1	Spatial geochemistry influences the home range of elephants.
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22 Abstract

23 The unique geochemistry surrounding the Palabora Mining Company (PMC) land may act as a 24 micronutrient hotspot, attracting elephants to the area. The PMC produces refined copper and 25 extracts phosphates and other minerals. Understanding the spatial influence of geochemistry on the 26 home range size of African elephants is important for elephant population management and 27 conservation.

28 The home ranges of collared elephants surrounding the PMC were significantly smaller (P=0.001) 29 than conspecifics in surrounding reserves, suggesting that their resource needs were met within these smaller areas. Environmental samples (soil, water and plants) were analysed from the mine 30 31 area and along six transects radiating from the mine centre. Tail hair and faecal samples from 32 elephants at the PMC, and conspecifics within the surrounding area were analysed. All samples were 33 analysed for minerals essential to health and potentially toxic elements (PTEs; As, Ca, Cd, Cu, Fe, K, 34 Mg, Mn, Na, P, Pb, Se, U, V and Zn). Results show that the geochemistry at the PMC is different 35 compared to surrounding areas, with significant elevations seen in all analysed minerals and PTEs in 36 soil closer to the mine, thereby drawing the elephants to the area. Additionally significant elevations 37 were seen in elements analysed in water and vegetation samples. Elephant tail hair from elephants 38 at the mine was significantly greater in Cd, whilst Mg, P, Cu, As, Cd, Pb and U concentrations were 39 significantly greater in elephant faecal samples at the mine compared.

40 When micronutrient hotspots overlap with human activity (such as mining), this can lead to poor 41 human-elephant coexistence and thus conflict. When managing elephant populations, the influence 42 of mineral provision on elephant movement must be considered. Such detailed resource information can inform conservation efforts for coordinated programmes (UN SDGs 15 and 17) and underpin 43 44 sustainable economic activity (UN SDG 8, 11 and 12).

45 Keywords: Loxodonta africana, minerals, mining, potentially toxic elements, elephant movement 46

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#### 49 1.0 Introduction

50 The increase in human population and global intensification of agriculture have significantly reduced 51 African savanna elephant (Loxodonta africana) populations, through habitat reduction and 52 fragmentation, causing the overlap of human and elephant habitation, leading to increased human-53 elephant conflict (HEC; Blanc, 2008). Elephants are forced into increasingly smaller areas, often 54 restricted by fencing or encroaching anthropogenic activities, resulting in increased pressures on 55 these areas to meet the elephants' resource needs. This can present nutritional challenges, resulting 56 in altered elephant movement patterns and distribution in efforts to seek out required minerals. 57 Elephants move to meet their mineral needs, and use available micronutrient hotspots, causing HEC, 58 when these overlap with human activities (Sach et al., 2019). Minerals are required by elephants for 59 a variety of biological processes including energy metabolism, organ and immune function, 60 reproduction and cellular growth (Ishiguro, Haskey and Campbell, 2018). 61 62 Geochemistry influences mineral availability in soils, and thereby in plants and water to elephants 63 (Prins and Langevelde, 2008). Understanding how the geochemistry of an area, and presence of 64 micronutrient hotspots, influences mineral provision to the animal, informs how geochemistry 65 influences home range size, especially when anthropogenic activities constrain long-distance 66 movements. Largely, plants reflect the soil mineral profile, plants growing in deficient areas lack key 67 minerals, which can result in deficiencies in the consumer (elephant). In contrast, plants growing in 68 mineral rich areas pass on the mineral abundance to the consumer (Joy et al., 2015). Geochemical 69 properties (including organic matter and soil pH), and the ability of plants to extract minerals from 70 the soil will influence the availability of these minerals to elephants (Bowell and Ansah, 1994; 71 Maskall and Thornton, 1996).

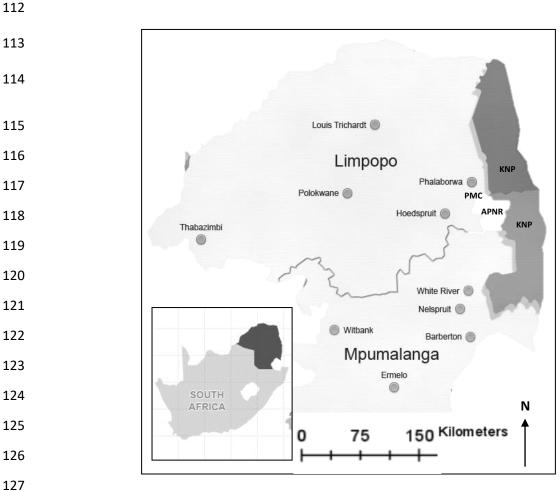
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African elephants move and adapt their food selection, to meet their target levels of (as yet
undetermined) minerals (Bax & Sheldrick, 1963). From here-on reference will be made to mineral in

