Eur J Cancer Clin Oncol, Vol. 23, No. 6, pp. 807–811, 1987.
Printed in Great Britain

0277-5379/87$3.00 + 0.00

Unchanged Pharmacokinetics of VP-16-213 (Etoposide, NSC 141540) During Concomitant Administration of Doxorubicin and Cyclophosphamide*

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Abstract—*In patients with small-cell lung cancer VP-16-213 is often given in combination with doxorubicin and cyclophosphamide. Little is known about possible interactions between these drugs. Therefore we investigated in 7 patients the pharmacokinetics of VP-16-213, with and without the other two drugs.*

We found no change in the pharmacokinetics. We also provide evidence that the pharmacokinetics did not change in two sequential administrations of the drug.

Pharmacokinetic data are in agreement with previous reports.

INTRODUCTION

VP-16-213 (etoposide, NSC 141540), a semisynthetic epipodophyllotoxin derivative, is an active agent against small-cell lung cancer (SCLC), testicular cancer, malignant lymphomas and some other malignancies [1].

For SCLC, VP-16 is one of the most active drugs [2]; the response seems to be schedule dependent [3]. Combination chemotherapy results in higher response rates for SCLC than single agent therapy of VP-16-213 [2]. In 1975 the three-drug combination, consisting of cyclophosphamide, doxorubicin and VP-16-213, was recognized as a highly active combination against SCLC by the Cancer Centre of the University of Maryland [4]. Pharmacokinetic studies of VP-16-213 in patients have been reported for medium [4–9] and for high dose treatment [10]. All three drugs are bound to protein, metabolized extensively and important amounts of the parent drugs or metabolites are excreted in the urine (see Discussion).

An influence on pharmacokinetic behaviour of VP-16-213 can be expected from these possible interactions. Therefore we compared the pharmacokinetic data of VP-16-213, after simultaneous administration of doxorubicin and cyclophosphamide, with data obtained after administration of VP-16-213 alone, in the same patient.

Table 1. *Main characteristics of the patients*

Patient No.	Sex	Age	Stage*
1	Male	59	LD
2	Male	72	ED
3	Female	74	ED
4	Female	57	ED
5	Male	59	ED
6	Male	65	LD
7	Male	48	LD

*LD = limited disease; ED = extensive disease.
No patient had abnormal liver or kidney function tests.

PATIENTS, TREATMENT AND SAMPLING PROCEDURE

We studied 7 patients with SCLC. The main characteristics of the patients are presented in Table 1.

In our hospital two alternating drug combinations have been given. Cyclophosphamide, doxorubicin and VP-16-213 alternate with cyclophosphamide, methotrexate, CCNU and vincristine. The first combination is given as follows: cyclophosphamide 800 mg/sqm (bolus, i.v.),

Accepted 2 December 1986.
*This study was supported by the Netherlands Cancer Foundation (grant No. KWF IKA 81–15).
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doxorubicin 40 mg/sqm (bolus, i.v.) and VP-16-213 100 mg/sqm (1 hr infusion) on day 1 and day 8. In our study, cyclophosphamide and doxorubicin on day 1 were given just before the administration of VP-16-213. Therefore we were able to investigate the pharmacokinetics of VP-16-213 with (day 1) and without (day 8) the other two drugs. Samples were taken on day 1 and 8 at 0 min, 30 min (during infusion), 60 min (directly after the infusion), 75 min, 90 min, 2 hr, 2.5 hr, 3 hr, 4 hr, 5 hr, 6 hr, 11 hr and 25 hr.

In all patients the study was done during the first course of treatment. The samples were directly put on ice, centrifuged and stored at −30° C.

Drug assay

VP-16-213 was determined with a HPLC system in combination with an electrochemical detection system. The method has been developed by Holthuis *et al.* [11]. VP-16-213 and VM-26 (teniposide) were kindly provided by Bristol-Myers (Syracuse, New York, U.S.A.). All other chemicals used were of analytical grade. VM-26 was used as an internal standard. VM-26, in a methanolic solution, was transferred into a polypropylene tube. The plasma (0.25–1 cc) was added after evaporating the methanol. After vortexing, dichloromethane (3 cc) was added. The extraction mixture was shaken and centrifuged (2500 rpm). The plasma was then removed and the dichloromethane was evaporated. The residue was redissolved by vortexing and sonification in 200 μl of the mobile phase. The mobile phase consisted of methanol/0.065 M phosphate buffer (60 : 40, v/v); 10–50 μl were injected in the HPLC system. We used a phenyl Bondapack column (Waters, Bedford, MA 01730, U.S.A.). The flow rate was 1 ml/min. Using this procedure the recovery was higher than 90%.

The electrochemical detector has been described elsewhere [12]. We measured at a constant electrode potential of + 0.60V (vs Ag/AgCl). The correlation coefficient of a standard plot, constructed by measuring seven concentrations in the expected range, was 0.99. Our limit of quantitation was 10 ng VP-16-213/ml of plasma.

Calculation of pharmacokinetic data

The area under the plasma concentration vs time curve (AUC) was calculated with the trapezoidal rule and the halflife time with the stripping method [13]. The mean residence time (MRT) was calculated according to van Rossum [14]. Total body clearance (Cl_{tot}) was calculated from $Cl_{tot} = D/AUC$ in which D is the dose and AUC is the area under the plasma concentration vs time curve. The distribution volume belonging to the beta phase was calculated according to Gibaldi and Perrier [13].

RESULTS AND DISCUSSION

Typical plasma concentration vs time curves of two patients are represented in Fig. 1.

The calculated pharmacokinetic parameters for VP-16-213 for 7 patients have been summarized in Table 2. No drug was detected in the pretreatment samples on day 8. In the first 25 hr the plasma levels of VP-16-213 describe a biexponential decay curve. On day 1 and 8 the individual differences in the patients and the mean differences were very small. Because of this, it was not reasonable to look for a correlation with toxicity or response in these few patients. Our data show that there is no influence of doxorubicin and cyclophosphamide administration on VP-16-213 plasma disappearance rate for the applied schedule.

Some comparative data of previous studies are summarized in Table 3. Our data are in accordance with those of earlier reports. Protein binding, metabolism and renal clearance are important variables of VP-16-213 and these can be influenced by other drugs. Although the body is a very complex system, only a few processes determine the shape and the height of the plasma concentration vs time curve [15]. Apparently these factors are not significantly influenced by the presence of the other drugs, resulting in a change of pharmacokinetic parameters of VP-16-213.

Protein binding of VP-16-213 is high (94%) [7]; doxorubicin is bound for 70% [16] and cyclophosphamide for 12–24% [17]. Both doxorubicin [18] and cyclophosphamide [19] are metabolized extensively. *In vivo* metabolism of VP-16-213 is largely unknown, but may be considerable. After administration of radiolabelled VP-16-213 50% of the radioactivity did not leave the body within 72 hr [20]. For high-dose VP-16-213 Holthuis *et al.* have found considerable amounts of the glucuronide in urine and low amounts of the *cis*-isomer and the *trans*-hydroxy acid [10]. Interesting metabolites, responsible for the antineoplastic effect, have been suggested by Van Maanen *et al.* [21]. Sinkule *et al.* [22] found a strong correlation between elevated liver enzymes in plasma and diminished systemic clearance.

Kidney clearance is important in the elimination of VP-16-213 (about 30% of the administered dose) [5, 21]. Also at this level the other drugs could have an effect since both doxorubicin [19] and cyclophosphamide [20] and metabolites are excreted in the urine.

CONCLUSION

This study has provided evidence that important factors, such as protein binding, metabolism and renal clearance were not affected by the other cytostatic drugs to the degree that the change resulted in a different plasma concentration vs time curve.

