Image processing techniques to improve characterization of composite ichnofabrics

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Abstract

Image processing techniques, including the Analyze Particles tool offered by Fiji software and the Intensity Profile by ICY (IP-ICY), were applied in core and outcrop examples to improve characterization of autocomposite ichnofabrics. Analyze Particles gives information about particle shape and size in the studied image. This tool was applied to Chondrites assemblages in composite ichnofabrics in view of selected images of modern marine hemipelagic cores from Site U1385 of IODP Expedition 339. Differences in size, relative abundance, density and distribution of Chondrites were interpreted as related to variations within the population of Chondrites tracemakers. Intensity Profile quantifies pixel values of the infilling material of traces, proving helpful to discriminate between specimens, evaluate the horizon of colonization, and approach the penetration depth. Its application to the analysis of Zoophycos improves characterization of composite ichnofabrics from deep-sea pelagic calcilutites of the Petra Tou Romiou section (Eocene Lefkara Formation; southern Cyprus). Several suites of Zoophycos were interpreted as associated with different phases of colonization from several horizons. Moreover, it was possible to discriminate structures pertaining to several specimens, as opposed to those from the same specimen.
**Keywords:** Composite ichnofabrics; image treatment; Fiji and ICY; core and outcrops

**Introduction**

In 2015, vol. 85 of *Annales Societatis Geologorum Poloniae* was dedicated to our colleagues Richard Bromley and Ulla Asgaard, including contributions presented in the Symposium ONE ICHNOLOGY (Bornholm, 14th-16th May, 2014) in honor of Richard and Ulla. During the Symposium and in the volume of ASGP published in its aftermath, we presented digital image treatment as a novel method for improving ichnofabric analysis in cores (Rodríguez-Tovar and Dorador, 2015). The applied methodology allows one to differentiate between biodeformational structures and trace fossils, meaning better identification and discrimination of ichnotaxa, observation of cross-cutting relationships, quantitative estimation of the percentage of bioturbation associated with each ichnotaxon, the whole ichnocoenosis, or a complete ichnofabric, as well as enhanced evaluation of the depth of penetration of a particular tracemaker and an approach to the tiering structure (Dorador and Rodríguez-Tovar, 2014, 2015, 2016a, b; Dorador et al., 2014a, b).

In the years since, this novel method has shown itself to be a powerful tool in numerous studies of modern marine cores, involving paleoenvironmental reconstructions, ocean-atmosphere dynamics and sedimentary basin analysis (e.g., Rodríguez-Tovar et al., 2015a, 2015b; Zeeden et al., 2015, 2017; Dorador et al., 2016; Hodell et al., 2017), mainly through improved ichnofabric characterization (see Dorador and Rodríguez-Tovar, 2018, for a recent review). However, as acknowledged in earlier papers (Ekdale and Bromley, 1983; Bromley and Ekdale, 1986), the analysis of composite ichnofabric is no easy matter given the complexity of the multitiered
macrobenthic tracemaker community, with continuous burrowing during sediment accretion.

Recently, two image processing techniques—Fiji and ICY—were evaluated in terms of their usefulness for ichnological analysis involving high to low contrast images from core and outcrop examples (Miguez-Salas et al., 2018). Fiji and ICY (+ Fiji) stand as rapid and valuable methods to quantify the whole of the bioturbated surface. The *Intensity Profile* plugin by ICY (IP-ICY) is useful for estimating penetration depth, as well to differentiate between specimens; moreover, its semi-automatic character favors faster calculations (Miguez-Salas et al., 2018).

The aim of the present paper is to assess the complexity of macrobenthic tracemaker communities producing composite ichnofabrics using new image processing techniques to look into ichnofabric features. Information on tracemakers, phases of colonization, and tiering structure can be gathered through an automatic evaluation of the pixel analysis. The technique allows for characterization of size and roundness of traces (i.e., *Chondrites*), assignation of different biogenic structures to a single or to different specimens of the same ichnotaxa (i.e., *Thalassinoides* and *Zoophycos*), evaluation of penetration depth, and quantification of bioturbation. The method was applied to core and outcrop examples.

