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A Fifteen-year Report of Serotype Distribution and Antimicrobial Resistance of *Salmonella* in the Philippines

Sonia Sia, Marietta Lagrada, Agnettah Olorosa, Marilyn Limas, Manuel Jamoralin Jr., Polle Krystle Macaranas, Holly Grace Espiritu, June Gayeta, Melissa Masim, Ferissa Ablola, Celia Carlos

Antimicrobial Resistance Surveillance Reference, Research Institute for Tropical Medicine

ABSTRACT

Background. *Salmonella enterica* ser. Typhi and *Salmonella enterica* ser. Paratyphi are agents of typhoid fever, a severe systemic disease, which remains to be a public health concern in the Philippines. Infection due to non-typhoidal *Salmonella* (NTS), on the other hand, most often results in a self-limiting acute gastroenteritis but may result in invasive disease in some cases. There is scarcity of information on the *Salmonella* serotypes in the Philippines which limits understanding of the distribution, transmission and antimicrobial resistance of these bacteria.

Objective. This study describes the serotype distribution and antimicrobial resistance of *Salmonella* in the Philippines over a 15-year period.

Methodology. *Salmonella* isolates were collected through the Philippine Department of Health-Antimicrobial Resistance Surveillance Program (DOH-ARSP) from January 1, 2004 to December 31, 2018. The isolates were serotyped using Sven Gard method for slide agglutination using antigens from Denka Seiken (Japan), and S and A serotest (Thailand). Antigenic formula obtained were classified according to White-Kauffmann-LeMinor scheme. Antimicrobial susceptibility testing for ampicillin, ceftriaxone, cefotaxime, chloramphenicol, ciprofloxacin, and trimethoprim-sulfamethoxazole, were performed using both automated and conventional methods (Kirby Bauer disk diffusion and gradient diffusion method). Antimicrobial susceptibility results were interpreted using Clinical and Laboratory Standards Institute (CLSI) 2018 interpretive criteria (M100Ed28E).

Results. A total of 2,387 isolates were collected from human specimens during the 15-year study period. There were 69 serotypes of *Salmonella* identified with the most common being *Salmonella enterica* ser. Typhi: n=1895 (79.39%), *Salmonella enterica* ser. Enteritidis: n=182 (7.62%), *Salmonella enterica* ser. Typhimurium: n=87 (3.64%), *Salmonella enterica* ser. Weltevreden: n=24 (1.00%), *Salmonella enterica* ser. Paratyphi A: n=17 (0.71%), *Salmonella enterica* ser. Stanley: n=17 (0.71%), *Salmonella enterica* ser. Anatum: n=13 (0.54%), *Salmonella enterica* ser. Heidelberg: n=12 (0.50%), *Salmonella enterica* ser. Choleraesuis var. Kunzendorf: n=9 (0.38%). The multidrug resistant *Salmonella* serotypes reported in this study were mostly resistant to ampicillin, cefotaxime, ciprofloxacin combinations.

Conclusion. This present study showed that prevailing *Salmonella* serotypes in the Philippines were similar with *Salmonella* serotypes reported from other Asian countries. Typhoidal isolates were high among 6-17 years old and were mostly from males. The antimicrobial resistance rates for typhoidal *Salmonella* isolates to ampicillin, chloramphenicol, trimethoprim-sulfamethoxazole, ciprofloxacin, ceftriaxone and cefotaxime were lower compared with the antimicrobial resistance rates for non-typhoidal *Salmonella* isolates. Multidrug resistance for both *Salmonella* Typhi and NTS were relatively low. Continued and enhanced surveillance is needed to monitor the rising levels of antimicrobial resistance, determine risk factors and exposures associated with *Salmonella* Typhi and NTS infection to guide prevention and control measures.

Key words: Salmonella Typhi, NTS, serotype distribution, antimicrobial resistance, multidrug resistant

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Corresponding author: Sonia B. Sia, MD
E-mail: ssiaphl@gmail.com

INTRODUCTION

Salmonella enterica is the agent of typhoid and paratyphoid fever (enteric fever), as well as of salmonellosis and non-typhoidal infections. Globally, 14.3 million estimated cases of enteric fever (10.9 M typhoid fever and 3.3 paratyphoid fever) and 535,000 cases of non-typhoidal *Salmonella* (NTS) invasive disease were reported in 2017.[1] Agents of enteric fever (*Salmonella enterica* serovar Typhi and *Salmonella enterica* serovar Paratyphi) are transmitted through fecal contamination of food or water by ill or asymptomatic chronic carriers while agents of salmonellosis and nontyphoidal *Salmonella* are transmitted through consumption of contaminated water, food animal products or fresh produce, and contact with animals or their environment.[2]

Enteric fever due to *Salmonella* Typhi (SAT) and *Salmonella* Paratyphi serotypes A, B and C is a severe systemic disease requiring antibiotic therapy. In the Philippines, there were 11,140 reported cases of typhoid fever from January 1 to July 2018, with 20 reported deaths.[3] Locally, cases of uncomplicated typhoid fever are treated with ampicillin, chloramphenicol, and trimethoprim-sulfamethoxazole.[4] Based on national AMR surveillance in the country, these antibiotics have remained effective against agents of enteric fever with resistance rates remaining under less than five percent for these antibiotic from 2004-2018 (Figure 1A and 1B).[5]

Infection due to Non-typhoidal *Salmonella* is commonly a self-limiting acute gastroenteritis. Treatment is thus primarily directed to replacement of fluid and electrolytes and antimicrobials are not routinely recommended for uncomplicated NTS gastroenteritis. Invasive infections, however, may occur in less than 5% of patients for which oral therapy may include fluoroquinolone, trimethoprim-sulfamethoxazole or amoxicillin. Figure 1C and 1D shows resistance rates of NTS to these antibiotics based on the national AMR surveillance in the country.

