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High Anti-Mullerian Hormone (AMH) Levels Do Not Impact Live Delivered Pregnancy Rates (LDPR) Per Transfer or Retrieval

Brooke Neumann DO[1], Jerome H. Check MD PhD[2,3] and Carrie Wilson MS[3]

[1]*Inspira Health Network Vineland, New Jersey, USA.*

[2]*Cooper Medical School of Rowan University Camden, New Jersey, USA.*

[3]*Cooper Institute for Reproductive and Hormonal Disorders, Mt Laurel, New Jersey, USA.*

***Correspondence:**
Brooke Neumann, DO, 7447 Old York Road, Melrose Park, PA 19027, TEL: 215-635-4400, FAX: 215-635-2304.

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ABSTRACT

Background: *Previous literature suggests high anti-Mullerian hormone (AMH), specifically ≥ 5 ng/mL, has a negative effect on pregnancy rates following in vitro fertilization transfer (IVF-ET) [1].*

Objective: *The aim of this study was to determine if women with increased serum AMH levels (≥ 5ng/mL) have a lower live delivered pregnancy rate per transfer than women with serum AMH between 1 and 4.99, and to evaluate the confounding effect of advancing age. Furthermore, if this study does corroborate other studies suggesting lower pregnancy rates in an in vitro fertilization embryo transfer (IVF-ET) cycle, the study would uniquely evaluate whether a lower live delivered pregnancy rate (LDPR) per transfer may be compensated related to more embryos i.e, frozen, thawed being transferred in subsequent cycles without undergoing another oocyte retrieval.*

Materials and Methods: *Women undergoing IVF-ET between the years of 2015-2023 were stratified by serum AMH ranges. (> 1-2.99, ≥ 3-<4.99, ≥ 5-<7.99 and a subset ≥ 8ng/mL). They were also stratified by age (≥ 35, 36-39, 40-44). LDPRs were recorded as seen in Table 1. LDPR was also calculated. The first frozen transfer would count as a woman's first transfer cycle if a fresh embryo transfer could not be completed if there was concern about the risk of the ovarian hyperstimulation syndrome.*

Results: *There were no significant differences in the LDPR per transfer or retrieval with higher serum AMH levels between any of the different age ranges or the subset of the age range of serum AMH ≥ 8 ng/mL.*

Conclusion: *High serum AMH levels do not have a negative impact on pregnancy rates per retrieval nor transfer by the method used by our IVF center for controlled ovarian hyper stimulation.*

Keywords

High anti-Mullerian hormone, Live delivered pregnancy rates, Polycystic ovarian syndrome.

Introduction

Anti-mullerian hormone (AMH), also known as Mullerian inhibiting substance (MIS), is part of the transforming growth factor-B superfamily [1]. It acts to suppress recruitment of primordial follicles and is produced by granulosa cells of primary, preantral and small antral follicles [2]. AMH is a sensitive marker for ovarian reserve [3]. AMH is a promising marker to predict ovarian response to gonadotrophins and the chance of a successful pregnancy as a result of *in vitro* fertilization-embryo transfer (IVF-ET) [4-7].

Polycystic ovarian syndrome (PCOS) is the most common endocrine disorder affecting women of reproductive age, with an estimated prevalence of 5–8% [8]. Diagnostic criteria include hyperandrogenism, anovulation and polycystic appearing ovaries after the exclusion of other conditions [9]. Serum AMH level

appears to be related to the severity of PCOS, and may not reflect their ovarian reserve, thus confounding the association between AMH and IVF outcomes [8].

The clinical utility of AMH in the PCOS population is difficult to define. AMH levels are higher in PCOS patients compared to patients without this diagnosis [10]. Patients with PCOS have a higher AMH likely due to both the number of preantral follicles and the increased production of AMH from granulosa cells [9]. Generally, a higher serum AMH greater than a level predictive of diminished oocyte reserve (DOR) would favor a greater chance of a live baby delivered, not only because women with DOR may produce somewhat fewer quality oocytes leading to less quality embryos, but also failing to provide a chance of conception by a subsequent frozen embryo transfer [11]. There are some studies however that find a lower LDPR per transfer if the serum AMH is too high, e.g., ≥5 ng/ml [12,13]. The difference is magnified by advancing age, but the greater number of embryos formed may provide a better LDPR from a given oocyte retrieved because of more embryos produced.

