

# Mercury contamination of cattle in artisanal and small-scale gold mining in Bombana, Southeast Sulawesi, Indonesia

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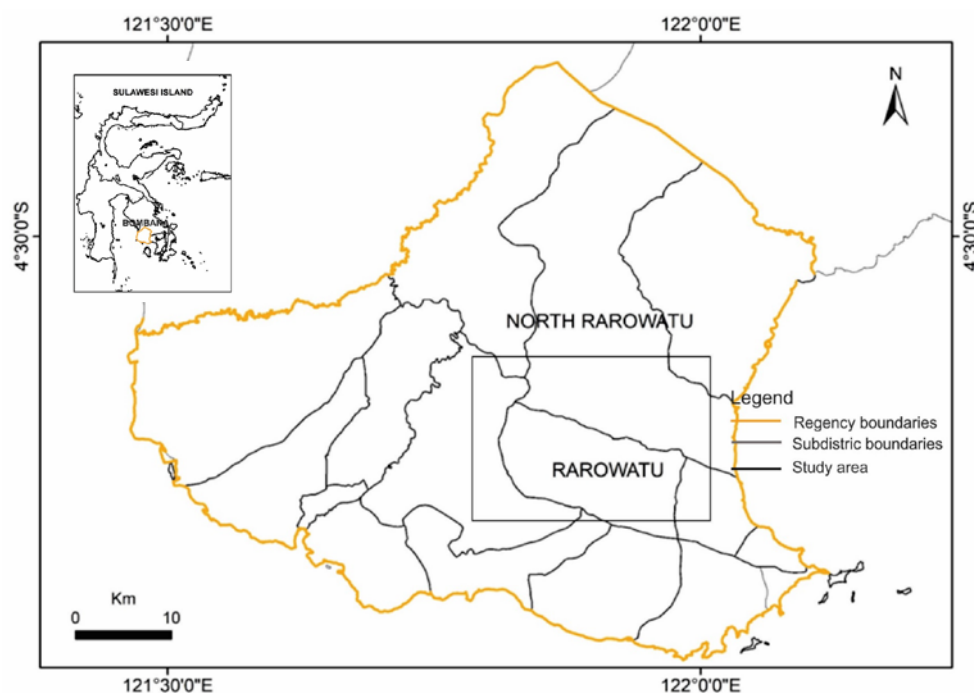
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## 1. INTRODUCTION

The World Bank recognizes that the ASGM sector plays an important role in developing communities so that it functions as a mechanism for poverty alleviation in developing countries. In some developing countries, small mines can be a major source of income for rural communities, and can provide income for investment. However, artisanal mining, and ASGM in particular, have an impact on environmental degradation, as small and artisanal miners can exploit mineral deposits that are considered uneconomical by modern industry [1].

Savanna landscape is a productive area for animal cultivation, especially for cattle colony. At regional scales, it becomes a center for beef production. Animal cultivation activity is generally generated by local inhabitants who stay temporarily in the Savannah area. At the same time, the savanna landscape contains the placer type of gold deposited and creates progressively ASGM sites. ASGM industry will attract migration from outside to create a socioeconomic problem. People believe that mercury pollution generated by ASGM activities will contaminate humans and cattle by the food chain process. However, this study proposes a hypothesis that mercury contamination occurs through atmospheric deposition.

Bioaccumulation of metals in the atmospheric contamination of large animals, including food animals such as cattle, and also in humans [2]. Diet in large mammals is the main route for metal accumulation, causing heavy metal accumulation in the kidneys, liver, bones, hair, and blood of these mammals. Cows, as large mammals, often swallow grass or other contaminated vegetation, drinking water, and small amounts of contaminated soil, although there is a possibility of exposure to metals through inhalation or treatment. The presence of Hg in meat and meat products is a major concern for public health and food security itself, causing widespread concern about public health. Therefore, the levels of these toxic metals in foodstuffs need to be controlled, and this is of particular interest in Bombana.



