



Compressive mechanical properties of dry antler cortical bone cylinders from different cervidae species

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ABSTRACT

Antlers are bony structures composed predominantly of primary osteons with unique mechanical properties due to their specific use by deer as weapon and shield. Antler bone fracture resistance has attracted prior scrutiny through experimental tests and theoretical models. To characterize antler mechanical properties, compression of cubes, or bending or tensioning of rectangular bars have been performed in the literature with variations in the protocols precluding comparisons of the data. Compression testing is a widely used experimental technique for determining the mechanical properties of specimens excised from cortical or cancellous regions of bone. However, the recommended geometry for compression tests is the cylinder, being more representative of the real performances of the material. The purpose of research was to report data for compressive strength and stiffness of antler cortical bone following current guidelines. Cylinders ($n = 296$) of dry antler cortical bone from either the main beam or the tines of *Cervus elaphus*, *Rangifer tarandus*, *Cervus nippon* and *Dama dama* were tested. This study highlights the fact that compression of antler cortical bone cylinders following current guidelines is feasible but not applicable in all species. Standardization of the testing protocols could help to compare data from the literature. This study also confirms that sample localization has no effect on the mechanical properties, that sample density has a significant impact and allows enriching the knowledge of the mechanical properties of dry antler cortical bone.

1. Introduction

In the realm of bone mechanics, an understanding of how bones respond to various stresses at different levels is paramount. Bones, at the macroscopic level, undergo compression, bending, and torsion forces. Zooming in to the tissue level, cortical bone experiences a complex interplay of loading stress, including tension, compression, and shear. This cortical layer, akin to an external shell, exerts a significant influence on the overall mechanical behavior of the entire bone structure (Attia and Willett, 2017; Zhao et al., 2018). Bone has two main mechanical properties: stiffness and strength. Measures of stiffness and strength need to be recorded and calculated during a mechanical test (An and Draughn, 1999). While stringent standards govern the experimental assessment of engineering materials, such norms are not uniformly established for bone compression testing. The application of standardized material testing protocols often encounters constraints due to the use of biological tissue. These challenges emanate from the intrinsic heterogeneity and finite size of bone specimens, complications in

gripping bone surfaces, and limitations on the magnitude of applicable loads. Consequently, variations in testing methodologies and specimen preparations proliferate across the literature, impeding direct study comparisons (Keaveny et al., 1993).

Compression testing is a widely used experimental technique for determining the mechanical properties of bulk tissue specimens excised from cortical or cancellous regions of bone (El Masri et al., 2012; Linde et al., 1988; Bayraktar et al., 2004; An and Draughn, 1999; Røhl et al., 1991; Zhao et al., 2018). However, there is wide variation in the literature about how compression testing of bone is performed, with no benchmark protocol (Zhao et al., 2018). Cortical bone can also be tested using configurations suited to measure behavior under pure shear, three-point bending, and four-point bending (Pal, 2017; Ma et al., 2017). Because bending combines tension and compression in a more complex stress distribution, material properties measured in bending differ from those measured in pure tension or pure compression. Moreover, cortical bone exhibits distinct mechanical characteristics under tension versus compression. The utility of compression testing lies

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in its straightforward execution, delivering measurements of apparent elastic modulus and other properties, such as ultimate strength (Donnelly, 2011). In this context, adhering to a cylindrical sample geometry is advocated (Attia and Willett, 2017; Zhao et al., 2018).

Antlers are organs of variable size and are present by pairs on the skull of males of most cervid species. Remarkably, these structures, composed predominantly of primary osteons, can weigh several kilograms per individual (Chen et al., 2009; Picavet and Balligand, 2016). Their specific use by deer (weapon and shield) is associated with particular mechanical properties namely a lesser stiffness but a higher fatigue resistance compared to skeletal bone (Clutton-Brock, 1982; Geist, 1966; Leslie Jr and Jenkins, 1985; Lincoln, 1992). Antlers can undergo high impact loading and large bending moments without fracture (Blob and Snelgrove, 2006). Antler bone fracture resistance has attracted prior scrutiny through experimental tests and theoretical models (Currey, 1999; Landete-Castillejos et al., 2007; Vashishth et al., 1997; Kitchener, 1991, 2000; Chen et al., 2009). Notably, extensive investigations by Blob and LaBarbera (2001) and Blob and Snelgrove (2006) into antler bones from white-tailed deer and moose highlighted that elastic moduli remain independent of longitudinal orientation, implying localization where the specimens were taken from does not significantly influence mechanical properties such as bending strength, ultimate tensile strength, and work of fracture. Currey (1989) and Zioupos et al. (1994), Zioupos and Currey, 1998 reported that the mineral content of the antler bone presents a strong relationship between the work of fracture and the elastic modulus of the bone. Namely, as the mineral content increases, the work of fracture decreases while the elastic modulus increases. Numerous other tests have been carried out by different investigators using various models and techniques. To characterize these mechanical properties, compression of cubes or bending or tensioning of rectangular bars (Shah et al., 2008; Rajaram and Ramanathan, 1982; Currey et al., 2009a; Launey et al., 2010; Chen et al., 2009; Blob and Snelgrove, 2006; Cappelli et al., 2017) have been performed. These differences between the tests render comparisons hazardous. Even though the recommended geometry for compression tests is the cylinder, being more representative of the real performances of the material, the authors of this study were unable to find documentation on the biomechanical properties of cortical antler bone cylinders tested in compression.

The objectives of this study were (1) to characterize mechanical properties of dry antler cortical bone cylindrical samples from different Cervidae species tested in compression; (2) to compare the data evaluating the effect of the species; (3) to confirm that sample localization (beam versus tines) has no effect on the mechanical properties of antler cortical bone.

2. Material and methods

2.1. Specimens

Antlers from 7 species of Cervidae and at least 3 individuals per species were harvested. The specimens originated from fallow deer (*Dama dama*), red deer (*Cervus elaphus*), reindeer (*Rangifer tarandus*), Sika deer (*Cervus nippon*), white-tailed deer (*Odocoileus virginianus*), muntjac (*Muntiacus muntjak*) and roe deer (*Capreolus capreolus*). All antlers were harvested after cast-off and belonged to adult animals. The antlers collected showed no sign of diagenetic alteration, no weathering (grey color) and no cracks. All antlers were stored at room temperature for at least 6 months prior to extraction and experimentation on bone specimens.

