







Waterford Institute of Technology

The Vincent Wildlife Trust and Waterford Institute of Technology Pine Marten Scat DNA Survey of England and Wales 2008-2009

John Messenger and Lizzie Croose¹ Peter Turner and Catherine O'Reilly²

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¹ The Vincent Wildlife Trust, 3&4 Bronsil Courtyard, Eastnor, Ledbury, Herefordshire HR8 1EP Tel: 01531 636441 Email: enquiries@vwt.org.uk Web: www.pinemarten.info/www.vwt.org.uk

² Waterford Institute of Technology, Cork Road, Waterford, Ireland. Tel: +353 051396049 Email: pturner@wit.ie Web: www.wit.ie

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Acknowledgements

1. Background

Until recently one of the most useful indicators of the presence of pine martens in an area was finding scats (droppings). Experienced field mammalogists were able to confirm the origin of the scat by shape, size, colour and odour. Kathy Velander used this method for her survey of Britain (Velander, 1983) and it was used again by McDonald et al., (1994) and Strachan et al., (1996) in surveys of England and Wales. In 1995 The Vincent Wildlife Trust (VWT) revived its interest in pine martens and explored a number of different methods of detecting the presence of martens, including scat surveys. During this period the Trust's experiences led to a growing scepticism about the reliability of the field identification of scats on the basis of physical attributes. By the end of the last century advances in the field of molecular biology made it possible (if somewhat expensive!) to extract DNA from scats and determine the species producing them. A study using DNA had already shown that otter, mink and polecat scats could be confused if using morphology alone (Hansen & Jacobsen, 1999). With this in mind, Johnny Birks hatched a project in which the best scat surveyors in Britain would collect scats from known marten areas in Scotland and the scats would be sent to the DNA laboratory for extraction and testing. The results showed that up to 29% of 'marten' scats collected were actually produced by foxes (Davison et al., 2002). When the study was extended to England and Wales (i.e. areas where the marten density was thought to be comparatively low) the misidentification rate increased to 97%. Clearly for areas with sparse marten populations this detection method was shown to be completely unsuitable. These results cast serious doubt on the results of the earlier scat surveys.



The Brechfa Forest survey team 2008

With DNA laboratory costs still too high for routine use in confirming the 'origins of faeces', using scat field surveys for detecting the presence of martens was put on hold. However, the Waterford Institute of Technology (WIT) began working on pine marten DNA and soon, largely as a result of the dedicated work of Catherine O'Reilly, the costs of this work fell to the point where testing of large batches of scats was financially feasible. Catherine and Peter Turner organised a very successful weekend marten workshop in June 2007, focussing on sparse marten populations and detection methods, which a number of English and Welsh marten workers attended. At this workshop Peter suggested that a way to make scat surveys work might be to undertake a 'blitzing' approach, where a large team of surveyors searched a site and collected all possible marten scats for testing in the DNA laboratory. Several of us took this idea away with thoughts of arranging a trial study as soon as possible.

2. Trial at Cwm Rheidol

It fell upon Kate Williamson of the Snowdonia Mammal Group to organise this trial study and Cwm Rheidol in mid Wales, where a high-quality marten sighting had been made recently, was selected as the study site. The summer of 2007 was exceptionally wet, which made the survival time of useable droppings in the field rather short. Nevertheless, on 26th June a group of 14 surveyors assembled at Cwm Rheidol to survey pre-determined transects using existing paths and tracks. The methodology used was refined subsequently but involved the surveyors collecting any scat found that could conceivably be from a pine marten. Details of the location of each scat and its situation were recorded. The scats were collected in labelled plastic bags. Only 25 scats were found and these were collected and sent to WIT for analysis. The results are displayed in Table 1. Although not obvious at the time, these results were unusual in that, not only was a marten scat identified, but fox scats accounted for only 8% of scats collected and 64% could not be determined. The marten scat was quite unusual in form, being a 'blob' of purple-coloured jelly, and was almost dismissed as not being worth collecting!

Species	Scats	% total
Pine marten	1	4.0
Fox	2	8.0
Dog	3	12.0
Pig/boar	1	4.0
Polecat	1	4.0
Hedgehog	1	4.0
ND (Not determined)	16	64.0
Total	25	100

Table 1: Scat DNA results for pilot survey in Cwm Rheidol, June 2007

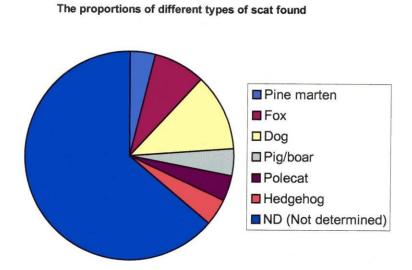


Figure 1: Scat DNA results for pilot survey in Cwm Rheidol, June 2007

It was perhaps fortunate (or perhaps unfortunate with the benefit of hindsight!) that one of these was shown to be from a pine marten of haplotype *a*, the type found in Scotland. Success at this early stage naturally suggested that this approach should be trialled in a more extensive programme of surveys in England and Wales.

3. Rolling out the survey

The trial provided valuable experience in how future scat surveys might be organised and how the methodology might be improved. However, it was clear that the next phase would still amount to a pilot study, and although we hoped that this approach would yield positive results, we knew that mainly we would be testing and refining this new approach to surveying pine martens.

4. Selection of sites

Because this project was aimed mainly at testing the technique, our intention was to select sites that would give the best chances of success. Therefore we focussed on those sites that had consistently produced high-quality pine marten sightings since 1995, when our recording began. A longlist of candidate sites was drawn up and from these the shortlist of 17 sites (8 in Wales and 9 in England) was produced. The criteria for this final selection included access arrangements and the availability of local volunteers.

Forest or Site	NGR (approx)
Claife Heights or Grizedale Forest	SD3798 / SD3394
Consall Wood/Churnet Valley	SJ9948
Ennerdale woodlands or Thirlmere	NY1513 / NY 3117
Gisburn Forest	SD7456
Hamsterley Forest	NZ0429
Kidland and Harbottle Forests	NT9304
North York Moors Woodlands	NZ5703
Upper Derwent Valley Woodlands	SK1689
Wark Forest	NY7573
Aberhirnant (Penllyn) Forest	SH9531
Brechfa Forest	SN5334
Clochaenog Forest	SJ0153
Crychan Forest	SN8440
Dyffryn Ffestiniog	SH6540
Dyfi Forest	SH7810
Gwydir Forest	SH7857
Hafren Forest	SN8588

Table 2: The original shortlist

This list was very much a working document and some changes took place by the time of the surveys. In the Lake District, we selected two pairs of sites intending only one from each pair would be surveyed. However, the enthusiasm of John Martin was such that all four sites were surveyed. The Derwent valley survey finally focussed on the mid-Derwent valley woodlands, in view of some very recent sightings being made. However, for various organisational reasons, the proposed survey of Gisburn Forest was not undertaken.

5. Methodology

5.1 Basic aims

This project was conceived as an exercise to evaluate and develop the scat survey/DNA testing survey method, not to produce results that could reveal absolute or relative marten population sizes. Consequently there was never an intention to ensure that a standardised 'search effort' was made at each site, which would have required more teams at the large sites compared with the smaller ones. The intention was to survey at least 50km of tracks and paths at each site and, based on prior experience, an average of 5km might be carefully surveyed by a team in five hours. On this basis 10 teams would be required, and with two as a minimum safe team size 20 surveyors would be needed.

