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**APPLYING DEEP LEARNING AND TRANSFER  
LEARNING FOR CLASSIFYING IMAGES OF  
BACTERIAL COLONIES**

By

**Batool Tawfeeq Awwad Shdaifat**

Supervisor

**Dr. Gheith Ali Abandah, Prof.**

Assistant supervisor

**Dr. Mohammad Abdul Majeed**

**This Thesis was submitted in Partial Fulfillment of the Requirements for the  
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## **COMMITTEE DECISION**

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**LIST OF ABBREVIATION AND SYMBOLS**

| <b>Abbreviation</b> | <b>Clarification</b>         |
|---------------------|------------------------------|
| AI                  | Artificial Intelligence      |
| ANNs                | Artificial Neural Networks   |
| CNN                 | Convolutional Neural Network |
| DL                  | Deep Learning                |
| DNN                 | Deep Neural Network          |
| RNN                 | Recurrent Neural Network     |
| SNNs                | Simulated Neural Networks    |

# **APPLYING DEEP LEARNING AND TRANSFER LEARNING FOR CLASSIFYING IMAGES OF BACTERIAL COLONIES**

**By**  
**Batool Tawfeeq Shdaifat**

**Supervisors**  
**Dr. Gheith Ali Abandah, Prof.**

**-Dr. Mohammad Abdul Majeed**

## **ABSTRACT**

Bacterial classification is critical in medical science for diagnosing many diseases, treating infections, and tracking disease spread. Manually identifying and classifying bacteria takes a significant amount of time and human effort. The task of recognizing images from digital electron microscopes is now performed by computers using machine learning and computer vision techniques, thanks to technological advancements. Furthermore, the most recent generation of convolutional neural networks (CNN) have recently achieved impressive results in the field of image classification. As a result, in this thesis, I investigated a method for automating the process of bacterial identification and classification. I use the transfer learning approach to train several models. 15% of the images in the data set are selected at random and separated to test the network classification accuracy. A number of experiments were performed using different models such as vgg16, mobilenet and resnet50, and they were applied to a number of datasets, some specific to bacteria species and some specific to families of bacteria only. We have applied balanced image technology and amplification technology and produced new data sets. The experiments showed that resnet50 model yielded the best results and was able to identify and classify different bacterial species with a test accuracy of about 91% using the DS\_Org\_Aug dataset, a dataset of bacterial species, to which only the augmentation technique was applied.

## **Chapter 1: Introduction**

This chapter includes a background of my thesis, explains the research problem, and states the main points of its importance.

### **1.1 Background**

In this section, I will talk about Background Information about Bacteria, Transfer Learning, and Deep Learning.

#### **1.1.1 Background Information about Bacteria**

Bacteria are the most basic prokaryotic microorganisms, lacking a nucleus and complex organelles. Because their size is less than ten micrometres, they are studied using microscopes. Bacterial classification is critical in medical science because it serves as the foundation for understanding many bacteria-contaminated diseases. Also bacteria are difficult to classify due to their diverse shapes, which range from spheres to rods and spirals. Clinicians and microbiologists now frequently use phenotypic typing schemes to better understand bacterial morphology and staining properties (Ferdous *et al.*,2019). However, because such manual systems are time-consuming and prone to errors, they rely on human expertise. As a result, computerized techniques are required to automate the classification process of bacteria in order to reduce human effort and save valuable time. Currently, computers based on machine-learning and computer-vision technologies are performing the task of recognizing images from digital electron microscopes with high accuracy (Ferdous *et al.*,2019).

There are many types of bacteria, including pathogenic and non-pathogenic, and some of them cause a number of diseases. Table 1 shows the diseases caused by the types of bacteria that we have and their spread in Jordan.

**Table 1: Diseases caused by these bacteria and their spread in Jordan.**

| The name of bacteria                                                              | Diseases caused by these bacteria and their spread in Jordan                                                                                                                                                                                                                       |
|-----------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| Enterococcus faecalis<br>Enterococcus faecium                                     | Both types infect the intestines at most and cause diarrhea, and the two types are widespread and they are done a lot                                                                                                                                                              |
| Escherichia coli                                                                  | It affects most of the human body and is the most common cause of urinary tract infection in Jordan, and its spread is also large                                                                                                                                                  |
| Klebsiella pneumoniae                                                             | One of the most important causes of injuries and the most common in Jordan is meningitis. It also leads to severe pneumonia in young people and the elderly, and also sometimes leads to urinary tract infection. It is also one of the most prevalent in Jordan.                  |
| Staphylococcus aureus                                                             | It leads to both tonsillitis and otitis media, and it is also very widespread                                                                                                                                                                                                      |
| Staphylococcus epidermidis<br>staphylococcus simulans<br>streptococcus agalactiae | They are considered non-pathogenic bacteria, but they may be pathogens in the case of people with low immunity, such as very old people, people with cancer, and those with acute injuries and acute infections. These types are less common and their spread is more in hospitals |

### 1.1.2 Transfer Learning

Transfer learning is a machine learning technique that uses a pre-trained neural network to solve a problem similar to what the network was originally trained for. For example, you could redesign a deep learning model to classify bacteria instead of creating your own. This can save you the trouble of finding an effective neural network architecture, spending time training, building a large amount of training data, and getting good results. You can spend years purchasing fifty layers of CNN to fully distinguish the types of bacteria, or you can simply reuse one of the many pre-trained image classification models available online.

### 1.1.3 Deep Learning

Deep learning is a type of machine learning and artificial intelligence (AI) that mimics the way people acquire certain types of knowledge. Deep learning is an important element of data science that includes statistics and predictive modeling. It is extremely useful for data scientists who need to collect, analyze, and interpret large amounts of data. Deep learning makes this process faster and easier. In its simplest form, deep learning can be thought of as a way to automate predictive analytics. While traditional machine learning algorithms are linear, deep learning algorithms stack up in a hierarchy of ever increasing complexity and abstraction.

## **1.2 Research Problem**

. In recent years, machine learning has become a pivotal tool for many projects in biology and medicine (Huang *et al.*, 2018). In microbiology, the bacterial identification may be an important step for diagnosis, treatment and infectious diseases prevention. The automatic image classification approach is introduced to spot the bacterial colonies counting on the morphology. This approach is often a good alternate to the standard strategies like the mass spectrum analysis system, VITEK2 System, and biological science technique (Huang *et al.*, 2018). Generally, these methods are expensive, need longer time to process, and depend upon instruments that don't seem to be offered in most laboratories. By exploitation the deep learning technique, the bacterial colony identification can be price and time-effective, straightforward and automated.