Emergence of multidrug resistance among typhoidal and non-typhoidal *Salmonella* attributed to transferable plasmids, however, have been observed. In a particular strain - SAT H58, antimicrobial resistance (AMR) genes have been associated with an IncHI1 plasmid.[6] In the 1990s, there was worldwide emergence of multidrug resistant *Salmonella* Typhimurium (phage type 104 or DT104) which were resistant to ampicillin, chloramphenicol, streptomycin, sulfonamides and tetracycline.

Salmonella enterica is highly diverse bacterial pathogen with over 2,600 known serotypes.[7] Identifying *Salmonella* serovar plays an important role in understanding the epidemiology of the pathogen, is used to establish link between cases as well as to track potential source of infection. The World Health Organization (WHO) reported that *Salmonella* serovars Enteritidis, Typhimurium, Typhi, Heidelberg, Infantis, Virchow, Hadar, Saintpaul, Montevideo and Agona were the most common serotypes isolated from human sources worldwide.[8]

There is scarcity of information on *Salmonella* serotypes in the Philippines, particularly for NTS, which limits understanding of the distribution and transmission of these bacteria. This report provides valuable information on the prevailing *Salmonella* serotypes in the country from 2004 to 2018 (Figure 2). Antimicrobial resistance among these bacteria will also be described.

METHODOLOGY

Isolates

Salmonella isolates from clinical specimens were collected through the Philippine Department of Health-Antimicrobial Resistance Surveillance Program (DOH-ARSP) from January 1, 2004 to December 31, 2018. The DOH-ARSP is a laboratory based antimicrobial resistance surveillance with 24 sentinel sites representing 16 regions in the country. Case finding for ARSP is based on priority specimens sent routinely to sentinel sites laboratories for clinical purposes. The initial bacterial identification and antimicrobial susceptibility of the isolates were confirmed by automated (Vitek 2) and conventional biochemical tests systems in the Antimicrobial Resistance Surveillance Reference Laboratory (ARSRL). Results are managed and analyzed using WHONET software.

Serotyping

The isolates were serotyped using Sven Gard method for slide agglutination using antigens from Denka Seiken (Japan), and S and A serotest (Thailand). Antigenic formula obtained were classified according White-Kauffmann-LeMinor scheme, as recommended by the World Health Organization Collaborating Centre for Reference and Research on *Salmonella*.[9]

Antimicrobial Susceptibility Testing

Antimicrobial susceptibility testing for ampicillin, ceftriaxone, cefotaxime, chloramphenicol, ciprofloxacin, and trimethoprim-sulfamethoxazole, were performed using both automated (Vitek 2) and conventional methods (Kirby Bauer disk diffusion and gradient diffusion method). Antimicrobial susceptibility results were interpreted using Clinical and Laboratory Standards Institute (CLSI) 2018 interpretive criteria (M100Ed28E).

Data Extraction

The research data were derived from the laboratory-based surveillance software (WHONET). Data gathered included identification (ID) and antifungal susceptibility (AST) and routine demographic information. No clinical information from the patients' chart were included thus, factor such as timing of specimen collection cannot be identified. A data collection tool was used to extract information of the isolates.

Ethical Considerations

During the biobanking (preservation) process, a biobank form is completed indicating the assigned accession number of the isolates. Isolate information in the biobank form includes ID and susceptibility profile, age, specimen type, and birthday of the patient; the name of the patient is not indicated. All of the information required in the data collection tool for this study were extracted exclusively from the biobank form. Since ARSRL department only handles referred isolates from its sentinel sites, no patient researcher interaction

