


# Detection of BABESIA BIGEMINA in Cattle Blood: AI and Impedance Methods

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**Abstract:** Haemoproteozoans are a diverse group of blood-borne parasites that cause significant economic losses in the veterinary field. In cattle, the three most common haemoproteozoans are Babesia, Theileria, and Anaplasma. Detection and treatment of these parasites are currently time-consuming and require laboratory facilities, which can delay treatment and lead to poorer outcomes, including increased anaemia and death. Infections also lead to diminished productivity, compromised reproductive performance, and increased vulnerability to secondary infections. To address this challenge, the project aims to develop novel technologies that will help in early detection of the parasite. The paper presents the design of an embedded AI software to detect the presence of Babesia bigemina protozoan within the cattle blood. The software used YOLO V8 model to train the system, and the software was integrated into a 3D printed open flexure microscope. The model yields a mean average precision of 66.2 percent for an IoU threshold of 0.5 and 34.7 percent for an IoU threshold of 0.5 to 0.9. The project also proposed research on the change in conductivity and impedance of the infected cattle blood and concluded that the presence of foreign particles, such as protozoans, in the blood samples resulted in a decrease in conductivity by values ranging between 2.2 to 3 milli siemens and an increase in impedance by a value within a range of approximately 330 to 450 milli ohm compared to normal blood samples.

## 1 INTRODUCTION

Haemoproteozoans are a diverse group of single-celled eukaryotic organisms transmitted by blood-feeding invertebrates, causing diseases in various animals and humans. They include genera like Babesia, Hepatozoon, Theileria, and Trypanosoma, with different species identified in wildlife such as cattle, dog and rats (A. S. Nair, 2011). Haemoproteozoan species affecting cattle include Theileria annulata, Trypanosoma evansi, and Babesia bovis. These parasites cause significant economic losses globally due to their impact on livestock health and productivity. Theileriosis, trypanosomiasis, and babesiosis are major haemoproteozoan diseases, with difficulties in diagnosis due to low parasitemia and concurrent infections. Tick-borne diseases, such as babesiosis, anaplasmosis, and theileriosis, are major constraints in the dairy industry, leading to decreased productivity and increased control costs. The hot and hu-

mid climate in tropical regions like India provides a favorable environment for haemoproteozoan parasites transmitted by vectors like ticks, posing a constant threat to susceptible animals (A. Tili and Jaffrezic-Renault, 2006). Haemoproteozoa species detected in cattle in Northern Kerala were Theileria like piroplasms and Babesia bigemina, with PCR revealing Trypanosoma evansi, Theileria sp., and B. bigemina (A. S. Nair, 2011). Detection and diagnosis of these diseases can be challenging as they require laboratory facilities and time consuming that even increases the rate of mortality. There is an alarming need of an on-field device that will accurately and precisely detect the presence of haemoproteozoan within the animal blood. This concept is the main building block of our device (C. S. Bhatnagar and Meena, 2015). A Non-invasive portable device can do the veterinarians and the farmers a good turn as they give out rapid results in the field detection fundamentally. The results would not only be rapid but also with towering accuracy. In particular this device facilitates to characterize to redline the different types of proto-

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zoans and its variations. Finding tick-borne diseases in cattle while out in the field poses significant challenges. Typically, diagnosing these diseases requires access to laboratory facilities equipped with specialized tools like microscopes for examining blood samples and PCR (Polymerase Chain Reaction) tests for identifying specific pathogens (B. R. Maharana and Sudhakar, 2016). However, such facilities may not always be readily available, especially in remote or rural areas where these diseases are prevalent. Furthermore, the cost associated with conducting these tests can vary depending on factors such as the type of test required and the location of the testing facility. In some cases, farmers may need to travel long distances to access testing centres, incurring additional expenses and time delays. Unfortunately, the delay in initiating treatment procedures due to the time-consuming nature of laboratory testing can have severe consequences. Which may even lead to the death of the animal, causing a devastating economic loss for the farmer. The primary outreach of this project is to concisely overcome the current challenges in detecting the presence of haemoprotezoan within the cattle blood, especially during the on-field examination. Thereby narrowing down the constraints which leads to the lagging of treatment initiation (B. R. Maharana and Sudhakar, 2016). Hence, the main aim of this project is to develop an on field low-cost portable device that could accurately and precisely detect the presence of protozoans from the blood sample under study. Additionally, the device may also be able to differentiate the genera of protozoans, since the drug administration for each protozoan is specific. The main objectives of the project are Analysing the relationship between protozoan infection and change in the electrical conductivity and impedance property of the blood due to the infection (I. Szymańska and Kaliszan, 2007). Developing a novel methodology based on the observation from the first objective to design a circuit to detect and measure the corresponding change in blood properties (G. C. McConnell, 2009). Designing and developing a software to accurately detect each genus of protozoan and concluding whether the blood sample under study is protozoan infected or not (B. K. Yap, 2018). For the time being the study only concentrates on one species. To successfully implement the designed software to a low-cost portable 3D printed microscope with high resolution.

