Monophasic Salmonella typhimurium and the pig's life cycle – a longitudinal study

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Introduction: With the worldwide rise of multidrug resistant bacteria, the assessment of control points where the chain of transmission from food animal to the human consumer can be broken is a key point in developing strategies to help staunch the diffusion of dangerous resistant pathogens through food products[1-3]. The purpose of this study was to characterize the dynamics of monophasic *Salmonella* Typhimurium transmission throughout the pig's productive cycle from birth to slaughter, based on the assessment of prevalence, antimicrobial resistance and clonality throughout the different phases of the cycle.

Methods: Three Portuguese industrial pig farms with similar preventive protocols based on the administration of ceftiofur, apramycin and colistin were included in the study. From each farm 10 litters were picked for sampling through fecal collection, each litter composed by the sow and 7 piglets (Figure 1). The piglets were further sampled 4 times throughout their life cycle until slaughter, where each carcass was then swabbed. Monophasic *S.* Typhimurium strains were isolated and identified according to European Food Safety Agency guidelines[1]. Antimicrobial susceptibility was determined by disk diffusion and MIC determination for 23 antimicrobials, and genetic determinants of antimicrobial resistance associated with the tested antimicrobials were identified by PCR. To assess the clonal relationship of isolates from the same cycle phase and between phases, PFGE was used[4].

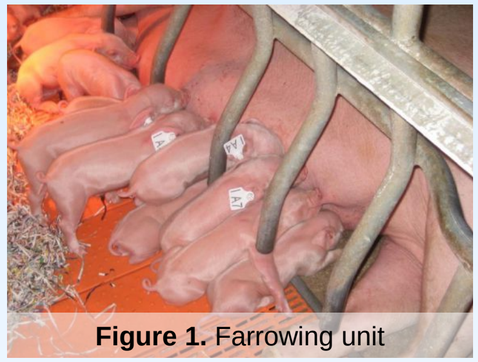

Figure 1. Farrowing unit

Results: Monophasic *S.* Typhimurium was isolated from samples of only one farm (n=43). Prevalence was variable between the sows and the different phases of the piglets' cycle, at birth, after weaning, at the nursery unit, at the finishing unit, before leaving the finishing unit and after slaughter: 70%, 24.3%, 1.5%, 0%, 29.7%, 0% and 0%. The antimicrobial resistance patterns and genes that were detected can be seen in Table 1. Three PFGE types were identified, with type A being the most predominant and the one type that was transmitted across all isolation positive phases. Type B was associated with 8 of the finishing phase isolates, namely with all gentamicin resistant/florfenicol susceptible isolates, and Type C counted only one sow (Table 2).

Age	Isolates	Antimicrobial Resistance Pattern[a]	Antimicrobial Resistance Genes	PFGE type
Sows	5	AKNNaSSuT	aphAI-IAB, bla_{TEM}, strA, strB, sul2, tetB (5)	A (5)
	1	ACGKNeSSxtTTo	aac(3')-IV, aadA, bla_{TEM}, cmlA, dfrA12, sul1, tetA (1)	C (1)
Piglets	14	AKNNaSSuT	aphAI-IAB, bla_{TEM}, sul2, tetB (1)	A (14)
			aphAI-IAB, bla_{TEM}, strA, strB, sul2, tetB (13)	
	1	AKNNaSSuT To	aphAI-IAB, bla_{TEM}, strA, strB, sul2, tetB (3)	A (3)
	2	AAcKNNaSSuT		
Pigs (4 weeks)	1	AKNNaSSuT	aphAI-IAB, bla_{TEM}, strA, strB, sul2, tetB (1)	A (1)
	4	AKNSSuT	aphAI-IAB, bla_{TEM}, strA, strB, sul2, tetB (4)	A (3)
				B (1)
Pigs (14 weeks)	6	AGKNNeSSuTTo	aac(3')-IV, aphAI-IAB, bla_{TEM}, strA, strB, sul2, tetB (7)	B (7)
	1	AGKNNaNe SSuTTo		
	5	ACFGKNNe SSuTTo	aac(3')-IV, aphAI-IAB, bla_{TEM}, floR, strA, strB, sul2, tetB (8)	A (8)
	3	ACFGKNNa NeSSuTTo		

Table 1. Antimicrobial resistance profiles and PFGE types of monophasic *Salmonella* Typhimurium.
[a]A: ampicillin; Ac: amoxicillin/clavulanic acid; C: chloramphenicol; F: florfenicol; G: gentamicin; K: kanamycin; N: neomycin; Na: nalidixic acid; Ne: netilmicin; S: streptomycin; Su: sulphonamides; Sxt: sulphonamides/trimethoprim T: tetracycline; To: tobramycin

Sows	Sow PFGE type	Piglets/pigs PFGE type			
		Farrowing	4 weeks	10 weeks	14 weeks
Sow A	A	A	A		A
Sow B	A	A			A
Sow C	Neg.				A
					B
Sow D	A	A			B
Sow E	Neg.	A			B
Sow F	ND				
Sow G	A				A
Sow H	C				A
					B
Sow I	Neg.				B
Sow J	A				A
					B

Table 2. *Salmonella* 4,[5],12:i:- isolates' PFGE types throughout the production cycle.
Continuous line represents a PFGE Type carryover; dotted line represents a PFGE Type change. Neg., negative to *Salmonella* isolation.; ND, non-monophasic *S.* Typhimurium serovar

Conclusions: We have identified not only the transmission of monophasic *S.* Typhimurium but also the downstream impact of antimicrobial usage throughout the pig's productive cycle, emphasizing the importance of longitudinal studies in finding additional control measures to aid in mitigating the role of production animals as a reservoir for multidrug resistant zoonotic bacteria. More research is needed to understand what triggers the transmission and which factors may act as barriers to monophasic *S.* Typhimurium transmission.

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