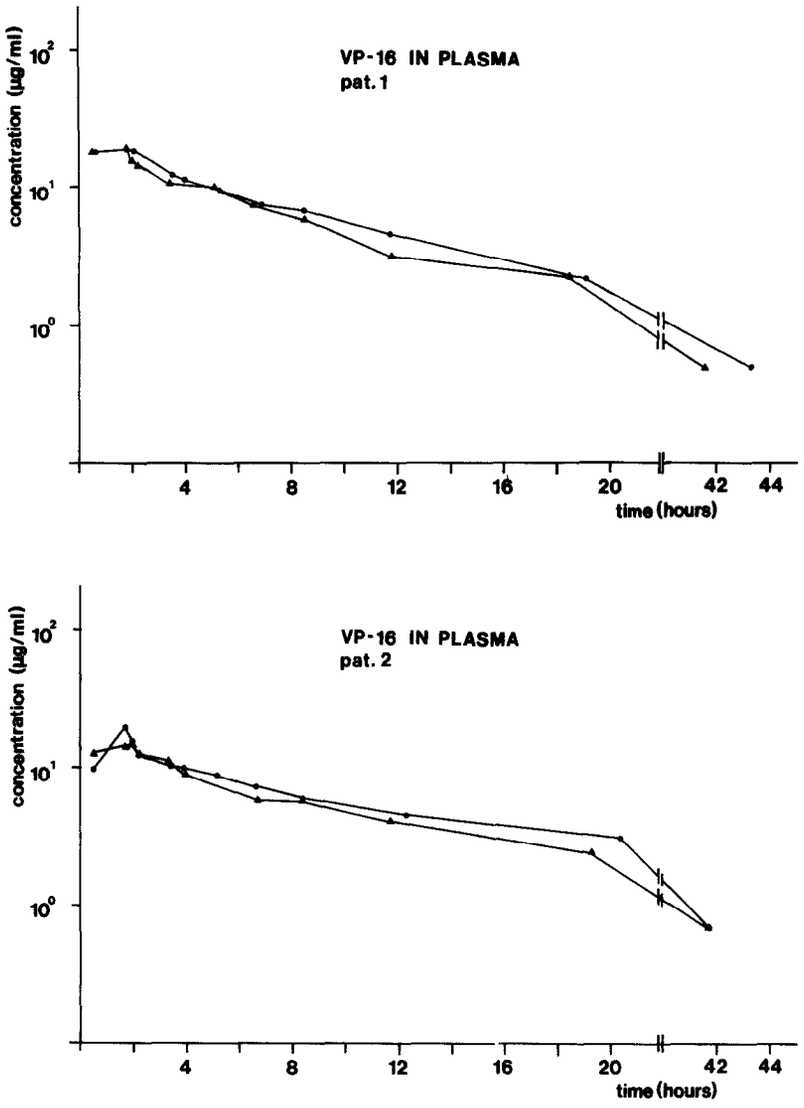

Fig. 1. *Typical plasma concentration vs time curves of* 2 *patients at day* 1 (●) *and day* 8 (▲) *during and after VP*-16-213 *infusion* (*dose* 100 mg/sqm).

Table 2. *Calculated pharmacokinetic parameters for VP*-16-213

Patients	Day	$t_{\frac{1}{2}}$ (min)	AUC (mg.hr/l)	MRT (min)	Cl_{tot} (ml/min.sqm)	V_d (l/sqm)
1	1	346	102	418	16.3	8.2
	8	316	88	392	18.9	8.6
2	1	384	104	478	15.9	8.8
	8	409	92	457	18.0	10.7
3	1	291	76	396	21.8	9.2
	8	348	76	369	22.0	11.0
4	1	346	82	418	20.3	8.7
	8	316	105	392	15.8	7.2
5	1	287	64	336	26.1	10.8
	8	311	56	320	29.9	13.4
6	1	290	105	479	15.9	6.7
	8	307	102	510	16.3	7.3
7	1	262	69	364	24.1	9.1
	8	279	78	325	21.4	8.6
Mean	1	315	86	413	20.1	8.7
	8	327	85	395	20.3	9.5

Table 3. Comparative data of previous studies

Study	Dose (mg/sqm)	No. of patients	$t_{\frac{1}{2}}$ (hr)	Cl_{tot} (ml/min.sqm)	Ref.
Hande	400–800	12	8.05	28.0	[5]
D'Incalci	200–250	14	5.80	19.5	[6]
Allen	100–200	10	7.05	26.8	[7]
Scalzo	80	6	5.95	19.2	[8]
Ho	40–120	12	3–14	16±7	[9]

The previous administration of VP-16-213 itself did not affect the pharmacokinetics either by e.g. induction or change in metabolism or clearance.

Acknowledgements—Helen Gall is gratefully acknowledged for her excellent help with collection of patient samples. Miguel Stuurman is thanked for skilful technical advice.