The goal of this study was to determine if women with increased serum AMH levels (≥ 5ng/mL) have a lower live delivered pregnancy rate per transfer than women with serum AMH between 1 and 4.99, and to evaluate the confounding effect of advancing age. Furthermore, if this study does corroborate other studies suggesting lower pregnancy rates in an *in vitro* fertilization embryo transfer (IVF-ET) cycle in women with high serum AMH levels, the study would uniquely evaluate whether a lower live delivered pregnancy rate (LDPR) per transfer may be compensated related to more embryos created, i.e., frozen-thawed embryos being transferred in subsequent cycles without undergoing another oocyte retrieval.

Materials and Methods

This was a retrospective study at a single private practice in women undergoing IVF-ET between the years of 2015-2023. All patients were included regardless of the cause of infertility (male factor, anovulation, tubal factor, etc.). With each patient, the pros and cons of transferring a fresh embryo or frozen embryo with delayed transfer was discussed. The decision was based on consultation between physical exam and patients' characteristics. Patients were all treated with a down-regulated gonadotropin releasing hormone (GNRH) antagonist protocol using either ganirelix or cetrorelix. The trigger injection was based on risk of OHSS and was generally given when there were 2 lead follicles with an average of 20mm diameter. The trigger injection was either 10,000 IU human chorionic gonadotropin or 1mg leuprolide acetate x3 every 12 hours. The latter was chosen if there was a risk of ovarian hyperstimulation (over 20 antral follicles or serum estradiol (E2) ≥ 5000 pg/nl on day of trigger). All patients were placed on vaginal progesterone for luteal phase support. Oocyte retrieval was completed in standard fashion and fertilization was performed using conventional insemination or intracytoplasmic sperm injection (ICSI) based on seven characteristics.

Women were stratified by serum AMH ranges: >1-2.99, ≥ 3-<4.99, ≥5-<7.99 and a subset ≥ 8ng/mL. Women were also stratified by age ≥35, 36-39, and 40-44. The first frozen transfer would count as a woman's first transfer cycle if a fresh embryo transfer could not be completed, e.g., if there was concern about the risk of the ovarian hyperstimulation syndrome. Cycles were cancelled if there was premature luteinization. The LDPR per transfer was defined as the achievement of a live delivery from the transfer of fresh embryos (fresh or frozen) obtained from the oocyte retrieval or the first transfer of frozen-thawed embryos if the transfer or fresh embryos was deferred. The LDPR rate per retrieval was defined as the birth of a live baby from any of the embryos created from the oocyte retrieval without the need to try a second IVF-ET cycle. All embryo transfers were performed with 3-day old embryos or blastocysts.

Results

Live delivered pregnancy rates (LDPRs) per retrieval and per transfer were recorded as seen in Table 1. As seen in Table 1, the serum level of AMH did not affect LDPR per retrieval or transfer at any age. As expected, the LDPR per transfer was less than that of LDPR per retrieval. It is difficult to determine whether the non-significant trend for higher LDPRs per retrieval of 33% and per transfer of 22% may be indicative of better quality of eggs in women with higher serum AMH despite advanced reproductive age (40-44) compared to 13.2% (23/174) and 11.2% (23/195) for women with normal egg reserve and normal (not high) serum AMH because of discrepancy in size of the two cohorts.

Discussion

The current study shows that higher levels of serum AMH did not adversely affect LDPR per retrieval or transfer at any age range. Higher AMH levels may correlate with a greater number of mature

Table 1: Live delivered pregnancy rate (LDPR) per retrieval and transfer for women according to 4 serum AMH ranges and by three different age ranges.