## 2 METHODOLOGY

The proposed solution for the particular problem that this project addresses, can be done mainly in two

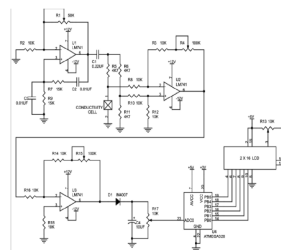


Figure 1: Circuit diagram of Microcontroller based Human Blood Conductivity Measurement System

ways, that is, one complete hardware and the other an AI software system embedded with 3D printed microscope. In this chapter the design of the hardware, components used to develop the circuit, role of each component in the circuit etc will be discussed first. Following session of the same chapter will introduce about the software design as well as the design and structure of the 3D printed microscope.

### 2.1 System Hardware: Detection of Haemoprotezoan in Cattle Blood by Analyzing the Conductivity and Impedance of the Blood Sample

This project aims to design a circuit to measure the conductivity and impedance of liquids (B. S. R. Bharati and Bhaskar, 2013), particularly blood samples, using important components like the LM741 op-amp, Arduino ATmega328, and a display as mentioned in figure 1. The system consists of three parts, namely:

- Signal Conditioning Circuit
- Conductivity Cell (Electrodes)
- Microcontroller and Display

#### Working Principle:

An AC signal is imposed on electrodes immersed in the sample. The ions in the liquid facilitate the flow of current, and the voltage across the electrodes reflects the liquid's impedance. The resulting signal is amplified, rectified, and converted to a readable format, from which conductivity is calculated (McAdams and Jossinet, 1995).

#### Signal Conditioning:

The signal conditioning unit consists of

- Wein Bridge Oscillator: It provides a 1kHz sine wave at an amplitude of 10Vpp by using the LM741 op-amp and an RC feedback network. The frequency is given by

$$f = \frac{1}{2\pi RC} \quad (1)$$

- **Impedance Bridge:** An AC signal from the oscillator excites the bridge, one arm of which contains the sample. Sample conductivity changes the impedance of the bridge, hence the voltage distribution.

The last component includes the Amplification and Processing which is followed by rectification and filtering to get a DC voltage representative of conductivity. This DC voltage is digitized through an ADC for processing.

## 2.2 System Software: Real Time Monitoring and Detection of the Haemoprotozoan from the Cattle Blood Sample Using Low-Cost Portable Microscope Embedded with AI Software

### 2.2.1 Blood Cell Classification Software

The developed software to classify the blood cells infected with haemoprotozoan will be architected to efficiently handle various tasks involved in processing the data, training and evaluation of models, and making inference(J. Knapper, 2022). The followings are included:

1. **Modular Components:** Dedicated modules for augmentation, model creation, training, evaluation, and inference of data. Reusability in functions and classes promotes maintainability and reusability of code.
2. **Flexibility:** Allows customization of model architectures, hyperparameters, pre-trained models, loss functions, and optimization techniques.
3. **Performance Optimization:** Batch processing, parallelization, and GPU acceleration are employed to ensure efficiency(C. Honrado, 2018).
4. **Error Handling and Platform Independence:** Robust error handling ensures smooth operation, while platform independence allows compatibility across operating systems.

### 2.2.2 Camera Module

The camera module includes a 4-megapixel CMOS sensor that can capture images at  $1920 \times 1920$  and record videos at  $640 \times 480$  pixels as shown in figure 2. This high resolution enables detailed analysis and live demonstrations. The CMOS sensors convert light into a digital signal, providing high-quality images suitable for microscopy applications(Berney and O'Riordan, 2008).



Figure 2: CMOS sensor obtained from a 1600x digital USB microscope.