2.2. Extraction and preparation of specimens

The extraction of the samples was carried out at the Laboratory of Mechanics of Materials and Structures at the Faculty of Applied Sciences, University of Liège (Belgium). Cortical bone segments were

extracted either from the proximal portions of the antler (beam) or at the distal portions of the tines. Antler palms were avoided for specimen harvesting. Extraction was done using a Startrite Band Saw (Model 30RWH) equipped with a 12 teeth/inch blade, rotating at 80 m/min. The localization of the samples was determined according to the amount of cortical bone as it was necessary to obtain specimens allowing the production of perfect cylinders. Each specimen was machined at approximately 40.0 mm in length and 15.0 mm in width. Cancellous bone and imperfections that could compromise the mechanical tests were removed. The longitudinal axis of each specimen was parallel to the longitudinal axis of the original antler. The samples were slit lengthwise in areas of cortical thickness allowing extraction of cylinder blanks of ± 6 mm in diameter. Each specimen was then machined in a Ernault Batignolles lathe (model AC 280) equipped for the occasion with a Schaublin bar clamp (type F28) and a HSS tool (High Speed Steel) with positive cut Ifanger to reach a test tube of ± 5.5 mm in diameter. A cylinder of 5 mm in diameter was then rotated by raising the reference face for setting to length. The test specimens were polished using a 1000-grit sandpaper. A 5-mm diameter cylinder was then rotated by setting the face that will serve as a reference for the lengthening. Specimens with a final length of 10 mm were polished with 1000-grit sandpaper (Fig. 1). Radiographs of each sample lot were taken (Digivex Medex *, 43 kVp, 12.5 mAs) to check for the presence of cancellous bone. The length and diameter of the samples were measured using a digital calliper. Samples were then weighed using a balance (Mettler PM200) capable of an accuracy of 0.001 g for a measurement of 200 g. The volume (V) was calculated from the formula $\pi \cdot r^2 \cdot h$ where r is the radius of the cylinder and h is the height. The density ($\rho = m/V$) was eventually calculated.

*: Digivex Medex, Belgium (120kV/300 mA, flat panel)

2.3. Mechanical tests

The cylindrical samples were tested in compression in a longitudinal direction. The machine used was an Instron 3366 press with a traverse speed set at 0.5 mm/min. Machine compliance was found to be negligible. All trials were conducted until fracture. Force-displacement curves were obtained (Bluehill® 2) indicating the values of maximum compressive strength (MCS) (N), maximum compressive stress (σ_c) (MPa), maximum energy absorbed (MEA) (J) and modulus of elasticity (EM) (GPa).

2.4. Statistical analysis

Descriptive statistics were carried out on all available variables. A linear mixed model was used to model MCS as a function of sample localization (beam or tines), sample species (red deer, fallow deer, reindeer or Sika deer) and the interaction between localization and species. To control for possible differences in sample density during sampling, sample density was used as an adjustment factor in the linear mixed model. The normality and homoscedasticity of the residuals were checked graphically. If necessary, a logarithmic transformation was applied to respect the assumptions of normality and homoscedasticity of the residuals. The same methodology was used to study MEA and EM. Comparison of sample density between the different species was carried out using a Kruskal-Wallis test and multiple comparisons. The same methodology was applied to compare density between the two localizations. For all comparisons, $p \leq 0.05$ was considered the significance level. All the statistical analyzes were carried out with the SAS (9.4) and R (4.2) software.

3. Results

3.1. Samples

A total of 296 samples were tested, harvested from the antlers of 15

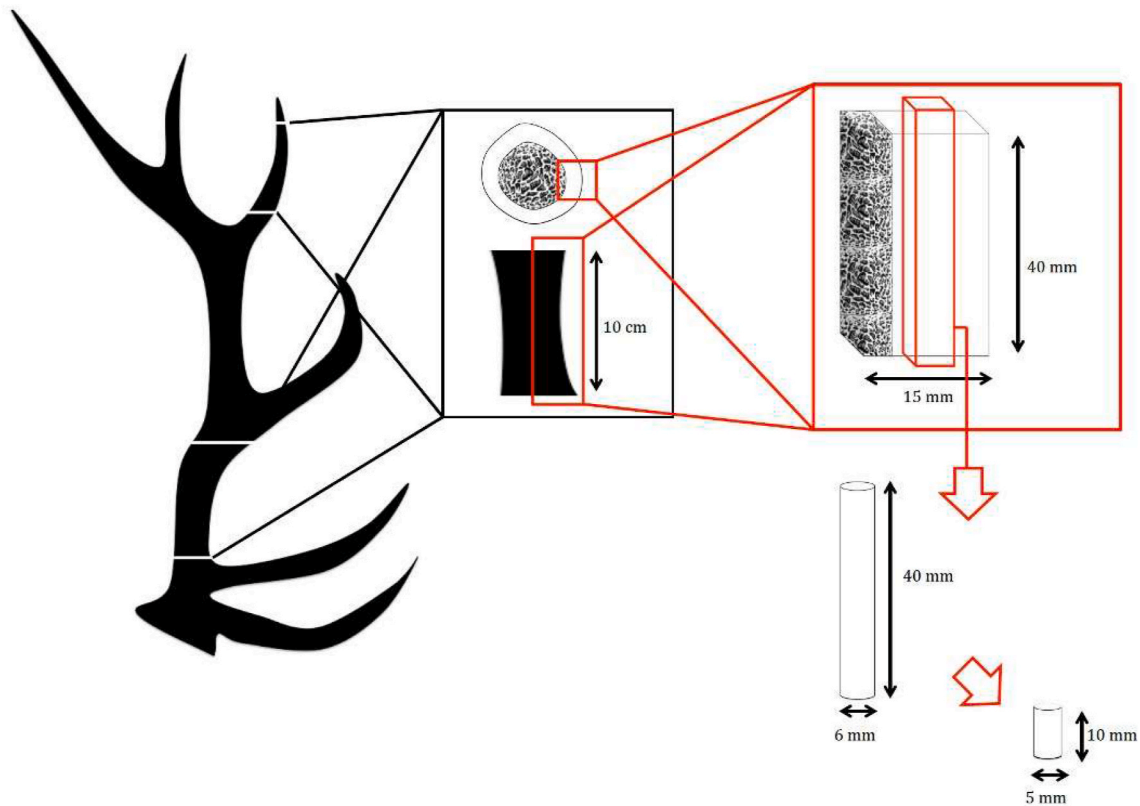


Fig. 1. Schematic illustration of the protocol for extracting the antler cortical bone cylinders.

different cervids. A limiting factor was observed for white-tailed deer, roe deer and muntjac for which the thin cortical layer made sample preparation impossible. These species were therefore excluded from the study. The median (Q1-Q3) number of samples per antler was 20.0 (15.0–27.0). The 15 cervids came from 4 different species: 3 red deer, 6 fallow deer, 3 reindeer and 3 Sika deer. Table 1 shows the distribution of the species and localization from which the samples were taken. Machining of 5 mm diameter antler cortical bone cylinders was not possible for reindeer 1 in the main beam, and for reindeer 3 in the tines.