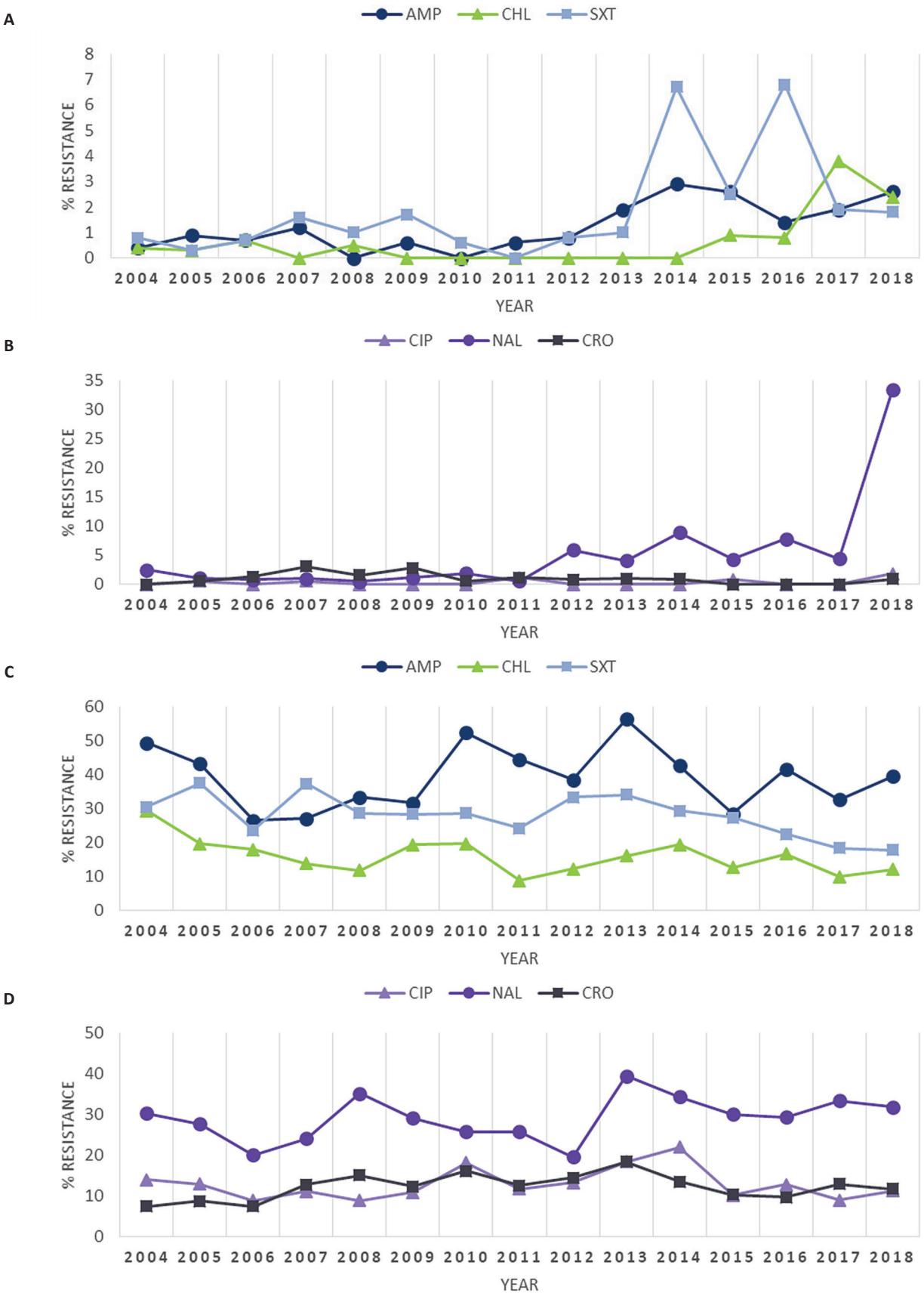

Figure 1. Yearly antimicrobial resistance rates of *Salmonella* Typhi isolates from 2004-2018 to **(A)** ampicillin, chloramphenicol and trimethoprim-sulfamethoxazole; **(B)** ciprofloxacin, nalidixic acid and ceftriaxone. Yearly antimicrobial resistance rates of NTS isolates from 2004-2018; **(C)** ampicillin, chloramphenicol and trimethoprim-sulfamethoxazole; **(D)** ciprofloxacin, nalidixic acid and ceftriaxone.

	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Total
2004	16	12	9	16	11	10	6	10	11	3	13	12	129
2005	17	13	9	7	11	17	9	15	15	12	25	14	164
2006	20	19	33	25	32	26	21	33	22	15	11	9	266
2007	14	14	12	13	14	7	9	16	13	11	9	7	139
2008	11	5	15	13	14	15	15	19	7	6	8	6	134
2009	8	10	14	15	13	4	11	6	14	17	16	15	143
2010	12	10	10	11	11	9	13	9	9	9	12	7	122
2011	17	12	7	9	14	12	12	16	16	6	7	9	137
2012	8	11	14	13	6	12	11	8	5	6	10	3	107
2013	5	9	2	6	6	15	14	5	4	3	3	7	79
2014	6	8	4	6	6	7	7	6	6	14	10	6	86
2015	8	10	6	6	5	9	10	14	8	16	7	8	107
2016	10	6	3	9	5	11	5	11	16	25	6	9	116
2017	9	16	12	17	9	12	5	9	13	11	6	4	123
2018	6	10	7	8	6	1	6	5	4	8	4	6	71
Average	11	11	10	12	11	11	10	12	11	11	10	8	128.2
	11	11	10	12	11	11	10	12	11	11	10	8	

LEGEND:

0	1-5	6-10	11-15	16-20	21-25	26-30	31-35	36-40	41-45	46-50	>50

Figure 2. Frequency distribution of Typhoidal *Salmonella* per year from 2004 to 2018 (n=1,923).

was established. It was noted that, data obtained for this study were part of an ongoing surveillance in the Philippines and surveillance reports in the country does not require ethical evaluation when used for research purpose, moreover, no informed consent were required from all the patients involved.

Statistical Analysis

No statistical software or tool was used in the study since the nature of the research is purely descriptive. Moreover, no analytical computations were required, and simple quantitative treatment of data were represented (e.g number of species over total number of isolates expressed in %).

RESULTS

A total of 2,387 isolates were collected from human specimens during the 15-year study period. There were 69 serotypes of *Salmonella* identified with the most common being *Salmonella* Typhi: n=1895 (79.39%), *Salmonella* Enteritidis: n=182 (7.62%), *Salmonella* Typhimurium: n=87 (3.64%), *Salmonella* Weltevreden: n=24 (1.00%), *Salmonella* Paratyphi A: n=17 (0.71%), *Salmonella* Stanley: n=17 (0.71%), *Salmonella* Anatum: n=13 (0.54%), *Salmonella* Heidelberg: n=12 (0.50%), *Salmonella* Choleraesuis var. Kunzendorf: n=9 (0.38%) (Table 1).

Salmonella Typhi was the most common typhoidal serotype observed in this study (n=1895, 98.54%). *Salmonella* Paratyphi A (n=17, 0.88%), *Salmonella* Paratyphi B (n=9, 0.46%) and *Salmonella* Paratyphi C (n=2, 0.10%) accounted for the rest. Most of the serotypes were from blood specimens (SAT=1753, *Salmonella* Paratyphi A=15, *Salmonella* Paratyphi B=6 and *Salmonella* Paratyphi C=1). Typhoidal serotypes was 15.8% more commonly isolated from males (n=1087; compared to females n=785) and most were from age group 6-17 years (n=915) (Figure 3). Many were collected from the Visayas region (Table 2) and 93.03% (1789 out of 1923) were from sterile sites. The most number of isolates were collected in 2006 (n= 266). No trend was observed on the number of salmonella isolates reported per month in the 15-year period.

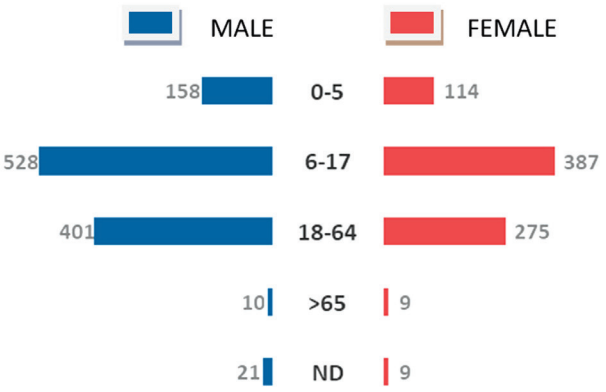

Figure 3. Distribution of Typhoidal *Salmonella* serotypes as to age group and sex.

