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Article

A new methodology, based on a CAD system, for evaluating partial and global asymmetries in deer antlers from hunting quality assessment data

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Abstract: In the field of biology, the "biomarkers" of the different animal species are of great relevance, as well as the relationships between them and with the quality of the habitat. A study has been proposed, analysing the antlers of the Iberian deer, to measure the bilateral asymmetry of its branches. The antlers are characterised in a CAD 3D-model of their axial structure obtained by photogrammetric restitution from two photographs. This method quantifies the global and partial asymmetries of the antler's structural features. The research evaluated 48 deer antlers from hunting days and taxidermy workshops. To quantify asymmetries, the measurements required are the same as those considered to assess the hunting quality of antlers. Bilateral deviations are estimated for each structural trait and for the whole antler. Preliminary results showed a convergence between the values of global asymmetry and hunting quality. The methodology proved to be fast, inexpensive and easy to implement, which facilitates its use in future asymmetry studies of other biological elements of animal or plant origin. It is only necessary that the starting data, the bilateral axial structure of the analysed element, is characterised in a 3D-model.

Keywords: Deer antler; Asymmetry value; Hunting quality; Biological applications; CAD 3D-model; Landmarks; 3D digital techniques; Computer graphic design.

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1. Introduction

In ungulate species the degree of stable development in an animal is determined by internal, genetic and external factors, level of pasture in the environment, relationships with other species, etc.; some of these may alter the development of the animal, causing geometric defects in certain morphological features of the antler. The antlers of ungulates are distinguished in that, in some species, such as bovids, their horns remain stable over time, and in other species, such as cervids, the antlers are cast off after the rutting season and are renewed every year. Some authors [1,2,3] point out the morphological and evolutionary differences between the antlers of both species of ungulates. Other authors [4,5,6] indicate the factors influencing the growth of cervid antlers and the 'shedding' period. The state of the antlers after the annual renewal represents a relevant biomarker that can be related to the influencing factors to which the specimen has been subjected. The function of the antler as a secondary sexual character and with biomechanical properties adapted to fighting during the rutting period are therefore greatly affected by environmental factors, and it has even been observed that the year of birth is a determining factor for the future antler quality of the individual [7].

One of these alterations is defined by several authors as 'fluctuating asymmetry' (FA), representing those random deviations in the symmetrical development of certain features of bilateral structures [8,9]. Studies on fluctuating asymmetry (FA) have been initiated on invertebrate, lower vertebrate and avian species [10,11,12]. Most studies have been carried out on the estimation of the degree of asymmetry in ungulate species, such as moose (*Alces alces*) [13], fallow deer (*Dama dama*) [14,15], reindeer (*Rangifer tarandus*) [16], and red deer (*Cervus elaphus hispanicus*) [17], as well as roe deer (*Capreolus capreolus*) [18], supporting the relationship between FA and stress of the animal caused by adverse environmental conditions in the year of antler growth or earlier.

Several studies assess the degree of bilateral divergence of animals and the causes of developmental instability as a function of different animal stress conditions [19,20,21,22,23]. In Iberian red deer [24] observed a direct correlation between the right and left branches of deer antlers being more significant in the thicknesses than in the lengths and number of tines, and [25] relate this asymmetry variable to age.

The study of FA in the antlers of iberian red deer, since they are renewed annually, can be a good indicator of their individual quality and the degree of environmental stress they have suffered during their development [17].

Several studies question the reliability of the results regarding the relationship of asymmetry with other variables: [25,26] state that studies with large sample sizes are needed to obtain conclusive results; others emphasize that methodologies with controlled measurement errors should be applied [22,27,28] and question the relationship between asymmetry and animal stress because the results obtained in numerous analyses are not consistent and homogeneous [23]; or question the fact that antler development is a factor in female selection [29,30].

