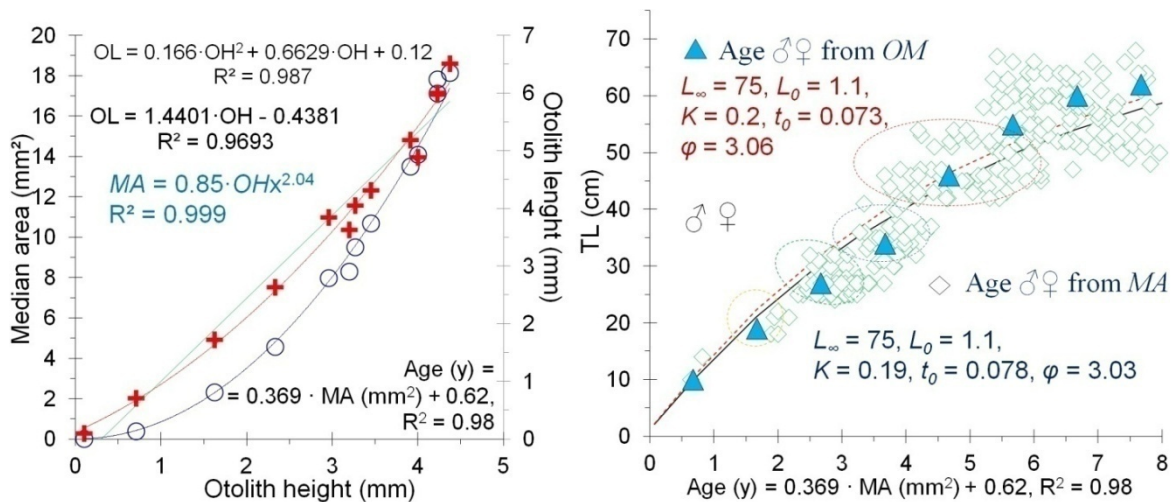


Fig. 25 Growth Bertalanffy from age estimated from otolith median area (MA) confirm age estimate from otolith mass. MA as expected has power dependence from otolith length.

years interruption for 40-50 cm TL fish being in age groups of 4 and 5 (Fig. 24). This seems to be related to the maturing process, during which the energy is used to gonad development at the expense of a reduction in the rate of somatic growth and in activity of swimming, which stimulates the shape of otolith. The first gonad maturity males *C. aceratus* is achieved in the end of age of 5 years, with a length of 50.6 cm, while females in the age of 6 years with a length of 61.5 cm TL (Fig. 26).

This means that the age and length at 50% of mature males at South Georgia was much lower than

females. This difference in maturing stage between sex was marked in otoliths by lowering elongation two times (Fig. 24).

4. Discussion

4.1 Daily Increments

There are only two case uses of daily increments to age determination of icefish: for *C. gunnari* and *Ps. georgianus*. This is a third case and first paper for *C. aceratus*. There are many reasons:

That is because otolith of icefish, which is very fragile, is difficult to get. This work present easy,

