Age validation of deepwater fish species, with particular reference to New Zealand orange roughy and oreos: a literature review

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Final Research Report for Ministry of Fisheries Research Project DEE2000/02 Objective 1

National Institute of Water and Atmospheric Research

July 2002
Final Research Report

**Report Title:** Age validation of deepwater fish species, with particular reference to New Zealand orange roughy and oreos: a literature review.

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1. **Date:** June 2002

2. **Contractor:** National Institute of Water and Atmospheric Research

3. **Project Title:** Examining feasibility of age validation in black and smooth oreos

4. **Project Code:** DEE2000/02

5. **Project Leader:** D.M. Tracey

6. **Duration of Project:**

   - **Start date:** February 2002
   - **Completion date:** October 2002

7. **Executive Summary**

Stock assessments used as the basis of TAC-setting rely upon estimates of the biomass of a species and its productivity. The latter is dependent on age and growth analyses. There is growing confidence in recent interpretations of the ages of deepwater species such as orange roughy, black oreo, smooth oreo and black cardinalfish. However, these studies are based on growth zone counts from otolith sections, and are unvalidated.

This report reviews available published research on ageing of deepwater fish species, summarises Fenton’s unpublished paper on radiometric ageing of oreos, and summarises radiometric age validation studies conducted at Moss Landing Marine Laboratories. It includes, as appendices, two recent reviews of orange roughy ageing.

The following ageing and age validation methods are covered: length frequencies; annual growth zones; daily growth zones; marginal increments; otolith weights; mark-recapture; biochemical ageing; radiometric ageing; elements and isotopes; and bomb radiocarbon. There are advantages and disadvantages to all of them. There are greater difficulties in ageing (and validating the ages of) deepwater fishes by all methods.
because of the narrowness of otolith growth increments. The most promising validation procedures are considered to be radiometric ageing (using the known decay rate of radioactive parent elements into daughter elements), and bomb radiocarbon (using the $^{14}\text{C}$ peak created by bomb tests in the early 1960s, and incorporated into otoliths, as a time marker). Both methods are relatively new, and still undergoing development.

A brief review is given on ageing studies on orange roughy, oreos, black cardinalfish, a deepwater rattail, rubyfish, toothfish, and sea perch. In general, deepwater species are considered to be slow-growing and long-lived.

Fenton’s unpublished radiometric study of oreo ages contains several deficiencies, and serves more to emphasis the complexities of this method than to advance age validation of these species.

Radiometric studies at Moss Landing Marine Laboratories use new analytical procedures which permit the study of much smaller samples than has previously been possible.

1. Literature review of recent developments in deepwater fish ageing

1.1 Introduction

The ageing of fishes, and consequently the determination of their growth and mortality rates, is an integral component of fisheries science. It is not an easy task; a variety of techniques are employed and continue to be developed, discrepancies between different workers are common and otolith-reading has been described as “...as much an art as a science” (Williams & Bedford 1974). An added complexity involves variability in growth rate of a species both geographically and over time. Validating fish ages is difficult, and in many cases not immediately possible.

Most studies of fish ageing, and reviews of such studies, are prefaced by a cautionary summary similar to the above. It is usually an admission that what follows will have deficiencies, although in most cases the information will be the best currently available. The risk in repeating such a preamble is that one key phrase can be overlooked: “ageing is an integral component of fisheries science.” Subsequent population modelling can be jeopardised, and the results misleading, if input parameters for growth and mortality are incorrect. This is particularly relevant to long-lived fishes, with very low natural mortality rates and low productivity.

It is useful to review an area of study from time to time. This report does not fully review all developments in the ageing of deepwater fishes and their relevance to New Zealand species, but addresses the directions that recent research has taken. New techniques for ageing, and/or validating the ages of deepwater fishes, are described, including improvements to the long-established procedure of counting growth increments in hard parts, particularly otoliths.

This report has three sections. The first (by L.J.P. & D.M.T) is a literature review, in two parts: (a) a brief account of previous reviews relevant to deepwater fishes, and a description of the procedures used to age fish and validate ages, with particular
reference to New Zealand species; and (b) a summary of published research on the main New Zealand deepwater commercial species or species-groups (orange roughy, oreos, black cardinalfish, rubyfish, and sea perch) plus a note on toothfish. The second section (by R.I.C.C.F.) reviews Fenton (n.d., [1996]). This report is treated separately as it is of particular interest to New Zealand (because of the species covered), although it has not undergone the usual quality-assurance process of peer review for formal publication. The third section (by D.M.T.) reviews recent radiometric studies at Moss Landing Marine Laboratories (MLML), where the otolith work for the other part of this study is being undertaken.

This report addresses Milestone 1 of MFish project DEE 2000/02, which is a feasibility study on the age validation of New Zealand deepwater fish species, particularly orange roughy, oreos, and black cardinalfish.

Milestone 2 will involve applying the “improved” radiometric technique to four samples of pooled orange roughy otolith cores, and determine if ages can be validated by comparing the measured radiometric ages with ages from growth increment counts on otolith thin sections. Milestone 3 will be a feasibility study to determine if otolith cores from black and smooth oreo and black cardinalfish can be isolated, and to measure the amount of material (and hence the size of otolith cores that represent the first few years of growth) required to perform radiometric age validation on these deepwater species. These two Milestones will be reported upon separately.

Two published reviews of orange roughy ageing studies are included here as Appendices, for ease of reference.

Common, scientific, and family names for the species mentioned in this review are listed in Table 1.

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<th>Common name</th>
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1.2 Previous reviews

The frequently cited paper of Beamish & McFarlane (1983) reviewed ageing studies from the standpoint of validation, and pointed out that relatively few such studies were properly validated. Often from necessity, the majority of published accounts are based on counts of assumed annual growth zones. Attempts at validation, such as recording seasonal changes in the growing edge of a scale or otolith were only possible for younger age groups; the marginal increments from older fish being too narrow. The assumption had to be made that the annual nature of early rings continued without change. This paper, more than any other, stimulated the continuing search for good age validation techniques, particularly of older fish. The infrequently cited paper on “ageing in fish” by Craig (1985) is not a review of ageing procedures, but is a useful account of the basic concepts of growth, age at maturity, longevity, physiological ageing, and senescence. It makes the point (as do many individual studies on fish ageing) that underestimating the age of long-lived species produces incorrect growth and mortality rates which are important to understanding population dynamics.

As a result of research carried out in the 1980s and 1990s, there was a growing realisation that cross-sectioned and thin-sectioned otoliths yielded higher estimated ages than did whole otoliths. There were numerous studies on otolith microstructure, both from a theoretical standpoint and from the perspective of understanding daily, seasonal, and annual growth marks. Campana (1999) provides a useful overview of this research. Most of this research was not directed at long-lived fish, but one outcome was a realisation that deepwater fish could reach high ages (Bergstad 1995, Munk 2001, Cailliet et al. 2001).

During this period, also, several innovative procedures were developed which could validate – generally, rather than precisely – the ages obtained by ring counting. Radiometric, bomb radiocarbon, and element/isotope methods are covered in separate sections below; useful summaries of these procedures are provided by Campana (1999, 2001) and Cailliet et al. (2001).

The 1980s and 1990s were also a period of increasingly detailed age and growth studies, with – prompted by Beamish & McFarlane (1983) – more emphasis on age validation. Special journal issues were devoted to ageing studies (e.g., Smith 1992), and two international conferences resulted in symposia volumes (Secor et al. 1995; Fossum et al. 2000). Campana (2001) gives an extensive account of the concepts of accuracy and precision in age determination, and a critical review of age validation methods.

Paul (1992) reviews all New Zealand marine fish ageing studies up to 1990. Kalish (1992) gives a much briefer review, with emphasis on future directions. The only review of ageing studies on a New Zealand deepwater species is that by Tracey & Horn (1999) on orange roughy.
1.3 Ageing methods and age validation

Determining a fish’s age, and validating that age, are separate but related tasks. Studies may cover only one, or mainly one, or may integrate both. In addition, some descriptive studies of otoliths (and other ageing structures) are relevant either to ageing or to validation. All these concepts are combined in the following account, in the sequence: length frequencies; annual growth zones; daily growth zones; marginal increments; otolith weights; mark-recapture; biochemical ageing; radiometric ageing; elements and isotopes; and bomb radiocarbon.

1.3.1 Length frequency analyses

Age groups can sometimes be inferred from length frequency modes. Length data are usually readily available. There are limitations: the growth rate must be moderate to fast, the spawning season annual and relatively short, and there must be little size-selective schooling or movements into or out of the sampling area. It is most suitable for juvenile fish, and often only the first two or three age groups. The size ranges of adult age groups overlap almost completely, but if a particularly strong year class is identifiable in successive annual length frequency samples it may be possible to determine (e.g., from otolith-reading) that its age increases by one each year.

This procedure is seldom applicable to deepwater species, the majority of which appear to be slow-growing, at least as adults, and long-lived. It was used for New Zealand orange roughy juveniles by Mace et al. (1990), who determined the growth of the first three age groups. Rodriguez-Marin et al. (2002) analysed the progression in size of an exceptionally large year class of a deepwater macrourid to partially validate a growth curve determined from zone counts in otolith thin sections; the two were in good agreement, but the year class involved was less than 10 years old. Paul et al. (2002) were also able to match the size progression of strong year classes of sea perch to a growth curve derived from otolith thin section counts, but also only from relatively young fish.

1.3.2 Annual growth zone analysis: scales and otoliths

Counting growth zones (rings, annuli, etc.; studies use different terminologies) has been the standard procedure for ageing fish. Over time, the structures/procedures have progressed from scales to whole otoliths, otolith cross-sections, and otolith thin sections. Scales have seldom been used for deepwater species, but when they have (e.g., Mel’nikov 1981) the low ages obtained are now considered unreliable.

Comparing different structures (otoliths, fin spines, even scales) can sometimes be useful in distinguishing true from false growth structures, or determining at least the early growth pattern, but as all are influenced by similar factors (physiological cycles and/or responses to environmental events) the results are seldom conclusive. Paul et al. (2000) observed that fin spine sections of rubyfish (*Plagiogeneion rubiginosum*) usually had five clear and prominent rings followed by numerous (and uncountable) narrow rings; otoliths had obscure growth zones, but these could be resolved into five broad early zones followed by numerous narrow zones.
As thin sections became the preferred otolith preparation, these narrow outermost zones could be more easily resolved; they were subjectively identified as annual structures, and higher counts were recorded for several deepwater species (e.g., Wilson & Boelhert 1990, Doonan 1993, Doonan 1994, Doonan et al. 1995, Doonan & Tracey 1997, Doonan et al. 1997, Allain & Lorance 2000, Beamish & McFarlane 2000). New Zealand researchers quickly adopted the thin section otolith preparation technique to orange roughy and the oreo species.

Investigation of internal structures and zone patterns took new directions (e.g., Gauldie 1987, 1990a, Gauldie et al. 1995; reviews by Campana & Neilson 1985, Campana 1999). Changes in the spacing and appearance of zones were described, notably a change from broad to narrow growth zones. This change (at zone 30, ±15) was termed a transition zone in orange roughy (Kalish et al. In Annala 1993), and related to the onset of maturity by Francis & Horn (1997). In some other deepwater species, e.g., oreos, rubyfish, and black cardinalfish, the change from wide to narrow zones occurs earlier, at about 5–7, and is suspected to mark a change from pelagic to demersal life (Smith & Stewart 1994; Tracey et al. 2000; Paul et al. 2000; NIWA, unpublished). In some species, notably rubyfish and sea perch (Helicolenus), and in some otoliths of other middle depth to deepwater species, the otolith region between these two positions (approximately zones 6 to 30) is difficult to interpret; the zones may be poorly defined and irregular, there may be two positions where zones change from wide to narrow, there may be only a gradual decline in zone width, and/or it may not be clear where the inner multi-banded zones change to the outer, more regular, single or paired zones. More generally, otolith thin sections from middle depth to deepwater species show a multiplicity of ‘zones’, ‘rings’, and ‘checks’. Without some independent information on the annual formation of zones, or on the age of the fish, it is possible to make high counts (all zones, etc.), intermediate counts (grouping some zones), or low counts (grouping most zones). This remains a significant problem for this group of fishes.

1.3.3 Image analysis

Efforts to develop computerised procedures to obtain objective counts of otolith growth zones continue (e.g., Cailliet et al. 1996a, Morison et al. 1998, Robertson et al. 1999, Troadec et al. 2000). However, this approach to the problem of distinguishing between true zones and false checks, or (more realistically) to the goal of obtaining standardised counts, remains experimental.

1.3.4 Daily growth zone (microincrement) analyses: otoliths

The demonstration by Pannella (1971) that daily growth rings could be detected in fish otoliths, stimulated many innovative studies in several areas of fish ageing. It soon became clear that in most fish species, particularly slow-growing ones, a full sequence of daily rings was present only for the first one or two years (and often only for the first summer-autumn growing season). For this reason, daily rings (also termed microincrements) have proved of limited value in studies on deepwater species. Some studies have been made of the microincrement pattern of orange roughy otoliths (Gauldie 1987, Gauldie & Nelson 1988, Gauldie et al. 1995); the authors infer a low age for the species, but appear to overlook the probability that microincrements and
associated structural and chemical features represent an incomplete growth record. A review of this work by Gauldie and co-authors is given by Tracey & Horn (1999).

The most profitable line of enquiry for future work on otolith microincrements in New Zealand deepwater fishes may be to identify the location of the first annual (probably winter) slow-growth zone or check.

1.3.5 Marginal increment analyses: otoliths

Marginal increment analysis is the most commonly used otolith age validation method. The theory is sound; if the outermost growth increment (zone, etc.) assumed to be annual does progress through a recognisable cycle (e.g., changing from translucent through opaque back to translucent) during a year the annual nature of its formation is confirmed. There are two ways of quantifying this cycle; (1) the proportion of otoliths at any one time with an edge in a defined state; and (2) the width of the outermost zone, relative to the penultimate zone. In reality, marginal increment analysis is highly problematic, and difficult to undertake correctly (Campana 2001). Difficulties include light refraction and reflection, differences between ‘zone forming’ and ‘zone visible’ times, differences in marginal increment size in different parts of the otolith, and regional and annual differences — probably environmental — in the timing of zone formation. The review by Beckman & Wilson (1995) of over 100 studies on the timing of otolith growth zone formation revealed numerous ambiguities attributable to the physical difficulties in defining marginal increments, and to inconsistencies in nomenclature. The procedure is appropriate for juvenile fish of fast-growing species, which have wide growth zones, but it is undesirable to extrapolate the results even to adults of these species. It is extremely difficult to use for slow-growing and long-lived deepwater species. In addition to all the problems listed above, the growth zones are too narrow for quantification of the marginal increment.

