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The evolution of body size, horn shape and social behaviour in crown Antilopini - an ancestral character state analysis

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Abstract

The earliest known bovids, commonly placed in the genus *Eotragus*, are small species with short straight horns that are located above the orbits. Among living bovids there are several species that show a similar horn morphology. These dwarf antelopes were historically united in the group „Neotragini“, which is now known to be a polyphyletic assemblage. The species in the genera *Ourebia*, *Raphicerus*, *Dorcatragus*, *Madoqua*, *Oreotragus*, *Nesotragus*, and *Neotragus* are scattered across the bovid phylogenetic tree. Nevertheless, the shared similarities, not only in body size and horn length but also in their social organisation, are interpreted to represent shared plesiomorphic traits that were present in the most recent common ancestor of Antilopinae. Most dwarf antelopes are closely related to gazelles and their allies, placed in the taxon Antilopini. Using a species level phylogeny of this group, I reconstructed ancestral states of several characters for the most recent common ancestor of Antilopini. While the results for the reconstruction of ancestral social organisation of the dwarf species are inconclusive, the small body size and short horns of *Ourebia*, *Raphicerus*, *Dorcatragus*, and *Madoqua* are most likely derived traits. The habitus of the ancestor of Antilopini was probably very similar to modern gazelles, consistent with the placement of the earliest fossil Antilopini in the genus *Gazella*.

Key words: Miniaturisation, Bovidae, dwarf antelopes, Bayesian ancestral state reconstruction.

1. Introduction

Antilopini are one of the major clades of horned ruminants (Bovidae, Artiodactyla). They comprise many gazelle-like animals, like the Asian *Procapra* and the African *Gazella*, *Eudorcas*, *Nanger*, and *Antidorcas marsupialis*. The other members of the clade are less well known, including the Asian Saiga antelope *Saiga tatarica* with its proboscis, the Indian blackbuck *Antilope cervicapra* with its spiral horns, and the East-African Gerenuk *Litocranius walleri* and Dibatag *Ammodorcas clarkei* that are both adapted to browsing bipedally on high shrubs and trees and have very elongated necks and legs. There are also several dwarf antelopes included in this clade, like the Oribi *Ourebia ourebi*, Steenbok and Grysboks of the genus *Raphicerus*, the Beira antelope *Dorcatragus megalotis* and the Dikdiks of the genus *Madoqua*. These species are commonly thought to represent an ancient type of bovid body plan, with small size and short straight horns (Kingdon 1989a; Gentry 1992; Kuznetsova & Kholodova 2003). They usually live in pairs (Estes 1992; Adamczak & Dunbar 2007) or small groups with one male and up to three females (*Dorcatragus*; Giotto & Gerard 2010) and occupy permanent territories that they mark with dung piles and secretions from their preorbital glands (absent in *Dorcatragus*). The other species of Antilopini show different types of social organisation. In most species, the males establish territories, often during the rut only, while females form loose herds and roam freely across territory boundaries (Kingdon 1989a, b; Estes 1992). Female *Litocranius* and *Ammodorcas* are usually solitary, and their home ranges overlap with the territories of one or two males (Estes 1992). *Saiga* males do not establish territories but defend a permanent harem during the rut (Sokolov 1974). Is has been speculated which of these different types of social organisation is ancestral for the whole clade of Antilopini, and again the permanent pair or family territories of the dwarf antelopes are usually regarded as primitive (Kingdon 1989a; Gentry 1992; Kuznetsova & Kholodova 2003; Giotto & Gerard 2010).

Recently, a species-level phylogeny based on a supermatrix analysis of four mitochondrial and five nuclear genes has become available for Antilopini (Bärmann et al. 2013). It resolves *Ourebia* as the sister-taxon to the remaining Antilopina (*Saiga*, *Litocranius*, *Antidorcas*, *Nanger*, *Eudorcas*, *Gazella*, and *Antilope*; *Ammodorcas* should also be included here according to Ropiquet et al. 2009, but molecular data for this species are not available on GenBank) and recovers a clade Raphicerina (a name introduced by Hassanin et al. 2012) comprising the other dwarf antelope genera (*Raphicerus*, *Madoqua*, *Dorcatragus*). The root of the antilopine tree was unstable, placed either between *Procapra*+Raphicerina and *Ourebia*+Antilopina, or with either *Procapra* or Raphicerina as sister-taxon to all other Antilopini.

The reconstruction of ancestral character states for species with known phylogenetic relationships is based on the fact that the evolutionary process creates a pattern of character change along the branches of a phylogeny. This pattern is preserved in the variation of characters among the tips of the phylogeny. Therefore, knowing the variation at the tips and the phylogeny is sufficient for inferring the evolutionary process and for estimating character states at ancestral nodes of the phylogeny (Pagel 1997). In the past decade, Bayesian methods have become popular for reconstructing ancestral character states, as they allow for incorporating parameter uncertainty as well as phylogenetic uncertainty (see review in Ronquist 2004). They have been used for reconstructing ancestral morphology of mosses (Vanderpoorten & Goffinet 2006), the evolution of waggle dance in honey bees (Raffiudin & Crozier 2007), male display signals in lizards (Feldman et al. 2011), ancestral states of virulence in primate malarias (Garamszegi 2011), or ancestral hosts and subsequent host-switches of parasitic nematodes (Jimenez et al. 2012).

In this study, I reconstruct the ancestral character states of body mass, horn length, territoriality, and female social organisation for the most recent common ancestor of Antilopini, using a set of phylogenetic trees derived from combined Bayesian analysis of molecular and morphological data. This allows for testing the long-held hypothesis about bovid evolution, namely that all living antelopes have evolved from a stock of small animals with short, piercing horns that lived solitarily or in pairs in widely spaced territories, and that the living dwarf antelopes are remnants of this early antelope radiation (Kingdon 1989a; Gentry 1992; Kuznetsova & Kholodova 2003).

