

基于SEER数据库建立和验证甲胎蛋白阴性肝细胞癌患者 癌症特异生存期的列线图

余孝鹏¹, 杨仁义¹, 贺佐梅², 曾普华^{2,3}

(1. 湖南中医药大学研究生院, 湖南 长沙 410208; 2. 湖南中医药研究院附属医院肿瘤二科, 湖南 长沙 410006; 3. 湖南中医药研究院中医肿瘤研究所, 湖南 长沙 410006)

[摘要] **目的:** 探讨甲胎蛋白(AFP)阴性肝细胞癌(HCC)患者预后相关因素, 构建列线图以预测患者生存时间。**方法:** 回顾性分析监测、流行病学和最终结果(SEER)数据库提取的2 064例AFP阴性HCC患者数据, 将所有患者按7:3比例随机分为训练集和内部验证集, 以湖南省中西医结合医院101例AFP阴性HCC患者作为外部验证集。将单因素Cox回归分析结果纳入多因素分析, 采用多因素Cox分析获得AFP阴性HCC患者的独立危险因素, 构建AFP阴性HCC患者癌症特异生存(CSS)预后列线图。采用时间依赖受试者工作特征曲线(ROC)、校准图和决策曲线分析(DCA)评估列线图的预测效能和临床实用性。将列线图所得总分进行风险分层, 比较列线图和美国癌症联合委员会(AJCC)分期系统的风险区分程度。**结果:** 采用多因素Cox回归分析筛选出10个独立危险因素, 构建AFP阴性HCC患者3、4和5年CSS预后列线图, 包括患者年龄、病理分级、手术情况、放疗情况、化疗情况、肺转移情况、肿瘤大小、肿瘤T分期、肿瘤M分期和婚姻状况。3、4和5年时间依赖ROC曲线下面积(AUC), 训练集分别为0.807(95%CI: 0.786~0.828)、0.804(95%CI: 0.782~0.826)和0.813(95%CI: 0.790~0.835), 内部验证集分别为0.776(95%CI: 0.743~0.810)、0.772(95%CI: 0.737~0.808)和0.789(95%CI: 0.752~0.826), 外部验证集分别为0.773(95%CI: 0.677~0.868)、0.746(95%CI: 0.620~0.872)和0.736(95%CI: 0.577~0.895)。校准图验证列线图能够很好地拟合到完美曲线上。DCA曲线显示列线图在某特定概率阈值上的净收益明显高于AJCC分期在某特定概率时净收益。与AJCC分期比较, 列线图具有较好的识别高风险人群的能力。**结论:** 血清中AFP表达是HCC患者的预后指标之一, 对于部分血清中AFP阴性表达的HCC患者应区别对待, 基于多个风险因素建立的列线图模型有望成为临床评估AFP阴性HCC患者CSS的有效工具。

[关键词] 甲胎蛋白阴性肝细胞癌; 列线图; 癌症特异生存; 预测模型; 生存时间

[中图分类号] R735.7 **[文献标志码]** A

Establishment and validation of nomogram of cancer specific survival of patients with hepatocellular carcinoma with negative alpha fetoprotein based on SEER Database

YU Xiaopeng¹, YANG Renyi¹, HE Zuomei², ZENG Puhua^{2,3}

[收稿日期] 2023-02-16

[基金项目] 国家自然科学基金项目(82074425, 82205227); 湖南省科技厅重点研发项目(2021SK2006); 湖南省科技厅自然科学基金项目(2021JJ30417); 湖南省科技厅自然科学基金青年基金项目(2021JJ40310); 湖南省中医药管理局青年基金项目(2021176)

[作者简介] 余孝鹏(1997-), 男, 广东省汕尾市人, 在读硕士研究生, 主要从事中西医结合防治恶性肿瘤方面的研究。

[通信作者] 贺佐梅, 主治医师(E-mail: 281144800@qq.com);

曾普华, 教授, 主任医师, 博士研究生导师(E-mail: zph120@126.com)

- (1. Graduate School, Hunan University of Chinese Medicine, Changsha 410208, China;
2. Department of Tumor, Affiliated Hospital, Hunan Academy of Traditional Chinese Medicine, Changsha 410006, China; 3. Institute of Traditional Chinese Medicine Oncology, Hunan Academy of Traditional Chinese Medicine, Changsha 410006, China)

ABSTRACT Objective: To discuss the factors related to the prognosis in the alpha fetoprotein (AFP) negative hepatocellular carcinoma (HCC) patients, and to construct the nomogram for predicting the survival time of the patients. **Methods:** The retrospective analysis on data of 2 064 cases of AFP negative HCC patients extracted from the Surveillance, Epidemiology, and End Results (SEER) Database was conducted, and all the patients were divided into training cohort and internal validation cohort at a ratio of 7:3, and 101 AFP negative HCC patients from the Integrated Traditional Chinese and Western Medicine Hospital in Hunan Province were regarded as the external validation cohort. The univariate Cox regression analysis results were incorporated into the multivariate analysis, and the independent risk factors for the AFP negative HCC patients were obtained by multivariate Cox analysis to build a cancer specific survival (CSS) prognosis nomogram for the AFP negative HCC patients. The predictive efficacy and clinical utility of the nomogram were evaluated by time-dependent receiver operating characteristic curve (ROC), calibration plots, and decision curve analysis (DCA). The total score obtained from the nomogram was used for the risk stratification to compare the degree of risk discrimination between the nomogram and the American Joint Committee on Cancer (AJCC) staging system. **Results:** Ten independent risk factors were selected by multivariate Cox regression analysis to construct 3-year, 4-year, and 5-year CSS prognostic nomograms for the AFP negative HCC patients, including the patient's age, pathological grade, surgical status, radiotherapy status, chemotherapy status, lung metastasis status, tumor size, tumor T stage, tumor M stage, and marital status. The area under curve (AUC) for the 3-year, 4-year, and 5-year time-dependent ROC in the training cohort were 0.807 (95% CI: 0.786–0.828), 0.804 (95% CI: 0.782–0.826), and 0.813 (95% CI: 0.790–0.835), respectively. In the internal validation cohort, they were 0.776 (95% CI: 0.743–0.810), 0.772 (95% CI: 0.737–0.808), and 0.789 (95% CI: 0.752–0.826), and in the external validation cohort, they were 0.773 (95% CI: 0.677–0.868), 0.746 (95% CI: 0.620–0.872), and 0.736 (95% CI: 0.577–0.895). The calibration plots verified that the nomogram fitted well with the perfect line. The DCA curve revealed that the net benefit of the nomogram was significantly higher than that of the AJCC staging system at certain probability thresholds compared with AJCC staging, the nomogram had a better ability to identify high-risk individuals. **Conclusion:** The serum AFP expression is one of the prognostic markers for the HCC patients. For those patients with AFP negative expression in serum, different considerations should be taken. The nomogram model based on multiple risk factors is a promising clinical tool for assessing the CSS in the AFP negative HCC patients.

