ANOSMIA IN ZnSO4-TREATED PIGEONS: LOSS OF OLFACTORY INFORMATION DURING ONTOGENY AND THE ROLE OF SITE FAMILIARITY IN HOMING EXPERIMENTS

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Summary

Depriving homing pigeons of olfactory information by spraying an 18% zinc sulphate solution onto their olfactory ephithelium results in anosmia that persists for at least 5 days (tested using the 'orienting response'). To study whether anosmic zinc-sulphate-treated pigeons ($ZnSO_4$-pigeons) were able to compensate for their loss of olfactory information by using familiar landmarks, we made releases from two different distances (9–24km, 54–70km). The birds had homed once before from each release site. At shorter distances, $ZnSO_4$-pigeons, sham-treated controls and untreated super controls were well oriented. At the longer distances, the anosmic pigeons and the sham-treated controls did not show any directional preference whereas the super controls were oriented towards home. In all cases, however, fewer anosmic pigeons homed successfully and their homing speed was slower than the speed of either control group.

In order to determine whether experience during ontogeny affects the orientation behaviour, a group of young pigeons was reared anosmically by application of $ZnSO_4$ solution once a week. A second group of experimental pigeons was made anosmic just before the critical release. When released at unfamiliar sites located between 15 and 24km from home, all but the group of short-term $ZnSO_4$-pigeons were well oriented. Neither permanently nor temporarily anosmic $ZnSO_4$-pigeons showed good homing performances. These data indicate that homing pigeons determine their home direction by relying on the navigational factors they have collected during rearing. The paradoxical finding that anosmically reared pigeons were oriented towards home but did not home successfully leads to the questions of whether intra-nasal irrigation with $ZnSO_4$ solution results in extensive brain damage or whether initial orientation and the rest of the homing process are based on different mechanisms.

Introduction

Among the many hypotheses put forward to explain pigeon homing (reviewed by Schmidt-Koenig, 1979, 1985, 1987; Wiltschko and Wiltschko, 1978, 1982, 1987;

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Waldvogel, 1989; Wallraff, 1988*a*; Papi, 1990), olfactory navigation remains the most contentious. A major source of dispute is the effectiveness of the techniques applied to achieve olfactory deprivation (Wallraff, 1988*b*) in tests of homing ability in anosmic pigeons.

Intra-nasal irrigation with zinc sulphate (ZnSO₄) is a new, rapid and simple method for olfactory deprivation in pigeons. Treatment with 18% ZnSO₄ solution results in anosmia that persists for at least 5 days. Effects on the pigeons' optical and acoustical systems could not be detected when tested by the 'orienting response' (Schlund, 1991*a*, 1992). Manipulations with ZnSO₄ caused weight loss of the bulbus olfactorius and damaged the nervus olfactorius in mice (Margolis *et al.* 1974; Harding *et al.* 1978), and this could not be excluded for pigeons.

Application of ZnSO₄ alters or destroys the olfactory mucosa and has been found to be effective in hamsters, mice and sea turtles (reviewed by Alberts, 1974). After some time, the olfactory mucosa regenerates, re-establishing olfactory capacity (Smith, 1951; Margolis *et al.* 1974; Matulionis, 1975, 1976; Harding *et al.* 1978; Cancalon, 1982).

The present paper reports on a series of experiments designed to test the homing ability of anosmic pigeons. The birds were made anosmic with zinc sulphate. With the help of the 'orienting response', we tested the olfactory capabilities of every individual.

Our first series of experiments was designed to test whether anosmic pigeons are able to compensate for the loss of olfactory information when released from familiar sites. For the second experimental series, we reared pigeons anosmically by repeated application of ZnSO₄. Thus, we prevented them from collecting olfactory information during ontogeny and studied how this affected their orientation at unfamiliar sites. In addition, we compared them with a second group of experimental pigeons which were made anosmic just before the critical releases.

Materials and methods

Release of temporarily anosmic pigeons from familiar sites

Pigeons (*Columba livia*), between 2 and 5 years old, were housed at our loft near Tübingen (Germany). They had all completed a standard training programme of flights of up to 40km from the cardinal compass directions. The birds had performed numerous single homing flights from various distances and directions in previous years.

In order to familiarise the pigeons with the release sites, they were released as a flock from that site a few days before the critical experiment. We used four sites between 9 and 24km (distance I) and four sites between 54 and 70km (distance II) from the loft, located as symmetrically as possible around it.

Pigeons that had homed from the flock releases were subdivided into three groups as uniformly as possible in terms of age, experience and number. In the critical experiment each individual was released only once.

(1) *Experimentals* (ZnSO₄-pigeons). Pigeons were treated on two successive days with an 18% ZnSO₄ solution: 11.2g of ZnSO₄·H₂O (179.45 g mol⁻¹), 0.1g of Tagat 02 (emulsifier; trade name) and 25g of Frigen 12 in 63.7g of distilled water. Approximately 0.2–0.3ml was sprayed through the choanes into the nasal cavities with an aerosol can.

The day before the critical release and immediately after they had arrived at the loft, the olfactory perception in $ZnSO_4$-pigeons was tested using cardiac responses. We tested the olfactory capabilities of every pigeon in response to odorous stimuli. Only pigeons individually confirmed to be unable to smell (subsequently called 'anosmic') were used for the release and subsequent statistical analysis. Intra-nasal irrigation with 18% $ZnSO_4$ solution results in anosmia that was found to persist for at least 5 days (Schlund, 1992). For further descriptions of the treatment, the experimental apparatus and the techniques for the olfactory test, see Schlund (1990, 1992).

(2) *Controls* (C). Control birds were handled like the $ZnSO_4$-pigeons. Instead of treatment with $ZnSO_4$ solution, they were sham-treated with a corresponding solution without $ZnSO_4$.

(3) *Super controls* (SC). These were completely untreated pigeons.

Releases of pigeons reared anosmically

We received pigeons at the age of 3–4 weeks from local breeders. On the day of arrival at our loft near Tübingen (Germany), the pigeons were divided into five groups.

(1) *Permanent anosmic pigeons* (P-$ZnSO_4$). Pigeons were reared anosmically by treating them with $ZnSO_4$ solution on the day of arrival and subsequently once a week. To ensure that the birds were anosmic, the olfactory capabilities of each pigeon were tested every week. Additionally, the olfactory perception in P-$ZnSO_4$-pigeons was tested immediately upon arrival at the loft after the critical releases.

