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MACHINE LEARNING AND DEEP LEARNING METHODS FOR PATHOGEN IDENTIFICATION AND CLASSIFICATION OF PATHOGENS

Asma Yousafzai^{1†}, Ayesha Nawaz^{2†}, Ali Hamza², Muhammad Sadiq³, Nousheen Arshad⁴, Muhammad Ali², Habiba Yasin², Akkasha Baig², Awais Ashraf², Eman Gulzar², Mehak Imran², Sabra Anwar⁵, Nimra Ali^{6*}, Amina Sajjad⁶

¹**Department of Biotechnology**, Balochistan University of Information Technology, Engineering and Management Sciences (BUIITEMS), Quetta, Pakistan

²**Department of Microbiology**, Faculty of Science and Technology, University of Central Punjab, Lahore, Pakistan

³**Principal Medical Officer**, CENAR Cancer Hospital, Quetta, Pakistan

⁴**Ripha College of Veterinary Sciences**, , Ripha International University, Lahore, Pakistan

⁵**Department of Biotechnology**, Faculty of Science and Technology, University of Central Punjab Lahore, Pakistan

⁶**Department of Basic and Applied Chemistry**, Faculty of Science and Technology, University of Central Punjab Lahore, Pakistan

***Corresponding Author:** Nimra Ali. E. mail: hnimra426@gmail.com

† Contributed Equally: Asma Yousafzai and Ayesha Nawaz

Abstract

Machine learning (ML) and deep learning (DL) have become powerful tools in medical sciences, offering rapid, low-cost, and accurate pathogen detection compared to conventional methods. A variety of algorithms, including SVM, NB, RF, and k-NNC, have been widely applied in microbiology for bacterial identification, genetic classification, and image analysis. Beyond bacteria, ML/DL approaches have improved detection of protozoan pathogens by recognizing different life-cycle stages and host pathogen interactions, while also advancing early viral diagnosis, particularly during the COVID-19 pandemic. Foodborne pathogen surveillance has further benefited from integration of ML with imaging techniques such as hyperspectral analysis. Despite these advances, challenges remain, including data bias, high computational requirements, and the need for better generalization across diverse datasets. This review summarizes current applications of ML/DL in pathogen identification, highlights recent progress, and discusses limitations and future opportunities for improving diagnostic accuracy and disease management.

Keywords: Bacteria, Biomedicine, Machine learning, Pathogen, Viruses

INTRODUCTION

Machine learning is an important part of artificial intelligence and plays an important role in the domain of bioinformatics. Machine learning is useful in obtaining meaningful information from complex datasets (1, 2). Machine learning involves the relationship between mathematics and computer sciences which makes it applicable in the fields of biomedicine, microbiology and phytology. Machine learning has also created paths from deep theoretical research into practical applications (3). The practical applications of machine learning cover biological image analysis, guessing the disease and diagnostic microbiology (4). Problems created by microbial infections can be treated by advancements in Artificial Intelligence technologies in the medical field (5). Various causative agents result infectious diseases such bacteria, fungi, viruses and protozoans (6). In the recent era machine learning has wide applications in microbiology such as predicting drug and vaccine candidates, tracking disease outbreaks, exploring microbial interactions, and detecting pathogens (7).

In machine learning input data can be of various types including numerical data, categorical data, time-series data, and information in the form of texts (8). Data preprocessing must be performed before data training in machine learning to ensure the reliability of output data. Data preprocessing depends upon the problem of interest (9). Biological data such as DNA, RNA contains a lot of irrelevant and unnecessary data so feature selection and feature extraction are needed to increase the accuracy of machine learning and to



