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Report of the NAFO Scientific Council Working Group on Reproductive Potential

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The NAFO Working Group on Reproductive Potential is comprised of 21 members representing 10 countries (Canada, Denmark, Germany, Greece, Iceland, Norway, Russia, Spain, United Kingdom, and USA).

Over the past year, Working Group members worked inter-sessionally by correspondence and ad hoc meetings at other scientific forums to address the ToRs approved by Scientific Council in June 2008. Several meetings of the EU COST Research Network Action Fish Reproduction and Fisheries (FRESH) (Coordinator: Fran Saborido-Rey, Spain) were also held in the past year. Following recommendations from the 2010 Scientific Council Meeting, the two groups continued to maintain an informal, mutual working relationship over the past year. This enabled the development of collaborations among scientists that benefited addressing NAFO ToRs and avoided duplication of effort between the groups allowing the WG to bring more results to the attention of Scientific Council.

A brief summary of progress and future plans of each ToR are given below:

ToR 1: Explore and conduct evaluation of underlying assumptions of protocols used to estimate total realized egg production of selected marine species and stocks. Co-Leaders: Rick Rideout (DFO, Canada) and Rosario Dominguez (CSIC, Spain)

Several marine laboratories in the North Atlantic have initiated routine fecundity estimation for key fish stocks. This information is being used to (i) help improve the estimation of stock reproductive potential (ii) understand population productivity and (iii) predict stock recovery rates. However, there is a lack of standardization and calibration of various methods to estimate fecundity among laboratories. For example, some laboratories have only recently initiated the autodiametric method and are developing appropriate calibration curves. On the other hand, observations have been made that indicate atresia and timing of sampling can influence estimates of total egg production. Techniques to quantify atresia (vitellogenic oocyte resorption) will be developed and evaluated in this ToR. This will involve histological analyses accompanied by computerized image analysis.

Establish Standard Operating Procedures:

- Provide uniform and standardized procedures for routine fecundity analyses in laboratories using a variety of methods, i.e. autodiametric method, image analysis
- Evaluate histological techniques for assessment of atresia

Validation of Assumptions:

- Test assumptions of different fecundity methods (i.e. the autodiametric method) and parameters associated with fecundity estimation
- Estimate down regulation of fecundity and quantification of atresia and non-annual spawning

The autodiametric method to expedite the estimation of fecundity of Greenland halibut was developed by Spanish and Canadian researchers (R. Domiqnguez-Petit, R. Rideout and others). The relationship between oocyte density

and diameter was significant (r2> 0.90) for fish from the Gulf of St. Lawrence and both inside and outside of Canada's 200 mile jurisdictional area off Newfoundland. Calibration curves were used to estimate and compare potential fecundity from the different areas. This method once validated will assist to develop long-term, fecundity data bases for this species. These findings were presented at the COST Fish Reproduction and Fisheries Conference, May 16-20, Vigo, Spain.

Additional work in this area by Spanish researchers highlighted the use of free software Govocitos which automates the process of counting, classifying and measuring different types of oocytes from histological images and estimates fecundity based on stereological principles.

A key deliverable for this ToR is the development of a resource handbook that provides detailed methodology for the estimation of input variables (e.g., age/size at sexual maturity, egg production) for the estimation of stock reproductive potential over a wide suite of fish species. This handbook will address one of the main aims of this ToR: to provide uniform and standardized procedures along with the pros and cons of various methods. An outline of the handbook's chapters is provided. Preparation of chapters has been ongoing for approximately one year, includes over 30 contributors and is showing strong progress, particularly in Chapters 3 and 4 which are the more comprehensive chapters. The next year's activities will see this contribution come near to completion. The outline of the handbook is given below:

Handbook of Applied Reproductive Biology for Fish Stock Assessment and Management

Editors: R. Dominguez-Petit, H. Murua, F. Saborido-Rey, and E.A. Trippel

1. Fish Reproductive Biology (Leaders: Olav S. Kjesbu & Mark Dickey-Collas)
- 1.1. Reproductive Biology: A Brief Historical Review
- 1.2. Reproductive strategies
- 1.3. Reproductive Potential
  - *Maturity estimate,Egg production*
- 1.4. Reproductive Potential and its importance for Fish Assessment and Management
  - *Stock-Recruitment Relationships*

2. Data Collection and Statistics for Reproductive Biology Studies (Leader: C. Stransky)
- 2.1. Sampling methods and design
- 2.2. Data collection. Data sources. Accuracy and precision.

