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Research Note

Survival and Growth of Wild-Type and *rpoS*-Deficient *Salmonella* Newport Strains in Soil Extracts Prepared with Heat-Treated Poultry Pellets

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ABSTRACT

Manure runoff can transfer pathogens to farmlands or to water sources, leading to subsequent contamination of produce. Untreated biological soil amendments, like manure, can be contaminated with foodborne pathogens, such as *Salmonella* Newport, which may lead to transfer of the pathogen to fruits or vegetables. Studies have reported the occurrence and survival of *Salmonella* in manure or manure slurries. However, data on the survival and growth of *Salmonella* Newport is lacking in matrices simulating runoff. We quantified the survival and growth of wild-type (WT) *Salmonella* Newport and *rpoS*-deficient (Δ*rpoS*) strains in sterile and nonsterile soil extracts prepared with (amended) or without (unamended) heat-treated poultry pellets at 25°C. *Salmonella* Newport WT and Δ*rpoS* populations reached a maximum cell density of 6 to 8 log CFU/mL in 24 to 30 h in amended and unamended soil extracts and remained in stationary phase for up to 4 days. *Salmonella* Newport in amended soil extracts exhibited a decreased lag phase (λ, 2.87 $\pm$ 1.01 h) and greater maximum cell densities (N_{max}, 6.84 $\pm$ 1.25 CFU/mL) compared with λ (20.10 $\pm$ 9.53 h) and N_{max} (5.22 $\pm$ 0.82 CFU/mL) in unamended soil extracts. In amended soil extract, the Δ*rpoS* strain had no measurable λ, similar growth rates (μ_{max}) compared with WT, and a lower N_{max} compared with the WT strain. Unamended, nonsterile soil extracts did not support the growth of *Salmonella* Newport WT and led to a decline in populations for the Δ*rpoS* strain. *Salmonella* Newport had lower cell densities in nonsterile soil extracts (5.94 $\pm$ 0.95 CFU/mL) than it did in sterile soil extracts (6.66 $\pm$ 1.50 CFU/mL), potentially indicating competition for nutrients between indigenous microbes and *Salmonella* Newport. The most favorable growth conditions were provided by amended sterile and nonsterile soil extracts, followed by sterile, unamended soil extracts for both *Salmonella* Newport strains. *Salmonella* Newport may grow to greater densities in amended extracts, providing a route for increased *Salmonella* levels in the growing environments of produce.

HIGHLIGHTS

- Soil extracts prepared with HTPP supported growth of *Salmonella* Newport by 4 to 5 log CFU/mL in 96 h.
- Lack of *rpoS* led to diminished growth or decline in survival of *Salmonella* Newport.
- Presence of indigenous microbes impaired *Salmonella* Newport growth in soil extracts.

Key words: Biological soil amendments; Heat-treated poultry pellets; Runoff; *Salmonella* Newport; Soil extract

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In recent years, foodborne disease outbreaks from *Salmonella* spp. have been increasingly associated with the consumption of raw vegetables and leafy greens *(15, 17)*. Each year, nontyphoidal *Salmonella* spp. are estimated to cause 1.2 million foodborne infections, 23,000 hospitalizations, and 450 deaths in the United States *(4)*. *Salmonella* spp. can be introduced to the fields used to grow fruits and vegetables through various means, such as the use of contaminated manure, irrigation water, wild animal scat, and bird droppings *(12, 23, 24)*. *Salmonella* spp. from soil can eventually transfer to vegetables and leafy greens, potentially leading to foodborne disease outbreaks *(13)*. *Salmonella enterica* subspecies *enterica* serovar Newport has been responsible for several outbreaks related to contaminated produce, such as cucumbers and tomatoes. The possible sources of *Salmonella* Newport associated with cucumbers in 2014 and with tomatoes in 2005 were attributed to environmental reservoirs, potentially including contaminated soils containing animal manure *(1, 9)*.

