

# HIV PROVIRAL REACTIVATION IS IMPAIRED IN EFFECTOR MEMORY CD4+ T CELLS BY IMMUNOMETABOLIC REPROGRAMMING WITH DASATINIB

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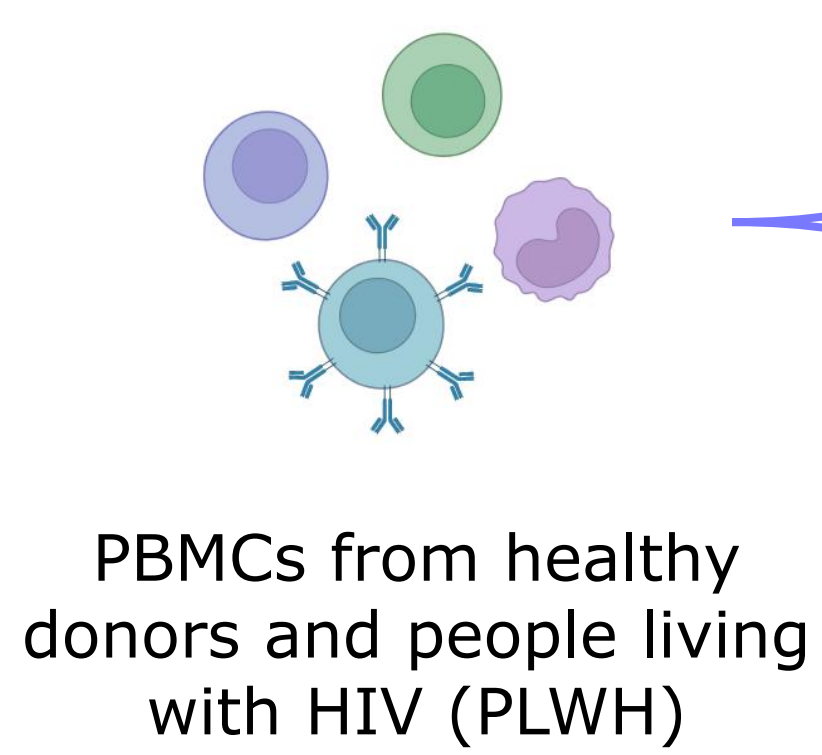
## BACKGROUND

- HIV selectively infects CD4+ T lymphocytes with high metabolic activity.
- TEM and TEMRA subpopulations**, which are essential for the maintenance of the reservoir, present a higher metabolic activity than the naive CD4+ lymphocytes and central memory (TCM).
- Dasatinib** is a tyrosine kinase inhibitor (TKI) that interferes with HIV infection in CD4 cells and macrophages.

## OBJECTIVE

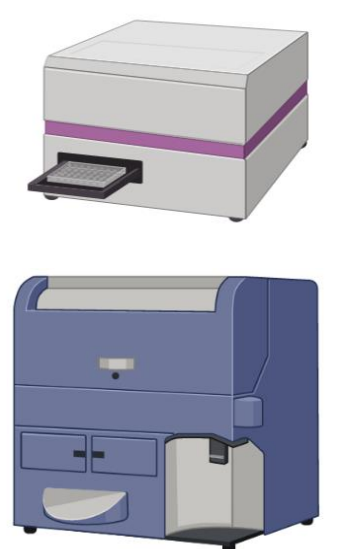
To evaluate the use of Dasatinib as a **proviral latency-promoting agent (LPA)** by reducing the metabolic activity of viable CD4+ T cells to interfere with reservoir maintenance

## METHODS



Phosphoproteome in presence and absence of Dasatinib → Liquid chromatography followed by tandem mass spectrometry in orbitrap  
 Proviral reactivation in presence and absence of Dasatinib  
 Analysis of metabolic activity in presence and absence of Dasatinib and an activating stimulus

- Myochondrial ATP and culture medium pH measurement
- Enzyme activity measurement by ELISA
- Measurement of GLUT-1 expression and 2-NBDG probe uptake
- Distribution of T lymphocytes memory subpopulations

