Knockout mouse lines presenting with welfare issues affecting their survival (abnormal survival [MP:0010769]) are processed through a bespoke sub-pipeline known as the "sick mouse procedure" (SMP) to maximise information collected on that mouse line. Matched wild-type controls are also processed to identify phenotypic abnormalities arising from the targeted allele.

Wnt16$^{tm2b(EUCOMM)Wtsi}$

Wingless-related MMTV integration site 16

Genetic Background: C57BL/6N;C57BL/6NTac

ES Cell Clone: EPD0561_6_A02

Tm2a construct:

Tm2b construct:

Affected genotypes

Homozygous (*Wnt16*$^{tm2b(EUCOMM)Wtsi/tm2b(EUCOMM)Wtsi}$).

Alternative breeding strategy

Following initial welfare observations, wild-type x heterozygous mating strategy was employed to complete phenotyping work in standard pipeline using heterozygous mice only.

Heterozygous mice showed no significant phenotypic findings on the primary screen.

Welfare observations

Homozygous mice exhibit:

- Increased incidence of bone fractures in tibia and fibula after 6 weeks of age: 5/6 (83%).

Homozygous Viability:

All genotyped mice from het x het intercross considered. When at least 28 mice are available, viability at p14 is calculated. *[>13% = Homozygous viable; >0% and <13% = Sub-viable; 0% = Lethal]*

- **Viable** : 20 Homs / 90 Total = 22.22%

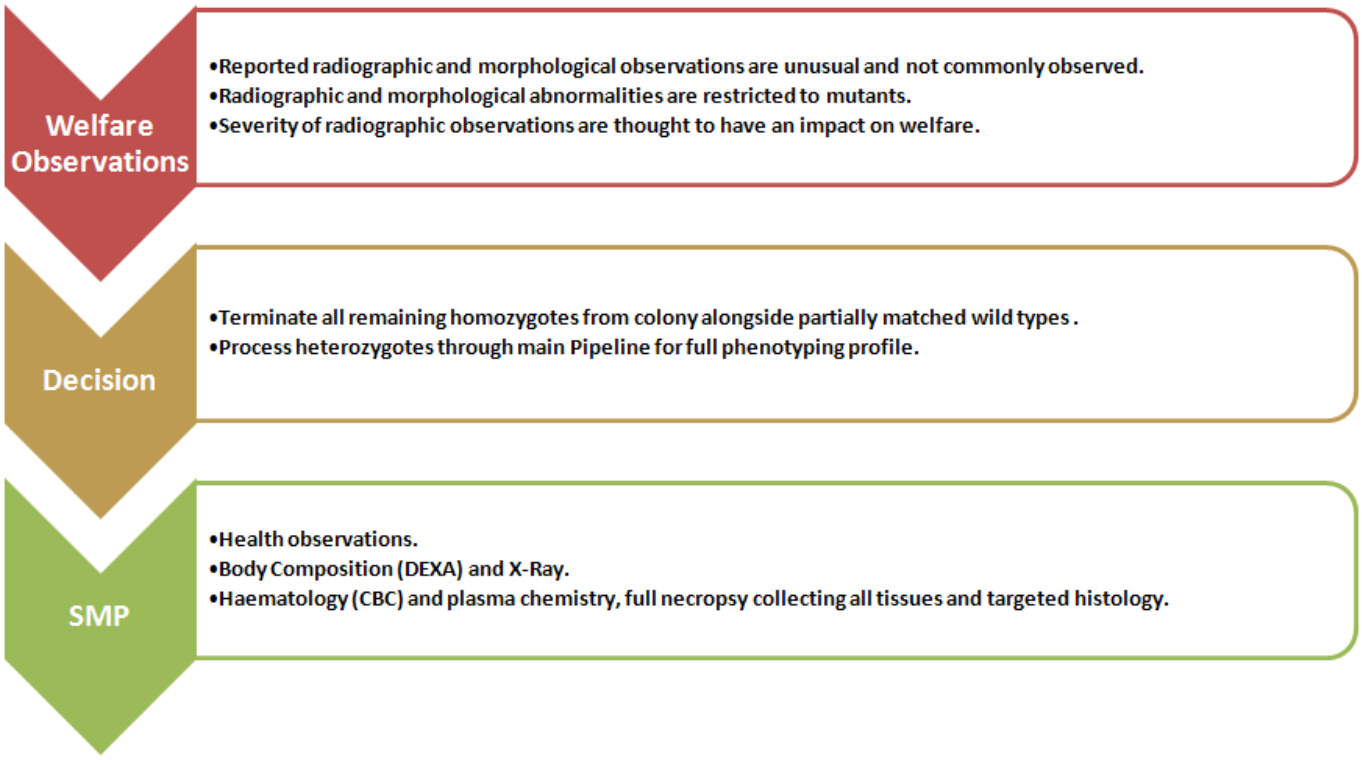
Sick Mouse Procedure (SMP)

