Is the development of infectious keratoconjunctivitis in Alpine ibex and Alpine chamois influenced by topographic features?

Fabien Mavrot, Fridolin Zimmermann, Edy M. Vilei & Marie-Pierre Ryser-Degiorgis

European Journal of Wildlife Research

ISSN 1612-4642
Volume 58
Number 5

Eur J Wildl Res (2012) 58:869-874
DOI 10.1007/s10344-012-0651-1

Springer

Eur J Wildl Res (2012) 58:869–874
DOI 10.1007/s10344-012-0651-1

SHORT COMMUNICATION

Is the development of infectious keratoconjunctivitis in Alpine ibex and Alpine chamois influenced by topographic features?

Fabien Mavrot · Fridolin Zimmermann · Edy M. Vilei · Marie-Pierre Ryser-Degiorgis

Received: 5 July 2011 / Revised: 20 June 2012 / Accepted: 28 June 2012 / Published online: 13 July 2012

Abstract Infectious keratoconjunctivitis (IKC) caused by *Mycoplasma conjunctivae* is a widespread ocular affection of free-ranging Caprinae in the Alpine arc. Along with host and pathogen characteristics, it has been hypothesized that environmental factors such as UV light are involved in the onset and course of the disease. This study aimed at evaluating the role of topographic features as predisposing or aggravating factors for IKC in Alpine chamois (*Rupicapra rupicapra rupicapra*) and Alpine ibex (*Capra ibex ibex*). Geospatial analysis was performed to assess the effect of aspect (northness) and elevation on the severity of the disease as well as on the mycoplasmal load in the eyes of affected animals, using data from 723 ibex and chamois (583 healthy animals, 105 IKC-affected animals, and 35 asymptomatic carriers of *M. conjunctivae*), all sampled in the Swiss Alps between 2008 and 2010. An influence of northness was not found, except that ibex with moderate and severe signs of IKC seem to prefer more north-oriented slopes than individuals without corneal lesions, possibly hinting at a sunlight sensitivity consequent to the disease. In contrast, results suggest that elevation influences the disease course in both ibex and chamois, which could be due to altitude-associated environmental conditions such as UV radiation, cold, and dryness. The results of this study support the hypothesis that environmental factors may play a role in the pathogenesis of IKC.

Keywords Aspect · Elevation · Environmental factors · Infectious keratoconjunctivitis · *Mycoplasma conjunctivae* · Wild Caprinae

Communicated by C. Gortázar

F. Mavrot · M.-P. Ryser-Degiorgis (✉)
Centre for Fish and Wildlife Health, Vetsuisse Faculty,
University of Bern,
Länggass-Strasse 122, PO Box 8466, CH-3001 Bern, Switzerland
e-mail: marie-pierre.ryser@vetsuisse.unibe.ch

F. Zimmermann
KORA,
Thunstrasse 31,
CH-3074 Muri, Switzerland

E. M. Vilei
Institute of Veterinary Bacteriology, Vetsuisse Faculty,
University of Bern,
Länggass-Strasse 122, PO Box 8466, CH-3001 Bern, Switzerland

Introduction

Infectious keratoconjunctivitis (IKC) caused by *Mycoplasma conjunctivae* is a complex disease of domestic and wild Caprinae, with great variations in the clinicopathological and epidemiological picture. In wildlife, IKC is sometimes associated with high mortality (Giacometti et al. 2002; Mavrot et al. 2012).

It has been suggested that the pathogenesis of IKC is influenced by host predispositions, virulence of *M. conjunctivae* strains, secondary infections, and environmental factors (Hars and Gauthier 1984; Nicolet 1985). Sex and age imbalance in affected populations were observed in severe outbreaks (Degiorgis et al. 2000; Tschopp et al. 2005), indicating that age and social behavior including sexual segregation may be important risk factors. While possible differences in virulence between different strains do not seem to play a major role, mycoplasmal load is obviously associated to the presence and severity of signs (Ryser-Degiorgis et al. 2009; Mavrot et al. 2012). However, the driver of mycoplasmal multiplication in the host is unknown. Environmental factors may play a role, regarding both the expression of the disease in individual cases and the onset of an outbreak in a population (Mavrot et al. 2012).

