THE RESTORATION OF ECOLOGICAL CONTINUITY CORRIDORS ON MOTORWAYS

MARCH 2023

REPORT

Feedback on experience 2: Wildlife structures and monitoring on the VINCI Autoroutes network

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NOTICE TO THE READER:
This report follows on from the first feedback on experience published in June 2016, *Restauration des continuités écologiques sur autoroutes – Retour d'expérience des aménagements et des suivis faunistiques sur le réseau VINCI Autoroutes* (Restoration of ecological continuities on motorways – Feedback on experience concerning adaptations for and monitoring of wildlife on the VINCI Autoroutes network).
It notably provides further information on the factors that condition the utilisation of wildlife crossings and attempts, by statistical analysis, to answer the much more complex question of the effectiveness of these adapted structures.
The first report nevertheless remains the document reference for any reader seeking technical information on the construction of such structures (cf. Method files).
The aim is still to hare the feedback on experience of a motorway concession company and its partners. In that respect, it is not intended to be used as a guide, as may be that of the CEREMA released in December 2021 and entitled *Les passages à faune. Préserver et restaurer les continuités écologiques avec les infrastructures linéaires de transport* (Wildlife crossings – Preserve and restore ecological continuities with linear transport).

Three specific reports are published alongside this main report (see § VIII).

This document is available in French:
"*Retour d'expérience n°2 des aménagements et des suivis faunistiques sur le réseau VINCI Autoroutes; 2023*".

*: *A glossary is available at the end of the book. Asterisks refer to the definition of the associated word in the glossary at the end of the document.*

PREFACE

Transport infrastructures such as motorways have documented effects on animal populations by acting, for example, as a barrier to the dispersion of individuals. This effect can be reduced by building wildlife crossings. These structures are integrated into infrastructure projects because their location - subject to the technical conditions allowing their construction - is defined on the basis of biological criteria, such as the presence of migration routes and/or known animal movements or the presence of environments likely to favour these movements (presence of woodland, wetlands, etc.).

Once set up, these structures must have characteristics that favour their use by fairly specific communities (e.g., mammals, reptiles, etc.). Ultimately, given the cost of transport infrastructure for animal populations and for the construction of these wildlife crossings, their effectiveness obviously needs to be assessed.

Published in 2016, the report *Feedback on fauna facilities and monitoring on the VINCI motorway network* describes the technical characteristics among the implementation of 96 facilities of different type (e.g., eco-bridges, eco-ducts, hydraulic structures, bridges for chiropterans, mixed crossings, fishways, including guidance systems such as fences and palisades). And the methods for monitoring their use by fauna (mammals, including chiropterans, birds, reptiles, amphibians) using a range of methodologies (photographic recorders, traps, surveys, etc.) and the analysis of data collected over five consecutive years.

This report is a reference document on road ecology providing a great quantity of diverse new information on the monitoring of fauna using wildlife crossings. The frequentation data are mainly the number of crossings detected and the number of refusals (thanks to the positioning of the detectors). The diversity of the situations and methods used limits the possibility of making more general interpretations, such as the influence of structural parameters (structure types and dimensions) and ecological parameters (landscape environments of the structures).

The positive correlation observed between the number of crossings detected and the number of days of monitoring carried out clearly shows the need to strengthen this monitoring. An important contribution of this report is the feedback on the fauna monitoring methods used, as well as the claim of needs for new methodological developments via the description of specific monitoring (footprint traps, detection of amphibians and chiropterans, monitoring of small mammals, fishways).

The difficulty to assess the effectiveness of wildlife crossings stems from several concomitant factors. The first is the biology of the species likely to use these structures. The presence of transport infrastructure in the animals' home range is likely to impact their dispersal (i.e., the movement of individuals from their birth area to other areas for their first mating or between two consecutive mating grounds), or other movements, for example seasonal ones, with return trips between feeding and wintering grounds.

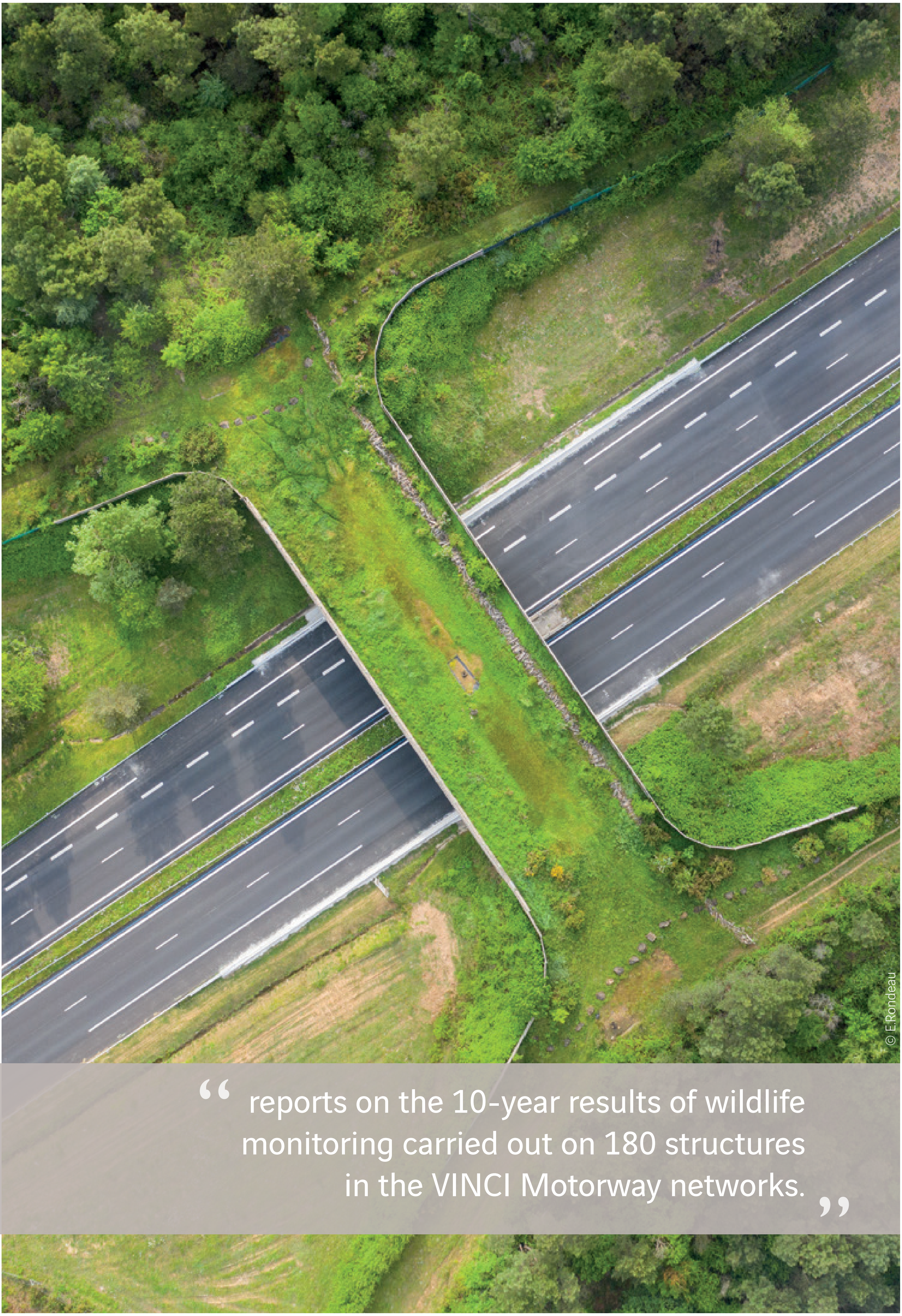
These movements may vary during animals' life cycles (e.g., differences between juveniles and adults or between sexes). It is therefore understandable that the use of a wildlife crossing by a species at a given stage and time period is the result of complex biological process that cannot be transposed from one species to another, or even from one context of structure implementation to another. The cognitive capacities of animals are also notably expressed by their ability to evaluate their environment, and the presence of a wildlife crossing is an element of the landscape that they consider during their movement choices within their home ranges.

The second difficulty relates to the scientific method. Testing the effectiveness of different wildlife crossing characteristics (e.g., length, width, substrates, etc.) should be based on experiments comparing structures with different characteristics in similar contexts. It is easy to see that this is not possible due to the design of these structures. The only experiments that have been carried out concern "small" tunnels where factors such as light, humidity or the type of substrate have been tested. When experimentation is not possible, the method consists in collecting enough data to study the relationships between the characteristics (e.g., of the structures) with the parameter of interest (e.g., wildlife use) using correlative approaches. The main problem is then having enough data so that the observed correlations can be interpreted as causal relationships.

This second report, *Feedback on fauna facilities and monitoring on the VINCI motorway network 2023*, was indeed complied using this approach. It reports on the 10-year results of wildlife monitoring carried out between 2011 and 2021 on 180 structures over 21 motorways in the VINCI Motorway network and its three concessionary companies: ASF (139 structures), Cofiroute (35 structures) and Escota (6 structures), as well as the mobilisation of 42 technical and scientific partners, which led to the collection of over 125,000 data about wildlife observation on and near these structures.

This report completes the practical recommendations from the 2016 feedback

Figure 1: Red Deer crossing the eco-bridge in La Lande Forest (A10) ©E. RONDEAU



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“ reports on the 10-year results of wildlife monitoring carried out on 180 structures in the VINCI Motorway networks. ”

report (e.g. amply illustrating the choice of structure location and their technical characteristics), puts these structures in their landscape context according to a standardised description, and presents the monitoring of structure use by clearly identifying the objectives, protocols, analyses and results.

The reader will thus find very precise protocol descriptions for the various monitoring activities carried out (sensors, traps, detectors, etc.) as a result of an outstanding field effort. 34 mammal taxa (mainly determined at species level and excluding domestic species) were detected, firstly species considered as common, such as the European Badger, Red Fox and European Roe Deer, and rarer species such as the European Otter and Stoat. This frequentation is described by the time of day and species phenology, the presence of other species (in particular humans and domestic species), and the types and characteristics of the structure.

In addition to the use of structures themselves, the sum of the accumulated data makes it possible to test hypotheses on the influence of landscape variables (density of roads, hedges, rivers, etc.) on the use of nearby structures for the species for which the most data were collected. This approach places this report in a short list of works in road ecology that focus on the analysis of explanatory variables for structure use. The limitations are also presented, because despite the quantity of devices monitored, a certain level of standardisation is observed which does not work well in testing, particularly for the use of structures in function of their dimensions.

The work carried out in this report is therefore substantial by its conception, organisation, animation, execution and valorisation. The effort made in the saving of acquired data in a database should also be highlighted, and will certainly prove useful in the future. We can only wish it the same success as the 2016 feedback report, and its appropriation by the various actors responsible for the design, implementation and evaluation of wildlife crossings.

I would like to express my personal thanks to the people behind this work - they will recognise themselves - which illustrates the value of a collaborative approach to these complex environmental assessment issues. It is to be hoped that this is a model approach that will be continued in the field of transport infrastructure, and also in other land use planning projects.

