



Predatory Bird
Monitoring Scheme
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Anticoagulant rodenticides in predatory birds 2010: a Predatory Bird Monitoring Scheme (PBMS) report

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Executive Summary

The Predatory Bird Monitoring Scheme (PBMS; <http://pbms.ceh.ac.uk/>) is the umbrella project that encompasses the Centre for Ecology & Hydrology's National Capability contaminant monitoring and surveillance work on avian predators. By monitoring sentinel vertebrate species, the PBMS aims to detect and quantify current and emerging chemical threats to the environment and in particular to vertebrate wildlife.

Anticoagulant rodenticides, and in particular second generation anticoagulant rodenticides (SGARs), can be toxic to all mammals and birds. Predators that feed upon rodents are particularly likely to be exposed to these compounds. The PBMS, together with other studies, have shown that there is widespread exposure to SGARs of a diverse range of predators in Britain and that some mortalities occur as a result. This report summarises the PBMS monitoring for anticoagulant rodenticides in barn owls (*Tyto alba*), kestrel (*Falco tinnunculus*) and red kites (*Milvus milvus*) that were found dead in 2010 and presents long term trend analysis for barn owls.

SGARs were detected in 91% of 53 barn owls analysed and the most prevalent compounds were difenacoum and bromadiolone. The majority of the residues were low and not diagnosed as directly causing mortality. The livers from 24 red kites analysed in 2010. Most (92%) had detectable liver SGAR concentrations, again mainly difenacoum and bromadiolone, although brodifacoum was also detected in over half the birds. Five of the red kites analysed showed signs of haemorrhaging thought possibly to be associated with rodenticide poisoning. However, only one of these birds had a sum SGAR liver concentration greater than 0.05 µg/g wet weight and the contribution of SGARs, if any, to the death of the other four birds is uncertain. SGARs were detected in 8 of 10 kestrels analysed. The most prevalent rodenticides detected in kestrel livers were those approved for outdoor use while the co-occurrence of multiple residues was also prevalent with 6 out of 10 kestrels having more than one SGAR present in their liver.

Difethialone, a SGAR marketed since late 2011, was not detected in any of the three species tested in 2010. This represents a base-line against which future results can be compared.

SGARs have been monitored in barn owls since 1983. Data on long-term trends have been adjusted to account for changes over time in sensitivity of analytical methods. This has meant that very low residues (<0.025 µg/g wet weight), which are now detectable, are not included in the time trend analysis. Overall, the proportion of barn owls with detectable liver concentrations of one or more SGAR has increased significantly over the course of monitoring. The highest value was recorded in 2008 but this was approximately twice that for the previous three years. The value for 2010 (23%) was lower than 2008 and is the lowest recorded since 2005.

The proportion of barn owls with detectable SGAR residues over the period 1990-2010 was twofold higher in England than in Scotland and Wales. Within England, the proportion of owls with datable residues was higher in the Defra Eastern region than elsewhere.

1. Introduction

1.1 Background to the PBMS

The Predatory Bird Monitoring Scheme (PBMS; <http://pbms.ceh.ac.uk/>) is the umbrella project that encompasses the Centre for Ecology & Hydrology's long-term contaminant monitoring and surveillance work on avian predators. The PBMS is a component of CEH's National Capability activities.



By monitoring sentinel vertebrate species, the PBMS aims to detect and quantify current and emerging chemical threats to the environment and in particular to vertebrate wildlife. Our monitoring provides the scientific evidence needed to determine how chemical risk varies over time and space. This may occur due to market-led or regulatory changes in chemical use and may also be associated with larger-scale phenomena, such as global environmental change. Our monitoring also allows us to assess whether detected contaminants are likely to be associated with adverse effects on individuals and their populations.

Overall, the PBMS provides a scientific evidence base to inform regulatory decisions about sustainable use of chemicals (for example, the [EU Directive on the Sustainable Use of Pesticides](#)). In addition, the outcomes from the monitoring work are used to assess whether mitigation of exposure is needed and what measures might be effective. Monitoring also provides information by which the success of mitigation measures can be evaluated.

