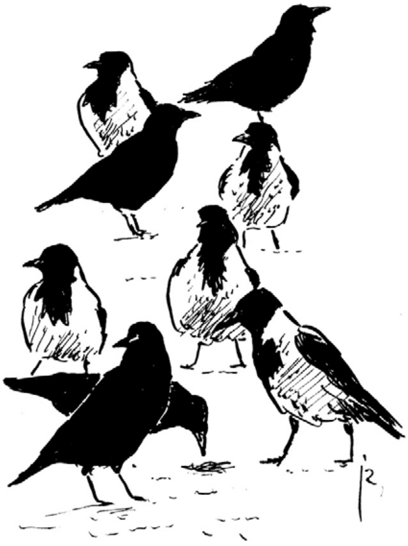
Assortative Mating of Carrion *Corvus corone* and Hooded Crows *C. cornix* in the Hybrid Zone in Eastern Germany

Author(s): Christoph Randler
Source: Ardea, 95(1):143-149.
Published By: Netherlands Ornithologists' Union
DOI: http://dx.doi.org/10.5253/078.095.0116
URL: http://www.bioone.org/doi/full/10.5253/078.095.0116

Assortative mating of Carrion *Corvus corone* and Hooded Crows *C. cornix* in the hybrid zone in eastern Germany

Christoph Randler[1]



Randler C. 2007. Assortative mating of Carrion *Corvus corone* and Hooded Crows *C. cornix* in the hybrid zone in eastern Germany. Ardea 95(1): 143–149.

Mating patterns in hybrid zones may be important for maintaining a stable hybrid zone. Theory suggests that hybrid zones are stable due to assortative mating outside the zone. To test whether assortative mating occurs in the crow hybrid zone in eastern Germany, I studied pairs of crows. The study area between Magdeburg and Dresden was divided into a grid and in each grid cell I recorded paired crows between 8 April and 12 July 2006. Carrion *Corvus corone* and Hooded Crows *C. cornix* mated assortatively given that the composition of observed pairs deviated significantly from what was expected by random mating; homotypic pairings, i.e. Carrion x Carrion or Hooded x Hooded Crows, were observed more and heterotypic pairings less often than expected. These findings contradict previous suggestions for this region but are in accordance with studies in other parts of Europe.

Key words: assortative mating, Carrion Crow, Hooded Crow, hybrid zone, hybridisation, mate choice

[1] University of Leipzig, Institute of Biology I, Johannisallee 21-23,
D-04103 Leipzig, Germany (randler@uni-leipzig.de)

INTRODUCTION

During the last ice age, many European animal populations became isolated in different refugia (allopatric populations), where they diverged into two (sub-)species. After the recession of the glaciation, these allopatric (sub-) species came into secondary contact (Mayr 1942, Barton & Hewitt 1985, Randler 2006, Swenson 2006). In the zone where the ranges of the two genetically and morphologically distinct populations overlap, they now interbreed and produce hybrids. Such hybrid zones provide opportunities to study factors affecting reproductive isolation, hybridisation and speciation (Barton & Hewitt 1985).

In birds, reproductive isolation between populations is often caused by the absence of mating (prezygotic isolation), rather than by reduced fitness of hybrids (postzygotic isolation; Grant & Grant 1992, 1997). One central aspect of prezygotic isolation is mate choice (Grant & Grant 1992, Randler 2002), which may foster assortative mating and reinforce premating barriers (Kirkpatrick 2000). The study of assortative mating in avian hybrid zones is interesting since it may provide evidence for stable as well as moving hybrid zones (Pearson 2000, Stein & Uy 2006). Assortative mating should narrow hybrid zones while female preferences for a specific trait of one species may make hybrid zones move (Stein & Uy 2006). Barton & Hewitt (1985) found that most avian hybrid zones were stable over time and narrow in comparison to both species' ranges. In their 'dynamic equilibrium model' Barton and Hewitt

