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# Repeat isolates and apparent trends in multidrug-resistant organism detection: a retrospective analysis of aggregated hospital surveillance data using an isolate-to-patient ratio

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## Abstract

# Background

Routine hospital reports of multidrug-resistant organisms (MDROs) often count every isolate, whereas surveillance standards count each patient once. Infection prevention teams that work from monthly aggregate reports cannot remove duplicates themselves. We examined whether repeat isolates from the same patient distorted the apparent trend in MDRO detection and whether the distortion could be measured from aggregate counts alone.

# Methods

We retrospectively analyzed 32 consecutive monthly surveillance reports (January 2024–August 2026) from a 1,200-bed general hospital in China, harmonized to one set of definitions. Repeat isolation was measured with the isolate-to-patient ratio (IPR; key MDRO isolates per MDRO-positive patient), which could be calculated from March 2025, and was monitored with a *u*-chart. For March–August 2025 versus March–August 2026 we compared isolate-based and patient-based MDRO densities per 1,000 patient-days and standardized the 2026 isolate-based proportion to the 2025 IPR.

# Results

For January–August, the isolate-based MDRO proportion rose from 13.9% in 2024 to 19.4% in 2026 (*P* for trend < 0.001). Between the two March–August periods, isolate-based density rose by 31% (rate ratio 1.31; 95% confidence interval [CI] 1.14–1.50) and patient-based density by 11% (rate ratio 1.11; 95% CI 0.93–1.33); the IPR rose from 1.60 to 1.89 (ratio 1.18; 95% CI 1.05–1.33). The IPR exceeded its upper control limit in May–July 2026, during an intensive care unit cluster of carbapenem-resistant *Klebsiella pneumoniae* (CRKP) in which three patients with bloodstream infection yielded 18 isolates in one month. Standardization to the 2025 IPR removed about half of the between-year difference in the isolate-based proportion (49%–59%, depending on the method). The rise in CRKP persisted in seasonally adjusted sensitivity analyses; the incidence of healthcare-associated infection caused by MDROs did not change measurably.

# Conclusions

Counting every isolate exaggerated an increase in MDROs: about half of the apparent rise reflected more isolates per patient rather than more patients. Where patient-level deduplication is not at hand, the IPR can be calculated from two counts in a routine report, and charting it marks the months in which repeat culturing and clustering both need review. The approach was tested in one hospital and should be validated against patient-level first-isolate data.

# Background

Bacterial antimicrobial resistance was directly responsible for an estimated 1.14 million deaths worldwide in 2021 and was associated with 4.71 million [1]. Carbapenem-resistant *Acinetobacter baumannii* (CRAB) and carbapenem-resistant Enterobacterales (CRE) are critical-priority pathogens on the World Health Organization list [2]. In China, the CHINET surveillance network recorded a rise in imipenem resistance among *Klebsiella pneumoniae* isolates from 2.9% in 2005 to 25.0% in 2018; in 2025 the figure was 21.9% for *K. pneumoniae* and 72.0% for *A. baumannii* [3, 4].

Hospitals track multidrug-resistant organisms (MDROs) to judge whether resistance is worsening and whether control measures work, and the answer depends on how isolates are counted. The Clinical and Laboratory Standards Institute (CLSI) M39 guideline keeps only the first isolate of a species from each patient in each analysis period [5, 6]. The position paper of the Society for Healthcare Epidemiology of America and the Healthcare Infection Control Practices Advisory Committee (SHEA/HICPAC) and the National Healthcare Safety Network MDRO module likewise exclude duplicates and use patient-days as the denominator [7, 8]. Including repeat isolates raises resistance estimates for hospital isolates [9] and is a recognized source of surveillance bias [10]; cumulative antibiograms shift with the deduplication rule [11], and later isolates are less susceptible than first isolates [12].

Infection prevention programs do not always work with deduplicated data. China's 2015 national quality indicators for infection control defined the MDRO "detection rate" as resistant isolates divided by all isolates of the species, with every isolate counted [13]. The 2024 revision replaced it with the incidence of healthcare-associated infection (HAI) caused by specified MDROs per 1,000 patient-days [14], but isolate-based rates remain common in monthly hospital reports, including ours. Using patient-level records from one hospital, Li et al. showed that deduplication lowered the proportion of carbapenem-resistant *K. pneumoniae* (CRKP) from 33.52% to 24.78% [15]. That study measured the bias in the level of resistance. What infection prevention teams have to interpret month by month, however, is the trend, and many of them work from aggregate reports that cannot be traced back to individual patients. A constant share of duplicates shifts the level of an isolate-based rate but leaves its trend intact; the trend is distorted only when the number of isolates per patient changes over time, as it may when a few critically ill patients are cultured repeatedly or when a cluster occurs. Whether such a change can be detected, and its effect on a trend estimated, from aggregate counts alone has not been examined.

We used 32 consecutive monthly MDRO reports from one hospital. Our objectives were (1) to describe the apparent trend in isolate-based measures of key MDROs after harmonizing definitions across a change in report format; (2) to estimate, from the isolate counts, patient counts and patient-days in the reports, how much of the change between comparable periods was associated with a change in the number of isolates per patient; and (3) to assess whether a control chart of the isolate-to-patient ratio (IPR) identifies the months in which an isolate-based rate should not be taken at face value. No hypotheses were prespecified.