Various biological soil amendments of animal origin (BSAAOs) are used in the organic cultivation of fruits and vegetables. Although the use of composted manure can minimize the transfer of pathogens from amended soils to

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crops, use of raw or untreated livestock animal manure is still practiced, leading to increased risks of contamination of produce by enteric pathogens present in manure *(10)*. Although studies have shown that both composted and raw manure can harbor pathogens, raw animal manure is known to contain more pathogens, such as *Salmonella* populations, as high as 10^5 to 10^7 CFU/g *(6, 8, 13)*. Leaching of manure runoff from animal farms to crop fields and eventually to water sources have been previously observed, which can lead to the spread of enteric pathogens on the farm or in the preharvest environment. For example, water wells that supplied drinking water were contaminated with cattle manure runoff containing *Escherichia coli* O157:H7 and *Campylobacter* spp. from nearby farms after rainfall in Walkerton, Ontario, Canada, in 2000, leading to hundreds of illnesses and multiple deaths *(2)*. Similarly, lettuce contaminated with Shiga toxin–producing *E. coli* in 2005 in Sweden, which caused 135 illnesses with 11 cases of hemolytic uremic syndrome, was retrospectively associated to a farm upstream of the irrigation point *(23)*. In addition, an outbreak of *E. coli* O157:H7 infection was associated with lettuce irrigated with water contaminated by cattle manure in the United States in 2006 *(24)*.

Salmonella spp. and *E. coli* O157:H7 can survive for several months in manure-amended soils and manure slurries *(18, 20, 27)*. Some BSAAOs are heat treated and pelletized to inactivate pathogens and stabilize nutrients, such as nitrogen. Heat-treated poultry pellets (HTPP) may not contain pathogens, but their nutrient levels, when amended to soils, may promote or affect survival of enteric pathogens already present in, or introduced to, soils through contaminated irrigation water or animal fecal deposits. Irrigation or rainfall events may result in growth of inoculated pathogens in soil *(16, 21),* which may be due to increased availability of nutrients with the addition of adequate amounts of water causing water-soluble nutrients to be readily available for bacterial growth. However, the extent of *Salmonella* spp. growth in amended or unamended soil extracts is unknown. It is possible that nutrients from soil or manure dissolve in water, allowing that runoff to support not only survival but also growth of *Salmonella* spp. In addition, soil has a diverse population of indigenous microbes that can affect pathogen survival by either competing with *Salmonella* spp. for available nutrients or providing pathogens nutrients through metabolic activity. It is important to understand the roles of such microbes regarding the survival of pathogens. Amended soils and runoff may pose environmental stresses to pathogens, where the general stress response regulator RpoS has been shown to contribute to prolonged survival of *Salmonella* and *E. coli* O157:H7 in manure-amended soil *(5, 25)*.

Our objectives were to (i) quantify the growth and survival characteristics of *Salmonella* Newport in soil extract prepared with or without HTPP, (ii) understand the role of *rpoS* in the survival of *Salmonella* Newport in soil extracts, and (iii) investigate the role of indigenous microorganisms in the survival and growth characteristics of wild-type (WT) *Salmonella* Newport and *rpoS*-deficient (Δ*rpoS*) strains. We have addressed these questions by quantifying the growth and survival of WT *Salmonella* Newport and Δ*rpoS* strains in soil extracts prepared from unamended and HTPP-amended soils in the presence and absence of indigenous microorganisms.

MATERIALS AND METHODS

Soil extract preparation. Soil (fine, loamy, mesic Aquic Hapludults) was obtained from the U.S. Department of Agriculture Agricultural Research Service (USDA-ARS) Beltsville Agricultural Research Center North Farm (Beltsville, MD). For preparation of unamended extract, 500 g of soil were added to 1 L of deionized water in a 2-L bottle. For amended extract, 30 g of HTPP, a commercial BSA (3-2-3 [N-P-K]) was added to 470 g of soil and then added to 1 L of deionized water into a 2-L bottle. The bottles were incubated at 25°C with shaking at 50 rpm for 24 h. That incubation temperature was selected to simulate stagnant water containing manure runoff and had the potential for indigenous microorganisms to grow during the incubation period. After incubation, the resulting liquid extract was transferred to 250-mL centrifuge bottles. Those extracts were centrifuged at 5,000 × *g* for 22 min at 25°C to remove heavy soil particles. After centrifugation, the supernatant was collected and used as nonsterile extracts. To obtain sterile extracts, supernatants were filtered with a 0.2-µm-pore-size filter (Thermo Fisher Scientific, Waltham, MA). The extracts were stored at –20°C for up to 72 h before use.