The mean cylinder height and diameter ± standard deviations (SD) were 10.06 ± 0.07 mm and 5.01 ± 0.06 mm respectively. The mean calculated volume and weight ±SD were 198.30 ± 5.462 mm³ and 0.33 ± 0.03 g respectively. The mean calculated density ±SD was 1.7 ± 0.155 g cm⁻³. All the values related to the dimensional data of the cylinders are presented in Table 2.

Species had a significant impact on sample density ($p < 0.0001$). Specifically, density was significantly higher for red deer than for reindeer (0.0017 (0.0017–0.0018) vs. 0.0016 (0.0015–0.0017), $p = 0.0013$). Conversely, the density of samples taken from red deer was significantly lower than the density of samples taken from Sika deer (0.0017 (0.0017–0.0018) vs. 0.0018 (0.0017–0.0018), $p < 0.0001$). The density of samples taken from Sika deer was also significantly higher

Table 1
Distribution of species and sample localization - N = 296.

Variable	Categories	N	Number (%)
Species	Red deer	296	87 (29.4)
	Fallow deer		117 (39.5)
	Reindeer		29 (9.8)
	Sika deer		63 (21.3)
Localization	Beam	296	172 (58.1)
	Tines		124 (41.9)

than the density of samples taken from fallow deer (0.0018 (0.0017–0.0018) vs. 0.0017 (0.0016–0.0018), $p = 0.009$) or reindeer (0.0018 (0.0017–0.0018) vs. 0.0016 (0.0015–0.0017), $p < 0.0001$).

Sample localization had also a significant impact on sample density ($p < 0.0001$). The density of samples taken from beam was significantly higher than the density of samples taken from tines (0.0018 (0.0017–0.0018) vs 0.0017 (0.0015–0.0018), $p < 0.0001$).

3.2. X-rays

Presence of cancellous bone or imperfections was monitored on radiographs. Only cortical bone remained after preparation and no imperfections were present (Fig. 2).

3.3. Compression tests

Table 3 summarizes the descriptive statistics of the mechanical properties of the samples studied, by species and localization.

3.3.1. MCS

In order to model the impact of the species from which the samples were taken, as well as the impact of the sampling localization on the maximum compression strength, a linear mixed model adjusted by sample density was carried out. Based on this model, we observed that MCS differs between species ($p = 0.038$) and that this difference is similar regardless the localization ($p = 0.41$). Sample localization had no significant impact on MCS ($p = 0.054$). The detailed results of this model are presented in Table 4. More specifically, whatever the localization, the mean MCS was significantly smaller for samples taken from fallow deer (-1072.25 ± 342.12 , $p = 0.0095$) or reindeer (-1075.45 ± 409.75 , $p = 0.024$) than for samples taken from red deer. There was no difference between Sika deer and red deer ($p = 0.43$). Density has a highly significant impact on MCS: it increased significantly with increasing density ($p < 0.0001$). Fig. 3 shows the estimated means of

Table 2
Descriptive statistics for sample sizes, globally, by species and localization - N = 296.

Variable	N	Mean	SD	Min	Q1	Median	Q3	Max
Height (mm) - All	296	10.06	0.07	9.85	10.01	10.05	10.10	10.36
Red deer	87	10.09	0.06	9.85	10.06	10.09	10.13	10.36
Fallow deer	117	10.03	0.07	9.90	9.99	10.02	10.07	10.19
Reindeer	29	10.05	0.07	9.92	9.99	10.03	10.12	10.17
Sika deer	63	10.05	0.05	9.94	10.01	10.05	10.08	10.21
Beam	172	10.06	0.07	9.90	10.01	10.05	10.10	10.36
Tines	124	10.06	0.07	9.85	10.01	10.05	10.11	10.26
Diameter (mm) - All	296	5.01	0.06	4.85	4.98	5.02	5.05	5.20
Red deer	87	5.01	0.06	4.85	4.98	5.02	5.05	5.18
Fallow deer	117	5.00	0.05	4.87	4.97	5.00	5.02	5.20
Reindeer	29	5.04	0.03	5.00	5.01	5.03	5.04	5.13
Sika deer	63	5.02	0.08	4.87	4.96	5.03	5.07	5.19
Beam	172	5.01	0.06	4.87	4.98	5.01	5.05	5.20
Tines	124	5.01	0.06	4.85	4.97	5.02	5.05	5.19
Volume (mm³) - All	296	198.30	5.01	185.02	195.27	198.42	201.39	212.26
Red deer	87	199.23	5.00	185.02	196.23	199.41	202.20	211.06
Fallow deer	117	196.89	4.49	186.94	194.10	196.64	199.80	212.26
Reindeer	29	200.05	3.41	195.66	197.63	199.40	201.00	209.27
Sika deer	63	198.82	5.92	188.04	194.09	199.80	203.20	211.45
Beam	172	198.31	4.77	186.94	195.27	198.42	200.99	212.26
Tines	124	198.29	5.34	185.02	195.14	198.50	202.19	211.45
Weight (g) - All	296	0.33	0.03	0.19	0.33	0.34	0.35	0.38
Red deer	87	0.34	0.01	0.30	0.34	0.34	0.35	0.37
Fallow deer	117	0.32	0.04	0.19	0.31	0.34	0.35	0.37
Reindeer	29	0.32	0.02	0.26	0.31	0.33	0.34	0.35
Sika deer	63	0.35	0.02	0.29	0.34	0.35	0.36	0.38
Beam	172	0.34	0.02	0.20	0.34	0.35	0.35	0.37
Tines	124	0.32	0.04	0.19	0.31	0.34	0.35	0.38
Density (g/mm³) - All	296	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Red deer	87	0.0017	0.0001	0.0015	0.0017	0.0017	0.0018	0.0019
Fallow deer	117	0.0016	0.0002	0.0010	0.0016	0.0017	0.0018	0.0019
Reindeer	29	0.0016	0.0001	0.0013	0.0015	0.0016	0.0017	0.0018
Sika deer	63	0.0018	0.0001	0.0015	0.0017	0.0018	0.0018	0.0018
Beam	172	0.0017	0.0001	0.0010	0.0017	0.0018	0.0018	0.0019
Tines	124	0.0016	0.0002	0.0010	0.0015	0.0017	0.0018	0.0019

