

The collection
Gustave Gilson
as a historical reference
framework for the
Belgian marine fauna:
feasibility study

SUSTAINABLE MANAGEMENT OF THE NORTH SEA 26107

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The collection of Gustave GILSON as a reference framework for the Belgian marine fauna: feasibility study

Royal Belgian Institute of Natural Sciences (RBINS)

Department of Invertebrates

# FINAL REPORT

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# SAMENVATTING

De Belgische mariene gebieden worden op dramatische en toenemende wijze beïnvloed door menselijke activiteiten. Vooraleer men overgaat tot bescherming van het mariene ecosyteem en een duurzaam beheer, moet de impact van de menselijke activiteiten worden bepaald. Daarvoor is informatie inzake de status van de mariene fauna in een zo natuurlijk mogelijke toestand essentieel. Zulke informatie kan worden afgeleid uit onderzoek van 'oorspronkelijke' gebieden of uit interpretatie van 'oude' gegevens. België beschikt over een unieke historische collectie van mariene monsters en gegevens, die het resultaat is van de omvangrijke bemonsteringen die professor Gustave Gilson, pionier in de mariene biologie, in het begin van de 20ste eeuw heeft uitgevoerd. Deze periode was gekenmerkt door een aanzienlijk lagere mate aan menselijke impact dan vandaag het geval is. De collectie Gilson wordt beheerd door het Koninklijk Belgisch Instituut voor Natuurwetenschappen (KBIN). Het doel van het project was de haalbaarheid te onderzoeken om deze collectie te gebruiken als 'referentie' voor de Belgische mariene fauna, ten einde lange termijn veranderingen en de invloed van de mens te kunnen vaststellen.

Het schema en de instrumenten inzake de bemonsteringen van Gilson werden in detail bestudeerd, waarbij vooral de nadruk lag op de 'bodemschaaf' ('dredge', benthos monsters) en de 'bodemverzamelaar' ('sondeur', sediment monsters). Beide instrumenten bleken betrouwbare gegevens op te leveren. Drie belangrijke onderzoeksprogramma's van Gilson werden in het huidige project behandeld.

Gilson heeft mariene monsters verzameld vanaf 1898 tot 1939, volgens een bijzonder intensief en systematisch schema in de periode 1899-1914. In totaal werden meer dan 14.000 monsters verzameld, als volgt verdeeld: ongeveer 3000 sedimentmonsters (hoofdzakelijk dmv de 'bodemverzamelaar'); 1500 waterstalen (verschillende flessen: Richard, Nansen en Petersen); 9500 fauna-monsters (dmv verschillende instrumenten: netten, trailers, bodemschaven, met de hand...). In de meeste gevallen werden plaatscoördinaten nauwkeurig bepaald. 841 monsters sedimenten zijn bewaard gebleven in het KBIN. Mariene biota werden oorspronkelijk in formol of alcohol bewaard en volgens de plaats van bemonstering. Sindsdien zijn de specimens geïdentificeerd, verdeeld en bewaard volgens soort (deelmonsters). Ook het oorspronkelijke etiket met het originele registernummer werd gekopieerd over de verschillende deelmonsters. Naar schatting twee derden van de Belgische mariene fauna-monsters in het KBIN behoren tot de collectie Gilson. Vele archieven en niet gepubliceerde documenten zijn bewaard gebleven, zoals logboeken, inventarissen, briefwisseling, informatie inzake bemonsteringinstrumenten, enz... Sommige documenten zijn niet teruggevonden.

We hebben ons in dit project beperkt tot een taxonomische revisie en een kwaliteitsonderzoek van de monsters en de bijhorende gegevens van de neogastropoden, bivalven en echinodermen. Alle beschikbare gegevens werden ingevoerd in een database die gedurende dit project werd ontwikkeld, de 'Southern North Sea Species Database' (SNNSD). De meeste monsters zijn goed bewaard gebleven en voorzien van de oorspronkelijke bemonsteringsgegevens. Bepaalde informatie zoals datum en tijd van de bemonsteringen, plaatscoördinaten, diepte, status van de getijden en substraatbeschrijving, zijn door Gilson op een systematische manier verzameld en genoteerd. Deze gegevens werden gedurende dit project ingebracht in een databank betreffende bemonsteringsplaatsen. Ook de sedimenten werden verwerkt in dit project. Van de oorspronkelijk 3000 sedimentmonsters zijn er 841 deelmonsters bewaard gebleven in het KBIN. Ze zijn voorzien van informatie inzake de bemonstering.

Vijf casestudy's werden uitgevoerd inzake mogelijke toepassingen van de collectie Gilson.

- 1. Onderzoek van 920 monsters neogastropoden (Mollusca, Gastropoda) van het KBIN, afkomstig van de collecties mbt de Belgische mariene fauna, waarvan de collectie Gilson een groot deel omvat. 12 van de 14 soorten vereisten een taxonomische revisie, 10 soortnamen moesten worden aangepast aan de huidige nomenclatuur, drie soorten waren bovendien onjuist geïdentificeerd. Van zeven soorten bestaat levend verzameld materiaal, de andere zeven zijn enkel vertegenwoordigd als lege schelpen, waarvan een deel (sub)fossiel. Historische verspreidingskaarten werden opgemaakt voor de levend verzamelde soorten (lege schelpen werden buiten beschouwing gelaten). De wulk, *Buccinum undatum*, was de meest voorkomende soort. Ondanks het feit dat de 10 zeemijlenzone veel intenser werd bemonsterd door Gilson, bleek deze soort opvallend meer voor te komen in het gebied van de Hinderbanken.
- 2. Het genus *Ensis* (Mollusca, Bivalvia) werd taxonomisch onderzocht. De taxonomie van sommige soorten van het genus is controversieel. Morfometrische metingen werden verricht op specimens van vijf soorten uit de zuidelijke Noordzee (*E. ensis, E. arcuatus, E. phaxoides, E. siliqua* en *E. minor*) en op de exoot *E. americanus*. Alle informatie inzake de soorten en specimens werd ingevoerd in de SNNSD. Een diepgaander studie is noodzakelijk om de complexe taxonomische problematiek binnen het genus *Ensis* te ontrafelen.
- 3. De haalbaarheid om de collectie Gilson te gebruiken voor genetische toepassingen werd onderzocht. Vooraleer de dieren van de KBIN-collecties werden bewaard in alcohol, werden ze gefixeerd in formol. Formol staat bekend om zijn beschadigende en degraderende werking op het DNA. Het project wou nagaan in hoeverre genetische studies konden worden uitgevoerd op historisch materiaal. DNA-extracties en amplicaties werden uitgevoerd op oude specimens van weekdieren. DNA-extracties en amplificaties bleken tot op heden niet succesvol voor *Littorina littorea* (alikruik of kreukel), daarentegen wel voor specimens van *Ensis* (mes– en zwaardscheden). Dit toont aan dat genetische toepassingen op 'oud' materiaal mogelijk zijn, maar dat een stap-voor-stap benadering voor specifieke genetische studies noodzakelijk is.
- 4. Een evaluatie van de bruikbaarheid van de monsters ten aanzien van eco-toxicologische toepassingen werd uitgevoerd. 20 sedimentmonsters werden geselecteerd voor een zware metalen analyse. De eerste resultaten in de 'totale' sediment fractie (< 2mm) wijzen niet op een abnormale hoeveelheid aan secundaire contaminanten. Cu (koper) niveaus waren zeer laag. Pb (lood) niveaus waren tamelijk hoog. Zn (zink) en Cd (cadium) niveaus waren onduidelijker te omschrijven, vanwege een hoger variatiepatroon. Alhoewel de studie nog niet werd vervolledigd, duiden deze analyses erop dat de sedimenten van G. Gilson tot interessante waarnemingen kunnen leiden, die bruikbaar kunnen zijn voor de studie van langetermijn-trends in de metaalniveaus van de Belgische mariene gebieden.
- 5. Een onderzoek werd verricht naar de historische habitatparameters (sedimentomschrijving en diepte) van benthische soorten. Alhoewel enkel 841 sedimentmonsters zijn bewaard gebleven, zijn er veel van de oorspronkelijk 3000 sedimentmonsters beschreven door Gilson. Het was de bedoeling om na te gaan of die beschrijvingen betrouwbaar waren, om ze te kunnen vertalen tot gestandaardiseerde granulometrische categorieën en om een gedetailleerde referentiekaart inzake sedimentologie op te maken. De onderzoekingen op granulometrische profielen tonen aan dat vier granulometrische hoofdcategorieën kunnen worden gehanteerd (slib, fijn zand, grof zand en grint). Verder werden deelcategorieën voorgesteld op basis van de aanwezigheid van schelpresten, hoeveelheid grint of slib. Ook gegevens over de diepte werden onderzocht en lijken betrouwbaar. Deze casestudy wijst aan dat historische habitatparameters kunnen worden gebruikt voor een lange termijn studie van de Belgische mariene gebieden.

Besluit: de collectie Gilson omvat monsters en archieven van een hoge wetenschappelijke en historische waarde. Een korte studie van de historische zoölogische collecties in de buurlanden van de Noordzee leert dat het onderzoek verricht door Gilson vrij uniek was in opzet, en bijgevolg ook in de resulterende gegevens en collectie. De informatie verzameld tijdens de bemonsteringen is gedetailleerd, waarbij vele parameters werden gemeten. Alhoewel een groot deel van het biologische materiaal intussen werd geïdentificeerd, zijn taxonomische revisies noodzakelijk. De ontwikkelde database zal worden geïncorporeerd in het 'Integrated and Dynamical Oceanographic Data management' (IDOD) wat

ongetwijfeld een belangrijke bijdrage zal zijn tot lange termijn onderzoek in de Belgische mariene gebieden, en mogelijk ook op ruimere schaal. De collectie Gilson is in het bijzonder aantrekkelijk om lange termijn veranderingen in habitats en fauna te bepalen, op voorwaarde dat het huidige marien onderzoek en de monitoringprogramma's vergelijkbare gegevens kunnen genereren (dus rekening houden met vergelijkbare bemonsteringsinstrumenten, intensiteit aan bemonstering, taxonomisch terrein). Kortom, de ontwikkeling van een historisch referentiepunt voor de biodiversiteit van de Belgische mariene gebieden, op basis van de verzameling Gilson, is haalbaar binnen goed gedefinieerde grenzen.

# **SUMMARY**

The Belgian marine areas (BMA) are dramatically and increasingly influenced by human activities. When ecosystem conservation and management practices are to be implemented, it is important to assess human impact on the ecosystem. For that purpose, information on the status of the marine fauna in absence of direct impact is essential. Such information can be derived from "pristine" areas or from observations on "old" material. Belgium holds a unique historical collection of marine samples thanks to the extensive sampling effort of Professor Gustave Gilson, a pioneer in marine ecology, at the very beginning of the 20<sup>th</sup> century, a period during which human pressure was considerably lower than today. This collection is held by the Royal Belgian Institute of Natural Sciences (RBINS). Our project aims at examining the feasibility of using this century-old collection as a "reference" for the Belgian marine fauna in order to investigate long term changes and human impacts.

Gilson's sampling scheme and instruments have been studied in detail, focusing on the dredge (benthos samples) and on the "ground-collector" (sediments samples) operated in the BMA. Both instruments seem to have provided reliable samples. Three main surveys are dealt with in the present project.

Gilson has collected marine samples between 1898 and 1939, with particular intensity and systematic scheme between 1899 and 1914. A total of more than 14,000 samples have been collected, distributed as follows: around 3,000 sediment samples (mainly using the "ground-collector"); around 1,500 water samples (various bottles: Richard, Nansen and Petersen); 9,500 fauna samples (using various gears: nets, trawls, dredges, hand-picking, etc.). In most cases, sampling coordinates were determined with a good precision. 841 sediment samples have been preserved in the RBINS. Marine biota were originally kept in formalin or alcohol, identified when possible and grouped by sampling station. Later on at the RBINS, the specimens were sorted out and classified by species in alcohol jars, often after taxonomic revision. These sub-samples were preserved with original sample numbers. In the RBINS, an estimated two third of the Belgian marine fauna samples belong to Gilson's material. Many archives and unpublished documents have also been preserved together with samples, such as log-books, inventories, letters, information on sampling gears, etc, although some documents could not be recovered yet.

We have focused our attention on some groups for "quality" assessment of samples and sampling data, *e.g.* neogastropods, razor clams and echinoderms. As a first step, a series of taxonomic checks and revisions were performed. All available data were entered in a database created during the project, the "Southern North Sea Species Database" (SNSSD). Most animals are well preserved and are labelled with the original sampling data. Some sampling information, such as date and time, coordinates, depth, tide status or bottom description, have been recorded in a systematic manner. These sampling data have been compiled in a separate databank of localities. The sediment collection was also assessed. Of the original 3,000 collected sediment samples, 841 sub-samples are preserved in the RBINS collections and are accompanied with sampling information.

Five case-studies were performed in order to assess possible exploitation of the collection.

1. A total of 920 available samples Neogastropods were recorded in the general collection of the Belgian marine fauna, most of which originated from the Gilson collection. Of 14 species, 12 required a taxonomic revision: 10 were subject to nomenclatural changes, three were incorrectly identified. Seven species were collected alive, while seven were represented by shells only, a part of which are of (sub)-fossil origin. The preservation of specimens is good. Historic distribution maps were drawn for alcohol preserved animals (empty shells were not considered). The common whelk *Buccinum undatum* was best represented. This species was surprisingly most abundant in the Hinders region, despite a more intense sampling effort in the coastal area.

- 2. The genus *Ensis* (Mollusca, Bivalvia) has been taxonomically investigated. Indeed, the taxonomy of some species of the genus is controversial. Morphometric measurements were performed on the specimens of five "species" from the southern North Sea (*E. ensis*, *E. arcuatus*, *E. phaxoides*, *E. siliqua*, *E. minor*) and an alien invasive species (*E. americanus*) in the collections of the RBINS. All species en specimens related information was entered in the SNSSD. A more profound systematic investigation should be performed on these taxa before they can be properly identified.
- 3. A feasibility study on genetic applications of the Gilson collection was performed. Until recently, animals collected for museum collections were usually first fixed in formalin before preservation in alcohol (ethanol). Unfortunately, formalin damages and degrades DNA. Therefore, our aim was to determine whether genetic studies could still be performed on this historic material. 18S ribosomal DNA extractions and amplifications were performed on old mollusc specimens. While the extractions failed for *Littorina littorea*, the tests were positive for the specimens of *Ensis*. A further attempt on mitochondrial DNA of *L. littorea* was positive. This demonstrates that DNA extractions and amplifications are feasible on Gilson's material. However, case-to-case feasibility investigations will be necessary for specific genetic studies.
- 4. An evaluation of the utility of Gilson's samples for eco-toxicological applications has been performed. 20 sediment samples were selected for trace metal analysis. The first results in the « total » sediment (< 2 mm fraction) indicate no abnormal result with reference to possible secondary contamination. Cu levels were very low. Pb levels were quite high. Zn and Cd levels were more unclear due to a higher variation pattern. Although incomplete, these analyses showed that the sediment of G. Gilson could provide interesting results for the study of long-term trends in the metal levels of the BMA.
- 5. A study has been performed on historic "habitat" (sediment nature and depth) for benthic species. Although only 841 sediment samples remain in the collection, many of the original 3,000 samples were qualitatively described by Gilson. Our aim was to determine whether this information is reliable, in order to translate it into standardized grain-size categories and to establish a detailed reference sedimentologic map. Our investigation on grain-size profiles shows that four main grain-size categories can be established (mud, fine sand, coarse sand, gravels). Sub-categories are further proposed using information on shell remains, gravel and mud presence. Depth data were investigated as well and seem reliable. This case-study indicates that these historical « habitat » parameters could be used in a long term investigation of the BMA.

