B-5745
[1-9]

RESEARCH ARTICLE Indian Journal of Animal Research

Integrated DNA Barcoding and QR Code-based Image Recognition for Identification of Indian Cattle Breeds

Dadala Mary Mamatha[1], Lakshmaiah Para Venkata[2], Damodar Naidu Thanikonda[3], Haripriya Kotagaram[4], Swetha Kumari Koduru[4]

10.18805/IJAR.B-5745

ABSTRACT

Background: The study aimed to establish molecular credentials for Indian bovine species through an integrated approach combining DNA barcoding, QR code generation and image analytics. Blood samples from 16 Indian bovine species were collected and mitochondrial COX-I gene amplification was targeted using universal primers.

Methods: DNA was isolated from blood samples and validated using agarose gel electrophoresis. PCR conditions for mitochondrial COX-I gene amplification were standardized and purified PCR products were sequenced. The resulting DNA barcodes were submitted to the Barcode of Life Data Systems (BOLD) database for authentication and barcode generation. Gene-based QR codes were generated using Python and image analyses integrating morphological and molecular features were performed for species authentication.

Result: Sixteen DNA barcodes representing Indian bovine species were successfully generated and uploaded to the BOLD database. Corresponding QR codes were created and image analytics supported accurate species identification. The integrated framework demonstrated effectiveness in precise identification, molecular characterization and authentication of Indian bovine genetic resources.

Key words: Biodiversity conservation, Cattle breeds, DNA barcoding, Genetic diversity, Indian bovine species.

INTRODUCTION

Indian bovine species

India continues to possess the largest bovine population in the world. According to the 20th Livestock Census (2019) conducted by the Government of India, the country is home to approximately 193.46 million cattle and 109.85 million buffaloes, accounting for over 300 million bovines in total. India alone contributes nearly 55% of the global buffalo population and about 13 to 14% of the world's cattle, underscoring its global significance in bovine genetic resources. Indigenous and non-descript cattle constitute the majority, with around 142 million indigenous/non-descript cattle, while crossbred and exotic cattle account for about 26.5% of the total cattle population. The Indian subcontinent is recognized as the primary centre of domestication of the riverine water buffalo (Bubalus bubalis). Although a substantial proportion of indigenous cattle remain non-descript, highlighting the need for systematic characterization, conservation and genetic improvement programmes. Twenty two buffalo breeds and fifty five recognized indigenous cow breeds make up the nation's bovine genetic resource (https://agriportal.cg.nic.in/).

Native animals are resilient and possess traits like heat tolerance, illness resistance and the capacity to flourish in harsh weather. While indigenous breeds of cattle are more resilient and resistant to ticks and disease, exotic breeds and crossbred cattle are more vulnerable to tropical diseases (Larcombe *et al*., 2022). Furthermore, only in a farm management system with expensive inputs do crossbred and exotic cattle perform at their best. Poor farmers raise most of the animals in India's Low-Input and Low-Output farm management system. The units like National Kamdhenu Breeding Centres (NKBC) were established to serve as a Centre of Excellence for the development and conservation of Indigenous Breeds in a comprehensive and methodical way. At AP Chintaladevi's NKBC Centre, bovine breeds namely Deoni, GIR,

[1]Department of Biosciences and Sericulture, Sri Padmavati Mahila Visvavidyalayam, (Women's University), Tirupati-517 502, Andhra Pradesh, India.
[2]Andhra Pradesh State Veterinary Council, Vijayawada-520 011, Andhra Pradesh, India.
[3]Department of Animal Husbandry, Vijayawada-520 011, Andhra Pradesh, India.
[4]Department of Biosciences and Sericulture, Sri Padmavati Mahila Visvavidyalayam, (Women's University), Tirupati-517 502, Andhra Pradesh, India.

Corresponding Author: Dadala Mary Mamatha, Department of Biosciences and Sericulture, Sri Padmavati Mahila Visvavidyalayam, (Women's University), Tirupati-517 502, Andhra Pradesh, India. Email: prof.mamatha@gmail.com

How to cite this article: Mamatha, D.M., Venkata, L.P., Thanikonda, D.N., Kotagaram, H. and Koduru, S.K. (2026). Integrated DNA Barcoding and QR Code-based Image Recognition for Identification of Indian Cattle Breeds. *Indian Journal of Animal Research.* 1-9. doi: 10.18805/IJAR.B-5745.

Submitted: 27-12-2025 **Accepted:** 18-02-2026 **Online:** 26-03-2026

Jafarabadi, Kangayam, Kankrej, Killari, Mahsena, Malanad Gidda, Murrah, Ongole, Pandharpuri, Punganuru, Rathi, Red Sindhi, Sahiwal and Tharparkar were selected and considered for blood sample collection of cattle breeds.

DNA barcoding-tool for molecular identification

The multidisciplinary effort of DNA Barcoding, Gene based QR coding will bring forth a significant contribution to support, conserve the Indigenous breeds, exotic and crossbreeds or lines of existing bovine livestock of the India and augment the bovine conservation, gene mapping of the bovine germplasm resources. Accelerating the inventory and quick analysis of the diversity, Identification of the new species, analysis of their molecular phylogeny will in turn contribute to traditional Knowledge, conservation; and bio piracy elimination (Yang *et al*., 2018).

Research on genetic diversity serves as a foundation for the creation of creative policies and plans for preserving the natural diversity found in native, exotic and crossbred animals. This information will also be useful in identifying important agricultural animal breeds, which paves the way for the identification and development of prospective future breeds for the creation of effective breeding programs that will increase the robustness and production of milk. Breeds or lines of current livestock will have legal protection through the use and preservation of bovine breed genetic resources (Sarang *et al*., 2024).

Image data analysis and cloud computing

Towards an innovative angle of bovine species identification and authentication Image data analysis can be employed. The machine learning methods, like convolutional neural networks (CNNs), can help classify species based on features like body size, shape, horn configuration and coat patterns (Sarızeybek *et al*., 2023). Cloud computing provides the infrastructure and resources for managing and analyzing large datasets without the need for physical data storage or high-performance computing hardware on-site.

Therefore, the present study objective is to identify and catalogue bovine genetic diversity through DNA barcoding, accompanied by Gene-based QR coding and image analysis to link individual animals to their genetic and phenotypic profiles. Further, this study generates a molecular atlas to understand genetic resources and variations by leveraging image data analysis for phenotypic classification and breed recognition. This would also support conservation efforts for endangered or indigenous bovine breeds and enhance breeding programs with genetic insights.

MATERIALS AND METHODS

Sample collection and DNA isolation

Sample collection

Bovine blood samples (2 nos/breed) were collected from National Kamadhenu Breeding Centre, Chintaladevi village, Nellore. The bovine breeds include Deoni, GIR, Jafarabadi, Kangayam, Kankrej, Killari, Mahsena, Malanad Gidda, Murrah, Ongole, Pandharpuri, Punganuru, Rathi, Red Sindhi, Sahiwal and Tharparkar as shown in Fig 1. The experiments were conducted in Dept. of Biosciences and Sericulture, SPMVV, Tirupati during 2021-23.

Genomic DNA isolation

The fresh samples were processed for isolation of Genomic DNA using CTAB method. The sample was transferred to a fresh microcentrifuge tube and pre-warmed CTAB and Beta-Mercaptoethanol and SDS were added. The reagents are mixed and proteinase K was added. Centrifugation was performed 14,000 rpm for 10 min. The pellet was discarded and to the supernatant, equal volumes of Phenol-Chloroform-Isopropanol was added and vortexed. The mixed sample was kept in -20°C for one hour, followed by centrifugation at 10,000 rpm for 10 min. The supernatant was discarded and 70% chilled ethanol with ammonium acetate was added to the pellet. The mix



Fig 1: Selected bovine breeds.

was centrifuged at 10,000 rpm for 10 min. The supernatant was discarded and TE buffer was added to the dried pellet.

