MicroCT reveals how male stag beetles can pinch forcefully with ungainly jaws

J. Goyens¹,², J. Dirckx², P. Aerts¹,³

¹ University of Antwerp, Laboratory of Functional Morphology, Universiteitsplein 1, B-2610 Antwerp, Belgium
² University of Antwerp, Laboratory of BioMedical Physics, Groenenborgerlaan 171, B-2020 Antwerp, Belgium
³ University of Ghent, Department of Movement and Sport Sciences, Watersportlaan 2, 9000 Ghent, Belgium

Aims
Male stag beetles fight aggressive battles over mating rights. In these fights, they grab their opponent between their elongated jaws and wrestle to detach it from the substrate. In the most spectacular battles, the loser is lifted into the air and thrown of the branch or tree log¹. Experimental measurements (see Figure 1) showed that, on average, male stag beetles bite 3 times as forcefully as females (after correction for the difference in body size)². It is impressive that males are able to produce such high bite force because they deliver this force with a very long output lever arm. Their high bite force could be facilitated by physiological improvements of the bite muscle fibers, by larger bite muscles or by an elongated input lever arm. Here we explore the last two options by microCT scanning.

Methods

Sample preparation
We obtained an adult male and female Cyclommatus metallifer individual from a commercial dealer (Kingdom of Beetle, Taiwan). After euthanasia, the anterior body parts (head + prothorax) were fixated in Bouin’s solution (Sigma-Aldrich, St. Louis, MO, USA) for 2 weeks. We dehydrated the samples in a graded ethanol series (in steps of 70, 80, 90, 96% and 100%). Finally, the samples were stained in a 1% iodine solution for 20 days (Sigma-Aldrich, St. Louis, MO, USA)².

Figure 1: Male stag beetle on the bite force measurement setup.
MicroCT scanning
The samples were scanned with a SkyScan 1172 high resolution microCT scanner (Bruker microCT, Kontich, Belgium) and with a custom built microCT scanner at the Centre for X-ray Tomography of Ghent University (UGCT) (see Table 1, Figure 2).

Table 1: Micro CT scan parameters

<table>
<thead>
<tr>
<th>SkyScan</th>
<th></th>
<th>UGCT</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Male</td>
<td>Male</td>
<td>Female</td>
<td></td>
</tr>
<tr>
<td>Voxel size</td>
<td>8.2 µm</td>
<td>38 µm</td>
<td>13 µm</td>
<td></td>
</tr>
<tr>
<td>Voltage</td>
<td>70 kV</td>
<td>130 kV</td>
<td>120 kV</td>
<td></td>
</tr>
<tr>
<td>Current</td>
<td>141 µA</td>
<td>108 µA</td>
<td>117 µA</td>
<td></td>
</tr>
<tr>
<td>Rotation angle</td>
<td>0.2°</td>
<td>0.3°</td>
<td>0.3°</td>
<td></td>
</tr>
</tbody>
</table>

Figure 2: MicroCT rendering of a male (a) and female (b) head. 1: jaw, 2: bite muscle, 3: hinge, 4: muscle attachment on jaw.

Segmentation
We selected the voxels belonging to a jaw and its bite muscles semi-automatically in Amira (3D image manipulation software; Amira 5.4.3, VSG systems, Mérignac, France). We separately segmented the attachment surface of the closer muscles to the head exoskeleton (see Figure 3). The muscle fibers converge from this convex head capsule to the muscle insertion on the jaw (see Figures 2,3). Hence, the area of the attachment surface on the head can be used as a proxy for the physiological cross-sectional area (PCSA) of the muscle and, therefore, of its force production. Further, we measured the length of the input lever arm (muscle attachment – hinge) and output lever arm (hinge – bite point) in Amira. For a certain muscle force, a longer input lever arm increases the produced bite force, while a longer output lever arm decreases the bite force.
Results

Adaptations for an enhanced male bite force

The male individual used in this study bit 6.5 N, while the female individual only produced a bite force of 1.6 N². After size normalization (by dividing by the posterior body mass), this corresponds to a bite force ratio of 2.06:

\[
\text{Measured bite force ratio} = \frac{\text{bite force}_{\text{male, normalized}}}{\text{bite force}_{\text{female, normalized}}} = \frac{7.9}{3.9} = 2.06.
\]

The male output lever arm (12.1 mm) is 3.6 times longer than the female output lever arm (3.4 mm, see Figure 3). Hence, males succeed in producing a twice as large bite force than females, despite their elongated output lever arm. This mechanical disadvantage is partially compensated by the 3.0 times longer male input lever arm (male: 3.6 mm; female: 1.2 mm, see Figure 3²).

Further, male stag beetles have enlarged bite muscles: their PSCA, which is directly correlated to their muscle force, is 2.4 times larger than in females.

With the anatomical data above, we can calculate the theoretical bite force ratio of males and females:

\[
\text{Theoretical bite force ratio} = \frac{\text{PSCA}_{\text{male}}}{\text{PSCA}_{\text{female}}} \cdot \frac{\text{input lever ratio}}{\text{output lever ratio}} = 2.4 \times \frac{3.0}{3.6} = 2.03.
\]

This theoretical bite force ratio (2.03), based on anatomical data only, is a very good prediction of the actual measured bite force ratio (2.06).

Figure 3: Segmentation of the microCT scans of the male and female heads. 1: jaw, 2: bite muscle, 3: hinge, 4: muscle attachment on jaw, 5: muscle attachment on head exoskeleton, 6: input lever arm, 7: output lever arm. The scale bars indicate 3 mm.
Muscle stress
Taking into account the male and female lever arm lengths, we find that their bite forces (6.5 and 1.6 N respectively) are generated by muscle forces of 9.9 and 2.4 N². By dividing these muscle forces by their PSCA, this shows that the male and female bite muscles have very similar muscles stresses of 18 and 17 N cm⁻², respectively. Muscle stress (or specific tension of a muscle) is the maximum force that a muscle can develop per unit of cross-sectional area.

Conclusion
Despite their ungainly long output lever arms, male stag beetles pinch on average three times as forcefully as females². Hence, they bite 526 times their own body weight, which is slightly more than specialized trap-jaw ants (which bite up to 504 times their own body weight)⁴,⁵. Our measurements, based on microCT scans, reveal that anatomical adaptations of the male head enable these extreme bite forces in stag beetles: males have elongated their input lever arm and enlarged their bite muscles for this purpose. As a result, no adaptation of their muscle physiology (i.e. muscle stress) was necessary². The enlarged male bite muscles come, however, at a significant cost to their owners: their weight increases both the energy costs of running and flying, and makes running stag beetles statically instable⁶,⁷.

References: