ISSN: 2475-7330 Volume 7, Number 2, June 2023

Scientific Research Publishing

Yangtze Medicine

ISSN: 2475-7330

https://www.scirp.org/journal/ym

Journal Editorial Board

ISSN Print: 2475-7330 ISSN Online: 2475-7349

https://www.scirp.org/journal/ym

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Editorial Board

Scientific Research Publishing

Yangtze Medicine, 2023, 7, 55-153
https://www.scirp.org/journal/ym
ISSN Online: 2475-7349
ISSN Print: 2475-7330

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Volume 7 Number 2 June 2023

Yangtze Medicine (YM)
Journal Information

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Scientific Research Publishing

Yangtze Medicine, 2023, 7, 55-62
https://www.scirp.org/journal/ym
ISSN Online: 2475-7349
ISSN Print: 2475-7330

Study on the Influencing Factors of Chemotherapeutic-Related Taste Changes in Cancer Patients

Xunya Xiong[1*], Zaonv Dong[1*], Gaopeng Zhang[2], Jun'e Hu[2#]

[1]Xianning Central Hospital, Xianning, China
[2]Jingzhou Central Hospital, Jingzhou, China
Email: [#]674209597@qq.com

How to cite this paper: Xiong, X.Y., Dong, Z.N., Zhang, G.P. and Hu, J.E (2023) Study on the Influencing Factors of Chemotherapeutic-Related Taste Changes in Cancer Patients. *Yangtze Medicine*, **7**, 55-62.
https://doi.org/10.4236/ym.2023.72006

Received: March 20, 2023
Accepted: May 5, 2023
Published: May 8, 2023

Abstract

Objective: To investigate the current situation of chemotherapy-related taste changes of cancer patients in Jingzhou area, and analyze the influencing factors of chemotherapy-related taste changes of cancer patients. **Methods:** In this study, 233 patients with malignant tumors who were confirmed by pathological examination and expected survival time of more than 6 months after admission to a tertiary general hospital in Jingzhou from January 2018 to October 2018 were selected by convenience sampling method. The Chinese version of The chemotherapy-induced Taste Alternation Scale (CiTAS) was used to investigate the baseline data and occurrence status, and multiple regression analysis was used to explore the influencing factors. **Results:** In this study, 171 tumor patients experienced chemotherapy related taste changes, accounting for 73.4% (171/233); The vast majority of chemotherapy patients have different types and severity of taste changes; Multiple regression analysis showed that the duration of chemotherapy, the number of consecutive days of chemotherapy, and dry mouth were the main influencing factors for chemotherapy related taste changes in cancer patients ($p < 0.05$). **Conclusion:** Nursing personnel should pay attention to the occurrence of chemotherapy-related taste changes in tumor patients, and provide predictive nursing interventions to improve their taste experience for tumor patients who have a long course of chemotherapy, many consecutive days of chemotherapy, or have dry mouth conditions.

Keywords

Cancer Patients, Chemotherapy, Taste Change, Influencing Factors

*Xunya Xiong and Zaonv Dong are the parallel first authors and have made equal contributions to this article.
[#]Corresponding author.

1. Introduction

According to statistics, about 3.12 million new cancer patients are added every year in China [1]. Chemotherapy is one of the important means of anti-tumor treatment for tumor patients, but chemotherapy drugs can kill tumor cells and damage normal taste bud cells and other structures, thus causing chemotherapy-related taste changes [2], the incidence of which is as high as 38% - 84% [3]. Once chemotherapy-related taste changes occur in tumor patients, it will not only stimulate negative emotions such as anxiety and psychological pain, but also induce symptoms such as malnutrition, and the severe cases will be affected by treatment interruption [4]. The purpose of this study is to investigate the current situation of chemotherapy-related taste changes in cancer patients and analyze the influencing factors, so as to provide a practical basis for clinical nurses to implement predictive nursing intervention, so as to improve the taste experience of cancer patients undergoing chemotherapy.

2. Research Object

With a convenient sampling method, 233 cancer patients who were to be treated with chemotherapy drugs for more than one course in a tertiary general hospital in Jingzhou City from January 2018 to October 2018 were selected as the study objects. The inclusion criteria of the patients were: 1) pathological diagnosis of malignant tumor; 2) age ≥ 18 years old; 3) chemotherapy; 4) expected survival time > 6 months; 5) clear consciousness of the patients, no language communication and cognitive dysfunction, able to independently complete the questionnaire questions and answers; 6) the patients agreed to accept the survey. Exclusion criteria: 1) patients with concurrent radiotherapy and chemotherapy; 2) patients with oral infection; 3) history of esophageal reflux; 4) those who can't eat by mouth; 5) those who have their own taste disorders.

3. Method

3.1. Research Tools

Including: 1) General information questionnaire: age, gender, nationality, residence, marital status, education level, working status after illness, medical treatment, chemotherapy plan, times of chemotherapy, days of chemotherapy, disease diagnosis, time of diagnosis, tumor stage, pathological type, metastasis, smoking, drinking, dry mouth, olfactory changes and complications. 2) Chinese version of the chemotherapy-induced taste alteration scale (CiTAS) [5]: prepared by Kano *et al.* in 2013 to evaluate the degree of taste alteration of chemotherapy patients. In 2017, Qian Lijing *et al.* authorized by the original author, Sinologized it. The scale includes taste change (6 items), unpleasant taste change (6 items), unpleasant symptoms and problems (6 items), and 18 items in total. The scale uses a Likert5-grade scoring method, from "no" to "very serious" respectively 1 - 5 points, all items are positive scoring. The higher the score is, the more serious the taste change is. The internal consistency coefficient was 0.766

and the retest reliability was 0.705.

3.2. Research Methods

The questionnaire was uniformly trained and explained by the researcher himself to the investigator, and was distributed and recycled on the spot. Before all questionnaires were distributed, the purpose of the study was explained to the patient and the caregiver to obtain the understanding and cooperation of the patient and the family members. When the respondents' physical, visual or educational level affects their answers, they should get the consent of the respondents, and read and explain the questions by the investigator. After the respondents understand and make a choice, they are authorized to fill in the questions on behalf of the investigator. In fact, 240 questionnaires were sent out and 233 effective questionnaires were recovered, the effective rate was 97.08%.

3.3. Statistical Methods

SPSS 19.0 statistical software was used to analyze the data. Frequency and percentage were used to describe the counting data, mean and standard deviation were used to describe the measurement data, and multiple linear regression was used to analyze the main influencing factors of chemotherapy-related taste changes, $p < 0.05$.

4. Results

4.1. General Information

In this study, 233 tumor patients participated in the investigation; 130 patients with dry mouth, 103 patients without dry mouth; 63 cases had olfactory changes and 170 cases had no olfactory changes. See Table 1 for other general information.

4.2. Current Status of Chemotherapy-Related Taste Changes

Among 233 cases, only 62 tumor patients did not have chemotherapy-related taste changes. Most of the chemotherapy patients had taste changes of different types and severity, as shown in Table 2.

4.3. Analysis of Influencing Factors

The results of logistic regression analysis showed that the duration of chemotherapy, the days of continuous medication, and dry mouth were the influencing factors for chemotherapy-related taste changes, as shown in Table 3.

5. Discussion

Of 233 patients, only 62 patients did not have chemotherapy-related taste changes, accounting for 26.6%. It can be seen that the incidence of chemotherapy-related taste changes in cancer patients is quite high. Paying attention to the taste changes of patients with cancer chemotherapy is the basis of tumor symp-

tom management, and also the key link to ensure the effect of chemotherapy [6]. The influencing factors of chemotherapy related taste changes in cancer patients were analyzed as follows.

Table 1. General information of 233 tumor patients.

Project		Number of cases (n)	Percentage (%)
Gender	female	95	40.77
	male	138	59.23
Age	<60 years old	127	54.51
	Over 60 years old	106	45.49
Nation	Han nationality	229	98.28
	Ethnic minority	4	1.72
Place of residence	City	56	24.03
	countryside	177	75.97
Marital status	unmarried	6	2.58
	married	213	91.42
	Divorce	6	2.58
	Widowed spouse	8	3.42
Degree of education	illiteracy	8	3.42
	primary school	95	40.78
	junior high school	87	37.34
	High school/technical secondary school	38	16.31
	College/University	5	2.15
Mode of seeking medical treatment	Medical insurance for residents	77	33.05
	Rural cooperative medical care	145	62.23
	Commercial insurance	1	0.43
	At their own expense	10	4.29

Table 2. 233 cases of tumor patients with chemotherapy-related taste changes.

Taste change type	Number of cases (n)	Percentage (%)	CiTAS score ($\overline{x} \pm s$)
No change	62	26.6	19.97 ± 0.275
Bad taste	53	22.7	22.91 ± 0.292
Absence of taste	41	17.6	23.66 ± 0.304
Phantom taste	36	15.5	23.14 ± 0.363
Hypofunction	31	13.3	23.39 ± 0.406
Gustatory error	10	4.3	22.60 ± 0.806

Table 3. Multiple regression analysis results of influencing factors of chemotherapy-related taste changes in tumor patients.

Item	β	S.E	Wald value	OR value	p value	95% CI
Constant term	−3.932	1.245	9.979	0.020	0.002*	
Age	−0.362	0.368	0.965	0.697	0.326	0.339 - 1.433
Course of disease	0.291	0.275	1.113	1.337	0.291	0.779 - 2.295
Course of treatment	1.379	0.389	12.557	3.969	0.000*	1.852 - 8.509
Days of medication	0.664	0.200	11.007	1.942	0.001*	1.312 - 2.874
Chemotherapy regimen	0.369	0.391	0.891	1.446	0.345	0.672 - 3.112
Smoking or drinking	−20.285	27604.277	0.000	0.000	0.999	0.000 - 0.000
Dry mouth or not	0.951	0.342	7.758	2.590	0.005*	1.326 - 5.058
Transfer or not	0.085	0.358	0.058	1.089	0.813	0.539 - 2.197

Note: "*" is $p < 0.05$, the difference is statistically significant.

5.1. Chemotherapy Time

The results of this study show that most cancer patients will have different degrees of taste changes at the beginning of chemotherapy, and its severity will gradually increase with the passage of time, until the most serious 3 - 5 days after chemotherapy. Previous studies have reported that chemotherapy-related taste changes may occur in cancer patients during chemotherapy or during the interval of chemotherapy, lasting for several hours or days or months [7], the most serious 5 - 7 days after general chemotherapy [8]. It can be seen that the results of this study show that the most serious change in chemotherapy-related taste is a little earlier than 1 - 2 days. The analysis of the causes may be related to the strengthening of health education for cancer chemotherapy patients by the specialized nurses of the oncology department, so that they gradually improve their awareness of the change in chemotherapy-related taste. Domestic research shows that chemotherapy related taste changes are usually temporary, and patients can gradually return to normal after chemotherapy, with a recovery time of about 6 months [9].

5.2. Whether There Are Complications

Based on the analysis of the results of this study, whether there is dry mouth in tumor patients is the influencing factor of chemotherapy-related taste changes, which is consistent with the conclusion of Qian Lijing and other researchers. The results showed that the type and degree of taste changes were related to dry mouth, olfactory changes and hypertension before chemotherapy. However, it is not clear that the mechanism of xerostomia is related to the decrease of saliva and the limitation of chemical receptors. Rich saliva can provide an ionic environment for taste cells to sense signals and conduct them. Chemotherapy drugs arrive at taste cells in the mouth with blood, producing bad taste, such as salty,

bitter or metallic taste [10]. Salivary gland is a very sensitive gland to chemotherapy drugs [11]. Once stimulated by chemotherapy drugs, the gland at the initial stage shows an increase in secretions and oral secretions, which can take away some bad taste. However, the serious damage of chemotherapy drugs to normal salivary gland cells has been irreversible, resulting in the decline of salivary secretion function, and finally, there is a small amount of saliva and viscosity, xerostomia, deterioration of chemotherapy-related taste changes in cancer patients [12]. In addition, relevant domestic research shows that the sensitivity of tumor patients with hypertension and diabetes to sweet and salty tastes is significantly lower than that of healthy people, which may cause tumor patients to take more sugar, salt or condiment, which may aggravate the degree of chemotherapy-related taste change of tumor patients [13].

5.3. Other Factors

The results of this study show that whether or not chemotherapy-related taste changes occur in cancer patients and the severity is not related to gender and age, which is consistent with the results of investigation and analysis by Qian Lijing and other domestic researchers [5], but through histological research, foreign researchers found that women have more fungiform taste buds and taste pores than men, and confirmed that women's sensitivity to taste is indeed higher than men's [14]. McGreevy J and other research report that the change of taste in female patients is often more serious than that in male patients, and with the increase of age, the degree of taste change will decrease [15]. The analysis of the reasons may be related to the gender and age distribution proportion of the study subjects. Early studies have pointed out that the incidence and duration of chemotherapy-related taste changes are different in patients with different tumor types [16]; when patients with head and neck tumors receive chemotherapy combined with radiotherapy, the incidence of taste changes increases, the occurrence time is earlier, and the duration is longer [17]. This study involves a large number of tumor types, and the sample size of some tumor types may be insufficient. Moreover, the inclusion criteria limit the inclusion of tumor patients undergoing concurrent radiotherapy and chemotherapy, leading to a certain deviation in the results of this study.

6. Summary

The nurses should pay more attention to the changes in taste in patients with cancer, effectively intervene and control the occurrence of xerostomia, chemotherapy or other complications, and improve the taste experience of patients with cancer chemotherapy. At the same time, we should pay attention to the compliance and tolerance of patients' chemotherapy plans, chemotherapy course and days of continuous chemotherapy, establish multi-disciplinary medical cooperation, give systematic and personalized predictive nursing intervention strategies in time, and reduce the occurrence of chemotherapy-related taste

changes of tumor patients. However, most of the cases in this survey are treated with several drugs combined with chemotherapy, and no single chemotherapy drug is involved. Whether there is a difference in chemotherapy-related taste changes of tumor patients caused by different chemotherapy drugs remains to be further studied.

States

The human data in this study were conducted according to the Helsinki Declaration with the informed consent of the patient. There are no interests or disputes in this article.

Acknowledgements

This study was supported by the 2018 Jingzhou Medical and Health Technology Development Plan project.

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Scientific Research Publishing

Yangtze Medicine, 2023, 7, 63-75
https://www.scirp.org/journal/ym
ISSN Online: 2475-7349
ISSN Print: 2475-7330

Risk Prediction Model for Surgical Treatment of Ruptured Corpus Luteum in the Ovary

Yuqi Qiu, Sufei Wang, Yong Chen, Wenrong He, Xiaowen Wang*

Department of Obstetrics and Gynecology, The First Affiliated Hospital of Yangtze University, Jingzhou, China
Email: *wxw0726@sohu.com

How to cite this paper: Qiu, Y.Q., Wang, S.F., Chen, Y., He, W.R. and Wang, X.W. (2023) Risk Prediction Model for Surgical Treatment of Ruptured Corpus Luteum in the Ovary. *Yangtze Medicine*, **7**, 63-75.
https://doi.org/10.4236/ym.2023.72007

Received: March 8, 2023
Accepted: May 8, 2023
Published: May 11, 2023

Abstract

Objective: To explore the related factors of surgical treatment of patients with corpus luteum rupture and establish a risk prediction model of surgical treatment of corpus luteum rupture. **Methods**: 222 patients with corpus luteum rupture treated in Jingzhou First People's Hospital from January 2015 to March 2022 were analyzed retrospectively, including 45 cases of surgery and 177 cases of conservative treatment. The training set and validation set were randomly assigned according to 7:3. We collected the basic information, laboratory and ultrasonic examination data of 222 patients. Logistic regression analysis was used to determine the independent risk factors and combined predictors of surgical treatment of corpus luteum rupture. The risk prediction model was established and the nomogram was drawn. The discrimination and calibration of the prediction model were verified and evaluated by receiver operating characteristic (ROC) curve, calibration curve and Hosmer-Lemeshow goodness of fit test; Decision curve analysis (DCA) was used to evaluate the clinical effectiveness of the prediction model. **Results**: Univariate logistic regression showed that whole abdominal pain (OR: 2.314, 95% CI: 1.090 - 4.912), abdominal muscle tension (OR: 2.379, 95% CI: 1.112 - 5.089), adnexal mass ≥ 4 cm (OR: 3.926, 95% CI: 1.771 - 8.266), hemoglobin < 12 g (OR: 11, 95% CI: 4.724 - 25.616), pelvic effusion depth ≥ 3 cm under ultrasound (OR: 10.606, 95% CI: 4.602 - 24.445) and positive cervical lifting pain (OR: 3.960, 95% CI: 1.831 - 8.563) were suspected risk factors for surgical treatment of corpus luteum rupture; Multivariate logistic regression analysis showed that hemoglobin < 12 g (OR: 5.398, 95% CI: 1.985 - 14.682), pelvic effusion depth ≥ 3 cm under ultrasound (OR: 6.256, 95% CI: 1.607 - 24.354) and positive cervical lifting pain (OR: 2.995, 95% CI: 1.19 - 7.538) were independent risk factors for surgical treatment of corpus luteum rupture ($P < 0.05$). The nomogram is drawn according to the prediction variables, and the prediction model is constructed. The prediction model predicted that the area under the ROC curve (AUC) of patients with corpus luteum rupture in the training set was 0.841, 95% CI (0.759, 0.922), and the area under the ROC

curve (AUC) of patients with corpus luteum rupture in the validation set was 0.919, 95% CI (0.821, 0.999). **Conclusion**: The nomogram prediction model containing three predictive variables (hemoglobin, depth of pelvic effusion under ultrasound and cervical lifting pain) can be used to predict the risk of surgical treatment in patients with corpus luteum rupture.

Keywords

Corpus Luteum Rupture, Surgical Treatment, Prediction Model

1. Introduction

Ovary luteal rupture is one of the most common gynaecological emergencies [1], usually with an acute onset and mostly unilateral rupture, with the right side being the most common [2]. Ovarian rupture of the corpus luteum can occur at all ages in women of childbearing age [3]. The most common clinical presentation is sudden onset of lower abdominal pain on one side, with atypical accompanying symptoms and non-specific ancillary investigations. If the diagnosis is not made early, timely and accurately, the condition is easily delayed and some patients may experience massive intra-abdominal bleeding, leading to haemorrhagic shock and even life-threatening. There are no clear guidelines on the choice of treatment for ruptured corpus luteum at home or abroad, and currently, there are clinical categories of surgical and conservative treatment, with each option having its own advantages and disadvantages [4]. Both open and laparoscopic surgery may result in long-term complications such as ovarian damage, pelvic adhesions and skin scarring, especially with the widespread use of monopolar and bipolar energy devices in laparoscopic surgery. The ensuing electrical and thermal radiation may cause irreversible damage to ovarian reserve function and may have long-term effects on patients with a ruptured corpus luteum who have a need for fertility. The trauma caused by thermal radiation is insidious and not easily detected intraoperatively, with symptoms often appearing around 2 weeks after the procedure [5].

There is currently no method of assessing the risk associated with surgery in the treatment of ruptured corpus luteum in national and international studies. The aim of this study is to investigate the factors influencing the surgical treatment of patients with ruptured corpus luteum and to develop a predictive model to provide an effective predictive tool for identifying high-risk patients, to further scientifically guide subsequent treatment, and to strictly control the indications for surgery so as to draw up individualized and more optimal treatment plans.

2. Subjects and Methods

2.1. Study Subjects

222 patients with ruptured corpus luteum admitted to the First People's Hospital

of Jingzhou City from January 2015 to March 2022 were selected, of which 45 were operated and 177 were conservatively treated. To establish and validate the prediction model the study population was randomly divided into two parts according to 7:3, with 172 cases (≈70%) in the training cohort and 50 cases (≈30%) in the validation cohort. Inclusion criteria: 1) admission with sudden onset of abdominal pain, ultrasound suggestive of a mass in the adnexal region and pelvic effusion; 2) diagnosis confirmed by postoperative pathology or conservative dynamic observation. Exclusion criteria: 1) previous history of pathological pelvic masses considered (untreated); 2) positive blood β-HCG; 3) no liver, kidney or haematological disease and no recent use of anticoagulant drugs.

2.2. Data Collection

Data were collected through a computer terminal, using the hospital's electronic medical record system and digital case system, according to a pre-designed clinical information questionnaire. One patient was included in the surgery group after 1 day of conservative treatment, when abdominal pain increased and was referred for surgery. Data collected included treatment modality, age, body mass index (BMI = weight (kg)/height $(m)^2$), marital status, regularity of menstruation (cycles of 21 - 35 days were considered regular), predisposing factors, smoking status (yes defined as ≥1 cigarette per day), alcohol consumption status (yes defined as ≥1 drink per week, history of previous pelvic surgery, whether abdominal muscle tension, cervical lifting pain on post-admission examination Laboratory and ultrasound indicators include hemoglobin (Hb), white blood cells (WBC), platelets (PLT), international standardized ratio (INR), C-reactive protein (CRP), and ultrasound monitoring of pelvic fluid depth and adnexal mass size through vagina (with sexual history) or rectum (without sexual history).

2.3. Statistical Methods

SPSS (Version 26.0; IBM, Armonk, New York) and Stata (Version 15.0; Stata Corporation) were used to analyse the data. Count data were statistically described using frequencies and two groups were compared according to data characteristics using Pearson's chi-square test, continuous corrected chi-square test and Fisher, s exact test, respectively. Variables with $P < 0.05$ were included in the multi-factor logistic regression analysis using one-way logistic regression with forced, forward, backward and stepwise regressions to obtain a total of four clinical prediction models, with the best model selected based on the AIC and likelihood ratio tests and plotted on column plots. The prediction models were assessed in terms of discrimination, calibration and net clinical benefit, with discrimination assessed by the area under the subject operating characteristic curve (ROC curve) (AUC); calibration assessed by the calibration curve and Hosmer-Lemeshow goodness of fit test; and clinical validity assessed by decision curve analysis (DCA). Differences were considered statistically significant at $P < 0.05$.

3. Results

3.1. Comparison of the Basic Clinical Characteristics of the Patients in the Training and Validation Sets

The p-values for body mass index, shock index and international normalized ratio in the training and validation sets were calculated using Fishers exact probability method as 0.553, 0.401 and 1, respectively; the p-values for regularity of menstruation, history of pelvic surgery, alcohol consumption and NRS pain score were calculated using the continuous corrected chi-square test as 1, 1, 0.275 and 0.719, respectively; the remaining variables were calculated using the Pearson chi-square test, none of the differences were statistically significant ($P > 0.05$), see Table 1.

3.2. Univariate and Multivariate Analysis for Predicting Risk of Luteal Rupture Surgery

Univariate analysis showed that there were six suspected risk factors associated with surgery for ruptured corpus luteum, namely cervical lifting pain, site of pain, abdominal muscle tension, haemoglobin, ultrasound monitoring of pelvic fluid depth and size of adnexal masses ($p < 0.05$, see Table 2). The statistically significant influencing factors from the univariate analysis were included in the multi-factor logistic regression analysis, which showed that cervical lifting pain, haemoglobin and ultrasound monitoring of pelvic fluid depth were statistically significant ($p < 0.05$, see Table 2).

3.3. Construction of the Prediction Model

A multivariate logistic regression prediction model was constructed using the mode of treatment for patients with ruptured corpus luteum as the dependent variable (assigned values: conservative = 0, surgery = 1) and three predictor variables screened by multivariate logistic regression analysis as the independent variables (assigned values shown in Table 1). The results showed that cervical lifting pain, haemoglobin and ultrasound monitoring of pelvic fluid depth were risk factors for surgical treatment of patients with ruptured corpus luteum ($p < 0.05$), and four clinical prediction models were constructed. Columnar plots, known as Nomograms, were also constructed based on the predictor variables (Figure 1). The column line graph allows the corresponding value of each variable to be scored, and then the scores of all variables are added together to obtain a total score, and a vertical line is drawn down from the total score to mark the estimated probability of surgical treatment risk for patients with ruptured corpus luteum.

3.4. Validation of the Prediction Model

The validation of the prediction model was based on the model discrimination and calibration, and the model discrimination was assessed by plotting the ROC curve of the prediction model predicting the risk of surgical treatment for patients

Table 1. Comparison of clinical basic information, laboratory and ultrasound examination between training set and verification set.

Variable	Training cohort (n = 172)	Validation cohort (n = 50)	χ^2	P	Variable	Training cohort (n = 172)	validation cohort (n = 50)	χ^2	P
Treatment					**Marital status**				
conservative (0)	136	41	0.206	0.65	unmarried (0)	108	28	0.753	0.386
surgical (1)	36	9			married (1)	64	22		
Age (y)					**Menstrual regularity**				
<35 (0)	164	49	0.184	0.403	yes (0)	165	48	0	1
≥35 (1)	8	1			no (1)	7	2		
BMI (kg/m^2)					**NRS Pain level (points)**				
<18.5 (0)	98	27		0.622	1 - 3 (1)	111	34	0.629	0.719
18.5 - 24 (1)	73	22			4 - 6 (2)	47	11		
>24 (2)	1	1			7 - 10 (3)	14	5		
Predisposing factors					**Cervical lifting pain**				
other (0)	67	18	0.143	0.705	no (0)	107	27	1.091	0.296
sex (1)	105	32			yes (1)	65	23		
Smoking					**Pain site**				
no (0)	155	40	3.711	0.054	lower abdomen (0)	95	30	0.358	0.55
yes (1)	17	10			whole abdomen (1)	77	20		
History of pelvic surgery					**Abdominal muscle tension**				
no (0)	166	48	0	1	no (0)	91	23	0.74	0.39
yes (1)	6	2			yes (1)	81	27		
Alcohol					**International standardized ratio**				
no (0)	159	49	1.194	0.275	<1.2 (0)	167	49		1
yes (1)	13	1			≥1.2 (1)	5	1		
Shock index					**C-reactive protein (mg/L)**				
0 - 1 (0)	171	49		0.401	<5 (0)	47	10	1.089	0.297
1 - 2 (1)	1	1			≥5 (1)	125	40		
Hemoglobin (g/L)					**Adnexal tumors (cm)**				
≥12 (0)	120	39	1.292	0.256	<4 (0)	106	35	1.172	0.279
<12 (1)	52	11			≥4 (1)	66	15		
Leukocyte (10^9/L)					**Pelvic effusion depth (cm)**				
<10 (0)	94	28	0.028	0.866	<3 (0)	123	39	0.827	0.363
≥10 (1)	78	22			≥3 (1)	49	11		
Platelets (10^9/L)									
<100 (0)	166	49	0.005	0.944					
≥100 (1)	6	1							

Table 2. Univariate and multivariate logistic regression analysis of the risk of surgical treatment of ovarian corpus luteum rupture.

