

# Associations between plasma anti-Müllerian hormone and fertility responses of seasonally calving grazing dairy cows

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## INTRODUCTION

Antral follicle count is highly variable among young adult cattle, repeatable within individuals, and associated positively with ovarian reserve and fertility traits.

Anti-Müllerian hormone (AMH) is produced by granulosa cells of healthy follicles and has been identified as a marker for antral follicle count and ovarian reserve, and could be used potentially to predict fertility of cattle.

## OBJECTIVE

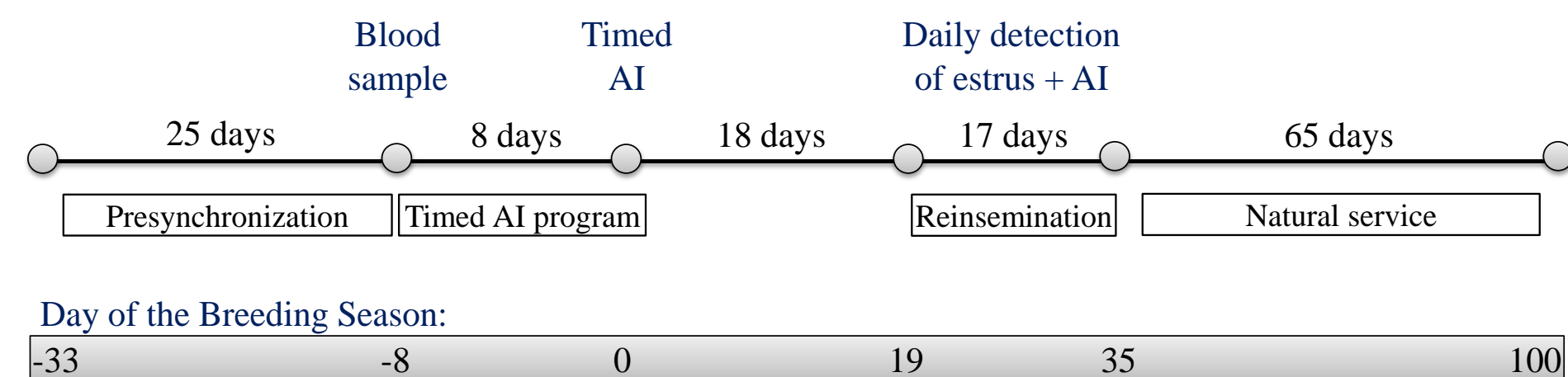
The objective was to investigate associations between plasma AMH and fertility of grazing dairy cows subjected to synchronized AI on the first day of a 100-day breeding season.

## MATERIALS and METHODS

### 100-day Breeding Season:

Timed AI + Estrous Detection + Natural Service

n = 1,237 lactating cows in two grazing dairies with a blood sample collected on study d -8



- ✓ AMH concentrations in plasma were determined using an available commercial kit (AnshLite™ Bovine AMH CLIA, AL-214)
- ✓ Additional 60 lactating cows from one grazing dairy had blood samples collected on days 7 and 15 of the estrous cycle for evaluation of the repeatability of AMH measurements within individuals

## STATISTICAL ANALYSIS

Continuous data were analyzed with models fitting a Gaussian distribution, and binary responses were analyzed by logistic regression fitting a binary data distribution, both using the GLIMMIX procedure of SAS. The interval to pregnancy was analyzed by the Cox proportional hazard model with the PHREG procedure of SAS.

AMH concentrations were analyzed as dependent variables of general independent variables (dairy, lactation number, breed of the cow, days in milk, BCS) and reproductive outcomes.

AMH concentrations were also tested as independent variable for prediction of reproductive outcomes using continues values (1 unit = 100 pg/mL) or quartile categorization (Q1: < 161 pg/mL, LSM = 102.5; Q2: between 160 and 266 pg/mL, LSM = 210.4, Q3: between 265 and 415 pg/mL, LSM = 333.6; Q4: > 414 pg/mL, LSM = 632.7).

