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Antler manipulation procedures for use in social and behavioral studies of deer

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Theories describing the role of antlers in reproductive biology have lacked substantive evaluation due to the inability to differentiate the effects of antler size from other allometrically related morphological traits. We developed an antler manipulation technique to facilitate comparisons of antler size while controlling for body size and age using white-tailed deer *Odocoileus virginianus*. We describe the process by which antlers can be manipulated to further investigate the functions of antlers. We believe this process is a viable option for manipulating antler size across Cervidae with appropriate modification to scale.

There has been much speculation as to the ecological justification for antler growth in Cervidae (Clutton-Brock 1982, Goss 1983). Antlers should enhance male fitness because they are costly to produce (Zahavi 1977, Ullrey 1983). Additional theoretical explanations of why males grow these structures range from predator defense to weapons for competition (Clutton-Brock 1982, Goss 1983), however few of these theories have been empirically evaluated. Supported theories of antler function include a display of dominance to other males, a weapon for intraspecific competition, and an ornament used by females to assess male quality (Demarais and Strickland 2011).

Although the dominance displaying function of antlers has some support in red deer (Bubenik 1983, Lincoln 1972, 1994), these studies were limited by the inability to manipulate antler size while controlling for allometrically related traits. Significant evidence suggests males use antlers as a weapon in intraspecific bouts against other males for access to females and to establish dominance (Bubenik 1983, Marchinton and Hirth 1984, Barrette and Vandal 1990, Miller et al. 2003). However, fighting ability and strength related to body size have also been related to success in malemale competition and reproductive success (Townsend and Bailey 1981, Jones et al. 2011). Thus, conflicting results have been reported when age and body size have been accounted for (Clutton-Brock 1982).

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To effectively assess the theorized functions of antlers, we must be able to manipulate antler size while controlling for the allometrically related trait of body size as well as age. Using an innovative antler-manipulation technique, we provided the first definitive support for female choice of larger antlered males (Morina et al. 2018). While a previous antler manipulation technique exists (Lincoln et al. 1970), the procedure was not described in enough detail to be reproducible. Here we describe our antler manipulation technique so that other researchers can use it for further investigations into the theories of antler functions.

Methods

Study area

We housed captive deer at the Mississippi State University Rusty Dawkins Memorial Deer Unit (MSU Deer Unit), Oktibbeha County, Mississippi, USA. The MSU Deer Unit is subdivided into five 0.4–0.8 ha housing pens and 6 0.05–0.07 ha holding pens. We provided water and two feeders with 20% crude protein deer pellets (Cargill Sportsman's Choice Record Rack) supplied ad libitum in each housing pen. Natural forages including white clover *Trifolium repens* and various grasses and forbs were also available within each pen.

Design and fabrication of antler coupling

A male–female sleeve and socket coupling device (Fig. 1) was designed to allow for natural positioning and rigid coupling of a foreign antler to a male after naturally grown

ITEM NO.	PART	DESCRIPTION	QTY.
1	Pedicle Base	Base Coupler	2/deer
2	Upper sleeve	Upper Antler Coupler	2/deer



Figure 1. Illustration of antler coupling assembly and parts.

antlers had been removed to a consistent height above the base of the pedicle. Preliminary measurements of representative antler base diameters to be coupled ranged from 19.1 to 44.5 mm. Therefore, the maximum inner diameter of the couplings was determined to be 44.5 mm to allow them to slip over any antler within the representative range. The couplings were machined from 50.8 mm outside diameter × 38.1 mm inside diameter 6061-T6 aluminum round tubing. The standard aluminum material is corrosion resistant and offers a strong and lightweight product that minimizes additional weight to the animal. Additionally, the dimensions of the standard tubing were consistent with the size needed for the couplings thus requiring minimal modification and machining. The couplings were designed and fabricated at the J. Charles Lee, Agricultural and Biological Engineering Machine Shop at Mississippi State Univ., Mississippi State, MS by means of standard precision machining procedures with a manual metal lathe to a dimensional tolerance of \pm 0.05 mm.

