Hair analysis in sled dogs (Canis lupus familiaris) illustrates a linkage of mercury exposure along the Yukon River with human subsistence food systems

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Abstract

The dog has been an important biomedical research model and hair samples from sled dogs could be used as a biomarker of exposure to metals. Hair samples were used as a non-invasive indicator of mercury exposure in sled dogs fed commercial food and traditional village diets. Sled dog populations living in rural New York and Alaska were sampled in 2005 and 2006. Total mercury (THg) content was determined on the entire hair sample in sled dogs from reference sites in North Creek, New York and Salcha Alaska. Both sites fed a commercial feed for high performance dogs and had mean THg levels of 36.6 ng/g for New York sled dogs while Alaskan sled dogs, occasionally supplemented with fish oil showed THg mean of 58.2 ng/g. These THg levels are below levels that are suggested to cause adverse effects and should be considered baseline levels. Yukon River sled dogs had higher THg, ranging from 139 to 15,800 ng/g and showed decreasing mean levels from the delta area to upriver. There were significant differences between THg in the dogs from Russian Mission (10,908.3±3028 ng/g), the farthest west village, and Ft. Yukon (1822.4±1747 ng/g), the farthest east village. All village dogs along the Yukon had higher THg levels than the THg mean level (657±273 ng/g) of hair from ancient dogs of the Seward Peninsula.

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1. Introduction

Climate change and natural resource development in the circumpolar north has the potential to impact the dietary consumption of human subsistence harvesters. This additional stress on ecosystem services is creating a compelling need for the development of bioindicators for monitoring environmental change (Hoekstra et al., 2003; Sobanska et al., 2005; Dehn et al., 2006). The presence of mercury in northern food chains, especially in traditional foods, has been a concern of health scientists because of its toxicity and bioaccumulation (Duffy et al., 2005; Jewett et al., 2003; Braune et al., 1999).

Hair from various mammals has been used as convenient biomarkers of mercury (Hg) pollution, including caribou (Duffy et al., 2005), wild boar (Sobanska, 2005), sea lions (Beckmen et al., 2002), seals (Sun et al., 2006), river otter (Ben-David et al., 2001), opossum (Burger et al.,...
1994), muskrats (Stevens et al., 1997) and dogs (Hansen and Danscher, 1995). Since mammals share many physiological and biochemical characteristics with man, Hg in dogs could impact the same mechanisms that influence human health. The dog has long been an important research model and is considered an omnivore when associated with man. Dogs share the same environment as humans and develop many of the same diseases, especially immunological syndromes (Dunlap et al., 2006; Felsburg, 2002), and therefore can be an ideal animal model for environmental impacts on northern people. In villages in the North, sled dogs are fed basically the same type of food as is consumed by humans (Andersen, 1992; Hansen and Danscher, 1995). These dogs are useful for biomonitoring because of their increased metabolism, shorter lifespan and use of a subsistence diet similar to rural Alaskans. Sled dogs as biomonitorers are especially important as younger Alaskans move away from traditional subsistence activities. Mercury studies on dogs also allow comparisons to past periods of time, since dogs are common in the archaeofauna (Larsen, 2001).

The objectives of this study were: 1) to investigate the usefulness of sled dogs (Canis lupus familiaris) hair as a non-invasive indicator of mercury, 2) survey the transfer of Hg from fish to a mammal population along the Yukon River, and 3) compare Hg levels of modern sled dogs with that of ancient dogs from Alaska. This research also tested the hypothesis that sled dogs fed commercial dog chow would have lower levels of mercury in their hair than sled dogs fed a more human like diet that included fish.

2. Materials and methods

2.1. Hair samples

Sled dog (C. lupus familiaris) hair was collected in Alaska and New York (2005–2006). A bout 0.2 g of front dorsal hair (lower neck) was cut near to the skin (proximal) by means of stainless-steel surgical scissors. Hair samples were washed and homogenized before analyses. The gender and age of the sled dogs were also recorded.

Samples were obtained from several Alaska sites and one New York site: 16 individuals from New York (45°N) and 20 individuals from Salcha, Alaska (65°N) both fed Purina Pro Plan were sampled in the summer of 2005 as reference kennels. Twelve Yukon River dogs were collected in the summer of 2006 from each village: Russian Mission (62°N), Galena (64°N), Rampart (65°N), Fort Yukon (66°N) and again from Salcha, Alaska. Yukon River dogs were maintained on a subsistence diet that consisted primarily of seasonal salmon for at least two months prior to collection. A two-month diet-recall was performed at every village. All kennel owners listed salmon as the primary food source and photo documentation of feed barrels and salmon drying racks was performed. Supplemented foods that were given to the dogs at some time in the past 2 months included black bear, pike, moose, chicken and commercial food, in agreement with the more extensive survey of Andersen (1992).