In conclusion, the Gilson collection constitutes a historic material of high scientific value. A short investigation on historic zoological collections in neighbouring countries of the North Sea shows that Gilson's survey was quite unique in its design and, consequently, in the resulting collections. Sampling information is detailed, and many parameters have been recorded. A large part of the biological material has been identified by previous researchers although further taxonomic revisions are necessary for future research. The SNSSD database, once incorporated within the database on the quality of the marine environment (IDOD, Integrated Dynamical Oceanographic Data Management) will be a valuable tool for long term research in the BMA and even at larger scale. The Gilson collection is particularly attractive for assessing long term changes in habitats and fauna in the BMA, provided that current marine research and monitoring programmes yield comparable data (sampling gear, sampling effort, taxonomic coverage). The establishment of a century-old semi-quantitative "reference point" for the biodiversity of the BMA with the Gilson collection is feasible.

# 1. INTRODUCTION

# 1.1. STATE OF THE ART

The Belgian marine areas (BMA) have been under an increasing stress as a consequence of human activities. The southern North Sea is a densely populated area and has heavily frequented shipping routes. Fishery, sand and gravel extraction, dredging works, pollution and recreation are further major human impacts, especially to the numerous sand banks (COENJAERTS, 1997; MAES *et al.*, 2000). These human activities have induced long-term changes in the biodiversity of the area.

Within a policy of sustainable development, it is important to manage the natural resources in order to preserve it and, when necessary, to restore it. This implies that the original condition of the ecosystem can be evaluated. Recent publications (ARNOLD, 1991; ALLMON, 1994; VAN GOETHEM et al., 1995; Mc Carthy, 1998; Shaffer et al., 1998) have underlined the importance of old museum collections for biodiversity applications. A revision of historical data with modern research and statistical techniques can lead to new, often unexpected information, and bring these collections 'back to life' again.

The Royal Belgian Institute of Natural Sciences (RBINS) holds the historic collection of Gustave GILSON, a unique set of samples and detailed environmental information on the fauna and the habitats of the southern North Sea, collected between 1898 and 1939. Being aware of the potential value of this collection for biodiversity research, the Departments of 'Invertebrates' and 'Marine Ecosytem Management' raised the following questions: can a revision of the century-old GILSON collection provide new information on the fauna and habitats of the southern North Sea, at a period when human impact was lower? Secondly, could this information support to a policy of sustainable development?

Two years ago, the OSTC has granted the Department of Invertebrates to execute a project, a so called 'feasibility study', to evaluate the scientific value of the Gilson collection and its possible use as a reference framework for the Belgian marine fauna.

# 1.2. OBJECTIVES

Two main objectives were identified within this project:

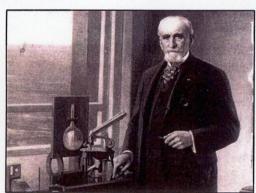
- a. An inventory of the collection in its present condition, with an evaluation of the samples of benthos and sediments and of the data on localities.
- b. An examination of the possible applications of the GILSON collection by five case studies with regard to:
  - Historical distribution maps
  - Taxonomic revision
  - Genetic applications
  - Contaminant analyses
  - Historical habitats mapping

Particular interest was given to four taxa: molluscs, crustaceans, echinoderms and fishes. In addition, particular attention was given to the time period 1899-1914, a period preceding severe anthropogenic stress in the North Sea.

# 2. THE GILSON COLLECTION

# 2.1. GUSTAVE GILSON

Gustave Gilson (1859-1944) was a biologist, a marine ecologist 'avant-la-lettre' and a pioneer in oceanography. In 1890 he was appointed as lecturer at the University of Leuven and became the



successor of Pierre-Joseph Van Beneden, who had build the very first marine laboratory worldwide, in 1842 in Oostende. In 1896, Gilson was engaged as a scientific collaborator at the Musée royal d'Histoire naturelle (the present RBINS), to conduct marine research in the southern North Sea. He was the representative of the Belgian government at the 'International Council for the Exploration of the Sea' (ICES) from 1903 to 1944. Gilson was the director of the RBINS from 1909 to 1925.

# 2.2. MARINE PROGRAMMES OF GILSON IN THE SOUTHERN NORTH SEA

The Gilson collection, comprising biota, sediment samples and archives, is the result of eight different types of explorations, subdivided in three marine programmes performed mainly between 1899 and 1914:

- > Explorations in the Belgian marine areas
- > Explorations in the southern North Sea
- > Explorations in the framework of the ICES

#### 2.2.1. EXPLORATIONS IN THE BELGIAN MARINE AREAS

A very systematic and intensive sampling was performed (GILSON, 1900). This programme consisted in the exploration of three areas (fig. 1):

- 1. A coastal area, from the French-Belgian border up to Walcheren (the Netherlands) and from the shore up to 10 nautical miles (n.m.) offshore, according to a grid based on minutes of longitude and latitude. Samples of benthic fauna were collected with a dredge (see also under 2.3.1.), towed over a distance of one n.m. (= one longitudinal minute). This area comprised more than 600 tows with the dredge. In addition, more than 2,000 sediments have been sampled with the cup-shaped 'ground collector' at every node of the grid and in between (see also under 2.3.2.). Besides this, GILSON has sampled the biota of the littoral zone.
- 2. At the West-, Oost- and Noord Hinder sandbanks (referred to as the 'Hinders'), Gilson performed sampling according 30 crosses, which were orientated towards the dominant SouthWest-NorthEast current. Biota and sediments were sampled along the arms of the crosses. His particular interest went to the fauna on and between the banks.
- 3. A transition area between the coastal area and the open sea was sampled by Gilson according to certain transects, such as from the lightship 'Wandelaar' to the Noord Hinder bank and from the 'Wandelaar' to the Thornton bank.

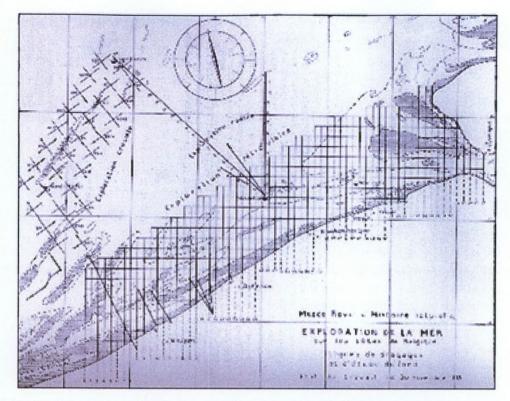


Figure 1: Explorations in the Belgian marine areas : the coastal area, the 'Hinders' area and the transition area

# 2.2.2. GENERAL EXPLORATIONS IN THE SOUTHERN NORTH SEA

Gilson also sampled numerous sites in the larger area of the southern North Sea (Fig. 2, red circled points).

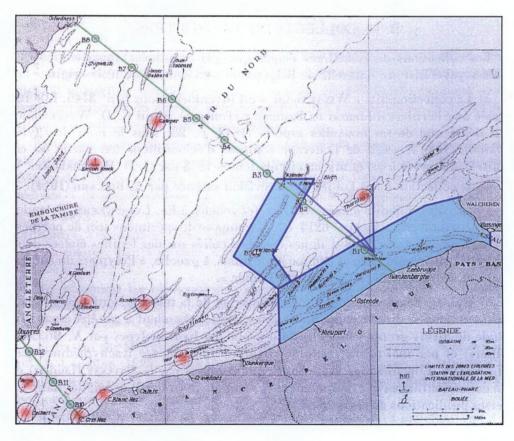


Figure 2. Overall view of the three marine programmes performed by Gilson (modified after Leloup, 1947)

# 2.2.3. EXPLORATIONS IN THE FRAMEWORK OF THE ICES

The third programme was done by Gilson in the framework co-ordinated by the ICES. Three-monthly sampling was performed along transects from Blankenberge to Oxfordnesss (B1-B8) and from Cap Gris-Nez to Dover (B10-B12) (fig. 2, green lines). This programme was chiefly carried out from 1903 to 1914 and included 44 sampling campaigns.

#### 2.3. SAMPLING INSTRUMENTS

# 2.3.1. DREDGES OF GILSON

For the study of benthic invertebrates, Gilson used several models of towed dredges. In various publications, Gilson insists on the fact that his dredges were tested long before the beginning of the "exploration of the sea". He used two major models: the "drague à anses" (fig. 3) and the "drague à large cadre". Both models are described in detail by GILSON (1900).

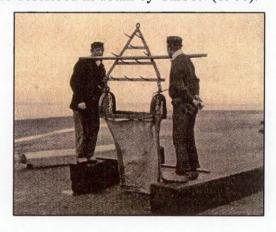


Figure 3: The "drague à anses" (source: GILSON, 1900)

The concept of these instruments was made in function of the bottom nature. In particular, soft bottoms were investigated with the "drague à anses", which ressembles an Agassiz trawl. The bottom fauna is collected in a bag made with sailing cloth. The concept of this dredge avoided, according to Gilson, the filling of the bag with sediment, although the instrument was towed on a standardized distance of one nautical mile (1,853 m). The field experience of Gilson in earlier years is confirmed by the fig. 4, which shows the instrument in operation. The dredge is equipped with a frontal rack (for soft bottoms) and is maintained on the bottom with lead weights. An ingenious system was built up in order to recover the dredge in case the cable was broken.

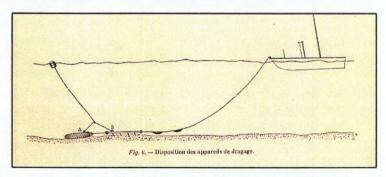


Figure 4: Illustration of the dredge with complete equipment (source: GILSON, 1900)

The instrument was basically designed for the collection of epibenthic fauna. The additional rack, equipped with vertical spines, allowed to further collect infauna of the surficial sediment.

In general, data obtained with dredges are nowadays considered as qualitative (absence-presence data) due to a low confidence in the gear efficiency. Although the dredge itself can be compared to an Agassiz trawl, we did not find an analysis of epibenthic data collected with a frontal rack as used by Gilson in the literature. When considering the Gilson collection within long-term studies on the biodiversity of the Belgian marine fauna, the use of such a particular instrument is of highest importance and should be taken into account.

In the « Explomer » databank (see 3.1), 1,140 samples are recorded as being collected with different models of dredges. Of these the instrument "drague n°5" is the most widely used (863 samples) and has been operated mainly in the exploration of the Belgian marine areas. The differences between models is not known so far. However, the model "n°5" being mostly used on soft bottoms, we can infer that it is a particular model of the "drague à anses".

# 2.3.2. THE GROUND COLLECTOR OF GILSON

The cup-shaped "ground collector" was invented by Gilson ("sondeur-collecteur à coupe") (GILSON, 1900, 1901, 1906; RICHARD, 1907; CARPINE, 1996). It consists of a large cup (roughly 20-60 cm in diameter), mounted on a central axis and closed with a mobile lid (figs. 5 - 6). In the very first model (years 1898-1899), the closing lid was a lead weight (see illustrations in GILSON, 1900). Shortly afterwards, the weight was definitely replaced by a forged steel plate.

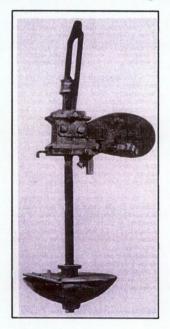




Figure 6: The ground-collector, over-filled with mud, is hauled onboard

Figure 5: Detail of the ground-collector. Source: CARPINE, 1996

10 models of 'ground collectors' ("Sondeur pyramidal", then "n°1" to "n°9") were used by G. Gilson. Only scattered information could be found in the archives on individual models.

It seems that the numbering of the model is linked to a particular cup shape and/or composition. Indeed, Gilson designed different cups for sampling within different substrate natures (sand, mud, clays, gravel, etc.; see Gilson, 1906). It seems that each model was fitted with a particular cup and was used in function of the substrate composition.

On the basis of available archives, the metal composition of the instrument was inferred for the different models. Model "n° 2" is in fact a different gear: Gilson intended to improve a coring instrument invented by Ekman; this model, made with a glass tube, was quickly broken and abandoned. The metal composition of model n° 3 could not be established. For model n° 4, Gilson

attempted to use copper, but this was also quickly abandoned due to the corrosion by sea water. All consecutive models are believed to be made of cast iron and forged steel.

When considering the sampling technique of the "ground-collector" (fig. 7), we can infer that the sampling depth could vary considerably in function of substrate hardness, from 20-30 cm in mud bottoms to perhaps 5 cm on hard (gravel or clay) bottoms. Gilson could clearly make a distinction between sediment layers in collected samples.

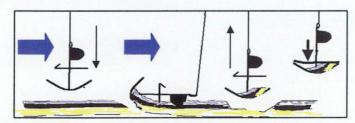


Figure 7: Operating the "ground-collector". While going downwards, the instrument is oriented by the current. When touching the floor, it lays on it's side allowing the cup to enter the sediment. Carefully hauled up, the instrument brings back some layers of surficial sediment. A messenger then allows the lid to close before hauling the sample onboard.

#### 2.3.3. OTHER METHODS

Aside from the ground collector and the dredges, many other sampling instruments have been used by Gilson during his explorations: various plankton nets operated near the bottom or in surface waters, various bottom trawls (mainly beam trawls), shrimp nets, various water bottles, static nets, hand-picking.

# 2.4. THE GILSON COLLECTION

Gilson has collected 14,035 samples from the southern North Sea during 18998-1939. These comprise roughly 3,000 sediment samples, 9,500 biota samples and 1,500 water samples. In addition *in situ* environmental parameters were measured, including substrate description, water temperature, depth, salinity, tide status, current- and wind direction, ...

The biota samples were originally stored per sampling site in the Maritime Institute of Oostende. Afterwards, these samples were sorted out to species level, and have since then been curated on a taxonomical basis among the other collections of the RBINS. This implies that an original Gilson's sample has been sorted out in species based 'subsamples'. The original label has been copied to the different 'subsamples'. When considering samples further on in this report and if not otherwise specified as original samples, samples will be mentioned in the sense of 'subsamples'.

The Gilson collection is held at the RBINS in seven depositories, managed by different sections and departments. Some original samples are still stored in formalin (some zooplankton samples), most samples have been kept in alcohol, dry or as microscopic slides.

Some 6,000 original samples of benthic biota such as molluscs (gastropods, bivalves, cephalopods), crustaceans (decapods, amphipods, isopods, mysidaceans, cumaceans), echinoderms (starfishes, brittle stars, sea urchins, sea cucumbers) and fishes (rays and numerous bonefish taxa) have been collected and are for the largest part identified. Among these samples, other benthic macroinvertebrate taxa such as polychaetes and other worm-like taxa identified as 'Vermes', bryozoans and cnidarians are also present. It remains unclear how many of these samples have been identified. An estimated 800 out of 3,700 zooplankton samples are still stored in their original state in formalin, others have been identified with their species name and are available as microscopic slides.

The water samples haven't been kept after their analysis.

From the original 3,000 sediment samples, 841 samples of 150 gram are still available at the RBINS.

Many researchers have used the material and/or data collected by Gilson for specific purposes; the resulting publications are listed in annex 1.

#### 2.4.1. ARCHIVES

The Gilson's surveys have generated large amounts of archived documents. These contain original log-books, maps, notes, publication manuscripts, etc.

A large part of this material was found in the archives of the Department of Invertebrates. Information on sediments was found in the Section Mineralogy / Petrography (Departement Marine Ecosystem Management). Further documents were found in the general archives and in few other sections of the RBINS.