Claude Miaud
Director of Studies at
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Montpellier, le 1er décembre 2022

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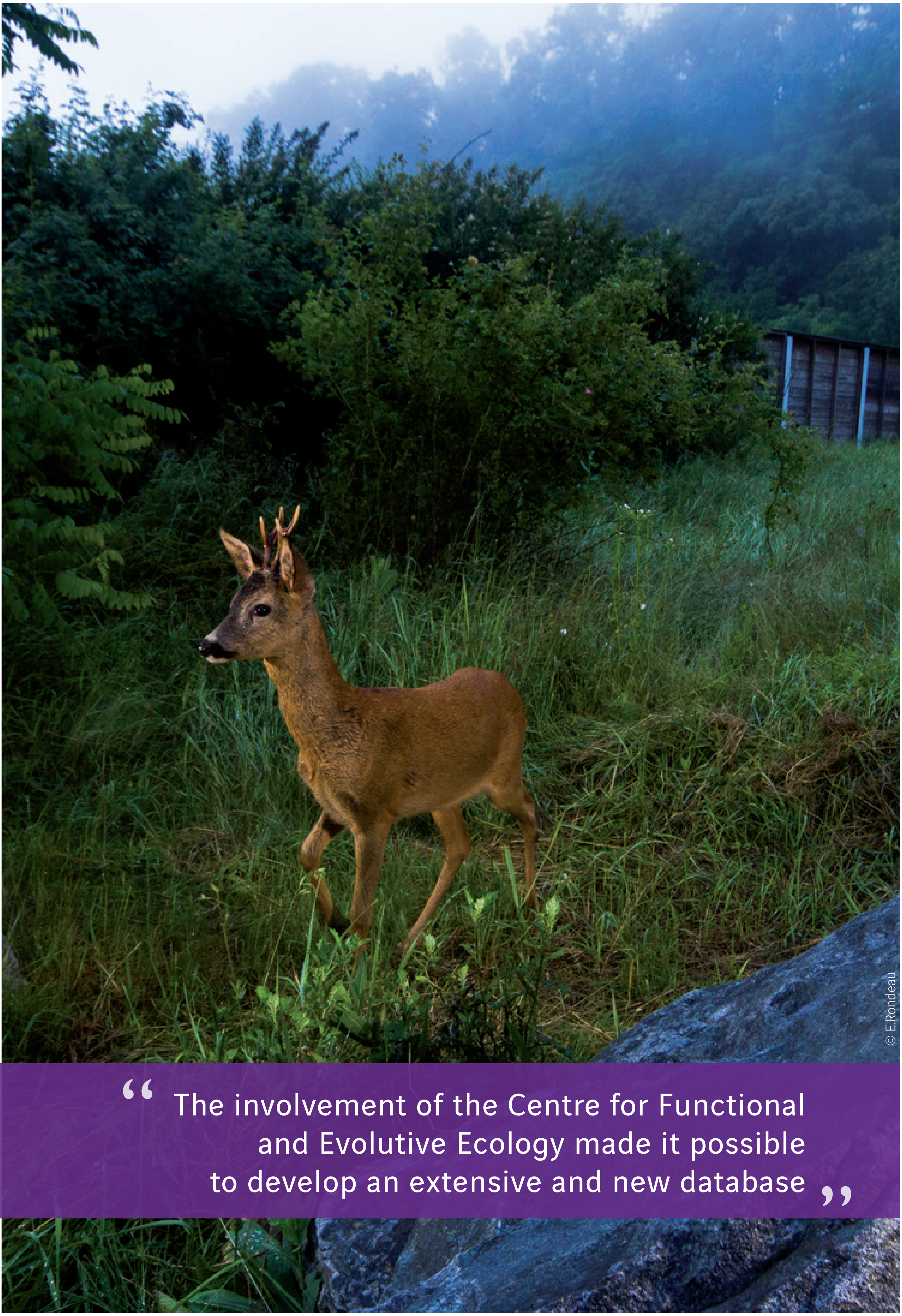


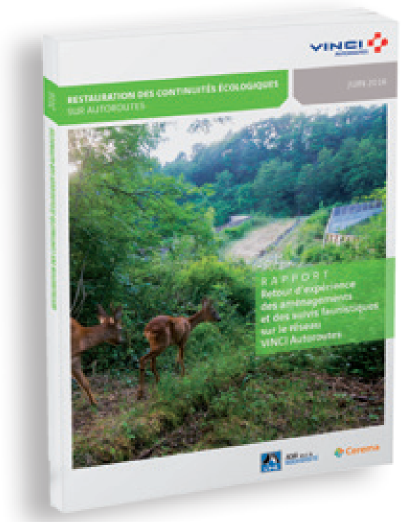
> “ The involvement of the Centre for Functional and Evolutive Ecology made it possible to develop an extensive and new database ”

1.1. FOLLOWING ON FROM THE FIRST REX 1 FEEDBACK REPORT PUBLISHED IN 2016

The first economic recovery plan in 2008 was an opportunity for VINCI Motorways to propose a programme dedicated to biodiversity conservation to the State, the Green Motorway Package (2009-2012), focusing on the restoration of ecological continuities through the construction of wildlife crossings. This experience of development and monitoring of fauna was shared in a report (referred to as *Rex 1* in this document) published in June 2016 under the title: *Restoration of ecological continuity on motorways - Feedback from the development and monitoring of fauna on the VINCI Motorway network*. This document can be downloaded from the Resource Centre website for the Implementation of the French Green and Blue Network of ecological corridor:
https://www.trameverteetbleue.fr/documentation/references-bibliographiques/retour-experience-amenagements-suivis-faunistiques-sur.

Encouraged by these initial results, new programmes followed, always within the framework of deciding opportunities jointly with the State, namely the Planning Contract (2012-2016), the Motorway Recovery Plan (2016-2020), the Motorway Investment Plan (2018-2021), but also within the framework of new projects or expansion projects, such as the creation of dual carriageways and relocation of the A9 motorway next to Montpellier (2017) or the western bypass of Strasbourg via the A355 (2021).

In total, over one hundred specialised wildlife structures have been added to those of the first programme, the Green Motorway Package (in French; PVA for *Paquet Vert Autoroutier*).



These structures have been monitored by 38 local technical and scientific partners and have resulted in the collection of over 125,000 data sets.

Specific monitoring was also carried out on specialised structures for fish fauna, semi-aquatic micromammals and chiropterans.

This document (named *Rex 2*) presents the results of the fauna monitoring carried out on all the structures built since 2009, including those of the PVA, i.e. over 180 structures. The involvement of a new scientific partner, the Centre of Functional and Evolutionary Ecology UMR 5175 (CEFE) in Montpellier, has made it possible via statistical analysis to develop an extensive and new database. Certain questions, such as the influence of structural parameters and their environment on their use by wildlife, were thus explored in greater depth, and answers were provided to new questions, particularly concerning the effectiveness of these structures.

Supported by 10 years of wildlife monitoring on a large number of structures, the document offers practical recommendations, advice on the use of assessment techniques and protocols, and suggests improvements.

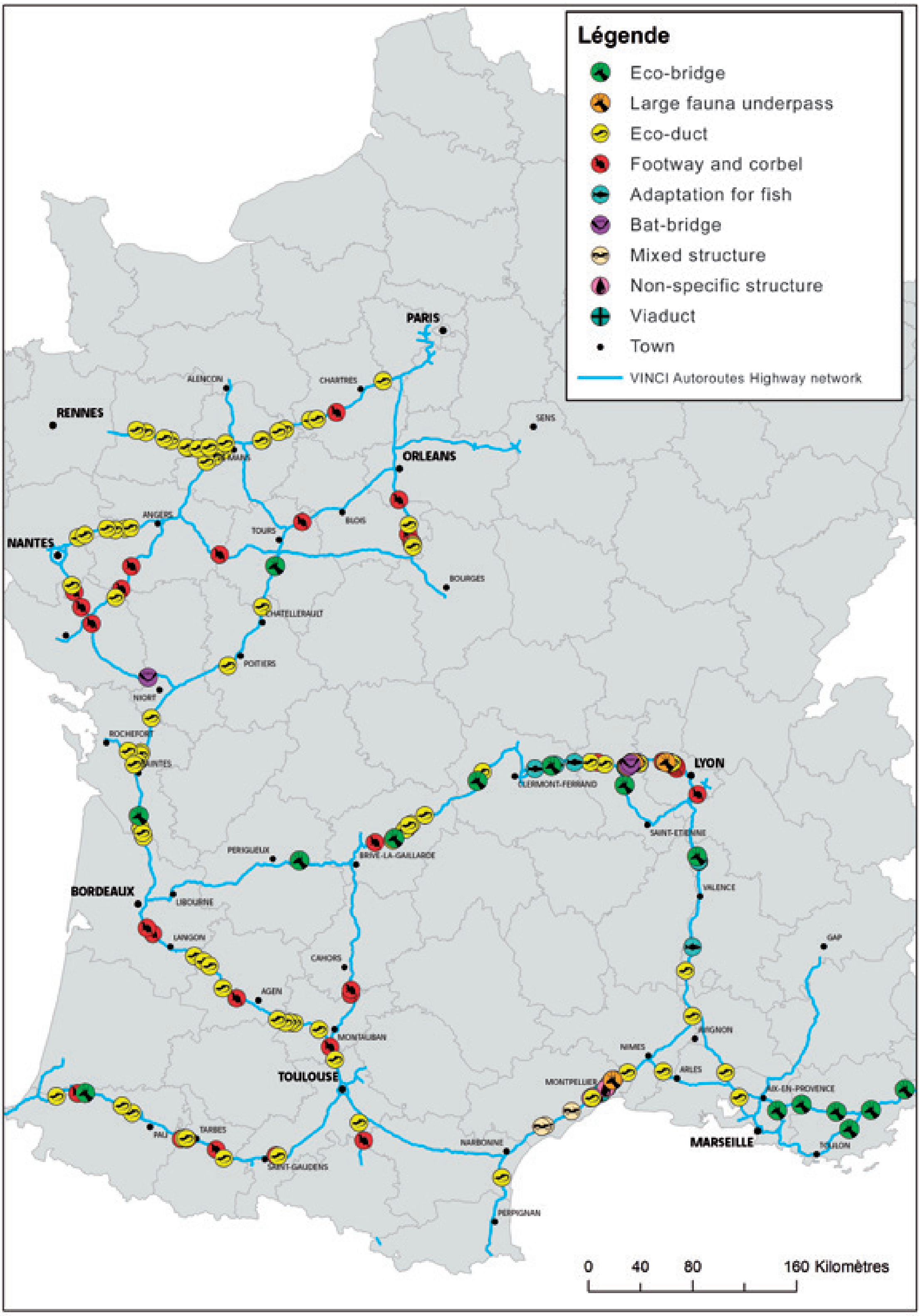


Figure 2: The VINCI Motorway network (4443 kilometres of motorways in 2022) and the location of 190 wildlife crossings: 91 eco-ducts, 39 adaptations inside hydraulic structures (footways and corbels), 20 non-specific structures, 15 eco-bridges, 7 adaptations for fish, 6 mixed structures, 5 large fauna underpasses, 4 viaducts and 3 bat-bridges constructed in the framework of requalification it programmes.

1.2. LEGISLATIVE BASIS AND PUBLIC POLICIES

1.2.1. EVOLUTION FROM SRCE TO SRADDET

In addition to Act No. 1976-629 of 10 July 1976 on the Protection of Nature, the founding laws concerning ecological continuity stem from the Grenelle Environmental Project:

1. Planning Act No. 2009-967 of 3 August 2009 for the implementation of the Grenelle Environmental Project, known as "Grenelle Act 1", which introduced the Green and Blue Network into French law and the establishment of Regional Ecological Cohesion Schemes (SRCE), which aim to preserve and restore ecological continuity by combating the fragmentation of natural areas.
2. Act no. 2010-788 of 12 July 2010 on a national commitment to the environment, known as "Grenelle Act 2", specified the applications stemming from "Grenelle Act 1": it foresees the development of national guidelines for the preservation and restoration of ecological continuity, which must be set out in the Regional Ecological Cohesion Schemes (SRCE) drawn up by the regions in partnership with the State. Thus, national level planning documents and projects (particularly major linear infrastructures owned by the State and its public establishments) must be compatible with these guidelines, while at the local level, planning documents and projects by local authorities must take into account the SRCEs and thus the green and blue corridors mapped therein (on a scale of 1:100,000).
3. Act No. 2015-991 of 7 August 2015 on the new territorial organisation of the Republic supplemented the previous regulatory framework: the regions must draw up Regional Planning Schemes, Regional Land Use and Sustainable Development Schemes (SRADDET), which are powered by the SRCEs, which are integrated de facto into the SRADDETs. Local authorities are also invited to take account of ecological continuity and therefore the ecological transparency measures implemented by transport infrastructure managers in their urban planning documents and territorial projects.

