THESE DE DOCTORAT DE

AGROCAMPUS OUEST

ECOLE DOCTORALE N° 598
Sciences de la Mer et du littoral
Spécialité : Ecologie marine

Par
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Structure spatiale et connectivité au sein du stock de sole commune de Manche Est

Apport d'une approche holistique multitraceur

Thèse présentée et soutenue à Rennes, le 13 janvier 2020
Unité de recherche : Ecologie et Santé des Ecosystèmes
Thèse N° : 2020-1_H-112

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À ma grand-mère Françoise

Foreword

This PhD project was conducted between October 2016 and December 2019 at the Ecology and Ecosystem Health research unit (UMR 985 ESE Agrocampus Ouest/INRA) in Rennes, France. This work was supervised by Pr. Olivier Le Pape and Dr. Elodie Réveillac.

The thesis is part of the 'Population spatial structure and connectivity' work package of the SMAC research program (Sole de Manche Est Amélioration des Connaissances pour une meilleure gestion du stock). This program aims at increasing the knowledge of the ecology and exploitation of the common sole of the Eastern English Channel stock to improve its management. The SMAC program is funded by "France Filière Pêche" and the "Direction des pêches maritimes et de l'aquaculture". Three research institutes are involved in this program, Ifremer (Boulogne-sur-Mer and Port-en-Bessin centers), Agrocampus Ouest and the UMR BOREA. Three regional fishermen committees (CRPM Nord Pas de Calais Picardie and Haute et Basse Normandie) and three producer organizations (FROM Nord, CME and OPBN) also participate to the program.

Over the past three years, three scientific papers emerged from the PhD project. The first one (Randon *et al.*, 2018) was published in Journal of Sea Research in September 2018 (special issue of the International Flatfish Symposium, Saint-Malo, France, 2017). The second publication (Randon *et al.,* sub) was submitted to Estuarine, Coastal and Shelf Science in November 2019 (special issue of the International Sclerochronology Conference, Split, Croatia, 2019). Finally, the third article (Randon *et al., in prep*) of this thesis will be submitted to Estuarine, Coastal and Shelf Science (special issue of the SWIMWAY conference, Hamburg, Germany, 2019).

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Remerciements

Voilà, le moment est venu pour moi de clore ces trois années de doctorat. Cette thèse fut pour moi une expérience enrichissante, tant du point de vue de l'apprentissage que du point de vue personnel.

Avant tout, je tiens à remercier mes deux encadrants de thèse, Olivier et Elodie, pour m'avoir donné la chance de vivre cette expérience et pour leur soutien dans les bons moments comme dans les moins bons. Merci à tous les deux pour votre positivisme à toute épreuve et votre humanité qui m'ont permis de me maintenir à flot dans bien des situations. Olivier, un grand merci pour ton efficacité, ta disponibilité et tes réflexions scientifiques (et pas que !) qui font que tu es unique. Une fois que j'ai su « décoder » ton langage (écrit et oral), ce fut extrêmement enrichissant de travailler avec toi. J'ai énormément appris en trois ans et je pense avoir muri scientifiquement grâce à toi. Elodie, merci pour ta bonne humeur et tes nombreux encouragements qui m'ont aidé à prendre confiance en moi. Ces dernières années ont également été intenses pour toi tant du point de vue personnel que professionnel. Je te souhaite le meilleur dans ta nouvelle vie 100% rochelaise. Un grand merci à vous deux donc et surtout merci pour la confiance que vous m'avez accordée.

Je tiens également à exprimer toute ma gratitude aux membres de mon jury de thèse. Merci aux Professeurs Henrique Cabral et Filip Volckaert pour le temps passé à évaluer ce manuscrit de thèse. Merci également à Hélène de Pontual et Bruno Ernande d'avoir accepté d'examiner ce travail.

Je tiens ensuite à remercier les financeurs de cette thèse, France Filière Pêche et la Direction des Pêches Maritimes et de l'Aquaculture. Ce travail de thèse s'inscrit dans le cadre du projet de recherche SMAC porté par Ifremer, l'UMR BOREA et Agrocampus Ouest. De nombreuses collaborations ont émergées de ce projet, notamment avec les scientifiques, pêcheurs et membres de comités des pêches et organisations de producteurs. Ce fut particulièrement stimulant pour moi de faire partie d'un tel programme de recherche. Un grand merci à toutes les personnes ayant participé aux discussions lors des réunions annuelles. Merci à Marie Savina d'avoir porté jusqu'au bout ce projet de recherche et d'avoir su concerter professionnels et scientifiques. Merci également à l'ensemble des professionnels pour leur implication dans les campagnes de marquage et leur apport pertinent dans les discussions.

Ce travail de thèse n'aurait jamais pu se faire sans la contribution de nombreux techniciens et scientifiques ayant participé aux échantillonnages en mer et aux extractions en laboratoire. Je remercie donc Sophie Parrad, Coline Lazard, Margaux Denamiel, Ronan Le Bras, David Osmond, Nicolas Goascoz, Ivan Schlaich, Jérôme Quinquis, Christophe Loots, Thomas Le Berre pour leur participation aux nombreuses campagnes et leur aide au laboratoire. Merci à Véronique Loizeau d'avoir accepté de nous fournir des soles d'estuaire de Seine. Merci également à Graham Doswell d'avoir si gentiment accepté que je vienne échantillonner dans les eaux anglaises sur son bateau de pêche. Je remercie également les collègues belges de l'ILVO et du KU Leuven pour m'avoir fourni des soles de mer du Nord. Merci notamment à Els Torreele, Loes Vandecasteele et Kris Hostens. Enfin, je remercie Johan Smith du Cefas pour son aide dans l'échantillonnage le long des côtes anglaises durant la campagne BTS.

Qui dit échantillonnage dit dissections et analyses en laboratoire. A ce titre je remercie du fond du cœur notre cher technicien de labo Thomas Le Berre. Merci pour ton aide cruciale dans la dissection de mes nombreux poissons (parfois odorants) et merci pour ton humour et ta joie de vivre au quotidien qui ont rendu les sessions dissections bien plus sympathiques ! Un grand merci aussi à Nathan Martin, stagiaire de Master 1, pour sa contribution dans les dissections, mais aussi dans les analyses ICPMS de microchimie des otolithes. Enfin, je remercie l'ensemble du pôle de sclérochronologie d'Ifremer Boulogne-sur-Mer et plus particulièrement Romain Elleboode pour son aide précieuse dans les acquisitions de formes des otolithes. Merci également à Alaia Morell, stagiaire de Master 2 d'avoir participé aux acquisitions de formes des otolithes.

Une thèse « multitraceur » sous-entend l'utilisation de multiples méthodes faisant appel à des scientifiques aux compétences variées. Pour commencer, merci à Etienne Rivot et Hubert Du Pontavice d'Agrocampus pour leurs enrichissantes contributions sur le premier article de la thèse. Ensuite, merci à Bruno Ernande et Kélig Mahé d'Ifremer Boulogne-sur-Mer pour leur aide et leurs conseils dans le traitement des données de formes des otolithes. Merci à Bleuenn Gueguen de l'IUEM de Brest de m'avoir permis de réaliser les analyses de microchimie des otolithes sur sa superbe machine ICPMS (parfois capricieuse). La thèse fût également l'occasion de collaborer avec le laboratoire LBEG du KU Leuven en Belgique. Un grand merci à Filip Volckaert d'avoir permis cette collaboration et surtout merci d'avoir rendu possible la réalisation du volet « génétique » de cette thèse. Je remercie également Sophie Delerue-Ricard pour ses conseils et le partage de ses protocoles d'analyses qui m'ont bien aidé pour démarrer. Cette partie génétique n'aurait tout simplement pas pu se faire non plus sans l'aide de Gilles Lassalle et Eric Petit de l'INRA. Gilles, je te suis infiniment reconnaissante pour ton aide en bio-informatique. Tu as été extrêmement patient et bienveillant avec moi. Tu as aussi été un réel soutien et je dois dire que ton humour et tes blagues ont rendu la bio-info bien plus rigolote ! Éric, merci pour ta disponibilité et tes nombreux conseils sans lesquels je n'aurais pas pu réaliser ce volet de génétique des populations.

Vient ensuite le laboratoire d'écologie halieutique. Merci à tous les permanents, postdocs, thésards et stagiaires sans qui cette ambiance conviviale et relax n'aurait pas sa place. Que serait le labo d’halieutique sans ses mythiques apéros du vendredi soir ?! Je remercie Catherine, Sophie et Mathilde, nos supers secrétaires sans qui nous n’irions pas bien loin. Merci à tous les titulaires du labo, Olivier, Didier, Hervé, Elodie, Etienne, Jérôme, Thomas, Marie, Catherine Laidin et Jean-Eude. Merci aux thésards Maxime, Hubert, Pierre-Yves, Erwan et Louise pour la bonne ambiance impulsée par vous dans le labo. Merci aussi aux postdocs Jean-Baptiste, Auriane, Lucille et Jennifer, aux nombreux stagiaires et CDD ayant fait un plus ou moins long séjour au labo. Merci aux copains de la CET pour avoir contribué à la bonne ambiance. La team « footing du midi », composée principalement de Jérôme, Olivier, Hubert, Pierre-Yves et Thomas, a également contribué à ma santé mentale et physique au cours de cette thèse. Merci aussi à Olivier, Hubert, Thomas et toute la bande « coquilles Saint-Jacques » de m’avoir initié à la différentiation des coquilles et des huitres. Je ne peux pas me permettre d’écrire des remerciements personnalisés à chacun d’entre vous faute de temps et de place, mais sachez que j’ai été heureuse de faire partie de la grande famille du labo EH et que vous allez tous me manquer.

Un projet de thèse ne se résume pas à une expérience professionnelle. Une thèse c’est aussi une aventure humaine qui se vit entourée de nos proches, familles et amis, qui nous soutiennent dans toutes les étapes à gravir. Merci à tous les copains de Rennes et d’ailleurs pour les encouragements et pour tous les bons moments partagés. Je remercie évidemment ma famille pour leurs nombreux encouragements et le soutien depuis toutes ces années. Enfin, et surtout, je remercie du fond du cœur David pour tout le bonheur que tu m’as apporté au cours de cette dernière année de thèse et pour ton immense soutien au cours de ces derniers mois intenses.

Résumé

La compréhension de la dynamique des populations et de leur distribution spatiale est particulièrement importante pour assurer une exploitation durable des ressources halieutiques. L'amélioration des connaissances sur la structure spatiale et la connectivité des populations est essentielle à l'évaluation non biaisée de ces populations exploitées par la pêche. En cas de discordance entre la définition des populations biologiques et les unités d'évaluation et de gestion des stocks, le risque de surexploitation s'accroit.

La sole commune (*Solea solea*, Linnaeus, 1758) de Manche Est (division VIId, Conseil International pour l'Exploration de la Mer), est un stock de poisson plat d'intérêt économique, surexploité au cours des dernières décennies. Cette espèce se reproduit au printemps au niveau de frayères hauturières. Suite à l'éclosion, les larves dérivent vers des zones de nourriceries côtières et estuariennes peu profondes. Les juvéniles sont cantonnés dans ces nourriceries durant environ deux ans avant de rejoindre les individus adultes pour se reproduire à leur tour. La connectivité induite par les larves est très faible au sein du stock de Manche Est; elles sont très majoritairement transportées vers les nourriceries les plus proches. Par la suite, les juvéniles sont sédentaires dans les nourriceries et ne participent pas à l'homogénéisation du stock. Enfin, malgré l'amplitude de distribution de la phase adulte, son rôle potentiel dans la structure et de la connectivité des populations à l'échelle du stock restait jusqu'alors peu renseignée. Dans ce contexte, la présence de barrières naturelles (i.e. plateaux rocheux) et la faible connectivité induite par les jeunes stades questionnent la possibilité d'une structuration spatiale de la population.

Cette thèse a donc eu pour objectif d'étudier la structure spatiale et la connectivité au sein de la population de sole commune de Manche Est ainsi que son alignement avec l'unité de stock servant à l'évaluation et la gestion, en se limitant aux stades préadulte (i.e. juvéniles quittant les nourriceries avant la première reproduction) et adulte. Pour ce faire, la structure spatiale de la population a été étudiée par une approche holistique combinant des analyses de traceurs à l'échelle populationnelle et individuelle. Cette approche consistait, à partir d'un large éventail de traceurs, à combiner des informations portant sur la structure du stock à différentes échelles spatiales et temporelles.

L'approche populationnelle a recherché des différences spatiales de croissance populationnelle et d'abondances aux âges à partir de séries temporelles de données de longueur aux âges issues de campagnes scientifiques. Il a été mis en évidence des variations spatiales des paramètres de croissance

entre trois sous-unités au sein du stock. L'analyse de la synchronie des séries d'abondance par cohorte a quant à elle souligné l'isolement de l'une de ces sous-unités par rapport au reste du stock. Ainsi, l'approche populationnelle a permis de montrer l'existence d'un signal à long terme de structure spatiale du stock.

L'approche individuelle a consisté en une combinaison d'analyses génétiques, de forme et de microchimie des otolithes. Les résultats de génétiques et de formes des otolithes ont convergé vers l'hypothèse d'une structure spatiale en trois sous-unités. Cependant, l'analyse de microchimie des otolithes n'a pas permis de conclure quant à la réassignation des individus adultes à leur nourricerie d'origine.

Les informations sur la structure spatiale de la population issue des approches populationnelles et individuelles ont été combinées aux résultats d'une étude de marquage-recapture conduite précédemment sur la sole commune de Manche Est afin de calculer un indice de différentiation de stock. Cet indice semi-quantitatif permet d'évaluer le niveau de structure interne au sein du stock par l'intégration de traceurs de résolutions spatiales et temporelles différentes. Cet indice a mis en évidence une structure spatiale forte et pérenne à l'intérieur du stock de sole commune de Manche Est.

En conclusion, la définition actuelle du stock de sole commune en Manche Est apparaît inadaptée à la réalité biologique de la population sous-jacente. L'approche holistique a démontré une structure en métapopulation composée de trois sous-unités, en discordance avec l'unité de stock actuelle servant à l'évaluation et à la gestion. Des stratégies alternatives d'évaluation et/ou de gestion devraient garantir une meilleure estimation du statut du stock, pour une exploitation durable de l'espèce sur ce secteur.

Abstract

In fisheries science, the understanding of population dynamics and spatial distributions is of great importance to ensure the sustainable exploitation of resources. Improving the knowledge of spatial structure and connectivity of marine populations is essential to provide unbiased assessment and management measures. Higher risks of declines of exploited populations result from the misalignment between biological populations and stock units.

The common sole (*Solea solea*, Linnaeus, 1758) of the Eastern English Channel (division VIId, International Council for the Exploration of the Sea) is a flatfish stock that has been overexploited over the last decades. This species reproduces on distinct spawning grounds in deep waters. After hatching, larvae drift towards shallow coastal and estuarine nurseries. Juveniles settle for about two years in these areas before they join adult feeding and spawning grounds, in deeper waters offshore. Larval-mediated connectivity is very low inside the stock since larvae are mainly advected towards the nearest nursery grounds. Then, juveniles are highly sedentary in the nursery grounds, inducing low mixing within the stock. Despite the potential role of sub-adult and adult stages in population structure and connectivity, their movements and mixing remain poorly shown. Though, natural barriers (i.e. rocky reefs) and the low connectivity induced by early life stages suggest potential spatial structure inside the stock.

This thesis aimed at investigating the common sole population structure and its alignment with the Eastern English Channel stock unit by focusing on subadult (i.e. juveniles leaving their nurseries before the first reproduction) and adult stages. To do so, the population spatial structure was studied through a holistic approach combining analyses of tracers at the population scale with analyses of tracers at the individual scale. More precisely, this approach consisted in combining information on the stock structure at different spatial and temporal scales using a range of natural or artificial tracers and assessing their convergence.

The population-based approach consisted in testing for spatial differences in size and abundance-at-age using long-term length-at-age time series retrieved from a scientific survey. Results highlighted spatial variation of growth parameters between three subunits inside the stock. Synchrony analysis of abundance-at-age throughout the cohorts highlighted isolation of one of these subunits. Thus, the population-based approach demonstrated a long-lasting signal of spatial structure.

The individual-based approach focused on a combination of genetic, and otolith shape and microchemistry analyses. Results of the genetic and otolith shape analyses were consistent and highlighted spatial structure in three subunits. However, otolith microchemistry analysis failed in assigning adult individuals to their nurseries of origin.

Information of spatial structure of the population and individual-based approaches were finally combined with the results of a previous mark-recapture study of the common sole of the Eastern English Channel, and integrated for the calculation of a stock differentiation index. This semi-quantitative index allowed for synthesizing information from tracers having different spatiotemporal scales, to assess the strength of stock spatial structure. This index demonstrated evidence of strong spatial structure in three subunits across the stock of sole of the Eastern English Channel.

Therefore, the current definition of the stock of the Eastern English Channel is not adapted to the underlying population structure of common sole. The holistic approach highlighted a metapopulation structure formed of three subunits, misaligned with the stock assessment and management unit. Alternative assessment and/or management strategies would help in improving the assessment of the stock status, to ensure the sustainable exploitation of this species.

Chapter 1

General introduction

General introduction

In the context of global change and increasing degradation of aquatic habitats worldwide, it is crucial to improve our understanding of ecological and biological processes shaping population and ecosystem functioning to ensure sustainable exploitation of resources. Overexploitation of aquatic species contributes to fish population declines or collapses (Le Pape *et al.*, 2017). However, expansion of human populations will increase future needs for fish products since fish are a key source of proteins worldwide (Merino *et al.*, 2012).

Upstream of the assessment-management process, marine fish stock assessment is based on hypotheses of spatial distribution of resources. Scientists assess fish populations using predefined areas, the stock units. Hypotheses on stock delineations are the basis of whichever assessment model. But simplification of, and/or misalignment on, population functioning and distribution can bias the assessment and lead to overexploitation (Tuck and Possingham, 1993; Fu and Fanning, 2004; Ying *et al.*, 2011). Hence, improving knowledge of population spatial distribution is crucial to provide reliable assessment, for sustainable exploitation of resources (Kerr *et al.*, 2017a; Cadrin, 2020).

This thesis focused on the common sole (*Solea solea*, Linnaeus, 1758), a flatfish species overharvested over the last decades in the Eastern English Channel. Uncertainties remain regarding the spatial structure of the stock and its alignment with the underlying population. The present thesis aimed at filling these gaps by combining different methods in a holistic approach to assess population structure. In the present introduction, population, metapopulation and stock concepts are first defined and the holistic approach is presented. Then, a focus is done on the case study. Finally the objectives of the thesis are exposed.

1.1. Alignment between marine populations and stock units

1.1.1. Population, metapopulation and connectivity in ecology

In ecology, population is a central concept and numerous definitions have been proposed, depending on the field of studies. Waples and Gaggiotti (2006) reviewed population definitions according to ecological, evolutionary and statistical paradigms. Following the ecological definition, a population could be viewed as a group of organisms interacting in a single patch (Holt, 1997). Regarding the evolutionary paradigm, a population is a group of interbreeding individuals coexisting in space and time. Consequently, a general definition of population could be a group of interbreeding individuals displaying spatiotemporal interactions. Populations compose metapopulations which altogether constitute a species (**Fig. 1.1**).

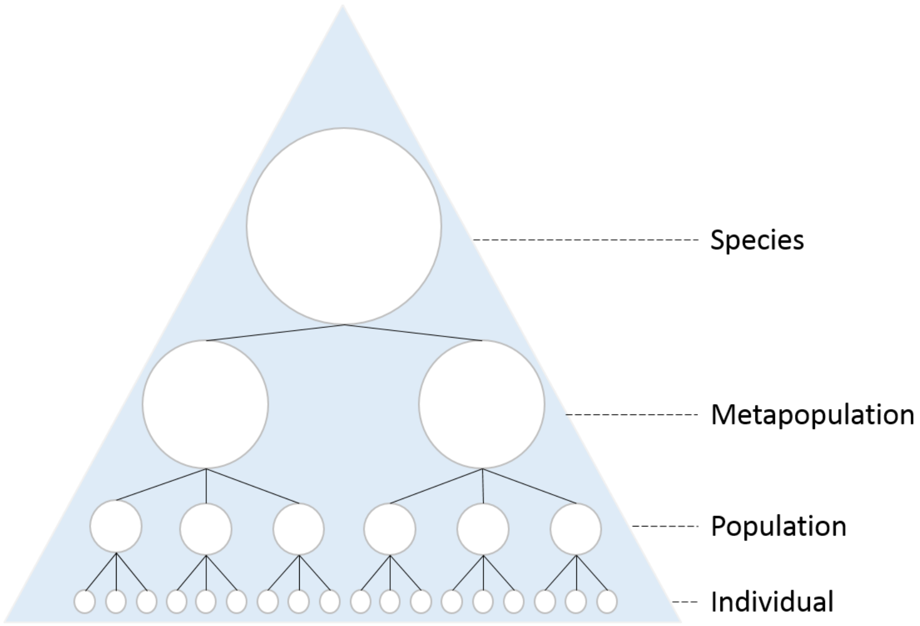

Figure 1.1 Levels of biological organization from individuals to species. The pyramid is a simplification of ecological levels and does not represent intermediate levels such as cohorts between the individual and population levels or subspecies between the metapopulation and species levels.

The concept of metapopulation has first been introduced by Levins (1969, 1970) who considered a metapopulation as a population of populations meeting three conditions: (1) populations have the same size and the strength of connections are equivalent, (2) population dynamics are asynchronous and (3) exchange rates between populations are too low to modify local dynamics. Various definitions have been proposed for metapopulation (Kritzer and Sale, 2004) considering that all these criteria are rarely fulfilled (Harrison and Taylor, 1997). Smedbol *et al.* (2002) and Grimm *et al.* (2003) defined the metapopulation as a set of populations connected by dispersal and characterized by extinction-recolonization events. Other authors considered a simpler definition with patchily discrete habitats linked by dispersal of intermediate intensity, allowing the independence, and sometimes the asynchrony, of local population dynamics (Hanski and Simberloff, 1997; Hanski, 1999; Hanski and

Gaggiotti, 2004; Kritzer and Sale, 2004). Because of the increasing magnitude of habitat degradation and fragmentation worldwide, the metapopulation concept has been widely used to investigate population dynamics (Hanski, 1999) and the management and conservation of resources (Southwell *et al.*, 2016). This concept has been broadly applied to terrestrial species (e.g. Hanski and Thomas, 1994; Esler, 2000; Hanski and Ovaskainen, 2000; Inchausti and Weimerskirch, 2002; Smith and Green, 2005; Heard *et al.*, 2013) and to a lesser extent to aquatic species, since the 1990s (Grimm *et al.*, 2003). The main barriers to this concept for aquatic species result from the movements of marine resources and the difficulty of observing and delineating populations: 'Counting fish is just like counting trees except that they are invisible and keep moving' (John Shepherd).

Closely linked to the metapopulation concept is connectivity. From an ecological perspective, connectivity could be defined as the strength of individual exchanges between populations across the distribution range of the species (Palumbi, 2003). Understanding the connectivity that links populations is particularly challenging and has been studied extensively over the past two decades, so that the 2000s was called 'the decade of connectivity' (Hixon, 2011). Connectivity can impact a large number of functions and processes at the scale of populations or metapopulations such as the flow of materials and energy and evolutionary divergence (Boström *et al.*, 2011; D'Aloia *et al.*, 2015; Bryan-Brown *et al.*, 2017). Connectivity shapes species distribution, movements of individuals and biology as well (Parrish, 1989). Investigating connectivity is also important for the recovery of populations, assemblages and ecosystems from perturbations (Bernhardt and Leslie, 2013) and is consequently considered in conservation and management of marine resources (Magris *et al.*, 2014; Beger *et al.*, 2015). Degrees of connectivity can vary along a continuum of population structure, from a single and homogeneous population (i.e. complete panmixia) to isolated populations (Waples and Gaggiotti, 2006; Cowen and Sponaugle, 2009; Ciannelli *et al.*, 2013). Intermediate situations of population or metapopulation structure between these two extreme cases are the most common (**Fig. 1.2**).

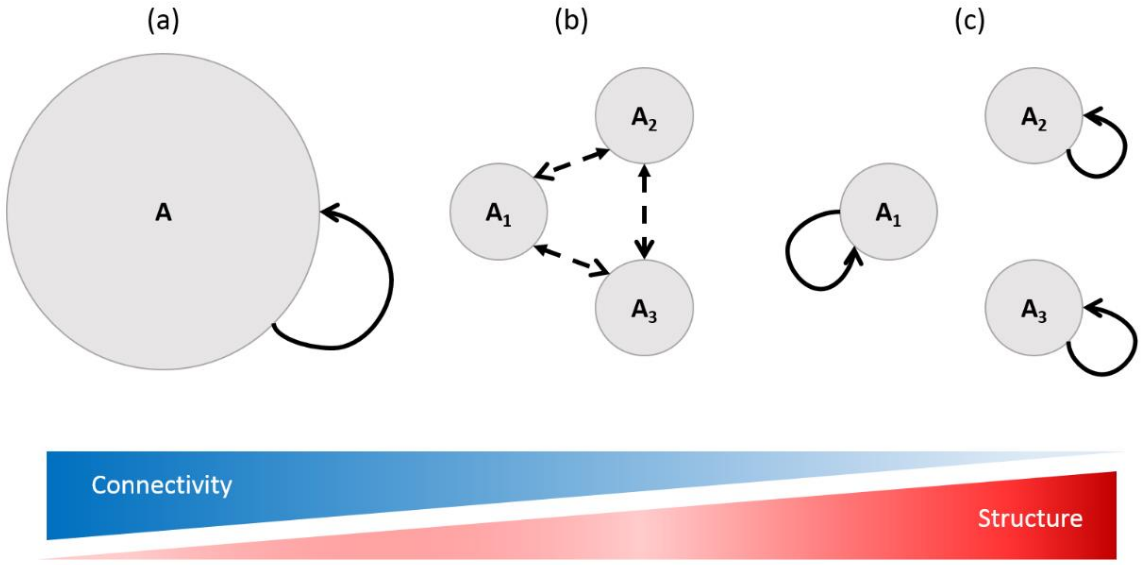

Figure 1.2 Continuum of connectivity and spatial structure. Circles represent the population or subpopulations and black arrows are the connection between populations. Dashed arrows mimic low connections whereas solid arrows represent strong connections. (a) Single homogeneous and well-mixed population. (b) Metapopulation composed of three subpopulations connected by dispersal. (c) Isolated subpopulations.

In the marine realm, the direction and intensity of connections are driven by geological history, topography, oceanography and spatial organization of ecosystems (Cowen *et al.*, 2007). Besides, the biophysical processes generating early stages (i.e. eggs and larvae) dispersal patterns and post-larval (i.e. juveniles, sub-adults and adults) movements through straying and homing behaviors, site fidelity (i.e. spawning and nursery grounds) or migration-related strategies are involved in connectivity (Secor, 2015). A combination of active (e.g. swimming, delay in the settlement, individual, and group behaviors) and passive processes (e.g. tides, wind, waves, currents, hydrodynamic processes, and stratification) are involved in larval dispersal (Cowen and Sponaugle, 2009). For instance, larvae can modify their vertical distribution by active swimming or change in buoyancy to reach different marine currents (Marchand and Masson, 1989; Lacroix *et al.*, 2013). They can also use chemical properties of freshwater from terrestrial inputs to orient themselves at low spatial scale (i.e. 10 km radius; Marchand, 1991). At subadult and adult stages, passive processes become less important compared to active behaviors (Secor, 2015). Fish can choose when, where and how to migrate according to species life cycle. For instance, natal homing is a specific behavior that consists of a within-generation return to the place of birth to reproduce (Secor, 2015). When the migration behavior occurs over multiple generations, the population becomes philopatric (Secor, 2015). Larval dispersal has been considered for a long time as the main driver shaping the connectivity in marine species (Cowen and Sponaugle, 2009). However, this paradigm was recently reevaluated and the importance of adult-mediated connectivity was highlighted (Mullon *et al.*, 2002; Frisk *et al.*, 2014). Hence, population connectivity needs to be investigated across the entire life cycle of species (Secor, 2015).

1.1.2. Stock unit in fisheries science

From a fishery perspective, the stock unit is basically considered as the spatial unit allowing the assessment and management of exploited fish species. Early definitions of stock appeared at the beginning of the twentieth century and was purely operational, referring to the portion of fish exposed to the fishery, i.e. “the harvest stock” (Waldman, 2005). Later, the Stock Concept International Symposium (1980, Ontario, Canada) debated and finally stated on a definition of stock proposed by Ihssen *et al.* (1981) who considered the stock as “an intraspecific group of randomly mating individuals with temporal or spatial integrity”. Then, a less restrictive definition was proposed by Hilborn and Walters (1992) who qualified the stock as arbitrary self-reproducing groups of individuals with similar life history traits within each group. The International Council for the Exploration of the Sea (ICES), an intergovernmental organization dedicated to marine science, defined the stock as “a part of a fish population usually with a particular migration pattern, specific spawning grounds, and subject to a distinct fishery; in theory, a unit stock comprises all the individuals of fish in the area, which are part of the same reproductive process”. Hence, defining a stock is a fuzzy art (Cadrin, 2020) and definitions will continue to change with technological advances allowing the delineation of stock units (Waldman, 2005). As suggested by Waldman (2005), stock definition according to Ihssen *et al.* (1981) can be considered accurate in fisheries ecology and is hereafter considered in this thesis.

1.1.3. Matching the population and stock definitions

In ecology and fisheries, defining a species, subspecies, metapopulation, population or stock is relative to the issue and the available tools allowing the question of delineation to be answered (Begg and Waldman, 1999; Waldman, 2005). Considering the previous definitions of population and stock, one may wonder why the stock unit would not directly correspond to the underlying population. The ICES considered the population as a “group of fish of one species which shares common ecological and genetic features; the stocks defined for the purposes of stock assessment and management do not necessarily coincide with self-contained populations”. Distinction between stock and population remains confusing. Secor (1999) proposed that the stock is at the intersection between the extent of population distribution and human activities (i.e. fishery) that impact population productivity. Therefore, the relevant question is ***does the stock delineation considered for assessment and management concerns is aligned with the underlying biological population*****?**

Stock delineation is a strong hypothesis introduced in stock assessment models and is a prerequisite to management measures (Kutkuhn, 1981; Smith *et al.*, 1990; Begg *et al.*, 1999a). However, marine populations are commonly structured in space and time (e.g. metapopulation) and habitat occupancy can vary along the life cycle (Waples and Gaggiotti, 2006; Reiss *et al.*, 2009; Ames and Lichter, 2013; Ciannelli *et al.*, 2013), which should be accounted for in stock assessments (Carson *et al.*, 2011; Petitgas *et al.*, 2013; Frisk *et al.*, 2014). For instance, individuals can share the same feeding grounds but reproduce in distinct spawning areas (or at different periods) so that reproductively isolated populations should be assessed and managed separately. The understanding of population structure and dynamics is crucial for marine resources especially because they experience many pressures among which habitat degradation, fishing exploitation and climate change (Cheung *et al.*, 2009). Some authors have shown the importance of considering spatial population structure in the optimization of the exploitation (see the optimal harvesting theory; Hilborn and Walters, 1992; Tuck and Possingham, 1993) and the adaptation of the exploitation to local productivities (Taylor *et al.*, 2011; Ying *et al.*, 2011). Whereas ignoring population structure can bias the estimates of life history (Punt, 2019), the current practice considers homogeneous vital rates when the understanding of stock structure is limited (Cadrin *et al.*, 2013; Kerr *et al.*, 2017). Consequently, mismatch between the stock and the underlying biological population arises (**Fig. 1.3**) and can induce overexploitation of less productive subunits (Tuck and Possingham, 1993; Fu and Fanning, 2004; Cadrin and Secor, 2009; Ying *et al.*, 2011; Goethel and Berger, 2017). Sometimes, such mismatch can lead to stock collapses (Hilborn *et al.*, 2003; Kritzer and Sale, 2004; Neat *et al.*, 2014). A large range of match or mismatch situations can be encountered in practice. In some cases, populations are well mixed and present homogeneous dynamics inside the stock unit so that the alignment between population and stock unit is perfect. However, a well-mixed population can be separated in distinct stock units for assessment and management, resulting in misalignment of population and stocks (**Fig. 1.3.a**). More complex situations are encountered when subpopulations have different internal dynamics with various vital rates. Mismatch in stock unit results from ignoring these different situations of spatial structure (**Fig. 1.3.b**).

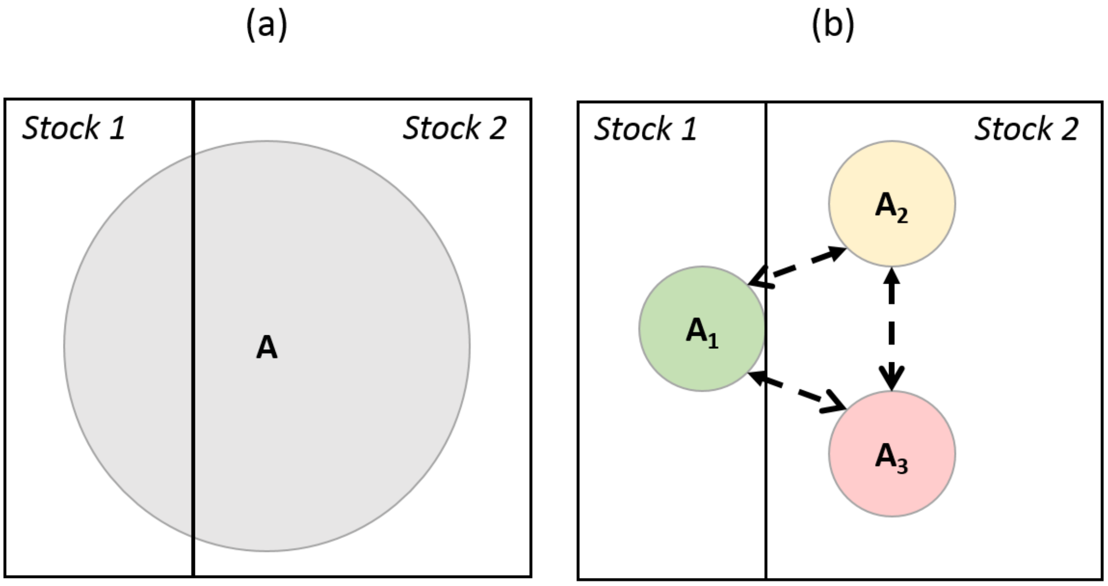

Figure 1.3. Theoretical examples of misalignment between population or metapopulation and stock units. Circles represent populations or subpopulations and squares are stock unit boundaries. Various colors in circles indicate contrasted internal dynamics and productivities. Dashed arrows mimic low connections between subpopulations. (a) Misalignment between biological population A and stocks 1 and 2. (b) Misalignment between low connected subpopulations (A_1, A_2, A_3) and stocks 1 and 2.

ICES reported that in early 2000s, around 50 out of about 150 stocks were misaligned with underlying populations (Stephenson, 2002). Originally, management units were convenient political boundaries rather than aligned with biological characteristics (Bosley *et al.*, 2019), although political delimitations are obviously not physical barriers for marine fish. Fortunately, a large range of methods exist to assess population structure and connectivity and solve the issue of the alignment between stock unit and population.

1.2. A holistic approach to assess population structure and connectivity

Fish stock identification is an interdisciplinary field in fisheries science that aims at discriminating among stock units using a large diversity of methods (Cadrin *et al.*, 2013). In the early 1990s, ICES initiated a working group on “stock identification protocols for finfish and shellfish stocks” which is now the "stock identification methods working group". This working group assesses the advances and current practices to evaluate stock identity. A process was detailed by Cadrin *et al.* (2014) in order to evaluate population structure and connectivity and inform the alignment between population and stock unit:

(1) Defining the current spatial assessment and management units and understanding the reasons for such delineation.
(2) Defining alternative hypotheses of spatial structure.
(3) Reviewing all methods and their spatiotemporal resolution to inform the stock structure and connectivity.
(4) Evaluating congruencies and discrepancies between methods and synthesizing information in an interdisciplinary (i.e. multi-tracer holistic) approach.
(5) Testing for *a priori* hypotheses of spatial structure and drawing conclusions.

At the end of this process, potential alternative assessment and management strategies have to be considered in case of misalignment between populations and stock units. Finally, practical limitations of those strategies have to be identified and biological, economic and social consequences need to be evaluated (Kerr *et al.*, 2017).

Over the past two decades, artificial (e.g. external or internal tags) and natural tracers (e.g. life history traits, genetic markers, geochemical tracers, stable isotopes, fatty acids, parasites and meristics) have been increasingly used to test for signals of population spatial structure and connectivity between populations or habitats (Cadrin *et al.*, 2013; Tanner *et al.*, 2016). A tracer is a piece of information located on or into the fish the *a posteriori* reconstruction of the environments experienced by an individual during its life. Depending on the spatial and temporal resolutions of the tracers, population structure and connectivity can be addressed at the population level, so that tracers are part of population-based approaches, or at the scale of individuals, also called individual-based approaches.

For the purpose of this thesis, hereafter are described the population and individual-based approaches used to address structure and connectivity of the common sole (*Solae solea*) population inside the Eastern English Channel stock. Other methods exist and are deeply described in Cadrin *et al.* (2013) but will not be developed here.

This thesis focuses exclusively on sub-adult and adult stages since the connectivity induced by larvae and juveniles was investigated previously (Coggan and Dando, 1988; Riou *et al.*, 2001; Rochette *et al.*, 2012; Le Pape and Cognez, 2016). The tracers developed hereafter are thus adapted to sub-adults and adults.

1.2.1. Population-based approaches

Population-based approaches aim at estimating differences of phenotypic and/or genetic characteristics at the population scale in order to highlight spatial structure. In this thesis, population-based approaches focus on abundances and growth, even if other life history parameters might be used when available (e.g. maturity, Cadrin *et al.*, 2013).

Available from routine surveys, abundance and growth parameters are underused in practice to discriminate among populations (Begg *et al.*, 1999b; Begg, 2005; Pita *et al.*, 2016). Hence, long-term time series are often available at no cost from these surveys (Begg and Waldman, 1999; Cope and Punt, 2009). It is advised to begin with the investigation of population distribution and abundance to provide an overview of the stock structure before analyzing spatial segregation at finer spatial and temporal scales (Begg *et al.*, 1999b; Begg, 2005).

- **Abundance**

Following the definition of Ihssen *et al.* (1981), individuals mate randomly and display spatiotemporal group integrity across the stock unit. Thus, the analysis of correlations in temporal fluctuations of demographic attributes (e.g. spatial synchrony; Walter *et al.*, 2017) is a valuable method to investigate the internal spatiotemporal structure of populations/stocks (Botsford and Paulsen, 2000; Rushing *et al.*, 2016; Rogers *et al.*, 2017). Synchrony among populations is mainly driven by dispersal and Moran effects (i.e. effect of correlated fluctuations in environmental drivers on synchrony among populations) (Ranta *et al.*, 1995; Liebhold *et al.*, 2004). Then, asynchrony in abundance time series across the stock would reveal stock structure (Begg *et al.*, 1999b; Begg, 2005) and call the stock integrity into question. More precisely, spatial asynchrony (i.e. lack of correlations) in abundance time series would indicate that the stock is spatially structured. Persistence in spatial structuring over the time series would indicate a long-term stock structure and should be considered in stock assessment and management (**Fig. 1.4**).

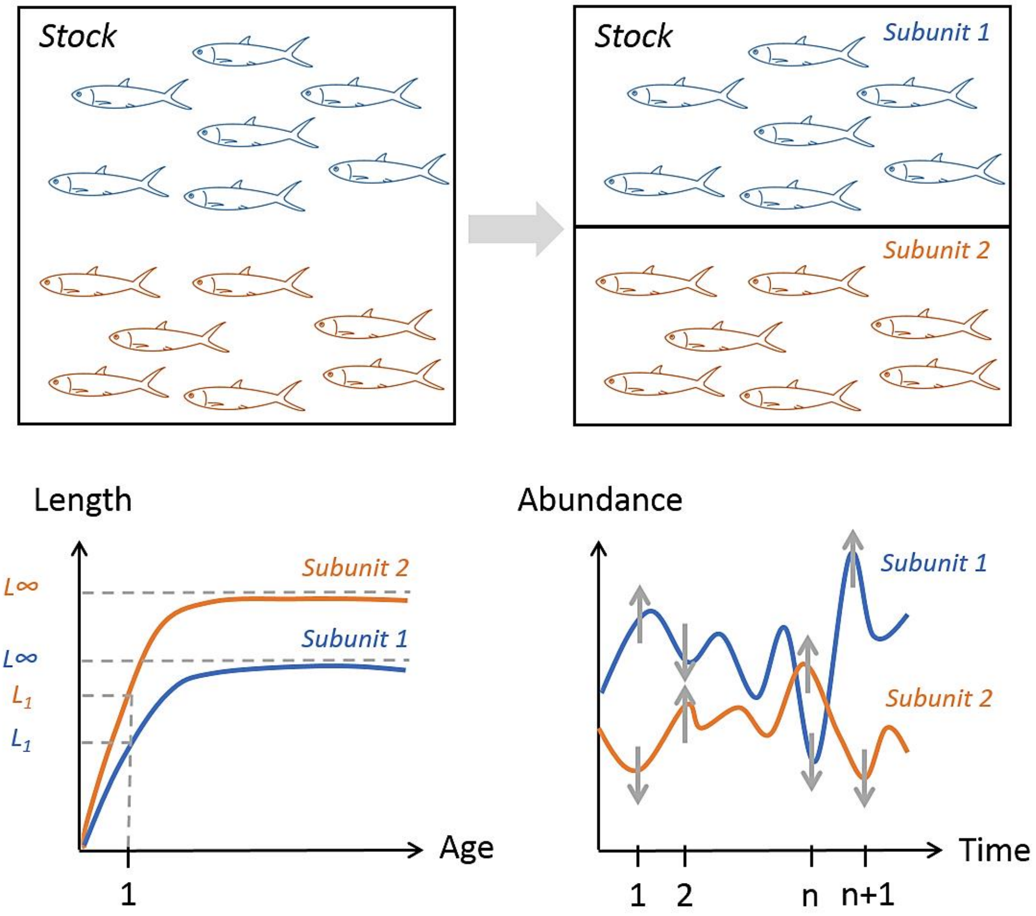

Figure 1.4. Spatial differences in von Bertalanffy growth parameters (L_∞ and L_1) and asynchrony among abundance throughout a cohort in the time series. These variations indicate that the stock (represented by a black square) is spatially structured in subunits 1 and 2 according to the growth and abundance analyses. Grey arrows on abundance curves underline that abundances vary in different directions along the time series (i.e. asynchrony).

- **Growth**

Growth characteristics are life history traits which can be viewed as the consequences of life history strategies. They are phenotypic expressions of the interaction between genotype and environment and reflect the underlying population dynamic (Begg, 2005). They are also sensitive to changes in fishing pressure and environmental conditions, so that life history traits need to be examined over consistent time frames through long-term survey data (Begg *et al.*, 1999b; Begg, 2005). Based on length-at-age data set, comparison of growth parameters is one of the most commonly used methods in life history-based stock structure analyses (**Fig. 1.4**). In particular, the von Bertalanffy growth model is very popular in fisheries science and allows for back calculate the age of fish relatively to the length (von Bertalanffy, 1957). Other growth models such as Gompertz, Schnute, Richards or logistic can be applied, depending on the species of interest. Then, spatial differences in growth parameters indicate that populations are geographically and/or reproductively isolated and consequently can be considered as distinct stock units for management purposes.

Population-based approaches provide baseline information on stock structure over the years. However, life history parameters or demographic attributes do neither allow to assign individuals to their stock of origin, nor to provide evidence of genetic isolation (Begg and Waldman, 1999). To fulfil these limitations, individuals-based approaches are particularly recommended in addition to population-based approaches.

1.2.2. Individual-based approaches

Individual-based approaches aim at estimating differences of phenotypic and/or genetic characteristics at the individual scale in order to highlight spatial structure or connectivity. In this thesis, individual-based approaches focus on otolith microchemistry and shape, genetic markers and external tagging.

- **Chemistry and shape of fish otoliths**

The otoliths of teleost fishes, also called ‘ear stones’, are calcified structures located in the inner ear and formed lifelong by crystallization of chemical elements extracted from the surrounding water. Three pairs of otoliths, the lapilli, asterisci and sagitta, are involved in hearing and balance (**Fig. 1.5**). During the egg and larval endotrophic stages, the central part of otoliths (i.e. the nucleus) forms and incorporates maternal material provided by the yolk (Campana, 1999; Campana *et al.*, 2000). Then, since gill-mediated respiration takes place and as the fish grows, otoliths grow incrementally as well, incorporating dissolved trace elements bio-available in the ambient water (Campana, 2001). Those trace elements are incorporated into the otoliths after being transported via the gills (or to a lesser extent via the intestine), blood plasma and finally endolymph (Campana, 1999). The otoliths are mostly composed of calcium carbonate and proteins (Campana, 1999).

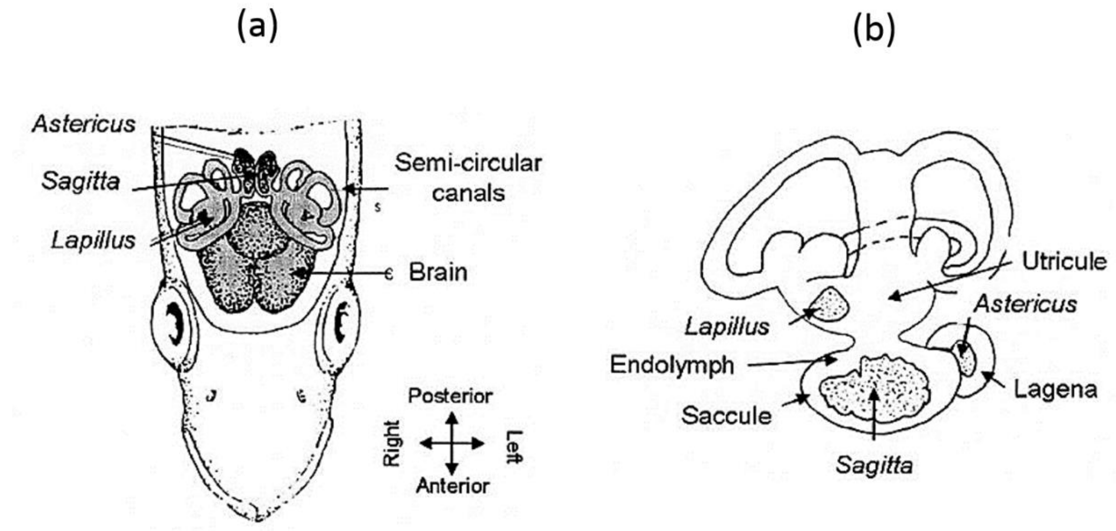

Figure 1.5 (a) Dorsal view of a teleost fish with the top of the cranium cut and apparent brain and inner ear. (b) Three pairs of otoliths located in the labyrinth system. Retrieved from Payan *et al.* (2004).

The otoliths are considered a metabolically inert timekeeper which grows by daily deposition of material without resorption during the entire life of the fish. Since otoliths embed trace elements of the surrounding environment, their composition mostly reflects the ambient water (Walther and Thorrold, 2006). Depending on the chemical elements, concentrations in water and otoliths could be equal or uptake could be partitioned (Bath *et al.*, 2000). In case of chemically distinct water masses among habitats, it is possible to discriminate habitats from otolith composition and retrace *a posteriori* their occupancy during the fish life. These properties make the otolith the 'black box' of fish. Coupling microchemistry and micro-increment analyses is particularly useful to reconstruct *a posteriori* the habitat use of the fish from birth (i.e. the core of the otolith) to death (i.e. the edge of the otolith; **Fig. 1.6**) (e.g. Thorrold *et al.*, 2001; Reis-Santos *et al.*, 2013; Tanner *et al.*, 2013; Randon *et al.*, 2017).

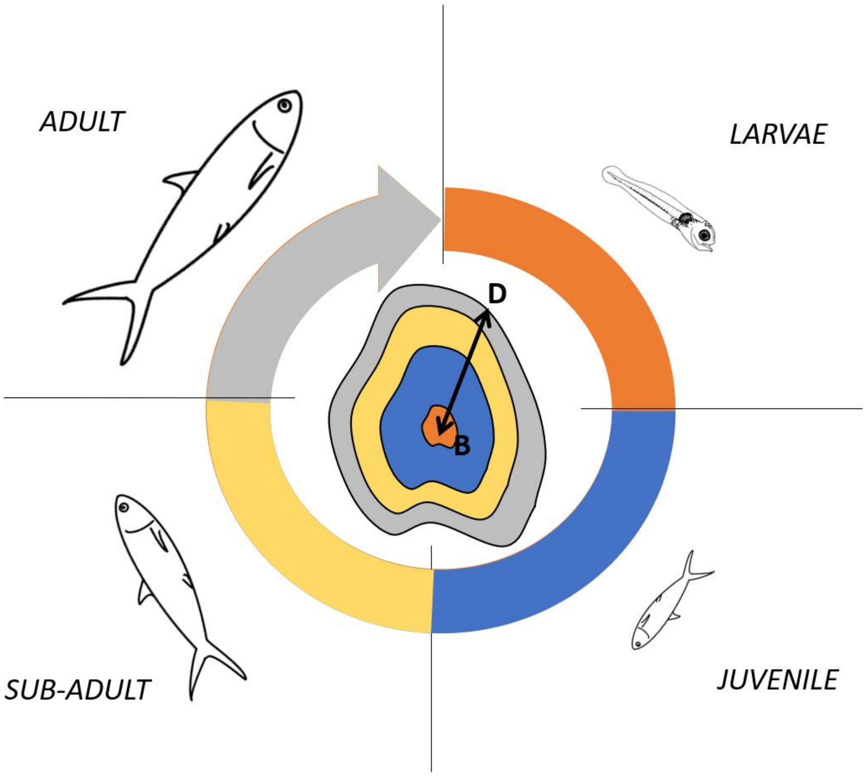

Figure 1.6. Integration of habitat chemical signatures in the otolith along the fish life cycle, from birth (B) to death (D).

Over the past decades, otoliths have been widely used to investigate fish age and growth rates (Campana, 2001), and to determine migration pathways and environmental history (Elsdon and Gillanders, 2002; Walther and Limburg, 2012).

In addition, environmental and genetic factors shape the otolith morphology (Cardinale *et al.*, 2004; Vignon, 2015). Spatial differences in otolith shape inside a stock would indicate stock spatial structure (**Fig. 1.7**).

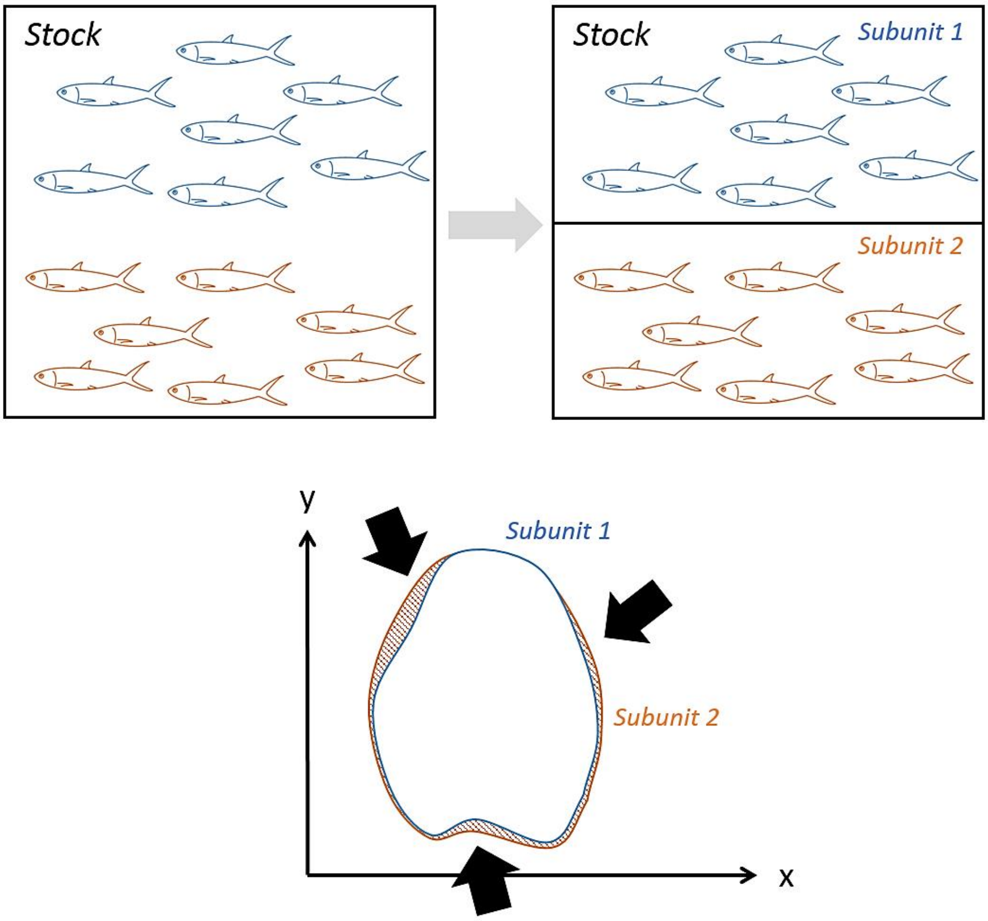

Figure 1.7. Schematic representation of (possible) otolith shape variations when the stock (represented by a black square) is spatially structured in two distinct subunits. Black arrows highlight the main shape variations between otoliths of subunits 1 and 2.

The well-known elliptical Fourier descriptors are among the most powerful methods based on otolith shape allowing to discriminate among fish populations (e.g. Hüssy *et al.*, 2016; Mahe *et al.*, 2016; Zhang *et al.*, 2016; Vasconcelos *et al.*, 2018). This approach consists of extracting shape parameters from Fourier harmonics and investigating spatial differences in parameters. Other methods using simpler shape indices based on otolith width, length, perimeter and area can also be used to detect population structure (Tuset *et al.*, 2003; Delerue-Ricard *et al.*, 2018). Moreover, recent investigation in bilateral asymmetry of otoliths (i.e. proportion of non-overlapping between left and right otoliths) has been suggested as a potential tool to highlight stock structure, but was yet underused (Mahé *et al., in press*).

Therefore, otoliths are a kind of ‘Swiss knife’ which allows to investigate population structure and connectivity using growth, microchemistry and/or shape analyses.

- **Genetic markers**

Genetic markers have been widely used over the last decades, notably to discriminate populations and retrace genetic connectivity (Hauser and Carvalho, 2008). Recently, the traditional view of genetic homogeneity in the marine realm and the role of extensive larval dispersal in population differentiation was reevaluated. Genetic tools have demonstrated their efficiency in discriminating among populations at low spatial scales, from tens to a few hundred kilometers (Ruzzante *et al.*, 1998; Knutsen *et al.*, 2003; Ciannelli *et al.*, 2010). Microsatellites and Single Nucleotide Polymorphisms (SNPs) are high resolution markers among the most up-to-date applied methods (Glover *et al.*, 2008; Nielsen *et al.*, 2012). The microsatellites are deoxyribonucleic acid (DNA) sequences composed of 2 to 8 bases forming repetitive motifs (**Fig. 1.8**). SNPs are abundant and widespread changes in single nucleotides at loci situated in coding or non-coding regions of the genome (Vignal *et al.*, 2002) (**Fig. 1.8**).

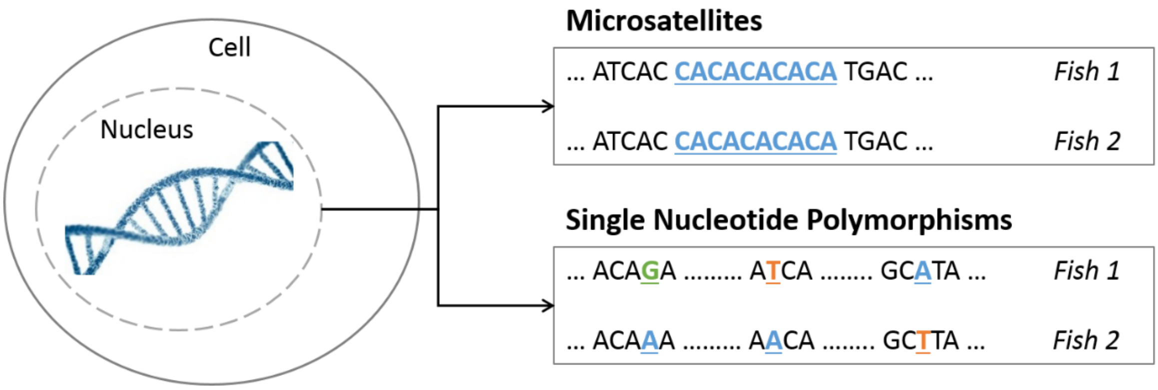

Figure. 1.8 Representations of microsatellites and Single Nucleotide Polymorphisms found in the nuclear DNA of a diploid species. Adapted from Morin *et al.* (2004).

Although microsatellites are highly polymorphic, some limitations emerged (e.g. homoplasy, null alleles, genotyping errors, low reproducibility between experiments, and low occurrence of motifs in some chromosomal regions) and resulted in the increasing success of SNPs in population genetic studies. Indeed, SNPs circumvent these limitations and advances in Next Generation Sequencing (NGS) allowed their improvements and spread, even for nonmodel species (Helyar *et al.*, 2011). Recent studies highlighted that a small fraction of SNPs can contain high information regarding population structure (Lao *et al.*, 2006; Paschou *et al.*, 2007), outperforming microsatellites (Liu *et al.*, 2005).

Various factors such as connectivity (e.g. migration levels between populations), population size and population history (e.g. secondary contact, founder event recolonization, range expansion) influence the level of population genetic differentiation. To measure genetic differentiation, a common practice is the estimate of F_{ST} (Weir and Cockerham, 1984) that increases with genetic differentiation.

• External tagging to track fish movements

Mark-recapture experiments are commonly used since the beginning of the twentieth century, to address population connectivity and migration routes (Jacobsen and Hansen, 2005). The principle of tagging experiments relies on the application of an internal or external tag (i.e. a mark) on the fish for the purpose of understanding individual movements across the distribution range of a species or at a finer spatial scale, within a stock or between habitats. An ideal mark allowing individuals to be precisely identified is permanent, inexpensive, easily applicable and without negative effects on behavior, survival and growth (Jacobsen and Hansen, 2005). Different kinds of marks exists and can be applied to fishes, depending on the species, the scientific question, the recovery method and the cost of the study. Among the most conventional tags, the well-known Peterson's disk consists of applying a little disk directly on the fish by piercing the muscle (**Fig. 1.9**). By comparing the release and recapture locations, one can retrace individual routes, and estimate connectivity. Such external mark has the advantage to be cheap, allowing for tagging many individuals.

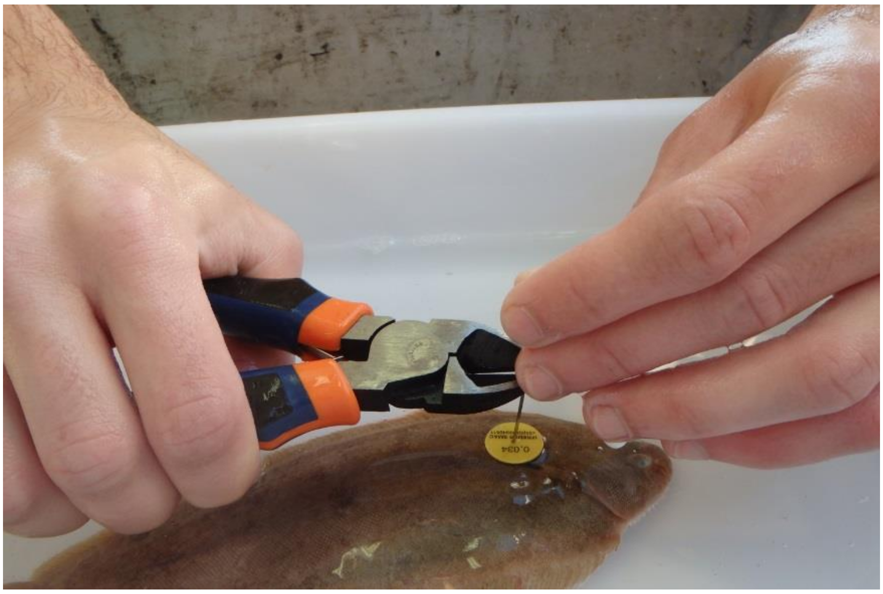

Figure 1.9. Positioning of a Peterson's disk on a common sole (*Solea solea*). (Picture ©Ifremer).

