Treatment-related Parameters Predicting Efficacy of Lym-1 Radioimmunotherapy in Patients with B-Lymphocytic Malignancies

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ABSTRACT

This study was designed to evaluate dosimetric, pharmacokinetic, and other treatment-related parameters as predictors of outcome in patients with advanced B-lymphocytic malignancies. Fifty-seven patients were treated with radiolabeled Lym-1 antibody in early phase trials between 1985 and 1994. Logistic regression and proportional hazards models were used to evaluate treatment parameters for their ability to predict outcome, taking into account patient risk group based on Karnofsky performance status and serum lactic dehydrogenase. The occurrence of a partial or complete response (31 of 57 patients) and development of human antimouse antibody (HAMA) predicted improved survival using a time-dependent proportional hazards model. The final multivariate model for survival with parameters significant at $P \leq 0.05$ included overall response and pretreatment risk group. Although some of the dosimetric and pharmacokinetic parameters were predictive in univariate analyses, only longer half-time of radionuclide in the blood showed any indication of improved prediction beyond that provided by the lactic dehydrogenase/Karnofsky performance status-based risk groups. Splenic volume, splenectomy, and malignant tissue Lym-1 reactivity were not contributory. In this patient group, the effect of radiolabeled Lym-1 treatment as indicated by measurable tumor response was associated with improved survival. Development of HAMA was also associated with improved survival, indicating that concern about HAMA should not preclude exploration of radioimmunotherapy. Although dosimetry has a role in determining safety based on dose to normal organs, when adjusted for baseline clinical features, dosimetric and pharmacokinetic parameters showed limited ability to improve outcome prediction.

INTRODUCTION

To effectively evaluate novel therapies such as RIT, it is important to identify and to take into account patient characteristics that might influence outcome. The need to determine factors that predict therapeutic outcome in patients with non-Hodgkin’s lymphoma has led to a number of studies (1–6), including the recent IPFP (7). In the IPFP, over 2000 patients treated with doxorubicin-based combination chemotherapy were evaluated to determine which pretreatment clinical features predicted for improved survival. Age, tumor stage, serum LDH, performance status, and number of extranodal disease sites were found to be significant in a multivariate stepwise analysis and were used to construct a model that predicted patients’ relative risk of death.

Candidates for radioimmunotherapy represent a different patient group than those considered in the IPFP. The histological types of the RIT patients are less homogeneous, and extent of disease is heavily weighted toward Ann Arbor stages 3 and 4. Furthermore, RIT patients have been extensively treated with combination chemotherapy (8–10) and external beam radiotherapy. Additionally, RIT represents a different therapeutic modality than chemotherapy or standard radiotherapy, and it cannot be assumed that the same set of pretherapy parameters would be predictive of outcome.

For that reason, the predictive value of 21 pretherapy parameters were evaluated in patients with B-lymphocytic malignancies that were subsequently treated with $^{111}$I- or $^{67}$Cu-labeled Lym-1 antibody (11–16). It was found that these patients with advanced disease could effectively be categorized into low, medium, and high risk groups for early failure and death based on LDH (percentage above normal range) and performance status, two of the five parameters found to be of importance in the IPFP. As an indication of the population differences, it is of interest that the IPFP risk factor of stage (I and II versus III and IV) was not applicable to these patients because they all were stage III or IV.

The present report evaluates parameters specifically relevant to RIT, including HAMA, dosimetry, and pharmacokineti-
ics. To reduce the possibility that these treatment-related parameters may serve as surrogates for overall patient status, general baseline risk factors were also included in the analyses. This type of analysis is critical to the evaluation of potential therapies, providing information relative to appropriate patient selection and radionuclide dosage planning.

