Preoperative Prediction of Lymphovascular Invasion of Colorectal Cancer by Artificial Neural Network

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Abstract

Background: Lymphovascular invasion (LVI) is considered to be important for metastasis of colorectal cancer (CRC). However, there is still no effective method to predict LVI before operation. Our research aimed to construct an artificial neural network (ANN) for the pre-operative prediction of LVI.

Methods: We obtained blood indexes and conditions of LVI (confirmed by pathological examination) of 288 cases of CRC patients from a tertiary hospital in China. ANN and logistic regression model were constructed based on randomly chosen 185 cases CRC patients (training group). The remaining 103 cases of CRC patients received tests of ANN and logistic model (validation group). Receiver operating characteristics curve (ROC) and decision curve analysis (DCA) was performed to assess the accuracy of constructed model respectively.

Results: In the training group, the area under curve (AUC) of ANN was higher than that of logistic model (0.832 vs 0.692). The ANN correctly predicted 92% cases of LVI, whereas logistic model only predicted 56% cases. Similar results were also tested in the validation model.

Conclusions: Our constructed ANN showed higher accuracy compared with a conventional linear model. The ANN based on blood indexes may provide value for pre-operative prediction of LVI.

Background

CRC is one of the most common digestive tract tumor and nearly one million people suffer from CRC every year with a mortality rate of 33%(1). Recurrence and metastasis are important causes of death of CRC. LVI was reported to be associated with poor differentiation, unfavorable survival as well as lymph node metastasis(2, 3). LVI is commonly detected by pathological examination after operation. However, pathological examination has hysteresis, which can not predict the metastasis and recurrence of patients in early stage(4). It is urgent to develop a new method to predict LVI before operation.

Traditional clinical prediction model is mainly based on linear regression model. Although it works effectively for small data size with simple relationships, it is difficult to construct lineal regression model for nonlinear data and express highly complex data(5). ANN is a good solution to the shortcomings of traditional linear regression with the characteristics of self-adaptive, self-organizing and real-time learning. It is a nonlinear and adaptive information processing system composed of a large number of processing units(6). At present, ANN has been more and more used in the field of biology and medicine(7, 8). Our research adopted common blood indexes in preoperative examination or physical examination to construct ANN for pre-operative prediction of LVI, thus provided value for surveillance of CRC in early stage.

Material And Methods

Patients

This study included 408 cases of CRC patients from Shanghai Pudong Hospital. These patients received CRC operation from April 2017 to April 2020. Patients with the following criteria were excluded from participation: receive anticancer treatment outside Shanghai Pudong Hospital; loss of lymphovascular invasion information, necessary basic characteristic information, necessary serological parameter or follow up; receive any other treatment. 288 cases of qualified CRC patients were finally recruited in the study (Fig. 1). Clinical data (sex, age and body Mass Index), pre-operative blood indexes (alanine aminotransferase, carcinoembryonic antigen, Carbohydrate antigen-199, etc.) and postoperative pathological data (LVI and histologic grade) were obtained from medical records. LVI was determined by D2-40 immunohistochemistry (IHC) combined with hematoxylin-eosin (HE) staining.

Construction of logistic regression model

In training group, the association of variables (clinical data and pre-operative blood indexes) and LVI were performed with Spearman correlation analysis, p-value less than 0.05 was adopted as the screening criteria. Screened variables were further performed with multivariate logistic regression analysis.

Construction of artificial neural network

Variables found to be significantly related to LVI in multivariate analysis were adopted to construct ANN. we built a three-layer feedforward ANN with four input nodes, five neurons in the hidden layer, and two output neurons. The learning rule used here was the back propagation of the error. The cut-offs of logistic regression and ANN outputs with the best relationship between sensitivity and specificity were adopted for classification.

