

Progress on the Global Research Agenda for Antimicrobial Resistance in Human Health in Pakistan: Findings and Implications

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Background and Objective: Antimicrobial resistance (AMR) poses a formidable challenge to global public health, with low- and middle-income countries (LMICs) including Pakistan being particularly vulnerable. This study assesses the progress made in Pakistan following the Global Research Agenda for AMR, which builds on the key activities and goals of its national action plan to reduce AMR. The intention is to identify key gaps, achievements, and future areas of focus to help reduce rising AMR rates in Pakistan.

Methods: Utilizing a systematic-narrative hybrid literature review methodology approach, recent research publication and policy initiatives related to AMR, including those published on the internet, were examined and documented.

Findings: The findings from 349 published studies were divided into the 40 research priority areas. This included 23 papers (9.95%) specifically related to prevention and 55 (22.9%) to diagnosis, 64 (26.7%) for treatment and care of patients with infectious diseases, 59 (24.5%) for cross-cutting, and 44 (18.33%) for drug-resistant tuberculosis (TB). Currently, research on AMR in Pakistan is primarily concentrated in major urban centers across a limited number of cities. This needs addressing going forward. To effectively combat AMR in Pakistan, prioritizing prevention is crucial to curb disease spread and reduce reliance on prophylactic treatments, especially inappropriate prescribing and dispensing of antimicrobials. Enhancing diagnostic facilities, strengthening antimicrobial surveillance systems and promoting appropriate management of patients with infectious diseases, supported by robust antimicrobial stewardship programs, can also help enhance judicious antibiotic use in Pakistan and reduce AMR going forward.

Conclusion and Interpretation: There are ongoing concerns regarding current research activities in Pakistan to reduce AMR. The pathway forward in Pakistan includes leveraging global partnerships to share knowledge, resources, and strategies to enhance the use of Access antibiotics as well as reduce AMR to reach agreed United Nations' goals.

Plain Language Summary: Antimicrobial resistance (AMR) poses a substantial threat to public health, as it reduces the number of effective antibiotics to combat infections, as well as increasing costs and the number of deaths. Pakistan is a critical country, with high and growing rates of AMR. Consequently, this issue must be addressed. The Global Research Agenda for AMR provided guidance on approaches that key stakeholders in Pakistan should undertake to reduce AMR. Our findings uncovered 20 published studies covering multiple aspects to reduce AMR. These included studies related to ways to reduce infections and the associated use of antibiotics and

improve the care of patients with infectious diseases, including tuberculosis (TB), resistant to the current antimicrobials. Going forward, health authorities and others in Pakistan need to prioritize activities to reduce infections, including better hygiene, as well as reduce unnecessary prescribing and dispensing of antibiotics. This includes activities called antimicrobial stewardship programs to improve antibiotic use. The pathway forward also included leveraging global partnerships to share knowledge, resources, activities and strategies to improve the future use of antibiotics. We believe that this is the first study among low- and middle-income countries to fully explore current activities as part of the Global Research Agenda for AMR to provide guidance to other researchers and countries operating in this field.

Keywords: antibiotics, antimicrobial resistance, antimicrobial surveillance, AWaRe, global research, health policy, Pakistan

Introduction

Antimicrobial resistance (AMR) is one of the greatest threats to global public health^{1–5} and occurs when pathogens become resistant to prescribed antimicrobials. As a result, making infections harder and more expensive to treat.⁶ AMR has a direct influence on morbidity and mortality, the length of hospital stays, the safety of patients including with other medical procedures, the overall costs of treatment and the global economy.^{2,4,7–9} Risk factors for emergence and transmission of AMR include challenges within the healthcare system, the water sanitation and hygiene (WASH) infrastructure, patient-related underlying health conditions and the inappropriate use of antibiotics especially those with appreciable resistance potential.^{6,10–12} In 2019, there were 1.27 million deaths directly attributable to AMR and 4.5 million deaths were associated with AMR, with the highest mortality observed in sub-Saharan and South Asia.² There are also concerns with increasing anti-fungal resistance especially in low- and middle-income countries (LMICs) and the implications going forward.^{13–15}

To help address rising rates of AMR and the resultant implications, the World Health Organization (WHO) launched the Global Action Plan (GAP) for AMR in 2015.¹⁶ The principal suggested activities to enhance appropriate antibiotic use included training and education to improve awareness of concerns regarding AMR, increasing surveillance and research, improvements in infection prevention and sanitation improvement, as well as developing the economic case for the development for sustainable investment.¹⁶ The GAP led to the development of National Action Plans (NAP) aligned with the objectives of GAP, which included Pakistan.^{17,18} However, there have been challenges with their implementation including resource issues of both personnel and available funding, which has resulted in variable implementation of the NAPs especially among LMICs.^{19–22} Various factors, including the impact of COVID-19, has hindered the progress of Pakistan's NAP in AMR, necessitating immediate rejuvenation of efforts to combat this silent pandemic.^{17,18,23}

In addition, the WHO launched AWaRe (Access, Watch, Reserve) for antibiotics to help limit the use of antibiotics, especially those that are prone to developing resistance. In this system, the Access group of antibiotics includes those with a lower potential of AMR development, the Watch group includes those with greater potential to develop resistance, with the Reserve group the last resort antibiotics, which should be reserved for life-threatening conditions given the rise in multi drug resistant (MDR) cases.^{24–26} The previous target for antibiotic use was at least 60% of antibiotics used in any given sector should be Access antibiotics.^{12,24,25} Guidance to reach this target was enhanced by the launch of the WHO AWaRe book giving suggested treatments for 35 common infectious diseases across sectors.^{24,27} More recently, the United Nations General Assembly (UN GA) on AMR set an updated target of 70% antibiotic use in sectors being from the Access list.^{5,28}

Other WHO initiatives include crafting a Microbiology Laboratories Database, a Microbiologists Database, and a Data Sharing Portal for GLASS (the Global Antimicrobial Surveillance System) and PASS (Pakistan Antimicrobial Surveillance System).²⁹ Leveraging whole-genome sequencing for AMR surveillance and fostering educational platforms including the “Microbiology and Antimicrobial Resistance” Virtual Journal Club underline Pakistan's holistic approach.^{30,31}

In contrast to the WHO's recommended benchmark of US\$ 86 per capita spending on health, Pakistan's per capita health expenditure currently stands at just US\$ 36.³² In addition to the US\$36 spent by the government, nearly 75% of total spending on medicines in Pakistan is out-of-pocket. As a result, there is substantial purchasing of antimicrobials out of pocket especially where citizens cannot afford physician fees in addition to the cost of medicines, with the potential of driving up AMR.^{17,33–35}

This is because studies have shown that excessive purchasing of antibiotics without a prescription from the Watch list, exacerbated by concerns with the current Drug Laws.^{33,34,36} Inappropriate dispensing of antibiotics without a prescription coupled with inappropriate prescribing practices urgently needs addressing in Pakistan given continued high rates of AMR.^{33,34,37–43} Alongside this, there is currently appreciable prescribing of antibiotics from the Watch list across both primary care and hospital sectors in Pakistan, including during the COVID-19 pandemic, where up to 100% of patients hospitalized with COVID-19 were prescribed antibiotics from the Watch and Reserve lists.^{44–48}

As a result of inappropriate and excessive prescribing of antimicrobials, in 2019 Pakistan recorded a number of deaths linked to antibiotic-resistant pathogens. This included 34,400 deaths attributed to *Klebsiella pneumoniae*, 31,300 to *Escherichia coli*, 28,600 to *Staphylococcus aureus*, 23,300 to *Salmonella Typhi*, and 20,300 to *Streptococcus pneumoniae*. These pathogens were identified as the primary causes of various infections, including bloodstream, lower respiratory tract, peritoneal, and intra-abdominal infections. Pakistan appears to be the principal country among the five leading Asian countries with the highest age-standardized mortality with an AMR burden of 221,300 deaths,⁴⁹ 611,000 TB incidence cases and 16,000 MDR-TB incidence cases in 2022.⁵⁰ This again needs to be urgently addressed to reduce deaths due to AMR as part of the NAP. Globally, there were an estimated 450,000 new cases of rifampicin and multi-drug-resistant tuberculosis in 2021. Alongside this, there are concerns that invasive fungal infections are increasing globally, with an associated impact on morbidity, mortality, and costs.^{14,15,20,51–53} This is also a concern, including among patients in Pakistan.^{47,54–56}

