

# A Systematic Literature Review of Deep Learning-Based Detection and Classification Methods for Bacterial Colonies

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**Abstract**—Deep learning is an area of machine learning that has substantial potential in various fields of study such as image processing and computer vision. A large number of studies are published annually on deep learning techniques. The focus of this paper is on bacteria detection, identification, and classification. This paper presents a systematic literature review that synthesizes the evidence related to bacteria colony identification and detection published in the year 2021. The aim is to aggregate, analyse, and summarize the evidence related to deep learning detection, identification, and classification of bacteria and bacteria colonies. The significance is that the review will help experts and technicians to understand how deep learning techniques can apply in this regard and potentially further support more accurate detection of bacteria types. A total of 38 studies are analysed. The majority of the published studies focus on supervised-learning-based convolutional neural networks. Furthermore, a large number of studies make use of laboratory-prepared datasets as compared to open-source and industrial datasets. The results also indicate a lack of tools, which is a barrier in adapting academic research in industrial settings.

**Keywords**—AI; bacterial-colonies; classification; deep learning; detection; literature review

## I. INTRODUCTION

Bacteria are unicellular microorganisms that are present on everything around us. A single drop of sea water contains at least a million cells of bacteria [1]. Knowledge of the bacterial genre is extremely important in microbiology. Accurate and quick recognition of the bacterial genre is crucial in various fields including clinical diagnosis, medicine, water and food industry, etc. [2] [3] [4].

In the traditional identification process, preparing samples requires not only substantial time but also specific equipment and costly chemicals. This step must take place before the identification process can even start. The resultant samples are observed by experts to identify features in traditional laboratory setups. Samples are also compared with standard reference images for accurate identification, which is a tedious and time-consuming task [5].

The most important feature for recognition of bacteria is shape. Bacteria can be classified into numerous categories based on its shape. Some of the prominent shapes include spiral, longitudinal, and cylindrical. Since various types of bacteria share very similar shapes, however, considering shape alone makes differentiation difficult. Therefore, other characteristics such as presence and shape of colonies and morphology are

exploited to correctly recognize bacteria. In short, identifying type of bacteria is arduous even for experienced microbiologists [5].

Image processing and computer vision techniques have revolutionized the approach to bacterial identification with the ability to process and classify large amounts of data. Yet identifying patterns to derive conclusions from complex, dynamic, and heterogeneous data created by computerized techniques is another challenge for scientists. Evolution of machine learning techniques has enabled scientists to classify, predict, and identify patterns from large amounts of data [6]. Deep learning is a subset of machine learning that has shown tremendous success in various fields including identification and classification of bacteria.

Systematic literature reviews (SLRs) aggregate, classify, and analyse state-of-the-art information from existing literature. SLRs are a type of secondary study that collects and summarizes the literature published in a particular area. The studies under review are referred to as primary studies [7].

This paper presents an SLR on deep-learning-based classification and identification of bacterial colonies. This work aggregates, analyses, and summarizes 38 studies related to deep-learning-based bacterial classification. The aim of this study is to benefit new researchers by providing organized insights from the literature. Furthermore, this SLR is also beneficial for practitioners since it highlights the latest tools, techniques, and frameworks in this area. Specifically, this paper provides the following contributions:

- Identification of the deep-learning-based bacterial classification techniques presented in the literature. This paper presents a taxonomy and classifies existing literature according to types of deep learning approach, types of learning, and tools used to perform bacterial colony classification.
- A descriptive analysis of quantitative data and a thematic analysis of qualitative data to provide insights into deep learning approaches and datasets. This paper also presents a comparative analysis to find the similarities and differences in performance evaluation metrics and tools available in existing studies.
- Insights for practitioners into the latest advancements in tools, techniques, and frameworks in the deep-learning-based classification of bacterial colonies.

- Reporting of benchmark datasets and performance evaluation metrics used to measure the performance of deep learning approaches for bacterial colony classification.

The rest of the paper is organized as follows: Section II discusses the background of various deep learning approaches in general. Section III presents the research methodology applied in this study. Section IV presents the results of this study. Finally, Section V presents a discussion followed by the conclusion.

## II. BACKGROUND

### A. A State-of-the-Art Survey on Deep Learning Theory and Architectures

Deep learning is an advanced subset of machine learning, which is now emerging in numerous traditional and new areas. Deep learning has gained tremendous attention from researchers and practitioners in recent years. In comparison with conventional techniques, deep learning has generated experimental results that show extraordinary success in different fields such as cyber security, natural language processing, image processing, biotechnology, speech recognition and translation, and robotics [8] [9] [10]. In deep learning architecture, there are quite a few intermediary layers between the input and output layers. These intermediary layers allow deep learning models to pick up patterns and perform classifications. Deep learning is a general solution that can apply to almost any field, whether new or old, and has proven successful in solving almost any kind of problem [11].

### B. Types of Deep Learning Techniques

Deep learning techniques can be classified into three categories: supervised, unsupervised, and semi-supervised learning techniques.

1) *Supervised deep learning techniques:* Supervised deep learning techniques make use of labelled input and output datasets. Examples of supervised deep learning techniques are Deep Neural Network (DNN), Convolutional Neural Network (CNN), and Recurrent Neural Network (RNN).

2) *Unsupervised deep learning techniques:* In machine learning, unsupervised deep learning techniques make use of unlabelled datasets. Clustering is an example of unsupervised deep learning. In unsupervised deep learning, learning agents identify and study indefinite relationships among input data. It promises to identify extremely intricate and nonlinear models with many free parameters. Deep unsupervised learning makes use of millions of parameters and unlabelled data. Generative Adversarial Network (GAN), Restricted Boltzmann Machine (RBM), and Auto Encoder techniques are used in unsupervised deep learning.

3) *Semi-supervised deep learning techniques:* This type of learning occurs when datasets are not completely labelled. It is a blend of unsupervised and supervised deep learning. First, the role of supervised deep learning is to identify key features from the data for which outputs are known. Then unsupervised deep learning takes place, in which less information about the data is

available, basically no information about the output. This unsupervised deep learning helps in exploiting just input data to identify other features. For instance, in a face recognition problem, supervised learning can identify that eyes are an important feature to differentiate faces from other objects. Then unsupervised learning identifies other new features such as eyebrows and noses and lips as important to identify a face in case eyes are not visible. Thus unsupervised learning improves the overall generalizability of a semi-supervised learning model [12]. Semi-supervised deep learning techniques include Deep Reinforcement Learning (DRL), GAN, and RNN.

Deep Neural Network (DNN): An Artificial Neural Network (ANN) has been mapped on human neurons to solve identification- and classification-related problems. A DNN is an advanced form of ANN that consists of several hidden layers between input and output layers. Each layer contributes to improving classification accuracy. The following sections discuss specialized forms of DNNs. Fig. 1 shows the architecture of a DNN.

Convolutional Neural Network (CNN): CNNs are a type of ANN that support recognition- and classification-related tasks. CNNs are similar to a multi-layered simple neural network, albeit with the difference that unlike in other neural networks, the layers in CNNs are stacked. A CNN works in a similar way to how humans process visuals, with the ability to process multidimensional images [11]. The basic architecture of a CNN consists of a feature extractor and a classifier. This architecture can be further distributed into three type of layers: convolutional, pooling, and fully connected layers. These layers are sandwiched between input and output layers. At the input layer, the input parameters are specified, including height, width, and depth. Odd numbered layers are for pooling, whereas even numbered layers are dedicated for convolution tasks. Convolutional layers extract and create feature maps, which are then processed through an activation function and biased to produce the final output. The function of each odd pooling layer is to reduce the dimensions of the output produced by the former convolution layer. This step is necessary because the exponential increase in dimensions make the dimensions increasingly difficult for the computer to process. Finally, the output produced by pooling and convolutional layers is given as an input to the classification layer, or the fully connected layer [13]. In the classification layer, the features are collected, and activation functions are applied. This layer is computationally expensive, so alternatives such as global and average pooling layers have been reported that reduce parameters, thereby reducing the overall complexity at this layer. Fig. 2 presents architecture of a CNN.

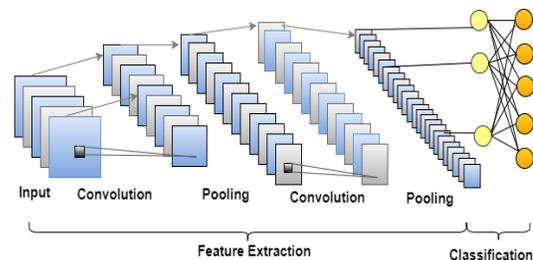


Fig. 1. Deep Neural Network.

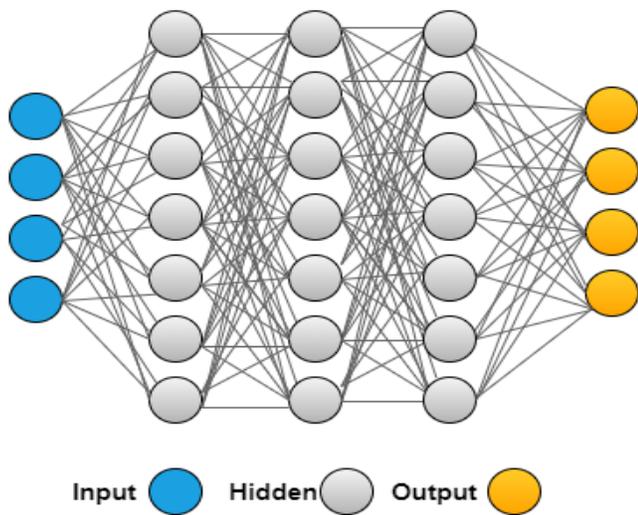


Fig. 2. Convolutional Neural Network.

Recurrent Neural Network (RNN): CNN and conventional neural networks work on fixed sized vectors as input and output, e.g., the images and probability of classes respectively. In contrast, the distinguishing quality of an RNN is that it can handle variably sized vectors. RNNs are iterative in nature, which allows them to pass on information through each iteration. RNNs can be considered as replicas of the same network. They can retain information from the past to interpret, present, and predict the future. For instance, an RNN model can retain information about previous video frames to understand the present and to predict and produce future frames. RNNs have also been used to solve natural language processing [14], text mining, time series, and financial data related problems [6]. The limitation of this neural network is that it faces a ‘vanishing gradient problem’, though this problem has been solved in the literature by the invention of Long Short-Term Memory (LSTM).