(2) *Permanent controls* (PC). Controls were treated similarly, except that the control spray, which was delivered once a week, did not contain $ZnSO_4$. The olfactory capability of each pigeon was tested several times during the experimental period.

(3) *Short-term experimentals* (ST-$ZnSO_4$). Pigeons were made anosmic a few days before the critical release by treating them with $ZnSO_4$ on two successive days. The second day of treatment corresponded with the time of $ZnSO_4$-application to the P-$ZnSO_4$-pigeons. On the following day and immediately after homing, the olfactory capability of every pigeon was tested.

(4) *Short-term controls* (STC). Controls were handled like the short-term $ZnSO_4$-pigeons, but sham-treated with control spray lacking $ZnSO_4$.

(5) *Super controls* (SC). These were completely untreated pigeons.

As soon as they were able to fly (at the age of 5–6 weeks), pigeons were usually allowed to fly around the loft. When they were about 10 weeks old, we began the standard training programme consisting of eight flock releases from the cardinal compass directions at distances of up to 6km from the loft. The critical releases began at the age of 15 weeks. In the critical experiment, each individual was released only once.

For the critical releases, we used four sites between 15 and 24km from the loft.

Release procedure

The pigeons were released singly, alternating between groups. They were followed visually using 7×50 binoculars with an incorporated magnetic compass until they vanished from sight. The vanishing bearings were recorded to the nearest 1°. Homing

times were also recorded. All releases were performed under sunny conditions in 1990 and 1991.

Data analysis

For circular statistics, we considered only vanishing bearings. For each sample and release distance the length (**a**) and direction (α) of the mean vector were calculated with respect to geographical north. Each sample was tested for uniformity preferences using the Rayleigh test (Batschelet, 1981). For samples with significant directional preferences, we calculated the 95% confidence interval of the mean direction ('Bootstrap' with 500 replica; Cabrera *et al.* 1991). If the 95% confidence interval of the mean direction did not include the home direction, the two directions were considered to differ.

Vanishing bearings of different samples were compared by means of first-order (Watson U^2-test) and second-order statistics (Hotelling test: d.f. 2,5).

Homing perfomances were compared using the Mann–Whitney *U*-test (Siegel, 1956). For this, homing speed per release was standardised by the median of super controls. Pigeons that homed within 3 days after release (lofts were checked every morning and evening) were assigned relative homing speeds: homed on the evening of the releasing day, 0.9; homed the next day, morning, 0.8; homed the next day, evening, 0.7; homed the second day, morning, 0.6; homed the second day, evening, 0.5; homed the third day, morning, 0.4; homed the third day, evening, 0.3; did not home, 0.1.

For each release, standardisation was performed separately before summarising the data per distance and per group. Tests were run on a PC with the help of SAS (1987).

Results

According to the results of heart rate tests after release, two ZnSO$_4$-pigeons used for releases from familiar sites (distance I and II) and nine ZnSO$_4$-pigeons in the second experimental series (eight ST-ZnSO$_4$, one P-ZnSO$_4$) were able to smell. They were eliminated from the analyses.

Releases of temporarily anosmic pigeons from familiar sites

Distance I

The data for all releases are given in Table 1. If pooled with respect to home, ZnSO$_4$-pigeons, controls and super controls showed directional preference (Rayleigh test). The 95% confidence interval of the direction of the mean vector of all three groups included the home direction (Fig. 1).

The two groups of controls were indistinguishable in comparisons using both first- and second-order statistics. Also, the ZnSO$_4$-pigeons did not differ from either of the two groups of controls (Table 2).

The homing speed of ZnSO$_4$-pigeons was slower than the speed of either group of controls. This was true for the single releases (Table 1) as well as for the combined releases pooled with respect to the median of super controls, as shown in Fig. 2 (Mann–Whitney *U*-test: SC-ZnSO$_4$: $z=4.88$, $P<0.001$; C-ZnSO$_4$: $z=4.16$, $P<0.001$). There was no difference between the two control groups (Mann–Whitney *U*-test: $z=0.67$, $P>0.05$).

Table 1. *Initial orientation of temporarily anosmic pigeons in single releases from familiar sites, distance I (9–24km)*

Site	β	km	Group	α	**a**	N_R	N_B	$km\,h^{-1}$	N_h
Pli	221°	23.9	SC	267°	0.74***	10	9	37.8	10
			C	268°	0.76***	10	9	40.7	10
			ZnSO₄	280°	0.60	8	7	23.4	8
Fro	45°	15.8	SC	31°	0.74***	10	10	28.6	10
			C	360°	0.63**	10	10	37.2	10
			ZnSO₄	24°	0.63*	10	9	8.6	7
Duß	342°	9.2	SC	324°	0.64*	10	7	17.9	10
			C	6°	0.31	10	7	28.3	10
			ZnSO₄	338°	0.08	10	7	16.7	10
Ler	140°	13.4	SC	151°	0.67*	10	7	32.9	10
			C	114°	0.72***	10	10	29.7	10
			ZnSO₄	146°	0.50	10	9	11.3	7

Sites (Pli, Plieningen; Fro, Frommenhausen; Duß, Dußlingen; Ler, Lerchenberg); β, home direction; km, distance of release site from home; group (SC, super controls; C, controls; ZnSO₄, experimentals); α, **a**, direction and length of mean vector (north=0°); levels of significance under the Rayleigh test (*$P<0.05$, **$P<0.01$, ***$P<0.005$); N_R, number of pigeons released; N_B, number of vanishing bearings; $km\,h^{-1}$, median of homing speed; N_h, number of pigeons homed within 3 days.

Table 2. *Initial orientation of temporarily anosmic pigeons at distances I (9–24km) and II (54–70km) tested by means of first-order and second-order statistics*

	C (N=36)	ZnSO₄ (N=32)
Distance I		
SC (N=33)	$U^2=0.063$	$U^2=0.117$
	$T^2=4.05$	$T^2=5.60$
C	–	$U^2=0.053$
	–	$T^2=0.66$

	C (N=32)	ZnSO₄ (N=31)
Distance II		
SC (N=35)	$U^2=0.081$	$U^2=0.169$
	$T^2=0.87$	$T^2=1.93$
C	–	$U^2=0.071$
	–	$T^2=0.41$

The results are pooled with respect to home.
First-order statistics, Watson U^2-test; second-order statistics, Hotelling test; d.f.=2, 5.
For further details, see Table 1.