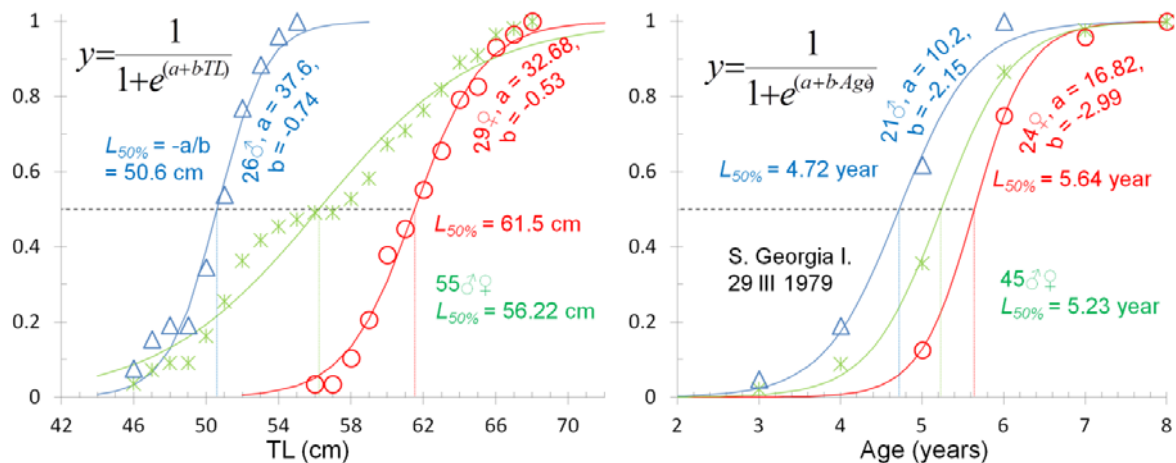


Fig. 26 Logistic curves of *C. aceratus* lengths and ages with gonadal maturity stage above 3.

quick and not expensive method of prepare clear daily increment for the light and SEM microscopy from 40 otolith in one time per one microscopic glass;

Another problem is how to choose and right account their numbers. In this paper, daily increments were interpreted from arrangement of collagen discovered by removing aragonite from collagen matrix. Collagen is first component to start arranging bicristal of otoliths. It is very conservative—little changing in the process of evolution. Its structure is characterized by high durability and strength, and it is made up by the same size unit: 260 nm tropocollagen, in the same pattern over all otolith. Taking above into account, the pattern of daily increments from larvae living together of close related icefish should be similar. It is true, because average width of daily increments of otolith larvae from similar species were: 0.001 mm for *C. aceratus*, 0.0018 mm for *Ps. georgianus* and 0.0015 mm for *C. gunnarii*, Averages of daily increments for the postlarvae for *C. aceratus*, *Ps. georgianus*, *C. gunnarii* are as follows: 0.0016 mm, 0.0028 mm and 0.0024 mm. Although there were not measured from the same otolith radii, there were measured automatically. Similar dimensions of micro-increments among above 3 species of *Channichthyidae* suggest that they are the same units well chosen for age determination more or less squeezed by being under different pressures of little different environments or swimming activities. There

are interspecies differences, because larvae of *Ps. georgianus* catch larvae *C. aceratus* and *C. gunnarii*, and eat them. But larvae of *Ps. georgianus* were not caught and eat by *C. aceratus* or *C. gunnarii*.

Another reason for this species problem is their bones undergo constant and large reduction in ossification. In this reason, the main bone elements should be changed at last. Otoliths play one of the most important roles. Changes of them will destroy the base of their life—the swimming possibility. Additionally, daily increment were measured from collagen pattern not from aragonite, where its amount of deposition is going down [5].

The last exclusively large for study fish are the troubles to get the age data from the old individuals. For all icefish for age interpretation, the transverse of otolith planes are used. Unfortunately, the radius (of growth) in dorsal part of otolith of *C. aceratus* do not increase over 50 cm in males and over 60 cm in females. In this case, for measurement of growth of large, older fish is better to use growth radius along the length of otolith (OL), which grows continuously. But this dimension does not provide easy opportunities for counting increments. Along the length of the otolith there are depressions forming discontinuities and many adhesions from additional primers creating bulges and horns (Fig. 19). In this situation, the age can be obtained by dividing the radius that increase otolith length by the average width

of the daily increments or from otolith shape analysis, such as analysis of the otolith growth on the median planes.

4.2 Body Growth and Environment Distributions

Studying the changes of otolith shape during growth and development of fish and comparing them with other species show that they reflect the body growth, differentiate fish on the age groups and life stages living in different environment and have increased with age swimming speed (Fig. 19). They have interspecies differences: in body size and shape, in swimming speed and strategy and in distribution in the environment. The otolith length of *C. aceratus* is larger than other icefish such as *Ps. georgianus* and *C. gunnari*. Large otolith *C. aceratus* indicate deeper depth of life more in bottom, up to 700 m depth. *Ps. georgianus* has semipelagic life up to 500 m depth and *C. gunnari* has pelagic life style [15]. Grenadier fish in compare have increased otolith length and size in

consequence influencing of larger deeps of life up to 1,200 m depth (Fig. 27).

