

Lead in game meat

Part 1 - Ammunition residues and chemical analysis.

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Preface

It is the task of the National Food Agency (NFA) to protect the interests of Swedish consumers by working to ensure safe foods of good quality, fair practices in the food trade and healthy dietary habits.

Several international studies have shown that lead residues from ammunitions can occur in very high concentrations in game meat. According to the European Food Safety Authority (EFSA), almost 30 percent of the samples of game meat analysed exceed the maximum permitted level of 0.1 mg/kg that applies to lead in meat from cattle, sheep, pigs and poultry that are offered for sale. In Sweden, it is estimated that around 10 percent of the population consume a large amount of game meat (approximately 300,000 hunters and their families). It is therefore important that the NFA investigates to what extent lead occurs in game meat, and whether there is any risk associated with the consumption of game meat.

In the autumn of 2011 a pilot study was conducted that led to NFA producing advice in respect of the consumption of meat deriving from game shot with bullets containing a lead core (Bly i viltkött – en riskhanteringsrapport [Lead in game meat - a risk management report], National Food Agency 2012). In 2012, NFA's Riksmaten [National Diet] investigation showed that consumers of game meat had higher contents of lead in their blood than other consumers (Riksmaten – vuxna [National Diet - adults] 2010-11) and a decision was made to continue studies concerning lead in game meat. The sub-reports that are hereby published answer the following questions:

- In which cuts of game meat do lead residues from ammunition occur, and how high are the concentrations?
- Can the lead residues be removed through adapted handling/cleaning?
- How much of the lead residues is available to the body through the consumption of shot game?
- How great a risk is entailed by the consumption of game meat, with respect to the effect that lead can have on the health of the consumer?
- What measure or measures are required to reduce the risk of the occurrence of lead in game meat?

These studies provide greater knowledge into the risks that can exist from the consumption of game shot with ammunition containing lead, and provide data and result for generating recommendations on how meat can be handled to minimise any possible risks.

The studies have been conducted on a collaborative basis by the National Food Agency, the Swedish Association for Hunting and Wildlife Management (SJF) and the National Veterinary Institute (SVA).

Report no. 18 *Lead in game meat* consists of four parts. *Part 1, Ammunition residues and chemical analysis*, investigates how the occurrence of lead residues from ammunition and lead contents vary between various cuts of game meat, depending on the choice of ammunition and the placement of the shot itself. This report also studies how lead residues dissolve in gastric environments. *Part 2,*

Lead contents in the blood of hunter families, investigates whether the content of lead in the blood affects consumers of game meat. Parts 1 and 2 provide data for the risk assessment of consumption of game meat shot with lead ammunition that is presented in *Part 3, Risk assessment*. The latter describes the risks entailed by residues of lead ammunition in game meat. Based on this assessment, a health-based critical level for lead fragments in game meat has then been established.

The information contained in these three scientific sub-reports and in other academic literature has then been evaluated in order to assess what measures could and should be used to reduce the risks associated with the occurrence of lead in game meat. Other relevant factors have also been considered within these assessments, such as whether it is possible for consumers to follow specific advice regarding the consumption of game meat that has been shot with lead ammunition; how advice such as this would be perceived; how it could be applied by the target groups; which supervisory authorities exist for this purpose; and whether the consequences of such a measure are proportionate in relation to the risks and benefits. *Part 4, Risk management*, describes the different considerations and assessments that led to the measures that NFA deems are necessary for the handling of occurrences of lead residues in game meat, and in order to minimise the risks that consumption of such meat can entail. The report aims to clearly describe the reasoning behind the measures that NFA has determined.

National Food Agency, 7 October 2014

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Definitions and abbreviations

Abbreviation	Definition
BfR	Bundesinstitut für Risikobewertung (German Federal Institute for Risk Assessment)
CRM	Certified reference material
EFSA	European Food Safety Authority
EU	European Union
ICP-MS	Inductively coupled plasma mass spectrometry
ICP-AES	Inductively coupled plasma atomic emission spectroscopy
NMKL	Nordic Committee on Food Analysis
Pb	Chemical symbol for lead
PT	Proficiency test
PTWI	Provisional tolerable weekly intake
SJF	Swedish Association for Hunting and Wildlife Management
SVA	National Veterinary Institute
Wound channel	Meat visibly affected by bullets, gunshot or fragments, including all bloodshot meat.
WHO	World Health Organisation

Summary

The results show that residues from lead ammunition are present in many of the meat samples analysed, both in minced moose meat and in other cuts of meat from roe deer, fallow deer fawns, wild boar and crows shot with lead ammunition. A total of nearly 200 samples have been analysed. The levels of lead vary immensely, from the detection limit (0.004 mg/kg) up to hundreds of mg/kg. The highest levels of lead are found in meat from the wound channels, which are not intended to be consumed. High levels have also been found in game meat that is intended for consumption.

In minced moose meat, 33 per cent of the samples were over the maximum permitted value (0.10 mg/kg) for lead in meat from cattle, pigs, sheep and poultry, and in the other cuts of meat, 43 per cent of the samples were over this limit. There was a significant connection between the lead content and the distance to the wound channel. The closer to the wound channel, the higher the lead content in the meat. The median value for the level of lead in various cuts of meat intended for consumption was 0.05 mg/kg (n=104) whilst the mean value was 9.9 (standard deviation= 38) mg/kg. In minced moose meat the median value was 0.03 mg/kg (n=54) and the mean value 0.9 (standard deviation =3) mg/kg. The results from game shot with bullets show that high lead levels occurred in cleaned meat from the shoulders of roe deer and fallow deer fawns (mean value 30 mg/kg, median 0.08 mg/kg). The median value for lead in meat from the loin, tenderloin, saddle and for the haunch was 0.004 mg/kg, which is the level that this analysis method is capable of detecting. The mean value in these samples was 0.25 mg/kg, which is 1,000 times lower than in the wound channel from game shot with lead core bullets (mean value 223 mg/kg, median value 89 mg/kg). The lead levels were also high for uncleaned game taken with lead shot (mean value 111 mg/kg). After cleaning, the lead level fell by approximately 100 times (mean value 0.78 mg/kg). The results from the solubility experiments showed that lead fragments from bullets dissolve in hydrochloric acid of the same concentration as in the stomach of humans. The solubility increases when the fragments in the acid are gently rocked to mimic the motion in the stomach. An alternative is to use lead-free ammunition.

Background

General

Swedish hunters annually shoot game that provide nearly 17,000 tonnes of meat (Wiklund & Malmfors, 2014). This corresponds to around 12 per cent of the total trade in beef (The Swedish Board of Agriculture's Yearbook of Agricultural Statistics 2010). Game meat is lean, is low in cholesterol and is a good source of trace elements (Jarzyńska & Falandysz 2011) and a valuable natural resource (Wiklund & Malmfors 2014). In recent years however, reports from overseas have shown that game meat can contain residues of lead from ammunition (EFSA 2010). Lead is poisonous and even low levels of exposure are thought to cause damage to the nervous system. This is especially the case when the brain is developing in the foetus or in small children. In epidemiological studies of children it has been estimated that blood lead levels of around 12 µg/litre can lead to a lower IQ (for further information regarding the toxicity of lead, see *Lead in game meat Part 3 – Risk assessment and Part 4 – Risk management*). A first investigation of Swedish conditions in 2011 showed that lead residues were present in around half of all minced moose meat collected from hunters' freezers (National Food Agency internal report from 2012, the results also presented here). This led to the National Food Agency issuing limiting advice regarding the consumption of meat from game shot with bullets containing a lead core (Bly i viltkött - en riskhanteringsrapport, Livsmedelsverket [Lead in game meat - a risk management report, National Food Agency] 2012). In the following year, the National Food Agency showed, through its Riksmaten – vuxna [National Diet - adults] 2010-11 investigation, that consumers of game meat had higher levels of lead in their blood than other consumers (Bjermo 2013).

At the same time as the Agency published advice concerning consumption, follow-up investigations were initiated to study:

- In which cuts of game meat do the lead residues from ammunition occur and how high are the contents?
- Can the lead residues be removed through adapted handling/cutting?
- How much of the lead residues can be absorbed by the body through the consumption of game meat?
- How great a risk is entailed by the consumption of game meat, with respect to the effect that lead can have on the health of the consumer?

The investigations aim to increase knowledge regarding the lead levels in various cuts of game meat, and to develop recommendations for adapted meat handling in order to minimise any possible risks. The studies have been conducted on a collaborative basis by the National Food Agency (NFA), the Swedish Association for Hunting and Wildlife Management (SJF) and the National Veterinary Institute (SVA). SJF has been responsible for the collection of samples, the statistical evaluation of results, and for knowledge in respect of hunting and ammunition. SVA has been responsible for x-ray analyses and has, together with SJF, cut and prepared the samples. In *Part 1, Ammunition residues and chemical analysis*, the NFA has been responsible for the homogenisation of samples, for chemical analysis and for the investigation of the solubility of lead

in gastric environments. All parties have contributed to the writing of texts within their respective areas and the NFA has acted as coordinator.

Lead in different types of ammunition

Ever since hand guns were first invented, lead has been used for bullets and shot since this metal has a number of positive ballistic qualities. At the same time, it is well established that lead from ammunition can have an impact on the environment (for example Axelsson 2009, Helander et al., 2012, Mateo et al., 2014). In recent years, the presence of residues of lead from ammunition has been noted in game meat along with raised levels of lead in the blood of consumers of game meat (Bjermo 2013, Meltzer 2013). Ammunition manufacturers have consequently produced alternatives to lead, both for bullets and gunshot. The alternative materials show some limitations compared with lead, and hunters have to balance the environmental advantages against the ballistic disadvantages. However, there are also compromises to be made when choosing between the various lead bullets available; there is no one bullet that is suitable in all situations.

Bullets

The vast majority of bullets used in hunting today have a lead core. This is enclosed within a jacket which is made of a brass alloy containing a high degree of copper. When a bullet expands, more impact energy is transferred to the target, which means the shot has a greater effect, leading to a quicker death. Consequently, larger game may only be hunted with bullets that are designed to expand in Sweden. However, roe deer and lynx may also be hunted using shot (NFS 2002: 18). In terms of weight, game that can only be shot with expanding bullets accounts for over 90 per cent of the game meat obtained from hunting in Sweden every year (Wiklund & Malmfors, 2014). For this reason, the majority of this report refers to this type of hunting.

When a bullet with a lead core expands, it also sheds fragments of the core and the jacket, and this contributes to the effect of the bullet. At the same time, the fragmentation must be limited, so that the bullet retains enough energy to penetrate and reach the vital organs. The most effective way of limiting expansion and fragmentation is currently to adapt the thickness of the jacket and simultaneously to chemically fuse the lead core together with the jacket so that they stay connected to each other during the expansion. Since the jacket and core are bonded together, these bullets are known as "bonded" bullets. Expanding bullets with lead cores are therefore currently divided up into conventional "non-bonded" bullets and modern "bonded" bullets. Bonded bullets hold together better and consequently do not lose as much lead when hitting the target.

The majority of lead fragments from an expanding bullet are small, soft and difficult to see when cleaning or slaughtering the animal (for example Hunt et al., 2009, Knott et al., 2010). It is important to clean thoroughly around the wound channel in order to minimise the risk of lead residues in the meat. One of the purposes of this study was to investigate how close to the wound channel cleaning should be conducted.

Lead-free bullets also exist; these are made of homogeneous copper or brass with a hollow tip. When the bullet hits the prey, the bullet expands as the tip is turned inside out. Copper bullets do not expand as easily as soft bullets with a lead core, which means that bullets made from copper

may have less effect on the prey. There are however comprehensive modern investigations that show that the shot effect of copper bullets is perfectly satisfactory (Gremse & Rieger 2014).

Figure 1 shows the bullets used in the investigation, which were shot into a plastic crate filled with soaked telephone directories in order to simulate the impact on prey.

In a survey of Fennoscandian moose hunters regarding their choice of bullet, 2.4 per cent of the Swedish hunters stated that they used copper bullets, compared with 4 per cent in Norway and 18 per cent in Finland (Stokke et al., 2010).

Shotgun shells and shot

Unlike rifle cartridges, shotgun shells contain many small projectiles called shot.

But like bullets, lead shot require great impact energy. Just as with bullets, and for the same reasons, lead is, ballistically, a very suitable material for the manufacture of gunshot. Lead shot have no jackets but may be plated, for example with nickel.

A charge of shot that hits its prey creates many small wound channels, unlike bullets, which create one large one. Lead shot that hits a bone or other hard material can form fragments when that material becomes deformed.

By far the most common alternative to lead is steel shot. This is lighter than lead shot of the equivalent size, and harder. Exactly as with bullets, the environmental advantages of alternative types of shot must be balanced against the ballistic disadvantages; see also Appendix 1.



Photograph: F Widemo, SJF

Figure 1. The various bullets that were used in the investigation; they were shot from a .308 calibre Winchester into a plastic crate filled with soaked telephone directories in order to simulate the impact on the prey. From the left, Nosler E-tip (homogeneous copper bullet; residual weight 99.2 per cent), Lapua Naturalis (homogeneous copper bullet; residual weight 99.6 per cent), Norma Oryx (bonded bullet with lead core; residual weight 94.1 per cent), Lapua Mega (non-bonded bullet with lead core; residual weight 89.0 per cent) and Norma Silverblix (older type non-bonded bullet with lead core; residual weight 60.7 per cent).

Analysis of lead in food

Several studies have shown that residues from ammunition, in the form of lead particles, are not evenly distributed throughout the meat; furthermore, the particles vary greatly in size (for example, Hunt et al., 2009, Knott et al., 2010). Therefore, in order to analyse the presence of residues from ammunition in a certain part of a cut, it is necessary to analyse the whole cut. This procedure differs from regular routine analyses of metals in meat where only a small sub-sample is taken from each part of a cut. A method where the homogenised (minced) meat is placed in its entirety in diluted nitric acid to dissolve any residues of lead without completely dissolving the meat was developed at the Norwegian University of Life Sciences (Lindboe 2012). Analysis is then conducted on a sub-sample taken from the acid-meat mixture (the extract). For this method to be able to provide a completely quantitative result, all metallic lead must be completely dissolved and distributed evenly throughout the sample mixture. If any of these criteria are not completely satisfied, then the method still provides results of a semi-quantitative nature that can be used to demonstrate the presence of lead residues in game meat.

Metallic lead, solubility and absorption

The lead found in lead ammunition is metallic lead. Metallic lead dissolves in the presence of hydrogen ions (Hägg 1989), i.e., in acids. When lead comes into contact with hydrochloric acid (which can be found in the stomach) the dissolved lead ions, together with the chloride ions from the hydrochloric acid, form a salt. This salt is known as lead chloride and it forms a protective layer on the surface of the lead metal and protects it, to a certain extent, from dissolving further. If the layer of lead chloride is damaged in some way or scraped off, then the underlying lead continues to dissolve. A layer of lead chloride is relatively easy to dissolve and not at all as hard as, for example, a layer of lead carbonate that can form in old lead water pipes. In the stomach and intestines, there is constant movement as nutrients are processed and transported. This is taken into consideration, for example, when one investigates how pharmaceuticals taken orally dissolve in the stomach and intestines. Various forms of stirring are then employed (European Pharmacopoeia 2012). The study of how metallic lead dissolves in the stomach and intestines should therefore include some kind of movement. In part of this report, a simple investigation is conducted regarding the amount of lead that can be dissolved from lead shavings in a gastric environment (weak hydrochloric acid) with and without stirring.

The percentage of lead dissolved also depends on how small or large the lead particles are, or, in other words, how large the total area exposed to the acid is, in relation to the volume. One gram of lead in a large, single piece has much less surface exposed to acid than 1 gram of small lead shavings. Therefore, lead should theoretically dissolve faster from 1 gram of small lead shavings than from one large 1 gram piece. In our tests, only smaller particles, of around 1 mm in diameter, have been studied. Another aspect of interest in discussions regarding the solubility of metallic lead is that, under normal conditions (in air and room temperature), the outermost layer of lead metal always forms lead oxide. The presence of lead oxide affects both the speed of dissolution and the weight ratio of lead in the sample being studied. In our tests, freshly prepared lead shavings have been used.

Method and material

Collection of data and samples

Minced moose meat

For the collection of minced moose meat from hunters, ten of the Swedish Association for Hunting and Wildlife Management's game wardens were asked to collect 4-5 packages of frozen minced moose meat each from local hunting teams that could certify that the slaughter had been conducted under private management and that the wardens had not themselves participated in the handling of the meat. Already frozen minced meat intended for consumption was collected, one random sample (≥ 200 grams each) taken from each hunting team. A total of 48 samples were collected from the counties of Norrbotten, Västerbotten, Västernorrland, Jämtland, Gävleborg, Dalarna, Uppsala, Västmanland, Kalmar and Kronoberg. The samples were marked with a serial number and were accompanied by information regarding the age of the animal (adult/calf) and the part of the carcass that had been used. However, meat from several animals often is mixed up prior to grinding, so comprehensive information about several of the samples was missing. Furthermore, six packets of minced moose meat, weighing 200 grams or more, were purchased by game wardens from food stores in five different towns. When making the purchase, they asked which county the minced meat had originally come from. The minced meat was frozen and treated in the same manner as the samples obtained from the hunters. These collections were made during November 2011.

Two muscle samples from moose that had died of other causes (game found dead that was examined by the SVA) were analysed in order to estimate the normal approximate level of lead in moose meat that had not been contaminated by lead from ammunition.

Meat from game shot with bullets

Roe deer and fallow deer fawns

To gather data regarding how bullet fragments dispersed around the wound channel and to analyse lead levels in the meat, a total of 10 roe deer, 11 fallow deer fawns, one wild boar and a moose calf were shot by rifle during 2012-2013. The hunters who shot the game were instructed, as far as possible, to try to shoot directly from the side and to hit the central part of the shoulder or just behind it, in line with the target area that is recommended in the training hunters receive prior to gaining their hunting licence (Christoffersson et al., 2010).

The actual result was that the shots either went through both shoulders, one shoulder, or, in some cases, failed to hit the shoulder at all. For two of the fallow deer fawns, the shots hit above the backbone, instead of through the chest. In all cases, the bullet's entrance and exit holes were in the front half of the body and the animal was felled with a single shot.

.308 Win calibre ammunition was used throughout for roe deer, fallow deer and wild boar. Eight roe deer and eight fallow deer fawns were shot with factory-loaded Norma Oryx 11.7 gram bonded bullets. One roe deer was shot with a Norma Silverblix bullet and three fallow deer fawns were

shot with Lapua Mega bullets. In these cases, the ammunition is a 11.7 gram bullet of conventional type, with lead cores that are not bonded to the bullet's jacket. One roe deer and a wild boar were shot with hand-loaded ammunition - a 9.7 gram homogeneous copper bullet of the Nosler E-tip brand in a .308 calibre Winchester. This bullet is manufactured completely in a copper alloy and is designed to expand without releasing any fragments. The moose calf was shot with hand-loaded ammunition of 9.3x74R calibre, with a 14.2 gram bullet of the Lapua Naturalis brand. This is a copper bullet of the same type as the Nosler E-tip (see also Figure 1).

Both the roe deer and the fallow deer fawns were gutted and skinned before they were x-rayed. After the front part of the body had been x-rayed, the shoulders were cut from the fallow deer fawns and x-rayed, prior to cleaning.

The carcasses were cleaned so that all the meat around the wound channel that was bloodshot or in any way visibly affected by the bullet, fragment or splinters from the bone were trimmed away. This included all the bloodshot meat. All such meat was collected in separate samples designated *Wound channel* for the fallow deer fawns. The roe deer were cleaned in the same manner, after which a further five centimetres of unaffected meat around the wound channel was cut away and added to the sample from the wound channel itself. Cutting away 5 cm of unaffected meat around the wound channel as a safety margin against fragments is in line with the SJF's previous recommendations for the handling of meat from game shot with lead bullets. These preliminary recommendations were produced whilst the project was in progress, pending the final recommendations.