Initial welfare observations were reported in homozygotes during the entry to the phenotyping pipeline (4 weeks).

Initial welfare observations in all homozygotes were deemed to be critical and in conjunction with a literature review* the decision was made to initiate SMP (see schematic below). 8 male and 6 female homozygotes were processed alongside 5 male and 3 female matched wild-types. No further homozygotes were phenotyped due to the aforementioned alternative breeding strategy employed to reduce further welfare implications.

*Zheng H-F, Tobias JH, Duncan E, Evans DM, Eriksson J, et al. (2012) WNT16 Influences Bone Mineral Density, Cortical Bone Thickness, Bone Strength, and Osteoporotic Fracture Risk. PLoS Genet 8(7): e1002745. doi:10.1371/journal.pgen.1002745

Schematic Outline of Bespoke SMP Pipeline



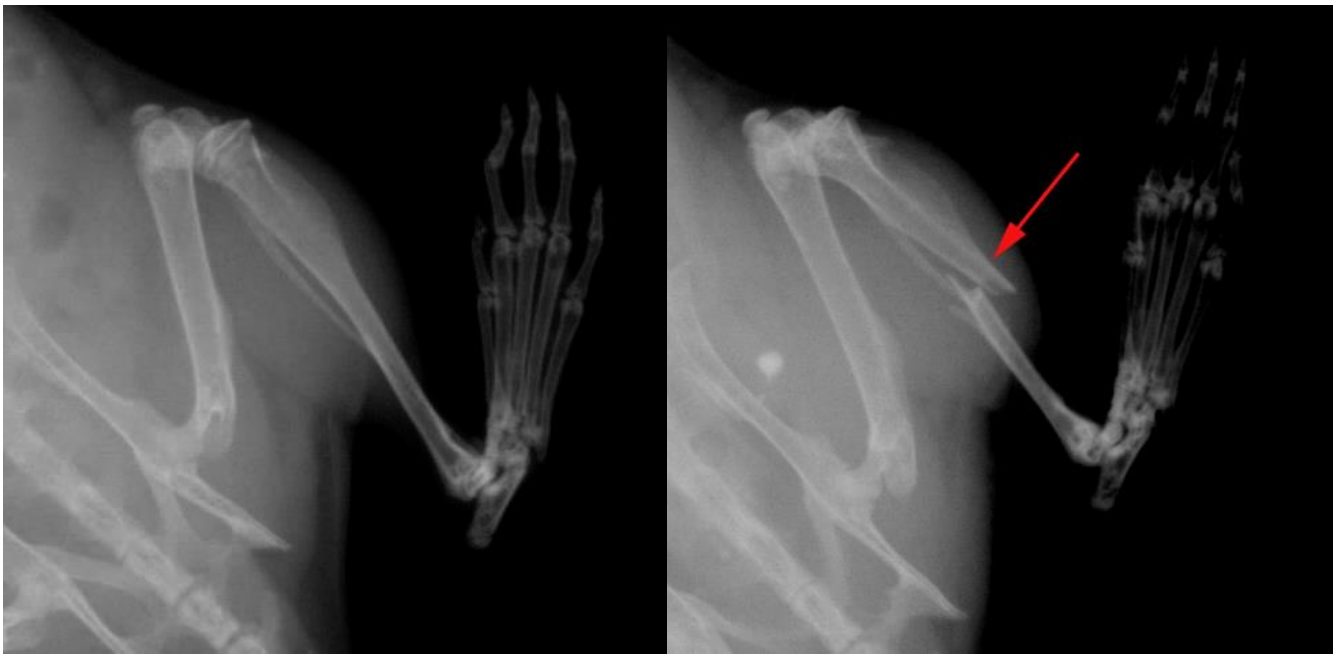
Phenotyping Heat Map

Colony Prefix	Allele Name	Genotype	Weight Curves	Body Composition (DEXA)	X-ray Imaging	Plasma Chemistry	Haematology (CBC)	Tissue Biobank
MUBC	*Wnt16* $^{tm2b(EUCOMM)Wtsi}$	Homozygous						

Significant / Interesting
Not significant
Resources available

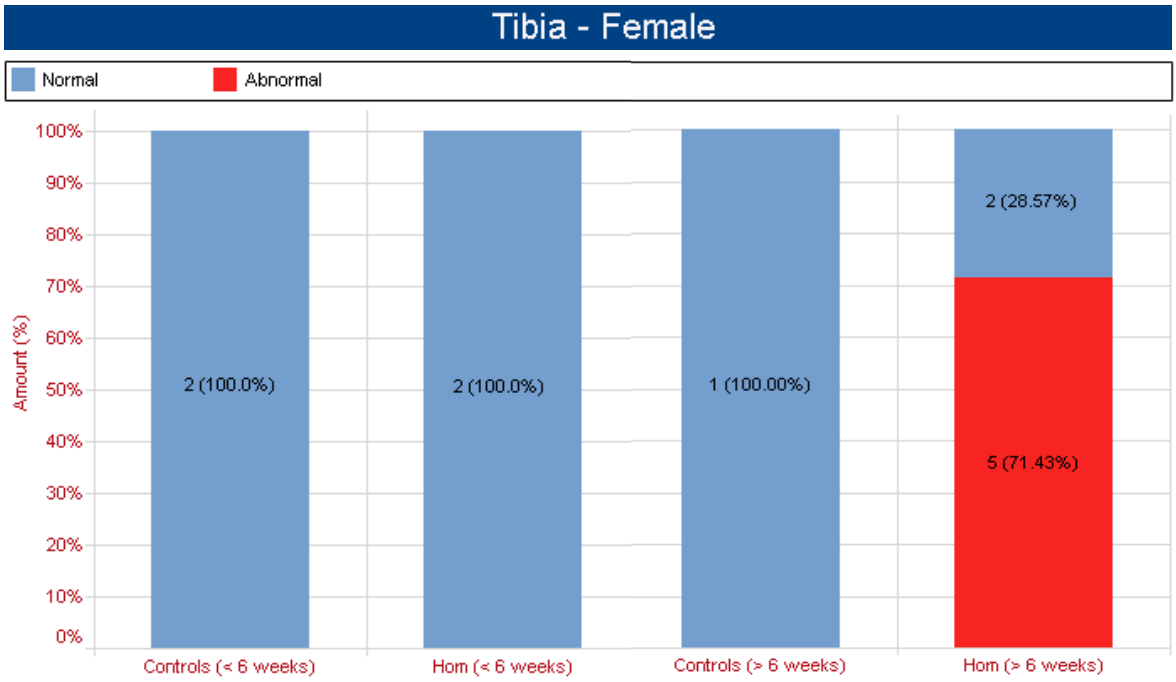
Phenotyping data of interest (significant changes)

