
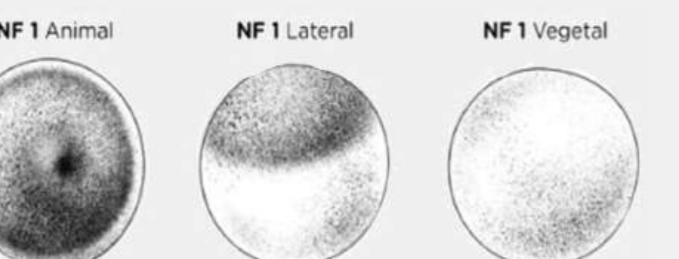

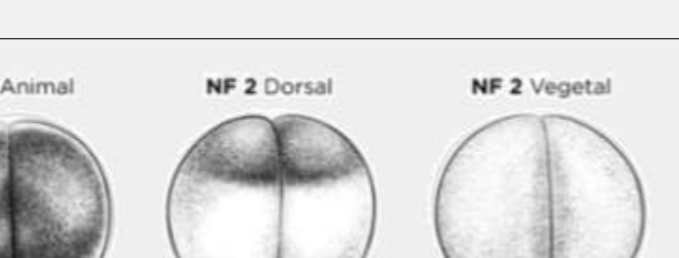
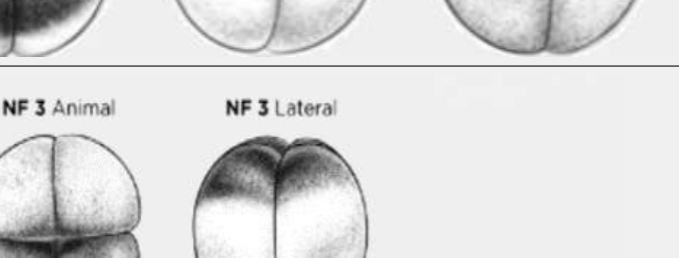
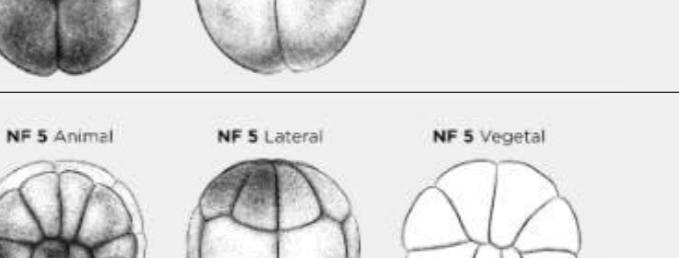



Introduction

Category	Class	Xenopus	Real Image
Unfertilized	S0	Ovulated unfertilized egg	
	S1	NF 1 Animal, NF 1 Lateral, NF 1 Vegetal	
	Sa	Abnormal unfertilized egg	
Fertilized	S2	NF 2 Animal, NF 2 Dorsal, NF 2 Vegetal	
	S4	NF 3 Animal, NF 3 Lateral	
	S8	NF 5 Animal, NF 5 Lateral, NF 5 Vegetal	
	Sb	Not properly developed fertilized egg	

- In order to accurately enumerate *Xenopus laevis* embryos [3] at various developmental stages within a Petri dish, we use a multi-class Machine learning algorithm based on Stardist, [2] and [5]. This is an ongoing project with the Aquatic Germplasm Genetic Research Center (AGGRC) and Marine Biological Laboratory (MBL).
- According to Nieuwkoop and Faber (1994), these embryos have been classified into blastomeres during distinct developmental stages, referred to as NF.

Objectives

- Our goal is to provide AGGRC and MBL researchers with a simple and reliable application for effective multiclass classification of embryos. We have tailored Stardist model that classifies embryos into fertilized (NF-2, NF-4, and NF-6) and unfertilized categories.