MATERIALS AND METHODS

Patient Selection. Fifty-seven heavily pretreated, adult patients with B-lymphocytic malignancies whose disease was progressing despite standard therapy were entered into clinical trials of Lym-1-based RIT between September 1985 and March 1994. Fifty-three patients received treatment with $^{131}$I-Lym-1 and 4 received treatment with $^{67}$Cu-Lym-1. The clinical trials were early phase studies (two were maximum tolerated dose trials). Patient doses were based on the assigned dose level at the time of patient enrollment. RIT doses were repeated at 2-6-week intervals. The median number of infusions was three (range, 1–16). Patients were evaluated immediately before and approximately 1, 4, 8, 12, 24, 36, and 48 weeks after radiolabeled antibody infusion and at 6-month intervals thereafter. Before treatment, all patients were advised of the investigational nature of the study and signed an informed consent for protocols that were approved by the University of California at Davis Human Subjects and Radiation Use Committees under an Investigational New Drug authorization from the United States Food and Drug Administration.

Fifty-two patients had non-Hodgkin’s lymphoma (11, 32, and 9 classified as low, intermediate, and high grade, respectively, based on the Working Formulation) and 5 had chronic lymphocytic leukemia. There were 34 men and 23 women. Median age was 55 years (range, 30–74). Median KPS was 70 (range, 40–90). The median number of prior chemotherapy regimens was 4 (range, 1–13). All 57 patients had malignancies that showed evidence for Lym-1 reactivity as demonstrated by immunophenotypic studies.

Patient Risk Groups for Early Failure and Death. For the purpose of this study, patients were categorized into three risk groups based on their LDH and KPS values using information from the proportional hazards model for survival developed previously (11). In that model, the patient's LDH as a ratio of 1, <0.9; 2, 0.9–1.11; 3, 1.12–1.9; and 4, >1.9. From the model, the formula for risk assignment was 0.70 (LDH grade)–0.05 (KPS). Using this model, an increase in LDH grade by one level increased the logarithm of the hazard ratio by 0.7. This increase roughly corresponded to a decrease in KPS of 10 units, which increased the log hazard by 0.5. The relationship between risk groups and histological grades was weak, and all histological grades were represented in each risk group.

Quantitative HAMA. Quantitative HAMA assays for avidity for a membrane-associated antigen found on most malignant B cells (18). $^{131}$I-labeled Lym-1 was prepared using chloramine-T at a mass ratio of about 1 μg of chloramine-T:10 μg of Lym-1 (19). $^{67}$Cu-labeled Lym-1 ($^{67}$Cu-2-iminothiolane-6-[p-(bromoacetamido)benzyl]-1,4,8,11-tetraazacyclotetradecane-N,N",N"",N""-tetraacetic acid-Lym-1) was prepared by the methods described previously (20). At least 90% of radioactivity was associated with Lym-1 by immunochemical characterization, and the pharmaceutical exhibited at least 65% immunoreactivity (19–21).

Pharmacokinetics and Radiation Dosimetry. Methods for collecting the pharmacokinetic data and calculating radiation absorbed dose have been described previously (22, 23). Briefly, planar images of conjugate views were acquired immediately, 2–6 h, and daily up to 10 days after administration of the radiopharmaceutical. The amount of activity in organs and tumors was determined using either geometric-mean or effective-point-source methods, depending on whether the source object could be identified on both or one conjugate view (22–25). Coincidence at high counting rates was corrected using a reference source (26). Blood samples were collected during imaging sessions, and the radioactivity in each sample was determined using a gamma well-counter.

Cumulated activity was determined by fitting pharmacokinetic data to a monoexponential function except for the blood, where a biexponential fit was used (22, 23, 27). Radiation dose was calculated based on Medical Internal Radiation Dose formalism (28, 29). A uniform distribution of radionuclide in the tissues was assumed, and Medical Internal Radiation Dose data for “standard man” was used for the organ S factors (30, 31) except for spleen, where actual volume was used. Spleen volume was determined using computed tomography images except for four patients, where single-photon emission computed tomography images were used (32) because computed tomography images of the spleen were not available. Patients that had a splenectomy were considered to have zero volume and zero cumulative activity for the purposes of analysis. For other spleen parameters, these patients were excluded from analysis.

