Prognostic models in patients with non-small-cell lung cancer using artificial neural networks in comparison with logistic regression

Taizo Hanai,[1, 5, 6] Yasushi Yatabe,[2, 6] Yusuke Nakayama,[1] Takashi Takahashi,[4] Hiroyuki Honda,[1] Tetsuya Mitsudomi[3] and Takeshi Kobayashi[1]

[1]Department of Biotechnology, Graduate School of Engineering, Nagoya University, Furo-cho, Chikusa-ku, Nagoya 469-8603, [2]Department of Pathology and Molecular Diagnostics, [3]Department of Thoracic Surgery, Aichi Cancer Center Hospital and [4]Division of Molecular Oncology, Aichi Cancer Center Research Institute, 1-1 Kanokoden, Chikusa-ku, Nagoya 464-8681

(Received January 16, 2003/Revised March 17, 2003/Accepted March 17, 2003)

It is difficult to precisely predict the outcome of each individual patient with non-small-cell lung cancer (NSCLC) by using conventional statistical methods and ordinary clinico-pathological variables. We applied artificial neural networks (ANN) for this purpose. We constructed a prognostic model for 125 NSCLC patients with 17 potential input variables, including 12 clinico-pathological variables (age, sex, smoking index, tumor size, p factor, pT, pN, stage, histology) and 5 immunohistochemical variables (p27 percentage, p27 intensity, p53, cyclin D1, retinoblastoma (RB)), by using the parameter-increasing method (PIM). Using the resultant ANN model, prediction was possible in 104 of 125 patients (83%, judgment ratio (*JR*)) and accuracy for prediction of survival at 5 years was 87%. On the other hand, *JR* and survival prediction accuracy in the logistic regression (LR) model were 37% and 78%, respectively. In addition, ANN outperformed LR for prediction of survival at 1 or 3 years. In these cases, PIM selected p27 intensity and cyclin D1 for the 3-year survival model and p53 for the 1-year survival model in addition to clinicopathological variables. Finally, even in an independent validation data set of 48 patients, who underwent surgery 10 years later, the present ANN model could predict outcome of patients at 5 years with the *JR* and accuracy of 81% and 77%, respectively. This study demonstrates that ANN is a potentially more useful tool than conventional statistical methods for predicting survival of patients with NSCLC and that inclusion of relevant molecular markers as input variables enhances its predictive ability. (Cancer Sci 2003; 94: 473–477)

Lung cancer is the leading cause of cancer death in Japan[1)] as well as in Western countries.[2)] Lung cancer is divided into two major morphological types; small-cell lung cancers (SCLCs) and non-small-cell lung cancers (NSCLCs). Although about 30% of NSCLC patients are candidates for potentially curative resection, their long-term survival rate remains unsatisfactory.[3–6)] Using existing prognostic tools, such as the commonly used TNM classification,[7, 8)] however, it is often difficult to accurately predict the outcome of each individual NSCLC patient.[3, 9)] Tumors that show similar morphology under the microscope may have different sets of genetic alterations and, thus, may exhibit different biological behavior in patients.

We have been evaluating various genetic or epigenetic changes of cancer-related genes in a search for clinically relevant prognosticators. In our previous studies, alterations of cancer-related genes, including cyclin D1,[10)] retinoblastoma (RB),[10)] p53[11)] and p27,[12)] have been shown to be of prognostic importance. However, no single variable was sufficiently predictive to precisely foresee a patient's outcome. To overcome this problem, statistical methods of regression have been developed to analyze multiple variables simultaneously. For studies with a binary endpoint, logistic regression (LR) has been used, while for studies with time-to-event data, Cox proportional hazards regression has been utilized as a standard method. However, these methods still have limitations and are not ideal tools for prediction of individual patient's outcome.