Electronic tags have the advantages to provide fishery independent locations and data archives allowing the decrease of the uncertainty of movements from mark to recapture locations (Sippel *et al.*, 2015). However, electronic tags are still relatively large/heavy and expensive, and are preferred for large fish species such as tuna or sharks (e.g. Hazen *et al.*, 2016; Hearn *et al.*, 2016). Therefore, the choice of a tag type is a trade-off between the quantity of information contained in the tags, the number of tags that can be applied (and then the spatial coverage of the study) and the probability to recover the tag. Tag-recapture data are considered as reliable to infer population structure (Goethel *et al.*, 2011).

1.2.3. Combining approaches and spatiotemporal scales

Recent advances in stock identification methods and movement ecology encourage the combined use of methods. To circumvent the respective limitations of each tracer, it is advised to combine different types of tracers in a holistic approach (Begg and Waldman, 1999; Waldman, 1999; Tanner *et al.*, 2016), i.e. a multi-tracer approach. Such holistic approach consists of reviewing all the stock identification information obtained from different methods to infer stock structure. The stock structure could be investigated in a single study using two or more methods on a range of samples; or preferably performing a wide range of methods on the same individuals (Begg and Waldman, 1999). Whereas the latter approach should be retained to resolve stock structure, it is rarely used in practice because of technical (e.g. combination of population and individual tracers) and financial limitations, and then, the use of a range of sample is frequent.

Different advantages arise from the use of a holistic approach. First, the use of a range of different methods often allows solving the discrepancies between methods, particularly for stocks demonstrating an apparently complex structure. The results from different methods (e.g. between genetics and meristics or other alternative methods) could be divergent (e.g. Leslie and Grant, 1990; Pepin and Carr, 1993). At the opposite, congruence between methods would allow the increase of confidence with regards to stock structure. Results from different stock identification methods must be compared and combined (e.g. Abaunza *et al.*, 2008; Tanner *et al.*, 2014; Reis-Santos *et al.*, 2015, 2018; Izzo *et al.*, 2017; Ulrich *et al.*, 2017) and a precautionary management strategy is advised when a lack of congruence between methods is found.

Before conducting an interdisciplinary approach, the expected results from each tracer and their respective spatiotemporal resolution must be considered. Indeed, some tracers could provide redundant information about the stock structure and will not necessarily contribute to increase the strength of the outputs. Following this idea, Begg and Waldman (1999) advised combining at least a genetic and a phenotypic approach. The genetic-based approach provides information of stock structure at an evolutionary time scale and generally produces fewer stock units (i.e. more conservative groupings) than phenotypic-based approaches or others (Elliott *et al.*, 1995). Indeed, genetic methods can fail in discriminating stocks since little gene flow can induce genetic homogeneity between units (Hawkins *et al.*, 2016).

Each kind of tracer has its own spatiotemporal resolution and different methods can inform the stock structure in a complementary manner. Mark-recapture studies generally provide an understanding of the stock structure at a broad spatial scale but fail in determining more complex structure (Begg *et al.*, 1997). Besides, morphometric, meristic and life history traits analyses inform the stock structure at a range of spatiotemporal scales but are sometimes limited because of environmental dependent variations (Casselman *et al.*, 1981; Elliott *et al.*, 1995). Microchemistry analyses can also contribute to segregating stock units, but intra-stock variations can limit its strength in differentiating stocks (Campana *et al.*, 1995). Then, tracers accounted for separately have advantages and drawbacks, depending on the question and sampling possibilities. But, altogether, they can converge toward a consensual diagnosis.

The combination of tracers has to be planned properly before conducting the study and regarding the expected spatiotemporal resolution of the results (Tanner *et al.*, 2016) (**Fig. 1.10**).

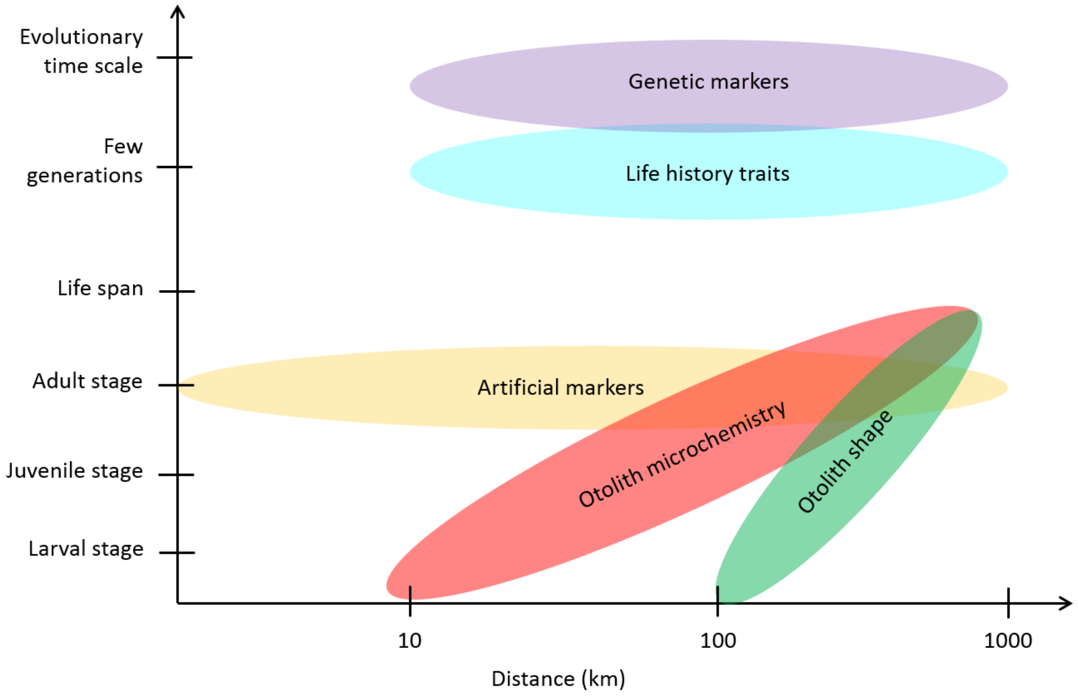

Figure 1.10 Overview of the spatiotemporal resolutions of the different kinds of tracers used in this thesis to assess population structure and connectivity. Adapted from Tanner *et al.* (2016).

Among the most commonly used combinations, genetic and otolith microchemistry approaches provide information on stock structure and connectivity at evolutionary and ecological time scales (i.e. across the life cycle of individuals), respectively (Smith and Campana, 2010; Tanner *et al.*, 2014; Reis-Santos *et al.*, 2018). Also, otolith microchemistry analyses have been combined with tagging experiments (e.g. Darnaude and Hunter, 2008; Darnaude *et al.*, 2014) or biophysical modelling (e.g. Ashford *et al.*, 2010), allowing the investigation of fine and large-scale movements of fish in early life and adult stages. Many other combinations have been applied for stock discrimination and connectivity over the last decades. However, statistical limitations have prevented the development of a really integrated approach, as developed by Neubauer *et al.* (2013) or Smith and Campana (2010) in a Bayesian framework. Recently, statistical packages such as *assignPOP* (Chen *et al.*, 2018) or MixSIAR (Stock *et al.*, 2018) have been developed for the purpose of combining different kinds of data but the range of spatiotemporal resolution of tracers was still a limiting point. To circumvent these difficulties, a synthetic Stock Differentiation Index (SDI) has been developed (Welch *et al.*, 2015; Izzo *et al.*, 2017) to easily combine tracers with various spatiotemporal resolutions into a single framework.

Therefore, the winning approach is holistic, and combined tracers must be chosen in a relevant manner before conducting the study considering the question asked and its spatiotemporal resolution, the life cycle of the targeted species, and the cost of the sampling and analyses.

1.3. Case study – The common Sole of the Eastern English Channel stock

Common sole (*Solea solea,* Linnaeus, 1758) is a nursery-dependent flatfish species of high economic importance living in coastal waters of the northeastern Atlantic Ocean. The species is distributed from western Scotland and Baltic Sea in the North to the Mediterranean Sea in the South, including the African coast as far as Senegal (Wheeler and Stebbing, 1978; Rijnsdorp *et al.*, 1992). Common sole is a benthic species highly linked to sandy and muddy substrates (Dorel *et al.*, 1991; Horwood, 1993; Gibson *et al.*, 2014). Depending on latitude, spawning period generally extends from winter to spring in coastal waters (Rochette *et al.*, 2012; Mollet *et al.*, 2013; Savina *et al.*, 2016), except in the Western English Channel and northern Bay of Biscay where spawning occurs offshore (Koutsikopoulos and Lacroix, 1992; Horwood, 1993). After hatching, larvae drift towards nursery grounds in estuarine and coastal areas where they undergo metamorphosis, become lateralized and settle for 2-3 years (Dorel *et al.*, 1991; Riou *et al.*, 2001). Juveniles are then recruited to the adult part of the population, in deeper waters. These are general features of the common sole life cycle but some differences exist between regions according to environmental conditions.

Currently, ICES considers three independent stocks in the English Channel-North Sea region: the Western English Channel (WEC; ICES area VIIe), Eastern English Channel (EEC; ICES area VIId) and the North Sea (NS; ICES area IVc) stocks (**Fig. 1.11**). A recent genetic study confirmed this separation through patterns of isolation by distance based on SNPs (Diopere *et al.*, 2018). The EEC measures around 35 000 km² with average depth of 35 m. Substrates are largely composed of sand and mud even if rocky reefs are found and could constitute natural barriers for the common sole (**Fig. 1.11**).

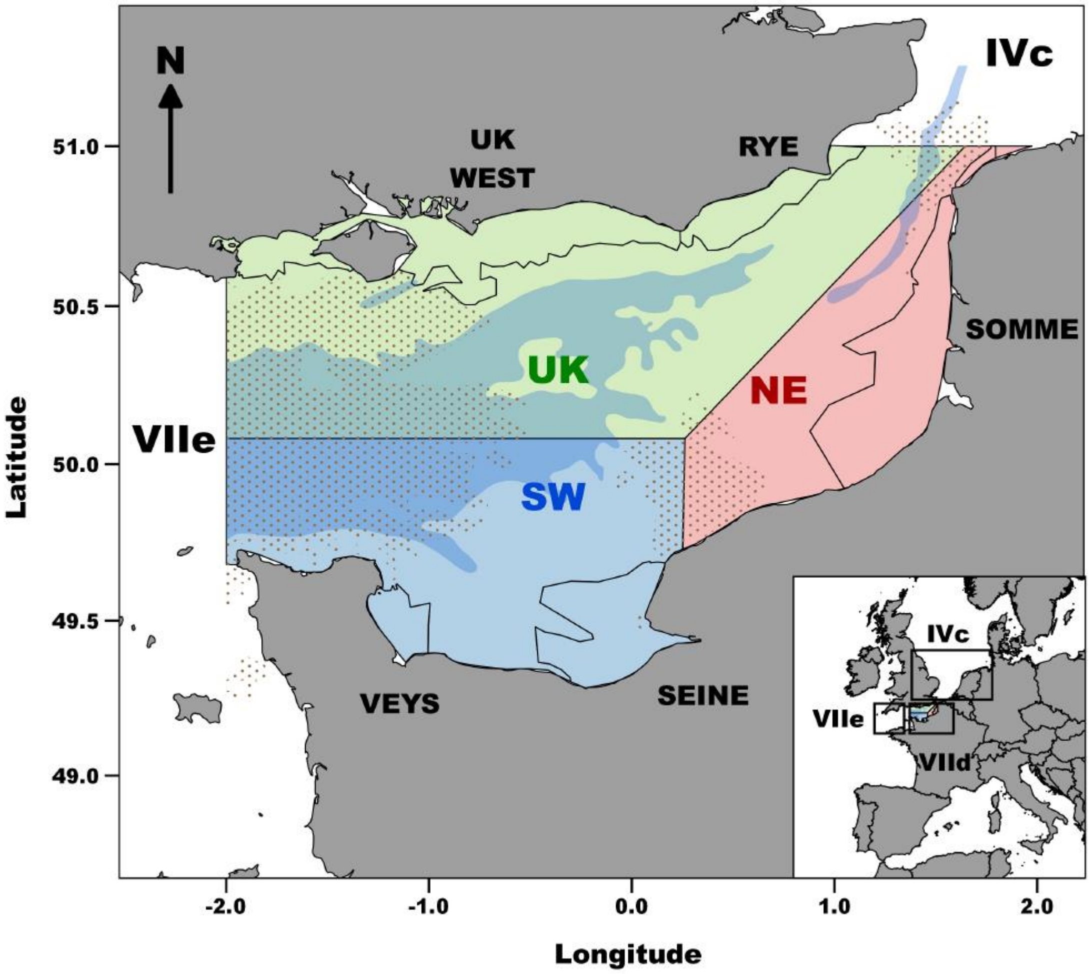

Figure. 1.11 Map representing the delineation of stocks and subunits in the English Channel-North Sea region. Three potential subunits are delineated within the Eastern English Channel stock (SW, NE and the UK). Five estuarine and coastal nurseries are delineated by solid lines (20 m isobaths). Light grey dots indicate rocky reefs. The dark blue area represents depth above 100 m.

In the EEC, spawning occurs in early spring (between March and May) near the coast, on spawning grounds of moderate depth (Riou *et al.*, 2001; Rochette *et al.*, 2012). After hatching, pelagic larvae drift for almost eight weeks (Grioche, 1998; Savina *et al.*, 2010; Rochette *et al.*, 2012) and settle in five potential nursery grounds (Rochette *et al.*, 2010): the bay of Veys and Seine estuary along the southwestern part of the French coast, the bay of Somme along the northeastern part of the French coast and the Rye and western UK nurseries along the southern English coast (**Fig. 1.11**). After metamorphosis, juvenile individuals spend for about 2.5 years in nurseries before they are recruited and move towards deeper offshore foraging areas (Riou *et al.*, 2001; Rochette *et al.*, 2010).

The ICES working group on assessment of demersal stocks in the North Sea and Skagerrak (WGNSSK) provides each year a stock assessment for the common sole in the EEC. Over the last decade, the stock has been overexploited with fishing mortality (F) above the fishing mortality threshold of sustainability (F_{MSY}, fishing mortality achieving maximum sustainable yield), except in 2017 (ICES, 2018a). Even if F was recently below F_{MSY} (in 2016 and 2017), the spawning stock biomass (SSB) was below $B_{trigger}$ (in 2016, 2017 and 2018), which means that the stock was not exploited in a sustainable manner. Conservation measures have been implemented after the drop in SSB and the poor recruitment in 2012. Belgian beam trawlers reduced the catch of undersized soles by changing the mesh size. Also, France reinforced the protection of nursery grounds, increased the nursery areas closed to fishing and increased the minimum catch size to 25 cm (ICES, 2018a).

In 2017, a benchmark was conducted in order to discuss and improve the assessment and management of common sole in the EEC (ICES, 2017a). The stock identity was questioned since the stock delineation is a strong hypothesis of stock assessment models (Cadrin *et al.*, 2013; Kerr *et al.*, 2017). Recent modelling studies have suggested three putative subunits in the EEC stock of sole (Rochette *et al.*, 2013; Archambault *et al.*, 2016). First, regarding the larval connectivity, a biophysical modelling approach found that larval advection occurred mostly towards the nearest nursery grounds (Rochette *et al.*, 2012). Additionally, the connectivity induced by juveniles was very low (Coggan and Dando, 1988) with very moderate/ small-scale movements (< 10 km ; Le Pape and Cognez, 2016) and low dispersal of immature individuals in nurseries (Riou *et al.*, 2001; Durieux *et al.*, 2010). Besides, natural barriers such as coastal rocky reefs (**Fig. 1.11**) and a deep central channel covered with gravel were expected to be unsuitable habitat for sole, preventing individuals from displaying movements between the three putative subunits: the southwestern (SW), the northeastern (NE) and English (UK) subunits (**Fig. 1.11**; Rochette *et al.*, 2012; Archambault *et al.*, 2016). However, the adult-mediated connectivity remained poorly documented (Burt and Millner, 2008; Archambault *et al.*, 2016) despite its importance in the understanding of population connectivity (Mullon *et al.*, 2002; Frisk *et al.*, 2014).

In particular, the connectivity between nurseries and spawning grounds and the level of exchange between putative subunits were unknown.

1.4. Objectives of the thesis

Considering the lack of knowledge about the structure and connectivity within the EEC stock of common sole at subadult (i.e. juveniles leaving their nurseries before the first reproduction) and adult stages, this thesis project aims at investigating the consistence of a single stock hypothesis. The current assessment and management strategies consider the stock as a single population displaying a homogeneous dynamic throughout the area. Then, **the purpose of this work is to challenge the spatial structure hypothesis under different scenarios by focusing on sub-adult and adult stages**. To handle this issue, a holistic approach was conducted and is summed up in **Fig. 1.12**.

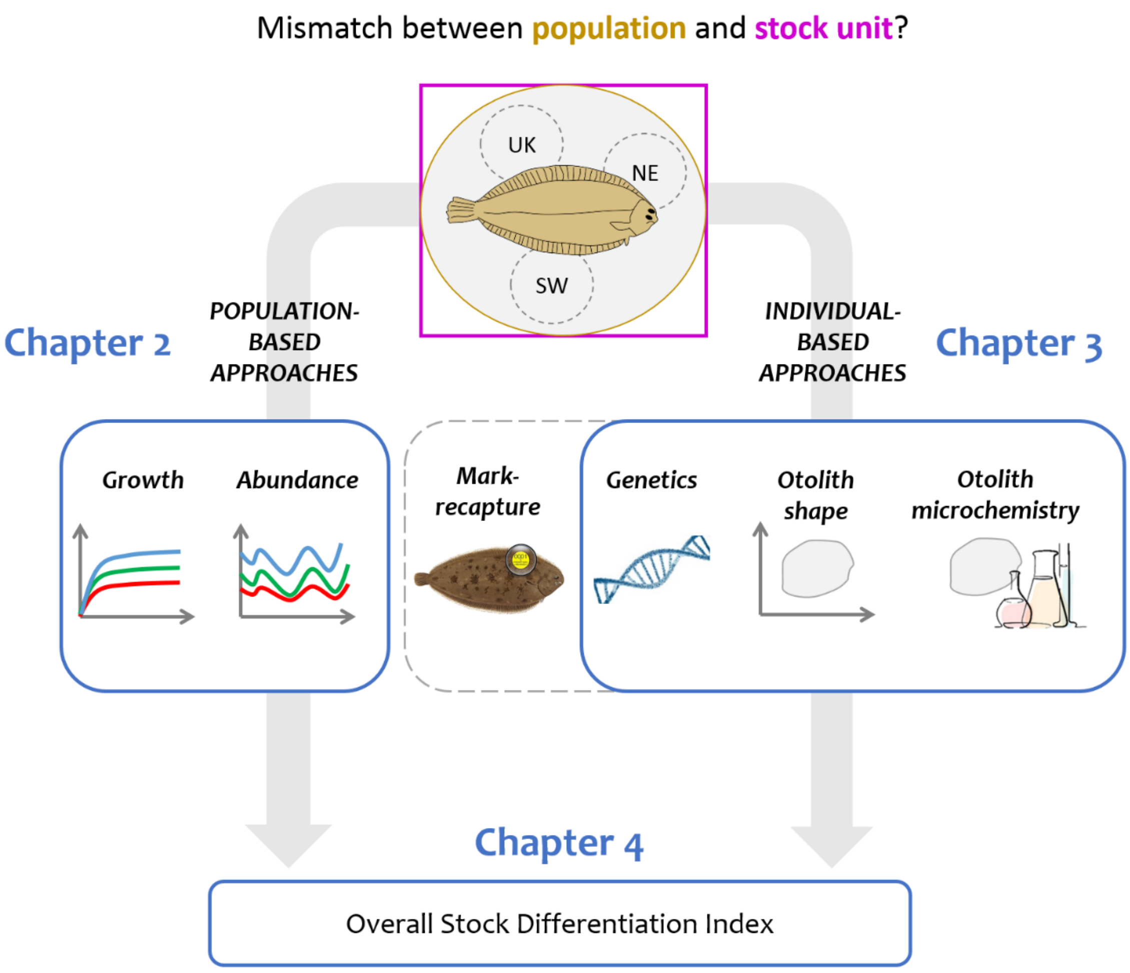

Figure. 1.12 Overview of the different approaches conducted during this thesis to assess the common sole (*Solea solea*) population structure inside the Eastern English Channel stock (ICES division VIId). The dotted line indicates that the mark-recapture analysis was not performed in this thesis but that the related findings of a previous study (Lecomte *et al.*, 2019) were included in chapter 4.

The population-based approach (**chapter 2**) investigates the EEC stock structure over a period of 20 years by combining growth and abundance analyses. Growth parameters are estimated from a von Bertalanffy growth function and spatial patterns are tested considering a single stock hypothesis (i.e. null hypothesis) and alternative spatial structures. In parallel, synchrony of abundance time series throughout the cohorts is assessed considering the null and alternative hypotheses of spatial structure. Congruence and discrepancies between the growth and abundance analyses are finally discussed and first conclusions regarding the spatial structure of the EEC stock of common sole are proposed.

Chapter 3 presents an individual-based approach to go deeper into the assessment of spatial stock structure and to investigate the connectivity within the stock. Using up-to-date genetic markers and otolith shape descriptors, the stock spatial structure is examined and alternative hypotheses of spatial are tested. Besides, analysis of otolith microchemistry is performed to estimate the nurseries of origin of adults caught in deeper waters during the spawning period and then rebuild *a posteriori* the connectivity along the life cycle. Focusing on the adult individuals used in genetic, otolith shape and microchemistry analyses, an integrated analysis is then conducted to test for congruence and divergence between tracers. Results from the single and integrated analyses are discussed and conclusions of spatial structure and connectivity are proposed.

Finally, **chapter 4** combines and discusses results from the population and individual-based approaches and integrates mark-recapture results (Lecomte *et al.*, 2019). Considering the difficulty of combining methods with various spatiotemporal scales, a Stock Differentiation Index (SDI; Welch *et al.*, 2015; Izzo *et al.*, 2017) is computed to synthetize and integrate the results of chapters 2 and 3. The relevance of each analysis is discussed and conclusions regarding the consistence of the current stock hypothesis are drawn. Alternative assessment and management strategies are discussed.

Chapter 2

Population-based approaches to investigate the spatial structure of the stock

Population-based approaches

2.1. Introduction of chapter

The mismatch between stock units and biological populations can induce overexploitation of the less productive subunits, leading to stock depletion (Fu and Fanning, 2004; Cadrin and Secor, 2009; Ying *et al.*, 2011; Goethel and Berger, 2017). Mismatch occurs when the extent of the stock is misaligned with the underlying population or when spatial structure exists within the population. To detect such misalignment, the recommended approach falls within the combination of tracers allowing the detection of a signal of population structure (Begg and Waldman, 1999; Waldman, 1999; Cadrin *et al.*, 2013). Among the large range of tracers available to investigate stock identity, population-based approaches should be used as the first step of the stock identification process (Begg, 2005). Indeed, population-based approaches provide an overview of the stock structure by estimating characteristics or parameters at the population scale. Spatial differences in life history traits or demographic attributes would suggest independent spatial units experiencing various environmental conditions and/or displaying low levels of mixing between subunits. However, the lack of spatial differences would suggest that (i) there is evidence of spatial homogeneity (i.e. the population is aligned with the current stock definition) or (ii) there is a type I error suggesting that no differences were found whereas there is effectively spatial heterogeneity between subunits. Besides, life history parameters and demographic attributes have the advantage to be easily available from historic data sets of scientific surveys and can be inferred from these long-term time series.

The Eastern English Channel and North Sea stocks of common sole are sampled every year during the summer by the UK Beam Trawl Survey (UK-BTS). This routine survey provides long-time series of length-at-age data for mature soles over 20 years. Then, estimates of growth parameters and abundance-at-age can be used to fulfil the poor understanding of stock structure. Du Pontavice *et al.* (2018) conducted a preliminary study of spatial stock structure based on an estimate of growth parameters over a short and recent period (2010-2015; see the article provided in supplementary material). The three-subunit hypothesis suggested by previous modelling studies (i.e. SW, NE and UK subunits; **Fig 1.11**; Rochette *et al.*, 2012; Archambault *et al.*, 2016) was tested and results highlighted that asymptotic length was significantly higher in the SW than in the UK subunit and that smaller fish were found in the NE subunit. This preliminary analysis presented two main limitations: (1) the short time span of the study prevented the authors from analyzing temporal evolution of spatial structure, which is essential for this kind of study (Begg, 2005), and (2) only one tracer was used, which is not recommended to conclude on the spatial structure of a stock (Begg and Waldman, 1999).

Therefore, in this second chapter, population growth and abundance analyses are presented in a single population-based approach to inform the stock structure over 20 years. Spatial differences in growth parameters and asynchrony between abundance-at-age time series (i.e. no correlation) are expected to be a signal of stock structure. Indeed, spatial contrasts in growth parameters between subunits can be found in case of contrasted growing conditions due to both patterns in environmental features and limited exchanges between subunits. Regarding the abundance-at-age analysis, times series are expected to be synchronous throughout the cohorts within a subunit in case of non-mixing with other subunits. At the opposite, synchrony of abundance-at-age time series is expected within a well-mixed population.

As recommended by Cadrin *et al.* (2014), different hypotheses of spatial structure were considered in this study: the null hypothesis (i.e. the current stock definition) and alternative hypotheses of spatial structure were tested by pooling by pair the primary subunits SW, NE and UK.

Could we consider a single stock when spatial subunits present lasting patterns in growth and asynchrony in cohort densities? A flatfish case study

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Accepted the 21 September 2018 in *Journal of Sea Research*
(https://doi.org/10.1016/j.seares.2018.09.012)

2.2. Abstract

An accurate representation of the spatial structure of marine fish populations is a prerequisite for unbiased stock assessment, to build appropriate management measures. The common sole (Solea solea, L.) of the Eastern English Channel (EEC) is a commercial flatfish species, whose stock is currently assessed as a single homogeneous population and has been overexploited over the last decade. Previous studies have highlighted the stock's low connectivity and the lack of understanding of sub-adults and adults mixing between putative subunits, raising the issue of a potential spatial structure of this stock. Here, we examined evidence of spatial structure by analyzing spatiotemporal patterns of length and density-at-age using time series (1989-2015) obtained from a scientific survey (UK-BTS). We tested for various hypotheses of spatial structure, based on both scientific and expert knowledge, including three isolated subunits, their combination, and no spatial structure. We combined two sets of analyses: (1) a selection of the von Bertalanffy growth model with spatial effects capturing the most accurate spatial structure of the stock and the analysis of long-term spatial patterns (gradients, trends, synchrony) in growth parameters; and (2) an analysis of the synchrony among density-at-age time series between spatial subunits. Growth analysis revealed a spatial structure in three subunits (i.e. the southwestern, northeastern and English parts of the EEC) and an overall decline of length-at-age, suggesting Fishery-Induced Evolution. The synchrony analysis revealed high spatiotemporal integrity at the level of the southwestern subunit of the EEC. Our two analyses thus detected a lasting signal of spatial stock structure with a probable isolation of the southwestern subunit from the rest of the EEC. Future research should build on our study by investigating the connectivity of sole throughout its entire life cycle, to improve stock assessment and fishery management.

Keywords

Population structure – Growth – Density – Synchrony – *Solea solea* – Eastern English Channel

2.3. Introduction

According to Ihssen *et al.* (1981), a stock is a monospecific group of individuals that randomly mate and displays spatiotemporal integrity. An accurate delineation of stocks is a prerequisite for setting appropriate fisheries management measures (Kutkuhn, 1981; Smith *et al.*, 1990; Begg *et al.*, 1999a). However, population structure at different geographic scales and life stages is common (Waples and Gaggiotti, 2006; Reiss *et al.*, 2009; Ames and Lichter, 2013; Ciannelli *et al.*, 2013), and should be considered in stock assessments (Carson *et al.*, 2011; Petitgas *et al.*, 2013; Frisk *et al.*, 2014). Inaccurate representation of the spatial structure of (meta)population, e.g. by ignoring the existence of independent subunits, or of connectivity and exchanges with other stocks, bias estimates of population life history (i.e., growth, maturity and mortality) (Cadrin *et al.*, 2013; Kerr *et al.*, 2017). When the understanding of stock structure and delineation is limited (Cadrin *et al.*, 2010; Zemeckis *et al.*, 2014; Mahe *et al.*, 2016), current practice assumes homogeneous vital rates without contrasts between putative subunits (Cadrin *et al.*, 2013). This can induce a mismatch between the management unit and ecological connectivity (Hawkins *et al.*, 2016; Kerr *et al.*, 2017), resulting in the overexploitation of less productive subunits and underexploitation of more productive ones (Fu and Fanning, 2004; Cadrin and Secor, 2009; Ying *et al.*, 2011; Goethel and Berger, 2017).

Different methods exist to identify and delineate stocks (Östman *et al.*, 2017). Genetic markers (microsatellites (e.g. Cuveliers *et al.*, 2012; Jasonowicz *et al.*, 2016), or Single Nucleotide Polymorphisms (e.g. Milano *et al.*, 2014; Laconcha *et al.*, 2015)); morphometry and meristics (Allaya *et al.*, 2016; Sley *et al.*, 2016); parasites (Catalano *et al.*, 2014; MacKenzie and Abaunza, 2014); otolith shape (Hüssy *et al.*, 2016; Mahé *et al.*, 2016) or microchemistry (Tanner *et al.*, 2016; Moreira *et al.*, 2018)); and tagging (Rogers *et al.*, 2017; Le Bris *et al.*, 2018) are widely used. Although easily available from survey data, life history traits such as abundance, growth, and maturity are rarely used to analyze the spatial structure of populations (Begg and Waldman, 1999; Begg *et al.*, 1999b; Cadrin *et al.*, 2013; Erlandsson *et al.*, 2017). Yet, long-term time series derived from field surveys are frequently available at no cost, allowing the assessment of spatial structure while accounting for temporal integrity (Begg *et al.*, 1999a; Cope and Punt, 2009).

The analysis of correlations in temporal fluctuations of life history traits and demographic attributes among populations (e.g. spatial synchrony; Walter *et al.*, 2017) is an underused but valuable method to investigate the spatiotemporal structure of natural populations (Botsford and Paulsen, 2000; Rushing *et al.*, 2016; Rogers *et al.*, 2017). Dispersal and Moran effects (i.e. effects of correlated fluctuations in environmental drivers on synchrony among populations) have repeatedly been highlighted as structuring observed patterns (Ranta *et al.*, 1995; Liebhold *et al.*, 2004). In the context of stock structure identification, if there were synchronous environmental drivers over stock subunits and spatially asynchronous life history traits, it would indicate that the stock is spatially structured. The stock is “spatially structured” in case of persistent spatial asynchrony in life history traits among subunits.

For decades, stock assessments of the common sole (*Solea solea*), a commercial species of main interest (ICES, 2017b), have considered three independent stocks in the English Channel-North Sea Region: the North Sea (ICES division IVc), the Eastern English Channel (EEC; ICES division VIId) and the Western English Channel (ICES division VIIe) stocks (**Fig. 2.1**). This separation is in accordance with patterns of isolation by distance (Diopere *et al.*, 2018). In the EEC, reproduction occurs in early spring on spawning grounds (Rochette *et al.*, 2012). Once hatched, pelagic larvae drift for almost two months towards shallow estuarine and coastal nursery grounds (Grioche, 1998; Savina *et al.*, 2010; Rochette *et al.*, 2012). After metamorphosis, juveniles grow on these shallow nursery grounds for about two years before moving to deeper offshore adult foraging grounds (Riou *et al.*, 2001; Rochette *et al.*, 2010). Uncertainty remains regarding the spatial unity of the stock (Rochette *et al.*, 2013; Archambault *et al.*, 2016; ICES, 2017a). Larval connectivity is low since spawning areas directly feed adjacent coastal and estuarine nursery grounds (Rochette *et al.*, 2012). Besides, very moderate movements of juvenile fish on small scales (<10 km; Le Pape and Cognez, 2016) and their strong dependence upon local nursery habitats (Riou *et al.*, 2001) result in low juvenile connectivity (Coggan and Dando, 1988). However, connectivity among subunits as a result of adult movement, a potentially important driver of population segregation (Mullon *et al.*, 2002; Frisk *et al.*, 2014) still remains partially unknown (Burt and Millner, 2008; Archambault *et al.*, 2016). Based on several lines of evidence, three subunits of the stock appeared a realistic hypothesis in the EEC (Rochette *et al.*, 2012; Archambault *et al.*, 2016): the Bay of Seine (southwest subunit, SW), the Northern French coast (northeast subunit, NE) and the southern English coast (English subunit, UK) (**Fig. 2.1**). Natural barriers with unsuitable habitats for adult sole (i.e. large and deep gravel grounds in the middle of Eastern Channel, wide rocky reefs from shallow to deep areas; Rochette *et al.*, 2012; Archambault *et al.*, 2016) separate these subunits. Considering metapopulation dynamics among these potential subunits in the EEC would drastically change inferences on population dynamics and stock assessment (Archambault *et al.*, 2016).

Based on a von Bertalanffy growth model (VB) to analyze length-at-age data from commercial landings and scientific survey over a short period (2010-2015), Du Pontavice *et al.* (2018) found spatial differences in asymptotic length and length-at-age 2 between the three subunits described above. However, limitations prevented to conclude on the spatial structure from this study. First, the use of a single stock identification method is not sufficient to provide robust conclusion about the stock structure. Indeed, different stock identification methods may provide inconsistent results about the stock structure, and the use of a multiple approach is recommended (Begg and Waldman, 1999; Waldman, 1999; Cadrin *et al.*, 2013). Second, authors investigated spatial patterns of growth parameters over a short period, but the temporal evolution was not examined, despite its importance to understanding spatial stock structure (Begg *et al.*, 1999b). Thirdly, Du Pontavice *et al.* (2018) only tested the three-subunit hypothesis whereas alternative hypotheses deserve to be tested to investigate the stock structure (Begg, 2005). Here, we investigate the spatiotemporal consistency of a stock structure in the EEC by analyzing spatiotemporal patterns of key life history traits over 26 years. We expanded from Du Pontavice *et al.* (2018) by analyzing, over a longer series (1989-2015), density-at-age in addition to length-at-age data, both estimated from a scientific survey dataset. We tested for different configurations of spatial structure, including three isolated subunits (Rochette *et al.*, 2012; Archambault *et al.*, 2016; ICES, 2017a); **Fig. 2.1**); two isolated subunits (combination of the 3) or a single stock (no spatial structure, i.e. the present management unit). Specifically, we combined two sets of analysis: (1) we used model selection to select the VB growth model that reflected the more probable spatial structure, then we investigated long-term spatial differences and synchrony in growth parameters; (2) we used multivariate time series analyses to assess intra-subunit synchrony among density-at-age time series. The growth analysis focused on patterns, trends and spatial synchrony among subunits, whereas the density-at-age analysis examined the strength of the intra-subunit synchrony to assess the spatial structure of the EEC stock of common sole. We assume that long-term differences and spatial asynchrony in growth parameters among stock units, combined with strong intra-subunit synchrony in density-at-age time series are convergent indices of a spatial structure.

2.4. Material and methods

2.4.1. Challenging various hypotheses of spatial structure

Growth and density-at-age data were analyzed using five different hypotheses of spatial structure in the EEC (**Fig. 2.1**, **Table 2.1**). The null hypothesis (H0) considered that the EEC is a single stock (i.e. no spatial structure), while the remaining four alternative hypotheses assumed for the stock to be partitioned into two or three subunits.

Table 2.1. Hypotheses of spatial structure inside the common sole stock of the Eastern English Channel. The three subunits (H1) considered are the UK (along the southern English coast), NE (North-East French coasts) and SW (South-West French coasts) subunits. These primary subunits are pooled to define the null hypothesis (H0) and alternative hypotheses (H2, H3 and H4). "+" means that primary subunits are pooled.

Hypotheses	Subunits	Number of subunits
H0	UK + NE + SW	1 single stock
H1	UK, NE and SW	3
H2	(NE + SW) and UK	2
H3	(UK + SW) and NE	2
H4	(UK + NE) and SW	2

2.4.2. Length-at-age and density-at-age datasets

The UK- Beam Trawl Survey (BTS) is an annual scientific survey conducted by the Center for Environment Fisheries and Aquaculture Science (Cefas) since 1989. It covers the EEC and the southern part of the North Sea. Sampling takes place in July and August, following the same survey design each year (**Fig. 2.1**), using a commercial 4 m beam trawl (ICES, 2009). Each sole caught is measured (total length), sexed and aged by reading otolith increments. Data from this survey provide consistent time series of lengths and abundances-at-age by sex throughout the EEC over the period 1989-2015.

However, to ensure sufficient numbers of common sole per age, sex and subunit categories, we removed three cohorts (1992, 2006 and 2007) from the dataset, due to the low occurrence and abundance of sole.

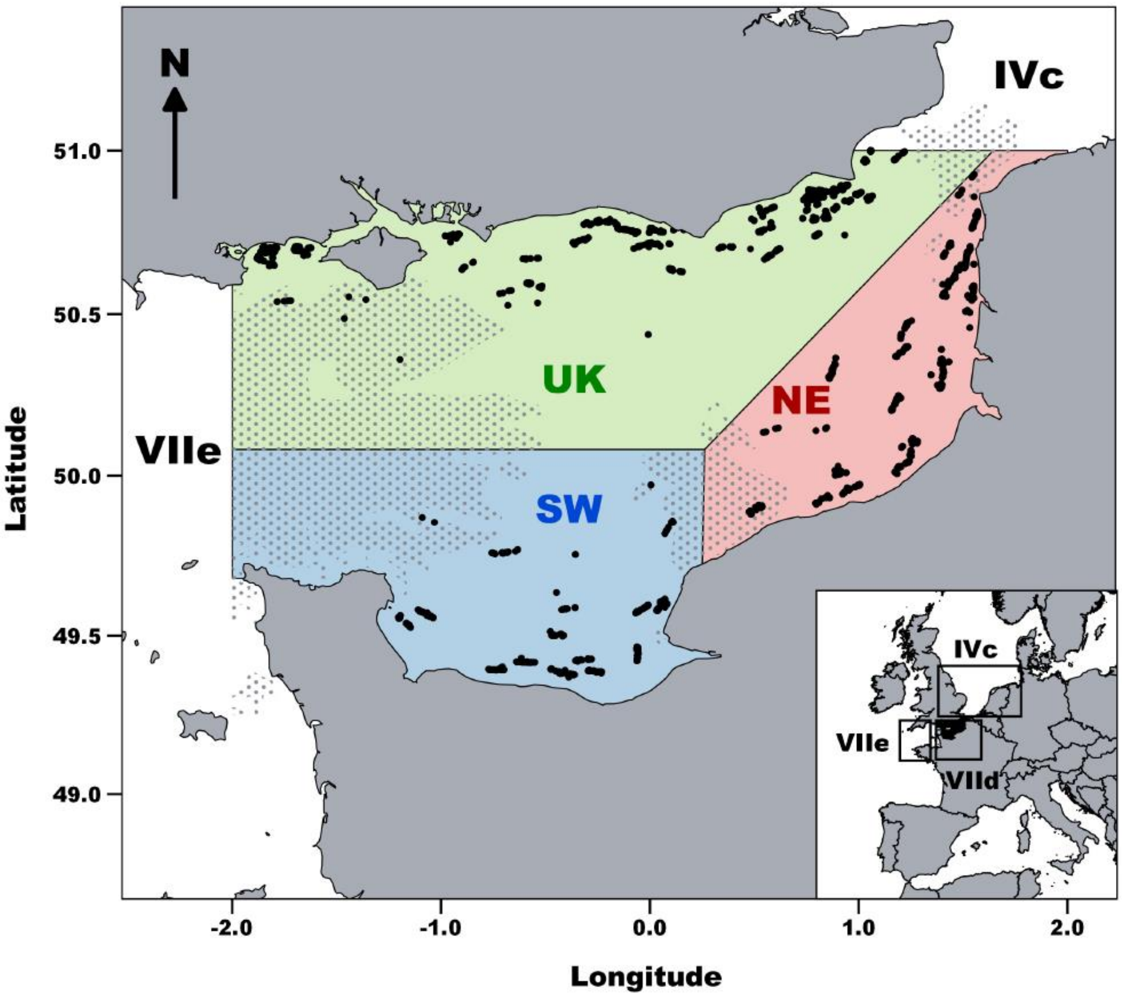

Figure. 2.1 Map of sampling of common sole inside the Eastern English Channel stock (ICES division VIId) including the three putative subunits tested for in this study (UK, NE and the SW). Light grey dots indicate rocky reefs. Black dots correspond to the location of the UK-BTS sampling stations from 1989 to 2015.

Regarding the growth analysis, to avoid bias in estimates of growth parameters, we removed post-2008 cohorts, as old individuals (i.e. age > 8) have not been caught yet, preventing to accurately estimate L_∞ (i.e. the asymptotic length) in VB growth models. Seventeen cohorts from 1989 to 2008, corresponding to a sufficient number of females and males (ages 1 to 19), caught in the three putative subunits were finally selected (**Table 2.2**).

Regarding the density-at-age analysis, to ensure sufficient numbers of fish per age, cohort, sex and subunit categories, we only included soles between ages 1 and 5 (19 cohorts, from 1989 to 2005; **Table 2.2**).

The full data set consists of 11,296 and 12,217 common soles available for growth and density-at-age analyses, respectively (**Table 2.2**).

Table 2.2. Number of common soles sampled by the UK-Beam Trawl Survey (BTS) in the Eastern English Channel and used in growth and density-at-age analyses.

	Selected ages	Selected cohorts	Number of fish			
			Subunit	Sex		Total
				Females	Males	
Growth	1-19	1989-1991+ 1993-2005+ 2008	UK	2903	2740	5643
			NE	1339	1704	3043
			SW	1348	1262	2610
Density	1-5	1989-1991+ 1993-2005+ 2008-2010	UK	3187	2954	6141
			NE	1511	1861	3372
			SW	1397	1307	2704

All growth and density-at-age analyses detailed below were performed using the R software (R Development Core Team, R.3.1.1, 2016).

2.4.3. Growth modelling

Length-at-age data were analyzed using the von Bertalanffy Growth Function (von Bertalanffy, 1957). We applied the same method as Du Pontavice *et al.* (2018) to assess estimates of growth parameters with a von Bertalanffy growth function, from age 1 (***Eq. 2.1***):

$$\mathbf{L_{t,i} = L_\infty - (L_\infty - L_1) \times exp\,(-K \times (t - 1)) + \varepsilon_i} \qquad \textbf{(\textit{Equation. 2.1})}$$

with $L_{t,i}$, representing total length of the sole i at age t (in mm); $\boldsymbol{\varepsilon_i}$**,** a normally distributed error term considered independent among all individuals and with a homogeneous variance; L_1, total length at age 1; L_∞, asymptotic length and K, growth rate (in year^{-1}). The three parameters were estimated using a maximum likelihood framework using the nonlinear least squares procedure (*nls* function) function of the R package *stats*.

- **Model selection**

Since sexual dimorphism in growth has been documented for the common sole (Rijnsdorp and Van Beek, 1991), we introduced a “sex” effect on growth parameters. Thus, for each sex, we investigated variations in growth parameters across time (cohorts) and space (subunits) to test for a signal of spatial stock structure.

We selected the most appropriate combination of “cohort”, “subunit” and “sex” effects on the three estimated parameters of the VB based on the resulting Akaike Information Criterion (AIC). As preconized by Burnham and Anderson (2003), we also included AIC weights to compare the probability of each model. AIC weights were assessed using the *Akaike.weights* function of the *qpcR* package.

We first tested models which considered the sequential introduction of effects on each parameter separately (L_1, L_∞ or K), on two parameters (L_1 and L_∞, L_1 and K, L_∞ and K) and on each parameter simultaneously (L_1, L_∞ and K). The sequential introduction of effects on the parameter(s) consisted of introducing the “sex” effect then the “cohort” effect and finally the ”subunit”, by contrasting the 5 hypotheses of spatial structure (H0 - H4; **Table 2.1**).

- **Assessing spatiotemporal patterns of growth parameters**

As estimates of K and L_∞ are negatively correlated in the VB (Schnute, 1981), the inter-cohort variability for both parameters can represent statistical flukes rather than an ecological signal in the data. Therefore, we used L_{10} (the estimated length at age 10) as an indicator of growth, as it is more robust to statistical correlation than parameters taken individually. We arrived at L_{10} by preliminarily testing the correlation between parameters K and L_∞ and L_{age} and found that L_{10} was the appropriate parameter to investigate growth in this study focused on adult stages. L_{10} was chosen to explore spatial structure since it is more influenced by L_∞ than by K and L_1, more relative to growth of juveniles.

Variance estimates were calculated using the delta method, which is a method for deriving the variance of a function of asymptotically normal random variables (Casella and Berger, 2002).

We qualitatively examined L_{10} trends per sex and subunit to evaluate the appearance, maintenance or disappearance of spatial patterns of growth over the time series.

- **Spatial synchrony in time series of growth parameters**

We tested the spatial synchrony of time series (cohort year) of L_{10} to investigate the covariation in time trends among subunits. Covariations were assessed using multivariate time series analysis with the *MARSS* package in R (Holmes *et al.*, 2013). This package allows for fitting time series models with (or without) covariates to a set of multivariate time series data. Given a set of $i=1,\dots,m$ time series of data of length $t=1,\dots,T$ (17 different cohorts are considered, $T=17$) denoted $y_{i,t}$, the overarching model comprises a state process (***Eq. 2.2***), which defines the time-series model including covariation between the m time series, and an observation process (***Eq. 2.3***), including observation errors in the data.

$$\mathbf{x_t} = \mathbf{x_{t-1}} + \mathbf{u} + \mathbf{w_t}, \text{ where } \mathbf{w_t} \sim \mathbf{MVN(0,Q)} \qquad \textbf{(\textit{Equation. 2.2})}$$

$$\mathbf{y_t} = \mathbf{Z_t x_t} + \mathbf{v_t}, \text{ where } \mathbf{v_t} \sim \mathbf{MVN(0,R)} \qquad \textbf{(\textit{Equation. 2.3})}$$

with $x_t = x_{i,t}$, $i = 1,\dots,m$ the vector of size m of the state at any time step t, u a vector of size m accounting for any systematic trend in the time series x_t and w_t the multivariate Normal (MVN**)** process error with Q an $m*m$ matrix describing the correlation between process deviations. y_t is the vector of observations at any time step t and v_t is the MVN observation error with $m*m$ covariance matrix R. Process and observation errors inform on model reliability.

This general framework was applied to estimate the spatial covariations in the time series (cohorts) of $L_{10i,t}$ previously estimated from the VB growth model, where i defines the indices of spatial subunits and t the cohort. There were twice as many time series m as subunits (separate females and males). The linear time trend u and covariation matrix Q were estimated without constraints. The covariance matrix of observation errors R was constrained to be diagonal (no covariation in observation errors), with all variance parameters on the diagonal to be equal (same observation error variance for all the time series).

The variance-covariance matrix Q was transformed into a correlation matrix using the *covtocor* function of the *hapsim* package and visualized using the function *corrplot* of the *corrplot* package in R.

2.4.4. Assessing the consistency in the variation of densities within subunits

In this analysis, we assessed to what extent variations of density-at-age across time propagates through a cohort at successive fish ages, and how the strength of this statistical signal changes given our hypotheses on the spatial structure of the stock (**Table 2.1**). High consistency in cohorts' density throughout age classes within a spatial unit would be an indication of isolation of this subunit.

- **Anomalies of density-at-age time series**

We used data on the number of individuals in **Table 2.2** to build abundance indices at age, per cohort, per sex and for each spatial unit considered in **Table 2.1**. Based on the density-at-age dataset (i.e. ages 1-5, from cohort 1989 to 2010), we first calculated the density per sex, age, cohort and subunit, as the abundance relative to the trawled surface (***Eq. 2.4***):

$$\mathbf{Density}_{\mathbf{Sex,Subunit,Age,Cohort}} = \frac{\mathbf{Abundance}_{\mathbf{Sex,Subunit,Age,Cohort}}}{\sum(\mathbf{Surface\ trawled}_{\mathbf{Sex,Subunit,Cohort+Age}})} \qquad \textbf{(\textit{Equation. 2.4})}$$

The decrease in abundance with age resulting from both natural and fishing mortality is strong. This decline could blur the statistical signal of covariations in abundance among age classes throughout a cohort. To avoid such limitation, we first standardized density indices from BTS surveys into anomalies using average density at age (***Eq. 2.5***):

$$\mathbf{Anomaly}_{\mathbf{Sex,Subunit,Age,Cohort}} = \frac{\mathbf{Density}_{\mathbf{Sex,Subunit,Age,Cohort}} - \mathbf{Mean}(\mathbf{Density}_{\mathbf{Sex,Subunit,Age}})}{\sigma(\mathbf{Density}_{\mathbf{Sex,Subunit,Age}})} \qquad \textbf{(\textit{Equation. 2.5})}$$

- **Spatial synchrony of density-at-age anomalies**

The *MARSS* package allowed for assessing the strength of the correlation in the variation of density (anomaly) across ages and cohorts within a spatial unit. Thus, referring to notations introduced in section 2.3.3, we assessed the covariations among m ($i=1,\ldots,m$) series $y_{i,t}$ where $y_{i,t}$ denote the anomalies of density of the cohort t ($t=1,\ldots,T$; nineteen different cohorts are considered here, $T=19$) and i denotes the combination of age classes (5) and spatial units considered (**Table 2.1**). For instance, under hypothesis H0 (no spatial structure), $m=5$ as only 5 series of abundance at ages 1 to 5 are considered. Under the hypothesis H1 (3 subunits), $m=15$ as 5 time series of density-at-age (ages 1 through 5) are considered for each of the 3 subunits.

We performed separate analyses for each of the 5 alternative hypotheses regarding spatial stock structure (**Table 2.1**), for males and females independently because no information allows for considering that they have the same dynamics. The linear time trend and covariation matrix where estimated without constraints. The covariance matrix of observation errors was constrained to be diagonal, with equal variance parameters on the diagonal. The variance-covariance matrices were transformed into correlation matrices using the *covtocor* function then plotted with the *corrplot* function.

2.5. Results

2.5.1. Spatiotemporal patterns of growth parameters

- **Model selection**

The “sex”, “cohort” and “subunit” effects were first tested on K, L_∞ and L_1 separately and then simultaneously. Models that simultaneously included effects on the three parameters systematically outperformed models that considered effects on each parameter separately (not presented). The following models therefore considered the sequential introduction of effects on the three parameters simultaneously.

As expected, preliminary results showed that the variable “sex” captured the greatest part of the variation, followed by “cohort” and “subunit” (**Table 2.3**). In other words, models including sex and cohort effects always outperformed models without sex or cohort effects, regardless of the spatial structure considered. Finally, regarding both AIC weights and evidence ratios, results showed that the best model (having the higher conditional probability among models) agreed with hypothesis H1 (**Table 2.3**). This model showed no violations of normality or homoscedasticity in the data (**Fig. S.2.1**).

Table 2.3. Akaike Information Criterion (AIC) and AIC weights of each model tested considering different spatial structure hypotheses (the null hypothesis H0 and 4 alternative hypotheses H1, H2, H3 and H4). Sex, cohort and subunit effects on the three parameters of the von Bertalanffy growth function were included simultaneously. H0 = single stock, H1 = NE, SW and UK subunits, H2 = (NE + SW) and UK subunits, H3 = (UK + SW) and NE subunits, H4 = (UK + NE) and SW subunits.

Structure hypotheses	Effects on parameters L_∞, L_1 and K	AIC	Akaike Weights
H0	Sex	109924	0
	Sex, Cohort	102762	0
H1	Sex, Cohort, Subunit	101149	1
H2	Sex, Cohort, Subunit	101347	0
H3	Sex, Cohort, Subunit	101925	0
H4	Sex, Cohort, Subunit	102514	0

- **Spatiotemporal trends in growth**

Growth parameters K, L_∞ and L_1 were estimated from the best model for each sex, cohort and spatial subunits (H1). The parameter L_{10} was then calculated from K, L_∞ and L_1 for each sex, cohort and subunit (**Fig. 2.2**; see the related growth curves in **Fig. S2.2**).

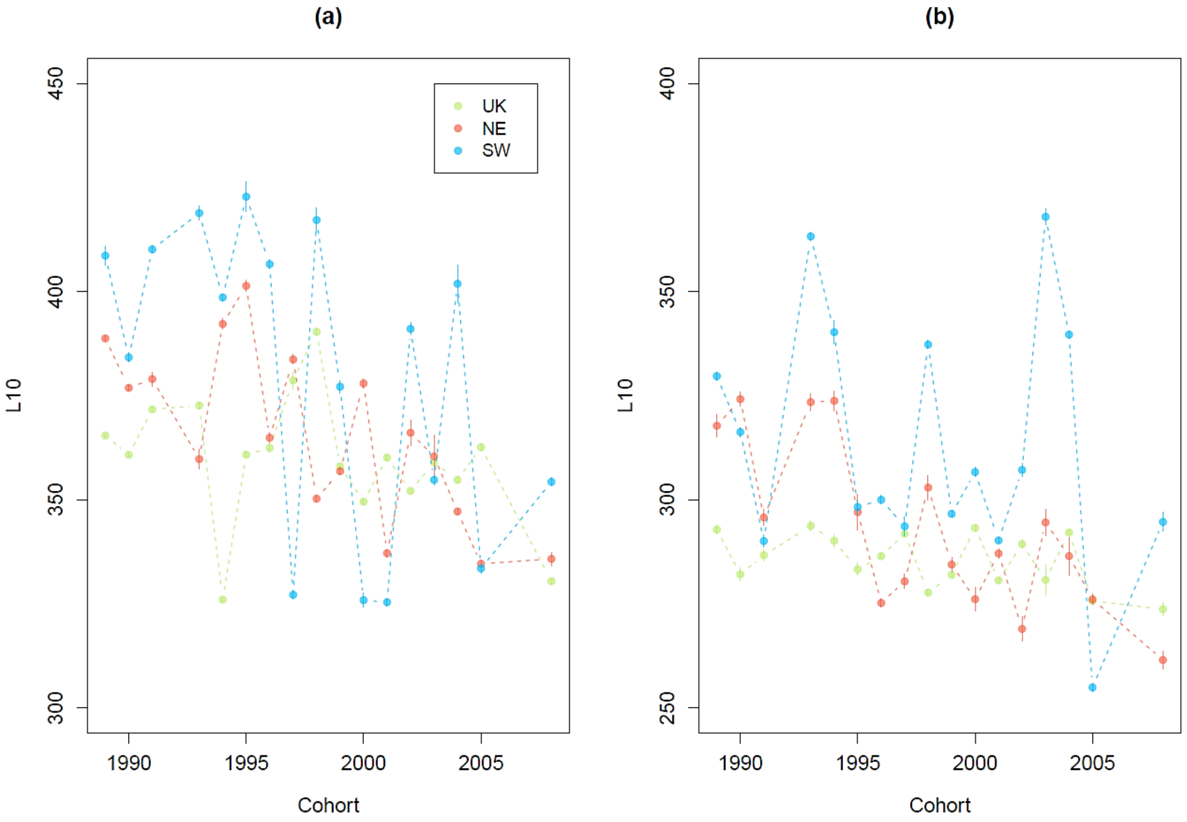

Figure. 2.2 L_{10} time series for females (a) and males (b) within each subunit (UK, NE and SW) with their associated standard errors.

The spatial synchrony analysis of growth parameters yielded negative values for vectors u (**Table S.2.1**), with L_{10} exhibiting a generally decreasing trend among cohorts from 1989 to 2008, regardless of sex and subunit (**Fig. 2.2**). Over this period, females' L_{10} declined by 35, 53 and 54 mm in the UK, NE and SW subunits, respectively (**Fig. 2.2a**), corresponding to a loss of 10 to 14% in size. For males (**Fig. 2.2b**) L_{10} decreased by 19, 56 and 35 mm in the UK, NE and SW subunits respectively, representing a loss of 7 to 18 %.

Over the whole time series, common sole growth, estimated from L_{10}, was generally lower in the NE than in the SW subunit. The situation was more complex along the UK coast where growth was comparable to estimates for the NE subunit for the first half of the time series, but declined less rapidly than in the two other subunits and was closer to growth estimates in the SW since around 2000. However, in the SW subunit, L_{10} presented high inter-cohort variability (**Fig. 2.2**).

- **Spatial synchrony in growth parameter time series**

The estimated correlation matrix did not reveal spatial synchrony in the L_{10} time series between the three subunits (**Fig. 2.3**).

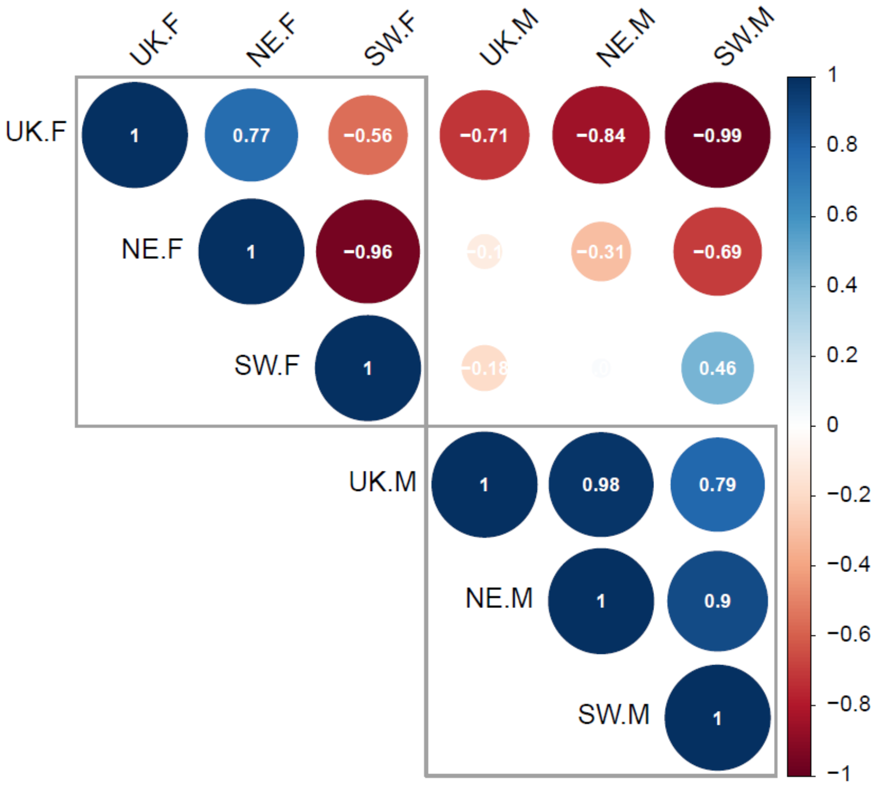

Figure. 2.3 Pair-wise correlation matrix between L_{10} time series by sex and subunits. F and M refer to female and male time series. The UK, NE and SW are the three subunits considered (hypothesis H1). The colors and sizes of the circles indicate the direction (positive or negative) and strength of the correlation. Grey squares separate females and males' spatial correlations.

We found high correlations (>0.79) in L_{10} time series among the three subunits for males (**Fig. 2.3**), indicating high growth synchrony. However, female growth exhibited strong negative correlation (< -0.90) between the SW and UK subunits (**Fig. 2.3**), and a positive correlation between the UK and NE subunits. The interannual variations in growth was asynchronous between the SW and the other subunits.

However, these results have to be interpreted with caution since observation and process errors were high, especially in the SW subunit for both sexes. Process error was > 1000 in the SW subunits whereas it varied between 6 and 100 in other subunits.

2.5.2. Spatiotemporal patterns of density-at-age anomalies

We did not find any trend in the time series of density-at-age anomalies regardless of the subunits and sex considered (**Fig. 2.4**). These results were supported by vector u estimates from the *MARSS* models (not presented).