After cleaning, samples were taken from the shoulders where there was still meat, and from the front part of the back, directly above the wound channel (chuck). For the two fallow deer fawns that had been shot partly through the back (and through the chuck), samples were taken instead from the front part of the loin, farther back. From four of the roe deer, samples were also taken from the loin, the tenderloins and the haunch. All these samples correspond to cuts of meat that are expected to be used for consumption. The samples were x-rayed again after cutting. The wild boar shot with a copper bullet was gutted, but not skinned before being x-rayed. The moose calf could not be x-rayed due to its size, but all the meat trimmed from the wound channel was saved and analysed with respect to its lead and copper content.

Prior to the analysis of the levels of lead and copper in the meat, the samples from the entrance and exit sides of the carcasses were combined, so as to reduce the total number of analyses. This is also often the case with the hunters' handling of the cuts. The roe deer and fallow deer fawns were subsequently classified based on whether the bullet hit the upper foreleg bone (humerus; strong resistance), the shoulder blade (scapula; medium resistance) or penetrated the chest cavity without hitting skeletal parts of the shoulders (light resistance). The two fallow deer fawns that were shot above the backbone were not included in the analyses of how impact on the skeletal parts of the shoulders affected fragments or lead levels.

Wild boar shoulders

The collection of samples from game shot with known ammunition was supplemented through the collection of 18 shot-through wild boar shoulders from Öster Malma's game handling facility. These were x-rayed, after which they were cut. All the meat around the wound channel that was

bloodshot or in some way visibly affected by the bullet, or from fragments or splinters from the bone was designated *Wound channel*. Around the wound channel samples were then cut out in concentric circles of 0-5 cm, 5-10 cm and 10-15 cm around the cleaned wound channel. The samples were then x-rayed again. In three cases the shoulder was too small for it to be possible to take a sample from the 10-15 cm category. It is not known what calibre of weapon or type of ammunition was used in respect of the wild boar shoulders. In one case a whole, expanded bullet was still in the wild boar's shoulder when it came to be cut. This was removed prior to grinding, in the same way as it would be with the normal handling of meat.

The wild boar were subsequently classified based on whether the bullet hit the upper foreleg bone (humerus; strong resistance), the shoulder blade (scapula; medium resistance) or if it just penetrated the shoulder's soft tissues (light resistance). All wild boar shoulders contained fragments from bullets that could be identified in x-ray images. The number and size of the fragments varied immensely. The number of fragments was classified for the whole shoulder as large (> 200), medium (c. 50-200) or small (< 50). At the same time, the size of the fragments was classified as large (> 4mm), medium (2-4 mm) or small (< 2mm). Following cutting and x-raying of the samples these were also classified on a six-point scale: no fragments, 1-5, 6-10, 11-15, 16-20 and more than 20 fragments.

The carcasses and shoulders were cleaned and cut whilst they lay on the autopsy table at the SVA. The protective plastic on the autopsy table was changed between each animal in order to prevent the lead fragments being contaminated between animals. During the cutting however, all the work for each animal was conducted on the same table surface. The knives were dried clean with paper between each new sample.

Meat from game shot with lead shot

In order to study fragments from lead shot and lead levels in the meat, a total of 20 crows were shot with shotguns. To start with, ten crows were shot with a 12 bore shotgun using Saga Elite Sporting size US 7 shot. Following the initial x-ray analyses of these, the study was extended to include a further ten crows, which were shot with a 12 bore shotgun using Gyttrorp Grouse size US 6 shot. Saga is marketed as a sport shooting shell, whilst Gyttrorp Grouse is marketed as a shell for use in hunting. However, many hunters use sport shooting shells when hunting smaller game.

The crows were debreasted, i.e. the whole breast bone and the breast muscles were prepared for analysis. These were then x-rayed, after which the breast muscles were cut free. At the same time, all bruising and visible wound channels from individual pieces of shot were trimmed off. If whole pieces of shot were encountered in the preparation, then these were removed prior to grinding, exactly as with normal meat handling. For the ten crows that were shot with Saga shot, the trimmed flesh from all the individual birds was combined to form one joint wound channel sample. For the crows shot with Gyttrorp Grouse shot, the trimmed flesh was kept separate for each individual and was weighed separately, after which the lead levels were analysed. In this way the untrimmed lead level could be calculated for each individual shot with Gyttrorp shot, based on the levels in the two analysed samples (cleaned cut + trimmed flesh). It was only possible to calculate a mean untrimmed value for the crows shot with Saga shot (10 cleaned individual samples + combined offal), however. Following cleaning, all breast muscles were x-rayed again.

In addition, one roe deer and one hare were shot with lead shot. The roe deer was shot with a 12 bore shot gun using Rottweil US3 size shot and the hare with a 12 bore shot gun using Gyttop Special US3 size shot. The carcasses were x-rayed in their entirety. Subsequently, the carcasses were coarsely chopped up into bone-free cuts. The sirloin of the roe deer was divided into two parts: the loin and chuck, and the entire bone-free thigh, for the right and left side of all parts. The hare was cut along the right and left thighs, as well as the whole of the loin (right and left side together). Bloodshot parts of the cuts were trimmed away without any particular cutting away of extra blood-free margins. Any visible shot was removed manually from the cuts in line with what was deemed to be normal hunting practice. Bone-free cuts were x-rayed separately. A randomly selected loin, chuck and thigh from the roe deer, and all parts of the hare were analysed in respect of lead levels.

Statistical evaluation of the results

Statistical analyses were conducted using Statistica 12. All distributions of fragments and lead levels deviated from a normal distribution and therefore nonparametric tests were used throughout. Roe deer and fallow deer fawns are similar in terms of their morphology, size and weight and there were no significant differences between the two in respect of lead levels for any particular cut, see Results below. To increase the sample size and the statistical power, the results for the fallow deer fawns and the roe deer were analysed together. There were no significant differences in the lead content of animals shot with bonded bullets or of those shot with unbonded bullets with a lead core. (see Results below). To increase the sample size and statistical power, the results for animals shot with different types of lead ammunition were therefore analysed together.

X-ray

All samples were x-rayed using the Rotopractix 90/20 together with a Regius Model 110 S direct digitizer. The dissolution of lead fragments in the digital x-ray images that were saved in JPEG format is estimated at a smallest interpretable size of approximately 0.1 mm. Bone chips from splintered bones can, like metal fragments, be seen as a radiopaque object, but the metal density is so much greater that the material can be distinguished when examining the samples.

Pooling of samples

In order to reduce the total number of samples prior to chemical analysis, the cuts from the right and left sides of the same roe deer or fallow deer were ground together (pooled). For example, left and right shoulders were pooled and left and right pieces of loin were pooled. The pooling provides a measurement of the lead levels in the cuts that are intended for consumption (shoulder, chuck, loin) and in those to be discarded (meat trimmed from the wound channel).

Determining the lead content

All chemicals used have been of analytical quality (p.a.) or better. All water used for dilutions and preparations of diluted acids comes from a Q-POD Element water purification unit (Merck Millipore, Darmstadt, Germany). All laboratory material is washed in acid prior to use.

All sample preparation and analysis has been conducted on de-identified meat samples.

Grinding

The samples are ground in mills with stainless steel knives until the mixture is minced and of an even consistency. For larger samples (> 200 g), a Multipurpose Food processor (HUG Electromechanic Engineering Ltd., Switzerland) is used, and for smaller samples (< 200 g), a Knife Mill Grindomax GM200 (Retsch, Düsseldorf, Germany). The mills are washed thoroughly between each sample. Some of the individual meat samples were ground together to form one sample (pooled). Samples of minced moose meat were considered sufficiently homogenised upon their arrival, and were not ground further.

Extraction/dissolution of lead fragments in meat

The dissolution of metal fragments was conducted mainly in accordance with Lindboe et al. (2012). The minced meat was weighed in glass or plastic beakers. The acid used for dissolution of fragments was 15 percent w/v nitric acid (HNO₃) and the amount added corresponded to twice the weight of the minced meat. The acid was added to the minced meat whilst it was stirred, either directly as 15 percent acid, or first with water and then with concentrated nitric acid which together gave a final nitric acid content of 15 percent. The latter procedure, where water was added before the acid, made stirring easier and prevented the formation of lumps which could occur when the acid was added. The minced meat was broken up into pieces and stirred with a porcelain spoon. It was covered with a lid and left overnight (17-20 hours). The mixture was stirred again and a sub-sample of the liquid was extracted with a pipette (10-20 ml) for analysis. The liquid extracted was turbid due to traces of muscle tissue and fat. Figure 2 shows the various interim stages in the extraction procedure.

To control the method of extraction, four samples of minced moose meat were extracted over an extended period of time. Sub-samples of the liquid were taken after two and three days and were analysed in respect of lead. X-ray images were also taken after the extraction of the leached minced moose meat, and also of the leached minced meat residues from wound channels, in order to see if any metal particles remained.

Analysis of lead

The lead level was principally determined by an Agilent 7700x ICP-MS (inductively coupled plasma mass spectrometry), following digestion of the liquid extract in a microwave oven (MARS Xpress) according to method NMKL no. 186. This method of analysis is accredited for samples of foodstuffs by SWEDAC in accordance with ISO/IEC 17025. The detection limit for lead is 0.002-0.004 mg/kg depending on the type of sample preparation. The performance of each individual analysis was verified through analyses of reference material with a known lead content.

For the minced moose meat samples, the level of lead was determined directly in the extract by ICP-AES (inductively coupled plasma atomic emission spectroscopy) of the brand Spectro Ciros^{CCD} at Uppsala University, Department of Chemistry, Division of Analytical Chemistry (Kollander 2010). The analysis method using ICP-AES is not accredited, but the results have been verified through repeated analysis of five samples using ICP-MS (accredited method, described above). The detection limit for lead using ICP-AES was estimated at 0.02 mg/kg per liquid sample.



Figure 2. Sample preparation for extraction/dissolution of metal particles.

Upper picture from the left: weighing in in 3 litre cups and the addition of 15 percent nitric acid. Lower picture from the left: minced moose meat before and after the addition of nitric acid.

Solubility of metallic lead in gastric environments

In order to estimate the amount of metallic lead that can be dissolved in a gastric environment, an experiment was conducted whereby metallic lead shavings were placed in diluted hydrochloric acid of the same concentration as is normally found in the stomach (0.1 M). The level of dissolved lead was then measured at different time intervals using ICP-MS. The metallic shavings were produced by using a stainless steel spatula to scrape out parts of the inside of a lead bullet that had previously passed through a moose. Around 8 mg of metal shavings (Figure 3) was then weighed into 4 different 50 ml test tubes, and 40 ml of 0.1 M hydrochloric acid (HCl) was added. Two of the tubes were placed in a shaker machine (Incubating mini-shaker, VWR, Radnor, Pennsylvania, USA) which was then set to produce a rocking motion to imitate movement in the stomach (Figure 4). The rocking motion continued for 2 days, after which the test tubes were left to stand still in a test tube rack. The two other test tubes were placed in test tube racks and remained still throughout the whole experiment. A blank test, with only 40 ml of hydrochloric acid in the test tube was prepared for shaking and another was left still throughout the experiment. The same experiments was repeated twice with different rocking settings on the shaker - "Rocking" with the setting "120" and "Increased rocking" with the setting "160". The intervals at which samples of the dissolved lead were taken were also different in both experiments.

In the experiment with "Increased rocking" sub-samples of the hydrochloric acid were taken from the various samples with plastic syringes after 1, 17 and 20 hours, whilst for the 'Rocking experiment', sub-samples were taken every 30 minutes for the first two hours, then one sub-sample per hour after three hours and four hours and then after 24, 51 and 120 hours. The sub-samples were extracted via the syringe and then filtered (0,45 µm) down into separate test tubes. For each sub-sample, a new syringe was used and a new filter, to avoid contamination. The sub-samples were diluted to 10,000 times with high purity water prior to analysis.

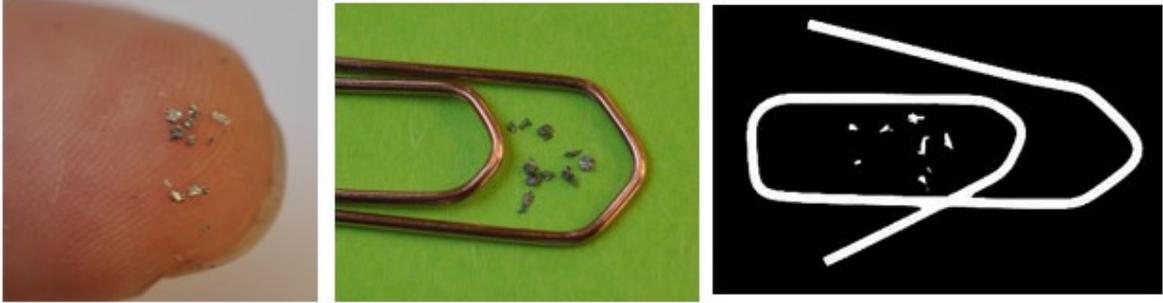


Figure 3. The pictures show 8 mg of lead shavings from a used lead bullet, photographed on a finger tip and alongside a paper clip. The black and white picture shows an x-ray image of a paper clip and 8 mg of lead shavings. The maximum permitted level for lead in meat from cattle, sheep, pigs and poultry is 0.1 mg/kg, which means that 8 mg contaminates 80 kg of meat up to the maximum permitted level. Photograph: SVA, 2014.



Figure 4. 8 mg of lead shavings were placed in 50 ml test tubes along with 40 ml of 0.1 M hydrochloric acid. Three samples, one of which was blank, were placed in a shaker machine and three in a test tube rack.

Results

Quality assurance of analysis results

The results are based on analysis of the extracted liquid and on the particulate lead dissolved during the prescribed 20 hours. When the extraction period was extended from one day to two and three days for five of the minced moose meat samples, there was no significant increase in the lead content in the liquid extracted. The x-ray of the extracted minced moose meat residues showed no visible traces of metal fragments remaining after one day of extraction. An estimation of the distribution of dissolved lead between the extract and the mince residue for the minced moose meat samples showed that around a fifth of the dissolved lead remained in the meat.

For many of the cut samples, especially those taken from the wound channel, the lead particles were bigger and greater in number than they were in the minced moose meat. In several of these samples, metal fragments could be seen by x-ray even after the extraction. The lead levels in these samples were amongst the highest measured in the study, between 250 and 1,829 mg/kg, despite the fact that not all of the lead residue dissolved. This means that it is not possible to estimate a general distribution of lead in the mince meat residue and the extract that applies for the whole concentration range. No attempt was therefore made to estimate the lead level in the minced meat residue remaining from the cuts. The analysis results for cuts are therefore based on the assumption that all the lead dissolved in the acid. This means that the measured levels of metallic lead in samples with large amounts of lead residues most probably are underestimations of the actual content.

The blank tests showed no quantifiable levels of lead and the analysed reference material agreed well with the known lead levels.

Lead levels in game meat

General

The results show that residues from lead ammunition are present in many of the meat samples analysed, both in minced moose meat and in other cuts of meat from other animals shot with lead ammunition. The results are presented in Tables 1-3 and in Appendices 2-5. The levels vary, from under the detection limit up to hundreds of mg/kg. The highest levels of lead were found in the wound channels, which are not intended for consumption. However, high levels were found also in game meat that is intended for consumption. In minced moose meat, 33 per cent of the samples were over the maximum permitted value (0.10 mg/kg) for lead in meat from cattle, pigs, sheep and poultry (Commission Regulation (EC) 1881/2006), and in the other cuts of meat, 43 per cent of the samples were over this limit.

There was a significant relationship between the lead content and the distance to the wound channel. The closer to the wound channel, the higher the lead content in the meat.

Minced moose meat

When the minced moose meat was x-rayed, visible fragments were present in 35 per cent of the samples. The lead levels in minced moose meat were within the range of < 0.020 - 31 mg/kg fresh weight. See Figure 5 and Appendix 2. The median value was 0.027 mg/kg. For the six samples that were purchased in stores, the results were within the range of < 0.02 - 2.5 mg/kg, with a median value of 0.05 mg/kg. Fifty-four percent of the samples contained quantifiable levels of lead and the level exceeded 0.1 mg/kg in 33 per cent of the samples (18/54). In 46 percent of the samples, the level of lead was under 0.02 mg/kg, which is the detection limit (LOD) for the ICP-AES method used. The measured level of lead was higher in the packets of minced meat that contained fragments of metal density visible under x-ray.

The two samples of muscle tissue from moose that died of non-hunting-related causes had a lead level of ≤ 0.002 mg/kg (measured with accredited routine method with ICP-MS).

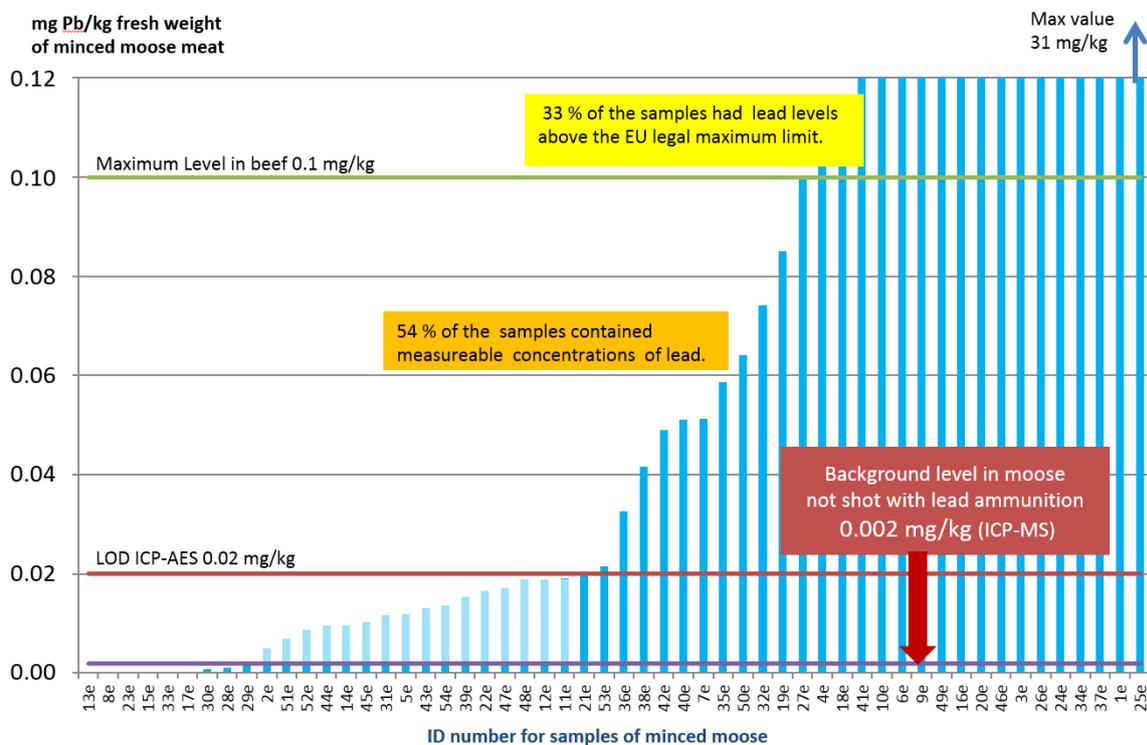


Figure 5. Lead levels in minced moose meat sorted in ascending order.

Different cuts of game

Analyses of lead were conducted in different cuts taken from 11 roe deer, 10 fallow deer fawns, 18 wild boar, one moose and 20 crows - a total of 61 individual animals and almost 200 samples. The lead levels in cuts of meat intended for consumption varied from a level under the detection limit to over 100 mg/kg. The median value for lead levels in cuts intended for consumption was 0.05 mg/kg (n=104). The mean value was 9.9 (standard deviation= 38) mg/kg. Forty-three percent of the cuts analysed had a level of lead that was over the maximum permitted value for lead (0.10 mg/kg) that applies for meat from cattle, pigs, sheep and poultry that are to be offered for sale.

Lead levels in game shot with bullets

Wild boar shoulders

Lead levels were significantly higher in wound channels from wild boar shoulders with many fragments (Kruskal-Wallis; $H= 10.45$; $n= 18$; $p= 0.005$) (Figure 6), and lead levels were higher in wound channels with larger fragments (Kruskal-Wallis; $H= 11.49$; $n= 18$; $p= 0.003$).

Lead levels decreased with increasing distance from the wound channel in the wild boar shoulders (Friedman; $\chi^2= 32.2$; $n=15$; $p< 0.0001$), (Figure 7, and Table 1), but there was only a tendency for a difference in the lead level between the samples taken from 5-10 and 10-15 cm from the wound channel (Friedman; $\chi^2= 3.27$; $n=15$; $p< 0.07$).

