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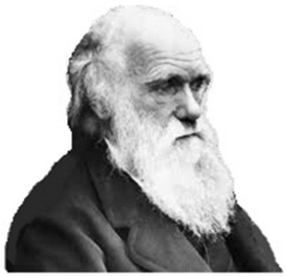

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Curbing Antimicrobial Resistance, for Better Development in the Health and Public Sector

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ABSTRACT

With 1.27 million deaths in 2019, antimicrobial resistance (AMR) has resulted in the deaths of more people than HIV/AIDS and malaria combined. AMR is listed among the top 10 global public health priorities by the WHO. The magnitude of the AMR burden is poorly prioritized in the public health sector, and its impact is difficult to accurately estimate due to a lack of sufficient data, especially in Africa. The misuse and overuse of antibiotics for various non-medical purposes is widely reported, especially in low-income developing countries, as one of the key contributors to AMR, yet little effort is put in place to curb the menace. Also, the clinical pipeline of new antimicrobials is scanty. This review explores the mechanism of AMR, the effects of AMR on health and the public sector with regard to morbidity, mortality, and economic losses, and the impact of COVID-19 on AMR. Recent potential therapies that are used to combat AMR, such as phage therapy, and how they could bring about development in the public health sector were discussed. Also, the impact of vaccines and preventive strategies to curb the spread of AMR in the public sector were highlighted.

***Keywords*:** Antibiotic resistance, public health, vaccines, phage therapy, COVID-19

1. INTRODUCTION

Antimicrobial resistance (AMR) can be defined as the ability of a microorganism to resist the effects of antimicrobial treatment, and sadly, the most prominent antimicrobial treatment antibiotics are rapidly losing their potency, thereby threatening the very stability of public health globally.

Resistance to antibiotics can occur either through mutation or acquisition[1]. Before the historic discovery of penicillin by Sir Alexander Fleming in 1928, humans died from various diseases that are curable today using first-line antibiotics. The discovery of antibiotics has revolutionized the practice of medicine over the years, saving the lives of millions, including postpartum mothers and their babies. However, humankind is fast backsliding into the pre-antibiotic era due to irrational use of antibiotics, non-compliance, and use of antibiotics for agricultural purposes, thereby resulting in the rapid evolution and spread of antibiotic-resistant strains.

In a recent review of the global burden of antimicrobial resistance (AMR) in 2019, about 4.95 million deaths were reported, of which 1.27 million were caused by antibacterial resistance (ABR). The Western region of sub-Saharan Africa has the highest mortality rate at 27.3 deaths per 100,000 deaths, making it a significant concern[3].

The impact of AMR burden is difficult to accurately estimate, especially in low and middle-income countries, due to the lack of sufficient data; however, some of the common indicators used to measure the impacts include the mortality rate, the economic cost on the patient and their family, and low productivity due to hospitalization. It is estimated that by 2050, the global economic burden of AMR will rise to between $300 billion and $1 trillion, and about 10 million deaths will be reported annually. Therefore, joint, proactive, and coordinated action must tackle this global threat to public health before it is too late.

The aim of this review work, therefore, is to bring to focus the mechanism of AMR, current efforts directed at combating AMR, strategies to curb AMR via the public sector, the impact of AMR in the health and public sectors, and the effect of the Coronavirus (COVID-19) pandemic on the fight against AMR.

2. ANTIMICROBIAL RESISTANCE MECHANISM

The most common classes of antibiotics being prescribed today exhibit either bacteriostatic or bactericidal effects by inhibiting bacterial DNA replication, disrupting the synthesis of peptidoglycan, lysis of the bacterial cell membrane, interrupting gene transcription and translation, as well as folate biosynthesis, as shown in Figure 1[5]. Decades of research prove that AMR occurs via intrinsic (naturally acquired) or extrinsic mechanisms. Intrinsic mechanism resistance occurs as a result of the pathogen's biology and survival instincts, while extrinsic resistance is acquired externally, either from one bacteria to another or from the environment. The use, misuse, and abuse of antibiotics by humans, especially for agricultural purposes (e.g fish farming), is one of the major contributing factors responsible for the spread of AMR [6]