Variable	Univariate logistic regression							Multivariate logistic regression						
	B	S.E	Wald	Sig	OR	95% CI for OR		B	S.E	Wald	Sig	OR	95% CI for OR	
						Lower	Upper						Lower	Upper
Age (y)														
<35	−0.641	1.086	0.349	0.555	0.527	0.063	4.423							
≥35														
BMI (kg/m²)														
<18.5	−0.096	0.372	0.066	0.797	0.909	0.439	1.883							
18.5 - 24														
>24														
Marital status														
unmarried	0.238	0.382	0.386	0.534	1.268	0.599	2.683							
married														
Menstrual regularity														
yes	0.433	0.859	0.254	0.614	1.541	0.286	8.291							
no														
Predisposing factor														
other	0.629	0.411	2.347	0.126	1.876	0.839	4.196							
sex														
Smoking														
no	−1.263	0.755	2.800	0.094	0.283	0.064	1.242							
yes														
Alcohol														
no	0.075	0.674	0.012	0.911	1.078	0.288	4.038							
yes														
History of pelvic surgery														
no	−19.919	16408.711	0	0.999	0	−19.91	16408.711							
yes														
Cervical lifting pain														
no	1.376	0.394	12.23	0.000	3.960	1.831	8.563	1.097	0.471	5.424	0.02	2.995	1.19	7.538
yes														
Pain site														
lower abdomen	0.839	0.384	4.776	0.029	2.314	1.090	4.912	0.537	0.488	1.21	0.271	1.71	0.657	4.451
whole abdomen														
Abdominal muscle tension	0.867	0.388	4.994	0.025	2.379	1.112	5.089	0.796	0.478	2.774	0.096	2.217	0.869	5.66
no														
yes														
Hemoglobin (g/L)														
≥12	2.398	0.431	30.91	0.000	11.000	4.724	25.616	1.686	0.51	10.911	0.001	5.398	1.985	14.682
<12														
Leukocyte (10^9/L)														
<10	0.236	0.375	0.396	0.529	1.267	0.607	2.644							
≥10														
Platelets (10^9/L)														
≥100	−0.663	0.887	0.559	0.455	0.515	0.091	2.931							
<100														

Continued

International standardized ratio <1.2 ≥1.2	0.959	0.933	1.056	0.304	2.608	0.419	16.231							
C-reactive protein (mg/L) <5 ≥5	0.546	0.461	1.401	0.237	1.726	0.699	4.263							
Benign adnexal tumors (cm) <4 ≥4	1.342	0.393	11.663	0.001	3.826	1.771	8.266	−0.58	0.696	0.695	0.405	0.56	0.143	2.191
Pelvic effusion depth (cm) <3 ≥3	2.361	0.426	30.723	0.000	10.606	4.602	24.445	1.834	0.693	6.991	0.008	6.256	1.607	24.354
NRS Pain level (points) 1 - 3 4 - 6 7 - 10	0.498	0.274	3.316	0.069	1.646	0.963	2.815							

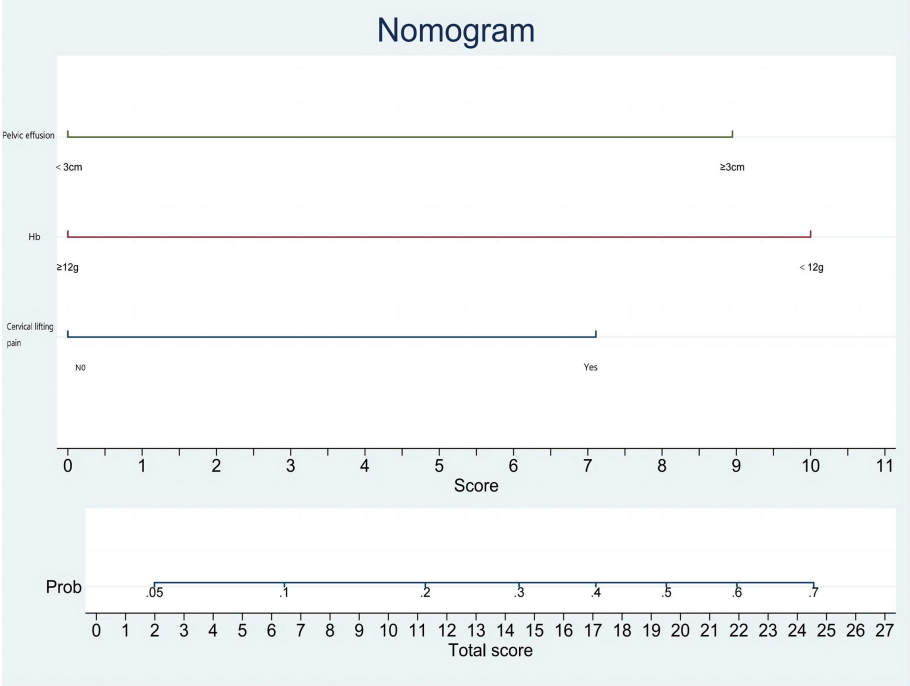


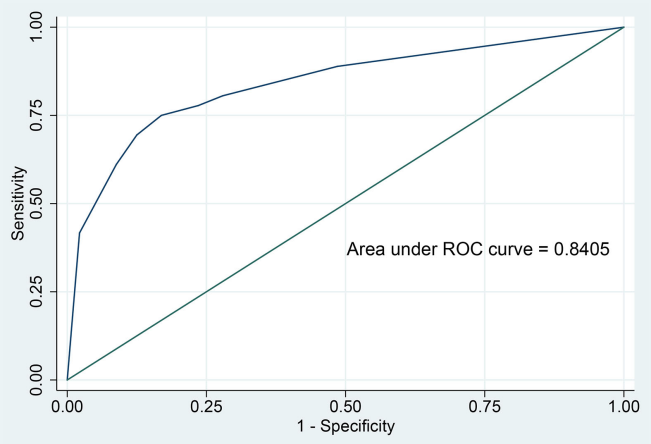
Figure 1. Nomogram of surgical risk in patients with ovarian corpus luteum rupture.

with ruptured corpus luteum. The AUC for the training cohort was 0.841 [95% CI (0.760, 0.922)] with a cut-off value of 0.263 (**Figure 2(a)**); the AUC for the validation cohort was 0.919 [95% CI (0.826, 0.999)] with a cut-off value of 0.128 (**Figure 2(b)**), indicating that the prediction model had good discriminatory power. Meanwhile, the Hosmer-Lemeshow goodness-of-fit test showed a good fit (training cohort P = 0.93; validation cohort P = 0.41), indicating that the pre-

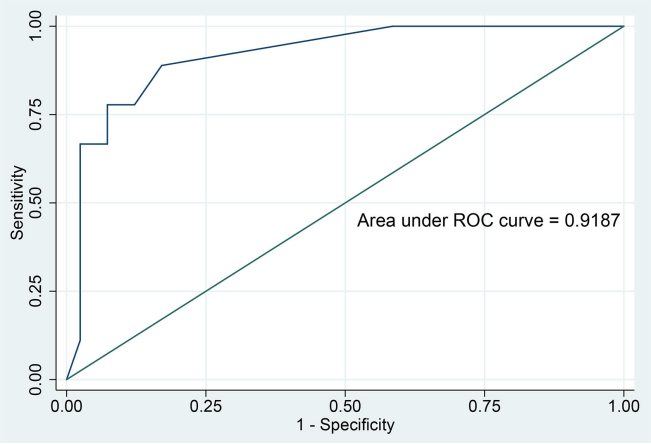
dicted probabilities of the model were generally consistent with the actual probabilities and had a good calibration. In addition, the calibration curves for the training and validation cohorts showed moderate agreement and the prediction model had good calibration ability (**Figure 3**). In summary, the Nomogram of the prediction model has good prediction capability.

3.5. Clinical Application

The clinical validity of the prediction model was assessed using the DCA for the column line graph of the probability of occurrence of surgical treatment for patients with ruptured corpus luteum is shown in **Figure 4**. The results show that using this column line graph to predict the risk of surgical treatment for patients with ruptured corpus luteum in the current study would have been more beneficial than implementing an intervention program for all patients if the threshold probabilities for patients and physicians were each >20%, and that the net benefit of the prediction model was significantly higher in this range than in two extreme cases, where all patients received clinical interventions.

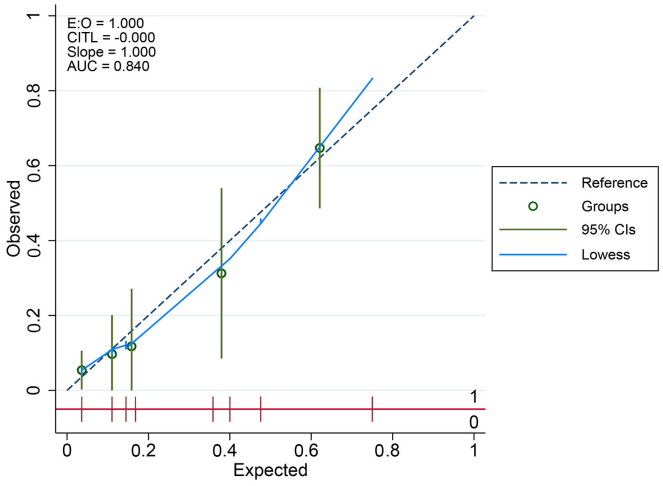


(a)

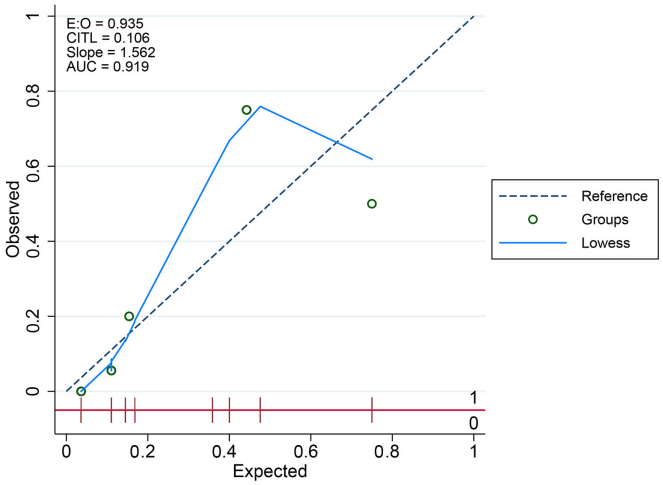


(b)

Figure 2. ROC curve of prediction model in training cohort and validation cohort. (a) Traning cohort; (b) Validation cohort.

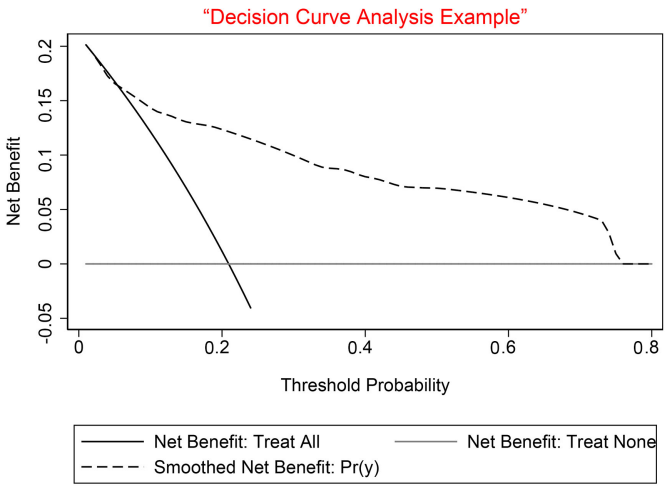


(a)

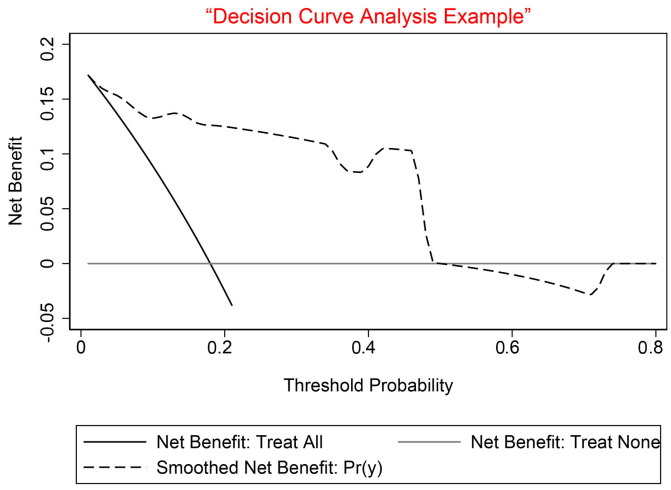


(b)

Figure 3. Calibration curve of prediction model in training cohort and validation cohort. (a) Traning cohort; (b) Validation cohort.



(a)



(b)

Figure 4. DCA curve of prediction model in training cohort and validation cohort. (a) Traning cohort; (b) Validation cohort.

4. Discussion

In recent years, nomograms have been widely used for risk prediction in oncology and chronic diseases, both nationally and internationally because of their high utility and reliability [6] [7]. They can be used to visualize and quantify the occurrence and prognosis of various diseases. However, there is no literature on the assessment of the risk of surgical treatment in patients with ruptured corpus luteum.

This study establishes and validates a new tool to predict the risk of surgical treatment in patients with ruptured corpus luteum by obtaining three surgically relevant and readily available influencing factors through multivariate logistic regression. The incorporation of these risk factors into a concise nomogram enables individualised prediction of the risk of surgvalidation cohortsical treatment in patients with ruptured corpus luteum. This study provides a relatively accurate predictive tool for predicting the risk of surgical treatment in patients with ruptured corpus luteum. The AUCs of the training and were 0.841 and 0.919 ($p < 0.05$) respectively, indicating that the constructed nomogram had good predictive ability. In addition, the DCA curves also suggested that the prediction model has good clinical validity.

This study showed that haemoglobin < 12 g, pelvic fluid depth > 3 cm on ultrasound and cervical lifting pain (+) were independent risk factors for the risk of surgery in patients with ruptured corpus luteum. In a retrospective study by Seok *et al.* [8], the results showed that CT suggestive of active bleeding from the lesion and depth of pelvic effusion made a significant difference in the choice of treatment modality with ORs of 3.773 and 1.318 respectively ($p < 0.01$). In the ROC curve with the depth of pelvic blood collection as the test variable, the best cut-off value (cut-off) was measured at 5.8 cm, where the sensitivity was 73.7% and the specificity was 58.6% (P = 0.004). The conclusions of the article suggest

that surgery is 5.786 times more risky than conservative treatment for patients with ruptured corpus luteum with active bleeding measured by CT to a depth of >5.8 cm in the pelvis. This finding is consistent with the findings of this study. A study by Mi Ju Kim *et al.* [9] showed that the surgical group had more pelvic blood loss (325 ± 250 ml vs 206 ± 146.5 ml, p = 0.002) and lower haemoglobin (11.3 ± 1.4 g/dL vs 12.2 ± 1.2 g/dL; p = 0.007) on preoperative assessment compared to the conservative treatment group and CT suggested a high volume of pelvic effusion (single deepest depth in the literature) (6.7 ± 2.2 cm vs. 5.1 ± 1.5 cm, p = 0.006), all differences being statistically significant and consistent with the findings of this study. However, active bleeding from the lesion requires enhanced CT for effective assessment [10] [11], which is less feasible for primary care and emergency patients, and ultrasound monitoring is more clinically appropriate in comparison. In a study by Wei *et al.* [4], it was shown that for both functional and non-functional ruptured ovarian cysts, the surgical group had a larger cyst volume and more pelvic fluid compared to the conservative treatment group, and the difference was statistically significant, consistent with the results of this study. This study also suggests that cervical lifting pain can be an independent risk factor for assessing the need for surgical treatment, but this has not been reported.

In 1993, the rate of surgery for patients with ruptured corpus luteum could reach 83% [12], but with continuous advances in all aspects of medical technology, conservative treatment has become the trend [3]. However, surgery remains the only treatment option for patients with unstable vital signs, unremitting symptoms, progressive worsening of anaemia and imaging suggestive of increased pelvic effusion [13]. Platelet anemia, systemic lupus erythematosus, renal failure, and coagulation abnormalities due to oral anticoagulants can all lead to severe abdominal bleeding in patients with luteal rupture [14] [15] [16] [17], while case reports suggest that deficiency of α-1 trypsin can even lead to recurrent luteal rupture [18].

Although this study was the first to establish a predictive model for the risk of surgical treatment in patients with ruptured corpus luteum, the degree of differentiation and calibration was fair, and the DCA curve suggested that the predictive model had good clinical validity. However, there are still shortcomings in this study. Firstly, the study excluded patients with early pregnancy and coagulation disorders, and only provided risk prediction for the surgical treatment of luteal rupture in normal non-pregnant women of childbearing age. Secondly, this study is a single-centre retrospective study and the predictive validity of the line graph prediction model needs to be validated with more external data, especially in multicentre, large-sample cohort studies with different regions and ethnicities; thirdly, this study did not refer to the basal haemoglobin level of patients, nor did it refer to the dynamic detection of haemoglobin changes in patients whose vital signs were basically stable after admission and who did not want to undergo surgery for the time being, so the assessment of haemoglobin on actual blood loss is not well guided. Finally, the model in this study included

fewer risk factors. Therefore, more risk factors should be included in the next validation studies to further improve the predictive power of the model.

In conclusion, this study found that total abdominal pain, abdominal muscle tension, adnexal mass ≥ 4 cm, haemoglobin < 12 g, pelvic fluid depth ≥ 3 cm on ultrasound, and positive cervical lift pain were suspected risk factors for surgical treatment of patients with ruptured corpus luteum. Using multivariate logistic regression analysis, we screened the three best risk factors and created a nomogram with relatively high accuracy to predict the risk of surgical treatment for patients with ruptured corpus luteum and to provide individualised treatment for patients.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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Scientific Research Publishing

Yangtze Medicine, 2023, 7, 76-86
https://www.scirp.org/journal/ym
ISSN Online: 2475-7349
ISSN Print: 2475-7330

Advances of Genetic Testing Technology in Etiology Diagnosis of Recurrent Spontaneous Abortion

Qing Xiao[1], Ziwei Li[2], Jinzhi Lu[3*]

[1]The First Affiliated Hospital of Yangtze University, Jingzhou, China
[2]Department of Pathology, The Central Hospital of Xiaogan, Xiaogan, China
[3]Hubei Provincial Clinical Research Center for Personalized Diagnosis and Treatment of Cancer, Department of Central Laboratory Medicine, The First Affiliated Hospital of Yangtze University, Jingzhou, China
Email: *jinzhilu2015@163.com

How to cite this paper: Xiao, Q., Li, Z.W. and Lu, J.Z. (2023) Advances of Genetic Testing Technology in Etiology Diagnosis of Recurrent Spontaneous Abortion. *Yangtze Medicine*, **7**, 76-86.
https://doi.org/10.4236/ym.2023.72008

Received: March 8, 2023
Accepted: June 4, 2023
Published: June 7, 2023

Abstract

Recurrent spontaneous abortion (RSA) is a complex and heterogeneous disorder with multiple etiologies. Genetic factors are thought to play an important role in the etiology of RSA. With recent advances in genetic testing technologies, there has been an increasing interest in using these tools to diagnose the etiology of RSA. This review discusses the different types of genetic testing methods, such as karyotyping, chromosomal microarray analysis, next-generation sequencing, and their applications in the diagnosis of the etiology RSA. The use of genetic testing in the diagnosis of RSA has the potential to improve the accuracy of diagnosis and the understanding of the underlying mechanisms of the disorder, which could lead to better management and treatment of affected individuals.

Keywords

Recurrent Spontaneous Abortion, Etiology, Genetic Testing Technology

1. Introduction

Abortion is the most common complication of human pregnancy, accounting for approximately 10% - 15% of clinically confirmed pregnancies [1]. According to the definition of American Society for Reproductive Medicine (ASRM) and European Society of Human Reproduction and Embryology (ESHRE), recurrent spontaneous abortion (RSA) refers to at least two spontaneous abortions occurring before 20 weeks of gestation. It is a common human reproductive disorder

*Corresponding author.

affecting about 3% of couples of reproductive ages [1] [2] [3] [4] [5]. The risk of miscarriage in RSA patients is relatively high in re-pregnancy. Finding the potential cause of miscarriage is an efficient method to estimate the recurrence risk and take preventive management measures.

The etiology of RSA is extremely complex, with significant heterogeneity [6]. It includes reproductive tract anatomy, genetics, endocrinology, immunology, coagulation, masculinity, and a number of additional unknown causes. Scholars believe chromosomal abnormalities or genetic imbalances in embryos or fetuses are the major cause of abortion [7]. Genetic analysis of miscarriage and stillbirth tissue is of great value in analyzing the causes of miscarriage and stillbirth, assessing recurrence risk, and prenatal diagnosis. This paper will review the application of genetic testing techniques to the etiological diagnosis of RSA to serve as a reference for the selection of clinical diagnosis and treatment.

2. Genetic Testing Technology

2.1. G-Banding Chromosome Karyotype Analysis

G-banding chromosome karyotype analysis has been the most common method for diagnosing chromosomal abnormalities [8]. This method involves culture of cells or tissues under sterile conditions, and chromosome specimens are prepared by a series of operations such as trypsin digestion and Giemsa staining, and chromosome morphology can be observed under a microscope for karyotype analysis.

2.1.1. Karyotype Analysis of Couple's Peripheral Blood

Couples with chromosomal abnormalities, known as carriers, account for 2% - 5% of the RSA population [9]. In the study of peripheral karyotype analysis in RSA couples, the detected chromosomal abnormalities included abnormal chromosome number and structural abnormalities, of which structural abnormalities are the most common, including chromosomal equilibrium translocations, Robertsonian translocations, inversions, duplications, deletions [10] [11]. Chromosomally balanced translocations are highest in the carrier population [12], which are characterized by the exchange of DNA segments between non-homologous chromosomes, with no DNA gain or loss at breakpoints, and thus a balanced rearrangement; individuals with balanced translocations are usually phenotypically normal unless the translocation breakpoint disrupts a dominant gene or the exchange of chromosomal segments has an effect on the expression of nearby genes [13]. Carriers commonly seek medical attention after pregnancy loss, and chromosomal abnormalities are detected through peripheral blood karyotyping.

2.1.2. Products of Conception (POC) Karyotyping

The G-banded karyotype analysis of POC has been used by numerous scholars to study the causes of abortion. POC includes aborted villi tissue and amniotic fluid cells. Compared with peripheral blood, the karyotype analysis of POC requires higher culture conditions, and there is inevitable maternal material con-

tamination. Due to the need for *in vitro* culture, POC tissues or cells are required to maintain favorable biological activity. In the karyotype analysis of POC, the detected chromosomal abnormalities include aneuploidy, polyploidy, deletion, duplication, translocation, etc, among which aneuploidy is the most common [14] [15]. Aneuploidy results from the nondisjunction of homologous chromosomes during germ cell meiosis, resulting in an abnormal number of chromosomes in the zygote.

G-banding karyotype analysis can detect a variety of chromosomal abnormalities, which provides a great reference value for the diagnosis of RSA. However, there are myriad factors involved in the whole culture and analysis process, such as culture environment, colchicine concentration and action time, hypotonicity, fixation, drip, trypsin digestion, Giemsa staining, etc. Currently, there is no unified quality control for experimental manipulation of chromosome preparation. At the same time, this method has elevated requirements for laboratory technicians, and karyotype reading has a certain subjectivity. Due to the limitation of band resolution, conventional G-banding karyotyping can only identify the abnormal fragments larger than 5 Mb, and cannot judge the deletion, duplication and structural abnormality of minor fragments.

2.2. Chromosomal Microarray Analysis (CMA)

CMA, known as "molecular karyotype technology", is a high-throughput gene detection technology developed in recent years. For RSA patients, CMA plays a more prominent role in detecting pregnancy products. Compared to conventional karyotyping, CMA has the advantage of higher resolution, shorter detection time, and the absence of tissue cell culture. CMA can also detect clinically significant genomic copy number variants (CNVs). In addition to numerical chromosome abnormalities, newborn fetuses may also inherit CNVs or acquire absence of heterozygosity (LOH) from their parents' genomes, which may be harmful to embryonic or fetal development. Recurrent CNVs result from DNA nonallelic homologous recombination repair in regions with low copy repeat sequences [16]. Fetal CNVs may be inherent in the parental genome or may be newly formed in the parental gametes. LOH in the reproductive system may be generated by close marriage or by abnormal gene repair early in embryonic development [17].

As abortion tissue is often old samples, CMA provides a more suitable choice for RSA patients to find the cause of abortion.

2.2.1. Array Comparative Genomic Hybridization (aCGH)

ACGH is created in an ordered fashion using a small number of DNA fragments, with probe sizes ranging from a few tens to 2 hundred thousand base pairs, DNA samples were denatured, mixed DNA hybridized with probes in the array, and various fluorescence signals were emitted according to copy number amplification, gain, loss, or deletion to generate fluorescence intensity maps to identify CNVs present in the test DNA [18]. Clinicians regard CMA as an essen-

tial tool for screening and diagnosing genetic diseases. It has been found that about 1.6% - 1.8% of RSA cases reported clinically significant CNVs by aCGH. ACGH improved the detection efficiency and detected CNVs that could not be identified by conventional karyotype analysis [19]. Studies have shown that aCGH is effective in identifying common genetic aberrations, submicroscopic genomic rearrangements, and genes whose mutations cause miscarriage [15] [20] [21] [22]. However, aCGH probes cannot cover all chromosome segments, nor detect polyploidy and low-proportion mosaicism.

2.2.2. Single Nucleotide Polymorphism Array (SNP-Array)

SNP-array hybridizes the tested samples to a set of normal genomic controls [23]. Compared with aCGH, SNP-array has the advantage of detecting long extension homozygote, which can detect not only uniparental disomy (UPD) but also LOH. Arrays based exclusively on SNP probes are biased to include common certain genomic segments, and an early SNP detection array can only detect about 26% of the CNVs detected by the phosphor-terminal sequence mapping strategy; Newer SNP detection arrays also contain non-polymorphic oligonucleotide probes designed for copy number testing to provide more reliable and uniform coverage [24]. Overall, all current array platforms are capable of providing sufficient sensitivity for clinical CMA testing due to sufficiently dense probe coverage. CMA detects genomic imbalances in clinical settings with higher resolution and less subjectivity. Most current clinical CMA platforms can detect copy number changes in the whole genome with a lower limit of resolution of about 400 kb, which is more than 10-fold higher than G-banding karyotype analysis. This level of resolution will provide a broad genomic survey and reliably identify all known recurrent microdeletion and microduplication syndromes mediated by segmental repeat structures, as well as the majority of non-recurrent pathogenic imbalances that are clearly pathogenic. In the diagnosis of the etiology of RSA patients, CMA is more commonly used in the detection of pregnancy products. In a study of 5003 miscarriage samples, 309 genes were identified as potential miscarriage candidates, and three recurrent submicroscopic CNVs (22q11.21, 2q37.3, and 9p24.3p24.2 microdeletions) were found to be significantly more common in miscarriage cases [25]. Existing studies shown that in CMA detection of pregnancy products, the incidence of aneuploidy is the highest, followed by chromosomal structural abnormalities, triploidy [9]. However, while obtaining higher resolution, many CNVs of unknown clinical significance will also be detected, which brings certain difficulties to clinical consultation. According to the latest technical standards for CNVs interpretation and reporting, CNVs are divided into five categories: Pathogenic (P), Likely pathogenic (LP), Uncertain Significance (VUS), Likely benign (LB), Benign (B) [26]. The practicability of clinical consultation will be enhanced by a standard approach to the interpretation of CNVs applicable to all technology platforms and a widely accessible database of CNVs.