A total of five samples were analyzed from the native village of Deering, located on the northern Seward Peninsula (Bowers, 2006; Gerlach et al., 2006). These samples were excavated from an archaeological site in 1950, and have been curated ever since at the Danish National Museum, Copenhagen (Larsen, 2001). In October 2006, Bowers studied the artifact collection and borrowed the samples for analysis. The materials consist of hair from dog feces from the anteroom of a Qaleti (men’s ceremonial house), dated to 1290±200 radiocarbon years before present (K-537), (calibrated to circa. AD 780: Larsen, 2001; Gerlach and M ason, 1992). A associated dog bones from the site are described as being “…very similar to the rather large, strongly built Eskimo type of dogs, as known from Alaska through Canada to Greenland” (Larsen, 2001). Human subsistence resources identified with associated faunal remains include seal, caribou, walrus, hare, birds, wolf and fox. From cultural historical perspective, the materials relate to the Ipiutak phase of Arctic prehistory, which in Deering persisted from about AD 600 to 900 (Bowers, 2006; Larsen, 2001).

2.2. Sample preparation and QC method

Hair samples were washed prior to digestion. An organic solvent wash was performed using consecutive 10 mL rinses of solvent. For the first two rinses, the samples were shaken every 10 min for an hour. The final three benzene rinses were performed by shaking the sample for 1 min in 10 mL of benzene followed by the removal of the benzene with a pipette. After the washing was complete, the hair was dried overnight in an oven at ~65 °C and then homogenized.

Total mercury (THg) concentration in the hair samples was measured at Frontier Geosciences (Seattle, WA) using the cold vapor atomic fluorescence spectrometry (CVA F) method (Duffy et al., 2005). The results are reported on a wet weight (w/wt) basis as ng/g (ppb). All analyses have been run with respect to a thorough quality control program using reference material (DOLT3-dog fish liver tissue) as well as spiked samples. The percent recovery of DOLT3 was 102%. The % matrix spike percent recovery averaged 84% for two dogs (82% and 86%).
Table 1

<table>
<thead>
<tr>
<th>Site</th>
<th>N</th>
<th>Mean</th>
<th>STD</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Russian Mission</td>
<td>12</td>
<td>10,908</td>
<td>3028</td>
<td>4420–15,800</td>
</tr>
<tr>
<td>Galena</td>
<td>11</td>
<td>4528</td>
<td>1125</td>
<td>2120–6410</td>
</tr>
<tr>
<td>Rampart</td>
<td>12</td>
<td>2446</td>
<td>1737</td>
<td>491–6010</td>
</tr>
<tr>
<td>Fort Yukon</td>
<td>12</td>
<td>1822</td>
<td>1747</td>
<td>139–5050</td>
</tr>
<tr>
<td>Salcha</td>
<td>11</td>
<td>91</td>
<td>18</td>
<td>63–125</td>
</tr>
<tr>
<td>Salcha (2005)</td>
<td>18</td>
<td>59</td>
<td>9</td>
<td>44–80</td>
</tr>
<tr>
<td>Deering (780)</td>
<td>5</td>
<td>657</td>
<td>273</td>
<td>441–982</td>
</tr>
</tbody>
</table>

Salcha and New York sled dogs were maintained on a commercial feed (Purina Pro Plan Performance®). Samples were collected in 2006 unless indicated otherwise. A ncient dog hair samples from a site on the Seward Peninsula, AK are dated at approximately 780 AD.

2.3. Sample digestion

For total mercury in tissue, approximately 0.1–0.2 g of each dog hair sample was digested with 10 mL of hot refluxing 70% HNO₃: 30% H₂SO₄ for approximately 2 h. The digests were then diluted to a final volume of 40 mL with a solution of 10% (v/v) 0.2 N BrCl. Following complete digestion on a hotplate, the samples were diluted up to 40 mL with methanol (neat). From each KOH methanol digest, an aliquot of 0.5 mL was removed and aliquoted into a 60 mL Teflon distilling vial containing 45 mL of double-distilled water.

2.4. Total Hg analysis

Aliquots of each digest were reduced in pre-purged double-distilled water to Hg⁺ with SnCl₂, and then the Hg⁺ purged onto gold traps as a preconcentration step. The Hg contained on the gold traps was then analyzed by thermal desorption into a cold vapor atomic fluorescence detector (CVAF) using the dual amalgamation technique.