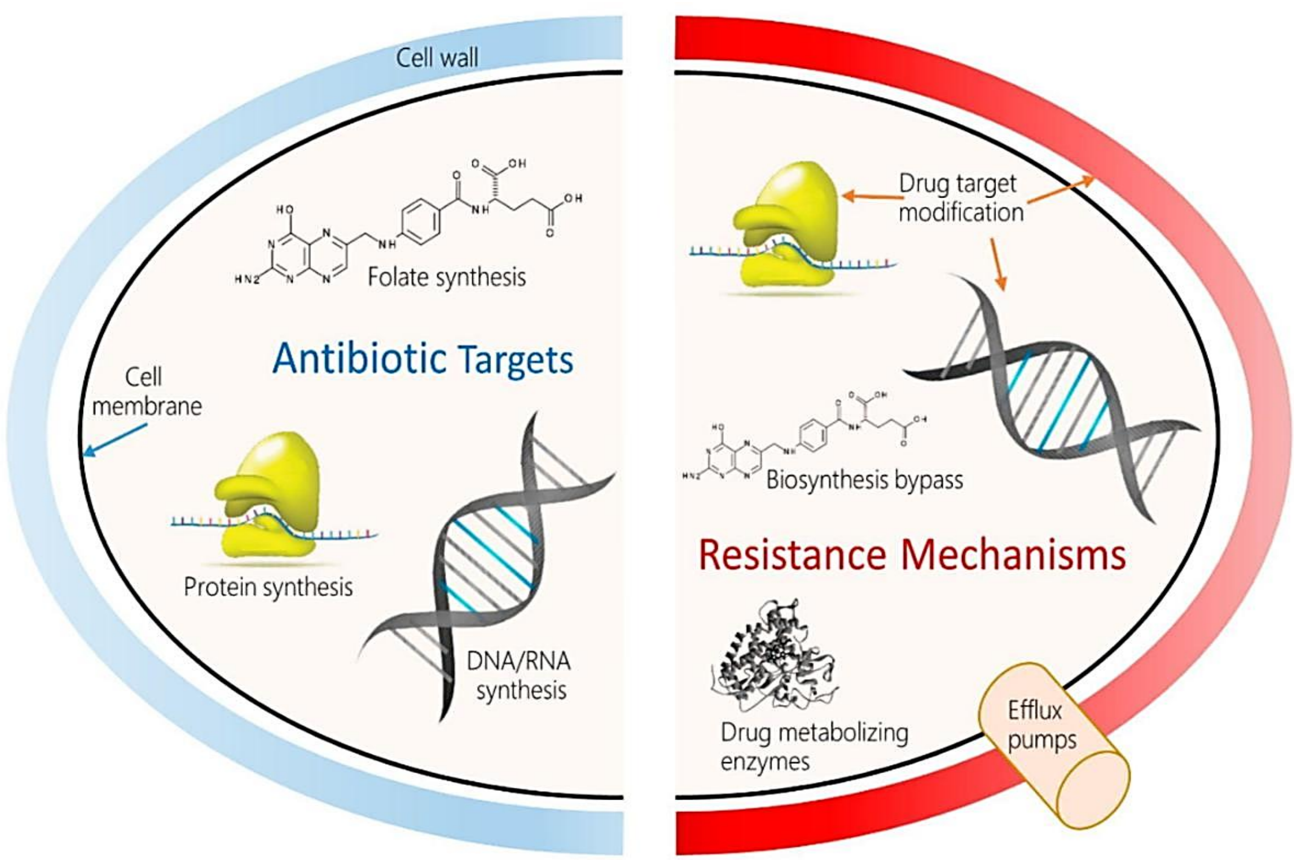

Figure 1. Left: The sites of action for the most common classes of antibiotics currently in use. Right: Mechanisms of resistance employed by resistant bacteria to evade the effects of antibiotics at their sites of action[5] (Pieter-Jan Van Camp *et al.*, 2020).

Intrinsic or naturally acquired resistance

Intrinsic resistance occurs when the target bacteria can survive the antimicrobial activities of a certain antibiotic due to its structure and possession of resistant genes. Internally acquired

AMR mechanisms can develop in several ways, depending on the active site of the antibiotic on the target bacteria. These mechanisms have been well researched and can be categorized broadly into four groups; restriction of antibiotic entry through the bacterial cell wall; which is exemplified by the resistance of Enterobacteriaceae to carbapenems, due to the reduced bacterial membrane permeability, activation of efflux pump systems to pump antibiotics out of the cell before antibiotic-target interactions occur, enzymatic degradation of the antibiotic, bacterial synthesis of -lactamases that degrade the -lactam class of antibiotics, and finally, antibiotic target site modification [6]

For instance, daptomycin is a potent antibiotic against Gram-positive bacteria but completely inactive against Gram-negative bacteria due to differences in the cell membranes between the Gram-positive and Gram-negative bacteria (Fig. 2). Recent studies into the different mechanisms of AMR have revealed several antibiotic resistance genes (ARG), or bacterial gene variants implicated in the phenotypic resistance to antibiotics such as fluoroquinolones, β- lactams and aminoglycosides[5]. The mechanism of action of many AMR genes or cellular pathways is now known. The important resistance genes include thioredoxin reductase (trxB), thioredoxin A (Trx A), SapC, DacA, FabI, and D-Ala-D-Ala carboxypeptidase [7]

Externally Acquired AMR mechanisms.

A susceptible bacteria can acquire resistance from the environment through an extrinsic resistance mechanism to survive an unfavorable environment that can threaten its survival. Usually, bacteria acquire a resistant gene from a resistant bacteria through:

- Horizontal gene transfer (from one bacteria to another)
- Recombination 8
- and Mobile gene transfer

Despite all we currently know about AMR, there are other complex mechanisms of resistance that we still do not know a lot about and, thus, simple association with phenotypic resistance would be impossible, especially when trying to make generalizations on Antibiotics Resistance Gene across multiple species or while researching and testing multiple antibiotics at once [7]

3. POTENTIAL THERAPY FOR ANTIMICROBIAL RESISTANCE

Since the discovery of the first antibiotic, AMR has been identified in bacteria. As new antibiotics are developed, they are seen with less concern. But currently, resistant bacteria strains are observed due to antibiotics' misuse, and there are no new antibiotics to counter these strains. However, looking at the adverse effects of AMR has necessitated the development of alternative strategies to combat the menace.

Phage therapy

Phage therapy in recent years has captivated a tremendous interest as one of the potential options for the treatment of antibiotic resistance. A phage infection in bacteria is established by injecting the viral genome into the bacterial cell. Phages overtake the host bacterial machinery. There are two life cycles involved: lysogenic or lytic infection.

