



Temporal effect of maternal heat stress during gestation on the fertility and anti-Müllerian hormone concentration of offspring in bovine

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ABSTRACT

Ovarian reserve has been suggested as an important contributing factor of reproductive success in bovine. Size of ovarian reserve is determined during fetal period and it could be altered by environmental factors, with which the dam is exposed. Maternal heat stress could impair placental function and fetal development; however, there is limited information on the impact of prenatal heat stress on fertility and ovarian reserve in the offspring. Therefore, a retrospective study was conducted, in which fertility parameters and AMH concentration, as a reliable marker of ovarian reserve in bovine, were studied in the offspring of dams that had been exposed to heat stress during the first (FTE), second (STE) or third (TTE) trimester of gestation and the offspring of dams unexposed to heat stress (US). Additionally, postpartum exposure of offspring with heat stress was considered in the model to adjust the statistical analysis in this regard. Days to first service (DFS) and calving to conception interval (CCI) were prolonged in exposed than unexposed cows ($P < 0.05$). Days to first service and CCI were also longer in STE compared with FTE cows ($P < 0.05$). First service conception rate was lower in TTE than UN cows ($P < 0.05$). The proportion of repeat breeders was higher in exposed compared with unexposed cows ($P < 0.05$). Service per conception was higher in STE and TTE than UN cows ($P < 0.05$). Culling rate between different periods of lactation was also higher in exposed than unexposed cows ($P < 0.05$). Finally, AMH concentration was lower in STE and TTE than UN cows ($P < 0.05$); moreover, it was lower in STE compared with FTE cows ($P < 0.05$). In conclusion, the present study revealed detrimental effects of maternal heat stress on fertility, productive longevity and ovarian reserve in the offspring. In this context, the second and third trimesters appeared to be more critical periods.

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1. Introduction

The size of ovarian reserve, the number of primordial follicles, has been observed to be associated with fertility in bovine [1–3]. Cows with low ovarian reserve have low oocyte quality [4], reduced embryonic developmental competence [5], low progesterone concentration [6], impaired endometrial growth [6] and earlier ovarian senescence [7]. As a result, lower pregnancy rates [1,2], higher service per conception [1], longer calving to conception interval [1] and shorter productive life [3] have been reported in cows with low ovarian reserve.

The formation of ovarian reserve occurs during fetal life in bovine [8,9]. Although the size of ovarian reserve is influenced by genetic factors [10], compelling evidence indicates that maternal environmental factors during pregnancy could substantially impact the size of ovarian reserve in the female offspring in bovine [11] as well as other mammals [12–16].

It is well-established that heat stress adversely affects reproduction in mammals including cattle [17,18]. Exposure to heat stress would impair oocyte competence [19] and disrupt early embryonic development [20]. Moreover, heat stress causes reduced fetal and placental weight [21], and concentrations of placental hormones [22]. Maternal exposure to heat stress during gestation has also been observed to compromise the growth and immune function of calves [23,24]. Furthermore, heifers born to cows exposed to heat stress during late gestation have been observed

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with more service per conception [24]. Nevertheless, the effect of maternal heat stress exposure during different stages of pregnancy on fertility and ovarian reserve of the subsequent offspring has not been investigated to our knowledge.

Anti-Müllerian hormone (AMH) is a member of transforming growth factor β superfamily, which is primarily secreted by granulosa cells of healthy growing follicles [25]. Circulating AMH concentration is highly positively associated with ovarian reserve in bovine [26,27], human [25] and murine [28], and has minimal day-to-day variation within individuals over the course of reproductive cycles in bovine [27,29] and human [30]. Therefore, a single measurement of AMH obtained on any day of the estrous cycle has been reasonably proposed to serve as a reliable phenotypic marker for ovarian reserve in cattle [27].

Accordingly, a retrospective study was performed to assess the effects of maternal exposure to heat stress during different trimesters of pregnancy on reproductive performance and AMH concentration in the next generation.

2. Materials and methods

2.1. Animals, location, climatic data

Animal Ethics Committee at University of Tehran approved the present study in terms of animal welfare and ethics.

