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Advances in Brief

Predictive Performance of a Pharmacodynamic Model for Oral Etoposide[1]

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Abstract

The objective of this work was to prospectively validate a pharmacodynamic model for 21-day oral etoposide. The model had been developed in 27 untreated patients with stage IIIB or IV non-small cell lung cancer. Treatment consisted of 50 mg/m^2/day, p.o., etoposide for 21 days in combination with 100 mg/m^2, i.v., cisplatin on day 1 every 28 days for up to 6 courses. Weekly evaluations included etoposide plasma concentrations (E_c, μg/ml) before the daily dose and WBC and neutrophil counts (ANC, $10^3/\mu l$). The relationship between E_c and the pretreatment (WBC_p, ANC_p) and nadir counts (WBC_n, ANC_n) in the first course was described as follows:

$$WBC_n = 0.35\ (1 + WBC_p \times e^{-1.12 \times E_c}))$$

$$ANC_n = 0.32\ (1 + ANC_p \times e^{-2.47 \times E_c})$$

The same study criteria were used to enter 26 additional patients, and 21 were evaluable for pharmacodynamics (5 had incomplete data). Predicted nadir counts were not significantly different from observed nadir counts (paired t test, $P > 0.4$). There were 12 and 7 patients correctly predicted to be above and below, respectively, the clinically important ANC_n of 0.5 × $10^3/\mu l$. The model performed reliably, and therapeutic drug monitoring appears warranted in future studies.

Introduction

Current clinical research tries to define the schedule dependency of etoposide. Traditionally, etoposide has been given as short courses of therapy in the order of 3 to 5 days. Based on the hypothesis that sustained inhibition of topoisomerase II may be more efficacious, prolonged treatment regimens have been developed and shown to be well tolerated (1). Such regimens commonly use oral etoposide for 21 days (1–8), but long-term continuous infusion regimens have also been explored (9–12). Hande *et al.* (13) investigated the bioavailability of etoposide and found that absorption is better at lower oral doses and that wide interpatient variability in bioavailability exists (13). Because of this variability, the relationship between etoposide concentrations and the ensuing hematological toxicity was determined in a population of patients who all received the same dose of oral etoposide (14). Specifically, 27 previously untreated patients with stage IIIB or IV non-small cell lung cancer received 50 mg/m^2/day etoposide for 21 days in combination with 100 mg/m^2 cisplatin on day 1 (every 28 days for up to 6 courses). Weekly evaluations included etoposide plasma concentrations before the daily dose (trough levels in μg/ml) and WBC[3] and neutrophil counts. The relationship between drug concentrations and the pretreatment and nadir counts in the first course were described by the following pharmacodynamic model for which counts were expressed as $10^3/\mu l$:

$$WBC_n = 0.35\ (1 + WBC_p \times e^{-1.12 \times E_c})$$

$$ANC_n = 0.32\ (1 + ANC_p \times e^{-2.47 \times E_c})$$

The objective of the continuation of this work was to validate the pharmacodynamic model prospectively.

Materials and Methods

Patients and Treatment. Patients with advanced non-small cell lung cancer were treated according to the same protocol on which the pharmacodynamic model was based (14). Eligibility criteria included a histological or cytological diagnosis of non-small cell lung cancer (stage IIIB or IV), measurable or evaluable disease, no prior chemotherapy, age 18–80 years, and performance status 0–2 (Eastern Cooperative Oncology Group criteria). Laboratory criteria for entry on the protocol were WBC, ≥4000/μl; hemoglobin, ≥10 g/dl; platelets, ≥100,000/μl; serum creatinine, <2 mg/dl; creatinine clearance, >60 ml/min; blood urea nitrogen, <1.5 × normal; and bilirubin, <1.5 × normal. Treatment consisted of 50 mg/m^2/day etoposide orally for 21 days in combination with 100 mg/m^2 i.v. cisplatin on day 1 every 28 days for up to 6 courses. Weekly plasma samples were obtained to measure etoposide trough concentrations before the morning dose. Etoposide was measured by high-performance liquid chromatography with UV detection (14). Hematological toxicity was assessed weekly in all patients with complete blood counts and differentials. The primary study purpose was to test the predictive performance of the pharmacodynamic model. Secondary end points were toxicity, tumor response, and survival.

Biostatistics. Validation of a prediction model involves testing goodness of fit of the model to data that were not used in model development. The prospective patient sample was drawn from the same population so that no differences in patient characteristics would occur. The aim was to assess how close predicted and actual nadir values were for a prospective patient sample. The deviation of predicted to actual nadir values is the residual. Under the normality assumptions, (*a*) the expected value of the residuals is 0, and (*b*) the residuals are not correlated with the predicted values. These assumptions provided the basis for two null hypotheses. The respective alternative hypotheses considered the effects of intrinsic curvature on the residuals: (*a*) the residuals are biased away from 0, and (*b*) there is a linear relationship between the residuals and the predicted values. The first hypothesis was tested with a paired t test on the differences between the actual and predicted nadir values. Paired t tests to determine the presence of significant bias were conducted on residuals from both the original pharmacodynamic model and the model for ANC_n after logarithmic transformation. The second hypothesis was tested by inspecting the residual plot of the residuals against the predicted nadir values and testing for a nonzero Spearman correlation coefficient. In this study, the pharmacodynamic model was validated with two data sets containing values for etoposide plasma concentrations and counts for WBCs and neutrophils (before treatment and nadir counts). The primary data set was obtained from a prospective patient sample and comprised values for the first treatment course only. The secondary data set comprised values from the subsequent courses (*i.e.*, 2–6) from the combined patient sample in the previous and current study.

Evaluation of the clinical usefulness of the model was based on correct prediction of grade 4 leukopenia or neutropenia from measured etoposide concentrations. Life-threatening, grade 4 toxicity on WBCs and neutropenia is defined as $WBC_n < 1.0 \times 10^3/\mu l$ and $ANC_n < 0.5 \times 10^3/\mu l$ (National Cancer Institute, Common Toxicity Criteria). Agreement between actual and predicted

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[3] The abbreviations used are: WBC_p (WBC_n), WBC count pretreatment (at nadir); ANC, absolute neutrophil count; ANC_p (ANC_n), ANC pretreatment (at nadir); E_c, etoposide concentration; CI, confidence interval.

Table 1 *Patient characteristics, tumor response, and pharmacodynamic model*

	Model development	Model validation
Patients entered	32[a]	26
Age (yr)		
Median	65	63
Range	39–77	43–74
Male/female gender	30/2	24/2
White/black race	23/9	16/10
Performance status		
0	2	3
1	23	14
2	7	9
Stage III B	6	8
Stage IV	26	18
Squamous cell carcinoma	16	13
Adenocarcinoma	10	11
Large cell carcinoma	6	2
Serum albumin (g/dl)		
Median	3.4	3.6
Range	2.0–4.1	2.4–4.5
Etoposide concentration (μg/ml)		
Median	0.88	0.80
Range	0.13–1.89	0.11–1.73
Complete response	3	0
Partial response	10	9
Objective response rate (%)	41	35
Survival (mo)		
Median	4	4
Range	1–26+	1–20+
Patients with pharmacodynamics	27	21
No pharmacodynamics due to		
Noncompliance	1	2
Rapid tumor progression	3	1
No nadir counts	1	2

[a] Results are numbers of patients unless other unit of measure is given.

toxicity was evaluated with the κ statistic. The model as written reflects a nonlinear situation. In addition, a linearization technique was applied to estimate etoposide concentrations for selected values of pretreatment counts. Pretreatment counts in all 48 patients with pharmacodynamic data were ranked and the following percentiles (minimum to maximum) selected: 0, 25, 50, 75, and 100. For each selected pretreatment count, the expected etoposide concentration and 95% CI associated with grade 4 toxicity was estimated using inverse regression and prediction techniques (15, 16). The κ statistic was used to evaluate the concordance between actual and predicted grade 4 neutropenia (17).

Results and Discussion

As reviewed by Mick and Ratain (18), the most preferable means to test the predictive performance of a model is to validate the model on an independent set of data. This approach was followed. The pharmacodynamic model was based on a group of 32 patients (14). In a subsequent group of 26 patients the model was validated prospectively (Table 1). No significant differences were apparent in the patient characteristics between the original and the prospective sample. The overall objective response rate in the 58 patients was 38% (95% CI, 26–50%), but the survival was poor (Table 1). The patient with the longest survival time in the group for model validation developed refractory anemia with excess blasts associated with partial loss of the long arm of chromosome 7 which occurred 20 months after chemotherapy.

Table 2 shows the etoposide concentrations and the observed and predicted counts for 21 patients with complete pharmacodynamic data. The nadir counts for WBC and ANC that were predicted by the pharmacodynamic model were not significantly different from observed nadir counts (paired t test, $P > 0.4$). The SD of the residuals was $1.05 \times 10^3/\mu l$ for WBC and $0.59 \times 10^3/\mu l$ for ANC, and this is a measure of precision of the model (18). The deviations of observed and predicted nadir counts (residuals) were compared to the observed nadir counts. The residuals and WBC_n counts were not correlated ($r = -0.06$; $P > 0.9$). The residuals and ANC_n were correlated ($r = 0.49$; $P < 0.03$) in that the residuals were higher for high ANC_n (low etoposide concentrations) compared to low ANC_n (high etoposide concentrations). If a logarithmic transformation for the ANC is used, the linearized model is:

$$\ln(ANC_n) = -0.11 + 0.24 \ln(ANC_p) + 1.66\, E_c$$

Figure 1 shows the relationship between etoposide concentrations and ANC_n. Based on the linearized model, the residuals and ANC_n were not correlated; thus, the correlation detected above was an effect of intrinsic curvature related to the nonlinear form of the model (15).

The dose-limiting toxicity of treatment was neutropenia as reported before (14), and this was described by the pharmacodynamic model. Using the clinically important cutoff point for grade 4 neutropenia, an ANC_n of $0.5 \times 10^3/\mu l$, we correctly predicted that 12 and 7 patients had values above and below this value, respectively (Table 2). Measured as a κ statistic, the concordance between actual and predicted grade 4 neutropenia was 0.8 ($P < 0.03$) which indicates excellent agreement (17). Expressed another way, characteristics of the model were 78% for sensitivity (exact 95% CI, 40–97%), 100% for speci-

Table 2 *Etoposide concentrations, observed leukocyte and neutrophil counts, and nadir counts predicted by the pharmacodynamic model for 21 assessable patients*[a]

Evaluable patient	Race/ gender	E_c (μg/ml)	Observed		Predicted	Observed		Predicted
			WBC_p	WBC_n	WBC_n	ANC_p	ANC_n	ANC_n
1	W/M	1.73	13.4	1.2	1.0	11.7	0.2	0.4
2	W/M	0.43	7.5	1.6	2.0	5.1	0.7	0.9
3	B/M	0.19	6.6	2.5	2.2	3.6	1.3	1.0
4	W/F	0.98	8.6	0.3	1.3	6.1	0.1	0.4
5	B/M	0.30	8.8	3.0	2.6	6.0	1.8	1.2
6	W/F	0.69	15.3	2.5	2.8	10.9	0.8	0.9
7	B/M	0.91	10.9	0.4	1.7	8.0	0.3	0.6
8	B/M	1.49	7.9	0.4	0.9	5.3	0.2	0.4
9	B/M	0.12	15.7	8.3	5.2	12.5	5.3	3.3
10	W/M	0.33	5.4	2.0	1.7	2.8	1.2	0.7
11	W/M	0.11	8.4	3.5	2.9	6.2	2.4	1.8
12	W/M	1.00	14.4	1.0	1.9	11.1	0.5	0.6
13	W/M	0.92	15.9	0.6	2.3	13.9	0.4	0.8
14	B/M	0.28	12.3	2.7	3.5	9.2	2.3	1.8
15	B/M	1.29	5.7	0.4	0.8	2.4	0.1	0.3
16	W/M	0.94	10.2	1.8	1.6	7.7	0.6	0.6
17	W/M	0.25	17.2	3.5	4.9	13.3	1.6	2.6
18	W/M	1.38	6.8	0.9	0.9	5.6	0.4	0.3
19	B/M	1.29	3.4	1.3	0.6	1.7	0.2	0.3
20	W/M	1.63	12.5	0.2	1.0	7.9	0.01	0.3
21	W/M	0.52	14.1	2.5	3.1	9.8	1.4	1.2
Median		0.91	10.2	1.6	1.9	7.7	0.6	0.7