KEYWORDS Hepatocellular carcinoma with negative alpha fetoprotein; Nomogram; Cancer specific survival; Prediction model; Survival time

肝细胞癌 (hepatocellular carcinoma, HCC) 是最常见的消化系统肿瘤之一, 占肝脏恶性肿瘤的75%, 在全球常见癌症中排第6位, 在癌症相关死亡原因中排第4位^[1-2]。血清甲胎蛋白 (alpha fetoprotein, AFP) 是诊断HCC可靠的肿瘤生物标志物, 与HCC复发、血管侵犯、转移和预后密切相关^[3-4]。血清AFP水平 $>400 \mu\text{g}\cdot\text{L}^{-1}$ 是诊断

HCC的指征之一^[5]。AFP水平越高, HCC患者预后越差、肿瘤侵袭性越高且治疗效果也越差^[6]。有30%~40%的HCC患者血清AFP为阴性表达, 即血清AFP水平 $<20 \mu\text{g}\cdot\text{L}^{-1}$, 该种情况可能会影响HCC患者的早期诊断、治疗和预后的判断^[7]。研究^[8-10]表明: AFP阴性HCC患者的肿瘤一般较小, 分化更好, 术后复发率和生存率优于AFP阳性患

者。但血清AFP是HCC临床筛查、诊断、复发和预后监测的主要生物标志物,因此AFP阴性HCC患者的预后预判应区别对待^[11-12]。目前对于AFP阴性HCC患者预后风险的相关研究较少,因此本研究通过分析影响AFP阴性HCC患者的危险因素,并以此建立预测癌症特异生存(cancer specific survival, CSS)列线图,以便临床工作者对AFP阴性HCC患者进行正确的预后判断。

1 资料与方法

1.1 病例筛选

采用SEER*Stat 8.4.0.1软件,从监测、流行病学和最终结果(Surveillance, Epidemiology, and End Results, SEER)数据库中筛选确诊的HCC患者,所有患者均来自美国。纳入标准:①组织学代码为8170/8171/8172/8173/8174/8175;②AFP阴性患者;③具有明确的生存状态和生存时间。排除标准:①缺少有关年龄、性别、婚姻状况、基于美国癌症联合委员会(American Joint Committee on Cancer, AJCC)标准的肿瘤分期、肿瘤大小、手术类型、放疗信息及骨和肺转移信息;②患者在1个月内死亡或自初步诊断后不到1个月内接受随访;③其他原因死亡或死因不明。基于SEER数据库病例筛选的独立危险因素,纳入湖南省中西医结合医院2012~2022年期间确诊为AFP阴性HCC患者101例作为外部验证。纳入标准:①确诊为HCC;②血清检测AFP为阴性,即 $AFP < 20 \mu\text{g}\cdot\text{L}^{-1}$ 。排除标准:①缺少年龄、病理分级、手术、放疗、化疗、肺转移、T分期、M分期、肿瘤大小(肿瘤最大直径)和婚姻状况信息的患者;②并发其他组织恶性肿瘤的患者;③非肿瘤疾病引起死亡的患者;④在1个月内死亡或自初步诊断后1个月内接受随访的患者;⑤失访的患者。

1.2 训练集和验证集的确定及筛选变量

采用R软件将2604例患者按7:3比例分为训练集和内部验证集,以确保结果事件在两个队列之间随机分布。训练集用于筛选变量和建立模型,内部验证集用于验证模型。从SEER数据库中筛选13个变量,包括患者年龄(诊断时)、性别、病理分级、肿瘤T分期、肿瘤N分期、肿瘤M分期、手术情况、放疗情况、化疗情况、骨转移情况、肺转移情况、肿瘤大小和婚姻状况。基于独立危险因素,

者作为外部验证集,纳入10个变量进行外部验证:患者年龄、病理分级、手术情况、放疗情况、化疗情况、肺转移情况、肿瘤T分期、肿瘤M分期、肿瘤大小和婚姻状况。

1.3 统计学分析

采用R软件进行统计学分析。患者年龄和肿瘤大小的最佳截断值通过受试者工作特征曲线(receiver operating characteristic curve, ROC)进行计算,组间比较采用卡方检验;训练集病例采用单因素和多因素Cox回归分析,并计算相应的风险比(hazard ratio, HR)及其95%CI。将单因素Cox回归分析所得到的危险因素纳入多因素Cox回归分析中,多因素Cox回归分析得出独立危险因素纳入列线图的构建;采用风险评分计算公式计算列线图的风险评分,通过时间依赖ROC曲线下面积(area under curve, AUC)评估列线图的判别能力,采用Calibration校准曲线评价列线图模型的效能和校准度。通过决策曲线分析(decision curve analysis, DCA)^[13]评估列线图的临床净收益。采用X-tile软件^[14]对列线图总分进行风险分层,采用Kaplan-Meier分析比较列线图和AJCC肿瘤分期的风险分层。以 $P < 0.05$ 为差异有统计学意义。

2 结果

2.1 AFP阴性HCC患者的基线资料

从SEER数据库中筛选2604例符合条件的AFP阴性HCC患者,按7:3比例随机分为训练集($n=1822$)和内部验证集($n=782$)。患者的基线资料见表1。