## RESULTS

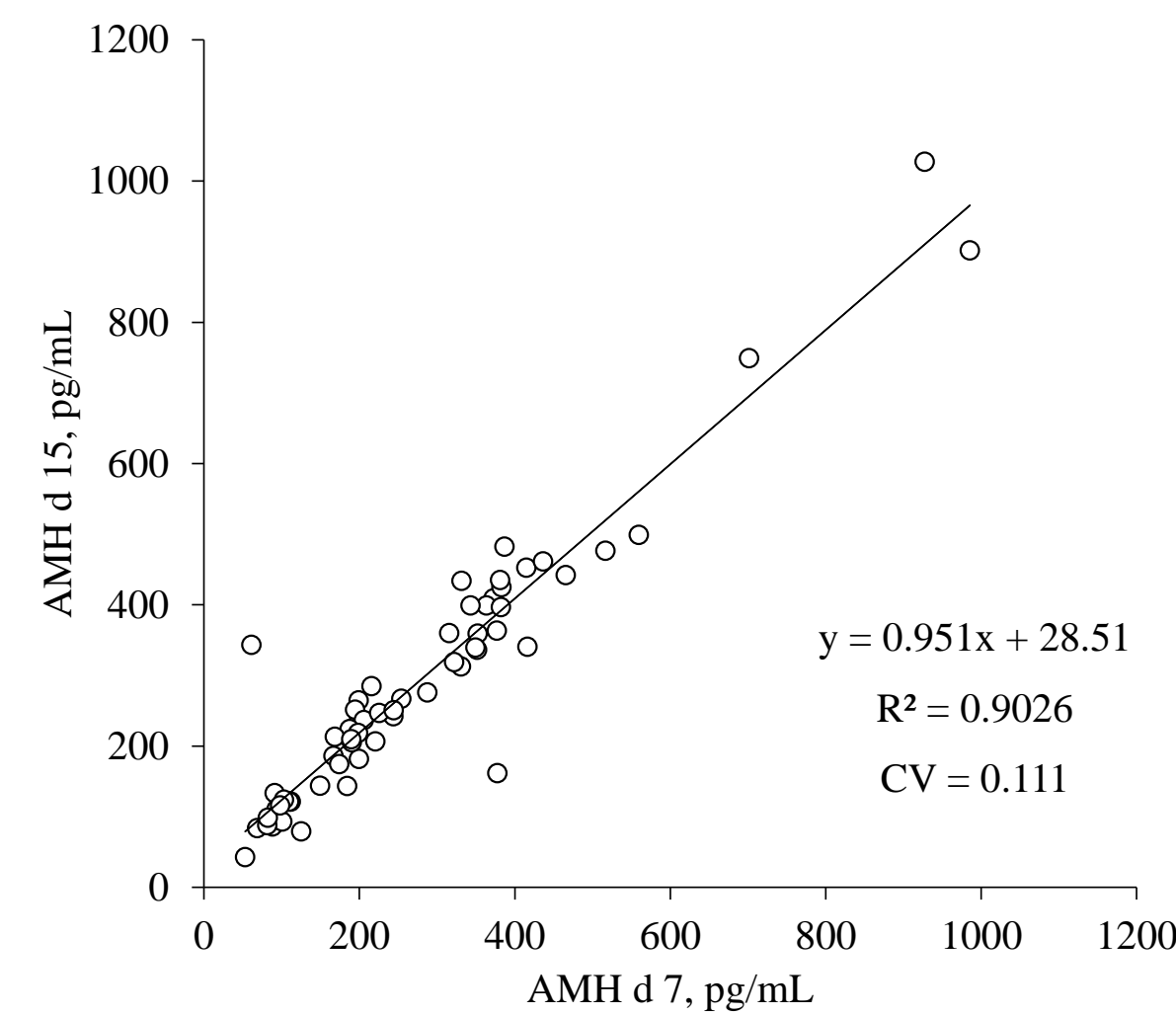
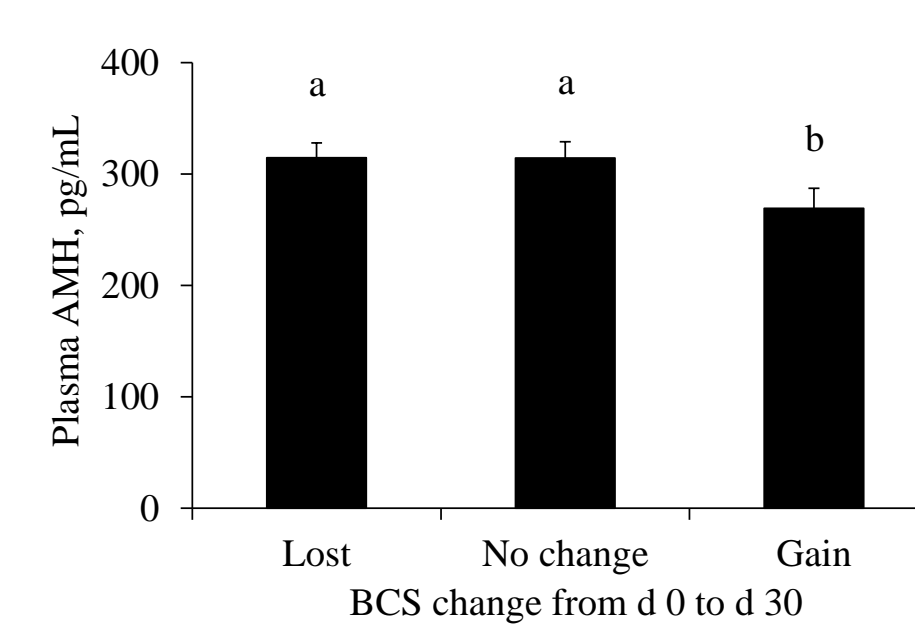