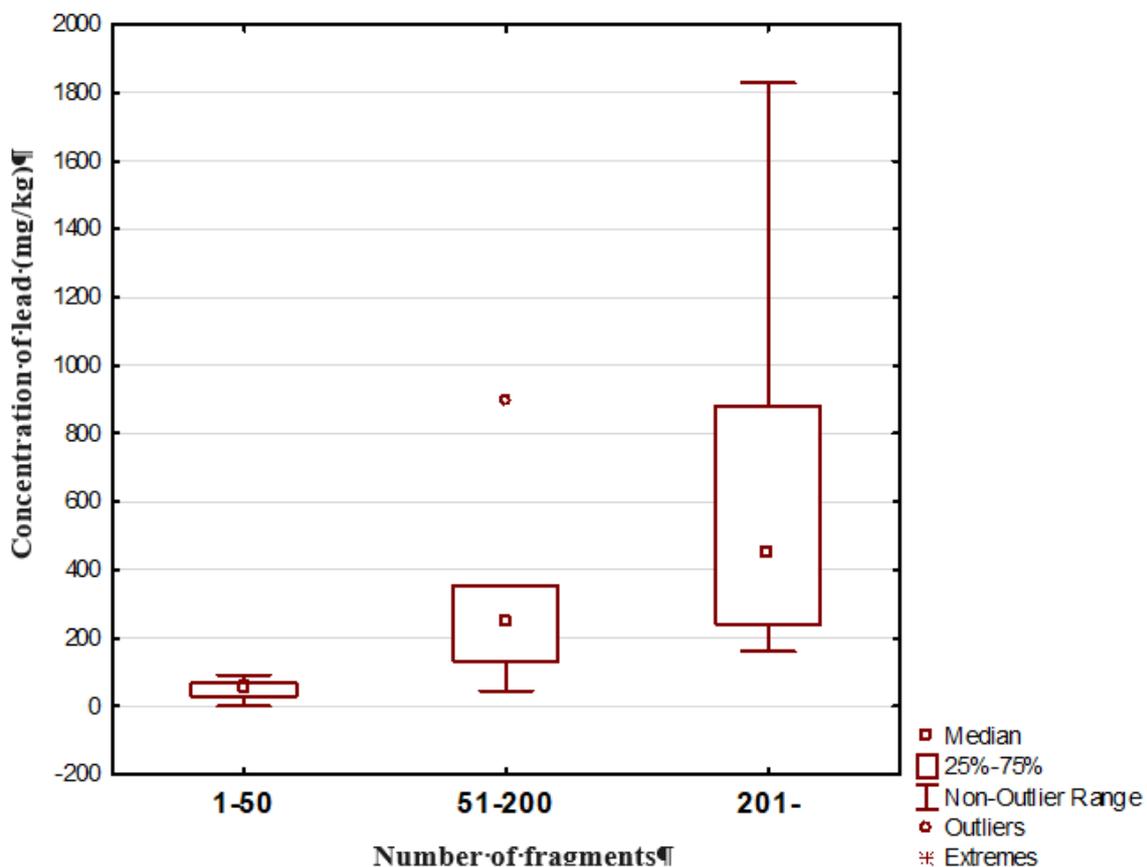


Figure 6. Wild boar wound channels with many fragments visible under x-ray had higher lead concentrations.

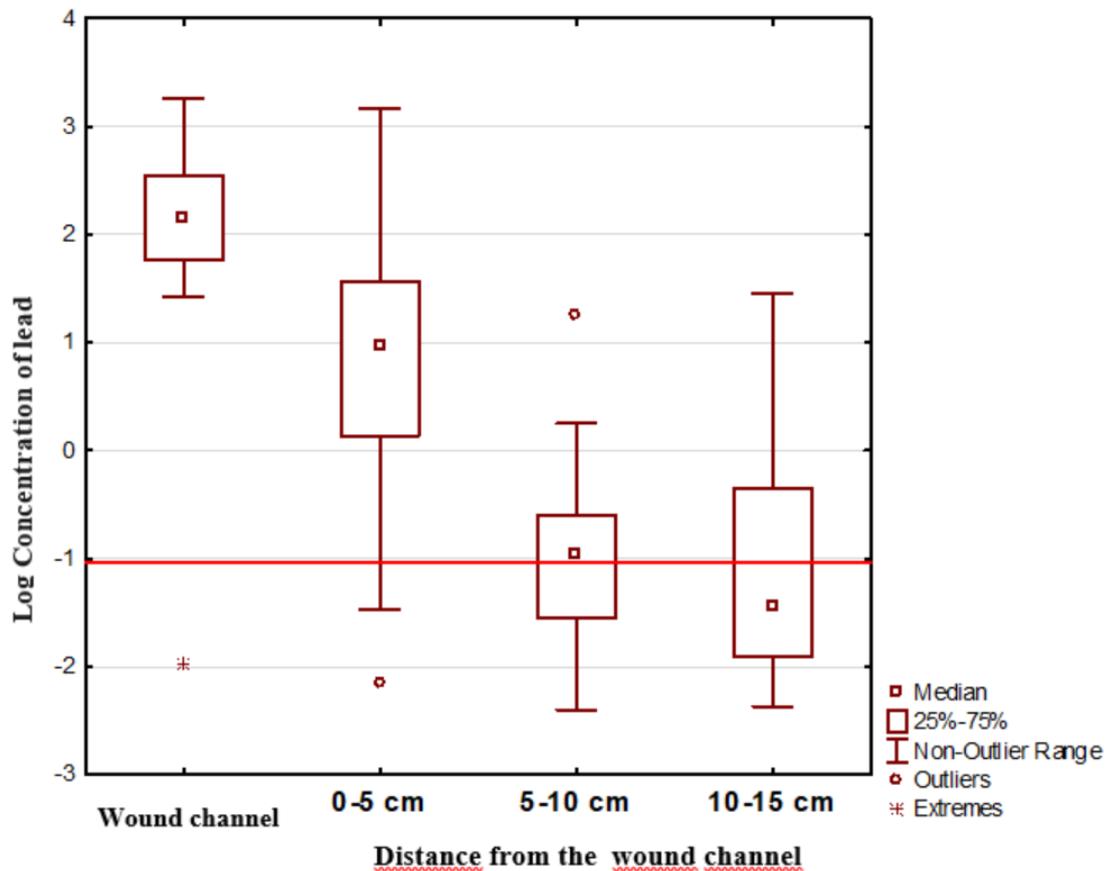


Figure 7. The lead levels decreased significantly with increasing distance from the wound channel in the wild boar shoulders. The natural logarithm of the lead levels are displayed, in order to illustrate the variations at lower concentrations. For comparison, the red line shows the maximum permitted value, 0.1 mg/kg, for lead in meat from cattle, pigs, sheep and poultry that are to be offered for sale. There is no maximum permitted level for game meat.

Table 1. Lead levels in various cuts of meat from each wild boar (V), mg/kg fresh weight.

Animal ID	Wound channel	Distance from the wound channel		
		0-5 cm	5-10 cm	10-15 cm
V1	352	24.6	1.07	28.5
V2	356	1.50	0.028	0.067
V3	69.1	0.034	0.050	0.038
V4	71.8	9.41	0.080	0.017
V5	131	103	0.153	0.013
V6	880	202	18.0	0.450
V7	1829	9.71	0.207	0.049
V8	92.0	0.821	0.022	0.011
V9	26.7	1.38	0.126	0.013
V10	160	1.59	0.063	0.014
V11	59.0	37.0	0.206	< 0.004
V12	0.011	0.007	< 0.004	
V13	242	18.6	0.251	0.036
V14	45.3	5.09	0.006	0.009
V15	44.8	0.166	1.78	1.58
V16	552	1466	0.523	7.37
V17 in*	250	52.4	0.024	
V17out*	895	9.18	0.089	
Mean	336	107	1.26	2.55
Median	146	9.30	0.11	0.04
Standard deviation	462	343	4	7
Max	1829	1466	18.0	28.5
No. of Individuals	17	17	17	15
No. of samples	18	18	18	15

* Both shoulders of this wild boar were analysed, both the entrance and exit of the wound.

Meat from the wound channel regularly contained hundreds of fragments, the samples from the region 0-5 cm from the wound channel often contained 5-10 fragments, whilst samples from 5-10 and 10-15 cm often lacked fragments distinguishable under x-ray. The analyses of the x-ray images showed that single fragments sometimes occurred in cuts far from the wound channel, even if there were no fragments in samples taken from regions closer to the wound channel (Figures 8 and 9).

Figure 8 A



Figure 8 B

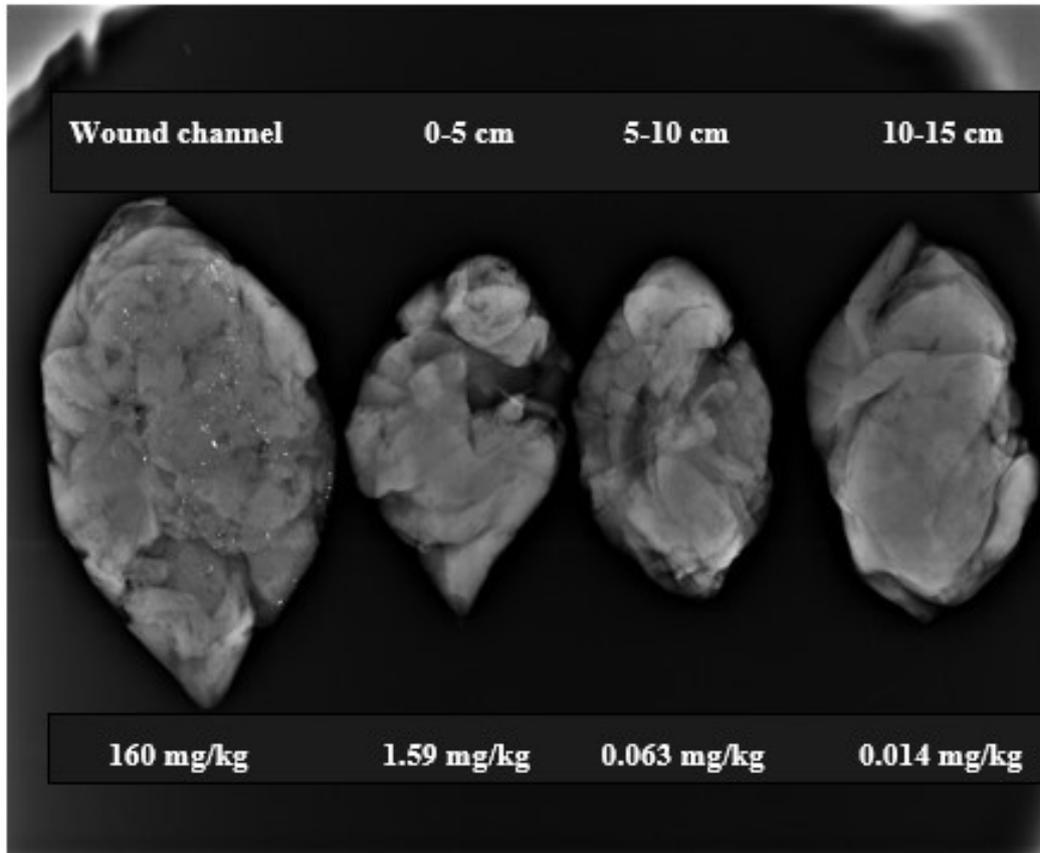


Figure 8. (A) Wild boar shoulder (individual V10) where the bullet hit the rear edge of the shoulder blade and fragmented. Based on the uniform size of the fragments, a bonded bullet has probably been used. **(B)** Cleaned cuts from the same shoulder prior to grinding. The sample from the wound channel contains many small fragments, the sample taken from 0-5 cm contains a few fragments whilst the remainder of the samples lack visible fragments. Photograph: SVA and SJF, 2014.

Figure 9A



Figure 9B

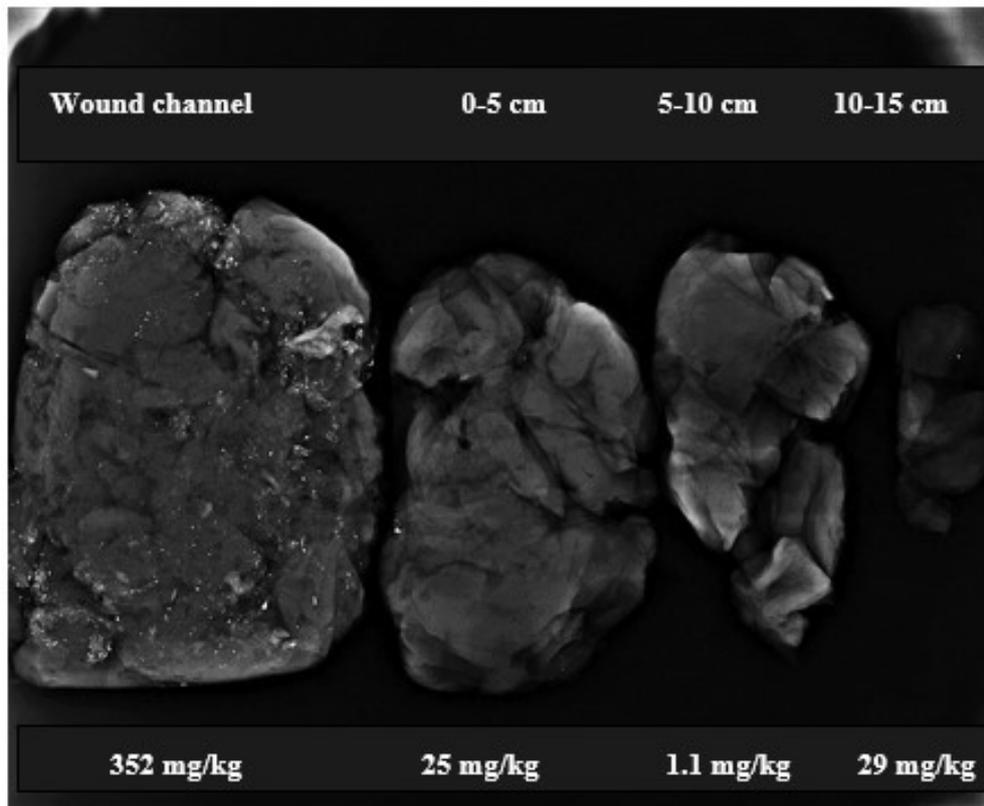


Figure 9. (a) Wild boar shoulder (individual V1) where a conventional, non-bonded bullet (bullet found in shoulder) hit the humerus and fragmented extensively. **(b)** Clean cuts from the same shoulder prior to grinding. The sample from the wound channel contained many small fragments, the sample taken from 0-5 cm contained isolated fragments, the sample from 5-10 cm lacked visible fragments whilst there was one visible fragment in the sample taken from 10-15 cm. Photograph: SVA and SJF, 2014.

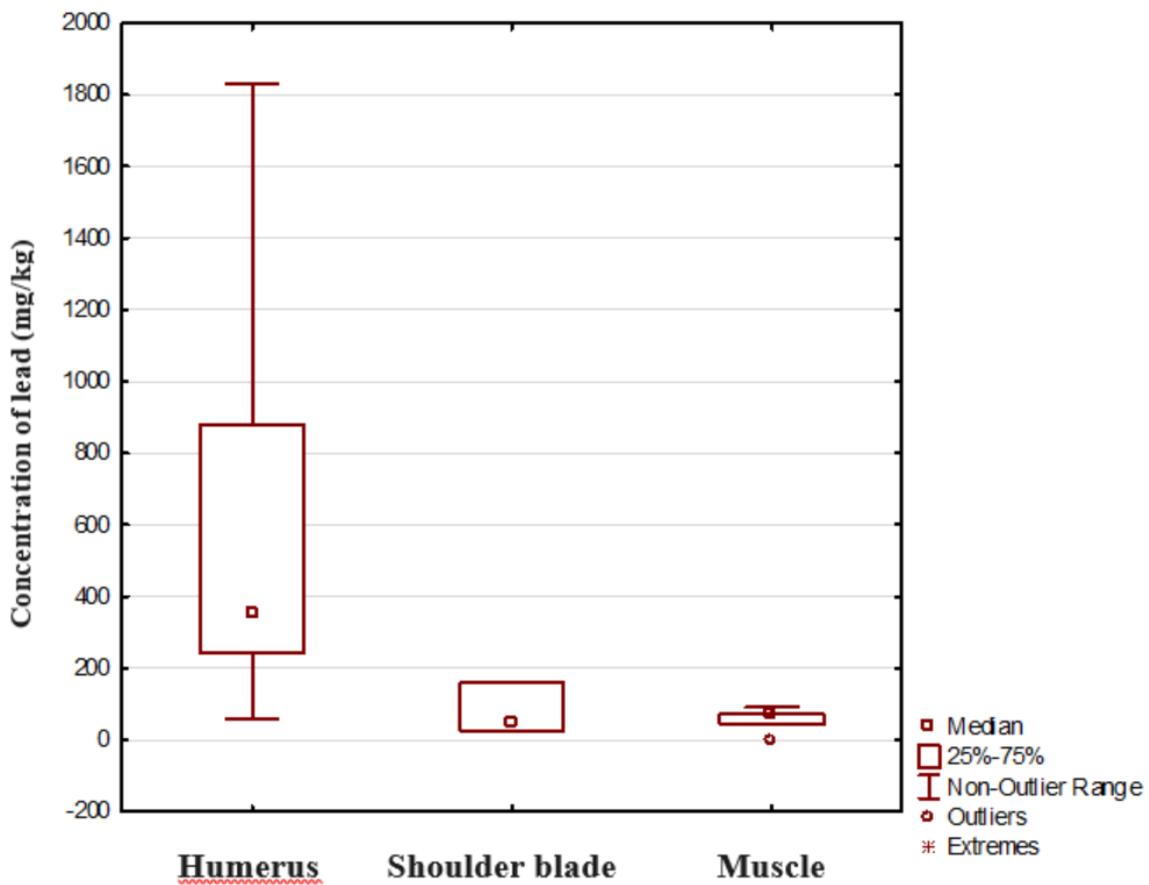


Figure 10. Lead levels in meat from wound channels where the bullet hit the humerus, shoulder blade or muscle (soft parts) of the wild boar. Significantly higher lead levels were found in the wound channel if the bullet hit the humerus, rather than if it only hit the shoulder blade or soft tissues.

Roe deer and fallow deer fawns

The results from roe deer and fallow deer fawns shot with bullets show that high levels of lead can occur in cleaned meat from the shoulder (mean value 30 mg/kg, median 0.08 mg/kg, see Table 2). However, the combined median value for lead in meat from the chuck, loin, tenderloin and the haunch was at or below 0.004 mg/kg, which is the level that this analysis method is capable of detecting. The mean value in these samples was 0.25 mg/kg, which is 1,000 times less than that measured in wound channels for game shot with bullets (roe deer, fallow deer and wild boar, mean value 223 mg/kg, median value 89 mg/kg) in this study.

X-raying of fallow deer fawns and roe deer showed that fragments were present around the wound channel, but also occurred spread throughout the chest cavity (Figure 11), as well as in clotted blood outside the chest cavity (Figure 12).

The lead levels of the various cuts (wound channel, shoulder meat and loin sections) for the fallow deer fawns and roe deer showed significant differences (Friedman; $\chi^2= 18.86$; $n=14$; $p< 0.00008$). For a subset of the roe deer, samples were also taken from the loin, tenderloins and steaks from the haunch. The more comprehensive comparisons also showed significant differences between cuts (Friedman; $\chi^2= 12.96$; $n=3$; $p= 0.02$). Regarding all samples as independent observations, there were significant differences between the various cuts (Kruskal-Wallis; $H= 36.56$; $n= 57$; $p< 0.0001$), and the lead levels fell with increasing distance from the wound channel (Figure 13, Table 2).

For fallow deer fawns and roe deer, there were no significant differences in lead levels in the meat from the wound channel whether the bullet hit the humerus, the shoulder blade or entered behind the shoulder (Kruskal-Wallis, $H= 1.99$; $n= 17$; $p= 0.37$).

