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AMR status in Uttarakhand

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ABSTRACT: Antimicrobial resistances (AMR) of bacteria are the burning issues globally which are closely related to public health and wealth. Since Fleming's great discovery of Penicillin to till now antibiotics of clinical therapy is a great advance in human and animal medicine. Due to haphazard and unregulated utilization of antibiotics in the field of poultry, dairy farming, agriculture, fish farming and clinical therapies results into development of antibiotic resistome. The overall population and high infection load led to higher total consumption. Hence, India is still the largest consumer of antibiotics for human health. WHO, FAO and OIE have agreed that antibiotic resistance- "One Health problem" that involved humans, animals and the environment. "One Health approach" is needed to meet the challenges of the rapidly increasing supply and demand of our global food system and to attain optimal health for people, animals and our environment. In Uttarakhand, more investigations are needed for searching the alternate source to antibiotics as well as containing the unregulated use of antibiotics in human, animal and agriculture. NAP-AMR has been formulated in our country but only one state i.e., Kerala has adopted it. There is necessary that all states and union territories must adopt and execute National Action Plan on AMR for the wellbeing of mankind.

Key words: Antimicrobial resistances (AMR), antimicrobial resistance genes (args), antibiotics, Campylobacter, NAP-AMR, one health approach, salmonella

India is a high consumer of antibiotics and antimicrobial resistant (AMR) bacteria are a major public health concern. Antimicrobial resistance (AMR) is one of the latest and burning issues to galvanise political and financial investment as an emerging global public health threat. The introduction of antibiotics to clinical therapy is a great advance in human medicine, since Fleming's great discovery. However, the successful use of antibiotics is gradually compromised by the development of antibiotic resistance. Currently, the problem of antibiotic resistance is receiving unprecedented attention all over the world. Now AMR has become National as well as world issue. The global collection of resistance genes in clinical and environmental samples is the antibiotic "resistome" and is subject to the selective pressure of human activity. The origin of many modern resistance genes in pathogens is likely environmental bacteria including antibiotic producing organisms that have existed for millennia. Most antibiotics in medical or agricultural use are derived from or produced by a group of soil-dwelling bacteria called the Actinomycetes (the most notable genus for

antibiotic production being *Streptomyces*). These organisms are prolific producers of specialized metabolites (so-called "natural products"), including the antibiotics streptomycin, tetracycline, chloramphenicol, erythromycin and vancomycin. The antibiotic resistance has a long history that is comparable to the discovery of antibiotics and the resistance even appeared prior to the clinical use of the drugs (Yongfei *et al.*, 2017). Antibiotic resistance is One Health problem that involved humans, animals and the environment (Robinson *et al.*, 2016). The One Health approach is defined as the collaborative effort of multiple disciplines-working locally, nationally, and globally – to attain optimal health for people, animals and our environment. A One Health approach is needed to meet the challenges of the rapidly increasing supply and demand of our global food system (AVMA, 2008). Antimicrobial resistance can spread from food animals to humans through a number of routes. Direct contact with livestock is an important route of exposure for agricultural workers, but the most likely sources of exposure for the general population are thought to be resistant bacteria in livestock-

derived food products and animal waste, which is used as a fertilizer of crops and can contaminate water supplies. Yet, antimicrobials are used in very large quantities in the human population and bacteria carrying resistance genes can spread directly within human populations or indirectly through sewage contamination of food, water or the wider environment. It is not clear how important the livestock-derived routes are compared with the human-derived routes (Woolhouse and Ward, 2013). It is also found that the environment is a nearly boundless reservoir of antibiotic resistance has resulted in several studies that seek to estimate the risk of gene transfer to pathogen (Manaia, 2016). The evidence is now clear that the environment is the single largest source and reservoir of antibiotic resistance. Soil, aquatic, atmospheric, animal-associated and built ecosystems are home to microbes that harbor antibiotic resistant elements and the means to mobilize them. (Surette and Wright, 2017). The investigation of novel approaches for tackling the antimicrobial resistance crisis must be part of any global response to this problem. Currently, every year, 700,000 patients die globally due to antimicrobial resistance (AMR). It has been estimated that this death toll will increase to 10 million by 2050, which would lead to a reduction of gross domestic product (GDP) by at least 2.5% (Gravitz, 2012). The fight against AMR is constant, and the discovery of new antibiotics is critical. Together, the facts and statistics raise an important question: is the time of antibiotics up? Although antibiotics have served humanity well for the last 70 years or so, the ability of bacteria to quickly evolve has made it imperative to look for other options. As more antibiotics are rendered ineffective by drug resistance bacteria, focus must be shifted toward alternative therapies for treating infection. Although several alternatives already exist in nature, the challenge is to implement them in clinical use. The main aim of this review is to know the AMR issues and one health problems for survival of human and animals in safe environment.

