

# MOL231: PTBP1 expression in endometrial cancer cells

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## Introduction:

Polypyrimidine tract-binding protein (PTBP1) is an RNA-binding protein shown to be overexpressed in multiple types of cancer.

The perinucleolar compartment (PNC) is a nuclear substructure containing PTBP1, which is localized near the nucleolus and observed in aggressive cancer cells.

Studies have shown that knockdown of PTBP1 expression inhibits tumor cell growth.

However, the regulation of PTBP1 levels is not clearly understood.

## Hypothesis:

PTBP1 binds to the signaling lipids PI(4,5)P<sub>2</sub> and PIP<sub>3</sub>, which may contribute to its protein stability.

## Aim:

To investigate the effects of PIP5K1A and PI3K p110β inhibition on PTBP1 expression and PNC formation in endometrial cancer cells.



## Methods:

Cell culture  
(with/without inhibitor)

Immunostaining

Quantification of  
PNC prevalence

SDS - PAGE

Western blot

Quantification of  
PTBP1 protein levels

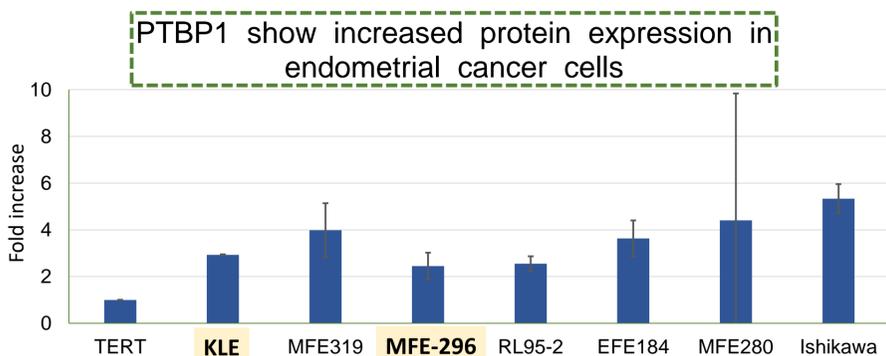


Figure 1: Quantification of western Immunoblotting from whole cell extracts obtained from the indicated endometrial cancer cell lines (n=2).

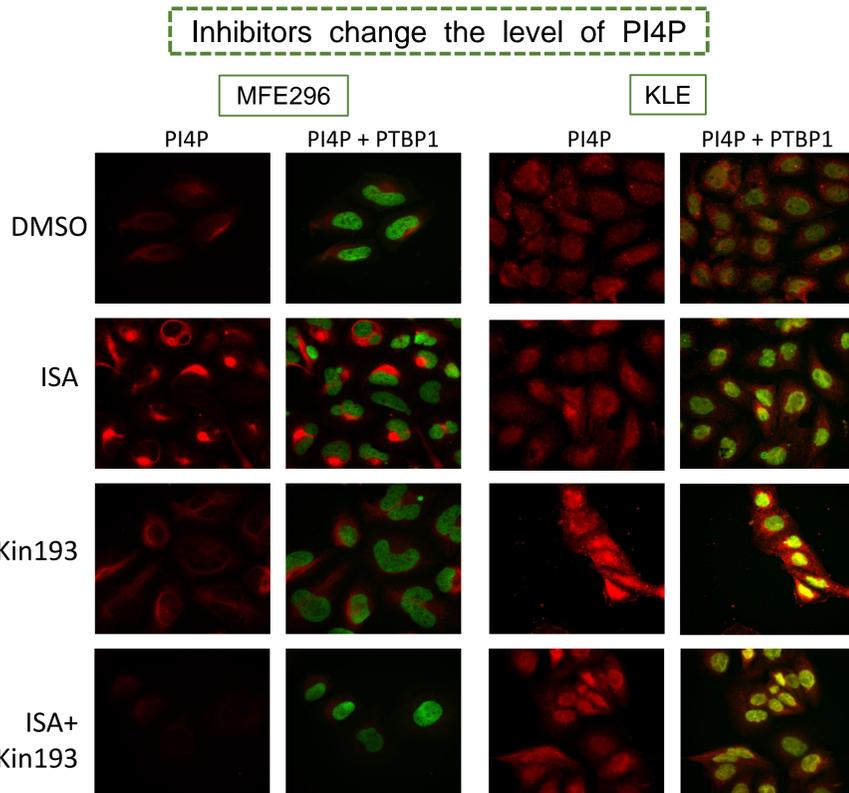
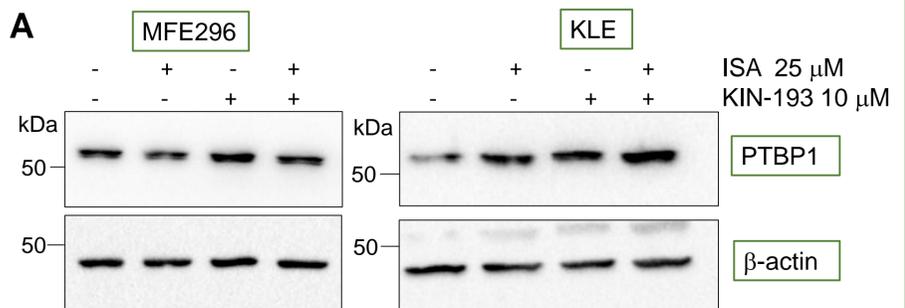