		AMH ≥1- ≤2.99 (ng/ml)	AMH ≥3- ≤4.99 (ng/ml)	AMH ≥5 (ng/ml)	AMH ≥8 (ng/ml)
≤35 years old	LDPR per retrieval	54.6% (100/183)	66.3% (69/104)	66.7% (84/126)	73.7% (42/57)
	LDPR per transfer	36.8% (100/272)	39.9% (69/173)	39% (84/215)	43% (42/97)
36-39 years old	LDPR per retrieval	41.2% (56/136)	48.9% (22/45)	48.6% (17/35)	66.7% (8/12)
	LDPR per transfer	32.4% (56/173)	40% (22/55)	32% (17/53)	38% (8/21)
40-44 years old	LDPR per retrieval	13.7% (20/145)	10.3% (3/29)	33.3% (6/18)	50% (1/2)
	LDPR per transfer	12.9% (20/155)	7.5% (3/40)	22% (6/27)	20% (1/5)

oocytes, thus more embryos, resulting in higher pregnancy rates [11-16].

Some previous studies showed that cumulative pregnancy rate or live birth rate was lower in women with high serum AMH group or similar to controls [17-19]. In addition, some studies suggest that women with PCOS and high serum AMH (mean 11.86) produce inferior quality embryos compared to women with normal serum AMH (3.3 ng/ml) i.e., 53.6% with PCOS and high AMH vs 66.9% with normal AMH [19]. Lower good-quality embryo rates in the PCOS group (11.86 ng/mL AMH) compared to the low AMH group (3.33 ng/mL) (66.9% vs. 53.6%, $p<0.001$) (19). Similarly, Tal et al found that high serum AMH predicted inferior LDPRs [20]. However, not all studies agree that high AMH levels may predict a lower LDPR per transfer. Thus, these reached similar conclusions to the study presented here. In addition, these authors found the high AMH group had increased number of oocytes and more good quality embryos compared to women with a slightly lower AMH level [20]. Liu et al. performed a retrospective review in PCOS patients undergoing IVF by AMH levels: ≤2.25, 2.25-5.71, and >5.71 and found that LDPR was 47.6%, 55.2% and 59.5% respectively ($p<0.001$) [21].

However, although they found a higher live birth rate with higher AMH compared to low and average, after controlling for age and egg yield, there was no difference [21]. Reichman et al. found that pregnancy rate per retrieval stratified by age, there was a trend toward higher pregnancy rates with higher AMH until age >40 years old and pregnancy rate per transfer had a positive trend but only in patients <35 and 38-40 years old [22]. Kaya et al. grouped patients by AMH level, <2.54- low, 2.54-3.85 moderate and high >3.85 and found that clinical pregnancy rate (CPR) were 33.1 (7/21), 46.1 (19/39) and 60% (12/20) respectively ($p<0.001$) [23]. In a meta-analysis in 4,324 women performed by Tal et al., the pooled diagnostic odds ratio for AMH as a predictor of higher clinical pregnancy rates was 2.10 [24]. In addition, in the ultrahigh AMH group >10 ng/mL, patients had better quality embryos. However, although these studies concluded similar findings, none of these studies reported live delivered pregnancy rate per retrieval and per transfer as we did in the current study. In addition, most of these studies reported live delivered pregnancy rates by fresh embryo transfers, whereas we reported the fresh embryo transfer unless it could not be completed due to risk of OHSS, then the first frozen transfer was included. As women age, it is unclear whether or not higher AMH levels may be associated with better LDPRs related to a greater ovarian reserve. A higher reserve max compensates for a decline in both oocyte quality and quantity and thus more oocytes retrieved and better pregnancy rates [25]. However, poor responders with low serum AMH levels at a young age typically have a better chance of achieving a pregnancy than those of advanced reproductive age [11,26-28]. It is hypothesized that at an older age, there is impairment of mitochondrial function and increased production of reactive oxygen species with older granulosa cells [29-30]. This could be because there are less high-quality embryos [11].

