Research Square

Preprints are preliminary reports that have not undergone peer review.
They should not be considered conclusive, used to inform clinical practice,
or referenced by the media as validated information.

Development and validation of a prediction model for multidrug-resistant infection in patients with HBV-related acute-on-chronic liver failure complicated by bacterial infection: a dual-center retrospective cohort study

Zhanhu Bi
Air Force Medical University
Haifeng Hu
Air Force Medical University
Hong Du
Air Force Medical University
Linxu Wang
Air Force Medical University
Jiayi Zhan
Air Force Medical University
Fei Hu
985th Hospital of Chinese People's Liberation Army
Denghui Yu
PLA General Hospital of Southern Theater Command
Hongkai Xu
PLA 960th Hospital
Xiaofei Yang
Air Force Medical University
Yidi Ding
Air Force Medical University
Chunlin Wang
The 901st Hospital of the Joint Logistics Support Force of the Chinese People's Liberation Army
Jianqi Lian
lianjq@fmmu.edu.cn

Air Force Medical University

Research Article

Keywords: Acute-on-chronic liver failure, Bacterial infection, Multidrug-resistant, Prediction model, Risk factors

Posted Date: November 12th, 2026

DOI: https://doi.org/10.21203/rs.3.rs-11418679/v1

License: This work is licensed under a Creative Commons Attribution 4.0 International License. Read Full License

Additional Declarations: No competing interests reported.

Abstract

Background

Multidrug-resistant (MDR) bacterial infections significantly increase mortality in patients with hepatitis B virus-related acute-on-chronic liver failure (HBV-ACLF). This study aimed to develop and validate a prediction model based on clinical indicators at the time of infection onset to early identify high-risk patients for MDR infection and guide empirical antimicrobial therapy.

Methods

This retrospective study enrolled 191 patients with HBV-ACLF and bacterial infection, divided into MDR (*n* = 125) and non-MDR (*n* = 66) groups. Clinical and etiological data were collected at the time of infection onset. Candidate variables were initially screened by univariate logistic regression and clinical relevance, followed by Spearman correlation analysis and least absolute shrinkage and selection operator (LASSO) regression for feature selection. A multivariable logistic regression model was constructed, and a nomogram was developed. Internal validation was performed using the bootstrap method (1000 resamples) and 10-fold cross-validation. Model performance was evaluated using receiver operating characteristic (ROC) curves, calibration curves, and decision curve analysis (DCA).

Results

A total of 191 pathogenic strains were isolated, including 111 (58.1%) Gram-negative and 80 (41.9%) Gram-positive bacteria. Multivariable logistic regression identified age (OR = 1.060, 95% CI 1.026–1.101), length of hospital stay (OR = 1.114, 95% CI 1.065–1.173), lactate (Lac, OR = 1.494, 95% CI 1.249–1.864), and procalcitonin (PCT, OR = 1.606, 95% CI 1.307–2.174) as independent risk factors for MDR infection (all $P < 0.001$). The apparent area under the curve (AUC) was 0.902 (95% CI 0.859–0.945), and the bootstrap-corrected AUC was 0.893. The Hosmer-Lemeshow test yielded a P value of 0.395. DCA demonstrated significant net benefit across a wide range of threshold probabilities.

Conclusions

The 4-variable prediction model based on age, length of hospital stay, Lac, and PCT demonstrated good discrimination, calibration, and clinical utility for assessing MDR risk in HBV-ACLF patients with bacterial infection, providing a quantitative basis for early empirical antimicrobial therapy.

Background

Acute-on-chronic liver failure (ACLF) is an acute deterioration of liver function in patients with chronic liver disease, characterized by organ failure and high short-term mortality [1]. Although the etiology

varies geographically, hepatitis B virus (HBV) infection remains the leading cause of ACLF in the Asia-Pacific region, including China [1]. Bacterial infection is one of the most common complications in patients with ACLF and is closely associated with progression of organ failure and significantly increased mortality [2, 3].

In recent years, with the widespread use of broad-spectrum antimicrobials, the proportion of multidrug-resistant (MDR) bacterial infections among patients with ACLF has been increasing [2, 4]. MDR infections not only substantially increase the rate of antimicrobial treatment failure and mortality but also significantly exacerbate the economic burden on the healthcare system [4, 5]. However, the early identification of MDR infections remains a formidable challenge. Conventional microbiological culture and antimicrobial susceptibility testing are time-consuming, often placing clinicians in a dilemma when prescribing initial empirical antimicrobial therapy: overuse of broad-spectrum antimicrobials increases selective pressure for resistance, whereas under-treatment may delay effective therapy and lead to disease deterioration [6]. Therefore, developing an MDR risk prediction model based on routine clinical indicators at the time of infection onset holds significant translational value for guiding precise empirical antimicrobial therapy and improving patient prognosis [7, 8].

Currently, although an increasing number of MDR infection prediction models have been reported [8], several limitations remain. On the one hand, the vast majority of existing models focus on Intensive Care Unit (ICU) or general hospitalized populations, while studies specifically targeting the high-risk HBV-ACLF population remain limited [7]. On the other hand, despite the considerable number of models, the robustness and generalizability of most models remain questionable [8]. Furthermore, existing models often incorporate complex variables that are not readily available, making them difficult to implement rapidly in real-world clinical decision-making scenarios [9].

Given these gaps, this study aimed to develop and validate a concise and practical MDR infection prediction model using routine clinical indicators at the time of infection onset, specifically during the critical window between the establishment of bacterial infection and the availability of pathogen results. We anticipate that this model will provide clinicians with an intuitive quantitative tool for risk stratification, aiding in the selection of initial empirical antimicrobial therapy intensity, thereby striking an optimal balance between avoiding over-treatment and preventing under-treatment.

Methods

Study design and participants

This retrospective cohort study was conducted at two centers: Tangdu Hospital of the Fourth Military Medical University and the 901st Hospital of the Joint Logistics Support Force of PLA. A total of 571 patients hospitalized for liver failure or severe hepatitis between January 1, 2010, and December 31, 2021, were initially screened. The detailed process of patient screening and enrollment is summarized in Fig. 1.

The inclusion criteria were as follows: (1) ACLF diagnosed according to the consensus criteria of the Asian Pacific Association for the Study of the Liver (APASL)—acute hepatic insult manifesting as jaundice (serum total bilirubin ≥ 5 mg/dL) and coagulopathy (INR ≥ 1.5 or prothrombin activity < 40%), complicated within 4 weeks by clinical ascites and/or encephalopathy in patients with chronic liver disease (non-cirrhotic or compensated cirrhosis) [10]; (2) Pathogens isolated from sputum, blood, urine, ascites, bile, bone marrow, pleural fluid, or skin/soft tissue cultures at the time of infection onset; (3) Availability of complete data for all pre-specified candidate variables at the time of infection onset.

The exclusion criteria were as follows: (1) Non-HBV etiologies (e.g., co-infection with other hepatotropic viruses, alcoholic, autoimmune, or cholestatic liver disease; *n* = 247); (2) Lacked documented microbiological evidence (culture-negative or not performed) to define MDR status (*n* = 133); (3) Concurrent active malignancy, including hepatocellular carcinoma or malignancies of other organs; (4) Severe underlying comorbidities that were expected to limit survival to less than the duration of the current infection episode, or that the investigators judged could significantly confound the assessment of infection-related clinical parameters (e.g., chronic kidney disease stage 4–5, New York Heart Association class III–IV heart failure, or active autoimmune disease); (5) Prior or current liver transplantation; (6) Missing data for any pre-specified candidate variable at infection onset.

After rigorous screening, a total of 191 patients with HBV-ACLF and bacterial infection were finally included and categorized into the MDR group (*n* = 125) and the non-MDR group (*n* = 66) based on antimicrobial susceptibility results at the time of infection.

Data collection and definitions

Clinical data at the time of infection onset were collected, including: (1) demographic characteristics: age, sex, and body mass index (BMI); (2) clinical features: length of hospital stay (time from admission to infection onset), hepatic encephalopathy (HE) grade, ascites, and upper gastrointestinal bleeding; (3) laboratory parameters: routine blood tests, liver and renal function, coagulation profile, inflammatory markers (CRP, PCT, and IL-6), ammonia, lactate (Lac), electrolytes, lipid profiles, and other available laboratory variables; (4) liver disease severity scores: Child-Pugh score, MELD, MELD-Na, and i-MELD. Data on pathogen distribution and antimicrobial susceptibility were also collected.

Microbiological methods

Specimens from the infection site (sputum, blood, urine, ascites, bile, etc.) were collected. Species identification was performed using an automated system, and antimicrobial susceptibility testing was conducted using the disk diffusion or broth microdilution method, with results interpreted according to the Clinical and Laboratory Standards Institute (CLSI) standards [11]. MDR was defined as acquired non-susceptibility to at least one agent in three or more antimicrobial categories [12].

