Assessment of dianhydrogalactitol (VAL-083) in the treatment of relapsed or refractory non-small cell lung cancer

Anne Steino[1], Guangan He[2], Beibei Zhai[3], Jeffrey A. Bacha[1], Shun Lu[4], Dennis M. Brown[1], Mads Daugaard[3] and Zahid H. Siddik[2]

[1]DelMar Pharmaceuticals, Inc., Vancouver, Canada and Menlo Park, CA, USA; [2]The University of Texas MD Anderson Cancer Center, Houston, TX; [3]Vancouver Prostate Centre, Vancouver, Canada; [4]Shanghai Lung Cancer Center, Shanghai, China

ABSTRACT # 4639:

VAL-083 is a bi-functional alkylating agent with proven activity against NSCLC in historical NCI-sponsored clinical studies[1-5].

VAL-083 is approved for the treatment of lung cancer in China; however, clinical adoption is limited by lack of modern data related to mechanism-of-action (MoA) and utility in the context of standard-of-care in NSCLC. We have previously demonstrated that VAL-083 circumvents cisplatin-resistance in ovarian cancer cells, *in vitro*. VAL-083 also exhibits superior activity to cisplatin in both *in vitro* and *in vivo* NSCLC models, including TKI-resistant NSCLC.[6] Here we further differentiate VAL-083 from current standard-of-care in NSCLC by investigating *in vitro* i) the distinct MoA of VAL-083, ii) VAL-083 cytotoxicity in a panel of NSCLC cell lines with varying p53 status and T790M and KRAS mutations, and iii) the combination of VAL-083 with cisplatin or oxaliplatin.

NSCLC: Lung cancer, including NSCLC, is treated with surgery and chemotherapy with tyrosine kinase inhibitors (TKIs) or platinum-based regimens. EGFR mutated tumors account for 10-15% and 40% of NSCLC in Western and Asian populations, respectively. In EGFR mutated NSCLC, TKI-treatment produces dramatic initial improvements, but tumors ultimately recur with new mutations, including T790M. Third generation TKI AZD9291 is effective against recurrent NSCLC with the T790M EGFR mutations, but acquired resistance appears to emerge through RAS signaling, including KRAS mutations. Cisplatin-resistance also represents an unmet clinical need, and long-term prognosis in NSCLC remains poor.

MECHANISM-OF-ACTION

VAL-083 targets N7 of guanine leading to DNA interstrand crosslinks, irreparable DNA double strand breaks, persistent S/G2-phase cell cycle arrest, and activation of the HR DNA repair pathway.[6,7]

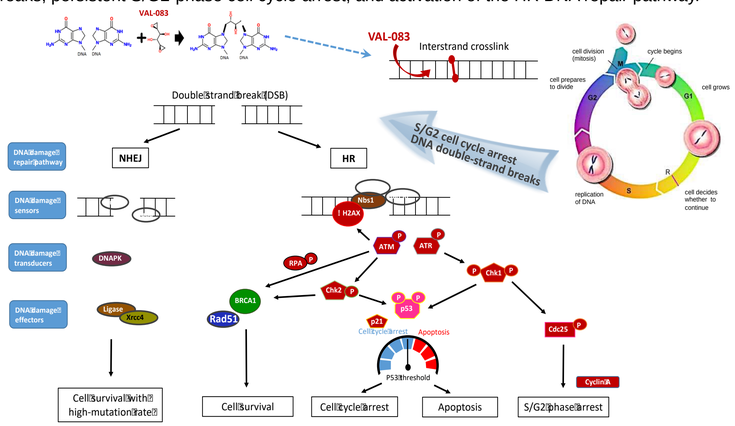


FIGURE 1. VAL-083 induces interstrand crosslink leading to double-strand breaks, S/G2 phase arrest and HR activation. Red color signifies demonstrated activation/expression after VAL-083 treatment.

CLINICAL BACKGROUND

VAL-083 demonstrated activity in prior clinical trials sponsored by the US National Cancer Institutes. VAL-083 is approved for the treatment of lung cancer in China.

