

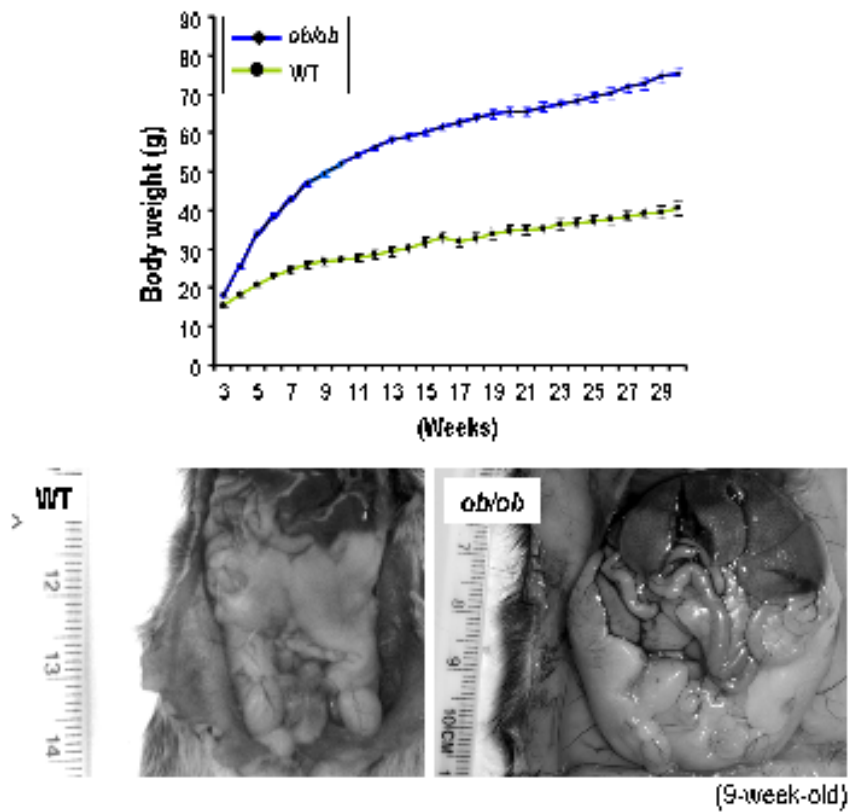
Genetic induction of phosphate toxicity significantly reduces the survival of hypercholesterolemic obese mice

Mutsuko Ohnishi, Shigeko Kato, M. Shawkat Razzaque

Department of Oral Medicine, Infection and Immunity, Harvard School of Dental
Medicine, Boston, MA 02115, USA

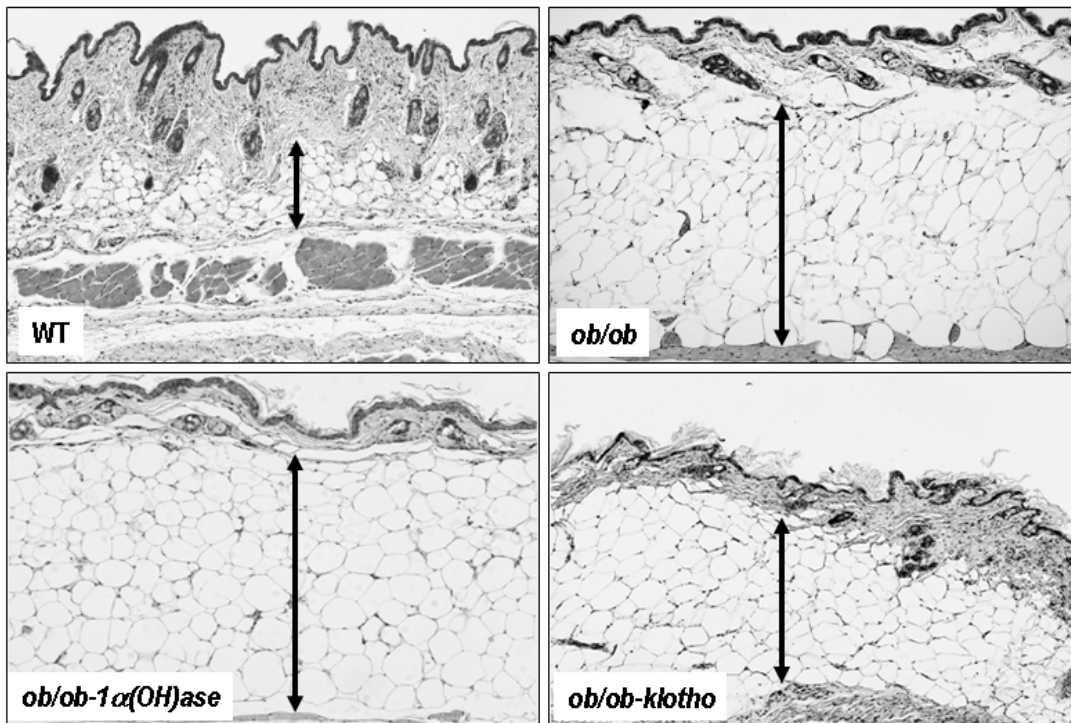
Suppl. Figure 1.

