Serum copper (Cu) and zinc (Zn) concentration in relation to super ovulatory response in embryo donor (Sahiwal) and conception rate in embryo recipient (crossbred) cattle

SHIV PRASAD1 and S N MAURYN

Govind Ballabh Pant University of Agriculture and Technology, Pantnagar, Uttaranchal 263145 India

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ABSTRACT

Serum copper (Cu) and zinc (Zn) levels were measured in embryo donor (Sahiwal) and embryo recipient (crossbred) cows in relation to super ovulatory response and conception rate respectively. Sahiwal cattle (19) were super ovulated with equine and porcine follicle stimulating hormone and they were divided into 3 groups on the basis of super ovulatory response (good, fair and non responders). The levels of copper and zinc in serum were higher in the animals, which had responded well to the gonadotrophin treatment in comparison to those responded fair and did not responded at all. The levels of copper ranged from 1.42±0.17 ppm to 2.15±0.11 ppm, 1.23±0.04 to 1.53 ±0.08 ppm and 1.36±0.10 to 1.75±0.13 ppm in good, fair and non responder animals, whereas, the levels of zinc (Zn) ranged between 2.38±0.18 to 2.83±0.22 ppm, 2.00±0.07 to 2.82±0.27 and 1.79±0.24 to 2.82±0.11 ppm in good, fair and non responder respectively. The animals, which conceived, had higher levels of copper in comparison to those, which did not become pregnant. The levels in pregnant animals varied from 1.95±0.16 ppm to 2.35±0.15 ppm, whereas in non-pregnant animals it varied from 1.73±0.13 to 2.23±0.13 ppm. The level of variation in the zinc among different days of pregnancy were not significant but variation in groups (pregnant and non-pregnant) was significant (P<0.01).

Key words: Animal reproduction, Copper, Embryo donor, Embryo recipient, Pregnancy, Superovulation, Zinc

Hypocuprosis in cattle is associated with female reproductive disorders; the most common symptom has been prenatal mortality, particularly embryonic loss. A low copper content in the diet either prevent implantation or induce embryonic loss and fetal death (McChow ell1968; McChow ell and Hall 1970). A high incidence of infertility in cattle in Australia (Bennets et al. 1948) was associated with cases of falling diseases, anaemia and retarded growth in areas where pasture copper was less than 3 ppm. The pituitary gland and ovaries of cows contained 9.5 and 4.8 µg copper/g dry weight (Stockl and Weiser 1968). The lower copper level in some repeat breeders cows was also observed by Prasad and Rao (1997). Zinc is an essential nutrient required for normal development, growth and tissue synthesis and may be a specific dietary factor modulating fetal growth. Many workers Dufty (1977) and Kumar and Vadve (1994) have documented low zinc levels in anestrous heifers than normally cycling cattle, as the plasma zinc levels tended to be higher at the time of estrus (1.50±0.27 µg/ml) than during the diestrus (1.01±0.16µg/ml). Zinc deficiency in relation to reproduction is documented as being responsible for reproductive infertility and defects in prostaglandin metabolism (Cunnae et al. 1983).

In the present study the relationship of copper and zinc was evaluated in relation to superovulation in embryo donor cattle and conception in embryo recipient cattle.

MATERIALS AND METHODS

The study was conducted at Livestock Research Centre of the University. The animals were selected on the basis of reproductive history, clinical and per rectal examination twice at the time of estrus. Embryo donors, Sahiwal (19) were super ovulated by using equine and porcine follicle stimulating hormone given as per following schedule. Donor animals were divided into 3 groups on the basis of super ovulatory response (good, fair and non responders). The blood samples from the embryo donor and embryo recipient cattle were collected as per following schedule.

**Super ovulatory treatment with FSH-P and FSH-E**

Super ovulatory treatment was initiated on day 10 of estrous cycle and was spread for 4 days. FSH-P was given @400 mg NIH in 8 divided doses I/M in descending dose manner; 130, 110, 90, 70 mg on day 10, 11, 12, 13 respectively. FSH-E was given @50mg in 8 divided doses and also in descending
dose manner 15, 13, 12, 10mg on day 10, 11, 12 and 13 respectively. Injecting prostaglandins on day 12 of estrous cycle at 25 mg i.m induced estrus.