Assessment of logistic regression model and artificial neural network

In training group and validation group, both logistic regression model and ANN were tested with ROC curve (Hanley-McNeil method) and represented in terms of negative predictive value (NPV), positive predictive values (PPV), and likelihood ratios (LR). Furthermore, DCA was performed to calculate the net benefit of LVI conditions by the logistic regression model and ANN respectively.

Statistical analysis

All analyses were performed using SPSS 23.0 and R 3.5.3. All statistical tests were two-sided, and a P value < 0.05 was considered statistically significant. Continuous variables that conformed to the normal distribution were compared with the use of an independent t test for comparison between groups, while continuous variables with skewed distribution were compared with the Mann-Whitney U test. Categorical data were tested using the Chi-square test.

Results

Baseline of Patients' characteristics and pre-operative blood indexes

The characteristics of all patients in both training group and validation group were shown in Table 1. There was no difference in all clinical characteristics and pre-operative blood indexes between training group and validation group. In training group, there were 70 cases of LVI, accounted for 37.8% of all patients. In validation group, there were 42 cases of LVI, accounted for 40.8% of all patients. There was no difference between LVI condition in two groups (p-value: 0.705).

Table 1
Baseline characteristics of the study population in the training and validation group

Variables	Training (n = 185)	Validation (n = 103)	p Value
Sex (Male)	112 (63%)	58 (57%)	0.274
Age (≤ 50 years)	20 (10.8%)	8 (7.8%)	0.534
Median BMI (IQR)	23.7 (22.5−25.1)	23.7 (21.6−26.0)	0.487
ALT (≤ 50 U/L)	150 (81.1%)	79 (76.7%)	0.446
AST (≤ 40 U/L)	162 (87.6%)	86 (83.5%)	0.376
TB (≤ 19 μmol/L)	144 (77.8%)	75 (72.8%)	0.388
CB (≤ 6.8 μmol/L)	135 (73%)	67 (65%)	0.180
ALB (≤ 40 g/L)	121 (65.4%)	71 (68.9%)	0.603
PLT (≤ 100 × 10^9/L)	8 (4.3%)	3 (2.9%)	0.752
Median CEA (ng/ml) (IQR)	2.63 (1.68−5.77)	2.7 (1.46−5.95)	0.584
Median AFP (ng/ml) (IQR)	2.6 (1.6−3.5)	1.8 (1.3−2.6)	0.230
Median CA199 (u/ml) (IQR)	16.36 (10.27−29.6)	15.13 (10.24−26.43)	0.331
Median CA242 (u/ml) (IQR)	6.98 (4.27−10.83)	6.33 (4.19−7.19)	0.221
Median CA125 (u/ml) (IQR)	9.7 (6.8−16.1)	8.9 (6.4−15.1)	0.192
Histologic grade (moderately/poorly differentiated)	123 (66.5%)	76 (73.8%)	0.232
lymphovascular invasion (present)	70 (37.8%)	42 (40.8%)	0.705
IQR: interquartile range			

Construction of logistic regression model and artificial neural network

First of all, we investigated the relationship of clinical characteristics and blood indexes with LVI via Spearman correlation analysis. We demonstrated glutamyl transpeptidase (CGT), neuron specific enolase (NSE), CA125, CA199, carcinoembryonic antigen (CEA), C reactive protein (CRP), fibrinogen and preoperative largest diameter of tumor were associated with LVI ($p < 0.05$). These variables were further performed with multivariate logistic regression model. Only CA125, CA199 and fibrinogen were finally adopted as independent prediction factors for LVI (Table 2). Meanwhile, CA125, CA199 and fibrinogen were adopted to construct ANN to improve performance of the logistic regression model.

Table 2
Preoperative clinical characteristics and blood indexes of the training group related to lymphovascular invasion.