Distance II

Table 3 summarises the principal data for single releases. If pooled with respect to home, it was only the super controls that showed a directional preference (Rayleigh test)
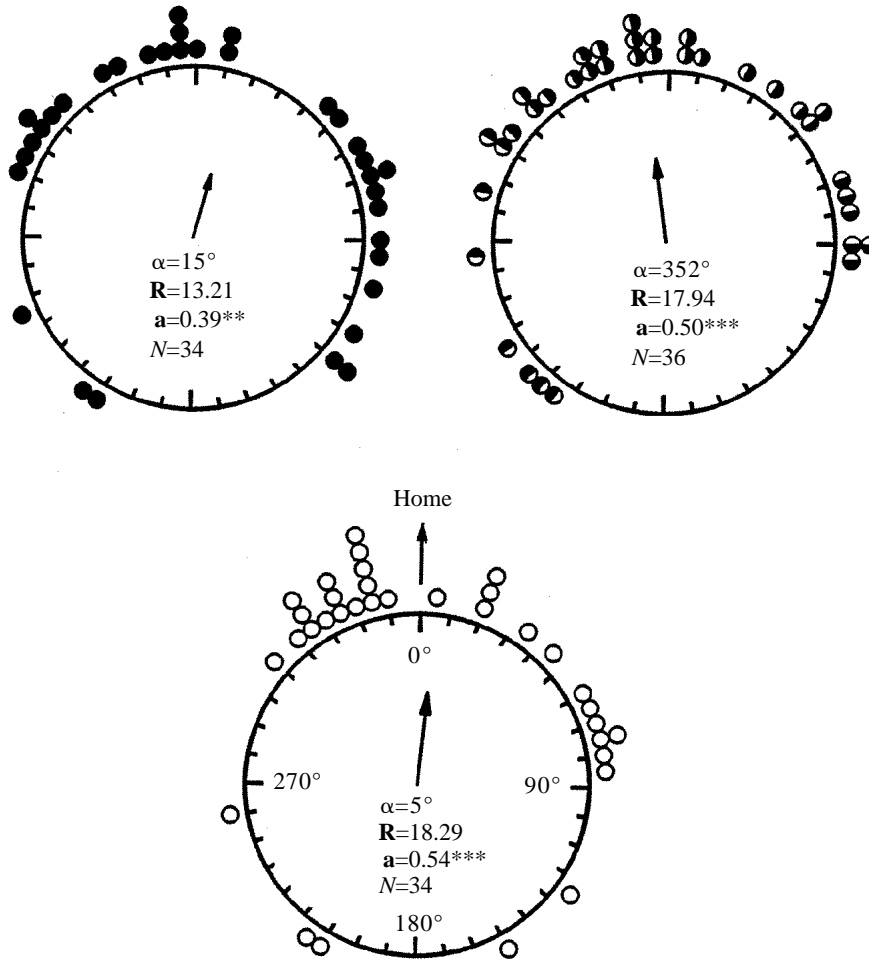
Fig. 1. Initial orientation of ZnSO$_4$-pigeons (●), C-pigeons (◐) and SC-pigeons (○) summarised with respect to home (0°). Releases from familiar sites, distance I. α, **R**, direction and length, respectively, of the mean vector; **a**, length of the mean vector with respect to 1; N, sample size; levels of significance using the Rayleigh test, *$P<0.05$, **$P<0.01$, ***$P<0.005$.

and for whom the 95% confidence interval of the direction of the mean vector included the home direction. The distribution of the pooled vanishing bearings of the ZnSO$_4$-pigeons and C-pigeons, however, did not differ from random (Fig. 3).

The two groups of controls were indistinguishable according to first-order and second-order statistics. Also, there was no difference between the ZnSO$_4$-pigeons and controls and between ZnSO$_4$-pigeons and super controls, even though the Watson-U^2 test approached significance ($P<0.1$) (Table 2).

As for releases from distance I, the homing speed of ZnSO$_4$-pigeons was slower than the homing speed of either group of controls (Fig. 4; Mann–Whitney U-test: $z=3.64$,
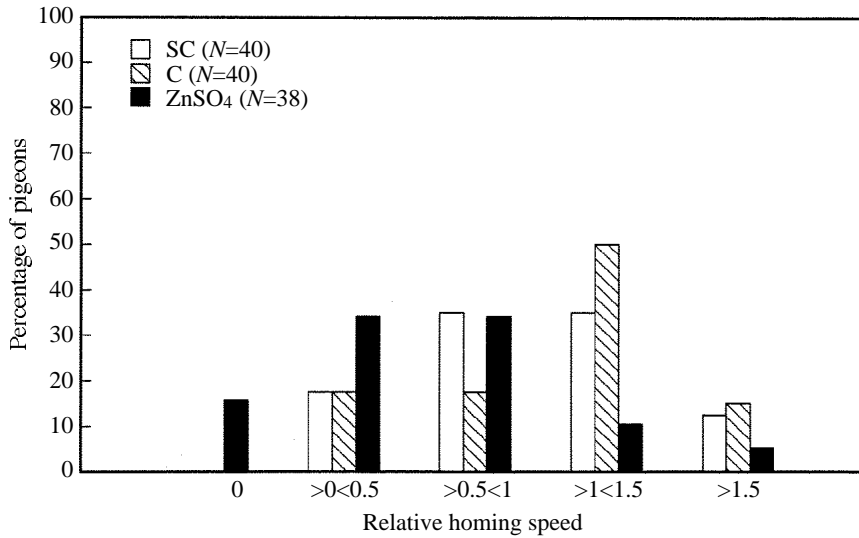
Fig. 2. Summary of relative homing speeds of the releases from familiar sites, distance I. For further details, see Materials and methods.

Table 3. *Initial orientation of temporarily anosmic pigeons in single releases from familiar sites, distance II (54–70km)*

Site	β	km	Group	α	a	N_R	N_B	km h^{-1}	N_h
Kol	11°	54.0	SC	345°	0.86***	12	9	15.2	11
			C	23°	0.51	13	10	23.0	11
			ZnSO$_4$	73°	0.47	10	8	0.1	5
Lei	185°	68.8	SC	192°	0.29	10	9	32.3	9
			C	178°	0.64*	10	3	27.7	7
			ZnSO$_4$	178°	0.46	9	9	0.1	3
Tom	275°	66.5	SC	325°	0.63*	11	8	0.9	9
			C	115°	0.31	12	10	4.6	8
			ZnSO$_4$	116°	0.73**	11	9	0.1	3
Dur	129°	69.3	SC	190°	0.16	12	9	0.9	10
			C	246°	0.59*	12	9	19.1	11
			ZnSO$_4$	39°	0.19	10	5	0.5	5

Kol, Kolbingen; Lei, Leingarten; Tom, Tomerdingen; Dur, Durmersheim.
For more details, see Table 1.