TABLE 1. **Historical clinical results with VAL-083 in lung cancer**

Reference	Patient Population	Reported Result
Haas et al. (1976[1]) VAL-083 single agent	Advanced lung cancer	ORR • 42% sqclc • 25% adeno
Eagan et al. (1977[2]) VAL-083 single agent	Advanced SQCLC	Regression: • 15%: sqclc
Eagan et al (1980[3]) VAL-083 combination therapy	Advanced SQCLC	Regression: • 27% VAL-083 + adriamycin • 53% VAL-083 + adriamycin + cisplatin
Haas et al. (1981[4]) VAL-083 single agent	Advanced lung cancer	ORR • 19%: recurrent disease • 26%: newly diagnosed
Eagan et al. (1981[5]) VAL-083 combination therapy	Advanced SQCLC	Regression: • 54%: VAL-083 + adriamycin + cisplatin

ORR = SD/PR/CR

References:

1. *Haas CD, et al. Cancer Treat. Rep. 1976;60(5):611-4*
2. *Eagan et al. Cancer Treat Rep. 1977;61(7):1339-45*
3. *Eagan et al. Cancer Treat Rap. 1980;64(1):87-91*
4. *Haas CD, et al. Cancer Treat Rep.1981;65(1-2):115-7*
5. *Eagan et al. Cancer Treat Rep. 1981;65(5-6):517-9*
6. *Institoris & Tamas. Biochem J. 1980:185,659-66.*
7. *Zhai et al. AACR annual meeting 2016*

CONCLUSIONS & FUTURE DIRECTIONS

- ***Historical clinical activity combined with a new understanding of the MoA supports the potential of VAL-083 as a possible treatment of chemo-refractory NSCLC***
  - ✓ **VAL-083 has a distinct MoA from other chemotherapeutics used in the treatment of NSCLC**
  - ✓ **VAL-083 overcomes TKI-resistance in NSCLC cell lines, including cells with the EGFR mutation T790M and KRAS mutations**
  - ✓ **VAL-083 displays super-additivity and synergy with both cisplatin and oxaliplatin in NSCLC cell lines, including TKI-resistant cells with T790M or KRAS mutations**
  - ✓ **VAL-083 activity is independent of p53 status in a panel of NSCLC cell lines**
  - ✓ **VAL-083 displays synergy with topoisomerase inhibitors etoposide and camptothecin, suggesting VAL-083 as part of novel combination therapies**
- **An open-label post-market clinical trial in China will investigate the activity of VAL-083 in relapsed/refractory NSCLC. Results will provide guidance to physicians under the context of VAL-083's current approval in China, and serve as proof-of-concept for expanded clinical development worldwide**

VAL-083 DISPLAYS SYNERGY WITH ETOPOSIDE, CAMPTOTHECIN, CISPLATIN AND OXALIPLATIN

The distinct mechanism-of-action of VAL-083 makes it a valuable partner for combination therapies with agents already used in the treatment of GBM and other CNS tumors.

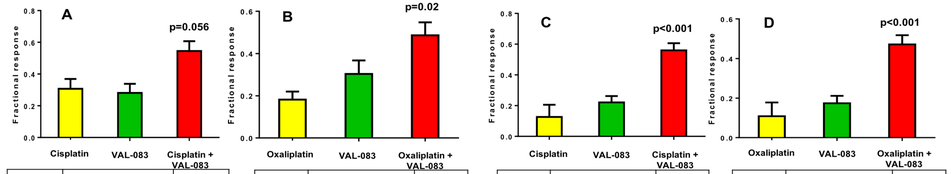
- As VAL-083 induce cell cycle arrest in S/G2-phase, we predicted synergy with agents that require cancer cells to be in S/G2-phase for maximum effect, including topoisomerase inhibitors. As expected, **VAL-083 demonstrated synergy with etoposide (topoisomerase II inhibitor) and camptothecin** (topoisomerase I inhibitor) (Table 2).
- **VAL-083 also demonstrated synergy with cisplatin and oxaliplatin** in NSCLC cell lines, suggesting distinct mechanism-of-action from the platinum-based agents (Figure 2).