The variation in studies could be due to multiple factors including heterogeneity in individual patient populations, IVF protocols, and specific serum AMH assay used. Although all included patients regardless of the cause of infertility (male factor, anovulation, tubal factor, etc.), higher AMH levels can be seen in patients with PCOS [20]. Although it is still largely unknown why patients with PCOS have a higher AMH, it is likely due from both the number of preantral follicles and the increased production from each granulosa cell [9]. Thus, in patients with PCOS who now have DOR, a physician may predict a better yield of oocytes based on serum AMH levels so that for PCOS a day 3 serum FSH may be a better predictor of a poor responder [26]. The androgen production in PCOS could possibly play a role in reducing LDPRs [31-32]. Studies have suggested that a higher AMH could suggest a more severe phenotype of PCOS [33-35]. Unfortunately, the sample size of this study was limited. In addition, the authors recognize that lifestyle factors such as smoking and medications such as oral contraceptives may play a role in suppressing AMH levels [36]. Lastly, the number of cycle cancellations was not included in this study and could be added for future studies. Acharya et al found that each unit increase in serum AMH >5 ng/mL was associated with a decrease in live birth per stimulated cycle because of increased cancellation rate, though the live birth rate per transfer was preserved [37]. This is in contrast to Reichman et al. who found that there was an inverse correlation between AMH level and cycle cancellation ($p<0.0001$) [22]. However, our findings suggest that AMH does not have a negative effect on LDPR, both by transfer and by retrieval. We did not measure serum testosterone (T) on all patients, which is why T levels are not included. Since higher serum AMH did not seem to impair LDPRs per transfer, and it is likely that women with higher serum AMH generally have higher serum testosterone levels (at least in part from the effect of AMH on inhibiting the FSH induced aromatase enzyme), it would not seem that higher levels of T impair pregnancy outcome following IVF-ET or frozen ET [38].

The intent of this study was not to perform a meta-analysis to answer the question whether higher serum AMH levels lead to lower, higher, or no difference in LDPRs in women with normal serum levels of AMH. Several authors agree with these findings, favoring our conclusion that a higher serum AMH does not negatively affect LDPRs following embryo transfer but, possibly, may even improve LDPRs per retrieval, i.e., achieving a live delivered pregnancy from a given cohort of embryos from an IVF cycle without having to go through a second round of controlled ovarian stimulation and oocyte retrieval, fertilization, and embryo formation [20-26]. This conclusion is in contrast to the other research whose data favored a lower chance of achieving a live delivery with higher serum AMH [17-19].

IRB statement

Neither Cooper Medical School of Rowan University or Cooper Hospital require an IRB for a study. The patients whose data we used to perform this retrospective study all gave written permission in their initial visit that their data could be used for future studies.