$P<0.001$). There was no difference between the two control groups (Mann–Whitney *U*-test: $z=0.77$, $P>0.05$).

For both distances, the vanishing bearings were pooled with respect to the mean of the super controls. No additional information could be gained.
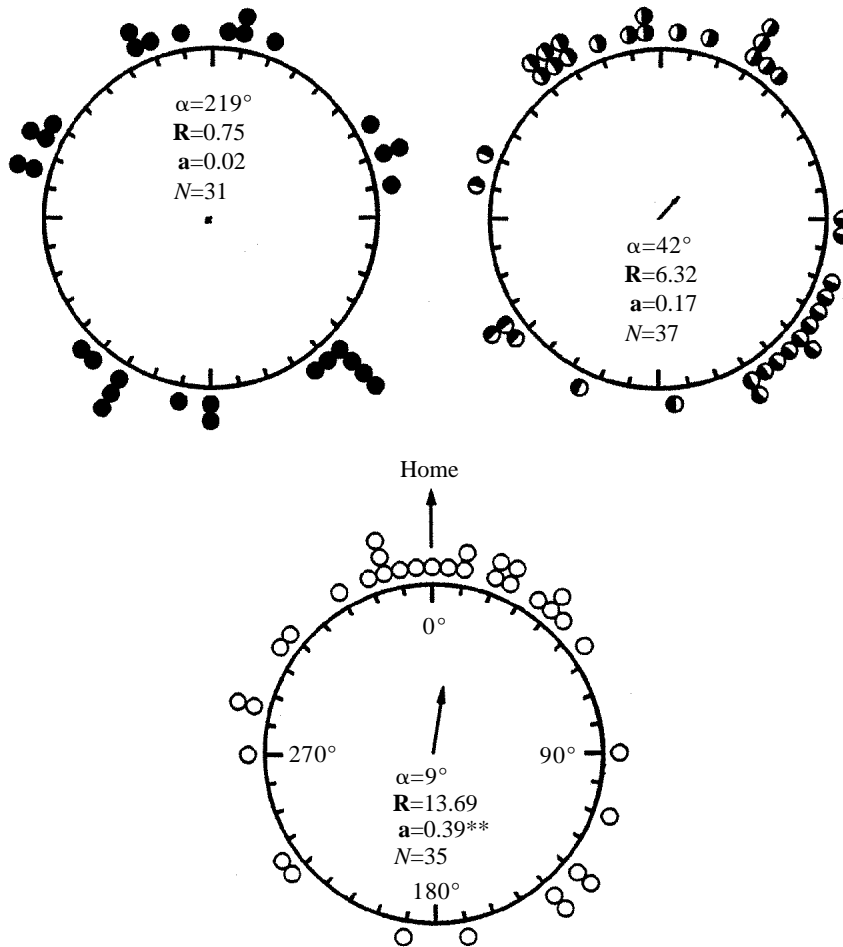
α=219°
R=0.75
a=0.02
N=31

α=42°
R=6.32
a=0.17
N=37

Home

0°

270° 90°

α=9°
R=13.69
a=0.39**
N=35

180°

Fig. 3. Initial orientation of ZnSO$_4$-pigeons (●), C-pigeons (◑) and SC-pigeons (○) summarised with respect to home (0°). Releases from familiar sites, distance II. For further details, see Fig. 1.

Releases of pigeons reared anosmically

The first time that pigeons were allowed to fly around the loft, 20 of 54 P-ZnSO$_4$-pigeons were lost (summarised for both years). After the entire training phase, only 32 P-ZnSO$_4$-pigeons remained for the critical releases.

The principal data for the single releases are summarised in Table 4. Fig. 5 shows the mean vectors for anosmic birds (P-ZnSO4, ST-ZnSO$_4$) and controls (PC, STC, SC). All three control groups and the P-ZnSO$_4$-pigeons showed significant directional preferences (Rayleigh test) when results were pooled with respect to home. The 95% confidence interval of the direction of the mean vector of all four groups included the home direction. The distribution of the pooled vanishing bearings of ST-ZnSO$_4$-pigeons did not differ from random. Table 5 compares the vanishing bearings of the different release groups by
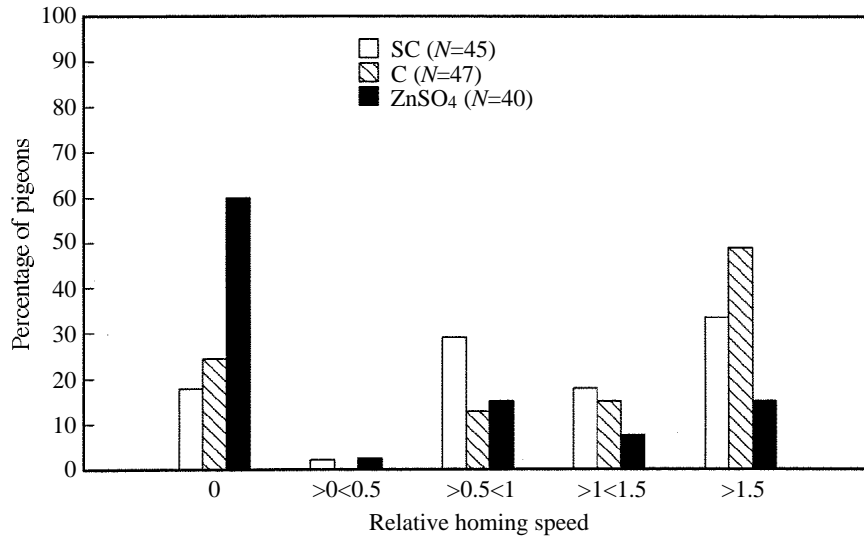
Fig. 4. Summary of relative homing speeds of the releases from familiar sites, distance II. For further details, see Materials and methods.

means of first-order (Watson-U^2 test) and second-order statistics (Hotelling test). The orientation of the permanently anosmic pigeons did not differ from that of SC- and PC-pigeons using both first- and second-order statistics. Although there was no difference between the ST-ZnSO$_4$-pigeons and the controls (Table 5), the results show a tendency towards significance. In addition, there were no differences either between P-ZnSO$_4$-pigeons and ST-ZnSO$_4$-pigeons or between any groups of controls.