75 terms of the nutrient requirement for elephants. It is suspected that in volcanic areas such as the 76 Palabora Mining Company (PMC), levels of micronutrients will be elevated, acting as a 77 micronutritent hotspot, with a reduction in elephant home ranges size (Greyling, 2004). This may be 78 beneficial or detrimental to elephants. In areas where the soil is generally deficient in minerals, it 79 may allow elephants to meet their mineral needs within a small area. However, as with other 80 mammals, dietary excess of minerals or potentially toxic elements (PTEs) can occur from 81 overconsumption, causing toxic effects; data is limited as to these threshold levels for elephants 82 (Sach et al., 2019). Elephants are large, slow-growing and can accommodate extended periods of 83 nutrient deficiency due to their nutrient stores (Prins and Langevelde, 2008). Excess consumption of 84 minerals or PTEs to harmful levels is likely to take several years (Ullrey, Crissey and Hintz, 1997). 85 86 As well as micronutrients, drivers for elephant movement include availability of food and water, 87 social interaction, human activities, safety and access to shade (Wall, Douglas-Hamilton and Vollrath, 88 2006). The distance travelled by elephants to meet their resource needs, will be reflected in their 89 home range size (de Knegt et al., 2011). Mineral provision influences elephant food selections; for 90 example, the Associated Private Nature Reserves (APNR), South Africa are suspected to have a 91 localised phosphorus (P) deficiency, elephants increased their consumption of leaves from trees that 92 had been fertilised with P (Pretorius et al., 2011, 2012). Secondly, females in family units maximised 93 P intake by ingesting leaves with higher P content, to meet their increased requirements, compared 94 to larger bodied males who consumed other lower P plant parts (Greyling, 2004). Phosphorus plays a 95 role in reproduction and lactation (Groenewald and Boyazoglu, 1980). It is predicted that if an area 96 such as the PMC is a micronutrient hotspot, elephants will remain within the locality, to meet their 97 resource needs for minerals as demonstrated by Tucker et al. (2018), especially if the surrounding 98 soils are poor in several essential micronutrients such as P, as suggested by Greyling (2004) and 99 Pretorius (2011, 2012).

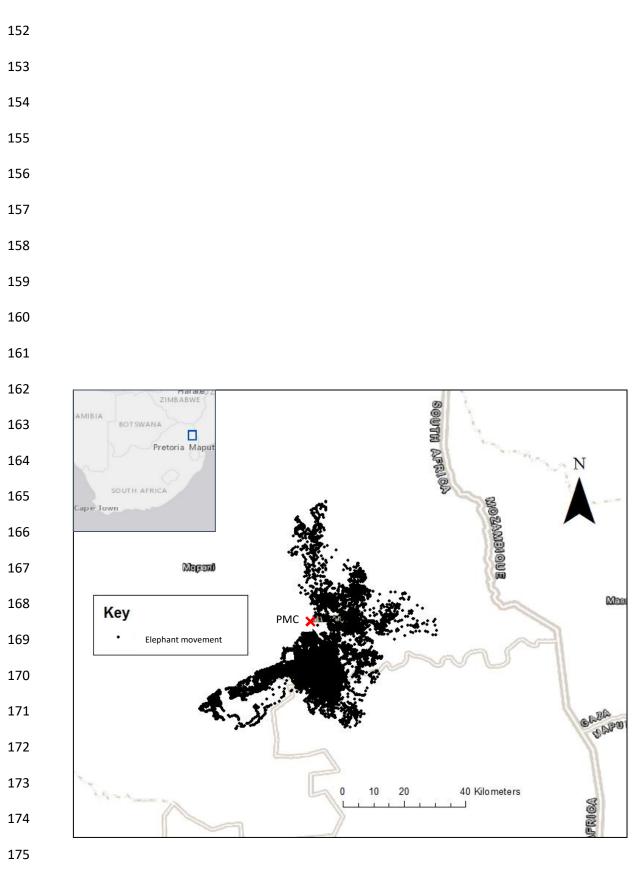
100 The aim of this study was to understand the spatial influence of geochemistry on the home range 101 size of elephants, using the Palabora Mining Company (PMC) property and surrounding national park 102 land as a case study of contrasting environments. The following objectives were used to achieve the 103 aim: (1) Determine if mineral levels in soil, forage and water near the mine are greater than the 104 nearby Kruger National Park (KNP)/APNR and hence may influence a reduced elephant home range 105 size; (2) Establish baseline levels for key minerals and PTEs in African elephant tail hair and faeces as 106 potential biomarkers, and (3) Determine if the elephant tissues (tail hair and faecal samples) 107 collected near the mine contain greater concentrations of essential minerals and PTEs, compared to 108 elephants in surrounding reserves, away from the mine.

- 109 2.0 Materials and methods
- 110
- 111



128 Figure 1: Study area showing the Kruger National Park (KNP), Associated Private Nature Reserves 129 (APNR) and Palabora Mining Company (PMC). 130 The study was conducted on the Palabora Mining Company (PMC) land near Phalaborwa town, 131 South Africa and adjacent areas within the KNP and the APNR (Fig. 1). From west to east, the 132 geological succession of the KNP changes from granitic to basaltic. Granites generally form nutrient poor substrates whereas basaltic rocks form nutrient rich substrates (Venter and Gertembach, 133 1986). The APNR is located on the western border of the KNP, and is made up of gneiss, granite or 134 135 magmatite (Venter and Gertembach, 1986). Elephants can move freely amongst the KNP, APNR and 136 PMC lands. Elephant incursion into the PMC can cause financial losses and risk to elephant and 137 human life. Elephants can damage infrastructure, inhibit mining operations and cause elephant, 138 vehicle and train collisions. 139 In this generally micronutrient poor environment, the Palabora Igneous Complex has a unique 140 mineral rich rock formation. Commercial mining began in 1954, with open-cast mining of foskorite 141 and pyroxenite, thereafter the PMC began mining the same ores for copper and magnetite, 142 developing into the country's main producer of refined copper, operating over 1950 ha (Roux et al., 1989). The NGO Elephants Alive (EA) have collared elephants throughout the APNR, and seven 143 144 elephants utilising the mine area (movements in Fig. 2). The home range of these mine collared 145 elephants was calculated using a-LoCoH 90% (Getz and Wilmers, 2004), and was smaller than that of 146 neighbouring elephants within the APNR (Table 1), animals of the same sex, age category and 147 wearing collars for the same time period were compared. Elephant census data, showed that 148 elephant density within the operational PMC (1.4 per km<sup>2</sup>) was larger than that within the surrounding KNP (0.8 per km<sup>2</sup>; Lerm and Swemmer, 2015). 149