Diversity proportion of otoliths is accompanied by changes in body proportions. *C. aceratus* having shorter and longer otolith has a body also less high as compared to high otoliths and high body *Ps. georgianus* [15]. Similarly, the proportions of otolith *Macrouridae* display his body proportions associated with changes in the depth of occurrence, resulting in the reversal of proportion to subject species living closer to the surface (Fig. 27). Just like the sawfish who has a long saw at the front of body, so, the otolith has a big horn in front (Fig. 28).

In general, otolith of *C. aceratus* are larger than those of other white blood. Their body also achieves greater size than the other *Channichthyidae* (Fig. 29).

Faster growth of body help *C. aceratus* to save before eaten out by *Ps. georgianus* as their larvae eat larvae of *C. aceratus* probably until they become large. The results obtained suggest the very fast growth rate

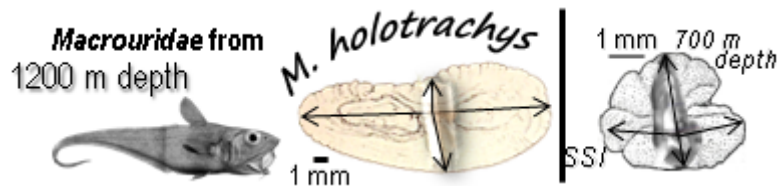


Fig. 27 Long otolith length, opposite dorsal radius as body.



Fig. 28 *Xiphias gladius*, 96 km·h⁻¹.

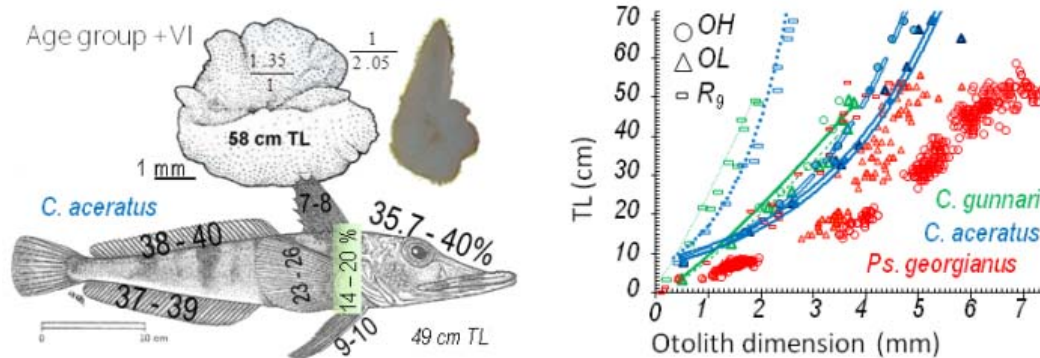


Fig. 29 *C. aceratus* attain the largest body and otoliths.

of *C. aceratus* during its first three years of life. It is in concord with fact, that not only *C. aceratus* but all *Channichthyidae* family also reach rapidly large body due to their adaptation to cold water [17]. With larger body, they have reduced heat loss due to the lower surface to volume rate and the Bergman's rule of energy benefits. Average size of adult *Channichthyidae* is 43 cm TL, with yearly increment = 6-10 cm. The other Antarctic fishes have smaller body size: *Nototheniidae* 36 cm TL, *Bathydraconidae* 26 cm TL and 15 cm TL [17]. Large body determine to generate additional adaptations. Large body has large resistance and need more power for swimming in the strong currents—*C. aceratus* advanced in swimming [6]. In order to spend less energy need in cold water, *C. aceratus* has low energy swimming strategy like all icefish, by vertical waving of the pectoral fins. Body with a little energy not waving has reduction in axial muscles, and use energy of current. There is also floating strategy, without swimming bladder supported by reduction of bone forming (less liming, replacing the bone by cartilage). Species has scaleless body for the reduction of friction between body and water, reducing the resistance of the body [15].