However one reasonably satisfactory application of this technique was applied to the long-lived deepwater (depth range 70–2000 m) species Patagonian toothfish (Dissostichus eleginoides) (Horn, 2002). Several otolith samples of Patagonian toothfish were collected throughout the year and the margins from fish of any age were generally opaque in summer and translucent in winter. This species appeared to deposit one translucent zone in its otoliths each year, and counts of these zones indicated a probable validation method to determine fish age. D. eleginoides appears to be moderately fast growing, at least to about age 10, and reasonably long-lived, reaching at least 50 years. Von Bertalanffy growth parameters were also calculated, separately by sex, for Antarctic toothfish (Dissostichus mawsoni). Otoliths of this species were interpreted similarly to those of D. eleginoides, but this method of ageing D. mawsoni is unvalidated. D. mawsoni appears to be moderately fast growing, at least to about age 10, and can live for at least 35 years. This species probably grows at a slightly faster rate, and reaches a larger size than D. eleginoides.
1.3.6 Otolith weight analysis

Otoliths weight has been increasingly used in recent years as a proxy for age, either alone or in combination with otolith and fish dimensions. Although not a validation procedure, the relationship between otolith weight and otolith zone count (= age?) is informative. It may be linear, or otolith weight increments will decline with age – with either a ‘broken-stick’ or asymptotic relationship. If weight increments are found to increase with assumed age, the count of growth zones is almost certainly in error and has underestimated age. Otolith weights have generally been used for inshore species with a moderate or fast growth rate, where individual age groups can be recognised, but the otolith weight-age relationship may also prove useful for deepwater species.

Otolith weight is usually considered to be a better measure of age than is fish length. However, a search for potential indicators of orange roughy biomass depletion showed that in this species they were only marginally better for this purpose (Francis & Smith 1995).

To determine whether age structure of the orange roughy Chatham Rise population had changed with exploitation, random samples of whole otoliths collected from North Chatham Rise surveys between 1984 and 1992 were weighed. Results showed that there was no significant change in otolith weight over time. There was however, a decline in the proportion of smaller otoliths over this period. These results are consistent with the hypothesis (based on no change in mean length of the population over time), that recruitment had been low in recent years (Francis and Tracey 1994).

Otolith-mass growth parameters are often incorporated in the radiometric procedure for determining ages. However, Francis (1995a), using orange roughy data, demonstrated that this need not be necessary.

1.3.7 Mark-recapture studies

Mark-recapture procedures are frequently used for growth studies and age validation. The simplest (in theory) is to use fish size at release and recapture to determine the intervening growth. It has several general shortcomings (a low recapture/return rate; abnormal post-tagging growth; problematic check-mark induced by tagging trauma; etc.). It has only occasionally proved useful in measuring growth, and it rarely provides information on longevity. A development of this procedure is to chemically mark the fish (with or without a physical tag) with a substance such as oxytetracycline (OTC), and after recapture interpret the growth increment visible outside the chemical mark. Successful studies of this nature have been undertaken on fish species from shallow to moderate depths. Mark-recapture studies of deepwater fishes are difficult to impossible, in view of the pressure-related damage (barotrauma) they suffer on capture, the difficulty of holding them in captivity, and the logistics of tagging or chemically-marking, and then recapturing, sufficient fish to be informative.

Attempts have been made to validate otolith microstructure periodicity for deep sea fish age estimation by capturing and holding the fish in situ (de Pontual et al. 1998). This experiment was a “world first” live capture and tagging in situ of a single orange roughy. The cage was retrieved after 1 month and while the fish had died the post
mortem state indicated it had only been dead for 1 or 2 days. The otoliths have yet to be analysed.

1.3.8 Biochemical ageing

The search for ageing procedures independent of growth zones in hard parts has involved some consideration of biochemical compounds that may change or accumulate at a known rate with age. One that initially held some promise was lipofuscin, a metabolically-accumulated cellular ageing pigment (e.g., Hill & Radke 1988, Hunter & Vetter 1988). However, other work (e.g., Nicol 1987, Hill 1991, Girven et al. 1993) revealed ambiguities and limitations, particularly for fish, and there appear to have been no recent studies. Another procedure for aquatic animals involves the racemisation of aspartic acid in biogenic carbonates (e.g., Goodfriend 1992, Goodfriend et al. 1995, George et al. 1999); it has been applied to marine invertebrates and whales, but not fish.

1.3.9 Radiometric ageing

More emphasis is now being placed on validating the counts of otolith growth increments by age estimates which are independent of the otolith's appearance and structure. The most frequently used method, particularly for the deeper water species which are increasingly found to be long-lived, is radiometric age determination—the use of radioactive disequilibria of naturally occurring radioisotopes in calcified structures, such as otoliths. Examples of such comparisons include Milton et al. (1995), Smith et al. (1995), and Stewart et al. (1995), but there is a considerable literature on radiometric ageing (see also reference list of MLML studies in Section 3).

This procedure infers age from measurements of the relative activities of two radionuclides from the same decay series and which have been incorporated into the otolith during its growth. The most common nuclide pair is $^{226}\text{Ra}$ and $^{210}\text{Pb}$, but $^{228}\text{Th}$ and $^{228}\text{Ra}$ have also been used for shorter lived species. It was pioneered by Bennett et al. (1982), and gained acceptance during the 1980s and 1990s as a validation technique. It is well-suited to long-lived fishes, and has been useful in demonstrating the longevity of some species where there were conflicting interpretations of growth zones resulting in widely divergent age estimates. However, the technique depends on some key assumptions which are not fully resolved. It has relatively low precision, and requires relatively large samples (pooling of several otoliths, or otolith cores).

Following the early studies on supposedly long-lived species, this procedure has been used for a number of taxonomically and ecologically different species, and there have been reviews of either the method (and its perceived shortcomings), or of the results on the range of species so far investigated. These reviews include: Smith et al. (1991), Fenton & Short (1992), West & Gauldie (1994), Kimura & Kastelle (1995), Cailliet et al (1996b), Andrews et al. (1999b), Burton et al. (1999), Campana (1999), and Cailliet et al. (2001).
An unpublished study by Fenton in the early 1990s on radiometric ageing of oreos is considered in Section 2 of this report. Oreo otoliths are particularly small, and several of this study's deficiencies result from the use of pooled whole otoliths instead of otolith cores.

After the original research on rockfishes by Bennett et al. (1982), work on this group of species from moderate depths to deep water was continued by Campana et al. (1990), Cailliet et al. (1996b), Kastelle et al. (2000), and Andrews et al. (2002). The improved method applied by Andrews and Cailliet (MLML) is summarised in Section 3, which includes a publication list of the MLML studies.

Kastelle et al. (1994) used the radiometric procedure to confirm burnt otolith cross-section ages up to 34 years in sablefish, a species also taken in moderate depths.

Radiometric studies on the ages of Australian hoki were reported by Fenton et al. (1990) and Fenton & Short (1995). The earlier study used whole otoliths and was unsuccessful. Fenton & Short (1995) used otolith cores, and obtained ages up to 23 yrs which gave a growth curve similar to that derived from conventional growth zone counts.

Although this method is usually employed for long-lived species, Campana et al. (1993) successfully used it for a tropical flyingfish with a lifespan of less than two years, and observed that it was a promising procedure for other short-lived tropical species with otoliths that are difficult to read by conventional methods.

A number of recent studies have been undertaken on coastal species, either to confirm otolith 'ages' from zone counts, or to determine ages where such counts were difficult and/or ambiguous, such as tropical species. Successful results have been obtained from short-lived as well as long-lived species. Milton et al. (1995) aged three species of tropical snapper from northern Australia. Baker et al. (2001a, 2001b) aged a tropical snapper and red drum from the Gulf of Mexico. Andrews et al. (2001) applied radiometric age determination to the Atlantic tarpon.

Whitehead & Ditchburn (unpublished, 1996) attempted to age New Zealand hapuku, but assumed a regular sequence of concentric otolith growth zones, instead of zones which mainly form on the convex (medial) surface. This simplification largely invalidated their results.

Deepwater oreos have particularly difficult otoliths to read, and the apparently high ages require validation. Australian radiometric studies (Stewart et al. 1995, Fenton, n.d.) are in agreement with zone counts, establishing a maximum age of at least 130 years.

Some grenadiers or rattails also appear, from zone counts, to reach high ages; radiometric studies by Cailliet et al. (1996b) and Andrews et al. (1999a) confirm ages of at least 55 and probably 70+ years.

Orange roughy have high ring counts, suggesting a maximum age of at least 100 and probably over 150 years. Radiometric studies so far undertaken have yielded comparable results (Fenton et al. 1991, Francis 1995b, Smith et al. 1995, Fenton & Short 1997, Allain & Lorance 2000). The reliability of the radiometric method has
been questioned, for orange roughy in particular, by West & Gauldie (1994), Gauldie (1998), and Gauldie & Cremer (1998) but their concerns have not been accepted by other workers using this method (Fenton & Short 1992, 1997; Milton et al 1995; Andrews et al 1999a; Baker et al 2001b).

1.3.10 Elemental and isotopic analysis

Otoliths contain trace elements, and elemental isotopes, which can be interpreted in terms of the fish's life history. The assumption is made that otolith microchemistry is influenced by the water in which the fish lives, and is a record of daily, seasonal, and annual cycles. Numerous studies have made use of this feature: determination of temperature history, sometimes over several generations or even centuries; determination of nursery grounds and subsequent migrations, including anadromy; stock identification; use as a natural tag; and use in age validation. Reviews of the methodology and its application have been written by Thorrold et al. (1997), Campana (1999), Clear & Kalish (2000), and Kalish (1997, 2000).

Otolith isotope records appear most useful in tracking environmental changes during the life of a fish; representative studies include Weidman & Millner (2000), Begg & Weidman (2001), and Andrus et al. (2002).

They are less useful for age determination or validation, apart perhaps from the first year or two where the growth zones are wide. Correlations with early growth zones do occur, but as both are assumed to be environmentally controlled (but may also reflect physiological stress) the isotope record is not an independent validation of age. Campana (2001) notes that chemical cycles within otoliths may be useful for confirming the presence of visually-observed growth increments, but of limited value in inferring what periodicity they might represent. They are likely to be of no value in validating the high ages of long-lived fishes because the growth increments are too narrow. Gauldie et al. (1995) explored age and growth characteristics of orange roughy by comparing strontium and calcium deposition with visible microincrements; their results are complex and unconvincing, but the authors interpret them as suggesting ages much younger than 50 years, rather than the maximum age range of 100–150 years increasingly accepted by other workers.

1.3.11 Bomb radiocarbon

A relatively-recent age validation procedure follows the demonstration by Kalish (1993) that an anthropogenic radiocarbon ($^{14}$C) "signal" formed in fish otoliths between the years 1950 and 1970, when nuclear testing in the Pacific region introduced a pulse of $^{14}$C first into the atmosphere and subsequently the terrestrial and marine environments. This was an extension of work on corals (Druffel & Linick 1978, Druffel 1989). Bomb radiocarbon quickly became incorporated in the carbonate of hard tissues (coral skeletons, fish otoliths) in concentrations proportional to those in the water column. The level of $^{14}$C in otolith cores of old fish, or in whole archived otoliths of young fish, can determine whether the fish were born before, during, or later than the main years (1958–1965) of $^{14}$C increase, using appropriate reference $^{14}$C chronologies for the region. Ages previously determined from growth increment counts can be shown to be either correct or incorrect. This is a powerful validation
procedure, particularly for fish born during the period 1958–1965 (Campana 2001). Published studies use single, birth-year values and a reference chronology; research in progress is directed at defining the $^{14}$C chronology within a single otolith. Studies have also been more successful for fish from shallow to intermediate depths, where the $^{14}$C history is best known, but good inferences can be made for deepwater species.

Reviews of the bomb radiocarbon method are given by Kalish (1995a), and Campana (1999, 2001), and briefer but useful accounts of particular issues are incorporated in most published accounts of individual species.

Coastal species studied have included the New Zealand snapper (Kalish 1993), haddock (Campana 1997), black drum (Campana & Jones 1997), and a tropical snapper (Baker & Wilson 2001).

The only pelagic species to have been studied is the southern bluefin tuna (Kalish et al. 1996).

Studies on species from moderate depths have included the bluenose (Morison & Robertson 1995, 1995b), Australian redfish (Kalish 1995b), and Australian hoki (Kalish et al. 1997). There has been a successful study on the toothfish from Subantarctic waters (Kalish & Timmiss 2001).

Only limited, exploratory studies have been made on ageing orange roughy and the oreo species using the bomb radiocarbon method (Morison et al. 1999, Sparks 2000 unpublished), with problematic results. For orange roughy, Morison et al. (1999) used the "radiocarbon age" method, making the assumptions that the fish lived in a stable deepwater environment, and that no bomb pulse (of elevated radiocarbon, $^{14}$C) would be present. Radiocarbon dating was considered feasible, and of potential value in validating age estimates from increment counts in otolith thin sections. Measurements of radiocarbon were made from individual otolith cores, as well as positions between the core and otolith edge. This study by Morison et al (1999) was considered unsatisfactory by Sparks (2000), as only one of the 8 samples showed fair agreement with the zone counts and the $^{14}$C ages based on the core to otolith edge difference in values. The stable environment assumptions were also criticized by Sparks (2000), as several of the samples were "contaminated" by post-bomb levels of radiocarbon.

Morison et al (1999) also considered that the "bomb chronometer" method of radiocarbon ageing could be used to validate age estimates derived from otoliths of black and smooth oreos. The assumption was that surface-dwelling juvenile oreos would pick up the 1960s bomb pulse in their otoliths (subsequently the cores), and the level of radiocarbon in each core could to used to define the approximate year of birth of that fish, and thus an age. These ages could then be compared with ages derived from growth zone counts in otolith thin sections. Sparks (2000) considered that the data presented by Morison et al. (1999) gave a poor correlation between $^{14}$C and section ages.
1.3.12 Radiometric and bomb radiocarbon studies by year

Both these procedures (radiometric and bomb radiocarbon) are expensive, relative to routine ageing studies based on otolith growth zone counts, and only about 30 have been undertaken during the 1990s, following on from the pioneer radiometric study in 1982 (Table 2). A comparison with routine growth zone studies is not valid. However, the radiometric and bomb radiocarbon techniques are attempts to validate ageing procedures, or at least to determine which (low or high) age interpretation (e.g., from growth zone counts) is more likely to be correct.