Currently, the PBMS has two key objectives:

- (i) to detect temporal and spatial variation in exposure, assimilation and risk for selected pesticides and pollutants of current concern in sentinel UK predatory bird species and in species of high conservation value
- (ii) in conjunction with allied studies, to elucidate the fundamental processes and factors that govern food-chain transfer and assimilation of contaminants by top predators.

Further details about the PBMS, copies of previous reports, and copies of (or links to) published scientific papers based on the work of the PBMS can be found on the [PBMS website](#).

1.2 PBMS monitoring of anticoagulant rodenticides

Second generation anticoagulant rodenticides (SGARs) can be toxic to all mammals and birds. Predators that feed upon rodents are particularly likely to be exposed to these compounds. The PBMS (see previous reports, also Newton et al., 1999, Shore et al., 2006, Walker et al., 2008a,b) together with other studies (Dowding et al., 2010, McDonald et al., 1998, Shore et al., 2003a,b) have shown that there is widespread exposure to SGARs of a diverse range of predators in Britain. Defra’s Wildlife Incident Monitoring Scheme (WIIS)² and the PBMS have shown that in the UK some mortalities result from this exposure.

In response to conservation concerns over the potential impacts of SGARs on predators, the PBMS has monitored trends in exposure to second generation anticoagulant rodenticides (SGARs) in a sentinel species, the barn owl (*Tyto alba*). This has been done since 1983 and the findings from previous years and analyses of long-term trends are given in previous PBMS reports and by Newton et al., (1990, 1999). The red kite (*Milvus milvus*) is a high conservation priority species that has been reintroduced to England as part of the red kite species recovery programme (Carter and Grice 2002). SGAR-induced deaths of kites have been detected by the Wildlife Incident Investigation Scheme. Until 2007 only a small number of red kites were received and analysed by the PBMS each year although this showed that a large proportion of reintroduced birds were exposed to SGARs (Walker, Shore et al. 2008). The development of a recent collaboration with the Institute of Zoology has meant that the number of liver samples available for analysis has now usually increased. Kestrels (*Falco tinnunculus*) have been monitored since 2000 following a pilot study that demonstrated a relatively high level of exposure compared with barn owls (Shore, Malcolm et al. 2001) and conservation concerns over declines in kestrel populations (<http://www.bto.org/birdtrends2009/wcrkestr.shtml>).

This report describes the results of PBMS monitoring of barn owls, kestrels and red kites submitted to the PBMS in 2010 (Table 1.1). This involved measuring liver residues in carcasses submitted to the PBMS by members of the public. The birds died from various causes, but mainly from road traffic collisions and from starvation. The provenance of the birds is shown in Figure 1.1.