## **1.3 Research Contributions**

One of the contributions I made to my thesis is that the dataset I worked on was not designed to be valid for the classification of bacteria, each image of this dataset contains several types of bacteria appearing inside squares of different sizes, so I extracted the squares from the images and selected some types of bacteria to get a New dataset suitable

for a machine learning project classifying bacteria and I put this dataset and its code on GitHub.

## **1.4 Research Importance**

The importance of the research lies in our ability to classify bacteria using machine learning through pictures of bacteria colonies without the need for bacteriologists or special equipment to classify bacteria in hospitals or laboratories, and this achieves less cost and more speed because in the traditional situation, bacteria are classified by dealing with bacterial cells. Through many steps from bacterial cultivation to Gram staining until microscopy. Some of these steps can take up to several hours. Alternatively, precise identification can be made by studying bacterial genetic codes. This genetic analysis includes DNA extraction and purification and polymerase chain reaction (PCR) or even sequencing in some cases, it is more expensive than the traditional method and requires some expensive tools.

## **1.5 Thesis Outline**

This thesis includes related works for my research in Chapter 2. Chapter 3 includes a thesis methodology. Chapter 4 includes the Results and Discussion. Finally, my thesis conclusion is in Chapter 5.

## **Chapter 2: Related Work**

In this section, I will talk about neural networks and the related work of bacteria classification.

### **2.1 Neural networks**

Neural networks, also known as artificial neural networks (ANNs) or simulated neural networks (SNNs), are a subset of machine learning that are at the heart of deep learning methods. Their name and structure are inspired by the human brain, replicating the way organic neurons communicate with one another.

Artificial neural networks (ANNs) are made up of node layers that include an input layer, one or more hidden layers, and an output layer. Each node, or artificial neuron, links to another and has its own weight and threshold. If the output of any individual node exceeds the defined threshold value, that node is activated and sends data to the next tier of the network. Otherwise, no data is sent to the next tier of the network.

### **2.2 Related Work of bacteria classification**

In recent years, Machine learning has become a pivotal tool for several projects in biology and medicine. And one in all the foremost spectacular and often used machine learning techniques in medical image process is deep CNN. It's discovered an incredible accuracy in classifying microscopic cell images, numeration microorganism colonies (Wahid *et al.*, 2019). And also the transfer learning overcome the main issues of deep learning like the requirement for vast datasets and long training times. Transfer learning permits us to use a pretrained model to resolve our problem. The pretrained model are retrained using our dataset that is smaller than the general dataset. Retraining on a small dataset is much quicker than training on a large one.

Bartosz *et al.* (2017) applied state of the art method for texture analysis to classify genera and species of bacteria. This method uses deep Convolutional Neural Networks to get image descriptors that are then encoded and classified with Support Vector Machine or Random Forest. To evaluate this approach and compare it against different approaches, they used DIBaS dataset (Digital Image of bacterial Species) that contains 660 pictures with thirty three different genera and species of bacteria. So as to verify if the method dedicated for texture recognition is with success applied to classify species of bacteria, they perform 2 experiments supported DIBaS dataset. The goal of the first experiment is to verify that native image descriptors and pooling encoders give the best accuracy. For classification, they used SVM or Random Forest with 3 kinds of representations: FV-SIFT, FC-CNN and FV-CNN. Within the second experiment, they examined the quantifiability of the considered method. They analyzed this attribute, as a result of there be fairly more than thirty three bacterial species and in the future they suppose to extend DIBaS dataset. To predict decrease in accuracy caused by an exaggerated variety of species. The results of the first experiment shows that either concatenating FC-M, FV-M, and FV-SIFT, or using FV-M itself provide the best accuracy (sometimes larger than 97%) for all of the classifiers. The results of the second experiment they observe that the accuracy of the considered methodology decreases nearly linearly with the amount of classes.

Lei Huang *et al.* (2018) presented a process bacterial colony classification system with 3 supervised and unsupervised neural networks. The supervised classification strategies embrace a conventional convolutional neural network and a special convolutional neural network named AlexNet designed by the supervision group for big scare visual recognition. Besides the input layer and output layer, both models are

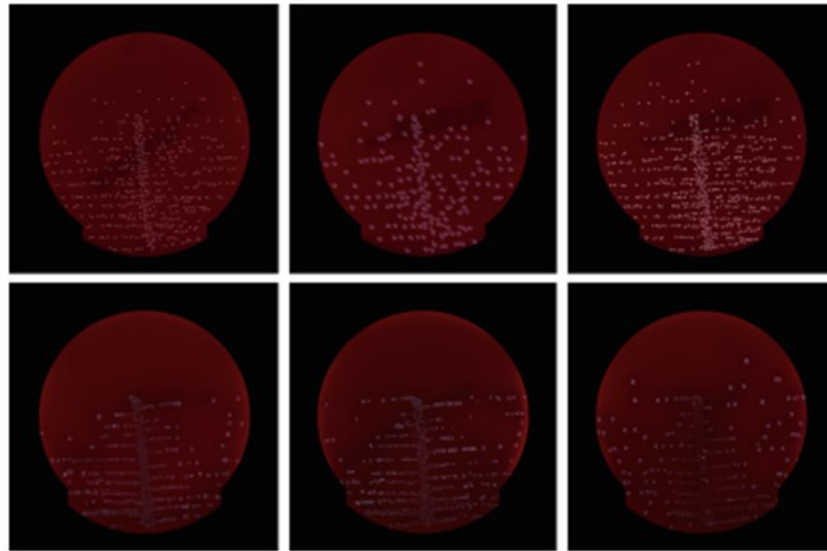
consisted of 4 varieties of layers, that is convolutional layer, ReLU layer, pooling layer and fully connected layer. Then, a form of unsupervised methodology named Autoencoder is introduced, to form comparison with the supervised methods and to verify the feasibilities of both strategies. The performance comparison was created among these 3 neural networks- conventional CNN, AlexNet and Autoencoder. In line with accuracy and precision results, CNN and AlexNet methods are comparable, each having higher precision performance than Autoencoder. The commonest 18 bacterial colony classes from Peiping University initial Hospital were used to train this framework, and different pictures out of those training dataset were utilised to check the performance of this classifier. The whole range of every bacterial class during this dataset is 4982. The classification accuracy of all eighteen bacteria classes is 73%, and therefore the accuracy and specificity of every reasonably bacteria can reach as high as 90%.

