



An introduction to Deep Learning

Part I - Fundamentals

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Goal of the training :

- Understand what an **Artificial Neural Network (ANN)** is and what are the main parameters to characterize them
- What is a **Convolutional Neural Network (CNN)** and why is it used for image processing
- What are the **fundamentals for building and training a CNN using Keras**
- Understand the **most common applications** and **where to find the tools for your applications**

Outline :

- I. Deep Learning : applications for image analysis**
- II. General introduction & definition of neural networks
- III. Example #1 : single neuron application
- IV. Example #2: building a multi-layer network

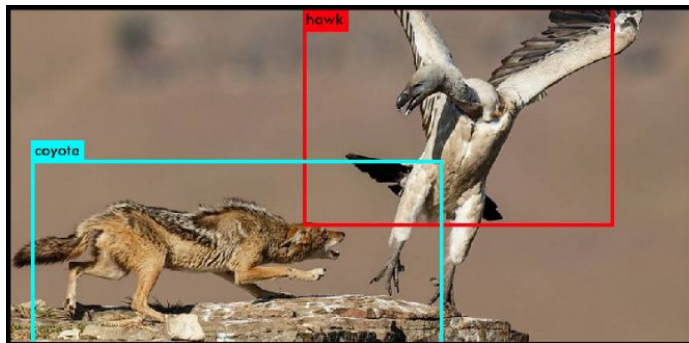
Most common applications for image analysis:

1- Image classification :

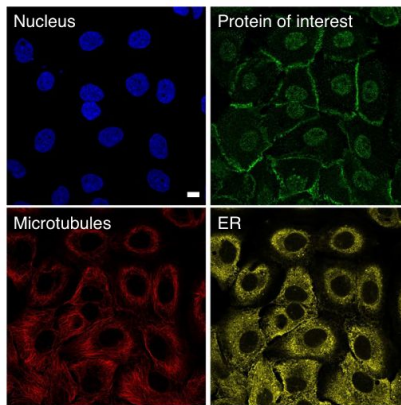


Pl@ntNet

<https://plantnet.org/en/>



Redmon & Farhadi - 2016 YOLO9000, better, faster, stronger.
Von Charmier et al. - 2020 ZeroCostDL4Mic: an open platform to use Deep-Learning in Microscopy.
<https://github.com/HenriquesLab/ZeroCostDL4Mic>



Classifier
→

Multi-label prediction

Nucleoplasm
Cytosol
Plasma membrane
Nucleoli
Mitochondria
Golgi apparatus
Nuclear bodies
Nuclear speckles
Nucleoli fibrillar c.
Centrosome
Cell junctions
Actin filaments
...

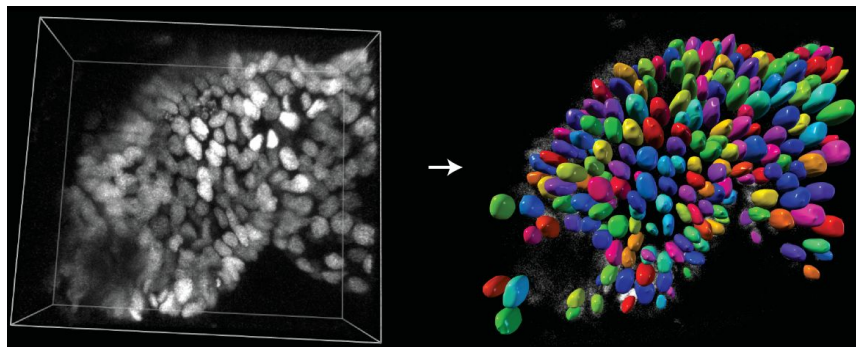
Ouyang et al. - 2019 Analysis of the Human Protein Atlas Image Classification competition.

Most common applications for image analysis:

1- Image classification :

2- **Image segmentation :**

2D / 3D segmentation of objects



<https://github.com/stardist/stardist> - Schmidt et al. - 2018 Cell Detection with Star-Convex Polygons

<https://github.com/hci-unihd/plant-seg> - Wolny et al. - 2020 Accurate and versatile 3D segmentation of plant tissues at cellular resolution

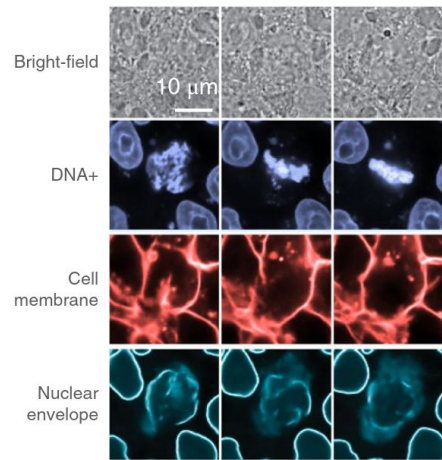
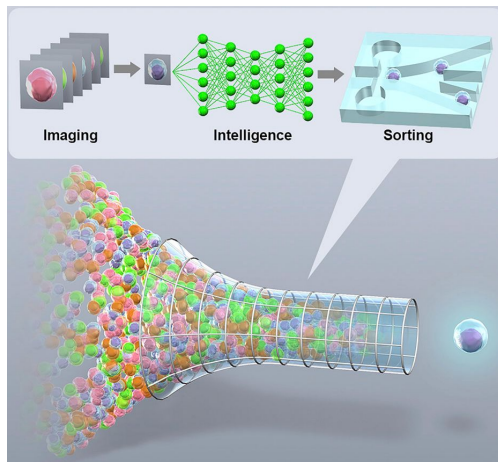
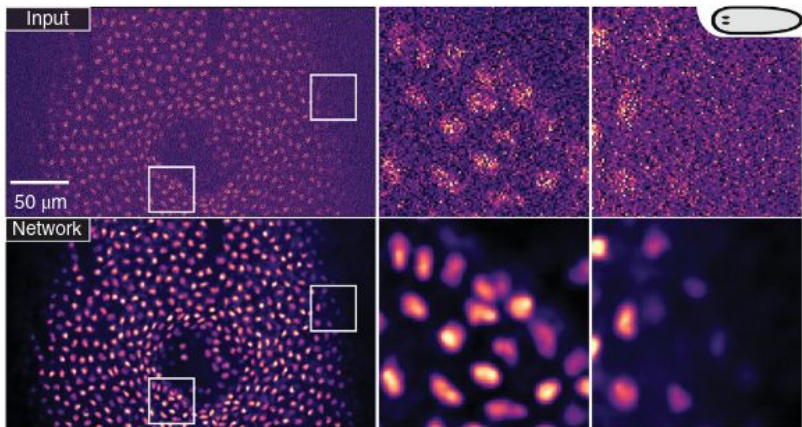
<https://github.com/MouseLand/cellpose> - Stringer et al. 2021 Cellpose: a generalist algorithm for cellular segmentation

<https://github.com/kevinjohncutler/omnipose> - Cutler et al. - 2022 Omnipose: a high-precision morphology independent solution for bacterial cell segmentation

<https://github.com/vanvalenlab/intro-to-deepcell> - Greenwald et al. - 2022 Whole-cell segmentation of tissue images with human-level performance using large-scale data annotation and deep learning

Most common applications for image analysis:

- 1- Image classification :
- 2- Image segmentation :
- 3- **Augmented microscopy :**



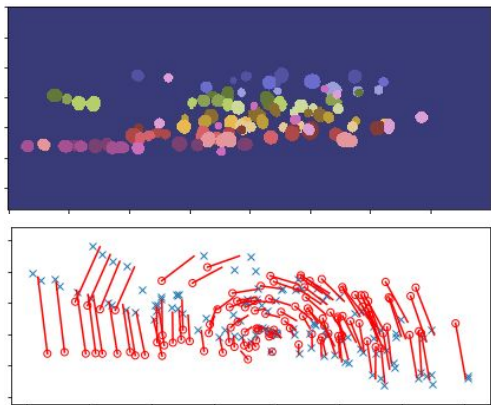
<https://github.com/CSBDeep/CSBDeep> - Weigert et al. 2017. Content-aware image restoration: pushing the limits of fluorescence microscopy

Nitta et al. 2018. Intelligent Image-Activated Cell Sorting

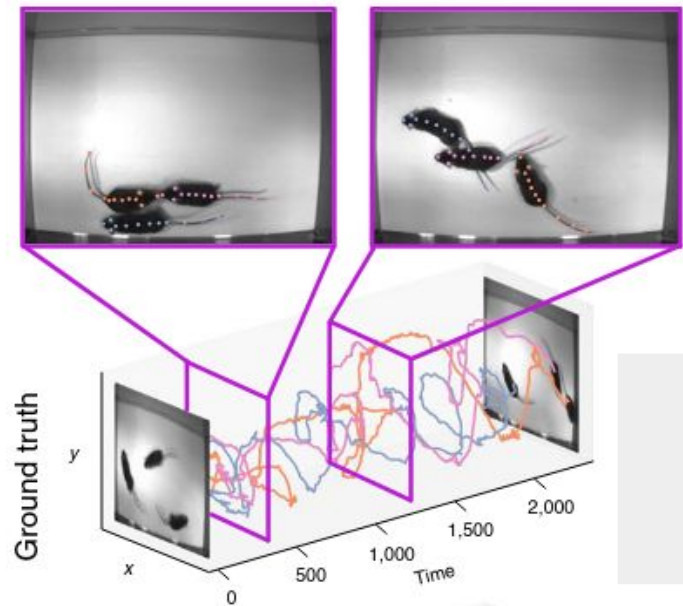
Ounkomol et al. 2018. Label-free prediction of three-dimensional fluorescence images from transmitted-light microscopy

Most common applications for image analysis:

- 1- Image classification :
- 2- Image segmentation :
- 3- Augmented microscopy :
- 4- Tracking :**

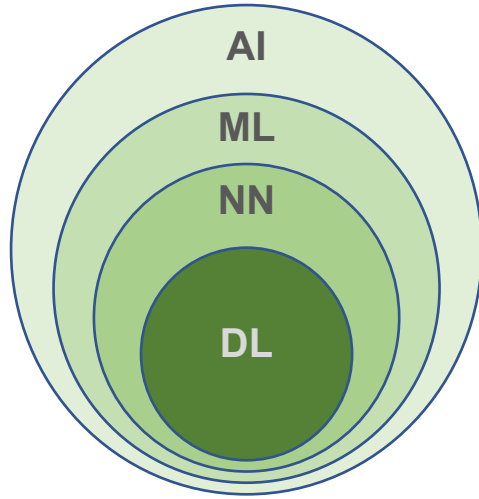


Wen et al. - 2021 3DeeCellTracker, a deep learning-based pipeline for segmenting and tracking cells in 3D time lapse images