Artificial neural networks (ANN) have been developed as an alternative statistical technique in the last 40 years[13)] and have been applied in the biochemical and medical fields.[14–17)] ANN is a computational methodology that performs multifactorial analyses. In analogy with networks of brain neurons, ANN contains layers of simple computing nodes that operate as nonlinear summing devices.[13)] These nodes are interconnected by weighted connection lines, enabling tasks such as predicting outcome values, classifying an object, approximating a function and recognizing a pattern in multifactorial data (Fig. 1).[13)] In addition, an ANN model has an output for each set of input variables. Therefore, using ANN, it is possible to predict outcome on an individualized basis.

In the present study, we attempted to predict the prognosis of individual patients using ANN, aiming at achieving better clinical management of the individuals based on the expected risk. The cohort used is a non-biased consecutive series, and the variables of the cohort included not only conventional clinicopathological factors but also biological markers; i.e., p53, p27, cyclin D1 and RB. The accuracy and efficacy of ANN were compared with those of LR.

Methods

Patients. A series of 125 consecutive patients with NSCLC (32–82 years old (median, 63.3), 88 females and 37 males who underwent potentially curative resection at the Department of Thoracic Surgery, Aichi Cancer Center, Nagoya, from 1986 through 1988) were used for the present study. There were 62 adenocarcinomas, 50 squamous cell carcinomas, five adenosquamous carcinomas and eight large-cell carcinomas. The variables listed in Table 1 were used for the analysis in the present study, which included age, sex, smoking index (number of cigarettes×year), tumor size, p factor (pleural involvement with 4 categories: i.e., p0, no invasion; p1, invasion that does not reach the surface; p2, invasion beyond visceral pleura; p3, invasion to chest wall, diaphragm, mediastinal pleura, parietal pericardium), pT, pN, pathological stage according to the 4th edition of the TNM classification, and histologic type according to the new World Health Organization (WHO) classification (1999). In addition, 48 patients who underwent surgery in 1996

[5]To whom correspondence should be addressed at Laboratory for Bioinformatics, Graduate School of Systems Life Sciences, Kyushu University, 6-10-1 Hakozaki, Higashi-ku, Fukuoka 812-8581, Japan.
E-mail: taizo@brs.kyushu-u.ac.jp
[6]These authors have an equal contribution to this article.

were used for validation of the models created with the 1986–1988 cohort (test cases). This group comprised 31 men and 17 women with ages ranging 43 to 76 years (median 64) and included 32 adenocarcinomas, eight squamous cell carcinomas, five adenosquamous carcinomas and three large-cell carcinomas. All cases were coded prior to the initiation of this study in order to keep patients' information anonymous.

Immunohistochemical analysis of biological markers. In the present study, we incorporated several biological variables to improve the efficacy of our models (Table 1). To this end, we used data sets comprising the expression status of p27, p53, cyclin D1 and RB, which have been reported as prognostic factors in our previous studies.[10–12] The procedures of preparation and analysis of tissue samples have also been described in detail.[10–12] Briefly, the standard avidin-biotin-peroxidase complex method was used for immunohistochemical examination of paraffin sections using monoclonal antibodies against $p27^{KIP1}$ (Transduction Laboratory, Lexington, KY), p53 (DO-7, DAKO, Copenhagen, Denmark), cyclin D1 (NCL-cyclin D1, Novocastra Laboratories, Newcastle, UK), and RB (3H9, Medical and Biological Laboratories, Nagoya). In the case of evaluation for p27 expression, both percentage and intensity were included (according to the criteria as reported).

Data preprocessing. In order to use these parameters as input variables for ANN, we standardized all these data into numerical data ranging from 0.05 to 0.95. In this study, three independent models to estimate patient survivals at 1, 3, and 5 years after surgery were constructed. For each estimation model, the output values of survival and death were set to 0.05 and 0.95, respectively. Multicolinearity, which may interfere with the construction of a proper model, would be expected to be present among some of the variables. For example, pT is determined by p factor and tumor size, and pathologic stage is based on pN and pT in the present series. However, calculation of the correlation coefficients for every combination of all variables in Table 1 revealed no disturbingly high correlation, and every correlation coefficient was found to be less than 0.85. Therefore, all 17 variables were used as potential inputs for the current analysis.

