Tuning the reconstitution conditions improves stability and handling of ZD2767P in an aqueous environment

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Introduction

ZD2767P is a bis-iodo phenol mustard prodrug used in a two-step approach of targeting chemotherapy involving either antibody-directed enzyme prodrug therapy (ADEPT) or gene-directed enzyme prodrug therapy (GDEPT) [1].

ZD2767P has very poor stability in aqueous solution. Therefore the existing method for ZD2767P delivery is to provide ZD2767P as a powder, with reconstitution in cold sodium bicarbonate solution immediately prior to intravenous administration through a Hickman line [2].

Materials and Methods

ZD2767P was supplied by AstraZeneca. For HPLC analysis of ZD2767P, a Spherisorb ODS column with a H_2O/acetonitrile gradient was used. Samples were diluted with NMP/H_2O and analysed immediately to minimise degradation. Microtitrimetry analysis was performed on a Sirius T3.

Results and Discussion

Microtitrimetry analysis of ZD2767P determined the following pKa values: 3.0 (carboxyl), 3.6 (amine), 4.8 (carboxyl). This indicates that a pH > 4.8 is required for good solubility of ZD2767P. However, hydroxyl ions strongly accelerate degradation of ZD2767P by nucleophilic substitution of the iodide in the molecule, as illustrated in the pH stability (Fig. 2). Hence, careful conversion of ZD2767P into the salt form while avoiding excess alkali is critical.

N-Methylpyrrolidone (NMP) emerged as a suitable solvent for formulation (Table 1) because ZD2767P shows excellent solubility as well as comparably good stability in this solvent. A high solubility is critical because relatively large amounts of ZD2767P (610mg) are administered.

Figure 1: Structure of ZD2767P

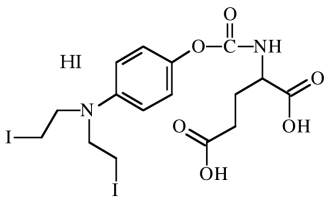

Figure 2: pH stability of ZD2767P at RT

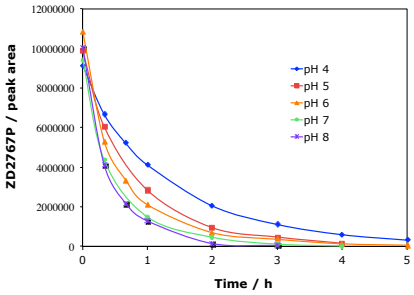

Table 1: Solubility and stability of ZD2767P

Solvent	Solubility (mg/ml)	Stability $T_{1/10}$* at RT
NMP	> 401	> 15 h
Methanol	> 64.7	~8 min
2-Propanol	11.6 - 15.4	18.2 h
Butanol	16.5 - 22.0	3.1 h
DMSO	> 58.8	-
8.4% w/v $NaHCO_3$	> 49.2	-
1M HCl	2.1 - 3.2	-
5% v/v Acetic acid	1.2 - 1.4	-
HPLC grade H_2O	~0.4	-

$T_{1/10}$: time within which 10% of the initial ZD2767P degrades

Table 2: Comparison of reconstitution methods

Preparation	Molar ratio $NaHCO_3$/ ZD2767P	NMP %	Storage Temp.	Stability $T_{1/10}$* (min)
Existing method	3	0	4°C	211
			20°C	53
			20°C	527
New method	1.6	20	26-28°C	151
			40°C	28

$T_{1/10}$: time within which 10% of the initial ZD2767P degrades

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Results and Discussion (*continued*)

An alternative method for reconstitution was developed whereby ZD2767P is dissolved in NMP (305 mg/ml), followed by addition of 8.4%w/v $NaHCO_3$ solution. The molar ratio of $NaHCO_3$:ZD2767P is 1.6. After the initial burst of CO_2 the solution is diluted with saline to adjust the NMP content to 20%v/v. ZD2767P reconstituted with the 'new' method shows improved stability (Table 2).

Advantages of dissolution in NMP are: i) no frothing which spreads ZD2767P over the surface of the vial (as occurs when dissolved directly in $NaHCO_3$) and ii) dissolution time and temperature are not critical because ZD2767P is comparably stable in NMP at RT. Overall, the 'new method' is a clean process because there are no solids present during the initial CO_2 burst following $NaHCO_3$ addition.

Conclusion

An alternative method for reconstitution of ZD2767P has been developed which provides improved handling characteristics and room temperature stability where previously cold temperature manipulation and storage was required. This allows a longer time-window between reconstitution and administration.

Acknowledgements

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References

[1] R. Begent, S. Sharma, K. Chester, "Antibody-Dependant Enzyme Prodrug Therapy (ADEPT)" in *Antibody Engineering Vol. 1*, R. Kontermann and S. Duebel, Springer-Verlag, 2010, ch. 28, pp. 431-451.

[2] R.J. Francis, S.K. Sharma, C. Springer, et al. "A phase I trial of antibody directed enzyme prodrug therapy (ADEPT) in patients with advanced colorectal carcinoma or other CEA producing tumours" *Br. J. Cancer*, 87 (2002) 600-607.