The relative homing speeds are illustrated in Fig. 6, giving the combined releases pooled with respect to the median of super controls. Table 6 gives comparisons of the relative homing speeds between all groups using the Mann–Whitney U-test. ST-ZnSO$_4$-pigeons and P-ZnSO$_4$-pigeons homed more slowly than the controls. Although the homing speed of P-ZnSO$_4$-pigeons was lower than that of the ST-ZnSO$_4$-pigeons, there was no difference between the groups. Additionally, there was no difference between super controls (SC) and short-term controls (STC) but, compared with the permanent controls (PC), the homing speed of the STC birds was significantly reduced.

Discussion

Releases of temporarily anosmic pigeons from familiar sites

The anosmic ZnSO$_4$-pigeons released at familiar sites were capable of correct initial orientation only at shorter distances (distance I). In releases from distance II, the distribution of vanishing bearings of anosmic ZnSO$_4$-pigeons was more scattered than the distribution of vanishing bearings of controls. Homing ability, however, was drastically reduced in ZnSO$_4$-pigeons of both series. Compared with anosmic ZnSO$_4$-pigeons released from unfamiliar sites (Schlund, 1992), relative homing speeds of ZnSO$_4$-pigeons

Table 4. *Experiments with pigeons reared anosmically: initial orientation in single releases*

Site	β	km	Group	α	a	N_R	N_B	km h^{-1}	N_h
Fro	45°	15.8	SC	56°	0.88***	7	7	24.3	5
			STC	42°	0.72**	9	9	30.6	8
			PC	41°	0.85**	6	6	13.9	6
			ST-ZnSO$_4$	77°	0.42	7	7	6.4	6
			P-ZnSO$_4$	77°	0.66	8	6	0.1	1
Hec	23°	23.6	SC	43°	0.81***	9	7	0.1	4
			STC	66°	0.97***	7	5	0.7	6
			PC	63°	0.90***	9	8	0.1	2
			ST-ZnSO$_4$	150°	0.46	7	5	0.1	3
			P-ZnSO$_4$	81°	0.45	8	7	0.1	1
Ber	224°	22.8	SC	134°	0.24	10	8	0.9	7
			STC	129°	0.53	10	9	7.5	7
			PC	135°	0.38	12	9	0.8	9
			ST-ZnSO$_4$	99°	0.27	6	6	0.4	2
			P-ZnSO$_4$	326°	0.43	8	4	0.1	1
Ler	140°	15.4	SC	157°	0.59**	13	12	5.6	13
			STC	72°	0.57*	9	9	16.4	8
			PC	112°	0.52	11	10	0.9	9
			ST-ZnSO$_4$	37°	0.51	8	6	0.1	3
			P-ZnSO$_4$	123°	0.94	8	4	0.6	4

Sites: Fro, Frommenhausen; Hec, Hechingen; Ber, Bernhausen; Ler, Lerchenberg.

Groups: SC, super controls; STC, short-term controls; PC, permanent controls; ST-ZnSO$_4$, short-term experimentals; P-ZnSO$_4$, permanent experimentals.

For more details, see Table 1.

Table 5. *Initial orientation of anosmically reared pigeons by means of first-order and second-order statistics*

	STC (N=32)	PC (N=33)	ST-ZnSO$_4$ (N=24)	P-ZnSO$_4$ (N=21)
SC (N=34)	U^2=0.148 T^2=0.45	U^2=0.070 T^2=0.31	U^2=0.165 T^2=0.68	U^2=0.639 T^2=0.56
STC	– –	U^2=0.042 T^2=0.15	U^2=0.128 T^2=6.58	– –
PC	– –	– –	– –	U^2=0.182 T^2=0.92
ST-ZnSO$_4$	– –	– –	– –	U^2=0.138 T^2=6.16

The results are pooled with respect to home.

First-order statistics, Watson U^2-test; second-order statistics, Hotelling test; d.f.=2, 5.
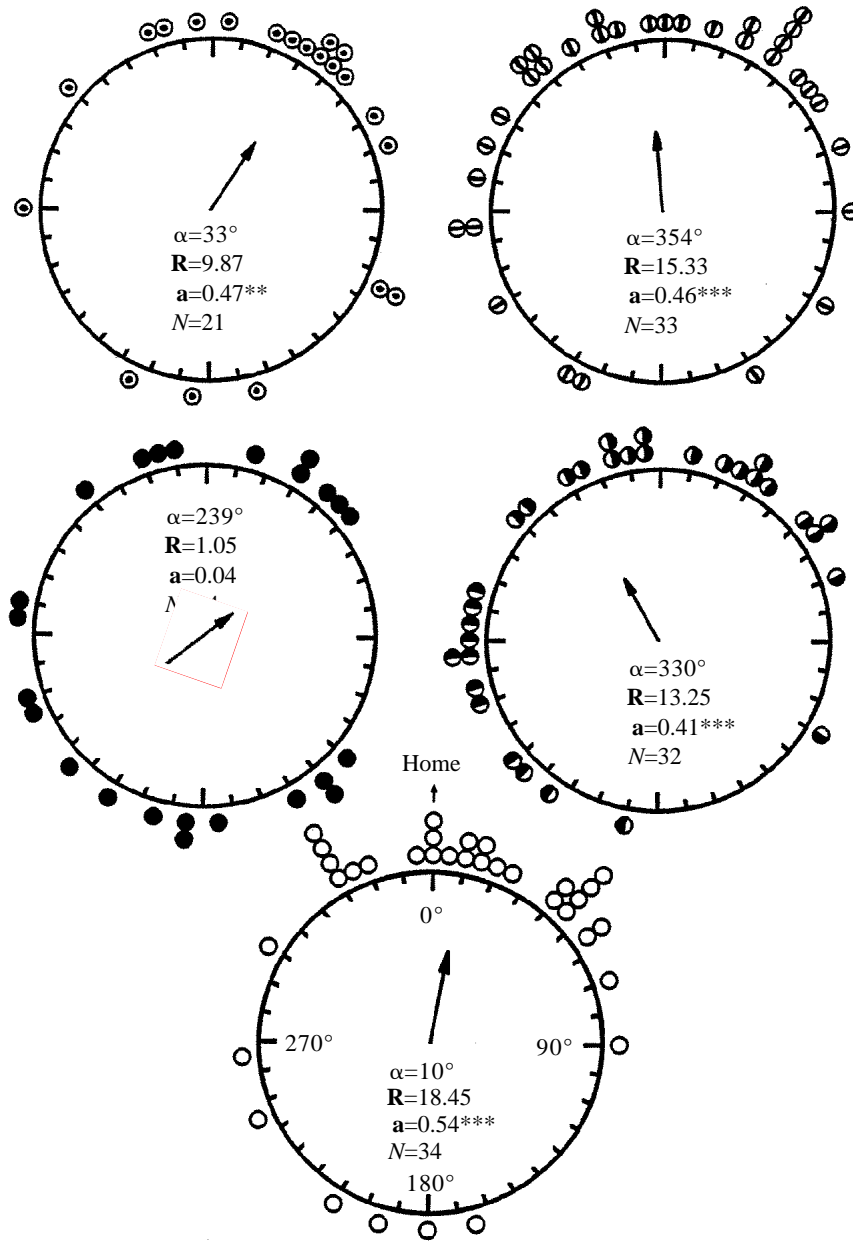
For further details, see Table 4.