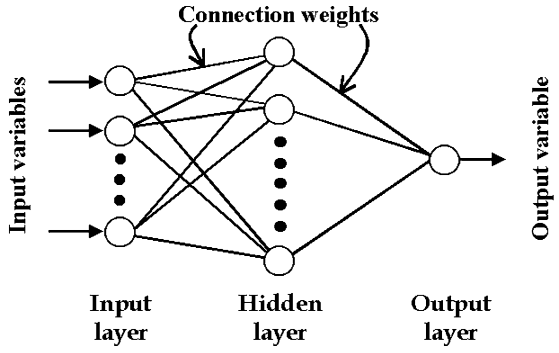

Fig. 1. Artificial neural network.

ANN. In the present study, a three-layered ANN composed of input, hidden and output layers, was designed as shown in Fig. 1. The output layer had only one unit, which represents the survival status of the patients. Initially, connection weights were randomly assigned values between 0 and 1, and subsequently they were automatically altered by the back propagation method[18] to identify the optimal relationships between input and output. This process is called "learning."

In cases where the number of connection weight parameters is much larger than that of the learning data set, the resultant model may have less generalizability and flexibility.[19] Therefore, in this case it was necessary to decrease the number of input and hidden units so that the number of connection weight parameters was decreased for optimization of the model. To this end, we used the parameter-increasing method (PIM).[20] Briefly, the initial step of PIM was to choose the input variable that was most crucial for accurate prediction. In the next step, the second most crucial variable was selected. By repeating this operation, the best combination of input units was selected in the prediction model. Similarly, the number of units in the hidden layer was decreased one by one from 10, and this procedure was continued until the accuracy of the model had dropped sharply. Practically, the numbers of input units and hidden layer units were different every time because of the random assignment of initial weight connections. Therefore, five optimizing procedures were independently done for both input and hidden layer units, and the input/hidden layer neurons that were most frequently selected were considered as the optimized input/hidden layer neurons.

Subsequently, in order to examine the flexibility of an ANN, cross-validation was performed as follows. At first, the data sets were divided into 5 groups. Then, groups 2, 3, 4 and 5 were used for learning data, and the remaining group 1 was used for evaluation of the trained ANN model. In the next step, groups 1, 3, 4 and 5 were used for learning data, and the remaining group 2 was used for evaluation. All groups of data

Table 1. Potential input variables for prognostic models

Input variables	Data type	Value
Age	Continuous	36–82
Sex	Categorical	1: male, 2: female
Smoking index	Continuous	0–2350 cigarettes×year
Tumor size	Continuous	36–110 mm
p factor	Categorical	1: p0, 2: p1, 3: p2, 4: p3
T factor	Categorical	1: T1, 2: T2, 3: T3, 4: T4
N factor	Categorical	1: N0, 2: N1, 3: N2
Pathological stage	Categorical	1: I, 2: II, 3: IIIa, 4: IIIb
Histological types adeno	Categorical	1: yes, 2: no
Histological types squamous	Categorical	1: yes, 2: no
Histological types adenosquamous	Categorical	1: yes, 2: no
Histological types large	Categorical	1: yes, 2: no
p27 percentage	Categorical	1: <5%, 2: 5–30%, 3: 31–60%, 4: 61%<
p27 intensity	Categorical	1: negative, 2: decreased, 3: normal, 4: increased
p53	Categorical	1: negative, 2: positive
Cyclin D1	Categorical	1: negative, 2: positive
RB	Categorical	1: negative, 2: positive

were used as evaluation data in the same way, and the average of all procedures was considered as the estimation ability of ANN. In addition, a completely independent data set was also used to validate the ANN model constructed as described above.