Statistical analysis

All analyses were performed using R software (version 4.2.2). All candidate variables were first evaluated by univariate logistic regression (Additional file 1: Table S3), and those with $P < 0.10$ or clinical relevance were selected as candidates for the subsequent two-stage strategy. A two-stage strategy was then applied for variable selection. First, Spearman correlation coefficients were calculated among candidate variables, and for pairs with $|r| \geq 0.9$, only one variable was retained. Second, the remaining variables were subjected to least absolute shrinkage and selection operator (LASSO) regression, with the optimal penalty parameter λ selected via 10-fold cross-validation. Variables retained by LASSO were entered into multivariable logistic regression to calculate odds ratios (ORs) and 95% confidence intervals (CIs), and a nomogram was constructed based on the regression coefficients. Model performance was evaluated as follows: discrimination by receiver operating characteristic (ROC) curves, calibration by the Hosmer-Lemeshow test and calibration curves, overall prediction error by the Brier score, and clinical utility by decision curve analysis (DCA). Internal validation was performed using the bootstrap method (1000 resamples) to calculate the corrected AUC, optimism, and calibration slope, with 10-fold cross-validation used to assess model robustness. A two-sided P value < 0.05 was considered statistically significant. Details of the univariate logistic regression and the complete pathogen distribution and antimicrobial susceptibility results are provided in Additional file 1: Tables S3 and S4.

Results

Baseline characteristics

A total of 191 patients with HBV-ACLF and bacterial infection were enrolled, including 125 (65.4%) in the MDR group and 66 (34.6%) in the non-MDR group. Statistically significant differences were observed between the two groups in terms of age, length of hospital stay, HE grade, ammonia, Lac, monocyte count, triglycerides, several coagulation parameters (PT, TT, PTA, and INR), inflammatory markers (IL-6, CRP, and PCT), and liver disease severity scores ($P < 0.05$) (Table 1, placed at the end of the document).

Table 1
Patient characteristics at enrolment

Variable	Overall (*n* = 191)	MDR (*n* = 125)	Non-MDR (*n* = 66)	*t*/*Z*/χ^2	*P* value
Prognosis (28-day mortality)	86 (45.0%)	66 (52.8%)	20 (30.3%)	7.946	0.005
Length of hospital stay (days)	21.00 (14.00, 28.00)	24.00 (16.00, 31.00)	17.00 (9.25, 23.00)	4.855	< 0.001
Demographics					
Age (years)	50.90 ± 13.62	53.32 ± 12.64	46.32 ± 14.31	3.477	< 0.001
Male sex	116 (60.7%)	74 (59.2%)	42 (63.6%)	0.195	0.659
BMI (kg/m²)	23.18 (21.22, 25.70)	22.84 (20.70, 25.78)	23.42 (21.32, 25.62)	-1.207	0.228
Medical history					
Family history of HBV	33 (17.3%)	21 (16.8%)	12 (18.2%)	0.002	0.969
Smoking	46 (24.1%)	31 (24.8%)	15 (22.7%)	0.020	0.888
Alcohol consumption	35 (18.3%)	26 (20.8%)	9 (13.6%)	1.041	0.308
Hypertension	15 (7.9%)	11 (8.8%)	4 (6.1%)	0.149	0.699
Diabetes	18 (9.4%)	10 (8.0%)	8 (12.1%)	0.444	0.505
Heart disease	16 (8.4%)	12 (9.6%)	4 (6.1%)	0.319	0.572
Fatty liver	29 (15.2%)	18 (14.4%)	11 (16.7%)	0.041	0.839
Cirrhosis	118 (61.8%)	77 (61.6%)	41 (62.1%)	0.000	1.000
Complications					
Upper gastrointestinal bleeding	14 (7.3%)	9 (7.2%)	5 (7.6%)	-	1.000
Ascites	152 (79.6%)	99 (79.2%)	53 (80.3%)	0.000	1.000
HE grade				-2.500	0.012
None	88 (46.1%)	48 (38.4%)	40 (60.6%)		
Grade 1	18 (9.4%)	14 (11.2%)	4 (6.1%)		

Data are presented as mean ± standard deviation, median (*P*25, *P*75), or n (%). Continuous variables were compared using Student's t-test (for normally distributed data with equal variance) or the Mann-Whitney U test (for non-normally distributed data); categorical variables were compared using the chi-square test (with Yates' continuity correction for 2×2 tables) or Fisher's exact test. Ordinal categorical variables (HE grade, Child-Pugh class) were compared using the Mann-Whitney U test.

Variable	Overall (*n* = 191)	MDR (*n* = 125)	Non-MDR (*n* = 66)	*t*/*Z*/χ^2	*P* value
Grade 2	14 (7.3%)	11 (8.8%)	3 (4.5%)		
Grade 3	9 (4.7%)	7 (5.6%)	2 (3.0%)		
Grade 4	62 (32.5%)	45 (36.0%)	17 (25.8%)		
Organ failure					
Renal failure	42 (22.0%)	26 (20.8%)	16 (24.2%)	0.131	0.717
Respiratory failure	35 (18.3%)	26 (20.8%)	9 (13.6%)	1.041	0.308
Circulatory failure	23 (12.0%)	15 (12.0%)	8 (12.1%)	0.000	1.000
Laboratory parameters					
WBC (×10⁹/L)	7.08 (4.95, 10.46)	7.43 (5.55, 10.82)	6.39 (4.59, 10.19)	1.793	0.073
Lymphocyte count (×10⁹/L)	0.90 (0.59, 1.27)	0.96 (0.60, 1.29)	0.88 (0.57, 1.26)	0.322	0.748
Monocyte count (×10⁹/L)	0.63 (0.34, 0.92)	0.70 (0.40, 1.01)	0.53 (0.32, 0.77)	2.651	0.008
Neutrophil count (×10⁹/L)	5.29 (3.38, 8.59)	5.84 (3.78, 8.71)	4.53 (3.16, 7.52)	1.919	0.055
HGB (g/L)	119.77 ± 26.58	122.34 ± 27.03	114.91 ± 25.19	1.850	0.066
PLT (×10⁹/L)	94.00 (56.50, 138.00)	92.00 (56.00, 133.00)	104.50 (61.00, 140.00)	-0.714	0.477
HCT (%)	34.73 ± 7.42	35.45 ± 7.57	33.36 ± 6.98	1.861	0.064
ALB (g/L)	30.50 (27.50, 33.60)	30.50 (27.60, 33.30)	30.45 (27.20, 34.40)	-0.481	0.632
ALT (U/L)	246.00 (72.00, 750.00)	297.00 (82.00, 883.00)	205.00 (58.00, 537.75)	1.843	0.066
AST (U/L)	228.00 (97.50, 673.50)	255.00 (106.00, 765.00)	167.00 (89.50, 405.00)	1.889	0.059
TB (μmol/L)	317.80 (214.00, 416.75)	304.30 (202.70, 422.42)	341.75 (230.48, 402.38)	-0.622	0.535

Data are presented as mean ± standard deviation, median (*P*25, *P*75), or n (%). Continuous variables were compared using Student's t-test (for normally distributed data with equal variance) or the Mann-Whitney U test (for non-normally distributed data); categorical variables were compared using the chi-square test (with Yates' continuity correction for 2×2 tables) or Fisher's exact test. Ordinal categorical variables (HE grade, Child-Pugh class) were compared using the Mann-Whitney U test.

Variable	Overall (*n* = 191)	MDR (*n* = 125)	Non-MDR (*n* = 66)	*t*/*Z*/χ^2	*P* value
Ammonia (μmol/L)	72.10 (42.25, 99.30)	83.80 (44.20, 109.10)	54.70 (41.70, 84.18)	2.724	0.006
GGT (U/L)	83.00 (46.50, 136.50)	84.00 (50.00, 132.00)	73.50 (45.25, 165.50)	-0.129	0.898
ALP (U/L)	134.00 (102.50, 187.50)	133.00 (101.00, 193.00)	136.00 (103.00, 184.00)	-0.081	0.936
ChE (U/L)	3056.00 (2234.00, 4223.00)	3130.00 (2312.00, 4183.00)	2768.00 (2018.50, 4194.00)	1.101	0.272
Lac (mmol/L)	3.59 (2.44, 5.09)	3.90 (2.94, 5.88)	2.67 (1.85, 3.69)	4.852	< 0.001
Cr (μmol/L)	62.80 (53.05, 81.40)	64.60 (53.70, 84.90)	60.50 (51.25, 74.97)	1.185	0.237
BUN (mmol/L)	5.00 (3.40, 7.65)	5.00 (3.32, 9.00)	4.95 (3.52, 6.38)	0.331	0.742
CP (mg/L)	285.50 (209.60, 356.55)	287.00 (207.70, 362.60)	281.40 (213.70, 349.77)	-0.223	0.825
Na^+ (mmol/L)	137.20 (132.30, 139.75)	137.10 (132.40, 139.80)	137.50 (131.60, 139.60)	-0.320	0.751
K^+ (mmol/L)	4.09 (3.57, 4.66)	4.17 (3.58, 4.89)	3.98 (3.54, 4.56)	1.241	0.215
PT (s)	23.60 (18.35, 29.65)	24.90 (19.70, 32.30)	21.15 (17.10, 27.78)	2.926	0.003
APTT (s)	53.50 (42.20, 63.90)	54.70 (43.50, 65.80)	50.70 (38.75, 59.28)	1.860	0.063
TT (s)	22.70 (20.70, 25.75)	23.20 (21.10, 26.30)	22.25 (20.35, 23.98)	2.530	0.012
Fib (g/L)	1.18 (0.91, 1.66)	1.11 (0.84, 1.59)	1.25 (1.00, 1.83)	-1.862	0.063
PTA (%)	32.00 (21.90, 43.85)	30.30 (21.00, 39.60)	37.30 (27.25, 49.00)	-2.858	0.004

Data are presented as mean ± standard deviation, median (*P*25, *P*75), or n (%). Continuous variables were compared using Student's t-test (for normally distributed data with equal variance) or the Mann-Whitney U test (for non-normally distributed data); categorical variables were compared using the chi-square test (with Yates' continuity correction for 2×2 tables) or Fisher's exact test. Ordinal categorical variables (HE grade, Child-Pugh class) were compared using the Mann-Whitney U test.

