Abstract

Effect of a bioflavonoid dietary supplement on acetaminophen-induced oxidative injury to feline erythrocytes

Robin W Allison, DVM; E. Duane Lassen, DVM, PhD, DACVP; Mary Jo Burkhard, DVM, PhD, DACVP; Michael R. Lappin, DVM, PhD, DACVIM, **Journal of the American Veterinary Medical Association.** Vol. 12, No. 8, October 2000:1157-1161

Flavonoids are naturally occurring compounds found in plants throughout the plant kingdom. They have been the subjects of a number of research studies in recent years investigating their antioxidant properties. This antioxidant characteristic is believed to be due to their ability to chelate metal ions, scavenge free radicals, and inhibit lipid peroxidation, hemolysis, and hemoglobin oxidation.

Erythrocytes are prone to oxidative injury when intracellular reducing pathways that utilize NADH, NADPH, and GSH (reduced glutathione) are insufficient to meet the oxidant challenge. Upon oxidation, hemoglobin becomes denatured and forms microscopically visible aggregates (Heinz bodies) that attach to the internal membrane of erythrocytes. Heinz body formation is associated with decreased deformability and shorter erythrocyte life span. Feline hemoglobin is very susceptible to oxidative injury, attributable to the 8 reactive sulfhydryl groups on the hemoglobin molecule and the easy dissociation of hemoglobin from the tetramer to dimer form.

Low erythrocyte GSH content, increased methemoglobin concentrations, and the formation of Heinz bodies have been reported in cats after acetaminophen challenge and exposure to other oxidants. The objective of this double blind research study was to determine the effects of a bioflavonoid antioxidant dietary supplement on measurable oxidative injury to feline erythrocytes induced by acetaminophen.

The study population was divided into three groups. Group one received one 10-mg. antioxidant (Proanthozone) capsule orally each morning from day one through day 14. This group did not receive an oxidative challenge and served as a control group. Group 2 cats received a single dose of acetaminophen (90 mg/kg) orally on day 7 and did not receive antioxidant. Group 3 cats received one 10 mg. antioxidant capsule orally each morning from day 1 through day 14 and also received a single dose of acetaminophen (90 mg/kg) orally on day 7.

Packed cell volume, Heinz body count, and GSH and GSSG determinations were performed on days 8, 10, 11, and 14 in all cats. All cats that received antioxidant alone (group 1) remained clinically healthy during the study period. Among group-2 and group-3 cats, transient cyanosis and lethargy were observed during the first 12 hours after administration of acetaminophen. Cats did not require medical intervention, and all were clinically normal within 24 hours.

Acetaminophen administration resulted in a sustained significant increase in percentage of Heinz bodies in groups 2 and 3 compared with values in the control group. Although Heinz body percentage steadily increased in group-3 cats, the magnitude of increase was significantly less ($p=0.018$) than that of group-2 cats and did not continue to increase after day 8. Results of the study indicate that administration of the antioxidant had a significant protective effect against Heinz body formation after acetaminophen challenge.

Oral administration of bioflavonoid antioxidants to cats at risk for oxidative stress may have a beneficial effect on their ability to resist oxidative injury to erythrocytes. Cats are uniquely sensitive to oxidative injury to erythrocytes, and Heinz body formation as the result of exogenous oxidant exposure as well as disease processes such as lymphoma, diabetes mellitus, and hyperthyroidism, has been well documented. Dietary treatment that would minimize oxidative injury could be of benefit to sick cats and may be a useful protective supplement in healthy cats. Results of the study reported here suggest that daily oral supplementation of cats with a bioflavonoid antioxidant provides therapeutic and protective support, at least when supplementation begins prior to exogenous oxidative stress.

**See reverse for graphs*

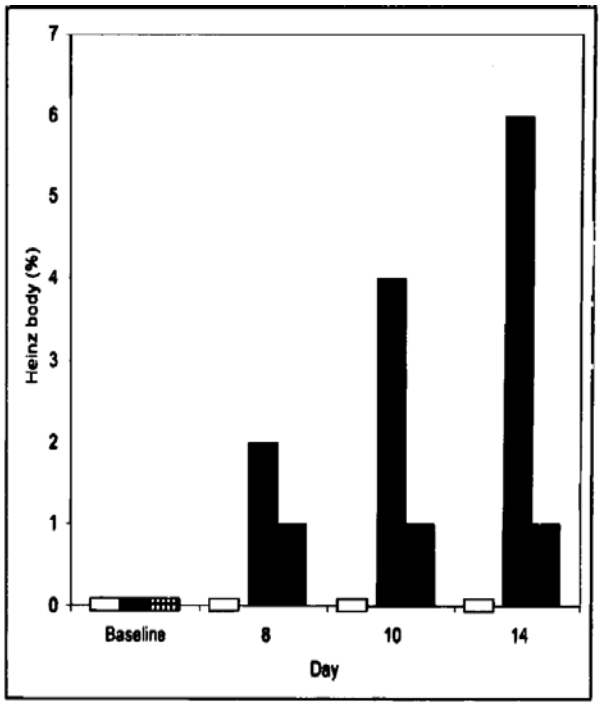

Figure 1—Median percentage of erythrocytes containing Heinz bodies in cats (n = 15/group) that received bioflavonoid antioxidant orally for 14 days (□), that did not receive antioxidant and were given acetaminophen on day 7 (■), or that received antioxidant orally for 14 days and were given acetaminophen on day 7 (⊞).