We evaluated the efficacy of our models in terms of two values; i.e., judgment ratio (*JR*) and accuracy. If the patient of interest was dead at a given time point, and the output was larger than the high threshold, the prediction was considered true positive (TP). Conversely, if the output was smaller than the low threshold, the prediction was considered false negative (FN). False positive (FP) and true negative (TN) predictions can be similarly determined. With these numbers, *JR* and accuracy are given in the following equations. The *JR* indicates the proportion of patients on which judgment can be achieved, while accuracy indicates the fraction of *JR* on which the correct judgment was achieved.

$$JR = \frac{N_{TP}+N_{TN}+N_{FP}+N_{FN}}{N_{all}} \times 100 \qquad (1)$$

$$Accuracy = \frac{N_{TP}+N_{TN}}{N_{TP}+N_{TN}+N_{FP}+N_{FN}} \times 100 \qquad (2)$$

where N_{TP}, N_{TN}, N_{FP}, N_{FN} and N_{all} are the number of TP, TN, FP, FN and all collected data, respectively.

LR modeling. As a control of the modeling method, a conventional statistical prediction model with logistic regression was also constructed. SPSS for Windows (SPSS Regression Models 10.0, SPSS, Inc., Chicago, IL) was used for LR modeling. The input variables for the LR model were optimized by PIM based on the likelihood ratio.

Results

Selected input variables for 1-, 3- and 5-year survival models. Table 2 shows the selected variables for 1-, 3- and 5-year survival models using ANN and LR in the order selected by PIM. Variables selected in the earlier steps are more essential for prognosis prediction. It was noted that several biological variables, especially p53 and p27, were repeatedly selected in the models.

Although a disturbingly high correlation was not observed, we also confirmed that multicolinearity among the input variables does not affect the results by eliminating one of the parameters, such as pathological stage, size, p factor, or p27 expression, from the constructed ANN models. In every attempt, the *JR* and accuracy of the models were lower than those of the models using all input variables (data not shown). The number of the units in hidden layers for the 1-, 3- and 5-year survival estimation models was optimized in a similar manner, resulting in 7, 9 and 10 units, respectively.

Table 2. Selected input variables by the parameter-increasing method

Order of selection	1-year survival		3-year survival		5-year survival	
	ANN	LR	ANN	LR	ANN	LR
1	Stage	Stage	pN	pN	Stage	p
2	SI	SI	p27 intens.	p	Size	pN
3	Age	—	Age	Sex	SI	p27 intens.
4	p	—	pT	SI	p	—
5	p53	—	SI	p27 intens.	p53	—
6	Size	—	Size		Age	—
7	pT	—	CCND1		—	—

Stage, pathological stage; p27 intens., p27 intensity; SI, smoking index; CCND1, cyclin D1.

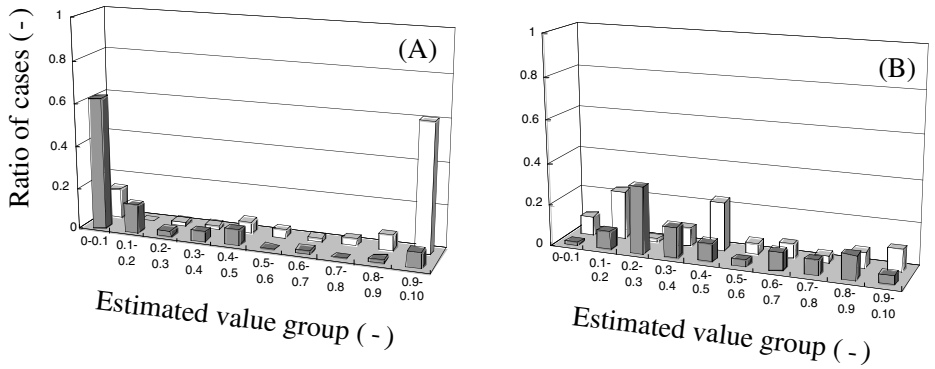

Fig. 2. Results of 5-year survival estimation model using ANN (A) and LR (B). Shaded boxes, "alive" patients; open boxes, "dead" patients.