2.2 训练集中AFP阴性HCC患者CSS的独立预后因素

采用单因素和多因素Cox回归分析,确定训练集中AFP阴性HCC患者的独立预后因素。单因素Cox分析结果显示:患者年龄、性别、病理分级、手术情况、放疗情况、化疗情况、骨转移情况、肺转移情况、肿瘤大小、婚姻状况、肿瘤T分期和肿瘤M分期是AFP阴性HCC患者的独立预后因素($P < 0.05$)。多因素Cox分析结果显示:患者年龄、病理分级、手术情况、放疗情况、化疗情况、肺转移情况、肿瘤大小、肿瘤T分期和肿瘤M分期是影响HCC患者CSS的独立预后因素($P < 0.05$)。见图1。

2.3 构建AFP阴性HCC患者CSS预后列线图

基于训练集多因素Cox回归分析结果,用训练

表1 训练集和内部验证集 AFP 阴性 HCC 患者的基线资料

Tab. 1 Basic data of AFP negative HCC patients in training cohort and internal validation cohort [n(η/%)]

Characteristic	Training (n=1 822)	Internal validation (n=782)	χ^2	P	Characteristic	Training (n=1 822)	Internal validation (n=782)	χ^2	P
Age(year)			0.58	0.450	Lung metastasis			0.20	0.650
<75	1 413 (77.55)	617 (78.90)			Yes	30 (1.65)	11 (1.41)		
≥75	409 (22.45)	165 (21.10)			No	1 792 (98.35)	771(98.59)		
Gender			2.56	0.110	Tumor size			3.45	0.180
Female	420 (23.05)	158 (20.20)			<38 mm	770 (42.26)	361(46.16)		
Male	1 402 (76.95)	624 (79.80)			38-82 mm	714 (39.19)	283(36.19)		
Grade			2.30	0.470	>82 mm	338 (18.55)	138(17.65)		
Grade I	771 (42.32)	312 (39.90)			Marital status			1.32	0.720
Grade II	865 (47.48)	383 (48.98)			Divorce	206 (11.31)	77 (9.85)		
Grade III	174 (9.55)	84 (10.74)			Married	1 119 (61.42)	490(62.66)		
Grade IV	12 (0.66)	3 (0.38)			Single	339 (18.61)	144(18.41)		
Surgery			0.02	0.900	Widowed	158 (8.67)	71 (9.08)		
Yes	1 076 (59.06)	464 (59.34)			T stage			2.39	0.500
No	746 (40.94)	318 (40.66)			T1	1 098 (60.26)	475(60.74)		
Radiotherapy			2.07	0.150	T2	408 (22.39)	183(23.40)		
Yes	164 (9.00)	57 (7.29)			T3	269 (14.76)	111(14.19)		
No	1 658 (91.00)	725 (92.71)			T4	47 (2.58)	13 (1.66)		
Chemotherapy			0.11	0.740	N stage			1.35	0.250
Yes	612 (33.59)	268 (34.27)			N0	1 764 (96.82)	750 (95.91)		
No	1 210 (66.41)	514 (65.73)			N1	58 (3.18)	32 (4.09)		
Bone metastasis			0.09	0.770	M stage			0.81	0.370
Yes	26 (1.43)	10 (1.28)			M0	1 737 (95.33)	739 (94.50)		
No	1 796 (98.57)	772 (98.72)			M1	85 (4.67)	43 (5.50)		

集病例构建 AFP 阴性 HCC 患者 3、4 和 5 年 CSS 预后列线图 (图 2), 主要由以下独立危险因素组成: 患者年龄、病理分级、手术情况、放疗情况、化疗情况、肺转移情况、肿瘤大小、肿瘤 T 分期、肿瘤 M 分期和婚姻状况。列线图中对应于每个变量的线的长度表示预测变量对生存结果的贡献值。

2.4 列线图的验证

基于训练集病例构建列线图, 采用内部验证集和外部验证集进行内部验证和外部验证^[15-16]。

2.4.1 外部验证集病例基线资料 选择 2012—2022 年于湖南省中西医结合医院确诊为 APF 阴性 HCC 患者 101 例作为外部验证集, 其基线资料见表 2。

2.4.2 时间依赖 ROC 曲线验证 AFP 阴性 HCC 患者 CSS 预后列线图的预测性能 采用时间依赖 ROC 曲线评估 CSS 预后列线图在不同组集中的预测性能。在本研究中, 训练集 3、4 和 5 年时间依赖

ROC 曲线 AUC 分别为 0.807 (95%CI: 0.786~0.828)、0.804 (95%CI: 0.782~0.826) 和 0.813 (95%CI: 0.790~0.835), 内部验证集 3、4 和 5 年时间依赖 ROC 曲线 AUC 分别为 0.776 (95%CI: 0.743~0.810)、0.772 (95%CI: 0.737~0.808) 和 0.789 (95%CI: 0.752~0.826), 外部验证集 3、4 和 5 年时间依赖 ROC 曲线 AUC 分别为 0.773 (95%CI: 0.677~0.868)、0.746 (95%CI: 0.620~0.872) 和 0.736 (95%CI: 0.577~0.895)。AJCC 分期 3、4 和 5 年时间依赖 ROC 曲线 AUC, 训练集分别为 0.609 (95%CI: 0.587~0.621)、0.604 (95%CI: 0.562~0.644) 和 0.605 (95%CI: 0.571~0.652), 内部验证集分别为 0.624 (95%CI: 0.588~0.661)、0.623 (95%CI: 0.586~0.660) 和 0.614 (95%CI: 0.574~0.654), 外部验证集分别为 0.582 (95%CI: 0.461~0.731)、0.676 (95%CI: 0.539~0.846)