Paolo *et al.* (2018) trained a deep convolutional neural network for bacterial colony segmentation in agar plate pictures. Despite the large kind of growing media and bacterial species which will be found on Petri dishes, public informationsets don't exist accounting for such a variability. For this reason, They proposed a replacement synthetic image generator, which can be used to cheaply manufacture large datasets of fully annotated images of Petri plates. The synthetically generated data can be utilized to train a convolutional neural network for semantic image segmentation, that they proved to be capable to generalize to real images.. To evaluate the contribution of the injection of synthetic data throughout training, they proceeded with the subsequent experimental setup:

- synthetic pictures – training on synthetic data only;
- Real images – training on real data from the MicrobIA dataset only;

- synthetic and real images – training on synthetic data and fine-tune on real data from the MicrobIA dataset.

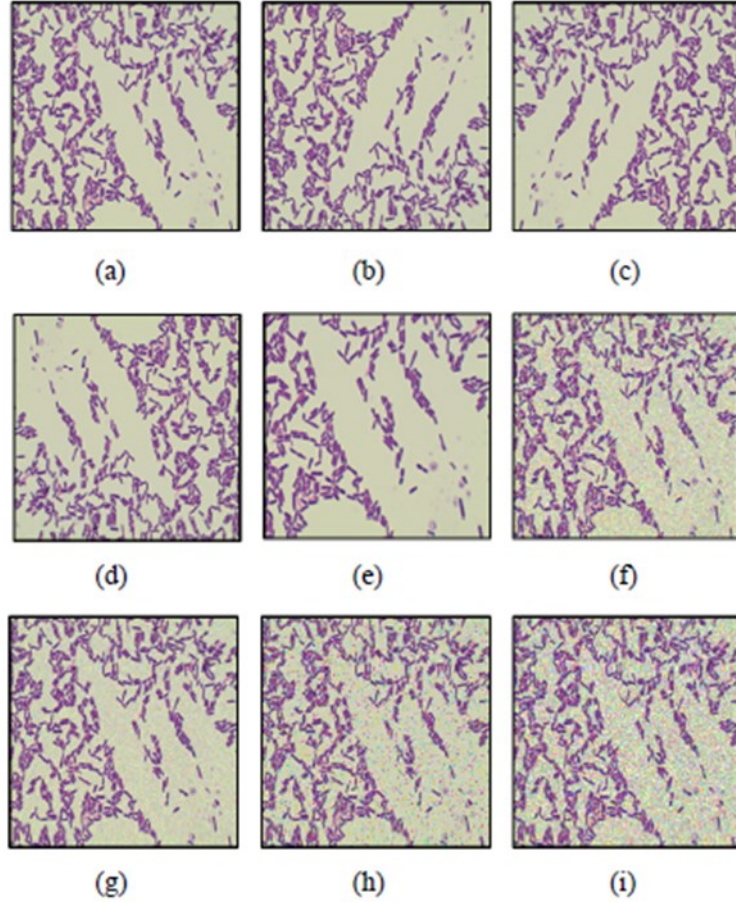
The segmentation network has been evaluated on the MicrobIA haematolysis Dataset, collection 324 pictures of blood agar plates. and a dataset of 120000 simulated blood agar plate images has been generated; some samples of synthetic images are shown in Figure 1. The segmentation results obtained using the 3 totally different experimental setups is synthetic images with accuracy 98.29%, Real images with accuracy 99.19% and synthetic and real images with accuracy 99.26%.



**Figure 1. Some examples of synthetically generated blood agar plate images (top row); Images taken from the MicrobIA Haemolysis dataset (bottom row).**

Khalifa *et al.* (2019) used augmentation techniques to come up with a bigger dataset and to decrease the overfitting problem. The first dataset employed in this research was restricted to 660 pictures for thirty three classes of a bacterial colony. Augmentation techniques raised the dataset size to 6,600 images for training and testing. The most typical methodology to beat overfitting is to extend the quantity of images used for training by applying label-preserving transformations. In addition, data augmentation schemes are applied to the training set to create the resulting model additional invariant to reflection,

zooming and little noise in values of a pixel. In order to use augmentation, every image within the training data is transformed for Reflection X, Reflection Y, Reflection XY, and Zoom and for four kinds of noise: includes Gaussian, Poisson, salt and pepper and speckle noise. this raises the dataset number of pictures from 660 images to 6,600 images, doubled ten times. this can result in a big improvement in neural network training phase. Also, will build the proposed design proof against memorise the data and be additional sturdy and answerable for the testing and verification phase. Figure 2 shows the output of applying previous data augmentation techniques on a sample image type dataset. The proposed design achieved a median accuracy of 98.22%.



**Figure 2. Augmentation techniques applied to image sample from DIBaS, (a) original image (b) reflection X axis, (c) reflection Y axis, (d) reflection X, Y axis, (e) zooming (f), Gaussian noise (g) Poisson noise, (h) salt and pepper noise, (i) speckle noise.**

Ferdous *et al.* (2019) presented an automatic system to acknowledge at the same time classify bacteria from microscopic image using deep convolutional neural network (CNN) namely, 'Xception architecture' (Ferdous *et al.*, 2019). Here, they have chosen 7 types of bacteria for recognition which could proof lethal for human and prepare a dataset of 1150 bacteria pictures where every selection contains at least a hundred and sixty images. They trained the projected system on 920 bacteria images of seven varieties from train dataset. Then, finally the performance has been evaluated on 230 bacteria images of 7 varieties from check dataset that shows promising performance with around 97.5% prediction accuracy in bacteria image classification. They used network exploiting transfer learning method that permits them to partially use the pre-trained network weight that decreases process complexness and total coaching parameters. Therefore, they used pretrained Xception design as feature extractor. So as to use the load of pretrained Xception architecture, they have removed last 5 layers of the pretrained Xception architecture and add one global average pooling, 2 fully connected, one softmax and a classification-output layer at the last transferred layer (block14\_acpconv2\_act) for classifying seven styles of bacteria. Subsequently they have frozen the lower thirteen layers (from input\_1 to conv2D\_1) to stop update their weights throughout training method as lower layers extract solely low-level features corresponding to corners, edges, blob and so on that is most typical features all told pictures. Then they retrain the active layers of the changed Xception model and update their pre-trained weights in step with the input images for extracting a lot of invariant, high-level and relevant features. Finally, all the extracted features are planar and fed into fully-connected layer to train the remainder of the modified Xception network for activity classification task.