make the results more comprehensive. Feature extraction transfers high dimensional space into lower dimensional space (10). This also improves the pattern recognition capability. In proteins the sequence information is converted into numerical vectors. Feature selection a subset of feature is transferred instead of a complete feature (11). ML tasks can be organized into three types: supervised, unsupervised, and semi-supervised learning. Supervised learning tasks contains labels to detect output of an unlabeled task and uses the data from ground truth while unsupervised learning tasks are without labels and targets but capable of predicting differences and similarities in unlabeled tasks (12). Semi-supervised learning contains both labeled and unlabeled tasks. This methodology participates to distinguish diverse parasitic morphologies and detecting morphological outliers at different lifecycle stages of the malaria parasite (13). There are many different algorithms can be used in different machine learning tasks. In the field of microbiology, the most commonly used algorithms are SVM, NB, RF, and k-NNC. The type of algorithm to be used depends upon the actual problem to be solved and also on the model for the data training (14). The algorithms used in deep learning include neural networks which processes information based on the phenomenon of synaptic connections of brains neurons to transfer information. DL algorithms have some advantages over machine learning including strong learning ability, wide-coverage, and good portability. The disadvantages faced by these algorithms include enormous computing power, high hardware cost, and complex model design (15). Fig.1 illustrates the schematic diagram of the ML approach.

Although several studies have explored individual applications of machine learning in microbiology, there is still a lack of comprehensive reviews that integrate its role across multiple pathogen types, including bacteria, protozoa, viruses, and foodborne contaminants. Most existing reviews focus on a single group of pathogens or a specific application such as imaging or genomic analysis. This review aims to bridge that gap by providing an integrated overview of how both ML and DL approaches are being applied in diverse areas of pathogen detection, highlighting not only recent advances but also the limitations and future opportunities in this evolving field.

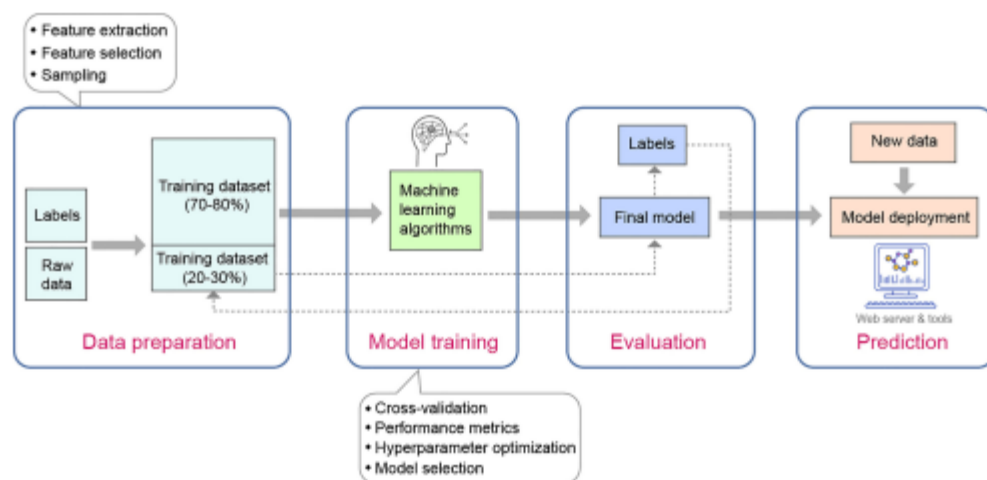


Fig. 1. Machine learning model

APPLICATION OF MACHINE LEARNING IN PATHOGEN DETECTION

Machine learning is increasingly applied for the detection of diverse pathogens, offering faster and more accurate alternatives to traditional diagnostic methods (16). In bacterial studies, algorithms can classify species from images and genetic data, while in protozoan infections such as malaria, ML models recognize different life-cycle stages with high precision. Viral detection has also advanced, with ML improving early diagnosis and supporting outbreak monitoring, as seen during COVID-19. In food safety, combining ML with imaging tools has enhanced the identification of microbial contaminants. These applications highlight the versatility of ML in tackling varied infectious threats, though challenges such as data quality and computational demands still need to be addressed.