**Composite ichnofabrics**

Composite ichnofabrics (Ekdale and Bromley, 1983; Bromley and Ekdale, 1986) refer to those generated by the superimposition of different (successive) suites of biogenic structures (Ekdale et al., 2012). The development of composite ichnofabrics (Ekdale and Bromley, 1983) may reveal the progressive upward migration of a single,
tiered benthic community during continuous sediment accretion, or the successive occupation of the sediment by multiple communities of organisms in response to autogenic or allogenic changes in environmental conditions within a depositional system. Accordingly, Savrda (2016) defined two main types of composite ichnofabrics: autocomposite ichnofabrics in reference to the first mechanism, characterized by a single ichnocoenosis, and self-generated by a particular assemblage of tracemakers; and heterocomposite ichnofabrics produced by the second mechanism, comprising two or more different ichnocoenoses.

Differentiation of heterocomposite ichnofabrics can be relatively simple in that the successive (two or more) ichnocoenoses most often reflect short- to long-term paleoenvironmental changes, determining overprinting of different ichnofacies assemblages (Savrda, 2016). Elsewise, characterization of autocomposite ichnofabric can be comparatively complicated, as occurs in the case of composite ichnofabrics of shelf-sea pelagic deposits, pointed by Ekdale and Bromley (1991) as “among the most complex and difficult to decipher of all ichnofabrics, because they represent a succession of diverse, multitiered, benthic communities occupying a succession of substrate types during continuous sedimentation under variable seafloor conditions”.

**Methodology**

In the present paper, Fiji and ICY software are used in ichnofabric analysis of modern marine hemipelagic cores from the IODP Expedition 339 and outcrop deep-sea pelagic calcilutites from the Petra Tou Romiou section (Cyprus). Both image processing techniques were recently evaluated for the characterization of ichnological features, and
found to be useful as less time-consuming alternative techniques, to increase visibility of trace fossils, quantify the percentage of bioturbated surface, and estimate penetration depth (Miguez-Salas et al., 2018).

Fiji software (Schindelin et al., 2012) is an extension of the well-known ImageJ open-source software. ImageJ has been applied in geological research (e.g., Grove and Jerram, 2011; Goldstein et al., 2017), but only occasionally in ichnological analysis to enhance the visibility of certain image attributes, and to estimate bioturbated surface and shape/length measurements (Francus, 2001; Nicolo et al., 2010; Lauridsen et al., 2011; Curth et al., 2014). Fiji was recently applied in neo-ichnological analysis for format conversion in reconstructed volumes obtained by computed tomography of invertebrate burrow systems (Hale et al., 2015).

To enhance image visibility, Fiji software offers a wide range of plugins. Among them, the Contrast Limited Adaptive Histogram Equalization (CLAHE) provides for quick, enhanced image visibility. This method is based on the modification of block size, controlling the size of the local region around a pixel for which the histogram is equalized, and defines the number of histogram bins used for histogram equalization.

To quantify the bioturbated surface, a black and white binary image is necessary. After applying CLAHE, the obtained image must be transformed into an 8-bit grayscale image; this image is then converted to a new black and white binary image based on the selection of an appropriate threshold value from the 8-bit grayscale image. Before quantification, the bioturbated area can be precisely determined by applying a wide range of easy-to-use binary tools and filters (i.e., Erode, Fill holes, and Minimum/Maximum filters, among others) available in the Fiji process. Finally, manual corrections can be conducted by means of eraser or painting tools, controlling that all the selected pixels pertain to trace fossils. Afterwards, output measurement results are
quickly obtained with the Fiji analyze menu, whose program measures the percentage of black pixels, thereby offering a quantification of the bioturbated surface.

In this research we apply two different tools: Analyze Particles, for the first time, and Intensity Profile.

**Analyze Particles**

The Analyze Particles tool offered by Fiji software (Schindelin et al., 2012), allows one to obtain information (shape, size) about particles in the studied image.