The yearly antimicrobial resistance rates for typhoidal *Salmonella* serotypes from 2004 to 2018 are shown in Figure 4A. Resistance to ampicillin, chloramphenicol and trimethoprim-sulfamethoxazole remained low from 2007-2018. Chloramphenicol resistance was reported only in 2004 (n=127) (0.8%) and 2006 (n=262) (0.4%). Resistance to ciprofloxacin, ceftriaxone and cefotaxime likewise have remained low throughout the study period (Figure 4B). There were statistically significant increases in ceftriaxone resistance in 2008 (0.4%) to 2010 (0.8%) as well as from 2012 (0%) to 2013 (1.3%).

Table 1. Frequency of *Salmonella* isolates, ARSP, 2004 TO 2018

Salmonella enterica Serotype	Total Number of Isolates
Typhoidal *Salmonella*	**1923**
Salmonella Typhi	1895
Salmonella Paratyphi A	17
Salmonella Paratyphi B	9
Salmonella Paratyphi C	2
Non-typhoidal *Salmonella*	**464**
Salmonella Enteritidis	182
Salmonella Typhimurium	87
Salmonella Weltevreden	24
Salmonella Stanley	16
Salmonella Anatum	13
Salmonella Heidelberg	12
Salmonella Choleraesuis var. Kunzendorf	9
Salmonella Group B	9
Salmonella Virchow	8
Salmonella enterica ss. arizonae	7
Salmonella Kentucky	6
Salmonella Rissen	6
Salmonella Albany	5
Salmonella Choleraesuis	5
Salmonella Group C	5
Salmonella Newport	4
Salmonella Schwarzengrund	4
Salmonella Bardo	3
Salmonella Derby	3
Salmonella Hissar	3
Salmonella Lexington	3
Salmonella Saintpaul	3
Salmonella Agona	2
Salmonella Bonn	2
Salmonella Corvallis	2
Salmonella Essen	2
Salmonella Abardeen *Salmonella* Ajiobo *Salmonella* Bareilly *Salmonella* Bournemouth *Salmonella* Brazaville *Salmonella* Breda *Salmonella* Chailey *Salmonella* Denver *Salmonella* Eastbourne *Salmonella* Emek *Salmonella* Farcha *Salmonella* Group A *Salmonella* Group E1 *Salmonella* Group K *Salmonella* Haifa *Salmonella* Hillingdon *Salmonella* Houtenae *Salmonella* Indica *Salmonella* Irumu *Salmonella* Itami *Salmonella* Javiana *Salmonella* Lome *Salmonella* Lomita *Salmonella* London *Salmonella* Mbandaka *Salmonella* Mgulani *Salmonella* Montevideo *Salmonella* Ndolo *Salmonella* Nessziona *Salmonella* Ohio *Salmonella* Potsdam *Salmonella* Regent *Salmonella* Sandiego *Salmonella* Sangera *Salmonella* Saphra *Salmonella* Seftenberg *Salmonella* Tallahassee *Salmonella* Tumodi *Salmonella* Wanatah	39 (1 isolate each)

Salmonella Enteritidis: n=182 (39.22%) were the most common NTS followed by *Salmonella* Typhimurium: n=87 (18.75%) and *Salmonella* Weltevreden: n=24(5.17%) (Table 1). It was noted that NTS were isolated more from males for all serotypes (Figure 6) except for *Salmonella* Stanley (Male= 7, Female= 10) and *Salmonella* Anatum (Male= 5, Female= 8). The serotypes with the highest number of isolates for 0-5 years old were *Salmonella* Typhimurium (n=45), *Salmonella* Stanley (n=13), *Salmonella* Anatum (n=6), and *Salmonella* Heidelberg (n=10). Highest number of *Salmonella* Weltevreden isolates were seen among 6-17 year olds. On the other hand, *Salmonella* Enteritidis (n=81) and *Salmonella* Choleraesuis var. Kunzendorf (n=5) have the most number of isolates for 18-64 years olds. For more than 65-years population, *Salmonella* Enteritidis (n=21) and *Salmonella* Typhimurium (n=15) were the two serotypes with the most number of isolates. Metro Manila shared the biggest number of isolates for all serotypes except for serotypes *Salmonella* enterica serovars Weltevreden and Heidelberg. Overall, NTS were mostly isolated from blood (n=227, 49%) and stool (n=159, 34%). The most number of NTS isolates were collected (n= 68) in 2018; however, no trend in the reported number of NTS isolates per month was observed for the 15-year period.

Resistance to ampicillin was the highest (63.3%) in 2004 with fluctuations on the resistance rates from 2005 to 2018 (Figure 7). Chloramphenicol and trimethoprim-sulfamethoxazole resistance among NTS was highest in 2008 (33.3%) and remained in the range of 10-20% from 2009-2018. *Salmonella* Typhimurium showed the highest resistance to ampicillin (65.1%) among the NTS. Trimethoprim-sulfamethoxazole resistance rates were lowest for *Salmonella* Stanley (50%) and *Salmonella* Heidelberg (66.7%). Combined ampicillin and ciprofloxacin resistant phenotypes in *Salmonella* enterica serovars Enteritidis (n=8), Typhimurium (n=10) and Choleraesuis var. Kunzendorf (n=3) were also noted.