BACTERIAL PATHOGENS DETECTION

Bacteria are prokaryotic organisms that lack a nucleus, with their genetic material dispersed in the cytoplasm. Morphologically, they are classified into five basic forms: spherical, rod-shaped, spiral, comma-shaped, and corkscrew (17). While many species are beneficial to humans, contributing to processes such as food production, biotechnology, and pest control, others are pathogenic and responsible for serious diseases including tetanus, typhoid fever, cholera, tuberculosis, and foodborne illnesses (18). Traditional bacterial classification relied on phenotypic characteristics such as shape, size, and color. Additional methods included motility testing, biochemical assays, and molecular techniques (19). Although valuable, these methods were often labor intensive, time consuming, and dependent on skilled operators. They also lacked accuracy when dealing with complex or mixed samples. The emergence of machine learning (ML) has helped to overcome these challenges by providing automated, reliable, and cost effective approaches to bacterial identification.

ML has been successfully applied in diverse biomedical fields, including the classification of medical images such as cancer cells, and its ability to process complex data has paved the way for bacterial classification. These approaches utilize feature extraction and selection techniques to analyze bacterial morphology and genetic information, enhancing precision compared to traditional methods (20). As shown in Fig. 2, a typical ML workflow for bacterial classification includes five phases: image acquisition, preprocessing to remove noise and improve clarity, segmentation to isolate relevant features, extraction and selection of critical parameters, and final classification into categories (21).

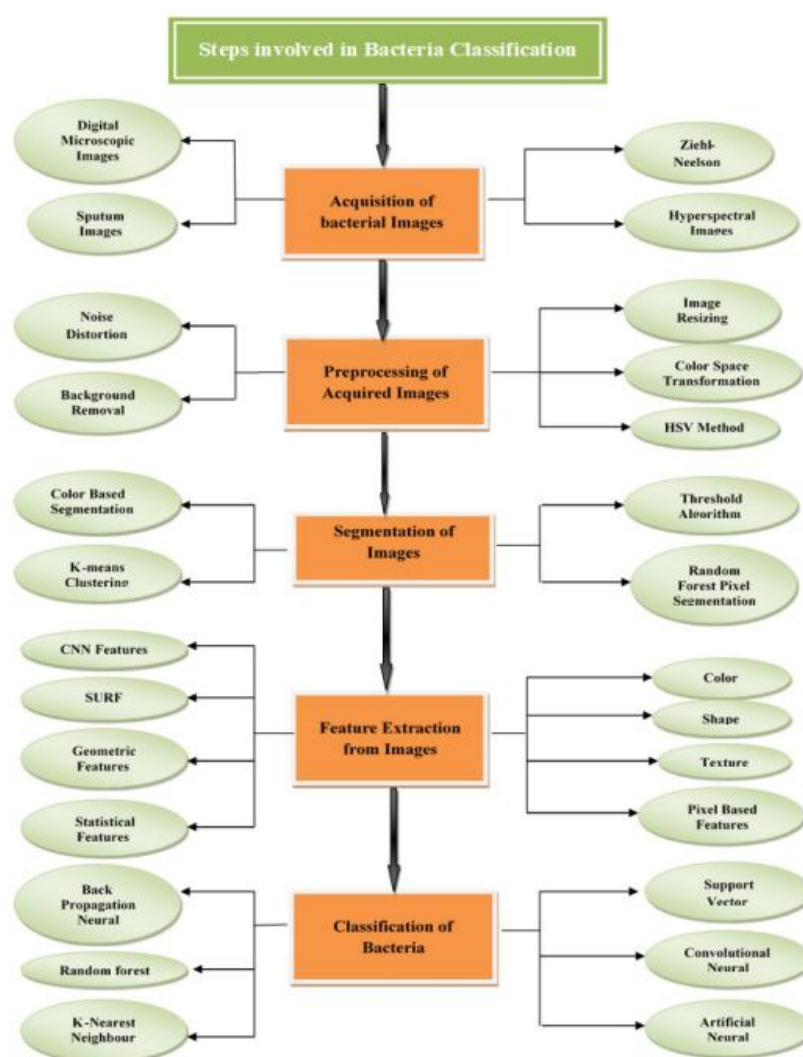


Fig. 2. Bacterial image classification system

By combining morphological and genetic data, ML systems can provide results that are both faster and more accurate than conventional approaches. Feature extraction and selection are particularly

important, as they reduce irrelevant data and highlight meaningful characteristics, enabling robust pattern recognition. Unlike traditional classification methods that primarily rely on observable traits, ML can integrate genetic information, thereby improving classification accuracy and reliability. This is crucial not only for distinguishing pathogenic species but also for identifying beneficial bacteria used in industrial and environmental applications. Several ML approaches for bacterial classification are summarized in Table I.