In life phenotyping

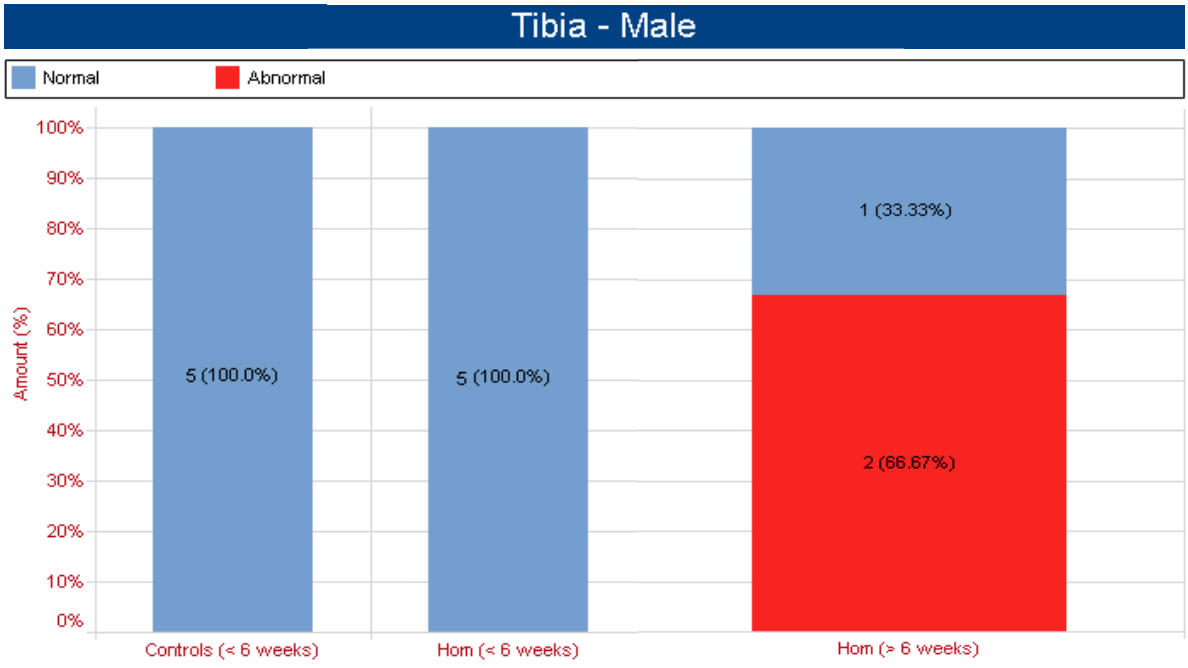


a b

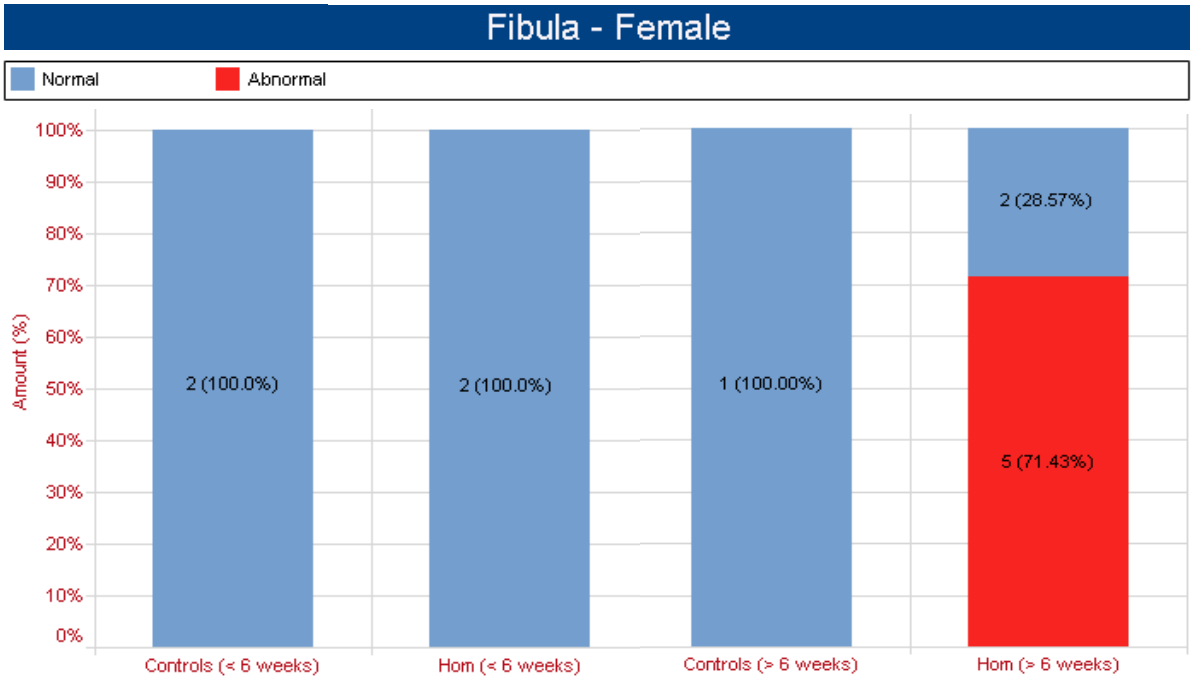
Figure 1. Dorsoventral radiograph of (a) wild-type and (b) homozygous mouse right hind limb displaying fractures of tibia and fibula (abnormal tibia morphology [MP:0000558] and abnormal fibula morphology [MP:0002187])