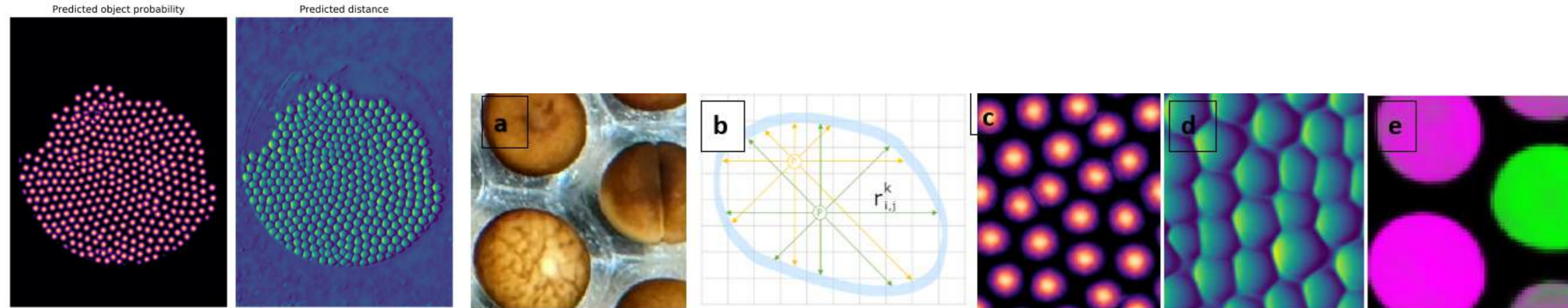


Figure 1. Stardist Predicted Probabilities and Distances

Architecture-Stardist and Multi Class

- The Stardist model utilizes a neural network that is built on the U-net architecture, object probability, and radial distances.
- Figure (1) illustrates the Stardist architecture: (a) a segmentation image; (b) star-convex polygons parameterized by the radial distances $r(i,j)$; and (c) the object probabilities $d(i,j)$.
- In Figure (1), (d) depicts the representation of a distance map of the predicted objects, and (e) shows the final instances are processed via non-maximum suppression (NMS).
- The dataset provided by AGGRC has been preprocessed, with developmental stages annotated using LabKit in FIJI.

Distance and Probability Loss

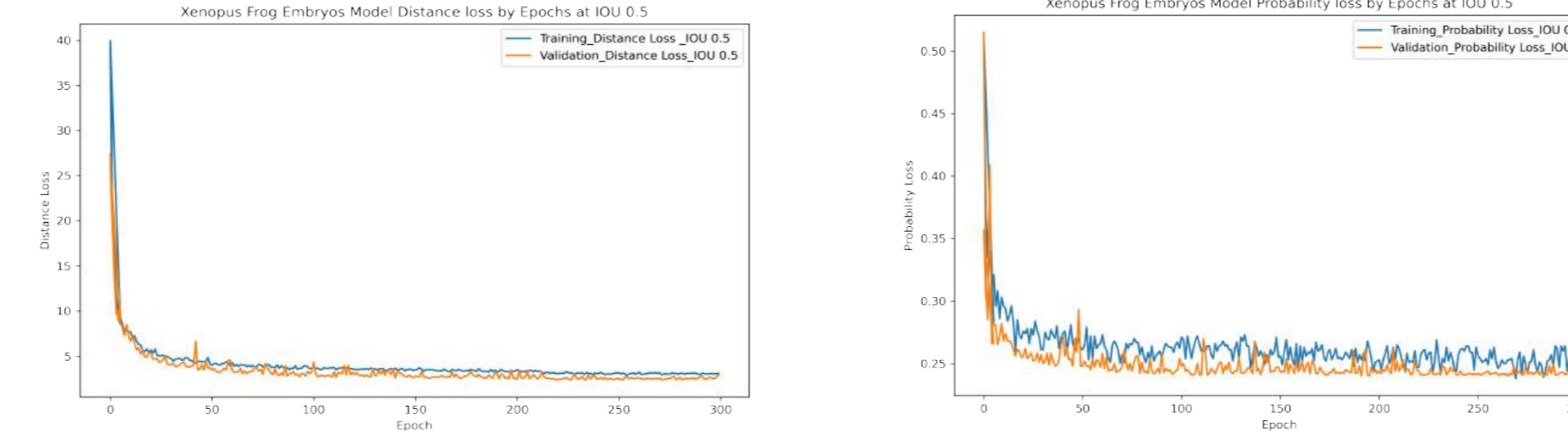


Figure 2. Distance and Probability Loss

- The first graph shows that training and validation losses began at 30 and decreased to 5 at epochs 50, 100, 150, 200, 250, 300.
- The second graph shows the probability loss, with both training and validation losses beginning at 0.6, reducing to 0.3 by epoch 10, and decaying to 0.25 around epoch 100. The dataset was divided into 80-20 ratios for training and testing, with 20 percent for validation from the training ratios.
- This signifies enhanced model performance and learning capability, resulting in more accurate predictions.