Several studies have demonstrated the effectiveness of ML in bacterial detection. For instance, research conducted in Sweden analyzed 100 petri plates containing urinary bacteria and extracted 48 distinct features, achieving a classification accuracy of 76%. Other investigations across different institutions and conditions have reported higher performances, with many ML models demonstrating accuracies between 90% and 100%. Among these, the K-means clustering algorithm achieved 100% sensitivity in classifying bacterial species (22). The ability of ML algorithms to achieve such results illustrates their potential to revolutionize diagnostic microbiology. By reducing the need for manual interpretation, these models not only improve efficiency but also minimize errors associated with human bias. With continuous refinement, ML based bacterial classification systems could become a standard component of clinical diagnostics, food safety monitoring, and microbiological research.

Table I. ML approached for bacterial classification

Machine learning techniques	Types of bacteria	Feature selection	Dataset	Results
ANN	Urinary bacteria	Shape features	Petri dish Images	Acc=76%
NN,Back propogation	Tuber-culosis bacilli	Shape features	ZN-stained Sputum smear Images	Acc=97.9% Se=94.1% Sp=99.1%
KNN	Different Bacteria species	Shape features	Digital Images	Acc=97%
K-means Clustering	Tuberculosis bacteria	Shape features	ZN-stained Sputum smear Images	Sp=93.54% Se=100%
SVM	Heterotrophic bacteria colonies	Shape features	Heterotrophic bacteria colony Images	Acc=98.7%
SVM and Radial basis function	Oral cavity bacteria	Color features	Petri Dish Images	Acc=96% Pre=0.97+-0.03 Re=0.96+-0.04

PROTOZOAN DETECTION

PCR and real-time PCR can detect the nucleic acid of parasite and these laboratory methods are often time saving, cost effective and simple to perform (23). Point of care detection of parasites can be appropriately achieved through microscopy methods but sometimes health care workers are needed to analyze large number of parasitic images (24). The best tool for detection of these images is machine learning (Fig. 3). Parasites present different images at different stages of protozoan life cycle. So machine learning can not only detect the presence of microscopic images but also the diverse life cycle stages (25). Different parasitic protozoans detected by machine learning are Toxoplasma, Babesia, Trypanosoma, Cryptosporidium, and Giardia. Plasmodium falciparum, P. vivax, P. ovale, P. malaria and P. Knowlesi are protozoa that can infect humans (26). Plasmodium parasites uses human as vertebrate host and mosquito as vector host and cause Malaria. Amongst the complex life cycle stages of parasites, intraerythrocytic stages cause Malaria (27). The images of these pathogenic microorganisms can be detected efficiently through machine learning. Watershed threshold algorithm used in SVM method of machine learning is highly accurate in determining the life cycle stages of Malaria (28).

Other algorithms for the detection of protozoan pathogens include k-NNC, NB, LDC, LR and CNN (Fig. 4). More than 20 studies have been published that prove CNN as a significant machine learning algorithm for the detection of Malaria (29). One of the studies proposed that 27,588 images were used for the automatic detection of malarial parasites which proved to be 99.96% accurate and 100% precise. DTGCN model was based on DL method and used certain single cell images to detect parasitized and uninfected cells. The classification was performed at ring, trophozoite, schizont, and Gametocyte stages. Machine learning models based on DL methods were combined with smartphones by the use of

microscopic images. This machine learning detection by the use of smartphones proved to be 99% accurate (30). Malaria screener is an android phone application that can detect malaria by the use of certain images scanned through mobile camera. The precision of this detection was 99% on average. Toxoplasma tachyzoites can destroy red blood cells and some other toxoplasmas can infect Brain cells or muscle tissues. Toxoplasma detection is also required to diagnose and treat the disease. Toxoplasmas can be determined through DL algorithm of machine learning. 28,127 single cell images were used in this method and this was more than 93% accurate in both datasets of T400 and T1000 (31). Cryptosporidium oocysts are the protozoa that can cause gastrointestinal diseases and various inflammations. This microorganisms are released through feces into public environment. This infection can be acquired through ingestion of infected water and food contaminated through this oocyte. Cryptosporidium oocysts are fluorescently labeled and detected by ANN algorithm (32).