Mitochondrial COX-I gene amplification, purification and sequencing

Gene amplification

The designing and selection of suitable primers is essential for a successful gene amplification. To amplify the mitochondrial COX-I gene, 3 pair of species-specific primers were used (Table 1). Cytochrome Oxidase subunit I gene (COX-I) sequences of various species belonging to different order of were amplified using the primers retrieved from BOLD database.

Partial mitochondrial COX-I gene was amplified among cattle Breeds for their molecular Identification and molecular phylogeny studies. PCR with kit components were used and PCR Conditions for the gene amplification was standardized by applying different annealing temperatures using Gradient Master cycler Nexus (Applied Biosystems) thermocycler. The gene got amplified with a initial denaturation at 94°C for 3 mins followed by 35 cycles of denaturation, annealing and extension at 94°C (1min), 48°C (40 sec) and 72°C (3 min) respectively and final extension at 72°C for 7 min.

Purification

The extra nucleotides, primer residues and buffer salts were removed in PCR product cleanup process and ethanol precipitation. Exo-SAP Digestion was performed to remove excess primers and nucleotides in the reaction mixture, 2.5 µl of PCR product was treated with 0.25 µl Exonuclease-I (1U/µl) and 0.5 µl (1U/µl) of Shrimp alkaline phosphatase (SAP). Later 10X SAP buffer was added and incubation at 37°C for 45 minutes. The samples were re-incubated at 80°C for 15 minutes to inactivate Exonuclease-1 and SAP enzymes (Hajibabaei *et al*., 2006).

Cycle sequencing was performed to amplify the PCR products prior to sequencing. Sequencing primers with 0.8 PM concentration were used in each PCR reaction and obtained products are purified by cleanup process. The temperature conditions include 5 stages with temperatures of 96°C (5 min), 96°C (30 sec), 50 °C (15 sec), 64°C (4 min) and 4°C respectively. The samples obtained from cycle sequencing were purified by following cleanup step to eliminate ddNTPs, leftover primers and salts in the product by Big Dye (R) X Terminator (Big Dye Terminator v3.1 clean up Applied Biosystems, USA).

Gene sequencing

The Big Dye (R) X Terminator (Big Dye Terminator v3.1 clean up Applied Biosystems, USA) was vortexed at ~2500 rpm for 45 minutes due to its viscous nature. The samples were kept for centrifugation at 1000 × g for 2 minutes prior to pipetting in sequencer (AB3130, USA). The sequencer contains four capillary tubes with the length of 50 cm. POP7(R) (AB1, USA) polymer was added to the reaction. Finally, the templates were digested with HiDi-formamide and sequenced using 'ABI 3130 genetic' bidirectional sequencer. DNA Sequences in Chromatogram file was generated by (ABI Sequencer) through Sangers Dideoxy sequencing.

Submission to bold database

Bioinformatic analysis

The amplified sequences were edited using Codon code aligner and MEGA software and submitted to BOLD - Barcode of Life Database (https://www.barcoding.si.edu). The trace files were edited by using "CodonCode aligner" software to remove ambiguous bases, noisy peaks. The trace files of the sequences obtained were in different lengths. Hence, it is compulsory to delete noisy or messy peaks, ambiguity codes. The ends of trace files were trimmed from each sequence to remove primers. 'N' s is placed in the position of low-quality bases. Later, raw sequence information was drawn from each trace file. Forward and reverse sequences of each specimen was combined. Consensus sequence was retrieved from the combined sequence which specifies complete contig of individual Bovine breed.

The Sequence analysis of corrected COX-I gene sequences of Bovidae species were carried out in MEGA (Molecular Evolutionary Genetics Analysis) (Tamura *et al*., 2021) software. It is a windows-based user-friendly program with graphical user interface. It also contains the multiple alignment program ClustalW to identify the conservation between the sequences. Comparative studies of COX-I sequences were studied through multiple sequence alignment. The ends of the alignments were clipped to remove flanking regions. The edited sequences were again compared using ClustalW in MEGA server to get more accurate alignment sets. These sequences were further corrected manually and adjusted where needed to avoid alignment errors. The sequence alignment using Codon code aligner and MEGA software analyses.

Table 1: Details of primer sequences used in the amplification of CoX-I gene.

Primer name	Direction	Sequence description
SNLF	Forward	5′GGTCAACAAATCATCATAAAGATATTGG3′
SNLR	Reverse	5′TAAACTTCAGGGTGACCAAAAAACAT3′
SNVF	Forward	5′TTCTCAACCAACCACAARGAYATYGG3′
SNVR	Reverse	5′TAGACTTCTGGGTGGCCRAARAAYCA3′
SNZF	Forward	5′TCTCAACTAAYCAYAAAGAYATYGG3′
SNZR	Reverse	5′TAAACTTCRGGGTGACCAAARAATCA3′

For comparative studies, BLAST (Basic local alignment search tool) (Altschul *et al*., 1997) tool has been used and it compared two sequences by identifying the similar local regions. BLAST-N algorithm was executed to identify the closely related species to the query sequences of each cattle specimen and compared against the reference database with nucleotide-nucleotide evaluation (BLAST-N) by customizing the settings.

Submissions

Genbank and the Barcode of life data systems (BOLD) are two major publicly available databases to access and submit DNA barcode data of animals and plants. After comparative studies using BLAST search, the accurate annotated COX-I gene sequences of Bovidae species belonging to eight different orders are made ready for submission to bold (The Barcode of Life Data systems) database for the creation of DNA barcodes.

To get an authenticated and validated barcode for a sequence, numerous files namely Voucher data file, Taxonomy file, Specimen data file, Collection data file and details are prepared and generated. For each partial mitochondrial COX-I sequence, name of the species, voucher data, institution where it was processed, details of catalogues, collection data, specimen identifier, information of sequence (~650 bps), primer sequence and raw sequence particulars must be provided.

Gene based-QR code generation

Based on the DNA Barcodes generated, Quick Response (QR) codes were developed to each Bovidae species selected for this study for their automatic identification. Python program was used for QR code creation. When it comes to encoding DNA barcode sequences, QR codes outperform other 2D codes (Matrix) in terms of compression efficiency. The Jupyter-Python kernel version (Anaconda)'s open source QR Code Library was modified to create a program that encodes DNA sequences. Upon scanning QR code, the complete details of the Cattle breed along with its DNA Barcode, Breed characteristics and image will be displayed. The steps involved in creating DNA based QR codes were shown in Fig 2.

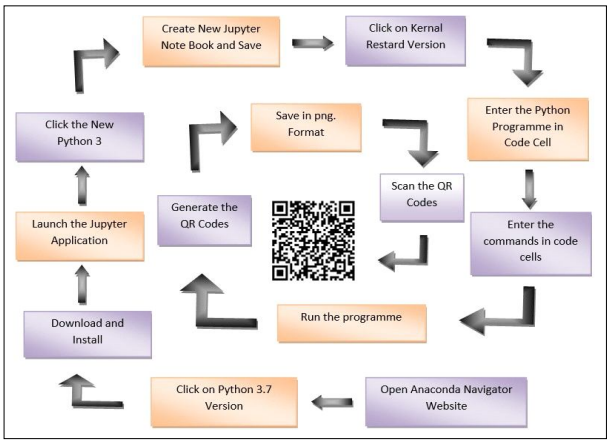


Fig 2: DNA barcode based QR code generation using python.