Microbial profiling of soil extracts. Microbial populations were quantified from 25 mL of prepared soil extracts after centrifugation. Quantification of total heterotrophs, fecal coliforms, *E. coli,* and fungi were performed with tryptic soy agar, MacConkey agar, tryptone bile X-glucuronide agar (Neogen, Lansing, MI), and potato dextrose agar (BD, Franklin Lakes, NJ), respectively. Appropriate dilutions of soil extracts were spread plated in a volume of 0.1 mL on the agar plates and incubated at 37°C for 48 h for total heterotrophs, 42°C for 24 h for fecal coliforms, 37°C for 24 h for *E. coli,* and 25°C for 5 days for fungi (yeasts and molds). All CFUs were counted on tryptic soy agar and potato dextrose agar for total heterotrophs and fungi, respectively. Pink or red colonies on MacConkey agar and blue and green colonies on tryptone bile X-glucuronide agar were counted as fecal coliforms and *E. coli,* respectively.

Chemical analyses of soil extract. Soil extracts were sent to University of Delaware Soil Testing Program (Newark) to determine pH and to quantify other nutrients, such as total carbon, total organic carbon, P_2O_5, NH_4-N, NO_3-N, and trace elements.

Strain preparation and inoculation of extracts. A rifampin-resistant *Salmonella enterica* subsp. *enterica* serovar Newport WT strain was obtained from the USDA-ARS Environmental Microbial and Food Safety Laboratory (Beltsville, MD) and has been described previously *(9)*. An *rpoS*-deficient, kanamycin-resistant *Salmonella* Newport strain was constructed using the λ-red recombination method *(14),* as previously described *(19)*. These strains were stored at –80°C and were streaked separately for isolation on xylose lysine Tergitol 4 (XLT4) agar plates (Neogen) containing 80 µg/mL rifampin and 25 µg/mL kanamycin for *Salmonella* Newport WT and Δ*rpoS* strains, respectively. An isolated colony of *Salmonella* Newport WT and Δ*rpoS* strains were transferred to separate 30 mL of tryptic soy broth, supplemented with 80 µg/mL rifampin and 25 µg/mL kanamycin, respectively, and incubated at 37°C for 24 h. After incubation, the overnight bacterial cultures were serially

TABLE 1. *Microbial profile of soil extracts*[a]

Soil extract	Total heterotrophs	Fecal coliforms	*E. coli*	Fungi
UNS	1.30×10^5	3.30×10^4	2.50×10^2	1.70×10^5
US	<10	<1	<1	2.50×10^2
ANS	3.55×10^5	7.30×10^4	6.50×10^1	1.50×10^5
AS	<10	<1	<1	3.50×10^2

[a] All counts are in CFU per milliliter. UNS, unamended, nonsterile; US, unamended, sterile; ANS, amended, nonsterile; AS, amended, sterile.

diluted, and 3 mL of the diluted culture was added to 297 mL of both sterile and nonsterile, amended and unamended soil extracts in sterile flasks for both WT and Δ*rpoS* strains to obtain an initial population of approximately 3 log CFU/mL. These flasks were incubated at 25°C with shaking at 125 rpm.