Cross sections of blood vessels and heart *C. aceratus* are large because of the need to transport a large volume of blood, in particular for greater distribution of dissolved oxygen which due to lack of active transport: hemoglobin—red blood cells eliminated in order to reduce the density of the blood and thus, to preserve its flow in cold waters. Adaptation of this species for cold water is greater – it has more bottom life style, where water are colder. It is confirmed anatomical and physiological because apart from the lack of myoglobin in the muscles species has not it also in the heart. As in deep, cold water affinity of myoglobin for O_2 increase it will impedes taking them to tissues for use. Lack of myoglobin in heart increase in cold deep water the use of O_2 , that are accumulated in large heart.

4.3 Geographical Distribution

C. aceratus like almost all ice fish live on shelf exclusively on Antarctic island. They do not live on open ocean. Their otolith are not very flattened that characterise pelagic fishes like *Elecrona antarctica* or very fast *Scombrus japonicus* ($21 \text{ km}\cdot\text{h}^{-1}$).

Their otolith are not very rounded, that characterise slow swimming medusa or with water propulsion slow swimming squid (Fig. 31), whose distribution are strong depended on dispersion by currents.

On Antarctic island, there are strong surface currents, that disperse icefish larvae to the open ocean (Fig. 32)—the otolith of icefish larvae are more rounded (Fig. 24).

With the increase, the larvae have more capacity to swim and finally adults swim 4 times faster (Fig. 33). At the same time, the shape of their otoliths becomes less rounded and more elongated (Fig. 24).

The transverse otolith plane of older icefish resemble a physical pendulum (in unstable equilibrium, which is very sensitive to deviation by high moment of inertia) to measure by its small oscillation the deviations in their vertical lifting and sinking in the depth range 0-700 m. The strongest marked daily vertical migrations has *Ps. georgianus* and accordingly, it has the largest dorsal otolith radius R_9 (l —length of the pendulum) and OH at the



Fig. 30 *Scombrus japonicus*, swimming with body waving $21 \text{ km}\cdot\text{h}^{-1}$.

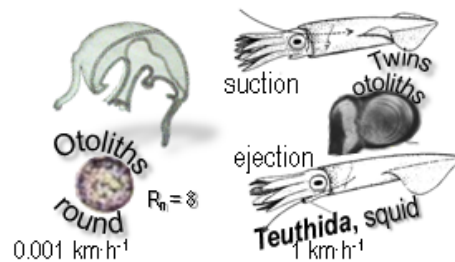


Fig. 31 Pulsed swimming of medusa and squids.

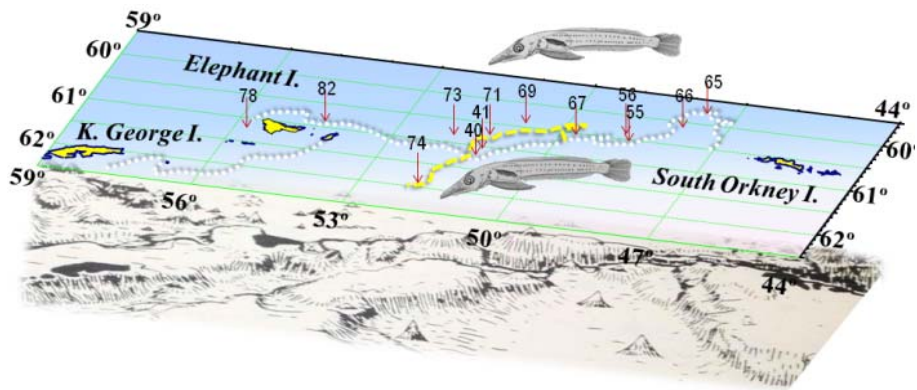


Fig. 32 Postlarvae of *C. aceratus* were found at station 40 and 66 in the water of open ocean in summer 1989 [15].