The limitation of CMA is that it cannot detect polyploidy, balanced transloca-

tions and low proportion mosaicism, and economic benefits are also part of the consideration due to the high chip cost.

2.3. Next-Generation Sequencing (NGS)

In recent years, with the rapid development of NGS, it has the advantage of more comprehensive genetic testing coverage and lower cost, and is widely used for disease diagnosis, especially prenatal diagnosis, providing additional options for the diagnosis of miscarriage in RSA patients.

2.3.1. Copy Number Variation Sequencing (CNV-Seq)

CNV-seq is the discovery of CNVs through bioinformatics analysis by sequencing samples and comparing the sequencing results with human reference genomes. It can detect CNVs of different sizes by adjusting the sequencing depth and changing the resolution. Numerous studies have reported the clinical use of CNV-seq to analyze the relationship between CNVs and miscarriage [25] [27] [28] [29] [30]. In a prospective chromosomal analysis of 3429 amniocentesis samples, the detection rate of pathogenic and potentially pathogenic CNVs increased from 1.8% to 2.8% using CNV-seq compared with karyotyping [31].

2.3.2. Whole Genome Sequencing (WGS)

WGS extracts DNA from the test sample, maps the sequenced reads to the reference genome and assigns them to a 20 kb sequencing box with a 5 kb slide to obtain higher resolution CNVs. In a study of 2186 pregnancy products with CNVs detected by WGS [32], chromosomal abnormalities were consistent with CMA; they found developmental genes that can be used to effectively identify pregnancy loss or congenital abnormal phenotypes. These genes were rich in genes related to embryonic development, especially neuronal development and differentiation.

2.3.3. Whole Exome Sequencing (WES)

WES is a gene detection technology for exon sequencing in protein coding regions, which is commonly used to detect the role of gene mutations in the mechanism of clinical diseases. In terms of prenatal diagnosis, if karyotype testing and CMA cannot determine the underlying cause of fetal malformations and structural abnormalities, WES can provide relevant information to aid in current pregnancy management. In the latest studies, many gene variants related to embryo abortion have been found by WES, suggesting that gene variants may be the potential etiology of RSA [33] [34] [35] [36] [37]. Future enriched WES results help to create a comprehensive database of genetic information containing mutations in genes that cause embryonic death, which will facilitate a broader understanding of the etiology of RSA and the development of strategies.

At present, NGS has been widely used in non-invasive prenatal screening, and it also plays an important role in the study of the etiology of RSA. Although NGS has many advantages in the detection of pregnancy products, its own limitations, such as the failure to detect UPD, balanced structural translocations and polyp-

loidy, should not be ignored. Due to the higher resolution of NGS, the interpretation of the results may increase the difficulty of genetic counseling.

2.4. Other Detection Techniques

In the process of using genetic testing technology to study the etiology of RSA, it is sometimes necessary to use some technical methods to support verification or technical exclusion of the detected abnormal results. Maternal cells may be involved in the collection of abortion tissue, which may affect the accuracy of the results. In addition, the next step of parental verification is required when chromosomal abnormalities are detected.

2.4.1. Fluorescent in Situ Hybridization (FISH)

FISH is a technique that uses a known nucleotide sequence labeled with fluorescence as a probe to hybridize with the target sequence in the chromosome to be tested, and then the fluorescence signal is observed under a fluorescence microscope to analyze the chromosome of the specimen to be tested. The detection efficiency of FISH is more dependent on the design of the probe and the corresponding detection. The chromosomal abnormalities of the abortion products detected by the above techniques can be verified by FISH for the parental chromosomes [38] [39], determining whether the mutation is new or inherited from the parents. The economic cost of using FISH probes is high because of the variation detected.

2.4.2. Quantitative Fluorescent Polymerase Chain Reaction (QF-PCR)

QF-PCR uses multiple pairs of fluorescent labeled primers for specific PCR amplification, and then the products are subjected to capillary electrophoresis, the chromosomal abnormalities are diagnosed according to the fluorescence signal intensity of the primers. In the detection of pregnancy products, QF-PCR is often used to exclude maternal cell contamination (MCC) in the sample to be tested [27] [40] [41]. If the specimen to be examined has a significant MCC, it should be excluded from the study.

3. Summary and Discussion

For pregnant couples, miscarriage can be a deeply distressing experience. Uncertainty about the cause of miscarriage and the concern that subsequent pregnancies may also fail gravely affect the physical and psychological well-being of RSA couples. Identifying the cause of a miscarriage can help predict the likelihood of continuing a pregnancy in the future and guide treatment. This is of great significance to couples who have experienced recurrent miscarriages.

This review discusses the application of chromosome karyotype analysis, CMA and NGS in RSA, and analyzes the advantages and limitations of each detection method by comparing the technical principle, type, distinguished and detection cost of different detection methods (**Table 1**), so as to provide certain guiding value for clinical selection of detection technology. Genetic testing is an

integral part of diagnosing the causes of abortion. A thorough understanding of the applicability of these testing methods and the selection of appropriate detection strategies will help to efficiently detect the etiology of RSA and provide more accurate information for subsequent genetic counseling. In addition, this paper provides a strategy for genetic testing of pregnancy products (**Figure 1**), and selects subsequent detection directions according to chromosome detection results to help determine whether chromosome abnormalities are original mutations or inherited from parents. Identification of specific diagnostic genetic variants can facilitate parental reproductive counseling and lead to improved management of future pregnancies by allowing prenatal or pre-implantation genetic diagnosis.

However, there are some limitations in this paper. Due to the rapid development of genetic testing techniques and the constant update of various testing methods, some of the contents of this paper may be partially biased and further literature reviews are needed to complement the relevant contents in the future.

Table 1. Advantages and limitations of various genetic testing methods.

	Aneuploid	Multiploid	Chimera	Microdeletion	Microdeletion, Microrepeat	Distinguishability	Cell culture	UPD	Cost	Others
G-banding chromosome karyotype analysis	√	√	√	√	×	Low	√	×	Low	Strong subjectivity
CMA	√	×	Low proportion of chimerism could not be detected	×	√	High	×	√	High	Uncertain significance CNVs
NGS	√	×	Low proportion of chimerism could not be detected	×	√	High	×	×	Relatively low	Uncertain significance CNVs

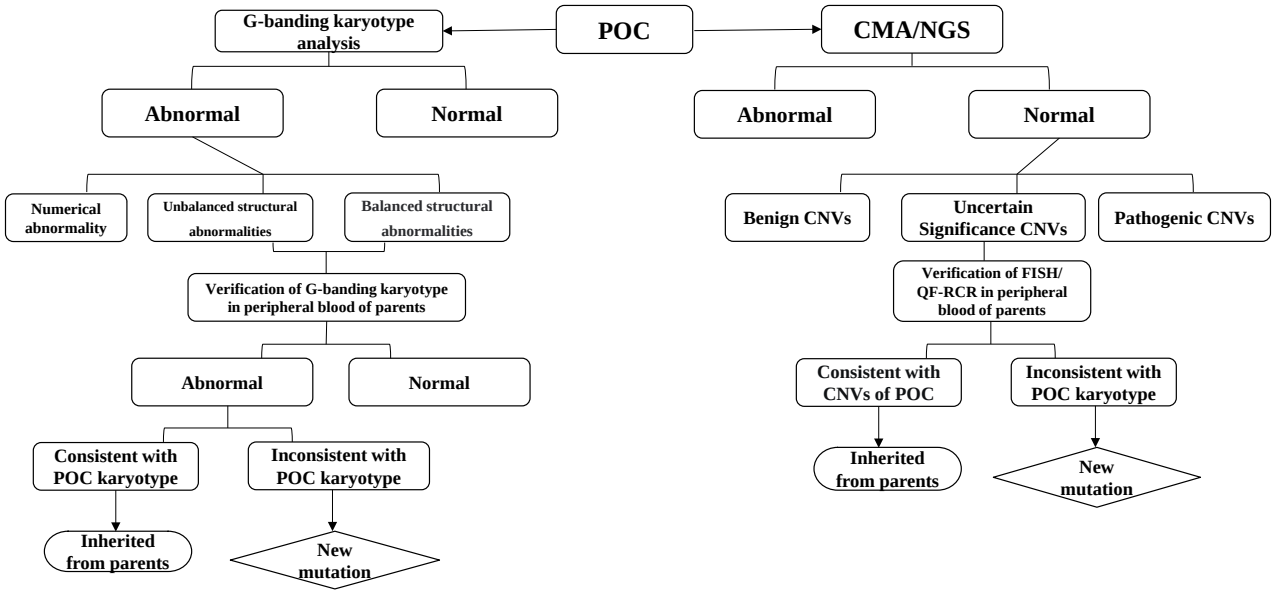

Figure 1. Strategies for genetic testing of pregnancy products.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

Funding

This research was funded by the Jingzhou science and technology development plan (key projects, 2015AC45, 2016AE51-2 and 2022CA48), Hubei Province health and family planning scientific research project (key projects, WJ2017Z024, WJ2018H175, WJ2019M085, WJ2019F126 and WJ2018H199), Hubei Provincial Natural Science Foundation of China (2018CFB775), and Fund of Hubei Clinical Medicine Research Center for individualized cancer diagnosis and therapy.

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Scientific Research Publishing

Yangtze Medicine, 2023, 7, 87-94
https://www.scirp.org/journal/ym
ISSN Online: 2475-7349
ISSN Print: 2475-7330

Application of Clinical Nursing Pathway in the Peri-Treatment Period of Immunoadsorption Therapy for Rheumatic Immune Diseases

Li Wang, Fan Fan, Yanrui Ren*

The First Affiliated Hospital of Yangtze University, The First People's Hospital of Jingzhou, Jingzhou, China
Email: *2245407657@qq.com

How to cite this paper: Wang, L., Fan, F. and Ren, Y.R. (2023) Application of Clinical Nursing Pathway in the Peri-Treatment Period of Immunoadsorption Therapy for Rheumatic Immune Diseases. *Yangtze Medicine*, **7**, 87-94.
https://doi.org/10.4236/ym.2023.72009

Received: February 27, 2023
Accepted: June 6, 2023
Published: June 9, 2023

Abstract

Objective: The paper aims to investigate the clinical nursing pathway (CNP) in the application of immunosorption therapy in patients with rheumatic immune disease. **Methods:** Convenience sampling method was used to select inpatients who received immunoadsorption therapy from January 2020 to December 2022 in the rheumatology and Immunology department of a 3A hospital in Jingzhou City. 30 patients from January 2020 to June 2021 were selected as control group, and 30 patients from July 2021 to December 2022 were selected as observation group. The control group was given routine nursing. On the basis of the control group, the observation group used a clinical nursing pathway for intervention during the perioperative period of immunosorbent therapy. The incidence of adverse reactions, patient satisfaction, and nurse satisfaction during immunosorbent therapy between the control group and the observation group were compared. **Results:** After intervention, the incidence of adverse reactions in the observation group was significantly lower than that in the control group, while patient satisfaction and nurse satisfaction in the observation group were significantly higher than those in the control group. The results are all statistically significant ($P < 0.05$). **Conclusion:** Clinical nursing pathway is beneficial to reduce the incidence of adverse reactions in patients with immunoadsorption during peri-treatment and improve the satisfaction of patients and nurses.

Keywords

Immunoadsorption, Clinical Nursing Pathway, Rheumatic Immune Disease,

Peri-Treatment Period

1. Introduction

Rheumatic immune disease is a common autoimmune disease, the main cause of which is the abnormal immune response of the body to normal tissues, resulting in excessive production of autoantibodies, which will attack the body's own cells, tissues and organs, cause inflammation, and eventually cause damage to the body [1]. At present, the treatment of rheumatism mainly uses hormones and immunosuppressants, but the above drugs have great side effects and take a long time to take effect, which has limitations for critical patients. In recent years, a new therapy to remove specific autoimmune components in the immune system: immunoadsorption (IA) has become a hot research direction in the treatment of autoimmune diseases [2]. It is a more targeted approach to the treatment of rheumatic immune diseases.

Immunoadsorption is a blood purification technology that combines highly specific antigens, antibodies or substances with specific physical and chemical affinity (ligand) and adsorbent materials (carrier) into adsorbent (column), selectively or specifically removes pathogenic immune complex or autoantibody from plasma to improve patients' clinical symptoms, so as to alleviate the disease.

Immunoadsorption therapy is to extract the patient's blood through peripheral or central vein, add anticoagulant into the blood, and then go through the filter to separate the plasma and blood cells in the blood, plasma pump to separate the plasma to the adsorbent [3], through the adsorption column with high-affinity substances to remove pathogenic factors, and finally the purified plasma and red blood cell components back to the patient.

Peri-treatment nursing of immunoadsorption is very important. Studies have shown that standardized care can reduce peri-treatment complications, such as bleeding tendency, allergic reactions, and pipe blockage. Clinical nursing pathway (CNP) is to optimize the nursing workflow and form a standardized nursing process under the premise of patient-centered, so as to guide nursing work in a more predictable, specific, standardized and active way. At the same time, it also enables patients to clarify their nursing goals [4], so that they can consciously participate in the whole process of disease management.

In clinical nursing work, peri-treatment nursing content of immunoadsorption is complicated and tedious. In order to further improve the standardization of treatment, reduce adverse reactions as far as possible, and improve nurses' work efficiency and patients' satisfaction; in this study, clinical nursing pathway method was used to intervene in patients receiving immunoadsorption therapy in the rheumatology and immunology department of a 3A hospital in Jingzhou City from January 2021 to December 2022, and the effect was good. The report is as follows.

2. Object and Method

2.1. Research Object

Patients who underwent immunosorption therapy from January 2020 to December 2022 in a Grade-A hospital in Jingzhou City were selected as subjects by convenience sampling method. Inclusion criteria: 1) older than 18 years; 2) Patients clearly diagnosed with rheumatic immune disease and clinically assessed by doctors to improve their condition by immunoadsorption; 3) Informed consent can cooperate with the completion of this study. Exclusion criteria: 1) patients with consciousness disorder; 2) Patients with severe infection or cardiovascular and cerebrovascular diseases. According to the above criteria, 30 patients from January 2020 to June 2021 were included as the control group, including 6 males and 24 females, aged from 19 to 63 years old, with an average of (32.5 ± 6.2) years old and a course of disease of 3 - 180 months. From July 2021 to December 2022, 30 patients were included in the observation group, including 4 males and 26 females, ranging in age from 21 to 60 years, with an average age of (31.6 ± 7.4) years. The course of disease lasted 4 - 178 months. There were no significant differences in age, gender, course of disease and other general data between the two groups ($P > 0.05$), indicating comparability. The patient has signed an informed consent form.

2.2. Methods

2.2.1. Control Group

All adopted DNA280 adsorption column or DNA230 adsorption column produced by Zhuhai Jianfan Biotechnology Company. Before immunoadsorption therapy, all patients signed informed consent after fully understanding the pros and cons of the therapy. The control group was carried out according to the nursing routine of immunoadsorption therapy in the department. The specialist nurses cooperated with the doctors to carry out deep venocentesis (generally selecting the internal jugular vein or femoral vein) with dual cavity catheterization. The whole course lasted for 5 days, and the adsorption therapy was performed 3 times, respectively on the 1st, 2nd and 4th day of catheterization, and the treatment time was generally 2 - 2.5 hours. The whole treatment process was monitored by a specialist nurse bedside, and the patient underwent ECG monitoring and oxygen inhalation. Specialist nurses carried out routine nursing education in the course of treatment, including the significance and process of immunosorption therapy, postoperative precautions, etc.

2.2.2. Observation Group

Observation group was on the basis of control group to take clinical pathway method intervention. Our department has set up a special working group, strictly following the principles of evidence-based medicine, sorting out, summarizing and summarizing the contents of immunoadsorption peri-treatment, and then using clinical nursing pathway methods, from the four aspects of "preparation and education before treatment", "disease monitoring during treatment", "ob-

servation and health guidance of complications after treatment", "guidance after extubation" to form a standardized, organized clinical path table.

The details are as follows: first, in the "pre-treatment preparation and education" aspect, from the environmental preparation, patient preparation two aspects to strengthen the management, in the environmental preparation is equipped with a single room ward, patients before and after treatment for ultraviolet disinfection; Patient preparation measures include responsible nurses to educate patients on the preparation of materials. Before treatment, the preparation list of materials for patients is distributed, including bedpans, urinals (male patients), toilets, disposable sheets, and two sets of cotton loose clothes. Perineal skin preparation and relevant laboratory examination are performed the day before treatment. The responsible nurse taught the process of immunosorption therapy through video, explained the key points of cooperation with patients, and conducted psychological counseling. Second, in terms of "condition monitoring during treatment", specialized nurses perform ECG monitoring and oxygen therapy for patients, connect perfusion machine for patients after detecting vascular access, monitor patients' breathing and pulse in real time before and after using the machine, monitor body temperature and blood pressure every half hour, and gradually increase blood flow to 200 - 250 ml/min according to patients' conditions [5]. Closely observe whether the patient has palpitation, chest tightness, blood pressure drop and other adverse reactions, at the same time pay attention to the function of the catheter, real-time adjustment of the direction of the catheter, to avoid the blockage of blood. During the whole treatment process, personalized anticoagulant therapy was carried out in strict accordance with the doctor's advice. Third, in the aspect of "observation and health guidance of complications after treatment", responsible nurses should take over every shift to observe whether the puncture site has oozed blood, and conduct standardized catheter maintenance operations to avoid tube blockage and infection. In the aspect of health guidance, it focuses on guiding the care of patients' catheters, guiding patients to reasonably relieve themselves, avoiding bending and discounting of pipes, and preventing tube removal. In addition, patients were instructed to prevent hypoglycemia and falls. The main measures included timely eating breakfast, activity guidance, and prevention of deep vein thrombosis. Fourthly, in the aspect of "post-extubation guidance", emphasis should be placed on guiding patients to immobilization of operative limbs, keeping the dressing at the puncture site clean and dry, and avoiding bleeding or infection caused by improper nursing after extubation.

2.3. Evaluation Indicators

1) Incidence of adverse reactions includes bleeding tendency, allergic reaction (fever, chest tightness, etc.), pipeline blockage, hypoglycemia, decreased blood pressure, fall of patients, etc. 2) Patient satisfaction and nurse satisfaction: self-designed satisfaction questionnaire was adopted, which was investigated and recovered within 6 hours after extubation of patients and divided into three levels: satisfaction, general and dissatisfaction.

2.4. Statistical Method

SPSS22.0 statistical software is used for data processing and analysis. The measurement data is expressed by mean ± standard deviation ($\overline{X} \pm s$), and independent sample t-test is used for inter-group comparison. The counting data were expressed in frequency and percentage (%), and the chi-square test was used for comparison between groups. The difference was statistically significant with $P < 0.05$.

3. Results

3.1. Incidence of Peritherapeutic Adverse Reactions in 2 Groups

In the observation group, one case of adverse reaction occurred, which was bleeding at puncture after extubation: The patient did not immobilization the affected limb after extubation according to the doctor's advice, and was eager to get out of bed and bleed at the puncture site, but did not bleed after being pressed again. A total of 7 adverse reactions occurred in the control group, including hemorrhage after extubation in 2 cases, catheter dysfunction in 2 cases, hypoglycemia in 1 case, fall in 1 case, and decrease in blood pressure in 1 case. The above adverse reactions were actively treated, and none of them caused damage to patients. There was statistical significance in the incidence of adverse reactions in peri-treatment between the two groups ($P < 0.05$) (see Table 1).

3.2. Comparison of Treatment Satisfaction between the Two Groups

In terms of satisfaction of patients and nurses, the satisfaction of the observation group was significantly higher than that of the control group, the difference was statistically significant ($P < 0.05$) (see Table 2 and Table 3).

Table 1. Comparison of the incidence of adverse reactions in the two groups during the treatment period [n(%)].

Group	n	Adverse reactions
Control group	30	7 (23.3)
Observation group	30	1 (3.33)
χ^2		4.57
P		<0.05

Table 2. Comparison of patient satisfaction between the two groups [n(%)].

Group	n	Patient satisfaction		
		Satisfied	Generally satisfied	Dissatisfaction
Control group	30	20 (66.67)	9 (30)	1 (3.33)
Observation group	30	28 (93.33)	2 (6.67)	0
χ^2			5.77	
P			<0.05	

Table 3. Comparison of nurse satisfaction between the two groups [n(%)].

Group	n	Nurse satisfaction		
		Satisfied	Generally satisfied	Dissatisfaction
Control group	14	8 (57.14)	6 (42.86)	0
Observation group	14	13 (92.86)	1 (7.14)	0
χ^2			5.56	
P			<0.05	

4. Discussion

In recent years, with the deterioration of living environment, the increase of social pressure and the change of diet structure, the incidence of rheumatic immune diseases has shown an obvious trend of rise, and has gradually become a common and frequently occurring clinical disease [6]. The treatment of immune system diseases is complex, incurable and highly personalized, and how to effectively treat them is a hot issue of clinical concern. Immunoadsorption therapy is a new clinical treatment of rheumatic immune diseases [7]. Through extracorporeal circulation and the use of antigen-antibody immune response, it can effectively remove related pathogenic factors in the body, which is an effective method of blood purification. Immunoadsorption therapy can provide a good treatment window for reducing disease activity. For rheumatic diseases with poor treatment effect with drugs alone or in critical and severe condition, Immunoadsorption therapy can rapidly relieve the clinical symptoms and improve the condition of patients [8].

However, unlike conventional treatments, the treatment requires deep vein catheterization, using extracorporeal circulation to remove disease-causing factors from the blood and then transfusing it back into the patient through an adsorbent device. For patients, due to the lack of understanding, acceptance and compliance of this new and invasive treatment, many patients have different degrees of psychological disorders, lack of confidence in treatment; will appear excessive tension, fear, anxiety and other adverse emotions. In this study, based on the advanced method of clinical nursing pathway (CNP), on the basis of patient-centered, the nursing work content of immunoadsorption peri-treatment is integrated and optimized, forming a standardized nursing process, which is more guiding for nurses' work [9]. At the same time, it also enables patients to have a clear understanding of their own preparation and coordination work, so that they can actively participate in the entire process of disease management [10]. Experienced specialist nurses focus on the two dimensions of patient education and condition observation in the three aspects of treatment before, during and after treatment. Through one-to-one nursing, personalized understanding of patients' concerns can effectively relieve patients' bad mood and improve patients' treatment compliance and satisfaction.

In addition, Immunoadsorption therapy is based on ideal vascular access to

ensure adequate blood flow. Therefore, it is necessary to actively maintain the pipeline during treatment, disinfect and fix the pipeline, seal the tube correctly, and actively prevent the occurrence of thrombus in the catheter. In this study, by establishing standardized operation procedures of immunoadsorption, the focus of disease observation and nursing is clarified, especially for the maintenance of deep vein catheterization, which effectively reduces the occurrence of various complications during peri-treatment. At the same time, the process and standardized form of clinical pathway can make the complex work content in the peri-treatment period organized and standardized [11], so that nurses have a clear idea of the work content and focus, so as to improve the work efficiency and satisfaction of nurses.

5. Conclusion

Through this study, it was found that clinical nursing pathway can effectively reduce various complications and improve the satisfaction of patients and nurses during the peri-treatment period of immunoadsorption for rheumatic immune diseases, which is worthy of clinical application. However, the clinical nursing pathway developed at this stage is not fixed. In future work, we should enrich and improve on the basis of evidence-based and clinical practice, and should focus on the needs of patients, in order to better provide comprehensive and high-quality services for patients with immunosorbent therapy. However, the shortcomings of this study are that it is only conducted in a single medical institution, lacks a multi-center sample size, and lacks evaluation indicators related to the patient's disease activity in the patient outcome indicators, which needs further research.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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Scientific Research Publishing

Yangtze Medicine, 2023, 7, 95-104
https://www.scirp.org/journal/ym
ISSN Online: 2475-7349
ISSN Print: 2475-7330

Progress in the Study of Vonoprazan Fumarate vs. Proton Pump Inhibitors in the Treatment of Gastroesophageal Reflux Disease

Yuanmei Dai[1*], Baofeng Liu[1], Xiaohui Shen[1], Lei Huang[2*]

[1]The First Clinical College, Changzhi Medical College, Changzhi, China
[2]Department of Endoscopy, Heji Hospital, Changzhi Medical College, Changzhi, China
Email: *dym17855356056@163.com, *dr.huanglei@qq.com

How to cite this paper: Dai, Y.M., Liu, B.F., Shen, X.H. and Huang, L. (2023) Progress in the Study of Vonoprazan Fumarate vs. Proton Pump Inhibitors in the Treatment of Gastroesophageal Reflux Disease. *Yangtze Medicine*, **7**, 95-104.
https://doi.org/10.4236/ym.2023.72010

Received: April 5, 2023
Accepted: June 6, 2023
Published: June 9, 2023

Abstract

Gastroesophageal reflux disease (GERD) is a common gastrointestinal disease, and proton pump inhibitors (PPIs) have been recommended as the first-line treatment for GERD. In recent years, studies on vonoprazan fumarate in the treatment of GERD have attracted widespread attention. In this paper, we review the research progress of vonoprazan fumarate and proton pump inhibitors in the treatment of GERD in recent years, and compare and analyze the efficacy, safety, tolerability, and advantages and disadvantages of long-term application of both. By reviewing the relevant literature, we found that vonoprazan fumarate has similar performance with proton pump inhibitors in terms of efficacy and safety, but has potential advantages in terms of tolerability and long-term application. Therefore, we believe that vonoprazan fumarate may become a new option for GERD treatment, helping clinicians to develop more appropriate treatment plans for patients and providing new ideas and directions for research in related fields.