***Salmonella* Newport populations and determination of growth curves.** To obtain potential survival and growth curves for the different extracts, *Salmonella* Newport populations were quantified at 0, 4, 8, 24, 30, 48, 72, and 96 h after inoculation. At those times, 1 mL of the inoculated extracts was removed from the bottle, and appropriate serial dilutions were prepared in phosphate-buffered saline, which were then plated on XLT4 agar plates containing 80 μg/mL rifampin and 25 μg/mL kanamycin for WT and Δ*rpoS Salmonella* Newport strains, respectively. Those plates were incubated at 37°C for 24 h, and black colonies were counted. Plate counts were used to determine populations (CFU per milliliter), which were log transformed, and those values were used to generate survival curves for *Salmonella* Newport in each extract type. Growth curves were fit using the Baranyi model in R software (version 3.4.2, R Foundation for Statistical Computing, Vienna, Austria) using the nlsmicrobio package *(3, 26)* with the equation [$y(t) = \ln x(t)$], where $x(t)$ is the cell concentration (in CFU/mL) at time (t), and $y(t)$ is the growth rate.

Statistical analysis. Growth experiments were arranged as a completely randomized design of a complete factorial combination of two strains (WT and Δ*rpoS Salmonella* Newport) and four extract types: (i) unamended, nonsterile (UNS); (ii) unamended, sterile (US); (iii) amended, nonsterile (ANS); and (iv) amended, sterile (AS). Treatments were replicated four times. The obtained parameters lag phase (λ), growth rate (μ_{max}), and maximum cell density (N_{max}) were treated as dependent variables, whereas the strains and types of soil extract were treated as independent variables. Two-way analysis of variance (ANOVA) was conducted in SAS software (version 9.4, SAS Institute, Cary, NC) using proc GLIMMIX, and corrections for multiple comparisons were performed with "tukey = adj" at an adjusted P value of 0.05 for significant interactions.

RESULTS

Microbial and chemical characteristics of soil extracts. Abundance of total heterotrophs, fecal coliforms, *E. coli,* and fungi were similar among HTPP-amended and UNS soil extracts, which shows that bacterial loads were not affected by the addition of HTPP (Table 1). To study the effect of the presence of indigenous microorganisms on *Salmonella* Newport survival, the nonsterile soil extracts were filter sterilized, leading to a substantial decrease in the counts of total heterotrophs, fecal coliforms, *E. coli,* and fungi in sterile soil extracts (Table 1). Chemical nutrients and pH values of UNS and US extracts were similar (Table 2). The same trend was observed for amended extracts (Table 2). However, nutrient and pH levels differed substantially between unamended and amended soil extracts. The pH of both UNS (6.40) and US (6.50) soil extracts were lower than those of ANS (7.39) and AS (7.36) soil extracts. The P_2O_5 in ANS (116.13 mg/L) and AS (118.97 mg/L) soil extracts were greater than those of UNS (2.27 mg/L) and US (3.67 mg/L) soil extracts. Similarly, NH_4-N was greater in ANS (118.18 mg/L) and AS (64.30 mg/L) soil extracts compared with UNS (0.43 mg/L) and US (0.42 mg/L) soil extracts. NO_3-N levels were found to be greater in unamended soil extracts compared with amended soil extracts. However, all forms of carbon, such as total inorganic carbon, total carbon, and total organic carbon, were present in greater concentrations in amended soil extracts than they were in unamended soil extracts (Table 2). All other trace elements, such as Al, B, Ca, Cu, Fe, K, Mg, Mn, Na, P, S, and Zn, were also found in higher concentrations in amended soil extracts than they were in unamended soil extracts. Observations for all nutrients were found to be similar between sterile and nonsterile soil extracts (Table 2).

Survival and growth of *Salmonella* Newport WT. Mean WT *Salmonella* Newport populations at 0 h ranged from 3.07 to 3.23 log CFU/mL for UNS, US, ANS, and AS extracts (Fig. 1). In UNS, an appropriate growth curve could not be fit with the Baranyi model, but a maximum population of 4.16 ± 1.45 log CFU/mL was observed at

