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PLASMA ANTI-MÜLLERIAN HORMONE ALLOWS REPRODUCTIVE SELECTION OF DONORS WITH GREATER POTENTIAL OF EMBRYO PRODUCTION

Niveles plasmáticos de hormona anti-mülleriana permite la selección de donadoras con alto potencial de producción de embriones

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RESUMEN

Las tecnologías reproductivas, tales como recuperación de ovocitos aspiración folicular (OPU) y producción de embriones in vitro (IVEP), son herramientas tecnológicas muy importantes para el progreso genético asociado con la eficiencia reproductiva. La población de folículos antrales puede afectar directamente el éxito de aspiración folicular y puede ser utilizada como una característica para seleccionar los ovocitos de donadoras de ovocitos con un gran potencial reproductivo que pudiera ser usado en programas de aspiración folicular y producción de embriones in vitro. Aunque es altamente repetible en el animal, la población e folículos antrales es extremadamente variable entre individuos. Por lo tanto, investigación en marcadores endocrinos consistentes y confiables para predecir la población de folículos antrales en bovinos, ha sido el objetivo de varios estudios recientes. En este contexto, los niveles plasmáticos de hormona antimulleriana (AMH) que se correlaciona con el tamaño de la población de folículos antrales y tengan el potencial de ser un marcador endocrino, pueden ayudar a identificar vacas donantes con mayor número de folículos disponibles para aspiración folicular (OPU), mayor respuesta superovulatoria y, en consecuencia incremento en la producción de embriones *in vivo e in vitro*. Esta revisión resume la información más reciente en relación con la población de folículos antrales y su relación con la AMH, y la posibilidad de utilizar AMH como un marcador para la selección de las vacas donantes que se inician los programas de reproducción para difundir la genética y mejorar la fertilidad en el bovinos

Palabras clave: AMH, folículos, embriones, bovinos

ABSTRACT

Reproductive technologies, such as ovum-pick-up (OPU) and in vitro embryo production (IVEP) are important tools to accelerate genetic gain associated with reproductive efficiency. The antral follicular population (AFP) can directly affect the success of OPU and may be used as a characteristic to select the oocyte donor cows with greater reproductive potential that will be used in OPU-IVEP programs. Although highly repeatable within animal, the AFP appears to be extremely variable across individuals. Thus, the investigation of consistent endocrine markers that can trustable predict AFP in cattle has been targeted in several recent studies. In this context, the plasmatic levels of anti-Müllerian hormone (AMH) have been reported to be correlated with the size of AFP and it has the potential of being an endocrine marker to help the identification of donor cows with greater number of follicles available for OPU, greater superovulation response and, consequently, increased in vivo and in vitro embryo production in cattle. This review summarizes recent information regarding AFP and its relationship with AMH, and the possibility of using AMH as a marker for the selection of donor cows that will start reproductive programs to disseminate genetics and eventually to enhance fertility in cattle.

Keywords: AMH, follicle, embryo, bovine

INTRODUCTION

The development of reproductive biotechnologies is strongly driven by the need of the breeding industry to increase genetic gain and pregnancy in dairy and beef cattle. The advances on ovum-pick-up (OPU) and in vitro embryo production (IVEP) enabled the increase on the number of offspring from genetically valuable animals. However, the success of OPU-IVEP is greatly dependent on individual ovarian characteristics that may influence the number and quality of the oocytes that are retrieved (Gandolfi *et al.*, 1998; Kastrop *et al.*, 1991; Lonergan *et al.*, 1994; Pavlok *et al.*, 1992; Tan and Lu, 1990; Wise, 1987). It is known, for example, that the number of antral follicles in the early follicular phase directly correlates with ovarian reserve (Frattarelli *et al.*, 2000). Indeed, the antral follicular population (AFP) directly represents the follicle cohort in the ovaries, which is associated with the number of oocytes retrieved for IVEP. For that reason, AFP is been accepted as a direct marker of the recruitable follicular cohort.

Ovarian AFP is positively related with several indirect measures of fertility in cattle such as ovarian function (Ireland *et al.*, 2009, 2008; Jimenez-Krassel *et al.*, 2009), superovulation responses (Cushman *et al.*, 1999; Kawamata, 1994; Singh *et al.*, 2004), IVEP (Pontes *et al.*, 2009; Taneja *et al.*, 2000), fertility (Erickson *et al.*, 1976; Maurer and Echternkamp, 1985; Mossa *et al.*, 2012) and herd longevity (Jimenez-Krassel *et al.*, 2015). Remarkably, AFP has also been associated to several blood compounds, comprising circulating concentrations of insulin, insulin-like growth factor I (IGF-1), and anti-Müllerian hormone [AMH; (Alvarez *et al.*, 2000; Batista *et al.*, 2014; Fortune *et al.*, 2010; Sales *et al.*, 2015; Satrapa *et al.*, 2013)].
AMH is a dimeric glycoprotein member of the TGFβ superfamily of growth factors synthesized from granulosa cells of preantral and small antral follicles (growing follicles up to the antral stage or to a diameter of approximately 6 mm) and represents the indirect activity of the follicular pool (Cate *et al.*, 1986; Durlinger *et al.*, 1999; Grootegoed *et al.*, 1994; Weenen *et al.*, 2004). In cattle, circulating AMH concentration can help veterinarians to predict AFP in ovaries (Batista *et al.*, 2014a, Ireland *et al.*, 2008, Rico *et al.*, 2009), response to superovulation treatments (Monniaux *et al.*, 2010a, 2010b; Rico *et al.*, 2009; Souza *et al.*, 2015), and more recently as a marker to predict IVEP performance of *Bos taurus* (Gamarra *et al.*, 2015; Guerreiro *et al.*, 2014; Vernunft *et al.*, 2015) and *Bos indicus* breeds (Guerreiro *et al.*, 2014).