Variable	Overall (*n* = 191)	MDR (*n* = 125)	Non-MDR (*n* = 66)	*t*/*Z*/χ^2	*P* value
INR	2.08 (1.63, 2.69)	2.16 (1.73, 2.97)	1.85 (1.48, 2.49)	2.919	0.004
FDP (μg/mL)	5.30 (3.14, 11.05)	5.76 (3.30, 11.20)	5.00 (2.96, 10.88)	0.586	0.561
D-dimer (μg/mL)	1.97 (0.69, 5.25)	2.17 (0.73, 5.19)	1.74 (0.65, 5.25)	0.255	0.801
TC (mmol/L)	2.16 (1.56, 3.05)	2.09 (1.45, 3.01)	2.41 (1.69, 3.14)	-1.660	0.097
TG (mmol/L)	1.07 (0.61, 1.66)	0.98 (0.54, 1.62)	1.19 (0.72, 1.73)	-2.128	0.034
IL-6 (pg/mL)	37.16 (11.54, 77.82)	48.09 (15.59, 106.80)	17.24 (7.74, 46.64)	4.420	< 0.001
CRP (mg/L)	25.50 (13.29, 52.73)	33.60 (19.91, 67.79)	11.11 (5.71, 29.39)	6.192	< 0.001
PCT (ng/mL)	1.09 (0.36, 5.92)	2.06 (0.67, 10.29)	0.47 (0.27, 0.98)	6.175	< 0.001
AFP (ng/mL)	21.39 (3.79, 144.50)	25.80 (5.41, 144.00)	12.16 (3.08, 145.95)	1.184	0.237
Scoring systems					
Child-Pugh class				1.160	0.246
Class A	8 (4.2%)	3 (2.4%)	5 (7.6%)		
Class B	25 (13.1%)	17 (13.6%)	8 (12.1%)		
Class C	158 (82.7%)	105 (84.0%)	53 (80.3%)		
Child-Pugh score	12.00 (10.00, 13.00)	12.00 (11.00, 14.00)	11.00 (10.00, 12.75)	2.893	0.017
MELD	26.99 ± 6.15	27.72 ± 6.29	25.60 ± 5.66	2.288	0.023
MELD-Na	28.00 ± 5.93	28.70 ± 5.96	26.68 ± 5.70	2.259	0.025
i-MELD	45.52 (40.70, 51.14)	46.69 (42.44, 51.79)	44.10 (37.79, 49.94)	2.336	0.020

Data are presented as mean ± standard deviation, median (*P*25, *P*75), or n (%). Continuous variables were compared using Student's t-test (for normally distributed data with equal variance) or the Mann-Whitney U test (for non-normally distributed data); categorical variables were compared using the chi-square test (with Yates' continuity correction for 2×2 tables) or Fisher's exact test. Ordinal categorical variables (HE grade, Child-Pugh class) were compared using the Mann-Whitney U test.

Abbreviations: MDR, multidrug-resistant; BMI, body mass index; HBV, hepatitis B virus; HE, hepatic encephalopathy; WBC, white blood cell; HGB, hemoglobin; PLT, platelet; HCT, hematocrit; ALB, albumin;

ALT, alanine aminotransferase; AST, aspartate aminotransferase; TB, total bilirubin; GGT, gamma-glutamyl transferase; ALP, alkaline phosphatase; ChE, cholinesterase; Lac, lactate; Cr, creatinine; BUN, blood urea nitrogen; CP, ceruloplasmin; PT, prothrombin time; APTT, activated partial thromboplastin time; TT, thrombin time; Fib, fibrinogen; PTA, prothrombin activity; INR, international normalized ratio; FDP, fibrinogen degradation products; TC, total cholesterol; TG, triglycerides; IL-6, interleukin-6; CRP, C-reactive protein; PCT, procalcitonin; AFP, alpha-fetoprotein; MELD, model for end-stage liver disease; MELD-Na, MELD combined with serum sodium; i-MELD, integrated MELD.

Specifically, patients in the MDR group were older (53.32 ± 12.64 vs. 46.32 ± 14.31 years, $P < 0.001$), had a longer length of hospital stay [24.00 (16.00, 31.00) vs. 17.00 (9.25, 23.00) days, $P < 0.001$], and a higher 28-day mortality rate (52.8% vs. 30.3%, $P = 0.005$) compared to the non-MDR group. Among the key predictors included in the final model, both Lac [3.90 (2.94, 5.88) vs. 2.67 (1.85, 3.69) mmol/L] and PCT [2.06 (0.67, 10.29) vs. 0.47 (0.27, 0.98) ng/mL] were higher in the MDR group (both $P < 0.001$). Other laboratory and scoring parameters are detailed in Table 1.

No statistically significant differences were found between the two groups regarding sex, BMI, family history of HBV, comorbidities (e.g., hypertension, diabetes), or complications such as upper gastrointestinal bleeding, ascites, and renal failure ($P > 0.05$).

A total of 191 pathogenic strains were isolated, including 111 (58.1%) Gram-negative and 80 (41.9%) Gram-positive bacteria. The predominant Gram-negative isolates were *Escherichia coli* (39 strains, 20.4%), *Klebsiella pneumoniae* (31 strains, 16.2%), and *Acinetobacter baumannii* (10 strains, 5.2%). Among Gram-positive bacteria, *Enterococcus faecium* (32 strains, 16.8%), *Staphylococcus hominis* (7 strains, 3.7%), and *Staphylococcus aureus* (6 strains, 3.1%) were the most common (Fig. 2).

The most common specimen sources were sputum (54 strains, 28.3%), blood (41 strains, 21.5%), urine (38 strains, 19.9%), and ascites (31 strains, 16.2%). Specifically, *E. coli* was predominantly isolated from blood and ascites (12 strains each), *K. pneumoniae* from sputum (16 strains), and *E. faecium* from urine (22 strains) (Fig. 2). Detailed distribution data for all 38 pathogens are provided in Additional file 1: Table S1 and Figure S1.

Pathogen distribution between groups

Differences in pathogen distribution were observed between the MDR and non-MDR groups. The detection rates of *Enterococcus faecium* (22.4% vs 6.1%, $P = 0.004$) and *Escherichia coli* (26.4% vs 9.1%, $P = 0.005$) were higher in the MDR group. Conversely, *Stenotrophomonas maltophilia* was more frequently detected in the non-MDR group (0.0% vs 12.1%, $P < 0.001$). No statistically significant differences were observed for other major pathogens, including *Staphylococcus hominis*, *Staphylococcus aureus*, *Staphylococcus haemolyticus*, MRSA, *Klebsiella pneumoniae*, *Acinetobacter baumannii*, and *Haemophilus influenzae* ($P > 0.05$) (Fig. 3).

Discussion

Pathogen distribution and antimicrobial strategy

Among the pathogens isolated in this study, *Escherichia coli* and *Klebsiella pneumoniae* were the predominant Gram-negative bacteria, while *Enterococcus faecium* was the predominant Gram-positive bacterium, which is generally consistent with the pathogen spectrum previously reported for ACLF complicated by infection [13]. However, this study further revealed that the detection rates of *E. faecium* and *E. coli* were significantly higher in the MDR group than in the non-MDR group, suggesting that infections with these two pathogens are highly associated with MDR risk. Antimicrobial susceptibility testing in our cohort revealed that the MDR group exhibited extremely low susceptibility rates to clinically common fluoroquinolones (e.g., levofloxacin, ciprofloxacin) and third-generation cephalosporins (e.g., cefepime). This finding is consistent with European multicenter data demonstrating that MDR infections in cirrhosis and ACLF are associated with the failure of empirical strategies based on third-generation cephalosporins or quinolones [14]. This phenomenon highlights a critical challenge in current clinical practice: during the decision-making window when infection is established but susceptibility results are not yet available, conventional empirical regimens relying on fluoroquinolones or third-generation cephalosporins carry a substantially high risk of inadequate coverage for high-risk MDR patients [14].