Antimicrobials and their Mechanisms

Antimicrobials – including antibiotics, antivirals,

antifungals and antiparasitics – are medicines used to prevent and treat infections in humans, animals and plants. Antimicrobial resistance (AMR) occurs when bacteria, viruses, fungi and parasites change overtime and no longer respond to medicines making infections harder to treat and increasing the risk of disease spread, severe illness and death. The emergence and spread of drug-resistant pathogens that have acquired new resistance mechanisms, leading to antimicrobial resistance, continues to threaten our ability to treat common infections. Especially, alarming is the rapid global spread of multi and pan-resistant bacteria (superbugs that cause infections that are not treatable with existing antimicrobial medicines such as antibiotics (WHO, 2021).

Antibiotic resistance poses a tremendous threat to human and animal health. To overcome this problem, it is essential to know the mechanism of antibiotic resistance in antibiotic-producing and pathogenic bacteria. Main mechanisms of antibiotic resistance are Antibiotic modification or degradation, Antibiotic Efflux, Antibiotic Sequestration, Target Modification, Bypass and Protection Mechanism (Van Boeckel *et al.*, 2014).

In case of aminoglycosides, chloramphenicol, and beta lactam antibiotic modification is commonly used strategy to make its ineffective. The aminoglycoside antibiotics are biosynthesized by *Actinobacteria*, so these prokaryotes must protect themselves against attacks by their own biosynthetic products. A large number of aminoglycosides modification enzymes (AMEs) including N-acetyl transferases (AAC), O-Phosphotransferases (APH) and O-Adenyl-transferase (ANT) that acetylate, Phosphorylate or adenylate the aminoglycoside antibiotic in producer organism. There is structural and sequence similarities between AMEs of producer and cellular metabolic enzymes. Modification enzymes may be co-opted from housekeeping enzyme. In contrast to modification, beta-lactam antibiotic is normally degraded by β -lactamase hydrolysing enzyme. This enzyme widespread among *Streptomyces* and similar enzymes are found in pathogenic and non pathogenic bacteria, they all constitute the ‘ β -lactamase superfamily’ (Ogawara, 2015).

Efflux of antibiotic is another commonly used mechanism for self protection, although it usually occurs in conjunction with other mechanism. The best studied example of antibiotic efflux among producer is *Streptomyces peucetius*, which produced two closely related anti-cancer antibiotic, daunorubicin (Dnr) and doxorubicin (Dox). These two antibiotics joined with DNA preventing further round of replication. Efflux of these antibiotics occurs by ABC family transporter (ATP binding cassette). Another example is OtrC found in oxytetracycline producer *Streptomyces rimosus*. It exhibit multidrug specificity. Self resistance in *S. rimosus* is conferred by two efflux protein: OtrB located in the biosynthesis cluster and OtrC located outside the cluster. OtrB belong to major facilitator superfamily (MFS) of transport family.

Sequestration involves the function of drug-binding proteins, which prevent the antibiotic from reaching its target. In producers of the bleomycin family of antibiotics, the primary mechanism of resistance involves sequestration of the metal-bound or the metal-free antibiotic by binding proteins TlmA, BlmA, and ZbmA in *S. hindustanus*, *S. verticillus*, and *Streptomyces flavoviridis*, respectively (Rudolf *et al.*, 2015). Each bleomycin-family producer member has one or more genes related to ABC transporters in their biosynthesis clusters which may be used to remove the antibiotics bound to binding proteins. The three self-resistance-related genes, *blmA*, *blmB* and *orf 7* encoding the bleomycin-binding protein, the bleomycin acetyltransferase, and ABC transporter respectively, are present at the end of the cluster. The gene *orf29*, which is located at another end of the cluster, may also be involved in the self-resistance by transporting the drug (Pozzi *et al.*, 2016).

