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# Deep Sea – Close Kin: A Genetic Approach for Improved Fisheries Management

Close Kin Analysis based on Genetics in Support of Improved Management under the Common Fisheries Policy – A Feasibility Assessment

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### Foreword

Being terrestrial animals, we human beings tend to forget that more than 90% of earth's liveable volume is inherent to the deep ocean, below water depths of 200m. The deep sea is a vast realm that is increasingly commercially exploited, while remaining to the greatest extent a "big unknown" and as of yet unexplored space.

Fish species living below 400m are an important natural renewable resource that is exploited by a rather recent fishery that has swiftly expanded in response to a growing demand for fish products and the decline of stocks of marine fish species that live in more accessible layers of the ocean.

We know little about the biology of deep sea fish, and much less still about the deepocean ecosystems they are part of. It is clear though that deep-sea ecosystems are fragile, and that deep sea fish stocks are very sensitive to the impact caused by fisheries.

The need to better understand the responses of deep sea fish stocks to exploitation Is reinforced by the increasing evidence of climate change effects. While climate change impacts in the deep ocean are less visible than those above sea level or in shallow waters, the longevity and slow pace of life in the deep makes that ecosystem uniquely sensitive to environmental variability, and climate change impacts might further accentuate changes in abundance and distribution of deep sea fish species.

A major step towards an improved deep-sea fisheries management would be the establishment of a fishery-independent tool to assess the abundance and the monitoring of changes in abundance of commercially exploited species. This report assesses whether a recent close-kin abundance estimate based on genetics, which was originally developed by CSIRO Australia for Southern bluefin tuna, might provide an abundance estimate tool also for deep-sea fish species, which could ultimately underpin stock assessments and fishery management frameworks.

### Abstract

Deep-sea fish stocks consist of species that live at depths of greater than 400 metres. While being important for EU fisheries, this natural renewable resource is particularly vulnerable to over-fishing, as many deep sea species are slow-growing and commonly of low fecundity. Generally little is known about the biology of deep sea species, and there prevails a substantial lack of scientific data on deep-sea stocks. This constitutes a major impediment to management strategies underpinning sustainable and profitable deep sea fisheries.

Europe's deep-sea fisheries began in the 1970's and were entirely unregulated. The fleet grew as rewards were high, but many species were rapidly depleted. It was only in 2003 that a management plan was brought into action. While some measures to better protect commercially exploited deep sea fish have been adopted, such as the limitation of fishing effort or total allowable catches, these have been insufficient to allow stocks to recover and there is a general consensus that most deep-water stocks remain below safe biological limits for exploitation.

In a recent communication to the Council and the European Parliament, the European Commission has emphasized the need to improve our knowledge on deep sea fish species to move away from the current prevailing unsustainable exploitation.

Ideally, this would be the development of a robust and practical approach to estimate the abundance of deep sea species to support stock assessments and reduce the uncertainty about the state and rebuilding rates of commercially exploited deep sea stocks.

The current rapid technology development and concurrent steep drop in costs of largescale genotyping offers major opportunities for fisheries management. This report explores whether the concept of genetic close-kin abundance estimation, recently applied to establish biomass estimates of Southern Bluefin Tuna, can be applied to fisheries management of deep sea fish species.

### 1. Introduction

Deep-sea fish stocks consist of species that live at depths of greater than 400 metres. At these depths, extreme but stable conditions have led to the development of marine ecosystems that are very susceptible to disturbances.

Since deep-sea fish tend to live long and grow slowly, while maturing late and having low fecundity, this natural renewable resource is particularly vulnerable to over-fishing.

Fisheries targeting deep-water species have developed relatively recently during the 1960s. Deep-sea fisheries are mostly considered "data-poor", *i.e.* there prevails a lack of reliable scientific data to ensure sustainable exploitation. Sustainable management is further impeded as generally knowledge about the biology of deep sea fish is limited.

To preserve this resource, both input and output measures, such fishing effort limitations or total allowable catches have been adopted. However, they are insufficient, as most deep-water stocks are considered to be below safe biological limits for exploitation.

The specific characteristics of deep-sea fisheries including complex ecosystems, their great depths and distances from the coast, and complex governance frameworks pose several management challenges.

Acknowledging the particular challenges inherent to deep sea fishery management, in 2009 the FAO has developed international guidelines for the management of deep-sea fisheries in the high seas. These guidelines are a voluntary international instrument intended to support States and Regional Fisheries Management Organizations (RFMOs) in formulating and implementing appropriate measures for the sustainable management of deep-sea fisheries in the high seas.

In the European Union (EU) deep-sea fisheries are covered by Council regulation No. 2347/2002 on "establishing specific access requirements and associated conditions applicable to fishing for deep sea stocks", which lists 24 species, including a number of deepwater sharks. For the EU, the setting of total allowable catches (TACs) for deep sea fish is based on the scientific advice developed established the International Council for the Exploration of the Sea (ICES) and the Scientific, Technical and Economic Committee for Fisheries (STECF). According to both advisory bodies, the majority of deep-sea stocks are subject to unsustainable exploitation. Therefore, the trend is to reduce fishing opportunities and currently no directed fishery is authorised for several species, including all deep-sea sharks.

