CHEMOSENSITIZATION TO CARBOPLATIN AND PACLITAXEL BY IAP INHIBITOR DEBIO 1143 IN OVARIAN CANCER CELL LINES: SIGNATURE IDENTIFICATION FOR POTENTIAL PATIENT STRATIFICATION

D Rechavi-Robinson[a], N Wiedemann[a], C Schusterbauer[a], S Rigotti[a], F Brichory[a], G Vuagniaux[a]

[a] *Debiopharm International SA, Switzerland*

Debiopharm Group™ WE DEVELOP FOR PATIENTS

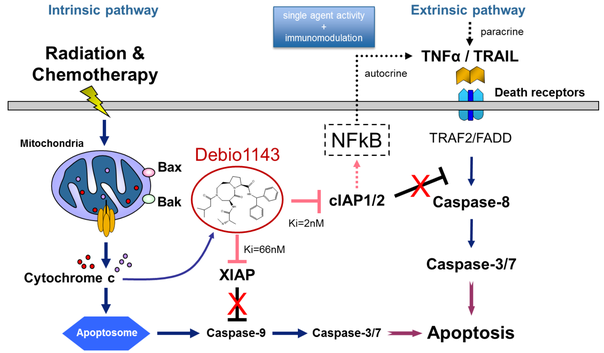
INTRODUCTION

Resistance to apoptosis is a typical hallmark of cancer. Inhibitor of Apoptosis Proteins (IAPs) block caspase activation, modulate NF-kB signaling pathways, and are involved in resistance to standard chemo and radiation therapies. IAP inhibitors such as Debio 1143 can reverse this effect and lead to apoptosis (**Figure 1**).

A phase I study of IAP inhibitor Debio 1143 in combination with standard of care (SOC) carboplatin and paclitaxel has recently shown that the combination is tolerable, with signs of activity observed in patients with heavily pre-treated epithelial ovarian cancer (EOC).

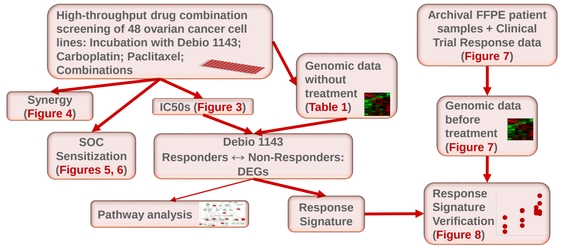
This study assessed the potential of Debio 1143-mediated sensitization of carboplatin and paclitaxel in *in vitro* models of human EOC, and provides a basis for the identification of biomarkers for response in a combination therapy setting.

Figure 1: Debio 1143 Mechanism of Action



METHODS OVERVIEW

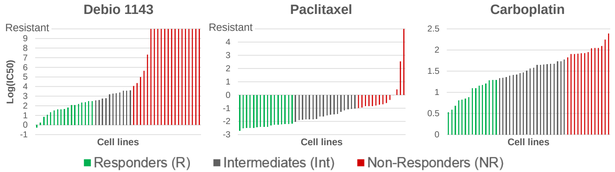
Figure 2: Methods and Results Workflow



High-throughput *in vitro* drug screening of 48 human ovarian carcinoma cell lines was performed at Horizon Discovery. Cell lines were treated with combinations of Debio 1143 with paclitaxel or with carboplatin using a dose matrix design. For each agent, IC50s were calculated, and sensitivity cut-offs based on the observed distribution were selected to categorize cell line response (**Figure 3**). Drug combination synergy was analyzed, and several cell lines were found to have a Synergy Score >10 (**Figure 4**). SOC sensitization by Debio 1143 was calculated using specific concentration ranges (**Figure 5**). Sensitization was defined as an increase in affected cells of at least 25% upon addition of Debio 1143 (**Figure 6**). A signature predictive of response to Debio 1143 was derived at Intomics by combining response and gene expression data from the cell lines with data from patient derived xenograft models. The signature was successfully tested in patient samples from a clinical trial (**Figures 7, 8**).

SINGLE AGENT IC50 RESULTS

Figure 3: Single agent IC50 distribution – Debio 1143 and SOCs



Cell viability was assessed in ATP lite assays using a dose matrix design, including zero + 8 concentrations (concentration ranges: 0.0455-100uM for Debio 1143 and carboplatin; 1.37x10^{-4}-0.03uM for paclitaxel). Cut-offs for response to Debio 1143, paclitaxel, and carboplatin were under 2.5, -2, and 1.3, respectively; cut-offs for non-response were above 4, -1, and 1.8, respectively (in log(IC50, uM) units). Cut-offs were chosen so as to create R and NR groups of approximately equal size and as large as possible, with a gap of at least one order of magnitude when possible, so as to avoid overlap of R and NR; in addition, all Responder IC50s needed to be well evaluable at the concentration range tested.

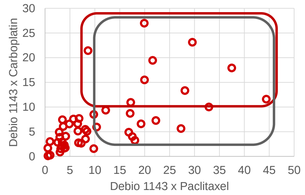
- **17/48 cell lines were sensitive to Debio 1143 alone.**
- **14 and 13 out of 48 cell lines were categorized as resistant to paclitaxel and to carboplatin respectively.**

SYNERGY AND SENSITIZATION

Figure 4: Synergy Score for Growth Inhibition

The Synergy Scores for combination were calculated by Chalice Analyzer[2] using growth inhibition (GI) data.