Target modification acts as a self-resistance mechanism against several classes of antibiotics, including β -lactams, glycopeptides, macrolides, lincosamides and streptogramins (MLS) and aminoglycosides. The β -lactam antibiotic has a similar structure to peptidoglycan precursor substrates (PBP), thus allowing the antibiotic to associate and cause acylation of the active site serine

resulting in its inhibition. The producer *Streptomyces* species, despite of being Gram-positive are highly resistant to penicillins, which is due to either overproduction of PBPs or synthesis of low-affinity PBPs. Analysis of the biosynthesis cluster of β -lactam producing bacteria showed that they contained gene for PBPs suggesting their role in self-resistance (Ogawara, 2015).

AMR in India and Uttarakhand state

Antimicrobial resistance (AMR) is a growing public health problem globally. In India, there are several factors responsible for the emergence of AMR such as –unregulated access to antibiotics, including over the-counter sale as well as sale without prescription or with invalid prescription; and perverse financial incentives for providers to prescribe antibiotics, often driven by patient demand and expectations, injudicious use of antibiotics for treatment of diseases in man and animals, use of antibiotics as a growth promoter in animal and poultry premixed feed (Laxminarayan *et al.*, 2016). The issue of AMR drew global attention through the controversy of the nomenclature of the New Delhi Metallo-beta-lactamase-1 (NDM-1), and has since received a lot of attention as a major public health concern. All the factors like the high burden of bacterial infections, poor sanitary and hygiene conditions and the increasing proportion of intensive animal farming, especially in small holder settings with minimal oversight and quality control makes emergent AMR a cause of special concern in the country. The problem is no longer restricted to the clinical or hospital setting and is now emerging as a wider concern involving the animal/food/livestock sector, as well as environmental contamination as a source for the spread of resistance genes and antibiotic residues promoting selection pressure (WHO, 2016). In India's report of National action plan on Antimicrobial Resistance 2017-21 (NAP-AMR) illustrated that world consumption of antibiotic was 12.9×10^9 units in 2010. India is the largest consumer of antibiotics for human health. Although the per capita consumption of antibiotics in India (10.7 units per capita) is still lower than that seen in many other countries (e.g. 22 units per

capita in USA), the overall population and infection load led to higher total consumption, hence, India is still the largest consumer of antibiotics for human health. Almost 23% of the increase in the retail antibiotics sale in the BRICS countries was attributable to India. With respect to consumption of antimicrobials in food animals, the global consumption was estimated to be 63,151 ($\pm 1,560$) units in 2010; India accounts for 3% of the global consumption and is the fourth highest in the world, behind China (23%), the United States (13%) and Brazil (9%). The consumption of antimicrobials in the food animals sector in India is expected to double by 2030. The absence of stringently framed and implemented regulatory frameworks to limit the use of antimicrobials in livestock and food animals, especially for non-therapeutic purposes like growth promotion has been one of the drivers of antibiotic overuse at the community level (MoHFW, 2017).

AMR in Human

The emergence of carbapenem resistance, particularly carbapenem resistant Enterobacteriaceae (CRE), has been a major concern, especially in the hospital/health care setting. There has been increase in carbapenem resistance in isolates of *E. coli*, *Klebsiella pneumoniae* and *Acinetobacter baumannii* from India considered as a major health concern (CDDEP, 2016). Owing to established effective antibiotic stewardship and/or infection prevention and control programmes, the proportion of Methicillin-resistant *Staphylococcus aureus* (MRSA) isolates has been decreasing. In India, extended spectrum beta-lactamase (ESBL) producing strains of Enterobacteriaceae have emerged as a challenge in hospitalized patients as well as in the community. In a multicentric study conducted in seven tertiary care hospitals in Indian cities, 61% of *E. coli* isolates were ESBL producers. In the same study, 31–51% *Klebsiella* species were carbapenem resistant, 65% *Pseudomonas* sp. were resistant to ceftazidime and 42% were resistant to imipenem (Mehta *et al.*, 2007). *Acinetobacter* species isolated from hospitalized patients in a tertiary care hospital in Delhi showed 57–80% resistance to imipenem/meropenem while 70%