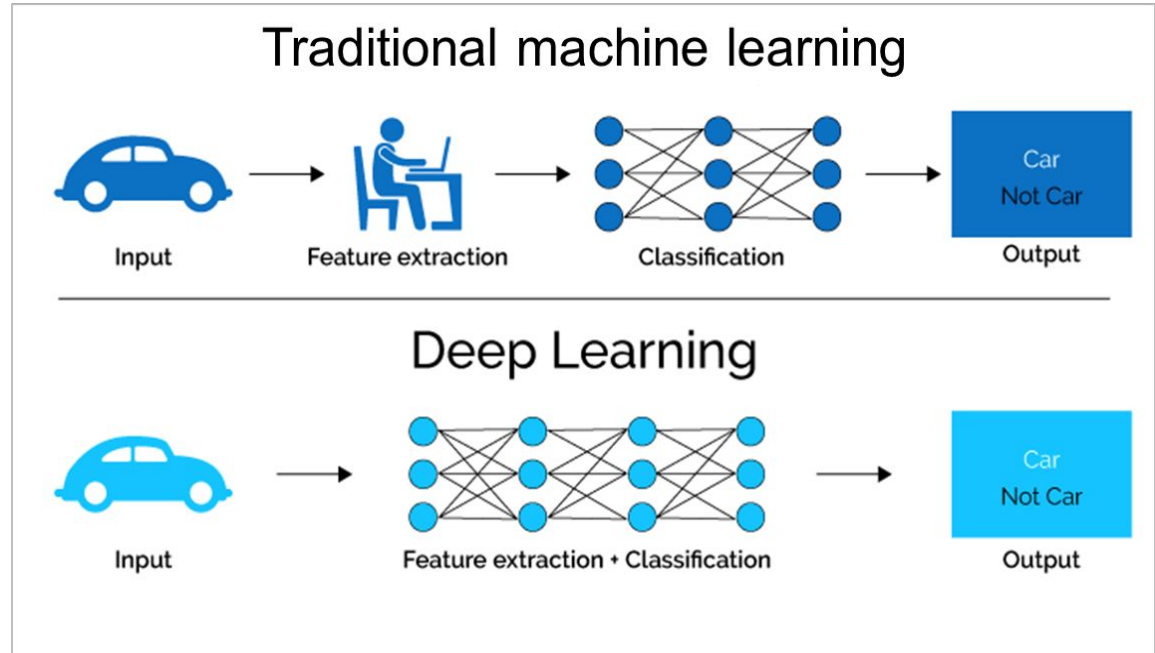


Lauer et al. - 2022 Multi-animal pose estimation, identification and tracking with DeepLabCut

Machine learning vs. Deep Learning :



AI = artificial intelligence
ML = machine learning
NN = neural network
DL = deep learning



When & why using Deep Learning?

When **classic image processing/analysis tools** are not efficient or do not exist for the task we want to perform (e.g. high throughput segmentation)



Need to have enough **analyzed & good-quality** data to train the network



Need to label the data in order to get database large enough for the training

Time consuming



Network are trained for a specific set of data. New type of data means new training.

Not (always) flexible



Deep Learning needs large computational resources for image analysis

Expensive

How to start with Deep Learning (for free)?



Python 3 – open source

For DL, the open-source
TensorFlow and **PyTorch**
libraries are used.



Colab (google)
free GPU
python jupyter

+



<https://www.youtube.com/c/DigitalSreeni>

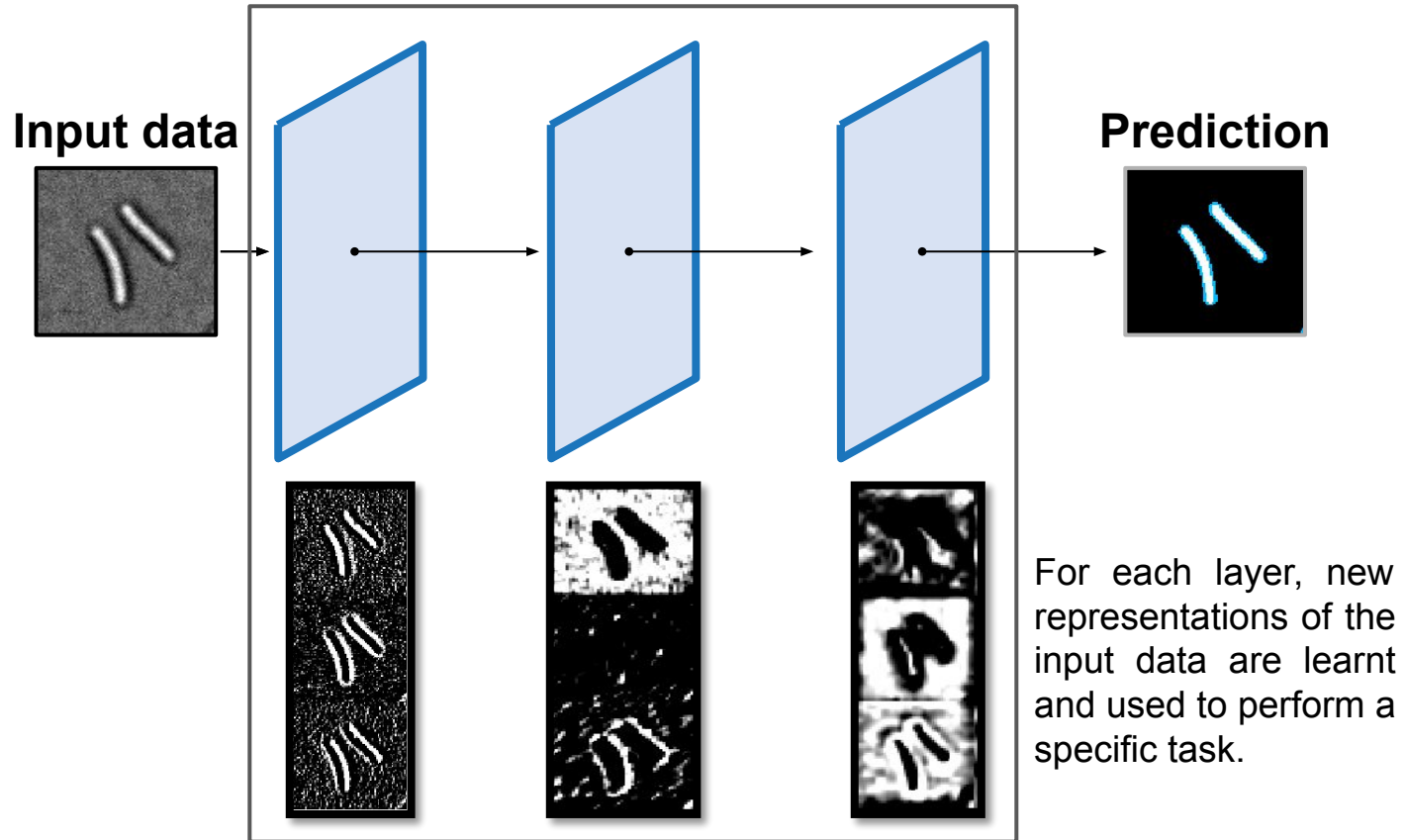
<https://www.youtube.com/c/CNRSFormationFI/DLE?app=desktop>

<https://cs230.stanford.edu/lecture/>

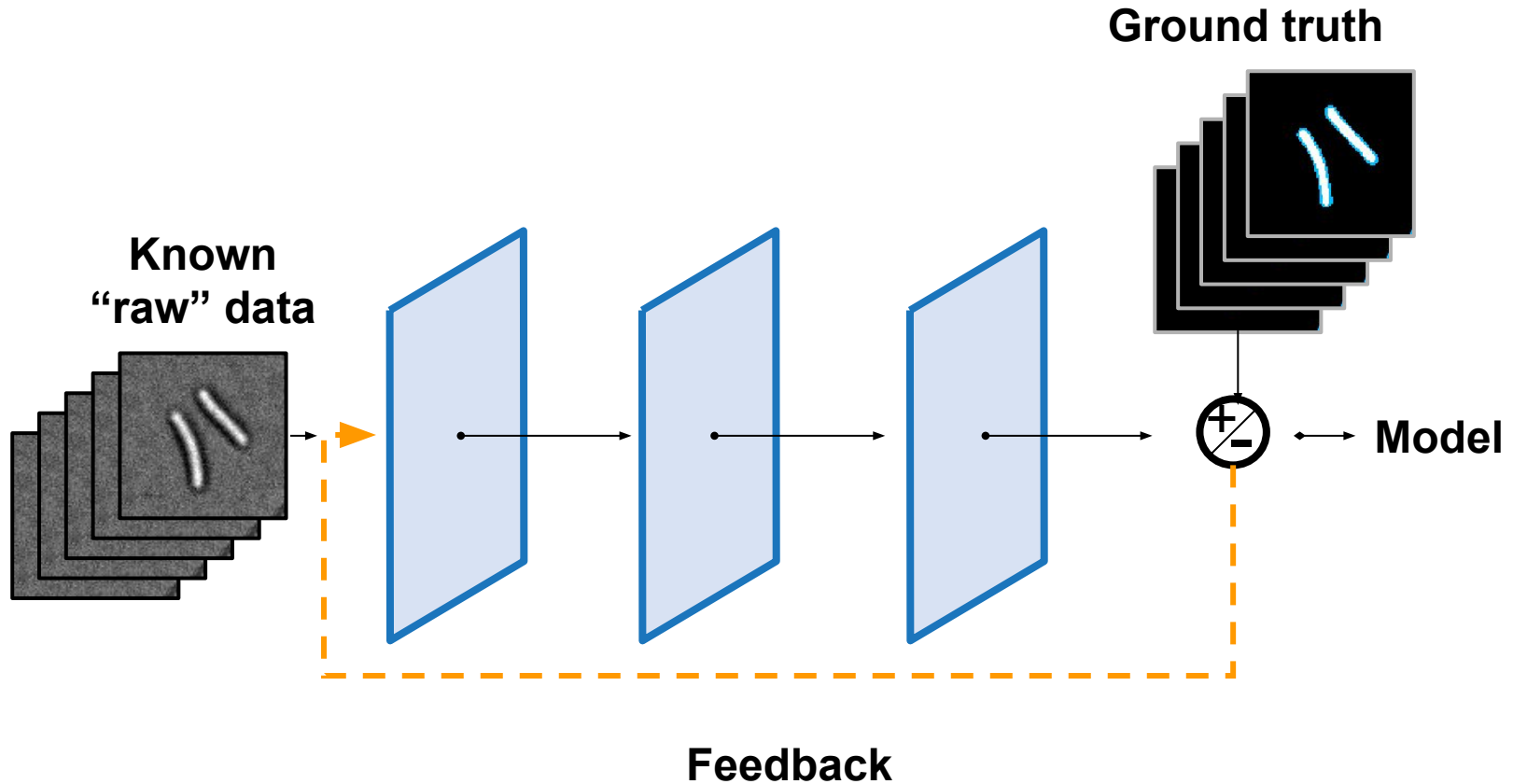
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- II. General introduction & definition of neural networks**
- III. Example #1 : single neuron application
- IV. Example #2: building a multi-layer network