## Chapter 3: Methodology

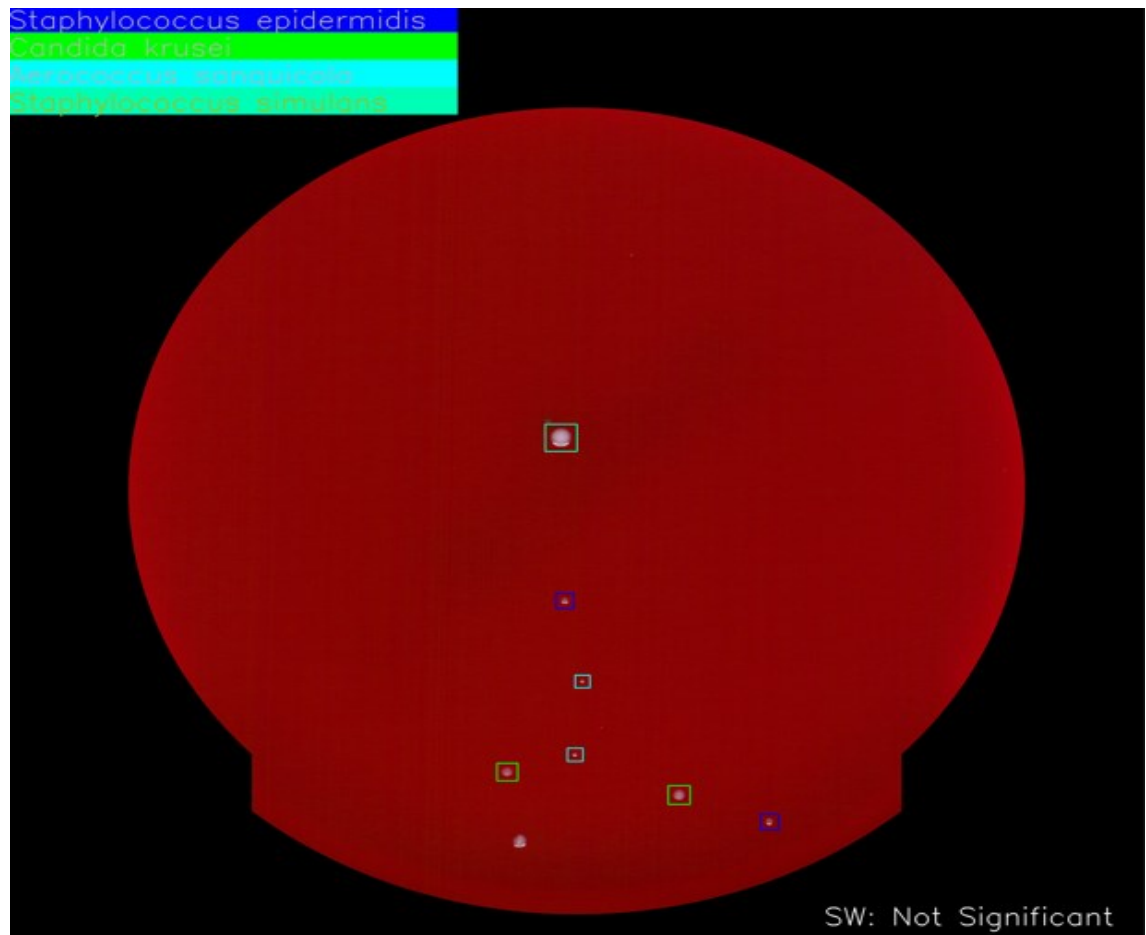
In this chapter, we will initially talk about the data set that I worked on related to bacteria species and their families, and then we will talk about the experiments we did use a number of models such as vgg16, mobilint and resnet50.

### 3.1 Dataset

In this section, I will talk about the dataset from several aspects. At first, about the preparation of the dataset, dataset augmentation, and the experiments that we have done.

#### 3.1.1 Data Preparation

Initially, we studied several types of the dataset for bacteria, and finally, MicrobIA Haemolysis Dataset was selected, and this data set was specialized for examination of the presence of hemolysis. Hemolysis consists of three types:  $\alpha$ -hemolysis,  $\beta$ -hemolysis, and  $\gamma$ -hemolysis, and since each type contains several types of bacteria that cause it. The sample agar plates are filled with samples of several types of bacteria, so we know that the dataset I worked on was not designed to be valid for bacteria classification, each image of this dataset contains several types of bacteria appearing inside different sized squares, so I extracted the squares from the images and selected some bacteria types to get a new dataset suitable for a machine learning project which classifies bacteria. I built the new dataset and publish it in two versions, DS\_Org and DS\_bal. I TFigure 3 shows one of the agar plates from the dataset with the regions with bacteria surrounded by a colored rectangle.



**Figure 3. In this agar plate have four type of bacteria: 1-staphylococcus epidermis  
2: staphylococcus simulans 3: candida krusei 4: Aerococcus sanguicola.**

After that, the types of bacteria present in the samples were counted, and the number of repetitions of each of them was counted, and the results were shown in the following Table 2.

**Table 2: The 33 types of bacteria and the number of images for each type.**

| <b>The Name of Bacteria</b>            | <b>The Number Of Images</b> |
|----------------------------------------|-----------------------------|
| Aerococcus sanguicola                  | 697                         |
| Aerococcus urinae                      | 243                         |
| Candida albican                        | 131                         |
| Candida glabrata                       | 0                           |
| Candida krusei                         | 55                          |
| Candida parapsilosis                   | 6                           |
| Candida tropicalis                     | 61                          |
| Citrobacter freundii                   | 8                           |
| Citrobacter koseri                     | 2                           |
| Enterobacter aerogenes                 | 0                           |
| Enterobacter cloacae                   | 0                           |
| Enterococcus faecalis                  | 1132                        |
| Enterococcus faecium                   | 790                         |
| Escherichia coli                       | 306                         |
| Klebsiella oxytoca                     | 0                           |
| Klebsiella pneumoniae                  | 127                         |
| Lactobacillus species                  | 218                         |
| Micro-colonies                         | 0                           |
| Morganella morganii                    | 24                          |
| Proteus mirabilis                      | 11                          |
| Providencia stuartii                   | 0                           |
| Pseudomonas aeruginosa                 | 147                         |
| Serratia marcescens                    | 18                          |
| Staphylococcus aureus                  | 141                         |
| Staphylococcus epidermidis             | 602                         |
| Staphylococcus lugdunensis             | 22                          |
| Staphylococcus saprophyticus           | 15                          |
| Staphylococcus simulans                | 184                         |
| Streptococcus agalactiae               | 358                         |
| Streptococcus dysgalactiae (Group C/G) | 13                          |
| Streptococcus mitis                    | 138                         |
| Streptococcus oralis                   | 39                          |
| Streptococcus pyogenes                 | 48                          |
| <b>Total images</b>                    | <b>5536</b>                 |

Then all bacteria containing less than 140 images were ignored. The number of samples of agar plates available is 279, and the number and types of bacteria in each plate differ from the other. Table 3 shows the types of bacteria and the number of images that were used in this thesis.

