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Development and validation of a video analysis software for marine benthic applications

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Abstract :

Our aim in the EU funded JERICO project was to develop a flexible and scalable imaging platform that could be used in the widest possible set of ecological situations. Depending on research objectives, both image acquisition and analysis procedures may indeed differ. Up to now the attempts for automating image analysis procedures have consisted of the development of pieces of software specifically designed for a given objective. This led to the conception of a new software: AVIExplore. Its general architecture and its three constitutive modules: AVIExplore – Mobile, AVIExplore – Fixed and AVIExplore – ScriptEdit are presented. AVIExplore provides a unique environment for video analysis. Its main features include: (1) image selection tools allowing for the division of videos in homogeneous sections, (2) automatic extraction of targeted information, (3) solutions for long-term time-series as well as large scale image acquisition, (4) real time acquisition and in some cases real time analysis, (5) a large range of customized image-analysis possibilities through a script editor. The flexibility of use of AVIExplore is illustrated and validated by three case studies: (1) Coral identification and mapping, (2) Identification and quantification of different types of behaviors in a mud shrimp, and (3) Quantification of filtering activity in a passive suspension-feeder. The accuracy of the software is measured comparing with visual assessment. It is: 90.2%, 82.7%, and 98.3% for the three case studies, respectively. Some of the advantages and current limitations of the software as well as some of its foreseen advancements are then briefly discussed.

Keywords : AVI, Imaging, Automation, Software, Macrofauna

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1. Introduction

Imaging technologies are currently used to address different questions related to marine benthic ecology (see Solan et al., 2003 for review) including: (1) the assessment of benthic biodiversity (Mallet and Pelletier, 2014; Spencer et al., 2005), (2) the study of faunal composition (Cuvelier et al., 2012; Duffy et al., 2014), (3) habitat mapping (Williams et al., 2012), (4) the characterization and quantification of behaviors and biological activities (Grémare et al., 2004; Jordana et al., 2000; Maire et al., 2007a; Matabos et al., 2011; Matabos et al., 2015), (5) the quantification of sediment reworking (Bernard et al., 2012; Maire et al., 2007b), and (6) ecological quality assessment (Rosenberg et al., 2009).

Imaging is a non-destructive technique that : (1) allows for the saving of initial raw information and thus for potential re-analysis, and (2) records both visible benthic organisms and other biological/ biogeochemical parameters resulting from biological activity such as the apparent Redox Potential Discontinuity on Sediment Profile Images (Romero-Ramirez et al., 2013). The reasons for choosing imaging technologies differ depending on the aim of each study. As an example, benthos and epibenthos sampling strategies at deep seabed present some difficulties (Jamieson et al. 2013): (1) the use of trawls and sleds does not allow for quantitative samplings, (2) the use of traps and suction samplers result in biased samplings resulting from differences in feeding regimes and motility, and (3) the use of grabs and box corers is complicated by the use of long wires and possible heteregoneities in sediment penetrability. Furthermore there are several designs for sampling epibenthos and their efficiencies are substrate-dependent (i.e. beam trawls for soft bottoms or suction samplers for hard bottoms). None of the sampling gears are recommended as a standard for a quantitative epibenthos assessment across all substrate types (Rees and Service, 1993). Moreover, some species from the epibenthos community are very mobile, making them difficult to capture when sampling. In this context, video and image analysis has proven to be an alternative and sometimes the only (i.e.

hydrothermal vents,(Cuvelier et al., 2012)) *in situ* tool adequate to assess the epibenthos at all substrate types in a qualitative and/or quantitative manner.

In or *ex situ* imaging devices for benthic surveys can be deployed from different platforms (Smith and Rumohr, 2013), which can be divided into two main types: static platforms such as benthic landers (Roberts et al., 2005), and (2) mobile platforms such as Remote Operated Vehicles (ROV) or Autonomous Underwater Vehicles (AUV). Each type of platform provides image sequences with their own specificities that have to be taken into account during processing. Both static and mobile platforms produce series of images often acquired under different light conditions (especially when deployed in shallow waters) because of: (1) changes in water turbidity, (2) changes in the intensity of natural light, and/or (3) the development of biofilms. In addition the series of images derived from mobile platforms need to be geo-referenced to produce sound habitat mapping.

Although the use of automated image acquisition systems is clearly currently spreading worldwide (Mallet and Pelletier, 2014), corresponding analyses are still most often achieved manually/visually (Cuvelier et al., 2012; Matabos et al., 2011; Sarrazin et al., 1997; Spencer et al., 2005; Vertino et al., 2010). Significant efforts have been made to automatically or semi-automatically process isolated images using either generic (Birchenough et al, 2012; de Moura Queirós et al., 2011) or specific software (Teixidó et al., 2011; Romero-Ramirez et al., 2013). With the arrival of newer video sensors, the tendency towards increasing video duration, image resolution and time frequency acquisition, makes corresponding image analysis very time-consuming (Edgington et al., 2006). Locating sequences of images containing important information would lighten this task (Lebart et al. 2003). Moreover, the complexity of the information contained in each image makes it highly operator-dependent (Cuvelier et al., 2012; Germano et al., 2011). There have been some attempts to generate automated image-analysis procedures (Aguzzi et al., 2011; Edgington et al., 2006; Kannappan and Tanner, 2013; Lebart et al.,2003; Mane and Pujari, 2014; Nowak et

al., 2008; Romero-Ramirez et al., 2013; Teixidó et al., 2011). These have included both the automatic detection (Aguzzi et al., 2011; Edgington et al., 2006; Kannappan and Tanner, 2013) and in some cases even the identification of large epibenthic organisms on videos using either fixed (Aguzzi et al., 2011) or mobile cameras (Edgington et al., 2006). Given the large variety of possible applications of imaging in benthic ecology, a flexible software, which could provide users with an assistance for the widest possible set of all applications is however clearly still missing.

One of the aims of the JERICO project (http://www.jerico-fp7.eu/) was to develop the use of image analysis techniques to monitor a large set of biological components and processes recorded either at high frequency and/or over large scales using automated or semi-automated procedures. In this context, we report here on the AVIExplore software that provides a unique flexible environment for automated video analysis. AVIExplore proposes different interfaces to analyze videos originating from imaging devices set on mobile and/or fixed platforms. The principal features of AVIEplore are: (1) to provide image selection tools allowing for the division of videos in homogeneous subsections, (2) to allow for the automatic extraction of targeted information, (3) to propose solutions for long-term time-series as well as large scale image acquisition, (4) to allow for real time acquisition and in some cases real time analysis, (5) to provide a large range of customized image-analysis possibilities through a script editor. This paper presents the principles and the structure of the AVIExplore software. Its capacities are also illustrated via the presentation of several case studies for which it has already been used. These include an: (1) automated search for coral colonies in a video recorded using a mobile imaging platform, (2) automated behavior identification of *Upogebia pusilla* within the sediment column of a thin aquarium based on a video recorded using a fixed device, and (3) automated assessment of temporal changes in the filtering activity of a passive suspension feeding benthic species based on a video of the sediment surface recorded with a fixed device.

2. AVIExplore software description

2.1. Generalities and common features

AVIExplore is a software that allows acquisition and analysis of standardised AVI videos. It presents a graphical user interface (Fig. 1) that allows access to three different modules:

(1) AVIExplore - Mobile. This module allows the extraction of information from videos taken with moving sensors and cameras. It has two working modes: real time and recorded video.

(2) AVIExplore - Fixed. This module allows for the survey of activity on surfaces by using videos taken with a fixed camera. It has two working modes: real time and recorded video.

(3) ScriptEdit. This module can be seen as a tool that allows writing and testing scripts in view of their use in the other two AVIExplore modules. It is however a standalone module that can be used to compute data from videos or images.

The three modules of AVIExplore use a similar internal structure (Fig. 2). Images are extracted from video sources; each incoming image being called ‘Origin Image’. The result of applying an operation (for example contrast enhancement) on an ‘Origin Image’ is saved as a ‘Work Image’. If the user needs to apply a second operation, he can either save it as ‘Secondary Work Image’ or rewrite on the initial ‘Work Image’. In order to speed up operations, it is possible to work with greyscale images, which are then saved as a ‘Grey Image’. Operations can be bidirectional as a ‘Work image’ can become an ‘Origin Image’. However, in this case, this transformation is not reversible as the return to the initial ‘Origin image’ is impossible. A mask is a tool used to select a subset of pixels in an image. These masks can be directly drawn by the user or computed using a script. Binary hexadecimal masks are used as internal structures to store intermediate information. They can be saved and loaded using an external file (*.msk). Other Regions Of Interests (ROIs) can also be drawn by the user and saved

in a different external file (*.roi). ROIs can be converted in internal binary regions for computations.

External files are similar within the three modules. They gather different types of information (Fig. 3): (1) the AVI input file (main input), (2) script files to process images (processing), (3) ROI files to store working areas (service), (4) mask files to get fast mask loading (service), (5) data and companion files to store intermediate results (information), (6) time and position files which can be of interest for image tracking collected with mobile carriers (information), and (7) export information in text files.

The graphic user interfaces of each of the three modules share a similar structure with: (1) a displayed window, (2) a strip menu, and (3) a scrollbar for video timing. However, depending on the module, the functionalities present on the left side of the window may differ (Fig.4).

AVIExplore can be used for different purposes namely: (1) image acquisition, (2) testing of image-analysis procedures and image analysis *per se* (Fig. 5). The user may consider AVIExplore real time acquisition mode (which is present within both the AVIExplore - Fixed and the AVIExplore - Mobile modules) for acquiring a video to be analyzed with another module. During testing, different functions can be tried out in order to optimize and automatize the image-analysis procedure, which will then be applied to a whole acquisition sequence or a batch of acquisition sequences. The AVIExplore - ScriptEdit module indeed allows for saving and then loading back any of the so-tested scripts

2.2. The AVIExplore Real Time acquisition mode

Real time acquisition can be achieved for both mobile and fixed platforms through AVIExplore - Mobile and AVIExplore - Fixed modules. The main advantages of monitoring real time acquisition using AVIExplore are: (1) an absolute control of image frequency acquisition, (2) the possibility for real time video treatments, which includes

the possibility of applying different filters to incoming images as well as the use of masks and the selection of objects. The user may then choose to directly save results and no longer original images, which can prove valuable when using acquisition memory limited devices. There is however a clear limitation for real time analyses: the flow of incoming images cannot be interrupted. Processing time must therefore remain less than the time lag between consecutive images. As a consequence, computing loops online is not possible. This can be avoided by saving the original video and analyzing it afterwards.

2.3. The AVIExplore – Mobile module

AVIExplore - Mobile handles both the video to be analyzed and a geolocalization file, which is used to locate each image and associated identified objects in space. Sophisticated Remotely Operated Vehicles (ROVs) usually produce this file when acquiring images but other mobile acquisition carriers do not. If the information concerning the location of the images is available through another way (e.g. a Log book), the user can generate a geolocalization file by himself. If not, the user can still proceed with image-analysis but in this case, identified objects will not be geographically located. The corresponding format for the geolocalization file is specified on Table 1.