Keywords

Vonoprazan Fumarate, Proton Pump Inhibitors, Gastroesophageal Reflux Disease, Efficacy, Safety, Tolerability, Long-Term Application

1. Introduction

1.1. Background

Gastroesophageal reflux disease (GERD) is a common digestive system disease, mainly manifested as gastric acid reflux into the esophagus, causing symptoms such as heartburn, cough, and chest pain [1]. Long-term gastric acid stimulation can cause esophageal mucosal damage, and even progress to Barrett's esophagus

and esophageal cancer [2]. In recent years, with the improvement of living standard and the change of diet structure, the incidence of GERD is increasing year by year and has become a worldwide public health problem [3]. The main drugs for treating gastroesophageal reflux disease include proton pump inhibitors (PPIs) and histamine H2 receptor antagonists (H2RAs) [4]. Among them, proton pump inhibitors are the most commonly used treatment drugs, which can effectively inhibit gastric acid secretion and relieve symptoms [5]. However, long-term use of proton pump inhibitors may cause some adverse reactions, such as malnutrition, intestinal infection and fractures, [6] so finding safe and effective alternative drugs is of great significance. Vonoprazan fumarate is a novel potassium-competitive acid blocker (P-CAB), which has been marketed in countries such as Japan and China for the treatment of gastroesophageal reflux disease, peptic ulcers, etc. [7]. Compared with proton pump inhibitors, vonoprazan fumarate has a faster onset of action, stronger acid suppression, and a longer duration of action [8]. Therefore, this article aims to review the research progress of vonoprazan fumarate and proton pump inhibitors in the treatment of gastroesophageal reflux disease, and compare their efficacy, safety, tolerability, and the advantages and disadvantages of long-term application [9].

1.2. Purpose

The purpose of this article is to provide more options for the treatment of gastroesophageal reflux disease by comparing and analyzing the research progress of vonoprazan fumarate and proton pump inhibitors in the treatment of gastroesophageal reflux disease, to help clinicians develop more suitable treatment plans for patients, and to provide new ideas and directions for research in related fields.

1.3. Methods

This article searched databases such as PubMed, Web of Science and CNKI to collect recent research on vonoprazan fumarate and proton pump inhibitors in the treatment of gastroesophageal reflux disease, including clinical trials, retrospective research, meta-analyses, and systematic reviews. The search keywords included: “vonoprazan fumarate”, “proton pump inhibitors”, “gastroesophageal reflux disease”, “efficacy”, “safety”, “tolerability”, “long-term application”, etc. During the screening process, priority was given to selecting recent and high-quality research. By comparing and analyzing the results of different studies, the advantages and disadvantages of vonoprazan fumarate and proton pump inhibitors in the treatment of gastroesophageal reflux disease, including efficacy, safety, tolerability, and long-term application, were summarized.

2. Mechanisms of Action of Vonoprazan Fumarate and Proton Pump Inhibitors

2.1. Mechanism of Action of Vonoprazan Fumarate

Vonoprazan fumarate is a potassium-competitive acid blocker (P-CAB), and its

mechanism of action is different from that of proton pump inhibitors. Vonoprazan mainly works by competitively binding to the H+/K+-ATPase on gastric acid-secreting cells, preventing hydrogen ions from entering the gastric cavity, thereby inhibiting gastric acid secretion [7]. Due to vonoprazan's competitive binding to H+/K+-ATPase, it has a strong effect in inhibiting gastric acid secretion and acts faster [8]. Vonoprazan also has some advantages, such as not being affected by the physiological rhythm of gastric acid secretion, so it can be administered at any time, without being limited to before or after meals [10]. In addition, due to the longer half-life of vonoprazan, its efficacy can last for a period of time, helping to reduce the daily dosage [8].

2.2. Mechanism of Action of Proton Pump Inhibitors

Proton pump inhibitors (PPIs) are a class of drugs widely used to treat gastroesophageal reflux disease and other gastric acid hypersecretion-related diseases. Proton pump inhibitors work by inhibiting the H+/K+-ATPase (proton pump) on gastric acid-secreting cells to reduce gastric acid production [4]. Proton pump inhibitors form a covalent bond with the H+/K+-ATPase, causing it to lose function, thus reducing gastric acid secretion [11]. Proton pump inhibitors mainly include omeprazole, lansoprazole, pantoprazole, esomeprazole, etc. These drugs have similar mechanisms of action in inhibiting gastric acid secretion, but have differences in pharmacodynamics and pharmacokinetics [12]. The efficacy of proton pump inhibitors is closely related to the timing of administration, usually taken before meals, in order to achieve the best effect during the peak gastric acid secretion [4]. Although proton pump inhibitors have significant efficacy in the treatment of gastroesophageal reflux disease, long-term use may increase the risk of certain side effects, such as kidney damage, fractures, and malnutrition [13].

3. Comparison of the Efficacy of Vonoprazan Fumarate and Proton Pump Inhibitors in the Treatment of Gastroesophageal Reflux Disease

3.1. Efficacy Evaluation Criteria

To compare the efficacy of vonoprazan fumarate and proton pump inhibitors in the treatment of gastroesophageal reflux disease, certain evaluation criteria must be applied. Efficacy evaluations usually include improvement in clinical symptoms, healing of gastroesophageal mucosal injury, and the degree of gastric acid secretion inhibition [14] [15] [16] [17].

3.2. Research Review

In recent years, several studies have compared the efficacy of vonoprazan fumarate and proton pump inhibitors in the treatment of gastroesophageal reflux disease. A randomized controlled trial (RCT) conducted by Ashida K *et al.* [18] compared the efficacy of vonoprazan fumarate and omeprazole in patients with

esophagitis. The study results showed that the symptom relief rate and mucosal healing rate in the vonoprazan fumarate group were higher than those in the omeprazole group, suggesting that vonoprazan fumarate may have superior efficacy in the treatment of gastroesophageal reflux disease. A study by Miyazaki H *et al.* [19] found that compared with lansoprazole, vonoprazan had better effects in inhibiting gastric acid secretion, improving clinical symptoms, and promoting gastroesophageal mucosal healing. This study indicates that vonoprazan may have higher efficacy in the treatment of gastroesophageal reflux disease. A systematic review and meta-analysis by Li M *et al.* [20] showed that, compared with proton pump inhibitors, vonoprazan fumarate had faster symptom improvement and higher mucosal healing rates in the treatment of gastroesophageal reflux disease. A study by Ma Yuan *et al.* [21] found that vonoprazan fumarate tablets had significant clinical efficacy in treating recurrent reflux esophagitis, significantly improved 24-hour esophageal pH, reduced inflammation factor levels, and did not increase the incidence of adverse drug reactions. A study by Zheng Yanhe *et al.* [22] showed that compared with lansoprazole, vonoprazan fumarate was more effective in promoting the healing of damaged esophageal mucosa, improving clinical symptoms, increasing MTL and GAS levels, and showing more significant clinical efficacy in the treatment of RRE.

In summary, current research generally believes that vonoprazan fumarate has superior efficacy to proton pump inhibitors in the treatment of gastroesophageal reflux disease. However, further large-sample, multicenter randomized controlled trials are still needed to verify the stability and reliability of this conclusion.

4. Comparison of Vonoprazan Fumarate and Proton Pump Inhibitors in Terms of Safety

Comparative studies on the safety of vonoprazan fumarate and proton pump inhibitors are insufficient. In a randomized controlled trial by Jenkins H *et al.* [8], the safety of vonoprazan fumarate and omeprazole in treating patients with gastroesophageal reflux disease was investigated. The results showed that there was no significant difference in the incidence of adverse events during treatment between the two groups, suggesting that vonoprazan fumarate has comparable safety to omeprazole. An open-label, randomized crossover trial by Sakurai Y *et al.* [7] investigated the safety of vonoprazan fumarate and proton pump inhibitors (esomeprazole and rabeprazole) in healthy adults. The study results showed that there was no significant difference in the incidence and severity of adverse events between vonoprazan fumarate and proton pump inhibitors. A study by Zheng *et al.* [23] found that the combination of vonoprazan fumarate tablets and lansoprazole in treating patients with gastroesophageal reflux-related pharyngolaryngeal disease showed superior clinical efficacy and good safety, making it worth promoting. A randomized, double-blind, double-dummy, parallel-controlled trial by Ashida K *et al.* [18] compared the side effects of vonoprazan fumarate

and omeprazole in treating patients with acute gastritis. The study results showed that there was no significant difference in the incidence of adverse reactions during treatment between the two groups. Furthermore, there was no significant difference in the incidence of severe adverse reactions between the two groups, suggesting that vonoprazan fumarate and omeprazole have similar safety in terms of side effects. A retrospective study by Miyazaki H *et al.* [19] compared the side effects of vonoprazan fumarate and proton pump inhibitors (lansoprazole) in treating patients with gastroesophageal reflux disease. The results showed that there was no significant difference in the incidence of adverse events during treatment between the two. However, this was a retrospective study, which may be subject to bias. More prospective studies are needed in the future to confirm this conclusion.

These research results indicate that the safety of vonoprazan fumarate in the treatment of gastroesophageal reflux disease is comparable to proton pump inhibitors. However, these studies have small sample sizes and are mostly short-term trials. To more comprehensively evaluate the differences in safety between the two, more large-scale, long-term studies are needed in the future.

5. Comparison of Tolerability between Vonoprazan Fumarate and Proton Pump Inhibitors

Tolerability is an important indicator for evaluating drug safety, which can help determine the possible adverse reactions patients may encounter during drug use.

5.1. Tolerability of Vonoprazan Fumarate

Vonoprazan fumarate, as a new potassium-competitive acid blocker, shows good characteristics in terms of tolerability. A randomized controlled trial by Jenkins H *et al.* [8] investigated the safety, tolerability, pharmacokinetics, and pharmacodynamics of vonoprazan fumarate in healthy male subjects. The study results showed that the incidence of adverse events during the treatment with vonoprazan fumarate was low, and most adverse events were mild to moderate, indicating good tolerability of vonoprazan fumarate. An open-label, randomized crossover trial by Sakurai Y *et al.* [7] evaluated the tolerability of vonoprazan fumarate versus esomeprazole and rabeprazole in healthy adults. These studies indicate that vonoprazan fumarate has a good performance in terms of tolerability.

5.2. Tolerability of Proton Pump Inhibitors

Proton pump inhibitors (PPIs), as commonly used gastric acid suppressants, have a good performance in tolerability. A randomized, double-blind, controlled trial by Kahrilas PJ *et al.* [24] evaluated the tolerability of omeprazole versus lansoprazole in patients with gastroesophageal reflux disease. The study results showed that there was no significant difference in the incidence of adverse events during treatment between the two, and both were low, suggesting good

tolerability of proton pump inhibitors. A randomized, double-blind, parallel-group, multicenter study by Röhss K *et al.* [25] compared the tolerability of esomeprazole and rabeprazole in patients with gastric ulcers. The results showed that there was no significant difference in the incidence of adverse events during treatment between the two, and most adverse events were mild to moderate, indicating good tolerability of proton pump inhibitors. In summary, proton pump inhibitors have a good performance in tolerability.

5.3. Tolerability Comparison

To gain a more comprehensive understanding of the differences in tolerability between vonoprazan fumarate and proton pump inhibitors, a randomized, double-blind, controlled trial by Ashida K *et al.* [18] compared the tolerability of vonoprazan fumarate and rabeprazole in patients with gastroesophageal reflux disease. The study results showed that the incidence of adverse events during treatment with vonoprazan fumarate was similar to that of rabeprazole, and most adverse events were mild to moderate, suggesting that the two have comparable tolerability. A randomized, open-label, controlled trial by Matsukawa J *et al.* [26] evaluated the tolerability of vonoprazan fumarate and esomeprazole in patients with chronic gastritis. The study results showed that the incidence of adverse events during treatment with vonoprazan fumarate was slightly lower than that of esomeprazole, but the difference was not statistically significant, indicating that the two have similar tolerability. A randomized, double-blind, controlled trial by Jenkins H *et al.* [8] assessed the tolerability of vonoprazan fumarate and omeprazole in patients with gastroesophageal reflux disease. The study results showed that there was no significant difference in the incidence of adverse events during treatment between the two, and both were low. Most adverse events were mild to moderate, suggesting that vonoprazan fumarate and omeprazole have comparable tolerability. A randomized, double-blind, multicenter trial by Xiao Y *et al.* [27] compared the tolerability of vonoprazan fumarate and lansoprazole in patients with gastric and duodenal ulcers. The study results showed that the incidence of adverse events during treatment with vonoprazan fumarate was similar to that of lansoprazole, and most adverse events were mild to moderate. These results further confirmed the comparability of the two in terms of tolerability.

In summary, the comparison of tolerability between vonoprazan fumarate and proton pump inhibitors indicates that both have similar incidences and severity of adverse events during treatment. However, due to the limited current research, more large-scale, long-term studies are needed in the future to comprehensively assess the differences in tolerability between the two.

6. Analysis of Advantages and Disadvantages of Long-Term Use

This chapter will analyze the advantages and disadvantages of vonoprazan fumarate and proton pump inhibitors in long-term use.

6.1. Advantages and Disadvantages of Vonoprazan Fumarate

Advantages: As previously mentioned, many studies have shown that vonoprazan fumarate is well tolerated in long-term applications [18] [26]. According to a study by Tanabe T *et al.* [28], vonoprazan fumarate showed good efficacy in long-term use, which looked at the improvement of symptoms in patients with GERD after 12 months of treatment with vonoprazan fumarate and showed significant improvement in symptoms.

Disadvantages: The safety and side effects of long-term use are not yet supported by sufficient evidence.

6.2. Advantages and Disadvantages of Proton Pump Inhibitors

Advantages: Proton pump inhibitors have been widely used in the clinic for many years, and several studies have shown that proton pump inhibitors have good efficacy in the long-term treatment of GERD [29] [30], and their long-term use has been widely validated for safety and efficacy [31].

Disadvantages: Long-term use of proton pump inhibitors may lead to some potential side effects, such as increased risk of fracture [32], increased risk of renal injury [33], and nutrient malabsorption [34], and some patients may develop drug tolerance leading to reduced efficacy after long-term use of proton pump inhibitors [4].

7. Conclusion and Prospects

Through a review of existing research, we found that both vonoprazan fumarate and proton pump inhibitors have good efficacy and high safety. However, there are differences in their mechanisms of action, side effect profiles, and indications. In actual clinical practice, doctors need to develop personalized treatment plans based on the patient's specific condition, drug response, and potential risks to improve treatment outcomes and reduce side effects. Furthermore, future research should continue to focus on the mechanisms of action, efficacy, safety, and patient compliance of vonoprazan fumarate and proton pump inhibitors, and conduct more high-quality randomized controlled trials to provide more robust evidence for clinical practice. In addition, drug research for specific patient populations and the development of new drugs are important directions for future research.

In conclusion, by comparing vonoprazan fumarate and proton pump inhibitors, we have provided valuable insights for the treatment of gastroesophageal reflux disease and pointed out directions for future research and clinical practice. We hope this review will bring more inspiration to the diagnosis and treatment of gastroesophageal reflux disease.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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Yangtze Medicine, 2023, 7, 105-115
https://www.scirp.org/journal/ym
ISSN Online: 2475-7349
ISSN Print: 2475-7330

Establishment of Risk Prediction Model and Nomogram for Lymph Node Metastasis of Cervical Cancer: Based on SEER Database

Sufei Wang*, Shiwei Li*, Yong Chen#, Ya Zhang†

Obstetrics and Gynecology, The First Clinical Medical College of Yangtze University, Jingzhou First People's Hospital, Jingzhou, China
Email: †43547235@qq.com

How to cite this paper: Wang, S.F., Li, S.W., Chen, Y. and Zhang, Y. (2023) Establishment of Risk Prediction Model and Nomogram for Lymph Node Metastasis of Cervical Cancer: Based on SEER Database. *Yangtze Medicine*, **7**, 105-115.
https://doi.org/10.4236/ym.2023.72011

Received: April 23, 2023
Accepted: June 26, 2023
Published: June 29, 2023

Abstract

Objective: To predict the risk factors of lymph node metastasis in cervical cancer by using large sample clinical data, and to construct and verify the nomogram for predicting lymph node metastasis. Methods: A total of 5940 patients with cervical cancer from 2004 to 2015 in the National Cancer Institute Surveillance Epidemiology and End Results database were retrospectively screened and randomly assigned to training group (n = 4172) and validation group (n = 1768). Multivariate Logistic regression analysis was used, and the optimal model was selected according to AIC or BIC and likelihood ratio test, and a nomogram was drawn. The accuracy and robustness of the prediction model were evaluated in three aspects: discrimination, calibration and clinical net benefit. Results: The prediction model based on race, tumor tissue differentiation degree, tumor histopathological type, distant metastasis of tumor, tumor diameter and other risk factors was successfully established and a nomogram was constructed. The AUCs of training group and validation group were: 0.736 and 0.714, respectively. And the p-values of the Hosmer-Lemeshow test were 0.28 and 0.11, respectively. The calibration curve was in good agreement with the ideal curve. It had high accuracy and applicability after internal verification. Conclusion: A prediction model is constructed based on the risk factors of lymph node metastasis of cervical cancer. The nomogram has a good effective prediction and can provide a theoretical basis for clinicians to assess the disease quickly before surgery.

Keywords

Cervical Cancer Lymph Node Metastasis, SEER Database, Logistic Regression, Nomogram

*First author.
#Second author.
†Corresponding author.

1. Introduction

A common malignant tumor, cervical cancer is also the second malignant tumor of the female reproductive system in Chinese women [1]. Cervical cancer is the fourth malignant tumor with the highest morbidity and mortality worldwide. At present, the treatment of cervical cancer is mainly operation and radiotherapy, while surgery is the main comprehensive treatment for early-stage cervical cancer. And individualized treatment plan can be made according to the relevant risk factors after operation [2]. The main high-risk factors for postoperative recurrence of cervical cancer include positive lymph nodes, para-uterine infiltration, positive resection margins and so on. Lymph node metastasis can occur in the early-stage cervical cancer, the incidence is about 10% - 20%, and once lymphatic metastasis, the 5-year survival rate of patients can be reduced by 20% - 45% [3]. Therefore, lymph node metastasis is an important factor affecting the treatment and prognosis of patients.

In recent years, nomogram is an important tool to evaluate and guide treatment in clinical prediction models, and it is widely used in clinical practice. Nomogram can be constructed by predicting high-risk factors, which can visualize and quantify the occurrence and prognosis of various diseases. At present, there is no risk assessment method for lymph node metastasis of cervical cancer at home and abroad. The purpose of this study is to retrieve a large number of cervical cancer data from SEER database, to explore the related influencing factors of cervical cancer patients with lymph node metastasis, and to establish a prediction model to provide an effective predictive tool for identifying high-risk patients, further scientific guidance for follow-up treatment, and strict grasp of surgical indications, so as to propose individualized and more optimized treatment plans.

2. Objects and Methods

2.1. Research Object

The data of 5941 cases included in this study were extracted from the SEER database (https://seer.cancer.gov/) using SEER*STATA software, which stores cancer surveillance data from various parts of the United States, covers approximately 28% of the population of the United States, and provides data for population studies of various cancers [4]. The ICD code C73.9 was used to screen the case data of uterine cancer patients whose pathological code was 8510 from 2004 to 2015. Due to the openness of the SEER database, this study does not require ethical approval or informed consent. Inclusion criteria: The pathological diagnosis was cervical cancer and had complete pathological characteristic data. Exclusion criteria: complicated with other types of malignant tumors. The clinicopathological data are incomplete or unknown.

The pathological features included in this study include: ID, age, race, marital status, primary site of tumor, tumor diameter, tumor tissue differentiation degree, tumor histopathological type, T stage, N stage, M stage, SEER historic stage

A, AJCC Stage Group. The TNM staging of the patients was based on the American Joint Commission on Cancer (AJCC) tumor staging, 6th edition.

2.2. Statistical Methods

EXCEL, SPSS 26.0 and Stata15 software were used to analyze the data. The patients who were included in the study were cleaned by EXCEL, and the data with missing data were removed. In SPSS, the patients were randomly divided into training group and validation group according to the proportion of 7:3. Chi-square test was used to test the difference of clinicopathological features between these training groups and validation groups. The counting data were described by frequency, and the chi-square test was used to compare the two groups. The variables with $P < 0.05$ were included in multivariate Logistic regression analysis by univariate Logistic regression. Four clinical prediction models were obtained by direct input, forward method, backward method and stepwise method. The optimal model was selected according to AIC or BIC and likelihood ratio test, and the nomogram was drawn. The prediction model was evaluated by discrimination, calibration and clinical net benefit. The discrimination was evaluated by the area under (AUC) the receiver operating characteristic curve (ROC curve), calibration curve and Hosmer-Lemeshow goodness-of-fit test to evaluate the calibration, and decision curve analysis (DCA) to evaluate clinical effectiveness. Finally, the nomogram is constructed according to the selection of the optimal model. The difference was statistically significant ($P < 0.05$).

3. Results

3.1. Comparison of the Balance of Basic Clinical Characteristics between Training Set and Validation Set

Training set and validation set patients' lymph node status, age, race, marital status, primary site of tumor, tumor tissue differentiation degree, tumor histopathological type, distant metastasis of tumor, tumor diameter, SEER historic stage, AJCC Stage Group. There was no significant difference in 6th ed. ($P > 0.05$), see Table 1.

3.2. Univariate and Multivariate Analysis of Lymphatic Positive Risk Prediction for Cervical Cancer

Univariate analysis showed that there were 7 suspected risk factors associated with positive lymph nodes of cervical cancer. They were race, marital status, primary site of tumor, tumor tissue differentiation degree, tumor histopathological type, distant metastasis of tumor, tumor diameter ($P < 0.1$, see Table 2). The influencing factors with statistical significance in univariate analysis were included in multivariate Logistic regression analysis. The results showed that race, tumor tissue differentiation degree, tumor histopathological type, distant metastasis of tumor, tumor diameter There were considered statistically significant ($P < 0.05$, see Table 2).

Table 1. Clinical information for training sets and validation sets.

Variable	Training set (n = 4172)	Validation set (n = 1768)	P	Variable	Training set (n = 4172)	Validation set (n = 1768)	P
Lymph node status				**Tumor tissue differentiation**			
Negative	3147	1339	0.80	Level 1	610	232	0.18
Positive	1025	429		Level 2	1890	806	
Age				Level 3	1564	670	
<40	1161	498	0.94	Level 4	108	60	0.98
40 - 60	2069	877		**Distant metastasis of tumor**			
≥60	942	393		No distant metastasis	3801	1605	0.68
Race				Distant metastasis	371	163	
Whites	3091	1301	0.62	**Tumor diameter**			
Blacks	510	232		≤6	3420	1456	0.73
Native American	63	27		>6	752	312	
Asian	491	195		**SEER stage**			
Unidentified race	17	11		Regional	1632	705	0.94
Marital status				Localized	2129	889	
Married	1938	828	0.89	Distant	405	171	
Unmarried and live apart	1118	466					
Unmarried	613	268		**AJCC Stage**			
Widowed	323	138		I	2218	928	0.98
Unknown	182	68		II	568	251	
Tumor histological type				III	943	402	
Cervical squamous cell carcinoma	2692	1138	0.88	IV	437	184	
Squamous adenocarcinoma	161	71		unstage	6	3	
Adenocarcinoma	1206	517					
Others	113	42					
Primary site							
Endocervix	901	405	0.52				
Cervix uteri	3111	1302					
Exocervix	82	35					
Overlapping lesion of cervix uteri	78	25					

Table 2. Univariate analysis and multivariate Logistic analysis of cervical cancer lymph node metastasis.

Features	Univariate Logistic analysis					Multivariate Logistic analysis				
	B	OR	95% CI Lower	95% CI Upper	p	B	OR	95% CI Lower	95% CI Upper	p
Age					0.21					
<40			reference					NA		
40 - 60	0.11	1.12	0.97	1.29	0.11					
≥60	0.14	1.15	0.99	1.36	0.76					
Race					0.04					0.01*
Whites			reference					reference		
Blacks	1.43	4.17	0.99	17.62	0.05	1.16	3.17	0.73	13.77	0.12
American Indians/Alaska Native	1.44	4.23	0.99	17.97	0.05	0.88	2.42	0.55	10.61	0.24
Asian	1.97	7.17	1.6	32.2	0.01	1.65	5.25	1.12	24.52	0.03
Marital status					0.06					
Married			reference							
Unmarried	0.06	1.06	0.77	1.44	0.73					
Divorce and diaspora	0.25	1.28	0.93	1.77	0.13			NA		
Widowed	0.2	1.22	0.88	1.71	0.24					
Unknown	0.19	1.21	0.84	1.75	0.3					
Tumor histological type					<0.05					<0.05*
Cervical squamous cell carcinoma			reference					reference		
Squamous adenocarcinoma	0.23	1.26	0.94	1.68	0.12	−0.12	0.88	0.60	1.29	0.51
Adenocarcinoma	−0.53	0.59	0.51	0.68	0.00	0.09	1.09	0.68	1.77	0.70
Others	0.91	2.49	1.80	3.44	0.00	−0.48	0.61	0.41	0.91	0.02
The primary site of the tumor					0					0.72
Endocervix			reference					reference		
Cervix uteri	−0.46	0.63	0.54	0.74	0.00	−0.16	0.83	0.44	1.59	0.58
Exocervix	−0.68	0.51	0.31	0.84	0.01	0.48	1.13	0.62	2.07	0.69
Overlapping lesion of cervix uteri	0.08	1.08	0.70	1.66	0.73	0.10	1.10	0.87	1.38	0.43
Tumor tissue differentiation					<0.05					<0.05*
Level 1			reference					reference		
Level 2	1.30	3.67	2.78	4.80	0.00	1.61	2.98	2.23	3.98	0.00
Level 3	1.82	6.16	4.68	8.12	0.00	1.41	4.09	3.05	5.49	0.00
Level 4	2.14	8.49	5.67	12.72	0.00	1.80	6.04	3.89	9.39	0.00
Distant metastasis	2.27	9.65	7.92	11.76	0.00	2.02	7.54	6.11	9.29	0.00*
Tumor diameter	1.25	3.48	3.03	4.00	0.00	0.82	2.28	1.95	2.66	0.00*

3.3. Building a Prediction Model

In SPSS and STATA software, the treatment mode of patients with positive

lymph nodes of cervical cancer was taken as dependent variables (assigned: lymph node negative = 0, lymph node positive = 1). Seven variables selected from univariate logistic regression were included in multivariate Logistic regression analysis. Four clinical prediction models were constructed by forward method, backward method and stepwise method. The AIC were: 4035.367/4029.592/4038.9/4034.221. The BIC were: 4105.065/4073.945/4078.574/4070.581. According to the selection of the minimum AIC or BIC value and the comparison of likelihood ratio under the same AIC value, the optimal model was selected, and drew the nomogram according to the prediction variables, namely Nomogram (Figure 1). The corresponding values of each variable could be scored by the nomogram, and then the total score could be obtained by adding the scores of all variables, and a vertical line could be drawn down according to the total score, the estimated probability of lymph node metastasis risk of cervical cancer could be marked.