The methodologies for asymmetry studies in cervids should be more rigorous, based on the study of those particular antler traits that represent the causes of developmental instability, thus causing asymmetry in the two branches of the antler [31]. Some authors emphasize the importance of directing asymmetry studies towards the analysis of particular geometric features of the antler, thus [17,22,23] establish partial asymmetry values in differential features of the antler, allowing us to detect and relate those features which are more exposed to stress so that the conclusive results are more rigorous in studies on the instability of the development of the antler. [31] relates the degree of FA in different antler traits and at different age intervals. Contrary to other studies, he states that FA caused by developmental instability is more pronounced in older males. In early stages there is little degree of asymmetry, however if asymmetry is detected in certain traits it remains in more mature stages of age. The most frequent studies on asymmetry in cervid antlers have consisted of assessing asymmetry in particular traits of the antlers. Thus, numerous authors have studied the degree of asymmetry, not as a composite value, but in several traits, from a few [13,14,18] to a large number [25,30,32]. Studies that offer a composite value for asymmetry are less frequent [17,33]. Similarly, [21,34] study the relationship of asymmetry with hunting quality values obtained as hunting trophies in Alaskan moose species and white-tailed deer (*Odocoileus virginianus*) in North America.

Traditional methodologies have been based on partial asymmetry assessments of some particular traits of the biological element under study and not on global asymmetry assessments for the whole structure. This is due to the difficulty in taking the necessary measurements and their precision regarding all the traits that form the structure, in order to finally obtain a global value of asymmetry.

Some authors [17,22,23] emphasize the importance of studying particular traits of the antlers and their relationship with the degree of stress of the animal.

The method presented allows extending this study on different traits of the antler structure and their relationships with the stress of the animal. The new methodology allows to extend this study and to infer later the causes of influence.

More recent studies have evaluated asymmetry values, not as an overall value but as partial values for differential traits of antler structure [13,14,18]. To a lesser extent, an overall value of asymmetry is evaluated and estimated for the entire antler [17,33].

The new method allows estimating partial values of asymmetry for those traits of antler structure taken into account in the evaluation of the biomarker of hunting quality. That is to say that with the same values it is possible to evaluate both biomarkers. It also allows estimating a global value of asymmetry for the whole antler.

Regarding the different methodological typologies for estimating the value of asymmetry, some authors [12,13,17,30] have created methods and tools for quantifying the asymmetry of biological elements using projective techniques of linear measurement in two dimensions. Bartoš et al. in 2007 [32] estimate the degree of asymmetry using three antler characteristics: weight, length and number of tips, without relying entirely on geometric values.

Ditchkoff and Defreese [35] use three-dimensional methodologies for the geometric definition of antlers for the subsequent analysis of asymmetries. The proposed method defines the antlers in three dimensions in order to estimate, in the same way, the values of partial asymmetry in its structural traits, without considering horizontal or vertical symmetry planes referenced on the antlers themselves.

Other authors [36] proposed a method for determining the global asymmetry of the entire antler. For this purpose, he uses the antler data characterised by a CAD 3D-model of its axial structure. In this case, the deviations of the extreme points of the tines of the antlers of both branches with respect to the reference of a symmetry plane linked to relevant points of the skull of the specimen are considered.

The new methodology presented in this study allows a more direct application, since it does not consider the previous definition of a symmetry plane. It will consider the divergences of the measurements on average of different related structural traits of the two branches of the antler of the Iberian deer (*Cervus elaphus hispanicus*). With these values of partial asymmetries, a global value of asymmetry for the whole antler structure is finally obtained. For the analysis, the characterisation of the antler is used as a 3D-model of its axial structure. The algorithm takes advantage of the measurements considered for the assessment of the degree of hunting quality [37].

The new method will provide a number of relevant advantages over other methods:

- For the analysis, the characterisation of the structure of the antler is used as input data by means of a CAD 3D-model, obtained by photogrammetric restitution from few data, only two photographs.
- The 3D-model obtained is accurate and therefore the estimated deviations on the two branches of the antler will also be accurate, finally obtaining reliable values of the degree of asymmetries.
- It is not necessary to refer the deviations, estimated between the two bilateral structures of the antler, to a previously defined symmetry plane.
- As the CAD definition of the antler by means of a 3D-model is relatively fast, it will allow further analyses with larger sample sizes, so that conclusive results and degrees of correlations with other biomarkers and influencing factors can be obtained.
- It will allow estimating a 'global' value of the degree of asymmetry for the whole antler.

Estimating a global asymmetry value is of great difficulty due to the large number of measurements that must be considered at significant points corresponding to all traits of the antler structure. For this reason, most of the previous methods have estimated asymmetry values of specific traits of the antler.

The proposed method resolves all of these drawbacks, since it allows us to quickly measure, on the CAD previously defined 3D-model, the values of the deviations in the traits of the two branches of the antler (partial asymmetries) and, based on these, to quantify a 'global' asymmetry value for the whole antler.