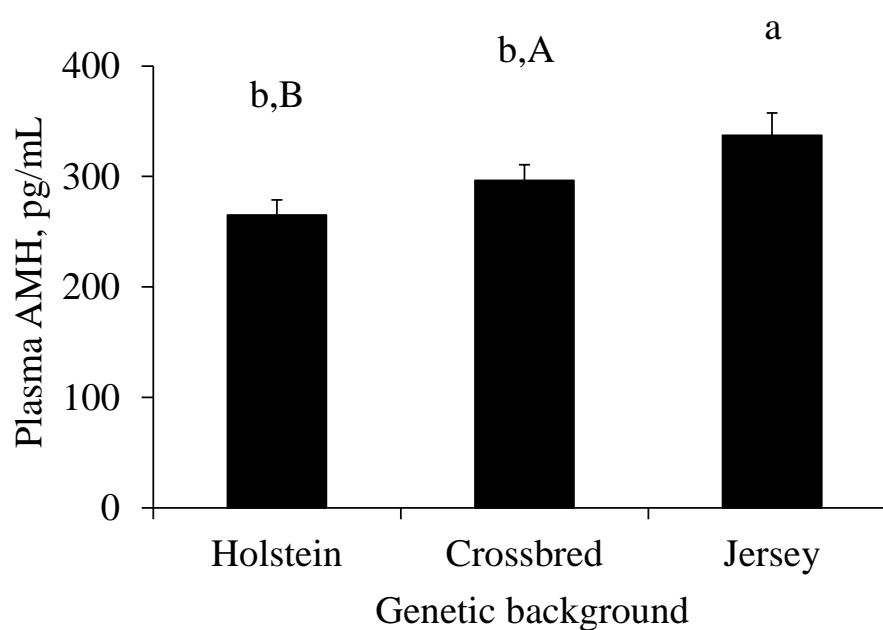
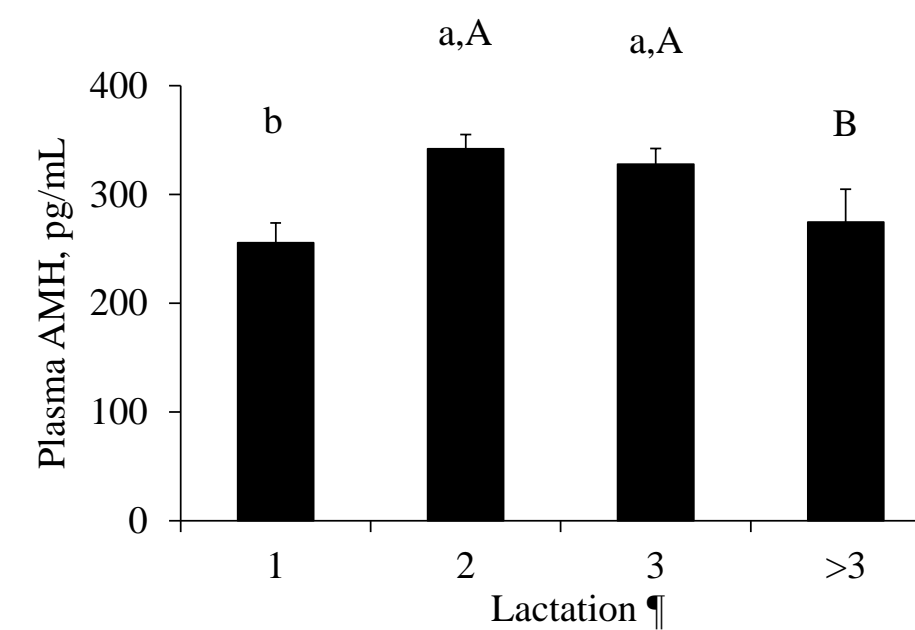