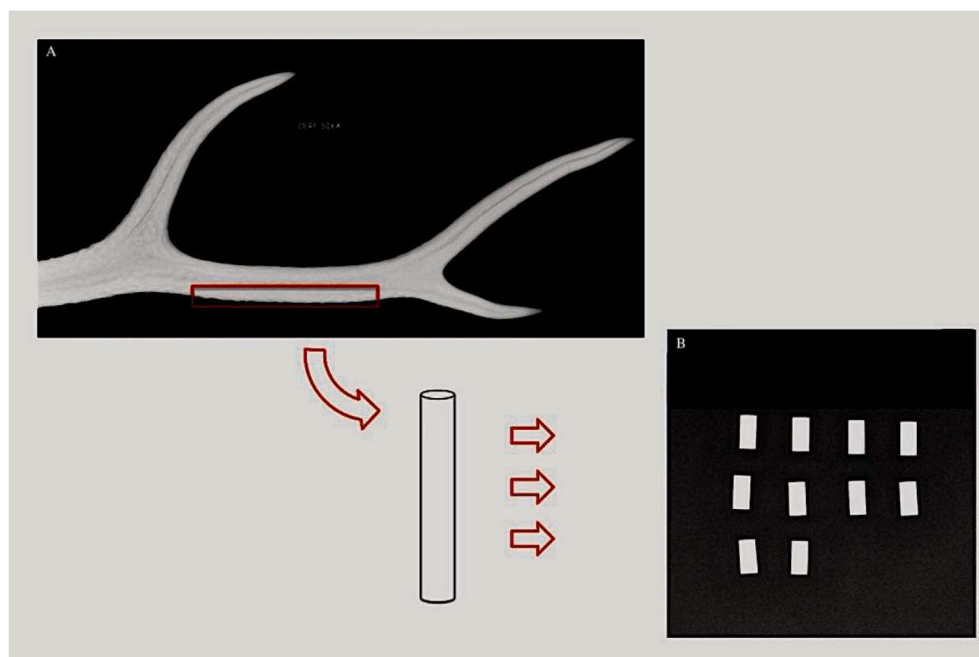


Fig. 2. Radiographic evaluation of the antler (A) of a Sika deer and the cylinders harvested from the main beam (B).

MCS by species and localization, based on the linear mixed model.

3.3.2. σ_c

A linear mixed model adjusted by sample density was performed to analyze the maximum compressive stress of samples as a function of

species and sampling localization. This linear mixed model showed a statistically significant impact of species ($p = 0.047$) and localization ($p = 0.047$). However, the impact of these two factors were independent of each other ($p = 0.39$). **Table 5** presents the detailed results of this linear mixed model. Despite a significant effect of localization, the regression

Table 3

Descriptive statistics of mechanical properties of samples, globally, by species and by localization - N = 296.

Variable	N	Mean	SD	Min	Q1	Median	Q3	Max
MCS (N) - All	296	3876.4	1016.7	508.50	3402.8	4158.9	4569.9	5777.7
Red deer	87	4484.3	329.86	2954.4	4310.9	4508.5	4656.1	5132.2
Fallow deer	117	3292.4	1133.4	508.50	2482.0	3680.5	4156.8	5012.2
Renne	29	3330.4	649.48	1937.3	2792.9	3479.5	3718.5	4570.1
Sika deer	63	4372.7	788.25	2563.3	3844.5	4452.1	5066.9	5777.7
Beam	172	3944.3	915.95	728.50	3489.1	4217.3	4553.3	5481.2
Tines	124	3782.1	1139.0	508.50	3070.0	4080.9	4616.6	5777.7
σ_c (MPa) - All	296	196.77	51.71	26.40	171.15	211.42	232.50	294.26
Red deer	87	228.38	16.80	150.47	219.55	229.62	237.13	261.38
Fallow deer	117	167.90	57.58	26.40	128.20	186.50	211.91	255.27
Reindeer	29	168.58	33.18	98.30	141.70	175.06	189.38	229.10
Sika deer	63	219.73	41.81	127.50	192.80	220.90	258.06	294.26
Beam	172	200.12	46.70	37.50	175.62	213.80	231.38	279.16
Tines	124	192.13	57.82	26.40	159.82	207.66	234.18	294.26
MEA (J) - All	296	1.29	0.86	0.10	0.90	1.26	1.52	7.99
Red deer	87	1.43	0.17	0.88	1.34	1.44	1.53	1.82
Fallow deer	117	1.33	1.28	0.10	0.80	1.10	1.43	7.99
Reindeer	29	0.86	0.28	0.30	0.70	0.90	1.10	1.27
Sika deer	63	1.25	0.46	0.50	0.90	1.20	1.60	2.80
Beam	172	1.27	0.83	0.20	0.90	1.30	1.52	7.99
Tines	124	1.33	0.90	0.10	0.98	1.25	1.55	7.43
EM (GPa) - All	296	6.80	0.87	2.49	6.84	7.00	7.08	8.16
Red deer	87	6.95	0.10	6.84	6.84	7.00	7.08	7.08
Fallow deer	117	6.45	1.24	2.49	6.26	6.87	7.08	8.16
Reindeer	29	6.99	0.60	5.43	6.84	7.04	7.38	8.06
Sika deer	63	7.16	0.42	5.72	6.85	7.08	7.58	8.08
Beam	172	6.97	0.71	2.73	6.84	7.00	7.34	8.16
Tines	124	6.57	1.02	2.49	6.84	6.87	7.08	7.71

Table 4

Results of the linear mixed model of maximum compressive strength as a function of species and sample localization - N = 296.

