

## *Supporting Information*

# **An automated and intelligent microfluidic platform for microalgae detection and monitoring**

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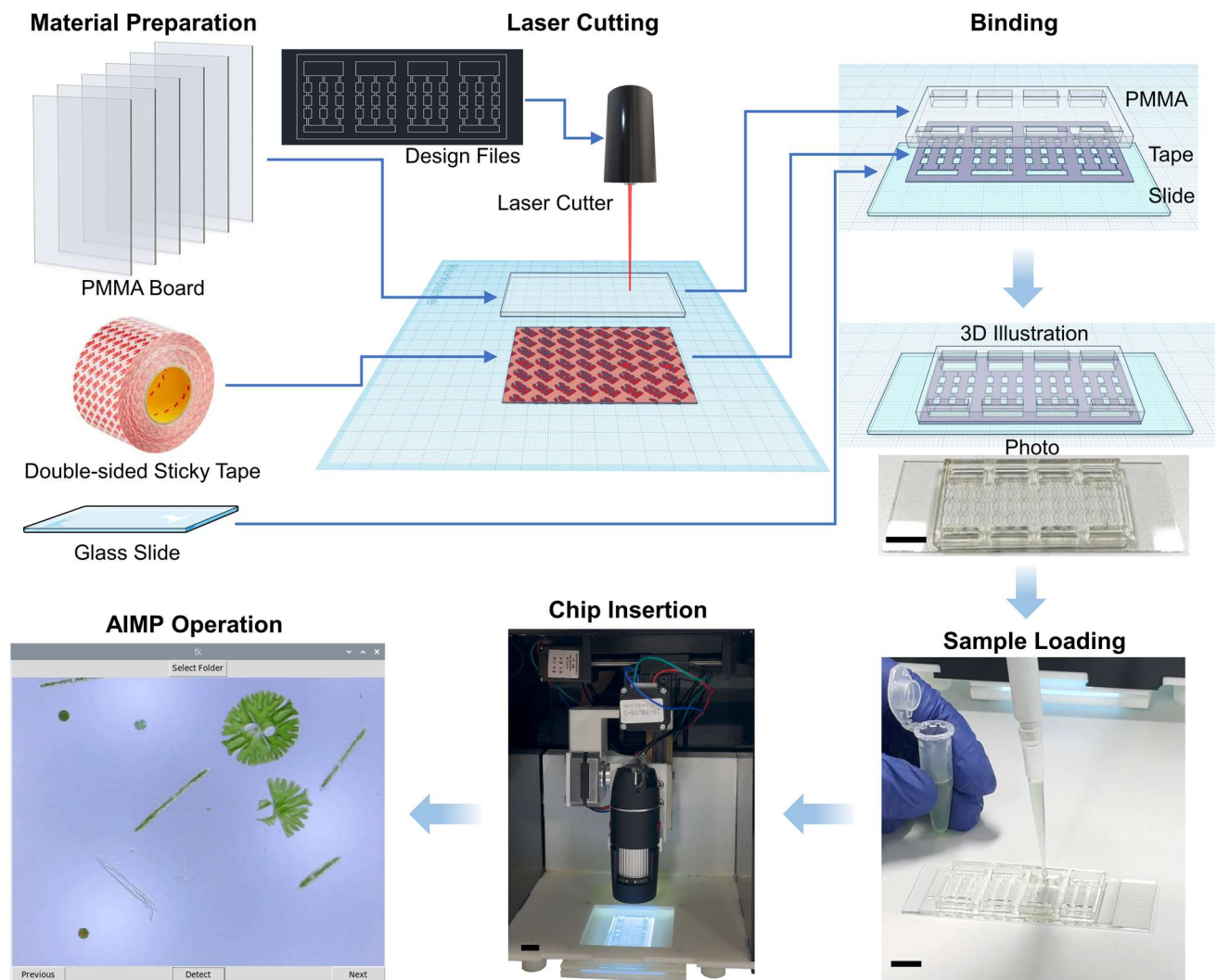
## List of costs for the main components of AIMP

Table S1 lists the names, quantities and costs of the main components used to build the AIMP. Ignoring the price of consumables such as glue, PMMA shells, etc., the total cost of building the AIMP was less than £170.

