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**Ilayaraja Selvaraj**

Wildlife SOS, Mathura, Uttar  
Pradesh, India

**Arun A Sha**

Wildlife SOS, Mathura, Uttar  
Pradesh, India

**E Gochalan**

Wildlife SOS, Mathura, Uttar  
Pradesh, India

**Rahul Prasad**

Wildlife SOS, Mathura, Uttar  
Pradesh, India

**Pramod Rajpoot**

Wildlife SOS, Mathura, Uttar  
Pradesh, India

**Lalit K Jangid**

Wildlife SOS, Mathura, Uttar  
Pradesh, India

**Corresponding Author:**

**Ilayaraja Selvaraj**

Wildlife SOS, Mathura, Uttar  
Pradesh, India

## Pulmonary tuberculosis with secondary aspergillosis in a captive Asian elephant (*Elephas maximus*): A case report

**Ilayaraja Selvaraj, Arun A Sha, E Gochalan, Rahul Prasad, Pramod  
Rajpoot and Lalit K Jangid**

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### Abstract

Tuberculosis (TB) remains a significant re-emerging zoonotic disease in captive elephants, characterized by diagnostic complexity, chronic progression, and substantial welfare implications. The present case report describes pulmonary tuberculosis caused by *Mycobacterium orygis* with concurrent secondary aspergillosis in a 15-year-old rehabilitated female Asian elephant (*Elephas maximus*). The animal exhibited progressive weight loss, anorexia, gastrointestinal disturbances, and severe respiratory compromise. Hematobiochemical evaluation revealed anemia, systemic inflammation, and multi-organ dysfunction. Serological testing (Mycopac Dual Kit) indicated exposure to *Mycobacterium tuberculosis* complex (MTBC), while conventional trunk wash cultures remained negative, highlighting diagnostic limitations. Post-mortem findings revealed extensive pulmonary cavitation and caseous granulomatous lesions. Histopathology confirmed caseo-calcified granulomas with acid-fast bacilli, and molecular analysis identified *M. orygis*. Concurrent fungal colonization (*Aspergillus* spp.) within cavitory lesions emphasized the role of opportunistic infections in advanced disease. This case underscores the critical need for early diagnostic surveillance, integration of serological and molecular tools, and proactive clinical management. From a welfare perspective, it highlights the profound impact of delayed diagnosis and chronic disease burden in captive elephants, reinforcing the necessity for structured health monitoring protocols in managed populations.

**Keywords:** Asian elephant, tuberculosis, opportunistic infection, diagnostics

### 1. Introduction

Tuberculosis in captive elephants represents a complex intersection of infectious disease, public health concern, and animal welfare challenge. Since its recognition as a re-emerging zoonotic condition, TB has posed persistent difficulties in early detection, clinical management, and containment within captive populations (Mikota *et al.* 2000; Mikota 2008) [6, 5]. Elephants often present with non-specific, insidious clinical signs, including gradual weight loss, anorexia, intermittent respiratory distress, and behavioural changes frequently leading to delayed diagnosis and advanced disease at presentation (Fowler & Mikota 2006; Lyashchenko *et al.* 2006) [2, 4].

Pathologically, tuberculosis is characterized by granuloma formation, caseation, and progressive pulmonary destruction (Mikota *et al.* 2000) [6]. Chronic infection compromises host immunity, predisposing affected individuals to secondary opportunistic infections, notably aspergillosis, which further exacerbates respiratory dysfunction (Tell 2005; Beernaert *et al.* 2010) [7, 1].

Despite increasing recognition of tuberculosis in elephants, documented cases involving concurrent fungal infections remain limited, particularly in the Indian context. Such coinfections not only complicate diagnosis and treatment but also significantly worsen clinical outcomes.

This case report aims to present a clinically and pathologically comprehensive account of pulmonary tuberculosis with secondary aspergillosis, while emphasizing the critical welfare implications, diagnostic gaps, and need for early intervention strategies in captive elephant management

## Methodology

A 15-year-old female Asian elephant, previously used in circus conditions and subsequently rehabilitated at the Elephant Conservation and Care Centre (Wildlife SOS, India), was monitored longitudinally. Clinical observations began in June 2023 with progressive weight loss and osteoarthritic changes. Body weight was recorded using a calibrated electronic scale, and body condition scoring (BCS) was conducted following established visual assessment criteria (Wijeyamohan *et al.* 2014) <sup>[10]</sup>. By November 2024, the animal developed gastrointestinal disturbances, anorexia, and progressive systemic decline. In January 2025, advanced respiratory distress, mucopurulent trunk discharge, and behavioural signs of discomfort were evident.