Figure 3: Immunofluorescence microscopy using anti-PI4P and anti-PTBP1 antibodies.

## A



## B

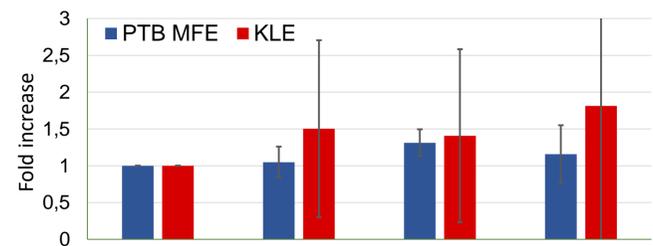
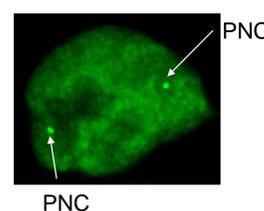


Figure 2: A. Representative Western Immunoblotting from whole cell extracts obtained following the indicated treatments for 24h. B. Quantification of PTBP1 levels relative to β-actin in MFE-296 cells (n=3) and KLE cells (n=2).

## Relative frequency of PNC in percentage

## A



## B

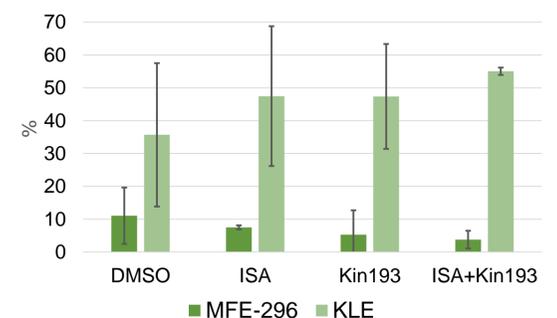


Figure 4: A. PNC revealed by anti-PTBP1 immunostaining in KLE cells (indicated by arrows). B. PNC prevalence were obtained from all experiments done on MFE296 cells (n=2) and KLE cells (n=2).

## Conclusion:

PTBP1 levels were increased upon PI3K and PIP5K1A inhibition. PI4P levels were used as an experimental control but its levels were not consistently changed in all experiments.

KLE cells showed a high % of PNCs, but this was not affected by any treatment.

Cell-death was induced in MFE296 cells upon PIP5K1A inhibition.

## References

- Takahashi et al. Significance of Polypyrimidine Tract-Binding Protein 1 Expression in Colorectal Cancer, *Mol Cancer Ther*, 2015, vol. 14, 7.
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- Wen et al., The perinucleolar compartment associates with malignancy, *Front Biol*, 2013, vol. 8, 4.