150



176 Figure 2: Fixes of collared elephants surrounding the Palabora Mining Company (PMC) site between
177 15.6.2012 and 23.7.2017

	Average Home Range calculated using LoCoH 90% (km <sup>2</sup> )	Standard error of mean	Min/max (km²)	Number of elephants
РМС	529	78	200/728	7
Neighbouring reserves	1305	265	498/2244	7

180 **Table 1:** Home ranges of elephants within the Palabora Mining Company (PMC) land and

181 neighbouring reserves. Full data given in Supplementary Information Table 9.

# 182 Sample site selection

183 Fifty-three sampling sites were selected on six transects radiating out from the PMC, to include

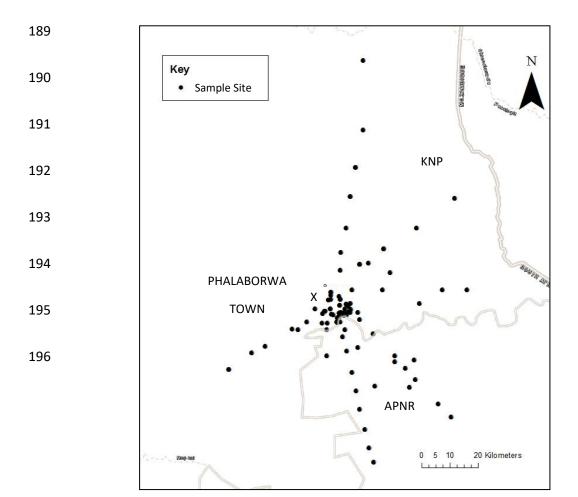
points within and outside of the area occupied by the collared elephants at the mine (Fig. 3).

185 Transects were used to observe if an elemental gradient from the PMC was present. Additionally, 43

186 sampling sites were identified within the PMC (Fig. 3). Sample sites were not selected to the north

187 west of the PMC area, this is a fenced urban area with minimal elephant movement.

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Figure 3: Sampling sites for environmental and faecal samples. KNP=Kruger National Park;
 APNR=Associated Private Nature Reserves. The PMC (Palabora Mining Company) is located where
 transects cross, south of Phalaborwa town.

203

# 204 Sample Collection

205 Environmental and faecal sampling was conducted during September 2017 and September 2018,

206 within a 50 m radius around each sampling point. Trace element free paper bags were used for

- 207 plant, soil, faecal and tail hair samples. All samples were transported to the lab within 8 h of
- 208 collection; plant and water samples in a cooler, tail hair, faecal and soil samples at ambient
- 209 temperature.

# 210 Environmental sampling

- 211 Plant parts (approx. 500g per sample, n=100) were sampled from seven species commonly
- consumed by elephants (Table 2; Smallei and O'Connor, 2000; Holdø, Dudley and McDowell, 2002;
- 213 Codron et al., 2006; Pretorius et al., 2011, M Henly pers. comm 2017). Not every species or part was
- found at the sampling site. Samples were taken randomly from the plant, mixed sized leaves were
- 215 sampled, branches/ roots of approx. 5cm in length were cut using secateurs and bark was scraped
- 216 off the trunk using a chisel.

Species	Common name	Part sampled
Colophospermum mopane	Mopane	Leaves
Grewia monticola	Silver Raisin	Bark
		Leaves
Senegalia nigrescens	Knob Thorn	Bark
Combretum apiculatum	Red Bushwillow	Roots
Lannea schweinfurthii	False Marula	Leaves
		Inner bark
Dichrostachys cinerea	Sicklebush	Branches

	Leaves
Dwarf Bush-cherry	Branches
	Bark
	Leaves
	Dwarf Bush-cherry

# 217 **Table 2:** Plant species and plant parts sampled within this study

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- 219 Soil samples (n=97, approx. 500g per sample) were collected at each site, from surface soil using a
- trowel, to a depth of 15cm, from five separate points within a 1-m<sup>2</sup> grid. Water was sampled
- 221 opportunistically at sample sites, key rivers or identified elephant drinking points (n=36); two 30 ml
- samples were collected in Nalgene HDPE bottles, filtered (0.45 μm).

## 223 Biological Sampling

- 224 Elephant faecal samples (n=94, approx. 500g per sample) were taken from the centre of fresh, intact
- boluses with circumference of >40cm, to indicate adult size (Jachmann and Bell, 1985), as calf
- samples on a pre-weaned diet could bias results (Cook *et al.*, 1994). On return to the laboratory,
- samples were oven dried at 50 °C for 24 h.
- Tail hair samples were plucked from the tail (1-3 hairs per animal) between March 2002 and July
- 229 2018 during routine collaring operations or management activities throughout the KNP, APNR and
- 230 PMC, as part of the South African National Parks Bio-bank (SANParks), or by EA. Tail hair samples
- were taken up to 170km from the PMC (n=200 from non-mine collected by SANParks and EA, n=7
- from mine area collected by EA). All sedations were carried out using the SANParks SOPs for Capture
- 233 Transport (Standard Operating Procedures for Capture Transport and maintenance in Holding
- 234 Facilities of Wildlife, 2017).