2. Material and methods

2.1 Reconstruction of ancestral character states

The program BayesTraits V1.1 beta (Pagel 1999; Pagel et al. 2004) was used for reconstructing body mass, horn length, territoriality, and female social organisation for the most recent common ancestors of Antilopini, Antilopina, and Raphicerina, as well as other internal nodes in the phylogeny of Antilopini. These nodes are defined with the command *addMRCA* which does not restrict the selected taxa to be monophyletic. Reconstructions are based on a set of 5000 post-burnin trees from the Bayesian phylogeny estimation (see Methods 2.2, the consensus tree is shown in Fig. 1). Bayes Factor (BF) comparison was used to select the best model of character evolution: For a given pair of models, the BF is determined as the ratio of the marginal likelihoods of the models multiplied by 2. The harmonic mean of the log-Likelihoods (lnL) in the stationary phase of the run (printed in the log file) is used to approximate the marginal likelihood for each model. A BF>2 is interpreted as 'significantly' better fit of the model with the higher lnL (Kass & Raftery 1995). If the difference is smaller, the simpler model with fewer parameters should be preferred. The reconstruction is different for continuously evolving characters, i.e., body mass and horn length, and discrete multistate characters, i.e., territoriality and female social organisation. Both types will be explained briefly in the following two sections, for more information please consult the manuals and publications of the author of the program (Pagel 1999; Pagel et al. 2004).

2.1.1 Discrete multistate characters: territoriality and female social organisation

Data on territoriality and social organisation were taken from numerous authors (Sokolov 1974; Schaller 1977; Jarman 1974; Dubost & Feer 1981; Walther et al. 1983; Walther 1984, 1988; Kingdon 1989a, b; Schaller et al. 1991; Estes 1992; Habibi et al. 1993; Mendelssohn et al. 1995; Yom-Tov et al. 1995; Kingswood & Blank 1996; Kingswood & Kumamoto 1996a, b; Sokolov & Lushchekina 1997; Nowak 1999; Cain et al. 2004; Prater 2005; Ostrowski & Williams 2006; Adamczak & Dunbar 2007; Giotto et al. 2008; Kingdon 2008; Leslie Jr. & Schaller 2008; Giotto & Gerard 2010; Leslie Jr. 2010; Leslie Jr. et al. 2010). For territoriality, every species was placed in one of three categories: living in permanent territories (0), establishing temporary territories during the rut (1), or not establishing territories (2). For female social organisation I distinguished two groups: solitary (0) or living in herds (1). State 0 also includes species where occasionally small groups of up to three females occur (e.g., *Dorcatragus*, *Ourebia*).

Defining the node of interest with the command *addMRCA* allows for calculating the probabilities for each character state at that node. For assessing whether the state with the highest probability is 'significantly' better than the other states, the character has to be fixed (command: *fossil*) to every possible state, running separate analyses for every state. These analyses can be compared using Bayes Factors.

For finding the most suitable priors (uniform, exponential, or gamma distributed priors) for the Bayesian analysis, I ran several short test analyses (1,050,000 generations) with the consensus tree, trying different prior distributions and rate deviation parameters. The ancestral state for the root of the tree was estimated for better comparison between the different models. A Maximum Likelihood estimation of rate change parameters (using the consensus tree) and the ancestral state at the root were used as a reference for the Bayesian analyses.

The final reconstructions of the ancestral states with the best-fit models (exponentially distributed hyperprior) used the original set of 5000 rooted trees and were run for 5,500,000 generations, sampling every 500th generation. Burn-in was set to 500,000. All analyses were run five times to check for convergence of the results.

2.1.2 Continuous characters: body mass and horn length

Data on body mass for males and females of each species were compiled from numerous sources (Walther 1988; Kingdon 1989a, b; Schaller et al. 1991; Estes 1992; Mendelssohn et al. 1995; Silva & Downing 1995; Yom-Tov et al. 1995; Kingswood & Blank 1996; Kingswood & Kumamoto 1996a; Sokolov & Lushchekina 1997; Nowak 1999; Kingdon 2008; Macdonald 2009). The data were log-transformed prior to analysis.

Data on horn length come from numerous skulls measured by the author (data were published in the dissertation of Bärmann (2012) and are available on request). Horn length was divided by skull length to scale it in relation to body size.

The basic model for the reconstruction of continuous traits in BayesTraits is model A, a constant-variance random walk model where the change in the studied character does not show a preferred direction. Therefore the model has only one parameter, the ‘variance of evolution’ parameter. Ancestral state reconstruction using this model will always reconstruct the ancestral condition to lie within the range of the values observed in the tips of the tree. Model B corresponds to a directional random walk. In addition to the ‘variance of evolution’ parameter it has a ‘directional change’ parameter. Using this model makes it possible to reconstruct an ancestral condition outside the range of the tips of the tree and therefore accommodates evolutionary trends, e.g., an increase in body mass.

Three parameters can be used to transform and scale the phylogeny and thereby test the tempo (*Delta*), mode (*Kappa*), and phylogenetic associations (*Lambda*) of trait evolution (Pagel, Continuous manual). If these parameters are estimated as close to 1.0, the phylogeny and its branch lengths are assumed to accurately describe the constant-variance random walk model (A or B). *Kappa* differentially scales the branch lengths and therefore tests for punctuational versus gradual trait evolution: *Kappa*>1 implies that trait evolution accelerates within long branches; if *Kappa*<1 a more punctuational mode of character evolution is assumed, diminishing the influence of branch lengths. *Delta* scales the overall path length in the phylogeny in order to detect a change in the rate of trait evolution over time: *Delta*<1 implies that shorter paths contribute to a higher degree to trait evolution, indicating adaptive radiation; *Delta*>1 implies that longer paths contribute more to trait changes, indicating accelerated evolution. *Lambda* accounts for the difference between expected and observed non-independence among species and adopts values between 0 and 1. In the extreme case of *Lambda*=0, the trait evolves as if species were completely independent of each other; *Lambda*=1 indicates that the phylogeny and the constant variance random walk model correctly represent the data.

