Blood Lead Levels of Common Ravens With Access to Big-Game Offal

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ABSTRACT Despite increased knowledge about environmental toxins and changes in lead use (i.e., the mandated use of nonlead paint, gasoline, and shotgun pellets used for hunting waterfowl on federal lands), lead poisoning continues to occur in terrestrial birds. The degree of exposure and its demographic effect, however, continue to be described, emphasizing the growing concern over lead exposure. We examined 302 blood samples from common ravens (*Corvus corax*) scavenging on hunter-killed large ungulates and their offal piles to determine if lead rifle-bullet residuum was a point source for lead ingestion in ravens. We took blood samples during a 15-month period spanning 2 hunting seasons. Of the ravens tested during the hunting season, 47% exhibited elevated blood lead levels ($\geq 10 \mu g/dL$) whereas 2% tested during the nonhunting season exhibited elevated levels. Females had significantly higher blood lead levels than did males. Our results confirm that ravens are ingesting lead during the hunting season and are likely exposed to lead from rifle-shot big-game offal piles. (JOURNAL OF WILDLIFE MANAGEMENT 72(1):240–245; 2008)

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Lead poisoning in birds has been documented in many species other than waterfowl. Fisher et al. (2006) list 59 terrestrial bird species worldwide that have been exposed to lead from ammunition sources, including raptors, galliforms, gruiforms, columbiforms, and gulls. In the United States, Kendall et al. (1996) found that upland game birds ingest substantial amounts of lead shotgun pellets and deduced that raptors must incur secondary ingestion of pellets because their prey ingested it. However, lead shot is not the only source of lead originating from firearms. Within recent years, researchers suspected rifle bullets as a point source for lead contamination through ingestion and most reported cases have been with raptors (Wiemeyer et al. 1988, Snyder and Snyder 1989, Wayland and Bollinger 1999, Garcia-Fernandez et al. 2005, Fisher et al. 2006).

Elevated lead levels have been documented in many species of raptors and vultures, particularly California condors (*Gymnogyps californianus*), bald eagles (*Haliaeetus leucocephalus*), and golden eagles (*Aquila chrysaetos*). The primary source for eagles was believed to be lead shot from waterfowl, but lead occurrence in these species did not significantly drop after the ban of lead shot on federal lands (Kramer and Redig 1997, Wayland and Bollinger 1999). Further, researchers have not documented differences between areas of high waterfowl occurrence and hunting and areas with low waterfowl occurrence, suggesting another lead source (Miller et al. 1998, Wayland et al. 2003).

Recently, Hunt et al. (2005) found that fragmented rifle bullets in hunter-discarded offal piles of harvested deer (*Odocoileus* spp.) and whole unrecovered deer pose a threat to California condors. Through radiographs, Hunt et al. (2005) found that offal piles contained hundreds of small lead particles that may be ingested by scavenging species. Further, Pauli and Buskirk (2007) and Knopper et al. (2006) found that rifle-shot prairie dogs and ground squirrels (respectively) may contain fragmented lead particles that could be ingested by scavengers or raptors. Recently, Church et al. (2006) linked isotopically labeled lead in California condors with rifle bullets sold in the same region, substantiating that condors were ingesting lead and dying from bullet fragments.

Many scavengers use offal piles in the Greater Yellowstone ecosystem (GYE), where a large-scale elk hunt occurs annually (Wilmers et al. 2003), and the most common hunting practice is to field-dress game, but some hunters remove the meat from the bones in the field and leave both the offal and skeletal frame (D. Craighead, Craighead Beringia South, personal observation). In a 1,530-km² study area in the northern GYE, Wilmers et al. (2003) estimated that >35,000 kg of offal (22.9 kg/km²) is available to scavengers annually. Wilmers et al. (2003) also quantitatively estimated that during the hunting season common ravens (Corvus corax) consumed roughly 25,000 kg of offal per year within that study area (16.3 kg/km²; including cached food), and that offal is the main diet of the raven during this time. Hunt et al. (2005) found a mean of 160 lead fragments in offal piles of rifle-shot deer; because elk hides and bones are thicker than deer, more bullet fragmentation may take place in elk. These studies imply that there is a large amount of lead deposited in the raven's main food source during autumn and early winter.

