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Prevalence, Determinants, and Temporal Dynamics of Multidrug-Resistant Gram-Negative Bacilli in Urinary Tract Infection Patients from Central Portugal (2018–2022)

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Abstract

Background: Multidrug-resistant (MDR) Gram-negative bacilli (GNBs) significantly compromise the effective management of urinary tract infections (UTIs) worldwide. As antimicrobial resistance varies across regions, locally tailored data are essential to guide empirical therapy. This study investigated the prevalence, determinants, and temporal dynamics of MDR GNBs in UTI patients from Central Portugal between 2018 and 2022. Methods: We conducted a retrospective observational study at a hospital center in Central Portugal, analyzing data from 2018 to 2022. Data from 5194 UTI patients with GNB-positive cultures were analyzed. Binary logistic regression was used to identify determinants of MDR GNBs, defined as resistance to ≥1 agent in ≥3 antibiotic classes. Results: The study population had a mean age of 64.5 ± 25.3 years, and females represented two-thirds of the sample (67.0%). The overall prevalence of MDR GNBs was 35.8%. Advanced age (≥75 years), male sex, and specific treatment contexts—particularly day treatment and laboratory-only cases—were independently associated with MDR. SBL-producing Enterobacterales and non-fermenting GNBs showed the highest risk levels. Conclusions: MDR GNBs are highly prevalent among UTI patients in Central Portugal, and their increasing trend—particularly in 2022—highlights an urgent need for strengthened surveillance and updated empirical treatment strategies. The observed temporal increase highlights the urgent need for strengthened regional surveillance and updated empirical treatment guidelines.

Keywords: Anti-Bacterial Agents; drug resistance; Gram-negative bacteria; Portugal; urinary tract infections

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1. Introduction

Urinary tract infections (UTIs) are among the most common bacterial infections worldwide, affecting individuals across all age groups and occurring in both community and healthcare settings. Most cases are successfully treated with standard antibiotics in primary care [1–3]. However, the growing emergence of antimicrobial resistance has significantly challenged the effectiveness of empirical therapies traditionally used for these infections.

Among UTI pathogens, Gram-negative bacilli (GNBs) such as *Escherichia coli*, *Klebsiella pneumoniae*, and *Pseudomonas aeruginosa* are of particular concern due to their increasing resistance to commonly used antimicrobials [4]. The rise in multidrug-resistant

(MDR) GNBs—defined as resistance to at least one agent in three or more antibiotic classes—represents a critical public health threat, leading to higher morbidity, mortality, prolonged hospital stays, and elevated healthcare costs in UTI patients [5,6].

Identifying the determinants and drivers of MDR GNBs is therefore essential for developing targeted prevention and risk-stratification strategies. These determinants can be broadly categorized into patient-related and exposure-related factors. Evidence shows that advanced age, male gender [7], previous or frequent antibiotic use [7,8], hospitalization [8], production of extended-spectrum beta-lactamases (ESBL), chronic steroid therapy [9], multiple comorbidities, recent healthcare contact, and admission to critical care units [10] all increase the likelihood of MDR infection.

Importantly, antimicrobial resistance (AMR) varies markedly between regions and even between hospitals within the same country. This variability reflects differences in antibiotic prescribing practices, infection control policies, and the circulation of specific resistant clones. Resistance profiles vary substantially between regions and even between hospitals within the same country, influenced by local prescribing practices, infection control policies, and the circulation of specific resistant clones [11]. Consequently, global or national guidelines often fail to capture local resistance trends, rendering them suboptimal for guiding empirical therapy at the regional level [12].

In Portugal, surveillance data on MDR urinary pathogens remain limited, particularly at the regional scale. Understanding the temporal dynamics and determinants of MDR GNBs in specific geographic contexts is vital for updating empirical treatment protocols and supporting national antibiotic stewardship initiatives.

However, regional-level AMR data in Portugal remain scarce, limiting the ability to design empirical therapy tailored to local resistance patterns. Therefore, this study aimed to determine the prevalence and determinants of MDR GNBs in UTI patients, and to analyze temporal trends in Central Portugal between 2018 and 2022.

2. Materials and Methods

2.1. Ethical Consideration

The study approval (letter no. CE-UBI-Pj-2023-020 dated 19 April 2023) was sought from the Ethics Committee and Data Protection Officer of the University of Beira Interior. Informed consent requirement was waived off by the committee.