## Methods

# Study design, setting, and data source

This retrospective study of routinely collected surveillance data is reported according to the RECORD statement [16] (Additional file 2). The setting was one campus of a general hospital in Chengdu, southwest China, with 1,200 beds, 23 of them in the intensive care unit (ICU). The data source was the 32 monthly MDRO surveillance reports issued by the infection prevention and control department from January 2024 through August 2026, which are compiled from the laboratory information system and the HAI surveillance system. Each report gives, by ward, culture submissions, isolates and key MDRO isolates, together with the origin of CRE isolates, HAI episodes caused by key MDROs, and patient-days. From March 2025, the reports also give the number of patients from whom a key MDRO was isolated. No new hospital-wide MDRO intervention was introduced during the study period.

Three investigators extracted the data with a single parsing program and checked them by hand against the reports. The database passed 18 internal consistency checks; 96 data issues were logged and their handling recorded (Additional file 1: Supplementary Methods). Only the monthly aggregate values in the reports were used. Patient-level laboratory records were not retrieved, so the analysis is confined to the information available to an infection prevention team that works from such reports.

## Analysis periods

Because patient counts began in March 2025 and the series ended in August 2026, three periods were used. The full 32 months were used for monthly trends in isolate-based proportions and in HAI incidence. January–August of 2024, 2025 and 2026 were compared for changes between years. The 18 months from March 2025 through August 2026 were used for every analysis that involved patient counts, with March–August 2025 and March–August 2026 as the paired comparison periods and March–December 2025 as the baseline for the control chart.

## Isolates and definitions

We included isolates of seven target organisms (*A. baumannii*, *Pseudomonas aeruginosa*, *Escherichia coli*, *K. pneumoniae*, *Staphylococcus aureus*, *Enterococcus faecium* and *Enterococcus faecalis*) from clinical specimens of inpatients. Specimens from outpatient and emergency departments, active screening specimens and environmental samples were excluded. Repeat isolates from the same patient were not removed, because they were the object of study; isolates classified as contaminants also stayed in the counts, as in the source reports.

Organisms were identified by matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (VITEK MS; bioMérieux, Marcy-l'Étoile, France). Susceptibility was tested with an automated system (VITEK 2 Compact; bioMérieux) and interpreted with the breakpoints of CLSI M100,

34th edition [17]. Methods and breakpoints were unchanged throughout the study, so breakpoint revisions cannot account for changes over time [18]. Key MDROs, defined with reference to international and national definitions [19, 20], were CRAB, carbapenem-resistant *P. aeruginosa* (CRPA), methicillin-resistant *S. aureus* (MRSA), vancomycin-resistant *Enterococcus* (VRE), carbapenem-resistant *E. coli* (CRECO) and CRKP, the organisms named in the 2024 national indicators [14] (Additional file 1: Supplementary Methods).

## Harmonization of definitions

The report format was revised in March 2025: CRE was limited to CRKP and CRECO, and other Enterobacterales were dropped from the denominator of the overall rate. We applied the revised definitions to the whole series, recalculating January 2024 through February 2025 from itemized counts in the original reports. CRE here means CRECO plus CRKP. The origin of CRE isolates could not be separated by species before the revision and was analyzed from March 2025 onward.

## Measures

The measures are summarized in Table 1.

Table 1
Surveillance measures derived from the monthly aggregate reports

| Measure | Numerator | Denominator | Months available | Inflated by repeat isolates |
|---|---|---|---|---|
| Isolate-based MDRO proportion, % | Key MDRO isolates | Isolates of the corresponding target species | 32 | Yes |
| Isolate-based MDRO density | Key MDRO isolates | Patient-days (per 1,000) | 32 | Yes |
| Patient-based MDRO density | MDRO-positive patients* | Patient-days (per 1,000) | 18 | No |
| Isolate-to-patient ratio (IPR) | Key MDRO isolates | MDRO-positive patients* | 18 | Measures it |
| Incidence of HAI caused by key MDROs | New HAI episodes | Patient-days (per 1,000) | 32 | No |

HAI, healthcare-associated infection; MDRO, multidrug-resistant organism.

*Counted once per patient and organism in each month; reported from March 2025.

*Isolate-based MDRO proportion.* This is the “detection rate” of the source reports: isolates of a given MDRO divided by isolates of the corresponding species in the same period, with no deduplication. The

overall proportion is the sum of the six key MDROs over the sum of the seven target organisms. Because it depends on species mix, we also standardized it directly to the pooled species distribution for January–August of the three years.

*HAI incidence.* HAIs were identified according to national surveillance and diagnostic standards [21, 22]. Following the 2024 indicator [14], incidence was the number of new HAI episodes caused by key MDROs per 1,000 patient-days. Repeated isolation of the same organism from the same infection site counted once, so this measure does not rise with repeat cultures.

*Repeat isolation.* The reports count patients with a key MDRO (MDRO-positive patients) per patient and organism each month: a patient with the same key MDRO from several wards or specimen types is counted once, and a patient with two different key MDROs is counted once for each. The IPR is the number of key MDRO isolates divided by the number of MDRO-positive patients in the same period. Because patients are counted per organism, the IPR reflects repeated isolation of the same organism from the same patient and not co-detection of several MDROs.

*Isolate-based and patient-based density.* Isolate-based density is the number of key MDRO isolates, and patient-based density the number of MDRO-positive patients, per 1,000 patient-days. We call these densities and not incidence densities because both include colonization and organisms present on admission, and a patient who remains positive is counted again in each month. In any period the ratio of the two densities equals the IPR. The ratio of the IPRs of two periods therefore equals the isolate-based rate ratio divided by the patient-based rate ratio, and measures directly how far the isolate-based change exceeds the patient-based change.

*IPR-standardized proportion.* This estimates what the 2026 isolate-based proportion would have been had repeat isolation stayed at its 2025 intensity. The IPR for March–August 2025 was multiplied by the number of MDRO-positive patients in the same months of 2026, and the resulting expected number of isolates was divided by the number of target-organism isolates in that period. This primary version adjusts only the numerator. Because the excess MDRO isolates (observed minus expected) are also counted in the denominator, we repeated the calculation with the excess removed from both. Neither version adjusts for a change in repeat isolation among the other isolates of the target organisms, which the reports do not allow us to measure.