We tested common raven blood lead levels over 15 months during and after the elk-hunting seasons in Jackson Hole, Wyoming, USA. We hypothesized that there was a change in raven blood lead levels that correlated with the big-game hunting season.

STUDY AREA

Jackson Hole, Wyoming (43°91'N, 110°40'W; elevation of the valley floor approx. 2,300 m), located in the southern extreme of the GYE, was home to one of the most dense

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breeding populations of common ravens in the world (Dunk et al. 1997, Bedrosian 2005). Although there were no data on raven nesting densities surrounding our study area, yearround foraging areas of breeding pairs were as much as 1,000 times larger than nesting territories (approx. 2.5 km²; B. Bedrosian, Craighead Beringia South, unpublished data), suggesting that many ravens outside of our study area can become locally concentrated within our study area based on food availability. The GYE landscape was composed of 2 National Parks (Grand Teton and Yellowstone National Parks), the National Elk Refuge, 3 designated wilderness areas, and 4 national forests. Because of this protection, shooting of wildlife rarely occurs outside the hunting season; however, we acknowledge that some recreational killing and predator control may occur on National Forest and private lands throughout the year. An annual elk hunt occurs everywhere within our study area except for the western half of Grand Teton National Park (from the Snake River to the western edge of the park; 7% of the study area). Protection of wildlife temporally controlled most exposure to lead ingestion by scavengers and allowed us to compare 2 periods, hunting and nonhunting.

METHODS

We defined our study area as the outer perimeter of the 11 designated hunt zones (Wyoming Game and Fish Department; 2004 elk hunt zones 73-82, 84, 85), which encompassed the winter foraging ranges of ravens we followed during the sampling period (approx. 2,700 km²; B. Bedrosian, unpublished data). Using the annual harvest reports, 2001–2004 (Wyoming Game and Fish Department 2001, 2002, 2003, 2004) we tallied annual elk harvest estimates within our study area. The 2004 elk-hunting season ended on 5 December, and the 2005 hunting season began on 10 September and ended on 4 December. We estimated amount of hunter-left carrion by assuming that each successful hunter left an offal pile in the field. We did not include rumen in estimates of available offal because ravens do not feed on undigested plant materials. We assumed that each offal pile (minus the rumen) weighed 14% of the animal's live weight and multiplied mean weights based on gender and age class by 0.14 to obtain estimates of total offal weight left in the field in 2004 (Jensen 2000, Wilmers et al. 2003). Based on an informal canvass of hunters, we assumed that bullets used for elkhunting were lead-core or solid-lead bullets.

Within our study area, Wyoming Game and Fish Department (2004, 2005) estimated an average of 2,925 (SD = 228) elk harvested annually from the 2004 and 2005 hunting seasons. We estimated an annual 108,674 kg of offal was left in the field during the hunting season (7.14 kg/km²). Based on Hunt et al.'s (2005) estimate of 160 lead fragments per offal pile in deer, we estimated 468,000 visible lead fragments were deposited annually within our study area. We confirmed presence of lead particles in 2 randomly chosen offal piles left by hunters. Offal piles were removed from the field, radiographed by a local veterinarian, and we

counted lead particles in each offal pile using 3 classification categories; fragments <2 mm, fragments between 2 mm and 5 mm, and fragments >5 mm. We only counted unambiguous fragments visible with an unaided eye. One offal pile showed 159 visible fragments (146 fragments <2 mm, 10 fragments between 2 mm and 5 mm, and 3 fragments >5 mm). The other offal pile showed 114 small fragments (<2 mm).