**Table 3: Types of bacteria and the number of images that were collected for them.**

| The Name Of Bacteria       | Number Of Images |
|----------------------------|------------------|
| Enterococcus faecalis      | 1132             |
| Enterococcus faecium       | 790              |
| Escherichia coli           | 306              |
| Klebsiella pneumoniae      | 127              |
| Staphylococcus aureus      | 141              |
| Staphylococcus epidermidis | 602              |
| Staphylococcus simulans    | 184              |
| Streptococcus agalactiae   | 358              |
| <b>Total images</b>        | <b>3640</b>      |

Then I decided to suggest four types of datasets as follows:

- DS\_Org
- DS\_Bal
- DS\_OrgFam
- DS\_BalFam.

DS\_Org and DS\_Bal are specific to all of our eight bacterial species, but DS\_OrgFam and DS\_BalFam are more general because they are only specific to the five bacteria families. The details of each dataset is as follows:

**DS\_Org (Dataset Original):** It consists of only clear images from the MicrobIA Haemolysis dataset. These images are used in our model training and evaluation as shown in Table 4.

**Table 4: The types of bacteria and the number of images that were collected for DS\_Org.**

| The Name Of Bacteria       | Train Images | Validation Images | Test Images | Total Images |
|----------------------------|--------------|-------------------|-------------|--------------|
| Enterococcus faecalis      | 178          | 31                | 36          | 245          |
| Enterococcus faecium       | 116          | 20                | 24          | 160          |
| Escherichia coli           | 102          | 18                | 21          | 141          |
| Klebsiella pneumoniae      | 82           | 14                | 16          | 112          |
| Staphylococcus aureus      | 95           | 16                | 19          | 130          |
| Staphylococcus epidermidis | 125          | 21                | 25          | 171          |
| Staphylococcus simulans    | 73           | 12                | 15          | 100          |
| Streptococcus agalactiae   | 87           | 15                | 18          | 120          |
| <b>Total Images</b>        | <b>858</b>   | <b>147</b>        | <b>174</b>  | <b>1179</b>  |

**DS\_Bal (Dataset Balanced):** Consists of clear images and used equal number of images for all types of bacteria and shown in Table 5.

**Table 5: The types of bacteria and the number of images that were collected for DS\_Bal.**

| The Name Of Bacteria       | Train Images | Validation Images | Test Images | Total Images |
|----------------------------|--------------|-------------------|-------------|--------------|
| Enterococcus faecalis      | 73           | 12                | 15          | 100          |
| Enterococcus faecium       | 73           | 12                | 15          | 100          |
| Escherichia coli           | 73           | 12                | 15          | 100          |
| Klebsiella pneumoniae      | 73           | 12                | 15          | 100          |
| Staphylococcus aureus      | 73           | 12                | 15          | 100          |
| Staphylococcus epidermidis | 73           | 12                | 15          | 100          |
| Staphylococcus simulans    | 73           | 12                | 15          | 100          |
| Streptococcus agalactiae   | 73           | 12                | 15          | 100          |
| <b>Total Images</b>        | <b>584</b>   | <b>96</b>         | <b>120</b>  | <b>800</b>   |

**DS\_OrgFam (Dataset Original for Families of bacteria):** In this dataset the bacteria classes are grouped into their biological families as shown in Table 6.

**Table 6: The types of bacteria and the number of images that were collected for DS\_OrgFam.**

| The Name Of Bacteria     | Train Images | Validation Images | Test Images | Total Images |
|--------------------------|--------------|-------------------|-------------|--------------|
| Enterococcus             | 294          | 51                | 60          | 405          |
| Escherichia coli         | 102          | 18                | 21          | 141          |
| Klebsiella pneumoniae    | 82           | 14                | 16          | 112          |
| Staphylococcus           | 293          | 49                | 59          | 401          |
| Streptococcus agalactiae | 87           | 15                | 18          | 120          |
| <b>Total Images</b>      | <b>858</b>   | <b>147</b>        | <b>174</b>  | <b>1179</b>  |

**DS\_BalFam (Dataset Balanced for Families of bacteria):** Similar to **DS\_OrgFam** dataset but with equal number of images in each family as shown in Table 7.

**Table 7: The types of bacteria and the number of images that were collected for DS\_BalFam.**

| The Name Of Bacteria     | Train Images | Validation Images | Test Images | Total Images |
|--------------------------|--------------|-------------------|-------------|--------------|
| Enterococcus             | 73           | 12                | 15          | 100          |
| Escherichia coli         | 73           | 12                | 15          | 100          |
| Klebsiella pneumoniae    | 73           | 12                | 15          | 100          |
| Staphylococcus           | 73           | 12                | 15          | 100          |
| Streptococcus agalactiae | 73           | 12                | 15          | 100          |
| <b>Total Images</b>      | <b>365</b>   | <b>60</b>         | <b>75</b>   | <b>500</b>   |

### 3.1.2 Image Augmentation

Image augmentation was used to increase the diversity and number of image information in datasets by resampling the image into a completely different image. There are several ways such as rotating, zooming in/out and flipping. During this message, image rotation 90, 180 and 270° was used and applied to DS\_Org, DS\_Bal, DS\_OrgFam and DS\_BalFam. And now we have four new datasets, which are as follows:

- DS\_Org\_Aug
- DS\_Bal\_Aug
- DS\_OrgFam\_Aug
- DS\_BalFam\_Aug

The tables from Table 8 to Table 11 show the data sets after data augmentation.

**Table 8: The DS\_Org\_Aug and the number of images that were collected for them.**

| The Name Of Bacteria       | Train Images | Validation Images | Test Images | Total Images |
|----------------------------|--------------|-------------------|-------------|--------------|
| Enterococcus faecalis      | 712          | 124               | 144         | 980          |
| Enterococcus faecium       | 464          | 80                | 96          | 640          |
| Escherichia coli           | 408          | 72                | 84          | 564          |
| Klebsiella pneumoniae      | 328          | 56                | 64          | 448          |
| Staphylococcus aureus      | 380          | 64                | 76          | 520          |
| Staphylococcus epidermidis | 500          | 84                | 100         | 684          |
| Staphylococcus simulans    | 292          | 48                | 60          | 400          |
| Streptococcus agalactiae   | 348          | 60                | 72          | 480          |
| <b>Total Images</b>        | <b>3432</b>  | <b>588</b>        | <b>696</b>  | <b>4716</b>  |