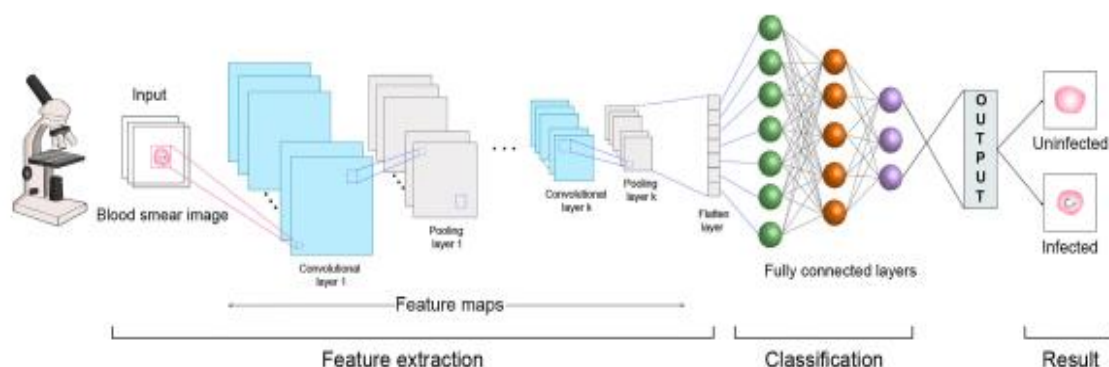


Fig. 3. CNN model for pathogen detection

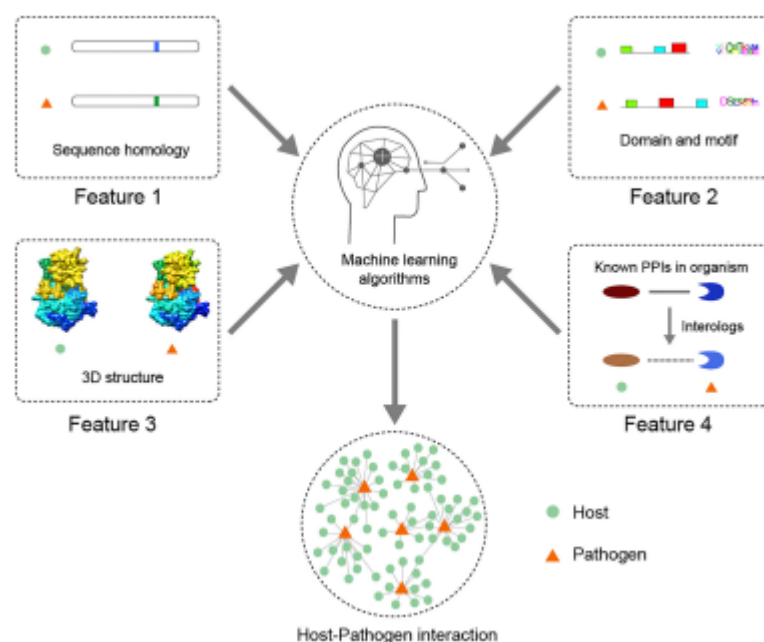


Fig. 4. Host parasite interaction and machine learning

VIRAL DETECTION

Typically, virus can be detected by two methods, one of which is PCR and immunosensing is another method. Polymerase chain reaction basically amplifies the DNA template and then portion containing variants can be analyzed. Real time PCR allows the amplification and analysis at the same time by the use of fluorescent probes. As the virus contains RNA so the genetic material is first converted into DNA and then further PCR process starts. Fluorescence observation is the key to viral detection. This method even though provides highly accurate results but the process is very time taking (37, 38). While in the immune-sensing technique virus is detected through the antigen-antibody interaction. Mostly in district