[a] All counts expressed as $10^3/\mu l$.

ficity (exact lower one-sided 95% CI, 83%), 100% for positive predictive value (exact lower one-sided 95% CI, 72%), and 86% for negative predictive value (exact 95% CI, 57–98%). In 2 patients, $ANC_n < 0.5 \times 10^3/\mu l$ was not predicted (Table 2, patients 7 and 13). Patient 7 had the lowest albumin concentration of the group (2.4 g/dl), and patient 13 had a pretreatment performance status of 2. Although performance status and albumin concentration were not found to be confounding variables during model development (14), their contribution was evaluated again, and neither improved the predictive performance of the model. Performance status and albumin concentration were not related to each other ($P > 0.3$), to etoposide concentration ($P > 0.6$), or hematological toxicity ($P > 0.4$).

Since it has long been recognized that the ANC in healthy white Americans is lower than in black Americans (19, 20), racial differences were evaluated in the combined sample including all patients. The ANC_p was significantly ($P < 0.03$) lower for black males than for white males with means of 6.0 and 8.1 $\times 10^3/\mu l$, respectively. However, the ANC_n was not significantly different in black *versus* white males ($P > 0.2$). In addition, no significant difference was observed for ANC_p or ANC_n in females ($P > 0.2$), and no significant differences for ANC_p ($P < 0.07$) or ANC_n ($P > 0.3$) were observed for males *versus* females. The etoposide concentrations were not significantly different in whites *versus* blacks or males *versus* females ($P > 0.3$).