2.2. Study Design, Setting and Duration

The retrospective observational study was carried out at a hospital center in central Portugal from January 2018 to December 2022.

2.3. Sample Size

All available patients' records of 5857 UTI patients, diagnosed positive on urine culture test, spanning over a five-years period from January 2018 to December 2022 were retrieved from the Urinary Tract Infections in Central Portugal (ITUCIP) project. Patients demonstrating growth of any Gram-negative bacilli (n = 5194) such as *Escherichia coli*, *Klebsiella pneumoniae*, *Morganella morganii*, *Proteus mirabilis*, and *Pseudomonas aeruginosa*, etc., were included. Other inclusion criteria were patients of any age, sex, and patient care setting. Patients demonstrating growth of Gram-positive cocci (GPCs, n = 661) such as *Enterococcus faecalis*, *Enterococcus faecium*, and *Staphylococcus aureus*, etc., and patients with missing data (n = 2) were excluded. Details are provided in Figure 1.

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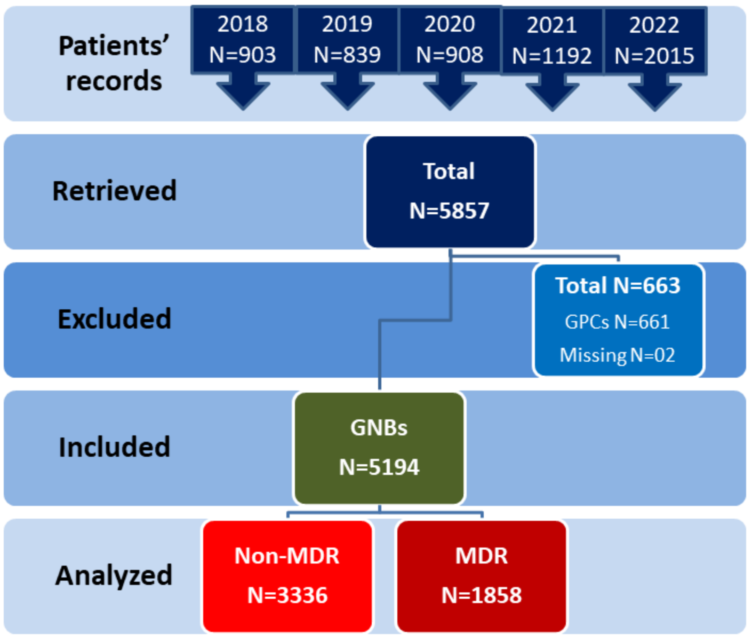

Figure 1. Flow diagram showing sample selection process.

2.4. Antimicrobial Susceptibility Testing (AST)

AST was performed using disk diffusion and automated methods (VITEK 2, bioMérieux, Lyon, France). Results were interpreted according to EUCAST 2022 (version 12.0) standards [13].

2.5. Independent Variables

The dataset included patient demographics, care setting, clinical specialty, bacterial isolate, antimicrobial susceptibility results, and the year of the culture test.

The retrieved dataset included patient age, sex, pregnancy status, patient care setting, clinical department/specialty, bacterial isolate, antimicrobial susceptibility test results, and the year of urine culture.

Age was categorized into six groups: <5 years, 5–18 years, 19–35 years, 36–55 years, 56–75 years, and >75 years. Sex and pregnancy status were combined into a three-level interaction variable: (1) non-pregnant female, (2) pregnant female, and (3) male.

Patient care settings were classified as (1) outpatient/community, (2) emergency department, (3) inpatient department, (4) day treatment, and (5) laboratory-only cases. Clinical departments/specialties were grouped as (1) low-risk/community, (2) general/internal medicine, (3) surgery/post-surgery, (4) high-risk/ICU/transplant, (5) urology/nephrology, and (6) other specialties.

Bacterial isolates (Gram-negative bacilli) were categorized into six major groups: (1) *Escherichia coli* (non-ESBL), (2) *Klebsiella*/*Proteus* spp. (non-ESBL), (3) *Enterobacterales* (ESBL-producing), (4) *Enterobacterales* (high-risk/AmpC-inducible), (5) non-fermenting GNBs, and (6) other or unspecified GNBs.

