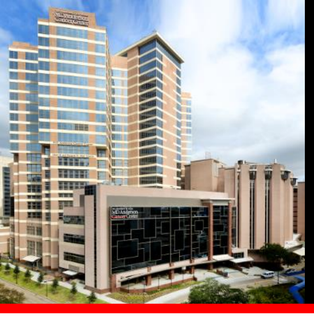

ATR Inhibitor BAY 1895344 Inhibited Proliferation of Triple-Negative Breast Cancer Cells *In Vitro* by Arresting Cell Cycle Progression and Inducing Apoptosis

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Background

- Triple-negative breast cancer (TNBC) is an aggressive type of breast cancer with high recurrence and mortality rates[1].
- Replication stress, caused by DNA damage, leads to genetic instability in cancer, which is the hall marker of cancer[2].
- Targeting ataxia telangiectasia and Rad3 proteins (ATR), a key sensor of DNA damage and replication stress, has emerged as an effective option for cancer treatment[2].
- Following DNA damage, several events driving TNBC pathogenesis increase reliance on ATR signaling[3].
- Targeting ATR can be a therapeutic option for TNBC[3].
- BAY 1895344 (BAY) exhibited antitumor efficacy in various cancer models by targeting ATR[3].

Objectives

- To evaluate the *in vitro* antitumor efficacy of ATR inhibitor BAY in TNBC cells.
- To investigate the action mechanism of BAY.

Hypothesis

BAY inhibits TNBC cell proliferation via inducing cell cycle arrest and apoptosis by specifically targeting ATR.

Methods

- The anti-proliferation effect of BAY was assessed using the CellTiter-Blue, sulforhodamine B, and clonogenic assays.
- The impact of BAY on target impression was assessed by Western blotting.
- The impacts of BAY on apoptosis and cell cycle progression were assessed by flow cytometry.

Acknowledgements

I am very grateful to the PCCSM program at MD Anderson Cancer Center and Dr. Naoto T. Ueno for giving me this great opportunity to learn knowledge of breast cancer and related technology.

Results

BAY inhibited proliferation of TNBC cells *in vitro*

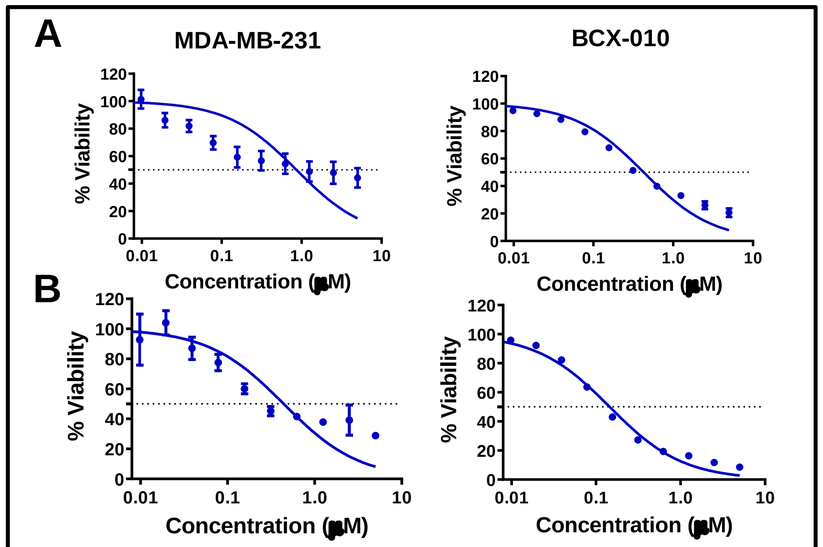

Fig 1. The anti-proliferation effect of BAY was determined using (**A**) CellTiter-Blue and (**B**) Sulforhodamine B assays at 72 h following treatment. MDA-MB-231: IC_{50} = 0.44 μM, BCX-010 cells: IC_{50} = 0.14 μM.

BAY induced apoptotic cell death

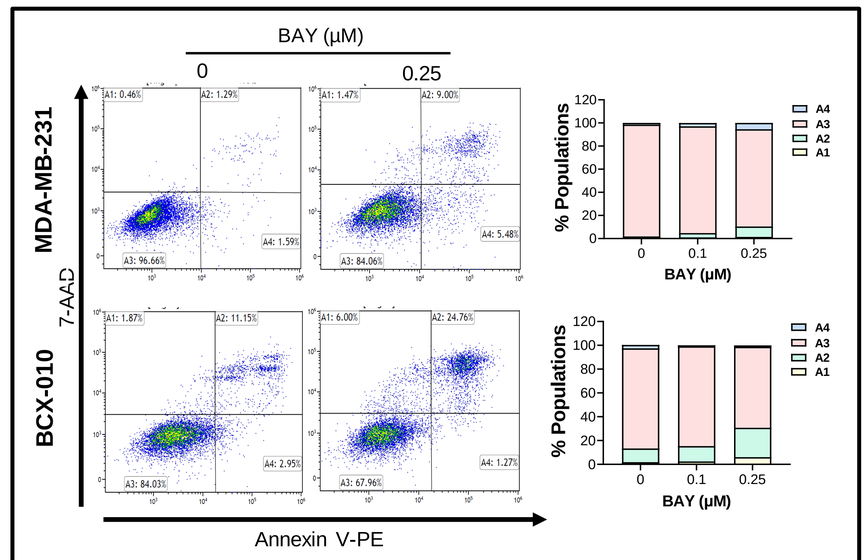

Fig 4. Cells were treated with BAY for 72h, stained with annexin V and 7-ADD, and then subjected to flow cytometry analysis.