Table 2. Distribution of *Salmonella sp.* ARSP, 2004 TO 2018

Demographics	Typhoidal (N=1,923)	NTS (N=464)
Island Group		
Metro Manila	321 (16.69%)	202 (43.53%)
Luzon	348 (18.09%)	116 (25%)
Visayas	815 (42.38%)	100 (21.55%)
Mindanao	430 (22.36%)	46 (9.91%)
Specimen Type		
Blood	1775 (92.30%)	227 (48.92%)
CSF	2 (0.10%)	5 (1.07%)
Stool	85 (4.42%)	159 (34.26%)
Urine	18 (0.93%)	17 (3.66%)
Respiratory	4 (0.20%)	3 (0.64%)
Wound	14 (0.72%)	23 (4.95%)
Tissue	5 (0.26%)	0
Fluid	7 (0.36%)	21 (4.52%)
Other	4 (0.20%)	9 (1.93%)

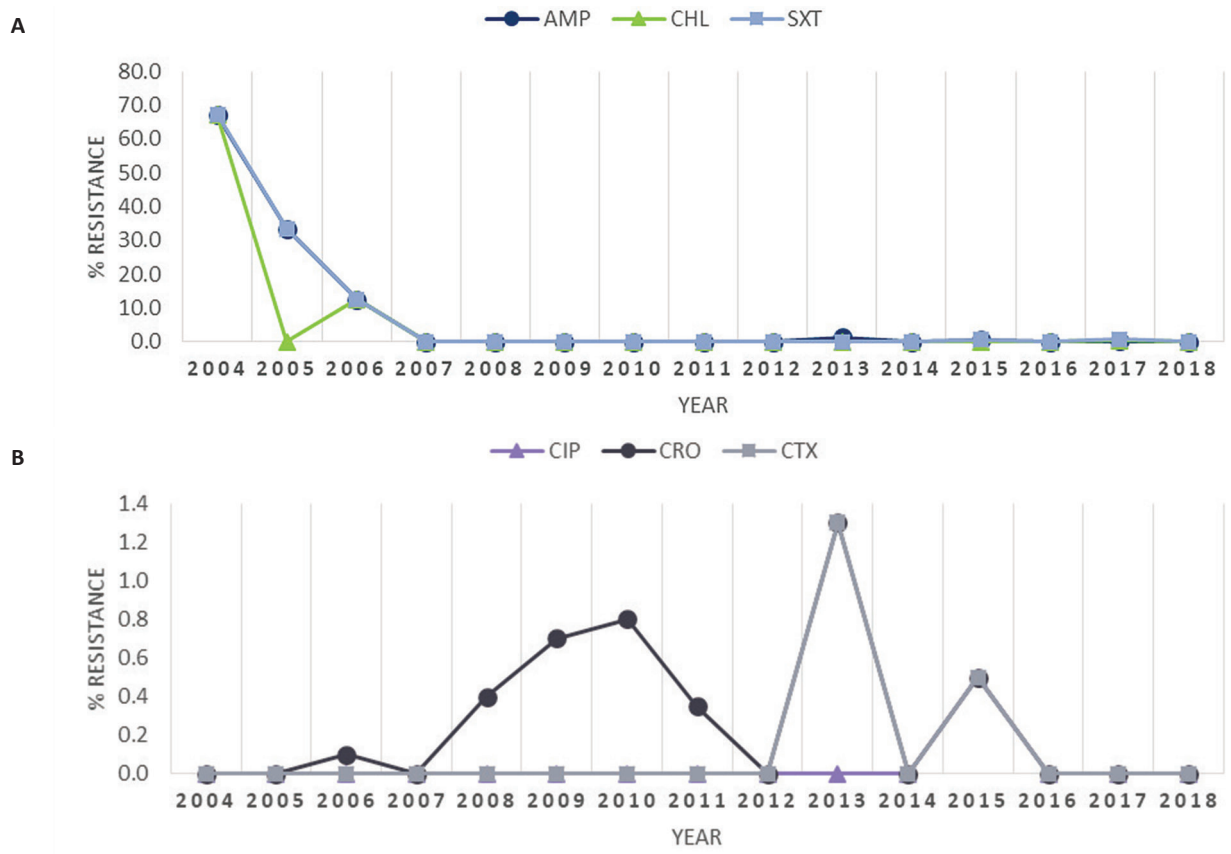

Figure 4. Yearly antimicrobial resistance rates of Typhoidal *Salmonella* serotypes from 2004-2018 to **(A)** ampicillin, chloramphenicol, trimethoprim-sulfamethoxazole; **(B)** ciprofloxacin, ceftriaxone and cefotaxime.

Among the typhoidal *Salmonella* isolates, combined cefotaxime-ciprofloxacin resistance phenotype was seen in 17 *Salmonella* Typhi isolates. Multidrug resistance (resistance to ampicillin-ciprofloxacin-chloramphenicol) was seen among *Salmonella* Paratyphi B and *Salmonella* Paratyphi C. No multidrug resistance was noted among *Salmonella* Paratyphi A. On the other hand, combined ampicillin- ciprofloxacin resistance was noted in *Salmonella* enterica serovars Enteritidis (n=8), Typhimurium (n=10) and Choleraesuis var. Kunzendorf (n=3).

DISCUSSION

A more recent report by Epidemiology Bureau Public Health Surveillance Division of the Department of Health in January 1-to February 23, 2019, revealed that there were 2,720 reported cases of typhoid fever collated nationwide.[10] The number of typhoid fever cases decreased by 16%, from 3,524 cases in 2018 to 2,720 in 2019. There were, however, two confirmed deaths (CFR=0.07%) out of the 2,720 cases reported. The report confirms the persistence of *Salmonella* Typhi infections in the country.

There is scarcity, on the other hand, of data on occurrence of NTS infections, including its distribution and antimicrobial resistance, in the country (Figure 5).

