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RECEIVED 15 May 2026

REVISED 31 July 2026

ACCEPTED 10 August 2026

PUBLISHED 03 September 2026

CITATION

Mortelmans J, Decrop W,
Heynderickx H, Cattrijsse A, Depaepe M,
van Walraven L, Scott J, van Oevelen D,
Deneudt K and Muñiz C (2026) Plankton
imager 10 monitoring in the southern
North Sea: an open workflow for
classification, morphometry and
DwC-A publication.
Front. Mar. Sci. 13:1882482.
doi: 10.3389/fmars.2026.1882482

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Plankton imager 10 monitoring in the southern North Sea: an open workflow for classification, morphometry and DwC-A publication

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Plankton are a key component in marine food webs and are vulnerable to climate-driven changes. Their phenology, distribution, and body size influence the biological carbon pump and higher trophic interactions. Plankton imaging enables high-frequency observations of community composition and size structure, but operational workflows for processing, validating and publishing large *in situ* image collections remain limited. This work describes the deployment of the Plankton Imager 10 (Pi-10) aboard the R/V Simon Stevin in the southern North Sea and presents an open pipeline for Pi-10 data processing, classification, validation and standards-based publication. An open-access data processing pipeline is presented, combining geotagged imaging, deep learning classification, and image-based metrics with applicability beyond the Belgian part of the North Sea. Best practices for documenting Pi-10 datasets in Darwin-Core Archives (DwC-A) are presented. Although developed and validated for Pi-10 imagery, the workflow is designed as a modular framework that can be adapted to other plankton imaging sensors.

KEYWORDS

biological carbon pump, Darwin core archive (DwC-A), deep learning classification, phenology, plankton imaging

Background and summary

Plankton are ubiquitous (Castellani & Edwards, 2017) and their importance to marine food webs thoroughly documented (Ratnarajah et al., 2023; Chivers et al., 2017). Many plankters are vulnerable to climate-driven changes: their phenology, range, and body size change with climate (Chivers et al., 2017; Beaugrand et al., 2014) having consequences for the biological carbon pump and higher trophic interactions (Ratnarajah et al., 2023).

Plankton size structure, from individual body length to community-level size spectra, affects trophic efficiency and predator-prey dynamics (Barton et al., 2013; Pitois et al., 2021; Atkinson et al., 2024). Plankton are a key part of marine food webs and thus essential for ocean's health and resilience (Falowski et al., 1998; Grigoratou et al., 2025).

The Belgian part of the North Sea (BPNS) is a small but dynamic marine coastal area and hosts a rich planktonic community shaped by strong seasonal cycles, riverine inputs, eutrophication, and climate variability (Nihoul and Hecq, 1984; Lacroix et al., 2004; Lescrauwaet et al., 2013; Mortelmans et al., 2021). Historically, plankton monitoring in the BPNS has relied on net sampling and manual microscopy, labor-intensive methods, relatively limited in resolution and prone to observer bias (Benfield et al., 2007; Wiebe and Benfield, 2003). While these traditional approaches have provided valuable long-term data, they fall short in capturing the fine-scale morphological, spatial, and temporal patterns that are crucial for understanding ecosystem change. Recent decades have seen a rapid and diverse shift in marine observation capabilities, particularly through the development of automated, high-resolution imaging technologies (Lombard et al., 2019; Giering et al., 2022; Irisson et al., 2022). These new imaging technologies, however, still cope with challenges including the need for expert-validated training data, uncertainty in automated classifications, class imbalance, instrument-specific biases, and reduced model performance when classifiers are applied to new samples, seasons, regions, or imaging systems (Eerola et al., 2024; Irisson et al., 2022; Giering et al., 2022). In the BPNS especially, new time series are being constructed based on imaging sensors, e.g., ZooScan for zooplankton time series, since 2012 (Mortelmans et al., 2019) and FlowCam for phytoplankton time series, since 2017 (Lagaisse et al., 2025); both based on physical samples.

A new and emerging technology is the onboard Plankton Imager 10 (Pi-10), developed by the Centre for Environment Fisheries and Aquaculture Science (Cefas) and Plankton Analytics (Culverhouse et al., 2016; Pitois et al., 2018; Scott et al., 2025). The Pi-10 is a standalone, automated line-scan imaging system designed for continuous monitoring of plankton and suspended particles in a flow-through system. Although other instruments, such as the *In Situ* Ichthyoplankton Imaging System (ISIIS 3.0; Cowen and Guigand, 2008), also use line-scan cameras, their sampling configurations differ substantially: the ISIIS is a towed, underwater shadowgraph system that images organisms directly in the surrounding water and can be vertically profiled to resolve their distribution through the water column. By contrast, the Pi-10 is installed onboard a vessel or other platform and images particles in water delivered through a controlled viewing chamber by a continuous water supply. This distinction results in different operational strengths. ISIIS is particularly suited to depth-resolved surveys and the observation of relatively rare, fragile, and gelatinous organisms in their natural environment, whereas the Pi-10 prioritizes autonomous operation, straightforward installation, and integration into routine vessel activities. The Pi-10 can operate continuously without dedicated towing operations and can be deployed on any platform with access to water and power. Its enclosed flow-through configuration also enables sampling in highly turbid waters where

open-water optical systems are constrained (Ollevier et al., 2022). However, unlike a profiling system such as ISIIS, the sampling depth of the Pi-10 is determined by the position of the platform's water intake. The Pi-10 is designed to capture images of plankton and other suspended particles between approximately 20 and 3000 μm while processing around 2 m^3 of water per hour. This comparatively high and continuous sampling volume helps address limitations associated with low-volume sampling (Barth et al., 2025). Its ability to operate autonomously with minimal human intervention makes it suitable for long-term deployment on research vessels, ships of opportunity, and other platforms equipped with a water and power supply.

In this paper we provide to the plankton imaging user community: (1) a Convolutional Neural Network (CNN) model for classification of Pi-10 images, with a regional focus on the BPNS; that can be adapted for use with other imaging sensors; (2) a python script to prepare, select and classify imagery; and automatically create workable export products; (3) an R Shiny application for the users to validate the Regions Of Interest (ROIs) classified by the model; (4) an R script that formats the annotated dataset into a standardized Darwin Core Archives (DwC-A), allowing dissemination of data amongst users and enabling a flow to public databases such as EurOBIS (EurOBIS, 2026); (5) a 2.4M image dataset from the southern North Sea that was iteratively classified with a CNN model and then validated, along with simultaneously collected environmental data; and finally (6) an interpretation how the modular pipeline here presented can process imagery collected by other plankton imaging sensors, such as ZooScan, FlowCam and VPR.

This methodological contribution is a validated, open, sensor-adaptable workflow that connects high-volume plankton image acquisition to quality control, classifier confidence assessment, expert validation, morphometric trait extraction, and standards-based biodiversity publication.