Table 2. Lead levels in various cuts taken from the same individual, fallow deer fawns (A, F) and roe deer (R) shot with lead bullets, mg/kg fresh weight.

	Trim- mings*	Shoulder	Chuck	Tenderloin	Loin	Haunch
A1	252	0.077	2.63			
A2	33.6	1.76	0.004			
A3	148	0.126	0.021			
A4	13.3	0.059	0.019			
A5	439	1.51	0.590			
A6	147	5.28	0.149			
A7	233					
A8	85.7					
F1	60	0.019	0.047			
F2	10.4	0.018	< 0.004			
F3	233	235	< 0.004			
R1	266		< 0.004			
R10	171	< 0.004	< 0.004			
R2	78.1	1.79	3.41	< 0.004	< 0.004	< 0.004
R4	95.3	0.037	0.014	< 0.004	< 0.004	< 0.004
R5		< 0.004	< 0.004	0.100		
R6	83.2	0.039	< 0.004	0.014	0.009	< 0.004
R8	10.2	207	< 0.004			
R9	166					
Mean value	140	30.2	0.43	0.03	0.006	< 0.004
Median	121	0.08	0.01	0.01	0.004	< 0.004
Standard deviation	113	78	1.0	0.05	0.003	
Max	439	235	3	0.10	0.009	< 0.004
No. of samples	18	15	16	4	3	3

* Trimmings here include both the wound channel and an additional 0-5 cm in accordance with SJF's 2012 recommendations.

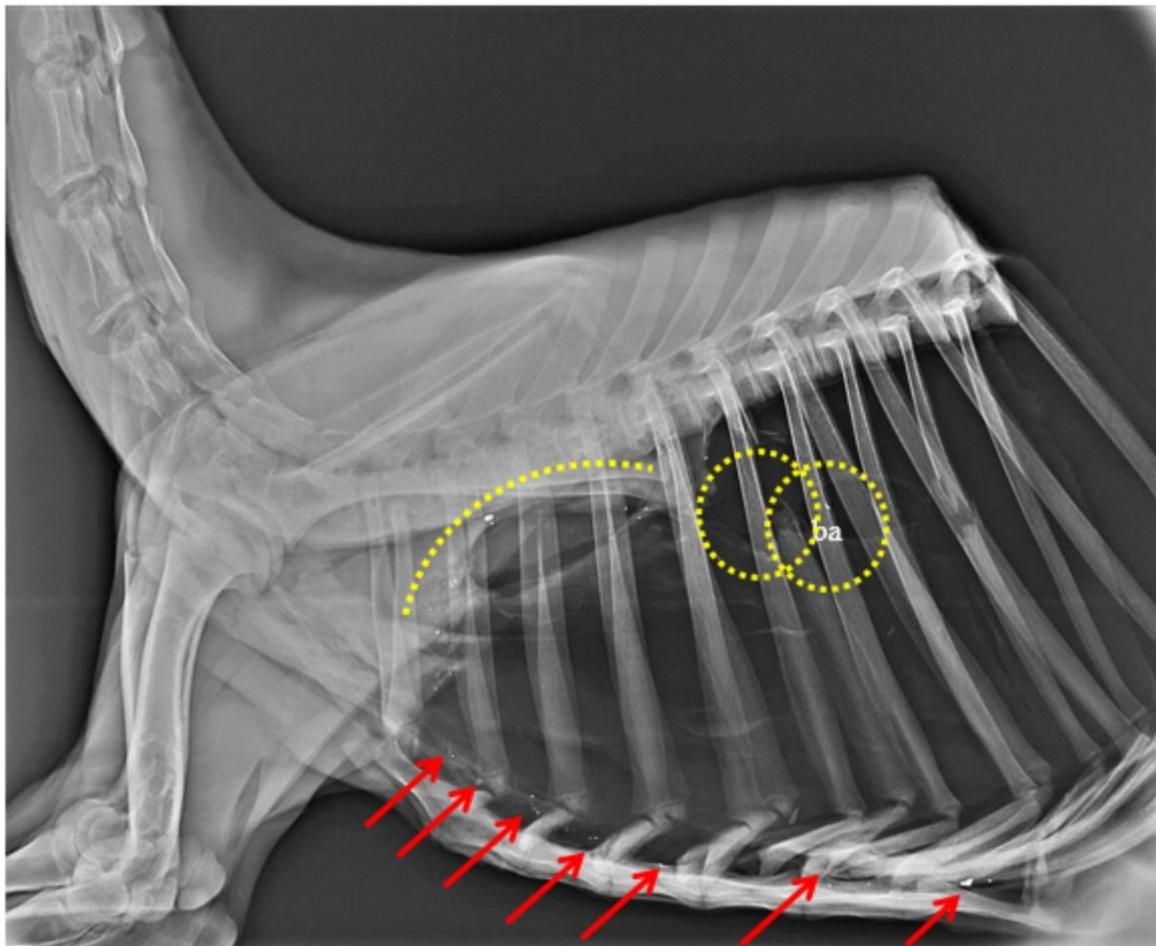


Figure 11. Gutted and skinned fallow deer calf shot with a bonded bullet just behind the shoulder. Fragments are seen both around the wound channel on the entrance and exit side (marked with yellow circles) and spread throughout the chest cavity where they have been carried by blood from the shot-through lungs (fragments marked with red arrows). The front legs have been stretched forward in the x-raying process, which the result that the wound channel at the rear edge of the shoulders (demarcated with a dotted curve) was displaced, compared with the holes through the chest cavity. Photograph: SVA and SJF, 2014.

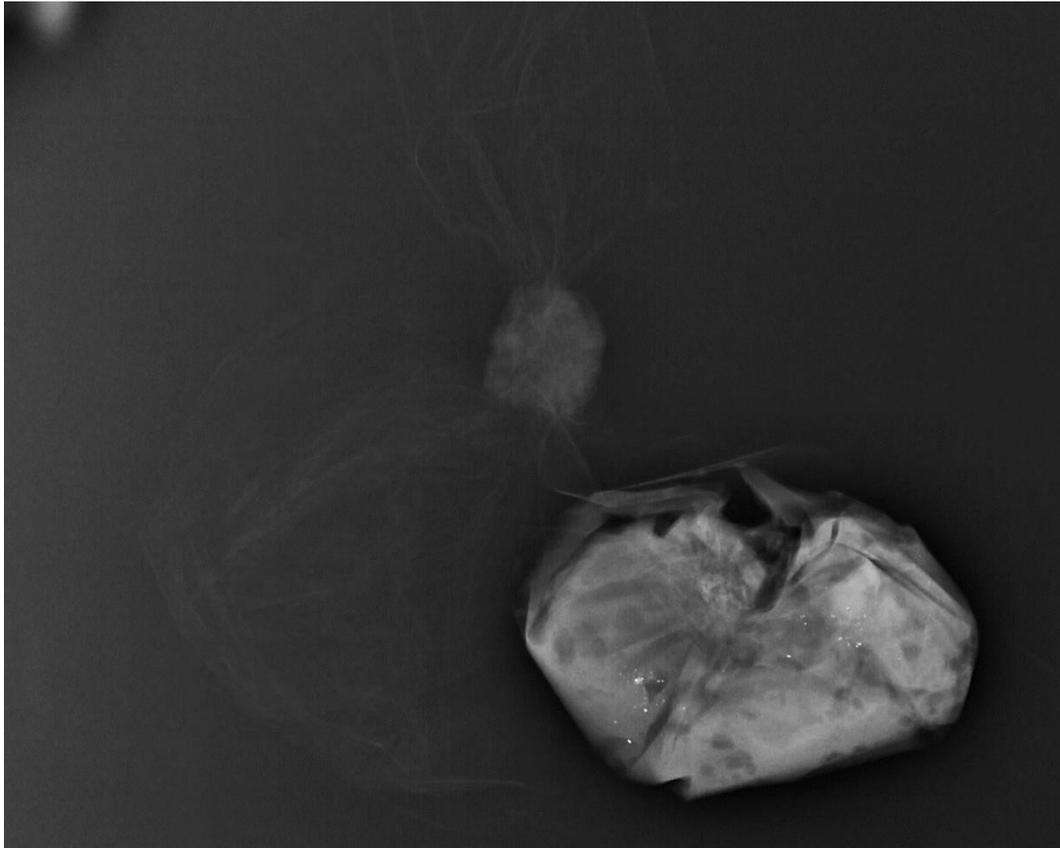


Figure 12. X-ray image of cut away blood-filled membranes that sat around the wound channel between the shoulder and thorax of a roe deer shot with a bonded bullet. The clotted blood contains a number of small lead fragments, some of which have been carried there by blood from the chest cavity. Photograph: SVA and SJF, 2014.

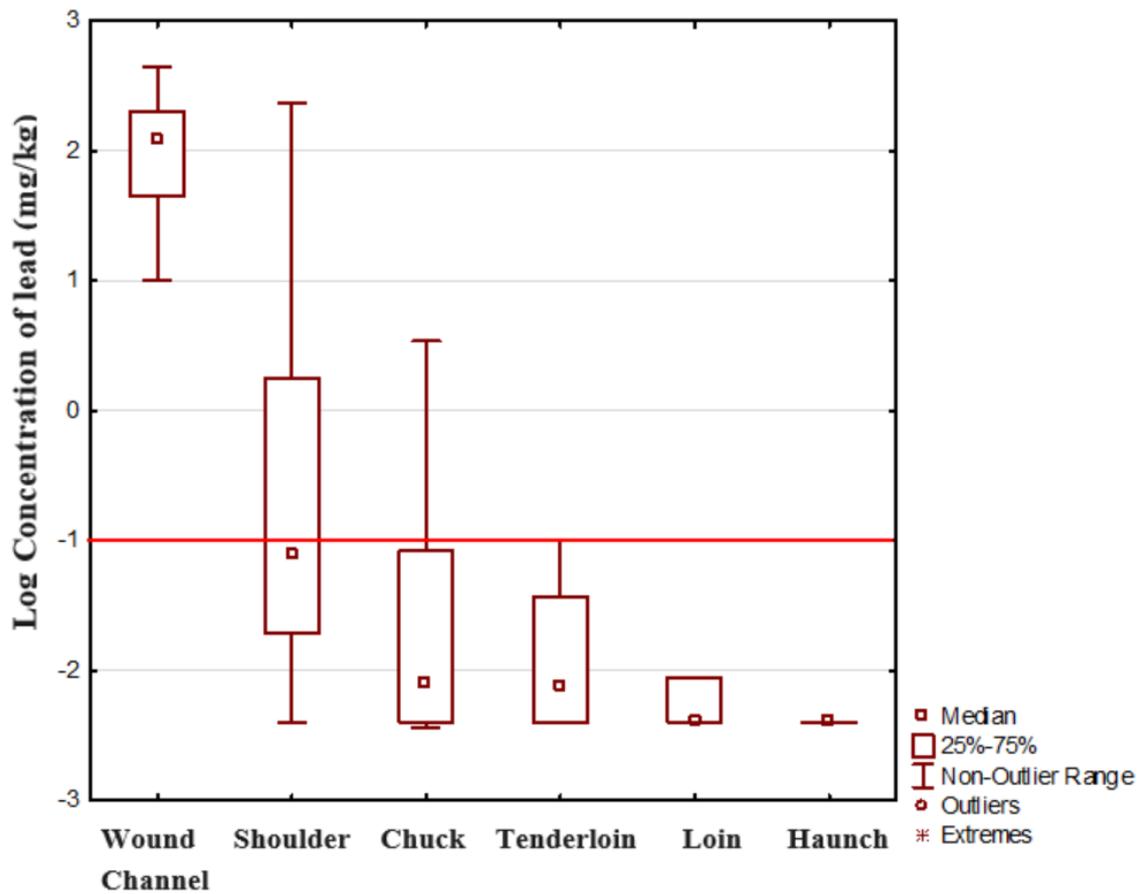


Figure 13. For fallow deer and roe deer, the lead levels decreased significantly with increasing distance from the wound channel. The natural logarithm of the lead levels are displayed, in order to illustrate the variations at lower concentrations. For comparison, the red line shows the maximum permitted value, 0.1 mg/kg, for lead in meat from cattle, pigs, sheep and poultry that are to be offered for sale. There is no maximum permitted level for game meat.

Game shot with copper bullets

The roe deer shot with an E-tip copper bullet only had quantifiable levels of lead in the tenderloin, which contained 0.02 mg lead/kg, whilst the wound channel from the moose calf shot with a Lapua Naturalis copper bullet contained 0.04 mg lead/kg. The wild boar shot with a copper bullet was not analysed with respect to lead. No bullet fragments could be seen in the x-ray of the animals shot with copper bullets (Figure 14).

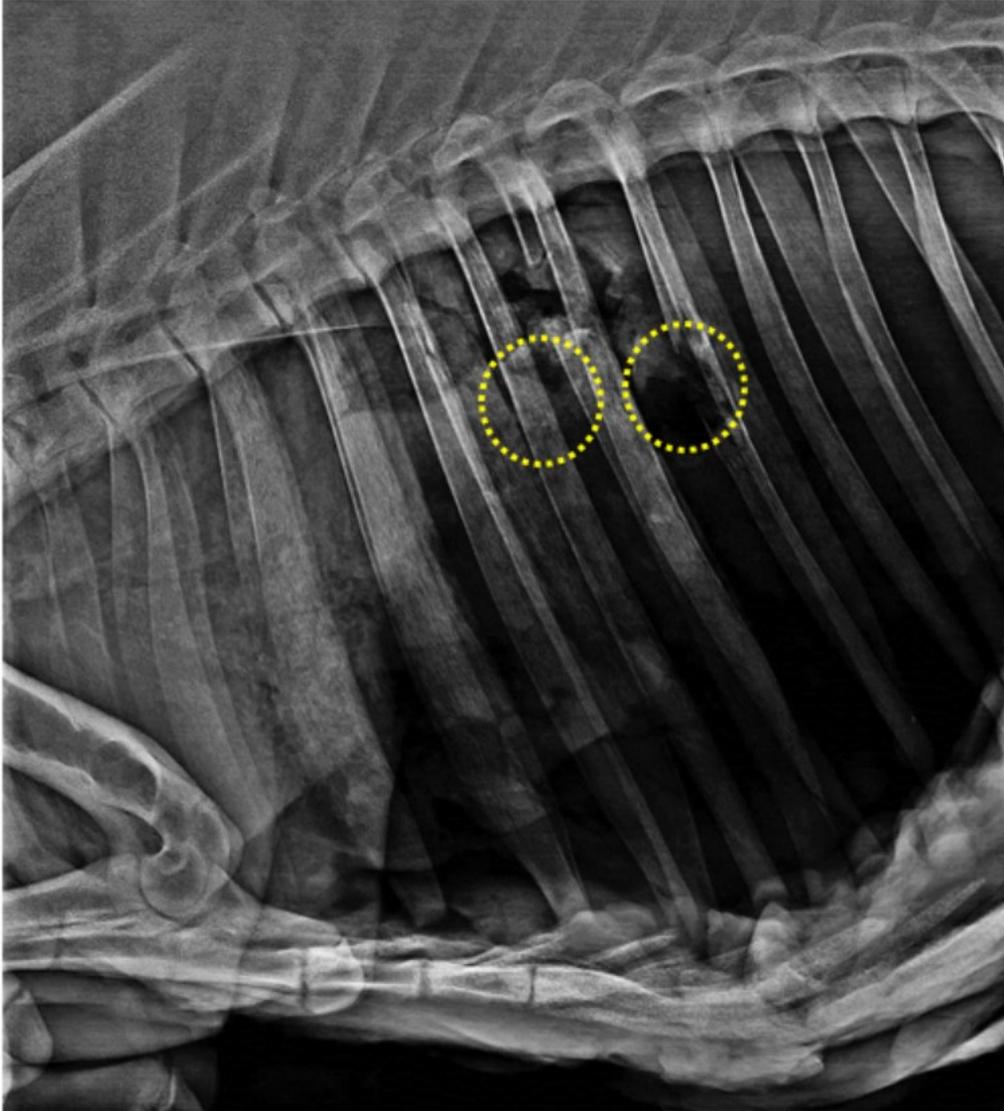


Figure 14. X-ray image of wild boar shot just behind the shoulders with a copper bullet from a distance of 40 m. No fragments from the bullet could be seen in the x-ray. Entrance (left) and exit (right) holes through the chest cavity are marked with circles. Photograph: SVA and SJF, 2014.

Lead levels in game taken with lead shot

The lead levels in crows shot with lead shot were high prior to cleaning of the wound channels (mean value 111 mg/kg, n= 20). After cleaning, the lead level fell by approximately 100 times (mean value 0.78 mg/kg). The cleaned breast muscles from crows shot with the Gyttorp Grouse hunting cartridge had significantly lower lead levels than the untrimmed breast muscles (Wilcoxon signed-pairs= ; Z= 1.99; n=10; p< 0.05). After trimming, one out of the ten crows shot with a hunting cartridge was over the maximum permitted value of 0.1 mg lead/kg for beef, compared with three crows in ten prior to trimming (Table 3). One of the untrimmed crows had a very high value, since a whole shot was still present in the breast muscle. Figure 15 shows x-ray images of crows shot with the Saga Elite sport shooting shell, with US 7 size shot. Four crows still had shot in their breast muscle and six crows had fragments that were visible under x-ray. Of the cleaned crows shot with the Sage Elite Sporting shell, eight out of ten were over 0.1 mg/kg. The untrimmed crows shot with the sport shooting shell had a mean content of 39.3 mg/kg, which equates to nearly 400 times the maximum permitted level in beef. The cleaned crows that had been shot with the hunting shell had significantly lower lead levels than the cleaned crows shot with the sport shooting shell (Mann-Whitney; Z= 3.14 n=10+10; p= 0.002), (Figure 16).

The cleaned cuts from the hare shot with lead shot varied between 0.02 and 5.5 mg/kg, whilst the samples from the roe deer shot with lead shot varied between 0.1 and 21 mg/kg. Fragments could be seen in the x-ray images of the roe deer taken with lead shot (Figure 17) and the hare.

Table 3. Lead levels in crows (K) shot with lead shot of various brands - Saga and Gyttorp. Lead level in mg/kg.

Type of ammunition	Saga		Gyttorp**		
	Crow no.	Untrimmed*	Cleaned cut	Crow no.	Untrimmed*
K1	39.3	0.148	K11	0.822	0.009
K2	39.3	0.102	K12	0.016	0.014
K3	39.3	0.931	K13	0.033	0.012
K4	39.3	0.111	K14	1835	0.072
K5	39.3	0.050	K15	0.680	0.862
K6	39.3	0.037	K16	0.009	0.007
K7	39.3	6.07	K17	0.023	0.007
K8	39.3	5.02	K18	0.014	0.011
K9	39.3	0.832	K19	0.021	0.005
K10	39.3	1.23	K20	0.006	< 0.004
Mean	39.3	1.45	Mean	184 (0.18)	0.10
Median	-	0.49	Median	0.02 (0.02)	0.01
Standard deviation	-	2.21	Standard deviation	580 (0.33)	0.27
Max	-	6	Max	1,835(0.86)	0.86
No. of samples	10	11	No. of samples	10 (9)	11

* Trimmings from all individuals were combined into a single sample which was analysed, and consequently an average for untrimmed breast fillets can be calculated for crows shot with Saga shot.