Typhoidal *Salmonella*

The present study showed that both *Salmonella* Typhi and *Salmonella* Paratyphi A were more commonly isolated from males. While available national census data shows higher percentage of males more than females in the country from 2004-2015,[11] the present observation may also be attributed to male behavioral factors such as higher risks in food handling, preparation and consumption.[12]

The most number of typhoidal isolates in this study were collected from the Visayas region. This is in contrast with the Department of Health data from January 1 to February 23, 2019 which showed that the Mindanao region has the most number of *Salmonella* Typhi cases. The relative numbers per island group in the present study may not necessarily indicate relative prevalence of *Salmonella* infections but could be a reflection of the diagnostic practices of clinicians as well as technical capabilities, including laboratory resources, of the sentinel sites of the ARSP. Nevertheless, it can be noted that most areas of these two island groups – Visayas and Mindanao – were rural areas.

The Philippine Progress Report on the Millennium Development Goals 2010 stated that in 1990-2008, 18% percent of the rural areas in the country still practice open defecation.[13] Moreover, the direct exposure of individuals to livestock animals which is prevalent in the country might also increase risk of human *Salmonella* infection. These factors may contribute to the continued presence of Salmonella infections in the country.

As antibiotic therapy is the mainstay for the treatment of typhoid fever and the complications associated with it, the emergence of resistance to antibiotics used against

	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Total
2004	16	12	9	16	11	10	6	10	11	3	13	12	129
2005	17	13	9	7	11	17	9	15	15	12	25	14	164
2006	20	19	33	25	32	26	21	33	22	15	11	9	266
2007	14	14	12	13	14	7	9	16	13	11	9	7	139
2008	11	5	15	13	14	15	15	19	7	6	8	6	134
2009	8	10	14	15	13	4	11	6	14	17	16	15	143
2010	12	10	10	11	11	9	13	9	9	9	12	7	122
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2013	5	9	2	6	6	15	14	5	4	3	3	7	79
2014	6	8	4	6	6	7	7	6	6	14	10	6	86
2015	8	10	6	6	5	9	10	14	8	16	7	8	107
2016	10	6	3	9	5	11	5	11	16	25	6	9	116
2017	9	16	12	17	9	12	5	9	13	11	6	4	123
2018	6	10	7	8	6	1	6	5	4	8	4	6	71
Average	11	11	10	12	11	11	10	12	11	11	10	8	128.2
	11	11	10	12	11	11	10	12	11	11	10	8	

LEGEND:

0	1-5	6-10	11-15	16-20	21-25	26-30	31-35	36-40	41-45	46-50	>50

Figure 5. Frequency distribution of NTS per year from 2004 to 2018 (n=464).

it – chloramphenicol, ampicillin, sulphamethoxazole-trimethoprim, quinolones and cephalosporins – is a concern. Although the origin of antibiotic resistance genes among *Salmonella* Typhi is still unclear, it has been recognized that the two factors that mediate antibiotic resistance are foreign genes acquisition via plasmids and chromosome mutation.[14] Moreover, resistance may be through inactivation of the antimicrobial agent, efflux or transport of the antimicrobial, modification of the antimicrobial target site and reduced permeability of the antimicrobial agent.

Antimicrobial resistance in developed countries has been linked to the utilization of antimicrobial drugs in livestock animals and environmental usage.

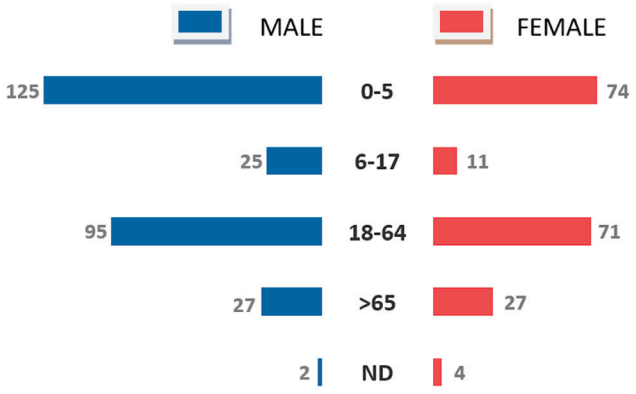

Figure 6. Distribution of NTS serotypes as to age group and sex.

Among developing countries, however, episodes of antimicrobial resistance of both Non-typhoidal and typhoidal *Salmonella* spp. were associated with the use antimicrobials for medication in humans.[15]

Multidrug resistant strains of *Salmonella* resistant to first line antibiotics (ampicillin, chloramphenicol, and cotrimoxazole) have been identified as early as 1989.[16] The widespread use of these drugs facilitated the emergence of resistance to chloramphenicol and subsequently to ampicillin and co-trimoxazole, leading to MDR typhoid. In the present study, however, the percent resistance of *Salmonella* Typhi among ampicillin (0.3%), ceftriaxone (0.1%), cefotaxime (0.4%), ciprofloxacin (0.1%) and sulfamethoxazole-trimethoprim (0.3%) have remained low in the country from 2004-2018.

The multidrug resistant *Salmonella* Typhi reported in this study were mostly resistant to ampicillin, cefotaxime, ciprofloxacin tandems or combinations. On the other hand, no multidrug resistance was noted for *Salmonella* Paratyphi A. No multi-locus sequence typing (MLST) was performed in the present study. It will be interesting to determine the genotypic characteristics of the MDR clone in the country and its relation, if any, with the multidrug-resistant (MDR) *Salmonella* Typhi H58 clone which is the dominant MDR type that circulates in the Indian subcontinent and Southeast Asia[17-19] as well as clones from India, Pakistan and Vietnam which had higher rates of MDR isolates of *Salmonella* Typhi than Indonesia and China.[20]

