## Selenium status in cattle herds in Wallonia (Belgium): overview and health management

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## Abstract

Aim: Monitoring was performed in order to determine the Selenium (Se) status of cattle herds in different agricultural areas in Wallonia (Belgium).

**Materials and Methods:** The study included 114 heifers and 184 cows (82 dairy and 102 beef cows) from 66 cattle farms situated in Wallonia. The Se status was assessed by measuring the glutathion peroxydase in red cells and converting it to the equivalent Se blood content.

**Results:** The average blood concentrations of Se were very low. The Ardennes region was an area where the lowest Se status was recorded. The highest levels of Se in dairy cows and beef cows were recorded respectively in the limoneuse region and Famenne areas. The Se content in beef herds was lower compared with that of dairy herds (35 vs. 56  $\mu$ g/L, p<0.01).

**Conclusion:** On average over all the regions, 87% of animals were classified as deficient and only 13% of the animals were classified as adequate. The heifers from both dairy and beef herds in Wallonia exhibited a deficiency in Se classified as moderate to severe. The beef herds showed larger deficiencies compared with the dairy herds. Selenium deficiency can be prevented by ensuring adequate supplementation of deficient animals in Se deficient regions. An increased consumption of vitamin-mineral supplements, the use of Se-enriched fertilizers and ingredients containing high levels of Se can help to reduce or correct deficiencies recorded in cattle herds in Wallonia.

Keywords: cows, glutathione peroxidase, heifers, selenium deficiency.

## Introduction

Selenium (Se) status is an important indicator in animal and human health [1-3]. Selenium is a trace element involved in the composition of specific selenoproteins. These and some other Se compounds are involved in the prevention of cardiovascular diseases, cancers and further pathologies in humans [4], or of white muscle disease in sheep and cattle [5,6]. Selenium deficiency also causes a decrease in neutrophil activity during the perinatal period, as well as an increase in placental retentions and fertility disorders [7]. This micronutrient possesses the ability to reduce oxidative stress and thus, the impact in some economically important diseases in dairy cattle, such as mastitis [2] and their associated effects on milk quality [3].

In Wallonia, the Southern part of Belgium, the agricultural regions are characterized by differing soil and elimatic conditions. Consequently, grass and crop growth can vary according to the agricultural region, thus affecting the diet of cattle.

The Se concentration in soil is low in Belgium, ranging between 0.14 and 0.70 mg/kg. Consequently,

it is low in plants (50  $\mu$ g/kg dry matter (DM)) [8,9]. Therefore, Se requirements estimated at 300  $\mu$ g/kg DM for all types of dairy cattle [10] are not met. Selenium deficiency in animals can induce negative consequences in humans, owing to the relationship between Se content in the fodder and blood of the animals and also the relationship between Se content in the blood and muscles, [11] along with the milk [12].

The purpose of this study was to assess Se status in farms, located in different agricultural regions in Wallonia, in order to highlight potential Se deficiencies and to measure their importance in heifers and cows of both beef and dairy herds. The purpose was also to classify the agricultural regions according to the deficiencies importance.

#### Material and Methods

#### **Ethical approval**

The research program was conducted in accordance with local laws and regulations.

## Description of the area of the study

The study was carried out on 66 cattle farms, belonging to an agricultural co-operative (SCAM), located in Wallonia (50° 30' North; 4° 45' East, Figure-1). There were 29 farms in Condroz, 21 farms in the limoneuse region, 10 farms in Ardennes and

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**Figure-1:** Map of Belgium and of the different agricultural areas in Wallonia.

five farms in Famenne. According to Peeters [13] these four agricultural regions could be described as follows: The soils of the Ardennes plateau are mainly made from schist or sandstone. They are essentially acidic and nutrient-poor. There is no limestone. Owing to an average altitude of 500 m above sea level, the climate is colder and rainier than other regions. The mean annual rainfall is higher than 1000 mm. Ardennes is a beef-oriented region. Grassland covers more than 75% of the agricultural area. Temporary pastures are the main source of conserved forages while permanent pastures are exclusively grazed. It is possible to produce spelt and some spring cereals if they are not exposed to severe frosts.

Famenne is a rather flat area characterized by an average altitude of about 100-200 m above sea level. It is a grassland-dominated region where cattle provide the majority of farmer's incomes. A most of the agricultural area, including permanent grasslands, is devoted to forage. However, there are still some cereals, and oilseed rapes included in the crop rotation.