**Table S1.** Cost for the main components of AIMP

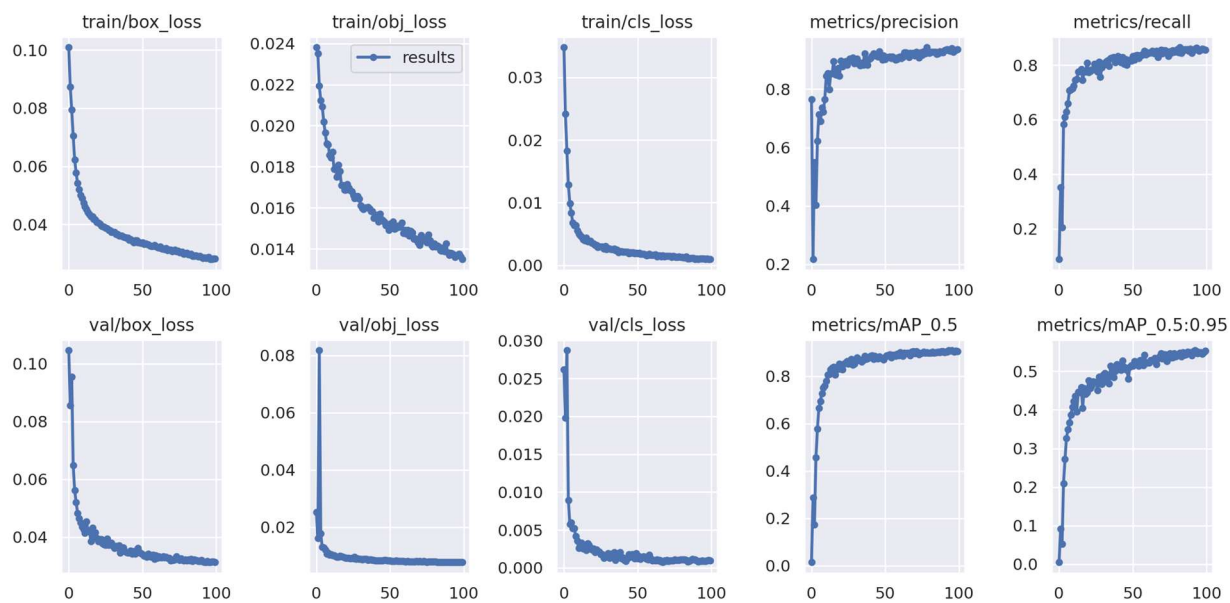
<b>Component</b>	<b>Quantity</b>	<b>Cost in Total (in British Pound)</b>
USB microscope	1	£21.38
Miniature Electric Linear Actuator	3	£64.98
Stepper Motor Driver (DRV8825)	3	£9.99
Raspberry Pi	1	£45.83
Backlight LED Board	1	£2.48
Battery	1	£23.00