# 235 Sample Preparation

Soil samples were air-dried, crushed and sieved to  $\leq 2$ mm particle size and further milled to  $\leq 40\mu$ m in an agate ball mill. Water samples were filtered with a hydrophilic 25mm Minisart filter and acidified to 1% HNO<sub>3</sub> and 0.5% HCl. Plant and faecal samples were oven dried at 50 °C for 24 h, and passed

- through a food blender as described by Watts et al. (2019). Elephant tail hair samples were cleaned
- as described in Middleton et al. (2016) and autoclaved in line with DEFRA requirements.

## 241 Sample Digestion for ICP-MS analysis

- Soil samples (0.25 g) were digested in a mixed acid solution (HF: 2.5 ml/HNO<sub>3</sub>:2 ml/HClO<sub>4</sub>:1
- 243 ml/H<sub>2</sub>O<sub>2</sub>:2.5 ml) on a programmable hot block; 0.5 g of plant samples or faecal samples were
- digested in HNO<sub>3</sub>:10 ml/H<sub>2</sub>O<sub>2</sub>:1 ml mixed solution in a closed vessel microwave heating system
- 245 (MARS Xpress) as described in Watts et al. (2019). Elephant tail hair samples (variable weight) were
- digested in HNO<sub>3</sub>:4 ml/H<sub>2</sub>O<sub>2</sub>:1 ml mixed solution in a closed vessel microwave heating system (MARS
- 247 Xpress) as described in Middleton et al. (2016). Tail hairs from the non-mine elephants were
- digested and analysed whole, and those from the collared elephants at the PMC were cut into 3–5
- 249 cm sections, down the length of the hair, for future profiling, prior to digestion and subsequent
- analysis. Soil, plant, faecal material and tail hair data is presented as dry weight.

#### 251 Elemental Analysis

Elemental analysis was conducted on all prepared samples by inductively coupled plasma mass spectrometry (ICP-QQQ; Agilent 8900x) using collision cell mode (gas modes: H<sub>2</sub> for Se, O<sub>2</sub> for As, He for all remaining elements). Fifteen biologically functional or potentially toxic elements were selected for this study; Ca, copper (Cu), iron (Fe), potassium (K), Mg, manganese (Mn), Na, P, selenium (Se), zinc (Zn), arsenic (As), cadmium (Cd), lead (Pb), uranium (U) and vanadium (V). Sample blanks were run to determine the practical limit of detection (LOD, 3\*STDEV).

258 Analytical Quality Control

259 The accuracy of the elemental analysis was verified by analysing the following certified reference260 materials (CRM)s:

- Human Hair (GBW07601, China)
- Spinach leaves (SRM1570a, NIST, USA)

- Tomato leaves (SRM1573a, NIST, USA)
- Basalt rock (BCR-2 United States Geological Survey, USA)
- Soil (SRM2711a, NIST, USA)
- Soil (BGS 102, British Geological Survey, UK)
- In house human toenail (BAPS 2014) reference material

268 The concentrations of all elements of interest in the reference materials had an acceptable 269 accuracy to the target values, of  $97\% \pm 39\%$ , data detailed in Supplementary Information Table 3.

# 270 Statistical Analysis

The evidence for differences between mine and non-mine elephant home range size was assessed by a Wilcoxon-test of the null hypothesis that the median home range size value was the same for the collared mine and non-mine elephants. Statistical analysis was conducted using 'R' Studio version 3.5.0.

275 The evidence for differences between mine and non-mine elephant tail hair and faecal samples with 276 respect to analytes was assessed by a Student's t-test of the null hypothesis that the mean value was 277 the same for samples from the mine and non-mine. Boundaries to define the mine and non-mine 278 were based on physical land ownership. The t-test was performed assuming that the variances 279 within the two groups were not necessarily the same, and computing effective degrees of freedom 280 for the resulting t-statistic according to the Satterthwaite-Welch equation (Welch, 1947). This is a 281 conservative approach when, as here, the sample sizes are unequal. 282 Each family of tests (t-tests on one matrix for the set of minerals, or tests of trend models for some

283 environmental matrix on the set of minerals) can be regarded as a multiple hypothesis testing

- 284 exercise, because each mineral was not considered in turn, but rather examined for evidence that
- specific minerals display behaviour of interest. For that reason we undertook false discovery rate

control (FDR) following Benjamini and Hochberg (1995). The FDR is the expected proportion of
rejected null hypotheses that should have been accepted. Here we controlled the FDR at 0.05,
computing adjusted P-values for each family of tests using the p.adjust command in the base
statistical library of the R package (R Core team, 2017).

290 The environmental data, on soil, water and plants, were examined for evidence that there is a 291 dependency of the measured concentration on distance from the mine. This was done using a 292 polynomial function of distance. For plants, leaves only were used to demonstrate spatial variation, 293 a full dataset and comparison for the plant data (all plants versus leaves) is in Supplementary 294 Information Table 4. The data on soil, water and plants were collected from transect points radiating 295 from the mine, sampling at more or less regular intervals. Because the samples are not collected 296 from sites selected independently and at random, it is not possible to make sound inferences based 297 on standard ordinary least squares methods (Lark and Cullis, 2004). Rather, it is necessary to fit a 298 linear mixed model (LMM) to the data, with the fixed effects comprising polynomial terms in 299 distance to the mine, and the random effect comprising both an independent and identically 300 distributed error term and a spatially correlated random effect. The models were fitted using the 301 Ime and update functions from the nlme library for the R platform (R Core Team, 2017; Pinheiro et al., 2018). 302