Cattle image analysis using RGB and deep feature extraction techniques

To accurately identify cattle breeds, high-resolution images of individual animals were captured from multiple angles, focusing on distinct anatomical features such as horns and muzzles. These images were then imported into Python using the latest image processing libraries (*e.g*., OpenCV, scikit-image, PIL) for advanced analysis.

Each imported image underwent preprocessing and segmentation to isolate specific regions of interest (ROIs) crucial for breed identification-primarily the horn structure and muzzle area, including the mouth, nose and upper jaws. A deep extraction method was used, which leverages deep learning or morphological operations to extract these features with high precision while minimizing the inclusion of irrelevant background or body parts (Fig 3).

Upon loading, the image dimensions were recorded as (3504, 6240) for grayscale images, indicating a single channel and (3504, 6240, 3) for RGB images, signifying three separate channels-Red, Green and Blue. These channels represent the color intensity values for each pixel and are crucial for distinguishing subtle texture and color differences in cattle features that might not be visible in grayscale images.

The use of RGB image decomposition allows for:

- **Channel-wise analysis:** Each color channel may highlight different aspects of the cattle's features (*e.g*., horns may have sharper contrast in the red channel).
- **Enhanced feature visibility:** Certain cattle breeds may have distinct pigmentation patterns that are more visible in specific channels.
- **Composite feature mapping:** Combining information from all three channels enables a richer and more accurate feature extraction process.

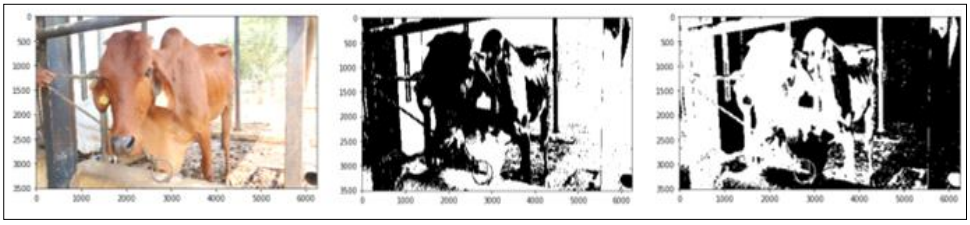


Fig 3: Cattle image analysis and applying deep extraction feature.

To streamline the classification process and improve computational efficiency, feature extraction techniques were applied to identify and quantify key morphological descriptors. Finally, all extracted features are compared against a custom-built image database of known cattle breeds. Machine learning or deep learning classifiers (*e.g.*, SVM, CNNs) can be trained on these features to accurately identify the breed based on horn shape, muzzle size and structural patterns revealed through RGB image analysis.

The PCR products were subjected Exo-SAP digestion and clean-up process. The purified amplified gene was sequenced. The obtained DNA sequences were trimmed, edited and aligned using Codon Code Aligner and MEGA tools. The output is as shown in Fig 4.

The final sequences were submitted to BOLD with the respective trace files and the taxonomy files. BOLD Database reviewed and has generated DNA Barcodes for the selected Bovine species. The generation of directory of Indian Bovine species' DNA Barcodes in BOLD database is first of its kind as shown in Fig 4. The DNA Barcodes for the selected Bovine breeds were tabulated in Table 2.

QR codes were generated based on the obtained DNA Barcodes using Python Code. Each characteristic of the breed is also linked with the QR code. Upon scanning, the morphological and molecular parameters of bovine species would be displayed. The output page for QR code shows cattle image, DNA Barcode and Characteristics as shown in Fig 5.

Further, using the deep feature extraction, image analysis was performed for unique identification of the cattle breeds. Different threshold values were applied to find the cattle image features and to know the breed, followed by extracting muzzle, horns, individually as shown in Fig 6.

The selected extraction point is compared with the different cattle images in the database which are created with basic structure of the cattle to find the breed type. Similar studies were conducted for all selected cattle breeds.

RESULTS AND DISCUSSION

In this study, 16 distinct bovine breeds from India were selected for comprehensive molecular profiling using an integrated approach that combines DNA barcoding, QR code tagging and advanced image processing. This multifaceted strategy aims to enhance the precision, traceability and sustainability of managing India's bovine genetic resources. Genetic diversity analysis is essential for breed conservation and improvement strategies. Studies on indigenous cattle populations have demonstrated significant mitochondrial diversity, which supports the need for structured conservation programs (Zhao *et al*., 2021; Sarang *et al*., 2024).

The mitochondrial cytochrome c oxidase subunit I (COX-I) gene has been widely accepted as a reliable molecular marker for species-level discrimination due to its conserved primer-binding regions and sufficient interspecific sequence divergence (Hebert *et al*., 2003; Folmer *et al*., 1994). Its applicability in livestock genetic studies has been supported by recent investigations demonstrating effective differentiation among cattle populations using COI-based molecular approaches (Utami *et al*., 2024).

Traditional breed identification methods primarily rely on morphological characteristics such as coat color, horn shape and body conformation. However, these traits can be influenced by environmental conditions, management practices and genetic admixture, potentially leading to misclassification. In contrast, DNA barcoding provides a stable genetic signature that remains unaffected by external variables. Molecular authentication has proven valuable not only for breed verification but also for ensuring transparency in animal-derived products and preventing species mislabeling within supply chains (Smith *et al*., 2018; Sultana *et al*., 2017). Furthermore, previous research has demonstrated the utility of DNA barcoding in diverse animal taxa, reinforcing its versatility as a standard identification tool (Alam *et al*., 2020).

Genetic diversity assessment is a critical component of sustainable livestock management. Indigenous bovine breeds possess adaptive traits such as thermotolerance, disease resistance and efficient feed utilization, which are particularly important under changing climatic conditions. Molecular studies targeting both mitochondrial and nuclear genes have provided valuable insights into breed-specific variation and adaptive potential. For example, characterization of stress-related genes such as HSP70 in buffalo breeds has highlighted the importance of gene-level analysis for understanding environmental resilience (Singh *et al*., 2024). Similarly, identification of single

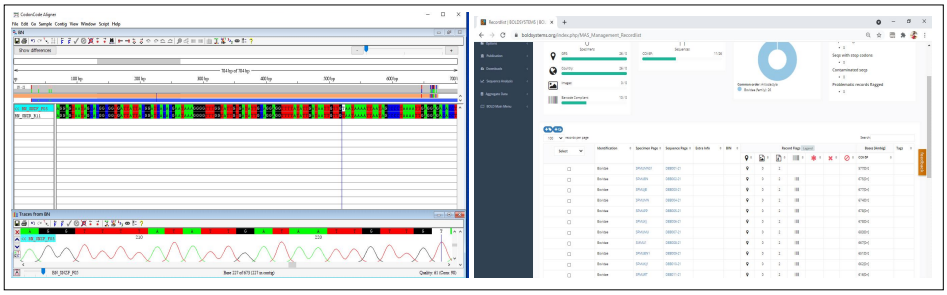


Fig 4: Sequence editing using MEGA and submission to bold database.

Table 2: CoX-I gene sequence and respective DNA barcodes of selected bovine species.