Figure 1. Regression and correlation analyses between plasmatic AMH concentrations on days 7 and 15 of the estrous cycle from 60 lactating grazing dairy cows.

Table 1. Factors affecting AMH concentrations

Independent variables	P value
Dairy	0.99
Breed	<0.01
Lactation	<0.01
Days in milk	0.88
BCS at d 0	0.57
BCS change from d 0 to d 30	0.06



Bars with different letters differs (a,b = P < 0.05; A,B = P < 0.10); ¶ Quadratic effect P < 0.01.

## RESULTS

Table 2. Plasma concentrations of AMH according to reproductive outcomes

Item	Incidence of event		P value
	No	Yes	
	---- AMH concentration, pg/mL ----		
Estrous cyclic	315.6 ± 20.2	294.8 ± 10.6	0.40
Estrus at timed AI	319.6 ± 11.3	269.9 ± 17.7	0.03
Pregnant from timed AI on d 30	303.1 ± 13.0	296.2 ± 14.4	0.75
Pregnant from timed AI on d 64	303.1 ± 12.1	296.0 ± 15.6	0.75
Pregnancy loss	296.0 ± 14.4	303.0 ± 47.7	0.89
Reinseminated	286.7 ± 25.5	311.6 ± 13.6	0.43
Pregnant from reinsemination	282.7 ± 20.9	338.1 ± 22.0	0.10
Pregnant from natural service	251.7 ± 22.2	311.3 ± 22.3	0.09
Pregnant at the end of breeding season	251.7 ± 24.9	310.1 ± 10.0	0.05

Table 3. Continuous values of AMH concentrations as predictor of reproductive outcomes

Item	Univariable analysis		Multivariable analysis	
	OR (95% CI)	P	AOR (95% CI)	P
Estrous cyclic	0.99 (0.94 - 1.04)	0.64	0.95 (0.90 - 1.01)	0.08
Estrus at timed AI	0.95 (0.91 - 1.01)	0.07	0.93 (0.28 - 0.85)	0.01
Pregnant from timed AI on d 30	0.99 (0.94 - 1.03)	0.54	0.97 (0.93 - 1.02)	0.29
Pregnant from timed AI on d 65	0.99 (0.95 - 1.04)	0.76	0.98 (0.93 - 1.03)	0.37
Pregnancy loss	0.97 (0.86 - 1.09)	0.60	0.97 (0.87 - 1.09)	0.59
Reinseminated	1.02 (0.95 - 1.10)	0.54	0.99 (0.92 - 1.07)	0.86
Pregnant from reinsemination	1.06 (0.98 - 1.14)	0.18	1.07 (0.99 - 1.16)	0.10
Pregnant from natural service	1.08 (1.00 - 1.17)	0.05	1.07 (0.99 - 1.16)	0.10
Pregnant at the end of season	1.07 (1.00 - 1.15)	0.05	1.03 (0.97 - 1.11)	0.34

Table 4. Quartile categorization of AMH concentrations as predictor of reproductive outcomes