5.2 Sites and sectors

Having selected the sites the first task was to define them. In many cases this was relatively simple, as the site comprised of a block of almost contiguous woodland, but

elsewhere this required careful consideration. Once the site had been delimited then survey sectors were defined, each being an area in which a single survey team would operate. These were created based on 1:25,000 scale OS maps or Forestry Commission (FC) maps if available. Maps were also provided by CCW for some Wales surveys and the Snowdonia National Park Authority (SNPA) for surveys in that area. Each sector was designed to have at least 5km of walkable tracks and paths, with as many circular routes as possible. Consideration was also given to ease of access and the avoidance of any identified hazards. The intention was also to avoid extensive areas of clear-fell, using information gathered either from a prior visit by VWT staff or from expert local knowledge.

5.3 Volunteers

Volunteer surveyors assembled at a prearranged location and were given a short introduction, briefed on the methodology and issued with all necessary equipment, including basic survival aids, recording forms and scat collecting kits. Health and safety, site safety matters and the emergency plan were explained and mobile 'phone numbers were swapped. At some of these meeting locations we benefited from the availability of an indoor facility where a short slide show could be given.

5.4 Field work

Survey teams selected the routes they would take prior to setting off, choosing circular routes wherever possible. The route(s) taken were marked clearly on the map of the sector that was provided. Teams were instructed to walk slowly along each route, searching for scats as they went and paying particular attention to track intersections, stream crossings, boulders etc. which might be preferred scatting points.

Any scats found that could not be dismissed as clearly from an animal other than a marten were collected after first having been photographed alongside a sequentiallynumbered collecting bag. This ensured that each photograph included a unique reference number as well as something for scale. All scat handling was undertaken using new disposable wooden spatulas for each specimen. Great care was taken to avoid cross-contamination. The finer details of the collecting technique appear in Appendix 1.



Scat and collecting bag photographed together

6. DNA lab methodology (Pete Turner)

DNA extracts were made and initially screened for fox and pine marten DNA using a real time PCR assay (O'Reilly *et al.*, 2007). In order to maximise the chances of success, several small samples were taken from various parts of the scat and pooled in the extract. Care was taken to avoid cross contamination from other samples processed in the lab; precautions included swabbing the bench area with dilute bleach, and using disposable forceps for sampling. Samples from other sources were never processed in the same batch. Scats were stored frozen.

One sample, from the pilot survey in Cwm Rheidol (see 2 above) was shown to contain pine marten DNA. This scat was re-extracted and the analysis repeated; the confirmed pine marten mitochondrial DNA was sequenced in order to determine the haplotype.

Samples not shown to be fox or pine marten but which were shown to contain significant levels of DNA were further analysed by DNA sequencing. The sequencing targeted two regions. Firstly a region of approximately 400 bps of the mitochondrial D-loop was amplified and sequenced using primers that would amplify a limited range

of mammals (e.g. mustelids, fox, dog, cat). Samples that failed with this assay were then tested using a ~400 bp region of the mitochondrial 16s RNA gene using more

generalised primers that will amplify a broader range of animals (e.g. mammals, birds, fish, amphibians). The rationale for this was primarily to identify samples that might originate from a *Martes americana* lineage. Several animals analysed from Northern England showed such a mitochondrial lineage (Kyle *et al.*, 2003), which is evidence of interbreeding between the two species at some point in their ancestry. This analysis also provided information on what other species had contributed samples to the survey.

7. Results

Overall the survey resulted in over 900km of tracks and paths being surveyed and 726 scats being collected. No scats were collected from which pine marten DNA was detected.

7.1 Combined results

The results from the whole survey (19 sites) have been consolidated and are presented in Tables 3 and 4 and Figure 2 below:

Number of surveyors	283
Number of teams	137
Total number of scats collected	726
Mean number of scats collected per team	5.3 (0 to 25)
Total distance surveyed (km)	901.2
Mean distance surveyed per team (km)	6.6 (2.1 to 12.5)
Mean scat density per survey (scats km ⁻¹)	0.75 (0.18 to 2.29)

Table 3: Combined surveying statistics

DNA result	Scats	% total
Fox	446	61.4
Bird	63	8.6
Mixed / misc	28	3.8
Dog	19	2.6
Deer	6	0.8
Polecat	4	0.5
Amphibian	3	0.4
Human	3	0.4
Sheep	3	0.4
Stoat	2	0.3
Otter	1	0.1
Hedgehog	1	0.1
Vole	1	0.1
ND (Not Determined)	146	20.5
Total	726	100.0

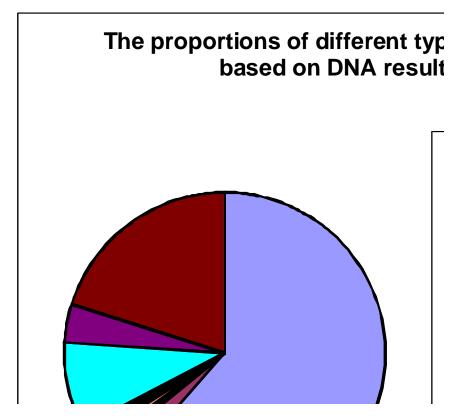


Figure 2: Combined DNA result statistics

The largest proportion of scats (61.4%) came from foxes, which was to be expected as foxes are relatively common, they scat on tracks and paths and the surveyors were urged to err of the side of caution by collecting any scats that could possibly be from a marten. The next largest group was 'not determined' (20.5%), which means that the laboratory was unable to extract DNA from them. In view of the comparatively wet weather experienced during both summers of the survey, this result might have been expected to be much worse. The greatest concentration of cells from the defecator is on the outside of the scats as they are collected during passage though the intestine. Old scats might lose these cells as a result of weathering.

7.2 Unexpected results

Of the remaining scat DNA results, some appear to be quite surprising. Birds featured quite strongly (8.6%) but weathered pheasant droppings and corvid pellets can be easily mistaken for carnivore scats. Perhaps the most curious results are those of vole and amphibian: could a surveyor really mistake a vole dropping for a marten scat? In these cases the faecal material clearly was not produced by the species whose DNA was extracted, and it seem likely that they represent prey remains in a scat from which all the predator cells had been washed out. This probably also accounts for the few scats from which only bacterial DNA was extracted.

A few scats were shown to contain mixed DNA. This could simply be contamination in the field or more likely, the detection of prey and predator DNA by the different sequencing assays (e.g otter and fish).



Dropping spotting

8. Discussion

8.1 Basic aims

Whilst the basic aims were not properly achieved at the majority of sites in terms of numbers of surveyor teams deployed, overall the discrepancy was small. The average distance surveyed by a team was 6.6km, slightly above the figure expected (5km), but the total number of teams deployed (137) fell significantly below target (190). Nevertheless the overall distance surveyed was 901.2km, only slightly short of the anticipated 950km (19x50km). All things considered these seemed to be acceptable results, showing that the basic aims were reasonably realistic.

Despite this, the level of survey effort did vary considerably from site to site, from the single team that surveyed the Staffordshire woodlands to the 30 surveyors in 12 teams that surveyed Wark Forest. However, the Staffordshire woodlands amounted to a comparatively small area whilst Wark Forest, really the southern end of the massive Kielder Forest, was massive and the survey felt rather like 'scratching the surface'. For some surveys (e.g. Hafren Forest), the survey sectors covered virtually all of the

site resulting in all parts being surveyed. Naturally it would have been ideal if this could have been true everywhere, but to achieve this would have meant assembling unrealistically large numbers of surveyors or, alternatively, excluding the larger sites from the survey. The intention was always to undertake all surveying on a single day, largely for logistical and health and safety reasons. Also past experience suggested that volunteer support declines after the first day.