All preserved documents are in a good quality status but often suffer paper ageing. It is anticipated that most of the available relevant scientific information has been identified. Missing documents, such as some log-books, may be archived somewhere else. We also know that the buildings of the Maritime Institute of Oostende have been destroyed during WW II: some documents may be definitively lost. A list of recovered archives has been compiled (Annex 2); it may still be further completed over time and should therefore not be considered as exhaustive.

# 2.4.2. QUALITY CONDITION OF THE COLLECTION

Biota and sediment samples have been preserved in good to excellent condition. No exceptional cases of desiccation have been observed for the alcohol samples. All samples are labeled with sampling information or with a register number, which by means of Gilson's log-books or cardindex system gives access to sampling information. However, information on the label of the sample is not always equivalent to the information in the archives. Case-study 1 has shown that about 1 % of the samples show locality data or sampling sites different from the information in the archives. 28 % of the Gilson neogastropod samples had descriptions of localities instead of geographic coordinates. However, many of these descriptions can be recalculated in geographic coordinates at different levels of precision.

Nomenclature and taxonomy are constantly subject to possible changes. Results from the case-study 1 show that the names of 12 of the 14 studied neogastropods had to be adapted to recent nomenclature (see also 4.1.3). Of these twelve, three species had to be re-identified.

Species based inventories have been made for the macrobenthic invertebrate taxa mentioned in 2.4, except for the polychaetes and the other wormlike-taxa. The oldest inventories date from 1939 and contain for each species a listing of all samples, including information of the sample labels. These 'old species lists' were translated in a more complete and handy card-index system for cumaceans, cirripedes, isopods and decapods in 1993. Half of the data and the samples of the bryozoans, some 200 records, have been digitalized into a database.

#### 3. DATABASES

Databases have been created in order to compile all relevant information obtained from the examination of the Gilson collection. A databank of « localities » has first been created in order to summarize sampling information and environmental parameters. Biological data were entered in a second database designed to comprise the complete information related to the species and the specimens.

The data gathered in the databases during this project will be entered within the Integrated Dynamical Oceanographic Database (IDOD), enhancing the added value of the collection for long-term studies.

#### 3.1. « EXPLOMER »

The EXPLOMER data bank is based on the exploration inventories and the card-index system developed by Gilson in order to summarize information from his sampling campaigns. For each sampling instrument, Gilson recorded similar information, even when two or three instruments were operated together (GILSON, 1900). Therefore, the cards theoretically contain information copied from different field log-books.

It has been decided to transpose the card-index system and the general directories into a Filemaker Pro data bank. Seven fields were filled: inventory type; sample number; type of exploration; IG number (RBINS collection identification number); "locality"; date; instrument.

The "locality" field comprises all information relative to the sampling point (coordinates, depths, etc.) and environmental parameters measured (temperature; salinity, ...).

Exactly 14,035 data were entered in the data bank.

Around 500 cards have been found in archives at the end of the project. Their content will be added to Explomer as well.

# Datasheet of archived sediments

No database was available for the Gilson sediments remaining in the RBINS collections. When these samples were examined for case-studies 4 and 5, it was decided to enter sample information from Explomer into a readily usable form for sample selection.

Firstly, old paper lists of Gilson sediments preserved in the RBINS collections were used to select data in the Explomer database. A total of 841 samples are archived and were identified with a specific field in "Explomer". 146 samples were taken with a dredge, three samples were collected with a bottom planktonic net, and one sample was collected with a glass jar on the beach. These 150 samples were considered not relevant and were not further investigated. The remaining 691 samples (77 %) were collected with the "ground-collector".

These 691 preserved samples were then copied to a separate MS-Excell sheet. The "locality" field from Explomer was cleared and subdivised in more specific fields (latitude; longitude; sediment description; depth; water temperature; tide status; water density; tide current), together with information on available weights. In this data set, data on coordinates, sediment descriptions and depths have been assessed for case-studies n° 4 and 5.

# 3.2. SOUTHERN NORTH SEA SPECIES DATABASE (SNSSD)

The Southern North Sea Species Database (SNSSD; Filemaker Pro 4.0.), was created to store data regarding the fauna of the southern North Sea. The database was initially focusing on data from the

faunistic-taxonomic revision of the neogastropods (see 4.1). However, it was designed in order to treat all Gilson samples, including information on the sampling localities, data from other southern North Sea collections in the RBINS, recent and future data.

This database will therefore constitute a tool for establishing a reference for the study of the long-term evolution of the Belgian marine fauna.

Although emphasis was given to data input in the table of samples and localities within this project, it is expected to complete the other tables in the future.

# 3.2.1. STRUCTURE OF THE SNSSD

The structure of the SNSSD is shown in fig. 8. There are six important tables in the database: samples, localities, maps of localities, taxonomy and systematics, distribution maps and related publications. There are further links with related websites.

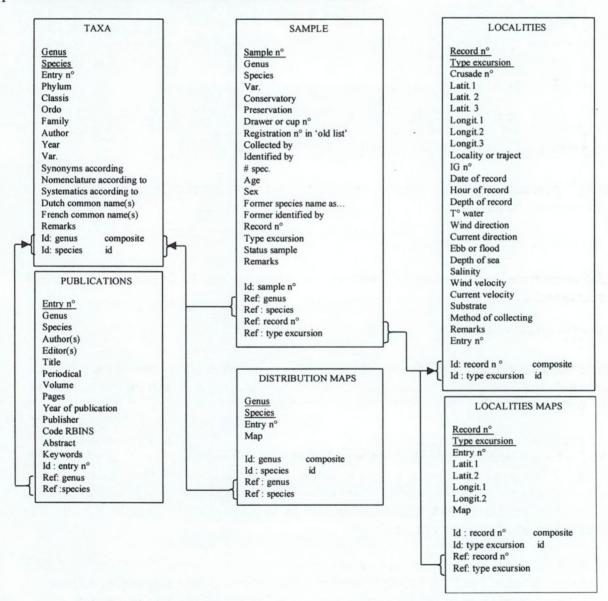


Figure 8: Scheme of the Southern North Sea Species Database (SNSSD)

The SNSSD contains the information described by the figs. 9-14, with an illustrative example of an alcohol sample of *Buccinum undatum* (a neogastropod mollusc). Details on the different fields of each table are explained in annex 3.

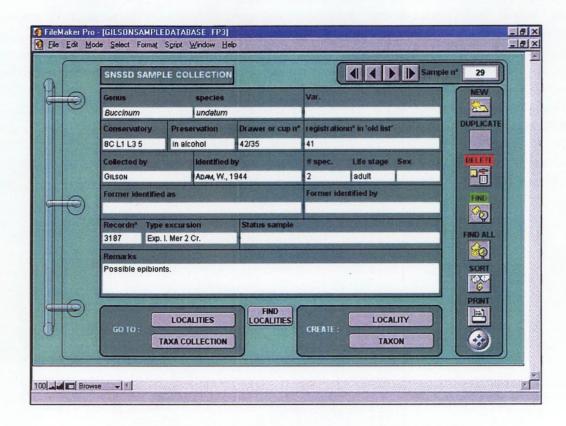


Figure 9: Table of samples: data on samples, including results of faunistic-taxonomic revision and quality check.

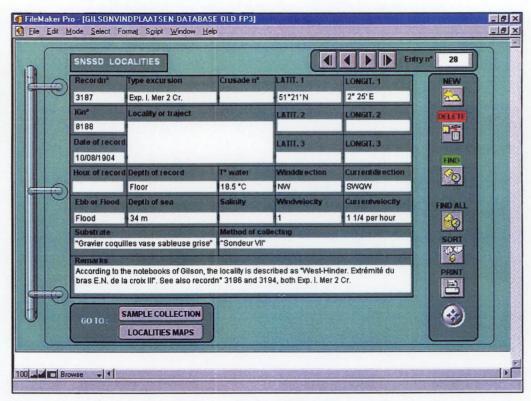


Figure 10: Table of localities: data on the sampling sites, including site description or geographic coordinates and in situ environmental parameters.

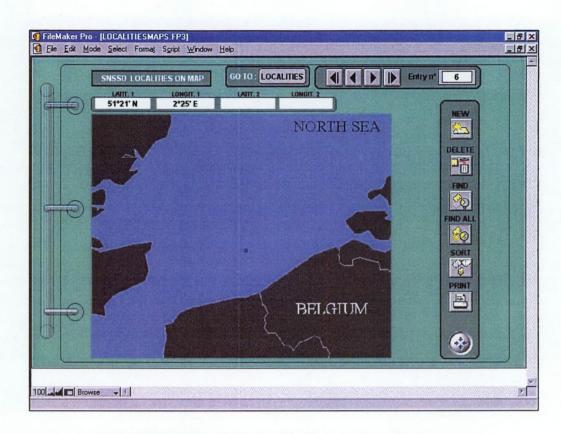


Figure 11: Table of of locality map: visualisation of the sampling site.

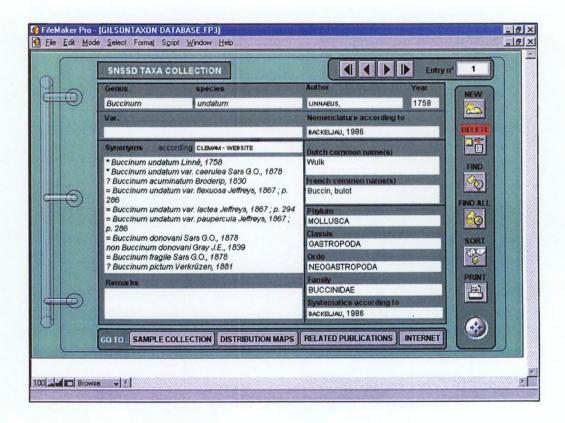


Figure 12: Table of systematics and taxonomy: all relevant data on taxonomy and systematics of each species, including synonyms and vernacular names.



Figure 13: Table of distribution maps: visualizing geographic distribution of species.

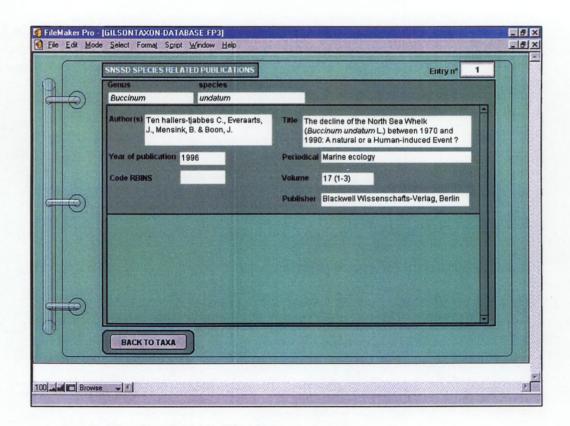


Figure 14: Table of species related publications.

# 3.2.2. PROTOCOL OF SAMPLE TREATMENT AND DATA INPUT

- Samples from a certain species were taken out of the depository.
- For each sample, the label and the specimen(s) were removed.
- Identification of the specimens was verified. The age class was evaluated (egg, juvenile or adult). Readily visible parasites, commensals or epibionts were looked for. Each gastropod shell was examined for biota, such as hermit crabs which are known to occupy empty shells. Furthermore, distinction was made between a 'fresh', subfossil or fossil gastropod shell. In case of re-identification, due to former false identification or synonym(s), former scientific name(s) and former 'identifier(s)' were recorded. Possible damage to specimen tissue, shells and level of alcohol in the jars were visually checked. Numbers of specimens mentioned on the label were verified. Data on the label of Gilson samples were compared with the explomer database, log-books and card-index system, identifying possible differences in date, sample site or recordn°.
- This examination was digitalized into the SNSSD in the two following tables:

# 1. Table of samples.

Comments, in particular on quality condition, were added when necessary. Additional data input into the table of samples enclosed the exact place of the sample in the depository, the way of preservation, drawer- or jar number and registration number in 'old list' (see also fig. 9).

#### 2. Table of localities.

The recordnumber and type of excursion, being the key, were copied automatically in the table of the localities. Additional information such as a cruise number, IGn° (inventory number which is given for each well described RBINS collection), date of record, locality (description) and/or geographic coordinates (latitude and longitude) were added. Furthermore, *in situ* data registered in the archives from Gilson were added too, such as time, depth of record, temperature from the surface water, wind direction and -velocity, water current direction and -velocity, tide status, depth of the sea, salinity, substrate description and method of collecting. Some relevant remarks on the locality of the sample, especially regarding possible differences with the archives, were highlighted (see also fig. 10).

- Aside these data input some additional treatment was planned and was performed in the scope of the feasibility study:
  - ➤ Some maps indicating each sampling site, trawl or dredge tow are available in the table of locality maps. Up to now 50 sampling sites maps are present in the SNSSD (see fig. 11).
  - > For every new species, a new datasheet was made in the table of taxonomy and systematics (see fig. 12).
  - A table of distribution maps was made (see fig. 13).
  - A table of species related publications was made (see fig. 14).

# 3.2.3. STATUS OF DATA INPUT IN THE SNSSD

Almost 2,200 samples have been treated according to the above protocol, whereby relevant data of the samples and results of the feasibility study have been registered in the different tables of the Southern North Sea Species Database (SNSSD). Data input involved several mollusc taxa, such as the neogastropods, the blue mussel *Mytilus edulis*, the razorclam genus *Ensis* and several echinoderm taxa such as starfish, brittle stars, sea urchins and sea cucumbers. Sample treatment and data input represent an intensive work, which for the neogastropods alone involved some 4,000 specimens ( see also 4.1.).

Some 1,400 samples originate from the Gilson collection; the remaining samples were taken from numerous smaller RBINS collections, such as DE MALZINNE, DUPUIS, GILTAY, LANSZWEERT, NYST, VERHAS...

17% of the fishes, crustaceans, molluscs and echinoderms from the Gilson collection (period 1899-1914) have been registered in the SNSSD.

# 4. POSSIBLE APPLICATIONS OF THE GILSON COLLECTION

The Gilson collection has a considerable potential for further scientific research and in particular as a historic reference framework for the Belgian marine fauna. Aside the detailed inventory of the collection, further « quality check » was performed by testing some scientific applications. Within this framework, five case-studies were performed.

- A first case-study examined the quality of the Gilson samples for the study of the historical distribution of benthic species in the Belgian marine areas. The study focused on neogastropod molluscs.
- A second case-study investigated the taxonomic study of complex species groups such as the genus *Ensis* (veneroid molluscs). Morphometric measurements were performed.
- > The feasibility of DNA extraction and amplification was investigated in case-study 3.
- > The quality of samples for trace contaminant analysis was evaluated, and a series of metal analyses in sediments were performed in case-study 4.
- Finally, an investigation was performed on the quality of sediments and depth data as parameters defining historical « habitats » for the benthic species.