** Values within parentheses were calculated without the extreme value from K14 untrimmed (1,835 mg/kg).

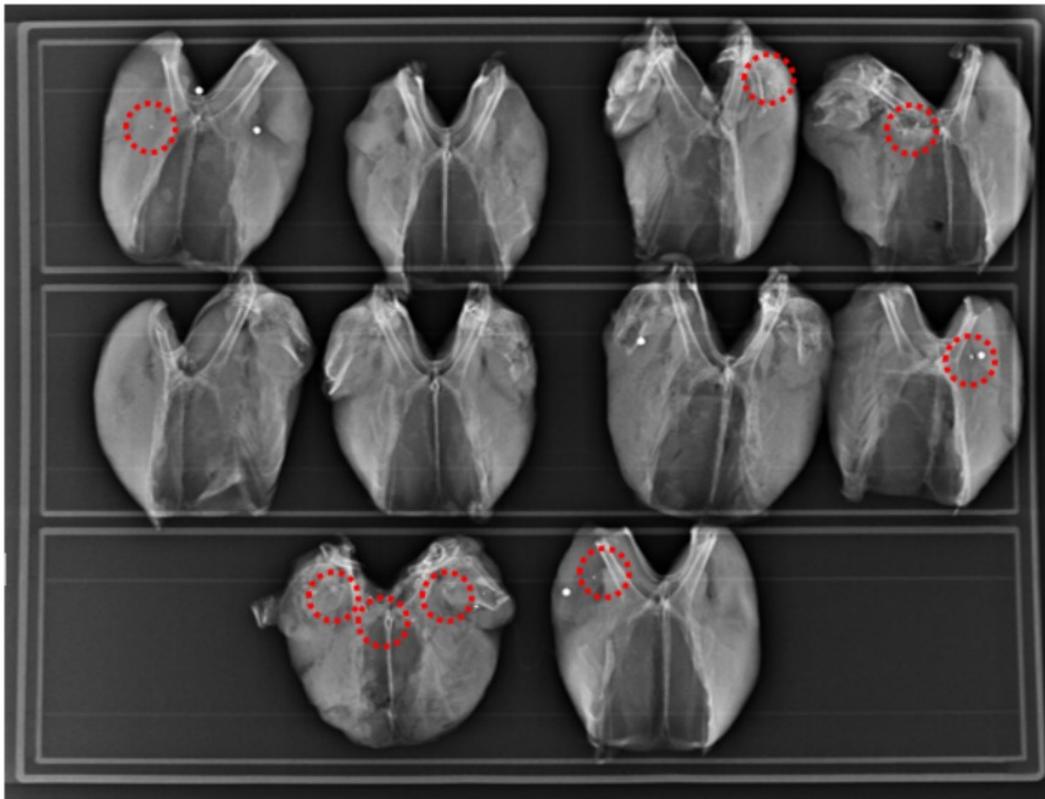


Figure 15. Ten debreasted crows that were shot with the Saga Elite shell intended for sport shooting. Four crows still had shot in their breast muscle and six crows had fragments that were visible under x-ray (marked with red rings). After the breast bone had been removed and the breast fillets trimmed, the lead level was still over the maximum permitted value for beef for eight of the ten crows. Photograph: SVA and SJF, 2014.

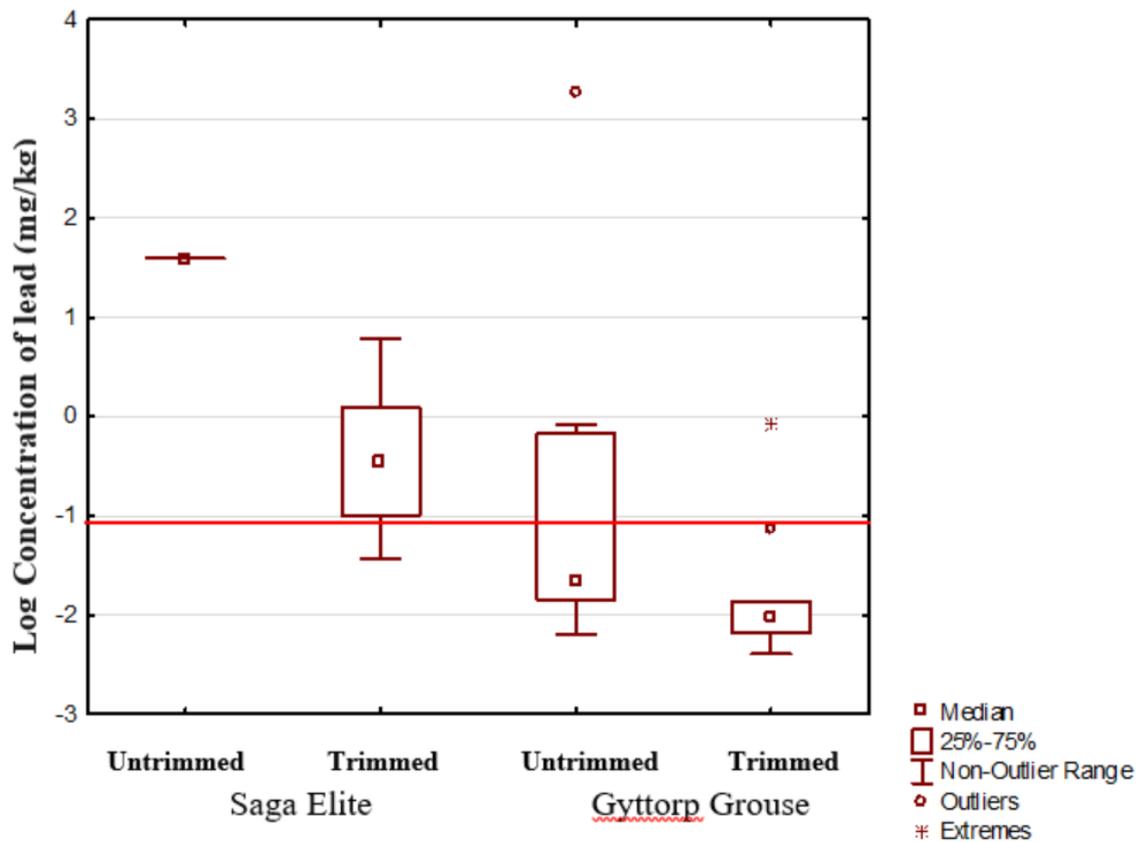


Figure 16. Crows shot with the sport shooting Saga cartridge had higher lead levels than crows shot with the Gyttop Grouse shell intended for hunting, at the same time as trimmed breast muscles clearly had lower lead levels than untrimmed ones. The natural logarithm of the lead levels are displayed, in order to illustrate the variations at lower concentrations. For comparison, the red line shows the maximum permitted value, 0.1 mg/kg, for lead in meat from cattle, pigs, sheep and poultry that are to be offered for sale. There is no maximum permitted level for game meat.

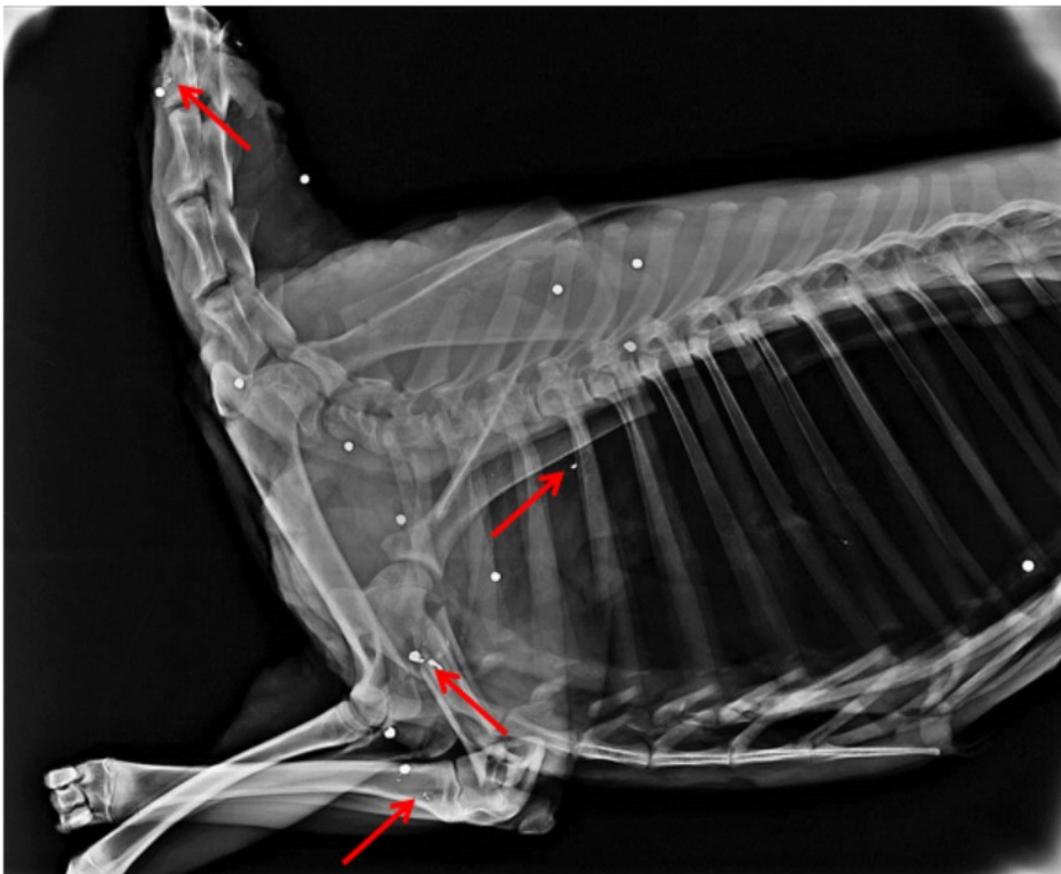


Figure 17. Roe deer shot with lead shot. The image shows both whole shot and fragments of shot that hit bone. Some of the fragments have been marked with red arrows. Photograph: SVA and SJF, 2014.

Solubility of metallic lead

The results from the solubility experiments can be seen in Figure 18, where the proportion of dissolved lead is described in percent as a function of time. The diagram shows that, after the first half hour, samples subjected to a rocking motion have a higher percentage of dissolved lead than those left still. After 1 hour, 1-2 per cent of the lead has dissolved in the rocking samples whilst the figure is just under 0.5 per cent for the lead left still. The difference between the still and the rocking samples increases over time. The diagram also shows that if the speed of the rocking motion is increased, then the lead dissolves quicker ("Increased rocking" compared with "Rocking"). After two days, the rocking motion was stopped and all samples were left still for the rest of the experiment. This can be seen in the upper diagram of Figure 18 where all four samples show the same speed of dissolution (the same inclination) from 51 hours onward. In the experiment with "Increased rocking" no sub-samples were taken after 20 hours. The solutions were instead left still and, after three months no visible traces of lead particles could be seen in any of the test tubes.

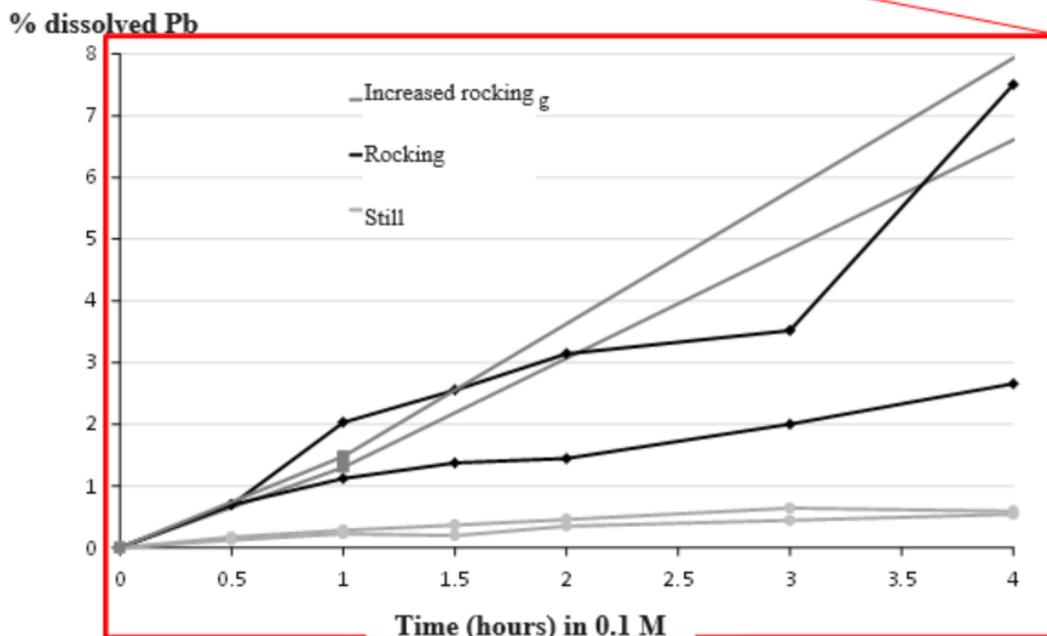
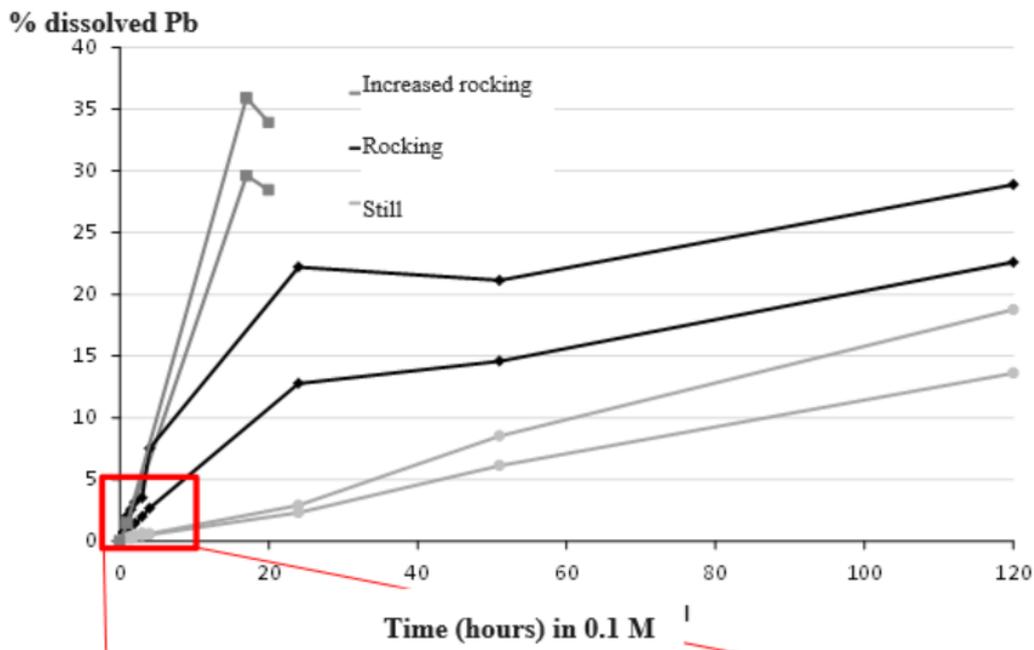


Figure 18. The proportion of lead (Pb) in percent that has dissolved in the gastric environment (0.1 M hydrochloric acid) after a certain time. At the start, 8 mg of metallic lead in the form of metal shavings was placed into 40 ml of nitric acid. The four darker curves at the top show the percentage of dissolved lead from samples that were rocked slowly at various speeds - "Rocking" and "Increased rocking" respectively. The two lighter curves at the bottom show the percentage of dissolved lead from the motionless (still) samples.

Results for other metals

In the minced moose meat that was analysed using ICP-AES, levels of cadmium, copper, zinc, manganese, nickel and vanadium were evaluated along with lead. The levels of cadmium, nickel and vanadium were very low throughout and lied under the detection limit. The levels of copper, zinc and manganese found correspond roughly to the levels normally found in the muscles of domestic animals. In no case was the level so high that a metal fragment could be suspected as being the cause.

In an ICP-MS analysis of the various cuts, levels of copper and antimony were also evaluated. No heightened levels of copper were noted in the samples taken from animals shot with bullets, compared with the control values taken from game that died of non-hunting-related causes. Only one wild boar showed a heightened level of antimony, 1.6 mg/kg compared with other samples that contained below 0.01 mg/kg.

Discussion

General

The results show that high levels of lead often occurred in game meat intended for consumption. Around 30 percent of the minced moose meat packets examined, and the cuts, contained lead levels over the maximum permitted level for lead in beef (0.10 mg/kg). The levels varied, from under the detection limit up to hundreds of mg/kg. There are several studies that show that high levels of lead occur in game meat in Europe and some of these results are compiled in Table 4. The median values in the various studies are relatively low and the majority lie around 0.02-0.03 mg/kg, while levels up to hundreds of mg lead/kg meat have been recorded. Thus, our results follow a general pattern in the literature. The median values in Table 4 are around 2-3 times higher than the normal level of lead in meat in the Swedish market, < 0.008 mg/kg (Table 5). The results in Table 5 are obtained from accredited analysis methods where small sub-samples of around 1 gram are analyzed. The levels obtained lie close to or under the quantification limit for the analysis methods. In an analysis of nutrients in muscles taken from moose (n= 6) lead levels were also analysed and the mean level was 0.011 (<0.005-0.029) mg/kg (National Food Agency's report 18/2008). Even here it could be seen that the lead level somewhat exceeded the normal level for uncontaminated meat. This means that with routine analyses, where only a small sub-sample is analysed, it is possible to detect traces of lead residues, albeit at lower levels than with Lindboe's method (Lindboe 2012).

The results from the solubility experiments showed that lead fragments from bullets dissolve in hydrochloric acid of the same concentration as in the stomach of humans. The solubility increases with increased movement.

Table 4. Comparison with other surveys of the presence of lead ammunition residues in game meat.

	Species	Ammunition	Number of samples	Median, mg/kg	Mean, mg/kg	Standard dev., mg/kg	Max. mg/kg	% exceeding 0.1 mg/kg
EFSA ^a	game	-	2,521	0.02	3.15	-	867	-
BfR ^b	wild boar	bullet	-	0.02	4.7	-	288	-
SLV ^c	wild boar, roe deer, fallow deer calves	bullet	104	0.050	9.9	35	235	43
NVI ^d	Minced moose meat	bullet	52	0.3	5.6	20	110	31
SLV ^c	Minced moose meat	bullet	54	0.027	0.9	3.0	31	33
IREC ^e	bird	lead shot	128	-	2.55	0.75	-	55
SLV ^c	bird	lead shot	20	0.06	0.78 (111) ^f	1.7	6	45

^a Efsa, 2010a. EFSA panel on contaminants in the food chain (CONTAM); Scientific opinion on lead in food. EFSA J. 8(4), 1570.

^b Bundesinstitut für Risikobewertung, "Bleibelastung von Wildbret durch Verwendung", Stellungnahme Nr. 040/2011.

^c National Food Agency, Sweden, 2014. Results from this report, Tables 1, 2 and 3 (without wound channel and trimmed meat) and Appendix 2.

^d Lindboe, M., et al. Lead concentration in meat from lead-killed moose and predicted human exposure using Monte Carlo simulation. Food Additives and Contaminants (2012) 1-6, iFirst.

^e Mateo, R., et al. 2010. Bioaccessibility of Pb from ammunition in game meat is affected by cooking treatment. PLoS One 6, e15892.

^f The value within parentheses, 111 mg/kg, shows the lead level in untrimmed crows.

Table 5. Analyses of lead in various types of meat conducted under the management of the National Food Agency, 2011-2013. The analyses were conducted using accredited routine methods with small sub-samples (approximately 1 gram). All game meat and the majority of other meat came from game handling facilities in Sweden. The remainder were purchased in food stores.

Animal	n		Mean, mg/kg		Max. mg/kg
Moose ^{1,2}	71		0.010		0.143
Wild boar ^{1,2}	23	<	0.008		
Roe deer ^{1,2}	18	<	0.008	<	0.008
Fallow deer ²	4	<	0.008	<	0.008
Red deer ²	1		0.003		
Reindeer ²	5	<	0.008	<	0.008
Lamb ²	4	<	0.002	<	0.002
Cattle ²	10	<	0.002	<	0.002
Turkey breast ²	1	<	0.002		

¹ The samples were analysed by ALS Scandinavia in Luleå. Source: The National Food Agency's report series no 9/2012. Kontroll av rests substanser i levande djur och animaliska livsmedel [Control of residual substances in living animals and animal-based foodstuffs]. Uppsala: National Food Agency.

² All samples consisted of combined samples where 4-10 different meat samples were included within each sample. Analyses were conducted by the National Food Agency. Source: The National Food Agency's report series no 24/2013. Kött - analys av näringsämnen [Meat - analysis of nutrients]. Uppsala: National Food Agency.

Meat from game shot with bullets

The analyses of x-ray images showed that game shot with expanding bullets with a lead core regularly contained hundreds of lead fragments of varying sizes prior to cleaning, and the analyses of lead levels showed that meat from the wound channel contained very high levels of lead. The results are in agreement with previous studies (for example, Dobrowolska & Mellosik 2008, Hunt et al., 2009, Knott et al., 2010, Lindboe et al., 2010).