8.2 Site selection

The selection of sites that had consistently produced good numbers of relatively recent, high-scoring sightings reports seemed at the time to be a good strategy. However, as the survey progressed and no marten scats were found, we began to consider the differences between the pilot study site in Cwm Rheidol and the rest of the sites. One clear difference exists, in that in addition to meeting all the basic criteria, Cwm Rheidol was the site of a high-scoring marten sighting only two weeks earlier. In this respect the survey there could be considered a 'hot pursuit' response to this sighting. As far as we are aware this was not the case elsewhere. Outside the scope of this report, but relevant here, are the results of scat surveys undertaken at Cwm Rheidol in 2008 and 2009, using the same survey routes used in the 2007 pilot survey. Neither of these resulted in a pine marten scat being collected and no recent sightings had been reported prior to either survey.

8.3 Organising and operating the survey

8.3.1 Sites and sectors

Some survey sites were 'modified' slightly after initial selection. Usually this involved including additional nearby blocks of forestry where new information had come to light, and / or when it was clear that sufficient surveyors would be available to cover more ground. Some sectors were redesignated when it was discovered that they had been recently clear-felled.

8.3.2 Site owners

Many of the survey sites selected are owned by the FC, the staff of which were most helpful in granting permits to operate the survey. The National Trust (NT), United Utilities (UU), the North Yorkshire Moors National Park Authority (NYMNPA) and the SNPA were also very accommodating. In some areas there was an understandable concern that highlighting the presence of pine martens in sites adjacent to game bird rearing estates might create difficulties with external relations, but this was never presented as an obstacle to the survey.

8.3.3 Volunteer surveyors

From the outset it was clear that assembling a large team of experienced volunteer surveyors might be a challenge in some of the more rural parts of England and Wales. Collaboration with local organisations such as mammal groups and the Wildlife Trusts was vital, and without this help many surveys would never have been undertaken. Despite the excellent support we received, of the 18 sites surveyed, at only four were the target numbers of surveyors and teams achieved (See Table 5). On reflection the numbers of volunteers that did attend the surveys was very good, bearing in mind the

nature of the task set i.e. walking all day with eyes fixed on the ground, and also collecting faeces.

Site	Surveyors	Teams
Aberhirnant	10	7
Brechfa Forest	21	11
Claife Heights	8	4
Clocaenog Forest	24	10
Crychan and Halfway	17	10
Forests		
Derwent Valley	14	6
Dyffryn Ffestiniog	19	9
Dyfi Forest	19	9
Ennerdale	12	6
Grizedale Forest	7	5
Gwydir Forest	18	9
Hafren Forest	16	9
Hamsterley Forest	19	8
Kidland and Harbottle	17	8
Forests		
North Yorkshire Moors	24	12
Forests		
Staffordshire Woodlands	3	1
Thirlmere Forest	4	2
Wark Forest	30	12

Table 5: Surveyors and survey teams for each survey site

Whilst it would have been ideal to have access to a team of 'crack' marten scat hunters for the survey work, we knew that some of our volunteers would have had little previous experience. Some basic guidance was given before each survey, including the opportunity to look at photographs showing a range of confirmed marten scats. Despite the limited experience of some surveyors, visual inspections of the material collected seemed to suggest that everyone had been 'on the right track'. Many had never undertaken scat surveys before and some had little experience of working with maps, but without exception every team produced useable results. We were tremendously impressed!



The Wark Forest survey team

8.3.4 Operating the survey

Whilst the methodology appeared to work as well as expected, the time and effort required in organising it exceeded expectations. Simple matters such as obtaining and preparing suitable maps for the survey, identifying the survey sectors and even running the survey on the day, were often problematic. Often reconnaissance visits by VWT staff were not possible and, whilst key local contacts were excellent, there is no substitute for the survey leader having first-hand experience of the site. With 20 surveyors arriving back at the meeting point at roughly the same time, many wanting not to have to wait around, it was a challenge to check and process all the paperwork and scats and collect all the photographs. Quickly it became apparent that three people were needed to process everything speedily and accurately. Nevertheless, a degree of duplication in the recording meant that, even though sometimes information was not recorded, a cross reference could resolve the issue.

8.3.5 Extrinsic factors

There were some factors that affected / might have affected the survey over which there was little or no control. The weather was one of these. The summer of 2008 was exceptionally wet, which not only would have affected the survival of scats but could have had impacted on the amount of excreted DNA present. The summer of 2009 was dryer but still with much rain. Personal experience suggests that surveying in the rain is less effective, as scats seem to be less obvious when wet and the rain can affect the functioning of the surveyor (e.g. rain on spectacles). The other major

factor is the presence of scats. An assumption is made that pine martens will scat on tracks and paths to mark territories. This is the received wisdom from Scotland and Ireland, but does it apply in England and Wales, where marten territories may not be contiguous? An intensive study undertaken in a wood in Ireland showed that martens scat away from tracks as well as on them (P. Turner pers. comm.). However, various constraints, especially health and safety issues, ruled out any off-track surveying. It seems possible that confining the survey to tracks and paths would have the effect of reducing the number of scats found in any given sector, without completely compromising the protocol. However, it is possible (if not particularly plausible) that martens might avoid scatting on tracks and paths in England and Wales and this would have a major impact on the effectiveness of the approach used.

9. Conclusions

The possibility that this survey would result in no pine marten scats being identified had been considered at the outset, and so the result came as no great surprise but clearly something of a disappointment. This method has been shown to work in Ireland and promised to solve the problem of the field identification of scats. This work had to be undertaken as it promised the best chance of finding a reliable field survey method for pine martens.

Although it is anecdotal evidence, the success of the pilot study was based on a single scat at a site where there had been a recent high-quality marten sighting. The difference between one scat and none is as small as it can be! Hindsight, as the saying goes, is a wonderful thing, and perhaps it would have been wiser to have attempted further pilot studies at conveniently-located sites to see if this result could be repeated or even improved on. Also, although not appreciated at the time, the recent sighting at the pilot study site could have been very significant in influencing the outcome of that survey.

Although some of the early stages could have been improved on, the project was very successful in that it confirmed that DNA testing scats collected in the field is a viable method, it gave valuable experience to the Trust and the volunteers in undertaking this type of work, and it pointed to new ways of applying the methodology in the future.

10. Suggestions for future work

10.1 'Hot pursuit'

It would be a valuable exercise to use this approach at sites where recent sightings had been made. It is possible that these 'hot-spot' areas in England and Wales are not permanently occupied by martens and so this approach could increase the chance of finding a marten scat. This would need a team of volunteer surveyors that could be mustered at relatively short notice.

10.2 Low intensity ongoing surveys

Something that the VWT is already encouraging and supporting is regular surveys by individuals or small groups of volunteers. These would best suit volunteers who live within a short distance of a hot spot, and who might visit the area regularly anyway. This approach, in the longer term, could result in a substantial survey effort. There may also be benefits in conducting surveys at different times of the year. An advantage of this approach is that it requires little organising and gives the surveyor(s) a high degree of flexibility.

10.3 Locally organised surveys

The complications of organising and running such surveys sometimes hundreds of miles away from the Trust's offices are not to be underestimated. The Trust had always hoped that this project would lead to interest in running scat-DNA surveys locally, and this appears to be by far the best way of proceeding. Organised and run by local mammal groups or the Wildlife Trusts, with support from the VWT, the 'hot pursuit' and ongoing survey approaches need to be evaluated.

If any individual or group would like assistance in setting up a local survey please contact the Trust at the address at the front of this report.