Table 2: Number of original radiometric and bomb radiocarbon ageing studies, 1982 to 2001. Critiques and reviews are not included.

<table>
<thead>
<tr>
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<th></th>
<th></th>
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<th></th>
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<tbody>
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<td></td>
<td>1</td>
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<td>2</td>
<td>1</td>
<td></td>
<td></td>
<td>3</td>
<td>2</td>
</tr>
<tr>
<td>Radiocarbon</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>3</td>
<td></td>
<td></td>
<td></td>
<td>1</td>
<td>2</td>
</tr>
</tbody>
</table>

1.4 Age estimation of New Zealand deepwater fish species

1.4.1 Orange roughy

The orange roughy fishery became commercially important in the New Zealand region in 1979, and ageing studies commenced shortly afterwards. Age determination in this species has been extremely difficult; there are many published accounts, plus a number of ‘grey-literature’ reports of research that proved inconclusive. There is general consensus that the species is slow-growing and long-lived (to at least 150 years), but this has been difficult to validate. A review of research to 1990 is included in Paul (1992), see Appendix 1. A later, more comprehensive review was published by Tracey & Horn (1999), see Appendix 2. Subsequent research includes an attempt to estimate of orange roughy recruitment using age- and length-based models, a Northeast Atlantic study to application of the bomb radiocarbon method to validate ages, and ageing studies of orange roughy from the Northeast Atlantic Ocean, Chilean, and Namibian waters. No further routine ageing of New Zealand orange roughy has been carried out at NIWA.
Table 3: Chronological list of ageing studies on orange roughy, *Hoplostethus atlanticus*.

<table>
<thead>
<tr>
<th>Author(s)</th>
<th>Year</th>
<th>Summary of work</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kotlyar</td>
<td>1981</td>
<td>Ages from scale reading (Indian Ocean)</td>
</tr>
<tr>
<td>van den Broek</td>
<td>1983</td>
<td>Ages from otolith zone counts (N.Z.)</td>
</tr>
<tr>
<td>Linkowski &amp; Liwoch</td>
<td>1986</td>
<td>Description of otolith zones (N.Z.)</td>
</tr>
<tr>
<td>Liwoch &amp; Linkowski</td>
<td>1986</td>
<td>Description of otolith zones (N.Z.)</td>
</tr>
<tr>
<td>Gauldie</td>
<td>1987</td>
<td>Fine structure of otolith check rings (N.Z.)</td>
</tr>
<tr>
<td>Gauldie</td>
<td>1988</td>
<td>Surface sculpturing of otolith zones (N.Z.)</td>
</tr>
<tr>
<td>Gauldie et al.</td>
<td>1989</td>
<td>K-selection growth characteristics (N.Z.)</td>
</tr>
<tr>
<td>Gauldie</td>
<td>1990a</td>
<td>Description of otolith zones and check marks (N.Z.)</td>
</tr>
<tr>
<td>Gauldie</td>
<td>1990b</td>
<td>Critique of growth curve based on possible false otolith zones (N.Z.)</td>
</tr>
<tr>
<td>Mace et al.</td>
<td>1990</td>
<td>Juvenile growth, from otolith zones and Lfs (N.Z.)</td>
</tr>
<tr>
<td>Fenton et al.</td>
<td>1991</td>
<td>Ages from radiometric analyses (Aust.)</td>
</tr>
<tr>
<td>Gauldie et al.</td>
<td>1991</td>
<td>Scale morphology and chemistry (N.Z.)</td>
</tr>
<tr>
<td>Smith &amp; Robertson</td>
<td>1992</td>
<td>Review of ageing problems (Aust.)</td>
</tr>
<tr>
<td>Doonan</td>
<td>1993</td>
<td>Use of ages to derive natural mortality (N.Z.)</td>
</tr>
<tr>
<td>Doonan</td>
<td>1994</td>
<td>Derivation of growth parameters from age data (N.Z.)</td>
</tr>
<tr>
<td>Francis</td>
<td>1995a</td>
<td>Otolith mass parameters and radiometric age estimates (N.Z.)</td>
</tr>
<tr>
<td>Francis</td>
<td>1995b</td>
<td>Reinterpretation of radiometric age data to give longevity (N.Z.)</td>
</tr>
<tr>
<td>Francis</td>
<td>1995c</td>
<td>Fish size, age, otolith weight; indicators of biomass depletion (N.Z.)</td>
</tr>
<tr>
<td>Francis &amp; Smith</td>
<td>1995</td>
<td>Fish size, age, otolith weight; indicators of biomass depletion (N.Z.)</td>
</tr>
<tr>
<td>Gauldie et al.</td>
<td>1995</td>
<td>Otolith microincrements, Sr &amp; Ca cycles, and age estimates (N.Z.)</td>
</tr>
<tr>
<td>Smith et al.</td>
<td>1995</td>
<td>Age comparison: zone counts cf. radiometric ageing (Aust.)</td>
</tr>
<tr>
<td>Doonan &amp; Tracey</td>
<td>1997</td>
<td>Use of ages to derive natural mortality (N.Z.)</td>
</tr>
<tr>
<td>Fenton &amp; Short</td>
<td>1997</td>
<td>Radiometric age determination (Aust.)</td>
</tr>
<tr>
<td>Francis &amp; Horn</td>
<td>1997</td>
<td>Otolith zone counts: transition zone and age at maturity (N.Z.)</td>
</tr>
<tr>
<td>Gauldie</td>
<td>1998</td>
<td>Critique of orange roughy ageing methods, short lifespan proposed (N.Z.)</td>
</tr>
<tr>
<td>Gauldie &amp; Cremer</td>
<td>1998</td>
<td>Critique of radiometric ageing: possible loss of Rn (N.Z.)</td>
</tr>
<tr>
<td>Horn et al.</td>
<td>1998</td>
<td>Regional differences in size and age at maturity (N.Z.)</td>
</tr>
<tr>
<td>Tracey &amp; Horn</td>
<td>1999</td>
<td>Detailed review of orange roughy ageing work, to 1997 (N.Z.+)</td>
</tr>
<tr>
<td>Morison et al.</td>
<td>1999</td>
<td>Bomb radiocarbon age determination (N.Z.)</td>
</tr>
<tr>
<td>Sparks</td>
<td>2000</td>
<td>Critique of Morison et al. bomb radiocarbon work (N.Z.)</td>
</tr>
<tr>
<td>Allain &amp; Lorance</td>
<td>2000</td>
<td>Radiometric age determination (NE Atlantic)</td>
</tr>
<tr>
<td>Bull et al.</td>
<td>2001</td>
<td>Orange roughy recruitment using age- and length-based models (NZ)</td>
</tr>
<tr>
<td>Tracey and Clark</td>
<td>2001</td>
<td>Analysis of Indian Ocean orange roughy otoliths (NZ)</td>
</tr>
<tr>
<td>Gili et al.</td>
<td>2002</td>
<td>Age and growth for orange roughy in Chilean waters (Chile / NZ)</td>
</tr>
</tbody>
</table>

1.4.2 Oreos

The black oreo and smooth oreo fisheries became commercially important in New Zealand in the late 1970s. Age determination proved difficult, but there is consensus that both species are slow-growing and long-lived, with growth parameters not unlike those of orange roughy. Most ageing of oreo species has been by otolith zone counts; with a few attempts to validate these by radiometric and bomb radiocarbon methods.
Table 4: Chronological list of ageing studies on oreos, family Oreosomatidae.

<table>
<thead>
<tr>
<th>Author</th>
<th>Year</th>
<th>Species</th>
<th>Summary of work</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mel’nikov</td>
<td>1981</td>
<td>warty</td>
<td>Ages from whole otoliths, scales (S Africa)</td>
</tr>
<tr>
<td>Gauldie et al.</td>
<td>1991</td>
<td>smooth</td>
<td>Scale morphology and chemistry (N.Z.)</td>
</tr>
<tr>
<td>Smith &amp; Stewart</td>
<td>1994</td>
<td>various?</td>
<td>Ages from otolith zone counts (Aust.)</td>
</tr>
<tr>
<td>Doonan et al.</td>
<td>1995</td>
<td>black, smooth</td>
<td>Ages from otolith zone counts (N.Z.)</td>
</tr>
<tr>
<td>Stewart et al.</td>
<td>1995</td>
<td>warty</td>
<td>Radiometric age determination (Aust.)</td>
</tr>
<tr>
<td>Fenton</td>
<td>1996</td>
<td>black, smooth, spiky</td>
<td>Application of radiometric technique (Aust)</td>
</tr>
<tr>
<td>Doonan et al.</td>
<td>1997</td>
<td>smooth</td>
<td>Derivation of growth parameters from age data (N.Z.)</td>
</tr>
<tr>
<td>McMillan et al.</td>
<td>1997</td>
<td>black</td>
<td>Derivation of growth parameters from age data (N.Z.)</td>
</tr>
<tr>
<td>George et al.</td>
<td>1997</td>
<td>smooth</td>
<td>Juvenile growth, from otolith zone counts (S America)</td>
</tr>
<tr>
<td>Morison et al.</td>
<td>1999</td>
<td>black, smooth</td>
<td>Bomb radiocarbon age determination (N.Z.)</td>
</tr>
<tr>
<td>Sparks</td>
<td>2000</td>
<td>black, smooth</td>
<td>Critique of Morison et al. bomb radiocarbon work (N.Z.)</td>
</tr>
</tbody>
</table>

Note:
1. Fenton’s unpublished study is undated; it appears to have started in 1992, with the MS completed in 1996. See Section 2 of this report.

1.4.3 Black cardinalfish

There has been only one study on the age and growth of black cardinalfish, based on otolith zone counts (Tracey et al. 2000). Interpretation of the zones in the whole juvenile otolith samples aided interpretation of the zones in the sections of the adult black cardinalfish. The patterns of zones were reasonably good although toward the edge of the otolith counting was difficult due to the fine spacing of the zones. A slow growth rate and apparent high ages at recruitment (about 45 years) were found. A high maximum age of 104 years was recorded. Adult ages were unable to be validated using the marginal increment technique, however the clear marginal increments visible in juvenile samples provided some evidence of annual periodicity.

1.4.4 Ridge-scaled rattail

A preliminary study of the age of the deepwater ridge-scaled rattail found this species had a maximum age of 55 years (range 9 to 55 y) indicating a reasonably slow growth rate and probable high age at maturity (Marriott & Horn 2000). A change in the zone pattern (i.e. a transition zone) occurred at age 10, from wide zones to regular narrow usually clear zones to the outer margin.

1.4.5 Rubyfish

There has been only one study on the age and growth of rubyfish, based on otolith zone counts (Paul et al. 2000). The zones were narrow, often comprised multiple banding, were interpreted with difficulty, and gave a maximum age of at least 80 years.
1.4.6 Patagonian toothfish / Antarctic toothfish

An ageing study of the deepwater (depth range 70–2000 m) Patagonian toothfish (*Dissostichus eleginoides*) and Antarctic toothfish (*D. mawsoni*) was made by Horn (2002). Counts of the transverse sections of otolith zones showed that Patagonian toothfish is moderately fast growing, at least to about age 10, and reasonably long-lived, reaching at least 50 years. Antarctic toothfish also appears to be moderately fast growing to about age 10, and can live for at least 35 years. Antarctic toothfish probably grow at a slightly faster rate, and reach a larger size, than Patagonian toothfish.

1.4.7 Sea perch

The sea perch (*Helicolenus* spp.) is included here because its depth range extends into deep water (at least 800 m), and because its otoliths have some similarity to those of deepwater species. A preliminary investigation of age and growth, based on otolith growth zones was made by Paul (1998), followed by a more detailed investigation also based on otolith zone counts (Paul et al. 2002). The zones were narrow, often comprised multiple banding, were interpreted with difficulty, and gave a maximum age of at least 80 years.

1.5 Discussion

From about 1980, as commercial fisheries developed on deepwater fish stocks, there has been an increasing number of studies on the age and growth of deepwater species. Most have continued with the established procedures of interpreting and counting growth zones on otoliths, maintaining the assumption that these zones, although narrow and somewhat different from those of shallower water species, represent annual physiological events. ‘Otolith-reading’ quickly moved away from viewing whole otoliths, which studies on many species from all depths showed did not provide complete counts of growth increments, to viewing burnt cross-sections and thin sections (both longitudinal and transverse) of otoliths. Thin sections, although more time-consuming to prepare, have generally proved the most suitable.

Otolith-reading soon revealed that deepwater fishes appeared to reach very high ages, over 100 years in several species. The likelihood that deepwater fishes, in general, live longer was investigated by Cailliet et al. (2001), who found that within a moderately well-studied group, the rockfishes (*Sebastes* spp., family Scorpaenidae) there was a good relationship between longevity and maximum age. Physiological and ecological reasons for this remain unexplained.

The apparent longevity of deepwater fishes indicates relatively low productivity of their stocks, and requires a precautionary approach to setting exploitation levels. This gave an impetus to validating the ages, and to the development of ageing methods which are not reliant on growth increment counts.

The two main methods of age validation, radiometric and bomb radiocarbon, have been successful for several fish species, particularly those of shallow to moderate depths as well as some from deep water (e.g., *Sebastes* spp – rockfishes,
Coryphaenoides spp. - grenadiers) NIWA researchers are optimistic that the radiometric age validation feasibility study of the oreo species, orange roughy and black cardinalfish currently being carried out at MLML will prove useful for these commercially important deepwater species.