**Figure 1.** The locality of the study (modified from [3]).

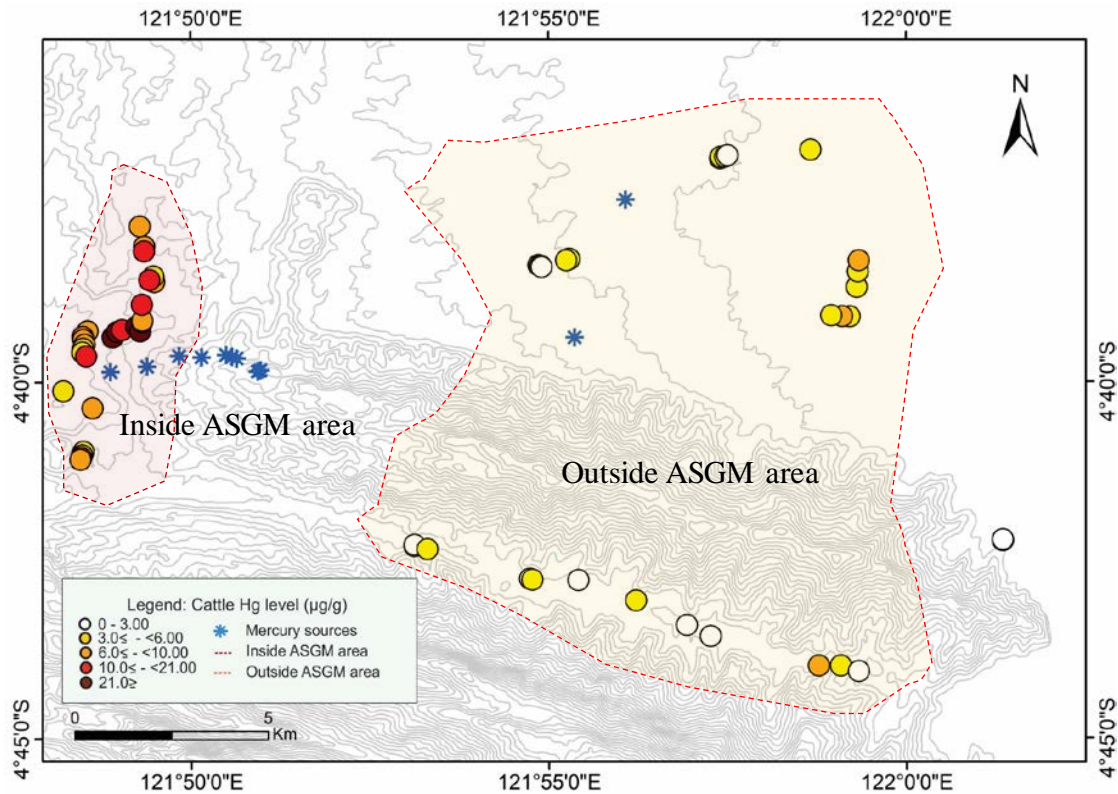
## 2. MATERIALS AND METHODS

### 2.1. Study Area

Bombana Regency is situated in the southeast peninsula of Sulawesi, southern part of the equator. The number of cattle in the Bombana area increased from 46,686 in 2014 to 54,029 in 2016, and they are currently the most important agricultural animal in this region. Bombana is the largest beef-producing center in Sulawesi, and accounts for about 70% of Sulawesi's total production. Altogether, there are about 5000 head of cattle in the sub-districts of Rarowatu and North Rarowatu, which have been the focus of the gold mining industry since 2008. Bombana has two ASGM sites that have expanded progressively over the past ten years [4] (Fig. 1).

### 2.2. Sample collection

To obtain cattle hair for analysis of total mercury (tHg) levels, we collected hair samples from similar numbers of male and female cattle aged 1–15 years, from cattle farmed in the two sampling areas, over the period from August 2016 to March 2017. We also attempted to determine the factors influencing the toxic levels of Hg in the samples.