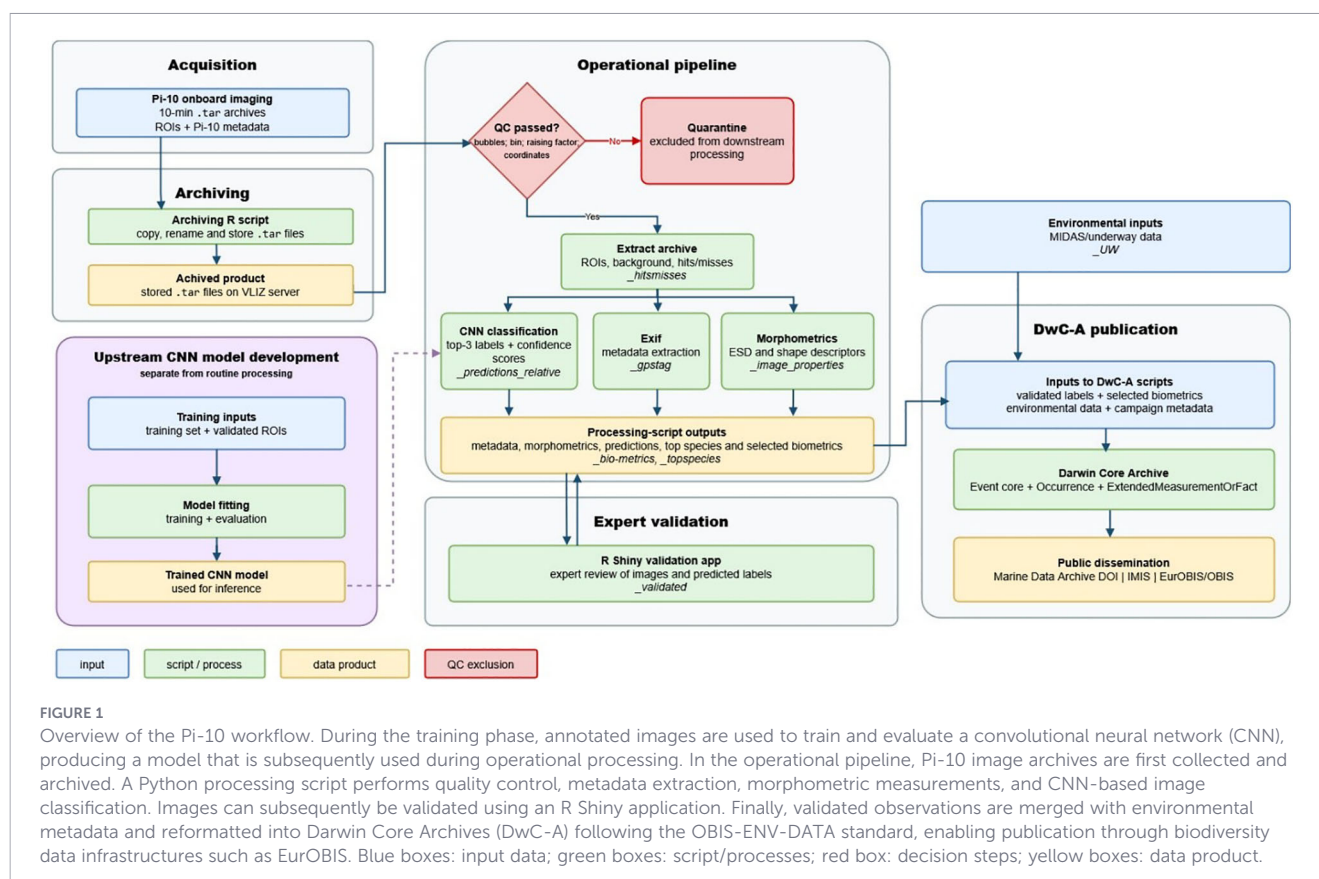
Methods

Data record and code availability

The entire pathway from sampling towards public data dissemination is described (Figure 1) and documented within this manuscript. Four scripts are involved to process Pi-10 imagery: (1) the R-script to copy and rename the compressed folders containing Pi-10 imagery (further named tar files), to a network-archive ('archiving' – Mortelmans, 2026a); (2) the python script to create all extensions associated to a certain tar ('operational pipeline' – Decrop and Mortelmans, 2026), including the CNN predictions ('upstream CNN model development'); (3) the R script to validate Pi-10 images ('expert validation' – Mortelmans, 2026b); (4) the suite of R-scripts to create DwC-A from validated data ('DwC-A publication' – Mortelmans, 2026c).

Sampling design

Data was collected between March 2025 and October 2025 aboard the R/V Simon Stevin in the southern North Sea (Figure 2A)



when the Pi-10 was continuously running, with a detail on the BPNS (Figure 2B). In total the Pi-10 was used on 32 days; totaling for 226 hours of data spread over 1,358 tar files (2.75 Tb), with each.tar file representing a 10-minute bin.