Hunt et al., (2008) delivered white-tailed deer that had been shot and x-rayed to various commercial abattoirs, which had the task of cutting the animals into bone-free meat and minced meat. After cutting, all the packets of minced meat were x-rayed. Fragments could be detected in a total of 32 per cent of all the packets, in at least one packet for 80 per cent of the carcasses and, on average, in 32 per cent of the packets from each individual deer (Hunt et al., 2009). Our investigations show that the wound channel in roe deer, fallow deer fawns and wild boar shot with bullets with a lead core contained hundreds of bullet fragments, just as in the investigations conducted by Hunt et al. (2009) and Knott et al. (2010). The packets of minced meat and cuts of meat intended for consumption contained fewer fragments or no fragments at all that could be distinguished under x-ray. Consequently, standard cleaning removes the vast majority of the fragments. However, occasional fragments are enough to give unacceptably high lead levels, and just removing the meat that looks to be affected by the bullet (here defined as ‘wound channel’) is not sufficient. Further cleaning is required in order to remove all the lead (Hunt et al. 2009, results above).

The probability that lead residues are present in game meat intended for consumption will largely depend on how much meat was cut away in the cleaning process. Hunt et al. (2009) found that fragments could be located as far away from each other as 45 cm, which means a spread of up to 22.5 cm from the wound channel. Dobrowolska & Melosik measured lead levels up to 30 cm from the wound channel in wild boar and red deer and found that the meat still contained lead levels over the maximum permitted value for lead in beef for 8/10 of the wild boar and 5/10 of the red deer. Furthermore, the authors noted that, with heavier individuals, the lead levels were higher in the meat on the entrance wound side than on the exit side, whilst the opposite was true for lighter individuals. It was proposed that this difference is due to the fact that a bullet that hits a heavier individual meets greater resistance and therefore expands more rapidly, at the same time as the wound channel is longer in a bigger, heavier animal. Consequently, a larger proportion of the fragments are deposited in the first half of the wound channel (Dobrowolska & Melosik 2008). The study encompassed ten red deer and ten wild boar that were shot with ammunition of an unknown type. The size of this random sample is therefore relatively small, at the same time as variation in the choice of ammunition almost certainly accounts for some of the variations in the lead levels. Despite this, the results clearly show that levels of lead can be high far away from the wound channel itself. Dobrowolska & Melosik (2008) drew the conclusion that it is necessary to clean a margin outside of the bloodshot meat around the wound channel.

Our results show that there were quantifiable levels of lead in 54 per cent of the packets of minced moose meat, and that there were fragments visible under x-ray in 35 per cent of the packets. However, in our investigation, only one packet of minced meat was collected from each hunter. It is probable that certain packets contained fragments and elevated levels of lead even from those moose where our analysed sample showed this was not the case. Our proportion of packets

containing fragments is very similar to that found by Hunt et al. (2009) (35 per cent compared with 32 per cent). The results from Hunt et al. (2009) may therefore give an indication of the percentage of moose that are not cleaned thoroughly enough to remove all the lead. White-tailed deer are, however, significantly smaller than moose. A bullet that hits a moose meets greater resistance and will provide a longer wound channel than a bullet that hits a white-tailed deer, probably resulting in a greater amount of lead deposited in the form of fragments around the wound channel. At the same time, there is a "diluting" effect due to the larger amount of moose meat that is cut and ground, which makes finding fragments in any given packet of minced meat less likely. The relative importance of these two factors cannot be determined, but it seems evident that the proportion of moose carcasses where at least one packet of minced meat contains fragments or quantifiable levels of lead is clearly higher than 35 or 54 per cent respectively. In a corresponding Norwegian survey where lead levels in packets of mince moose meat from hunters were investigated, elevated levels were found in 80 per cent of the packets (Lindboe et al. 2012).

We found quantifiable levels of lead in five out of six packets of minced moose meat bought over the counter and the mean level was higher than in the minced meat from the hunters' own freezers. However, the sample size is not sufficient to allow a comparison of minced meat from private hunters with minced meat bought over the counter with a reasonable statistical power. Lindboe et al. (2012) also found lead levels over the maximum permitted level for beef in minced moose meat purchased over the counter in Norway, and in an investigation of blood lead levels in consumers of game meat in Norway, those who had purchased game meat over the counter had higher blood lead levels than those who ate privately supplied game meat (VKM 2013). Furthermore, Hunt et al.'s (2009) investigation, (see above) is based on slaughtered game from commercial processing plants. The problem with lead residues from ammunition in game meat is therefore not exclusive to game slaughtered privately for own consumption; it also applies to meat from commercial game processing plants, which is offered for sale.

As far as the wild boar shoulders are concerned, the amount and size of the fragments was affected by whether or not the bullet met hard resistance. The difference in the amount and size of the fragments was also reflected in the difference in lead level for meat from the wound channel, which is a result also found in previous studies (for example Dobrowolska & Melosik 2008).

Corresponding patterns for roe deer and fallow deer fawns are however lacking in our study. There are several possible reasons for this. Firstly, our sample size is small, which means that it will only be possible to statistically demonstrate relatively large effects and differences. Secondly, the humerus of a roe deer or fallow deer provides less resistance than those of a wild boar, reducing the variation explained by differences in shot placement. Thirdly, samples from the entrance and exit wounds were added together in the analysis of levels for roe deer and fallow deer fawns. There is therefore a "diluting effect" with less affected meat when the bullet has only hit the humerus on one side, which was often the case.

The lead levels from the wound channels were considerably higher for the wild boar shoulders than they were for roe deer and fallow deer fawns. However, the latter also contained on average several hundred mg lead/kg, at the same time as the cleaned shoulder meat contained several tens of mg lead/kg.

The lead level fell rapidly from the wound channel to 10 cm from the wound channel for the wild boar shoulders, whilst there were no significant differences between 5-10 cm from the wound channel and 10-15 cm. The level of lead also decreased for roe deer and fallow deer fawns as the distance from the wound channel increased, and the rate of the decrease was smaller as the distance increased. In the cuts of meat farthest from the wound channel, no quantifiable levels of lead were found (< 0.004 mg/kg).

The German Federal Institute for Risk Assessment (BfR), together with Deutsche Jagdverband (The German Hunting Association), has conducted equivalent studies in Germany, but with a considerably more extensive number of roe deer, wild boar and red deer. According to the BfR, they also found elevated levels of lead in the haunch of roe deer shot with bullets with a lead core (Lahrssen-Wiederholt 2014). However, in the steaks analysed in our study we found no elevated levels of lead. There are many different factors that can contribute to any differences, such as the number of game shot and studied, the calibres used, the choice of ammunition types, shot placement and the distance from which the shot was taken, gutting and cleaning procedures and sampling routines. It is impossible to determine what has led to the differences in the results in the two investigations.

The majority of lead fragments are small, flat and soft. They can easily be rinsed away with the blood in the chest cavity, something that was apparent when the x-ray images were analysed. There was also an abundance of lead fragments in the clotted blood between shoulders and the chest cavity. This is something that should be taken into consideration when gutting and cleaning. It is also easy to spread lead fragments with a knife or chopping board. In several cases it appears that fragments may have been spread from the region around the wound channel to other parts of the carcass, since isolated fragments were found 10-15 cm from the wound channel despite the meat 5-10 cm from the wound channel lacking any fragments. It seems unlikely that small, light fragments were transported there by the bullet's shock wave. Handling procedures should be designed so that there is minimal risk of transferring of fragments between different parts of the carcass.

Factors that affect the amount of fragments from bullets

The bullet expansion and the amount of lead fragments that become detached from the bullet depend on the bullet's construction, its velocity and the resistance the bullet encounters. The bullet's velocity at impact depends on the muzzle velocity, the weight and shape of the bullet and the distance to the target.

Hunt et al. (2009) x-rayed 30 gutted white-tailed deer, *Odocoileus virginianus*, that had been shot with the bullet with a lead core most commonly used in Wyoming, USA. The carcasses contained on average 136 fragments, which were noted to consist of lead of the same isotopic composition as ammunition lead. Knott et al. (2010) had ten red deer and two roe deer shot in the same way. The gutted carcasses contained on average 356 fragments that could be seen using x-ray, and an average fragment weighed 3.7 mg. Based on the number of fragments and their size, Knott et al. (2010) calculated that an average of 1.2 grams of bullet fragments was deposited in the carcasses (Knott et al. 2010). If this method is used to count and measure fragments, then the amount of fragments deposited will probably be underestimated, however, since really small fragments cannot be seen under x-ray.

Stokke et al. (2010) have investigated a large number of bullets collected from moose shot in Sweden, Norway and Finland. On average, the bullets investigated lost 2.69 grams in weight, and the hunters who collected the bullets stated that they would take, on average, 1.4 shots per moose. The authors came to the conclusion that 3.77 grams of lead were deposited as fragments in an average moose shot in Fennoscandia. However, they disregard the fact that parts of the bullet's weight loss is down to loss of the jacket. This portion differs greatly between various bullet makes and models, but for the bullets used by Hunt et al. (2009) for example, the lead core constituted 68 per cent of their weight. Furthermore, Stokke et al. (2010) did not investigate the bullets that had been shot through moose. Through and through shots are more common than shots where the bullet remains in the moose, something that Stokke et al. (2010) also demonstrated. Hard bullets that give off less lead are more likely to penetrate and exit the animal, at the same time as bullets that meet great resistance in the form of bones leave more lead and are more likely to remain in the animal. Consequently, an investigation that only examines the bullets that remained within the moose will overestimate the amount of lead deposited in the average moose.

The difference between the methods for calculating the amount of lead deposited is of no great significance to the conclusions as far as handling recommendations are concerned, bearing in mind the amounts of lead involved. If one proceeds from the maximum permitted value of 0.1 mg/kg, which applies for lead in meat from cattle, sheep, pigs and poultry that are to be offered for sale, then even the lower figure of 1.2 grams of deposited lead is enough to contaminate 12 tonnes of game meat up to the maximum permitted level. One single, average fragment of 3.7 milligrams is enough to contaminate 37 kilos of game meat. This equates to roughly three times the carcass weight for a roe deer or fallow deer calf. Even if the weight of the deposited bullet fragment is adjusted to reflect the amount of lead by bullet weight, the figures are still very high, with regard to the maximum permitted value. However, it is not the amount of lead deposited that is of interest, rather how much remains after the shot has been cleaned.

Calibre

Hunt et al. (2009) had all their white-tailed deer shot with a standard bullet with a lead core fired from a 7 mm Remington Magnum calibre, whilst Knott et al. (2010) used the same procedure but with a .270 Winchester calibre. The roe deer and fallow deer fawns analysed here were all shot with a .308 calibre Winchester, which is one of the most common calibres used in Sweden. The .308 Win. exceeds the energy requirements for a class 1 weapon, which is required for shooting the largest species of game in Sweden (e.g. moose, wild boar, bear, red deer and fallow deer). However, this calibre lies in the lower part of the segment of calibres that satisfy the requirements for class 1 weapons. Had a larger calibre (larger bullet diameter) or a faster calibre (higher muzzle velocity) been used, then a greater amount of lead would probably have been deposited in the form of fragments, regardless of the construction of the bullet, the shot placement or the distance to the prey. The calibres that Hunt et al. (2009) and Knott et al. (2010) used are faster than the .308 Winchester, but the results seem at least qualitatively comparable.

Distance to the prey

The roe deer and fallow deer fawns were shot at a distance of 60-130 metres, which is considered to be a normal distance in Sweden, when hunting this quarry with rifles. Longer and shorter distances do of course also occur, which would result in lower and higher impact velocities,

respectively. Accordingly, the size of the wound channel and the amount of lead in the carcass would also vary more than is the case in the investigation.

The physical and chemical design of the bullet

The expansion of a lead core bullet at a given velocity and a given resistance is determined by the thickness of the jacket, its construction, whether the lead core has been fused together with the jacket (bonded or non-bonded), and the extent to which antimony has been added to the lead in the core (see also Appendix 1). Here, only three different bullets with lead cores were used to shoot the roe deer and the fallow deer fawns. There are however, hundreds of different bullets of the .308 Win. calibre other than those investigated; some are softer (will release more lead) whilst others are harder (will release less lead). As a result, there will be a greater variation in the size of the wound channel and the amount of lead fragments even for a single calibre in than in the investigation performed.

Bonded bullets have a greater residual weight than non-bonded ones and higher lead levels should therefore be expected for meat in the wound channel where conventional bullets have been used, rather than bonded ones. Our investigation indicated no difference, however. The small sample size in our studies makes it difficult to distinguish anything other than very clear differences. A limited expansion firing of the bullets used showed that the difference in residual weight between the two bullets primarily used was very small. Furthermore, it is possible that the wound channel is larger for conventional bullets than for bonded ones, which means that more meat is removed in cleaning. Therefore, a greater absolute quantity of lead will be "diluted" with more meat, which gives a lower level than if the same amount of lead is deposited in a smaller wound channel. This might explain the lack of a difference in the levels for the wound channels. Looking at the cuts that are to be consumed, the levels do not need to be different at all if sufficient cleaning of the shot has been conducted for carcasses shot with both types of bullets.

Another possible explanation for why we did not find any differences between the bullet types is that not all the lead may have dissolved during the analyses. When we re-x-rayed the leached samples from the wild boar shoulders, we noted that there were still some metal fragments that had not dissolved completely in some of the samples. This applied primarily to wound channel samples that contained a greater number, and larger fragments. It takes longer for large fragments to dissolve than small fragments. This means that there is probably a greater risk that not all the lead fragments dissolve from conventional bullets dissolve, than there is with bonded bullets. This could also be a reason why we did not find any differences in the lead levels for the wound channels.

The German BfR also compared bonded and unbonded bullets in its more extensive study. They did not find lower lead levels for bonded bullets compared to conventional bullets in their cuts of meat, and in some case they even found higher levels in cuts from animals shot with bonded bullets (Lahrssen-Wiederholt 2014).

The distribution of fragment sizes is probably important for absorption if game meat containing lead fragments is ingested. Small fragments have a greater area in relation to their volume, and will therefore dissolve faster than larger fragments in the acidic gastric environment. Larger fragments will probably only partially dissolve before they are transported further to the intestines' more

alkaline environment. It is therefore possible that a small amount of lead, in the form of small fragments, can lead to a greater amount of absorbed lead than a larger total amount of lead that is divided up into large and small fragments. However, there are cases described in literature where people have suffered lead poisoning as a result of larger particles becoming lodged in their stomach/intestinal system (e.g. Gustavsson 2005). The symptoms disappeared following removal of the lead particles.

Neither the Swedish nor the German results indicate that the risk of consuming lead residues from ammunition is less with game taken using bonded as compared to non-bonded bullets.

The handling of game shot with bullets

As can be seen above, there are many different factors that affect the amount of lead deposited, and this makes it difficult to provide unequivocal handling recommendations. In general, a higher velocity and a larger, expanded diameter will produce a greater shock wave and, consequently, a larger wound channel. A greater number of fragments will also contribute to a larger wound channel. Consequently, a great deal of the variation depends on the size of the actual wound channel. A recommendation that is based on the removal of all affected meat (the wound channel and any bloodshot meat), along with a margin of unaffected meat, is probably more in line with the existing variation in the spreading of lead fragments than a recommendation to just "cut away xx cm". Furthermore, a recommendation based around the wound channel is probably easier for hunters and game handling facilities to apply. At the same time, less meat will be cut away unnecessarily.

The total amount of meat that will need to be removed will depend on the resistance the bullet has encountered, the calibre and construction of the bullet, and the impact velocity. Higher velocity and higher resistance lead to more fragments, which means that the wound channel itself will be larger in diameter. In the animals investigated, the wound channel itself (defined as all affected meat) normally extended to a 5-15 cm radius.. Therefore, a 15-25 cm radius around the actual bullet hole would normally have to be removed (wound channel, including all affected meat + 10 cm unaffected meat), in order to achieve an acceptable probability that the game meat no longer contains residues from the lead core. For smaller ungulates such as roe deer and fallow deer fawns, it is not therefore possible to eat shoulder meat from an animal shot through or just behind the shoulder without a relatively high probability of encountering lead fragments in the meat.

Prevent the spreading of lead fragments when gutting and cutting

Lead fragments were found throughout the chest cavity and in clotted blood between the shoulders and the chest cavity. To minimise the risk of lead residues in game meat intended for consumption, care must be taken to both trim bruised areas and to adapt the gutting process. Traditionally, game is often gutted out in the forest; a cut is made through the diaphragm and all the internal organs are removed. Blood from the chest cavity will then typically flow back into the abdominal cavity, and this may take some lead fragments with it. A more suitable procedure is to avoid cutting through the diaphragm if the bullet has only penetrated the chest cavity, but to just remove the organs from the abdominal cavity. The heart and lungs can be removed after the game has been transported from the forest. Ideally, the chest cavity should be opened whilst the animal is hanging by its hind legs, which means that the blood runs out of the body without lead residues passing into the abdominal cavity. This procedure is already recommended as a normal procedure for good slaughter hygiene, but the reduced spreading of lead fragments is yet another reason not to cut through the diaphragm unnecessarily. It is of course possible to hang the animal by its hind legs and gut it out in the forest if so desired, if it is possible to hoist the animal up.

Unintentional spreading of fragments will occur in the hunters' practical handling of the meat if measures are not taken to prevent this. Cleaning hanging game reduces the risk that fragments are spread between cuts that are handled on the same chopping board. In addition to this, it is also necessary to carefully wipe off or wash the knife after the wound channel has been cut away, and again after the margin of unaffected meat has been removed.

After gutting, the inside of the chest cavity will still have some lead fragments. Some of these can be wiped off with paper, or rinsed away with clean water at low pressure. It is very important however to ensure that fragments are not spread to other parts of the carcass, either with the paper or the water. There are investigations that show that the rinsing of carcasses leads to the spread of fragments (Cornicelli & Grund 2008). Furthermore, it is recommended for reasons of hygiene that, generally speaking, carcasses should only be rinsed in professional facilities with controlled water and where air humidity can be controlled (SJF's training in meat handling).

Meat from game shot with lead shot

Crows, roe deer and hares revealed heightened levels of lead in game meat and lead shot and fragments of shot were present in certain cuts of meat. There were major differences in the lead levels of the crows shot with sport shooting shells and those shot with shells intended for hunting. Crows shot with sport shooting shells still contained comparatively high levels of lead even after all visible wound channels from each individual piece of shot had been trimmed away. As far as the hunting shells are concerned, one in ten crows still contained levels over the maximum permitted level for beef to be offered for sale after cleaning, compared with three in ten prior to cleaning. It is impossible to say whether this pattern is representative for sport shooting shells or hunting shells, respectively, based on a comparison of one shell from each category. It is obvious, however, that there are major differences between various types and brands of shells.

The results concur with Pain et al. (2010), who found shot or fragments of shot in 87 per cent of untrimmed wildfowl purchased over the counter or direct from hunters. Several previous studies have indicated heightened blood lead levels in groups that consume large amounts of game shot with lead shot (for example, Tsuji 2008 a,b). According to Green & Pain (2012), 2-3 portions per month of wildfowl shot with lead shot is enough to have an effect on a child's emotional development. If wildfowl shot with lead shot are eaten several times a week, there can be a heightened risk of chronic kidney disease, or an increased risk of miscarriage. The average levels Green & Pain used were 1.18 mg/kg, which is lower than the mean value for both untrimmed (39.2 mg/kg) and trimmed (1.45 mg/kg) crows shot with Saga ammunition in our investigation. On the other hand, the mean values for untrimmed (0.18 mg/kg) and trimmed crows (0.1 mg/kg) shot with the Gyttorp Grouse hunting ammunition were clearly lower than the level that Green & Pain (2012) used, if one disregards the untrimmed breast fillet that contained a whole piece of shot.

The investigation of game shot with lead shot was primarily conducted on crows which, in terms of size, correspond to smaller game such as wood pigeon. Exactly as with game shot with bullets, there is a greater probability that lead shot will fragment if it encounters hard resistance than if it meets light resistance. If fragments are found in smaller game, then more fragments can consequently be expected in larger game, for any given shell and size of shot. Pain et al. (2010) found more fragments and more shot in larger wildfowl, which could be both an effect of a larger body being hit by more shot, and of the shot fragmenting more easily. At the same time, however there is a diluting effect caused by the larger amount of meat in larger game, which means that the lead level per kilo need not be greater. Finally, the whole situation is further complicated by the fact that larger shot is used for larger game, which could also affect the probability of the shot fragmenting.