Altitude, air quality, and UV light have been discussed as possible predisposing factors for IKC in wild ungulates along with overcrowding (Nicolet 1985; Costa 1986). In this study, we evaluated whether elevation and aspect (i.e., slope orientation) may be predisposing or aggravating factors for IKC, using a sample of Alpine chamois (*Rupicapra rupicapra rupicapra*) and Alpine ibex (*Capra ibex ibex*) systematically examined for IKC signs and tested for infection with *M. conjunctivae*. The fact that geospatial distribution of wild Caprinae varies throughout the year (Parrini et al. 2003; Nesti et al. 2010) was considered in the analysis.

Material and methods

Samples

Data on 723 chamois and ibex in 17 regions of the Swiss Alps between 2008 and 2010 were available for this study. Regions are limited by geographical barriers and considered as epidemiological units (Fig. 1, Mavrot et al. 2012). Convenience sampling was carried out by hunters, game keepers, and biologists on animals that were hunted, shot because of disease, found dead, or captured for marking procedures. Each eye was sampled separately with a sterile swab. These were sent to the Centre for Fish and Wildlife Health within 48 h. Biological data of sampled animals, geographical coordinates, date and time of sampling, and a standardized description of ocular signs were systematically recorded (Mavrot et al. 2012). All samples were tested for the presence of *M. conjunctivae* by quantitative real-time PCR according to established procedures (Vilei et al. 2007), the obtained mycoplasmal load corresponding to the estimated amount of mycoplasma in the sample.

Eyes included in the analysis were classified according to previous clinical and histological observations (Mavrot et al. 2012) as (a) healthy (eyes from asymptomatic animals, negative for *M. conjunctivae* in both eyes; ibex = 844, chamois = 322), (b) asymptomatic carriers (eyes from asymptomatic animals, positive for *M. conjunctivae*; ibex = 32, chamois = 13), (c) mild IKC (ocular discharge; ibex = 16, chamois = 17), (d) moderate IKC (ocular discharge, corneal opacity; ibex = 31, chamois = 45), and (e) severe IKC (ocular discharge, corneal opacity, corneal neovascularization, or perforation; ibex = 32, chamois = 45). Eyes were also classified as sampled during summering season (ibex: June–October, chamois: May–October) or wintering season (remaining months). These seasons were defined according to knowledge on annual shifts in habitat of wild Caprinae in Switzerland (Meile et al. 2003; Schnidrig-Petrig and Salm 2009.)

Geoprocessing and data analysis

For each animal, elevation and northness were computed using the exact coordinates recorded at sampling location with a 100-m grid cell resolution. Since one-point location may not be representative of the environment in which the animals live and could be subject to bias due to sampling method, average altitude and northness were additionally assessed for the circular sampling area (CSA, i.e., surface around the sampling location) of each individual, as proposed by Ramp et al. (2005). Size of CSA was derived using home-range sizes from studies conducted in similar habitats (Hardenberg et al. 2000; Meile et al. 2003; Boschi and