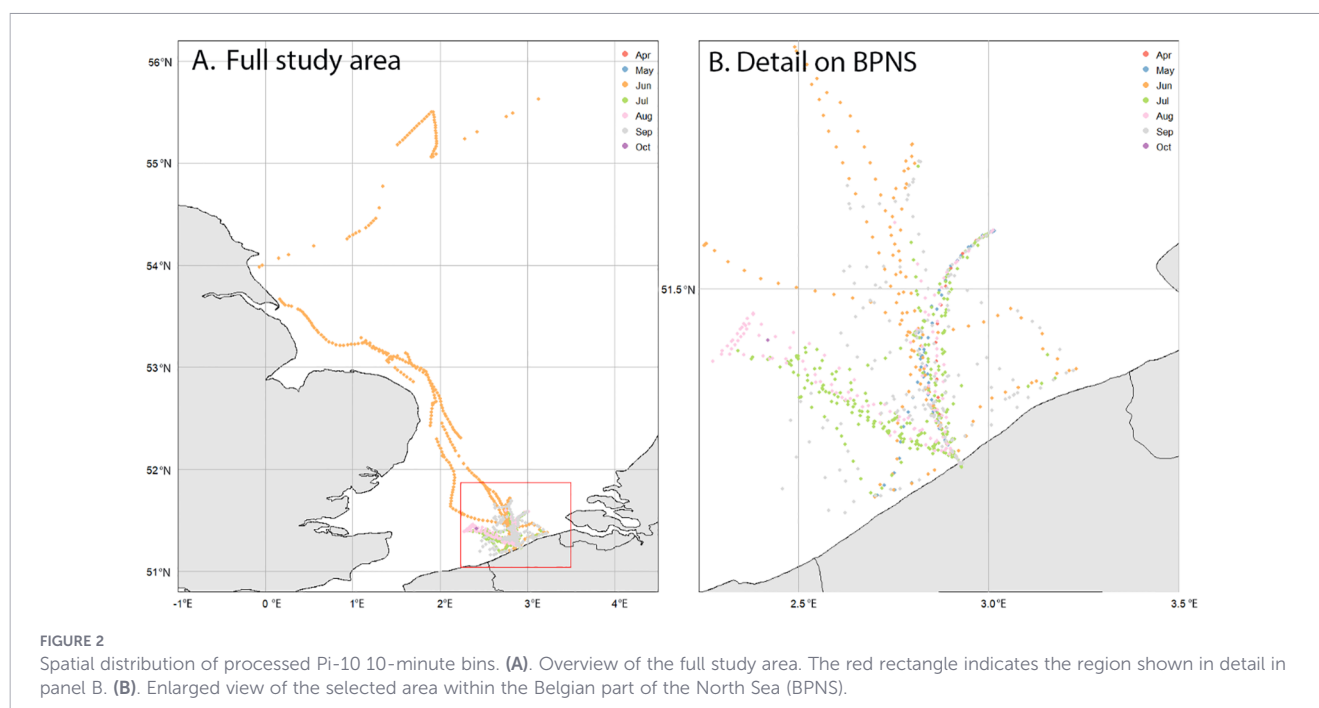
Data acquisition

The Pi-10 is housed within a robust metal frame ($75 \times 100 \times 30$ cm) and features internal climate control to maintain stable temperature and reduce humidity around the optical components. At its core is a downward-facing Basler 2048-70kc line-scan camera, capable of capturing 70,000 lines per second at a resolution of $2048 \times 10 \mu\text{m}$ pixels, enabling high-resolution imaging of particles in the 20 – 3000 μm size range. Illumination is provided by an LED lighting system, optimized to enhance contrast and reduce motion blur. The water flows through a custom flow cell, where particles are imaged as they pass through the camera's field of view. Captured images are transmitted via fiber optic cable to an onboard PC (the Pi-PC), where ROIs are created and further processed and stored in real time. The system can record up to 100,000 regions of interest (ROIs) per minute, although in turbid or particle-rich waters, not all ROIs are saved to manage data volume. Nonetheless, total particle counts are retained, allowing subsampling-based estimates to be made during post-processing. The total number of particles and the total number of saved particles are documented in the *hits-and-misses.txt* file. Further technical performance details are available (Scott et al., 2025; Pitois et al., 2025).

On the R/V Simon Stevin, the water inlet that the Pi-10 is connected to is situated at 0.6 m below the surface, which is

especially shallow in rough weather or when performing actions at the Dynamic Positioning (DP) system of the vessel. Communities sampled are therefore biased toward surface-water communities, which are then affected by wind, tide and currents (Blauw et al., 2012; Ramond et al., 2021). These conditions also result in an intake of air bubbles which negatively affect the number of imaged particles (hence, larger fractions are considered). Although the Pi-10 can image particles from 10 μm upwards, we choose 200 μm as the lower size threshold, in accordance with the 200 μm mesh size of the WP2 nets which are used in the VLIZ monitoring schedule. This size limit is standard for mesozooplankton research (Fraser, 1966) and it is routinely used for plankton monitoring in the BPNS (Mortelmans et al., 2019). We opted to collect 7,500 ROIs per minute to keep processing and validating time feasible, as well as to guard storage space. The Pi-10 is capable of collecting up to 100,000 ROIs per minute (Scott et al., 2025; Pitois et al., 2025).

With the Pi-10 being part of an underway data acquisition system, coupling to metadata and associated parameters is straightforward. The same underway data acquisition system acquires samples for ICOS (ICOS Belgium, 2026) performing standardized greenhouse gas measurements. It is equipped to measure the partial pressure of carbon dioxide ($p\text{CO}_2$) continuously, and to take discrete samples for dissolved inorganic carbon (DIC), pH and Total Alkalinity. Being on the same system creates possibilities to match biological parameters collected through the Pi-10 with the ICOS measurements. Finally, temperature readings from three sensors on the R/V Simon Stevin underway data acquisition system are available: the SBE21 (Sea-Bird Thermosalinograph), SBE38 (Sea-Bird Hull Sensor) and the SBE3 (Sea-Bird



Temperature Probe). In addition to temperature, the underway data collection includes water flow, salinity, and vessel speed measurements. All these associated data is available through the Flanders Marine Institute Data Explorer (Flanders Marine Institute, 2026).

CNN-models: training set and fitting

A FAIR training set was made available (Van Walraven et al., 2026) consisting of 50,354 ROIs in 87 classes. The ROIs in this training set were collected on board the R/V Tridens and R/V Endeavor during various fisheries surveys in 2023, mainly from the Dutch part of the North Sea (Van Walraven et al., 2026).

Before fitting our model, four modifications to the training set provided by Van Walraven et al. were performed: (1) In the current study 2,412,153 ROIs from the southern North Sea were manually validated; of which 57,802 were used to augment the original training set by Van Walraven et al. (2026). The remainder of the validated ROI's were used to evaluate model performance. We contributed especially to the rarer classes, to reach a balanced training set, with an equal number of ROIs per class. (2) Irrelevant or rarely occurring taxa in the BPNS which could create model imbalance, were excluded from the training set. For the BPNS-model it was decided to keep the number of classes relatively low to improve model performance. In total, 35 taxa were selected for image recognition (see Supplementary Information 1), aligning with the OSPAR- lifeform categories (Holland et al., 2023). (3) Several new classes were introduced to the training set based on size fractions (e.g., 'bubbles' were split in to 'large bubbles', 'small bubbles' and 'multiple bubbles'), and morphological differences (e.g., 'cumacea' were split into 'cumacea-curved' and 'cumacea-straight'). This step increased the number of classes from 22 to 30, but the number of taxa remained identical (see Supplementary Information 1). These new classes greatly improved the model's accuracy as intra-class variation among ROIs dropped substantially. (4) Subgroups within each class are identified based on particle level morphometrics obtained by the

processing script (see Supplementary Information 2, e.g., equivalent spherical diameter, eccentricity, etc.). In this study, particle-level morphometric descriptors were projected into a two-dimensional UMAP space, allowing visually similar ROIs to appear close together and making intra-class structure, outliers, and potential misclassifications easier to detect. UMAP, or Uniform Manifold Approximation and Projection, is a non-linear dimensionality-reduction method that projects high-dimensional measurements into a low-dimensional space while preserving neighborhood relationships between similar objects (McInnes et al., 2018; Healy and McInnes, 2024). For this we relied on Spotlight by Renumics (<https://github.com/Renumics/spotlight>), a Python package designed to help users quickly explore and visualize unstructured datasets. Such computer-assisted intra-class sorting helps to identify critical clusters in the data that would otherwise remain unnoticed. Subsequently, these more uniform clusters are then more easily picked up by CNN-algorithms. Spotlight also enables users to visualize a single parameter to find distinctions between ROIs and finetune a certain class in their training sets.