*Control chart.* To identify months with an unusually high IPR, we applied *u*-chart principles [23]. With March–December 2025 as the baseline, we calculated the mean number of additional isolates per patient, $\bar{u}$ = (isolates − patients)/patients. The upper control limit for month $i$ was $1 + \bar{u} + 3\sqrt{(\bar{u}/n_i)}$, where $n_i$ is the number of MDRO-positive patients in that month. The limits were calculated from the baseline months only and then applied to all 18 months. Ten baseline months are fewer than the 20 to 25 usually advised for setting control limits, so the limits should be regarded as provisional. We also applied a simple alert threshold of IPR greater than 2.0, chosen after inspection of the data.

# Statistical analysis

Proportions were compared with the $\chi^2$ or Fisher exact test, with Bonferroni correction for pairwise comparisons among the three years ($\alpha = 0.0167$). Linear trend across years was assessed with the Cochran–Armitage test, with Holm adjustment across the six MDROs. Monthly trends were modeled by quasi-binomial logistic regression with time in years as the covariate, and results are given as the odds ratio (OR) per year. A segmented model added a level-change term for 2026 and, to test continuity of the harmonized series, a second term at the March 2025 revision [24]. We adjusted for seasonality with one sine–cosine pair of 12-month period and checked residual autocorrelation with the Ljung–Box test. The two densities and HAI incidence were compared between periods with rate ratios (RRs), and monthly HAI incidence was modeled by Poisson regression with log patient-days as the offset.

The IPRs of the two March–August periods were compared as a ratio. Its 95% confidence interval (CI) came from a parametric bootstrap with 20,000 resamples, in which the numbers of MDRO-positive patients and of repeat isolates (isolates minus patients) in each period were drawn from Poisson distributions. We checked this comparison in two ways. First, we estimated the Pearson dispersion of the monthly numbers of repeat isolates, with MDRO-positive patients as the exposure, and widened the interval by the square root of the dispersion, using the *t* distribution with 10 degrees of freedom. Second, we compared the IPR of each calendar month of 2026 with that of the same month of 2025 with an exact sign test. The 95% CIs of the IPR-standardized proportion and of its difference from 2025 came from the same kind of bootstrap.

In sensitivity analyses we excluded months in which the IPR exceeded the upper control limit or 2.0, used March–December 2025 as the reference period for standardization, and adjusted for seasonality. The excluded months, the alert threshold and the model breakpoint were chosen after the data had been examined, so these analyses are exploratory. For non-significant comparisons of rates we report the smallest RR detectable with 80% power. Analyses were done in Python 3.12; tests were two-sided, with $P < 0.05$ considered significant. Details are in Additional file 1: Supplementary Methods.

## Results

# Overview

Over the 32 months, 101,598 culture submissions yielded 18,735 isolates. Of 11,982 isolates of the seven target organisms, 1,886 (15.74%) were key MDROs: CRKP, 715 (37.91%); MRSA, 448 (23.75%); CRAB, 374 (19.83%); CRPA, 223 (11.82%); CRECO, 113 (5.99%); and VRE, 13 (0.69%). Case mix shifted over time: for January–August the ICU's share of culture submissions rose from 17.24% in 2024 to 19.17% in 2026, and the distributions of target organisms and of specimen sources also changed (all $P \leq 0.001$; Additional file 1: Table S1).

# Annual comparison

For January–August, the overall isolate-based proportion of key MDROs was 13.92% in 2024, 15.23% in 2025 and 19.44% in 2026 (*P* for trend < 0.001; Table 2); standardized for species mix, it was 13.82%, 15.85% and 18.70% (*P* for trend < 0.001). CRKP and CRAB showed upward trends (Holm-adjusted *P* < 0.001 and *P* = 0.024), whereas CRPA, MRSA, CRECO and VRE did not change significantly. Pairwise, the overall proportion did not differ between 2024 and 2025 (*P* = 0.152) and was higher in 2026 than in either earlier year (both *P* < 0.001).

Table 2
Isolate-based proportions of key multidrug-resistant organisms, January–August 2024 to 2026

| Organism | 2024 | 2025 | 2026 | $\chi^2$ | *P* value | *P* for trend |
|---|---|---|---|---|---|---|
| CRAB | 75/168 (44.64) | 85/153 (55.56) | 139/235 (59.15) | 8.56 | 0.014 | 0.005 |
| CRPA | 50/317 (15.77) | 49/329 (14.89) | 66/365 (18.08) | 1.39 | 0.499 | 0.397 |
| MRSA | 123/465 (26.45) | 104/392 (26.53) | 86/351 (24.50) | 0.51 | 0.774 | 0.548 |
| VRE | 3/327 (0.92) | 2/311 (0.64) | 3/297 (1.01) | — | 0.910* | — |
| CRECO | 26/1,072 (2.43) | 28/1,142 (2.45) | 35/1,109 (3.16) | 1.46 | 0.482 | 0.288 |
| CRKP | 138/633 (21.80) | 185/648 (28.55) | 258/663 (38.91) | 46.09 | < 0.001 | < 0.001 |
| Overall | 415/2,982 (13.92) | 453/2,975 (15.23) | 587/3,020 (19.44) | 36.82 | < 0.001 | < 0.001 |
| Overall, species standardized† | 13.82 (12.66–14.97) | 15.85 (14.64–17.05) | 18.70 (17.46–19.93) | — | — | < 0.001‡ |

Values are No. of resistant isolates/No. of isolates of the species (%) unless otherwise indicated. *P* for trend is from the Cochran–Armitage test; after Holm adjustment, *P* < 0.001 for CRKP and *P* = 0.024 for CRAB. For carbapenem-resistant Enterobacterales (CRECO plus CRKP), the proportions were 9.62% (164/1,705), 11.90% (213/1,790) and 16.53% (293/1,772) (*P* for trend < 0.001).