In our study, shot had fragmented when it hit bone both in the case of the hare and the roe deer; both animals had levels that were around 10 to 100 times the maximum permitted level of lead (0.10 mg/kg) that applies for lead in meat from cattle, pigs, sheep or poultry that is to be offered for sale. The material from furred game shot with lead shot is far too small for any certain quantitative conclusions to be drawn, but it is evident that high levels of lead also can occur in furred game shot with lead shot.

Further studies are required in order to ascertain the differences in fragmentation associated with different shells and types of game.

The handling of game shot with lead shot

All hunters probably clean the wound channel from a bullet, albeit to differing degrees. For game shot with lead shot, many probably remove bloodshot areas, whilst few cut away the meat around each wound channel. Based on the crows shot, however, it appears as if both the bloodshot meat and the meat around the shot channels should be trimmed off in order to reduce the risk of getting lead in the game meat. It is not sufficient to just remove the bruised or bloodshot areas. At the same time, the trimming of wound channels also reduces the risk of whole pieces of shot remaining in the meat, which, in rare cases, can have serious consequences in the form of severe lead poisoning (Gustavsson 2005, Treble 2002). Bearing in mind that the analyses also show both fragments and high levels of lead for furred game, the same procedure should be adopted here.

The sporting shell examined, Saga Elite Sporting, left very high levels of lead, even in trimmed meat, but it cannot be said that this is a pattern that applies in general for sport shooting shells.

Metals in addition to lead

Both bullets and shot also contain other metals in addition to lead, e.g., copper, antimony and zinc. As copper and zinc are minerals that occur naturally in meat, they generally occur in considerably higher levels than lead, 1-3 mg/kg and 10-70 mg/kg respectively, (National Food Agency's report series 24/2013) so it is not as easy to see the effect of ammunition residues for these metals. Low levels of antimony, a few mg per kg, are normally found in meat (unpublished data, National Food Agency 2014) and only one sample in this study provided a noticeably higher level of antimony.

The solubility and absorption of lead in gastric environments

The results show that the rocking motion contributes to a more rapid dissolution of lead particles. The relatively soft layers of lead chloride, which are formed on the surface of the lead shavings when it comes into contact with the hydrochloric acid, are probably scraped off through the motion and this allows more lead to dissolve. When the rocking motion stops, this scraping stops and means that the dissolution of the lead continues at a slower rate. The rate of dissolution is then the same in all the samples, regardless of whether they were rocked at the start or left still. The dips and unevenness that can be seen in some of the curves can be attributed to the lead chloride that had formed becoming stuck to the walls of the test tube, and subsequently becoming loose to different degrees in the different sub-samples. There are publications that show that the level of lead in the blood increases when metallic lead is ingested by humans (Gustavsson 2006, Treble 2002) and other animals (for example Oscarsson 1992, Hunt 2009), which suggests that metallic lead is, to a certain extent, available for the body to absorb in the stomach and intestines. There are also studies that show that the body's capacity to absorb lead is affected by its iron status, its calcium status, whether it is ingested with other foodstuff or on an empty stomach (see more information in *parts 3 and 4*). Our results only show the chemical dissolution of lead in the presence of weak hydrochloric acid, which is also found in the stomach. The results clearly show that metallic lead is dissolved and that the dissolution is quicker with increased stirring/motion. Exactly how quickly a lead particle of a certain size could be able to dissolve in the stomach together with ingested game meat prepared in a specific manner together with other foodstuff, and drink, is more difficult to determine. There is also the question of the size distribution of the lead fragments in the game meat, and how this could affect dissolution.

Quality of the chemical analyses

The analytical method used is not a common routine method; it has been especially developed for the analysis of residues of lead ammunition in game meat (Lindboe 2012). The method is not accredited and it suffers from a certain degree of uncertainty when game samples with larger lead particles are analysed, since these particles may not manage to completely dissolve. This leads to an underestimation of the level of lead in the game meat. The quality controls conducted with the method employed show that the lead levels measured come from game meat that has been contaminated with lead, and that the methods provide a sufficiently good estimation of the lead level in game meat.

The analytical instruments ICP-AES and ICP-MS that were used in the studies have both advantages and disadvantages. The advantage of ICP-AES is that it simplifies the processing of samples considerably since the extract can be analysed directly. Another advantage of ICP-AES is that the instruments manage to measure higher levels of lead without getting damaged. One disadvantage is that the detection limit is worse than for ICP-MS if the aim is to measure low levels.

Concluding remarks

There is no doubt that high levels of lead from ammunition residues can be found in game meat intended for consumption. There is also no doubt that there is game meat that does not contain elevated levels of lead, i.e., that is completely unaffected by lead from ammunition residues. With the correct handling, it is possible to reduce the level of lead in game shot with bullets by 1,000 times. Another alternative is to use lead-free ammunition.

References

- Axelsson, J. 2009. Bly från ammunition som förgiftningsrisk hos rovfåglar- en kunskapsöversikt. Viltforum 1/2009. Svenska Jägareförbundet.
http://jagareforbundet.se/Documents/Viltforum/Viltforum1_2009_Bly.pdf
- AEWA 2009. Phasing Out the Use of Lead Shot for Hunting in Wetlands: Experiences made and Lessons Learned by AEWA Range States. <http://www.unep-aewa.org/en/publication/phasing-out-use-lead-shot-hunting-wetlands-experiences-made-and-lessons-learned-aewa>
- Bjeremo, H., Sand, S., Nälsén, C., Lundh, T., Enghardt Barbieri, H., Pearson, M., Lindroos, A.K., Jönsson, B.A.G., Barregård, L., Darnerud, P.O. 2013a. Lead, mercury, and cadmium in blood and their relation to diet among Swedish adults. *Food Chem. Toxicol.* 57, 161-169.
- Bly i viltkött – Riskhanteringsrapport, Livsmedelsverket juni 2012, Uppsala, Sverige
- Christofferson, S., Karlsson, B., Bengtsson, G. & T. Mörner. Jägarskolan. Svenska Jägareförbundets förlag, Öster Malma.
- Cornicelli L. & M. Grund. 2008. Examining variability associated with bullet fragmentation and deposition in white-tailed deer and domestic sheep: preliminary results.
http://files.dnr.state.mn.us/fish_wildlife/lead/bulletstudy/resources/publicsummary.pdf
- Dobrowolska, A. & M. Melosik. 2008. Bullet-derived lead in tissues of the wild boar (*Sus scrofa*) and red deer (*Cervus elaphus*). *European Journal of Wildlife Research* 54:231–235
- European Pharmacopoeia 01/2012:20903, 2.9.3 Dissolution test for solid dosage forms.
- EFSA. 2010. EFSA panel on contaminants in the food chain (CONTAM); scientific opinion on lead in food. *The EFSA Journal*, 8(4):1570.
- Gremse, C. & S. Rieger. 2014. Ergänzende Untersuchungen zur Tötungswirkung bleifreier Geschosse. http://www.bmel.de/SharedDocs/Downloads/Landwirtschaft/Wald-Jagd/BLE-Forschungsbericht-Jagdmunition.pdf?__blob=publicationFile
- Gustavsson P, Gerhardsson L. Intoxication from an accidentally ingested lead shot retained in the gastrointestinal tract. *Environ Health Perspect.* 2005 Apr;113(4):491-3.
- Helander, B., Räikkönen, J., Ågren, E. & H. Borg. 2012. Rapportering från projekt om undersökning av bly i leverprover från havsörn 2005 – 2011. *Naturhistoriska riksmuseets rapport* 16: 2012.
- Hunt, W.G., Watson, R. T., Oaks, J.L., Parish, C.N., Burnham, K.K., Tucker, R.L., Belthoff, J.R., & G. Hart. 2009. Lead Bullet Fragments in Venison from Rifle-Killed Deer: Potential for Human Dietary Exposure. *Plos ONE* 4: 1-6.
- Hägg, G., *Allmän och oorganisk kemi*, 1989, Stockholm : Almqvist & Wiksell, ISBN: 9789120090153
- Iqbal, S. Blumenthal, W., Kennedy, C., Yip, F. Y., Pickard, S., Flanders, W. D., Loring K., Kruger, K., Caldwell, K.L., Brown, M.J. 2009. Hunting with lead: Association between blood lead levels and game consumption. *Environmental Research* 109: 952–959
- Jarzyńska, G. & J. Falandysz. 2011. Selenium and 17 other largely essential and toxic metals in muscle and organ meats of Red Deer (*Cervus elaphus*) — Consequences to human health. *Environment International* 37: 882–888.

Jani, P., Halbert, G.W., Langridge, J., Florence, A. 1990. Nanoparticle uptake by the rat gastrointestinal mucosa: Quantitation and particle size dependency. *J. Pharm.Pharmacol.* 199, 42: 821-826

Jordbruksstatistisk årsbok 2010, Sveriges officiella statistik, Jordbruksverket, Statistiska centralbyrån, ISBN 978-91-618-1527-2

von der Kammer, F., Legros, S., Larsen, E.H., Loeschner, K., Hofmann, T. 2011. Separation and characterization of nanoparticles in complex food and environmental samples by field-flow fractionation. *Trends in Analytical Chemistry*, 30:3, 425-436

Knott, J., Gilbert, J., Hoccom, D.G. & R.E. Green. 2010. Implications for wildlife and humans of dietary exposure to lead from fragments of lead rifle bullets in deer shot in the UK. *Science of the Total Environment* 409 (2010) 95–99

Kollander, B., Andersson, M., Pettersson, J.(2010) Fast multielement screening of non-digested biological materials by slurry introduction to ICP-AES. *Talanta* 80:2068-2075

Lahrssen-Wiederholt 2014. "Lebensmittelsicherheit von jagdlich gewonnenem Wildbret".
Powerpointpresentation.

<http://www.bfr.bund.de/cm/343/lebensmittelsicherheit-von-jagdlich-gewonnenem-wildbret.pdf>

Lindboe M, Henrichsen EN, Høgåsen HR, Bernhoft A. Lead concentration in meat from lead-killed moose and predicted human exposure using Monte Carlo simulation. *Food Additives and Contaminants Part A Chem Anal Control Expo Risk Assess.* 2012;29(7):1052-7.

The National Food Agency's report series 18/2008. Älgkött - analys av näringsämnen [Moose meat - analysis of nutrients]. Uppsala: National Food Agency

The National Food Agency's report series 9/2012. Kontroll av rests substanser i levande djur och animaliska livsmedel [Control of residual substances in living animals and animal-based foodstuffs]. Uppsala: National Food Agency

National Food Agency. 2012. Bly i viltkött - en riskhanteringsrapport [Lead in game meat - a risk management report]. Appendix: risk assessment of lead in minced moose meat.

The National Food Agency's report series 24/2013. Kött - analys av näringsämnen [Meat - analysis of nutrients]. Uppsala: National Food Agency

Mateo, R., Baos, A.R., Vidal, D., Camarero, P.R., Martinez-Haro, M., Taggart, M.A., 2010. Bioaccessibility of Pb from ammunition in game meat is affected by cooking treatment. *PLoS One* 6, e15892.

Mateo R, Vallverdú-Coll N, López-Antia A, Taggart MA, Martínez-Haro M, Guitart R, Ortiz-Santaliestra ME. Reducing Pb poisoning in birds and Pb exposure in game meat consumers: the dual benefit of effective Pb shot regulation. *Environ Int.* 2014 Feb;63:163-8.

Meltzer HM, Dahl H, Brantsæter AL, Birgisdottir BE, Knutsen HK, Bernhoft A, Oftedal B, Lande US, Alexander J, Haugen M, Ydersbond TA. 2013. Consumption of lead-shot cervid meat and blood lead concentrations in a group of adult Norwegians. *Environmental Research* 127:29–39.

NFS 2002: 18, Naturvårdsverkets föreskrifter och Allmänna råd om jakt och statens vilt, ISSN 1403-8234, Naturvårdsverket, Sverige, <http://www.naturvardsverket.se/Documents/foreskrifter/nfs2002/NFS2002-18.pdf>

NMKL 186. 2007. Trace elements - As, Cd, Hg, Pb and other elements. Determination by ICP-MS after pressure digestion.

Oskarsson. A., Jorhem, L., Sundberg, J., Nilsson, N-G., Albanus, L., Lead poisoning in cattle – transfer of lead to milk. *The Science of the Total Environment*. (1992) 111: 83-94.

Pain, D.J, Cromie, R.L., Newth, J., Brown, M.J., Crutcher, E., Hardman, P.,

Hurst, L., Mateo, R., Meharg, A.A., Moran, A.C., Raab, A., Taggart, M.A. &

R.E. Green. 2010. Potential Hazard to Human Health from Exposure to Fragments of Lead Bullets and Shot in the Tissues of Game Animals. *Plos ONE* 4: 1-17.

Riksmaten - vuxna 2010-11 Livsmedels- och näringsintag bland vuxna i Sverige, [National Diet - adults 2010-11 Dietary habits and nutrient intake in Sweden] © Livesmedelverket [National Food Agency], Uppsala September 2012 ISBN 978 91 7714 216 4

Stokke. S. et al. Blyrester från kulammunition i viltkött – en hälsorisk? *Svensk Veterinärtidning* (2010) 15: 21-24

Treble, R.G., Thompson, T.S. Elevated Blood Lead Levels Resulting from the Ingestion of Air Rifle Pellets. *Journal of Analytical Toxicology*, Vol. 26, September 2002

Tsuji LJS, Wainman BC, Martin ID, Sutherland C, Weber J-P, Dumas, P. & E. Nieboer. 2008a. The identification of lead ammunition as a source of lead exposure in First Nations: the use of lead isotope ratios. *Science of the Total Environment* 393: 291–298. DOI:10.1016/j.scitotenv.2008.01.022.

Tsuji LJS, Wainman BC, Martin ID, Sutherland C, Weber J-P, Dumas, P. & E. Nieboer. 2008b. Lead shot contribution to blood lead of First Nations people: the use of lead isotopes to identify the source of exposure. *Science of the Total Environment* 405: 180–185. DOI:10.1016/j.scitotenv.2008.06.048.

Wiklund & Malmfors. 2014. *Viltkött*. Rapport i Naturvårdsverkets rapportserie.

VKM 2013. Risk assessment of lead exposure from cervid

meat in Norwegian consumers and in hunting

dogs. Vitenskapskommittén for mattrygghet ISBN: 978-82-8259-096-9.

Appendices

Appendix 1. Description of ammunition types

Information available at the Swedish Association for Hunting and Wildlife Management's web site describing various types of ammunition.

<http://jagareforbundet.se/jakten/kotthantering/>

Hunting ammunition can be divided up into bullets, where each cartridge contains just one projectile, and shot, where each shell contains several projectiles. A special case is 'slugs' or Brenneke bullets, where shells contain just one large projectile. These are relatively rare and have therefore not been investigated in this study.

Ever since handguns first were invented, lead has been used for bullets and shot. This is because lead is both malleable and of high density, that is to say, it has a high weight per volume. Both of these qualities are important in this context. Projectiles of a given size made from a material with high density allow a higher impact energy than projectiles of a lighter material. Bullets and shot manufactured from a soft metal also deform to fit the bore of the barrel better, which means that less of the gases that propel the projectiles "escape". At the same time, a lower pressure is required to fire a soft projectile through a barrel than a hard projectile. Consequently, the weapon can function with a lower working pressure than would otherwise be the case. Furthermore, soft projectiles normally give off more of their energy when they hit the target, since they are deformed rather than just penetrating the target. This guarantees a high 'killing efficacy', which means that the animal dies quickly. Consequently, lead has a number of positive ballistic qualities.

However, lead is poisonous and severe poisoning can result in many different symptoms, such as tiredness, constipation and loss of appetite. Lead also damages the red blood cells and this can lead to anaemia. A more serious case of poisoning can also mean the loss of nerve functions in the extremities (e.g., the arms), which can lead to partial paralysis. Thus, lead is poisonous and even low levels of exposure are thought to cause damage to the nervous system. This is especially the case when the brain is developing in the foetus or in small children. In epidemiological studies of children it has been estimated that blood lead levels of around 12 µg/litre can have negative effects in the form of a lower IQ. These effects are those that are thought to occur at the lowest levels of exposure. It is also well established that lead from ammunition can have an impact on the environment (for example Axelsson 2009, Helander et al., 2012, Mateo et al., 2014). In recent years, the presence of residues of lead from ammunition have been noted in game meat along with raised levels of lead in the blood of consumers of game meat (Bjermo 2013, Meltzer 2013).

Ammunition manufacturers have consequently produced alternatives to lead, both for bullets and gunshot. The alternative materials have a number of limitations compared with lead, and hunters have to balance the environmental advantages against the ballistic disadvantages. But there are also compromises to be made when choosing between the various lead bullets available; there is no one bullet that is suitable in all situations.

Bullets

The vast majority of bullets used in hunting today have a lead core. This is enclosed within a jacket which is made of a brass alloy containing a high degree of copper. Larger game, such as moose, deer, bison, European bison, muskox, moufflon sheep, wolves, bear, seal, wild boar, roe deer, wolverine, lynx and beaver may only be hunted in Sweden with bullets that are manufactured to expand on impact (roe deer and lynx may however also be hunted using shot) (NFS 2002: 18). When a bullet expands, more impact energy is transferred to the target, which means the shot has a greater effect, leading to a quicker death. Solid lead bullets with no jacket may be used for the same game, provided that they are constructed to expand. The use of solid lead bullets is however very unusual for the hunting of larger game in Sweden today. On the other hand, lead bullets with no jacket are sometimes used for smaller game, such as ptarmigan. In terms of weight, game that can be shot with expanding bullets only accounts for over 90 per cent of the game meat obtained from hunting in Sweden every year (Wiklund & Malmfors 2014).

The majority of expanding bullets with a lead core have a jacket that is enclosed at the back but open at the front. Sometimes the lead core is exposed at the front of the bullet, sometimes the tip is instead equipped with a plastic tip that retains its form better than lead, gives the bullet a more aerodynamic shape and allows the expansion to start faster. Bullets with a lead core that are not made to expand have a jacket that is enclosed at the front, but open at the back. Since there is no opening in the visible part of the bullet that sticks out of the cartridge, these are known as 'full metal jackets'. This type of ammunition normally leaves little or no lead residue in game meat, since the bullet goes straight through the animal without expanding and fragmenting.

The speed of bullet expansion depends on the bullet construction, impact velocity and the amount of resistance the bullet encounters. When hunting larger game it is important that enough of the bullet's original weight is retained through its expansion, in order to guarantee that the bullet reaches and damages the vital organs. Hunters often strive for the bullet to go straight through the animals' body; this increases the bleeding, has a greater shot effect and makes it easier to find the prey by following the trail of blood, if the animal does not fall on the spot where it was hit. At the same time, it is not desirable for the bullet to retain too much of its energy after having penetrated the body, since this energy will not contribute to the shot effect. Consequently, the bullet should not expand and fragment more than necessary to ensure that it reaches and damages the vital organs. At the same time, it should not fragment less than is necessary to produce a satisfactory effect.

Several different methods are employed in the construction of bullets to control the expansion. The lead in the bullet's core is normally alloyed with some percentage of antimony, to make the core harder and limit fragmentation and expansion. At the same time the jacket is normally constructed so that it limits the expansion. The thickness of the jacket is of great significance with regard to how quickly and how much the bullet expands. Some bullets have mechanical "lead locks" to ensure that a part of the bullet stays intact. One extreme example is to have two separate lead cores, where one is completely enclosed in the jacket and will remain intact. The other is allowed to expand and fragment, which guarantees a good shot effect whilst the rear part of the bullet remains intact and ensures penetration. A simpler variant is to manufacture a fold in the jacket that goes into

the lead core. The front part of the bullet will then expand, but not farther back than the fold in the jacket.

The most effective way of limiting expansion and fragmentation is currently to adapt the thickness of the jacket and simultaneously to chemically fuse the lead core together with the jacket so that they stay connected during the expansion. Since the jacket and core are bonded together, these bullets are known as "bonded" bullets. Expanding bullets with lead cores are therefore currently divided up into conventional "non-bonded" bullets and "bonded" bullets. Bonded bullets hold together better and consequently do not lose as much lead when hitting the target. Such bullets often retain over 90 per cent of their original weight upon expansion, whilst conventional bullets typically lose 10-40 per cent of their original weight in the form of fragments. There is however great variation within the groups of conventional and bonded bullets, and there are "hard" conventional bullets that have a higher residual weight than "soft" bonded bullets.