The phages get assimilated into the bacterial chromosome and continue dwelling as prophages in the lysogenic cycle. Meanwhile, in the lytic cycle, phage particles are produced inside the bacterial host cell, and the enzymes (holin and endolysin) help with the release of mature phages [8]. A phage-delivered resistance eradication with a suitable antibiotic treatment (PRESA) strategy was developed by Liu et al. which resulted in a potent antimicrobial effect against *E.coli* and a decrease in the bacterial load. In contrast to the lytic phage strategy, PRESA shows continuous new inhibitory effects on resistant bacteria with no emergence of new mutational resistance [9].

Therefore, with PRESA, there could be the restoration of low-cost antibiotics, and it can be further developed as a reassuring therapy to control AMR. Additionally, Samir et al demonstrated that isolated and characterized lytic bacteriophage specific to methicillin-resistant *Staphylococcus aureus* (MRSA) displayed a wide host range against MRSA and can be commercially fit for treatment as lysate preparation in the future [10].

Phages are known to be valuable tools to curb infections caused by different microorganisms, which include multidrug-resistant (MDR) pathogens. MDR bacteria can obtain antimicrobial resistance through inherent mechanisms and attributes, such as the existence of an outer membrane or horizontal genetic transfer. [11] (Blasco et al., (2019) investigated the antimicrobial action of the combinatory therapy (novel lytic phages, Ab105-2phiΔCI, combined with antibiotics of the carbapenem class, meropenem, and imipenem) against a carbapenem-resistant strain of *Acinetobacter baumannii.* The result showed improved antimicrobial activity of the antibiotic and the phage such that the bacterium became sensitive with regards to the antibiotics and a reduction in the emergency degree of phage-resistant bacteria was observed [12].

In another study carried out in Taiwan, an aerosol formulation which contains bacteriophage in the ICU was used, which resulted in a significant reduction in new acquisitions of carbapenem-resistant [13]. In a related study, it was reported that *in situ* anti-*S. aureus* phage therapy combined with intravenous antibiotics and surgical debridement was employed as a restorative therapy for polymicrobial chronic osteitis of the pelvic bone allograft, which resulted in satisfactorily short-term results with respect to both the clinical microbiological levels. [14]

With regards to bacteria persistence through phage therapy, Zaldastanishvili et al., 2021, carried out a retrospective study on three patients that went through phage therapy at the Elivara Phage Therapy Center (EPTC) in Georgia.

The patients were among the few EPTC patients whose pathogens continued through phage therapy. Genome restriction-based Pulsed Field Gel Electrophoresis (PFGE) profiling of the strain before and after phage therapy was used to investigate the adaptation strategies that were utilized by these pathogens. In the cases of two patients with chronic infectious diseases related to *Pseudomonas aeruginosa* (lower respiratory tract infection) and *Klebsiella pneumoniae* (urinary tract infection), the adaptation strategy of the bacteria appeared to have resulted in the heterogeneity of infecting strains of the new species, while in the other patient case with chronic infection that were related to *Pseudomonas aeruginosa,* bacterial strains were identical before and after phage therapy, but are distinguishable in their phage susceptibility properties [15].

Certainly, phage therapy is a potential solution to antibiotic resistance, however, attention needs to be geared towards preventing phage resistance in general and also enhancing the capacity of phage therapy. A challenge ahead for phage therapy is the management of regulatory policies to ensure misuse and abuse are prevented.

Therefore, scientists and clinicians should engage regulatory bodies to see that this area is moved forward within a short time frame.

The Role of Vaccines

A vaccine works by exposing the immune system to a pathogen so that the immune system will identify and react to that pathogen, thereby averting the establishment of an infection or at least reducing severity of disease. Most vaccines also protect individuals who are not vaccinated by a process called "herd immunity." By preventing the spread of diseases, which leads to a decrease in the use of antibiotics, vaccines are important in decreasing the rate of AMR [16].

In a study conducted to observe the impact of influenza vaccination on the usage of antibiotics in the USA between 2010-2017, Klien et al. observed that increasing the influenza vaccination point by 10% resulted in a 6.5% decrease in antibiotic use. It was gathered that vaccination coverage resulted in a decreased prescription rate of antibiotics the most in the pediatric population and the elderly aged 65 and above [17]. Also, it is expected that the introduction of the typhoid conjugate vaccine (TCV) will cure 44% of typhoid, of which 35% are antibiotic resistant [18].

P. Buchy et al. highlighted that vaccines can help avert AMR by preventing diseases and the multiplication of bacteria, reducing the use of antibiotics due to fewer infections, preventing resistant strains from transpiring and transmitting, preventing misuse of antibiotics, and mechanisms of action less liable to activate resistance [19]. Many vaccines are in the stage of clinical development and will be used to prevent infections caused by the major AMR bacterial pathogens including *M. tuberculosis* [20], *S. typhi* [21], *S. aureus* [22], and pathogenic *E. coli* [23].

Vaccine hesitancy is a problem highly associated with the treatment of diseases.[24] interventions are needed to solve vaccine hesitancy among individuals. An internet-based intervention with vaccine information and social media components was conducted by Daley et al. 2018 which resulted in improved parents' attitudes towards vaccines [25].

The World Health Organization (WHO), in an effort to control AMR, has come up with a strategy to express the impact of vaccines on AMR, a technical inclusion in the immunization agenda 2030, and an action framework that identifies a vision for vaccines to fully contribute to the prevention and control of AMR by preventing infections and a significant reduction in the use of antimicrobials (WHO, 2021).

It usually takes between 10-20 years to develop a new vaccine, which is a long time. To tackle the crisis of AMR, pharmaceutical companies should change the process of vaccine development, which comprises up to date technologies and new vaccine platforms, and it would be important to facilitate clinical studies and change the synergy with regulatory bodies [27].

