

SUPPLEMENTARY DOCUMENT 1

Seminiferous tubule diameter

1. Analyzed diameter of 125 seminiferous tubules in each testicle (5 tubules in 5 fields of 5 cuts) was measured
2. Images were captured under $\times 100$ magnification
3. ImageJ software for analysis
4. Open the tool file and after that, open the ruler with the same resolution and size of the image captured for calibration
5. Open the image file to be analyzed
6. Open the straight line tool
7. Use the straight line tool, drawing a line from the tunica propria of one pole to the other of the tubule passing through the center of the tubule. The result of the analysis of each testicle was considered to be mean of the 125 diameters measured, expressed in μm .

Seminiferous epithelium height

1. Analyzed diameter of 125 seminiferous tubules in each testicle (5 tubules in 5 fields of 5 cuts) was measured
2. Images were captured under $\times 200$ magnification
3. ImageJ software for analysis
4. Open the tool file and after that, open the ruler with the same resolution and size of the image captured for calibration
5. Open the image file to be analyzed
6. Open the straight line tool
7. Draw three lines (using the same tool used for the diameter) that extend from the tunica propria to the lumen of the seminiferous tubule, excluding spermatozoa. Therefore, here we do not consider the spermatozoa as cells that make up the epithelium seminiferous. The three lines were distributed in such a way that they were equidistant. The average of these three lines was considered as the height of the epithelium of that seminiferous tubule.

Volumetric density protocol

1. 25 fields randomly captured were analyzed for each rat
2. Images were captured under $\times 400$ magnification
3. ImageJ software for analysis
4. Open the image file to be analyzed, and measure its area in pixels by choosing "measure" in the "analyze" bar.
5. Open the plugin "Grid" and insert the measured area of the image and choose grid type: crosses – **Supplementary Figure 2**
6. Open the plugins analyze cell counter. With the tool "cell counter," each structure touched by a point was counted, and its density was determined as a percentage of the analyzed field
7. We quantified the V_v of the tunica propria, seminiferous epithelium, tubular lumen, and intertubular compartment. The sum of the V_v (tunica propria), V_v (seminiferous epithelium), and V_v (tubular lumen) was considered the V_v (tubular compartment)
8. The V_v of the interstitial space was considered as the intertubular compartment
9. For each parameter, the result was expressed as a percentage and calculated by the average of the results of each analyzed image
10. Further, the absolute volume (A_v) of each structures mentioned was calculated by multiplying the testicular volume by each structure's V_v . This parameter was expressed in milliliters. For this, the following formula was used:
$$Y \text{ (analyzed structure) volume} = (V_v [Y]/100) \times \text{testicular volume}$$