Two patients in this study had an immunoadsorption procedure at 6 h after infusion of the $^{131}$I-Lym-1 dose to reduce the radiation dose to normal tissues (33). The cumulated activity was calculated for these patients by summing the activity over time that was separately determined before and after the immunoadsorption procedure.

Tumor radionuclide uptake measurements were made only for those tumors for which adequate dosimetry could be expected based on previous work using tumor phantoms (34). This included a criterion that the tumor be at least 2 cm in diameter. Eight patients had no tumors that met these criteria; 13 patients had only one tumor that met the criteria. To provide an overall picture of tumor dosimetry when multiple tumors were present, the tumors that had the least and greatest rads/MCi dose were selected for each patient from among those that could be quantitated. When only one tumor met the criteria, the dose to this tumor was included in analysis of lowest tumor dose and greatest tumor dose as predictors of outcome.

To assess radiation to the marrow, penetrating radiation from the body, nonpenetrating radiation from the blood (27, 35),
and nonpenetrating radiation from targeting of the marrow (36) were considered.

**Outcome Assessment.** Complete staging by X-ray, physical exam, and computerized tomography was performed prior to RIT. Tumor sites were reevaluated during treatment and at 1–6-month intervals thereafter.

To qualify as a response, tumor regression must have persisted for at least 4 weeks. Responses were classified as: CR, the complete absence of demonstrable disease including negative bone marrow examination; PR, a decrease in the sum of the products of all tumor dimensions by at least 50%, or all tumor volumes by at least 70%; stable disease; and progression, an increase of at least 25% in the size of any lesion or the development of new lesions.

TTP was measured from the start of treatment. Survival time was measured from the start of treatment to death or the date of the last follow-up for surviving patients.

**RIT-specific Predictive Factors.** All dosimetric and pharmacokinetic parameters potentially related to treatment outcome were considered for analysis. In general, these parameters were selected based on their relevance to either antitumor activity or normal tissue toxicity. A list of these parameters together with definitions are provided in Table 1 (dosimetry) and Table 2 (pharmacokinetics).

**Statistical Methods.** Statistical analyses were done using SAS version 6.08 (37). Analysis of potential predictors of CR and CR + PR (overall response) was done using logistic regression. Analysis of TTP and survival was done using proportional hazards models (38). A multivariate analysis including the LDH/KPS-based risk group was done for all parameters significant at $P = 0.1$ in univariate analysis to evaluate the degree to which the new parameters would increase the ability to predict beyond prediction based on the general information known about the patient. All parameters that continued to be significant at $P = 0.1$ were considered for the final multivariate analysis with retention based on $P \leq 0.05$. Initial multivariate analyses included only those patients with values for all parameters under consideration, as required by the modeling method. The final model included all patients because all patients had the required information based on the final parameters selected.

Parameters that varied with time were included as time-dependent variables in the analysis of TTP and survival (38). For example, patients were not grouped as responders versus nonresponders with time to event calculated separately for the two groups, a method known to be biased (39). Instead, at each follow-up time, all patients without an event to that point were categorized as having had a response by that time or not, and this information was used in the calculation of a hazard ratio.

The usual proportional hazards model assumes a linear increase in the log hazard as a function of the predictor. However, when data include extreme values, the relationship can often be more accurately described assuming a linear relationship with a log transformation of the predictor values. For this reason, natural logarithms of the HAMA titers were used for the purpose of the time-dependent analysis of maximum HAMA.

All patients were assigned a score of zero at baseline (based on the fact that all were considered HAMA negative at start of study). For patients that became HAMA positive, at each subsequent evaluation the log of the maximum HAMA titer to that point was used.

Analyses involving pharmacokinetics and dosimetry (administered radionuclide dose and radiation dose) included only those patients that received $^{131}$I-Lym-1 to prevent confounding due to differences in the radionuclide. The two patients that
received immunoadsorption were also excluded for analyses involving clearances from tumor, total body and blood and extrapolated zero time tumor uptakes because values for these patients were modified by the immunoadsorption.