Table 3. Comparison of predictive models for 1-, 3- and 5-year survival using ANN and LR

	1-year survival		3-year survival		5-year survival	
	ANN	LR	ANN	LR	ANN	LR
JR (%)	77.6	73.6	72.8	50.4	83.2	36.8
No. of patients	97/125	92/125	91/125	63/125	104/125	46/125
Accuracy (%)	93.8	95.7	91.3	87.3	86.8	78.3
No. of patients	91/97	88/92	83/91	55/63	90/104	36/46

Threshold determination. Fig. 2 shows the result of the estimation of 5-year survival using ANN (A) or LR (B). The ANN model discriminated the survival status well, whereas the LR model did not. This clear distinction was also evident in the models estimating 1- and 3-year survival. Since the outputs of the models are given with contiguous values, we configured thresholds for the predictive judgment such that the accuracy of the model was maximized. An increasing accuracy is, to some extent, in conflict with a high *JR*; therefore, we gave accuracy a higher priority than *JR*. By fluctuating the threshold values, the maximal accuracy (86.8%) was achieved with values of 0.2 and 0.8 for the low and high thresholds, respectively. These thresholds were also suitable for the estimation models of 1- and 3-year survival and were therefore used for all estimation models using ANN and LR.

***JR* and accuracy of the estimation models.** Table 3 shows the *JR* and accuracy of the 1-, 3- and 5-year survival estimation models using ANN and LR. Both *JR* and accuracy were superior in the models using ANN to those using LR in most of the cases. For the ANN estimation model of 5-year survival, the *JR* was 83.2% and the accuracy was 86.8%. In contrast, with a 5-year survival model using LR, *JR* was 36.8% and accuracy was 78.3%.

To assess the importance of biological variables, we also constructed models eliminating those variables. *JR* and accuracy of these models were lower than those of the models using all input variables (Tables 4 and 5), suggesting that these biological markers are important for optimal performance of the models. In addition, the impact of p53 and p27 on the prediction was examined by eliminating either of these two biological variables in a similar manner. This also resulted in lower *JR* and accuracy than if all input variables were used in the models. Taken together, these results suggest that biological variables (i.e., p53 and p27) were as important as other clinicopathological variables (Tables 4 and 5).

Validation with an additional independent data set (Table 6). In order to examine the generality of the constructed ANN model, additional data of an independent cohort of 48 patients, who underwent surgery in 1996, were used for validation. *JR* was 77.8% and 81.3% for 3- and 5-year survival, respectively. The *JR* differences between the cohorts with test data (1998 cohort) and learning data (1986–88 cohort) were +5.0% and −1.9%, respectively, for 3-year and 5-year survival. Accuracies with the validation data set (1998 cohort) were 85.7% for 3-year survival and 76.9 % for 5-year survival, which were only 5.6% and 9.9% lower than those with the learning data set (1986–88 cohort).

Table 4. Selected input variables by the parameter-increasing method

Order of selection	5-year survival	5-year survival without variables for p53 and p27	5-year survival without biological variables
1	Stage	Stage	Stage
2	Size	Size	Size
3	SI	SI	SI
4	p	p	p
5	p53	CCND1	Age
6	Age	Age	Sex
7	—	—	—

Stage, pathological stage; SI, smoking index; CCND1, cyclin D1.

Table 5. Comparison of predictive models for 5-year survival

	5-year survival	5-year survival without variables for p53 and p27	5-year survival without biological variables
JR (%)	83.2	59.2	55.2
No. of patients	104/125	74/125	69/125
Accuracy (%)	86.8	83.8	81.2
No. of patients	90/104	62/74	56/69

Table 6. Estimation of 3- and 5-year survival for unlearned data set from 1996 cohort

	3-year survival		5-year survival	
	Learning data	Validation data	Learning data	Validation data
JR (%)	72.8	77.8	83.2	81.3
No. of patients	91/125	14/18	104/125	39/48
Accuracy (%)	91.3	85.7	86.8	76.9
No. of patients	83/91	12/14	90/104	30/39

Learning data were from 1986–1988 cohort and validation data were from 1996 cohort.

Discussion

We have created models for prediction of outcome of each individual NSCLC patient using ANN with immunohistochemical data of biological markers, as well as with conventional clinicopathological variables. *JR* and accuracy of 1-, 3- and 5-year survival estimation models using ANN were superior to those of LR in most cases, except for the accuracy of prediction of 1-year survival by LR, for which only two variables of pathological stage and smoking index were found to be useful (see Table 2). This suggests that the relationship between the input variables and survival status may be correlated rather simply for the prediction of short-term outcome, and that 3- or 5-year survival status may be configured by more complex factors.