1.2.2. THE 2016 BIODIVERSITY ACT

Act 2016-1087 of 8 August 2016 for the recovery of biodiversity, nature and landscapes requires petitioners to demonstrate the effectiveness of reduction and compensation measures: "They must result in an obligation to achieve results and be effective throughout the duration of the damage".

In this respect, the present work (*Rex 2*) promotes the application of this law. It contributes to strengthening results by providing methods and specifying their application for the monitoring of wildlife crossings, which in combination with fences, constitute a measure to reduce wildlife-vehicle collisions. More rarely, it may also be a compensatory measure when the equipment and monitoring of wildlife crossings are carried out on a linear infrastructure not owned by the manager. This is the case, for example, with the compensatory measures for the Southern Europe - Atlantic High-Speed Line (LGV SEA), which includes a programme to restore ecological continuity on the departmental roads of three catchment areas linked to the LGV SEA.

1.2.3. AN APPROACH INTEGRATED INTO PUBLIC POLICIES

By restoring continuity at identified priority black spots areas in the SRADDETs and thus helping to reduce fragmentation of the territory, the creation of specialised wildlife structures is an indirect part of the national policy for the Green and Blue Corridors. It will thus enrich the Wildlife Crossing Information System (SIPAF), which is currently being developed and lists all wildlife crossings on linear transport infrastructures in metropolitan France. It also contributes to the improvement and harmonisation of wildlife crossing monitoring techniques in order to strengthen monitoring results. This contribution will be included in the future guide on wildlife monitoring of wildlife crossings that will accompany the SIPAF.

Finally, *Rex 2* also addressed objective 39a of the government's July 2018 Biodiversity Plan, which aims to address 20 of the main SRCE/SRADDET black spots.

II. CONSTRUCTION OF STRUCTURES

This chapter addresses the design of structures and adaptations for wildlife. It is intended to supplement the "construction of structures" section of Rex 1 *by presenting techniques and achievements not covered in the 2016 document.*

2.1. CHOICE OF LOCATION OF CROSSINGS FOR WILDLIFE

Supplement to Rex 1*, pages 18 to 20.*
As a reminder, the essential steps in determining the location of a wildlife crossing are as follows:

- Carry out preliminary studies (spatial analysis and ecological expertise).
- Multi-criteria analysis (issues by scale, target species, technical constraints, sustainability, etc.).
- Consultation with local stakeholders (nature protection associations, local authorities, etc.).

The main existing ecological continuities and those to be restored in a local area are now identified at different scales in the planning and development documents of the territory as well as the associated land-use planning documents. Regional Plan for Planning, Sustainable Development and Equality of Territories include in their Orientation and Objectives Documents a mapping of the green and blue networks at a supra-territorial scale. These data are in principle included in the LUP and LUPi of the municipalities and inter-municipalities. It is difficult for an infrastructure manager to guarantee the sustainability of the corridors leading to the structure. It is therefore beneficial to use these documents in order to position wildlife crossings in the continuity of the identified corridors, which should not exclude the possibility of building crossings outside of these referenced areas if expert assessments show this to be the case (limits of the SRCE/SRADDET assessments, changes in land use).

The operator's data on animal mortality is unreliable, firstly because they are incomplete due to predation and the corpses of small species that are difficult to find, and they may also be erroneous due to difficulties in identifying the species. Moreover, they are insufficient for identifying an accident site, as the animals hit may have entered the right-of-way from a point far from the collision. These data are therefore indicative[1] for the purposes of standardised diagnoses that would make it possible to objectively determine the proposed locations.
Numerous landscape ecology tools are being developed to model ecological networks and highlight potential theoretical solutions for restoring ecological continuity. These software programs use methods that are cumbersome to implement and require powerful tools for modelling at the scale of a motorway network. They provide a first level of information that requires cross-referencing with infrastructures but cannot replace field assessments.
These expert reports, based on field observations in collaboration with local naturalists (associations, hunting and fishing federations, etc.), make it possible to evaluate wildlife movements with greater precision, to refine the position of structures or to define them.
It should be noted that in order to optimise the use of the structure by wildlife, organising a consultation with the hunting community is strongly recommended and should make it possible to establish non-hunting zones, particularly around the funnels, to limit the number of hunts as well as the number of hunting stations in the immediate vicinity. This consultation should be carried out under the auspices of the State in order to involve all the partners involved in the area of the funnels, but outside the land ownership control of the manager of the linear transport infrastructure.

1. Etude comparative de deux méthodes de relevé des collisions entre la faune et le trafic (Guinard, 2019)
https://www.ittecop.fr/en/content_page/item/231-comercar

2.2. FEEDBACK ON ECO-BRIDGE CONSTRUCTION AND ECOLOGICAL ENGINEERING TECHNIQUES

Supplement to Rex 1*, pages 25 to 34.*

By acing in favour of ecological engineering, and also the environment, civil engineering enables sustainable solutions to be sought based on the choice of materials and techniques with a reduced impact on human health and our environment, notably concerning global warming. It is thus advisable to incorporate into the multi-criteria analysis, a report on CO^2 emissions related to the nature and volumes of the materials used (wood, cement, concrete, metal.). For this, we can refer to the CO^2 emission factors provided by the CEREMA.

2.2.1. TECHNIQUES FOR BUILDING ECO-BRIDGES

The choice of a type of structure follows the classic path of developing an engineered structure by seeking to meet the requirements of the target animal species. Civil engineering is thus in a way at the service of ecological engineering. The optimal solution will therefore be a response appropriate to the target animal species and to all the constraints identified during multi-criteria analysis: technical constraints (height of the embankment or excavation, width of the gap, existing networks, necessary right-of-way around the location of the eco-bridge, etc.), operating constraints (inconvenience to users, closure or reversal of traffic), financial constraints.

For example, single-span bridges are less inconvenient for operations, but require a construction area for the deck close to the final facility site from where it will then be transported, which is not always possible. Generally speaking, increasing the number of spans, which depends on the possibility of setting up intermediate supports (presence of sensitive networks to be diverted, etc.), impacts the sizing of the spans and broadens the range of solutions.

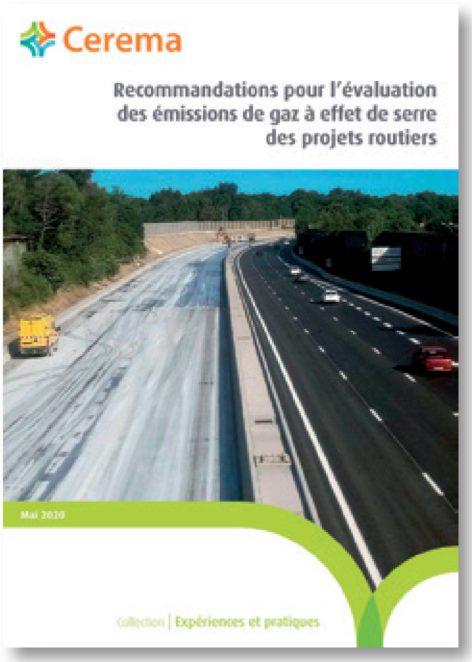


Figure 3: CEREMA Guide, Recommendations for assessing greenhouse gas emissions from road projects (https://www.cerema.fr/fr/centre-ressources/boutique/recommandations-evaluation-emissions-gaz-effet-serre-projets)

Decks and funnels

Supplement to Rex 1*, page 25.*

The deck of an eco-bridge must be watertight and resistant to the roots of plants in order to ensure the durability of the structure. An inspection should be performed to ensure good waterproofness over time. This check should be performed before the bridge is covered with materials (Figure 11).

The funnelling effect of an eco-bridge is best achieved by trapezoidal funnels and a rectangular deck rather than a curved deck (more expensive and complicated to implement under operation). The asymmetry of the funnels makes it possible to adapt to the configurations of the facility site (Figure 12).


Figure 4: Fuveau A50 Eco-bridge. © VINCI Autoroutes photo library.


Figure 5: La Pologne A89 Eco-bridge. ©KOOX.


Figure 6: Le Causse Eco-bridge - Les Grands Genévriers A89. ©KOXX.


Figure 7: Col du Grand-Bœuf A7 Eco-bridge. ©VINCI Autoroutes Photo Library.

Figure 8: Pourcieux A8 Eco-bridge. ©MARTINI.

Figure 9: Vidauban A8 Eco-bridge. © VINCI Autoroutes photo library.

Figure 10: Forêt de la Lande A10 Eco-bridge. ©MOULET.

Figure 11: Deck of an eco-bridge before being covered by material. ©M. MARTINI.

Land cover

Supplement to Rex 1 *page 25.*
A compromise is required between civil and ecological engineering to be able to implement the ecological measures in a sustainable manner. Insufficient soil thickness can increase hydric stress for plants (a problem compounded by the necessity of a bridge drainage system).
The soil is spread on site and then shaped to give natural movement to the ground, with a substrate depth of between 20 and 80 centimetres, depending on the location on the deck. It is important to work this soil in good conditions, avoiding periods that are too wet or too dry, and to compact it by rolling (spreading backward).
On flat deck structures, 80-centimetre-thick bench terraces of topsoil for planting wooded strips are created on both sides of the deck, approximately 5 metres wide (see Figure 2 of *Rex 1*, page 25). Between these bench terraces, which can be delimited and supported by low dry-stone walls (see the concept of eco-terraces), a thickness of 20 centimetres is sufficient to maintain a low herbaceous cover, without development of ligneous plants, and thus an open environment.
On vault type structures, for technical reasons, there is less filling at the top of the vault than on the sides, which are previously filled with materials before being covered by topsoil.

Sight screens

Supplement to Rex 1 *page 25.*
Sight screens can be designed from unstained vertical 2.20 to 2.60 metre high wood slats. It is advisable to use class 4 treated wood, guaranteed 15 years against fungal attacks and xylophagous insects, or wood made hydrophobic by cross-linking. Animal outlines can then be affixed to the outside of these screens used as supports showing the function of the structure to users of the infrastructure. On the inside, they equipment can be fixed to monitor the use of the structure. Note also that they provide guidance for bats.

View breaker and fences

Supplement to Rex 1 *page 27.*
The use of heather panels as a visual barrier is not recommended in areas exposed to strong winds. If the wind is too strong, the fence may break. The fence should be reinforced with struts at regular intervals. Indeed, fencing is an essential complementary measure to ensure the effectiveness of wildlife crossings (Rytlwinski *et al.*, 2016), especially for guiding animals to the structures.
Some species of micromammals, reptiles and amphibians are able to get through (fine mesh) fences set up for microfauna. To improve the effectiveness of these fences, it is often recommended to install cage-type lower liners (Figure 16). However, this type of arrangement is difficult to instal and maintain. Conan *et al.* (2022), based on tests of the effectiveness of fences with and without a liner, showed that its presence improved the effectiveness of the fence for amphibians, but not for micromammals. They recommend using opaque fencing for these species instead.
Also, in order to limit the problems of damage to the small mesh (difficulty installing, breakage of the mesh during clearing), it is recommended that the finest mesh (6.5 x 6.5 mm) be placed between the large fauna fence and the small fauna fence as reinforcement (25 x 13 mm or 25 x 25 mm) (cf. *Rex 1*, Figure 3, page 27) Experiments with other (notably opaque) arrangements are still to be conducted to find suitable solutions.

Ditch/drainage channel crossings

Supplement to Rex 1 *page 37.*
The crossing of a ditch under the fence is a real limit to the effectiveness of small fauna fences by leaving a way in especially for all semi-aquatic

Figure 12: Arial views of eco-bridges: Fuveau A50, © VINCI Autoroutes photo library; Brignoles A8, ©OLYA; Le Causse les Genévriers, ©KOOX; Forêt de la Lande A10, ©E.RONDEAU.