From 14 December 2004 to 30 March 2006, we captured common ravens from 22 randomly selected trap sites throughout the study area. Because of large foraging areas used by ravens, their highly gregarious nature, and information-sharing about food resources, we assumed any raven had an equal chance of visiting each trap site (Marzluff et al. 1996; Boarman and Heinrich 1999; D. Craighead, unpublished data). We used a net launcher (Coda Ent., Mesa, AZ) and drop-in traps baited with roadkilled carrion and offal piles left by hunters (offal piles were not moved; Engle and Young 1989). We only used available carrion and did not add bait to the study area that was suspected of containing lead. We banded each raven with a unique United States Geological Survey band and a plastic, alpha-numeric color band (Forsman et al. 1996), and we collected a 0.4-cubic cm (cc) blood sample from the brachial vein. We placed one 0.2–0.3-cc portion of the blood sample in a lithium heparin Microtainer® blood tube (Becton Dickinson, Franklin Lakes, NJ) for blood lead analysis and we placed the remainder in lysis buffer for later polymerase chain reaction gender analysis (Longmire et al. 1988, Fridolfsson and Ellegren 1999). We aged ravens as hatchyear or after-hatch-year birds based on plumage characteristics (Heinrich 1994, Pyle 1997). We temporarily retained 7 ravens that tested with high lead blood-levels, which we kept in an aviary and fed uncontaminated food, and tested their blood lead levels (BLLs) every 2-3 days to determine a species-specific depuration rate. After ≤ 2 weeks blood lead levels became undistinguishable from baseline levels, suggesting a fairly short half-life in vivo. Because of this rapid depuration rate, we considered any recaptures 2 weeks after initial capture independent BLL samples; we considered birds captured within 2 weeks of the end of the hunting season potentially under the influence of the hunting season and we combined them with hunting-season captures for analysis. As a control group, we took blood samples from one randomly selected nestling raven from each of 19 nests in May and June 2005.

We analyzed blood samples for lead levels (μ g/dL) using a Leadcare[®] portable blood lead analyzer (ESA Biosciences Inc., Chelmsford, MA). We tested each blood sample within 24 hours after collection. We tested bovine controls periodically to confirm accuracy of the tester and to check for contamination. We considered blood samples <10 μ g/dL baseline exposure, and we considered samples with BLLs \geq 10 μ g/dL as birds that had lead exposure within the past 2 weeks. During our testing, Leadcare issued a recall because of a calibration error in the test kits that we were using. Although Leadcare issued a calibration equivalent for

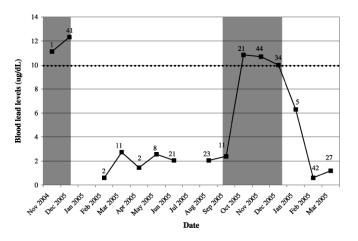


Figure 1. Median monthly blood lead levels (μ g/dL) of common ravens in Jackson Hole, Wyoming, USA, 14 December 2004–30 March 2006. Sample sizes appear above each monthly median. The dotted line indicates the threshold in which we considered birds to have incurred lead exposure within the past 2 weeks and the shaded areas represent the elk-hunting season (the Dec 2005 sample represents birds tested during and after the hunting season combined). We did not collect data in January or July 2005.

human samples, we designed our own calibration equivalent for raven samples using a subset of samples and a simple linear regression, setting the *y*-intercept at 0.0 for the recalled kits and the correctly calibrated kits. We tested samples we used for calibration on both the recalled kit and a correctly calibrated kit using the same reagent. Different techniques used to analyze heavy metal concentrations can result in different BLLs for the same sample but should be directly comparable. Therefore, because some researchers use other methods for determining BLLs, we had a subset of correctly calibrated samples (n = 10) analyzed by graphite furnace atomic absorption spectrometry (GFAAS) at ESA Inc. and we compared the 2 techniques using simple linear regression.

We tested for differences in raven BBLs between the hunting season, nonhunting season, and nestlings using a Kruskal–Wallis test because of nonnormality. We first

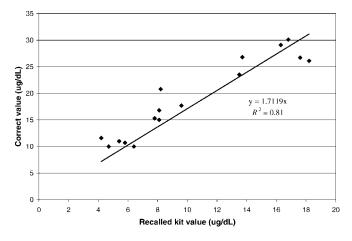


Figure 2. Regression of recalled Leadcare[®] (ESA Biosciences Inc., Chelmsford, MA) blood lead tests and correctly calibrated Leadcare tests based on a random subset of common raven blood samples taken in Wyoming, USA (n = 17).