In at least one case in EU waters, an area was closed for the exploitation of deep sea species. Council Regulations (EC) No 2270/2004 and 1359/2008 set up and maintain closed areas for the protection of vulnerable aggregations of orange roughy (*Hoplostethus atlanticus*). ICES advises for 2015 that, due to the very low stock size, this fishery should remain closed.

Owing to the lack of data mentioned above, quantitative population modelling is not feasible for all exploited deep sea stocks. Therefore alternative approaches are required to assess the status and/or the risk exposure of populations that cannot be assessed by established fisheries science approaches.

This report discusses and evaluates the possibility to transfer genetic close-kin abundance estimation, recently applied to establish biomass estimates of Southern Bluefin Tuna, into fisheries management schemes for marine deep sea fish species. The report starts out by looking into the current status of deep sea fisheries management under the Common Fisheries Policy, with emphasis of knowledge on stock status, knowledge gaps, needs and challenges. This is followed by a discussion of the principles of close kin analysis and its possible application for deep sea fisheries. Steps needed to foster an uptake and technology transfer into existing management schemes are outlined. The observations of this report will also be submitted to the 2016 ICES Expert Working Group on Applied Genetics for Fisheries and Mariculture (WGAGFM) for further elaboration.

# 2. The Management of Commercial deep Sea Species under the Common Fisheries Policy

In recent times deep sea fish species gained increasing relevance for commercial exploitation, also for the EU fishing industry, which is also reflected in EU legislation. This chapter depicts relevant EU legislation, and discussed knowledge as well as knowledge gaps of deep sea fish biology and their management.

#### 2.1 The EU Legal framework

In the European Union (EU) deep-sea fisheries are covered by Council Regulation No. 2347/2002 on "establishing specific access requirements and associated conditions applicable to fishing for deep sea stocks" (The Council of the European Union, 2002). This regulation emphasizes that scientific advice concerning certain stocks of fish found in the deep sea indicates that these stocks are vulnerable to exploitation, and that fishing opportunities for these stocks should be limited or reduced in order to assure their sustainability. Moreover it stresses that appropriate, verifiable and up-to-date information for scientific advice about fisheries and the marine environment should be made available to the relevant scientific and management bodies as soon as possible. Annex I of this regulation lists 24 species, including a number of deepwater sharks. It applies to Community fishing vessels carrying out fishing activities in ICES sub-areas I to XIV inclusive, and community waters of CECAF areas 34.1.1, 34.1.2, 34.1.3 and 34.2 (Figures 1 and 2).

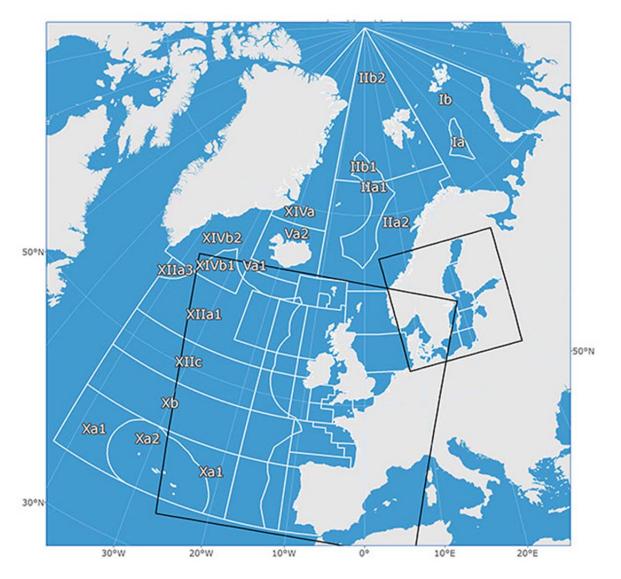


Figure 1 Boundaries of the Atlantic, Northeast (FAO Major Fishing Area 27) corresponding to the ICES fishing areas for statistical purposes. © FAO 1990-2015. FAO Major Fishing Areas. ATLANTIC, NORTHEAST (Major Fishing Area 27). CWP Data Collection. In: FAO Fisheries and Aquaculture Department [online]. Rome. Updated 11 December 2008. (<u>http://www.fao.org/fishery/area/Area27/en#NB10D3</u>)

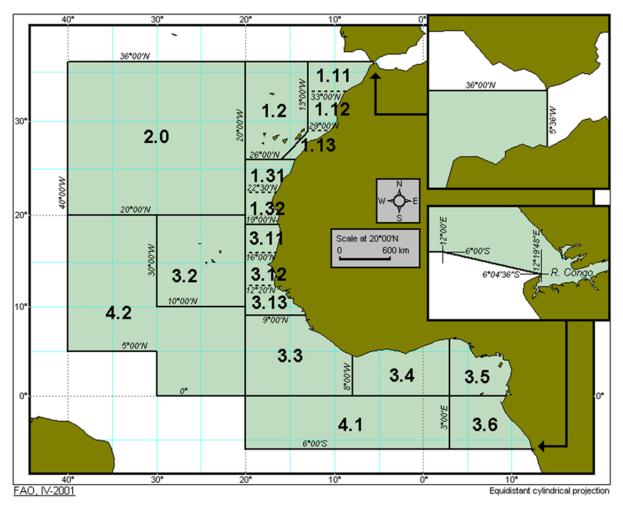


Figure 2: Area covered and Members States of CECAF - Fishery Committee for the Eastern Central Atlantic. ATLANTIC, EASTERN CENTRAL (FAO Major Fishing Area 34). © FAO 1990-2015. FAO Major Fishing Areas. ATLANTIC, EASTERN CENTRAL (Major Fishing Area 34). CWP Data Collection. In: FAO Fisheries and Aquaculture Department [online]. Rome. Updated 1 October 2004. (http://www.fao.org/fishery/area/Area34/en)