- The proposed method also makes it possible to take advantage of the same measurements used in the assessment of the hunting quality of the antler. These correspond to the geometric deviations produced in the structural traits of the two antler branches. Therefore, with the same measurements two biomarkers can be estimated: the degree of hunting quality and the values of partial and global asymmetry.

Other subsequent studies, of biological and ecological interest, will be aimed at relating the values obtained for this biomarker to other influential factors.

2. Materials and Methods

In order to validate the methodology, a set of 48 red deer specimens was used: 21 of them from three sessions of official approval processes as hunting trophies (homologation sessions) and taxidermy workshops, and 27 specimens shot in two hunting days in hunting areas between 2014 and 2019, aged between 5 and 9 years, in the Sierra de Andújar Natural Park, in southern Spain.

2.1. Equipment used

For all the specimens studied photographs were taken from different points of view and in which a metric reference appears, panel sized in DIN-A3 format (594x420 mm), in order to enable photogrammetric restitution [38] to obtain the 3D spatial representation of the antler represented by axial structure. Photographs were taken with an SLR reflex camera *Nikon 300D*, resolution 4,288 x 2,848 pixels, with 28-80 mm zoom lens. Exposure was kept at values of around 35 mm during photography. In order for the photogrammetric restitution process to be reliable and accurate, it was verified that the photographs taken with SLR cameras showed less deformation on the centre and periphery than those taken with compact cameras, without the use of expensive metric cameras or subsequent editing and adjustment treatments of the photographs to correct deformations.

In terms of hardware, no major resources are required. The graphics card used is an '*Nvidia GeForce RTX*' with 6 Gb of memory. In studies with CAD 3D-objects it is recommended that the graphics card be independent of the motherboard and has a large amount of memory in order to achieve good performance in image processing.

The 3D-modeling software '*SolidWorks v.2020*' from *Dassault Systèmes* was used for the restitution of the antler, represented in the two photographs, by creating a 3D-scenario for the definition of the 3D-model of the antlers and for the measurement of the different geometric parts of its axial structure.

For the manual measurements of the different parts of the antler, necessary for the hunting valuation process, it is only necessary to use a flexible tape measure and adapt it to the different external surfaces of the antler as indicated in previous studies [37].

2.2. Object of the study

For the new proposed methodology, the study of deer antler asymmetry, it is necessary to start from a model that represents the structure of the antler, so that the lengths and perimeters of the different structural parts of the antler considered can be measured on it.

The process of defining the 3D-model of the antler, using a photogrammetric restitution methodology from two photographs of the antler taken from different angles, is described in [38] (Figure 1). Similarly, the digital CAD geometry of the antler is also validated on the physical model of the antler, demonstrating its degree of approximation and geometric accuracy.

In the proposed analysis the starting data is the characterisation of the antler in a CAD 3D-model of its axial structure, which faithfully describes the geometric elements of its structure: main beam, main tines (eye tines and trez tines) and the cross sections of relevant parts of the main beam of the antler (Figure 2).

Figure. 1. Deer antlers represented by two photographs and 3D-model with axial structure and cross sections.

2.3. Geometrical data of the antler structure

For the present study different relevant structural parts of the antler geometry, represented by its axial structure, will be considered in order to quantify the length of each of them (Figure 2).

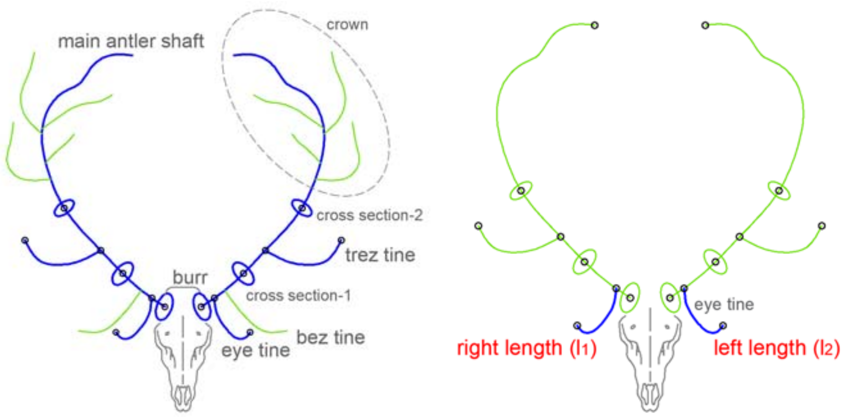

Figure 2. Geometric elements of the antler considered for the study: main beam, eye tines, trez tines and cross sections. Lengths considered (right and left) in the structural trait considered (eye tine) for the estimation of the (PAI) value.

