

Mechanical, chemical, structural, and radiological changes in pigeon bone, associated with the dietary intake of nickel recovery slag as a grit source

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Introduction

Slag tailings containing high concentrations of base metals from over 130 years of continuous nickel smelting operations in the Sudbury, Ontario basin, have become ubiquitous. This material rich in heavy metals upon ingestion, has the potential to effect changes in bony structures of biological systems. Most birds depend on a continuous uptake of pebble-sized grit to assist in the grinding functions of the gizzard. In this work, we analyse the effects of dietary slag ingestion as a grit source, on seven different quantitative, and two qualitative parameters of the tibio-tarsal bones in pigeons (*Columbia Livia Domesticus*). The specimens were divided into a control group ($n = 9$) provided normal grit, and an experimental group ($n = 9$) fed slag-based grit, both for a period of 1 year. Their tibio-tarsal bones were then harvested for analysis. Quantitative analytical methods included measurement of compact bone thickness (TH), conventional density measurements (CD), bone mineral density (BMD) measurements using Dual Energy X-Ray Absorptiometry (DEXA), Ca and Fe concentrations using mass spectrometry, and the determination of Young's Moduli (YM) and breaking strength (BS) using a universal testing machine (UTM). Qualitative microscopy studies - both optical and electron were also carried out for both treatment groups.

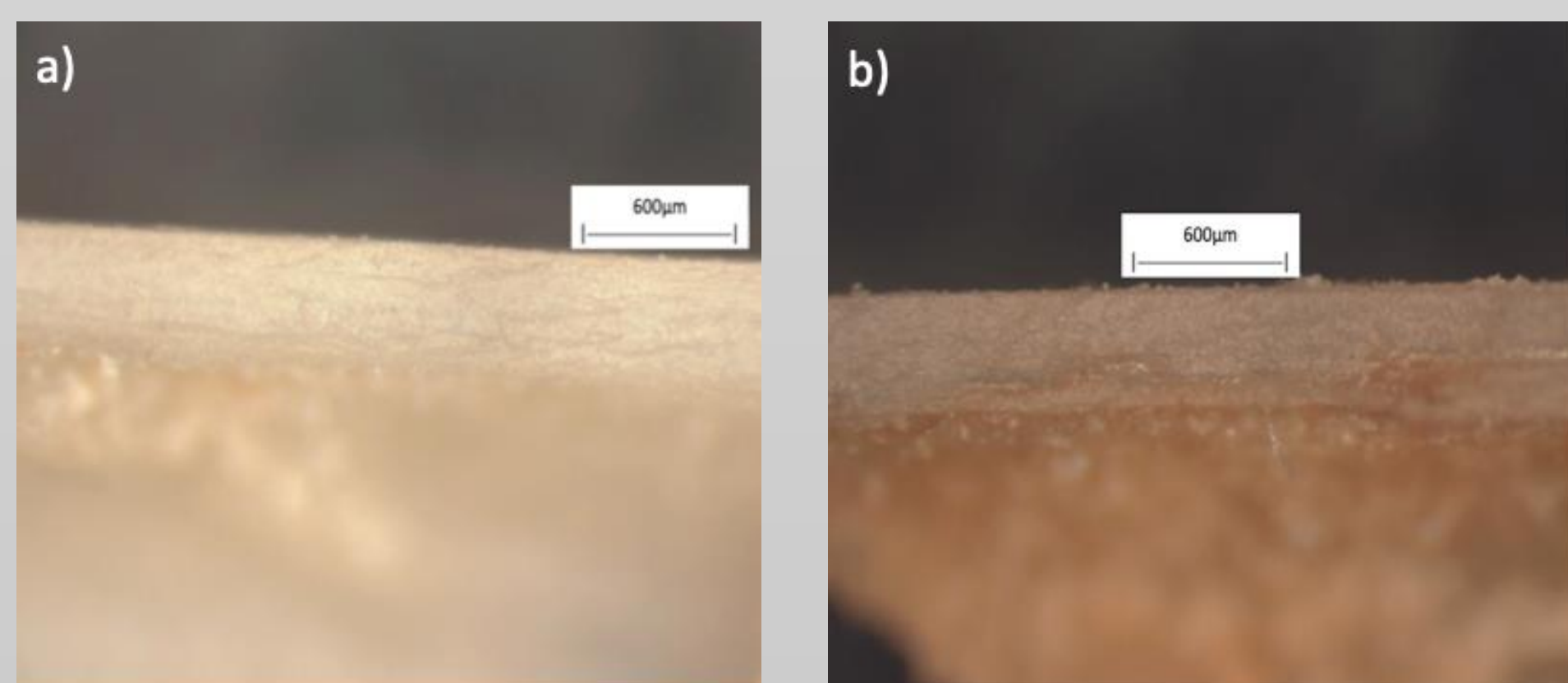


Figure 1: Optical micrographs of cortical tibio-tarsi of a) control group sample (normal grit), and b) experimental group sample (slag grit). Note the thinning of the cortical thickness, as well as the increased irregularity of the periosteal and endosteal surfaces in the experimental sample.