2.5. Statistical analysis

One-way ANOVA was performed to compare the THg between the New York group and the Alaska group. Significance was established with a p value less than 0.05. In the Alaska 2005 Salcha group, two outlying data points were removed from the analysis because they were more than four times the mean (Gamberg and Braune, 1999). Similarly, one sample was also removed from the 2006 group. Samples from the Alaskan 2006 Salcha site and the Yukon River villages were analyzed using SAS statistical software. A nalysis of variance was used to analyze all the data to evaluate the effects of sampling location on THg concentrations in hair. Significant differences between sampling sites was determined using Tukey’s Studentized range test. Differences were considered significant at p ≤ 0.05.

3. Results

The THg levels of the sled dogs fed commercial diets in 2005 are reported in Table 1. The mean concentration of 37 ng/g ± 14 for the New York sled dogs was lower than the THg mean concentration for the Salcha, Alaska sled dogs which was 59 ng/g. The range for the Salcha sled dogs was 45–80 ng/g and the range for the New York sled dogs was 20–59 ng/g. Comparisons of these two commercial fed groups show that the Salcha group had more females (40% vs. 25%). While the Salcha

Fig. 1. Total mercury concentrations in hair in village sled dogs subsisting on local salmon along the Yukon River and mercury concentrations from a reference kennel in Salcha, Alaska in 2006. On the inset map, (A) Russian Mission, (B) Galena, (C) Rampart, (D) Fort Yukon, (E) Salcha, where the reference kennel is located and (F) Deering, where the ancient dog hair was located.
group had a wider range of ages, both groups had similar mean ages (3.6±2.7 vs. 3.9±3.9). There was no correlation of age with THg concentration.

The THg levels in Yukon River sled dogs (Table 1) progressively decreased as samples were collected upriver from the river delta (Fig. 1). The reference dogs in Salcha had significantly lower THg levels compared to all other sampling sites, with the exception of Ft. Yukon (Table 2). Russian Mission dogs had significantly higher THg concentrations than all other sampling sites. For intermediate sites, significant differences were found between Galena and Ft. Yukon (Table 2). A single sled dog had lower levels than the Yukon River dogs but higher than the reference dogs.

4. Discussion

Average mercury concentrations (Table 1; Fig. 1) for Yukon River dogs fall within the range for domestic dogs. No THg concentrations from any dog approached concentrations indicative of potential toxic effects (30,000 ng/g; Thompson, 1996). Smith and Armstrong (1975) reported Inuit sled dogs, subsisting largely on seal meat, which had levels of Hg, up to 11,500 ng/g in the liver, without apparent harm.

Smaller mammals such as mink, cats, dogs, and river otters appear to be more resistant to Hg than larger mammals (Eisler, 2006; Hinck et al., 2006). Eisler (2006) suggests that these differences are related to metabolism and possible higher Hg detoxification rates. For mammals, the main concern is the Hg exposure levels during the early stages of pregnancy—the first trimester in humans. K. hera (1979) reported that domestic dogs exposed to 0.1 mg/kg to 0.25 mg/kg of body weight during their entire pregnancy showed a higher incidence of stillbirths. The mean values of THg for the Yukon River village sled dogs exceed the no effects hazard concentrations for river otters (660 ng/g) and both mean levels for Russian Mission and Galena exceed the low effects hazard concentration (3290 ng/g) (Hinck et al., 2006). Since sled dogs are usually larger than river otters, the current risk to the sled dogs through the consumption of fish is minimal but biomarkers of cardiovascular and immune functions should be examined.

Published data on mercury in dog hair (or C. lupus, in general) is sparse (Hoekstra et al., 2003; Gamberg and Braune, 1999; Hansen and Danscher, 1995). In a study on sled dogs, Hansen and Danscher (1995) reported mercury levels in the hair of 10 dogs fed meat from marine mammals. The hair concentration ranged from 4105 to 34,743 ng/g, which is twice as high as our observed high value in Russian Mission (Table 1). Hair THg levels were also higher than concentrations in liver and kidney in their younger dogs. According to Hansen and Danscher (1995) the tissue distributions of THg levels in dogs exposed to high levels of THg are: hair: muscles (1:0.05); hair:liver (1:0.13); hair:kidney (1:0.25). Our results support Hansen's study in that we observed a significant difference between the fish fed dogs and commercially fed dogs from both New York and Salcha, Alaska (ANOVA, p.b.001). The hair THg concentration in the village dogs ranged from 139 to 15,800 ng/g. Hansen and Danscher (1995) found a biphasic relationship between THg hair concentration and age. We found no relationship with age.