Variables	Spearman correlation analysis		Multivariate analysis	
	r value	p value	b coefficient	p value
Age (years)	-0.091	0.175		
Sex (male, female)	0.090	0.175		
Hepatic virus infection (absent, present)	-0.003	0.965		
ALT (U/L)	0.040	0.557		
AST (U/L)	0.025	0.711		
ALP (U/L)	0.078	0.249		
GGT (U/L)	0.176	0.046		0.533
WBC	0.094	0.163		
RBC	0.013	0.852		
PLT	0.048	0.480		
Lipase	-0.775	0.220		
TB (umol/L)	-0.029	0.661		
CB (umol/L)	-0.003	0.966		
ALB (g/L)	-0.012	0.862		
Prealbumin	-0.023	0.739		
GLOB (g/L)	-0.026	0.699		
CA242	0.215	0.166		
NSE	0.732	0.039		0.103
CA50	-0.024	0.905		
AFP	-0.106	0.128		
CA125	0.172	0.013	0.019	0.028

AFP, alpha fetoprotein; ALB, albumin; ALP, alkaline phosphatase; ALT, alanine aminotransferase; Apo A1, apolipoprotein A1; Apo B, apolipoprotein B; AST, aspartate aminotransferase; CA242, carbohydrate antigen 242; CB, conjugated bilirubin; CEA, carcinoembryonic antigen; CRP, C reactive protein; D-D, d-dimer; GGT, glutamyl transpeptidase; GLOB, globulin; PLT, blood platelet; PT, prothrombin time; RBC, red blood cell; TB, serum total bilirubin; TBA, total bile acid; WBC, white blood cell; neuron specific enolase, NSE.

	Spearman correlation analysis		Multivariate analysis	
CA199	0.181	0.008	0.006	0.031
CEA	0.201	0.004		0.650
CRP	0.149	0.046		0.703
Fibrinogen	0.142	0.036	0.168	0.042
Antitrypsin	0.161	0.117		
Serum phosphorus	0.179	0.175		
Serum magnesium	-0.194	0.152		
Serum sodium	0.059	0.382		
Preoperative largest diameter of tumor	0.166	0.019		0.648

AFP, alpha fetoprotein; ALB, albumin; ALP, alkaline phosphatase; ALT, alanine aminotransferase; Apo A1, apolipoprotein A1; Apo B, apolipoprotein B; AST, aspartate aminotransferase; CA242, carbohydrate antigen 242; CB, conjugated bilirubin; CEA, carcinoembryonic antigen; CRP, C reactive protein; D-D, d-dimer; GGT, glutamyl transpeptidase; GLOB, globulin; PLT, blood platelet; PT, prothrombin time; RBC, red blood cell; TB, serum total bilirubin; TBA, total bile acid; WBC, white blood cell; neuron specific enolase, NSE.

Comparison of logistic regression model and artificial neural network

We further assess the performance of logistic regression model and ANN in both training group and validation group via ROC curve (Fig. 2A, Table 3). We confirmed best cutoff value by learning Youden's index (Sensitivity + specificity-1). The sensitivity of ANN is 92% in training group and 75% in validation group, significantly higher than those of logistic regression model (56% in training group and 45% in validation group). However, high sensitivity of ANN seems to sacrifice specificity. The specificity of ANN is 74% in training group and 67% in validation group, lower than those of logistic regression model (83% in training group and 85% in validation group). Similarly, ANN showed higher PPV and positive LR but lower NPV and negative LR compared with logistic regression model in both training group and validation group. The AUC of ANN was 0.839 in training group and 0.76 in validation group, significantly higher than those of logistic regression model (0.692 in training group and 0.682 in validation group). ANN significantly improved AUC compared with logistic regression model (Training group: $P < 0.001$, Validation group: $P < 0.05$). In DCA, we also determined ANN provided a better net benefit to predict LVD compared with logistic regression model (Fig. 2B).