The first step when analyzing a video is to extract its images (Fig. 6A). When using AVIExplore, it is recommended to first proceed with a global scene analysis. This procedure extracts features (Table 2) from the images and generates a companion file containing relevant metadata, which are used for a fast comparison between images. This process allows for the identification of common: (1) properties, and (2) objects between images. It can be used to define sections of the original video where the presence of the same specific object is suspected and which therefore can be processed using the same image analysis procedure (script). The values of the extracted features can be exported on a text file for post-treatment. Object detections can be done either on this module or on the Script Editor module. The section on

AVIExplore - ScriptEdit module describes the type of extracted information that can be exported.

2.4. The AVIExplore - Fixed module

This module allows for surveying biological activity based on videos collected using a fixed carrier either *in* or *ex situ*. The analysis of a fixed video starts by extracting its images, which are then subtracted between each other (Fig. 6B). Three different types of subtraction are proposed: (1) 'Previous', (2) 'Gap', and (3) 'Reference image'. When the 'previous' type is selected, the subtraction is carried out between all pairs of consecutive images. When the 'Gap' type, the subtraction is carried out between all pairs of images separated by a fixed number of images. When the 'Reference image' type is selected, the subtraction is always carried out between the current and a fixed reference image (which usually corresponds to the start of the recording and/or to a zero level of activity). Subtraction results can be expressed in three different modes: (1) absolute values, (2) positive values and (3) negative values. Both the type of subtraction and its combination with other parameters (Table 3) must be adjusted by the user depending on image quality and desired analyses, although some default values are proposed. The user may choose to focus on some areas of the images by drawing a ROI or a mask. Image subtractions are then only achieved within the selected area and no longer in the whole image. At the end of the analysis a 'Data File' is created and loaded. The 'Data File' is a binary file that can be understood as a metadata companion file which allows the user to directly visualize the subtraction results. On this module, the user can choose to export those results obtaining two different text files. The first file includes for each image the position (barycenter) and size (number of pixels) of each isolated region where the subtraction result is bigger than the specified threshold. The second file provides the mean position (barycenter and standard deviation) and the total size (total number of pixels) of all the regions where the subtraction result is bigger than the specified threshold found on an image. The section describing the AviExplore - ScriptEdit module provides more examples of the extracted information that could be exported.

2.5. The AVIExplore – ScriptEdit module

The AVIExplore – ScriptEdit module allows writing and testing scripts, which can later be used in another AVIExplore module. However the AVIExplore – ScriptEdit module is also a standalone program that can proceed with the complete analysis of a video and produce a result file. Available functions and commands that can be used within this module can be divided into:

(1) Video management and visualization functions.

(2) Image processing operations. Some of these operations allows for: color filtering, image segmentation, edge detection, image and histogram equalization, morphological processing and noise removal.

(3) Looping. Set of commands that allow applying one operation to a set of images.

(4) Export and log. Functions that allow either to have a log or a result text file.

(5) Masks and ROIs. Set of functions allowing the use of masks and ROIs within a script. Those masks and ROIs can either be the result of an image processing operation or designed by the user.

The detailed description of the high number of functions available is provided in the AVIExplore manual, which is embedded in the software and available online (http://www.epoc.u-bordeaux.fr/index.php?lang=fr&page=eq_ecobioc_aviexplore).

Depending on the scope of the analysis, dedicated searches must be carried out to identify/classify objects. The user can choose which information to export, some examples are: the number of objects on an image, the surface and the perimeter of each object, the width and the length of the minimum bounding rectangle of an object, the position of each object, the mean and standard deviation of all the positions and surfaces of all the objects on an image.

The ScriptEdit module proposes a functionality to help the user write scripts. This functionality provides the users with the parameters required by all specific functions. The current version of the software includes examples of object detection and

identification routines (needed for the mobile module) to help the user building their own scripts and to provide insights on which kind of operations are relevant to obtain specific information. Scripts can be saved within a Microsoft access database and then become easily accessible for subsequent analyses. The AVIExplore – ScriptEdit module has a complementary utility for video edition, including: (1) cropping, (2) reducing duration, (3) reducing resolution, (4) changing frame frequency acquisition, (4) extracting images, and (5) building a video from extracted images.

3. Case studies

3.1. AVI-Explore – Mobile: Mapping of deep corals

3.1.1. General context

In the last decade live deep-water corals have been reported in the Ionian Sea at the Santa Maria de Luca (SML) Province (Taviani et al., 2005; Tursi et al., 2004). The most reported predominant species in this area are *Lophelia pertusa* and *Madrepora oculata,* both species present a multi-branch morphology that can provide ecological niches, refuges and substrates for a multitude of other species (Tursi et al., 2004). The SML Province contains living and dead isolated colonies of those deep-water corals as well as coral patches (Savini et al., 2005; Vertino et al., 2010). Different tools have been used to identify and characterize both macrofauna and habitats within the SML province (Mastrototaro et al., 2010; Tursi et al., 2004, Savini and Corselli, 2010, Taviani et al., 2005).

In this context, video imagery acquired with ROVs has been used to detect macrohabitat and determine main seafloor features that includes the percentage of recognizable dead and/or live coral colonies (Vertino et al., 2010). This last analysis was achieved but at a low spatial resolution and apparently by direct visual video observation (Vertino et al., 2010). We report here on the use of the AVIExplore software to analyze another video collected in the same area. Our specific aims

consisted in automatically: (1) detecting corals, and (2) differentiating between apparently dead and living coral patches.

3.1.2. Materials and methods

The analyzed video was recorded with a remotely operated vehicle (ROV) *Victor-6000* during the MEDECO cruise, which took place aboard the French R/V *Pourquoi Pas?* This ROV was equipped with a SONY DXC990 3 CCD camcorder (resolution of 720x576 pixels, frame rate of 25fps, duration 2 hours and 5 seconds). The study area covers 0.022 km². The video contained a total of 180,133 images. Because of the deployment of an automatic flash device, overexposed images were removed every 298 images. A total of 179,529 images were thus considered for the analysis. The video was processed using the AVIExplore – Mobile module. A region of Interest (ROI) was created to exclude imbedded text from the analysis (Fig. 7). Two different scripts were executed to assess the surface of: (1) apparently dead and living corals within each image (Table 4A), and (2) only apparently living corals within each image (Table 4B). The main feature used to determine living corals was its white-color (Fig. 8). A visual assessment was completed on a subsection (1330 images) to validate the so-obtained results. Images were then visually classified as: images with corals and images without corals. A comparison between the two assessment modes (visual and automatized via AVIExplore) was achieved. Total accuracy was computed as the number of images for which both classification modes were similar divided by the total number of compared images. An analysis of the types of errors and their frequency of occurrence was also carried out. False positive errors corresponded to the fact that AVIExplore classified an image as containing corals whereas the same image was visually classified as not containing corals. False negative errors corresponded to the fact that AVIExplore classified an image as not containing corals whereas the same image was visually classified as containing corals.

3.1.3. Results

The dissimilarity in the Grey-Level Co-occurrence Matrix (GLCM) computed from each image on the video was computed during the global scene analysis (Table 2). This allowed us to identify (Fig. 7) a continuous sequence of images featuring high dissimilarity of their GLCM. The direct visualization of those images showed that they were recording the water column and no longer the water-sediment interface. They were thus discarded and only 161,000 images were considered for the identification of corals. An example of the results of the two executed scripts is shown in Fig.8. These results were combined with the geolocalization file allowing for the mapping of apparently dead and apparently living coral colonies (Fig. 9). Agreement between images classified by the software and visually was achieved for 90.2% of the 1330 tested images. False positive and false negative error frequencies were 5.4 and 4.3%, respectively. Main sources of false positive errors were the presence of: (1) other megafauna, (2) fishes and (3) suspended sediment. Negative errors were mainly due to small corals and/or bad light conditions. Also, in some cases, corals were not so clearly visible but the human operator tended to infer their existence because the same corals could be seen on the previous image.

3.2. AVIExplore – Fixed: Identification and quantification of different types of behaviors in the mud shrimp *Upogebia pusilla*

3.2.1. General context

Burrowing crustaceans are among the most efficient bioturbators inhabiting marine soft sediments (Pillay and Branch, 2011). These organisms dig large galleries deep (up to 1m) in the sediment column and therefore rework large amount of particles. They also strongly increase the rate of exchanges between the sediment column and the overlying water through the ventilation of their burrows (Coelho et al., 2000; Webb and Eyre, 2004). High frequency time series of ventilation by burrowing invertebrates have been collected both *in situ* using pressure sensors (Volkenborn et al., 2012; Wethey and Woodin, 2005; Wethey et al., 2008) and *ex situ* using planar optodes (Volkenborn et al., 2007; Volkenborn et al., 2010; Volkenborn et al., 2012). Corresponding studies however remain largely

descriptive since ventilation is highly dependent on animal behavior, which is very difficult to monitor in burrowing crustaceans because these organisms spend most of their time in their burrow. Up to now the description or the identification of different types of behaviors of burrowing crustaceans has only been achieved visually (Coelho et al., 2000; Dworschak, 1987; Stamhuis et al., 1996). The establishment of a quantitative relationship linking activity and sediment reworking, such as the one recently established by Maire et al., (2007d) for sediment reworking by the bivalve *Abra ovata*, is therefore still lacking for these bioturbators in spite of their potentially major role in the functioning of some coastal ecosystems. Here, we report on the use of AVIExplore to automatically assess and quantify different behaviors by the mud shrimp *Upogebia pusilla*, which is common within the seagrass meadows of the Arcachon Bay (France). Our study is based on a video of a lateral view of a flat aquarium recorded in the laboratory.

3.2.2. Materials and methods

Our experiment was carried out in a large flat aquarium (40 × 25 × 2 cm) filled with 25 cm of natural sieved (0.5 mm mesh) sediment overlaid with 15 cm of aerated seawater. The sediment was left to settle for 48 h in the aquarium and the mud shrimp was introduced 1 week prior the beginning of the video recording the experiment to fully establish its burrow. The video (duration: 13h 20min, frequency of image acquisition: 0.1Hz, resolution: 2560 × 1920 pixels) was then recorded under infra-red light using a µEye camera (IDS®) controlled by a computer. The visual analysis of this movie allowed for the identification of several types of behaviors, namely: 'Resting/ventilating', 'Burrowing' and 'Walking'. During 'Resting/ventilating', the mud shrimp remains at the same location within its burrow and its movements are restricted (e.g. to abdomen contractions and pleopod beating). During 'Burrowing', the mud shrimp moves within its burrow but only over a limited distance and it occasionally displaces sediment. During 'Walking', the mud shrimp moves within its burrow over large distances. During the present study, we used the AVIExplore – Fixed module to automatically analyze the video and to quantify these three behaviors. A ROI corresponding to the visible part of the

burrow was first drawn (Fig.10A). Each image was then subtracted from the preceding one and pixels featuring a positive differences higher than 30 (threshold value on an overall scale between 0 and 255 set after preliminary trials) were identified. For each pair of consecutive images, the AVIExplore – Fixed module generated: (1) the number of pixels within the ROI differing between the two images, and (2) the barycenter of those pixels. A signal treatment combining these two pieces of information was then defined to assess the behavior of the mud shrimp at each given time of the monitoring (Fig. 11). If the shape of the signal related to the number of pixels modified between two images is flat or consists either in a single peak or consecutive isolated peaks, then the attributed behavior for the corresponding images is 'Resting/Ventilating'. If the signal has another shape, the range of positions of related pixels is used to differentiate between 'Burrowing' (small range) and 'Walking' (large range).