A number of supplementary analyses were done to confirm the results. These included a second multivariate analysis using the same parameters but including only patients in the two better risk groups, because it is possible that identifying a patient as high risk precludes further distinctions and, therefore, would invalidate the modeling assumptions. Analyses of parameters where it was felt chronic lymphocytic leukemia patients might be inherently different from lymphoma patients were repeated excluding the four patients, compared, for example, to the IPFP study, so that some risk factors might have been missed. Therefore, the two parameter analyses, including risk group and either HAMA or overall response as predictors of survival, were repeated using the IPFP risk groups. Because all of these supplementary analyses were consistent with the primary analysis, only the primary analysis is reported.

RESULTS

Thirty-one patients achieved at least a PR, including 11 with CRs. Eighty-seven % of the responders had at least a partial response in less than 9 weeks from start of treatment, with a median of 4 weeks. At the time of this analysis, all patients had progressed. Four patients (all in the low risk group) remained alive at 6.3, 4.2, 3.5, and 2.1 years after initiation of treatment.

**Treatment-related Predictors.** The values for key treatment-related parameters are summarized in Tables 1–4. Table 5 lists characteristics found significant on a univariate basis for one or more of the outcome measures. The only pretherapy parameter found significant at P = 0.1 was larger initial dose (mCi), which was predictive for CR. Among parameters measured following the first treatment dose, longer α and β half-times (h) for blood clearance predicted for CR and improved TTP. Increased cumulative activity in the blood (µCi/hr), increased bone marrow nontargeted radiation dose (rads), increased peak concentration (%ID/g), and extrapolated zero time concentration (%ID/g) for the tumor receiving the maximum dose (rads/mCi) were also predictive for CR.

Among the time-dependent variables, CR and overall response (CR or PR) predicted for prolonged time to progression and survival. Nineteen of 57 patients developed positive serum HAMA response (CR or PR) predicted for prolonged time to progression and survival. Nineteen of 57 patients developed positive serum HAMA levels, and higher HAMA score analyzed as a time-dependent variable predicted for improved survival. HAMA activity interrupted therapy in only seven patients (12%). With a limited number of patients, it is possible that predictive parameters might not achieve statistical significance. For this reason, the distributions of key parameters not found significant in univariate analyses were reviewed for patients who had a CR and for patients who had any response. Complete response occurred in patients with a total radiation dose to a tumor site from the first treatment dose as low as 31 rads and with a spleen volume as large as 968 ml. The median spleen volume for all patients with responses was 25 ml. These numbers suggest that responses can occur in patients often considered unlikely to benefit from RIT and that although there may be an association between these parameters and outcome, it is limited in degree.
tors have reported promising results from clinical trials using clearance $alpha$ and $beta$ half-times ($P_{ \text{non}}$ = $P_{ \text{as time-dependent variables remained predictive by this crite-}

versus $0.001$) and overall response

risk group in the model, none of the treatment-related parame-

Lym-1 (I2, 15, 16, 40, 41

DISCUSSION

point but without a response. This indicates that for each point in time, a patient with a response to high). The hazard ratio for overall response was 0.4. This category was 3.5. This indicates that, based on the model, at any

teraction with a high degree of significance

vival with a high degree of significance

they predicted improved UP and survival from the time of

missions were to be expected. Therefore, the presence of re-

parameters found in this study is of interest because of other

studies and were, therefore, not enrolled in the therapeutic

assessable tumor predicted to receive a higher absorbed dose of

biodistribution for patients receiving $^{111}$I antibody therapy (45,

ligation and facilitating host recognition of tumor (49, 50). Whether or not high HAMA titers ultimately prove to be pre-

ductive, it is clear from this study and others that concern about HAMA should not a priori preclude use of RIT.