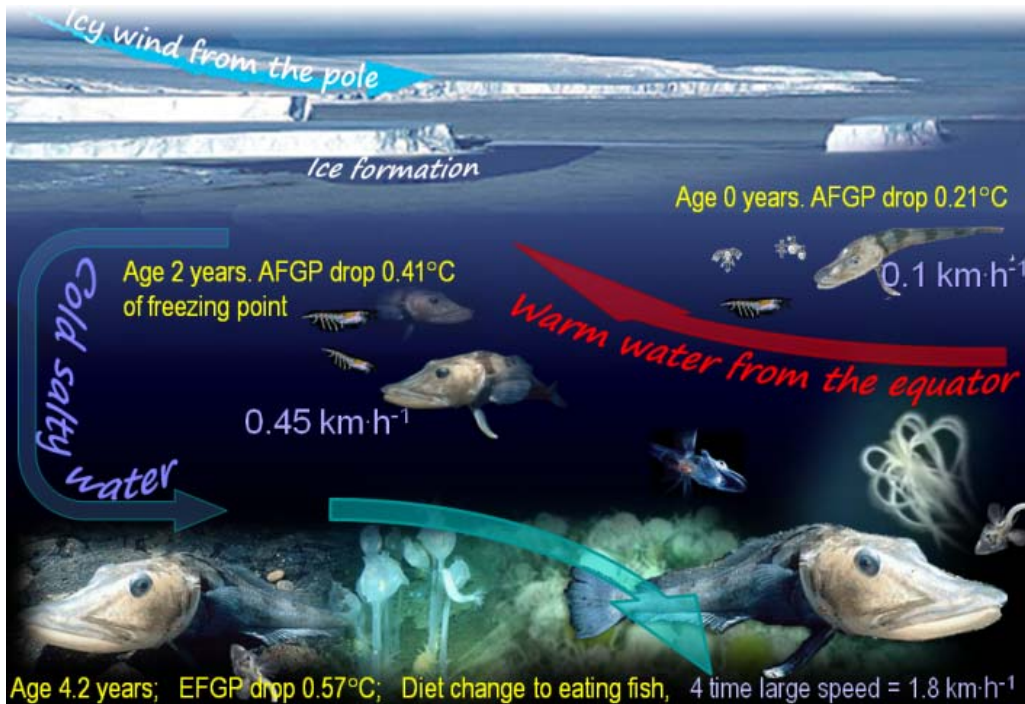


Fig. 33 Adult swim 4 time faster than young [18].

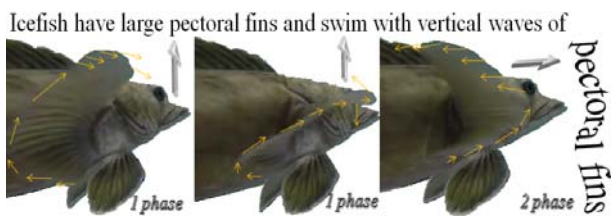


Fig. 34 In first phase of vertical waves fins are lifting body, in second (backing way) push it forward.

transverse section (Fig. 31). The oscillations of the pendulum described by wave equation are used in various apparatuses measuring time and mass. In this case, influence force during migrations equilibrated with force of otolith attachment. This otolith shape may be related as above with less energy swimming

strategy of icefish in their vertical waves of pectoral fins (Fig. 34).

C. aceratus live on the bottom, in deep water where the currents are much slower than at the surface. It was confirmed in the catch. Old *C. aceratus* dominated on West of South Georgia I. (Fig. 35).

Otolith in early stages of development as a more rounded indicate their more dispersion: larvae *C. aceratus* occur in the open ocean, near the edge of the ice, but adult fish were not. The propagation of larvae between the islands confirms the broad geographical distribution of species [15] (Fig. 36). These species has characteristics which give wide spread north into