**Figure 2.** The location of the samples site and distribution of Hg level (modified from [3]).

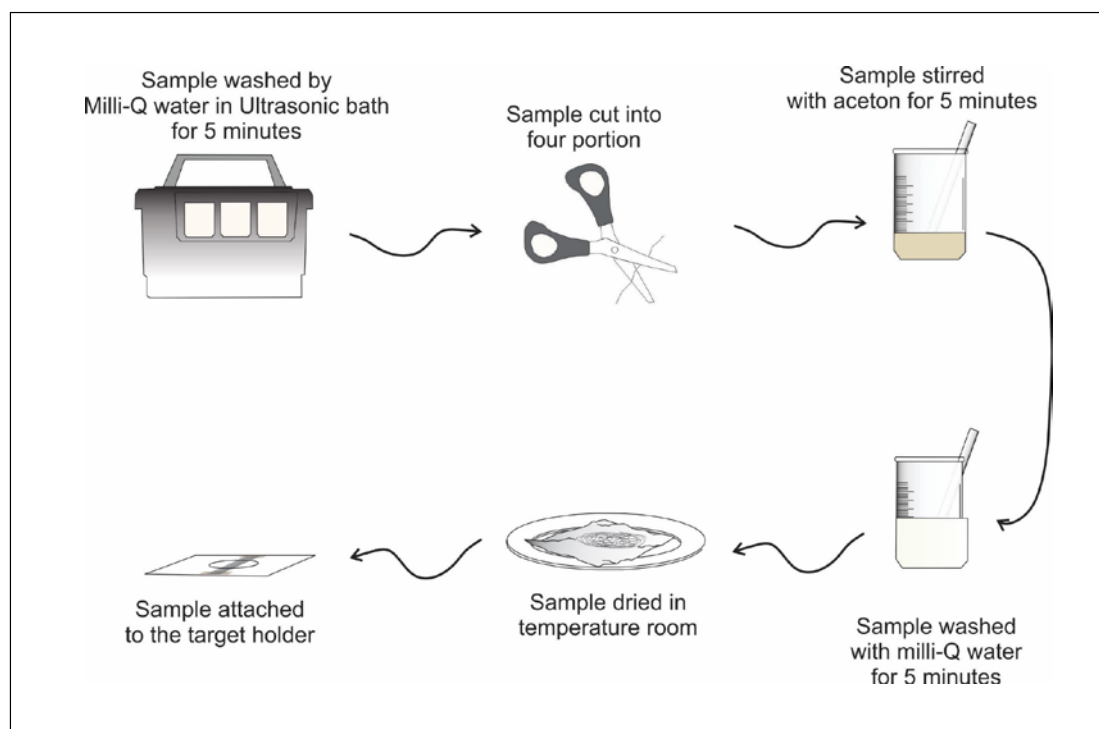
Hair samples were taken from cows representing each of the colonies using a non-random approach. The selection of each territory was based on the factors of distance and permitted access from the local community. The cattle live in groups and move within a 3 km<sup>2</sup> range for food. The sample distribution within the ASGM area was denser than outside of ASGM site. The landscape at the ASGM sites was dominated by tropical savanna hill, while outside the ASGM sites, landscape conditions were widely distributed among rivers, forest edges, and rice field locations (Fig. 2).

### 2.3. Analytical Method

The samples were shaken with Milli-Q water (18.2 MΩ/cm) in an ultrasonic cleaning bath (Sharp) for 5 min to remove dust, dirt, bacteria, and other contaminants. The samples were then dried with sterile paper on a clean glass plate for 10 min. The dried samples were stirred in acetone solution (Wako Pure Chemical Industries, Ltd., Osaka, Japan) for 5 min to remove any organic material that was not water soluble. The samples were then washed again with Milli-Q water and dried using sterile tissue at room temperature. Several strands of hair were randomly selected from each dried sample and cut into sections that included either the root or the end of the strand. About eight hair samples were attached parallel to the sample holder during preparation of the target (Fig. 3).