The geometric elements considered for each of the two branches of the antler are: the 'main beam', the 'eye tine', the 'trez tine', the 'burr' and the two cross sections of the 'main beam', one taken between the 'eye tine' and the 'trez tine', and the other between the 'trez tine' and the 'crown'.

2.4. Description of the proposed Methodology

The proposed methodology allows the estimation of the degree of antler asymmetry, which represents the symmetry defect between the two branches of the antlers of the deer studied, using measurements taken in previous studies to quantify the hunting quality of the antlers of the specimen.

Among the different items used in the official scale [37] for the evaluation of hunting quality, we considered those related to quantifiable distances, three lengths and three perimeters, belonging to the structural features of the antler in each of the two branches of the antler, mentioned above (Figure 2).

The method allows estimating the asymmetry value, represented by the variable (PAI) (partial asymmetry index) for each of the six estimated geometric traits of the antler and a global asymmetry value for the whole antler, defined by the variable (GAI) (global asymmetry index).

In this proposal the measurements pertaining to the same geometric trait of the antler under consideration taken on each of the two sides of the antler were compared; the difference between the two measurements was then evaluated and compared with the average length of the antler element studied (Figure 2) (Equation 1).

The algorithm implemented in the methodology will actually estimate the relative error representing the geometric divergence in the structural traits of the antler under study. The two measurements of the studied structural trait, of the right and left branches of the antler, are usually close. Therefore, the resulting numerical value is usually a decimal value between 0-1, which will be expressed with a precision of three decimal places.

The numerical value obtained (Equation 1), will be dimensionless, without units, since it represents the metric difference between two related elements referred to the average value of these.

$$(PAI)(eye\ \ tine) = \frac{|l_1 - l_2|}{\frac{(l_1 + l_2)}{2}} \quad (1)$$

(PAI)(eye tine), represents the 'partial asymmetry index' for the specific trait studied; the 'eye tine'.

Applying this procedure, the values of asymmetry for each of the six traits considered corresponding to the geometric elements of the antler are obtained. The global asymmetry value for the antler, (GAI) (global asymmetry index) is taken as the weighted sum of each of the six traits of (PAI).

In order to establish the partial weights assigned to the six structural traits of the antler, the weight assigned in the official scale of evaluation of the degree of hunting quality as a hunting trophy [37] to each of the traits of the structure of the antler was considered.

The quantification of these weights has been referenced on the specimen with the highest hunting quality, taken as a reference, of the sample studied. This specimen gad a score of 187 points (gold medal), compared to the scores obtained (138.7) in the six antler traits of interest in the study of asymmetries. The partial weights assigned to each of the six antler traits (Table 1) were determined based on these scores.

In the evaluation of hunting quality, apart from the items that express characteristics of the geometric development of the antler other items are considered, some of which are subjective, such as ‘wingspan’, ‘total number of tines’, and ‘weight’, ‘separation’ of the antlers without skull. These are not considered for the estimation of partial weights.

Table 1. Weights of each antler’s structural traits for the calculation of (PAI).

structural Trait of the antler	**Score 'Hunting valuation'**	**Weight (%)**
main beam	48	**34.6**
eye tine	7	**5.1**
trez tine	7.28	**5.2**

burr	23	**16.6**
cross section-1 (eye tine-trez tine)	27.2	**19.6**
cross section-2 (trez tine-crown)	26.2	**18.9**
Total	**138.7**/187	100

Once the weights represented by each estimated trait have been established, the calculation of the overall asymmetry value for the antler (GAI) is represented by the weighted sum of the six traits considering the weight represented by each one of them (Table 2) (Equation 2).