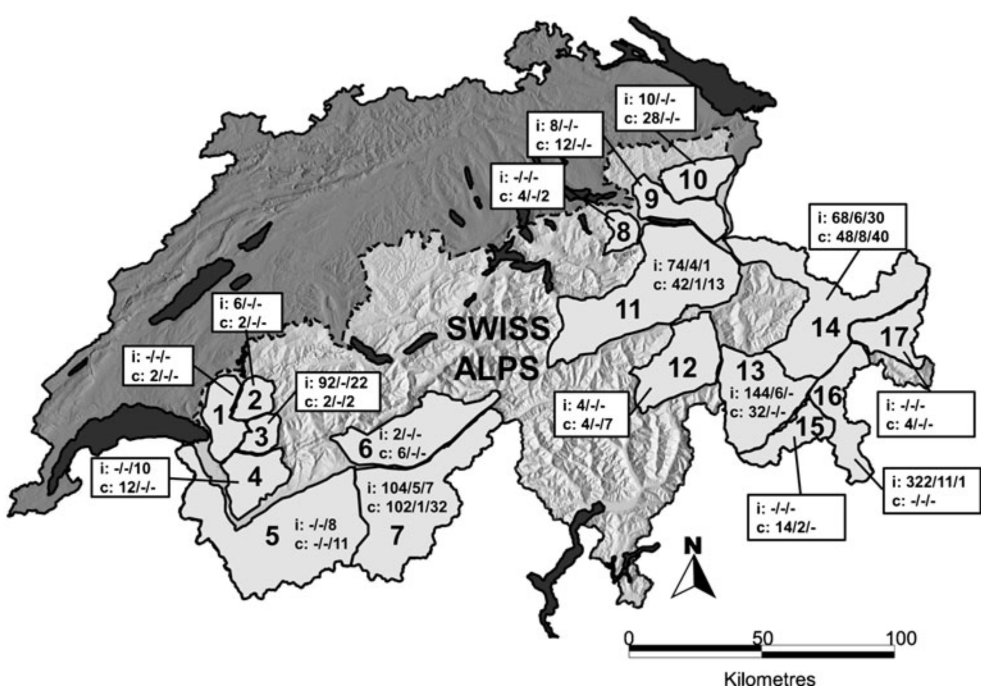

Fig. 1 Map of Switzerland showing main lakes (*black*), the Alps (*light grey*), and the study regions (*1* Dent-de-Lys; *2* Vanil Noir; *3* Cape-au-Moine; *4* Diablerets; *5* Valais central left riverside; *6* Oberwallis right riverside; *7* Oberwallis left riverside; *8* Längenegg; *9* Churfristen; *10* Alpstein; *11* Oberalp-Calanda; *12* Rheinwald; *13* Julier; *14* Flüela; *15* Bregaglia; *16* Albris; *17* Macun). For each region, the number of sampled eyes (healthy/healthy carriers/symptomatic) is given for ibex (*i*) and chamois (*c*) (made with gvSIG and Microsoft Office Power Point)

Nievergelt 2003; Grignolio et al. 2004; Baumann 2005; Nesti et al. 2010). Estimates (in hectares) depend on the species (ibex = i, chamois = c), sex (female = f, male = m), and season (wintering = w, summering = s): i-f-w = 30, i-f-s = 250, i-m-w = 125, i-m-s = 450, c-f-w = 70, c-f-s = 500, c-m-w = 10, and c-m-s = 50. Northness was defined as the degree of orientation towards north or south at sampling location. It was determined based on the formula of Hengl et al. (2004) and ranged from −90 to +90 with +90 being a full north orientation, −90 a full south orientation, and 0 either a full east or a full west orientation. Geoprocessing analyses were effectuated with ArcView (Environmental Systems Research Institute, Redlands, CA, USA) and gvSIG (Generalitat Valenciana, Spain).

Statistical significance was set at 0.05 for all tests. The Kruskal–Wallis test for multiple groups was used to assess differences in northness and elevation between sampled eyes of different IKC status (healthy, healthy carrier, mild, moderate, and severe) and to evaluate possible confounding related to sampling time of the day. *Post hoc* analysis of pairs of groups was performed using a two-tailed Mann–Whitney–Wilcoxon test. To further check for confounding, pairs with significant differences were tested at seasonal (wintering/summering) and regional levels (only for regions with more than five samples in at least three different IKC status groups, i.e., Flüela and Oberwallis left riverside for chamois and Flüela and Cape-au-Moine for ibex). Finally, a possible association between mycoplasmal load and northness or elevation was evaluated by Spearman rank correlation coefficient.

Because summarizing ophthalmological findings at the individual level can result in loss of information (Murdoch et al. 1998), statistical analyses were based on eyes rather than individuals. *P* value was adjusted for an estimated inter-eye correlation of 0.70 for IKC stages (Kendall's tau, $P<0.001$) and of 0.75 for mycoplasmal load (Spearman's rho, $P<0.001$) as previously described (Rosner et al. 1979; Rosner 1982; Griffin and Gonzalez 1995).

For healthy animals, which, per definition, yield exactly the same information in both eyes (no IKC signs, negative to *M. conjunctivae*), the inter-eye correlation was 1.

Results and discussion

Sampling time ranged from 6 a.m. to 10 p.m., and there was no statistical difference between healthy, healthy carriers, and IKC-affected animals (ibex, $P=0.655$; chamois, $P=0.205$).