2. Summary and critique of Fenton’s report (n.d.) on ageing of oreo species

2.1 Summary

Fenton (n.d.) used a radiometric method, based on the isotopes $^{210}\text{Pb}$ and $^{226}\text{Ra}$, to estimate ages of three species: black oreo (*Allocyttus niger*), smooth oreo (*Pseudocyttus maculatus*), and spiky oreo (*Neocyttus rhomboidalis*). For each species, nine otolith samples were analysed, with each sample weighing about 1 g and containing many otoliths (between 12 and 222) of about similar weight, and from fish of similar lengths (and of the same sex, for samples from mature fish). Activities of the two target isotopes were measured and the mean age of each sample was estimated using a formula which assumes that otolith mass increases linearly with age. Two estimates were obtained for each sample, based on two values of $^{226}\text{Ra}$ activity: the measured rate, and a “mean” rate. Maximum estimated ages were 134 y (or 92 y, using mean Ra activity) for black, 39 y (46 y) for smooth, and 106 y (125 y) for spiky oreos. The precision claimed for these ages is not high, particularly for black oreos, where cited errors are typically 30%, and sometimes more than 50% (these are presumably standard errors, but this is not stated). Estimates of age at maturity (based on published estimates of length at maturity for females) were about 60 y for black, 30 y for smooth, and 10–20 y for spiky oreos.

Fenton also analysed the concentrations of Ca, Sr, Pb, and Ba in the otoliths. She inferred that smooth oreos move from cooler water to warmer water as they age because Sr/Ca ratios were negatively correlated with mean fish length. The Pb and Ba concentrations were very low, often below the detection threshold. This supports the important assumption that that the only substantial source of $^{210}\text{Pb}$ in otoliths is from the decay of $^{226}\text{Ra}$ already in the otolith.

2.2 Critique

There are three main reasons to doubt the age estimates in this report. First, there seems to be no basis for the key assumption that otolith mass increases linearly with age. On p. 20, Fenton comments that most other studies have used a double (broken-stick) model for otolith-mass growth, and are thus based on circular reasoning. She goes on to say that “the data here suggest that was not required”, but it is unclear what data are referred to. The only apparent reason for using the linear model is that it is “simplest” (p. 16). Figures 12 and 13 show apparent linear relationships between otolith weight and age but, as the author admits, the ages used are based on the linear assumption.

A second, and related, problem is that, even if otolith-mass growth is linear, it is most unlikely that the growth rate is the same for all otoliths, as is implicitly assumed (see equation (20), and associated text, in Francis 1995a).
The third problem concerns the Ra measurements, and this problem has two parts. Figure 7 shows that these measurements are correlated with fish length. This appears to violate one assumption of the method, that the rate of incorporation of $^{226}$Ra in the otolith is constant through the life of the fish. Fenton writes, "Because of these [correlations] all age calculations have been presented using both individual $^{226}$Ra and mean $^{226}$Ra levels" but it is unclear how these mean values were calculated or how they fix the problem of a non-constant rate of incorporation of $^{226}$Ra. If this rate increases through life (as it appears to do for black and smooth oreos) then age will be underestimated. The second part of the problem is that some Ra measurements appear to be suspect because of low rates of recovery of $^{133}$Ba (see Table 5 and associated text). It is not clearly stated which readings were considered inaccurate or why.

It should be noted that the first and third of these problems are avoided when, as is proposed for the present study, otolith cores are used in place of whole otoliths.

There are two statistical issues related to regression which are not well dealt with in this paper. First, because there is no testing of statistical significance, some conclusions are not well substantiated. For example, the above inference about smooth oreos moving to warmer water is based on a very small correlation, $r^2 = 0.257$ (Figure 5B), but the standard test fails to reject the null hypothesis of zero correlation (the associated $P$ value is 0.58; to get a significant result ($P \leq 0.05$) for $n = 9$, we need $r^2 \geq 0.444$). Second, the usual regression approach ($y$ on $x$) is not appropriate when, as for most graphs in this report, there are substantial errors in both $x$ and $y$ (the same criticism presumably applies to the method used to fit von Bertalanffy curves in Figure 16, although this method is not described).

Fenton's report appears to have been hastily compiled, without the rigorous review that is normal with formal publication. As a result there are many other minor ways in which the report is unclear (e.g., the text distinguishes between samples of mature and immature fish but there is no indication as to which is which). Had the report been subject to rigorous review it seems probable that it would have been clearer, and that at least some of the problems mentioned above would have been dealt with. There seems no reason to doubt the quality of the radiochemical procedures used because these are similar to those used in earlier published work by the same author.

The copy of Fenton's report reviewed here did not contain the four appendices which are referred to in its text.

3. Radiometric studies at Moss Landing Marine Laboratories

3.1 Further development of the radiometric method

The radiometric method of age validation uses the known decay rates of naturally occurring radionuclides in otoliths (e.g., $^{226}$Ra decays to $^{210}$Pb). The calculated age is then usually compared with ages determined by some other method, usually ring counts.
The earliest studies required moderate sample sizes, and it was necessary to pool several whole otoliths to get sufficient material. This procedure was used by Fenton and co-workers in their studies of orange roughy and oreos (see comment on cores cf. whole otoliths in Section 2).

Current studies at Moss Landing Marine Laboratories (MLML) have overcome many of the earlier difficulties with the radiometric method by using new ion exchange separation procedures and thermal ionisation mass spectrometry (TIMS). These improvements make it possible to apply the technique to smaller sample masses (Andrews et al. 1999b). Consequently, analyses can now be run on otolith cores, rather than whole otoliths, resulting in greater precision. In addition, by using cores, two important assumptions of the radiometric approach are clearly met (see below).

Several scientific papers detailing this method (see appended list below), have been applied to animals such as deep-sea corals and sea pens as well as long-lived fishes, including some from deep water. Details of these radiometric procedures are given in the papers listed in 3.2; they are clearly described in Andrews et al. (2002).

The technique utilizes a known radioactive decay series in the cores of previously aged fish otoliths to provide an independent age estimate. Of the otolith pair collected for each fish, one otolith is aged using a traditional ageing technique such as counting growth zones along a transverse or longitudinal thin section. The other otolith has the outer (younger) portion removed and only the core (approximately the first three years of growth) is used. To obtain enough material the core samples are pooled into age groups.

After cleaning, the pooled cores are then analysed. The method exploits the disequilibria of $^{210}\text{Pb}$ and $^{226}\text{Ra}$ in otoliths as a natural chronometer. The naturally occurring uranium-238 decay series contains the radioisotope pair $^{226}\text{Ra}$ and its daughter product $^{210}\text{Pb}$; the pair used for age determination of long-lived fishes. $^{210}\text{Pb}$ is the more stable isotope, and has a half life of 22.26 years.

Initially the activity from $^{210}\text{Pb}$ is negligible. As the fish ages, $^{210}\text{Pb}$ activity builds in from the decay of $^{226}\text{Ra}$. This process is called "ingrowth". Ingrowth is non-linear and as the activity of $^{210}\text{Pb}$ approaches the activity of $^{226}\text{Ra}$ it levels off and approaches equilibrium at a ratio of 1.0. Age determination from ingrowth, as in the example plotted for the deepwater yelloweye rockfish (*Sebastes ruberimimus*) (Figure 1; Andrews et al. 2002), becomes more uncertain as the ratio approaches 1.0.
Three assumptions are made with the radiometric technique:

1. the calcified structure acts as a closed system for radium and its daughter products.
2. the initial activity ratio of $^{210}\text{Pb}$ and $^{226}\text{Ra}$ in the calcified structure (i.e., during formation of the otolith core) should be negligible or close to zero. This can be cross-checked by analysing very young, known-age otoliths.
3. the uptake rate of $^{226}\text{Ra}$ is proportional to mass growth of the structure during the lifetime of the fish. Because of uncertainty over this, using otolith cores representing only the first few years of growth minimises the effect of variability or trends in uptake rate.

The procedure currently in use at MLML is a more robust radiometric method to age fish because it is based on otolith cores. When otolith cores are used the last two assumptions can be measured or largely circumvented.

### 3.2 Radiometric publications by MLML and associated staff


4. Acknowledgments

We thank Allen Andrews, Moss Landings Marine Laboratories, for assistance with some details of this report. Don Robertson contributed useful suggestions when reviewing the manuscript. This project was funded by the Ministry of Fisheries under Project DEE2000/02.

5. References


**Hoplostethus atlanticus** Orange roughy TRACHICHTHYIDAE

An extremely important deepwater species, whose age and growth characteristics are vital for management decisions. Otoliths have been collected since the late 1970s, but because of numerous crowded rings have proved extremely difficult to interpret. Early unpublished results from New Zealand samples varied widely, and results from elsewhere (e.g., Kotlyar 1981) provided little guidance. Van den Broek (1983) attempted to age from surface ridges on the otoliths, but cautioned that they did not appear annual; his very provisional results gave maturity at 4-5 y, maximum age at 21 y, and von Bertalanffy parameters. Liwoch & Linkowski (1986) presented some biological data on the species, including length frequencies, but conceded failure in ageing attempts. In a more detailed account Linkowski & Liwoch (1986) described the otolith's morphology, including length-weight relationships, and looked for some sensible pattern of annual bands in whole otoliths, broken and burned sections, and thin sections, without success. Sullivan & Parkinson (1987) attempted some exploratory back-calculations from small whole otoliths.

Gauldie began his exploration of the orange roughy otolith at about this time. Ageing the species had high research priority at FRC, as trawl surveys were beginning to suggest a rapidly declining biomass. He used a variety of approaches in all his papers cited below: whole otoliths examined for annual and daily markings in a variety of ways, including SEM, TEM, and high magnification light microscopy, broken surfaces, thin sections, acetate peels, and the crystalline structure and chemistry of the otolith. Gauldie (1987) noted two types of “structural check rings” which might invalidate the normal procedure of counting what appeared to be annual growth zones. Gauldie (1988a) describes surface sculpturing in relation to opaque and translucent zones, concluding (as had van den Broek and Linkowski & Liwoch) that the patterns were complex and unrelated; he also described the difficulties in viewing these otoliths under different conditions, concluding that “three different (and out of phase) [age counts] can be made from opaque zones observed by incident light, ... transmitted light, and ... in sections”. Gauldie & Nelson (1988) postulated that if daily rings were regular there is some potential to estimate ages from the length of the “principal growth axis”. However, in a contemporary paper (Gauldie 1988b) he emphasised the sound-detection purpose of otoliths, and expressed doubt in the validity of check rings for “time-keeping” [ageing]. Gauldie et al. (1989) integrated ages (with a maximum of 18 y) from daily ring numbers estimated along the growth axis, with length frequency data, in an attempt to explain the unusual size frequency curves generally observed for orange roughy. In a largely descriptive paper Gauldie (1990a) concludes that the otolith's crystalline structure complicates age determination from check rings. Gauldie (1990b) included orange roughy in a general comparison of length-at-weight and length-at-age curves for several species, and argues that check rings may represent weight changes, rather than age markers. Gauldie (1990c) briefly discussed the “metabolic work history” of orange roughy otoliths, with only indirect age/growth implications. Gauldie & Nelson (1990b) return to the question of how regular and complete the principal growth axis microincrement are, as well as otolith crystalline structure, and again question the validity of opaque zones and check rings as age markers.
In this whole sequence of papers Gauldie describes the otolith's microstructure in great detail and speculates on its growth characteristics and natural function, but does little to clearly relate the patterns of crystal growth and “daily” microincrements to the structures (in various species as well as orange roughy) to the more prominent features optimistically interpreted by other fisheries workers as annual growth rings. If anything, his latest writings imply great doubt on the value of otolith reading for age, and weaken his argument that the microincrements he described earlier are daily, but they have had little influence on the current efforts by FRC [Fisheries Research Centre, now NIWA] staff to age orange roughy from otoliths.

A different approach was adopted by Mace et al. (1990), who examined whole otoliths under reflected light, with emphasis on a collection taken from very small fish, assumed to be 0+ and 1+, at regular intervals over a one-year period. Two modal length frequency peaks shifted one unit to the right, with a matching unit increase in translucent rings, and partial validation from otolith edge growth. (This work supported the earlier exploration by Sullivan & Parkinson.) Similar rings further out were assumed to be annual also, and were counted with moderate confidence out to 6 or 7, allowing construction of a von Bertalanffy curve. Extrapolation gave an age at maturity of about 20 y, and a maximum age over 50 y. Otoliths with ring counts over about seven are ambiguous, but efforts are continuing at FRC [NIWA] to validate their ageing.

References


Appendix 2: Review of orange roughy ageing studies (Tracey & Horn 1999).

0028-8330/99/3301-0067 $7.00 © The Royal Society of New Zealand 1999

Background and review of ageing orange roughy (Hoplostethus atlanticus, Trachichthyidae) from New Zealand and elsewhere

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Abstract  Studies on New Zealand orange roughy (Hoplostethus atlanticus) otoliths, and of orange roughy ageing conducted in New Zealand and elsewhere are described. Ageing studies have concentrated on three aspects: the interpretation of daily growth increments, the interpretation of annual growth increments, and radiometric analyses. All the methods suffer from problems relating to validation. Daily growth zones have not been validated, annual zones have been validated for juvenile fish only, and assumptions necessary for the application of radiometric techniques may be flawed. However, the weight of current evidence indicates that orange roughy are a slow-growing, long-lived species. A review of otolith morphology and microstructure studies, and a summary of the productivity parameters used in stock assessments of orange roughy, are also presented. Standard protocols used to prepare and interpret otoliths in current investigations are described. This review highlights the complexities of ageing long-lived, deepwater fish, and stresses the importance of obtaining accurate productivity parameters for stock assessment. The key area for future research is the age validation of post-juvenile fish, which should lead to the development of an accurate ageing technique.

Keywords orange roughy; Hoplostethus atlanticus; review; ageing techniques; growth; otolith; age validation

INTRODUCTION

Orange roughy (Hoplostethus atlanticus Collett 1889) is one of New Zealand’s most important commercial finfish occurring in depths between 700 and 1500 m throughout much of the New Zealand 200-mile Exclusive Economic Zone (EEZ). Target trawl fisheries commenced c. 1979 and now occur in several offshore areas (Fig. 1). Commercial fisheries for this species also developed off south-eastern Australia c. 1989, in the north-east Atlantic Ocean c. 1990, and off south-west Africa in 1995 (Fig. 2).

In New Zealand, the orange roughy fishery developed on the Chatham Rise in 1979 (Annala et al. 1998). Other fisheries developed in the late 1980s on the south-west Challenger Plateau, off the central west coast of the South Island, and off the east coast between Gisborne and Kaikoura. More recently, commercial operations have been targeting orange roughy in other areas including near East Cape, Bay of Plenty, the Puysegur Bank, and the Macquarie Ridge.