3. Maturity (Leader: Cindy Van Damme)
- 3.1. Stock structure
  - *Length/Age stock composition, Age-Length Keys ,Sex ratio*
- 3.2. Maturity staging
  - 3.2.1. Microscopic Maturity Staging
    - *Histological Processing,Staining Processing*
  - 3.2.2. Macroscopic Maturity Staging
    - *Females (*Oviparous/Viviparous), *Males*
  - 3.2.3. Validation and methodological calibration
- 3.3. Alternative Methods to Estimate Maturation
  - *Biochemical methods,Somatic indices*
- 3.4. Maturity Ogives and Spawning Proportion
  - *Age based, Length based, Cohorts based,Seasonal based*

4. Egg Production: Leader: Kostas Ganias)
- 4.1. Fecundity
  - 4.1.1. Fecundity measures (Laboratory issues)
    - *Gravimetric Method + Image Analysis, Autodiametric Method, Image analysis*
    - *Stereological Methods (Disector, Fractionator)*
  - 4.1.2. Potential Fecundity
  - 4.1.3. Batch fecundity

4.2. Spawning Fraction and batch frequency
Sampling, Laboratory (POF staging and ageing method), Analysis, Alternative to the POFs methods
4.3. Regulation of egg production
Fecundity down-regulation, Skip spawning,Non annual spawning
How to move from individual to population scale
4.4. Population Egg Production
4.4.1 Determinate spawners
Annual Egg Production, Daily Egg Production
4.4.2 Indeterminate spawners
Annual Egg Production, Daily Egg Production

5. Sperm Production (Leader: E.A. Trippel)
5.1. Experimental techniques to evaluate male reproductive potential
5.2. Testes development
Recruitment of spermatocytes, gonadosomatic index, histological techniques
5.3. Sperm density
Spermatocrit, Number of sperm per ml
5.4. Sperm motility
Sample preparation, Recording of motility, Software/data analysis
5.5. Sperm morphology and biochemistry
Sperm head shape ,size, tail length,staining techniques, available software
Sperm viability kits, seminal plasma composition, pH, ATP, LDH
5.5. Population sperm production
Body metrics and predictions of male fertility and moving from individual to population scale, consideration of wild vs. laboratory results, sperm limitation, sex ratio.

6. Elasmobranch Reproductive Potential (L. Serrano)
6.1 Modes of reproduction
Oviparity, Aplacental yolk-sac viviparity, Aplacental viviparity with trophenomata or uterine villi
Aplacental viviparity with oophagy or intra-uterine cannibalism, Placental viviparity
6.2 Reproductive traits
Females, Males
6.3 Maturity staging
Macroscopic staging, Microscopic staging
6.4 Reproductive cycle
Ovarian cycle, Testis and spermatogenic cycle
Timing and location of copulation, Embryonic development and gestation period
Timing and location of egg-laying (oviposition),Timing and location of parturition
Resting years
6.5 Reproductive parameters
Gonad weight and gonadosomatic index, Clasper length
Maturity ogive, Maternity ogive, Ovarian fecundity, Uterine fecundity
Pup size, Sex ratio at birth, Temporal and spatial differences in reproductive parameters

Annex I: Glossary: S.Lowerre-Barbieri)

This annex defines and standardizes terminology used in fish reproductive ecology studies based on the most recent discussions at the 4th Workshop on Gonadal Histology of Fishes (Cadiz, Spain).

ToR 2: Explore and investigate the potential effects of changes in water temperature and food supply on reproductive success in selected marine species and stocks. Co-Leaders: Richard McBride (NMFS, USA) and Stylianos Somarakis (HCMR, Greece)

Environmental factors can modify the reproductive potential of fish stocks and thereby influence recruitment. Annual variations and potential increases in water temperature due to climatic warming will presumably act strongly

to influence gonadal development and reproductive success. Prey resources also vary and influence fish condition which in turn affects reproductive output. In this ToR, using data on specific stocks and laboratory experiments, the influence of specific abiotic and biotic factors on gonadal development and spawning will be evaluated pending available data.