The Condroz is a hillier region with many small crests, at an average altitude of about 250 m above sea level. Permanent pastures are found in calcareous depressions. Crop production happens in places with adequate loess deposits. It is a transition region between the arable farming systems of the loamy region and the livestock-based systems of Ardennes. Cash crops (winter wheat, winter barley, oilseed rape) and forage crops are important. Permanent grasslands cover 36% of agricultural area. Most farms are involved in both livestock and annual crop productions. Beef cows are much more abundant than dairy cows.

At an altitude of about 150-200 m above the sea level, the soils in the limoneuse region are loamy and deep, up to 20 m deep in some areas. The conditions are favourable for annual crop production. Cash crops cover almost 80% of the agricultural area. The areas producing cereals, sugar beet, potato and chicory are the largest of the country. Permanent grasslands are mainly located on the wet soils of the valleys and are not easily ploughed. So, they are mainly grazed by beef animals and dairy heifers, often at very high stocking rates. Some pastures, situated on well-drained soils and close to farms, are used for dairy cows.

## Animals sampled

A total of 298 animals were evaluated, including 114 heifers (26 dairy and 88 beef) and 184 cows (82 dairy and 102 beef). They belonged to 36 dairy cattle herds and 52 beef cattle herds. The beef herds were composed mainly of Belgian blue cattle, and the dairy herds were composed of the Holstein breed. The number of sampled animals was proportional to the cattle population found in the four agronomic regions. Average ages were  $44\pm15$  months for the cows and  $18\pm6$  months for the heifers. Two to five blood samples were obtained from each farm. The samples were taken from healthy animals at the end of the winter period before the animals went to the grass. Blood was taken from the jugular vein in the morning, 1-2 h after feeding.

## Selenium assessment

The glutathione peroxidase (Gpx) activity in erythrocytes was measured according to the method described by Paglia and Valentine [14]. Selenium concentration in the blood was then estimated from Gpx activity according to the equation developed by Kinoshita *et al.* [15]: y=20.94x-6.04 (y represents Se concentration in ng/ml and x the Gpx in mmol/min per ml).

## Statistical analyses

Statistical analyses were conducted by age and animal category, using a generalized linear model (Proc Glm, SAS 1999) and included the effect of the region. Owing to similar management of heifer's herds during the growing period and due to the lack of significant differences in Se status between dairy and beef heifers, heifers from both herd types were analysed as a sole group. Multiple comparisons of means between region and animal type were performed using the Student's t-test adjusted by the Tukey method. The means were considered significantly different at p<0.05.

## **Results and Discussion**

The thresholds used to classify the Se status of cattle monitored were: Severely deficient: 0-50  $\mu$ g/L; marginally deficient: 51-80  $\mu$ g/L; adequate: 81-161  $\mu$ g/L and highly adequate: 161  $\mu$ g/L or higher [16].

There are several reference values on Se status in cattle reported in the literature. In order to assess this status, measurements are performed directly (by analysis of Se) or indirectly (measuring the Gpx activity) on different matrices. In fact, these references may correspond to measurements performed on whole blood [17-21], plasma [22] or whole milk [23]. Other authors consider the activity of Gpx in the blood [24] or in the tissues such as kidney [25]. There are also equations [11] to estimate the Se content in a matrix through a measurement made on another matrix.

## Classification of individual animals according to selenium blood levels

The Se status of most of the animals recorded was below 80  $\mu$ g/L, a concentration regarded as adequate.

On average, across all regions, 87% of the animals were classified as deficient in Se and only 13% of the animals were classified as adequate (Figure-2). Similar frequencies of Se deficiency in cattle were observed in other European countries. In a similar study in Estonia [26], 34% of animals were classified as non-deficient. The same percentage was recorded in Slovenia [27]. The slaughter of 205 cattle in Greece showed that only 5% of the animals were character-ized by an adequate level of Se [28]. The percentage of animals deficient in Se in the Czech Republic was 50% [29]. A study two decades ago in the Western part of Germany [30] showed that the proportion of cattle with an adequate Se status was only 25%.

## Selenium blood concentration in cattle herds according to the animal category

The average Se blood concentrations of the three categories of animal studied (heifers, dairy cows and beef cows) were all low (Table-1). The Se content in blood of beef cows was lower than that of dairy cattle (35 vs. 56  $\mu$ g/L, p<0.01). The difference recorded between the two types of herds has to be associated with the ration of each herds. The diets of cattle on farms in Wallonia consist of a varying mix of cereals, fresh or preserved grass, maize as whole maize silage, protein and/or energy supplements, or by-products



**Figure-2:** Classification of the animals according to the Selenium content in the blood. Adequate= $80-161 \mu g$  Se/L, marginally deficient= $51-80 \mu g$  Se/L, severely deficient= $0-50 \mu g$  Se/L.