Hg enters the food chain in the north as input from both regional geology, fires and atmospheric transport (Kelly et al., 2006; Nriagu, 1989; Peterson et al., 2007). Hg is generally found at greater concentrations in higher trophic level marine biota relative to terrestrial mammals due to the biomagnification of organic mercury forms and a longer food chain. While interspecies comparisons of THg are difficult because of feeding selection and behavior, our data may serve as reference for closely related canine species such as wolves and foxes in Alaska.

Sled dogs from Russian Mission, about 260 km from Norton Sound on the Bering Sea, had the highest mean level of THg in their hair (10,908 ng/g) and Fort Yukon, some 1300 km upriver from the delta, had the lowest (1822 ng/g) THg concentrations (Table 1). Galena (4528 ng/g) and Rampart (2446 ng/g) sled dogs were intermediate. In particular, the THg levels in Russian Mission sled dogs were among the highest reported from hair in Alaska, similar to those reported in hair for Steller sea lions (Beckman et al., 2002), fur seals (Beckman et al., 2002), and river otters (Ben-David et al., 2001). Importantly, the concentrations and patterns in Salcha, the Alaska reference site, are 10-fold lower than the sled dogs with the coastal subsistence diet. Mercury concentrations in fish can be highly variable in subarctic waters (Zhang et al., 2001; Jewett et al., 2003) and can differ with ecosystem
structure (Ben-David et al., 2001), wetland areas (Hinck et al., 2006) and forest fire frequency (Taylor, 2007; Kelly et al., 2006). The presence of large salmon runs is related to the diet of the dogs (and humans) along the Yukon and needs to be monitored as system dynamics change, including the marine-nutrient pump (Krummel et al., 2003).

The amount of THg transported by fish to sled dogs is greater at the river delta. The spawning salmon act as contaminant pumps by transporting Hg inland (Zhang et al., 2001). Sampling in this study was performed from August through September when salmon is plentiful and this was represented in diets fed to the dogs. Hair samples in sled dogs however, are representative of the past year of mercury accumulation, when dogs are fed dried fish, caribou and other supplements. Decreasing THg concentrations upstream from the river delta may be due to salmon availability for the rest of the year as well as other supplemented subsistence foods. All participating mushers fed salmon when available and supplemented the diet when supply was limited and when racing season began, in December. The number of salmon, both available and harvested, is greater in number and variety the closer in proximity the village is to the mouth at the Bering Sea. A second possibility for the decreasing trend as you travel upriver from the delta may be a consequence of contaminant elimination. During migration, salmon stop eating and rely on protein and lipids stores. As these stores are mobilized for energy use, Hg bound to protein and lipids may also be mobilized and eliminated from the animal. Limited data comparing Chinook salmon in Beaver (approximately 95 km downriver from Fort Yukon) with Pilot Station (approximately 95 km downriver from Russian Mission) does not support this hypothesis. More research is needed to understand the flow of THg from fish to ecoreceptors, including subsistence users.

It is interesting to compare our Yukon River sled dog data to archaeological samples of dog hair from Deering on the Seward Peninsula in western Alaska. The mean THg level for our archaeological samples is below the means for the Yukon River sled dogs. These THg levels from circa 780 AD are also well below the different thresholds for biological effects (Gamberg et al., 2005). This limited data suggests that Alaskan sled dogs can inform scientists about community health exposures not only in the present but also in the past. The archaeological samples also can be used to provide baseline information on non-anthropogenic THg exposure, albeit with the limitation of the lack of gender and age data for individuals.

As climate change impacts global marine systems and wildfire regimes, the release of Hg and other toxic trace elements may increase. It has been implied that remote western Arctic biota will be accumulating the highest levels of Hg in history as industrial development increases in Asia and is transported to higher latitudes (Fitzgerald et al., 1998). Continued biomonitoring of sled dogs and increased analysis of archaeofauna for THg is needed to document trends in exposure to subsistence users. This biomonitoring study was initiated years ago, when Elders from the Yukon–Kuskokwim drainages wondered if metals such as Hg posed a health threat. They knew Hg could accumulate in fish and be transferred up the food chain. They also knew that Hg has always been present in the environment (Gerlach et al., 2006) but wondered how the increased development such as mining and subsequent erosion of geological deposits would affect their food, and thus the health of future generations. Unfortunately we still cannot fully answer that question.

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