Characteristic	HR(95% CI) (Univariate analysis)	P (Univariate analysis)	HR(95% CI) (Multivariate analysis)	P (Multivariate analysis)
Age		<0.001		
<75	0.523 (0.458-0.597)	<0.001	0.726 (0.628-0.839)	<0.001
≥75	1.912 (1.676-2.181)	<0.001	1.377 (1.191-1.592)	<0.001
Gender		0.869		
Male	0.988 (0.857-1.139)	0.869	-	-
Female	1.012 (0.878-1.166)	0.869	-	-
Grade		<0.001		
Grade I	Reference			
Grade II	0.895 (0.787-1.017)	0.088	1.101 (0.965-1.256)	0.154
Grade III	1.448 (1.185-1.770)	<0.001	1.622 (1.312-2.006)	<0.001
Grade IV	3.038 (1.668-5.534)	<0.001	2.707 (1.474-4.971)	0.001
Surgery		<0.001		
Yes	0.236 (0.208-0.268)	<0.001	4.186 (3.591-4.880)	<0.001
No	4.233 (3.735-4.797)	<0.001	0.239 (0.205-0.278)	<0.001
Radiation		<0.001		
Yes	1.781 (1.475-2.149)	<0.001	0.783 (0.635-0.964)	0.021
No	0.562 (0.465-0.678)	<0.001	1.277 (1.037-1.574)	0.021
Chemotherapy		<0.001		
Yes	1.590 (1.407-1.798)	<0.001	0.744 (0.644-0.858)	<0.001
No	0.629 (0.556-0.711)	<0.001	1.345 (1.165-1.552)	<0.001
Bone metastasis		<0.001		
Yes	3.995 (2.681-5.952)	<0.001	1.007 (0.606-1.673)	0.979
No	0.250 (0.168-0.373)	<0.001	0.993 (0.598-1.650)	0.979
Lung metastasis		<0.001		
Yes	9.050 (6.257-13.088)	<0.001	2.306 (1.452-3.664)	<0.001
No	0.111 (0.076-0.160)	<0.001	0.434 (0.273-0.689)	<0.001
Tumor size		<0.001		
<38	Reference			
38-82	1.739 (1.513-1.998)	<0.001	1.274 (1.098-1.477)	0.001
>82	2.601 (2.214-3.057)	<0.001	1.622 (1.350-1.949)	<0.001
Marital status		<0.001		
Married	Reference			
Single	1.345 (1.111-1.629)	0.002	1.194 (0.981-1.452)	0.076
Divorced	1.343 (1.151-1.567)	<0.001	1.277 (1.089-1.498)	0.003
Widowed	1.664 (1.364-2.030)	<0.001	1.119 (0.908-1.378)	0.291
T stage		<0.001		
T1	Reference			
T2	0.912 (0.779-1.068)	0.254	1.037 (0.882-1.219)	0.663
T3	2.482 (2.126-2.898)	<0.001	1.495 (1.256-1.780)	<0.001
T4	3.161 (2.303-4.341)	<0.001	1.737 (1.240-2.432)	0.001
N stage		<0.001		
N0	0.364 (0.274-0.482)	<0.001	0.953 (0.695-1.306)	0.763
N1	2.748 (2.073-3.644)	<0.001	1.050 (0.766-1.439)	0.763
M stage		<0.001		
M0	0.193 (0.154-0.243)	<0.001	0.462 (0.323-0.660)	<0.001
M1	5.171 (4.116-6.497)	<0.001	2.165 (1.515-3.096)	<0.001

“-”: No data.

图1 AFP阴性HCC患者单因素和多因素Cox回归分析森林图

Fig. 1 Forest map of univariate and multivariate Cox regression analysis on AFP negative HCC patients

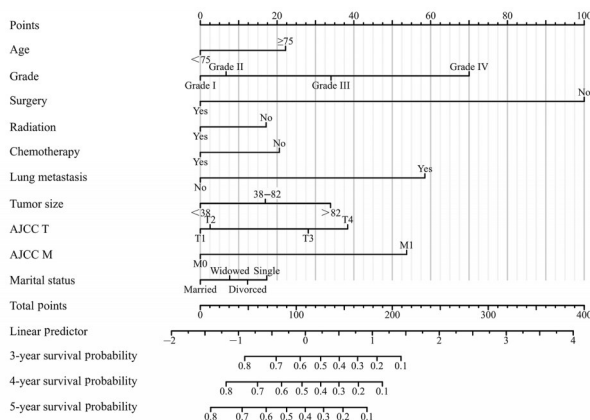


图2 AFP阴性HCC患者CSS预后列线图

Fig. 2 Nomogram of CSS prognosis of AFP negative HCC patients

和0.646 (95%CI: 0.486~0.825)。本研究结果表明: 构建的AFP阴性HCC患者CSS预后列线图预测性能明显优于AJCC分期。见图3。

2.4.3 列线图的校准曲线 采用校准图将列线图与校准图的完美曲线进行比较。灰色线表示理想的参考线, 黑色、红色和蓝色实线分别代表3、4和5年列线图预测线, 预测线越贴近灰色线拟合情况越好, 列线图预测的生存率就越准确。列线图的校准曲线结果显示: 本研究中列线图预测结果与观察到的AFP阴性HCC患者生存率之间具有较高的一致性。见图4。

2.4.4 训练集、内部验证集和外部验证集的DCA曲线 将列线图的临床净收益与基于AJCC分期的临床净收益进行比较。训练集、内部验证集和外部验证集的DCA曲线显示: 列线图在某特定概率阈

表2 训练集和外部验证集AFP阴性HCC患者基线资料
Tab. 2 Basic data of AFP negative HCC patients in training cohort and external validation cohort [n(η/%)]

Characteristic	Training (n=1 822)	External validation (n=101)	χ^2	P
Age(year)			11.05	< 0.05
<75	1 413 (77.55)	93 (92.08)		
≥75	409 (22.45)	8 (7.92)		
Grade			36.55	< 0.05
Grade I	771 (42.32)	42 (41.58)		
Grade II	865 (47.48)	31 (30.69)		
Grade III	174 (9.55)	28 (27.72)		
Grade IV	12 (0.66)	0 (0)		
Surgery			55.63	< 0.05
Yes	1 076 (59.06)	21 (20.79)		
No	746 (40.94)	80 (79.21)		
Radiotherapy			11.68	< 0.05
Yes	164 (9.00)	20 (19.80)		
No	1 658 (91.00)	81 (80.20)		
Chemotherapy			17.48	< 0.05
Yes	612 (33.59)	55 (54.46)		
No	1 210 (66.41)	46 (45.54)		
Lung metastasis			67.35	< 0.05
Yes	30 (1.65)	15 (14.85)		
No	1 792 (98.35)	86 (85.15)		
Tumor size			95.86	< 0.05
<38 mm	770 (42.26)	14 (13.86)		
38—82 mm	714 (39.19)	28 (27.72)		
>82 mm	338 (18.55)	59 (58.42)		
Marital status			61.43	< 0.05
Divorced	206 (11.31)	0 (0)		
Married	1 119 (61.42)	101 (100.00)		
Single	339 (18.61)	0 (0)		
Widowed	158 (8.67)	0 (0)		
T stage			217.05	< 0.05
T1	1 098 (60.26)	14 (13.86)		
T2	408 (22.39)	23 (22.77)		
T3	269 (14.76)	36 (35.64)		
T4	47 (2.58)	28 (27.72)		
M stage			117.57	< 0.05
M0	1 737 (95.33)	69 (68.32)		
M1	85 (4.67)	32 (31.68)		