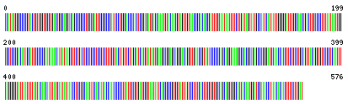
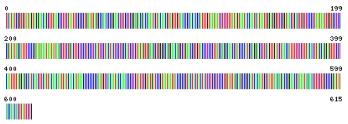
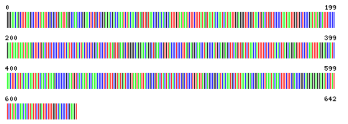
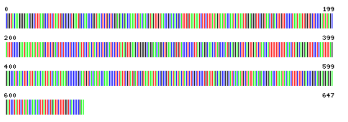
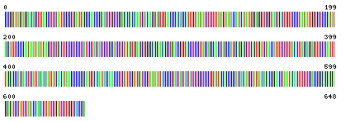
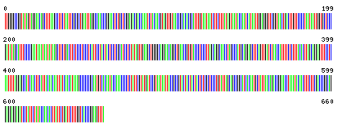
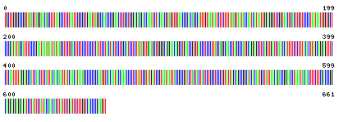
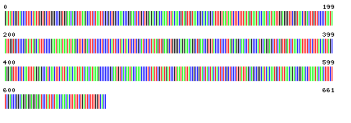
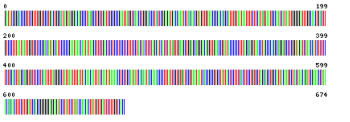
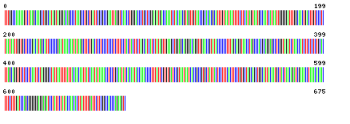
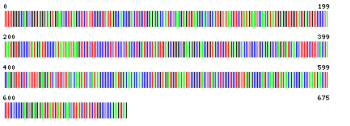
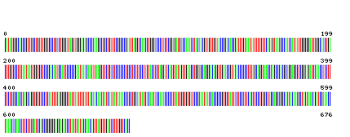
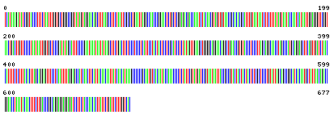
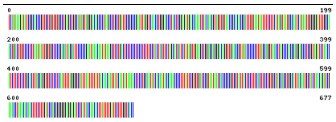
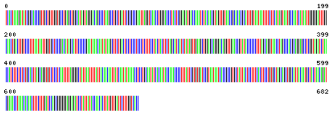
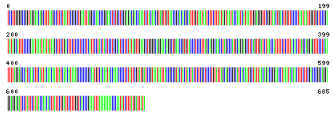
Sample ID	Sequence	Barcode
SPMUMNS1	TCATATCGGCCCGTACTGTTGGTGCCTGACTGGCAGGACAGCCCGCCTGCTGATTCGCG CCGAATCCTGGGACCCTACTCGGAGATGACCAAATCTACAACGTAGTTGTAACCGCACAC GCATTTGTAATAATTTTCTTCATAGTAATGCCAATCATAATTGGGGGATTTGGCAACTGA CTTGTCCCTTTAATAATTGGTGCTCCCGACATAGCATTCCCCGAATAACAATATAAGC TTCTGACTCCTCCCTCCCTCTTTCCTACTACTTCTAGCATCATCTATATTGAAGCTGGAG CAGGGACAGGTTGAACCGTATACCCCCCAGCAGGTAAAGCCCACGCAGAGCCTCGGATCT AACCATCTTCTCTGCACTTACAGGTGTCTCCTCAATTTTAGGGGCTATTAATTTTATT ACAACAATTATCAATATAAACCTCCTGCAATATCACAATACCAAACCCCTTTATTCTGTG ATCCGTAATAATCACGCCGTGCTATTACTCCTTTCACTTCCTGTGCTAGCAGCTGGCAT TACAATGGATCGAAACCTAAACACAACTTTCTTTGAT	
SPMRT	TCATATCGGTGTATAGTAGGAACAGCTTTAAGCCTTCTAATTCGCGCTGAATTAGGCCAA CCCGGAACTCTGCTCGGAGACGACCAAATCTACAACGTAGTTGTAACCGCACACGCATTT GTAATAATCTTCTTTATAGTAATACCAATCATAATTGGAGGGTTCGGTAACTGACTTGTT CCCCTAATAATTGGTGCTCCCGATATAGCATTTCCCCGAATAAATAATATAAGCTTCTGA CTTCTCCCTCCCTCATTCCTACTACTCCTCGCATCCTCTATAGTTGAAGCTGGGGCAGGA ACAGGCTGAACCGTGTACCCTCCCTTAGCAGGCAACCTAGCCCATGCAGGAGCTTCAGTT GATCTAACCATTTTCTCTTTACACTTAGCAGGAGTTTCCTCAATTTTAGGAGCCATCAAC TTCATTACAACAATTATCAACATAAAGCCCCCGCAATGTCACAATACCAAACCCCTCTTA TTCGTATCGTACTATAATTACCGCCGTACTACTACTCTCGCTCCCTGTATTAGCAGCCGGC GCCGGCATCACAATGCTATTAACAGACCGGAACCTAAATACAACTTTCTTGCCCGGATAT ACCAACACTTATTCTG	
SAHIWAL	GGGAACAGCTTTAAGCCTTCTAATTCGCGCTGAATTAGGCCAACCCGGAACTCTGCTCGG AGACGACCAAATCTACAACGTAGTTGTAACCGCACACGCATTTGTAATAATCTTCTTTAT AGTAATACCAATCATAATTGGAGGGTTCGGTAACTGACTTGTTCCCCTAATAATTGGTGC TCCCGATATAGCATTTCCCCGAATAAATAATATAAGCTTCTGACTTCTCCCTCCCTCATT CCTACTACTCCTCGCATCCTCTATAGTTGAAGCTGGGGCAGGAACAGGCTGAACCGTGTA CCCTCCCTTAGCAGGCAACCTAGCCCATGCAGGAGCTTCAGTTGATCTAACCATTTTCTC TTTACACTTAGCAGGAGTTTCCTCAATTTTAGGAGCCATCAACTTCATTACAACAATTAT CAACATAAAGCCCCCCGCAATGTCACAATACCAAACCCCTCTATTCGTATGACAGCCGGCA TCACAATGCTATTAACAGACCGGAACCTAAATACAACTTTCTTGCCCGGATAGGAGGAGA TCCTATTCTATACCAACACTTATTCTGATTCTTTGGTCACCCTGAAGT	
SPMUOL	CGGTACAAGGGGAACGGCTTTAAGCCTTCTAATTGCGCGCTGAATGCCAACCCGGTAACT CTGCTCGGAGACGACCAAATCTACAACGTAGTTGTAACCGCACACGCATTTGTAATAATC TTCTTTATAGTAATACCAATCATAATTGGAGGGTTCGGTAACTGACTTGTTCCCCTAATA ATTGGTGCTCCCGATATAGCATTTCCCCGAATAAATAATATAAGCTTCTGACTTCTCCCT CCCTCATTCCTACTACTCCTCGCATCCTCTATAGTTGAAGCTGGGGCAGGAACAGGCTGA ACCGTGTACCCTCCCTTAGCAGGCAACCTAGCCCATGCAGGAGCTTCAGTTGATCTAACC ATTTTTCTTTACACTTAGCAGGAGTTTCCTCAATTTTAGGAGCCATCAACTTCATTACA ACAATTATCAACATAAAGCCCCCCGCAATGTCACAATACCAAACCCCTCTATTCGTATGA TCCGTAATAATTACCGCCGTACTACTACTACTCTCGCTCCCTGTATTAGCAGCCGGCATC ACAATGCTATTAACAGACCGGAACCTAAATACAACTTTCTTCGACCCGGCAGGAGGAGGA GATCCTATTCTATACCAACACTTATTCTGATTCTTTGGTCACCCCGAA	
SPMUPG	GCCGGTATAGTAGGAACAGCTTTAAGCCTTCTAATTCGCGCTGAATTAGGCCAACCCGGA ACTCTGCTCGGAGACGACCAAATCTACAACGTAGTTGTAACCGCACACGCATTTGTAATA ATCTTCTTTATAGTAATACCAATCATAATTGGAGGGTTCGGTAACTGACTTGTTCCCCTA ATAATTGGTGCTCCCGATATAGCATTTCCCCGAATAAATAATATAAGCTTCTGACTTCTC CCTCCCTCATTCCTACTACTCCTCGCATCCTCTATAGTTGAAGCTGGGGCAGGAACAGGC TGAACCGTGTACCCTCCCTTAGCAGGCAACCTAGCCCATGCAGGAGCTTCAGTTGATCTA ACCATTTTCTCTTTACACTTAGCAGGAGTTTCCTCAATTTTAGGAGCCATCAACTTCATT TCATTACAACAATTATCAACATAAAGCCCCCGCAATGTCACAATACCAAACCCCTCTATTC TGATCCGTAATAATTACCGCCGTACTACTACTACTCTCGCTCCCTGTATTAGCAGCCGGC ATCACAATGCTATTAACAGACCGGAACCTAAATACAACTTTCTTCGACCCGGCAGGAGGA GGAGATCCTATTCTATACCAACACTTATTCTGATTCTTTGGTCACCCGA	
SPMUKM	TTGGGGCCGGTATAGTAGGAACAGCTTTAAGCCTTCTAATTCGCGCTGAATTAGGCCAAC CCGGAACTCTGCTCGGAGACGACCAAATCTACAACGTAGTTGTAACCGCACACGCATTTG TAATAATCTTCTTTATAGTAATACCAATCATAATTGGAGGGTTCGGTAACTGACTTGTTC CCCTAATAATTGGTGCTCCCGATATAGCATTTCCCCGAATAAATAATATAAGCTTCTGAC TTCTCCCTCCCTCATTCCTACTACTCCTCGCATCCTCTATAGTTGAAGCTGGGGCAGGAA CTGAGCTGAACCGTGTACCCTCCCTTAGCAGGCAACCTAGCCCATGCAGGAGCTTCAGTTG ATCTAACCATTTTCTCTTTACACTTAGCAGGAGTTTCCTCAATTTTAGGAGCCATCAACT TCATTACAACAATTATCAACATAAAGCCCCCGCAATGTCACAATACCAAACCCCTCTAT TCGTATGATCCGTAATAATTACCGCCGTACTACTACTACTCTCGCTCCCTGTATTAGCAG CCGGCATCACAATGCTATTAACAGACCGGAACCTAAATACAACTTTCTTCGACCCGGCAG GAGGAGGAGATCCTATTCTATACCAACACTTATTCTGATTCTTTGGTCACCCGGAAGTTT A	