References

1. La Marca A, Sighinolfi G, Radi D, et al. Anti-Mullerian hormone (AMH) as a predictive marker in assisted reproductive technology (ART). Hum Reprod Update. 2010; 16: 113-30.
2. Pellatt L, Rice S, Mason HD. Anti-Mullerian hormone and polycystic ovary syndrome: a mountain too high? Reproduction. 2010; 139: 825-833.
3. Van Rooij IA, Broekmans FJ, Te Velde ER, et al. Serum anti-Mullerian hormone levels: a novel measure of ovarian reserve. Hum Reprod. 2002; 17: 3065-3071.
4. Broer SL, Mol BW, Hendriks D, et al. The role of antiMullerian hormone in prediction of outcome after IVF: comparison with the antral follicle count. Fertil Steril. 2009; 91: 705-714.
5. Lekamge DN, Barry M, Kolo M, et al. Anti-Mullerian hormone as a predictor of IVF outcome. Reprod Biomed Online. 2007; 14: 602-610.
6. Hazout A, Bouchard P, Seifer DB, et al. Serum antimullerian hormone/mullerian-inhibiting substance appears to be a more discriminatory marker of assisted reproductive technology outcome than follicle-stimulating hormone, inhibin B, or estradiol. Fertil Steril. 2004; 82: 1323-1329.
7. Eldar-Geva T, Ben-Chetrit A, Spitz IM, et al. Dynamic assays of inhibin B, anti-mullerian hormone and estradiol following FSH stimulation and ovarian ultrasonography as predictors of IVF outcome. Hum Reprod. 2005; 20: 3178-3183.
8. Nardo LG, Yates AP, Roberts SA, et al. The relationships between AMH, androgens, insulin resistance and basal ovarian follicular status in non-obese subfertile women with and without polycystic ovary syndrome. Hum Reprod. 2009; 24: 2917-2923.
9. Norman RJ, Dewailly D, Legro RS, et al. Polycystic Ovary Syndrome. Lancet. 2007; 370: 685-697.
10. Pellatt L, Hanna L, Brincat M, et al. Granulosa cell production of anti-Mullerian hormone is increased in polycystic ovaries. J Clin Endocrinol Metab. 2007; 92: 240-245.
11. Check JH, Wilson C. The younger the patients the less adverse effect of diminished oocyte reserve on outcome following *in vitro* fertilization-embryo transfer as long as the proper ovarian stimulation protocol is used. J Reprod Contracep. 2013; 24: 221-227.
12. Fallat ME, Siow Y, Marra M, et al. M€ullerian-inhibiting substance in follicular fluid and serum: a comparison of patients with tubal factor infertility, polycystic ovary syndrome, and endometriosis. Fertil Steril. 1997; 67: 962-965.
13. La Marca A, Malmusi S, Giulini S, et al. Anti-mullerian hormone plasma levels in spontaneous menstrual cycle and during treatment with FSH to induce ovulation. Hum Reprod. 2004; 19: 2738-2741.
14. Hazout A, Bouchard P, Seifer DB, et al. Serum anti-mullerian hormone/mullerian-inhibiting substance appears to be a more discriminatory marker of assisted reproductive technology outcome than follicle-stimulating hormone, inhibin B, or estradiol. Fertil Steril. 2004; 82: 1323-1329.
15. Lekamge DN, Barry M, Kolo M, et al. Anti-m€ullerian hormone as a predictor of IVF outcome. Reprod Biomed Online. 2007; 14: 602-610.
16. Van Rooij IA, Broekmans FJ, Hunault CC, et al. Use of ovarian reserve tests for the prediction of ongoing pregnancy in couples with unexplained or mild male infertility. Reprod Biomed Online. 2006; 12: 182-190.
17. Xi W, Gong F, Lu G. Correlation of serum Anti-Müllerian hormone concentrations on day 3 of the *in vitro* fertilization stimulation cycle with assisted reproduction outcome in polycystic ovary syndrome patients. J Assist Reprod Genet. 2012; 29: 397-402.
18. Guo Y, Liu S, Hu S, et al. High Serum Anti-Müllerian Hormone Concentrations Are Associated With Poor Pregnancy Outcome in Fresh IVF/ICSI Cycle but Not Cumulative Live Birth Rate in PCOS Patients. Front Endocrinol. 2021; 12: 673284.
19. Chen Y, Ye B, Yang X, et al. Predicting the outcome of different protocols of *in vitro* fertilization with anti Mullerian hormone levels in patients with polycystic ovary syndrome. J Int Med Research. 2017; 45: 1138-1147.
20. Tal R, Seifer CM, Khanimov M, et al. High serum Antimullerian hormone levels are associated with lower live birth rates in women with polycystic ovarian syndrome undergoing assisted reproductive technology. Reprod Biol Endocrinol. 2020; 18: 20.
21. Liu S, Hong L, Mo M, et al. Association of antimüllerian hormone with polycystic ovarian syndrome phenotypes and pregnancy outcomes of *in v*itro fertilization cycles with fresh embryo transfer. BMC Pregnancy Childbirth. 2022; 22: 171.
22. Reichman DE, Goldschlag D, Rosenwaks Z. Value of antimüllerian hormone as a prognostic indicator of *in vitro* fertilization outcome. Fertil Steril. 2014; 101: 1012-1018.e1.
23. Kaya C, Pabuccu R, Satıroglu H. Serum antimüllerian hormone concentrations on day 3 of the *in vitro* fertilization stimulation cycle are predictive of the fertilization, implantation, and pregnancy in polycystic ovary syndrome patients undergoing assisted reproduction. Fertil Steril. 2010; 94: 2202-2207.
24. Tal R, Tal O, Seifer BJ, et al. Antimüllerian hormone as predictor of implantation and clinical pregnancy after assisted conception: a systematic review and meta-analysis. Fertil Steril. 2015; 103: 119-130.e3.
25. Tal R, Seifer DB, Khanimov M, et al. Characterization of women with elevated antimüllerian hormone levels (AMH): correlation of AMH with polycystic ovarian syndrome phenotypes and assisted reproductive technology outcomes. Am J Obstet Gynecol. 2014; 211: 59.e1-e8.
26. Silberstein T, Maclaughin DT, Shai I, et al. M€ullerian inhibiting substance levels at the time of HCG administration in IVF cycles predict both ovarian reserve and embryo morphology. Human Reprod. 2006; 21: 159-163.