Gross phenotype of the *ob/ob* mice. Body weight chart of wild-type (WT) and *ob/ob* mice. The *ob/ob* mice were significantly larger compared to controls (**upper panel**). By 9 weeks, a massive accumulation of abdominal fat was observed in the *ob/ob* mice compared to the WT mice (**lower panel**) [23].



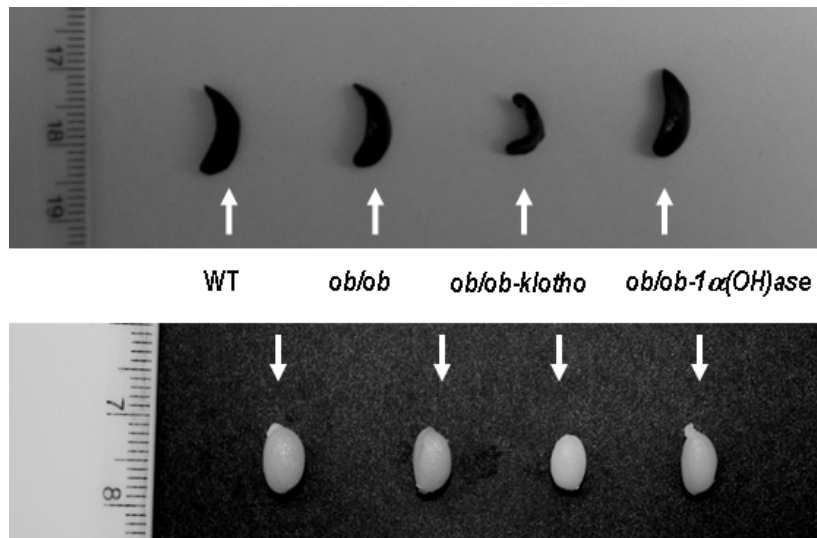
Suppl. Figure 2.

Skin histology. Compared to the skin of the wild-type (WT) mice, there was an increase in the subcutaneous fat tissue layer present in all three mutant mice (*ob/ob*, *ob/ob-klotho*^{-/-} and *ob/ob-1 α (OH)ase*^{-/-}); however, the *ob/ob-klotho*^{-/-} mice had relatively less fat tissue (represented as a bar) (10x magnification).