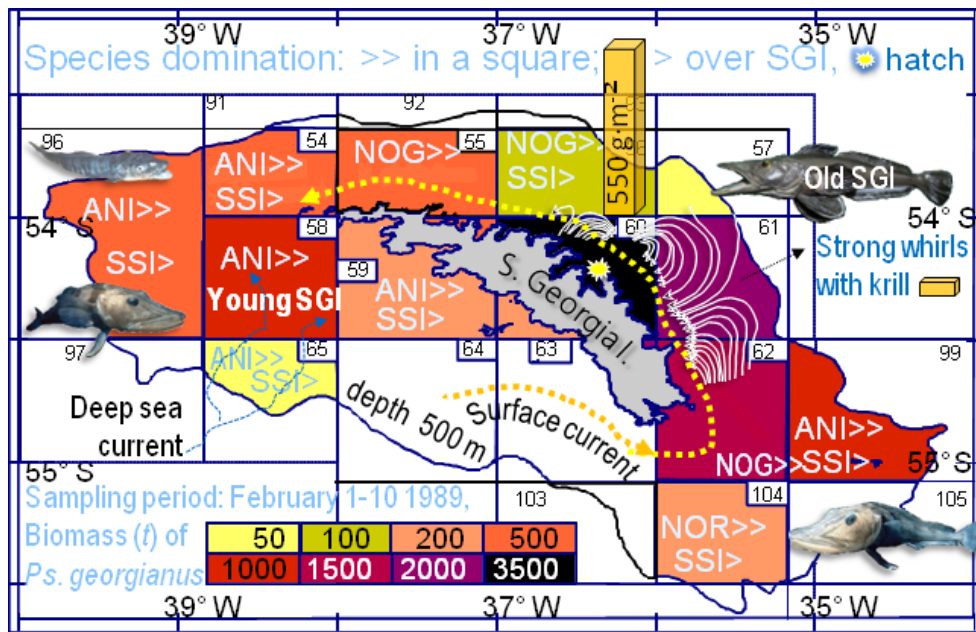


Fig. 35 Old *C. aceratus* (SSI) live on west in cold deep sea current (*C. gunnari* (ANI) and *Ps. georgianus* (SGI)).