Comprehensive hematological and biochemical analyses were performed. Serological testing for MTBC was conducted using the MycoPac Dual Kit (CisGEN Biotech Discoveries Pvt. Ltd., Tamil Nadu, India), while repeated triple trunk wash cultures were attempted in accordance with standard tuberculosis surveillance protocols (USAHA 2012) <sup>[9]</sup>.

Despite intensive clinical management, the animal succumbed to disease progression. A full necropsy was conducted, with tissue samples submitted for histopathology, Ziehl–Neelsen

staining, molecular diagnostics, and fungal culture.

## Results and Discussion

### Clinical progression and systemic decline

The elephant demonstrated a marked decline in body condition, with weight reduction from 3440 kg (BCS 7/10; Fig. 1a) to 2510 kg (BCS 4/10; Fig. 1b), reflecting chronic disease progression and sustained metabolic compromise. Hematobiochemical findings indicated progressive anemia, significant hepatic enzyme elevation, renal compromise and systemic inflammatory response. These findings collectively suggest multi-organ involvement, consistent with chronic infectious disease and prolonged physiological stress. The detailed hematobiochemical parameters are presented in Table 1.

Repeated triple trunk wash cultures were negative, consistent with previous reports highlighting the limited sensitivity of trunk wash culture in early or latent stages of tuberculosis in elephants (Mikota *et al.* 2000; Greenwald *et al.* 2009) <sup>[3, 6]</sup>. In contrast, serological testing demonstrated strong reactivity (Fig. 2), supporting the growing evidence that serology and molecular diagnostics are essential tools for early and reliable detection (Lyashchenko *et al.* 2006) <sup>[4]</sup>.

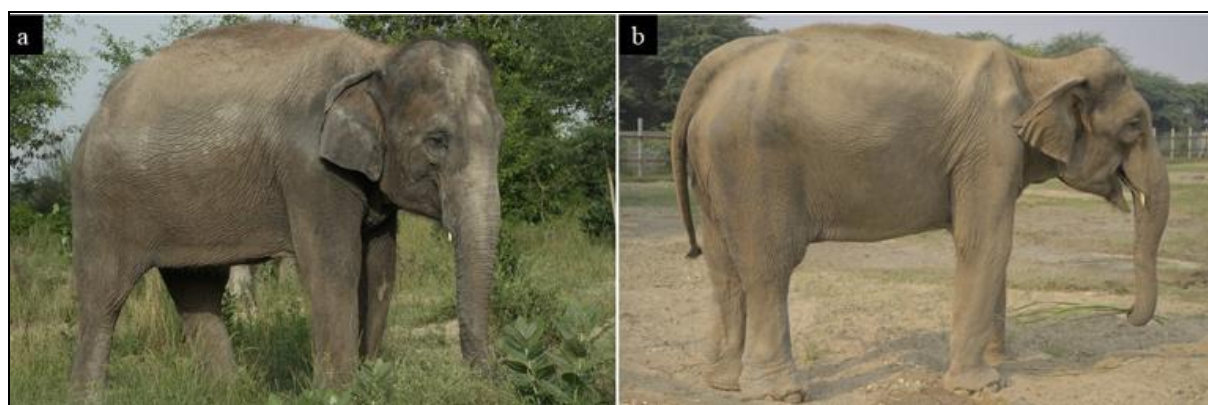


**Fig 1:** Longitudinal decline in body condition score

**Table 1:** Comparative haematological and serum biochemical parameters of the affected *E. maximus*

| Measured (unit)                                   | Results   |               |
|---------------------------------------------------|-----------|---------------|
|                                                   | June 2023 | November 2024 |
| Red blood cell count (Millions/cm m)              | 3.82      | 3.16          |
| Mean corpuscular volume (fL)                      | 113.0     | 113.1         |
| Mean corpuscular haemoglobin (pg)                 | 39.7      | 42.0          |
| Mean corpuscular haemoglobin concentration (g/dL) | 35.2      | 37.1          |
| Packed cell volume (%)                            | 43.1      | 35.8          |
| Haemoglobin (gm %)                                | 15.2      | 13.3          |
| Total leucocyte count (/cu mm)                    | 12200     | 15200         |
| Neutrophils (%)                                   | 70        | 74            |
| Lymphocytes (%)                                   | 28        | 26            |
| Monocytes (%)                                     | 02        | 00            |
| Eosinophils (%)                                   | 00        | 00            |
| Platelet count (per cu mm)                        | 517000    | 448000        |
| Erythrocyte sedimentation rate (mm/1st hr.)       | 40        | 42            |
| Iron (µg/dl)                                      | 85        | 83            |
| C-reactive protein (mg/l)                         | 0.5       | 1.5           |
| Total bilirubin (mg %)                            | 0.3       | 0.4           |
| Aspartate aminotransferase (U/L)                  | 39        | 254           |
| Alanine aminotransferase (U/L)                    | 3.0       | 14            |
| Alkaline phosphatase (U/L)                        | 61        | 61            |
| Total protein (gm %)                              | 7.5       | 8.2           |
| Albumin (gm %)                                    | 1.4       | 1.6           |
| Globulin (gm %)                                   | 6.1       | 6.6           |
| Gamma-glutamyl transferase (U/L)                  | 12        | 26            |
| Lactate dehydrogenase (U/L)                       | 2119      | 2324          |
| Blood urea nitrogen (mg %)                        | 21.8      | 22.4          |
| Serum chloride (mmol/L)                           | 79        | 81            |