In view of growing issues, concerns and challenges, national policies and international health commitments are now being integrated into a framework in Pakistan established by the Ministry of National Health Services Regulations and Coordination (MNHSR&C) to help tackle AMR through the NAP. The establishment of an Intersectoral Core Steering Committee by the MNHSR&C marked the first step in this process, leading to the creation of the National AMR Strategic Framework in 2016 coupled with the activation of the AMR NAP in 2017. The National Institutes of Health (NIH) emerged as a crucial entity in Pakistan,⁵⁷ serving as both the national focal point for AMR and International Health Regulation (IHR) activities.⁵⁸ The launch of GLASS by NIH in 2016,⁵⁹ and subsequently PASS in 2018,²⁹ laid the groundwork for effective AMR monitoring and management in Pakistan. Pakistan's efforts to improve future antibiotic use, as well as reduce AMR, also incorporate initiatives including the AMR Tricycle program and repeated point prevalence surveys (PPS) on antimicrobial usage generating critical insights.^{46,60–63} Further national and international efforts to reduce AMR in Pakistan include: Pakistan Global Antibiotic Resistance Partnership (GARP),¹⁷ Pakistan AMR surveillance system,²⁹ and the National TB Control Program.⁵⁰ At a micro level, AMR is being tackled through the introduction of the Antibiotic Stewardship Initiative in Pakistan (ASIP) and the Pakistan Antimicrobial Resistance Network (PARN) under the umbrella of Medical Microbiology and Infectious Diseases Society of Pakistan (MMIDSP).⁶⁴

To fill the research gap created by a lack of knowledge, the WHO developed a Global Research Agenda for AMR.⁶⁵ This initiative aims to provide a transparent and global control and prevention of resistant bacteria-related knowledge gaps assessment in the human health sector, which includes drug-resistant *M. Tuberculosis*, sexually transmitted diseases (STD) and fungal infections, within a time frame of 2023–2030. Alongside this, catalyze scientific interest, as well as focus attention, on the development of new and critical evidence on AMR to assist in the implementation of additional global, regional and national policies to reduce AMR especially among LMICs. This agenda, with its focus on the human health sector, will complement the WHO one health priority research agenda currently being developed in parallel, which focuses on interfaces between humans, animals, and plants and their shared environment.²³ Consequently, the primary aim of this review was to analyze current AMR research activities in Pakistan under the Global Health Research Agenda, identifying study types, geographic and thematic distributions, as well as existing gaps. As a result, guide future research, policy-making, and AMR control strategies in Pakistan in line with the Global Research Agenda for AMR.⁶⁵ The findings can be subsequently be used to guide all key stakeholders in Pakistan concerning future initiatives to reduce rising rates of AMR and their implications to meet NAP and UN GA goals.

Method

A systematic-narrative hybrid literature review covering five thematic areas and 40 research priorities of the global research agenda for AMR in human health was undertaken to evaluate the advancement of this agenda within Pakistan.⁶⁶ This approach was used in order to try and include all relevant articles and documents, which included those published

on the internet. We have adopted this approach before when reviewing ongoing activities and their implications to improve future antibiotic use across LMICs.^{20,67–71} The findings will be used to provide suggestions for all key stakeholder groups in Pakistan to achieve the goals of global research agenda to reduce antibiotic resistant pathogens incidence by investigating preventive strategies. In addition, increasing activities including antimicrobial stewardship programs (ASPs), building on current activities.

Global Health Research Agenda for AMR in Human Health

This policy brief outlines a comprehensive research agenda on AMR with five main themes, 13 subthemes, and 40 specific research areas. These five main themes include Prevention (3 subthemes, four areas), Diagnosis (one subtheme, six areas), Treatment and Care (three subthemes, 11 areas), Cross-Cutting Research (3 subthemes, 12 areas), and Drug-Resistant Tuberculosis (TB) (3 subthemes, 7 areas). Each theme addresses critical aspects such as WASH, rapid diagnostics, antimicrobial stewardship (AMS), epidemiology, and drug-resistant TB interventions aimed at guiding evidence-based practice policies in the future and improve AMR management both in Pakistan and globally.

Developing the Protocol

A protocol specifying the objectives, inclusion criteria, and methods for the narrative review was developed based on the study objectives. This included a number of subsections.

Eligibility Criteria and Population

The inclusion and exclusion criteria were based on the scope of the review. For example, studies focusing on AMR only on human health within Pakistan and published within a specific timeframe. The time frame included studies published from January 2000 until October 2024 to capture the most relevant and recent data on AMR research progress in Pakistan, building upon the launch of the NAP. Consequently, the most emphasis was placed on publications following the launch of the NAP and its initial challenges.¹⁷ The general population, physicians, students, pharmacists, hospitalized patients, outpatients, nurses, dentists, healthcare workers (HCWs) and other healthcare professionals (HCPs) were included.

Concept

Papers for the narrative review included those dealing with the following categories in Pakistan:

(i) prevention (WASH) and infection control practices; (ii) immunization against antibiotic resistant infections; (iii) diagnosis through biochemical, multiplex, and antigen-based testing; (iv) implementation of ASPs and their components; (v) antibiotic use and consumption, especially those broken down by the AWaRe classification; (vi) antibiotic treatment regimens for drug resistant typhoidal and non-typhoidal infections; (vii) treatments regimen for bloodstream infections; (viii) treatment of critical group fungi; (ix) treatment of sexually transmitted diseases caused by *Neisseria gonorrhoeae*; (x) epidemiology, burden of urinary tract, respiratory tract, and bloodstream infections as common infections in Pakistan; (xi) antibiotic resistance awareness and education; (xii) implementation of policies, regulations, and frameworks to mitigate AMR including those surrounding ASPs; (xiii) drug-resistant TB prevention through BCG vaccine; (xiv) diagnosis of drug-resistant, extrapulmonary, and pulmonary TB infections and (xv) new and existing regimens for TB treatment.

Recognizing and Generating Research Questions (RQ)

As mentioned, the primary objective of this review is to comprehensively analyze AMR research currently being conducted in Pakistan, as reported under the Global Health Research Agenda.

This analysis aims to identify the types of studies performed, the geographic distribution of these studies, the specific themes and sub-themes of ongoing research efforts, and the time periods over which the studies were conducted. In addition, this review seeks to identify the prevailing gaps in AMR research and practice in Pakistan, with the ultimate goal of informing future research directions, policy-making, and the implementation of effective AMR control strategies in Pakistan to meet NAP targets.

Search Strategy

Google Scholar and PubMed were used to search for relevant literature. The search strategies were drafted according to the database protocol. These included the terms related to prevention practices, antibiotic infection control practices in hospitals, antibiotic infection immunization, antibiotic infection diagnosis, ASPs, fungal infection diagnosis, antibiotic use and consumption across sectors, treatment of typhoidal and non-typhoidal infections, the epidemiology of urinary tract, respiratory tract, and blood stream infections in community and healthcare settings as well as drug-resistant TB infection prevention, diagnosis and treatment.

The grey literature and government reports were also searched to ensure comprehensive coverage in this area since many Government and other documents would not typically be published in scientific journals.

Selecting Studies and Screening Process

This study used a two-stage screening process. This began with the titles and abstracts, followed by full-text screening according to inclusion criteria. At least two reviewers were included in this study independently assessing studies for eligibility to minimize bias.

Charting the Data

Data Extraction

An agreed standardized form was used to extract data from included studies. Information collected included the study characteristics (eg, year, study design), focus areas of AMR research, findings, and implications for the global research agenda and health policy in Pakistan.

Collecting, Summarizing, and Reporting Results

The findings, in line with the objectives of the review, were organized and categorized. Tables were used to summarize key information and trends. Any gaps in research were highlighted and suggested areas for future investigation.

Stakeholder Consultation

Engaging stakeholders (eg, researchers, policymakers, and clinicians) was also considered to add depth to the review, validate the findings, and identify additional resources including unpublished studies.

A structured approach was used to help ensure that the narrative review comprehensively captures and analyses the progress and implications of AMR research in human health in Pakistan, aligning with global research agendas and identifying areas needing further investigation. Moreover, key recommendations, expected outcomes, and Priority levels are based on the considerable experience of the co-authors working in this field.

Ethical Review

No ethical approval was required for this study as it did not involve humans or animals. It was a review of published studies and references.