Previous to the application of the Analyze Particles tool, three steps must be taken: i) Image visibility improvement with Contrast Limited Adaptive Histogram Equalization (CLAHE); ii) threshold draft and generation of black and white binary image (B/W); and iii) a filtering process and manual corrections.

After obtaining the binary B/W image, it is treated with the Analyze Particles tool for trace fossil selection in function of shape and size. Before running the plugin, two main input parameters should be delimited. The first is the minimum and maximum pixel area of the bioturbation, in order to remove small background and trace fossils that are not of interest. The second is roundness (resemblance to a circle), to exclude unwanted bioturbation. In the study case, the minimum size pixel area is 0.5 mm$^2$ and the maximum 8 mm$^2$; the roundness value selected was above 60%.

**Intensity Profile by ICY (IP-ICY)**

The Intensity Profile plugin by IP-ICY has been successfully applied previously for trace fossil penetration depth in core analysis (Miguez-Salas et al., 2018). Following
the same steps and plotting the intensity value of each pixel, the methodology was here applied for the first time in outcrop examples in order to discern not only penetration depth but also distinct trace fossil generations.

This plugin calculates the intensity value of each pixel (providing three values per pixel, one for each channel) along a particular Region Of Interest (ROI). Then, values are plotted on intensity profile graphics from a given ROI (vertical line along the core), after which the values must be manually checked and filtered to identify potential measurement errors.

Results: Composite ichnofabrics and processing techniques

Several cases involving core and outcrop examples were analyzed.

Case 1: Composite ichnofabric at modern marine hemipelagic cores from IODP Expedition 339

The Analyze Particles tool was applied to composite ichnofabrics of modern marine hemipelagic cores from Site U1385 of IODP Expedition 339 (Figs. 1, 2). Site U1385 (southwestern Iberian margin; 37°34.285’N, 10°7.562’W) is dominated by Pleistocene hemipelagic mud- and claystones. Previous analysis allowed characterization of a multi-tiered assemblage: biodeformational structures revealed colonization of the uppermost tiers, above or just below the seafloor, with Planolites, Palaeophycus and even Taenidium as upper tier traces. Thalassinoides-like/Thalassinoides occupied a middle tier, and Zoophycos and Chondrites were found
to be deep tier forms associated with the final phases of colonization (Rodríguez-Tovar and Dorador, 2014). Paleoenvironmental conditions were envisaged as presenting a major incidence of pore-water oxygen conditions and organic matter availability.

In this case, we focus analysis on the *Chondrites* tracemaker community. Taking images from Site U1385 of IODP Expedition 339 (Fig. 1), visibility was enhanced with Contrast Limited Adaptive Histogram Equalization (CLAHE), then a black and white binary image (B/W) was generated, and finally a filtering process and manual corrections were conducted (Fig. 1). The results reveal the presence of a mottled background on which *Chondrites*, together with *Planolites* and *Thalassinoides*, are seen.

The visual differentiation in size between two *Chondrites* assemblages is noteworthy. To quantify the shape and size of *Chondrites* in the two assemblages, the *Analyze Particles* tool was applied on the filtered image (Fig. 2). Based on size and roundness, the structures can be isolated and studied in greater detail. A total of 148 spots were recognized, 52 belonging to small *Chondrites* and 96 to large *Chondrites*. Small *Chondrites* correspond to specimens between 0.5 and 3 mm\(^2\) in area, whereas large *Chondrites* would be specimens in the range of 3-8 mm\(^2\) in area. Size histograms show a significant variation within the large *Chondrites*, the specimens of 3 mm\(^2\) in area being more abundant, while small *Chondrites* are more homogenous in size. The total percentage of bioturbation corresponding to *Chondrites* is 4.5%, being significantly higher for large forms (4.1%) than for small ones (0.4%). Size distribution of large *Chondrites* is more or less uniform in the studied area, while small *Chondrites* are located in patches.