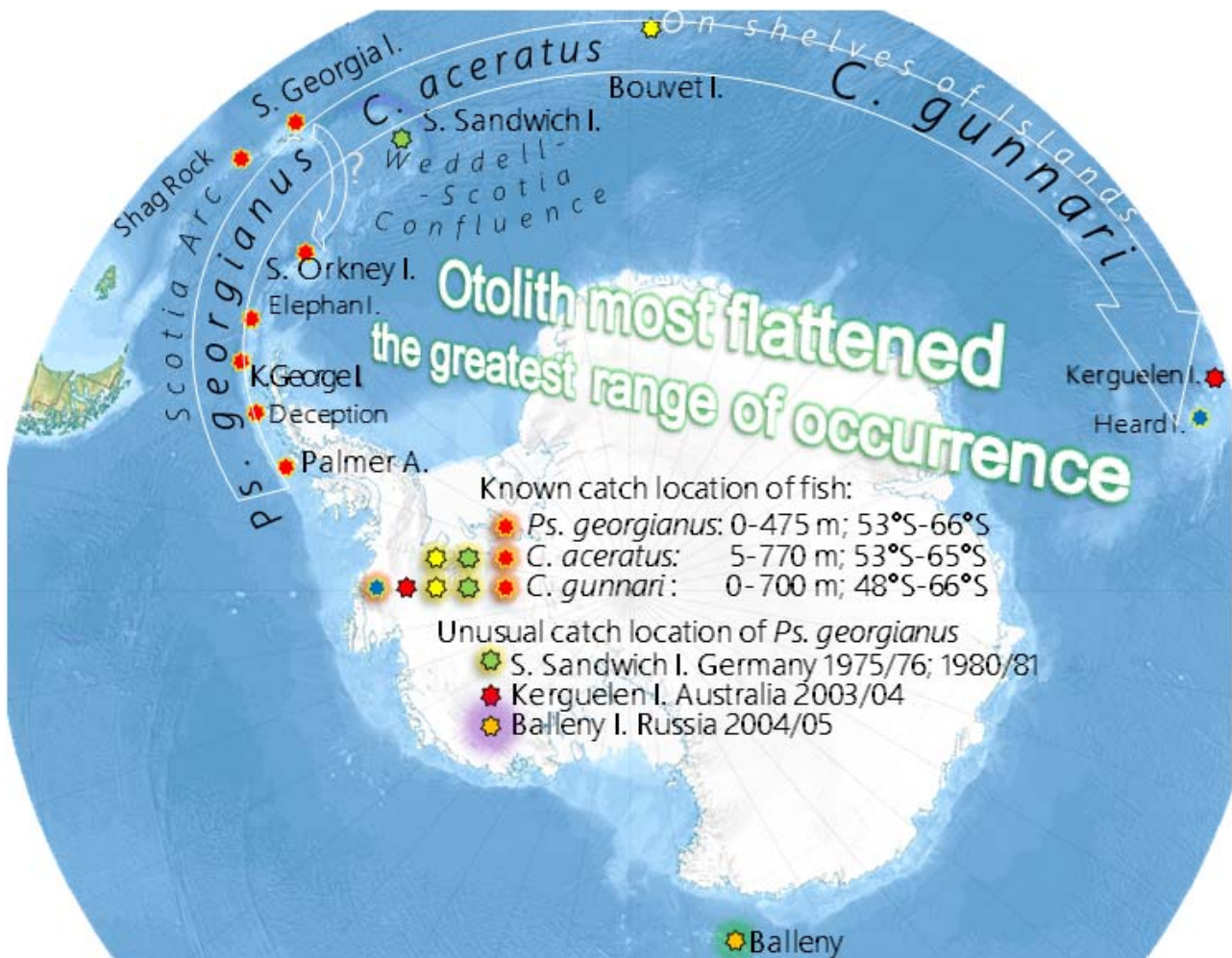


Fig. 36 Geographical distribution of icefish.

warmer waters. Larval stages have greater tolerance for warm water what increases the adaptation of species [18] (Fig. 33). Similar larvae spreading beyond the shelf on the water of the open sea has *C. gunnari*. In contrast, *Ps. georgianus* is less distributed across by the currents of the ocean. Its larvae in the open ocean cannot be found as these two species. Its otoliths as the body are very high, so, it has greater success in swimming in currents and more useful to the vertical migration for food and better living conditions.

5. Conclusions

At the same time, methods prepare 40 otolith slides, glued on a microscope slide (to measure daily increments and otolith shapes): (a) large saving time in prepare slices; (b) saving material (gold and platinum for SEM); (c) save otolith growth boundary before a clash and (d) it allows to obtain very thin layers of very fragile otoliths. This work present easy, quick and not expensive method of prepare clear daily increment for the light and SEM microscopy from 40 otolith in one time per one microscopic glass.

Traditional reading age of older fish from the annual growth rings, as well as daily on transverse sections is difficult even for *C. aceratus*. The radius of the dorsal section is not incremented with the length of the fish (so also not with its age). In the process of desorption, which characterise this species, aragonite storage may be performed repeatedly for one and the same part of otolith. Crystallization aragonite may not extend from the surface of the collagen network, but from its center. So, the surface could be the subject of many crystallizing and re-crystallizing cycles.

Age of this species could be obtained by otolith length interpretation, and by shape analysis, taking into account parameter such as median area surface.

Proportion of otolith confirm that this species has bottom life style.

Otolith more rounded can indicate dispersion to open sea for larvae, having greater tolerance to warm

waters, which increases the adaptation of species.

Life stages, their strategy, their anatomy and their changes of swimming speed seems to be adopted to environment conditions to the currents and countercurrents. As growing large and move to bottom, they prevent for being the floating drift in large Antarctic Current. *C. gunnari* is pelagic but it has not like *C. aceratus* the reduction in axial muscles it has muscles very similar to muscles of trout.

The width of daily increments in otoliths of larvae and post larvae of *C. aceratus* and their pattern are similar to *Ps. georgianus* and *C. gunnari*, confirming that there are the same units of otolith growth made by matrix of collagen fibres of 260 nm tropocollagens.

C. aceratus is fast growing species, reach large body, which is due to one of special icefish adaptation to cold water.

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