BAY inhibited colony formation of TNBC cells *in vitro*

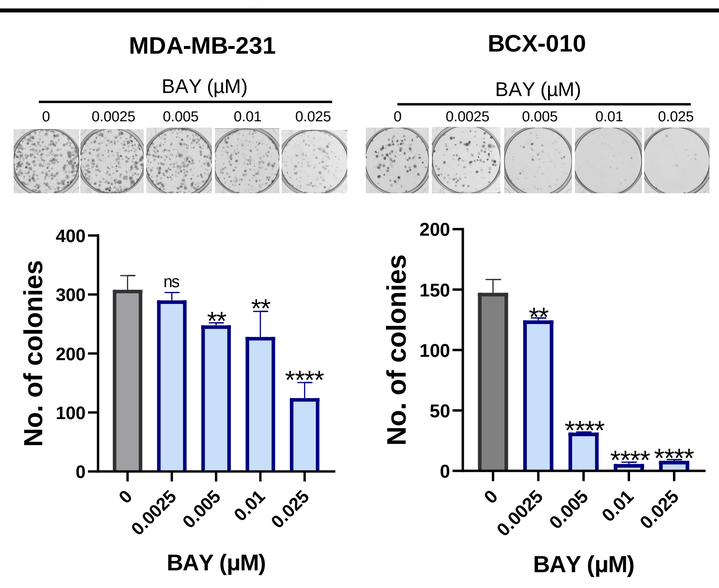

Fig 2. The impact of BAY on long-term growth was determined using a clonogenic assay at days 10 following treatment. ns: not significant, ** $P < 0.01$, **** $P < 0.0001$ for BAY treatment vs. the vehicle control.

BAY induced G2/M and S phase arrest

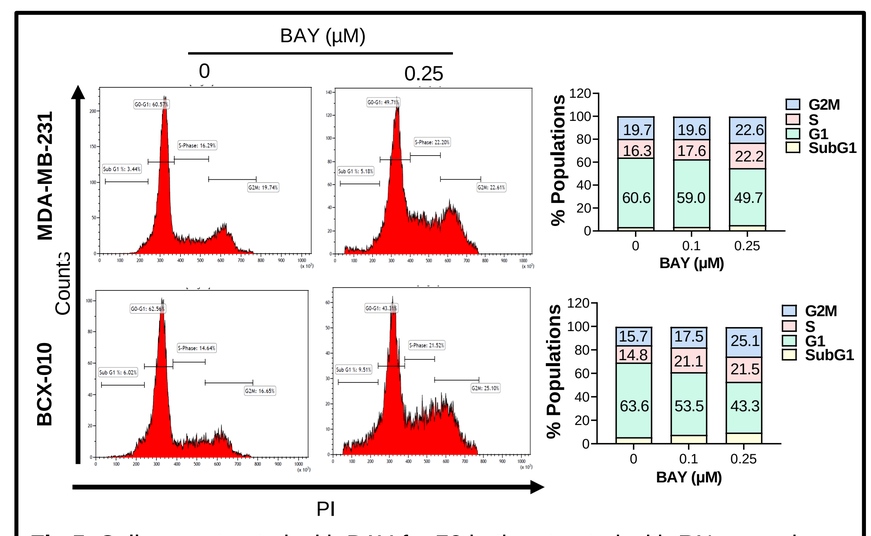

Fig 5. Cells were treated with BAY for 72 h, then treated with RNase and stained with propidium iodide (PI), followed by flow cytometry analysis.

BAY inhibited colony formation of TNBC cells *in vitro*

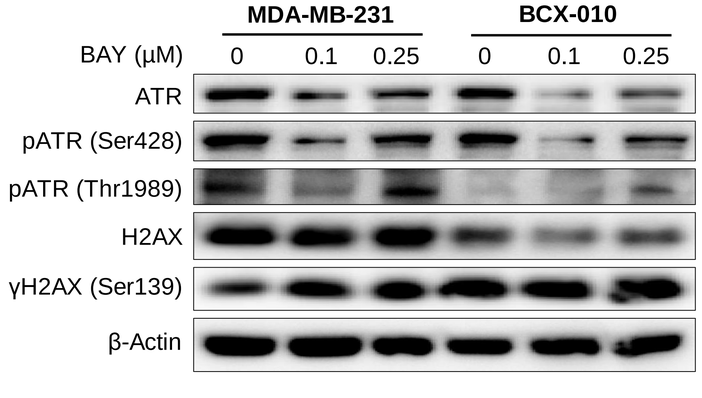

Fig 3. The effect of BAY on expression of ATR and H2AX (a substrate of BAY) was determined by Western blotting at 48 h after treatment.

Conclusion

- BAY suppressed TNBC cell growth by inducing cell cycle arrest and apoptosis.
- Targeting ATR can be an effective treatment strategy for TNBC.

Future studies

- Assess whether BAY inhibits TNBC cell growth by specifically targeting ATR.
- Evaluate the *in vivo* antitumor efficacy of BAY.
- Identify a potential partner whose inhibition potentiates BAY's antitumor efficacy.

References

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