## Microfluidic Channel Design and AIMP Operating Process



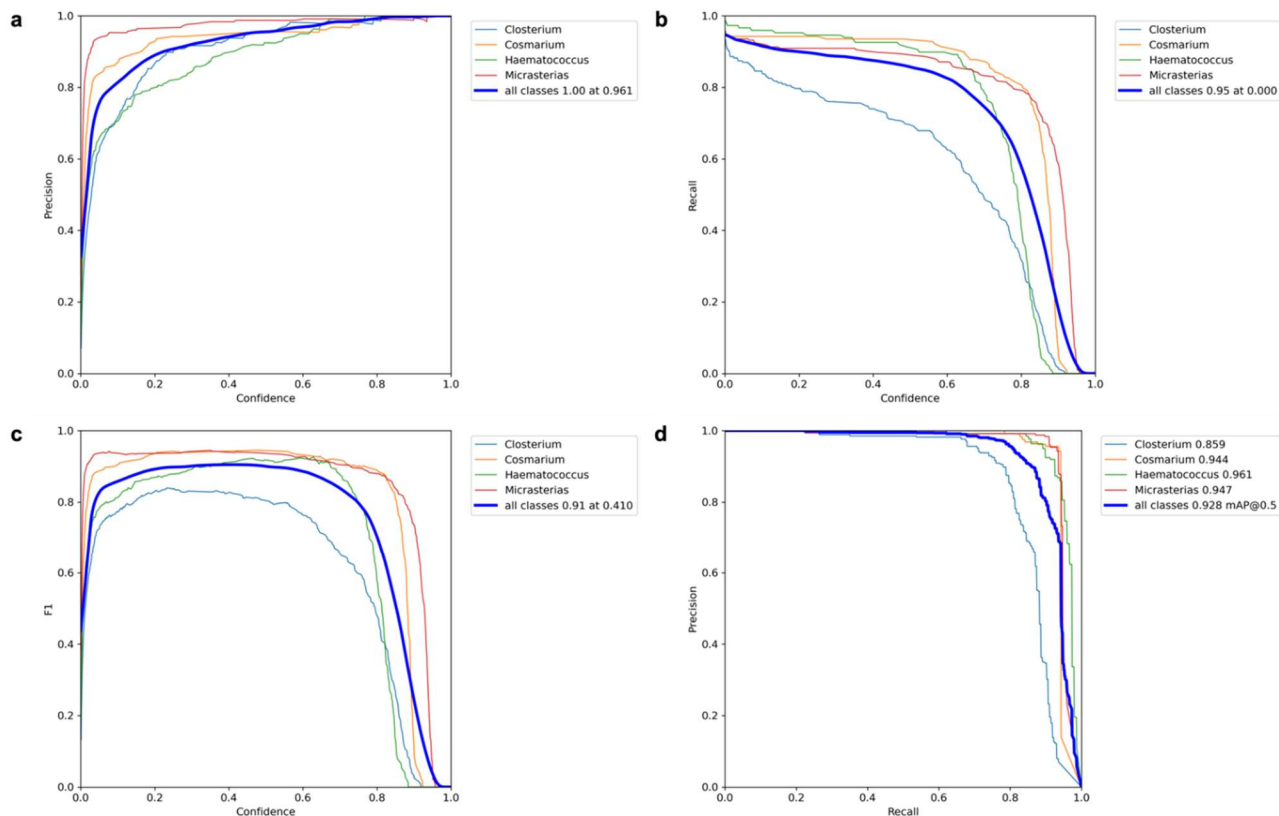
**Figure S1:** Microfluidic chip fabrication process and the AIMP operation procedures. Scale bars are 1 cm.

## Training Process Recorded for the Microalgae Species Detection Model



**Figure S2.** Training process (trained on YOLOv5s, 100 epochs, 16 batch size)

## Microalgae Detection Model Evaluation



**Figure S3.** Evaluation of the trained microalgae species detection model.

Based on YOLOv5 (more specifically YOLOv5s), a microalgae species detection deep convolutional neural network with a depth of 283 layers was trained on the collected microalgae dataset. To evaluate the trained model, we calculated Precision ( $P$ ), Recall ( $R$ ) and  $F1$  score using three basic evaluation values, TP, Background FN and Background FP for each microalgae species to be detected, where

$$Precision (P) = \frac{TP}{TP + FP}$$

$$Recall (R) = \frac{TP}{TP + FN}$$

$$F1 = \frac{2PR}{P + R}$$

Since this is an object detection task, we induced a confidence score for better evaluation. The confidence score (Conf) is associated with each bounding box that the model predicted, which can indicate the model's belief in the presence of the microalgae being detected and its species within the predicted bounding box. With this value induced, the following P vs Conf (Fig. S3a), R vs Conf (Fig. S3b) and F1 vs Conf (Fig. S3c) could be found. These curves were obtained by calculating P, R and F1 values under different confidence scores (from 0 to 1), respectively. The PR curves (Fig. S3d) are composed of the values of precision and recall plotted at different confidence thresholds for each species.