Effect	Localization	Species	Coefficient \pm SE ^a	P-value
Localization (ref = beam)	Tines		33.01 \pm 82.55	0.70
Species (ref = red deer)		Fallow deer	-1072.25 \pm 342.12	0.0095
Species (ref = red deer)		Reindeer	-1075.45 \pm 409.75	0.024
Species (ref = red deer)		Sika deer	-326.30 \pm 394.86	0.43
Localization*Species	Tines	Fallow deer	24.09 \pm 116.55	0.84
Localization*Species	Tines	Reindeer	442.24 \pm 256.44	0.12
Localization*Species	Tines	Sika deer	70.77 \pm 129.16	0.60
Density			3,250,509 \pm 218,006	<.0001

^a Coefficient \pm SE: Regression coefficient and standard error.

coefficient showed that localization did not significantly impact σ_c (1.71 \pm 4.17, $p = 0.70$). In terms of species, samples from fallow deer antlers (-54.44 \pm 17.68, $p = 0.011$) or reindeer antlers (-56.23 \pm 21.14, $p = 0.022$) had a significantly lower σ_c than samples from red deer antlers. The σ_c of Sika deer antler samples was similar to that of red deer antler samples (-19.74 \pm 20.40, $p = 0.35$). Maximum compressive stress increased significantly as sample density increased ($p < 0.0001$). The average σ_c estimated by the linear mixed model are shown by species and location in Fig. 4. In order to explain the discrepancy between the overall p-value showing a significant effect of location ($p = 0.047$) and the non-significant effect of location via the regression coefficients, contrasts were made to compare the maximum compressive stress between the two sample locations within each species, despite the non-significant effect of the interaction between species and localization ($p = 0.39$). It should be noted that the overall p-value ($p = 0.047$) compares the localization to an average level of the species. When the

contrasts are made, a similar maximum compressive stress is observed between the two localizations for each species (red deer: $p = 0.69$, fallow deer: $p = 0.44$, reindeer: $p = 0.071$, Sika deer: $p = 0.38$).

3.3.3. MEA

The impact of species and sample localization on MEA was modelled using a linear mixed model adjusted by sample density. The interaction between localization and species was not significant ($p = 0.41$). Similarly, neither localization ($p = 0.16$) nor species ($p = 0.40$) had a significant impact on MEA. The results of this model are presented in Table 6. The impact of density on maximum energy absorbed was statistically significant; maximum energy absorbed increased significantly with increasing density ($p < 0.0001$). Fig. 5 shows the average logarithmic transformation of the maximum absorbed energy estimated by the linear mixed model, by species and localization.

3.3.4. EM

A density-adjusted linear mixed model modeling the modulus of elasticity of the samples as a function of the species from which the samples were taken, as well as the localization where the samples were taken, was carried out. The detailed results of this linear mixed model are presented in Table 7. Based on this model, the interaction between species and localization was not statistically significant ($p = 0.17$). Also, the modulus of elasticity was not impacted by sample localization ($p = 0.063$) or species ($p = 0.087$). The modulus of elasticity increased significantly with increasing density ($p < 0.0001$).

The means estimated by the linear mixed model of the modulus of elasticity are shown in Fig. 6 by species and localization.

4. Discussion

The research described here was performed with the goal of characterizing the compressive mechanical properties of dry antler cortical bone from different Cervidae species using cylindrical samples. We obtained data for red deer, fallow deer, reindeer and Sika deer. To our knowledge, no data has ever been published to date concerning the mechanical properties of fallow deer antlers.

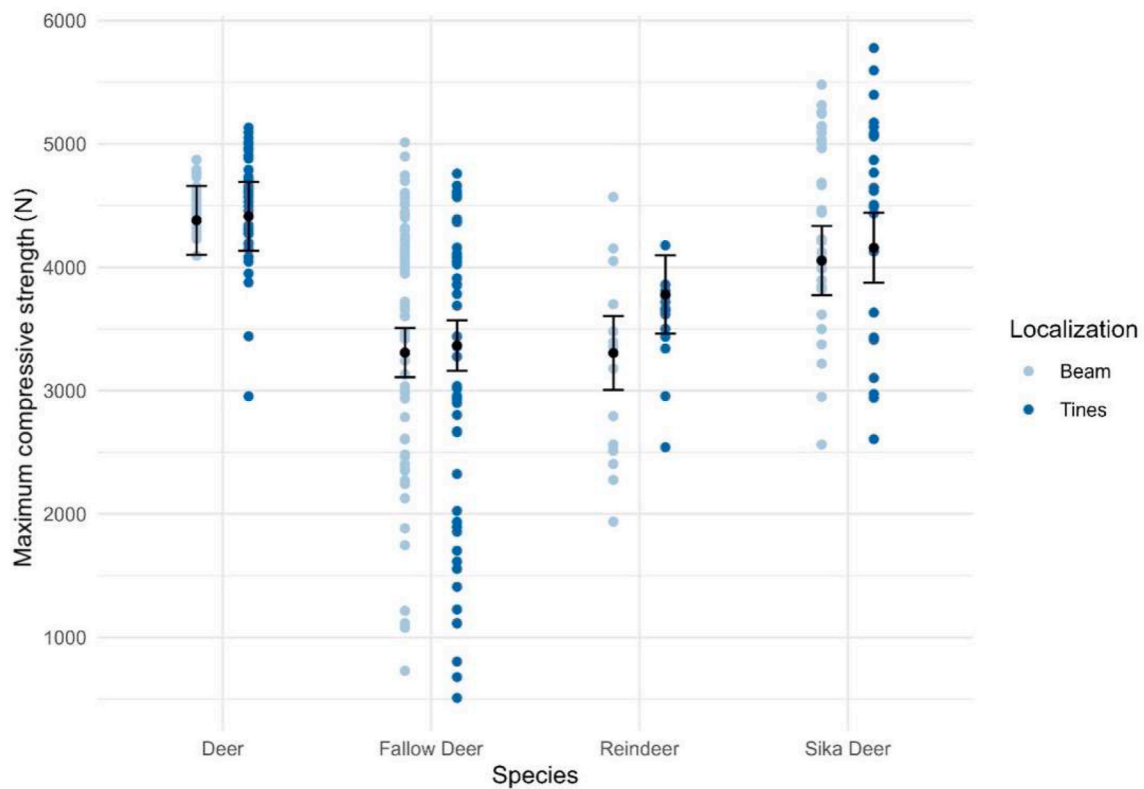


Fig. 3. Estimated mean (mean and standard error) MCS of samples by species and localization - N = 296. Points represent individual sample values.

Table 5

Results of the linear mixed model of maximum compressive stress as a function of species and sample localization - N = 296.