A practical step here taken is the alignment of Pi-10 outputs with OSPAR-aligned lifeform categories, with categories such as diatoms, dinoflagellates, copepods, gelatinous zooplankton, fish larvae, holoplankton, and meroplankton, while retaining non-taxonomic particles as a separate group (Holland et al., 2023). This provides a useful model for translating detailed Pi-10 classifications into broader monitoring categories suitable for regional assessment, while preserving the original detailed labels for ecological interpretation and future model refinement.

CNN-models: predictions

For image recognition, we relied on a CNN, a type of deep learning algorithm specifically designed for image recognition and processing (LeCun et al., 2015). CNNs automatically learn hierarchical patterns, detecting simple features in early layers and

progressively more complex structures in deeper layers. Recent studies have increasingly applied CNNs to plankton image classification (LeCun et al., 2015; Lumini and Nanni, 2019; Kareinen et al., 2026), demonstrating their effectiveness for automated image recognition.

The Pi-10 zooplankton image classifier builds directly on the FlowCam phytoplankton identification model developed by VLIZ within the iMagine project (Decrop and Lagaisse, 2025). The FlowCam phytoplankton identification model provided reusable models, FAIR-compliant outputs, and tools for retraining and performance analysis. The model architecture is based on EfficientNetV2B0, a deep CNN known for its use of residual connections to facilitate the training of very deep networks. EfficientNetV2B0 is part of the EfficientNetV2 family designed by the Google Brain team (Tan and Le, 2021). The network outputs the five most probable taxonomic classes for each image along with their confidence levels (Decrop et al., 2025), in the Pi-10 pipelines, the three most probable taxonomic classes are kept.

The Pi-10 classifier adopts this architecture but is specifically trained for ROIs collected by the Pi-10 (see section “CNN models: training set and fitting”). The training pipeline included data preparation, model training, and performance monitoring. After prediction the artificially created subclasses (e.g., ‘cnidaria 1’, ‘cnidaria 2’, ‘cnidaria 3’, ‘cnidaria 4’) are merged back into their original taxonomic category (e.g., ‘cnidaria’). This overall approach of splitting functionally or taxonomically identical groups into shape-based subgroups during the CNN procedure led to a significant improvement in model performance. When evaluated on a fully independent test set of ROIs that were not used during model training or validation, the classifier achieved a Top-1 accuracy of 86.34% and a Top-3 accuracy of 97.12%. The resulting training set and associated CNN model is published open access (Mortelmans et al., 2026).

Finally, classifier performance was evaluated by comparing model predictions with an independently validated reference dataset that was not used for model training or validation. For each ROI, the classifier assigned the three most likely taxonomic labels, each with an associated confidence score. These predictions were compared with the expert reference labels, and precision, recall, and F1-score were calculated for each class (Powers, 2007). Precision measures how often positive predictions are correct, i.e., the proportion of ROIs assigned to a given class that were correctly classified. Recall measures the completeness of detection, i.e. the proportion of ROIs belonging to a given reference class that were successfully retrieved by the classifier. The F1-score, calculated as the harmonic mean of precision and recall, provides a single metric that balances these two aspects of performance (Goodwin et al., 2022; Ciranni et al., 2024).

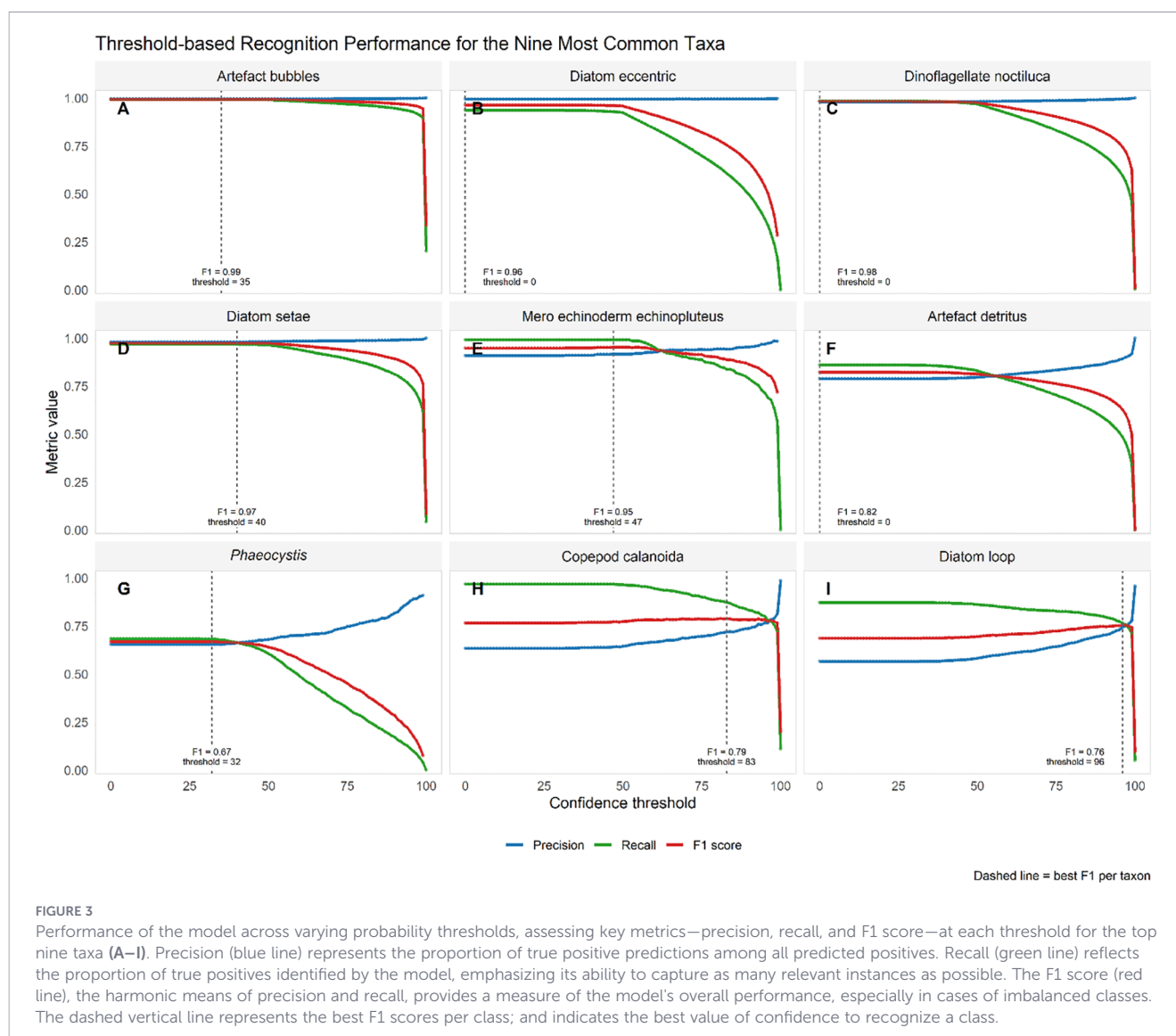
Processing script

After collecting data at sea, all.tar files are copied to a server, where they remain for storage. For copying data to the server, we use a R-script which changes the filename provided by the Pi-10 software to contain the date in addition to the time (and hence, making it unique) (Mortelmans, 2026a). In the processing pipeline

(Decrop and Mortelmans, 2026) each.tar archive undergoes four assessments whether to proceed further downstream or not.

1. The script generates a preview dataset by randomly sampling 200 TIFF images from the extracted content. These ROIs are subjected to an initial classification step using a CNN. The model assigns each of these 200 ROIs a top-1 predicted class, and the resulting distribution of class labels is statistically analyzed. If the relative frequency of “bubbles” exceeds a predefined threshold (default: 40%), the archive is flagged as low-quality input and is automatically moved to a quarantine directory for exclusion from further processing. As bubbles are recognized with a precision over 99% (Figure 3), it is a very suited method to save time in downstream processing.
2. Per minute, the total number of particles and the total number of saved particles are documented in the *hits-and-misses.txt* file (Scott et al., 2025). The second assessment is based on this *hits-and-misses.txt* file – if a certain.tar file contains less than the expected 10 rows (i.e., it measured not the full 10 minutes), the file is moved to a quarantine directory, for exclusion from further processing. That implies, only.tar files are processed from complete 10-minute bins avoiding corrupt datafiles.
3. Third, we calculated a quality-control metric referred to here as the “raising factor”. This metric describes the fraction of missed detections relative to the total number of accepted detections in a file. In practice, a high raising factor indicates that a substantial correction would be required before converting image counts into particle densities. Such files are therefore more likely to produce biased downstream abundance or density estimates. In the current workflow, files with a raising factor above 0.05 were moved to a quarantine directory for manual inspection. This threshold should be interpreted as a conservative empirical cut-off rather than a universal value. It was selected to flag files in which more than approximately 5% of detections required correction or were affected by processing uncertainty. Future applications may recalibrate this threshold using independent validation data, especially when the workflow is transferred to new regions, seasons, instruments, or ecological applications.
4. A final image-quality assessment is performed by evaluating the grayscale intensity along the outer edges of a random subset of ROIs. For each archive, the script samples 20 images and calculates the mean grayscale value of the outer image border. If this mean edge intensity falls below the predefined threshold, this indicates that the images are overloaded with microbubbles, which negatively affect downstream processing.

Once these escape rules are cleared, eight types of extensions are created for each.tar. All processing times are logged to identify bottlenecks in the pipeline. Eight types of extensions are available documented in Table 1.



Computational analyses were performed in a virtualized environment running Microsoft Windows Server 2022 Standard, version 10.0.20348, Build 20348, hosted on a Nutanix AHV platform. The CPU environment was allocated 16 Intel Xeon Cascade Lake CPU cores/16 logical processors running at approximately 3.59 GHz, with 98.2 GB physical RAM. GPU-accelerated processing was performed using two NVIDIA A16 GPUs, each with 15,356 MiB memory, for a total of approximately 30 GiB GPU memory. The NVIDIA driver version was 595.71.05, and nvidia-smi reported CUDA version 13.2. Python processes were used for GPU computation. The script takes on average 3.12 hours per tar, ran on CPU; and on average 19.8 minutes on GPU; where most computing time is spent on the CNN-predictions and exif extraction.

Validation

After model prediction, ROIs can be manually validated using the rShiny annotation tool provided with this workflow (Mortelmans, 2026b). For each ROI, the script extracts the two most likely predicted classes and their associated confidence scores.

Predictions with very high confidence (>0.999) and a large margin over the second-best prediction (>0.9) are labelled with the suffix “_AI99,” indicating that the classifier assigned the object almost exclusively to one class and showed little confusion with the runner-up class. Predictions that do not meet these criteria are retained without this suffix and remain available for manual validation. These thresholds should be interpreted as conservative heuristic cut-offs rather than formally optimized universal values. They were chosen to prioritize precision and reduce the risk of automatically accepting ambiguous predictions, especially in a dataset containing many visually similar plankton and particle classes.

Data publishing

The validated data are then reformatted according to the OBIS-ENV-DATA standard, an extension of the Darwin Core Archive (DwC-A) specifically tailored for sample-based marine biological datasets (De Pooter et al., 2017; Wiczorek et al., 2012). This format enables the integration of taxonomic, environmental, and sampling metadata within a single, self-contained dataset. It allows users to

store information from the Pi-10 in an interoperable format, allowing quick and clean dissemination through public biodiversity data systems like EurOBIS. There are three scripts documented (Mortelmans, 2026c), resulting in one DwC-A holding three (data)files:

1. the core file is an Event table, which captures hierarchical information about the sampling event, such as location, date, time, and depth. Nine different measurementTypes are associated to each eventID (see Table 2). This Event core is linked to two extensions:
2. the Occurrence extension, which documents the presence or absence of biological taxa; and where all taxon names are matched against the WoRMS taxonomic backbone. For each taxon, the validation status was recorded as: 'NA' (not assessed), '0' (assessed for but not found), and '1' (assessed for and found).
3. the ExtendedMeasurementOrFact (eMoF) extension, which holds quantitative and qualitative data. The eMoF includes biotic measurements (e.g., abundance), environmental variables (e.g., temperature, salinity), and metadata on sampling devices and protocols. The extension allows a scheme as proposed by Van Walraven et al. (2026); which allows multiple labels per image, separating taxonomic classes (e.g., 'Oithona') from descriptive attributes (e.g., 'egg sac'), enabling flexible grouping and future additions without altering original labels. To document size measurements per taxon, two measurementTypes are suggested, each explicitly linked to an occurrenceID (see Table 3). These measurement types allow users to report (1) the mean volume calculated on the equivalent spherical diameter (ESD) of all detected objects assigned to a given taxon,

and (2) the total volume calculated on ESD of all detected objects assigned to that taxon.

To run the script successfully, users need to provide their input data in a simple format, documented in the [Supplementary Information 3](#); and provide minimal metadata on scientific campaigns in a separate.csv table, containing spatiotemporal information on the campaigns specified, documented in the [Supplementary Information 4](#).

For archiving, the obtained DwC-A is uploaded to the Marine Data Archive (<https://marinedataarchive.org/>), an online repository specifically developed to independently archive data files in a fully documented manner. Subsequently, the archive is assigned a digital object identifier (DOI) for traceability. All datasets described above are linked in the Integrated Marine Information System (IMIS), an ISO-19115 compliant catalogue for metadata discovery catalogue. Finally, the script is now designed for validated data but works on predictions equally well. An overview of each specific field in the archive; and a brief description of the content, is provided in [Supplementary Information 5](#).

Results and discussion

Classifier performance

The performance of the CNN classifier varies across taxa. Distinguishing between morphologically similar or highly heterogeneous taxa remains challenging and negatively affects the performance of the model (Ciranni et al., 2024). The optimal confidence threshold that balances precision and recall was determined for each class (Figure 3); the vertical dashed line indicates the class-

TABLE 1 Standardized output extensions generated by the Pi-10 processing pipeline.

Output extension	Description
_background.tif	Contains the background image of a .tar file. Extracting the _background.tif is essential for quick assessment of the metadata, as it allows quick access to metadata without having to unzip entire .tar files.
_gpstag.csv	Contains a timestamp and GPS coordinates for each ROI in the .tar. The script extracts .tif files, reads GPS metadata using exif, and compiles these into a cleaned dataset.
_hitsmisses.txt	Keeps track of the raising factor considered in each minute of the .tar file. The hitsmisses.txt file counts the number of hits, i.e. particles seen and stored, and misses, i.e. particles seen but not stored, allowing fractions to be calculated. This file is copied to the standardized output to allow quick access without having to unzip entire .tar files.
_image_properties	Stores all metrics for each ROI. Deep-learning predictions and shape-descriptor extraction are performed. In total, 25 morphological features of the dominant object in each ROI, such as area, eccentricity, orientation, and Hu moments, were extracted using region-based segmentation with scikit-image (van der Walt et al., 2014). In downstream calculation of the surface area, or "biovolume", the parameter object_additional_area is used, representing the total area occupied by the object in pixels. The full list is given in Supplementary Information 2 .
_predictions_relative	A JSON file containing the classification results of the CNN algorithm. The top-3 most probable taxonomic labels, together with their associated confidence scores, are saved.
_topspecies	An extension of _predictions_relative that stores the best-fitting class for each ROI; this is: one ROI, one name, and one confidence score. Keeping this information in a simple .csv allows users to quickly assess the content of a processed .tar file and obtain a general characterization of the .tar.
_validated	Contains the result of manual validations. This extension may be absent if no validations have been performed yet.
_bio-metrics	Contains concatenated biodiversity data per minute for classes where the model reaches high recall and precision, and a high F1-score at low confidence thresholds. For these classes, counts, mean ESD, and total ESD are calculated and then merged with information on the raising factor and essential metadata, including coordinates and timestamp, followed by the calculation of densities.

The table summarizes the purpose and content of each output file created for a processed .tar archive.

TABLE 2 Suggestion for event related measurementTypes in DwC-A.

Measurement type ID	Measurement value	Measurement value ID	Measurement unit	Measurement unit ID
	Plankton Analytics Pi-10 Plankton Imager	http://vocab.nerc.ac.uk/collection/L22/current/TOOL1844/		
http://vocab.nerc.ac.uk/collection/P01/current/VOLXXXX/	340		litre	http://vocab.nerc.ac.uk/collection/P06/current/ULIT/
http://vocab.nerc.ac.uk/collection/P01/current/SSAMPC01/	0.13513			
http://vocab.nerc.ac.uk/collection/P01/current/APSABT01/	1.451		Knots	http://vocab.nerc.ac.uk/collection/P06/current/UKNT/
http://vocab.nerc.ac.uk/collection/P01/current/DISTPHMS/	172.758		Metres	http://vocab.nerc.ac.uk/collection/P06/current/ULAA/
http://vocab.nerc.ac.uk/collection/P01/current/TEMPPR01/	14.3298		Degrees Celsius	http://vocab.nerc.ac.uk/collection/P06/current/UPAA/
http://vocab.nerc.ac.uk/collection/P01/current/PSALPR01/	31.4843		Grams per kilogram	http://vocab.nerc.ac.uk/collection/P06/current/UGKG/
http://vocab.nerc.ac.uk/collection/P01/current/NMSPPF01/	Simon Stevin	http://vocab.nerc.ac.uk/collection/C17/current/11SS/		

TABLE 3 Suggestion for taxon related measurementTypes in DwC-A.

Measurement type	Measurement type ID	Measurement value	Measurement unit	Measurement unit ID
Surface area of biological entity specified elsewhere in analysed sample from the water body by image analysis and computation from Equivalent Spherical Diameter (ESD)	http://vocab.nerc.ac.uk/collection/P01/current/AREBPESD/	0.195094	Square micrometres	http://vocab.nerc.ac.uk/collection/P06/current/SQUM/
Surface area mean of biological entity specified elsewhere in analysed sample from the water body by image analysis and computation from Equivalent Spherical Diameter (ESD)	http://vocab.nerc.ac.uk/collection/P01/current/AREAVESD/	3.55094	Square micrometres	http://vocab.nerc.ac.uk/collection/P06/current/SQUM/

specific confidence threshold at which the F1-score is maximized. For each threshold, predictions with confidence scores below that value are not accepted as accurate classifications; therefore, increasing the threshold generally improves precision but reduces recall by rejecting more true positives. Improving the current model falls outside the scope of this paper but is improved by an iterative process of improving the learning set, retraining the model, and validating the model (Ciranni et al., 2024; Goodwin et al., 2022; LeCun et al., 2015).

Five classes – *Noctiluca*, diatom-setae, diatom-eccentric, echinoderm_echinopluteus and bubbles – are correctly identified even at low confidence thresholds; and hold few false positives (i.e. ‘there are no other taxa wrongly identified as these classes’) (Figures 3A–E). These groups yield high recall and precision. The best F1 is achieved even at low confidence thresholds. For these groups it seems acceptable to keep the model’s predictions without additional validation, as further manual checking would yield minimal improvement to the dataset but would cost a lot of time. Other classes – e.g., copepod-calanoidea and detritus – are reasonably well identified even at low confidence thresholds; but hold many false positives (i.e. ‘a lot of other taxa are wrongly identified as these classes’) (Figures 3F, H). These groups yield high recall but very low precision. The best F1 is achieved only at high confidence thresholds. In other words: while all true members of a specific class are recognized, many individuals from other classes are incorrectly included. Finally, other classes – e.g., *Phaeocystis*, Cnidaria – are poorly identified by the model. It is clear manual validations remain essential (Figures 3G, I).