Northness

In both species, there was no correlation between northness and mycoplasmal load and no significant differences in CSA northness among IKC stages. In contrast, there was a significant difference in northness at sampling location among IKC stages in ibex ($P=0.017$), but not in chamois: northness at sampling location was significantly higher for ibex eyes with severe signs than for eyes without corneal lesions (healthy, $P=0.002$; asymptomatic carriers, $P=0.005$; mild signs, $P=0.037$, Fig. 2, panel a). Similar results were

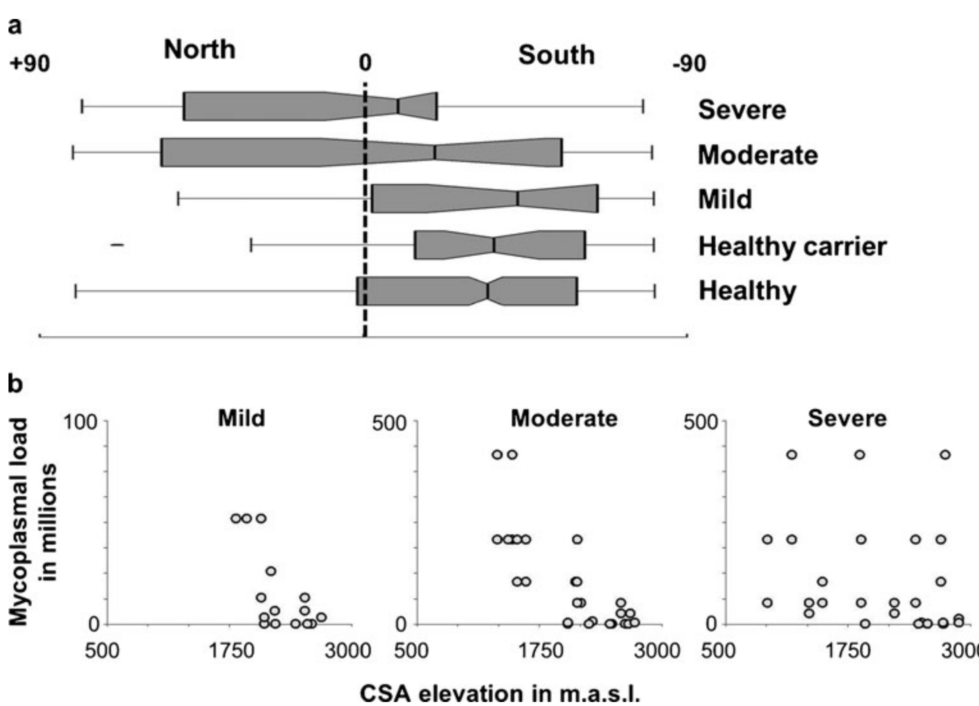

Fig. 2 Influence of aspect and elevation on the course of infectious keratoconjunctivitis (IKC) in ibex. **a** Northness of eyes sampled in ibex depending on the IKC status. **b** Mycoplasmal load in symptomatic ibex eyes depending on the circular sampling area (CSA) elevation. Load is given in millions of mycoplasma and altitude in meters above sea level (mild, $n=16$; moderate, $n=31$; severe, $n=32$). Since mycoplasmal load is lower in early stages of IKC (Mavrot et al. 2012), the scale of the *y*-axis was adapted for the IKC group with mild signs (made with NCSS and Microsoft Office PowerPoint)

found when considering seasons: northness was higher for eyes with severe signs than healthy carriers (P=0.008) and eyes with mild signs (P=0.034) during summering and than healthy eyes during wintering (P=0.007). Since differences were found only when considering sampling location and not the CSA, they may be due to an influence of IKC on the behavior (choice of slope exposure) of affected ibex rather than to a causal relationship between slope exposure and IKC: individuals with severe signs may prefer more north-oriented, less sunny slopes. Photophobia has been described in IKC-affected sheep and goats (Baas et al. 1977; Dagnall 1994), and free-ranging ungulates may behave similarly. Especially in winter, avoidance of south-oriented slopes could further weaken affected individuals since ibex are known to normally seek favorable environmental conditions on sunny slopes (Meile et al. 2003; Parrini et al. 2003). Significant difference in northness between eyes with and without corneal lesions was not found at regional level, but this may be due to the small sample size. While results indicate that confounding due to seasonal variation in habitat use is unlikely, relationships may be confounded by topographic differences among regions.