In preliminary analyses for model selection (one or two-parameter random walk, incorporation of *Kappa*, *Delta* and *Lambda*) I used the Bayesian consensus tree that was transformed into a rooted, strictly bifurcating tree by introducing zero length branches. Analyses were run for 1,050,000 generations, sampling every 100th generation and discarding the first 50,000 generations as burn-in. BF comparison was used to select the model with the fewest parameters that best fitted the data.

The final reconstructions of the ancestral states with the best-fit models used the original set of 5000 trees and were run for 5,500,000 generations, sampling every 500th generation. Burn-in was set to 500,000.

2.2 Phylogenetic analysis

For the phylogenetic analysis, the molecular data set of Bärmann et al. (2013) was complemented with 16 indels and 88 skull characters from Bärmann (2012) (available on Morphobank, project 352). The taxonomic sampling was similar to Bärmann et al. (2013) but also included *Ammodorcas clarkei* that was sampled for morphological data only. The total number of taxa was 41, comprising all 29 species of Antilopini and 12 species from all other major clades of bovids.

Bayesian analysis was performed with MrBayes v. 3.1.2 (Ronquist & Huelsenbeck 2003) using the default priors (dirichlet distributions for base frequencies as well as substitution rates, exponential branch length priors). The molecular data were partitioned as in Bärmann et al. (2013) (separate partitions for the three codon positions of mitochondrial protein coding regions, 12S, 16S, the three codon positions of nuclear protein coding regions, and one partition for the nuclear non-coding regions). Indels were analysed with the Restriction Sites model (*datatype=restriction*) for binary data to account for different rates of gains and losses of a character (e.g., it will be very unlikely to regain a similar sequence

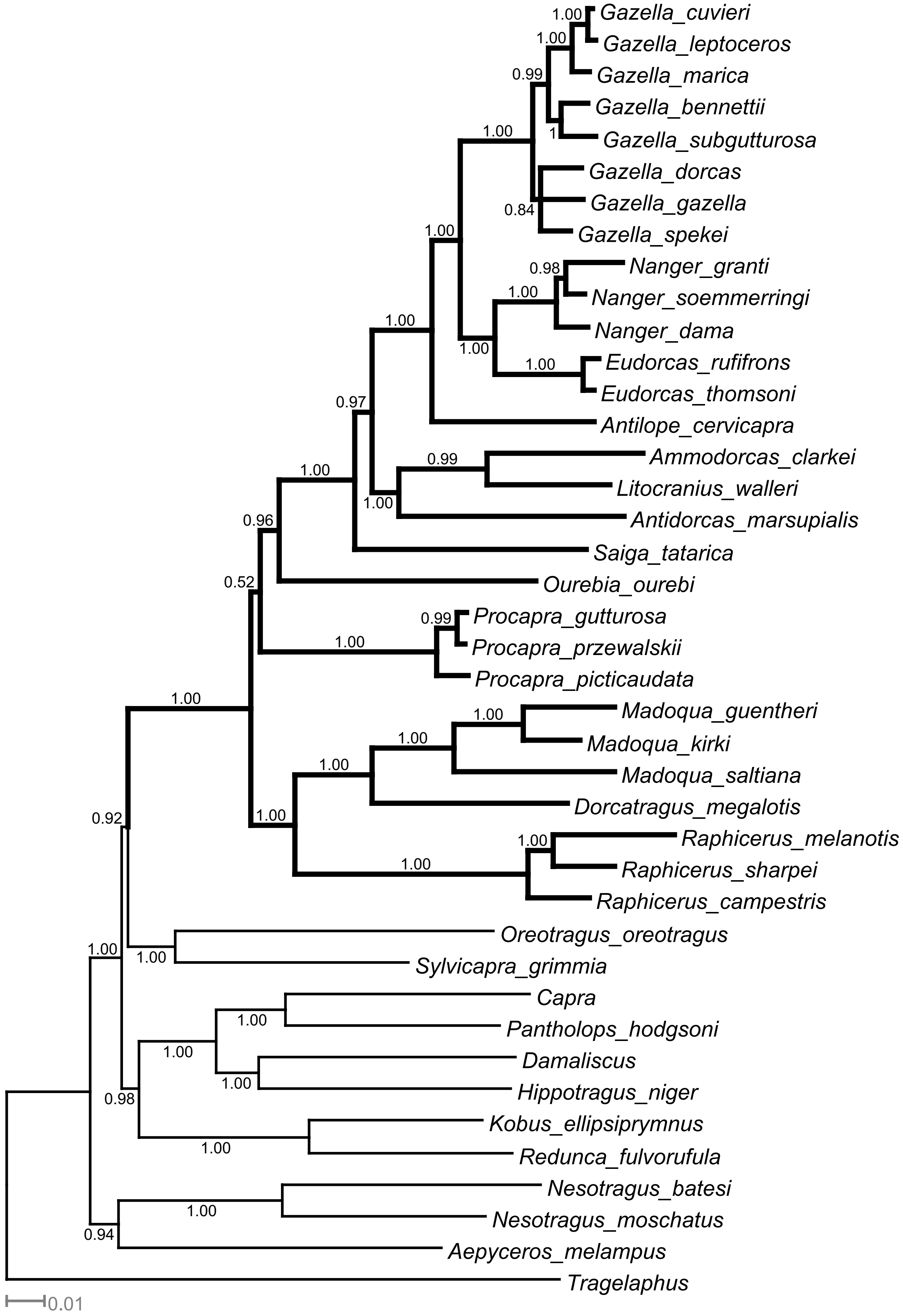

Figure 1: Phylogeny of Antilopinae. Consensus tree of 5000 post-burnin trees from combined Bayesian analysis of molecular and morphological data. Branch labels = posterior probabilities. The clade Antilopini is highlighted with bold branches.

in the place where a deletion has occurred previously). Morphological data were analysed with the Standard Discrete model (*datatype=standard*), which allows multiple character states, and the command *lst coding=variable* was used to account for coding bias (as non-variable characters are generally not recorded in morphological studies; Ronquist & Huelsenbeck 2003). Analyses were run for 50-million generations, sampling every 5,000th generation. Burn-in was determined by visual inspection of likelihood values, tree lengths and parameter estimates in Tracer v. 1.5 (Rambaut & Drummond 2007).