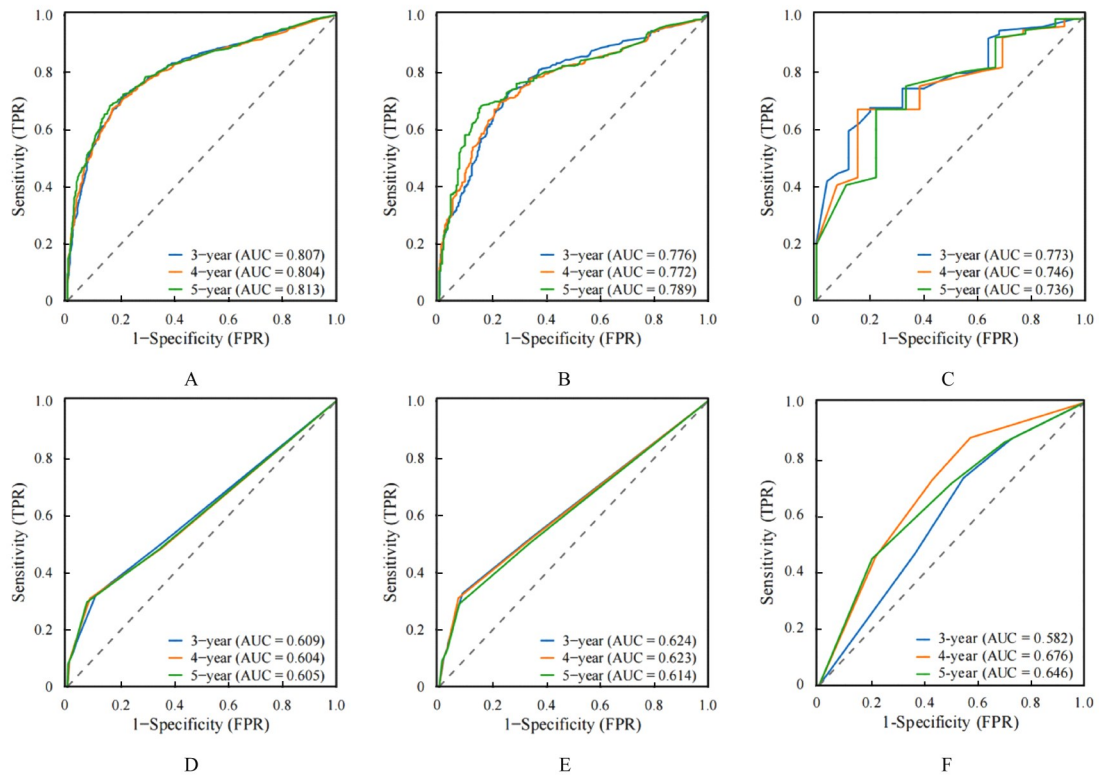
值(x轴线)的净收益(y轴线)明显高于AJCC分期在某特定概率阈值时净收益。见图5。

2.4.5 列线图和AJCC分期的风险分层 将训练集患者的生存状态、生存时间和风险评分总分整合到X-tile软件中计算列线图风险评分临界值^[14],以风险评分临界值进行列线图风险分层,分为低风险组(<130分)、中风险组(130~220分)和高风险组(>221分)。AJCC分期风险分层以SEER分期划分。以内部验证集和外部验证集对该临界值风险分层的稳定性进行验证。Kaplan-Meier生存曲线分析结果显示:在所有队列中,不同风险亚组患者生存率比较差异均有统计学差异($P<0.01$);与AJCC分期比较,列线图具有较好的识别高风险人群的能力。见图6。

3 讨论

HCC是全球发病率和死亡率较高的癌症之一,尽管治疗方法多种多样,但其复发率和死亡率仍然很高。肝癌预后评估系统如巴塞罗那分期、TNM分期和中国肝癌分期等分期系统较为完善,但在临床应用上仍有不足之处,其忽略了许多重要的危险因素,如年龄、性别和治疗经过等。在许多癌症中,列线图已被证实较传统的分期系统更为准确^[2, 4, 12, 17]。血清AFP是HCC临床筛查、诊断、复发和预后监测的主要生物标志物^[18]。但在临床上,并非所有HCC患者AFP均为阳性。研究^[10]显示:HCC患者中AFP阴性占52%(261/502),在TNM I期患者中AFP阴性患者占53.5%(51/95),在TNM IV期HCC患者中AFP阴性患者占41.5%(24/58),表明约一半的HCC患者为AFP阴性。AFP阴性HCC患者在随访中增加了检测成本,且临床价值有限^[19],因此对于该类患者应区别对待。目前,AFP阴性HCC患者复发和预后等监测主要依靠影像学方法^[20]。但是,影像学检查价格较昂贵,不适合在短时间内重复。AJCC分期系统作为HCC患者预后的评估系统并非为AFP阴性HCC患者制定,因此对HCC患者预测价值有限^[21-22]。

