A cell-based neuroprotection assay targeting poly (ADP-ribose) polymerase (PARP) reveals oncology-based potency ranking of clinically used PARP inhibitors (PARPi)

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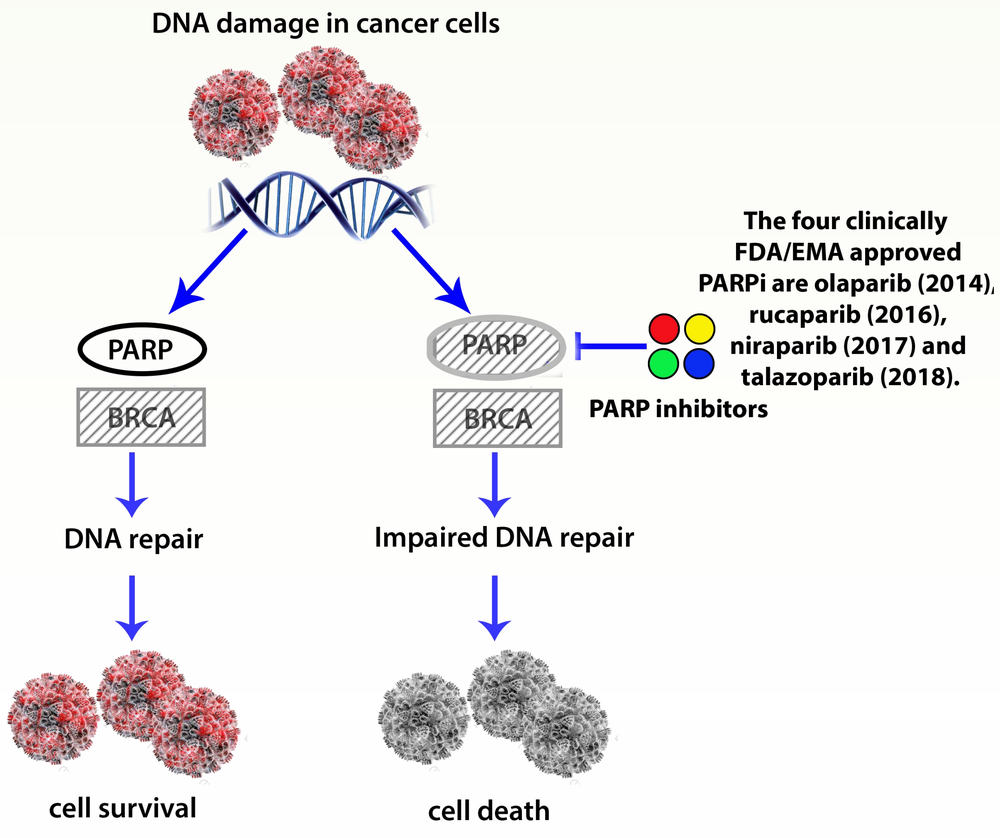
1. Introduction

- Poly (ADP-ribose) polymerase (PARP) inhibitors (PARPi) selectively kill cancer cells that harbour mutation in tumour suppressor genes, such as *BRCA1* and *BRCA2*.
- Four PARPi are currently used in the clinic as monotherapy for *BRCA*-mutated cancers.
- PARPi have also been suggested for repurposing for neurological and neurodegenerative indications, wherein excessive PARP activation leads to neuronal death and PARP inhibition is neuroprotective [1].



2. Aim

To examine whether our cell-based assay, originally designed for interrogating PARP-mediated neuronal death, is adaptable to other application contexts by revealing the established, oncology-related rank order of potencies for the PARPi.

3. Methods

- PARP-dependent cell death was induced in HeLa cells by treatment with the alkylating agent, *N*-methyl-*N*'-nitro-*N*-nitrosoguanidine (MNNG) (50μM, 25 minutes) [2].
- Cells were treated with each of four PARP inhibitors: Olaparib, niraparib, rucaparib and talazoparib (each up to 10μM).
- Protection against cell death (evidence of PARP inhibition) was assessed 24 h after MNNG treatment using the MTT assay.
- EC_{50} values reported as means ± SEM; n=3-6.
- Statistical analysis (in GraphPad 9.2.0): ANOVA with Tukey's post-hoc test; $P<0.05$ considered statistically significant.