Metrics of F1 score, Precision, Accuracy and Recall over Epochs

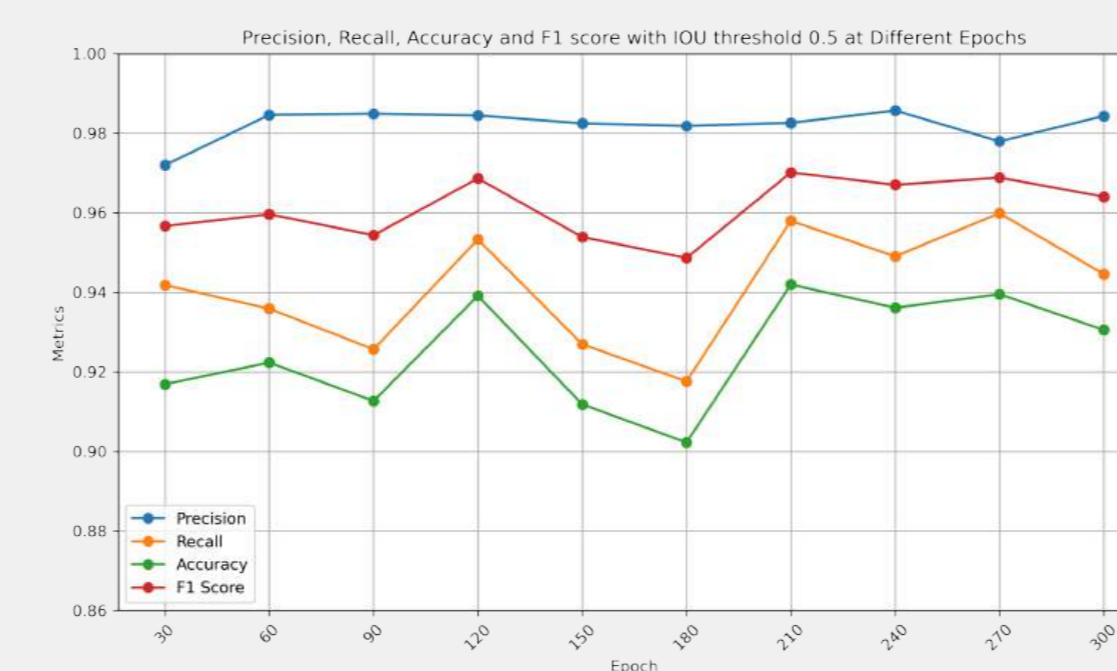


Figure 3. Illustration of the Metrics-F1 score, Precision, Accuracy, and Recall on Epochs 50 to 250

- The graph depicts the variations in F1 score, Accuracy, Recall, and Precision for varying epochs during the training of U-Net CNN architecture and StarDist. The models were evaluated using an Intersection over Union (IoU) threshold equal to 0.5.