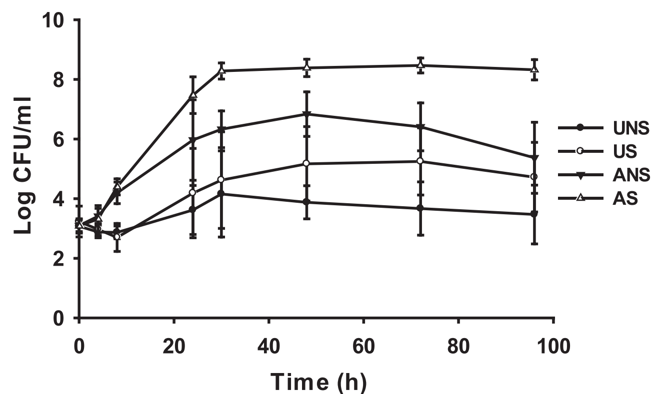

FIGURE 1. *Growth and survival of* Salmonella *Newport WT in soil extracts: unamended, nonsterile (UNS); unamended, sterile (US); amended, nonsterile (ANS); and amended, sterile at 25°C from four experimental replicates. Error bars indicate standard deviations.*

TABLE 2. *Chemical characteristics of soil extracts*

	Unamended soil extract		Amended soil extract	
Parameter[a]	Nonsterile	Sterile	Nonsterile	Sterile
pH	6.4	6.5	7.39	7.36
P_2O_5	2.27[b]	3.67	116.13	118.97
NH_4-N	0.43	0.42	118.18	64.3
NO_3-N	7.96	4.45	2.67	3.89
TIC	1.44	0.26	15.02	12.2
TC	33.15	40.04	1,270.65	988.2
TOC	31.71	39.78	1,255.63	976
Al	0.33	0.09	0.37	0.27
B	0.01	0.01	0.7	0.63
Ca	4.24	6.79	41.07	37.87
Cu	0.01	0.02	1.96	1.73
Fe	0.17	0.1	2.78	2.56
K	4.73	8.59	410.92	385.64
Mg	1.26	2.08	36.07	37.67
Mn	0.09	0.14	0.79	0.84
Na	0.52	0.8	162.89	151.05
P	0.99	1.6	50.71	51.95
S	0.93	1.34	202.5	182.86
Zn	0.02	0.02	0.61	0.56

[a] TIC, total inorganic carbon; TC, total carbon; TOC, total organic carbon.
[b] All concentrations are presented in milligrams per liter.

30 h in UNS. Significant ($P < 0.05$) increases in cell densities (N_{max}) were observed in other soil extracts within 96 h (Table 3). The highest N_{max} was 8.40 ± 0.31 log CFU/mL in the AS extract, which was significantly greater than that observed in ANS (6.59 ± 0.77 log CFU/mL) and US (6.19 ± 0.97 log CFU/mL) soil extracts ($P < 0.05$) (Table 3). Overall, significantly greater WT *Salmonella* Newport populations (N_{max}) were observed in amended soil extract than they were in the unamended soil extract ($P < 0.05$). In addition, sterile soil extracts supported a greater N_{max} than the nonsterile soil extracts did for WT *Salmonella* Newport populations ($P < 0.05$). Similarly, growth rates (μ_{max}) were estimated to be 0.60 ± 0.17, 0.48 ± 0.12, and 0.53 ± 0.07 h^{-1} for US, ANS, and AS, respectively, which were not significantly different from each other ($P > 0.05$). However, the λ observed in US extract was estimated to be 20.17 h, which was significantly greater than those observed for AS (2.86 ± 1.39 h) and ANS (2.89 ± 0.66 h) soil extracts ($P < 0.05$) (Table 3).

Survival and growth of Δ*rpoS Salmonella* Newport. The mean Δ*rpoS Salmonella* Newport population at 0 h ranged from 2.76 to 2.86 log CFU/mL for UNS, US, ANS, and AS extracts (Fig. 2). Similar to the WT *Salmonella* Newport strain, an appropriate growth curve could not be fit with the Baranyi model for the UNS soil extract for *Salmonella* Newport Δ*rpoS* strain, and a decrease in N_{max} was observed over time, with a density of 1.59 ± 0.41 log CFU/mL observed at 96 h for UNS soil extract. In contrast, N_{max} observed in US, ANS, and AS soil extracts were significantly ($P < 0.05$) greater than those observed in UNS. The highest N_{max} was 7.09 ± 0.16 log CFU/mL for