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Appendices

Appendix 1- Scat Surveys

North Wales

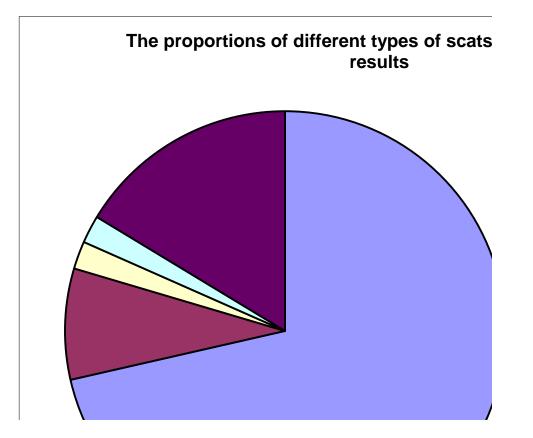
1.1 Aberhirnant Survey - 1st August 2009

Aberhirnant woodland is a predominantly coniferous upland plantation to the east of Bala. It comprises part of Penllyn forest and amounts to 1,900 ha in total. Survey efforts were concentrated in the western area of the forest.

Number of surveyors	10
Number of teams	7
Total number of scats collected	49
Scats collected per team	3 to 12
Total distance surveyed (km)	57.2
Distances surveyed per team (km)	4.6 to 11.7
Overall scat density (scats km ⁻¹)	0.82

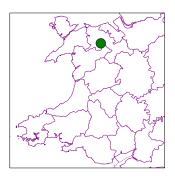
DNA	Scats	%
Fox	35	71.4
Waterfowl/Bird	4	8.2
Mixed	1	2
Bacteria	1	2
ND (Not Determined)	8	16.4
Total	49	100





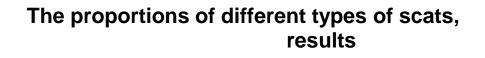
1.2 Clocaenog Forest Survey - 12th April 2008

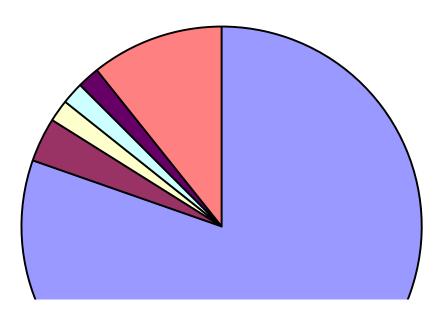
Clocaenog Forest comprises of a number of contiguous conifer blocks amounting to about 6000ha in total. It has produced many good-quality sightings reports in recent years. This was the first of the surveys and was undertaken slightly early in the season and before the official start date of 1st May, to fit in with the availability of surveyors. Snow fell early in that day and hindered surveying during the morning. The results include a high proportion of fox scats but the collection of some mustelid scats is encouraging.



Number of surveyors	24
Number of teams	10
Total number of scats collected	56
Scats collected per team	2 to 10
Total distance surveyed (km)	58.5
Distances surveyed per team (km)	2.2 to 9.2
Overall scat density (scats km ⁻¹)	0.96

DNA result	No. of scats	%
Fox	45	80.3
Polecat	2	3.6
Otter	1	1.8
Otter or Stoat	1	1.8
Dog	1	1.8
ND (Not Determined)	6	10.7
TOTAL	56	100.0





1.3 Crychan and Halfway Forests Survey - 18th July 2009

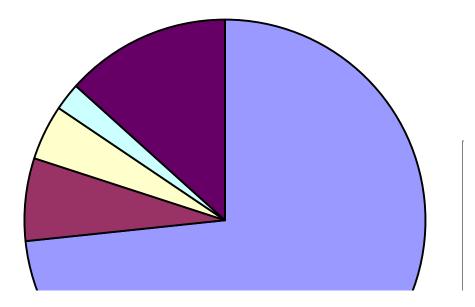
These two forestry blocks are almost contiguous and fall on the Powys/Carmarthenshire border. They have both produced good-quality sightings over many years. Mynydd Epynt, a military training area, adjoins Crychan Forest and is a haven for wildlife. The survey covered significant areas of both forests but little of interest was found.



17
10
45
2 to 9
53.1
4.3 to 10.0
0.85

Scats	% total
33	73.3
3	6.8
2	4.4
1	2.2
6	13.3
45	100
	33 3 2 1 6

The proportions of different types of scats, results

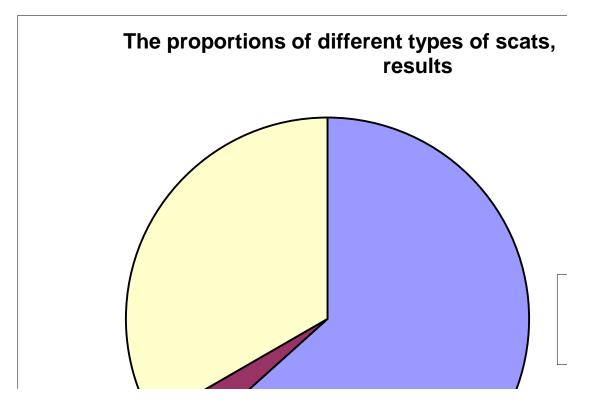


1.4 Dyffryn Ffestiniog Survey - 23rd August 2008

The survey site comprises of a number of forestry plantations on the north side of the valley, between Blaenau Ffestiniog and Penrhyndeudraeth. There is also extensive broadleaf woodland in this area, and the total area of tree cover probably exceeds 15,000ha.

Number of surveyors	19
Number of teams	9
Total number of scats collected	30
Scats collected per team	1 to 6
Total distance surveyed (km)	44.7
Distances surveyed per team (km)	3.4 to 8.7
Overall scat density (scats km ⁻¹)	0.67

DNA result	Scats	% total
Fox	19	63.3
Bird	1	3.4
ND (Not determined)	10	33.3
Total	30	100





1.5 Dyfi Forest Survey - 22nd August 2009

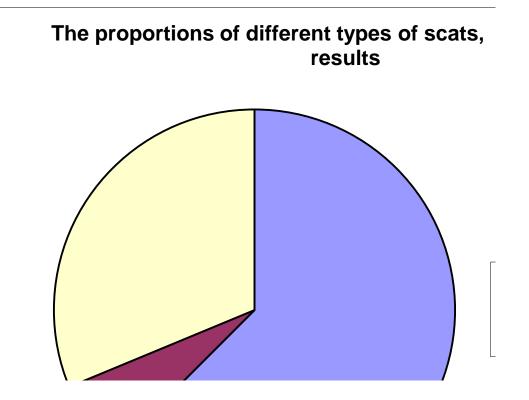
Dyfi Forest is an extensive area of forestry, encompassing 6,000 ha, to the south-east of Cader Idris. Despite having a good number of surveyors, the size of the forest meant that only a limited area was surveyed and efforts were focused on the western area of the forest. The overall scat density was very low, possibly a result of the recent forestry works that had occurred in much of the forest.



Number of surveyors	19
Number of teams	9
Total number of scats collected	16
Scats collected per team	0 to 6
Total distance surveyed (km)	88.9
Distances surveyed per team (km)	5 to 13
Overall scat density (scats km ⁻¹)	0.18

DNA result	Scats	% total
Fox	10	62.5
Corvus spp.*	1	6.2
ND (Not Determined)	5	31.3
Total	16	100

* Corvus spp. is the crow family



1.6 Gwydir Forest Survey - 20thJune 2009

Gwydir Forest is a large area of forestry that extends over 7,250 ha. A scat, confirmed by DNA analysis to be pine marten, was found in the forest in 1996. Since then, the area has produced many good quality sightings reports. Due to its size, we were unable to cover the whole forest and our efforts were mainly focused in the northern section of the forest, to the north of Betws-y-Coed, where most sightings have been made.