### 2.2.3 Parasite Detection Methodology

1. **Data Preparation:** Blood sample images containing Babesia parasites are formatted in the YOLO structure, with annotation files marking bounding boxes around parasites. The dataset is split into training and validation sets.
2. **Model Selection:** YOLOv8, a state-of-the-art object detection model, is chosen for its high accuracy and real-time detection capabilities.
3. **Model Training:** The model is trained for 100 epochs using the AdamW optimizer. Data augmentation techniques involved include blur, median blur, grayscale conversion, and CLAHE to enhance robustness.
4. **Evaluation:** Model performance was measured in terms of precision, recall, and mean average precision.
5. **Deployment:** The developed model is deployed after validation in the identification of haemoprotozoon infections from blood sample images.,
6. **Improvement:** To perform well continuously, make iterations in fine-tunes, additional data, or advance the object detection algorithm.

### 2.2.4 Affordable Portable Microscope Design

The microscope utilizes a 3D-printed Open Flexure Microscope which resolves samples  $2\text{--}5\ \mu\text{m}$  in size. This open-sourced created, by Dr. Richard Bowman at the University of Bath, is a cost effective, portable solution, well suited for education and research. Images are acquired live and analyzed by the software to identify in real time infections caused by haemoprotozoa(J. Knapper, 2022), (C. Honrado, 2018), (Berney and O'Riordan, 2008).



Figure 3: Prototype of the hardware



Figure 4: PCB and circuit components

### 3 RESULT

#### 3.1 Blood Conductivity and Impedance

The objective of this project is to measure the conductivity and impedance of blood samples using a conductivity measuring circuit with a Wein bridge oscillator, LM741 IC, copper electrode arrangement, and an Arduino microcontroller as shown in the figure 3.

A conductivity measuring circuit was constructed comprising a Wein bridge oscillator with an LM741 IC as the signal source generator. A copper electrode arrangement was used as the conductivity cell, immersed in the blood samples. An Arduino microcontroller was employed to process the signals and calculate conductivity and impedance values. Figure 4 shows the PCB fabrication.

##### 3.1.1 Observations

The conductivity measurements ranged from 2.2 ms to 5.8 ms. A total of 20 readings were obtained within this range, with incremental changes in conductivity values. Impedance values were calculated as the reciprocal of conductivity.

The presence of foreign particles, such as protozoans, in the blood samples resulted in a decrease in conductivity to a range of 2.2 to 3ms and an increase in impedance within a range of 330 to 450 mohms compared to normal blood samples. This suggests that the electrical properties of blood are influenced by the presence of contaminants, highlighting the im-

Table 1: Observations of conductivity and impedance measurements of blood samples.

Reading	Normal Blood Sample		Infected Blood Sample	
	Conductivity (mS)	Impedance (mΩ)	Conductivity (mS)	Impedance (mΩ)
1	4.4	227.27	2.8	357.14
2	4.35	229.89	3.2	312.50
3	4.3	232.56	3.0	333.33
4	4.25	235.29	2.4	416.67
5	4.2	238.10	2.9	344.83
6	4.1	243.90	3.4	294.12
7	4.15	240.96	2.3	434.78
8	4.3	232.56	3.3	303.30
9	3.95	253.60	2.6	384.62
10	3.8	263.16	2.2	454.44

portance of monitoring conductivity and impedance for detecting abnormalities.

#### 3.2 Result of the Software System

The project integrated YOLOv8 into a 3D-printed inverted microscope to develop an automated system for detecting Babesia Bigemina in blood samples. Positive and negative samples were tested, and the software effectively analyzed, classified, and detected the parasite. Due to dataset availability, the study focused on Babesia Bigemina.

##### 3.2.1 Dataset and Training

The dataset consisted of blood sample images with bounding boxes for Babesia parasites. Training and validation sets were made.

- **Model Training:** YOLOv8 was trained for 100 epochs using the AdamW optimizer with automatic learning rate and momentum.
- **Data Augmentation:** Techniques include blur, median blur, grayscale conversion, and contrast-limited adaptive histogram equalization.

##### 3.2.2 Model Evaluation and Results

The model's performance was evaluated on the validation set based on metrics such as precision, recall, mAP50, and mAP50-95 computed at various IoU thresholds. Observations can be seen in Table 4.2.

Table 2: Results of the Model.

SL. No	Performance Metrics	Babesia	Extracellular
1	Precision	0.685	0.462
2	Recall	0.736	1.000
3	mAP50	0.892	0.995
4	mAP50-95	0.621	0.895

##### 3.2.3 Metric Definitions

- **Precision:** Ratio of true positives to total predicted positives, measuring prediction accuracy.