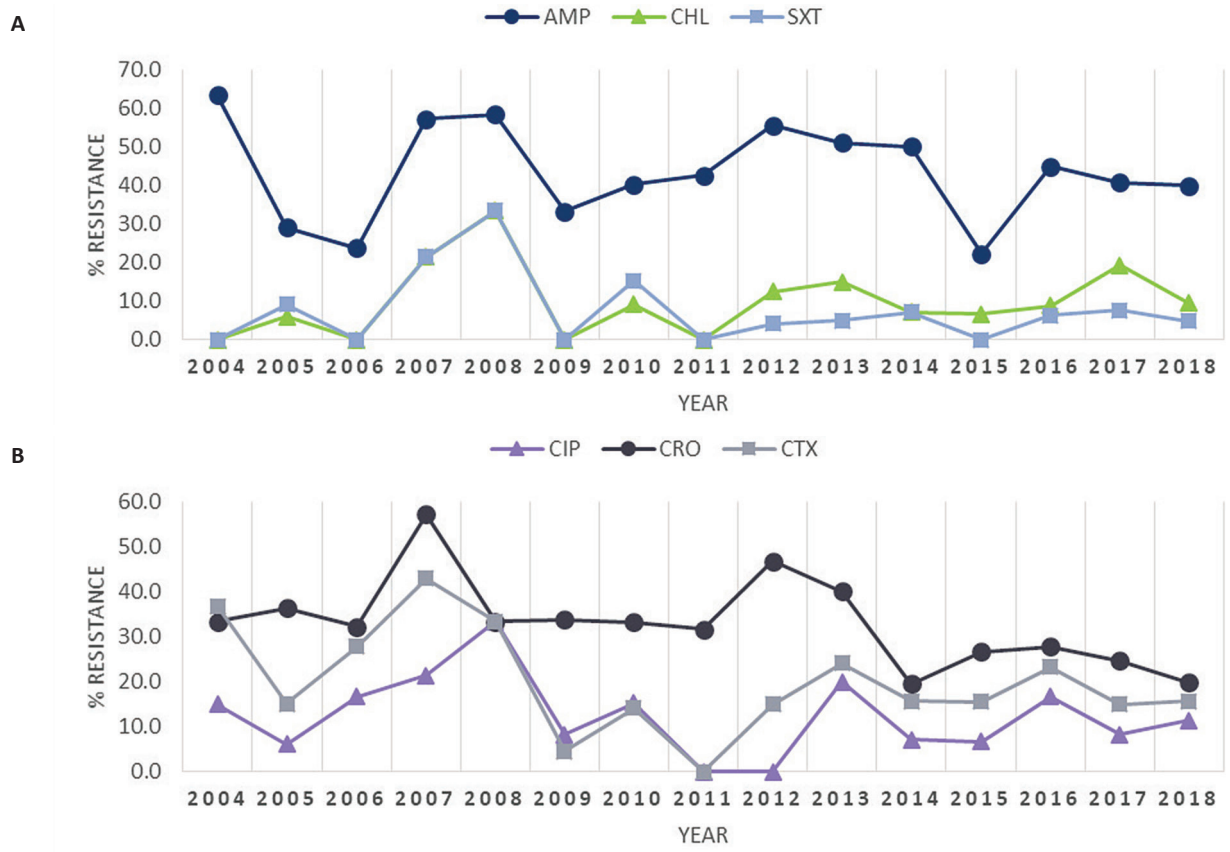

Figure 7. Yearly antimicrobial resistance rates of Non-typhoidal *Salmonella* serotypes from 2004-2018 to **(A)** ampicillin, chloramphenicol, trimethoprim-sulfamethoxazole; **(B)** ciprofloxacin, ceftriaxone and cefotaxime.

Non-typhoidal *Salmonella*

Three of the most frequently isolated NTS serotypes in this present study – S. Enteritidis, S. Typhimurium, and S. Weltevreden - are included in the 2006 WHO list of commonly isolated *Salmonella* strains. ARSP had been part of the WHO Salmonella Surveillance and has contributed data to this surveillance since 1994. It has been reported by WHO that serotype distribution varies with geographical location, age group affected and socio-economic status of the region. It was noted that NTS serotypes reported in the present study were similar to that of Thailand, a developing nation like the Philippine. We report in this study the sporadic occurrence over the15-year period of the following isolates which were not reflected in the 2006 WHO list of commonly isolated *Salmonella* strains: *Salmonella* Stanley, *Salmonella* Anatum, *Salmonella* Choleraesuis var. Kunzendorf.

Although NTS gastroenteritis are typically self-limiting and non-fatal to immunocompetent individuals, invasive NTS (iNTS) infections can lead to death among susceptible populations suffering from malaria, malnutrition and people living with HIV.[21] Case fatality rate of NTS related community acquired bacteremia in African regions was reported to be as high as 20.6%.

In this present study, there were 212 invasive NTS isolates with the most common serotypes being *Salmonella enterica* serovars *Enteritidis*, *Dublin*, and *Typhimurium*. Serovars associated with iNTS in the present study is similar with serovars causing iNTS In Vietnam (*Salmonella* Enteritidis and *Salmonella* Typhimurium). Most of the patients with iNTS were adult males, HIV positive and with history of drug use.[22] Future studies on iNTS may consider identifying risks for this type of infection. Further consideration for risk factor evaluation may also include other potential predisposing factors such as previous medication (acid blockers and antimicrobial pre-treatment).[23]

NTS infections in this study were noted to be higher in urbanized area such as Metro Manila. While this could be due to the urban in-migration that caused expansion and generation of urban slums, wherein there is resultant increased risk of food and waterborne diseases linked to poor water, sanitation and hygiene infrastructures,[24] the high number of NTS may also be accounted for by the diagnostic practices of physicians, capacity of the laboratories, and the relative number of patients in the area.

Reservoir of NTS was previously thought to be exclusively of animal origin. Personal hygiene and regulated food handling were then considered sufficient control measures to prevent NTS infection. Human reservoir of iNTS from Burkina Faso in West Africa, however, has since been confirmed.[25] With the emerging reports of human iNTS reservoir, vaccines has been increasingly explored as a control measure for iNTS. Continuous and comprehensive surveillance of iNTS would be an invaluable information towards vaccine development for agents of iNTS.