303 A quartic polynomial (first, second, third and fourth order terms) in distance was initially fitted to the 304 data by ordinary least squares, and summary statistics and the histogram of the residuals were 305 examined to decide whether to analyse the data on their original units or after transformation to 306 natural logarithms. The full model was then fitted as a LMM using residual maximum likelihood 307 (REML), and models with spherical and exponential correlation functions for the spatially-dependent 308 random effect were compared on their likelihood. The selected spatial correlation function was then 309 retained for all further models for this variable on the matrix being considered. The full quartic 310 model was then re-estimated using ordinary maximum likelihood to allow comparisons with

alternative models with different fixed effects. A cubic model was then fitted (i.e., dropping the
quartic term), and the quartic and cubic models were compared on the log-likelihood ratio statistic
to test the null hypothesis that the coefficient for the quartic term was zero. If this null hypothesis
was rejected then the full model was retained and compared with a null model in which the only
fixed effect was a constant mean. This latter test was recorded as the strength of evidence for a
trend with distance to the mine. If, on the other hand, the null hypothesis was accepted, then the
quartic term was dropped and the cubic model compared with a quadratic, and so on.

As with the comparisons between the mine and non-mine areas by the t-test, each set of spatial

319 models over all elements on a particular matrix was treated as a family of multiple hypotheses to be

320 tested with FDR control. The same method was used to do this as described above for the t-tests.

321 One data point furthest from the mine was removed, because of the considerable leverage that this

could have on a trend model. It was also necessary to "jitter" some of the spatial coordinates,

moving them 1 metre in a random direction. This is because, although none of the environmental

324 samples on any matrix were actually from the same location, the GPS coordinates were duplicated

as GPS readings are only precise within 6 m. It was one observation out of any such pair that was

326 "jittered" in this way using the jitterDupCoords function from the geoR package in R (Ribeiro and

327 Diggle, 2018).

328 Statement of Ethical Approval

329 Required ethical clearance and permits were obtained from relevant authorities

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331 3.0 Results

332 Home range size

333 The null hypothesis that the mean home range size was the same for the mine and non-mine areas

334 could be rejected. The Wilcoxon test showed a significant difference between mine and paired

conspecifics outside of the mine (P= 0.001; Table 1; Supplementary Information Table 9).

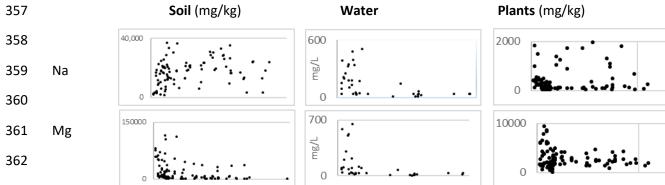
#### 336 Environmental samples

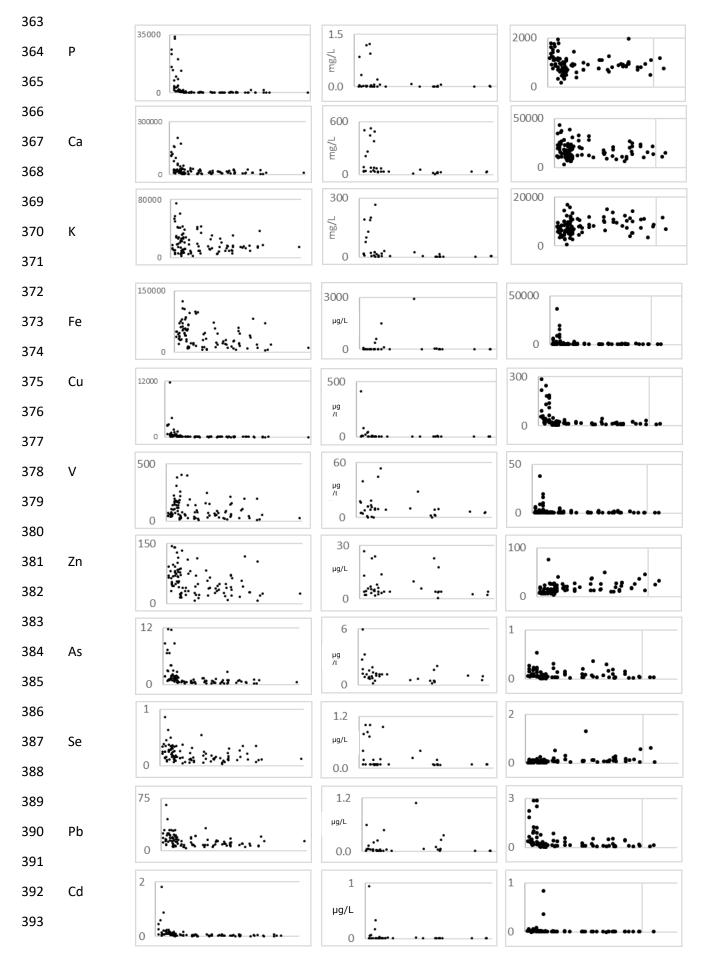
With FDR control at 0.05, the null hypothesis of no spatial trend in concentration in the soil with 337 338 distance from the mine could be rejected for all investigated elements. Inspection of the trend 339 model shows that in all cases concentrations decline with distance (Fig. 4; Table 3; Supplementary Information Table 3 and 8). With FDR control at 0.05, the null hypothesis of no spatial trend in 340 341 concentration in the water with distance from the mine could be rejected for four investigated 342 elements (Ca, K, Fe and Cu), with concentrations declining with distance (Fig. 4; Table 3; 343 Supplementary Information Table 5 and 8). With FDR control at 0.05, the null hypothesis of no 344 spatial trend in concentration in plants (leaf samples) with distance from the mine could be rejected 345 for nine investigated elements (P, Mg, Mn, Fe, Cu, Zn, Se, Cd, and U), with concentrations declining 346 with distance (Fig. 4; Table 3; Supplementary Information Table 4 and 8).