In 2007 the European Commission issued a communication to the Council and the European Parliament entitled: Review of the management of deep-sea fish stocks (European Commission, 2007). This communication assesses the status of deep sea fisheries management measures and efficiency. It highlights the point that, measures to protect deep sea fish species have been adopted, such as the limitation of fishing effort or total allowable catches but that these they are insufficient, as most deep-water species are still fished above sustainable levels. Regulations concerning deep-sea fisheries are relatively recent. TACs were first introduced in 2002 for the period 2003-2004 and are updated every two years for the majority of species. Due to the lack of knowledge regarding the species concerned and the lack of information on what makes up the catches, discards, geographical distribution of the stocks, etc., the TACs were initially set rather arbitrarily and for only nine of the 48 deep-sea species which are listed in Annexes I and II to Regulation (EC) No 2347/2002 ((The Council of the European Union, 2002); -see Annex). After 2004 TACs have been set according to scientific advice provided by ICES and STECF. Both advisory bodies indicate that the majority of deep-sea stocks are subject to unsustainable exploitation. As a consequence, the trend is to reduce fishing opportunities and currently no directed fishery is authorised for several species, including all deep-sea sharks.

As a complementary measure to TACs, fishing effort limitations were introduced reducing the fishing effort of vessels with licences by 10 % in 2005 and a further 10 % in 2006, as compared to 2003 levels. The communication observes however that this capacity ceiling failed to limit the expansion of deep-sea fisheries, since certain deep-sea stocks, such as ling, tusk and argentines, are taken as by-catches. Generally the reductions in fishing effort proved to be ineffective as they have not led in practice to a reduction of the exploitation rate of deep-water stocks. Better information is needed on the various fisheries exploiting deep-sea species so that fishing effort levels can be adjusted in each of them individually according to the target species and by-catch species.

The communication states that scientific sampling programmes were carried out aiming at enhancing the insufficient scientific knowledge of deep-water stocks. Generally though, it concludes that current legislation does not provide sufficient guidance on how to proceed. Sampling schemes drawn up by the Member States differ in quality and content, rendering their exploitation for assessment purposes difficult. The recommendation is to design a reporting format facilitating the aggregation of the data received and to improve their quality.

Deep sea fisheries management is also accompanied by monitoring and control measures. An example are closed areas such as those introduced for orange roughy (The Council of the European Union, 2004, 2008). Vessels with deep-sea fishing licences entering such areas must observe certain rules: During transit in the area in question, they must maintain an average speed of at least eight knots and all gears carried on board must be lashed and stowed.

The communication recommends that the supervisory authorities of the Member States should make more use of the satellite-based vessel monitoring system (VMS). This would allow to warn inspectors of suspect activities in the areas concerned and to intercept the vessels on entering port. Moreover fisheries monitoring centres should be set up in each Member State to inspect the vessels in transit or fishing in the closed areas. Control efficiency is however limited as a great number of fishing vessels hold a deep-sea fishing licence while their catches of deep-sea species are only marginal. That limits the effectiveness of deep-sea effort limitations and can lead to control problems for non-deep-sea stocks. Vessels with such licences can legitimately fish in areas where a Member State has deep-sea quotas, without necessarily targeting this type of stocks.

Generally Member States are required to notify the Commission of the inspection and surveillance procedures they apply in the ports designated for landings of deep-sea species.

The latest EU legal act concerning deep sea fisheries is Council Regulation (EU) No 1367/2014 fixing for 2015 and 2016 the fishing opportunities for Union fishing vessels for certain deep-sea fish stocks (The Council of the European Union, 2014). It states in the preamble under (6) that the latest scientific advice from ICES and STECF indicates that most deep-sea stocks are still harvested unsustainably and that fishing opportunities for those stocks, in order to assure their sustainability, should be further reduced until the evolution of the stocks shows a positive trend. ICES has further advised that no directed fishery should be allowed for orange roughy in all areas and for certain stocks of red seabream and roundnose grenadier.

#### 2.1.1 Current knowledge about deep sea fish biology

The deep-sea fishes of the north-east Atlantic have a long history of study over the last century (Gordon, 2003). Despite this, new species are still being described and for many species there exists only basic estimates of life-history parameters, stock structure, movements and distribution. The fish assemblage is very diverse (well over 200 species are found between depths of 400-2000m) and diversity increases with depth to around 1200-1400m (Campbell et al., 2011; Clarke et al., 2015). There are of course far fewer species that are of commercial interest (Annexe 1). The degree of biological knowledge

and information for these species varies from species to species and consequently so does the capacity to manage effectively. There has also been mixed stock responses to exploitation; many deepwater species declined with the onset of fishing (Bailey et al., 2009), some dramatically such as orange roughy, blue ling and 'siki' sharks, others less obviously, such as black scabbardfish and roundnose grenadier.

