AI-aided at-line microscopy for monitoring of microalgae culture contamination

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Contamination in Microalgal Cultures

Biological contamination is one of the biggest bottlenecks in microalgae cultivation, both in open ponds and in closed photobioreactors, preventing the commercial viability of large-scale processes especially for bulk products. A large number of organisms are known to contaminate microalgae cultures and cause stress, disturb production or even destroy the culture. Early detection of contamination would enable intervention and possibly rescue of the culture, but current monitoring approaches for contaminant detection are largely based on offline methods necessitating manual sampling and expert evaluation, while results are available only with a time delay.

Materials and Methods

A commercially available online microscope (Pixcell measurement device by Pixact, **Fig. 1**) was used to image cultures of *Chlorella vulgaris*, both pure and contaminated with flagellate *Poterioochromonas malhamensis*, dinoflagellate *Oxyrrhis marina* and rotifer *Brachionus plicatilis.* A total of 2870 images in Portable Network Graphics (PNG) format were collected from these cultures. The integrity of the image files was verified with GNU *pngcheck*, and the few identified erroneous files were excluded from further analyses.

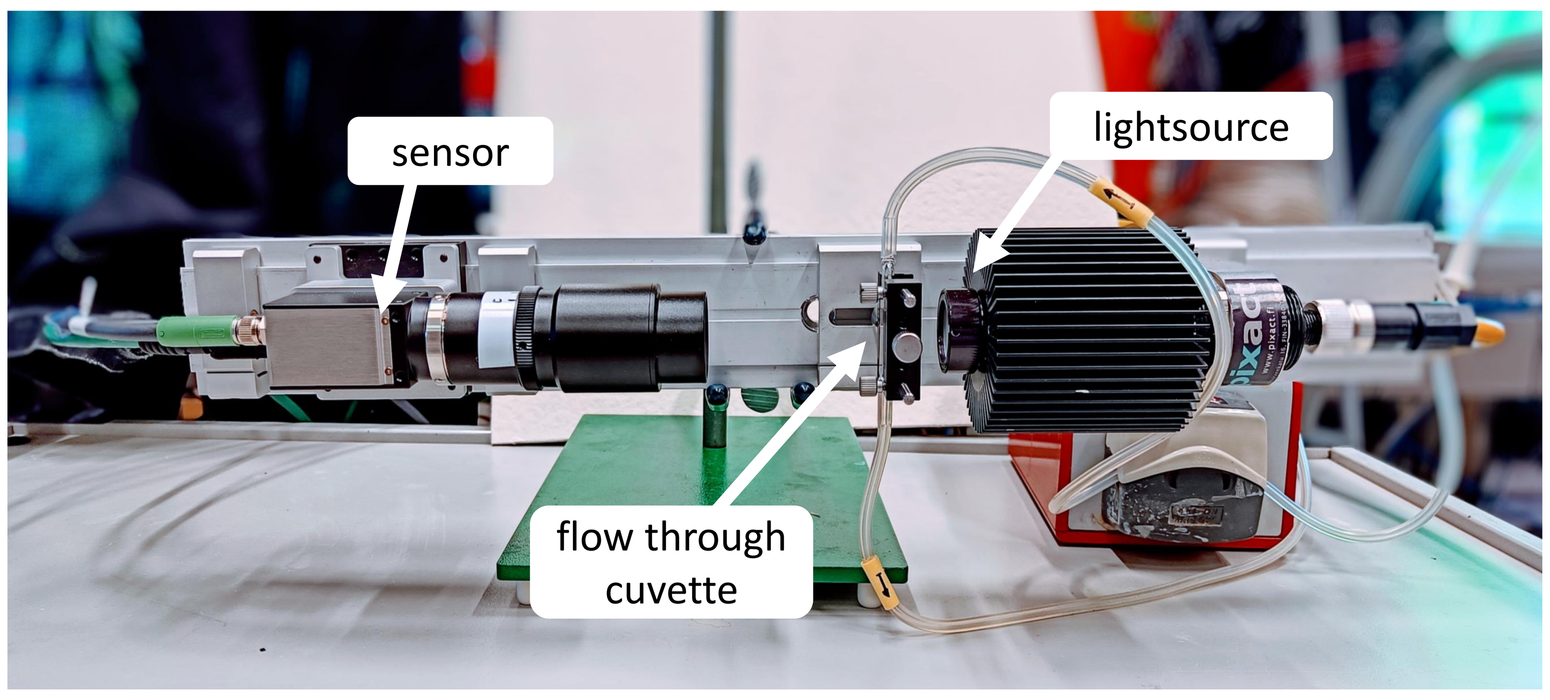


Fig 1. Pixcell measurement device (Pixact).