REFERENCES

1. O'Dwyer PJ, Leyland JB, Alonso MT, Marsoni S, Wittes RE. Etoposide, current status of an active anticancer drug. *New Engl J Med* 1985, **312**, 692–700.
2. Bunn PA. The role of chemotherapy in small-cell lung cancer. In: Issell BF, Muggia FM, Carter SK, eds. *Etoposide (VP-16), Current Status and New Developments*. Orlando, Academic Press, 1984, 141–163.
3. Cavalli F, Goldhirsch A, Joss R. Single agent activity and European experiences with etoposide in the treatment of small-cell lung cancer. In: Issell BF, Muggia FM, Carter SK, eds. *Etoposide (VP-16), Current Status and New Developments*. Orlando, Academic Press, 1984, 141–163.
4. Aisner J, Whitacre M, Van Echo DA, Fuks JZ. Cyclophosphamide, doxorubicin and etoposide (CAE) for the treatment of small-cell lung cancer. In: Issell BF, Muggia FM, Carter SK, eds. *Etoposide (VP-16), Current Status and New Developments*. Orlando, Academic Press, 1984, 171–182.
5. Hande KR, Wedlund PJ, Noone RM, Wilkinson GR, Greco FA, Wolff SN. Pharmacokinetics of high-dose etoposide (VP-16-213), administered to cancer patients. *Cancer Res* 1984, **44**, 379–382.
6. D'Incalci M, Farina P, Sessa C *et al.* Pharmacokinetics of VP-16-213 given by different administration methods. *Cancer Chemother Pharmacol* 1982, **7**, 141–145.
7. Allen LM, Creaven PJ. Comparison of the human pharmacokinetics of VM-26 and VP-16-213. Two antineoplastic epipodophyllotoxin glucopyranoside derivatives. *Eur J Cancer* 1975, **11**, 697–707.
8. Scalzo AJ, Comis R, Fitzpatrick A *et al.* VP-16 pharmacokinetics in adults with cancer as determined by a new high-pressure liquid chromatography (HPLC) assay. *Proc Am Soc Clin Oncol* 1982, **1**, 129, abstract.
9. Ho DHW, Kanellopoulos KA, Yap HY *et al.* Clinical pharmacology of etoposide by radioimmunoassay. *Proc Am Assoc Cancer Res* 1983, **24**, 131, abstract.
10. Holthuis JJM, Postmus PE, van Oort WJ *et al.* Pharmacokinetics of high dose etoposide (VP-16-213). *Eur J Cancer Clin Oncol*, 1986, **22**, 1149–1155.
11. Holthuis JJM, Römkens FMGM, Pinedo HM, van Oort WJ. Plasma assay for the antineoplastic agent VP-16-213 (etoposide) using high performance liquid chromatography with electrochemical detection. *J Pharm Biomed Anal* 1983, **1**, 89–97.
12. Lankelma J, Poppe H. Design and characterization of a coulometric detector for high speed, high efficiency liquid chromatography. *J Chromatogr* 1976, **125**, 375–388.
13. Gibaldi M, Perrier D. *Pharmacokinetics*. New York, Marcel Dekker, 1975.
14. Van Rossum JM, Van Ginneken CAM. Pharmacokinetic system dynamics. In: Gladtke E, Heimann G, eds. *Pharmacokinetics*. Stuttgart, Gustav Fisher, 1980, 53–73.
15. Van Rossum JM, Van Lingen G, Maes RA. Pharmacokinetics from a dynamical systems point of view. In: Stoechel H, ed. *Quantitation, Modelling and Control in Anaesthesia*. Stuttgart, George Thieme, 1985, 72–98.
16. Eksborg S, Ehrsson H, Ekqvist B. Protein binding of anthraquinone glycosides with special reference to adriamycine. *Cancer Chemother Pharmacol* 1982, **10**, 7–10.
17. Grochow LB, Colvin M. Clinical pharmacokinetics of cyclophosphamide. In: Ames MM, Powis G, Kovach JS, eds. *Pharmacokinetics of Anticancer Agents in Humans*. Amsterdam, Elsevier, 1983, 135–154.
18. Takanashi S, Buchur NR. Adriamycin metabolism in man. *Drug Metab Disp* 1976, **4**, 79–87.
19. Bagley CM Jr, Bostick FW, De Vita VT Jr. Clinical pharmacology of cyclophosphamide. *Cancer Res* 1973, **33**, 226–233.

20. Creaven PJ, Allen LM. EPEG, a new antineoplastic epipodophyllotoxin. *Clin Pharmacol Ther* 1975, **18**, 221–226.
21. Van Maanen JMS, de Ruiter C, Kootstra PR *et al.* Inactivation of ØX174 DNA by the *ortho*-quinone derivative or its reduction product of the antitumour agent VP-16-213. *Eur J Cancer Clin Oncol* 1985, **21**, 1215–1218.
22. Sinkule JA, Hutson P, Hayes FA, Etcubanas E, Evans W. Pharmacokinetics of etoposide (VP-16) in children and adolescents with refractory solid tumours. *Cancer Res* 1984, **44**, 3109–3113.