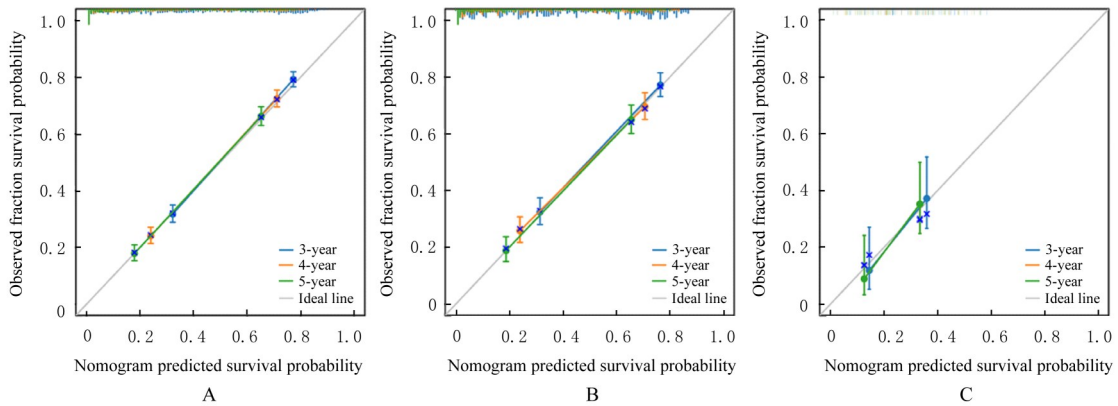
研究^[21, 23]表明:AFP阴性HCC患者的预后优于AFP阳性HCC患者,因此对AFP阴性HCC患者进行预后预测具有重要的临床意义,可对预后危险因素的早干预和早治疗提供依据。本研究中,多因素Cox回归分析确定了年龄、病理分级、手术情况、放化疗情况、肺转移情况、肿瘤大小、婚姻状况、肿瘤T分期和肿瘤M分期是影响HCC患者CSS的独立预后因素,与有关研究结果^[2, 17]具有



A-C: Nomogram; D-F: AJCC stage; A, D: Training cohort; B, E: Internal validation cohort; C, F: External validation cohort.

图3 时间依赖ROC曲线图评估CSS列线图 and AJCC分期在不同组集中的预测性能

Fig. 3 Prediction abilities of CSS nomogram and AJCC stage in different cohorts assessed by time-dependent ROC curve



A: Training cohort; B: Internal validation cohort; C: External validation cohort.

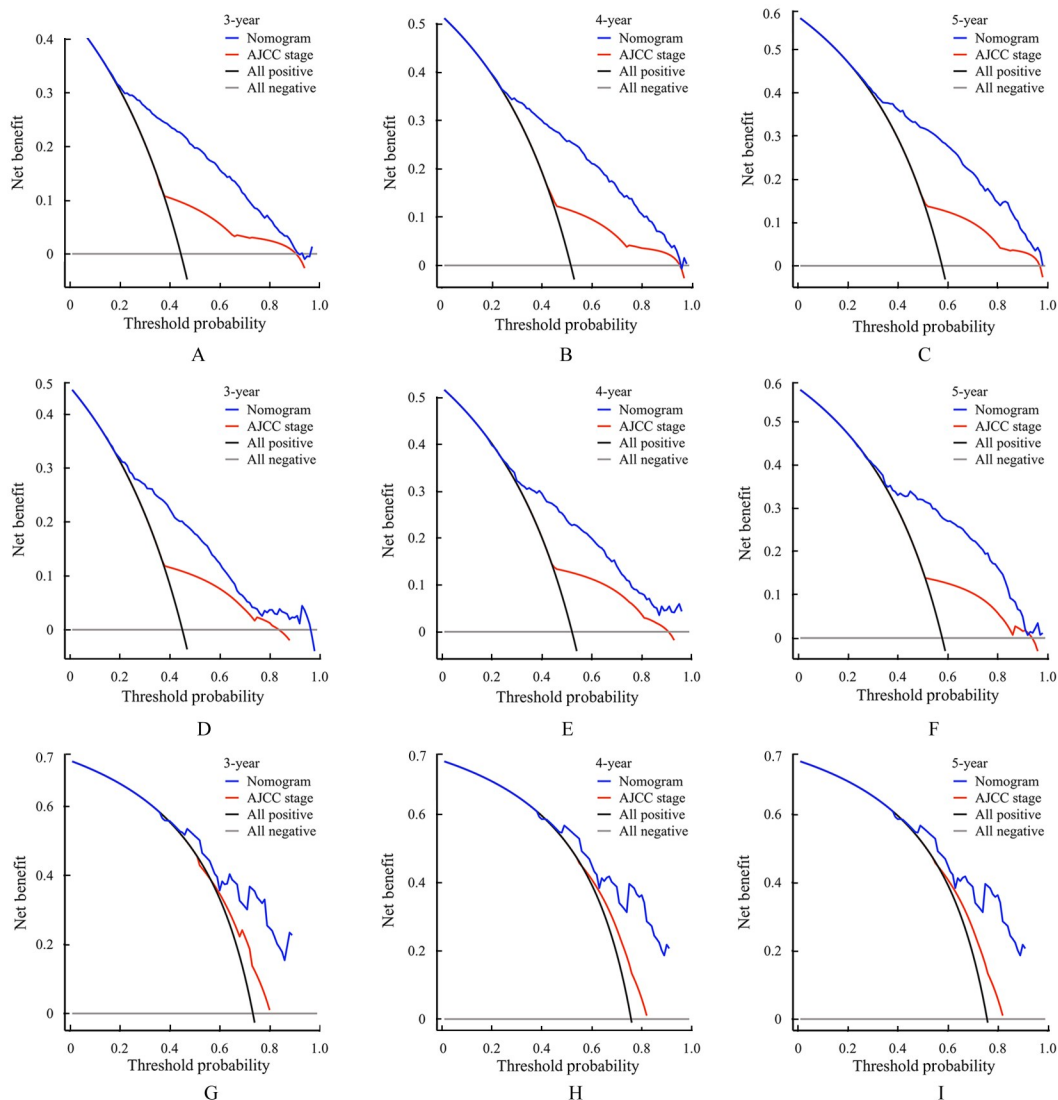
图4 AFP阴性HCC患者列线图的校准曲线图

Fig. 4 Calibration curves of nomogram of AFP negative HCC patients

一致性。骨转移是癌症患者的独立危险因素^[24-25]，但在本研究AFP阴性HCC患者的结果中并未得出该结果，可能与患者接受的治疗有关^[26]。本研究结果表明：接受手术、放疗和化疗等治疗的HCC患者预后更好，该结论可用于指导医生和患者的选择。另外，年龄也是患者的危险因素之一^[27]，在本研究中，年龄>75岁的患者生存率更低，可能

与老年患者的免疫力低下和身体退化有关^[28-30]。

最后，对列线图进行了内部数据验证和中国病例外部数据验证。本研究中，CSS预后列线图的时间依赖ROC曲线AUC值超过0.70，在验证集的结果也相同，预测AFP阴性HCC患者CSS的能力优于AJCC分期，在校准图中也证实了列线图预测的CSS概率与实际概率高度一致，与传统AJCC分期



A-C: Training cohort; D-F: Internal validation cohort; G-I: External validation cohort; A, D, G: 3 years; B, E, H: 4-year; C, F, I: 5-year.