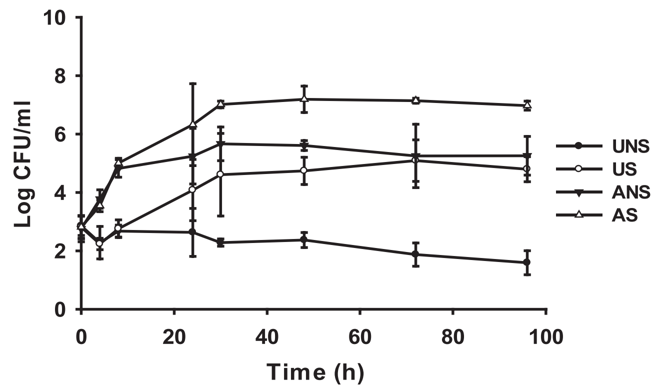

FIGURE 2. *Growth and survival of* Salmonella *Newport Δ*rpoS *in soil extracts: unamended, nonsterile (UNS); unamended, sterile (US); amended, nonsterile (ANS); and amended, sterile at 25°C from four experimental replicates. Error bars indicate standard deviations.*

the AS extract, which was significantly greater than that of the ANS (5.29 ± 0.63 log CFU/mL) and US (4.76 ± 0.46 log CFU/mL) extracts ($P < 0.05$) (Table 3). The AS soil extracts had greater N_{max} than did the UNS soil extracts ($P < 0.05$). The maximum N_{max} for *Salmonella* Newport Δ*rpoS* strain in each of those extracts was significantly lower than that observed for *Salmonella* Newport WT ($P < 0.05$) (Table 3).

For the Δ*rpoS Salmonella* Newport strain, the μ_{max} were observed to be 0.65 ± 0.09 and 0.61 ± 0.19 h^{-1} in UNS and AS soil extracts, respectively, which were not significantly different from each other ($P > 0.05$). The μ_{max} for Δ*rpoS Salmonella* Newport strain in US soil extract was observed to be 0.29 ± 0.11 h^{-1}, which was significantly lower than in UNS and AS extracts ($P < 0.05$) (Table 3). No significant differences were observed between WT and Δ*rpoS Salmonella* Newport strains for μ_{max} ($P > 0.05$). Conversely, no λ was observed for the Δ*rpoS Salmonella* Newport strain, except in the US soil extract. The observed λ for *Salmonella* Newport Δ*rpoS* strain in the US soil extract was 20.04 ± 12.25 h, which was similar to the λ for *Salmonella* Newport WT (20.17 ± 8.79 h) in the US soil extract ($P > 0.05$).

DISCUSSION

Manure runoff from animal farms has been implicated in previous outbreaks associated with produce commodities *(2)*. Cattle manure and poultry litter can contain harmful pathogens, and *Salmonella* has been shown to survive for a prolonged period in those matrices *(6, 8)*. If manure runoff can support the growth of *Salmonella* spp., there is increased likelihood for longer environmental persistence leading to elevated risk of subsequent transfer to fresh fruits and vegetables. In this study, soil extracts amended with HTPP supported growth of both WT and Δ*rpoS Salmonella* Newport strains with an increase in cell densities of 4 to 5 log CFU/mL over 96 h. However, in UNS soil extracts, *Salmonella* Newport WT populations remained similar over 96 h. The increased levels of nutrients from the addition of HTPP promoted higher N_{max} compared with that of the

TABLE 3. *Growth parameters of WT and Δ*rpoS Salmonella *Newport strains in soil extracts using the Baranyi model*