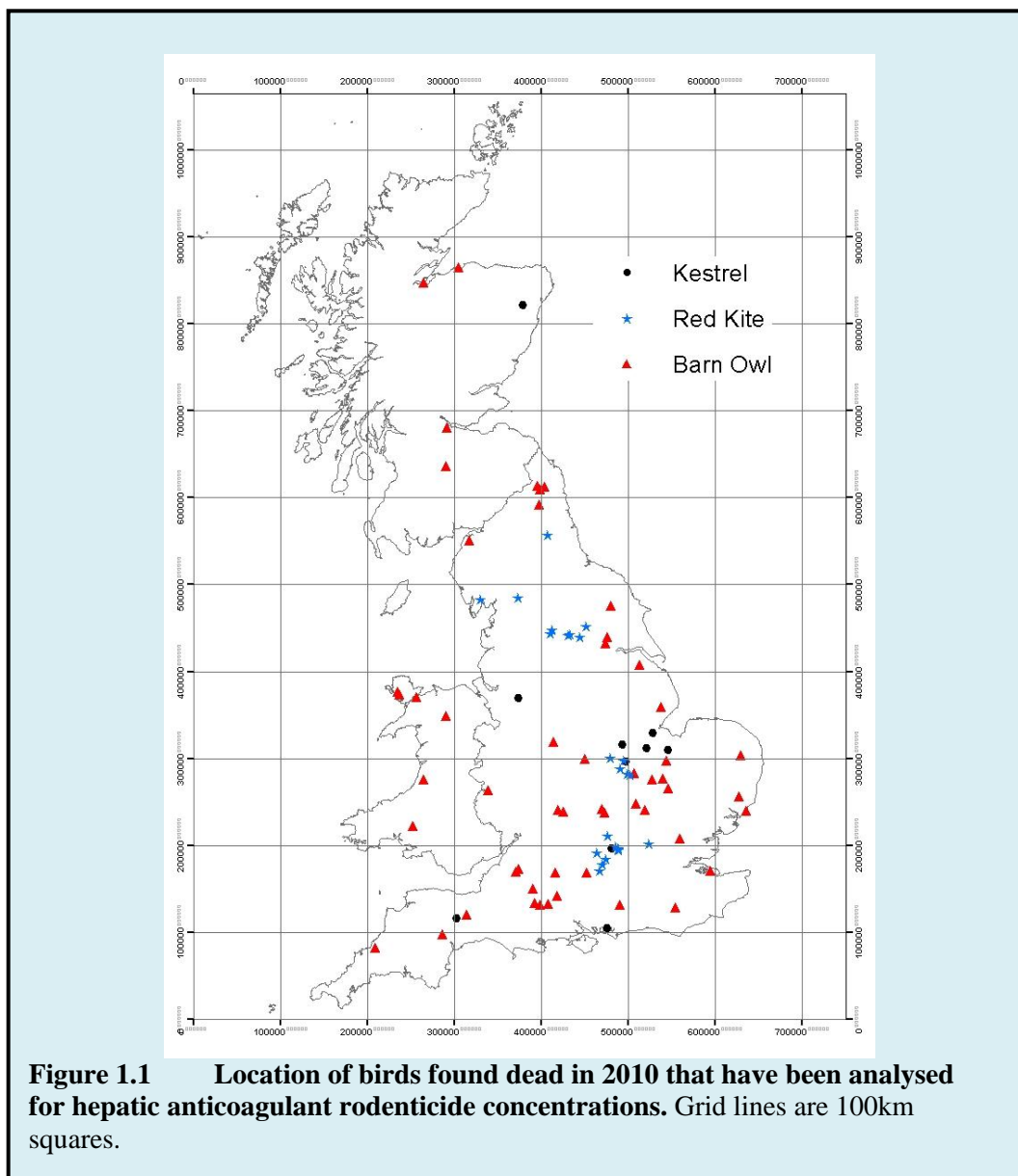
Species		Received	Analysed
barn owl	<i>Tyto alba</i>	100	53
kestrel	<i>Falco tinnunculus</i>	15	10
red kite	<i>Milvus milvus</i>	24	24
Total		139	87

All red kites were autopsied and analysed. All the barn owls received were autopsied but,

² Annual WIIS reports are available at www.pesticides.gov.uk/environment.asp?id=58

because of the large number, a sub-sample of just over 50 birds per year (stratified by date found) were analysed. Similarly a sub-sample of 10 kestrels have been analysed. Tissues from all birds received were archived in the PBMS tissue and egg archive where they are available for future research purposes.

Since 2006, the concentrations of warfarin and coumatetralyl (first generation hydroxycoumarins) and the presence or absence of diphacinone and chlorophacinone (indandiones) have been quantified in addition to SGARs. A summary of the analytical methods can be downloaded from the PBMS website (http://pbms.ceh.ac.uk/docs/AnnualReports/PBMS_Rodenticides_Methods.pdf). Anticoagulant rodenticide concentrations are reported as $\mu\text{g/g}$ wet weight (wet wt) throughout this report.



2. Anticoagulant rodenticide concentrations in birds submitted to the PBMS in 2010

Summary statistics for the incidence of detectable concentrations of anticoagulant rodenticides in the barn owls and red kites that were analysed are given in Table 2.1. Results for individual birds are given in a downloadable addendum to this report ([Addendum to “Anticoagulant rodenticides in predatory birds 2010: a Predatory Bird Monitoring Scheme \(PBMS\) report”](#)).

Table 2.1. Number of birds with detectable liver concentrations of anticoagulant rodenticides (No/) and the percentage this comprised of all birds analysed (%). Total number of barn owls, kestrels and red kites analysed was 53, 10 and 24, respectively.