isolates were resistant to tigecycline. In a study conducted at 15 tertiary care centres on *S. aureus* isolates, the Indian Network for Surveillance of Antimicrobial Resistance (INSAR) found MRSA prevalence rate of 41% which also showed a high rate of resistance to ciprofloxacin, gentamicin, cotrimoxazole, erythromycin, and clindamycin (INSAR, 2013). Emergence of multi drug resistance (MDR) in isolates of *Vibrio cholera*, *Neisseria gonorrhoeae*, *Salmonella typhi*, *Enterococcus faecalis* and *E. coli* are drawing our attention for implementation of effective surveillance and monitoring as well as its strategic prevention and control strategies. Bacteria differ in terms of the mechanisms by which they develop antibiotic resistance. Over the last couple of decades, novel mechanisms and dissemination of antibiotic resistance have been identified. Examples include the New Delhi metallo beta lactamases in Gram negative bacteria, AmpC (a type of beta-lactamase) mediated drug resistance in Enterobacteriaceae, Vancomycin resistant Enterococci (VRE), and XDR *Mycobacterium tuberculosis*. Of late, drug resistant mechanisms for carbapenems and colistin (which are important last resort antibiotics) have been identified among Gram negative organisms, and cases of colistin resistance are being reported (Aggrawal *et al.*, 2018). The rise of “superbugs” is another cause for alarm. They denote multi drug resistant organisms that can be treated only with the use of high end antibiotics; examples of superbugs include the “ESKAPE” group of organisms i.e., *Enterococcus faecium*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, and *Enterobacter* species (Santajit *et al.*, 2016). More studies from India are required and a scaling up is essential to identify patterns of resistance as well as to quantify the nationwide mortality burden owing to AMR.

AMR in livestock/food animals

The burden of AMR in livestock and food animals has been poorly documented in India. Increasing the use of antibiotics in animal sector is resulting in a greater selection of pathogens, is being driven by

increased demand for milk, meat and chicken. Aside from sporadic, small, localized studies, evidence that can be extrapolated to the national level is lacking. Given that there are few regulations against the use of antibiotics for non-therapeutic purposes in India, with no stringent implementation protocols even when there are regulations, the emergence of AMR from antibiotic overuse in the animal sector is likely to be an unmeasured burden in India. India does not have a surveillance system that accounts for use and/or consumption of antibiotics in the animal/food/livestock sector. India manufactured about a third of the total antibiotics produced globally. Hence, it is likely that the consumption patterns and emerging AMR through antibiotic use in the livestock sector is an important target for policy-makers. The detection of antibiotic residues in milk and dairy products has been the main approach for estimating the use of antibiotic in dairy cattle, followed by non-compliance with withholding periods to allow the drug to be completely metabolized before consuming milk. A survey conducted by the National Dairy Research Institute (NDRI) identified that the most commonly used antibiotics in cattle were – tetracycline, oxytetracycline, gentamicin, ampicillin, amoxicillin, cloxacillin, and penicillin due to their lower costs (Grover *et al.*, 2013). Use of enrofloxacin, lincomycin, streptomycin and chloramphenicol was also common when veterinarians treated for clinical conditions. More recent studies have shown that the most common indication for using antibiotics in dairy cattle is mastitis, and the preferred antibiotics include beta-lactams and streptomycin (Unnikrishnan *et al.*, 2005). A high level of AMR was reported from Shiga toxin-producing *E. coli* isolated from calves with diarrhoea in Gujarat and the Kashmir Valley. Resistance was ubiquitous for kanamycin and cephalexin and was above 50% for seven of the antibiotics tested (Arya *et al.*, 2008). On analyzing milk samples for the estimation of AMR in livestock, 48 per cent of Gram-negative bacilli detected in cow and buffalo milk were extended-spectrum β -lactamases (ESBL) producers (West Bengal) and 47.5 per cent were resistant to oxytetracycline (Gujarat) (Das *et al.*, 2017). Among the Gram-positive organisms isolated from these milk samples,