*Cases 2 and 3: Composite ichnofabrics in outcrop deep-sea pelagic calcilutites from the Petra Tou Romiou section (Cyprus)*
Deep-sea pelagic calcilutites are registered in the lowermost part of the Petra Tou Romiou section (Eocene Lefkara Formation; southern Cyprus). These chalky calcilutites record a well-developed and moderately diverse trace fossil assemblage, consisting of *Chondrites* isp. (*Ch. intricatus* and *Ch. targionii*), *Planolites* isp., *Taenidium* isp., *Thalassinoides* isp., and *Zoophycos* isp., typical of the *Zoophycos* ichnofacies (Miguez-Salas and Rodríguez-Tovar, 2018). A composite chalky ichnofabric reveals a multi-tiered benthic community: the uppermost tier determines a mottled background, the upper tier shows the highest trace fossil abundance and diversity (*Planolites*, *Taenidium* and *Thalassinoides*), the middle tier features *Zoophycos* and large *Chondrites*, and the deep tier consists mainly/exclusively of small *Chondrites*. This ichnofabric has an autocomposite character, associated with bioturbation by a single ichnocoenoses and gradual upward migration of the tiered macrobenthic community as the pelagic calcilutite sedimentation slowly progresses.

To improve characterization of the composite chalky ichnofabric, the Intensity Profile plugin of IP-ICY was applied to two outcrop examples (Cases 2 and 3; Figs. 3, 4), including in both cases an upper part of greenish calcilutites and a lower (dominant) part of greenish/whitish calcilutites. In both examples, intensity profiles were conducted, from top to bottom, crosscutting different ichnotaxa, but focusing on *Zoophycos* structures.

In the first example (Case 2), two different intensity profiles were carried out (Fig. 3a, b).

The first profile (Case 2a in Fig. 3) comprises *Zoophycos*, as the dominant trace, together with *Planolites* in the upper part, and vertical burrows in the lower part. Similar intensity pixel values were obtained from the analyzed *Planolites*, vertical
burrow, and most of the traces of *Zoophycos*, in the range of those values corresponding to the upper greenish calcilutite. The values are significantly lower than the ones corresponding to the host greenish/whitish calcilutites. Considering that the bioturbation horizon for these traces is within the upper greenish calcilutite, the penetration depth values of *Planolites*, *Zoophycos* and the vertical structure would be at least 2 cm, 3-6 cm, and 7 cm, respectively. Only in the case of *Zoophycos* registered at the lower part are the intensity pixel values intermediate between those of the greenish and the greenish/whitish calcilutites, similar to those observed in the upper part of the greenish/whitish calcilutites. If we assume that this structure comes, in fact, from the upper part of the greenish/whitish calcilutites, a penetration depth of 6 cm could be estimated.

The second profile (Case 2b in Fig. 3) consists exclusively of several specimens of *Zoophycos*, as revealed by the cross-cutting relationships observed. All the analyzed *Zoophycos* structures have similar intensity pixel values, in the range of those values corresponding to the upper greenish calcilutite, irrespective of the penetration depth (minimums of 2 cm, 4 cm, 5 cm and 7 cm).

In the second example (Case 3, Fig. 4), the intensity profile includes two specimens of *Planolites* and *Zoophycos* and one of *Thalassinoides*. Similar intensity pixel values are obtained from the lower structure of *Zoophycos*, the two specimens of *Planolites*, and the burrow of *Thalassinoides*, within the range of the values corresponding to the upper greenish calcilutite, allowing assignation of minimum penetrations depths of 4.5 cm, of 5 cm and 6 cm, and of 8.5 cm, respectively. Especially significant, however, are the higher values of the upper structure of *Zoophycos*, observed between 2 and 3 cm, very similar to those from the host greenish/whitish calcilutites.
Discussion

The examples described here underline the usefulness of the *Analyze Particles* and *Intensity Profile* tools to improve characterization of autocomposite ichnofabrics and shed light on the complexity of the associated macrobenthic tracemaker community. Tiering structures, as reflecting development of the tracemaker community, can be addressed by depth of penetration, cross-cutting relationships, and vertical variations in ichnological features. In some cases this is not an easy matter, but it can be facilitated by means of the proposed methodology.

