

Electromagnetic 0.1–100 kHz noise does not disrupt orientation in a night-migrating songbird implying a spin-coherence lifetime of less than 10 μ s

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Abstract

According to the currently prevailing theory, the magnetic compass sense in night-migrating birds relies on a light-dependent radical-pair-based mechanism. It has been shown that radio waves at MHz frequencies disrupt magnetic orientation in migratory birds, providing evidence for a quantum-mechanical origin of the magnetic compass. Still, many crucial properties, e.g. the lifetime of the proposed magnetically sensitive radical pair, remain unknown. The current study aims to estimate the spin coherence time of the radical pair, based on the behavioural responses of migratory birds to broadband electromagnetic fields covering the frequency band 0.1–100 kHz. A finding that the birds were unable to use their magnetic compass under these conditions would imply surprisingly long-lived ($> 10 \mu$ s) spin coherence. However, we observed no effect of 0.1–100 kHz radiofrequency fields on the orientation of night-migratory Eurasian blackcaps (*Sylvia atricapilla*). This suggests that the lifetime of the spin coherence involved in magnetoreception is shorter than the period of the highest frequency radiofrequency fields used in this experiment (i.e. $\sim 10 \mu$ s). This result, in combination with an earlier study showing that 20–450 kHz electromagnetic fields disrupt magnetic compass orientation, suggests that the spin-coherence lifetime of the magnetically sensitive radical pair is in the range 2–10 μ s.

Keywords: magnetoreception, radical pair mechanism, radiofrequency field, broadband noise

1. Introduction

Night-migratory songbirds have a magnetic compass that they use for migratory orientation [1-3]. The compass is light-dependent [4] (for a review see [5]) and the information it provides is processed in a specific part of the birds' visual system [6-9]. Furthermore, the sensor responds to changes in the inclination of the Earth's magnetic field (the angle between the field vector and the horizontal direction), but not to its polarity [1]. These observations have led to the proposal that a light-activated, radical-pair based chemical reaction forms the basis for the avian magnetic compass [3, 10-13].

According to this model, photoexcitation of receptor molecules in the birds' retinas can trigger an electron transfer pathway that results in the formation of radical pairs [13]. The magnetic moments of the unpaired electron spins in the two radicals interact with each other (exchange and dipolar interactions) [14] and with the magnetic nuclei in the radicals (hyperfine interactions) [15]. The latter cause interconversion of the two allowed quantum states of the radical pair: a singlet (antiparallel electron spins) and a triplet (parallel electron spins). The time-dependent proportion of radical pairs in each of the two states is influenced by the strength and direction of an external magnetic field. In this way, the spin state of radical pairs formed by this photochemical reaction can depend on the orientation of the receptor molecule with respect to the Earth's magnetic field. For this effect to form the basis of a magnetic compass sensor, at least one of the reaction pathways of the radical pair must be spin-selective, i.e. it must proceed with different rate constants for the singlet and triplet states. The consequence is that the yield of one of the reaction products depends on the extent and timing of the singlet-triplet interconversion and therefore carries information on the direction of the external magnetic field (for a review see [13]).

In animals, the only candidate magnetoreceptor molecules known to form long-lived radical pairs on photoexcitation are the cryptochromes [16, 17]. These proteins are expressed in birds' retinas [18-21] and are activated by blue light [16, 22] (also see [23] for a plant cryptochrome). Of the four known avian cryptochromes, Cry4 seems the most likely to be a magnetoreceptor: it has been found in the outer segments of the double-cone photoreceptor cells, it binds FAD (flavin adenine dinucleotide) under physiological conditions (without

which there can be no radical pairs formed), and it does not show the circadian expression pattern characteristic of the other avian cryptochromes [21, 24-27].

So far the most direct *in vivo* evidence for radical-pair-based magnetoreception comes from behavioural experiments showing the effect of oscillating electromagnetic fields on the orientation of migratory birds. Radiofrequency (RF) fields can alter the spin state of the unpaired electron spins in a radical pair [28-30] if the frequency of the oscillating field matches one of the frequencies of singlet-triplet interconversion. This resonant effect could theoretically override the effect of the Earth's magnetic field and so prevent the bird from using its magnetic compass.

The disruptive effect of oscillating electromagnetic fields has been reported in several studies, both for monochromatic [29, 31-33] and broadband fields [29, 34, 35]. However, the reported all-or-nothing effect of monochromatic fields on magnetic orientation was recently challenged, when Schwarze et al. (2016) [35] and Malkemper et al. (2015) [36] did not observe an expected effect of the Larmor frequency field. The possible reasons behind these discrepancies in the results are debatable and could include differences in the sample sizes, as well as equipment and measurement quality [3, 13, 30, 37].

The effect of broadband electromagnetic noise on magnetic orientation behaviour has been reported in several independent studies.

1) Ritz et al. (2004) [29] showed that European robins were disoriented when exposed to oscillating fields in the range 0.1–10 MHz. 2) Engels et al. (2014) [34] and Schwarze et al. (2016) [35] observed the same disruptive effect on the orientation of robins using broadband electromagnetic noise in the low MHz range with a “maximal, total magnetic field intensity” as low as 32 nT (see Table 1 for a detailed description).

3) Malkemper et al. (2015) [36] exposed wood mice (*Apodemus sylvaticus*) to a 0.9–5.0 MHz frequency sweep and found effects on their nest-building behaviour.

Sensitivity to RF fields can be used as a general diagnostic test for the radical pair mechanism [28] and also to estimate some crucial properties of the radical pair, e.g. its spin-coherence lifetime which is important in determining its performance as a magnetoreceptor. The longer the coherence lasts, the weaker is the field to which the radical pair is sensitive [13, 38]. An oscillating electromagnetic field can only have a resonant effect on the spin dynamics of the radical pair if the period of its oscillation is smaller than the lifetime of the spin coherence. If this is not the case, the time-dependent field hardly changes during the

lifetime of the coherence and would therefore be expected to have no more of an effect on the operation of the sensor than an additional static field of intensity ca. 1 nT.

There have been several attempts to infer the coherence lifetime from the intensity of the RF fields found to disable the bird's compass sense. These estimates have generally led to implausibly long spin relaxation times. Kavokin (2009) [39] concluded that the disruptive effect of 0.1–10 MHz RF fields with average amplitude of 85 nT (reported by Ritz et al. 2004 [29]) could only be explained if the longitudinal relaxation time of the electron spins were ~ 1 s. Using a different approach, Kattnig et al. (2016) [40] arrived at a much smaller, but still unrealistically long, decoherence time of 600 μ s based on the disruptive effect of a 10 nT monochromatic field oscillating at the Larmor frequency. The same authors calculated spin relaxation rates for radical pairs in cryptochrome using all-atom molecular dynamics simulations, density functional theory for the hyperfine tensors, and Redfield relaxation theory. Their conclusion was that the spin-coherence lifetime could be in the region of 1 μ s [40] (Kattnig et al. 2016). This value is close to optimum for sensing the Earth's magnetic field [11, 40–42] (but see [38]), but cannot explain the sensitivity of the magnetic compass to the extremely weak electromagnetic fields used in the studies that have reported RF-induced disorientation (e.g. [29, 31, 33–35]).

An approximate lower limit on the spin-coherence lifetime can be derived from the frequency range of the electromagnetic fields that have been found to affect birds' ability to use their magnetic compass. In the study by Engels et al. (2014) [34], European robins were disoriented by a 20–450 kHz broadband electromagnetic field. Since 450 kHz has a period of 2.2 μ s, this suggests that the spin coherence persists for at least 2.2 μ s.

The aim of the present study was to estimate the upper limit of the spin-coherence lifetime of magnetosensitive radical pairs in a night-migratory songbird. To this end we exposed Eurasian blackcaps (*Sylvia atricapilla*) to electromagnetic white noise in the range 0.1–100 kHz and looked for an effect on their magnetic compass orientation. The expectation is that if the spin-coherence lifetime is longer than the 10 μ s period of the 100 kHz component, the birds would be disoriented. If the birds could still orient magnetically in the presence of 0.1–100 kHz noise, the conclusion would be that the spin coherence probably persists no longer than about 10 μ s.

2. Methods

Experiments were conducted at the University of Oldenburg, Germany, in the Springs of 2017 and 2018. Blackcaps were wild-caught, housed in on-site aviaries in a windowless room with a light regime identical to that of the local photoperiod, and provided with food and water *ad libitum*.

2.1. Testing Location

Experiments took place in a custom-built laboratory constructed entirely of non-magnetic materials (full description in [35]), in an aluminium-shielded chamber constructed as a Faraday cage that allowed undistorted penetration of static magnetic fields and attenuation of fluctuating magnetic fields in the range 10 kHz–10 GHz by a factor $\geq 10^5$ [35]. The experimental chamber was grounded via a single electrode loop integrated into the concrete base of the laboratory, while all electrical equipment involved in generating the radiofrequency fields was grounded by means of a separate 8m-deep grounding rod.

2.2. Generation and Measurement of Static Magnetic Fields

Static magnetic fields were generated by a double-wrapped, 3-axis Merritt 4-coil system [35, 43]. The coils were ca. 2 m $\times$ 2 m, and experiments took place in the centre of the coils where the field homogeneity was $\geq 99\%$. Each of the three sets of four coils was driven by a separate constant-current power supply (BOP 50-2M or BOP 50-4M, Kepco Inc., Flushing, NY, USA). The local and rotated static magnetic fields were recorded using a flux-gate magnetometer (FVM-400, Meda Inc.), with measurements taken daily in the centre and at alternating sets of corners of the experimental space. All behavioural tests were performed in two magnetic field conditions: a normal magnetic field (NMF) and a changed magnetic field (CMF) where the horizontal component of the field had been rotated 120° anticlockwise. In the NMF condition, identical antiparallel currents passed through duplicate coil windings so that the birds experienced only the local geomagnetic field. In the CMF condition the currents flowed in parallel so that the fields produced by the two windings added instead of cancelling. The measured NMF field averaged over all days on which tests were performed was: inclination $67.52 \pm 0.22^\circ$ (mean $\pm$ standard deviation, SD) and intensity $48,022 \pm 187$ nT. For CMF the mean field was: inclination $67.56 \pm 0.16^\circ$, horizontal direction $-120.2 \pm 0.9^\circ$ and intensity $48,014 \pm 186$ nT.

2.3. Generation and Measurement of Time-dependent Electromagnetic fields

Time-dependent electromagnetic fields were generated using a signal generator (Wandel & Goltermann RG1, Germany) in which the broadband noise was produced by the in-built resistor R101, wideband-amplified by transistors T101 to T108 and then filtered using two external filter boxes (Krohn-Hite 3202, Germany). Two custom-built flat cable loops (outside dimensions 2.14 m × 2.14 m) were mounted horizontally around the Merritt coil system to deliver the time-dependent magnetic fields. Birds were subjected to two experimental conditions: “RF-on” (broadband noise in the frequency range 0.1–100 kHz) (Fig. 1a, b) and “RF-off” (signal generator running but with internal amplifier set to zero gain) (Fig. 1c, d).

The magnetic component of the field was measured prior to each behavioural test using a calibrated passive loop antenna (ETS Lindgren, Model 6511, 20 Hz to 5 MHz, Germany) and an active loop antenna (Schwarzbeck Mess-Elektronik, HFS 1546, 150 kHz to 400 MHz). The signal generator was fine-tuned to the correct frequency range using a signal analyser (Rohde and Schwarz, FSV 3 Signal and Spectrum Analyzer, 10 Hz–3.6 GHz, Germany). The electric component of the field was measured with a calibrated active bi-conical antenna (Schwarzbeck Mess-Elektronik, EFS 9218, 9 kHz–300 MHz, Germany). Signals were measured with a resolution bandwidth of 100 Hz for the stimulus (0.1–100 kHz) (Fig. 1, magnetic fields presented as pT/√Hz) and 10 kHz for the extended range measurements (500 kHz–100 MHz) (Fig. 2, magnetic fields presented as pT/√Hz). The extended measurements up to 100 MHz were performed to demonstrate the absence of non-specific electromagnetic noise at higher frequencies. We recorded the *maxhold* signal per square-root-bandwidth with the “Maxpeak” detector (in keeping with [34]) and the *average* signal per square-root-bandwidth with the “RMS” detector. The measuring antennas were placed in the centre of the coils and recorded the electric and magnetic components averaged over most of the area inside the coils.

The biophysical mechanism of the magnetic compass in Eurasian blackcaps is almost certainly the same as in other night-migratory songbirds such as European robins (*Erithacus rubecula*). On the assumption that blackcaps and robins have similar sensitivity to RF fields, we used electromagnetic noise of intensity comparable to that found by Engels et al. (2014) [34] to disrupt the magnetic orientation of robins (see Table 1).

Using a spectrum analyser, Engels et al. [34] measured their noise-modulated radiofrequency fields in *max hold* mode, i.e. the maximum magnetic noise intensity (in Tesla) at each frequency over a period of 1 hr. We denote such measurements with a superscript

“max”. We have used the same equipment to measure the radiofrequency noise in both *max hold* and *average* modes, the latter being the root-mean-square intensity (in Tesla) at each frequency during a 1 hr period. For white noise, which has a Gaussian distribution, the *max hold* intensity is ca. 10 dB (i.e. a factor of $\sqrt{10} \approx 3.16$) larger than the *average* intensity.

In all their experiments, Engels et al. measured and quoted radiofrequency noise intensities in Tesla using a resolution bandwidth, $BW = 10$ kHz. Because of the narrower range of frequencies employed in our study, we used a smaller resolution bandwidth, $BW = 100$ Hz. To allow the two measurements to be compared, the noise intensities measured by the spectrum analyser (in Tesla) should be divided by $\sqrt{BW}$ to obtain noise densities (in $\text{T Hz}^{-1/2}$) that are independent of the resolution bandwidth of the measurement. To distinguish these two quantities, we use upper and lower case symbols (B and b) for magnetic noise intensity and magnetic noise density, respectively, with the numerical values related by $b = B / \sqrt{BW}$.

We use two measures of the overall radiofrequency magnetic field in a frequency band $\Delta f = f_{\text{max}} - f_{\text{min}}$. Not enough is known about the operation of the magnetoreceptor to say which is more relevant. The first is the mean noise density,

$$\bar{b} = \frac{1}{N} \sum_{j=1}^N b_j, \quad (1)$$

for *average* measurements, and similarly for *max hold* measurements with $\bar{b}$ and b_j replaced by $\bar{b}^{\text{max}}$ and b_j^{max} respectively. The sum runs over the N equally spaced frequencies in the frequency band, Δf . Note that the values of the “maximal, total magnetic field intensity”, B_{total} previously given in Engels et al. can be converted to $\bar{b}^{\text{max}}$ using

$$\bar{b}^{\text{max}} = B_{\text{total}} \frac{\sqrt{BW}}{\Delta f}. \quad (2)$$

The other measure is the root-mean-square noise intensity, defined as

$$B_{\text{rms}} = \sqrt{\Delta f} \sqrt{\frac{1}{N} \sum_{j=1}^N b_j^2} \quad (3)$$

for *average* measurements, and similarly for *max hold* measurements with B_{rms} and b_j replaced by $B_{\text{rms}}^{\text{max}}$ and b_j^{max} respectively. B_{rms} , determined in the spectral domain, Eq. (3), is identical to the root-mean-square amplitude in the time domain, and has a clear physical

meaning in terms of the power density, which is proportional to B_{rms}^2 . Values of $\bar{b}^{\text{max}}$, $\bar{b}$, $B_{\text{rms}}^{\text{max}}$, and B_{rms} for the experiments of Engels et al. (European robins) and our own (Eurasian blackcaps) are shown in columns 3-6 of Table 1. Also included in Table 1 are values of the B_{total} parameter used by Engels et al.

Engels et al. (Table 1) found that robins were disoriented by radiofrequency noise for $\bar{b}^{\text{max}}$ between 1.4 and 30 pT Hz^{-1/2}, $B_{\text{rms}}^{\text{max}}$ between 2.3 and 23 nT, and B_{total} between 33 and 280 nT. In our “RF-on” condition, the corresponding values were $\bar{b}^{\text{max}} = 23 \text{ pT Hz}^{-1/2}$, $B_{\text{rms}}^{\text{max}} = 8.0 \text{ nT}$, and $B_{\text{total}} = 230 \text{ nT}$. By all three measures, the RF fields we have used are similar strength to those that caused disorientation in European robins in the Engels et al. study.

frequency band / kHz	species	$\bar{b}^{\text{max}}$ / pT Hz ^{-1/2}	$\bar{b}$ / pT Hz ^{-1/2}	$B_{\text{rms}}^{\text{max}}$ / nT	B_{rms} / nT	B_{total} / nT	orientation	source
10 – 5000	ER	0.07	~0.02	1.18	~0.37	3.30	+	[34], Fig. 4f, blue trace
10 – 5000	ER	5.58	~1.8	23.2	~7.35	279	–	[34], Fig. 4f, red trace
20 – 450	ER	30.1	~9.5	23.1	~7.29	129	–	[34], Fig. 4f, green trace
600 – 3000	ER	1.35	~0.43	2.30	~0.73	32.5	–	[34], Fig. 4f, black trace
0.1 – 100	BC	23.3	6.6	7.98	2.26	233	+	current study, Fig. 1a, RF-on
0.1 – 100	BC	0.92	0.32	1.29	0.51	9.17	+	current study, Fig. 1c, RF-off

Table 1. Comparison of different measures of the overall intensity of the broadband electromagnetic noise used here and in Engels et al. Species: ER – European robin, BC – Eurasian blackcap. $\bar{b}^{\text{max}}$, $\bar{b}$, $B_{\text{rms}}^{\text{max}}$ and B_{rms} are defined by Eqs(1) and (3). For the experiments of Engels et al., where measurements were done only in the *max hold* mode, the *average* mode values, $\bar{b}$ and B_{rms} , were estimated by dividing $\bar{b}^{\text{max}}$ and $B_{\text{rms}}^{\text{max}}$, respectively, by $\sqrt{10} \approx 3.16$ (i.e. by assuming white noise). Orientation: whether the birds were found to be oriented (+) or disoriented (–) in the behavioural experiments.

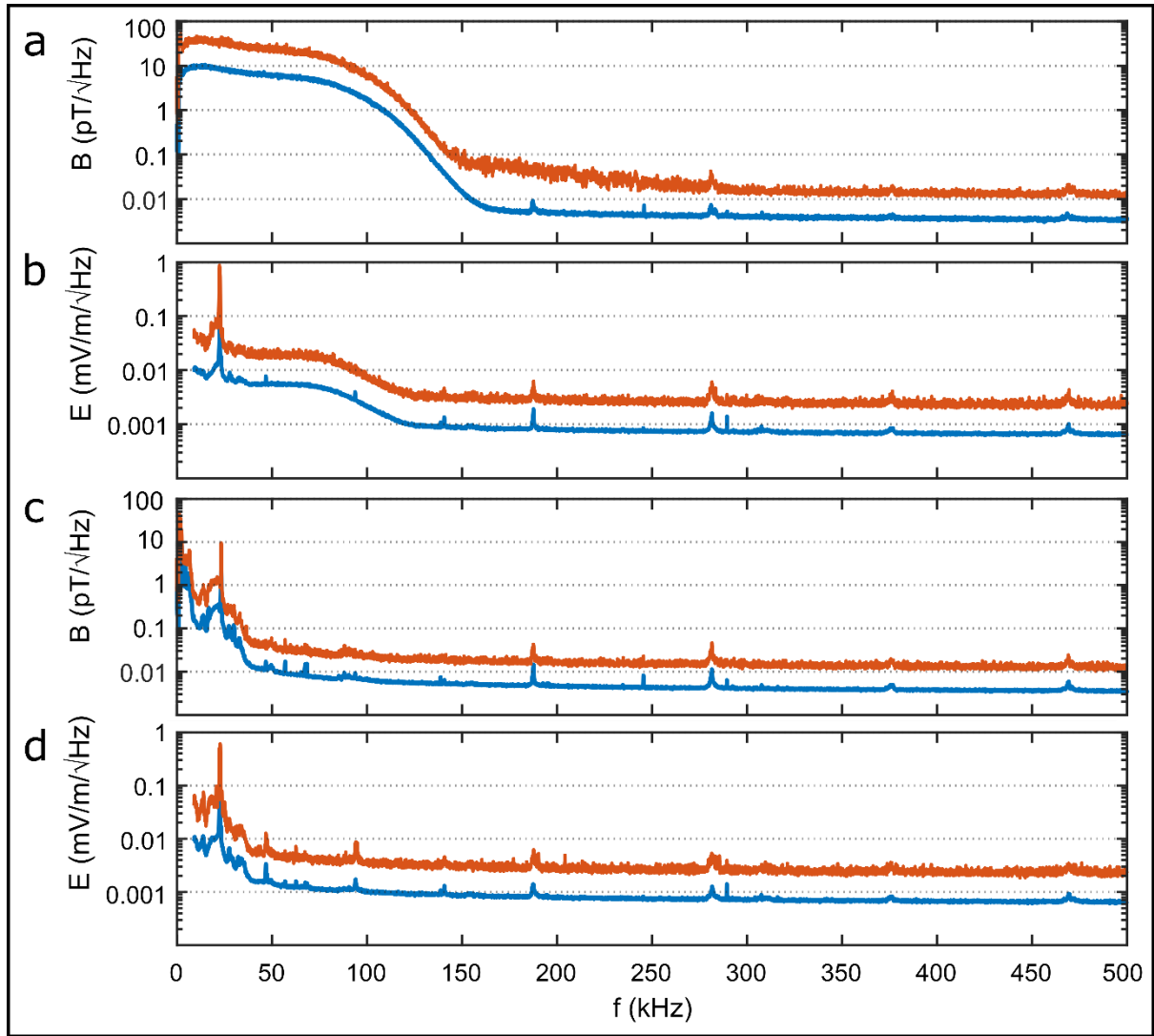


Figure 1

Measurements of the magnetic and electric components of the 0.1–100 kHz RF fields to which birds were exposed. (a) Magnetic component of the “RF on” condition; (b) Electric component of the “RF on” condition; (c) Magnetic component in the “RF off” condition; (d) Electric component in the “RF off” condition. Blue: signals measured in the “average” mode. Red: signals measured in the “maxhold” mode.

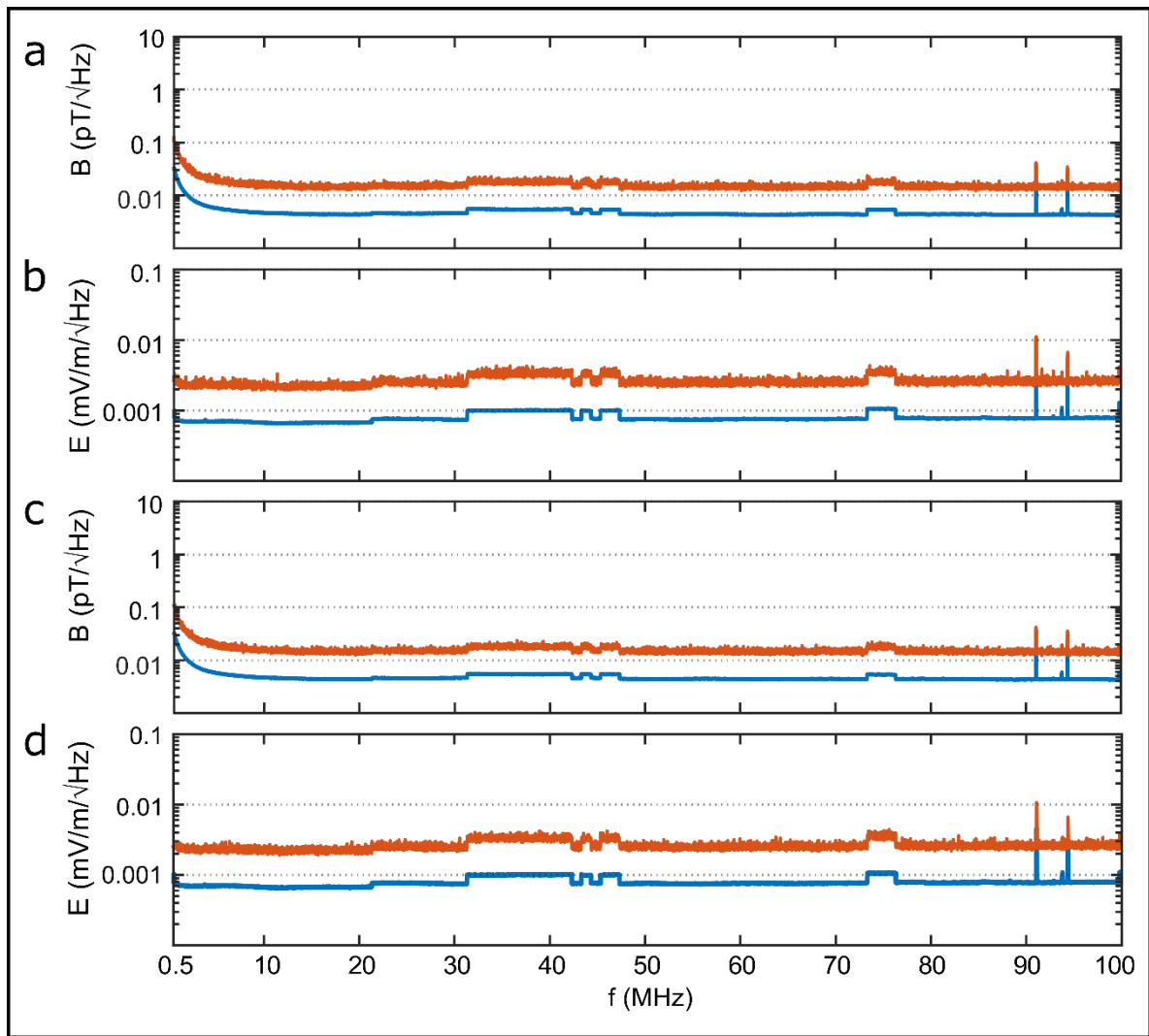


Figure 2

Extended (500 kHz–100 MHz) measurements of the magnetic and electric components of the electromagnetic fields to which birds were exposed. Note that there were no differences in this frequency range between control “RF off” and experimental “RF on” conditions. The increase in the measured spectrum below 10 MHz in (a) and (c) reflects the antenna sensitivity. (a) Magnetic component when 0.1–100 kHz RF fields were applied (“RF on” condition); (b) Electric component when 0.1–100 kHz RF fields were applied (“RF on” condition); (c) Magnetic component in the “RF off” condition; (d) Electric component in the “RF off” condition. Blue: signals measured in the “average” mode. Red: signals measured in the “maxhold” mode.

2.4. Behavioural Experiments and Statistical Analysis

Birds were first removed from their aviary ca. half an hour before sunset, and left outside for an hour, giving them the opportunity to recalibrate their magnetic compass [44]. At the end of civil twilight (when the Sun is 6° below the horizon), birds were transferred to modified plastic Emlen funnels [45] placed at 9 locations within the magnetic coil system. The funnels were lined with scratch-sensitive paper [46] and a translucent lid was placed atop each funnel. In the funnels the birds tend to jump in the direction in which they would fly if released [47, 48] and, in so doing, they leave scratches on the papers that line the funnels [45, 46]. Birds were left in the funnels for an hour under dim white light of intensity 2.5 mWm⁻². In some cases another group of 9 birds was tested in a second round immediately following the first. The join of the scratch paper was oriented to one of the four cardinal directions by one experimenter, and the mean scratch direction relative to the join was judged by eye to the nearest 10° by two other experimenters acting independently. The other experimenter set up the time-dependent and static magnetic fields to ensure double-blinding with respect to the experimental conditions for the two who assessed the birds' orientation. If there were fewer than 30 scratches or if the two estimated directions differed by more than 30°, the birds were deemed 'not active' or 'random', respectively. In the case of an active non-random bird, its mean direction was calculated from the independent values reported by the two assessors.

First, birds were pre-tested in the NMF and CMF conditions, and the birds that did not adjust their orientational preference to the rotated magnetic field were eliminated from subsequent testing.

All active, non-random birds were tested again in three conditions: NMF with no RF field, NMF with a 0.1–100 kHz broadband field (NMF-RF) and CMF with the same broadband field (CMF-RF). Birds were tested at least 4 times in each condition. Additional tests were performed if the birds did not pass the inclusion criteria (see below) in all conditions and if time permitted us to get larger sample sizes (Table S1 in Supplementary Information). The decision on the number of times to test a bird was not influenced by the direction of its responses.

In the final analysis we included only those birds that showed at least two directed non-random orientation values in each condition and a length of the mean vector r per bird in the control condition of more than 0.2 (7 birds in 2017, 8 birds in 2018) (Table S1).

To compare results from the two migratory seasons we used the rank-sum test for small sample sizes as described in Batschelet (1981) [49]. Since there was no significant difference in orientation in the NMF between the seasons (rank-sum test, $U = 21$, $p > 0.1$), we analysed data from the two seasons together.

Overall consistency in group migratory direction in a given magnetic condition was tested using the Rayleigh test of uniformity, whilst the significance of differences between conditions was tested using the Mardia-Watson-Wheeler test [49].

To compare the number of ‘not active’ and ‘random’ trials in the different field conditions we calculated the number of random or not active trials divided by the total number of trials for each bird in each condition. The data were analysed by ANOVA (type III from the “car” package) of the Generalized Linear Mixed Model (GLMM, glmer function from “LmerTest” package) for the binomial distribution. The directedness (r -values) of individual birds in the different conditions was also compared using ANOVA of GLMM.

All statistics were computed in R [50], and graphs were produced in MATLAB R2016a.

3. Results

Birds tested during the spring migratory season oriented significantly in the season-specific north-easterly direction in the normal magnetic field both in the control condition (NMF, Fig. 3a; mean = 68.4° , SD = 65.9° ; Rayleigh test; $r = 0.5164$, $p = 0.0157$) and when exposed to very “clean” (Fig. 1 and Fig. 2) 0.1–100 kHz RF fields (NMF-RF, Fig. 3b; mean = 47.5° , SD = 68.8° ; $r = 0.4859$, $p = 0.0262$). Orientation in NMF-RF did not differ from that in the NMF control condition (Mardia-Watson-Wheeler test; $w = 0.995$, $p = 0.61$). After rotation of the magnetic field through -120° , the birds changed their orientation accordingly and remained significantly oriented (CMF-RF, Fig. 3c; mean = 311.3° , SD = 59.3° ; $r = 0.5852$, $p = 0.0042$). This rotation in the mean direction between NMF-RF and CMF-RF conditions was significant (Mardia-Watson-Wheeler paired test; $w = 7.71$, $p = 0.021$) and did not differ from the expected -120° shift (Mardia-Watson-Wheeler paired test, $w = 1.543$, $p = 0.462$).

Furthermore, we found no significant difference in the proportion of birds that oriented randomly between conditions (ANOVA, $p = 0.30$) as well as no difference in the proportion of inactive trials between conditions (ANOVA, $p = 0.32$).

We also found no difference in directedness at the individual level by comparing the r -values from Rayleigh statistics for each individual in the different experimental conditions

(mean $\pm$ SD: NMF = 0.52 ± 0.22 , NMF-RF = 0.56 ± 0.26 , CMF-RF = 0.51 ± 0.26 ; ANOVA, $p = 0.538$).

In short, under conditions in which 20–450 kHz RF fields had previously been found to disrupt birds' ability to use their magnetic compass, we found no evidence that 0.1–100 kHz fields caused a similar effect.

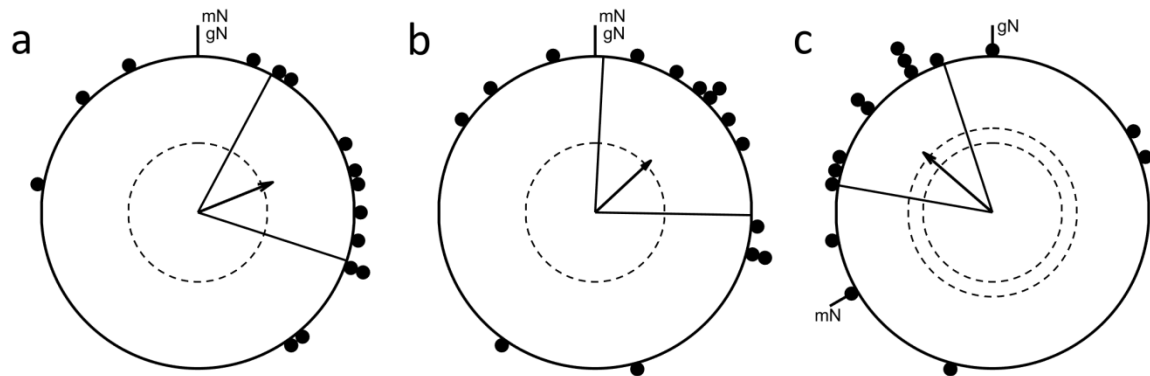


Figure 3

Magnetic compass orientation of European Blackcaps in (a) NMF – normal Earth's magnetic field in Oldenburg, (b) NMF-RF – normal Earth's magnetic field plus 0.1–100 kHz RF fields, (c) CMF-RF – 120°-turned Earth's magnetic field plus 0.1–100 kHz RF fields. Each dot represents the mean direction for an individual bird. Arrows show the group mean orientation, flanked by the confidence intervals ($\pm 95\%$); dashed circles indicate threshold p -levels (0.05 for inner circle, 0.01 for outer circle) of the Rayleigh test for the corresponding sample size. gN – geographic North, mN – magnetic North.

4. Discussion

In the experiments reported here, we found significant magnetic orientation of Eurasian blackcaps in both the control condition and when a noise-modulated, broadband, 0.1–100 kHz RF field was applied, in both the Earth's magnetic field and when its horizontal component had been rotated by -120° (Fig. 3 and Table S1 in Supplementary Information).

For an RF field to have a resonant effect on the singlet-triplet interconversion of a radical pair, the lifetime of the spin coherence in the radical pair must exceed the period of the RF oscillation. If this condition is not met, the spin dynamics of the radical pair would be

unchanged and the operation of the magnetic compass unaffected [30]. Thus, the absence of disorientation caused by a 0.1–100 kHz broadband field leads to the conclusion that the coherence lifetime does not exceed 10 μ s (the period of the highest frequency component of those fields). In their 2014 study, Engels et al. [34] observed disorientation in European robins (*Erithacus rubecula*) exposed to 20–450 kHz broadband electromagnetic noise. Disruption of the compass sensor by a 450 kHz field suggests a coherence lifetime of 2.2 μ s or longer. If the current theoretical model of radical pair magnetoreception is reliable [30], the results of these two studies therefore indicate that the spin-coherence lifetime of the avian magnetoreceptor is likely to be somewhere between 2 μ s and 10 μ s.

Clearly, in order to have a spin-coherence lifetime of 2–10 μ s, the radical pair must live for at least 2–10 μ s. If the disappearance of the radicals were much slower than this, the magnetic sensitivity would be reduced for the following reason. Once the coherence has gone, and the spin system is at thermal equilibrium, an external magnetic field can no longer influence the spin dynamics and thereby affect the yield of the reaction product that acts as the signalling state [13]. For sensitive detection of the Earth's magnetic field, the majority of the radical pairs should react while significant spin coherence remains. Therefore it seems probable that the radical pairs have a lifetime similar to that of their spin coherence, i.e. 2–10 μ s. Our data agree with the results reported by Ritz and co-authors (2009) [31], who did not observe an effect of monochromatic RF fields at frequencies up to 100kHz on the orientation of European robins. However, we have had difficulties replicating the effects of other monochromatic RF-fields reported in the same paper [35]. In any case, the broadband RF field used in the current study has an important advantage over monochromatic fields when testing for the properties of the radical pair mechanism. The hyperfine interactions between the unpaired electrons and the nuclear spins in any realistic receptor molecule produce a variety of energy levels, meaning that a range of radiofrequencies can affect the singlet-triplet interconversion [13, 30]. According to this model, radical pairs would have an almost uniform distribution of resonances spread over a range of frequencies up to at least 100 MHz rather than just a few resonances in this range [30]. Hence, a broadband field should have stronger cumulative effects on the radical-pair spin dynamics than a single-frequency field of comparable root-mean-square amplitude as observed in a recent orientation experiment on European robins [35]. Even using broadband 0.1–100 kHz RF fields, we saw no effect on the magnetic orientation of blackcaps. The intensity of the RF-field is an important factor influencing the resonance effect on the radical pair. The broadband field used in our study

was ca. 3 times stronger than the lowest threshold that was shown to have an effect on the orientation of European robins [34] (see Table 1). Although we consider it unlikely, we cannot exclude species-specific differences in the sensitivity to RF fields (as has been suggested for the Larmor frequency [31, 33]). The conclusions of this study rest on the assumption that in the absence of spin relaxation the radical pairs are equally sensitive to different frequencies of broadband noise provided one compares frequency bands with similar overall intensities as judged by measures such as $\bar{b}$ and B_{rms} (Table 1). Until considerably more is known about the properties of the radicals *in vivo*, it is impossible to be certain that this is reliable. We also assume that if there exists an unknown amplification mechanism, as suggested in Ref. 30, it too operates equally efficiently at different frequencies.

Several previous studies of the effects of RF-fields on magnetic compass orientation have resulted in seemingly contradictory findings ([29, 31, 33], but see [35]). The possible reasons behind these discrepancies are debatable and could include differences in the sample sizes, as well as equipment and measurement quality [3, 13, 30, 37]. In our study we provide: 1) All the parameters of the static magnetic fields and their homogeneity (see Methods 2.2); 2) The measurements of the electric and magnetic field components in the range up to 100MHz both for control and experimental conditions (Fig. 2); 3) The amplitude of the RF field measured both in *average* and *maxhold* modes (Fig. 1 and Fig. 2); 4) Root-mean-square RF noise intensities to allow comparison with RF fields that have different frequency ranges (see Methods 2.3); 5). The spectral amplitude in T/ $\sqrt{\text{Hz}}$ and not simply T. The latter is meaningless without also quoting the measurement bandwidth. As such, we are confident that the stimuli described were actually the only electromagnetic noise stimuli the birds were exposed to, and that our birds were not exposed to resonant frequencies or physical anomalies caused by the experimental apparatus.

A spin-coherence lifetime of 2-10 μs is compatible with the “quantum needle” proposed by Hiscock et al. (2016) [38]. Spin dynamics simulations suggest that flavin-containing radical pairs in cryptochromes could respond particularly sensitively to certain directions of the Earth’s magnetic field if the coherence persists for more than about 1 μs . In some situations, the yield of the magnetic signalling state is predicted to show a pencil- or needle-shaped dependence on the orientation of the protein that could afford a bird with a precise compass bearing. For a three-dimensional representation of the highly anisotropic response when the spin-coherence lifetime is 10 μs , see Fig. 1F in [38].

While considerable work is still required to establish the mechanism beyond reasonable doubt, our finding of a 2-10 μ s coherence lifetime provides an important basis for future *in silico* and *in vivo* studies of radical-pair based magnetoreception.

5. Ethics

All experimental procedures were approved by the Animal Care and Use Committees of the Niedersächsisches Landesamt für Verbraucherschutz und Lebensmittelsicherheit (LAVES, Oldenburg, Germany, 33.19-42502-04-13/1065, 33.19-42502-04-17/2724).

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8. Author contributions

H.M., P.J.H., H.H. and M.W. designed the research; D.K., J.W., J.X. and R.C. performed the experiments; D.K., J.W. and M.W. performed the statistical analyses; H.M. provided reagents and facilities; all authors worked on the first draft of the paper which was written by D.K. and J.W.

9. Data accessibility

Original data are available in the supplementary materials.

10. Declaration of interests

The authors declare no competing interests.

11. References

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Electromagnetic 0.1–100 kHz noise does not disrupt orientation in a night-migrating songbird implying a spin-coherence lifetime of less than 10 μ s

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Supplementary Information

ID	NMF				NMF-RF				CMF-RF				year
	<i>dir</i>	<i>r</i>	<i>N</i>	<i>Nval</i>	<i>dir</i>	<i>r</i>	<i>N</i>	<i>Nval</i>	<i>dir</i>	<i>r</i>	<i>N</i>	<i>Nval</i>	
12 grey	31.6	0.73	7	4	305.5	0.72	8	4	278.2	0.28	5	3	2018
83 green	280.0	0.71	5	2	45.0	0.91	7	2	260.0	1.00	4	2	2018
14 orange	108.7	0.27	7	3	66.4	0.80	5	3	67.5	0.92	6	2	2018
38 grey	108.4	0.28	7	5	215.6	0.19	9	4	329.3	0.87	4	4	2018
27 grey	102.1	0.60	4	4	317.9	0.39	6	6	238.0	0.41	4	4	2018
20 grey	147.3	0.90	7	3	42.1	0.37	7	3	195.0	0.58	8	3	2018
19 grey	138.5	0.63	9	5	105.9	0.68	6	3	328.6	0.36	4	3	2018
15 grey	80.4	0.86	3	3	165.9	0.46	9	6	285.0	0.33	6	3	2018
81 green	32.9	0.43	6	6	42.5	0.84	6	2	339.7	0.28	5	5	2017
30 green	74.9	0.22	6	4	53.0	0.36	5	4	330.8	0.45	5	5	2017
59 green	89.0	0.58	6	6	347.2	0.21	5	5	1.8	0.30	5	5	2017
74 green	313.3	0.50	10	7	95.0	0.87	5	2	290.0	0.82	5	2	2017
40 grey	65.3	0.36	6	4	29.2	0.87	5	5	57.5	0.45	6	6	2017
79 green	337.0	0.45	10	6	105.1	0.45	6	4	308.9	0.26	5	3	2017
80 green	17.5	0.26	10	9	13.7	0.31	7	5	310.0	0.34	6	2	2017

Table S1. Summary of experimental results for each individual (ID) with mean orientation (*dir*) and Rayleigh value (*r*) for this orientation; *N* – overall number of trials, *Nval* – number of trials in which orientated activity was observed, NMF – normal static magnetic field in Oldenburg, NMF-RF: normal static magnetic field with RF field present, CMF-RF: –120°-turned static magnetic field with RF field present.