Table 2. Calculation process of the '*Global Asymmetry Index*' (GAI).

structural Trait of the antler	(PAI)	Weight (%)	**(PAI) weighted**
main beam	X1	34.6	$\mathbf{Z_1}$
eye tine	X2	5.1	$\mathbf{Z_2}$
trez tine	X3	5.2	$\mathbf{Z_3}$
burr	X4	16.6	$\mathbf{Z_4}$
cross section-1 (eye tine-trez tine)	X5	19.6	$\mathbf{Z_5}$
cross section-2 (trez tine-crown)	X6	18.9	$\mathbf{Z_6}$
			(GAI) value

$$(GAI) = \sum_{i=1}^{6} (Z_i) \quad (2)$$

The (PAI) and (GAI) values quantify the asymmetry defect of the antlers. For a given trait, a '0' PAI value indicates no asymmetry; values close to zero represent reduced degrees of asymmetry.

This methodology uses the same measurements taken on the antlers to study their hunting quality. The proposed methodology allows its application on the 3D-model of the antler or on the physical model of the antler.

3. Results

The process of calculating the values of (PAI) and (GAI) for each of the antlers is detailed in (Table 3). These results are derived from the same measurements used for the estimation of hunting quality, metric values in (cm) of the six structural features of the antlers considered. The results belong to the specimen of the sample, taken as reference, which presents the highest hunting quality, 187 points (gold medal).

Table 3. Process of calculation of (PAI) and (GAI)

(PAI) and (GAI) values (reference deer specimen)				
Starting Data **(Hunting valuation):** Lengths (cm)		**(PAI)'s** Partial Asymmetry Indexes		**(GAI)** Global Asymmetry Index
Right Main Beam	97.6	(PAI)-(Main Beam)	**0.015**	34.6%
Left Main Beam	94.8			
Right Eye tine	26.9	(PAI)-(Eye tine)	**0.039**	5.1%
Left Eye tine	29.1			
Right Trez tine	28.7	(PAI)-(Trez tine)	**0.014**	5.2%
Left Trez tine	29.5			
Right Burr	22.7	(PAI)-(Burr)	**0.013**	16.6%
Left Burr	23.3			
Right Cross Section-1	13.5	(PAI)-(Cross Section-1)	**0.007**	19.6%
Left Cross Section-1	13.7			
Right Cross Section-2	13.1	(PAI)-(Cross Section-2)	**0**	18.9%
Left Cross Section-2	13.1			
		Total Value **(GAI)**		**0.011**

The assessment analysis of the partial asymmetry (PAI) and global asymmetry (GAI) values of all the specimens evaluated was carried out on the input data of the antler characterized by its CAD 3D-model (Table 4). The results show the arithmetic mean of the (PAI) and (GAI) values in the specimens of each of the samples, corresponding to the three 'homologation sessions' and the two 'hunting days'.

Table 4. Results of 'PAI' and 'GAI' in antlers as 'CAD 3D-model'.

Antlers as CAD Model-3D measurements								
('Homologation' sessions')								
	PAI						**GAI**	**Hunting Quality** (score)
	Lengths			Perimeters				
	Main Beam	Eye tines	Trez tines	Burrs	cross section1	cross section2		
(2014-03-27)	0.012	0.045	0.039	0.022	0.024	0.025	**0.022**	177
(2015-04-06)	0.015	0.034	0.042	0.025	0.014	0.020	**0.020**	167
(2017-10-25)	0.016	0.042	0.083	0.028	0.026	0.020	**0.026**	150
Mean	**0.014**	**0.040**	**0.055**	**0.025**	**0.021**	**0.022**	**0.022**	
cross section-1: (between Eye tine-Trez tine) cross section-2: (between Trez tine-Crown)								
('Hunting sessions')								
	PAI						**GAI**	**Hunting Quality** (score)
	Lengths			Perimeters				
	Main Beam	Eye tines	Trez tines	Burrs	cross section1	cross section2		
(2014-10-23)	0.022	0.051	0.082	0.075	0.025	0.029	**0.037**	150

(2019-12-18)	0.009	0.045	0.075	0.077	0.057	0.038	**0.040**	153
Mean	**0.015**	**0.048**	**0.078**	**0.076**	**0.041**	**0.033**	**0.039**	

The 'GAI' values obtained on CAD-measurements are between 0.020 and 0.040.

4. Discussion

The (PAI) and (GAI) values detect some relationships on the results obtained in the different structural traits studied and on the origin of the specimens, from 'homologation sessions' or 'hunting days'. The convergence between the preliminary results obtained in the asymmetry biomarker and the hunting quality biomarker is also highlighted.