Results

This comprehensive review synthesized findings from 349 published studies in 40 research priority areas from 2000 until 2024. This included 23 publications (9.95%) specifically relevant to prevention, 55 (22.9%) to diagnosis, 64 (26.7%) to treatment and care, 59 (24.5%) to cross cutting, ie interconnected factors that influence the development and spread of resistance, and 44 (18.33%) relevant to drug-resistant TB. The research spans various cities in Pakistan, although predominantly in major urban centers including Karachi, Lahore, and Islamabad, focusing on different aspects of AMR (Figure 1). The types of studies included observational and intervention-based research that explored WASH practices as well as those researching diagnostic techniques, AMS, and the effectiveness of immunization against drug-resistant pathogens.

During 2001–2020, prevention-related studies mostly came from Karachi and endorsed the WASH and Infection, prevention and control (IPC) practices in curbing infectious diseases especially diarrhea and, by extension, AMR (Table 1). Encouragingly, an appreciable increase in vaccination uptake resulted in a significant step forward in

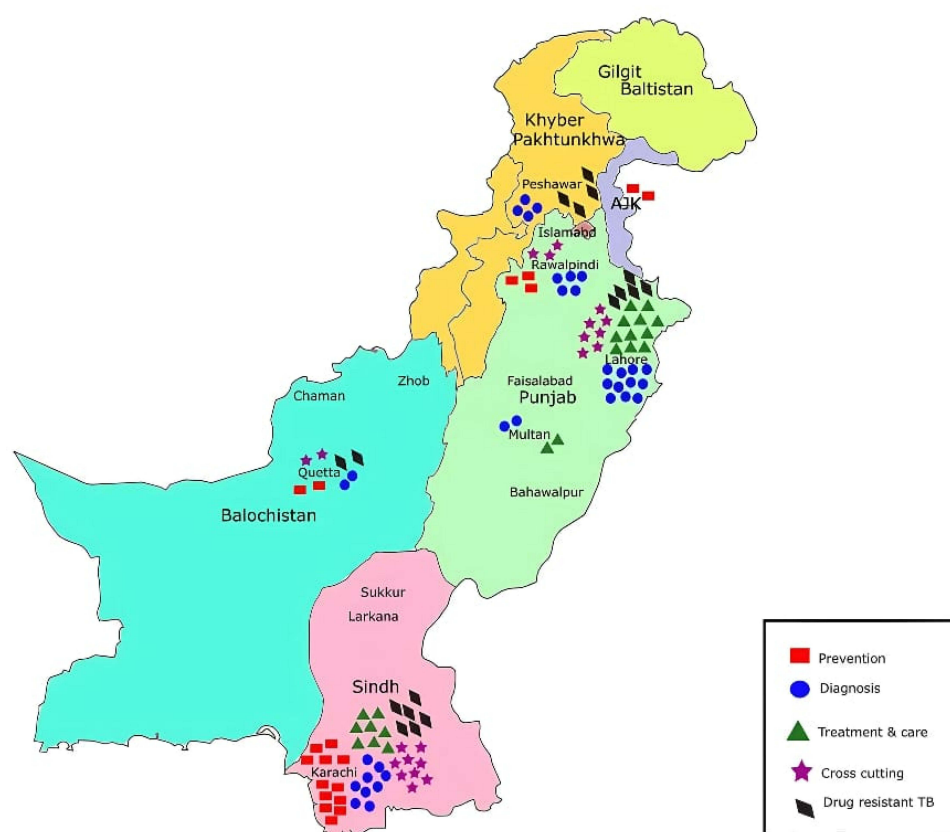


Figure 1 Geographical distribution and focus of 5 AMR areas of GHRA in Pakistan.

vaccine-driven AMR prevention in the country. Nevertheless, the persistent gap in effective practices in both hospital and community settings suggests a need for more robust policy implementation, including public health initiatives, to further reduce infectious diseases and AMR in Pakistan.

In terms of diagnosis, the review highlights diagnostic methods for resistance pathogens, susceptibility detection, use of minimum inhibitory concentration (MIC) determination and the use of advanced diagnostics, all showing the potential of distinguishing bacterial infections from viral infections (Table 2). However, the findings to date highlight a deficiency in current routine laboratory surveillance in Pakistan and the integration of ASPs at the diagnostic level, suggesting that diagnostic capacities are currently being underutilized for AMR management in Pakistan, similar to other LMICs.⁷¹ This is seen by currently limited use of culture and sensitivity testing even in tertiary hospitals in Pakistan because of the costs involved. As a result, high levels of empiric prescriptions in hospitals Pakistan, which need addressing going forward.^{60,95}

Encouragingly in recent years, treatment and care strategies have seen improvements with an increasing number of ASPs being undertaken to promote the judicious antibiotic use in Pakistan given the previous concerns (Table 3).^{43,44,95,151,162,207} However, whilst good ASP knowledge was observed in studies among doctors, pharmacists and nurses, there were concerns with the actual implementation of ASPs in practice. This suggests the need for implementing education surrounding ASP and policies to all key stakeholder groups going forward as well as ensuring that pharmacist and other HCPs play a key role in their implementation after receiving appropriate ASP training to overcome potential barriers in their implementation.^{151,152,154–156}

Cross-cutting efforts highlight the importance of an integrated approach for tackling resistant pathogens (see Table 4). This involves health system strengthening, emergency preparedness, and enhanced sanitation practices especially in countries such as Pakistan. Surveillance studies across various Pakistani regions have provided invaluable data on AMR

Table 1 Theme 1 of GHRA-Prevention

Sub Themes of GHRA	Research Priority Areas Covered as Per GHRA	References	Year of Published Studies	No. of Studies	Targeted Population	Key Findings	Study Highlights
1.WASH:	Area 1 WASH practices effect on AMR and diarrhea burden	[72–81]	2003-2022	10	Physicians, children, HCP, Quack, children and their mothers, LHV's, LHW's	Diarrhea management by <ul style="list-style-type: none"> • Hand washing • WASEP and HOPE interventions • Diarrhea pack • Bismuth sub-salicylate 	Study demonstrating the effective diarrhea management without antibiotics
	Area 2: Effect of WASH-related interventions on burden of HAI	[82–85]	2001-2022	4	HCP's	<ul style="list-style-type: none"> • Awareness on importance of adopting preventive measures • Improvement in outcomes of health-care waste, water and sanitation and hand hygiene by WASH-FIT 	Studies showing the effectiveness of WASH interventions in infection prevention and control
2.Infection, prevention and control	Area 4 Effect of different IPC practices on mitigation of HAI	[86–90]	2005-2023	5	Auxiliary health-care staff, nurses, HCW's	Improved waste disposal rate, hand hygiene, use of personal protective equipment by educational interventions and training session	Studies highlighting reduction in infection burden in health-care setting by use of IPC practices
3. Immunization	Area 4 Infection prevention by vaccination	[91–94]	2012-2023	4	9 months-15 year of children	Prevention of drug-resistant infections like typhoid by TCV, Typbar TCV	Studies showing the preventive effect of vaccines
Implications	Poor WASH practices in hospital and community highlights need of simple, effective preventive measures against AMR						

Abbreviations: LHSs, Lady Health Supervisors; LHWs, Lady Health Workers; WASEP, Water and Sanitation Extension Program; HCP, Healthcare provider; WASH-FIT, Water, Sanitation, and Hygiene Facility Improvement Tool; HAI, healthcare-associated infections; TCV, typhoid conjugate vaccine; XDR, extensive drug resistance.