The majority of lead fragments from an expanding bullet are small, soft and difficult to see when cleaning or slaughtering the animal. This is especially true for bonded bullets, where there are normally many small fragments instead of a mix of large and small. As shown in recent reports, it is important to clean thoroughly around the wound channel in order to minimise the risk of lead residues in the meat.

Lead-free bullets also exist; these are usually made of homogeneous copper or brass with a hollow tip. When the bullet hits the prey, it expands as the tip is turned inside out, just like a soft-point lead bullet. The majority of copper bullets are manufactured so that they stay in one piece upon expansion. Accordingly, there are no fragments that cause damage outside of the wound channel, in addition to the effect from the shock wave of the bullet. The copper alloys are considerably harder than lead, which in addition means that the bullet often does not expand to the same diameter as an equivalent bullet with a lead core. All in all, this means that the shot effect from expanding copper bullets can be somewhat less than for expanding lead bullets. As the bullet is constructed to stay in one piece its weight is retained however, which ensures certain penetration to the vital organs and copper bullets usually yield a through and through shot. In recent years copper bullets have been developed that are made to partially fragment, exactly like bullets with a lead core. A new, major study in Germany shows that there is no difference between lead bullets and lead-free bullets in terms of the estimated escape distance of prey after being hit. Furthermore, there were no differences in how satisfied the hunters were with the bullet's function (Gremse & Rieger 2014).

Figure 1 shows the bullets used in the investigation, which were shot into a plastic crate filled with soaked telephone directories in order to simulate the effect of impact on prey. The diameter of the bullets fired did not differ noticeably, whilst the residual weights did. The copper bullets Lapua Naturalis and Nosler E-tip had the highest residual weights, followed by Norma Oryx, which is a bonded bullet with a lead core. The non-bonded bullets with a lead core had the lowest residual weight, and Lapua Mega had a higher residual weight than Norma Silverblix which is no longer sold in Sweden. Bullets with a higher residual weight will penetrate deeper, but at the same time the bullets that lose a great deal of weight will deposit a lot of energy and lead fragments to the carcass. The shot effect is trade-off between penetration and expansion, but will be greater for bullets with great expansion as long as they penetrate sufficiently.

In a survey of Fennoscandian moose hunters regarding their choice of bullet, 2.4 per cent of the Swedish hunters stated that they used copper bullets, compared with 4 per cent in Norway and 18 per cent in Finland (Stokke et al., 2010).



Figure 1. The various bullets that were used in the investigation were shot from a .308 calibre Winchester into a plastic crate filled with soaked telephone directories in order to simulate the impact on the prey. From the left, Nosler E-tip (homogeneous copper bullet; residual weight 99.2 per cent), Lapua Naturalis (homogeneous copper bullet; residual weight 99.6 per cent), Norma Oryx (bonded bullet with lead core; residual weight 94.1 per cent), Lapua Mega (non-bonded bullet with lead core; residual weight 89.0 per cent) and Norma Silverblix (older type of non-bonded bullet with lead core; residual weight 60.7 per cent). Photograph: F Widemo, SJF

Lead shot

Unlike rifle cartridges, shotgun shells contain many small projectiles. But like bullets, lead shot require great impact energy to be effective. Just as with bullets, lead is a very suitable material for the manufacture of shot from a ballistic point of view. Lead shot are fired at lower velocities and have no jackets, but can be plated for example with nickel.

Shot guns are adapted for hunting game that is moving, for example flying fowl, and for short shooting distances (normally less than 30 metres). The dispersal of the pellets increases with the distance from the weapon. This increases the chances of bringing down a moving target. At the same time, the density of the charge of shot is lower and fewer pellets will hit a quarry of a given size. Due to the low weight in each projectile, the impact energy for each pellet is low, and an effective shot effect is ensured by a number of shot hitting the prey's vital organs.

A charge of shot that hits its prey creates many small wound channels, unlike bullets, which create one large wound cavity. Lead shot that hits a bone or other hard material fragment just like expanding bullets. Previously, hunting was conducted exclusively with shot made of lead, with a small amount of antimony and tin added to make the shot suitably hard. As early as the late 1800s, it was noted that ducks were becoming poisoned with lead as they picked up and ate lead shot from the bottom of shallow wetlands and lakes (for example Mateo et al., 2014). Today, there are international agreements to ban hunting with lead shot in wetlands (AEWA 2009) and Denmark has banned lead shot completely. As a result, a market has emerged for shot manufactured from materials other than lead.

By far the most common alternative to lead is steel shot. This is lighter than lead shot of the equivalent size, and harder. Steel shot of a given size therefore have a lower impact energy, does not deform as well as lead, requires higher pressure in the weapon and entails a greater risk of ricochets against hard objects. The reduced efficacy can however be mitigated by the selection of larger and therefore heavier shot, and by shooting from a shorter distance. Steel shot shells cost roughly the same as lead shot shells. There are also alternatives made from bismuth and tungsten. These metals are considerably heavier than steel and also considerably more expensive. Shells with shot made from these materials can cost up to ten times as much as lead and steel shells, which means that very few Swedish hunters currently use them. This might change if there was to be a total ban on lead in shotgun ammunition. Today, Swedish hunters primarily use steel shot in wetlands and lead shot over dry land.

Appendix 2. Lead levels in minced moose meat.

Analysis conducted using ICP-MS, detection limit 0.02 mg lead/kg fresh weight.

Species/	Id	Weight,	Lead content, mg/kg
Minced moose	1e	532	3.40
Minced moose	2e	233	<0.02
Minced moose	3e	735	0.90
Minced moose	4e	498	0.11
Minced moose	5e	178	<0.02
Minced moose	6e	241	0.18
Minced moose	7e	1000	0.05
Minced moose	8e	478	< 0.02
Minced moose	9e	558	0.30
Minced moose	10e	534	0.17
Minced moose	11e	391	<0.02
Minced moose	12e	554	< 0.02
Minced moose	13e	269	<0.02
Minced moose	14e	651	< 0.02
Minced moose	15e	366	< 0.02
Minced moose	16e	626	0.70
Minced moose	17e	463	< 0.02
Minced moose	18e	444	0.11
Minced moose	19e	420	0.09
Minced moose	20e	480	0.82
Minced moose	21e	241	0.02
Minced moose	22e	422	< 0.02
Minced moose	23e	501	< 0.02
Minced moose	24e	468	2.54
Minced moose	25e	250	31.00
Minced moose	26e	466	1.45
Minced moose	27e	432	0.10
Minced moose	28e	376	< 0.02
Minced moose	29e	389	< 0.02
Minced moose	30e	313	< 0.02
Minced moose	31e	235	< 0.02
Minced moose	32e	249	0.07
Minced moose	33e	473	< 0.02
Minced moose	34e	199	2.54
Minced moose	35e	261	0.06
Minced moose	36e	460	0.03
Minced moose	37e	119	3.40
Minced moose	38e	78	0.04
Minced moose	39e	420	<0,02
Minced moose	40e	386	0.05
Minced moose	41e	291	0.14
Minced moose	42e	508	0.05

Species/	Id	Weight,	Lead content, mg/kg
Minced moose	43e	244	<0.02
Minced moose	44e	651	<0.02
Minced moose	45e	509	<0.02
Minced moose	46e	367	0.88
Minced moose	47e	571	<0.02
Minced moose	48e	499	<0.02
Minced moose	49e	371	0.40
Minced moose	50e	200	0.06
Minced moose	51e	583	<0.02
Minced moose	52e	358	<0.02
Minced moose	53e	196	0.02
Minced moose	54e	202	<0.02

Appendix 3. Lead levels in wild boar shoulders from various individuals and shot locations.

Analysis conducted using ICP-MS, detection limit 0.004 mg lead/kg fresh weight.

Species	Individual	Sample type	Shot location	Lead content (mg/kg)
Wild boar	V1	Wound channel	Humerus	352
Wild boar	V1	0-5 cm	Humerus	24.6
Wild boar	V1	5-10 cm	Humerus	1.07
Wild boar	V1	10-15 cm	Humerus	28.5
Wild boar	V2	Wound channel	Humerus	356
Wild boar	V2	0-5 cm	Humerus	1.50
Wild boar	V2	5-10 cm	Humerus	0.028
Wild boar	V2	10-15 cm	Humerus	0.067
Wild boar	V3	Wound channel	Soft tissue	69.1
Wild boar	V3	0-5 cm	Soft tissue	0.034
Wild boar	V3	5-10 cm	Soft tissue	0.050
Wild boar	V3	10-15 cm	Soft tissue	0.038
Wild boar	V4	Wound channel	Soft tissue	71.8
Wild boar	V4	0-5 cm	Soft tissue	9.41
Wild boar	V4	5-10 cm	Soft tissue	0.080
Wild boar	V4	10-15 cm	Soft tissue	0.017
Wild boar	V5	Wound channel	Humerus	131
Wild boar	V5	0-5 cm	Humerus	103
Wild boar	V5	5-10 cm	Humerus	0.153
Wild boar	V5	10-15 cm	Humerus	0.013
Wild boar	V6	Wound channel	Humerus	880
Wild boar	V6	0-5 cm	Humerus	202
Wild boar	V6	5-10 cm	Humerus	18.0
Wild boar	V6	10-15 cm	Humerus	0.450
Wild boar	V7	Wound channel	Humerus	1829
Wild boar	V7	0-5 cm	Humerus	9.71
Wild boar	V7	5-10 cm	Humerus	0.207
Wild boar	V7	10-15 cm	Humerus	0.049

Species	Individual	Sample type	Shot location	Lead content (mg/kg)
Wild boar	V8	Wound channel	Soft tissue	92.0
Wild boar	V8	0-5 cm	Soft tissue	0.821
Wild boar	V8	5-10 cm	Soft tissue	0.022
Wild boar	V8	10-15 cm	Soft tissue	0.011
Wild boar	V9	Wound channel	Shoulder blade	26.7
Wild boar	V9	0-5 cm	Shoulder blade	1.38
Wild boar	V9	5-10 cm	Shoulder blade	0.126
Wild boar	V9	10-15 cm	Shoulder blade	0.013
Wild boar	V10	Wound channel	Shoulder blade	160
Wild boar	V10	0-5 cm	Shoulder blade	1.59
Wild boar	V10	5-10 cm	Shoulder blade	0.063
Wild boar	V10	10-15 cm	Shoulder blade	0.014
Wild boar	V11	Wound channel	Humerus	59.0
Wild boar	V11	0-5 cm	Humerus	37.0
Wild boar	V11	5-10 cm	Humerus	0.206
Wild boar	V11	10-15 cm	Humerus	0.004
Wild boar	V12	Wound channel	Soft tissue	0.004
Wild boar	V12	0-5 cm	Soft tissue	0.011
Wild boar	V12	5-10 cm	Soft tissue	0.007
Wild boar	V13	Wound channel	Humerus	242
Wild boar	V13	0-5 cm	Humerus	18.6
Wild boar	V13	5-10 cm	Humerus	0.251
Wild boar	V13	10-15 cm	Humerus	0.036
Wild boar	V14	Wound channel	Shoulder blade	45.3
Wild boar	V14	0-5 cm	Shoulder blade	5.09
Wild boar	V14	5-10 cm	Shoulder blade	0.006
Wild boar	V14	10-15 cm	Shoulder blade	0.009
Wild boar	V15	Wound channel	Soft tissue	44.8
Wild boar	V15	0-5 cm	Soft tissue	0.166
Wild boar	V15	5-10 cm	Soft tissue	1.78
Wild boar	V15	10-15 cm	Soft tissue	1.58
Wild boar	V16	Wound channel	Humerus	552

Species	Individual	Sample type	Shot location	Lead content (mg/kg)
Wild boar	V16	0-5 cm	Humerus	1466
Wild boar	V16	5-10 cm	Humerus	0.523
Wild boar	V16	10-15 cm	Humerus	7.37
Wild boar	V17	Wound channel	Humerus	250
Wild boar	V17	0-5 cm	Humerus	52.4
Wild boar	V17	5-10 cm	Humerus	0.024
Wild boar	V18	Wound channel	Humerus	895
Wild boar	V18	0-5 cm	Humerus	9.18
Wild boar	V18	5-10 cm	Humerus	0.089

Appendix 4. Lead levels in various cuts of meat taken from fallow deer fawns and roe deer.

Analysis conducted using ICP-MS, detection limit 0.004 mg lead/kg fresh weight.

Species	Animal ID	Bullet	Sample type	Shot location	Lead content (mg/kg)	Remarks
Fallow deer calf	A1	Bonded	shoulder	Shoulder blade	0.077	
Fallow deer calf	A1	Bonded	trimmed cut	Shoulder blade	252	
Fallow deer calf	A1	Bonded	loin	Shoulder blade	2.63	
Fallow deer calf	A2	Bonded	shoulder	Soft tissue	1.76	
Fallow deer calf	A2	Bonded	trimmed cut	Soft tissue	33.6	
Fallow deer calf	A2	Bonded	loin	Soft tissue	0.004	
Fallow deer calf	A3	Conventional	shoulder	Humerus	0.126	
Fallow deer calf	A3	Conventional	trimmed cut	Humerus	148	
Fallow deer calf	A3	Conventional	loin	Humerus	0.021	
Fallow deer calf	A4	Bonded	shoulder	Soft tissue	0.059	
Fallow deer calf	A4	Bonded	trimmed cut	Soft tissue	13.3	
Fallow deer calf	A4	Bonded	loin	Soft tissue	0.019	
Fallow deer calf	A5	Bonded	shoulder	Soft tissue	1.51	
Fallow deer calf	A5	Bonded	trimmed cut	Soft tissue	439	
Fallow deer calf	A5	Bonded	loin	Soft tissue	0.590	

Species	Animal ID	Bullet	Sample type	Shot location	Lead content (mg/kg)	Remarks
Fallow deer calf	A6	Bonded	shoulder	Shoulder blade	5.28	
Fallow deer calf	A6	Bonded	trimmed cut	Shoulder blade	147	
Fallow deer calf	A6	Bonded	loin	Shoulder blade	0.149	
Fallow deer calf	A7	Bonded	shoulder	Backbone	0.007	Shot upper side of backbone
Fallow deer calf	A7	Bonded	trimmed cut	Backbone	233	Shot upper side of backbone
Fallow deer calf	A7	Bonded	loin	Backbone	0.119	Shot upper side of backbone
Fallow deer calf	A8	Bonded	Shoulder	Backbone	0.005	Shot upper side of backbone
Fallow deer calf	A8	Bonded	Trimmed cut	Backbone	85.7	Shot upper side of backbone
Fallow deer calf	A8	Bonded	Loin	Backbone	0.942	Shot upper side of backbone
Fallow deer calf	F1	Conventional	shoulder	Shoulder blade	0.019	
Fallow deer calf	F1	Conventional	trimmed cut	Shoulder blade	60.1	
Fallow deer calf	F1	Conventional	loin	Shoulder blade	0.047	
Fallow deer calf	F2	Bonded	shoulder	Soft tissue	0.018	
Fallow deer calf	F2	Bonded	trimmed cut	Soft tissue	10.4	
Fallow deer calf	F2	Bonded	loin	Soft tissue	<0.004	
Fallow deer calf	F3	Conventional	Shoulder	Humerus	235	
Fallow deer calf	F3	Conventional	Trimmed cut	Humerus	233	

Species	Animal ID	Bullet	Sample type	Shot location	Lead content (mg/kg)	Remarks
Fallow deer calf	F3	Conventional	Loin	Humerus	<0.004	
Roe deer	R1	Bonded	Trimmed cut	Humerus	266	
Roe deer	R1	Bonded	Loin	Humerus	<0.004	
Roe deer	R10	Bonded	Shoulder	Soft tissue	<0.004	
Roe deer	R10	Bonded	Trimmed cut	Soft tissue	171	
Roe deer	R10	Bonded	Loin	Soft tissue	<0.004	
Roe deer	R2	Bonded	Shoulder	Soft tissue	1.79	
Roe deer	R2	Bonded	Tenderloin	Soft tissue	<0.004	
Roe deer	R2	Bonded	Trimmed cut	Soft tissue	78.1	
Roe deer	R2	Bonded	Loin	Soft tissue	3.41	
Roe deer	R2	Bonded	Saddle	Soft tissue	<0.004	
Roe deer	R2	Bonded	Haunch	Soft tissue	<0.004	
Roe deer	R3	Copper	Shoulder	Soft tissue	<0.004	
Roe deer	R3	Copper	Tenderloin	Soft tissue	0.022	
Roe deer	R3	Copper	Trimmed cut	Soft tissue	<0.004	
Roe deer	R3	Copper	Loin	Soft tissue	<0.004	
Roe deer	R3	Copper	Saddle	Soft tissue	<0.004	
Roe deer	R3	Copper	Haunch	Soft tissue	<0.004	
Roe deer	R4	Conventional	Shoulder	Humerus	0.037	
Roe deer	R4	Conventional	Tenderloin	Humerus	<0.004	
Roe deer	R4	Conventional	Trimmed cut	Humerus	95.3	
Roe deer	R4	Conventional	Loin	Humerus	0.014	
Roe deer	R4	Conventional	Saddle	Humerus	<0.004	
Roe deer	R4	Conventional	Haunch	Humerus	<0.004	
Roe deer	R5	Bonded	Shoulder	Soft tissue	<0.004	
Roe deer	R5	Bonded	Tenderloin	Soft tissue	0.100	
Roe deer	R5	Bonded	Loin	Soft tissue	<0.004	
Roe deer	R6	Bonded	Shoulder	Humerus	0.039	
Roe deer	R6	Bonded	Tenderloin	Humerus	0.014	
Roe deer	R6	Bonded	Trimmed cut	Humerus	83.2	
Roe deer	R6	Bonded	Loin	Humerus	<0.004	
Roe deer	R6	Bonded	Saddle	Humerus	0.009	

Species	Animal ID	Bullet	Sample type	Shot location	Lead content (mg/kg)	Remarks
Roe deer	R6	Bonded	Haunch	Humerus	<0.004	
Roe deer	R8	Bonded	Shoulder	Soft tissue	207	
Roe deer	R8	Bonded	Trimmed cut	Soft tissue	10.2	
Roe deer	R8	Bonded	Loin	Soft tissue	0.004	
Roe deer	R9	Bonded	Trimmed cut	Soft tissue	166	

This report is available in both Swedish and English.

During 2012-2014 the National Food Agency investigated lead levels in game meat taken with lead ammunition and whether the consumption of game meat could be a risk for the consumers. The work is described in the report *Lead in game meat* in four parts. Part 1, Ammunition residues and chemical analysis, investigates how the occurrence of lead residues from ammunition and lead contents vary between various cuts of game meat, depending on the choice of ammunition and the placement of the shot itself. This report also studies how lead residues dissolve in gastric environments. Part 2, Lead contents in the blood of hunter families, investigates whether the content of lead in the blood affects consumers of game meat. Parts 1 and 2 provide data for the risk assessment of consumption of game meat shot with lead ammunition that is presented in Part 3, Risk assessment. The latter describes the risks entailed by residues of lead ammunition in game meat. Based on this assessment, a health-based critical level for lead fragments in game meat has then been established.

The information contained in these three scientific sub-reports and in other academic literature has then been evaluated in order to assess whether, and if so which, measures could and should be used to reduce the risks associated with the occurrence of lead in game meat. Other relevant factors have also been considered within these assessments, such as whether it is possible for consumers to follow specific advice regarding the consumption of game meat that has been shot with lead ammunition; how advice such as this would be perceived; how it could be applied by the target groups; which supervisory authorities exist for this purpose; and whether the consequences of such a measure are proportionate in relation to the risks and benefits. Part 4, Risk management, describes the different considerations and assessments that led to the measures that NFA deems are necessary for the handling of occurrences of lead residues in game meat, and in order to minimize the risks that consumption of such meat can entail. The report aims to clearly describe the reasoning behind the measures that NFA has determined.

This report might be of interest for those with a general interest in science, hunting and cooking game, especially hunters, analytical chemists, toxicologists and risk managers.

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