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Nutrition

Effects of selenium supplementation on selenium status of farmed fallow deer in outdoor pens

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ABSTRACT

The study investigated the effects of selenium (Se) supplementation on Se status in farmed fallow deer. Fallow deer were housed on grass pasture and adapted to consume ~200 g of pelleted grain daily. Animals were divided into two groups. One group received pelleted grain enriched with sodium selenate for 12 weeks (Se+ group, $N = 10$). Se intake for the first 7 weeks was 0.18 mg/kg dry matter (DM) and 0.32 mg/kg DM for the subsequent 5 weeks. The control group was fed pelleted grain without extra Se (Se− group, $N = 9$, 0.06-0.08 mg/kg DM). Blood samples were collected at the beginning and the end of the experiment. After the animals were slaughtered, tissue samples were collected for analysis of Se concentrations and Se-dependent glutathione peroxidase 1 (GPx1) activity. In addition, Se-independent α-glutathione-S-transferase (α-GST) activity was analyzed in liver tissue. Se supplementation significantly increased Se levels in plasma and in tissues as follows: liver > spleen > skeletal muscle > myocardium > kidney. Se supplementation also significantly increased GPx1 activity in tissues in the following order: liver > skeletal muscle > spleen = myocardium > kidneys. However, hepatic α-GST activity did not differ between Se+ and Se− groups. As expected, Se supplementation increased blood and tissue Se concentrations and GPx1 activity, which suggests a better antioxidant status. However, the activity of α-GST, an important Se-independent antioxidant enzyme, was not altered, presumably because GPx provided an adequate antioxidant capacity even though Se intake was low.

Introduction

Studies on selenium (Se) metabolism in domesticated ruminants, including cattle, sheep and goats, have clarified dietary requirements and helped to develop protocols to diagnose and prevent deficiencies [1–3]. Se concentrations in serum and various tissues, particularly the liver, are reliable indicators of Se metabolism [3,4]. Additionally, Se-dependent glutathione peroxidase (GPx) activity serves as an indicator of Se metabolism since GPxs have antioxidant functions. Of the eight different GPx isoforms, only five, including ubiquitous cytosolic GPx (GPx1), plasma GPx (GPx3) and phospholipid hydroperoxide GPx (GPx4), can reduce hydrogen peroxide and lipid hydroperoxides by using glutathione as a cofactor [5]. Compared to plasma GPx3 whole blood GPx activity (GPx1 activity in erythrocytes and plasma GPx3) may have a more delayed response to changes in Se intake as it strongly depends on newly formed erythrocytes [3].

Muscular dystrophy has been associated with an insufficient Se intake in ruminants [1]. In subclinical Se deficiency, reduced reproductive performance or diminished immunity may occur [6].

In wild cervids like moose, red deer, roe deer, white-tailed deer and reindeer, serum and liver Se levels suggest low to medium Se intake [4,7–12]. Se intake strongly varies by geographic location and animal population density. In Europe, most plants contain low Se levels; however, lichens in areas of Norway contain comparatively higher Se concentrations [4].

The health significance of low Se intake in cervids remains an open question as only a few reports of nutritional myodegeneration have been published [13–15]. Thus the question is raised if the low

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Se intake in Europe is considered marginal for wild cervids as it is for cattle. Reference values have only been established so far for red deer and reindeer [8,10], but not for fallow deer.

In Europe, interest is increasing for raising fallow deer (*Dama dama*) as a meat-producing animal. Currently, information is sparse regarding Se metabolism in fallow deer kept in outdoor pens. It is already well known in various mammal species that Se supplementation increases Se concentration and GPx activities in blood and tissues according to their hierarchy [1,2]. Se supplementation strategies in such systems may include pasture Se fertilization, Se injection or oral Se supplementation via minerals [16]. The present study was aimed at investigating the effects of Se supplementation on Se concentrations and GPx activity in blood, serum and various tissues of fallow deer. Se supplementation was expected to increase Se levels in the various tissues and lead to higher GPx activity.

Materials and methods

Animals

Nineteen fallow deer (male, age 16 months, mean body weight [BW] ± SD 44 ± 2.4 kg) were used in this experiment. The herd of fallow deer is owned by the Faculty of Veterinary Medicine in Leipzig. Fallow deer were kept on a grass pasture (in total 12 ha, divided into smaller sections) with a shelter hut (3 m × 4.1 m × 2.45 m) on each section. Fallow deer were regularly dewormed twice a year. Immediately before starting with the study, all animals were dewormed with 0.2 mg ivermectin/kg BW subcutaneously.

Reproduction rate varied around 74–78% (~70 female fallow deer, 52–55 living offspring each year). In the herd, there were no clinical symptoms of disorders before starting the study. During the experimental period, there were no clinical signs of health problems in fallow deer.