CRAB, carbapenem-resistant *Acinetobacter baumannii*; CRECO, carbapenem-resistant *Escherichia coli*; CRKP, carbapenem-resistant *Klebsiella pneumoniae*; CRPA, carbapenem-resistant *Pseudomonas aeruginosa*; MRSA, methicillin-resistant *Staphylococcus aureus*; VRE, vancomycin-resistant *Enterococcus*.

*Fisher–Freeman–Halton exact test. †Directly standardized to the pooled distribution of target organisms for January–August of the three years; values are % (95% CI). ‡Weighted linear trend test.

# Monthly trends

Across the 32 months, the odds that a target-organism isolate was a key MDRO rose by 17% per year (OR per year 1.17; 95% CI 1.04–1.31; *P* = 0.008). For CRKP the OR per year was 1.46 (95% CI 1.17–1.81; *P* < 0.001), and 1.45 (95% CI 1.21–1.74) after adjustment for a seasonal pattern with lower values from September through November. The monthly trend for CRAB was not significant (OR per year 1.22; 95% CI 0.92–1.62; *P* = 0.159). In the segmented model the 2024–2025 slope was flat (OR per year 0.99; 95% CI 0.84–1.17), with a level increase in 2026 (OR 1.44; 95% CI 1.09–1.92; *P* = 0.012). Adding a level-change term for the March 2025 report revision showed no step at that point (OR 0.78; 95% CI 0.55–1.12; *P* = 0.178) and lowered the 2026 estimate to 1.32 (95% CI 0.96–1.80; *P* = 0.084).

The increase in 2026 did not start in January: the overall proportion for January–February was 15.60% (105/673), against 15.36% (98/638) a year earlier (*P* = 0.904). The rise was concentrated in March through August. In the second quarter the CRKP and overall proportions reached 45.67% and 22.36%, and both fell back somewhat in July–August (Fig. 1).

# Repeat isolation and the IPR control chart

From March 2025 through April 2026 the monthly IPR ranged from 1.30 to 1.81. It rose to 2.16 in May 2026 (119 isolates from 55 patients) and 2.11 in June (80 from 38), was 1.95 in July (76 from 39), and returned to 1.58 in August (Fig. 2). Against a baseline of 1.56, these three months were above the upper control limit and all other months were within it. The 2.0 threshold flagged May and June.

The out-of-control months coincided with a cluster in the ICU. In May 2026 the ICU had 49 key MDRO isolates, 32 of them CRKP from nine patients. Four of these patients had CRKP HAIs, including three with bloodstream infection who alone contributed 18 isolates. The infection prevention department opened a cluster investigation that month and found that cases clustered in time and place and shared a susceptibility profile. No new cases occurred after environmental disinfection and reinforced control measures. Molecular typing was not done (Additional file 1: Supplementary Results).

# Isolate-based versus patient-based change

Between March–August 2025 and March–August 2026, key MDRO isolates increased from 355 to 482 and MDRO-positive patients from 222 to 255 (Table 3). Isolate-based density rose by 31% (RR 1.31; 95% CI 1.14–1.50). Patient-based density rose by 11% (RR 1.11; 95% CI 0.93–1.33), an increase that the data could neither confirm nor exclude: the comparison had 80% power only for an RR of about 1.29 or more. The IPR rose from 1.60 to 1.89, a ratio of 1.18 (95% CI 1.05–1.33), so the isolate-based increase exceeded the patient-based increase by more than chance would explain. The monthly numbers of repeat isolates were overdispersed (Pearson dispersion 2.10); with the interval widened accordingly, the ratio remained above 1 (95% CI 1.02–1.37; *P* = 0.028). The IPR was higher in 2026 than in 2025 in each of the six calendar months (exact sign test, *P* = 0.031).

The isolate-based proportion rose from 15.19% to 20.54%, a difference of 5.35 percentage points. At the 2025 IPR, the 255 MDRO-positive patients of 2026 would have yielded 408 isolates instead of 482. The

IPR-standardized proportion for 2026 was 17.37% (95% CI 14.99%–20.06%), which left a difference of 2.18 percentage points (95% CI − 0.68 to 5.25); on the point estimates, 59% of the increase was associated with the higher IPR. With the 74 excess isolates also removed from the denominator, the standardized proportion was 17.94% (95% CI 15.84%–20.17%) and the residual difference 2.75 percentage points (95% CI 0.18 to 5.40), so that 49% of the increase was associated with the higher IPR. With March–December 2025 as the reference, the residual difference under the primary version was 1.75 percentage points (95% CI − 0.90 to 4.58).