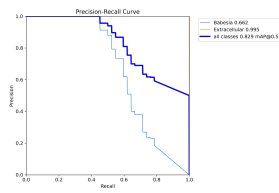


Figure 5: Precision-Recall Curve

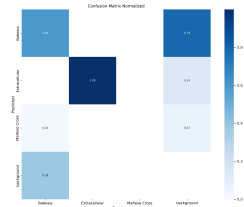


Figure 6: Normalized confusion matrix

- Recall: Ratio of true positives to total actual positives, assessing the model's ability to detect all instances.
- mAP50: Mean average precision at IoU 0.5, providing a balance between precision and recall.
- mAP50-95: Mean average precision across IoU thresholds (0.5–0.95), offering a comprehensive evaluation.
- Class-Specific Metrics: Precision, recall, and mAP for individual classes, enabling detailed performance analysis.

### 3.2.4 Observations

For YOLOv8, satisfactory metrics were obtained with more room for improvement in detecting Babesia parasites. Future optimizations can be done for better results with increased accuracy and generalization.

## 4 DISCUSSION AND CONCLUSION

### 4.1 Conductivity and Impedance of Protozoan Affected Blood

The project developed hardware and software technologies for the detection of haemoprotozoan in cattle

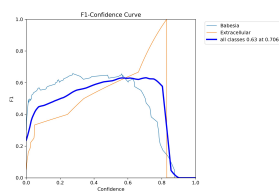


Figure 7: F1- Confidence Curve

blood. The hardware consists of a conductivity measurement circuit consisting of a Wein bridge oscillator, LM741 IC, copper electrodes, and an Arduino microcontroller. This setup facilitates portable blood conductivity and impedance measurement, which is of great value during the identification of changes in blood properties.

While blood impedance measurement is preferred for the detection of blood-borne parasites, results may be affected by factors such as temperature and foreign particles that limit accuracy. These disadvantages could be overcome by incorporating advanced sensors and electrodes.

In conclusion, as much as the LM 741-based conductivity circuit is not fully accurate for the detection of haemoprotozoa, it effectively detects changes in impedance and conductivity of blood.

### 4.2 Real Time Monitoring and Detection of Haemoprotozoan Using Embedded Ai System

The proposed study is to design a portable, non-invasive device for the detection of haemoprotozoan parasites in cattle blood, keeping in mind the limitations of existing methods of diagnosis that are time-consuming, expensive, and require laboratory facilities. The focus is on the development of an on-field, low-cost device for identifying protozoan genera so that appropriate drugs can be administered. The study focuses on the relationship between protozoan infection and changes in blood conductivity and impedance properties. A software integrated with a low-cost 3D-printed high-resolution microscope is developed to analyze blood samples and detect infections. The project emphasizes the potential of biomedical engineering in enhancing veterinary healthcare, supporting cattle farming—a key contributor to Indian GDP and milk/meat production (E. O. Adekanmbi and Srivastava, 2023).

Haemoprotozoan diseases have a major impact on livestock, resulting in financial losses from infections and mortalities (Garcia and Sabuncu, 2019). Facilities for diagnosis are generally insufficient, relying on conventional techniques of microscopic examination and serological tests like ELISA, which are laborious and time-consuming (Technologies, 2017). Modern molecular techniques such as PCR detect the disease during its latent phase more effectively (E. O. Adekanmbi and Srivastava, 2023). Electrochemical Impedance Spectroscopy and immunosensor-based methods show potential in the diagnosis of diseases such as Babesia bovis through the analysis of electrical properties (Cole, 1941) (Macdonald and Johnson,



2005)(Garcia and Sabuncu, 2019).

Although the novel diagnostic approach was introduced, certain constraints occurred. The presence of other pathogens, such as viruses, bacteria, or other protozoan genera, was not considered; temperature dependencies during experiments were not considered, either. Low-cost components reduced accuracy, and software analysis was restricted to Babesia, excluding complex genera like Theileria and Anaplasma due to the limitation of the dataset. These factors constrained the model's total mean average precision. Future research can address these limitations by replacing low-cost components with precise ICs or sensors like AD5933(H. Cho and Baek, 2021), designing specific electrodes for impedance-based detection, and expanding datasets through more blood sample collection and annotation.

Enhancing training models to differentiate haemo-protozoan genera accurately and employing advanced microscopic technologies like lensless microscopy or muscope can broaden the device's applications.

These methodologies could also be applied to the diagnosis of human parasites such as Plasmodium and Trypanosoma, laying the foundation for innovative veterinary diagnostics with significant societal and economic benefits.

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