Figure 13: On the western bypass of Strasbourg, the panels are linked together by a metal cable and connected to the top of the metal posts. In the event of a possible breakage of one of the panels, this prevents them falling onto the roadway (4 mm diameter stainless steel cable). ©VINCI Autoroutes.

Figure 14: Case of an embankment structure (Bas-Bry - Cofiroute). The sight screens on the structure extend outside, beyond the wing walls and at the start of the access ramps for approximately 15 metres on both sides of the structure. ©VCT.

Figure 15: Broken posts of a fence fitted with heather panels after Storm Ana in 2017. ©VINCI Autoroutes.

Figure 16: When using heather for visual barriers, reinforce the fence with struts at regular intervals (every second fence post). ©VINCI Autoroutes.

Figure 17: Fine mesh liner fitted to a fence for small fauna. The most successful arrangement has a tensioning wire attached to angle irons. ©Cabinet X-AEQUO.

Figure 18: SANIEZ prototype stainless steel ditch module to secure the ditches by replacing conventional concrete sills. ©Cabinet X-AEQUO.

species (traditional grids are not suitable). Specific devices are essential to ensure maximum watertightness while limiting the filling of the outlet. The device presented (Figure 18) is based on the principle of a flap gate, but it should be improved by providing a small pit in front of it for the decantation of earthy or gravelly materials to prevent them from blocking the flap gate. Alternative solutions have yet to be found, taking into consideration the constraints of these devices once in operation.

2.2.2. ECOLOGICAL ENGINEERING TECHNIQUES ON ECO-BRIDGES

Plants

Supplement to Rex 1 *page 28.*

In general, the climatic context complicates the creation of plant cover and ensuring recovery. Plants are subjected to increasingly frequent hydric stress. It is therefore necessary to think about the selection of the species used and the difficulty of carrying out watering due to water shortages. Seeds and shrubs are also preferable to planting large stem trees because of the risk of fire amongst other things. The success of planting depends on the implementation of techniques such as working the soil, inputting organic matter and mulching to maintain humidity and limit competition from herbaceous plants.

Various tests have been implemented in the Mediterranean area:

- Adaptation of the range of plants in favour of drought-resistant plants. We can mention oleasters (wild olive trees), as an original innovation used in the framework of planting the Pignans and Brignoles eco-bridges. The issue is not to plant big, beautiful trees, but to benefit from the survival capacities of the olive trunks.
- Increasing the water retention capacity of the soil by combining massive inputs of organic matter (shreds) with incorporating cross-linked potassium acrylate-acrylamide copolymer-based water-retention agents,

Figure 19: Setup of a drip system. ©X-AEQUO.

Figure 20: Setup of a holding tank fitted with a solar panel and supplied by a borehole. ©X-AEQUO.

Figure 21: Revegetation by seeding the slopes of double-arch bridges prevents gullying. ©M. MARTINI.

installing a fixed irrigation network from the local network or from a holding tank fitted with a solar panel, and supplying water from a borehole.

To facilitate servicing the structure, it is recommended to keep a space of about 2 metres between the plants and the sight screens.

It is recommended to have a maintenance and guarantee period of at least 3 years, with two visits per year to limit the potential development of invasive species as well as to monitor the recovery of the vegetation.

Figure 22: In the dry Mediterranean context, creation of ditches to feed the ponds. ©M. MARTINI.

Feedback on the development of ponds by LPO PACA (on the SNCF network):

"The development of temporary ponds on the eco-bridges or in their immediate vicinity provides real ecological added value by forming the stopover habitats necessary for species dependent on wetlands, and by offering a point to attract wildlife (water point, hunting area). Amphibians breeding in temporary habitats particularly appreciate these adaptations. However, although temporary, it is important to avoid drying out the ponds too quickly, which is particularly the case in Mediterranean environments where rainfall is episodic between seasons. The use of a waterproofing layer is crucial to ensure that ponds are effective in this type of climate. When creating ponds, it is important to consider the durability of the waterproofing layer and to invest in a device to protect it from scouring and perforation by the hooves of ungulates. The setting up of large stone slabs (which cannot be moved by wild boar) has shown good results, but requires machinery to place them, given their weight"

Micaël GENDROT, LPO PACA.

Figure 23: Creating a pond by laying large stone slabs. ©M. Gendrot.

Ponds

Supplement to Rex 1 *page 30*

These ponds take the form of depressions with variable slopes, different depths (up to 1 metre) and a surface area of approximately 25 m^2 or 50 m^2. The setup must take into account the topography, which conditions the pond's water. The materials resulting from earthworks are reused on site.

Rockfill may be placed on all or part of the perimeter using 60- to 100- centimetre-diameter-blocks in to limit the passage of people, bicycles or motorcycles. The implementation of this rockfill needs to be carried out in accordance with the latest developments in order to ensure their durability.

If a pond is located in an area frequented by livestock, in order to avoid any damage, an agricultural-type fence (wooden stakes with 2 rows of barbed wire) is set up around the periphery.

The watertightness of the ponds is sometimes problematic, in particular due to the presence of wild boar.

Figure 24: Rockfill around the pond to protect it from wild boar. © VINCI Autoroutes photo library.

Figure 25: Testing of protection using a buried sheet of mesh (Adrets-de-l'Estérel eco-bridge). The alternation of sand, geotextiles, membranes (Bidim/ EPDM) combined with a mesh grid with anchored plating irons gives good results after one year. ©Cabinet X-AEQUO.

Figure 26: A pond trampled by wild boar. Due consideration needs to be made when creating the pond to ensure its durability and the efficacy of the seal. ©VINCI Autoroutes.

Eco-terraces

Inspired by Provençal terraces, these low walls made of prefabricated bricks, logs and dry stones compatible with the load-bearing capacity of the engineered structures, make it possible to create banks of planted soil for hedges and refuge zones for small animals and microfauna. To avoid the corridor effect, soil depth is limited to 70-80 centimetres (minimum compatible with the root development of planted trees). Figure 27 illustrates a terrace that has been deliberately collapsed to create intermittent breaks reaching level.

For further details, a data file is available on the Green and Blue Network website:

http://www.trameverteetbleue.fr/retours-experiences/amenagement-ecopont-haute-fonctionnalite-ecologique.

Windrow

Supplement to Rex 1 *page 30.*

Evolution over 18 years of windrow laid by O.G.E for VINCI Autoroutes:

As the windrow re vegetates over time, it is not necessary to replace it. Woody plants will develop on the structure and recreate a shrubby corridor.

It has also been shown to be necessary to make the windrow as long as possible in order to optimise the connection with the habitats surrounding the structure.

Figure 27: Eco-terrace deliberately collapsed to reach soil level. © VINCI Autoroutes photo library.

Figure 28: Ocellated Lizard on an eco-terrace. ©V. MARIANI

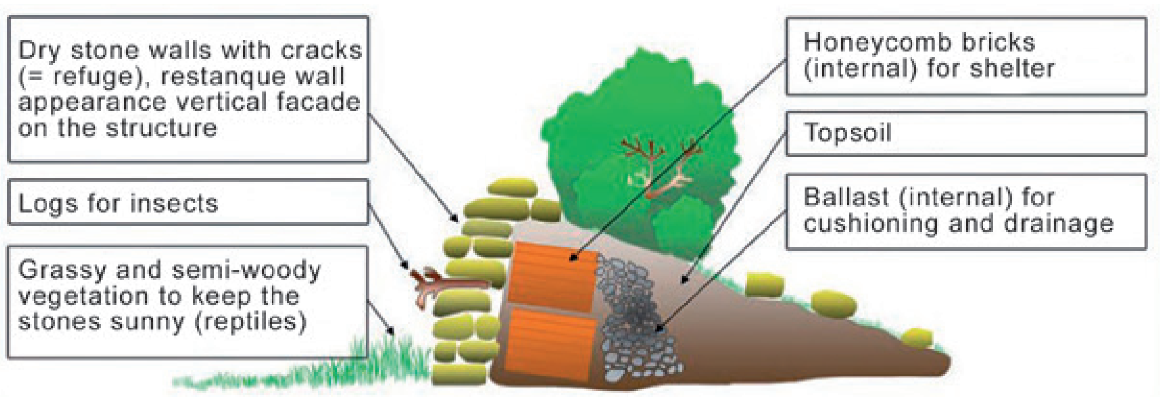


Figure 29: Eco-terrace inspired by the Provençal terrace (dry stone embankment), this provision provides a refuge structure for microfauna, while optimising the volumes of soil necessary for planting hedges. ©Cabinet X-AEQUO

Bat guidance systems on upper structures

The testing of a bat guidance system for low-flying species to compensate - at least temporarily - for the small size of the trees, did not show convincing results. (Figure 30, line of Styrofoam balls linked by a rope suspended on posts 2 metres above the ground).
Observations made at dusk on the Vidauban eco-bridge, which is equipped with this device, seem to show that the species sometimes use the fences as structures to guide their flight without particularly favouring the cable-and-ball system. Specific protocols will need to be set up in order to evaluate precisely the effectiveness of such a system and its added value with respect to fences.
These observations, linked to the difficulty of obtaining tall trees given the constraints of the soil in the Mediterranean context (shallow, dry soils), argue for reconsidering the objectives of revegetation on these structures. For guiding bats, the objective can undoubtedly be achieved by long-lasting features (fences, barriers, etc., see Chapter 8.2). Consequently, the use of small tree species could meet other ecological objectives (ecological niches, shade, etc.), without necessarily seeking to have high crowns (at least on the decks of footbridge-type structures).

Anti-intrusion systems

Supplement to Rex 1 *page 30.*
The use of anti-machine barriers (patented) has been generalised on ESCOTA structures (Figure 32). Monitoring by camera traps attests to the transparency of the system for all large and medium-sized wildlife (Red Deer, Roe Deer, Wild Boar, Wolf, etc.).
In addition, information panels on the adaptations carried out and their objectives ask people not to disturb the quietness of the site (Figure 33).

Setting up footprint traps

Some eco-bridges were equipped with 2 to 3 footprint traps per structure (1 at each end of the structure and 1 in the centre). These traps are in the form of flat strips across the whole width of the structure. These 3-metre-wide strips can complete the detection and identification of the wildlife present on the eco-bridge or help to

Figure 30: Ball-and-cable guidance system to compensate for the low development of hedges on the structure. ©VINCI Autoroutes and ©X-AEQUO.

This specific structure was the first in France to be fitted with wooden windrow and rocks (the largest trunk is indicated with yellow arrows in the photos on this page. In the photo on the right, taken from a plane on 24th June 2004, the pathway of the large fauna that uses this crossing (Wild Boar, Roe Deer, Red Deer) can be clearly seen either side of the windrow (width of the structure 12 m). The structure is 4 years old ©V. Vignon

Detailed view of the windrow five years after it was set up, 5th October 2005. A hedge is developing, in particular thanks to seeds brought by birds that perch on the structure. The heathland is recovering on the edges. The windrow is gradually "naturalising". ©V. Vignon

Detailed view of the windrow 18 years after it was set up, 19th October 2018. The sight screens have been repainted brown. The hedge, bushes and shrubby layer that developed have been managed by shredding. All that remains of the windrow is the discontinuous rocks, the wood has disappeared apart from the big pieces more than 50 cm in diameter (the large trunk indicated with yellow arrows is the only piece of wood that has hardly changed). The hedge that has replaced the wood now performs that role on the structure.

Figure 31: Analysis of the current status of wildlife crossings on the A28 motorway: assessment of management practices, management recommendations for restoring their effectiveness. Note on the management of the "Canyon" site near Alençon (72). © O.G.

Figure 32: Anti-intrusion barriers. ©VINCI Autoroutes.

Figure 33: Information panel for people using areas close to the structure asking them not to disturb the quiet nature of the site. ©VINCI Autoroutes photo library.