3.4. Verification of Prediction Model

The verification of the prediction model was mainly based on the discrimination and calibration of the model. The model discrimination was evaluated by drawing a prediction model to predict the ROC curve of cervical cancer lymph node metastasis risk. The AUC of the training set was 0.736 (95% CI (0.72)) (Figure 2(a)). The AUC of the verification set was 0.714 (95% CI (0.685, 0.742)) (Figure 2(b)). It showed that the prediction model had good discriminant ability. At the

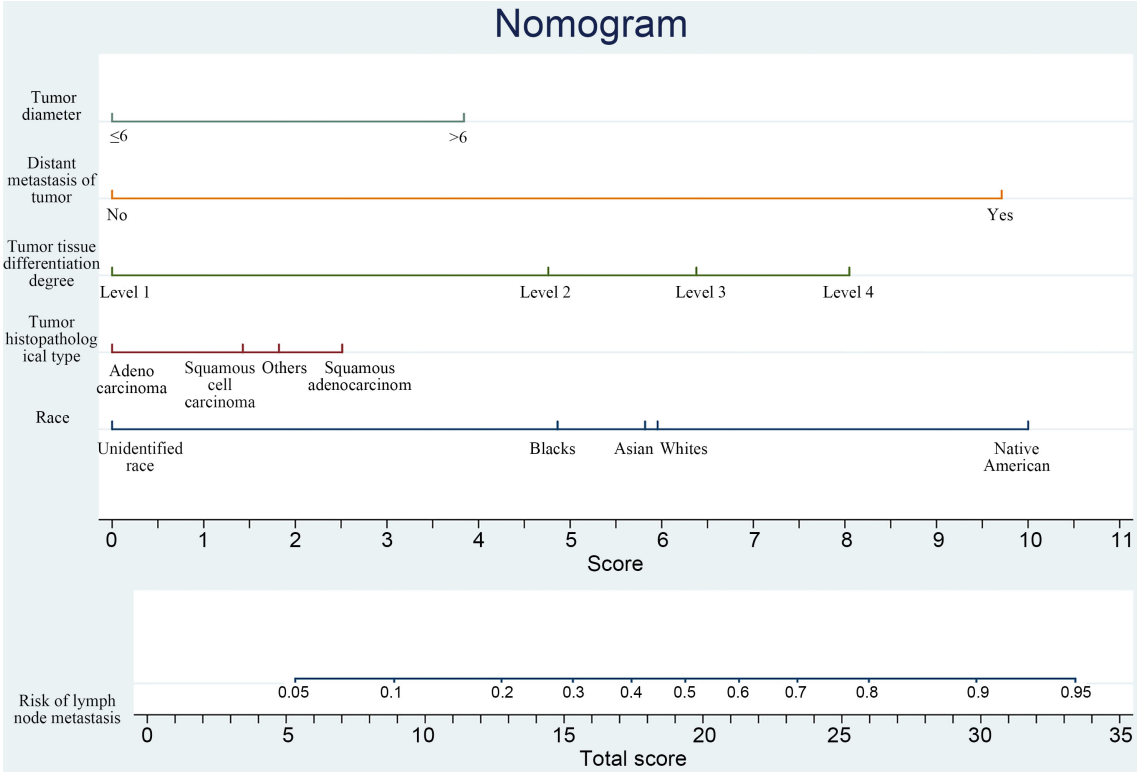

Figure 1. Nomogram.

same time, the Hosmer Lemeshow goodness-of-fit test showed a good degree of fit (training set 0.28; verification queue 0.11), which showed that the prediction probability of the model was basically consistent with the actual probability, and the model had a good calibration. In addition, the calibration curves of the training set and the verification set showed moderate consistency, and the correction ability of the prediction model was good (Figure 3 calibration curve). To sum up, the Nomogram of the prediction model had medium prediction ability.

3.5. Clinical Application

The clinical validity of the prediction model was evaluated by DCA. The DCA of the occurrence probability nomogram of cervical cancer lymph node metastasis was shown in Figure 4. The results showed that if the threshold probability of the patient and the doctor are more than 20% respectively, in the current study, the risk of positive lymph nodes in patients with cervical cancer using this nomogram will be more beneficial than that of all patients implementing intervention programs. Within this range, the net benefit of the prediction model was significantly higher than that of the two extremes. All patients have received clinical intervention.

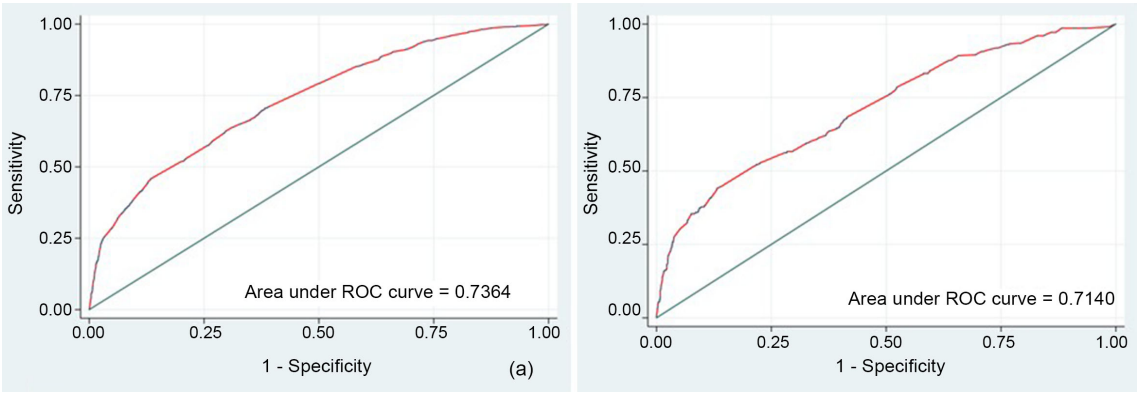

Figure 2. ROC curve classification of two sets of nomograms.

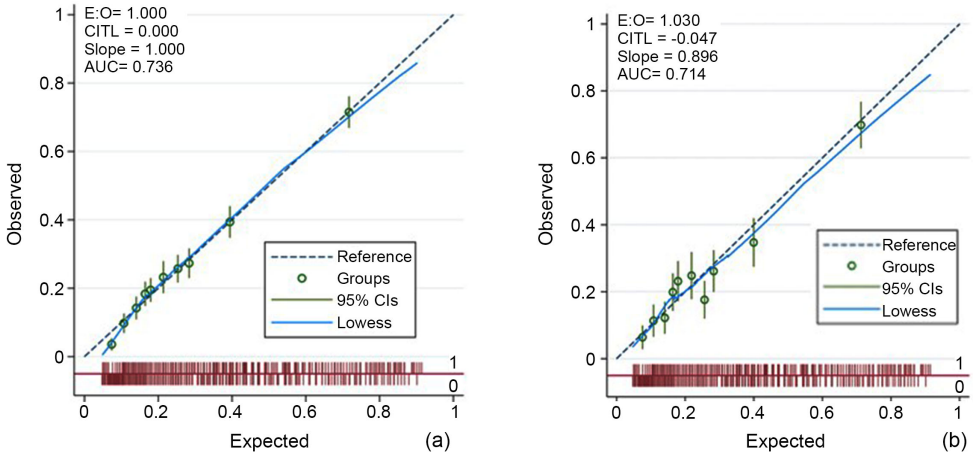

Figure 3. Calibration curves of two sets of nomograms.

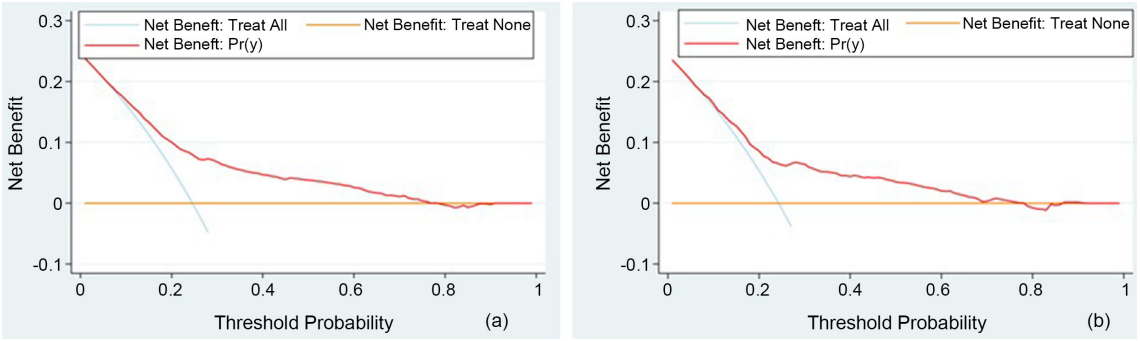

Figure 4. Decision curve analysis of two groups of nomograms.

4. Discussion

Cervical cancer is one of the four most common female malignant tumors in the world. It ranked second in cancer-related deaths among young women between the ages of 20 and 39 in 2020. The global incidence of cervical cancer is about 500,000 cases every year. More than one-fourth of all new cases and fatalities worldwide occur in China. With the exception of cervical and uterine body cancers, survival rates for all of the most prevalent cancers have increased since the middle of the 1970s. As we all know, the main way of metastasis of cervical cancer is through direct spread and lymph node metastasis, and many studies have shown that lymph node metastasis is an independent risk factor to evaluate the prognosis of cervical cancer. There were studies have shown that patients with pathological risk factors (lymph node metastasis, para-uterine infiltration, positive resection margins of vaginal stump) have a higher recurrence rate [5]. Moreover, lymph node status has been included in the staging criteria in the 2018 FIGO guidelines. There is currently controversy over the scope of cervical cancer surgery. The status of lymph nodes has guiding significance for the scope of surgery and postoperative adjuvant therapy. Preoperative evaluation of lymph node status in clinical work is very important for patients' treatment. Accurate preoperative assessment of lymph node status can reduce unnecessary lymph node dissection and reduce operative complications caused by lymph node dissection, such as pelvic vascular injury, Lymphocyst, Chylous fistula and so on. The examination methods for evaluating lymph node metastasis of cervical cancer in clinical practice mainly include imaging examination: MRI, PET-CT, PET-MRI, etc. [6] but its sensitivity is low. Studies by Sato [7] and others showed that CA125 was valuable in evaluating preoperative lymph node metastasis. In addition, SLNB (sentinel lymph node biopsy) is considered to be the most positive rate for lymph node metastasis. However, it is only applicable to cervical tumors with a diameter of less than 2cm, which cannot be widely carried out in clinical practice due to its invasive examination [6]. Currently, there is no quantitative index for comprehensive judgment of cervical cancer lymph node metastasis. Valuable high-risk factors of cervical cancer lymph node metastasis are calculated by statistics, and an effective prediction model is established. It can help clinicians identify high-risk patients and guide individualized treatment.

The SEER database (Surveillance by the National Cancer Institute/Epidemiology and final Database) collects a large number of patient information data, including demographics/disease diagnosis/tumor staging/treatment information and prognosis, which can provide a large amount of systematic data for clinicians. This study used a large population provided by SEER database and constructed and validated a nomogram for predicting lymph node metastasis of cervical cancer.

In this study, cervical cancer cases from 2004 to 2015 were screened by SEER database. Five risk factors for cervical cancer lymph node metastasis were calculated by statistical method: race, tumor tissue differentiation degree, tumor histopathological type, distant metastasis of tumor, tumor diameter. And clinical prediction model was constructed ($P < 0.1$, see **Table 2**). Similar studies have shown that the 5-year survival rate of cervical cancer patients younger than 35 years is lower than average. Moreover, the cancerous lesion is large, the recurrence rate is high, and the prognosis is poor. [8] Some similar studies have suggested that cervical cancer lymph node metastasis is associated with age. It is different from the results of this study, which may be due to the different number of cases and selection bias. Studies by Zhuang Jinman [9] and others showed that no cervical erosion, tumor maximum diameter > 3 cm, Para-uterine infiltration and lymphatic vascular interstitial infiltration were the high-risk factors affecting lymph node metastasis in early-stage cervical cancer. There are some similar predictive factors to this study. A retrospective study of 302 cases of cervical cancer by Yi-Fang Dai [10] *et al.* showed that there was no significant correlation between lymph node metastasis rate and tumor size (>4 cm), but it was related to tumor differentiation, depth of uterine myometrium infiltration, lymphatic vascular infiltration and other factors. However, the study of Zhuang *et al.* showed that the risk of lymph node metastasis of tumor maximum diameter > 3 cm was 1.98 times higher than that of tumor maximum diameter ≤ 3 cm. That is, the larger the maximum diameter of the tumor, the greater the possibility of lymph node metastasis. Zhuang *et al.*'s study included three factors: maximum diameter of tumor, para-uterine infiltration and infiltration of lymphatic vascular space to construct a nomogram to predict pelvic lymph node metastasis. In this study, a more detailed prediction model was constructed based on a large sample of clinical data, and the results showed that the model has better prediction ability. Finally, the results of decision curve analysis showed that our model is also of guiding significance for clinicians to make clinical decisions.

Although the prediction ability of this study is good, there are still the following shortcomings. 1) This study is a retrospective study, excluding a large number of incomplete cases, which may lead to selective bias. Therefore, more large samples of prospective studies are needed for further verification. 2) This study is based on the SEER database, there is no vascular invasion/nerve tissue invasion and para-uterine invasion and other data, so there are fewer risk factors. Therefore, more risk factors should be included in the following research to fur-

ther improve the prediction ability of the model. 3) The samples included in this study cover early and late-stage cases, so the score of distant tumor metastasis in the model construction is relatively high.

5. Conclusion

In summary, this study identified the risk factors associated with lymphatic metastasis in patients with cervical cancer by analyzing a large number of data obtained from the SEER database. Finally, we constructed a model with high predictive performance based on the five best risk factors, and constructed a nomogram to help clinicians assess the risk of lymphatic metastasis. Through individual risk assessment, clinicians and patients can choose personalized treatment plans and take necessary intervention measures in advance to extend the survival time of patients.

Conflicts of Interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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https://doi.org/10.1177/0300060518785827

Scientific Research Publishing

Yangtze Medicine, 2023, 7, 116-133
https://www.scirp.org/journal/ym
ISSN Online: 2475-7349
ISSN Print: 2475-7330

CIB1 as a Potential Diagnosis and Prognosis Biomarker in Uveal Melanoma

Xianwang Wang[1*#], Xiao Zhang[1*], Shujuan Hu[2*], Lei Ge[3], Zhiming Zou[3], Yingying Lu[4#]

[1]Department of Biochemistry and Molecular Biology, Health Science Center, Yangtze University, Jingzhou, China
[2]Institute of Education and Sports Sciences, Yangtze University, Jingzhou, China
[3]Department of Ophtalmology, The Second School of Clinical Medicine & Jingzhou Central Hospital, Yangtze University, Jingzhou, China
[4]The Seventh Affiliated Hospital, Sun Yat-sen University, Shenzhen, China
Email: #500851@yangtzeu.edu.cn, #luyy39@mail.sysu.edu.cn

How to cite this paper: Wang, X.W., Zhang, X., Hu, S.J., Ge, L., Zou, Z.M. and Lu, Y.Y. (2023) CIB1 as a Potential Diagnosis and Prognosis Biomarker in Uveal Melanoma. *Yangtze Medicine*, **7**, 116-133.
https://doi.org/10.4236/ym.2023.72012

Received: May 3, 2023
Accepted: June 27, 2023
Published: June 30, 2023

Abstract

Background: Uveal melanoma (UVM) is the most common primary intraocular tumor in adults. However, identification of the effective biomarker for the diagnosis and treatment of UVM remains to be explored. Calcium and integrin-binding protein 1 (CIB1) is emerging as an important factor in tumor progression. **Purpose:** To determine the contribution of CIB1 in the diagnosis of UVM. **Method:** Immunohistochemical staining is used to detect the CIB1 expression level, while Gene Expression Profiling Interactive Analysis 2 (GEPIA2) and UALCAN online tools were used to analyze patient survival and CIB1 correlation genes in UVM. Integrative analysis using STRING and GeneMANIA predicted the correlated genes with CIB1 in UVM. **Results:** CIB1 expression level in UVM was significantly enhanced when compared with that in paracancerous tissues. A higher CIB1 expression level resulted in a significantly worse disease-free survival as well as overall survival. Moreover, the survival probability of patients was associated with body weight and gender of the patients with UVM. The correlated genes with CIB1 in UVM, and the similarity of the genes in UVM expression and survival heatmap were verified. Furthermore, Gene ontology enrichment analysis revealed that CIB1 and its correlated genes are significantly enriched in ITGA2B-ITGB3-CIB1 complex, regulation of intracellular protein transport and regulation of ion transport. **Conclusions:** Our novel findings suggested that CIB1 might be a potential diagnostic predictor for UVM, and might contribute to the potential strategy for UVM treatment by targeting CIB1.

*These authors contributed equally to this work and are joint first authors.
#Correspondence author.

Keywords

CIB1, Uveal Melanoma, TCGA, Prognostic Biomarker, Patient Survival

1. Introduction

Melanomas are originating from melanocyte transformation and represent a fatal form of highly malignant cancer. There are two subtypes of melanomas have been classified, cutaneous melanoma (SKCM) and ocular melanoma (OM) [1]. As the most frequent (85%) primary intraocular malignancy arising in adults, uveal melanoma (UVM) is a clinically distinct and specifically lethal subset of melanoma deriving from eye melanocytes [2]. The large majority of UVM arises from the uvea (95%), comprising the posterior uvea (choroid 90% and ciliary body 5%) and the anterior uvea (5%) of eye [3]. Little is understood concerning the molecular pathogenesis of UVM in contrast to SKCM, particularly in associated genes or tumor suppressor pathways owning to perplexing host and environmental factors influencing the aggression of UVM. Currently, the treatment of localized UVM involves radiation, immunotherapy, laser therapy, or surgery excision [4] [5] [6] [7]. However, there are no effective therapeutic strategies for patients with UVM [1], and the prognosis of metastatic UVM remains not adequate, with an overall survival of less than one year in most objects [8]. Cytogenetic has identified several alterations, which include monosomy 3, chromosome 6 abnormalities and *MYC* amplification at 8q24 are strong diagnostic and prognostic markers in UVM [9] [10], to investigate the novel prognosticators of UVM and an enhanced interpretation of the potential molecular mechanisms and gene networks associated in the development and prognosis of UVM are still required.

Calcium and integrin-binding protein 1 (CIB1) is an EF-hand calcium-binding protein, which is ubiquitously expressed and is associated with various human cancers [11]. Accumulating evidence has fuelled the conclusion that CIB1 performs crucial roles in facilitating cell survival, proliferation and migration, thereby mediating tumor growth and tumor-induced angiogenesis [12] [13] [14]. In particular, Chung *et al.* recently documented that CIB1 depletion with docetaxel or TRAIL enhances triple-negative breast cancer cell death [15]. However, the potential diagnostic value of CIB1 on UVM is unclear.

In the present study, CIB1 expression level of the typical patients with malignant UVM was analyzed. Moreover, bioinformatics analysis from The Cancer Genome Atlas (TCGA) by UALCAN interactive web-portal [16] and Gene Expression Profiling Interactive Analysis 2 (GEPIA2) online tools [17] were used to identify the critical role of CIB1 gene in UVM diagnosis and prognosis. CIB1 expression level in UVM, effect of CIB1 expression level on patient survival, promoter methylation levels of CIB1 in UVM and the correlated genes with CIB1 in patients with UVM were also presented.

The STRING database (http://string-db.org) provides a critical evaluation and integration of protein-protein interactions (PPI), including physical and functional relevance [18]. GeneMANIA (http://genemania.org) is a flexible user-friendly website for generating hypotheses regarding gene function, analyzing gene lists and prioritizing genes for functional assay [19]. Metascape tool (http://metascape.org) provides a resource for biologists for the analysis of systems-level datasets [20]. Thus, CIB1 PPI network and Gene ontology (GO) enrichment analysis were highlighted by using STRING/GeneMANIA and Metascape, respectively. Taken together, the diagnostic and prognostic role of CIB1 in UVM was identified. Thus, the results from the current study might advance the development of antagonist CIB1 strategies for patients with UVM.

2. Materials and Methods

2.1. Patients and Ethics Statement

Patients were recruited from the Ophtalmology department of the Second School of Clinical Medicine & Jingzhou Central Hospital (Jingzhou, China). Inclusion criteria: 1) Deformities, diminutions, visual field defects, retinal detachment etc.; 2) diagnosis confirmed by post-operative pathology. Exclusion criteria: 1) other diseases of the eyes; 2) negative results of post-operative pathology. The study was approved by the ethics committee of the Health Science Center of Yangtze University (approval number: YZLL2020-019). All 10 participants gave written informed consent prior to UVM tissue sampling.

2.2. CIB1 Gene Expression and Mutations Analysis

UALCAN is an interactive web resource for analyzing cancer transcriptome data. To analyse the expression of CIB1 across TCGA tumors, the online software tools UALCAN (http://ualcan.path.uab.edu/analysis.html) and Gene Expression Profiling Interactive Analysis 2 (GEPIA2, http://gepia2.cancer-pku.cn/#index) were used. The expression level of CIB1 in patients with UVM are generated by both UALCAN and GEPIA2, and the CIB1 expression level in UVM based on individual cancer stage, tumor histology, and gender, weight, age of the patients were analyzed using UALCAN online tool. To verify the two isoforms structural of CIB1 and compare the two isoforms usage in patients with UVM, GEPIA2 platform was performed. X: cancers, Y: isoforms, datasets from UVM. The cBioPortal for Cancer Genomics (http://www.cbioportal.org/) provides a web resource for exploring, visualizing, and analyzing multidimensional cancer genomics data [21]. In the present study, cBioPortal for Cancer Genomic was used to further detect the CIB1 gene expression and mutations in UVM.

2.3. Survival Analysis of Patients with UVM

To detect the effect of CIB1 expression level on UVM patient survival, UALCAN web-portal was used to generate the survival plot. In order to assess the combined survival effect of CIB1 expression and clinical parameters such as the body

weight, sex, race of the patient, multivariate Kaplan-Meier (KM) survival analysis was applied. R scripts were written to divide all patients into these six categories and to generate KM plot. The *P* value obtained from log-rank test was used to indicate statistical significance of survival correlation between groups. The overall survival (OS) and disease free survival (DFS), based on the expression status of CIB1 gene in patients with UVM was generated using GEPIA2. GEPIA2 uses Mantel-Cox test for the hypothesis evaluation. The cox proportional hazard ratio and the 95% confidence interval information can be included in the survival plot. Half of the patients with UVM had high expression levels of CIB1 and half had low expression levels of CIB1.

2.4. Correlation Genes Analysis

To detect the correlation genes of CIB1 in UVM, correlation analysis was used. The 20 top-rank correlation genes of CIB1 expression in UVM were obtained using GEPIA2 online tool. The expression dataset is UVM tumor from TCGA. Pearson's correlation coefficient (Pearson CC) was used to screen the top 20 correlated genes that were ≥0.78. Moreover, the top 50 positive and negative correlated genes of CIB1 expression in UVM were analyzed from UALCAN dataset. If pearson CC > 0, represent genes positively correlated with CIB1 in UVM; when, pearson CC < 0, genes negatively correlated with CIB1 in UVM. Concerning top 50 positive correlated genes, pearson CC are selected more than 0.73; Negative correlated genes, pearson CC are screened less than minus 0.53. Furthermore, the 5 top-rank positively correlated genes (ORMDL2, MRPS34, MRPS11, ATOX1 and ZDHHC12) and negatively correlated genes (RPL32, RPL14, C6orf48, LTA4H and FBL) with CIB1 in UVM were also analyzed using UALCAN online tool.

2.5. Expression and Survival Heatmap Analysis

The heatmap profile of correlation genes of CIB1 expression in UVM and additional TCGA cancer types were analyzed using an interactive heatmap, and the multiple gene comparison tools in GEPIA2 was applied. The tumor data from TCGA was selected. To compare the survival contribution of top 20 positive and negative correlated genes of CIB1 expression in UVM and additional TCGA cancer types, the survival map was calculated from TCGA-tumor specimens using the Mantel-Cox test.

2.6. Promoter Methylation Analysis

To analyze the CIB1 promoter methylation levels in patients with UVM, the CIB1 promoter methylation profile based on individual cancer stage, and gender, weight, age of patients was analyzed using UALCAN online tool.

2.7. Protein-Protein Interaction (PPI) Networks and GO Enrichment Analysis

The STRING database (http://string-db.org) provides a critical assessment and

integration of PPI, including physical as well as functional associations [18]. The PPI network of CIB1 was contrasted using STRING version 10.0 online tools. GeneMANIA (http://genemania.org) is a flexible user-friendly web site for generating hypotheses concerning gene function, analyzing gene lists and prioritizing genes for functional assays [19]. GeneMANIA was used to further analyze the related genes of CIB1. GO enrichments were analyzed using Metascape tool (http://metascape.org) explored in 2019 [20].

3. Results

3.1. Elevated Expression Level of CIB1 in Patients with UVM

To determine the expression level of CIB1 in UVM, immunohistochemical (IHC) staining was used firstly. As shown in **Figure 1A**, a significant increase expression level of CIB1 was observed from UVM tissues (middle) when compared with that in normal tissues (surrounding normal choroidal melanocytes) and that in negative control groups. Socio-demographic characteristics of patients in this study suggested that the proportion of elderly patients is increasing, and the proportion of men decreased and the proportion of women increased. To detect the CIB1 expression profile in UVM based on individual cancer stage, tumor histology, and sex, weight, age of patients, UALCAN web resource was analyzed. As shown in **Figures 1B-F**, there were no significantly difference expression levels of CIB1 based on gender (**Figure 1D**) and weight (**Figure 1E**) of the patients. However, individual cancer stages (**Figure 1B**), patient's age (**Figure 1C**) and tumor histology (**Figure 1F**) might be the potential impact factor for CIB1 expression in patients with UVM. In particular, the expression level of CIB1 is significantly elevated in stage 4 when compared with that in stage 3 (**Figure 1B**, $p < 0.001$). As shown in **Figure 1C**, older patients (81 - 100 years) exhibited higher CIB1 expression level than that in young patients (21 - 40 years, $p < 0.05$). Epithelioid cells expressed more CIB1 than that in spindle cells (**Figure 1F**, $p < 0.01$).

As shown in **Figure 1G**, cBioPortal for Cancer Genomic analysis demonstrated that there are several gains, diploid and shallow deletions were seen in TCGA-UVM samples. There are two isoforms of CIB1 have been identified, CIB1-001 and CIB1-201. To verify the two structural isoforms of CIB1 and compare the occurrence of the two major isoforms in patients with UVM, GEPIA2 dataset analysis was further used. As shown in **Figures 1H-I**, two isoforms CIB1-001 and CIB1-201 consist of 191aa and 231aa, respectively. Each of them contains the EF-hands for their multiple functions. The occurrence of CIB1-001 is more frequent in patients with UVM compared with that in CIB1-201 (**Figure 1H**).