|                           |     |     |
|---------------------------|-----|-----|
| Blood sugar random (mg %) | 45  | 47  |
| Triglycerides (mg %)      | 21  | 40  |
| Blood urea (mg %)         | 15  | 48  |
| Creatinine (mg %)         | 1.0 | 1.5 |
| Uric acid (mg %)          | 0.2 | 0.3 |
| Sodium (mmol/L)           | 127 | 127 |
| Potassium (mmol/L)        | 4.7 | 6.5 |
| Calcium (mg/dl)           | 8.9 | 8.4 |
| Phosphorus (mg/dl)        | 4.8 | 4.5 |

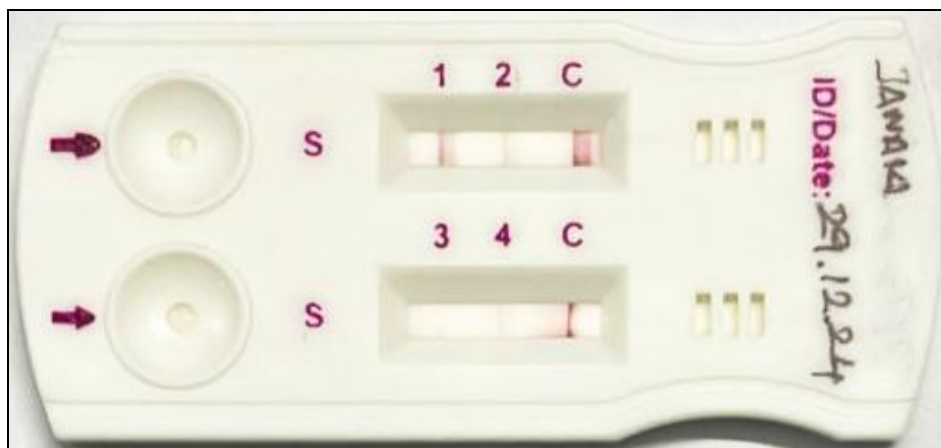


**Fig 1:** MycoPac Dual Kit showing seropositivity to *Mycobacterium tuberculosis* complex (MTBC) characterized by a high-intensity T1 band and low-intensity T2–T4 bands

### Therapeutic Limitations

Despite initiation of anti-tubercular therapy (INH and PZA), clinical response was poor. This highlights a critical reality that in elephants, therapeutic success is heavily dependent on early diagnosis. Advanced-stage disease is often refractory to treatment (Fowler & Mikota 2006) [2]. Challenges such as poor drug acceptance and advanced pathology further complicate management.

**Pathological findings and disease severity:** Post-mortem

examination revealed extensive pulmonary cavitation, diffuse caseous granulomas, severe pulmonary destruction (Fig. 3). Cytological examination of impression smears further demonstrated clusters of acid-fast bacilli (Fig. 4), histopathology confirmed chronic active tuberculosis, while PCR identified *Mycobacterium orygis*, an emerging pathogen of regional importance. The presence of *Aspergillus* spp. within necrotic lung cavities is particularly significant, indicating severe immunosuppression, advanced disease stage and compounded respiratory dysfunction.