**Table 9: The DS\_Bal\_Aug and the number of images that were collected for them.**

| The Name Of Bacteria       | Train Images | Validation Images | Test Images | Total Images |
|----------------------------|--------------|-------------------|-------------|--------------|
| Enterococcus faecalis      | 292          | 48                | 60          | <b>400</b>   |
| Enterococcus faecium       | 292          | 48                | 60          | <b>400</b>   |
| Escherichia coli           | 292          | 48                | 60          | <b>400</b>   |
| Klebsiella pneumoniae      | 292          | 48                | 60          | <b>400</b>   |
| Staphylococcus aureus      | 292          | 48                | 60          | <b>400</b>   |
| Staphylococcus epidermidis | 292          | 48                | 60          | <b>400</b>   |
| Staphylococcus simulans    | 292          | 48                | 60          | <b>400</b>   |
| Streptococcus agalactiae   | 292          | 48                | 60          | <b>400</b>   |
| <b>Total Images</b>        | <b>2336</b>  | <b>384</b>        | <b>480</b>  | <b>3200</b>  |

**Table 10: The DS\_BalFam\_Aug and the number of images that were collected for them.**

| The Name Of Bacteria     | Train Images | Validation Images | Test Images | Total Images |
|--------------------------|--------------|-------------------|-------------|--------------|
| Enterococcus             | 292          | 48                | 60          | <b>400</b>   |
| Escherichia coli         | 292          | 48                | 60          | <b>400</b>   |
| Klebsiella pneumoniae    | 292          | 48                | 60          | <b>400</b>   |
| Staphylococcus           | 292          | 48                | 60          | <b>400</b>   |
| Streptococcus agalactiae | 292          | 48                | 60          | <b>400</b>   |
| <b>Total Images</b>      | <b>1460</b>  | <b>240</b>        | <b>300</b>  | <b>2000</b>  |

**Table 11: The DS\_OrgFam\_Aug and the number of images that were collected for them.**

| The Name Of Bacteria     | Train Images | Validation Images | Test Images | Total Images |
|--------------------------|--------------|-------------------|-------------|--------------|
| Enterococcus             | 1176         | 204               | 240         | <b>1620</b>  |
| Escherichia coli         | 408          | 72                | 84          | <b>564</b>   |
| Klebsiella pneumoniae    | 328          | 56                | 64          | <b>448</b>   |
| Staphylococcus           | 1172         | 196               | 236         | <b>1604</b>  |
| Streptococcus agalactiae | 348          | 60                | 72          | <b>480</b>   |
| <b>Total Images</b>      | <b>3432</b>  | <b>588</b>        | <b>696</b>  | <b>4716</b>  |

### 3.2 Experiments

I used three types of models which are Resnet50, Vgg16, and Mobile net.

The experiments were conducted in two phases, in the first phase, I trained the models only on the output layer, while in the second phase I did a deep study of all the models, By utilizing previous research, and by training and freezing layers to obtain the best results for each model.

Experiments were performed on 8 types of datasets, four of which contain 8 types of bacteria which are DS\_Org, DS\_Bal, DS\_Org\_Aug, and DS\_Bal\_Aug and the other four contain only 5 families of bacteria which are DS\_OrgFam, DS\_BalFam, DS\_OrgFam\_Aug, and DS\_BalFam\_Aug.

In addition, I made a simple model with multiple layers of CNN and then tested datasets on it, and I use 400 epochs with early stopping patience = 5 for all experiments.

## Chapter 4: Results and Discussion

In this chapter, in the first two parts of it, we will talk about the results that I obtained after experiments on eight classes of bacteria and on five classes of bacteria, and in the third part we will discuss these results.

### 4.1 Bacteria Species Datasets

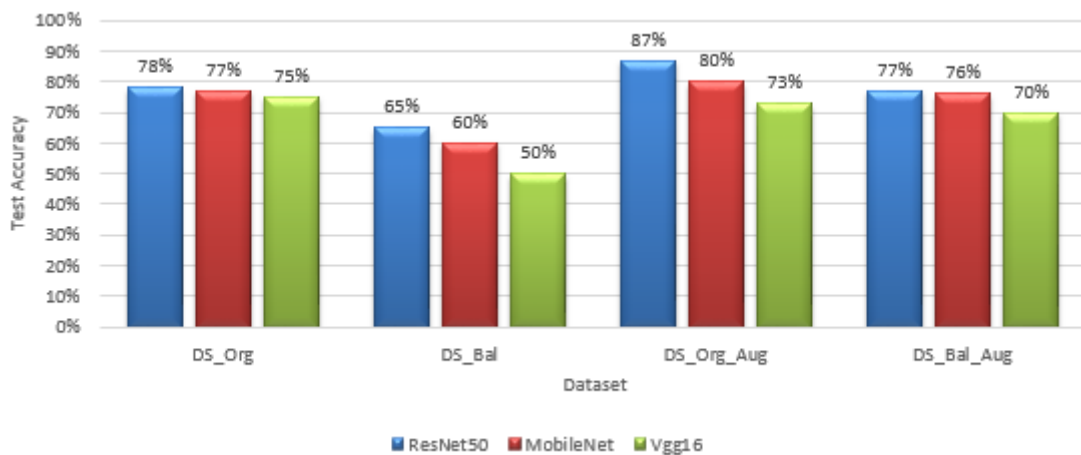
This section describes the results that we obtained in the case of eight classes of bacteria and is divided into two parts: Investigating Three Alternative models and Selecting the Number of Trained Layers.

#### 4.1.1 Investigating Three Alternative models

In this section, we used a number of ready-made models, namely ResNet50, Vgg16, and MobileNet. In the first, we trained this model on the output layer only, and Table 12 shows the results.

**Table 12: The Test accuracy results on bacteria species datasets when trained this model on the output layer only.**

| Models Name | DS_Org | DS_Bal | DS_Org_Aug | DS_Bal_Aug |
|-------------|--------|--------|------------|------------|
| ResNet50    | 80%    | 65%    | 87%        | 77%        |
| Vgg16       | 75%    | 50%    | 73%        | 70%        |
| MobileNet   | 78%    | 60%    | 80%        | 76%        |

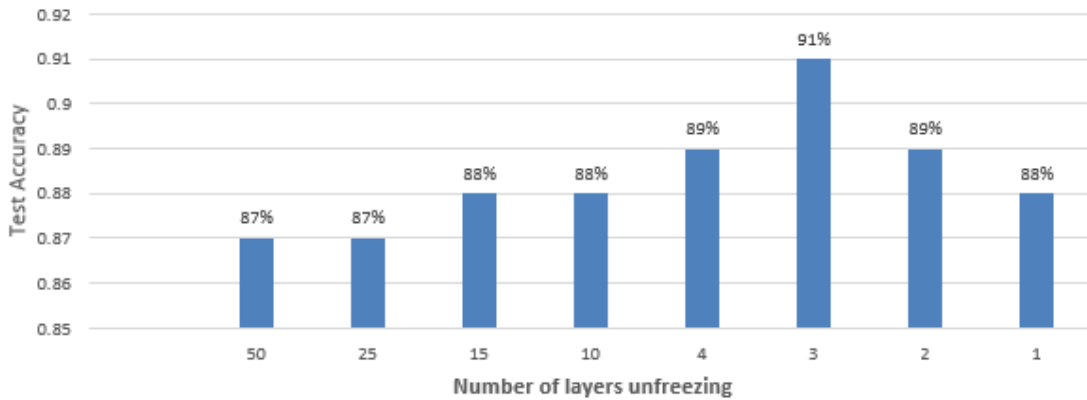


**Figure 4. Test accuracy for ResNet50, Vgg16, and MobileNet models for bacteria species datasets when trained this model on the output layer only.**

Figure 4 shows us the accuracy of the test, and it is clear that the ResNet50 model achieved the best results compared to the other models.