3. Results

3.1 Combined-data phylogeny of Antilopini

The phylogeny from the combined analysis of molecular and morphological data (Fig. 1) in most parts resembles the molecular phylogeny of Bärmann et al. (2013). *Ammodorcas clarkei* was placed as the sister species to *Litocranius* (posterior probability (PP) =1). However, there were some small changes in the topology upon the inclusion of morphological characters: *Antilope* was no longer resolved as the sister group of *Gazella*, but was placed as sister taxon to *Gazella+Eudorcas+Nanger* with high support (PP=1). Reduncini were resolved as the sister group to the clade comprising Caprini, Hippotragini, and Alcelaphini (CHA-clade) with high support (PP=0.95), a relationship that was unresolved in the molecular analysis. Cephalophini+*Oreotragus*, previously placed as sister taxon to Antilopini+Reduncini+CHA-clade by the molecular data (PP=0.67), were grouped with only the Antilopini in the combined analysis (PP=0.92).

3.2 Ancestral states reconstruction

3.2.1 Male territoriality

Reconstruction of male territoriality for the MRCA of Antilopini favoured state 0: ‘temporary territory during the rut’ (Fig. 2, Tab. 1). The mean probability estimation for this character state was 0.72, whereas the other two character states were much less probable (p=0.19 for state 1: ‘permanent territories’; p=0.09 for state 2: ‘no territories’). When constraining the analysis to the three different character states, the Bayes Factor comparison between the lnL estimations for state 0 (lnL=-31.99) and the other

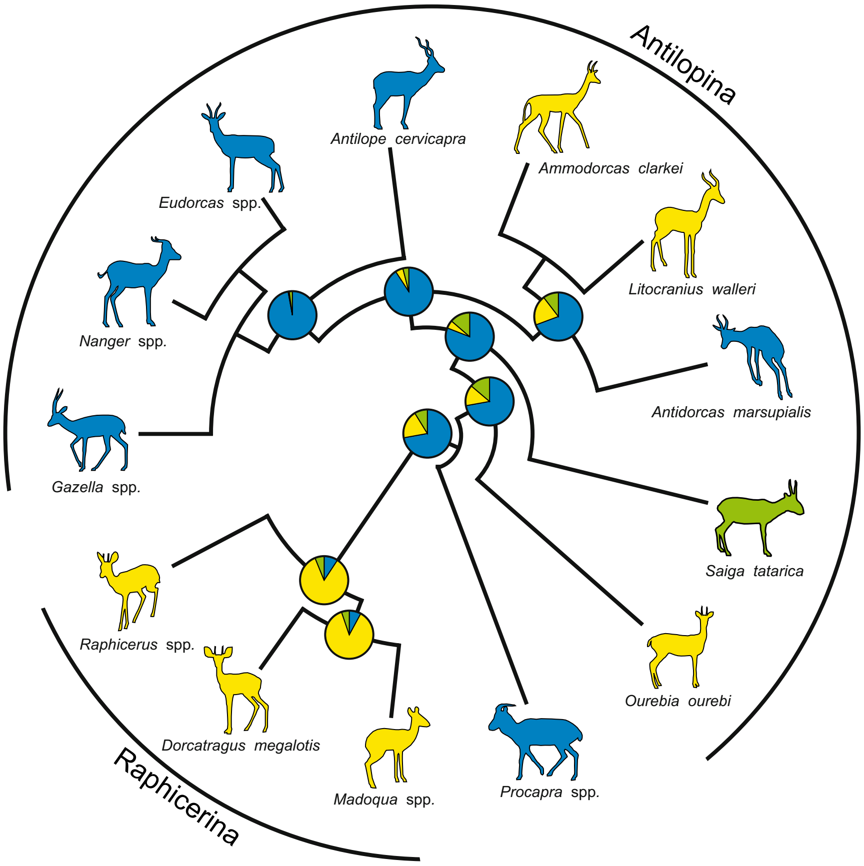

Figure 2: Reconstruction of male territoriality for eight internal nodes in the phylogeny of Antilopini. Blue = temporary territories during the rut, yellow = permanent territories, green = no territories. The relative amount of each colour in the pie charts at the internal nodes represents the probability of the respective character state at that node. Icon colours reflect the character state of each terminal taxon.