# 4.1. CASE-STUDY 1: HISTORICAL DISTRIBUTION MAPS

#### 4.1.1. INTRODUCTION

Biodiversity, ecological and ecotoxicological related issues have stimulated a worldwide interest on molluscs during the last two decades. Aside other molluscan taxa, marine neogastropods (Mollusca: Caenogastropoda) have been the object of this increasing attention. Such is the case in countries near or bordering the North Sea such as France, Germany, the Netherlands and the U.K. It has been demonstrated that the dog-whelk Nucella lapillus (LINNAEUS, 1758) and the common whelk Buccinum undatum LINNAEUS, 1758 have declined in the North Sea (HUET et al., 1996 a,b; OEHLMANN et al., 1996; CADEE et al., 1995; TEN HALLERS-TJABBES et al., 1996 a,b; LAVALEYE, M & BERGMAN, M., 2000; MENSINK et al., 2000; BRYAN et al., 1986; GIBBS, P., 1993; KIDEYS, A., 1993; TUCK et al., 1998). Moreover, B. undatum has been listed in the status category 'Endangered' of the Red Lists of marine and coastal species and biotopes in the Wadden Sea area (PETERSEN et al., 1996). On the other hand, RUMOHR et al. (1998) state that B. undatum has been found more frequently in the North Sea in 1986 than in the period 1902-1912. The Belgian marine areas have been regularly sampled during the past two decades by the Laboratory of Marine Biology of the Ghent University, the Sea Fisheries Department (supervised by the Ministry of Small Enterprises, Traders and Agriculture) and by members of the «Strandwerkgroep», a Belgian society for marine biology. While the Ghent team sampled mainly macrobenthos from 'de Westkust' and 'de Vlaamse banken', the Sea Fisheries Department sampled different stations on edible target species in the Belgian marine areas in the framework of a monitoring programme. The « Strandwerkgroep » based their observations on all biota of the coastal zone and on stranded specimens. To get more insight in the different neogastropod species and their evolution in the Belgian marine areas, however, one needs to have reliable data at larger spatial and temporal scales.

In this contribution, a quality check on the Gilson samples was performed and the distribution of the neogastropods in the southern North Sea, in particular in the Belgian marine areas is presented and discussed.

# 4.1.2. MATERIAL AND METHODS

#### 4.1.2.1. COLLECTIONS OF THE RBINS

A total of 920 neogastropod samples, comprising more than 4,000 specimens from the southern North Sea, covering a sampling period from 1869 till 1978, have been revised according to the protocol described in 3.2.2.

The samples are stored as intact animals in alcohol (live samples) or as empty shells, which have a 'fresh' look or are subfossil or fossil. For some samples the species name had to be corrected according to the nomenclature used in BACKELJAU (1986) and in CLEMAM (= Check List of European Marine Mollusca: an up-to-date database edited by the Musée Nationale d'Histoire Naturelle, Paris; www.mnhn.fr). Identifications and information on the habitats and European distributions of the neogastropods were based on FRETTER & GRAHAM (1985), GRAHAM (1971), HAYWARD & RYLAND (1995), HOUART (1981), SEAWARD, R. (1990) and VAN REGTEREN ALTENA et al. (1956, 1957 & 1961).

Results have been registered in the Southern North Sea Species Database (SNSSD) (see also 3.2). Some two thirds of this material belongs to the Gilson collection, is mainly related to the Belgian marine areas and to the period 1899-1914.

#### 4.1.2.2. DISTRIBUTION DATA

Distribution maps have been made using the software Carto Fauna-Flora (Y. BARBIER AND P. RASMONT, 1995, *Cartography of biological data*, Université de Mons, Belgium). Species distribution maps have the following particularities:

Dots on the maps represent occurrences of live neogastropod specimens.

The dots, when not mentioned otherwise, are based on sites with geographic coordinates and on sites with descriptions which can be easily converted to coordinates, such as littoral sampling sites. Other sampling sites were discarded.

#### 4.1.3. RESULTS AND DISCUSSION

Table I gives an overview of all neogastropods from the RBINS collections found in the southern North Sea, with respect to the taxonomy of the species and way of preservation of the samples.

Species	Total samples	Live samples	Empty shell samples
Nucella lapillus (LINNAEUS, 1758)	126	33	93
Ocenebra erinacea (LINNAEUS, 1758)	117	6	111
Neptunea antiqua (LINNAEUS, 1758)	16	3	13
Buccinum undatum LINNAEUS, 1758	308	171	137
Nassarius incrassatus (STRÖM, 1768)	41	2	39
Nassarius reticulatus (LINNAEUS, 1758)	86	12	74
Oenopota turricula (MONTAGU, 1803)	97	3	94
Colus gracilis (DA COSTA, 1778)	13	0	13
Bellaspira rufa (MONTAGU, 1803)	42	0	42
Bellaspira septangularis (MONTAGU, 1803)	2	0	2
Boreotrophon clathratus (LINNAEUS, 1767)	9	0	9
Boreotrophon truncatus (STRÖM, 1768)	59	0	59
Trophonpsis muricatus (MONTAGU, 1803)	3	0	3
Nassarius pygmaeus (LAMARCK, 1822)	1	0	1

Table I: Taxonomy of the neogastropod species and way of preservation of the samples.

All species except *Buccinum undatum* and *Neptunea antiqua* had to be adapted to recent nomenclature. Samples of *Boreotrophon clathratus*, *Boreotrophon truncatus*, and *Trophonopsis muricatus* had to be re-identified.

All samples have been stored well. No dessication of the alcohol samples was detected. Samples were provided with a label or a registernumber, which refers mainly to data on the sampling site. An average of 72% of the labels on the Gilson samples have been supplied with geographic coordinates, 28 % were supplied with a spatial description of the sampling site. Most of the labels of the other RBINS collections were not provided with geographic coordinates, but with vaguely spatial descriptions of sampling sites.

Seven neogastropods are represented only by empty shells, mostly as subfossil or fossil specimens: Colus gracilis, Bellaspira rufa, Bellaspira septangularis, Boreotrophon clathratus, Boreotrophon truncatus, Trophonopsis muricatus and Nassarius pygmaeus. According to FRETTER & GRAHAM (1985) and HAYWARD & RYLAND (1995) these species are not or rarely found alive in the southern North Sea.

The neogastropod species sampled alive in the southern North Sea and in particular in the Belgian marine areas between 1899 and 1971 are: *Nucella lapillus*, *Ocenebra erinacea*, *Neptunea antiqua*, *Buccinum undatum*, *Nassarius incrassatus*, *Nassarius reticulatus* and *Oenopota turricula*. Distribution maps of live species are illustrated in fig. 15. Fig. 2 can be of aid to locate some of the sampling sites.

# Nucella lapillus

This species was dredged only once near the English Channel, not far from the Southeastern coast of the U.K. 32 records have been found on hard substrates such as stones, break waters and piers in the neighbourhood of the cities Oostende and Zeebrugge. Sampling was probably performed by hand-picking.

#### Ocenebra erinacea

One juvenile specimen was dredged near the 'Hinder' banks. Two records were found in the English Channel, while a fourth record was collected at the 'Spuikom' (Oostende).

#### Neptunea antiqua

Although according to HAYWARD & RYLAND (1995) widely distributed from Biscay to Arctic only one sample of three adult specimens was found near de 'Wandelaar'(B1), another adult specimen was found just outside the Belgian marine areas at 'de vlakte van Raan' (The Netherlands). Both records were surprisingly not sampled by the dredge, but with a Pettersen net towed in bottom waters.

#### Buccinum undatum

A total of 101 samples were collected, in the southern North Sea, mainly between 1899 and 1914, with only seven samples dating from after 1913. Occurrence was relatively very high near the 'Hinders'.

#### Nassarius incrassatus

One adult specimen was found at Cap-Griz-Nez (France), three adult specimens were collected near the lightship West-Hinder (at the 'Hinders').

# Nassarius reticulatus

One sample was collected in the Belgian coastal zone, eight samples were taken in the Sluice dock (Oostende), during a period 1954-1971 and one sample was found in the English Channel.

# Oenopota turricula

Two samples were collected in the Belgian coastal zone, a third was found in the Schelde estuarium near the Dutch shore.

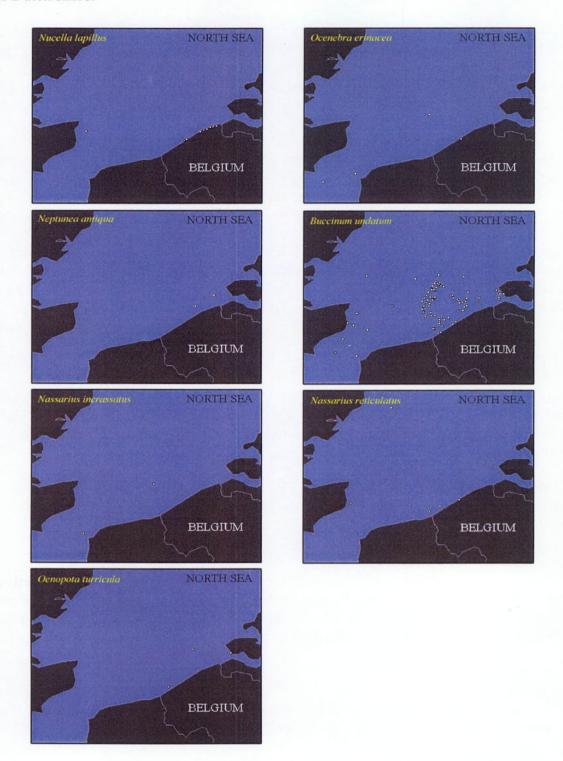


Figure 15: Distribution of live neogastropods in the southern North Sea, based on all RBINS collections and different instruments

According to the data from the RBINS collections, *Buccinum undatum* is the most common neogastropod in the Belgian marine areas during the period 1899-1971 (see Fig. 15). *Nucella lapillus*, *Ocenebra erinacea*, *Neptunea antiqua*, *Nassarius incrassatus*, *Nassarius reticulatus* and *Oenopota turricula* were rather rare.

Considering the collection of Gilson and the dredge n°5 only, B. undatum was found in 32% of all tows at the 'Hinders', whereas in the Belgian coastal area the species was gathered in only 4% of all

tows. This relatively higher occurrence of *B. undatum* in the 'Hinders' is even more remarkable because Gilson has towed the 'Hinders' three times less than the Belgian coastal area (see Fig. 16 and 17).



Figure 16: Total sampling sites: Gilson; dredge n°5. (Data with exact coordinates only)



Figure 17: Distribution of *B. undatum*; dredge n°5. (Data with exact coordinates only)

The 'Hinders' were already mentioned by GILSON (1900) to be of particular interest. In Leloup (1931, 1934), Wesenberg-Lund (1933) and Holly (1938), who all performed taxonomical and zoogeographical studies on the macrobenthos of the GILSON collection, distribution maps indicate the 'Hinders' as the area in the Belgian marine waters with relatively the highest occurrences of the taxa. This is confirmed by our results on the distribution of *Buccinum undatum*. Cattrijsse & Vincx (2001) who summarised scientific information on the benthos and avifauna of the Belgian marine areas since 1970, show a more diversified view. Their examination of 13 different studies on infaunal macrobenthos showed a decrease offshore by species number and by density, except for the study of Govaere (1977), who described an increasing average density, average number of species and species diversity towards the open sea. Furthermore, data on epifaunal benthos in Cattrijsse & Vincx (2001) tend to indicate that species number is increasing offshore and relatively is highest at the 'Hinders'.

- Considering the above findings, following two questions were raised:
  - To what extent did our distribution results represent the 'real' situation?
  - What are the distributions for the neogastropods in the Belgian marine areas today?

The answer to the first question is related to species ecology, to sampling intensity and especially to gear efficiency.

- With regard to the intensive sampling by Gilson, with regard to the efficiency of dredge n°5 (see also 2.3.1.) and with regard to the fact that *B. undatum*, *N. antiqua* and *O. turricula* are being described as living on the sandy soils of sublittoral areas (FRETTER & GRAHAM, 1985; HAYWARD & RYLAND, 1995; SEAWORD, 1990), we can expect that their distribution maps are giving a truly good representation. *B. undatum* can be seen as a relatively common species for the Belgian marine areas during 1899-1914,
  - while N. antiqua and O. turricula were consequently rare.

N. reticulatus, buried in sediments up to 15 cm deep (WEINBERG, 1999), was possibly under-recorded by Gilson.

- N. lapillus, O. erinacea and N. incrassatus live on at rocky shores, under stones or in crevices (HAYWARD & RYLAND, 1995). Such biotopes are not typical for the Belgian marine areas, which might explain the relative low number of records for these three species.
  - The second question can hardly be answered, since recent (published) data are scarce and scattered. At the RBINS most recent data for live neogastropod specimens are from 1971. More recently, live specimens of *B. undatum* have been collected with a 8 m beam trawl at some stations in the Belgian

marine areas by the Sea Fisheries Department monitoring programme (supervised by the Ministry of Small Enterprises, Traders and Agriculture) (HILLEWAERT, pers. comm., 2001). Members of the « Strandwerkgroep », have observed *B. undatum* alive at their regular strand visits during the period 1981-2000 (KERCKHOF, 1981; ENEMAN & KERCKHOF, 1983; ENEMAN, 1984; KERCKHOF, 1986; VANDERPERREN, 1991; VANHAELEN, 1998, VANHAELEN, 1999).

According to KERCKHOF (1988), *Nucella lapillus* has been declining rapidly at the Belgian coast since 1979 and has not be seen anymore (KERCKHOF, pers. comm., 2001.) An ongoing survey (VOLCKAERT, pers. comm., 2002) tends to indicate similar conclusions.

Live specimens of *N. reticulatus* have stranded frequently at the Belgian coast in recent years (Vanderperren, 1991; Vanhaelen, 1992, 1994, 1996a, b; Severijns, 1993, 1996; Vanwalleghem, 1994; Jonckheere, 1993; Jacobs, 1995a, b; Kerckhof, 1998). In addition, *N. reticulatus* has been sampled with a van Veen grab by the laboratory of marine biology of the Ghent University at different localities in the Belgian marine areas, in particular at the West Coast and the Flemish Banks (Degraer & Van Hoey, unpubl. data, 2001).

A debate exists on *Nassarius nitida* (Jeffreys, 1867), found alive in 'de Spuikom' (Oostende). According to Fretter & Graham (1985) *N. nitida* should be considered as a variety of *N. reticulatus*, whilst Rolan AND LUQUE (1994) state that these are two valid species.

#### 4.1.4. CONCLUSIONS

Samples of the Gilson collection are preserved very well.

Detailed and reliable information on the sampling stations is available.

Historical distribution maps make a new and interesting contribution regarding the biodiversity of the macrobenthos in the Belgian marine areas. Results indicate the importance of the 'Hinder' banks as an area with a potential high biodiversity.

The case-study shows also the importance of museum collections for long-term changes regarding biodiversity.

# 4.2. CASE-STUDY 2: MORPHOMETRIC AND TAXONOMIC STUDY

#### 4.2.1. INTRODUCTION

The systematics of the razor clam genus *Ensis* is only based on shell characteristics. Because these characteristics are very variable and because morphological differences between some of the species of *Ensis* are not always very clear, this has led to confusion and misidentifications within the genus *Ensis*.

The Gilson collection contains many specimens belonging to the genus *Ensis* and can add more insight in clarifying possible changes in shell characteristics and related taxonomical problems.

#### 4.2.2. MATERIAL AND METHODS

Collections of the RBINS. The genus *Ensis* from the Gilson collection and from other Belgian collections in the RBINS has been revised and verified. The indigenous 'species' from the southern North Sea, *E. ensis*, *E. arcuatus*, *E. siliqua*, *E. minor* and the alien invasive species *E. americanus* were investigated. The results of this taxonomic revision have been entered in the Southern North Sea Species Database (SNNSD) (see 3.2.).

The collections of the Naturalis Museum (Leiden, The Netherlands). The collections of *Ensis*, including the holotype and paratypes of *E. minor var. subarcuata*, were examined in order to get more insight on the different morphometric characteristics. Identifications of the specimens at Naturalis are based on shell characteristics and morphometric descriptions made by VAN URK (1964 a,b, 1987).

Morphometric data. Twenty five different shell characteristics, comprising length and height of the shell, length/height ratio, position of adductor and retractor (muscles)... were measured with a calliper. Only adult specimens were selected.