Effect	Localization	Species	Coefficient ±SE ^a	P-value
Localization (ref = beam)	Tines		1.71 ± 4.17	0.69
Species (ref = red deer)		Fallow deer	-54.44 ± 17.68	0.011
Species (ref = red deer)		Reindeer	-56.23 ± 21.14	0.022
Species (ref = red deer)		Sika deer	-19.74 ± 20.40	0.35
Localization*Species	Tines	Fallow deer	1.79 ± 5.89	0.77
Localization*Species	Tines	Reindeer	23.55 ± 12.99	0.10
Localization*Species	Tines	Sika deer	2.91 ± 6.52	0.67
Density			166,773 ± 11,021	<.0001

^a Coefficient±SE: Regression coefficient and standard error.

The protocol used in this study is innovative in the field of biomechanical tests in antler bone as compression of cylinders of cortical bone have never been performed so far, to the author’s knowledge.

Compression testing is a widely used technique for measuring the stiffness of bone. Theoretically, a compression test is not as solid as a tensile test due to the edge effect and the axial inaccuracies (Reilly et al., 1974; Wall et al., 1970) and a tensile test is the standard engineering test for determining material properties (An and Draughn, 1999). However, because of the difficulties of specimen fabrication or testing, this test has not been utilized as commonly as compression testing for mechanical properties of bone (An and Draughn, 1999). Also, it is now well accepted that various types of mechanical tests are required to fully assess the antler mechanical properties (Launey et al., 2010; Skedros et al., 2014).

For compression tests, the specimens are typically cylindrical or

parallelepiped (Zhao et al., 2018). Opting for cylinders was driven by their suitability for compression analysis (Attia and Willett, 2017). The cylindrical shape facilitates uniform force distribution at all points, a distinctive advantage over other geometric shapes that might induce a restraining effect due to stress distribution modification at edges (Linde et al., 1992). Zhao et al. (2018) mentioned that the following aspects should be considered when selecting specimen geometry: (1) the aspect ratio (i.e. the height to width ratio of the specimen) should be between 1 and 2 to avoid buckling of the specimen during the compression test and thus maintain axial load application (Wang et al., 2010); (2) cylindrical cross-section specimens should be used where possible, with 2:1 cylinder of minimum 5 mm diameter as suggested by Keaveny et al. (1993) would be recommended; (3) the method of strain measurement. In general, larger specimens should be used if using machine crosshead motion for strain measurement to minimize the effect of the structurally compromised free surface layers. Zhu et al. (1994) recommended a minimum height of 10 mm and cross-sectional area of 100 mm². We followed these recommendations, machining cortical bone cylinders of 10 mm height and 5 mm diameter. The preparation of a cylinder remains also easier than that of a cube, which requires erecting strictly parallel faces. However, the preparation process was not without its challenges. Indeed, few regions in the antlers had a regular thickness of at least 5 mm of only cortical bone. This necessitated the multiplication of sawing localizations to find suitable areas. This also involved taking samples from adult individuals in good condition to maximize the chances of having a significant cortical layer. The search for a sufficiently thick cortical layer is also a constraining factor when switching to other cervid species. Indeed, other species were envisaged in this study (white-tailed deer, roe deer, muntjac) but, at no point throughout the antlers it was possible for us to harvest 5 mm of cortex for the specimens in our possession. These species were thus excluded from the study. Also, machining of the cylinders was only possible in one of the two localizations (beam or tine) for two reindeer (1 and 3). Importantly, the radiographs attested to the quality of machining, revealing the absence of

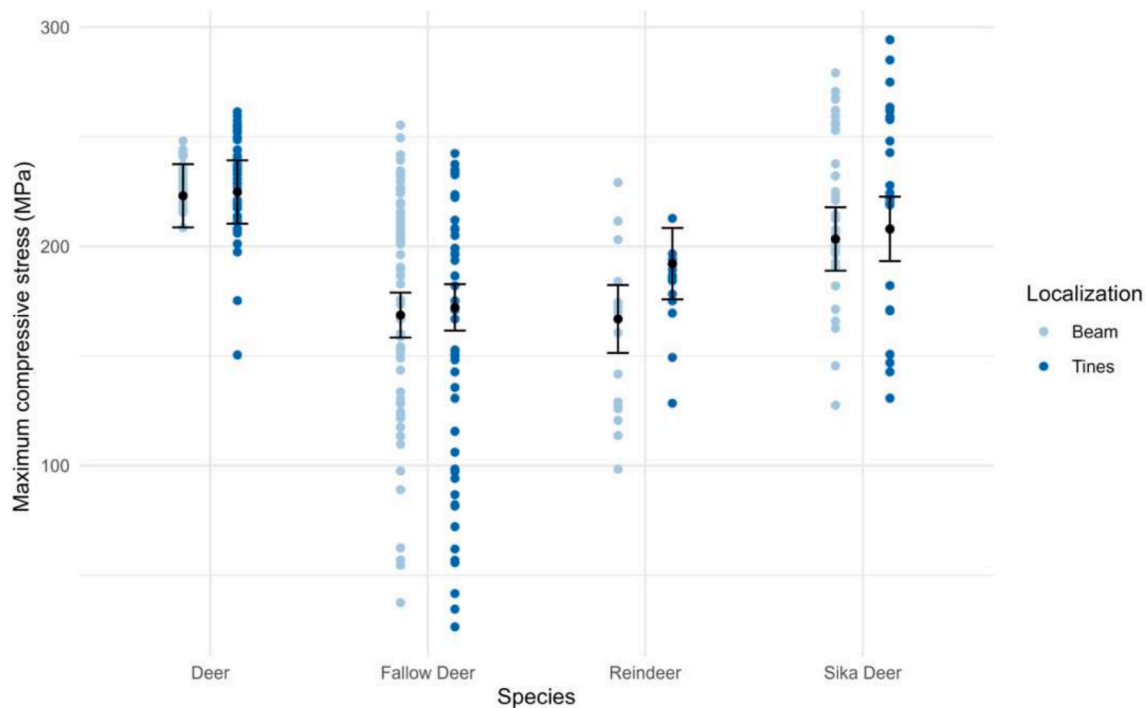


Fig. 4. Estimated mean (mean and standard error) σ_c of samples by species and location - N = 296. Points represent individual sample values.

Table 6
Results of the linear mixed model of the logarithmic transformation of maximum energy absorbed as a function of species and sample localization - N = 296.