For Gram-negative infections, carbapenems (e.g., meropenem) remain the preferred first-line treatment for invasive extended-spectrum β-lactamase (ESBL)-producing Enterobacteriaceae infections outside the urinary tract [15]. However, considering the specific pathophysiological status of patients with cirrhosis and liver failure, indiscriminate empirical use of carbapenems would significantly exacerbate selective pressure for resistance [6]. Our findings suggest that for high-risk MDR patients (i.e., those presenting with advanced age, prolonged hospital stay, elevated Lac or PCT at infection onset, and a high predicted risk on the nomogram), empirical therapy based on carbapenems is reasonable and necessary; whereas for low-risk MDR patients, carbapenem-sparing strategies should be guided by the suspected pathogen and infection site (e.g., trimethoprim/sulfamethoxazole or fluoroquinolones for susceptible urinary isolates), and third-generation cephalosporins should not be routinely used when ESBL-producing organisms are suspected [6, 15]. Furthermore, although novel β-lactamase inhibitor combinations (e.g., ceftazidime-avibactam) exhibit favorable activity against ESBL-producing Enterobacteriaceae, current guidelines recommend reserving them preferentially for carbapenem-resistant organisms [15].

Regarding Gram-positive bacteria, *E. faecium* was predominant in the MDR group [2, 13]. In our cohort, in vitro susceptibility testing indicated high sensitivity rates to both linezolid (91.4%) and vancomycin (96.6%) in the MDR group. However, safety concerns must be highly vigilant in clinical application [16]. Patients with liver failure have severely impaired liver function; long-term use of linezolid may lead to thrombocytopenia and anemia [17], while vancomycin is prone to drug accumulation and nephrotoxicity in patients with cirrhosis [18]. Therefore, for high-risk MDR patients with suspected Enterococcus infections, the selection and combination of these agents should be cautious, guided by liver function status and therapeutic drug monitoring [17, 18].

In summary, current empirical antimicrobial selection often faces the dilemma between "treatment failure due to inadequate coverage" and "exacerbated resistance due to overuse of broad-spectrum antimicrobials." The 4-variable model constructed in this study precisely bridges this decision-making gap. Prior to the availability of microbiological results, clinicians can use the nomogram to rapidly stratify MDR risk: for high-risk patients, a broad-spectrum strategy based on carbapenems (combined with glycopeptides or linezolid if necessary) should be initiated early to avoid treatment delay; whereas for low-risk patients, narrow-spectrum antimicrobials can be safely prescribed to minimize unnecessary exposure to broad-spectrum agents. Furthermore, once susceptibility results become available within 24–72 hours, de-escalation therapy should be promptly implemented for high-risk patients initially on broad-spectrum therapy if MDR infection is excluded. Through this "early precise stratification and dynamic de-escalation" strategy, an optimal balance is ultimately achieved between improving patient prognosis and curbing the selective pressure of antimicrobial resistance.

Biological rationale of independent predictors

Among the independent risk factors identified in this study, procalcitonin (PCT) demonstrated the strongest predictive efficacy (OR = 1.606). PCT is a specific biomarker for bacterial infection; its elevated levels not only indicate severe bacterial infection but, in the context of MDR infections, may also reflect the greater virulence and higher bacterial burden of MDR pathogens [19]. A prospective cohort study confirmed that patients with MDR bacteremia had significantly higher PCT levels than those with non-MDR bacteremia (median 9.6 vs. 1.5 ng/mL), and the predictive performance of PCT for MDR bacteremia (AUC = 0.71) was significantly superior to that of the traditional inflammatory marker CRP (AUC = 0.47) [19]. This aligns well with our findings, further supporting the core value of PCT in the early identification of MDR infections.

Lactate (Lac) is also an important independent risk factor in this model. As a well-recognized biomarker of illness severity and tissue hypoperfusion, elevated Lac levels strongly indicate severe infection [20]. In clinical practice, severe infection status (often accompanied by high Lac) prompts physicians to initiate broad-spectrum antimicrobials earlier and more aggressively [21], and early empirical use of broad-spectrum antibiotics has been recognized as a major driver in promoting antimicrobial resistance [22]. Therefore, there is a reasonable pathophysiological and clinical association between the infection severity reflected by Lac and the risk of MDR infection [20, 23]. Our findings indicate that in the specific population of HBV-ACLF, the combination of Lac and PCT can more precisely delineate the high-risk profile of MDR infection.

Advanced age may contribute to the increased risk of MDR infection through immunosenescence, the accumulation of comorbidities, and more frequent healthcare exposure and prior antimicrobial use [24]. While age was not consistently identified as an independent predictor in all cirrhosis cohorts [7], older age has been repeatedly linked to an increased risk of MDR infection in hospitalized patients with liver disease. In our study, advanced age was confirmed as an independent risk factor for MDR infection in patients with HBV-ACLF.

Furthermore, the length of hospital stay directly reflects cumulative exposure to the hospital environment. Prolonged hospitalization increases opportunities for contact with medical devices, cross-transmission, and exposure to broad-spectrum antimicrobials, thereby elevating the risk of colonization and infection with MDR strains. Consistent with our findings, hospitalization-related factors, such as nosocomial infection and recent hospitalization, have been explicitly identified as independent risk factors for MDRO infections in patients with decompensated cirrhosis [4, 14].

Clinical utility, comparison with previous models, and limitations

The model incorporates only four variables: age, length of hospital stay, Lac, and PCT, all of which are routinely available clinical parameters at the time of infection onset. Requiring no additional costs or complex equipment, the model is highly feasible in resource-limited clinical settings. Through the nomogram, clinicians can rapidly perform risk stratification during the window period before microbiological results are available, providing a quantitative basis for empirical antimicrobial decision-making. The net benefit demonstrated by DCA further supports the potential value of this model in real-world decision-making scenarios.

Compared with previous MDR prediction models, this study offers targeted improvements in the following aspects. Regarding the target population, the vast majority of existing models focus on ICU or general hospitalized patients [8, 9], while studies specifically addressing the HBV-ACLF population—which carries both severe liver dysfunction and high infection risk—remain limited [25]. Regarding clinical operability, whereas other models incorporate variables that are not immediately available or rely on complex algorithms [9], this study uses only four routinely available clinical indicators at the time of infection, maximizing discrimination while minimizing implementation thresholds, thereby facilitating rapid application in bedside decision-making.

This study has several limitations. First, although it includes data from two centers, it is essentially a retrospective study with a limited sample size (n = 191). Second, consistent with the methodological limitations of many existing models [8], our model has not yet undergone external validation. Third, previous antimicrobial exposure history and MDRO colonization status were not included in the analysis. A substantial body of research has established their roles as independent risk factors for MDR infection: antimicrobial exposure can promote the colonization and proliferation of resistant bacteria by altering the microbiome and selective pressure [26]; patients colonized with multidrug-resistant Gram-negative bacteria (such as CRE) have a significantly increased risk of subsequent infection with the same organism, as demonstrated by a recent umbrella review and meta-analysis [27]. These variables are difficult to collect accurately and completely in a retrospective design, which is the main reason for their exclusion and which warrants further prospective cohort studies. Fourth, the definition of MDR relied on clinical culture and susceptibility results; MDR infection cases that are culture-negative or not submitted for testing were not captured, which may introduce a false-negative bias in outcome classification.

Conclusions

In this study, a 4-variable prediction model based on age, length of hospital stay, Lac, and PCT demonstrated good discrimination, calibration, and clinical utility for assessing the risk of MDR infection in patients with HBV-ACLF and bacterial infection. The model provides an intuitive quantitative tool for the early identification of high-risk MDR patients and the optimization of empirical antimicrobial therapy. Future multicenter, large-sample prospective studies are still warranted to externally validate the model and further evaluate its generalizability.

Abbreviations

ACLF
Acute-on-chronic liver failure
APASL
Asian Pacific Association for the Study of the Liver
AUC
Area under the receiver operating characteristic curve
BMI
Body mass index
CI
Confidence interval
CLSI
Clinical and Laboratory Standards Institute
CRE
Carbapenem-resistant Enterobacteriaceae
CRP
C-reactive protein
DCA
Decision curve analysis
EBV
Epstein-Barr virus
ESBL
Extended-spectrum β-lactamase
HBV
Hepatitis B virus
HCV
Hepatitis C virus
HE
Hepatic encephalopathy
HEV
Hepatitis E virus

ICU
Intensive Care Unit
IL-6
Interleukin-6
INR
International normalized ratio
i-MELD
Integrated model for end-stage liver disease
Lac
Lactate
LASSO
Least absolute shrinkage and selection operator
MDR
Multidrug-resistant
MDRO
Multidrug-resistant organism
MELD
Model for end-stage liver disease
MELD-Na
Model for end-stage liver disease combined with serum sodium
MRSA
Methicillin-resistant *Staphylococcus aureus*
OR
Odds ratio
PCT
Procalcitonin
ROC
Receiver operating characteristic
VRE
Vancomycin-resistant *Enterococcus*

Declarations

Ethics approval and consent to participate: This study was conducted in accordance with the Declaration of Helsinki. The study protocol was approved by the Ethics Committee of Tangdu Hospital of the Fourth Military Medical University (Approval No. GKJ-Y-202303-111), which served as the lead ethics committee for this two-center study. Due to the retrospective nature of the study, the requirement for written informed consent was waived by the ethics committee.