The Environmentally Certified Reference Materials (CRMs) from the National Institute for Environmental Studies was prepared for the use in the evaluation of the accuracy of tHg of

animal hair. The samples were analyzed by particle-induced X-ray emission (PIXE) at the Cyclotron Research Center, Iwate Technical University, Japan, using a proton energy beam of 2.5–3 MeV. In this method, the X-ray emissions from the sample that pass through the target area were detected by a Si (Li) detector. Low-energy X-rays were attenuated by a 300- $\mu$ m Mylar Mixer filter. Rays of uniform density were combined to have a diameter of 6 mm with a thick nickel foil and a diffuser in a graphite collimator system. The samples were placed on the target rectangle at an angle of  $\sim 35^\circ$  to the horizontal beam axis.



**Figure 3.** General preparation procedure of cattle hair sample.

#### 2.4. Data Analysis

The researcher in this study used the Student t-test, Mann-Whitney U test to identify differences in Hg concentrations in hair from cattle in three sampling area, with reference to sex, and sample site. Pearson correlation test and Spearman correlation test applied to determine correlation between Hg concentration, duration of exposure and age. Differences were considered significant for p values  $< 0.05$ . In all statistical analyses, Paleontological Statistic (PAST) Ver. 3.17 and IBM SPSS Statistic 21 Ver. 21.0 were used for data analyses.

### 3. RESULTS

#### 3.1. Laboratory Data

The analytical results are listed in Table 1. Of the 63 samples of hair collected, 34 were taken from cattle farmed within the two ASGM areas, and 29 were from cattle farmed outside

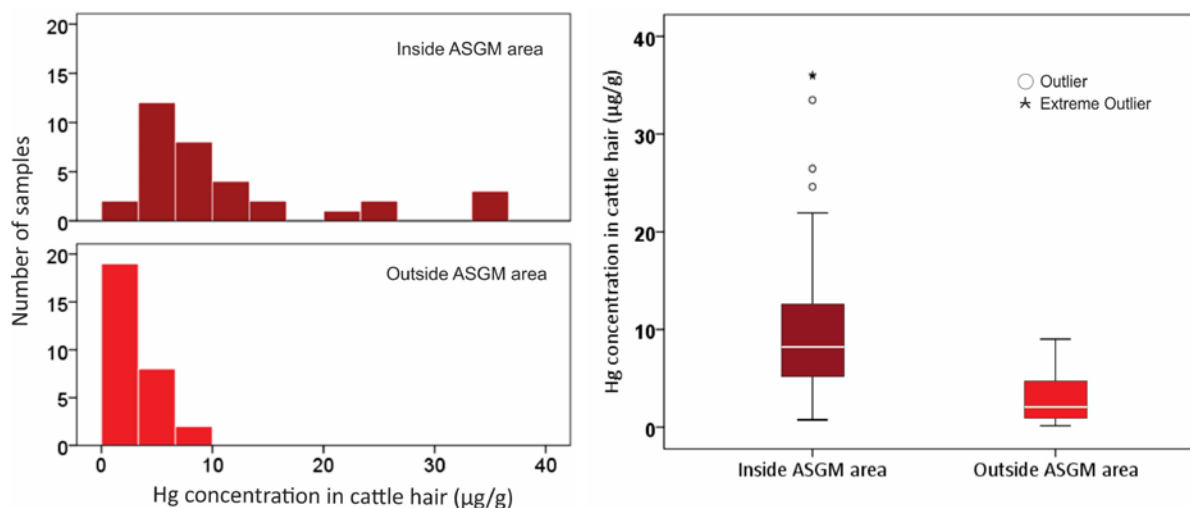
the ASGM areas (total female,  $n = 42$ ; male,  $n = 21$ ). Fewer males were sampled, because they are farmed in smaller numbers than females. There was no statistically significant relationship between sex and sampling location ( $p > 0.05$ ).