The results were compared with those originating from a visual analysis of the video. Accuracy was assessed as the proportion of images which were attributed the same behavior by the two procedures. An error analysis is carried out to understand the sense of miss classified behaviors.

3.2.2.1. Results

Temporal changes in the occurrence of the three types of behaviors are shown in Fig. 10B. The mud shrimp spent most of its time (i.e., 70.1%) 'Resting/ventilating', versus 26.1% 'Burrowing' and only 3.7% 'Walking'. These figures can be compared with those originating from the visual observation of the video: 67.5% 'Resting/ventilating', 30.2% 'Burrowing', and 2.3% 'Walking'. The total accuracy was 82.7% with higher discrepancies between the two methods corresponding to confusion between 'Burrowing' with either 'Resting/ventilating' (Table 5). Except for 'Resting/ventilating', we observed that there was a lag of a few images between the end of each period of time corresponding to a given behavior as recorded visually and using AVIExplore. This discrepancy was attributed to differences in sensitivity between the sensor and the eye of the operator. When adjusting this lag for 'Burrowing' and 'Walking', there was only a

slight improvement in the overall accuracy (83.7%). The existence of these lags therefore cannot be considered as the main source of discrepancies between automated and visual analyzes. Besides this general agreement, the analysis of the time series derived from automated and visual assessments clearly showed some difficulties in assessing positive 'Walking' and to a lesser extent positive 'Burrowing' behaviors. The proportions of agreement between automatically and visually detected positive behaviors were indeed 89.2, 75.2 and only 13.1% for 'Resting/ventilating', 'Burowing' and 'Walking', respectively.

3.3. AVIExplore – Fixed: Quantification of filtering activity in the passive suspension-feeding polychaete *Ditrupa arietina*

3.3.1. General context

The composition of littoral benthic macrofauna show major temporal changes in the Gulf of Lions apparently in relation with climatic oscillation such as the North Atlantic Oscillation (Grémare et al., 1998a; Grémare et al., 1998b) and the Western Mediterranean oscillation (Bonifácio, 2015). Within the Littoral Fine Sands community (Labrune et al., 2008), the polychaete *Ditrupa arietina* is by far the species featuring the largest changes in abundances with densities between 0 and more than 11,000 individuals.m^{-2} (Grémare et al., 1998b). This species may have an impact on carbon fluxes through: (1) the construction of its calcareous tusk-shaped tube (Medernach et al., 2000), and (2) nutrition. The corresponding impact depends on the intensity of filtering activity. Jordana et al. (2000) have therefore described and quantified the filtering activity of this species using a software specifically designed. During the present study, we use AVIExplore to analyze one of their videos and to show the ability of this new software to quantify the filtering activity of *D. arietina* in a completely automatized way.

3.3.2. Materials and methods

The experimental setup is described in (Jordana et al., 2000). Briefly, worms were collected by dredging and kept in the laboratory in tanks filled with a thin layer

of well-sorted fine sand supplied with running ambient sea water. During the experiment per se, 15 worms were geometrically positioned upward, with tube openings, 1 cm above the sediment surface (Fig. 12A) in a 60.3 x 40.3 x 20 cm aquarium filled with a 10cm thick layer of well sorted fine sand and 30 l of filtered seawater. Polychaete worm filtering activity was monitored during 9 h using a CCD-ICS059 AI video sensor (VLSI Vision Ltd®) with an image frequency acquisition of 0.017 Hz. The video was analyzed using the AVIExplore – Fixed module. Fifteen ROIs were drawn close to the shadow of the extremity of each tube, where the openings of gills crowns were most visible (Fig. 12B). A fixed reference image with all gill crowns fully retracted was subtracted from each current images. 'Dark' pixels featuring negative differences lower than -30 (threshold fixed after preliminary trials) were identified and considered as indicative of gill crown deployment. They were summed for each ROI and the corresponding data were exported as text files. Filtering activity was defined as the ratio of the current number of 'dark' pixels divided by the maximum number of 'dark' pixels recorded at any given time within the corresponding ROI. Assuming, that maximal extension of the gill crown was achieved for all 15 worms during the observation period, this index allows for direct comparisons between worms irrespective of their size and position within the recorded field.

Validation was achieved on one of the 15 specimens and accuracy was assessed as the proportion of images for which there was an agreement between visual and AviExplore assessment.

3.3.3. Results

Temporal changes in the filtering activity of each of the 15 studied worms are shown in Fig. 12C. The general pattern and the degree of cycle were clearly worm dependent. The level of synchronicity between individual worms was low since the number of simultaneously filtering worms varied between 2 and 13 and did not follow any clear pattern (Fig. 13A). The cumulated time devoted to filtration also

varied strongly between individuals as indicated by the fact that, by the end of the 9 h monitoring period, the time spent filtering by each worm was between 1 h 33 min and 7 h 10 min. (Fig. 13B). Overall, these results and conclusions are in full agreement with those of Jordana et al. (2000).

Accuracy was 98.3%. A systematic error has been identified; the filtering activity of the analyzed specimen started one image before when visually assessed.

4. Discussion

The increase of the use of imaging in benthic ecology has resulted in a high diversity of image acquisition modes (including mobile and fixed platforms as well as ex and in situ deployments) and image characteristics (stability or variability in illumination and image quality). Image analysis, especially when applied to video clearly constitutes a bottleneck when using imaging for ecological studies. Another possible flaw of this technique is that it may prove highly operator-dependent due to the complexity of the information: (1) contained, and (2) to be extracted from each image (Cuvelier et al., 2012). Software developments may clearly help overcoming these two drawbacks. At present such developments have been mostly customized to specific acquisition procedures and ecological applications. Along this line, and within JERICO, our own research group has recently produced the SPIArcBase software, which is specifically dedicated to the processing of sediment profile images (Romero-Ramirez et al., 2013). The aim of the development of the AVIExplore is much more ambitious. The goal is to provide a single platform flexible enough to automate the image analysis procedures requested by the largest possible sets of image acquisition platforms and ecological implications. In order to do so, we have followed a three-pronged approach: (1) building two modules specifically designed to process the video acquired with mobile and fixed image acquisition platforms and to tackle associated ecological questions, and (2) providing a module specifically designed to elaborate and test new specific image analysis procedures, which could prove necessary to tackle specific ecological questions, and (3) leaving the software open to new developments through the

possibility of including new libraries depending on their compatibility with the AVIExplore. The three case studies carried out aimed at showing the real flexibility of the AVIExplore software. The following discussion will show some of the advantages of having used the AVIExplore software based on the experiments gained during these trials.

4.1. The AVIExplore – Mobile module

Edgington et al. (2006) emphasized the fact that the potential of ROVs and underwater observatories for collecting long-term observations is constrained by the time and effort to view and quantify the data. They thereby underlined the need for new software developments for processing videos obtained from mobile platforms. A first difficulty when processing those videos is that they often cover different habitats, which may correspond to very different images, which may therefore require different image analysis procedures. Applying all of those procedures to the whole video may rapidly become time consuming and could possibly lead to significant errors as well. AVIExplore therefore offers the possibility of defining sections of videos that are composed of similar images, and therefore can be processed using the same script. This is based on image comparison achieved on the Grey-Level Co-occurrence Matrix. Moreover, this procedure may prove useful even when a single habitat is monitored. This was the case during the coral mapping case study, where it facilitated the reduction of the number of analyzed images from 179,529 to 161,000 (i.e., by 10%) of the images to analyze. The reason for that was that the video recorder was no longer recording the sediment water interface but the water column by the end of the video. We strongly believe that in the future, this procedure will also prove useful in defining section of videos characterized by dissimilarities due to various characteristics including: light conditions, image quality levels, angle of view, contrast, clarity, and presence of objects.

The AVIExplore – Mobile module also presents a high potential for assessment of benthic (mega)fauna. The visual assessment of benthic epibenthos is indeed often restricted to presence/absence data or to operator-dependent semi-quantitative data,

which led Cuvelier et al. (2012) to plea for the development of automated image analysis procedures. A difficulty then consists in correcting the surfaces within the image to account both for the altitude and the inclination of the camera. During the 'Coral mapping' case study, the surface of corals was only quantified as pixel² because the information regarding both sensor altitude and inclination were lacking. Should such information be available during future uses, we would recommend to proceed to appropriate corrections (e.g., as briefly described in Robert and Juniper, 2012) on the raw surface data exported by AVIExplore. Along the same line, during the 'Coral mapping' study, we did not account for possible overlap between images. Here again, this could be handled through appropriate post-treatments (e.g. according to Morris et al., 2014) of AVIExplore exported raw surfaces.

4.2. The AVI – Explore – Fixed module

The AVIExplore – Fixed module gathers the experience gained by our research group through the developments of pieces of software specifically designed to process videos in view of assessing movements and activities of several benthic invertebrates and communities (Bernard et al., 2012; Duchêne et al., 2000; Duchêne and Queiroga, 2001; Duchêne and Rosenberg, 2001; Grémare et al., 2004; Hollertz and Duchêne, 2001; Maire et al., 2007a; Maire et al., 2007c). In its present form the AVIExplore – Fixed module mainly involves image subtraction procedures, which can first be achieved between the current image and a fixed reference image. This procedure is especially appropriate for the quantitative assessment of activity as was for example the case in the 'Quantification of filtering activity in the passive suspension-feeding polychaete *Ditrupa arietina*' case study. However, when used alone, it is clearly limited in the recognition of different kinds of behavior, which would require a time-series analysis of the so-recorded activity, such as achieved during the mud shrimp case study or by Grémare et al. (2004) for categorizing inhalant siphon activity by the deposit-feeding clams *Abra ovata* and *A. nitida*.

The AVIExplore – Fixed module offers a much more straightforward possibility to assess movements and to categorize activities. It consist in achieving subtraction between each pair of consecutive (or separated by a constant lag) images. This procedure was used during the 'Identification and quantification of different types of behaviors in the mud shrimp *Upogebia pusilla*' case study. It generated a limited number of parameters including the distance of the barycenters of the pixels modified between two consecutive pairs of images, which proved efficient in attributing three different types of behavior to the mud shrimp *Upogebia pusilla*. However, we believe that the use of other features and/or machine learning algorithms may result on better classification results.