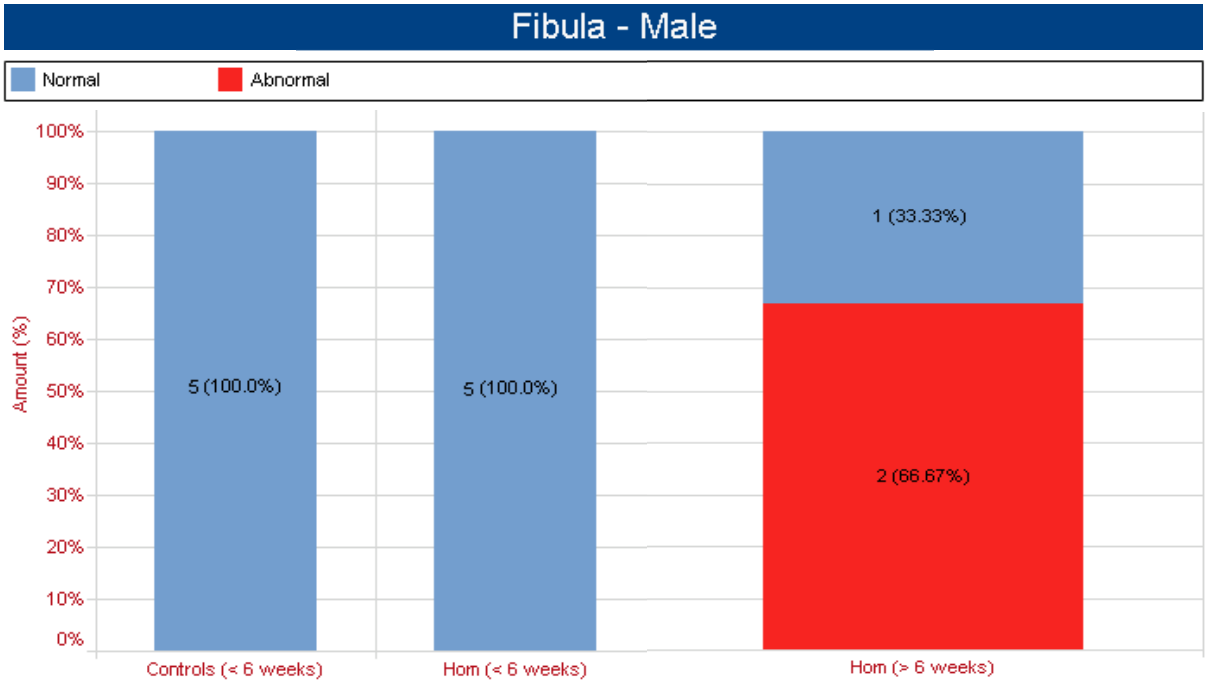
Females – Abnormal Tibia Morphology [MP:0000558]