Effect	Localization	Species	Coefficient \pm SE ^a	P-value
Localization (ref = beam)	Tines		-0.04 \pm 0.07	0.63
Species (ref = red deer)		Fallow deer	-0.37 \pm 0.24	0.14
Species (ref = red deer)		Reindeer	-0.60 \pm 0.29	0.062
Species (ref = red deer)		Sika deer	-0.33 \pm 0.27	0.25
Localization*Species	Tines	Fallow deer	0.10 \pm 0.10	0.34
Localization*Species	Tines	Reindeer	0.25 \pm 0.21	0.27
Localization*Species	Tines	Sika deer	0.17 \pm 0.11	0.16
Density			1036.92 \pm 184.66	<.0001

^a Coefficient \pm SE: Regression coefficient and standard error.

trabecular bone, a noteworthy aspect considering cortex’s pivotal role in bone mechanics (Currey, 2002; Davison et al., 2006).

The use of external strain measurement methods (extensometer, strain gauge, linear variable differential transformer or digital image correlation) is now recommended for bone compression testing (Zhao et al., 2018). The decision to use traditional machine crosshead displacement for strain measurement was driven by its quick and easy implementation, the specimens’ small size preventing the use of some of these external strain measurements methods, and the fact that machine compliance was found negligible thus avoiding potential accuracy limitations. However, we cannot exclude that results may have been impacted by this decision and that more differences between species could have been shown with the use of an external strain measurement method.

The decision to test cortical bone from dry deer antlers was grounded in the observation that antlers, when utilized in combat, are inherently dry (Currey et al., 2009; Landete-Castillejos et al., 2019). Dry antlers exhibit superior mechanical suitability for their role in combat compared to wet bones, demonstrating a combination of stiffness and

resistance to impact (Rajaram and Ramanathan, 1982; Currey et al., 2009a, 2009b; Chen et al., 2009).

Five intrinsic parameters were investigated in our study. It was shown that species had a significant effect on MCS and σ_c . Sampling localization had no effect on any of the parameters evaluated. Sample density had a highly significant effect on each of them. Studies have already shown that there was no effect of the sampling localization on antler for the elastic modulus (Blob and LaBarbera, 2001; Blob and Snelgrove, 2006) in white-tailed deer and moose. We confirm our hypothesis and this finding for red deer, Sika deer, reindeer, and fallow deer. These same authors suggested that other mechanical properties may also be insensitive to localization, and this has been confirmed by several subsequent studies with different testing techniques. We found the same results with this protocol. A localization effect has been found first for σ_c with the overall p-value but was adjusted via regression coefficients and contrasts. A possible explanation could be the lack of samples for reindeer in both antler localizations compared with the number of samples available for both localizations in the other species. Species influenced MCS and σ_c . For both parameters, fallow deer and reindeer had significantly lower values than those from red deer and Sika deer. This could maybe be partially explained by the differences in antler morphology and subsequent fighting behavior as suggested by Blob and LaBarbera, 2001; Blob and Snelgrove, 2006 attempting to explain the differences found between white-tailed deer and moose. Red deer, Sika deer or white-tailed deer have all in common a long antler beam with tines extending from this central beam. Cervidae species having palmate antlers with small tines like moose, fallow deer and at a lesser extent reindeer cannot interlock their antlers while fighting as do red, Sika and white-tailed deer.

Sample density had a significant impact on mechanical properties. Bone mineral density is known to play an important role in bone strength (An and Draughn, 1999; Schaffler and Burr, 1988; McCalden et al., 1993; Mosekilde, 1993). Mineral profile was not evaluated although it has been proven that mineral composition of the antlers (nutrition) may have an impact on mechanical properties and density of cortical bone (Landete-Castillejos et al., 2010). Antlers have the lowest mineral content among all bone tissues and a high content of collagen

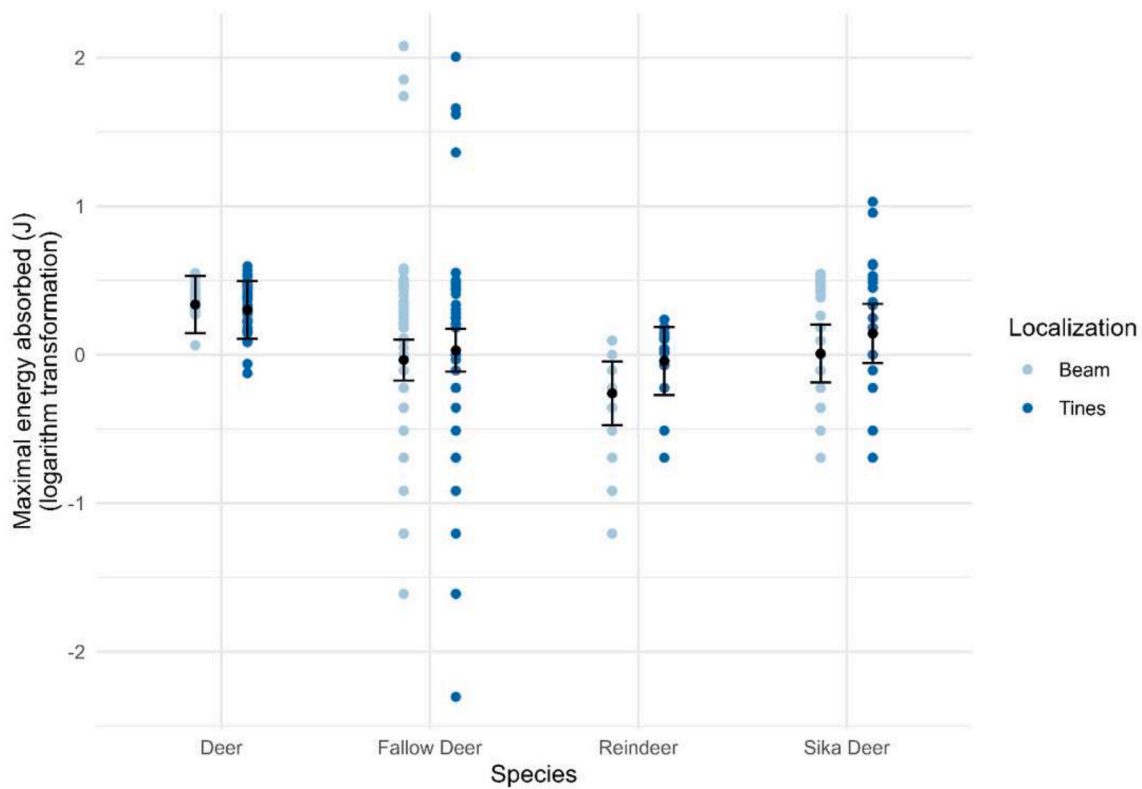


Fig. 5. Estimated mean (mean and standard error) of the logarithmic transformation of the MEA of samples by species and localization - N = 296. Points represent individual sample values.

