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- and the pathogenesis of multiple B-cell malignancies ¹.

- clinical phase I trials (NCT05006716 and NCT05294731)⁶.





- Luminescent cell viability assay in both TMD8 and OCI-LY10 lymphoma cell lines where various known clinically emergent BTK resistance mutants were overexpressed.
- Cells were treated with multiple doses of BGB-16673, Compound-ED or pirtobrutinib. IC50s were calculated from the dose response curves. - A homogeneous time resolved fluorescence assay and western blot were conducted to evaluate BTK mutants' degradation by BGB-16673 in the TMD8 cell line.

Bruton Tyrosine Kinase (BTK) Protein Degrader BGB-16673 Is Less Apt To Cause, And Able To P1239 **Overcome Variable BTK Resistance Mutations Compared To Other BTK Inhibitors**

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A, BTK cDNA sequencing analysis; B, Heatmap of average IC50 in multiple ENU clones with the same BTK mutation; C, Western blot analysis of high frequent and ENU mutagenesis screen/clinic overlapped BTK mutations.

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• In ENU mutagenesis screens, fewer resistant clones and lower BTK mutation frequency were seen with BTK CDAC treatment as compared to non-covalent BTKi treatment (Figure 3 and Table 1).



*Resistant clone rate is defined by the proportion of the number of emerged resistant clones to the number of seeded wells.

Compound-ED (E3 dead non-covalent control)

100% (80 out of 80)

100% (144 out of 144)

Pirtobrutinib

(non-covalent inhibitor)

**BTK mutation frequency is defined by the proportion of the number of resistant clones harboring BTK mutations to the number of

• BGB-16673 appears to have a unique BTK resistance mutation profile vs. pirtobrutinib. Much fewer

- No BTK mutations with frequency >10% were observed in the BGB-16673 arm. In comparison, more clones resistant to pirtobrutinib had mutations in BTK, many of which arose in the kinase domain. Among the pirtobrutinib clinically derived BTK mutations, V416L and T474I were identified as high frequent mutations, and one L528W clone was detected (**Figure 4A**).



- pirtobrutinib screens (**Figure 4B**).



Figure 6. Degradation of the BTK mutants by BGB-16673



A, BTK degradation was evaluated in TMD8 overexpression cell lines by HTRF assay; B, Further verified by western blot analysis; C, Summary of BTK degradation data from HTRF assay; Abbreviations: DC, degradation constant; Dmax, maximum degradation

CONCLUSIONS

ACKNOWLEDGMENTS

DISCLOSURE

• All authors have no conflicts of interest to disclose. **CONTACT INFORMATION**

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BGB-16673 was more potent against BTK mutations derived from compound-ED and

BTK kinase activities of BTK mutations that were both frequently observed in the ENU screen and known to be clinically emergent displayed significant variability (Figure 4C).

• BGB-16673 could overcome all BTK resistance mutations from both covalent and noncovalent BTKi trials, except A428D which was tested to be resistant to all BTKi (Figure 5).

Figure 5. Heatmap of average IC50 in the BTK mutant overexpression cell lines

• BGB-16673 could degrade BTK in the presence of clinically relevant resistance mutations (V416L, M437R, T474I, C481S, C481F, C481Y, and L528W) (Figure 6).

• Relative to other BTKi, the BTK CDAC BGB-16673 is less apt to cause on-target resistance mutations, demonstrated a unique on-target resistance mutation profile. Further studies exploring the dominant BGB-16673 resistance mechanisms not reliant on BTK are in progress. • BGB-16673 could overcome a wide variety of BTK resistance mutations derived from both ENU mutagenesis screens and relapsed patients.

• These findings suggest that BGB-16673 is a promising novel BTK degrader that could benefit patients who develop BTKi on-target resistance mutations.

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