Total annual landings from the New Zealand EEZ peaked at c. 50 000 t in the mid 1980s, but have subsequently declined to a current catch of c. 20 000 t. The progressive lowering of catch quotas has been based on scientific evidence which revealed a rapid decline in biomass (Clark 1995a; Francis et al. 1995), and on early ageing studies (Mace et al. 1990) which suggested a very slow growth rate, and hence, low productivity. Approximate peak annual landings levels in other fisheries are 40 000 t off south-eastern Australia, 12 000 t off south-west Africa, and 4000 t in the north-east Atlantic Ocean.

Information on age and growth is fundamental to the management of any fish stock. Although numerous researchers have attempted to use otoliths to interpret the ages of orange roughy with the intention
of developing growth parameters, the task has proven very difficult. The rate of orange roughy growth up to 3 years has been validated (Mace et al. 1990) but a validated technique to age adults of this species has yet to be achieved. However, investigations since the mid 1970s have produced a considerable body of data concerning orange roughy ageing, generally indicating a slow growing, long-lived species.

Paul (1992) reviewed fish age and growth studies of New Zealand marine fish and briefly summarised early work on orange roughy. A summary by Merrett & Haedrich (1997) of available information on orange roughy ageing noted the difficulties encountered in various studies. They concluded that although most of the evidence suggests that orange roughy are slow-growing, the debate about their longevity was still not settled. The current report summarises in detail studies on New Zealand orange roughy otoliths, and of orange roughy ageing in general.

Standardised otolith terminology (Secor et al. 1995) has been used throughout this report unless directly quoting other authors' work. Zones viewed on sectioned and whole otoliths illuminated by transmitted light are either opaque (the zone that resists the passage of light and appears dark) or translucent (the zone that allows the passage of greater quantities of light, appears bright). The term hyaline is synonymous with translucent, but the latter
is preferred. Unless otherwise stated, all references to orange roughy length denote standard length (SL), the measurement from the snout to the base of the caudal peduncle.

**ORANGE ROUGHY OTOLITH**

Orange roughy otolith shape is unusual and highly variable. Nolf (1985) describes obvious ontogenetic changes where the whole rim shape and sulcus pattern alters with fish size. Otoliths of juvenile fish (<10 cm) display a smooth oval shape exhibiting little morphological structure (Fig. 3A). In prerecruits and adults the morphological structure includes a pronounced rostrum separated from the antirostrum by a large excisural notch (Fig. 3B). The posterior is very blunt. Linkowski & Liwoch (1986) described the morphology over a range of sizes of orange roughy otoliths collected on the Chatham Rise and Challenger Plateau. They showed otolith shape to vary greatly between individuals (and occasionally, between otoliths of a single individual).

Smith & Robertson (1992) examined all bony parts of orange roughy to test their suitability as material to age fish. Generally all, except otoliths, showed little structure or zonation useful for ageing.

**EARLY STUDIES OF OTOLITH ZONES**

The first published study of orange roughy ageing was by Kotlyar (1981) who examined otoliths and scales from fish caught in the Indian Ocean (West Australia Ridge and Madagascar Ridge) and the North Atlantic Ocean (Irish continental slope). No useful zonation pattern was observed on scales. Zones were apparent in otoliths when they were examined whole in glycerine, under reflected light. No sectioned otoliths were examined. Kotlyar concluded that zones were clearer if the otoliths had been stored in ethyl alcohol with a little glycerine, rather than dry. Otoliths from fish 7–58 cm were examined, although over 95% of the fish were smaller than 32 cm. Kotlyar reported an age range of 1–24 years, and a slow growth rate of 2–3 cm per year after a first year's growth of 6–7 cm. From photos presented in the paper, it is apparent that the first translucent zone on the otolith was ignored in all counts.

Van den Broek (1983) attempted to interpret New Zealand orange roughy otoliths using several preparation techniques, i.e., thin sections through both baked and untreated otoliths, breaking the otolith through the nucleus and examining the broken surface after heating it in a spirit flame, and
Fig. 3 Distal face of whole orange roughy (*Hoplostethus atlanticus*) otolith from: A, a juvenile; B, an adult showing the position of a thin section; and C, a longitudinal thin section through an otolith from an adult fish viewed with transmitted light and showing a clear example of the transition zone (arrowed).

examining whole baked or untreated otoliths under water in a black dish. He concluded that examining whole, untreated otoliths was the best method. A series of ridges on the otolith surface were noted and assumed to represent episodes of growth, though they were probably not annual. Hence, he concluded that multiple rings were laid down in the first few years, and interpreted the most pronounced rings as annual. The ring pattern was considered to become more conventional (i.e., one ring per year) after maturity. Based on this interpretation of 643 otoliths from fish with lengths ranging from 9 to 42 cm, van den Broek concluded that orange roughy grew to c. 18 cm in 2 years, matured at c. 4–5 years, and lived to c. 21 years. He presented preliminary von Bertalanffy parameters. In summary, van den Broek (1983) noted numerous zones in the otoliths, but assumed that multiple bands were laid down in the first few years.

Attempts to age a small sample of New Zealand orange roughy using otoliths were also described by Linkowski & Liwoch (1986). Otoliths had been stored dry. They investigated two preparation techniques; whole otoliths after immersed the otolith in water for 20 h, and whole baked otoliths. In both instances otoliths were illuminated by both transmitted and reflected light and all preparations were immersed in glycerine during observation. The
method which best allowed the internal structure to be viewed was soaking whole, untreated otoliths in water and examining the lateral surface under either reflected or transmitted light. Linkowski & Liwoch reported numerous clear rings, but noted that they exhibited large variations in width and that ring width did not appear to decrease uniformly towards the otolith edge. Complex structure within individual rings was also observed. Hence, they concluded that the well-developed zones did not correspond to regular periods of growth. Some zone counts were obtained but this process was deemed a failure as readings for an individual otolith could differ by as much as 100%.

Sullivan & Parkinson (1987) examined 109 otoliths from orange roughy caught in the Bay of Plenty, and calculated a linear relationship between fish length and otolith length. This regression equation was then used to predict the length of orange roughy from the size of the first four growth check zones in 66 whole otoliths. The first zone was a fairly constant size, and it was predicted that a fish would be c. 3 cm at its time of formation. The spread about the mean size of a zone increased with increasing zone number, but distinct peaks in the predicted length distribution were still produced. They concluded that fish length at the completion of the first four zones would be c. 3, 5, 7, and 8.5 cm, respectively, and noted that if these zones were formed annually then a very slow growth rate was indicated for this species. An estimate of 40 years to reach 40 cm, and an instantaneous natural mortality rate \( M \) of less than 0.1, were suggested. Sullivan & Parkinson (1987) were the first to provide data pointing to the slow growth rate and long life of orange roughy.

Williams (1987) described two ageing methods for a sample of New Zealand orange roughy. The first was to examine whole otoliths in water and count and measure translucent and opaque bands that were presumed to be annual. Band counts ranged from 7 for a 16.4 cm fish, to 15 for the largest fish examined (40.4 cm). The second approach was to section otoliths embedded in resin, and then read and measure the widths of assumed daily growth increments (DGI). Williams found it difficult to distinguish separate presumed annual groupings when examining translucent and opaque markings on the sectioned otoliths. Also, there did not appear to be any distinction between fast growth and slow growth for this species as growth rates appeared to be the same between opaque and translucent zones. Williams (1987) obtained conflicting estimates of age when he compared assumed annual banding structure on whole otoliths with presumed daily growth increments on sectioned otoliths. Translucent-opaque zone counts showed no correlation with the DGI data, and he suggested that interrupted growth may have resulted in fewer than 365 DGIs being deposited in any one year. The study was considered inconclusive.

The first study to provide a partial validation of the rate of orange roughy growth was by Mace et al. (1990) who examined whole otoliths from small Chatham Rise fish (<14 cm SL). Samples were obtained from trawl surveys conducted in February,
May, and September 1988. Translucent zones were counted, the distances from the primordium to each zone were measured, and the otolith margin was classified as either translucent or opaque and wide or narrow. There was a distinct relationship between peaks in the length frequency histograms and translucent zone count for fish with 0–3 zones (Fig. 4). The analysis of the otolith margins showed that fish began laying down a new translucent zone between February and May and it continued forming through to September. Mace et al. (1990) concluded that "the most parsimonious interpretation" was that the first three translucent zones were formed annually. The rate of progression of length frequency peaks also supported the hypothesis that there was an annual deposition of translucent zones. It indicated that average lengths after 1, 2, and 3 years of growth were 3.1, 5.5, and 7.6 cm, respectively. Counts out to about seven zones could be made with moderate confidence, after which interpretation became increasingly uncertain. However, von Bertalanffy parameters were calculated using data for 242 fish ranging from 2 to 38 cm, and aged from 0 to 29 years. Extrapolating from the growth curve, an average age at maturity of c. 20 years (from a length of 30 cm), and a maximum age in excess of 50 years were proposed. These values indicated that $M$ was probably less than 0.1.

In summary, early studies to age orange roughy examined both whole and sectioned otoliths and concluded that sections were not useful (Kotlyar 1981; van den Broek 1983; Linkowski & Liwoch 1986). Although these studies dealt mainly with ageing problems, most also suggested ages and a growth rate. Kotlyar (1981) was close to correctly describing the rate of growth in the first few years, but he ignored the first translucent zone, and consequently combined 2 year's growth into one. Linkowski & Liwoch (1986) rejected the zones as annual phenomena because they did not fit with the "classical" pattern of growth zones in otoliths, i.e., they varied in width, and did not uniformly decrease in width towards the otolith edge. Van den Broek (1983) noted numerous zones, but assumed that multiple bands were laid down in the first few years. These authors constrained themselves by trying to interpret the otolith zonation pattern as they would an otolith from a fish with moderate growth and longevity. Sullivan & Parkinson (1987) were the first to indicate a slow growth rate and long life for orange roughy. Their conclusions were confirmed by Mace et al. (1990) who validated the first 3 year's growth, and indicated that translucent zones were laid down annually in otoliths.

**OTOLITH MICROSTRUCTURE**

In a sequence of papers, Gauldie described the microstructure of orange roughy otoliths in great detail and also speculated on their growth characteristics. He used a variety of techniques in the works cited below: whole otoliths examined for annual and daily markers using electron and light microscopy; broken surfaces; thin sections; and acetate peels. Crystalline structure and otolith chemistry were also investigated. Gauldie (1987) described two kinds of check rings: "diffuse deposits" which were not visible on otoliths which had been broken through the nucleus and burned, and "structural check rings" which were represented by fine lines on the surface and dark bands in the body of the otolith. He found no relationships between the frequency of the two types of band, and noted that the structural check rings were deposited irregularly. He concluded that the diffuse deposits were most likely related to growth in the conventional sense, but that they were hard to see. He reported two types of "structural check rings", those with protein and those without, which combined to give rise to the appearance of "conventional check rings", but claimed they were not related to age. Although it is not clear from Gauldie's descriptions, these are probably the zones that were later validated as being formed annually (Mace et al. 1990).

Gauldie (1988a) examined the surface sculpturing in relation to opaque and translucent zones, and reported that some otoliths exhibited opaque zones with little or no related surface sculpturing, whereas others showed opaque zones both related and unrelated to surface structure. He noted that there were difficulties in viewing otoliths under different conditions and concluded that different opaque zone counts could be obtained when observing otoliths under incident light, transmitted light, and in section.

Gauldie (1988b) presented relationships between fish length and otolith length and weight, showing that fish and otolith lengths were linearly related over the length range 6–34 cm. He concluded that none of the check rings in otoliths were valid for "time-keeping" (ageing) in orange roughy. However, in a contemporary paper, Gauldie & Nelson (1988) reported regular microscopic growth increments of varying width (1–5 μm) and suggested they were daily zones, based on a conclusion that daily
increments were an obligatory feature of otolith growth. They postulated that if daily rings were regular, they could be used to estimate fish age by measuring the length of the “principal growth axis” (presumably the longest radius from the nucleus to the margin) of the otolith. This ageing method was described in a manuscript (Gauldie 1988c) that was formally cited in several of Gauldie’s papers, but never published. Essentially, a section of assumed daily growth increments were measured to obtain a mean increment width, which was then related linearly to the length of the principal growth axis. Gauldie et al. (1989) proposed ages for orange roughy up to about 20 years, based on the daily growth zone method described above, although the validity of the zones as daily growth checks has never been established. This work looked at the theory of dissipative structures (Johnson 1981) and analysed length frequency structures and length-at-age to explain why the size-frequency distributions observed for this species had remained unchanged despite apparent marked declines in biomass. However, one of the authors’ basic assumptions was that they knew the age structure of the populations, and this is flawed at least for the youngest fish. Based on assumed daily growth increments, they concluded that 2+ and 3+ fish had length ranges of 10–13 and 12–16 cm respectively, which differs from the ranges of 4–9 and 6–11 cm for 2+ and 3+ fish derived from the validated growth in Mace et al. (1990). Gauldie et al. (1989) claimed that the stability of the “intermediate” length frequency modes (and instability in modes of large and small fish) argued against extreme ages and slow growth, yet their length distributions exhibit a clear stability in the length of the largest mode.

Gauldie (1990a) described the crystalline structure of orange roughy otoliths. He concluded that crystal growth differed in different parts of the otolith, and postulated that this could explain why his age determinations from check rings differed from counts along an otolith section. Gauldie (1990b) compared length-at-weight and length-at-age curves for several species (including orange roughy) and argued that otolith check rings represented changes in fish weight rather than age markers. Gauldie & Nelson (1990) further discussed crystalline structure, and concluded that microincrements fluctuated widely in width and were deposited at different rates across the otolith (hence questioning the validity of these zones as accurate age markers).

Gauldie et al. (1991) described the morphology and chemistry of orange roughy scales and proposed that patterns of calcium, fluorine, carbon, and oxygen across scale sections could be used to age the fish. They concluded that the fish grew to c. 20 cm in 2 years and 35 cm in 7 years.

A microprobe analysis of cycles of calcium and strontium in orange roughy otoliths was reported by Gauldie et al. (1995). They sampled at 200 points across an 8 mm radius, but then smoothed the data using an 0.8 mm window. Periodicities in chemical composition were concluded to be unrelated to patterns of “checks” in the otolith, but similar to patterns of microincrement widths (though the mean number of microincrements in a cycle was only 252). The authors claimed that a count of assumed daily increments was consistent with an interpretation of otolith strontium variation as an annual growth marker.