Table 3
ROC curve analysis of Logistic regression model and artificial neural network

	Logistic regression		Artificial neural network	
Group	Training	Validation	Training	Validation
AUROC (95% CI)	0.692	0.682	0.839	0.760
	(0.606−0.778)	(0.595−0.770)	(0.780−0.899)	(0.685−0.835)
Cut-off values	0.31	0.31	0.66	0.41
Sensitivity/specificity (%)	56/83	45/85	92/74	75/67
Correctly classified (%)	75	72	78	75
PPV/NPV (%)	67/82	51/82	54/97	43/89
Positive/negative LR	3.33/0.53	3.09/0.64	3.48/0.11	2.25/0.38
Comparison of AUROC:				
Logistic vs. ANN (Training)	$p < 0.001$			
Logistic vs. ANN (Validation)	$p < 0.05$			

Note: ROC, receiver operating characteristic; AUROC, area under the ROC curve; LR, likelihood ratio; NPV, negative predictive value; PPV, positive predictive value.

Discussion

Recent studies also showed that LVI can be regulated and promoted by lymphangiogenic of growth factors. Growth factors include glycoprotein, vascular endothelial growth factor-C (VEGF-C) and VEGF-D, and chemokines may also play roles by attracting tumor cells to lymphatic vessels(9). Lymphangiogenic factors can be released not only by tumor cells, but also by tumor related active matrix and immune cells(10). Further understanding of these mechanisms may improve future treatment strategies to inhibit the spread of cancer metastasis and prolong the survival of CRC patients. Current AJCC guidelines do not adopt LVI as a prognostic indicator for CRC in TNM staging systems. However, studies have shown that LVI is an independent risk factor for survival in CRC patients(11). LVI also indicated resistance to neoadjuvant chemoradiotherapy(12). Although several research had constructed models to predict LVI, part of them adopted postoperatively pathological data to predict LVI, which seems to be disadvantaged in early prediction(13, 14). There were also studies established pre-operative prediction models based on imaging data, including magnetic resonance (MR) and computed tomography (CT)(15, 16). However, imaging data is not as easy to obtain as blood indexes, and the analysis results are greatly affected by imaging quality. The additionally, most research adopted lineal regression model, but rare research adopted ANN. ANN is a good solution to the shortcomings of traditional linear regression with the characteristics of self-adaptive, self-organizing and real-time learning. Therefore, our research aimed to construct pre-operative prediction model for LVI based on blood indexes via ANN.

Our research determined CA125, CA199 and fibrinogen were independent predicted factors for LVI in CRC patients. Among them, CA125 and fibrinogen were also reported to be associated with LVI in endometrial cancer(17). Based on these 3 factors, we constructed logistic regression model and ANN respectively. ANN showed inspiring AUC and sensitivity compared with logistic regression model (AUC: 0.839 *vs* 0.692; sensitivity: 92% *vs* 56%). The performance of ANN was similar as the multimodal radiomics model reported in rectal cancer (AUC: 0.839 *vs* 0.884; sensitivity: 92% *vs* 93.8%)(15). Blood indexes are faster, simpler and cheaper than imaging indexes. The specificity of ANN was lower than logistic regression model, but higher than reported multimodal radiomics model (74% vs 83% & 72.7%). On the premise of high sensitivity, this sacrifice in specificity is acceptable.

Conclusion

Based on accessibly blood indexes (CA125, CA199 and fibrinogen), we constructed an ANN for pre-operative prediction of LVI in CRC patients. The model showed satisfied performance compared with conventional lineal regression model. The ANN may provide potential value in predicting metastasis and recurrence of CRC patients.

Declarations

Conflict of Interest

The authors declare that they have no competing interests.

Author Contributions

DW and ZY contributed to the statistical analysis of the data. SW and YQ contributed to the data collection. ZM and HH contributed to the design of the study. All authors read and approved the final version of the manuscript.

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Not applicable.

Availability of data and materials

The datasets used and analyzed during the current study are available from the corresponding author on reasonable request.

Ethics approval and consent to participate

All procedures involving human participants were performed in accordance with Shanghai Pudong Hospital ethical committee (2020 No. W2-007) and with the 1964 Declaration of Helsinki and its later amendments or comparable ethical standards. All patients provided their written informed consent. The study protocol was approved by the Pudong Hospital Committee on human research.