Elevation

In contrast to the results on northness, multiple-group comparison indicated significant differences in elevation among IKC stages for chamois, but not for ibex, both when considering CSA (P=0.014) and sampling location (P=0.014). In chamois, CSA elevation was significantly higher for eyes with severe signs than for eyes with moderate signs (P< 0.001) and healthy eyes (P=0.008, Table 1). These differences were highly significant during wintering (P=0.003 and P<0.001, respectively) but absent during summering. A higher CSA elevation for severely affected eyes during wintering was also detected at regional level (Table 1).

Although no clear association between elevation and severity of IKC signs was found in ibex, there was a weak negative correlation between CSA elevation and mycoplasmal load in IKC eyes of all stages in this species (Spearman's coefficient correlation r_s=−0.498, P<0.001). When IKC stages in ibex were considered separately, this negative correlation was relatively strong for eyes with mild (r_s=−0.6441, P=0.021, Fig. 2, panel b) and moderate signs (r_s=−0.7545, P<0.001) but was absent for eyes with severe signs (r_s=−0.3443, P=0.139). When considering only the ibex eyes sampled during an outbreak of IKC in the regions of Les Diablerets/Cape-au-Moine in winter 2008–09, similar results were obtained for eyes with moderate signs (r_s=−0.7545, P=0.036). Thus, a lower mycoplasmal load may be needed at higher than at lower elevation to provoke the same damage to the eye. The lack of a correlation in later stages may be due to other factors such as overgrowth of secondary bacterial agents (Giacometti et al. 2002) influencing the mycoplasmal load and hiding a possible effect of elevation. There was no significant correlation between mycoplasmal load and elevation in eyes of healthy carriers (r_s=−0.319, P=0.118). In contrast to our results on northness, effect of elevation (severity of IKC or mycoplasmal load) was also detected at regional level for both species, thus suggesting a real impact of this factor. A selection bias of asymptomatic animals due to preferred hunting location (e.g., low altitude, proximity to roads) may account for these differences, but phenotypical selection and hunting plan constraints are expected to enhance distribution of hunters in ibex and chamois habitat and to reduce systematic sampling errors.

Overall, our results suggest that elevation may be a predisposing or aggravating factor for IKC in both species. However, it is probably rather the associated environmental conditions that influence the course of the disease, than elevation itself: UV light is a direct cause of damage to all eye tissues and is considered a predisposing factor of infectious keratitis in humans (Ellerton et al. 2009). UV exposure is known to increase with altitude (10 % per 1,000 m, Marín et al. 2005) and is expected to be particularly high in winter due to snow cover (Dolin and Johnson 1994). Relative humidity drops with elevation (Duane et al. 2008), which is considered along with sun, wind, and cold, to contribute

Table 1 Differences in circular sampling area elevation depending on the status of infectious keratoconjunctivitis (IKC) in chamois eyes (a healthy, b healthy carrier, c mild IKC, d moderate IKC, e severe IKC)

Home range	Region	Medians of groups (m.a.s.l.)		Difference between medians (m)	P value
Whole year	All	1,891 (a)	2,174 (e)	−283	0.008
		1,721 (d)	2,174 (e)	−453	<0.001
Wintering	All	1,624 (a)	2,211 (e)	−587	<0.001
		1,810 (d)	2,211 (e)	−401	0.003
Wintering	Oberwallis left riverside	1,703 (a)	2,332 (e)	−629	0.009
Wintering	Flüela	1,987 (d)	2,249 (e)	−262	0.038

Only significant differences are listed

to increased tear evaporation and dry eye syndrome in human populations living at high altitude (Gupta et al. 2008), the latter being a predisposing condition for microbial keratitis (Dart 1988).

Conclusion

We attempted to evaluate the importance of elevation and aspect in the pathogenesis of IKC in Alpine chamois and ibex. Results differed between the two species: only ibex seem to search for less exposed slopes when severely affected by IKC. Furthermore, an association between IKC signs and elevation was clearly present in chamois, but not in ibex, while in the latter, an association was observed between mycoplasmal load and elevation. These differences may be explained by species-related differences in physiology and metabolism such as sensibility to *M. conjunctivae* (Gauthier 1991; Giacometti et al. 1997; Ryser-Degiorgis et al. 2009; Mavrot et al. 2012). To our knowledge, this study is the first to investigate the potential role of environmental factors on the pathogenesis of IKC in wild Caprinae. While northness does not seem to influence the IKC course, elevation may be an important factor.