Development of Resistance Mechanism Inhibitors

Studies on the mechanisms of antibiotic resistance have aided the development of several resistance mechanism inhibitors, such as AMR gene silencers, which silence the AMR genes, e.g.: CRISPR-Cas system; ribosomal inhibitors, which bind with ribosomal subunits and alter the protein production so that the bacteria cannot fight by proteins; and efflux pump inhibitors as shown in figure 2 [6].

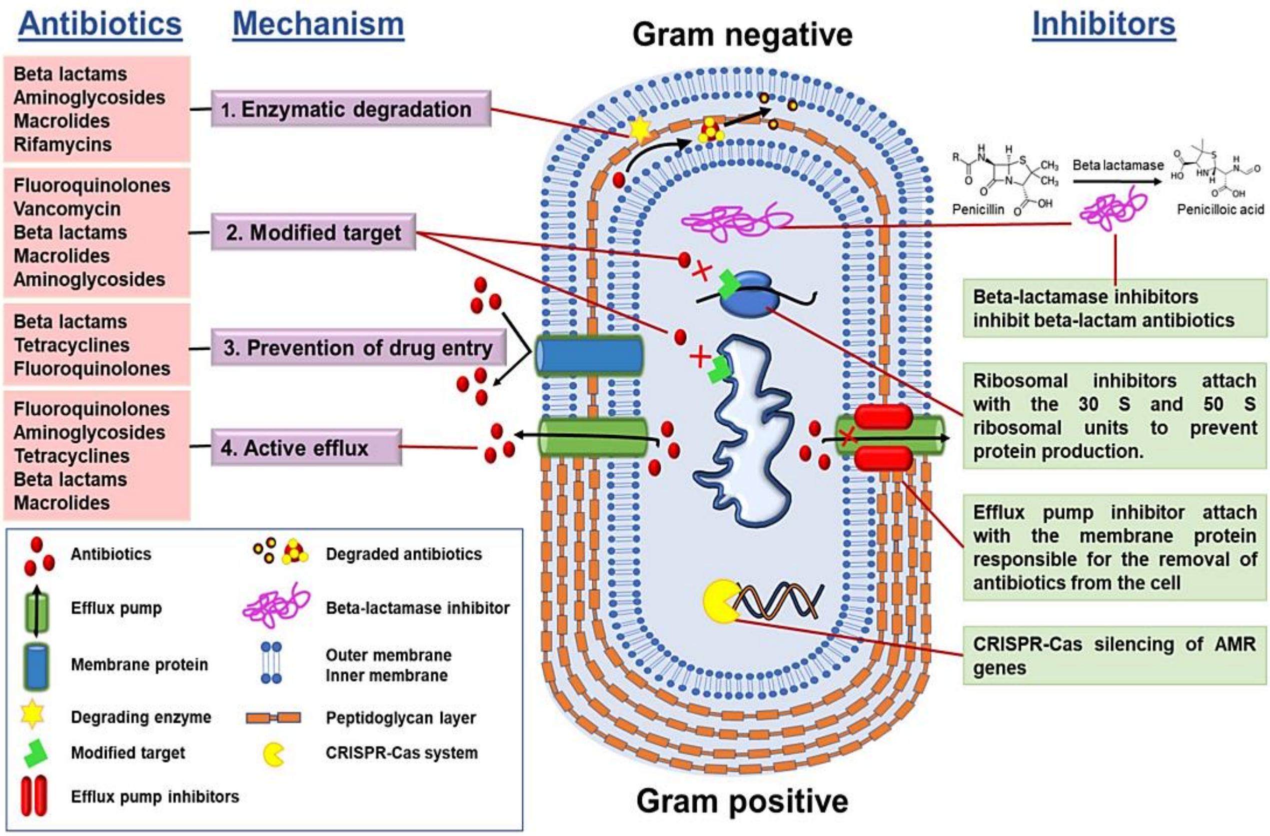

Figure 2. Classes of antibiotics, mode of action, and inhibitors (Murugaiyan, J. *et al.,* 2022).

4. STRATEGIES FOR COMBATING ANTIMICROBIAL RESISTANCE IN THE PUBLIC SECTOR

There is a need for coordinated action in combating AMR, like the One Health approach, which is a multisectoral and transdisciplinary approach that brings together different sectors and disciplines ranging from those in human, animal, plants, food, and the environment together to work in the creation and implementation of policies that will help for better outcomes in public health.

The best way to prevent AMR is to reduce the usage of antimicrobials and to do that, we must prevent infections that trigger the use of antimicrobials.

Beverly Merz highlighted some simple precautions that can help individuals from getting sick as a result of an infection.

She mentioned good hygiene as the primary way to prevent infections, which includes hand washing, covering the mouth and nose when sneezing or coughing, and washing and bandaging cuts; practicing good food safety techniques by rinsing meat, poultry, fish, fruits, and vegetables, and by thorough cooking of food and others; Vaccination to prevent illness, including vaccinations for children, adults, and pets; taking safety precautions before sexual intercourse and avoiding infections by using animal control [28].