Romanek & Gauldie (1996) described a model that predicted otolith growth rate (i.e., the width of a daily increment) as a function of endolymph chemistry (expressed as a saturation rate) and temperature. On the basis of the model they concluded that orange roughy could not be as old as 130 years as inferred from the radiometric method of Fenton et al. (1991) because the chemistry of the endolymphatic fluid surrounding the otolith implied that the otolith of a fish of that age would be very much bigger than is observed. However, their model produces estimates with very wide bounds (almost two orders of magnitude), and is based on what they described as “almost non-existent” observations of pH in fish endolymph (a parameter to which the model is very sensitive). Also, the temperature at which orange roughy live is outside the range covered by the observations on which the model is based. The modelled endolymphatic chemistry is assumed to be simple, but the authors admit that some non-modelled components may impart some control on crystal growth.

Gauldie’s series of work described the crystal structure in detail and speculated on the natural function of otoliths, but did little to establish a method to age orange roughy. Gauldie (1988a,b) concluded that counts of otolith growth checks were probably not a valid method to age orange roughy. He claimed to have identified daily rings (Gauldie 1988c), and estimated fish age by applying the mean width of a daily ring to the “principal growth axis” (Gauldie et al. 1989). However, in Gauldie & Nelson (1990) he appears to question the validity of daily growth rings. No formal validation of daily growth rings has ever been published.
RECENT DEVELOPMENTS IN OTOLITH INTERPRETATION

Following the confirmation that zones are formed annually in orange roughy otoliths (at least in the first few years of life), New Zealand studies on the growth of adult fish concentrated on zone counts in otolith sections. This work aimed to provide a refined reading protocol, revised productivity parameters, and a more complete validation of growth.

Estimates of productivity parameters

Otolith zones were counted on samples of pre-recruit (<32 cm) fish from the East Coast and pre-recruit and adult (>32 cm) fish from Chatham Rise, Challenger Plateau, and East Coast North Island fisheries. Interpretation of presumed annuli for fish >10 cm was difficult. Otoliths from adult fish were read whole and a small number (n = 25) compared with readings from broken and burnt otoliths from the same fish. There was poor agreement (<60%) when comparing the readings from these two methods, and presumed annual zone counts were concluded to be unreliable (Gabriel in: Annala & Tracey 1989).

Because interpreting growth zones in older fish appeared to be unreliable, effort focused on other techniques to obtain information on age and growth. A project was proposed primarily to develop a multivariate model to estimate orange roughy age based on objective measurements of otolith dimensions (Kalish et al. in: Annala 1993). A random sample of 447 otoliths was collected during a trawl survey of the Chatham Rise in winter 1990. Otoliths were weighed and measurements taken of the otolith width, length, area, and perimeter using a computer based video microscopy system (Image Analysis). These variables were analysed to see if any were useful in a predictive model to estimate age. Von Bertalanffy parameters were estimated using the otolith weight data, with $L\text{\lowercase{w}}$ being estimated as the asymptote of the curve describing mean length as a function of otolith weight (Francis et al. 1993).

By 1992, a technique had been developed to produce fine longitudinal sections of orange roughy otoliths (Appendix 1) which enabled counts of opaque and translucent zones. The mean distance from the primordium to the third annual zone was known from the measurements made on whole juvenile otoliths by Mace et al. (1990), and this was used as a guide for readers counting these zones on the adults. About 400 otoliths from the 1990 Chatham Rise survey were sectioned and examined by two readers to obtain maximum age estimates. Consistency between readers was examined by Doonan (in: Clark & Hurst 1994). For ages less than 15, there was no bias. Between 15 and 60, there was 8% bias between readers. After 60 years, bias increased. About 10% of the differences in ages between the two readers were greater than 20%. Only 9% of the readings were the same. The Beamish & Fournier (1981) index of average precision was 5.7%. These results were not used to revise age parameters owing to the perceived high level of between-reader variability. Subsequent effort focused on developing a standard reading protocol. An analysis of the relationship between otolith weight and estimated age (which would be expected to intersect near the origin), lead to a reinterpretation of juvenile growth.

Otoliths of 432 adult fish sampled from a 1984 survey on the Chatham Rise were sectioned and read in 1994. Using a standard reading protocol ages were more consistent between readers for this sample. These data enabled revised estimates of $M$ (0.045) and growth parameters ($L\text{\lowercase{w}} = 36.4$ for males and 38.0 for females, Fig. 5) (Doonan 1994). The previously aged 1990 sample was re-read and produced ages c. 10% older than those obtained initially (Tracey et al. in: Clark 1995b).

The estimate of $M$ based on otolith readings from the Chatham Rise population in 1984 was possibly underestimated because exploitation of that stock before 1984 could have selected out older fish (Clark 1996). A new estimate was obtained from two sets of readings from the relatively unexploited population of orange roughy in the Bay of Plenty area (Doonan & Tracey 1997). Between-reader variability was estimated to be 7.2%, and age was taken as the average from the two readers. $M$ was estimated to be 0.037 and was not statistically different from the estimate derived using the Chatham Rise data.

![Fig. 5 Fit of the estimated growth to the Chatham Rise orange roughy (Hoplostethus atlanticus) data, by sex. (SL = standard length.)](image-url)
Estimation of maturity and recruitment parameters

To investigate recruitment variability, a random sample of 220 pre-recruit fish (<31 cm) from off east coast North Island was aged. There was a possible mode at 12 years, but it considered unlikely that this mode could be followed between years owing to reader variability and sample variance (Tracey et al. in: Clark 1996).

Kalish et al. (in: Annala 1993) noted that many orange roughy otoliths exhibited a "transition zone" (Fig. 3C) where the width of the assumed annuli decreased markedly at a point where the otolith surface begins curving. This transition zone, which was presumed to represent the age at maturity, has since been described as the point on the otolith where zone width decreases markedly from relatively wide, dark opaque zones to a pattern of fine, lighter zoning (Francis et al. 1993; Doonan 1994; Francis & Horn 1997). The position of the transition zone was independently determined and its distance from the primordium measured by three readers on a sample of whole otoliths: Counts (assumed annual) were then made of zones inside the transition zone on thin longitudinal sections of these otoliths. Age at maturity was revised from 23 to 22.2 years based on these data (Francis et al. 1993).

Francis & Horn (1997) tested the hypothesis that the transition zone forms at the time of first maturity by examining otoliths from fish which had had their sexual maturity stage classified. They found that for fish sampled on the Chatham Rise during the spawning season, most otoliths of immature fish did not have a transition zone and most mature fish otoliths did. For females, the occurrence of transition zones was much higher in those actively spawning than in those maturing but not spawning that season. Mature fish with otoliths that were classified as not having the zone tended to be small. It was suggested that small fish were likely to have been mature for fewer years than large ones, and hence, their transition zone might be harder to detect because it would have fewer annuli outside it. In summary, the data collected during the spawning season were consistent with the hypothesis that the transition zone marks the onset of maturity, after allowing for some error in gonad staging and identification of the zone in recently mature fish. Readings to the transition zone provided estimates of age at maturity of 30 years for both males and females (Francis & Horn 1997).

Horn et al. (1998) carried out a study looking at the differences in age and length at first maturity of orange roughy populations in New Zealand waters (Fig. 1). Counts of the number of opaque zones inside the transition zone were used to estimate these parameters. Samples from Namibia and north-west Atlantic (Hatton Bank) were included in the analysis, and results were compared with the Australian estimates compiled by Bax (1997). Significant between-area differences were apparent for the mean age and length at maturity (Table 1). The authors showed a proportional relationship between age at onset of maturity and the modal size of adult fish. A greater onset of maturity was associated with a greater modal length, and this is probably linked to the extra years of faster, pre-reproductive growth experienced by the later-maturing populations.

North Atlantic orange roughy often attain standard lengths between 50 and 65 cm (Charuau et al. 1995), in comparison with New Zealand orange roughy which have an adult population size range

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<td>Hatton Bank</td>
<td>44</td>
<td>32.2–46.5</td>
<td>40.80</td>
</tr>
<tr>
<td>Tasmania</td>
<td></td>
<td>34.0</td>
<td>34.0</td>
</tr>
</tbody>
</table>
between 25 and 40 cm. Charuau et al. (1995) presented some age keys derived from otoliths of young (13–35 cm) fish, but concluded that because of the wide range of ages (±5 years) for lengths >24 cm it was impossible to age orange roughy. Despite the widely scattered ages, the data points indicated a growth rate comparable to that presented by Smith et al. (1995) for Australian orange roughy up to c. 30 cm. Nine large otoliths from North Atlantic fish of unknown length were sectioned and aged using the New Zealand reading protocol (Tracey et al. in: Clark 1995b). Four were aged older than 100 years, with one producing estimates between 156 and 174 years.

**Between-institution comparisons of readings**

The zonation pattern in orange roughy otolith sections can be complex and confusing. However, Smith & Robertson (1992) carried out a between-reader comparison on whole Australian otoliths and concluded that counts could be consistent, and thus, that the level of precision was relatively high. Smith et al. (1995) had 90% of paired readings being within ±10 zones and their estimates had a Beamish & Fournier index of 4.4%. Readings from a single set of otoliths by two readers from NIWA, New Zealand, and one reader from the Central Ageing Facility, Marine and Freshwater Resources Institute, Australia, were compared.

Initially, comparisons of New Zealand and Australian readings of a 1984 Chatham Rise sample (n = 50) were made (Tracey et al. in: Clark 1995b). Results showed that the median of total counts for the Australian reader was 6 years greater than those of the New Zealand readers, but the median of the transition zone counts was 2 less than the New Zealand median. Following these readings, the New Zealand reading protocol was revised as described above.

A sample of Australian-prepared otoliths was then obtained for reading comparisons. The resulting data were combined with those from the initial comparisons of New Zealand prepared otoliths (but with revised New Zealand readings). Otoliths that either reader considered had a readability category of 4 (considerable doubt about zone count) or worse, were excluded from the comparison. Otoliths were prepared at both laboratories, and after exclusions, gave a sample size of 67 (25 Australian fish and 42 from New Zealand). Counts were made from the nucleus to the transition zone, and from the transition zone to the otolith margin. Some slight differences in interpretation are apparent between institutions. In 90% of instances, both readers agreed whether the otolith did (n = 51) or did not (n = 9) have a transition zone. However, in six of the seven disagreements, the New Zealand reader identified a transition zone whereas the Australian reader did not. A summary of the differences in zone counts between readers is presented in Table 2. A bias is apparent, with the Australian reader producing slightly higher pre- and post-transition zone counts. The overall mean difference between readers is only 1.6 years for the pre-transition, and 1.5 years for the post-transition counts, although the average of the absolute difference for the total count is 6.3 years. Most of the fish in the sample were aged in the range of 30–60 years, so an overall mean difference between readers of c. 3 years equates to 5–10% of total age. A comparison of the plots of total age against length for each reader (data not presented here) showed no systematic differences, though the sample size was small.

**Table 2 Between-reader comparisons of 67 orange roughy (Hoplostethus atlanticus) otoliths. Comparisons are shown for pre-transition, post-transition, and total zone counts. Difference, the extent by which the age allocated by the NIWA reader differed from that of the CAF reader.**

<table>
<thead>
<tr>
<th>Difference</th>
<th>Pre-transition</th>
<th>Post-transition</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>+(16–20)</td>
<td>0</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>+(11–15)</td>
<td>0</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>+(7–10)</td>
<td>1</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>+6</td>
<td>0</td>
<td>3</td>
<td>1</td>
</tr>
<tr>
<td>+5</td>
<td>0</td>
<td>3</td>
<td>0</td>
</tr>
<tr>
<td>+4</td>
<td>1</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>+3</td>
<td>1</td>
<td>2</td>
<td>4</td>
</tr>
<tr>
<td>+2</td>
<td>4</td>
<td>5</td>
<td>4</td>
</tr>
<tr>
<td>+1</td>
<td>7</td>
<td>3</td>
<td>5</td>
</tr>
<tr>
<td>0</td>
<td>13</td>
<td>12</td>
<td>2</td>
</tr>
<tr>
<td>-1</td>
<td>7</td>
<td>4</td>
<td>2</td>
</tr>
<tr>
<td>-2</td>
<td>11</td>
<td>3</td>
<td>2</td>
</tr>
<tr>
<td>-3</td>
<td>7</td>
<td>3</td>
<td>6</td>
</tr>
<tr>
<td>-4</td>
<td>4</td>
<td>3</td>
<td>5</td>
</tr>
<tr>
<td>-5</td>
<td>6</td>
<td>2</td>
<td>8</td>
</tr>
<tr>
<td>-6</td>
<td>1</td>
<td>0</td>
<td>3</td>
</tr>
<tr>
<td>-(7–10)</td>
<td>2</td>
<td>8</td>
<td>13</td>
</tr>
<tr>
<td>-(11–15)</td>
<td>2</td>
<td>6</td>
<td>8</td>
</tr>
<tr>
<td>-(16–20)</td>
<td>0</td>
<td>2</td>
<td>2</td>
</tr>
</tbody>
</table>
Fig. 6  Normalised length frequency distributions for orange roughy (Hoplostethus atlanticus) with otoliths exhibiting 0–5 translucent zones, from 10 trawl surveys combined (Doonan unpubl. data). Lengths were made relative to the median length of the group with one ring in each sample so that the effects of seasonal growth from the different sampling times were cancelled out. This is why the mode for 1 ring sits on 0.

Age validation studies
Trawl surveys to obtain samples of small orange roughy were conducted on the Chatham Rise in 1989 (January, May, August, October, and December) and 1990 (May) with the intention of extending the study by Mace et al. (1990). Subsequently, the first 4 years of orange roughy ages, estimated from translucent zones in whole otoliths, were validated using the seasonality of otolith edges over a 2-year period and by tracking the progression of unimodal length-at-age distributions (Doonan unpubl. data). This work confirmed the estimates of size-at-age in Mace et al. (1990) and indicated that a 5-year-old fish would be c. 12.4 cm (Fig. 6). Inter-annual variations in total growth of c. 18% were apparent. Growth also varied seasonally, with rates being c. 3 times higher in summer (January–February) than in winter (late May–early September).