The average precision (AP) for each category can be derived by calculating the area under the PR curve. The area under the PR curve can usually be obtained using the trapezoidal rule:

$$AP = \text{Area Under PR Curve} = \sum_{i=1,2,\dots,n} \frac{(R_i - R_{i-1})(P_i + P_{i-1})}{2}$$

where  $n$  is the number of unique confidence scores,  $R_i$  is the R value at the  $i$ -th threshold, and  $P_i$  is the P value at the  $i$ -th threshold. Based on the AP, the mean average precision (mAP) can be derived by using:

$$mAP = \frac{1}{S} \sum AP$$

where  $S$  is the number of species to be detected (in this case,  $S = 4$ ).

The intersection-over-union (IoU) threshold is also a consideration when evaluating the model. To calculate  $mAP@k$  ( $k$  is the IoU value), we first consider a prediction as a true positive (TP) only if its IoU with the ground truth bounding box is greater than or equal to  $k$ , resulting in  $P@k$  and  $R@k$ . The corresponding  $mAP@k$  is then derived according to the above process.

## Performance of Other Network Architectures

In addition to the YOLOv5 architecture, other YOLO series (YOLOv7, YOLOv8)<sup>1,2</sup> are used for comparison (Table S2). For YOLOv8 we compared architectures with different numbers of parameters (from n, s, m, l, with increasing numbers of parameters in that order). Same as YOLOv5, YOLOv7 and v8 were trained for 100 epochs with a batch size of 16 using the same dataset. As indicated in Table S2, they all have a decent performance on microalgae detection. As the depth and parameters of the model increase, YOLOv7 and v8 do not show a significant advantage in microalgae detection accuracy. However, deeper model architectures and more parameters clearly make the model run slower.

To facilitate comparison, we trained a cosmarium classifier based on Haar Cascade<sup>3</sup> (see Table S2). It is important to note that both training and deploying low-resource-cost models like Haar Cascade offer advantages in terms of reduced computing power requirements. However, it often demands a more extensive dataset to achieve the desired results. In our case, we used the same raw dataset, designating images containing cosmarium as positive samples and the remaining images as negative samples. To create a test set for subsequent model evaluation, we extracted 5% of positive samples and 5% of negative samples from the raw dataset. Considering the typical size of cosmarium in the images, we employed a  $20 \times 20$  window size until reaching an Acceptance Ratio Break Value of  $10^{-6}$ .

In comparison to the YOLOv5-based MSDN, which achieved 87.0% precision and 94.0% recall in cosmarium detection, the Haar Cascade Classifier exhibited much poorer performance. This discrepancy is likely attributed to the relatively small dataset size and the relatively uniform background in the microalgae images provided by AIMP. Therefore, to mitigate the data collection costs for individual microalgae species and facilitate the future inclusion of additional microalgae species, we selected YOLOv5 as an example to showcase the capabilities of AIMP.