#### 4.2.3. RESULTS AND DISCUSSION

Our study of the collection in Naturalis has not given the necessary information to help recognize and identify each specimen of Ensis in the RBINS collections. Doubts remain on the status of  $E. \ phaxoides$  and on the  $E. \ siliqua/minor$  complex.

Table II gives an overview of all *Ensis*-species in the RBINS collections, with respect to taxonomy and number of specimens which have been morphometrically examined:

Ensis ensis (LINNAEUS, 1758)	25
E. siliqua/minor complex	145
E. americanus (CONRAD, 1843)	25
E. arcuatus (JEFFREYS, 1865)	54
Total number specimens	259

Table II: numbers of specimens for each considered "species".

Although ressembling *E. ensis*, *E. phaxoides* was described by VAN URK (1964 b) as a distinct 'species'. BACKELJAU (1986) refers also to *E. phaxoides* as a sixth possible Belgian 'species'. However, we were not able to identify or distinguish *E. phaxoides* in the RBINS collections. Only *E. ensis* could be detected. According to DE BRUYNE, GITTENBERGER and GOUD (pers. comm., 2000), scientific collaborators at Naturalis, *E. phaxoides* has to be seen as an ecological form of *E. ensis*. All ressembling samples have been treated as *E. ensis* and have been digitalized in that sense in the SNSSD. Futhermore, our examination of the RBINS collections, based on the characteristics described by VAN URK (1964 b), shows no straightforward distinction between *E. minor* and *E. siliqua*. Even not by using

the recent table of MOERDIJK (2000), who mentions some additional characteristics.

21

Specimen related information of 218 *Ensis* samples has been digitalized in the SNSSD, according to the procedure described in 3.2.2.

# 4.2.4. CONCLUSIONS:

The distinction between *E. ensis* and *E. phaxoides* on the one hand, and *E. siliqua* and *E. minor* on the other hand, are not straightforward.

A more profound investigation should be performed on these taxa. Morphometric data have been processed for further statistical treatment.

# 4.3. CASE-STUDY 3: GENETIC APPLICATIONS

## 4.3.1. FRAMEWORK OF THE CASE-STUDY

Biological diversity can be studied at various levels of organisation. Genetic investigations allow to investigate the natural variations occurring at the population level.

The collection of G. Gilson provides a possibility to investigate the genetic material of century-old specimens of the fauna from the southern North Sea. However, in most Museums old biota samples have generally been fixed with formalin, although formalin is often replaced with ethanol for storage. Unfortunately, formalin is known to induce severe DNA degradation and mutations. The possibility of DNA studies is thus questionable in old samples.

Our aim is to determine whether the material of Gilson could be suitable for DNA extraction.

#### 4.3.2. MATERIAL AND METHODS

Four specimens of molluscs, including two bivalves and two gastropods, were selected. These samples are currently stored in alcohol (table III), but the original fixative solution is not known.

Species	Sample n°	Origin and date	Analysed part	
Ensis arcuatus	(Exp. I. Mer. 7 Nect,) 653	Oostende-Nieuwpoort,	Muscular tissue	
		24/05/1913	18S Ribosomal	
Ensis siliqua	-	Oostduinkerke,	Muscular tissue	
		02/07/1938	18S Ribosomal	
Littorina littorea	(Exp. I. Mer. 9. Litt)	Walcheren,	Foot muscle	
	44	28/09/1904	18S Ribosomal	
Littorina littorea	(Exp. I. Mer. 9. Litt)	Walcheren,	Foot muscle	
	44	28/09/1904	Cyt. Ox., mit. DNA1	
Littorina littorea	(Exp. I. Mer. 9. Litt)	Zeebrugge,	Foot muscle	
	217	17/03/1911	18S Ribosomal	

Table III: sample selected for DNA extraction and amplification.

DNA extraction and PCR amplification were performed with the CTAB technique described in Winnepenninckx *et al.* (1993) on 18S ribosomal DNA. A further extraction of the cytochrome oxidase (subunit 1) sequence of the mitochondrial DNA was performed on *L. littorea*, sample n° 44.

#### 4.3.3. RESULTS AND DISCUSSION

18 S DNA extraction was successful for two specimens of *Ensis* but failed for *Littorina littorea*. The extracted fragments of 18s ribosomal DNA (500 BP) were amplified and sequenced. The additionnal extraction of mitochondrial DNA (550 BP) in a specimen of *L. littorea* was successfull.

The test failure in *L. littorea* could be due to the closure of operculum during the fixation process with formalin, resulting in a poor replacement rate inside the shell when ethanol rinses are performed. However, the original fixative is not known.

#### 4.3.4. CONCLUSIONS

This limited study clearly shows that DNA extraction is feasible in samples of the Gilson collection. Genetic applications on this old material can be considered but will require a case-to-case investigation.

# 4.4. CASE-STUDY N°4: CONTAMINANT ANALYSIS

#### 4.4.1. FRAMEWORK OF THE CASE-STUDY

The southern bight of the North Sea is heavily polluted due to the high industrial activity, demographic pressure and intense maritime traffic occurring in this area.

Intergovernmental organisations, such as UNEP<sup>1</sup> or OSPAR<sup>2</sup>, are concerned with these problems and set recommendations to their parties towards a significant reduction of the amounts of pollutants reaching the seas. It is thus of prime importance to be able to identify or estimate the "baseline" levels of organic and inorganic contaminants in the environment (OSPAR, 2000).

Within this framework, the sediments, animals and shells collected by G. Gilson in the early twentieth century are potential samples for the determination of historic "reference" levels of contaminants in the Belgian Continental Shelf (BCS). It has therefore been decided to investigate this particular application with a limited contaminant analysis on selected Gilson's samples.

#### 4.4.2. SELECTION OF SAMPLES AND CONTAMINANTS

For contaminant analysis, the samples of Gilson can be classified into three distinct groups: alcohol preserved animals, dry shells and sediments.

For each type of sample, sampling information is generally detailed. Each group has been briefly evaluated for sample selection.

#### 4.4.2.1. ALCOHOL PRESERVED ANIMALS

Contaminant analysis in fluid preserved animals has been proposed with enthusiasm by some authors in the past, mainly for trace metal analyses (e.g.: ZITCO et al., 1971; MILLER et al., 1972; BARBER and CROSS, 1972). Many of these results are questionable. GIBBS et al. (1972) and more recently RENAUD et al. (1995) indicate that metal concentrations obtained for tissues are doubtful due to risks of contamination by formalin and alcohol (low purity), jars, tags, pencil notes, etc. Furthermore, the specimen processing is expected to allow a leaching of contaminants: after a tissue fixation with formalin, samples are rinsed several times with alcohol as preservative fluid. Finally, we know that the specimens collected by Gilson at a sampling site were firstly preserved together in one jar. Specimens were sorted and classified by species later on. Cross contaminations are thus also expected.

These considerations led us to the conclusion that trace contaminant analysis in alcohol preserved samples will be unreliable.

However, one application can be considered: for animals with internal skelettons (such as echinoderms), calcified pieces could be of interest provided that their structure has not been altered during preservation. A microscopic investigation on calcified parts would answer this question.

# 4.4.2.2. DRY SHELLS

Empty shells of molluscs collected by Gilson and others have been stored dry in covered boxes and/or plastic bags. Many specimens have been manipulated after their collection.

A large part of the preserved shells is of sub-fossil to fossil origin. Traces of organic remains, indicative of a live catch, are present in only few specimens.

<sup>&</sup>lt;sup>1</sup> United Nations Environmental Programme

<sup>&</sup>lt;sup>2</sup> Oslo and Paris Convention

As is the case for most samples, the material from Gilson's surveys is accompanied with detailed sampling information. Analyses in dry shells would lead to results in a field were data are still scarce. These samples were therefore not considered for this case-study. However, such analysis should be feasible.

#### **4.4.2.3. SEDIMENTS**

Sediment samples appear to be the most attractive samples for the determination of "historical" contaminant levels. We have therefore investigated the sampling and storing conditions as well as the quality condition of preserved samples.

# 4.4.2.3.1. Sampling and storing

- Most samples were collected with the "ground-collector", although a part (18 %) of the preserved sediments were taken with a dredge.
- The metal composition of the "ground-collectors" could partly be inferred from scattered archived information (see 2.3.2). In general, it seems that around 1 Kg of collected sediment was initially stored at the "Oostende Maritime Institute".
- No more information is available for the period 1900-1956. In the latter year, 841 samples were transferred from Oostende to the RBINS. For that purpose, sample size has been reduced to 150 g in so-called "alkathene" (polyethylene) tubes.
- The record numbers of every preserved sample have been kept. The samples have seemingly not been manipulated since that time.

#### 4.4.2.3.2. Examination of samples

- The samples are preserved in wooden drawers closed with a glass lid, classified in function of Gilson's surveys.
- The alkathene tubes are not *sealed*, but are closed by a single alkathene cap. Consequently, there must have been exchanges with the atmosphere throughout the conservation, at least after 1956 (so far, no information was found regarding original storing conditions). However, the double enclosure of samples (closed alkathene jar + closed wooden box) has probably limited air exchanges; furthermore, samples have most probably not been manipulated since.
  - Although GILSON (1900) describes that the collected sediments are generally fixed with formalin, a brief visual investigation showed that most samples are in fact free of formalin. A total of 32 samples were opened for analytical purposes in the present project (20 metal analyses, 12 grain-size analyses). Of these, only one showed evidence of the presence of formalin (aggregation of sand, odour).

#### Selection of contaminants

The sediment samples have not been isolated from the ambient atmosphere, although air circulation was probably limited. This means that semi-volatile organic contaminants (such as PAH or dioxins) probably exhibit patterns of congeners for which interpretation will be difficult. Secondary contamination can of course be expected. Losses of contaminants may be feared (volatilisation of some congeners within a family). Furthermore, matrix ageing as well as bacterial activity have probably, over decades, influenced the relative abundance of the extractable compounds in the samples.

Metals are expected to be less labile contaminants, since secondary contamination of samples will be the main problem. Metal analysis in sediment samples was therefore retained for this case-study.

To date, only one sample of Gilson ("s135") has been analysed as a long-term "reference" for an investigation on trace metal levels (Cd, Cu, Pb, Zn, Fe, Hg, As, Cr, Ni, Al) in the BCS (Strubbe and Pichot, 1993). The levels mentioned are in general quite high.

# 4.4.3. TRACE METAL ANALYSES IN SEDIMENTS

#### 4.4.3.1. METHODS

# 4.4.3.1.1. Sample selection

Secondary contamination with metals can be expected from sampling operations (ground-collector composition) and from sample storing conditions. As a rule, it is generally acknowledged that high metal levels in sediments are mainly associated with a high proportion of fine fractions (clay and mud), although the correlation is not always obvious. These are monitored by analysing the Al content (indicative of the clay content) and the metal content of the fraction  $< 63 \mu m$  (mud).

Therefore, our hypothesis is that a secondary contamination could be detected when high levels are observed without a correlation to the clay or mud content. A very high variability of metal levels in samples is also expected if secondary contamination occurs.

We have selected 20 samples (table IV). Various combinations of instruments, sampling dates and sampling sites were made. The samples were further selected with respect to their vicinity to current monitoring stations in order to allow long-term comparisons (see fig. 18).

# 4.4.3.1.2. Trace metal analysis

Samples were processed in the Department of Sea Fisheries (Oostende) for sample preparation (isolation of fine fractions) and in the CODA Laboratory (Tervuren) for metal analyses (Al, Pb, Cu, Cd, Zn). Approximately 2 g of sediment were sieved on a 2mm sieve. 0.5 g of the < 2 mm fraction was dissolved in a mixture of nitric acid, perchloric acid and fluorhydric acid for a total decomposition. For Cd, Cu and Pb, analyses were performed with Induced Coupled Plasma -Mass Spectrometry (ICP-MS). Al and Zn levels were measured with ICP-Atomic Emission Spectrometry (ICP-AES). Concentrations are corrected with the reagent blank. Two certified reference materials ("BCSS-1", "MESS-1"; National Research Council Canada) were analysed with samples.

<63  $\mu$ m fractions were isolated (wet sieving) on approximately 40 g sample. These were isolated on samples 316, 317, 1311, 2414, 2586, 2081, 2090, 2258, 2706, 2582 and 2986.

# 4.4.3.2. RESULTS AND DISCUSSION

The metal analysis in the fine fraction could not be completed yet. For this reason, our results of "historical levels" are incomplete and should be examined with caution for long-term considerations. Furthermore, the number of samples in the selected areas (near current monitoring stations) is limited. However, some interesting trends can already be observed.

Gilson's samples					N	Monitoring stations	
N°	Position	Date	Description	Instrument	Name	Position	
3899	51°28'30N - 2°28'E, Croix XXIX	13/6/1906	Sable assez grossier coquillier	Sondeur VIII.	421	51,2883 - 2,2700	
3903	51°28'30N - 2°28'E, Croix XXIX	13/6/1906	Sable grossier coquillier	Sondeur VIII.	421	51,2883 - 2,2700	
3908	51°28'30N - 2°28'E, Croix XXIX	13/6/1906	Sable grossier coquillier	Sondeur VIII.	421	51,2883 - 2,2700	
2467	51°25'30N - 3°32'E	23/9/1902	Sable grossier coquillier	Sondeur VI.	SO1	51,2500 - 3,3420	
2468	51°25'30N - 3°32'E	23/9/1902	Sable grossier coquillier vase grise	Sondeur VI.	SO1	51,2500 - 3,3420	
1690	51°24'N - 3°13'E	21/9/1901	Sable grossier néritique, vase grise	Sondeur V.	700	51,2260 - 3,1320	
2083	51°24'N - 3°13'E	30/7/1902	Sable grossier coquillier	Sondeur VI.	700	51,2260 - 3,1320	
2081	51°26'N - 3°12'E	30/7/1902	Sable fin	Sondeur VI.	B03	51,2600 - 3,1230	
2258	51°26'N - 3°12'E	21/8/1902	Sable fin coquillier	Sondeur VI.	B03	51,2600 - 3,1230	
2090	51°26'30N - 3°13'E	30/7/1902	Sable fin	Sondeur VI.	B03	51,2600 - 3,1230	
637	51°11'N - 2°40'E	24/8/1900	Sable très coquillier	Sondeur I.	120	51,1110 - 2,4207	
651	51°11'30N - 2°42'E	25/8/1900	Sable un peu coquillier	Sondeur I.	120	51,1110 - 2,4207	
316	51°17'N - 2°51'E	10/8/1899	Sable avec coquilles pulvérisées	Sondeur I.	230	51,1850 - 2,5100	
317	51°17'30N - 2°51'E	10/8/1899	Sable pur	Sondeur I.	230	51,1850 - 2,5100	
1311	51°17'N - 2°51'E	02/8/1901	Sable grossier, très néritique	Sondeur III.	230	51,1850 - 2,5100	
2414	51°18'30N - 2°53'E	15/9/1902	Sable fin vaseux	Sondeur VI.	230	51,1850 - 2,5100	
2586	51°18'N - 2°53'E	11/5/1903	Sable fin vase grise et noire	Sondeur VII.	230	51,1850 - 2,5100	
2706	51°26'N - 3°8'E	05/8/1903	Sable fin coquillier	Sondeur VI.	710	51,2600 - 3,0800	
2582	51°26'N - 3°9'E	05/5/1903	Sable fin coquillier vase grise	Sondeur VII.	710	51,2600 - 3,0800	
2986	51°26'N - 3°9'E	13/6/1904	Sable fin gris	Sondeur VI.	710	51,2600 - 3,0800	

<u>Table IV</u>: Samples from the Gilson collection selected for trace metal analysis. The samples are grouped in function of their vicinity to current monitoring stations

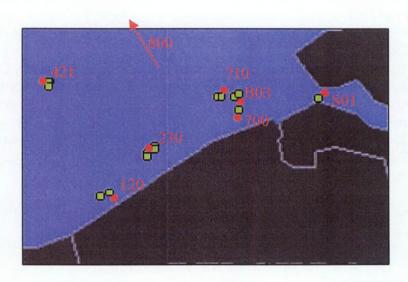
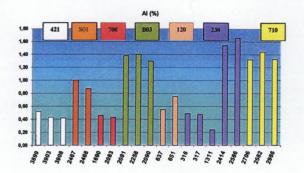
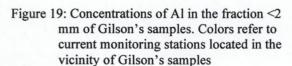


Figure 18: Geographic distribution of samples (CFF Edit map). Green: Gilson samples; Red: current monitoring stations (station 800: out of map). Comparative results are obtained in the literature (GUNS et al., 1997) for stations 700, 710 and 800

Al contents of the samples vary from 0,24 to 1,66 % of the sediment weight (fig. 19). Within the samples located in the vicinity of station "230", Al content vary to a large extent.