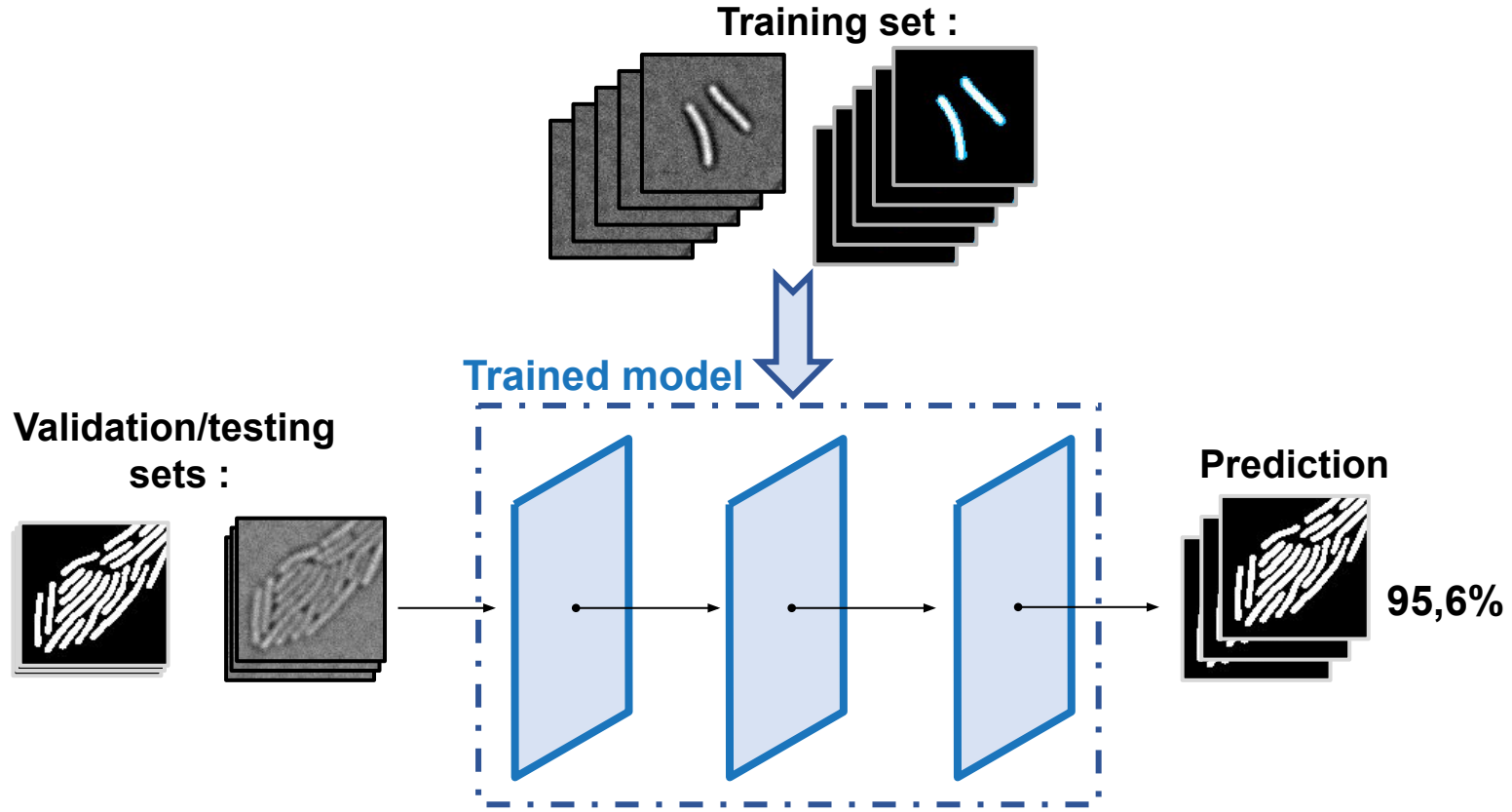
Deep Learning : why “Deep”?



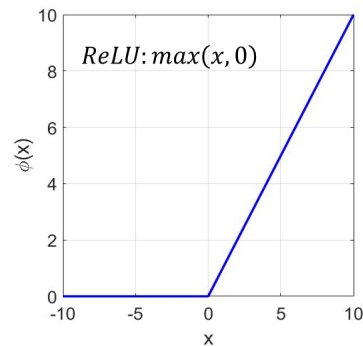
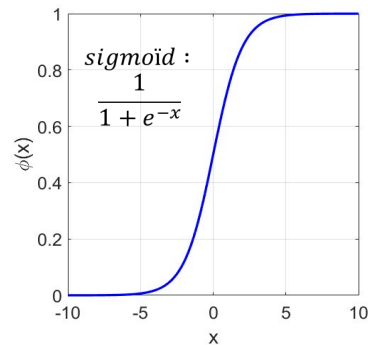
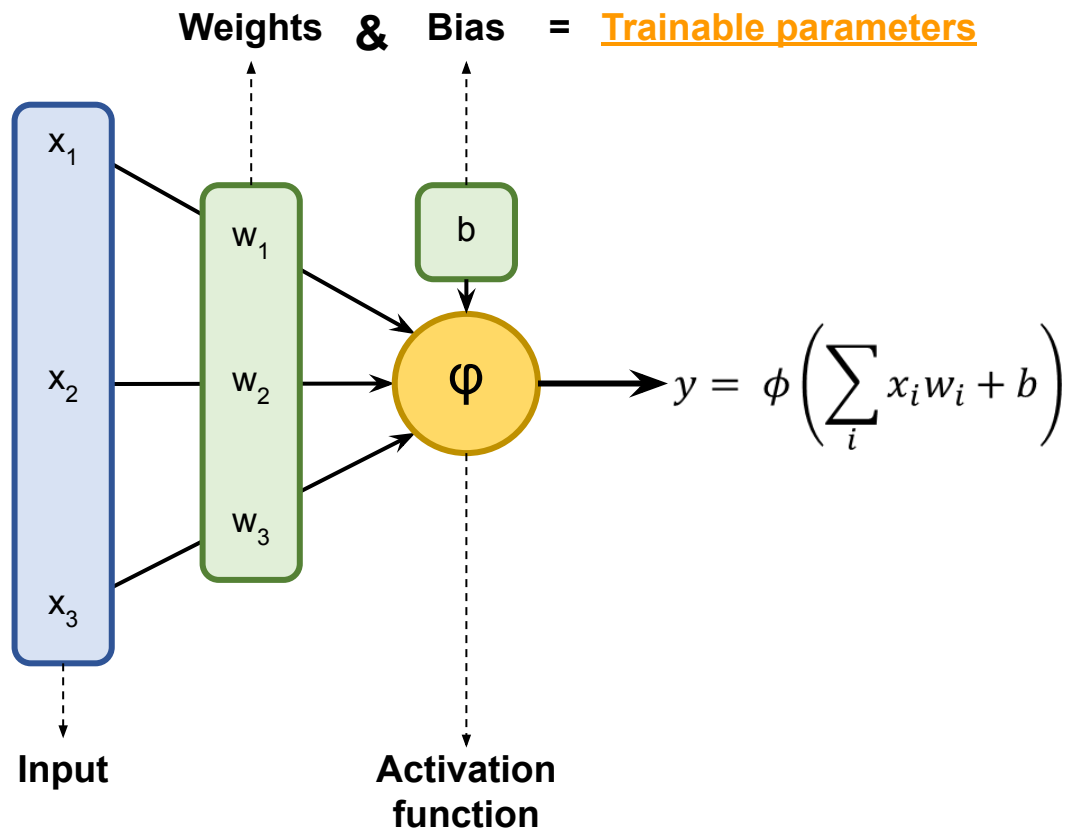
“Learning” under supervision :



Supervised deep learning network :