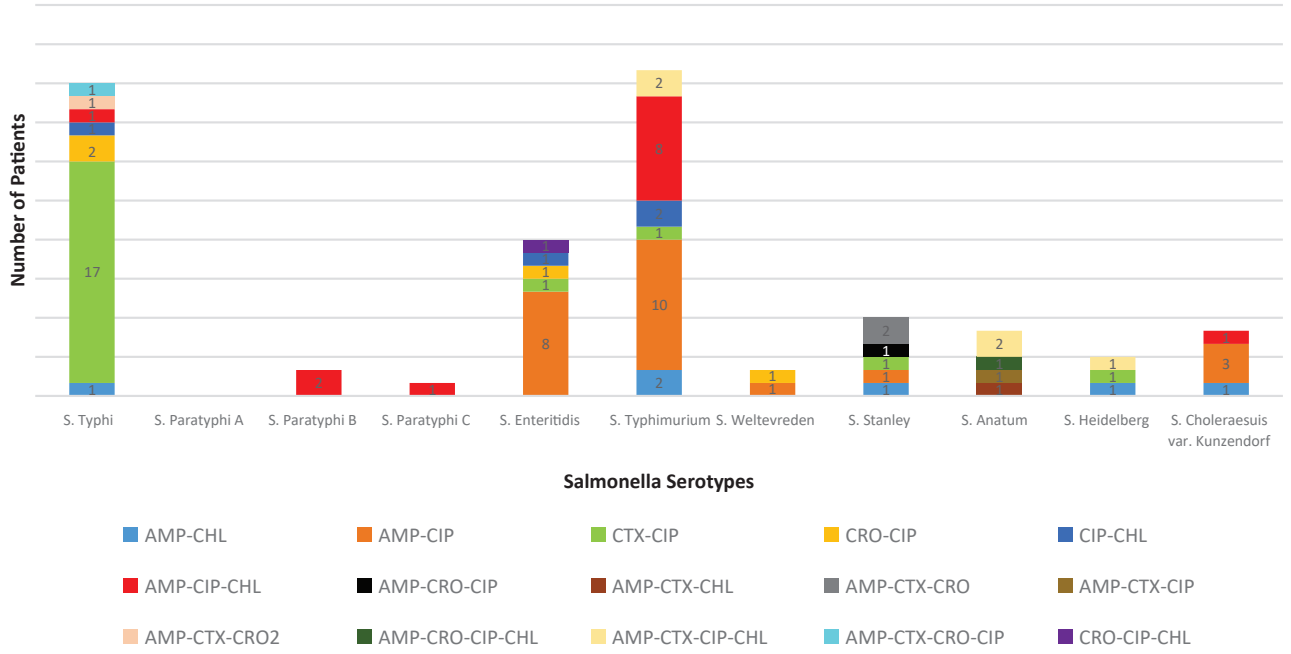

Figure 8. Multidrug resistant *Salmonella* serotypes from 2004-2018.

For NTS in this present study, resistance to antibiotics – chloramphenicol, ampicillin and sufamethoxazole-trimethoprim among *Salmonella* enterica serovars Typhimurium, Anatum, Stanley and Heidelberg were noted. Given, however, the very few number of isolates for respective NTS serotypes in this present study, the results should be interpreted with caution.

Antimicrobial resistance to ciprofloxacin (1.2%), third-generation cephalosporins (12.2%), ampicillin (68.2%) and TMP/SMX (17.0%) have been reported in the first population-based estimates of NTS from Southeast Asia. Similar rates have been found in a tertiary-care setting in Bangkok among children, where rates were 68.3%, 33.9%, 3% and 17.4% respectively.[26] Further, a 2009 multi-national study showed a high prevalence of reduced susceptibility to ciprofloxacin among non-typhoidal *Salmonella* strains from Taiwan (48.1%), Thailand (46.2%), Korea (36.5%), and Sri Lanka (8.0%).[27] In the present study, resistance to third generation cephalosporins (cefuroxime and ceftriaxone) were likewise high at 33.07% as was also observed in Taiwan and Thailand.[28] It is noteworthy that the present study herewith reports occurrence of invasive NTS (n=212) with isolates showing resistance to chloramphenicol, ampicillin and sufamethoxazole- trimethoprim.

The combination of ampicillin and ciprofloxacin resistance were mostly seen in *Salmonella* Enteritidis and *Salmonella* Typhimurium isolates in this present study. In addition, *Salmonella* Typhimurium mostly possess ampicillin-ciprofloxacin- chloramphenicol resistance. Similarly, *Salmonella* Typhimurium and *Salmonella* Enteritidis were the two main NTS serotypes that showed multiple resistance to amoxicillin, ciprofloxacin, cefotaxime and gentamicin in Kenya.[29]

CONCLUSION

The present study showed that prevailing *Salmonella* serotypes in the Philippines were similar with *Salmonella* serotypes reported from other Asian countries. There were 69 serotypes of *Salmonella* identified with the most common being *Salmonella* Typhi: n=1895 (79.39%), *Salmonella* Enteritidis: n=182 (7.62%), *Salmonella* Typhimurium: n=87 (3.64%), *Salmonella* Weltevreden: n=24 (1.00%), *Salmonella* Paratyphi A: n=17 (0.71%), *Salmonella* Stanley: n=17 (0.71%), *Salmonella* Anatum: n=13 (0.54%), *Salmonella* Heidelberg: n=12 (0.50%), *Salmonella* Choleraesuis var. Kunzendorf: n=9 (0.38%). Typhoidal isolates were high among 6-17 years old and were mostly from males.

Multidrug resistance for both *Salmonella* Typhi and NTS were relatively low. The multidrug resistant *Salmonella* serotypes reported in this study were mostly resistant to ampicillin, cefotaxime, ciprofloxacin combinations (Figure 8). Continued and enhanced surveillance is needed to monitor the rising levels of antimicrobial resistance, determine risk factors and exposures associated with *Salmonella* Typhi and iNTS infection to guide prevention and control measures.

STATEMENT OF AUTHORSHIP

All authors certified fulfillment of ICMJE authorship criteria.

AUTHOR DISCLOSURE

The authors declared no conflict of interest.

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