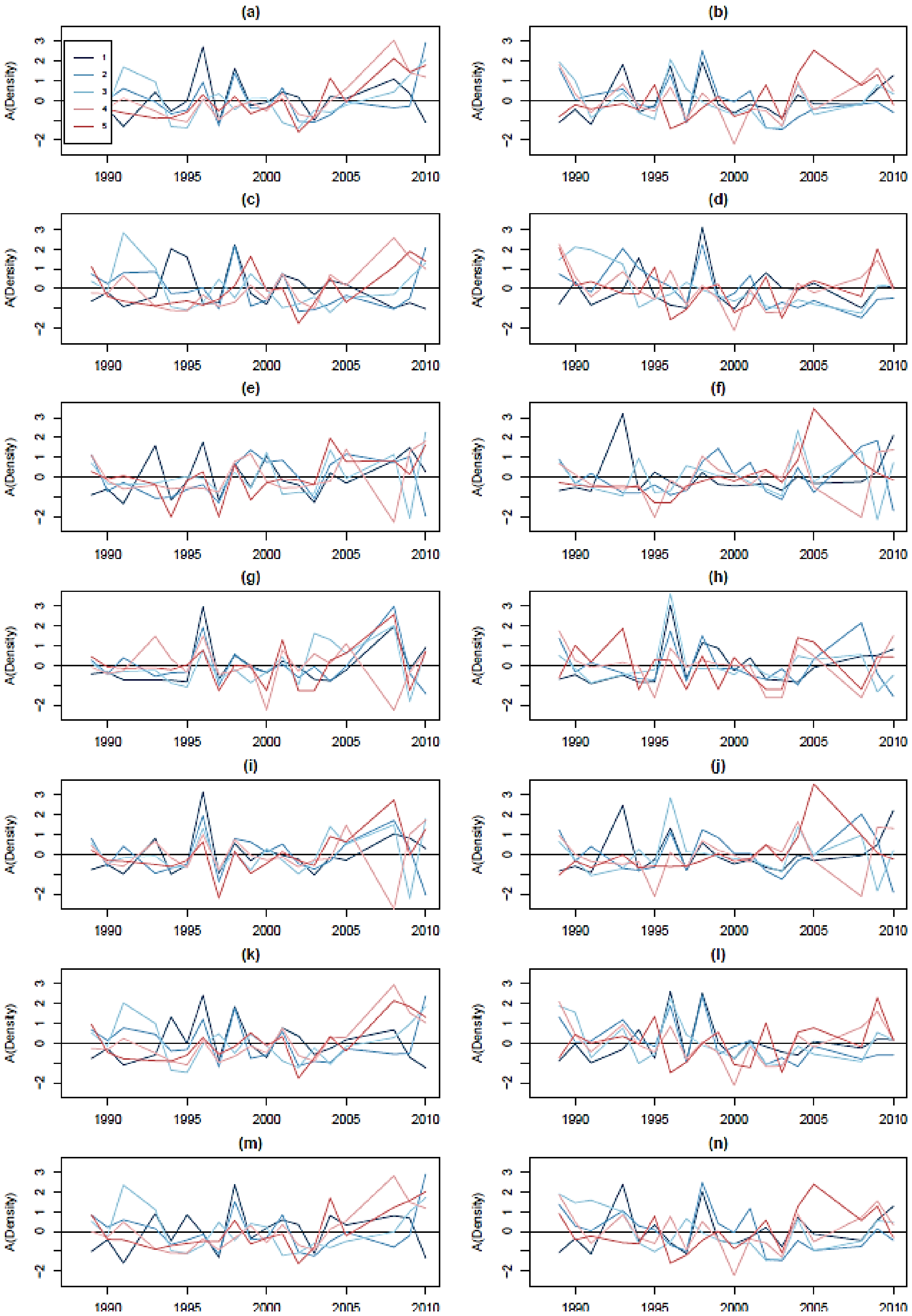

Figure. 2.4 Times series of density anomalies for common soles of ages 1 to 5 in the Eastern English Channel for cohorts between 1990 and 2010. Left and right panels correspond to female and male,

respectively. Individual panels correspond to the different spatial structuration hypotheses tested. Single stock (a,b); UK (c,d); NE (e,f); SW (g,h); NE + SW (i,j); UK + SW (k,l); UK + NE (m,n).

Correlation matrices between density-at-age time series per subunit for each hypothesis (**Fig. S.2.3**) are synthetized in **Fig. 2.5**. Whatever the hypothesis, process errors were close to 1 (between 0.02 and 2.1), allowing for interpretations.

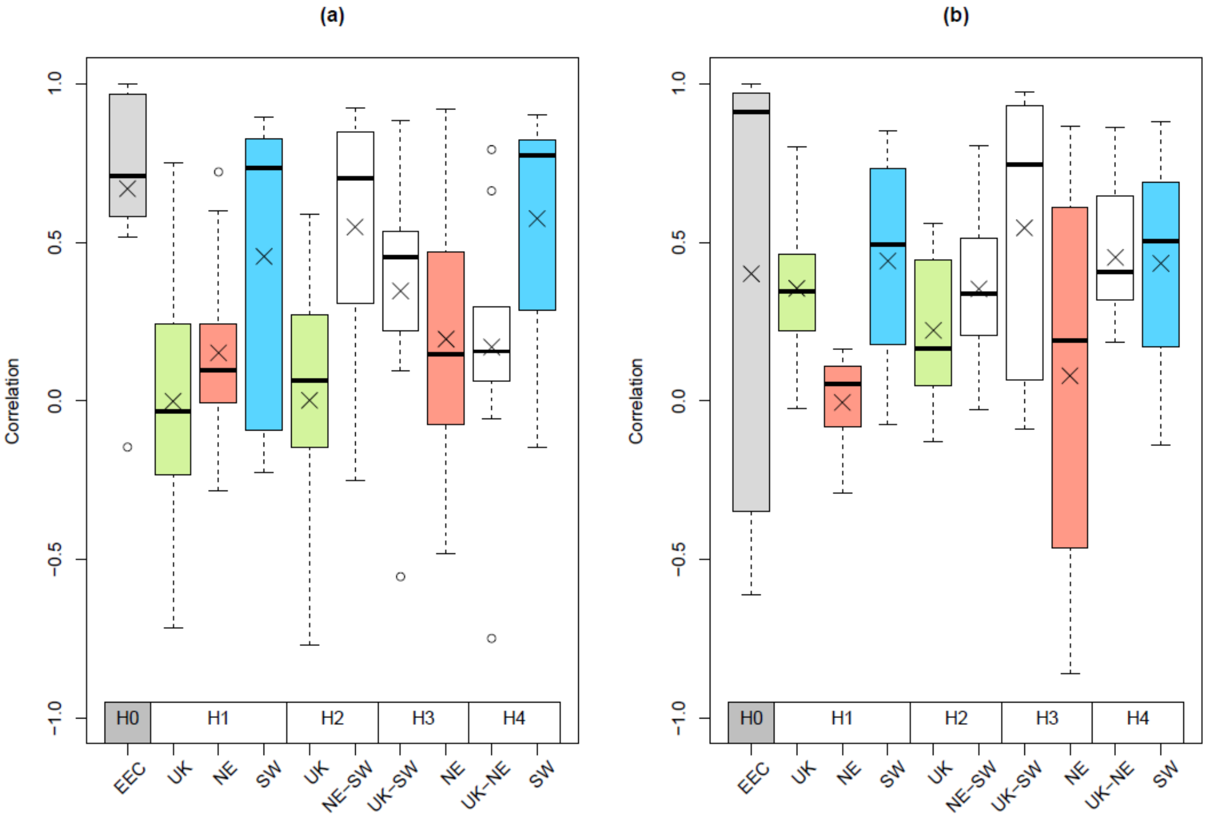

Figure. 2.5 Distributions of the correlations between density-at-age time series (ages 1-5, 10 pairwise correlations in each boxplot, crosses indicate the mean) for each subunit and spatial structuration hypothesis, for females (a) and males (b). Colors represent the subunits considered (in white when 2 subunits are pooled). H0 = single stock, H1 = 3 subunits (UK, NE and SW); 2 subunits for H2 = UK and (NE + SW), H3 = NE and (UK + SW) and H4 = SW and (UK + NE).

For both sexes, the strength of the synchrony across density-at-age time series was high considering the null hypothesis, i.e. no spatial structure (**Fig. 2.5**). Regarding females, the null hypothesis presented the highest level of synchrony among the structure hypotheses (**Fig. 2.5a**). Regarding males, the strength of the synchrony considering H0 was also high (**Fig. 2.5b**), but was not the highest. These results indicated that the null hypothesis (i.e. no spatial structure) was credible for both sexes and that mixing across the EEC stock could be important.

However, spatial structure was also probable for both sexes since the synchrony between density-at-age time series were also high within the SW subunit (H1 and H4, **Fig. 2.5**). Mixing between putative subunits appeared higher for other subunits, with low synchrony across density-at-age time series, especially in the UK subunit for females (H1 and H2; **Fig. 2.5a**), and in the NE subunit for males (H1 and H3; **Fig 2.5b**).

2.6. Discussion

The understanding of population structure is of major importance for the management of marine species (Kerr *et al.*, 2010a, 2010b; Ciannelli *et al.*, 2013). Inconsistencies between population structure and stock units may bias stock assessment results and the quantification of risks for sustainable fisheries management (Kerr *et al.*, 2017). In the EEC stock of common sole, spatial structure was shown for larvae (Rochette *et al.*, 2012) and juveniles (Le Pape and Cognez, 2016). For adults, the existence of spatial structure was also suggested by Du Pontavice *et al.* (2018), considering recent spatial contrasts in growth between the UK, NE and SW subunits. Herein, we expanded this study by assessing two different analyses of long-term growth and density to inform on this spatial structure.

2.6.1. Growth analysis revealed long-term stock structure

The von Bertalanffy growth modelling exhibited spatial stock structure for the common sole between the three predefined subunits in the EEC. This was consistent with Du Pontavice *et al.* (2018), who found higher length-at-age in the SW than in the NE and the UK subunits over the recent period 2010-2015, for both sexes. Herein, the analysis of long-term time series (1989-2015) highlighted a global decreasing trend of L_{10} over the last two decades, whatever sex and subunit. This finding corroborates the decreasing length-at-age trends in commercial landings since 2004 (ICES, 2017a). L_{10} in the SW remained the highest among subunits all along the study period, with dramatically higher inter-cohort variability. The common soles from the UK subunit presented similar L_{10} values as from the NE subunit before 2000, and higher values after 2000.

Growth is a phenotypic expression of genetic and environmental factors (Swain *et al.*, 2007; Cadrin *et al.*, 2013). Disentangling the relative effects of these factors is difficult for wild populations (Sinclair *et al.*, 2002). Growth is impacted by global change (Brunel and Dickey-Collas, 2010; Baudron *et al.*, 2014); as increasing sea temperature advantages early maturation and smaller body size. For the common sole of the EEC, changes of growth in response to global warming may explain the general decreasing trend of L_{10} observed over the last two decades. Indeed, increasing trends of water temperature is homogeneous throughout the EEC (Gohin *et al.*, 2010). Considering these homogeneous trends, contrasts in evolution of spatial growth between the UK and NE subunits could not be attributed to divergent environmental changes. Growth of the common sole is also impacted by intensity and size selectivity of the fishery (Mollet *et al.*, 2007) and fishery-induced evolution could have induced these contrasts in observed growth. Removing the larger individuals acts as a non-random genetic selection (Sinclair *et al.*, 2002; Law, 2007; Enberg *et al.*, 2012) and favor smaller size-at-age and early maturation (Enberg *et al.*, 2012). In the case of common sole of the EEC, contrasted size-selective fishing or exploitation rates between subunits could lead to spatial differences in growth trends (Mollet *et al.*, 2013; Hunter *et al.*, 2016) and is a relevant candidate mechanism to explain both the decreasing trend of length-at-age and the contrasted evolution of growth between subunits. Indeed, under the spatial hypothesis H1, Archambault *et al.* (2016) suggested contrasted estimates of fishing mortality between the UK, NE and SW subunits between 1980 and 2010: fishing mortality was the highest in the NE and the lowest in the SW subunit, with increasing difference between the UK and NE subunits after 2000. A lower fishing pressure in the SW might also have partly contributed to a lower selective pressure thus more variable growth potential and higher variability in growth in this area. Therefore, our findings may indicate a combined effect of global warming and fishery-induced evolution for the common sole of the EEC, although further investigations about the relative contribution of environmental factors and exploitation rate are required.

Despite these various trends, spatial contrasts in growth remained large during the whole period, especially between the NE and SW, revealing a long-lasting signal of spatial structure between these subunits (Erlandsson *et al.*, 2017). In addition to the analysis of spatiotemporal differences in growth parameters, we investigated the spatial structure through the synchrony analysis in L_{10} time series among subunits. Interpretation of differences in spatial synchrony of growth parameters was spurious because the observation and state process errors were high. In particular, process error was high in the SW subunit, with a probable link to the high inter-cohort variability of L_{10}. Besides, for growth parameters estimated at a multiyear time span, environmental drivers have potentially synchronous inter-annual variations between the subunits (Moran, 1953) thus have similar impacts on different cohorts (inter-annual autocorrelation in growth). Hence, synchronous variations in growth parameters are not a reliable indicator of the lack of spatial structure.

2.6.2. Density-at-age analysis highlighted isolation of the SW subunit

The assessment of covariability between abundance at different locations highlighted the potential existence of spatial structure in the EEC and was thus particularly valuable to describe the dynamic structure of subpopulations (Bjørnstad *et al.*, 1999; Botsford and Paulsen, 2000; Östman *et al.*, 2017). Whatever the sex, the synchrony analysis of density-at-age revealed a synchronous signal on the stock scale, but also at the SW subunit scale, and demonstrated a lack of synchrony in the UK for females and NE subunits for both sexes.

Understanding the mechanisms behind spatial synchrony (environmental stochasticity versus dispersal) remains a challenge in ecology (Liebhold *et al.*, 2004). Spatial synchrony in population dynamics may arise from two mechanisms (Liebhold *et al.*, 2004; Walter *et al.*, 2017), (1) dispersal among populations, and (2) congruent dependence of population dynamics on a synchronous exogenous factor. Dispersal among different subunits can lead to different levels of metapopulation structure, from very low connectivity between subunits to a single population (Östman *et al.*, 2017). Distinct demographic changes (i.e. high synchrony within subunits and low synchrony between subunits) may result from low dispersal rates and fine scale environmental processes, and synchronous demographic changes from high dispersal rates between subunits. In the case of the common sole of the EEC, concluding between a metapopulation and a single population from synchrony in cohort abundance was not possible although, the SW subunit appeared isolated from the others. The intra-subunit synchrony detected in the SW may result from low exchange rates with the other subunits of the EEC.

2.6.3. Congruence between analyses and perspectives

Stock identification methods may provide inconsistent results about the stock structure, some may detect stock structure where others fail to. Perception of spatial levels of stock structure might change with the method sensitivity and congruence between contrasting methods improves confidence (Cadrin *et al.*, 2013; Izzo et al., 2017). A multiple approach is hence preconized (Begg and Waldman, 1999; Waldman, 1999; Cadrin *et al.*, 2013) but remains underused (Pita *et al.*, 2016).

Here, we performed in parallel two distinct analyses of long-term life history parameters (growth and density-at-age) using survey data, underused for that purpose despite their easy access (Östman *et al.*, 2017). Our analyses were partially consistent to detect a lasting signal of stock structuration for common sole in the EEC. On the one hand, growth analysis highlighted long-term structure in three distinct subunits, previously defined (Rochette *et al.*, 2012; Archambault *et al.*, 2016). On the other hand, synchrony analysis of density-at-age time series underscored that two hypotheses of structure were probable, without concluding between the single stock hypothesis and spatial structure, with pronounced segregation in the SW subunit. Therefore, consistent evidence of spatial structure emerged with relatively strong indications of isolation of the SW subunit from the rest of the EEC. The SW subunit is physically isolated from the NE subunit and from the Western English Channel (ICES division VIIe) by rocky reefs forming a natural barrier (Rochette et al., 2010; **Fig.2.1**), and a deep central Channel covered by gravel separates the UK and SW subunits (Rochette et al., 2012). These unsuitable habitats for the common sole limit the migration of sub-adults and adults and isolate fish in the SW subunit. In addition to the low connectivity induced at the larval (Rochette et al., 2012) and juvenile stages (Riou et al., 2001; Le Pape and Cognez, 2016), our findings likely indicate low exchanges between the SW subunit and the rest of the EEC. For the two other subunits, isolation remains a question, suggesting higher exchange rates of sub-adults and adults between the UK and NE subunits, and potentially with the adjacent North Sea stock (ICES division IVc, **Fig.2.1**).

These assumptions have to be investigated in depth since a heterogeneous population dynamics among subunits could be inconsistent with the present assumption of the fishery assessment, which assumes a single stock. Archambault *et al.* (2016) examined the effect of a metapopulation structure on the estimates of the reference point for the common sole of the EEC. They suggested that such structure leads to contrasted sub-stock assessment, with full exploitation of the SW subunit but over-exploitation of the UK and NE subunits. If further investigations confirm the sub-stock segregation, it would be necessary to integrate their outcomes to improve the management of common sole in the EEC (Ulrich *et al.*, 2017) and to ensure the persistence capacity of populations (Heino *et al.*, 1997). At the same time, the decreasing trend of length-at-age has to be accounted for, since the consequences of smaller adult body length could reduce reproductive rates (Rijnsdorp *et al.*, 2010) and decrease population resilience (Hsieh *et al.*, 2006).

2.8. Conclusion of chapter

The second chapter represents a first step toward the understanding of the EEC stock structure of common sole. Interesting results emerged from a population-based approach associating growth and abundance analyses over 20 years.

Regarding the growth analysis, two main results emerged. **First, a decreasing trend of length-at-age was found over the time series, whatever the subunit considered**. Disentangling between the Fishery Induced Evolution theory and the climate change effect was impossible in this study and deeper investigations are required to answer this question. Recently, similar observations of smaller fish at age was described for the common sole in Danish waters (Boje *et al.*, 2019). Potential crossed effects of high fishing pressure and environmental changes are a credible hypothesis that has to be tested for maintaining sustainable exploitation of this flatfish species in the future across its distribution range. The second result that deserves to be underlined is the **long-lasting signal of growth differences between the SW, UK and NE subunits, suggesting long-term structuring of the stock** (**Fig. 2.6**). Despite that it was impossible to discriminate among the environmental and genetic factors influencing such differences, it suggested low exchanges between subunits, lasting at least over 20 years.

Results from the population growth analysis were partially in line with the results of abundance-at-age time series. Indeed, the **isolation of the SW subunit from the rest of the EEC stock was highly probable regarding the strength of the synchrony of abundance-at-age time series in this subunit** (**Fig. 2.6**). The presence of rocky reefs and high depth covered with gravels in the central channel between the SW subunit and the rest of the EEC is likely a factor driving the isolation of the SW.

Therefore, this second chapter highlighted the potential isolation of one subunit in the EEC and suggested that the stock structure was maintained over at least two decades which shed evidence of stock structure. However, this population-based approach shaped an overview of the stock structure at a large spatiotemporal scale and did not provide information at a fine spatial scales. Besides, this study did not assess the connectivity between nursery and spawning grounds. The next chapter proposes to complete the population-based approach by investigating fine spatial scale indices of spatial structure and connectivity using an individual-based approach (**Fig. 2.6**).

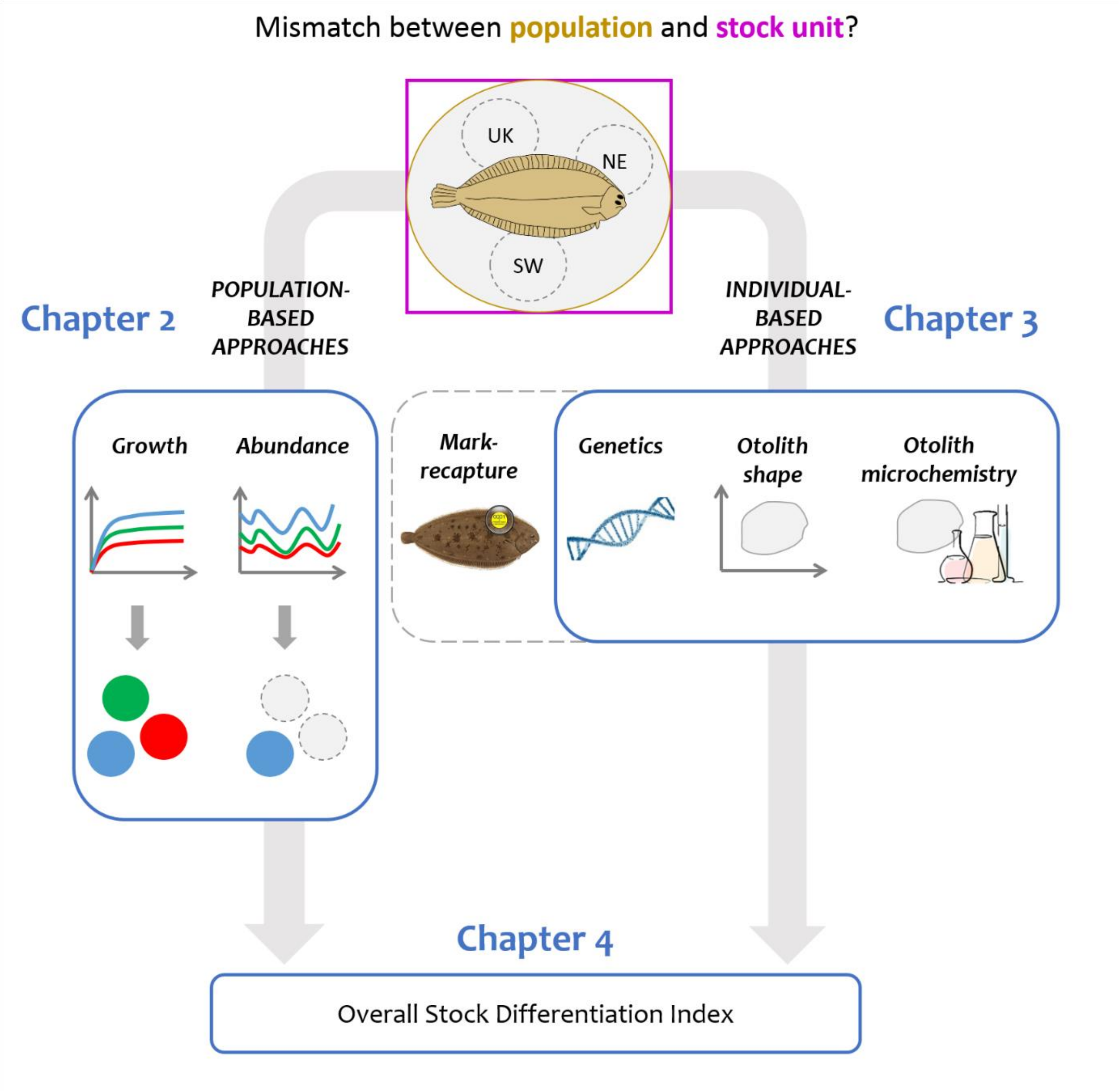

Figure. 2.6 Overview of the results of the common sole (*Solea solea*) population structure inside the Eastern English Channel stock (ICES division VIId) found in Chapter 2 of the thesis. The dotted line indicates that the mark-recapture analysis was not performed in this thesis but that the related findings of a previous study (Lecomte *et al.*, 2019) were included in chapter 4.

Chapter 3

Individual-based approaches to investigate the spatial structure and connectivity within the stock

Individual-based approaches

3.1. Introduction of chapter

In chapter 2, a long-lasting signal of spatial structure was evidenced by the common sole of the EEC. Spatial variations in growth parameters suggested a spatial structure made of three subunits, the SW, NE and UK. Additionally, high levels of synchrony among time series of abundance-at-age throughout the cohorts was found in the SW subunit, indicating its potential isolation from the rest of the stock. This population-based approach was a first step toward the understanding of the common sole population structure in the EEC. However, at this level it is not possible to conclude on the alignment or misalignment of the EEC stock unit with the common sole population. Indeed, information of the stock structure has to be completed with other tracers having various spatial and temporal resolutions. In Chapter 3, an individual-based approach is conducted through the use of genetic markers, otolith shape and otolith microchemistry analyses. Similarly to chapter 2, alternative hypotheses of spatial structure are tested.

Previous investigations of genetic differentiation using Single Nucleotide Polymorphisms (SNPs) demonstrated the isolation of the EEC from the adjacent North Sea and Western English Channel (Diopere *et al.*, 2018). However, the internal genetic structure of the EEC stock of common sole remains unknown, so far. Hereafter, genetic analysis consists of the investigation of spatial genetic differentiation using SNPs. Genetic analysis provides information of spatial structure at the evolutionary time scale which is necessary to understand the history of the stock structuring.

In parallel, the analysis of otolith shape allows for completing the understanding of population structure. Otolith shape is defined by environmental, ontogenetic and genetic factors (Cardinale *et al.*, 2004; Vignon, 2015) and could provide information of the stock structure at the lifespan scale (e.g. Hüssy *et al.*, 2016; Mahe *et al.*, 2016; Zhang *et al.*, 2016; Vasconcelos *et al.*, 2018).

Moreover, otolith microchemistry is a relevant method to assess connectivity between nursery and spawning grounds (Campana *et al.*, 2000; Gillanders, 2002a). The otoliths of fish grow continuously by incorporating chemical elements from the surrounding water and spatial variation of water chemical signatures imply related patterns in otolith chemical signatures. Hereafter, an atlas of juvenile otolith signatures is built to provide a baseline of otolith signatures in nursery grounds. By comparing the juvenile otolith composition of adults sampled on spawning grounds with the juvenile baseline, the reallocation of adults to their original nurseries is performed.

Finally, genetic and alternative tracers analyzed on the same individuals are combined in a single machine learning framework to assess the strength of population structure.

Combining genetic, morphological and chemical markers to assess population structure and trace connectivity within a flatfish stock

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3.2. Abstract

In the field of marine connectivity and stock identification, the current best practice is the combination of tracers that provide a holistic understanding of metapopulation structure. The common sole (*Solea solea*) of the Eastern English Channel (EEC) is currently assessed and managed as a single and homogeneous population. The presence of subpopulations has recently been suggested but has not been thoroughly investigated so far. Here, we hypothesized a metapopulation structure made of three subpopulations. We assessed connectivity by analyzing complementary natural tracers. First, we investigated population structure using up-to-date genetic markers (Single Nucleotide Polymorphisms). Also, we tested for spatial segregation of sub-populations using the adults' otolith shape as an integrative tracer of life history. In parallel, we assessed ontogenetic connectivity between the juvenile and adult stages of the otolith composition in trace elements. Finally, we combined the genetic, otolith shape and otolith microchemistry data sets in a supervised machine learning framework to assign probabilistically adults to subpopulations. Genetic assignments and otolith shape analyses provided congruent results and suggested a metapopulation structure. However, otolith microchemistry failed in assigning adults to their original nurseries because of the low discriminant capacity of juvenile signatures used as a baseline. The integrated analysis did not provide realistic reallocation probabilities because of the low number of fish available for the integrated approach. Our findings thus support the hypothesis of a metapopulation structure of three subpopulations, questioning the present management of the stock. The use of several tracers highlights the importance of a holistic approach to accurately conclude on the nature of stock structure. However, this study highlights the need for a large number of individuals, each analyzed for all the tracers, to prepare a reliable integrated assessment of stock structure.

Key-words: Otolith – SNP marker – Integration – Connectivity – Metapopulation

3.3. Introduction

Recent advances in population structure and connectivity of marine fish suggest that fine scale structuring is common in open habitats (Waples and Gaggiotti, 2006; Reiss *et al.*, 2009; Ciannelli *et al.*, 2013). The degree of connectivity varies along a continuum of population segregation, from complete mixing (i.e. panmixia) to full isolation (e.g. Smedbol and Wroblewski, 2002; Abaunza *et al.*, 2008; Cadrin *et al.*, 2010). Between these two opposite situations, metapopulations display a more or less consistent spatial structure through variable degrees of connection between subpopulations (Waples and Gaggiotti, 2006). Mechanisms underlying spatial structuring are mainly (i) biophysical processes involved in egg and larval dispersal patterns (Cowen and Sponaugle, 2009; Kerr *et al.*, 2017b) and (ii) post-larval (i.e. juvenile, sub-adult and adult) movements related to homing or straying behavior, spawning and nursery site fidelity or migration strategies (Secor, 2015). More interesting, the paradigm suggesting that larval dispersal acts as the main driver of population structure and connectivity (e.g. Cowen and Sponaugle, 2009) was reevaluated and the importance of adult-mediated connectivity was put forward (Mullon *et al.*, 2002; Frisk *et al.*, 2014). Hence, understanding of population structure and connectivity is a challenging and crucial issue depending on the life cycle and environmental conditions.

A wide range of methods exist to address the structure and connectivity of marine populations (Cadrin *et al.*, 2013; Östman *et al.*, 2017). Insights into population segregation can be provided, among others, by larval dispersal modelling (e.g. Savina *et al.*, 2016; Stuckas *et al.*, 2017), mark-recapture experiments (e.g. Le Bris *et al.*, 2018; Lecomte *et al.*, 2019), morphometry and meristics (e.g. Allaya *et al.*, 2016; Sley *et al.*, 2016), otolith shape (Hüssy *et al.*, 2016; Mahé *et al.*, 2016) and microchemistry (e.g. Tanner *et al.*, 2016; Moreira *et al.*, 2018). These tools make it possible to estimate population structure over time and space across the full life cycle (Tanner *et al.*, 2016). In addition, population structure can be informed at the evolutionary time scale through approaches that use genetic markers such as microsatellites or Single Nucleotide Polymorphisms (SNPs) (Saenz-Agudelo *et al.*, 2009; Mullins *et al.*, 2018). In general, limited exchanges of individuals suffice to maintain genetic homogeneity, so that genetic approaches sometimes fail in detecting population segregation over an evolutionary time scale (Hawkins *et al.*, 2016). However, genetic markers are useful to detect population structure at a fine spatial scale because of the high power to detect subtle population differentiation (Morin *et al.*, 2009). Therefore, population structure and connectivity studies would undoubtedly gain from combining genetic and alternative methods that have complementary biological, spatial and temporal resolutions. There are an increasing number of studies that aim at combining genetic and alternative analyses (e.g. Smith and Campana, 2010; Tanner *et al.*, 2014; Martin *et al.*, 2015; Marengo *et al.*, 2017; Reis-Santos *et al.*, 2018). The annual review of stock identification methods showed that those studies using interdisciplinary approaches are consistently increasing (ICES, 2018c). However, most couple only one tracer (generally otolith microchemistry) with genetic markers, thus limiting the interpretation of the results. Indeed, genetic markers sometimes fail to detect population structure and in this case, conclusions would be drawn on a single non-genetic tracer. Also, analyses on different tracers are often performed on different individuals, thus challenging the interpretation of the results (Waldman *et al.*, 1997; Cadrin *et al.*, 2014). Performing several analyses on the same individuals allows for assessing the congruence and discrepancies between tracers, and thus informs about the performance of each tracer.

The stock of common sole (*Solea solea,* Linnaeus, 1758) living in the Eastern English Channel (EEC; ICES division VIId; **Fig. 1**) is genetically distinct from the two adjacent stocks of the Southern North Sea (ICES division IVc) and the Western English Channel (ICES division VIIe) (Diopere *et al.*, 2018). However, the structure of this stock at a fine-spatial scale, i.e. within the EEC, has been questioned (Rochette *et al.*, 2013; Archambault *et al.*, 2016; ICES, 2017a) but the understanding of connectivity at such scale remains fragmentary. According to hydrodynamic modelling, larval connectivity is very low across the EEC stock, with spawning grounds provisioning the closest nursery grounds (Rochette *et al.*, 2012). High sedentariness of juveniles in nursery grounds also induces low connectivity at this stage (Riou *et al.*, 2001; Rochette *et al.*, 2010; Le Pape and Cognez, 2016). However, adult-mediated connectivity is poorly documented (Burt and Millner, 2008; Archambault *et al.*, 2016) and putative subunits within the EEC sole stock have been suggested (Rochette *et al.*, 2013; Archambault *et al.*, 2016). Recent studies tested for the spatial structure of the EEC stock of sole using life history traits of adults in population-based approaches (Du Pontavice *et al.*, 2018; Randon *et al.*, 2018). Three putative subunits of the EEC stock were hypothesized: the Bay of Seine (southwest subunit, SW), the northern French coast (northeastern subunit, NE) and the southern English coast (English subunit, UK; **Fig. 1**). Estimation of von Bertalanffy growth parameters highlighted long-term differences between these spatial subunits in the EEC (Randon *et al.*, 2018). In addition, density-at-age analysis suggested the isolation of the SW subunit from the rest of the stock and highlighted a long-lasting signal of spatial structure that remains to be thoroughly investigated.

In this study the former population-based approaches were complemented by focusing on fine-scale population structure and connectivity using an individual-based approach. More precisely, genetic connectivity was investigated using SNP genotypes of adults to assess population structure over an evolutionary timescale. In parallel, ecological connectivity was investigated by using analyses of otolith shape and microchemistry. The external outline of the otoliths allowed examining the population structure through subunit detection whereas otolith microchemistry aimed at providing estimates of connectivity linking spawning and nursery grounds. Finally, genetic and otolith tracers were combined using a supervised machine learning approach to test for their congruence on the bulk of adult individuals analyzed for the three tracers.

3.4. Material and methods

3.4.1. Spatial structure hypotheses

Several hypotheses of spatial structure were considered for the analyses of otolith shape and genetic structure. The null hypothesis H0 corresponded to a homogeneous population without spatial structure (i.e. the current assessment and management unit; **Fig.3.1**). Four alternative hypotheses of spatial structure were also tested: H1 = SW, NE and UK subunits separated, H2 = SW and NE pooled and separated from the UK, H3 = SW and UK pooled and separated from the NE, H4 = UK and NE pooled and separated from the SW.

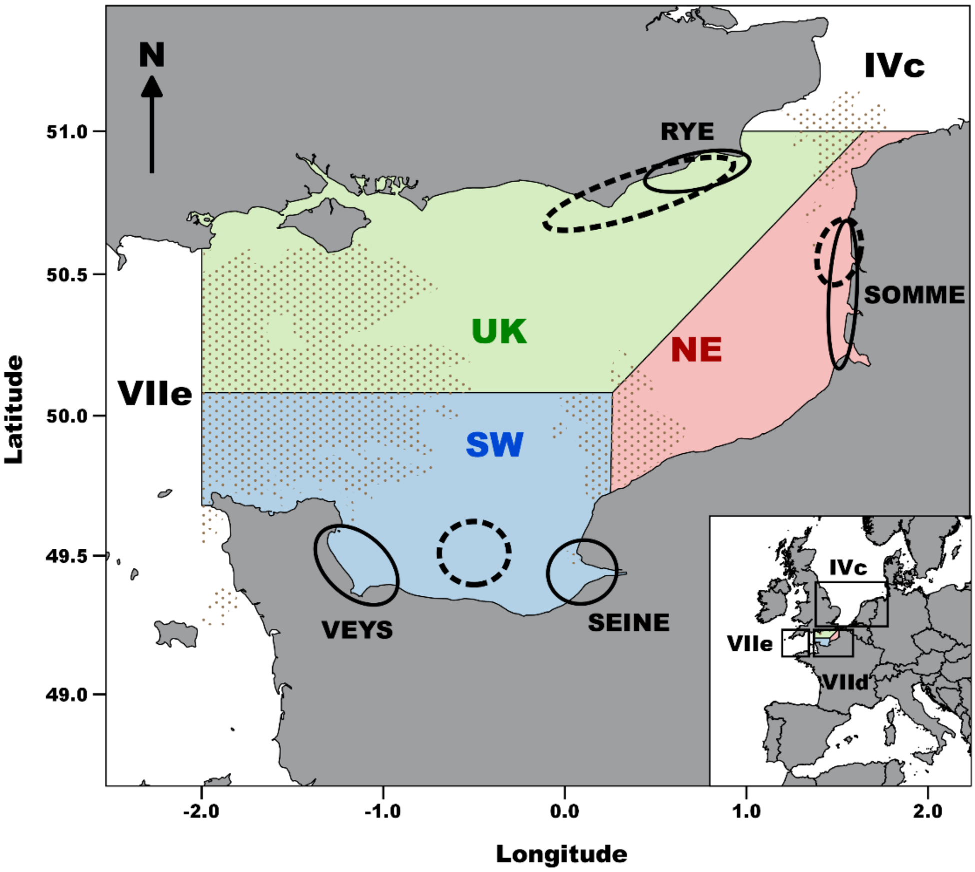

Figure. 3.1. Map of the sampling sites of common sole (*Solea solea*) in the three putative subunits (SW, NE and UK) of the Eastern English Channel stock (VIId). Solid and dashed black ellipses show the sampling location of juveniles in the nurseries and adults on the spawning grounds, respectively. Light grey dots are rocky reefs. VE = Bay of Veys, SE = Bay of Seine, NE = Bay of Somme, and UK = Rye.

3.4.2. Sample collection

The otolith shape and genetic analyses were based on adult individuals to investigate population structure. The otolith microchemistry analysis was composed of a juvenile baseline and an adult data set to assess connectivity between nursery and spawning grounds. The extent of the data sets varied depending on the tracers but a subset of adult individuals were analyzed for the three of them, aiming at combining the tracers in an integrated analysis (**Table 3.1**).

Soles were collected during scientific surveys or on professional fishing ships during tagging experiments (Lecomte *et al.*, 2019). After sex determination, total length was measured (in cm), sagittal otoliths were removed, and a caudal fin clip was sampled and stored in pure ethanol for genetic analysis. After picturing both otoliths for shape analysis, the right sagittal otolith was used for age determination and the left one for microchemistry.

Adult soles were sampled on the spawning grounds from April to May of 2017 and 2018 (**Fig. 3.1**, **Table 3.1**). There was an exception in 2018, when adults from the English part of the EEC were collected from the UK Beam Trawl Survey during summer since no sampling opportunity was available during the spawning period. Among the collected individuals, genetic sampling was performed on a larger number of adult individuals (n = 215) (**Table 3.1**) than otolith sampling for microchemistry and shape analyses (n = 157). Otolith sampling for shape analysis was then completed with additional otoliths (n= 602) collected from fish markets every month between 2016 and 2018 (**Table 3.1**). Overall, adults were estimated to be between 3 and 9 years-old, corresponding to cohorts 2009 to 2015.

For the microchemical analysis needs, additional juvenile soles (n = 137) were sampled on the nursery grounds in July and September 2016 and 2017 (**Fig. 3.1**, **Table 3.1**). Age-0 (length ranges: 4.70-15.5 cm; mean ± sd = 9.95 ± 1.90) and age-1 (length ranges: 10.5-18.0 cm; mean ± sd = 16.2 ± 1.52) juveniles were selected to build a baseline of the nurseries, corresponding to cohorts 2015 to 2017.

Table 3.1. Number of adult and juvenile individuals sampled within each subunit of the EEC used for otolith microchemistry (Nm), otolith shape (Ns), genetic (Ng) and combined (Nc) analyses.

Stage	Subunit	Nm	Ng_{2017}	Ng_{2018}	Ns	Nc_{2017}	Nc_{2018}
Juvenile	SW (VE + SE)	81 (24 + 57)	-	-	-	-	-
	NE	45	-	-	-	-	-
	UK	11	-	-	-	-	-
Adult	SW	42	47	12	522	11	7
	NE	64	31	42	139	13	22
	UK	51	42	41	98	34	4

3.4.3. Genetic analysis

DNA extraction from fin clips followed Cruz *et al.* (2017). Double digest restriction-site associated DNA (ddRAD) was performed (Peterson *et al.*, 2012) and fragments were amplified by PCR prior to paired-end sequencing. Samples from 2017 and 2018 were sequenced and independent libraries were built.

De novo assembly was performed using the *dDocent* variant calling pipeline (Puritz *et al.*, 2014) after demultiplexing. More details about the molecular, sequencing protocols, *de novo* assembly and SNP calling are available in **Appendix A**.

Regarding the library of 2017, after demultiplexing, 421 390 451 reads were available among which 20 995 bi-allelic SNPs were retained by the SNP calling. Regarding the library of 2018, 234 348 163 reads were obtained, resulting in 67 169 bi-allelic SNPs. These SNPs were filtered following several criteria of allelic depth, allelic balances, allelic frequencies, occurrence over all individuals, minimum heterozygosity threshold, Hardy-Weinberg Equilibrium (HWE) and threshold of linkage disequilibrium (LD). Information on the filters are provided in **Appendix A**. Considering that the ddRAD experiments were not reproducible between years, independent analyses of libraries were conducted. In 2017, 2 902 SNPs were retained for 120 individuals after SNP filtering. In 2018, the number of SNPs decreased to 435 for 95 fish.

Global and pairwise F_{ST} (Weir and Cockerham, 1984) were evaluated using the *hierfstat* R package (Goudet, 2005). Significance of pairwise F_{ST} was obtained from 95% confidence intervals (i.e. 95% CI). F_{ST} values were estimated for each hypothesis of spatial structure (H1, H2, H3 and H4). Finally, a Discriminant Analysis of Principal Components (DAPC) was computed with the *adegenet* R package (Jombart, 2008). Details about computational options are given in **Appendix A**.

3.4.4. Otolith shape analysis

After removing and cleaning sagittal otoliths from the fish, they were scanned with the sulcus facing up under reflected light with high resolution (3200 dpi). Individual images were extracted with the *TNPC* software (www.tnpc.fr). Five shape indices computed using four otolith size measures, namely length L_0 (i.e. the longest distance along the antero-posterior axis), width l_0 (i.e. the longest distance along the ventro-dorsal axis), perimeter P_0 and area A_0, were used to test for spatial differences (Tuset *et al.*, 2003) (**Table 3.2**).

Table 3.2. Shape indices as a function of otolith size measures (Tuset *et al.*, 2003). L_0, l_0, P_0 and A_0 are the length, width, perimeter and area of otoliths, respectively.

Shape indices	Formulae
Ellipticity	$(L_0-l_0)/(L_0+l_0)$
Circularity	P_0^2/A_0
Rectangularity	$A_0/(L_0\times l_0)$
Roundness	$(4A_0)/(\pi L_0^2)$
Form coefficient	$(4\pi A_0)/P_0^2$

Redundancy between shape indices was tested using the Pearson correlation test. Negative correlations were found between circularity and form coefficient (r = -0.99, p < 0.001) and between ellipticity and roundness (r = -0.89, p < 0.001). Each shape index was kept included in later discrimination analyses because they were not correlated with more than one index. Also, after checking for normality and homogeneity, residuals of linear models were used for ellipticity and roundness indices to account for their significant relationship with total fish length. Analysis of variance and post-hoc tests were conducted to test for spatial differences according to the five alternative hypotheses H0 to H4 in each shape index separately. Then, Linear Discriminant Analyses (LDA) based on all shape indices were performed for each hypothesis of spatial structure using the *MASS* R package (Ripley *et al.*, 2013). Total reclassification success was tested using jack-knife cross-validation. Finally, Wilks'λ (i.e. the ratio between intra-class variance and total variance) was used to assess the quality of reclassification results using the *rrcov* R package (Todorov and Filzmoser, 2009).

Complementary elliptical Fourier descriptors and directional bilateral asymmetry analyses were conducted and are presented in **Appendix B**.

3.4.5. Otolith microchemistry analysis

Left sagittal otoliths were prepared following the protocol of Martin *et al.* (2015) for microchemistry analysis conducted by LA-ICPMS. Triplicate spots were sampled using laser ablations in the first translucent zone corresponding to the first summer spent in nursery ground (**Appendix C**). Elemental concentrations in ^{7}Li, ^{23}Na, ^{43}Ca, ^{55}Mn, ^{59}Co, ^{63}Cu, ^{66}Zn, ^{85}Rb, ^{88}Sr, ^{107}Ag, ^{111}Cd, ^{138}Ba, ^{208}Pb, ^{238}U were measured relatively to Ca (µg.g^{-1}). Standardization procedure and limits of detection calculation are provided in **Appendix C**. Signatures (i.e. elemental concentrations) were obtained from the mean concentrations over the three ablations (**Fig. C.2**) and were log-transformed when needed to achieve normal distribution and homoscedasticity of variances. Overall spatial differences in the elemental concentrations of the nurseries were investigated using multivariate analysis of variance (MANOVA) and differences between nursery grounds and cohorts were analyzed using an ANOVA and Tukey post-hoc tests. Since interannual variations of the baseline could bias the reallocation of adult individuals to their nurseries of origin (Gillanders, 2002b), analyzing a cohort effect on the elemental compositions was needed. The analysis of temporal variability of the juvenile baseline composed of age-0 and age-1 juveniles, corresponding to cohorts 2015-2017, and adults corresponded to cohorts 2009-2015 allowed the selection of microchemical tracers that were temporally stable.

Elemental fingerprint indices (EFI) were computed to measure the similarity between two otolith compositions (Moll *et al.*, 2019) and ultimately highlight overlaps between signatures of nurseries. For fish i and j, $EFI_{i,j}$ was defined by Equation1.

$$\boldsymbol{EFIi,j} = \frac{2\times\sum_{x=1}^{n}\min(Cxi;Cxj)}{\sum_{x=1}^{n}(Cxi+Cxj)} \qquad \textit{(Equation 3.1)}$$

with n the number of elements considered and Cx the elemental concentration of element x.

This overlap index ranged between 0 and 1. A value of 0 indicated that fish i and j have different otolith signatures whereas a value of 1 meant similar otolith signatures.

Based on a Bayesian model (Randon *et al.*, 2017; **Appendix C**), adult individuals were probabilistically reallocated to their nurseries of origin using the juvenile baseline. At each iteration of the reallocation process, soles were reallocated to one nursery by comparing their juvenile composition (i.e. the composition of the part of the adult otolith bio-mineralized during the juvenile stage) with the juvenile baseline. At the end of the reallocation process, frequencies of reallocation were calculated with highest frequencies (F_{max}) interpreted as a high confidence in assignment. The final aim was to estimate the connectivity between juvenile and adult habitats by comparing their estimated origin with their subunit of sampling.

A preliminary validation procedure was necessary before interpreting the reallocation of adults since it directly depended on the discriminatory capacity of the juvenile baseline. To do so, the model was run 100 times. At each run, one juvenile per nursery was randomly removed from the juvenile dataset and introduced in the adult dataset to mimic an adult individual. These samples were then reallocated to nurseries at each run of the Bayesian model. The predicted nurseries of origin were compared to the “true” (i.e. known) nursery of origin of the fish (i.e. the sampling location). Finally, the percentage of reclassification success in each nursey was calculated based on the 100 runs.

3.4.6. Integrated analysis

An integrated analysis of individual tracers was performed using the *assignPOP* R package (Chen *et al.*, 2018) that allows combining genetic and alternative tracers to estimate the membership probabilities of adults to the three putative subunits. This package provides a machine learning framework whose principle is to assign individuals from different populations by dividing the entire dataset in training and test datasets and building a user-chosen machine learning classification function. The predictive model is then applied to all unknown individuals to reallocate them to their population of origin in a probabilistic way. The predictive model was built using Linear Discriminant Analysis of the *MASS* R package. Assignment accuracy of the training data sets were provided by the Monte Carlo cross-validation procedure. Membership probabilities were estimated using the K-fold cross-validation method. The integrated analysis was performed on individuals that had been analyzed for the three types of individual tracers with genetic data from 2017 only (higher number of samples in 2017 than 2018; **Table 3.1**).

3.5. Results

3.5.1. Genetic analysis

Global genetic structure was weak in 2017 and 2018 (low F_{ST} values) but statistically significant (**Table 3.3**). Pairwise F_{ST} comparisons between subunits revealed distinct genetic pools of individuals between the three subunits (**Table 3.3**), whatever the hypothesis of spatial structure considered (**Table A. 1**).

Table 3.3. Pairwise F_{ST} values and the corresponding 95% confidence interval (upper and lower limits) for the hypothesis H1 of spatial structure (SW, NE and UK subunits). '*' indicate significant values.

Year	Spatial comparison	Lower limit of 95% CI	F_{ST} value	Upper limit of 95% CI
2017	SW/NE	0.0019	0.0031*	0.0043
	SW/UK	0.0030	0.0044*	0.0060
	UK/NE	0.0045	0.0058*	0.0074
	Global	0.0035	0.0045*	0.0063
2018	SW/NE	0.0004	0.0031*	0.0058
	SW/UK	-0.0037	0.0028	0.0108
	UK/NE	-0.0033	0.0024	0.0095
	Global	0.0008	0.0029*	0.0132

These results were confirmed by the DAPC conducted on samples from 2017 and 2018 separately (**Fig. 3.2**). 30 PCs and 20 PCs were retained for 2017 and 2018, respectively. Weak overlap between subunits was observed, especially for 2017, supporting spatial genetic variation.

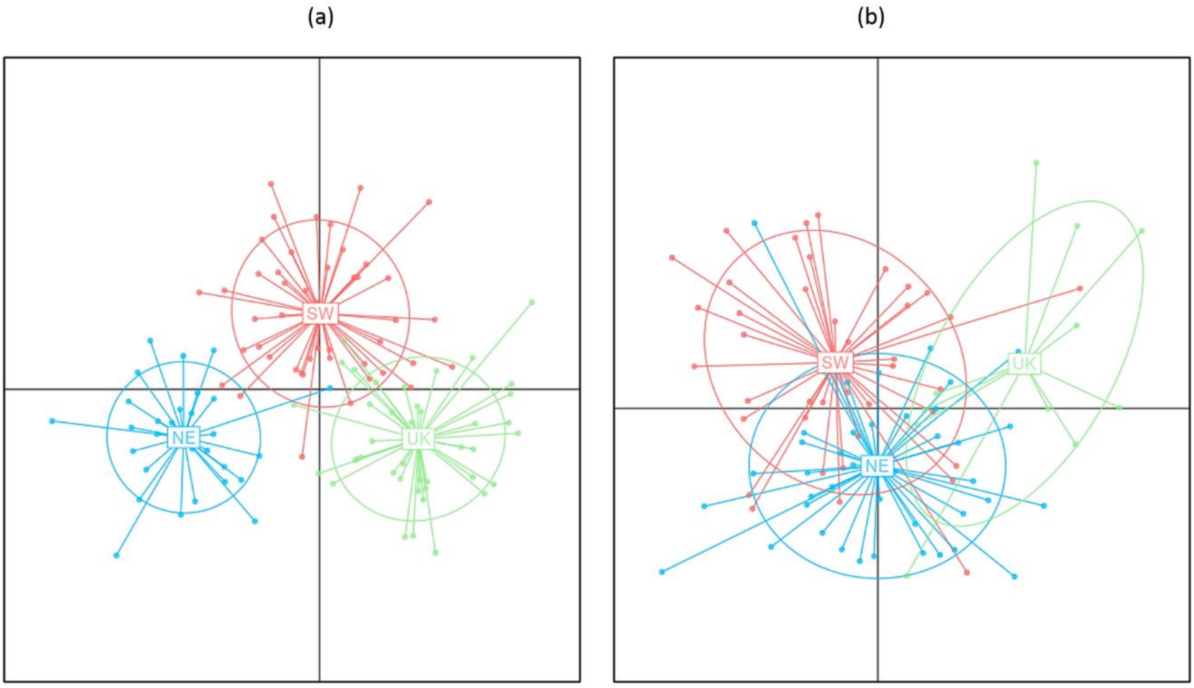

Figure. 3.2. Discriminant Analysis of Principal Components performed on the SNP genotypes of soles collected in 2017 (a) and 2018 (b).

Finally, using the *assignPOP* R package, a consistent signal of spatial structure was found for data from 2017 (**Fig. A.1 and Fig A.2**).

3.5.2. Otolith shape analysis

Regarding hypothesis H1, analysis of variance indicated that shape indices varied significantly between subunits (**Fig. B.1**), except rectangularity (**Table 3.4**). Based on *post hoc* tests, the SW and NE subunits presented significant differences in ellipticity, circularity, roundness and form coefficient. UK and SW subunits differed significantly in ellipticity, roundness and form coefficient. Similar signal of spatial structure was found under H3 and H4 (**Table B.1**).

Table 3.4. Mean comparisons (± SD) of shape indices between subunits using ANOVA for spatial structure hypothesis H1. Stars indicate significance. Left and right otoliths were pooled. Statistical significance: '***' P < 0.001; '**' P < 0.01; '*' P < 0.05

Shape indices	Mean (± SD)			DF	F	P
	SW	NE	UK			
Ellipticity	0.117 (± 0.025)	0.117 (± 0.022)	0.120 (± 0.025)	2	24.56	<0.001 ***
Circularity	14.2 (± 0.318)	14.1 (± 0.315)	14.1 (± 0.333)	2	4.424	0.01 *
Rectangularity	0.767 (± 0.017)	0.766 (± 0.017)	0.766 (± 0.019)	2	0.333	0.452
Roundness	0.773 (± 0.037)	0.771 (± 0.038)	0.767 (± 0.038)	2	24.67	<0.001 ***
Form coefficient	0.888 (± 0.018)	0.894 (± 0.019)	0.892 (± 0.021)	2	6.892	0.01 *

Whatever the otoliths used (left, right or both otoliths) for the LDA, reclassification success ranged between 47 and 63% and Wilks' λ ranged between 0.89 and 0.96, which suggested relatively poor discriminatory power of shape indices (**Table B.2**). However, lower values of Wilks' λ and higher reclassification success were found for hypotheses H1 and H4, indicating again a spatial structure.

Complementary analyses (**Appendix B**) led to contrasting findings. Otolith bilateral asymmetry confirmed spatial structure between the three subunits (H1) and especially evidenced contrast in the NE (H3). However, elliptic Fourier descriptors did not evidence spatial contrasts.

3.5.3. Otolith microchemistry analysis

- **Analysis of the juvenile baseline**

Among the 14 elements analyzed, Li, Na, Mn, Sr and Ba were detected in more than 90% of otoliths and were kept in the following analyses (**Table C.1**). Only Ba concentration was log-transformed to achieve normality.

Multi-elemental signatures differed significantly between nurseries (MANOVA: Pillai's trace = 0.72188, F = 8.0322, $p < 0.001$). Differences in elemental concentrations between nurseries were found for all elements (**Table C.2**) and a cohort effect was highlighted for Ba and Na, indicating that these two elements were not temporally stable. The percentages of variance explained by the nursery and cohort effects revealed that temporal effect was low for Ba (31% and 12% of the total variance, respectively) but was major for Na (12% and 30% of the total variance explained by the spatial and temporal effects respectively). Therefore, in the following spatial analysis, only elements Li, Mn, Sr and Ba were kept (**Fig. C.2**).

Analysis of EFI indicated that juvenile otolith signatures were similar within nurseries (average EFI = 0.919). Highest mean EFI were found within the SE and within the UK nurseries and the lowest EFI values within the NE nursery.

Comparisons of otolith signatures between nurseries revealed high values of mean EFI (i.e. similar signature), between UK and VE. Conversely, low EFI were found between the SE and NE nurseries, indicating fewer overlapping signatures (**Table 3.5**).

Table 3.5. Comparisons of mean signature similarity index EFI and standard deviation (± SD) between nurseries.

	VE	SE	NE	UK
VE	-	0.922 (±0.054)	0.918 (±0.063)	0.934 (±0.047)
SE	-	-	0.903 (±0.074)	0.906 (±0.058)
NE	-	-	-	0.923 (±0.057)
UK	-	-	-	-

Overlaps in juvenile otolith signatures were confirmed by the average expected signatures of nurseries predicted by the Bayesian model of reallocation (**Fig. 3.3**).

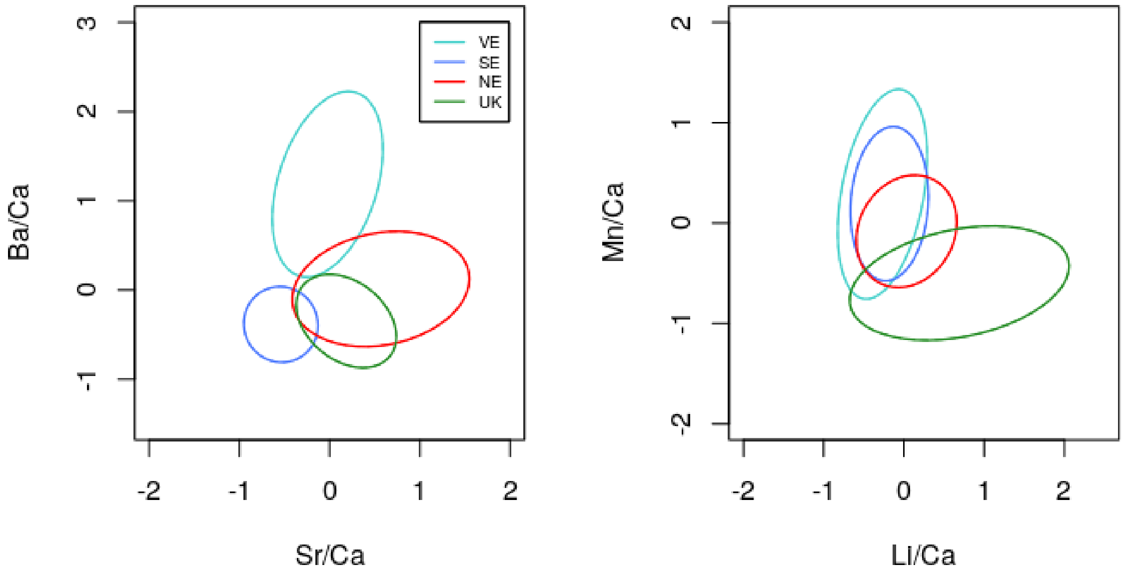

Figure. 3.3 Otolith elemental compositions of nurseries predicted by the Bayesian model. Concentrations were centered and scaled. Ellipses represent 75% confidence intervals.

- **Model validation**

Reclassification success obtained from the validation procedure was consistent with the EFI analysis (**Fig. 3.4.a**). Indeed, juveniles from the VE and SE nurseries were well reassigned with 75% and 59% mean reclassification success, respectively, which was not the case for individuals from the NE and UK nurseries (33% and 25% mean reclassification success, respectively).

- **Reallocation of adults**

Among adult individuals, 59% were reallocated in the SW subunit of the EEC (i.e. VE and SE nurseries), 21% in the UK and 20% in the NE nursery. These unbalanced proportions of fish reallocated in the SW nurseries were observed in each subunit (**Fig. 3.4.b**). However, the poor discriminant capacity of the juvenile baseline and the low reclassification success in the NE and UK nurseries limits the robustness of adult reallocation.

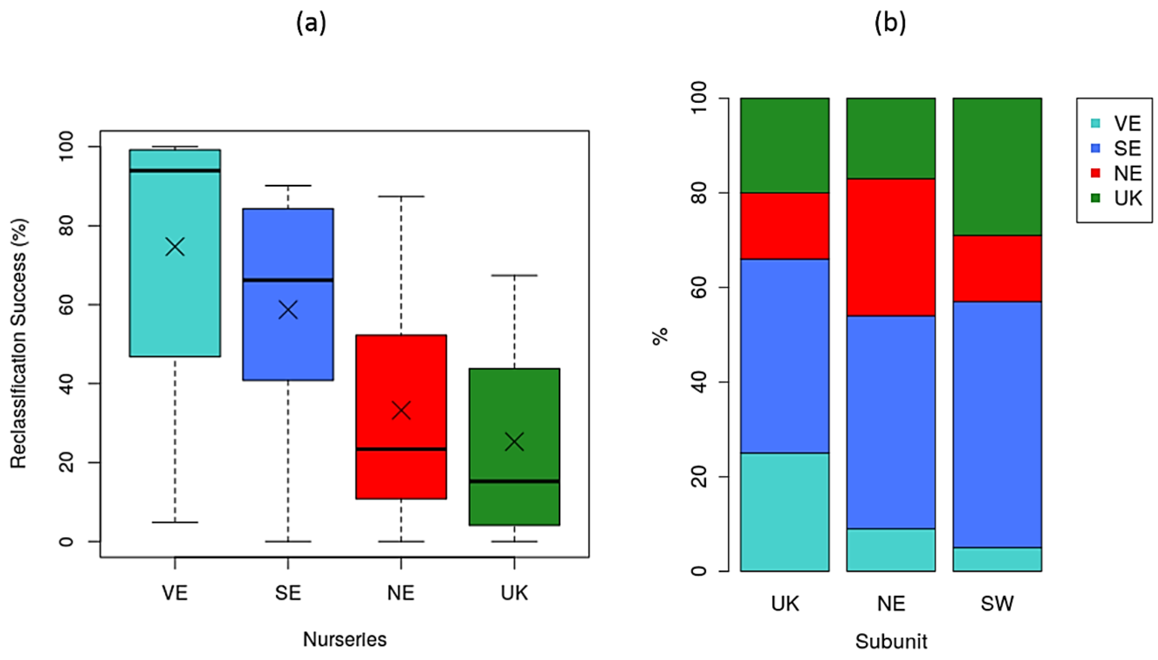

Figure. 3.4. (a) Reclassification success of juveniles to the nurseries. Crosses are the means of the distributions. (b) Proportion of adults reallocated in nurseries estimated by the Bayesian model relatively to their subunit of sampling.

