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HIgher concentration of haptoglogin indicates transient inflammation and negative energy balance in transition cows*

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Abstract

Haptoglobin is an acute phase protein that is of diagnostic significance in dairy cattle. In the present study, haptoglobin level was assessed during the periparturient period in 15 clinically healthy pregnant cows. The mean concentration of haptoglobin during transition was found to be 5.60 ± 0.54 mg/dL, whereas the concentration during the period immediately before and after transition was 4.80 ± 0.59 mg/ dL. The concentration obtained was greater than the reported concentration of 2mg/dL or less, in healthy ruminants. The higher concentrations observed were also associated with a higher concentration of serum non-esterified fatty acids of 0.576 ± 0.10 mmoles/L during transition and 0.328 ± 0.03 mmoles/L immediately outside the transition period. Increased haptoglobin might indicate the transient inflammation and a response to negative energy balance during transition in dairy cows.

Key words: *Haptaglobulin, Non esterified fatty acid, Transition*

Acute phase proteins (APPs) are plasma proteins synthesized by the liver in response to infection, inflammation or trauma. They play an important role in the trapping of microbes and their products, in triggering the complement system, in the binding of cellular remnants, in the neutralizing of enzymes which can otherwise harm the body, in scavenging free hemoglobin and radicals, and in altering the immune response of the host. APPs can be used as a tool for monitoring the health status of animals.

Haptoglobin (Hp) is one of the major positive APPs in cattle. It is a haemoglobin binding protein and has antioxidant and angiogenic properties. The hepatic synthesis of haptoglobin is stimulated by cytokines and glucocorticoids (Marinkovic and Baumann, 1990). This study was undertaken with the objective of gaining an understanding of the concentration of haptoglobin

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in periparturient dairy cows and to explore the possibility of it being used as a marker for stress during transition period (3 weeks before to three weeks after calving).

Materials and Methods

Blood was collected at fortnightly intervals, starting from four weeks before to four weeks after parturition, from 15 clinically healthy pregnant crossbred dairy cows in second to fifth parity maintained at University Livestock Farm and Fodder Research Station, College of Veterinary and Animal Sciences, Mannuthy and Cattle Breeding Farm, Thumburmuzhy. Serum was separated and stored at -20°C till use. Haptoglobin was assessed using haptoglobin (Hp) kit (Randox Laboritories Ltd.). The estimation of haptoglobin is based on the reaction between the anti-haptoglobin antibody and haptoglobin present in the sample and measuring the decrease in the absorbance at 340 nm. Five µL of sample and one ml of reagent 1 (haptoglobin buffer) were mixed and incubated for five min at 25°C. Readings were taken in a semi-automatic analyzer after the addition of 100 µL of Reagent 2 (reconstituted haptoglobin antibody and buffer) and further incubation for five min.

Non-esterified fatty acid content of the samples was estimated directly in a semiautomatic analyzer based on a colorimetric assay as defined by Hosaka *et al*. (1981) using the Randox NEFA kit from Randox Laboratories Ltd. The assay consisted of the measurement of hydrogen peroxide from free fatty acids by Acyl CoA synthetase and Acyl CoA oxidase and the quantitative determination of the absorbance at 550nm in the presence of peroxidase, 4-aminoantipyrine and N-ethyl-N-(2hydroxy-3-sulphopropyl) m-toluidine.

Results and Discussion

The mean concentration of haptoglobin during the transition period was 5.60 ± 0.54 mg/ dL and the concentration immediately outside the transition period (4 weeks before and after) was 4.80 ± 0.59 mg/ dL. The difference in haptoglobin levels between transition and outside transition was found to be significant ($p<0.05$) statistically.

The concentration of haptoglobin obtained during the entire study period was higher than the reported concentration of not more than 2 mg/dL in healthy ruminants (Kaneko *et al.*, 2008). Eckersall and Bell (2010) reviewed the importance of APP in veterinary medicine and reported that the concentration of serum haptoglobin was less than 2µg/dL in healthy cattle and increased in case of inflammation to a value greater than 200mg/dL.

Wide variations in haptoglobin concentration have been reported in apparently healthy dairy cattle by several authors (Chan *et al.*, 2004; Trevisi *et al.*, 2012). An elevated concentration of haptoglobin at calving which decreased to basal levels during later stages of lactation was observed by Bionaz *et al.* (2007). This has been reiterated by Bossaert *et al.* (2012) who reported an increase in the level of positive acute phase proteins in transition Holstein Friesian cows, which also returned to a basal level in the days following parturition. The increased haptoglobin reported in the present study might reflect transient inflammation associated with parturition and onset of lactation as reported by Bertoni *et al.* (2008) and Aleri *et al.* (2016). A marked increase in the concentration of haptoglobin and ceruloplasmin after calving was reported by Trevisi *et al.* (2015), especially during first week of lactation suggesting associated inflammatory events mediated by pro-inflammatory cytokines. The study was conducted on 21 multiparous Holstein Friesian cows during transition period. It could also be that crossbred cows in tropical countries possess higher haptoglobin level which needs to be verified in a larger population.

