

# Effect of a Soluble Cobalt, Selenium and Zinc Glass Bolus on Humoral Immune Response and Trace Element Status in Lambs

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## Introduction

Trace element deficiencies are common in many countries and affect animal health, productivity and welfare. Trace elements that can be limiting in animal nutrition include copper, cobalt, selenium and zinc. Deficiency diseases may be manifested as a consequence of a single or a multiple element deficiency. Deficiency in any of the above trace elements can result in an increased disease susceptibility and a decreased immune function. A soluble glass bolus has been developed for the sustained release of cobalt, selenium and zinc to sheep. This bolus is similar to the commercial Co/Se/Cu soluble glass (Cosecure®) but has zinc replacing copper.

## Materials and Methods

Thirty-four Suffolk cross store lambs were allocated to two groups by restricted randomisation of body weight (mean 27.8 kg, s.d.  $\pm$  3.6), and were kept at pasture on the University of Leeds Farms throughout the trial. On day 0, one group was bolused with the cobalt, selenium and zinc soluble glass boluses (bolused), whilst the other group was left unbolused (controls). The boluses weighed approximately 33g with a composition of 13.1% zinc, 0.5% cobalt and 0.15% selenium. Immune function was measured in the lambs by measuring their humoral immune response to a novel antigen, keyhole limpet haemocyanin (KLH). On day 34, lambs were immunised with 1 mg KLH (Calbiochem, San Diego, California, USA) precipitated in alum and given subcutaneously at a site over the ribs. Blood samples were taken for the antibody response on days 20, 42, 49 and 63 and serum was stored at  $-20^{\circ}\text{C}$ . Lamb anti-KLH IgG responses were measured by a direct ELISA method (Mackenzie et al. 1996). Blood samples were taken for assessment of trace element status on days 0, 20, 42 and 63. Cobalt status was assessed by measuring vitamin B<sub>12</sub> concentrations by radioassay kit (Becton Dickinson, Oxford England). Selenium status was assessed by colorimetric assay of erythrocyte glutathione peroxidase activity (Telfer et al. 1984). Plasma zincs and coppers were diluted 1:5 with 0.1 M HCl (Analar, BDH) and read by Atomic Absorption spectrophotometry (Pye SP9 AA spectrophotometer, zinc at 213.9 nm with background correction, copper at 324.8 nm). The sheep were slaughtered for recovery of boluses and collection of liver samples. Livers were analysed after freeze drying and microwave wet digestion (70% HNO<sub>3</sub>, Analar) using the same instrument parameters as for plasma. Recovered boluses were oven dried and weighed for calculation of dissolution rates.

Table 1. Effect of bolusing on lamb liveweight (kg).

Day	Bolused	Controls	SE
0	27.77	27.79	0.896
20	32.32	33.29	1.006
42	35.27	35.71	0.993
63	37.32	36.24	1.046

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## Results

Figure 1 shows the bolused lambs having significantly greater anti-KLH IgG levels on day 42 ( $p < 0.05$ ) and day 63 ( $p < 0.01$ ). Serum vitamin B<sub>12</sub> concentrations are illustrated in figure 2 and were significantly greater ( $p < 0.001$ ) in the bolused group on all post-bolusing samplings with many of the controls being deficient (mean ~260 pg/mL) at day 42. Erythrocyte glutathione peroxidase activities are illustrated in figure 3 and were significantly increased ( $p < 0.001$ ) in the bolused group on all post-bolusing samplings. Figure 4 shows the bolused lambs having higher plasma zinc concentrations on day 42 ( $p < 0.05$ ) and day 63 ( $p < 0.01$ ).

There was no significant effect of bolusing on liver copper and zinc concentrations, plasma copper concentrations or liveweight of the lambs. The average bolus dissolution rate was 326 mg glass/ day (s.d 30 mg/day) giving a daily release of 45.6 mg zinc, 1.6 mg cobalt and 0.5 mg selenium.