3.5.4. Integrated analysis

The input data set included 58 individuals, each analyzed for otolith shape, otolith microchemistry and genetic markers in 2017. To highlight the differences when combining the tracers, the genetic dataset was first analyzed separately and then otolith shape and microchemistry datasets were introduced. Whatever the type of data used (only genetic or integrated datasets), the proportions of training datasets (50%, 70% or 90% of individuals) and the proportions of loci used (10%, 25% or 50% of the highest F_{ST} or all loci), assignment accuracy was low in the SW and NE subunits (below the random probability of 0.33). However, assignment accuracy in the UK subunit was high and outperformed the overall situation (**Fig. 3.5.a**). Combining otolith shape and microchemistry datasets with genetic data increases the overall, SW and NE assignment accuracy (**Fig. 3.5.b**). However, assignment accuracy remained below 0.33 for samples of the SW and NE subunits (**Fig. 3.5.b**).

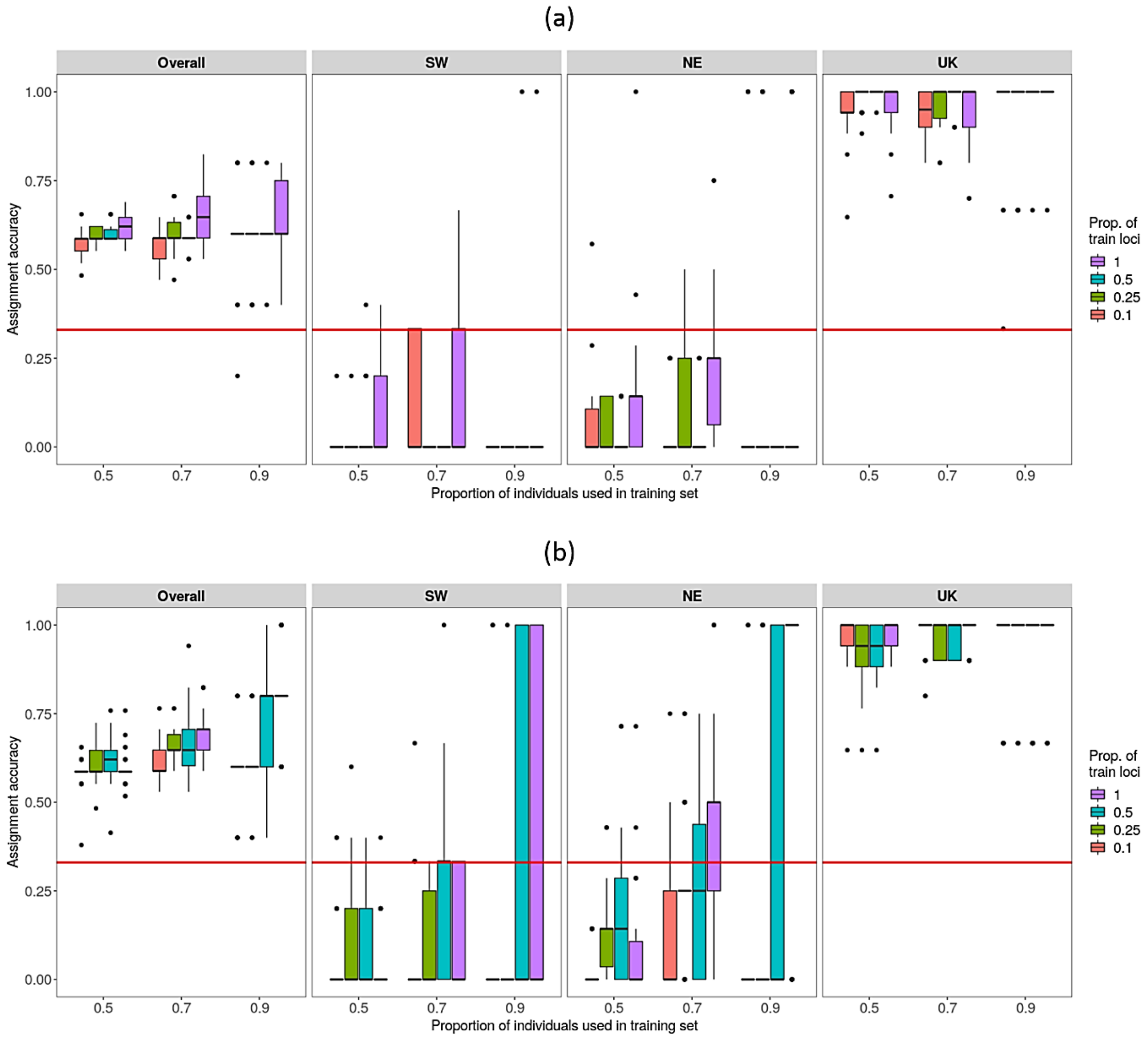

Figure. 3.5. Assignment accuracy estimates performed by Monte Carlo cross-validation and support vector machine methods using (a) only genetic and (b) genetic, otolith shape and microchemistry datasets. Training datasets were composed of four proportions of training loci (top 10% in orange, 25% in green and 50% in blue of the highest F_{ST} loci and all loci in purple), four shape indices and four otoliths elemental measurements. Three subsets of the datasets were considered for training (0.5%, 0.7% and 0.9%) and 30 iterations were computed. The horizontal red line represented the null subunit assignment rate (0.33 in the case of three subunits).

The low assignment accuracy in the SW and NE in contrast to the UK contributed to the low capacity of the model to reallocate individuals. When keeping all genetic loci, a weak signal of spatial structure was found (**Fig. 3.6.a**). When combining the genotypes with otolith shape and microchemistry data sets, spatial structure was lost. Indeed, whatever the subunit of sampling, individuals were assigned to the NE and SW with mean probabilities of 0.16, whereas they were assigned in the UK with probabilities above 0.5 (**Fig. 3.6.b**).

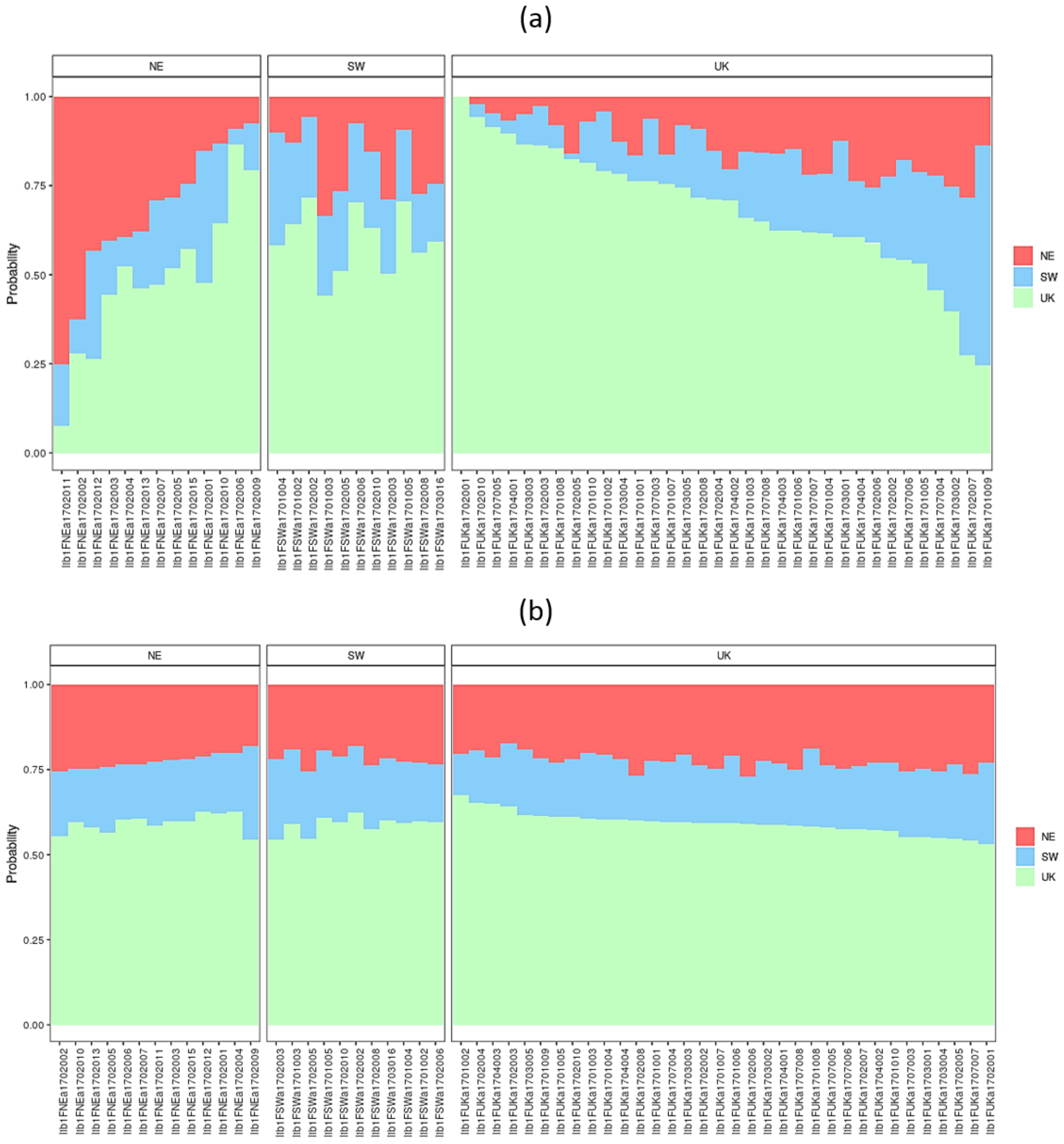

Figure. 3.6. Reallocation probabilities of individuals in the three subunits using (a) only genetic and (b) genetic, otolith shape and microchemistry datasets. Panels correspond to the subunits where individuals were sampled. Probabilities were estimated using the K-fold cross-validation method (K = 3) and all loci (2902 SNPs).

3.6. Discussion

Understanding the structure of marine populations and their functioning remains a challenge and merits to be thoroughly investigated to facilitate the sustainable exploitation of marine resources and the management of marine ecosystems. In this study, an individual-based approach combining genetic markers, otolith shape and microchemical composition was developed to complement the lack of knowledge on the stock structure of common sole living in the EEC. Compiling congruent information from tracers with different spatiotemporal resolutions is currently considered the best practice to provide robust information on population structure and allow relevant decision in fisheries and conservation (Reiss *et al.*, 2009; Cadrin *et al.*, 2014; Kerr *et al.*, 2017). Individual tracers were first analyzed separately and then combined to assess the benefits of an integrated approach.

3.6.1. Partial synergy between tracers revealed a signal of stock structure

Based on genetic and otolith shape analyses of common sole, spatial contrasts were found between three regions of the EEC. The genetic approach pointed to a low but significant spatial structure, which should be interpreted at an evolutionary time scale. Otolith shape-based approaches provided clues of spatial structure at the scale of a single generation, despite considerable genetic and physiological influences (Sturrock *et al.*, 2014; Izzo *et al.*, 2018).

For a long time, genetic approaches have been considered as the most relevant method to detect population structure and assess connectivity between populations (Ward, 2000; Hauser and Carvalho, 2008). However, failure to detect marine population structure with genetic tools is not a proof of the lack of population structure (Selkoe *et al.*, 2008; Hawkins *et al.*, 2016). Demographic tracers focusing on ecological time scales can reveal population structure where genetic approaches fail to do so. In the case of sole, results suggest that population structure is either relatively limited and/or recent or did not disperse over an evolutionary time scale (Selkoe *et al.*, 2008). Genetic structure of common sole at the scale of the North-East Atlantic Ocean based on microsatellites and mtDNA markers split in four groups, among which the North Sea and the EEC was distinct from the Bay of Biscay and to a lesser extent the Irish/Celtic Seas (Cuveliers *et al.*, 2012). Through the use of the state-of-the-art Single Nucleotide Polymorphisms, Diopere *et al.* (2018) highlighted a separation of sole populations between the North Sea/English Channel and the Bay of Biscay/Atlantic Iberian coast. Whereas SNPs allow spatial analysis of population structure on a fine scale, the internal structure of the EEC stock of common sole was not explored in great detail previously. In this study, genetic analysis based on the largest number of SNPs ever revealed spatial differentiation between subunits of the EEC. Even if F_{ST} values were low, suggesting low genetic differentiation, pairwise F_{ST} values were different between subunits, whatever the hypothesis of spatial structure. Since a low level of gene flow may homogenize the genetic structure and maintain genetic homogeneity (Hawkins *et al.*, 2016), the present genetic analysis suggested potential partial isolation of the three subunits from each other over an evolutionary time scale.

The otolith shape analysis resulted in consistent findings. The otoliths are incrementally built, from birth to death, by accretion of elements from the surrounding water and their growth is closely related to the somatic fish growth. Otolith shape is known to be influenced by numerous confounding factors such as ontogeny (i.e. the developmental stage), genetic and environmental factors (e.g. water salinity and temperature, depth, substrate and diet composition) (Cardinale *et al.*, 2004; Vignon, 2015). Otolith shape integrates the whole life history of the fish and could be considered as an individual tracer at the lifespan scale. In this study, a large data set of otoliths (> 700) of adult soles was available and allowed to detect differences in otolith shape between the three subunits of the EEC. Analysis of variance and post-hoc investigation of pairwise differences revealed significant variation of the outline of otoliths, which is a clue of spatial stock structure. However, elliptical Fourier descriptors failed to detect spatial variations (Appendix B) whereas the simpler shape indices highlighted significant spatial differences. Fourier descriptors are considered highly sensitive to fine variations in the otolith shape. Shape indices are usually considered as less powerful proxies of Fourier's harmonics (Russ, 1990; Tuset *et al.*, 2003; Delerue-Ricard *et al.*, 2018) and are used as supplement (e.g. Tuset *et al.*, 2003; Mérigot *et al.*, 2007). It was thus counter-intuitive to find spatial differences based on the shape indices and no spatial effect on the elliptical Fourier descriptors. Fourier series provide a large number of parameters (392 in this study, before reduction of the dimensions) that are used as discriminant variables. If the number of samples is insufficient for each combination of factors tested on these variables, potential effects of individual factors could not be detected. The number of samples required to highlight differences increases as a function of the number of variables (Saila and Martin, 1987). Herein, 28 harmonics were kept and summarized in 7 principal components. One may wonder if the number of variables compared to the number of samples in each combination of factors was sufficient to detect fine spatial variation in shape using Fourier series. This might explain why spatial variation was found in four shape indices but not in the elliptical Fourier descriptors.

In addition to the analyses of otolith shape indices and elliptical Fourier descriptors, directional bilateral asymmetry (DA) in shape between the left and right otolith (i.e. percentage of non-overlapping shape between the left and right otolith) was used and highlighted spatial differences (**Appendix B**). The degree of DA between otoliths has recently been proposed as a new tool to investigate population discrimination but has so far remained underused (Mahé *et al., in press*). In flatfish species, otolith DA occurs after metamorphosis of pelagic larvae and settlement in the benthic realm (Graf and Baker, 1983; Toole, 1993). During metamorphosis, common soles experience strong morphological and physiological modifications and adopt a dextral flat form (i.e. left blind side) with a 90° rotation of the stato-acoustic system (Graf and Baker, 1983). This lateralization process implies differences in biomineralization (i.e. carbonate accretion rates) of the right and left otoliths with the blind side otolith having higher growth rates (Sogard, 1991; Fischer and Thompson, 2004; Mille *et al.*, 2015). DA has been described for numerous flatfish species such as *Solea solea* (Mérigot *et al.*, 2007; Mille *et al.*, 2015; Delerue-Ricard *et al.*, 2018), *Pleuronectes platessa*, *Limanda limanda* and *Lepidorhombus whiffiagonis* (Mille *et al.*, 2015). DA in skeletal or calcified structures could be a genetic and/or phenotypic tracer regarding population discrimination studies (Mahé *et al., in press*). In this study, the degree of DA of the common sole was contrasted among subunits and reinforced the results of the analysis based on otolith shape indices.

To reassign adults to a source habitat (nursery ground) in order to identify potential sub-populations, a chemical baseline covering the source locations is required (Thorrold *et al.*, 2001; Gillanders, 2002a; Randon *et al.*, 2017; Reis-Santos *et al.*, 2018). By comparing the otolith composition of an individual to the baseline, assignment probabilities were estimated. Three main assumptions are assumed to characterize a baseline: (1) all potential sources have been defined and considered, (2) fingerprints are stable over time, and (3) significant differences between sources allow to clearly reallocate unknown individuals (Campana, 1999; Campana *et al.*, 2000; Gillanders, 2002b). In this paper, the first assumption was fulfilled since all potential nurseries had been identified previously (Riou *et al.*, 2001; Rochette *et al.*, 2010). Regarding the second assumption, only elements presenting stable concentrations over time (i.e. between year classes) were kept in the analysis, which reduced the number of tracers. Investigating the temporal stability of otolith signatures was essential since temperature, salinity and freshwater inputs in nurseries may vary between years and thus affect the chemical composition of water masses (Gillanders and Kingsford, 2000). Finally, the third assumption was not met. Although significant overall differences in otolith composition were found between nurseries, otolith signatures of juveniles were largely overlapping in Elemental Fingerprint Index. This resulted in a low reclassification success of the juveniles in their nurseries estimated by the Bayesian reallocation model. Hence, interpretation of adult reallocations was limited by the low reclassification success of the juveniles that composed the baseline. This suggests that the set of elements (Sr, Ba, Li and Mn) was not sufficiently discriminant between nurseries to allow precise reallocations. The uptake of an element from the water into the otolith depends on its bioavailability in the environment (Campana, 1999), physiological and genetic influences (Sturrock *et al.*, 2014; Izzo *et al.*, 2018) and environmental conditions such as temperature and salinity (Bath *et al.*, 2000; Martin *et al.*, 2004; de Vries *et al.*, 2005; Webb *et al.*, 2012; Izzo *et al.*, 2015). Consequently, the otolith elemental composition is supposed to reflect the chemical properties of the water masses. Along the French coast of the EEC, large volumes of freshwater impact the chemical composition of waters in the nursery grounds. In addition, salinity contrasts between the English coast, where salinity is high, and the French coast experiencing a relatively low salinity (Napoléon *et al.*, 2012). The different salinities should influence the water composition and provide contrasted signatures between the nurseries of the baseline. Otolith composition of juveniles were indeed significantly different between locations, however, not enough to discriminate nurseries.

To conclude, spatial structuring within the population of common sole of the EEC was supported by genetic and otolith shape analyses. Both tracers suggested three subpopulations as highlighted by former population-based approaches (Du Pontavice *et al.*, 2018; Randon *et al.*, 2018).

3.6.2. Interest of a quantitative (combined) vs qualitative holistic approach?

A current trend in the field of marine connectivity and population structure is the use of multiple methods (e.g. Cadrin *et al.*, 2014; Tanner *et al.*, 2016; Marengo *et al.*, 2017; Reis-Santos *et al.*, 2018). The philosophy behind this trend is that subtle differences in patterns might have a higher chance to be revealed through the combination of various analytical approaches. A holistic approach consists of reviewing all the stock identification information from different methods to infer stock structure; investigating the stock structure in a single study using two or more methods in combination on a range of samples; or preferably performing a wide range of methods on the same samples (Begg and Waldman, 1999). A holistic approach presents two main interests: (1) tracers provide a complementary view of the stock identity through the combination of spatial and temporal scales and (2) some tracers may fail in detecting spatial structure where others can detect signals. In this study, genetic markers provided a view of the stock structure over an evolutionary time scale whereas otolith shape indices and microchemistry rather focused on scales across a lifespan.

Combining tracer analyses on the same specimen is in theory the most attractive and relevant way to interpret the results and compare the performance of approaches having different ecological or evolutionary processes (Hawkins *et al.*, 2016; Tanner *et al.*, 2016; Chin *et al.*, 2017). The combination of tracers is especially valuable when the genetic signal is relatively weak (Selkoe *et al.*, 2008) and does not allow for a conclusion. Few studies have combined tracers from the same set of samples into a single analysis to assess population structure. Smith and Campana (2010) combined, in a Bayesian model, genetic and otolith microchemistry information into a single likelihood. Even if Bayesian modelling offers appealing perspectives, limitations emerged from the modelling skills required to build such complex models. Besides, Smith and Campana (2010) concluded that assignment success was better when tracers were analyzed separately compared to the integrated analysis. Tanner *et al.* (2016) suggested that "user-friendly" implementations should be developed. To circumvent such limitations, the use of multivariate analyses has been proposed and allowed accounting for the issue of the population structure and of the relative importance of each tracer (e.g. Marengo *et al.*, 2017; Reis-Santos *et al.*, 2018).

Here, the "user-friendly" *assignPOP* framework (Chen *et al.*, 2018) was used on a subset of fish for which the genotype, otolith shape and microchemistry were analyzed. By coupling genetic and non-genetic tracers into the *assignPOP* framework, no signal of spatial structure was observed on the restricted sample, in spite of the spatial contrast evidenced previously from genetics and otoliths shape in separate approaches. An explanation could be that the decreasing number of samples (from 120, 759 and 120 for separate tracer datasets, to 58 in the integrated approach) degraded the signal. By coupling genetic and non-genetic tracers into the *assignPOP* framework on a dramatically reduced sample, the weak signal of structure observed previously was lost. This is evidenced by genetic tracers analyzed alone in *assignPOP*: a signal of population structure was observed using the 120 individuals and confirmed the genetic differentiation suggested by F_{ST} values and DAPC (**Fig. A.1** and **A.2**), but the decrease in the number of samples (from 120 to 58) degraded the signal of genetic structure.

Each approach used to analyze population structure has its own discrimination capacity and does not require the same number of samples. Indeed, while up to date genetic tools (i.e. SNPs) and microchemistry here involved restricted sample size (order of magnitude near one hundred), analysis of otolith shape require large samples to be discriminant (> 700 samples for Tuset *et al.*, (2003), as herein). The differences in sample sizes are mainly due to financial and time constraints, and bioinformatics and modelling skills required to conduct genetic or microchemistry analyses. Technical advances in image processing allow for an increase in the sample size of otolith shape by extracting shape descriptors automatically from an image database (**cf. 2.4**). Therefore, each natural tracer has its own range of sample size, adapted to its discriminatory abilities and costs. Hence, when combining tracers in an integrated approach (**cf. 3.4**), the number of samples dramatically decreases, which alters the statistical accuracy and the related interpretations.

To conclude, this study advocated a holistic approach (i.e. qualitative approach) to investigate population structure and connectivity by analyzing different parallel tracers differing in their spatiotemporal resolution. In contrast, an integrated analysis combining tracers in a single framework (i.e. quantitative approach) did not appear to be the best option. The combination of tracers with different discriminating powers and contrasting costs of data acquisition, leading to an unbalanced sample size, may lead to a restricted number of samples in the integrated analysis, hence preventing to improve the output compared to the single-method analyses.

3.7. Conclusion of chapter

This third chapter proposed an individual-based approach to assess the spatial structure and connectivity of the common sole of the EEC. Interesting results emerged from the individual-based approach associating genetic and non-genetic tracers.

Regarding the genetic analysis, **three distinct genetic pools of individuals emerged and suggested associated spatial population structure made of three subunits,** the SW, NE and UK (**Fig. 3.7**). This analysis provided information of the stock structure over an evolutionary time scale. The low but significant signal of genetic spatial structure suggested a stabilized process at the scale of the EEC.

Similar spatial structure in three subunits emerged from the otolith shape analysis (**Fig. 3.7**). Surprisingly, the up-to-date Fourier descriptors failed to detect a signal of spatial structure whereas shape indices showed significant spatial variation. This analysis informed the stock structure at the lifespan scale and was congruent with genetic findings.

Connectivity between nursery and spawning grounds was investigated through the otolith microchemistry. The juvenile baseline was poorly discriminant among nurseries which biased the reallocation of adults to their nursery of origin (**Fig. 3.7**). Hence, this **promising tracer of connectivity throughout the life cycle failed in assessing the exchanges between nurseries and spawning grounds** because of low contrasted otolith signatures.

Finally, the **combination of tracers into a machine learning framework failed to detect a clear signal of structure spatial**. Indeed, the combined analysis of of genetic and non-genetic tracers was performed on a small number of specimens which limited the performance of the approach. Then, a qualitative investigation of the congruence/divergence of tracers was preferred in this study.

Therefore, a congruent signal of spatial structure was found through the genetic and otolith shape analyses and suggested a metapopulation structure composed of the SW, NE and UK subunits. These findings were in line with the results of the population-based approach proposed in chapter 2 (**Fig. 3.7**). Regarding the importance of a holistic approach in the assessment of stock spatial structure (Begg and Waldman, 1999; Cadrin *et al.*, 2014; Kerr *et al.*, 2017), the next chapter proposes to combine individual and population-based approaches.

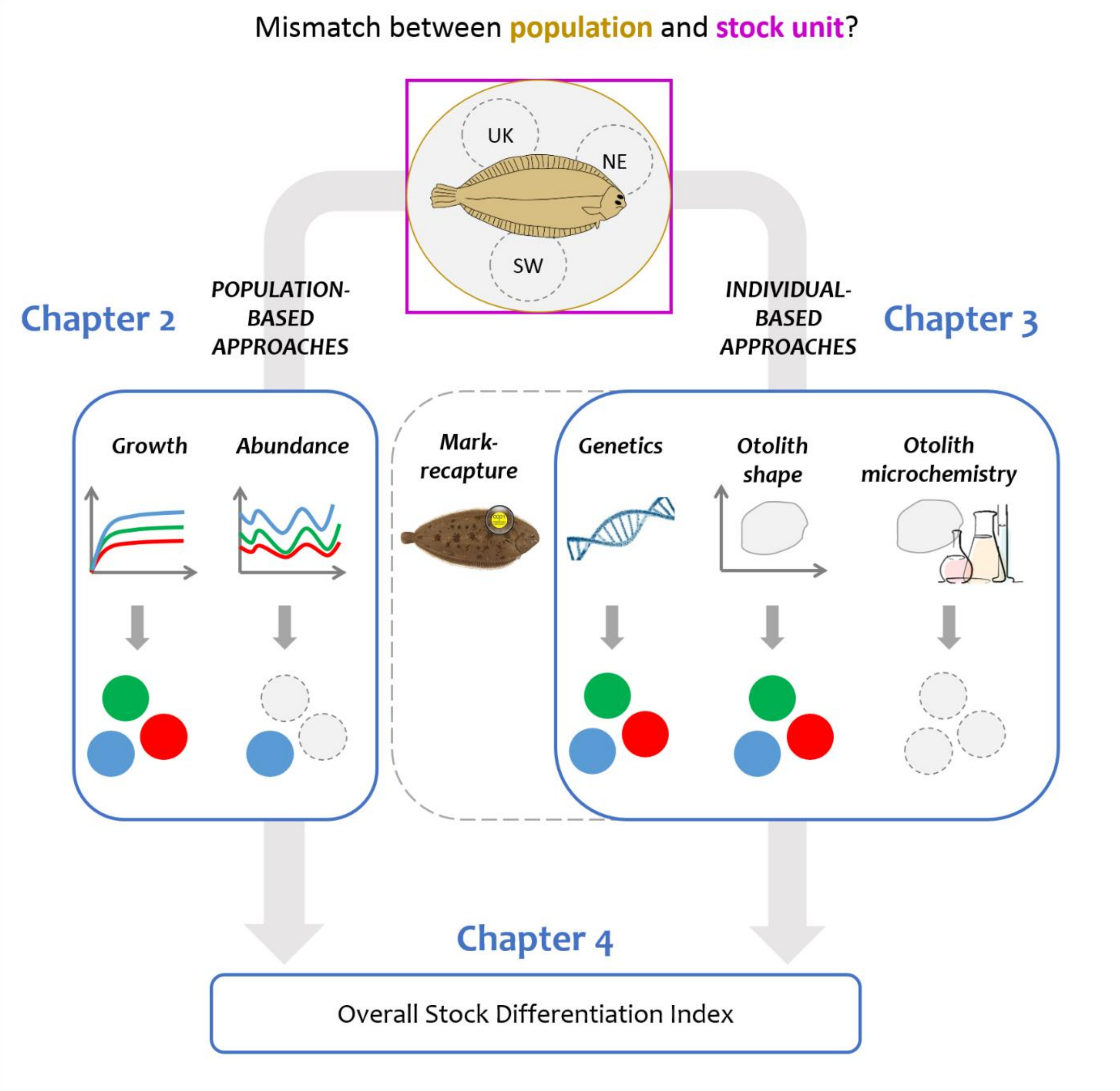

Figure. 3.7 Overview of the results of the common sole (*Solea solea*) population structure inside the Eastern English Channel stock (ICES division VIId) found in chapters 2 and 3 of the thesis. The dotted line indicates that the mark-recapture analysis was not performed in this thesis but that the related findings of a previous study (Lecomte *et al.*, 2019) were included in chapter 4.

Chapter 4

General discussion through a holistic approach

General discussion

4.1. Introduction of chapter

In chapter 2, population-based approaches evidenced a long-lasting signal of spatial structure of the common sole population of the EEC, and potential isolation of the SW subunit. In chapter 3, individual-based approaches highlighted a metapopulation structure made of three subunits, the SW, NE and UK. In this chapter the previous population and individual-based approaches are discussed and combined in a holistic framework.

A holistic approach is considered as the best practice to assess the spatial structure of fish stocks (Begg and Waldman, 1999; Cadrin *et al.*, 2014; Kerr *et al.*, 2017). It consists of reviewing all the available information of the stock structure and providing a synthetic view of the alignment between the stock and the underlying biological population.

The combination of tracers having various spatiotemporal resolutions is challenging (Tanner *et al.*, 2016). Consequently in this chapter, after reviewing available information of the stock structure, the synthetic semi-quantitative Stock Differentiation Index (SDI; Welch *et al.*, 2015; Izzo *et al.*, 2017) is calculated. For this purpose, population and individual-based approaches are considered in the holistic framework and a recent mark-recapture study conducted on the common sole of the EEC to assess movements of sub-adult and adult individuals is also accounted for.

This chapter corresponds to the general discussion of this thesis but is presented as an article that will be submitted in a scientific journal.

A holistic investigation of tracers at population and individual scales highlighted population structure for the common sole of the Eastern English Channel

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4.2. Abstract

The exploitation of marine resources is based on the understanding of population distribution, structure and functioning. The mismatch between stock units and populations can dramatically bias the assessment and lead to irrelevant measures in fisheries management. The common sole of the Eastern English Channel (ICES division VIId) is a flatfish species of high interest that has been overexploited over the last decades. Low connectivity induced by early life stages across the stock was previously informed, but gaps remained regarding the role of adults in population connectivity and spatial structure. In this study we filled these gaps by (1) reviewing all the information of stock identity through a large range of population and individual-based approaches, and (2) combining this information in a semi-quantitative framework, i.e. the Stock Differentiation Index (SDI). Regarding the population-based approaches, growth and abundance-at-age analyses highlighted a lasting signal of population structure, with three subunits. Regarding the individual-based approaches, genetic analysis of SNPs highlighted low but significant structure in three subunits over an evolutionary time scale. Moreover, a mark-recapture study demonstrated a low level of exchange between these subunits. Finally, an analysis of otolith shape pinpointed to spatial variation of shape between the three subunits. Overall, the SDI = 0.78 suggested evidence of spatial structure. We revisited the benefits of the holistic approach in the identification of a metapopulation structure of the common sole of the EEC. Misalignment of the stock with the underlying biological metapopulation calls for improvement of the assessment-management to ensure its sustainable exploitation.

Keywords

Stock – Metapopulation – Flatfish – Holistic approach – Assessment-management options

4.3. Introduction

Despite that the stock concept is central in fisheries science, several definitions could be adopted, depending on the scientific question and methods used (Begg *et al.*, 1999a; Abaunza *et al.*, 2008; Cadrin, 2020). From a fisheries management perspective, the stock is basically defined as a working unit for assessment models and management decisions (Kerr *et al.*, 2017; Cadrin, 2020). Ihssen *et al.* (1981) defined the stock as a monospecific group of individuals mating randomly to display spatiotemporal group integrity. In other words, the stock spatial unit is supposed to fit with the underlying biological population, stock assessment and management being provided at the population scale. Stock assessment models basically suppose that a population is closed and well mixed (Cadrin, 2020) and then, population vital rates and productivity are assumed to be homogeneous across the stock (Cadrin *et al.*, 2013; Bosley *et al.*, 2019). However, the stock delineation is frequently unclear (Cadrin *et al.*, 2010; Zemeckis *et al.*, 2014; Mahé *et al.*, 2016) inducing a misalignment between the stock and the biological population (Hawkins *et al.*, 2016; Kerr *et al.*, 2017). Such mismatch could lead to bias in stock assessment, thus to unsuitable exploitation of subunits having different productivity dynamics (Fu and Fanning, 2004; Cadrin and Secor, 2009; Goethel and Berger, 2017).

Delineating marine populations and understanding their functioning are difficult aims because biology is “messy” (Tawfik, 2010) and because aquatic species are rarely observed directly (“Counting fish is like counting trees except you can’t see them and they move”; John Sheperd). Marine connectivity is thus more complex to observe but is higher than in the terrestrial and freshwater realms. Through the exchanges of individuals that link populations (Secor, 2015), marine connectivity is involved in population structure (Parrish, 1989) at various spatial scales (Waples and Gaggiotti, 2006; Reiss *et al.*, 2009; Ciannelli *et al.*, 2013; Costello and Connor, 2019). Degrees of connectivity vary from panmixia to complete isolation of populations (e.g. Smedbol and Wroblewski, 2002; Abaunza *et al.*, 2008; Cadrin *et al.*, 2010), going through metapopulations that display a more or less consistent spatial structure (Waples and Gaggiotti, 2006). However, the increasing magnitude of habitat fragmentation and degradation worldwide and the environmental shifts induced by climate change are shaping new distribution areas and are challenging the definition of marine populations (Link *et al.*, 2010). Uncertainties regarding the spatial scope of stocks and populations thus arise from multiple factors that need to be considered in stock assessment and management.

Challenging such issue, Kerr *et al.* (2017) evidenced the need for adapting fisheries exploitation to the underlying population structure. In order to identify accurate management units, integrated approaches are recommended to capture the prevailing stock structure (Welch *et al.*, 2015). To do so, the first step consists of reviewing all available information of the stock identity into a holistic approach. The holistic understanding of population structure is currently considered as the best practice to draw robust conclusions regarding the stock structure (Begg and Waldman, 1999; Waldman, 1999; Abaunza *et al.*, 2008; Cadrin *et al.*, 2014). Indeed, population structure is induced by processes ranging from ecological to evolutionary time scales. Bringing together spatial variations in phenotypic and genetic characteristics can help to elucidate the stock identity (Cadrin and Secor, 2009; Cadrin *et al.*, 2014). Combining different methods allows increasing the likelihood of identifying the “true” population structure since one tracer can detect a signal where another fails to do so (Begg and Waldman, 1999; Abaunza *et al.*, 2008; Zemeckis *et al.*, 2014; Pita *et al.*, 2016). The identification of complex marine population structure and the associated uncertainty found through holistic approach (Kerr *et al.*, 2017) allows for further evaluation of the consequences of alternative assessment and management strategies regarding biological, economic and social purposes, through Management Strategy Evaluation (MSE). Using this state-of-the-art management decision-making method, the performances of each alternative option are assessed by providing observation, process and implementation errors (Sainsbury *et al.*, 2000; Bunnefeld *et al.*, 2011).

The common sole (*Solea solea*) of the Eastern English Channel (EEC; ICES division VIId; **Fig.4.1**), a substantively harvested flatfish species, has been overexploited over last decades (ICES, 2017b). It was found to be genetically distinct from the adjacent Western English Channel (ICES division VIIe) and the North Sea (ICES division IVc) stocks (**Fig.4.1**; Diopere *et al.*, 2018). However, misunderstanding regarding the internal stock structure remained (Rochette *et al.*, 2013; Archambault *et al.*, 2016; ICES, 2017a). Reproduction takes place in early spring on several distinct spawning grounds (**Fig.4.1**; Rochette *et al.*, 2012). After hatching, pelagic larvae drift passively towards shallow coastal and estuarine nursery grounds (**Fig.4.1**; Grioche, 1998; Rochette *et al.*, 2010; Savina *et al.*, 2010) where individuals metamorphose and settle as juveniles for about 2.5 years before mature soles join spawning areas in deeper waters (Riou *et al.*, 2001; Rochette *et al.*, 2010). Larval and juvenile-induced connectivity is low at the scale of the EEC stock. Indeed, biophysical modelling highlighted that larvae were mostly advected towards the nearest nursery grounds (Rochette *et al.*, 2012). Juveniles have been found to display very moderate movements away from their nursery habitats (< 10 km; Le Pape and Cognez, 2016) and high sedentarity in the local nursey grounds (Riou *et al.*, 2001). However, the sub-adult and adult-mediated connectivity were poorly documented despite its potentially high importance in population structuring (Mullon *et al.*, 2002; Frisk *et al.*, 2014). Subunits in the EEC stock of common sole have been hypothesized, based on several lines of evidence: the low connectivity induced by early life stages and the presence of natural barriers with unsuitable habitats for the common sole, benthic after metamorphosis, such as rocky reefs (**Fig.4.1**) and a deep central channel covered by gravels (Rochette et al., 2012; Archambault et al., 2016). However, this hypothesis remained poorly investigated (Randon et al., 2018). Ignoring such spatial structure could have led to a "myopic view" of productivity across the stock (Orensanz and Jamieson, 1998) and to biases in stock assessment (Archambault *et al.,* 2016).

Focusing on sub-adult and adult stages, recent studies have focused on the structure of the EEC stock of sole using a large range of approaches, from population-based (abundances and population growth; Du Pontavice *et al.*, 2018; Randon *et al.*, 2018) to individual-based approaches (mark recapture, genetics, otolith shape and otolith microchemistry; Lecomte *et al.*, 2019; Randon *et al., sub.*). The present study aims at (1) synthetizing all available information regarding the stock structure and (2) discussing alternative assessment and management strategies for the stock of sole of the EEC. To do so, a semi-quantitative approach, the Stock Differentiation Index (SDI; Welch *et al.*, 2015; Izzo *et al.*, 2017), was calculated to collate in a holistic approach (Kerr *et al.*, 2017) the various populations and individual-based analyses that previously provided information on the stock structure.

4.4. Material and methods

4.4.1. Current delineation of the Eastern English Channel stock of sole and alternative hypotheses

The EEC stock of sole is currently assessed and managed as one single stock (**Fig.4.1**), but the existence of three putative subunits has been hypothesized (Rochette *et al.*, 2013; Archambault *et al.*, 2016): the southwest subunit (SW; along the southwestern French coast of the EEC), the northeastern subunit (NE; along the northern French coast of the EEC) and the English subunit (UK; along the southern English coast of the EEC) (**Fig.4.1**).

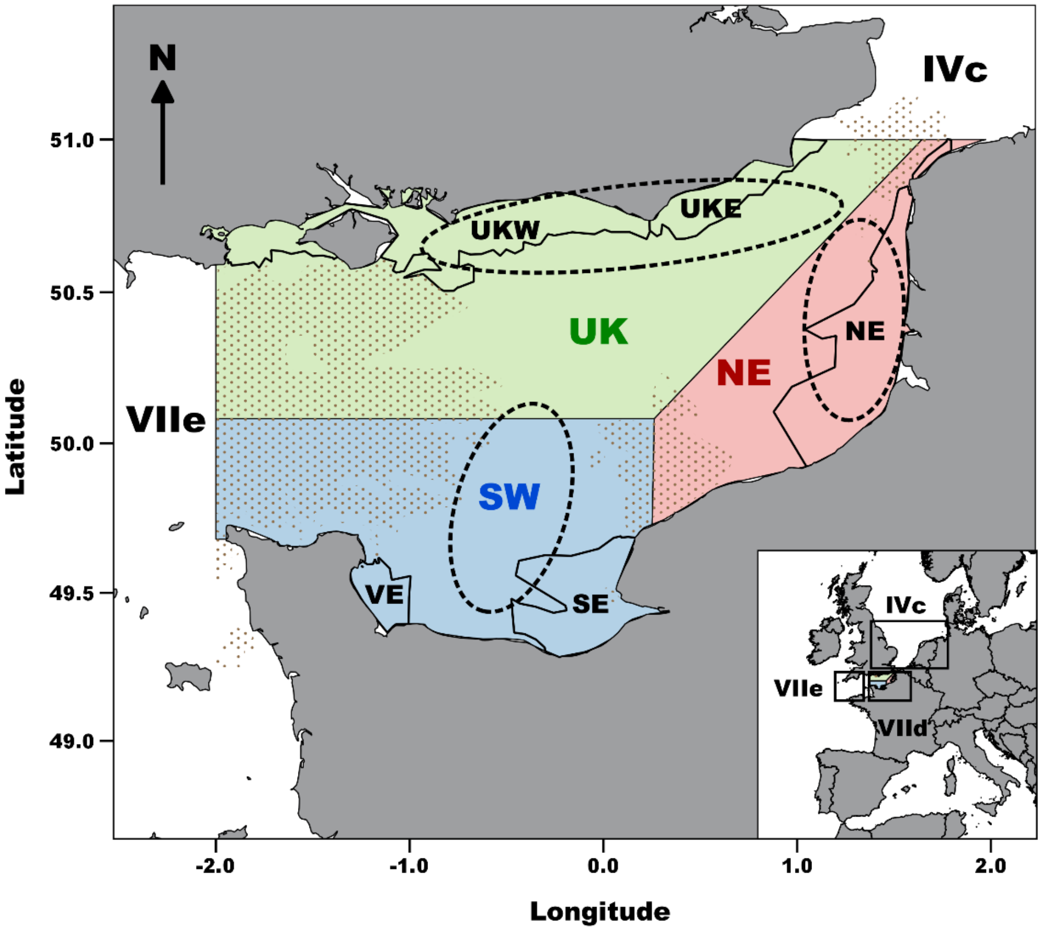


Figure. 4.1 Map of the EEC stock of common sole (ICES division VIId) and the three putative subunits (SW, NE and UK). Light grey dots represent rocky reefs. Coastal and estuarine nursery grounds (25 m isobath) and spawning areas (Rochette *et al.*, 2012) are delineated by solid and dotted lines, respectively. VE = Bay of Veys, SE = Bay of Seine, NE = Bay of Somme, UKE and UKW = the Eastern and Western parts of the UK coasts.

As suggested by Kerr *et al.* (2017), alternative hypotheses were proposed and tested in population and individual based approaches (Randon *et al.*, 2018). These alternative hypotheses consisted in pooling subunits : the null hypothesis (H0) considered the EEC as a single stock (i.e. the current assessment and management unit), while the remaining four alternative hypotheses assumed the stock to be partitioned into two or three subunits (**Table 4.1**).

Table 4.1. Hypotheses of spatial structure within the EEC stock of common sole (ICES division VIId). The three subunits (H1) considered are the UK (along the UK coast), NE (North-East French coasts) and SW (South-West French coasts) subunits. These primary subunits are pooled to define the null hypothesis (H0) and alternative hypotheses (H2, H3 and H4). "+" means that primary subunits are pooled.

Hypotheses	Subunits	Number of subunits
H0	UK + NE + SW	1 single stock
H1	UK, NE and SW	3
H2	(NE + SW) and UK	2
H3	(UK + SW) and NE	2
H4	(UK + NE) and SW	2

4.4.2. Review of information obtained through population and individual-based approaches

Six approaches were developed recently to analyze potential structure in the EEC stock of common sole (**Table 4.2**). Abundance-at-age throughout the cohorts and growth aimed at estimating differences at the population scale. Individual-based approaches focused on the estimate of individual movements or inter-individual differences in phenotypic and/or genotypic characteristics. Based on the assumption that a well-mixed stock should present homogeneous spatial patterns of demographic, genetic and phenotypic variables (Cadrin, 2020), population and individual-based approaches aimed at detecting spatial differences in these variables to highlight population structure. Each approach had its own spatiotemporal scale (**Table 4.2**) so that synthetizing results in a holistic approach allowed increasing the likelihood of detecting spatial structure and the reliability of the assessment (Waldman, 1999; Abaunza *et al.*, 2008; Cadrin *et al.*, 2014; Tanner *et al.*, 2016).

Table 4.2. Review of the reference studies dealing with the spatial stock structure of the common sole of the EEC (ICES division VIId). Spatial and temporal scales represent the resolution of tracers. Temporal scales in brackets represent the extent of the data used in each study.

Focus	Type of tracer	Spatial scale	Temporal scale	Reference
Population-based approach	Abundance-at-age	VIId	Generational (1990-2015)	Randon *et al.*, (2018)
	Growth			
Individual-based approach	Mark-recapture	VIId + VIIe +IVc	Individual lifespan (1970-2018)	Lecomte *et al.*, (2019)
	Genetics	VIId	Evolutionary (2017-2018)	Randon *et al.*, *sub.*
	Otolith shape	VIId	Individual lifespan (2016-2018)	
	Otolith microchemistry	VIId	Individual lifespan (2017-2018)	

4.4.2.1. Population-based approaches

Analyzing spatiotemporal patterns of growth and abundance-at-age, Randon *et al.* (2018) tested for a long-lasting signal (**Table 4.2**) of spatial structure inside the EEC stock of sole. They retrieved length-at-age data from the UK Beam Trawl Survey (UK-BTS) and estimated both von Bertalanffy growth parameters and synchrony (i.e. correlation) between trends in density-at-age throughout cohorts, to be used as structure markers (Begg *et al.*, 1999b; Cope and Punt, 2009; Erlandsson *et al.*, 2017; Walter *et al.*, 2017).

Growth

Heterogeneous growth parameters across the stock suggested population structure. The hypothesis of three subunits was supported (H1; **Table 4.1**). In spite of congruent decreasing trends over the time series in each subunit, higher asymptotic length was found in the SW compared to the UK and NE subunits.

Abundance-at-age

The asynchrony (i.e. different patterns) in density-at-age throughout the cohorts between subunits also evidenced spatial stock structure. High synchrony among time series was observed in the SW subunit, suggesting high spatiotemporal integrity inside this subunit and indicating its potential isolation from the rest of the EEC stock.

Combining analyses of growth and abundance-at-age over two decades, Randon *et al.* (2018) highlighted a long-lasting signal of stock structure with isolation of at least one subunit.

4.4.2.2. Individual-based approaches

- **Mark-recapture**

Investigation of post-larval dispersal was evaluated through a mark-recapture study covering the three putative subunits of the EEC stock and the adjacent North Sea (ICES division IVc) and Western English Channel (ICES division VIIe) stocks (Lecomte *et al.*, 2019). Adult connectivity was estimated using a state-space mark-recovery model that integrated mark-recapture (i.e. Peterson disks) data from multiple release experiments over three decades (**Table 4.2**).

Results suggested very low movements of adult soles between the three subunits of the EEC. Besides, exchanges with adjacent stocks were even lower. This study supported the hypothesis of segregated subunits (H1; **Table 4.1**) within the EEC.

- **Genetics**

Using up-to-date genetic markers (Single Nucleotide Polymorphism; SNPs), population structure was interrogated over an evolutionary time scale (Randon *et al., sub.*). Focusing on adult individuals on spawning grounds, this analysis aimed at understanding potential segregation across the stock. Spatial variations in genetic features were expected to reveal reproductive isolation.

Results indicated that genetic differentiation was low but significant between subunits, whatever the hypothesis of spatial structure. Genetic analysis highlighted isolated genetic pools within the EEC stock, reinforcing the hypothesis of stock structure of the common sole across the EEC.

- **Otolith shape**

A large data set of otolith shape descriptors was investigated in order to test for potential spatial variations (Randon *et al., sub.*). Since the shape of the otolith results from a combination of ontogenetic (i.e. development stage), environmental and genetic factors (Cardinale *et al.*, 2004; Vignon, 2015), spatial differences would indicate population segregation and thus stock structure from an assessment-management perspective.

Shape indices were significantly different between subunits, particularly regarding hypotheses H1, H3 and H4. Otolith shape analysis allowed detecting a signal of stock structure but did not allow for deciding about the best structure hypothesis.

- **Otolith microchemistry**

Dispersal between nursery and spawning grounds was investigated over an ecological time scale using otolith microchemistry (Randon *et al., sub.*). Otolith microchemistry is a key tool to assess connectivity between habitats, or populations (Campana *et al.*, 2000; Tanner *et al.*, 2016), since otoliths grow continuously without resorption and its chemical composition mainly reflects the surrounding water chemical properties (Campana, 1999).

However, using a Bayesian model, spawners failed to be confidently reallocated to their original nurseries. Despite significant spatial differences in elemental compositions, signatures of the juvenile baseline (i.e. atlas of otolith composition of juveniles sampled in all nurseries of the EEC; **Fig. 4.1**) were partially overlapping. As a result, reclassification success of juveniles was very low in nurseries of the NE and UK subunits. Then, poor discriminatory capacity of the juvenile baseline prevented the accurate reallocation of adult individuals using their juvenile signatures. Otolith microchemistry failed to assess connectivity between nursery and spawning grounds and prevented from drawing conclusions regarding the stock structure.

4.4.3. Stock Differentiation Index calculation

To integrate the previous findings on the EEC stock structure of common sole into a holistic approach (Kerr *et al.*, 2017), a Stock Differentiation Index (Welch *et al.*, 2015; Izzo *et al.*, 2017) was chosen to combine tracers in a synthetic manner regardless of their spatial and temporal resolutions (Begg and Waldman, 1999; Waldman, 1999).

The SDI is a semi-quantitative method that aims at underlying spatial structure inside a stock (Welch *et al.*, 2015). When a tracer revealed spatial differences between two subunits, a difference value $DV = 1$ is assigned to the pairwise comparison. However, when a tracer failed to detect spatial differences between two subunits, a difference value $DV = 0$ is assigned. Then, pairwise SDI is calculated as:

$$\boldsymbol{SDI = \sum DV / Count\ DV}$$

Where $\sum DV$ corresponds to the sum of DVs for one pair of subunits and $Count\ DV$ is the total number of tracers used. The overall SDI (i.e. across the stock) measures the relative differences among subunits. SDI ranges between 0 and 1, 0 indicating no spatial structure and 1 suggesting maximal spatial differences between subunits. As suggested by Welch *et al.* (2015), the null hypothesis (H0) has to be retained when the overall SDI = 0, even if there is no clear evidence of a single stock. Besides, thresholds have been defined by Izzo *et al.* (2017) to evaluate the strength of the spatial segregation. SDI < 0.33 may indicate a weak spatial structure, 0.33 ≤ SDI ≤ 0.66 provides moderate evidence of spatial structure and SDI > 0.66 would highlight strong evidence of stock spatial structure.

In order to facilitate the SDI calculation and because it was difficult to select a particular hypothesis of spatial structure among the different configurations tested, the three-subunit hypothesis H1 was retained. Then, three pairwise comparisons among subunits were examined through pairwise SDI calculations (i.e. SW vs NE, SW vs UK and NE vs UK). The six available tracers (**Table 4.2**) were weighted equally in the calculation of the SDI since the purpose of this approach was not to account for their relative importance in the discrimination among subunits. The otolith microchemistry was kept in the calculation of the SDI despite its inefficiency in discriminating nursery grounds.

4.5. Results

Pairwise SDI were calculated between each pair of subunits and provided strong evidence of spatial separation between subunits, particularly between the SW and the rest of the stock (**Table 4.3**). These highest values of SDI between the SW and the two other subunits were due to the differences found with the abundance-at-age analysis (cf. 4.2.2.1).

Table 4.3. Difference values (DV) between pairs of subunits of the EEC regarding the six available tracers applied to the common sole. Pairwise SDI values are indicated for each pair of subunits. SDI < 0.33 indicates weak spatial differences, 0.33 ≤ SDI ≤ 0.66 evidences moderate spatial differences and SDI > 0.66 highlights strong evidence of spatial differences.

Pairwise subunits	Tracers						Pairwise SDI values
	Abundance	Growth	Mark-recapture	Genetics	Otolith shape	Otolith microchemistry	
SW vs NE	1	1	1	1	1	0	0.83
SW vs UK	1	1	1	1	1	0	0.83
NE vs UK	0	1	1	1	1	0	0.67

Finally, the overall SDI = 0.78 across the EEC stock of common sole, indicating strong evidence of spatial structure (**Fig. 4.2**).

4.6. Discussion

Whereas the connectivity induced by early life stages has been informed previously (Riou *et al.*, 2001; Rochette *et al.*, 2012; Le Pape and Cognez, 2016), gaps remained regarding the adult-mediated connectivity (Mullon *et al.*, 2002; Frisk *et al.*, 2014). The holistic approach developed here contributed to fill the gap in the population structure of this flatfish stock by focusing on sub-adult and adult stages. The present review of available information on the EEC stock structure and the use of a semi-quantitative SDI provided evidence of spatial structure and isolation of subunits for the common sole of the EEC.

4.6.1. The need for a holistic approach to delineate structure in complex marine metapopulation

Over the last decades, there was a growing interest in both population and stock identification issues (Begg *et al.*, 1999a; Abaunza *et al.*, 2008; Caldrin, 2020) for marine fish. For instance, the International Council for the Exploitation of the Sea (ICES) reported that in early 2000s, around 50 out of about 150 stocks were misaligned with underlying populations (Stephenson, 2002). Originally, management units were convenient political boundaries rather than aligning with biological characteristics (Bosley *et al.*, 2019), although political delimitations are obviously not physical barriers for marine fish.

Defining accurate stock boundaries is a “fuzzy art” (Cadrin, 2020). Both the preliminary politically oriented delineation of stocks and the complex fish metapopulation processes result in inconsistent stock identity. This could dramatically bias stock assessment (Reiss *et al.*, 2009; Cadrin *et al.*, 2013; Kerr *et al.*, 2017). When population structure and functioning are unknown, the current practice is to consider homogeneous vital rates and productivity across the stocks (Cadrin *et al.*, 2013; Bosley *et al.*, 2019). As a result, subunits with lower productivity in stocks could be overharvested or even lead to collapse, as observed over the last decades (e.g. Wooster, 1992; Stephenson *et al.*, 1999; Hutchings, 2005). Such mismatches between populations and stocks increase the risk of unsustainable exploitation of marine resources (Fu and Fanning, 2004; Cadrin and Secor, 2009; Ying *et al.*, 2011; Goethel and Berger, 2017).

A large range of methods emerged for the purpose of highlighting and delineating population structure for marine fish (Cadrin *et al.*, 2013; Östman *et al.*, 2017). Overriding methods are the use of artificial tags (e.g. Rogers *et al.*, 2017; Le Bris *et al.*, 2018; Lecomte *et al.*, 2019) and natural tracers such as genetics (e.g. Cuveliers *et al.*, 2012; Jasonowicz *et al.*, 2016; Diopere *et al.*, 2018), otolith microchemistry (e.g. Tanner *et al.*, 2016; Moreira *et al.*, 2018) and shape (e.g. Hüssy *et al.*, 2016; Mahe *et al.*, 2016), morphometry and meristics (e.g. Allaya *et al.*, 2016; Sley *et al.*, 2016), fatty acid (e.g. Joensen and Grahl-Nielsen, 2004; Grahl-Nielsen, 2005), parasites (e.g. Catalano *et al.*, 2014; MacKenzie and Abaunza, 2014) and life history traits (e.g. Begg, 2005; Erlandsson *et al.*, 2017). Biological and ecological fish processes are complex, and each tracer has its own spatial and temporal resolution. For instance, genetic markers provide information of gene flows at the evolutionary time scale whereas biological markers integrate information at the individual lifespan scale (Randon *et al., sub*). Genetic has long been considered as the more relevant technique to discriminate populations (Ward, 2000). However, low levels of gene flow are known to homogenize genetic structure of marine populations (Exadactylos *et al.*, 2003; Hauser and Carvalho, 2008; Hawkins *et al.*, 2016). A lack of genetic differentiation can be reported despite other tracers suggest spatial population structure (Selkoe *et al.*, 2008; Cuellar-Pinzon et al., 2016; Hawkins *et al.*, 2016). Such a situation would indicate that genetic divergence is too recent at the evolutionary time scale to be detected by up-to-date genetic markers (Selkoe *et al.*, 2008). This evidences the need for combining genetic and non-genetic tracers for an accurate assessment, especially in a changing world.

In addition, the extent of sampling is often constrained by financial and technical limitations (e.g. genetics and otolith microchemistry are expensive techniques compared to morphometric of meristics for instance, Randon *et al., sub*.), resulting in various sizes of data sets and resolution power (Randon *et al., sub*.). Consequently, the recommended combination of different methods, with various spatiotemporal scales, into a holistic approach is challenging (Begg and Waldman, 1999; Tanner *et al.*, 2016). The spatiotemporal overlap between tracers might be viewed as redundant information. However, it is not a waste of time and money to perform overlapping analyses since it allows to assess the congruence of the methods and to conclude on the relevance of the tracers used to draw solid conclusions on the degree of spatial structure. Then, combining redundant and complementary information from multiple tracers is undoubtedly the winning method to elucidate stock identity.

Combining in a single framework the results of population-based and individual-based approaches is challenging since resolutions vary from evolutionary to lifespan scales. Yet, no universal quantitative method exists to combine such kind of information into a single framework. Focusing on individual tracers, few studies have combined complementary information into multivariate analyses, Bayesian models or supervised machine learning framework (e.g. Smith and Campana, 2010; Marengo *et al.*, 2017; Reis-Santos *et al.*, 2018; Randon *et al., sub.*). However, pooling in a single analysis various individual tracers belonging to the same specimens with tracers at population scale is not feasible in many cases. Sometimes it is preferable to perform independent analyses and discuss the combination qualitatively instead of combining tracers in a quantitative way (e.g. Smith and Campana, 2010; Randon *et al., sub.*). Such semi-quantitative holistic approach was proposed through the SDI calculation (Welch *et al.*, 2015; Izzo *et al.*, 2017). Despite the SDI could be viewed as an inflexible method, it presents the main advantages to be easy to understand, to compute and to integrate population and individual-based approaches into a single framework. SDI is a simplification of the status of the stock structure because it does not take into account complex biological and ecological processes. However, the SDI is relevant to feed the discussion between scientists and stakeholders.

4.6.2. Evidence of long-lasting signal of spatial structure across the EEC stock of sole

Marine population structure occurs at a much smaller scale than expected by the traditional paradigm (Hauser and Carvalho, 2008). For a long time, larval dispersal has been considered the main driver shaping the connectivity in marine species (Cowen and Sponaugle, 2009). However, recently this paradigm was reevaluated and the importance of adult-mediated connectivity was highlighted (Mullon *et al.*, 2002; Frisk *et al.*, 2014). The new paradigm rather focuses on population connectivity (i.e. connectivity along the life cycle) than larval connectivity (Secor, 2015).

The stock of common sole of the EEC has been overexploited over the last decades (ICES, 2017b, 2018b). Combined with low recruitment levels experienced in 2012 and 2013, the drop-in spawning biomass has led to a critical exploitation status over the last years. At the same time, internal structure in the EEC stock was hypothesized (Rochette *et al.*, 2012), with potential consequences for stock dynamics and sustainable levels of exploitation (Archambault *et al.*, 2016, 2018). Accordingly, in 2017, a benchmark pinpointed the potential misalignment of the EEC stock with the underlying population of common sole (ICES, 2017a).