Fig. 5. Initial orientation of P-ZnSO₄-pigeons (⊙), ST-ZnSO₄-pigeons (●), PC-pigeons (◍), STC-pigeons (◑) and SC-pigeons (○) summarised with respect to home (0˚). Releases with anosmically reared pigeons from unfamiliar sites. For more details, see Fig. 1.

from familiar sites at distances I and II were significantly faster (Mann–Whitney *U*-test; distance I: *z*=3.24, *P*<0.02; distance II: *z*=3.34, *P*<0.001). There was no significant difference between the relative homing speeds of control groups.
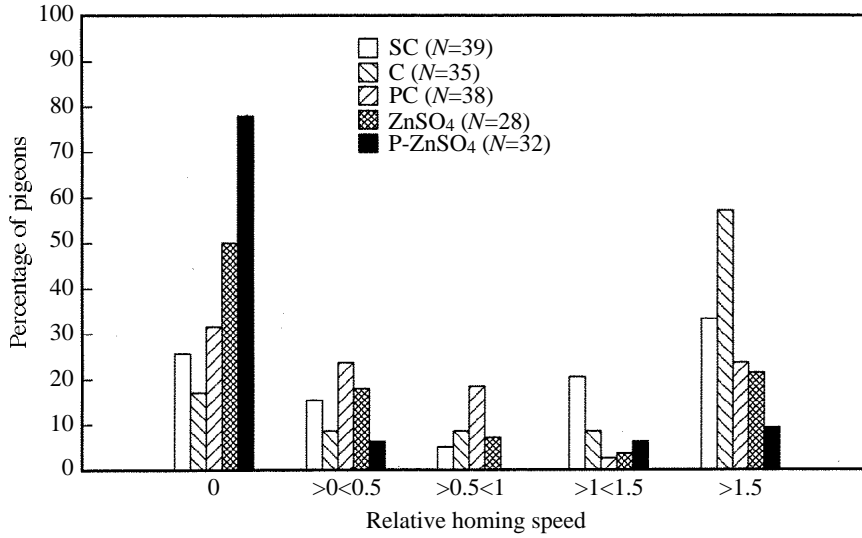
Fig. 6. Summary of relative homing speeds of the releases of anosmically reared pigeons from unfamiliar sites. For further details, see Materials and methods.

Table 6. *Comparison of relative homing speeds of the releases with pigeons reared anosmically*

	STC (N=39)	PC (N=35)	ST-ZnSO$_4$ (N=29)	P-ZnSO$_4$ (N=33)
SC (N=39)	z=1.178	z=1.440	z=2.411*	z=3.529***
STC	–	z=2.496*	z=3.048**	–
PC	–	–	–	z=2.651**
ST-ZnSO$_4$	–	–	–	z=1.130

The homing speed of each release was standardised on the median of super controls.
Mann–Whitney U-test: *P<0.05, **P<0.01, ***P<0.001.
For further details, see Table 4.

There is evidence, however, that familiarity with the release site can affect the initial orientation and the homing success of pigeons (Benvenuti *et al.* 1973; Hartwick *et al.* 1977; Kiepenheuer, 1982; Lednor and Walcott, 1988; Papi, 1986; Wallraff and Neumann, 1989; Grüter and Wiltschko, 1990; Braithwaite and Guilford, 1991). On the basis of their findings, Wallraff and Neumann fell back on the hypothesis that pigeons rely on two independent homing methods, olfactory navigation and non-olfactory pilotage. Griffin (1952, 1955) and Matthews (1955) considered orientation along familiar visual landmarks as the simplest homing strategy. According to Wallraff and Neumann, the use of visual landmarks should enable homing of anosmic pigeons over distances of 50km and more.

Our findings, however, argue against this hypothesis. In releases from distance II,

familiar landmarks did not determine the course of departure of anosmic pigeons and, in releases from both distances, they showed poor homing performances. Benvenuti and Fiaschi (1983) demonstrated that familiarity with the release site does not depend on visual input. Pigeons were homeward-oriented even when made anosmic and wearing frosted lenses. There is no evidence of pilotage using landmarks. At shorter distances, orientation could also be a navigational process.

Thus, the nature of the features which make a site 'familiar' remains unknown. This leads back to the idea of the navigational 'map' described by Wallraff (1974) and Wiltschko and Wiltschko (1982, 1987), which is hypothesised to be extrapolated beyond the area of immediate experience. By obtaining additional local experience, the pigeons can update their 'map'. Improving initial orientation and homing appears to depend on this modification of the birds' map as a result of previous familiarity with the area.

Our findings, however, indicate that familiarity with the release site has no effect on initial orientation in releases from distance II. This may be because the birds do not know the area well enough after only one previous flight. At the release sites located only 9–24km away (distance I), it seems possible that pigeons recognised a site known from former experience, for example during their training flights. So these birds were, in fact, more familiar with the general region around and beyond the test site than were birds released from distance II.

Releases of pigeons reared anosmically

The data obtained with permanently anosmic pigeons from releases in summer 1990 (Fro, Hec) and 1991 (Ber, Ler) are consistent in both years (see Table 4): the vanishing bearings of permanently anosmic pigeons and of all control groups showed a directional preference. The distribution of the pooled vanishing bearings of temporarily anosmic pigeons did not differ from random but, nevertheless, there was no significant difference in the mean vanishing directions between all groups of experimentals and controls (Table 4). This might be an effect of the small sample size of these pigeons because only the data for birds found to be still anosmic upon return to the loft were analysed statistically.