**Table-1:** Mean concentrations of blood selenium ( $\mu$ g Se/L whole blood) in heifers and cows according to the animal categories in Wallonia (Belgium)\*.

Animal categories	n	Mean	Standard error	Animal classification %			
				SD	MD	Α	НА
Heifers	114	46ª	2.4	63	26	11	0
Dairy cows	82	56⁵	2.8	52	21	27	0
Beef cows	102	35°	2.5	80	16	4	0

a,b,c=Means with a different letter are significantly different, n=Number of animals, SD=Severely deficient, MD=Marginally deficient, A=Adequate, HA=Highly adequate, \*Se was determined indirectly by Gpx activity measurement. Se concentration was calculated from results of Gpx determination according to the equation y=20.94x-6.04 where y represents Se concentration in ng/ml and x the Gpx in m mol/min per ml, Gpx=Glutathione peroxidase,

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of beet (fresh, ensiled or dried beet pulp, molasses) and mineral mixtures [31]. Dairy herds receive commercial feedstuffs and mineral mixtures, both generally supplemented with Se. By contrast, beef herds are usually fed with locally produced feedstuffs, like grass and preserved grass. As mentioned in the introduction, plants in Belgium are low in Se (0.05 mg/kg DM) [8]. Similarly, the Se content in Belgian soil is also low at 0.14-0.70 mg/kg [9]. This influences the content of Se in some locally grown products [32]. According to Waegeneers et al. [33], the Se content of beef meat in Belgium is estimated at 140 µg/kg and at 30 µg/kg for milk. The Se content in Belgian eggs from commercial farms is 197 µg/kg of egg [34] or 230  $\mu$ g/kg [33]. 31% of Se consumed by the Belgian population stems from meat and meat products, 4% originates from milk and milk products and 4% from eggs [33].

There is little information on the ability of plants in Belgium to accumulate Se. This lack of information is due to the deficiency of plant-available forms/ levels of this trace element in the soils, the absence of seleniferous soils in Western Europe and the floristic composition of the forages, which changes from year to year [8]. In trials conducted in Belgium, only a fraction of the Se applied by the nitrogen fertilizer, varying from 23% to 52%, was absorbed by plants [35]. There are many factors influencing the bioavailability of Se in soil, such as redox status, pH, rainfall, type of available Se in the soil, and the low or high capacity of cultivated fodder to store Se [36]. Furthermore, the presence of other minerals such as calcium, sulphur, copper and arsenic in the soils can interfere with the Se utilization by plants [37].

# Selenium blood concentrations in heifers, beef cows and dairy cows according to the agricultural area

Mean Se concentrations in the blood of heifers, according the agricultural areas, are given in Table-2. Blood Se concentration ranged from 34 to 52  $\mu$ g/L. No region, neither in dairy nor in beef herds was characterized by an adequate Se status. Se content in the blood varied from 44 to 64 µg/L and 32 to 55 µg/L for dairy and beef cows, respectively (Tables-3 and 4). Animals in Ardennes had the lowest Se concentration and were all classified as deficient. This observation can be explained mainly by the type of local agronomic productions and the rations distributed to cattle. In practice, the diets of Ardennes herds consist of grazed grass in summer and grass silage in winter [38]. It should be noted that, according to the data of the agricultural census in 2010, grasslands represented 75% of the usable agricultural area (UAA) in this region. The highest levels of Se in dairy cows were recorded in the limoneuse region (64  $\mu$ g/L) where grasslands cover only 19% of the UAA of the region. Large areas in limoneuse region are devoted to cereals which are crops storing Se in their seeds as Selenomethionine (over 60%) [39]. Consequently, the Se supply in cow diet was more diversified in the

**Table-2:** Mean concentrations of blood selenium ( $\mu$ g Se/L whole blood) in heifers according to the agricultural areas in Wallonia (Belgium)\*.

Areas	n	Mean	Standard error	Area classification	Animal classification %			
					SD	MD	Α	НА
Ardennes	16	34ª	3.7	SD	69	31	0	0
Condroz	46	52 <sup>b</sup>	2.2	MD	59	21	20	0
Famenne	11	47 <sup>bc</sup>	4.4	SD	55	36	9	0
Limoneuse region	41	43°	2.4	SD	68	29	3	0

a,b,c=Means with a different letter are significantly different, n=number of animals, SD=Severely deficient, MD=Marginally deficient, A=Adequate, HA=Highly adequate, \*For calculation of Se concentration from determined Glutathione peroxidase activity (see equation in Table-1).