Perhaps the most basic requirement of fisheries science is the data needed to define the distribution of the stock in space and depth. For some species there is reasonable data for this, for example, the round-nose grenadier (*Coryphaneoides rupestris*) is widely distributed across the north Atlantic at depths between 500-2000 m. Orange roughy (*Hoplostethus atlanticus*) is much more patchily distributed throughout the northeast Atlantic at depths below 1000m but is only found on seamounts, the mid-Atlantic ridge and steep-sided sections of the continental slope. The black scabbard-fish (*Aphanopus carbo*) is known to spawn in the south around Madeira and then migrate north for feeding. For many others, however, there is only scant information on preferred habitats, migration patterns and spawning grounds and generally such data are available for particular deep sea areas.

Before formulating a management plan, the next step to understand is stock structure. In recent years much progress on this has been made, thanks largely to population genetic approaches. The round-nose grenadier, which is currently managed a single stock unit, has been shown to be comprised several genetically distinguishable population units across its range (Knutsen et al., 2012). On the other hand the black scabbard-fish (*Aphanopus carbo*), which is currently managed as 2 stock units, appears to be single genetic population (Longmore et al 2014). Greenland halibut is a species that shows gradual genetic differentiation with increasing geographic distance throughout its range(Knutsen et al., 2007). Some deep sea shark species such as Portuguese dogfish (*Centroscymnus coelolepis*) and long nosed velvet dogfish (*Centroselachus crepidator*) appear to be globally distributed species (Catarino et al., 2015) and how no significant stock structure. For most other species, however, it simply not known if they represent a single pan-Atlantic stock or multiple semi-independent units.

Knowledge of life history variation is also crucial for predicting responses to exploitation; long lived, slow growing and low fecundity species will in general be highly vulnerable. The roundnose grenadier is estimated to be slow growing and to live to upto 60 years. It is thus not surprising that the stock declined during the 1980-90s (Bailey et al., 2009), although in more recent years it has at least been stable (ICES, 2015a; Neat and Burns, 2010). The orange roughy (*Hoplostethus atlanticus*) is exceedingly long lived (max age ~ 150 years) and slow growing. It has been drastically depleted from some areas by fisheries. For the black scabbard fish, longevity estimates vary from 10 to 30 years which indicate little more than how uncertain the state of knowledge is. Blue ling (*Molva dypterygia*) is estimated to have a maximum age of 25. Unlike many deep-sea species, however, blue ling is highly fecund and should in theory support a sustainable fishery. The large deep sea sharks such as the leafscale gulper shark (*Centrophorus squamosus*) live in excess of 75 years , have very slow growth and only produce a handful of offspring every second year, all of which renders them highly vulnerable to exploitation.

Stock trends and impacts of past fishing are reasonably well informed for recent years. There are fisheries independent trawl survey time series, e.g. Neat & Burns (2010), Neat et al. (2015) that give insight into change in relative abundance over time and there are some pre-fishery data sets from the 1970s for comparison (Bailey et al 2009). Information on fisheries activity and landings is largely lacking for the period prior to regulation, i.e. before 2003, and there is little consensus on how the large stocks such as blue ling and orange roughy must have once been to have supported the short lived, but lucrative fisheries they did.

In summary, relative to shallow water commercial species, deepwater species must still be considered 'data-poor' from a stock assessment point of view. There is just about enough information from some species, such as roundnose grenadier, to benchmark assessments (ICES, 2015b), but for most other species this is not realistic. Without major investment in coordinated surveys, it is unlikely that this situation will change. Thus alternative approaches are required to attempt to assess population parameters. There are signs that stocks are no longer declining and some cases there is evidence of gradual recovery, but it will probably take several more years before they are sufficiently abundant to support sustainable harvesting.

#### 2.1.2 Current knowledge on stock status

The commercial exploitation of fish and shellfish stocks within the European exclusive economic zone (EEZ) is evaluated through assessments carried out by ICES (FAO area 27), GFCM and STECF (FAO area 37).

These organizations compile large datasets with information about the fisheries targeting the stocks (fishing effort, landings and discards of each species), which is complemented by scientific surveys providing indications about stock fluctuations and demographic structure, as well as individual growth and reproduction patterns. These datasets are made available to a panel of scientists, which using statistical models estimate the status of the stock with regards to maximum sustainable yield (MSY).

The most recent evaluation (STECF, 2015) showed that 38 out of 62 stocks assessed within the ICES area were outside safe biological limits in 2013, the last year of data. In the Mediterranean and Black Sea (FAO area 37) the situation is worse, with about 95% of the stocks being exploited above the maximum sustainable yield levels (Scientific, Technical and Economic Committee and for Fisheries (STECF), 2016).

Although these figures show a dire situation, the trend over time shows that more stocks are now exploited within safe biological limits than ever in the past.

Nevertheless, the number of deep sea species assessed and about which the status of the stock can be evaluated is minor. STECF (2016) assessed only a small number of deep sea species with Total Allowable Catches set by the European Union, namely some stocks of deep-water shrimps, Norway lobster and anglerfish.