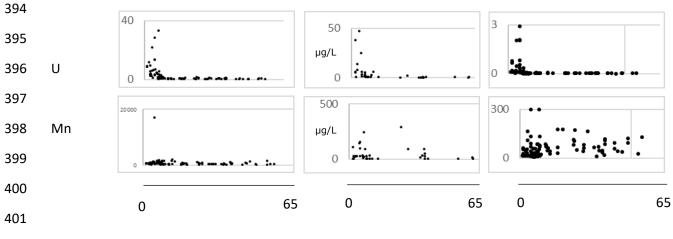
#### 347 Elephant biomarkers

- 348 With FDR control at 0.05, the null hypothesis of no difference between the mine and non-mine
- faecal samples could be rejected for Mg, P, Cu, As, Cd, Pb and U (larger concentrations in the
- elephants near the mine) and Na, Mn, Zn and Se (smaller concentrations in the elephants near the
- 351 mine; P<0.05; Figure 5; Supplementary Information Table 7; 8).
- With FDR control at 0.05, the null hypothesis of no difference between the mine and non-mine tail hair samples could be rejected for Cd (larger concentrations in the elephants near the mine) and K and Se (smaller concentrations in the elephants near the mine; P<0.05; Figure 5; Supplementary Information Table 6; 8).

#### 356 Environmental data







Distance from the mine (km)

**Figure 4:** Overview of elemental analysis of environmental samples (y-axis), against distance from

404 the mine (x-axis). Plant data=median of all samples collected (leaves, twigs and branches).

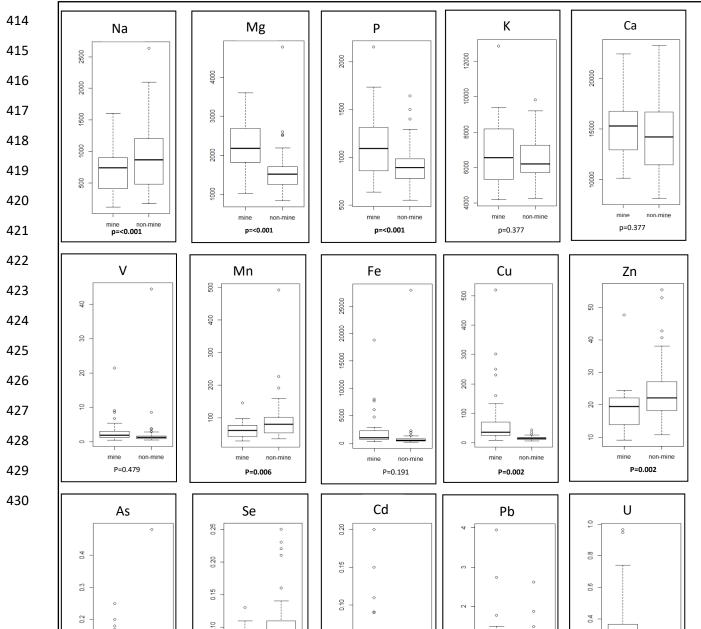
	Soil		Water		Plants/ leaves	
Element	P-value	Adjusted P-value	P-value	Adjusted P-value	P-value	Adjusted P-value
Са	0.05	0.043	0.42	0.038	0.336	0.360
l ratio	5.84		0.65		0.926	
number	2		1		1	
Р	<.0001	0.000	0.03	0.060	<.0001	0.005
l ratio	40.27		4.82		19.52	
number	4		1		4	
Mg	<.0001	0.000	0.02	0.075	<.0001	0.005
l ratio	31.88		5.42		68.08	
number	4		1		2	
Na	0.04	0.050	0.01	0.525	0.5317	0.532
l ratio	4.44		6.05		1.26	
number	1		1		2	
К	0.04	0.043	0.01	0.038	0.33	0.360
l ratio	4.44		7.88		0.93	
number	1		1		1	
V	0.002	0.003	0.31	0.423	0.10	0.136
l ratio	9.18		1.02		4.60	
number	1		1		2	
Mn	0.001	0.002	0.60	0.692	<0.0001	0.020
l ratio	10.47		1.86		6.9662	
number	1		3		1	
Fe	0.0003	0.001	<.0001	0.000	0.03	0.050
l ratio	12.94		42.50		6.77	
number	1		1		2	
Cu	0.009	0.012	0.01	0.038	0.01	0.021
l ratio	9.42		8.78		8.68	
number	2		2		2	
Zn	0.02	0.025	0.96	0.960	0.003	0.009
l ratio	7.96		0.00		8.78	
number	1		2		1	
As	<.0001	0.000	0.16	0.267	0.30	0.360
l ratio	47.96		1.95		3.66	
number	2		1		3	

Se	0.00	0.008	0.11	0.206	0.02	0.038
l ratio	9.60		2.51		5.65	
number	1		1		1	
Cd	<.0001	0.000	0.08	0.171	<0.0001	<0.0001
l ratio	51.20		0.08		25.54455	
number	2		1		1	
Pb	<.0001	0.000	0.77	0.825	0.04	0.060
l ratio	114.46		0.08		6.24	
number	1		1		2	
U	0.002	0.003	0.26	0.390	0.0027	0.009
l ratio	14.44		1.28		11.84	
number	3		1		2	

- **Table 3:** Results of linear mixed model to show significant differences in soil, water and plant (leaf)
- 408 concentrations as distance from the mine increased. P-value (<0.05) and adjusted P-values to control</li>
   409 the false discovery rate (FDR) reported.