Item	Quartile of AMH concentration				P	Linear	Quad
	Q1	Q2	Q3	Q4			
Estrous cyclic	78.6 <sup>AB</sup>	78.4 <sup>AB</sup>	80.4 <sup>A</sup>	73.8 <sup>B</sup>	0.32	0.31	0.21
Estrus at timed AI	42.2 <sup>a</sup>	34.1 <sup>b</sup>	34.3 <sup>b</sup>	29.3 <sup>b</sup>	0.01	<0.01	0.65
Pregnant from timed AI on d 30	51.6	49.6	49.5	47.4	0.80	0.34	0.99
Pregnant from timed AI on d 65	40.0	40.3	41.5	37.1	0.73	0.55	0.41
Pregnancy loss	14.2 <sup>a</sup>	9.2 <sup>ab</sup>	6.5 <sup>b</sup>	11.2 <sup>ab</sup>	0.17	0.29	0.05
Reinseminated	72.0	75.3	71.1	72.1	0.85	0.81	0.74
Pregnant from reinsemination	33.4 <sup>B</sup>	40.6 <sup>AB</sup>	40.8 <sup>AB</sup>	45.0 <sup>A</sup>	0.39	0.10	0.72
Pregnant from natural service	59.9 <sup>b</sup>	58.3 <sup>b</sup>	68.2 <sup>ab</sup>	74.2 <sup>a</sup>	0.05	<0.01	0.37
Pregnant at the end of season	84.9 <sup>B</sup>	85.2 <sup>B</sup>	88.1 <sup>AB</sup>	89.5 <sup>A</sup>	0.22	0.04	0.72

Different superscripts differ (<sup>a,b</sup> = P < 0.05; <sup>AB</sup> = P < 0.10) within the same column

## RESULTS

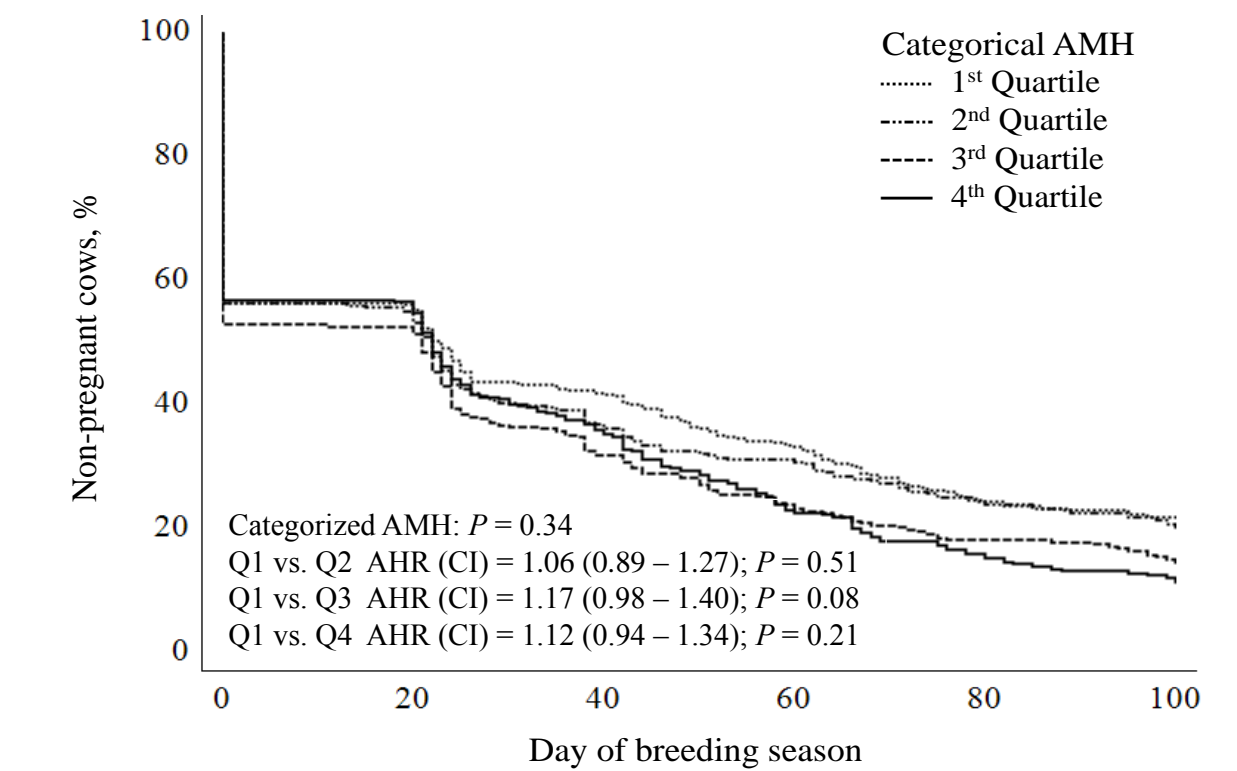


Figure 2. Survival probability of pregnancy throughout the breeding season.