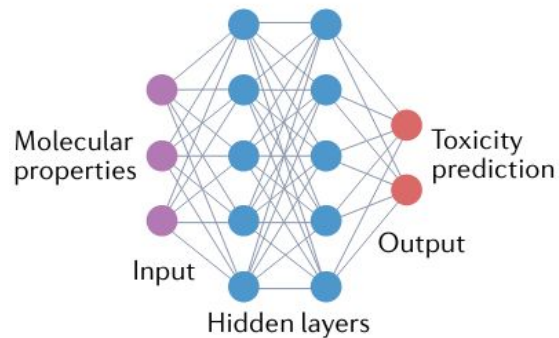


Definition of a single neuron

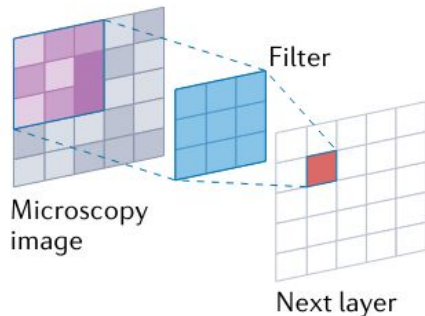


Many network architectures

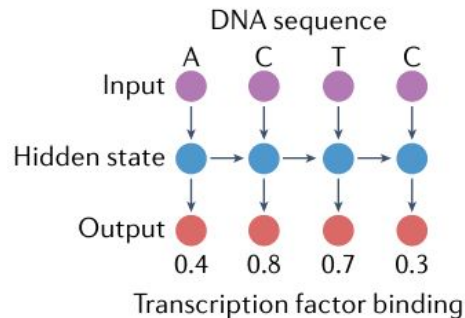
a Multilayer perceptron



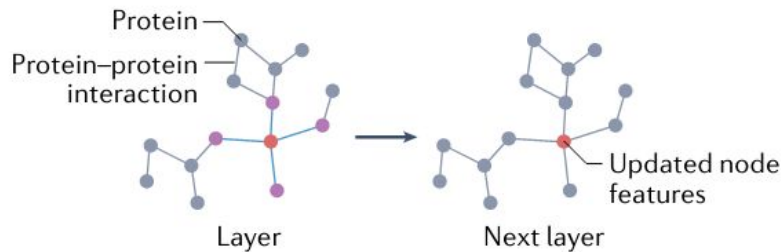
b Convolutional neural network



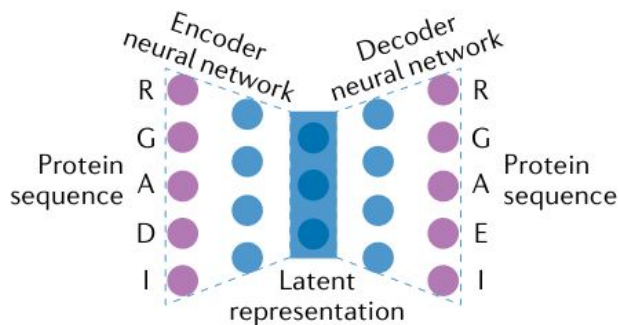
c Recurrent neural network



d Graph convolutional network

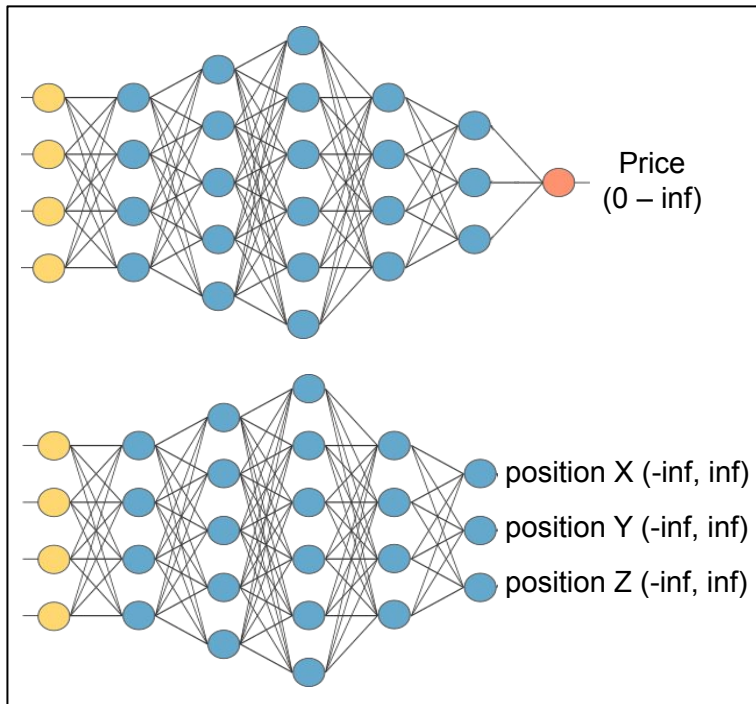


e Autoencoder

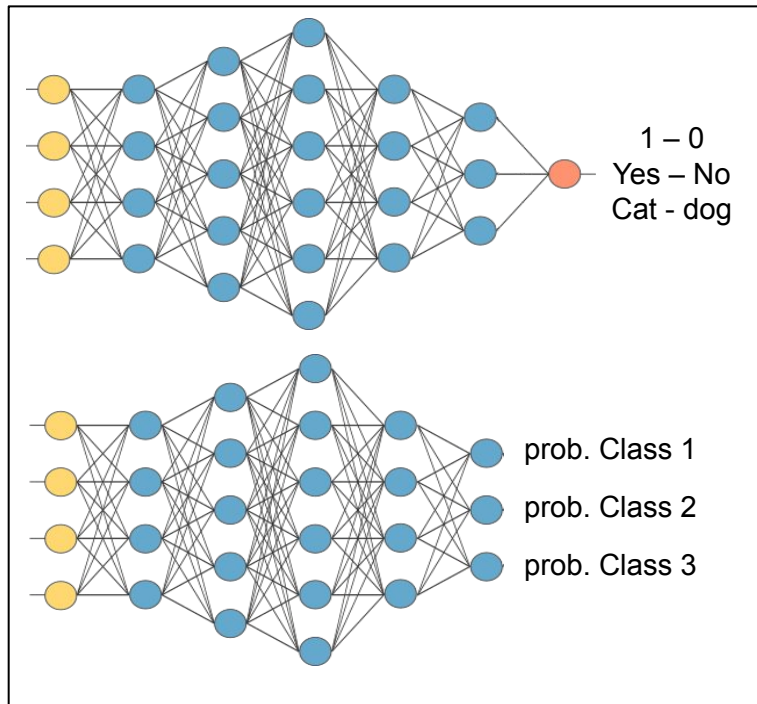


Regression vs. Classification

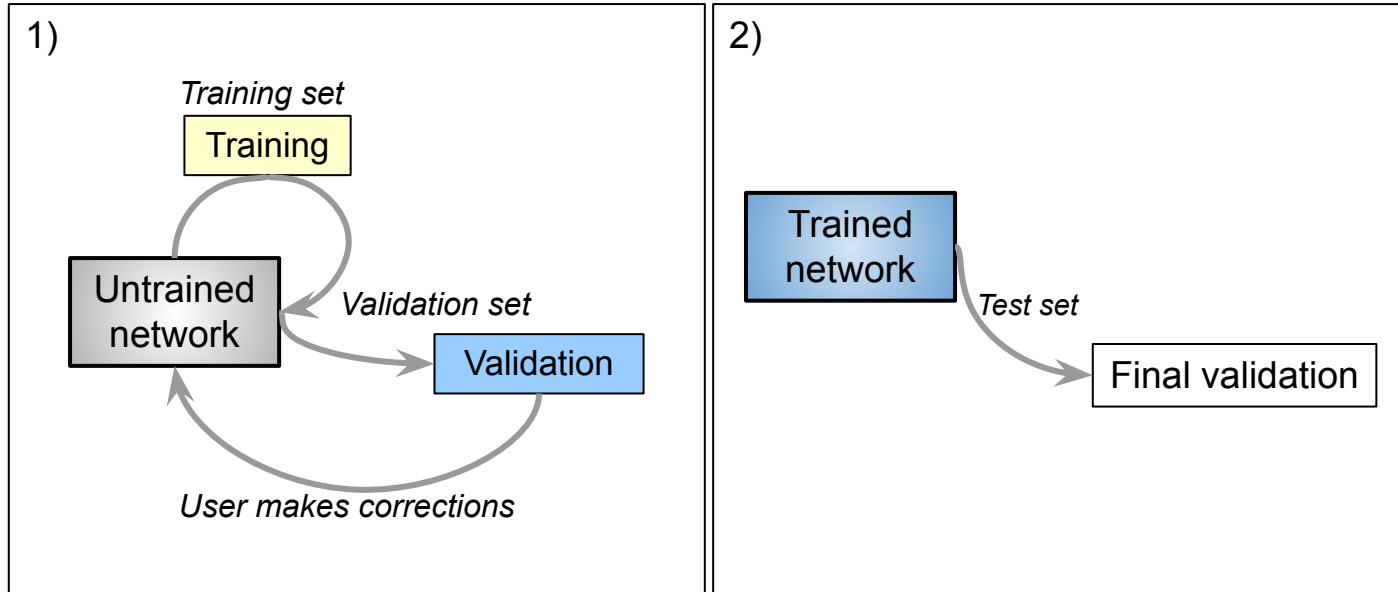
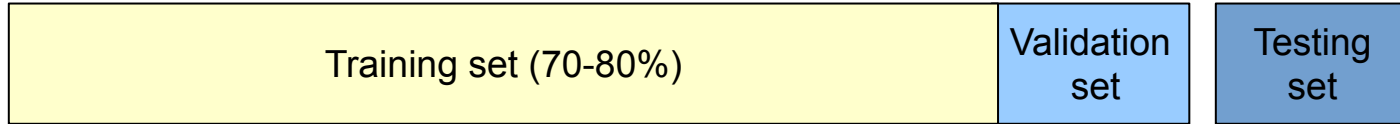
Regression : output is one or more real numbers



Classification : output is the probability that input belong to one or more classes



Training, testing and validation sets



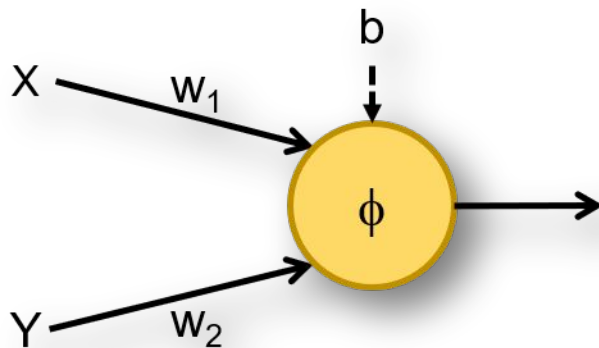
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- IV. Example #2: building a multi-layer network