Number of surveyors	18
Number of teams	9
Total number of scats collected	26
Scats collected per team	0 to 5
Total distance surveyed (km)	56.6
Distances surveyed per team (km)	2.9 to 9.4
Overall scat density (scats km ⁻¹)	0.46

DNA result	Scats	% total
Fox	16	61.5
Waterfowl	5	19.2
Bird	2	7.7
ND (Not determined)	3	11.6
Total	26	100



The proportions of different types of scats, results

Mid Wales 1.7 Hafren Forest Survey - 1st June 2008

The main concentration of recent sightings falls slightly to the west of Hafren Forest in an area that is poorly defined. Good quality sightings have been obtained from the forest and so, for operational reasons, this was selected as the survey site. The weather was fine and sunny and a good number of scats were found.

Number of surveyors	16
Number of teams	9
Total number of scats collected	56
Scats collected per team	2 to 10
Total distance surveyed (km)	70.9
Distances surveyed per team (km)	5.5 to 11.8
Overall scat density (scats km ⁻¹)	0.79

DNA result	Scats	% total
Fox	39	69.6
Polecat	2	3.6
Dog	1	1.8
Dog?	1	1.8
Sheep	1	1.8
ND (Not Determined)	12	21.4
Total	56	100



The proportions of different types of scats, results

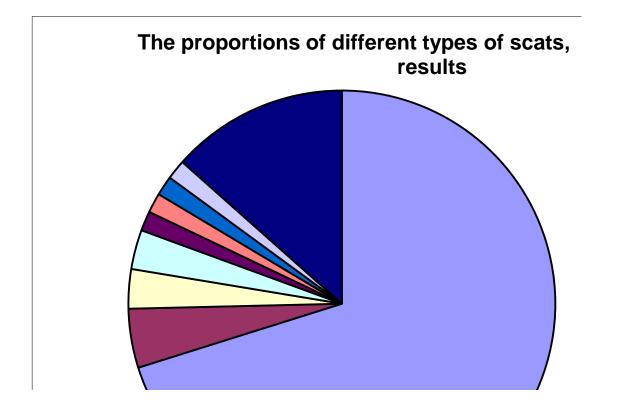
South Wales 1.8 Brechfa Forest Survey - 28th June 2008

The Brechfa Forest area produced some excellent quality sightings in the late 1990s but more recently extensive forestry operations have had a big impact on the character of some parts of the forest. Also the provision of off-road cycleways has resulted in a noticeable increase in visitors of late. The area has extensive broadleaf woodland, which may contribute to the pine marten interest.



Number of surveyors	21
Number of teams	11
Total number of scats collected	67
Scats collected per team	0 to 13
Total distance surveyed (km)	78
Distances surveyed per team (km)	3.6 to 10.2
Overall scat density (scats km ⁻¹)	0.86

Number	%
47	70.1
3	4.5
2	3.0
2	3.0
1	1.5
1	1.5
1	1.5
1	1.5
9	13.4
67	100.0
	47 3 2 2 1 1 1 1 1 9



England

Midlands

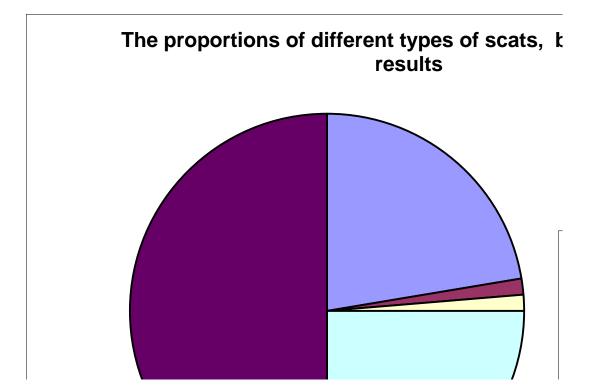
1.9 Derwent Valley Survey - 30th August 2009

This survey was undertaken in the small blocks of mixed woodland in the mid Derwent Valley, running from Ambergate to Cromford, following the River Derwent. These woodlands included Crich Chase and Shining Cliff Woods; the location of a two particularly good quality sightings in the mid-1990s. Two sites north of Matlock were also surveyed. The majority of the areas surveyed were open access and thus fairly busy with walkers. Half of the scats collected were not determined; a rather high percentage compared to other surveys.



Number of surveyors	14
Number of teams	6
Total number of scats collected	38
Scats collected per team	1 to 16
Total distance surveyed (km)	39.7
Distances surveyed per team (km)	2.8 to 6.8
Overall scat density (scats km ⁻¹)	1.0

DNA result	Scats	% total
Fox	17	44.8
Dog	1	2.6
Mixed	1	2.6
ND (Not Determined)	19	50.0
Total	38	100.0



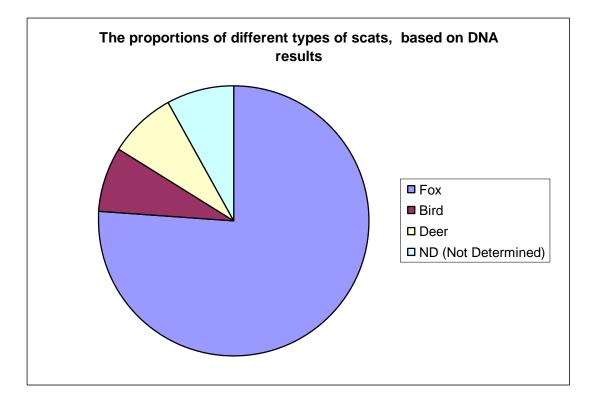
1.10 Staffordshire Woodlands Survey - 13th August 2008

This survey was undertaken in north Staffordshire, covering Consall wood in the Churnet Valley and Gib Torr. These areas have produced fairly recent pine marten records; firstly, a photo taken of a pine marten in a tree at Consall wood in 2004 and a scat, confirmed by DNA analysis to be pine marten, found at Gib Torr following a good quality sighting in 2001. The efforts of the survey were somewhat limited by only three surveyors and one team, the originally-planned survey in July having been postponed due to heavy rain.



Number of surveyors	3
Number of teams	1
Total number of scats collected	25
Scats collected per team	25
Total distance surveyed (km)	10.5
Distances surveyed per team (km)	2.8 to 7.8
Overall scat density (scats km ⁻¹)	2.29

DNA result	Scats	% total
Fox	19	76.0
Bird	2	8.0
Deer	2	8.0
ND (Not Determined)	2	8.0
Total	25	100.0



North-east England

1.11 Hamsterley Forest Survey - 8th August 2009

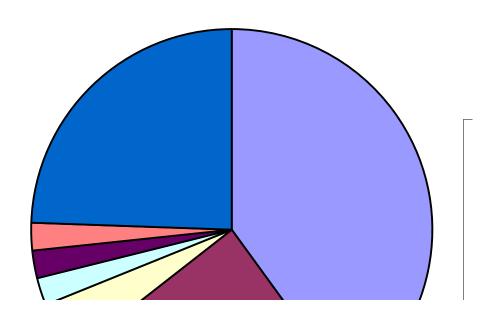
Hamsterley Forest is a c2,000ha site of mixed woodland on the edge of the North Pennines Area of Outstanding Natural Beauty. The forest is a popular visitor attraction and the tracks and trails are heavily used by walkers, cyclists and horse riders. This is probably the reason why in some areas of the forest scats were sparse. We were lucky to have a good number of surveyors and dry weather on the day of the survey and during the week leading up to it. The unusually high number of pheasant scats and no fox scats is possibly a result of the widespread game-keeping activity in the area. See Section 7.2 for information on pheasant droppings