Generative Adversarial Network (GAN): A GAN involves unsupervised deep learning where the dataset is not labelled. It uses input data to generate previously unknown patterns and then uses learnt patterns to generate new but similar samples of data. GANs make use of two neural networks, i.e., a generator and a discriminator. These neural networks compete against each other in an adversarial fashion. The generator generates sample data, whereas the discriminator compares the sample with input data and determines whether the generated sample is close enough to be acceptable. The feedback is fed back to the generator, which helps it to perform better. GANs have been used to solve numerous problems in diverse fields. GANs use deep learning methods, which is why they are categorized as both semi-supervised and unsupervised learning. For instance, GANs support image, video, and voice generation. GANs are also being used in game development.

Restricted Boltzmann Machine (RBM): RBM is another example of unsupervised learning. Just like GAN, RBM is also a generative approach that generates samples based on unlabelled input data. An RBM is composed of two layers, i.e., hidden and visible. The standard Boltzmann Machine is known for its slow learning process. The training part in an RBM can be exhibited using a Boltzmann Machine, which is a two-

layered network. It makes use of randomly distributed probability-based binary pixel and feature detectors. The RBM is based on hidden variables and undirected graphs. The stochastic nature of the RBM and its slow training phase makes the overall approach computationally expensive. RBMs can be used to solve classification and dimension reduction problems.

Deep Reinforcement Learning (DRL): This learning approach learns from an unknown environment. DRL is different from other supervised and unsupervised deep learning approaches since these approaches model data while DRL models environments. DRL can be thought of as an approach that suggests actions to be performed based on a given environment. Other deep learning approaches are exploited by DRL to model data based on the environment being modelled. DRL can be applied in numerous fields to determine actions, for instance, in engineering and mathematics. In addition, it can be used for decision making in investment markets.

Deep Auto Encoder (DAE): This approach is categorized as unsupervised deep learning. Auto encoders are used for encoding input data and to learn features from it. DAE consists of two parts: the encoder and decoder. In the encoding part, input data is encoded. In the decoding part, real features are generated. DAE encoders can be thought of as stacked data-driven auto encoders and are famous for solving dimension reduction problems. However, there are a few limitations to DAE approaches. For example, they are highly sensitive to input errors and face the ‘vanishing error’ problem [11].

Transfer Learning (TL): In conventional machine learning practices, a model is designed, created, and trained to produce accurate results. Mostly weights are initialized randomly before the start of a training process. As a result, models learn slowly, adjusting the weights on each iteration to reach a certain level of accuracy. TL is different from the traditional machine learning training process. It makes use of source information to improve the learning rate of a related target model. TL is a process in which pre-trained models are used to initialize the weights of a new model. This process can greatly improve the time required by a target model to reach higher classification accuracy. Performance of TL is dependent upon the DL algorithm being used by the model. Usually the last layer of the pre-trained model is removed, and a fully connected model is attached with the number of classes in the target model. If the target model performs well as compared to a model that learnt from scratch, then this process will be considered as positive TL. However, if the target model does not benefit from pre-trained model and performance is degraded as compared to the model that learnt from scratch, then this will be considered as negative TL [15]. This learning produces the best results when there is a limited amount of training data [13].

### III. PROPOSED METHODOLOGY

This study presents an SLR by following the guidelines presented by [16] [17]. These guidelines are well accepted in the software engineering community and have been followed by a number of studies [18] [19]. Fig. 3 shows the review protocol. This review is divided into three phases. The first phase presents the plan. This phase presents the research questions that are formulated based on the objectives of this study. The second phase is divided into three parts: definition

of the search strategy with the help of keywords, formulation of inclusion and exclusion criteria for study selection, and data extraction of selected attributes to answer the research questions. Finally, the data is synthesized. In the third phase, the results are reported by answering each research question. The following sections align with the phases and steps as outlined in Table I.

A. Research Questions

The following research questions align with the objectives of this study.

RQ 1. What is most state of the art in the field of deep-learning-based detection and classification of bacterial colonies?

This question is further sub-divided into the following questions:

RQ 1.1 What are the techniques/deep learning models that have been used in the primary studies?

Motivation: This research question is answered by identifying the deep learning models used in each primary study, for instance, the CNN and RNN. The summarized research methodology of deep learning models presented in primary studies are reported. The reporting covers, for instance, the number of layers and steps used during the pre-processing, training, and testing phases of each study.

RQ 1.2 Which types of learning have been applied?

Motivation: This research question aims to present the types of learning used by the techniques. For instance, supervised learning, unsupervised learning, and semi-supervised learning. This is assessed by mapping each deep learning model on the taxonomy presented in Fig. 4.

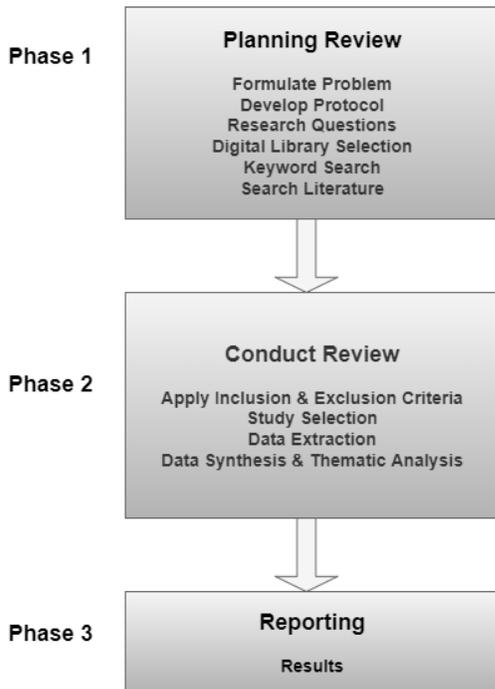


Fig. 3. Review Protocol.

TABLE I. STEPS FOLLOWED IN EACH PHASE

Phases	Steps Followed
Planning	Problem Formulation
	Protocol Development
	Research Questions
	Online Digital Library Selection
	Formulation of Query String
	Inclusion and Exclusion Criteria Definition
Conducting Review	Study Selection
	Attribute Identification
	Data Extraction
	Data Analysis
Reporting	Report Results

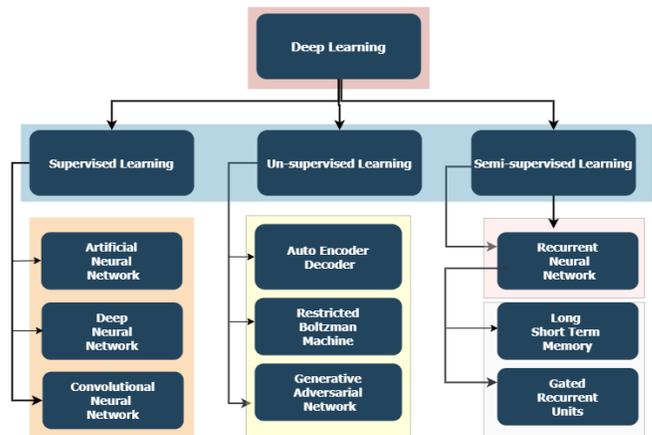


Fig. 4. Taxonomy of Deep Learning Methods.

RQ 1.3 What tools are available for deep-learning-based detection and classification of bacterial colonies?

Motivation: Availability of tools is an important concern for practitioners. To answer this research question, we aim to discover and report the tools developed and used by primary studies. The answer to this research question includes a list of tools presented and used in primary studies.

RQ 2. What type of datasets have been used for evaluation in the primary studies related to deep-learning-based detection and classification of bacterial colonies?

Motivation: The answer to this research question involves identifying the name and types of datasets used for the training and testing of deep learning models presented in primary studies. The datasets are categorized into three categories: academic, open-source, and industrial. Datasets constructed in lab environments that are not affiliated with any organization or institute are considered academic. Open-source datasets are those that are publicly available, well known, and used in similar studies. Datasets received from a specific organization that are not publicly available and solely used for the scope of a particular primary study are called industrial datasets. This research question is further divided into the following sub-questions.

### RQ 2.1 What are the characteristics of the datasets?

Motivation: This research question aims to identify the characteristics of data, such as type of sample (microscopic images, spectroscopic data, genomic data, etc.); number and size of dataset; and number of classes. The answer to this research question is a comparative analysis presented in a table listing dataset name, number of classes, type, and size.

RQ 2.2 What performance evaluation metrics do the studies use to evaluate the performance of deep-learning-based techniques for the detection and classification of bacterial colonies?

Motivation: This research question aims to investigate the performance of a deep learning model based on its ability to perform the classification accurately. To answer this research question, the evaluation data of each model were extracted for a given dataset. Different studies incorporate different metrics for performance evaluation depending on the dataset and deep learning technique used. Performance metrics can be accuracy (in percentage), Area Under the ROC Curve (AUC), precision, receiver operating curve, F1 score, sensitivity, confusion metrics, etc. The answer to this research question is a comparative analysis presented in a table providing the name of the deep learning model, dataset, and highest accuracy achieved on that dataset.

#### B. Digital Library Selection

The search was carried out using the large extensive databases shown in Table II. We searched a total of five digital databases: Google Scholar, IEEE Explore, ACM, PubMed, and Springer. We searched on the basis of keywords, titles, abstracts, and (in the case of Google Scholar) full texts to identify the relevant primary studies. These libraries provided almost complete sets of relevant studies. To complete the search process, a manual search was also conducted by looking into relevant journals and identifying articles from references of primary and secondary studies.

TABLE II. DIGITAL LIBRARIES AND URL

Digital Library	Uniform Resource Locator
Google Scholar	<a href="https://scholar.google.com">https://scholar.google.com</a>
ACM	<a href="https://dl.acm.org">https://dl.acm.org</a>
IEEE Explorer	<a href="https://ieeexplore.ieee.org">https://ieeexplore.ieee.org</a>
PubMed	<a href="https://pubmed.ncbi.nlm.nih.gov">https://pubmed.ncbi.nlm.nih.gov</a>
Springer	<a href="https://springer.com">https://springer.com</a>

#### C. Query String Formulation

By using the keywords identified in Table III, query strings were formulated. Keywords were identified based on the research questions. Synonyms and alternate terms and spellings were used to perform the advanced search. Query strings were used to perform advanced searches on the digital databases. The Boolean operator OR was used for synonyms and alternate keywords, and the operator AND was used for connecting keywords and phrases. Multiple versions of query strings were developed and executed on different databases in order to find as many relevant studies as possible.

TABLE III. QUERY STRING

Digital Library	Key Words Searched
Google Scholar	("Deep Learning" AND Bacteria*) AND (Classification OR Identification) AND Microscope* AND Image
IEEE Explore	("Document Title": Deep Learning ) AND ("Document Title": Bacteria Classification)
Springer Link	"Deep Learning" AND Bacteria AND Classification
PubMed	((Deep Learning) AND (Bacteria[Title/Abstract])) AND (Classification[Title/Abstract])
ACM	[Publication Title: deep learning] AND [Abstract: bacteria*] AND [Abstract: classification identification] AND [Publication Date: (01/01/2021 TO 12/31/2021)]

Table III presents the main query strings applied to the digital databases.