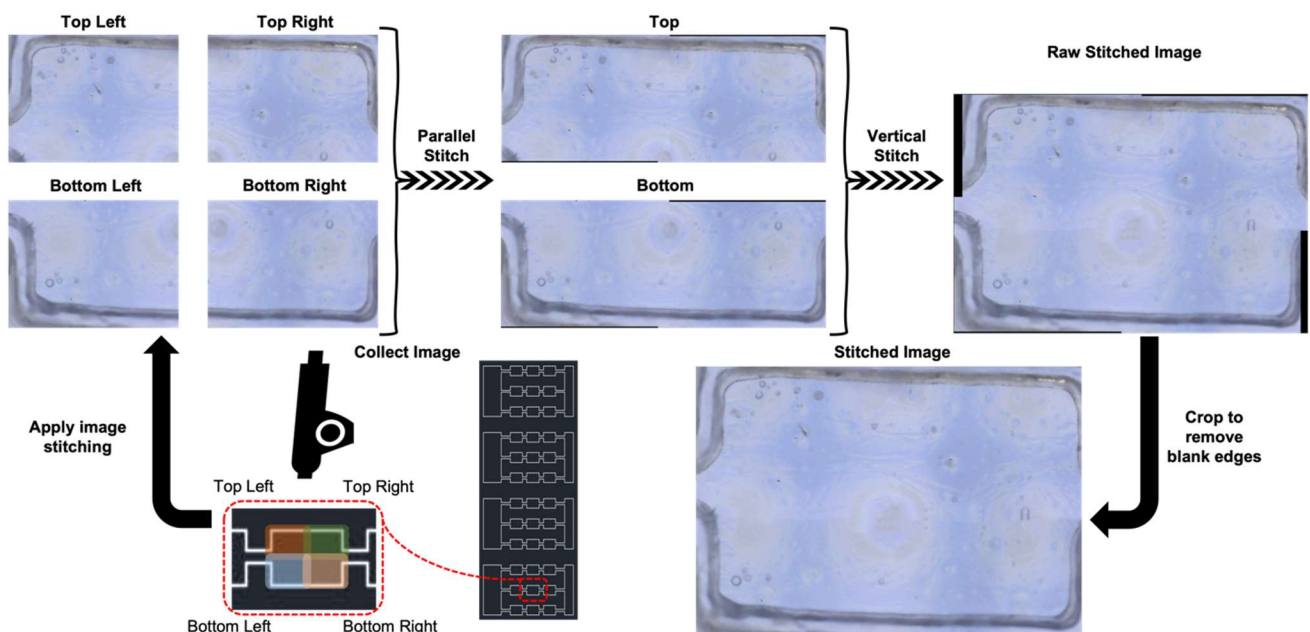
**Table S2.** Evaluation of the trained microalgae species detection model.

<b>Network Architecture</b>	<b>Precision</b>	<b>Recall</b>	<b>F1 Score</b>	<b>mAP@0.5</b>	<b>Speed (Tesla T4 GPU)</b>
YOLOv7	0.946	0.842	0.89	0.902	12.3 ms
YOLOv8n	0.937	0.845	0.89	0.921	7.6 ms
YOLO v8s	0.933	0.863	0.90	0.927	11.6 ms
YOLO v8m	0.948	0.843	0.89	0.915	18.9 ms
YOLOv8l	0.932	0.868	0.90	0.920	34.3 ms
Haar Cascade (Cosmarium Only)	0.562	0.409	-	-	-



## Panoramic Image of an Individual Observation Chamber Recovery

To be able to recover panoramic images of the observation chamber, the AIMP integrates an automatic stitching function (Fig. S4). The USB microscope first takes images of the upper part of the observation chamber by moving along the x-axis. Then, the microscope moves along the y-axis to take images of the lower part of the chamber. This process is repeated until the acquired images cover the entire observation chamber. By using the stitcher tools in OpenCV, each row of captured images is first parallelly stitched together. The resulting horizontally stitched set of images is rotated 90 degrees and then stitched again using the stitcher tool to complete the raw stitched image. The raw stitched image that covers the whole observation chamber then rotates back to its original orientation. As the raw stitched image has black edges due to non-ideal shifts that occur during the USB microscope movement, we use the pixel indexing method to crop the outermost black edges to obtain the final stitched image. The microalgae exhibit minimal movement during the process of generating panoramic images.



**Figure S4.** Automatic stitching function of the AIMP to recover panoramic image of an individual observation chamber.

## Supplementary References

1. C.-Y. Wang, A. Bochkovskiy and H.-Y. M. Liao, presented in part at the Proceedings of the IEEE/CVF Conference on Computer Vision and Pattern Recognition, 2023.
2. G. Jocher, A. Chaurasia, A. Stoken, J. Borovec, Y. Kwon, K. Michael, J. Fang, Z. Yifu, C. Wong and D. Montes, *Zenodo*, 2022.
3. S. Soo, *Institute of Computer Science, University of Tartu*, 2014, **2**, 1-12.