27. Check JH. A follicle stimulating hormone (FSH) receptor up-regulation technique as a method for follicular reserve for *in vitro* fertilization-embryo transfer in women with diminished oocyte reserve. Ed. Leon V. Berhardt; In: Advances in Medicine and Biology, Nova Science Publishers. 2022; 195: 119-137.
28. Ulug U, Ben-Shlomo I, Turan E, et al. Conception rates following assisted reproduction in poor responder patients: a retrospective study in 300 consecutive cycles. Reprod Biomed Online. 2003; 6: 439-443
29. May-Panloup P, Boucret L, Chao de la Barca JM, et al. Ovarian ageing: the role of mitochondria in oocytes and follicles. Hum Reprod Update. 2016; 22: 725-743.
30. Xu L, Haixiang Sun, Mei Zhang, et al. MicroRNA-145 protects follicular granulosa cells against oxidative stress-induced apoptosis by targeting Kruppel-like factor 4. Mol Cell Endocrinol. 2017; 452: 138-147.
31. Pigny P, Merlen E, Robert Y, et al. Elevated serum level of anti-Mullerian hormone in patients with polycystic ovary syndrome: relationship to the ovarian follicle excess and to the follicular arrest. J Clin Endocrinol Metab. 2003; 88: 5957-5962.
32. Eldar-Geva T, Margalioth EJ, Gal M, et al. Serum anti-Mullerian hormone levels during controlled ovarian hyperstimulation in women with polycystic ovaries with and without hyperandrogenism. Hum Reprod. 2005; 20: 1814-1819.
33. Teede H, Misso M, Tassone EC, et al. AntiMüllerian Hormone in PCOS: A Review Informing International Guidelines. TEM. 2019; 30: 467-478.
34. Dewailly D, Gronier H, Poncelet E, et al. Diagnosis of Polycystic Ovary Syndrome (PCOS): Revisiting the Threshold Values of Follicle Count on Ultrasound and of the Serum AMH Level for the Definition of Polycystic Ovaries. Hum Reprod. 2011; 26: 3123-3129.
35. Nardo LG, Yates AP, Roberts SA, et al. The Relationships Between AMH, Androgens, Insulin Resistance and Basal Ovarian Follicular Status in non-Obese Subfertile Women With and Without Polycystic Ovary Syndrome. Hum Reprod. 2009; 24: 2917-2923.
36. Dolleman M, Verschuren WM, Eijkemans MJ, et al. Reproductive and lifestyle & determinants of anti-Mullerian hormone in a large population-based study. J Clin Endocrinol Metab. 2013; 98: 2106-2115.
37. Acharya KS, Harris BS, Weber JM, et al. Impact of increasing antimüllerian hormone level on *in vitro* fertilization fresh transfer and live birth rate. F S Rep. 2022; 3: 223-230.
38. Chang HM, Klausen C, Leung PC. Antimüllerian hormone inhibits follicle-stimulating hormone-induced adenylyl cyclase activation, aromatase expression, and estradiol production in human granulosa-lutein cells. Fertil Steril. 2013; 100: 585-592.e1.