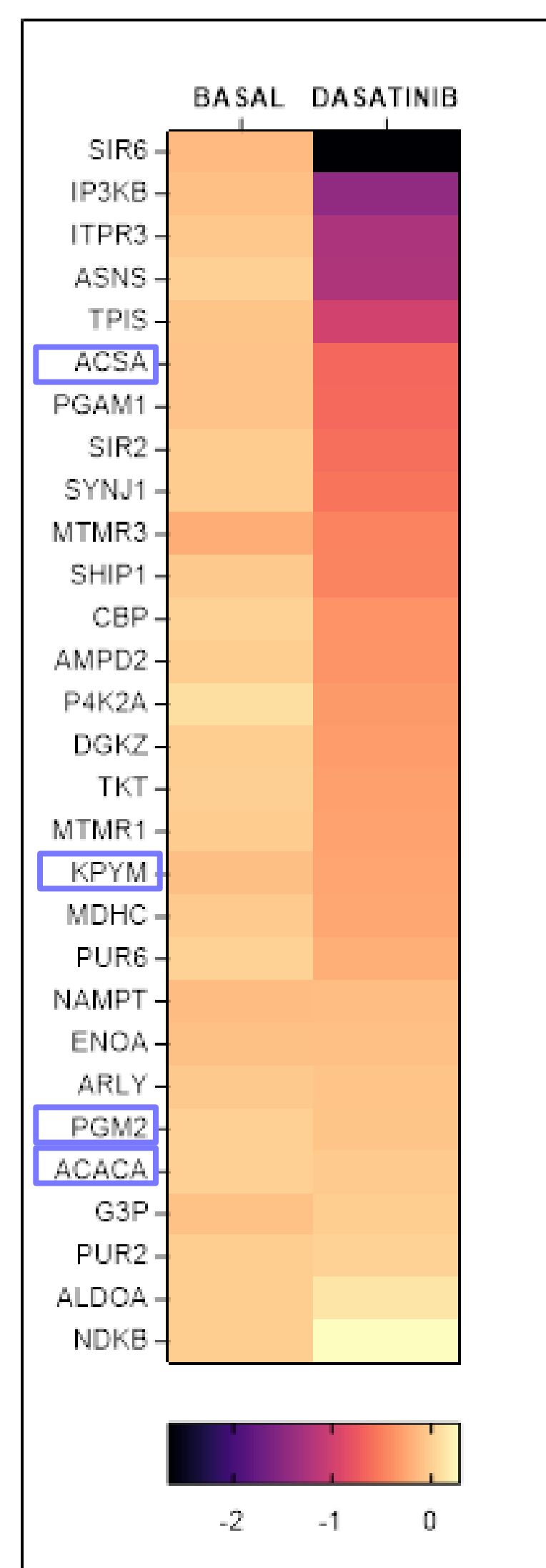


## RESULTS

### 1. Dasatinib modified the phosphorylation of >130 proteins involved in metabolic pathways.

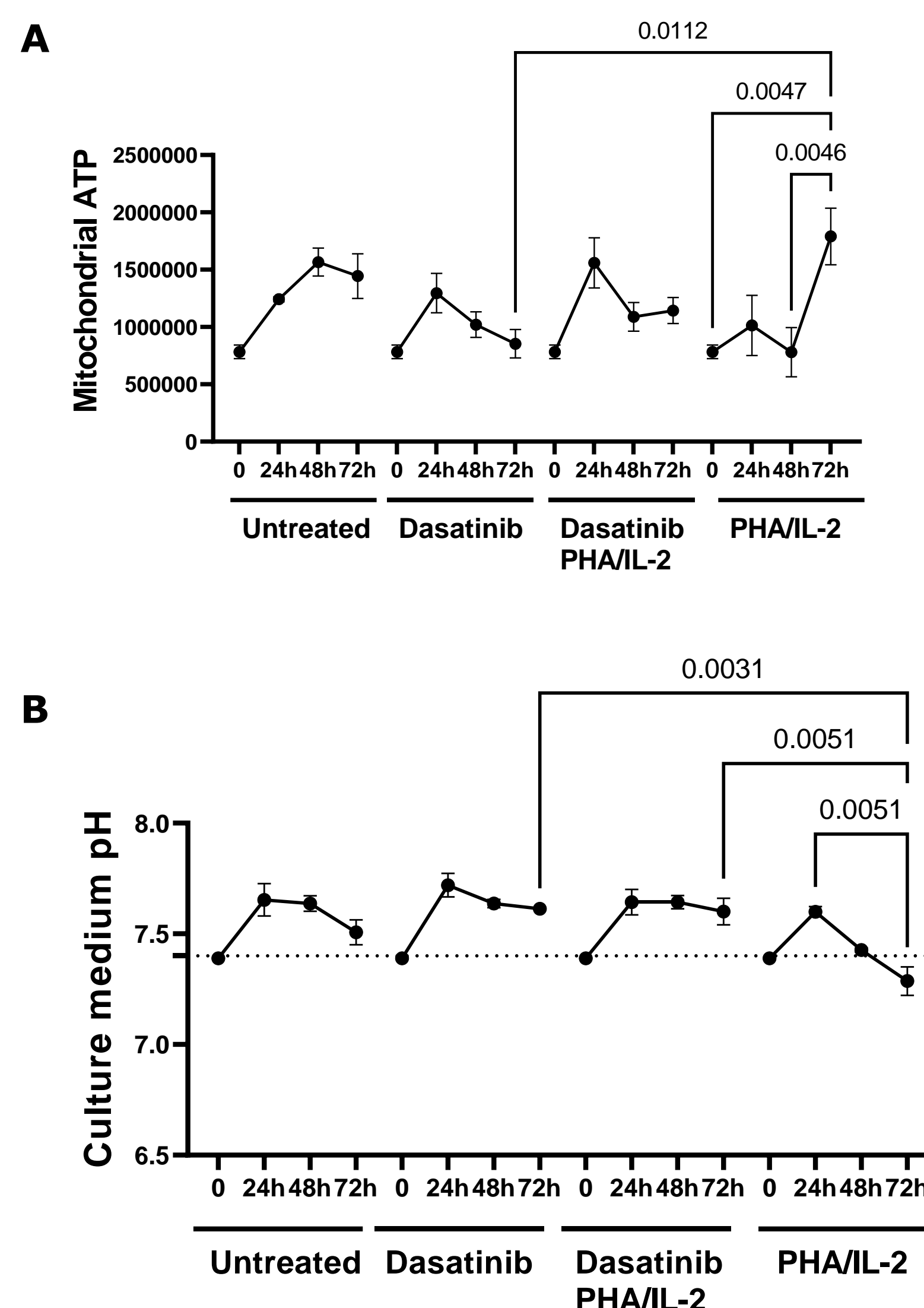
#### Deregulated proteins

- Glycolysis/gluconeogenesis