The following nine antibiotic classes were included in the MDR definition:

(1) Fluoroquinolones,
(2) Aminoglycosides,
(3) Carbapenems,
(4) Cephalosporins,
(5) Penicillins,
(6) β-lactam/β-lactamase-inhibitor combinations,
(7) Fosfomycin,

(8) Polymyxins,
(9) Folate-pathway inhibitors.

2.6. Dependent Variable

GNB isolates resistant to at least one antimicrobial agent in three or more distinct antibiotic classes were defined as multidrug-resistant (MDR). Isolates resistant to two or fewer classes, or susceptible to all nine, were considered non-MDR.

2.7. Statistical Analysis

All statistical analyses were performed using IBM SPSS Statistics version 26 (IBM Corp., Armonk, NY, USA). Categorical variables were summarized as frequencies and percentages. Binary logistic regression was applied to identify determinants of MDR GNBs, with MDR status (yes/no) as the binary outcome variable.

We first conducted univariate logistic regression for each covariate, followed by a multivariate model including all predictors. An additional model assessed the temporal trend using year as a continuous variable. The first category within each variable served as the reference group.

To evaluate the overall temporal trend in MDR occurrence during 2018–2022, an additional logistic regression was performed using "year of culture test" as a continuous variable. The model coefficient was used to estimate the Average Annual Percent Change (AAPC) in MDR odds.

Because ESBL phenotype is inherently linked to cephalosporin resistance and MDR classification includes resistance across multiple classes, some degree of overlap is expected. Therefore, ESBL status was included as a microbiological variable of interest, but its adjusted associations with MDR must be interpreted cautiously and not as causal.

Graphical representation of antimicrobial resistance profiles (100% stacked column charts) was created using Microsoft Excel. A two-sided p-value ≤ 0.05 was considered statistically significant.

This work is part of an ITUCIP study (Urinary Tract Infections in the Central Interior of Portugal).

3. Results

Data from 5194 UTI patients with GNB infection were subjected to final analysis. The mean age of the population was 64.5 $\pm$ 25.3 years (range 1 month–105 years). Females represented 67% of the sample. When patients were categorized into six age groups, the elderly had the highest proportion 2316 (44.6%), followed by older adults 1446 (27.8%). Female proportion was twice that of males (67.0% vs. 33.0%). Among females, 111 (2.1%) were pregnant. Among patient care settings, the highest proportion of patients 2173 (41.8%) visited the emergency department, followed by the inpatient department with 1438 (27.7%). Among specialties, nearly half of the patients 2508 (48.3%) sought medical treatment from the medicine department. *E. coli* (non-ESBL) was the most isolated GNB, detected in 3035 (58.4%) patients. The highest proportion of GNB UTIs, 1730 (33.3%), was noted in 2022.

The overall frequency of MDR GNBs was 1858 (35.8%). MDR prevalence increased progressively with age. Elderly patients (>75 years) showed the highest proportion of MDR isolates. Non-pregnant females had a higher MDR-positive rate as compared to males (53.5% vs. 45.4%). The emergency setting (729, 39.2%), the inpatient setting (637, 34.3%), and the medicine department (952, 51.2%) showed high MDR rates. Non-ESBL *E. coli* (525, 28.3%) and *Klebsiella*/*Proteus* species (522, 28.1%) showed similarly higher MDR rates. The highest MDR frequency 654 (35.2%) was noted in 2022, as shown in Table 1.

Table 1. Demographic and clinical characteristics of study population stratified by multi-drug resistance status.

		Multi-Drug Resistance				Total (N = 5194)	
		No (N = 3336)		Yes (N = 1858)			
Age (years)	<5 (Preschool children)	189	5.7%	39	2.1%	228	4.4%
	5–18 (Children and adolescents)	162	4.9%	48	2.6%	210	4.0%
	19–35 (Young adults)	292	8.8%	65	3.5%	357	6.9%
	36–55 (Middle-aged adults)	460	13.8%	177	9.5%	637	12.3%
	56–75 (Older adults)	923	27.7%	523	28.1%	1446	27.8%
	>75 (Elderly)	1310	39.3%	1006	54.1%	2316	44.6%
Sex	Non-pregnant female	2373	71.1%	994	53.5%	3367	64.8%
	Pregnant female	91	2.7%	20	1.1%	111	2.1%
	Male	872	26.1%	844	45.4%	1716	33.0%
Patient care setting	Outpatient/Community	841	25.2%	343	18.5%	1184	22.8%
	Emergency department	1444	43.3%	729	39.2%	2173	41.8%
	Inpatient department	801	24.0%	637	34.3%	1438	27.7%
	Day treatment	55	1.6%	58	3.1%	113	2.2%
	Laboratory only	195	5.8%	91	4.9%	286	5.5%
Department	Low risk/Community	398	11.9%	139	7.5%	537	10.3%
	General/Internal medicine	1556	46.6%	952	51.2%	2508	48.3%
	Surgery/Post-surgery	202	6.1%	141	7.6%	343	6.6%
	High risk/ICU/Transplant	112	3.4%	87	4.7%	199	3.8%
	Urology/Nephrology	276	8.3%	262	14.1%	538	10.4%
	Other specialties	792	23.7%	277	14.9%	1069	20.6%
Bacterial isolate	*E. coli* (non-ESBL)	2510	75.2%	525	28.3%	3035	58.4%
	Klebsiella/Proteus (non-ESBL)	654	19.6%	522	28.1%	1176	22.6%
	Enterobacterales (ESBL)	23	0.7%	420	22.6%	443	8.5%
	Enterobacterales (high risk/AmpC-inducible)	106	3.2%	173	9.3%	279	5.4%
	Non-fermenting GNBs	27	0.8%	208	11.2%	235	4.5%
	Other/Unspecified	16	0.5%	10	0.5%	26	0.5%
Year of culture test	2018	528	15.8%	292	15.7%	820	15.8%
	2019	505	15.1%	263	14.2%	768	14.8%
	2020	534	16.0%	300	16.1%	834	16.1%
	2021	693	20.8%	349	18.8%	1042	20.1%
	2022	1076	32.3%	654	35.2%	1730	33.3%

ICU: Intensive care unit; ESBL: extended-spectrum beta-lactamase; GNBs: Gram-negative bacilli.

ESBL Enterobacterales demonstrated absolute resistance to penicillins (100.0%), near-total resistance to cephalosporins (99.8%), and high resistance to fluoroquinolones (89.4%) and folates (71.1%). However, ESBL Enterobacterales showed low resistance to carbapenems (3.8%) and absolute susceptibility to polymyxins (100.0%). High-risk Enterobacterales also demonstrated near-total resistance to penicillins (99.6%) but had their lowest resistance to folates (12.2%). Non-fermenting GNBs demonstrated extremely high resistance to folates (97.1%), penicillins (94.5%), cephalosporins (88.0%), carbapenems (85.1%), and fosfomycins (84.6%). Conversely, non-fermenting GNBs showed their lowest resistance to polymyxins (4.9%) and absolute susceptibility to fosfomycin (100.0%), as shown in Figure 2.

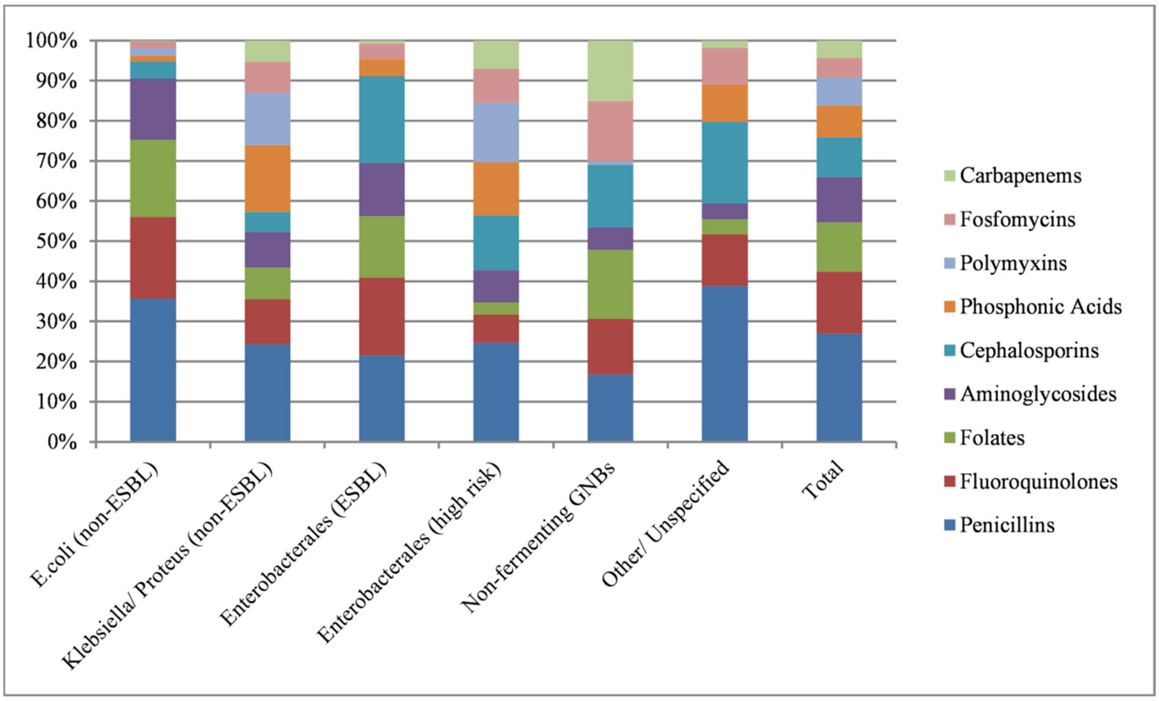

Figure 2. Antimicrobial resistance profiles of different GNBs in UTI patients.

In univariate analyses, middle-aged adults had a 2-fold higher risk of MDR GNBs (OR = 1.865; $p = 0.002$), older adults had a 2.7-fold higher risk (OR = 2.746; $p < 0.001$), and the elderly showed the highest risk at 3.7-fold (OR = 3.722; $p < 0.001$). Males carried a 2.3-fold greater risk (OR = 2.311; $p < 0.001$), while pregnant females demonstrated a 48% protective effect (OR = 0.525; $p = 0.010$). This risk of MDR GNBs varied significantly by setting, with the emergency department (OR = 1.238), inpatient department (OR = 1.950), and day treatment (OR = 2.586). Likewise, specialty departments such as urology (OR = 2.718), ICU (OR = 2.224), surgery (OR = 1.999), and medicine (OR = 1.752) showed elevated risk. The strongest risk factors were the causative organisms themselves, with non-fermenting GNBs conferring a 36.8-fold higher risk (OR = 36.831; $p < 0.001$) and ESBL-producing Enterobacterales conferring an overwhelming 87.3-fold higher risk (OR = 87.304; $p < 0.001$). Year of culture test (as categoric variable) did not show any association with the risk of MDR GNBs (all $p > 0.05$). Time (year as continuous variable) showed a trend for an annual increase in MDR GNB odds by 2.4% (OR = 1.024); however, this relationship was not statistically significant ($p = 0.229$), as shown in Table 2.

In multivariate analysis, the elderly had a two-fold higher risk of MDR GNBs (OR = 2.062; $p = 0.005$). Males carried a 42% greater risk (OR = 1.424; $p < 0.001$). By setting, Day treatment was associated with an 86% increased risk (OR = 1.868; $p = 0.011$), and laboratory-only cases showed a 94% greater risk (OR = 1.941; $p = 0.003$). Among causative organisms, ESBL-producing *Enterobacterales* conferred the highest risk of MDR GNBs (OR = 92.318; $p < 0.001$), followed by non-fermenting GNBs (OR = 29.065; $p < 0.001$). Non-ESBL *Klebsiella/Proteus* and high-risk *Enterobacterales* were also linked to a 3- to 6-fold

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greater risk. Finally, the Year 2022 was associated with a 55% higher risk of MDR GNBs (OR = 1.559; $p < 0.001$), as shown in Table 3.

Table 2. Determinants of MDR GNBs in UTI patients by univariate analyses.

Variable(s)	Covariate(s)	OR	95% C.I. for OR		
			Lower	Upper	*p*
Age (years)	<5 (Preschool children)	Reference category			
	5–18 (Children and adolescents)	1.436	0.896	2.301	0.133
	19–35 (Young adults)	1.079	0.697	1.670	0.734
	36–55 (Middle-aged adults)	1.865	1.268	2.743	0.002
	56–75 (Older adults)	2.746	1.914	3.940	<0.001
	>75 (Elderly)	3.722	2.611	5.304	<0.001
Sex	Non-pregnant female	Reference category			
	Pregnant female	0.525	0.322	0.856	0.010
	Male	2.311	2.049	2.606	<0.001
Patient care setting	Outpatient/Community	Reference category			
	Emergency department	1.238	1.061	1.444	0.007
	Inpatient department	1.950	1.656	2.295	<0.001
	Day treatment	2.586	1.751	3.818	<0.001
	Laboratory only	1.144	0.866	1.512	0.343
Department	Low risk/Community	Reference category			
	General/Internal medicine	1.752	1.421	2.160	<0.001
	Surgery/Post-surgery	1.999	1.497	2.669	<0.001
	High risk/ICU/Transplant	2.224	1.583	3.126	<0.001
	Urology/Nephrology	2.718	2.103	3.513	<0.001
	Other specialties	1.001	0.790	1.269	0.991
Bacterial isolate	*E. coli* (non-ESBL)	Reference category			
	Klebsiella/Proteus (non-ESBL)	3.816	3.289	4.427	<0.001
	Enterobacterales (ESBL)	87.304	56.785	134.227	<0.001
	Enterobacterales (high risk/AmpC-inducible)	7.803	6.020	10.114	<0.001
	Non-fermenting GNBs	36.831	24.399	55.599	<0.001
	Other/Unspecified	2.988	1.348	6.621	0.007
Year of culture test (categoric)	2018	Reference category			
	2019	0.942	0.766	1.158	0.569
	2020	1.016	0.831	1.242	0.878
	2021	0.911	0.751	1.104	0.340
	2022	1.099	0.925	1.306	0.284
Year of culture test (continuous)	Time (year)	1.024	0.985	1.065	0.229

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Table 3. Determinants of MDR GNBs in UTI patients by multivariate analyses.

Variable(s)	Covariate(s)	AOR	95% C.I. for AOR		*p*
			Lower	Upper	
Age (years)	<5 (Preschool children)	Reference category			
	5–18 (Children and adolescents)	1.395	0.806	2.415	0.234
	19–35 (Young adults)	1.116	0.620	2.009	0.714
	36–55 (Middle-aged adults)	1.474	0.867	2.505	0.152
	56–75 (Older adults)	1.625	0.980	2.693	0.060
	>75 (Elderly)	2.062	1.249	3.404	0.005
Sex	Non-pregnant female	Reference category			
	Pregnant female	1.241	0.649	2.371	0.514
	Male	1.424	1.224	1.657	<0.001
Patient care setting	Outpatient/Community	Reference category			
	Emergency department	1.228	0.917	1.645	0.169
	Inpatient department	1.202	0.920	1.571	0.177
	Day treatment	1.868	1.154	3.025	0.011
	Laboratory only	1.941	1.262	2.987	0.003
Department	Low risk/Community	Reference category			
	General/Internal medicine	0.919	0.649	1.302	0.636
	Surgery/Post-surgery	1.028	0.682	1.549	0.896
	High risk/ICU/Transplant	1.230	0.778	1.944	0.376
	Urology/Nephrology	1.290	0.938	1.774	0.117
	Other specialties	0.791	0.541	1.158	0.229
Bacterial isolate	*E. coli* (non-ESBL)	Reference category			
	Klebsiella/Proteus (non-ESBL)	3.396	2.906	3.968	<0.001
	Enterobacterales (ESBL)	92.318	59.625	142.939	<0.001
	Enterobacterales (high risk/AmpC-inducible)	6.569	5.013	8.607	<0.001
	Non-fermenting GNBs	29.065	19.061	44.319	<0.001
	Other/Unspecified	2.481	1.101	5.595	0.028
Year of culture test	2018	Reference category			
	2019	0.888	0.681	1.157	0.378
	2020	0.954	0.738	1.235	0.722
	2021	0.837	0.654	1.070	0.156
	2022	1.559	1.255	1.938	<0.001

Variable(s) entered in step 1: age (years), sex, patient care setting, department, bacterial isolate, year of culture test. ICU: intensive care unit; ESBL: extended-spectrum beta-lactamase; GNBs: Gram-negative bacilli.

Table 4 summarizes the distribution of MDR GNBs between outpatient and inpatient settings. As expected, MDR prevalence was higher among inpatients; however, a substantial proportion of MDR isolates was also observed in community-based cases, underscoring the growing public health relevance of community-associated MDR organisms.

Table 4. Distribution of MDR GNBs between outpatient and inpatient.

		MDR No	MDR Yes	Total	OR (95% CI)	*p*-Value	aOR (95% CI)	*p*-Value
Patient care setting (N = 2622)	OPD	841	343	1184	Ref.	<0.001	Ref.	<0.001
		51.2%	35.0%	45.2%				
	IPD	801	637	1438	1.950 (1.656–2.295)		1.950 (1.656–2.295)	
		48.8%	65.0%	54.8%				

4. Discussion

Multidrug-resistant (MDR) Gram-negative bacilli (GNBs) continue to challenge the management of urinary tract infections (UTIs) worldwide. Our findings demonstrate that MDR GNBs remain highly prevalent and are strongly associated with specific pathogens—particularly ESBL-producing Enterobacterales and non-fermenting GNBs—as well as patient age and care setting. The growing prevalence of antimicrobial resistance has transformed UTIs from largely manageable infections into a major therapeutic and public health concern. As antimicrobial resistance is shaped by local antibiotic use and healthcare practices, region-specific data are indispensable for optimizing empirical treatment and informing antibiotic stewardship strategies [11].

This retrospective study provides a comprehensive five-year analysis of MDR GNBs among UTI patients in Central Portugal, revealing a substantial prevalence (35.8%) and identifying key determinants of resistance. The findings underscore a critical and escalating challenge to the empirical management of UTIs in this region, consistent with the broader European concern regarding the increasing resistance among Enterobacterales and non-fermenting GNBs.

The 'laboratory-only' category included cases referred exclusively for microbiological testing, often representing repeated or follow-up samples from patients under outpatient monitoring. This subgroup therefore comprises individuals with more complex or persistent infections, which may explain the higher MDR prevalence observed in this setting.

Recent Portuguese studies have also documented a sustained increase in UTI prevalence and pathogen diversity across several hospitals in Central Portugal, highlighting an upward trajectory in UTI incidence and complexity. However, most of these studies primarily addressed overall prevalence and demographic trends rather than antimicrobial resistance profiles, underscoring the added value of the present investigation focused on MDR Gram-negative bacilli [14–17].

Comparable studies across different geographical contexts show wide variability in MDR prevalence, reflecting differences in antibiotic prescribing, infection control, and diagnostic capacity. For instance, reported MDR rates were 50.9% in East Africa [18], 46.2% in Saudi Arabia [19], and 44.0% in Northwest Ethiopia [20], all higher than the rate observed in Central Portugal. The comparatively lower prevalence in the present study may reflect regional differences in infection control policies, outpatient antibiotic consumption, or surveillance methodologies. Nevertheless, the predominance of *E. coli* as the leading uropathogen aligns with findings from diverse settings, confirming its persistent role as the principal etiologic agent of UTIs [18,20].

Resistance patterns observed here also mirror global trends, though with some distinctive features. Borcan et al. reported that GNBs exhibited >50% resistance to penicillins and >40% to third/fourth-generation cephalosporins, whereas *E. coli* maintained low resistance to fosfomycin (3%) and nitrofurantoin (2%) [21]. Similarly, Kasew et al. documented resistance exceeding 75% for several first-line agents, including ampicillin

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and trimethoprim-sulfamethoxazole [19]. In contrast, our data indicate near-universal resistance of ESBL-producing *Enterobacterales* to penicillins (100%) and cephalosporins (99.8%), and high resistance to fluoroquinolones (89.4%), yet low resistance to carbapenems (3.8%) and complete susceptibility to polymyxins (100%). These results suggest that while beta-lactamase-mediated resistance remains the dominant mechanism, carbapenems and polymyxins retain critical therapeutic potential against MDR GNBs in this region.

Although pivmecillinam is not routinely used in Portugal, it is recommended as a first-line option for uncomplicated UTIs in several Northern European countries and international guidelines. Importantly, it retains activity against many ESBL-producing Enterobacterales, making it a valuable oral alternative in settings where it is available. Its absence in our dataset reflects regional prescribing practices rather than lack of therapeutic relevance.

Non-fermenting GNBs, such as *Pseudomonas aeruginosa* and *Acinetobacter* spp., also demonstrated extensive resistance profiles, particularly to folate pathway inhibitors (97.1%), penicillins (94.5%), and cephalosporins (88.0%). These findings are consistent with reports describing non-fermenters as reservoirs of multidrug resistance genes and biofilm formation capacity, which enhance their persistence in hospital environments and resistance to disinfection protocols. The observed susceptibility to fosfomycin and low resistance to polymyxins indicate that these agents remain among the few viable options for treating infections caused by these high-risk pathogens.

Regarding risk factors, the present study corroborates the existing literature identifying male gender, advanced age, and specific healthcare exposures as major predictors of MDR infection [19,22]. Older adults and elderly individuals are particularly vulnerable due to age-related immunosenescence, higher comorbidity burden, and frequent healthcare contact. The higher odds among males likely reflect anatomical and functional urinary tract factors, as well as differential antibiotic exposure patterns. Furthermore, treatment setting emerged as an independent determinant: day treatment and laboratory-only cases showed significantly increased MDR risk, likely reflecting patients with recurrent infections or those under continuous medical surveillance.

The high adjusted ORs for ESBL Enterobacterales and non-fermenters reflect, in part, the shared resistance mechanisms underlying MDR. As ESBL positivity is inherently linked to cephalosporin resistance, and MDR classification includes resistance across multiple antimicrobial classes, the association must be interpreted as overlapping phenotypes rather than a causal effect.

Consistent with other reports [19,22], the type of bacterial isolate was the strongest determinant of MDR. ESBL-producing *Enterobacterales* and non-fermenting GNBs showed the highest adjusted odds, emphasizing that intrinsic resistance mechanisms—particularly beta-lactamase production, efflux pumps, and porin loss—remain central to the MDR phenotype. These findings reinforce the need for molecular surveillance to monitor the dissemination of high-risk clones and resistance genes, including CTX-M, OXA, and NDM families, which are increasingly reported across Europe.

The temporal increase in MDR observed in 2022 is particularly concerning and may reflect pandemic-related effects on antibiotic prescribing practices, diagnostic delays, or infection control measures. Similar post-pandemic increases in MDR rates have been documented elsewhere in Europe, suggesting that disruptions in routine stewardship and the expanded use of broad-spectrum antibiotics during COVID-19 may have contributed to the selection pressure for resistant organisms [23–25]. Several studies reported that COVID-19-related increases in broad-spectrum antibiotic prescribing reduced outpatient care continuity, and disruption of stewardship programs contributed to temporary rises in antimicrobial resistance [26–28].

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When considered alongside recent Portuguese epidemiological analyses of UTI trends, these findings provide a more comprehensive picture of the evolving infectious landscape in Central Portugal. By extending the evidence base from prevalence to antimicrobial resistance dynamics, the present study offers critical insights needed to guide local empirical therapy and strengthen regional antibiotic stewardship efforts [14–17].

Despite its strengths, including a large, regionally representative sample and a five-year analytical timeframe, this study has some limitations. Its retrospective design limits the ability to establish causal inferences. Additionally, clinical variables such as prior antibiotic use, hospitalization duration, and comorbidity index were not available, which could provide further explanatory power. The dataset, however, offers a rare regional snapshot of resistance dynamics that can guide future prospective and interventional research. This retrospective design limits causal inference and lacks clinical details such as comorbidities, prior antibiotic use, hospitalization duration, and molecular typing of isolates.

From a clinical perspective, these findings have direct implications for empirical therapy. The high prevalence of MDR GNBs calls for urgent revision of empirical antibiotic protocols in Central Portugal, with greater reliance on local susceptibility data and risk-based treatment algorithms. On a broader scale, the results support the reinforcement of antibiotic stewardship programs, continuous microbiological surveillance, and educational initiatives targeting prescribers and the public.

In summary, this study provides robust evidence that MDR GNBs are a persistent and evolving threat to effective UTI management in Central Portugal. The clear association with specific pathogens and patient characteristics underscores the necessity for tailored empirical therapy, regional resistance monitoring, and strategic interventions to preserve the efficacy of last-line antibiotics.

5. Conclusions

MDR Gram-negative bacilli are highly prevalent among UTI patients in Central Portugal. ESBL-producing Enterobacterales and non-fermenting GNBs are the strongest predictors of multidrug resistance. The increase observed in 2022—potentially influenced by pandemic-related disruptions—highlights the urgent need for strengthened regional surveillance, updated empirical treatment guidelines, and robust antibiotic stewardship initiatives.

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Informed Consent Statement: Patient consent was waived due to it being a retrospective study, with data consultation on a hospital basis, without access to patient identification.

Data Availability Statement: The data forms part of a database that can be made available for consultation, based on plausible justifications. All data is anonymized.

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