Number of surveyors	19
Number of teams	8
Total number of scats collected	45
Scats collected per team	1 to 15
Total distance surveyed (km)	58.5
Distances surveyed per team (km)	5.0 to 10.4
Overall scat density (scats km ⁻¹)	0.75

DNA result	Scats	% total
Pheasant	18	40.0
Mixed	11	24.5
Hedgehog	2	4.4
Stoat	1	2.2
Roe Deer	1	2.2
Bacteria	1	2.2
ND (Not Determined)	11	24.5
Total	45	100

The proportions of different types of scats, results



1.12 Kidland and Harbottle Forests Survey - 8th October 2008

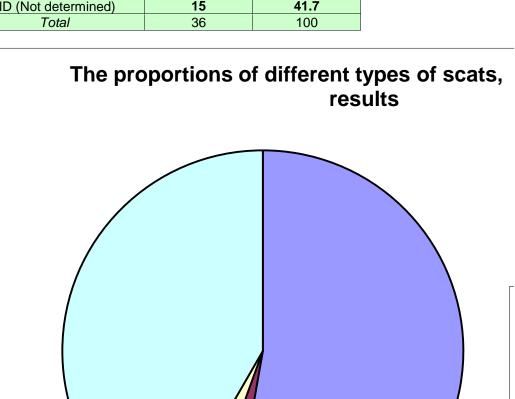
Kidland and Harbottle Forests are forestry plantations covering c2,000ha in the Cheviot Hills. A part of the Kielder Forest district, this area has produced many good quality sightings reports over the past 15 years. The survey was undertaken a little later in the year than desired, which may have resulted in more difficult surveying conditions, as the visibility of scats on the ground can be reduced by leaf-fall.

Number of surveyors	18
Number of teams	7
Total number of scats collected	36
Scats collected per team	2 to 9
Total distance surveyed (km)	36*
Distances surveyed per team (km)	3.2 to 9.2*
Overall scat density (scats km ⁻¹)	1.0**

* excludes the survey distance of one team whose route was not recorded

** as a result of the above this figure will appear to be higher

DNA result	Scats	% total
Fox	19	52.7
Dog	1	2.8
Dog Stoat	1	2.8
ND (Not determined)	15	41.7
Total	36	100



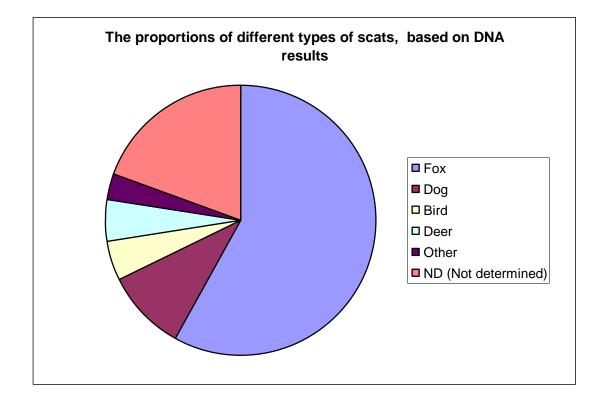


1.13 Wark Forest Survey - 9th July 2008

Wark Forest is the southern end of Kielder Forest, which total covers about 65,000ha and is the largest forest in England. Wark Forest includes wetlands areas managed Northumberland Wildlife Trust. This survey was largely organised by Kevin O'Hara who enlisted the help of his conservation volunteers.

Number of surveyors	30
Number of teams	12
Total number of scats collected	62
Scats collected per team	2 to 11
Total distance surveyed (km)	64.8
Distances surveyed per team (km)	2.1 to 8.0
Overall scat density (scats km ⁻¹)	0.96

DNA result	Scats	% total
Fox	36	58.1
Dog	6	9.7
Bird	3	4.8
Deer	3	4.8
Other	2	3.2
ND (Not determined)	12	19.3
Total	62	100

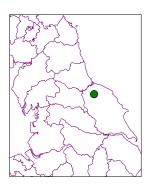




in by the

1.14 Yorkshire Moors Forests Survey - 15th August 2009

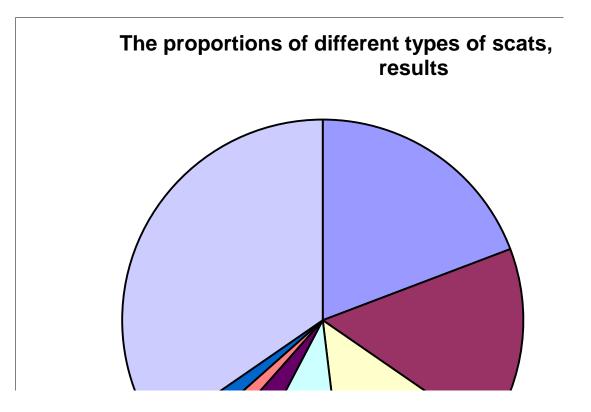
This survey was initially based on Broughton and Greenhow Plantations, but recent good quality sightings in some of the plantations further south, combined with a large and very enthusiastic team of volunteers, allowed for some of these to be visited. The low number of fox scats was unexpected. Over one third of the scats returned negative results, perhaps because of the wet weather prior (and during!) the survey. This area is possibly the top 'hot-spot' for sightings in England at present.



Number of surveyors	24
Number of teams	12
Total number of scats collected	52
Scats collected per team	1 to 11
Total distance surveyed (km)	66.9
Distances surveyed per team (km)	2.9 to 10.3
Overall scat density (scats km ⁻¹)	0.77

DNA result	Scats	% total
Fox	10	19.2
Pheasant	8	15.4
Dog	7	13.5
Bacteria	5	9.6
Waterfowl	2	3.8
Grouse	1	1.9
Corvus*	1	1.9
ND (Not Determined)	18	34.7
Total	52	100

* Corvus means a member of the crow family



North-west England

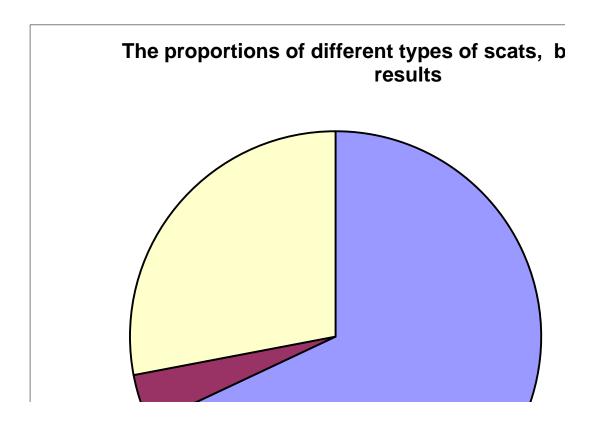
1.15 Claife Heights Survey - 17th August 2008

Claife Heights is an area of oak woodland on the western edge of Lake Windermere and it is linked to the north-eastern edge of Grizedale Forest. As Claife Heights is a comparatively small area, most of the woodland was surveyed even though there was a below average number of surveyors.