2.4 per cent of *S. aureus* were vancomycin resistant (West Bengal) (Bhattacharyya *et al.*, 2016) while the rate of methicillin resistance was 21.4 per cent for *S. aureus* and 5.6 per cent for coagulase-negative Staphylococci (Karnataka) (Preethirani *et al.*, 2015). India, with 9579×103 tonnes of fish is produced per annum is becoming an important hub of aquaculture industry. In the lakes of Maharashtra, 48 per cent Enterobacteriaceae isolated from the gut of Tilapia fish were ESBL producers (Marathe *et al.*, 2016). *Vibrio cholera* and *V. parahaemolyticus*, isolated from the retail markets of shrimps, shellfish and crabs in Kerala were 100 per cent resistant to ampicillin, 100 per cent susceptible to chloramphenicol while resistance to ceftazidime ranged from 67 to 96 per cent (Sudha *et al.*, 2014). The studies conducted on AMR in poultry reported the presence of *Salmonella* species in broilers to vary from 3.3 percent in Uttar Pradesh to 23.7 per cent in Bihar with 100 per cent isolates being resistant to ciprofloxacin, gentamicin and tetracycline in Bihar and West Bengal. ESBL producing Enterobacteriaceae have been documented from Odisha (9.4 per cent), Odisha and Madhya Pradesh (33.5 per cent) and Punjab (87 per cent) (Gandra *et al.*, 2017).

AMR in Environment

Different water sources such as rivers, lakes, ponds, springs, hand pumps and tube-wells are important environmental samples used for isolation of antibiotics resistant microbes. The major sources are the pharmaceutical waste waters and hospital effluents that are released into the nearby water bodies (Rivers mostly) without adequate treatment. The polluted water bodies may be one of the source for transfer of antibiotic resistant genes (*bla*NDM-1 and *bla*OXA48) to man and animals. Detection of third generation cephalosporin resistant *E. coli* and ESBL producing Gram-negative bacteria poses a great health issues (Kumar *et al.*, 2013).

AMR in Uttarakhand

During 2009-2020, studies were conducted in the Department of Veterinary Public Health and

Epidemiology, College of Veterinary and Animal Sciences, GBPUA&T, Pantnagar, Uttarakhand on antimicrobial resistance profile of non-typhoidal serotypes of *Salmonella* species and thermophilic species of *Campylobacter* under the ICAR funded project 'Outreach Programme on Zoonotic Diseases'.

Phenotypic AMR study was done using Kirby Bauer method on the isolates obtained from milk, meat, chicken, water and faecal samples from different places of Uttarakhand. The isolates obtained during this period were tested against various antibiotics. The resistance observed by the non-typhoidal *Salmonella* species were as: Sulphamethizole/Sulphamethazine, Cefalexin, Furazolidone (100%), Erythromycin (96.6%), Nalidixic acid (75.34%), Cefazolin (65.77%), Ampicillin (64.71%), Gatifloxacin (62.69%), Tetracycline (60.04%), Sulfafurazole/ Sulfisoxazole (59.7%), Ciprofloxacin (58.46%), Kanamycin (50%), Cefotaxime (48.84%), Levofloxacin (44.89%), , Enrofloxacin (26.09%), Cefoxitin (25.09%), Streptomycin (23.23%), Gentamicin (14.66%), Tobramycin (10%), Chloramphenicol (8.06%), Amikacin (8%), Amoxicillin/Clavulanic acid (6%) and Norfloxacin (4%). All the isolates were sensitive to Co-trimoxazole and Oxytetracycline. The isolates showed 100 % sensitivity against Co-trimoxazole and Oxytetracycline. The isolates exhibited less resistance (<50%) against Cefotaxime (48.84%), Levofloxacin (44.89%), Sulfisoxazole (38.78%), Enrofloxacin (26.09%), Cefoxitin (25.09%), Streptomycin (23.23%), Tobramycin (10%), Chloramphenicol (8.06%), Amikacin (8%), Amoxicillin/Clavulanic acid (6%) and Norfloxacin (4%). The anti-microbial resistance was observed in a range of 0-100% that is extreme variation in the sensitivity of *Salmonella* isolates against different anti- microbial agents. In another study, high level of sensitivity in *Salmonella* isolates was observed against those antibiotics such as Chloramphenicol (100%) and Tetracycline (92%) which were not commonly in use for past few years due to availability of new classes of anti-microbial agents. All *Salmonella* isolates (100%) exhibited resistance against 2 or more anti-microbial agents.