(1985) suggest hybrid zones are stable due to assortative mating outside the zone which prevents them from widening. However, individuals reared outside the hybrid zone and dispersing into it may mate in a non-assortative manner. Evidence for assortative mating has been found in several avian hybrid zones such as in skuas *Catharacta* sp. of the Antarctic peninsula (Ritz *et al.* 2006), sapsuckers *Sphyrapicus* in California (Johnson & Johnson 1985) and buntings *Passerina* (Baker & Boylan 1999). However, in some hybrid zones assortative mating was absent (Bronson *et al.* 2005) or extremely weak (Good *et al.* 2000). Even in the same hybridising species pair individuals in some populations mated assortatively and others not (Moore 1987, Wiebe 2000).

The Carrion Crow *Corvus corone* is all black and breeds predominantly in western Europe while the Hooded Crow *Corvus cornix* is black and grey and breeds from eastern Europe through Asia. The crow hybrid zone extends from Scotland in the northwest through much of Central Europe to northern Italy. The continental stretch of the zone is approximately 1300 km long and between 50 and 150 km wide (Meise 1928). The crow hybrid zone is considered to have arisen due to secondary contact (Cook 1975, Barton & Hewitt 1985, Rolando 1993).

Assortative mating between crow species was found in Italy (Saino & Villa 1992, Rolando 1993), but not in Scotland (Picozzi 1976) and was suggested not to occur in Germany (Meise 1928; Glutz von Blotzheim & Bauer 1993). These contradicting results from different parts of the hybrid zone encourage further analyses (Haas & Brodin 2005). Previous results might reflect differences in the ecological situations (subalpine valleys in Italy, atlantic climate in Scotland and northern Germany). Surprisingly, few studies were carried out in the eastern German part of the zone. Most studies published in the German literature claim a much wider hybrid zone compared to Meise (1928), suggesting that random mating might increase the width of the zone (Bährmann 1960; Steffens *et al.* 1998). I studied mating patterns to test these suggestions.

METHODS

Data collection

I observed 242 pairs of crows (n = 484 individuals) between 8 April and 12 July 2006. Observations of crows were mapped on 1:100 000 survey maps to avoid sampling the same individuals twice. Pairing status could be assessed with some practise (Risch & Andersen 1998, Loman 1985, Haas & Brodin 2005) by observing social behaviour and territorial defence, e.g. when pair mates were close together and defended food against others or when they were near their nests. During April to June this is easily possible because crows possess large territories (Loman 1985).

Crows were observed and pair status was checked in the core area of the hybrid zone in eastern Germany, between Magdeburg and Dresden along a stretch of approximately 200 km in length. The study area ranged between 52°15'N, 11°28'E in the northwest and 51°00'N, 13°36'E in the southeast (Fig. 1). Here, the hybrid zone stretches

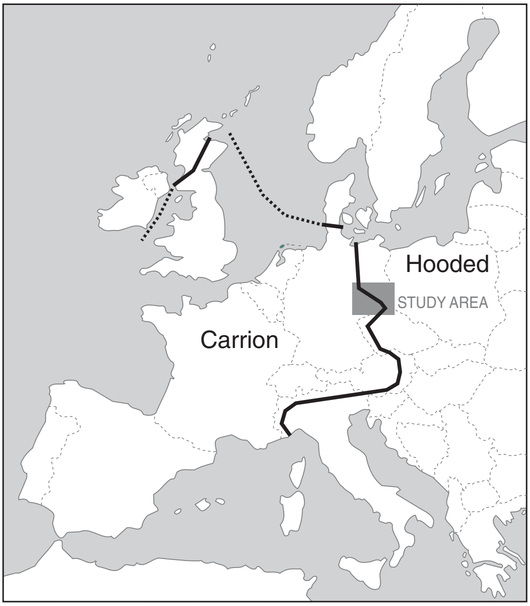


Figure 1. Approximate extent of the crow hybrid zone in central Europe and the location of the study area.

in a diagonal manner from NW to SE. The undulating landscape is dominated by arable land (mainly cereals (wheat), and some grassland). To cover the zone systematically, I divided the study area into a grid of approximately 18 x 18 km (ten geographical minutes in latitude and twelve in longitude; Gauß-Krüger-grid). Each of the grid cells was visited at least once, travelling slowly by car on public roads to search for crows using 10 x 50 binoculars and a 30x telescope.