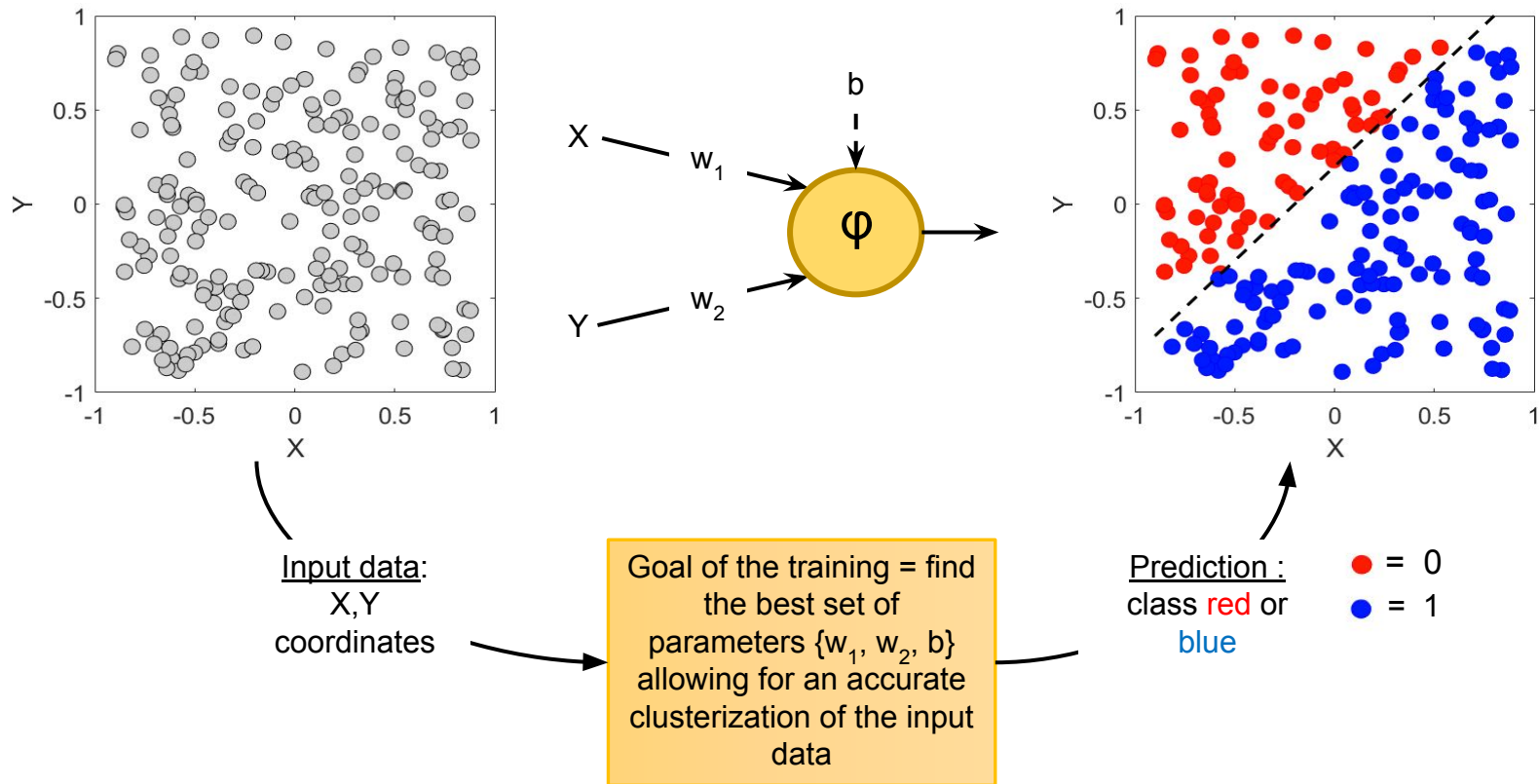
Train a single neuron classifier

Example n°1: Ex1_Clusterization_linearly_separated.ipynb

1. Understand the principle of the training
2. Train the classifier and test its accuracy
3. First step with Keras/TensorFlow



Train a single neuron classifier



Definition of a classifier with Keras

1- Definition of the network architecture

```
from keras import models
from keras.models import Sequential
from keras.layers import Dense

model = Sequential([
    Dense(1,activation='sigmoid', input_shape=(2,))
])
```

2- Definition of the training options

```
model.compile(optimizer='adam',
              loss='binary_crossentropy',
              metrics=['accuracy'])
```

3- Training

```
history = model.fit(Training_data,
                    Training_label,
                    epochs = 100,
                    validation_data = (Validation_data, Validation_label))
```


Definition of a single neuron

1- Definition of the network architecture

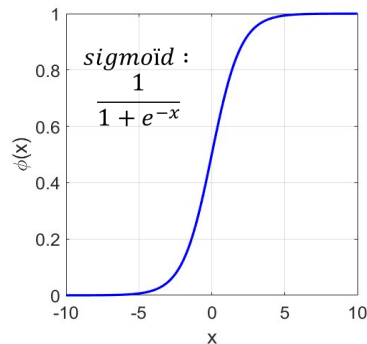
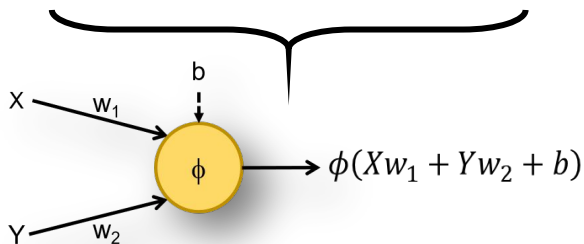
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```

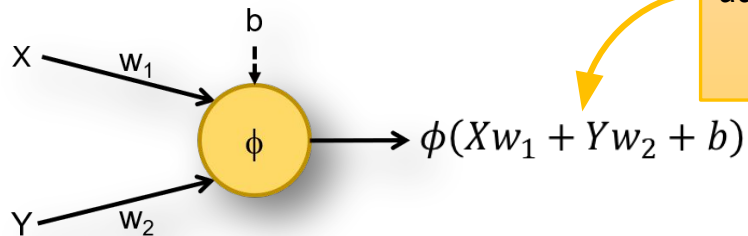
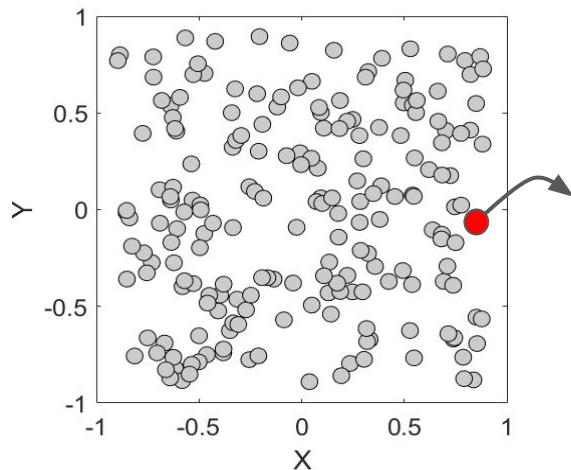
of neurons

ϕ

X,Y

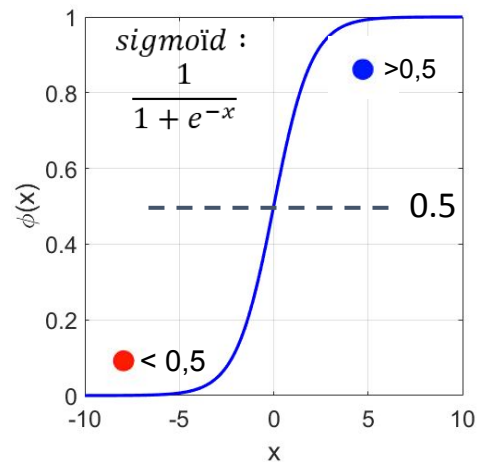


How the neuron works ?



Here we choose the **sigmoid** as activation function and a "prediction" is calculated

The weights are randomly initialized and the bias set to zero.



Definition of the training options

1- Definition of the network architecture

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from keras import models
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])
```

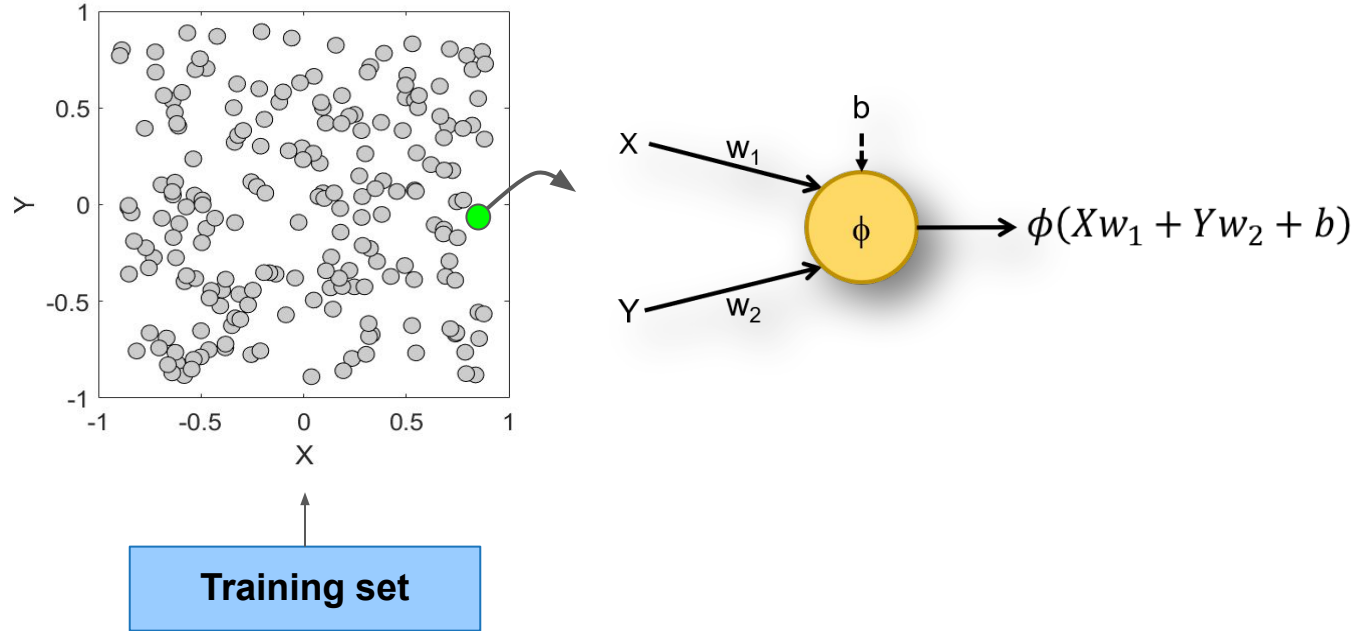
2- Definition of the training options