Population structure of common sole inside the EEC was partially informed for young stages. Regarding the early life stages (i.e. eggs and larvae), connectivity was low between subunits of the EEC, with larval drift and settlement occurring at small spatial scale (Rochette *et al.*, 2012). Therefore, moderate larval dispersal could have contributed to shaping a metapopulation (i.e. a set of subpopulations linking by dispersal; Kritzer and Sale, 2010) in the EEC. Also, juveniles displayed high fidelity to the nursery grounds where they settled after metamorphosis (Coggan and Dando, 1988; Riou *et al.,* 2001; Le Pape and Cognez, 2016). These findings were confirmed by a mark-recapture experiment that highlighted that 95% of the juveniles released in the UK nurseries were recaptured within 50 km (Burt and Millner, 2008). Therefore, nursery fidelity involved relatively small distances which contributed to maintaining a metapopulation structure in the EEC.

However, potential subadult and adult movements of soles, through individual dispersal after sexual maturity (Secor, 2015) were poorly documented. Migration distances were shown to be greater for adult than juvenile individuals in the EEC (Burt and Millner, 2008) but dispersal between subunits remained unknown. Herein, the use of a semi-quantitative SDI synthetized all the available information regarding the EEC stock structure by focusing on sub-adult and adult stages. The synergy of information from various tracers and the strength of the overall SDI (i.e. largely above the threshold of 0.66 fixed by Izzo *et al.*, 2017) highlighted a strong and long-lasting signal of spatial structure inside the stock **(Fig. 4.2)**. Spatial structure was found at the evolutionary (i.e. genetics), generational (i.e. population growth and abundances) and individual lifespan scales (i.e. otolith shape and mark-recapture). Low but significantly different genetic pools of individuals between the three subunits indicated a likely moderate and/or recent divergence.

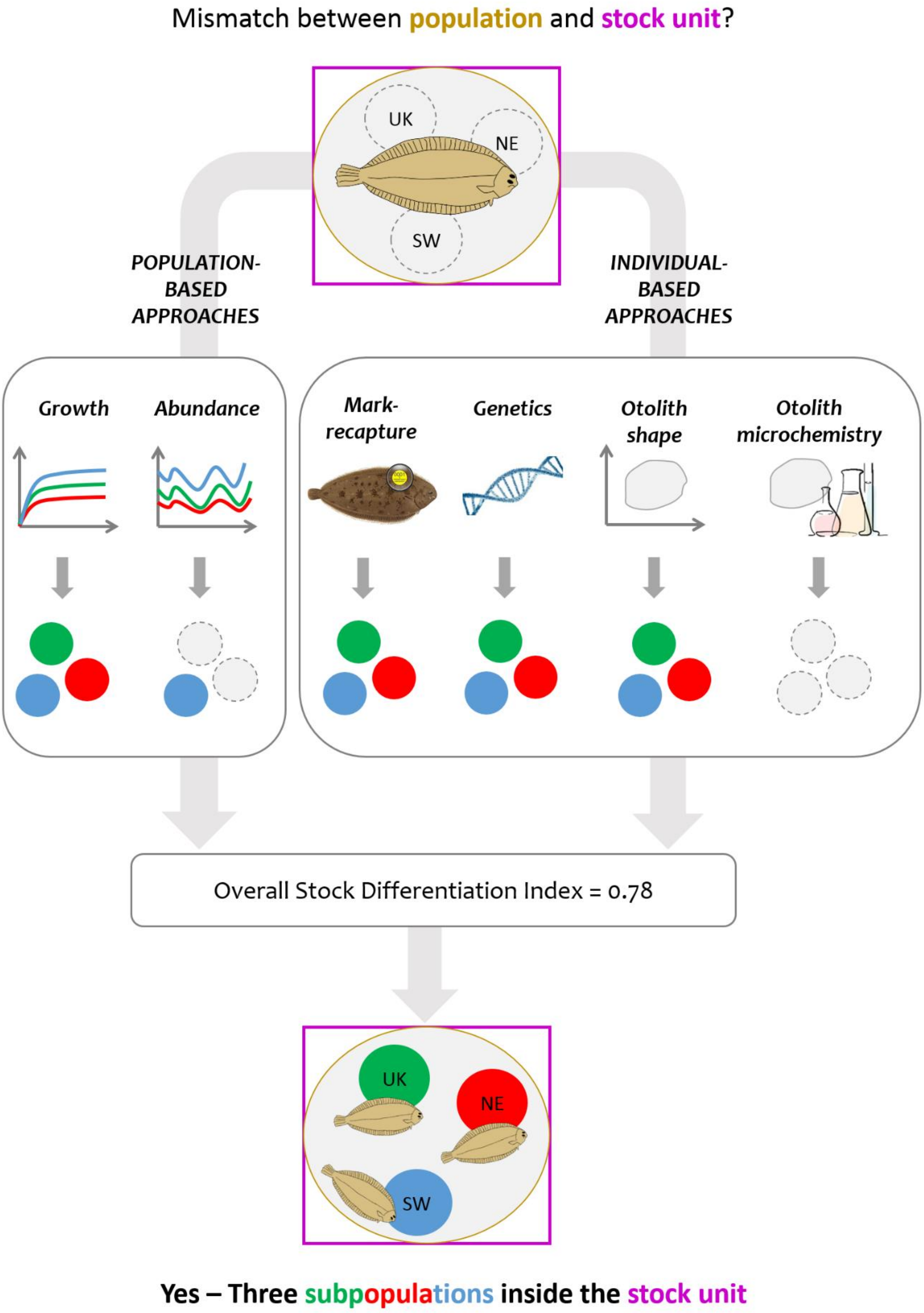


Figure. 4.2. Overview of the holistic investigation of population structure of common sole inside the Eastern English Channel stock.

Therefore, although fine scale ecological processes were not resolved (i.e. exchanges between spawning and nursery grounds; Randon *et al., sub*.), this holistic approach supported a metapopulation structure made of three subunits connected by low dispersal along the life cycle.

4.6.3. Practical considerations regarding alternative assessment and management strategies

As suggested by Kerr *et al.* (2017), after providing a review of information of the stock structure and contributing to a synthetic view of the stock (through the SDI), following steps consist of proposing alternative assessment and management options and evaluating their limitations.

Here, alternative spatial stock structures were tested by pooling subunits in different spatial structure hypotheses (**Table 4.1**). Although tracers did not converge towards a single spatial structure, the three-subunit hypothesis was retained with good confidence level regarding the pool of information and the SDI score. Considering three relatively independent subunits, different options could be proposed to improve the assessment and management of the stock.

The first option could be a change in stock unit boundaries by considering the three subunits as independent and assessing them separately. This option is certainly not the simplest alternative to answer the issues of the misalignment (Kerr *et al.*, 2017; Cadrin, 2020) since it would imply both higher amount of work for stock assessment and deep changes in the assessment and management process of the common sole and other species (e.g. trawl mixed fisheries).

Another option could be the implementation of spatially structured stock assessment (Cadrin and Secor, 2009; Berger *et al.*, 2017; Punt, 2019; Cadrin, 2020). Spatially explicit models allow incorporating population structure and connectivity to provide an estimate of the outcomes of ignoring spatial structure (Kerr and Goethel, 2014; Goethel *et al.*, 2016). By incorporating different data to inform the spatial structure (e.g. tagging data), spatial assessment improve the estimates of biological reference points (Goethel and Berger, 2017), particularly when growth varies spatially (Punt, 2019). Despite unanimous appreciation of these models to estimate the bias in assessment when spatial structure is ignored, they are still not broadly adopted because they require large data sets (increased cost of monitoring) and extensive knowledge of population structure to provide unbiased estimates (Goethel *et al.*, 2015). Furthermore, institutional inertia is probably the highest limiting point (Punt, 2019) and might explain that no spatially explicit models have been used to manage stocks in North Atlantic fisheries, to date (Kerr *et al.*, 2017). A Bayesian spatial integrated life cycle model explored the exploitation of the common sole of the EEC under a three subunits hypothesis to assess the impact of stock structure on the estimates of reference points and productivity (Archambault *et al.*, 2016). Exploitation was far above MSY (Maximum Sustainable Yield; $F/F_{MSY} = 1.8$) considering a single well-mixed stock (H0), but with contrasted patterns when considering three subunits; the NE and UK subunits being exploited above MSY (i.e. $F/F_{MSY} = 2$ and 1.9, respectively) and the SW subunit approaching full exploitation ($F/F_{MSY} = 1.05$). Thus, considering a metapopulation structure would undoubtedly help in providing unbiased estimates of reference points for the stock of sole of the EEC. However, it would be challenging to modify the current assessment models and then, other alternative strategies need to be considered.

A third option would consist of a different scenario of a trade-off between theory and management requirements. One can imagine adjusting the exploitation to the lowest productivity among the three subunits. It would prevent the stock from being overharvested. Another strategy could be a spatial management of fishing effort to adapt the exploitation level to local productivity. Finally, an alternative option could be to allocate the quota relatively to local contrasted productivities in the EEC stock of common sole (Archambault *et al.*, 2016). Therefore, local management strategies could be interesting and feasible options (Cadrin *et al.*, 2010; Wright *et al.*, 2019) and have to be evaluated relatively to data/methods, but also social, economic and institutional limitations (Punt, 2019).

Quantitative evaluation of the outcomes of alternative management options could be performed through MSE (Management Strategy Evaluation). MSE are currently viewed as the state-of-the-art for management decision-making since it evaluates biological and economic consequences of a range of management strategies (Sainsbury *et al.*, 2000; Bunnefeld *et al.*, 2011; Kerr *et al.*, 2017). Evaluation of management options is an interface between biology/ecology, fishery and management and should improve the assessment-management of the common sole of the EEC.

Conclusion Générale

Conclusion

Au cours de cette thèse, la structure du stock de sole (*Solea solea*) de Manche Est (division VIId, ICES) a été étudiée au moyen de traceurs aux résolutions spatiales et temporelles variées, en se focalisant sur les stades de vie préadultes et adultes (**chapitre 2 et 3**). Cette approche holistique a permis de mettre en évidence un consensus entre les résultats issus des différents traceurs (**chapitre 4**). À la lumière de ces résultats, des stratégies d'évaluation et de gestion du stock ont pu être discutées (**chapitre 4**).

5.1. Le contexte de l'étude

La sole commune est une espèce de poisson plat fortement exploitée en Manche Est par les pays limitrophes que sont la France et l'Angleterre, mais aussi par la Belgique (ICES, 2017b). En France, de nombreuses pêcheries dépendent de cette ressource halieutique. C'est pourquoi la gestion durable du stock de sole en Manche Est apparait comme une priorité. Depuis plusieurs décennies, ce stock a subi un déclin des abondances de poissons reproducteurs (i.e. biomasse féconde) ainsi que de très faibles recrutements entre 2011 et 2012 (ICES, 2018a). La combinaison de ces deux facteurs a mené le CIEM à considérer ce stock comme surexploité, mettant à mal sa gestion durable (ICES, 2018a).

La méconnaissance du fonctionnement des populations et de leur structure spatiale (et/ou temporelle) représente l'une des causes potentielles pouvant mener à une gestion non durable des ressources marines. En effet, lorsque le modèle d'évaluation du stock et les mesures de gestion ne sont pas adaptés à la réalité biologique, le risque de surexploitation du stock s'accroît (Kutkuhn, 1981; Smith *et al.*, 1990; Begg *et al.*, 1999a). Les limites d'unités de stocks ont été mises en place au XXe siècle et les quotas de pêche ont été calculés depuis 1983 sur l'hypothèse que les unités de gestions correspondent aux unités fonctionnelles (Association Française d'Halieutique, 2016). En général, lorsque la structure spatiale d'un stock est méconnue, la règle est de considérer que le stock correspond à une population unique et bien mélangée, et a une dynamique homogène (Cadrin *et al.*, 2013; Kerr *et al.*, 2017). Lorsque cette hypothèse n'est pas vérifiée, les modèles d'évaluation qui déterminent les mesures de gestion sont biaisés, ce qui peut conduire au déclin des stocks (Tuck and Possingham, 1993; Cadrin and Secor, 2009; Neat *et al.*, 2014; Goethel and Berger, 2017). Dans de nombreux cas, les populations de poissons se sont avérées structurées spatialement et temporellement (Waples and Gaggiotti, 2006; Reiss *et al.*, 2009; Ciannelli *et al.*, 2013). Pour un tiers des stocks de l'atlantique nord, la délimitation des stocks (unités de gestion) au début des années 2000s n'était pas alignée sur celle des populations biologiques (unités fonctionnelles) (Stephenson, 2002).

En Manche Est, la connaissance préalable à ce projet sur la dynamique spatiale de la population de sole commune était limitée. En particulier, la connectivité au sein du stock était partiellement renseignée. La sole commune est une espèce nourricerie-dépendante se reproduisant au printemps sur trois frayères hauturières en Manche Est (Rochette *et al.*, 2012). Suite à l'éclosion des œufs, les larves sont transportées vers des zones de nourriceries côtières ou estuariennes (Grioche, 1998; Savina *et al.*, 2010; Rochette *et al.*, 2012). Après la métamorphose, les juvéniles séjournent environ 2 ans et demi dans ces nourriceries avant de migrer vers des zones de nourrissage des adultes et les frayères, plus au large (Riou *et al.*, 2001; Rochette *et al.*, 2010). De précédentes études de modélisation biophysiques ont montré une faible connectivité induite par les larves à l'échelle du stock (Rochette *et al.*, 2012). Les larves sont advectées avec de très fortes probabilités vers les nourriceries les plus proches de leurs frayères d'origine, induisant peu de mélange au sein du stock. De plus, les mouvements des juvéniles apparaissent limités (Le Pape and Cognez, 2016), et ils restent sédentaires au sein de leur secteur côtier ou estuarien de nourricerie (Riou *et al.*, 2001; Durieux *et al.*, 2010). Ainsi, la connectivité induite par les jeunes stades de vie est limitée et une hypothèse de structure du stock en trois sous-unités a été émise précédemment (Rochette *et al.*, 2013; Archambault *et al.*, 2016). Ces trois sous-unités, les zones sud-ouest (SW), nord-est (NE) et anglaise (UK) (**Fig. 1.11**), ont été supposées sur la base de la faible connectivité induite par les larves et les juvéniles, ainsi que de la présence de plateaux rocheux et d'un chenal central profond et couvert de graviers constituant des barrières naturelles à la migration de la sole commune. En revanche, jusqu'ici, la connectivité induite par les stades préadultes (i.e. juvéniles quittant leur nourricerie avant maturité sexuelle) et adulte n'a pas été suffisamment renseignée (Burt and Millner, 2008; Archambault *et al.*, 2016) et l'état de structure du stock était largement méconnu. Le rôle des stades préadultes et adultes dans la connectivité des populations est pourtant potentiellement important (Mullon *et al.*, 2002; Frisk *et al.*, 2014) puisque les mouvements migratoires d'individus en vue de la reproduction sont susceptibles de participer au mélange du stock et à son homogénéité (Secor, 2015).

L'objectif de cette thèse était de tester l'existence d'une structure spatiale interne au sein du stock de sole commune de Manche Est en se focalisant sur la connectivité induite par les stades de vie préadulte et adulte. Pour ce faire, une approche holistique a été mise en œuvre en combinant des traceurs populationnels et individuels aux résolutions spatiales et temporelles variées. L'hypothèse de base d'une structure en trois sous-unités proposée par Rochette *et al.* (2013) et Archambault *et al.*

(2016) a été testée ainsi que d'autres configurations spatiales. Enfin, une méthode semi-quantitative a permis de combiner les deux types d'approches et de fournir une vision synthétique sur la structure spatiale au sein du stock.

5.2. Les méthodes et principaux résultats

L'approche holistique, sous la forme d'approche interdisciplinaire ou multitraceur, est recommandée dans l'étude de la structure spatiale et de la connectivité des populations de poissons. Cette approche consiste en premier lieu à réaliser plusieurs analyses en parallèle en utilisant des traceurs et/ou des marqueurs de résolutions spatiales et temporelles variables. Dans un second temps, les résultats de ces analyses sont combinés afin de déterminer s'il existe un consensus entre les hypothèses privilégiées par les différents traceurs et de parvenir à une synthèse sur la structure du stock. Dans la présente étude, deux grands types de méthodes, les approches populationnelles (**chapitre 2**) et individuelles (**chapitre 3**), ont d'abord été réalisées séparément avant d'être combinées et synthétisées (**chapitre 4**).

5.2.1. Les approches populationnelles

L'étude des traits d'histoires de vie et des attributs démographiques est recommandée avant d'approfondir l'analyse de la structure et de la connectivité des populations via d'autres traceurs. Ces approches populationnelles reposent sur des séries de campagnes scientifiques dont l'objectif est l'évaluation des stocks de poissons (Begg *et al.*, 1999b). Ce type d'approche est également profitable en premier lieu puisqu'elle peut permettre la mise en évidence de la structure du stock à une large échelle spatiale et temporelle. Ainsi, deux approches populationnelles ont été développées en premier lieu afin d'obtenir une vision sur le long terme de l'état de structure spatiale du stock de sole commune de Manche Est (**chapitre 2**). Pour cela, des séries temporelles de longueurs aux âges (i.e. 1989 – 2015) obtenues à partir d'une campagne scientifique (UK-BTS) ont permis d'étudier (1) les paramètres de croissance populationnelle et (2) la synchronie des abondances aux âges de poissons dans les sous-unités potentielles du stock. Différentes configurations de la structure spatiale ont été testées (**Table 2.1**).

- **La croissance populationnelle**

Le but de cette analyse était de détecter d’éventuelles différences spatiales de croissance entre les sous-unités, pouvant indiquer l’existence d’une structure spatiale pérenne du stock.

Un rétro calcul de la croissance des soles via un modèle de von Bertalanffy a permis l’estimation les paramètres de croissance par sexe, cohorte et sous-unité spatiale. En particulier, une sélection de modèles a été réalisée afin de tester la combinaison optimale de facteurs et de connaitre la structure spatiale la plus probable. Un proxy de la longueur asymptotique, le L_{10}, a été estimé à partir de ces courbes de croissance et son évolution temporelle a été étudiée pour chaque sexe et sous-unité.

Plusieurs résultats issus de cette analyse de la croissance populationnelle à long terme sont à retenir :

(1) La configuration la plus probable comprend trois sous-unités SW, NE et UK (hypothèse H1).
(2) Une diminution du L_{10} généralisée aux deux sexes et aux trois sous-unités suggère une évolution induite par la pêche (« Fishery-Induced Evolution »).
(3) Les différences de croissance entre les trois sous-unités sont pérennes sur la période couverte par la série temporelle.

Ainsi, l’étude de la croissance populationnelle a permis de détecter un signal stable de structure spatiale en trois sous-unités, sur le long terme.

- **La synchronie des abondances aux âges**

L’objectif de cette analyse était de rechercher le niveau de synchronie au sein des cohortes dans les séries d’abondances aux âges dans chaque sous-unité potentielle du stock. Une forte cohérence entre les années dans les séries d’abondances par cohorte au sein d’une sous-unité est le signe de son intégrité spatiale et de son isolement. A l’inverse, l’asynchronie dans les séries d’abondances aux âges au sein d’une sous-unité et/ou une meilleure cohérence au niveau du stock dans son ensemble suggère d’éventuels échanges avec d’autres sous-unités et donc une absence de structure spatiale.

Les séries d’abondances des âges 1 à 5 ont été transformées en anomalies de densités afin de rendre comparables les cohortes de fortes et de faibles abondances. Les corrélations intracohortes dans les séries temporelles d’anomalies de densités ont ensuite été estimées à l’échelle du stock entier (hypothèse H0) puis à l’échelle des différentes sous-unités selon les hypothèses alternatives de structure spatiale.

Deux principaux résultats sont à retenir de cette analyse :

(1) A l'échelle du stock entier (hypothèse H0), la synchronie par cohorte dans les séries temporelles de densités aux âges était forte pour les mâles et les femelles, indiquant une certaine intégrité du stock.
(2) Une structure spatiale avec isolement du SW a pu être mise en évidence puisque la synchronie au sein de cette unité surpassait celle des autres sous-unités, et celle du stock dans son ensemble.

Ainsi, l'étude des abondances aux âges a permis de détecter un signal de structure spatiale sur le long terme, avec un isolement de la zone SW.

Ce **chapitre 2** a donc permis de mettre en évidence un signal pérenne de structure spatiale du stock de sole commune en Manche Est. En revanche, à ce stade, aucune hypothèse prépondérante de structure spatiale n'était ressortie, même si l'isolement de la zone SW apparaissait probable. Ces résultats à l'échelle intergénérationnelle ont constitué une première ébauche de l'état de structure du stock. Des analyses supplémentaires dans le cadre d'approches individuelles ont ensuite été menées afin de confirmer ou d'infirmer les hypothèses de structure spatiale.

5.2.2. Les approches individuelles

Un grand nombre de traceurs individuels peuvent être considérés pour tester la structure spatiale et la connectivité des populations marines (Cadrin *et al.*, 2013; Östman *et al.*, 2017). Dans le **chapitre 3**, trois types de traceurs individuels ont été utilisés. D'abord, une analyse de la structure génétique du stock a été conduite à partir de marqueurs génétiques de type SNPs (Single Nucleotide Polymorphisms). Ensuite, une analyse de la morphologie des otolithes a été réalisée à partir d'indice de formes afin de tester un signal de structure spatiale du stock. Enfin, la connectivité entre les habitats de nourriceries et les frayères a été étudiée au moyen de la microchimie des otolithes adossée à un modèle Bayésien de réassignation. En synthèse, une analyse intégrée de ces trois traceurs sur les mêmes spécimens a été développée de manière à tester la congruence des traceurs et à synthétiser les acquis sur la structure spatiale du stock. Les mêmes hypothèses alternatives de structure spatiale du stock que celles utilisées au chapitre 2 ont été testées.

- **Analyses génétiques**

Le but de cette analyse était de détecter un éventuel signal de différentiation génétique entre les individus issus des différentes sous-unités spatiales testées. La mise en évidence de différentiations génétiques entre pools d'individus à fine échelle spatiale peut s'avérer laborieuse puisque de faibles flux de gènes peuvent suffire à homogénéiser les populations du point de vue génétique (Exadactylos *et al.*, 2003; Hauser and Carvalho, 2008; Hawkins *et al.*, 2016). En ce sens, le récent développement des SNPs a constitué une avancée considérable dans la détection des structures constituées à fine échelle spatiale et néanmoins stables à l'échelle évolutive.

Le séquençage d'individus adultes échantillonnés sur frayères pendant la période de reproduction (2017 et 2018) a été réalisé puis un jeu de SNPs par année d'échantillonnage a été déterminé. La différentiation génétique a été analysée au moyen des F_{ST} globaux et par paires (Weir and Cockerham, 1984) et d'une Analyse Discriminante des Composantes Principales (DAPC).

Les résultats à retenir concernant l'analyse génétique sont :

(1) Une différentiation génétique faible, mais significative pour les deux années analysées.

(2) Aucune hypothèse alternative de structure mise en exergue. Les différences étant significatives, quelle que soit l'hypothèse considérée, la ségrégation en 3 sous-unités paraissent réalistes.

Ainsi, cette étude génétique a mis en évidence l'existence d'une structure spatiale du stock à l'échelle évolutive avec un relatif isolement reproductif des trois sous-unités spatiales considérées.

- **Analyses de forme des otolithes**

L'objectif de cette analyse était de comparer la forme des otolithes issus des différentes sous-unités afin de mettre en évidence une éventuelle structure spatiale du stock. La forme des otolithes de poissons étant déterminée en partie par la génétique et l'environnement, sa variation spatiale constitue un signal de structure du stock, en intégrant l'histoire du poisson à l'échelle de son cycle de vie (Campana and Casselman, 1993; Cardinale *et al.*, 2004; Vignon, 2015).

A partir des images d'otolithes droits et gauches de soles adultes, les mesures de longueur, largeur, périmètre et aire ainsi que l'extraction des harmoniques de Fourier ont été réalisées de manière automatisée. Les indices d'ellipticité, de rondeur, de coefficient de forme, de circularité et de rectangularité ont été calculés. Une analyse de variance ainsi que des tests post-hoc ont été conduits sur chaque indice de forme afin de détecter un effet spatial dans la forme de l'otolithe. En parallèle, une analyse similaire à partir des descripteurs de Fourier a été réalisée (**Annexe B**). Enfin, la reconstruction de la forme des otolithes grâce aux descripteurs de Fourier a permis d'étudier l'asymétrie bilatérale des otolithes (i.e. pourcentage de non-recouvrement entre les otolithes droits et gauches) (**Annexe B**).

Les résultats issus de l’analyse de forme ont montré que :

(1) Les indices de formes (hormis la rectangularité) ainsi que l’asymétrie bilatérale variaient entre les trois sous-unités.
(2) Les descripteurs de Fourier n’ont pas permis de détecter un signal de structure spatiale (**Annexe B**).
(3) L’asymétrie bilatérale était variable entre les sous-unités, et spécifique en zone NE (cf. **Annexe B**).

Les différents indices n’ont donc pas abouti à un consensus entre les hypothèses alternatives de structure du stock, mais ont unanimement proposé l’existence d’une structure spatiale en trois sous-unités.

- **Analyses de microchimie des otolithes**

La microchimie des otolithes est un traceur particulièrement utilisé pour retracer la connectivité entre habitats ou entre populations (Campana *et al.*, 2000; Tanner *et al.*, 2016). L’otolithe se forme, sans résorption, de la naissance à la mort du poisson par accrétion de composés présents dans l’eau (Walther and Thorrold, 2006). La composition de l’otolithe permet de retrouver celle de l’eau au sein des habitats du poisson au cours de sa vie (à un coefficient ou une fonction de partitionnement près) (Bath *et al.*, 2000).

Dans le cas de la sole commune de Manche Est, l’objectif était d’utiliser la microchimie des otolithes pour identifier la nourricerie de provenance d’individus adultes pêchés sur frayères et ainsi pouvoir évaluer la connectivité entre les nourriceries côtières et les frayères identifiées dans les sous-unités spatiales de la Manche Est. Pour cela, la composition microchimique de chaque individu (juvénile ou adulte) a été caractérisée au niveau de la zone de l’otolithe correspondant à la phase juvénile en nourricerie (**Fig. C.1**). Les signatures des individus juvéniles ont ainsi permis de construire un atlas de référence de signatures de nourriceries. Par inférence Bayésienne, les individus adultes devaient être réassignés à leur nourricerie d’origine de manière probabiliste.

Cette analyse de microchimie des otolithes a mis en évidence que :

(1) Les signatures des juvéniles composants l’atlas de référence étaient significativement discriminantes.
(2) Leur succès de reclassification dans les nourriceries était néanmoins trop faible pour permettre de réassigner de manière robuste les individus adultes.

Ainsi, l'analyse de microchimie des otolithes n'a pas permis d'estimer la connectivité entre nourriceries et frayères à cause du faible pouvoir discriminant de l'atlas de référence.

- **Approche intégrée des traceurs individuels**

Parmi les approches holistiques possibles, la combinaison de traceurs issus des mêmes individus est particulièrement recommandée (Begg and Waldman, 1999). En effet, cela permet (1) de récupérer de l'information lorsque l'un des traceurs détecte une différence alors qu'un autre n'en détecte pas et (2) de cumuler les différences détectées par différents traceurs afin de synthétiser les signaux sur la structure spatiale.

Dans la présente étude, un cadre de *machine learning* a été utilisé afin de coupler les données génétiques (SNPs) aux données non génétiques (formes et microchimie des otolithes) et d'estimer de manière probabiliste l'origine des poissons (Chen *et al.*, 2018). L'objectif était d'analyser la force de signal de structure spatiale détectée par les trois traceurs combinés et d'estimer la proportion de soles classifiées dans leurs sous-unités d'échantillonnages.

Cette analyse intégrée des traceurs individuels a mis en évidence que :

(1) En réduisant le jeu de données génétiques aux seuls individus analysés pour les trois traceurs, le signal de structure spatiale préalablement mis en évidence est perdu, du fait du trop faible nombre d'individus.
(2) La combinaison des traceurs génétiques et non génétiques ne permet pas de détecter de signal de structure spatiale, à cause là aussi du faible nombre de spécimens analysés conjointement pour les trois traceurs.

Dans une telle situation, il est préférable de conduire une approche holistique par comparaison qualitative des résultats plutôt que de combiner les données, la réduction de l'échantillon aux individus analysés pour tous les traceurs conduisant à annihiler les signaux considérés individuellement.

Ce **chapitre 3** a mis en évidence l'existence d'une structure spatiale stable à l'échelle évolutive grâce aux analyses génétiques ainsi qu'à l'échelle du cycle de vie du poisson avec les analyses de formes des otolithes. En revanche, la connectivité entre nourriceries et frayères n'a pas été résolue par la microchimie des otolithes.

5.2.3. La combinaison des approches dans un cadre holistique multitraceur

Le **chapitre 4** de cette thèse était consacré à la combinaison des approches populationnelles (**chapitre 2**) et individuelles (**chapitre 3**) dans une approche holistique multitraceurs. La difficulté inhérente à la combinaison de ces deux types d'approches vient notamment de la différence des échelles d'estimation des paramètres (individuelle vs populationnelle). Pour pallier cette difficulté, un cadre de travail holistique semi-quantitatif a été sélectionné afin d'estimer la structure spatiale au sein du stock de sole de Manche Est.

Après avoir passé en revue les informations à disposition traitant de la structure du stock de sole de Manche Est, l'Index de Différentiation de Stock (SDI) a été calculé. Le SDI est une méthode semi-quantitative d'estimation de l'état de structure d'un stock. Cet indice varie entre 0 et 1, 0 indiquant une absence de structure spatiale (i.e. cas d'un stock homogène) et 1 indiquant un stock très fortement structuré. Les analyses utilisées pour calculer le SDI étaient :

- La croissance populationnelle (Chapitre 2; Randon *et al.*, 2018) ;
- Les abondances aux âges (Chapitre 2; Randon *et al.*, 2018) ;
- La capture-marquage-recapture (Lecomte *et al.*, 2019) ;
- La génétique (Chapitre 3 ; Randon *et al. soumis*) ;
- La morphométrie des otolithes (Chapitre 3 ; Randon *et al. soumis*) ;
- La microchimie des otolithes (Chapitre 3 ; Randon *et al. soumis*)

Les résultats d'une analyse de capture-marquage-recapture n'ont pas été développés dans le cadre de cette thèse (Lecomte *et al.*, 2019), mais ont été pris en compte dans le calcul du SDI. Cette étude avait permis de montrer de très faibles mouvements d'individus préadultes et adultes entre les trois sous-unités de SW, NE et UK et donc une forte rétention spatiale à ces stades de vie.

La valeur du SDI global de 0,78 s'est avérée être bien supérieure à la valeur seuil de 0,66 indiquant un stock fortement structuré (**Fig. 5.1**).

Ainsi, cette approche holistique a mis en évidence l'existence d'une structure spatiale du stock de Manche Est. La forte synergie des différents traceurs a permis de renforcer l'hypothèse de départ d'un stock structuré en 3 sous-unités. De plus, la complémentarité des traceurs et de leurs résolutions spatiales et temporelles s'est avérée particulièrement importante dans la compréhension de la structure spatiale de ce stock. En effet, une approche génétique (échelle évolutive) a montré des résultats de structure spatiale en trois sous-unités similaires aux résultats de l'approche de croissance populationnelle (échelle générationnelle) et de l'approche de formes des otolithes (échelle du cycle de vie). Cette concordance des résultats issus des différentes approches nous informe sur la structure spatiale à long terme du stock de sole commune de Manche Est.

Cumulée à une faible connectivité induite par les larves et les juvéniles, la faible connectivité induite par les stades préadulte et adulte contribue à structurer spatialement le stock de sole de la Manche Est. Une telle structure en trois sous-unités faiblement connectées renvoie à un fonctionnement en métapopulation (Kritzer and Sale, 2004) qui se doit d'être considérée dans le processus d'évaluation et de gestion du stock afin d'atteindre les objectifs d'une exploitation durable (Hilborn and Walters, 1992; Tuck and Possingham, 1993; Ying *et al.*, 2011; Goethel *et al.*, 2016), *a fortiori* au vu de sa surexploitation avérée (ICES, 2018a).

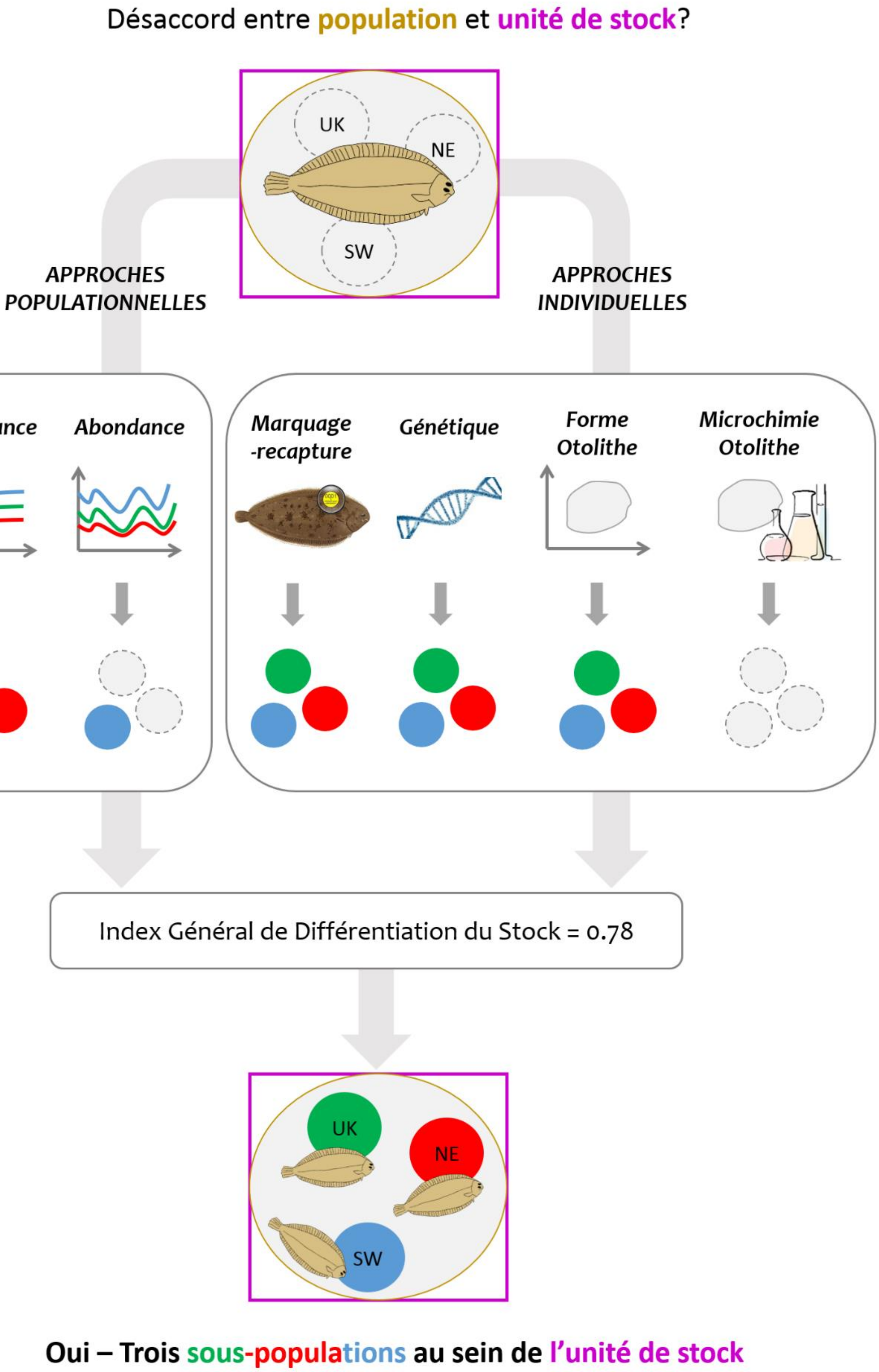


Figure. 5.1. Schéma récapitulatif de l'approche holistique multitraceur appliquée à la sole commune de Manche Est.

5.3. Les perspectives d’évaluation et de gestion

Le SDI calculé au cours du **chapitre 4** est une simplification de l’état de structure du stock puisqu’il ne tient pas compte des processus écologiques sous-jacents. En revanche, le SDI a permis de mettre en lumière un état de structure spatiale qui ne devrait pas être négligé dans l’évaluation et la gestion du stock. En 2017, un benchmark avait suggéré qu’une structure spatiale au sein du stock de sole de Manche Est était possible et que celle-ci devrait être considérée dans la gestion (ICES, 2017a).

Cette thèse a permis de mettre en évidence la structure de type métapopulation pour la sole commune de Manche Est. Bien que la connectivité entre nourriceries et frayères n’ait pas pu être résolue via la microchimie des otolithes, le niveau de ségrégation entre les sous-unités SW, NE et UK s’est avéré élevé d’après l’approche holistique multitraceur menée sur les stades préadulte et adulte. L’étape suivant l’analyse de la structure spatiale d’un stock consiste à en intégrer les acquis pour son évaluation et de sa gestion (Kerr *et al.,* 2017). Dans le cas présent, **le décalage entre l’unité de stock VIId et la métapopulation de sole commune va dans le sens d’une évaluation et d’une gestion actuellement inadaptée à la réalité biologique**.

Parmi les options envisageables, la remise en cause de la délimitation du stock n’est certainement pas la stratégie la plus simple à adopter (Kerr *et al.*, 2017; Cadrin, 2020). Elle impliquerait, outre un travail accru des scientifiques qui réalisent ces évaluations, de redécouper les limites du stock VIId avec des conséquences à l’échelle des différentes pêcheries ciblant ou non la sole commune dans cette zone. Les délimitations des stocks sont des décisions politiques dont les enjeux dépassent la connaissance de l’écologie des espèces exploitées (Bosley *et al.*, 2019).

Une autre option envisageable serait d’intégrer au modèle d’évaluation de stock une dimension spatiale, comme c’est le cas des modèles spatialement explicites d’évaluation (Cadrin and Secor, 2009; Berger *et al.*, 2017; Punt, 2019; Cadrin, 2020). L’intérêt de ce type de modèles est qu’ils permettent d’intégrer comme données d’entrées additionnelles des informations sur la structure du stock et d’estimer leurs conséquences sur l’évaluation, notamment pour l’estimation des points de références (Goethel and Berger, 2017). Cependant, malgré la pertinence écologique de ce type de modèle, ils n’ont pas été largement adoptés à ce jour dans les évaluations de stocks de l’atlantique nord (Kerr *et al.*, 2017). Ces modèles sont en effet très exigeants en données d’entrées (augmentation du coût des suivis) et requièrent une connaissance fine de la structure des populations (Goethel *et al.*, 2015). Un modèle spatialisé de cycle de vie de la sole commune de Manche Est avait été développé en considérant une ségrégation spatiale, alors hypothétique, en trois sous-unités (Archambault *et al.*, 2016). Ce modèle avait permis de questionner les biais liés à la structure du stock sur les points de référence. D’après cette étude, sous l’hypothèse d’un stock unique et homogène, l’exploitation serait supérieure au RMD (Rendement Maximum Durable) avec un rapport $F/F_{RMD} = 1{,}8$. Sous l’hypothèse de trois sous-unités, l’exploitation du stock paraissait contrastée avec une exploitation au-dessus du RMD en zone NE et UK (i.e. $F/F_{RMD} = 2$ et 1,9, respectivement) et proche du RMD en zone SW (i.e. $F/F_{RMD} = 1{,}05$). La prise en compte de la structure de type métapopulation de la sole commune en Manche Est mise en évidence dans cette thèse est donc nécessaire afin de fournir des estimations non biaisées des points de référence. Cependant, la modification du modèle d’évaluation actuel sera compliquée à mettre en œuvre, d’autant qu’une autre limite majeure dans l’application de ces modèles provient de l’inertie institutionnelle (Punt, 2019). Des changements profonds dans le processus d’évaluation prennent nécessairement plusieurs années.

Un compromis envisageable, au moins à court/moyen terme, consisterait à ajuster le niveau d’exploitation à la plus faible productivité parmi les trois sous-populations (Archambault et al., 2016). Cela permettrait d’éviter la surexploitation des sous-unités les moins productives comme c’est souvent le cas lorsque la structure spatiale d’un stock est ignorée (Tuck and Possingham, 1993; Fu and Fanning, 2004; Cadrin and Secor, 2009; Ying *et al.*, 2011; Goethel and Berger, 2017). Alternativement, une gestion spatialisée de l’effort de pêche ou des quotas pourrait être prodiguée afin de s’adapter aux productivités locales, contrastées en Manche Est.

Dans cette perspective, ces propositions de gestion locales nécessiteraient d’être discutées du point de vue des besoins en données et des prérequis de modélisation, mais aussi du point de vue social, économique et institutionnel (Punt, 2019). Une évaluation quantitative des conséquences des scénarii de gestion alternative pourrait être menée dans le cadre d’une MSE (Management Strategy Evaluation). Les MSE sont particulièrement pertinentes dans l’évaluation des conséquences à la fois biologiques et économiques de toute une gamme de stratégies de gestion (Sainsbury *et al.*, 2000; Bunnefeld *et al.*, 2011; Kerr *et al.*, 2017). L’utilisation de cette interface entre biologie, pêche et gestion des ressources devrait permettre une amélioration du processus d’évaluation et de gestion de la sole commune de Manche Est.

References

References

Abaunza, P., Murta, A. G., Campbell, N., Cimmaruta, R., Comesaña, A. S., Dahle, G., García Santamaría, M. T., *et al.* 2008. Stock identity of horse mackerel (*Trachurus trachurus*) in the Northeast Atlantic and Mediterranean Sea: Integrating the results from different stock identification approaches. Fisheries Research, 89: 196–209.

Allaya, H., FALEH, A. B., Rebaya, M., Zrelli, S., Hajjej, G., Hattour, A., Quignard, J.-P., *et al.* 2016. Identification of Atlantic Chub mackerel *Scomber colias* population through the analysis of body shape in Tunisian waters. Cahier de Biologie Marine, 57: 195–207.

Ames, E. P., and Lichter, J. 2013. Gadids and Alewives: Structure within complexity in the Gulf of Maine. Fisheries Research, 141: 70–78.

Andrews. 2010. FastQC A Quality Control tool for High Throughput Sequence Data. Available online at: http://www.bioinformatics.babraham.ac.uk/projects/fastqc/.

Archambault, B., Le Pape, O., Baulier, L., Vermard, Y., Véron, M., and Rivot, E. 2016. Adult-mediated connectivity affects inferences on population dynamics and stock assessment of nursery-dependent fish populations. Fisheries Research, 181: 198–213.

Archambault, B., Rivot, E., Savina, M., and Le Pape, O. 2018. Using a spatially structured life cycle model to assess the influence of multiple stressors on an exploited coastal-nursery-dependent population. Estuarine, Coastal and Shelf Science, 201: 95–104.

Ashford, J., La Mesa, M., Fach, B. A., Jones, C., and Everson, I. 2010. Testing early life connectivity using otolith chemistry and particle-tracking simulations. Canadian Journal of Fisheries and Aquatic Sciences, 67: 1303–1315.

Association Française d'Halieutique. 2016. Fixation des quotas de pêche 2017: pour de trop nombreux stocks les ministres ne pourront pas suivre l'avis scientifique. Available online at: https://www.association-francaise-halieutique.fr/wp-content/uploads/Fixation_des_quotas_de_p%C3%AAche_Note_danalyse_de_lAFH.pdf.

Bath, G. E., Thorrold, S. R., Jones, C. M., Campana, S. E., McLaren, J. W., and Lam, J. W. 2000. Strontium and barium uptake in aragonitic otoliths of marine fish. Geochimica et cosmochimica acta, 64: 1705–1714.

Baudron, A. R., Needle, C. L., Rijnsdorp, A. D., and Tara Marshall, C. 2014. Warming temperatures and smaller body sizes: synchronous changes in growth of North Sea fishes. Global Change Biology, 20: 1023–1031.

Beger, M., McGowan, J., Treml, E. A., Green, A. L., White, A. T., Wolff, N. H., Klein, C. J., *et al.* 2015. Integrating regional conservation priorities for multiple objectives into national policy. Nature Communications, 6: 8208.

Begg, G. A., Cameron, D. S., and Sawynok, W. 1997. Movements and stock structure of school mackerel (*Scomberomorus queenslandicus*) and spotted mackerel (*S. munroi*) in Australian east-coast waters. Marine and Freshwater Research, 48: 295–301.

Begg, G. A., and Waldman, J. R. 1999. An holistic approach to fish stock identification. Fisheries Research, 43: 35–44.

Begg, G. A., Friedland, K. D., and Pearce, J. B. 1999a. Stock identification and its role in stock assessment and fisheries management: an overview. Fisheries Research, 43: 1–8.

Begg, G. A., Hare, J. A., and Sheehan, D. D. 1999b. The role of life history parameters as indicators of stock structure. Fisheries Research, 43: 141–163.

Begg, G. A. 2005. Chapter 6 - Life History Parameters. *In* Stock identification methods, pp. 119–150. Academic Press.

Berger, A. M., Goethel, D. R., Lynch, P. D., Quinn, T., Mormede, S., McKenzie, J., and Dunn, A. 2017. Space oddity: The mission for spatial integration. Canadian Journal of Fisheries and Aquatic Sciences, 74: 1698–1716.

Bernhardt, J. R., and Leslie, H. M. 2013. Resilience to climate change in coastal marine ecosystems. Annual Review of Marine Science, 5: 371–392.

Bjørnstad, O. N., Ims, R. A., and Lambin, X. 1999. Spatial population dynamics: analyzing patterns and processes of population synchrony. Trends in Ecology & Evolution, 14: 427–432.

Blanchet, F. G., Legendre, P., and Borcard, D. 2008. Forward selection of explanatory variables. Ecology, 89: 2623–2632.

Boje, J., Brown, E. J., Davies, J. O., Edelvang, K., Frandsen, R., Jakob, H., Herrmann, B., *et al.* 2019. Improvement of the biological advice for Common Sole in Danish waters. DTU Aqua Report, 337–2019.

Bolger, A. M., Lohse, M., and Usadel, B. 2014. Trimmomatic: a flexible trimmer for Illumina sequence data. Bioinformatics, 30: 2114–2120.

Bosley, K. M., Goethel, D. R., Berger, A. M., Deroba, J. J., Fenske, K. H., Hanselman, D. H., Langseth, B. J., *et al.* 2019. Overcoming challenges of harvest quota allocation in spatially structured populations. Fisheries Research, 220: 105344.

Boström, C., Pittman, S. J., Simenstad, C., and Kneib, R. T. 2011. Seascape ecology of coastal biogenic habitats: advances, gaps, and challenges. Marine Ecology Progress Series, 427: 191–217.

Botsford, L. W., and Paulsen, C. M. 2000. Assessing covariability among populations in the presence of intraseries correlation: Columbia River spring-summer Chinook salmon (*Oncorhynchus tshawytscha*) stocks. Canadian Journal of Fisheries and Aquatic Sciences, 57: 616–627.

Brunel, T., and Dickey-Collas, M. 2010. Effects of temperature and population density on von Bertalanffy growth parameters in Atlantic herring: a macro-ecological analysis. Marine Ecology Progress Series, 405: 15–28.

Bryan-Brown, D. N., Brown, C. J., Hughes, J. M., and Connolly, R. M. 2017. Patterns and trends in marine population connectivity research. Marine Ecology Progress Series, 585: 243–256.

Bunnefeld, N., Hoshino, E., and Milner-Gulland, E. J. 2011. Management strategy evaluation: a powerful tool for conservation? Trends in Ecology & Evolution, 26: 441–447.

Burnham, K. P., and Anderson, D. R. 2003. Model selection and multimodel inference: A practical information-theoretic approach. Springer Science & Business Media. 512 pp.

Burt, G. J., and Millner, R. S. 2008. Movements of sole in the southern North Sea and eastern English Channel from tagging studies (1955–2004). Science Series Technical, Report, 144.

Cadrin, S. X., and Secor, D. H. 2009. Accounting for spatial population structure in stock assessment: Past, present, and future. *In* The future of fisheries science in North America, pp. 405–426. Academic Press.

Cadrin, S. X., Bernreuther, M., Daníelsdóttir, A. K., Hjörleifsson, E., Johansen, T., Kerr, L., Kristinsson, K., *et al.* 2010. Population structure of beaked redfish, *Sebastes mentella*: evidence of divergence associated with different habitats. ICES Journal of Marine Science, 67: 1617–1630.

Cadrin, S. X., Kerr, L. A., and Mariani, S. 2013. Stock identification methods: Applications in fishery science. Academic Press. 589 pp.

Cadrin, S. X., Kerr, L. A., and Mariani, S. 2014. Chapter twenty two - Interdisciplinary evaluation of spatial population structure for definition of fishery management units. *In* Stock identification methods (Second Edition), pp. 535–552. Academic Press.

Cadrin, S. X. 2020. Defining spatial structure for fishery stock assessment. Fisheries Research, 221: 105397.

Campana, Gagné, and McLaren. 1995. Elemental fingerprinting of fish otoliths using ID-ICPMS. Marine Ecology Progress Series, 122: 115–120.

Campana, S. 2001. Accuracy, precision and quality control in age determination, including a review of the use and abuse of age validation methods. Journal of Fish Biology, 59: 197–242.

Campana, S. E., and Casselman, J. M. 1993. Stock discrimination using otolith shape analysis. Canadian Journal of Fisheries and Aquatic Sciences, 50: 1062–1083.

Campana, S. E. 1999. Chemistry and composition of fish otoliths: pathways, mechanisms and applications. Marine Ecology Progress Series, 188: 263–297.

Campana, S. E., Chouinard, G. A., Hanson, J. M., Frechet, A., and Brattey, J. 2000. Otolith elemental fingerprints as biological tracers of fish stocks. Fisheries Research, 46: 343–357.

Cardinale, M., Doering-Arjes, P., Kastowsky, M., and Mosegaard, H. 2004. Effects of sex, stock, and environment on the shape of known-age Atlantic cod (*Gadus morhua*) otoliths. Canadian Journal of Fisheries and Aquatic Sciences, 61: 158–167.

Carson, H. S., Cook, G. S., López-Duarte, P. C., and Levin, L. A. 2011. Evaluating the importance of demographic connectivity in a marine metapopulation. Ecology, 92: 1972–1984.

Casella, G., and Berger, R. L. 2002. Statistical inference. Duxbury, Pacific Grove. 660 pp.

Casselman, J. M., Collins, J. J., Grossman, E. J., Ihssen, P. E., and Spangler, G. R. 1981. Lake Whitefish (*Coregonus clupeaformis*) Stocks of the Ontario waters of Lake Huron. Canadian Journal of Fisheries and Aquatic Sciences, 38: 1772–1789.

Catalano, S. R., Whittington, I. D., Donnellan, S. C., and Gillanders, B. M. 2014. Parasites as biological tags to assess host population structure: Guidelines, recent genetic advances and comments on a holistic approach. International Journal for Parasitology: Parasites and Wildlife, 3: 220–226.

Catchen, J., Hohenlohe, P. A., Bassham, S., Amores, A., and Cresko, W. A. 2013. Stacks: an analysis tool set for population genomics. Molecular Ecology, 22: 3124–3140.

Chen, K.-Y., Marschall, E. A., Sovic, M. G., Fries, A. C., Gibbs, H. L., and Ludsin, S. A. 2018. *assignPOP:* An R package for population assignment using genetic, non-genetic, or integrated data in a machine-learning framework. Methods in Ecology and Evolution, 9: 439–446.

Cheung, W. W. L., Lam, V. W. Y., Sarmiento, J. L., Kearney, K., Watson, R., and Pauly, D. 2009. Projecting global marine biodiversity impacts under climate change scenarios. Fish and Fisheries, 10: 235–251.

Chin, A., Simpfendorfer, C. A., White, W. T., Johnson, G. J., McAuley, R. B., and Heupel, M. R. 2017. Crossing lines: a multidisciplinary framework for assessing connectivity of hammerhead sharks across jurisdictional boundaries. Scientific Reports, 7: 46061.

Chong, Z., Ruan, J., and Wu, C.-I. 2012. Rainbow: an integrated tool for efficient clustering and assembling RAD-seq reads. Bioinformatics, 28: 2732–2737.

Ciannelli, L., Knutsen, H., Olsen, E. M., Espeland, S. H., Asplin, L., Jelmert, A., Knutsen, J. A., *et al.* 2010. Small-scale genetic structure in a marine population in relation to water circulation and egg characteristics. Ecology, 91: 2918–2930.

Ciannelli, L., Fisher, J. A. D., SkernMauritzen, M., Hunsicker, M. E., Hidalgo, M., Frank, K. T., and Bailey, K. M. 2013. Theory, consequences and evidence of eroding population spatial structure in harvested marine fishes: a review. Marine Ecology Progress Series, 480: 227–243.

Coggan, R. A., and Dando, P. R. 1988. Movements of juvenile Dover sole, *Solea solea* (L.), in the Tamar Estuary, South-western England. Journal of Fish Biology, 33: 177–184.

Cope, J. M., and Punt, A. E. 2009. Drawing the lines: resolving fishery management units with simple fisheries data. Canadian Journal of Fisheries and Aquatic Sciences, 66: 1256–1273.

Costello, M. J., and Connor, D. W. 2019. Connectivity is generally not important for marine reserve planning. Trends in Ecology & Evolution, 34: 686–688.

Cowen, R., Gawarkiewicz, G., Pineda, J., Thorrold, S., and Werner, F. 2007. Population connectivity in marine systems: An Overview. Oceanography, 20: 14–21.

Cowen, R. K., and Sponaugle, S. 2009. Larval dispersal and marine population connectivity. Annual Review of Marine Science, 1: 443–466.

Cruz, V. P., Vera, M., Pardo, B. G., Taggart, J., Martinez, P., Oliveira, C., and Foresti, F. 2017. Identification and validation of single nucleotide polymorphisms as tools to detect hybridization and population structure in freshwater stingrays. Molecular Ecology Resources, 17: 550–556.

Cuveliers, E. L., Larmuseau, M. H. D., Hellemans, B., Verherstraeten, S. L. N. A., Volckaert, F. A. M., and Maes, G. E. 2012. Multi-marker estimate of genetic connectivity of sole (*Solea solea*) in the North-East Atlantic Ocean. Marine Biology, 159: 1239–1253.

D'Aloia, C. C., Bogdanowicz, S. M., Francis, R. K., Majoris, J. E., Harrison, R. G., and Buston, P. M. 2015. Patterns, causes, and consequences of marine larval dispersal. Proceedings of the National Academy of Sciences, 112: 13940–13945.

Danecek, P., Auton, A., Abecasis, G., Albers, C. A., Banks, E., DePristo, M. A., Handsaker, R. E., *et al.* 2011. The variant call format and VCFtools. Bioinformatics, 27: 2156–2158.

Darnaude, A., and Hunter, E. 2008. Coupled use of data storage tags and otolith microchemistry to assess population dispersal and intra-specific diversity in migratory behaviour in North Sea plaice (*Pleuronectes platessa* L.). Comparative Biochemistry and Physiology-Part A Molecular & Integrative Physiology, 150(3), S205.

Darnaude, A. M., Sturrock, A., Trueman, C. N., Mouillot, D., EIMF, Campana, S. E., and Hunter, E. 2014. Listening In on the Past: What can otolith δ18O values really tell us about the environmental history of fishes? PLOS ONE, 9: e108539.

de Vries, M. C., Gillanders, B. M., and Elsdon, T. S. 2005. Facilitation of barium uptake into fish otoliths: Influence of strontium concentration and salinity. Geochimica et Cosmochimica Acta, 69: 4061–4072.

Delerue-Ricard, S., Stynen, H., Barbut, L., Morat, F., Mahé, K., Hablützel, P. I., Hostens, K., *et al.* 2018. Size-effect, asymmetry, and small-scale spatial variation in otolith shape of juvenile sole in the Southern North Sea. Hydrobiologia, 1-14.

Diopere, E., Vandamme, S. G., Hablützel, P. I., Cariani, A., Van Houdt, J., Rijnsdorp, A., Tinti, F., *et al.* 2018. Seascape genetics of a flatfish reveals local selection under high levels of gene flow. ICES Journal of Marine Science, *75*(2), 675-689.

Dorazio, R. M. 2009. On selecting a prior for the precision parameter of Dirichlet process mixture models. Journal of Statistical Planning and Inference, 139: 3384–3390.

Dorel, D., Koutsikopoulos, C., Desaunay, Y., and Marchand, J. 1991. Seasonal distribution of young sole (*Solea solea* (L.)) in the nursery ground of the bay of Vilaine (Northern Bay Of Biscay). Netherlands Journal of Sea Research, 27: 297–306.

Du Pontavice, H., Randon, M., Lehuta, S., Vermard, Y., and Savina-Rolland, M. 2018. Investigating spatial heterogeneity of von Bertalanffy growth parameters to inform the stock structuration of common sole, *Solea solea*, in the Eastern English Channel. Fisheries Research, 207: 28–36.

Durieux, E. D. H., Duigou, M. L., Millot, S., Sasal, P., and Begout, M.-L. 2010. Sedentary behaviour establishment in O-group common sole *Solea solea*: a laboratory video-tracking study. Journal of the Marine Biological Association of the United Kingdom, 90: 1257–1262.

Elliott, N. G., Haskard, K., and Koslow, J. A. 1995. Morphometric analysis of orange roughy (*Hoplostethus atlanticus*) off the continental slope of southern Australia. Journal of Fish Biology, 46: 202–220.

Elsdon, T. S., and Gillanders, B. M. 2002. Interactive effects of temperature and salinity on otolith chemistry: challenges for determining environmental histories of fish. Canadian Journal of Fisheries and Aquatic Sciences, 59: 1796–1808.

Enberg, K., Jørgensen, C., Dunlop, E. S., Varpe, Ø., Boukal, D. S., Baulier, L., Eliassen, S., *et al.* 2012. Fishing-induced evolution of growth: concepts, mechanisms and the empirical evidence. Marine Ecology, 33: 1–25.

Erlandsson, J., Östman, Ö., Florin, A.-B., and Pekcan-Hekim, Z. 2017. Spatial structure of body size of European flounder (*Platichthys flesus* L.) in the Baltic Sea. Fisheries Research, 189: 1–9.

Esler, D. 2000. Applying metapopulation theory to conservation of migratory birds. Conservation Biology, 14: 366–372.

Exadactylos, A., Geffen, A. J., Panagiotaki, P., and Thorpe, J. P. 2003. Population structure of Dover sole *Solea solea*: RAPD and allozyme data indicate divergence in European stocks. Marine Ecology Progress Series, 246: 253–264.

Fischer, A. J., and Thompson, B. A. 2004. The age and growth of southern flounder, *Paralichthys lethostigma*, from Louisiana estuarine and offshore waters. Bulletin of Marine Science, 75(1), 63-77.

Frisk, M. G., Jordaan, A., and Miller, T. J. 2014. Moving beyond the current paradigm in marine population connectivity: are adults the missing link? Fish and Fisheries, 15: 242–254.

Fu, C., and Fanning, L. P. 2004. Spatial considerations in the management of Atlantic cod off Nova Scotia, Canada. North American Journal of Fisheries Management, 24: 775–784.

Fu, L., Niu, B., Zhu, Z., Wu, S., and Li, W. 2012. CD-HIT: accelerated for clustering the next-generation sequencing data. Bioinformatics, 28: 3150–3152.

Garrison, E., and Marth, G. 2012. Haplotype-based variant detection from short-read sequencing. arXiv preprint arXiv:1207.3907.

Gelman, A., and Rubin, D. B. 1992. Inference from iterative simulation using multiple sequences. Statistical science: 457–472.

Gibson, R. N., Nash, R. D. M., Geffen, A. J., and Veer, H. W. V. der. 2014. Flatfishes: Biology and exploitation. John Wiley & Sons. 579 pp.

Gillanders, B. 2002a. Connectivity between juvenile and adult fish populations: do adults remain near their recruitment estuaries? Marine Ecology Progress Series, 240: 215–223.

Gillanders, B. M., and Kingsford, M. J. 2000. Elemental fingerprints of otoliths of fish may distinguish estuarine nursery habitats. Marine Ecology Progress Series, 201: 273–286.

Gillanders, B. M. 2002b. Temporal and spatial variability in elemental composition of otoliths: implications for determining stock identity and connectivity of populations. Canadian Journal of Fisheries and Aquatic Sciences, 59: 669–679.