Recently, it was indicated that cows with lower AFP have lower fertility (Mossa *et al.*, 2012). Thus, because circulating AMH is an indirect measure of the size of the ovarian follicle pool, later studies have investigated the use of AMH to predict field fertility in cattle (Jimenez-Krassel *et al.*, 2015; Ribeiro *et al.*, 2014). Nevertheless, the significance of AMH on predicting cows fertility may vary according to the reproductive management employed in the farm, since it appears that AMH is only associated to fertility in cows bred following estrus detection and not in those bred after timed AI (TAI) protocols (Ribeiro *et al.*, 2014). Therefore, the present review target to discuss some key points related to AMH and AFP, OPU-IVEP, superovulation responses, in vivo embryo production and fertility.

Anti-Müllerian hormone concentration, antral follicular population and in vitro embryo production in Bos indicus and Bos taurus cattle

A large variability of AFP is reported among different cows, however AFP count is highly repeatable within animal (Burns *et al.*, 2005; Ireland *et al.*, 2007), and AMH can be considered a reliable endocrine marker of ovarian reserve (Ireland *et al.*, 2007, 2008; Monniaux *et al.*, 2012). Our group recently conducted a series of studies aiming to determine the relation between AMH and AFP in different genetic groups with great importance for beef and milk production. Nelore cattle are the major breed raised in tropical areas such as Brazil and have a huge economic importance for the beef industry. As for milk production, Holstein is the predominant breed used in intensive systems while Gir is the predominant breed used in extensive systems in Brazil. Some production systems are also specialized on producing milk from Buffalo cows. In this context, four studies were performed by our group aiming to investigate if AMH was correlated with AFP in cattle of different genetic groups, *Bos indicus*, *Bos taurus* and *Bubalus bubalis* (Baldrighi *et al.*, 2014; Batista *et al.*, 2016, 2014; Guerreiro *et al.*, 2014).

In the first study (Baldrighi *et al.*, 2014), despite the high variability in AFP between individuals within each genetic background, the AFP count was greater in Gir (*Bos indicus*) heifers than in Holstein (*Bos taurus*) and Murrah (*Bubalus bubalis*) heifers (P = 0.01; Fig. 1). Similarly, AMH concentration was lower (P < 0.01) for Holstein and Murrah heifers than for Gir heifers. A positive relationship between AFP and plasmatic AMH concentration was observed for the three genetic groups studied (Fig. 2).

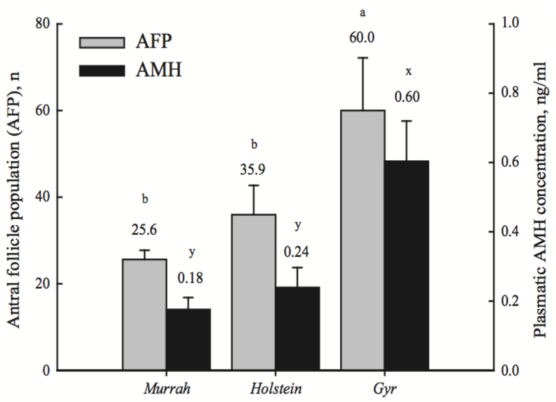

Figure 1. Number of antral follicle population (AFP) and plasma anti-Müllerian hormone (AMH) concentration in Murrah (Bubalus bubalis; n = 13), Holstein (*Bos taurus*; n = 15) and Gir (*Bos indicus*; n = 10) heifers. Data are shown as the means ± SEM. Different letters within columns of the same color are significantly different [AFP: a≠b; P = 0.01 and AMH concentration: x≠y; P < 0.001]. Baldrighi *et al.*, 2014.

The second study (Batista *et al.*, 2014), supported the results previously reported (Baldrighi *et al.*, 2014). The AFP (P < 0.05) and the AMH concentration (P < 0.0001) were also higher in Nelore (*Bos indicus*) than in Holstein (*Bos taurus*) heifers (Fig. 3), and they were correlated (Fig. 4). Additionally, when heifers were classified as to have high or low AFP (based on the mean AFP within each genetic group), high-AFP heifers had greater (P < 0.0001) AMH concentration than low-AFP heifers, regardless of the genetic group.

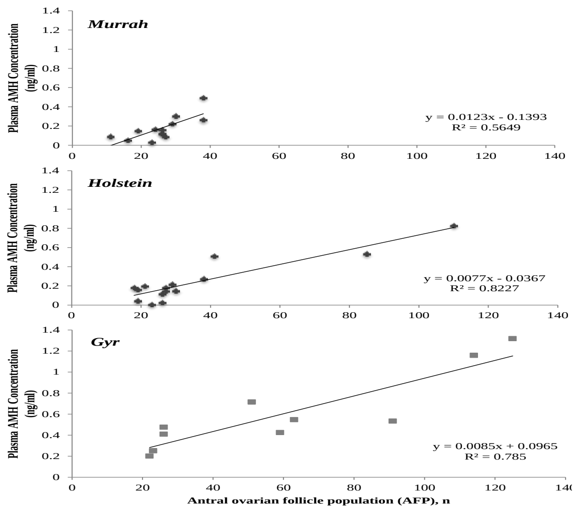

Figure 2. Relationship between antral follicle population (AFP) and plasma anti-Müllerian hormone (AMH) concentration in Murrah (*Bubalus bubalis*, n = 13), Holstein (*Bos taurus*, n = 15) and Gyr (*Bos indicus*, n = 10 heifers). Baldrighi *et al.*, 2014.

