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## ACCEPTED MANUSCRIPT

# Stable infection of a bovine mammary epithelial cell line (MAC-T) with bovine leukemia virus (BLV)

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### Highlights:

- Bovine mammary epithelial cell line is susceptible to infection with BLV.
- BLV infection increases TNF-α expression on infected cells.
- Infected mammary epithelial cell lines expresses BLV actively.

#### **Abstract**

Bovine leukemia virus (BLV) is a retrovirus that affects cattle causing a lymphoproliferative disease. BLV infection has been associated with misbalance of the immune response causing a higher incidence of other infections. Mastitis is one of the most important conditions that affect milk production in cattle. The aim of this study was to stably infect a bovine mammary epithelial cell line (MAC-T). MAC-T cell line was successfully infected with BLV and the infection was confirmed by nested PCR, qPCR, immunocytochemistry, western blot and transmission electron microscopy. This is the first report of a bovine mammary epithelial cell line stably infected with BLV. This new cell line could be used as an *in vitro* model to study the effect of BLV on the immune response in the mammary gland and the relationship with other agents causing mastitis.

#### Abbreviations:

BLV Bovine leukemia virus; MAC-T Bovine mammary epithelial cell line; EBL Enzootic bovine leukosis; BHV-1 Bovine Herpesvirus 1; BHV-4 Bovine Herpesvirus 4; BVDV Bovine viral diarrhea virus; FDM Foot and Mouth Disease; PBMCs peripheral blood mononuclear cells; FLK fetal lamb kidney; HPL high proviral load; p.p.i passage post-infection

**Keywords:** Bovine Leukemia Virus; MAC-T; Infection; Mastitis; TNF-α1. Introduction

Bovine leukemia virus (BLV) is a  $\delta$ -retrovirus responsible for Enzootic Bovine Leukosis (EBL), a lymphoproliferative disease that affects cattle. With a size of 60 to 125 nm, this enveloped virus naturally infects B-lymphocytes causing a chronic infection. Its genomic organization is typical of

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