Papi *et al.* (1972, 1973) advanced a hypothesis of navigation based on an olfactory 'mosaic' map. According to this hypothesis, pigeons associate specific odours with the direction of the winds carrying these odours to the loft. Permanently anosmic birds, having been prevented from establishing such an olfactory 'map', should not be able to determine the home direction. Nevertheless, our pigeons were capable of doing so, in contrast to pigeons made anosmic by severing the olfactory nerve (Papi *et al.* 1989). It should be borne in mind that nerve section results in side effects (Wenzel and Rausch, 1977; Wenzel, 1982, 1983). In addition, if pooled with respect to the mean of super controls, the vanishing bearings of P-ZnSO4-pigeons showed a bimodal distribution (Fig. 7) that corresponds to an orientation along an axis. After doubling the angles, the resulting cluster is well oriented ($\alpha=6°$, $a=0.62$, $N=21$, $P<0.005$).

These results support the hypothesis of the multifactorial navigational system (Keeton, 1974), which is at present the most widely accepted. According to this hypothesis, the
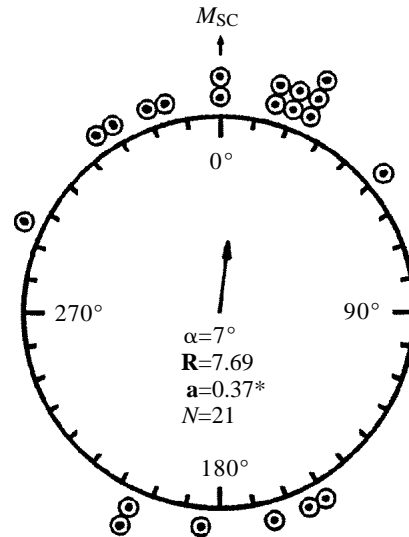
Fig. 7. Initial orientation of P-ZnSO$_4$-pigeons ($\odot$) pooled with respect to the mean of super controls (M$_{SC}$=0°).

pigeon can use different cues for navigation. The cluster analyses of Ganzhorn (1990) yielded a similar picture, in which pigeons in various regions were assumed to differ in strategies and in the factors they use to orient. In all probability, there is a certain hierarchy in choosing the navigational factors. If a type of olfactory information is missing during ontogeny, a 'map' will perhaps be established that depends on the available navigational cues but not on olfactory cues (Wiltschko and Wiltschko, 1985, 1987; Wiltschko *et al.* 1987). According to this theory, it is not surprising that short-term manipulations with ZnSO$_4$ caused disorientation. Releases of temporarily anosmic pigeons treated with xylocain showed similar results (Papi *et al.* 1989), although local anaesthetics have been shown to induce systemic effects (Schlund, 1990) and not to be very dependable (Wallraff, 1988*b*; Schlund, 1991*b*). The learned system of temporarily anosmic birds during rearing depends on a functioning olfactory system that was not available at the critical release.

Homing speed and homing success were drastically reduced in permanently and temporarily anosmic pigeons. The decrease in homing performance of the permanently anosmic pigeons is in contrast to their good initial orientation. This phenomenon is also described by Keeton *et al.* (1977). Birds obviously 'knew' the home direction but were not able to complete the route home. In the P-ZnSO$_4$ group some of the birds were hunted down by goshawks and many more landed, perhaps because of a reduced motivation for homing rather than because of orientation deficits. Unfortunately, we never received reports about permanently anosmic ZnSO$_4$-pigeons that did not home. Thus, there is no evidence of the distances they had covered. This discrepancy supports the idea that the navigational strategies are based on two

different mechanisms: one for initial orientation at the release site and another for the rest of the homing process.

Furthermore, little is known about the possible side effects of treatment of pigeons with $ZnSO_4$. In mice, treatment with $ZnSO_4$ caused weight loss of the bulbus olfactorius (Harding *et al.* 1978). The action of $ZnSO_4$ in the olfactory system and even adjacent parts of the brain of pigeons could have effects on learning, motivation, circadian rhythms or magnetic perception (discussed in detail by Schlund, 1992).

It should be emphasised that the results, although suggestive, do not prove olfactory navigation. The experiments do not provide evidence that odorous substances are involved in homing or that the experimental interference interacts exclusively with olfactory functions.

References

ALBERTS, J. R. (1974). Producing and interpreting experimental olfactory deficits. *Physiol. Behav.* **12**, 657–670.

BATSCHELET, E. (1981). *Circular Statsitics in Biology.* London, New York: Academic Press.

BENVENUTI, S. AND FIASCHI, V. (1983). Pigeon homing: combined effect of olfactory deprivation and visual impairment. *Comp. Biochem. Physiol.* A **76**, 719–723.

BENVENUTI, S., FIASCHI, V., FIORE, L. AND PAPI, F. (1973). Homing performances of inexperienced and directionally trained pigeons subjected to olfactory nerve section. *J. comp. Physiol.* **83**, 81–92.

BRAITHWAITE, V. A. AND GUILFORD, T. (1991). Viewing familiar landscapes affects pigeon homing. *Proc. R. Soc. Lond. B* **245**, 183–186.

CABRERA, J., SCHMIDT-KOENIG, K. AND WATSON, G. S.(1991). The statistical analysis of circular data. In *Perspectives in Ethology*, vol. 9 (ed. P. P. G. Bateson and P. H. Klopfer), pp. 285–306. New York: Plenum Press.

CANCALON, P. (1982). Degeneration and regeneration of olfactory cells induced by $ZnSO_4$ and other chemicals. *Tissue & Cell* **14**, 713–733.

GANZHORN, J. U.(1990). Towards the map of the homing pigeon? *Anim. Behav.* **40**, 65–78.

GRIFFIN, D. R. (1952). Bird navigation. *Biol. Rev.* **27**, 359–400.

GRIFFIN, D. R. (1955). Bird navigation. In *Recent Studies in Avian Biology* (ed. A. Wolfson), pp. 154–197. Urbana: University of Illinois Press.

GRÜTER, M. AND WILTSCHKO, R. (1990). Pigeon homing: the effect of local experience on initial orientation and homing success. *Ethology* **84**, 239–255.

HARDING, J. W., GETCHELL, T. V. AND MARGOLIS, F. L. (1978). Denervation of the primary olfactory pathway in mice. Long term effect of intranasal $ZnSO_4$ irrigation on behavior, biochemistry and morphology. *Brain Res.* **140**, 271–285.

HARTWICK, R. F., FOA, A. AND PAPI, F. (1977). The effect of olfactory deprivation by nasal tubes upon homing behaviour in pigeons. *Behav. Ecol. Sociobiol.* **2**, 81–89.