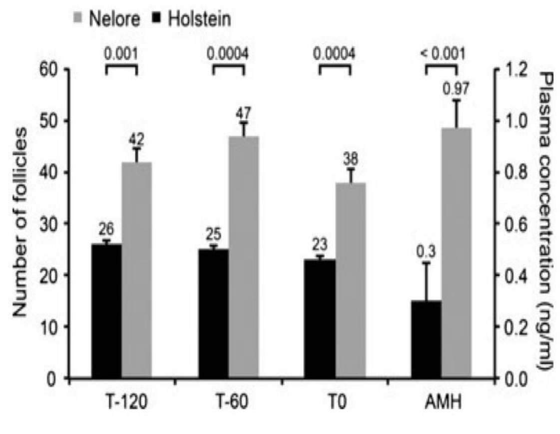

Figure 3. Number of antral follicle population (AFP) and plasma anti-Müllerian hormone (AMH) concentration in Nelore (*Bos indicus*; n = 16) and Holstein (*Bos taurus*; n = 16) heifers. Data are shown as the means ± SEM. Batista *et al.*, 2014.

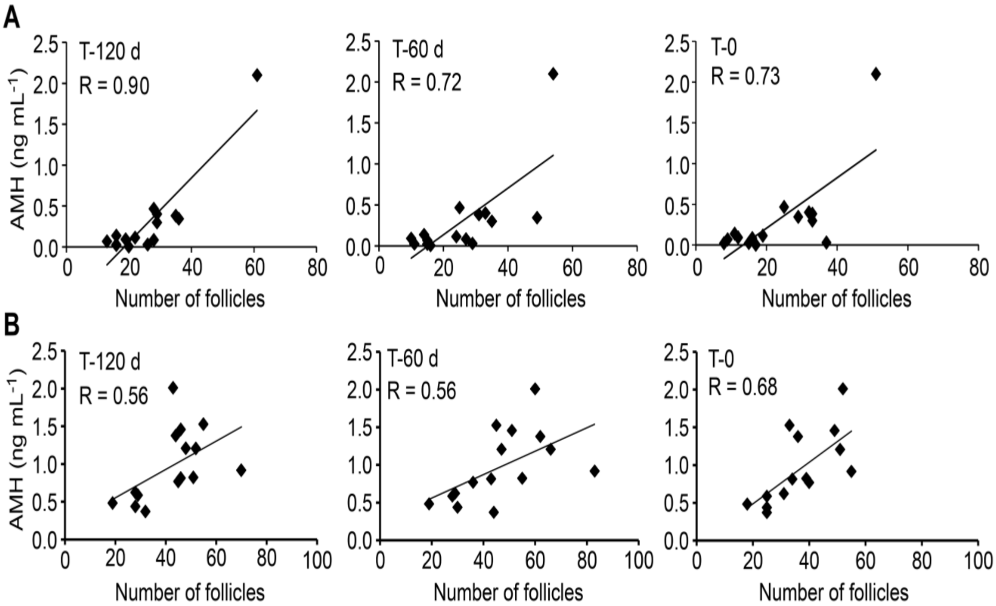

Figure 4. Relationship between the numbers of antral follicles counted 120 (T-120) or 60 (T-60) hours previous or at (T0) AMH dosage, and plasma AMH concentration in Holstein (n=16; A) and Nelore (n=16; B) heifers. Batista *et al.*, 2014.

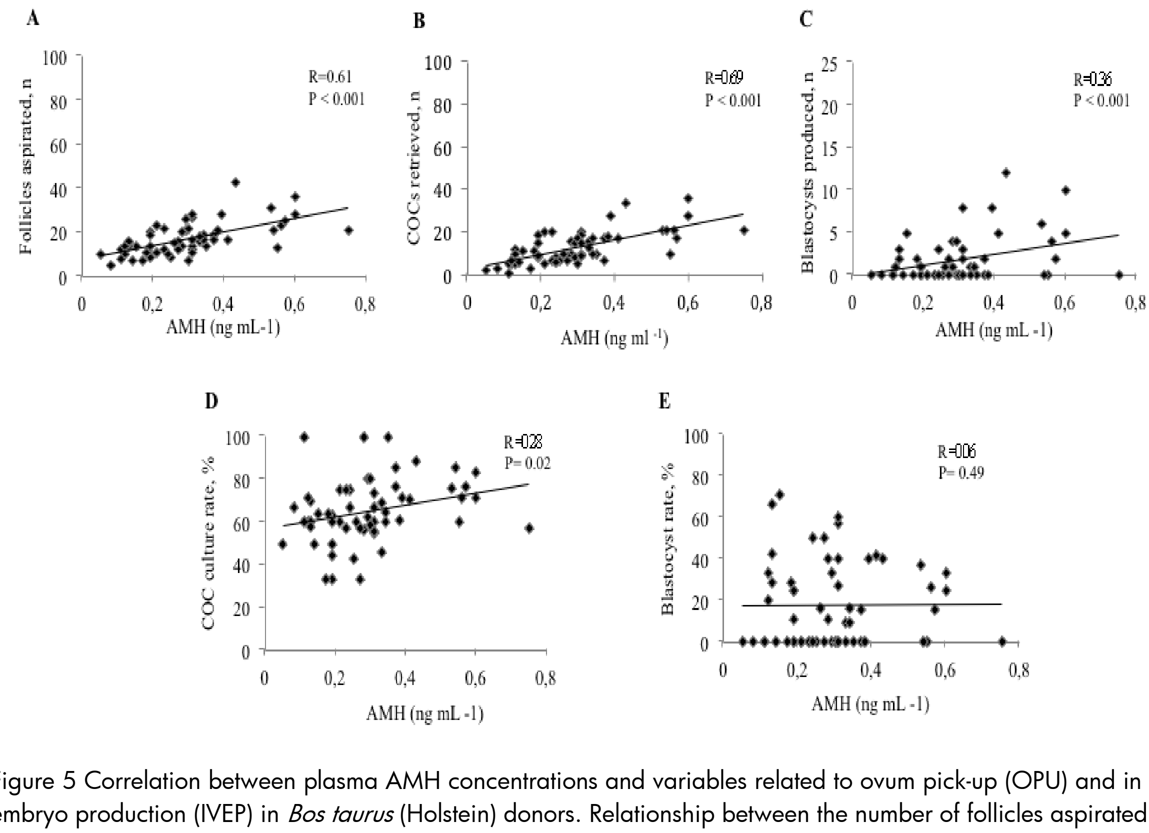

Figure 5 Correlation between plasma AMH concentrations and variables related to ovum pick-up (OPU) and in vitro embryo production (IVEP) in *Bos taurus* (Holstein) donors. Relationship between the number of follicles aspirated (A), the total COCs retrieved (B), the number of blastocysts produced (C), the COC culture rate (%, D) and the blastocyst rate (%, E) and plasma anti-Mullerian hormone (AMH) concentration. Guerreiro *et al.*, 2014.