	Limit of Detection ²	barn owls		kestrels		red kites	
		No/	%	No/	%	No/	%
<i>2nd Generation (SGAR)</i>							
bromadiolone	1.4	42	79	8	80	20	83
difenacoum	1.2	37	70	6	60	17	71
flocoumafen	1.1	0	0	0	0	0	0
brodifacoum	1.4	24	45	2	20	16	67
difethialone ¹	1.0	0	0	0	0	0	0
Any SGAR	-	48	91	8	80	22	92
Multiple SGARs	-	37	70	6	60	20	83

¹ 30, 10 and 16 livers from barn owls, kestrels and red kites, respectively, were tested for difethialone.
² LoDs reported in ng/g wet wt.

2.1 Difethialone

Difethialone has recently been licensed for indoor use only in Britain. This compound (CAS Number 104653-34-1), has now been added to the analysis suite for the PBMS, although only in time for a sub-set of all the birds sampled to be analysed. Therefore a set of 30 barn owls, 10 kestrels and 16 red kites were tested for difethialone. It was not detected in any liver sample. This year’s analysis represents a base-line against which future monitoring can be compared.

2.2 Barn Owls collected in 2010

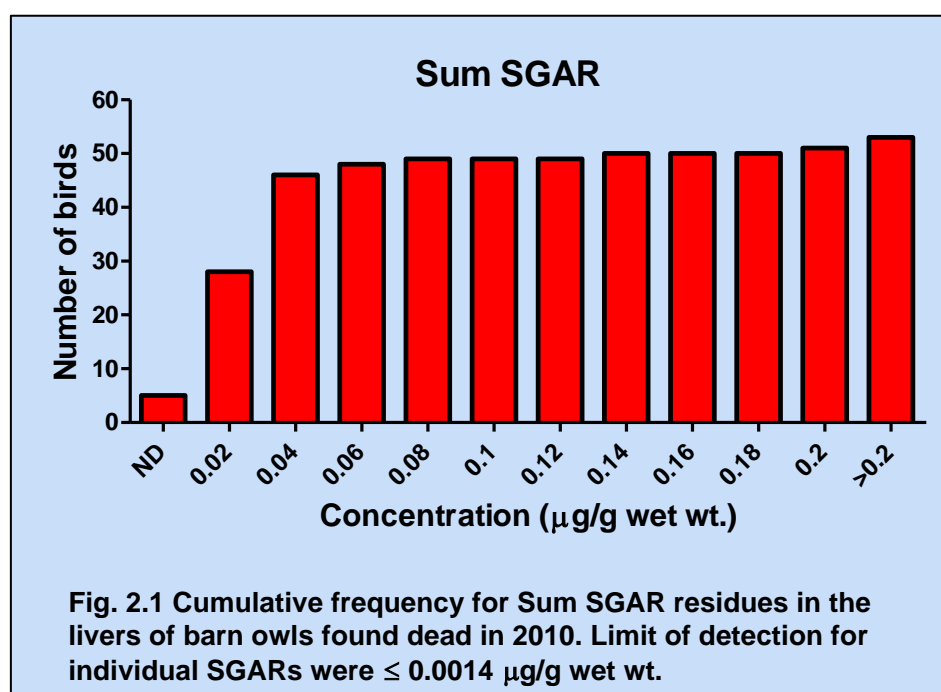
Fifty-three barn owls were analysed by PBMS in 2010; all had died in that year. Forty-eight (91% of the sample) contained detectable liver concentrations of one or more SGAR (Table 2.1) which is similar to those values reported for 2007 to 2009.



As in previous years, the majority of exposure was to bromadiolone and difenacoum (89% of barn owls analysed). Brodifacoum was detected less frequently (45% of birds analysed; Table 2.1). None of the other anticoagulant rodenticides were detected in any of

the 53 owls analysed. Overall, multiple SGAR residues were detected in 70% of the livers analysed.