The value of 'GAI' is lower in the specimens from the 'homologation sessions' as hunting trophies; 0.022 of mean value, 44% lower than the mean value obtained from 0.039 of those specimens from hunting days. This is due to the fact that the antlers of the deer from the 'homologation sessions' present a higher degree of hunting quality, and are therefore selected by their owners to be treated in taxidermy workshops for their definition as hunting trophies.

With respect to the asymmetry value evaluated in the different traits of the antler structure, 'PAI', it has been concluded that over the three lengths considered the lowest 'PAI' values, with mean values between 0.014 and 0.015, correspond to the length of the 'main beam' of the antler, thus representing the most symmetrical element. The next element of the antler, somewhat less symmetrical, corresponds to the 'eye tine', with mean 'PAI' values of between 0.040 and 0.048; the least symmetrical element is the one corresponding to the 'central tine or trez tine', with mean values ranging between 0.055 and 0.078.

With respect to the perimeters of the 'burrs' and of the two cross sections considered in structural parts of the antler, it is observed that the 'burrs' present the greatest asymmetry defect, with mean values of 0.025 in specimens from 'homologation sessions' and 0.076 in specimens from hunting days. The mean values of 'PAI' at the perimeters of the cross sections considered is 0.0215 in antlers from 'homologation sessions' and 0.037 in those from hunting days, 72% higher.

Finally, an approximation can be established for the 'GAI' value represented by a single 'PAI' trait out of the six that have been considered represented by different geometric parts of the antler. As can be seen in (Table 4) the 'overall asymmetry' 'GAI' value of the antler is very closely approximated by the 'partial asymmetry' 'PAI' value corresponding to the perimeter of the cross section taken between the 'central tine or trez tine' and the 'crown' of the antler; therefore, in a first approximation the overall 'GAI' value could be represented by the 'PAI' value for that geometric trait due to the great convergence of these values.

The advantages of the proposed methodology are based on its ease of implementation and the economy of resources required. Therefore, its implementation may be appropriate in the study of biomarkers, asymmetries and hunting quality of large populations and samples, such as the case of dead animals shot on hunting days [39] on selected specimens and with samples from different sources. Subsequently, these results can be related to other influencing factors. Furthermore, this new methodology could extend the study of naturally dead and older specimens [40]. The asymmetry relationship of various antler structural traits with respect to other mean antler values could be demonstrated. The degree of consistency of the relationships of partial asymmetries with age could also be studied. The proposed method allows the study of particular antler traits where the causes of instabilities in antler development at various age intervals can subsequently be detected [31].

This proposed methodology will also allow us to relate two very relevant biomarker variables in biological studies, the degree of hunting quality and the global asymmetry 'GAI' of the antler. Based on the data obtained, a strong direct relationship has been obtained between the value of 'hunting quality' and that of asymmetry; the specimens from the 'homologation sessions' present high values of 'hunting quality' (Table 4) together with low values of 'GAI' representing low defects and, therefore, high degrees of symmetry. The specimens that are evaluated in 'homologation sessions' are those whose hunting quality is recognized at first sight in terms of their degree of geometric development, number of points, colour, etc., being selected by hunters for their valuation and subsequent conservation as hunting trophies.

Therefore, this new variable 'GAI' is proposed as a 'biomarker' that quantifies the degree of antler symmetry defect on the two branches of the deer antlers so that future studies will be able to establish relationships and causalities with other internal and/or external environmental factors in which the specimens have developed.

Subsequent studies of populations of individuals, with larger samples, will allow inferring the results obtained with other internal variables (biomarkers) such as age, antler density and the degree of geometric development determined by hunting quality, or other external variables such as the level of pasture in the specific place, the level of rainfall, the relationship of the environment with other animal species, etc., in which the population has developed.

Other studies may relate the asymmetry of specific structural traits of the antler to age or to geometric variables such as wingspan or hunting quality.

The species analysed in order to define the proposed methodology is the red deer (*Cervus elaphus hipanicus*). Given that the proposed method is initially based on the measurements taken for the valuation of hunting quality, based on quantifying the differences in the lengths and perimeters of the geometric features of the right and left branches of the antler, the applicability of the proposed methodology could be generalized to other species indicated in the 'Manual de Homologación de Trofeos de Caza Mayor en España' [37]: to other cervids, such as fallow deer (*Dama dama*) and roe deer (*Capreolus capreolus canus*) and other ungulate species, such as wild boar (*Sus scrofa baeticus*), mouflon (*Ovis orientalis musimon*), mountain goat (*Capra pirenaica victoriae*), barbary sheep (*Ammotragus lervia lervia*) and chamois (*Rupicapra pyrenaica parva*).