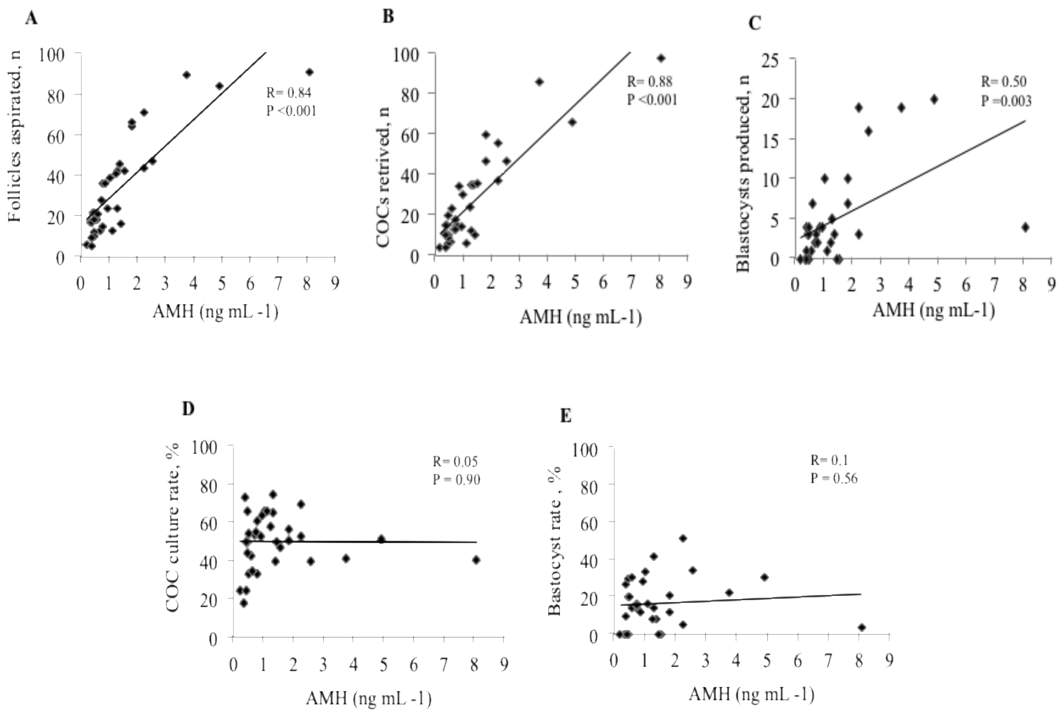

Figure 6. Correlation between plasma AMH concentrations and variables related to ovum pick-up (OPU) and in vitro embryo production (IVEP) in *Bos indicus* (Nelore) donors. Relationship between the number of follicles aspirated (A), the total COCs retrieved (B), the number of blastocysts produced (C), the COC culture rate (%, D) and the blastocyst rate (%, E) and plasma anti-Mullerian hormone (AMH) concentration. Guerreiro *et al.*, 2014

As observed, the relationship between AFP and AMH within genetic group concentration is similar regardless of time of blood collection. Indeed, it was reported previously that AMH levels variation is minimal during the estrous cycle (Ireland *et al.*, 2010; Rico *et al.*, 2009; Souza *et al.*, 2015), therefore blood samples can be taken at any time to evaluate circulating AMH. Thus, there is a practical advantage in utilizing AMH to predict AFP instead of direct counting AFP with ultrasound equipment. The exception would be the period just after superstimulatory treatments with FSH, in which the plasma AMH concentration appears to be greater than normal physiological levels. This increase in AMH concentration following FSH treatment may be due to growth of small follicles that were not detected by ultrasonography or the FSH treatment may have increased AMH secretion by granulosa cells. However, this hypothesis needs further investigation (Rico *et al.*, 2012, 2009).

A subsequent study was performed with the same genetic groups, but also considering different categories of age. A total of 59 Holstein (15 prepubertal heifers aged 8–10 months, 15 cyclic heifers aged 12–14 months, 14 lactating cows, and 15 nonlactating cows) and 34 Nelore (12 prepubertal heifers aged 10–11 months, 10 prepubertal heifers aged 21–23 months, and 12 cyclic heifers aged 24–26 months) females were enrolled in this study. Blood samples for plasma AMH determination were collected by coccygeal venipuncture immediately before the OPU sessions. In agreement to the previous findings, plasma AMH in *Bos indicus* and *Bos taurus* heifers showed a positive correlation with the total number of follicles aspirated, and also with the total number of complex cumulus oocytes (COCs) retrieved and the number of COCs cultured, and the number of embryos produced per OPU session (Figures 5 and 6). However, cleavage and blastocyst rates don't seem to have any correlation with circulating AMH (Figures 5 and 6; Guerreiro *et al.*, 2014).

Because genomic information allow producers to know genetic merit of their animals at early ages, we have recently explored the possibility of producing embryos retrieved from young female calves that were only 2-4 months old. In a previous study, it was shown that AMH concentrations decrease in parallel to the number of ovarian follicles as rodents (Kevenaar *et al.*, 2006) and women (Piltonen *et al.*, 2005) age. In agreement, in a recent study from our research group, we have found greater plasma AMH concentrations in calves compared to cycling heifers in both genetic groups, *Bos indicus* and *Bos taurus* (Fig. 7; Batista *et al.*, 2016).

