# Signaling dynamics of DNA damage response invoked by combination therapy are dose-dependent

DOI 10.1074/jbc.L119.007381

**Siang-Boon Koh**[1]

*From Harvard Medical School, Boston, Massachusetts 02115 and Massachusetts General Hospital Cancer Center, Boston, Massachusetts 02114*

Edited by Patrick Sung

Warren and Eastman (1) showed that gemcitabine sensitization by CHK1i was due to CDC7/CDK2-dependent replication stress. They suggested that this mechanism was unique to delayed administration of CHK1i relative to gemcitabine and not to concurrent gemcitabine + CHK1i.

A presumption in the report is that gemcitabine depletes deoxyribonucleotides (dNTP). Although this effect is known to be induced by gemcitabine diphosphate metabolite, the major mechanism of gemcitabine in causing cell death has been attributed to its triphosphate metabolite, which is incorporated into nascent DNA to halt chain elongation (2, 3). Thus, the extensive ssDNA observed following gemcitabine and CHK1i exposure may be only partially explained by dNTP exhaustion. Likely, the high-dose gemcitabine used in the study (which caused irreversible S-phase arrest within 6 h) could also have rapidly stalled replication forks, uncoupling gemcitabine-inhibited polymerases and CHK1i-reactivated helicases. In fact, with concurrent gemcitabine + CHK1i at relatively low, but synergistic, doses, we have identified increased gemcitabine incorporation following CHK1i-enforced origin firing as the key basis of replication stress (4). A conceivable cause of the nuances in our mechanistic findings is the different dose ratios employed. As we have demonstrated recently, tumor-cell signaling dynamics orchestrated by polytherapy are often dose-dependent, and such scenarios could have profound implications in the design of drug scheduling (5).

In sum, Warren and Eastman (1) have further illuminated our understanding of CHK1i. The efficacy of delayed CHK1i administration vis-à-vis gemcitabine or concurrent gemcitabine + CHK1i remains to be validated in humans; it will likely depend on parameters such as intratumoral pharmacokinetics and patient tolerance.

The author declares that he has no conflicts of interest with the contents of this article.

[1] To whom correspondence should be addressed. E-mail: sbkoh@mgh.harvard.edu or siangboon.koh@cantab.net.

## References

1. Warren, N. J. H., and Eastman, A. (2019) Inhibition of checkpoint kinase 1 following gemcitabine-mediated S phase arrest results in CDC7- and CDK2-dependent replication catastrophe. *J. Biol. Chem.* **294,** 1763–1778 CrossRef Medline
2. Mini, E., Nobili, S., Caciagli, B., Landini, I., and Mazzei, T. (2006) Cellular pharmacology of gemcitabine. *Ann. Oncol.* **17,** Suppl. 5, v7–v12 CrossRef Medline
3. Plunkett, W., Huang, P., Xu, Y. Z., Heinemann, V., Grunewald, R., and Gandhi, V. (1995) Gemcitabine: metabolism, mechanisms of action, and self-potentiation. *Semin. Oncol.* **22,** 3–10 Medline
4. Koh, S.-B., Courtin, A., Boyce, R. J., Boyle, R. G., Richards, F. M., and Jodrell, D. I. (2015) CHK1 inhibition synergizes with gemcitabine initially by destabilizing the DNA replication apparatus. *Cancer Res.* **75,** 3583–3595 CrossRef Medline
5. Koh, S.-B., Wallez, Y., Dunlop, C. R., Bernaldo de Quirós Fernández, S., Bapiro, T. E., Richards, F. M., and Jodrell, D. I. (2018) Mechanistic distinctions between CHK1 and WEE1 inhibition guide the scheduling of triple therapy with gemcitabine. *Cancer Res.* **78,** 3054–3066 CrossRef Medline