GUI

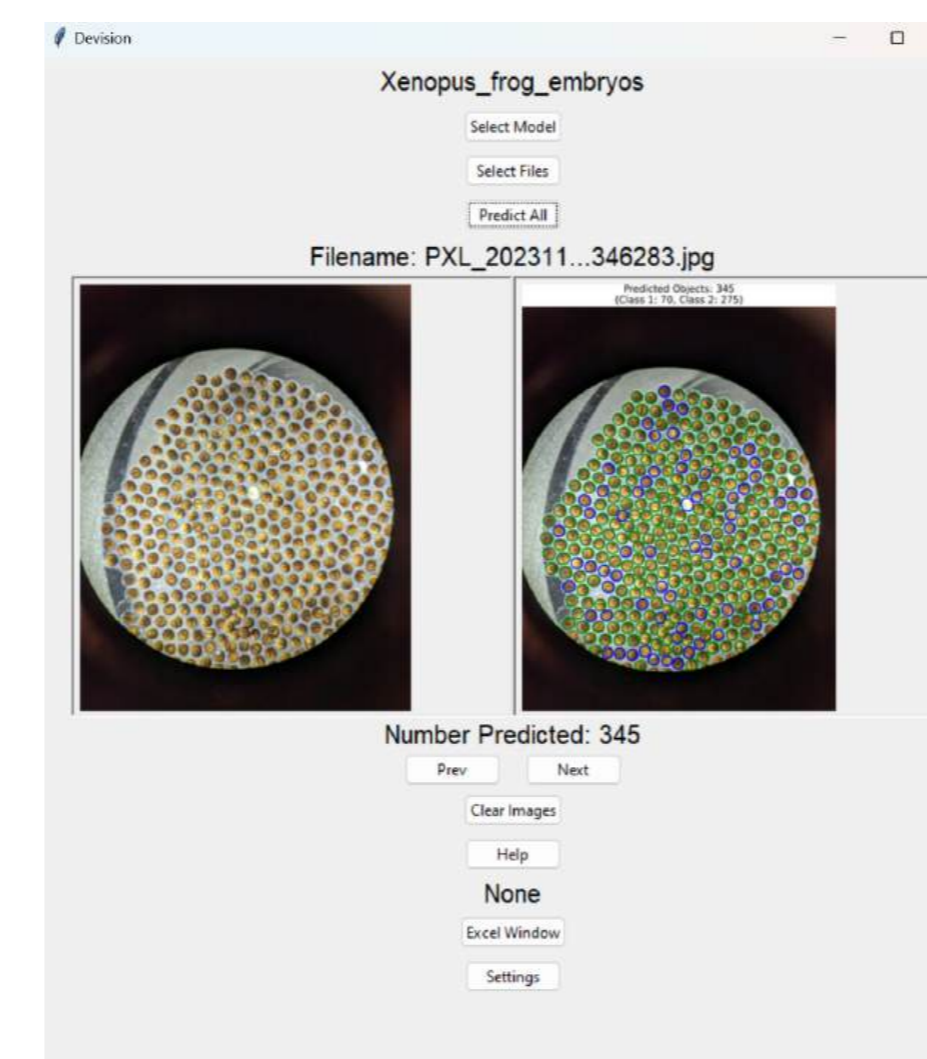


Figure 4. GUI-Tool for Prediction

- We have developed a user-friendly graphical user interface (GUI) for our models.
- This GUI facilitates the prediction process by providing object counts using a counting model and both counts and classifications using a classification model, based on the input images.

Multi-Class Prediction of Xenopus Frog Embryos

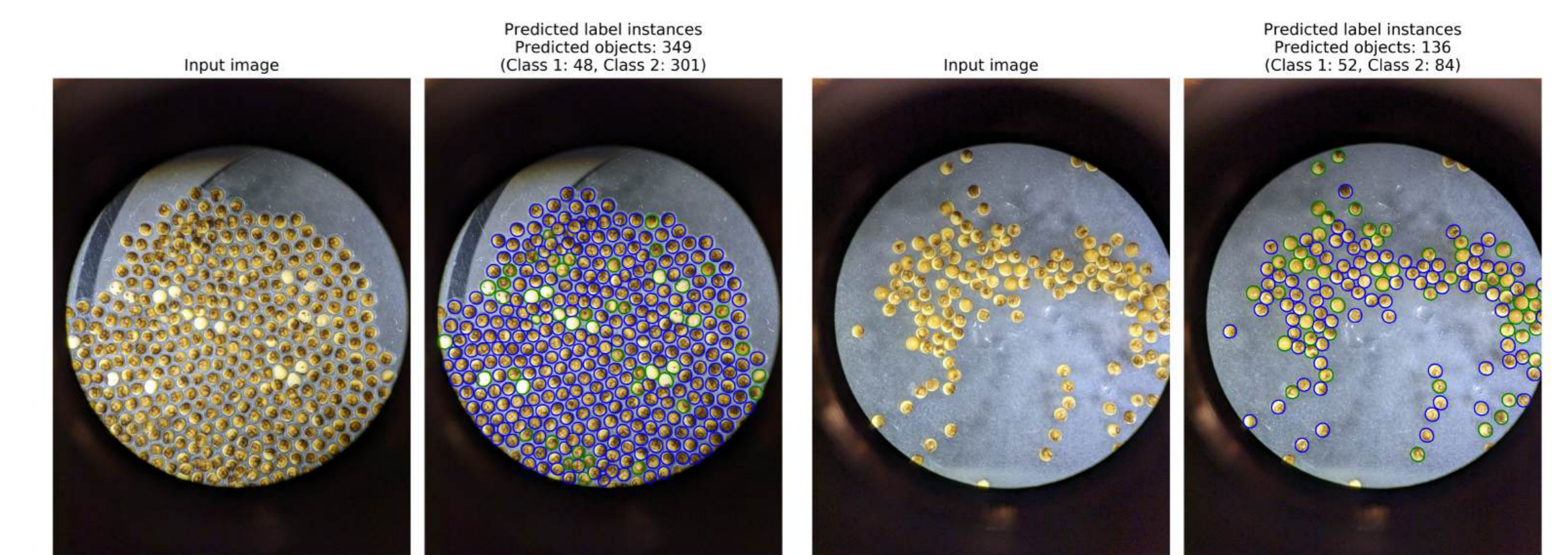


Figure 5. Predictions on Different Classes and Counts

- The prediction image shows the predicted *Xenopus laevis* embryos (left) count to be 349, classified into class 1 (green) as unfertilized with a count of 48, and class 2 (blue) as fertilized with a count of 301.
- The predicted image on the right shows a count of 136 embryos, with class 1 (green) unfertilized being 52 and class 2 (blue) fertilized being 84.

Future work

- We aim to develop the Stardist model to effectively handle multiple stages (S2, S4, S6).
- Additionally, we plan to focus on building a stardist Multi class model on different stages of *Xenopus* frog embryos, particularly stages 15 to 21, which includes neural groove formation. This will involve a detailed examination of the neural folds of the embryo.
- Another goal is to use GUI as a tool for different classification methods making it accessible to the general public.
- We will also produce a video demonstrating how to operate the GUI.

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- We want to acknowledge and thank Nikkos Svoboda, Computer Analyst for introducing and making us familiar with the workings of the high-performance computing system "Chaos".

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