Number of surveyors	8
Number of teams	4
Total number of scats collected	25
Scats collected per team	0 to 14
Total distance surveyed (km)	31
Distances surveyed per team (km)	5 to 8
Overall scat density (scats km ⁻¹)	0.77



DNA	Scats	% total
Fox	17	68.0
Dog	1	4.0
ND (Not Determined)	7	28.0
Total	25	100

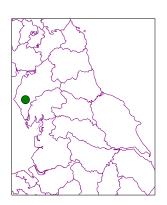


1.16 Ennerdale Forest Survey - 2nd August 2008

The Ennerdale Valley is situated on the north-western side of the Lake District National Park. The Valley extends to an area of 4,711 ha in its entirety. The remote mainly coniferous woodlands surrounding Ennerdale Water benefit from fewer visitors than other survey sites in the National Park, such as Grizedale Forest. For a discussion on the vole DNA result see Section 7.2.

Number of surveyors	12
Number of teams	6
Total number of scats collected	59
Scats collected per team	3 to 21
Total distance surveyed (km)	29.3
Distances surveyed per team (km)	2.8 to 6.0
Overall scat density (scats km ⁻¹)	0.5

DNA result	Scats	% total
Fox	50	84.7
Bird	3	5.1
Sheep	1	1.7
Deer	1	1.7
Vole	1	1.7
ND (Not determined)	3	5.1
Total	59	100



The proportions of different types of scats, results

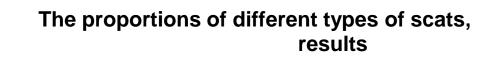
1.17 Grizedale Forest Survey – 16th August 2008

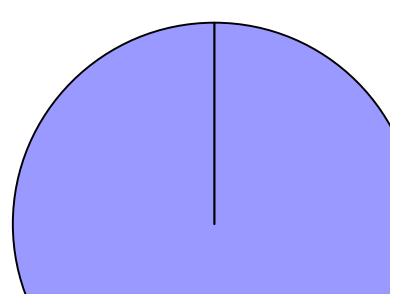
Grizedale Forest is the largest forest in the Lake District, comprising 2,447 ha of woodland in total. This area has produced the highest number of good quality sightings out of any of the survey sites in the Lake District. Due to the size of the forest and the limited number of surveyors, only the eastern and central areas of the forest were surveyed. This was the only survey that produced only fox scats. The forest is a popular tourist attraction and the forest tracks and trails are heavily used by walkers and cyclists, which may have influenced the result.



Number of surveyors	7
Number of teams	5
Total number of scats collected	29
Scats collected per team	3 to 10
Total distance surveyed (km)	38.5
Distances surveyed per team (km)	3.5 to 12.5
Overall scat density (scats km ⁻¹)	1.01

DNA result	Scats	% total
Fox	29	100
Total	29	100



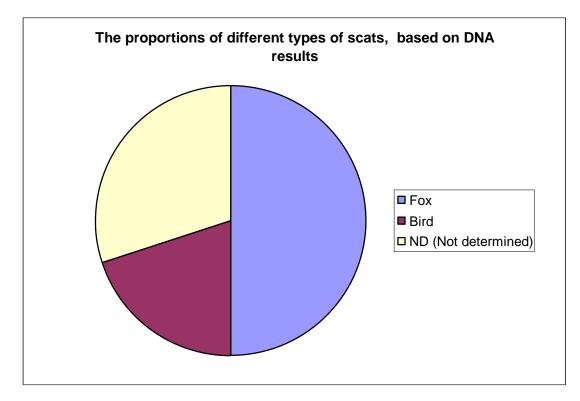


1.18 Thirlmere Forest Survey - 3rd August 2008

The woods surrounding Thirlmere reservoir comprise c4,800 ha of mostly coniferous forest. The area covered during the survey was limited as there were only two teams of surveyors. As a result, only the woodlands on the western edge of the reservoir were surveyed.

4
2
10
4 to 6
18.1
8.8 to 9.3
0.55

DNA result	Scats	% total
Fox	5	50.0
Bird	2	20.0
ND (Not determined)	3	30.0
Total	10	100





The VWT / WIT marten scat survey

Guide for Surveyors

Before starting any fieldwork please ensure that:

- You have read and understood the risk assessment and safety statement for this work.
- Each of you has completed the Surveyor Contact sheet.
- You understand the arrangements made for reassembling at the end of the survey.

Scats: Recent work has shown that not all classical pine marten scats, as identified in the field, originate from pine martens. More recent work has shown that scats whose appearance, smell etc are nothing like classical pine marten scats can be from pine martens. This can make it difficult to know what to collect. Any scats that are not obviously too big to have been produced by a marten should be collected. Some scats can be very soft and almost amorphous, and it might not be clear that they originate from a mammal, but these should also be collected. If there is any doubt then the scat should be collected. Some photographs of various scats should be available on the day, but please remember that these are just examples.

Searching: Specific tracks have not been identified in advance so the team should identify those that they intend to survey within their assigned location. Circular routes make the best use of the time. The length of the route that can be covered by one team in one day will vary according to the nature of the transect, but previous work indicates that between 4 and 7 km might be possible in a day. If you find yourself covering much more than this then you might not be searching as thoroughly as the survey requires. Similarly, if you cover much less ground then you might not be searching your sector as fully as the survey requires. Each team should mark the survey transects clearly on the *numbered* map provided (other maps ma be provided for general use but please use only the numbered map for recording). This should be done with a solid line drawn with a ballpoint pen. Make sure that the start and end points are clearly identifiable.

Where to search: Try to undertake as much of the survey on tracks and paths that have trees on at least one side, as past experience has shown that martens prefer the safety of nearby tree cover. In some survey sectors, more open areas will have to be surveyed. As many of the tracks you will be searching will be quite wide it is most effective if you have at least one surveyor on each side of the track. The surveyors should walk the transect slowly enough to be able to examine all parts of the track, especially the edges, any raised spots such as rocks, ant hills, tree stumps of fallen trees and any intersections with other linear features such as other tracks, streams, walls and fences. Stream or river intersections have been particularly productive in previous surveys. Where such intersections occur the surveyors should examine the intersectioning feature for some distance either side, as these areas can be good

scatting points. Please remember that we are still learning; don't use these guidelines as a rulebook.

Collecting: The scat should be photographed *in situ* with the collecting bag to be used alongside. Each bag carries a unique number, which should prevent confusion later, and it will also provide a scale. These photographs help to build a database of the morphological variation of scats. Once photographed, the location details, NGR, date and surveyors names should be written on the bag with a ballpoint *pen*. Then, using fresh disposable wooden spatulas, the scat should be placed in the bag and the bag sealed. Great care must be taken to avoid the scat coming into contact with the outside of the bag. The bag should then be placed in the rigid container provided to avoid any crushing in the field. This container should be used only for collected scats. Do not put anything else in it. In the past some surveyors have put clean collecting bags, spatulas etc in the scat container, which damages the scats and risks contamination. Cross-contamination is the enemy of DNA work, especially now that the laboratory methods are extremely sensitive. **DO NOT** use fingers to pick up scats, as you will then contaminate scats found subsequently with earlier DNA. Use fresh spatulas for every scat. Carefully place the used spatulas in the 'USED' bag provided and reseal. Finally, mark the location of the scat on the map with a clear dot labelled with the unique number and record the scat on the survey form along with supporting information. *Please use a pen for the recording form* and make sure that the NGR is written very clearly. We already have a number of forms where the NGRs are illegible, smudged, pencil marks! This might seem like a lot of unnecessary 'fussing' and a lot of duplication, but it is designed to minimise the risk of important information being lost or compromised.