- **Synergy scores above 10 were observed for combinations of Debio 1143 with both paclitaxel (in 18 cell lines, gray box), and carboplatin (in 10 cell lines, red box).**



Sensitization to SOC by Debio 1143

Sensitization to paclitaxel and carboplatin by Debio 1143 was measured as the difference between the percent of cells surviving in the presence of SOC alone vs. those surviving in the presence of SOC + Debio 1143, at an average of the following concentrations: concentrations of 3.7-11.1uM Debio 1143, 11.1-33.4uM carboplatin, and 0.001-0.01uM paclitaxel. This allows to calculate a difference even in cases where the IC50 cannot be calculated (**Figure 5**). Response improvement of at least 25% upon addition of Debio 1143 was considered as sensitization (**Figure 6**). The cut-off was chosen so as to be truly different from 0%.

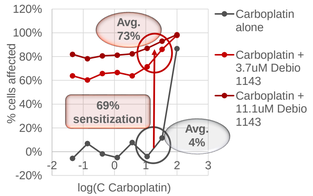


Figure 5: Example of a sensitization calculation

Carboplatin alone at 11-33uM affects 4% of the cells; when 3.7-11 uM of Debio 1143 are added, 73% of the cells are affected. Thus the sensitization is of 69%.

References:
1. Chou, Pharmacol. Rev. 2006, 621, Theoretical basis and experimental design of synergism and antagonism in drug combination studies
2. See chalice.horizondiscovery.com
3. Wiedemann N, Langdon CG et al., ELCC 2014 Abstract 317. Activity of IAP antagonist Debio 1143 as a monotherapy and in combination with standard of care agents in models of human lung cancer of different histotypes.

SIGNATURE AND VERIFICATION

Genomic Signature of response to Debio 1143

Gene expression data for the cell lines were taken from public datasets (CCLE – GSE36133, and Garnett – E-MTAB-3610), and in addition RNA-seq gene expression was acquired for 20 of the cell lines (using AmpliSeq Transcriptome). Previous results from 3D in vitro PDX models and their corresponding Affymetrix expression data were also used.[3]

Table 1: Number of cell lines with available genomic data in the platform used for genomic analysis

Response to Debio 1143	CCLE	Garnett	AmpliSeq RNA-seq	Total with data in any platform
R	16	11	13	17
Int	8	4	2	10
NR	12	13	5	18

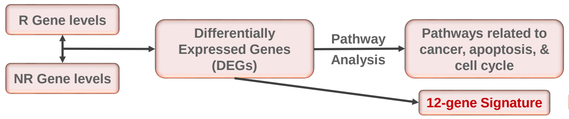
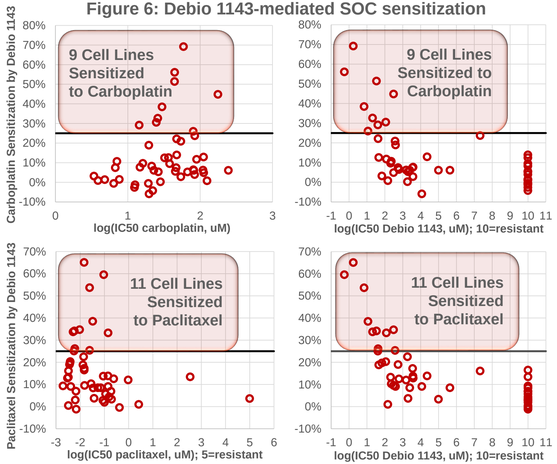
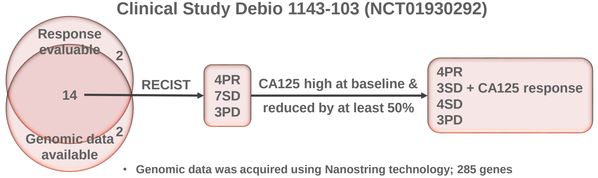


Figure 6: Debio 1143-mediated SOC sensitization



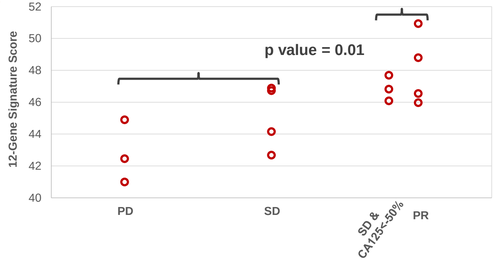
- **Sensitization was observed more frequently in cell lines with relatively low Debio 1143 IC50.**
- **No desensitizations were observed.**

Figure 7: Patient Response and Genomic Data available from Clinical Study Debio 1143-103 (NCT01930292)



- Genomic data was acquired using Nanostring technology; 285 genes considered as related to the mechanism of action were quantified.
- PD=Progressive Disease; SD=Stable Disease; PR=Partial Response.

Figure 8: Successful Signature Verification in FFPE Samples from Platinum-Resistant Ovarian Cancer Patients



- **The difference between responding patients (by BOR or by CA125) and others was found to be significant: the p value for the Wilcoxon rank-sum test was 0.01.**

CONCLUSIONS

- **Preclinical *in vitro* data of response to Debio 1143, SOC (carboplatin and paclitaxel) and their combinations were acquired in 48 ovarian cancer cell lines, and shows sensitization by Debio 1143 in about a quarter of the cell lines, suggesting that Debio 1143 is a good candidate for combination with SOC.**
- **A signature based on genomic expression was derived from in vitro data of response and successfully verified in patient samples from a clinical Phase I study. This confirms potential clinical relevance for selection of responsive patients, and will be verified in an ongoing Phase II trial in view of patient selection.**

CONTACT

Debiopharm International S.A.,
Lausanne, Switzerland.
www.debiopharm.com
Gregoire.Vuagniaux@debiopharm.com

DOWNLOAD

This poster is available via:
www.debiopharm.com/medias/publications