The potentially lethal range for SGAR residues in barn owls has variously been described as $> 0.1 \mu\text{g/g}$ wet wt (Newton, Dale et al. 1998) and $> 0.2 \mu\text{g/g}$ wet wt (Newton, Shore et al. 1999) and is so classed on the basis of two sets of observations. The first was that owls diagnosed at post-mortem of having died from rodenticide poisoning (because they had characteristic signs of haemorrhaging from such organs as the heart, lungs, liver, brain and/or subcutaneous areas) almost all had liver residues $>0.1 \mu\text{g/g}$ wet wt. The second was that owls that had been experimentally poisoned had residues of the range $0.2\text{-}1.72 \mu\text{g/g}$ wet wt (Newton, Shore et al. 1999). This range has been used in this report as an indicator of concern that SGARs may have an adverse effect on individuals although recent analysis (Thomas, Mineau et al. 2011) suggests that effects on some individuals may be associated with residues $<0.1 \mu\text{g/g}$ wet wt.



Most owls had concentrations below the potentially lethal range (Figure 2.1) but four (7.5% of the sample) had residues (summed values for all four SGARs) greater than $0.1 \mu\text{g/g}$ wet wt; two of these exceeded $0.2 \mu\text{g/g}$ wet wt. The maximum liver concentration amongst these seven owls was $0.243 \mu\text{g/g}$ wet wt (the majority of which was brodifacoum with traces of difenacoum & bromadiolone) which was detected in a bird that had been diagnosed as a probable road casualty. The second highest liver residue was $0.227 \mu\text{g/g}$ wet wt ($0.085 \mu\text{g/g}$ wet wt. brodifacoum, $0.142 \mu\text{g/g}$ wet wt. bromadiolone) in a bird diagnosed of dying from starvation; no internal haemorrhaging was apparent. Although the remaining two owls had liver residues between 0.1 and $0.2 \mu\text{g/g}$ wet wt., no internal haemorrhaging was apparent.

2.3 Red kites collected in 2010

Liver samples from 24 red kites that had died in 2010 were analysed. Twenty-two (92%) of the birds contained detectable concentrations of anticoagulant rodenticides (Table 2.1) with 20 birds having been exposed to more than one SGAR.



Interpretation based on such a small sample has to be limited. However, as with barn owls and kestrels, the most prevalent rodenticide detected in red kite livers was difenacoum (Table 2.1). Similarly to 2007 to 2009 a large proportion (67%) of red kite livers also contained brodifacoum, although unlike in 2007 & 2008 this was not significantly higher than the proportion of owls in which brodifacoum was detected.

The sum SGAR liver concentrations in red kites were generally higher than those observed in barn owls. Concentrations in livers with detectable SGAR residues ranged between 0.003 and 0.769 $\mu\text{g/g}$ wet wt with a median concentration of 0.120 $\mu\text{g/g}$ wet, which was 6-fold higher than in barn owls. The maximum liver concentration was 0.769 $\mu\text{g/g}$ wet wt. which was entirely due to brodifacoum.

Post mortem examinations by the Institute of Zoology indicated that five of the 24 red kites received showed internal hemorrhages not associated with detectable trauma and therefore consistent with anticoagulant poisoning. The sum SGAR liver concentration in these in one of these birds was 0.769 $\mu\text{g/g}$ wet wt; all of which was brodifacoum. Thus it seems likely that brodifacoum was the cause of death in this bird. The sum SGAR liver concentrations in the other four birds ranged between non-detected and 0.040 $\mu\text{g/g}$ wet wt. Similar residues (up to 0.644 $\mu\text{g/g}$ wet wt.) were detected in birds thought to have died due to other causes. Therefore the contribution, if any, of rodenticides to the death of these four individuals is uncertain.

2.3 Kestrels collected in 2010

Liver samples from 10 kestrels that had died in 2010 were analysed. Eight (80%) of the birds contained detectable concentrations of one or more anticoagulant rodenticides.



The most prevalent rodenticides detected in kestrel livers were those approved for outdoor use (80% of birds, Table 2.1). Bromadiolone was found in every bird which had detectable SGAR residues while difenacoum co-occurred with bromadiolone in six of the birds. Flocoumafen was not detected in any of the kestrels tested while brodifacoum was found in two birds. As these

figures suggest the co-occurrence of multiple residues was high with more than one rodenticide being detected in 6 birds.

Concentrations in livers with detectable SGAR residues ranged between 0.002 and 0.164 µg/g wet wt with a median concentration of 0.077 µg/g wet, which was 3-fold higher than that measured in barn owls. However, due to the large variance within the sample, the difference in sum SGAR liver concentrations between kestrels and barn owls was not statistically significant.