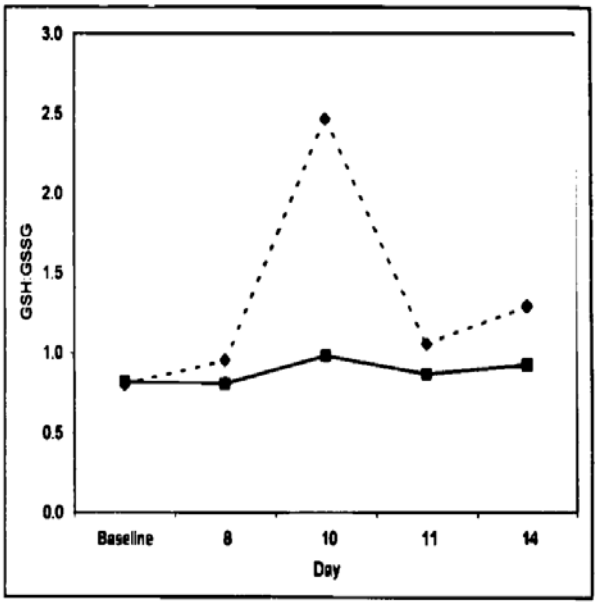

Figure 2—Median ratios of the concentrations of reduced (GSH) and oxidized (GSSG) glutathione in blood specimens of cats (n = 15/group) that did not receive antioxidant and were given acetaminophen on day 7 (dashed line) or that received oral administration of antioxidant for 14 days and were given acetaminophen on day 7 (solid line).

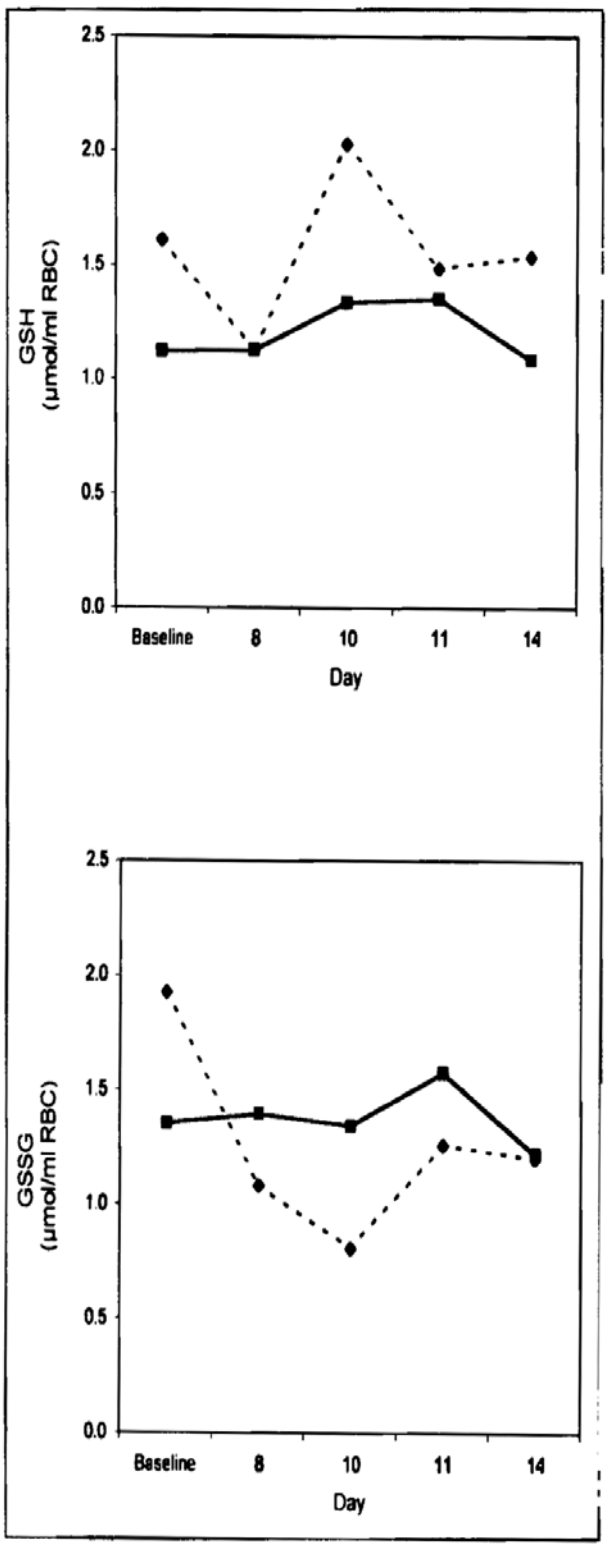

Figure 3—Median GSH (upper panel) and GSSG (lower panel) concentrations in blood specimens of cats (n = 15/group) that did not receive antioxidant and were given acetaminophen on day 7 (dashed line) or that received antioxidant orally for 14 days and were given acetaminophen on day 7 (solid line).