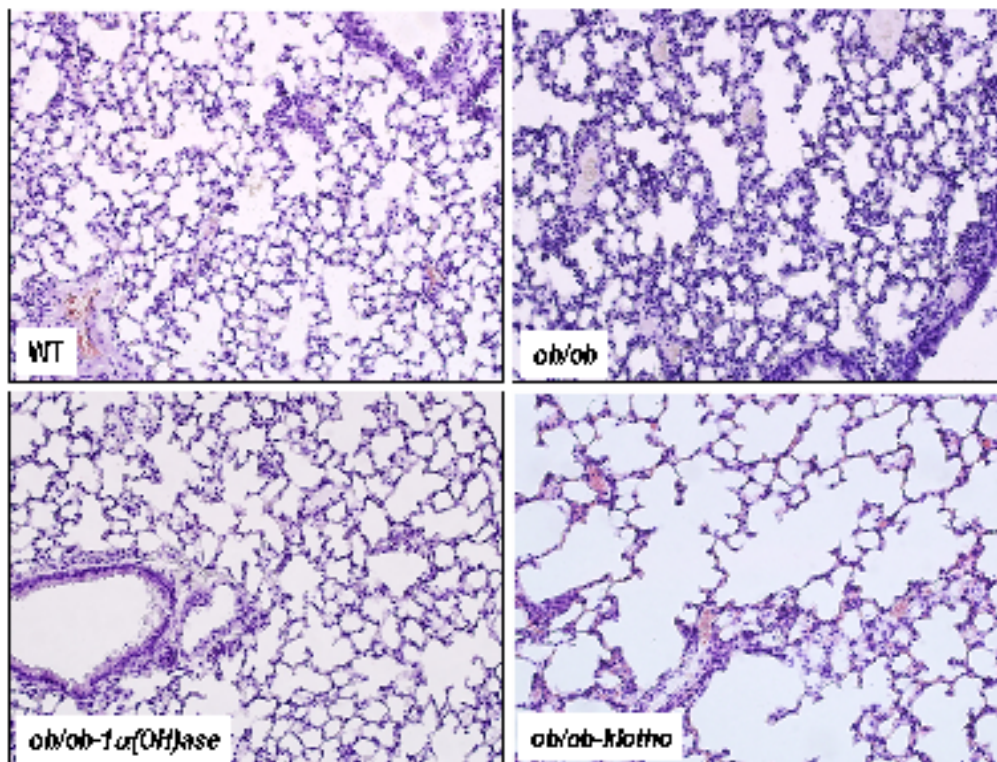
Suppl. Figure 3.

Gross appearance of the spleens and testes in the mouse models. The testes and spleens were collected from wild-type (WT), *ob/ob*, *ob/ob-klotho*^{-/-} and *ob/ob-1 α (OH)ase*^{-/-} mice. Compared to the *ob/ob* and *ob/ob-1 α (OH)ase*^{-/-} mice, the spleens were smaller in hyperphosphatemic *ob/ob-klotho*^{-/-} mice. The animals were age-matched (11 weeks; **upper panel**). The testes from hyperphosphatemic *ob/ob-klotho*^{-/-} mice were smaller than those from the *ob/ob* or *ob/ob-1 α (OH)ase*^{-/-} mice (**lower panel**).



Suppl. Figure 4.

Lung tissue histology. Hematoxylin and eosin-stained sections of the lung tissues from 6-week-old wild-type (WT), *ob/ob*, *ob/ob-klotho*^{-/-} and *ob/ob-1 α (OH)ase*^{-/-} mice. Compared to the WT mice, there was marked expansion of alveolar spaces (emphysema) in the hyperphosphatemic *ob/ob-klotho*^{-/-} mice. Similar changes were not observed in the *ob/ob-1 α (OH)ase*^{-/-} mice (20x magnification).



Suppl. Figure 5.

Survival plot. Survival analysis of wild-type (WT) (n=10), *ob/ob* (n=10), *ob/ob-klotho*^{-/-} (n=11) and *ob/ob-1 α (OH)ase*^{-/-} (n=10) mice. The survival time of hyperphosphatemic *ob/ob-klotho*^{-/-} mice was reduced compared to the *ob/ob* or *ob/ob-1 α (OH)ase*^{-/-} mice. All the *ob/ob-klotho*^{-/-} mice died by 20 weeks, while none of the WT, *ob/ob* or *ob/ob-1 α (OH)ase*^{-/-} mice died within the 25 week observation period [23].