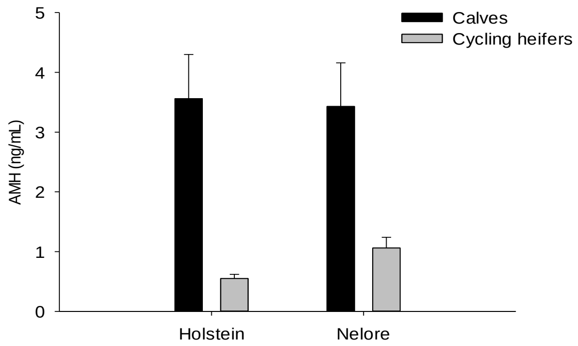

Figure 7. AMH plasma concentration (ng/mL) in calves (aging 2 to 4 monthes, Holstein: n = 24 and Nelore: n = 30) and cycling heifers (Holstein: n = 10 and Nelore: n = 12). Batista *et al.*, 2016.

Also, a positive correlation was observed between the plasma AMH concentration and the number of follicles (P < 0.0001), number of retrieved COCs (P < 0.0001), COCs cultured (P < 0.0001), cleaved COCs (P < 0.0001 and P = 0.001), and produced blastocysts (P = 0.0003 and P = 0.009) from *Bos indicus* (Nelore; Fig. 8) and *Bos taurus* (Holstein; Fig. 9) donor calves. However, there was no correlation between circulating AMH levels and cleavage rate (P = 0.24 and P = 0.36), COC culture rate (P = 0.28 and P = 0.07), or blastocyst rate (P = 0.52 and P = 0.08).

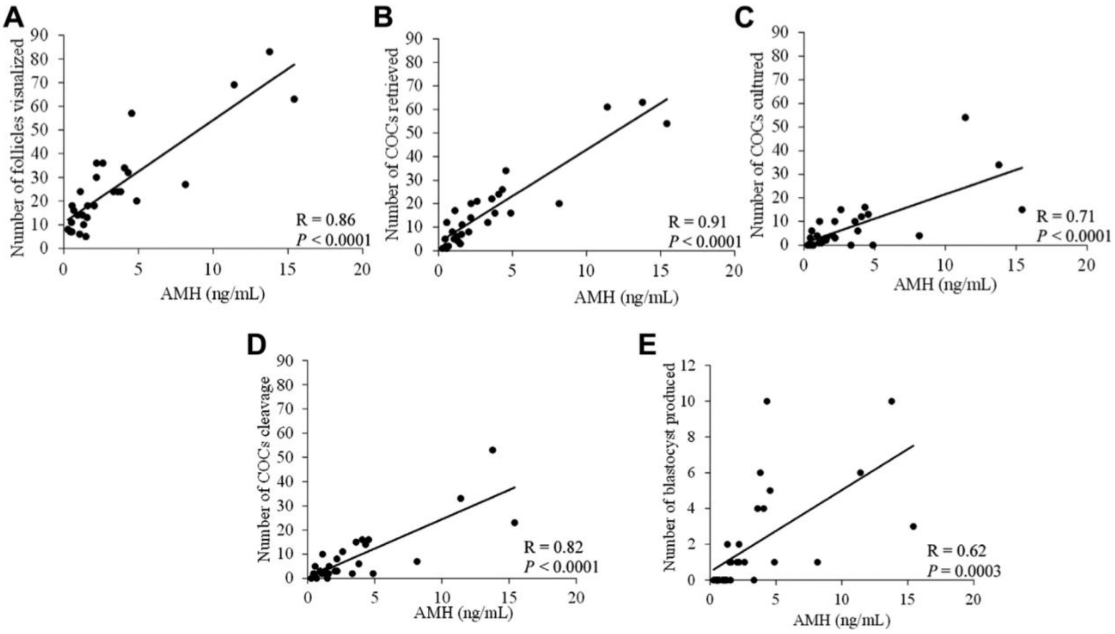

Figure 8. Correlations between plasma anti-Müllerian hormone (AMH) concentrations, the number of follicles and variables related to laparoscopic ovum pickup, and in vitro embryo production in B indicus donor calves (n=29). Relationships between the number of follicles (A), cumulus-oocyte complexes retrieved (B), cultured (C), and cleavage (D), blastocysts produced (E), and AMH concentration (ng/mL). Batista *et al.*, 2016.