Figure 34: Footprint trap set up on the eco-bridge. © VINCI Autoroutes photo library.

position camera traps on identified pathways. The material used (sand/clay) must be suitable. The use of fine limestone (risk of mass capture), and clay (only marks if wet) should be limited. Sandy-loam granulometry should be tested to ensure good marking and the durability of the prints over time. It is therefore recommended that any sample be validated with the monitoring operator before implementation.

A 20-centimetre-deep "trench" is dug mechanically, and the materials spread or filled in the area. At least 270 g/m² anti-contaminant felt is placed on the bottom of the trap. The whole trap is filled using two specific substrates whose characteristics provide easy, long-lasting and recognisable markings of the prints left by the passage of wild animals. A mixture of soil and stone is placed on the first 10 centimetres at the bottom of the pit. The remaining 10 centimetres on the surface consist of a sandy substrate that meets the above-mentioned requirements. .

It is also necessary that this material can be easily raked as part of the regular monitoring of the trap.

Regulations

Supplement to Rex 1 *page 34.*

Some eco-bridge projects may be subject to exemption decrees from the prohibition on disturbing, moving or destroying protected species or habitats. Despite their positive impact on the restoration of ecological continuity and the measures implemented as part of the ERC approach, a residual impact may indeed persist in the long term. These decrees thus specify reduction and/or compensation measures, as well as accompanying and monitoring measures. Additional delays in the project planning are to be expected if necessary.

In addition, in the framework of the ministerial decisions authorising the construction of eco-bridges, the project owner must provide the conditions for the management, maintenance and monitoring of the structures to the State services (Transport Infrastructures Directorate), after validation by the DREALs concerned.

2.3. FEEDBACK ON VARIOUS PARTICULAR CASES OF ADAPTATIONS

2.3.1. USE OF SUBSTRATE IN ECO-DUCTS

On the Cofiroute network, a sandy substrate was systematically spread over the floor of eco-ducts. This type of substrate has the advantage of remaining quite smooth on the

Figure 35: Example of a clayey substrate with dry clods in a 120-centimetre-diameter eco-duct. ©Alexis Orseau/LPO France.

floor of the structure compared to a clayey substrate, which can be "ploughed up" by animals passing through it and then harden, leaving the structure with a very irregular floor. Figure 35 illustrates this phenomenon, which could potentially obstruct the passage of small wildlife (amphibians, micromammals, etc.). This observation of an excessively irregular substrate for salamanders, for example, versus a lack of substrate was also made in *Rex 1* (*Rex 1*, page 130).

2.3.2. ADAPTATIONS FOR FISH

The sizing of structural adaptations for fish are based on the SETRA's memo 96 on small hydraulic structures and ecological continuities (*Petits ouvrages hydrauliques et continuités écologiques. Cas de la faune piscicole, 2013*) and the ONEMA guide on ecological continuity (*Information sur la Continuité Écologique – ICE, 2014*). It is essential to carry out a hydraulic study for implementing fish continuity measures. The objective of the adaptations is to ensure the ecological continuity of the river while respecting the hydraulic constraints, both usual and specific to the structure. These aspects are defined in concertation with the State services (DDT, OFB). Sizing needs to take into consideration the target species of the rivers concerned and enable crossing at discharges between QMNA 5 (the statistical low-water monthly discharge over 5 years used in France) up to 2.5 times the modulus (mean interannual discharge).

In application of Article R. 181-46 of the French Code of the Environment, the competent authorities need to be informed. In function of the works and the clauses of the Law on Water applicable to the project, a declaration or authorisation in accordance with the Law on Water is required, accompanied by an impact assessment if it involves a Natura 2000 site.

The main passability problems encountered in structures are as follows:

- Insufficient roughness inside the structure (flow speed too high);
- Restrictive depth of water;
- Waterfall downstream of the structure;
- Artificial weir;
- Shade inside the structure.

To respond to these issues, various works have been implemented on rivers of the VINCI Autoroutes network.

These structures were subject to specific monitoring operations (cf. Chapter 8.1).

EXAMPLE OF THE DUROLLE

Objective: Re-establish passability for Brown Trout and European Bullhead through upstream, downstream and internal adjustments to the hydraulic structure consisting in reducing the speed of flow and creating numerous hiding places and rest areas for the fish. The adaptations carried out are as follows:

- Creation of a riprap strip on the riverbed over a stretch of about 20 metres.
- Protection of the banks with loose rockfill.
- Addition of staggered large rough elements in the bed of the structure, on the concrete apron.
- Removal of the 40-centimetre fall downstream of the metal culvert.
- Filling-in of the stilling basin and slight levelling of the spillway crest.

This morphological adaptation of the structure responds to the fish's need to swim upstream and to sediment continuity, taking into account the issues of stabilising the bed and banks of the river.

Figure 36: Creation of a test strip with large rough elements arranged in staggered fashion, separated by protruding blocks of rock (+/- 10 centimetres). ©VINCI Autoroutes Photo Library

Figure 37: Setting up large rough elements in the bed of the structure to reduce the flow speed and create rest areas for fish. ©VINCI Autoroutes Photo Library

Figure 38: Setting up cofferdams and diverting the river to be able to work in the dry. ©VINCI Autoroutes Photo Library

Figure 39: Before/After: The stilling basin has been filled in, the 40-centimetre fall at the exit of the culvert removed and rough elements added inside the culvert to influence the flow speed. ©VINCI Autoroutes Photo Library

EXAMPLE OF THE ALLIER

Objective: Reestablishment of passability for high-stake species, notably large migratory salmonids, with a major issue pour to reduce upstream delays. Two weirs under ASF concession hinder the circulation of fish and sediment. They enter into the field of application of the 10th July 2012 decree of classification in Lists I and II and Article L.214-17 of the Environmental Code, implying an obligation of result.
Weir A89 does not provide satisfactory flow conditions that are totally compatible with the passing capacities of all the high-stake species. It is globally impassable for small species.
The Joze Weir is considered as passable with low water levels and high flows, but with a morpho-ecological impact due to the formation of a sediment barrier.
Studies including modelling, topometric and bathymetric profiles, sounding, sampling, diachronic analyses, ICE* diagnoses and multicriteria analysis of adaptation variants s resulted in the sizing of a project for each weir under the authority of a steering committee made up of the ONEMA, DREAL, DDT and Water Agency. These projects sere also validated by the DRJSCS* and the French Canoeing and Kayaking Federation.
The adaptations carried out are as follows:

- For Weir A89
  - Levelling of the weir crest by +/- 0.80 metres over 96 metres.
  - Creation of an asymmetrical breach 5 metres wide and 25 metres long with a rough bed and a slope of 4 % (passable for fish and canoes).
  - Restoration by rockfill of the impacted side protections.
- For the Joze Weir:
  - Lowering the whole height of the weir (by about 1 metre) over a length of 84 metres.
  - Levelling of the Allier riverbed.
  - Restoration by rockfill of the impacted side protections.

Figure 40: Making a test strip for the creation of a ramp for fish and canoes to pass. ©VINCI Autoroutes Photo Library

Figure 41: The works were carried out progressively from one bank to the other, in the riverbed upstream of the weir and requiring the displacement of large blocks for the formation of the ramp. ©VINCI Autoroutes Photo Library

2.3.3. BAT BRIDGES

Structures for guiding bats are not yet common. Those constructed by VINCI Autoroutes are experimental, with two designs: the first being cradle-shaped and the second in the form of a sign gantry. The success of these structures largely depends on their location and the landscape context.

Two "cradle" structures have been constructed on the eastern section of the A89, between Lyon and Balbigny. Studies prior to the construction of the motorway identified the main flight paths of the target species, notably the Western Barbastelle. These metallic structures are relatively big (several tens of metric tons of steel) and required considerable technical feasibility studies. Certain particular constraints needed to be taken into consideration such as the height difference, seismic risks, impacts by vehicles and the accumulation of frost or snow on the structure. The length of 40 metres spans the whole width of the infrastructure. The "cradle" structure positioned more than 9 metres above the ground therefore guides the bats at a sufficient height to avoid collisions.

2.3.4. WILDLIFE OVERPASSES

To reduce as much as possible the distance between ecologically transparent structures in areas with embankments where structures are complicated to set up (vehicle overpass with a long breach) and costly, the construction of wildlife crossings, notably for hamsters, called "*bioducs*" has been implemented. These wildlife overpasses are an ecological transparency measure perpendicular to the infrastructure.

The wildlife overpasses are positioned laterally (north-side to reduce exposure to the sun and the heat inside) on the decks of vehicle overpass structures crossing the embankments. They create transversal crossings over the motorway, for hamsters and other species of small fauna.

Adding a wildlife overpass to an existing structure requires a specific study and load calculation to avoid compromising its integrity and perennity.

The useful dimensions of the wildlife overpasses are 60 centimetres high by 40 centimetres wide. Adaptations are made around the access points (gentle slope, plant cover) in order to make the structures attractive to increase their effectiveness.

The wildlife overpass is fixed to the edge of the deck, inside the corniche of the structure. A 10-centimetre layer of free-draining material (sand) is added in order to reconstitute a natural soil in the crossing. An artificial gallery (10-centimetre-diameter PVC pipe) will enable the hamsters to circulate safely. Several small-diameter openings (8 centimetres) are included in the gallery all along the vehicle overpass in order for individuals using the wildlife overpass outside this gallery to be able to flee.

The wildlife overpass is made using a U-shaped box beam whose upper part is in perforated sheet steel to provide natural ventilation.

The slope of the access ramps for wildlife overpasses, along the embankments of the vehicle overpasses, is limited to 25 % (1/4).

In order to facilitate the utilisation of these specific structures, the access ramps for wildlife overpasses are set up in such a way as to provide a favourable habitat for the movements of small fauna. The aim is not to create habitats for living but to provide favourable cover made up of grassland vegetation.

The vehicle overpasses reconnecting roads above the motorway are fitted with safety features (concrete crash barriers) which, combined with an L-shaped curb perpendicular to the entrance to the wildlife overpass act as an enclosure or obstacle to prevent animals entering or leaving the wildlife overpass from accessing the reconnected road.

The effectiveness of these innovative measures is being monitored in their operating phase by camera traps. The initial results are encouraging.

	A89 bat bridges: cradle structure	A83 bat bridge: gantry structure
	 *A89 Moulin-Paris bat bridge. ©P.Bouffard.*	 *A83 bat gantry. ©Koox.*
Location	2 experimental structures on the A89, Loire department (1 structure at PR 496,1, commune of Saint Marcel-de-Félines and 1 structure at PR 493,7, commune of Saint-Just-la-Pendue).	1 experimental structure on the A83, Deux-Sèvres department (PR 119,5, commune of Saint Pompain).
Choice of sites	Flight paths identified in the framework of studies prior to the construction of the A89 motorway, notably the impact study. a particular issue concerning the Western Barbastelle was observed, especially in the Vallon du Bernand. It should also be noted that there are several disaffected tunnels in the study area, used in winter.	The site chosen is located in a large intensive agricultural area with few linear elements (hedges) favourable to bat movements. A former railway line, abandoned and left fallow since 1971, crosses this agricultural area and meets the A83 perpendicularly, thus forming the only local ecological corridor between two valleys. The experimental gantry was set up in the continuity of this corridor, above the traffic lanes.
Techniques / Dimensions	Metallic structure consisting of a 12-millimetre-thick solid steel sheet mounted on a founded tubular structure. Project management (calculations) = SETEC Constructer = Baudin Chateauneuf Length = 40 metres Width = 4.82 metres Max. height = 9.66 metres Bat guidance height: variable, from 1.65 to 2.45 metres. Quantities pour 1 structure: ▸ 49 tons of S355K2+N, S355N, S355J2H NF construction steel. ▸ 35 m³ of C30/37 concrete. ▸ 2 tons of HA 500 ironwork. ▸ 860 m² of a complex (3 coats) of C3ANV930 light grey paint, RAL 7035.	The structure is a modified sign gantry, 29,4 metres in length, with a (clearance) height varying from 6 to 6.5 metres. The gantry is topped by a grid whose diamond-mesh is 4.13 x 1.3 centimetres. These standard dimensions were chosen with a view to facilitating reproducibility. The gantry is "NF" certified, having been calculated according to norm "XP P98550-1". Ten 5-metre-tall high-crown trees were planted either side of the structure in order to fill the area empty of woody plants and improve the guidance of the bats towards the structure in combination with the vegetation of the former railway line.
Constraints	Seismic, topographic height difference. Impacts of vehicles. Accumulation of snow, frost.	Seismic, topographic height difference. Impacts of vehicles.
Suivi	Cf. Chapter 8.2.	Cf. Chapter 8.2.