Consent for publication: Not applicable.

Availability of data and materials: The datasets generated and/or analysed during the current study are available from the corresponding author on reasonable request.

Competing interests: The authors declare that they have no competing interests.

Funding: This work was supported by the Shaanxi Provincial Major Disease Diagnosis and Treatment Center Construction Project (Department of Infectious Diseases, Second Affiliated Hospital, Air Force Medical University). The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Authors' contributions: BZH, HHF, DH, and LJQ conceived and designed the study. BZH, ZJY, HF, YDH, and XHK collected the data. BZH, WLX, YXF, and DYD analyzed the data. BZH wrote the first draft. WCL and LJQ supervised the study and revised the manuscript. All authors read and approved the final manuscript.

Acknowledgements: The authors would like to thank all the patients and healthcare workers who participated in this study.

References

1. Goran LG, Lita FA, Fierbinteanu-Braticevici C, editors. Acute-on-chronic liver failure: steps towards consensus. Diagnostics (Basel). 2025;15(6):751. 10.3390/diagnostics15060751
2. Xu Z, Zhang X, Chen J, Shi Y, Ji S. Bacterial Infections in Acute-on-chronic Liver Failure: Epidemiology, Diagnosis, Pathogenesis, and Management. J Clin Transl Hepatol. 2024;12(7):667–76. 10.14218/JCTH.2024.00137.
3. Cao Z, Wong F, Choudhury AK, Kamath PS, Topazian M, Torre A, CLEARED Consortium, et al. Global prevalence and characteristics of infections and clinical outcomes in hospitalised patients with cirrhosis: a prospective cohort study for the CLEARED Consortium. Lancet Gastroenterol Hepatol. 2024;9(11):997–1009. 10.1016/S2468-1253(24)00224-3.
4. Trad N, Mohamed G, Bizid S, Ben Abdallah H, Bouali R, Abdelli MN. Clinical impact of multidrug-resistant bacterial infections in patients with cirrhosis. Future Sci OA. 2024;10(1):FSO945. 10.2144/fsoa-2023-0160.
5. Terra C, de Mattos AZ, Chagas MS, Torres A, Wiltgen D, Souza BM, et al. Impact of multidrug resistance on the management of bacterial infections in cirrhosis. World J Clin Cases. 2023;11(3):534–44. 10.12998/wjcc.v11.i3.534.
6. European Association for the Study of the Liver. EASL Clinical Practice Guidelines on acute-on-chronic liver failure. J Hepatol. 2023;79(2):461–91. 10.1016/j.jhep.2023.04.021.
7. Marciano S, Piano S, Singh V, Caraceni P, Maiwall R, Alessandria C, et al. Development and external validation of a model to predict multidrug-resistant bacterial infections in patients with cirrhosis. Liver Int. 2024;44(11):2915–28. 10.1111/liv.16063.

8. Li P, Bai Y, Yuan X, Zhang H, Li F, Liu J, et al. Clinical prediction models for hospital-acquired infection of multi-drug-resistant organism in intensive care units: a systematic review. J Hosp Infect. 2026;173:294–306. 10.1016/j.jhin.2026.04.022.
9. Wang H, Yang C, Zhao M, Ren F, Wang X, Yan H, et al. Comparison and validation of multiple machine learning algorithms for predicting MDRO infection in catheter-related bloodstream patients: a multicenter cohort study. Microbiol Spectr. 2026;14(3):e03713–25. 10.1128/spectrum.03713-25.
10. Sarin SK, Choudhury A, Sharma MK, Maiwall R, Al Mahtab M, Rahman S, et al. Acute-on-chronic liver failure: consensus recommendations of the Asian Pacific association for the study of the liver (APASL): an update. Hepatol Int. 2019;13(4):353–90. 10.1007/s12072-019-09946-3.
11. Clinical and Laboratory Standards Institute. Performance standards for antimicrobial susceptibility testing. 34th ed. CLSI supplement M100. Wayne, PA: Clinical and Laboratory Standards Institute; 2024.
12. Magiorakos AP, Srinivasan A, Carey RB, Carmeli Y, Falagas ME, Giske CG, et al. Multidrug-resistant, extensively drug-resistant and pandrug-resistant bacteria: an international expert proposal for interim standard definitions for acquired resistance. Clin Microbiol Infect. 2012;18(3):268–81. 10.1111/j.1469-0691.2011.03570.x.
13. Toll JF, Sola E, Perez MA, Piano S, Cheng A, Subramanian AK, et al. Infections in decompensated cirrhosis: Pathophysiology, management, and research agenda. Hepatol Commun. 2024;8(10):e0539. 10.1097/HC9.0000000000000539.
14. Fernández J, Prado V, Trebicka J, Amoros A, Gustot T, Wiest R, et al. Multidrug-resistant bacterial infections in patients with decompensated cirrhosis and with acute-on-chronic liver failure in Europe. J Hepatol. 2019;70(3):398–411. 10.1016/j.jhep.2018.10.027.
15. Tamma PD, Heil EL, Justo JA, Mathers AJ, Satlin MJ, Bonomo RA. 2024 Guidance on the Treatment of Antimicrobial-Resistant Gram-Negative Infections. Clin Infect Dis. 2024;ciae403. 10.1093/cid/ciae403.
16. Turner AM, Kinsella P, Miller WR, Carter GP, Tran TT, Howden BP, et al. Therapeutic approach to difficult-to-treat multidrug-resistant enterococcal infections. Antimicrob Agents Chemother. 2025;69(10):e01060–24. 10.1128/aac.01060-24.
17. Gou J, Li Q, Fan N, Zhang C, Tang H, Wang X, et al. High accumulation of linezolid and its major metabolite in the serum of patients with hepatic and renal dysfunction is significantly associated with thrombocytopenia and anemia. Microbiol Spectr. 2025;13(7):e02493–24. 10.1128/spectrum.02493-24.
18. Bamogaddam RF, Alamer A, Alqarni S, Almotairi MM, Almakrami AA, Alharbi AM, et al. Incidence and predictors of vancomycin nephrotoxicity and mortality in patients with chronic liver disease: a two-center retrospective cohort study. BMC Infect Dis. 2025;25(1):375. 10.1186/s12879-025-10763-3.
19. Meena DS, Bohra GK, Malik M, Lodha N, Kumar D, Midha N, et al. Association of procalcitonin levels with multidrug-resistant bloodstream infections: a prospective cohort study. BMC Infect Dis. 2026;26:1417. 10.1186/s12879-026-13680-1.

20. Garcia-Alvarez M, Marik PE, Bellomo R. Sepsis-associated hyperlactatemia. Crit Care. 2014;18(5):503. 10.1186/s13054-014-0503-3.
21. Evans L, Rhodes A, Alhazzani W, Antonelli M, Coopersmith CM, French C, et al. Surviving Sepsis Campaign: international guidelines for management of sepsis and septic shock 2021. Crit Care Med. 2021;49(11):e1063–143. 10.1097/CCM.0000000000005337.
22. Nazir A, Nazir A, Zuhair V, Aman S, Sadiq SUR, Hasan AH, et al. The Global Challenge of Antimicrobial Resistance: Mechanisms, Case Studies, and Mitigation Approaches. Health Sci Rep. 2025;8(7):e71077. 10.1002/hsr2.71077.
23. Korkmaz N, Senturk GC, Yilmaz FA, Kul G, Guldoğan B, Yilmaz HM, et al. Early procalcitonin kinetics as an independent predictor of mortality in multi-drug resistant Klebsiella spp. bloodstream infections: a retrospective cohort study. BMC Infect Dis. 2026;26:632. 10.1186/s12879-026-12898-3.
24. Theodorakis N, Feretzakis G, Hitas C, Kreouzi M, Kalantzi S, Spyridaki A, et al. Antibiotic resistance in the elderly: mechanisms, risk factors, and solutions. Microorganisms. 2024;12(10):1978. 10.3390/microorganisms12101978.
25. Wang N, Tao S, Chen L. Construction and validation of HBV-ACLF bacterial infection diagnosis model based on machine learning. BMC Infect Dis. 2025;25:847. 10.1186/s12879-025-11199-5.
26. Ho CS, Wong CTH, Aung TT, et al. Antimicrobial resistance: a concise update. Lancet Microbe. 2025;6(1):100947. 10.1016/j.lanmic.2024.07.010.
27. Woodhouse EW, Alsoubani M, Roach DJ, Flynn DB, LaValley M, Sheridan K, et al. Infection risk associated with colonization by multidrug-resistant gram-negative bacteria: an umbrella review and meta-analysis. Open Forum Infect Dis. 2025;12(7):ofaf365. 10.1093/ofid/ofaf365.