Glover, K. A., Skilbrei, O. T., and Skaala, Ø. 2008. Genetic assignment identifies farm of origin for Atlantic salmon *Salmo salar* escapees in a Norwegian fjord. ICES Journal of Marine Science, 65: 912–920.

Goethel, D. R., II, T. J. Q., and Cadrin, S. X. 2011. Incorporating spatial structure in stock assessment: movement modeling in marine fish population dynamics. Reviews in Fisheries Science, 19: 119–136.

Goethel, D. R., Legault, C. M., Cadrin, S. X., and Wilberg, M. 2015. Testing the performance of a spatially explicit tag-integrated stock assessment model of yellowtail flounder (*Limanda ferruginea*) through simulation analysis. Canadian Journal of Fisheries and Aquatic Sciences, 72: 582–601.

Goethel, D. R., Kerr, L. A., and Cadrin, S. X. 2016. Incorporating spatial population structure into the assessment-management interface of marine resources. Management science in fisheries: an introduction to simulation based methods. Edited by CTT Edwards and DJ Dankel. Routledge, New York: 319–347.

Goethel, D. R., and Berger, A. M. 2017. Accounting for spatial complexities in the calculation of biological reference points: effects of misdiagnosing population structure for stock status indicators. Canadian Journal of Fisheries and Aquatic Sciences, 74: 1878–1894.

Gohin, F., Saulquin, B., Bryère, P., and Hocer, S. 2010. Atlas de la Température, de la concentration en chlorophylle et de la turbidité de surface du plateau continental français et de ses abords de l'ouest européen. Ifremer. Available online at: http://archimer. ifremer. fr/doc/00057/16840.

Goudet, J. 2005. hierfstat, a package for r to compute and test hierarchical F-statistics. Molecular Ecology Notes, 5: 184–186.

Graf, W., and Baker, R. 1983. Adaptive changes of the vestibulo-ocular reflex in flatfish are achieved by reorganization of central nervous pathways. Science, 221: 777–779.

Grahl-Nielsen, O. 2005. Fatty acid profiles as natural marks for stock identification. *In* Stock identification methods, pp. 247–269. Elsevier.

Grimm, V., Reise, K., and Strasser, M. 2003. Marine metapopulations: a useful concept? Helgoland Marine Research, 56: 222–228.

Grioche, A. 1998. Dynamique de l'écophase ichtyoplanctonique en Manche orientale et sud Mer du Nord. Approche multispécifique et description de deux espèces cibles: *Solea solea* (L.) et *Pleuronectes flesus* (L.). Littoral.

Hanski, I., and Thomas, C. D. 1994. Metapopulation dynamics and conservation: A spatially explicit model applied to butterflies. Biological Conservation, 68: 167–180.

Hanski, I., and Simberloff, D. 1997. The metapopulation approach, its history, conceptual domain, and application to conservation. Metapopulation dynamics: ecology, genetics, and evolution: 5–26.

Hanski, I. 1999. Metapopulation ecology. OUP Oxford. 332 pp.

Hanski, I., and Ovaskainen, O. 2000. The metapopulation capacity of a fragmented landscape. Nature, 404: 755–758.

Hanski, I., and Gaggiotti, O. E. 2004. Ecology, genetics, and evolution of metapopulations. Academic Press. 718 pp.

Harrison, S., and Taylor, A. D. 1997. Empirical evidence for metapopulation dynamics. Metapopulation biology: ecology, genetics, and evolution: 27–42.

Hauser, L., and Carvalho, G. R. 2008. Paradigm shifts in marine fisheries genetics: ugly hypotheses slain by beautiful facts. Fish and Fisheries, 9: 333–362.

Hawkins, S. J., Bohn, K., Sims, D. W., Ribeiro, P., Faria, J., Presa, P., Pita, A., *et al.* 2016. Fisheries stocks from an ecological perspective: Disentangling ecological connectivity from genetic interchange. Fisheries Research, 179: 333–341.

Hazen, E. L., Carlisle, A. B., Wilson, S. G., Ganong, J. E., Castleton, M. R., Schallert, R. J., Stokesbury, M. J. W., *et al.* 2016. Quantifying overlap between the Deepwater Horizon oil spill and predicted bluefin tuna spawning habitat in the Gulf of Mexico. Scientific Reports, 6: 33824.

Heard, G. W., McCarthy, M. A., Scroggie, M. P., Baumgartner, J. B., and Parris, K. M. 2013. A Bayesian model of metapopulation viability, with application to an endangered amphibian. Diversity and Distributions, 19: 555–566.

Hearn, A. R., Green, J., Román, M. H., Acuña-Marrero, D., Espinoza, E., and Klimley, A. P. 2016. Adult female whale sharks make long-distance movements past Darwin Island (Galapagos, Ecuador) in the Eastern Tropical Pacific. Marine Biology, 163: 214.

Heino, M., Kaitala, V., Ranta, E., and Lindström, J. 1997. Synchronous dynamics and rates of extinction in spatially structured populations. Proceedings of the Royal Society of London B: Biological Sciences, 264: 481–486.

Helyar, S. J., Hemmer-Hansen, J., Bekkevold, D., Taylor, M. I., Ogden, R., Limborg, M. T., Cariani, A., *et al.* 2011. Application of SNPs for population genetics of nonmodel organisms: new opportunities and challenges. Molecular Ecology Resources, 11: 123–136.

Hilborn, R., and Walters, C. J. 1992. Quantitative fisheries stock assessment: Choice, dynamics and uncertainty. Reviews in Fish Biology and Fisheries, 2: 177–178.

Hilborn, R., Quinn, T. P., Schindler, D. E., and Rogers, D. E. 2003. Biocomplexity and fisheries sustainability. Proceedings of the National Academy of Sciences, 100: 6564–6568.

Hixon, M. A. 2011. 60 Years of coral reef fish ecology: past, present, future. Bulletin of Marine Science, 87(4), 727-765.

Holmes, E., Ward, E., and Wills, K. 2013. MARSS: Multivariate autoregressive state-space modeling. R package version, 3.

Holt, R. D. 1997. From metapopulation dynamics to community structure: Some consequences of spatial heterogeneity. *In* Metapopulation biology, pp. 149–164. Academic Press.

Horwood, J. 1993. The Bristol Channel sole (*Solea solea* (L.)): A fisheries case study. *In* Advances in marine biology, pp. 215–367. Ed. by J. H. S. Blaxter and A. J. Southward. Academic Press.

Hsieh, C., Reiss, C. S., Hunter, J. R., Beddington, J. R., May, R. M., and Sugihara, G. 2006. Fishing elevates variability in the abundance of exploited species. Nature, 443: 859–862.

Hunter, A., Speirs, D. C., and Heath, M. R. 2016. Investigating trends in the growth of five demersal fish species from the Firth of Clyde and the wider western shelf of Scotland. Fisheries Research, 177: 71–81.

Hüssy, K., Mosegaard, H., Albertsen, C. M., Nielsen, E. E., Hemmer-Hansen, J., and Eero, M. 2016. Evaluation of otolith shape as a tool for stock discrimination in marine fishes using Baltic Sea cod as a case study. Fisheries Research, 174: 210–218.

Hutchings, J. A. 2005. Life history consequences of overexploitation to population recovery in Northwest Atlantic cod (*Gadus morhua*). Canadian Journal of Fisheries and Aquatic Sciences, 62: 824–832.

ICES. 2009. Manual for the Offshore Beam Trawl Surveys, Revision 1.2. Working Group on Beam Trawl Surveys.

ICES. 2017a. Report of the benchmark workshop on the North Sea stocks (WKNSEA). ICES CM 2017/ACOM: 34. Copenhagen, Denmark.

ICES. 2017b. Report of the working group on the assessment of demersal stocks in the North Sea and Skagerrak. ICES HQ. ICES CM 2017/ACOM: 21.

ICES. 2018a. Sole (*Solea solea*) in Division 7.d (Eastern English Channel).

ICES. 2018b, June 29. ICES Advice on fishing opportunities, catch, and effort Greater North Sea Ecoregion. Sole (*Solea solea*) in Division 7.d (Eastern English Channel).

ICES. 2018c. Interim report of the stock identification methods working group (SIMWG). Portland, USA.

Ihssen, P. E., Booke, H. E., Casselman, J. M., McGlade, J. M., Payne, N. R., and Utter, F. M. 1981. Stock identification: Materials and methods. Canadian Journal of Fisheries and Aquatic Sciences, 38: 1838–1855.

Inchausti, P., and Weimerskirch, H. 2002. Dispersal and metapopulation dynamics of an oceanic seabird, the wandering albatross, and its consequences for its response to long-line fisheries. Journal of Animal Ecology, 71: 765–770.

Izzo, C., Doubleday, Z. A., Schultz, A. G., Woodcock, S. H., and Gillanders, B. M. 2015. Contribution of water chemistry and fish condition to otolith chemistry: comparisons across salinity environments. Journal of Fish Biology, 86: 1680–1698.

Izzo, C., Ward, T. M., Ivey, A. R., Suthers, I. M., Stewart, J., Sexton, S. C., and Gillanders, B. M. 2017. Integrated approach to determining stock structure: implications for fisheries management of sardine, *Sardinops sagax*, in Australian waters. Reviews in Fish Biology and Fisheries, 27: 267–284.

Izzo, C., Reis-Santos, P., and Gillanders, B. M. 2018. Otolith chemistry does not just reflect environmental conditions: A meta-analytic evaluation. Fish and Fisheries, 19: 441–454.

Jacobsen, J. A., and Hansen, L. P. 2005. Chapter 20 - Internal and external tags. *In* Stock identification methods, pp. 415–433. Academic Press.

Jasonowicz, A. J., Goetz, F. W., Goetz, G. W., and Nichols, K. M. 2016. Love the one you're with: genomic evidence of panmixia in the sablefish (*Anoplopoma fimbria*). Canadian Journal of Fisheries and Aquatic Sciences, 74: 377–387.

Joensen, H., and Grahl-Nielsen, O. 2004. Stock structure of *Sebastes mentell*a in the North Atlantic revealed by chemometry of the fatty acid profile in heart tissue. ICES Journal of Marine Science, 61: 113–126.

Jombart, T. 2008. adegenet: a R package for the multivariate analysis of genetic markers. Bioinformatics, 24: 1403–1405.

Jombart, T., Devillard, S., and Balloux, F. 2010. Discriminant analysis of principal components: a new method for the analysis of genetically structured populations. BMC Genetics, 11: 94.

Kamvar, Z., Tabima, J., and Grünwald, N. 2014. Poppr: an R package for genetic analysis of populations with clonal, partially clonal, and/or sexual reproduction.

Kerr, L. A., Cadrin, S. X., and Secor, D. H. 2010a. The role of spatial dynamics in the stability, resilience, and productivity of an estuarine fish population. Ecological Applications, 20: 497–507.

Kerr, L. A., Cadrin, S. X., and Secor, D. H. 2010b. Simulation modelling as a tool for examining the consequences of spatial structure and connectivity on local and regional population dynamics. ICES Journal of Marine Science, 67: 1631–1639.

Kerr, L. A., and Goethel, D. R. 2014. Simulation modeling as a tool for synthesis of stock identification information. *In* Stock identification methods, pp. 501–533. Academic Press.

Kerr, L. A., Hintzen, N. T., Cadrin, S. X., Clausen, L. W., Dickey-Collas, M., Goethel, D. R., Hatfield, E. M. C., *et al.* 2017. Lessons learned from practical approaches to reconcile mismatches between biological population structure and stock units of marine fish. ICES Journal of Marine Science, 74: 1708–1722.

Knutsen, H., Jorde, P. E., André, C., and Stenseth, N. C. 2003. Fine-scaled geographical population structuring in a highly mobile marine species: the Atlantic cod. Molecular Ecology, 12: 385–394.

Koutsikopoulos, C., and Lacroix, N. 1992. Distribution and abundance of sole (*Solea solea* (L.)) eggs and larvae in the Bay of Biscay between 1986 and 1989. Netherlands Journal of Sea Research, 29: 81–91.

Kritzer, J. P., and Sale, P. F. 2004. Metapopulation ecology in the sea: from Levins' model to marine ecology and fisheries science. Fish and Fisheries, 5: 131–140.

Kritzer, J. P., and Sale, P. F. 2010. Marine metapopulations. Academic Press. 573 pp.

Kutkuhn, J. H. 1981. Stock Definition as a necessary basis for cooperative management of Great Lakes fish resources. Canadian Journal of Fisheries and Aquatic Sciences, 38: 1476–1478.

Laconcha, U., Iriondo, M., Arrizabalaga, H., Manzano, C., Markaide, P., Montes, I., Zarraonaindia, I., *et al.* 2015. New nuclear SNP markers unravel the genetic structure and effective population size of Albacore tuna (*Thunnus alalunga*). PLOS ONE, 10: e0128247.

Lacroix, G., Maes, G. E., Bolle, L. J., and Volckaert, F. A. 2013. Modelling dispersal dynamics of the early life stages of a marine flatfish (*Solea solea* L.). Journal of Sea Research, 84: 13–25.

Lao, O., Duijn, K. van, Kersbergen, P., Knijff, P. de, and Kayser, M. 2006. Proportioning whole-genome Single-Nucleotide-Polymorphism diversity for the identification of geographic population structure and genetic ancestry. The American Journal of Human Genetics, 78: 680–690.

Law, R. 2007. Fisheries-induced evolution: present status and future directions. Marine Ecology Progress Series, 335: 271–277.

Le Bris, A., Fisher, J. A. D., Murphy, H. M., Galbraith, P. S., Castonguay, M., Loher, T., and Robert, D. 2018. Migration patterns and putative spawning habitats of Atlantic halibut (*Hippoglossus hippoglossus*) in the Gulf of St. Lawrence revealed by geolocation of pop-up satellite archival tags. ICES Journal of Marine Science, 75: 135–147.

Le Pape, O., and Cognez, N. 2016. The range of juvenile movements of estuarine and coastal nursery dependent flatfishes: estimation from a meta-analytical approach. Journal of Sea Research, 107, Part 1: 43–55.

Le Pape, O., Bonhommeau, S., Nieblas, A.-E., and Fromentin, J.-M. 2017. Overfishing causes frequent fish population collapses but rare extinctions. Proceedings of the National Academy of Sciences, 114: E6274–E6274.

Lecomte, J.-B., Le Pape, O., Baillif, H., Nevoux, M., Vermard, Y., Savina-Rolland, M., Veron, M., *et al.* 2019. State-space modeling of multi-decadal mark-recapture data reveals low adult dispersal in a nursery-dependent fish metapopulation. Canadian Journal of Fisheries and Aquatic Sciences.

Legendre, P., and Legendre, L. F. J. 1998. Numerical ecology. Elsevier. 1007 pp.

Leslie, R. W., and Grant, W. S. 1990. Lack of congruence between genetic and morphological stock structure of the southern African anglerfish *Lophius vomerinus*. South African Journal of Marine Science, 9: 379–398.

Levins, R. 1969. Some demographic and genetic consequences of environmental heterogeneity for biological control. Bulletin of the Entomological Society of America, 15: 237–240.

Levins, R. 1970. Extinction - Some mathematical problems in biology. American Mathematical Society, Providence, RI: 77–107.

Li, H. 2013. Aligning sequence reads, clone sequences and assembly contigs with BWA-MEM. arXiv preprint arXiv:1303.3997..

Liebhold, A., Koenig, W. D., and Bjørnstad, O. N. 2004. Spatial synchrony in population dynamics. Annual Review of Ecology, Evolution, and Systematics, 35: 467–490.

Link, J. S., Nye, J. A., and Hare, J. A. 2010. Guidelines for incorporating fish distribution shifts into a fisheries management context. Fish and Fisheries, 12: 461–469.

Liu, N., Chen, L., Wang, S., Oh, C., and Zhao, H. 2005. Comparison of single-nucleotide polymorphisms and microsatellites in inference of population structure. BMC Genetics, 6: S26.

MacKenzie, K., and Abaunza, P. 2014. Chapter ten - Parasites as biological tags. *In* Stock identification methods (Second Edition), pp. 185–203.Academic Press.

Magris, R. A., Pressey, R. L., Weeks, R., and Ban, N. C. 2014. Integrating connectivity and climate change into marine conservation planning. Biological Conservation, 170: 207–221.

Mahé K., Ider D., Massaro A., Hamed O., Jurado-Ruzafa A., Gonçalves P., Anastasopoulou A., Jadaud A., Mytilineou C., Reton M., Elleboode R., Morell A., Ramdane Z., Smith J., Bekaert K., Amara R., de Pontual H., Ernande B., 2019. Directional bilateral asymmetry in fish otolith: is it a potential tool to evaluate the stock boundaries? *In*: Lestrel P.E. (Ed.) Biological shape analysis: Proceedings of the 5th International Symposium of Biological Shape Analysis. World Scientific, Pub Singapore and New Jersey, USA: *In press*

Mahé, K., Oudard, C., Mille, T., Keating, J., Gonçalves, P., Clausen, L. W., Petursdottir, G., *et al.* 2016. Identifying blue whiting (*Micromesistius poutassou*) stock structure in the Northeast Atlantic by otolith shape analysis. Canadian Journal of Fisheries and Aquatic Sciences, 73: 1363–1371.

Marchand, J., and Masson, G. 1989. Process of estuarine colonization by 0-group sole (*Solea solea*): Hydrological conditions, behaviour, and feeding activity in the Vilaine estuary. *In* ICES Marine Science Symposia. 1989.

Marchand, J. 1991. The influence of environmental conditions on settlement, distribution and growth of 0-group sole (*Solea solea* (L.)) in a macrotidal estuary (Vilaine, France). Netherlands Journal of Sea Research, 27: 307–316.

Marengo, M., Baudouin, M., Viret, A., Laporte, M., Berrebi, P., Vignon, M., Marchand, B., *et al.* 2017. Combining microsatellite, otolith shape and parasites community analyses as a holistic approach to assess population structure of *Dentex dentex*. Journal of Sea Research, 128: 1–14.

Martin, G. B., Thorrold, S. R., and Jones, C. M. 2004. Temperature and salinity effects on strontium incorporation in otoliths of larval spot (*Leiostomus xanthurus*). Canadian Journal of Fisheries and Aquatic Sciences, 61: 34–42.

Martin, J., Rougemont, Q., Drouineau, H., Launey, S., Jatteau, P., Bareille, G., Berail, S., *et al.* 2015. Dispersal capacities of anadromous Allis shad population inferred from a coupled genetic and otolith approach. Canadian Journal of Fisheries and Aquatic Sciences, 72: 991–1003.

Mérigot, B., Letourneur, Y., and Lecomte-Finiger, R. 2007. Characterization of local populations of the common sole *Solea solea* (Pisces, Soleidae) in the NW Mediterranean through otolith morphometrics and shape analysis. Marine Biology, 151: 997–1008.

Merino, G., Barange, M., Blanchard, J. L., Harle, J., Holmes, R., Allen, I., Allison, E. H., *et al.* 2012. Can marine fisheries and aquaculture meet fish demand from a growing human population in a changing climate? Global Environmental Change, 22: 795–806.

Milano, I., Babbucci, M., Cariani, A., Atanassova, M., Bekkevold, D., Carvalho, G. R., Espiñeira, M., *et al.* 2014. Outlier SNP markers reveal fine-scale genetic structuring across European hake populations (*Merluccius merluccius*). Molecular Ecology, 23: 118–135.

Mille, T., Mahe, K., Villanueva, M. C., Pontual, H. D., and Ernande, B. 2015. Sagittal otolith morphogenesis asymmetry in marine fishes. Journal of Fish Biology, 87: 646–663.

Mille, T., Mahe, K., Cachera, M., Villanueva, C.-M., De Pontual, H., and Ernande, B. 2016. Diet is correlated with otolith shape in marine fish. Marine Ecology Progress Series, 555: 167–184.

Moll, D., Kotterba, P., Jochum, K. P., von Nordheim, L., and Polte, P. 2019. Elemental inventory in fish otoliths reflects natal origin of Atlantic herring (*Clupea harengus*) from Baltic Sea juvenile areas. Frontiers in Marine Science, 6, 191.

Mollet, F. M., Kraak, S. B. M., and Rijnsdorp, A. D. 2007. Fisheries-induced evolutionary changes in maturation reaction norms in North Sea sole *Solea solea*. Marine Ecology Progress Series, 351: 189–199.

Mollet, F. M., Engelhard, G. H., Vainikka, A., Laugen, A. T., Rijnsdorp, A. D., and Ernande, B. 2013. Spatial variation in growth, maturation schedules and reproductive investment of female sole *Solea solea* in the Northeast Atlantic. Journal of Sea Research, 84: 109–121.

Moran, P. a. P. 1953. The statistical analysis of the Canadian Lynx cycle. Australian Journal of Zoology, 1: 291–298.

Moreira, C., Froufe, E., Sial, A. N., Caeiro, A., Vaz-Pires, P., and Correia, A. T. 2018. Population structure of the blue jack mackerel (*Trachurus picturatus*) in the NE Atlantic inferred from otolith microchemistry. Fisheries Research, 197: 113–122.

Morin, P. A., Luikart, G., Wayne, R. K., and the SNP workshop group. 2004. SNPs in ecology, evolution and conservation. Trends in Ecology & Evolution, 19: 208–216.

Morin, P. A., Martien, K. K., and Taylor, B. L. 2009. Assessing statistical power of SNPs for population structure and conservation studies. Molecular Ecology Resources, 9: 66–73.

Mullins, R. B., McKeown, N. J., Sauer, W. H. H., and Shaw, P. W. 2018. Genomic analysis reveals multiple mismatches between biological and management units in yellowfin tuna (*Thunnus albacares*). ICES Journal of Marine Science, 75: 2145–2152.

Mullon, C., Cury, P., and Penven, P. 2002. Evolutionary individual-based model for the recruitment of anchovy (*Engraulis capensis*) in the southern Benguela. Canadian Journal of Fisheries and Aquatic Sciences, 59: 910–922.

Napoléon, C., Raimbault, V., Fiant, L., Riou, P., Lefebvre, S., Lampert, L., and Claquin, P. 2012. Spatiotemporal dynamics of physicochemical and photosynthetic parameters in the central English Channel. Journal of Sea Research, 69: 43–52.

Neat, F. C., Bendall, V., Berx, B., Wright, P. J., Ó Cuaig, M., Townhill, B., Schön, P.-J., *et al.* 2014. Movement of Atlantic cod around the British Isles: implications for finer scale stock management. Journal of Applied Ecology, 51: 1564–1574.

Neubauer, P., Shima, J. S., and Swearer, S. E. 2013. Inferring dispersal and migrations from incomplete geochemical baselines: analysis of population structure using Bayesian infinite mixture models. Methods in Ecology and Evolution, 4: 836–845.

Nielsen, R., Korneliussen, T., Albrechtsen, A., Li, Y., and Wang, J. 2012. SNP calling, genotype calling, and sample allele frequency estimation from new-generation sequencing data. PLOS ONE, 7: e37558.

Orensanz, J. L., and Jamieson, G. S. 1998. The assessment and management of spatially. *In* Proceedings of the North Pacific symposium on invertebrate stock assessment and management, p. 441. NRC Research Press.

Östman, Ö., Olsson, J., Dannewitz, J., Palm, S., and Florin, A.-B. 2017. Inferring spatial structure from population genetics and spatial synchrony in demography of Baltic Sea fishes: implications for management. Fish and Fisheries, 18: 324–339.

Palaiokostas, C., Bekaert, M., Khan, M. G., Taggart, J. B., Gharbi, K., McAndrew, B. J., and Penman, D. J. 2015. A novel sex-determining QTL in Nile tilapia (*Oreochromis niloticus*). BMC Genomics, 16: 171.

Palumbi, S. R. 2003. Population Genetics, Demographic Connectivity, and the Design of Marine Reserves. Ecological Applications, 13: 146–158.

Parrish, J. 1989. Fish communities of interacting shallow-water habitats in tropical oceanic regions. Marine Ecology Progress Series, 58: 143–160.

Paschou, P., Ziv, E., Burchard, E. G., Choudhry, S., Rodriguez-Cintron, W., Mahoney, M. W., and Drineas, P. 2007. PCA-correlated SNPs for structure identification in worldwide human populations. PLOS Genetics, 3: e160.

Payan, P., De Pontual, H., Bœuf, G., and Mayer-Gostan, N. 2004. Endolymph chemistry and otolith growth in fish. Comptes Rendus Palevol, 3: 535–547.

Pepin, P., and Carr, S. M. 1993. Morphological, meristic, and genetic analysis of stock structure in juvenile Atlantic cod (*Gadus morhua*) from the Newfoundland shelf. Canadian Journal of Fisheries and Aquatic Sciences, 50: 1924–1933.

Peterson, B. K., Weber, J. N., Kay, E. H., Fisher, H. S., and Hoekstra, H. E. 2012. Double Digest RADseq: An inexpensive method for de novo SNP discovery and genotyping in model and non-model species. PLOS ONE, 7: e37135.

Petitgas, P., Rijnsdorp, A. D., Dickey-Collas, M., Engelhard, G. H., Peck, M. A., Pinnegar, J. K., Drinkwater, K., *et al.* 2013. Impacts of climate change on the complex life cycles of fish. Fisheries Oceanography, 22: 121–139.

Pita, A., Casey, J., Hawkins, S. J., Villarreal, M. R., Gutiérrez, M.-J., Cabral, H., Carocci, F., *et al.* 2016. Conceptual and practical advances in fish stock delineation. Fisheries Research, 173, Part 3: 185–193.

Plummer, M. 2003. JAGS: A program for analysis of Bayesian graphical models using Gibbs sampling. In Proceedings of the 3rd international workshop on distributed statistical computing (Vol. 124, No. 125, p. 10).

Punt, A. E. 2019. Spatial stock assessment methods: A viewpoint on current issues and assumptions. Fisheries Research, 213: 132–143.

Puritz, J. B., Hollenbeck, C. M., and Gold, J. R. 2014. dDocent: a RADseq, variant-calling pipeline designed for population genomics of non-model organisms. PeerJ, 2: e431.

Randon, M., Réveillac, E. and Le Pape, O. A holistic investigation of tracers at population and individual scales highlighted population structure for the common sole of the Eastern English Channel. *In prep*.

Randon, M., Le Pape, O. and Réveillac, E. Combining genetic and non-genetic tracers to assess the population structure and connectivity within a flatfish stock. *Submitted* to Estuarine, Coastal and Shelf Science.

Randon, M., Daverat, F., Bareille, G., Jatteau, P., Martin, J., Pecheyran, C., and Drouineau, H. 2017. Quantifying exchanges of Allis shads between river catchments by combining otolith microchemistry and abundance indices in a Bayesian model. ICES Journal of Marine Science, 75: 9–21.

Randon, M., Réveillac, E., Rivot, E., Du Pontavice, H., and Le Pape, O. 2018. Could we consider a single stock when spatial sub-units present lasting patterns in growth and asynchrony in cohort densities? A flatfish case study. Journal of Sea Research, 142: 91–100.

Ranta, E., Kaitala, V., Lindström, J., and Lindén, H. 1995. Synchrony in population dynamics. Proc. R. Soc. Lond. B, 262: 113–118.

Reiss, H., Hoarau, G., Dickey-Collas, M., and Wolff, W. J. 2009. Genetic population structure of marine fish: mismatch between biological and fisheries management units. Fish and Fisheries, 10: 361–395.

Reis-Santos, P., Tanner, S. E., Elsdon, T. S., Cabral, H. N., and Gillanders, B. M. 2013. Effects of temperature, salinity and water composition on otolith elemental incorporation of *Dicentrarchus labrax*. Journal of Experimental Marine Biology and Ecology, 446: 245–252.

Reis-Santos, P., Tanner, S. E., França, S., Vasconcelos, R. P., Gillanders, B. M., and Cabral, H. N. 2015. Connectivity within estuaries: An otolith chemistry and muscle stable isotope approach. Ocean & Coastal Management, 118: 51–59.

Reis-Santos, P., Tanner, S. E., Aboim, M. A., Vasconcelos, R. P., Laroche, J., Charrier, G., Pérez, M., *et al.* 2018. Reconciling differences in natural tags to infer demographic and genetic connectivity in marine fish populations. Scientific Reports, 8: 10343.

Rijnsdorp, A. D., and Van Beek, F. A. 1991. Changes in growth of plaice *Pleuronectes platessa* L. and sole *Solea solea* (L.) in the North Sea. Netherlands Journal of Sea Research, 27: 441–457.

Rijnsdorp, A. D., Van Beek, F. A., Flatman, S., Millner, R. M., Riley, J. D., Giret, M., and De Clerck, R. 1992. Recruitment of sole stocks, *Solea solea* (L.), in the Northeast Atlantic. Netherlands Journal of Sea Research, 29: 173–192.

Rijnsdorp, A. D., Damme, V., G, C. J., and Witthames, P. R. 2010. Implications of fisheries-induced changes in stock structure and reproductive potential for stock recovery of a sex-dimorphic species, North Sea plaice. ICES Journal of Marine Science, 67: 1931–1938.

Riou, P., Le Pape, O., and Rogers, S. I. 2001. Relative contributions of different sole and plaice nurseries to the adult population in the Eastern Channel: application of a combined method using generalized linear models and a geographic information system. Aquatic living resources, 14: 125–135.

Ripley, B., Venables, B., Bates, D. M., Hornik, K., Gebhardt, A., Firth, D., and Ripley, M. B. 2013. Package ‘mass’. Cran R.

Rochette, S., Rivot, E., Morin, J., Mackinson, S., Riou, P., and Le Pape, O. 2010. Effect of nursery habitat degradation on flatfish population: Application to *Solea solea* in the Eastern Channel (Western Europe). Journal of Sea Research, 64: 34–44.

Rochette, S., Huret, M., Rivot, E., and Le Pape, O. 2012. Coupling hydrodynamic and individual-based models to simulate long-term larval supply to coastal nursery areas: Modelling larval supply to coastal nurseries. Fisheries Oceanography, 21: 229–242.

Rochette, S., Le Pape, O., Vigneau, J., and Rivot, E. 2013. A hierarchical Bayesian model for embedding larval drift and habitat models in integrated life cycles for exploited fish. Ecological Applications, 23: 1659–1676.

Rogers, L. A., Storvik, G. O., Knutsen, H., Olsen, E. M., and Stenseth, N. C. 2017. Fine-scale population dynamics in a marine fish species inferred from dynamic state-space models. Journal of Animal Ecology, 86: 888–898.

Rushing, C. S., Ryder, T. B., Scarpignato, A. L., Saracco, J. F., and Marra, P. P. 2016. Using demographic attributes from long-term monitoring data to delineate natural population structure. Journal of Applied Ecology, 53: 491–500.

Russ, J. C. 1990. Computer-assisted microscopy: The measurement and analysis of images. Springer Science & Business Media, New York. 461 pp.

Ruzzante, D. E., Taggart, C. T., and Cook, D. 1998. A nuclear DNA basis for shelf- and bank-scale population structure in northwest Atlantic cod (*Gadus morhua*): Labrador to Georges Bank. Molecular Ecology, 7: 1663–1680.

Saenz-Agudelo, P., Jones, G. P., Thorrold, S. R., and Planes, S. 2009. Estimating connectivity in marine populations: an empirical evaluation of assignment tests and parentage analysis under different gene flow scenarios. Molecular Ecology, 18: 1765–1776.

Saila, S. B., and Martin, B. K. 1987. A brief review and guide to some multivariate methods for stock identification. Tech. Mem., Proceedings of the Stock Identification Workshop, NMFS-SEFC. NOAA.

Sainsbury, K. J., Punt, A. E., and Smith, A. D. M. 2000. Design of operational management strategies for achieving fishery ecosystem objectives. ICES Journal of Marine Science, 57: 731–741.

Savina, M., Lacroix, G., and Ruddick, K. 2010. Modelling the transport of common sole larvae in the southern North Sea: Influence of hydrodynamics and larval vertical movements. Journal of Marine Systems, 81: 86–98.

Savina, M., Lunghi, M., Archambault, B., Baulier, L., Huret, M., and Le Pape, O. 2016. Sole larval supply to coastal nurseries: Interannual variability and connectivity at interregional and interpopulation scales. Journal of Sea Research, 111: 1–10.

Schnute, J. 1981. A versatile growth model with statistically stable parameters. Canadian Journal of Fisheries and Aquatic Sciences, 38: 1128–1140.

Secor, D. H. 1999. Specifying divergent migrations in the concept of stock: the contingent hypothesis. Fisheries Research, 43: 13–34.

Secor, D. H. 2015. Migration ecology of marine fishes. JHU Press. 305 pp.

Selkoe, K. A., Henzler, C. M., and Gaines, S. D. 2008. Seascape genetics and the spatial ecology of marine populations. Fish and Fisheries, 9: 363–377.

Sinclair, A. F., Swain, D. P., and Hanson, J. M. 2002. Disentangling the effects of size-selective mortality, density, and temperature on length-at-age. Canadian Journal of Fisheries and Aquatic Sciences, 59: 372–382.

Sippel, T., Paige Eveson, J., Galuardi, B., Lam, C., Hoyle, S., Maunder, M., Kleiber, P., *et al.* 2015. Using movement data from electronic tags in fisheries stock assessment: A review of models, technology and experimental design. Fisheries Research, 163: 152–160.

Sley, A., Jawad, L. A., Hajjej, G., Jarboui, O., and Bouain, A. 2016. Morphometric and meristic characters of blue runner *Caranx crysos* and false scad *Caranx rhonchus* (Pisces: Carangidae) from the Gulf of Gabes, Tunisia, Eastern Mediterranean. Cahier de Biologie Marine, 57: 309–316.

Smedbol, R. K., and Wroblewski, J. S. 2002. Metapopulation theory and northern cod population structure: interdependency of subpopulations in recovery of a groundfish population. Fisheries Research, 55: 161–174.

Smedbol, R. K., McPherson, A., Hansen, M. M., and Kenchington, E. 2002. Myths and moderation in marine 'metapopulations'? Fish and Fisheries, 3: 20–35.

Smith, M. A., and Green, M. D. 2005. Dispersal and the metapopulation paradigm in amphibian ecology and conservation: are all amphibian populations metapopulations? Ecography, 28: 110–128.

Smith, P. J., Jamieson, A., and Birley, A. J. 1990. Electrophoretic studies and the stock concept in marine teleosts. ICES Journal of Marine Science, 47: 231–245.

Smith, S. J., and Campana, S. E. 2010. Smith and Campana 2010. Integrated mixture analysis for continuous and categorical data, with application to genetic-otolith combinations. Canadian Journal of Fisheries and Aquatic Sciences, 67: 1533–1548.

Sogard, S. M. 1991. Interpretation of otolith microstructure in juvenile winter flounder (*Pseudopleuronectes americanus*): Ontogenetic development, daily increment validation, and somatic growth relationships. Canadian Journal of Fisheries and Aquatic Sciences, 48: 1862–1871.

Southwell, D. M., Hauser, C. E., and McCarthy, M. A. 2016. Learning about colonization when managing metapopulations under an adaptive management framework. Ecological Applications, 26: 279–294.

Stephenson, R. L., Rodman, K., Aldous, D. G., and Lane, D. E. 1999. An in-season approach to management under uncertainty: the case of the SW Nova Scotia herring fishery. ICES Journal of Marine Science, 56: 1005–1013.

Stephenson, R. L. 2002. Stock structure and management structure: an ongoing challenge for ICES. *In* ICES Marine Science Symposia, pp. 305–314.

Stock, B. C., Jackson, A. L., Ward, E. J., Parnell, A. C., Phillips, D. L., and Semmens, B. X. 2018. Analyzing mixing systems using a new generation of Bayesian tracer mixing models. PeerJ, 6: e5096.

Stuckas, H., Knöbel, L., Schade, H., Breusing, C., Hinrichsen, H.-H., Bartel, M., Langguth, K., *et al.* 2017. Combining hydrodynamic modelling with genetics: can passive larval drift shape the genetic structure of Baltic Mytilus populations? Molecular Ecology, 26: 2765–2782.

Sturrock, A. M., Trueman, C. N., Milton, J. A., Waring, C. P., Cooper, M. J., and Hunter, E. 2014. Physiological influences can outweigh environmental signals in otolith microchemistry research. Marine Ecology Progress Series, 500: 245–264.

Swain, D. P., Sinclair, A. F., and Hanson, J. M. 2007. Evolutionary response to size-selective mortality in an exploited fish population. Proceedings of the Royal Society of London B: Biological Sciences, 274: 1015–1022.

Tanner, S. E., Reis-Santos, P., Vasconcelos, R. P., Thorrold, S. R., and Cabral, H. N. 2013. Population connectivity of *Solea solea* and *Solea senegalensis* over time. Journal of Sea Research, 76: 82–88.

Tanner, S. E., Pérez, M., Presa, P., Thorrold, S. R., and Cabral, H. N. 2014. Integrating microsatellite DNA markers and otolith geochemistry to assess population structure of European hake (*Merluccius merluccius*). Estuarine, Coastal and Shelf Science, 142: 68–75.

Tanner, S. E., Reis-Santos, P., and Cabral, H. N. 2016. Otolith chemistry in stock delineation: A brief overview, current challenges and future prospects. Fisheries Research, 173: 206–213.

Tawfik, D. S. 2010. Messy biology and the origins of evolutionary innovations. Nature Chemical Biology, 6: 692–696.

Taylor, N. G., McAllister, M. K., Lawson, G. L., Carruthers, T., and Block, B. A. 2011. Atlantic Bluefin Tuna: A novel multistock spatial model for assessing population biomass. PLOS ONE, 6: e27693.

Thorrold, S. R., Latkoczy, C., Swart, P. K., and Jones, C. M. J. 2001. Natal homing in a marine fish metapopulation. Science, 291(5502), 297-299.

Todorov, V., and Filzmoser, P. 2009. An object-oriented framework for robust multivariate analysis. pp 1-47.

Toole, C. L. 1993. Relationships between otolith microstructure, microchemistry, and early life history events in Dover sole, *Microstomus pacificus*. Fish. Bull. US, 91: 732–753.

Tuck, G. N., and Possingham, H. P. 1993. Optimal harvesting strategies for a metapopulation. Bulletin of Mathematical Biology, 56: 107–127.

Tuset, V. M., Lozano, I. J., González, J. A., Pertusa, J. F., and García-Díaz, M. M. 2003. Shape indices to identify regional differences in otolith morphology of comber, *Serranus cabrilla* (L., 1758). Journal of Applied Ichthyology, 19: 88–93.

Ulrich, C., Hemmer-Hansen, J., Boje, J., Christensen, A., Hüssy, K., Sun, H., and Clausen, L. W. 2017. Variability and connectivity of plaice populations from the Eastern North Sea to the Baltic Sea, part II. Biological evidence of population mixing. Journal of Sea Research, 120: 13–23.

Vasconcelos, J., Vieira, A. R., Sequeira, V., González, J. A., Kaufmann, M., and Gordo, L. S. 2018. Identifying populations of the blue jack mackerel (*Trachurus picturatus*) in the Northeast Atlantic by using geometric morphometrics and otolith shape analysis. Fishery Bulletin, 116: 81–92.

Vignal, A., Milan, D., SanCristobal, M., and Eggen, A. 2002. A review on SNP and other types of molecular markers and their use in animal genetics. Genetics Selection Evolution, 34: 275.

Vignon, M., and Morat, F. 2010. Environmental and genetic determinant of otolith shape revealed by a non-indigenous tropical fish. Marine Ecology Progress Series, 411: 231–241.

Vignon, M. 2015. Disentangling and quantifying sources of otolith shape variation across multiple scales using a new hierarchical partitioning approach. Marine Ecology Progress Series, 534: 163–177.

von Bertalanffy, L. 1957. Quantitative laws in metabolism and growth. The Quarterly Review of Biology, 32: 217–231.

Waldman, J. R., Richards, R. A., Schill, W. B., Wirgin, I., and Fabrizio, M. C. 1997. An empirical comparison of stock identification techniques applied to striped bass. Transactions of the American Fisheries Society, 126: 369–385.

Waldman, J. R. 1999. The importance of comparative studies in stock analysis. Fisheries Research, 43: 237–246.

Waldman, J. R. 2005. Chapter 2 - Definition of stocks: An evolving concept. *In* Stock identification methods, pp. 7–16. Academic Press.

Walter, J. A., Sheppard, L. W., Anderson, T. L., Kastens, J. H., Bjørnstad, O. N., Liebhold, A. M., and Reuman, D. C. 2017. The geography of spatial synchrony. Ecology Letters, 20: 801–814.

Walther, B. D., and Thorrold, S. R. 2006. Water, not food, contributes the majority of strontium and barium deposited in the otoliths of a marine fish.

Walther, B. D., and Limburg, K. E. 2012. The use of otolith chemistry to characterize diadromous migrations. Journal of Fish Biology, 81: 796–825.

Waples, R. S., and Gaggiotti, O. 2006. What is a population? An empirical evaluation of some genetic methods for identifying the number of gene pools and their degree of connectivity. Molecular Ecology, 15: 1419–1439.

Ward, R. D. 2000. Genetics in fisheries management. Hydrobiologia, 420: 191–201.

Webb, S. D., Woodcock, S. H., and Gillanders, B. M. 2012. Sources of otolith barium and strontium in estuarine fish and the influence of salinity and temperature. Marine Ecology Progress Series, 453: 189–199.

Weir, B. S., and Cockerham, C. C. 1984. Estimating F-statistics for the analysis of population structure. Evolution, 38: 1358–1370.

Welch, D. J., Newman, S. J., Buckworth, R. C., Ovenden, J. R., Broderick, D., Lester, R. J. G., Gribble, N. A., *et al.* 2015. Integrating different approaches in the definition of biological stocks: A northern Australian multi-jurisdictional fisheries example using grey mackerel, *Scomberomorus semifasciatus*. Marine Policy, 55: 73–80.

Wheeler, A., and Stebbing, P. 1978. Key to the fishes of Northern Europe; a guide to the identification of more than 350 species. London: Warne.

Wooster, W. S. 1992. King crab dethroned. Climate, Variability, Climate Change and Fisheries: 14–30.

Wright, P. J., Christensen, A., Régnier, T., Rindorf, A., and van Deurs, M. 2019. Integrating the scale of population processes into fisheries management, as illustrated in the sandeel, *Ammodytes marinus*. ICES Journal of Marine Science.

Ying, Y., Chen, Y., Lin, L., and Gao, T. 2011. Risks of ignoring fish population spatial structure in fisheries management. Canadian Journal of Fisheries and Aquatic Sciences, 68: 2101–2120.

Zemeckis, D. R., Martins, D., Kerr, L. A., and Cadrin, S. X. 2014. Stock identification of Atlantic cod (*Gadus morhua*) in US waters: an interdisciplinary approach. ICES Journal of Marine Science, 71: 1490–1506.

Zhang, C., Ye, Z., Li, Z., Wan, R., Ren, Y., and Dou, S. 2016. Population structure of Japanese Spanish mackerel *Scomberomorus niphonius* in the Bohai Sea, the Yellow Sea and the East China Sea: evidence from random forests based on otolith features. Fisheries Science, 82: 251–256.

Supplementary material

Supplementary material of chapter 2

Table S.2.1 Outputs vectors u (i.e., temporal trends estimated from the synchrony analysis) of the1989-2008 times series of estimate length-at-age 10 for females (F) and males (M) in the three spatial subunits (UK, UK coasts; NE, North-Eastern part of the French coast; SW South-Western part of the French coast).

	UK	NE	SW
F	-0.87	-2.95	-3.31
M	-0.73	-3.44	-2.28

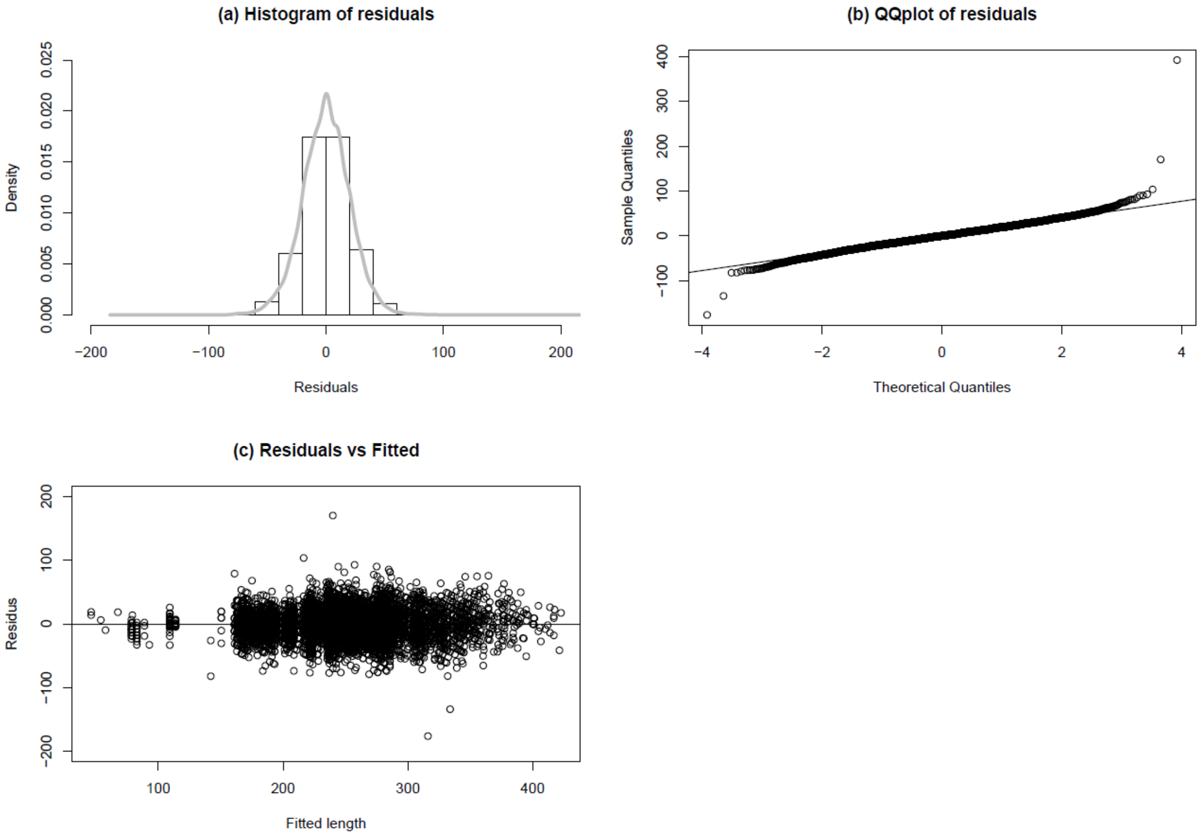


Fig. S.2.1 Analysis of the residuals of the Von Bertalanffy growth model. Panels (a) and (b) show the normality of residuals. Panel (c) presents the variance homoscedasticity.

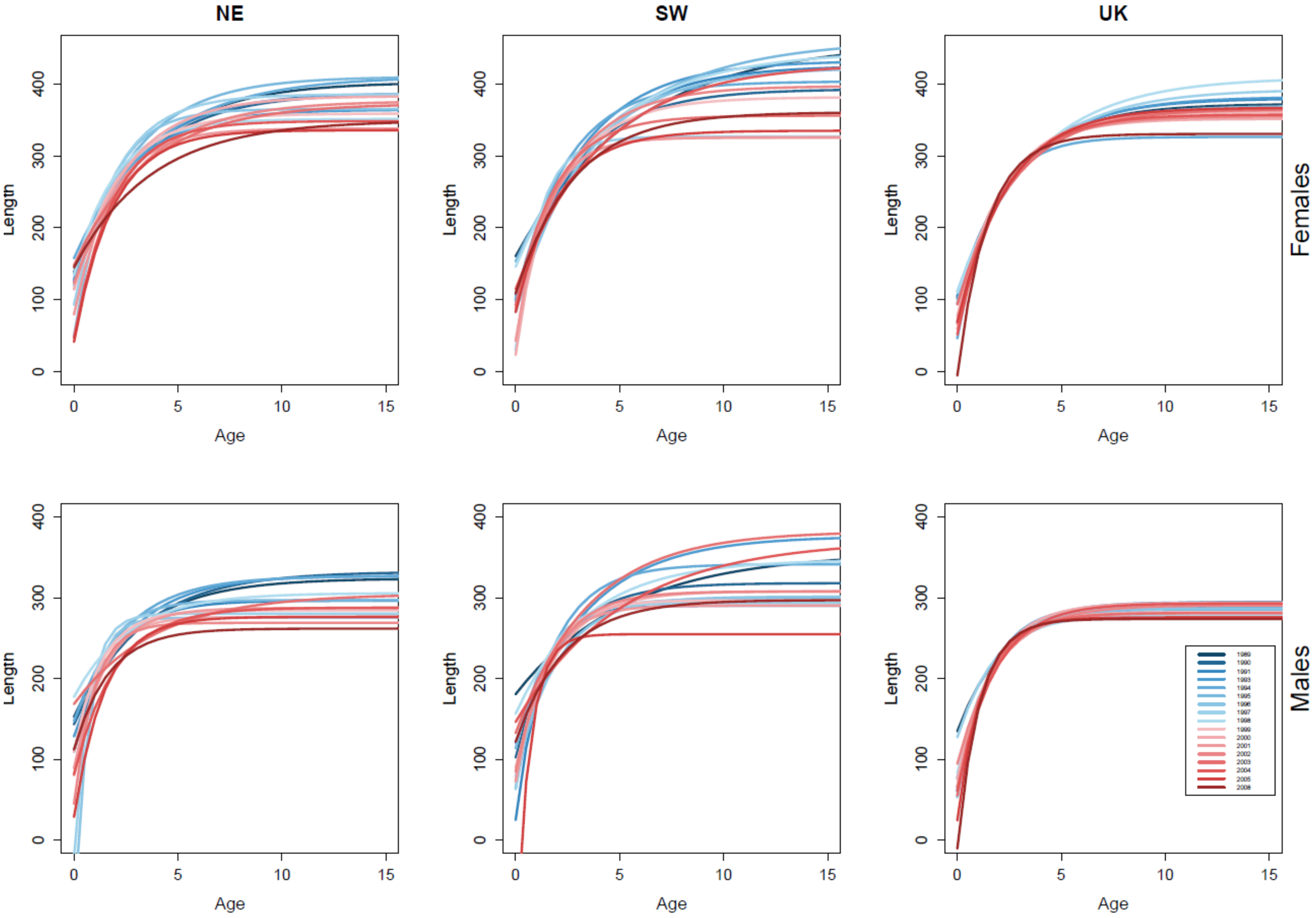
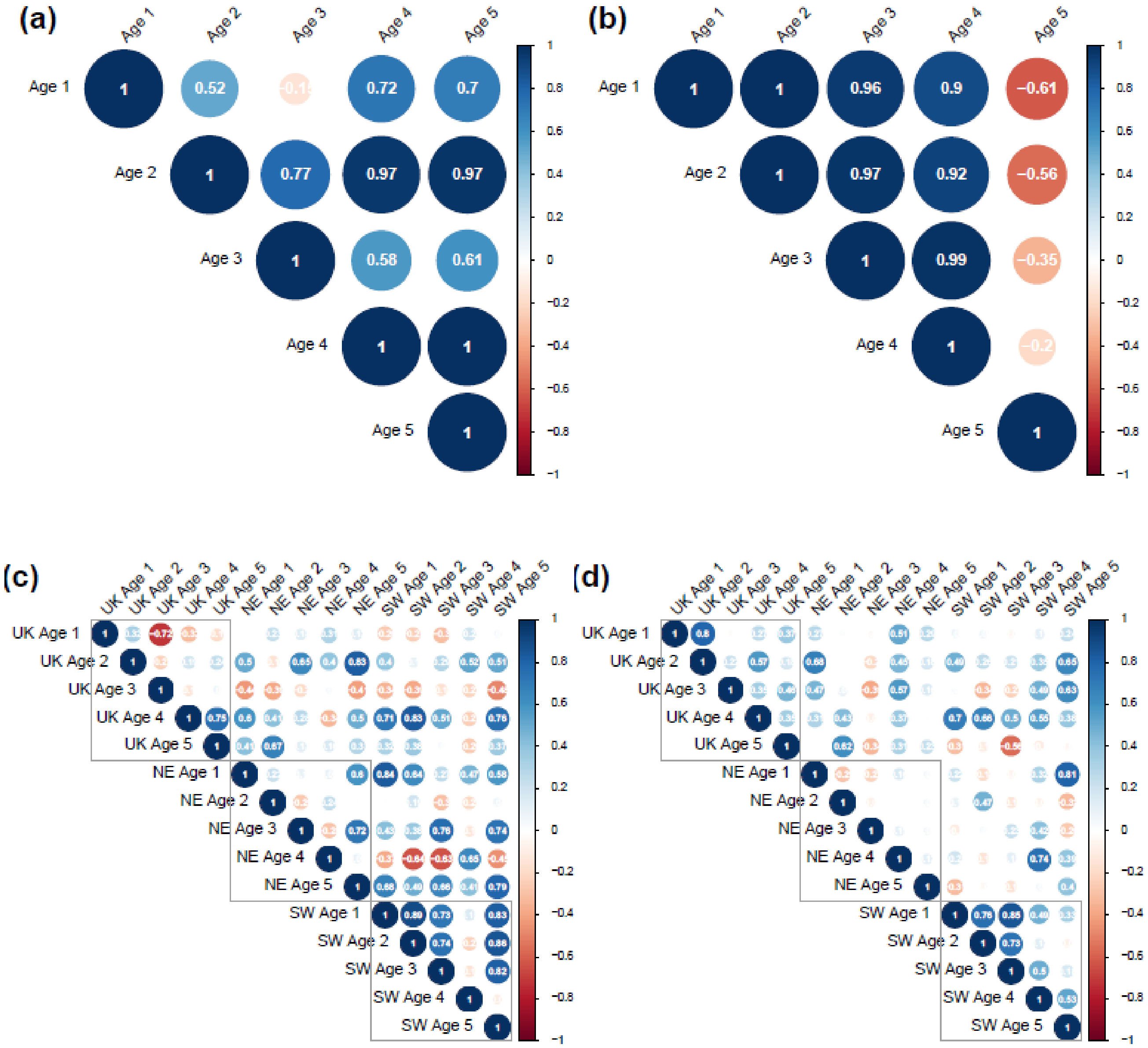
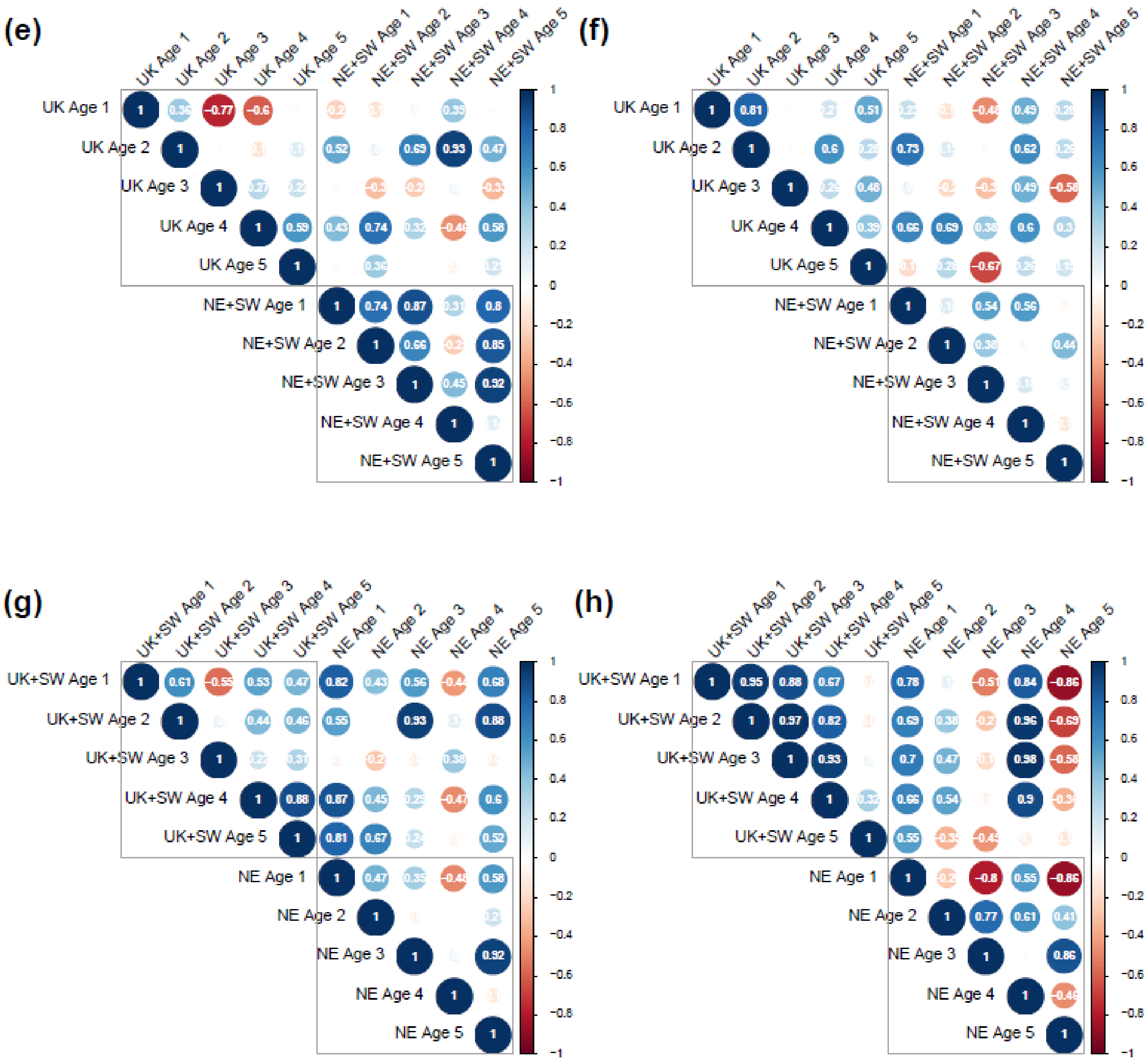


Fig. S.2.2 Curves of the von Bertalanffy growth model per sex, subunits (UK, UK coasts; NE, North-Eastern part of the French coast; SW South-Western part of the French coast) and cohorts.