Table 1: Comparison of the characteristics of the two types of bat bridges tested on the VINCI-Autoroutes network.

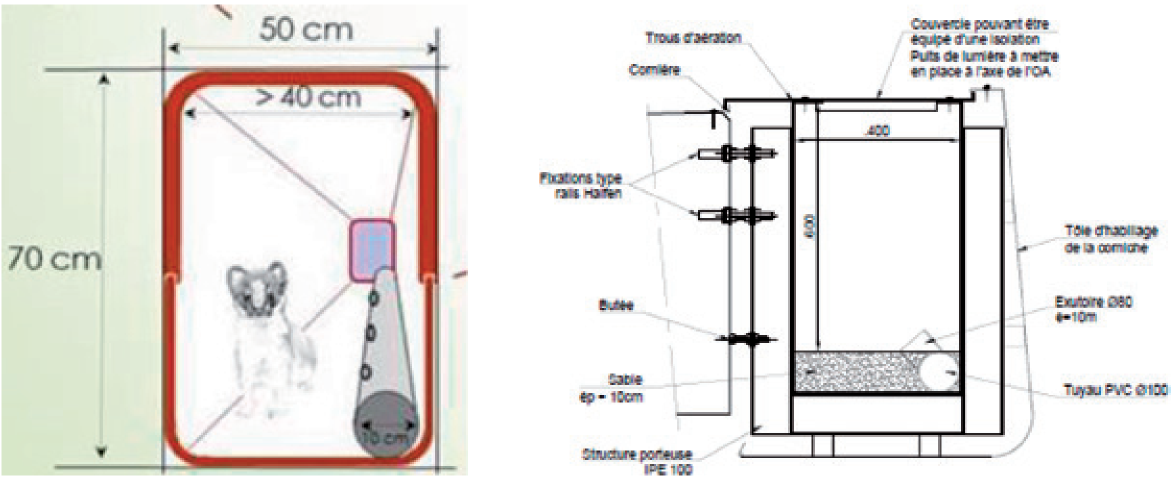


Figure 42: Block diagram of a wildlife overpass. ©SOCOS.

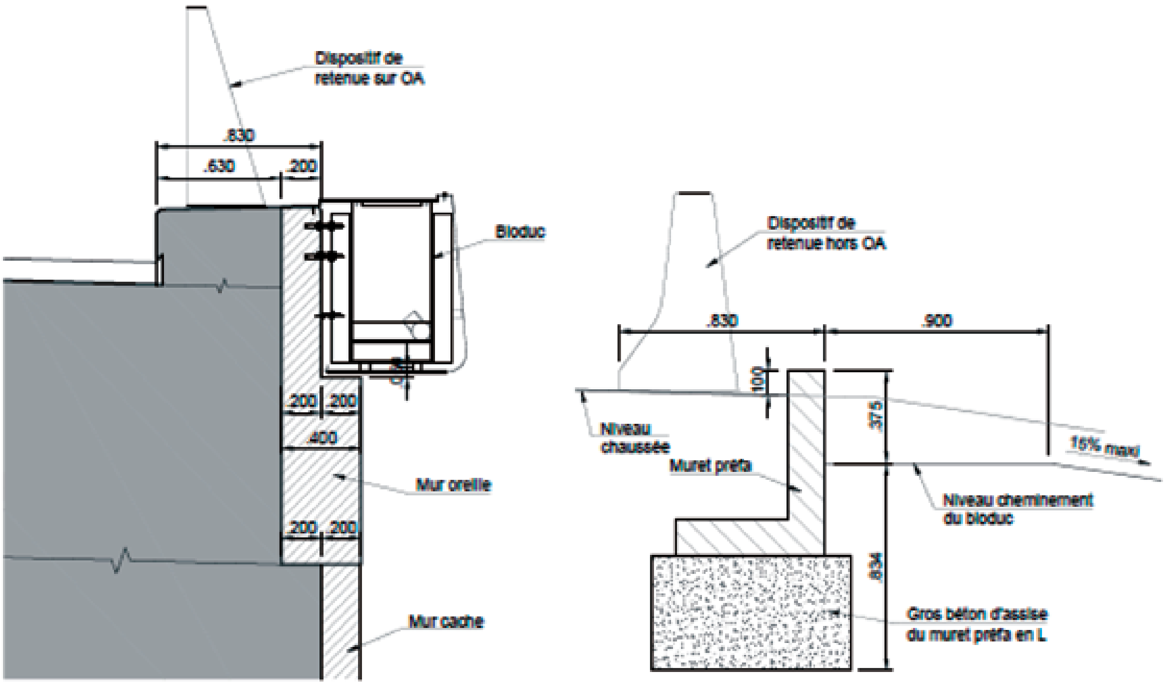


Figure 43: Principle of setting up the wildlife overpass on the deck and connection to the road ramp of the vehicle overpass. ©SOCOS.

Figure 44: © VINCI Autoroutes Photo Library

Figure 45: 70 x 40 cm gutter in 2-mm thick aluminium alloy sheeting, fixed to the support with A4 stainless steel screws. © VINCI Autoroutes Photo Library

> "Numerous species have already been observed crossing the structure, notably the European Hamster, the flagship species targeted by this adapted structure."

Badger

Weasel

Red Fox

Mole

Wildcat

European Hamster

Figure 46: Species crossing the "bioduc" (from top to bottom and left to right): Badger; Weasel; Red Fox; Woodland Mouse; Forest Cat; European Hamster. Photo Trap Captures. ©VINCI Autoroutes Photo Library.

2.3.5. FOCUS ON THE TWINNING OF TWO WILDLIFE CROSSINGS ON TWO PARALLEL INFRASTRUCTURES: THE BAS-BRY ECO-BRIDGE ABOVE THE A10 AND THE HIGH-SPEED TRAIN LINE

In the case of twinning a wildlife crossing project with another wildlife structure (on a different infrastructure, for example), it is important to ensure the passability of both structures in ecological continuity. In order to avoid creating a "tunnel" effect (long, narrow corridor) with the two structures, it is better to position them in a staggered way (slight staggering) with an undisturbed area between the two structures. Moreover, the curved lines need to give an integrated appearance to the structure. Work also needs to be done on the architectural plan of the passage between the two structures with appropriate landscaping and revegetation to maximise the chances of the two structures being used in continuity. An undisturbed area between the two structures needs to be set up to ensure a restful and safe place for the animals between two crossings.

2.4. MAINTENANCE OF STRUCTURES AND ADAPTATIONS

In order to ensure the lasting effectiveness of adaptations for wildlife, regular maintenance and checks of the amenities need to be set up.

Figure 47: Bas-Bry Eco-bridge, a crossing for wildlife above two infrastructures. ©VINCI Construction Terrassement. ©VINCI Construction Terrassement.

Figure 48: Satellite view of the site. ©Ortophotos Geoportail IGN.

Fences	Fences and small-mesh grid reinforcements guide the fauna towards the crossing structures. These fences must be maintained in satisfactory condition and replaced in the event of deterioration. Particular attention must be paid to the attachment points with structures and amenities and to the amenities themselves (gates, doors, etc.) so that the arrangement is completely impassable.
Sight screen	The aim of sight screens is to consolidate and prolong cover around the structures. Sensitive to storms and especially strong winds, they require regular checking.
Vegetation	After maintenance to ensure successful regrowth, progressive checking and removing of the plant supports, attachments and anti-rodent protection devices is required. If pruning is necessary, the clippings are left on the site, cut up and et placed at the foot of planted trees or shrubs or on the heap to act as a refuge for small fauna. Concerning spontaneous vegetation, which will develop naturally in the immediate vicinity of entrances to underpasses and on eco-bridges, occasional clearing needs to be planned… For underpasses, especially eco-ducts, footways and corbels, the maintenance objective is to prevent this vegetation from blocking or hiding the immediate access points to the structure so it can be visited. For eco-bridges, the thinking is different with the width and funnelling of the structure enabling the development of different zones. The objective is to obtain a patchwork of open and wooded habitats to conserve cleared and open areas to attract large fauna and, in parallel, covered areas for small and medium-sized fauna which will seek to move under cover. Attention also needs to be paid to pruning any high-crown trees that develop above the crossings. The frequency of the maintenance work, mainly dependent on climatic conditions, will be in the order of once every 2 or 3 years. To respect the periods favourable to wildlife, this maintenance must be carried out between September and late February. For mechanical or manual mowing/brush cutting care must be taken concerning the fencing specific to small fauna, which is more fragile, in order not to damage it.

Figure 49: Excessive vegetation development in a pond ©FNE Loire

Ecological adaptations	Specific adaptations (ponds, small fauna shelters) must be checked regularly. Ponds must be scraped out to avoid gradual silting up and/or cleared of excess vegetation, which develops naturally, in order to maintain water capacity. This operation can be carried out every 3 to 5 years in function of the evolution of the habitat and on at least 2/3 of the surface area. The pond is scraped out manually to avoid piercing any watertight geomembrane, in September or October during the dried-out period and outside the breeding season for amphibians (February/March to June). The heaps of wood, hibernacula, which are shelters for small fauna, must be checked and reloaded with branches, trunks or stumps if necessary (natural, deliberate or accidental degradation) to maintain dimensions of about 1 metre by 1 metre.
Fish ramps	Fish ramps must be visited regularly to remove any jams (waste, vegetation) that may obstruct them and prevent the circulation of fish, taking the flood pattern into consideration. Branches etc. should be reused on the bank as shelters for small fauna.
Illegal tipping	Care must be taken to ensure the hygiene and tranquillity of the sites for wildlife by preventing any degradation or storage, removing illegal tips and repairing damaged amenities.

Figure 50: Jam at the entrance to a fish ramp, making the adapted structure unusable by fish ©VINCI Autoroutes Photo Library

III. DATABASE OF MONITORED STRUCTURES

3.1. TYPOLOGY AND CHARACTERISTICS OF MONITORED STRUCTURES

3.1.1. MONITORED STRUCTURES SELECTED

The structures selected for compiling the initial database are those having been monitored at least once by camera trap whose standardised protocols since the first feedback are considered sufficiently homogeneous. Monitoring carried out between June 2011 and March 2021 concerns **180 structures** spread over 21 motorways of the VINCI Autoroutes network and its three concessionary companies: ASF (139 structures), Cofiroute (35 structures) and Escota (6 structures).

On average, the initial operating of the motorway sections where the structures were built and monitored dates from 1990 (±ET 15; minimum: 1961; maximum: 2017). These structures are therefore located on fairly old motorway sections, with the exception of the structures monitored on the last section of the A89 freeway operating in January 2013 between Balbigny (Loire) and La Tour de Salvagny (Rhône), and the doubled and relocated A9 motorway near Montpellier (Hérault) completed in 2017. The structures monitored went into operation between February 2011 and February 2019 as part of the Green Motorway Package (2009-2012), a contract plan (2012-2016), the Motorway Recovery Plan (2016-2020), or as part of new motorway construction projects such as the A89 or A9 motorways.