Taken together, the result revealed that CIB1 had high expression in UVM compared with that in normal tissues.

3.2. Effect of CIB1 Expression on Patient Survival

To evaluate the prognostic value of CIB1 in patients with UVM, the overall

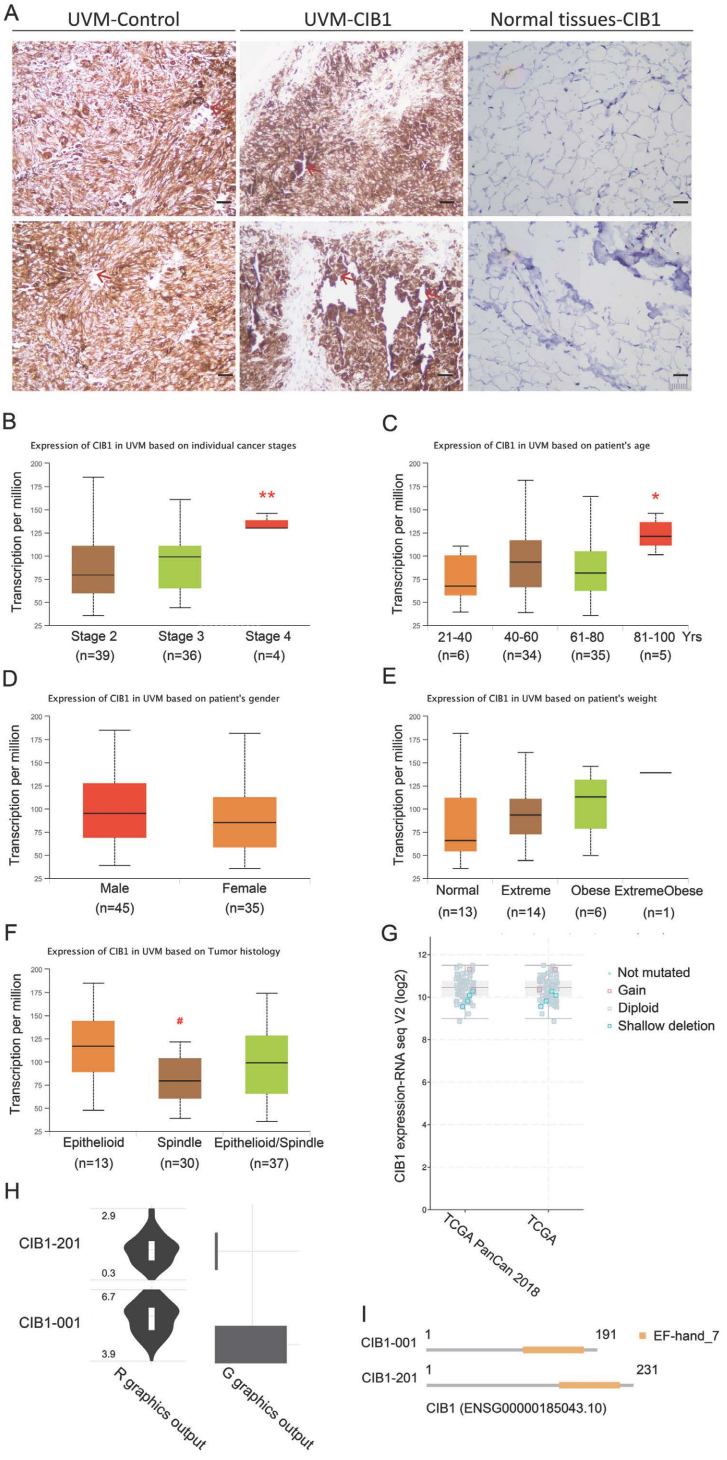

Figure 1. Expression of CIB1 in patients with UVM. A. The CIB1 expression levels in patients with UVM. Images of CIB1 immunohistochemical staining in normal tissue with CIB1 positive staining (control), UVM with CIB1 positive staining and negative staining (200x, bar = 10 μm; the lesion sites have been marked with arrows); Expression level of CIB1 in patients with UVM based on individual cancer stage (B, **$p < 0.001$, stage 4 vs stage 3), and age (C, *$p < 0.05$, 81 - 100 years vs 21 - 40 years), gender (D), weight (E) of the patients, tumor histology (F, #$p < 0.01$, epithelioid vs spindle and epithelioid/spindle vs spindle), respectively; G. The expression levels and mutations of CIB1 in patients with UVM analyzed using cBioPortal for Cancer Genomic; H. Occurrence of two CIB1 isoforms in patients with UVM. X: cancers, Y: isoforms, datasets from UVM. I. Two structural isoforms of CIB1.

survival (OS) and disease free survival (DFS) of CIB1 expression level in UVM were analyzed. GEPIA2 results demonstrated that a higher CIB1 expression level resulted in significantly lower survival probability (**Figure 2A**, OS, $p < 0.0011$ and **Figure 2B**, RFS, $p = 0.031$). UALCAN web portal analysis further exhibited the consist conclusion (**Figure 2C**, $p < 0.0001$).

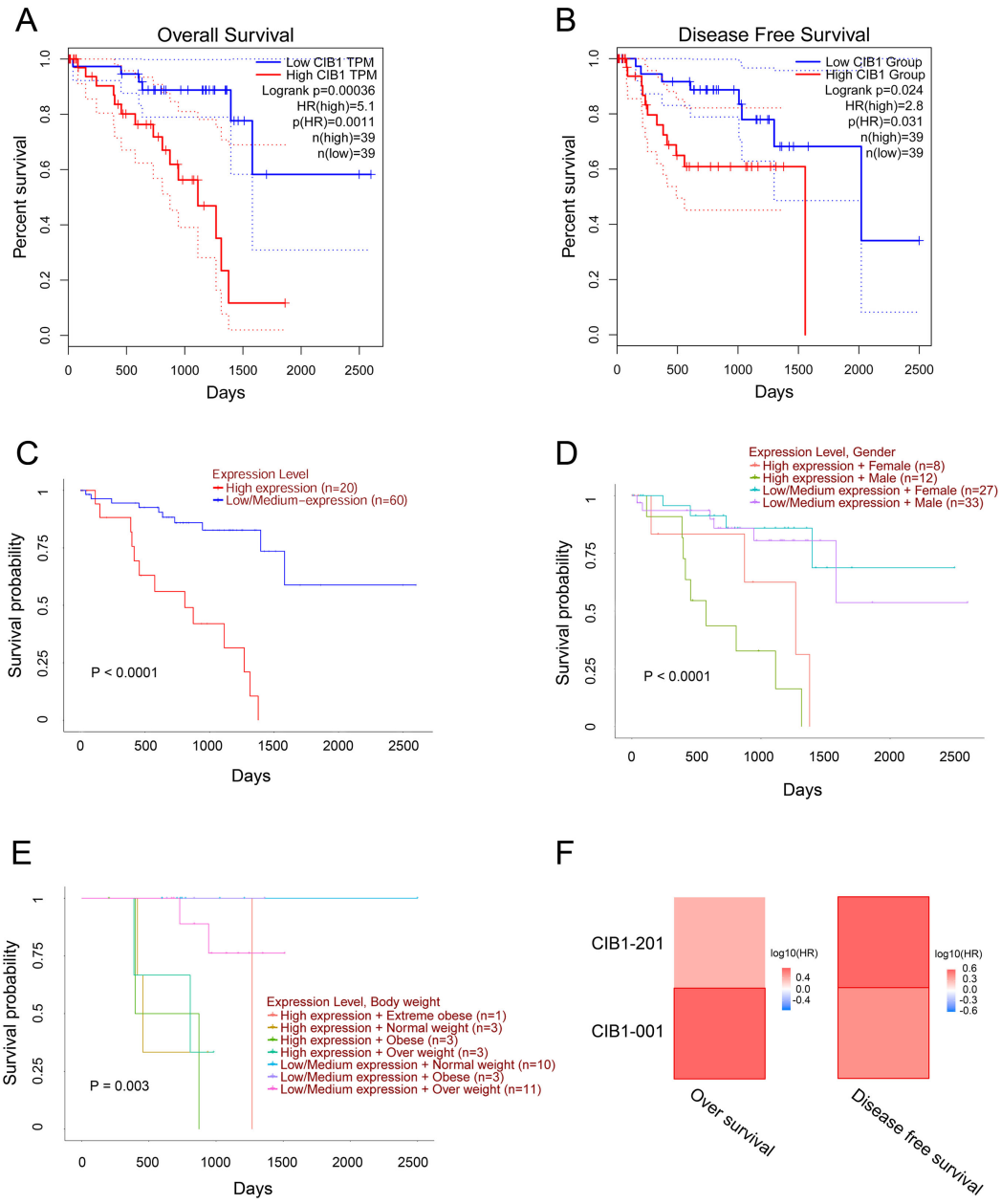

Figure 2. Effect of CIB1 expression level on survival probility in patients with UVM. A-B. A higher expression level of CIB1 was associated with poorer OS (A) and DFS (B) in patients with UVM, respectively; Median cut-off model (cut-off high and cut-off low are 50%) was selected for analysis, calculation of the hazards ratio was based on Cox PH Model. Add the 95% confidence intervals as dotted line. Axis Units are days. High groups and low groups are represented by red and blue, respectively. C. Effect of CIB1 expression levels on patient's survival was mined from UALCAN interactive web-portal; D. Effect of CIB1 expression levels & gender on UVM patient's survival; E. Effect of CIB1 expression levels & body weight on UVM patient's survival; F. Survival heatmap of two CIB1 isoforms in patients with UVM.

In addition, the survival probability was also associated with body weight and sex of the patients. As shown in Figure 2D, the male patients exhibited poorer survival probability when compared with that in female patients. Body weights of patient also effect the patient survival obviously, as shown in Figure 2E, obese and overweight patients demonstrated shorter survival time. The contribution of the CIB1 isoforms to the survival probability of patients with UVM was determined. As shown in Figure 2F, survival map analysis verified that CIB1-001 is the major isoform effect the survival of patient with UVM. These data indicated that CIB1 as a promising prognostic biomarker in patients with UVM.

3.3. Correlation Genes with CIB1 in Patients with UVM

To identify genes correlated with CIB1 in patients with UVM, the top 20 correlated genes with CIB1 expression in UVM were mined using GEPIA2 online tool and summarized in Table 1 (P CC > 0.78). Besides, the 50 top-rank positive and negative correlated genes with CIB1 expression in UVM were investigated using UALCAN tool and listed in Table 2.

Table 1. Top 50 ranked CIB1-positively and -negatively correlated genes in TCGA-UVM dataset.

Genes Number	Similar Genes	Genes ID	Pearson CC
1	ORMDL2	ENSG00000123353.9	0.85
2	MRPS34	ENSG00000074071.13	0.85
3	MRPS11	ENSG00000181991.15	0.84
4	ATOX1	ENSG00000177556.11	0.83
5	ZDHHC12	ENSG00000160446.18	0.82
6	MYL6	ENSG00000092841.18	0.81
7	POR	ENSG00000127948.13	0.80
8	MSRB1	ENSG00000198736.11	0.80
9	MPV17L2	ENSG00000254858.9	0.80
10	LAMTOR2	ENSG00000116586.11	0.80
11	DHRS7B	ENSG00000109016.17	0.80
12	ARPC1B	ENSG00000130429.12	0.80
13	PAGR1	ENSG00000238045.9	0.79
14	NDUFAF1	ENSG00000137806.8	0.79
15	GLA	ENSG00000102393.9	0.79
16	DNAJB12	ENSG00000148719.14	0.79
17	AIFM2	ENSG00000042286.14	0.79
18	ADCK5	ENSG00000173137.11	0.79
19	HM13	ENSG00000101294.16	0.78
20	ALG1	ENSG00000033011.11	0.78

Table 2. Top 20-ranked similar genes with CIB1 in patients with UVM.

Positive Correlated Genes	Pearson CC	Negative Correlated Genes	Pearson CC
ORMDL2	0.82	RPL32	−0.7
MRPS34	0.82	RPL14	−0.67
MRPS11	0.81	C6orf48	−0.66
ATOX1	0.81	LTA4H	−0.65
ZDHHC12	0.79	FBL	−0.64
C15orf63	0.79	RPL15	−0.64
NDUFAF1	0.78	RPSA	−0.63
GLA	0.78	RPL24	−0.62
ARPC1B	0.78	RPSAP58	−0.62
SEPX1	0.78	RPL3	−0.61
ROBLD3	0.78	QARS	−0.61
ADCK5	0.77	RPS3	−0.61
TNFRSF1A	0.77	CSNK2A2	−0.6
POR	0.77	RPS5	−0.6
DNAJB12	0.77	NCRNA00219	−0.59
MYL6	0.77	RPL35A	−0.59
HM13	0.77	EEF1G	−0.59
MPV17L2	0.77	RPSAP9	−0.59
DHRS7B	0.77	RPL12	−0.58
DYNLL1	0.76	LGTN	−0.58
SFXN3	0.76	SNHG7	−0.58
AIFM2	0.76	RPS4X	−0.58
S100A13	0.76	RPS13	−0.57
P4HA2	0.76	ZNF677	−0.57
SRA1	0.76	RPL10A	−0.57
WDR25	0.75	RPS18	−0.56
TCIRG1	0.75	LYRM4	−0.56
ALG1	0.75	RPL29	−0.56
CHAC1	0.75	EIF3L	−0.56
POLR3K	0.75	CTF1	−0.56
C12orf62	0.75	RPL23	−0.56
ATP6V0B	0.74	RPL38	−0.55
SIL1	0.74	MTUS1	−0.55
GSDMD	0.74	RPS2	−0.55
PFN1	0.74	RPL37	−0.55
IDH2	0.74	C14orf93	−0.55

Continued

FKBP2	0.74	RPL10	−0.55
SLC22A18	0.74	IMPDH2	−0.55
STX4	0.74	GLTSCR2	−0.55
MVP	0.74	RPS9	−0.55
IMP3	0.74	CDCA7L	−0.55
COTL1	0.74	GAS5	−0.54
DOK1	0.73	SLC25A38	−0.54
KDELR3	0.73	RPS23	−0.53
IRAK1	0.73	HNRNPA1	−0.53
NCS1	0.73	LETMD1	−0.53
MAGIX	0.73	RPL22	−0.53
MFSD5	0.73	GNB2L1	−0.53
RAB15	0.73	NMNAT3	−0.53
PSMB3	0.73	SNHG8	−0.53

As shown in **Figure 3A**, correlation analysis of the top 5 CIB1-positively correlated genes (ORMDL2, MRPS34, MRPS11, ATOX1 and ZDHHC12) and CIB1-negatively correlated genes (RPL32, RPL14, C6orf48, LTA4H and FBL) were analyzed. To further identified the similarity of the top 20 correlated genes, the expression heatmap and survival heatmap were evaluated. As shown in **Figure 3B**, the positively correlated genes expressions across TCGA tumors are very similar to that in CIB1, especially in UVM cancer types. In contrast, the negatively correlated genes expressions in patients with UVM are widely divergent to that in CIB1 (**Figure 3C**). To further analysis the survival heatmaps, as shown in **Figure 3D**, the results confirmed that the top-20 positively correlated gene showed high hazards ratio in UVM, but not in additional cancers. The top-20 negatively correlated gene showed low hazards ratio in UVM (blue) when compared with that in additional cancer types (**Figure 3E**).

3.4. Promoter Methylation Levels of CIB1 in Patients with UVM

Promoter methylation usually consider as a marker of gene inactivation [22]. To investigate the CIB1 promoter methylation profile based on individual cancer stage, and gender, weight and age of the patients, UALCAN online tool was used. As shown in **Figure 4**, the results revealed that the individual cancer stage (A), and age (C), gender (D) of the patients might not contribute greatly to the CIB1 promoter methylation in patients with UVM. However, patient's weight could alter the promoter methylation level of CIB1 in patients with UVM (**Figure 4B**, normal vs obese, $p < 0.05$).

3.5. PPI Network and GO Enrichment Analysis of CIB1

The functional interactions between proteins can provide important information

of the molecular mechanism involved. PPI network of CIB1 was presented using the STRING database (**Figure 5A**). The result indicated that CIB1 has interactions with 11 functional partners, including GUF1, CRY2, PLK3, ACTN4, PSEN2, ITGA2B and ITGB3, MAP3K5, CABP1, SPICE1, and ARHGEF1. To be specific, translation factor GUF1 [23], which promotes mitochondrial protein synthesis, and CRY2, transcriptional repressor which forms a core component of the circadian clock [24].

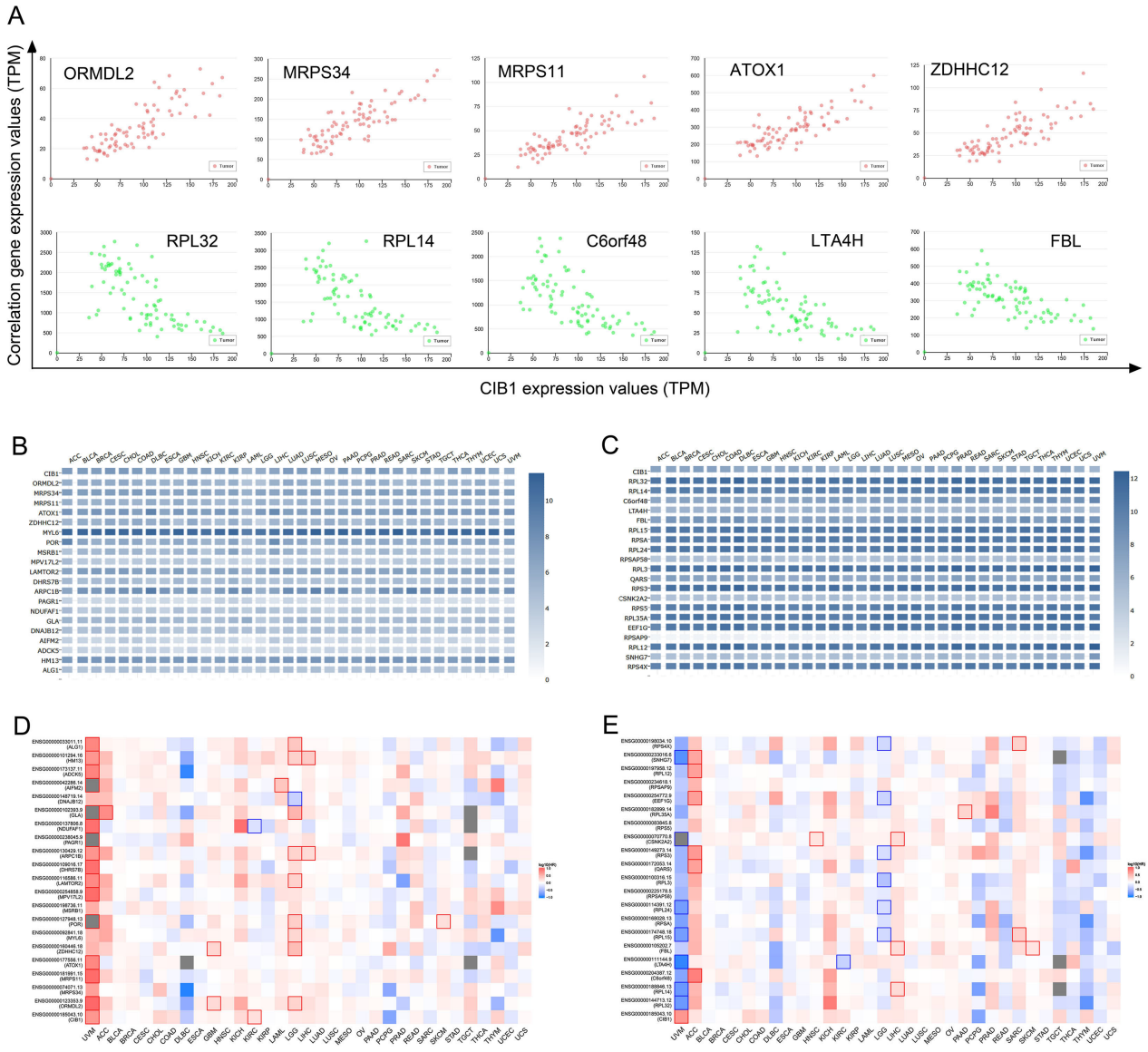

Figure 3. Correlation genes with CIB1 in patients with UVM. A. The correlations between the top 5 positively associated genes (ORMDL2, MRPS34, MRPS11, ATOX1 and ZDHHC12) and top 5 negatively correlation genes (RPL32, RPL14, C6orf48, LTA4H and FBL) with CIB1 in patients with UVM; B. Expression heatmap of 20 top-rank CIB1 positively correlated genes and CIB1 in patients with UVM and additional TCGA tumors; C. Expression heatmap of top 20 CIB1 negatively correlated genes and CIB1 gene in patients with UVM and additional TCGA tumors; D. Survival heatmap of top 20 CIB1 positively correlated genes and CIB1 gene n patients with UVM and additional TCGA tumors; E. Survival heatmap of top 20 CIB1 negatively correlated genes and CIB1 gene in patients with UVM and additional TCGA tumors.

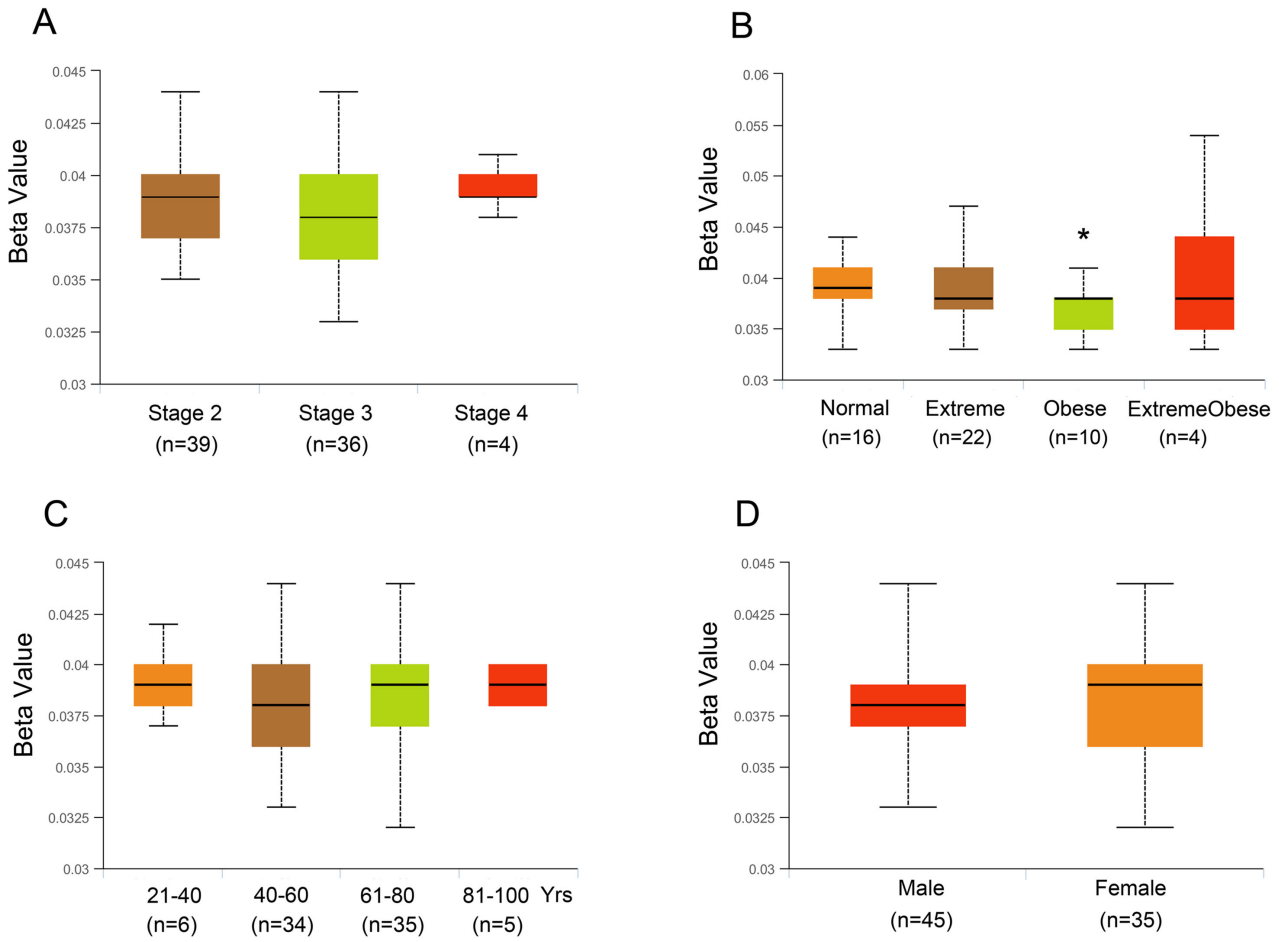

Figure 4. Promoter methylation levels of CIB1 in patients with UVM. Promoter methylation level of CIB1 in patients with UVM based on individual cancer stages (A), and weight (B), age (C), gender of the patient (D) (*$p < 0.05$, normal vs obese).

As shown in Figure 5B, 20 interactive genes of CIB1 were further identified using GeneMANIA, including PSEN2, PLK3, ITGA2B, and ITGB3, which consistent with the aforementioned correlated CIB1 genes from Figure 5A. Moreover, 16 other correlated genes from GeneMANIA dataset are PAX3, RAC3, PLK2, PSEN1, POLG, IDH2, FEZ1, ATP6V0E1, CD151, LRP10, GPX4, UBR5, POLD4, S100A10, CSTB, and C21orf33. Consistent with the aforementioned correlation analysis in Figure 3, IDH2 is just belonging to the top 50 ranked CIB1 positively correlated genes in patients with UVM.

To determine the potential function CIB1 in UVM, GO enrichment analysis of CIB1 and the genes it interacts with were performed using Metascape tool (Figure 5C and Figure 5D). The results suggested that CIB1 and the genes it interacts with are significantly enriched in ITGA2B-ITGB3-CIB1 complex, regulation of intracellular protein delivery, regulation of ion transport, cellular response to tumor necrosis factor and positive regulation of apoptotic process.