Table 2 Theme 2 of GHRA-Diagnosis

Sub Themes of GHRA	Research Priority Areas Covered as per GHRA	References	Year of Published Studies	No. of Studies	Targeted Population	Key Findings	Study Highlights
4. Diagnosis and diagnostics:	Area 5 Discrimination of viral and bacterial infections by using diagnostic algorithm or by biomarkers	[96–98]	2019-2022	3	Patients' samples	<ul style="list-style-type: none"> Raman spectroscopy by using PCA and LDA algorithm techniques for HCV detection Detection by using specific biomarkers like PCT, CRP, TLC 	Studies demonstrating the use of advanced diagnostics for differentiation of bacterial and viral infections
	Area 6 Antimicrobial resistance and susceptible detection by using phenotypic and genotypic methods	[99–117]	2013-2023	19	Patients	<ul style="list-style-type: none"> Phenotypic detection by Kirby Bauer disc diffusion, double disc synergy, combined disk Genotypic detection by monoplex and multiplex PCR 	Successful identification of resistant, susceptible and gene responsible for conferring resistance was achieved
	Area 7 Identification of resistant and susceptible antimicrobials by using rapid point of care tests	[118–140]	2004-2023	23	Patients' samples	Point-of-care tests include phenotypic methods like double disk synergy, combination disk, biochemical testing and gram staining, genotypic characterization by single plex, multiplex, nested and regular PCR, qualitative assay by real-time PCR	Highlight the use of novel point-of-care diagnosis tests for resistance and susceptibility detection
	Area 8 Detection of WHO critical group fungal priority pathogens	[141–145]	2015-2023	5	Patients' samples	For <i>Candida</i> spp detection carried out by biofilm method, by micro titer plate method, by disk diffusion method while <i>Aspergillus</i> spp were detected by IgG cut-off diagnosis and by dilution method	Studies demonstrating the effectiveness of different diagnostics test for identification of resistant fungal pathogens
	Area 9 Determination of diagnostic accuracy of phenotypic antifungal susceptibility testing	[146,147]	2013-2020	2	Patients' samples	Diagnostic tests like antifungal agar screening method, MIC determination by microdilution method used for detection of <i>Candida</i> and <i>Aspergillus</i> spp	Studies showing the importance of MIC determination for selection of antifungal treatment regimen
	Area 10 Evaluate the performance of rapid point-of-care test for detection of <i>Neisseria gonorrhoeae</i>	[148–150]	2016-2021	3	Patients' samples	<i>Neisseria gonorrhoeae</i> was successfully detected by multiplex PCR, by multiantigen sequence typing, calibrated dichotomous sensitivity disk diffusion, MIC and by E test	Studies showing the identification of resistant antibiotic for <i>Neisseria gonorrhoeae</i> to reduce inappropriate prescribing
Implications	Lack of laboratory surveillance, antibiograms, low capacity of laboratory for ASP integration supports the designing of appropriate antibiotic regimen and rational use of antibiotics						

Abbreviations: HCV, hepatitis C virus; PCT, procalcitonin; ASP, antimicrobial stewardship program; ESBL, extended spectrum beta lactamase; MDR, Multi-Drug resistance; PCR, polymerase chain reaction; MRSA, methicillin-resistant *Staphylococcus aureus*; MSSA, methicillin-susceptible *Staphylococcus aureus*; S. Typhi, *Salmonella Typhi*; MIC, minimum inhibitory concentration.

Table 3 Theme 3 of GHRA-Treatment & Care

Sub Themes of GHRA	Research Priority Areas Covered as per GHRA	References	Year of Published Studies	No. of Studies	Targeted Population	Key Findings	Study Highlights
5.Antimicrobial stewardship:	Area 11 Implementation of antimicrobial stewardship interventions to reduce antimicrobial misuse	[151–153]	2020-2022	3	HCP's	Following AMS related interventions were adopted <ul style="list-style-type: none"> • Consideration of regular audit & feedback by nurses, physicians & pharmacists • Participation in educational sessions 	Studies demonstrating the importance of ASP for AMR mitigation in community as well as in health-care settings
	Area 12 Role of pharmacist dispensing practices in implementation of antimicrobial stewardship	[154–158]	2018-2023	5	Hospital staff and community pharmacies staff	Pharmacists played significant role in <ul style="list-style-type: none"> • Discouragement of non-prescription antibiotic dispensing practices • Patient education • Compliance with DTC's guidelines 	Studies highlighting the positive role of pharmacist in ASP implementation and practices
	Area 13 Optimization of empirical antibiotic therapy by various strategies	[159–161]	2019-2023	3	Hospitalized patients	Empirical therapy practices were improved by <ul style="list-style-type: none"> • Selection and initiation of antibiotics according to recommended guideline • De-escalation of antibiotics • PAP audit 	Studies highlighting the need of evidence based empirical therapy
6.Antimicrobial use and consumption	Area 14 Methods to monitor antimicrobial use and consumption	[45, 46, 95, 162–171]	2015-2023	13	Patients from OPD's, emergency, medicine, gynecology, physicians, general population HCW's	Methods to determine irrational antibiotic use were <ul style="list-style-type: none"> • Dispensing of partial antibiotic course by pharmacists • Absence of guidelines or policies for antibiotic prescription • Completion of incomplete antibiotic course by patients • Self-medication and empirical therapy 	Studies determining the overall consumption and use of antibiotics by different methods
	Area 15 Use and consumption of AWARe group antibiotics	[33, 35, 62, 172–182]	2017-2023	14	Term neonates, hospitalized patients, student, pharmacy staff, drugstore staff, children, pharmacy technician	Highly prescribed/used antibiotics were from watch group followed by access and reserve group	Studies showing the overall consumption of AWARe group antibiotics
	Area 16 Monitoring of antimicrobial consumption and resistance surveillance data to inform antimicrobial stewardship programs and treatment guidelines	[183–187]	2015-2023	5	Inpatients and outpatients	Emergence of MDR, XDR organisms especially in enteric fever confirmed by surveillance methods like active surveillance which demand the ASP implementation	Studies demonstrating the importance of antimicrobial surveillance data in ASP

(Continued)

Table 3 (Continued).

Sub Themes of GHRA	Research Priority Areas Covered as per GHRA	References	Year of Published Studies	No. of Studies	Targeted Population	Key Findings	Study Highlights
7. Antimicrobial medicines	Area 17 Safe and effective antibiotic monotherapy & combination therapy for ESBL enterobacterales or CRE	[188–190]	2022-2023	3	Patients	Efficacy of in-vitro Ceftazidime-Avibactam (CAZ-AVI) and Meropenem-Vaborbactam (MEV) combination is decreasing day by day due to MBL and serine presence while cefiderocol was highly effective as only 4% isolated were resistant against this antibiotic	Studies showing the effective antibiotic options for <i>Enterobacterales</i>
	Area 18 Safe and effective antibiotic regimen for infection by typhoidal and non-typhoidal <i>Salmonellae</i>	[91, 94, 191–195]	2020-2023	7	Children, patients, general population	For XDR typhoid monotherapy with meropenem, azithromycin was more effective as compared to combination therapy with meropenem + azithromycin TCV also proved as beneficial	Studies showing the efficacious antibiotics for typhoidal and non-typhoidal infections
	Area 19 Safe and effective antibiotic regimen for bloodstream infections and sepsis	[196–201]	2012-2023	6	NICU neonates, patients, infants, hematology children	Use of colistin alone or in combination therapy was effective	Studies showing the effective antibiotics for BSI and sepsis
	Area 20 Safe and effective treatment regimen for infections caused by WHO fungal priority pathogens	[202–205]	2012-2021	4	Hospitalized patients	Effective antibiotics for <i>Candida</i> spp were • Fluconazole 150 mg in a single dose and itraconazole 200 mg twice for 1 day For <i>Aspergillus</i> spp were • Voriconazole 400 mg per day and surgery	Studies demonstrating the overall efficacious antibiotics for critical group fungal pathogens
	Area 21 Safe and effective antibiotic treatment regimen for STD's caused by <i>Neisseria gonorrhoeae</i> and <i>Mycoplasma genitalium</i>	[206]	2022	1	Patients	Effective treatment for <i>N. gonorrhoeae</i> was injection of 500mg ceftriaxone and 2gm spectinomycin	Studies showing the antibiotics used for STD's management
Implications	Barriers in implementation of ASP like knowledge gaps, incomplete compliance, absence of ASP core elements, absence of effective methods and metrics for estimating the national or regional level antibiotic consumption need to be addressed						

Abbreviations: OPD, outpatient department; AWaRe, Access, Watch, Reserve; BSI, bloodstream infection.