Because blood represents the input function of radiolabeled

antibody to organs and tumor, parameters related to radionuclide

kinetics in the blood were considered potentially predictive. Longer $alpha$ and $beta$ blood clearance half-times were found to be predictive of CR and TTP in the univariate analysis. Even with risk included in the model, the $beta$ half-time was almost signifi-

cant for TTP at $P = 0.06$. Prediction based on longer half-times is consistent with the concept that rapid clearance from the blood prevents sufficient radiolabeled antibody from reaching the tumors, and these parameters are analogous to parameters describing the area under the curve for a chemotherapeutic agent.

The lack of association between outcome and spleen pa-

rameters found in this study is of interest because of other

reports indicating a relationship of spleen size to radionuclide biodistribution for patients receiving $^{111}$I antibody therapy (45,

46). In those studies, patients received therapy only if they were considered likely to have a favorable biodistribution, with every assessable tumor predicted to receive a higher absorbed dose of radiation than key normal organs. Patients with enlarged spleens generally did not have favorable biodistribution based on tracer studies and were, therefore, not enrolled in the therapeutic portion of the trial. A larger spleen is associated with a greater number of normal and malignant B-lymphocytes that can take

<table>
<thead>
<tr>
<th>Table 3 Pretherapy parameters evaluated for predictive potential</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Initial dose (mCi/m^2)</strong></td>
</tr>
<tr>
<td>57; 36 (14–100)$^a$</td>
</tr>
<tr>
<td>% Lym-1 reactivity (%)</td>
</tr>
<tr>
<td>49; 60 (10–97)</td>
</tr>
<tr>
<td>Spleen volume (ml)</td>
</tr>
<tr>
<td>47; 251 (0–2077)</td>
</tr>
<tr>
<td>Splenectomy 8</td>
</tr>
</tbody>
</table>

$^a$ Number of patients; median (range).
Radioimmunotherapy Predictive Parameters

Table 4 Time-dependent parameters evaluated for predictive potential

<table>
<thead>
<tr>
<th>Description</th>
<th>CR</th>
<th>Response</th>
<th>TTP</th>
<th>Survival</th>
<th>Better outcome</th>
</tr>
</thead>
<tbody>
<tr>
<td>Peak HAMA (µg/ml)</td>
<td>19: 88 (7–1802)(^a)</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>Higher dose</td>
</tr>
<tr>
<td>Cumulative dose (mCi/m²)</td>
<td>57: 131 (15–508)</td>
<td>CR</td>
<td>11: 9 (3–38)(^b)</td>
<td>Response</td>
<td>31: 4 (0.3–16)(^b)</td>
</tr>
</tbody>
</table>

\(^a\) Number of patients; median (range) for maximum per patient.
\(^b\) Number of patients; median (range) for time to event in weeks for those with event.

Table 5 \(P\) for treatment-related parameters significant in univariate analysis at \(P = 0.1\) for one or more of the outcome measures.

<table>
<thead>
<tr>
<th>Description</th>
<th>CR</th>
<th>Response</th>
<th>TTP</th>
<th>Survival</th>
<th>Better outcome</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial dose (mCi/m²)</td>
<td>0.03</td>
<td>NS(^a)</td>
<td>NS</td>
<td>NS</td>
<td>Higher dose</td>
</tr>
<tr>
<td>Peak concentration (%ID/g) for the high dose tumor</td>
<td>0.09</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>Higher peak</td>
</tr>
<tr>
<td>Extrapolated zero time concentration (%ID/g) for the high dose tumor</td>
<td>0.1</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>Higher concentration</td>
</tr>
<tr>
<td>Blood cumulative activity (µCi/hr)</td>
<td>0.1</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>Higher cumulative activity</td>
</tr>
<tr>
<td>Alpha half-time (h)</td>
<td>0.01</td>
<td>NS</td>
<td>0.02</td>
<td>NS</td>
<td>Longer time</td>
</tr>
<tr>
<td>Beta half-time (h)</td>
<td>0.06</td>
<td>NS</td>
<td>0.08</td>
<td>NS</td>
<td>Longer time</td>
</tr>
<tr>
<td>Nontargeted marrow dose (rads)</td>
<td>0.09</td>
<td>NS</td>
<td>NS</td>
<td>0.02</td>
<td>Higher dose</td>
</tr>
<tr>
<td>Peak HAMA (µg/ml)</td>
<td>NA</td>
<td>NA</td>
<td>0.04</td>
<td>0.005</td>
<td>Higher HAMA</td>
</tr>
<tr>
<td>CR</td>
<td>NA</td>
<td>NA</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td>Response</td>
</tr>
</tbody>
</table>