The prevalence of such a large number of MDR *Salmonella* isolates (100%) indicated widespread and extensive use of anti-microbial agents in these areas which are of great public health concern.

Antimicrobial resistance genes (args)

The rampant use of antibiotics has led to the emergence of bacteria resistant to the antibiotics and the genes responsible for the resistance can be transferred horizontally to other bacteria of same or different genus. Thus, the detection of resistance genes backs the phenotypic resistance exhibited by the *Salmonella* isolates. In the study, all resistant *Salmonella* isolates were assessed for 16 corresponding antimicrobial resistance. The distribution of 4 aminoglycoside (Streptomycin) resistance genes *aadA1*, *aadA2*, *strA* and *strB* in the resistant 50 *Salmonella* isolates was diverse. The

Table 1: Phenotypic antibiotic resistance of *Salmonella* isolates

S. No.	Antibiotic	Class	Resistance (%)*
1.	Sulphamethizole	Sulphonamide	100
2.	Furazolidone	Nitrofurans	100
3.	Kanamycin	Aminoglycoside	50
4.	Gentamicin	Aminoglycoside	14.66
5.	Nalidixic acid	Quinolone	75.34
6.	Tobramycin	Aminoglycoside	10
7.	Amikacin	Aminoglycoside	8
8.	Ampicillin	Penicillin	64.71
9.	Streptomycin	Aminoglycoside	23.23
10.	Tetracycline	Tetracycline	60.04
11.	Amoxicillin/Clavulanic acid	Penicillin	6
12.	Norfloxacin	Quinolone	4
13.	Ciprofloxacin	Quinolone	58.46/48.72
14.	Cefotaxime	Cephalosporin	48.84
15.	Chloramphenicol	Chloramphenicol	8.06
16.	Cefalexin	Cephalosporin	100
17.	Co-trimoxazole	Sulphonamide	0
18.	Oxytetracycline	Tetracycline	0
19.	Erythromycin	Macrolide	96.6
20.	Gatifloxacin	Quinolone	62.69
21.	Cefazolin	Cephalosporin	65.77
22.	Levofloxacin	Quinolone	44.89
23.	Sulfisoxazole/Sulfafurazole	Sulphonamide	59.7%
24.	Cefoxitin	Cephalosporin	25.09
25.	Enrofloxacin	Quinolone	26.09

*Average percentage of resistance observed in different studies

aadA1, *aadA2*, *strA* and *strB* genes were present in 42 (84%), 20 (40%), 24 (48%) and 5 (10%) isolates respectively. Sulphafurazole resistance encoding *sul1* and *sul2* genes were harboured by 51.13% (68/133) and 3.76% (5/133) isolates, respectively. β -lactam resistant genes (*blaTEM*, *blaPSE*, *blaCMY*) were screened on β -lactam (Ampicillin, Cefoxitin, Cefazolin and Cefotaxime) resistant isolates. The *blaTEM* gene was the predominant β -lactam genes, detected in 36% (63/175) of the β -lactam resistant isolates while *blaCMY* was present in two isolates (1.14%, 2/175) and one isolate (0.57%, 1/175) harboured the *blaPSE* gene. The Tetracycline resistant isolates were tested against three Tetracycline resistant genes (*tetA*, *tetB*, *tetG*). The resistance gene *tetA* was present in 61.85% (107/173) isolates, whereas none of the isolates carried *tetB* and *tetG* genes (Table 2).