4. Discussion

Patients with UVM at the highest risk for progression to metastatic disease [25],

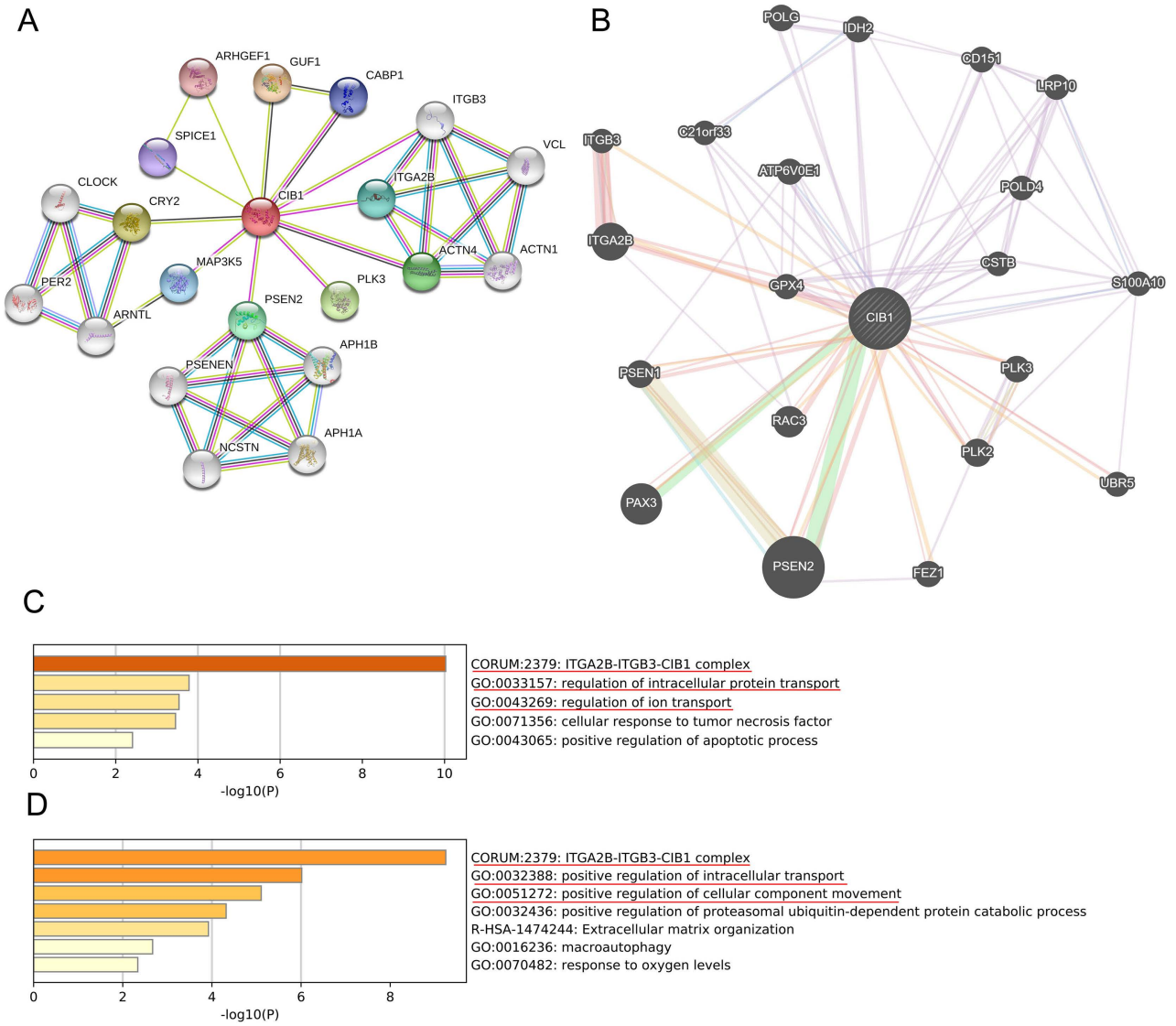

Figure 5. PPI networks and GO enrichment analysis of CIB1. A. PPI network of CIB1 and its interactive genes were constructed. Network nodes represent proteins; colored nodes represent CIB1 and the first shell of interacting proteins; white nodes represent the second shell of interacting proteins; B. PPI network of CIB1 was created using GeneMANIA. A total of 20-associated genes, with 21 total genes and 134 total links are shown. Interactions with CIB1 are indicated using stripes. C. Bar graph of enriched terms across input CIB1 and its interactive genes from Figure A, colored by p-values. Top 5 clusters with their representative enriched terms (one per cluster) were showed. Log10 (P) is the p-value in log base 10. D. GO enrichment analysis of CIB1 and the top 20 interactive genes obtained from Figure B, colored by p-values. Top 7 clusters with their representative enriched terms (one per cluster) were showed.

[26] often results in high rates of mortality; effective prognostic indicators used to evaluate the survival of patients with UVM are limited [27], [28]. In the present study, the most important discovery is reported that CIB1 as a potential diagnostic and prognostic biomarker in patients with UVM and might shed light on the UVM treatment by targeting CIB1. Our novel findings suggest that elevated CIB1 expression level in UVM compared with that in normal tissues, and several gain, diploid and shallow deletion were seen in TCGA samples. The results might assist us to understand the underlying carcinogenesis or progression of UVM better.

Moreover, the higher CIB1 transcripts per million (TPM) resulted in a significantly worse RFS and OS. The expression level of CIB1 in patients with UVM was obviously associated to body weight and gender of the patients. Besides, female gender and overweight body of the patients are considered as the important factors for facilitating their poorer survival probability. Most individuals who are obese harbor inflamed adipose tissue, which resembles chronically injured tissue, with immune cell infiltration and remodeling. Within this tumor microenvironment, several pathophysiologic changes are revealed that might contribute to cancers [29]. The positively correlated genes almost exhibited the similar expression and survival profiles to that of CIB1 in patients with UVM. The genes might have been implicated in poor prognosis and recurrence of patients with UVM. To be specific, it is identified that copper chaperone ATOX1 is required for MAPK signaling and growth in BRAF mutation-positive melanoma [30]. The mitochondrial ribosomal protein of the small subunits 34 (MRPS34) is explored to compromise protein synthesis and influence mitochondrial dysfunction [31]. Owning to similar expression and survival heatmap with CIB1 in patients with UVM, the data might provide more clues for prognostic judgment of UVM in the future. However, MYL6, ARPC1B and HM13 genes indicated the even higher expression levels than CIB1 in UVM from the top 20 ranked CIB1 correlated genes. Intriguingly, three CIB1-negatively correlated genes, C6orf48, RPSAP58 and CSNK2A2, showed similar levels of expression to CIB1 in patients with UVM. Thus, the molecular mechanism and possible applications of the correlated genes in UVM required intensively disclosed in the further study.

From the PPI network analysis, STRING and GeneMANIA identified 11- and 20-associated genes, respectively. Several CIB1-interactive genes, such as PLK3, PSEN2, ITGA2B, PAX3, RAC3, PLK2 and FEZ1, have been welled elaborated previously [11] [32]. Four same genes PSEN2, PLK3, ITGA2B and ITGB3 were identified using both STRING and GeneMANIA. It is also an interesting question that if the genes are associated with CIB1 in patients with UVM? To be specific, isocitrate dehydrogenase 2 (IDH2), which mined by GeneMANIA, is belonging to the 50 top-ranks CIB1 positively correlated genes in patients with UVM. IDH2, an important mitochondrial metabolic enzyme involved in the tricarboxylic acid cycle, is mutated in a variety of cancers [33] [34]. Increasing evidence has described that IDH2 play crucial roles in the prognosis and treatment of acute myeloid leukemia patients [35] [36]. Therefore, IDH2 is likely to be a potential biomarker for patients with UVM by further identification. Finally, Metascape tool analysis revealed that CIB1 and its correlated genes in UVM are intimately associated with ITGA2B-ITGB3-CIB1 complex, regulation of intracellular protein transport and regulation of ion transport. ITGA2B-ITGB3 receptor complex (αIIbβ3) is the paradigmatic integrin receptor. The binding of CIB1 to αIIbβ3 is verified to hamper phospholipase C (PLC)/IP3 signaling and intracellular Ca^{2+} release from ER store by IP_3R channel [11]. Combing with the GO enrichment analysis, we hypothesized that the pathogenic mechanism of CIB1 in UVM probably associated with αIIbβ3-mediated cell migration and/or intracel-

lular Ca^{2+} signaling transduction. However, further studies are required to intensively unveil the molecular mechanism and implication of CIB1 or correlated genes in UVM tumorigenesis and treatment through animal experiments or even cell experiments rather than the biogenic analysis. Moreover, the study had a pretty limited sample size, so more work needs to be done to verify and confirm the results.

5. Conclusion

In summary, the present study has identified that CIB1 is highly expressed in patients with UVM and elevated expression level of CIB1 is negatively correlated with survival probability in patients with UVM for the first time. The study might aid the identification of the underlying mechanism in UVM progression, and provide the high prognostic value of CIB1 in UVM. However, the detailed molecular pathogenesis and alteration in signaling pathways of CIB1 and the correlated genes in UVM are required to investigate in the future.

Declarations

Ethics approval and consent to participate:

This study was approved by the ethics committee of Yangtze University Health Science Center (approval number: YZLL2020-019) and participants provided written informed consent.

Author Contributions

SJ and XW designed the study. X, SJ and XW drafted the manuscript. SJ, X and XW analyzed the data and filled the tables. XW and YY revised the manuscript. All authors contributed to the article and approved the submitted version.

Funding

This work was supported by grants from the Central Government guides local funds for scientific and Technological Development (XZ202201YD0024C), Key R & D Program of Hubei Province (2021BGD010), Hubei Province Scientific and Technological Research Project (D20201306), Hubei Province Health Research Project (WJ2019-01), Hubei Medical Youth Tip-Top Talent, Leading Talent Program of Yangtze Talent Project and the College Students Innovative Entrepreneurial Training Program in Yangtze University (YZ2021296 and YZ2022307).

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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Scientific Research Publishing

Yangtze Medicine, 2023, 7, 134-146
https://www.scirp.org/journal/ym
ISSN Online: 2475-7349
ISSN Print: 2475-7330

Sodium Aescinate Alleviates Neuropathic Pain in Rats by Suppressing the TLR4/NF KB Pathway Activation after Paclitaxel Chemotherapy

Miao Huo[1], Qian Zhang[2], Ziyu Zhao[1], Ruifen Xu[1], Xingxing Zheng[1], Guang Yang[1], Jiao Guo[1*]

[1]Department of Anesthesiology, Shaanxi Provincial People's Hospital, Xi'an, China
[2]Department of Burn and Plastic Surgery, Shaanxi Provincial People's Hospital, Xi'an, China
Email: *hxyylp@163.com

How to cite this paper: Huo, M., Zhang, Q., Zhao, Z.Y., Xu, R.F., Zheng, X.X., Yang, G. and Guo, J. (2023) Sodium Aescinate Alleviates Neuropathic Pain in Rats by Suppressing the TLR4/NF KB Pathway Activation after Paclitaxel Chemotherapy. *Yangtze Medicine*, **7**, 134-146.
https://doi.org/10.4236/ym.2023.72013

Received: April 28, 2023
Accepted: June 27, 2023
Published: June 30, 2023

Abstract

Background: Emerging evidence suggests that chemotherapy-induced peripheral neuropathy (CIPN) is a significant side effect of chemotherapeutic drugs. Many experiments have proved that sodium aescinate (SA) has definite pharmacological effects such as anti-infection, anti-exudation, anti-edema, anti-tumor as well as neuroprotection, and the drug side effects are mild. However, no study has explored whether SA is involved in the analgesic effect of paclitaxel (PAC) induced neuropathic pain in rats. **Methods:** Rats were given an intraperitoneal injection of PAC (2.5 mg/Kg intraperitoneally on days 1, 3, 5, and 7), while SA 25 mg/kg intraperitoneally was administered daily for 14 consecutive days. The mechanical withdrawal threshold (MWT) and thermal withdrawal latency (TWL) of rats were examined on experimental days 3, 5, 7, 11, 14. All rats were sacrificed on day 15 of the experiment, and L4-6 spinal cords were removed. Subsequently, immunohistochemistry, HE staining, ELISA, RT-qPCR, Western blotting were applied to evaluate cytoskeletal protein expression (NF-L and NF-M), spinal nerve structural integrity, proinflammatory factor contents (TNF-α, IL-1β, and IL-6), and protein content of the TLR4/NF-κB pathway, respectively. **Results:** After the rats developed PAC induced pain behaviors, multiple injections of SA rendered the rats with elevated MWT and TWL values, decreased expression of NF-L and NF-M in the spinal cord, materially downregulated content of proinflammatory factors, and reduced amounts of TLR4 and p-NF-κB protein levels. **Conclusions:** The results of the present study preliminarily indicate that SA has an analgesic effect on rats with CIPN induced by PAC injection, and the mechanism may be related to blocking the TLR4/NF-κB signaling pathway, inhibiting the expression of proinflammatory factors, and alleviating cytoskeletal disorders.

Keywords

Chemotherapy-Induced Peripheral Neuropathy, Sodium Aescinate, The TLR4/NF-κB Pathway, Paclitaxel

1. Introduction

Cancer seriously endangers human health, and its incidence is increasing year by year. There are currently approximately 7 million new cancer patients occurring annually worldwide according to the World Health Organization [1]. Chemotherapy is one of the current important modalities for treating tumors, and although it can prolong the survival time of patients, it is accompanied by some distressing side effects [2]. Among them, chemotherapy-induced peripheral neuropathy (CIPN) is one of the common side effects in cancer patients and refers to the use of a specific class of chemotherapeutic drugs (phytoalkaloids, paclitaxels, platinum, etc.) resulting in the impairment of sensory, motor, autonomic nerve conduction, of which the symptom of sensory nerve damage is the most common [3]. The current clinical use of platinum, paclitaxel (PAC), and vincristine-based chemotherapeutics has been associated with a higher incidence of CIPN [4]. Statistically, acute onset of CIPN occurs in 65% - 96% and conversion to chronic CIPN occurs in 40% - 93% following oxaliplatin use [5]. The incidence of CIPN following cisplatin use is 12% - 85% [6]. Vincristine exposure occurs in 20%. The highest incidence of CIPN is seen with paclitaxels, ranging from 61% - 92%, and 30% of patients continue to endure the pain of CIPN 6 months after completion of chemotherapy [7]. At present, the lack of information related to the treatment of CIPN makes the patient's symptoms not effectively controlled, which in turn affects the progress or even the termination of chemotherapy. CIPN persists even after the completion of chemotherapy, leaving patients facing physical and functional challenges, experiencing social difficulties, emotions, sleep disturbances, etc., and severely affecting quality of life. Therefore, the development of effective drugs against CINP is a very urgent thing for cancer chemotherapy patients.

Aescinate is a natural mixture of triterpenoid saponins extracted from the seeds of *S. aestivum* after mature drying, and its major components are aescinate A, aescinate B, aescinate C and aescinate D [8]. In clinic, it is mostly applied in the form of tablets, gels, lyophilized powder of sodium aescinate (SA), and so on, and the oral form of SA tablets is most widely used [9]. SA has high solubility in water and good stability. A large number of experiments have proved that SA has definite pharmacological effects such as anti-infection, anti-exudation, anti-edema, anti-tumor as well as neuroprotection, and the drug side effects are mild [9] [10] [11]. At present, SA is widely used in the clinic for the treatment of diseases such as intracerebral hemorrhage, edema due to exudation after tissue

trauma, acute facial neuritis, lumbar disc herniation, herpes zoster, and venous thrombosis [12] [13]. In recent years, the study of SA in analgesia has also received industry attention. Li *et al.* [14], in their study of formalin induced pain, found that pain in rats was relieved by intrathecal administration of SA. A recent study found that SA significantly reduced the duration of licking, the number of flinches and increased paw edema in rats with neuropathic pain induced by chronic constriction injury of the sciatic nerve, showing great therapeutic effects on inflammatory pain responses [15]. However, no study has yet confirmed whether SA similarly has a mitigating effect on CINP.

Taken together, we speculate that SA has the potential to be an effective agent for the treatment of CINP. Since SA has made preliminary attempts and achieved certain effects in the direction of neuropathic pain, it will provide more substantial experimental evidence for its application in the field of neuropathic pain. In addition, since CINP incidence is highest after the use of taxanes, PAC induced peripheral neuropathic pain was adopted as a subsequent CINP research model in this study. Taking this as a hint, this study hopes to explore its alleviating effect on PAC induced neuropathic pain in rats by establishing the administration of SA to CINP rats, and to make a preliminary study on the mechanism of drug action.

2. Materials and Methods

2.1. Animal Experiment Grouping

Thirty-six SPF grade male SD rats aged 8 - 10 weeks with a body mass of 250 - 300 g were purchased from the Animal Experiment Center of Xi'an Jiaotong University. The rats were placed in animal facilities recognized by the International Laboratory and Nursing Evaluation and Certification Association, and were fed adaptively for a week using standard bedding during a 12 - 12 hour light and dark cycle at 24˚C, with free access to food and water. The rats were randomly divided into four groups according to the experimental design, including control, paclitaxel (PAC), and PAC + SA, and with 12 rats in each group. Animal experiments were approved and supervised by the Animal Ethics Committee of Shaanxi Provincial People's Hospital.

2.2. Establishment of the CINP Model

For the paclitaxel group, rats were administered PAC intraperitoneally on days 1, 3, 5, and 7 at the start of the experiment at a single dose of 2.5 mg/Kg for a total of four injections, and the final cumulative dose of paclitaxel was 10 mg/Kg. The mechanical withdrawal thresholds (MWT) and thermal withdrawal latency (TWL) were significantly reduced on the 3rd day, indicating the successful construction of CINP model. For the PAC + SA group, in addition to the intraperitoneal injection of PAC (2.5 mg/Kg intraperitoneally on days 1, 3, 5, and 7), SA 25 mg/Kg intraperitoneally was administered daily for 14 consecutive days after the start of the experiment. The same dose of saline was injected intraperito-

neally on days 1, 3, 5, and 7 in the control group. The rats were sacrificed on the 15th day of the experiment, and the spinal cords of the L4-6 group were taken for subsequent studies.

2.3. Behavioral Detection of Neuralgia in Rats

Behavioral tests were performed using von Frey filaments and thermal stimuli on rats before (day 0) and on days 3, 5, 7, 9, and 11 after the start of the experiment. Mechanical stimulus paw withdrawal thresholds (MWT) determined from Von Frey filament experiments and thermal withdrawal latency (TWL) determined from thermal stimuli experiments were used as behavioral measures. TWL: A single rat was placed in a clear glass box and allowed to acclimate in a quiet environment for 30 min. A thermal pain stimulus instrument was used to irradiate the bottom of the left hind foot of the rats, respectively, and the irradiation was stopped when the rats showed a characteristic licking or paw lifting response. Subsequently, the latency time (s), *i.e.*, TWL, of the rats in response to thermal stimuli was recorded. Each rat was tested 5 times with 5-min intervals between each session. MWT: Rats were acclimated for 30 min in a plastic box, and the right hind foot of the rats was stimulated with a Von Frey probe representing different pressures (minimum 0.4 g, maximum 26.0 g), each lasting 5 s. A rapid paw withdrawal was defined as a positive response when the rats presented, at which time the stimulus pressure was recorded. Subsequently, the MWT (g) was calculated for each rat, and each rat was tested five times with an inter stimulus interval greater than 2 min and averaged.

2.4. Immunohistochemical Staining

Spinal cord sections from rats in each group were deparaffinized, hydrated, antigen retrieval, serum blocking, NF-L (ab223343, Abcam, UK) and NF-M (ab7794, Abcam, UK) primary antibody and corresponding secondary antibodies were incubated, washed, DAB developed, hematoxylin counterstained, and the mounting of sections was performed sequentially.

2.5. HE Staining

The spinal cord tissues of rats in each group were preserved and fixed using 4% paraformaldehyde at 4˚C for one week. After tissue paraffin embedding and wax block sectioning, hematoxylin, and eosin (HE) staining (Thermo-fisher, USA) was used according to the manufacturer's requirements. Finally, slides were blocked after dehydration with ethanol, and then the staining was observed under a microscope.

2.6. Enzyme Linked Immunosorbent Assay (ELISA)

The levels of tumor necrosis factor-α (TNF-α, KALANG, USA), interleukin-1β (IL-1β, KALANG, USA), and interleukin-6 (IL-6, KALANG, USA) were measured by enzyme-linked immunosorbent assay, and the experimental procedures

were strictly performed according to the kit instructions.

2.7. RT-qPCR

The spinal cords of rats with different treatments in each group were collected, and total RNA was extracted. The cDNA was then used as template for mRNA (TNF-α, IL-1β, IL-6) amplification on a Bio-Rad CFX90 Real time PCR. GAPDH was used as an internal reference, and the relative expression levels were calculated by using the $2^{-\Delta\Delta CT}$ method.

2.8. Western Blotting

Each group of rat spinal cords was washed 2 times with cold PBS and lysed according to the application in RIPA lysis buffer (Roche Diagnostics, Basel, Switzerland) filled with protease inhibitors. The BCA method was used to perform protein quantification, followed by SDS-PAGE electrophoresis, in which 50 μg of sample was added to each well and the constant pressure was 80 V. A constant voltage of 120 V was set after 30 min for 75 - 90 min. A final constant voltage of 20 V was applied semi dry electric rotation for 1 h, and the proteins were transferred to a PVDF membrane. After blocking the membrane with 5% nonfat dry milk in PBST for 1 h, the primary antibody was incubated overnight at 4˚C. The following primary antibodies: rabbit anti-β-actin (ab8227, Abcam, UK), rabbit anti-TLR4 (ab13556, Abcam, UK), rabbit anti-NF-κB p65 (ab32536, Abcam, UK), rabbit anti-p-NF-κB p65 (ab76302, Abcam, UK). After finally washing the membrane with PBST, the membrane was incubated with secondary antibody at room temperature for 1 h, and the membrane was washed again for ECL (Pierce, Rockford, IL, USA). β-actin served as an endogenous contrast.

2.9. Statistical Analysis

Data are processed using SPSS 22.0 statistical software and presented as the mean ± SEM of results from at least three independent experiments. Differences among more than two groups in the above assays were estimated using one-way ANOVA, with $P < 0.05$ considered significant

3. Results

3.1. Analgesic Effects of Multiple Injections of SA on Established Paclitaxel Induced Neuropathic Pain

Firstly, the rats were found to have normal weight gain after SA treatment was administered in the PAC induced CINP model (**Figure 1(a)**). Subsequently, it was shown 3 days after the start of the experiment that the values of both MWT and TWL were conspicuously decreased in the paclitaxel group compared with the control group, suggesting that the simulated CINP model was constructed successfully. In addition, the values of both MWT and TWL in rats after administration of multiple injections of SA were elevated compared with the PAC group (**Figure 1(b)**, **Figure 1(c)**). These above illustrate that multiple injections of SA have analgesic effects on rats with paclitaxel induced neuropathic pain.

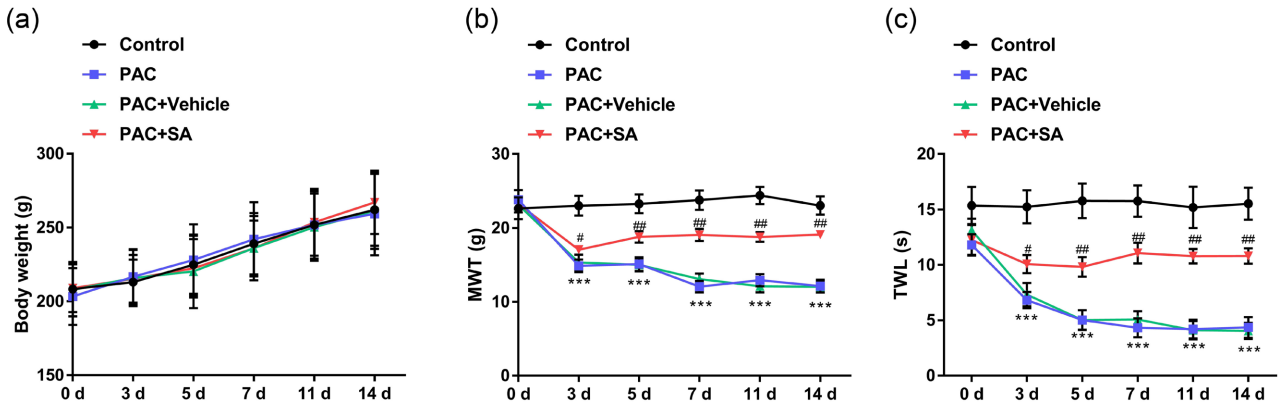

Figure 1. Analgesic effects of multiple injections of SA on established paclitaxel induced neuropathic pain. (a) The time courses of body weight in paclitaxel (PAC)-induced neuropathic pain in rats following SA treatment. (b) The time course of paclitaxel (PAC)-induced mechanical withdrawal thresholds (MWT) for neuropathic pain in rats following SA treatment. (c) The time course of paclitaxel (PAC)-induced thermal withdrawal latency (TWL) for neuropathic pain in rats following SA treatment. Data were presented as mean ± SEM. N = 10. ***$P < 0.001$ vs Control group; #$P < 0.05$, ##$P < 0.01$ vs PAC group.

3.2. SA Ameliorates Paclitaxel Induced Neuropathic Pain by Inhibiting Spinal Neurofilament Protein Expression in Rats

Next, the expression of the neurofilament proteins NF-L and NF-M in the rat spinal cord was detected using immunohistochemistry. Abnormal accumulation of neurofilament proteins in the neuronal soma or axon is known to be a hallmark of motor neuron disease and is also considered a marker of neuronal axonal degradation. The results of this study display that NF-L and NF-M are barely expressed in the spinal cord of control rats, and NF-L and NF-M expression is preeminently elevated in the PAC group (**Figure 2(a)**, **Figure 2(b)**), suggesting that axonal damage resulting from PAC may contribute to the development of pain, whereas SA injection decreases NF-L and NF-M expression and alleviates neuropathic pain induced by PAC.

3.3. SA Suppresses Paclitaxel Induced Neuroinflammation in the Rat Spinal Cord

Inflammatory factors TNF-α, IL-1β, and IL-6 have been shown to contribute to neuropathic pain, and these receptor antagonists are effective in alleviating hyperalgesia in animal models of neuropathic pain. According to this, the contents of pro-inflammatory factors of mice in each group were further analyzed in this study. RT-qPCR and ELISA results suggested that neuroinflammation was activated in the rat spinal cord after PAC repeated injections, as indicated by a significant increase in the mRNA and protein content of the proinflammatory factors. However, the contents of the above proinflammatory factors in the spinal cords of rats treated with SA repeated injection were materially downregulated (**Figure 3(a)**, **Figure 3(b)**). Further, it was observed from HE staining of rat spinal cords that the control group had an intact spinal nerve structure and no inflammatory cell infiltration, whereas the PAC Group had obvious inflamma-

tory cell invasion, which was improved by SA injection (Figure 3(c)). Taken together, these results demonstrated that SA inhibits paclitaxel induced neuroinflammation in the rat spinal cord.