#### D. Inclusion and Exclusion Criteria

An SLR includes and excludes papers from the study pool with the help of well-defined criteria. In order to select relevant papers, a simple yet straight forward inclusion and exclusion criteria was formulated. In the first phase of study selection, studies were selected based on their titles. In the next phase, abstracts of studies were reviewed. Finally, full texts of included studies were reviewed, and any study that was not in alignment with the inclusion criteria was excluded. The inclusion and exclusion criteria are stated below:

##### 1) Inclusion Criteria

IC1: Published in the year 2021

IC2: Written in the English language

IC3: Full text of paper is available

IC4: Peer reviewed

IC5: Discusses deep learning methods to detect and classify bacterial colonies

##### 2) Exclusion Criteria

EC1: Published before 2021

EC2: Written in a language other than English

EC3: A version other than the most recent version (if multiple versions are available)

EC4: Non-peer reviewed (e.g., presentations or books)

EC5: Duplicate article

EC6: Discusses classification of other microorganisms such as viruses and other non-bacterial microorganisms

#### E. Conducting Systematic Literature Review

This section presents the study selection, data extraction, and synthesis process.

1) *Selection of primary studies:* The search across five online digital libraries retrieved a total of 310 articles. In the first pass, the titles and abstracts of all the articles were

analysed. As a result, 50 articles were selected (IC2, EC2, IC5, and EC6). In the second pass, the duplicate articles (i.e., papers appearing more than once) were removed (EC5). Furthermore, any non-peer reviewed articles such as books, magazines, lecture notes, editorials, and presentations were removed (IC4, EC4). Articles whose full text was unavailable were also excluded (IC3, EC3). A manual search was also conducted by looking into relevant journals and identifying articles from the references of primary and secondary studies. As a result, a total of 38 articles were present in the study pool. Fig. 5 shows the study selection process.

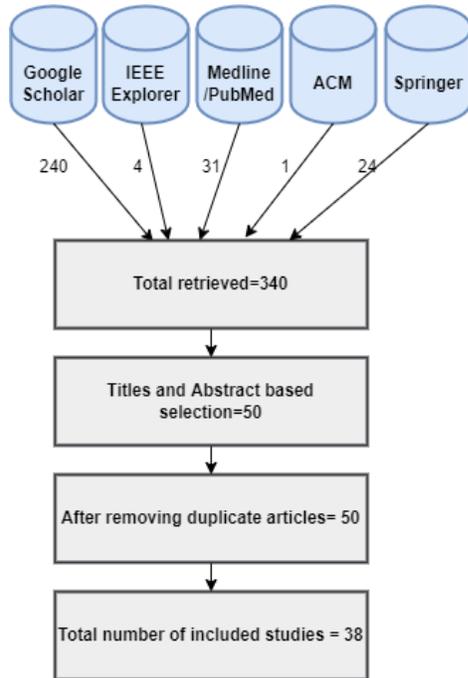


Fig. 5. Study Selection Process.

2) *Data extraction and synthesis:* The selected attributes were extracted from the primary studies and stored in a spreadsheet for analysis. We also extracted the quantitative data, such as number of datasets, size, number of classes, and accuracy, and the qualitative data, such as type of learning, type of dataset, and steps followed to develop the deep learning model. Data synthesis is a technique to summarize reported evidence from included primary studies to answer research questions. Table IV presents the data attributes extracted from each of the primary studies and maps those attributes on each respective research question. To synthesize qualitative data, a descriptive analysis was conducted, and to synthesize quantitative data, a thematic analysis was performed.

#### IV. RESULTS

This section presents the results and answers the research questions presented in section 3.1.

RQ 1. What is most state of the art in the field of deep-learning-based detection and classification of bacterial colonies?

TABLE IV. DATA ATTRIBUTES' MAPPING ON RESEARCH QUESTIONS

Data Attributes	Possible Values	Research Question
Deep Learning Model	e.g. CNN, RNN, DBN, AE	RQ 1.1
Type of Learning	Supervised Unsupervised Semi-Supervised	RQ 1.2
Tool	Tool Name	RQ 1.3
Tool Language	e.g. MATLAB, Python etc.	RQ 1.3
Tool Availability	Available Not available	RQ 1.3
Name of Dataset	e.g. DIBaS	RQ 2
Dataset Type	Academic Open source Industrial	RQ 2
No. of Classes	e.g. 3, 5	RQ 2.1
Type of data sample	Spectroscopic, microscopic, etc.	RQ 2.1
Highest Accuracy Achieved	Value in percentage	RQ 2.2
Evaluation Metrics	F1 score, precision, sensitivity, etc.	RQ 2.2

To identify what is most state of the art in deep learning approaches used for bacteria classification and identification, the information presented in Table IV was extracted and used to answer all sub questions.

RQ 1.1 What deep learning models/architectures have been used in the primary studies?

The primary studies were classified using thematic analysis, which involves identifying different patterns. A word cloud based on the extracted keywords is presented in Fig. 6. More than 75% of the primary studies are based on different architectures of CNN. Fig. 7 shows the distribution of deep learning architectures. Studies were further classified according to various CNN architectures, for example, ResNet, UNet, SqueezeNet, MobileNet, and InceptionNet. Fig. 8 shows the distribution of architectures within CNNs. A large number of primary studies incorporate ResNet architecture because it can accommodate as many as one thousand layers to achieve greater performance. The following sections discuss the research summaries of the studies categorized in each of these architectures.

1) *CNN-Based architectures:* In [20], the authors identify and classify three types of food-borne bacteria with a cytometric approach on micro-fluidic impedance. The three type of bacteria, namely 'Salmonella Enteritidis', 'Vibrio Parahaemolyticus', and 'Escherichia coli', were classified with the help of a CNN with 100% accuracy. The authors claim that this impedance-based technique can classify unlabelled data. This system can also detect pathogenic bacteria, thus it could be prolific in clinical diagnosis. Regarding the architecture of the CNN, the CNN was composed of one input layer, two convolutional layers, one fully connected layer, and one output layer. The depth of convolution was 8, and Rectified Linear Unit (ReLU) was used as an activation function. The same



In [26], the authors develop a spectra identification technique to identify the bacteria responsible for causing urinary tract infections (UTIs). A CNN was developed to perform the classification of bacterial species and their sensitivity to antibiotics. The proposed CNN model consists of four convolutional and max pooling layers. Two fully connected layers were used for classification. The results indicate that, as compared to traditional UTI bacterial identification techniques, this technique based on surface-enhanced Raman spectroscopy is faster and has an accuracy level of 96%.

In [27], the authors develop a deep learning approach to identify the geometrical characteristics of sulfate-reducing bacteria in order to avoid corrosion. They use a deep CNN model on Scanning Electron Microscopic (SEM) images. A modified watershed algorithm was used for bacterial cell segmentation before counting of the cells and identification of geometric properties such as height and width. The model takes unprocessed SEM images as input and produces geometric properties (height and width) as output. The model uses Deeplabv3+ pre-trained on ResNet50, ResNet18, and MobileNet. The results show that the proposed model can detect individual and clustered bacterial cells effectively in the presence of other objects.

In [28], the authors developed a CNN-based classifier to distinguish between two types of bacteria, i.e., MRSA and MSSA. The proposed CNN is a shallow model that takes 1D spectra as input and extracts feature maps. The model classifies spectra into two classes, so it can be considered as a binary classifier. The results indicate that the proposed approach can identify and distinguish two types of bacteria with an accuracy of 100%. In [29], the authors made use of a CNN-based model called VGG-16 for classification of bacterial colonies. Pre-trained VGG-16 was used with atrous convolution instead of a conventional CNN. The proposed model altered the standard VGG16 model by introducing additional dilated convolutional layers; the rest of its architecture was similar to original VGG-16. The performance of the proposed approach on test data was 94.8%.

In [30], the authors investigate the identification of growth paths of E.coli bacteria on non-transparent metallic mediums. Their aim was to identify multiple stages of bacterial image formation. These steps included identification of base, colony formation, bacterial dispersion, and crystallization. They incorporate a deep CNN model with four convolution layers followed by max pooling layers and a classification layer followed by a SoftMax layer. The authors in [31] attempt to develop a system to automatically calculate the number of bacterial cells in a colony in water samples. A CNN was developed to identify a specific type of bacteria called E-coli. The CNN model comprises six convolutional followed by two fully connected layers. Another deep learning model R-CNN was employed to automatically count the number of bacterial cells in a colony. This model was developed with the help of previously trained ResNet-50, hence use of TL. Classification layers of ResNet were replaced by a layer with nine neurons representing nine classes of R-CNN. The results indicate that the proposed models works significantly well with an accuracy

of 97% in automatic detection and cell counting of E-coli bacteria in colonies.

The authors in [32] use a CNN to detect bacterial cells from three-dimensional (3D) fluorescent microscopic images. The CNN is composed of 11 layers in total, distributed as follows: two convolutional layers followed by max pooling layers and two fully connected dense layers that use SoftMax function. The results indicate that the proposed approach works effectively with an accuracy of 95% and can detect bacterial cells from 3D fluorescent images.

2) *ResNet architecture*: In [33], the authors aim to identify the structural features of the G20 bacteria that are normally present on steel surfaces and cause corrosion. Data samples are SEM images. Mask Region CNN (RCNN) and Deep Convolutional Neural Network (DCNN) were used for segmentation of bacterial instance and identification of grouped bacteria respectively. The authors incorporate a previously developed platform named DeepLABv3+ for implementing deep learning architectures. RCNN is a pre-trained architecture with pre-extracted feature maps of images. Comparison was performed with deductions of experienced human experts. The results indicate that RCNN and DCNN are far faster with an accuracy of 81% as compared to manual and conventional approaches to detect bacteria from biofilms.

In [34], the authors present a CNN-based approach to classify three species of gram-positive bacteria through Whole Slide Images. Data were pre-processed to segment bacteria from background. The segmented bacteria images were then fed to a classifier, which classified them into three classes. Pre-trained ResNet was used to segment the bacteria from the background. The authors conclude that ResNet architecture can be effective in differentiating among three gram-positive bacteria with an accuracy of 81%.

In [35], the authors propose a bacilli detection approach based on deep CNN. Specifically, ResNet, SqueezeNet, and VGG-16 are used for training and testing to identify which models work best in bacilli identification. Use of pre-trained architecture indicates that the models used TL. Bacterial images were segmented from the background using K-means clustering and colouring techniques. Images were resized to 224x224x3 and fed to the model. ReLU and sigmoid activation functions were used between layers. Overall, SqueezeNet outperformed the other CNN models with an accuracy of 97%.