Table 3
Isolate-based and patient-based measures of key multidrug-resistant organisms, March–August 2025 and 2026

| Measure | March–August 2025 | March–August 2026 | Effect estimate (95% CI) | *P* value |
|---|---|---|---|---|
| Key MDRO isolates/MDRO-positive patients, No. | 355/222 | 482/255 | — | — |
| Isolate-to-patient ratio (IPR) | 1.60 | 1.89 | Ratio 1.18 (1.05–1.33)* | 0.028† |
| Isolate-based density, per 1,000 patient-days | 1.46 | 1.91 | RR 1.31 (1.14–1.50) | < 0.001 |
| Patient-based density, per 1,000 patient-days | 0.91 | 1.01 | RR 1.11 (0.93–1.33) | 0.259 |
| Isolate-based proportion, % (n/N) | 15.19 (355/2,337) | 20.54 (482/2,347) | OR 1.44 (1.24–1.68) | < 0.001 |
| IPR-standardized proportion, %: numerator adjusted | 15.19 | 17.37 (14.99–20.06)* | 2.18 (− 0.68 to 5.25)‡ | 0.145 |
| IPR-standardized proportion, %: numerator and denominator adjusted | 15.19 | 17.94 (15.84–20.17)* | 2.75 (0.18 to 5.40)‡ | 0.036 |

Patient-days were 243,903 in March–August 2025 and 252,569 in March–August 2026. The IPR-standardized proportion is the March–August 2025 IPR (1.60) multiplied by the number of MDRO-positive patients in March–August 2026 (expected isolates, 408) and divided by the number of target-organism isolates in that period; in the second version the 74 excess isolates were also removed from the denominator. For patient-based density, the comparison had 80% power to detect only an RR of about 1.29 or more.

IPR, isolate-to-patient ratio; MDRO, multidrug-resistant organism; OR, odds ratio; RR, rate ratio.

*95% CI from a parametric bootstrap (20,000 resamples). †From the analysis that allows for overdispersion between months (95% CI 1.02–1.37); the bootstrap *P* value was 0.005. ‡Difference from 2025 in percentage points, with 95% CI from the same bootstrap.

# Sensitivity analyses

When the three out-of-control months were excluded, the upward monthly trend in the overall proportion was no longer statistically significant (Table 4). For CRKP the trend was not significant without seasonal adjustment (*P* = 0.062) but remained so with it (OR per year 1.31; 95% CI 1.07–1.59; *P* = 0.009).

Table 4
Monthly trend in the overall and CRKP isolate-based proportions under different analysis sets

| Analysis set | Model | Overall, OR per year (95% CI) | *P* value | CRKP, OR per year (95% CI) | *P* value |
|---|---|---|---|---|---|
| All 32 months | Unadjusted | 1.17 (1.04–1.31) | 0.008 | 1.46 (1.17–1.81) | < 0.001 |
| | Seasonally adjusted | 1.16 (1.04–1.30) | 0.010 | 1.45 (1.21–1.74) | < 0.001 |
| Excluding May–June 2026 | Unadjusted | 1.09 (0.98–1.21) | 0.105 | 1.30 (1.05–1.62) | 0.017 |
| | Seasonally adjusted | 1.10 (0.98–1.22) | 0.092 | 1.36 (1.13–1.64) | 0.001 |
| Excluding May–July 2026 | Unadjusted | 1.07 (0.95–1.19) | 0.253 | 1.25 (0.99–1.58) | 0.062 |
| | Seasonally adjusted | 1.08 (0.96–1.21) | 0.209 | 1.31 (1.07–1.59) | 0.009 |

Quasi-binomial logistic regression with time in years as the covariate. Seasonal adjustment used one sine–cosine pair with a 12-month period.

CRKP, carbapenem-resistant *Klebsiella pneumoniae*; OR, odds ratio.

# Distribution by ward

For January–August, the proportion of key MDROs rose between 2024 and 2026 in the ICU (21.30% to 41.46%), rehabilitation and other wards (20.53% to 42.07%) and surgical wards (6.23% to 11.51%) (each *P* for trend < 0.001; Additional file 1: Table S2). Outside the ICU, the CRKP proportion was 19.01% (92/484) in 2024, 22.85% (117/512) in 2025 and 33.40% (168/503) in 2026 (*P* for trend < 0.001). With May–July removed from both years, to exclude patients transferred out of the ICU during the cluster, it was 22.56% (67/297) in 2025 and 31.23% (94/301) in 2026 (*P* = 0.017). The non-ICU wards with the highest CRKP proportions over the 32 months were the burn unit (61.76%), neurosurgery (56.36%) and rehabilitation medicine (47.13%).

# CRE origin and HAI incidence

Of 520 CRE isolates with an origin classification, 252 (48.5%) were present on admission, 125 (24.0%) represented colonization, 112 (21.5%) came from HAIs and 31 (6.0%) were contaminants. There were

153 HAI episodes caused by key MDROs in 1,317,128 patient-days (0.116 per 1,000 patient-days), 81 of them (52.9%) due to CRKP. For January–August the incidence was 0.117 in 2024, 0.116 in 2025 and 0.130 in 2026 (40, 37 and 42 episodes; *P* for trend = 0.644), and the monthly RR per year was 1.06 (95% CI 0.86–1.30; *P* = 0.594). With about 40 episodes per period, only an RR of about 1.86 or more could have been detected with 80% power.

## Discussion

In 32 monthly reports rebuilt under one set of definitions, the isolate-based proportion of key MDROs was clearly higher in 2026 than in the two preceding years. Over the same months, however, the number of isolates per MDRO-positive patient rose by 18% and went out of control from May through July. Isolate-based density increased by 31% and patient-based density by 11%, and standardizing for the IPR removed about half of the between-year difference in the isolate-based proportion. The rise in CRKP persisted in seasonally adjusted sensitivity analyses and extended beyond the ICU, while HAI incidence did not change measurably.

# Isolate-based rates amplify changes in repeat isolation

An isolate-based rate taken from routine reports can therefore exaggerate an increase in resistance. This agrees with studies based on patient-level data: resistance among hospital isolates was higher when duplicates were included [9], later isolates were less susceptible than first isolates [12], fewer than half of *K. pneumoniae* isolates in a 10-year series from a large Chinese teaching hospital remained after deduplication [25], and proportions of CRKP, CRAB, CRPA and MRSA fell once duplicates were removed [15]. Those studies concern the level of resistance. The present analysis concerns its trend, and shows that the effect on a trend can be estimated from aggregate counts alone, provided the reports give the number of patients as well as the number of isolates. What matters for a trend is not how many duplicates there are but whether their number per patient has changed: in our data, a rise in the IPR from 1.60 to 1.89 was enough to turn an 11% increase in patients into a 31% increase in isolates.