For the classes in the first group, the script already provides one self-contained output file (the bio-metrics.csv extension) which includes all information required for integration into DwC-A, even without validation. For other classes, automation can reduce manual annotation effort (Luo et al., 2018), however, the risk of misclassification, particularly for key ecological indicators, necessitates continued expert validation (González et al., 2017). Furthermore, classification uncertainty may affect downstream ecological interpretation when predictions are used for abundance, density, or biomass estimates. This is particularly important for calanoid copepods, which are ecologically important in marine food webs and monitoring frameworks (Mauchline, 1998; Kiørboe, 2011). The observed combination of high recall and low precision indicates that most true calanoid copepods are recovered, but that some non-target particles are also classified as copepods. Without correction, this may lead to overestimation of copepod abundance and biomass.

Relation to the broader plankton imaging field

Within the landscape of plankton imaging, the Pi-10 occupies a specific niche as it is an onboard instrument tailored for continuous, high-frequency monitoring of plankton communities without interfering with other research activities such as CTD profiling or transits (Scott et al., 2025; Pitois et al., 2025). The full spectra of *in situ* imaging systems are presented in Lombard et al. (2019) and include the Video Plankton Recorder (Davis et al., 2005), the *In Situ*

Ichthyoplankton Imaging System (ISIIS-3, and predecessors) (Cowen and Guigand, 2008), the Imaging FlowCytobot (Olson & Sosik, 2007; Sosik & Olson, 2007), the Lightframe On-sight Keyspecies Investigation (LOKI, Schulz et al., 2010) and ZooGlider (Ohman et al., 2018), as well as other profiling or semi-automated plankton observation systems such as UVP6 (and predecessors) (Picheral et al., 2022). The Pi-10 stands out in the field of *in situ* imaging for its operational simplicity and its unique capacity for high-throughput, continuous data acquisition. Every *in-situ* imaging sensor relies on distinct acquisition strategies making them unique in several aspects (Lombard et al., 2019; Giering et al., 2022; Irisson et al., 2022). However, once images are segmented into ROIs, the resulting image-based outputs are structurally comparable to the Pi-10. For that reason, the presented end-to-end pipeline for Pi-10 imagery can be adapted to other plankton imaging sensors, because the pipeline requires two mandatory inputs: a compressed.tar file containing ROIs, and a sensor-specific training set and model, which can be constructed following Decrop and Lagaisse (2025).

This approach has been tested on imagery from multiple sensors, with models constructed for datasets originating from ZooScan, FlowCam, and the Video Plankton Recorder (Decrop et al., 2026). Compressed image archives from these sensors successfully completed the presented pipeline, with limitations such as missing EXIF information and missing hits-and-misses information, being translated into the pipeline’s output. These limitations were overcome by merging external metadata sheets to the pipeline. While ROIs from both benchtop sensors or *in-situ* sensors can be processed with the current pipeline, there is a great need for comparison among instruments, due to sampling mechanisms, volumes, and statistical uncertainties that differ among devices (Barth et al., 2025). The standardization of image processing, however, is an incremental step forward especially in a broad and quickly evolving landscape (Goodwin et al., 2022; Ciranni et al., 2024).

Limitations, solutions and future directions

A first limitation is model performance. Model performance is expected to be regionally and seasonally specific, because classifiers trained on one plankton community, imaging setup, or environmental context may perform less well when transferred to new regions, seasons, instruments, or taxonomic compositions (Goodwin et al., 2022; Plonus et al., 2021; Conradt et al., 2022; Eerola et al., 2024). Therefore, the current classifier should be interpreted as regionally calibrated to the Belgian part of the North Sea. In addition, expert annotations should be considered reference labels rather than absolute ground truth. Manual plankton annotation is inherently subject to observer bias and inter-annotator variability. These uncertainties can propagate into both model training and performance evaluation, especially when ambiguous images are forced into a single taxonomic class (Culverhouse et al., 2003; Schröder et al., 2020; Schmarje et al., 2021; Eerola et al., 2024). For the current dataset, each image in the 2.4 million-image dataset was validated by at least two annotators. However, formal inter-annotator agreement metrics were not

calculated, and no systematic uncertainty score was assigned to individual annotations. Beside environmental context, improved training sets can make a model output more performant: the classical method of building training sets should be avoided, and in-depth morphometry should help users to distinct classes which cannot be separated to the human eye, but are easily recovered by clustering (Schröder et al., 2020). Although UMAP-based clustering is suggested here, cluster-assisted annotation tools such as MorphoCluster have already demonstrated their value for efficient plankton image validation (Schröder et al., 2020). And finally, scalability becomes especially important in obtained Pi-10 datasets – the more you collect, the longer the processes run. An associated limitation is the taxonomic resolution being limited by image-based analysis: many plankters cannot be reliably distinguished from two-dimensional images alone, classifications are often limited to broader morphotaxa or higher taxonomic levels than can typically be achieved using traditional microscopy (Culverhouse et al., 2006; Sorochan et al., 2026; Savineau et al., 2026).

Second, although automated classification reduces repetitive manual work after a model has been trained, it does not remove the need for expert annotation. Training, validation, and performance evaluation all depend on high-quality, taxonomically reliable reference labels, and the creation and maintenance of such labelled datasets remains time-consuming (Schröder et al., 2020; Irisson et al., 2022; Eerola et al., 2024). Addressing manual validation efforts, and how to keep these efforts as low as possible, is identified as a huge step forward in plankton imaging pipelines (Schröder et al., 2020; Irisson et al., 2022; Sorochan et al., 2026). Although a UI is here provided to validate big data volumes, it may also be possible to use existing cloud-based validation platforms for Pi-10 imagery, with EcoTaxa (Picheral et al., 2026; Schröder et al., 2020) being a promising option. This platform is already a widely adopted community platform for plankton image annotation, quality control, taxonomic management and data sharing across multiple imaging systems (Picheral et al., 2026). Despite the platform's ease of use, we were unable to upload the full Pi-10 image bins because of its exceptional data volume: EcoTaxa uploads are currently limited to 10Mb; whereas here we process 10minutes bins of roughly 1Gb each. Working with subsetted data may be a feasible alternative, but this would require careful selection of representative image subsets to avoid introducing bias into the validation process. Further discussion within the larger consortium is needed to address data storage strategies, including where and how to store data and what proportion of acquired data should be retained. First steps are taken by Scott et al. (2025) who uses Azure data packs for storage. Similarly, the workflow requires sufficient computational capacity for image processing, model inference, data storage, and quality control, particularly during high-particle-load conditions when image volumes can become very large (Lombard et al., 2019; Schmid et al., 2023; Pitois et al., 2025) which is partly covered by running the process on GPU.