The remaining data set consisted of a total of 2863 images of pure chlorella (n = 349) cultures and chlorella cultures contaminated with flagellates (n = 527), dinoflagellates (n = 1124) or rotifers (n = 863). The data set was randomly divided into training (805 of all images), validation (10%) and test (10%) sets. A few examples images shown in **Fig 2** together with microscopic images of the organisms..

A convolutional neural network (CNN) model was trained to classify the images into the four classes using the labels in the training data set. The image channel data were rescaled to have a maximum pixel intensity of one unit. The CNN model consisted of a series of seven 2-dimensional convolution (with Rectified Linear Unit or *ReLU* activation) and Maxpooling layers followed by a dense layer of 128 neurons and the output layer of four neurons with *softmax* activation function. The model training used Adam optimization algorithm and categorical cross-entropy as the loss function. A batch size of ten and ten epochs were used for training. The CNN model was trained using Tensorflow v2.12 and Python 3.9.13 on a single Nvidia Tesla V100-SXM2-32GB GPU.

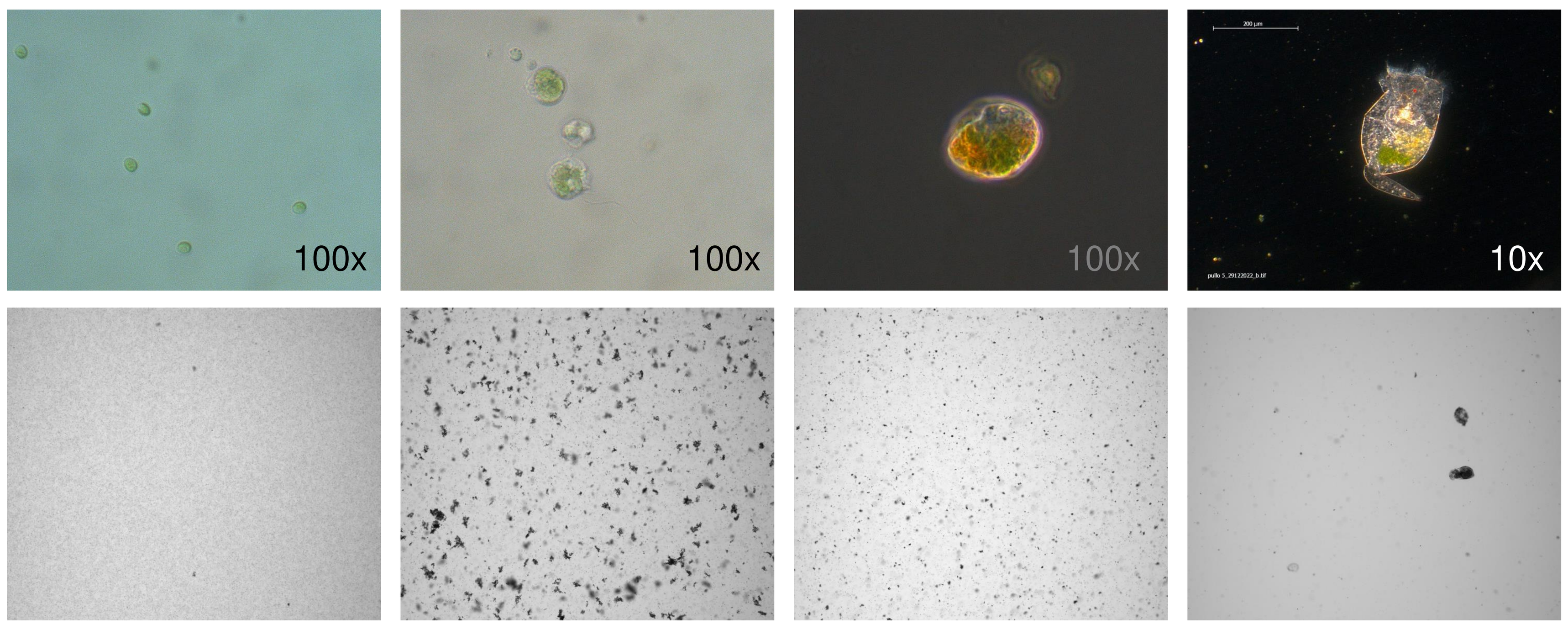


Fig 2. upper row: microscopic pictures of Chlorella culture and the contaminating organisms; lower row: corresponding Pixcell images of pure and contaminated culture, respectively.

Results and Discussion

The predictive performance of the CNN model during the training is shown in **Fig 3** in terms of the predictive accuracy and categorical cross-entropy loss as a function of epochs. The overall predictive performance of the model on training, validation and the test data set are summarized in **Table 1**. With the accuracy of 0.979 on the preliminary test data, the model demonstrates good generalizability. The future work will focus on verifying the generalizability of the model on a larger collection of images and improving the proof-of-concept model presented here.