labs a kit is used for this purpose. In this kit procedure, antibodies are already attached to the cellulose membrane on the kit with which the corresponding viral antigen binds (39). If the viral antigen is present in the sample, then labelled antibodies will produce a color which can be visualized. The intensity of the color produced will indicate the concentration of certain pathogenic virus (40). This requires the skillful experimental handling and is an effective tool in the medical field. But these typical methods can only detect viruses when they have reached a certain stage of severity. Still there was a gap to identify viruses at the earlier stages of infection. Moreover, the characteristics of individual viral particles cannot be detected. To overcome these limitations, nanodevices play a major role that can more closely characterize the virus. These nanodevices use modified nanowires with viral antibodies and technique uses electrical conductance. The drop in electrical conductance indicates the presence of viral antigens. Whenever a virus adheres to the nanowire an electrical signal is generated. With the advancement in this technique, single viral particle can be detected by the use of micro-spherical cavities. The principle of this technique is based on the optical resonance shift which shows the binding of one viral particle to the cavity (41, 42). Machine learning approaches are playing an important role in viral infection identification through different mechanisms as depicted in Fig. 5.

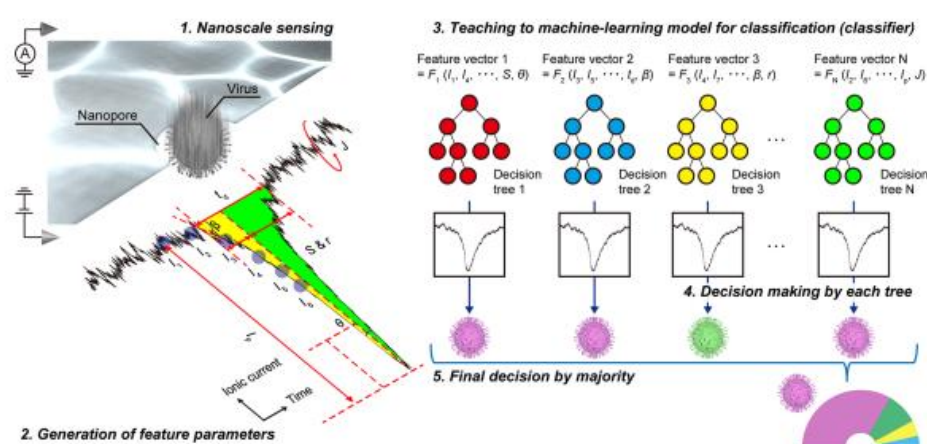


Fig. 5. Machine learning approach for viral infection identification

The first case of covid-19 was reported in 2019 in Wuhan, China. The disease spread its network in 218 countries and affected more than 10.6 million people around the world and almost 500,000 people also lost their important lives. So, it was very important to diagnose and overcome this viral disease as early as possible. Medical field required new technologies to get rid of this crisis because the clinical data was very limited (43). With this limited data, machine learning algorithms can be useful. Machine learning architecture was trained through for the prediction of covid-19 pandemic by using models such as Linear Regression (LR), Multi-Layer Perceptron (MLP), and Vector Auto-Regression (VAR) learning algorithms (44, 45). XGBoost classifier is machine learning based tool for diagnosing the covid-19 because previously used methods were cumbersome for the physicians because of certain limitations (46). This device was trained with certain features such as epidemiological, clinical, demographic, medication, laboratory, and nursing records. These records were electronic and were obtained from more than two thousand patients (47).