The authors in [36] employed five CNN-based architectures to differentiate between 33 different species of pathogenic microbes. These architectures included ResNet50, Mobile Net, ResNetv2, Inception Net, and DenseNet. All the architectures used TL, i.e., they were pre-trained to reduce the time required to achieve better performance. Fine tuning was used to vary the weights of parameters in deep and shallow layers. Shallow layers identified basic line features whereas deeper layers identified other complex features. The input to the models was a 224x224 image. Stochastic Gradient Approach was used to prevent the technique from getting stuck in local minima. MobileNet performed better than the other architectures with an accuracy of 96.8%.

In [37], the authors developed a tool called Motility-J to identify bacteria and detect surfaces that are covered with pathogenic bacteria. The authors emphasize that pathogenic bacteria develop features such as flagella to move across surfaces in order to survive in a constrained environment. The tedious task of labelling datasets was alleviated by labelling images with a semi-automatic technique. The authors used image segmentation to identify the part of image where bacteria are present from the background image and used that part as input to the model. A number of image processing techniques such as noising and erosion were used to obtain bacteria segments. A number of CNNs were used including ResNet50, ResNet101, FBNet, and EfficientNet B3 with fine tuning and TL. Fine tuning was done such that the last convolutional layer was replaced by a linear layer. All the models were trained using input images.

In [38], the authors differentiate longitudinal bacterial divisions (Fission) from horizontal and other divisions. They make use of a pre-trained CNN called ResNet with TL. Comparison was conducted of a pre-trained model with an un-trained model. The results indicate that classification accuracy for pre-trained ResNet was 99.67%, better than un-trained ResNet.

3) *UNet architecture*: In [39], the authors developed an approach for segmentation and classification of six bacterial species. They employed UNet architecture for segmenting the SEM image into foreground and background images. In the classification part, VGG16 was used to classify bacteria species before counting the number of cells. The authors kept the segmentation and classification independent so that they could learn independently and improve their individual accuracies. The results indicate that the performance of the proposed approach as compared to standard CNN was better with a classification accuracy of 95.8%.

The authors in [40] aim to identify bacteria and other harmful pathogens that contaminate the bloodstream and cause life-threatening health problems. The authors incorporated CNN-based architecture called UNet to identify the presence or absence of bacterial cells in dark field microscopic images. The authors emphasize that UNet architecture works well in image segmentation and object detection. The authors divided the architecture into four models. The first model used the concept of early stopping to prevent over fitting in case the performance on the training set does not increase even after a few iterations. The second model does not apply early stopping. The third model implements a loss function. The results indicate that the proposed UNet third model performed well with an accuracy of 96.6%.

In [41], the authors build on a previously proposed BCM3D approach. BCM3D is a combination of image processing techniques and CNN to detect, count, and segment single cell bacteria from biofilms. BCM3D2.0 addresses the challenge of segmenting bacterial cells from dense biofilms and a low signal-to-background ratio. Fluorescence microscopic images were used to train and test the new approach. The authors processed the images and created two types of transitional images for object localization and boundary detection. The proposed

approach not only creates a 3D outline of an object but also measures its distance from nearby objects. A UNet-based CNN was incorporated with two convolutional layers followed by max pooling and a linear function followed by a classifier. Another CNN was trained to determine the physiological shape of segmented objects. This CNN consisted of three convolutional layers: two average pooling layers followed by a sigmoid layer.

In [42], the authors aim to measure the number and length of bacterial cells and the overall area covered by biofilms. They claim to have solved the object segmentation problem that occurs in closely located or overlapping cells. The authors combined deep-learning-based UNet model with 'region-based ellipse fitting technique' to segment, count, and measure bacterial cell instances from biofilms. UNet architecture is an encoding decoding model where convolution and deconvolution take place on input images. The final output of the model is a logically outlined mask of bacteria cell instance where each pixel is categorized to a particular class. In the second phase, the ellipse technique was applied, in which centroids and distances were calculated and ellipses with similar angles were combined iteratively. This technique performed well with a recall of 93.6%.

4) *SqueezeNet*: The authors in [43] propose 12 CNN-based models to classify bacterial species. The proposed models have minor differences, though all were pre-trained and fine-tuned. An open-source dataset called DIBaS was used to train and test the models. Furthermore, the dataset was augmented to increase its size. Augmentation was done by cropping and zooming in multiple times. A total of 24,073 images (including the originals) were present after the augmentation. Models were trained by resizing the images to 224x224 pixels. Some architectures used in this paper are Efficient-net, SqueezeNet, Mobilenetv2, Mobilenetv3, and ShuffleNet. Almost all the architectures used pre-trained models and fine-tuned them such that the last layer was modified to 32 neurons representing the number of bacterial classes. Fine-tuning also reduced the number of parameters drastically thus enabling the models to consume less resources. Comparison of proposed models was done with other techniques with and without data augmentation. The results indicate that augmentation can have a great impact on performance of a model.

In [35], the authors propose a bacilli detection approach based on deep CNN. Specifically, SqueezeNet, ResNet, and VGG-16 are used for training and testing to identify which models work best in bacilli identification. The open-source dataset ZNSM-iDB, which has 2,000 images, was pre-processed with colouring techniques. Bacterial images were segmented from the background using K-means clustering and colouring techniques. Images were resized to 224x224x3 and fed to the model. ReLU and sigmoid activation functions were used between layers. Overall, SqueezeNet outperformed rest of the CNN models with an accuracy of 97%.

5) *Inception net*: The authors in [44] developed a classifier to identify the presence and absence of filamentous bacteria in waste water. They used pre-trained inception v3, an open-

source CNN-based architecture, to train the classifier into two classes. Inception v3 was developed by Google. It consists of 22 layers and 24 parameters. It has been trained on an image-net dataset, which is well known and large. Using a pre-trained inception net allowed the authors to use it as it was for identifying bacteria in waste water. The results indicate that the Inception v3 classifier was able to identify filamentous bacteria in abundance.

6) *Stacked auto encoder*: In [45], the authors present a DNN-based approach to differentiate between two types of bacteria: MSRA and MSSA. Stacked Auto Encoder is used to perform the training and testing of the unlabelled dataset. The authors highlight that auto encoders are good at learning complex features from unlabelled data. They stack a number of auto encoders such that the first's hidden layer will feed the second's hidden layers followed by SoftMax layers. Comparison of the proposed approach was conducted with six machine learning approaches including K nearest neighbours, Support vector machine, and decision trees. The results indicate that the proposed stacked Auto Encoder accurately detects MSSA and MSRA bacteria with an accuracy of 97.6% and AUC of 0.99.

7) *Recurrent neural network long short-term memory*: in [46], the authors aim to identify bacteria from marine water. A model based on Recurrent Neural Network (RNN) called Long Short-Term Memory (LSTM) was employed to do the task. The results were compared with a simple CNN mode with two convolutional layers and max pooling and a classification layer. The LSTM model consisted of two LSTM layers, each with 64 neurons followed by a classifier with 8 neurons representing eight marine bacterial species. Both models were trained, validated, and tested on the same data so that performances would be comparable. The results indicate that the proposed LSTM method performed better than the conventional CNN with an accuracy of 94%. Furthermore, the LSTM-based model was faster and more accurate.

The authors in [47] investigate the use of LSTM for identification and classification of food-borne bacteria. The authors make use of Hyper Spectral Microscopic Images, which were pre-processed and fed to an LSTM model as well as to three other models: PCA KNN, SCM, and LDA. The results of the LSTM model were compared with those of the three classifiers. The LSTM model is composed of several blocks, a dense layer and a SoftMax layer that contains five neurons representing five classes of bacteria. Three types of Region of Interest (ROI) were utilized to extract features of bacteria: inner cell, outer layer and boundary ROIs. The results indicate that centre ROI is a better feature to consider for bacterial classification. The proposed RNN-based LSTM model performed better than other models with an accuracy of 92.9 %.

#### RQ 1.2 Which types of learning have been applied?

This research question is answered by mapping each type of deep learning model on the taxonomy proposed in Fig. 3.

In [20], the authors make use of unlabelled impedance data, which means the proposed technique is unsupervised. The

authors in [26] use unlabelled Raman spectroscopy data to identify UTI-causing bacteria. The authors in [28], proposed binary classifiers using unsupervised learning. The classifiers detect two types of bacteria: MSSA and MRSA. The authors in [45] make use of stacked auto encoders for training and testing an unlabelled dataset, so we categorize this study under unsupervised learning models. The authors in [35] perform unsupervised learning as they detect bacilli-shaped bacteria from an open-source dataset of microscopic images. In [48], the authors develop a binary classifier using supervised learning. The proposed classifier classifies bacteria into two classes: harmful and benign bacteria. The authors in [49] propose a graph-based unsupervised technique called M-Lcuts that identifies numerous bacterial clusters in 3D space.

The authors in [47] make use of live spectral analysis to train an RNN-based LSTM model. The authors in [46] propose an RNN-based model called LSTM to identify bacteria from marine water. Since the authors consider the LSTM model semi-supervised, we categorize it under semi-supervised learning.

The authors in [21] collected six types of bacteria through a gram-staining method from a university in Malaysia. The dataset was annotated hence this technique can be classified as supervised learning. In [50], the authors differentiated between three strains of 'Klebsiella pneumonia' by using supervised learning'. The study [22] use a labelled dataset to detect bacterial vaginosis. The authors in [38] and [40] performed supervised learning by manually labelling the dataset into two classes. The authors in [23] performed bacteria sequence classification by using labelled barcode sequences of an open-source dataset. In [51], the authors performed supervised learning as they annotated the dataset into two classes: Escherichia coli and 'Mysococcus Xanthus'. The authors in [39], [27], and [33] used a labelled SEM-based dataset for identifying and classifying bacteria species. In [52], the authors used a labelled dataset to investigate the classification accuracies of some deep learning architectures on three types of bacteria. In [44], the authors used supervised learning and annotated microscopic images obtained from waste water. The authors in [24] used supervised learning for identification of food-borne bacteria, specifically 'Salmonella Typhimurium'. The authors in [53] make use of an annotated dataset of microscopic images to identify bacteria from images. In [25], the authors performed supervised learning as they made use of an annotated dataset composed of microscopic images. The authors in [43] proposed a supervised learning approach for quick identification of bacteria. In [54], the authors propose a supervised learning method to differentiate between different species of gram-positive bacteria through Hyper-spectral Microscopic Images. The authors in [29] performed supervised-learning-based bacterial colony classification by employing TL. The authors in [37], [31], and [41] made use of supervised learning to train models. In [36], the authors propose a fine-tuning-based supervised learning approach for pathogenic bacteria identification. Similarly, the authors in [30], [42], and [32] also made use of supervised learning techniques.