Hand hygiene

The COVID-19 pandemic necessitated the implementation of strategies like social distancing, wearing masks, and hand hygiene to combat the disease. In a retrospective study conducted in Taiwan, a decrease in cases of influenza, enterovirus, and all-cause pneumonia was observed as a result of precautions taken to prevent COVID-19, suggesting that hand hygiene may have an impact not only on COVID-19 but also on other respiratory infectious diseases [29]. A problem with hand hygiene practice is inadequate compliance, whether among health professionals or the general public. In research carried out on some third-year medical students at the University of Bratislava, 32.9% of them had poor hand-washing hygiene performance. It was concluded that this was due to not being acquainted with the protocols of hand washing or forgetting to wash the hands. [30]

Practicing good food safety

To prevent foodborne infections, the practice of good food safety is paramount. The Centers for Disease Control and Prevention have highlighted four steps to ensuring food safety. These include: Keeping hands and surfaces clean; avoiding cross contamination by separating raw meat, poultry, and seafood from ready-to-eat food; cooking food at the right temperature, and prompt refrigeration. Amodio et al. conducted a knowledge, attitude, and practice survey, and their findings confirmed the link between foodborne diseases and food handling at home with unsafe food practices [31].

Precaution before sexual intercourse

There are concerns about the treatment of sexually transmitted infections (STIs) with the increase of antimicrobial resistance as seen particularly in *gonorrhea* [32-33] and *Mycoplasma genitalium* [34]. Researches has been carried out on the effectiveness of condom use in preventing STIs and HIV [35].

5. CURRENT IMPACTS OF ANTIMICROBIAL RESISTANCE ON HEALTH AND THE PUBLIC SECTOR

AMR has been declared as one of the top 10 global public health concerns threatening humans by the World Health Organization in 2019. [34] The Centers for Disease Control (CDC) in 2013 reported that at least 23,000 people died out of 2 million people that got an antimicrobial-resistant infection. In recent times, more than 2.8 million antimicrobial-resistant infections have been reported in the United States alone and this results in over 35,000 deaths annually. [35]

An extensive evaluation of the global burden of AMR was provided by a study published in the Lancet. In the study, it was statistically forecated that there were an estimated 4.5 million deaths attributable to AMR in 2019, with 1.27 million deaths connected to bacterial AMR alone. The highest all-age death rate connected to resistance was estimated in Western Saharan Africa, at 27.3 deaths per 100,000, and the lowest in Australia, at 6.5 deaths per 100,000. Lower respiratory infections are identified as the most burdensome infection syndrome, with more than 1.5 million deaths attributable to resistance in 2019. Furthermore, *E.coli* is identified as the top six leading pathogens associated with resistance, others include *Staphylococcus aureus*, *Klebsiella pneumoniae, Streptococcus pneumoniae, Acinetobacter baumannii,* and *Pseudomonas aeruginosa* [35]. The UN ad hoc interagency coordinating on AMR warns that unless an action is taken, drug-resistant diseases could cause damage to the economy, similar to the 2008-2009 financial crisis by the year 2050. Also, by 2030, AMR could force about 24 million people into extreme poverty [36]. AMR is indeed a serious threat to humanity. It is beneficial for everyone to play a role in combating this menace.

6. COVID-19 AND THE IMPACT ON ANTIMICROBIAL RESISTANCE

In 2020, the world was hit with the Severe Acute Respiratory Syndrome Coronavirus 2 (SARS COV 2) pandemic, and the impact on AMR can be best described as two-fold; an increase in the use, misuse, and overuse of antibiotics primarily due to irrational prescription, irrational counter dispensing and an increase in the reported incidents of Multidrug-Resistant Organisms (MDROs) implicated infections [37]

Many countries, particularly the low and middle-income nations, were already plagued with issues like over-the-counter dispensing of antibiotics without prescriptions, antibiotic misuse and overuse, lack of access to prescribers, and the prevalence of counterfeit medicines, which are all factors increasing AMR, [38] but with the highly infectious nature of the SARS COV 2 virus, the initial lack of rapid testing tools to aid prompt detection, and in a bid to reduce the soaring mortality rate due to the virus and manage its upper respiratory symptoms, many prescribers opted to start management with antibiotics, with some reports of nearly 80% of hospitalized COVID-19 patients administered antibiotics [39]. Azithromycin (18.1%) was one of the first-line agents said to show promise in clinical trials for the treatment of the coronavirus disease (COVID-19) at the onset of the pandemic and it was shown to have been administered [40] as were ceftriaxone (24.6%) and moxifloxacin (64.5%) according to a study involving 138 hospitalized patients in Wuhan, China [41].

Also, in the wake of the COVID-19 pandemic, several studies have reported an increase in the number of infections involving MDROs including carbapenem-resistant Klebsiella pneumonia, Panechinocandin-resistant *Candida glabrata,* Multi-triazole-resistant *Aspergillus fumigatus,* multidrug-resistant *A. baumannii*, *Methicillin-Resistant Staphylococcus aureus* (MRSA), multidrug-resistant *Candida auris*, extended-spectrum β-lactamase (ESBL)-producing *K. pneumoniae, S. maltophilia, Burkholderia cepacia,* and *Pseudomonas aeruginosa*. [41-49]. Some of the causes of the spike in MDROs implicated infections were patients requiring prolonged hospitalization, longer contact hours of healthcare workers with infected patients, higher susceptibility to infections due to the COVID-19 virus, etc. [37]

Although in the short term, the use of antibiotics in the management of COVID-19 symptoms may have resulted in a lowering of the mortality rate due to SARS COV-2 infection, with results being largely inconclusive at this time, it can be estimated that its long-term effects may include worsening AMR and an increase in mortality due to superbugs and MDRs which might mean that we might reach the predicted 10 million deaths per year much earlier than the year 2050.