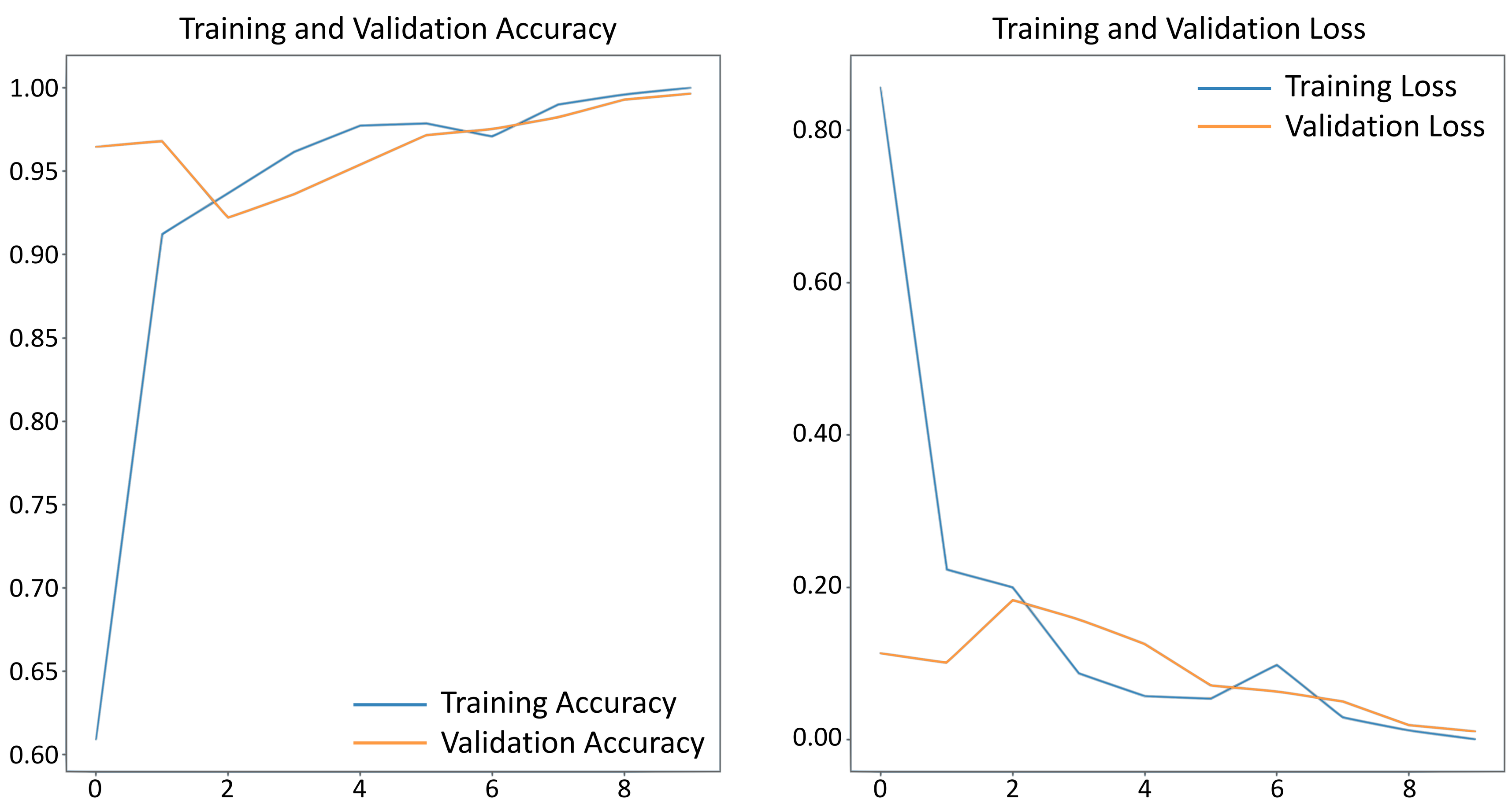


Fig 3. Classification accuracy as function of number of epochs during training of the Convolutional Neural Network (CNN) model

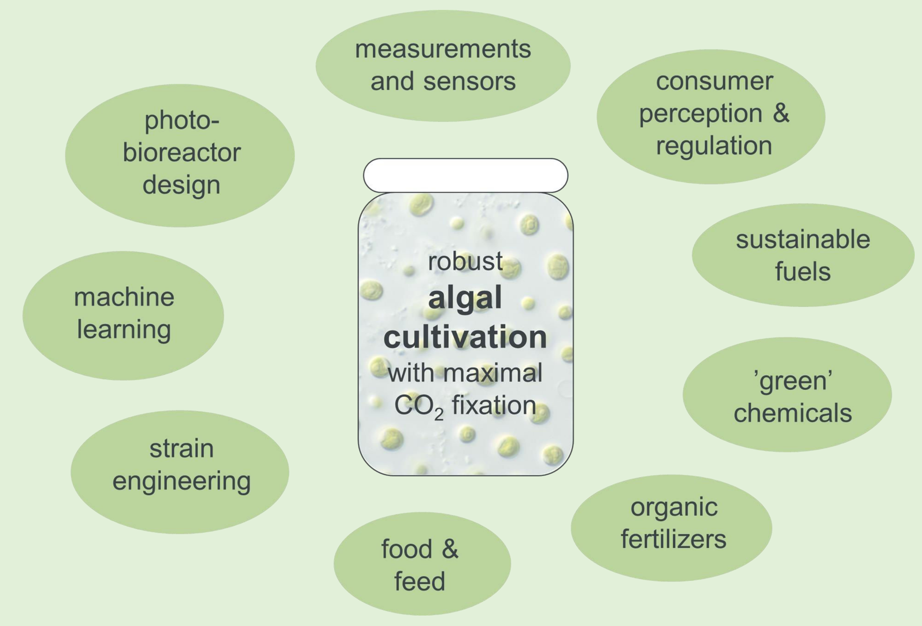
Table 1. Predictive performance of the CNN classifier on training, validation and test data sets presented in terms of the average accuracy over multi-class classification and the categorical cross-entropy loss.

Data set (size)	Accuracy	Loss
Training (2291)	0.979	0.0536
Validation (290)	0.954	0.0976
Test (282)	0.979	0.0506

About project ROBA

ROBA is a 2-year (1/2022-12/2023) Business Finland funded project (grant ID 6563/31/2021) with a budget of 1.4 M€, coordinated by VTT. **ROBA aims to develop intensification methods for large scale algal cultivations**, that allow optimization of CO_2 uptake and algal biomass production: an on-site algae pilot frame is outlined in terms of reactor design using CFD modelling, AI assisted monitoring is improving robustness of algae cultures, while crude techno-economic and life cycle assessments ensure viability of the process.

The project partners (research partners: VTT, Jyväskylä University; company partners: Agri-Biotech, Aircohol, Neste, Owatec, Timegate, Pixact, Soilfood, Specim) **combine excellence and skills** in bioprocess engineering, multi-product biorefinery concepts, bioreactor design, measurement technologies, process equipment development, applications from algal biostreams and more. ROBA establishes new collaborations between companies to explore **CO_2 mitigation potential and algae derived products**.



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