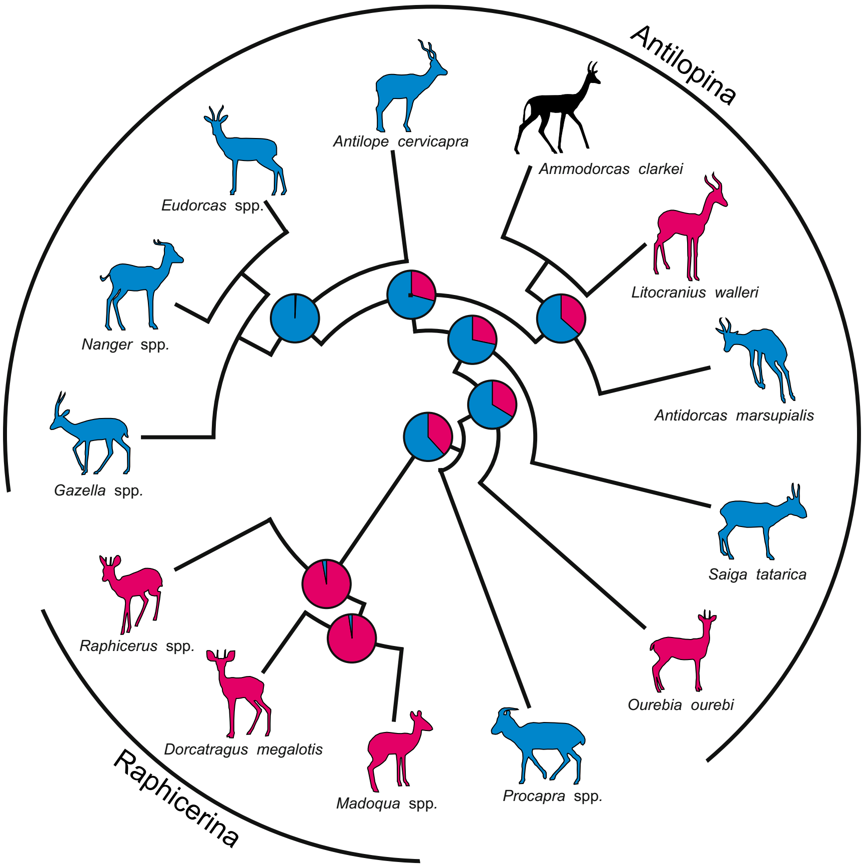

Figure 3: Reconstruction of female social organisation for eight internal nodes in the phylogeny of Antilopini. pink = solitary; blue = living in herds. The relative amount of each colour in the pie charts at the internal nodes represents the probability of the respective character state at that node. Icon colours reflect the character state of each terminal taxon. The character state for *Ammodorcas clarkei* (black icon) is unknown.

two states (state 1: lnL=-33.13; state 2: lnL=-34.08), respectively, favoured state 0 as 'significantly' better (Tab. 1).

For the MRCA of Antilopina, the reconstruction also clearly favoured temporary territories during the rut (state 0, p=0.72), whereas the males of the MRCA of Raphicerina most likely inhabited permanent territories (state 1, p=0.85). In the latter case, however, the difference in the lnL of the favoured state 1 and state 0 (temporary territories) was not 'significant' using Bayes Factor comparison (Tab. 1).

3.2.2 Female social organisation

The reconstruction of female social organisation for the MRCA of Antilopini was not conclusive. State 1 'living in herds' was favoured (p=0.62; Tab. 1) over state 0 'solitary' (p=0.38) in the unconstrained reconstruction (Fig. 3). However, when constraining the analysis to the respective character state, BF comparison of the lnL estimates favoured state 0 'solitary' over state 1 (Tab. 1).

For the MRCA of Antilopina, state 1 'living in herds' (p=0.66) was more likely than state 0 'solitary' (p=0.34). The females of the MRCA of Raphicerina, in contrast, very likely lived solitarily (state 0, p=0.97). BF comparison indicates that for both of these nodes there are 'significant' differences between the lnL of the favoured state and the alternative state (Tab. 1).

3.2.3 Body mass

For the reconstruction of body mass a constant-variance random walk model was used. Estimated values for *Lambda* (0.98 ± 0.01) and *Kappa* (0.99 ± 0.12) were close to 1, indicating that the topology and branch length adequately represent the evolution of body mass. The estimated value for *Delta* was 0.69 ± 0.19. Including any of these three additional variables or a directional change parameter did not 'significantly' increase the likelihood value of the analysis and was therefore regarded as an unnecessary increase in parameters.

The reconstructed body mass for the most recent common ancestor (MRCA) of all Antilopini was 19.7 ± 1.3 Kg for males and 17.8 ± 1.3 Kg for females; the same result was found for the MRCA of Antilopina (Fig. 4). Within Antilopina the body mass increased to 29.1 ± 1.3 Kg for males and 24.1 ± 1.3 Kg for females in the last common ancestor of Gazelles, *Antilope*, *Ammodorcas*, *Litocranius*, *Antidorcas* and