Materials and Methods

The specimens were sacrificed after one year of controlled feeding and the tibio-tarsal bones were excised from the carcasses.

A calliper was used to measure the outer and inner diameters of cortical diaphyses. Compact bone thickness (TH) for each sample was calculated by subtracting the inner from the outer diameters.

Conventional density (CD) measurements of the samples were made using a sealed scale for mass determination. Sample volumes were obtained by water displacement, and the mass to volume ratios computed.

Bone mineral density (BMD) measurements involved the use of a dual energy x-ray (DEXA) beam, directed at the samples to infer a ratio of high atomic number material (bone) to low atomic number material (soft tissue). Samples from the control and experimental groups were subjected to DEXA scanning.

Young's Modulus (YM) is a physical parameter which quantifies the elasticity of a solid. It is the ratio of the solid's stress to its strain, under an applied compressive (or tensile) load. Breaking strength (BS) is the minimum axial force required to induce mechanical failure of the solid. It is common practice to acquire YM and BS data simultaneously, using a Universal Testing Machine (UTM). The machine automatically produces a value of YM (in MPa), and a breaking strength (BS) value (in N). Left side tibio-tarsal bones from the control and experimental groups were subjected to destructive compressional YM and BS measurements.

Mass spectrometry is a high-precision and sensitivity method for elemental analysis, based on the electromagnetic separation of atomic nuclei of different masses. Tibio-tarsal bones from samples in both groups were subjected to mass spectrometry with special emphasis on Ca and Fe measurements, reported in units of parts per million (ppm).

Additional qualitative analyses of samples from the two groups were undertaken, in particular electron and optical microscopy studies, followed by energy dispersive spectroscopy (EDS). Electron micrographs of mid-diaphysis cortical tibio-tarsi of from the control and experimental groups (both $n = 1$), were obtained. Image acquisition parameters were: accelerating voltage 30 kV, beam current 1.0×10^{-11} A. These were followed up with conventional microscopy of the same samples, with particular attention to the cortical, mid-diaphysis.