Table 4 Theme 4 of GHRA-Cross Cutting

Sub Themes of GHRA	Research Priority Areas Covered as per GHRA	References	Year of Published Studies	No. of Studies	Targeted Population	Key Findings	Study Highlights
8.Antimicrobial resistance epidemiology, burden and drivers	Area 22 Epidemiology and socioeconomic impact of health-care associated and community acquired infections	[208–217]	2014-2023	10	Patients including Indoor, outdoor, pediatric), neonates	Varied incidence, prevalence, mortality and morbidity of different bacterial infections among different age groups were observed	Studies showing the epidemiology of respiratory tract, urinary tract, bloodstream and surgical site infections
	Area 23 Incidence, prevalence, morbidity, mortality caused by WHO fungal priority pathogens among co morbid and vulnerable population	[55, 218, 219]	2019-2022	3	Patients from Gynae& Obstetrics, pediatric patients	High prevalence of <i>Candida</i> spp especially <i>Candida albicans</i> was observed in females especially in pregnant women and high mortality rate in hospitals occurred due to <i>Candida auris</i>	Studies determining the overall epidemiology of WHO critical group fungal pathogens
	Area 24 Impact of health system factors such as hospital microbiome on bacterial and fungal colonization	[220–226]	2013-2024	7	Neonates, hospital personnel, ICU patients, pediatric patients	Hospital microbiome was found to be associated with the high colonization rate of vancomycin-resistant and vancomycin-susceptible enterococci among hospitalized neonates, pediatrics	Studies showing the relation between hospital microbiome and drug-resistant infections colonization rate
	Area 25 Optimal surveillance data on epidemiology and burden of AMR in community and health-care settings	[183, 184, 227–231]	2006-2020	7	Patients (inpatients, outpatients pediatrics, community)	Burden of multi-drug-resistant organisms estimated by surveillance methods like <ul style="list-style-type: none"> • Population based • Laboratory based • Active • Sentinel • Prospective • Passive 	Studies defining the role of surveillance methods for estimation of AMR burden

(Continued)

Table 4 (Continued).

Sub Themes of GHRA	Research Priority Areas Covered as per GHRA	References	Year of Published Studies	No. of Studies	Targeted Population	Key Findings	Study Highlights
	Area 26 Short- and long-term impact of AMR on use of antimicrobial medicines especially among vulnerable population	[232–238]	2019-2023	7	Medical inpatients, outpatients, neonates and children, pediatrics	Some long- and short-term impact of antimicrobial resistance were increased risk of HAI, increased risk of side effects, increased antibiotic resistance and increase in drug resistance of different infections like MDR and XDR typhoid	Studies demonstrating the long term and short-term effects of AMR
	Area 27 Impact of unnecessary and inappropriate antibiotic prescribing and potential consequences caused by <i>Neisseria gonorrhoeae</i>	[239–241]	2011-2022	3	Female patients, infertile women	Harmful antimicrobial consequences of currently recommended STD's management were increase in antibiotic resistance of <i>N. gonorrhoeae</i> , increased fertility rate, increased risk of pelvic inflammatory disease	Studies demonstrating the potential consequence of inappropriate antibiotic prescribing in STD's
9. Antimicrobial resistance awareness and education	Area 28 Mitigation of antimicrobial resistance by awareness and education among general population	[242–248]	2018-2022	7	Students, adults, patients, clinical staff	Awareness regarding AMR was raised by ASP introduction, AMR week, student moderated session, Global Antimicrobial Resistance Surveillance System (GLASS) and campaigns for antimicrobial misuse and abuse	Studies showing that how AMR reduction can be achieved by increasing awareness

10.Policies and regulations related to antimicrobial resistance	Area 29 Mitigation of AMR by implementation of AMR related policies and regulations	[249–251]	2019-2023	3	Doctors, pharmacist, administrative staff, patients, clinical microbiologists, associate professor, head of clinical department	Treatment failure and high mortality rate due to irrational antibiotic use can be controlled by introduction of ASP as carbapenem ASP intervention resulted in low mortality and improved clinical outcomes	Studies demonstrating the role of AMR policies
	Area 30 Cost-effective implementation of national policies, regulation to improve patient care and infection prevention	[252, 253]	2007-2017	2	ICU patients, pharmacists, residents, MS, AMS, DMS, HOD's of different wards	To tackle the situations like lack of IPC policies lack of well-defined system for human resources different interventions like 7S McKinsey framework and restriction of broad-spectrum antibiotics use were adopted	Studies highlighting the role of AMR regulations and policies in infection prevention
	Area 31 Cost-effective interventions for the mitigation of AMR in human health sector	[254–256]	2009-2024	3	Community pharmacists, hospitalized patients, health-care students	Educational interventions, training sessions and ASP knowledge helped to raise awareness regarding AMR	Studies demonstrating the importance of AMR related intervention
	Area 32 Integration of antimicrobial resistance intervention into broader health to reduce AMR burden	[43, 257–259]	2020-2023	4	AMS, MS, DMS, outpatients, sick infants	AMR problems like incorrect prescribing, dispensing, high antibiotic usage rate of AWWaRe group antibiotics can be addressed by emphasizing all the health-care stakeholders to take substantial measures for implementation of ASP and its core elements	Studies depicting the expansion of AMR interventions at a large scale
	Area 33 Effect of existing regulatory framework and sustainable financing models on development and availability of new antimicrobial medicines	[260–262]	2017-2022	3		Current gaps in drug monitoring laws, policy investment in drug regulation, absence of guidelines for sale of OTC antibiotics led to emergence of resistant pathogens which can be addressed by use of antibiotics that are highly susceptible against these pathogens should be considered	Studies highlighting the role of regulatory framework in antibiotic development and its availability
Implication	Lack of enhanced infrastructure, education, training, proper surveillance, poor public awareness, lack of antimicrobial audit and feedback in hospitals highlights the need for a multifaceted strategy against AMR						

Abbreviations: MDRO, Multi Drug Resistant Organisms; VRSA, Vancomycin Resistant Staphylococcus aureus; HOD, Head of Department.

trends in active, passive, and sentinel surveillance pinpointing the high prevalence of drug-resistant pathogens across healthcare settings in the country.

Although there are currently considerable efforts to map the epidemiology of resistant pathogens in Pakistan, studies highlight a lack of comprehensive data collection that integrates findings into national health policies. This suggests the urgent need for a unified national surveillance system to improve understand and respond to AMR trends. Moreover, there is also an evident lack of widespread AMR awareness and education among both the general population and healthcare workers in Pakistan.^{71,162} This underscores the necessity for national campaigns and regular training programs to enhance understanding and appropriate responses to AMR among all key stakeholder groups.⁷¹

Antitubercular therapy (ATT) and fluoroquinolones have proven effective in treating and preventing drug-resistant TB both pulmonary and extrapulmonary drug-resistant TB, particularly when used in combination regimens. This is important in Pakistan, given the current concerns (Table 5). The addition of medicines, including bedaquiline and delamanid, into treatment regimens have resulted in higher success rates and addressed the safety and efficacy concerns, which is encouraging.

Diagnostic advancements, including molecular assays and public-private models, enhance TB detection, which is also encouraging. Strategies adopted to combat drug resistant TB include vaccination (BCG),^{263–267} early screening (by Genotypic MTBDRplus and MTBDRsl assay and/or GeneXpert MTB/RIF assay),^{271–276} and complete adherence to TB treatment protocols (shorter treatment regimens, longer treatment regimen either in the form of monotherapy or combination therapy, and DOT therapy).^{290,292–294}

Cost-effective preventive treatments and strategies including DOT proved impactful in increasing treatment completion rates and managing comorbid conditions. These should be routinely available in Pakistan.

Discussion

We believe this study represents the first extensive examination of the Global Research Agenda for AMR in human health and specific to any LMIC. In this case, highlighting key areas for research gaps in AMR within Pakistan to meet NAP and UN GA goals.

Notably, the studies predominantly originate from urban centers, reflecting a common trend in global health research where major cities act as focal points for scientific inquiry, owing to their better healthcare infrastructures and access to resources. In addition, Universities are typically located in urban versus rural areas. Geographically, the concentration of AMR research in urban areas highlights a significant gap in our understanding of AMR dynamics in rural settings.³⁰⁶ This disparity suggests potential variations in AMR prevalence and management, which may be driven by differences in healthcare access, community health practices, and local policy enforcement.^{307,308} For instance, we are aware that there can be greater purchasing of antibiotics without a prescription in rural areas of LMICs where there are high co-payment levels due to the costs and time involved with seeing HCPs in primary care clinics versus the convenience of community pharmacies.^{69,309,310} In addition, community pharmacists may often be the principal HCP available in rural settings.^{36,69,311} Addressing this gap is crucial for developing region-specific strategies that are inclusive of rural healthcare dynamics. Implementing comprehensive strategies including preventive measures, rapid diagnostics, awareness strategies among patients and HCPs and implementing pertinent ASPs, will significantly reduce AMR levels and the subsequent deaths caused by resistant pathogens in Pakistan.³⁰⁷

The GHRA prioritizes WASH practices and infection prevention measures as essential for reducing AMR burden in community and healthcare settings. Among the studies evaluated in Pakistan,^{72–75,82} adopting hand hygiene interventions as part of IPC practices appeared to result in a reduction in antibiotic associated diarrhea. However, there was insufficient knowledge and lack of awareness among HCWs regarding hand hygiene in Pakistan,^{83,84} alongside an absence of IPC standards in Islamabad public hospitals in 2019. This also needs addressing going forward to reduce AMR in the future.

Encouragingly, during a catch-up campaign in Karachi, the Typhoid Conjugate Vaccine (TCV) proved to be 98% effective for children 9 months–15 yr during 2020.⁹¹ Similar findings have also been observed in other LMICs including Bangladesh, Zimbabwe, and Malawi. They also revealed the efficacy of existing vaccines including PCV, TCV against pneumonia and typhoid resistant pathogens in children, adults and neonates.^{312–317} As a result, it is imperative that these vaccines be encouraged going forward in Pakistan.³¹⁸

Table 5 Theme 5 of GHRA-Drug-Resistant TB

Sub Themes of GHRA	Research Priority Areas Covered as per GHRA	References	Year of Published Studies	No. of Studies	Targeted Population	Key Findings	Study Highlights
Prevention	Area 34 Impact of TB preventive practices on infection prevention, recurrence, on incidence of drug-resistant TB	[263–270]	1998-2023	8	Index patients, adults, infants, children residing with index patients	Preventive treatment with fluoroquinolones and BCG vaccine proved effective against pulmonary and extra pulmonary TB especially among children	Studies showing the preventive measures for reduction of TB burden
Diagnosis	Area 35 Detection of drug resistance among people with pulmonary and extra-pulmonary TB from non-respiratory specimens	[271–285]	2002-2021	15	Patients (inpatients & outpatients) samples	<ul style="list-style-type: none"> • Detection by conventional diagnostic methods along with molecular assays Xpert MTB/RIF assay, Genotype MTBDRplus and MTBDRsl assay • Phenotypic and genotypic detection by probe line assay • Phenotypic detection by BACTEC MGIT 960 medium • Direct detection by FAST Plaque TB assay • Molecular assay by drug proportion method 	Studies assessing the overall performance of TB diagnostics methods
	Area 36 Optimal diagnostic and treatment models to improve drug-resistant TB detection especially among vulnerable population	[286–289]	2004-2019	4	Patients' samples	<ul style="list-style-type: none"> • Improved TB case detection by models like public private mix model • Detection by acid –fast bacilli among prisoners' Serological test for TB detection especially among co morbid patients like HIV, hepatitis 	Studies showing effective and feasible methods adopted for TB detection

(Continued)

Table 5 (Continued).

Sub Themes of GHRA	Research Priority Areas Covered as per GHRA	References	Year of Published Studies	No. of Studies	Targeted Population	Key Findings	Study Highlights
Treatment and care	Area 37 Effective, optimally dosed and shorter combination regimen for treating all forms of drug-resistant TB	[201, 290, 291]	2021-2023	3	Patients, children, adolescents	Improved treatment outcomes by <ul style="list-style-type: none"> • Shorter regimen with isoniazid and combination regimen with isoniazid + rifapentine • Use of Antitubercular therapy (ATT) and anti-retroviral therapy (ART) co-administration among co-infected patients 	Studies demonstrating the effectiveness of TB treatment with shorter combination regimen
	Area 38 Cost effective, safest and shortest duration TB preventive treatment for contact of people with drug-resistant TB	[292–297]	2006-2023	6	Patients	Use of shorter treatment regimen, interventions like educational, nutritional, psychological and economic support showed successful treatment completion rate among TB patients	Studies showing the efficacy of safe TB treatment regimen prescribed for short duration
	Area 39 Strategies to improve treatment outcomes among vulnerable population and those who have known risk factors and co-occurring conditions	[298–302]	2001-2021	5	Patients	DOT strategy, use of 2 nd line antibiotics was adopted to improve treatment outcomes not only among TB patients but also in co morbid patients like with diabetes, depression, HIV	Studies showing the use of various strategies like directly observed treatment, short course (DOTS) for improving outcomes
	Area 40 Safety of currently used WHO recommended treatment regimen including combinations with bedaquiline, delamanid and/ or pretomanid for drug resistant TB	[303–305]	2021-2022	3	Children and patients	Conventional treatment without bedaquiline showed a high death rate, treatment failure rate while addition of bedaquiline and delamanid with longer treatment regimen result in high treatment completion rate	Studies demonstrating the effectiveness of combination treatment regimen with bedaquiline and delamanid
Implications	Lack of preventive measures in rural areas, inaccurate diagnostics, partial or incomplete compliance to existing TB treatment protocol emphasize the importance of rapid detection methods, DOTS strategy and evidence-based use of WHO recommended TB treatment regimen						

One significant barrier to AMR management in Pakistan is the limited availability and usage of reliable diagnostic facilities including culture and susceptibility testing within hospitals, including even within tertiary hospitals.^{62,319} This diagnostic gap leads to high dependence on empirical prescribing across facilities in Pakistan, a common trend seen also across many LMICs.^{67,68,320} Diagnostic advancements, including point-of-care testing and phenotypic and genotypic methods for pathogen resistance identification, have significantly contributed to AMR management.³²¹ The use of rapid diagnostics helps differentiate bacterial from viral infections, minimizing unnecessary antibiotic prescriptions. Different biomarkers levels including Procalcitonin (PCT), C-reactive protein (CRP) levels and total leukocyte count (TLC) have also been used to discriminate between bacterial from viral infections.³²² These are possibilities for Pakistan in the future to achieve its NAP goals.

Our findings also reveal appreciable AMR level in Pakistan, which again needs to be urgently addressed for Pakistan to achieve its NAP as well as UN GA goals. This includes a high prevalence of resistant pathogens including *K. pneumoniae*, *S. Typhi*, *E. coli*, and *S. aureus*, which have been documented to cause bloodstream, respiratory, and abdominal infections.^{323–325} This aligns with findings from neighboring countries, including India and Bangladesh, where overuse of antibiotics, particularly in urban healthcare centers, has resulted in similar high rates of resistant bacterial pathogens.^{326,327} In Pakistan, *S. Typhi* pathogens were found to be resistant against 1st and 2nd line antibiotics, which incorporate nalidixic acid, azithromycin, and 3rd generation cephalosporins detected by phenotypic and genotypic methods.^{99–101} However, in another study, *S. Typhi* pathogens were found to be resistant against chloramphenicol, ampicillin and ciprofloxacin while showing a higher susceptibility against azithromycin, imipenem and meropenem detected by molecular characterization.¹⁰² Resistant rates need continual monitoring to guide future empiric prescribing in the locality.

Epidemiological data regarding resistant bacterial and fungal pathogens has also been used to understand how AMR evolves and spreads over time.³²⁸ In addition, how AMR impacts on public health both immediately and in future. Some common short and long-term effects include an increase in drug resistance cases of *S. Typhi* from MDR to XDR.^{232–234} Alongside this, an increase in healthcare-associated infections among hospitalized patients in Pakistan, including among neonates and children.^{235,236} Alongside this, increased risk of side-effects among medical inpatients.²³⁷ Due to an increase in resistant *E. coli*, which is the causative agent for urinary tract infections (UTIs), limited treatment options are now available in Pakistan with imipenem, amikacin, and tazobactam.^{118–121} However, in Nishtar Hospital of Multan, the susceptibility patterns were different. *E. coli* showed 80% resistance to imipenem; however, tazocin, nitrofurantoin, and amikacin were found as highly susceptible antibiotics in urinary tract infections.¹²² We believe these different susceptibility patterns are due to different antibiotic usage practices and resistance profiles of bacteria in the different regions of Pakistan. This knowledge is essential to help health-care providers select effective antibiotic treatments for the different bacterial infections they see in the different regions in Pakistan. The different resistance patterns identified can subsequently be incorporated into revised WHO AWaRe guidance by locality and region in Pakistan.^{24,25,27}