Acknowledgments The authors thank the cantonal hunting authorities of the study regions for their participation in this study and the state game keepers, hunters, and collaborators of the Swiss National Park for collecting the samples. Many thanks to J. Frey (Institute of Veterinary Bacteriology, University of Bern, Switzerland) for the logistical support, E. Pesenti (KORA, Switzerland) for the statistical advice, and C. Willisch (FaunAlpin GmbH, Switzerland) and H. Segner (Centre for Fish and Wildlife Health, University of Bern, Switzerland) for the critical reading of the manuscript. This study was funded by the Swiss Federal Office of the Environment.

References

Baas EJ, Trotter SL, Franklin RM, Barile MF (1977) Epidemic caprine keratoconjunctivitis: recovery of *Mycoplasma conjunctivae* and its possible role in pathogenesis. Infect Immunol 18:806–815

Baumann PW (2005) Die Alpen-Gämse. Ein Leben auf Gratwanderung. Ott-Verlag, Bern

Boschi C, Nievergelt B (2003) The spatial patterns of Alpine chamois (*Rupicapra rupicapra rupicapra*) and their influence on population dynamics in the Swiss National Park. Mamm Biol 68:16–30. doi:10.1078/1616-5047-1610058

Costa R (1986) Contribution à l'étude étiologique de la kératoconjonctivite du chamois (*Rupicapra rupicapra*) et du bouquetin (*Capra ibex*). Dissertation, University Claude Bernard of Lyon

Dagnall GJR (1994) An investigation of colonization of the conjunctival sac of sheep by bacteria and mycoplasmas. Epidemiol Infect 112:561–567

Dart JK (1988) Predisposing factors in microbial keratitis: the significance of contact lens wear. Br J Ophthalmol 72:926–930

Degiorgis M-P, Frey J, Nicolet J, Abdo E-M, Fatzer R, Schlatter Y, Reist S, Janovsky M, Giacometti M (2000) An outbreak of infectious keratoconjunctivitis in Alpine chamois (*Rupicapra r. rupicapra*) in Simmental-Gruyères. Schweiz Arch Tierheilkd 142:520–527

Dolin PJ, Johnson GJ (1994) Solar ultraviolet radiation and ocular disease: a review of the epidemiological and experimental evidence. Ophthalmic Epidemiol 1:155–164. doi:10.3109/09286589409047224

Duane WJ, Pepin NC, Losleben ML, Hardy DR (2008) General characteristics of temperature and humidity variability on Kilimanjaro, Tanzania. Arct Antarct Alp Res 40:323–334. doi:10.1657/1523-0430(06-127)[DUANE]2.0.CO;2

Ellerton JA, Zuljan I, Agazzi G, Boyd JJ (2009) Eye problems in mountain and remote areas: prevention and onsite treatment—official recommendations of the International Commission for Mountain Emergency Medicine ICAR MEDCOM. Wilderness Environ Med 20:169–175. doi:10.1580/08-WEME-REV-205R1.1

Gauthier D (1991) La kérato-conjonctivite infectieuse du chamois. Étude épidémiologique dans le Département de la Savoie 1983–1990. Dissertation, University Claude Bernard of Lyon

Giacometti M, Degiorgis M-P, Mayer D, Krawinkler M, Nicolet J (1997) Épidémiologie des infections à *Mycoplasma conjunctivae* chez le bouquetin, le chamois et le mouton dans les Alpes suisses. Bull Soc Neuchâtel Sci Nat 120:27–34

Giacometti M, Janovsky M, Belloy L, Frey J (2002) Infectious keratoconjunctivitis of ibex, chamois and other Caprinae. Rev Sci Tech 21:335–345

Griffin D, Gonzalez R (1995) Correlational analysis of dyad-level data in the exchangeable case. Psychol Bull 118:430–439

Grignolio S, Rossi I, Bassano B, Parrini F, Apollonio M (2004) Seasonal variations of spatial behaviour in female Alpine ibex (*Capra ibex ibex*) in relation to climatic conditions and age. Ethol Ecol Evol 16:255–264