Case 1 (Figs. 1, 2) is an illustrative example, featuring a detailed study of *Chondrites*, which is a common trace fossil of deep-sea environments, and a significant component of the composite ichnofabrics. As presented in the paper of Ekdale and Bromley (1991) and latterly corroborated in a number of research studies, the lower tiers of composite ichnofabrics are mainly characterized by the presence of *Chondrites* of different sizes (small, medium and large), the size decreasing with depth. Thus, the dimensions of *Chondrites* prove to be an ichnological feature of major interest in composite ichnofabrics. The size of the tunnels in *Chondrites* is moreover considered indicative for the differentiation of ichnospecies (Fu, 1991; Uchman, 1999; Knaust, 2017). Accordingly, a detailed quantification of size of the burrows of *Chondrites* can improve characterization of the tracemaker community and composite ichnofabric analysis. Different *Chondrites* populations can be characterized by a quantification of respective sizes, relative abundance, and distribution. Large *Chondrites* (3-8 mm² in area) show a higher abundance and more or less homogeneous distribution in the
studied area, while small *Chondrites* (0.5-3 mm² in area) are less abundant and distributed in patches. Variations in size could be related to ontogenetic changes in the population or to different tracemakers. Larger *Chondrites* are dominant in number and occupation, possibly reflecting dominance of older (larger) tracemakers and thus a madurate population. The patchy distribution of smaller (younger, infant, juveniles) tracemakers could be related with more gregarious behavior. The concentration in patches might also reflect an opportunistic (r-selected strategy) character of the smaller forms linked to local concentration of organic matter (*Chondrites* is commonly associated with organic-rich deposits; Vossler and Pemberton, 1988).

As occurs with *Chondrites*, *Zoophycos* is a main trace in composite ichnofabrics (Ekdale and Bromley, 1991; Savrda, 2014). *Zoophycos* is also a very controversial trace, profusely studied, produced by a yet-undetermined organism(s), and with no consensus regarding the mode of construction or its ethological explanation (see Kotake, 2014; Löwermark, 2015; Zhang et al., 2015a, b; Dorador et al., 2016; Monaco et al., 2017). However, as a general rule, *Zoophycos* is interpreted as a deep component in multi-tiered communities in pelagic/hemipelagic sediments of different ages, reflecting late colonization and related to increasing firmness and decreasing oxygen in pore waters deep in the sediment (Rodríguez-Tovar and Uchman, 2004, 2008, 2017; Rodríguez-Tovar et al., 2009; Rodríguez-Tovar and Dorador, 2014; Giraldo-Villegas et al., 2016; Łaska et al., 2017). Furthermore, Gong et al. (2010) differentiated *Zoophycos* composite ichnofabrics (ZCI) comprising two or more suites of *Zoophycos*, with differentiation of shallowest, shallow, middle and deepest *Zoophycos* tiers on the basis of cross-cutting relationships, the soft-sediment deformation and the contrast in color between *Zoophycos* and its host rock. These authors consider that the multiple tiers may
represent variations in substrate consistency, constructed in response to the gradual accretion and lithification of sediment layers on the seafloor (Gong et al., 2010).

In the cases studied here (Cases 2a, b, 3; Figs. 3, 4), where color differentiation of the infilling material is weak, analysis of intensity pixel values could serve to identify different specimens of Zoophycos and several phases of colonization. Thus, in Case 2a, different pixel values are associated with more than one Zoophycos structure produced at different moments and in several colonization horizons during sediment accretion, as revealed by pixel values similar to those corresponding to the upper part of the greenish/whitish calcilutites and to the upper greenish calcilutite. In Case 2b, several structures of Zoophycos show similar intensity pixel values that can be associated with a single horizon of colonization within the upper greenish calcilutite, and could correspond to one specimen or to several. Especially interesting in Case 2b is the record of similar values in cross-cutting structures, suggesting different specimens produced from the same horizon of colonization (coetaneous?). In Case 3, the presence of several structures of Zoophycos with different intensity pixel values is illustrated, interpreted as several phases of colonization and diverse suites of Zoophycos. A first specimen registered at around 2-3 cm, showing higher values in the intensity profile values, might therefore reveal an initial colonization phase from the host greenish/whitish calcilutites, while the second specimen of Zoophycos, together with Planolites and Thalassinoides having similar values, would be produced latterly from the upper greenish calcilutite. The presence of Zoophycos structures above Planolites and Thalassinoides could indicate the upward migration of the tiered community during accretion of the upper greenish calcilutite, although we cannot discard the presence of minor erosions, having no record of erosional surfaces.
Conclusions