The epidemiological data for urinary, respiratory and blood stream infections in different cities in Pakistan were compared. For UTIs, *E. coli* was found as causative agent followed by *K. pneumoniae* and its prevalence was lower in males (8.9%) compared to females (13.8%) in Khyber Pakhtunkhwa.²⁰⁸ Similar results were obtained in Gilgit Baltistan in which the prevalence of UTIs in males was 33.6% versus females at 66.5%.²⁰⁹ The rising UTI burden, especially in females, was also observed globally with *E. coli* found as the most predominant causative organism.^{329–332} Most prevalent causative agents for blood stream infections in Pakistan include coagulase negative *Staphylococci* at 33.1%, *S. aureus* at 23.1%,²¹⁰ and *P. aeruginosa* and *S. Typhi* at 21.6%, with the prevalence of blood stream infections high in children aged ≤48 months (69.30%) or those with low socioeconomic status (56.8%).²¹¹ A varied prevalence pattern of respiratory tract infections (RTIs) was observed among different age groups in Pakistan, which included 61.6% in males, 38.4% in females and 36.2% in infants aged ≤6 months.²¹² However, adult males with a low socioeconomic status (54.16%) and infected with UTIs were more prone to RTIs.²¹³ Different surveillance methods, including active, passive, prospective, population based and hospital-based surveillance, have been used to estimate the epidemiology of different pathogens in Pakistan. These include *Proteus* spp, *Acinetobacter* spp, *K. pneumoniae*, *Haemophilus influenza* type B, *S. pneumonia*, *Neisseria meningitides*, *Shigella* spp, *S. Typhi* and *Paratyphi* in Pakistan.^{183,184,227–231}

Epidemiological data of fungal pathogens indicate a high prevalence *Candida albicans* in vulvovaginal candidiasis (37.2%) and oropharyngeal candidiasis (68.6%). Alongside this, a high prevalence of vulvovaginal candidiasis among females aged 26–30 (65.6%) as well as in pregnant women (43.2%).²¹⁸ A few studies have demonstrated the effectiveness of antifungals including itraconazole for vaginal candidiasis and voriconazole for mycetoma, which emphasize the importance of choosing the appropriate anti-fungal regimen for treatment based on resistance patterns to specific fungi.^{202–204} Otherwise, rates of resistant fungal pathogens like *Candida auris* will continue to grow if antifungal agents are prescribed inappropriately.³³³

Epidemiological data from hospitals has also shown that hospital microbiomes play an important role in infection transmission either by high colonization with vancomycin-resistant and vancomycin susceptible *enterococci*²²⁰ or through transmission of *S aureus* by hospital personnel.²²¹ There was also an appreciable presence of *S. aureus*, resistant vancomycin strains, and MRSA among hospitalized patients in Pakistan,²²² with tourniquets the potential transmission source for MRSA.²²³

There are also concerns for the management of sexually transmitted diseases (STDs) in Pakistan, including gonorrhea, for which there was an increase in the resistant pathogen rate from 0% in 1992 to 70.8% in 2009.²³⁹ Alongside increased infertility rates,²⁴⁰ and an increased risk of pelvic inflammatory disease among women infected with *Chlamydia trachomatis* infection was also observed.²⁴¹ This increase in antibiotic resistant strains to STDs in Pakistan, particularly gonorrhea, necessitates urgent action to develop new antibiotic treatment strategies. About 29.17% susceptibility of ciprofloxacin against *N. gonorrhoeae* suggests its use as first line therapy is currently in question in Pakistan.¹⁴⁸ This is in contrast with the findings of Karachi, which recommend the use of ceftriaxone and spectinomycin as first line therapy in gonorrhea due to 100% susceptibility to *N. gonorrhoeae*. Alongside this, the exclusion of ciprofloxacin from 1st line antibiotic therapy owing to 86% resistance.¹⁴⁹ In a study conducted in Kohat and Bannu, the response rate for 2g spectinomycin injection was high as compared to a 500 g ceftriaxone injection for *N. gonorrhoeae*. However, there was concern that spectinomycin must be used cautiously because it is contraindicated in pregnancy.²⁰⁶ These studies implicate that the current treatment guidelines for STDs need to be updated in Pakistan to address emerging resistance patterns and to ensure effective treatment.

Previously, the implementation of ASPs has faced barriers across LMICs including a lack of funding and pertinent personnel. However, this is now changing in LMICs with an appreciable number of ASPs now being implemented across sectors.^{68,71,242,334–337} When an ASP was introduced for patients who were on carbapenem therapy in Karachi, there was an improvement in antibiotic use, duration, and dose in 84.6% of patients, with an acceptance rate of 87.3%.²⁴⁹ Other ASPs in Pakistan have also resulted in a reduction in irrational antimicrobial prescribing and dispensing across sectors alongside improved communication between prescribers and pharmacists to improve future prescribing.^{243,338} High rates of antibiotic prescribing among for instance pediatric patients in Pakistan, with the majority from the Watch group (72.1%), coupled with a limited number from Reserve group (2.2%), similar to other studies in Pakistan, demand robust ASPs going forward.^{318,339} However, a current concern is the lack of knowledge and application of ASPs among physicians treating pediatric patients even in tertiary hospitals in Pakistan. Consequently, ASPs should be urgently instigated in these hospitals, with trainee physicians in these hospitals becoming the prescribers of tomorrow when qualified.³⁴⁰ In addition, the effectiveness of ASPs typically requires the need for DTCs for approval for the prescribing of restricted antibiotics in formularies to enhance their appropriate use.^{153,155,156} However, a concern going forward is the appreciable dispensing of non-prescribed antibiotics especially in Punjab (96.9%) Swat (78.4%), Lahore (67.1% for diarrhea and 50.9% for URTI) and Karachi (52%) due to a lack of education among key stakeholder groups and the absence of guidelines relating to their rational use.^{33,163,164} Alongside this, high rates of empiric antibiotic prescribing were the main cause of unnecessary antibiotic use, with metronidazole, ciprofloxacin, ceftriaxone and vancomycin all highly consumed antibiotics across the sectors.^{35,46,163,165,166,172}

As mentioned, another identified area of concern in Pakistan was generally the high prescribing of antibiotics, particularly those from the Watch group, among children and neonates.^{172,173,318} The overuse of antibiotics from the Watch group has also been observed in other LMICs, enhanced by dispensing of antibiotics without a prescription, which demands targeted AWaRe-based ASPs, with increasing use of the AWaRe system in practice to document utilization patterns.^{71,341,342} Similar results were obtained from the study involving 76 countries, which also showed a high consumption rate of antibiotics from the Watch

group. As a result, meeting the previous WHO target of at least 60% of antibiotic consumption being from the Access group is difficult to achieve with increasing AMR and the need for new antibiotics.^{12,26,71,343} There are also concerns in Pakistan with the appreciable availability of antibiotics from the Watch list, including multiple branded generics, exacerbating their use.³⁴⁴ This also needs addressing going forward to help attain UN GA targets for Access antibiotics.²⁸ Several studies in Pakistan have also highlighted the importance of education regarding antibiotics and AMR. Alongside this, how increased awareness through educational campaigns, changes in behavior, have helped to reduce antibiotic resistance and resulted in a reduction of nosocomial infections.^{71,206,254,338,341} Improved surveillance as well as ASPs have also helped.^{67,68,148,244}

Pakistan currently faces a high burden of drug-resistant TB, exacerbated by incomplete adherence to treatment protocols and limited access to timely diagnostics. Different studies were conducted to evaluate the efficacy of different preventative therapies for drug-resistant TB. In different cities of Pakistan, the BCG vaccine scar presence (55.19%) or absence (44.18%) ratio was less than one, which results in the lack of effectiveness of BCG vaccine against TB.²⁶⁷ Although in other studies conducted in Greenland, Kazakhstan, and the UK BCG vaccine was effective against *M. tuberculosis*.^{345–347} The efficacy of fluoroquinolones-based preventive therapy was demonstrated by a 70% complete cure rate with no side-effects among index patients in Karachi,²⁶³ coupled with a 65% reduction in TB exposure rate among children and adults residing with index patients.^{264,345–347}

Different conventional susceptibility testing methods, which include MTBDRplus and MTBDRsl assays, along with GeneXpert MTB/RIF assay, were used to detect gene mutation that confer drug resistance in *M. tuberculosis*.^{271,272} In a study conducted in Karachi, the *rrs* gene mutation was shown by all XDR TB strains, and this mutation was responsible for conferring resistance to amikacin and kanamycin.²⁷⁷ The *rpoB* gene mutation was responsible for conferring resistance to rifampicin isolates detected by phenotypic methods,²⁷⁸ especially important in areas with high TB burden. However, there is still a need for specific probe assays,²⁷¹ improvement in infrastructure,²⁷⁵ and the introduction of the GeneXpert MTB/RIF assay especially in rural areas in Pakistan, for improved diagnosis and management of TB in the country.²⁷⁹