Scats, recorded information and photographs: At the end of the survey all scats, forms and maps should be checked to ensure no details are missing before being handed to the co-ordinator. Scat photographs should be collected at the end of the survey and a laptop with a card reader, USB cables etc will be available when possible. Failing that please email your photographs to: jmessenger@vwt.org.uk

Timings: Please adhere to the timings given, especially returning to the rendezvous point or survey base, so that the rescue plan is not set in motion needlessly.

Although the final report is not scheduled for completion until June 2010, summary results of each local survey will be distributed as soon as they are available. Please ensure that you complete the 'Personal Details' form so that you are included.

Survey Team Sheet

Forest or site name	Date

Team No.

Surveyors' names: 1

(please give full names) **2**

Map number Number of scats collected (Complete table below)

Scat records:

NGR	ID No.	Photo?	Notes

VWT use only: Distance surveyedkm Scat photos received Y/N

The VWT/WIT Pine Marten Scat Survey

Leaders' Notes

There are two very important aspects of this fieldwork, the welfare of the surveyors and the correct operation of the survey.

Welfare of the volunteer surveyors

This is paramount. The volunteers should receive (by email if possible) copies of the fours documents (Health & Safety Guide for Volunteers, Surveyor's Details form, Surveyors Guide ands Survey Team Sheet) in advance of the survey and be asked to read them all. They should be asked to bring completed and signed copies of the H&S Guide and the Surveyor's Details form with them to the survey. Please ensure that you have spare copies of these as some may well turn up without them! All other paperwork can be supplied on the day if they haven't made copies. A reminder about bringing appropriate clothing etc might also be advisable at this stage.

On the day ensure that these forms are collected. Get each volunteer team to write down the mobile phone numbers and networks of the leader and all other surveyors if possible. This might be achieved by reading them out from the forms. If you are aware of any specific hazards at that site please make the volunteers aware of this. Please check the local weather forecast for the day and advise them of this. If you have any doubts about the fitness of a volunteer to undertake the work please be diplomatic but, if you remain unconvinced then it is up to you to ask them to withdraw. Similarly if any volunteer presents themselves with clothing/footwear inappropriate for the terrain/weather forecast then ultimately you might have to consider asking them to withdraw. Good judgement here is vital. Remember we cannot eliminate risks, only reduce them by good planning.

Record the names of those in each team, there approximate starting positions and the area in which they should be operating

The optimum number of people in a team is two. Lone working should be avoided unless the numbers of surveyors available is small AND there are volunteers present who are experience at this type of work and 'robust' enough to cope on their own. Only those who offer to work alone should be considered and avoid selecting those from the more vulnerable groups. These might include older volunteers, young women etc, but your judgement will be needed here.

Ensuring that the survey is carried out correctly

This is vital if we are to get useful results from it. You will need to ensure that the surveyors understand how to survey and collect the scats, and that they should collect *any* material that cannot be confidently dismissed as non-marten. Avoid putting inexperienced surveyors together as a team. Please ask the volunteers, in advance, to bring and clipboard and a digital camera if they have one. Correct use of the paper work is vital if we are to avoid having scat material that is not properly documented and traceable. Please stress the need to be rigorous in this matter. There is some duplication in the paperwork but this is designed to minimise the risk of important information being lost. Scats should be photographed in situ is possible, but we accept that for various reasons this might not be possible. Please arrange for photographs to be collected after the survey. These can be emailed directly to me after the survey (jmessenger@vwt.org.uk). Please stress the need to avoid contamination, by using fresh spatulas for colleting each sample, ensuring that the sample bags are properly sealed and that all samples are placed in the crush-proof box supplied. Also ensure that each team

has a map copy appropriate for the area in which they will be searching. If your knowledge of the survey site is good then these can be prepared easily beforehand. If not then try to ensure that the area is covered evenly by the teams. Please discuss this with me in advance if there are any problems.

Emergency plan

Please study this and make any alterations/additions appropriate to the survey site.

'Scatalogue'

Below is a photo record of some of the 'scats' found during the survey period. The photos show the remarkable similarities between the scats or droppings of a range of species.

Mammals

Pine marten



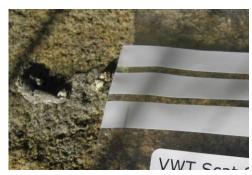
Polecat



Otter



Stoat



Fox













Hedgehog



Deer





Dog



Sheep



Birds

Pheasant



Corvus (crows and ravens)



Phoenicopterus (water fowl)









Acknowledgements

Kate Williamson, Rob Strachan and the rest of the Snowdonia Mammal Group, without whom the survey would have struggled in North and mid Wales. Kate, in particular, made a huge input into the design of this survey and continued to provide valuable ideas throughout. John Martin, for his enormous energy and enthusiasm which resulted in the survey visiting twice as many sites in the Lake District as first planned. Johnny Birks, for his valuable input in the early planning stages of the project. Kevin O'Hara for mobilising his volunteers at the Northumberland Wildlife Trust and for running two surveys in the north-east.

Others who contributed beyond the call of duty include Chris Hall, Tony Braithewaite, Mick Douche, Brian Walker, Karen McArthur (Durham Wildlife Trust), Dave Mallon (Derbyshire Mammal Group) and Derek Crawley (Staffordshire Mammal Group).

We are indebted to Natural England, the Countryside Council for Wales and the SITA Trust for financial assistance with this work.

We are also greatly indebted to all those volunteer surveyors who gave up a day of their time to walk all day looking only at the ground! Without you we simply could not have undertaken this work. Their names are listed below. We apologise if you were there and your name is not listed. Please let us know and we will correct the list and update this version.

VWT pine marten scat survey surveyors 2008-09:

Chris Addy Mike Ashford David Atkinson **Pauline Barber** Paul Beard Chris Berry Joanne Bissell Ian Blacklock Ian Bond Tony Braithwaite John Breffit Adam Britt Emma Broad Julia Brown David Brown Brian Burnett Susan Burnett Anne Butler Natalie Buttriss Simon Cahill **Derek Capes** John Carss Mike Castle Roger Chappell Rona Charles Sue Charlton Bekka Close Mary-Anne Collis Brian Copland Emma Cox Grace Crabb Dana Cracknell Derek Crawley **Richard Crompton** Shirley Cross **Rob Davies** Huw Denman **Geoff Dobbins** Mick Douch Joanna Dulat Sam Dver Ethan Earnshaw Mehdi El-Radhi Carol Emmons M.C. Flechard **Emily Foot** Ali Forbes Nick Forkin Zoe Foster Sara Frisby Chris Fry Frances Gillett Joyce Gillies

Laura Grant Mike Green Janice Gwilliam Rhys Gwynn Chris Hall Helen Halliday Liz Halliwell Stella Hammond Jacqueline Hartley Suzi Hawkins Christina Hawkins Ailsa Hav Stephen Hewitt Jenny Higgins Margaret Hobby Brian Hobby Jonathan Hodgson Michael Hogan Keira Holt Peter Holt Karl Holtby John Hooson **Charlie Horsford** Frances Horsford Duncan Hovle Paul Hudson Alison Hughes Duncan Hutt Suzanne luppa Kara Jackson **Rachel Jackson** Abigail Jackson **Elaine Jaggs Huw Jenkins Paul Jenkins** Dai Jermyn Kelly Johnson Gareth Jones **Rachel Jones Iorweth Jones** Carmen Jones Neil Jordan Sarah Jupp David Kennedy Mike Klymko Janet Langsford Eliza Lawler Molly Lear Ellen Lees Francesca Leslie Sarah Lewington Molly Lear **Ellen Lees**

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