SPMUTP	TGCTTGGGGCCGGTATAGTAGGAACAGCTTTAAGCCTTCTAATTCGCGCTGAATTAGGCC AACCCGGAACTCTGCTCGGAGACGACCAAATCTACAACGTAGTTGTAACCGCACACGCAT TTGTAATAATCTTCTTTATAGTAATACCAATCATAATTGGAGGGTTCGGTAACTGACTTG TTCCCCTAATAATTGGTGCTCCCGATATAGCATTTCCCCGAATAAATAATATAAGCTTCTG GACTTCTCCCTCCCTCATTCCTACTACTCCTCGCATCCTCTATAGTTGAAGCTGGGGCAG GAACAGGCTGAACCGTGTACCCTCCCTTAGCAGGCAACCTAGCCCATGCAGGAGCTTCAG TTGATCTAACCATTTTCTCTTTACACTTAGCAGGAGTTTCCTCAATTTTAGGAGCCATCA ACTTCATTACAACAATTATCAACATAAAGCCCCCGCAATGTCACAATACCAAACCCCTCTC TATTCGTATGATCCGTAATAATTACCGCCGTACTACTACTACTCTCGCTCCCTGTATTAG CAGCCGGCATCACAATGCTATTAACAGACCGGAACCTAAATACAACTTTCTTCGACCCGG CAGGAGGAGGAGATCCTATTCTATACCAACACTTATTCTGATTCTTTGGTCACCCGAAGTT TT	
SPMUKJ1	GATATCGGTACCCTTTCTATTGGTGCTTGGGCCGGTGGAACAGCTTTAAGCCTTCTAATT CGCGCTGGCCAACCCGGAACTCTGCTGGAGACGACCAAATCTACAACGTAGTTGTAACCG CACACGCATTTGTAATAATCTTCTTTATAGTAATACCAATCATAATTGGAGGGTTCGGTA ACTGACTTGTTCCCCTAATAATTGGTGCTCCCGATATAGCATTTCCCCGAATAAATAATA TAAGCTTCTGACTTCTCCCTCCCTCATTCCTACTACTCCTCGCATCCTCTATAGTTGAAG CTGGGGCAGGAACAGGCTGAACCGTGTACCCTCCCTTAGCAGGCAACCTAGCCCATGCAC GAGCTTCAGTTGATCTAACCATTTTCTCTTTACACTTAGCAGGAGTTTCCTCAATTTTAG GAGCCATCAACTTCATTACAACAATTATCAACATAAAGCCCCCGCAATGTCACAATACCAA AAACCCCTCTATTCGTATGATCCGTAATAATTACCGCCGTACTACTACTACTCTCGCTCC CTGTATTAGCAGCCGGCATCACAATGCTATTAACAGACCGGAACCTAAATACAACTTTCT TCGACCCGGCAGGAGGAGGAGATCCTATTCTAACCAACACTTATTCTGATTCTTTGGTCA CC	
SPMUBN	GATATCGGCACCCGTTCGTTTGGTGCCTGACTGGCATAGTAGGGACAGCCCTAAGCCTGC TGATTCGCGCCGAATTAGGTCAACCTGGGACCCTACTCGGAGATGACCAAATCTACAACG TAGTTGTAACCGCACACGCATTTGTAATAATTTTCTTCATAGTAATGCCAATCATAATTG GGGGATTTGGCAACTGACTTGTCCCTTTAATAATTGGTGCTCCCGACATAGCATTCCCCC GAATAAATAATATAAGCTTCTGACTCCTCCCTCTTTCCTACTACTTCTAGCATCATCAT TATAGTTGAAGCTGGAGCAGGGACAGGTTGAACCGTATACCCCCCTTTAGCAGGTAACC AGCCCACGCAGGAGCCTCGGTAGATCTAACCATCTTCTCTCTGCACTTAGCAGGTGTCT CTCAATTTTAGGGGCTATTAATTTTATTACAACAATTATCAATATAAAACCTCCTGCAA TACCACAATACCAAACCCCTTTATTCGTGTGATCCGTAATAATCACGCCGTACTACTACT CTCGACCCGGCAGGAGGAGGAGATCCTATTCTAACCAACACTTATTCTGATTCTTTGGTCA CC	
SPMUJBS1	TTTGGCCAAAAAGTCTAAGCAAGCTGGCATAGTAGGGACAGCCCTAAGCCTGCTGATTCG CGCCGAATTAGGTCAACCTGGGACCCTGCTCGGAGATGACCAAATCTACAACGTAGTTGT AACCGCACACGCATTTGTAATAATTTTCTTCATAGTAATGCCAATCATAATTGGGGGATT TGGCAACTGACTTGTCCCTCTAATAATTGGTGCTCCCGACATAGCATTCCCCCGAATAAA CAATATAAGCTTCTGACTCCTCCCTCCTCTTTCCTACTACTTCTAGCATCATCTATAGT TGAAGCTGGAGCAGGACAGGTTGAACCGTATACCCCCCTTTAGCAGGTAACCTAGCCCA CGCAGGAGCCTCGGTAGATCTAACCATCTTCTCTCTGCACTTAGCAGGTGTCTCCTCAAT TCTAGGGGCTATTAATTTTATTACAACAATTATCAATATAAAACCTCCTGCAATATCACA ATACCAAACCCCTTTATTCGTGTGATCCGTAATAATCACCGCCGTGCTATTACTCCTTTC ACTTCTGTGCTAACAGCTGGCATTACAATGCTACTACAGATCGAACCTAAATACAACAAC TTTCTTTGATCCAGCGGGGGGAGGAGATCCTATTCTATACCAACACTTATTTGATTCTT CGGCCACCCAGAAGTC	
SPMUPGS1	TTTTTGGGGCCCAGCACGGTGAGGGATAGTAGGAACAGCTTTAAGCCTTCTAATTCGCGC TGAATTAGGCCAACCCGGAACTCTGCTCGGAGACGACCAAATCTACAACGTAGTTGTAAC CGCACACGCATTTGTAATAATCTTCTTTATAGTAATACCAATCATAATTGGAGGGTTCGG TAACTGACTTGTTCCCCTAATAATTGGTGCTCCCGATATAGCATTCCCCGAATAAATAA TATAAGCTTCTGACTTCTCCCTCCCTCATTCCTACTACTCCTCGCATCCTCTATAGTTGA AGCTGGGGCAGGAACAGGCTGAACCGTGTACCCTCCCTTAGCAGGCAACCTAGCCCATGC AGGAGCTTCAGTTGATCTAACCATTTTCTCTTTACACTTAGCAGGAGTTTCCTCAATTTT AGGAGCCATCAACTTCATTACAACAATTATCAACATAAAGCCCCCGCAATGTCACAATA CCAAACCCCTCTATTCGTATGATCCGTAATAATTACCGCCGTACTACTACTACTCTCGCT CCCTGTATTAGCAGCCGGCATCACAATGCTATTAACAGACCGGAACCTAAATACAACTTT CTTCGACCCGGCAGGAGGAGGAGATCCTATTCTATACCAACACTTATTCTGATTCTTCGG CCACCCAGAAGTCAAG	
SPMUJB	GATATCGGCACCCGTTTGCTGTTTGGTGCCTGACTGGCATAGTAGGGACAGCCCTAAGCC TGCTGATTCGCGCCGAATTAGGTCAACCTGGGACCCTACTCGGAGATGACCAAATCTACA ACGTAGTTGTAACCGCACACGCATTTGTAATAATTTTCTTCATAGTAATGCCAATCATAA TTGGGGGATTTGGCAACTGACTTGTCCCTTTAATAATTGGTGCTCCCGACATAGCATTCC CCGAATAAACAATATAAGCTTCTGACTCCTCCCTCCTCTTTCCTACTACTTCTAGCAT CATCTATAGTTGAAGCTGGAGCAGGGACAGGTTGAACCGTATACCCCCCTTTAGCAGGTA ACCTAGCCCACGCAGGAGCCTCGGTAGATCTAACCATCTTCTCTCTGCACTTAGCAGGTG TCTCCTCAATTTTAGGGGCTATTAATTTTATTACAACAATTATCAATATAAAACCTCCTG CAATATCACAATACCAAACCCCTTTATTCGTGTGATCCGTAATAATCACCGCCGTGCTAT TACTCCTTTCACTTCCTGTGCTAGCAGCTGGCATTACAATGCTACTAACAGATCGAAACC TAAACACAACTTTCTTTGATCCAGCGGGGGGAGGAGATCCTATTTATACCAACACTTATG TTGATTCTTTGGTCACC	