**Table 1.** Number, sex, and age of cattle, distribution by location and Hg concentration.

Animal Data		Sample Location		Mann-Whitney <i>U</i> Test	
		Inside of ASGM Site ( $n = 34$ )	Outside of ASGM Site ( $n = 29$ )		
Quartile	Q1	5.20	1.03		
	Q3	8.22	4.57		
Hg levels ( $\mu\text{g/g}$ ) and sample location	Mean $\pm$ SD	11.44 $\pm$ 9.52	2.89 $\pm$ 2.45	$p(0.000) < 0.05$	
	Median	8.22	2.14	*	
Age (years) and sample location	Mean $\pm$ SD	6.00 $\pm$ 2.90	4.00 $\pm$ 3.07	$p(0.003) < 0.05$	
	Median	6	3	*	
Hg levels ( $\mu\text{g/g}$ ) sex and sample location	Male ( $n = 21$ )	Mean $\pm$ SD	10.17 $\pm$ 7.13	2.41 $\pm$ 2.40	$p(0.037) < 0.05$
		Median	7.52	1.89	*
	Female ( $n = 42$ )	Mean $\pm$ SD	11.98 $\pm$ 10.45	3.19 $\pm$ 2.51	$p(0.084) > 0.05$
		Median	8.62	3.12	**

\* = significant at  $p < 0.05$ ; \*\* = non-significant at  $p > 0.05$ .

Minimum and maximum ages were the same in both of the sampling sites, inside and outside the ASGM area, and the mean and median ages were higher in the two ASGM sites than outside the sites. Age w We used the Student t-test, Mann-Whitney U test to identify differences in Hg concentrations in hair from cattle in three sampling area, with reference to sex, and sample site. Pearson correlation test and Spearman correlation test applied to determine correlation between Hg concentration, duration of exposure and age. Differences were considered significant for  $p$  values  $< 0.05$  (Fig. 4).



**Figure 4.** Hg concentration of cattle. (A) The histogram of cattle Hg concentration; (B) The boxplot of cattle Hg concentration on two group sampling.

The mean Hg concentration in hair from ASGM areas ( $n = 34$ ; 11.44  $\mu\text{g Hg/g}$  hair) was almost three times than that in hair from outside the ASGM areas ( $n = 29$ ; 2.89  $\mu\text{g Hg/g}$  hair). The Mann-Whitney *U* test revealed a statistically significant difference in Hg concentration in

hair from inside and outside the ASGM sites ( $p < 0.05$ ) (Fig. 4). The values for the two ASGM sites combined were as follows: range, 0.15–36.08  $\mu\text{g/g}$ ; mean, 7.60  $\mu\text{g/g}$ ; standard deviation, 8.33  $\mu\text{g/g}$ .

## 4. DISCUSSION

In general, the toxic effect of Hg depends on the form and dose of Hg, the exposure duration, and the ingestion route. Exposure to Hg has been found to be toxic to both humans and animals [5]. There are similarities and differences in the toxic effects of the various forms of Hg [6]. Organic Hg is the more toxic form, and causes poisoning if ingested [7]. The major targets of toxicity from inorganic and organic Hg are the kidneys and the central nervous system, respectively. The clinical signs of Hg poisoning in cattle vary greatly, and include ataxia, neuromuscular incoordination, and renal failure, followed by convulsions and a moribund state [8].