- Pyruvate metabolism
- Pentose phosphate pathway
- Inositol phosphate metabolism
- Glucagon signaling pathway



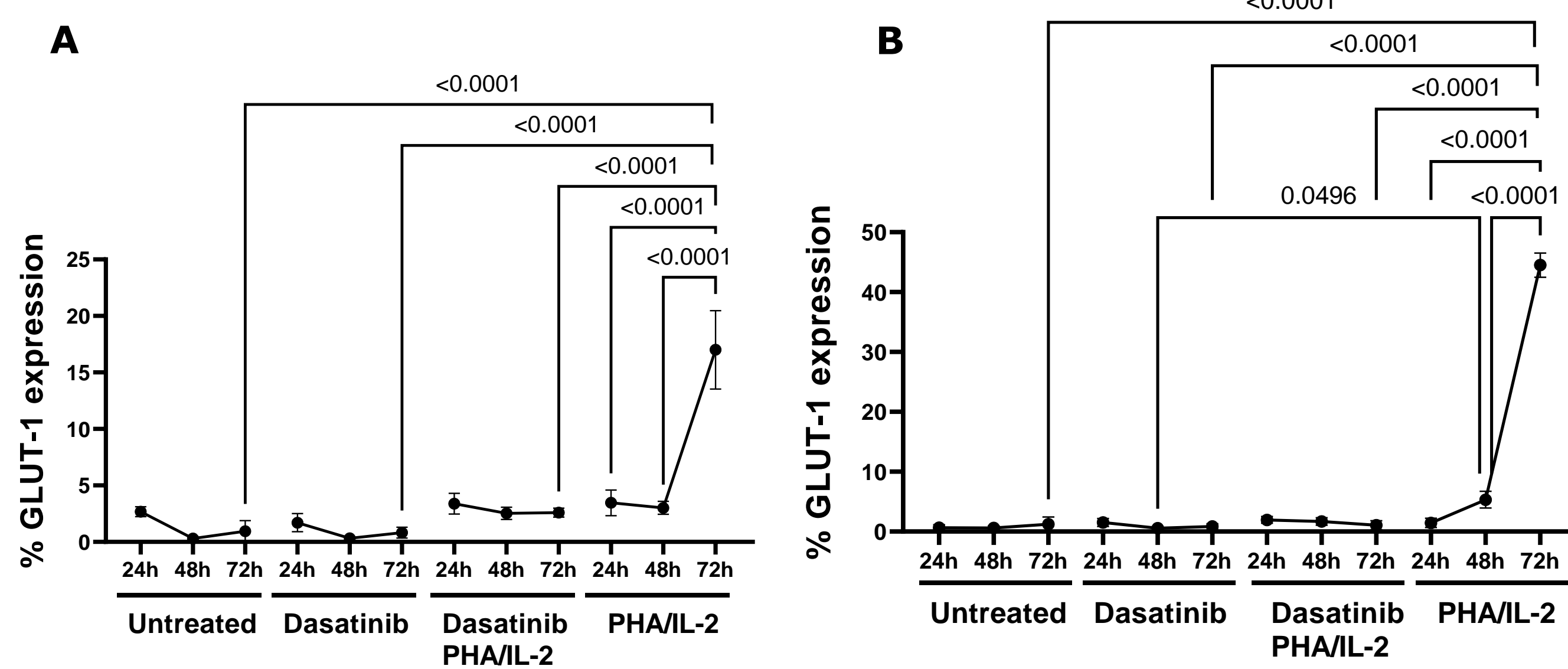
### 3. Dasatinib interferes with the metabolic activity of PBMCs