Figures

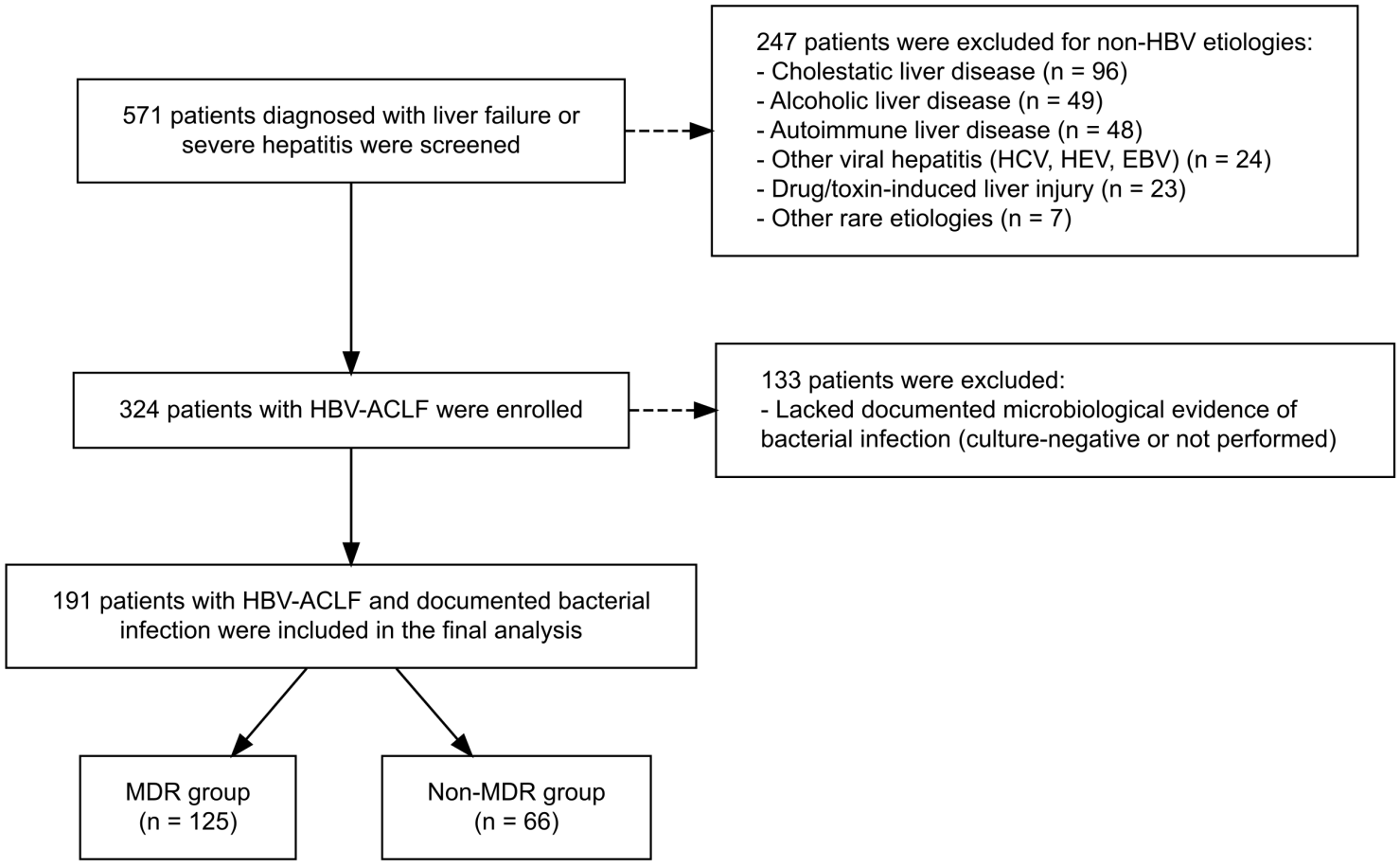


Figure 1

Flowchart of patient enrollment. A total of 571 patients with liver failure or severe hepatitis were initially screened. After excluding 247 patients with non-HBV etiologies, 324 patients with HBV-ACLF were enrolled. Following the exclusion of 133 patients lacking documented microbiological evidence, 191 patients with bacterial infection were included in the final analysis, comprising 125 in the MDR group and 66 in the non-MDR group. HBV-ACLF, hepatitis B virus-related acute-on-chronic liver failure; MDR, multidrug-resistant.

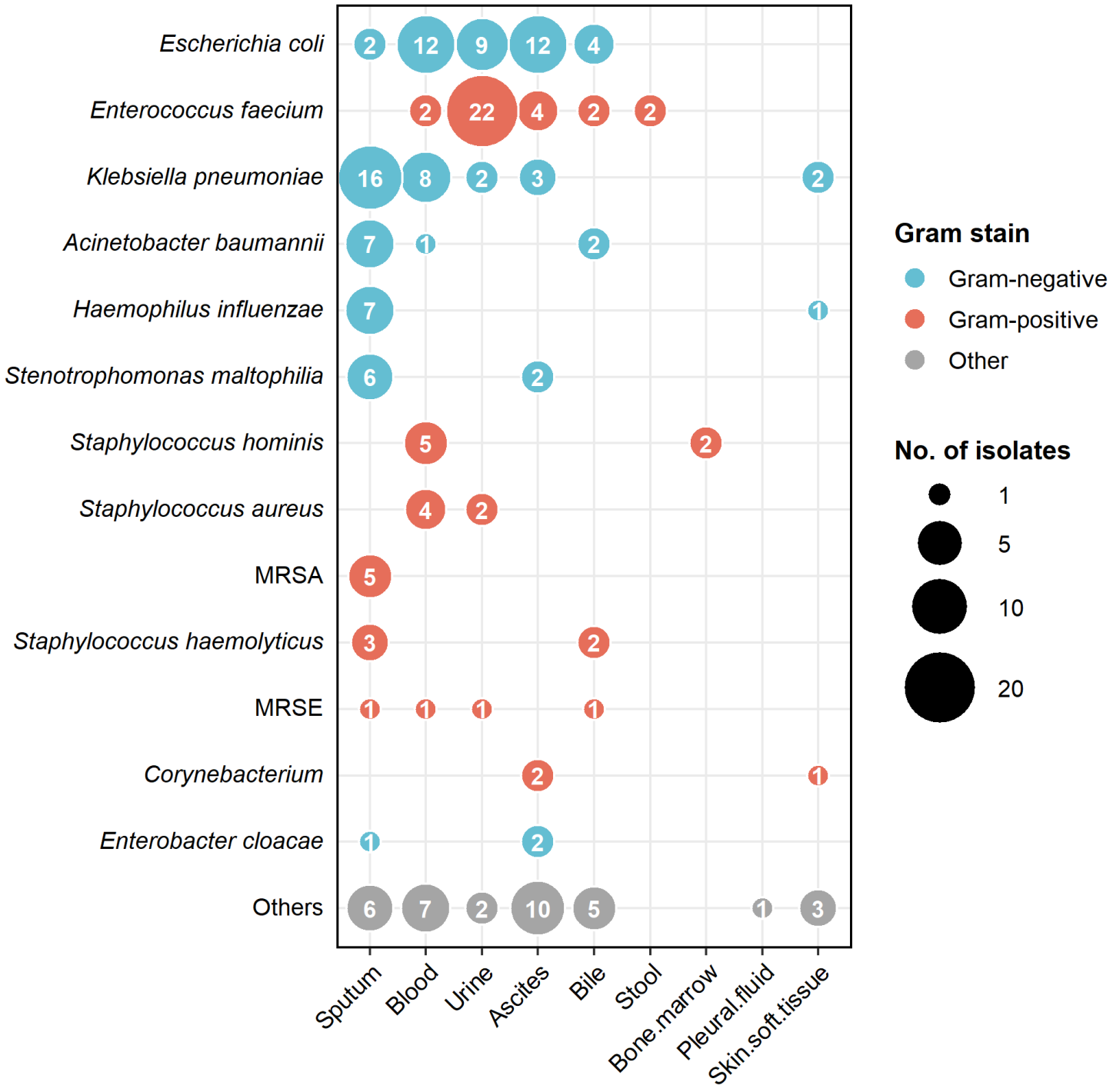


Figure 2

Distribution of bacterial isolates by specimen source. Distribution of bacterial isolates by specimen source in patients with HBV-ACLF and bacterial infection (*n* = 191). Bubble size represents the number of isolates. Colors indicate Gram stain: red for Gram-positive bacteria, blue for Gram-negative bacteria, and gray for "Others" (rare species with ≤2 isolates each, grouped together). Species are ordered by total isolate count. MRSA, methicillin-resistant *Staphylococcus aureus*; MRSE, methicillin-resistant *Staphylococcus epidermidis*.