Males – Abnormal Tibia Morphology [MP:0000558]



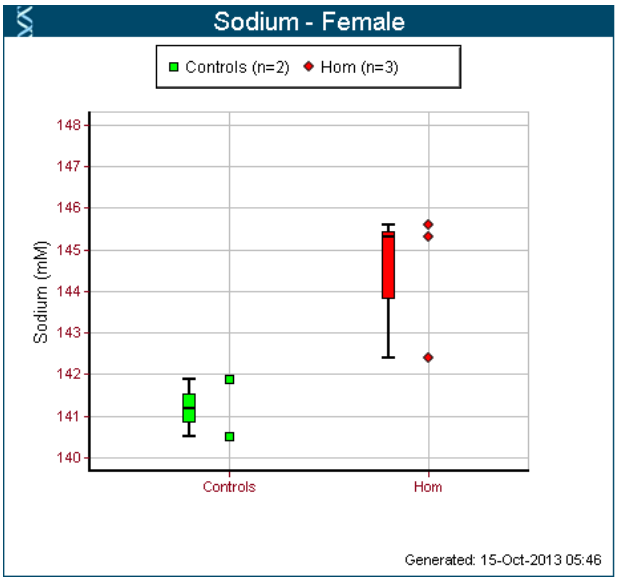
Females – Abnormal Fibula Morphology [MP:0002187]

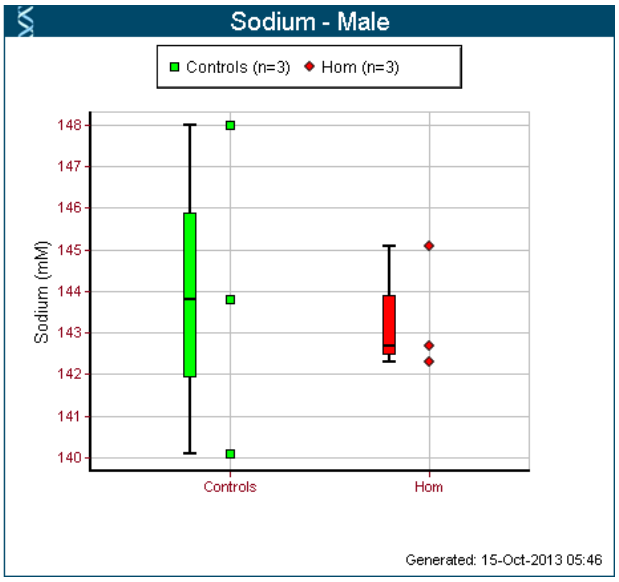


Males – Abnormal Fibula Morphology [MP:0002187]