Gupta N, Prasad I, Himashree G, D'Souza P (2008) Prevalence of dry eye at high altitude: a case controlled comparative study. High Alt Med Biol 9:327–334. doi:10.1089/ham.2007.1055

Hardenberg A, Bassano B, Peracino A, Lovari S (2000) Male Alpine chamois occupy territories at hotspots before the mating season. Ethol 106:617–630. doi:10.1046/j.1439-0310.2000.00579.x

Hars J, Gauthier D (1984) Suivi de l'évolution de la kérato-conjonctivite sur le peuplement d'ongulés sauvages du Parc National de la Vanoise en 1983. Trav Sci Parc Nat Vanoise 14:157–210

Hengl T, Gruber S, Shrestha DP (2004) Reduction of errors in digital terrain parameters used in soil-landscape modelling. Int J Appl Earth Obs Geoinfo 5:97–112. doi:16/j.jag.2004.01.006

Marín MJ, Sola Y, Tena F, Utrillas MP, Campmany E, de Cabo X, Lorente J, Martínez-Lozano JA (2005) The UV index on the Spanish Mediterranean coast. Photochem Photobiol 81:659. doi:10.1562/2004-11-25-RA-380.1

Mavrot F, Vilei EM, Marreros N, Signer C, Frey J, Ryser-Degiorgis M-P (2012) Occurrence, quantification and genotyping of *Mycoplasma conjunctivae* in wild Caprinae with and without infectious keratoconjunctivitis. J Wildl Dis 48:619–631

Meile P, Giacometti M, Ratti P (2003) Der Steinbock. Biologie und Jagd. Salm Verlag, Bern

Murdoch IE, Morris SS, Cousens SN (1998) People and eyes: statistical approaches in ophthalmology. Br J Ophthalmol 82:971–973. doi:10.1136/bjo.82.8.971

Nesti I, Posillico M, Lovari S (2010) Ranging behaviour and habitat selection of Alpine chamois. Ethol Ecol Evol 22:215–231. doi:10.1080/03949370.2010.502316

Nicolet J (1985) Experiences on keratoconjunctivitis in chamois in Switzerland. In: Balbo T, Lanfranchi P, Rossi L, Stero P (eds) Atti Simposio internazionale sulla cheratocongiuntivie infettiva del camoscio. Univercittà, Torino, pp 53–61

Parrini F, Grignolio S, Luccarini S, Bassano B, Apollonio M (2003) Spatial behaviour of adult male Alpine ibex *Capra ibex ibex* in the

Gran Paradiso National Park, Italy. Acta Theriol 48:411–423. doi:10.1007/BF03194179

Ramp D, Caldwell J, Edwards K, Warton D, Croft D (2005) Modelling of wildlife fatality hotspots along the Snowy Mountain Highway in New South Wales, Australia. Biol Conserv 126:474–490. doi:10.1016/j.biocon.2005.07.001

Rosner B (1982) Statistical methods in ophthalmology: an adjustment for the intraclass correlation between eyes. Biometrics 38:105–114

Rosner B, Donner A, Hennekens CH (1979) Significance testing of interclass correlations from familial data. Biom 35:461–471

Ryser-Degiorgis M-P, Bischof DF, Marreros N, Willisch C, Signer C, Filli F, Brosi G, Frey J, Vilei EM (2009) Detection of *Mycoplasma conjunctivae* in the eyes of healthy, free-ranging Alpine ibex: possible involvement of Alpine ibex as carriers for the main causing agent of infectious keratoconjunctivitis in wild Caprinae. Vet Microbiol 134:368–374. doi:10.1016/j.vetmic.2008.08.005

Schnidrig-Petrig R, Salm UP (2009) Die Gemse - Biologie und Jagd. Salm Verlag, Bern

Tschopp R, Frey J, Zimmermann L, Giacometti M (2005) Outbreaks of infectious keratoconjunctivitis in alpine chamois and ibex in Switzerland between 2001 and 2003. Vet Rec 157:13–18

Vilei EM, Bonvin-Klotz L, Zimmermann L, Ryser-Degiorgis M-P, Giacometti M, Frey J (2007) Validation and diagnostic efficacy of a TaqMan real-time PCR for the detection of *Mycoplasma conjunctivae* in the eyes of infected Caprinae. J Microbiol Methods 70:384–386. doi:10.1016/j.mimet.2007.05.007