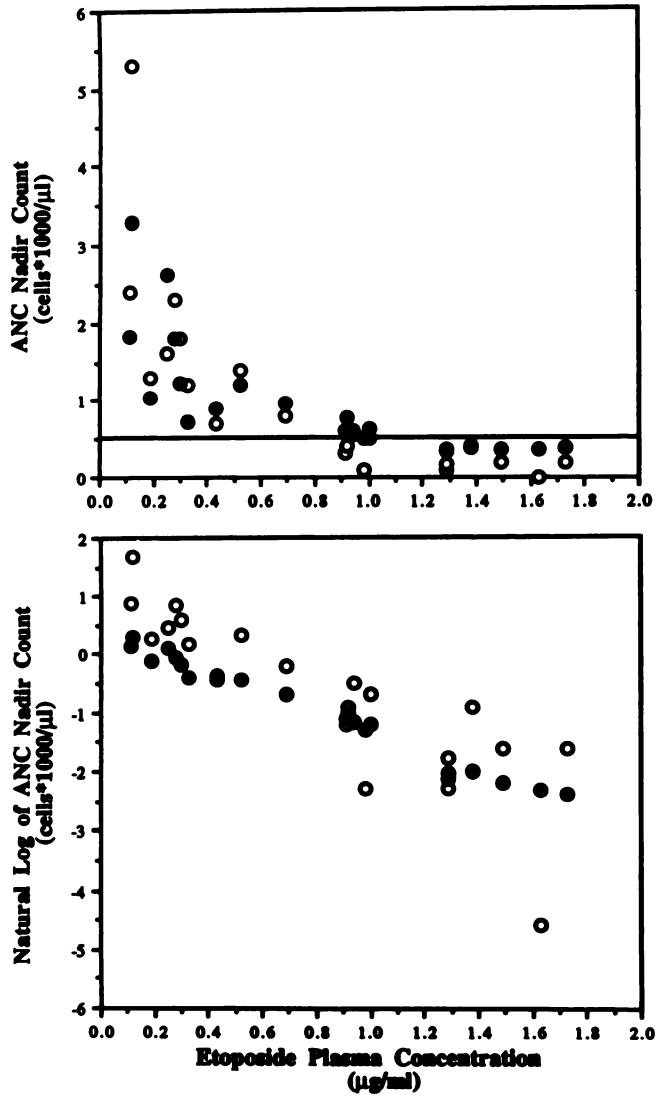

Fig. 1. Two-dimensional representations of the relationship between etoposide plasma concentrations and ANC based on the model $ANC_n = 0.32\ (1 + ANC_p \times e^{-2.47 \times E_c})$ (*top*) and the linearized model $\ln(ANC_n) = -0.11 + 0.24 \ln(ANC_p) + 1.66\ E_c$ (*bottom*). ○, actual counts; ●, predicted counts.

Table 3 *Etoposide concentrations (μg/ml) associated with grade 4 leukopenia[a] or neutropenia[b] in the first course, given selected pretreatment counts ($10^3/\mu l$)*

	Leukocytes		Neutrophils	
Percentile	WBC_p	E_c	ANC_p	E_c
0	3.4	0.87	1.7	0.75
25	7.9	1.33	5.3	1.00
50	9.4	1.42	6.7	1.05
75	13.4	1.62	10.1	1.14
100	17.2	1.72	14.6	1.22

[a] $WBC_n < 1.0 \times 10^3/\mu l$; 95% confidence limits for etoposide concentrations for all selected WBC_p, 0.27 to 3.88 μg/ml.

[b] $ANC_n < 0.5 \times 10^3/\mu l$; 95% confidence limits for etoposide concentrations for all selected ANC_p, 0.28 to 1.83 μg/ml.

Figure 1 shows the relationship between ANC_n and etoposide concentrations as a two-dimensional graph with a horizontal line at $ANC_n = 0.5 \times 10^3/\mu l$. The model, however, is three-dimensional with ANC_p as the third variable. Whether grade 4 toxicity develops depends not only on the etoposide concentration but also on ANC_p. The higher the pretreatment count, the higher the etoposide concentration that is tolerated. Table 3 lists etoposide concentrations that, given specific pretreatment counts, can be expected to lead to grade 4 toxicity after the first treatment course. For instance, grade 4 neutropenia may be expected with a relatively low E_c of 0.75 μg/ml if the ANC_p is only 1.7 $\times 10^3/\mu l$. On the other hand, it takes a higher E_c of 1.14 μg/ml to produce grade 4 neutropenia if the ANC_p is 10.1 $\times 10^3/\mu l$. The etoposide concentration that, if exceeded, was associated with grade 4 neutropenia in 88% of all patients (previous and current study) is 1.00 μg/ml. A patient with an etoposide concentration ≥ 1.0 μg/ml was almost four times as likely (relative risk, 3.9; 95% CI, 2.1 to 7.3) to develop grade 4 neutropenia as a patient with a concentration < 1.0 μg/ml.

In conclusion, the model performed reliably. Although the pretreatment ANC was lower for black than for white males, the nadir counts were not significantly different, and the model predicted ANC_n without significant difference for race. Therapeutic drug monitoring is the objective of an ongoing study.