```
model.compile(optimizer='adam',
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              metrics=['accuracy'])
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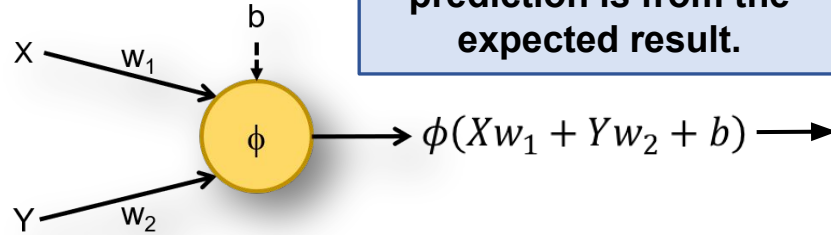
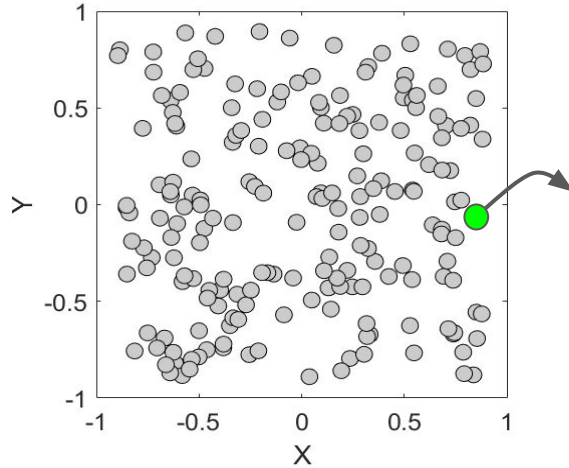
3- Training

```
history = model.fit(Training_data,
                    Training_label,
                    epochs = 100,
                    validation_data = (Validation_data, Validation_label))
```

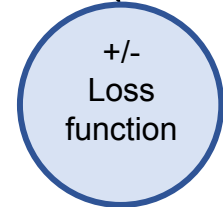
Compiling : defining training process



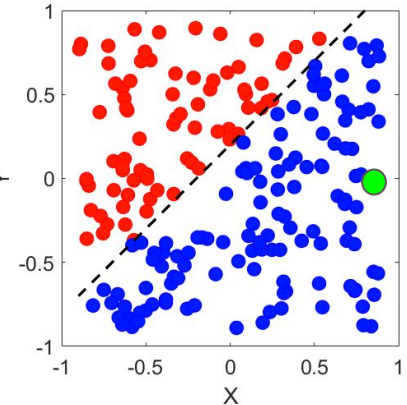
Compiling : defining training process



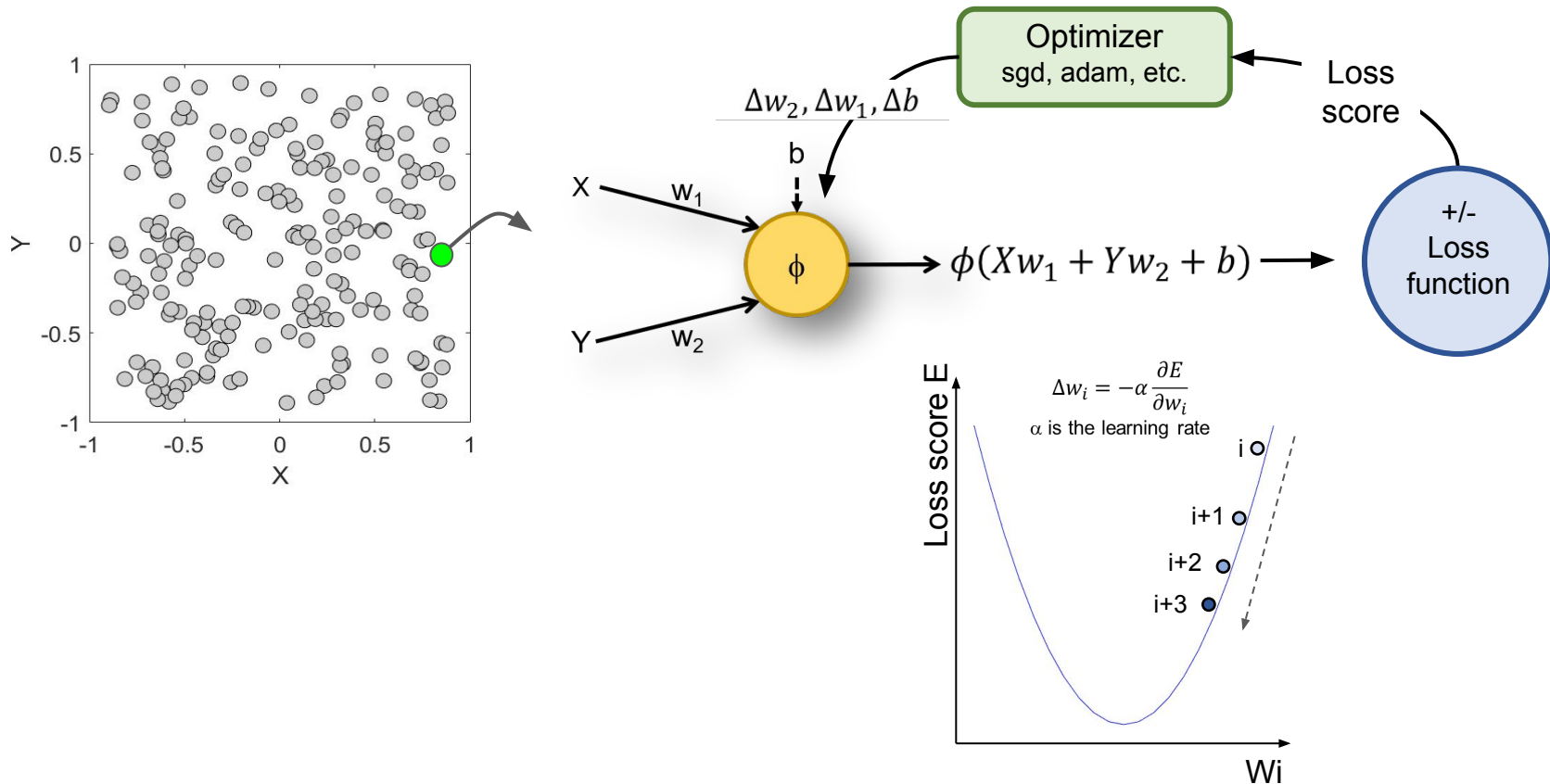
The **loss function** is used to measure how far the prediction is from the expected result.



The **neuron output** is compared to the **expected result**
=
ground truth



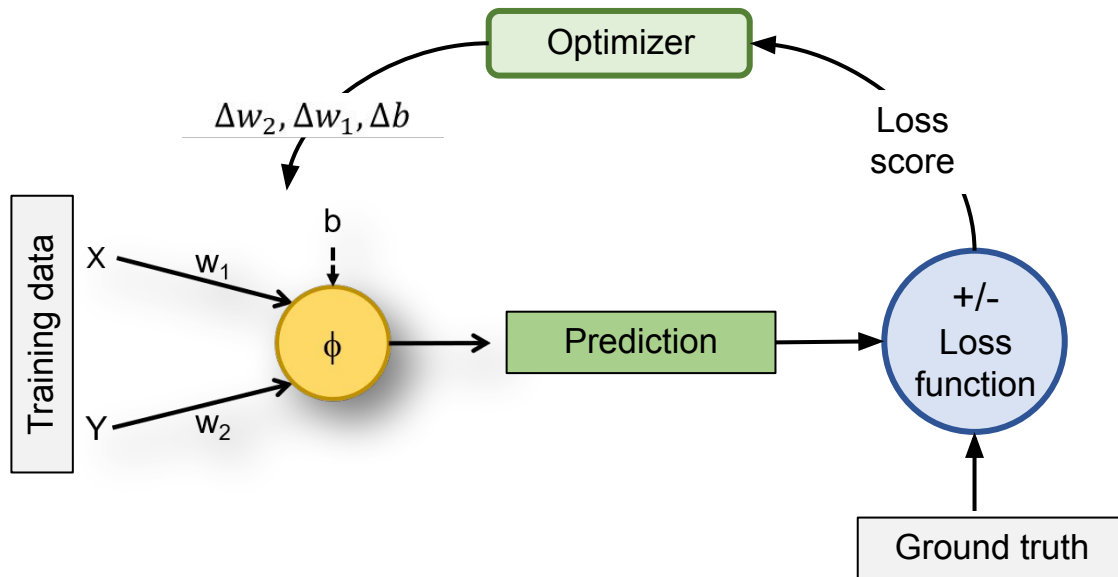
Compiling : defining training process



Model compiling

2- Definition of the training options

```
model.compile(optimizer='adam',  
              loss='binary_crossentropy',  
              metrics=['accuracy'])
```