Crows were classified into five phenotypes (according to Saino & Villa 1992, Rolando 1993, Haas & Brodin 2005): Carrion Crows, Carrion-type hybrids, intermediates, Hooded-type hybrids and Hooded Crows. Hybrids are depicted e.g. in Parkin *et al.* (2003) or Spiridonova & Kryukov (2004) and are easy to identify (Randler 2004). Fig. 2 shows an example of a hooded-type hybrid.

Statistical analysis

I used expected frequencies as calculated following the methods proposed by Rolando (1993). For each 18 x 18 km grid cell, I calculated the expected frequencies assuming that the sampling probability for each mate is independent of that of the other (Rolando 1993). The expected frequencies were calculated from numbers of phenotypes in pairs occurring in each grid cell (details in Rolando 1993, p. 81). Assume, for example, in one grid cell the proportion of Carrion and Hooded Crows were 0.40 and 0.60, with 10 pairs observed. Accordingly, the expected frequency of Hooded x Hooded pairs was 3.6 (0.6 x 0.6 x 10), that of Carrion x Carrion was 1.6 (0.4 x 0.4 x 10) and that of Carrion x Hooded was 4.8 (0.6 x 0.4 x 2 x 10). The expected frequencies for each grid cell were added and compared with the observed frequencies of the total area. To compare expected with observed frequencies, I used the G-test (Sokal & Rohlf 1991) in a spreadsheet version provided by John H. McDonald (for more details see: http://udel.edu/~mcdonald/statgtestgof.html). All tests were two-tailed.

Figure 2. 'Hooded-type' hybrid between Carrion and Hooded Crow.

RESULTS

A total of 134 Hooded Crows (27.7%), 275 Carrion Crows (56.8%) and 75 hybrids (15.5%) were observed. The hybrids were composed of 25 (33.3%) Carrion type, 23 intermediate type (30.7%) and 27 (36.0%) Hooded type. The three hybrid types were equally common ($G_2 = 0.32$; $P = 0.85$). Mating patterns of crosses among the five phenotypes are summarized in Table 1.

For the G-tests I categorized the phenotypes into pure Hooded Crow, pure Carrion Crow and hybrid (Hooded-type, intermediate, Carrion-type). Between these categories, crow phenotypes did not mate assortatively ($G_5 = 8.44$, $P = 0.13$; Table 1), i.e. mating patterns did not differ significantly from what was expected if random mating occurred. However, the mating preferences of hybrids could not be predicted *a priori* (Rolando 1993). It is unclear whether hybrids should pair

Table 1. Mating patterns of crow phenotypes in the hybrid zone between Carrion Crow and Hooded Crow in eastern Germany. $N = 242$ pairings.

		Partner 1				
		Carrion	Carrion-type	Intermediate	Hooded-type	Hooded
Partner 2	Carrion	110				
	Carrion-type	15	2			
	Intermediate	12	0	2		
	Hooded-type	7	2	3	4	
	Hooded	21	4	4	7	49

Table 2. Studies on assortative mating in Hooded and Carrion Crows. Details about ecology, geographic region and width of the zone given, studies organised from west to east and north to south. Test statistics recalculated from original data. Data from the present study included.