## Discussion

The bolus release rate (quoted in parentheses) was adequate for all three trace elements with dietary requirements of 20–30 (45.6) mg Zn/d, 0.1–0.2 (0.5) mg Se/d and 0.1–0.2 (1.6) mg Co/d given a dietary intake of ~1 kg dry matter. These release rates are below the maximum tolerable levels of 750, 2 and 10 mg/kg DM for zinc, selenium and cobalt respectively (N.R.C. 1985). The bolused lambs had significantly higher serum vitamin B<sub>12</sub> concentrations and erythrocyte glutathione peroxidase activities on all

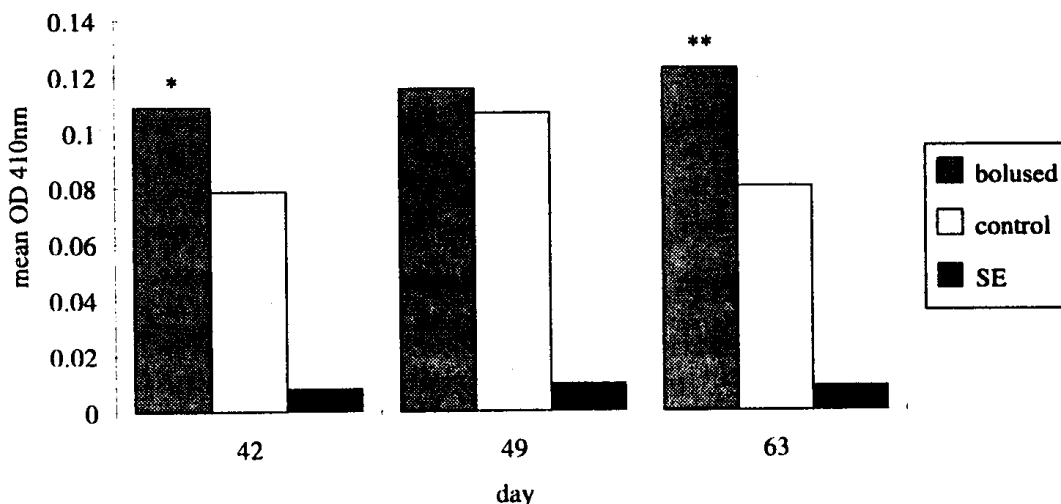


Figure 1. Humoral immune response after challenge on day 34 after bolusing.

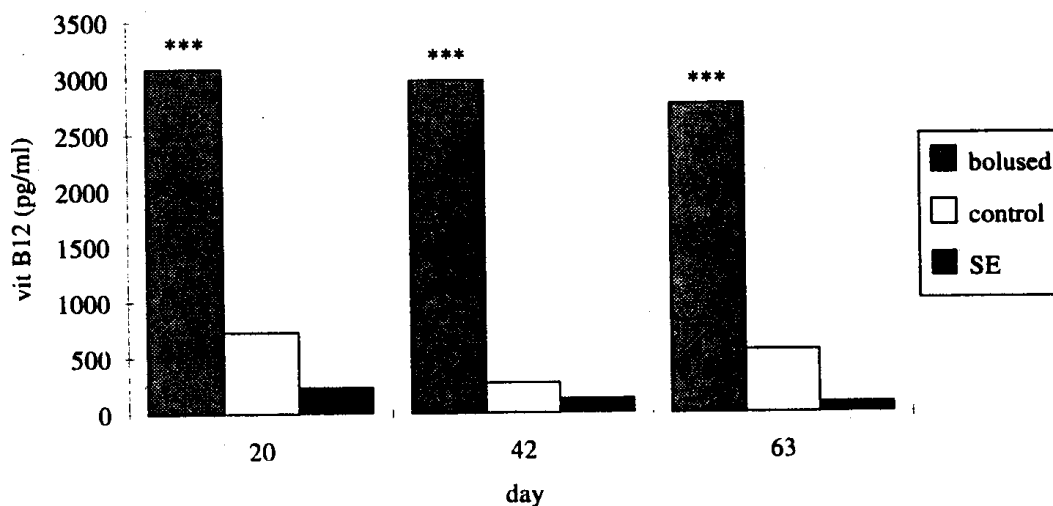


Figure 2. Vitamin B<sub>12</sub> concentrations after bolusing.