Repeat isolates come mainly from severely ill patients who are cultured often; in May 2026, three ICU patients with bloodstream infection were enough to move the hospital-wide figure. That is not the same as unnecessary culturing: follow-up blood cultures in persistent bacteremia are clinically indicated, and sampling from several sites is routine in critical illness. The IPR says nothing about whether cultures were appropriate. It shows only that the isolate count and the patient count have diverged.

China's 2024 indicators replaced the isolate-based rate with HAI incidence [14], and our findings are consistent with the reasoning behind that change. Read on its own, an isolate-based rate can turn a change in culturing practice, or in a handful of critically ill patients, into an apparent change in resistance. Where such rates are still reported monthly, the IPR can be calculated from numbers already in the report.

# What an out-of-control IPR means

An IPR above its control limit has two possible explanations, which are not mutually exclusive: a few patients cultured repeatedly, or a cluster with new cases. Both were present in May through July 2026. The investigation opened under the national outbreak guideline [26] found clustering in time and place with matching antibiograms, and at the same time follow-up blood cultures from three patients raised the IPR. Without molecular typing we could not separate the two components further.

Dropping the out-of-control months therefore removes real new cases along with the repeat isolates, so the loss of statistical significance shows only that the rise depended on those months, not that it was an artifact. IPR standardization keeps the new patients but assumes the same intensity of repeat culturing in both years, which may not hold when follow-up cultures were clinically necessary. The two versions of the standardization place the share of the increase associated with the IPR at about half, and the remaining 2 to 3 percentage points are the part that repeat isolation does not explain.

We therefore treat an out-of-control IPR as a signal for review, neither noise nor proof that resistance has worsened. Two steps follow from it. First, list the patients who contributed the most isolates that month and check whether the repeats were follow-up cultures from one site or cultures from different sites. Second, check whether MDRO-positive patients and HAI episodes rose in step; if several HAIs with the same organism occurred on one ward within a short period, an outbreak investigation should begin and isolates should be saved for typing [26]. In our series the control chart and a fixed threshold of 2.0 picked out nearly the same months. One episode in one hospital cannot show how sensitive or specific either rule is, and the threshold was chosen after the data were seen; other hospitals would need to set limits from their own baseline.

# CRKP beyond the ICU

CRKP did not behave like the overall measure. The trend persisted after exclusion of May–June 2026 and, once seasonality was taken into account, after exclusion of May–July. With only 32 months the seasonal terms are unstable, so this is supportive rather than conclusive evidence; CRKP counts are themselves subject to repeat isolation, so the size of the rise may still be overstated. The increase outside the ICU, which held when May–July were removed from both years, suggests that the problem is not confined to the ICU.

Nearly half of the classified CRE isolates were present on admission, which points to patients colonized on arrival as an important source. World Health Organization and European guidance recommends active screening, contact precautions, patient placement and environmental cleaning as a combined approach [27, 28]. CRKP proportions were near or above 50% in the burn unit, neurosurgery and rehabilitation medicine. If admission screening is introduced, the ICU and these three units would be the logical places to start, and resistance status should be passed on reliably when patients are transferred. We had no screening data with which to test this.

# Isolate-based rates and HAI incidence

The isolate-based proportion and HAI incidence did not move together. Isolates include colonization, organisms present on admission and contaminants, so the proportion reflects the burden of resistant organisms among cultured inpatients more than transmission in the hospital, whereas HAI incidence counts new infections and is unaffected by repeat cultures. SHEA/HICPAC recommend incidence density for evaluating control efforts [7], and both kinds of measure should be reported. With about 40 HAI episodes per period, however, the absence of an observed increase should not be read as no change.

# Strengths and limitations

The study used every monthly report over 32 months, rebuilt under one set of definitions and checked for internal consistency. It compared the same calendar months across years, and it set the isolate-based measures beside two measures that repeat cultures cannot inflate, patient-based density and HAI incidence. The method needs only two counts per month.

The main limitation is that the IPR was not validated against patient-level deduplication. We worked from aggregate reports, so first-isolate counts as defined by CLSI M39 could not be derived, and the IPR is a proxy for the number of duplicates. Patients were counted per organism and per month, and totals for longer periods were sums of monthly counts, so a patient who stayed positive across two months was counted twice. This inflates the patient count most when long-stay patients accumulate, as they did during the cluster. The likely effect is to understate the IPR and its rise, and to overstate rather than understate the 11% increase in patient-based density. Comparing the IPR with first-isolate data extracted from the laboratory information system is the necessary next step.

Second, this was a single-center study with patient counts for only 18 months and one between-year comparison. The control limits rest on 10 baseline months, and the out-of-control signal coincided with a single cluster. The size of the effect, about half of the apparent increase, describes this hospital in this period and should not be taken as a general estimate. What can be carried over to other hospitals is the mechanism and the way of checking for it.

Third, the IPR mixes repeat culturing, multisite infection and severity of illness, and culture submissions per 1,000 patient-days were lower in 2025 and 2026 than in 2024 (Additional file 1: Table S1), so sampling intensity was not constant. The standardization is approximate, and the CI of the residual difference included zero in the primary version. Isolate-level tests ignore correlation within patients, which makes the intervals for isolate-based measures too narrow. The excluded months, the alert threshold and the model breakpoint were chosen after inspection of the data, most comparisons were not adjusted for multiplicity, and the series is short for estimating seasonality.