A global survey that queried the impact of COVID-19 on Infection and Control Practices, amongst others, reported a positive correlation to combat AMR(14). Hand hygiene, the use of Personal Protective Equipment (PPE), and social distancing were some measures that became mainstream due to COVID-19 that helped reduce infection spread and subsequently reduced the use of antibiotics. The same survey also reported a marked increase in antibiotic consumption which negatively impacts the fight against AMR [50]. It is advisable, however, that even though some evidence exists to show the impact of COVID-19 across AMR surveillance, prevention, and control, most of the evidence may be biased and should be cited with caution[50]

7. CONCLUSION

The menace of antimicrobial resistance is fast spreading across the globe, and if immediate and well-coordinated action is not taken, the world is about to witness a global pandemic, one worse than the recent COVID-19 outbreak. One major problem identified by this study is the lack of reliable data and efficient tracking systems to monitor the trend of AMR in middle and low-income countries such as Africa. Without reliable data, it is difficult to estimate, plan, and effectively curb the spread of AMR. In addition, Government agencies and other policymakers rely heavily on such data to come up with guidelines and policies aimed at fighting AMR and curtailing the spread. It is therefore imperative that scientists, researchers, and all care providers document accurately every case of resistance and also come up with more effective strategies to combat antibiotic resistance. Some of the strategies highlighted in this review such as phage therapy and vaccination could help push the impending doom day further into the future buying mankind another century or more.

The health of every nation remains the collective responsibility of the Government and her citizens hence, governments across all levels should ensure that the public is well sensitized to adopt safe and hygienic practices such as regular handwashing, and use of sanitizers, as was employed during the recent COVID-19 pandemic. Since no Country exists independent of another, international trade and migration facilitate the spread of microbes including resistant genes from one country to another at an unprecedented rate. Therefore, there is a need for international collaboration, resource sharing, and funding for developing regions if we want to achieve the UN 2030 third sustainable development goal (SDG-3) of good health and well-being for all and to win the fight against AMR.

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References

[1] Samreena, Iqbal Ahmada, Hesham A. Malak b, Hussein H. Abulreeshb. Environmental antimicrobial resistance and its drivers: a potential threat to public health. *Journal of Global Antimicrobial Resistance* 27 (2021) 101–11

[2] Sameer Dhingra, Nor Azlina A. Rahman, Ed Peile, Motiur Rahman, Massimo Sartelli, Mohamed Azmi Hassali, Tariqul Islam, Salequl Islam, and Mainul Haque Microbial Resistance Movements: An Overview of Global Public Health Threats Posed by

Antimicrobial Resistance, and How Best to Counter Frontiers in Public Health (2020) | Volume 8 | Article 535668

[3] Dr. Mohsen Naghavi, Christopher J L Murray, Kevin Shunji Ikuta, Fablina Sharara, Lucien Swetschinski, Gisela Robles Aguilar, Authia Gray, Chieh Han, Catherine Bisignano, Puja Rao, Eve Wool, Sarah C Johnson, Annie J Browne, Michael Give Chipeta, Frederick Fell, Sean Hackett, et al., Global burden of bacterial antimicrobial resistance in 2019: a systematic analysis. *Lancet* 2022; 399: 629–55

[4] Thiruchelvi Pulingam, Thaigarajan Parumasivam, Amirah Mohd Gazzali, Azlinah Mohd Sulaiman, Jiun Yee Chee, Manoj Lakshmanan, Chai Fung Chin, Kumar Sudesh Antimicrobial resistance: Prevalence, economic burden, mechanisms of resistance and strategies to overcome. *European Journal of Pharmaceutical Sciences* 170 (2022) 106103

[5] Pieter-Jan Van Camp, David B. Haslam, and Aleksey Porollo. Bioinformatics Approaches to the Understanding of Molecular Mechanisms in Antimicrobial Resistance. *Int. J. Mol. Sci.* 2020, 21, 1363; doi:10.3390/ijms21041363

[6] Murugaiyan, J.; Kumar, P.A.; Rao, G.S.; Iskandar, K.; Hawser, S.; Hays, J.P.; Mohsen, Y.; Adukkadukkam, S.; Awuah, W. A.; Jose, R.A.M.; et al. Progress in Alternative Strategies to Combat Antimicrobial Resistance: Focus on Antibiotics. *Antibiotics* 2022, 11, 200. https://doi.org/10.3390/antibiotics11020200

[7] Liu, A., Tran, L., Becket, E., Lee, K., Chinn, L., Park, E., Tran, K., and Miller, J.H., 2010. Antibiotic Sensitivity Profiles Determined with an Escherichia coli Gene Knockout Collection: Generating an Antibiotic Bar Code. *Antimicrobial Agents and Chemotherapy*, 54 (4), 1393–1403

[8] Azam A.H, Tan XE, Veeranrayanam S, Kiga K, Cup L. Bacteriophage Technology and Modern medicine. *Antibiotics.* 2020,10,999.