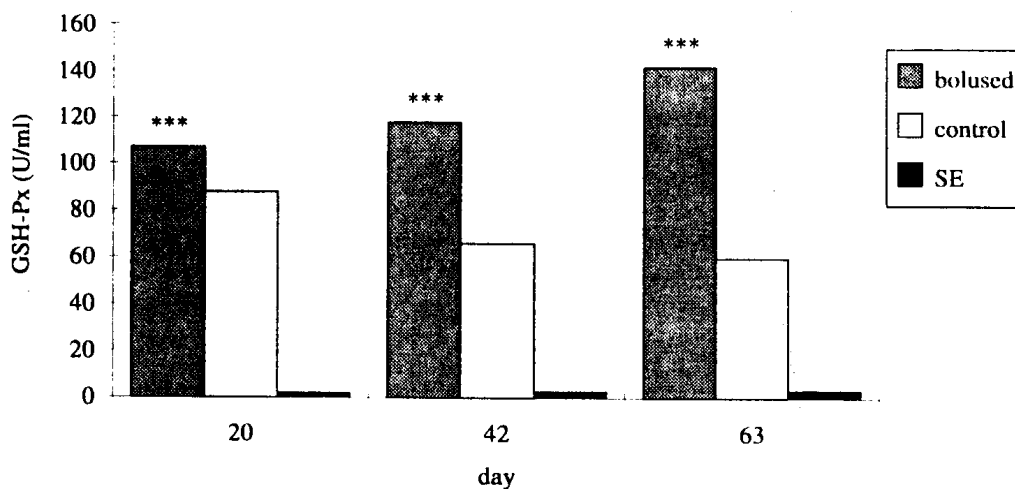


Figure 3. Glutathione peroxidase activities after bolusing.

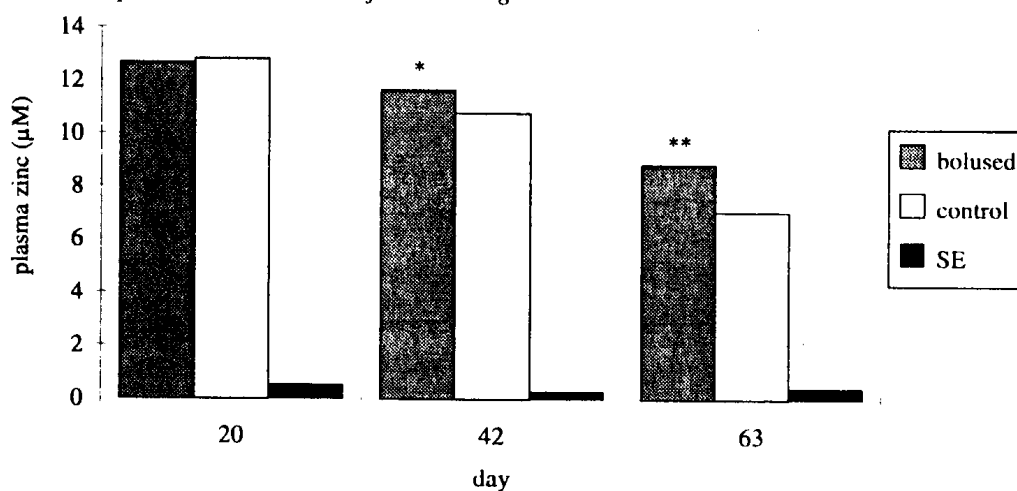


Figure 4 Plasma zinc concentrations after bolusing.

sampleings after bolusing, with significantly greater plasma zinc concentrations on days 42 and 63 with no depression in plasma copper. The significant increase in the levels of anti-KLH IgG in the bolused lambs may be due to an increase in the secretion rate of the antibody or an increase in the number of plasma cells. Previous reports have shown that cobalt, selenium and zinc affect the immune response in ruminants (MacPherson et al. 1987, Pollock et al. 1994). In this study it is not clear if the effect of the bolus on lymphocyte function is due to a single element or a combination of two or three elements. To conclude, bolusing with the cobalt, selenium and zinc bolus resulted in an increased antibody response and an elevated cobalt, selenium and zinc status of the lambs.

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