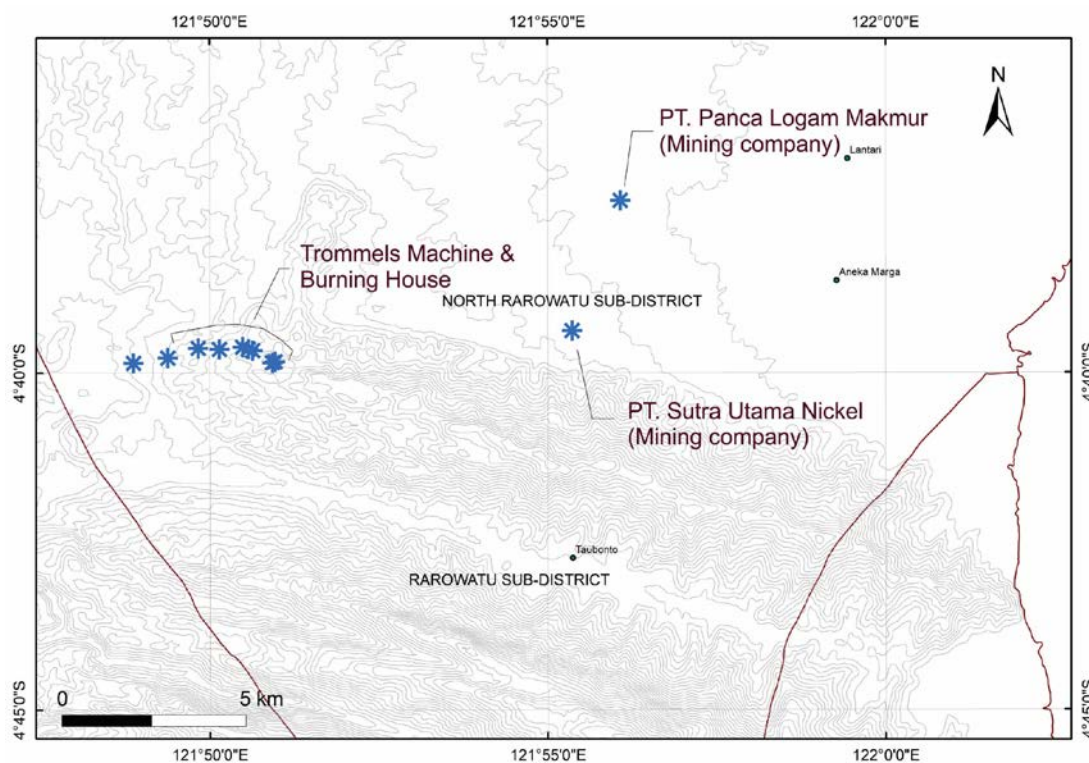
The response of an animal population to a certain amount of toxicant may differ among individuals, because various factors can modulate the overall toxic response [9]. Drugs and toxins may be substituted in some kinetic or dynamic interactions when binding to plasma proteins or receptors at the active site of the enzyme involved in [1]. The main factors affecting chemical toxicity in individuals and in populations are breed, age, sex, pathophysiological events, diet composition, environmental conditions, the source, and route of exposure [7]. Here, we examine the influence of environmental conditions, age, maturity, sex, and hormonal status.

### 4.1. Environmental Condition

In the mid-2008 a new gold mining site was found in Bombana area, a new region in Southeast Sulawesi Province, Indonesia (Fig. 5). The first source of pollution, derived from burning house working on the processing of gold ore material. The location is the same as the trommel house because it is in the same building and managed by the same person. At this facility, several stages of gold processing have the potential to produce mercury (Hg) pollution. Generally, through grinding in trommels process, amalgam burning and tailings disposal. In the burning houses, 50 % of the Hg added to small ball mills (trommels machine) is lost: 46 % with tailings and 4 % when amalgam is burned. The Hg lost through ball mills and tailings will break into the environment and contaminate the soil, body water and plants. While Hg lost through amalgam combustion will be released into the atmosphere and move in the direction of the wind. Hg particles will fall to earth and attached to plants and animals.

The second source of pollution come from the four gold mining companies, PT. Panca Logam Makmur (PLM), and PT. Sultra Utama Nickel (SUN), are since working on 2008. Unlike the ASGM industry, this sector manages large-scale gold seeds, using advanced technology and heavy equipment, resulting in uncontrolled natural exploitation. Mining companies do not use

traditional Hg officially in employment, but workers do illegal tailings processing within the company's work area.