Hiss *et al*. (2009) reported that negative energy balance in cows can be related to the increased haptoglobin levels, as cows with increased milk haptoglobin also showed increased concentration of NEFA in serum. It is pertinent to note that in the present study, animals were in a state of negative energy balance as indicated by an elevated NEFA concentration of 0.576 ± 0.10 mmoles/L during transition and 0.328 ± 0.03 mmoles/L immediately outside the transition period.

Increased NEFA can lead to fatty liver which in turn can adversely affect liver function. Haptoglobin was reported to have a bacteriostatic action in cows with fatty liver and associated diseases and has a role in liver regeneration by scavenging the triglyceride accumulated hepatic cells (Kim *et al*., 1995). Kanno and Katoh (2001) determined the presence of haptoglobin in HDL and VLDL fractions of dairy cows with fatty liver suggesting its role in lipid metabolism. The higher concentration of haptoglobin could be protective to the liver in conditions that could lead to fatty liver.

Though a non-specific marker of inflammation, haptoglobin level after calving was found to be associated with a higher concentration of serum non-esterified fatty acids, an indicator of negative energy balance in transition dairy cows. The role of haptoglobin in fatty liver and associated disorders is suggestive of its use as a marker for transition risk in dairy cows.

References

Aleri, J. W., Hine, B. C., Pyman, M. F., Mansell, P. D., Wales, W. J., Mallard, B. and Fisher, A.D. 2016. Periparturient immunosuppression and strategies to improve dairy cow health during the periparturient period. *Res. Vet. Sci.* **108**: 8-17.

Bertoni, G., Trevisi, E., Han, X. and Bionaz, M. 2008. Effects of inflammatory conditions on liver activity in puerperium period and consequences for performance in dairy cows. *J. Dairy Sci.* **91**: 3300-3310.

Bionaz, M., Trevisi, E., Calamari, L., Librandi, F., Ferrari, A. and Bertoni, G. 2007. Plasma paraoxonase, health, inflammatory conditions, and liver function in transition dairy cows. *J. Dairy Sci.* **90**: 1740-1750.

Bossaert, P., Trevisi, E., Opsomer, G., Bertoni, G., De Vliegher, S. and Leroy, J. L. M. R. 2012.The association between indicators of inflammation and liver variables during the transition period in high-yielding dairy cows: An observational study. *Vet. J.*.**192**: 222-225.

Chan, J. P.W., Chu, C.C., Fung, H.P., Te chuang, S., Lin, Y. C., Chu, R. M. and Lee, S.L., 2004. Serum haptoglobin concentration in cattle. *J. Vet. Med. Sci.* **66**: 43-46.

Eckersall, P. D. and Bell, R. 2010. Acute phase proteins: Biomarkers of infection and inflammation in veterinary medicine. V*et. J.* **185**: 23-27.

Hiss, S., Weinkauf, C., Hachenberg, S. and Sauerwein, H., 2009. Relationship between metabolic status and the milk concentrations of haptoglobin and lactoferrin in dairy cows during early lactation. *J. Dairy Sci.* **92**: 4439-4443.

Hosaka, K., Kiteucki T., Mitsukida, N., and Kawagucki A. J. 1981. A new colorimetric method for the determination of free fatty acids with acyl-coA synthetase and acylCoA oxidase. *J. Biochem*. **89**: 1799-1803.

Kaneko, J. J., Harvey, J. W. and Bruss, M. L. 2008*. Clinical biochemistry of domestic animals.* (6th Ed.). Academic Press, San Diego. 916p.

Kanno, H. and Katoh, N. 2001. Analysis by enzyme-linked immunosorbent assay and 2-dimensional electrophoresis of haptoglobin in the high-density lipoprotein fraction in cows. *J. Vet. Med. Sci.* **63**: 61-66.

Kim, I. K., Lee, J. H., Kim, H. S., Kwon, O. J. and Shim, B. S., 1995. A novel function of haptoglobin: haptoglobin-haemoglobin complex induces apoptosis of hepatocarcinomatous Hep 3B cells. *Scand. J. Clin. Lab. Investigation*. **55***:* 529-535.

Marinkovic, S. and Baumann, H. 1990. Structure, hormonal regulation and identification of the interleukin 6 and dexamethasone responsive element of the rat haptoglobin gene. *Mol. Cell. Biol.* **10**: 1573-1583.

Trevisi, E., Amadori, M., Cogrossi, S., Razzuoli, E. and Bertoni, G. 2012.Metabolic stress and inflammatory response in high-yielding, periparturient dairy cows. *Res. Vet. Sci.* **93**: 695-704.

Trevisi, E., Jahan, N., Bertoni, G., Ferrari, A. and Minuti, A. 2015. Pro-inflammatory cytokine profile in dairy cows: consequences for new lactation. *Ital. J. Anim. Sci.* **14**: 3862. ■