The framework of this model analyzed three components to diagnose the risk of covid-19 disease. These key diagnostic components include lactic dehydrogenase (LDH), lymphocyte, and high sensitivity C-reaction protein (hs-CRP). Another model for same purpose was designed by a scientist and the machine learning tool was trained on the basis of vector machine, neural network, gradient boosting trees, random forests, and logistic regression. The machine learning algorithms designed for the sake of diagnostic purposes covered a range of parameters as shown in Fig.6 (48). These parameters are gender, age, hemoglobin, red blood cells, platelets, etc. out of which lymphocytes, eosinophils, and leukocytes were the most important variables. Chest X-ray images by the machine learning methods can distinguish patients of covid-19 from those without the disease that were not affected by the virus (49). Different machine learning

models were used to extract some important features from the x-ray images to diagnose the patients of covid-19. The method was highly accurate with the accuracy of more than 96.09% and 98.09% from the datasets. Medical equipment is not readily available to diagnose the covid-19 patients so as the manpower. Machine learning tools are the best alternatives to deal the major crises by facilitating the diagnostic procedure. Two distinguishing benefits of machine learning models which make them the most useful advancement is the fastest diagnostic process and the procedures are reasonably accurate (50).

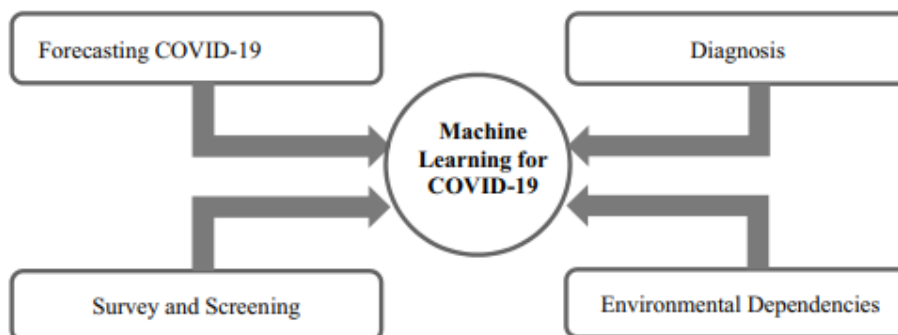


Fig. 6. Machine learning in COVID-19 detection

FOODBORNE PATHOGEN DETECTION

These pathogens can result in food poisoning and also spoil the food products because of which the food products are deprived of their life earlier than the normal (51). These pathogens include bacteria, viruses, fungi, yeast, and protozoa. Spore-forming and non-spore forming strains of bacteria interfere normal human biology and can introduce toxins to food. Some bacteria are capable of food spoilage but are non-pathogenic. These species cause off-odors and can change the texture and taste of food products (52). *Pseudomonas fluorescens*, *Pseudomonas putida*, *Pseudomonas fragi*, and *Pseudomonas perolens* are some of the bacterial species that can spoil food even in the refrigerator (Table II). These pathogens can alter the odor of cheese by adding blue pigment to it and same kind of changes are also caused in the milk. Bacterial pathogens are also involved in meat spoilage by producing gas in the meat and causing discoloration to some extent (53). In addition to bacteria, fungal pathogens are responsible for food spoilage. Some fungal species are beneficial as these are edible and can be used as fermenters. Harmful fungi are those of types of mycotoxins that cause certain cancers, compromised growths, immunosuppressive activities and defects in the neural tube (54). *Aspergillus* and *candida* are the examples of pathogenic fungi that can compromise the immune system of the individuals. These fungi related spoilage of food is due to mishandling of food and inappropriate storage conditions.

Some viruses can also cause food spoilage but these viruses do not directly affect the food products. These viruses make use of human host and then with the mechanism not well-understood yet, virus spoils the food products. Conventionally these food-borne pathogens were detected by growing them on the solid media composed of agar. Bacteria and fungi can be detected by growing those pathogens on the agar and removing the background flora. This detection of pathogens through agar media takes 3 to 7 days and then satisfied results are produced. Food borne bacteria can also be detected by using PCR and real-time PCR, ELISA and biosensor methods. Fungal pathogens can be detected by high profile liquid chromatography (55). Recently, a tool called electronic nose is also used to detect fungi in the food. Viruses can be detected by the use of real-time PCR and other amplification methods. HYPERSPECTRAL IMAGING is the detection method to detect contaminants in the food. This technique scans different areas of interest to detect food pathogens. This technique can classify spore-forming and non-spore forming bacteria and some physical contaminants in the food. Some of the challenges faced by hyperspectral imaging is background interference and limits of detection (56). Other challenges include food matrix is of heterogenous nature, diverse number of populations in the sample etc. data can be extracted from hyperspectral imaging to train the machine learning tool. All three models of machine learning can be combined with hyperspectral

imaging to detect food borne pathogens. Machine learning in hyperspectral imaging can reduce the background noise and increase the efficiency of food borne contaminants detection methods.