\(^a\) NS, not significant at \(P = 0.1\); NA, not applicable (time-dependent variables were not considered as possible predictors for CR or Response).

up antibody, resulting in less antibody available for tumor binding outside the spleen. In our studies, patients with enlarged spleens were not excluded, and many patients had spleen volumes that deviated greatly from normal. Therefore, the data were useful for determining whether spleen size and/or dose to spleen were predictive of outcome from Lym-1 RIT. Analysis showed no relation between spleen parameters and outcome, even in univariate models. However, the antibodies studied target distinctly different antigens: CD20 and CD37 versus HLA-Dr (Lym-1)(18, 45). An enlarged spleen may present an overwhelming antigen sink for some antibodies, requiring adjustments such as pretherapy loading doses or splenectomy for successful therapy. This will need to be evaluated on an individual antibody basis.

Bone marrow dosimetric parameters were considered because of their potential for limiting RIT doses. The only parameter significant in univariate analysis was marrow radiation contributed by blood and body sources, which predicted higher likelihood of CR with higher dose (rads). This result may reflect the higher initial injected doses, which tended to be given to the patients without extensive marrow malignancy. However, none of the bone marrow variables proved predictive once patient risk group was taken into account.

Because radiotherapeutic response results from energy deposition in the tumor tissue, parameters relevant to radionuclide uptake, clearance, and radiation dose for tumor could be useful predictors. Past studies have shown that radiation dose to tumor can vary considerably and is not easily predicted from the amount of radioconjugated-antibody or radionuclide given (41). In a study reported by Kaminski et al. (44), pretreatment antibody loading dose was selected based on the ratio of radiation dose to tumor and total body observed in tracer studies. As noted above, Press et al. (45, 46) selected patients for RIT by requiring that tumor dose be greater than that to normal organs. For patients reported here, dose adjustments were not made based on tumor uptake, allowing evaluation of tumor dose as a predictor. In our analysis, some tumor dose-related parameters were predictive in univariate analyses (Table 5), but none improved predictive ability beyond information already provided by risk group category for any of the outcomes studied. The lack of outcome prediction based on tumor dose may have multiple causes. It is generally recognized that radiation dose is not homogeneously distributed throughout the tumor and differs from tumor to tumor. Additionally, radiation dosimetry for radionuclide treatment has not reached the level of accuracy of radiation dosimetry for external beam radiotherapy. Finally, tumor response and survival are the result of complex interactions of variables including the inherent biology and radiosensitivity (radiobiology) of the tumor and the clinical and immunological status of the patient.

In summary, our intent is to present a model for how to approach evaluating RIT therapies, rather than a model for RIT therapy. Even with the limitations presented by relatively small sample size and the many sources of variability, there was still
an indication of positive long-term outcomes based on response to therapy. This reinforces the belief that even heavily pretreated patients can derive clinical benefit from treatment with Lym-1 RIT. It also demonstrates that concern about HAMA need not a priori preclude consideration of such therapy. Review of the extensive information available to us concerning pharmacokinetics and dosimetry on these patients found them to be of limited predictive value once basic patient characteristics were considered. It will be important for others to complete similar reviews of their data. In the interim, although the use of radiation dosimetry to project safe dose levels based on potential organ toxicity may have a role in treatment planning, we should be cautious in using tumor dosimetry to determine patient eligibility or to select administered dose of radionuclide.

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REFERENCES


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