The base coupler serves as the male component of the coupling device (Fig. 2). It is designed to slip over and be affixed to the cut-off portion of the antler above the base of the pedicle and in line with the natural direction of the antler's main beam. The mass of the base coupler is 60.8 g. The upper antler coupling serves as the female sleeve component of the coupling device (Fig. 3). It is also designed to slip over and be affixed to the end of selected foreign antlers. Mounting holes drilled at strategic locations radially and axially through the sidewall of the upper coupling allow the couplings to be secured to the antler

with threaded mechanical fasteners. The mass of the upper antler coupling is 63.93 g. Both the upper and base couplers are designed to be installed axially in line with the natural direction of the antler main beam. The two-piece design allows the couplings to be preinstalled separately on selected antlers and deer. The radial design allows couplings to interlock concentrically upon installation and be rotated until a natural alignment and spread between antler beams is achieved. Combined mass of the coupling devices installed on both antlers of the animal is 124.75 g.

Mounting of antler in upper antler coupling

Prior to beginning the antler installation process, we prepared the upper antler couplings by preinstalling antlers with traits related to our trials (Morina et al. 2018). To evaluate female preference for antler size Morina et al (2018) selected smaller and larger antlers relative to the natural range of variation present in their study area. We placed each selected antler inside an upper coupling and positioned it to look as natural as possible when fully installed. We then held the antler in place with no. $10-16 \times 34$ hex drive washer head 410 stainless steel self-drilling screws at three evenly spaced locations around the coupler connection. We then filled the interstitial spaces between the antler base and the coupling using Technovit (Jorgensen Laboratories, Loveland, CO, USA), a strong acrylic binding adhesive material (Fig. 4).

Removal and preparation of antler

The antler installation procedure begins with the removal of the naturally grown antlers. For the easiest installation, we sedated the deer prior to installing the antlers and followed sedation procedures outlined in Morina et al. (2018). We used a reciprocating saw to remove antlers at a point 5 cm above the burr at the base of the pedicle. Some antlers are more oval than round while others have burrs projecting above the base burr. These conditions dictated that we use a rotary tool with a cutting blade to reduce the burrs or alter the shape to allow the base coupler to fit.

Installation of base coupling

We slid the base coupler down onto the antler base, leaving 2 cm between the bottom of the base coupler and the antler burr (Supplementary material Appendix 1, BaseCou-

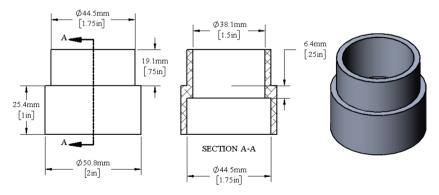


Figure 2. Illustration of base coupler and dimensions.

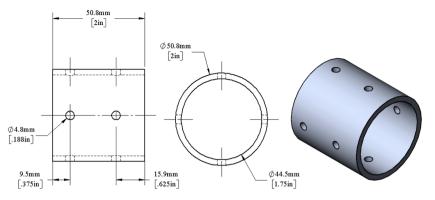


Figure 3. Illustration of upper coupler and dimensions.

pler.mp4). One must ensure the coupler is in line with the natural direction of the antler main beam. Then, we drilled a pilot hole 10 mm from the bottom of the base coupler through both walls of the coupler and the antler base using a 4.76 mm (3/16" imperial) drill bit of sufficient length (≥50 mm). We inserted a no. 10–24 × 3" 6-lobe drive truss head grade 18–8 stainless steel machine screw (Fastenal Company, Winona, MN, USA) into the pilot hole and secured it using a no. 10–24 grade 18–8 stainless steel nylon insert lock nut (Fastenal Company, Winona, MN, USA). We removed the excess length of the machine screw that extended past the lock nut and filed down the new end to prevent any possible skin irritation. We then drilled a second pilot hole perpendicular to and 10 mm above the first fastener location and repeated the same fastening procedure.

After the fasteners were in place, we filled the interstitial spaces between the antler base and the coupling using anchoring adhesive (AnchorFix-1, Sika Corporation, Lyndhurst, NJ, USA). We applied tape at the bottom of the base coupler to prevent leakage of the anchoring adhesive onto the animal tissue at the base of the antler during the drying period. We then repeated the entire procedure for the second antler base and coupler (Fig. 5).

Installation of upper antler coupling

After we secured the base couplers, we slid each of the upper antler couplings onto their respective base coupling

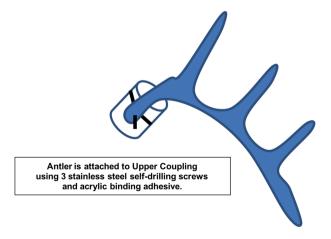


Figure 4. Schematic representing antler attachment to upper coupling.