For free-ranging specimens that develop in areas or habitats of countries that protect specific cervid species, such as the huemul (*Hippocamelus bisulcus*) in Argentina, the application of this methodology is complicated for conservation and species management reasons. To obtain the metric measurements of the antlers, for the estimation of asymmetries, it is necessary to have the physical antlers or their characterization in the form of a CAD 3D-model from two photographs. The photographs of the antler, used as starting data to obtain the 3D-model, must include a metric reference that makes the photogrammetric spatial restitution process possible. With free-ranging specimens, this process is very complicated to carry out and not very efficient due to the costly infrastructure and controlled equipment requirements, such as, for example, well-balanced photo-trapping cameras with validated scaling.

Therefore, in estimating the biomarker of asymmetry in bilateral cervid antler structures, the implementation of the methodology in live animals is complicated. In protected areas, where specimens in danger of extinction or other causes are conserved, the methodology could be applied to specimens found dead. In protected species, on specimens found dead, the proposed methodology would provide additional information in complete analyses, such as necropsy procedures, implemented to determine the causes and other data of interest on the circumstances of the specimen's death. The asymmetry data obtained can be used later to infer with other estimated biomarkers, but conditioned by the favorable environmental setting. Another case of obtaining the physical antler can be by obtaining the antler branches, 'cast-off', collected in the field at the end of the winter season when cervids shed their antlers annually, as do studies in which the cast-offs are used for different studies, either genetic [41] or to validate methodologies for calculating antler volume and density [42].

But this is complicated by the difficulty of finding and associating the two antler branches to the same specimen. Another drawback the consistency of the results in the study due to the possibility of sample size limitation.

4.1. Comparison of methodologies.

The results obtained, with the proposed methodology 'Met-2' were compared with those obtained with another previous methodology proposed by the same author [36], called 'Met-1'. It is observed (Table 5) that the mean AI values (asymmetry index) obtained with ''Met-1, were 0.063 for specimens studied in 'homologation sessions', 43% lower than for specimens from hunting days, with a mean value of 0.110; indicating a higher symmetry in specimens from 'homologation sessions'. The same proportion occurs in the results applying 'Met-2', 0.024 and 0.039, 39% lower, so that the results of the two methodologies converge, each in its own scale of values.

Table 5. Results of 'AI' (Met-1) and (Met-2) with data on the 'CAD 3D-Model'.

Results (AI): (Met-1) and (Met-2) on the 'CAD 3D-Model'				
	Met-1		Met-2	
	AI	Mean	**GAI**	Mean
Homologation session (2014-03-27)	0.056		0.022	
Homologation session (2015-04-06)	0.065	**0.063**	0.024	**0.024**
Homologation session (2017-10-25)	0.068		0.028	
Hunting day (2014-10-23)	0.085	**0.110**	0.037	**0.039**
Hunting day (2019-12-18)	0.134		0.040	

On the new proposed methodology, 'Met-2', the results obtained are convergent with those obtained in 'Met-1', each in its own scale of results. The values of asymmetry in the specimens from 'homologation sessions' are somewhat lower over those from 'hunting days'; 57% in 'Met-1' at a and 61% in 'Met-2'.

Due to the great convergence of results obtained with the two methodologies, 'Met-1' [36] and this new proposal, 'Met-2', in the assessment of the degree of antler asymmetry, it will be necessary to decide, depending on the scope of the study, which of them will be applied.

The decision as to the implementation of one or the other method will be determined firstly by the type of data available, whether from CAD or manual measurements with a tape measure on the physical model, secondly by the number of variables to be estimated, estimates of the degree of hunting quality, the degree of asymmetry or both, and thirdly by the size of the population of specimens to be studied.

If only the asymmetry study is desired the application of the 'Met-1' is optimal, since it only requires the spatial restitution of the pairs of points necessary for the study represented by the start and ends of the main points and the crown of the antler. 'Met-2' is more appropriate when it is desired to study the hunting quality, and to take advantage of the measurements for this study pertaining to the lengths and perimeters of the geometric elements of the two branches of the antler to perform the study of the asymmetry, either globally 'GAI' or partially 'PAI', in order to then carry out the study of each particular feature of the antler.