**RADIOMETRIC STUDIES**

The technique of using decay rates of the naturally occurring radionuclides in otoliths ($^{226}$Ra decays to $^{210}$Pb at a known rate) to age fish (Bennett et al. 1982) was first applied to orange roughy by Fenton et al. (1991). Because of a required minimum sample of 1 g, individual samples comprised multiple otoliths ($n = 2–135$) pooled on the basis of similarity of fish length and otolith weight. The extent of the variation in fish length and otolith weight within samples was not stated, but the mean fish length of individual samples ranged from 11 to 40 cm. To avoid any complications as a result of sexual differences in growth rate, all otoliths from mature fish were from females. The authors calculated two estimates of age for each sample: one based on a constant relationship between fish age and otolith weight (3.94 mg yr$^{-1}$), and the other based on a "broken stick" relationship where annual otolith weight gain decreased after maturity to an arbitrarily chosen value of 1.77 mg yr$^{-1}$ (45% of the pre-maturity growth rate). However, they concluded that the constant growth model was not appropriate for orange roughy, so they used the broken stick model with an assumed age at maturity of 32 years. Based on this analysis, they stated that orange roughy is a very slow-growing and long-lived fish, with individuals 38–40 cm being 77–149 years old.

West & Gauldie (1994) criticised the application of the $^{210}$Pb-$^{226}$Ra disequilibrium method to age orange roughy. They maintained that the gas $^{222}$Rn could diffuse out of the otolith, that the sources and sinks of $^{210}$Po, $^{210}$Pb, and $^{226}$Ra could vary over the lifetime of the fish, and that the radiometric ages depended on an assumed otolith mass growth model derived from some other ageing method thus involving a circular argument.

Smith et al. (1995) presented data from sectioned otoliths which confirmed the two-stage otolith mass growth model, and indicated that the pre-maturity growth rate was 5.3 mg yr$^{-1}$, compared with 3.3 mg yr$^{-1}$ after maturity. Their age data were obtained from annulus counts, which indicated an age at maturity of c. 25 years (based on a length of 30–32 cm). This new information enabled the reanalysis of the radiometric data of Fenton et al. (1991), and had the effect of reducing the calculated ages, i.e., fish 38–40 cm were 59–101 years old.

Francis (1995a,b) provided a new interpretation of the radiometric data used by both Fenton et al. (1991) and Smith et al. (1995). He presented two analyses: estimates of “most probable” ages which...
avoided the difficulty of needing to specify otolith mass growth rates in order to apply radiometric methods; and a lower bound on the maximum age of fish in the sample. The most probable ages for 38-40 cm fish were 95-194 years. However, Francis drew attention to the large standard errors around these estimates and noted that they did not include uncertainty related to the otolith mass growth rates. Thus, although the analysis produced most probable ages it gave little information about confidence intervals for these ages. The problem of between-individual variability in otolith mass growth rates could be overcome by using otolith cores (see Campana et al. 1990), rather than complete otoliths. This problem is also avoided in the second analysis to produce a lower bound for the maximum age. Francis showed that 84 years was the lower 95% confidence bound for the maximum age of the sample. He stressed that as a lower bound for the population, that value must be viewed as conservative. He also showed that the estimate is not much affected if a 10-fold increase in uptake of $^{210}\text{Pb}$ at maturity is assumed.

The analyses presented by Francis (1995a,b) addressed two of the criticisms West & Gauldie (1994) made of the radiometric ageing method, i.e., that estimated ages depend on the otolith mass growth model, and that the accretion rates of $^{210}\text{Pb}$ and $^{226}\text{Ra}$ may change over the life of the fish. He showed that, for the problem of estimating the longevity of orange roughy, these do not appear to pose serious difficulties. This work also supported the general conclusions of the earlier analyses that orange roughy attain ages at least approaching, and very possibly exceeding, 100 years.

Whitehead & Ditchburn (1996) serially dissolved orange roughy otoliths (dissolving c. 15% of otolith weight at each step) and analysed the chemical composition of the consequent dissolved layers. Based on the assumption that the outermost otolith layers contain only young material, they claimed that $^{210}\text{Pb}$ is incorporated in the outer (i.e., young) layers of the otoliths, and that there was a variable uptake of $^{226}\text{Ra}$ into otoliths over the life of the fish, thus questioning assumptions of the radiometric method. However, as is apparent when viewing a cross-section of an orange roughy otolith (see Fig. 3C), the first, and subsequent, dissolutions would comprise both young and old material, as new material appears to be deposited primarily on the proximal side of the otolith. In fact, the first dissolution is likely to contain a large proportion of the oldest material deposited on the distal face of the otolith, and hence, contain the highest level of $^{210}\text{Pb}$ (see fig. 3 of Whitehead & Ditchburn 1996). Thus, the fundamental assumption of Whitehead & Ditchburn (1996) is invalid.

The investigations based on radiometric analyses of otoliths, although not providing any further formal validation, provide support for the argument of Mace et al. (1990) that orange roughy are very slow-growing and long-lived. The comparison of age estimates based on radiometric data and counts of assumed annual zones (Smith et al. 1995) also supports the hypothesis that opaque and translucent zones are laid down annually in otoliths of orange roughy throughout their lifetime. However, although some of the criticisms of West & Gauldie (1994) have been answered, the possibility that $^{222}\text{Rn}$ could diffuse into or out of the otolith (and the general problem of mobility of elements within an otolith) has not been resolved. Such a diffusion could result in higher or lower estimates of age.

**ANALYSES OF OTOLITH DIMENSIONS**

There was no significant difference between the weights of left and right orange roughy otoliths (Francis & Tracey 1994; Smith et al. 1995).

Francis & Tracey (1994) addressed the question of how much, if at all, had the average orange roughy otolith weight changed on the Chatham Rise over the period 1984-92. This work was an indirect way of measuring a change in average age, as otolith weight continues to increase as a fish ages. A decrease in mean otolith weight would support the hypothesis that biomass had declined or recent recruitment had been high, whereas no decrease would imply either that biomass has not declined or that recent recruitment has been very low. It was found that there had been no significant change in average otolith weight over the time period examined and, given that other research had shown that the Chatham Rise orange roughy biomass had progressively declined, this result supported the hypothesis that recruitment had been low in recent years. There had also been a decline in the proportion of smaller otoliths over time, which was consistent with the low recruitment hypothesis.

At the time this work was planned it was assumed that mean otolith weight (being a proxy for mean age) would be a more sensitive indicator of biomass decline than mean fish length. However a study by Francis & Smith (1995) showed this to be not true. They noted that although the biomass of orange roughy on the Chatham had declined by an estimated
80%, the mean length of fish in the population had not declined. They suggested that because orange roughy are slow growing, mean age or mean otolith weight would likely be more sensitive indicators of biomass decline than mean fish length. A simulation study compared the three indicators and found them to be equally sensitive, assuming constant recruitment (although this conclusion is unlikely to change with any changes in recruitment patterns).

Smith et al. (1995) presented relationships between otolith weight and estimated age and suggested that the best fit was a broken stick linear equation, with the break being at about the age and size at first maturity (i.e., 25 years and 30 cm). Such a relationship implied that pre-mature fish had an otolith mass growth rate of c. 5.3 mg yr\(^{-1}\), but it reduced to c. 3.3 mg yr\(^{-1}\) after maturity.

Relationships between standard length of orange roughy and otolith length and weight are presented in Table 3. Kotyar (1981) stated that the relationship between fish and otolith length was linear, but raw data were not presented. No significant sexual or areal differences were apparent in relationships between total fish length and otolith weight or length (Linkowski & Liwoch 1986). Smith & Robertson (1992) plotted raw data for both these relationships for fish of length 13–43 cm, but provided no regression equations. They did claim that fish length was linearly related to otolith length, but the plotted line on their Fig. 2A clearly has unbalanced residuals.

Table 3. Relationship for orange roughy (Hoplostethus atlanticus) between fish length and otolith length and weight. (SL, standard fish length (cm); OL, otolith length (mm); W, otolith weight (g); Lgth, length range of fish in sample (cm); \(R^2\), regression coefficient.) Linkowski & Liwoch (1986) measured total fish length (TL), so their equations were adjusted to SL using the following relationships (NIWA unpubl. data): SL = 0.839 TL – 1.53 (Chatham Rise), SL = 0.828 TL – 1.35 (Challenger Plateau).

<table>
<thead>
<tr>
<th>Equation</th>
<th>Area</th>
<th>(R^2)</th>
<th>n</th>
<th>Length</th>
<th>Source</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Otolith length–fish length</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>OL = 0.418 SL – 0.758</td>
<td>Chatham</td>
<td>0.99</td>
<td>138</td>
<td>16–44</td>
<td>Linkowski &amp; Liwoch (1986)</td>
</tr>
<tr>
<td>OL = 0.420 SL – 0.644</td>
<td>Challenger</td>
<td>0.98</td>
<td>235</td>
<td>10–40</td>
<td>Linkowski &amp; Liwoch (1986)</td>
</tr>
<tr>
<td>OL = 0.38 SL*</td>
<td>Chatham</td>
<td>–</td>
<td>110</td>
<td>3–34</td>
<td>Gauldie (1988b)</td>
</tr>
<tr>
<td>OL = 0.436 SL – 0.831</td>
<td>Chatham</td>
<td>0.86</td>
<td>438</td>
<td>12–41</td>
<td>Kalish (NIWA unpubl. data)</td>
</tr>
<tr>
<td><strong>Otolith weight–fish length</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>W = 1.30 \times 10^{-6} (1.207 SL + 1.625)(^{3.146})</td>
<td>Challenger</td>
<td>0.99</td>
<td>683</td>
<td>10–40</td>
<td>Linkowski &amp; Liwoch (1986)</td>
</tr>
<tr>
<td>W = 2.71 \times 10^{-5} SL(^{2.38})</td>
<td>Chatham</td>
<td>0.83</td>
<td>94</td>
<td>14–33</td>
<td>Gauldie (1988b)</td>
</tr>
<tr>
<td>W = 4.87 \times 10^{-7} SL(^{3.62})</td>
<td>Chatham</td>
<td>0.60</td>
<td>7152</td>
<td>25–45</td>
<td>Francis (NIWA unpubl. data)</td>
</tr>
</tbody>
</table>

*Equation estimated from plotted data.

USE OF AGEING DATA IN STOCK ASSESSMENTS

As an understanding of the growth of orange roughy developed and ageing techniques improved, biological parameters used for stock assessment modelling were revised (Table 4). As ageing focused primarily on Chatham Rise fish, life history parameters from this work were used initially in assessments for all populations of orange roughy in New Zealand waters, although in more recent assessments some population-specific parameters have been estimated (Annala et al. 1998).

Initial assessments either selected parameters based on an understanding of other marine fish, or used those proposed by Gauldie (1987) and van den Broek (1983). It was emphasised that the assessments were highly uncertain owing to the ageing techniques being unvalidated. Trawl surveys indicated that there had been rapid declines in biomass, suggesting that \(M\) was underestimated, and age at recruitment was incorrect (Robertson et al. 1988). The parameters changed markedly in 1988–89, following the work of Mace et al. (1990) showing slow growth and low productivity. Subsequently, only minor changes were made until 1993–94 when Francis et al. (1993) revised the von Bertalanffy...
parameters, by estimating $L_w$ as the asymptote of the curve describing mean length as a function of otolith weight.

In 1993–94, new data based on zone counts in otoliths of adult fish provided age at maturity and recruitment estimates, and a new value of $L_w$ (Doonan 1994). In 1996–97 and 1997–98, re-examinations of pre-transition zone growth resulted in revisions of the age at maturity for various fishing grounds (Francis & Horn 1997; Horn et al. 1998).

The choice of productivity parameters for any fish can have a major effect on assessments of biomass and yields. Initial assessments of orange roughy assumed that the species had relatively low productivity. However, parameters have been modified over time in ways that represent even lower productivity, i.e., a reduction in $M$ and the von Bertalanffy $k$, and increases in age at maturity and recruitment (Table 4). The continued uncertainty about maximum age of orange roughy impacts on estimates of $M$ and $k$ (parameters strongly correlated with maximum age).

**CONCLUSION**

This is the first detailed review of techniques used to age orange roughy, an important deepwater commercial species. It highlights the difficulty of determining age of a long-lived species, and stresses the importance of obtaining accurate productivity parameters for stock assessment. Age at maturity, and growth and mortality rates are directly related to the sustainable yield of a fishery, and early (probably incorrect) assumptions of life history parameters had a considerable effect on catch quotas, and subsequent stock size, when the New Zealand orange roughy fishery was developing.

Most investigations of age and productivity of orange roughy have used otoliths, though studies using scales (Gauldie et al. 1991) and parasite loadings (Gibson & Jones 1993) have also been reported. The otolith investigations have concentrated on three areas: interpretation of daily growth increments, interpretation of annual growth increments, and radiometric analyses. All three areas have been fraught with problems relating to the validation of the ageing technique.

The existence of daily growth increments has been advocated, but never validated, in a series of works by Gauldie. Gauldie et al. (1989) claimed that the formation of daily micro-increments is obligatory in fish otoliths, yet it has been shown for some species that they may not be formed on a daily basis, especially in older fish (e.g., Radtke et al. 1985; Morales-Nin 1987). The predictive model of daily increment growth proposed by Romanek & Gauldie (1996) is based on few data from species living in environments very different to those experienced by orange roughy, and also assumes that the rate and conditions of biogenic crystal growth is similar to

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**Table 4** Biological parameters used for Chatham Rise orange roughy (*Hoplostethus atlanticus*) assessments, 1984–98. (Sex: $B$, both; $M$, male; $F$, female. $M$, instantaneous natural mortality; $A_r$, age at recruitment; $A_m$, age at maturity; $S_r$, gradual recruitment; $S_m$, gradual maturity; $L_w$, $k$, $t_0$, von Bertalanffy growth parameters; $-$, not estimated.)