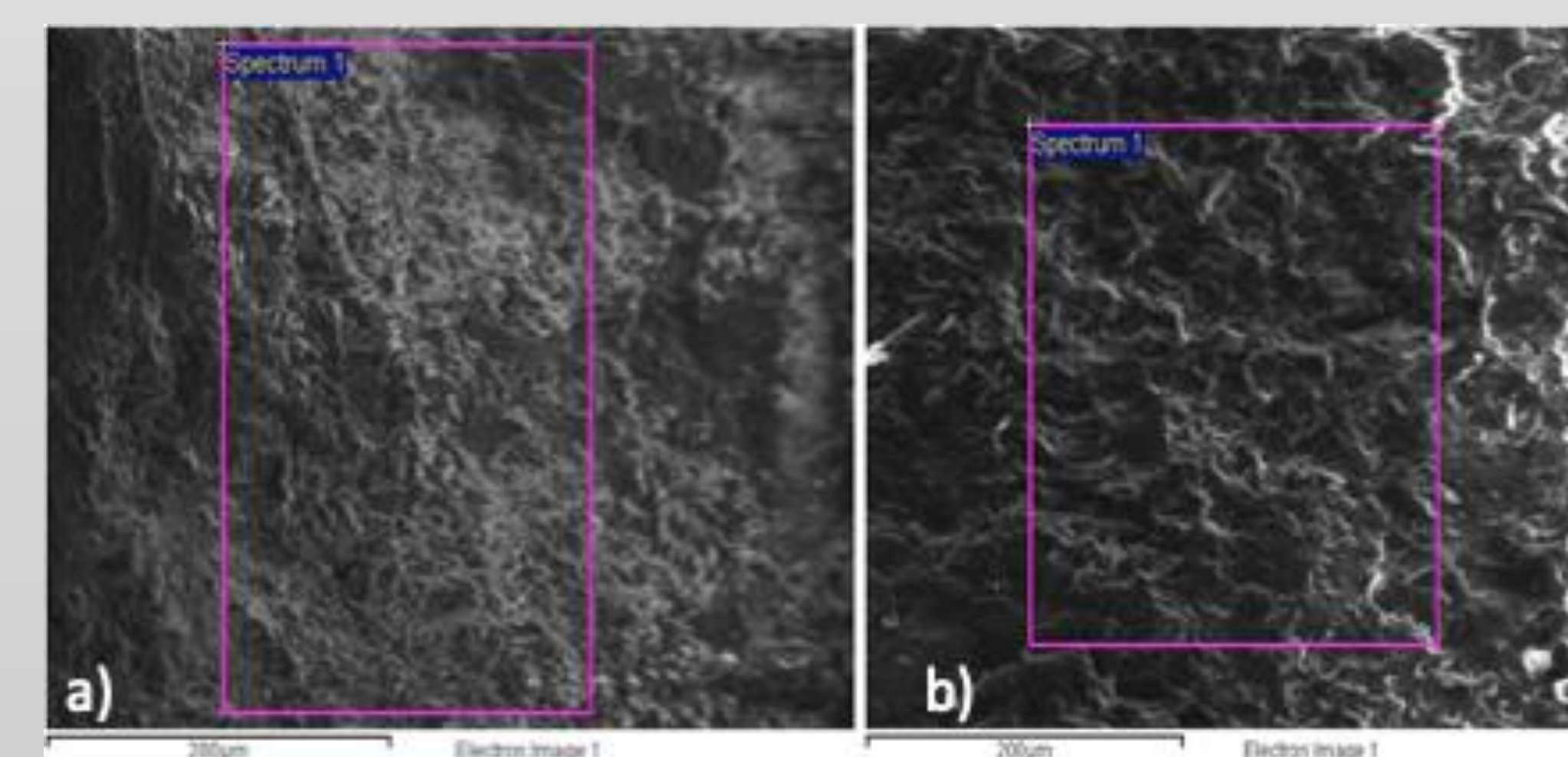


Figure 2 (a and b): Electron micrographs of cortical tibio-tarsus of a) the control group (normal grit), and b) experimental group (slag grit). Note structural changes in the experimental sample, consistent with resorption, woven formations, and fibroplasia.

Results

	Control	Experimental	p (one-tail)
Th (mm)	0.61 ± 0.07 (n=6)	0.48 ± 0.03 (n=6)	0.005
CD (g/cm^3)	1.9785 ± 0.1948 (n=9)	1.7698 ± 0.2223 (n=9)	0.0251
BMD (g/cm^2)	0.152 ± 0.005 (n=6)	0.143 ± 0.005 (n=6)	0.0089
Ca (ppm)	82621.8 ± 11046.7 (n=9)	70912.8 ± 11952.7 (n=10)	0.0272
Fe (ppm)	80.874 ± 35.274 (n=9)	242.76 ± 180.59 (n=10)	0.0127
BS (N)	451.57 ± 133.78 (n=7)	328.50 ± 139.62 (n=8)	0.0511
YM (MPa)	3483.6 ± 1853.7 (n=7)	2835.9 ± 879.6 (n=8)	0.0662

Table 1: Results for the seven measured parameters, for both the control and experimental groups, with relevant "n" and "p" values. Note that six of seven parameters showed a statistically significant degradation ($p \leq 0.05$).

Conclusions

In summary, based on the measurements performed in this work, we conclude that the dietary intake of nickel recovery slag as a grit source, is associated with measurable mechanical, chemical, and radiological changes - as well as detectable qualitative structural changes - in the tibio-tarsi of *Columbia Livia Domesticus* pigeons.

References

Parker, G. H., and Van Der Kraak, G., Use of Metal-Rich Slag as a Source of Grit and Its Effects on Pigeon Health and Fitness. *J. Env. Sci. Pollut. Res.* 4(2) (2018) pp 256-267.

Cork, S.C., Iron storage disease in birds. *Avian. Pathol.* 29 (2000) pp 7-12.

Zhang, B. and Coon, C.N., The relationship of various tibia bone measurements in hens. *Poultry Science* 76.12 (1997): pp 1698-1701.

"The t-Test" in *Statistics for the Life Sciences* 5th Ed. by M. L. Samuels, J. A. Witmer and A. Schaffner, Pearson, 2016.