Patient consent for publication

Written informed consent for publication was obtained from all the participants.

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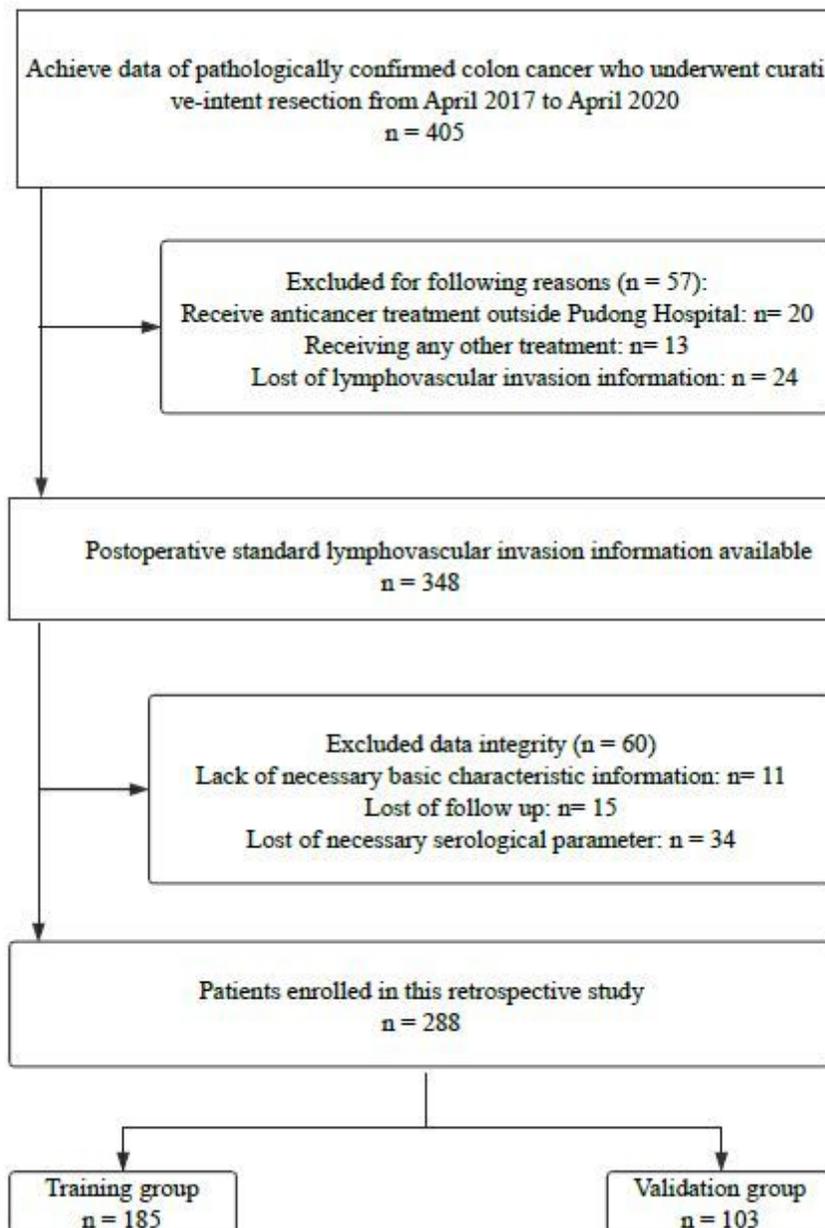
Figures

