



UNIVERSITY OF LEEDS

This is a repository copy of *AZD1775 induces toxicity through double-stranded DNA breaks independent of 5-FU activity in P53 mutated colorectal cancer cells*.

White Rose Research Online URL for this paper:
<http://eprints.whiterose.ac.uk/117254/>

Version: Accepted Version

Article:

Webster, PJ, Littlejohns, AT, Beech, DJ et al. (1 more author) (2017) AZD1775 induces toxicity through double-stranded DNA breaks independent of 5-FU activity in P53 mutated colorectal cancer cells. *British Journal of Surgery*, 104 (S3). pp. 43-44. ISSN 0007-1323

<https://doi.org/10.1002/bjs.10531>

© 2017 The Authors. BJS © 2017 BJS Society Ltd. This is the peer reviewed version of the following article: Webster, PJ, Littlejohns, AT, Beech, DJ et al. (2017) AZD1775 induces toxicity through double-stranded DNA breaks independent of 5-FU activity in P53 mutated colorectal cancer cells. *British Journal of Surgery*, 104 (S3). pp. 43-44. doi:10.1002/bjs.10531, which has been published in final form at <https://doi.org/10.1002/bjs.10531>. This article may be used for non-commercial purposes in accordance with Wiley Terms and Conditions for Self-Archiving. Uploaded in accordance with the publisher's self-archiving policy.

Reuse

Items deposited in White Rose Research Online are protected by copyright, with all rights reserved unless indicated otherwise. They may be downloaded and/or printed for private study, or other acts as permitted by national copyright laws. The publisher or other rights holders may allow further reproduction and re-use of the full text version. This is indicated by the licence information on the White Rose Research Online record for the item.

Takedown

If you consider content in White Rose Research Online to be in breach of UK law, please notify us by emailing eprints@whiterose.ac.uk including the URL of the record and the reason for the withdrawal request.



eprints@whiterose.ac.uk
<https://eprints.whiterose.ac.uk/>

O135 AZD1775 INDUCES TOXICITY THROUGH DOUBLE-STRANDED DNA BREAKS INDEPENDENT OF 5-FU ACTIVITY IN P53 MUTATED COLORECTAL CANCER CELLS

PJ Webster, AT Littlejohns, DJ Beech, DA Burke

School of Medicine, University of Leeds, Clarendon Way, Leeds

Introduction: Colorectal cancer is a global health problem with an estimated 700,000 deaths per year. AZD1775 is a small molecule WEE1 inhibitor used in combination with DNA-damaging agents to cause premature mitosis and cell death in p53 mutated cancer cells. Recently, AZD1775 has been shown to have monotherapeutic activity by causing double-stranded DNA (DS-DNA) breaks as a result of nucleotide exhaustion. We sought to determine the dominant mechanism of action of AZD1775 in combination with 5-FU in a p53-mutated colorectal cancer cell line.

Method: HT29 cells (p53 mutated) were treated with 5-FU (1 μ M) for 24 hours followed by AZD1775 (300 nM). Proliferation was assessed using a WST-1 assay. Flow cytometry was used to quantify levels of double-stranded DNA breaks and premature mitosis using the specific markers γ H2AX and pHH3. Caspase-3 dependent apoptosis was quantified using an Incucyte Imaging system.

Result: AZD1775 significantly improved the cytotoxicity of 5-FU decreasing the IC50 from 9.3 μ M to 3.5 μ M. It caused significantly more mitosis (3.8% vs 56.2%), DS-DNA breaks (5.1% vs. 60.5%) and caspase-3 dependent apoptosis (4% vs. 13%) compared to 5-FU alone. The addition of exogenous nucleosides significantly rescued the increased DS-DNA breaks (60.5% vs 6.9%) and caspase-3 dependent apoptosis (13% vs 4.8%) caused by AZD1775, suggesting this to be the dominant mechanism of action, not premature mitosis.

Conclusion: AZD1775 has independent cytotoxic effects from 5-FU in p53-mutated colorectal cancer cells. This finding is important for designers of future clinical trials when considering the timing and duration of AZD1775 treatment.

Take-home message:

AZD1775 causes DS-DNA breaks and has independent cytotoxic effects from 5-FU in p53-mutated colorectal cancer cells.