## **Supplementary Material**

## Mice

All sampling localities are listed in Table S1. Brothers within the fraternal pairs were individually marked by fur clipping and housed together in standard polycarbonate cages ( $43 \times 30 \times 22$  cm) under a 14-h light/10-h dark regime at a room temperature 20 °C. Pelleted food (ST1, VELAZ, Prague, Czech Republic) and water were available *ad libitum*. The two subspecies were kept in separate rooms and in the absence of females. The breeding facility of the Institute of Vertebrate Biology in Studenec has been licensed for maintaining small mammals according to Czech law. Animals were handled by authorised persons only, and adequate measures were taken to minimise the pain or discomfort of the experimental subjects.

**Table S1**: List of sampling localities of *M. m. musculus* and *M. m. domesticus* mice used as the parental generation for the experimental animals under study (see also Fig. 1). *N*: number of pairs examined.

M. m. domesticus			M. m. musculus		
Locality	Coordinates	Ν	Locality	Coordinates	Ν
Benk	50° 11' N, 11° 52' E	2	Buškovice	50° 13' N, 13° 22' E	2
Lehsten	50° 07' N, 11° 55' E	2	Mirotice	50° 07' N, 13° 00' E	2
Neudorf	50° 02' N, 11° 39' E	2	Přílezy	50° 06' N, 12° 57' E	2
Ottmannsreuth	49° 53' N, 11° 37' E	2	Úhošťany	50° 21' N, 13° 16' E	2
Straas	50° 11' N, 11° 46' E	2	Vrbice	50° 09' N, 13° 14' E	2

## Methods

All mice were checked at five-day intervals starting from the age of 20 days; after about 50 days of age, the intervals were extended to 5–7 days. During the controls, samples of fresh urine were collected. The animals usually spontaneously urinated when handled. If a sufficient sample was not obtained, a gentle abdominal massage was applied, or the animal was kept individually in a clean, dry cage for a while. After collecting at least 10  $\mu$ l of urine, the samples were stored at -20 °C until processing.

Raw total MUP quantities were normalised using a dilution coefficient based on creatinine concentrations as reliable indicators of the volume of liquid filtered through the kidney. The dilution coefficient was calculated as  $C_{creat}/C_{ref}$ , where  $C_{creat}$  is the creatinine concentration in each sample, and  $C_{ref}$  is the highest concentration over all measured creatinine concentrations.

The following generalised additive models were considered to fit the data:

M1: one curve for all males, i.e., differences neither between subspecies nor ranks; M2: two subspecies-specific curves, no difference between ranks within subspecies; M2dd: two curves, one for dominant *domesticus* males and one for all other categories; M2ds: two curves, subordinate *domesticus* males differ from all other males; M2md: two curves, dominant *musculus* males differ; M2ms: two curves, subordinate *musculus* males differ; M3d: one joint curve for *musculus*, two rank-specific curves for *domesticus*; M3m: one joint curve for *domesticus*, two rank-specific curves for *musculus*; M4: distinct curves both for subspecies and ranks. The models were compared using the Akaike information criterion (AIC) and Akaike weights, expressed as

$$W_i = \exp(-0.5 \times \Delta \text{AIC}_i) / \sum_{j=1}^k \exp(-0.5 \times \Delta \text{AIC}_j),$$

where  $W_i$  is the Akaike weight for the *i*-th model,  $\Delta AIC_i$  is the difference between AIC for the *i*-th model and the best-fitting model, and the numerator is the sum over *k* models.

Results



**Figure S1.** Creatinine concentrations in *musculus* and *domesticus* males measured between the 20<sup>th</sup> and 100<sup>th</sup> day of age, fitted with the M2ds model, which best fitted the data (one distinct curve for subordinate *domesticus* males (light blue), one curve for all other males shown in grey).



**Figure S2.** Normalised MUP concentrations (ng/ml) as in Fig. 1, but the *y*-axis is not in log-scale (A). Predicted curves for testosterone levels for the same individuals (B); light, thin curves depict 95% confidence intervals (data adopted from Hiadlovská et al. 2015).