Regarding the treatment of drug-resistant TB, different strategies have been introduced in Pakistan to try and address the problem. These include both short treatment regimen, long treatment regimen and directly observed treatment (DOT) strategies. The first study in Pakistan to assess the outcomes of the short treatment regimen for TB treatment was undertaken between 2018 and 2019. This showed an 87.3% treatment success rate,²⁹² with only a 73.2% treatment success with the long treatment regimen.²⁹⁴ Having said this, treatment with the longer treatment regimen of bedaquiline and delamanid resulted in an 81.7% treatment success rate, 18.3% treatment failure rate and 16.4% death rate among rifampicin resistant MDR-TB patients.³⁰³ Encouragingly, a study in eight TB facilities of Karachi showed a treatment completion rate higher than 90% among 678 household contacts with one month of daily isoniazid and rifapentine (IHP).²⁹⁰ The findings of these studies were consistent with the findings from other countries showing the efficacy of a shorter treatment regimen for patients with TB in terms of the treatment completion rate, successful treatment outcomes, and lower costs compared with the longer treatment regimen.^{348–352} A DOT strategy was more effective, with a 64% cure rate,²⁹⁸ as well as costs for key groups.²⁹⁹ However, owing to higher costs and time, including those associated with travelling to clinics, the DOT strategy was less effective for the poor with monthly incomes of less than \$7.2 as the cost of DOT was more than \$10.8.²⁹⁵ Contrastingly, community-based DOT was seen as more effective, with enhanced accessibility for patients living in remote areas, as there was enhanced adherence especially when administered within community compared with the health facility-based DOT.^{353–355} When drivers and risk factors for unfavorable TB outcomes were checked, being male above 60 years and having co-morbidities including Human Immunodeficiency Virus (HIV), diabetes, and XDR-TB, and a treatment history with 1st and 2nd line anti-TB drugs, were found as key drivers of drug-resistant TB in Pakistan.^{300,304} All these factors highlight the need of comprehensive care strategies and targeted interventions for the prevention of drug-resistant TB in vulnerable population in Pakistan. This aspect will be explored in future studies.

Suggested Next Steps

Several activities have evolved from the findings of this review to help reduce AMR in Pakistan. These are summarized in Table 6.

We will be following up a number of these suggestions in future studies.

Table 6 Recommendations

Theme	Recommendations	Target Stakeholders	Expected Outcomes	Priority Level
Prevention	- Enhance WASH Initiatives: Strengthen WASH programs in healthcare and community settings.	Government, NGOs, Community Leaders	Reduced incidence of healthcare-associated and community-acquired infections.	High
	- Expand Infection Prevention Controls: Develop and enforce stricter IPC measures and provide routine training for healthcare workers.	Hospital Administrators, Healthcare Workers	Improved compliance with IPC protocols, reducing the spread of AMR and use of antimicrobials as prophylaxis.	Very High
Diagnosis	- Broaden Access to Diagnostic Tools: Increase accessibility to advanced diagnostic tools, especially in rural areas.	Government, Healthcare Providers	Enhanced early detection and targeted therapy based on antibiogram.	High
	- Integrate Diagnostic Training: Incorporate training on the use and interpretation of diagnostic tests into medical education.	Educational Institutions, Healthcare Providers	Increased accuracy and efficiency in AMR diagnosis.	Medium
Treatment and Care	- Strengthen ASPs: Enhance the implementation of antimicrobial stewardship programs across all healthcare facilities.	Hospitals, Clinics, Community Pharmacies	Optimized the use of available antimicrobials, reducing the inappropriate sale of antibiotics without a prescription especially Watch antibiotics through reducing their availability enhanced by targeted educational programs among community pharmacists and patients	Very High
	- Promote Evidence-Based Practices: Encourage the use of evidence-based guidelines for antibiotic use among healthcare providers.	Healthcare Providers, Policy Makers	Routine adoption of AWaRe antibiotic classification system and the WHO AWaRe book guidance by all healthcare workers to help meet UN GA target of 70% Access antibiotics	Very High
Cross-Cutting Issues	- Implement National Surveillance Systems: Develop comprehensive AMR surveillance systems to monitor and respond to AMR trends.	Government, Research Institutions	Better informed public health strategies and resource allocation based on accurate AMR data.	High
	- Policy and Regulation Enforcement: Ensure consistent enforcement of existing policies and develop new regulations as needed.	Policy Makers, Regulatory Bodies	Stronger regulatory compliance and more effective containment of AMR. This includes Regulatory Agencies focusing on priority antibiotics for registration	Very High
	- Public Health Campaigns: Launch extensive public health campaigns to raise awareness about AMR and promote responsible antibiotic use.	Government, Public Health Officials	Increased public and professional awareness of antibiotics and AMR leading to more judicious use of antimicrobial agents - especially with the growing influence of patients requesting antibiotics in Pakistan	Medium to High

(Continued)

Table 6 (Continued).

Theme	Recommendations	Target Stakeholders	Expected Outcomes	Priority Level
TB Specific	- Enhance TB Prevention Measures: Implement targeted interventions to reduce transmission and recurrence of TB, particularly drug-resistant strains.	Government, Healthcare Providers	Reduced transmission and improved management of TB, particularly drug-resistant forms.	High
	- Improve TB Diagnostic Tools and Strategies: Enhance the use of rapid diagnostic tests for early and accurate detection of TB and drug-resistant TB.	Healthcare Providers, Diagnostic Labs	Faster and more accurate diagnosis of TB, leading to timely and effective treatment interventions.	High
	- Optimize TB Treatment Protocols: Develop and disseminate updated guidelines for the treatment of all forms of drug-resistant TB.	Healthcare Providers, Policy Makers	Improved treatment outcomes and reduced TB mortality rates through effective use of treatment protocols.	High

We are aware our approach has several limitations that may influence the interpretation of our findings. Primarily, as a narrative review, we focused on synthesizing broad thematic areas within the published literature, which may have led to the exclusion of certain studies, particularly those not indexed in major databases. This methodological approach, while effective for mapping out the landscape of AMR research in Pakistan, might have missed nuanced details or emerging research in academic publications. The selection criteria also prioritized published studies, potentially overlooking insights from unpublished research and ongoing projects that could offer valuable perspectives on AMR strategies. Additionally, the geographical representation of AMR research within Pakistan might be skewed towards more accessible urban centers, thereby under representing rural and remote areas where AMR challenges may be greater. The review also relied on the availability and accuracy of reported data that may not capture the full spectrum of AMR-related initiatives, or the nuances of their implementation across different regions. Finally, the evolving nature of AMR and the continuous emergence of new resistance patterns, necessitate ongoing research, suggesting that our review, while extensive, represents a snapshot within a dynamic field of study. However, despite these limitations, we believe that our findings will be of interest to all the key stakeholders in Pakistan.

Conclusion

Overall, current research regarding research activities relating to AMR in Pakistan is primarily concentrated in major urban centers across a limited number of cities including Karachi, Islamabad, Lahore and Rawalpindi. This is a concern going forward. Alongside this, GHRA's research priority areas were addressed in only limited scope across various cities. This included the fact that studies focusing on prevention were conducted in Karachi, studies related to diagnosis were concentrated in Lahore, studies related to treatment and care took place in both Karachi and Lahore and studies related to cross-cutting and drug-resistant TB were carried out in Karachi. As a result, a number of gaps and potential activities were identified to address AMR in Pakistan going forward. These included addressing current poor IPC practices, absence or incomplete implementation of AMS activities alongside typically a lack of ASPs across sectors according to WHO AWaRe classification and guidance, lack of laboratory surveillance, educational gaps among all key stakeholder groups, poor diagnostic accuracy, lack of enhanced infrastructure, and the loss of TB treatment efficacy. To address these challenges, Pakistan's second National Action Plan emphasizes comprehensive IPC strategies, improved surveillance systems and ASPs, as well as educational campaigns aimed at all key stakeholder groups including physicians, community pharmacists and patients. Pakistan must prioritize its policies that support AMR sustainable practices and international collaboration. The pathway forward includes leveraging global partnerships to share knowledge, resources, and strategies, ensuring that Pakistan can effectively contribute to and benefit from the global efforts to mitigate AMR. We will be following this up in future research projects.

Author Contributions

All authors made a significant contribution to the work reported, whether that is in the conception, study design, execution, acquisition of data, analysis and interpretation, or in all these areas; took part in drafting, revising or critically reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

Funding

This research received no external funding.

Disclosure

The authors declare no relevant conflicts of interest.

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