(a)
Age 1
Age 2
Age 3
Age 4
Age 5
1
0.52
−0.1
0.72
0.7
0.77
0.97
0.97
0.58
0.61
(b)
0.96
0.9
−0.61
0.92
−0.56
−0.35
0.99
−0.2
1
0.8
0.6
0.4
0.2
0
−0.2
−0.4
−0.6
−0.8
−1
(c)
(d)
UK Age 1
UK Age 2
UK Age 3
UK Age 4
UK Age 5
NE Age 1
NE Age 2
NE Age 3
NE Age 4
NE Age 5
SW Age 1
SW Age 2
SW Age 3
SW Age 4
SW Age 5


(e)
UK Age 1
UK Age 2
UK Age 3
UK Age 4
UK Age 5
NE+SW Age 1
NE+SW Age 2
NE+SW Age 3
NE+SW Age 4
NE+SW Age 5
(f)
UK Age 1
UK Age 2
UK Age 3
UK Age 4
UK Age 5
NE+SW Age 1
NE+SW Age 2
NE+SW Age 3
NE+SW Age 4
NE+SW Age 5
(g)
UK+SW Age 1
UK+SW Age 2
UK+SW Age 3
UK+SW Age 4
UK+SW Age 5
NE Age 1
NE Age 2
NE Age 3
NE Age 4
NE Age 5
(h)
UK+SW Age 1
UK+SW Age 2
UK+SW Age 3
UK+SW Age 4
UK+SW Age 5
NE Age 1
NE Age 2
NE Age 3
NE Age 4
NE Age 5
1
0.8
0.6
0.4
0.2
0
−0.2
−0.4
−0.6
−0.8
−1

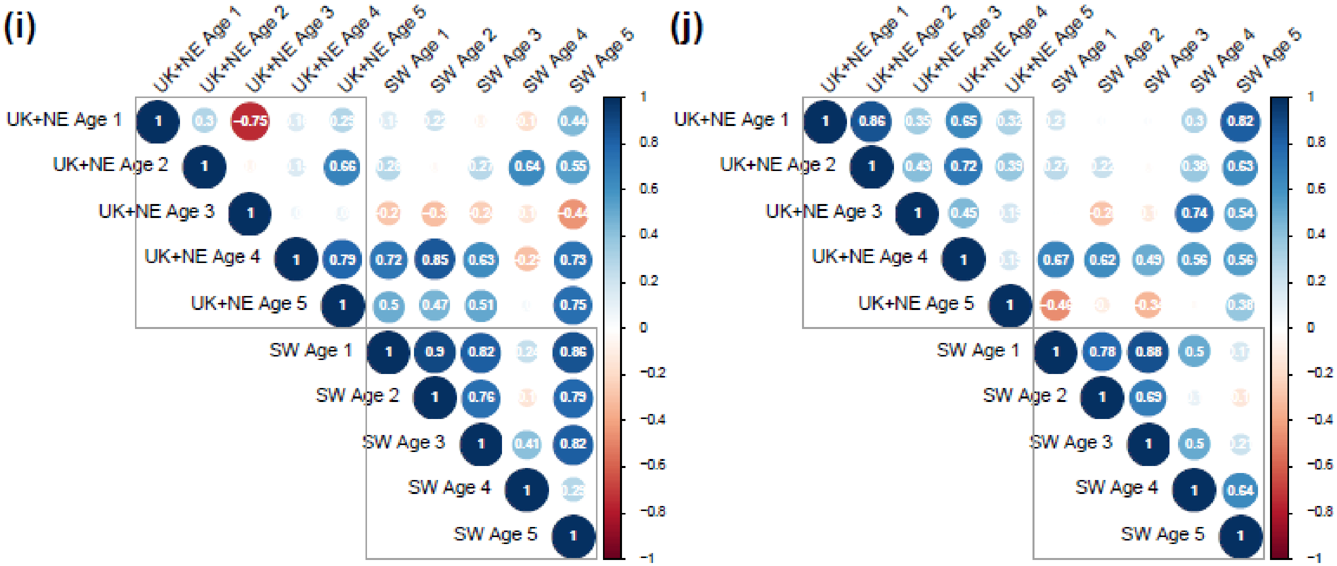


Fig. S.2.3 Correlation matrices of density-at-age anomalies time series from ages 1 to 5 between subunits. Left and right panels correspond to females and males correlations, respectively. The colors and sizes of the circles indicate the direction (positive or negative) and intensity of the correlation. Grey rectangles highlight within subunit correlations. (a) and (b): H0 (i.e. single stock). (c) and (d): H1 (i.e. UK, NE and SW subunits). (e) and (f): H2 (i.e. UK and NE-SW subunits). (g) and (h): H3 (i.e. NE and UK-SW subunits). (i) and (j): H4 (i.e. SW and UK-NE subunits).

Supplementary material of chapter 3

Appendix A – Genetic analysis

A.1. Material and methods

- **Molecular techniques and sequencing**

Genomic DNA was extracted following Cruz *et al.* (2017). SNP markers were identified using double digest restriction-site associated DNA (ddRAD) (Peterson *et al.*, 2012). Two libraries were built (adult samples of 2017 and 2018) based on the protocol of Palaiokostas *et al.* (2015) with the restriction enzymes *SbfI* and *SphI*. After enzymatic digestion and adapter ligation, sequences were size-selected (320-590 bp) and PCR amplified (16 cycles). Fragments between 300 and 600 bp were selected and libraries were sequenced paired-end on an Illumina HiSeq 2500 platform (Genomics Core of the KU Leuven, Belgium).

- ***De novo* assembly, SNP calling and filtering**

De novo assembly was chosen since no reference material was available for *Solea solea*. SNP calling was performed with the *dDocent* pipeline (Puritz *et al.*, 2014). First, quality of reads was checked with FastQC (Andrews, 2010) and forward and pair-end files were then demultiplexed with *process-radtags* of STACKS (Catchen *et al.*, 2013). Quality trimming was performed using *Trimmomatic* (Bolger *et al.*, 2014) and sequences were assembled with the *Rainbow* RadSeq assembly program (Chong *et al.*, 2012). Reference sequences were clustered using the *CD-HIT* program (Fu *et al.*, 2012). Then, the quality-trimmed reads were mapped to the reference contigs with the *BWA-MEM* alignment algorithm (Li, 2013). SNP calling was performed by the assembled haplotypes sequences with *FreeBaye*s variant detection software (Garrison and Marth, 2012). Finally, SNP were concatenated into a single variant call file (VCF) using *VCFtools* program (Danecek *et al.*, 2011).

Several filters were applied to reduce the number of SNPs and select relevant markers: minimum allelic depth = 4, allelic balance range = 0.25-0.75, minimum allele frequency = 0.05, exclusion of loci with missing data > 10% over all individuals. Loci with observed heterozygosity above 0.5 were removed. The Hardy-Weinberg equilibrium was estimated and distribution of missing data across spatial subunits and individuals and linkage disequilibrium were assessed using the *poppr* package

(Kamvar *et al.*, 2014). Loci out of the Hardy-Weinberg equilibrium for more than 2/3 of individuals were removed. Also, loci in Linkage Disequilibrium above 0.7 were eliminated.

- **Spatial structure of genetic variation**

Genetic differentiation between individuals was assessed using different approaches, following the protocol of Mullins *et al.* (2018). First, global and pairwise F_{ST} values (Weir and Cockerham, 1984) were calculated with the *hierfstat* R package. Significance of pairwise F_{ST} tests was computed by bootstrap (1000 permutations) and resulted in 95% interval credibility. Finally, assessment of genetic structure was completed with Discriminant Analysis of Principal Component (DAPC) of *adegenet* R package (Jombart *et al.*, 2010). The number of principal components was fixed using the a-score optimization method provided by *adegenet*.

A.2. Supplementary results

Whatever the alternative hypotheses of spatial structure H2, H3 and H4, pairwise and global FST were found to be significant (**Table A.1**).

Table A.1. Pairwise F_{ST} values and the corresponding 95% confidence interval (upper and lower limits) for hypotheses of spatial structure H2, H3 and H4. '*' indicate significant values.

Hypothesis	Year	Spatial comparison	Lower limit of 95% CI	F_{ST} value	Upper limit of 95% CI
H2	2017	SW-NE/UK	0.0028	0.0042*	0.0056
	2018		0.0001	0.0024*	0.0049
H3	2017	UK-SW/NE	0.0038	0.0052*	0.0065
	2018		-0.0005	0.0017*	0.004
H4	2017	UK-NE/SW	0.0015	0.0025*	0.0036
	2018		-0.036	0.0018*	0.0083

Using all genetic samples from 2017 (Ng_{2017} = 120), the *assignPOP* R package was applied to estimate reallocation probabilities and investigate spatial population structure. Using 90% of individuals in the training data set and 50-100% of genetic loci allowed maximazing assignment accuracy. However, using only 10-25% of the highest F_{ST} loci did not allow assigning correctly (above 0.33) in the NE subunit (**Fig. A.1**).

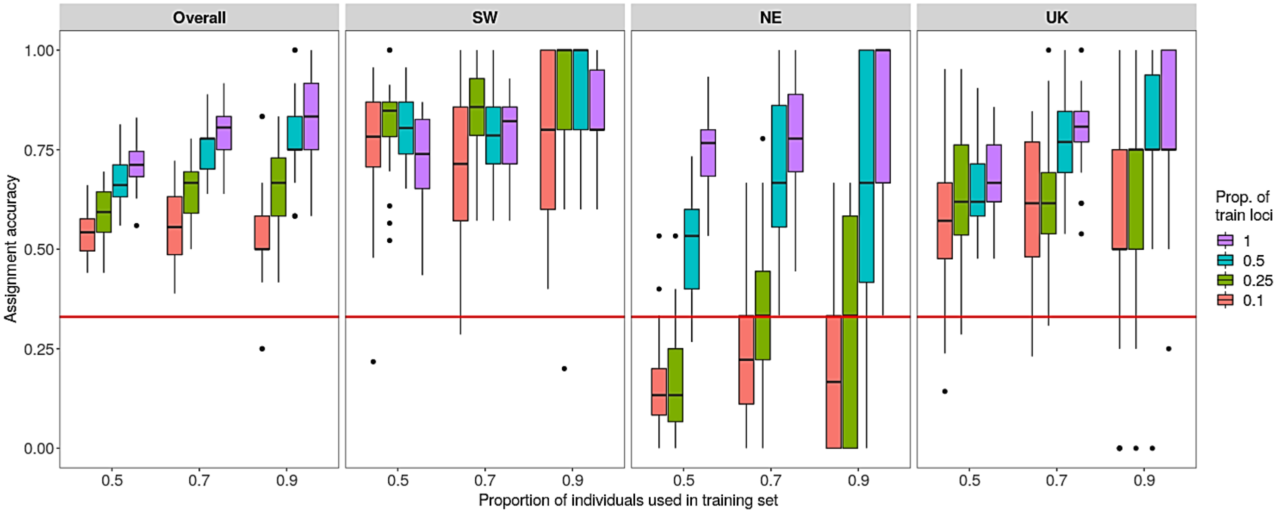


Figure. A.1. Estimated assignment accuracy performed by Monte Carlo cross-validation and support vector machine methods using all individuals of the genetic data set of 2017. Training data sets were composed of four proportions of training loci (top 10% in orange, 25% in green and 50% in blue of the highest F_{ST} loci and all loci in purple). Three levels of training data sets were considered (0.5, 0.7 and 0.9) and 30 iterations were computed. The horizontal red line represented the null subunit assignment rate (0.33 in the case of three subunits).

Also, **Fig. A2** showed evidence of spatial structure with individuals mainly reallocated in their subunit of sampling with high probabilities. This result was in line with the genetic structure of **part 3.1**.

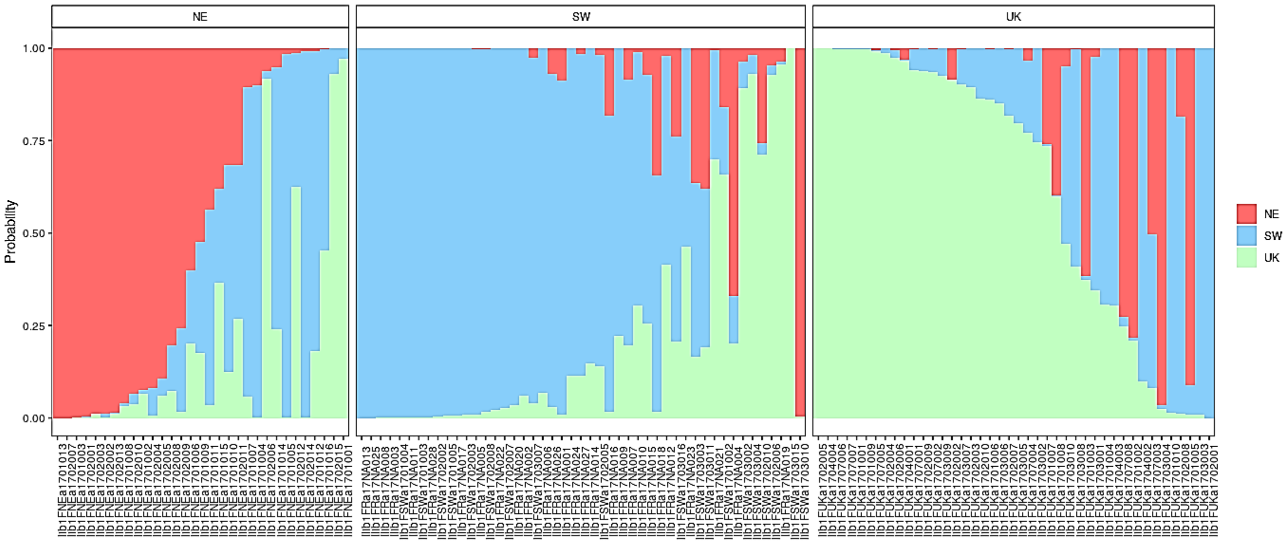


Figure. A.2. Probabilities of reallocation of individuals in the three subunits using only the SNP genotypes of 2017. Panels correspond to the subunits where individuals were sampled. Probabilities were estimated using the K-fold cross-validation method (K = 3) and all loci (2302 SNPs).

Appendix B – Otolith shape analysis

B.1. Material and methods

- **Shape indices**

Distribution of shape indices in subunits are presented in **Fig. B.1**.

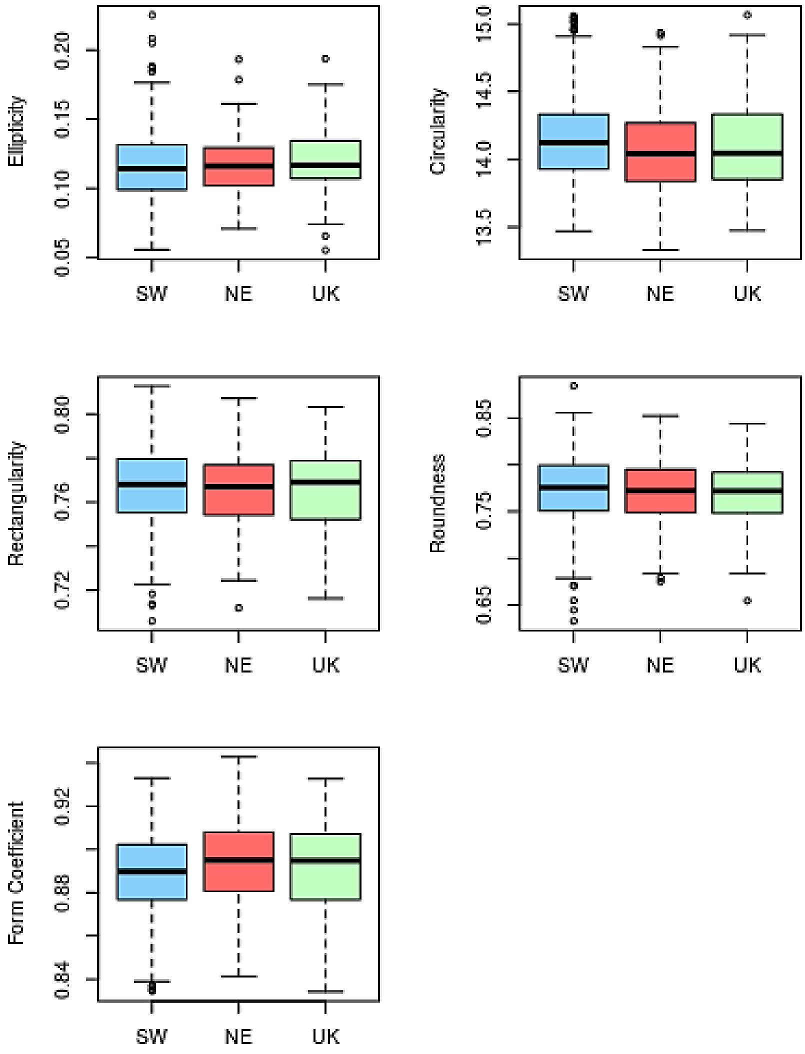


Figure. B.1. Shape indices of otoliths sampled in putative SW, NE and UK subunits of the EEC.

After removing the size effect from shape indices, spatial differences were tested regarding alternative hypotheses of spatial structure (**Table B.1**).

Table B.1. Mean comparisons of shape indices between subunits using one-way ANOVA for spatial structure hypotheses H2, H3 and H4. Stars indicate significance. Left and right otoliths were pooled. Statistical significance: '***' P < 0.001; '**' P < 0.01; '*' P < 0.05

Hypothesis	Shape index	DF	F	p
H2	Ellipticity	1	23.12	<0.001 ***
	Circularity	1	3.108	0.0783
	Rectangularity	1	0.258	0.611
	Roundness	1	23.09	<0.001 ***
	Form coefficient	1	3.431	0.0644
H3	Ellipticity	1	16.03	<0.001 ***
	Circularity	1	6.416	0.0115 *
	Rectangularity	1	0.287	0.592
	Roundness	1	16.17	<0.001 ***
	Form coefficient	1	7.888	0.005 **
H4	Ellipticity	1	48.25	<0.001 ***
	Circularity	1	11.59	<0.001 ***
	Rectangularity	1	0.665	0.415
	Roundness	1	48.42	<0.001 ***
	Form coefficient	1	13.71	<0.001 ***

Result of the Linear Discriminant Analysis performed in the shape indices is provided in **Table B.2**.

Table B.2. Results of discriminant analyses performed on the left (L), right (R) and both otoliths (L+R) for each hypothesis of spatial structure. Statistical significance: '***' P < 0.001; '**' P < 0.01; '*' P < 0.05

Hypothesis	Results	L	R	L+R
H1	Wilks' λ	0.910	0.890	0.909
	p	<0.001 ***	<0.001 ***	<0.001 ***
	Reclassification success (%)	47	48	48
H2	Wilks' λ	0.963	0.958	0.961
	p	0.007 **	0.01 *	<0.001 ***
	Reclassification success (%)	61	58	59
H3	Wilks' λ	0.960	0.951	0.963
	p	0.004 **	0.006 **	<0.001 ***
	Reclassification success (%)	58	58	57
H4	Wilks' λ	0.912	0.892	0.911
	p	<0.001 ***	<0.001 ***	<0.001 ***
	Reclassification success (%)	62	63	62

- **Otolith bilateral asymmetry**

The use of spatial variation in bilateral directional asymmetry (DA) of otolith shape has been suggested as a new tool to discriminate populations (Mahé *et al., in press*). Shape analyses were complemented by assessing variation in the degree of DA between subunits. DA was estimated as the percentage of non-overlapping surface between average reconstructed left and right otoliths. Since the common sole is a flatfish, DA was expected but spatial differences of such asymmetry could indicate different environmental conditions and low connectivity between subunits. This analysis was conducted on 330 pairs of otoliths. Average otolith shapes were reconstructed based on EFD.

- **Elliptical Fourier Descriptors**

For each otolith, the first 99 elliptical Fourier harmonics were extracted from the scanned image via TNPC 7 software. Normalization with respect to the first harmonic insured the invariance of harmonics from the otolith size, rotation and starting point of contour description. Each harmonic k is described by 4 coefficients a_k, b_k, c_k and d_k so that 392 shape descriptors (EFD) were available (98 harmonics and 4 coefficients per harmonic). To reduce the number of descriptors, the number of harmonics n_j of each otolith j was adjusted so that its contour was reconstructed with a precision of 99.9% (i.e., the proportion of variance in contour coordinates accounted for by the harmonics was 99.9%) as measured by the cumulative Fourier power F:

$$\mathbf{F}(\boldsymbol{n}_\mathrm{j}) = \sum_{\boldsymbol{k}=\mathbf{1}}^{\boldsymbol{n}_\mathrm{j}} \frac{\boldsymbol{a}_\mathrm{k}^{\mathbf{2}} + \boldsymbol{b}_\mathrm{k}^{\mathbf{2}} + \boldsymbol{c}_\mathrm{k}^{\mathbf{2}} + \boldsymbol{d}_\mathrm{k}^{\mathbf{2}}}{\mathbf{2}} = \mathbf{99.9}\%$$

The maximum number of harmonics $n = \max(n_j)$ across all otoliths was then used to describe their contour to ensure a precision of at least 99.9% for each of them.

Then, the number of EFD was further reduced using a principal component analysis (PCA) with the *prcomp* function of the *stats* package. The number of principal components was then chosen using a broken stick model (Legendre and Legendre, 1998). The matrix of chosen principal components S represents thus the otolith shape matrix.

Genotype and environment are known to influence otolith shape but additional variables linked to the developmental stage of the fish are also expected to affect otolith shape such as total length, age, sex and maturity stage (Cardinale *et al.*, 2004; Vignon and Morat, 2010; Mille *et al.*, 2016). Therefore, when testing the variation in the otolith shape matrix S between subunits of sampling, variables linked to the developmental stage, i.e. total length, sex, and age where added as covariables. Potential temporal variations were also accounted for by adding the effect of year of sampling and cohort (i.e. year-class). Finally, the asymmetry between left and right otolith was taken into account by adding a side effect. These various effects on the otolith shape matrix (S) were tested using a redundancy analysis (RDA) with the *rda* function of the *vegan* package. An automatized stepwise model selection was applied using the *ordiR2step* function of the same package. This procedure used the adjusted R squared (R^2) as a selection criterion. It was applied in a stepwise direction so that effects were added or removed sequentially to the starting model (M0) until the model with the highest R^2 was found. A "starting" model (M0) and a "maximum" model (Mmax) were included in the ordiR2step function (Blanchet *et al.*, 2008):

$$(\boldsymbol{S}) \sim \boldsymbol{1} \qquad (M_0)$$

$$(\boldsymbol{S}) \sim \boldsymbol{Length} + \boldsymbol{Age} + \boldsymbol{Year} + \boldsymbol{Cohort} + \boldsymbol{Side} + \boldsymbol{Sex} + \boldsymbol{Subunit} \qquad (M_{max})$$

The "starting" model included no effect on the otolith shape matrix. The "maximum" model considered all potential effects on otolith shape except the genotypic and environmental effects. The Subunit effect tested for potential spatial differences in otolith shape matrix and the four alternative hypotheses of spatial structure (H1, H2, H3 and H4; Randon *et al.*, 2018) were considered so that four models Mmax were computed (MmaxH1, MmaxH2, MmaxH3 and MmaxH4).

Finally, hierarchical clustering was performed on the shape matrix S to assess spatial consistency between emerging clusters of otolith morphology and the different subunits considered for each spatial structure hypothesis. This clustering analysis was computed on the residual otolith shape matrix (S') of the redundancy analysis. The function *hclust* of the *stats* package was applied on a dissimilarity matrix using the Ward's minimum variance method. The dissimilarity matrix was calculated from the otolith shape matrix using the *dist* function of the *stats* package with Euclidean distance method. Then, results were visualized using functions *fviz_dend* and *fviz_cluster* of the *factoextra* package. The number of clusters was decided visually using the *fviz_nbclust* function with the silhouette method. This method measures how well samples are clustered and estimated the average distance between clusters.

B.2. Supplementary results

- **Otolith bilateral asymmetry**

Results (**Table B.3**) indicated a significant side effect on the shape matrix S based on EFD. Following hypothesis H1 (i.e. 3 subunits), spatial differences in bilateral asymmetry was found, especially between the NE subunit and the SW and UK subunits (**Fig. B.2.a-c**). Besides, spatial variation in otolith shape was higher considering the left otoliths (i.e. bling side for common sole) (**Fig. B.2.d-e**).

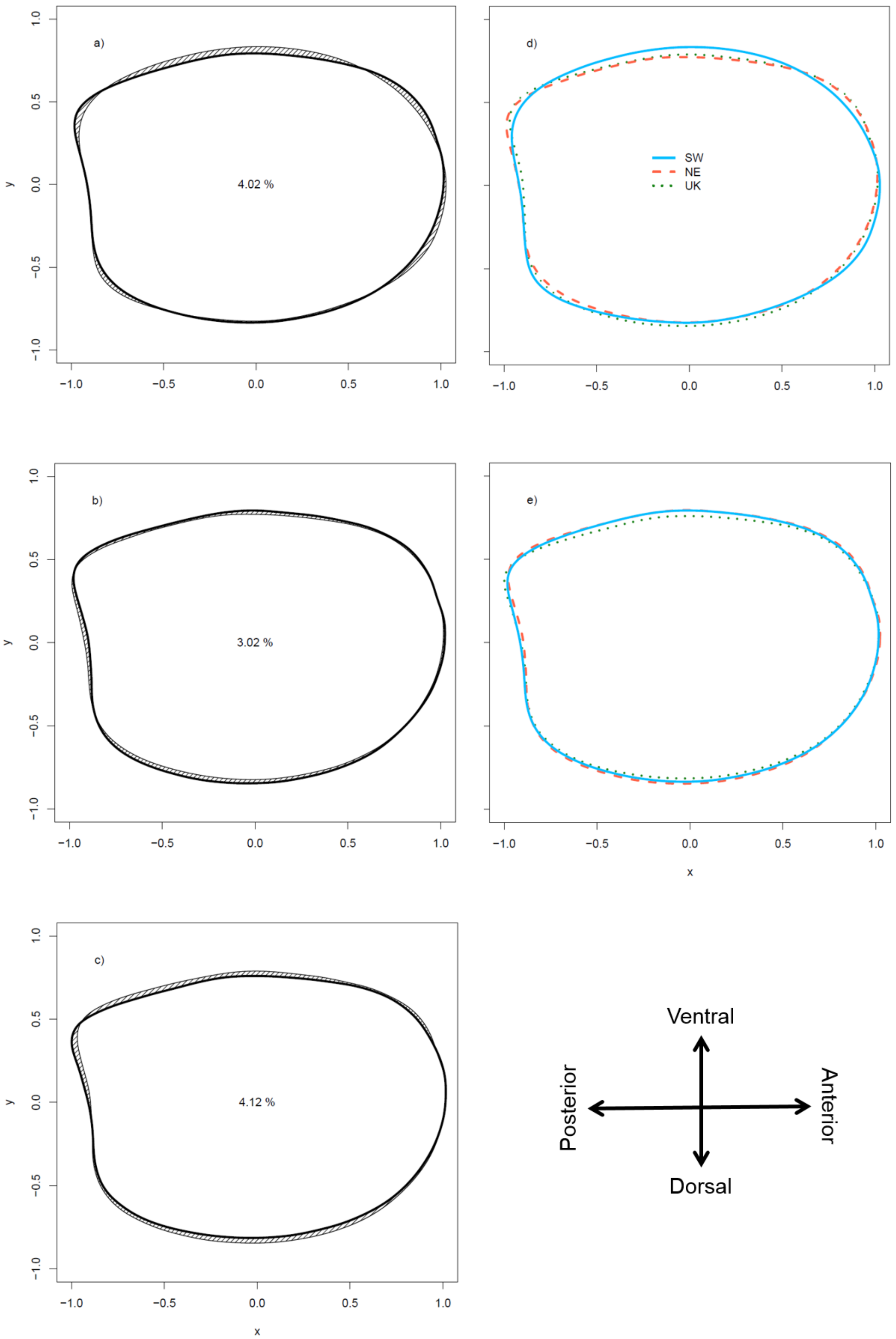


Figure. B.2. Left panel column: Comparison of average reconstructed otolith shape between left (full lines) and right otoliths (dotted lines) from the SW (a), NE (b) and UK (c) subunits. Crosshatching indicate non-overlapping zones between the left and right otoliths. Percentages of bilateral asymmetry are mentioned for each subunit. Right panel column: Spatial differences between average left (d) and right (e) otolith shapes.

Fig. B.2 a-c indicated that the NE subunit had a lower level of bilateral asymmetry than the others. This could result from different environmental conditions and low connectivity with the rest of the EEC stock thus suggesting that the NE subunit is partly isolated from the rest of the stock. However, another interpretation could be that the SW and UK subunits presented similar environmental conditions, or different environmental conditions with high levels of mixing between subunits (i.e. homogenization). Therefore the analysis of bilateral asymmetry suggested spatial structure with potential partial isolation of the NE subunit but did not allow concluding about the two other subunits.

- **Elliptical Fourier Descriptors**

On average, otoliths were reconstructed at 99.99% with 9 harmonics but the maximum number of harmonics $\max(n_j)$ = 28 was preferred to reconstruct all individual otoliths with the precision of 99.99%. At this stage, 112 EFD were available (28 harmonics and 4 coefficients per harmonic).

<u>Principal Component Analysis</u>

A principal component analysis was applied in the shape matrix composed of 112 EFD. Based on a broken stick model, the first seven principal components (PC) were kept and corresponded to 89.4% of total inertia. The shape matrix used in the following analysis was composed of these 7 PC in columns and individual otoliths in rows. The first 2 PC are presented in **Fig. B.3**.

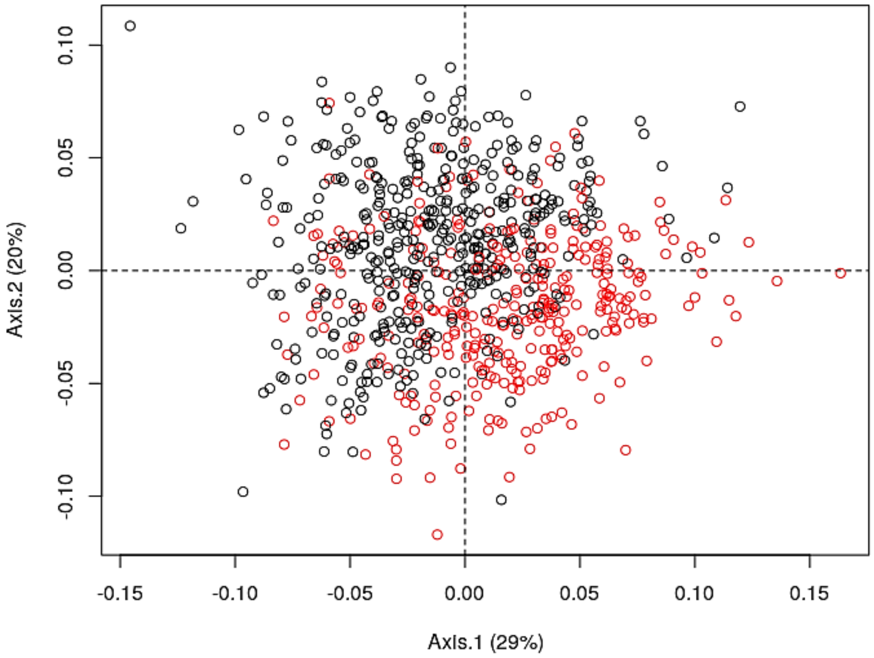


Figure. B.3. Plot of the first two principal components of the PCA applied on the shape matrix based on elliptical Fourier descriptors. Circles represent otoliths. Black and red circles correspond to the left and right otoliths, respectively.

Redundancy Analysis

Whatever the spatial structure considered, the stepwise procedure selected a model (M_3) that included the effects of $Side$, $Cohort$ and $Length$ on the otolith shape matrix (**Table B.3**).

Table B.3. Results of the stepwise model selection for redundancy analysis considering a staring model M0 with no effect on the otolith shape matrix (S). “+” means that effects are added to the previous model during the stepwise procedure. Adjusted R-squared (R^2) and p-value are indicated at each step of the stepwise selection procedure. Model M3 is the selected model with the highest R^2. Statistical significance: ‘***’ P < 0.001; ‘**’ P < 0.01; ‘*’ P < 0.05

Hypothesis	Model	Model formulation	R^2	p-value
H1	M0	$(S) \sim 1$	0	-
	M1	$(S) \sim 1 + Side$	0.161	0.002**
	M2	$(S) \sim 1 + Side + Cohort$	0.177	0.002**
	M3	$(S) \sim 1 + Side + Cohort + Length$	0.179	0.03*

This result suggests that the otolith shape based on EFD did not vary spatially and was invariant between females and males. Also, it suggested a potential otolith asymmetry and a cohort effect (i.e. year-class effect). Finally, the adjusted R-squared of the selected model (M3) was particularly low, suggesting that other major factors (environmental or genotypic) potentially impacted the otolith shape.

Hierarchical Clustering

Hierarchical clustering was performed to assess the potential consistency between otolith morphological groups and subunits. This analysis revealed three clusters (**Fig. B.4**).

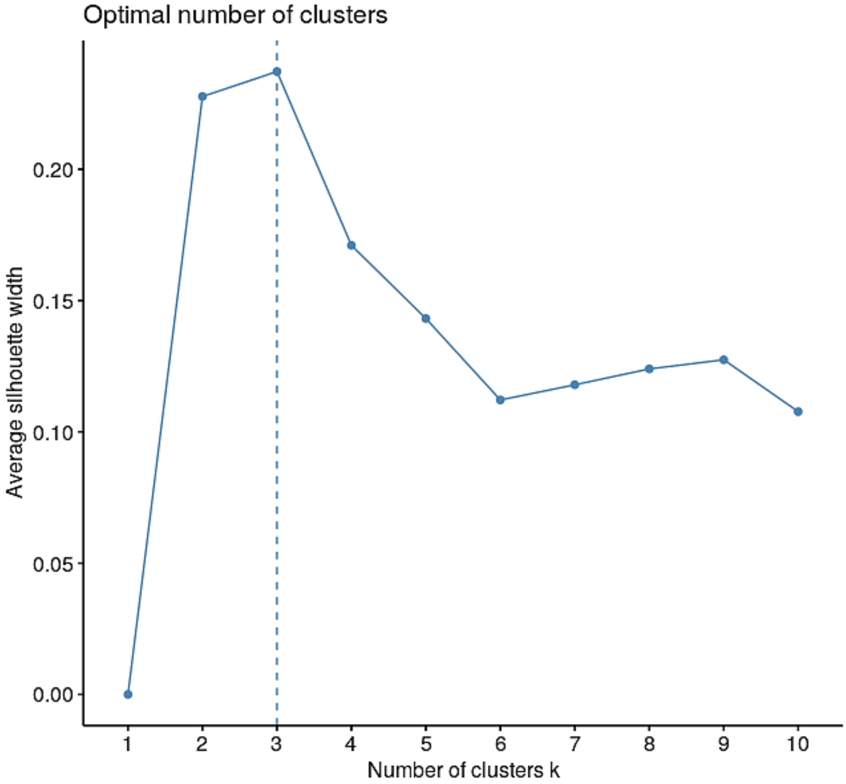


Figure. B.4. Optimal number of clusters using the Silhouette method as validation of clustering consistency based on Fourier descriptors. The vertical line indicates the number of clusters that maximizes the silhouette, i.e. the optimal number of clusters.

The resulting tree and corresponding scatterplot (**Fig. B.5.a** and **B.5.b**) revealed some overlap between the clusters, suggesting poor differences in otolith shape based on EFD.

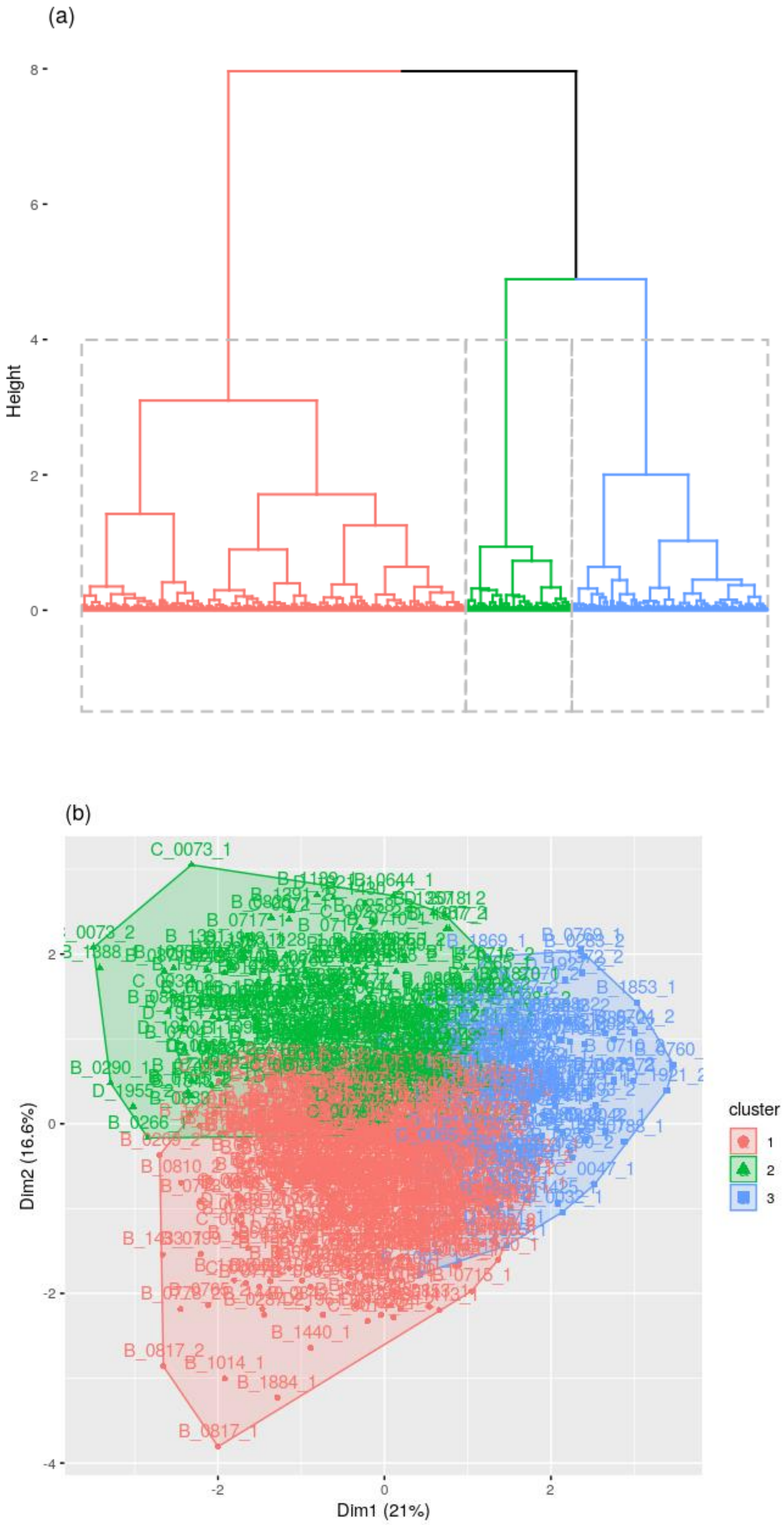


Figure. B.5. (a) Dendrogram and (b) principal components scatterplot of hierarchical clustering analysis on residual matrix (S') of EFDs.

In addition, no spatial consistency between clustering and subunits was found since otoliths were mainly classified in clusters 1 whatever their subunit of sampling (**Table B.4**). Thus, there was no signal of spatial structure using the EFDs as otolith shape descriptors.

Table B.4. Number of otoliths per cluster resulting from hierarchical clustering analysis of residual shape matrix (S'). Results are specified for each hypothesis of spatial structure and corresponding subunits. Grey cells indicate the highest number of otoliths for each subunit.

Hypotheses	Subunit(s)	Cluster 1	Cluster 2	Cluster 3
H1	SW	287	144	74
	NE	91	47	30
	UK	53	29	15
H2	NE+SW	378	191	104
	UK	53	29	15
H3	SW+UK	340	173	89
	NE	91	47	30
H4	UK+NE	144	76	45
	SW	287	144	74

Appendix C – Otolith microchemistry analysis

C.1. Material and methods

- **Otolith preparation and analysis**

Otolith preparation followed the protocol of Martin *et al.* (2015). Sagittal otoliths were removed from the fish, cleaned under a binocular with Milli-Q water, air-dried and stored in plastic vials preliminary washed with nitric acid (5%). Left and right otoliths were first scanned for shape analysis. Then, left otoliths were embedded in epoxy resin with the sulcus facing upward before grinding on a sagittal plan with different sanding pads (gran 1200 and 2400) and Milli-Q water, and polished with pads (9 µm and 3 µm) and diamond suspension. Polishing was stopped until the core of the otolith and its periphery were reached. Juvenile stage was targeted on both adult and juvenile otoliths to obtain microchemical signatures of nursery grounds (i.e. juvenile signatures).

Laser ablations were conducted using an Element XR (Thermo Scientific) LA-ICP-MS coupled to a 193 nm laser ablation system (Coherent Complex Pro) (Laboratory of Geosciences, IUEM, Brest, France). Laser conditions were set so that ablations were 40 µm diameter and 30 µm depth (10 Hz frequency and 800 pulses). Juvenile microchemical signatures of adult and juvenile individuals were obtained from three circular ablations localized in the postero-ventral region of the otolith (**Fig. C.1**). More precisely, ablations were positioned after the end of the metamorphosis zone of the otolith after the opaque zone, corresponding to the first summer spent in nursery ground (**Fig. C.1**).

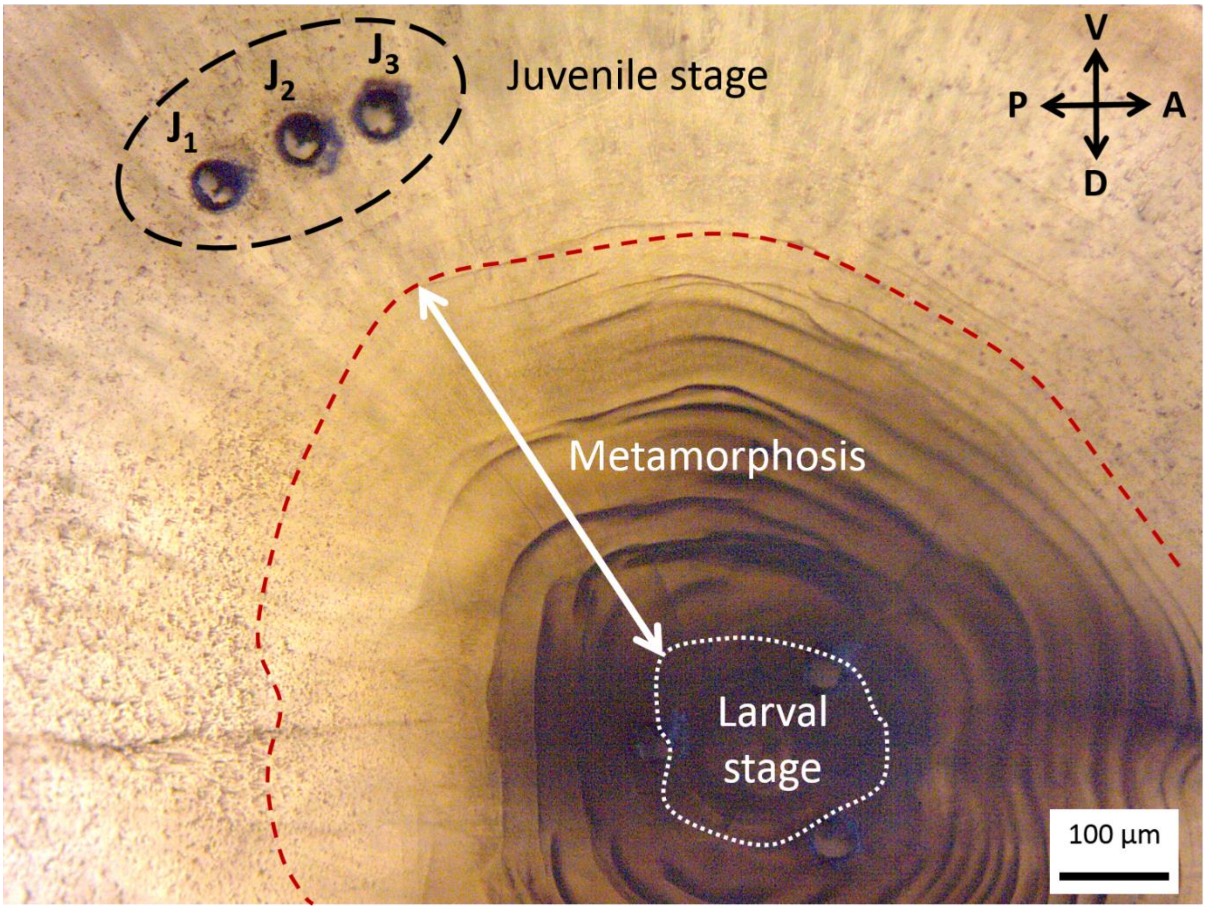


Figure. C.1. Microscopic view (transmitted light) of the central zone of a left sagittal otolith of an age-1 common sole (sagittal plan) after LA-ICP-MS. J_1, J_2 and J_3 indicate the three juvenile ablations, corresponding to the juvenile signature (black dotted lines). White and red dotted lines represent the beginning and the end of the metamorphosis, respectively.

For each ablation, ^{7}Li, ^{23}Na, ^{43}Ca, ^{55}Mn, ^{59}Co, ^{63}Cu, ^{66}Zn, ^{85}Rb, ^{88}Sr, ^{107}Ag, ^{111}Cd, ^{138}Ba, ^{208}Pb, ^{238}U were measured at low resolution. Ca was used as the internal standard for concentration calculations (µg.g^{-1}). Standard reference material NIST (National Institute of Standards and Technologies) 610, 612 and 614 were used to calibrate measurements every 3 otoliths.

After laser ablation, relative standard deviation of elements were: 1.77% for Li/Ca, 8.42% for Na/Ca, 1.93% for Mn/Ca, 8.24% for Co/Ca, 5.21% for Cu/Ca, 6.67% for Zn/Ca, 3.14% for Rb/Ca, 4.48% for Sr/Ca, 5.12% for Ag/Ca, 4.85% for Cd/Ca, 5.10% for Ba/Ca, 10.12% for Pb/Ca and 8.19% for U/Ca.

- **Limits of detection**

Limits of detection (LOD) were defined as 3 × Standard Deviation of the blank (**Table C.1**). Values below the LOD were set to the corresponding LOD.

Table C.1. Mean limits of detection (LOD) for each element (µg.g^{-1}) and percentages of data above the limits of detection (% > LOD).

	Li	Na	Mn	Co	Cu	Zn	Rb	Sr	Ag	Cd	Ba	Pb	U
LOD	0.250	21.3	0.245	3.56	0.675	0.920	0.117	3.12	0.224	0.431	0.0623	0.0613	0.00424
% > LOD	91	100	100	0	46	57	58	100	0	0	100	15	6

- **Bayesian model**

The Bayesian model was based on Martin *et al.* (2015) and Randon *et al.* (2017) and adapted to marine nursery-dependent flatfish species.

Adults and juveniles are denoted by a and j respectively. Brackets { } correspond to vectors and matrices are indicated by braces [ ]. The otolith elemental concentrations were centred and scaled for juveniles and adults to allow single scale of variations between elements.

The otolith composition of an adult $Oto(a)$ followed a multinormal distribution (MN) with $mu(n)$ the average composition and $\sum(n)$ the variance-covariance matrix of an otolith in nursery n (Equation C.1).

$$(\ \{\boldsymbol{Oto(a)}\}\mid \boldsymbol{N(a)=n}\ ) \sim \boldsymbol{MN}(\ \{\boldsymbol{mu(n)}\}, [\boldsymbol{\Sigma(n)}]\ ) \qquad \text{(Equation C.1)}$$

$N(a)$ corresponds to the nursery of origin of an adult a. Uninformative priors were chosen for $mu(n)$ and $\sum(n)$ (Equation C.2 and C.3).

$$\{\boldsymbol{mu}\} \sim \boldsymbol{N}(\ \boldsymbol{0, 0.01}\ ) \qquad \text{(Equation C.2)}$$

$$[\boldsymbol{\Sigma}] \sim \boldsymbol{Wishart}(\ [\boldsymbol{I}], \boldsymbol{df}\ ) \qquad \text{(Equation C.3)}$$

With [I] the identity matrix of dimensions 4 × 4 (4 elements) and df the degree of freedom (number of elements + 1).

The otolith composition of a juvenile $Oto(j)$ also followed a multinormal distribution (MN) (Equation C.4).

$$(\{\boldsymbol{Oto}(\boldsymbol{j})\}) \sim \boldsymbol{MN}(\{\boldsymbol{mu}(\boldsymbol{N}(\boldsymbol{j}))\}, [\boldsymbol{\Sigma}(\boldsymbol{N}(\boldsymbol{j}))]) \tag{Equation C.4}$$

With $N(j)$ the nursery of origin of a juvenile j (i.e. its catch location).

Then, reallocation of adults to their nursery of origin was described by a categorical distribution (Equation C.5).

$$\mathbf{N}(\mathbf{a}) \sim \boldsymbol{Categorical}(\{\boldsymbol{\Theta}_{\mathbf{1},\ldots,\boldsymbol{n}}\}) \tag{Equation C.5}$$

With $\Theta_{1,\ldots,n}$ the probabilities of originating from nursery 1 to n. This vector of probabilities of origin followed a Dirichlet distribution (Equation C.6).

$$(\{\boldsymbol{\Theta}_{\mathbf{1},\ldots,\boldsymbol{n}}\}) \sim \boldsymbol{Dirichlet}(\boldsymbol{\alpha} * \{\boldsymbol{p}_{\mathbf{1},\ldots,\boldsymbol{n}}\}) \tag{Equation C.6}$$

With $\{p_{1,\ldots,n}\} = \{\frac{1}{n}, \ldots, \frac{1}{n}\}$ the base vector of probabilities and α a concentration parameter following a Gamma distribution as suggested by Dorazio, (2009) to mimic an uninformative prior (Equation C.7).

$$\boldsymbol{\alpha} \sim \boldsymbol{Gamma}(\boldsymbol{s}, \boldsymbol{r}) \tag{Equation C.7}$$

With s and r the shape and rate hyperparameters, respectively. They were preliminary computed so that (s,r) minimized the Stirling Number of first kind (See Dorazio, (2009) for details).

The Bayesian model and all statistical analyses were performed with R (R Development Core Team, R 3.4.3). The *rjags* package provided an interface from R Just Another Gibbs Sampling library (Plummer,

2003) and Monte Carlo Markov Chains (MCMC) were used to compute simulations from posterior distributions. Three chains were run in parallel with 40 000 iterations and a burn-in period of 10 000 iterations. The Gelman and Rubin, (1992) diagnosis allowed convergence checking of parameters using the *coda* package.

C.2. Supplementary results

- **Adult and juvenile otolith compositions**

Juvenile elemental concentrations of juvenile and adult individuals in Li, Mn, Sr and Ba (in ratio Ca) are presented in **Fig. C.2**.

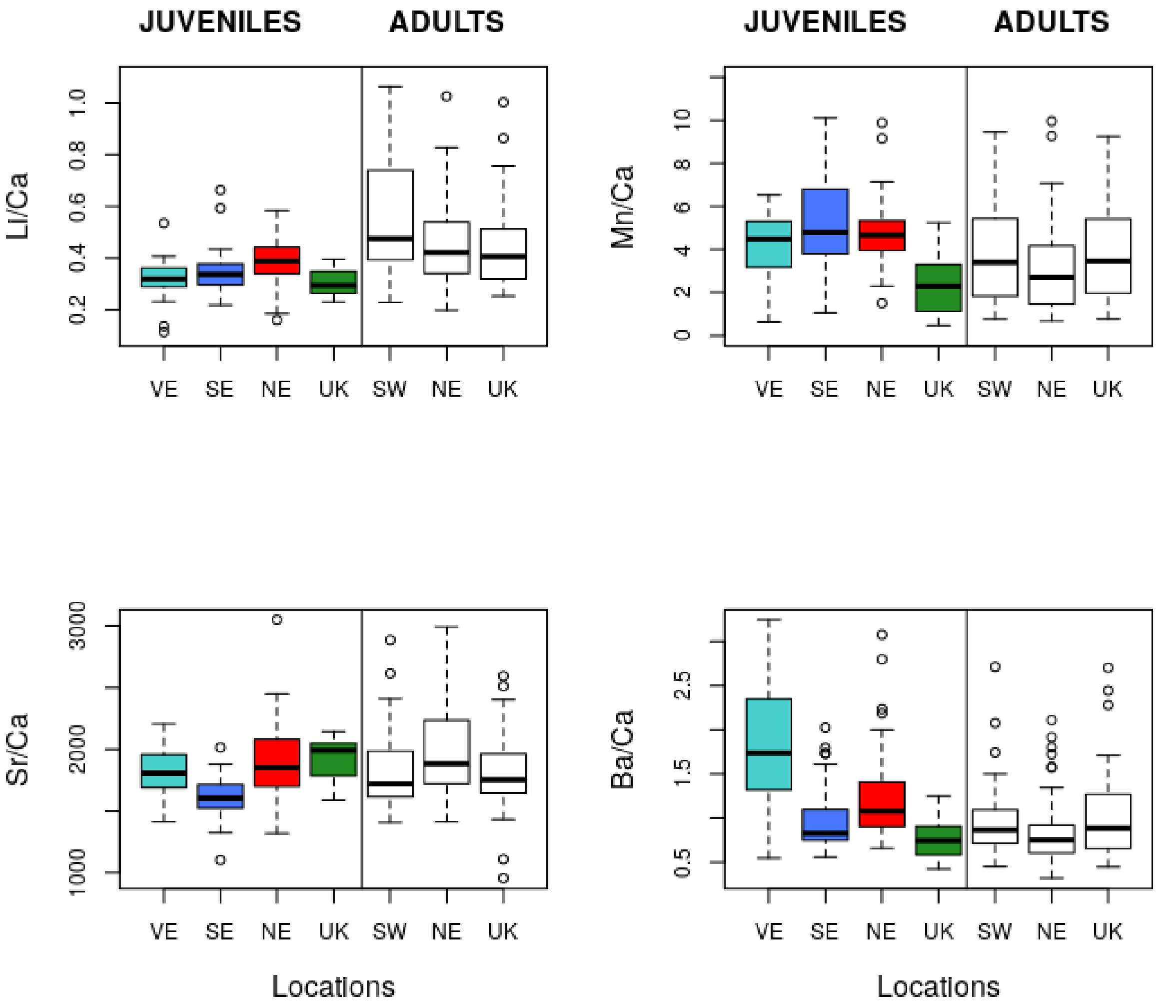


Figure. C.2. Juvenile (i.e. nursery) elemental concentrations in ratio Ca (µg.g^{-1}) of juvenile and adult soles at each location of the EEC.

- **Spatial variation in juvenile otolith composition**

Results of the analysis of variance conducted on the juvenile baseline is presented in **Table C.2**.

Table C.2. Results of ANOVA testing spatial (i.e. nursery) and temporal (i.e. cohort) differences in elemental concentrations. ‘×’ denotes interactions between factors. Statistical significance: ‘***’ P < 0.001; ‘**’ P < 0.01; ‘*’ P < 0.05

Elements	Effects	DF	F	p-value
Li	Nursery	3	6.757	<0.001 ***
	Cohort	2	2.631	0.0759 ·
	Nursery × Cohort	5	32.019	<0.001 ***
Na	Nursery	3	10.09	<0.001 ***
	Cohort	2	39.02	<0.001 ***
	Nursery × Cohort	5	5.28	<0.001 ***
Mn	Nursery	3	10.104	<0.001 ***
	Cohort	2	0.956	0.387
	Nursery × Cohort	5	3.395	0.00653 **
Sr	Nursery	3	15.710	<0.001 ***
	Cohort	2	2.922	0.0575 ·
	Nursery × Cohort	5	2.769	<0.05 *
Ba	Nursery	3	25.421	<0.001 ***
	Cohort	2	15.163	<0.001 ***
	Nursery × Cohort	5	3.352	0.00708 ***

List of publications

Randon M., *in prep*. A holistic investigation of tracers at population and individual scales highlighted population structure for the common sole of the Eastern English Channel. *In Prep.*

Randon M., *et al*. Combining genetic, morphological and chemical markers to assess population structure and trace connectivity within a flatfish stock. *Submitted in Estuarine Coastal and Shelf Science.*

Mahé K., Ider D., Massaro A., Hamed O., Jurado-Ruzafa A., Gonçalves P., Anastasopoulou A., Jadaud A., Mytilineou C., **Randon M.**, Elleboode R., Morell A., Ramdane Z., Smith J., Bekaert K., Amara R., de Pontual H., Ernande B., 2019. Directional bilateral asymmetry in fish otolith: is it a potential tool to evaluate the stock boundaries? *In* : Lestrel P.E. (Ed.) Biological Shape Analysis: Proceedings of the 5th International Symposium of Biological Shape Analysis. World Scientific, Pub Singapore and New Jersey, USA: *In press.*

Randon, M., Réveillac, E., Rivot, E., Du Pontavice, H., & Le Pape, O. (2018). Could we consider a single stock when spatial sub-units present lasting patterns in growth and asynchrony in cohort densities? A flatfish case study. *Journal of sea research*, *142*, 91-100.

Du Pontavice, H., **Randon, M**., Lehuta, S., Vermard, Y., & Savina-Rolland, M. (2018). Investigating spatial heterogeneity of von Bertalanffy growth parameters to inform the stock structuration of common sole, Solea solea, in the Eastern English Channel. *Fisheries research*, *207*, 28-36.

Randon, M., Daverat, F., Bareille, G., Jatteau, P., Martin, J., Pecheyran, C., & Drouineau, H. (2017). Quantifying exchanges of Allis shads between river catchments by combining otolith microchemistry and abundance indices in a Bayesian model. *ICES Journal of Marine Science*, *75*(1), 9-21.

Le Luherne, E., Le Pape, O., Murillo, L., **Randon, M.**, Lebot, C., & Réveillac, E. (2017). Influence of green tides in coastal nursery grounds on the habitat selection and individual performance of juvenile fish. *PloS one*, *12*(1), e0170110.

Du Pontavice *et al*., (2018)

Fisheries Research 207 (2018) 28–36

Contents lists available at ScienceDirect

Fisheries Research

journal homepage: www.elsevier.com/locate/fishres

Investigating spatial heterogeneity of von Bertalanffy growth parameters to inform the stock structuration of common sole, Solea solea, in the Eastern English Channel

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ARTICLE INFO

Handled by A.E. Punt.

Keywords:
Key-words
von Bertalanffy growth function
Stock structuration
Solea solea
Flatfish
Life-history traits
Metapopulation

ABSTRACT

In fisheries science, a mismatch between the delineation of a fish stock and the underlying biological population can lead to inaccurate assessment and management. Previous results suggested a potential spatial structuration of the Eastern English Channel (EEC) stock of common sole, *Solea solea*, in three sub-populations. In this article, we propose to investigate the spatial population structure of common sole in the EEC using the von Bertalanffy Growth Function parameters as indicators of population segregation. In order to test the sub-population hypothesis and evaluate its robustness to data sources, we developed three models, all including an area effect on growth parameters. The first model was aimed at testing a potential data source effect (in addition to the area effect) using commercial and scientific survey data jointly. The two other models used either scientific survey or commercial fishery data and focused on spatial differences in growth parameters. Our results showed that the growth parameter estimates indeed differed depending on the type of data used, with higher estimated asymptotic length and length at age two (L_2) using commercial data. They also highlighted spatial differences in asymptotic length, consistent between models, which tend to confirm a spatial structuration of sole in the EEC. While these results need to be strengthened by marking and genetic studies, they constitute a first step towards a better understanding of the population spatial structuration of common sole in the EEC.

1. Introduction

Harvested species are usually assessed and managed at the stock-unit scale, a stock being defined as an intraspecific group of individuals randomly mating and maintaining its integrity in time and space (Ihssen et al., 1981). Boundaries of these stock units are supposed to reflect the underlying population structures in terms of biological rates such as mortality and growth (Cadrin et al., 2014; Kerr et al., 2016). Recent research suggests that a strong population structuration in marine fish is a relatively common situation (Ames and Lichter, 2013; Ciannelli et al., 2013; Reiss et al., 2009; Waples and Gaggiotti, 2006), and is mainly driven by oceanographic and environmental factors and larval diffusion (Cowen, 2006; Jorgensen et al., 2005), but also by species' specific migratory behaviour (Secor, 2015) and sequential occupancy of various habitats throughout the lifespan (Petitgas et al., 2013, 2010). An assumed single homogeneous population can sometimes turns out to be a set of sub-populations linked by dispersal, i.e., a metapopulation (Alex Smith and Green, 2005; Hanski, 1998; Kritzer and Sale, 2004). Mismatch between the stock-unit delineation and the true metapopulation structure may impede our capacity to provide adequate management recommendations (e.g., Total Allowable Catch) and may lead to overfishing of the less productive sub-populations while the more productive sub-populations are underexploited (Cadrin and Secor, 2009; Frank and Brickman, 2000; Fu and Fanning, 2004; Ricker, 1981). In the past decade, stock identification has been explored for numerous fish stocks, such as blue whiting, Atlantic cod, and horse

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Abbreviations: VBGF, von Bertalanffy growth function; EEC, eastern english channel; SW, southwest; NE, northeast; ICES, International Council for the Exploration of the Sea; UK-BTS, UK beam trawl survey; NLR, non-linear regression; GNLS, generalized nonlinear least squares

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https://doi.org/10.1016/j.fishres.2018.05.009
Received 25 October 2017; Received in revised form 9 May 2018; Accepted 10 May 2018
0165-7836/

mackerel (Abaunza et al., 2008; Mahe et al., 2007; Zemeckis et al., 2014) and led to revise stock boundaries.

The Eastern English Channel (EEC) common sole, *Solea solea,* is a nursery-dependent flatfish species harvested across its entire range, from the Mediterranean to Baltic Sea (Wheeler, 1978). Reproduction occurs from winter to spring throughout the distribution area, resulting in several weeks of pelagic larval drift before settlement and metamorphosis in coastal and estuarine nursery grounds (Rochette et al., 2012). After two years, mature common soles are recruited to the stock and can in turn reproduce (Dorel et al., 1991; Riou, 2001). The common sole is a high value targeted fish in the EEC with some fleets highly dependent on it. Fishing mortality on EEC common sole has decreased over the last decade and was estimated below Fmsy - the fishing mortality that produces the maximum sustainable yield (ICES, 2017) - for the first time in 2017 (ICES, 2017) due to a series of low recruitments. However, the biomass is still below the targeted value (MSY Btrigger; ICES, 2017).