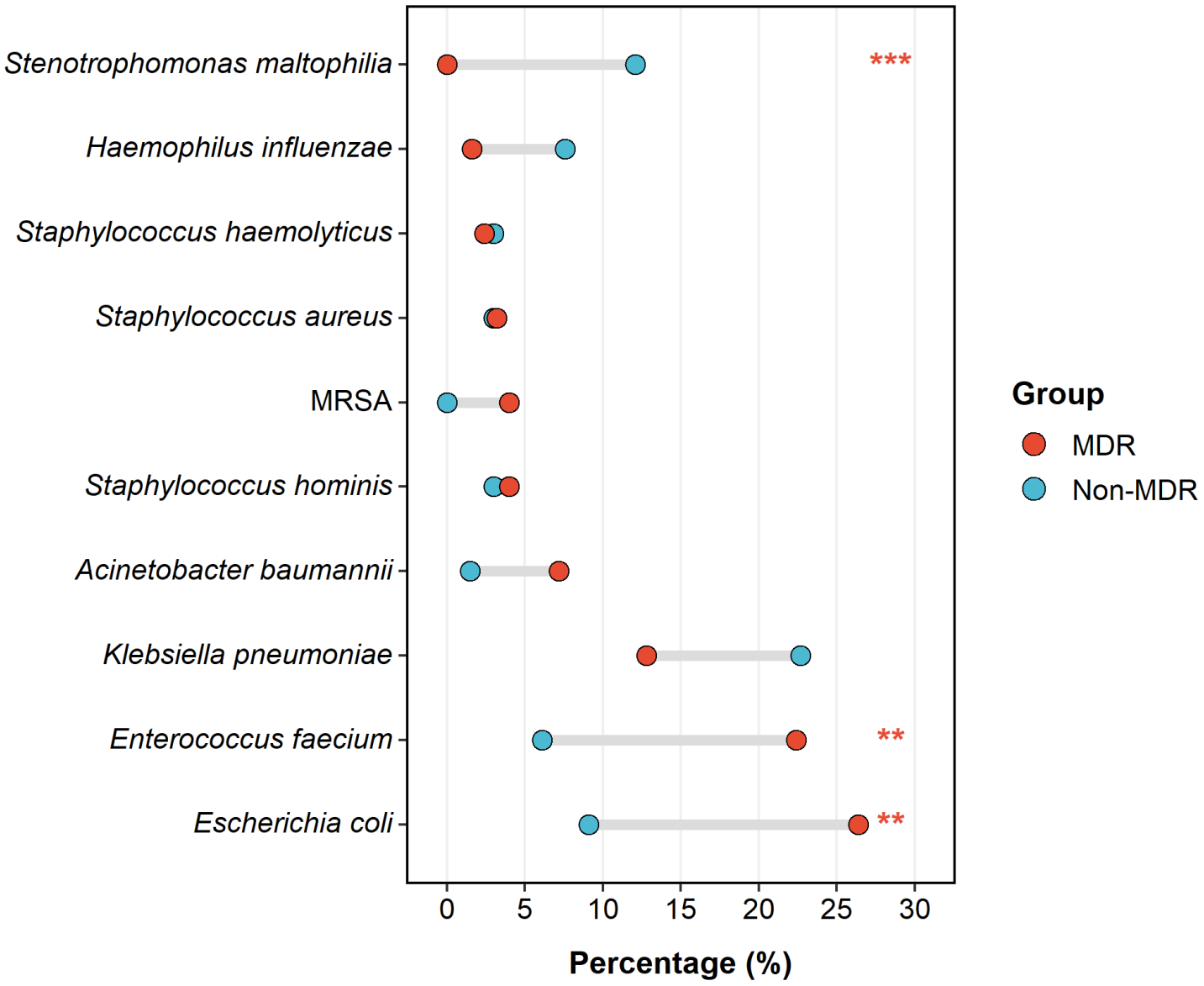


Figure 3

Distribution of predominant bacteria in MDR and Non-MDR groups. Dumbbell plot showing the proportion of each bacterial species within the MDR group (red) and the Non-MDR group (blue). Species are ordered by ascending MDR proportion. Asterisks indicate statistically significant differences between the two groups: *P < 0.05, **P < 0.01, ***P < 0.001. MDR, multidrug-resistant.

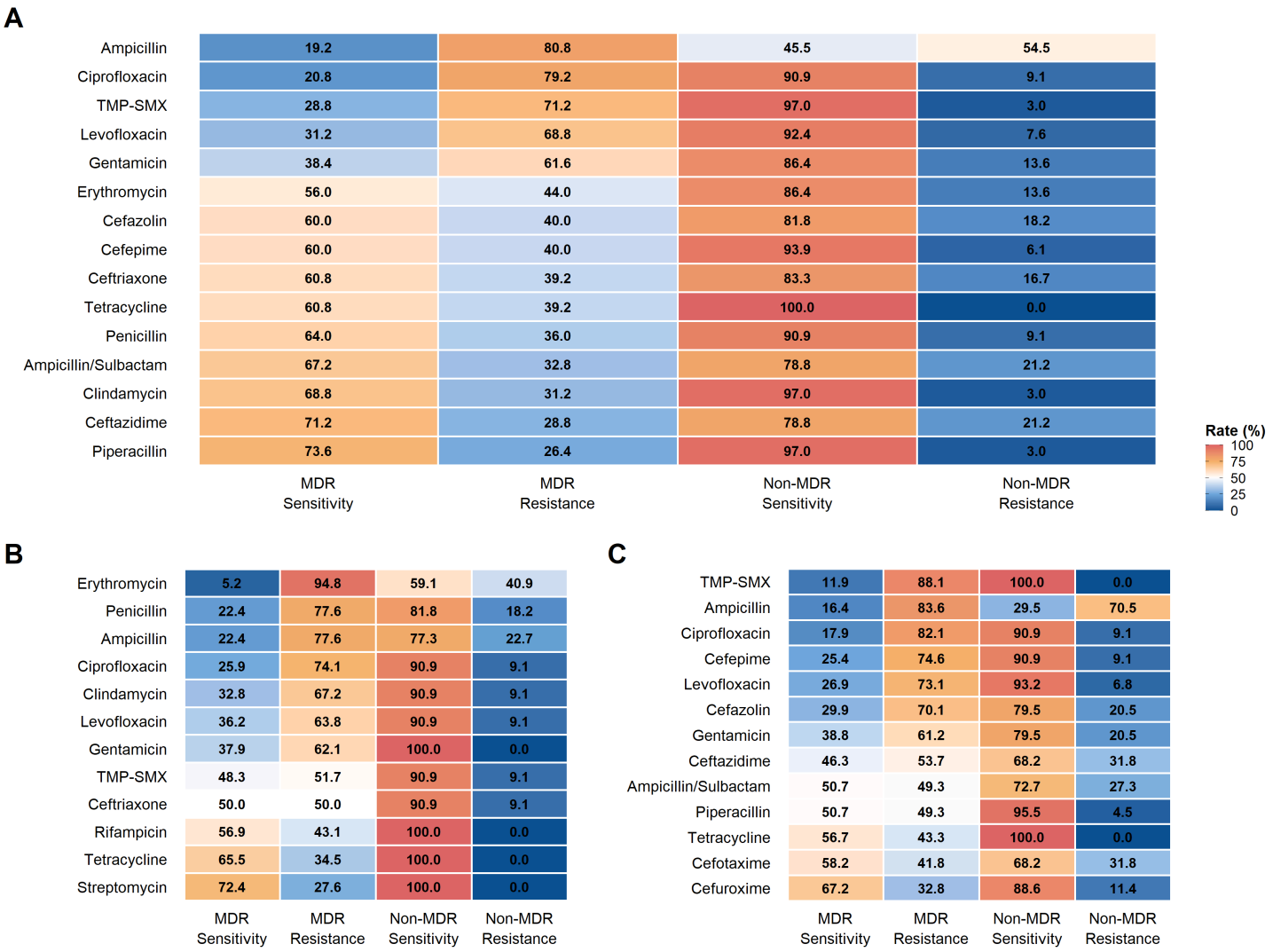


Figure 4

Antibiotic sensitivity and resistance profiles of MDR and Non-MDR isolates. Heatmaps showing the sensitivity and resistance rates of (A) overall bacteria (*n* = 191), (B) Gram-positive bacteria (*n* = 80), and (C) Gram-negative bacteria (*n* = 111) to selected antibiotics. For each antibiotic, four consecutive columns represent: MDR sensitivity, MDR resistance, Non-MDR sensitivity, and Non-MDR resistance. Color intensity indicates the rate (%): blue represents sensitivity, red represents resistance, and white indicates 0%; darker colors correspond to higher rates. Antibiotics within each panel are ordered by ascending MDR sensitivity rate. MDR, multidrug-resistant.