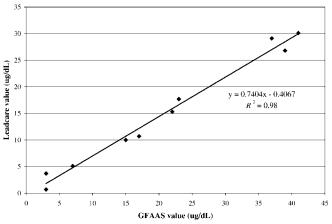


Figure 3. Regression of a Leadcare[®] (ESA Biosciences Inc., Chelmsford, MA) portable lead tester and graphite furnace atomic absorption spectrometry (GFAAS) using a subset of common raven samples from Wyoming, USA (n = 10).

tested for differences in age classes (excluding nestlings) of birds that had lead exposure versus those with no lead exposure during the hunting season using a Mann–Whitney test. If we found no differences, we combined age classes and tested for differences between genders and BLLs using a Mann-Whitney U. Because mass can vary among genders and individuals over time, we determined a body condition index for all captured ravens (Boarman and Heinrich 1999). Similar techniques have been used to create body condition indices for other species using structural measurements and mass (e.g., Dufour et al. 1993, Griebel and Savidge 2003). We used tarsus length as an indicator of structural body size because most measures of structural size are highly correlated for common ravens (Webb et al. 2004). We then used our raven measurement data to create a regression of tarsus length and mass, which indicated an average mass for a bird of a given size within our study area. Then, by using residuals of that regression, each individual can be assessed for condition in relation to the specific sample. We tested this index against BLLs during the hunting season using regression to determine if BLLs could be predicted by body condition. We weighed ravens on an Adam CPWplus-6 digital bench scale (Danbury, CT) and we took tarsus measurements with digital calipers following Pyle (1997).

RESULTS

We obtained 302 blood samples from ravens, including 26 recaptures >2 weeks apart. Of these, we captured 136 during hunting seasons, (43 and 93 in 2004 and 2005, respectively), 147 during nonhunting seasons (before and after hunting combined), and 19 nestlings (Fig. 1). We calibrated recalled test kits with corrected values kits using a simple linear regression without fitting the *y*-intercept (P < 0.01; $R^2 = 0.81$; Fig. 2). We also found that the Leadcare field tester tended to systematically measure lower BLLs when compared to GFAAS (P < 0.01; $R^2 = 0.98$; Fig. 3). The median BLL was 10.7 µg/dL during the hunting

season, 1.8 μ g/dL during the nonhunting season, and 1.2 μ g/dL for nestlings. We found that 47% (n = 64) of the

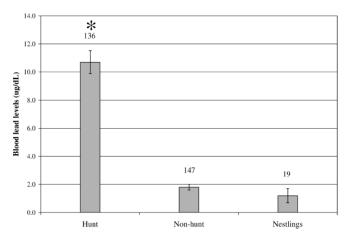


Figure 4. Median blood lead levels ($\tilde{x} \pm SE$) of common ravens in Wyoming, USA during the elk-hunting season, outside of the hunting season, and nestling controls, 14 December 2004–30 March 2006. Samples appear above standard error bars and * denotes a significant difference at $\alpha = 0.01$.

ravens tested during the hunting season exhibited blood lead levels $\geq 10 \ \mu g/dL$, whereas only 2% (n = 3) exhibited elevated blood lead during the nonhunting season. We found no differences between the controls and the samples collected during the nonhunting season whereas the blood lead levels of birds tested during the hunt were significantly higher (P < 0.01; $H_{adj} = 158.94$; Fig. 4). We documented all 3 cases of elevated BLLs during nonhunt from 15 April to 24 April 2006, which corresponds with an increase in ambient temperature that caused some offal piles buried in snowfall to reemerge due to snow melt.

We found no difference between hatch-year (n = 45) and after-hatch-year (n = 89) for BLLs during the hunting season (P = 0.51). After pooling age classes, we did find a difference in BLLs during the hunting season between genders, with females displaying higher median blood lead levels (P = 0.03; 95% CI = 0.2–5.3 µg/dL; Fig. 5). We found no relationship between blood lead levels during the hunting season and body condition index (P = 0.24).