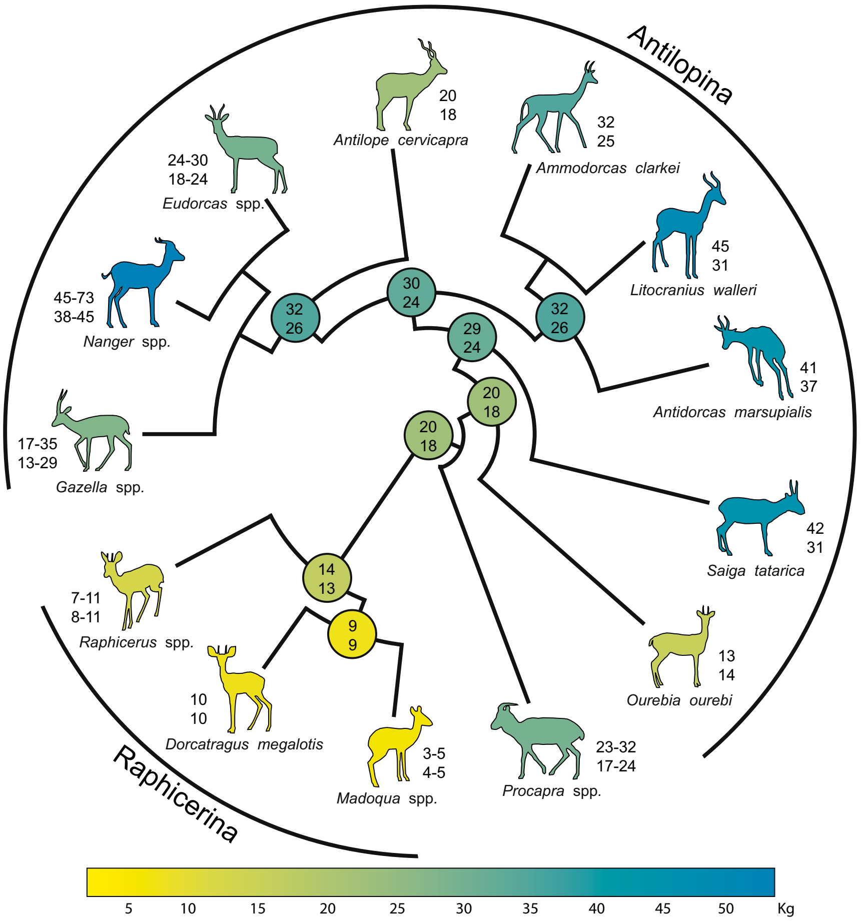

Figure 4: Reconstruction of male (top value) and female (bottom value) body mass (in Kg) for eight internal nodes in the phylogeny of Antilopini. The tips of the cladogram are labelled with the mean value of the body mass of the terminal taxon or with the range of mean body masses in the respective genus. A colour change from yellow to blue is used to visualise the body mass changes from the internal nodes to the tips of the cladogram.

Saiga. The MRCA of Raphicerina was estimated to have had a smaller body mass of only 14.2 ± 1.4 Kg in males and 13.4 ± 1.3 Kg in females, decreasing further in the last common ancestor of *Dorcatragus* and *Madoqua* that was estimated to have weighed 9.4 ± 1.4 Kg in both sexes.

3.2.4 Male horn length

A constant-variance random walk model was used for the reconstruction, including the *Lambda* parameter (0.88 ± 0.06) to account for the difference between expected and observed non-independence among species. Incorporation of other parameters (including a directional change parameter) did not result in a 'significant' increase in the likelihood values and was therefore not considered for the final reconstruction of male horn length.

The male horn length of the MRCA of Antilopini was estimated to 100.7 ± 1.2 % of the basal cranial length (Fig. 5). The MRCA of Antilopina had only slightly longer horns (104.1 ± 1.2 %), but within Antilopina the horn length increased further to about 152.1 ± 1.3 % in the last common ancestor of gazelles and *Antilope*. The MRCA of Raphicerina had horns that were a little shorter than the skull (86.6 ± 1.3 %), and horn length decreased further in the last common ancestor of *Dorcatragus* and *Madoqua* (76.8 ± 1.3 %).

4. Discussion

4.1 Ancestral states reconstruction

Although it seems to be commonly agreed upon that the dwarf antelopes are remnants of the early radiation of Antilopinae, and that their small size, short horns, and social organisation therefore represent the ancestral condition of antelopes (Kingdon 1989a; Gentry 1992; Kuznetsova & Kholodova 2003), this is the first attempt to test these hypotheses using phylogeny-based ancestral states reconstructions. Surprisingly, these reconstructions of four characters do not all agree with the classical view of bovid evolution.

4.1.1 Territoriality and female social organisation

The most likely form of territoriality present in the most recent common ancestor of Antilopini is the

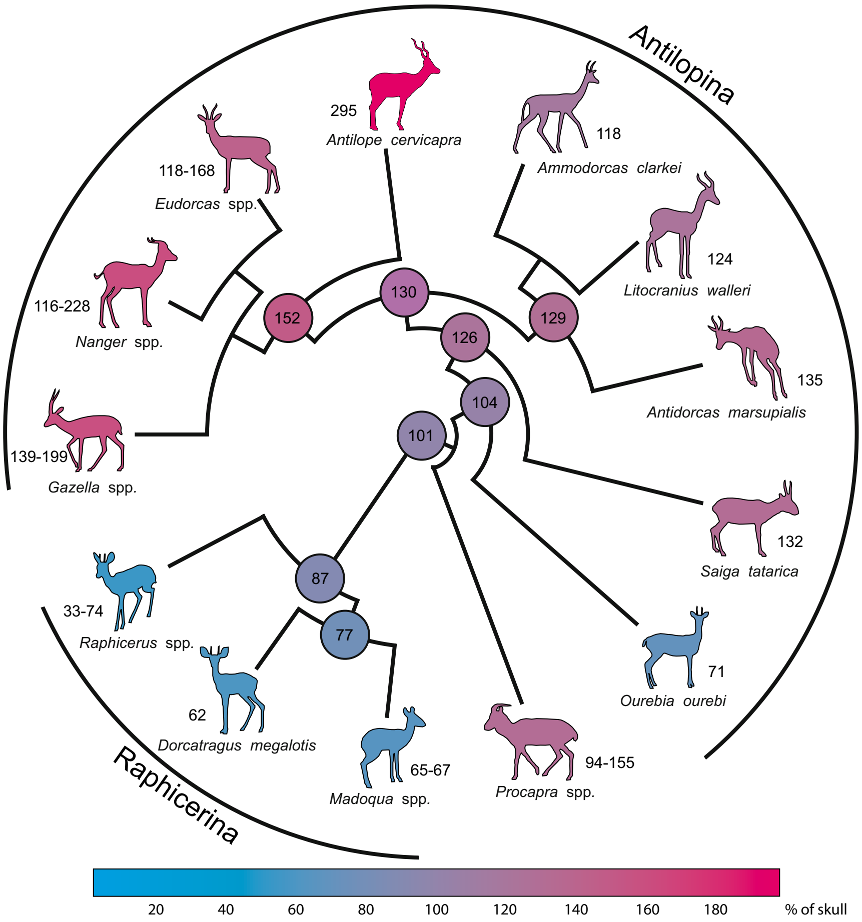

Figure 5: Reconstruction of male horn length (in % of skull length) for eight internal nodes in the phylogeny of Antilopini. The tips of the cladogram are labelled with the mean value of the horn length of the terminal taxon or with the range of mean horn lengths in the respective genus. A colour change from blue to pink is used to visualise the horn length changes from the internal nodes to the tips of the cladogram.

defence of 'temporary territories during the rut'. This character state is present in living *Procapra*, *Antidorcas*, *Antilope* and all three gazelle genera, thus in species inhabiting more open landscapes and feeding on a mixed or grass-dominated diet. The permanent territoriality of the dwarf species as well as of *Litocranius* and *Ammodorcas* might therefore be a secondary adaptation that could be correlated with their specialised diet as browsers: while in species with temporary territories the males establish them mainly for gaining access to trespassing female herds, browsing species mainly defend the limited food resources in their territory. As a trade-off they usually share their territory with only one female. Interestingly, in *Ourebia* two females may share the territory of the same male if the perceived predation risk is high, but only if food is sufficiently abundant (Adamczak & Dunbar 2007).