One clear limitation of the proposed method was that we were not able to clearly distinguish between 'Resting' and 'ventilating', which may result from a too low: (1) frequency acquisition (0.1 Hz) relative to the beating frequency of pleopods (0.5 Hz, LP personal observation), and (2) number of considered movement characteristics. Regarding this last point, a complexity of the number and characteristics of considered parameters (i.e., a holistic feature extraction approach) may certainly prove very useful as it has already been the case in several other imagery fields (Madhvanath and Govindaraju, 2001; Rodriguez et al., 2007; Turk and Pentland, 1991).

In its present forms the AVIExplore – Fixed module allows for the assessment of the trajectory of isolated objects/organisms through the plots of the position of consecutive barycenters of modified pixels identified by image subtraction. Such a capacity has proven useful in the assessment of swimming capacities in pelagic larvae of benthic organisms (Duchêne and Nozais, 1994; Nozais et al., 1997). Benthic ecologists are nevertheless more and more interested in the study of individual movements occurring within groups of objects/organisms. Bernard et al. (2012) for example developed a specific application to monitor the individual displacement of fluorescent sediment particles (luminophores). This requires the elaboration of a complex research algorithm (involving both the analysis of distances and directions of movements between consecutive pairs of images), which are currently not included in the AVIExplore – Fixed

module. Depending on the interaction with future users, we may integrate such an approach within later version of AVIExplore.

4.3. The AVI – Explore – ScriptEdit module

The Avi-Explore – ScriptEdit module allows for the analysis, processing and visualization of videos and images. In its current version, it includes a large set of functions for analyzing individual images. As demonstrated during the present study, it already constitutes a powerful tool that can be used for different purposes, e.g.,: detection and quantification of organisms (see the Deep corals case study) and the detection/quantification of different kinds of activities. Other applications can be already foreseen such as: (1) the analysis of swimming (fishes and benthic invertebrate larvae) patterns, and (2) the study of specific processes such as the dynamics of benthic settlement and early recruitment.

As it stands, the Avi-Explore – ScriptEdit module nevertheless does not allow for the quantitative assessment of some specific processes. As an example, recent imaging applications in benthic ecology have included the 2D analysis of sediment reworking, which requires the establishment of vertical profiles of luminophores concentrations. The first step of this procedure consists in defining the water-sediment interface, which can either be achieved manually or (Romero-Ramirez et al., 2013). Its second step consists in flattening this interface (Maire et al., 2006), which consists in transferring its position to the first pixel row of each pixel column. At present, none of the corresponding procedures are included in the AVIExplore – ScriptEdit module. Here again, and depending on interactions with future users, we may integrate them within a later version of AVIExplore.

Acknowledgements

This work was carried out within the JERICO Integrated Infrastructure Intiative project (grant agreement n°262584). It was also supported by the BIOMIN project (LEFE CYBER and EC2CO-PNEC). We would like to thank Ifremer for providing access to the video of the MEDECO cruise used during the 'Coral mapping' case study. Ifremer is not responsible for the use of this video. All rights reserved. Alicia Romero was financially supported by the French Muséum d'Histoire Naturelle. Ludovic Pascal was supported by a doctoral grant of the French Ministère de l'Enseignement Supérieur et de la Recherche (ED – 304 Science et Environnement).

Figure Captions

Fig. 1. AVIExplore graphical user interface: (1) AVIExplore – Mobile module, (2) AVIExplore – Fixed module, (3) AVIExplore – ScriptEdit module, (4) summary of system characteristics, (5) extension files for future versions, and (6) software manual and information.

Fig. 2. Internal structure of AVIExplore. Origin, Work and Secondary Work images are 24 bit bitmaps for color images and video frames. Grey images are 8 bit images. Regions Of Interest (ROIs) and Masks are structures saved in external files, which can be loaded on demand. Op: Operation

Fig. 3. External files generated by AVIExplore. Continuous arrows refer to files that must to be loaded by the user. Dashed arrows refer to files automatically loaded by the software.

Fig. 4. AVIExplore – Mobile, AVIExplore – Fixed and AVIExplore – ScriptEdit module's general graphic user interface: (1) video display window, (2) strip menu, (3) video time scrollbar, and (4) area differing between modules.

Fig. 5. Interactions between different modules and modes of AVIExplore.

Fig. 6. A: AVIExplore – Mobile: Typical flowchart of a video obtained with a mobile device, B: AVIExplore – Fixed: Typical flowchart of a video obtained with a fixed device.

Fig. 7. Coral mapping case study: dissimilarities of Grey-Level Co-occurrence Matrix for each image of the video. Numbered vertical dashed lines indicate different images which are shown on the right. 1 and 2: images of the water-sediment interface, 3: image of the water-sediment interface at high altitude and 4: image of the water column.

Fig. 8. Coral mapping case study: results of the execution of the two scripts on a single image. (A) Original image showing the delimitation of ROI in yellow, (B) image with white overlay covering apparently dead and living corals, and (C) image with a red overlay covering only apparently living corals.

Fig. 9. Coral mapping case study: Mapping of (A) apparently dead and living corals, and (B) apparently living corals

Fig. 10. Mud shrimp case study: (A) Example of an image extracted from the video with an ROI corresponding to the delimitation of the visible part of the burrow, (B) Automatic and (C) Visual assessment of the temporal changes in the occurrence of the three monitored behaviors: 'Resting/ventilating', 'Burrowing' and 'Walking'

Fig. 11. Mud shrimp case study: Combination of AVIExplore extracted features to assess the three different behaviors of *Upogebia pusilla*.

Fig. 12. Passive suspension-feeder case study: (A) Reference image showing the disposition of the 15 studied worms with their retracted gill-crowns, (B) Localization of the 15 ROIs around the shadow of the top extremity of the tubes where extended gill-crowns (10 worms) can be observed, (C) Temporal changes in individual filtering activities.

Fig. 13. Passive suspension-feeder case study: (A) Temporal changes in the number of worms filtering (B) Temporal changes in individual cumulated filtering activities.

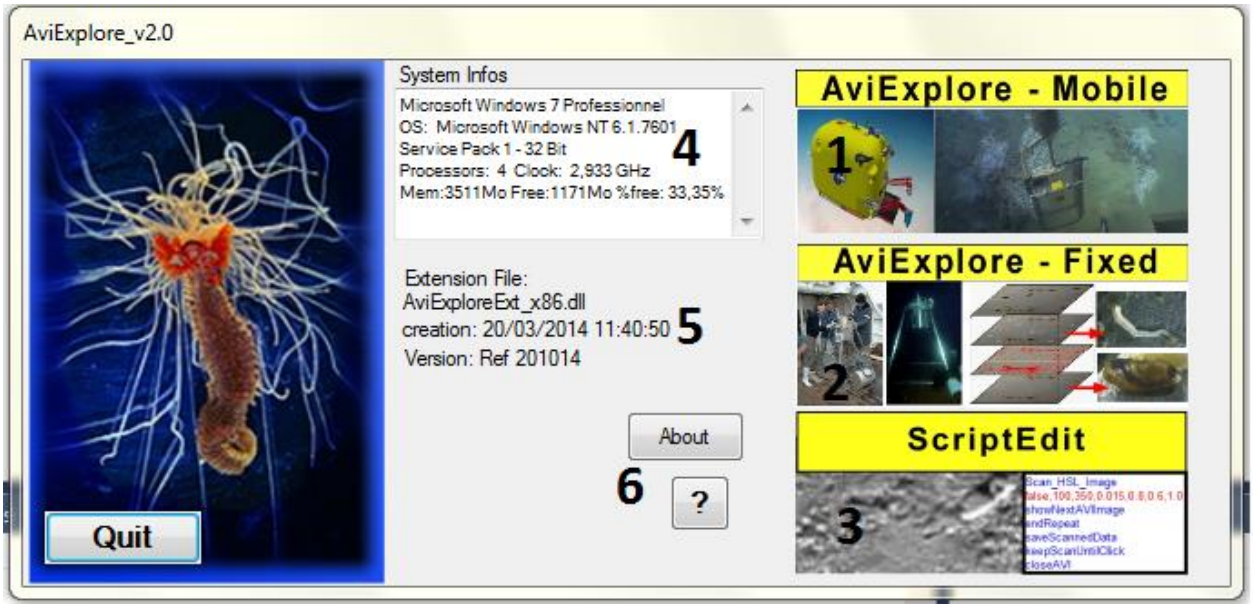

AviExplore_v2.0
Quit
System Infos
Microsoft Windows 7 Professionnel
OS: Microsoft Windows NT 6.1.7601
Service Pack 1 - 32 Bit
Processors: 4 Clock: 2,933 GHz
Mem:3511Mo Free:1171Mo %free: 33,35%
4
Extension File:
AviExploreExt_x86.dll
creation: 20/03/2014 11:40:50
5
Version: Ref 201014
About
6
?
AviExplore - Mobile
1
AviExplore - Fixed
2
ScriptEdit
3