Next year will include analysis of a further 10 kestrel livers which will be combined with this year's results in order to test for temporal trends of SGARs in this species.

3. Long term trends in liver SGAR concentrations in barn owls

A common limit of quantification (LoQ) was applied to the long-term dataset for SGARs. This was 0.025 µg/g wet wt. and was applied to each of the four compounds as described in Walker et al. (2010). Any detected values below this 0.025 µg/g wet wt. LoQ were re-assigned as non-detected values and the percentage occurrence of SGARs were then recalculated for each year - these are termed “adjusted % detected” values. The use of adjusted % detected values under-estimates the true occurrence of liver SGAR residues for compounds and years where the limit of quantification was substantially lower, but it eliminates biases in the long-term data due to improvement in the sensitivity of analysis over time. The adjusted % detected values therefore provide a measure of temporal changes but do not necessarily indicate the actual scale of exposure. Adoption of a common limit of detection for different SGARs eliminates detection biases when comparing % detection values for different rodenticides.



The adjusted % detected values for one or more SGAR in barn owl livers has increased from zero in 1983 (based on a small sample size of 4 livers), when monitoring began, to a maximum of 49% in 2008 (Figure 3.1). This long-term change primarily reflects an increase over time in the proportion of birds with detectable residues of difenacoum and/or bromadiolone; the proportion of birds that have multiple compounds in their livers has also increased (Figure 3.1). Brodifacoum, and to a lesser extent flocoumafen, have been detected in barn owls during the course of the monitoring period but there is no evidence of any significant progressive change in exposure over time (Figure 3.1).

The adjusted % detected value for birds in 2010 was 23% and is the lowest value reported since 2005 (Figure 3.1). However there is considerable inter-year variation over the last 10 years (Coefficient of Variation: 25.8%) and further monitoring is required to determine if there is any long-term decline in exposure.