The following is a comparison between both methods, where the characteristics of each of them, their functionalities and the suitability of their implementation according to the type of biological application are defined.

1. Characteristics of the methodology:
   'Met-1': We need to define a 'plane of symmetry'; defined by three points on the skull. The asymmetry study is performed by comparing the perpendicularity and the distances on the plane of symmetry of the segments joining the pairs of related points of the antler considered.
   'Met-2': No need to define a 'plane of symmetry'. The proposed methodology consists of comparing the lengths of the different geometrical traits considered corresponding to the two branches of the antler.
2. Characteristics of the initial data:
   Met-1: Only admits that the starting data is in digital format; its spatial position is obtained from the points considered for the study, represented in two photographs, by means of photogrammetric spatial restitution.
   Met-2': The evaluation of the degree of asymmetry is based on the results of the measurements, lengths and perimeters, carried out for the evaluation of the hunting quality. These measurements can be obtained on the physical model of the antler by tape measure, or be defined in a CAD 3D-model representing the structure of the antler.
3. Obtaining process:
   Met-1': It is not necessary to obtain the entire CAD structure of the antler by photogrammetric restitution. It is only necessary to render in 3D the relevant 'homologous' points of the antler necessary for the study: the start and ends of the main tips and the centre of the crown corresponding to the two branches of the antler.
   Met-2': It is necessary to define in 3D the whole antler by representing its axial structure formed by the main beam, the main tines and the geometrical center of the crown tines.
4. Biological applications:
   'Met-1': It is appropriate if it is only implemented for the assessment of the ‘degree of asymmetry' as this requires more simplified, smaller and quicker data to obtain. Without fully characterizing the structural trait analysed by means of a 3D-model, it is sufficient to define the three-dimensional spatial position of the significant points of the structural feature evaluated; all this is achieved by means of a 3D photogrammetric restitution process of these points represented in two photographs. Recommended for the study of a large set of specimens, 'populations', located at a specific place.
   'Met-2': Appropriate in studies of 'hunting valuation' and of the global 'degree of asymmetry' or of specific geometric traits of the antler; in this case the study consists of knowing the two variables and being able to determine their degree of correlation. It can be implemented on specimens from ‘homologation sessions’ or groups of specimens from the same hunting day.

5. Conclusions

The present study validates the usefulness of CAD measurements taken on deer antler structure to assess the degree of asymmetry in antler structure traits. The algorithm uses the same measurements used in the evaluation of hunting quality to estimate the values of antler asymmetry. The same measurements will allow the estimation of two biomarkers. The input data for the analysis is the characterisation of the antler by means of a CAD 3D-model represented by its axial structure. This is achieved by spatial restitution of two photographs, taken from different angles.

The implementation of the methodology is relatively fast; on the physical model of the antlers, it will only be necessary to make the calculation from the available measurements that were previously used to perform the evaluation of the hunting quality. For the definition of the 3D-model of the antler, the time invested is somewhat longer, due to the photogrammetric restitution and CAD modeling processes.

The antlers, digitally characterised using a 3D-model, allow analyses to be carried out in order to estimate the degree of asymmetry of deer specimens from 'homologation sessions' or in larger samples of specimens from 'hunting days'.

The methodology presented here can also be implemented in asymmetry studies of other biological elements of animal or plant origin, since the starting data, the bilateral axial structure of the geometric features under study, are relatively simple to obtain and to digitally characterise in the form of a 3D-model.

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Data Availability Statement: The data supporting the findings of this study are available from the corresponding author upon request.

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Institutional Review Board Statement: All specimens used in this study came from authorized hunts during the official hunting season in the study area. Current regulations on animal protection and bioethics were complied with. This study complied with all Andalusian, Spanish and European legal requirements and guidelines regarding experimentation and animal welfare. It is developed by the 'Animal Experimentation Ethics Committee' of the 'University of Jaén' and is authorized by the 'General Directorate of Agriculture and Livestock of the Department of Agriculture, Fisheries and Environment' of the Regional Government of Andalucía (Junta de Andalucía). No special permits from the bioethics committees were required. The datasets and specimens of the current study, stored at the University of Jaén, may be available upon reasonable request to the corresponding author and head of the Research Group PAI-RNM-175 and Vertebrate Laboratory.

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