Abiotic: Examine changes in water temperature (short and long-term) and their effects on timing and duration of spawning, fecundity, egg size and fertilization success

Biotic: Assess variation in prey resource type and abundance and their effects on egg production and gamete quality

ToR 2 was divided into two components

(a) Explore and investigate the potential effects of changes in water temperature on reproductive success in selected marine species and stocks.

For this Sub-ToR, the effects of age and temperature on spawning time in cod and haddock stocks of both sides of the Atlantic were investigated. Results showed that age of fish influences spawning time in a step function rather than linear (analyses based of proportion spend fish in research surveys). There was significant annual variation in spawning time but little evidence of a direct effect of temperature. Spawning time in the Northwest Atlantic cod stocks has become later in recent years (J. Morgan, P. Wright and R. Rideout). These findings were presented at the COST Fish Reproduction and Fisheries Conference, May 16-20, Vigo, Spain.

Viability and development during early life phases of Atlantic cod and Baltic cod in relation to paternity and water temperature were investigated through laboratory experiments. Results showed that paternity and temperature interacted to influence embryonic survival and larval characteristics to a significant extent indicating adaptive ability to temperature changes exists and can be expressed through female mate choice (F. Dahlke, S. Politis, M. Peck and E. Trippel). These findings were presented at the COST Fish Reproduction and Fisheries Conference, May 16-20, Vigo, Spain.

Fecundity, egg characteristics and embryonic development of captive Greenland halibut from the Gulf of St. Lawrence were investigated. Findings demonstrated that contrary to most groundfish in the NAFO area, Greenland halibut is a single batch spawner. It produces eggs of a large size (3.4-4 mm in diameter) and hatching time is strongly dependent on incubation temperature. At 2ºC, 4°C and 6°C, the time at 50% hatching is 46, 30, and 24 days, respectively. Egg buoyancy experiments also confirm that the eggs are bathypelagic and that important changes in buoyancy in the last 3-4 days before hatching result in hatching of the larvae in the upper part of the water column. (Y. Lambert, R. Dominguez-Petit, and P. Ouellet).

(b) Explore and investigate the potential effects of changes in food supply on reproductive success in selected marine species and stocks.

Fish reproductive strategies in relation to trophic dynamics of their environment are being investigated for over 30 fish species. The reproductive traits being evaluated include between year spawning frequency (semelparity vs iteroparity), synchrony of vitellogenic follicle development (synchronous, group-synchronous or asynchronous), recruitment of vitellogenic follicles with the ovary each year (determinate vs indeterminate) and with-in year spawning frequency (batch vs. total spawning). These reproductive traits are being matched with evidence that food amount or food type affect egg production in fishes. A manuscript for publication is underway and preliminary results were presented at the COST/FRESH Conference in Vigo, Spain (lead R. McBride).

Examination of reproductive strategies and fecundity type regulation through food availability in marine fish was made. Food availability was shown to be the most important factor regulating fecundity type (capital (determinate) vs. income (indeterminate) spawners). From a principal component analysis, it was revealed that other important factors for the determinate fecundity type include body condition, egg dry weight and latitude. For the indeterminate fecundity type, the additional significant factors were relative fecundity, spawning period and temperature. A manuscript for publication is being pursued. (C. van Damme, A. Rijnsdorp, M. Dickey-Collas and O.S. Kjesbu).

ToR 3: Undertake appraisal of methods to improve fish stock assessments and fishery management advice that incorporate new biological data for highly exploited and closed fisheries. Co-Leaders: Joanne Morgan (DFO, Canada) and Loretta O'Brien (NMFS, USA)

The depressed and age-altered state of many marine fish stocks has led to reduced landings and in some instances fishery closures. New biological data associated with these altered states will be used to forecast recruitment and improve the accuracy of stock assessment advice. Building on information from previous WG ToRs, the intrinsic rate of population increase will be utilized to assess the timeframe for selected stocks to recover under various fishing and environmental conditions.

Recruitment prediction: Improve prediction of incoming year class size and develop new stock-recruitment models and biological reference points based on better estimates of stock reproductive potential. This includes testing whether more complex indices of reproductive potential result in better estimates of recruitment and limit reference points. Develop scenarios which model population reproductive responses to extrinsic factor data developed in ToR 2.