References

1. Greco, F. A., Johnson, D. H., and Hainsworth J. D. Chronic daily administration of oral etoposide. Semin. Oncol., *17:* 71–74, 1990.
2. Hainsworth, J. D., Johnson, D. H., Frazier, S. R., and Greco F. A. Chronic daily administration of oral etoposide—a phase I trial. J. Clin. Oncol., *7:* 396–401, 1989.
3. Johnson, D. H., Greco, F. A., Strupp, J., Hande, K. R., and Hainsworth J. D. Prolonged administration of oral etoposide in patients with relapsed or refractory small-cell lung cancer: a phase II trial. J. Clin. Oncol., *8:* 1613–1617, 1990.
4. Einhorn, L. W., Pennington, K., and McClean, J. Phase II trial of daily oral VP-16 in refractory small cell lung cancer: a Hoosier Oncology Group study. Semin. Oncol., *17:* 32–35, 1990.
5. Smit, E. F., Carney, D. N., Harford, P., Sleijfer, D. T., and Postmus, P. E. A phase II study of oral etoposide in elderly patients with small cell lung cancer. Thorax, *44:* 631–633, 1989.
6. Miller, J. C., and Einhorn, L. H. Phase II study of daily oral etoposide in refractory germ cell tumors. Semin. Oncol., *17:* 36–39, 1990.
7. Hainsworth, J. D., Johnson, D. H., Frazier, S. R., and Greco, F. A. Chronic daily administration of oral etoposide in refractory lymphoma. Eur. J. Cancer, *26:* 818–821, 1990.
8. Waits, T. M., Johnson, D. H., Hainsworth, J. D., Hande, K. R., Thomas, M., and Greco, F. A. Prolonged administration of oral etoposide in non-small cell lung cancer: a phase II trial. J. Clin. Oncol., *10:* 292–296, 1992.
9. Robert, F., Wheeler, R. H., Miller, A. A., and Lee, J. Y. Phase I and pharmacologic study of continuous intravenous (IV) etoposide (VP-16) administered as a 7 or 21-day infusional schedule (SCH). Proc. Am. Soc. Clin. Oncol., *11:* 131, 1992.
10. Levin, J., Parkhurst, G., and Bonomi, P. Serum drug levels following continuous infusion etoposide. Proc. Am. Soc. Clin. Oncol., *12:* 166, 1993.
11. Thompson, D. S., Hainsworth, J. D., Hande, K. R., Johnson, D. H., and Greco, F. A.

Prolonged continuous infusion of low dose etoposide: seeking the best dose and schedule. Proc. Am. Soc. Clin. Oncol., *12:* 134, 1993.
12. Kunitoh, H., and Watanabe, K. A phase I/II study of long-term continuous infusion of etoposide combined with cisplatin in advanced non-small cell lung cancer (NSCLC). Proc. Am. Soc. Clin. Oncol., *12:* 136, 1993.
13. Hande, K. R., Krozely, M. G. Greco, F. A., Hainsworth, J. D., and Johnson, D. H. Bioavailability of low-dose oral etoposide. J. Clin. Oncol., *11:* 374–377, 1993.
14. Miller, A. A., Tolley, E. A., Niell, H. B., Griffin, J. P., and Mauer, A. M. Pharmacodynamics of prolonged oral etoposide in patients with advanced non-small cell lung cancer. J. Clin. Oncol., *11:* 1179–1188, 1993.
15. Seber, G. A. F., and Wild, C. J. Nonlinear Regression: Measures of Curvature of Nonlinearity. New York: John Wiley and Sons, 1989.
16. Sokal, R. R., and Rohlf, F. J. Biometry: The Principles and Practice of Statistics in Biological Research, Ed. 2. New York: W. H. Freeman and Co., 1981.
17. Agresti, A. Categorical Data Analysis: Models for Matched Pairs. New York: John Wiley and Sons, 1990.
18. Mick, R., and Ratain, M. J. Statistical approaches to pharmacodynamic modeling: motivations, methods, and misperceptions. Cancer Chemother. Pharmacol., *33:* 1–9, 1993.
19. Broun, G. O., Herbig, F. K., and Hamilton, J. R. Leukopenia in negroes. N. Engl. J. Med., *275:* 1410–1413, 1966.
20. Lichtman, M. A. Classification and clinical manifestations of neutrophil disorders. *In:* W. J. Williams, E. Beutler, A. J. Erslev, and M. A. Lichtman (eds.), Hematology, Ed. 4, p. 802. New York: McGraw-Hill, 1990.