Study	
1	Picozzi (1976), NE Scotland; no ecological segregation, width of the zone: no details given, no pure Hooded Crows present.
2	Risch & Andersen (1998), Amrum, Wadden Sea, Germany; no ecological segregation, no pure Hooded Crows present, width of zone not reported.
3	Randler (present study), eastern Germany; arable land, no sharp ecological transitions, no ecological segregation, width of the zone: 50–100 km.
4	Rolando (1993), Northern Italy, Alpine Valley; altitudinal segregation of Crow species, ecotone, width of the zone: less than 50 km.
5	Saino & Villa (1994), Northern Italy, Alpine Valley and intensively cultivated plains; differences in habitat use, width of the zone: 10–13 km.
6	Kryukov & Blinov (1994), Siberia; assortative mating occurs, Hooded and Carrion crow *C. c. orientalis* differ in migration behaviour and Carrion Crows start nesting earlier, width of the zone: 150 km, no details about pairings given. Share of hybrids up to 32% in the centre.

		Carrion x Carrion	Hybrid x Hybrid	Hooded x Hooded	Carrion x Hybrid	Carrion x Hooded	Hooded x Hybrid	G-statistic	*P*
1	Expected[a]	20.8	4.8	-	20.2	-	-	$G_2 = 1.61$	0.45
	Observed	19	3	-	24	-	-		
2	Expected	26.63	2.63		16.74	-	-	$G_2 = 6.74$	0.03
	Observed	30	6		10	-	-		
3	Expected	102.95	9.06	41.71	37.69	31.39	19.17	$G_2 = 13.44$ (see text)	0.001
	Observed	110	13	49	34	21	15		
4	Expected	42.15	3.26	42.28	11.18	27.28	15.65	$G_5 = 13.71$	0.02
	Observed	51	1	41	9	16	24		
5	Expected	15.1	2.9	54.7	11.1	42.1	25.1	$G_5 = 147.16$	< 0.001
	Observed	35	6	70	3	10	27		

[a] Expected frequencies calculated after Rolando (1993).

with another hybrid or backcross with a Hooded or a Carrion Crow. The information about hybrid-hybrid pairings is irrelevant when testing whether Hooded and Carrion Crows mate assortatively with conspecifics (Rolando 1993). Therefore, I calculated the G-test based on both parental types and their mixed pairs. Hooded and Carrion Crows mated assortatively and the pattern of observed pairings deviated significantly from what was expected by random mating ($G_2 = 13.44$; $P = 0.001$). Homotypic pairings, i.e. Carrion x Carrion or Hooded x Hooded Crows, were observed more and heterotypic pairings (Hooded x Carrion Crow) less often than expected (Table 2).

DISCUSSION

This study provides evidence for assortative mating in the crow hybrid zone in eastern Germany. The data suggest that homotypic pairings are more common and mixed pairings between Carrion and Hooded Crow less common than expected by random pairing. This contradicts previous suggestions (Meise 1928, Glutz von Blotzheim & Bauer 1993) as both sources suggested random mating in the eastern German hybrid zone. Instead, my results are in accordance with other recent studies that also reported assortative mating in crows. A total of five studies found evidence in favour of assortative mating and one not (exception: Picozzi 1976, see Table 2). Unfortunately, these results are insufficient for testing predictions using meta-analytical tools. In detail, in Scotland, evidence for assortative mating could not be found, perhaps because sample size was low and Hooded Crows did not occur. Risch & Andersen (1998) found assortative mating on the island of Amrum, where homotypic pairs were more frequent, although Hooded Crows did not occur on Amrum. In the alpine hybrid zone in Italy, assortative mating occurred, but (sub-) species showed a different altitudinal distribution (Rolando & Saino 1994, Saino & Villa 1992, Rolando 1993). In eastern Germany there is no altitudinal gradient separating both forms (see also Haas & Brodin 2005) and in contrast to Italy (Saino & Villa 1992) there is no ecotone between the parental species habitats where hybrids may be superior. Further, there seems no relationship between the width of the hybrid zone and assortative mating as it occurs in narrow zones in the Alps (Rolando 1993, Saino & Villa 1992) as well as in wider zones in Siberia (hybridization between Hooded Crow and Carrion Crow *C. c. orientalis*; Kryukov & Blinov 1994).