#### 4.1.2 Selecting the Number of Trained Layers

We conducted a deeper study and tried to train different layers of models. For example, in the Vgg16 model, it was found that the freeze layers from 16-20 achieved the best results as mentioned (Hasib *et al.*, 2018), and in the MobileNet model, training three layers of the convolutional neural network achieved the best results as mentioned (Suharjito *et al.*, 2021), For the Resnet50 model, I decided to try training a different number of layers, and training the last 3 layers had the best result. Figure 5 shows the results.

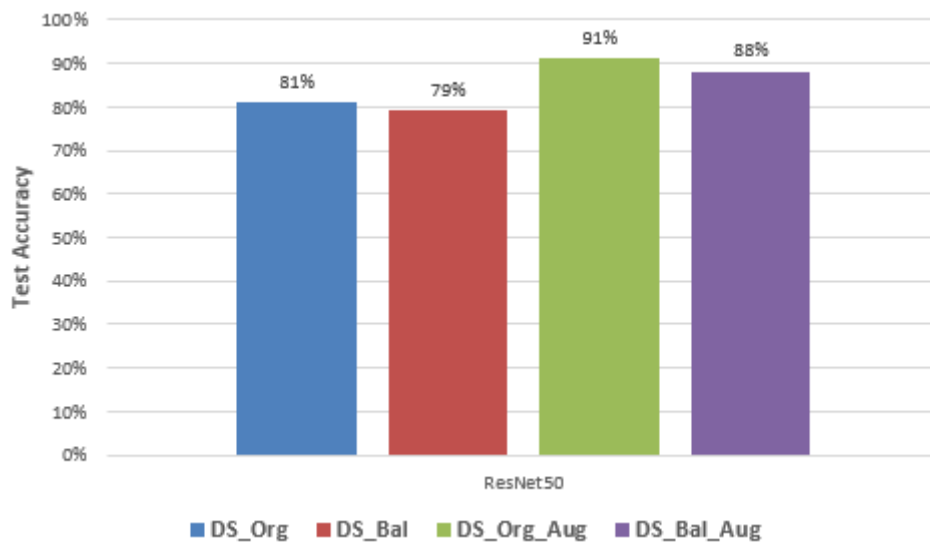


**Figure 5. Test accuracy ResNet50 model in different layers training cases, using DS\_Org\_Aug dataset.**

In addition to the models that were used, I built a simple model consisting of a few CNN layers, and in Table 13 the final results of the second phase are shown in addition to the results of the simple model, and as it is clear the Resnet50 model achieves the best results.

**Table 13: The results of the final test accuracy of the selecting the number of trained layers in addition to the test accuracy of the simple model.**

| Models Name  | DS_Org | DS_Bal | DS_Org_Aug | DS_Bal_Aug |
|--------------|--------|--------|------------|------------|
| ResNet50     | 81%    | 79%    | 91%        | 88%        |
| Vgg16        | 78%    | 77%    | 83%        | 81%        |
| MobileNet    | 78%    | 76%    | 83%        | 80%        |
| simple model | 76%    | 77%    | 86%        | 86%        |



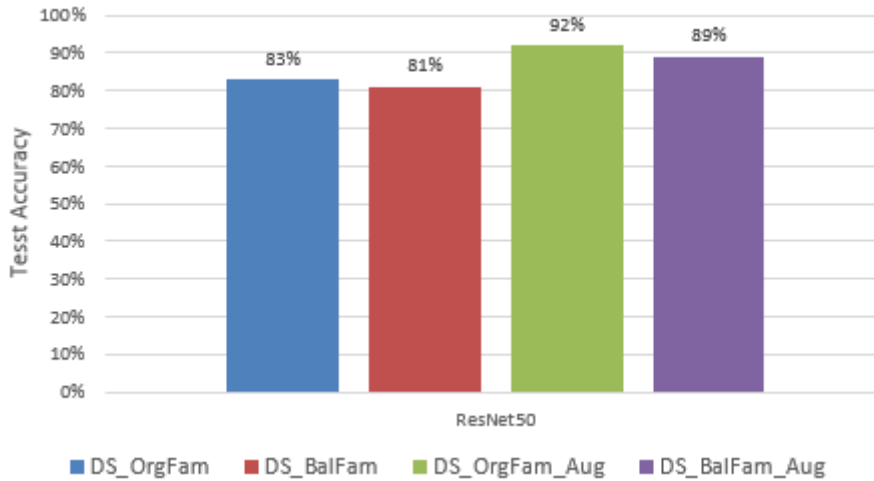
**Figure 6. Test accuracy for ResNet50 model for selecting the number of trained layers.**

## 4.2 Bacteria Families Datasets

In this section describes the results of the final experiments obtained for five classes of bacteria families, I decided to adopt the Resnet50 model only and conduct the remaining experiments on it. In this case, we trained the Resnet50 model on the last three layers that proved its advantage, and it was clear that the best results were using the DS\_OrgFam\_Aug dataset with results up to 92% and Table 14 shows these results:

**Table 14: The Test accuracy results in bacteria families datasets.**

| Models Name | DS_OrgFam | DS_BalFam | DS_OrgFam_Aug | DS_BalFam_Aug |
|-------------|-----------|-----------|---------------|---------------|
| ResNet50    | 83%       | 81%       | 92%           | 89%           |



**Figure 7. Test accuracy for ResNet50 model for bacteria families datasets.**

Figure 7 shows that the Resnet50 model achieves the best results using the dataset DS\_OrgFam\_Aug