<table>
<thead>
<tr>
<th>Assessment years</th>
<th>Sex</th>
<th>$M$</th>
<th>$A_r$</th>
<th>$A_m$</th>
<th>$S_r$</th>
<th>$S_m$</th>
<th>$L_w$</th>
<th>$k$</th>
<th>$t_0$</th>
<th>Source</th>
</tr>
</thead>
<tbody>
<tr>
<td>Up to 1984–85</td>
<td>$B$</td>
<td>0.1</td>
<td>$-$</td>
<td>$-$</td>
<td>$-$</td>
<td>$-$</td>
<td>$-$</td>
<td>$-$</td>
<td>$-$</td>
<td>Robertson (1985)</td>
</tr>
<tr>
<td>1985–86</td>
<td>$B$</td>
<td>0.1</td>
<td>5</td>
<td>$-$</td>
<td>$-$</td>
<td>$-$</td>
<td>0.2</td>
<td>$-$</td>
<td>$-$</td>
<td>Robertson (1986)</td>
</tr>
<tr>
<td>1986–87</td>
<td>$B$</td>
<td>0.1</td>
<td>5</td>
<td>53.5</td>
<td>$-$</td>
<td>0.22</td>
<td>2.79</td>
<td>$-$</td>
<td>$-$</td>
<td>Robertson et al. (1988)</td>
</tr>
<tr>
<td>1987–88</td>
<td>$B$</td>
<td>0.1</td>
<td>6</td>
<td>$-$</td>
<td>$-$</td>
<td>41.2</td>
<td>0.65</td>
<td>$-$</td>
<td>0.065</td>
<td>Mace &amp; Doonan (1988)</td>
</tr>
<tr>
<td>1988–89</td>
<td>$B$</td>
<td>0.05</td>
<td>20</td>
<td>$-$</td>
<td>$-$</td>
<td>42.5</td>
<td>0.059</td>
<td>$-$</td>
<td>$-$</td>
<td>Robertson (1989)</td>
</tr>
<tr>
<td>1989–90–1991–92</td>
<td>$B$</td>
<td>0.05</td>
<td>23</td>
<td>3</td>
<td>42.5</td>
<td>0.059</td>
<td>$-$</td>
<td>$-$</td>
<td>$-$</td>
<td>Robertson (1990, 1991)</td>
</tr>
<tr>
<td>1992–93</td>
<td>$B$</td>
<td>0.04</td>
<td>23</td>
<td>3</td>
<td>42.5</td>
<td>0.059</td>
<td>$-$</td>
<td>$-$</td>
<td>$-$</td>
<td>Francis &amp; Robertson (1992)</td>
</tr>
<tr>
<td>1993–94</td>
<td>$B$</td>
<td>0.04</td>
<td>22</td>
<td>6.6</td>
<td>37.3</td>
<td>0.07</td>
<td>$-$</td>
<td>$-$</td>
<td>$-$</td>
<td>Francis et al. (1993) Doonan (1993)</td>
</tr>
<tr>
<td>1994–95–1995–96</td>
<td>$M$</td>
<td>0.045</td>
<td>33</td>
<td>9</td>
<td>36.4</td>
<td>0.070</td>
<td>$-$</td>
<td>$-$</td>
<td>$-$</td>
<td>Doonan (1994)</td>
</tr>
<tr>
<td></td>
<td>$F$</td>
<td>0.045</td>
<td>34</td>
<td>8</td>
<td>38.0</td>
<td>0.061</td>
<td>$-$</td>
<td>$-$</td>
<td>$-$</td>
<td>Francis et al. (1995)</td>
</tr>
<tr>
<td>1996–97</td>
<td>$M$</td>
<td>0.045</td>
<td>30</td>
<td>4</td>
<td>36.4</td>
<td>0.070</td>
<td>$-$</td>
<td>$-$</td>
<td>$-$</td>
<td>Francis &amp; Horn (1997)</td>
</tr>
<tr>
<td></td>
<td>$F$</td>
<td>0.045</td>
<td>30</td>
<td>4</td>
<td>38.0</td>
<td>0.061</td>
<td>$-$</td>
<td>$-$</td>
<td>$-$</td>
<td>Francis &amp; Horn (1997)</td>
</tr>
<tr>
<td>1997–98</td>
<td>$M$</td>
<td>0.045</td>
<td>29</td>
<td>3</td>
<td>36.4</td>
<td>0.070</td>
<td>$-$</td>
<td>$-$</td>
<td>$-$</td>
<td>Doonan et al. (1998)</td>
</tr>
<tr>
<td></td>
<td>$F$</td>
<td>0.045</td>
<td>29</td>
<td>3</td>
<td>38.0</td>
<td>0.061</td>
<td>$-$</td>
<td>$-$</td>
<td>$-$</td>
<td>Horn et al. (1998)</td>
</tr>
</tbody>
</table>
that of pure chemical crystals. Recent work refutes
this assumption (Dove 1998) and has also shown that
the concentration of chemical elements and pH in the
endolymph surrounding the otolith are not homoge­
nous (Payan et al. 1998).

Several workers attempted to interpret the
patterns of opaque and translucent zones visible in
whole and sectioned orange roughy otoliths, and
relate them to annual growth episodes. These studies
were largely unsuccessful as their authors expected
to find the "classical" zonation pattern of zone width
uniformly decreasing towards the otolith edge
(Kotlyar 1981; van den Broek 1983; Linkowski &
Liwoch 1986). Mace et al. (1990) were the first to
obtain very small fish and relate the number of
translucent zones visible in whole otoliths to the
progression of peaks in length frequency distri­
butions. This work constitutes the only published
validation of orange roughy ageing, but it only
confirms the deposition of annual zones up to age
3. It also demonstrated the very slow initial growth
of the juveniles. This work has been enhanced by
Doonan (unpubl. data) who extended the validation
to the first four translucent zones.

Attempts to validate the growth of older fish by
following the progression of strong or weak year
classes in samples from consecutive years, as
determined from counts of presumed annual zones,
were unsuccessful owing to reader variability
(Tracey et al. in: Clark 1996). The interpretation of
the zonation pattern is often complicated because of
varying zone width and the occurrence of multiple
banding structure in some individual zones (see
Appendix 1). However, these investigations have
identified a point of transition in otoliths where zone
width changes markedly from broad to narrow, and
is associated with the onset of sexual maturity.

Radiometric techniques have been used to age
numerous relatively long-lived fish species. How­
ever, some of the assumptions of the technique
have been criticised (West & Gauldie 1994), primar­
ily with regard to possible mobility of elements
within an otolith and changes in elemental accretion
rates over time. The most recent radiometric
evaluation of orange roughy otoliths (Francis 1995b)
addressed the second of these criticisms and still
concluded that the fish could live more than 100
years. However, the problem of elemental mobility
(particularly concerning the diffusion of radon gas
into or out of the otolith) has not been resolved and
could result in higher or lower estimates of age using
this technique. Proctor & Thresher (1998) showed
that different methods of post-mortem handling of
fish can influence the measurements of certain trace
elements in otoliths.

Clearly, future research needs to address age
validation of post-juvenile orange roughy. Vali­
dating otolith zones by examining changes in otolith
margins over a year, or the progression of strong or
weak year classes over time, does not appear to be
viable. Radiometric techniques hold promise, but
only if the problem of elemental mobility is first
addressed. The validation of the periodicity of zones
in otoliths using chemical markers is another
technique being investigated through attempts to
capture and hold deepwater fish species in situ, and
tag them with chemical markers (de Pontual et al.
1998). Although the extent and composition of any
otolith structure laid down outside the chemical mark
would provide information on growth, this type of
validation trial is difficult because of the necessity
to hold marked fish in their normal environment for
a period of time.

Improved tools and more efficient techniques are
being developed to measure the elemental compo­
sition at sites within the otolith. Cycles in the ratio
of strontium to calcium along otolith radii have been
linked to growth in some species (e.g., Sadovy &
Severin 1992), so similar investigations of orange
roughy otoliths may indicate whether any visual
zonation pattern is correlated with cycles of chemical
composition. Age validation studies have been
provided for some species from bomb radiocarbon
methods (e.g., Kalish 1995). The viability of using
radiocarbon from nuclear testing as a dated marker
in the otoliths of deepwater species is currently being
examined, and may provide definitive age validation.

ACKNOWLEDGMENTS
Thanks to Peter Marriott for contributing to the section
on otolith preparation techniques, Darren Stevens for the
otolith drawings, Ian Doonan for Fig. 6, and to Malcolm
Clark, Larry Paul, Rosie Hurst, and an anonymous ref­
eree for commenting on the manuscript. Recent ageing
work developed from earlier proposals by John Kalish,
and we acknowledge his studies on orange roughy. This
project (DEOR11) was funded by the New Zealand Min­
istry of Fisheries.

REFERENCES
Annala, J. H. 1993: Deepwater projects review 1992:
MAF Fisheries Greta Point Internal Report No.
200. Unpublished report held in NIWA library,
P. O. Box 14 901, Wellington, New Zealand. 57 p.


Tracey & Horn—Background and review of ageing orange roughy


Gauldie, R. W. 1988c: Microscopic growth increments in the otoliths of orange roughy, Hoplostethus atlanticus. (Variations of this title have been cited as New Zealand Fisheries Research Bulletin No. 28 or 29, 1989, or in press. Current status is unpublished manuscript.)


Appendix 1  Otolith collection, preparation, and reading.

Otolith collection and storage
Orange roughy otoliths have been collected in New Zealand since the early 1970s. Otoliths were removed from the fish, cleaned of any adherant organic material, and either stored dry in paper envelopes (1979–83, and from 1992 onwards), or wet in a solution of 70% isopropyl alcohol with added glycerine (5% by volume) (1984–91). Otoliths have been obtained from three sources: trawl surveys of various areas (since 1979), samples from fish processing sheds (1988–91), and scientific observer samples taken at sea on commercial vessels (since 1986).

Reading whole otoliths
Otoliths from small fish can be read whole with reasonable confidence. Otoliths were cleaned of any remaining organic material, placed distal face up in black plastic trays filled with paraffin oil, and examined with a binocular microscope using reflected light. Smith et al. (1995) used a similar method except that the paraffin oil was replaced by isopropyl alcohol with added glycerine (5% by volume) (1984–91). Otoliths were cleaned of any adherant organic material, placed distal face up in black plastic trays filled with paraffin oil, and examined with a binocular microscope using reflected light. Smith et al. (1995) used a similar method except that the paraffin oil was replaced by isopropyl alcohol with added glycerine (5% by volume) (1984–91). Otoliths have been obtained from three sources: trawl surveys of various areas (since 1979), samples from fish processing sheds (1988–91), and scientific observer samples taken at sea on commercial vessels (since 1986).

Sectioning (New Zealand method)
One otolith from each pair from an individual fish was sectioned longitudinally. The left otolith was consistently chosen for sectioning, but if it was chipped, broken or calcified then the right otolith was used. If both were damaged or calcified, the fish was rejected from the sample.

The otolith was examined distally (sulcus side down) under a binocular microscope on low magnification, and a straight line was drawn on the otolith through the intended sectioning plane with a fine (0.35 mm) ink cartridge pen. The optimal sectioning plane was from the primordium through the most uniform postero-dorsal axis (Fig. 3B). This was generally orientated close to the dorsal edge of the postrostrum. The otoliths were individually embedded in slow curing epoxy resin (Araldite K142) in disposable vinyl specimen moulds. Curing was accelerated by placing the moulds in an oven for 4 h at 50°C. The moulds were left overnight to finish curing, then the embedded otoliths were removed from the vinyl moulds and labelled.

The embedded otoliths were sectioned on a Struers Accutom-2 sectioning saw, with twin 0.37 mm diamond edged blades separated by a 1 mm thick spacer. Each embedded otolith was aligned so that one blade passed each side of the line, and the mould was clamped in the chuck. Moulds were sectioned at 1800 rpm, using a 3% oil/water coolant.

The cut sections were hand polished on one side using a graded series of wet-and-dry carborundum papers (400, 1200, and 4000 grit) and a suspension of 0.1 μm alumina powder (Linde A). The sections were then mounted, polished face down, on glass slides using quick curing epoxy resin ("Five minute Araldite"). The exposed side of the mounted section was polished on a Struers Planopol-2 polishing wheel, using a graded series of carborundum papers and polishing powder as above. The sections were ground to a thickness of c. 0.3 mm at the primordium and 0.4 mm at the terminal edge. The fine band structure at the terminal edge became harder to
resolve if the section was much less than 0.4 mm thick. But at that thickness, the more opaque centre of the otolith was harder to read, so this portion of the otolith was ground slightly thinner. The prepared section was then ready for reading.

**Sectioning (Australian method)**

At the Australian Central Ageing Facility, whole otoliths were embedded individually in clear polyester resin and cut, longitudinally, along the anterior-posterior axis using a 0.15 mm thick diamond edged circular saw (Smith et al. 1995). Two or three sections, each c. 0.3 mm thick, were taken from each otolith, and mounted on glass slides with a clear mountant under a coverslip. Taking multiple sections ensured that at least one was through the primordium.

**Reading otolith sections**

Mounted otolith sections were examined under a binocular microscope with illumination by transmitted light and at ×36 magnification. Varying the angle of a rotating polarising filter sometimes helped to clarify the zonation pattern. The area near the primordium can be quite dense and dark, so zones may be difficult to differentiate. The long axis diameter of the first three zones on whole otoliths is c. 3.3 mm (Mace et al. 1990), so on sections, the expected distance from the primordium to the outer edge of the third zone is c. 1.7 mm. An eyepiece graticule was used to measure out that distance, and thus, indicate the likely position of the third zone. The first three zones did tend to be wider than all the others. Subsequent zones were generally more easy to interpret. Otolith readers had no knowledge of fish length at any reading, nor of their first readings when making duplicate counts.

A count of zones was made from the primordium to the transition zone at ×36 magnification. In the longitudinal section of most otoliths, this point was clear and unambiguous, and generally occurred where the otolith surface begins curving (Fig. 3C). In c. 10% of otoliths, band widths decreased steadily rather than suddenly, and the transition zone was less obvious. In this situation, the transition zone was assigned to the point where the surface began curving (Fig. 3C). The pre-transition zones were often irregularly spaced and seldom showed a regular decreasing width as distance from the primordium increased.

Zone counts from the transition zone to the section margin were made at ×64 magnification, as the zonation structure is much finer than in the pre-transition zone region, particularly at the margins of otoliths from large fish (Fig. 3C).

Some otolith sections were difficult to interpret because of complicated zone structure and multiple banding just outside the transition zone. It was sometimes difficult to determine whether individual bands should be counted or whether some bands should be grouped to represent 1 year’s growth. For interpretation problems in the post-transition area, subtle changes in the colour of the section would sometimes mark the zone boundaries and, hence, indicate whether multiple banding was present within a zone.


Mace, P. M.; Doonan I. J. 1988: Biomass and yield estimates for North Chatham Rise orange roughy. 42 p. (Draft report prepared for the Standing Committee on orange roughy.) Unpublished report held in NIWA library, P. O. Box 14 901, Wellington, New Zealand.