In samples for which the content of "mud" (fraction < 63  $\mu$ m) was determined, low values were obtained (fig. 20). Two samples have a mud content of a few percents.





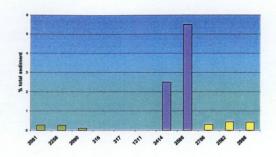


Figure 20: Percentage of <63 μm fraction determined for some Gilson's samples (groups of samples in the vicinity of stations B03, 230 and 710)

When considering sampling dates, positions and instruments, the analysis of measured levels is difficult at this stage. Within "geographic" groups, variation is observed for the considered metals in the <2mm fraction. Detailed figures of metal levels in the total sediment (<2 mm, mg/Kg and normalisation to Al content) are provided in fig. 21.

When the levels of sample n° 1311 are normalised to the Al content, they are out of the range. This sample is also the only one to be collected with the "ground-collector n°3". This could due to a problem in the analysis (in particular the Al content) as well as from a secondary contamination. A re-analysis of this sample is necessary to confirm the result. Therefore, it will not be further considered in the following discussion. Another sample (2090) presents relative high levels of Pb, Cu and Cd. However, Al normalisation indicates that this sample is in the range of the sample set.

It was feared that instrument "n°1", suspected to contain lead parts, could induce a Pb contamination. Amongst the five samples located near the station 230, two sediments were sampled with this instrument (n° 316 and 317). No abnormal level is observed. This is also true for samples near station 120.

The differences observed between samples located near the station 230 are interesting and can already be attributed to a difference in sediment composition (mud and Al contents).

In general, we observe large variations in Zn, and to a lesser extent Cd levels, even for samples collected in similar conditions. However, Zn levels are varying with a similar amplitude in results from recent analyses of coastal stations (Guns *et al.*, 1997). Pb and Cu levels are relatively similar within stations when normalized to Al.

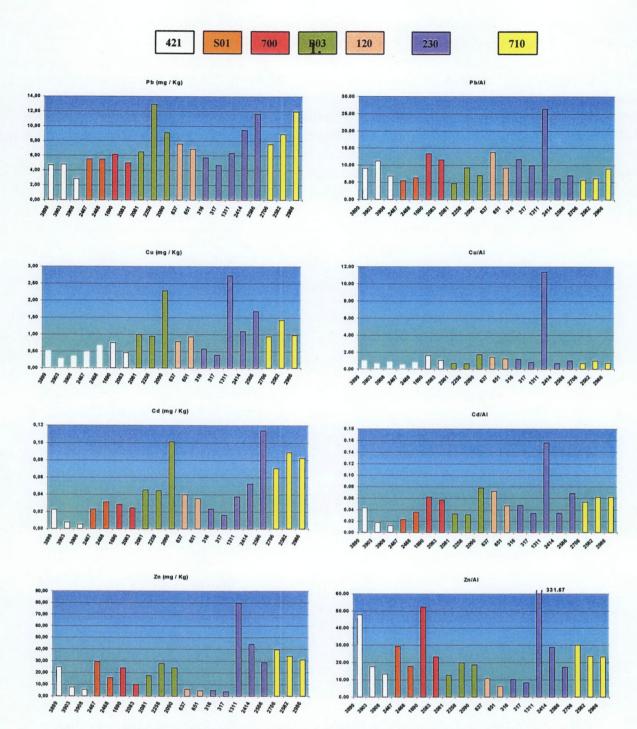


Figure 21: Gilson's sediment samples: Concentrations of four metals (Pb, Cu, Cd, Zn) in the <2mm fraction (mg / Kg) (left) and normalised to Al content (right). Colours refer to geographic area of samples.

#### Comparison with levels measured in recent monitoring campaigns (1979 – 1995)

In order to evaluate the range of "old" metal concentrations as compared to the nowadays situation, levels measured in sediments (<2 mm fraction) in monitoring stations of the BCS during the period 1979 – 1995 were taken from GUNS *et al.* (1997). Mean level and standard deviation of the period 1979 – 1995 were calculated for Cu, Pb and Zn in three stations. For Cd, no result was available on the 2 mm fraction in Guns *et al.* (1997). "ZO" ("Zeebrugge Oost", = station n° 700) and "S2" ("Loswal S2" = station n° 710) are sites of dumping of dredged material. "NO" ("Noord" = station n° 800) is an offshore station of the BCS. These levels were plotted against mean levels of Gilson's samples (sample n° 1311 excepted) (fig. 22). Cu levels are, in most Gilson's samples, lower than current levels in any station. This allows us to

consider that the secondary contamination for this metal is minimal, if ever existing. Historic levels are particularly low.

Levels of Pb are not low in Gilson's samples. In the years 1900, this metal was already largely used and significant levels could be expected. Historical and current offshore levels are similar (see fig. 21). However, the levels measured appear to be higher than nowadays in many Gilson's samples. For Zn and Cd (see also fig. 21), a large variation pattern is observed in Gilson's samples, even within the considered areas. However, large geographical variation is also observed nowadays (GUNS *et al.*, 1997). Further discussion on these metals is difficult at this stage of the research.

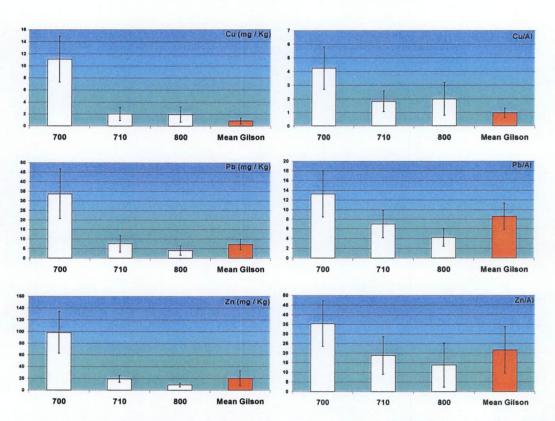


Figure 22: Comparison of Pb, Cu and Zn mean levels in three monitoring stations (1979 – 1995; source: GUNS et al., 1997) (white) and Gilson samples.

#### 4.4.4. CONCLUSIONS.

- The results obtained with the "ground-collector n° 1", suspected to contain lead parts, tend to indicate that the metal composition of the instrument shouldn't be considered when sediment samples of the Gilson collection are selected for metal analysis. However, this parameter is available if aberrant results are recorded, as in sample n° 1311.
- ➤ Only one of the twenty sediments analysed showed evidence of abnormal levels. For Cu and Pb, the levels can be considered as relatively similar when Al normalisation is applied. If secondary contamination induced by storing conditions accounts for the levels observed, it should thus be low, although occasional stronger contamination is always possible.
- We can already state that historical Cu levels are lower than nowadays. Although Pb levels are quite high, the levels observed for this metal are relatively uniform. For Cd and Zn, the situation is rather unclear due to a large variation pattern. However, this is also the case in the recent monitoring results. It is well known that the geographic variation in trace metal levels of marine sediments is a complex problem.
- > The results of metal analysis in the < 63 μm fraction will be necessary for more decisive conclusions. The Gilson's samples will certainly be most interesting for an investigation of long-term trends of the considered trace metals in sediments of the BCS.

#### 4.5. CASE-STUDY 5: HISTORIC HABITATS MAPPING

As mentioned in 2.4, nearly 3000 sediments have been collected with the "ground-collector" and documented by Gilson. In particular, qualitative descriptions of sediments and depth measurements were recorded, together with tide status, time and geographic information.

CATTRIJSSE and VINCX (2001) and others have recently highlighted the importance of sediment grain-size and depth data for the definition of macrobenthic species habitats in the BCS. Therefore, our aim is to determine whether these parameters, recorded by Gilson, could be used for a research on the distribution of "historic habitats" of the benthic fauna in the BCS.

Although only 691 samples remain in the RBINS collections, a large part of the qualitative descriptions of non-stored sediments were preserved in the original log-books. Our aim is to investigate the reliability of grain-size categories established on the basis of Gilson's descriptions. For that purpose, we studied the sub-set of 691 stored sediments.

#### 4.5.1. SEDIMENT DATA

#### 4.5.1.1. GRAIN-SIZE CATEGORIES: ANALYSES AND RESULTS

In the "sub-set" of 691 data (see 3.1, archived sediments database), a total of 439 data (64 %) were found to be directly usable (documented with coordinates and sediment descriptions). More points should be usable provided that a recalculation of coordinates is performed. For this study, we have focused on the directly usable information.

For these 439 data, we have examined the sediment descriptions. The descriptions are generally performed in a "standardised" format. The descriptions have been extracted from the "Explomer" databank, verified with the available original log-books and investigated. On the basis of this analysis, a series of sediment categories and sub-categories can be proposed (table V).

Mud
Mud with sand
Mud with sand and shell remains
Mud with sand, shell remains and gravels
"Fine" sand
Fine sand with mud
Fine sand with mud and shell remains
Fine sand with shell remains
Fine sand with mud, shell remains and gravels
"Medium" sand
Medium sand with mud
Medium sand with mud and shell remains
Medium sand with shell remains
Medium sand with mud, shell remains and gravels
"Coarse" sand
Coarse sand with mud
Coarse sand with mud and shell remains
Coarse sand with shell remains
Coarse sand with mud, shell remains and gravels
Gravel
Fine gravel
Coarse gravel

Table V: Grain-size categories proposed on the basis of Gilson's description of sediment samples.

We have studied the adequation between the descriptions and grain-size profile of samples. Two approaches were considered:

- The study of 82 grain-size analyses performed by Gilson.
- 12 new grain-size analyses on preserved samples.

## 4.5.1.1.1. Original "old" grain-size analyses

## 4.5.1.1.1.1. Gilson's grain-size analysis method

GILSON (1900) describes the techniques used for the mechanical separation of grain-sizes. A series of 6 sieves of different meshes were used, the lowest mesh corresponding to a sieve aperture of 0.5 mm (table VI). For smaller particles, Gilson used an elutriation instrument adapted from THOULET (1900): three fine fractions were separated using an ascendant siphon. Particles remaining at the basis of the siphon were classified as "fine sands" (VII). Two further mud categories were assigned to particles going through the siphon, which respectively settled down after one hour (VIII) or did not settle down (IX). The mean diameter of these three fractions can probably be estimated using settling parameters.

Grain-size fractions (Gilson, 1900)	n°	Sieve n°	Sieve aperture (mm)	
PIERRES	I	1	15	
GRAVIER GROSSIER	II	2	5	
GRAVIER MOYEN	III	3	2,5	
GRAVIER FIN	IV	4	1,5	
SABLE GROSSIER	V	5	1	
SABLE MOYEN	VI	6	0,5	
SABLE FIN	VII	NOT THRU SIPHON	SIPHON	
"VASE"	VIII	THRU SIPHON, SETTLING in 1 hour	SIPHON	
"CORPS LEGERS"	IX	THRU SIPHON, no SETTLING after 1 hour	SIPHON	
•	Х	THRU SIPHON, no SETTLING after 1 hour, PRECIPITATION WITH ALUN	SIPHON	

<u>Table VI</u>: Summary of information provided by Gilson (1900) about sieving operations for grainsize analysis.

A total of 82 analysis sheets were found. In these sheets, the grain-size fraction n° VIII is called "very fine sand" instead of "mud". Fraction IX, and a further fraction X ("precipitation with Alun") are together summed to constitute the "mud" percentage in the sample. The fraction X has only been determined on rare occasions and is always negligible.

Of these 82 samples, only one (sample n° 925) has been preserved as a whole sample in the RBINS. The grain-size fractions isolated in the other samples have been stored in glass tubes and were preserved, but the original whole sample was lost. Samples n° 915 to n° 1000 (77 data) were collected in 1901. These samples belong to a series of close sampling sites along a coastal transect off Oostduinkerke. They were performed in order to obtain a bathymetric and sedimentologic profile along this line, up to 10 nautical miles offshore. Exact coordinates of each point have not been found so far.

## 4.5.1.1.1.2. Analysis of grain-size profiles and discussion

#### Methods

All results compiled in the old datasheets were copied in a MS-Excel file for further processing. The weight of every grain-size fraction (from 1 to 9) was normalised to the total weight. When the sum of all fractions was different from 100 +/- 0.5 %, Gilson's calculations were checked and appropriately corrected. We have selected samples 915 to 1000 (77 samples), considering that they were of particular interest (Oostduinkerke transect). Sediment descriptions were extracted from the "Explomer" databank and corrected on the basis of original log-book information.

On the basis of these descriptions, we have divided the samples into two groups of sediments: "homogenous" (42 samples) and "inhomogenous. For the first group, a classification based on descriptions was confronted with a multidimensional classification of grain-size profiles (classification with entropy theory, R<sub>s</sub>; JOHNSTON and SEMPLE, 1983). For the second group, no multivariate analysis could be applied.

#### Results

With the multidimensional classification, 92.06 % of the differences between homogenous samples is explained when they are classified into 6 groups. The analysis of dominating fractions within groups (annex 4) indicates that

- In group 1, fraction IX dominates samples described as "mud".

- In group 2, fraction VIII dominates samples described as "fine sand". The description of mud presence in the sample n° 970 is not evidenced

- The group 3 is characterised by fraction VIII with 10-20 % of fraction VII, and comprises all remaining "fine sand" descriptions. All samples are described to contain only traces of mud, which is consistent with the low content (1 %) observed.

- The groups 4 and 5 comprise all samples described as "pure sand" except one. They are constituted of fraction VIII and VII, with increased proportions of fraction VII in group 5.

- The group 6 is dominated by the fraction VII and contains some 10 % of fraction VI (0.5 mm). One sample is described as "coarse sand" and the second as "pure sand".

## The following observations can be made:

1. Most samples are classified as expected from the analysis of descriptions.

2. Most samples are constituted of sediments separated on the basis of the elutriation procedure. The three elutriated fractions are clearly distinct.

3. The grain-size fractions are by far smaller than expected from table VI. The "coarse sand" description seems to be characterised by the fraction VII (< 0.5 mm), which should correspond to "fine sand" in table VI.

4. Mud samples are distinct from other samples. The "sandy mud" description (sample n° 940) is even evidenced by a higher proportion of fraction VIII, although fraction IX still dominates the pattern.

5. Except in one case (sample n° 998), "pure sand" description is clearly different from "fine sand". It should correspond to a "medium sand".