Table 2: Continue...

Table 2: Continue...

SPMUKJ	TAAGCCTTCTAATTCGCGCTGAATTAGGGCCAACCCGGAACTCTGCTCGGAGACGACCAAA TCTACAACGTAGTTGTAACCGCACACGCATTTGTAATAATCTTCTTTATAGTAATACCAA TCATAATTGGAGGGTTCGGTAACTGACTTGTTCCCTAATAATTGGTGCTCCCGATATAG CATTTCCCCGAATAAATAATATAAGCTTCTGACTTCTCCCTCCCTCATTCCTACTACTCC TCGCATCCTCTATAGTTGAAGCTGGGGCAGGAACAGGCTGAACCGTGTACCCTCCCTTAG CAGGCAACCTAGCCCATGCAGGAGCTTCAGTTGATCTAACCATTTTCTCTTTACACTTAG CAGGAGTTTCCTCAATTTAGGAGCCATCAACTTCATTACAACAATTATCAACATAAAGC CCCCCGCAATGTCACAATACCAAACCCCTCTATTCGTATGATCCGTAATAATTACCGCCG TACTACTACTACTCTCGCTCCCTGTATTAGCAGCCGGCATCACAATGCTATTAACAGACC GGAACCTAAATACAACTTTCTTGACCCGGCAGGAGGAGGAGATCCTTCTACCAACACTTA TTCTGATTCTTTGGTCAC	
SPMUPP	TCATAAAGATATCGGCACCCGTTGCTGTTGTGCCTGAGCTGGCATAGTAGGGACAGCCCT AAGCCTGCTGATTCGCGCCGAATTAGGTCAACCTGGGACCCTACTCGGAGATGACCAAAT CTACAACGTAGTTGTAACCGCACACGCATTTGTAATAATTTCTTCATAGTAATGCCAAT CATAATTGGGGGATTTGGCAACTGACTTGTCCCTCTAATAATTGGTGCTCCCGACATAGC ATTCCCCCGAATAAACAATATAAGCTTCTGACTCCTCCCTCCCTCTTTCCTACTACTTCT AGCATCATCTATAGTTGAAGCTGGAGCAGGGACAGGTTGAACCGTATACCCCCCTTTAGC AGGTAACCTAGCCCACGCAGGAGCCTCGGTAGATCTAACCATCTTCTCTCGACTTAGC AGGGTGTCTCCTCAATTTTAGGGGCTATTAATTTTATTACAACAATTATCAATATAAAACC TCCTGCAATATCACAATACCAAACCCCTTTATTCGTGTGATCCGTAATAATCACCGCCGT GCTATTACTCCTTTCACTTCCTGTGCTAGCAGCTGGCATTACAATGCTACTAACAGATCG AAACCTAAACACAACTTTCTTTGATCCAGCGGGGGGAGGAGATCCTATACCAACACTTAT TTTGATTCTTTGGTCACC	
SPMUMU	TCACTAAAGATATCGGCACCCGTTGCTGTTGGTGCCTGAGCTGGCATAGTAGGGACAGCC CTAAGCCTGCTGATTCGCGCCGAATTAGGTCAACCTGGGACCCTACTCGGAGATGACCAA ATCTACAACGTAGTTGTAACCGCACACGCATTTGTAATAATTTCTTCATAGTAATGCCA ATCATAATTGGGGGATTTGGCAACTGACTTGTCCCTTAATAATTGGTGCTCCCGACATA GCATTCCCCCGAATAAACAATATAAGCTTCTGACTCCTCCCTCCCTCTTTCCTACTACTT CTAGCATCATCTATAGTTGAAGCTGGAGCAGGGACAGGTTGAACCGTATACCCCCCTTTA GCAGGTAACCTAGCCCACGCAGGAGCCTCGGTAGATCTAACCATCTTCTCTCGCACTTA GCAGGTGTCTCCTCAATTTTAGGGGCTATTAATTTTATTACAACAATTATCAATATAAAA CCTCCTGCAATATCACAATACCAAACCCCTTTATTCGTGTGATCCGTAATAATCACCGCC GTGCTATTACTCCTTTCACTTCCTGTGCTAGCAGCTGGCATTACAATGCTACTAACAGAT CGAAACCTAAACACAACTTTCTTTGATCCAGCGGGGGGAGGAGATCCTATTATACCAACA CTTATGTTGATTCTTTGGTCACC	
SPMUGIR	TCCTTGGGGCCGGTAGGAACAGCTTTAAGCCTTCTAATTCGCGCTGAATTAGGCCAACCC GGAACTCTGCTCGGAGACGACCAAATCTACAACGTAGTTGTAACCGCACACGCATTTGTA ATAATCTTCTTTATAGTAATACCAATCATAATTGGAGGGTTCGGTAACTGACTTGTTCCC CTAATAATTGGTGCTCCCGATATAGCATTTCCCCGAATAAATAATATAAGCTTCTGACTT CTCCCTCCCTCATTCCTACTACTCCTCGCATCCTCTATAGTTGAAGCTGGGGCAGGAACA GGCTGAACCGTGTACCCTCCCTTAGCAGGCAACCTAGCCCATGCAGGAGCTTCAGTTGAT CTAACCATTTTCTCTTTACACTTAGCAGGAGTTTCCTCAATTTTAGGAGCCATCAACTTC ATTACAACAATTATCAACATAAAGCCCCCCGCAATGTCACAATACCAAACCCCTCTATTC GTATGATCCGTAATAATTACCGCCGTACTACTACTACTCTCGCTCCCTGTATTAGCAGCC GGCATCACAATGCTATTAACAGACCGGAACCTAAATACAACTTTCTTCGACCCGGCAGGA GGAGGAGATCCTATTCTATACCAACACTTATTCTGATTCTTTGGTCACCCGAAATTTAAA AAAAACCCAATATCTTTATGATTATA	