Fig. 1

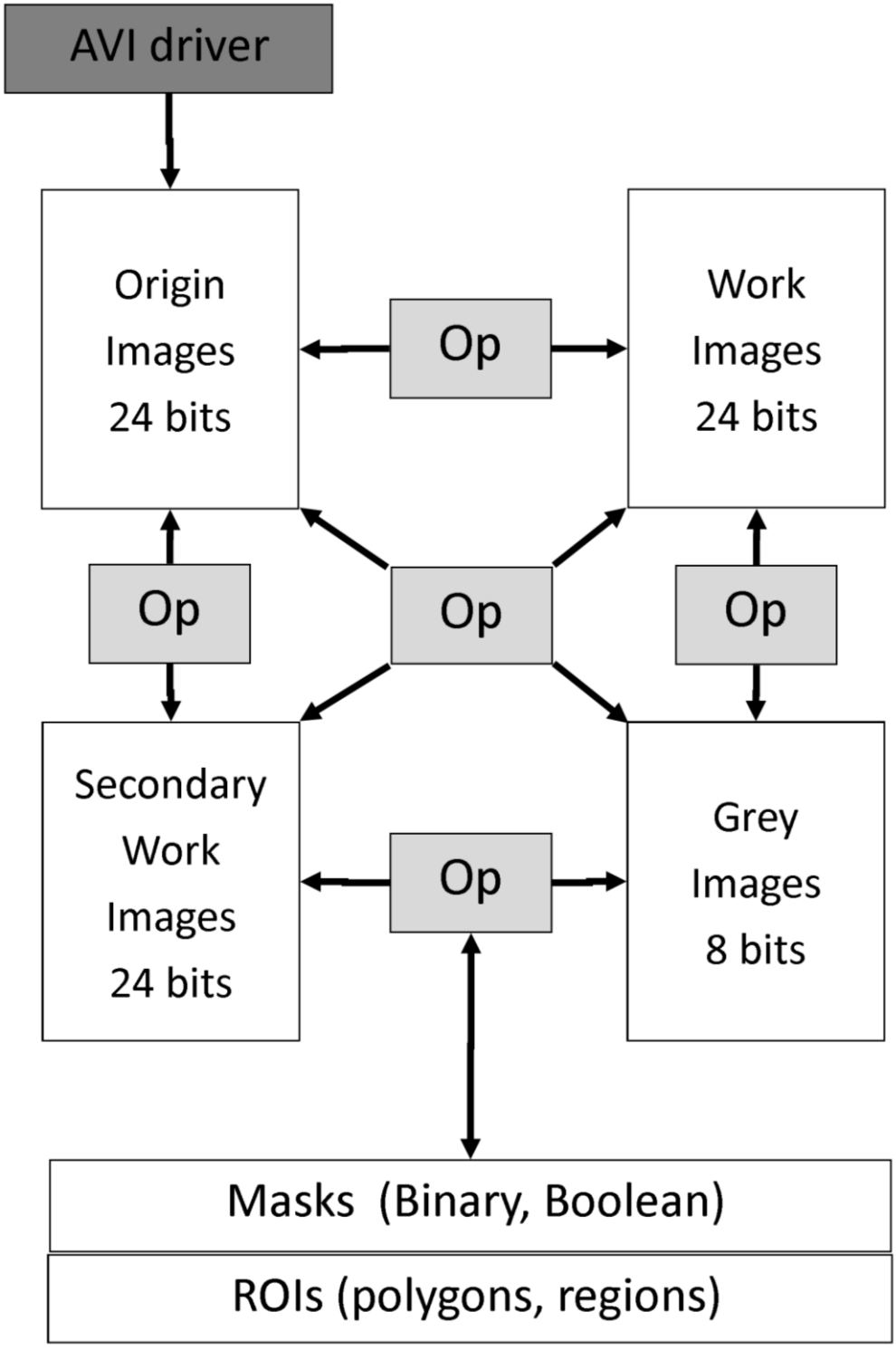

AVI driver
Origin
Images
24 bits
Op
Work
Images
24 bits
Op
Op
Op
Secondary
Work
Images
24 bits
Op
Grey
Images
8 bits
Masks (Binary, Boolean)
ROIs (polygons, regions)

Fig. 2

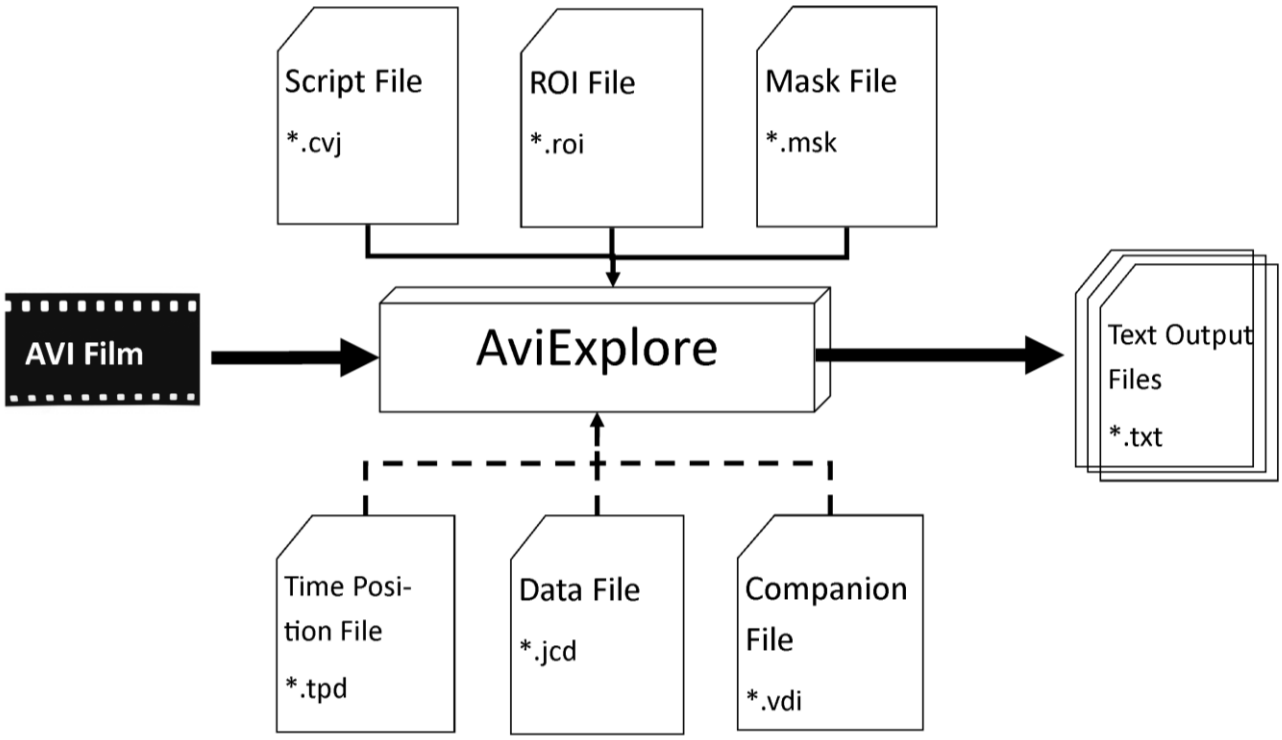

Script File
*.cvj
ROI File
*.roi
Mask File
*.msk
AVI Film
AviExplore
Text Output Files
*.txt
Time Posi-tion File
*.tpd
Data File
*.jcd
Companion File
*.vdi

Fig. 3

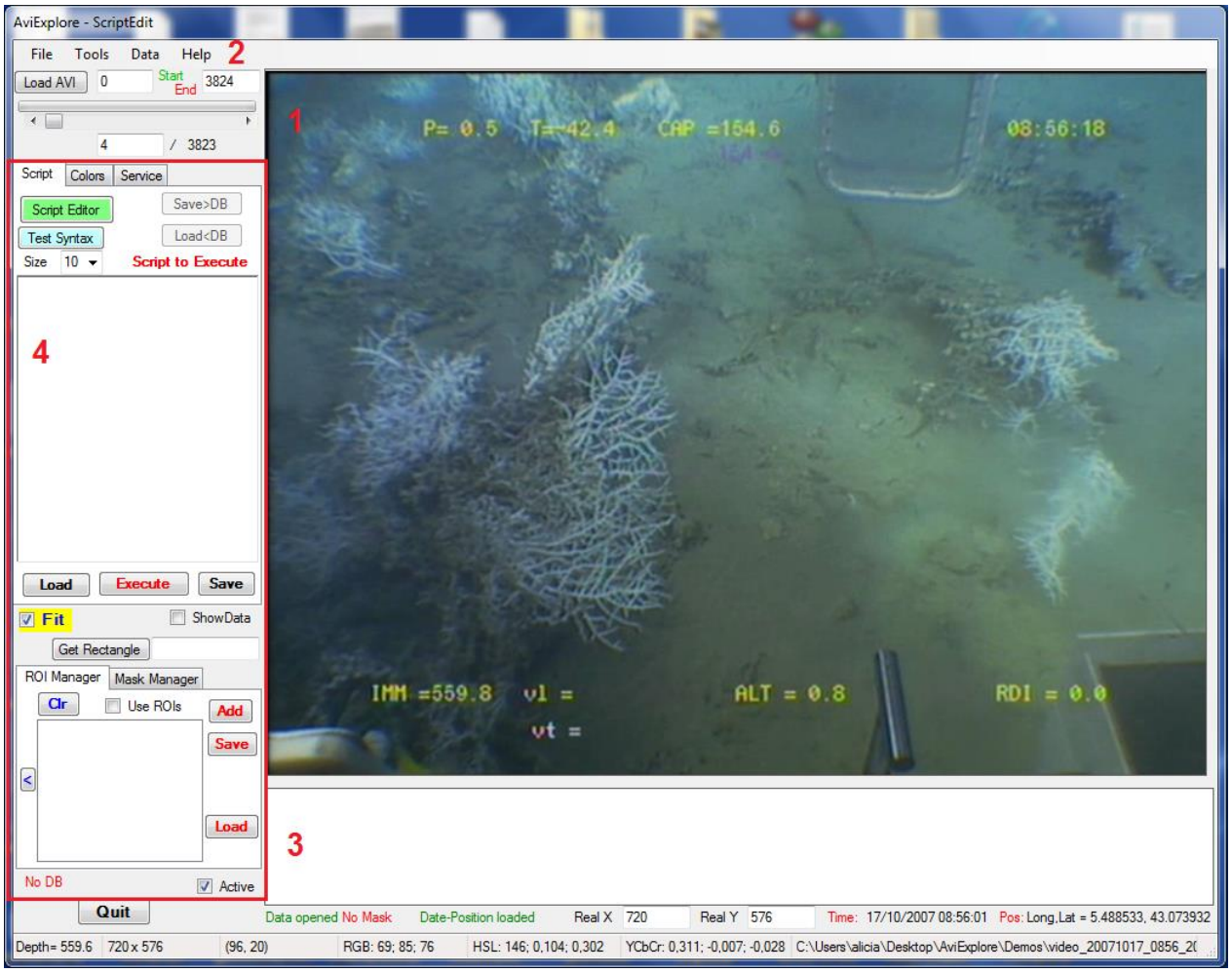

AviExplore - ScriptEdit
File
Tools
Data
Help
2
Load AVI
0
Start
End
3824
4
/ 3823
Script
Colors
Service
Script Editor
Save>DB
Test Syntax
Load<DB
Size
10
Script to Execute
4
Load
Execute
Save
Fit
ShowData
Get Rectangle
ROI Manager
Mask Manager
Clr
Use ROIs
Add
Save
Load
No DB
Active
Quit
1
P= 0.5
T=42.4
CAP =154.6
08:56:18
IMM =559.8
vl =
vt =
ALT = 0.8
RDI = 0.0
3
Data opened No Mask
Date-Position loaded
Real X
720
Real Y
576
Time: 17/10/2007 08:56:01
Pos: Long,Lat = 5.488533, 43.073932
Depth= 559.6
720 x 576
(96, 20)
RGB: 69; 85; 76
HSL: 146; 0,104; 0,302
YCbCr: 0,311; -0,007; -0,028
C:\Users\alicia\Desktop\AviExplore\Demos\video_20071017_0856_2(