Sampling schedule of embryo donor and recipient animals

**Embryo donor:** Blood serum samples from embryo donor animals were collected on day of I P.G. treatment, day of estrus, second day of estrus, fifth day of estrus, day of initiation of FSH treatment (10), day of II P.G. treatment (12), last day of FSH treatment (13), day of superovulation (S1), third day of superovulation (S2) and seventh day of superovulation (S3) or day of embryo collection.

**Embryo recipient:** Blood serum samples were collected on day of embryo transfer and on days 4, 7, 14, 21 and 28 post ET. The samples were preserved at -20°C in deep freezer till analysis.

Blood (15ml) was collected from jugular vein of embryo donor and recipient cow and blood serum was separated. The serum was stored at -20°C till analysis.

Before analysis of copper and zinc, the serum samples were first digested by nitric acid and triple acid mixture for the analysis. For digestion, 5ml of concentrated nitric acid was added to test-tube containing 1 ml of serum. It was then kept over a hot plate for 30 min. After cooling, 5 ml of triple acid (nitric acid, perchloric acid and sulphuric acid in the ratio of 10: 4: 1) was added into it. It was again kept on hot plate till the contents reduced to 1 ml. It was then cooled and volume was made to 10ml by adding triple glass distilled water and the estimation was done by atomic absorption spectrophotometer using following instrumental settings.

<table>
<thead>
<tr>
<th>Elements</th>
<th>Max. lamp Wave length (nm)</th>
<th>Slit setting (nm)</th>
<th>Flame</th>
</tr>
</thead>
<tbody>
<tr>
<td>Copper</td>
<td>5.0</td>
<td>324.75</td>
<td>0.4</td>
</tr>
<tr>
<td>Zinc</td>
<td>10.0</td>
<td>213.86</td>
<td>0.4</td>
</tr>
</tbody>
</table>

**RESULTS AND DISCUSSION**

In the present study the level of copper, in good responder animals ranged from 1.42±0.17 ppm to 2.15±0.11 ppm and in non-responders animals it was 1.36±0.10 to 1.75±0.13 ppm (Table 1). The higher level of copper in the animals which responded well to the gonadotrophin treatment are in agreement with Pradhan et al. (1998) and Vhora et al. (1998) who have reported higher values of copper in normal cyclic animal than post-partum anestrous animal.

Present findings agree well with the findings of Kalita et al. (1999). They have reported variations in the levels of copper in normal cycling, repeat breeder and anestrous cows though the levels were statistically non-significant (1.69±0.18 ppm, 1.48±0.19 ppm and 1.46±0.16 ppm respectively). The variation in the level of copper could be due to the stage of estrous cycle of the cow.
animals, which conceived, had higher levels of copper in comparison to those, which did not become pregnant (Table 2). Present findings of copper in pregnant and non-pregnant animals are also fitted well with the findings of Sarmah et al. (1999). They found significant increase in pregnant groups of non-descriptive cattle.

The values of zinc (Zn) differ significantly among different groups and they were significantly lower in non-responder animals to the super ovulatory treatments (Table 3). The levels were higher during the early pregnancy in comparison to non-pregnant animals.

In the present study, the level of zinc variations during pregnancy was significant (<0.05) in comparison to non-pregnant (Table 4). The levels were higher during the early pregnancy in comparison to non-pregnant animals.

Table 3. Serum zinc (Zn) levels (ppm; mean±SE) in embryo donor cows

<table>
<thead>
<tr>
<th>Donor response</th>
<th>Days from estrus</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>3 (PG)</td>
</tr>
<tr>
<td>Good</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Ba</td>
</tr>
<tr>
<td></td>
<td>±0.22</td>
</tr>
<tr>
<td>Good</td>
<td></td>
</tr>
<tr>
<td></td>
<td>±0.07</td>
</tr>
<tr>
<td>Non-responders</td>
<td></td>
</tr>
<tr>
<td></td>
<td>±0.59</td>
</tr>
</tbody>
</table>

Means within a row with different small superscript are significantly different (P<0.05); means within a column with different capital superscript are significantly different (P<0.05); means within a column with different capital superscript are significantly different (P<0.05).

REFERENCES


Sarmah B C, Kalita D J and Bhattacharya B N. 1999. Certain mineral profile and haemoglobin concentration in local nondescript and