Campylobacter isolates were subjected to antibiotic sensitivity testing against 8 antimicrobial agents (ampicillin, gentamicin, erythromycin, levofloxacin, ciprofloxacin, nalidixic acid, ceftriaxone and cotrimoxazole) by disc diffusion method. All the isolates were found to be completely resistant to nalidixic acid and completely sensitive to erythromycin and cotrimoxazole. Almost all the isolates were sensitive to all the studied antibiotics up to some extent except nalidixic acid. The isolates showed no resistance against erythromycin,

levofloxacin, ciprofloxacin, ceftriaxone and cotrimoxazole while 5.3%, 2.6% and 100% isolates were resistant against ampicillin, gentamicin and nalidixic acid, respectively (Garhia *et al.*, 2020, Rajagunalan, 2010).

Dhayananth (2019) reported that *Campylobacter* isolates from poultry farms showed antibiotic sensitivity pattern in which Cefoxitin had highest resistance as 95% followed by Ciprofloxacin 80%, Nalidixic acid-25%, Cefatoxime-22.5%, Ampicillin-20%, Tetracycline-12.5%, Clindamycin-7.5% and Erythromycin-5%. The antibiotics Gatifloxacin and Streptomycin were susceptible to all the isolates screened but some intermediate resistance was found in Sulfisaxazole and Levofloxacin.

Antibiotic resistance profile *Campylobacter* isolates were determined using eight antibiotics according to the CLSI 2015. Comparison of antibiotic susceptibilities of the isolates from different sources (water, milk, meat, chicken and faecal samples) were accessed. All isolates (100%, n=38) were resistant to NA, while all were sensitive to ERY and COT. Only 2 (5.3%) *C. coli* isolates were resistant to AMP and 1 (2.6%) *C. jejuni* was resistant to GEN. Of all the isolates, 13.16%, 18.42%, 23.68%, 36.84%, and 23.68% were intermediately resistant to AMP, GEN, LE, CIP, and CTR, respectively probably reflecting a shift toward resistance.

Table 2: Distribution of antimicrobial resistant genes in the *Salmonella* isolates

S.N.	Resistance genes	Antimicrobial	No. of resistant isolates	Resistance gene positive
1.	<i>blaTEM</i>	Beta-lactam	175	63 (36%)
2.	<i>blaPSE</i>	Beta-lactam	175	1 (0.57%)
3.	<i>blaCMY</i>	Beta-lactam	175	2 (1.14%)
4.	<i>aadA1</i>	Streptomycin	50	42 (84%)
5.	<i>aadA2</i>	Streptomycin	50	20(40%)
6.	<i>strA</i>	Streptomycin	50	24 (48%)
7.	<i>strB</i>	Streptomycin	50	5 (10%)
8.	<i>sul1</i>	Sulphafurazole	133	68 (51.13%)
9.	<i>sul2</i>	Sulphafurazole	133	5 (3.76%)
10.	<i>tetA</i>	Tetracycline	173	107 (61.85%)
11.	<i>tetB</i>	Tetracycline	173	0
12.	<i>tetG</i>	Tetracycline	173	0
13.	<i>gyrA</i>	Flouroquinolones	46	42 (91.3%)
14.	<i>parC</i>	Flouroquinolones	46	42 (91.3%)
15.	<i>qnrA</i>	Flouroquinolones	46	0
16.	<i>qnrS</i>	Flouroquinolones	46	1 (2.17%)

The development of resistance in the poultry and animals of the study area is a potential threat to human health. There is much fear that this resistance may spread to environment which may further lead to difficult to treat cases (Rawat *et al.*, 2018). Upadhyay and Ipshta, 2017 reported that all the *C. jejuni* isolates (100%) were found to be sensitive to Amoxiclav, while 93.75% showed resistance towards Penicillin G. Among 7 *Campylobacter coli* isolates, 5 (71.43%) were sensitivity to Amoxycyclav and Ampicillin. Only Penicillin G was found to be resistant in 5 (71.43%) of the *C. coli* isolates. Upadhyay and his coworker in 2016 also found that all the *C. jejuni* and *C. coli* isolates (chicken, chevon and pork) were resistant to Cephalothin (100%) and sensitive to Gentamicin and Erythromycin (100%). While 80%, 50% and 40% resistance was observed against Suphamethoxazole, Ampicillin and Ciprofloxacin respectively. Among *C. coli*, 70%, 80% and 50% sensitivity was observed against Ciprofloxacin, Nalidixic acid and Gentamicin respectively and 50% isolates were resistant to Ampicillin. This study indicates significance of chickens as important reservoirs of this enteric pathogen and in transmission and dissemination of campylobacter associated diseases to human being.