Figure 1

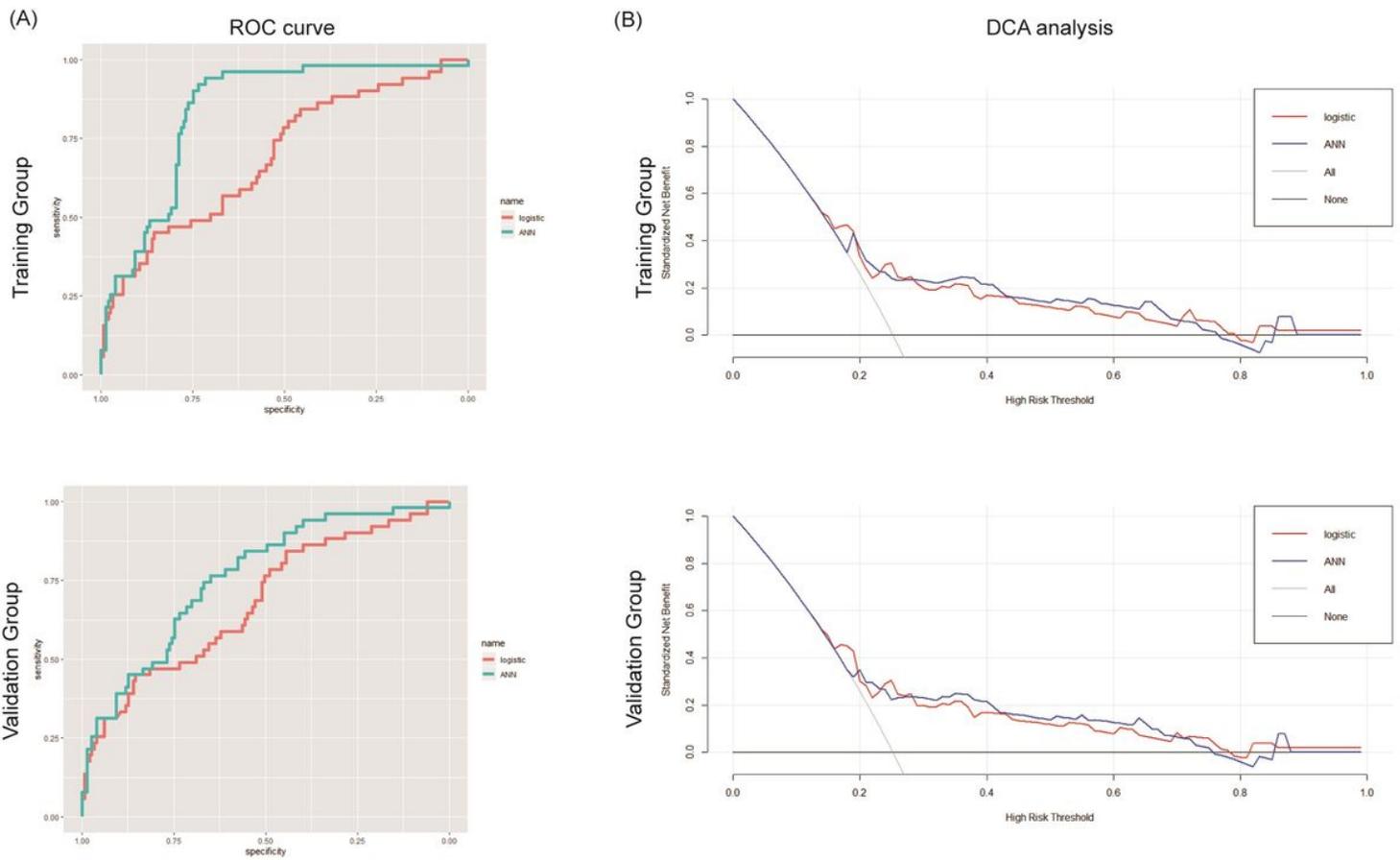Screening process of enrolled patients

Figure 2

Comparison of logistic regression model and artificial neural network. A. ROC curve analysis of logistic regression model and artificial neural network. B. Decision curve analysis (DCA) for each model. The y-axis measures the net benefit, which is calculated by summing the benefits (true-positive findings) and subtracting the harms (false-positive findings), weighting the latter by a factor rela