[9] Liu H, Li H, Liang Y, Du X, Yang C, Yang L, Xie L, Zhao R, Tong Y, Qui S, Song H. Phage delivered sensitization with subsequent antibiotic treatment reveals sustained effect against antimicrobial resistant bacteria. *Theranosties* 2021.10(4):6310-6321

[10] Samir S, El-Far A, Okasha H, Raina M, Samir F, and Nasr S. Isolation and Characterization of lytic bacteriophages from sewage at an Egyptian tertiary care hospital against methicillin-resistant Staphylococcus aureus clinical isolates. *Saudi Journal for Biological Sciences* 2022. 29(5): 3097-3106

[11] Blasco, L., Ambroa, A., Lopez, M., Fernandez-Garcia, L., Bleriot, I., Trastoy, R., Ramos-Vivas, J., Coenye, T., Fernandez-Cuenca, F., Vila, J., Martinez-Martinez, L., Rodriguez-Baño, J., Pascual, A., Cisneros, J. M., Pachon, J., Bou, G., & Tomas, M. (2019). Combined Use of the Ab105-2φΔCI Lytic Mutant Phage and Different Antibiotics in Clinical Isolates of Multi-Resistant Acinetobacter baumannii. *Microorganisms* Vol. 7, Issue 11, p. 556. https://doi.org/10.3390/microorganisms7110556

[12] Martins W.M, Toleman M.A, Gales A.C. Clinical utilization of bacteriophages: a new perspective to combat the antimicrobial resistance in Brazil. *BRAZ J INFECT DIS.* 2020, 24(3): 239-246

[13] Zaldastanishvili E, Leshkasheli L, Dadiania M, Nadareishvili L, Askilashvili L, Kvatadze N, Goderadzishvili M, Kutateladze M, and Balarjishvili N. Phage therapy experience at the eliava phage therapy center: Three cases of Bacterial persistence. *Viruses.* 2021, 13, 1901. https://doi.org/10.3390/v131010901

[14] Jansen KU, Knirsch C and Anderson A.S. The role of vaccines in preventing bacterial antimicrobial resistance. *Nature Medicine* 2018. Vol. 24

[15] Klien EY, Schuella E, Tseng KK,Morgan DJ, Laximinarayan R and Nandi A. The impact of influenza vaccination on antibiotic use in the United States, 2010-2017. *OFID.* 2020. doi:10.1093/ofid/ofaa223

[16] Kaufold S, Yaesoubi R and Pitzer VE. Predicting the impact of typhoid conjugate vaccine on antimicrobial resistance. *Clinical Infectious Diseases* 2019. 68(2): S96-S104

[17] Buchy P, Ascioglu S, Buisoon Y, Data S, Nissen M, Tambyah P.A and Vong S. Impact of vaccines on Antimicrobial resistance. *International Journal of Infectious Diseases.* 2020. 188-196

[18] Schrager LK, Vekemens J, Drager N, Lewinsohn DM and Olesen OF. The status of tuberculosis vaccine development. *The Lancet Infectious Diseases* 2020. 20(3): 28-37

[19] Syed AK, Saluja T, Cho H, Haiao A, Shaikh H, Wartel TA,Mogasale V, Lynch J, Kim JH, Ecler J and Sahastrabuddhe S. Review on the recent advances on typhoid vaccine development and challenges ahead. *Clinical Infectious Diseases* 2020. 7(2): S141-150

[20] Clegg J, Soldaini E, Mc Loghlin RM, Rittenhouse S, Bagnoli F and Phogat's. Staphylococcus aureus vaccine research and development. *Front Immunol* 2021. 12: 705360

[21] Kowarik M, Wether M, Haeptle MA, Braun M, Steffen M, Stefan K, Ravenscroft N, De Benedetto G, Z Uppiger M, Sirena D, Cesutti P and Wacker M. The development and characterization of an E.coli 025B bioconjugate vaccine. *Glycoconj J.* 2021. 38(4): 421-435

[22] Dror AA, Eisenbach N, Taiber S, Morozor NG, Mizrachi M, Zigron A, Srouji S, and Sela E. Vaccine hesitancy: the next challenge in the fight against COVID-19. *European Journal of Epidemiology* 2020. 35: 775-779

[23] Daley MF, Narwaney KJ, Shoup JO, Wagner NM, and Glanz JM. Addressing patients vaccine concerns: A Randomized trial of a social media intervention. *AM J Prev Med.* 2018. 55(1): 44-54

[24] https://www.who.int/publications/m/item/leveraging-vaccines-to-reduce-antibiotic-use-and-prevent-antimicrobial-resistance

[25] Micoli F, Bagnoli F, Rappuoli R, Serruto D. The role of vaccines in combating antimicrobial resistance. *Nature Reviews/Microbiology* 2021. Volume 19, 287

[26] Merz Beverly. How to prevent infections. Harvard Health publishing. 2020. https://www.health.harvard.edu/staying-healthy/how-to-prevent-infections

[27] Chiu N, Chi H, Tai Y, Peng C, Tseng C, Chen C, Tan BF and Lin C. Impact of wearing masks, hand hygiene, and social distancing on influenza, enterovirus and all-cause

pneumonia during the coronavirus pandemic: Retrospective National Epidemiological Surveillance Study. *Journal of Medical Internet Research* 2020. 22(8)

[28] Novak M, Breznicky J, Kompanikova J, Hudecková H. Impact of hand hygiene knowledge on hand hygiene compliance. *Med Glass.* 2020. 17(1): 194-199

[29] Amodio E, Calamusa G, Tiralongo S, Lombardo F, and Genovese D. A survey to evaluate knowledge, attitude, and practices associated with risk of foodborne infections in a sample of Silicon general population. *AIMS Public Health* 2022. 9(3): 458-470

[30] Whiley DM, Jennison A, Pearson J, Lahra MM. Genetic characterization of Neisseria gonorrhoeae resistant to both ceftriaxone and azithromycin. *The Lancet Infectious Diseases.* 2018. 18(7): 717-718