4. Results

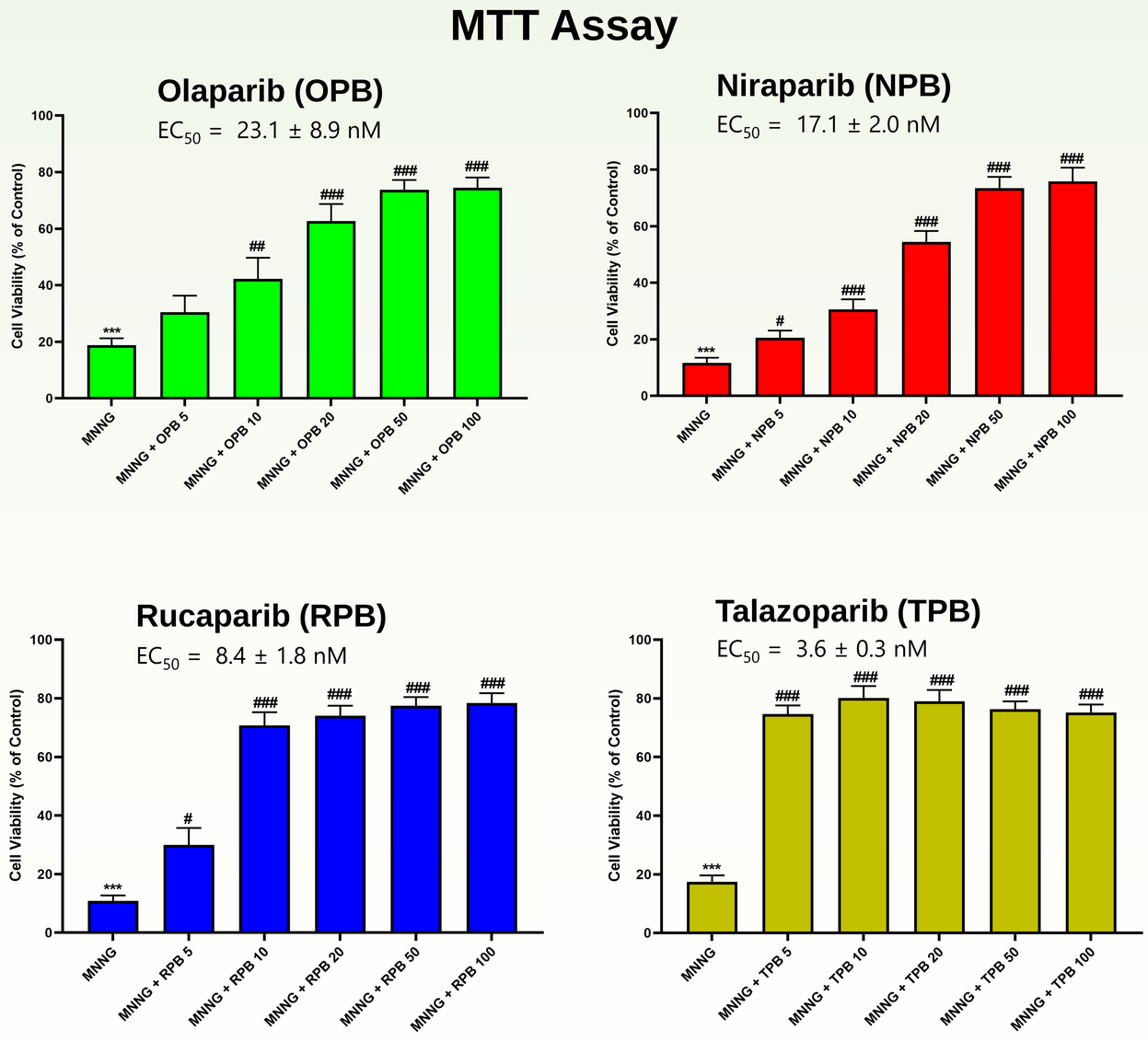


Figure 1: Each of the four PARPi (5-100nM) shows concentration-dependent protection against MNNG-induced reduction in cell viability, as quantified by the MTT assay. ***P<0.001 with respect to the control; ###P<0.001, ##P<0.05 and #P<0.01 with respect to MNNG alone.

Table 1: EC_{50} values (Mean ± SEM) for the four PARPi and their relative potencies. The EC_{50} values revealed their potencies in the following order: talazoparib > rucaparib > niraparib > olaparib.

	EC_{50} /IC_{50} (nM)		Potency ratio vs. olaparib	
	EC_{50} (our **cell-based** assay)	IC_{50} (literature-reported, **cell-free** assay)	Our Cell-based Assay	Literature-reported Cell-free Assay
Olaparib	**23.1 ± 8.9**	**5**	**1**	**1**
Niraparib	**17.1 ± 2.0**	**3.8**	**1.3**	**1.3**
Rucaparib	**8.4 ± 1.8**	**2**	**2.8**	**2.5**
Talazoparib	**3.6 ± 0.3**	**0.57**	**6.4**	**8.8**

5. Conclusions

- Although the cell-based EC_{50} values are higher than the literature-reported IC_{50} values in cell-free assays, the order of potency was the same and the relative potencies compared to olaparib were similar [2].
- Our cell-based assay is therefore robust for the characterisation of PARPi for oncological and repurposing applications.

References:

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2. Slade D (2020). PARP and PARG inhibitors in cancer treatment. *Genes Dev.* **34**:360-394.