3.1.2. TYPOLOGY OF STRUCTURES

The classification of structures is based on the VINCI Autoroutes reference system adopted at the end of Rex 1, which differentiates the structures monitored into **11 types of structures (Table 2)**. While most structures are dedicated to wildlife crossing (150 structures), some mixed structures (6) and non-dedicated structures (24) were also monitored:

- Mixed crossing: crossing partially adapted for fauna (for example: road with vegetated sidepath).
- Non-dedicated: structure not dedicated to fauna (viaduct, overpass/underpass for road reconnection, hydraulic structure) that may enable the crossing of wildlife.

Table 10 in Appendix 1 indicates by type of structure the number of structures monitored (by camera trap) included in the database.

3.1.3. PARAMETERS SELECTED TO DESCRIBE THE STRUCTURES

Certain adaptations and design features may influence the use of structures by wildlife. A set of 10 variables describing the structures and their adaptations were tested to explain the weekly occurrence of species in the structures:

- Type of structure
- Operational width (metres)
- Operational height (metres)
- In-ground large fauna fence
- Small fauna fence
- Foldable small fauna fence
- Bottom panel of small fauna fence
- Acoustic protection
- Type of soil substrate
- Light shafts

The structures monitored are relatively homogeneous in length. As there were not enough structures of different lengths, this variable related to the size of the structures was not included in the analysis.

Table 10 in Appendix 1 specifies, by type of structure, the average dimensions of the structures monitored by camera trap in the database.

Types of structures monitored	Description	Ilustration
DEDICATED ADAPTATIONS		
Éco-duct	Circular, dry culvert (without hydraulic purpose). Small to medium sized underpass (up to about 2 metres wide) enabling the passage of small and medium-sized fauna under the transport infrastructure (with or without earthen substrate added to the ground)	
Footway	Single or multiple steps, set up on the apron, laterally within a hydraulic structure enabling the passage of fauna without getting wet (1 or both banks can be adapted).	
Corbelled arch	Structure fixed to the walls of a generally hydraulic structure, connected to the bank, enabling the passage without getting wet (1 or both banks can be adapted).	
Rockfill/natural embankment	Reconstructed bank or rockfill enabling the passage of wildlife without getting wet in a hydraulic structure (1 or both banks can be adapted).	
All-fauna underpass	An underpass of sufficient width designed for all wildlife (small, medium and large fauna). A variety of attractive habitats can be created around the structure (seedlings, plants, ponds, etc.) or inside (windrow, shelters, etc.) to enable the passage of as many animal species as possible under the transportation infrastructure.	
Eco-bridge (or overpass for all fauna)	A vegetated overpass at least 10 metres wide, designed for all wildlife (small, medium and large), creating a diversity of habitats (seedlings, plants, ponds, windrow, etc.) to enable the passage of as many animal species as possible over the transportation infrastructure.	

Table 2: Classification of the monitored structures (next page also)

Types of structures monitored	Description	Ilustration
MIXED STRUCTURES		
Mixed underpass	An engineered structure that was originally designed exclusively for road traffic and was later modified (usually with a natural pathway) to facilitate wildlife movement through the structure while maintaining road traffic.	
Mixed overpass	An engineered structure initially designed exclusively for road traffic and whose upper part, at a later stage, was modified to facilitate the movement of wildlife, while maintaining road traffic.	
NON-DEDICATED STRUCTURES		
Non-dedicated underpass	Underpass (road, agricultural, forestry, etc.) not adapted for wildlife crossing, but whose configuration seems suitable for the passage of certain species.	
Non-dedicated hydraulic structure	Hydraulic structure not adapted for the passage of fauna, but whose configuration seems suitable to the passage of certain species, particularly in dry periods.	
Viaduct	Overpass spanning a talweg (valley, river) and the associated transport axes.	

All combinations of these variables were also tested to explain the occurrence of species by incorporating structure coordinates into the two axes of mixed data factor analysis (AFDM, Lê *et al.*, 2008) as an explanatory variable for the models (c.f. §4.4. modelling).

3.2. ENVIRONMENT OF MONITORED STRUCTURES

Ten variables describing the environment around the structures were selected (the source layers are specified in brackets):

- the percentages of land use: (1) wooded areas (database Forêt® 2014), (2) built-up areas (database TOPO®, v.2020), (3) pasture and (4) fodder (Graphic parcel register, RPG v.2020);
- the densities of (5) roads (Bd ROUTE 500®, v.2020), (6) hedges (database TOPO®, v.2020), (7) rivers (database Topage®, v.2019);
- distances to the nearest (8) rivers (database Topage®, v.2019), (9) woodland edges (database Forêt®, 2014) and (10) protected areas (SCIs, SPAs, NNRs, RNRs, RNCFS, ZNIEFF1, ZNIEFF2 and APPB sites; INPN 2021).

These variables were extracted from the 500-metre, 1-kilometre, 5-kilometre, and 10-kilometre buffer zones, respectively, around the wildlife crossings (QGISis.org, 2021; R Core Team, 2021; PEBESMA, 2018; Bivand *et al.*, 2021), representing four scales of study.

The analysis of these variables shows that the monitored structures are distributed along two habitat gradients, for the four scales studied:

- a gradient from forested habitats to open landscapes (grasslands, crops);
- a gradient from habitats with natural, agricultural or forest soils toward habitats with artificial soils (buildings/roads).

No single group of structures stands out, i.e., the structures are distributed along these two axes. A few structures are found in purely forested or open landscapes, but most are located in landscapes with a variety of land uses. This pattern is observed at all four scales studied.

To select the most relevant scale for each of the landscape variables, the four scales of variables were selected one by one by model comparison, selecting the models that best explained the weekly occurrence of the species (c.f. §4.4. modelling).

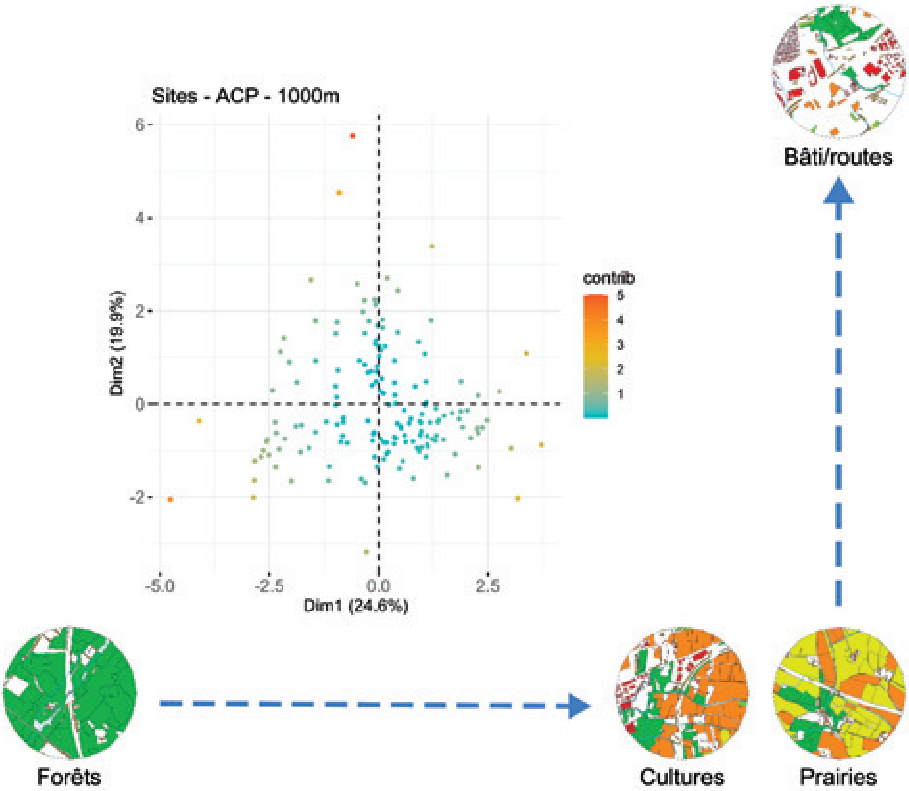


Figure 51: Distribution of structures (dots) in the first two axes of the ACP factor space (Principal Component Analysis; Bates et coll., 2015) focused on the environment surrounding the structures (a one km buffer zone here). Axis 1 (horizontal) represents a forest habitat gradient toward open habitats, and axis 2 (vertical) represents a habitat gradient with natural, agricultural or forest soils toward habitats with artificial soils.

IV. MONITORING THE FREQUENTATION OF STRUCTURES: OBJECTIVES, PROTOCOLS AND ANALYSES

Figure 52: Group of European Badgers crossing a 120 cm culvert. ©ASF/LPO Sarthe.

4.1. OBJECTIVES

The primary objective of the structures studied here is to completely (restoration) or partially (reduction) re-establish the movements of wildlife on both sides of motorways which, in France, are fenced off from large fauna to limit the risks of collisions between vehicles and wildlife.

The objective of wildlife monitoring conducted since 2011 on the VINCI Autoroutes network has been firstly to answer the question of use: **does the adaptation implemented allow wildlife to cross the motorway?** The objective of the proposed monitoring is to estimate **the species that use the structures and their frequency**. The data collected for nearly 10 years by standardised protocols also offer the opportunity today to compare the results of monitoring obtained on nearly 180 structures to study **the factors specific to the environment and the structures that influence the frequency of passages of species.**

The question of the effectiveness of the structures and the means necessary to be set up to answer this issue is addressed in Chapter 8. This question, which focuses not on the use of the structure alone, but on its usefulness in maintaining a population of a given animal species in good conservation status, is in fact much more complex and difficult to address. For example, a single crossing by a Red Deer over an entire season may ultimately make a structure more efficient and useful than one that is heavily used by an abundant species on both sides of the highway.

Movements and dispersal

Animals can make many movements during their life cycle. There are many classifications of these movements, but a scientific consensus is emerging through a unified definition of dispersal. Dispersal is defined as the movement of an individual from its birth area to another area to breed for the first time (natal dispersal), or the movement between two successive breeding areas (reproductive dispersal). This definition is therefore very closely linked to the phenomenon of reproduction, as the movement of individuals may be accompanied by gene flow if reproduction is successful. Dispersal thus differs from other movements that animals make for their basic needs such as the search for food or shelter, for example, or for wintering. These migratory movements are often seasonal, moving back and forth between feeding, summering and wintering grounds. Dispersal and migratory movements are different processes. Dispersal is described as a three-step process: emigration (the movement away from the birth or breeding area), transit movement through the landscape matrix, and migration (movement to arrive at the breeding ground).

Dispersal is a core process for ecology and evolution, as it greatly influences the dynamics of populations (which are distributed in favourable, but discontinuous spaces separated by an unfavourable landscape matrix), gene flow and consequently evolutionary phenomena. Individuals may be faced with a choice (to move or not) at different stages of dispersal. The determinism of dispersal may depend on individual factors (dispersal may be dependent on the condition of individuals such as their stage - juveniles versus adults - or their body condition) and environmental factors (dispersal is context dependent, such as resources or density).

In the context of man-made landscapes, animal populations are often confined to favourable spaces and surrounded by an unfavourable landscape matrix and/or infrastructure serving as barriers. The various movements of (terrestrial) animals and dispersal can therefore be affected. Wildlife crossings can thus allow for daily movements (e.g., for an otter to feed in its territory) and seasonal movements (e.g., for the feeding grounds of different Roe Deer between winter and spring). They contribute to successful dispersal by allowing individuals to breed in areas distributed across the transportation infrastructure.

Figure 53: Camera trap set up in a 120 cm eco-duct. ©Alexis Orseau/LPO France.

4.2. PROTOCOLS SET UP FOR VINCI AUTOROUTES MONITORING

The monitoring concerned by this study includes all VINCI Autoroutes (ASF, COFIROUTE and ESCOTA) wildlife crossing monitoring carried out from 2011 until the beginning of 2021.
In the meantime, *Rex 1*, published in 2016, was an opportunity to partially standardise data collection and entry, for example by giving instructions on the installation of camera traps in structures (Fagart *et al.*, 2016), on protocols for monitoring small fauna on eco-bridges, and on protocols for entering collected data according to standardised formats.
However, despite efforts towards standardising protocols, some heterogeneity still remains that is notably related to the diversity of stakeholders involved in monitoring (e.g., different motorway operating companies) and to constraints at some sites (risk of equipment theft, accessibility, etc.).