Launching the training

1- Definition of the network architecture

```
from keras import models
from keras.models import Sequential
from keras.layers import Dense

model = Sequential([
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```

2- Definition of the training options

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model.compile(optimizer='adam',
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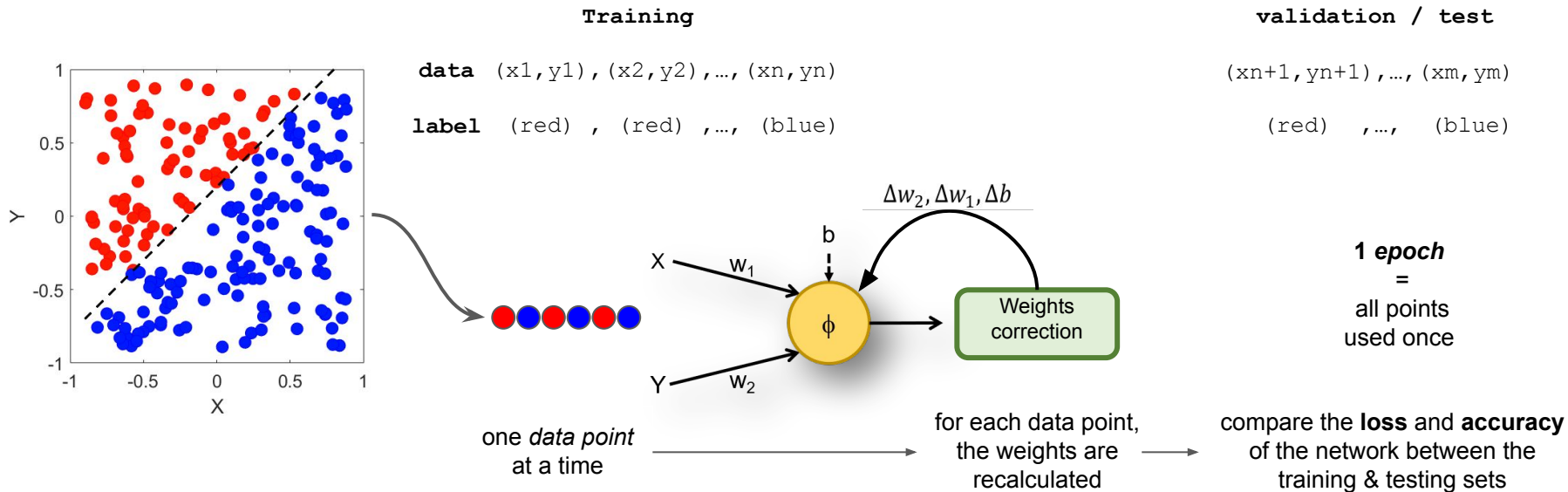
3- Training

```
history = model.fit(Training_data,
                    Training_label,
                    epochs = 100,
                    validation_data = (Validation_data, Validation_label))
```

Start the training

3- Training

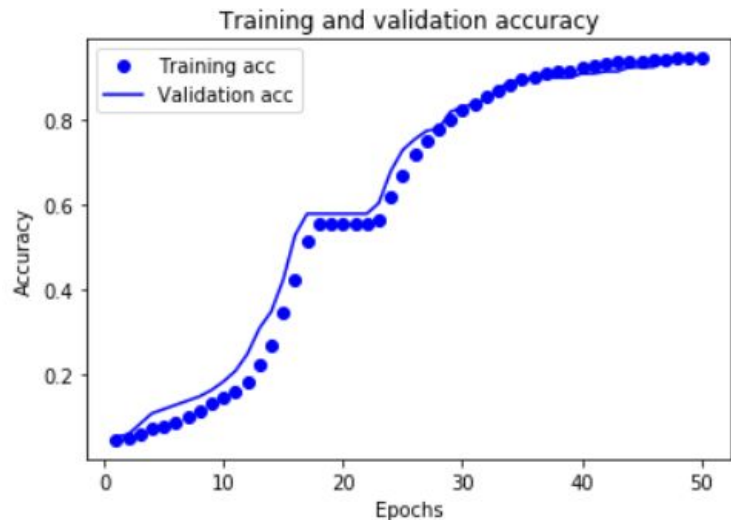
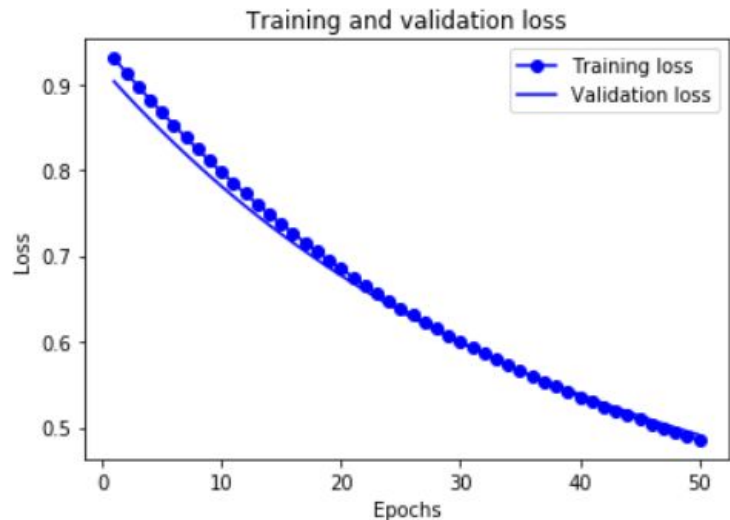
```
history = model.fit(Training_data,  
                    Training_label,  
                    epochs = 100,  
                    validation_data = (Validation_data, Validation_label))
```



Training results

3- Training

```
history = model.fit(Training_data,  
                    Training_label,  
                    epochs = 100,  
                    validation_data = (Validation_data, Validation_label))
```



Summary

1- Definition of the network architecture

```
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from keras.models import Sequential
from keras.layers import Dense

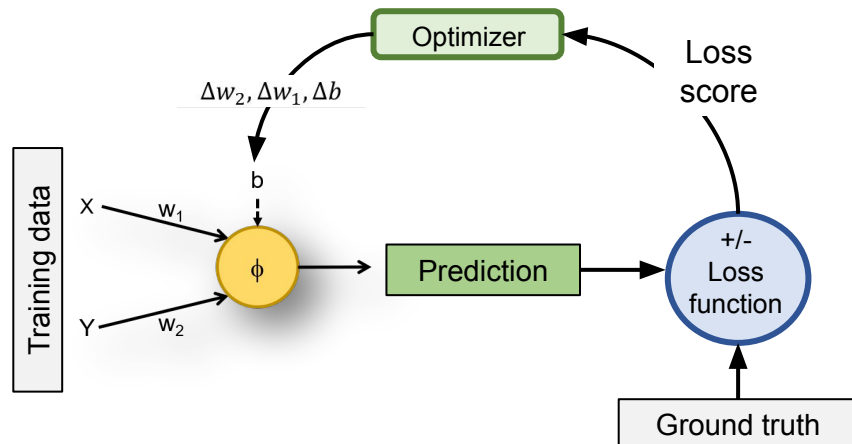
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])
```

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```
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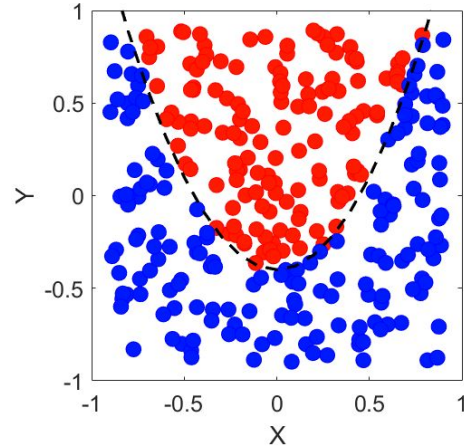
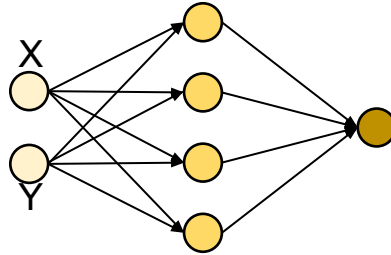
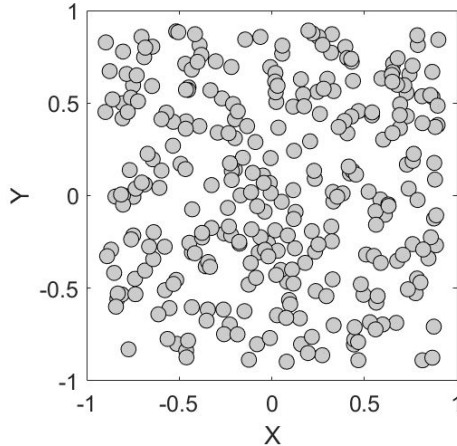
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Example 2: classify non-linearly separable data

Example n°2: Clusterization_not_linearly_separated_parabole

1. Observe the limitations of single-layer model
2. Find a simple architecture able to solve this classification problem



Multiple layers :

Activation function: try sigmoid or relu

1st layer needs input specified

number of neurons

1st layer

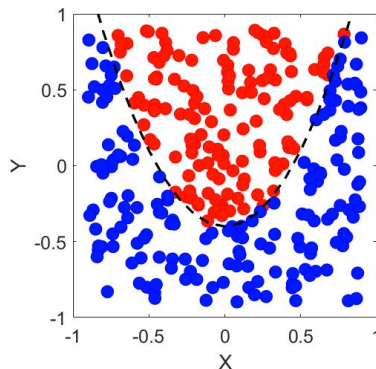
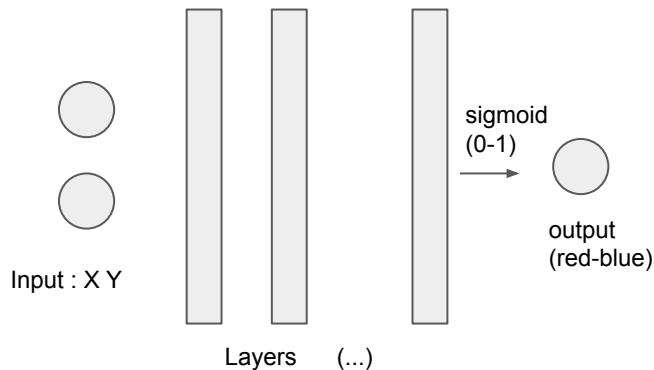
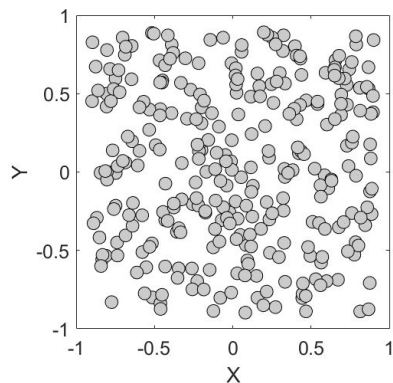
2nd layer

...

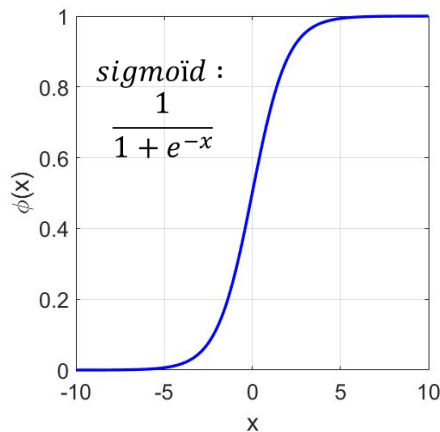
```
from keras.models import Sequential
from keras.layers import Dense

model = Sequential([
    Dense(32, activation='relu', input_shape=(2,)),
    Dense(16, activation='relu'),
    Dense(1, activation='sigmoid')
])