图5 AFP阴性HCC患者DCA曲线

Fig. 5 DCA curves of AFP negative HCC patients

比较, 列线图预测 AFP 阴性 HCC 患者生存率具有更高的净收益, 且列线图中 3 个风险组患者 CSS 比较差异均有统计学意义, 与传统的 AJCC 分期比较, 列线图也有较好的识别高风险人群的能力。本研究也存在一定的局限性, 首先大样本回顾性研究基于 SEER 数据库, 该数据库可能存在一些固有的偏倚; 其次, SEER 数据库中缺少一些重要预后相关因素的数据, 如乙型肝炎病毒表面抗原和血管浸润情况及并发其他疾病等, 上述指标均为 HCC 的预后危险因素。

综上所述, 本研究结果表明: 年龄、组织学分级、手术情况、放疗情况、化疗情况、肺转移情况、肿瘤大小、肿瘤 T 分期、肿瘤 M 分期和婚姻

状况是 AFP 阴性 HCC 患者的重要预后因素, 可用于评估患者的生存期, 本研究构建的 CSS 列线图能够很好地指导临床实践。

利益冲突声明:

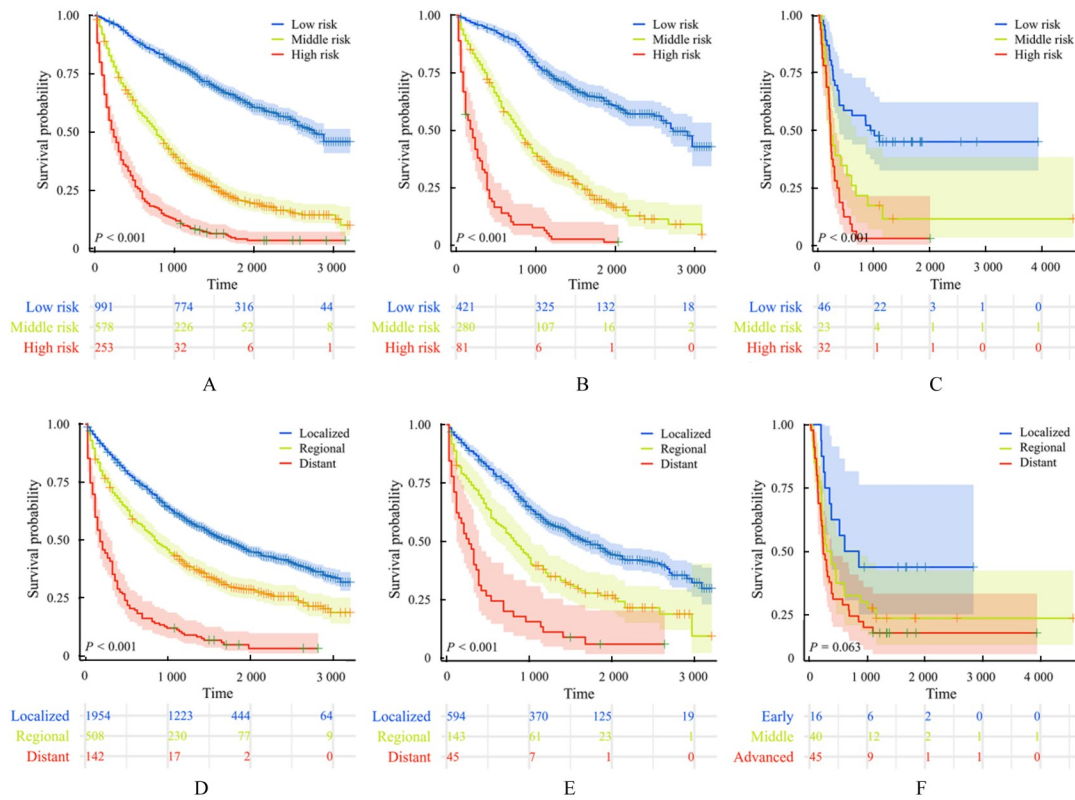
所有作者声明不存在利益冲突。

作者贡献声明:

余孝鹏参与数据整理和分析及论文撰写, 余孝鹏和杨仁义参与研究设计, 余孝鹏、杨仁义、贺佐梅和曾普华参与论文审校。

[参考文献]

- [1] BRAY F, FERLAY J, SOERJOMATARAM I, et al. Global cancer statistics 2018: GLOBOCAN estimates of



A, D: Training validation cohort; B, E: Internal validation cohort; C, F: External validation cohort.

图6 AFP阴性HCC患者列线图(A-C)和AJCC分期(D-F)风险分层

Fig. 6 Nomograms(A-C) and AJCC stage(D-F) of risk stratifications of AFP negative HCC patients

incidence and mortality worldwide for 36 cancers in 185 countries[J]. *CA Cancer J Clin*, 2018, 68(6): 394-424.

[2] YAN B, SU B B, BAI D S, et al. A practical nomogram and risk stratification system predicting the cancer-specific survival for patients with early hepatocellular carcinoma[J]. *Cancer Med*, 2021, 10(2): 496-506.

[3] GALLE P R, FOERSTER F, KUDO M, et al. Biology and significance of alpha-fetoprotein in hepatocellular carcinoma[J]. *Liver Int*, 2019, 39(12): 2214-2229.

[4] YANG D S, ZHU M Q, XIONG X Y, et al. Clinical features and prognostic factors in patients with microvascular infiltration of hepatocellular carcinoma: development and validation of a nomogram and risk stratification based on the SEER database [J]. *Front Oncol*, 2022, 12: 987603.

[5] 中华人民共和国国家卫生健康委员会.原发性肝癌诊疗指南(2022年版)[J]. *肿瘤综合治疗电子杂志*, 2022, 8(2): 16-53.