The present study was conducted at a commercial Holstein dairy farm located in Varamin county, Tehran province, Iran (Latitude: $35^{\circ} 46' N$; Longitude: $51^{\circ} 65' E$; Altitude: 1200 m) with arid climate. Based on climatic data, cows were exposed to heat stress (temperature humidity index ≥ 72) [31] in June, July and August over the last 15 years (2001–2015; Fig. 1). Temperature-humidity index (THI) was calculated as follows: $THI = \text{dry bulb temperature} + (0.36 - \text{dew point temperature}) + 41.2$ [32]. In the herd, voluntary waiting period was 42 days and cows were inseminated 12 h after the observation of standing estrus. Estrus detection was performed thrice daily by visual observation for at least 30 min each time. All artificial inseminations were conducted by the same technician and pregnancy diagnosis was performed 40–45 days after AI by transrectal palpation.

2.2. Data

To investigate the effect of maternal exposure during different trimesters of gestation, we assumed that in addition to exposure,

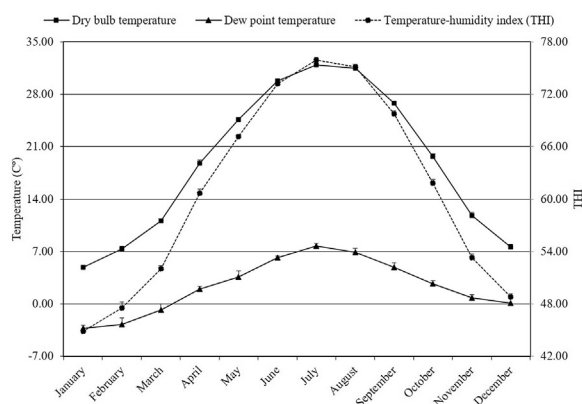


Fig. 1. Mean values for dry bulb temperature, dew point temperature and THI in different month from 2001 to 2015. Data were retrieved from the nearest meteorological station.

time of exposure would be of significance. As a result, our inclusion criteria was to choose the female calves of dams that had been exposed to heat stress for ≥ 2.5 months during the first, second or third trimester of pregnancy. Additionally, to ease the data collection and analysis, the length of gestation in bovine was considered nine months, and months 1–3, 4–6 and 7–9 were presumed as the first, second and third trimester of pregnancy, respectively. Time of successful insemination was considered as the commencement of conception; therefore, cows that had been conceived from June 1 to 15 were considered as first trimester exposed; cows that had been conceived from March 1 to 15 were considered as second trimester exposed; cows that had been conceived from December 1 to 15 were considered as third trimester exposed; and cows that had been conceived from September 1 to 15 were considered as unexposed. Data associated with calves conceived from 2003 to 2007 were collected from the herd database. Since we intended to investigate fertility parameters in the same population of each group over the first, second, third and fourth lactation periods, cows with missed data in this regard were excluded from the analysis. In total, data consisted of 206 cows, out of which 42, 61, 55 and 48 cows belonged to unexposed, first trimester exposed, second trimester exposed and third trimester exposed groups, respectively. Given that offspring in various aforementioned categories of the present study were born at different times of year and the corresponding difference would probably lead to disparity in time of calving and postpartum breeding among different groups and considering the adverse effects of heat stress on fertility of cows [17,18], data associated with heat stress exposure of the offspring at the termination of VWP (Day 42 postpartum) were collected as well to avoid this confounding effect and adjust the statistical analysis in this regard.

2.3. Reproductive parameters

Days to first service (DFS) was defined as the interval from parturition to first insemination. First service conception rate (FSCR) was defined as the proportion of cows diagnosed pregnant following first insemination postpartum. Cows that failed to conceive after 3 services were considered as repeat breeder (RB) cows. Service per conception (SPC) was defined as the number of services implemented to achieve conception in cows. Calving to conception (CCI) interval was defined as the number of days from parturition to conception. Days to first service, FSCR, RB, SPC and CCI were calculated using the data of cows diagnosed pregnant and the data of cows failed to conceive were not considered for calculation of the respective parameters. Culling rate (CR) was defined as the proportion of cows that failed to enter the next lactation period.

2.4. AMH assay

As aforementioned, cows with different history of prenatal exposure to heat stress were randomly subjected to blood sampling from various periods of lactation including the first, second, third and fourth periods. Cows received two administrations of PGF2 α (500 μg i.m.; Vetaprost[®]; Aburaihan Pharmaceutical Co., Tehran, Iran) 14 days apart beginning at day 28 postpartum. Afterwards, cows were monitored for detection of standing estrus, and blood samples were taken using venipuncture from tail vein of cows observed in estrus. Blood samples were centrifuged for 10 min at $2000 \times g$ within 2 h after collection. Serum was stored at -20°C until hormonal assay. AMH concentration was measured using AMH (Bovine) ELISA kit (Ansh Labs, TX, USA). In addition, data associated with heat stress exposure at the time of blood

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