A	VAL-083 in combination with etoposide (topoisomerase II inhibitor), 72 hr treatment.		
Cell line (molar ratio VAL-083:etoposide)	Cytotoxic effect (**Fa**)	Combination index (**CI**)	
PC3 (5:1)	**ED50**	0.65	
	ED75	0.59	
	ED90	0.55	
A549 (5:1)	**ED50**	0.72	
	ED75	0.88	
	ED80	0.94	

B	VAL-083 in combination with camptothecin (topoisomerase I inhibitor), 72 hr treatment.		
Cell line (molar ratio VAL-083:camptothecin)	Cytotoxic effect (**Fa**)	Combination index (**CI**)	
PC3 (250:1)	**ED75**	0.83	
	ED90	0.66	
	ED95	0.56	
A549 (212:1)	**ED85**	0.94	
	ED90	0.87	
	ED95	0.77	

TABLE 2. VAL-083 demonstrates synergy with A) etoposide (topoisomerase II inhibitor) and B) camptothecin (topoisomerase I inhibitor) in PC3 prostate and A549 NSCLC cancer cells. The tables show CI values for the cytotoxic effect (Fa), achieved at indicated drug concentrations. CI<1 shows synergy. N=3.

Human NSCLC cell line H1975 (TKI-resistant T790M mutant) | **Human NSCLC cell line A549 (KRAS mutant)**



Cytotoxic Level (Fa)	Concentration (μM) VAL-083	Cisplatin	CI
ED75	0.97	0.49	0.78
ED90	2.01	1.00	0.68
ED95	3.30	1.65	0.63

Cytotoxic Level (Fa)	Concentration (μM) VAL-083	Oxaliplatin	CI
ED75	0.78	0.44	0.65
ED90	1.49	0.84	0.53
ED95	2.31	1.31	0.46

Cytotoxic Level (Fa)	Concentration (μM) VAL-083	Cisplatin	CI
ED75	1.79	0.67	0.87
ED90	3.09	1.16	0.82
ED95	4.48	1.68	0.79

Cytotoxic Level (Fa)	Concentration (μM) VAL-083	Oxaliplatin	CI
ED75	1.55	0.46	0.91
ED90	2.26	0.68	0.78
ED95	2.92	0.88	0.71

FIGURE 2. VAL-083 demonstrates synergy with cisplatin (A,C) or oxaliplatin (B,D) on H1975 (A,B) and A549 (C,D) NSCLC cells. The tables shows CI values for the cytotoxic level (Fa) shown, achieved at indicated drug concentrations. CI<1 shows synergy. Mean +/- SE, N=4-7.

VAL-083 ACTIVITY IN A PANEL OF 11 NSCLC CELL LINES

Consistent with prior published research, VAL-083 was active against all 11 NSCLC cell-lines tested, irrespective of their p53, EGFR (T790M) and KRAS status, **suggesting a MoA that differs from other chemotherapeutic agents** in the treatment of NSCLC, including cisplatin.

These results further suggest VAL-083 as a **treatment option for chemo-resistant NSCLC, irrespective of the presence of p53, EGFR or KRAS mutations** known to induce resistance to other chemotherapeutics used in the treatment of NSCLC.

TABLE 3. **VAL-083 is active against all 11 NSCLC cell lines independent of p53, EGFR and KRAS status.**

NSCLC cell line	IC_{50} ± SE (μM)	p53	EGFR	KRAS
H460	**0.5 ± 0.1**	wild type	wild type	mutant
A549	**1.8 ± 0.3**	wild type	wild type	mutant
H226	**6.1 ± 1.0**	wild type	wild type	wild type
H1792	**4.6**	mutant	wild type	mutant
H1975	**0.9 ± 0.2**	mutant	mutant (T790M)	wild type
SkLU1	**2.7 ± 0.0**	mutant	wild type	mutant
H2122	**2.8 ± 0.3**	mutant	mutant	mutant
H157	**4.5 ± 0.4**	mutant	wild type	mutant
H23	**2.6**	mutant	wild type	mutant
H1299	**2.4 ± 0.1**	null	wild type	wild type
H838	**4.6 ± 0.4**	null	wild type	mutant