(Supplementary material Appendix 1, Upper.Coupling. mp4). We rotated the antlers to achieve a natural alignment and spread between antler. We then secured the antlers using no. $10-16 \times \frac{1}{2}$ ° hex drive washer head 410 stainless steel self-drilling screws (Fastenal Company, Winona, MN, USA) at three evenly spaced locations around the coupler connection (Fig. 5). To provide a more natural look to the antlers when presented to females in trials, we wrapped the antler couplings with four layers of beige Color Duck Tape Brand duct tape (ShurTech Brands, Avon, OH, USA) (Supplementary material Appendix 1, NaturalLook.mp4).

Results

During the breeding seasons of 2015-2016 and 2016-2017, we installed antlers on six male white-tailed deer. Antlers remained installed for 10-52 days. The group of manipulated male deer was comprised of two six-yearolds, two three-year-olds and two yearlings. The time needed to adapt to the new antler size differed between age classes. The six-year-old males behaved normally after antler installation and did not require any acclimation period. The three-year-old males carried their heads slightly lower than normal and required a 2-24-h acclimation period, after which they could carry larger antlers with a natural, consistent appearance of the head and neck. The yearling males initially hung their heads significantly lower than normal and had trouble stabilizing movement of the large size antlers. They required a 24–48-h acclimation period. Qualitatively, there was no detectable differences in the behavior of manipulated and non-manipulated males after the acclimation period.

The six-year-old males had manipulated antlers installed for 52 days and the three-year-old males had manipulated antlers installed for 34 days. The yearling males had manipulated antlers installed for 10 days, at which point the yearling male with the large antlers installed broke them off at the fastening location, where the antler integrity was weakened. Upper antler couplings were removed from each male at the end of a set trial period conducted for a separate study (Morina et al. 2018). Base couplings remained on the animals until they fell off during the natural antler shedding period.

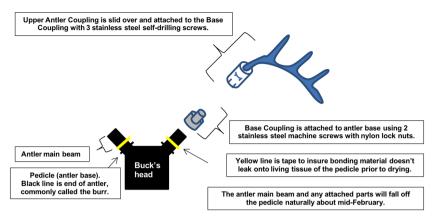


Figure 5. Schematic representing base coupling attachment to main beam and upper antler coupling to base coupling.

Discussion

Using our innovative process for size manipulation of deer antlers, we have provided a technique that can be used to disentangle the influence of antlers from other allometrically related traits in social dominance, intrasexual competition and mate choice. For instance, we used this method to test the ornament function of antlers by manipulating antler size while controlling allometrically related traits, like body size, to isolate the influence of antler size on female mate choice (Morina et al. 2018). Thus, the use of our antler manipulation process can be used in future social and behavioral studies to test current theories for the functions of antlers.

Our method provides more control of antler size manipulation than some previous methods attempted since it allows for increasing antler size in addition to simply size reduction (McComb and Clutton-Brock 1994). Lincoln et al. (1970) artificially increased antler size in red deer *Cervus elaphus*, but lack of a thorough description of the manipulation process warranted publication of a full description of a similar technique that others could repeat. We believe our technique will be a viable option across all species of Cervidae, although slight modifications may be required for some species due to antler diameter. Both the size of the couplings and the attachment hardware may need to be increased for species with larger and heavier antlers.

In manipulating the antlers of deer, the diameter of the antler bases need to be considered. The yearling male we attached a large set of antlers to had them break off with a forceful collision into a fence. His antler base diameter was approximately 2 cm and was not strong enough to withstand the force exerted on the attachment point of the base antler coupler. Though we did not determine the threshold of antler size to pedicle diameter, we would recommend limiting installation of large antlers to males with appropriate pedicle diameters.

Manipulated antlers worn by older males withstood abuse related to breeding behavior of male deer. Males rubbed their antlers on many surfaces within our research facility including fences, feeders and trees. No antlers manipulated using this technique came loose or broke off. In fact, one of the males in our study broke an antler tine off of his installed antlers with no damage to the antler coupling device. However, we did not allow males to directly interact with manipulated antlers installed, so it is possible

there may be some limitation for their use in competition events. Based on their durability during our trials, we believe they would hold up in male—male combat but we cannot say what the failure threshold would be. Since the devices are relatively small, it would be possible to increase the length or thickness of the coupler devices to increase strength as needed.

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