Stock recovery: Evaluate the intrinsic rate of increase of selected stocks under differing conditions of reproductive potential and levels of fishing mortality to aid in the development of reopening criteria. Estimate recovery time for specific stocks to achieve target biomass levels.

Egg production methods can estimate spawner biomass and/or stock numbers independently of commercial fisheries data. Improved information on stock reproductive potential is improving the accuracy of these methods. The daily egg production method is being explored to evaluate adult stock size for determinate spawning species in the Baltic and North Seas.

An excellent contribution was made that clearly reveals the environmental and fishing limitations to the rebuilding of the northern Gulf of St. Lawrence cod stock (published in Can. J. Fish. Aquat. Sci. 68: 618-631). Time series of life history traits were analysed during the time period of stock collapse and the period of expected but failed recovery. Estimates of the *r*, instrinsic rate of increase (measure of stock productivity) were negative when the stock collapsed, indicating the biomass would have decreased even without fishing. Population abundance projections for the recent period suggest a potential increase in population size of 7.3% per year, with a doubling time of 10.5 y in the absence of exploitation and a near 0% rate with current fishing mortality, indicating that present harvesting does not allow any rebuilding of the stock. Given the similarities in environmental conditions and key life history traits, the pattern for this stock may have been repeated in other low productive Northwest Atlantic cod stocks over the past 20 years (Y. Lambert).

The mean lengths of spawners were estimated annually for five stocks: Barents Sea cod, Grand Banks cod and Grand Banks American plaice, North Sea haddock and North Sea plaice and varied greatly (range 10-30 cm depending on stock). An examination was made as to whether stocks dominated by small individuals have lower reproductive rates (total egg production divided by SSB) than those with large individuals. In each stock, positive correlations were found between size diversity and reproductive rate. Current stock assessment protocols, including application of biological reference points to evaluate stock status, are insensitive to the impact that interannual variation in length composition is having on reproductive rates (T. Marshall, J. Morgan, P. Wright, A. Rijnsdorp). These findings were presented at the COST Fish Reproduction and Fisheries Conference, May 16-20, Vigo, Spain.

Environmental influences on SRP were investigated using the intrinsic rate of population increase r, derived from life table analysis, that incorporates characteristics (e.g. growth rate, fecundity, etc.) which are also associated with SRP. Trends in r were compared among nine Atlantic cod Gadus morhua stocks (Northern Gulf of St. Lawrence, Northeast Arctic, Georges Bank, Gulf of Maine, Baltic, Icelandic, Irish Sea, Flemish Cap, and West of Scotland) with time series varying in length between 22 and 56 years for the time period 1946-2005. Cod west of Scotland had the highest r over the observed period while Northern Gulf of St. Lawrence cod had the lowest r. Tests for significant differences of mean r between stocks indicated five distinct groups, for example, cod from Northern Gulf of St. Lawrence and West of Scotland were significantly different from each other and the other three stock-groups. Stock specific environmental variables, including a global climatic variable (Northwest Atlantic Oscillation) and life

history characteristics relative to r were analyzed to further investigate the environmental influences on SRP. (L. O'Brien and 9 others).

Notice has been made that more fully exploring a stock's reproductive potential is most important or critical when a stock becomes depleted, here a multitude of factors such as sex ratio, spawning sites, number of brooders, etc may come into play, over and above the actual level of egg production. A review was made of the potential factors that can have a significant affect on reproductive potential of a stock over a broad range of species and life histories. These should be included, particularly under stressed conditions, to more appropriately represent a stock's ability to produce viable eggs and larvae (R. Nash and O.S. Kjesbu). These findings were presented at the COST Fish Reproduction and Fisheries Conference, May 16-20, Vigo, Spain.

A study was conducted to determine if recruitment is better predicted using more complex indices of reproductive potential (IN PRESS Can. J. Fish. Aquat Sci.). This study examined four NAFO stocks: 3LNO American plaice, 3NO cod, 3M cod and 2+3KLMNO Greenland halibut. Stock recruit models paired with complex indices of RP gave a better estimate of recruitment in slightly more than half of the tests conducted. When there were larger trends in the reproductive biology (maturity at age, sex ratio and egg production) more complex indices of RP were more likely to provide a better estimate of recruitment. For 3M cod recruitment prediction was clearly improved by using more complex indices of reproductive potential, while for Greenland halibut the best predictions of recruitment came from 10+ biomass. The results for 3LNO American plaice and 3NO cod were intermediate (J. Morgan, A. Perez-Rodriguez and F. Saborido-Rey).

The highlight for ToR 3 is the Workshop on Implementation of Stock Reproductive Potential into Assessment and Management Advice for Harvested Marine Species that was recently held at the University of Aberdeen, Scotland on April 12-14, 2011. This Workshop has been a key deliverable for the NAFO WG on Reproductive Potential for sometime and has now been successfully completed. This workshop was held in conjunction with the EU COST Action Fish Reproduction and Fisheries (FRESH).

The objectives of this workshop were to provide workshop participants with expert advice in implementing information on reproductive potential into the assessment of their stocks and to review and recommend best practices for incorporating information about growth, maturation, condition and fecundity into management of harvested marine species.

The workshop was organized by Tara Marshall (UK), Joanne Morgan (Canada), Loretta O'Brien (USA), Iago Mosqueira (UK) and Santiago Cervino (Spain). Invited presenters were Bridget Green (Australia), Adriaan Rijnsdorp (Netherlands), Peter Wright (UK), Coby Needle (UK), Paul Spencer (USA) and Liz Brooks (USA). Presentations were also made by Joanne Morgan and Santiago Cervino (Spain). The presentations were made under 4 themes: ESTIMATING STOCK REPRODUCTIVE POTENTIAL; IMPLEMENTING ESTIMATES INTO ASSESSMENTS; ARE WE DOING IT BETTER, WORSE OR JUST DIFFERENTLY?; and CODING IT UP. Following the presentations in each theme, areas for further discussion were identified and groups formed to participate in these discussions and report back to plenary. The presentations and discussions allowed people with diverse backgrounds to become familiar with the techniques used to compute SRP, the potential impact of SRP on our perception of stock status and some of the issues around incorporating SRP into scientific advice.

In addition to the organizers and invited speakers, 16 others participated in the workshop. Two NAFO Designated Experts participated; F. González Costas and Dawn Maddock Parsons. The support of NAFO for the attendance of these two DEs and for the organizer L. O'Brien is gratefully acknowledged.

Conclusions of the workshop

It is clear that the incorporation of more complex indices of SRP can make a difference in the perception of stock status. The past trajectory of the stock can be affected as can the estimated limit reference points and current stock status relative to those reference points. In addition, perceptions of projected stock status can vary depending on the information that is incorporated into estimates of SRP. It is also clear that there are no real technical impediments to incorporating this information.

Trends in biological parameters and the quality of the data on these parameters are both important components. There will be a greater difference in perception of stock status if there are large trends in reproductive parameters and advice is more likely to be improved by the incorporation of these data into estimates of SRP. The ability to detect trends in biological parameters will be affected by the quality of the data that are collected (one must be able to detect the signal in the noise). The quality of the data will also affect the ability to detect any difference in various estimates of SRP and will have an impact on the likelihood of improving advice.

Variation in weight at age and in maturity at age are both common and can have a large impact on perceived SRP. Often weight at age is from commercial catch at age and is calculated using an invariant length weight relationship. It is likely that variation in weight at age is greater than currently thought as a result of variation in condition. Consideration should be given to updating length weight relationships on an ongoing basis. Maturity at age should be estimated where possible by cohort and macroscopic classification scales verified with histology.

In general it has been found that changes in fecundity are small and have not had a great impact in variation in SRP. However, data on fecundity tend to be limited more data should be collected to determine if this is indeed the case.

The collection of data on weight, maturity, sex ratio and fecundity is encouraged. Only through the collection of good quality data on these factors can we begin to fully determine how much of an influence there is of not incorporating them into our advice.

Work on whether or not advice is improved by incorporating more biology into our estimates of SRP is only beginning. These studies should be continued and applied to more stocks and species with more varied reproductive strategies. The best approach is likely to be within a management strategy evaluation context. This type of process would require the input of both modelling experts and experts in species biology.

FUTURE ACTIVITIES

The 10th Meeting of the NAFO Working Group on Reproductive Potential is proposed to be held in the latter half of 2012 (further details to be defined by local organizers). This will be the third and proposed final meeting for the 3rd Set of ToRs.