Fig. 4

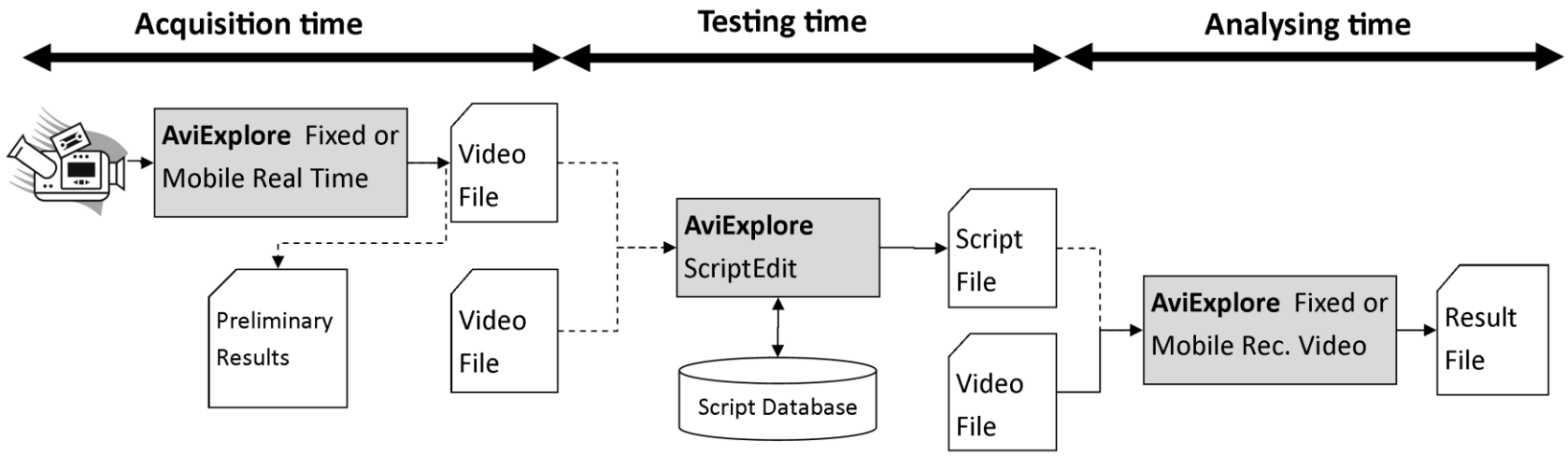

Acquisition time
Testing time
Analysing time
AviExplore Fixed or Mobile Real Time
Video File
Preliminary Results
Video File
AviExplore ScriptEdit
Script Database
Script File
Video File
AviExplore Fixed or Mobile Rec. Video
Result File

Fig. 5

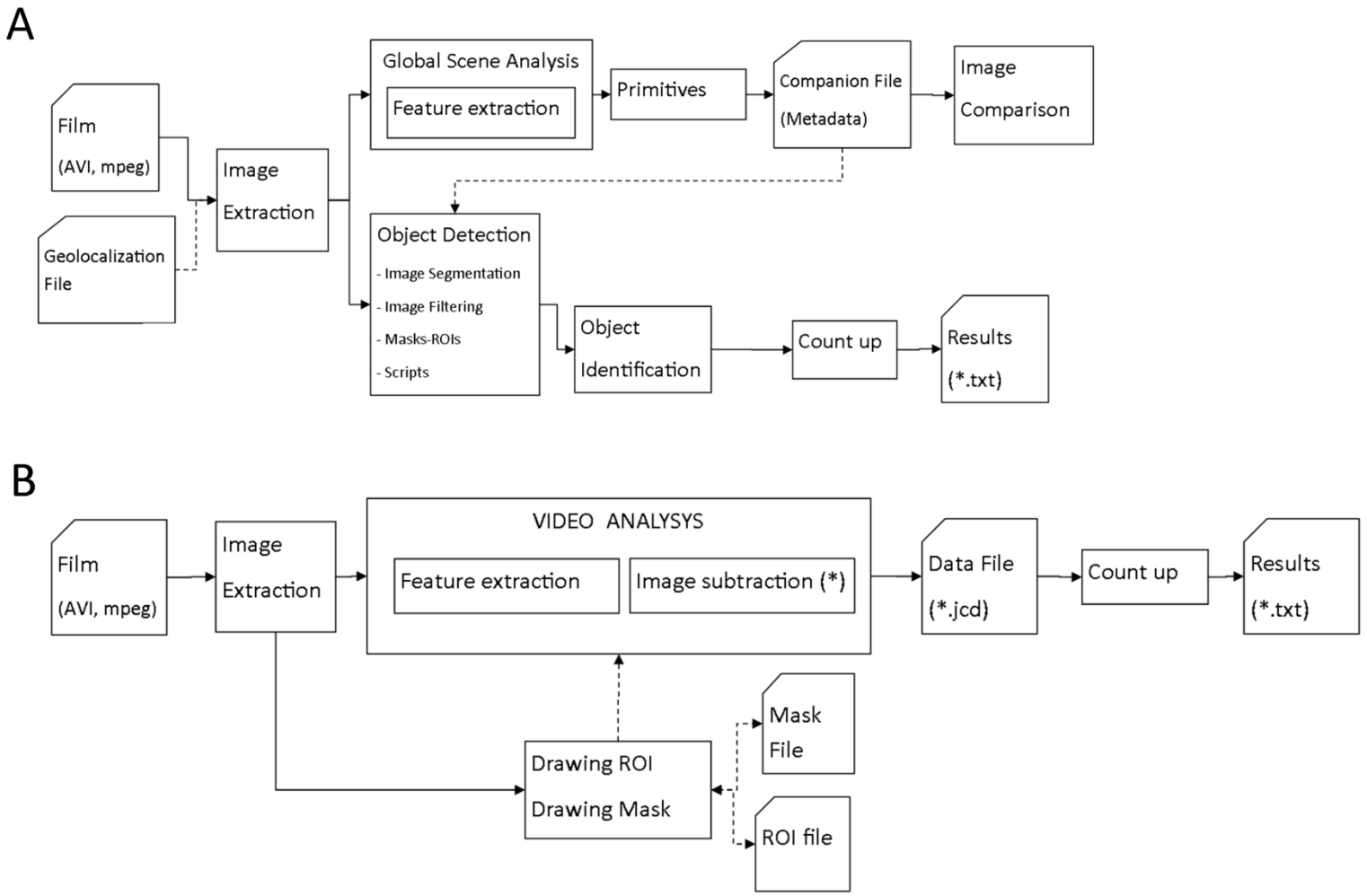

A
Film
(AVI, mpeg)
Geolocalization
File
Image
Extraction
Global Scene Analysis
Feature extraction
Primitives
Companion File
(Metadata)
Image
Comparison
Object Detection
- Image Segmentation
- Image Filtering
- Masks-ROIs
- Scripts
Object
Identification
Count up
Results
(*.txt)
B
Film
(AVI, mpeg)
Image
Extraction
VIDEO ANALYSYS
Feature extraction
Image subtraction (*)
Data File
(*.jcd)
Count up
Results
(*.txt)
Mask
File
Drawing ROI
Drawing Mask
ROI file

Fig. 6

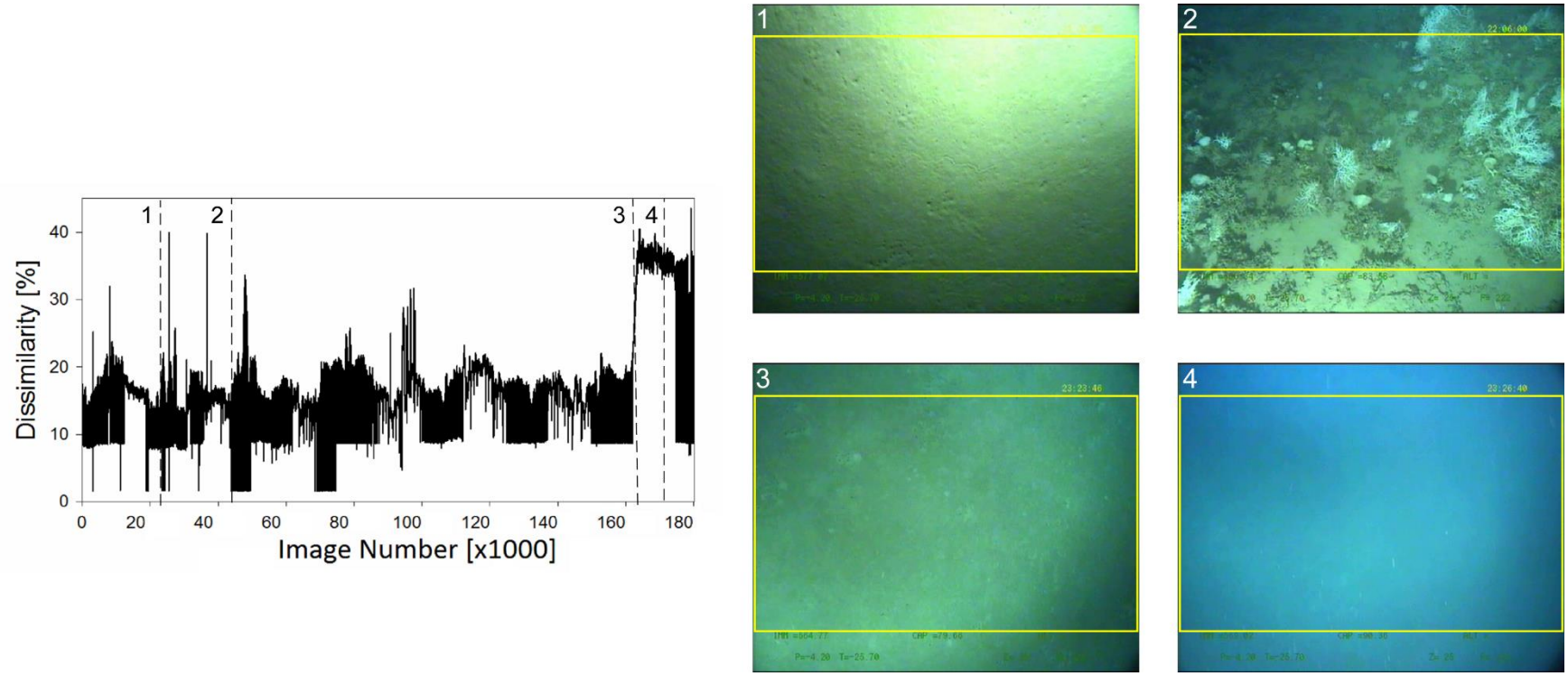
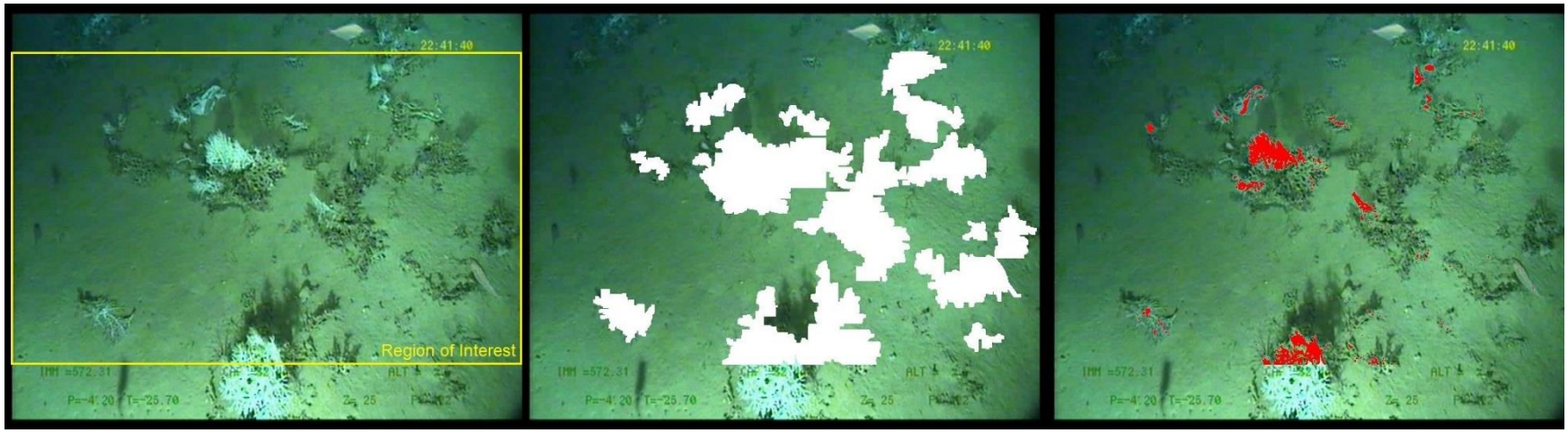