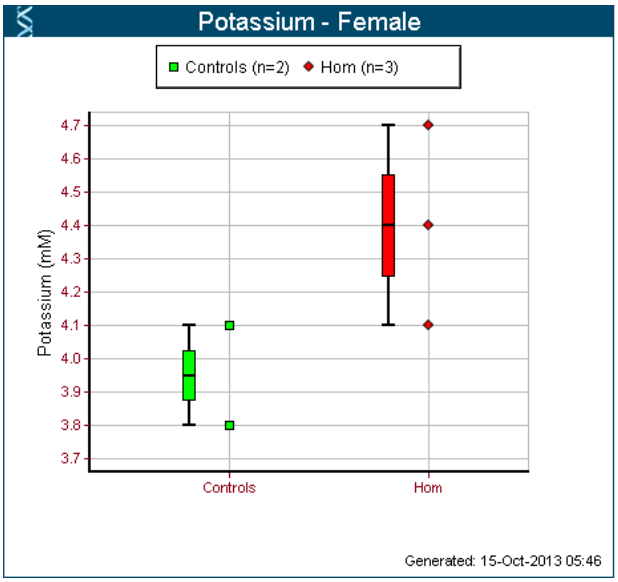
Plasma Chemistry

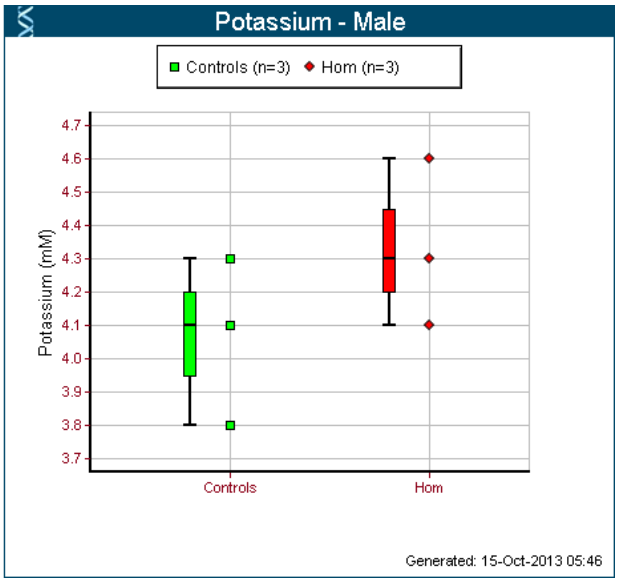
NB: Results are for blood collected from mice below 6 weeks of age.



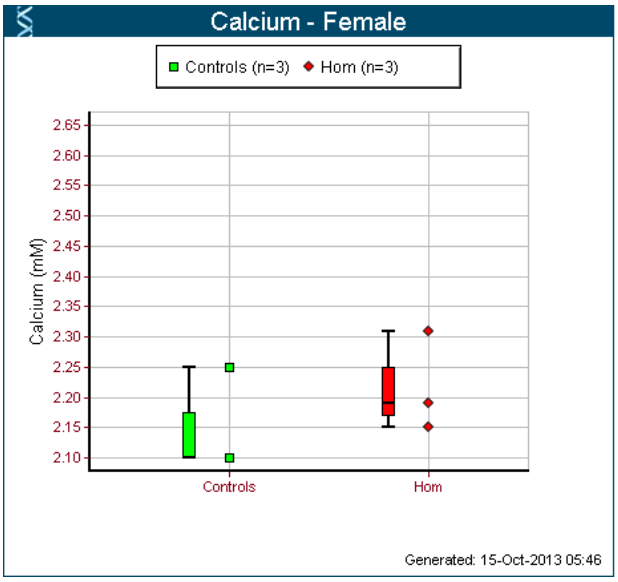


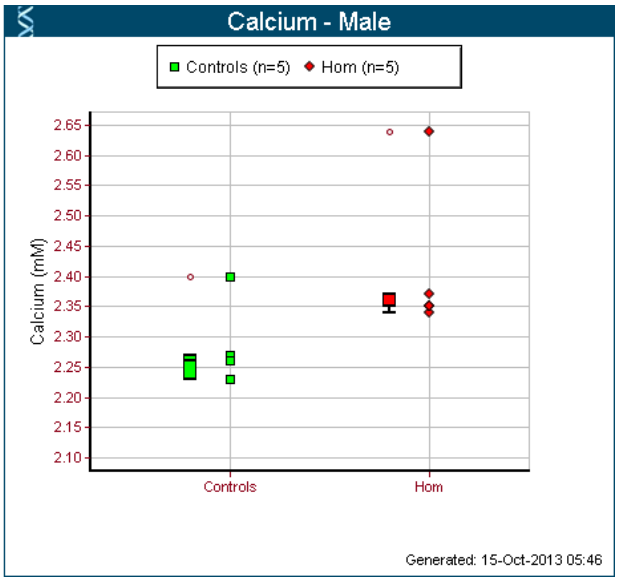
Females only - Increased circulating sodium level [MP:0005633]





Females only - Increased circulating potassium level [MP:0005627]





Males only - Hypercalcemia [MP:0000194]