**Table-3:** Mean concentrations of blood selenium ( $\mu$ g Se/L whole blood) in dairy cows according to the agricultural areas in Wallonia (Belgium)\*.

Areas	n	Mean	Standard error	Area classification	Animal classification %			
					SD	MD	Α	НА
Ardennes	6	44ª	6.4	SD	83	17	0	0
Condroz	39	48ª	2.5	SD	61	18	21	0
Famenne	5	51 <sup>ab</sup>	6.8	MD	60	20	20	0
Limoneuse region	32	64 <sup>b</sup>	2.7	MD	35	30	35	0

a,b=Means with a different letter are significantly different, n=number of animals; SD=Severely deficient, MD=Marginally deficient, A=Adequate, HA: Highly adequate, \*For calculation of Se concentration from determined Glutathione peroxidase activity (see equation in Table-1).

**Table-4:** Mean concentrations of blood selenium ( $\mu$ g Se/L whole blood) in beef cows according to the agricultural areas in Wallonia (Belgium)\*.

Areas	n	Mean	Standard error	Area classification	Animal classification %			
					SD	MD	Α	НА
Ardennes	25	32ª	3.7	SD	88	12	0	0
Condroz	48	35ª	2.5	SD	79	19	2	0
Famenne	6	55 <sup>b</sup>	7.3	MD	67	33	0	0
Limoneuse region	23	40 <sup>ab</sup>	3.7	SD	78	18	4	0

a,b=Means with a different letter are significantly different, n=number of animals, SD=Severely deficient, MD=Marginally deficient, A=Adequate, HA=Highly adequate, \*For calculation of Se concentration from determined Glutathione peroxidase activity (see equation in Table-1).

limoneuse region than in Ardennes [40]. A lack distribution of vitamin-mineral complexes could also be the cause of the deficiencies.

## Selenium supplementation and health management

Selenium supplementation of heifers and dairy cows before calving could be a key strategy to reduce productivity losses and the risk of pathologies due to a Se deficiency [41]. Selenium supplementation reduced the prevalence of intra-mammary infections, the incidence of metritis, ovarian cysts and the incidence of retained placenta [2,7]. There are many methods that can be employed to prevent Se deficiencies. These are: the inclusion of mineral or organic Se in the diet, application of fertilizers containing Se, incorporation of Se in drinking water, Se injections, Se implants, Se microencapsulation and Se boluses [42,43].

Selenium level may be increased in feedstuffs by use of a mineral fertilizer containing Se. In Finland, Se-bio fortification program started in 1984 over the whole country. This has been very successful in increasing the Se status of humans and livestock. In plants, mineral Se in fertilizer is mostly transformed available for herds over the whole year. The Se content of a ration can be increased from 70 to 220  $\mu$ g/kg DM, using forages and cereals produced with Se-enriched fertilizers [45]. The use of alfalfa hay produced with fertilizer enriched Se also improved both the Se status of calves and their body weight [46]. Similarly, Hall et al. [47] using different strategies of Se supplementation, with a forage produced by Se enriched fertilizers, induced adequate Se concentrations in blood of cattle, where a supply was offered over a period of 6 weeks. Even after 6 months of pasture without Se supplement, Se status remained high compared to the inorganic Se supplemented group. Similar results were reported in sheep in USA [48]. The use of plants with a high storage capacity of Se, such as Lolium perenne, may also be implicated in improving the Se status in the cattle herds of Wallonia. L. perenne produced on soil with Se-enriched fertilizers concentrated more Se (0.15 vs. 0.03 mg/kg DM) than other grass species found in the grasslands of Wallonia [8].

into various types of organic Se [44]. It is, therefore, possible to increase the Se content in grass, grass

silage, maize silage and cereals and to make it more

### Conclusion

Owing to large Se deficiencies in the cattle herds studied - including all types of animals and all regions - strategies are needed to improve the Se status of animals. It is possible to provide Se in compound feedstuffs or in supplementary mineral mixtures. It is also possible to use crops with a high Se content, if they are produced in soil with a high Se content, or produced with a Se enriched fertilizer. By doing so, one can reduce Se deficiencies in the cattle herds of Wallonia. Similarly effects can also be expected over the whole of Belgium and over Europe.

## **Author's Contributions**

The experiment was performed under the guidance of ID. Data encoding was done by JFC. Data processing was done by JLH and MY. The article was prepared by MY and was supervised, reviewed and edited by JFC, JLH, LI and ID. All authors read and approved the final manuscript.

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### **Competing Interests**

The authors declare that they have no competing interests.

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