Sulindac, a COX1/COX2 Inhibitor, Exhibits Anti-tumorigenic Effects in Obesity-driven Models of Endometrioid Endometrial Cancer

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BACKGROUND

- **Endometrial cancer (EC)**: 4th most common cancer among women in U.S.[1]
  - Increasing in frequency and mortality due to the obesity epidemic.[2]
- **Obesity** induces a chronic inflammatory state which is proposed to drive development of endometrial cancer (EC).[3]
- **Adipocytes** → complex endocrine organ[4]
  - Main source of aromatase in post-menopause → unopposed estrogen
  - Modulates activity of IGF1 and IGF1R → stimulate endometrial proliferation through MAPK and AKT signaling pathways
- Secretion of pro-inflammatory adipokines such as leptin, IL-6, TNF-α → leads to insulin resistance and increased levels of IGF1[4], promoting hyperactivity of the MAPK and PI3K/AKT/mTOR pathways.
- **Sulindac** → potent anti-inflammatory effects through inhibition of the COX1 and COX2 pathways.
  - Sulindac has demonstrated promising effects in pre-clinical models of obesity driven GI cancer

Given the intimate relationship between obesity, inflammation, and EC progression, anti-inflammatories may have potential in the prevention and treatment of obesity-driven EC. However, the impact of sulindac in EC pathogenesis remains unexplored.

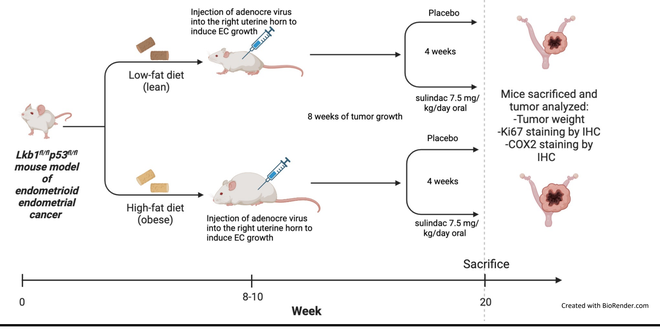
OBJECTIVE: To analyze the effects of sulindac on tumorigenesis and tumor development in endometrioid EC cell lines and in a transgenic mouse model of endometrioid EC.

METHODS

1. Human endometrioid endometrial cancer cell lines: KLE, HEC-1

- Exposed to **sulindac** at varying concentrations and studied by:
  - Cell proliferation (MTT assay, colony count assay)
  - Apoptosis (Cleaved Caspase-3, -8, -9 assays)
  - Cellular stress (Reactive oxygen species (ROS), JC-1, and TMRE assays)
  - Migration determined by wound healing assay
  - Western immunoblotting to assess sulindac's effects on downstream targets related to cellular stress, apoptosis, cell cycle control and DNA damage

2. *Lkb1$^{fl/fl}$p53$^{fl/fl}$* mouse model of endometrioid endometrial cancer



1. Seigel et al. Cancer Statistics. 2021 | 2. Smrz et al. AJOG. 2021 | 3. Onstad et al. Journal of Clinical Oncology. 2016 | 4. Yang et al. Frontiers in Oncology. 2019

RESULTS

Figure 1. Sulindac inhibited cell proliferation in dose-dependent fashion by MTT assay (left) and colony count assay (right).

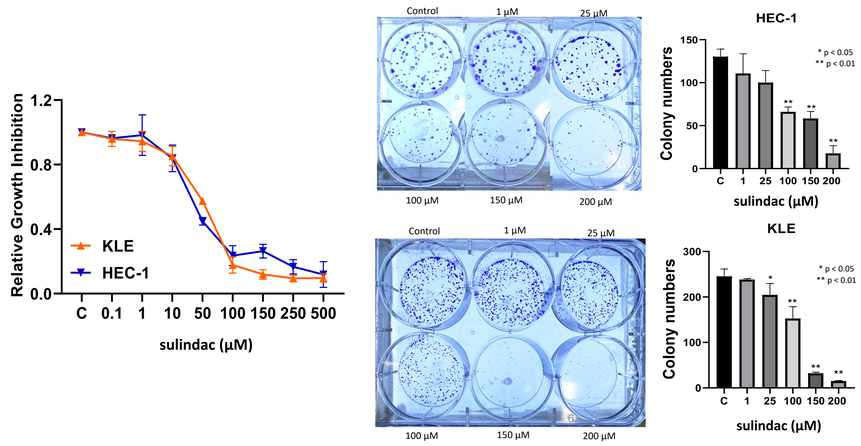


Figure 2. Induction of apoptosis in a dose-dependent fashion by sulindac.