#### 2.1.3 Pitfalls, needs and challenges

The evaluation of stock status is hampered by the fact that for a large number of stocks their exploitation status is unknown. Limitations in scientific knowledge, that underpins stock assessments, are mostly due to limited data availability and/or the poorly understood biology and response to environmental conditions. This is particularly true for deep sea fish. Furthermore, individual stocks that are not subject to management measures or are aggregated in groups, tend to get less attention, both in terms of funding and human resources. This is why the number of (deep sea) stocks for which there is no information about the exploitation status may be large, an issue that is difficult to tackle.

Traditional stock assessment methods rely on solid and ample information about growth, reproduction, abundance fluctuations, stock-recruitment relationships and fleet dynamics. For most of deep sea stocks such information is scattered or simply non-existing. For example, deep-sea scientific surveys, that do normally support stock assessments, are difficult to carry out, due to large areas that have to be covered together with limited accessibility to deep sea waters. This renders the application of traditional stock assessment models almost impossible, very unreliable and burdened with high levels of uncertainty.

This situation requires from the scientific community the development of methodologies that do not depend on complex and vast cross-disciplinary datasets, nevertheless being able to provide information about the fluctuation of stock's abundance and their reaction to fishing pressure. One approach that might possibly fulfil such a requirement is Close-Kin Abundance Analysis.

# 3. Close-Kin Abundance Analysis and the Management of Deep Sea Species

To assess the feasibility of genetic close-kin analysis for deep sea species this chapter explains the concept of this approach and its application for SBT abundance estimates. Added value but also pitfalls are discussed and put into the context of deep sea fisheries.

#### 3.1 The Theory of Close Kin Abundance Analysis based on Genetics

Close kin abundance analysis is based on the principles of DNA based paternity testing, i.e. the use of DNA profiling to determine whether two individuals are parent and offspring. In that case they form a Parent-Offspring Pair (POP). Secondly close kin abundance analysis builds on the fact that all offspring have two parents.

DNA testing is currently the most advanced and accurate technology to determine parentage. In a DNA parentage test, the result ('probability of parentage') is 0% when the alleged parent is not biologically related to the offspring and the probability of parentage is typically 99.99% when the alleged parent is biologically related to the offspring.

In diploid cells or organisms chromosomes are available in pairs, one from each parent, i.e. each gene has two copies. As genes are subject to mutation they exist in variant forms in populations, so called alleles. Each individual has a unique "DNA fingerprint" (with the exception of for identical twins) and inherits half of its DNA from one parent and the other half from the other parent. A parent and its offspring must have at least one matching allele at every diploid locus. For parentage testing a number of loci will be analysed and compared between potential parent-offspring pairs.

When examining a large number of genetic loci in a sample of animals the probability that two unrelated animals have the same allele at every locus is very low, which means that, provided the analysis is carried out properly, the probability of identifying false positive POP is equally low.

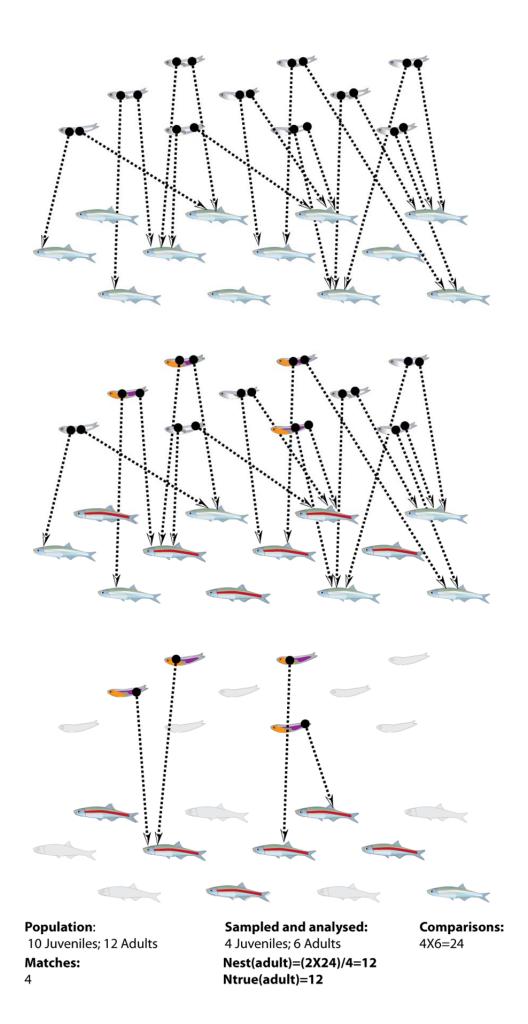
False negatives, the rejection of true POPs, can occur in rare cases through spontaneous mutations from one generation to the next or through genotyping errors.

The identification of POPs can be used for abundance estimates of fish as explained in Bravington et al's report (Bravington et al., 2014): Let's suppose in a sample there are  $n_p$  randomly selected adult animals, and  $n_j$  juveniles. Both adults and juveniles are genotyped using a number of genetic loci that excludes the identification of false positives. What is the probability that a chosen adult is actually a parent of a juvenile? - Since the juvenile must have had two parents, the probability that the chosen adult is one of those two is 2/N, with N being the number of adults alive when the juveniles were spawned. This comparison is repeated for the same juvenile and all other sampled adults. The expected number of hits between that juvenile and the entire set of  $n_p$  adults is  $2n_p/N$ . This process is repeated for all sampled juveniles and the expected total number of hits, is  $2n_jn_p/N$ . If h is the actual number of hits (matches), an estimate of N can be deduced:

#### $N_{est} = 2n_j n_p / h$

This estimate represents the number of specimen in an assessed area. The concept of close-kin abundance estimates is illustrated in Figure 3.

