Universiteit Utrecht

Faculty of Veterinary Medicine
Institute for Risk Assessment Sciences

Strong dose-response relationship between antimicrobial use and livestock-associated MRSA in pig farming: results from a pragmatic intervention study

Alejandro Dorado García[1,2] | Wietske Dohmen[1] | Marian E.H. Bos[1] | Koen M. Verstappen[2] | Jaap A. Wagenaar[2] | Dick J.J. Heederik[1,3]

1 Utrecht University, Faculty of veterinary Medicine, Institute for Risk Assessment Sciences

2 Utrecht University, Faculty of veterinary Medicine, Department of Infectious Diseases and Immunology

3 University Medical Center Utrecht, Julius Center for Health Sciences and Primary Care

Background

Livestock-associated methicillin resistant *Staphylococcus aureus* (LA-MRSA) of sequence type ST398 is widely spread in pig farms in the Netherlands with around 70% of them testing positive. This animal reservoir poses a public health threat since the resistant bacteria and resistance genes can be transferred to people living or working on farms and they can be introduced in hospitals and the community.

Objectives

- To assess trends in antimicrobial use (AMU) during the study period and its relationship with LA-MRSA.
- To identify other intervention measures aimed at reducing MRSA in pig herds.

Methods

Study design and laboratory analysis

- 36 pig farms in the Netherlands.

Table 1. Number of farms by type of production and external supply of gilts from other farms.

Type of production	External supply of gilts		Total
	CLOSED	*OPEN*	
FARROW-TO-FINISHING (delivering fatteners for slaughter)	11	13	24
MULTIPLIERS (delivering pigs for fattening or gilts)	3	9	12
Total	14	22	36

- Sampling at 0, 6, 12 and 18 months.
- Nasal swabs from 60 animals per farm (10 pools of 6 animals)
- Selective enrichment in MRSA Brilliance™ (Oxoid®) and RT-PCR targeted at mecA and C01 genes to confirm presence of ST398.

Data collection

- Data on AMU per farm: Animal Defined Daily Dosages per year (ADDD/Y) for the 6 months preceding each sampling moment.
- Tailor made intervention protocol focused on: improving personal and farm hygiene, animal contact structure and reduction in AMU.
- Questionnaire at 0, 6, 12 and 18 months.

Data analysis (SAS and R)

In all farms and stratifying by *open/closed* farms and *farrow-to-finish /multipliers*:

I. Variable reduction: logistic regression with PROC GENMOD (GEEs) in the 4 cross-sections. Selection when p<0.2 in at least 2 cross-sections.

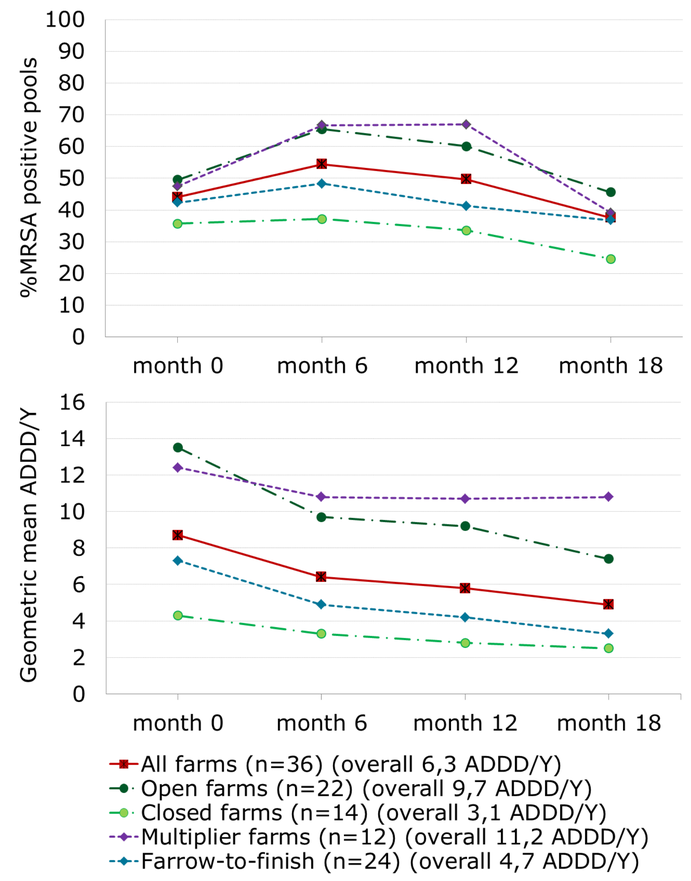
II. Univariate analysis: random intercept non-linear mixed model (PROC GLIMMIX) for a pool to be MRSA positive adjusting for sampling moment (as factor) and age group of the pool.