1
2
3
4
Dissimilarity [%]
0
10
20
30
40
0
20
40
60
80
100
120
140
160
180
Image Number [x1000]

Fig. 7


22:41:40
Region of Interest
IMM =572.31
ALT
P=-4.20 T=-25.70
Z= 25
22:41:40
IMM =572.31
ALT
P=-4.20 T=-25.70
Z= 25
22:41:40
IMM =572.31
ALT
P=-4.20 T=-25.70
Z= 25

A B C

Fig. 8

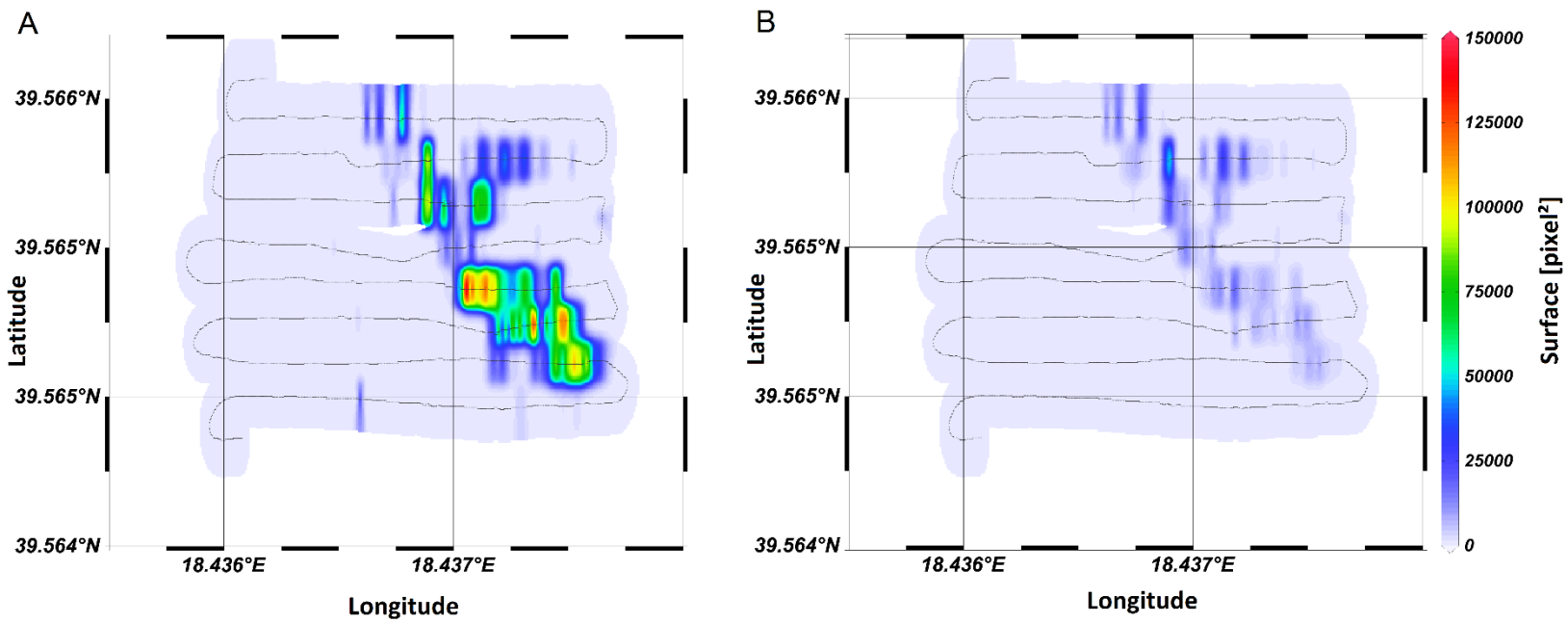

A
B
39.566°N
39.565°N
39.565°N
39.564°N
Latitude
18.436°E
18.437°E
Longitude
150000
125000
100000
75000
50000
25000
0
Surface [pixel²]

Fig. 9

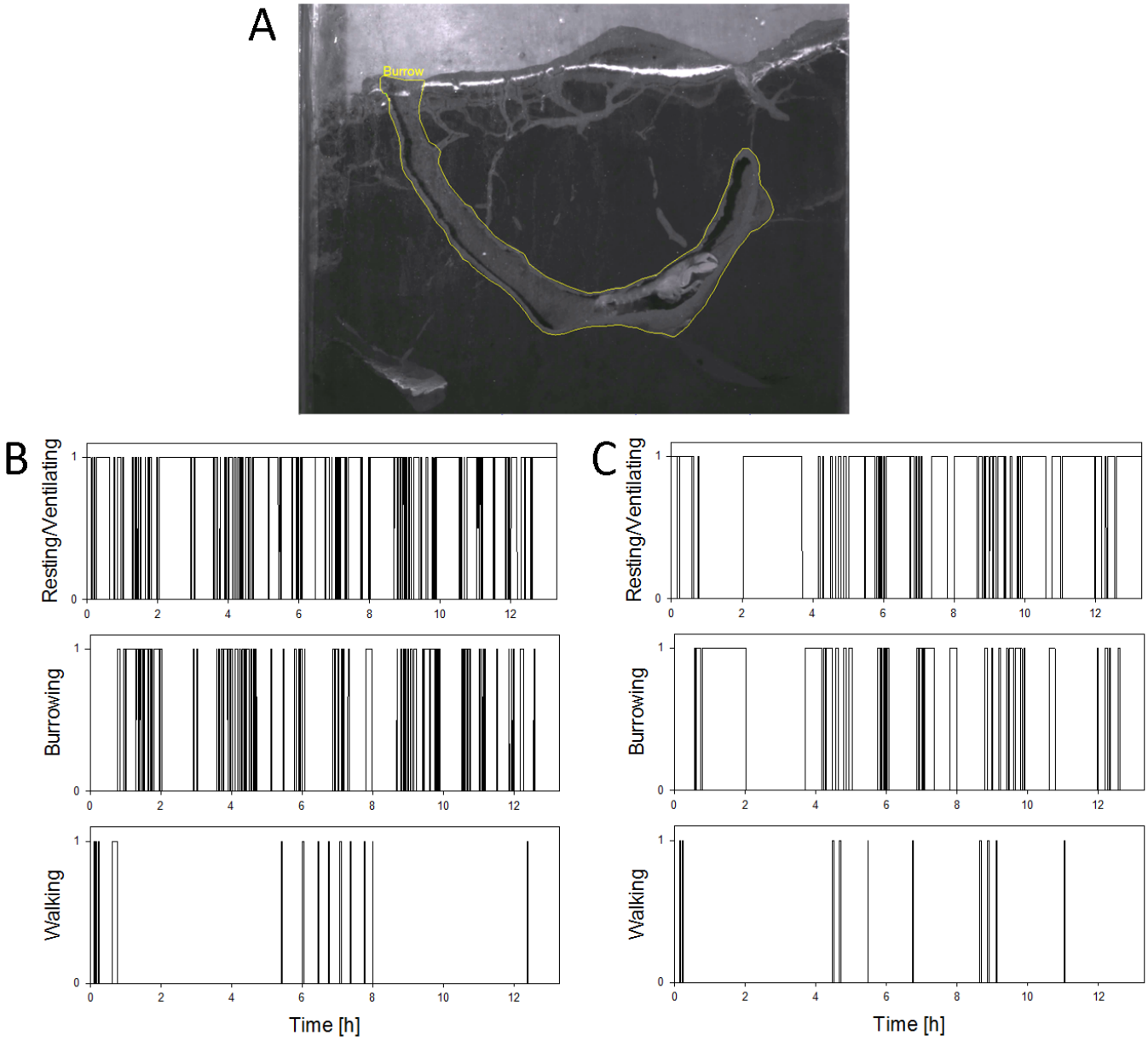

A
Burrow
B
Resting/Ventilating
Burrowing
Walking
Time [h]
C
Resting/Ventilating
Burrowing
Walking
Time [h]

Fig.10

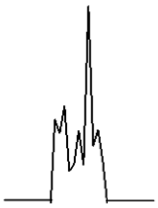
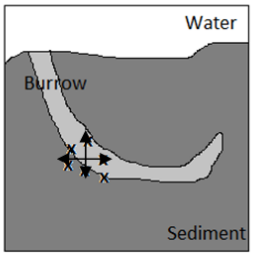
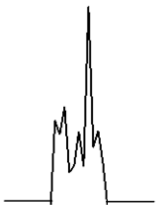
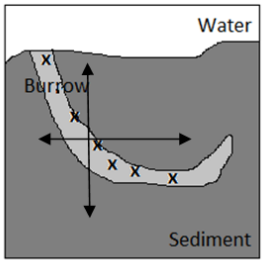
Extracted Information from film		Asociated Behaviour
Shape of signal (Number of pixels modified between two consecutive images)	Range of position (Barycenter)	
	Zero	Resting or ventilation
	Zero	Resting or ventilation
	Small	Resting or ventilation
	Small 	Burrowing
	Large 	Walking