- 412 Biological data





**Figure 5:** Elemental analysis data (y-axis, mg/kg) for faecal samples. Box plots show median, Q2, Q3,

- 441 max and min. Outliers are defined as 1.5\*IQR. Adjusted P-values are reported to control for false
- 442 discovery (p<0.05). For mine samples n =37, non-mine n=57.

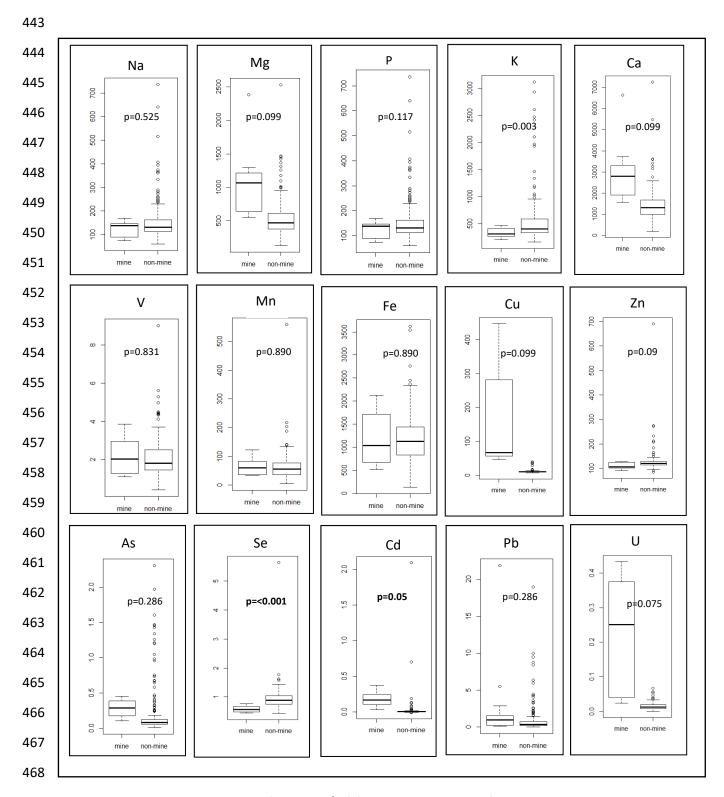


Figure 6: Elemental analysis data (y-axis mg/kg) from tail hair samples from mine and non-mine
elephants, y axis=element concentration (mg/kg). Box plots show median, Q2, Q3, max and min.
Outliers are defined as 1.5\*IQR. Adjusted P-values are reported to control for false discovery

472 (p<0.05). For mine samples n=7, for non-mine samples n=200.

473

# 475 4.0 **Discussion**

476	Mineral provision at the PMC was greater than the surrounding areas (Table 3; Fig. 4). Home ranges
477	of the mine elephants were significantly smaller (59% P=0.001) than those in the surrounding areas
478	(Fig. 2; Table 1), suggesting that their resource needs, including minerals, were met within this
479	smaller area, close to the PMC (Tucker et al., 2018). A trade-off is likely whereby elephants consume
480	soil and water (or plants) near the PMC to obtain increased levels of Ca, Mg, P, Cu, Zn and Se but
481	also consume PTEs (Pb, U and V). Selenium and Zn are fertility augmenters, benefiting elephants in
482	early life (Hidiroglou and Knipfel, 1984; Mistry et al., 2012), whereas the effects on fertility from
483	consuming PTE's to toxic levels may take decades to realise, having a lesser effect on total
484	reproductive output (Kincaid, 1999). An evolutionary advantage may be gained in consuming
485	increased micronutrients at the PMC, at the cost of the increased consumption of PTEs. High
486	consumption of macro-minerals (seen in plants) are under homeostatic control within the elephant,
487	and thus the elephant can buffer increased intake (Kincaid, 1999).
488	Biological samples
489	Tail hair reflects up to 18 months residence, whereas faecal material reflects short-term dietary
490	intake (Bencko, 1995; Wittemyer, Cerling and Douglas-Hamilton, 2009). The differences in tissue
491	biomarkers indicated that short-term environmental differences in availability of minerals consumed
492	by the elephants, appeared to be reflected directly by faecal samples. Whereas, the tail hair data
	by the elephants, appeared to be reflected directly by factor samples. Whereas, the tail half data
493	suggested that the elephants moved to obtain required minerals over time, thereby not showing