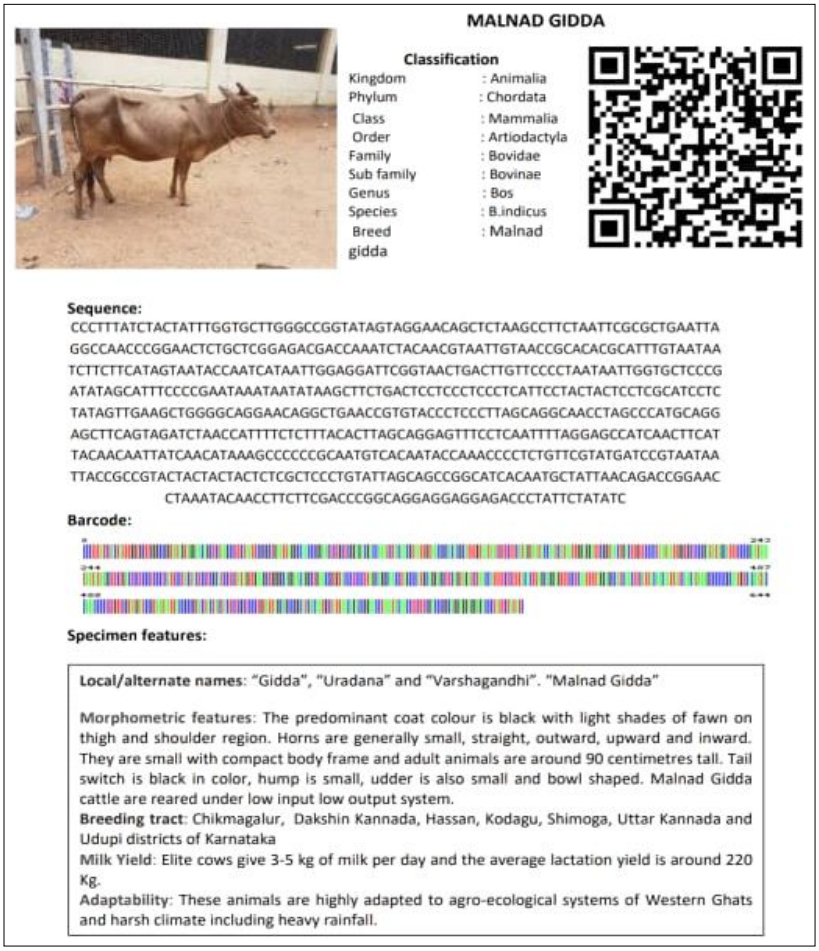


Fig 5: QR code scan-output page-showing the characteristics of a breed.

nucleotide polymorphisms associated with thermal tolerance traits has contributed to marker-assisted selection strategies in bovines (Saikia *et al*., 2019). These studies complement the present COX-I barcoding approach by illustrating how multiple molecular markers collectively strengthen genetic documentation and conservation planning.

The deposition of validated sequences in the Barcode of Life Data Systems (Ratnasingham and Hebert, 2007) enhances global accessibility, reproducibility and long-term

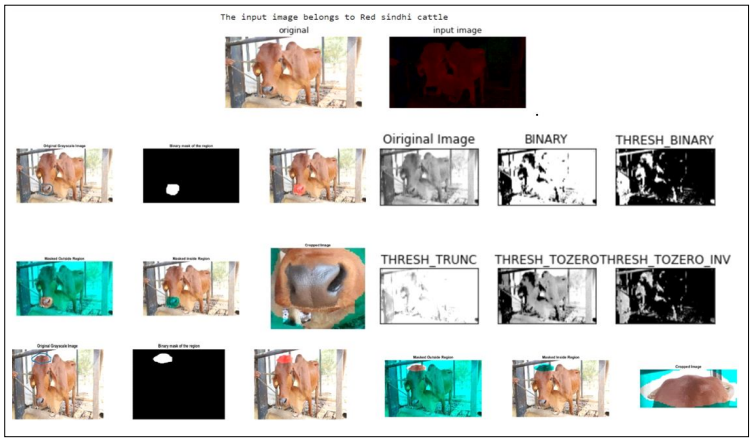


Fig 6: Input Image, its identification and extracting features individually for each part.

preservation of genetic data. Creating a structured barcode repository for Indian cattle breeds contributes significantly to biodiversity documentation and establishes a reference framework for future taxonomic, phylogenetic and breeding studies.

In addition to molecular tools, the incorporation of image-based classification improves phenotypic validation and automation of breed recognition. Machine learning and deep learning algorithms have demonstrated strong performance in classifying cattle breeds using morphological descriptors extracted from digital images (Hussain *et al*., 2020). Advances in drone-assisted imaging and computer vision technologies have further expanded opportunities for real-time herd monitoring and large-scale livestock assessment (Kim *et al*., 2019). In the present study, RGB decomposition and deep feature extraction enabled precise identification of anatomical regions such as horns and muzzle patterns, improving discrimination accuracy while reducing background interference.

The integration of gene-based QR codes provides an innovative layer of digital traceability. By linking DNA barcode data, morphological features and breed-specific information into a scannable format, this system facilitates rapid on-site verification through mobile devices. Such a digital interface supports efficient livestock record management, authentication processes and transparency in breeding programs.