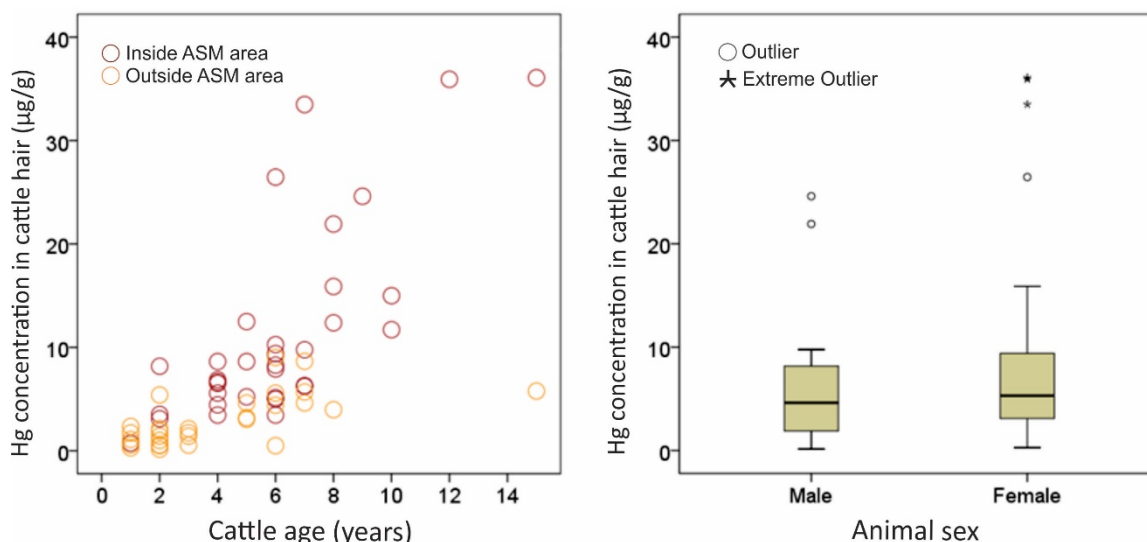


**Figure 5.** New source of Hg contamination in Bombana area.

#### 4.2. Age and sex of animal

Previous animal studies have reported an increasing sensitivity to toxins with age, and that the effect was more pronounced in food-producing species that have a long life cycle [5]. Pharmacokinetic conditions in groups such as older people, different ethnic groups, and pregnant women will have different pathological effects (e.g., renal and hepatic insufficiency, cardiac dysfunction, and obesity). The xenobiotic cleansing function of the liver and kidneys becomes less effective with increasing age due to a reduction in renal blood flow and glomerular filtration, rather than to a significant reduction in biotransformation capacity (Fig. 6A) [10].

Variations in response to certain chemicals that are related to physiological differences between the sexes can affect hormonal systems and functions. Biologically, the difference is large if not exclusively, related to gonadal hormone secretion. Biological differences between the sexes contribute to many diseases and sex disorders [1] [8]. Because male and female might have specific pharmacokinetic and pharmacodynamic differences, sex could affect the response to chemicals [10] (Fig. 6B).



**Figure 6.** The main factors affecting chemical toxicity in cattle. (A) The positive correlation between Hg concentration and cattle aged; (B) The boxplot of cattle Hg concentration based on animal sex.

Previous studies have reported Hg accumulation in terms of the sex of the animal in cattle, studies of other species have shown higher levels of Hg accumulation in females than males [4]. For example, studies of deer and calves have shown higher levels of Hg accumulation in females [6]. It is of note that that study found no significant difference in Hg levels in the livers and kidneys between male and female deer. Experimental studies [6] have shown that the excretion and accumulation of Hg in animal organs are essentially influenced by the status of the reproductive hormones; however, as yet our understanding of the underlying mechanisms is incomplete.

## 5. CONCLUSIONS

This study evaluates the environmental effects of ASGM activities in Bombana mining area based on cattle hair data. The environmental effect assessment results revealed that mercury (Hg) contamination of cattle was attributable to the uncontrolled use of Hg in gold processing. Levels of Hg in cattle exceeded the international permissible guideline values. The Hg levels in cattle designates that the Hg pollution is a severe problem in Bombana area. High concentrations of Hg were found in the hair of cattle farmed in Bombana, both within and outside of ASGM areas. The mean Hg concentration was higher in females than in males, but the difference was not statistically significant. Further, Hg concentration in cattle hair was positively correlated with the age of the cattle.

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