3.4. SA Inhibited Paclitaxel Mediated Activation of the TLR4/NF-κB Pathway

Previous studies have proposed that activation of the TLR4/NF-κB pathway is also involved in the development of neuropathic pain resulting from PAC. The results of this study display that TLR4 and p-NF-κB protein expression, but not NF-κB total protein, were substantially upregulated in the spinal cords of rats with repeated PAC injections, whereas they were suppressed after SA injection treatment (Figure 4). These results suggest that SA may be able to target the TLR4/NF-κB pathway to ameliorate PAC-mediated neuropathic pain in the rat spinal cord by inhibiting its activation.

4. Discussion

Because of the poor selectivity of chemotherapeutic drugs, it is also inevitable to damage normal human cells while killing cancer cells, resulting in the adverse effects of drugs, among which pain is a common type of adverse effect when chemotherapy is applied [16]. At present, CIPN has become an important reason that seriously affects the survival quality of cancer patients, and how to effectively avoid or slow the occurrence of CIPN has become an urgent problem during chemotherapy [17]. Therefore, to effectively reduce pain when cancer patients are treated with chemotherapy, the rational use of analgesic drugs has become the most important clinical means for the treatment of cancer pain at

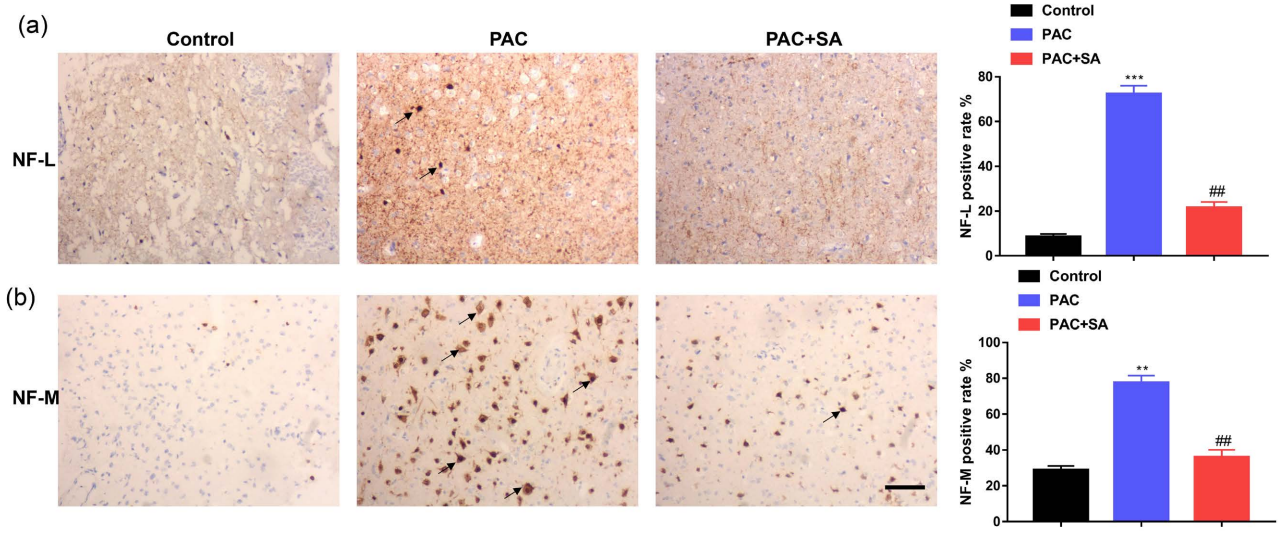

Figure 2. SA ameliorates paclitaxel induced neuropathic pain by inhibiting spinal neurofilament protein expression in rats. (a) Immunohistochemistry detection of the expression of neurofilament proteins NF-L in rat spinal cords after SA treatment. (b) Immunohistochemistry detection of the expression of neurofilament proteins NF-M in rat spinal cords after SA treatment. Scale bar: 50 μm. Data were presented as mean ± SEM. N = 10. $^{**}P < 0.01$, $^{***}P < 0.001$ vs Control group; $\#\#P < 0.01$ vs PAC group.

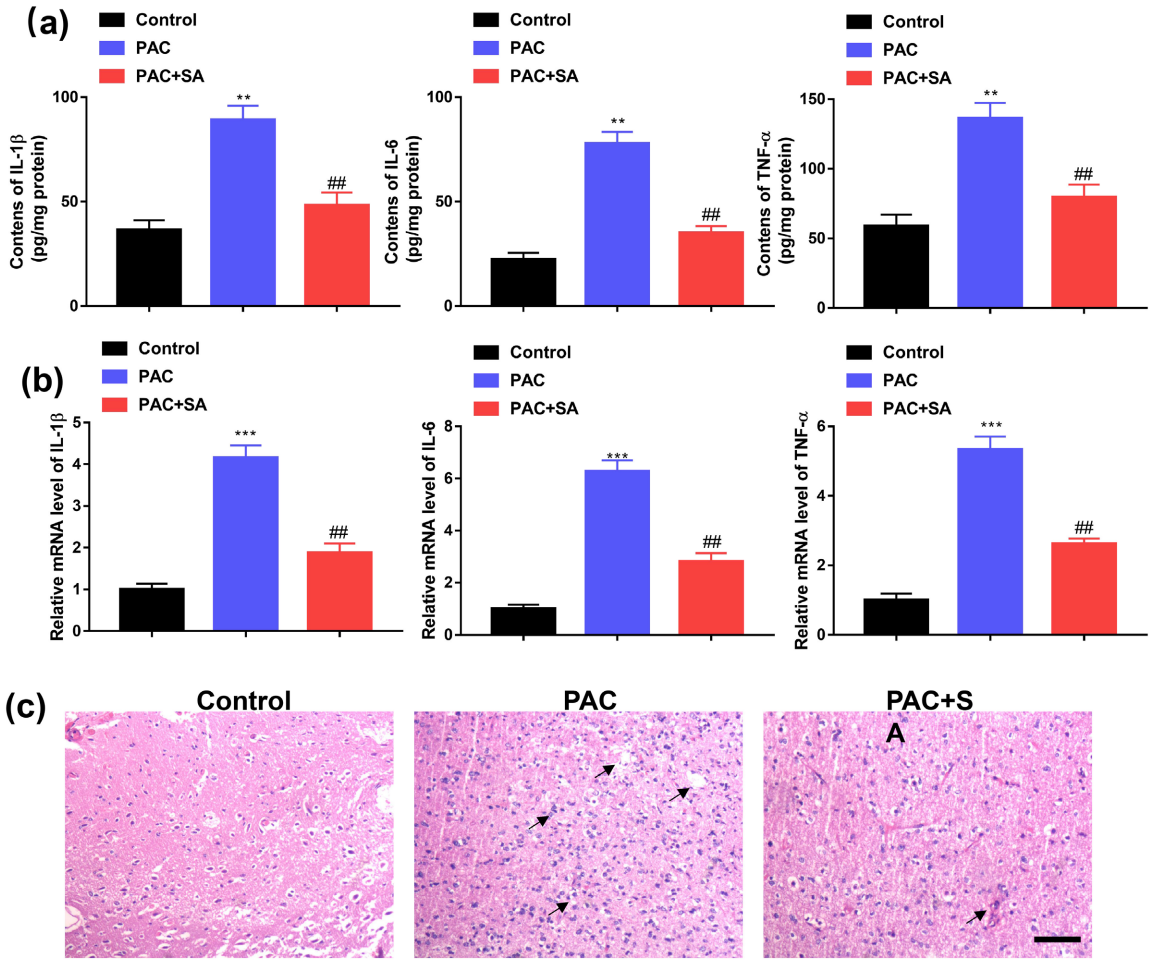

Figure 3. SA suppresses paclitaxel induced neuroinflammation in the rat spinal cord. (a) ELISA assessment of the protein contents of proinflammatory factors TNF-α, IL-1β, and IL-6 in rat spinal cords after SA treatment. (b) RT-qPCR estimate of the mRNA expression of proinflammatory factors TNF-α, IL-1β, and IL-6 in rat spinal cords after SA treatment. (c) H&E staining of spinal cords from rats in each group, with red arrows pointing to disrupted neural structures. Scale bar: 50 μm. Data were presented as mean ± SEM. N = 10. $^{**}P < 0.01$, $^{***}P < 0.001$ vs Control group; $^{\#\#}P < 0.01$ vs PAC group.

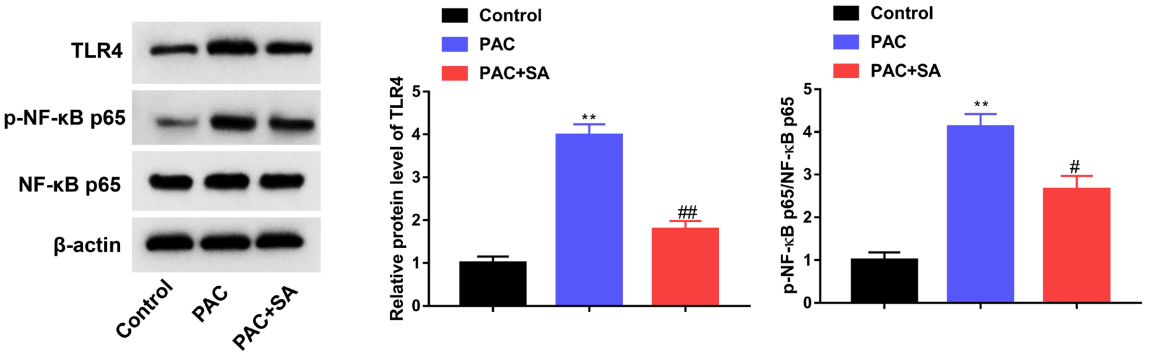

Figure 4. SA inhibited paclitaxel mediated activation of the TLR4/NF-κB pathway. Western blotting assessment of the protein levels of the TLR4/NF-κB pathway related proteins TLR4, p-NF-κB, and NF-κB in rat spinal cords after SA treatment. Data were presented as mean ± SEM. N = 10. $^{**}P < 0.01$ vs Control group; $^{\#}P < 0.05$, $^{\#\#}P < 0.01$ vs PAC group.

present. In this study, we established a CIPN rat model by referring to the dosage and manner of PAC administration commonly used in the literature, and the results showed that intraperitoneal administration of PAC at 2.5 mg/Kg every 2 days for four consecutive administrations was able to effectively induce mechanical pain behaviors in rats. However, MWT, TWL and other behaviors of rats were improved after administration of SA, suggesting that the mechanism of SA application may be associated with neuropathic pain. Previous studies have also discovered that SA is associated with multiple types of neuropathic pain. Yao *et al.* [18] demonstrated that SA can attenuate ischemia-reperfusion injury of nervous tissue due to nerve ligation and play a promoting role in the recovery of neurological function, thereby alleviating neuralgia. In addition, SA has also been confirmed to be able to exert antitumor effects in a variety of cancers [19] [20]. Subsequently, the present study further found that PAC induced neuropathic pain may be associated with its mediated disturbance of neurofilament proteins in the spinal cord. Neurofilament proteins are the major structural units of the cytoskeleton specifically expressed in neurons and their axons in the central and peripheral nervous systems [21]. Neurofilament proteins consist of light (NF-L), medium (NF-M), heavy (NF-H) proteins and α-catenin. Abnormal accumulation of neurofilament proteins in the neuronal soma or axon is a hallmark of motor neuron disease and is also considered a marker of neuronal axonal degradation [22]. The results of this study suggest that PAC treatment of rats significantly elevated NF-L, NF-M expression in spinal cord tissue, suggesting that axonal damage resulting from PAC may contribute to the development of pain, whereas administration of SA treatment reduced NF-L, NF-M expression and alleviated neuropathic pain induced by PAC. The latest studies have also shown that the levels of NF-L are markedly elevated in the serum of patients with chronic pain following rehabilitation from COVID-19, and NF-L may serve as a potential biomarker for persistent neuropathic pain on COVID-19 [23]. Furthermore, Yang *et al.* [24] found that the levels of NF-L polypeptide were elevated in a pain model constructed by spinal nerve ligation in rats, and miR-7a could block transcriptional signaling pathway activators by inhibiting NF-L polypeptide and then ameliorate neuropathic pain.

Cytokines are proteins that regulate systemic immune responses. Cytokines involved in innate immune signaling have also been identified as key players driving the pathophysiology of CIPN [25]. Cytokines are released not only by immune cells but also by glia and neurons [26]. They can directly or indirectly act on primary afferent fibers, dorsal root ganglia, and spinal dorsal horn neurons and are involved in the development and progression of inflammatory, cancerous pain, and morphine tolerance [27]. The production and release of proinflammatory factors, which can be conspicuously increased by chemotherapeutic drugs, is considered to be one of the main mechanisms regulating CIPN [28]. The results of this study show that the levels of the proinflammatory cytokines are remarkably upregulated in the PAC administered group. However, the con-

tents of the above proinflammatory factors in the spinal cords of rats treated with SA repeated injection were materially downregulated.

TLR4 in the spinal cord plays an important role in models of neuropathic pain and nerve injury [29]. Studies have shown that chemotherapeutic agents are responsible for the activation of TLR4 leading to increased proinflammatory cytokine expression in the peripheral and central nervous systems [30]. The activation of TLR4 is ultimately manifested in the activation of nuclear factor κB (NF-κB), which promotes inflammatory factor and chemokine synthesis and secretion, initiating acquired immune responses [31]. Chang *et al.* [32] showed that kaempferol treatment strikingly reduced neuropathic pain and proinflammatory cytokine production, and attenuated TLR4/NF-κB pathway activation. Wang *et al.* [33] suggested that dexmedetomidine could alleviate neuropathic pain and reduce inflammatory responses by preventing the activation of TLR4/NF-κB p65 pathway. These above suggest that inhibiting the activation of TLR4/NF-κB pathway may be an effective drug target for alleviating neuropathic pain. However, whether the TLR4/NF-κB p65 signaling pathway is expressed and functions in the PAC induced CIPN model remains unknown. The results of the present study are consistent with previous findings and that SA can restrain the TLR4/NF-κB p65 pathway to ameliorate PAC mediated neuropathic pain in the rat spinal cord.

5. Conclusion

The results of the present study preliminarily indicate that SA has an analgesic effect on rats with CIPN induced by PAC injection, and the mechanism may be related to blocking the TLR4/NF-κB signaling pathway, inhibiting the expression of proinflammatory factors, and alleviating cytoskeletal disorders. This study provides an initial exploration of the therapeutic effects of SA on PAC induced CIPN. However, the experimental content is relatively insufficient, and more experimental data are needed next to do further validation and refine the relevant mechanism of action studies of the drug.

Funding

This work was supported by grants from the Xi'an Innovation Ability Strong Foundation Program—Medical Research Project (22YXYJ0107).

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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Scientific Research Publishing

Yangtze Medicine, 2023, 7, 147-153
https://www.scirp.org/journal/ym
ISSN Online: 2475-7349
ISSN Print: 2475-7330

Comparative Observation on Nursing Effect of Nursing Intervention and Routine Nursing in Patients with Renal Calculi and Gastric Ulcer and the Impacts on Epidermal Growth Factor

Qianping Jiang, Yu Sang*

Department of Urology Surgery, The First Affiliated Hospital of Yangtze University, Jingzhou, China
Email: *77150681@qq.com

How to cite this paper: Jiang, Q.P. and Sang, Y. (2023) Comparative Observation on Nursing Effect of Nursing Intervention and Routine Nursing in Patients with Renal Calculi and Gastric Ulcer and the Impacts on Epidermal Growth Factor. *Yangtze Medicine*, **7**, 147-153.
https://doi.org/10.4236/ym.2023.72014

Received: November 18, 2022
Accepted: June 27, 2023
Published: June 30, 2023

Abstract

Objective: To explore the comparative observation on nursing effect of nursing intervention and routine nursing in patients with renal calculi and gastric ulcer and the impacts on epidermal growth factor. **Methods:** A total of 72 patients with renal calculi and gastric ulcer were selected and treated in our hospital from January 2018 to December 2018. They were divided into the observation group and the control group, 36 for each. Comprehensive nursing intervention was implemented in the observation group, whereas routine nursing was implemented in the control group. The level of epidermal growth factor, nursing satisfaction, renal calculi recurrence rate, average hospital stay and postoperative blood loss were compared between the two groups after nursing. **Results:** There was no significant difference in the level of epidermal growth factor between the two groups before nursing ($P > 0.05$), while after nursing, the level in the observation group was higher compared with the control group, and the difference between the two groups was significant ($P < 0.05$). Similarly, in comparison with the control group, the nursing satisfaction score of the observation group was higher, the average hospital stay was shorter, the postoperative blood loss was less, and the calculi recurrence rate was lower. The between-group differences for all mentioned indexes were significant ($P < 0.05$). **Conclusion:** With regard to patients with renal calculi and gastric ulcer, comprehensive nursing intervention can improve nursing satisfaction and quality of patients' lives, reduce calculi recurrence rate, and increase the level of epidermal growth factor, which has clinical application value.

Keywords

Nursing Intervention, Epidermal Growth Factor, Renal Calculi, Gastric Ulcer

1. Introduction

Gastric ulcer (GU) is a chronic ulcer that occurs between the cardia and pylorus, and is a type of peptic ulcer. At present, it has developed into a ubiquitous disease clinically, with the characteristics of long course and slow development. The disease is associated with the patient's living rules, food habits, psychological factors, etc. It is difficult to achieve a complete cure in a short period of time, which will have a certain impact on its treatment [1]. In recent years, people have gradually attaches importance to the applied research on the nursing of renal calculi. In this study, comprehensive nursing intervention was adopted for patients with renal calculi and gastric ulcer. The results are as follows.

2. Materials and Methods

2.1. Case Information

A total of 72 patients with renal calculi and gastric ulcer were selected and treated in our hospital from January 2018 to December 2018. They were divided into the observation group and the control group, 36 for each. Comprehensive nursing intervention was adopted for patients in the observation group, whereas routine nursing was adopted for those in the control group. The level of epidermal growth factor, nursing satisfaction, calculi recurrence rate, average hospital stay and postoperative blood loss were compared between the two groups after nursing. All participants provided signed informed consent. Those with high blood pressure and high blood sugar were excluded from the experiment. Patients in the observation group (20 males and 16 females) aged from 30 to 45 years, with an average age of 42.2 ± 1.1 years. Their average disease duration was 3.48 ± 1.43 years, and the average weight was 65.25 ± 5.54 kg. As their counterparts, patients in the control group (19 males and 17 females) aged from 31 to 46 years, with an average age of 41.8 ± 1.3 years. Their average disease duration was 3.52 ± 1.26 years, and the average weight was 65.21 ± 5.39 kg. There was no statistical difference in case data between the two groups.

2.2. Inclusion and Exclusion Criteria

Inclusion criteria: 1) Patients aged 18 years or older; 2) Informed consent can effectively cooperate with therapeutic intervention patients; 3) Patients diagnosed with kidney stones and gastric ulcers. Exclusion Criteria: 1) Patients with hypertension; 2) patients with diabetes; 3) patients with mental disorders; 4) Severe loss of renal function, severe heart disease patients. All patients signed informed consent forms. The study was approved by the ethics committee officer.

2.3. Methods

Two different nursing modes were implemented in the two groups, underpinned by the basic treatment of gastric mucosal protective agents and proton pump inhibitors for both groups.

2.3.1. The Control Group

Routing nursing was adopted. Starting from the time of admission, the relevant disease diagnosis and knowledge education were carried out. First of all, it was necessary to encourage patients to fully understand the basic knowledge about the disease, and comprehensively educate patients about the knowledge of renal calculi and gastric ulcer. Secondly, we tried to change their long-standing psychological state of fear and anxiety, and implemented health education. Finally, we had to encourage patients to quit smoking, live life in a scientific and reasonable way, and do appropriate physical exercise.

2.3.2. The Observation Group

Comprehensive nursing intervention

Before surgery, patients were excluded from surgical contraindications, and various routine examinations were implemented. In order to ensure that reasonable nutrition could be provided to patients, a special diet plan was formulated according to their own conditions. It is necessary to obtain the expected effect of surgical treatment before surgery, and implement psychological care according to the patient's own character to avoid postoperative emergencies [2]. ECG monitoring was provided to the patient after surgery. Various physical indexes and blood oxygen saturation of the patient were detected. Medicinal water was guaranteed to be 37˚C, and the operating room temperature was about 29˚C. During postoperative nursing, the vital signs of patients should be monitored, and their underlying conditions should be closely observed [3]. It was necessary to actively prevent the occurrence of related complications. When complications occurred, corresponding measures should be taken to deal with them immediately. Additionally, as for the physical condition of patients after surgery, there was a need to give health guidance for them, and adjust the diet plan in time, so as to prevent the occurrence of complications.

Medication guidance

During the treatment, acidic foods and dairy products should be banned for patients who were not treated with antacids. Taking the relevant drugs before meals were exactly those taking bismuth therapy [4].

Psychological care

Due to the long course of the disease, it was more difficult to obtain a complete cure. Patients were therefore lack of strong confidence in the treatment and had low enthusiasm, which led to the emergence of anxiety, depression and other negative emotions, so it was necessary to encourage patients, step up psychological intervention for patients, and increase their confidence in treatment.

2.4. Observation Indexes

The level of epidermal growth factor, satisfaction, calculi recurrence rate, nursing satisfaction, average hospital stay, postoperative blood loss were compared between the two groups after nursing, postoperative rehabilitation effect. The Likert level 5 scoring method is adopted, with the nurse's service attitude and the

attention to patients as the evaluation indicators. Among them, 0 points for very dissatisfied, 1 point for dissatisfaction, generally 2 points, 3 points for satisfaction, and 4 points for satisfaction. The total score of all items is transformed into an overall "feeling score" of 0 - 100 points, the higher the score, the higher the satisfaction. The percentage conversion formula is: the sum of the scores of effective answer items/number of effective answer items/4 × 100 [5].

2.5. Statistical Analyses

SPSS23.0 Software was utilized for data analysis. All data used were analyzed after being processed by Epidata. A value of 0.05 was the test standard, and t test was used for comparison between groups. A value of $P < 0.05$ was considered statistically significant.

3. Results

3.1. Comparison of the Level of Epidermal Growth Factor between the Two Groups

Before nursing, there was no significant difference in the level of epidermal growth factor between the two groups ($P > 0.05$), while after nursing, the level in the observation group was higher compared with the control group, and the difference between the two groups was significant ($P < 0.05$). See Table 1.

3.2. Comparison of Nursing Satisfaction between the Two Groups

Compared with the control group, the nursing satisfaction score of the observation group was higher, and the difference between the two groups was significant ($P < 0.05$), as shown in Table 2.

3.3. Comparison of the Average Hospital Stay and Postoperative Blood Loss between the Two Groups

Compared with the control group, the average hospital stay of the observation group was shorter, and the postoperative blood loss was less. The differences between the two groups were significant ($P < 0.05$), as shown in Table 3.

3.4. Comparison of Renal Calculi Recurrence Rate between the Two Groups

The renal calculi recurrence rate of the observation group was lower than that of the control group, and the difference between the two groups was significant ($P < 0.05$). See Table 4.

Table 1. Comparison of the level of epidermal growth factor between the two groups.

	Number of cases	Before nursing	After nursing
The observation group	36	248.33 ± 29.57	477.77 ± 65.24
The control group	36	250.14 ± 31.03	580.24 ± 71.14

Table 2. Comparison of nursing satisfaction score between the two groups (x ± s, point).

	Number of cases	Score
The observation group	36	95.26 ± 2.10
The control group	36	80.02 ± 3.44

Table 3. Comparison of the average hospital stay and postoperative blood loss between the two groups (x ± s).

	Number of cases	Average hospital stay (d)	Postoperative blood loss (mL)
The observation group	36	7.12 ± 1.24	30.01 ± 3.31
The control group	36	11.02 ± 2.47	62.35 ± 5.35

Table 4. Comparison of calculi recurrence rate between the two groups [n (%)].

	Number of cases	Renal calculi recurrence rate (%)
The observation group	36	2 (5.56)
The control group	36	9 (25.00)

4. Discussion

Addiction to tobacco and alcohol, irregular diet, mental stress and other factors can cause the occurrence of chronic gastric ulcer in patients [6]. Because patients often do not understand its inducing factors and lack of the knowledge about the disease, the cure of the disease is delayed in the treatment. However, patients do not pay attention to change lifestyle after the cure, so there is no significant improvement in the inducing factors, resulting in repeated attacks of the disease. In addition, it is relatively difficult to treat canal calculi clinically. Surgical treatment can easily induce renal failure and cause great damage to patients [2]. In the current stage, extracorporeal shock wave therapy is a satisfactory means. The concept of fast rehabilitation has gradually gained attention in surgical nursing work in recent years [3], and in perioperative patient care, fast rehabilitation nursing can shorten the length of hospital stay and accelerate recovery, which plays an extremely critical role [4].

Additionally, renal calculi combined with gastric ulcer often occurs in clinic, with the characteristics of being difficult to treat and slow onset, so it is emphasized to do nursing work from various aspects. It is necessary to be comprehensive. In clinical practice, nursing intervention is a commonly used method, which can provide patients with better and comprehensive nursing services [7]. Preoperative intervention can make good preoperative preparations to effectively prevent the occurrence of postoperative emergencies, and ensure patients to receive treatment in the best state. Postoperative intervention can reduce the disease recurrence rate, reduce the occurrence of complications, and ensure the efficacy of surgery. In this study, all the indexes that we investigated in the obser-

vation group were better than those in the control group. The results of this study have confirmed that helicobacter pylori infection and imbalance of pepsin secretion are the main factors for the disease in patients with renal calculi and gastric ulcer. The main use of the comprehensive nursing intervention can somewhat increase the level of epidermal growth factor, improve nursing satisfaction, reduce renal calculi recurrence rate, and improve the quality of life of patients.

5. Conclusion

In a nut shell, traditional clinical nursing plan adopts the "disease-centered" nursing mode, and the nursing staff is often difficult to deal with the various needs of patients, which leads to poor nursing quality, which increases the incidence of medical disputes. Comprehensive nursing intervention is a pertinent nursing mode with "patient-centered". When different patients appear untimely due to various factors, they adopt an individualized and humanized nursing method to help patients reduce pain and improve the quality of nursing. If the sample size is small, it is suggested to adjust the sample screening criteria to expand the sample size and scope. For patients with renal calculi and gastric ulcer, the comprehensive nursing intervention can bring a series of better effects, which has clinical application value.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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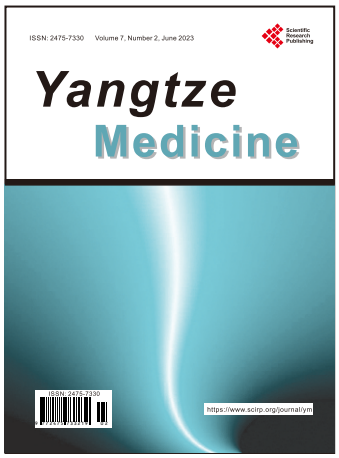

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