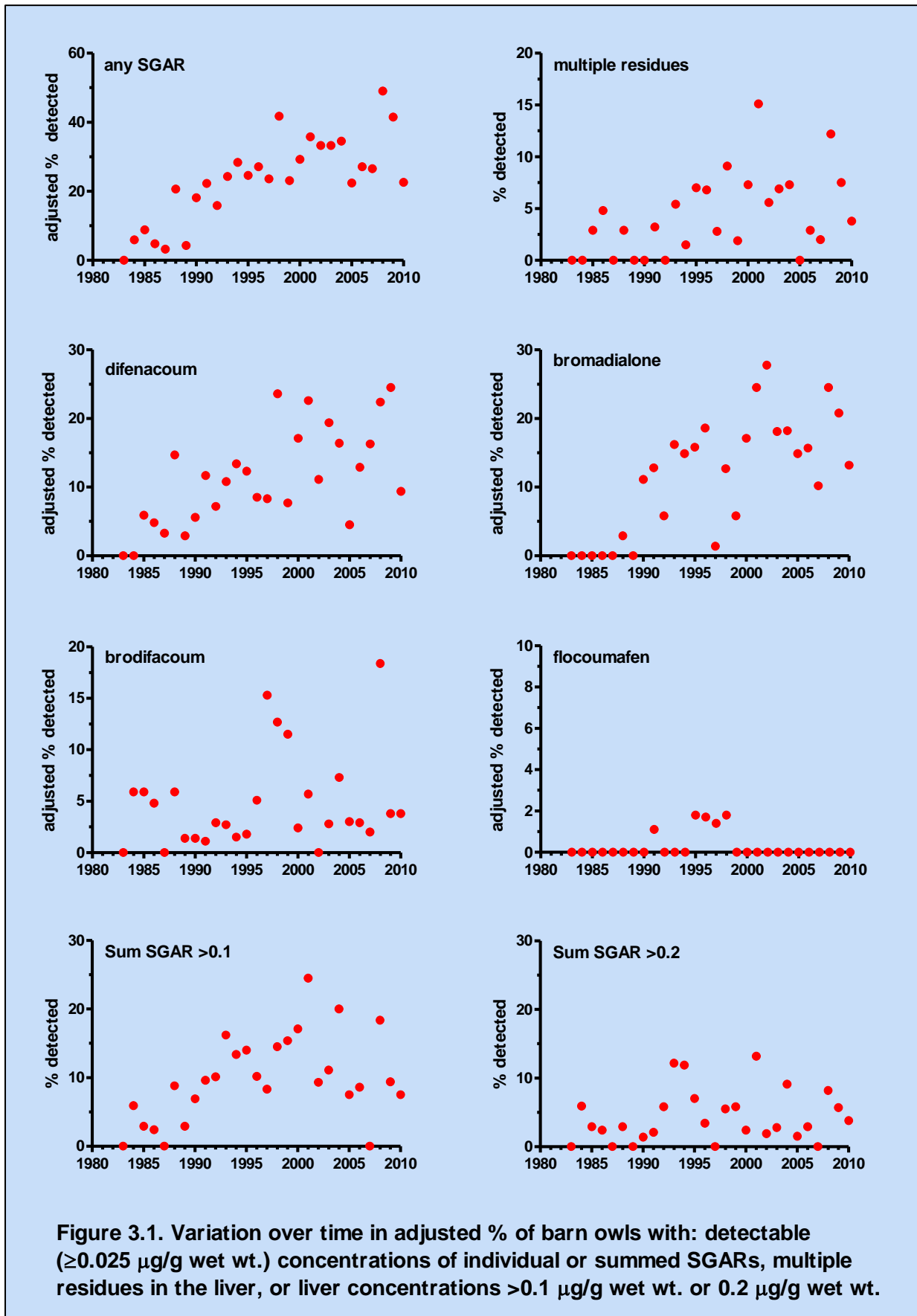
In terms of potential adverse effects, the 2010 results are consistent with those previously reported (Walker et al., 2010) in that the proportion of barn owls with liver concentrations above 0.1 µg/g wet wt. has risen during the course of monitoring over time ($r_s=0.417$, $P<0.05$) but there has been no significant change in the proportion of birds with liver residues > 0.2 µg/g wet wt. (Figure 3.1). Overall, the average proportion of owls analysed that had SGAR residues > 0.2 µg/g wet wt is 4.5%, but the cause of death in many of these birds has not been attributed to anticoagulant rodenticides.

Table 3.1. Number (n) of owls and the number as a percentage of all birds tested (%) from England, Scotland and Wales between 1990 and 2010 that had detectable liver SGAR concentrations ≥ 0.025 $\mu\text{g/g}$ wet wt. (common limit of quantification applied to all compounds and samples).

	number (% of whole sample tested) of owls with detected residues						Chi Squared statistic ¹
	England (n=1055)		Scotland (n=113)		Wales (n=111)		
Bromadiolone	173	(16%)	12	(11%)	7	(6.3%)	9.89 (**)
Difenacoum	151	(14%)	7	(6.2%)	10	(9.0%)	7.71 (*)
Flocoumafen	2	(0.2%)	1	(0.9%)	0	(0%)	-
Brodifacoum	57	(5.4%)	4	(3.5%)	1	(0.9%)	4.87 (ns)
Any SGAR	319	(30%)	20	(18%)	17	(15%)	17.5 (***)
Multiple SGAR	59	(5.6%)	4	(3.5%)	1	(0.9%)	5.21 (ns)

¹ ns = not significant, * = P<0.05, *** = P<0.001; unable to test flocoumafen

The scale of exposure of barns owls in England, Scotland and Wales has also been compared using the data available pooled for the years 1990-2010 to provide sufficient sample size for analysis. The adjusted % of owls with detected residues of any SGAR was approximately two-fold higher in England than in either Scotland or Wales and the difference between the countries was significantly different (Table 3.1). At a smaller scale there were also significant differences among regional areas of Great Britain as defined by Defra ($\chi^2=31.0$, $P<0.001$; Figure 3.2). In contrast to the previous report (Walker, Llewellyn et al. 2010), if Scotland and Wales were excluded from the analysis then there was still a significant difference between the English regions ($\chi^2=12.96$, $P=0.012$). The highest proportion of barn owls with SGAR residues was for birds from Eastern England (33%).