KEETON, W. T. (1974). The orientation and navigational basis of homing in birds. *Adv. Study Behav.* **5**, 47–132.

KEETON, W. T., KREITHEN, M. L. AND HERMAYER, K. L. (1977). Orientation by pigeons deprived of olfactory by nasal tubes. *J. comp. Physiol.* **114**, 289–299.

KIEPENHEUER, J.(1982). The effect of magnetic anomalies on the homing behaviour of pigeons. In *Avian Navigation* (ed. F. Papi and H. G. Wallraff), pp. 120–128. Berlin, Heidelberg: Springer Verlag.

Lednor, A. J. and Walcott, C. (1988). Orientation of homing pigeons at magnetic anomalies: the effect of experience. *Behav. Ecol. Sociobiol.* **22**, 3–8.

Margolis, F. L., Roberts, N., Ferriero, D. and Feldman, J. (1974). Denervation in the primary olfactory pathway of mice: biochemical and morphological effects. *Brain Res.* **81**, 469–483.

Matthews, G. V. T.(1955). *Bird Navigation.* Cambridge: Cambridge University Press.

Matulionis, D. H.(1975). Ultrastructural study of mouse olfactory epithelium following destruction by $ZnSO_4$ and its subsequent regeneration. *Am. J. Anat.* **142**, 67–90.

Matulionis, D. H. (1976). Light and electron microscopic study of the degeneration and early regeneration of olfactory epithelium in the mouse. *Am. J. Anat.* **145**, 79–100.

Papi, F. (1986). Pigeon navigation: solved problems and open questions. *Monit. zool. ital. (N.S.)* **20**, 471–517.

Papi, F.(1990). Olfactory navigation in birds. *Experientia* **46**, 352–362.

Papi, F., Fiaschi, V., Benvenuti, S. and Baldaccini, N. E. (1973). Pigeon homing: outward journey detours influence the initial orientation. *Monit. zool. ital. (N.S.)* **7**, 129–133.

Papi, F., Fiore, L., Fiaschi, V. and Benvenuti, S.(1972). Olfaction and homing in pigeons. *Monit. zool. ital. (N.S.)* **6**, 85–95.

Papi, F., Gagliardo, A., Fiaschi, F. and dall'Antonia, P. (1989). Pigeon homing: does early experience determine what cues are used to navigate? *Ethology* **82**, 208–215.

SAS/STAT (1987). *Guide for Personal Computers.* Cary.

Schlund, W. (1990). Auswirkungen der Lokalanästhesie der Riechschleimhaut auf Sinnesleistungen bei Brieftauben (*Columba livia*). *J. Orn.* **131**, 325–332.

Schlund, W. (1991*a*). Auswirkungen von $ZnSO_4$ auf die olfaktorische Wahrnehmung, die Anfangsorientierung und den Heimkehrerfolg von Brieftauben (*Columba livia*). Diplomarbeit, Fakultät für Biologie, Universität Tübingen.

Schlund, W. (1991*b*). Auswirkungen der Lokalanästhesie der Riechschleimhaut auf Sinnesleistungen bei Brieftauben (*Columba livia*). *Verh. dt. zool. Ges.* **84**, 359.

Schlund, W.(1992). Intra-nasal zinc sulphate irrigation in pigeons: effects on olfactory capabilities and homing. *J. exp. Biol.* **164**, 171–187.

Schmidt-Koenig, K.(1979). *Avian Orientation and Navigation.* London: Academic Press.

Schmidt-Koenig, K.(1985). Hypothesen und Argumente zum Navigationsvermögen der Vögel. *J. Orn.* **126**, 237–252.

Schmidt-Koenig, K. (1987). Bird navigation: Has olfactory orientation solved the problems? *Q. Rev. Biol.* **62**, 31–47.

Siegel, S.(1956). *Nonparametric Statistics.* New York, Toronto, London: McGraw-Hill.

Smith, C. G. (1951). Regeneration of sensory olfactory epithelium and nerves in adult frogs. *Anat. Rec.* **109**, 661–671.

Waldvogel, J. A.(1989). Olfactory orientation by birds. *Current Orn.* **6**, 269–321.

Wallraff, H. G. (1974). *Das Navigationssystem der Vögel. Schriftenreihe 'Kybernetik'.* München, Wien: R. Oldenbourg Verlag.

Wallraff, H. G. (1988*a*). Navigation mit Duftkarte und Sonnenkompaß: das Heimfindevermögen der Brieftauben. *Naturwissenschaften* **75**, 380–392.

Wallraff, H. G.(1988*b*). Olfactory deprivation in pigeons: Examination of methods applied in homing experiments. *Comp. Biochem. Physiol.* **89**A, 621–629.

Wallraff, H. G. and Neumann, M. F. (1989). Contribution of olfactory navigation and non-olfactory pilotage to pigeon homing. *Behav. Ecol. Sociobiol.* **25**, 293–302.

Wenzel, B. M. (1982). Functional status and credibility of avian olfaction. In *Avian Navigation* (ed. F. Papi and H.G. Wallraff), pp. 352–361. Berlin: Springer Verlag.

Wenzel, B. M. (1983). Chemical senses. In *Physiology and Behaviour of the Pigeon* (ed. M. Abs), pp. 149–167. London: Academic Press.

Wenzel, B. M. and Rausch, L. J.(1977). Does the olfactory system modulate affective behaviour in the pigeon? *Ann. N.Y. Acad. Sci.* **290**, 314–330.

Wiltschko, R. and Wiltschko, W. (1985). Pigeon homing: change in navigational strategy during ontogeny. *Anim. Behav.* **33**, 583–590.

Wiltschko, W. and Wiltschko, R. (1978). A theoretical model for migratory orientation and homing in birds. *Oikos* **30**, 177–187.

Wiltschko, W. and Wiltschko, R. (1982). The role of outward journey information in the orientation

of homing pigeons. In *Avian Navigation* (ed. F. Papi and H. G. Wallraff), pp. 239–252. Berlin: Springer Verlag.

WILTSCHKO, W. AND WILTSCHKO, R. (1987). Cognitive maps and navigation in homing pigeons. In *Cognitive Processes and Spatial Orientation in Animals and Man* (ed. P. Ellen and C. Thinus-Blanc), pp. 201–216. Dordrecht: M. Nijhoff.

WILTSCHKO, W., WILTSCHKO, R. AND WALCOTT, C.(1987). Pigeon homing: Different effects of olfactory deprivation in different countries. *Behav. Ecol. Sociobiol.* **21**, 333–342.