Strain	Soil extract	Lag phase (λ)	μ_{max} (h^{-1})	N_{max} (log CFU/mL)
WT	UNS	NA[a]	NA	NA
	US	20.17 ± 8.79 B[b]	0.60 ± 0.17 AB	6.19 ± 0.97 AC
	ANS	2.89 ± 0.66 A	0.48 ± 0.12 AB	6.59 ± 0.77 A
	AS	2.86 ± 1.39 A	0.53 ± 0.07 AB	8.40 ± 0.31 B
Δ*rpoS*	UNS	NA	NA	NA
	US	20.04 ± 12.25 B	0.29 ± 0.11 B	4.76 ± 0.46 C
	ANS	—[c]	0.65 ± 0.09 A	5.29 ± 0.63 AC
	AS	—	0.61 ± 0.19 A	7.09 ± 0.16 AB

[a] NA, model could not be generated with the Baranyi model.
[b] Within each column, different letters indicate significantly different values at $P < 0.05$.
[c] —, no parameters were estimated.

unamended extracts. Similarly, AS soil extracts supported higher N_{max} than the ANS soil extracts did, which may have been influenced by competition for available nutrients between *Salmonella* Newport and indigenous microorganisms. *Salmonella* Newport Δ*rpoS* strain in AS soil extracts showed no measurable λ and a similar μ_{max} compared with the WT strain; however, a lower N_{max} for the Δ*rpoS* strain compared with the WT strain was observed. In UNS soil extracts, *Salmonella* Newport Δ*rpoS* populations declined by 1 log CFU/mL over 96 h, whereas no decrease in the *Salmonella* Newport WT population was observed. This indicates that a functional *rpoS* may aid growth of *Salmonella* Newport in amended soil extract. Importantly, populations of the Δ*rpoS* strain declined over 96 h in unamended soil extracts, whereas WT *Salmonella* Newport strain did not decline and survived during that period, showing that RpoS improved *Salmonella* Newport survival under those nonhost conditions. RpoS supports the survival of *Salmonella* spp. and *E. coli* O157:H7 in nonhost environments, including in aged broiler litter under desiccation *(5)*. Furthermore, *E. coli* O157:H7 strains with a mutation in *rpoS* had shorter survival durations compared with the WT strains in manure-amended soil *(25)*. Taken together, these studies and our data indicate that RpoS has an important role in improved survival of *Salmonella* Newport in soil extracts.

In our study, growth characteristics for both *Salmonella* Newport WT and *rpoS*-deficient strains were greater in sterile than in nonsterile soil extracts. Similar observations were made for *Salmonella* spp. in sterile and nonsterile biosolids *(22)*. In the latter study, a lower N_{max} and reduced μ_{max} was observed in nonsterile biosolids compared with that in sterile biosolids, which indicated that competition from indigenous microorganisms for available nutrients limited the growth of *Salmonella* Newport. Other studies investigated growth of *Salmonella* and *E. coli* O157:H7 in amended soil extracts. *Salmonella* Enteritidis phage type 30 populations increased by up to 3 log CFU in soil prepared with almond hull extract *(7)*. Populations of *Salmonella* spp. in various composts types (biosolids, manure, and yard wastes) increased by 1.5 log CFU during storage at 25°C over 3 days *(16)*. Growth of *Salmonella* spp. in autoclaved composts with 40 and 50% moisture content supported up to a 4-log increase in *Salmonella* counts within 3 days of storage *(11)*. These studies show that *Salmonella* spp. may grow in biological soil amendments or amended soil extracts given sufficient nutrients and a lack of microbial competition.

Our study shows that *Salmonella* Newport growth in soil extracts is affected by the nutrients present and that nutrients from HTPP resulted in increased populations of both WT and *rpoS*-deficient *Salmonella* Newport strains. The *rpoS* status of *Salmonella* Newport also affected the maximum population attained, indicating that a functional *rpoS* gene may promote higher population levels of *Salmonella* Newport in soil extracts and amended soil runoff. Examination of other strains, genetic factors, microbial competition factors, and nutrient factors in nonhost environments is required to fully characterize the survival of *Salmonella* Newport in soil and soils containing BSAAOs. Our study demonstrates that *Salmonella* Newport can not only survive but also grow to high populations in unamended or amended soil extracts, which can lead to potential contamination of soils, water sources, and produce commodities.

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