Table II. Common foodborne pathogens and contaminations caused by them

Microbial Strains	Food safety or related concerns	Food items reported to be contaminated
<i>Campylobacter jejuni</i>	Diarrhea (often bloody), fever, stomach cramps, irritable bowel syndrome, temporary paralysis, and arthritis in immunocompromised individual	Undercooked or raw chicken, beef, and pork, unpasteurized milk
<i>Listeria monocytogenes</i>	Listeriosis is often associated with septicemia, meningitis, gastroenteritis, pneumonia, and spontaneous abortion with a mortality rate of 20–30% in immunocompromised individuals	Vegetables, milk, and raw refrigerated meat and poultry products
<i>Aspergillus flavus</i> , <i>Aspergillus parasiticus</i> , and <i>Aspergillus nomius</i> (Aflatoxin M1)	Carcinogenic, hepatotoxic, teratogenic, and mutagenic effects	Cereals-maize and rice
<i>Aspergillus flavus</i> (Aflatoxin A1)	Carcinogenicity and immunosuppression	Medical plants considered edible: <i>Alysicarpus vaginalis</i> and <i>Aerva lanata</i>
Norovirus	Foodborne gastroenteritis including vomiting and diarrhea with acute onset	Raw or undercooked oysters, herbs, vegetables, and fruits
Ebola virus	Ebola virus disease (EVD)	Bushmeat—meat from wildlife (e.g., monkeys and bats) slaughtered and prepared for human consumption

LIMITATIONS AND FUTURE DIRECTIONS OF ML/DL IN PATHOGEN DETECTION

Despite the remarkable progress of machine learning and deep learning in pathogen detection, several limitations hinder their routine clinical and laboratory use. Many models rely on large, well-annotated datasets, but biological data are often incomplete, noisy, or unbalanced, which can introduce bias and reduce accuracy. Deep learning approaches also require high computational power and advanced hardware, making them less accessible for laboratories in low-resource settings. Another issue is generalizability, as models trained on one dataset may fail to perform equally well on samples from different populations, geographical regions, or sequencing platforms. Over fitting remains a frequent problem, especially when training data are limited. Moreover, the limited interpretability of many neural networks makes explanation difficult, and clinicians often prefer tools that provide more transparent results.

Looking ahead, future research should prioritize the development of lightweight models that require less computational power but still provide high accuracy. Expanding open-access, standardized datasets will also be critical to reduce bias and improve reproducibility. Integration of ML with genomic sequencing, imaging, and real-time surveillance systems could provide more holistic diagnostic platforms. Efforts should also be directed toward explainable AI, allowing clinicians to better understand and trust model predictions. Finally, designing ML systems that can be applied in point of care and low resource settings will ensure that these technologies benefit global health, not just well equipped laboratories. Together, these advances will help overcome current challenges and unlock the full potential of ML/DL in pathogen detection and disease management.

CONCLUSION

Machine learning is transforming pathogen detection and classification in bacterial, protozoan, viral, and foodborne domains by overcoming the limits of conventional methods. These approaches have

improved diagnostic speed and accuracy, from bacterial morphology studies to early detection of viral infections such as COVID-19, and have advanced foodborne pathogen surveillance through imaging techniques. Future progress will depend on integrating ML with genomic sequencing, developing real-time surveillance systems, and creating models suitable for low-resource settings. Such advancements will strengthen public health, improve food safety, and support effective infectious disease management.

Authors' contribution:

AN, AL, NA, MA & HY Conceptualization of study and Data collection; AB, AA, EG & MI Manuscript writing and Formatting of manuscript; SA; Review and Editing; NA & AS Reverences cross check and Editing.

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