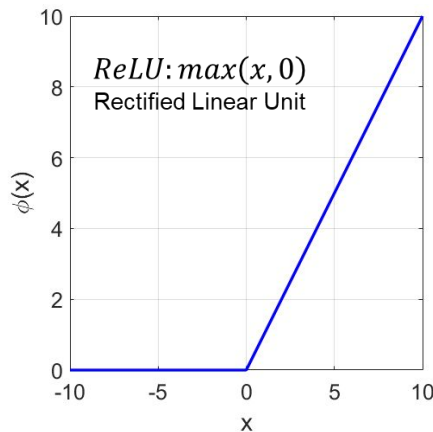
model.compile(optimizer = 'adam',
              loss='binary_crossentropy',
              metrics=['accuracy'])
```



The two most popular activation function



Gradient is saturating for large output values and $\exp()$ is a computer-expensive operation

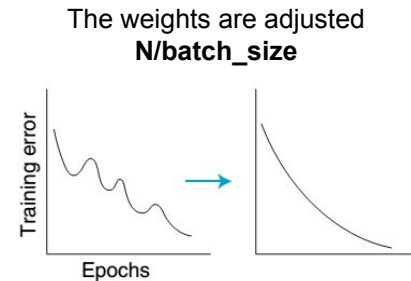
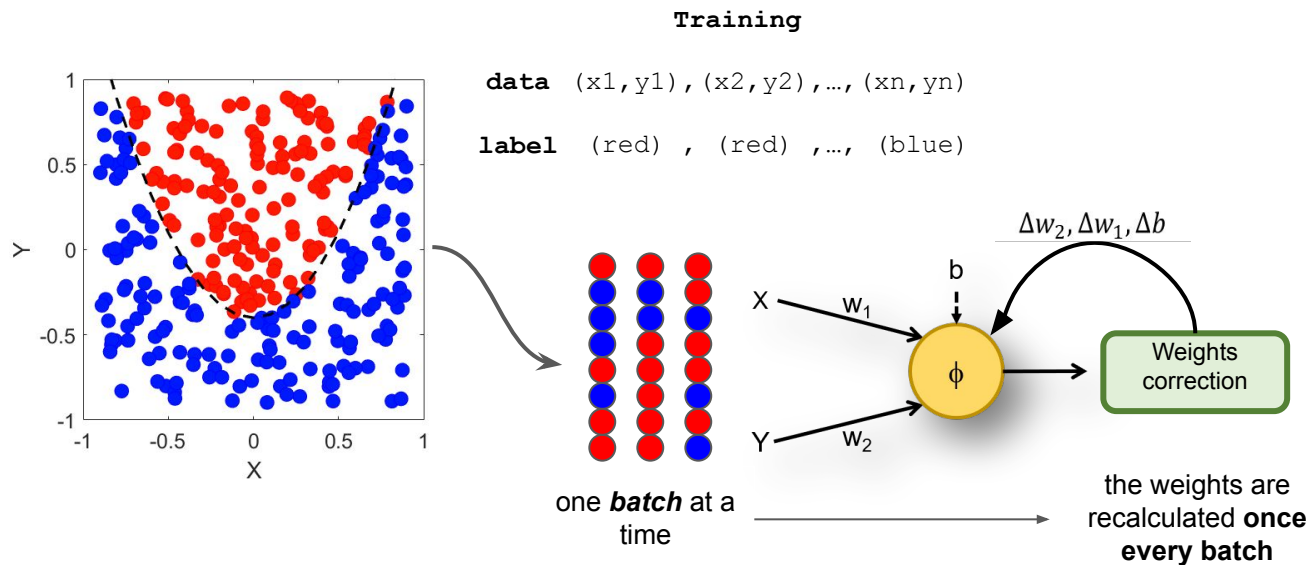


Non-saturating gradient and very fast operation. However, negative values are discarded.

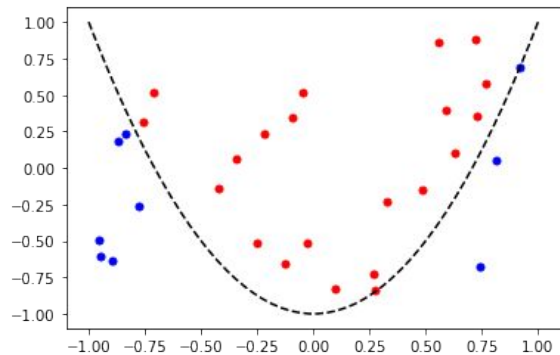
Training with mini-batch

```
history = model.fit(Training_data,  
                    Training_label,  
                    epochs = 150,  
                    batch_size = 8,  
                    validation_data = (Validation_data, Validation_label))
```

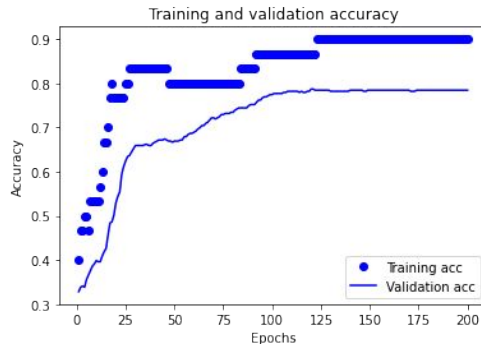
Good practice to
use a power of 2



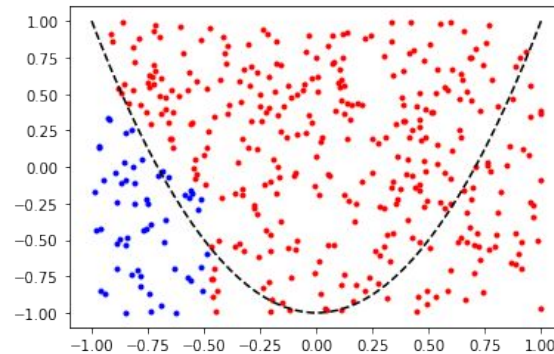
Effect of imbalanced classes



Training set



Accuracy ~ 0.8



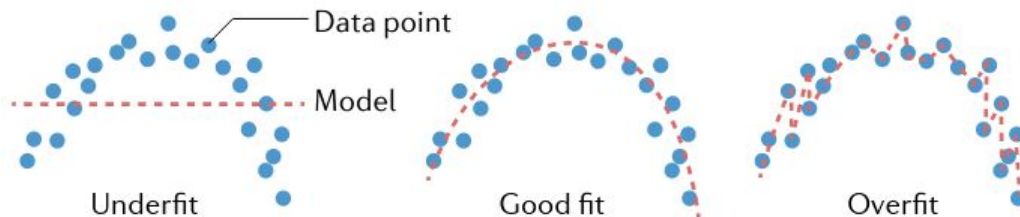
Prediction on
test set

Overfitting

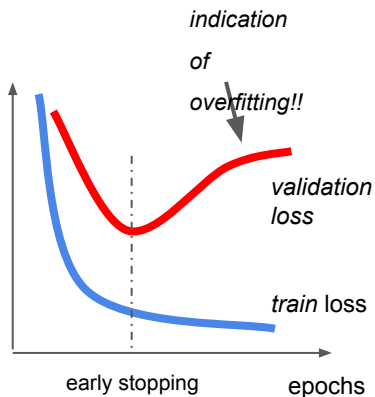
The production of an analysis that corresponds too closely or exactly to a particular set of data, and may therefore fail to fit additional data or predict future observations reliably

Possible problem when

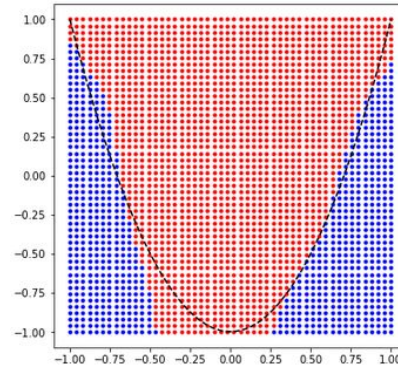
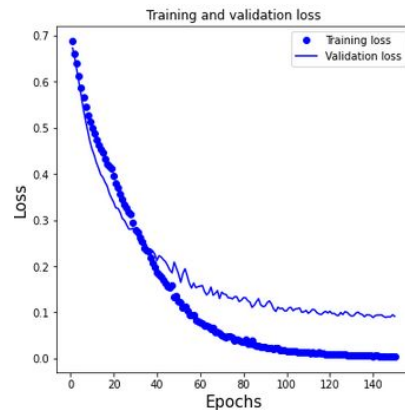
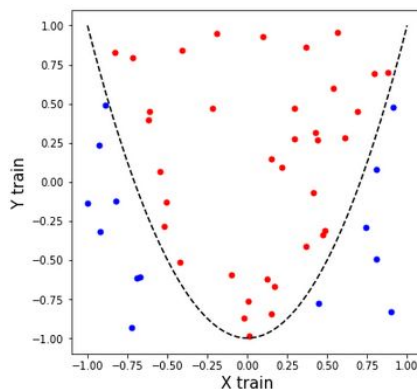
- too many layers
- too many neurons
- too **few** training data



Greener et al. - 2021 A guide to machine learning for biologists



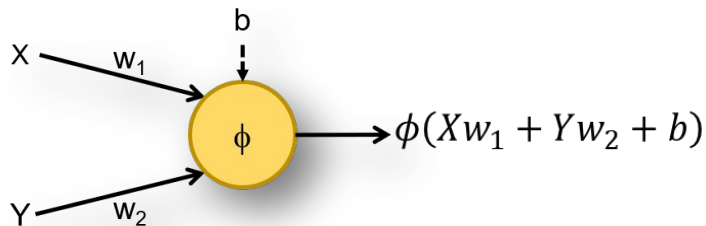
In our example:



What did we learn ?

- How a single neuron works :

- **Activation function**
 - **Input / output**
 - **Weights and bias**
- } To be defined by the user



- How the training works :

- **Loss function**
- **Optimizer**
- **Learning rate**