DISCUSSION

We found that common ravens experience elevated blood lead levels during the hunting season in the Jackson Hole valley of Wyoming. Median blood lead levels for ravens during the hunting season (10.2 μ g/dL) were 5.1 times higher than BLLs that are regarded as safe for humans (2 μ g/dL; Menke et al. 2006). We also confirmed that unretrieved offal piles of hunter-killed game were a point source for lead contamination. It is clear that ravens were ingesting more lead during the hunting season than other parts of the year. Given the strong seasonality of lead exposure in ravens, the incidence of lead fragments in offal, and that offal was the primary food source of ravens during time of exposure, it is likely that offal was the source of blood lead in ravens within our study area (Wilmers et al. 2003, Hunt et al. 2005). Redig et al. (1983), Pain et al. (1993), and Redig et al. (1998) discuss advantages and disadvantages of sampling method biases in bird populations

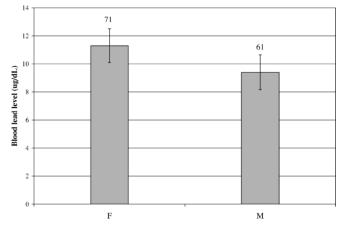


Figure 5. Median blood lead levels (BBLs) of female and male common ravens in Wyoming, USA, during the hunting seasons of 2004 and 2005, and 2005 and 2006. Females exhibited significantly higher BLLs than males (P = 0.02). Sample sizes are given above standard error bars.

exposed to lead and though we acknowledge that trapping may be inherently biased, regardless of biases, 47% of ravens sampled during the hunting season exhibited elevated BLLs.

We found female ravens had higher BLLs than males, a scenario that has been documented in a wild population of western marsh harriers (*Circus aeruginosus*) and with laboratory experiments on mallards (*Anas platyrhynchos*; Sanderson and Bellrose 1986, Pain et al. 1993). Observed disparities in BLLs between sexes may be due to differences in metabolic rates and differential mobilization of energy resources between genders. Also, Burger and Gochfeld (2003) found that lead storage in herring gulls (*Larus argentatus*) can be partly ameliorated with increased levels of exercise, which may suggest male ravens are more active than females. Finally, the difference may simply reflect differential amounts of lead consumed by either gender.

We found 3 ravens that exhibited elevated BLLs (i.e., >10 μ g/dL) during the spring. Although the proportion of the population latently exposed during the spring (2%) is markedly lower than during the hunting season (47%), the mean spring melt dates correspond with the average egg laying dates for ravens in our study area (Dunk et al. 1997). If lead is passed from females to their young during egg production, this scenario may lead to decreased population fitness because of increased susceptibility of developing birds to negative impacts of lead (Burger and Gochfield 2000).

Little or no quantitative data delineate background or exposure levels for blood lead in ravens. The BLLs we observed in nestlings and ravens during the nonhunting period suggest that defining 10 μ g/dL as positive exposure is a reasonable, if not an overestimated, value. A threshold toxic level is difficult to assign because effects on the nervous system can be subtle and difficult to detect without specific measurable behaviors. Although we have no data on the sub-lethal effects of lead exposure on raven behavior or survival, sub-lethal lead exposure has been documented to affect most body systems and result in a variety of mental and physical ailments for various animals (Fisher et al. 2006). Small amounts of lead ingestion causing even a

minor decrease in fitness of a bird surviving in a hostile and competitive environment may result in a proximate death from many causes. For example, sub-lethal lead exposure has been found to delay behavioral response time, increase the risk of collision with overhead power lines, and decrease weight and muscle mass (Burger and Gochfeld 2000, Carpenter et al. 2003, Kelly and Kelly 2005). Futher, lead ingestion has also been documented to cause hemolytic anemia, stunt neurological development, increase blood pressure, lower bone density, and cause paralysis of the neuromuscular system (Pattee et al. 1981, Victery 1988, Staessen et al. 1994, Mateo et al. 2003, Bagchi and Preuss 2005). In long-lived bird species, such as condors, eagles, and ravens, periodic and chronic exposure has the potential to skew the normal age structure toward younger and nonbreeding birds and negatively influence long-term population viability.

MANAGEMENT IMPLICATIONS

Better understanding of the distribution and amount of lead contributed to the environment from hunting is needed globally. We have proposed a likely mechanism to explain the phenomenon of high lead levels in ravens during hunting season and this should help managers to predict when and where to expect this phenomenon in other species. The implications of our study are that lead contamination should be suspect in all species that feed on hunter-killed animals and at spatial and temporal scales other than what we documented. As carcasses and offal decompose, lead can potentially migrate to soil, where it is available for further bioaccumulation in other systems. Fortunately, using nonlead bullets is one relatively easy solution to curb this form of lead contamination.

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