Overall, the convergence of mitochondrial DNA barcoding, molecular data archiving, image analytics and QR-based digital tagging offers a comprehensive strategy for the authentication and conservation of bovine genetic resources. This study presents a unified model for molecular authentication and digital traceability of Indian cattle breeds using DNA barcoding, gene-based QR codes and image recognition. COX-I barcodes for sixteen breeds were generated and submitted to BOLD, creating India's first Bovine DNA Barcode Directory. QR-linked genomic and phenotypic data enable instant verification, while image analytics achieved over 95% classification accuracy. The framework supports efficient livestock management, breed conservation and national digital agriculture initiatives.

CONCLUSION

This study presents a practical approach for accurately identifying Indian cattle breeds by combining DNA barcoding, QR code technology and image analysis. Using the mitochondrial COX-I gene, DNA barcodes were successfully generated and authenticated for sixteen bovine breeds and stored in the BOLD database. The integration of QR codes allows easy access to genetic and phenotypic information through simple scanning. Image analysis further strengthens breed identification by examining unique physical traits such as horns and muzzle patterns. Together, these tools create a reliable and user-friendly system for breed authentication. This integrated framework can support better livestock management, conservation of indigenous cattle and improved breeding strategies in the future.

ACKNOWLEDGEMENT

The present study was supported by National Kamadhenu Breeding Centre, NKBC. The authors sincerely acknowledge Late Prof. S. Jyothi, SPMVV for her kind support in carrying out Image Analysis.

Disclaimers

The views and conclusions expressed in this article are solely those of the authors and do not necessarily represent the views of their affiliated institutions. The authors are responsible for the accuracy and completeness of the information provided, but do not accept any liability for any direct or indirect losses resulting from the use of this content.

Informed consent

All animal procedures for experiments were approved by the Committee of Experimental Animal care and Handling Techniques were approved by the University of Animal Care Committee.

Conflict of interest

The authors declare that there are no conflicts of interest regarding the publication of this article. No funding or sponsorship influenced the design of the study, data collection, analysis, decision to publish, or preparation of the manuscript.

REFERENCES

Alam, A., Chadha, N.K., Kumar, A.P., Chakraborty, S.K., Joshi, K.D., Sawant, P.B., Das, S.C.S., Kumar, J. and Kumar, T. (2020). DNA barcoding and biometric investigation on the invasive *Oreochromis niloticus* (Linnaeus, 1758) from the River Yamuna of Uttar Pradesh. *Indian Journal of Animal Research.* **54(7):** 856-863. doi: 10.18805/ijar.B-3833.

Altschul, S.F., Madden, T.L., Schäffer, A.A., Zhang, J., Zhang, Z., Miller, W. and Lipman, D.J. (1997). Gapped BLAST and PSI-BLAST: A new generation of protein database search programs. *Nucleic Acids Research.* **25(17):** 3389-3402. https://doi.org/10.1093/nar/25.17.3389.

Folmer, O., Black, M., Hoeh, W., Lutz, R. and Vrijenhoek, R. (1994). DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology.* **3(5):** 294-299.

https://www.barcoding.si.edu (Barcode of Life Data Systems (2024). BOLD Systems..

https://agriportal.cg.nic.in/.

https://www.data.gov.in/catalog/20th-livestock-census.

Hajibabaei, M., Smith, M.A., Janzen, D.H., Rodriguez, J.J., Whitfield, J.B. and Hebert, P.D.N. (2006). A minimalist barcode can identify a specimen whose DNA is degraded. *Molecular Ecology Notes*. **6(4):** 959-964. https://doi.org/10.1111/j.1471-8286.2006.01470.x.

Hebert, P.D.N., Cywinska, A., Ball, S.L. and deWaard, J.R. (2003). Biological identifications through DNA barcodes. *Proceedings of the Royal Society B: Biological Sciences*. **270(1512):** 313-321.

Hussain, A., Sun, D.W. and Cheng, J.H. (2020). Deep learning-based classification of cattle breeds using image processing techniques. *Computers and Electronics in Agriculture*. **175:** 105533. https://doi.org/10.1016/j.compag.2020.105533.

Kim, J.S., Lee, S.H. and Park, J.H. (2019). Drone-based image analysis for livestock monitoring and breed identification using deep learning algorithms. *Sensors.* **19(22):** 5036. https://doi.org/10.3390/s19225036.

Larcombe, S.D., Capewell, P., Jensen, K., Weir, W., Kinnaird, J., Glass, E.J. and Shiels, B.R. (2022). Susceptibility to disease (tropical theileriosis) is associated with differential expression of host genes that possess motifs recognised by a pathogen DNA binding protein. *PLoS One*. **17(1):** e0262051. doi: 10.1371/journal.pone.0262051. PMID: 35061738; PMCID: PMC8782480.

Ratnasingham, S. and Hebert, P.D.N. (2007). Bold: The barcode of life data system. *Molecular Ecology Notes*. **7(3):** 355-364.

Saikia, J., Verma, A., Gupta, I.D., Hazarika, D., Deshmukh, B. and Das, R. (2019). Novel SNP identified in HSBP1 gene and its association with thermal tolerance traits in murrah buffalo. *Indian Journal of Animal Research.* **54(3):** 282-285. doi: 10.18805/ijar.B-3767.

Sarang, S.K., Sreekumar, D. and Sejian, V. (2024). Indigenous cattle biodiversity in India: Adaptation and conservation. *Reproduction and Breeding*. **4(4):** 254-266. https://doi.org/10.1016/j.repbre.2024.09.001.

Sarızeybek, A., Tezcan, A. and Işık, A. (2023). Detection of bovine species on image using machine learning classifiers. *Gazi University Journal of Science*. **37:** 1-15. https://doi.org/10.35378/gujs.1203685.

Singh, R., Gurao, A., Mishra, S.K., Niranjan, S.K., Vohra, V., Mukesh, M., Rajesh, C. and Kataria, R.S. (2024). Molecular characterization of the coding region and 5' UTR of *HSP70* gene in Indian riverine buffalo breeds. *Indian Journal of Animal Research.* **58(2):** 196-199. doi: 10.18805/IJAR.B-4423.

Smith, A.M., McKeown, P.C. and Reilly, A. (2018). Application of DNA barcoding for the authentication of beef products and detection of species substitution. *Food Control.* **92:** 59-67. https://doi.org/10.1016/j.foodcont.2018.04.018.

Sultana, S., Ali, M.E., Hossain, M.A., Naquiah, N. and Zaidul, I.S.M. (2017). DNA barcoding: A powerful tool for species identification. *Bangladesh Journal of Agricultural Research.* **42(1):** 1-11.

Tamura, K., Stecher, G. and Kumar, S. (2021). MEGA11: Molecular evolutionary genetics analysis. *Molecular Biology and Evolution.* **38(7):** 3022-3027. https://doi.org/10.1093/molbev/msab120.

Utami, S., Jamil, A., Reviany, N.V., Muhlis, N., Haris, P., Dhani, P., Arif, A.N.M., Indrasari, S. and Chairul, N.A. (2024). Genetic diversity of crossbred cattle using cytochrome oxidase subunit I (COI) gene in South Sulawesi, Indonesia. *Indian Journal of Animal Research.* **58(1):** 28-34. doi: 10.18805/IJAR.BF-1600.

Yang, F., Ding, F., Chen, H., He, M., Zhu, S., Ma, X., Jiang, L. and Li, H. (2018). DNA barcoding for the identification and authentication of animal species in traditional medicine. *Evidence-Based Complementary and Alternative Medicine*. pp 5160254. https://doi.org/10.1155/2018/5160254.

Zhao, Y., Zhang, H., Liu, X. and Wang, J. (2021). Mitochondrial DNA diversity and population structure analysis of Chinese indigenous cattle breeds. *Animals.* **11(4):** 1033. https://doi.org/10.3390/ani11041033.