Third, despite the recent technical development of the Pi-10, its application for calculating ecological indicators remains only partly explored (Pitois et al., 2018; Scott et al., 2021; Van Walraven et al., 2026). This represents an important limitation, because image-derived plankton data do not automatically translate into robust

indicators of ecosystem state. Nevertheless, the instrument has strong potential in this context, particularly because it generates standardized size and morphological measurements for every detected particle, going beyond biodiversity tools that focus mainly on taxonomic detection. Such trait-based information (Litchman et al., 2013) is highly relevant for functional ecology and ecosystem modeling, as size structure reflects community dynamics, trophic interactions, grazing pressure, and biogeochemical fluxes (Benedetti et al., 2018; Lombard et al., 2019; Hirai et al., 2020). A key strength of the Pi-10 imagery is the generation of high-quality ROIs with minimal object overlap, allowing robust calculation of 2D size measurements for imaged particles. However, image-derived morphometrics can still be influenced by particle orientation, image resolution, optical distortion, segmentation uncertainty, partial objects at image boundaries, and occasional touching or overlapping objects (Lu et al., 2017; Miyazaki et al., 2026). These limitations are especially relevant for fragile, elongated, colonial, or irregularly shaped plankton. Therefore, the resulting morphometric measurements should be interpreted as consistent image-based approximations (Merz et al., 2021). The selected morphometric descriptors were included to capture complementary aspects of particle size, shape, and image structure (Gorsky et al., 2010). Size-related variables, including area, equivalent spherical diameter, and major and minor axis length, describe particle dimensions, while shape descriptors such as eccentricity, solidity, and convexity capture elongation, compactness, and boundary irregularity. Moment-based features, including Hu moments and inertia tensor metrics, provide additional information on particle symmetry and pixel distribution. Together, these variables form a general-purpose feature set for image classification, quality control, and future ecological analyses. In this study, 'object_additional_area' was used as the main morphometric variable for ecological summaries because it is directly interpretable and suitable for describing particle-size distributions. The remaining descriptors were retained because they may support taxonomic discrimination, detection of atypical objects, and future trait-based analyses, although their individual ecological interpretation was not the primary focus here. Further validation could compare Pi-10-derived size distributions with independent measurements, such as co-located Laser *In-Situ* Scattering and Transmissometry instrument (LISST; Agrawal and Pottsmith, 2000) observations or controlled laboratory images, but such an intercomparison would require a dedicated validation framework beyond the scope of this workflow paper (Merz et al., 2021).

Finally, Pi-10-derived ROIs could contribute to indicator calculations such as Copepod Mean Size and Total Abundance (CMSTA) (Pitois et al., 2021), while zooplankton biomass and diversity metrics may support broader assessment frameworks such as those used in OSPAR's Quality Status Report (Holland et al., 2023). Such indicators allow to translate complex plankton community data into interpretable signals of ecosystem change (Holland et al., 2023), often before these changes become visible at higher trophic levels (Mackas et al., 2004). Future work should therefore focus on converting high-frequency Pi-10 observations into validated, interpretable ecological indicators, including biodiversity indices, bloom-risk alerts, and ecosystem health assessments

(Ohman et al., 2018; Lombard et al., 2019; Kraft et al., 2022). In the longer term, onboard or near-real-time classification could enable adaptive sampling and early-warning applications, while also facilitating integration with satellite observations, ecosystem models, and digital ocean-twin frameworks (Ohman et al., 2018; Scott et al., 2025).

Conclusion

This study documents the operational use of the Plankton Imager 10 (Pi-10) aboard the R/V Simon Stevin and presents an open and reproducible end-to-end workflow for processing, classifying, validating, and publishing high-frequency plankton image data, accompanied by a dataset of 2.4 million validated images. By combining continuous *in situ* imaging, CNN-based classification, expert validation, particle-level morphometry, and standards-based publication, the workflow demonstrates how large image collections can be transformed into FAIR and reusable biodiversity datasets. Beyond species occurrence, the integration of size and shape measurements enables analyses of plankton size structure and functional traits, thereby extending the ecological value of image-based observations. By mapping image-derived observations to Darwin Core Archives using the OBIS-ENV-DATA standard, the workflow provides a bridge between emerging plankton imaging technologies and established biodiversity data infrastructures such as OBIS, facilitating interoperability, long-term preservation, and data reuse.

Although developed for the southern North Sea and applied here to Pi-10 imagery, the framework is not limited to a single instrument. The modular structure of the workflow has been tested for processing ROI-based image archives originating from ZooScan, FlowCam and the Video Plankton Recorder, demonstrating its potential as an adaptable processing framework. However, this should not be interpreted as quantitative cross-platform validation of the Pi-10 classifier. Broader application to other sensors requires platform-specific preprocessing, retraining, threshold calibration and independent validation. As imaging technologies continue to generate increasingly large and complex datasets, reproducible and standards-based workflows will become essential for integrating observations across platforms and regions.

Effective marine biodiversity research relies on standardized data formats to support global data sharing, integration, and reuse. The Darwin Core Archive (DwC-A) is a widely recognized standard for biodiversity data and provides a practical structure for publishing Pi-10-derived observations in a consistent and interoperable format (Wieczorek et al., 2012). In this workflow, abundances are stored per 10-minute bin. Although finer temporal resolution is possible, it would substantially increase data volume. By linking image acquisition, machine-learning classification, expert validation, morphometry, and FAIR data publication within a single framework, this workflow provides a practical foundation for automated ecosystem monitoring and contributes to the development of interoperable, data-driven plankton observation systems, including future digital representations of marine ecosystems and digital ocean twins (Wilkinson et al., 2016).

Data availability statement

The original contributions presented in the study are included in the article/Supplementary Material. Further inquiries can be directed to the corresponding author.

Author contributions

JM: Supervision, Conceptualization, Data curation, Investigation, Validation, Writing – review & editing, Methodology, Formal analysis, Software, Writing – original draft, Resources, Visualization. WD: Methodology, Writing – review & editing, Software. HH: Methodology, Writing – review & editing. AC: Resources, Writing – review & editing. MD: Conceptualization, Writing – review & editing, Investigation, Methodology, Resources. LV: Writing – review & editing. JS: Writing – review & editing. DO: Writing – review & editing. KD: Investigation, Resources, Funding acquisition, Project administration, Writing – review & editing, Conceptualization. CM: Resources, Writing – review & editing, Funding acquisition, Project administration, Methodology, Conceptualization.

Funding

The author(s) declared that financial support was received for this work and/or its publication. This work was supported by data and infrastructure provided by VLIZ as part of the Flemish contribution to LifeWatch [I000819N-LIFEWATCH, I002021NLIFEWATCH].

Acknowledgments

This work was made possible thanks to the intense involvement of the crew of the R/V Simon Stevin, DAB VLOOT and colleagues from VLIZ. For the initial round of validations, we thank especially Irina Lung for her endurance in validating. Finally, we thank the three reviewers for their suggestions and corrections, largely improving the manuscript.

Conflict of interest

The author(s) declared that this work was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Supplementary material

The Supplementary Material for this article can be found online at: <https://www.frontiersin.org/articles/10.3389/fmars.2026.1882482/full#supplementary-material>

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