[31] Martin I, Sawatzky P, Allen V, Lefebvre B, Hoang LMN, Naidu P, Minion J, Van Caeseele P, Halvare D, Gad RR, Zahariadis G, Corriveau A, German G, Tomas K, Mulvey MR. Multidrug-resistant and extensively drug-resistant *Neisseria gonorrhoeae* in Canada, 2012-2016. *Can Commun Dis Rep* 2019 Feb 7; 45(2-3): 45-53. doi: 10.14745/ccdr.v45i23a01

[32] Hamasuna R, Hanzawa H, Moritomo A, Matsumoto M, Hisami A, Tomisaki I, Akasaka T, Naohiro F, and Jensen JS.Analysis of fluoroquinolone resistance using mic determination and homology modeling of parc of contemporary Mycoplasma genitalium strains. *Journal of Infection and Chemotherapy* 2022. 28, 3, 377-383

[33] Wiyeh AB, Mome RKB, Mahasha PW, Kongnyuy EJ and Wiysonge CS. Effectiveness of the female condom in preventing HIV and sexually transmitted infections: a systematic review and meta-analysis. *BMC Public Health.* 2020. 30(319)

[34] Stover J and Tent Y. The impact of condom use on the HIV epidemic. *Gates Open Research.* 2020. 5: 911

[35] Chih-Cheng Lai, Shey-Ying Chen, Wen-Chien Ko, Po-Ren Hsueh. Increased antimicrobial resistance during the COVID-19 pandemic. *Int J Antimicrob Agents.* 2021 Apr; 57(4): 106324, 2021 Mar 19. doi: 10.1016/j.ijantimicag.2021.106324

[36] Hyacinth O. U. The interrelationships between antimicrobial resistance, COVID-19, past, and future pandemics. *J Infect Public Health* 2021 Jan;14(1):53-60. doi: 10.1016/j.jiph.2020.10.018. Epub 2020 Dec 9

[37] Xu X-W, Wu X-X, Jiang X-G, Xu K-J, Ying L-J, Ma C-L, et al. Clinical findings in a group of patients infected with the 2019 novel coronavirus (SARS-Cov-2) outside of Wuhan, China: retrospective case series. *BMJ* 2020; 368: m606, http://dx.doi.org/10.1136/bmj.m606

[38] Gautret, P. et al. Hydroxychloroquine and azithromycin as a treatment of COVID-19: results of an open-label non-randomized clinical trial. *Int J Antimicrob Agents.* 2020 Jul; 20. doi: 10.1016/j.ijantimicag.2020.105949

[39] Chen N, Zhou M, Dong X, Qu J, Gong F, Han Y, et al. Epidemiological and clinical characteristics of 99 cases of 2019 novel coronavirus pneumonia in Wuhan, China: a descriptive study. *Lancet* 2020; 395: 507–13

[40] Tiri B, Sensi E, Marsiliani V, Cantarini M, Priante G, Vernelli C, et al. Antimicrobial stewardship program, COVID-19, and infection control: spread of carbapenem-resistant Klebsiella pneumoniae colonization in ICU COVID-19 patients. What did not work? *J Clin Med* 2020; 9: E2744

[41] Li J, Wang J, Yang Y, Cai P, Cao J, Cai X, et al. Etiology and antimicrobial resistance of secondary bacterial infections in patients hospitalized with COVID-19 in Wuhan, China: a retrospective analysis. *Antimicrob Resist Infect Control* 2020; 9: 153

[42] Contou D, Claudinon A, Pajot O, Micaëlo M, Longuet Flandre P, Dubert M, et al. Bacterial and viral co-infections in patients with severe SARS-CoV-2 pneumonia admitted to a French ICU. *Ann Intensive Care* 2020; 10: 119

[43] Sharifipour E, Shams S, Esmkhani M, Khodadadi J, Fotouhi-Ardakani R, Koohpaei A, et al. Evaluation of bacterial co-infections of the respiratory tract in COVID-19 patients admitted to ICU. *BMC Infect Dis* 2020; 20: 646

[44] Fu Y, Yang Q, Xu M, Kong H, Chen H, Fu Y, et al. Secondary bacterial infections in critically ill patients with coronavirus disease 2019. *Open Forum Infect Dis* 2020; 7: ofaa220

[45] Nori P, Szymczak W, Puius Y, Sharma A, Cowman K, Gialanella P, et al. Emerging co-pathogens: New Delhi metallo-β-lactamase producing Enterobacteriaceae infections in New York City COVID-19 patients. *Int J Antimicrob Agents* 2020: 106179

[46] Farfour E, Lecuru M, Dortet L, Guen ML, Cerf C, KarnycheffF, et al. Carbapenemase-producing Enterobacterales outbreak: another dark side of COVID-19. *Am J Infect Control* 2020; 48: 15336. DOI: 10.1016/j.ajic.2020.09.015

[47] Posteraro B, Torelli R, Vella A, Leone PM, De Angelis G., De Carolis E, et al. Panechinocandin-resistant Candida glabrata bloodstream infection complicating COVID-19: a fatal case report. *J Fungi* (Basel) 2020; 6: 163

[48] Sara Tomczyk, Angelina Taylor, Allison Brown, et al. Impact of the COVID-19 pandemic on the surveillance, prevention, and control of antimicrobial resistance: a global survey. *J Antimicrob Chemother* 2021; 76: 3045–3058. doi:10.1093/jac/dkab300

[49] Janeway CA Jr, Travers P, Walport M, et al. Immunobiology: The Immune System in Health and Disease. 5th edition. New York: Garland Science; 2001. Manipulating the immune response to fight infection. Available from: https://www.ncbi.nlm.nih.gov/books/NBK27131/