Finally, without molecular typing, antimicrobial consumption data or admission screening results, we cannot say whether the ICU cluster was clonal or what drove the rise in CRKP. Contaminants remained in the isolate counts, and the source reports contained documented data issues (Additional file 1: Supplementary Methods).

## Conclusions

Isolate-based MDRO rates are sensitive to changes in repeat isolation from the same patient and can exaggerate an increase in resistance; in this hospital, about half of an apparent rise reflected more isolates per patient. Routine reports should give patient-based density and HAI incidence alongside isolate-based rates. Where only aggregate counts are available, the IPR can be calculated from the numbers of isolates and of MDRO-positive patients, and a control chart of it marks the months in which both repeat culturing and clustering should be checked. Where possible, deduplicated first-isolate data should be extracted from the laboratory information system, and the IPR should be validated against them. In our hospital the rise in CRKP persisted in most sensitivity analyses and reached wards outside the ICU, and it remains the priority for MDRO control.

## Abbreviations

CI
confidence interval
CLSI
Clinical and Laboratory Standards Institute
CRAB
carbapenem-resistant *Acinetobacter baumannii*
CRE
carbapenem-resistant Enterobacterales
CRECO
carbapenem-resistant *Escherichia coli*
CRKP
carbapenem-resistant *Klebsiella pneumoniae*
CRPA
carbapenem-resistant *Pseudomonas aeruginosa*
HAI
healthcare-associated infection
ICU
intensive care unit
IPR
isolate-to-patient ratio
MDRO
multidrug-resistant organism
MRSA
methicillin-resistant *Staphylococcus aureus*
OR
odds ratio
RR

rate ratio
SHEA/HICPAC
Society for Healthcare Epidemiology of America and Healthcare Infection Control Practices Advisory Committee
VRE
vancomycin-resistant *Enterococcus*.

## Declarations

**Ethics approval and consent to participate**

The study used only aggregate routine surveillance data without patient identifiers. It was approved by the Ethics Committee of Chengdu Second People's Hospital (approval No. 202604124), which waived the requirement for informed consent. The study was conducted in accordance with the Declaration of Helsinki.

**Consent for publication**

Not applicable.

# Competing interests

The authors declare that they have no competing interests.

# Clinical trial number

Not applicable.

# Additional file 1 (.docx).

Supplementary Methods; Supplementary Results; Table S1, patient-days, culture submissions and distribution of target organisms, January–August 2024 to 2026; Table S2, isolate-based proportion of key multidrug-resistant organisms by ward group, January–August 2024 to 2026.

**Additional file 2 (.docx).**

RECORD checklist.

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# Author Contribution

Authors' contributionsSZ conceived the study, developed the methods, curated and analyzed the data, and drafted the manuscript. WZ, LX and MG collected and validated the data. LZ supervised the study and revised the manuscript critically. All authors read and approved the final manuscript.

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During the preparation of this work the authors used Claude (Anthropic) to translate the manuscript from Chinese into English and to improve its language, readability and organization. The authors reviewed and edited the output, checked all data and analyses, and take full responsibility for the content of the publication.

# Availability of data and materials

The study database (including the harmonization procedure, logical checks and log of data issues) and the analysis code are available from the corresponding author on reasonable request.