Figure 3 (next page): The concept of close-kin abundance estimates. The DNA-profile of the juveniles ( ) identifies ("tags") its two parents ( ). Top: All parents and juveniles and parental relationships are displayed. Middle: highlights all sampled adults ( ) and juveniles ( ). Bottom: All sampled fish that are analysed genetically and the identified POPs as well as the process of calculating the abundance estimate. Note: For demonstration purposes in this example the estimate is exactly equal to the true number of adults. Also if two juveniles match the same adult two POPs are counted. Details see text. Changed from Bravington et al. (2014).



# 3.2 Close-Kin Abundance Analysis using genetic markers applied to Southern Bluefin Tuna Management: A paradigm?

Southern bluefin tuna (*Thunnus maccoyii*) is a highly exploited species which is classified as 'critically endangered' on the IUCN's Red List of threatened species (International Union for Conservation of Nature (IUCN), n.d.). This large pelagic and highly migratory fish occurs in temperate to cold seas. At maturity, which is reached after 10–12 years, SBT undertake annual migrations between feeding grounds in temperate waters to a single spawning ground in the northeastern Indian Ocean near Java.

The biology and life history traits of SBT are very well known (reviewed in (Shomura et al., 1994)), including its reproductive behaviour (Farley et al., 2015). SBT stocks are consistently monitored, abundant effort is put in their management (Kurota et al., 2010) and the SBT fishing industry is extensively documented (Miyake and Food and Agriculture Organization of the United Nations, 2010). Also genetic and genomic research on this species is progressing swiftly (Yasuike et al., 2015).

The abundant knowledge about SBT renders it an ideal species for close-kin analysis, with one of the major advantages being the fact of a single existing (and known) spawning ground. To start with this feature facilitates the sampling strategy development and effort tremendously.

In summary, for the study by Bravington *et al.* (2014) 14000 SBT were sampled, ensuring a good coverage of spawning ground and spawning season. The authors of the study state that an optimal sampling scheme would aim to enable the genotyping of an equal numbers of juveniles and adults, as that is likely to yield the greatest number of POPs for a fixed amount of genotyping effort. In their case however, for logistical reasons, the balance was shifted slightly towards juveniles, without a great impact on the study itself.

An initial genotyping of 5000 fish led to the development of 25 genetic markers, here microsatellites, to identify POPs. Using pairwise genotype comparison on the sampled fish 45 POPs were identified. Employing a "close-kin abundance model for SBT" led to an estimate of numbers of 10+yr-old SBT by year, 10+ yr-old biomass of SBT by year, and an estimated annual recruitment over the period covered by the project (2002-2010). The results were compared to results obtained with the Operating Model (OM) that is employed by the Commission for the Conservation of Southern Bluefin Tuna for stock assessments. For example the estimated abundance of 10+ adults for 2004 was 1.73X10<sup>6</sup> fish with a biomass of 141kT as compared to 2.04X10<sup>6</sup> fish, with a biomass of 157kT for the OM. While there was a declining trend in the estimated 10+ biomass observed over the period (14%), estimates of annual recruitment to the spawning population were relatively stable around the average for the period.

The close-kin analysis can in principle answer three crucial questions for fisheries management: the number of POPs (given the number of comparison) essentially sets the scaling of absolute abundance, the age and length distribution within the POPs informs on selectivity/fecundity, and the distribution of time-gaps within the POPs essentially determines survival.

However, even if the application of the close-kin abundance estimate occurred under rather ideal conditions for SBT, some drawbacks need to be highlighted.

The first one refers is the fact that the success of this approach depends on an extensive knowledge about the biology and life-history traits of the target species, a condition that is not given for many commercially exploited marine fish species.

Secondly the sampling effort needed to support the close-kin abundance estimate is considerable: Almost 14000 SBT individuals were genotyped for this study. Such a high number may well not be feasible for many other species.

The authors propose that an optimum cost-effective number and frequency of adult and juvenile samples to collect in future should be established. They also emphasize that

that the annual sample sizes and costs are likely to be much lower than for the study as it was carried out. This is because of the "quadratic efficiency" of close-kin methods. Each new sample (whether juvenile or adult) gets compared against each pre-existing sample of the other type. Because a large catalogue of older samples has already been built up, the number of new samples required to find a given number of new POPs will be lower than before.

The authors state that that the close-kin data could be incorporated into the current Operating Model (OM) of the Commission for the Conservation of Southern Bluefin Tuna (CCSBT) without major structural modifications. On the other hand the estimates of absolute adult abundance from the close-kin estimate are about 3 times those from the base case from the CCSBT OM. The independent estimate of adult mortality is similar to that estimated from the CCSBT OM. The authors note that currently a direct comparison to results stemming from the CCSBT OM is not possible for a variety of reasons, but believe that the close-kin estimate does indicate that the absolute abundance of spawning SBT is considerably higher than previously thought. They conclude that ultimately these questions can only be settled through a full incorporation of the close-kin data into the full CCSBT OM.