Steps for containment of AMR

Antimicrobial resistance is a growing public health problem. Resistance has emerged even to newer & more potent antimicrobial agents like Carbapenems. India has given due cognizance to the problem of Antimicrobial resistance (AMR). To tackle this issue, Government of India launched a “National Programme on AMR Containment” during the 12th five-year plan (2012-2017) which is being coordinated by National Center for Disease Control (NCDC), New Delhi. The main objectives of this programme are

1. Establish a laboratory-based AMR surveillance system in the country to generate quality data on antimicrobial resistance
2. Carry out surveillance of antimicrobial usage in different health Care settings

3. Strengthen infection control practices and promote rational use of antimicrobials through Antimicrobial stewardship activities
4. Generate awareness amongst health care providers and community on antimicrobial resistance and rational use of antimicrobials.

Under the programme, National Antimicrobial Surveillance network (NARS-Net) has been established to determine the magnitude and trends of AMR in different geographical regions of the country. The network labs under NARS-Net are required to submit AMR surveillance data of seven priority bacterial pathogens of public health importance: *Klebsiella* spp., *Escherichia coli*, *Staphylococcus aureus*, and *Enterococcus* spp., *Pseudomonas* spp, *Acinetobacter* spp., *Salmonella enteric* serotypes Typhi and Paratyphi. The lists of 29 labs in 24 states/UTs were currently included in the network. From Uttarakhand state Govt. Medical College, Haldwani is the only college selected. Along with the collection submission of AMR surveillance data, NCDC have formulated other guidelines – such as National Treatment Guidelines, Infection Prevention & Control guidelines and surveillance of Health care associated infections and different awareness programmes. The Ministry of Health & Family Welfare (MoHFW) identified AMR as one of the top 10 priorities for the ministry’s collaborative work with World Health Organization (WHO).

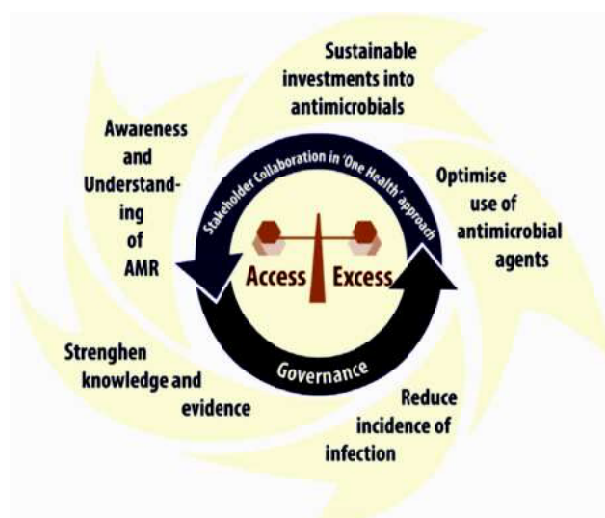


Fig. Activities under NAP-AMR

In 2016, Govt. of India has formulated National Action Plan for containment of AMR (NAP-AMR). Its goal is to effectively combat antimicrobial resistance in India, and contribute towards the global efforts to tackle this public health threat. The scope of the NAP-AMR focuses primarily on resistance in bacteria. With defined objective and strategic priorities central government implemented the programme for 2017-2021 but presently only few states have adopted this programme while Uttarakhand is still lacking in NAP-AMR adoption in full swing. Following activities were included in NAP-AMR programme.

CONCLUSION

Antimicrobial resistance (AMR) is a major public health problem globally. While all types of AMR are concerning, antibacterial resistance (ABR) is seen as currently posing the most serious health threat. Bacteria are present everywhere, including in every living being and in the soil, water and air. With the interconnected ecosystems (humans, animals, the environment), the exchange of bacteria is continuous, and thus the antibiotic resistance problem is no longer limited to medical science alone. It requires effective collaboration among several disciplines (Medical, Veterinary, Agriculture, Fisheries and Environmental sciences) of different departments/institutions within and outside the country for AMR/MDR containment.

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