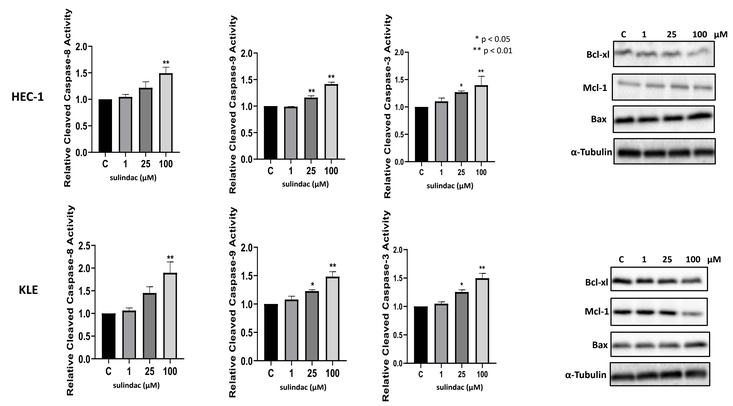


Figure 3. Sulindac Increased cellular stress in a dose-dependent fashion.

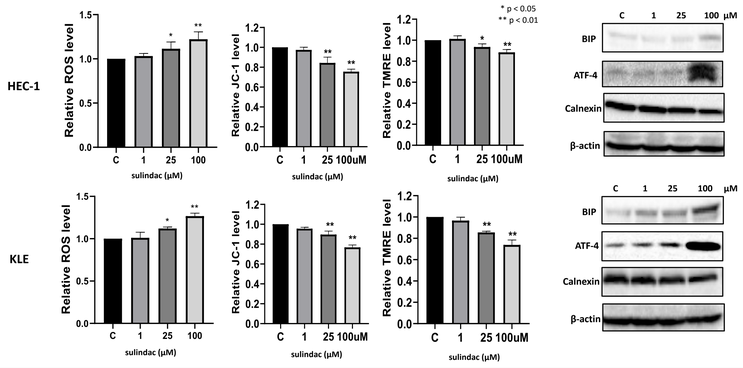


Figure 4. Sulindac inhibitd cell invasion *in vitro*.

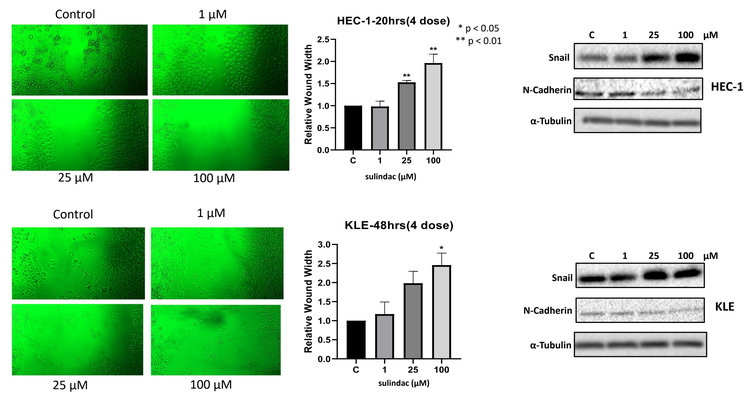


Figure 5. Sulindac decreased cell cycle proteins (A) and downregulated COX-2 (B).

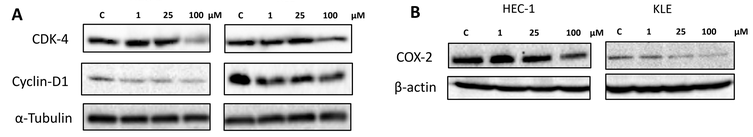
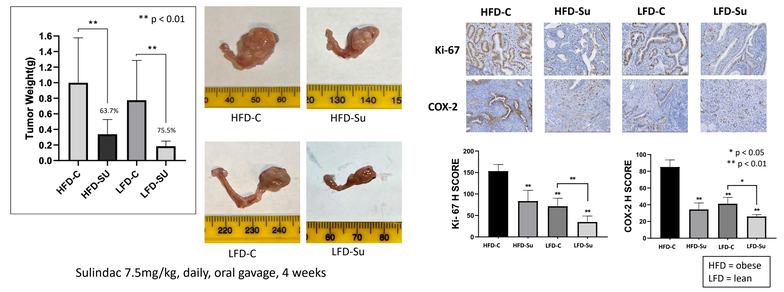


Figure 6. Sulindac inhibited tumor growth and decreased Ki-67 and COX-2 expression in the *Lkb1$^{fl/fl}$p53$^{fl/fl}$* mouse model.



Sulindac 7.5mg/kg, daily, oral gavage, 4 weeks

CONCLUSIONS

Sulindac has **anti-tumorigenic** and **anti-proliferative** effects in endometrioid EC:

- Inhibition of cell proliferation, induction of apoptosis, increased cellular stress, inhibition of cell invasion and decreased cell cycle proteins.
- Significant reduction in EC tumor weight, Ki-67 expression and COX-2 expression. These effects were more pronounced in the setting of obesity.

Take-away: Further studies are needed to assess if sulindac would be a novel intervention in the treatment of endometrial cancer.

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