Fig. 11

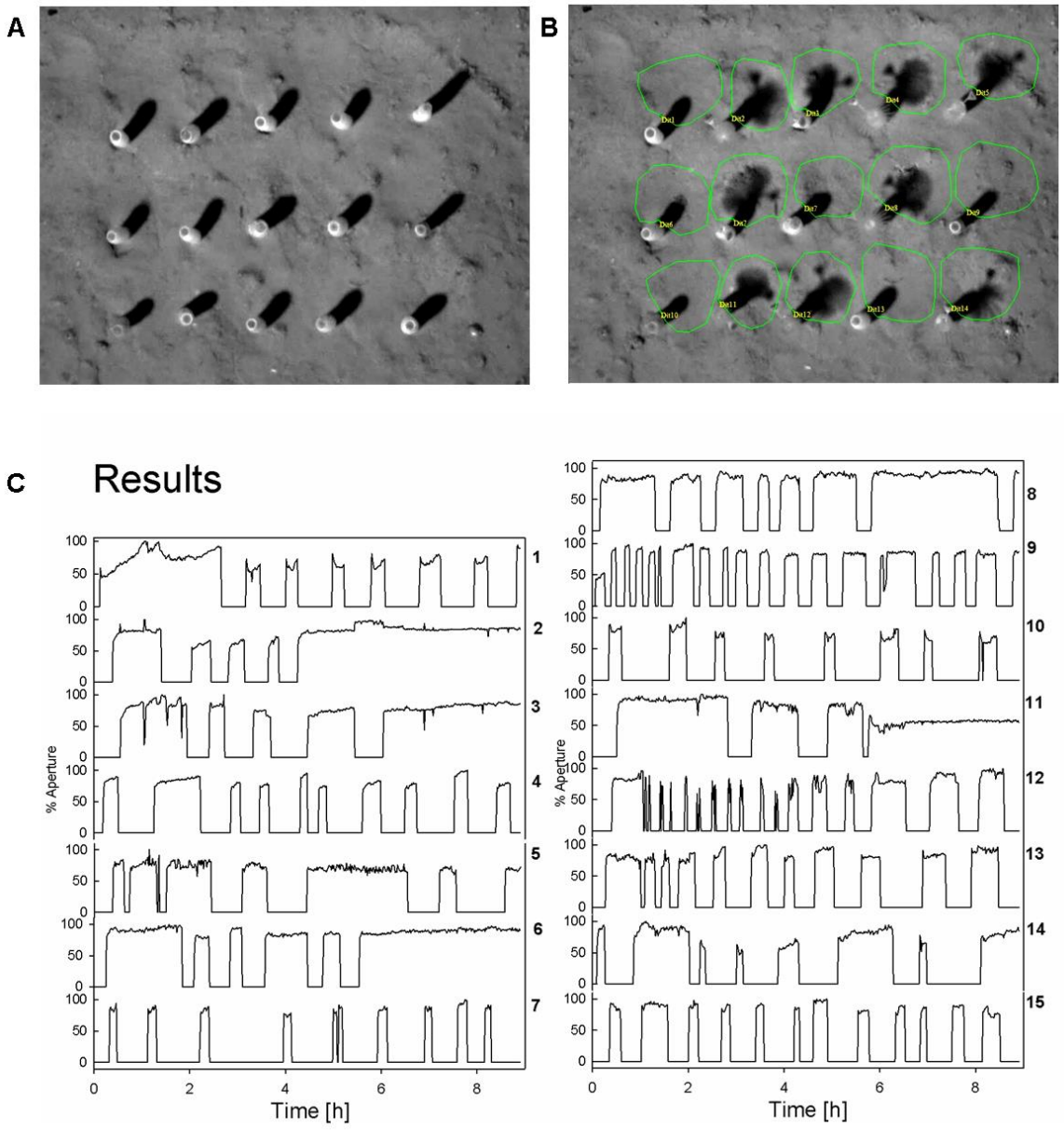


Fig. 12

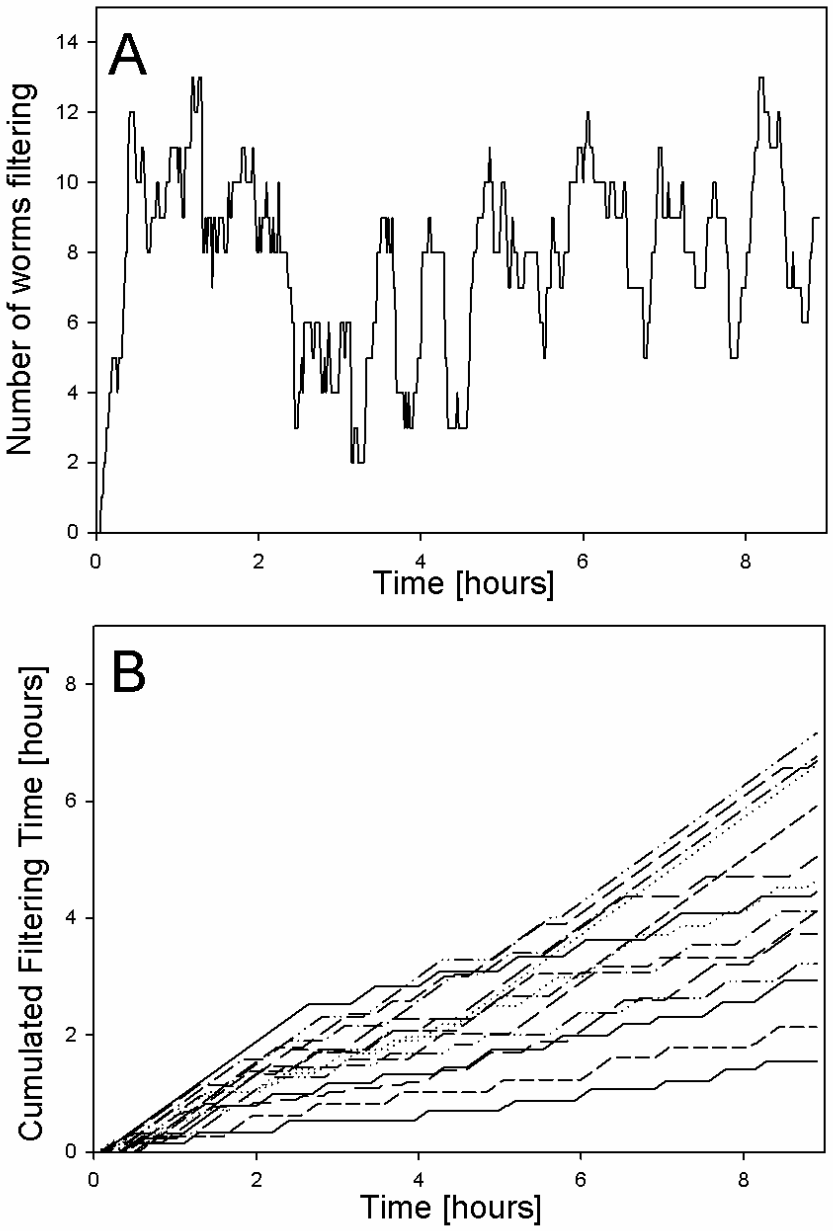

A
Number of worms filtering
Time [hours]
B
Cumulated Filtering Time [hours]
Time [hours]

Fig. 13

Table 1. Format of Geolocalization File: (1)Explanation, (2) Example

Number of images	1340			
Date and time [mm/dd/yyyy hh:mm:ss.ss]	10/14/2007 21:35:04.10			
Complementary information	Device info			
Date and time [mm/dd/yyyy hh:mm:ss.ss] Latitude Longitude Depth	10/14/2007 21:35:04.10	39,5655667	18,4361983	548
	10/14/2007 21:35:09.10	39,5655667	18,43618	548
	…			
(1)	(2)			

Table 2. List of extracted features by the AVIExplore - Mobile module. In (1-6) *i,j* are the horizontal and vertical coordinates in the Greylevel Co-occurrence Matrix, *P* is the cell value.

List of Extracted Features	**Description**
Number of objects (SURF)	The number of objects found on an image when using "Speeded Up Robust Feature" for object detection
Object size (SURF)	The size of objects found on an image when using "Speeded Up Robust Feature" for object detection
Spatial moment	Weighted average of the intensities of the image pixels
Variance	Mean variance of the greyscale image
Hu1	First Hu moment for pattern recognition (Hu, 1962)
Hu2	Second Hu moment for pattern recognition (Hu, 1962)
Angular second moment (GLCM)	Also known as uniformity, it is the sum of squares of entries in the **greylevel co-occurrence matrix (GLCM)** (1): $\sum_{i,j=0}^{N-1} P_{i,j}{}^2$
Energy (GLCM)	Square root of the angular second moment
Entropy (GLCM)	Amount of information of the image needed for the image compression, it represents spatial disorder (2): $\sum_{i,j=0}^{N-1} -P_{i,j} \ln P_{i,j}$
Contrast (GLCM)	Measure of the intensity contrast between a pixel and its neighbor over the whole image. Sum of squares variance in the GLCM (3): $\sum_{i,j=0}^{N-1} P_{i,j} (i-j)^2$
Dissimilarity (GLCM)	Measure that defines the variation of grey level pairs (pixel and its neighbor) in an image (4): $\sum_{i,j=0}^{N-1} P_{i,j} \lvert i-j \rvert$
Inverse Difference Moment (GLCM)	Also called homogeneity. Measures of the closeness of the distribution of elements in the GLCM to the GLCM diagonal (5): $\frac{\sum_{i-1}^{N-1}\sum_{j-1}^{N-1} P_{i,j}}{1+(i-j)^2}$
Mean_i (GLCM)	The mean based on the reference pixels (μ_i)
Mean_j (GLCM)	The mean based on the neighbor pixels (μ_j)
Variance_i (GLCM) Variance_j (GLCM)	Measures the dispersion around the mean, of values within the GLCM (σ_i,σ_j)
Correlation (GLCM)	Measure of how correlated a pixel is to its neighbor over the whole image (6): $\sum_{i,j=0}^{N-1} P_{i,j} \left[\frac{(i-\mu_i)(j-\mu_j)}{\sqrt{\sigma_i^2+\sigma_j^2}}\right]$
Hue	The Hue histogram
R	The red channel histogram
G	The green channel histogram
B	The blue channel histogram
Grey	The greylevel histogram

Table 3. List of parameters to select for analyzing when using the AVIExplore - Fixed module.

Parameters to adjust	Description
Diff Mode	Type of subtractions
diffType	Expression of subtraction
Th	Threshold for subtraction difference
Min Sz	Minimum size of search objects
Max Sz	Maximum size of search objects
Use ROI	Use of a drawn region of interest
Use Mask	Use of a drawn Mask
Gap	Number of images between subtraction Reference image number

Table 4. Coral mapping case study: Details of the two scripts used to: detect, quantify the surface, visualize and save each analyzed image. For (A) apparently living and dead corals, and (B) apparently living corals.

A) Living and dead corals	B) Living corals
openLogFile *true* repeat *0,NbImg* getRawHSLMask *80,200,true,110,350, 0.1,1.0,0.1,1.0* CopyOriginToBinBuf DilateMask *binBuf* eraseMaskBlobs *1,600,100000* fillBinBufHoles *140* reloadCurAVIImage ShowMask *Origin,binBuf,0* CountPixelsInMask *1* WriteOneINTTextValue saveJpeg *(DirectoryFolder\),100* showNextAVIImage *1* endRepeat closeLogFile	openLogFile true repeat 0,NbImg getRawHSLMask *50,200,true,110,350, 0.1,1.0,0.1,1.0* CopyOriginToBinBuf DilateMask *binBuf* eraseMaskBlobs *1,300,100000* fillBinBufHoles *140* reloadCurAVIImage BinarizeRGB *100,255,150,255,150,255, 1,1,1,0,0,true,Mask,Origin,true* CountPixelsInMask *0* WriteOneINTTextValue saveJpeg *(DirectoryFolder\),100* showNextAVIImage *1* endRepeat closeLogFile

Table 5. Mud shrimp case study: Frequency of occurrence of the different types of discrepancies between the visual and the automated analysis.

		Visual Analysis		
		Resting or ventilating	Burrowing	Walking
Automated Analysis	‘Resting/ ventilating’		7.9 %	0.8%
	‘Burrowing’	4.1 %		1.2 %
	‘Walking’	2.0%	1.1 %	

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