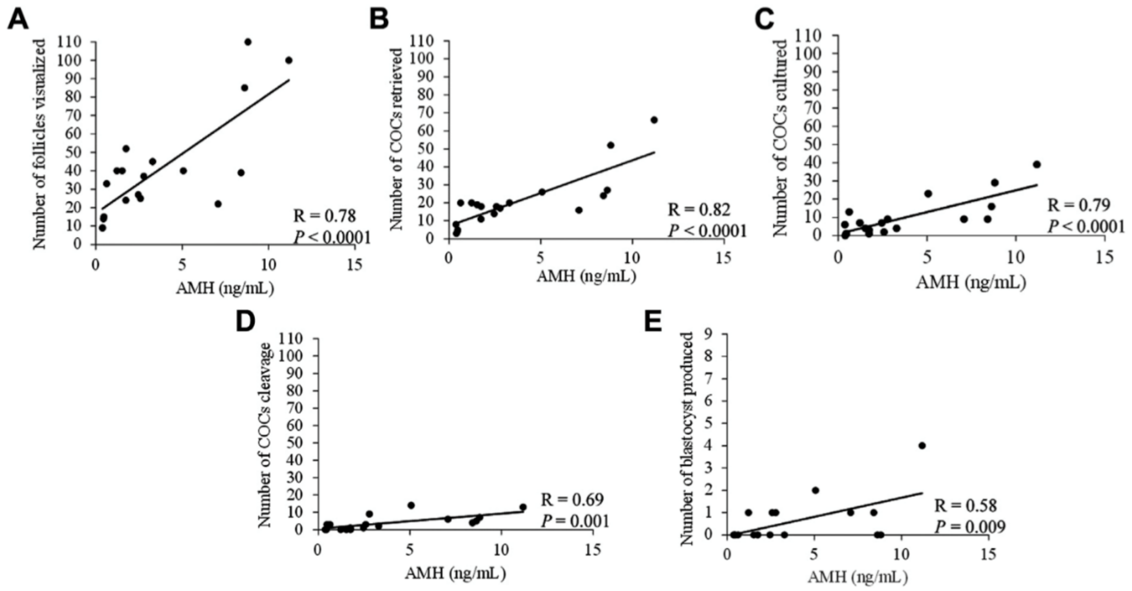

Figure 9. Correlations between plasma anti-Müllerian hormone (AMH) concentrations, the number of follicles and variables related to laparoscopic ovum pickup, and in vitro embryo production in B taurus donor calves (n=19). Relationships between the number of follicles (A), cumulus-oocyte complexes retrieved (B), cultured (C), and cleavage (D), blastocysts produced (E), and AMH concentration (ng/mL). Batista *et al.*, 2016.

In most situations, because examining ovaries from very young calves with an ultrasound can be difficult and unpractical, the determination of circulating AMH concentration in this animal category can be an important tool to select best oocyte-donors for in vitro embryo production, overcoming some of the technical limitations involved in utilizing an ultrasound in young calves. Therefore, we forecast that with the availability of genomic technology for the identification of animals with superior genetics at early ages and AMH measurement to facilitate identification of best oocyte-donors, that the use of calves as oocyte donors have its place in IVEP programs and will allow faster genetic gains by dramatically decreasing generation intervals (Armstrong *et al.*, 1992; Camargo *et al.*, 2005; Lohuis, 1995).

In conclusion, *Bos indicus* (Nelore) heifers and calves were found to have greater plasma AMH concentrations and larger AFP than *Bos taurus* (Holstein) heifers and calves. The AFP was positively correlated with plasma AMH concentrations in *Bos indicus* (Nelore), *Bos taurus* (Holstein) and *Bubalus bubalis* (Murrah) heifers, and *Bos indicus* (Nelore) and *Bos taurus* (Holstein) calves. We also found that plasma AMH concentrations could be a consistent endocrine marker of AFP and IVEP, even in calves, regardless of the genetic group. Therefore, the measurement of circulating AMH concentrations associated with genomic information can help select heifers and calves at very young ages as oocyte donors, accelerating the genetic gain of the herds.

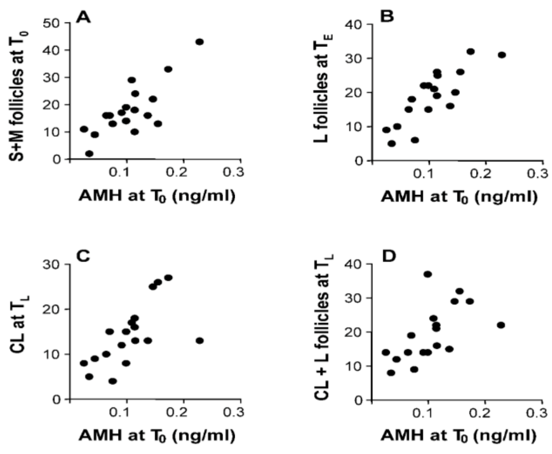

Figure 10. Relationships between AMH concentrations measured in plasma before superstimulatory treatment (T0) and (A) the numbers of small (S) and medium (M) follicles before superovulatory treatment or (B-D) the number of large follicles (L) and corpus luteum (CL) after the superovulatory treatment (n = 18 cows). Each circle represents data from one cow. Adapted from Rico *et al.*, 2009.

Anti-Müllerian hormone, superovulation and in vivo embryo production

A strong positive relationship between circulating AMH and the production of embryos in vivo following superovulation in dairy cattle was previously showed (Monniaux *et al.*, 2010a; Rico *et al.*, 2012; Souza *et al.*, 2015). AMH has been correlated with the number of small and medium follicles before the superstimulatory treatment (Fig. 10; Rico *et al.*, 2009), and with large follicles and CLs after superovulation (Fig. 10 and 11; Rico *et al.*, 2012, 2009; Souza *et al.*, 2015) and the number of embryos produced (Monniaux *et al.*, 2010a; Souza *et al.*, 2015) in primiparous and multiparous cows.

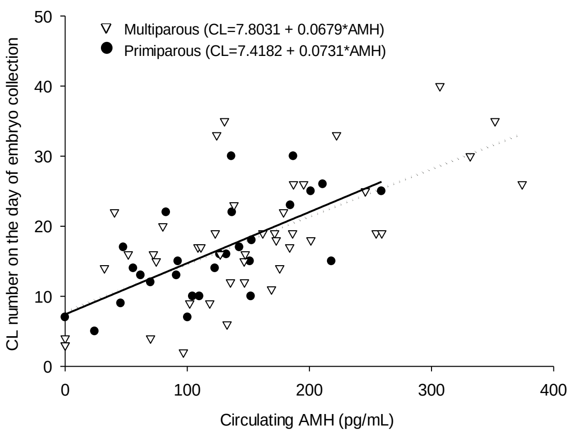

Figure 11. Average circulating AMH (pg/mL) and number of CL structures on the day of embryo collection for primiparous and multiparous dairy cows. Souza *et al.*, 2015.

The type of blood anti-coagulant factor may influence AMH measurements and it appears to be important to consider when interpreting AMH results. In this basis, different thresholds were reported for samples collected with heparin (87 pg/mL) and with EDTA (123 pg/mL) to identify dairy cows producing less than 15 ovulatory follicles after FSH treatment and near the time of estrous. Thus, AMH is reported to show a strong relationship with the number of small and medium follicles after superstimulation and also large follicles and CL after the treatment and may be used as a marker of potential candidates to integrate a reproductive program as embryo donors. Furthermore, measuring AMH before enrolling cows to FSH programs will likely allow practitioners to improve numbers of embryos produced and, thereby, reduces costs per embryo produced.

Anti-Müllerian hormone and Fertility

A positive association between AMH and fertility in dairy cows was recently reported in two studies (Jimenez-Krassel *et al.*, 2015; Ribeiro *et al.*, 2014). In one of them it was reported that cows with low AMH concentrations had lower pregnancy results following first service and greater incidence of pregnancy loss between day 30 and 65 of gestation (Ribeiro *et al.*, 2014). Moreover, dairy cows with relatively low circulating AMH concentrations as heifers also had the lowest survival rate after freshening for the first time compared with age-matched herdmates having greater AMH concentrations (Jimenez-Krassel *et al.*, 2015). However, it seems that AMH positive correlation with pregnancy is only valid when dairy cows are inseminated following estrus detection and lacks after TAI (Ribeiro *et al.*, 2014). Thus, it appears that the use of TAI protocols may override possible associations of AMH with field fertility and that may help explain some of contrasting results we have recently observed when working with Nelore cows.

Contrasting with data found for Holstein cows, when working with mature Nelore cows (n = 758) and heifers (n = 1,113) we found no correlation between AFP and (animals likely having greater AMH) and conception rates following TAI protocols (Table 1; Baruselli *et al.*, 2015). This finding is in accordance with previous (Santos *et al.*, 2014) reporting that high AFP positively influenced IVEP, but not conception rate of TAI Nelore cattle. More recently our group found similar results regarding Holstein cows subjected to TAI (Viziack *et al.*, 2016, unpublished data). In this study, regardless of having high or low AFP, conception rates following TAI were kept similar (Table 2).

Table 1. Number of animals enrolled in the trial, body condition score (BCS), age (months), ovulation and pregnancy rate after timed AI in Nelore heifers or cows according to the antral follicle population (AFP; measured at D4 of timed AI protocol; the expected moment of wave emergence) category in which animals were assigned. Data is shown as percentage or average ± standard error of the mean (SEM).

	Antral follicle population categories			Total	*P value*
	Low	Medium	High		
		COWS			
Number of animals	255	250	253	758	-
BCS (1 – 5)	3.00 ± 0.02	3.02 ± 0.02	3.02 ± 0.02	3.01 ± 0.01	0.29
Antral follicle population, n	24.5 ± 0.5[C]	39.2 ± 0.9[B]	56.3 ± 1.4[A]	40.0 ± 0.7	< 0.0001
Pregnancy rate, %	47.1	53.6	45.5	48.7	0.89
		HEIFERS			
Number of animals	371	371	371	1,113	-
Age, months	15.0 ± 0.1	14.9 ± 0.1	14.8 ± 0.1	14.9 ± 0.1	0.22
BCS (1 – 5)	3.27 ± 0.02	3.27 ± 0.02	3.31 ± 0.02	3.28 ± 0.01	0.74
Antral follicle population, n	7.1 ± 0.1[C]	11.3 ± 0.1[B]	17.2 ± 0.2[A]	11.8 ± 0.2	< 0.0001
Ovulation rate, %	82.5	78.3	79.8	80.2	0.56
Pregnancy rate, %	39.6	36.4	36.7	37.6	0.57

Table 2. Average and mean standard deviation of production and reproduction-linked characteristics of lactating Holstein cows with high or low antral follicle population (AFP).

	High AFP	Low AFP	P value
Number of lactation	1.0 ± 0.2	1.0 ± 1.0	> 0.05
Number of insemination	3.1 ± 2.9	2.5 ± 2.6	> 0.05
Milk production (Kg/day))	30.6 ± 9.8	31.4 ± 9.4	> 0.05
Number of follicles	30.6 ± 9.7	12.6 ± 4.2	< 0.05
Pregnancy rate 30d after TAI	24.6%	24.5%	> 0.05

Additionally, although AFP has been respectably associated with embryo production, no effect of donor (non lactating Holstein cows) AFP was observed on pregnancy establishment of recipients after transferring the embryos produced in vitro (Fig. 12; Bragança *et al.*, 2014). Similarly, an absence of relationship between circulating levels of AMH and pregnancy probability was reported for crossbred recipients that received an in vitro produced embryo from Holstein donors (Fig. 13; Guerreiro *et al.*, unpublished data). We also failed to observe any association of circulating AMH neither with calving to conception intervals in Holstein and Jersey lactating cows (r = -0.05, P > 0.10) nor with age at conception in Holstein and Jersey nulliparous heifers (r = -0.06, P>0.10), inseminated after estrous detection or TAI (Carvalho *et al.*, 2015; Fig. 14). Thus, circulating AMH doesn't correlate with reproductive performance of lactating cows or heifers from Holstein and Jersey breeds.

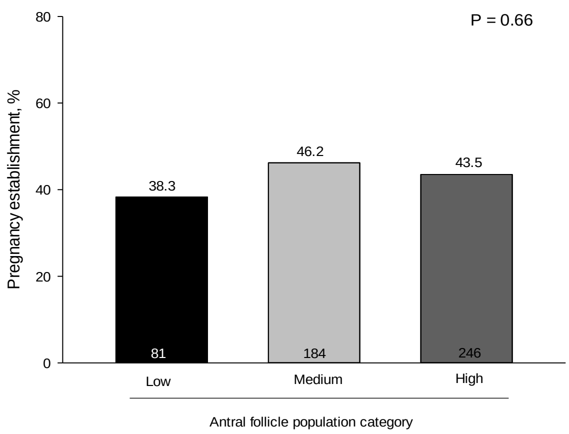

Figure 12. Pregnancy in crossbred (*Bos indicus* x *Bos taurus*) recipients after embryo transfer of in vitro produced embryos according to the non-lactating Holstein donor antral follicle population category (low, medium or high). Adapted from Bragança *et al.*, 2014.