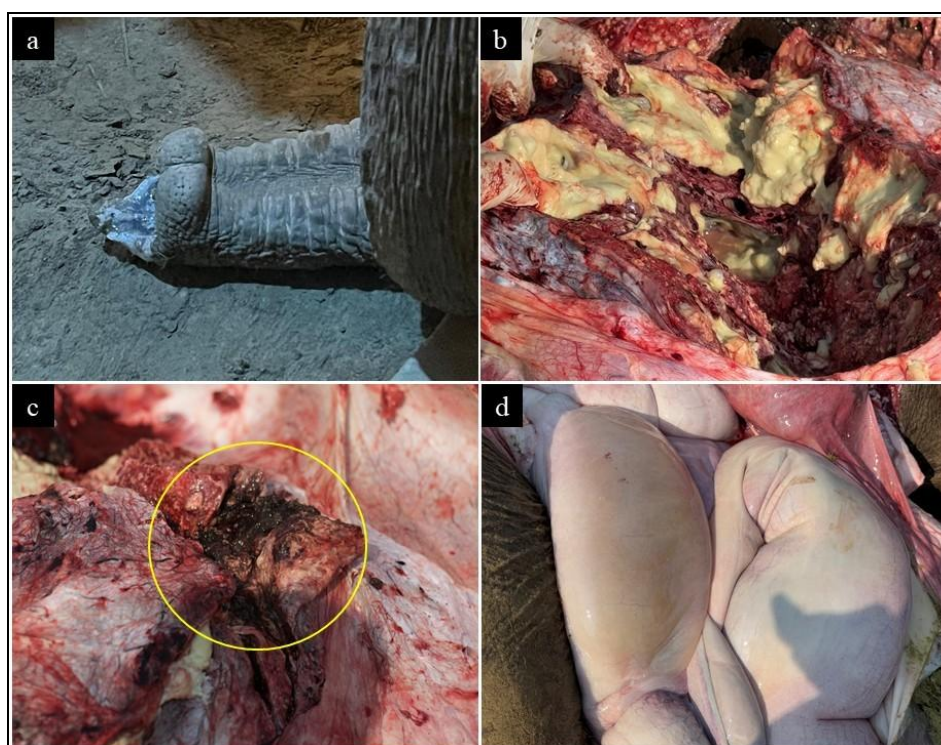


**Fig 2:** (a) Antemortem mucopurulent trunk discharge; (b) multiple pulmonary cavitory lesions containing thick purulent material; (c) blackish, fine, coffee powder–like material (marked with yellow circle) within a pulmonary cavity; (d) oedematous intestinal walls

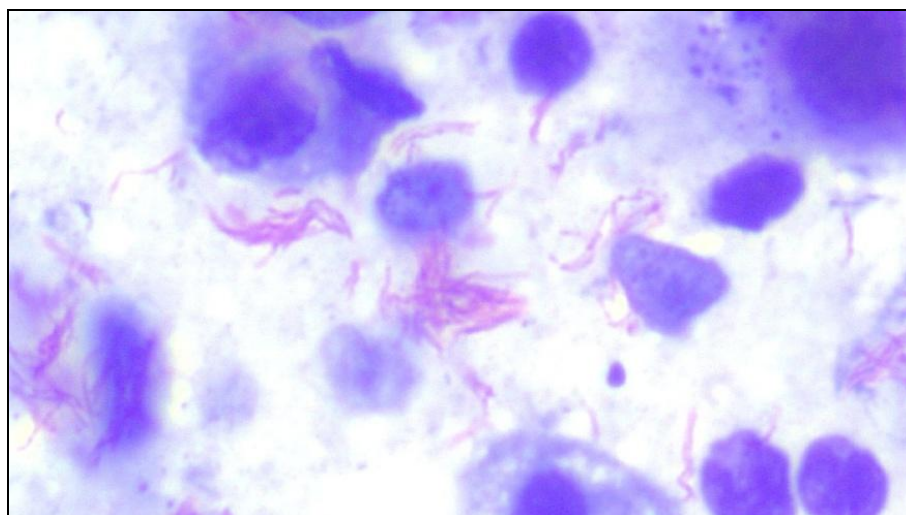


**Fig 4:** Ziehl–Neelsen-stained lung impression smear showing acid-fast bacilli

### Welfare Interpretation

This case exemplifies the silent progression of chronic disease in captive elephants, where clinical signs often become evident only at advanced stages. Key welfare concerns include; prolonged undetected suffering, chronic systemic stress, reduced quality of life due to respiratory compromise and metabolic decline and co-morbid conditions such as osteoarthritis likely contributed to immunosuppression, further accelerating disease progression.

### Conclusion

This case highlights pulmonary tuberculosis caused by *Mycobacterium orygis*, complicated by secondary aspergillosis, in a captive Asian elephant. The findings emphasize: 1) the chronic and insidious nature of tuberculosis in elephants, 2) the limitations of conventional diagnostics, 3) the critical role of opportunistic infections in disease severity, and 4) the urgent need for early, proactive disease surveillance systems.

### Key Recommendations

Key recommendations for veterinary and management practice include: 1) implementation of routine TB surveillance using serological and molecular tools, 2) development of standardized health monitoring protocols for captive elephants, 3) early identification and management of co-morbid conditions, 4) strengthening biosecurity and zoonotic risk mitigation measures, and 5) adoption of a preventive, welfare-centered veterinary approach.

### Final impact statement

Early detection is not merely a diagnostic goal-it is a welfare obligation. In captive elephants, timely intervention defines the difference between managed disease and prolonged, silent suffering.

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