Contrasts in the length structure of French landed common sole between the North and South of the EEC raise concerns about a possible misunderstanding of the stock structure (Du Pontavice et al., personal communication). The fleets fishing on the EEC sole are segregated in space, across areas corresponding to potentially different components of the population. It is therefore crucial to improve our knowledge of spatial structuration and population connectivity within the EEC stock and to check whether the hypothesis of a unique stock matches the underlying population structure. A body of research already investigated the level of connectivity of common sole population in the EEC at different stages of the life cycle. First, larval advection to coastal nursery grounds has been shown to limit the connectivity between the different spawning and nursery grounds (Rochette et al., 2012). Second, previous analyses evidenced juveniles common sole as sedentary in their nursery grounds during the two first years of life (Coggan and Dando, 1988; Le Pape and Cognez et al., 2016). Finally, former mark-recapture surveys suggested low mobility of adult common sole (Burt and Millner, 2008; Kotthaus, 1963). On the basis of these results, recent modelling studies hypothesized the existence of three sub-populations spatially structured within the EEC (Fig. 1; (Archambault et al., 2016; Rochette et al., 2012). Archambault et al., (2016) assessed the effect of adult-mediated connectivity on population dynamics and stock assessment and concluded that ignoring possible metapopulation could lead to overexploitation of local populations in the EEC. The authors also suggested that research should focus on the adult-mediated connectivity, which remained largely unknown and which magnitude could be a strong driver of spatial structuration within the EEC (Frisk et al., 2014). Recently, the improvement and the multiplication of stock identification tools has made stock delineation increasingly precise (Cadrin et al., 2014; Kerr et al., 2016; Pita et al., 2016). In particular, the use of life-history parameters (e.g., age, growth and mortality) is particularly relevant and cost-efficient (Cadrin et al., 2014). For instance, Barrios et al. (2017) coupled individual growth trajectories and length-at-age datasets in mixed-effects models to investigate stock identification of whiting in the North East Atlantic. Erlandsson et al. (2017) suggested a reconsideration of assessment models of European Flounder based on the spatiotemporal structure of body size in the Baltic Sea. Given the before-mentioned differences in EEC common sole length structure, we propose to study the spatial heterogeneity in length at age as a way to identify spatial structuration within the stock and to test the three common sole sub-populations hypothesis mentioned earlier. Growth is usually modelled using the von Bertalanffy Growth Function (VBGF; Von Bertalanffy, 1938) which provides a non-linear relationship between length and age of organisms. In the present work, we do not aim at describing the growth processes but rather at summarising the characteristics of the length-at-age relationship in delineated subareas in the EEC stock using the three parameters of the VBGF. To avoid bias in the analysis, length-selection in data collection must be avoided and the spatial and temporal coverage has to be representative of the spatial entity considered. In the EEC, two types of data were available to us: data from a scientific survey (the UK Beam Trawl Survey, UK-BTS) and sampling data from French commercial landings. Differences in length-at-age between survey and commercial data can be expected: scientific survey are designed to reflect the length structure of the population, while the length structure of commercial landings is influenced by the minimum landing size imposed by EU on common sole, through fishing strategy (e.g., commercial fisheries tend to fish in areas with the biggest common soles), size-selectivity, and discarding practices.

In this study, we investigated the spatial variability in VBGF parameters within the EEC stock of common sole in order to inform current

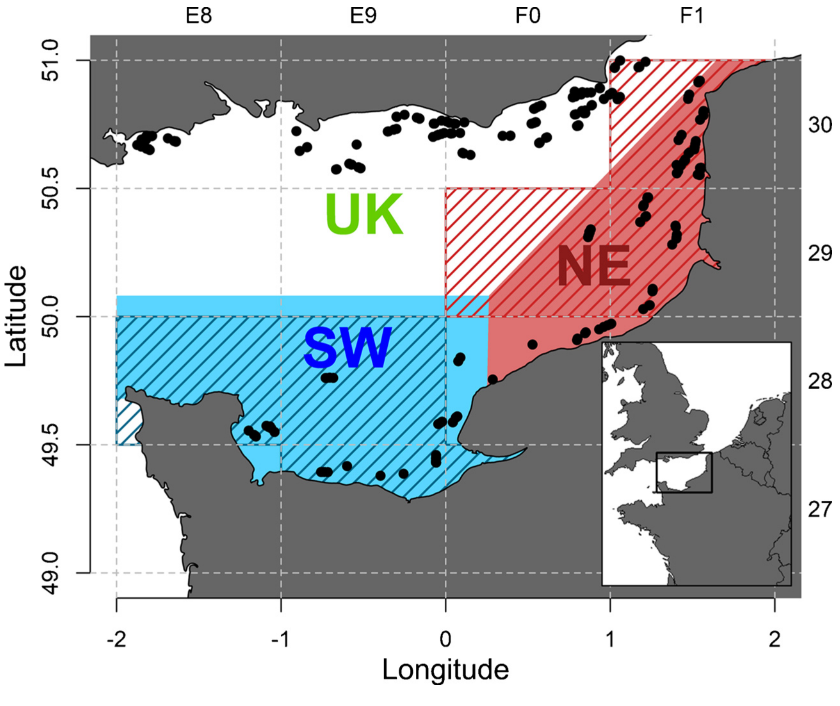

Fig. 1. The three subareas of the Eastern English Channel as proposed in (Archambault et al., 2015; Rochette et al., 2012) (UK: United Kingdom, NE: Northeast, SW: Southwest). The points represent haul positions of the UK-BTS (UK Beam Trawl Survey). The two coloured areas SW and NE in represent the subareas with French commercial sampling and the cross-hatching ICES (International Council for the Exploration of the Sea) statistical rectangles were assigned to the Northeast and Southwest subareas respectively. Top and right axes correspond to the ICES statistical rectangle coordinates.

interrogations about population spatial structuration. Based on the three subpopulations hypothesis (Archambault et al., 2016; Rochette et al., 2012), (Archambault et al., 2016; Rochette et al., 2012), we first evaluated the spatial differences in VBGF parameters accounting for potential bias due to the type of data used (i.e., commercial or survey data). Then, we used the two types of data separately to inform spatial growth differences between the three subareas of the EEC stock of common sole.

2. Materials & methods

2.1. Data

Biological data for common sole in the EEC were extracted from the French commercial sampling program and onboard sampling during the UK Beam Trawl scientific Survey.

2.1.1. Data from French commercial fisheries

Samples from commercial fisheries were collected in fish markets and provided individual information on age and total length (10 mm classes) by quarter, year, ICES division and fishing gear. In order to determine the subarea of origin, the fishing location of each sample had to be identified (Fig. 1): Each sampled fish was linked to a statistical rectangle by using logbook information for the corresponding fishing trip. Two main assumptions had to be made for this association: (i) sampled common soles were allocated to the statistical rectangle where the fishing vessel caught the largest volume of common sole during the trip and (ii) in the NE subarea, common soles caught by netters in a statistical rectangle shared by two subareas were assigned to the subarea closest to the shore (for instance in ICES statistical rectangle 30 F1 (Fig. 1), common soles caught by netters were assigned to the NE subarea). Indeed French netters usually operate in French coastal zones within subareas NE and SW (Fig. 1). Biological data from commercial sampling comes from trammel netters in the NE of the EEC and from trammel netters (31%), bottom trawlers (63%), and unidentified gears (5%) in the SW. The French sampling program operates quarterly and individuals are randomly selected in the fish markets using a stratified sampling by sex and length-class. The data used cover the period 2010 to 2015.

2.1.2. Data from scientific survey

The UK-BTS survey has been carried out annually in July/August since 1989 by the Centre for Environment Fisheries and Aquaculture Science (CEFAS), using a commercially rigged 4 m steel beam trawl (ICES, 2009). Samples from UK-BTS provided individual information on age and total length with the exact haul position (Fig. 1). At each haul, individuals are randomly selected using a stratified sampling by sex and length-class. UK-BTS data are publically available and were extracted from the International Council for the Exploration of the Sea (ICES) database (www.ices.dk/marine-data/) for the period 2010 to 2015.

2.1.3. Differences between the two data sources

The two data sources differ by their spatial and temporal coverage, and observed age composition. In both biological data sources, age estimation was conducted by otoliths sagitta reading by IFREMER in France for commercial data and by CEFAS for survey data. Reader effects on otolith reading have been tested in workshops (involving both IFREMER and CEFAS) and agreement between institutes was 91% for all ages (ICES, 2009). Both data series are available over the period between 2010 and 2015. Survey data are substantial from age 1 (common soles at age 1 represent 21% of total number of fish) while commercial data become substantial only from age 2 (< 1% of age 1 in the third quarter). This is likely due to the minimum landing size of 24 cm in place for sole (Supplementary material S1).

2.2. Model formulation

Differences in length at age were investigated through analyses of the VBGF parameters. As in Rindorf et al. (2016), cohort effects were ignored considering the short period of observations in comparison with common sole life span. Year effects, usually included as proxies for annual environmental conditions, were also neglected because the purpose was not to assess precisely growth parameters but rather to focus on differences between subareas and data sources. The growth equation was parameterized from age 2,

$$L_{t,i} = L_{\infty} - (L_{\infty} - L_2) \times exp(-K \times (t_i - 2)) + \varepsilon_i \tag{1}$$

where ε_i is a normally distributed error term.

$L_{t,i}$ is the length of the individual i at age t, L_2 the length at age 2, L_∞ the asymptotic maximum length, and K (year^{-1}) is the intrinsic somatic length growth rate (i.e., the speed at which the asymptotic length is reached).

To identify the influence of subareas and data sources on the estimates of the growth parameters, a generalized nonlinear least squares (GNLS) model was used. This model was formulated using a two-stage framework (Pinheiro and Bates, 2000).

First, the length of the i-th common sole is modelled by Eq. (1).

Second, covariates were introduced in the model. The covariates tested included (i) the three subareas (denoted “subarea”), (ii) the two data sources (denoted “data”), (iii) the sexual dimorphism (denoted “sex”), and (iv) the annual growth continuity (denoted “quarter”). The three growth parameters can be expressed as a vector: $\phi_i = [K_i, L_{\infty,i}, L_{2,i}]$ with $\phi_i = A_i.\beta$. A is the designed matrix whose size depends on the number of covariates and the number of groups in each covariate, and β is the vector of parameters for the covariates.

For example, in a model considering the three subareas in the EEC (NE, SW, and UK) and the sexual dimorphism (Male or Female) (i.e., Model 2), K, L_∞, and L_2 can be expressed as: $\{K = \beta_1 + \beta_2.SubareaSW + \beta_1.SubareaUK + \beta_4.SexMale\}$, $\{L_\infty = \beta_5 + \beta_6.SubareaSW + \beta_7.SubareaUK + \beta_8.SexMale\}$, $\{L_2 = \beta_9 + \beta_{10}.SubareaSW + \beta_{11}.SubareaUK + \beta_{12}.SexMale\}$.

Subarea and data were the covariates of interest: subarea captures potential variations in length-at-age between subareas in the EEC and data documents the potential differences in the perception of length-at-age relationships between commercial and survey data.

Both sex and quarter effects were used to avoid bias in estimation. Indeed, sexual dimorphism was described for common sole by several authors (De Veen, 1976; Rijnsdorp and Van Beek, 1991). Moreover, the fishing strategy (fishing area and period) of commercial fisheries leads to a high share of female in the capture (81%) compared to the scientific survey (45%). The quarter effect aimed at capturing the variability caused by the annual growth continuity and reducing the incidence of unbalanced sampling in the commercial fisheries. Indeed, in the NE of the EEC common sole are sampled homogeneously throughout the year, while, in the SW, 83% of common sole are sampled in the 2nd and the 3rd quarter.

This general form was then applied in three alternative models which differed by the datasets and the covariates considered (Summarised in Table 1).

2.3. Model description

2.3.1. Model 1: commercial and survey data

The first model (Model 1) was aimed at verifying if commercial and scientific data may be jointly used in estimation or if a data-source bias exists. To allow comparison between the two data sources, we only considered commercial data collected during quarter 3 (because the scientific survey is carried out in July and August), and survey data from NE and SW subareas due to the absence of catch by French commercial fisheries in the UK subarea. The model was fitted on a resulting dataset of 3113 length-age couples from survey (45%), and

Table 1

Summary of the three models implemented to analyse the effects of sex, subarea, quarter, and data source on growth parameters K, L_∞ and L2, across the three subareas NE, SW and UK. For each model, the ticks represent the inclusion (or not) of effects tested on each growth. The green boxes are the effects selected by the models. Q stands for Quarter. (For interpretation of the references to colour in this table legend, the reader is referred to the web version of this article).

	Data effect		Growth comparison between subareas				
	Model 1		Model 2		Model 3		
Data Period	BTS + Commercial Q 3 - SW NE		BTS data Q 3 – All areas		Commercial All quarters - SW NE		
Effects included in full models	Sex	data x subarea	Sex	Subarea	Sex	Subarea	Quarter
K	√	√	√	√	√	SW, NE	√
L_∞	√	√	√	√	√	SW, NE	Not tested
L2	√	√	√	√	√	SW, NE	√

commercial fishery (55%).

The effect of sex (β_1), subarea (β_2), data source (β_3) and the interaction between subareas and data source (β_4) were included in the model. Consequently, the model produces independent estimations for each combination of data source and subarea.

2.3.2. Model 2 and 3: independent analysis of commercial and survey data: subarea effect on the estimates of growth parameters

Model 2: Survey data

The second model was fitted to survey data only and aimed at detecting potential differences in length at age between the three subareas, independent from bias due to different fishing practices across regions (e.g., differences in mesh size, targeting of sizes and discards). The non-linear model tested the effects of subarea (β_1) and sex (β_2).

We considered 2863 individuals of age 1 to age 23 from UK-BTS for the 3 subareas: NE (33%), SW (16%) and UK (50%) (the age structure in the dataset is described in Supplementary material S2). We noted that the quantity of data in the SW subarea is relatively low in comparison with the NE and UK subareas.

Model 3: Commercial data

Model 3 was fitted to data from commercial fisheries and tested the effects of subarea (β_2), season (β_3) and sex (β_1). As for model 1, data were available only in the two subareas fished by the French commercial fleets (i.e., NE and SW subareas). Commercial data are sampled throughout the year, thus a quarter effect was added in order to consider the annual growth continuity.

The interest of this model lies in the quantity of available data (5391 individuals of age 1 to age 24) particularly in the SW subarea wherein the quantity of data was low in the scientific survey in comparison with the NE and the UK subareas (Supplementary material S3 and S4). We considered a quarter effect on K, because the speed of growth may depend on environmental seasonal conditions. Inversely we did not consider a quarter effect on the asymptotic length, which is viewed as the maximum length reached by the population and reflects the cumulative growth history of fish thus constant throughout the year at population scale.

2.4. Model selection

The three models were implemented in the statistical software R (R Core Team, 2016) using the 'gnls' function (package 'nlme'; (Pinheiro and Bates, 2000). Backward stepwise procedure was performed and the Akaike Information Criterion (AIC) and the Bayesian Information Criterion (BIC) were used to select the best model. The initial model was the full model (all covariate effects on all three parameters) and the impact on the criteria of the sequential deletion of each covariate on each parameter was assessed. The covariates whose deletion either improved the model (by reducing AIC and BIC) or did not deteriorate its quality (not significant difference in AIC and BIC) were eliminated. The process was repeated until no further improvement was possible. The significance of the parameters was assessed using a Wald chi-square test. Graphical methods (e.g., residual diagnostic plots) were used to check that the final models verified the normality assumption (Supplementary material S12).

Finally, in order to evaluate the robustness of the conclusions to the shape of the growth curve, we estimated a logistic growth model as an alternative to the VBG model using the UK-BTS data in the SW of the English EEC and the effects selected in Model 2 (Described in the following section; Supplementary material S13). Models were compared using AIC and BIC.

3. Results

3.1. Data source effect on growth parameters

Model 1 was based on commercial fisheries and survey data in the two subareas NE and SW of the EEC and considered the effects of sex, subarea, data and interaction subarea x data on growth parameters. It revealed a data source effect on K and L_2 with different magnitude depending on the subarea globally higher in the NE than in the SW. It also suggested differences in growth between subareas.

The model considering the four effects on the three growth parameters (K, L_∞ and L_2) was always selected based on the AIC selection criteria (Supplementary material S6 and S7). In the case of the more conservative BIC criteria, the gain obtained by considering the effect on the growth parameter was K quite low ($\Delta BIC_{S1\text{-}S0} = 1.07$ and $\Delta BIC_{S4\text{-}S0} = 0.4$). Consequently, we kept the full model keeping in mind that the four effects on K might be unclear.

Wald chi-square test indicated that all the parameters were significant with a p-value < 0.001, except for the parameter L_∞ associated to the commercial data in the SW subarea (p-value = 0.055).

The sex effect on the three growth parameters was significant and of similar amplitude as the three other effects (Supplementary material S6 and Table 2).

Interaction effect in Model 1 allowed to provide independent estimates for each data source in each subarea. The results showed that differences depending on the data sources in L_∞, L_2 and K. L_∞ was 14.5% (46.5 mm) higher using commercial data than using UK-BTS in NE, and 5.5% (19 mm) in the SW. L_2 was 14.5% (32.5 mm) higher using commercial data than using UK-BTS in NE and 9% (20 mm) in SW. K was 33% lower using commercial data than using UK-BTS in NE whereas, in the SW, K was 4% higher using commercial data.

The asymptotic length and the length at age 2 were significantly higher using the commercial data compared to the UK-BTS data in the SW and the NE of the EEC, while K were significantly lower using commercial data in the NE and higher in the SW.

The results also showed differences in growth parameters between the two subareas with the two data sources (Fig. 2), which were consistent across data source for L_∞, i.e., higher L_∞ in the SW of the EEC.

In the following part, the subarea effect was analysed in details

Table 2
Growth parameters of the von Bertalanffy equation computed from the model fitted to data from the scientific survey UK-BTS and commercial sampling, considering data source effect and sex effect. The columns' % variation' specifies the percentage of variation in each subarea between the data from commercial fisheries and the data from UK-BTS.

			Male		Female	
Growth Parameters	Subarea	Sampling type	Value	% difference (ref BTS in the same area)	Value	% difference (ref BTS in the same area)
K	NE	BTS	0.40		0.33	
	NE	COM	0.28	−30	0.21	−36
	SW	BTS	0.39		0.32	
	SW	COM	0.41	+ 4	0.33	+ 4
L_∞	NE	BTS	284		352	
	NE	COM	330	+ 16	399	+ 13
	SW	BTS	317		385	
	SW	COM	336	+ 6	404	+ 5
L_2	NE	BTS	207		224	
	NE	COM	239	+ 15	255	+ 14
	SW	BTS	218		234	
	SW	COM	238	+ 9	254	+ 9

separating data from scientific survey and from commercial fisheries.

3.2. Variations in growth parameters across subareas in the EEC

3.2.1. Scientific survey UK-BTS data within the three subareas in the EEC

In Model 2, which considered the subarea and sex effects using scientific survey data, the complete model considering all effects on all parameters was selected based on the AIC selection criteria (Supplementary material S8 and S9). On the basis of the BIC criterion, the sub-model S4 would be selected (which releases the subarea effect on K). Even if AIC and BIC criteria did not provide the same message on the model selection, we chose to keep the subarea effect on K to be able to look at the potential subarea effect. Wald chi-square test indicated that all the parameters were significant with a p-value < 0.001.

In this model, sexual dimorphism had a larger effect on the three growth parameters than subareas. L_∞ and L_2 were lower in males than females, and K was higher in males (Fig. 3 and Table 3).

The asymptotic length, L_∞, showed the largest variation across the three subareas. L_∞ was much higher in the SW than in the NE of the EEC (+10% for males and +9% for females) and in the UK (+7% for males and +6% females) (Table 3). The same result was observed, to a lesser extent, for L_2 (+4% and +5% between the NE and SW subareas and +2% between the UK and SW subareas). The subarea effect on K followed a different pattern with K around 0.40 for males (and 0.31 for females) in the SW and NE subareas whereas it reached 0.46 for males (and 0.37 for females) in the UK subarea.

Both L_∞ and L_2 were higher in the SW of the EEC than in the UK and the same parameters were higher in the UK than in the NE. This means that the differences in terms of length-at-age exist from youngest age and spread throughout the life cycle of the common sole.

3.2.2. Commercial fisheries data within the two subareas in the EEC

Model 3 is based on commercial fisheries data with three effects (subarea, quarter and sex) on the three growth parameters. Unlike the two previous models, the fit of this model required four steps and led to the removal of the subarea effect on K and L_2 and the sex effect on K, based on both AIC and BIC (Supplementary material S10 and S11). The Wald chi-square test indicated that all the parameters were significant with a p-value < 0.001.

The subarea effect and the sex effect were retained for L_∞. This parameter was significantly higher in the SW than in the NE subarea (+14% for male and +11% for female) (Fig. 4 and Table 4). This was consistent with the results obtained with the survey data. K showed quarterly variation with higher values for the quarters 2 and 3 that were

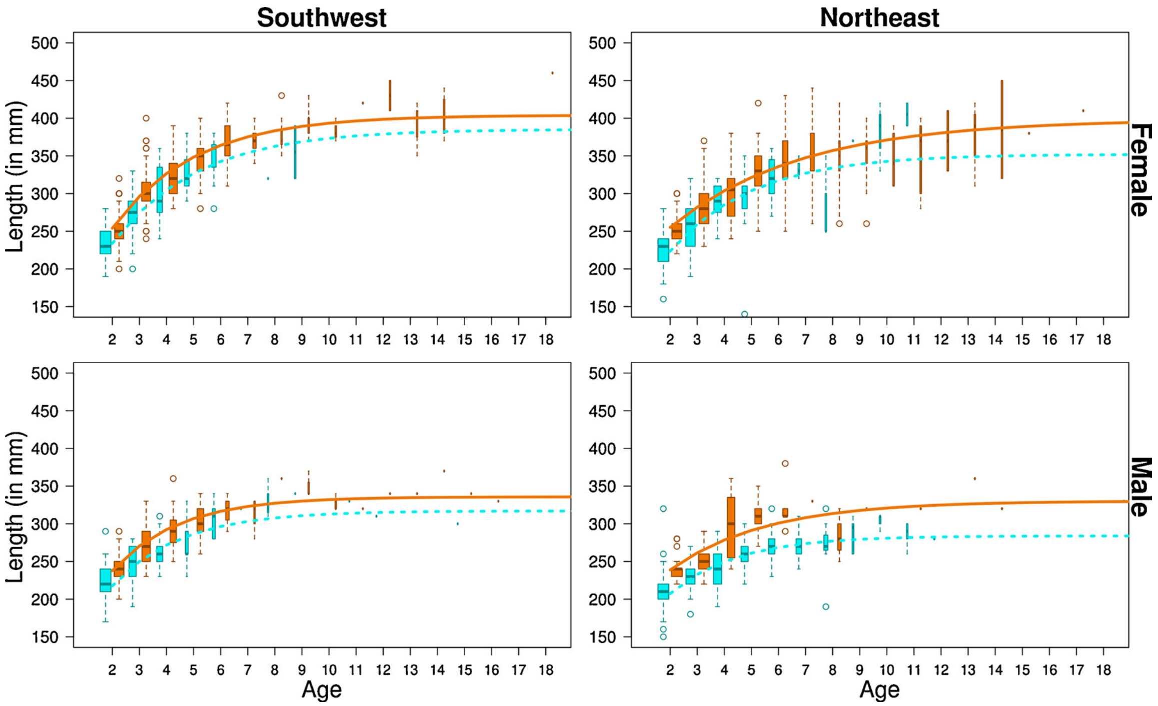

Fig. 2. Von Bertalanffy growth curves for the Northeast and Southwest subareas of the Eastern English Channel for each data source, UK-BTS (UK Beam Trawl Survey) (dashed line in blue) and French commercial fisheries (solid line in orange). The curves separate female and male and were plotted from growth parameters computed from the model fitted with the two data sources. The boxplots represent the data distribution at each age, their widths are proportional to the number of data at each age and for each data source and the dots are the outliers at each age (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article).

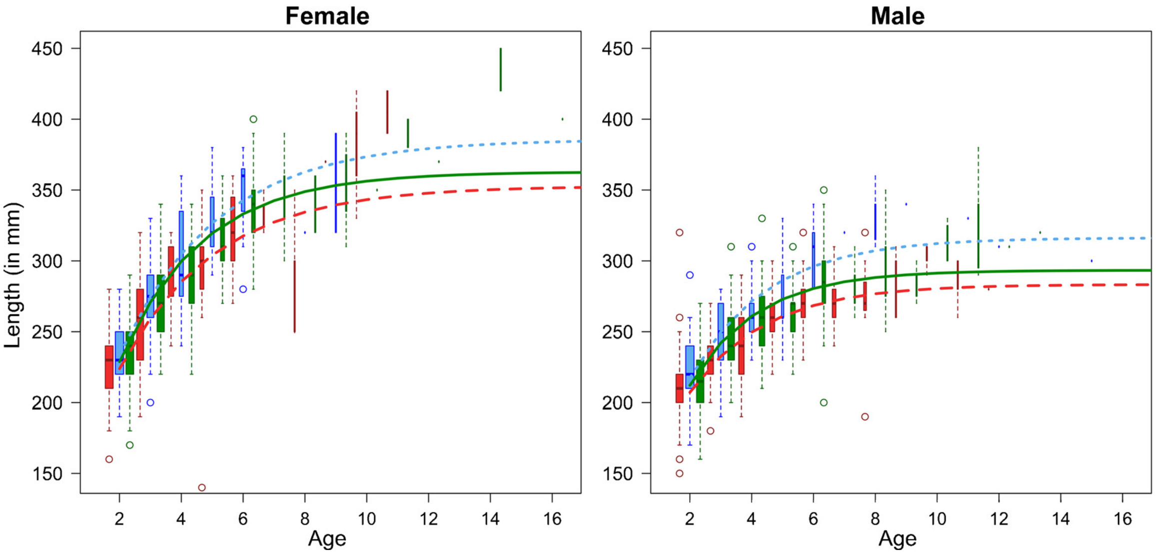

Fig. 3. Von Bertalanffy growth curves for Northeast (dashed line in red), Southwest (dotted line in blue) and UK (solid line in green) subareas of the Eastern English Channel separating female and male, plotted from growth parameters computed from model fitted with scientific survey. The boxplots represent the data distribution at each age, their widths are proportional to the number of data at each age and in each subarea and the dots are the outliers at each age (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article).

Table 3
Growth parameters of von Bertalanffy equation computed from the model fitted on data from French commercial fisheries considering subarea effect and sex effect. The columns '% variation' indicates the percentage of variation between the subareas with the SW subarea as the reference.

			Male		Female	
Growth Parameters	Subarea	Data source	Value	% difference (ref SW subarea)	Value	% difference (ref SW subarea)
K	SW	BTS	0,40		0,31	
	NE	BTS	0,41	+2%	032	+2%
	UK	BTS	0,46	+16%	037	+16%
L_∞	SW	BTS	316		386	
	NE	BTS	283	−10%	353	−9%
	UK	BTS	293	−7%	363	−6%
L_2	SW	BTS	217		234	
	NE	BTS	207	−5%	224	−4%
	UK	BTS	212	−2%	229	−2%

not consistent with intra-annual growth. Finally, sexual dimorphism induced variations in L_∞ and L_2 but not in K.

The results showed significant differences across subareas in asymptotic length, L_∞, in all models, and differences in length at age 2 (L_2) with the scientific survey data (Model 1 and Model 2) (Table 1 in green). Asymptotic length was systematically higher in the SW of the EEC than in the NE. Same results were found to a lesser extent for length at Age 2 (L_2) which was higher in the SW of the EEC than in the NE. The growth rate (K) showed small variations between the SW and the NE. The UK subarea was explored only with the data from scientific survey in the Model 2. Nevertheless, the results indicated that K was much higher in the UK subareas than in the two other subareas, whereas the value of L_∞ was intermediate between those in the SW and in the NE subareas.

4. Discussion

The use of life-history traits, especially growth parameters, is a well-known method to inform stock identity (Abaunza et al., 2008; Barrios et al., 2017; Cadrin et al., 2014; Sequeira et al., 2012). Herein, we investigated the stock spatial structuration of common sole in the EEC using VBGF to estimate growth parameters, according to an existing three sub-populations hypothesis (Archambault et al., 2016; Rochette et al., 2012). From a methodological perspective, we compared growth parameter estimates using length-at-age data from a scientific survey and a commercial fisheries sampling program. While the estimation of VBGF parameters, K and L_∞, for common sole was not the primary aim of the study, the parameters obtained are in the range (or slightly lower for L_∞; Deniel, 1990; Lorenzen and Enberg, 2002) of those estimated for the same species in surrounding regions (Carpentier et al., 2009; De Veen, 1976; Deniel, 1990; Lorenzen and Enberg, 2002).

4.1. *Effects of the data source on the growth parameter estimates*

4.1.1. *Different fishing practices provided different perceptions of length-at-age population structure*

Model 1 was developed to test a potential data source effect on the growth parameter estimates. We found that the three growth parameters estimates were indeed strongly influenced by the data source used in the VBGF model. More precisely, in the two subareas considered in Model 1 (i.e., the SW and NE subareas), L_∞ and L_2 estimates were higher using commercial data compared to survey, particularly in the NE subarea.

This data source effect can be attributed to size-selectivity differences between the commercial and scientific fishing practices. The smallest common soles at age 2 (and notably the males, which are smaller) are not captured or not landed by commercial fisheries, while the scientific survey catches common soles from age 1. Commercial fleets are much more size-selective than scientific survey due to a minimum landing size (24 cm) imposed by the European Union regulation, and the low commercial value of the smallest commercial category of common sole. This selectivity is induced by gear characteristics (such as mesh size) and by the targeting behaviour. Commercial fisheries tend to fish in areas with the biggest common soles, while scientific survey aims at sampling the entire population using a stratified random design. The perception of the population structure is consequently dependent of the type of data used. Differences in the magnitude of the data source effect between the NE and the SW of the EEC were presented and they may be due to differences in the fishing gears used by the commercial fishery in these two subareas. In the NE, the fishery is mainly composed of trammel nets with smaller mesh size

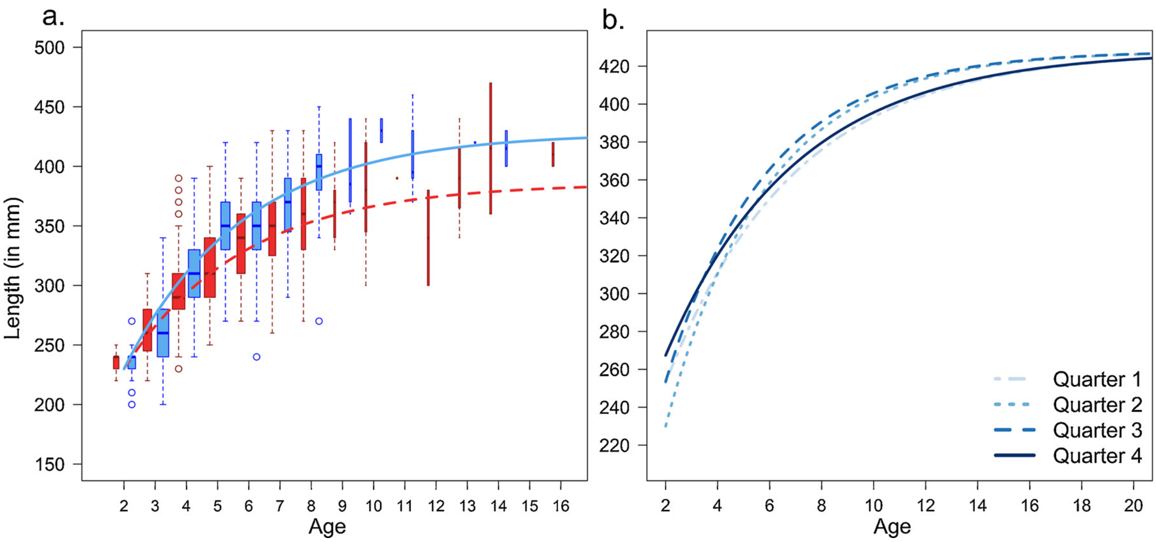

Fig. 4. Von Bertalanffy growth curves plotted from growth parameters computed from model fitted with commercial fisheries: a. for Northeast (dashed line in red) and Southwest (solid line in blue) subareas for females during the quarter 2. b. for each quarter for females in the SW subareas. The boxplots represent the data distribution at each age, their widths are proportional to the number of data at each age and in each subarea and the dots are the outliers at each age (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article).

Table 4
Growth parameters of von Bertalanffy equation computed from the model fitted on data from French commercial fisheries considering subarea effect, sex effect and quarter effect.

			Male		Female	
Growth Parameters	Subarea	Quarter	Value	% difference (ref NE subarea)	Value	% difference (ref NE subarea)
K	NE/SW	1	0.20		0.20	
	NE/SW	2	0.26		0.26	
	NE/SW	3	0.26		0.26	
	NE/SW	4	0.20		0.20	
L_∞	NE	1/2/3/4	310		386	
	SW	1/2/3/4	352	+ 14	428	+ 11
L_2	NE/SW	1	243		253	
	NE/SW	2	220		230	
	NE/SW	3	244		254	
	NE/SW	4	258		267	

whereas, in the SW, it is composed of trammel nets with bigger mesh size, and bottom trawls.

Ones should be careful when combining length-at-age data from various data sources and especially commercial and survey data to build models. Ignoring data source can lead to skew the perception of length-at-age population structure.

4.1.2. Methodological limitations: significance of the estimates and correlation between von Bertalanffy growth parameters

In the paper, we used the well-known VBGF as a way to summarise the length-at-age in the population. A logistic growth model was also tested and suggested significant differences between subareas as well. However, the best fits were obtained using VBG model (Supplementary materials S13).

We included a season effect in the model applied to commercial data as a covariate instead of using decimal ages to account for continuous growth. Indeed, as evidenced by the estimates of the quarter effect, intra-annual continuous growth was not supported by the data (Supplementary materials S14). This surprising result will lead to further analysis in the future. Meanwhile, accounting for the quarter effect avoids propagating the bias in VBGF estimates.

It is important to highlight the correlation between the growth rate (K) and the asymptotic length (L_∞). In Model 1, in the NE subarea, K estimate was lower with commercial data compared to survey data whereas the two other growth parameters L_∞ and L_2 were higher with commercial data. In the same way, in Model 2 and 3, K was found to vary oppositely to L_∞. Correlation matrix of the estimates of Model 1 and Model 2 showed a relatively high degree of correlations between in K and L_∞ estimates (between -0.87 and -0.81 for the same effect in both models; (Supplementary materials S15 and S16). This correlation between growth parameters may be an artefact of the models' parametrization (Schnute, 1981). However, it may also result from an interaction between two biological processes, growth and maturation especially between K and L_∞ (Brunel et al., 2013; Lester et al., 2004; Quince et al., 2008).

4.2. A first insight into the stock structuration of common sole in the EEC

4.2.1. Spatial differences in the asymptotic length (L_∞), in length-at-age 2 (L_2) and in growth rate (K)

The second and third models integrated length-at-age data from a scientific survey (Model 2) and commercial fisheries (Model 3), respectively. Both models highlighted that asymptotic length was significantly higher in the SW than in the NE subareas, with the same magnitude between models. Model 2 showed that the length-at-age 2 were also higher in the SW than in the UK subarea. This shows that differences in terms of length-at-age exist from the youngest age and spread throughout the life cycle of the common sole. Furthermore, model 2 highlighted that the growth rate (K) was much higher in the UK subarea compared to the NE and the SW of the EEC. However, we could not confirm these results using the French commercial data. Genetic (intrinsic) and environmental (extrinsic) components are likely to be explicative factors of such differences (Swain et al., 2007). Among extrinsic components, water temperature and density-dependent factors are particularly cited in the literature (e.g., Brett et al., 1969; Brett, 1979; Weatherley, 1990; Rijnsdorp and Van Beek, 1991; Sinclair et al., 2002; Castillo-Jordán et al., 2010). Mollet et al. (2013) showed latitudinal variations in asymptotic length of female *Solea solea* linked to the temperature in the Northern Atlantic. In our case, further research is needed to understand if growth differences in the EEC could be explained by abiotic or biotic factors.

Spatial differences in growth parameters support the hypothesis of low mobility of common soles in the EEC. While the movements of common soles at early stages are partially understood in the EEC (Archambault et al., 2015, 2016; Rochette et al., 2012), the mobility of adults remains poorly resolved, but preliminary reprocessing of former mark-recapture surveys data (Burt and Millner, 2008; Kotthaus, 1963) suggested little exchanges between the three subareas (Véron and Rivot, pers. comm). Additional mark-recapture data are currently collected to further support this conclusion.

4.2.2. *A differential fishing impact within the EEC?*

Given the strong fishing pressure on common sole in the EEC, these differences in the estimated growth parameters between subareas may be induced, at least partly, by heterogeneous fishing pressure on common sole across areas.

One of the first and direct effect of size-selective fishing is to reduce the share of larger individuals in the population, which can lead to a genetic change toward smaller individuals (Law, 2000; Ricker, 1981). The low asymptotic length in the NE in the EEC could therefore be due to a higher fishing pressure in this subarea compared to the SW and the UK.

More generally, fish growth can be subject to a high degree of genetically-based variations and therefore has the potential to evolve rapidly in response to harvesting (Law, 2000; Lorenzen, 2016). Evolutionary effects of fishing on growth may arise from multiple mechanisms including size selective fishing (Enberg et al., 2012). Fishing induced evolution can lead to a decrease of asymptotic length, as shown by Edeline et al., (2009 and 2007) in *Esox Lucius*. The lower growth rate and asymptotic length in the NE of the EEC where the fishing effort is the highest seems to follow this pattern. This is in line with (Archambault et al., 2016) who, under the hypothesis of the existence of distinct sub-populations, estimated a higher fishing mortality in the NE than in the UK since 2000, with the lowest fishing mortality in the EEC in the SW since 1985.

The analysis presented herein was conducted on a short and recent period (2010-2015). To explore the hypothesis of differential fishing impact on the potential of common sole in the EEC, it would be necessary to study the size-at-age data throughout a longer time-series in the three subareas (using UK-BTS survey data from 1989 to 2016 for instance).

Finally, our analysis was a first step towards the investigation and the understanding of the potential spatial stock structuration of common sole in the EEC. Spatial differences in the asymptotic length appeared consistent between the models, whatever the type of data used. Moreover, the analysis of the survey showed spatial differences in growth rate and length at age 2 in Model 1 and Model 2. These results were in favour of a potential spatial stock structuration following a three subpopulations hypothesis. The confirmation of the existence of three isolated subareas could have major implications on our perception of the stock and consequently for management. Currently, both stock assessment and management assume a single and homogeneous population and until 2016, the EEC stock has been exploited above MSY (ICES, 2016). Accounting for metapopulation dynamics is essential in assessment models to avoid local over-exploitation (Archambault et al., 2016; Tuck and Possingham, 1994; Ying et al., 2011). To inform this potential metapopulation structure, complementary studies are required (using genetics, otoliths or other life history traits) and would deserve to be integrated in an interdisciplinary approach which is considered to be the best approach to investigate stock identity (Begg and Waldman, 1999; Cadrin et al., 2014; Pita et al., 2016).

Acknowledgment

We authors are grateful to Bruno Ernande (Ifremer Boulogne-sur-Mer), who provided help and advice on statistical methods. This study was funded by the research program SMAC (France Filière Pêche, DPMA, IFREMER, Agrocampus Ouest, région Hauts-de-France and région Normandie). Publically available data from the UK-BTS survey operated by CEFAS were used in this study. French commercial data were made available by Ifremer SIH – Système d'informations Halieutiques.

Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi: https://doi.org/10.1016/j.fishres.2018.05.009.

References

Abaunza, P., Murta, A.G., Campbell, N., Cimmaruta, R., Comesaña, A.S., Dahle, G., García Santamaría, M.T., Gordo, L.S., Iversen, S.A., MacKenzie, K., Magoulas, A., Mattiucci, S., Molloy, J., Nascetti, G., Pinto, A.L., Quinta, R., Ramos, P., Sanjuan, A., Santos, A.T., Stransky, C., Zimmermann, C., 2008. Stock identity of horse mackerel (Trachurus Trachurus) in the northeast Atlantic and Mediterranean Sea: integrating the results from different stock identification approaches. Fish. Res. 89, 196–209. http://dx.doi.org/10.1016/j.fishres.2007.09.022.

Alex Smith, M., Green, D.M., 2005. Dispersal and the metapopulation paradigm in amphibian ecology and conservation: are all amphibian populations metapopulations? Ecography 28, 110–128. http://dx.doi.org/10.1111/j.0906-7590.2005.04042.x.

Ames, E.P., Lichter, J., 2013. Gadids and Alewives: structure within complexity in the Gulf of Maine. Fish. Res. 141, 70–78. http://dx.doi.org/10.1016/j.fishres.2012.09.011.

Archambault, B., Le Pape, O., Baulier, L., Vermard, Y., Véron, M., Rivot, E., 2016. Adult-mediated connectivity affects inferences on population dynamics and stock assessment of nursery-dependent fish populations. Fish. Res. 181, 198–213. http://dx.doi.org/10.1016/j.fishres.2016.03.023.

Archambault, B., Rivot, E., Savina, M., Le Pape, O., 2015. Using a spatially structured life cycle model to assess the influence of multiple stressors on an exploited coastal-nursery-dependent population. Estuar. Coast. Shelf Sci. http://dx.doi.org/10.1016/j.ecss.2015.12.009.

Barrios, A., Ernande, B., Mahé, K., Trenkel, V., Rochet, M.-J., 2017. Utility of mixed effects models to inform the stock structure of whiting in the Northeast Atlantic Ocean. Fish. Res. 190, 132–139. http://dx.doi.org/10.1016/j.fishres.2017.02.005.

Begg, G.A., Waldman, J.R., 1999. An holistic approach to fish stock identification. Fish. Res. 43, 35–44. http://dx.doi.org/10.1016/S0165-7836(99)00065-X.

Brett, J.R., 1979. Environmental factors and growth. Fish Physiology. Elsevier, pp. 599–675. http://dx.doi.org/10.1016/S1546-5098(08)60033-3.

Brett, J.R., Shelbourn, J.E., Shoop, C.T., 1969. Growth rate and body composition of fingerling sockeye salmon, Oncorhynchus nerka, in relation to temperature and ration size. J. Fish. Res. Board. Can. 26, 2363–2394. http://dx.doi.org/10.1139/f69-230.

Brunel, T., Ernande, B., Mollet, F.M., Rijnsdorp, A.D., 2013. Estimating age at maturation and energy-based life-history traits from individual growth trajectories with non-linear mixed-effects models. Oecologia 172, 631–643. http://dx.doi.org/10.1007/s00442-012-2527-1.

Burt, G.J., Millner, R.S., 2008. Movements of sole in the southern North Sea and eastern English Channel from Tagging Studies (1955-2004) (No. 144). Cefas Lowestoft.

Cadrin, S.X., Kerr, L., Mariani, S. (Eds.), 2014. Stock Identification Methods: Applications in Fishery Science, 2nd ed. Academic Press, Amsterdam.

Cadrin, S.X., Secor, D.H., 2009. Accounting for spatial population structure in stock assessment: past, present, and future. In: Beamish, R.J., Rothschild, B.J. (Eds.), The Future of Fisheries Science in North America. Springer, Netherlands, Dordrecht, pp. 405–426. http://dx.doi.org/10.1007/978-1-4020-9210-7_22.

Carpentier, A., Martin, C., Vaz, S., 2009. Channel Habitat Atlas for Marine Resource Management, Final Report (CHARM Phase II). INTERREG 3a Programme, IFREMER. Boulogne-sur-mer, France.

Castillo-Jordán, C., Cubillos, L.A., Navarro, E., 2010. Inter-cohort growth rate changes of common sardine (Strangomera bentincki) and their relationship with environmental conditions off central southern Chile. Fish. Res. 105, 228–236. http://dx.doi.org/10.1016/j.fishres.2010.05.012.

Ciannelli, L., Fisher, J., Skern-Mauritzen, M., Hunsicker, M., Hidalgo, M., Frank, K., Bailey, K., 2013. Theory, consequences and evidence of eroding population spatial structure in harvested marine fishes: a review. Mar. Ecol. Prog. Ser. 480, 227–243. http://dx.doi.org/10.3354/meps10067.

Coggan, R.A., Dando, P.R., 1988. Movements of juvenile Dover sole, Solea solea (L.), in the Tamar Estuary, South-western England. J. Fish. Biol. 33, 177–184. http://dx.doi.org/10.1111/j.1095-8649.1988.tb05571.x.

Cowen, R.K., 2006. Scaling of connectivity in marine populations. Science 311, 522–527. http://dx.doi.org/10.1126/science.1122039.

De Veen, J.F., 1976. On changes in some biological parameters in the North Sea sole (Solea solea L.). J. Cons. 37, 60–90. http://dx.doi.org/10.1093/icesjms/37.1.60.

Deniel, C., 1990. Comparative study of growth of flatfishes on the west coast of Brittany. J. Fish. Biol. 37, 149–166. http://dx.doi.org/10.1111/j.1095-8649.1990.tb05936.x.

Dorel, D., Koutsikopoulos, C., Desaunay, Y., Marchand, J., 1991. Seasonal distribution of young sole (Solea solea (L.)) in the nursery ground of the Bay of Vilaine (Northern Bay of Biscay). Neth. J. Sea Res. 27, 297–306. http://dx.doi.org/10.1016/0077-7579(91)90032-V.

Edeline, E., Carlson, S.M., Stige, L.C., Winfield, I.J., Fletcher, J.M., James, J.B., Haugen, T.O., Vøllestad, L.A., Stenseth, N.C., 2007. Trait changes in a harvested population are driven by a dynamic tug-of-war between natural and harvest selection. Proc. Natl.

Acad. Sci. 104, 15799–15804. http://dx.doi.org/10.1073/pnas.0705908104.

Edeline, E., Le Rouzic, A., Winfield, I.J., Fletcher, J.M., James, J.B., Stenseth, N.C., Vollestad, L.A., 2009. Harvest-induced disruptive selection increases variance in fitness-related traits. Proc. R. Soc. B Biol. Sci. (276), 4163–4171. http://dx.doi.org/10.1098/rspb.2009.1106.

Enberg, K., Jørgensen, C., Dunlop, E.S., Varpe, Ø, Boukal, D.S., Baulier, L., Eliassen, S., Heino, M., 2012. Fishing-induced evolution of growth: concepts, mechanisms and the empirical evidence: Fishing-induced evolution of growth. Mar. Ecol. 33, 1–25. http://dx.doi.org/10.1111/j.1439-0485.2011.00460.x.

Erlandsson, J., Östman, Ö, Florin, A.-B., Pekcan-Hekim, Z., 2017. Spatial structure of body size of European flounder (Platichthys flesus L.) In the Baltic Sea. Fish. Res. (189), 1–9. http://dx.doi.org/10.1016/j.fishres.2017.01.001.

Frank, K.T., Brickman, D., 2000. Allee effects and compensatory population dynamics within a stock complex. Can. J. Fish. Aquat. Sci. 57, 513–517. http://dx.doi.org/10.1139/f00-024.

Frisk, M.G., Jordaan, A., Miller, T.J., 2014. Moving beyond the current paradigm in marine population connectivity: are adults the missing link? Fish Fish. 1 (5), 242–254. http://dx.doi.org/10.1111/faf.12014.

Fu, C., Fanning, L.P., 2004. Spatial Considerations in the Management of Atlantic Cod off Nova Scotia, Canada. North. Am. J. Fish. Manag. 24, 775–784. http://dx.doi.org/10.1577/M03-134.1.

Hanski, I., 1998. Metapopulation dynamics. Nature 396, 41. http://dx.doi.org/10.1038/23876.

ICES, 2017. WGNSSK. Report of the Working Group on Assessment of Demersal Stocks in the North Sea and Skagerrak (WGNSSK), 26 April - 5 May 2017, ICES Headquarters, Copenhagen. ICES CM 2017/ACOM:21: 1091pp.

ICES, 2016. Solea (Solea solea) in Division VIId (Eastern English Channel).

ICES, 2009. Manual for the Offshore Beam Trawl Surveys. Revision 1.2., Working Group on Beam Trawl Surveys.

Ihssen, P.E., Booke, H.E., Casselman, J.M., McGlade, J.M., Payne, N.R., Utter, F.M., 1981. Stock identification: materials and methods. Can. J. Fish. Aquat. Sci. 38, 1838–1855. http://dx.doi.org/10.1139/f81-230.

Jorgensen, H.B.H., Hansen, M.M., Bekkevold, D., Ruzzante, D.E., Loeschcke, V., 2005. Marine landscapes and population genetic structure of herring (Clupea harengus L.) in the Baltic Sea. Mol. Ecol. 14, 3219–3234. http://dx.doi.org/10.1111/j.1365-294X.2005.02658.x.

Kerr, L.A., Hintzen, N.T., Cadrin, S.X., Clausen, L.W., Dickey-Collas, M., Goethel, D.R., Hatfield, E.M.C., Kritzer, J.P., Nash, R.D.M., 2016. Lessons learned from practical approaches to reconcile mismatches between biological population structure and stock units of marine fish. ICES J. Mar. Sci. J. Cons. http://dx.doi.org/10.1093/icesjms/fsw188. fsw188.

Kotthaus, A., 1963. Tagging experiments with the North Sea sole (Solea solea) in 1959 and 1960. Spec. Publ. Int. Commn. NW. Atlan. Fish. 123–129.

Kritzer, J.P., Sale, P.F., 2004. Metapopulation ecology in the sea: from Levins' model to marine ecology and fisheries science. Fish. Fish. 5, 131–140. http://dx.doi.org/10.1111/j.1467-2979.2004.00131.x.

Law, R., 2000. Fishing, selection, and phenotypic evolution. ICES J. Mar. Sci. 57, 659–668. http://dx.doi.org/10.1006/jmsc.2000.0731.

Le Pape, O., Cognez, N., 2016. The range of juvenile movements of estuarine and coastal nursery dependent flatfishes: estimation from a meta-analytical approach. J. Sea Res. 107, 43–55. http://dx.doi.org/10.1016/j.seares.2015.06.001.

Lester, N.P., Shuter, B.J., Abrams, P.A., 2004. Interpreting the von Bertalanffy model of somatic growth in fishes: the cost of reproduction. Proc. R. Soc. B Biol. Sci 271, 1625–1631. http://dx.doi.org/10.1098/rspb.2004.2778.

Lorenzen, K., 2016. Toward a new paradigm for growth modeling in fisheries stock assessments: Embracing plasticity and its consequences. Fish. Res. 180, 4–22. http://dx.doi.org/10.1016/j.fishres.2016.01.006.

Lorenzen, K., Enberg, K., 2002. Density-dependent growth as a key mechanism in the regulation of fish populations: evidence from among-population comparisons. Proc. R. Soc. Lond. B Biol. Sci. 269, 49–54. http://dx.doi.org/10.1098/rspb.2001.1853.

Mahe, K., Delpech, J.-P., Carpentier, A., 2007. Synthèse bibliographique des principales espèces de Manche orientale et du golfe de Gascogne.

Mollet, F.M., Engelhard, G.H., Vainikka, A., Laugen, A.T., Rijnsdorp, A.D., Ernande, B., 2013. Spatial variation in growth, maturation schedules and reproductive investment of female sole Solea solea in the northeast Atlantic. J. Sea Res. 84, 109–121. http://dx.doi.org/10.1016/j.seares.2012.12.005.

Petitgas, P., Rijnsdorp, A.D., Dickey-Collas, M., Engelhard, G.H., Peck, M.A., Pinnegar, J.K., Drinkwater, K., Huret, M., Nash, R.D.M., 2013. Impacts of climate change on the complex life cycles of fish. Fish. Oceanogr. 22, 121–139. http://dx.doi.org/10.1111/fog.12010.

Petitgas, P., Secor, D.H., McQuinn, I., Huse, G., Lo, N., 2010. Stock collapses and their recovery: mechanisms that establish and maintain life-cycle closure in space and time. ICES J. Mar. Sci. (67), 1841–1848. http://dx.doi.org/10.1093/icesjms/fsq082.

Pinheiro, J.C., Bates, D.M., 2000. Mixed-Effects Models in S and S-PLUS. Springer, New York.

Pita, A., Casey, J., Hawkins, S.J., Villarreal, M.R., Gutiérrez, M.-J., Cabral, H., Carocci, F., Abaunza, P., Pascual, S., Presa, P., 2016. Conceptual and practical advances in fish stock delineation. Fish. Res. 173, 185–193. http://dx.doi.org/10.1016/j.fishres.2015.10.029.

Quince, C., Abrams, P.A., Shuter, B.J., Lester, N.P., 2008. Biphasic growth in fish I: theoretical foundations. J. Theor. Biol. 254, 197–206. http://dx.doi.org/10.1016/j.jtbi.2008.05.029.

Reiss, H., Hoarau, G., Dickey-Collas, M., Wolff, W.J., 2009. Genetic population structure of marine fish: mismatch between biological and fisheries management units. Fish Fish. 10, 361–395. http://dx.doi.org/10.1111/j.1467-2979.2008.00324.x.

Ricker, W.E., 1981. Changes in the average size and average age of pacific salmon. Can. J. Fish. Aquat. Sci. 38, 1636–1656. http://dx.doi.org/10.1139/f81-213.

Rijnsdorp, A.D., Van Beek, F.A., 1991. Changes in growth of plaice Pleuronectes platessa L. and sole Solea solea (L.) in the North Sea. Neth. J. Sea Res. 27, 441–457. http://dx.doi.org/10.1016/0077-7579(91)90045-3.

Rindorf, A., Wright, P.J., Jensen, H., Maar, M., 2016. Spatial differences in growth of lesser sandeel in the North Sea. J. Exp. Mar. Biol. Ecol. 479, 9–19. http://dx.doi.org/10.1016/j.jembe.2016.02.007.

Riou, P., 2001. Contribution respective de différentes nourriceries côtières aux populations adultes de sole et de plie: étude par couplage de modèles linéaires généralisés avec un système d'information géographique. Aquat. Living Resour. 14, 125–135. http://dx.doi.org/10.1016/S0990-7440(01)01110-X.

Rochette, S., Huret, M., Rivot, E., Le Pape, O., 2012. Coupling hydrodynamic and individual-based models to simulate long-term larval supply to coastal nursery areas: modelling larval supply to coastal nurseries. Fish. Oceanogr. 21, 229–242. http://dx.doi.org/10.1111/j.1365-2419.2012.00621.x.

Schnute, J., 1981. A versatile growth model with statistically stable parameters. Can. J. Fish. Aquat. Sci. 38, 1128–1140. http://dx.doi.org/10.1139/f81-153.

Secor, D.H., 2015. Migration Ecology of Marine Fishes. Johns Hopkins University Press, Baltimore.

Sequeira, V., Neves, A., Paiva, R.B., de Lima, J.P., Vieira, A.R., Gordo, L.S., 2012. Life history parameters as possible discriminators of bluemouth Helicolenus dactylopterus (Delaroche, 1809) populations in Portuguese waters. Fish. Res. (125–126), 69–76. http://dx.doi.org/10.1016/j.fishres.2012.02.009.

Sinclair, A.F., Swain, D.P., Hanson, J.M., 2002. Disentangling the effects of size-selective mortality, density, and temperature on length-at-age. Can. J. Fish. Aquat. Sci. 59, 372–382. http://dx.doi.org/10.1139/f02-014.

Swain, D.P., Sinclair, A.F., Mark Hanson, J., 2007. Evolutionary response to size-selective mortality in an exploited fish population. Proc. R. Soc. B Biol. Sci. 274, 1015–1022. http://dx.doi.org/10.1098/rspb.2006.0275.

Tuck, G.N., Possingham, H.P., 1994. Optimal harvesting strategies for a metapopulation. Bull. Math. Biol. 56, 107–127. http://dx.doi.org/10.1016/S0092-8240(05)80207-1.

Von Bertalanffy, L., 1938. A quantitative theory of organic growth (inquiries on growth laws II). Hum. Biol. 10, 181–213.

Waples, R.S., Gaggiotti, O., 2006. What is A population? An empirical evaluation of some genetic methods for identifying the number of gene pools and their degree of connectivity: what is a population? Mol. Ecol. 15, 1419–1439. http://dx.doi.org/10.1111/j.1365-294X.2006.02890.x.

Weatherley, A.H., 1990. Approaches to understanding Fish growth. Trans. Am. Fish. Soc. 119, 662–672. http://dx.doi.org/10.1577/1548-8659(1990)119<0662:ATUFG>2.3.CO;2.

Wheeler, A.C., 1978. Key to the Fishes of Northern Europe: a Guide to the Identification of More than 350 Species. F. Warne, London.

Ying, Y., Chen, Y., Lin, L., Gao, T., Quinn, T., 2011. Risks of ignoring fish population spatial structure in fisheries management. Can. J. Fish. Aquat. Sci. 68, 2101–2120. http://dx.doi.org/10.1139/f2011-116.

Zemeckis, D.R., Martins, D., Kerr, L.A., Cadrin, S.X., 2014. Stock identification of Atlantic cod (Gadus morhua) in US waters: an interdisciplinary approach. ICES J. Mar. Sci. 71, 1490–1506. http://dx.doi.org/10.1093/icesjms/fsu032.

Titre : Structure spatiale et connectivité au sein du stock de sole commune de Manche Est – Apport d’une approche holistique multitraceur.

Mots clés : *Solea solea* – Métapopulation – Croissance - Abondances – Otolithe – Génétique

Résumé : La sole commune (*Solea solea*) est un poisson plat nourricerie-dépendant ayant été surexploité en Manche Est (division VIId). L’une des causes de ce déclin pourrait être le décalage entre l’unité de gestion (stock) et l’unité fonctionnelle (population). L’objectif de cette thèse était de déterminer la structure spatiale du stock de sole commune de Manche Est en se focalisant sur les stades de vie pré-adultes et adultes et de statuer sur le potentiel décalage entre l’unité de stock et la population sous-jacente. Pour cela, une approche holistique multitraceur a été développée en combinant des approches populationnelles et individuelles.

D’abord, une étude de la croissance populationnelle et de la synchronie des abondances aux âges a mis en évidence l’existence d’un signal à long terme de structure spatiale du stock en trois sous-unités avec l’isolement probable de l’une d’entre elles. Ensuite, des analyses génétiques, de forme des otolithes et de microchimie des otolithes ont mis en évidence une structure spatiale en trois sous-unités. Enfin, un Indice semi-quantitatif de Différentiation du Stock a suggéré une forte structure spatiale en trois sous-unités. Ainsi, cette thèse a démontré une structure de type métapopulation de la sole commune en Manche Est qui devrait être prise en compte dans le processus d’évaluation et de gestion du stock afin d’atteindre une gestion durable.

Title : Spatial structure and connectivity within the Eastern English Channel stock of common sole – Contribution of a multitracer holistic approach

Keywords : *Solea solea* – Metapopulation – Growth – Abundances – Otolith – Genetics

Abstract: The common sole (*Solea solea*) is a nursery-dependent flatfish that has been overexploited in the Eastern English Channel stock (division VIId). An explanation of such decline could be the misalignment between the management unit (stock) and the biological unit (population). This thesis aimed at assessing the spatial structure of the Eastern English Channel stock of common sole by focusing on sub-adults and adults and determining the potential mismatch between the stock unit and the underlying population. To do so, a multitracer holistic approach was developed by combining population and individual-based approaches.

First, analyses of population growth and synchrony of abundances-at-age series revealed the existence of a long-lasting signal of spatial stock structure made of three subunits with potential isolation of one of these subunits. Then, genetics, otolith shape and otolith microchemistry analyses highlighted three subunits within the stock. Finally, a semi-quantitative Stock Differentiation Index suggested a strong spatial structure in three subunits. Therefore, this thesis evidenced a metapopulation structure of the common sole of the Eastern English Channel that should be integrated in the assessment – management process to provide a sustainable exploitation of the stock.