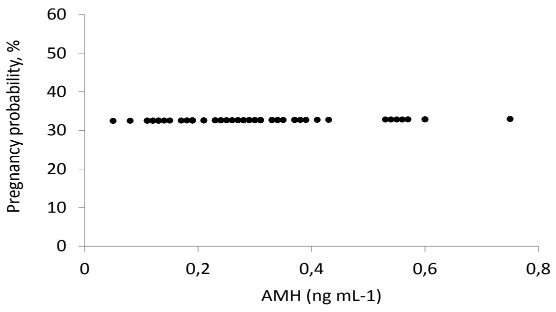

Figure 13. Relationship between circulating levels of AMH (ng/mL) and pregnancy probability in crossbred receptors after transferring in vitro produced embryos from Holstein donors (n=107). Adapted from Guerreiro *et al.*, unpublished data.

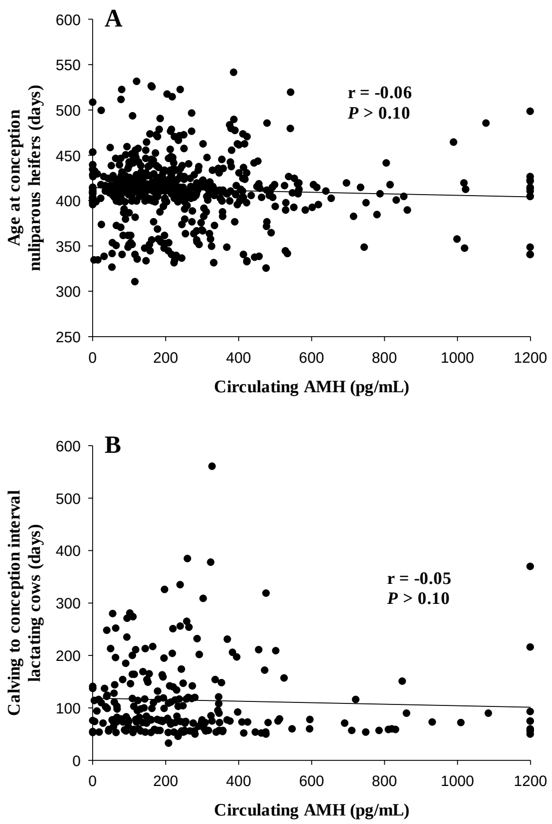

Figure 14. Relationship between circulating levels of AMH (pg/mL) and age (days) at conception in heifers (n = 528; panel A); and interval from calving-to-conception (days open) in lactating cows (n = 223; panel B). Adapted from Carvalho *et al.*, 2015.

Furthermore, because number of ovarian antral follicles appears to be correlated in cattle dam-daughter pairs (Walsh *et al.*, 2014), and that may allow for selection of animals with greater AFP, we have recently looked into possible associations between circulating AMH in dam-daughter pairs in Holstein and Jersey breeds (Figure 15; Batista *et al.*, 2015 – non-publish data). Although significant, the correlation in circulating AMH in dam-daughter pairs, was somewhat low (r = 0.18). Although, AFP in cattle is moderately heritable (0.31; Walsh *et al.*, 2014), epigenetic factors such as levels of negative energy balance during early fetal life (Evans *et al.*, 2012) as well as dam-age and lactation status (Walsh *et al.*, 2014) might likely influence antral follicle count in offspring. These epigenetic factors might then explain the poor correlation found in circulating AMH between dam-daughter pairs. Overall, the value of using AMH measurement to predict field fertility is still controversial and further studies using large numbers of animals are needed to draw final conclusions.

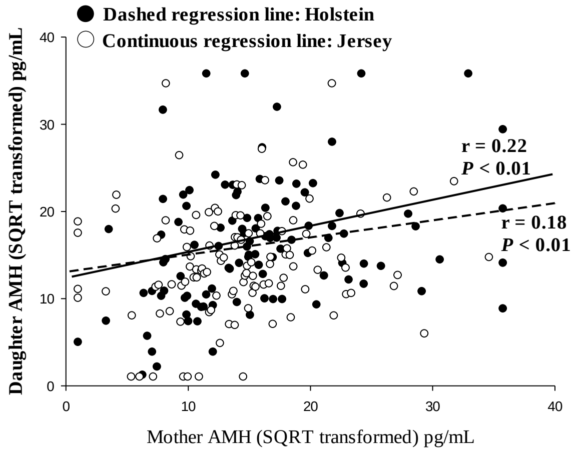

Figure 15. Correlation between circulating levels of AMH (pg/mL) in Holstein (n=116) and Jersey (n =106) dam-daughter pairs. AMH values are squared-root transformed. Batista *et al.*, 2015 - non-publish data.

CONCLUSIONS AND IMPLICATIONS

The plasmatic concentration of AMH has been reported to positively correlate with the size of AFP, superovulatory response (number of large follicles and CL after treatment) and in vitro and in vivo embryo production. However, the association of AMH and fertility (pregnancy outcomes) is still controversial, and seems to be affected by other factors. Thus, AMH has the potential of being a reliable endocrine marker of ovarian reserve (AFP), and it may be used to predict the results of in vivo and in vitro embryo production. The levels of AMH is emerging as a new tool to help the identification of donor cows with greater potential from embryo production (more follicles available for OPU, and greater superovulation), accelerating the dissemination of selected genetics and improving herds genetic gain.

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