6. Unclear descriptions (for example sample n° 942) should be discarded

For sample 998, the wrong result could be due to an isolated copying error. If we except this sample, we can consider that "mud", "fine sand", "pure sand" and "coarse sand" are all reliable. At this stage of the research, it is not feasible to attribute a particle size for each category. However, these can be ranked as follows:

"Mud": fraction IX

"Fine sand": fraction VIII

"Pure sand" (= "medium" grain-size?): fractions VII – VIII in similar proportions

"Coarse sand" (provisional): fraction VII dominating

"Gravel" (not investigated): > fraction VI (0.5 mm)

The "pure sand" description should be interpreted as a medium sand. However, in the descriptions of the sub-set of the 691 sediments remaining in the RBINS collections, this description is rare. The "medium sand" description was found more often.

The cumulative grain-size profiles plotted for the "<u>inhomogenous</u>" group shows that the descriptions of presence of mud or shell remains are clearly evidenced (fig. 23.). "Mud", "fine sand" and "coarse sand" are, again, distinct profiles. This demonstrates that further sub-categories can be assigned to the main categories. This heterogeneity parameter is of prime interest when considering habitats for benthic species.

In addition, other peculiarities of samples were also recorded by Gilson. Black mud presence, indicating anoxic conditions in the sediment, is mentioned, as well as the occurrence of clay, peat or pebbles (such as flintstone).

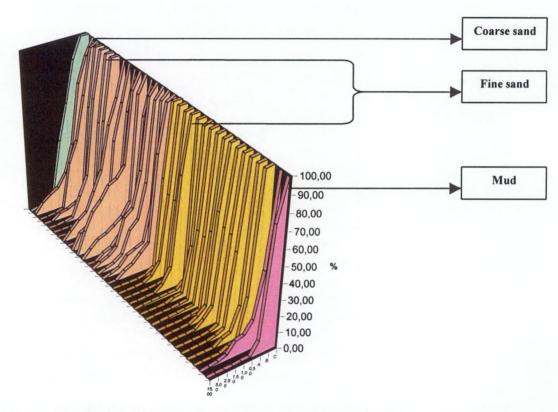


Figure 23: Cumulative frequencies of grain-size fractions I to IX for the "inhomogenous" group of sediment descriptions. Text boxes: empiric classification on basis of descriptions (mud, fine sand, coarse sand).

We can thus conclude that the transcription of the considered descriptions into reliable grain-size categories seems to be feasible on most occasions. However, at this stage of the research, we are not able to attribute a grain size measure to the considered fractions. This could only be achieved by considering a Gilson's elutriation system.

## 4.5.1.1.2. New grain-size analyses.

In the "old" available analyses, descriptions of "medium sands" were not represented and "coarse sands" were rare. Therefore, new analyses were performed focusing on these categories. 12 of the 439 samples were selected on the basis of sediment descriptions (table VII).

Sample n°	Description						
1390	Sable moyen un peu néritique						
1621	Sable moyen piqueté						
3730	Sable moyen						
4028	Sable moyen brunâtre						
s161	Sable moyen						
s226	Sable moyen						
3485	Sable grossier extrêmement néritique						
3493	Sable grossier légèrement néritique						
3495	Sable grossier très finement néritique						
s96	Sable grossier						
s171	Sable grossier						
s135	Vase noire, grisâtre, un peu sableuse et néritique						

Table VII: samples used for new grain-size analyses and their original description by Gilson.

## 4.5.1.1.2.1. Analysis methods

Samples were processed in the laboratory of the Petrography / Sedimentology section of the RBINS. After homogenisation of whole samples (shaking), 20 + -0.1 g of each sample was weighed. Diluted  $H_2O_2$  (28%  $H_2O_2$ , 1:3 in water) was added to the sample on a hot plate for the removal of organic material. Carbonates were not removed. After rinsing with demineralised water, samples were dried in an oven at 105 °C overnight.

The fine fraction (mud + clay) was separated on a 63  $\mu$ m sieve with addition of a solution of 1.33 g Na carbonate and 8.93 g Na oxalate per liter water.

Further sieving has been performed for larger grain-sizes (mm): 2, 1, 0.5, 0.250, 0.125.

Samples s161 and s135 were not properly processed. However, at least 28 % (underestimation) of sample s135 is constituted by fraction < 63  $\mu$ m, confirming the description as "mud with sand". Strubbe and Pichot (1993) have analysed this sample and found a mud content of 70 %.

Sample 1621 was analysed twice.

Sample n°3485 was identified as containing formalin (odour, grains aggregation).

#### 4.5.1.1.2.2. Results and discussion

The cumulative grain-size profiles, grouped in function of categories assigned with original descriptions, show that only 2 "medium sand" profiles (1621 and 4028) on a total of 5 can be considered as clearly different from coarse sand profiles (total 5) (fig. 24.).

The two analyses performed on 1621 gave identical results (not illustrated).

The "neritic" description (presence of shell remains) is confirmed by grain-size profiles (samples 1390 and 3485).

We observe that the "coarse sands" are characterised by a mode at 250  $\mu$ m (fraction 4). This observation is consistent with the unique result of the "old" analyses. However, only two "medium sand" descriptions are different (125 – 250  $\mu$ m) from the "coarse sands" profiles.

This tends to indicate that "medium sand" description is doubtful.

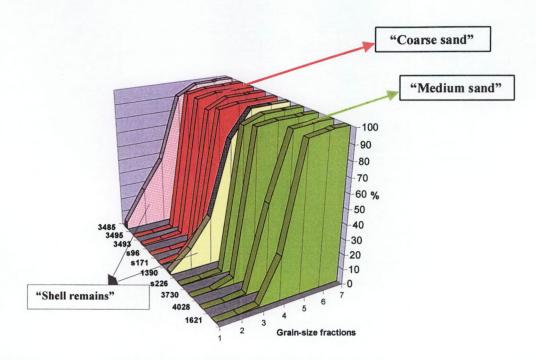


Figure 24. Cumulative frequencies of grain-size fractions in new analyses. Fraction numbers 1 to 7 (μm): 2000, 1000, 500, 250, 125, 63, <63. Red profiles: "coarse sand"; green profiles: "medium sand". Text boxes: original description.

## 4.5.1.2. GENERAL CONCLUSION OF THE SEDIMENT DATA ANALYSIS

The "old" analyses have provided an evidence that the considered samples could be ranked in function of the description. At this stage of the research, we can consider that "mud", "fine sand", "coarse sand" and "gravel" are most probably reliable categories. Furthermore, the description of presence of mud, shell remains, pebbles, etc. can also be considered as reliable. However, the new grain-size analyses tend to indicate that "medium sands" are not as reliable as "pure sand" in old analyses.

Further grain-size analyses will be necessary for more decisive conclusions on the possible use of "medium sand" descriptions and to assign a grain-size range to each category.

#### 4.5.1.3. PLOTTING GILSON'S DESCRIPTIONS ON VAN MIERLO'S SEDIMENTOLOGIC MAP

In order to make a very first attempt of sediment mapping, we have plotted some of the sediment descriptions of Gilson on an old sedimentologic map of the BMA (VAN MIERLO, 1899) (Fig. 25). This map was originally produced on the basis of 300 sampling sites. The sediment categories assigned by VAN MIERLO (1899) are based on qualitative descriptions. This map has been used or quoted as the long term "reference" sedimentologic map of the Belgian Continental Shelf (BCS) (BASTIN, 1974; Strubbe and Pichot, 1993).

There is a general good agreement of Gilson's descriptions with Van Mierlo's general appreciation, although local heterogeneities are highlighted by Gilson samples. For example, the large area described by Van Mierlo as "sandy mud", in front of Blankenberge, harbours various combinations from pure mud to pure fine sand according to Gilson. There is a good agreement between authors in the "Hinders" area. In the Schelde mouth, Gilson finds coarse sands while Van Mierlo only mentions fine sands.

According to our limited study, the information yielded by the map of Van Mierlo can be ameliorated on the basis of Gilson's data, since:

- (1) The number of samples will be much higher.
- (2) The categories of Gilson could hopefully be defined with reference to a grain-size range, allowing direct long-term comparisons.

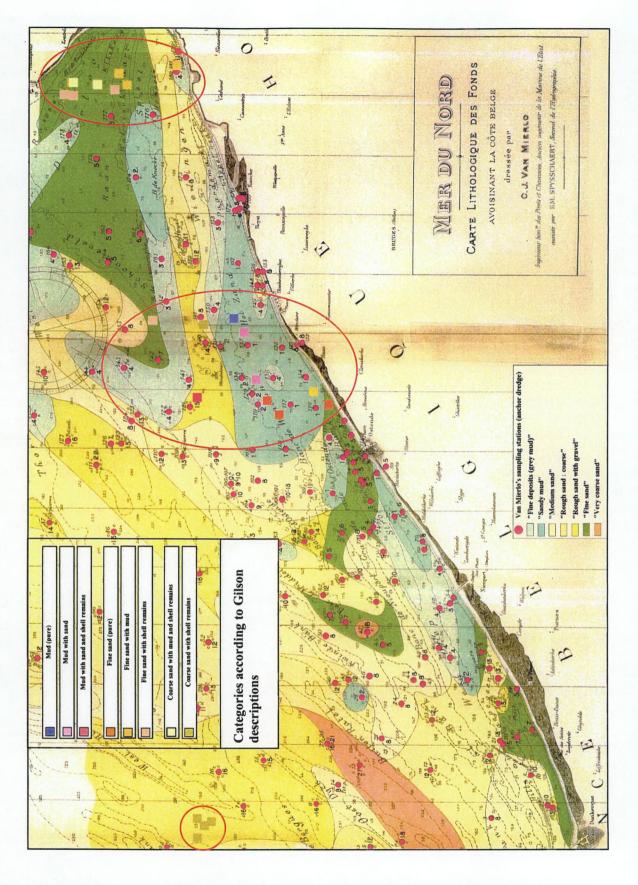


Figure 25: sedimentologic map of VAN MIERLO (1899) modified with addition of some Gilson's descriptions. Background colours: grain-size categories according to Van Mierlo (see map legend). Red points: Van Mierlo's sampling points. Colour squares (in red circles): grain-size category of the sampling site according to Gilson's descriptions (see text-box).

### **4.5.2. DEPTH DATA**

## 4.5.2.1. DATA ANALYSIS

As detailed in point 3.1 (database of sediments in the RBINS collections), many depth data are associated with the collected sediments. The quality and possible use of these data is examined in the aforementioned sub-set of 691 data.

A total of 542 depth data were found in "Explomer" for this sub-set. In the sea log-books, 466 measured depths are registered. The latter data were added to the archived sediment database (see 3.1). The two information sources are found together in 389 cases (56 % of total data-set).

By comparing original sea log-books to the Explomer (card-index system) data, we have found that part of the depth data have seemingly been reduced to a common low tide reference level (corrections occur until august 1903). The presence or absence of such a reduction is not mentioned in Explomer. Furthermore, some copying errors have been detected in Explomer. It will therefore be necessary to verify all depth data in available log-books in order to know the original depth measured (feasible on 71 % of the depth data of Explomer for this sub-set). Exact time of sampling is not mentioned systematically in Explomer but can also be found in the field log-books, together with date and approximate tidal status.

#### 4.5.2.2. DISCUSSION AND CONCLUSION

The presence of depth reduction in a part of the sub-set, as well as copying errors, indicate that only data in original log-books should be used in order to obtain "measured" depths.

The question resulting from the observation of depth reduction in a part of the data set is to determine the original reference level used by Gilson. The answer was found in GILSON (1907). The reference level is the mean lower low water level of spring tides. This level is described to be the reference (zero) level of the "Service des Ponts et Chaussées". Most interestingly, it is situated exactly 1,48 meter above the basis of the sea lock of the dock "bassin du commerce" (Handelsdok) in the Oostende port. From this paper, we also learn that Gilson reduced a part of his measured depth using tide amplitude measured at coast.

In brief, all measured depths of Gilson can be referenced to a common level, using mathematical models for tidal reduction offshore. The reference level can be exactly situated with reference to a concrete building, the handelsdok being today the Mercator's dock.

It is thus theoretically feasible to associate data of benthic fauna to relative depths in different geographic sites of the Gilson sampling surveys and to evaluate the faunistic long-term changes with respect to this parameter.

For that purpose, further examination of precision of depths and geographic coordinates measurements are necessary. It is anticipated that the relative error will be small since:

- GILSON (1900) has given much attention to the determination of the "localities" sites, providing a detailed method of geographical positioning.
- The Belgian coastal waters are shallow. Therefore, most hand measurements of depth should not be much biased.

## 4.5.3. CONCLUSIONS

Our aim was to investigate whether the information of sediment nature and depths provided by Gilson could be used as "habitat" parameters for the benthic fauna of the BCS. The results obtained in this short case-study are promising.

The sediment descriptions of G. Gilson are, at least partly, reliable for a mapping of grain-size categories. More grain-size analyses will be necessary for stronger conclusions as well as for a characterisation of the grain size range of each category. Furthermore, a reliable reference to the sediment heterogeneity can be performed. Provided that the considered sub-set of 691 data is representative of the total sediment data-set, an estimated 1900 sediment data should be usable. If "medium sand" descriptions are considered not reliable, the "gap" on the resulting map could probably be solved by the application of interpolation techniques. An estimated 2000 depth data should also be theoretically usable.

The characterisation of "habitats" of the benthic fauna in the early century as a reference for a long-term investigation on benthic biodiversity appears to be feasible at medium scale in the coastal waters (up to 10 miles offshore) and the area of the "Hinders" banks.

## 5. END PRODUCTS OF THE FEASIBILITY STUDY

This report constitutes a general quality investigation of the GILSON collection and provides some insight in its suitability for particular applications.

The end products as defined in the project proposal (1999) are:

- 1. A <u>bibliography</u> comprising publications referring or relevant to the collection and the explorations of GILSON. The list is annexed. The publications by GILSON himself are listed in DEBAISIEUX (1937).
- 2. Two databases with biotic and abiotic data on the southern North Sea. Firstly, <u>EXPLOMER</u> comprises all geographic data and *in situ* environmental measurements of the explorations of GILSON. Specific information on the archived sediments was extracted and copied into a MS-Excel file. Secondly, the <u>SOUTHERN NORTH SEA SPECIES DATABASE (SNSSD)</u> has been created to store the results of faunistic-taxonomic revisions, such as performed in the case-studies. This database will be developed by adding all data from the GILSON collection and other relevant RBINS collections. These databases will be incorporated into the Integrated and Dynamical Oceanographic Data management (IDOD).
- 3. <u>Historical distribution maps</u> of the neogastropods. Maps of echinoderm and ray species, not presented in this report, have also been prepared.
- 4. A historical map of dredge n°5 sampling intensity for the period 1898-1914 has been developed.
- 5. A paper on the historical distribution of the neogastropods is in preparation.
- 6. A <u>short investigation on historic zoological collections in neighbouring countries</u> of the North Sea was performed, showing that Gilson's survey was quite unique in its design and, consequently, in the resulting collection. Contacts have been taken at national and international level in order to participate in cooperative projects on long-term changes in the marine fauna.

## 6. CONCLUSIONS

A feasibility study has been performed on the Gilson collection in order to evaluate its scientific value and its possible use as a reference framework for the Belgian marine fauna.

The intensive and systematic explorations conducted by Gilson, as well as the resulting sedimentological and zoological collections, are quite unique for the North Sea. The preserved collection comprises tens of thousands of biotic and abiotic samples and measurements.

The archives and samples of the Gilson collection are well preserved and managed. The quality of the considered samples ranges from good to excellent. Most of the biota are labeled and identified. However, taxonomic revisions will be necessary for further research. The feasibility study has focused on molluscs, echinoderms, crustaceans, fishes and sediments.

This detailed information is compiled into a database format, such as the SNSSD. The SNSSD will constitute a valuable tool when applications on the faunistic collection are to be considered. The data will be incorporated into the Integrated and Dynamical Oceanographic Data management (IDOD).