III. Multivariate analysis: backward elimination of non-significant terms from full model (made of variables with P<0,10 from step II).

Results

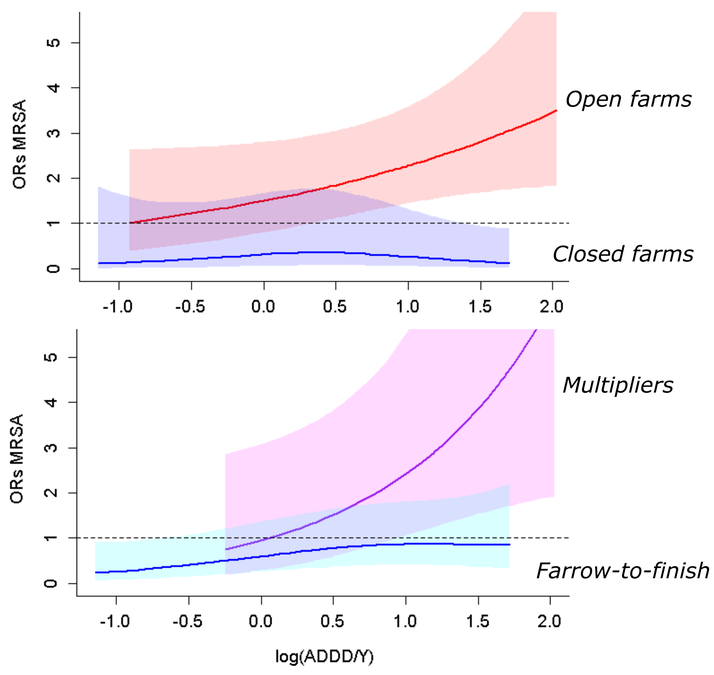
- MRSA prevalence and AMU differed between *open* and *closed* farms, and *multipliers* and *farrow-to-finish* farms.

Figure 1. Slight decrease in MRSA pool-prevalence and significant reduction in AMU over time.



- AMU and MRSA were positively associated (ORs per 5 ADDD/Y increase) in all farms analysis (OR=1,4; P=0,02) and specifically in high AB consumers: open farms (OR=1,3; P=0,12) and multipliers (OR=2,1; P=0,03).

Figure 2. Non-parametric spline (using GAMM in R) for a pool to be MRSA positive (OR) with increasing AMU (log ADDD/Y).



- Over time, the decrease in AMU was associated with a reduction of MRSA-pool prevalence in all farms and *open* farms analysis:
  - Significant interaction term between ADDD/Y and sampling moment (P=0.01, consecutive ORs of 0.7, 1.9, 1.5 and 1.1).

- Univariate analysis in questionnaire items:
  - Significant risk factors in all, *open* and *closed* farms: injection of AB in piglets in the first week, tooth clipping in piglets, vaccination of piglets and/or fatteners.
  - Biosecurity items were found to be protective mainly in *closed* farms: phases of production with different compartments, boarding platform for sows, washing overalls.
- AMU was not a compendium of underlying determinants: pairwise Spearman's rho coefficients between ADDD/Y and reduced set of variables <0,5.

Table 2. Multivariable model for a pool to be MRSA positive (n=1351 pools) in all farms (n=36).

Variable	Category	N	OR	95% CI
Sampling moment	0	342	1.39	0.90-2.13
	6 months	340	2.84	1.86-4.34***
	12 months	338	2.45	1.63-3.69***
	18 months	331	Ref.	-
Age group of pool	Gilts	268	1.03	0.66-1.60
	Finishers	178	3.36	2.04-5.52***
	Suckling piglets	273	3.99	2.59-6.15***
	Weaned piglets	362	7.85	5.12-12.0***
	Sows	270	Ref.	-
Animal external supply	Open	827	6,34	2,06-19,49**
	Closed	524	Ref	-
Delivery room for materials	Yes	1031	0.40	0.22-0.74**
	No	320	Ref.	-
AMU (log10 (ADDD/Y))	Per 5 ADDD/Y	1351	1.42	1.02-1.97*
Hygiene piglet compartment	Disinfection	179	0.41	0.17-0.99*
	Soaking agents	270	2.55	1.17-5.56*
	Soaking & dis.	658	1.17	0.60-2.27
	None	244	Ref.	-

*P<0,05; **P<0,005; ***P<0,0001

Conclusions

- Important differences in prevalence, AMU and selection of variables existed by type of production and by external supply of animals. Approaches for MRSA control should take these differences into account.
- AMU has a strong and positive dose-response relationship with MRSA carriage in pigs especially in farms with high AMU (*open* and/or *multipliers*).
- In addition to the reduction in AMU, we identify other determinants that can define attainable measures for MRSA control (e.g. tooth clipping, increased biosecurity).