The reconstruction of female group size for the most recent common ancestor of Antilopini is ambiguous. While the unconstrained analysis slightly favours 'living in herds' (p=0.66), the constrained analysis with female social organisation set to 'solitary' has the higher likelihood value. The reconstructions for Antilopina and Raphicerina are more conclusive and favour, 'living in herds' for Antilopina and 'solitary' for Raphicerina. However, the analysis took a very simplified perspective, as all female groups larger than three were treated as the same character state. In reality, there are huge differences in the size of the female groups, as well as large intraspecific variation correlated with food availability, seasonal climate changes and predator abundance (see Estes 1992, Brashares et al. 2000, and references therein). Accounting for these differences might influence the outcome of the analysis. However, especially the intraspecific behavioural plasticity is problematic, firstly because the reality rarely fits the strict categories that we invent to classify it, and secondly because for most species the known social behaviour might only be a fraction of the possible behavioural spectrum.

4.1.2 Body mass and horn length

The body mass for the most recent common ancestor (MRCA) of Antilopini, reconstructed to have been about 20 Kg in males and slightly less in females (Fig. 4), lies in the range of present day gazelles in the genus *Gazella*. The same ancestral condition was reconstructed for the MRCA of Antilopina. This implies that the body mass reconstruction

Table 1: Reconstruction of male territoriality and female social organisation for three ancestral nodes: Antilopini, Antilopina, and Raphicerina. p = posterior probability of the character state (average from the post-burnin phases of five independent runs); lnL = natural logarithm of the harmonic mean of the likelihoods of the post burn-in phase of the Bayesian analysis with fixed character states (average from 5 independent runs); BFC = 2*log(BF), Bayes Factor comparison between the state with the highest lnL (**bold**) and the other states, values larger than 2 are regarded as positive evidence for the model with the higher lnL.

Discrete characters	Antilopini			Antilopina			Raphicerina		
	p	lnL	BFC	p	lnL	BFC	p	lnL	BFC
Male territoriality									
0: temporary territory during the rut	**0.72**	**-31.99**		**0.72**	**-32.21**		0.09	-33.07	0.66
1: permanent territory	0.19	-33.13	2.28	0.14	-33.54	2.66	**0.85**	**-32.74**	
2: no territory	0.09	-34.08	4.17	0.14	-34.46	4.51	0.06	-35.29	5.09
Female social organisation									
0: solitary	0.38	**-14.28**		0.34	-16.7	4.34	**0.97**	**-14.38**	
1: living in herds	**0.62**	-15.50	2.45	**0.66**	**-14.5**		0.03	-16.89	5.03

is not affected by the unstable position of the root of the Antilopini tree that could either be between Raphicerina and Antilopina+*Procapra* or between Raphicerina+*Procapra* and Antilopina. In any case, the body mass most likely decreased twice within Antilopini: within Raphicerina and in the ancestral lineage of *Ourebia*. The small body size of the dwarf antelopes in Antilopini (*Raphicerus*, *Dorcatragus*, *Madoqua* and *Ourebia*) should therefore be regarded as a derived condition. This miniaturisation took place relatively early in the evolutionary history of the clade. According to the dated phylogeny provided by Bibi (2013), the split between Raphicerina and Antilopina occurred between 12-10 Ma, right at the start of the radiation of crown-Antilopini. The body mass changes should therefore be regarded as part of the adaptive ration, and are not a recent phenomenon.

Repeated miniaturisation within a clade is a phenomenon that has recently also been shown for other mammals, i.e. Madagascan dwarf lemurs (Masters et al. 2014). For lemurs as well, the small body size was previously thought to represent the primitive condition. The authors present several lines of evidence for paedomorphosis, including morphometric similarities of adult dwarf lemurs and juveniles of their larger relatives. The body size reduction in correlation with truncated ontogeny (progenesis) could have been selected for by high environmental unpredictability and might also be correlated with dietary changes in the small species. For antelopes, additional analyses like morphometric comparison of developmental sequences of dwarf and non-dwarf species could help to further understand the process of miniaturisation and might shed light on the underlying selective pressures.

The reconstruction of the ancestral horn lengths within Antilopini shows a similar pattern (Fig. 5) as the body size. The condition reconstructed for the MRCA of Antilopini and the MRCA of Antilopina, a horn that has about the length of the skull, is most similar to the Mongolian gazelle *Procapra gutturosa* among the living species. Again, the unstable root of the tree does not affect the ancestral character state reconstruction. While the horn length increased dramatically within Antilopina, reaching 300 % of the skull length in *Antilope*, the horns of the ancestor of the living Raphicerina species and of the *Ourebia* ancestors got shorter.

Perhaps the miniaturisation in combination with a decrease in horn length happened convergently not only twice within Antilopini, but also in the ancestral lineages of *Oreotragus*, *Nesotragus* and *Neotragus*, i.e., in all species formerly united in the taxon “Neotragini” on the basis of these traits. This should be tested in a larger-scale ancestral-state analysis including all living bovid species.