[6] CUCCHETTI A, PISCAGLIA F, GRIGIONI A D, et al. Preoperative prediction of hepatocellular carcinoma tumour grade and micro-vascular invasion by means of

artificial neural network: a pilot study [J]. *J Hepatol*, 2010, 52(6): 880-888.

[7] WANG M J, DEVARAJAN K, SINGAL A G, et al. The doylestown algorithm: a test to improve the performance of AFP in the detection of hepatocellular carcinoma[J]. *Cancer Prev Res*, 2016, 9(2): 172-179.

[8] GIANNINI E G, MARENCO S, BORGONOVO G, et al. Alpha-fetoprotein has no prognostic role in small hepatocellular carcinoma identified during surveillance in compensated cirrhosis [J]. *Hepatology*, 2012, 56(4): 1371-1379.

[9] BAI D S, ZHANG C, CHEN P, et al. The prognostic correlation of AFP level at diagnosis with pathological grade, progression, and survival of patients with hepatocellular carcinoma[J]. *Sci Rep*, 2017, 7(1): 12870.

[10] FARINATI F, MARINO D, DE GIORGIO M, et al. Diagnostic and prognostic role of alpha-fetoprotein in hepatocellular carcinoma: both or neither? [J]. *Am J Gastroenterol*, 2006, 101(3): 524-532.

[11] JIANG S J, ZHAO R J, LI Y R, et al. Prognosis and nomogram for predicting postoperative survival of duodenal adenocarcinoma: a retrospective study in China and the SEER database[J]. *Sci Rep*, 2018, 8(1): 7940.

- [12] WU J, ZHANG H B, LI L, et al. A nomogram for predicting overall survival in patients with low-grade endometrial stromal sarcoma: a population-based analysis[J]. *Cancer Commun*, 2020, 40(7): 301-312.
- [13] VICKERS A J, ELKIN E B. Decision curve analysis: a novel method for evaluating prediction models[J]. *Med Decis Making*, 2006, 26(6): 565-574.
- [14] CAMP R L, DOLLE-FILHART M, RIMM D L. X-tile: a new bio-informatics tool for biomarker assessment and outcome-based cut-point optimization[J]. *Clin Cancer Res*, 2004, 10(21): 7252-7259.
- [15] TAN X J, WANG J K, TANG J, et al. A nomogram for predicting cancer-specific survival in children with wilms tumor: a study based on SEER database and external validation in China [J]. *Front Public Health*, 2022, 10: 829840.
- [16] HUANG X, LUO Z, LIANG W, et al. Survival nomogram for young breast cancer patients based on the SEER database and an external validation cohort [J]. *Ann Surg Oncol*, 2022, 29(9): 5772-5781.
- [17] LIU K, HUANG G B, CHANG P K, et al. Construction and validation of a nomogram for predicting cancer-specific survival in hepatocellular carcinoma patients[J]. *Sci Rep*, 2020, 10(1): 21376.
- [18] TERENTIEV A A, MOLDOGAZIEVA N T. Alpha-fetoprotein: a renaissance [J]. *Tumour Biol*, 2013, 34(4): 2075-2091.
- [19] TOYODA H, KUMADA T, TADA T, et al. Clinical utility of highly sensitive *Lens culinaris* agglutinin-reactive alpha-fetoprotein in hepatocellular carcinoma patients with alpha-fetoprotein <20 ng/mL [J]. *Cancer Sci*, 2011, 102(5): 1025-1031.
- [20] XU Y Y, LU X, MAO Y L, et al. Clinical diagnosis and treatment of alpha-fetoprotein-negative small hepatic lesions [J]. *Chung Kuo Yen Cheng Yen Chiu*, 2013, 25(4): 382-388.
- [21] ZHANG X F, QI X, MENG B, et al. Prognosis evaluation in alpha-fetoprotein negative hepatocellular carcinoma after hepatectomy: comparison of five staging systems[J]. *Eur J Surg Oncol*, 2010, 36(8): 718-724.
- [22] GÓMEZ-RODRÍGUEZ R, ROMERO-GUTIÉRREZ M, ARTAZA-VARASA T, et al. The value of the Barcelona Clinic Liver Cancer and alpha-fetoprotein in the prognosis of hepatocellular carcinoma [J]. *Rev Esp Enferm Dig*, 2012, 104(6): 298-304.
- [23] GAN W, HUANG J L, ZHANG M X, et al. New nomogram predicts the recurrence of hepatocellular carcinoma in patients with negative preoperative serum AFP subjected to curative resection [J]. *J Surg Oncol*, 2018, 117(7): 1540-1547.
- [24] REN N, QIN L X, TU H, et al. The prognostic value of circulating plasma DNA level and its allelic imbalance on chromosome 8p in patients with hepatocellular carcinoma [J]. *J Cancer Res Clin Oncol*, 2006, 132(6): 399-407.
- [25] WANG X P, MAO M J, HE Z L, et al. Development and validation of a prognostic nomogram in AFP-negative hepatocellular carcinoma [J]. *Int J Biol Sci*, 2019, 15(1): 221-228.
- [26] ZHAN H, ZHAO X, LU Z X, et al. Correlation and survival analysis of distant metastasis site and prognosis in patients with hepatocellular carcinoma [J]. *Front Oncol*, 2021, 11: 652768.
- [27] ZHANG W, JI L C, WANG X J, et al. Nomogram predicts risk and prognostic factors for bone metastasis of pancreatic cancer: a population-based analysis [J]. *Front Endocrinol*, 2021, 12: 752176.
- [28] WANG H M, SHAN X F, ZHANG M, et al. Homogeneous and heterogeneous risk and prognostic factors for lung metastasis in colorectal cancer patients [J]. *BMC Gastroenterol*, 2022, 22(1): 193.
- [29] TONG Y X, HUANG Z H, HU C, et al. Independent risk factors evaluation for overall survival and cancer-specific survival in thyroid cancer patients with bone metastasis: a study for construction and validation of the predictive nomogram [J]. *Medicine*, 2020, 99(36): e21802.
- [30] HUANG Z H, HU C, LIU K W, et al. Risk factors, prognostic factors, and nomograms for bone metastasis in patients with newly diagnosed infiltrating duct carcinoma of the breast: a population-based study [J]. *BMC Cancer*, 2020, 20(1): 1145.