To summarize, 77% of the architectures belong to supervised learning and thus make use of annotated or labelled datasets. Furthermore, 5% of the architectures belong to semi-

supervised learning. The remaining 18% of architectures belong to unsupervised learning. Fig. 9 presents the distribution of deep learning techniques for bacterial classification using three types of learning approaches. Fig. 10 presents the distribution of techniques within unsupervised learning architectures.

RQ 1.3 What tools are available for detection and classification of deep-learning-based detection and classification of bacterial colonies?

Development of tools has important implications for practitioners. Development of automated deep learning tools allows academic research to support practitioners. It also helps novice researchers and beginners to adapt to deep learning methods by overcoming the technology barrier. Table V presents the list of available tools in the field of bacterial detection and classification in deep learning. It also presents their languages, frameworks, and usage information.

Seven deep learning tools are available for detection and classification of bacteria and their colonies. These tools perform various functions and work on different levels. Functions such as image segmentation, de-noising, cell counting, and labelling can be performed. For instance, the authors in [53] develop a tool called eHooke for automatic analysis of images to detect bacteria. In particular, spherical bacteria are targeted. An ANN was used as a deep learning model to detect this bacteria. eHooke was developed in Python. The eHooke tool is available publicly for download. ZeroCostDL4Mic was used in [52]. This tool supports different architectures like U-Net, CARE, pix2pix, StarDist, and SplineDist. Furthermore, the authors in [51] propose Mistic, which is a tool based on CNN and UNet architecture that helps in image segmentation in dense and multi-species environments. Mistic can work on numerous types of images such as two-dimensional (2D) fluorescence, bright field, and phase contrast images, and it does not depend on microscopes. It makes use of Shape Index map (SI), which can be derived from microscopic image data, so diverse types of sample data can be used for bacterial identification. A CNN-based UNet architecture is trained to identify the shapes of bacteria by extracting their features. The general workflow of Mistic is to take input of any type of image, convert that input into SI, and then segment by UNet. Mistic-Pip and Mistic-GUI are available for download. The authors in [37] developed a tool called Motility-J to identify bacteria and detect surfaces covered with pathogenic bacteria. The tedious task of labelling datasets was also improved by labelling images with semi-automatic technique. eHooke, Mistic, and MotilityJ have been used for image segmentation and classification. Interestingly, the majority of the tools have been developed using Python language. Fig. 11 presents the common frameworks reported by primary studies for development of deep learning architectures whereas Fig. 12 presents languages used by primary studies for implanting architectures. The majority of studies have used TensorFlow, PyTorch, and Keras framework and Python for implementing architectures.

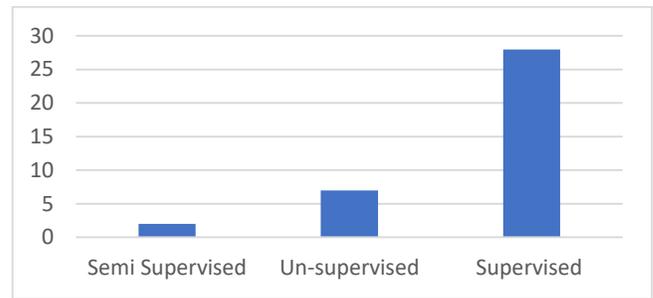


Fig. 9. Distribution of Deep Learning Techniques.

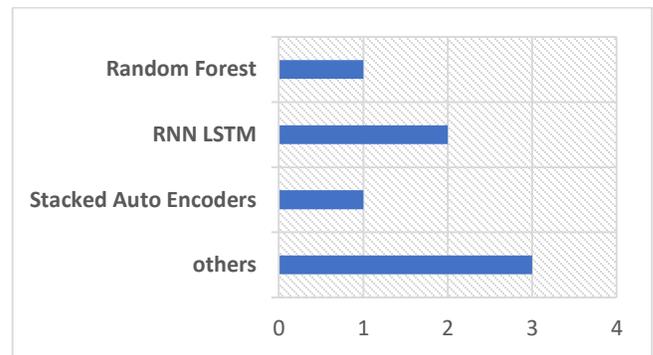


Fig. 10. Unsupervised Learning Architectures.

TABLE V. LIST OF AVAILABLE TOOLS IN BACTERIA DETECTION AND CLASSIFICATION

Ref.	Tool Name	Language	Framework	Usage
[37]	MotilityJ	Java, Python	PyTorch	Classification and segmentation
[31]	Mobile Application GUI	MATLAB	TensorFlow	Bacteria colony quantification
[38]	Anonymous	Python	PyTorch	Classification of longitudinal bacteria division
[51]	Mistic-Pip, Mistic-GUI	Python	TensorFlow	Segmentation in dense colonies
[52]	ZeroCostDL4Mic	Python	Google Colab	Image segmentation, Image de-noising, labelling for rod and spherical shape bacteria
[44]	Automated Microscopic Image Acquisition System	Python	TensorFlow	Obtaining image from waste water
[53]	eHooke	Python	eHooke	Classification, segmentation and quantification of spherical bacteria

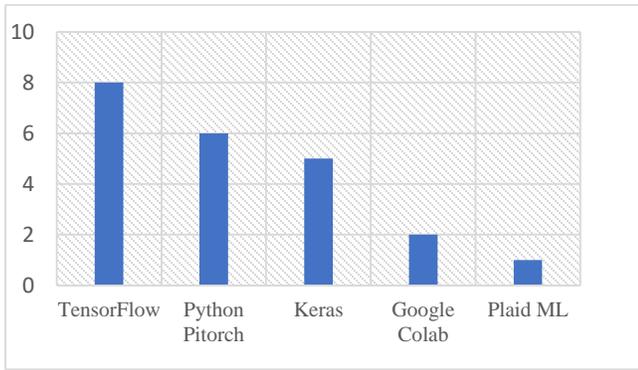


Fig. 11. Frameworks used to Implement Deep Learning Architectures.

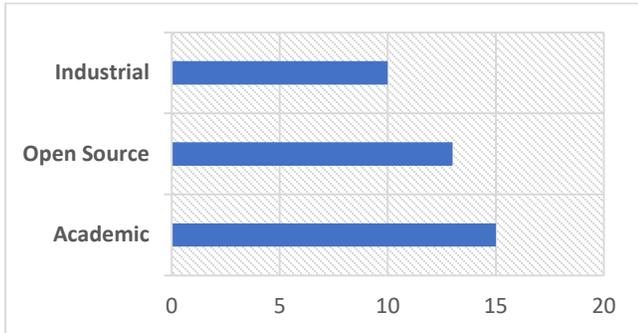


Fig. 12. Languages used to Implement Deep Learning Architectures.

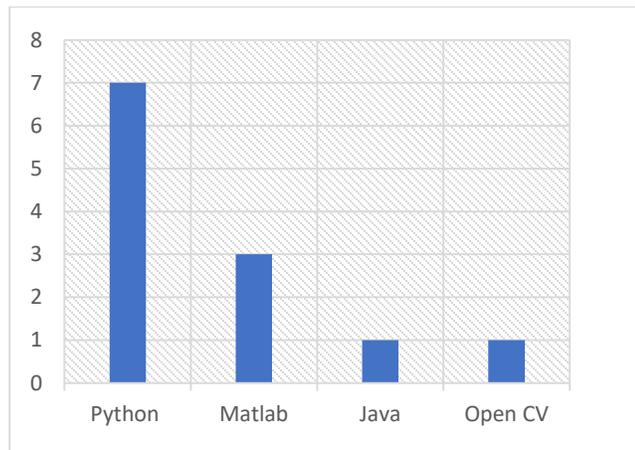


Fig. 13. Types of Datasets.

RQ 2 What type of datasets have been used for evaluation in the primary studies related to deep-learning-based detection and classification of bacterial colonies?

Fig. 13 shows the distribution of type of datasets used by the primary studies. Most of the datasets are academic. Namely, 40% (15) of the primary studies use academic datasets, meaning these datasets were constructed in lab environments and are not associated with any organization or institute. For instance, one study grew bacteria in lab settings under certain temperatures using chemical reagents [33]. Industrial datasets were used in 26% (10) of the primary studies. These datasets come from a specific organization, are not publicly available, and are solely used for the scope of that particular primary study. For instance, one study collected water samples from five domestic waste

water treatment plants in Japan [44]. Since a particular type of waste water treatment plant in Japan was targeted for sample collection, this dataset is categorized as industrial. Finally, open-source datasets were used in 34% (13) of the studies. Open-source datasets are publicly available, well known, and have been used in similar studies. For instance, one study used an open-source database called DIBaS, which is freely available and has been used in other similar studies [25].

The authors in [55] classify bacterial species in the DIBaS dataset. DIBaS is an open-source dataset with 33 classes of bacteria and other microorganisms. This dataset was augmented to increase its size and to avoid over-fitting. Images were resized as 224x224x3. Another open-source dataset of 205 images was incorporated by [48]. In this case, the dataset was divided into different ratios of training and testing data. The open-source dataset DIBaS was also used by [25] for training and testing. The dataset was reduced to 1,669 relevant microscopic images that were resized to 128x128x3. The authors in [35] used the open-source dataset ZNSM-iDB, with 2,000 images pre-processed with colouring techniques. Bacterial images were segmented from the background using K-means clustering and colouring techniques. Images were resized to 224x224x3 and fed to the model. The authors in [29] also used the DIBaS dataset for training, validating, and testing the proposed model. Since the number of relevant images in original dataset was small, data augmentation was used to increase dataset size. Augmentation was done by zooming, flipping, and cropping the original images multiple times. An open-source dataset named ‘‘Bacteria Detection with Dark-field Microscopy’’ (BDDM) used by [40] for training and testing UNet architecture. It contains 366 images of size 128x128, which were manually labelled. The authors in [36] and [43] used DIBaS for training and testing the models. The dataset was augmented to increase its size. Augmentation was done by cropping and zooming in multiple times. A total of 24,073 images (including originals) were present after augmentation. Models were trained by resizing the images to 224x224 pixels. Table VI presents list of open-source datasets in the context of primary studies.

RQ 2.1 What are the characteristics of the datasets?