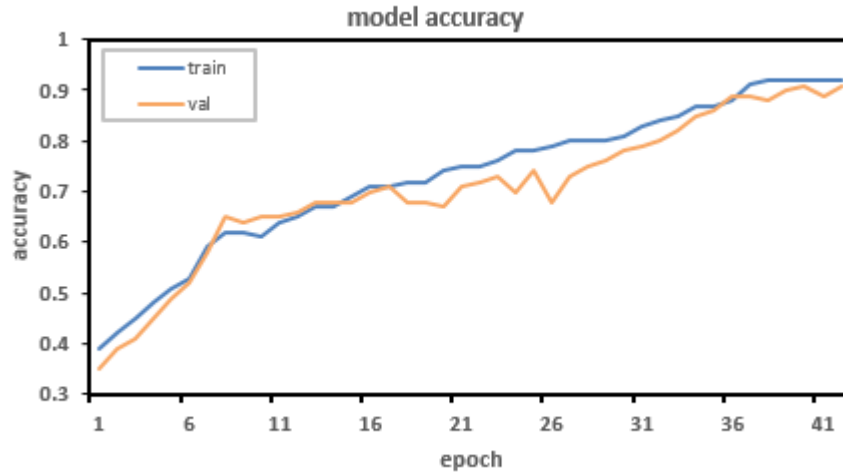
### 4.3 Discussion

In this section, we will discuss the best case that has been tried and analyzed more and more.

The best case that has been tried is when I used DS\_Org\_Aug Dataset, which is a dataset of eight types of bacteria and using all the images without using the image balanced technique and with the use of the Augmentation technique and the Resnet50 model was applied to it with training the last three layers of it.

Accordingly, it was found that using the data set that contains separate types of bacteria is better than using the data set that contains only the families of bacteria, although the bacteria families obtained an accuracy of 92%, which is higher than the accuracy obtained by the types of isolated bacteria with a value of 1% But this increase is not good because it is very small, and we lost with it information about the separate types of bacteria.

Figure 8 shows the train and validation curve as a function of epoch for the best case achieved.



**Figure 8. Train and validation curve as a function of epoch for the best case achieved.**

And in Table 15 shows the values of Precision, recall, and F1 score for the best case obtained.

**Table 15: Value of Precision, recall, and F1 score for the best case obtained.**

| The Name Of Bacteria       | Precision | recall | F1_score |
|----------------------------|-----------|--------|----------|
| Enterococcus faecalis      | 1         | 0.9    | 0.95     |
| Enterococcus faecium       | 0.83      | 0.95   | 0.88     |
| Escherichia coli           | 1         | 0.94   | 0.97     |
| Klebsiella pneumoniae      | 0.97      | 0.98   | 0.98     |
| Staphylococcus aureus      | 0.92      | 0.95   | 0.94     |
| Staphylococcus epidermidis | 0.92      | 0.93   | 0.93     |
| Staphylococcus simulans    | 0.89      | 0.57   | 0.69     |
| Streptococcus agalactiae   | 0.75      | 1      | 0.86     |

## Chapter 5: Conclusion

Manually identifying bacteria is possible in a variety of ways. Most of the time, these methods necessitate the use of live bacteria samples and highly trained personnel who perform various writing schemes and use electron microscopes to observe the structural patterns and colony formations of bacteria. Thus, manual methods necessitate impressive skills and valuable time, both of which can be saved by computerizing the bacteria classification process through the use of machine learning and computer vision techniques. In this thesis, several transfer learning models, namely Resnet50, Vgg16, and Mobilenet, were used to classify bacterial forms. I've used a number of datasets, including those for the types of bacteria I have, and others more general just for bacteria families, and then divided them into balanced and unbalanced datasets. She also used the Augmentation technique and applied it to the datasets, after that used 15% of each dataset to test set and 12% of the remaining for the validation set and the rest was for the training set. After performing multiple steps of preprocessing the image data, and using two phases for models, We finally found that Through the experiments, it was found that using the augmentation technique is better than not using it, as the increase in the number of images in the dataset enabled us to get better accuracy, while the use of the balanced dataset technique did not give good results, Conversely, not using it and experimenting with the complete dataset produced better results. In addition, experiments on a data set containing bacteria families achieved better results than the data set containing bacteria types separately, but the improvement was very small, which means that it is not worth losing some information about the separate types of bacteria.

Accordingly, we can say that the best case was using the ResNet50 model by training the last three layers of it and also using the dataset to which the augmentation technique

was applied and not to which the balanced dataset technique was applied, this case achieved an accuracy of 91% using the DS\_Org\_Aug dataset.

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## تطبيق التعلم العميق ونقل التعلم لتصنيف صور المستعمرات البكتيرية

إعداد

بتول توفيق عواد شديفات

المشرفين

الأستاذ الدكتور غيث علي عبنده

الدكتور محمد عبدالمجيد

### ملخص

يعتبر التصنيف البكتيري أمرًا بالغ الأهمية في العلوم الطبية لتشخيص العديد من الأمراض وعلاج العدوى وتتبع انتشار المرض. من ناحية أخرى ، يستغرق تحديد البكتيريا وتصنيفها يدويًا قدرًا كبيرًا من الوقت والجهد البشري. يتم الآن تنفيذ مهمة التعرف على الصور من المجاهر الإلكترونية الرقمية بواسطة أجهزة الكمبيوتر باستخدام تقنيات التعلم الآلي ورؤية الكمبيوتر ، وذلك بفضل التقدم التكنولوجي. علاوة على ذلك ، حقق أحدث جيل من الشبكات العصبية التلافيفية (CNN) مؤخرًا نتائج رائعة في مجال تصنيف الصور. نتيجة لذلك ، في هذه الأطروحة ، بحثت في طريقة لأتمتة عملية التعرف على البكتيريا وتصنيفها. أستخدم نهج التعلم في النقل لتدريب عدة نماذج ، بما في ذلك "نموذج Resnet50". يتم اختيار 15٪ من الصور في مجموعة البيانات عشوائيًا وفصلها لاختبار دقة تصنيف الشبكة. تم إجراء عدد من التجارب باستخدام نماذج مختلفة مثل vgg16 و mobilenet و resnet50 ، وتم تطبيقها على عدد من مجموعات البيانات ، بعضها خاص بأنواع البكتيريا وبعضها خاص بعائلات البكتيريا فقط. لقد طبقنا تقنية الصورة المتوازنة وتكنولوجيا التضخيم وأنتجنا مجموعات بيانات جديدة. ثم بينت لنا التجارب أن نموذج resnet50 حقق أفضل النتائج وكان قادرًا على تحديد وتصنيف الأنواع البكتيرية المختلفة بدقة اختبار تبلغ حوالي 91٪ باستخدام مجموعة بيانات DS\_Org\_Aug ، وهي مجموعة بيانات للأنواع البكتيرية ، والتي تم تطبيق تقنية الزيادة عليها فقط. .