We found several biological markers, especially p53 and p27, that were selected as important input variables. Addition of these biological markers to conventional clinico-pathological variables potentiated the predictive ability of the model.[8] Although it is well recognized that the extent of invasion and metastasis is associated with a patient's outcome, additional parameters have been sought to improve accuracy of prediction. However, no single marker known to date has definitive prognostic significance in NSCLC patients. Even for p53, which is one of the molecules that play a central role in cancer development, the prognostic significance is controversial. Although many reports have shown a prognostic significance of p53 status, which was confirmed by meta-analysis,[21] the significance was marginal or at best rather modest in most reports. In the present study, biological markers, including p53, p27 and cyclin D1, were often selected by PIM as more significant input variables for prognostic prediction by ANN, suggesting that the ANN model could resolve their entangled nonlinear relationships. Jefferson *et al*. created predictive models for the outcome of 620 NSCLC patients at 12, 18 and 24 months using a genetic algorithm neural network (GANN, a type [kind] of ANN).[22] The accuracy of 1-year survival estimation using these authors' model was 10% lower than that using our model. This difference may be in part attributable to the fact that these investigators only used clinico-pathological variables (stage, sex, T, N, histologic type and differentiation).

Bellotti *et al*. also constructed an estimation model for prognosis of NSCLC patients using a three-layered ANN.[23] They used 12 variables, including four biological parameters (i.e., S phase fraction, proliferating nuclear antigen, MIB-1 staining

and p53), for the analysis of 67 patients' data.[23] In contrast to our attempt to use PIM to reduce the number of input variables and hidden layers, these investigators used all 12 input variables, 12 units in hidden layers, 1 output variable and 156 connection parameters, which may well be too many for the 67 learning data. Unfortunately, they used all 67 data as learning data and did not evaluate the validity of the model by using an independent data set, which we believe is of crucial importance for the evaluation of generality of the constructed model. In our study, the accuracy observed with the additional independent data set for validation was 9.9% lower than that for the training data set. Taking into account the fact that the validation data set was obtained with patients who underwent surgery about 10 years later, there may have been small but nevertheless important differences in patient selection and management. In addition, there were considerable changes in the distribution of histologies of lung cancers during the decade. Therefore, a decrease in the prediction accuracy by less than 10% may be considered to be reasonably small.

The superiority of ANN to regression models, however, should be interpreted with caution. In a collective review of 28 studies comparing ANN with logistic or Cox regression models,[24] ANN outperformed regression models in 10 of the 28 studies, but was outperformed by regression in 4 studies, and the 2 methods had similar performance in the remaining 14 studies. It is noteworthy that in the 8 largest studies (sample size >5000), regression and ANN tied in 7 cases.

In conclusion, survival estimation models at 1, 3 and 5 years after potentially curative surgery were created using ANN in patients with NSCLC. After optimization of ANN, *JR* and accuracy of ANN models were generally higher than those of LR models. The selected input variables of ANN by PIM contained not only conventional clinico-pathological variables, but also several biological markers. Recent progress in biotechnology now enables us to perform multiple genetic or immunohistological analyses with a short turn-around. cDNA microarray[25, 26] and tissue microarray technologies[27] are representatives of these techniques. If they were combined with ANN, this would give the best opportunity to create a highly accurate prognostic model. In this connection, Khan *et al*. have recently shown the potential of combinatorial use of expression profiling and ANN for classification of small round blue-cell tumors into four specific diagnostic categories, which are sometimes difficult to distinguish by light microscopy.[28] In a future study, a larger number of patients should be collected to increase the reliability of the prognostic model. It should become possible to correctly identify patients who are at greater risk of poor clinical outcome using ANN. Such patients are candidates for investigational therapeutic approaches. In clinical trials, this kind of model may also be useful to stratify patients into different prognostic groups.

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