Table 7
Results of the linear mixed model modeling modulus of elasticity as a function of species and sample localization - N = 296.

Effect	Localization	Species	Coefficient ±SE ^a	P-value
Intercept			-0.93 ± 0.43	0.053
Localization (ref = beam)	Tines		0.13 ± 0.08	0.17
Species (ref = red deer)		Fallow deer	-0.18 ± 0.24	0.47
Species (ref = red deer)		Reindeer	0.30 ± 0.30	0.34
Species (ref = red deer)		Sika deer	0.16 ± 0.28	0.58
Localization*Species	Tines	Fallow deer	-0.08 ± 0.12	0.53
Localization*Species	Tines	Reindeer	0.40 ± 0.25	0.15
Localization*Species	Tines	Sika deer	-0.20 ± 0.13	0.17
Density			4565.65 ± 220.11	<.0001

^a Coefficient±SE: Regression coefficient and standard error.

(especially collagen type I). This results in low-yield strength, and two to three times lower stiffness compared to human cortical bone (Currey, 2002; Launey et al., 2010), and a reported lowest elastic modulus (Currey, 1999; Rho et al., 1998). Species and localization had both a significant impact on sample density. Whereas variations of the density according to species can be expected, an effect of the localization on sample density is surprising. This unlikely impact could be attributed to a type I error.

There are numerous studies on the mechanical properties of cortical bone from human and animal sources. However, a large range in values is reported, which may be caused by interspecimen variability, differences in bone microstructure at different locations in the same bone, diversity in experimental conditions, etc (Attia and Willett, 2017). Also, the mechanical properties of cortical bone depend on the type of

mechanical testing and the loading directions of the testing method (An and Draughn, 1999). Our data may thus be slightly different from what has been published so far on antlers' mechanical properties and unsuitable for comparison.

This study has several limitations. The reduced number of individuals per species prevents us of making any definitive conclusion. In the future, multiplications of the individuals for each species in order to validate our data would be beneficial. Mineral composition has not been evaluated and could thus vary between each individual. Considering the significant impact that mineral composition can have on the mechanical properties, a thorough analysis must be conducted conjointly with the mechanical tests. A combination of testing procedures is necessary to characterize completely mechanical properties of antlers. A combination of bending, tensile/compressive and torsion tests are necessary to characterize the stiffness and strength of antler cortical bone. Our study focused only on the compressive mechanical properties. Finally, the protocol used here did not allow us to test several species. However, other methods of testing may have been also difficult if standardized guidelines are followed.

5. Conclusions

In conclusion, this research allows enriching the knowledge of the mechanical properties of dry antler cortical bone. Cylinders of cortical bone were tested in compression following current recommendations, highlighting that this type of testing is feasible in some species. Five intrinsic parameters are reported for red deer, Sika deer, fallow deer and reindeer: sample density, maximum compressive strength, maximum compressive stress, maximum energy absorbed and modulus of elasticity. Standardization of the tests used to characterize mechanical properties is advisable if comparisons are wanted. Data for fallow deer, so far unreported, are now available.

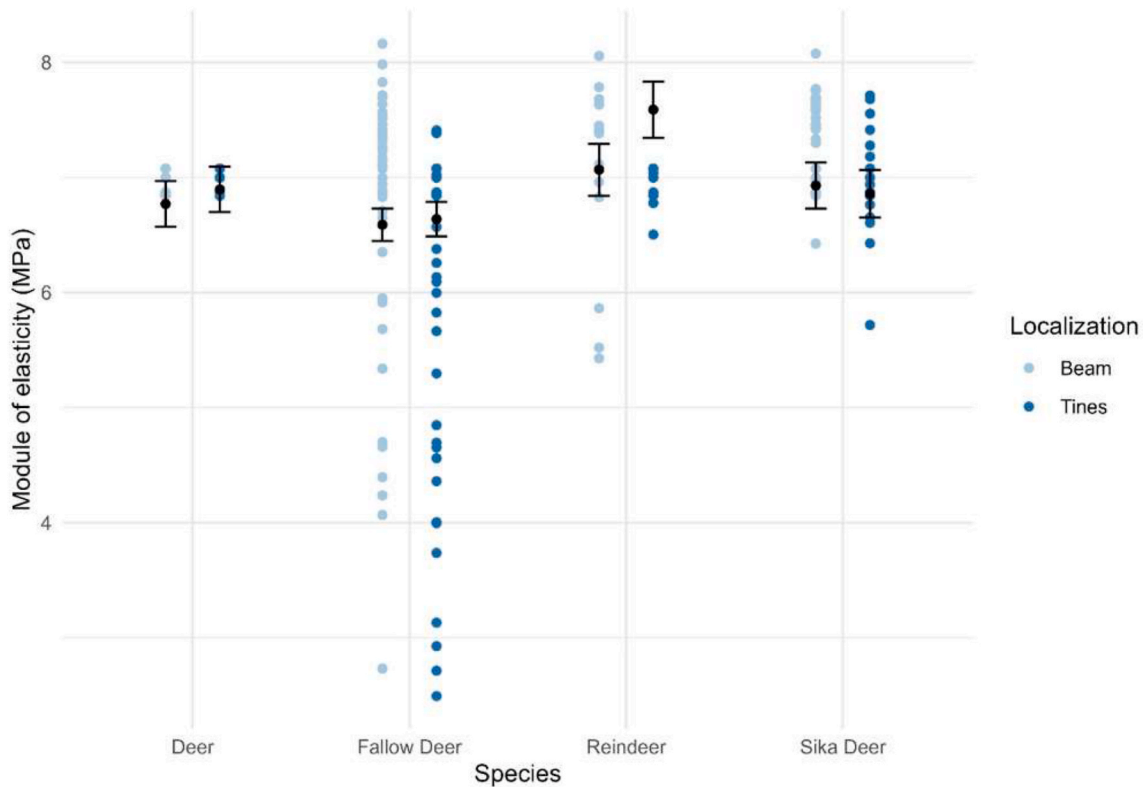


Fig. 6. Estimated mean (mean and standard error) EM of samples by species and localization - N = 296. Points represent individual sample values.

CRedit authorship contribution statement

Pierre P. Picavet: Writing – original draft, Methodology, Investigation, Formal analysis, Conceptualization. **Stéphanie Claeys:** Visualization. **Etienne Rondia:** Validation, Resources, Methodology. **Marc Balligand:** Writing – review & editing, Supervision, Resources, Funding acquisition, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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