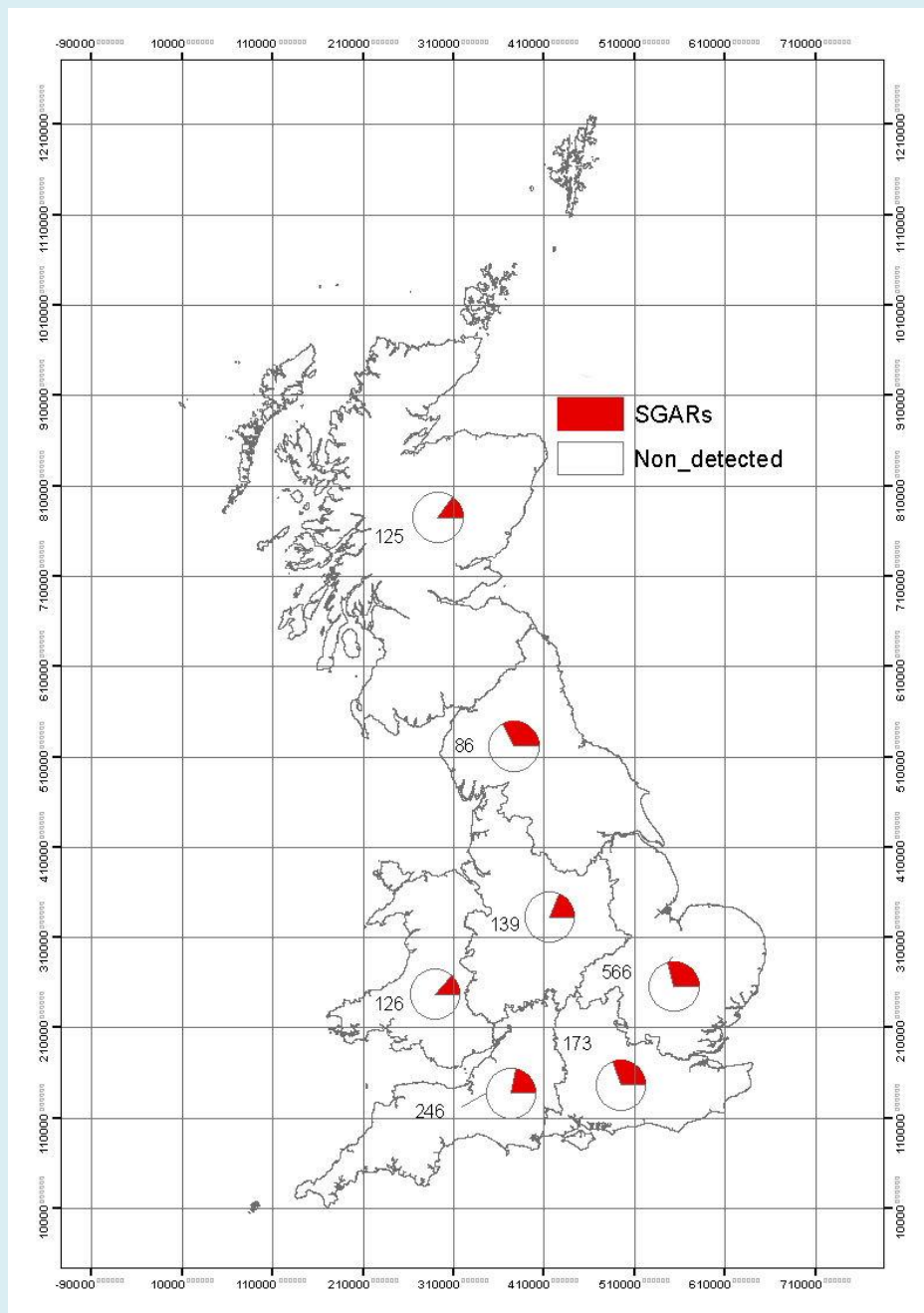


Figure 3.2 Proportion of barn owls (*Tyto alba*) that have detectable residues greater than $0.025\mu\text{g/g}$ wet wt. in their liver for each Defra region (Dawson and Garthwaite 2004). Number in next to each pie chart indicates number of samples. Grid lines are 100km squares.

4. Acknowledgements

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