The approach and results have been thoroughly reviewed by CCSBT, but to date a peerreviewed publication is not available. A manuscript appears though to be under revision (personal communication, 2015). This would help to further assess the value of the presented approach, particularly its application for other species.

# 3.3 Close-Kin Abundance Analysis using genetic markers applied to deep sea species: considerations

The authors of the study by Bravington *et al.* (2014) claim that there is considerable potential for applying close-kin methods to other species that are hard to monitor, whether commercial targets, non-commercial targets, or by-catch.

Currently it is envisaged to extent this approach to Pacific bluefin tuna (*Thunnus orientalis*) (NOAA, 2015) and some shark species of Australian waters (CSIRO – personal communication, 2015).

It is generally recommended that in each case, careful consideration should be given to the details of life-cycle, stock structure, sampling logistics, and likely adult abundance, to determine whether the approach is feasible and/or likely to be cost-effective. As already stated above, the most obvious limitation is that very abundant populations require very large numbers of samples (proportional to the square root of abundance) to find enough POPs for useful estimates, implying high effort and costs in most cases.

While the single-stock adults-vs-juvenile comparison of SBT is perhaps the simplest version of close-kin that could be imagined, some theory has now been developed for other variants such as adult-vs-adult comparisons, which would be more appropriate for many commercial target species. It is even possible, using the newest genetic technology, to work only with juvenile samples, looking for Half-Sibling Pairs rather than POPs. Currently CSIRO is testing this on elasmobranchs.

For the deep sea species listed in Annex I of Council Regulation No. 2347/2002 most of the caveats mentioned above are valid. In particular, while frequently the simultaneous catch of spawners and adults is possible, knowledge about spawning areas is very limited. Moreover juvenile samples are scarce and generally collecting enough samples to sufficiently support a close-kin abundance estimate study on deep sea species would require a major effort and resources.

These considerations do not preclude an added value of a genetic close-kin abundance analysis for a number of deep-sea species, but hint to the fact that embarking on such a study bears considerable risks.

While discussing options and strategies it emerged that white anglerfish (*Lophius piscatorius*) might be a potential test species. Strictly speaking not a deep-sea species,

but sharing some common features, it is a highly commercial species. White angler fish is assessed in ICES divisions VIIIc and IX (Cantabrian Sea, Atlantic Iberian waters), which constitutes an asset as there would be a reference. The ICES technical advice on white anglerfish states that the adult stock size is unknown, and that fishing pressure is too high to ensure an optimal use in the long term. It was exploited above MSY in 2014 (International Council for the Exploration of the Sea, 2015c).

The stock assessment of white angler fish would benefit from an independent approach such as the close-kin analysis. Obtaining a sufficient number of samples is feasible. Moreover Marine Scotland undertakes an annual trawl survey for this species – albeit only covering only a fraction of its range. Therefore, currently absolute abundance estimates of the stock are tentative and indeed the true stock size remains highly controversial and its response to the present fishing pressure remain poorly understood. Nevertheless Marine Scotland holds robust data on trends in relative abundance over the past 10 years, including distribution and maturity, all valuable information in the context of an eventual close-kin study.

A variety of studies have addressed the population structure of the white anglerfish providing some evidence for geographical isolation of the early life stages (Swan et al., 2004) but generally limited genetic structure (Charrier et al., 2006) (Farina et al., 2008).

In summary, it appears that knowledge about *Lophius piscatorius* and its commercial exploitation as well as management combine features which render this species a good candidate for a close-kin abundance estimate study which might ultimately also serve as a paradigm for deep-sea species.

# 3.4 Integration of genetic close-kin abundance analysis with other fisheries science approaches

Fisheries science is a research field covered a broad array of disciplines such as marine biology, ecology, population dynamics, (bio-)economics, sociology and management. Understanding and accommodating the complexity inherent to the exploitation of marine living resources as to move towards management schemes underpinning profitable as well as sustainable exploitation comes along with need to integrate these originally disparate disciplines. While this is generally acknowledged (Dörner et al., 2015)(WFC 2016 - Session 1 and 7 [http://www.wfc2016.or.kr/english/02\_program/02\_program.asp]), moving towards the implementation of a truly integrative approach proves difficult, but can be achieved at least partially.

As outlined above, classically stock identification and abundance estimates, or stock assessments, relies on the collection and compilation of large datasets with information about the fisheries targeting the stocks (effort, landings and discards for each species), which is complemented by scientific surveys providing indications about stock fluctuations and demographic structure, and individual growth and reproduction patterns.

There is a long debate about the benefits emerging from the addition genetic information to stock identification and assessment which has in recent times been fueled by the enormous progress in genetics and genomics (Cuéllar-Pinzón et al., 2016; Ovenden et al., 2015). Particularly for stock identification the value of genetic information is increasingly acknowledged and applied (Fiorentino et al., 2014) with Pacific salmon management being a paradigm (http://www.adfg.alaska.gov/index.cfm?adfg=fishinggeneconservationlab.bbaysockeye baseline; https://vimeo.com/110201354) and Baltic cod providing an impressive example for a major commercially exploited marine fish species (Eero et al., 2014).