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# Figures

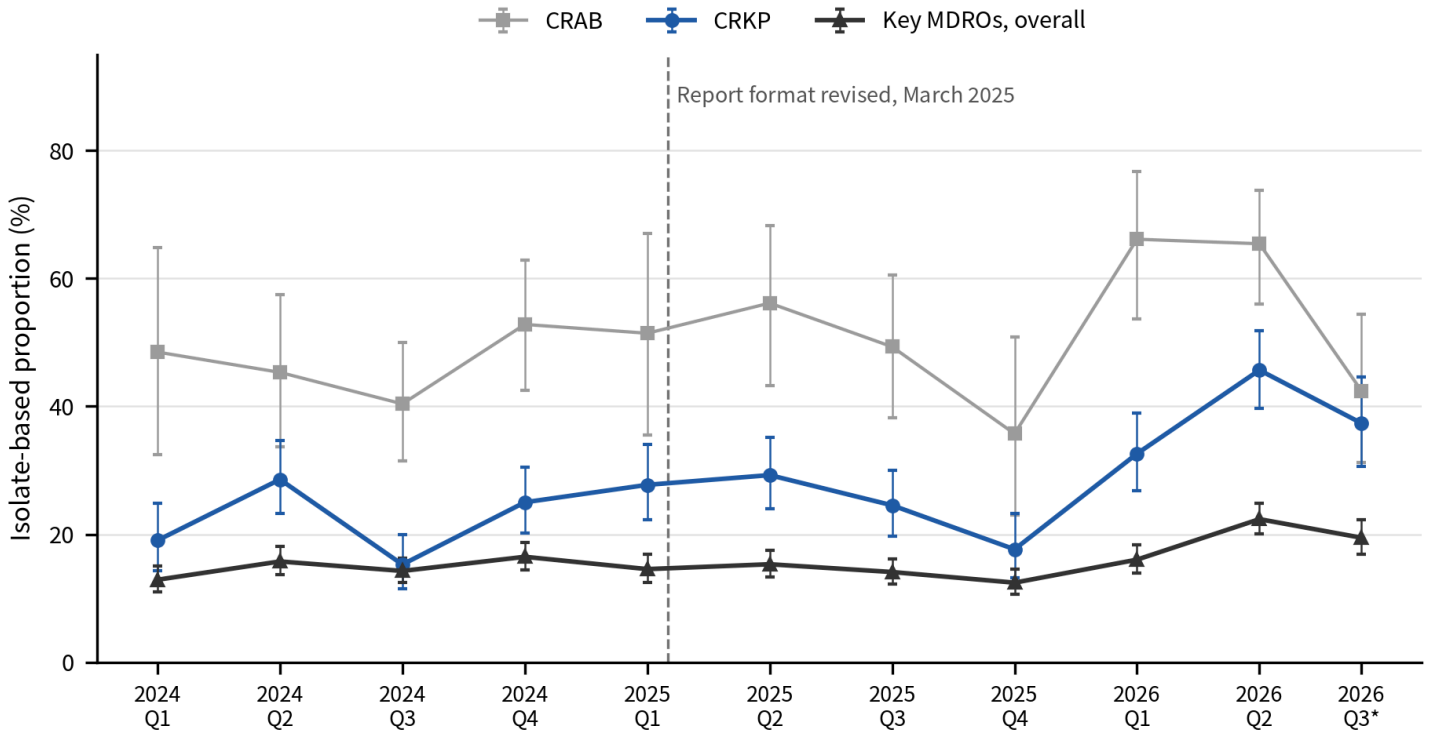


**Figure 1**

Quarterly isolate-based proportions of key multidrug-resistant organisms (MDROs) overall, carbapenem-resistant *Klebsiella pneumoniae* (CRKP) and carbapenem-resistant *Acinetobacter baumannii*(CRAB), first quarter of 2024 through third quarter of 2026. Error bars are 95% CIs (Wilson method). The dashed vertical line marks the revision of the report format in March 2025; earlier data were recalculated under the harmonized definitions, and the first quarter of 2025 includes two prerevision months. *The third quarter of 2026 includes July and August only. Proportions are isolate based, without patient-level deduplication.

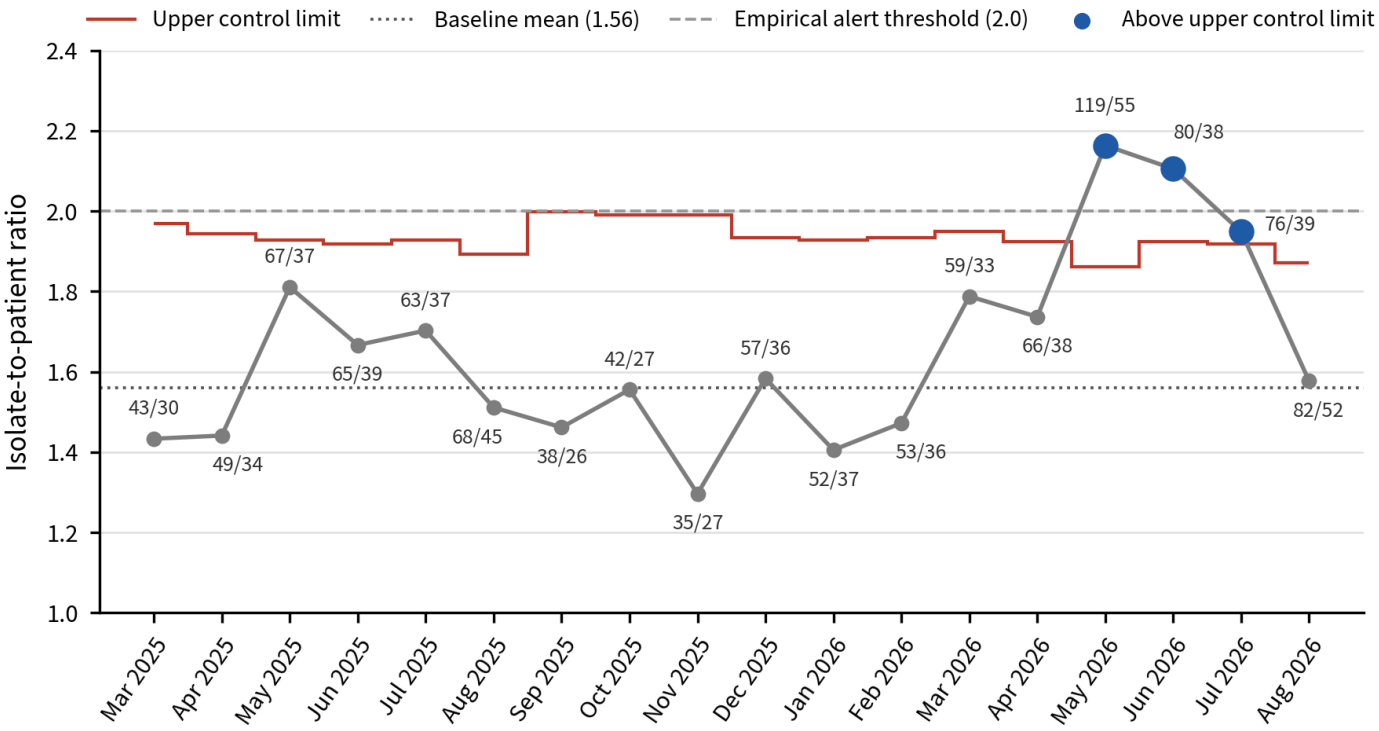



**Figure 2**

Control chart of the monthly isolate-to-patient ratio for key multidrug-resistant organisms (MDROs), March 2025 through August 2026. Data labels give key MDRO isolates/MDRO-positive patients. The dotted line is the baseline mean (March–December 2025), the stepped line is the upper control limit, which varies with the number of patients in each month, and the dashed line is the empirical alert threshold of 2.0. Large filled circles mark months above the upper control limit. Patient counts were not reported before March 2025.

# Supplementary Files

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

- Additionalfile1Supplementary.docx

- Additionalfile2RECORDchecklist.docx