Of course, a secondary miniaturisation in the living dwarf antelope species does not rule out that the earliest bovids also possessed a similar body mass and horn morphology. A possible scenario is that horn evolution started with very short horns in species like the early Middle Miocene *Eotragus noyei* Solounias et al., 1995 (from the Kamlial Formation, Pakistan, 18-18.3 Ma), *Eotragus artenensis* Ginsburg and Heintz, 1968 (from Córcoles, Spain, around 18 Ma; Alférez et al., 1980) and *Namacerus gariepensis* Morales et al., 2003 (from Arrisdrift, Namibia, 17-17.5 Ma), proceeded to longer and more elaborate horns in the ancestor of Antilopinae, and returned to simple, short horns in the lineages leading to Raphicerina, *Ourebia*, *Nesotragus*, *Neotragus*, *Oreotragus*, and Cephalophini.

Based on this scenario it is possible to make predictions about the genes and developmental pathways that are responsible for generating the various horn shapes in bovids. I hypothesize that the genetic basis for lyre-shaped horns, the most common type of horns in Antilopini, can also be found in *Ourebia* and Raphicerina. However, the expression of these genes has been suppressed in these species, possibly because horn development in dwarf antelopes exhibits a heterochronic change, i.e., paedomorphosis, terminating at a comparatively premature state. While the genetic basis of horn shape is not yet fully understood (Davis et al. 2011), this prediction of similarity between small-horned bovids such as *Ourebia* and raphicerines provides a phenotypic basis for future studies.

4.2 Comparison with the fossil record

Of course the fossil record of antelopes provides us with the possibility to evaluate the reconstructed ancestral states by comparing them with data from fossil species. One of the most complete antelope fossils from the Late Miocene is †*Gazella lydekkeri* Pilgrim, 1937 from the Dhok Pathan level of the Middle Siwaliks in Pakistan (Fig. 6). Its phylogenetic position was estimated in the combined-data analysis in Bärmann (2012), which placed it within Antilopini, but not as a close relative of recent gazelles. Instead it was part of a basal polytomy, together with Raphicerina, *Procapra*, *Ourebia*, and the remaining Antilopina. The most frequent topologies in the Bayesian tree sample showed a sister-taxon relationship of †*G. lydekkeri* and *Ourebia*. †*G. lydekkeri* may therefore be a good model for the ancestral habitus of Antilopini.

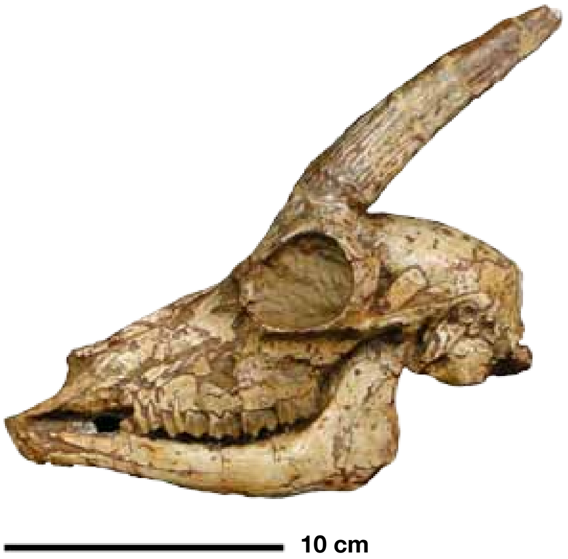

Figure 6: †*Gazella lydekkeri*, male holotype skull (AMNH 19663) housed in the American Museum of Natural History in New York.

The body mass of fossil species can be inferred with linear regressions based on skull, tooth or limb bone dimensions (Janis 1990; Scott 1990; Köhler 1993). Here, I used the regressions based on comparative data from 74 extant bovid species in Janis (1990) for estimating the body mass of †*G. lydekkeri* (AMNH 19663, male holotype). Skull length had the highest correlation coefficient with body mass in bovids ($r^2=0.958$), and can be used thanks to the exceptional preservation of the specimen. The estimated body mass (29.2 ± 10 Kg) is higher than the reconstructed ancestral state of the MRCA of Antilopini (but the estimated value from the ancestral state reconstruction lies within the standard deviation of the estimated weight of †*G. lydekkeri*); therefore it does not contradict the hypothesis that the ancestor of Antilopini was larger than previously thought.

The (longer) right horn core of the specimen measures 116 mm. In living Antilopini, the average ratio of horn core to total horn length is 0.63 ± 0.09 (0.47–0.72; values from 14 individuals representing eight species for which both measurements were available). Assuming that †*G. lydekkeri* horns had a similar horn core/horn sheath ratio, the specimen had a horn length of about 184 mm (161–214 mm). This translates to 99 % (87–115 %) of the skull length, a value that very well matches the estimated ancestral condition for Antilopini (100.7 ± 1.2 %).

5. Conclusions

The reconstruction of ancestral states based on a phylogeny is useful for testing long-held hypotheses about bovid evolution. Even if the phylogeny is not perfectly resolved, Bayesian methods allow for incorporating this uncertainty. For Antilopini, the reconstructed ancestral states for body mass yielded an unexpected result, as the most recent common ancestor is estimated to have weighed approximately 20 Kg. This is much larger than the assumedly “primitive“ dwarf antelopes with a body mass of 3-14 Kg. The horn length of these small species, i.e., *Ourebia*, *Madoqua*, *Dorcatragus* and *Raphicerus*, is also estimated to be secondarily reduced, possibly caused by paedomorphic early termination of horn growth. These results of the ancestral horn length estimation presented here could serve as a starting-point for studies on the evolution and development of bovid horns. An independent test for ancestral states reconstructions, at least for fossilizing traits, can be provided by the fossil record, given that the phylogenetic position of the fossils are determined beforehand, ideally using a combined-data phylogenetic analysis.

Reconstructions for the female social organisation and male territoriality also challenge the long-held idea that the pair- or family territories of the dwarf antelopes represent the ancestral condition for Antilopini: according to the analysis permanent territories most likely represent a derived trait and evolved twice within Antilopini. However, intraspecific behavioural plasticity can hardly be incorporated into current analyses, so the reconstructions of territoriality and female social organisation should be treated with caution.

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