Using genetic information to support abundance and biomass estimates for marine fish, and their integration into novel fisheries modelling frameworks has been discussed amongst geneticists and modelers during the kick-off meeting of the JRC Assessment for All initiative in 2012 (a4a - <u>https://fishreg.jrc.ec.europa.eu/web/a4a</u>). While at that time

no concrete conclusion was drawn genetic close-kin abundance analysis, if feasible, can complement other established stock assessment approaches.

Beyond that, once genetic close-kin abundance analysis has shown its feasibility and value, a reflection amongst fisheries geneticists and fisheries assessments experts will be needed how to best integrate this approach into existing data collection, assessment and fisheries management frameworks.

# 4. Conclusion and Outlook: Towards development, application and technology transfer

In the course of a visit to the JRC December 2013, CSIRO presented its SBT close-kin analysis approach, from which a discussion emerged whether and to which marine fish species with CFP relevance out could be applied. At that time, due to the difficulties their management faces, along with some of the biological characteristics that are known, deep-sea species were considered among potential candidates. The present Technical Report serves to provide a first assessment about feasibility and challenges. Generally the conclusion is that probably none of the deep-sea species listed in Annex I of Council Regulation No. 2347/2002 is currently ideally suited for such an approach. However, as depicted above, white anglerfish might be a species that would be worthwhile to be studied with close-kin analysis, and which might serve as a paradigm for commercially exploited deep-sea species.

As has been shown in the report of by Bravington et al. (2014), this new approach can in principle be integrated into existing management and assessment frameworks and valuably complement those as it is a fishery-independent abundance estimate.

This, along with estimated resource needs will be further explored and reported in the frame of one of the Terms of Reference of the 2016 ICES Working Group meeting (May 2016, Queens University, Belfast, UK).

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### List of abbreviations and definitions

CCSBT: Commission for the Conservation of Southern Bluefin Tuna CECAF: Fishery Committee for the Eastern Central Atlantic CFP: Common Fisheries Policy CSIRO: Commonwealth Scientific and Industrial Research Organisation EEZ: Exclusive Economic Zone EU: European Union FAO: Food and Agriculture Organization of the United Nations GFCM: General Fisheries Commission for the Mediterranean ICES: International Council for the Exploration of the Sea MS: EU Member State(s) MSY: Maximum Sustainable Yield POP: Parent-Offspring Pair (POP) RFMO: Regional Fisheries Management Organizations SBT: Southern bluefin tuna STECF: Scientific, Technical and Economic Committee for Fisheries TACs: total allowable catches VMS: Vessel Monitoring System WGAGFM: ICES Expert Working Group on Applied Genetics for Fisheries and Mariculture

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Figure 3 (p.14): The concept of close-kin abundance estimates. The DNA-profile of the juveniles identifies ("tags") its two parents. Top: All parents and juveniles and parental relationships are displayed. Middle: highlights all sampled adults and juveniles. Bottom: All sampled fish that are analysed genetically and the identified POPs as well as the process of calculating the abundance estimate. Note: For demonstration purposes in this example the estimate is exactly equal to the true number of adults. Also if two juveniles match the same adult two POPs are counted. Changed from Bravington et al. (2014).

### **Useful links**

DEECON: Unravelling population connectivity for sustainable fisheries in the Deep Sea. <u>http://www.imr.no/prosjektsiter/deecon/en</u>

POPSIZE: Estimating effective population size in populations of marine fish: an approach using interannual fluctuations of the genetic composition <u>http://ices.dk/community/icessciencefund/Pages/POPSIZE.aspx</u>.

### ANNEX

Deep Sea Species listed in Annex I of Council Regulation No. 2347/2002

Scientific name	Common name	TAC
Aphanopus carbo	Black scabbardfish	
Apristuris spp.	Iceland catshark	
Argentina silus	Greater silver smelt	
Beryx spp.	Alfonsinos	
Centrophorus granulosus	Gulper shark	
Centrophorus squamosus	Leafscale gulper shark	
Oracher and Himme for heide !!	Dia di da 26 di	
Centroscyllium fabricii	Black dogfish	
Centroscymnus coelolepis	Portuguese dogfish	
Coryphaenoides rupestris	Roundnose grenadier	
Dalatias licha	Kitefin shark	
Densis selector	Divide a de aña la	
Deania calceus	Birdbeak dogfish	
Etmopterus princeps	Greater lanternshark	
Etmopterus spinax	Velvet belly	
Galeus melastomus	Blackmouth dogfish	
Galeus murinus	Mouse catshark	
	WOUSE CAISIIAIN	
Hoplostethus atlanticus	Orange roughy	
Molva dypterigia	Blue ling	
Phycis blennoides	Forkbeards	
Centroscymnus crepidater	Longnose velvet dogfish	
	FouPunge Active moBusit	
Scymnodon ringens	Knifetooth dogfish	
Hexanchus griseus	Six-gilled shark	

Chlamydoselachus anguineus	Frilled shark	
Oxynotus paradoxus	Sailfin roughshark (Sharpback shark)	
Somniosus microcephalus	Greenland shark	

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