TABLE VI. OPEN-SOURCE DATASETS REPORTED AND USED IN PRIMARY STUDIES

Dataset	Size	Type	Classes
16SsRNA	393	Gene barcode	3
DIBaS	3000	Microscopic images	33
2DBEST	66	SEM Images	4
BDDM	366	Dark-field Microscopic Images	2
ZNSM-iDB	800	Microscopic Images	2

In [20], the authors detect and classify three types of food-borne bacteria: ‘Salmonella Enteritidis’, ‘Vibrio Parahaemolyticus’, and ‘Escherichia coli’. An academic dataset of 600 microfluidic data was used for training the model. TL is used when there is limited availability of training data. In [21], the authors augmented an industrial dataset with random image transformations such as rotations and reflections in order to reduce the dataset’s bias and increase its size. A total of 44,985

images divided into six classes of bacteria were in the laboratory-provided datasets. Images were resized to 224x224. The authors in [50] used 610 gram-stained microscopic images of dimension 3500x5760. This dataset was provided by a hospital and prepared in a laboratory setting. Three classes were used in the dataset to distinguish between the clones of *Klebsiella pneumoniae*. The authors in [22] obtained three datasets from three different hospitals in China to automatically detect bacterial vaginosis from gram-stained microscopic images. Several data augmentation methods like scale jittering and image flipping were used to reduce sampling bias. A total of 29,095 images were used and divided into three smaller datasets. All images were resized to 1024x768. Since the datasets used by [22] and [50] were provided by hospitals, they are categorized as industrial datasets. Another study used an open-source dataset of 15,090 microscopic images to differentiate longitudinal bacterial divisions (Fission) from horizontal and other divisions [38]. This dataset was also used by [56] and [57]. In these studies, the dataset was manually labelled and divided into two types of classes, 'longitudinal' and 'other', to classify bacterial division. Apart from labelling, a number of pre-processing steps were performed on the open-source dataset, and images were resized to 128x128. The authors in [23] used the open-source dataset 16SrRNA for classifying bacterial sequences into a taxonomy. The authors concluded that performance can be enhanced by increasing the size of dataset, i.e., by including more samples in datasets. The authors in [37] addressed the tedious task of labelling datasets by labelling images with a semi-automatic technique. In this technique, images are segmented to identify the part of the image where bacteria are present from the background image, and that part is used as input for the model. A number of image-processing techniques such as noising and erosion were used to obtain bacteria segments. The dataset was augmented by applying filters, zooming, and flipping to increase its size. Models were trained using input images of two sizes. A total of 2,772 images were used for training and testing; these images are available for download. An industrial dataset of microscopic images was used to train the models. Images were resized to 100x100x3.

Fig. 14 presents the different data samples and their frequencies. Many studies have used microscopic data. Many have also used spectroscopic data samples in their datasets.

In [51], the authors used two academic datasets composed of 695 fluorescence, bright-field, and phase contrast images to train models for bacterial cells detection and classification in complex multi-cellular environments.

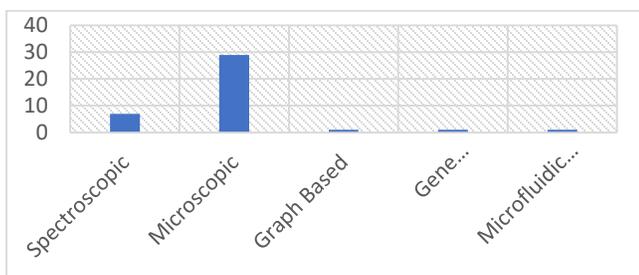


Fig. 14. Types of Data Samples.

In [52], the authors developed different datasets in order to segment three types of bacteria. Since the datasets were developed and labelled in laboratory settings, they are categorized as academic datasets. Datasets contain different types of samples such as bright field, wide field, and fluorescence images. Datasets were augmented to increase the size. These datasets are available for download. The authors in [41] prepared an academic dataset composed of fluorescence microscopic images to train and test deep learning models. The authors processed the images and created two types of transitional images for object localization and boundary detection. The augmented dataset contains 733 images. In [32], the authors detected bacterial cells from the 3D fluorescent microscopic images of an academic dataset. The authors classify the detected cells into two classes: bacteria and non-bacteria. The dataset was prepared by using 3D images of Zebrafish intestine. The image size is 10x30x30. The authors in [24] applied a number of pre-processing steps to their datasets. For instance, a magnetic field was used to convert 3D signals into 2D data. Each 5000x3000 image was divided into one hundred 500x300 images. Two datasets were used for training and verification, including an academic and an open-source dataset named 'VOC 2007'.

The authors in [39] used an academic dataset composed of SEM images. Three datasets created in a laboratory environment were used for training and testing: the first contained two types of bacteria; the second contained two different types of bacteria; and the third contained six types of bacteria, including those present in the first and second dataset. The third dataset was considered challenging because bacteria present in this dataset resemble each other in shape. Images were resized to 400x400 before being fed to UNet architecture. The authors in [58] used an open-source dataset composed of spectral data. This dataset was pre-processed and augmented before it was fed to the deep learning model. In [26], a 3000 Raman spectra from an industrial dataset was used for identification of UTI-causing bacteria. This dataset was divided for training and testing purposes. In [45], the authors used 33,951 unlabelled spectroscopic data from an academic dataset to train and test the model, which was able to differentiate between two types of bacteria: MSRA and MSSA. The authors in [46] also used spectral data from an academic dataset to classify bacteria into eight classes. In [28], an industrial dataset obtained from a hospital was used. This dataset contains 1,000 spectra from 25 different bacterial species. Data were randomly divided for training and testing purposes in 9:1.

The authors in [44] used a pre-trained Inception v3 model to identify the presence and absence of filamentous bacteria in waste water. An industrial dataset of 13,860 images was prepared in a laboratory setting by obtaining waste water samples from eight different waste water plants in Japan. In [31], the authors prepared an extensive dataset by collecting water samples from 1,301 locations. This dataset was pre-processed, and images were resized to 3228x3215.

In [34], pre-processing steps were performed to segment bacteria from the background of Whole Slide Images into three bacterial classes. Three types of industrial datasets were used to train models. A labelled dataset was used for segmentation whereas an unlabelled dataset was fed to the fine-tuned CNN

classifier. An academic dataset was used by [53], where training data comprised of around 11,000 wide-field microscopic images and around 9,000 SIM images. In each case, 10% of the images were set aside randomly and used for a test set.

One study made use of 6,900 SEM images of size 1280x960 as an academic dataset [30]. A sliding window of size 40x40 was used to scan them. In [33], the authors used SEM images as data samples for identification of structural features of G20 bacteria. An academic dataset with 66 images and two classes was divided into validation, testing, and testing data. The authors in [42] used a dataset of 290 SEM images. The data were pre-processed to remove any noise and resized to 512x512. In [27], an open-source dataset called 2DBEST composed of 66 SEM images was used to train the DCNN model. This dataset contains images of size 229x256.

In [54], the authors differentiate between different species of gram-positive bacteria through Hyper-spectral Microscopic Images of an industrial dataset. Bacteria were segmented from a 512x512 image background with the help of a binarization image-processing technique. The authors in [47] made use of a pre-processed industrial dataset of Hyper Spectral Microscopic Images to classify food-borne bacteria into five classes.

RQ 2.2 What performance evaluation metrics do the studies use to evaluate the performance of deep-learning-based techniques for the detection and classification of bacterial colonies?

Performance evaluation of bacteria detection and classification techniques involves well-known evaluation metrics. Metrics such as accuracy, F1 score, precision, and confusion matrix are commonly used to compare the performance of a newly proposed technique with previously available techniques. Apart from these metrics, criteria such as computation complexity, cost, size of dataset, and level of pre-processing activities are also used to evaluate the performance

of deep learning classifiers and extracted features. Deep learning models are inherently complex in nature. For instance, the number of layers in a CNN may increase overall computational cost and complexity.

$$Accuracy = (TP + TN)/(TP + FP + TN + FN) \quad (1)$$

$$Precision = TP/(TP + FP) \quad (2)$$

$$Recall/Sensitivity = TP/(TP + FN) \quad (3)$$

$$Specificity = TN/(TN + FP) \quad (4)$$

$$F1\ Score = (2 * Sensitivity * Precision)/(Sensitivity + Precision) \quad (5)$$

$$Dice\ Index = (2(A \cap B))/(A + B) \quad (6)$$

Equations 1–6 can calculate accuracy, precision, sensitivity, and F1-score. Here ‘TP’ stands for ‘true positive’, or the correct identification of bacteria presence. ‘TN’ stands for ‘true negative’, indicating correct identification of absent bacteria. ‘FP’ represents ‘false positive’, or the false presence of bacteria as absent, whereas ‘FN’ stands for ‘false negative’, or the incorrect identification of absent bacteria as present. Accuracy is the sum of correct identifications (both present and absent) divided by total samples. Precision is the fraction of correctly identified positive instances over all positive instances. Sensitivity is the proportion of correctly identified positive instances out of all positive instances. Specificity is calculated as correctly identified negative instances over all negative instances. The receiver operating curve (ROC) presents the true positive rate against the true negative rate and is also known as the precision-recall rate. F1-score is a statistical measure that is calculated as the geometric mean of specificity and sensitivity. Confusion matrix represents the rate of misclassified bacterial instances. Rows in the confusion matrix represent actual classes while columns represent predicted classes. The evaluation metrics used by the studies included in this review are presented in Table VII.

TABLE VII. EVALUATION METRICS USED IN PRIMARY STUDIES

Reference	Highest Accuracy	Precision	Recall	Sensitivity	Specificity	FP Rate	Confusion Matrix	F1-Score	AUC/ROC
(Zhang et al., 2021b)	100%						✓		
[21]	99.24%				✓	✓	✓		
[50]	65%				✓	✓	✓		
[22]	89.3			✓	✓				✓
[38]	99.6	✓	✓					✓	
[23]	91.7	✓	✓			✓		✓	✓
[51]	76%								
[51]	95.8%		✓						
[52]	98%	✓	✓	✓	✓				
[44]		✓	✓	✓	✓				
[24]	86%	✓							
[33]	81%	✓	✓					✓	
[53]	86%						✓		
[26]	96%						✓		

[40]	96%								
[25]	97%								
[27]	74%								
[45]	97.66%			✓	✓		✓		✓
[28]	100%						✓		
[43]		✓	✓					✓	
[29]	94.8%	✓	✓					✓	
[34]	81%	✓	✓					✓	✓
[35]	97%	✓	✓				✓	✓	
[36]	96.8%						✓		
[48]	95%	✓						✓	
[55]	96.2%	✓		✓	✓		✓	✓	
[47]	92.9%						✓		
[49]	✓							✓	
[46]	94%								
[30]	93%						✓		
[54]	98%			✓	✓				
[41]	90%							✓	
[42]	93%	✓	✓					✓	
[31]	97%	✓		✓			✓	✓	
[32]	95%								
[58]	92%								
[37]	100%	✓	✓					✓	

## V. DISCUSSION AND IMPLICATIONS

This secondary study, an SLR, has reviewed a total of 38 primary studies. This section discusses the results and their implications.