Five case-studies have demonstrated that the quality of the samples and sampling information allows several scientific applications: faunistics, taxonomy, population genetics, trace metal analysis, sedimentology...

Our investigation highlights the particular interest of this collection for biodiversity research. Hence, the Gilson collection is highly suitable for the establishment of a century-old reference point for the biodiversity of the Belgian marine areas.

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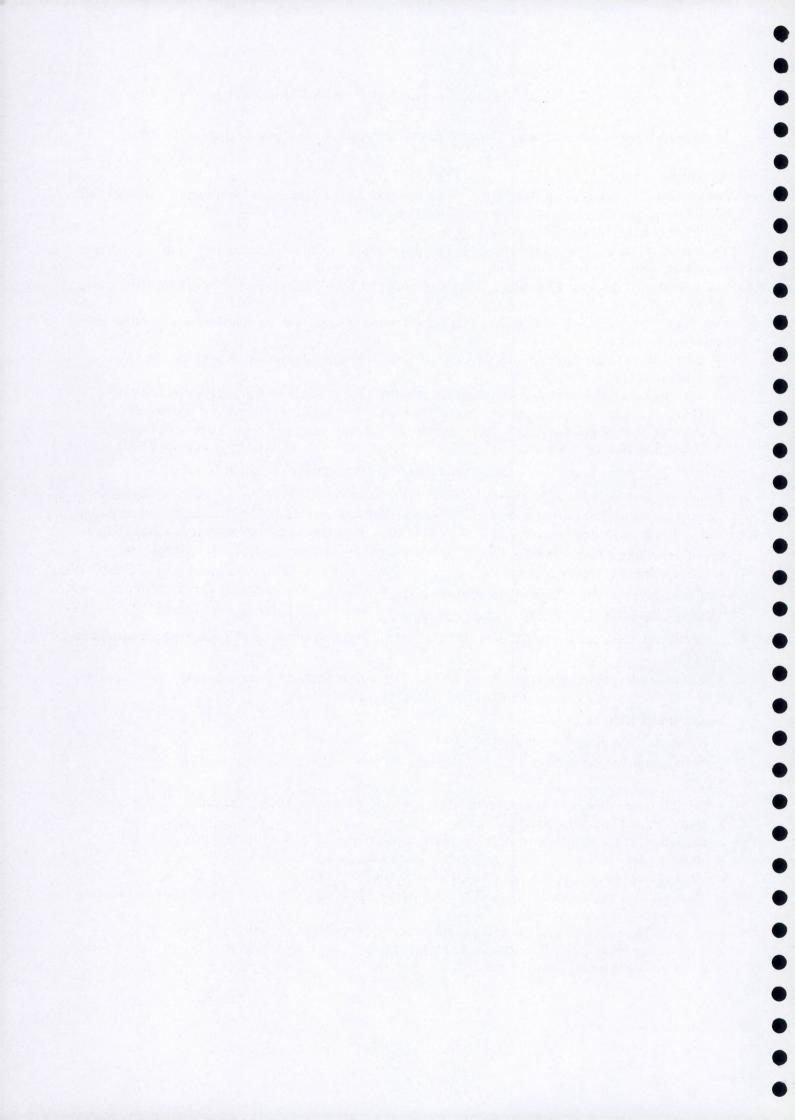
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# ANNEX 2: List of compiled archived documents

- General archives of the Institute, G. Gilson (letters, scientific correspondence, personal documents, notes, etc.)
- Log-books
  - ➤ Plankton: 19 books (Exp. Mer. I. Rét.) + 46 books (ICES) + data sheets from various cruises + folder with information on localities of some plankton samples
  - > "Observations bi-horaires": 99 books
  - ➤ Various meteorological and oceanographic observations: 63 books + data sheets on t°, salinity and density of water
  - > Sediments: 13 books (Exp. Mer I. Rét.) + 19 books (ICES) + 132 sheets (n° 68 to 200; Exp. Mer. I. Rét)
  - > "Pêches" (trawl): 27 books (ICES) + data sheets (fish species, size and numbers of specimens per station)
  - ➤ Inventory of sediments and dredging: n° 501 to 980 (1900) [information on locality + environmental parameters]
  - Fish sold at Oostende "Minque" (numbers of baskets / sp + origin of fish) in December 1908
  - > One folder containing information on many localities (equivalent to the general inventory?)
  - > A series of log-books of temperatures, salinity and density measures in the harbour of Oostende
  - A series of data sheets from ICES cruises
- Manuscripts: many original manuscripts and data of Gilson publications could be found
- A card-index system of more than 14,000 cards (sampling "localities": see 3.1, Explomer databank)
- General inventories of Gilson's samples: "Répertoire Général Exp. I Mer"; "Croisières diverses Exp. I. Mer"; "Etude du milieu marin P. PO Exp. I. Mer" (data West-Hinder); "Exploration du littoral Exp. I. Mer" (Estran, bateaux-phares et Entrées); "Etude du Necton, pêches C, Cm, Cr, L.P Exp. I. Mer"
- A series of bibliographic cards
- Construction plan of the "Eggnet" (planktonic net)
- Typical specimens of log-books (including dredging)
- General map of the sample collection in the maritime institute of Oostende (= before sample transfer to the RBINS)
- List of mollusk species caught by Gilson with locality information (n°, cruise and geographic position) (NB with catches in the "Spuikom (Oostende) from years 1947-1948)
- 82 Grain-size analysis data sheets
- A folder with samples of net clothes
- A maregraphic recording from the Rijkwaterstaat, 01 06 / 1938
- Maps:
  - > Original map of the current experiment (bottles) with positions of bottles indicated
  - ➤ Map with first sampling sites (bad quality status)
  - > 17 Maps of marking-recapture data for *Pleuronectes platessa*, 1904-1908, large format
  - Maps of bathymetric profiles of the two ICES transects studied by Gilson
  - Maps of plancton tracks at coast; 1904, 1905 and 1907
  - ➤ Hydrographic map BCS, 1901 1905, "Serv. Spécial de la côte, Ponts et Chaussées"; with notes on stations positioning
  - > Maps of 7 bathymetric profiles in the southern North Sea, Gilson et Lauwers
  - > Very large format map of the Belgian coast: reporting of sampling sites in Expl. Mer I Rét. (incomplete)
  - Distributions maps on several taxa



# ANNEX 3: SNSSD glossary

# Table of samples

Sample n°: entry number.

Genus, species: the fields of genus and species comprise the scientific name of the species (in latin), with possible differentiation to subspecies name or variety. According to recent nomenclature.

Var.: abbreviation for 'variety.

Conservatory: exact place in the depository room of the RBINS.

Preservation: way of preservation; in alcohol, dry, in formol or as a microscopic slide.

- Drawer or cup n°: in this example the sample is conserved in cup or jarnumber 42/35. Dry samples will refer to a drawer number.
- Registrationn° in 'old list': number of registration on the 'old species lists' or species based inventories.

Collected by: name of the institute or person who has collected the sample.

Identified by: name of the person who has identified the sample, year of identifying.

# spec: number of specimens in the sample.

Age: lifestage of the specimens in the sample. Can be all eggs, juvenile or adult specimens, mixed or unknown...

Sex: male, female, hermafrodite, indefinite, or mixed...

Former identified as: only in case of former synonym names or misidentifications. When there are numerous former names, they can be listed chronologic.

Former identified as: name and year of the former identifier(s).

Recordn° (recordnumber): registration of a sample in the database. Coupled with the type of excursion it is the key to define a sample. Regarding to the GILSON collection, the recordnumber defines a sampling on a certain hour, date, locality and with a certain instrument and has been registered in the GILSON inventories. Any

change of one of these four parameters will imply for the GILSON samples a change in recordnumber.

Type of excursion: field to foresee a possible differentiation in excursions, such as in the case of GILSON who during his 3 overall marine programmes has made 8 different types of excursions in the southern North Sea:

Exp. I. Mer 1 Rét., Exp. I. Mer 2 Cr., Exp. I. Mer 3 Rad., Exp. I. Mer 4 Crois. Div., Exp. I. Mer 5 Mil., Exp. I.

Mer 6 Pl., Exp. I. Mer 7 Nect. and Exp. I. Mer 9 Litt.

Status sample: specific remarks on status such as dessication, damaged, not enough label information or doubtful label information...

Remarks: other relevant information than mentioned in the above fields.

#### Table of localities

Recordn° and type of excursion: key to define a sample, see also table of samples.

Cruisen°: in case of different cruises.

LATIT. 1 & LONGIT. 1: latitudinal and longitudinal coordinates of a sampling site.

LATIT. 2 & LONGIT. 2 and LATIT. 3 & LONGIT. 3: in case of dredging or trawling; refers to the

latitudinal and longitudinal coordinates from an endpoint, or from a sampling site in between.

IGn°: number assigned to each collection which enters the RBINS.

- Date of record.
  - Locality or traject: sometimes descriptions are given in stead of geographic coordinates.

Hour of record: only for a relatively low number of GILSON samples

Depth of record: measurement in meters or description, such as e.g. floor

T° water: temperature of the watersurface

- Winddirection.
  - Windvelocity.
  - Currentdirection.

Current velocity.

Tide status: ebb or flood.

Depth of sea: measurement in meters.

Salinity.

Substrate: description. For GILSON samples, a description of the sediments was made.

Method of collecting: name or description of the sampling instrument.

Remarks: other relevant information on the sampling sites than mentioned in the above fields.

# Table of systematics and taxonomy

Genus, species, Author, Year, Var.: scientific name of the species, with possible differentiation to subspecies name or variety...and according to recent nomenclature. In addition, fields are available for name and year of the author.

Nomenclature according to: source of reference.

Synonyms according to: listing of all synonyms, with source of reference.

Dutch common name(s).

French common name(s).

Phylum, Classis, Ordo and Family: refer to systematic classification and are according to a source of reference.

Remarks: other relevant information than mentioned in the above fields.

# Table of species related publications

Genus and species: scientific name of the species.

Author(s) of the publication.

Year of publication.

Code RBINS: code applied by the RBINS library to identify the paper or periodical.

Title of the publication.

Periodical, volume, pages and publisher.

# ANNEX 4: results of multivariate classification of sediments

Results of entropy classification in 6 groups ( $R_s = 92.06$  %; Johnston and Semple, 1983) of the relative frequency of grain-size fractions. The typical features of groups are highlighted.

	n°	Grain-size fractions (relative frequency						frequ	ency)		
Group (Entropy)		15 mm	5 mm	2 mm	1.5 mm IV	1 mm V	0.5 mm VI	A VII	B VIII	C (mud) IX	Description
		I	II	III							
-	939	0.00	0.00	0.00	0.00	0.00	0.00	0.02	0.03	0.92	Vase noire, très léger vestige de sable
	940	0.00	0.00	0.00	0.00	0.00	0.02	0.08	0.21		vase noire, trace grise, vase sableuse noire, trace de sable jaunâtr
9	969	0.00	0.00	0.00	0.00	0.00	0.00	0.02	0.96	The last two last to the last	sable fin gris
	970	0.00	0.00	0.00	0.00	0.00	0.00	0.01	0.97		sable fin gris, un peu vaseux, et vase grise séparée
	971	0.00	0.00	0.00	0.00	0.00	0.00	0.02	0.96		sable fin un peu vaseux
3 9	915	0.00	0.00	0.00	0.00	0.00	0.00	0.08	0.90	0.02	Sable fin, grumeaux de vase noire, traces de grise
	917	0.00	0.00	0.00	0.00	0.00	0.00	0.15	0.84	0.01	
	918	0.00	0.00	0.00	0.00	0.01	0.00	0.12	0.86		Sable fin, grumeaux de vase noire, traces de grise
	919	0.00	0.00	0.00	0.00	0.00	0.00	0.16	0.83		Sable fin, grumeaux de vase noire, traces de grise
	920	0.00	0.00	0.00	0.00	0.00	0.00	0.11	0.87		Sable fin - faible trace de vase noire
	921	0.00	0.00	0.00	0.00	0.00	0.00	0.11	0.87	0.01	sable fin, très petit grumeau de vase noire
	922	0.00	0.00	0.00	0.00	0.00	0.00	0.13	0.85		sable fin, faible trace de vase grise
9	923	0.00	0.00	0.00	0.00	0.00	0.01	0.19	0.78	0.01	sable fin, trace de vase noire et grise
	924	0.00	0.00	0.00	0.00	0.01	0.00	0.20	0.78	0.01	sable fin, pas de vase
4	943	0.00	0.00	0.00	0.00	0.00	0.00	0.44	0.56	0.00	Sable pur
	944	0.00	0.00	0.00	0.00	0.00	0.00	0.39	0.60	0.00	sable pur
	945	0.00	0.00	0.00	0.00	0.00	0.00	0.29	0.71	0.00	sable pur
	946	0.00	0.00	0.00	0.00	0.00	0.01	0.35	0.64	0.00	sable pur
	947	0.00	0.00	0.00	0.00	0.00	0.01	0.38	0.61	0.00	sable pur
	949	0.00	0.00	0.00	0.00	0.00	0.00	0.38	0.61	0.00	sable pur
	950	0.00	0.00	0.00	0.00	0.00	0.01	0.51	0.48	0.00	sable pur
	951	0.00	0.00	0.00	0.00	0.00	0.01	0.46	0.53	0.00	sable pur
	953	0.00	0.00	0.00	0.00	0.01	0.01	0.44	0.54		sable pur
	954	0.00	0.00	0.00	0.00	0.01	0.01	0.46	0.51		sable pur
	960	0.00	0.00	0.00	0.00	0.02	0.00	0.36	0.59		sable pur, très peu de tubes d'annélides
	961	0.00	0.00	0.00	0.00	0.01	0.00	0.33	0.64		sable pur - tubes d'ann., pas de coquille
	962	0.00	0.00	0.00	0.01	0.01	0.01	0.29	0.67		sable pur - tubes d'ann., pas de coquille
	963 965	0.00	0.00	0.00	0.00	0.01	0.00	0.44	0.54		sable pur - tubes d'ann., pas de coquille
	966	0.00	0.00	0.00	0.00	0.00	0.00	0.43	0.56		sable pur tubes d'annélides
	967	0.00	0.00	0.00	0.00	0.00	0.00	0.43	0.56		sable pur - tubes d'ann., pas de coquille
	968	0.00	0.00	0.00	0.00	0.00	0.00	0.50	0.50	0.00	sable pur sable pur
	982	0.00	0.00	0.00	0.00	0.00	0.00	0.40	0.33		sable pur
	983	0.00	0.00	0.00	0.00	0.00	0.00	0.57	0.43		sable pur
5	942	0.00	0.00	0.00	0.00	0.00	0.00	0.65	0.32	0.00	
3	984	0.00	0.00	0.00	0.00	0.00	0.00	0.60	0.32		sables vaseux gris et vaseux noir, pas très vaseux ni l'un ni l'autr sable pur
	985	0.00	0.00	0.00	0.00	0.00	0.00	0.69	0.39	0.00	•
	986	0.00	0.00	0.00	0.00	0.00	0.00	0.09	0.31		sable pur
	987	0.00	0.00	0.00	0.00	0.00	0.00	0.77	0.23		sable pur
	988	0.00	0.00	0.00	0.00	0.00	0.01	0.82	0.17		sable pur
6	941	0.00	0.00	0.01	0.00	0.01	0.10	0.55	0.27	Street Squares	
0	998	0.00	0.00	0.01	0.00	0.01	0.10	0.55	0.27	0.03	sable grossier jaunâtre et sable vaseux gris



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