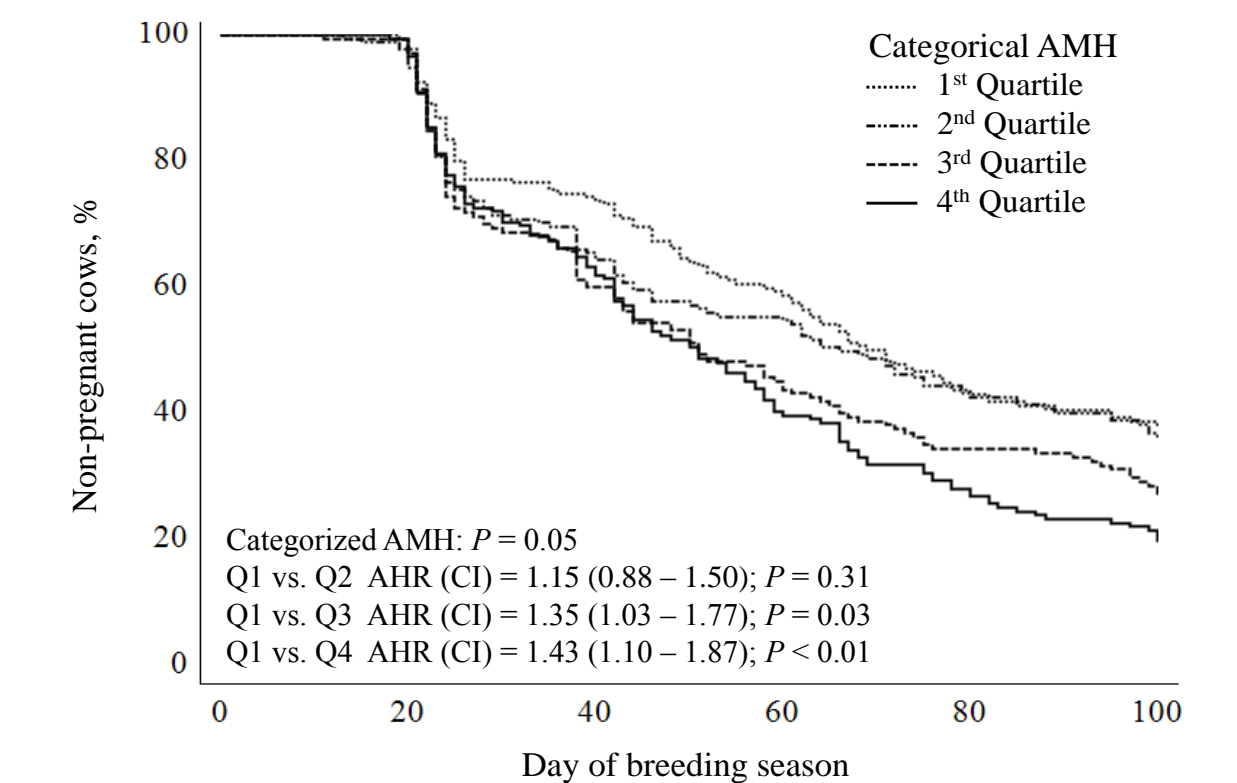


Figure 3. Survival probability of pregnancy throughout the breeding season for cows not pregnant from timed AI on the first day of the breeding season.

## SUMMARY

AMH concentrations in plasma were affected by lactation, genetic background of the cow and change in body condition score.

High concentrations of AMH were associated with reduced expression of estrus at timed AI but did not affect expression of spontaneous estrus for reinsemination.

AMH concentrations were not associated with fertility of cows subjected to timed AI with a synchronized ovulation but were associated positively with fertility of cows that failed to become pregnant from timed AI and were subsequently bred after spontaneous estrus.

Synchronization of ovulation might override any association of AMH and fertility in lactating dairy cows.