Most of the articles reviewed focus on supervised learning techniques. Only a few papers discuss unsupervised learning techniques. More contributions to semi-supervised and unsupervised learning techniques are needed. Overall, 77% of the primary studies focus on supervised learning, 18% focus on unsupervised learning, and only 5% focus on semi-supervised learning techniques. In short, semi-supervised learning is a potential area that can be further explored for bacterial colony detection and classification. This implication is noteworthy for researchers who want to contribute in the area of deep-learning-based identification and classification of bacterial colonies. Within supervised learning, the majority of studies have focused on CNNs and their different architectures. Around 86% of the supervised learning techniques focus on different CNN architectures. ResNet (48%) and UNet (20%) dominate as compared to other CNN architectures such as SqueezeNet and AlexNet. There is a need to investigate the evidence regarding how to select a particular CNN architecture according to type and size of dataset. Such research may enable new researchers and practitioners to select the best deep learning model according to the size and type of their data.

Development of tools plays an important role in applying academic research to industrial practices. Lack of tools can be a great barrier when it comes to adopting academic research into industrial practices. Only a few tools are available publicly. The majority of the primary studies did not develop end-to-end

tools, with some providing only implementation details and others mentioning no implementation details at all. Most deep learning methods are computationally expensive, with optimization demanding an extremely large number of parameters and with memory constraints that necessitate simultaneous use of Graphics processing unit (GPUs). There is a need for stand-alone tools that can operate without technological constraints such as memory and that are accessible even to beginners or practitioners who might not possess technical understanding of architectural details.

Datasets are the fundamental entities that determine performance in deep learning methods. It was observed that 40% percent of the primary studies used academic datasets. Thus most data samples were constructed in laboratory settings. While 34% of the primary studies employed open-source datasets, only 26% used industrial datasets. Datasets created in laboratory settings under specific conditions always have an inherent bias, which is a threat to validity of results. Future research must incorporate more industrial datasets so that academic research can solve real industrial problems. Furthermore, a large number of benchmark open-source datasets must be readily available to researchers so that performance of different methods can become comparable. Hence, creating and updating of open-source datasets also needs researchers' attention.

A number of studies performed data augmentation to increase the size of datasets and reduce the bias of sampling. Images were augmented in a number of ways. For instance, the authors in [22] performed jittering and horizontal and vertical flips. Yet there is little discussion on the cost of augmentation and comparison of performance of deep learning models with

and without augmentation. It would be interesting to conduct such empirical studies and compare the cost and accuracies.

Performance evaluation of the primary studies was conducted using well-known evaluation metrics such as precision, recall, FP rate, AUC/ROC, and F1-score. Accuracy, precision, F1-score, confusion matrix, and recall were most frequently used for performance evaluation.

This study can benefit researchers and practitioners by providing them a bird's eye view as well as an in-depth analysis of existing research in the area of deep-learning-based identification and classification of bacterial colonies. Researchers can benefit from the classification of existing studies on the taxonomy of deep learning models. This study helps to identify where primary studies are lacking so that researchers can target and contribute to those areas. This study also benefits practitioners by outlining recent developments in academic research so that they can adopt those developments in the industry with confidence and evidence.

## VI. CONCLUSION AND FUTURE DIRECTIONS

This study analysed 38 articles and identified trends in most commonly used deep learning techniques, commonly used datasets, and availability of tools. The study has presented a taxonomy of deep learning techniques and mapped existing primary studies to identify the gaps in the literature. Furthermore, a thematic and descriptive analysis was conducted on qualitative and quantitative data respectively to answer the research questions and provide insights into deep learning approaches. The study reports on benchmark datasets used by deep learning approaches for bacterial colony classification. This study also presents a comparative analysis to find the similarities and differences in performance evaluation metrics used in primary studies. The results indicate that most of the articles focus on supervised learning techniques. Within supervised learning, the majority of the articles focus on CNN. Only a few used unsupervised learning techniques. Development of tools plays an important role in applying academic research to industrial practices, yet only a limited number of tools are publicly available. The results also indicate that a majority of the primary studies use academic datasets. Furthermore, accuracy, precision, F1-score, confusion matrix, and recall were the most frequently used performance evaluation metrics. This study is beneficial for researchers as it helps to identify areas where they can contribute.

In the future, more contributions towards semi-supervised and unsupervised learning techniques are needed. Future research works must incorporate more industrial datasets so that academic research can solve real industrial problems. Furthermore, there is a need for stand-alone tools that can operate without technological constraints so that beginners and practitioners can use them.

## REFERENCES

- [1] Wassenaar TM. Bacteria: the benign, the bad, and the beautiful. John Wiley & Sons; 2011 Nov 3.
- [2] Joy, Chethna, G. Naveen Sundar, and D. Narmadha. "AI Driven Automatic Detection of Bacterial Contamination in Water: A Review." In 2021 5th International Conference on Intelligent Computing and Control Systems (ICICCS), pp. 1281-1285. IEEE, 2021.
- [3] Kc, K., Yin, Z., Wu, M., and Wu, Z.: 'Evaluation of deep learning-based approaches for COVID-19 classification based on chest X-ray images', Signal, Image and Video Processing, 2021, 15, (5), pp. 959-966.
- [4] Du, Y., Han, D., Liu, S., Sun, X., Ning, B., Han, T., Wang, J., and Gao, Z.: 'Raman spectroscopy-based adversarial network combined with SVM for detection of foodborne pathogenic bacteria', Talanta, 2022, 237, pp. 122901.
- [5] Zieliński, B., Plichta, A., Misztal, K., Spurek, P., Brzywczy-Włoch, M., and Ochońska, D.: 'Deep learning approach to bacterial colony classification', PloS one, 2017, 12, (9), pp. e0184554.
- [6] Mahmud, M., Kaiser, M.S., McGinnity, T.M., and Hussain, A.: 'Deep learning in mining biological data', Cognitive Computation, 2021, 13, (1), pp. 1-33.
- [7] Xiao, Y., and Watson, M.: 'Guidance on conducting a systematic literature review', Journal of Planning Education and Research, 2019, 39, (1), pp. 93-112.
- [8] Berman, D.S., Buczak, A.L., Chavis, J.S., and Corbett, C.L.: 'A survey of deep learning methods for cyber security', Information, 2019, 10, (4), pp. 122.
- [9] Shvets, Alexey A., Alexander Rakhlin, Alexandr A. Kalinin, and Vladimir I. Iglovikov. "Automatic instrument segmentation in robot-assisted surgery using deep learning." In 2018 17th IEEE international conference on machine learning and applications (ICMLA), pp. 624-628. IEEE, 2018.
- [10] Young, T., Hazarika, D., Poria, S., and Cambria, E.: 'Recent trends in deep learning based natural language processing', IEEE Computational Intelligence Magazine, 2018, 13, (3), pp. 55-75.
- [11] Alom, M.Z., Taha, T.M., Yakopcic, C., Westberg, S., Sidike, P., Nasrin, M.S., Hasan, M., Van Essen, B.C., Awwal, A.A., and Asari, V.K.: 'A state-of-the-art survey on deep learning theory and architectures', Electronics, 2019, 8, (3), pp. 292.
- [12] Valpola, H.: 'From neural PCA to deep unsupervised learning', in 'Book Advances in independent component analysis and learning machines' (Elsevier, 2015), pp. 143-171.
- [13] Subramanian, N., Elharrouss, O., Al-Maadeed, S., and Chowdhury, M.: 'A review of deep learning-based detection methods for COVID-19', Computers in Biology and Medicine, 2022, pp. 105233.
- [14] Socher, Richard, Cliff C. Lin, Chris Manning, and Andrew Y. Ng. "Parsing natural scenes and natural language with recursive neural networks." In Proceedings of the 28th international conference on machine learning (ICML-11), pp. 129-136. 2011.
- [15] Perkins, D.N., and Salomon, G.: 'Transfer of learning', International Encyclopedia of Education, 1992, 2, pp. 6452-6457.
- [16] Petersen, K., Feldt, R., Mujtaba, S., and Mattsson, M.: 'Systematic mapping studies in software engineering', in Editor (Ed.) (Eds.): 'Book Systematic mapping studies in software engineering' (2008, edn.), pp. 1-10.
- [17] Kitchenham, B., Brereton, O.P., Budgen, D., Turner, M., Bailey, J., and Linkman, S.: 'Systematic literature reviews in software engineering—a systematic literature review', Information and Software Technology, 2009, 51, (1), pp. 7-15.
- [18] Kumar, S., Kar, A.K., and Ilavarasan, P.V.: 'Applications of text mining in services management: A systematic literature review', International Journal of Information Management Data Insights, 2021, 1, (1), pp. 100008.
- [19] Imtiaz, J., Sherin, S., Khan, M.U., and Iqbal, M.Z.: 'A systematic literature review of test breakage prevention and repair techniques', Information and Software Technology, 2019, 113, pp. 1-19.
- [20] Zhang, S., Han, Z., Feng, Z., Sun, M., and Duan, X.: 'Deep learning assisted microfluidic impedance flow cytometry for label-free foodborne bacteria analysis and classification', in Editor (Ed.) (Eds.): 'Book Deep learning assisted microfluidic impedance flow cytometry for label-free foodborne bacteria analysis and classification' (IEEE, 2021, edn.), pp. 7087-7090.
- [21] Akbar, S.A., Ghazali, K.H., Hasan, H., Mohamed, Z., and Aji, W.S.: 'An enhanced classification of bacteria pathogen on microscopy images using deep learning', in Editor (Ed.) (Eds.): 'Book An enhanced classification of bacteria pathogen on microscopy images using deep learning' (IEEE, 2021, edn.), pp. 119-123.