Two tools, namely *Analyze Particles* offered by Fiji software and *Intensity Profile* by ICY (IP-ICY), are shown to be highly useful for enhanced characterization of autocomposite ichnofabrics in core and outcrops. Both tools allow ichnological features to be quantified, supporting a more objective analysis than those based on semi-quantitative approaches.

*Analyze Particles* allows for the quantification of shape (roundness) and size of structures. Its application to the analysis of *Chondrites* assemblages in composite ichnofabrics from selected images of modern marine hemipelagic cores obtained at Site U1385 of IODP Expedition 339 reveals differences in size, relative abundance, density and distribution; they are interpreted as associated with variations within the population of *Chondrites* tracemakers.

*Intensity Profile* provides quantification of the intensity pixel values of the infilling material of traces, an aid to discriminate between specimens, evaluate the horizon of colonization, and appraise penetration depth. Application to the analysis of *Zoophycos* in composite ichnofabrics from deep-sea pelagic calcilutites of the Petra Tou Romiou section (Eocene Lefkara Formation; southern Cyprus) led us to differentiate between several suites of *Zoophycos*, with discrimination between structures belonging to several specimens, as well as among those from the same specimen. Moreover, different phases of colonization from several horizons could be characterized through this methodological approach.

Acknowledgements
The study was funded by project CGL2015-66835-P (Secretaría de Estado de I+D+I, Spain), Research Group RNM-178 (Junta de Andalucía), and Scientific Excellence Unit UCE-2016-05 (Universidad de Granada). The research of Olmo Miguez-Salas is funded through a pre-doctoral grant from Spain’s Ministerio de Educación, Cultura y Deporte. The research of Javier Dorador is funded through a Newton International Fellowship by the Royal Society and for the European Union’s Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No 792314 (ICON-SE). The paper benefited from comments and suggestions by Co-Editor (L. H. Vallon) and two anonymous reviewers.

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**Figure captions**

**Fig. 1.** Case 1. From left to right: Original image from Site U1385 of IODP Expedition 339. Treated image with Contrast Limited Adaptive Histogram Equalization (CLAHE) to improve visibility of trace fossils showing *Chondrites* (*Ch*), *Planolites* (*Pl*) and *Thalassinoides* (*Th*). Enlarged image of the selected part. Generated black and white binary image (B/W). Filtering process and manual corrections on the B/W image.

**Fig. 2.** Case 1. *Analyze Particle* treatment applied to filtered image of Case 1 (Fig.1). Differentiation between small (0.5-3 mm²) and large (3-8 mm²) *Chondrites*, with distribution into the treated image, respective percentage of bioturbated area and size histograms.

**Fig. 3.** Case 2. Composite ichnofabrics at outcrop deep-sea pelagic calcilutites from the Petra Tou Romiou section (Cyprus). Original image (left) showing an upper part of greenish calcilutites and a lower (dominant) part of greenish/whitish calcilutites. Treated image by CLAHE (upper right) with indication of the two conducted intensity
profiles. Intensity profiles (a, b), showing differentiated trace fossils (*Planolites, Pl*; *Zoophycos, Zo*; vb, vertical burrows), and the corresponding Intensity Pixel Values.

**Fig. 4.** Case 3. Composite ichnofabrics in outcrop deep-sea pelagic calcilutites from the Petra Tou Romiou section (Cyprus). Original and treated image by CLAHE with indication of the conducted Intensity image, showing an upper part of greenish calcilutites and a lower (dominant) part of greenish/whitish calcilutites. Intensity profile, showing differentiated trace fossils (*Planolites, Pl*; *Th, Thalassinoides*; *Zoophycos, Zo*), and the corresponding Intensity Pixel Value.