495	elements in faecal material versus 3 of 15 elements in tail hairs). Such temporal variability must be
496	considered in evaluating the use of biomarkers for assessing nutrient status/habitat quality.
497	This study covers the widest range of minerals and PTE analysis in elephant faeces to date (n=97; Fig.
498	5; Supplementary Information Table 7). In Hwange National Park, Zimbabwe, Mg, Na and K data
499	were similar to concentrations found in this study (Holdø, Dudley and McDowell, 2002). However, in
500	this study, faecal Ca concentrations, both from mine and non-mine samples were substantially larger
501	than reported by Holdø et al. (2002) with the minimum and maximum level in this study being 8,100
502	and 23,100 mg/kg DM, respectively, versus 920 and 12,000 mg/kg DM. Additionally, in the APNR,
503	Greyling (2004) reported similar P levels in faecal samples (median 1100 versus 990 mg/kg DM in
504	this study). Faecal samples reflect Ca intake (Sach et al., 2020 unpublished data), therefore increased
505	Ca levels found in in this study could be attributed to increased intake.
506	Faecal samples may not represent a specific location or plant consumed; elephants have a total gut
507	transit time of 11–46 h (Clauss <i>et al.</i> , 2003) and walk over 22 km daily (Thomas, Holland and Minot,
508	2012). Faecal samples were a reliable indicator of Ca, P, Se, Cu and As intake (Sach et al., 2020
509	unpublished data) and thus a proxy for elemental status. Significantly greater levels of faecal P, Cu
510	and As were seen in mine samples compared to non-mine samples, indicating that intake of these
511	elements were greater in mine versus non mine. This is also seen in soil, in Cu in water and P and Cu
512	in plants (leaves), supporting this increased intake. Additionally, elephants are frequently
513	documented to participate in geophagy (Holdø, Dudley and McDowell, 2002), and although not

specifically reported at PMC, could be in part obtaining these increased elemental levels viageophagy.

516	This study provides the largest multi-element dataset on mineral and PTE analysis data in elephant
517	tail hair (Fig. 6; Supplementary Information Table 6). Hair analysis is routinely used in humans and
518	livestock to assess Se and As levels (Bencko, 1995; Middleton et al., 2016). Duer, Tomasi and
519	Abramson, (2016) analysed an elephant tail hair from a deceased healthy individual from Tsavo
520	National Park, Kenya and reported 11 elements for which concentrations were comparable to the
521	non-mine elephants within this study. However, levels of Mg, Ca, Mn, Cu and Pb in the mine
522	elephant tail hairs were considerably greater than those reported by Duer, Tomasi and Abramson
523	(2016).
524	Environmental samples
525	This study agrees with work reported by Ramahlo (2013) within the Phalaborwa region, regarding

the impact of mining on soil at surrounding farms, where P, As and Pb levels in soil decreased with
increasing distance from the mine. African soils contain high levels of Fe (Siyame *et al.*, 2013), and
thus a significant difference between mine/ non-mine faecal or tail hair samples may not be seen
(Fig. 5 and 6), as all animals may be consuming to excess. Studies demonstrated elephants
selectively drank water with elevated mineral levels; notably Na, iodine (I), sulphur (S), Zn, Ca, Mg,
Mn and Fe (Weir, 1972; Sienne, Buchwald and Wittemyer, 2014). Additionally, elephants may spend
more time at the PMC site during the dry season, either due to mineral deficiencies in natural forage

being heightened in the dry season, or simply for increased water availability within the PMC area(Purdon and van Aarde, 2017).

535	Elemental analysis of plant samples do not always reflect soil due to a variety of factors including soil
536	pH, organic matter and differences in the capacity of individual plant species to accumulate certain
537	elements (Bowell and Ansah, 1994; Maskall and Thornton, 1996). In the Sabi Sands Reserve, South
538	Africa, grasses were analysed from soils of higher mineral levels, yet they accumulated less minerals
539	compared to grasses from soils where the minerals were lower (Ben-Shahar & Coe, 1992), due to
540	differences in soil-to-plant transfer between plant species and the effect of the local micro-climate.
541	Similarly, this variation in soil-to-plant transfer was reflected in this study for Ca, Na, K, V, As and Pb
542	(Fig. 4; Table 3). These elements decreased significantly in soil with distance from the mine, although
543	plants did not follow the same trend. The igneous Phalaborwa apatite would be expected to have
544	low reactivity (i.e. low solubility), hence the elements in the soil may be less available for uptake by
545	plants (Appleton, 2002). Finally high soil Fe, typical of African soils, could also reduce the availability
546	of P to animals via plants, thus the increase in soil P may not be reflected within mine tail hair
547	samples (Fordyce, Masara and Appleton, 1996).

## 548 5.0 **Conclusion**

The home ranges of the collared PMC elephants are considerably smaller (59% smaller) than elephants in surrounding areas, implying that their resource needs are being met within this relatively compact area. Drivers for elephant movement are multifactorial, yet evidence suggests that these key differences in the geochemistry of the mine compared to the surrounding areas, could act as a driver for elephant movement, resulting in reduced home range size compared to other elephants within this geographical region. Mineral provision to the elephants at the PMC is

significantly greater than in surrounding areas, seen most significantly in the soil where all
investigated mineral and PTE levels decreased significantly with increasing distance from the mine.
These differences suggest that elephants are attracted to this micronutrient hotspot at the PMC, to
obtain required minerals.

559 The increased mineral provision and trade off of increased PTE levels were reflected in biological

560 samples of elephant tail hair and faeces. Baseline levels of key minerals and PTEs in African elephant

tail hair and faeces were established from this work. The methods described within this natural

562 experiment to investigate how environmental geochemistry influences elephant home range size

and potentially movement, facilitates the consideration of intervention to reduce associated HECs at

- the PMC. This approach could be applied to similar situations, with wider benefits to a variety of
- 565 stakeholders, informing broader conservation efforts.

## 566 6.0 CRediT author statement

- 567 FS, MW, LY, SLE, ED, MH and AS conceptualization and methodology; FS and AB data curation; FS, MH,
- 568 AG, EH and PB investigation; MW, MH, AS and PB resources; FS and RML formal analysis; FS writing
- 569 original draft. All authors contributed critically to drafts and final approval for publication.

## 570 7.0 Declaration of Interest

- 571 Ellen Dierenfeld is employed by Ellen Dierenfeld Consulting LLC
- 572 No other declarations of interest

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