- [22] Wang, Z., Zhang, L., Zhao, M., Wang, Y., Bai, H., Wang, Y., Rui, C., Fan, C., Li, J., and Li, N.: 'Deep neural networks offer morphologic classification and diagnosis of bacterial vaginosis', *Journal of Clinical Microbiology*, 2021, 59, (2), pp. e02236-02220.
- [23] Helaly, M.A., Rady, S., and Aref, M.M.: 'Deep learning for taxonomic classification of biological bacterial sequences': 'Book Machine learning and big data analytics paradigms: Analysis, applications and challenges' (Springer, 2021), pp. 393-413.
- [24] Hu, Q., Wang, S., Duan, H., and Liu, Y.: 'A fluorescent biosensor for sensitive detection of salmonella typhimurium using low-gradient magnetic field and deep learning via faster region-based convolutional neural network', *Biosensors*, 2021, 11, (11), pp. 447.
- [25] Mai, D.-T., and Ishibashi, K.: 'Bacteria shape classification using small-scale depthwise separable CNNs', in Editor (Ed.) (Eds.): 'Book Bacteria shape classification using small-scale depthwise separable CNNs' (IEEE, 2021, edn.), pp. 2940-2943.
- [26] Fu, Q., Zhang, Y., Wang, P., Pi, J., Qiu, X., Guo, Z., Huang, Y., Zhao, Y., Li, S., and Xu, J.: 'Rapid identification of the resistance of urinary tract pathogenic bacteria using deep learning-based spectroscopic analysis', *Analytical and Bioanalytical Chemistry*, 2021, 413, (30), pp. 7401-7410.
- [27] Rahman, M.H., Duckworth, J., Ragi, S., Chundi, P., Gadhamshetty, V.R., and Chilkoor, G.: 'Deep learning approach to extract geometric features of bacterial cells in biofilms': 'Book Advances in data science and information engineering' (Springer, 2021), pp. 359-368.
- [28] Wang, S., Dong, H., Shen, W., Yang, Y., Li, Z., Liu, Y., Wang, C., Gu, B., and Zhang, L.: 'Rapid SERS identification of methicillin-susceptible and methicillin-resistant *Staphylococcus aureus* via aptamer recognition and deep learning', *RSC Advances*, 2021, 11, (55), pp. 34425-34431.
- [29] Patel, S.: 'Bacterial colony classification using atrous convolution with transfer learning', *Annals of the Romanian Society for Cell Biology*, 2021, pp. 1428-1441.
- [30] Wong, P.-C., Fan, T.-E., Lee, Y.-L., Lai, C.-Y., Wu, J.-L., Chang, L.-H., and Su, T.-Y.: 'Detection and identification of the stages of DH5-Alpha *Escherichia coli* biofilm formation on metal by using an artificial intelligence system', *Microscopy and Microanalysis*, 2021, 27, (5), pp. 1218-1225.
- [31] Mohammad Khan, F., Gupta, R., and Sekhri, S.: 'Automated bacteria colony counting on agar plates using machine learning', *Journal of Environmental Engineering*, 2021, 147, (12), pp. 04021066.
- [32] Mhathesh, T., Andrew, J., Martin Sagayam, K., and Henesey, L.: 'A 3D convolutional neural network for bacterial image classification': 'Book Intelligence in big data technologies—Beyond the hype' (Springer, 2021), pp. 419-431.
- [33] Ragi, Shankarachary, Md Hafizur Rahman, Jamison Duckworth, Jawaharraj Kalimuthu, Parvathi Chundi, and Venkataramana Gadhamshetty. "Artificial intelligence-driven image analysis of bacterial cells and biofilms." *IEEE/ACM Transactions on Computational Biology and Bioinformatics* (2021).
- [34] Alhammad, Sarah, Kun Zhao, Anthony Jennings, Peter Hobson, Daniel F. Smith, Brett Baker, Justin Staweno, and Brian C. Lovell. "Efficient DNN-Based Classification of Whole Slide Gram Stain Images for Microbiology." In 2021 Digital Image Computing: Techniques and Applications (DICTA), pp. 01-08. IEEE, 2021.
- [35] Shwetha, V., Keerthana Prasad, Chiranjoy Mukhopadhyay, Barnini Banerjee, and Abir Chakrabarti. "Automatic Detection of Bacilli Bacteria from Ziehl-Neelsen Sputum Smear Images." In 2021 2nd International Conference on Communication, Computing and Industry 4.0 (C2I4), pp. 1-5. IEEE, 2021.
- [36] Jha, Nirajan, and Dibakar Raj Pant. "Stochastic Gradient Descent and Discriminative Fine Tuning on ResNet, DenseNet, Inception-ResNet and MobileNet for the Multi Class Pathogenic Microbes Classification." (2021).
- [37] Casado-Garcia, A., Chichón, G., Domínguez, C., García-Domínguez, M., Heras, J., Ines, A., López, M., Mata, E., Pascual, V., and Saenz, Y.: 'MotilityJ: An open-source tool for the classification and segmentation of bacteria on motility images', *Computers in Biology and Medicine*, 2021, 136, pp. 104673.
- [38] Garcia-Perez, C., Ito, K., Geijo, J., Feldbauer, R., Schreiber, N., and Zu Castell, W.: 'Efficient detection of longitudinal bacteria fission using transfer learning in deep neural networks', *Frontiers in Microbiology*, 2021, 12.
- [39] Kakishita, Y., Ray, A., Hattori, H., Hisada, A., Ominami, Y., Baudoin, J.-P., Khalil, J.Y.B., and Raoult, D.: 'Quantitative analysis system for bacterial cells in SEM image using deep learning', in Editor (Ed.) (Eds.): 'Book Quantitative analysis system for bacterial cells in SEM image using deep learning' (IEEE, 2021, edn.), pp. 01-06.
- [40] Sundar, G.N.: 'Effective deep learning model to identify harmful bacteria in blood samples for healthcare application', *Turkish Journal of Computer and Mathematics Education (TURCOMAT)*, 2021, 12, (6), pp. 3475-3487.
- [41] Zhang, J., Wang, Y., Donarski, E., and Gahlmann, A.: 'BCM3D 2.0: Accurate segmentation of single bacterial cells in dense biofilms using computationally generated intermediate image representations', *bioRxiv*, 2021.
- [42] Abeyrathna, Dilanga, Terrance Life, Shailabh Rauniyar, Shankarachary Ragi, Rajesh Sani, and Parvathi Chundi. "Segmentation of Bacterial Cells in Biofilms Using an Overlapped Ellipse Fitting Technique." In 2021 IEEE International Conference on Bioinformatics and Biomedicine (BIBM), pp. 3548-3554. IEEE, 2021.
- [43] García, R.G., Rodríguez, S.J., Martínez, B.B., Gracidas, C.H., and Torres, R.M.: 'Efficient deep learning architectures for fast identification of bacterial strains in resource-constrained devices', *arXiv preprint arXiv:2106.06505*, 2021.
- [44] Satoh, H., Kashimoto, Y., Takahashi, N., and Tsujimura, T.: 'Deep learning-based morphology classification of activated sludge flocs in wastewater treatment plants', *Environmental Science: Water Research & Technology*, 2021, 7, (2), pp. 298-305.
- [45] Ciloglu, F.U., Caliskan, A., Sarıdag, A.M., Kilic, I.H., Tokmakci, M., Kahraman, M., and Aydin, O.: 'Drug-resistant *Staphylococcus aureus* bacteria detection by combining surface-enhanced Raman spectroscopy (SERS) and deep learning techniques', *Scientific Reports*, 2021, 11, (1), pp. 1-12.
- [46] Yu, S., Li, X., Lu, W., Li, H., Fu, Y.V., and Liu, F.: 'Analysis of Raman spectra by using deep learning methods in the identification of marine pathogens', *Analytical Chemistry*, 2021, 93, (32), pp. 11089-11098.
- [47] Kang, R., Park, B., Ouyang, Q., and Ren, N.: 'Rapid identification of foodborne bacteria with hyperspectral microscopic imaging and artificial intelligence classification algorithms', *Food Control*, 2021, 130, pp. 108379.
- [48] Akshay, S. "Classification of Beneficial and non-Beneficial Bacteria using Random Forest Algorithm." In 2021 Third International Conference on Inventive Research in Computing Applications (ICIRCA), pp. 1-9. IEEE, 2021.
- [49] Wang, J., Zhang, M., Zhang, J., Wang, Y., Gahlmann, A., and Acton, S.T.: 'Graph-theoretic post-processing of segmentation with application to dense biofilms', *IEEE Transactions on Image Processing*, 2021, 30, pp. 8580-8594.
- [50] Borowa, Adriana, Dawid Rymarczyk, Dorota Ochońska, Monika Brzywczy-Włoch, and Bartosz Zieliński. "Deep learning classification of bacteria clones explained by persistence homology." In 2021 International Joint Conference on Neural Networks (IJCNN), pp. 1-8. IEEE, 2021.
- [51] Panigrahi, S., Murat, D., Le Gall, A., Martineau, E., Goldlust, K., Fiche, J.-B., Rombouts, S., Nölmann, M., Espinosa, L., and Mignot, T.: 'Mistic, a general deep learning-based method for the high-throughput cell segmentation of complex bacterial communities', *Elife*, 2021, 10, pp. e65151.
- [52] Spahn, C., Laine, R.F., Pereira, P.M., Gómez-de-Mariscal, E., von Chamier, L., Conduit, M., de Pinho, M.G., Jacquemet, G., Holden, S., and Heilemann, M.: 'DeepBacs: Bacterial image analysis using open-source deep learning approaches', *bioRxiv*, 2021.
- [53] Saraiva, B.M., Krippahl, L., Filipe, S.R., Henriques, R., and Pinho, M.G.: 'eHooke: A tool for automated image analysis of spherical bacteria based on cell cycle progression', *Biological imaging*, 2021, 1.
- [54] Liu, K., Ke, Z., Chen, P., Zhu, S., Yin, H., Li, Z., and Chen, Z.: 'Classification of two species of Gram-positive bacteria through hyperspectral microscopy coupled with machine learning', *Biomedical Optics Express*, 2021, 12, (12), pp. 7906-7916.

- [55] Mai, D.-T., and Ishibashi, K.: 'Small-scale depthwise separable convolutional neural networks for bacteria classification', *Electronics*, 2021, 10, (23), pp. 3005.
- [56] Pende, N., Leisch, N., Gruber-Vodicka, H.R., Heindl, N.R., Ott, J., Den Blaauwen, T., and Bulgheresi, S.: 'Size-independent symmetric division in extraordinarily long cells', *Nature Communications*, 2014, 5, (1), pp. 1-10.
- [57] Leisch, N., Pende, N., Weber, P.M., Gruber-Vodicka, H.R., Verheul, J., Vischer, N.O., Abby, S.S., Geier, B., Den Blaauwen, T., and Bulgheresi, S.: 'Asynchronous division by non-ring FtsZ in the gammaproteobacterial symbiont of *Robbea hypermnestra*', *Nature Microbiology*, 2016, 2, (1), pp. 1-5.
- [58] Tafintseva, V., Shapaval, V., Blazhko, U., and Kohler, A.: 'Correcting replicate variation in spectroscopic data by machine learning and model-based pre-processing', *Chemometrics and Intelligent Laboratory Systems*, 2021, 215, pp. 104350.

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