The project (A23/07) was approved by the Ethics Committee for Animal Rights Protection of the District government, in accordance with German legislation for animal rights and welfare.

Diets

Fallow deer were adapted to consume ~200 g of pelleted grain (barley and wheat) per animal once a day, which was provided in large buckets placed in the pasture. During the adaptation period, all animals were trained to come to the buckets at 5:00 p.m. Within 14 days, all animals consumed the whole grain meal with no leftovers.

At the beginning of the study, fallow deer were placed on an idle pasture allocated into two groups by dividing the pasture into two parts (~5000 m^2 each) by an electric fence (height 2 m). A shelter hut (3 m × 4.1 m × 2.45 m) was located in each area.

Ten animals received Se-enriched pelleted grain daily (Se+ group; sodium selenate, ~200 g pelleted grain per animal). From week 1 to 6, Se intake in the total ration (grass + Se-enriched pelleted grain) was approximately 0.18 mg/kg dry matter (DM), assuming a dry matter intake (DMI) around 3% of BW. The Se intake was adjusted according to the Se requirement for small ruminants (0.1–0.2 mg Se/kg DM). Due to a mixing error, Se intake was approximately 0.32 mg/kg DM from week 7 to 12.

Another nine animals received daily a pelleted grain without Se-enrichment (Se− group, ~200 g pelleted grain per animal). From week 0 to 12, Se intake in the total ration (grass + pelleted grain) was about 0.06–0.08 mg/kg DM given an estimated DMI around 3% of BW.

Table 1
Se content in feedstuffs and calculated Se content in total rations for Se+ and Se− groups, values are expressed in mg/kg DM.

Week	Se+			Se−		
	Grain	Grass	Se total ration[a]	Grain	Grass	Se total ration[a]
0–6	1.1	0.04	0.18	0.15	0.04	0.06
7–12	1.9	0.07	0.32	0.15	0.07	0.08

[a] Assuming an estimated DMI around 3% of BW according to Kamphues et al. [17].

The pelleted grain was provided once a day at 5:00 p.m., and constant feed intake of individuals was monitored daily for the entire experiment.

Feed samples of grass and pelleted grain (with and without Se enrichment) were collected at weeks 0, 6 and 12 of the supplementation period. For the grass samples, 10 small grass samples (~500 g) were collected and mixed to a large sample size (~5 kg) for each pasture and collection day. The large sample was manually homogenized and ~1 kg was stored at −20 °C until analysis. The pelleted grain samples (2 kg) were collected after processing and stored at −20 °C until analysis. Crude nutrients and Se content of the feedstuffs and estimated Se intake are summarized in Tables 1 and 2.

Fallow deer had free access to tap water (no nutrient analysis) ad libitum which was provided in buckets.

Blood and tissue collection

On the morning of the first day of the experiment, prior to supplementation, fallow deer were captured and restrained. Each animal was weighed (Wiegesystem FX1, Texas Trading, Windach, Germany) and given an identifying earmark. Blood was collected by venipuncture of the V. jugularis externa. Blood was placed in two tubes containing lithium-heparin (15 I.E. heparin/mL blood, Sarstedt, Germany). One tube was centrifuged (12,000 × *g* for 10 min) within 10 min of collection, and the plasma was stored at −80 °C until analysis. One tube was cooled until analysis.

After deworming, animals were randomly separated into two groups (Se+, *N* = 10; Se−, *N* = 9). Groups were housed separately in neighbouring sections on the grass pasture.

After week 12 all animals were slaughtered by captive-bolt-stunning. Immediately after captive-bolt stunning, blood was collected from the carotid artery. Clotting of whole blood reduced the sample number of animals at the second blood sampling point (Se+, *N* = 6; Se−, *N* = 3). Blood samples were handled as already described.

After bleeding, skinning and exenteration carcass weight was obtained by scale (MHW 10, Fa. ADE, Hamburg, Germany).

Tissue samples (~60 g wet weight) from spleen (margo cranialis), liver (lobus quadratus), right kidney (renal cortex), skeletal muscle (M. adductor magnus) and myocardium (apex) were collected after a standardized protocol postmortem, shock-frozen in liquid nitrogen and stored at −80 °C until analysis.

Table 2
Crude nutrients in pasture grass for Se+ and Se− groups, values are expressed in % DM.

Week	Se+					Se−				
	CA	CP	CF	CL	NfE	CA	CP	CF	CL	NfE
0	9.0	16	26	3.0	47	10	15	26	3.0	46
6	10	15	27	2.0	47	10	15	27	2.0	46
12	9.0	14	28	2.0	47	12	13	27	2.0	46

CA, crude ash; CP, crude protein; CF, crude fibre; CL, crude fat, NfE, nitrogen-free extractives.