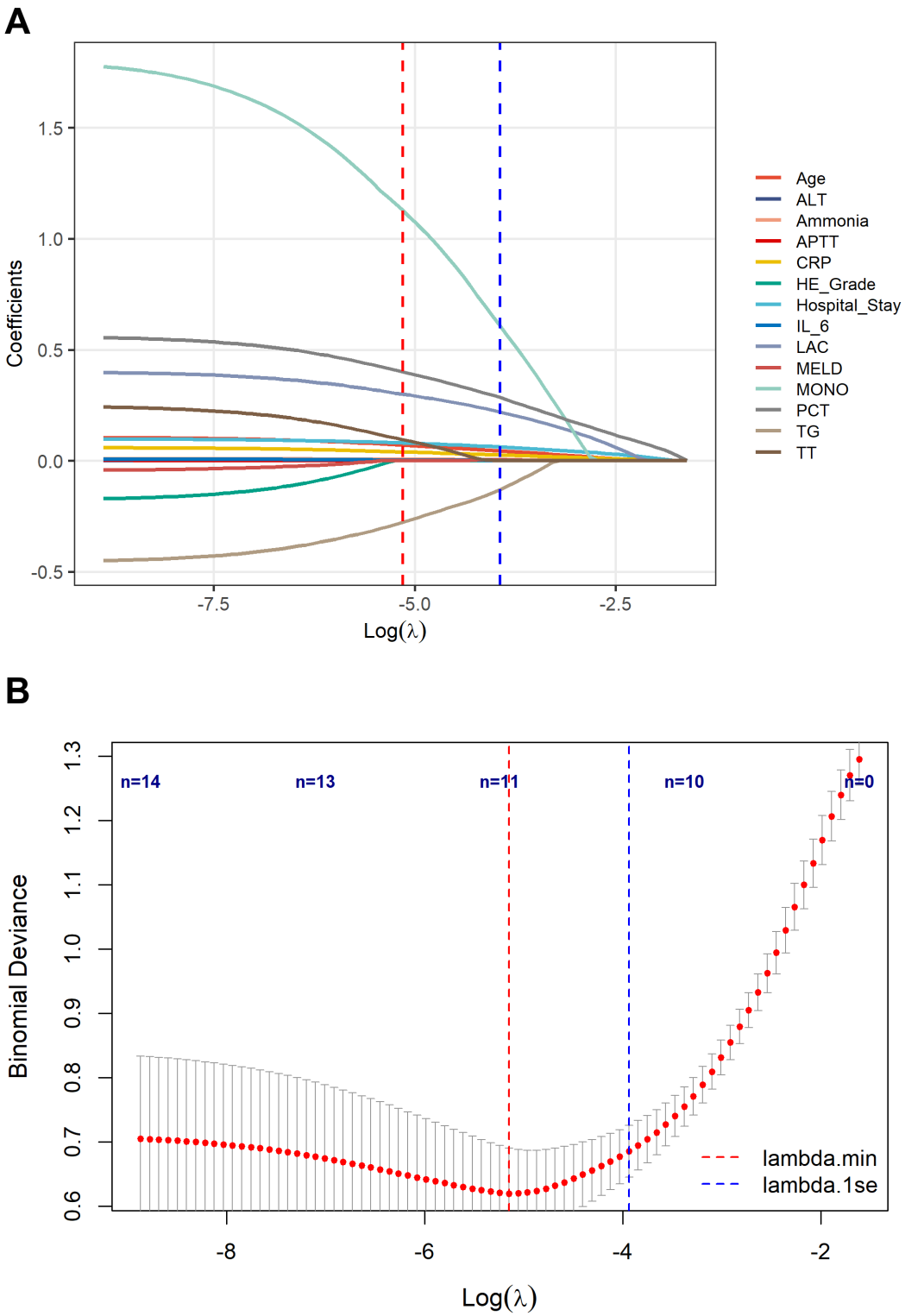


Figure 5

LASSO regression for variable selection. (A) LASSO coefficient paths of the 14 candidate variables. The x-axis represents log(λ) and the y-axis represents the coefficient value. Each curve corresponds to one variable. The red dashed vertical line indicates the optimal λ selected by 10-fold cross-validation (lambda.min), and the blue dashed vertical line indicates the most parsimonious λ within one standard error (lambda.1se). (B) Ten-fold cross-validation curve. The x-axis shows log(λ), and the y-axis shows the

binomial deviance. Red dots represent the mean cross-validated error, and error bars represent standard errors. The red and blue dashed lines indicate lambda.min and lambda.1se, respectively. Numbers inside the panel (*n* = …) indicate the number of non-zero coefficients at selected λ values. LASSO, least absolute shrinkage and selection operator.

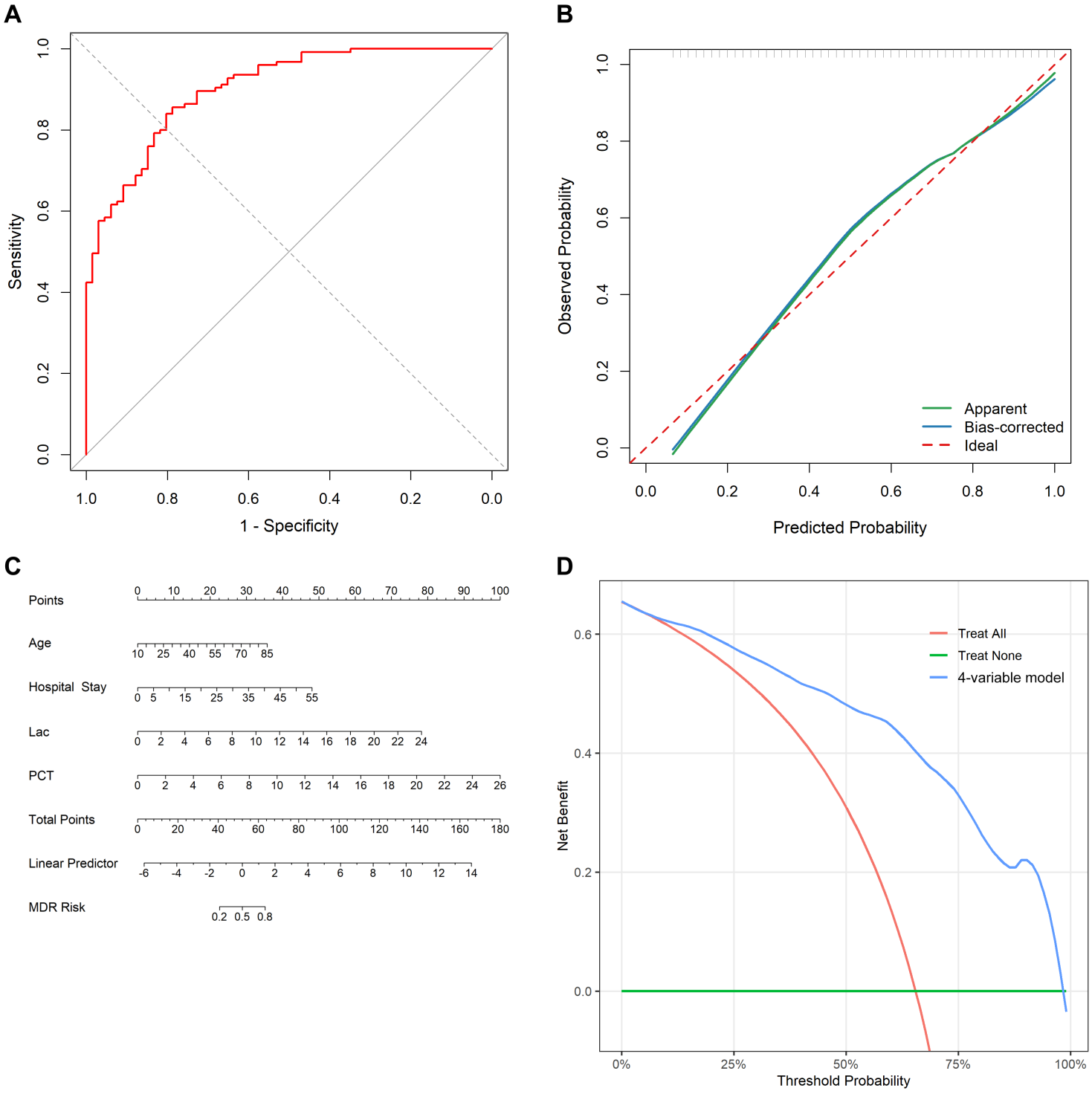


Figure 6

Construction and internal validation of the 4-variable prediction model for MDR infection.(A) Receiver operating characteristic (ROC) curve of the 4-variable model. The area under the curve (AUC) was 0.902

(95% CI 0.859–0.945). (B) Calibration curve with 1000 bootstrap resamples. The green solid line indicates apparent performance, the blue solid line indicates bias-corrected performance, and the red dashed line indicates the ideal reference. (C) Nomogram for predicting the risk of MDR infection based on age, hospital stay, Lac, and PCT. To use the nomogram, locate each variable value on its axis, read the corresponding point score, sum the points, and project the total points onto the MDR Risk axis to estimate the probability of MDR infection. (D) Decision curve analysis (DCA). The red curve represents the "treat all" strategy, the green horizontal line represents the "treat none" strategy, and the blue curve represents the 4-variable model. The model provides a higher net benefit than the two reference strategies across a wide range of threshold probabilities (approximately 0%–90%). MDR, multidrug-resistant; Lac, lactate; PCT, procalcitonin; AUC, area under the receiver operating characteristic curve; DCA, decision curve analysis.

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- Additionalfile1.pdf