Assortative mating need not be the results of active mate preferences, but may result from the unequal spatial and temporal distribution of various phenotypes (‘prevalence-hypothesis’; Findlay 1987, Saino & Villa 1992, Rolando 1993). For example, when one species arrives earlier in spring than another, this must inevitably lead to assortative mating. Also, at each end of the hybrid zone, usually one species is more common than the other, again leading to a higher probability of assortative mating. However, these hypotheses were rejected in the present data set because the expected frequencies were calculated on the basis of each of the grid cells separately (see also Rolando 1993) and, second, both crow species are largely resident in eastern Germany.

If assortative mating is almost absent, hybridisation occurs for several reasons (see below). Resulting hybrids may be inferior to either or both their parental species and there may be selection against hybrids (Picozzi 1976, Saino & Villa 1992). Therefore, proximate cues must exist on which the choosing sex bases its decision. Such cues may be types of behaviour, vocalisations, and plumage traits. Different factors may account for ongoing hybridisation in crows. First, mistakes in mate recognition may lead to hybridisation (Randler 2002). However, crow species are clearly distinguishable by plumage, and Brodin & Haas (2006) found in a computer simulation using neural networks that both Hooded and Carrion Crows could be easily trained to recognise conspecifics. Therefore, plumage cues seem sufficient for species recognition. However, the choice of a heterospecific partner may be a result of misimprinting (Randler 2005). As long as mixed pairings between both species occur, imprinting would lead to heterospecific mate choice of their offspring, and, hence, maintain the hybrid zone. Studies about imprinting in crows in the hybrid zone are needed to support this argument and to distinguish between visual and audio imprinting.

A second reason for ongoing mixed pairings could be the scarcity of conspecific partners (Randler 2002, 2006). Crows may be able to discriminate conspecifics from heterospecifics but a lack of conspecific partners might lead to mixed pairing. Although reproductive success in mixed pairings or in pairs with one hybrid is reduced (in another part of the hybrid zone; Saino & Villa 1992), resulting hybrids are fertile. Mixed pairing when conspecific partners are scarce could be interpreted as 'making the best of a bad job' as proposed by Baker & Boylan (1999). An interesting question in this context might be whether hybrid costs could be avoided as in the hybrid zone of flycatchers *Ficedula* where females pair heterospecifically when conspecifics are absent but gather conspecific sperm from extrapair copulations (Veen *et al.* 2001; discussion in Randler 2006a).

The results show that Carrion and Hooded Crows mate assortatively. By itself, assortative mating should narrow a hybrid zone until interbreeding stops and two distinct species coexist (Barton & Hewitt 1985). However, the crow hybrid zone is rather narrow and remains more or less spatially stable, indicating that other processes counteract the effect of assortative mating. One such process may be the immigration of inexperienced individuals from outside the hybrid zone that are more likely to pair with a heterospecific. However, dispersal of individuals into and out of a hybrid zones has not been investigated in crows.

ACKNOWLEDGEMENTS

I highly appreciate the suggestions made by an anonymous reviewer and by the associate editor, Ken Kraaijeveld.

REFERENCES

Bährmann U. 1960. Untersuchungen an einer Krähenpopulation im Mischgebiet der Raben- und Nebelkrähe in Deutschland östlich der Elbe. Abh. Ber. Staatl. Mus. Tierkunde Dresden 25: 71–79.

Baker M.C. & Boylan, J.T. 1999. Singing behavior, mating association and reproductive success in a population of hybridizing Lazuli and Indigo buntings. Condor 101: 493–504.

Barton N.H. & Hewitt G.M. 1985. Analysis of hybrid zones. Ann. Rev. Ecol. Syst. 16: 113–148.

Brodin A. & Haas F. 2006. Speciation by perception. Anim. Behav. 72: 139–146.

Bronson C.L., Grubb T.C., Sattler G.D. & Braun M.J. 2005. Reproductive success across the black-capped chickadee (*Poecile atricapillus*) and carolina chickadee (*P. carolinensis*) hybrid zone in Ohio. Auk 122: 759–772.

Cook A. 1975. Changes in the Carrion/Hooded Crow hybrid zone and the possible importance of climate. Bird Study 22: 165–160.

Cooke F., Finney G.H. & Rockwell R.F. 1976. Assortative mating in lesser snow geese. Behav. Genet. 6: 127–140.

Delestrade A. 2001. Sexual size dimorphism and positive assortative mating in Alpine Chough (*Pyrrhocorax graculus*). Auk 118: 553–556.

Findlay C.S. 1987. Non-random mating: a theoretical and empirical overview with specific reference to birds. In: Cooke F. & Buckley P.A. (eds) Avian Genetics: 289–319. Academic press, London.

Glutz von Blotzheim U.N. & Bauer K.M. 1993. Handbuch der Vögel Mitteleuropas. Band 13, Passeriformes 4. Aula, Wiesbaden.

Good T.P., Ellis J.C., Annett C.A. & Pierotti R. 2000. Bounded hybrid superiority in an avian hybrid zone: effects of mate choice, diet, and habitat choice. Evolution 54: 1774–1783.

Grant P.R. & Grant B.R. 1992. Hybridization of bird species. Science 256: 193–197.

Grant P.R. & Grant B.R. 1997. Hybridization, sexual imprinting, and mate choice. Am. Nat. 149: 1–28.

Haas F. & Brodin A. 2005. The Crow hybrid zone in southern Denmark and northern Germany. Ibis 147: 649–656.

Johnson N.K. & Johnson C.B. 1985. Speciation in sapsuckers (Sphyrapicus): II. Sympatry, hybridization, and mate preferences in *S. ruber dagetti* and *S. nuchalis*. Auk 102: 1–15.

Kirkpatrick M. 2000. Reinforcement and divergence under assortative mating. Proc. R. Soc. Lond. B 267: 1649–1655.

Krüger O., Lindström J. & Amos W. 2001. Maladaptive mate choice maintained by heterozygote advantage. Evolution 55: 1207–1214.

Kryukov A.P. & Blinov V.N. 1994. Hybrid Zone of Hooded and Carrion Crow in Siberia. Research Notes on Avian Biology 1994: Selected Contributions from the 21st International Ornithological Congress. J. Ornithol. 135: 47.

Loman J. 1985. Social organization in a population of the hooded crow. Ardea 73: 61–75

MacDougall A.K. & Montgomerie R. 2003. Assortative mating by carotenoid plumage colour: a quality indicator in American goldfinches, *Carduelis tristis*. Naturwissenschaften 90: 464–467.

Mayr E. 1942. Animal species und evolution. Harvard University Press, Harvard.

Meise W. 1928. Die Verbreitung der Aaskrähe (Formenkreis *Corvus corone* L.). J. Ornithol. 76: 1–204.

Moore W.S. 1987. Random mating in the Northern Flicker hybrid zone: implications for the evolution of bright and contrasting plumage patterns in birds. Evolution 41: 539–546.

Parkin D., Collinson M., Helbig A.J., Knox A.G., Sangster G. 2003. The taxonomic status of Carrion and Hooded Crows. Brit. Birds 96: 274–290.

Pearson S.F. 2000. Behavioral asymmetries in a moving hybrid zone. Behav. Ecol. 11: 84–92.

Picozzi N. 1976 Hybridization of Carrion and Hooded Crows *Corvus c. corone* and *Corvus c. cornix* in northeastern Scotland. Ibis 118: 254–257.

Randler C. 2002. Hybridization, mixed pairing and female choice. Anim. Behav. 63: 103–119.

Randler C. 2004. Frequency of bird hybrids: does detectability make all the difference? J. Ornithol. 145: 123–128.

Randler C. 2005. Do forced extrapair copulations and interspecific brood amalgamation facilitate natural hybridisation in wildfowl? Behaviour 142: 477–488.

Randler C. 2006. Behavioural and ecological correlates of natural hybridization in birds. Ibis 148: 459–467.

Randler C. 2006a. Extrapair paternity and hybridization in birds. J. Avian Biol. 37: 1–5.

Risch M. & Andersen L. 1998. Selektive Partnerwahl der Aaskrähe (*Corvus corone*) in der Hybridisierungszone von Rabenkrähe (*C. c. corone*) und Nebelkrähe (*C. c. cornix*). J. Ornithol. 139: 173–177.

Ritz M.S., Hahn S. Janicke T. & Peter H.U. 2006. Hybridisation between South polar skua (*Catharacta maccormicki*) and Brown skua (*C. antarctica lonnbergi*) in the Antarctic Peninsula region. Polar Biol. 29: 153–155.

Rolando A. 1993. A study on the hybridization between Carrion and Hooded Crow in Northwestern Italy. Ornis Scand. 24: 80–83.

Rolando A. & Saino N. 1994. Assortative mating among Eurasion Crow Phenotypes across a Hybrid Zone. Research Notes on Avian Biology 1994: Selected Contributions from the 21st International Ornithological Congress. J. Ornithol. 135: 48.

Roulin A. 1999. Non-random pairing by male Barn Owls (*Tyto alba*) with respect to a female plumage trait. Behav. Ecol. 10: 688–695.

Saino N. & Villa S. 1992. Pair composition and reproductive success across a hybrid zone of carrion and hooded crows. Auk 109: 543–555.

Sokal R. R. & Rohlf F.J. 1995. Biometry: the principles and practice of statistics in biological research. Third edition. Freeman & Co., New York.

Spiridonova L.N. & Kryukov A.P. 2004. Genetic variability of Carrion and Hooded crows and their hybrids according to RAPD-PCR data. Cytol. Genet. 38: 27–35.

Steffens R., Saemann D. & Größler K. 1998. Die Vogelwelt Sachsens. Gustav Fischer, Jena.

Stein A.C. & Uy J.A.C. 2006. Unidirectional introgression of a sexually selected trait across an avian hybrid zone: a role for female choice? Evolution 60: 1476–1485.

Swenson N.G. 2006. Gis-based niche models reveal unifying climatic mechanisms that maintain the location of avian hybrid zones in a North American suture zone. J. Evol. Biol. 19: 717–725.

Veen T., Borge T., Griffith S.C., Sætre G.-P., Bures S., Gustafsson L. & Sheldon B.C. 2001. Hybridization and adaptive mate choice in flycatchers. Nature 411: 45–50.

Wiebe K.L. 2000. Assortative mating by color in a population of hybrid northern flickers. Auk 117: 525–529.

SAMENVATTING

Zwarte Kraaien *Corvus corone* en Bonte Kraaien *Corvus cornix* hybridiseren met elkaar in een smalle, langgerekte zone die zich uitstrekt van Schotland tot in Italië. Het bestuderen van factoren die bijdragen aan het instandhouden van zulk een hybridisatiezone kan mogelijk inzicht verschaffen in soortsvorming. In dit onderzoek werd nagegaan of Zwarte en Bonte Kraaien in de hybridisatiezone in het oosten van Duitsland een voorkeur hadden voor partners van de eigen soort. Hiertoe werden de partners van 242 kraaienpaartjes getypeerd als Zwarte Kraai, Bonte Kraai of Zwarte x Bonte Kraai. Pure Zwarte en Bonte Kraaien bleken vaker met de eigen soortgenoot te paren dan wanneer ze een willekeurige partner hadden gekozen. Hybride vogels bleken geen voorkeur te hebben. Een overzicht van de literatuur liet zien dat dergelijke soortspecifieke paarvorming op alle plekken in de hybridisatiezone voorkomt, behalve in Schotland. Eerdere suggesties dat de mate van soortspecifieke paarvorming samenhangt met de plaatselijke ecologie werd hiermee niet bevestigd. (KK)

Corresponding editor: Ken Kraaijeveld
Received 6 December 2006; accepted 27 March 2007