- In absence of dasatinib, there is an increase in mitochondrial ATP production. Dasatinib interferes with mitochondrial ATP production in response to an activating stimulus.
- In absence of Dasatinib, acidification of the pH of the culture medium occurs due to increased mitochondrial respiration.



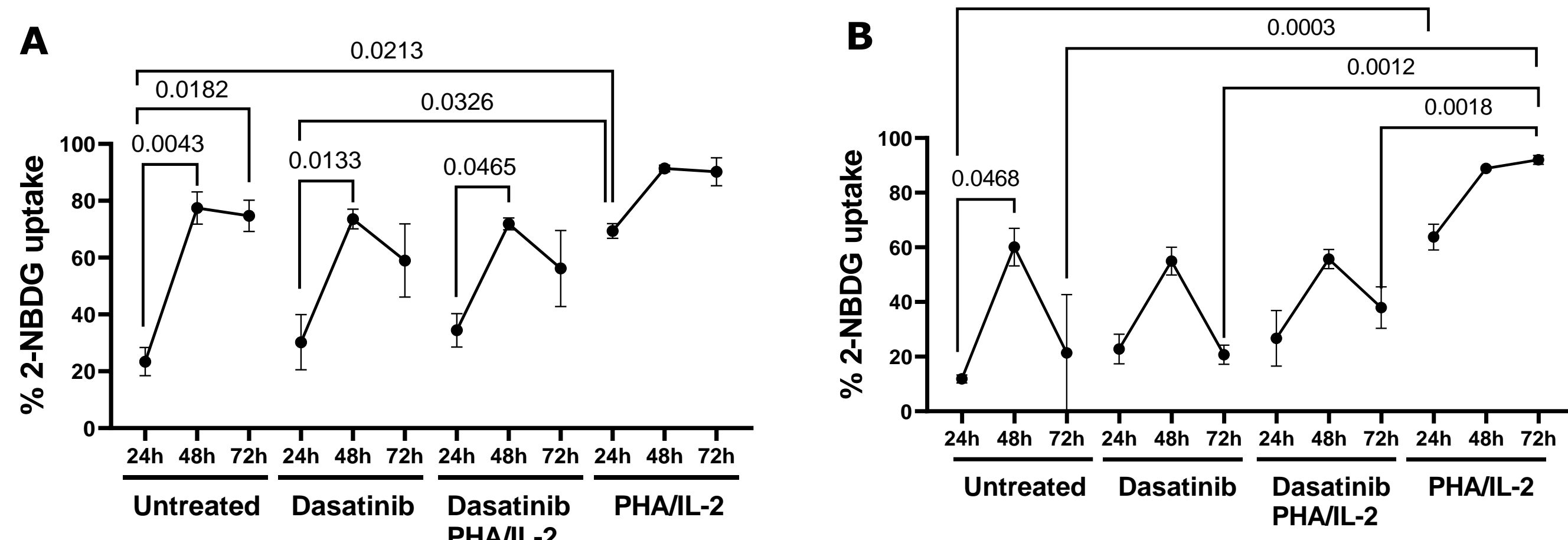
### 5. Dasatinib decreased the expression of the glucose transporter GLUT-1 in:

- CD4+ TEM lymphocytes
- CD4+ TEMRA lymphocytes

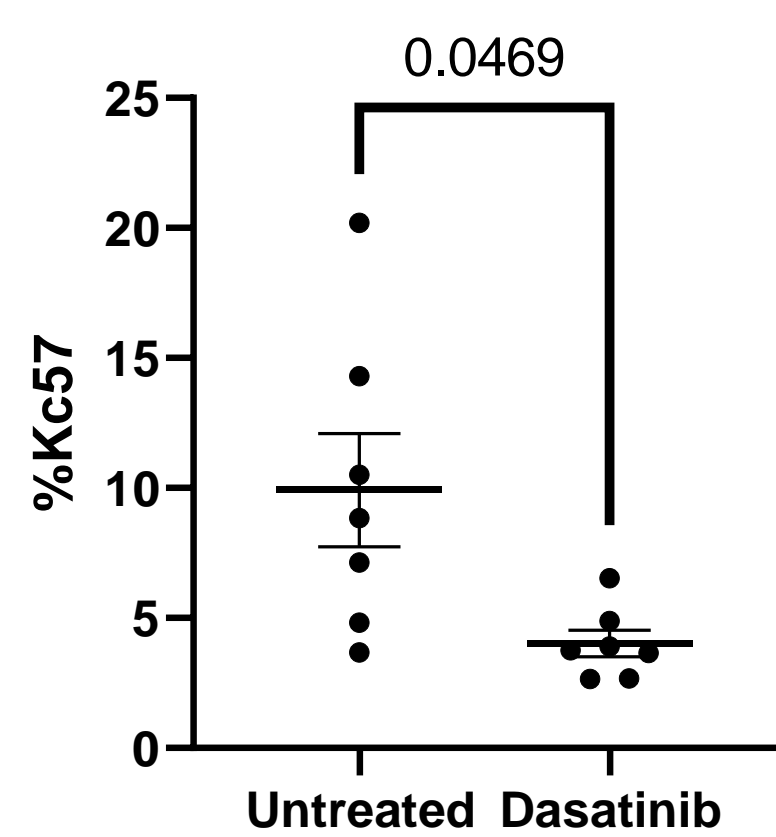


### 6. Dasatinib decreased the 2-NBDG probe uptake in:

- CD4+ TEM lymphocytes
- CD4+ TEMRA lymphocytes

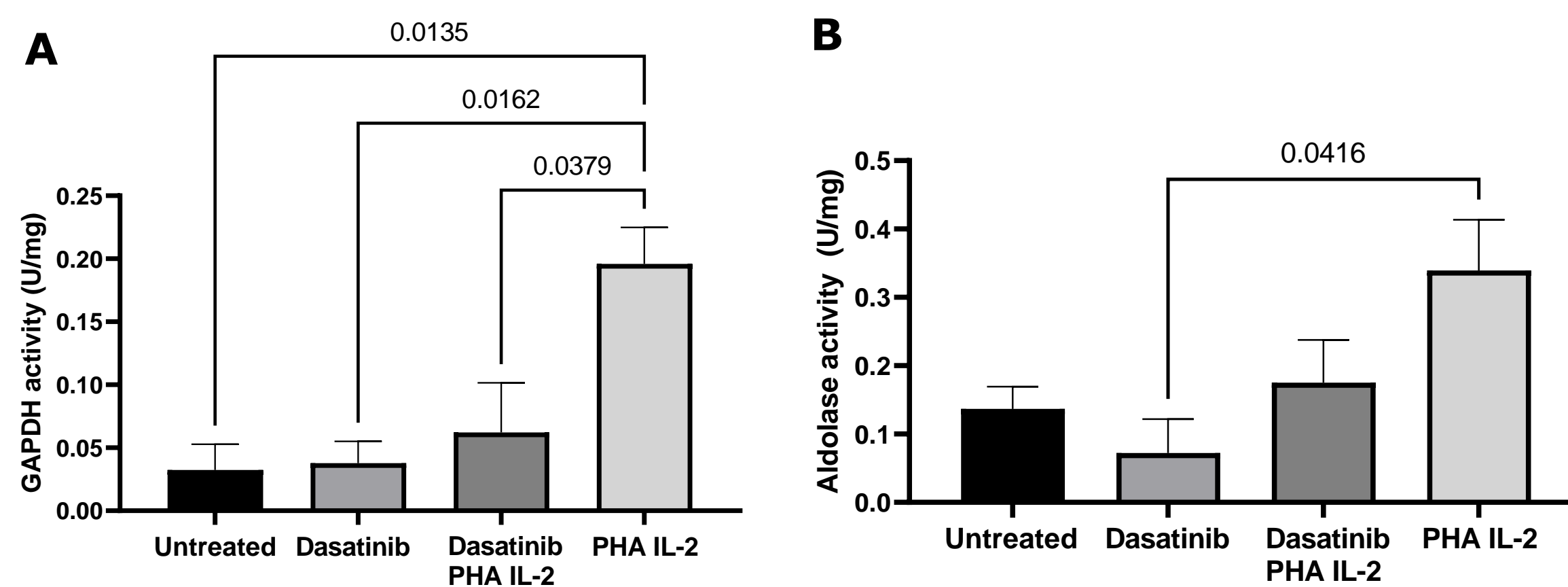


### 2. Dasatinib reduced HIV provirus reactivation in CD4+ T cells from PLWH



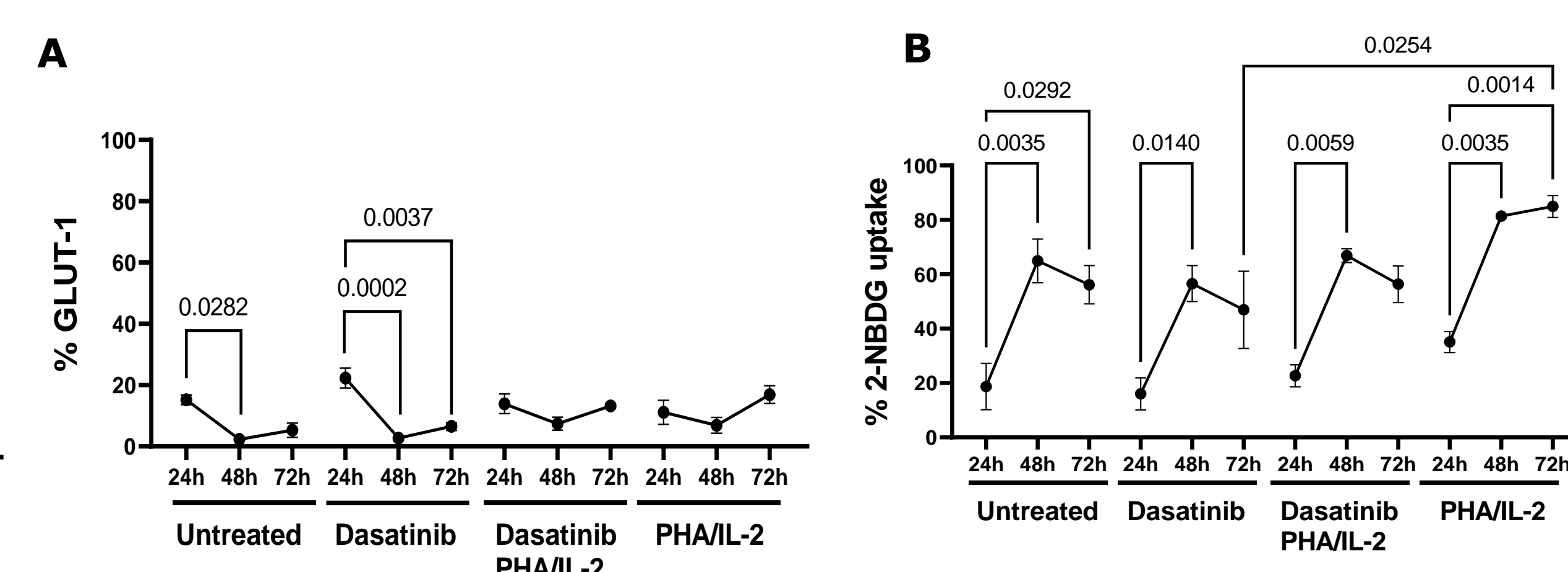
### 4. Dasatinib decreased the enzyme activity of:

- Glyceraldehyde 3-phosphate dehydrogenase (GAPDH) enzyme
- Aldolase enzyme



### 7. In NK cells:

- Dasatinib did not affect GLUT-1 expression
- Dasatinib decreased the uptake of the 2-NBDG probe



## CONCLUSIONS

- Dasatinib acts as an **LPA**, inducing a resting state in viable CD4+ T cells by blocking glycolysis and mitochondrial ATP synthesis.
- Dasatinib **prevents both HIV infection and reservoir reactivation**.
- Although glucose metabolism was partially affected in NK, PCVs treated with ART and Dasatinib do not show increased susceptibility to opportunistic infections or provirus reactivation.
- Treatment with **dasatinib and ART could silence the reservoir** as part of the **block & lock** strategy.

## ACKNOWLEDGMENTS

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