4.2.1. PROTOCOLS SET UP FOR STRUCTURES MONITORED BY CAMERA TRAPS (EXCEPT ECO-BRIDGES)

To answer the question of the use by large and medium-sized fauna of variously dimensioned underpasses (from eco-ducts less than 1-metre wide to 3-4 metre-wide underpasses), VINCI Autoroutes' monitoring is based mainly on multispecies monitoring carried out using camera traps (Table 3) positioned within the structures (Figure 53). The performance of these devices makes it possible to detect in a relatively homogeneous way the use of underpasses by large and medium-sized fauna (ungulates, lagomorphs, foxes, mustelids, etc.).
The limit of the devices used is the detection distance/animal size ratio, so the position of the equipment must be adapted according to the size of the structure monitored and the target species. Various triggering systems (infrared, vibration, Time-Lapse*) can also be used (see §4.2.). In all cases, a minimum number of recommendations is necessary in order to optimise the camera traps detectability and

	ASF	COFIROUTE
Equipment	Infrared camera trap (Reconyx HC600)	Infrared camera trap (Reconyx HC600) or vibration camera trap (Cuddeback E3)
Position/ orientation of equipment	► positioned inside the structure, ► directed toward inside (longest side), ► height of attachment between 30 cm and 130 cm depending on targeted species, with adapted inclination (see recommendations Rex 1, p. 118-119) ► placing an item (large rock) to allow the fauna to mark their territory in front of the camera trap	
Camera trap adjustments	Photo mode / maximum sensitivity / triggering 24 hours a day / no time lapse	
Duration of monitoring	3 years	3/6/12 months
Entry recommendations	1 line entered for every passage detected including at least: date / hour / species / number of individuals / direction of movement / refusal / comment	
	Enter passages of humans and domestic animals	
	Standardised Excel template file	Excel template file

Table 3: Principal recommendations requested by motorway operating companies for monitoring structures except eco-bridges by camera traps.

to collect homogeneous data throughout the monitoring.
The following table summarises the main recommendations transmitted to monitoring operators since 2016 on the collection and entry of multispecies monitoring data with the objective of evaluating the use of structures by wildlife.

4.2.2. PROTOCOL SET UP FOR MONITORING ECO-BRIDGES

Due to their large size and specific adaptations (vegetation, windrow, ponds, etc.), eco-bridges target a multitude of species. In order to answer the question of the species that use the eco-bridges (in terms of crossing or living habitat), various monitoring protocols are set up.
The installation of several camera traps is recommended to assess the use of the structure by large and medium-sized fauna. The layout of the camara traps must be adapted to the configuration of the eco-bridges to maximise the detection of passages.
For small fauna, i.e., micromammals, bats, reptiles and amphibians, specific protocols have been adapted to each group and to the assessment objectives. This monitoring can be used to determine whether the eco-bridge is used as a corridor (movement across the highway), and also to determine whether it is used as a habitat for settling by certain species with small home ranges (micro-habitats created by pools, windrows, etc.). Other monitoring for avifauna or flying entomofauna (*Rhopalocera*, etc.) has been carried out, but without a protocol adapted to the problem of crossing a motorway. For these species, it is a separate issue (Jones and Pickvance, 2013) which is not addressed here.
Table 4 lists the types of monitoring protocols used by VINCI Autoroutes for each group studied.

4.3. DESCRIPTION OF MONITORING OPERATIONS

4.3.1. ORGANISATIONS IN CHARGE OF MONITORING

42 organisations (associations for the protection of nature, departmental hunting federations, environmental consultancies) were responsible for organising the monitoring of fauna by camera traps of all the structures according to protocols and specifications defined by each concession holder (see Table 3). The organisations in charge of the monitoring and the sampling carried out are listed in **Appendix 2**. Each monitoring operation is accompanied by progress reports and a final report.

	ASF	ESCOTA	COFIROUTE
Mammals (large and medium-sized fauna)	4 infrared camera traps Reconyx HC600 positioned in the middle of the apron, directed toward the centre of the structure, and set 100-130 cm off the ground.	Infrared camera traps and footprint traps	2 camera traps and 3 footprint trackers per eco-bridge
Small mammals	CMR* (INRA traps)	Ink traps, INRA traps	-
Reptiles	Detection techniques described in POPReptile (SHF)	Targeted surveys	-
Amphibians	Detection techniques described in POPAmphibien (SHF)	Targeted surveys	-
Bats	Specific studies (cf. Naturalia Environnement study)	Active listening, passive recording	-
Duration of monitoring	3 years	5 years	-

Table 4: Principal protocols used by motorway operating companies for monitoring eco-bridges.

Figure 54: Various monitoring devices along a windrow: reptile trace tracker and INRA traps for small mammals. ©ASF/LPO Auvergne.

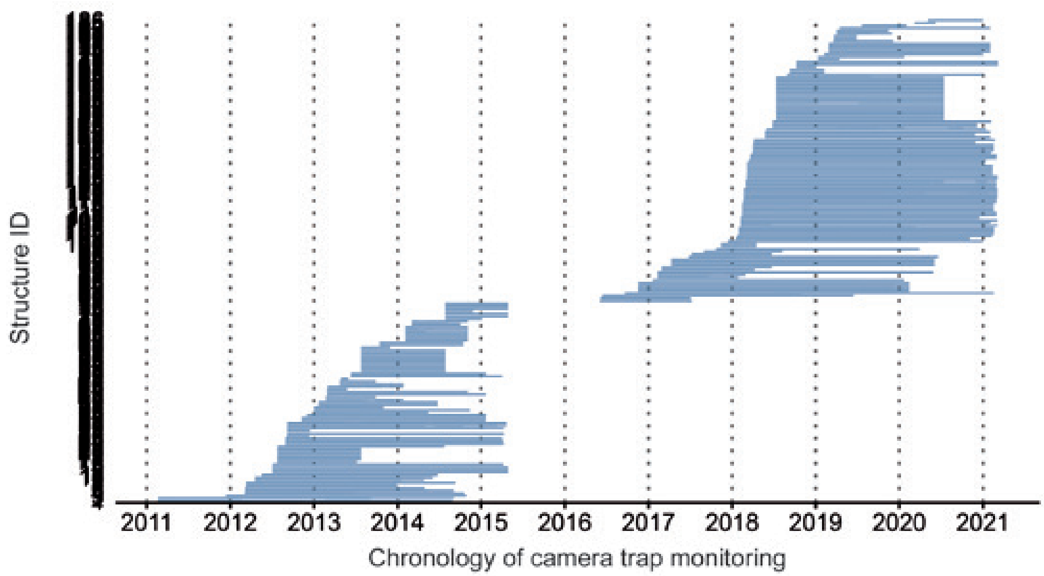


Figure 55: Observation pressure (period of operation) of the camera traps on the 178 monitored structures retained for the analysis (178 structures retained out of 180 monitored).

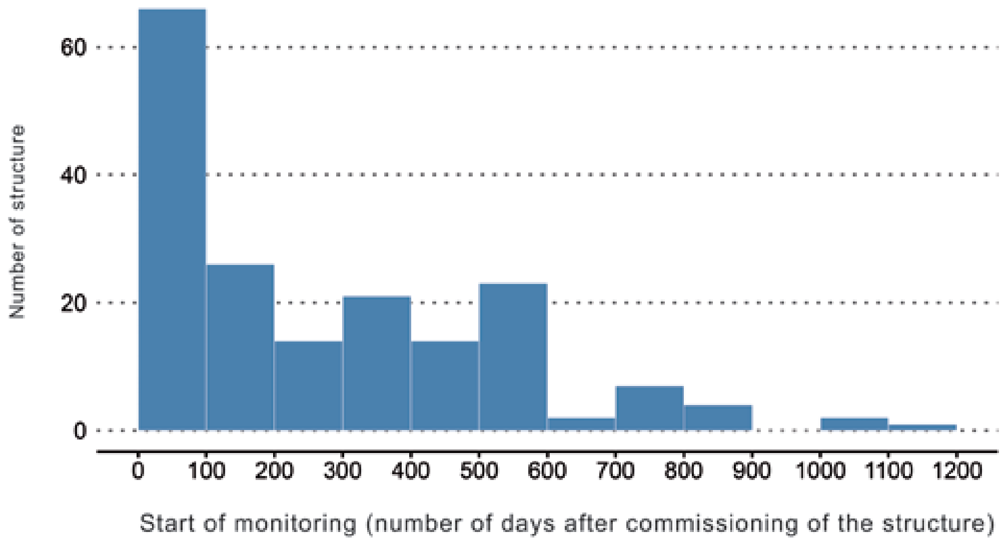


Figure 56 : Start day(s) of beginning of monitoring with regard to initial operation of structures (180 structures).

4.3.2. DESCRIPTION OF PERIODS AND DURATION OF MONITORING

The duration of the monitoring, as well as the period of deployment in relation to the installation of the structures, are also factors known to influence the frequencies of detected passages.

In the case of the present feedback, the average duration of monitoring is **572 days, i.e., 1 year and 7 months** (SD = 362 days; min = 32, max = 1,204 days, i.e., 3 years and 4 months; Q 25% = 242, Q 75% = 970). The majority of the monitoring operations are thus of rather short duration (67% < 2 years and 33% < 1 year) and only 17 structures reach 3 years of monitoring. Figure 32 shows the distribution and duration of monitoring between 2011 and 2021. Of the 180 structures in the database, 178 structures were selected for analysis.

With a median start date of 194 days (6.4 months) after the initial operation of the structures, the majority of the monitoring operations are done very quickly after the setting up of the structures (Figure 55).

4.3.3. DESCRIPTION OF DEVICES: CAMERA TRAPS USED

While the first feedback proposed elements towards a harmonising the positioning and camera trap devices, in practice, the monitoring differs depending on the models of camera traps used, the triggering system and their positioning. These factors are known to influence the detection rates of the different target species.

Camera trap model:

Nearly **15 camera trap models**, across the range of the most recent models developed by different distributors, were used to monitor the structures studied. The performance of camera traps is known to vary between brands and different generations of models (SWAN *et al.* 2014; DRIESSEN *et al.* 2017; APPS & MCNUTT, 2018). For monitoring focused on multisite comparisons and/or time series, it is then advisable to favour the use of a single camera trap model and similar positionings. Monitoring spans over more than 10 years, the models used between 2011 and 2015 (*Rex 1*) are already outdated compared to the latest generations of equipment chosen for the most recent monitoring.

Detection system and camera trap adjustments:

Currently, the majority of camera trap models are equipped with passive infrared (PIR) sensors that are triggered by moving objects with a different surface temperature than the background environment (Welbourne *et al.*, 2016; Wearn & Glover-Kapfer, 2017). This detection method is specific in that it will miss part of the endothermic species that produce too little heat (micromammals or small vertebrates) and most of the ectothermic species that do not produce heat (reptiles, amphibians, and invertebrates).

All camera traps are configured with at least one **infrared trigger**. These devices can be coupled with other external active detection systems. Thus, for about 10% of the monitoring operations (23 structures), the camera traps are coupled with vibration detection systems (cloth placed on the ground to capture the vibrations, *Rex 1*, p. 122) enabling increased detection of small fauna. It should be noted that the adjustments and positioning of the vibrating cloth are intricate, and it may not be very effective if it is badly positioned. It can also physically disturb the passage of fauna (an accustomization period is sometimes necessary).

For a particular monitoring operation aiming at detecting the movements of amphibians, the camera trap was configured in "Time-Lapse" mode, i.e., taking a photo at a fixed and regular time interval. This technique requires tests in order to define the ideal time step that will avoid missing any amphibian passages between two photographs (this depends on the camera's field of view). This frequency of photographs makes it possible to calculate the autonomy of the memory card of the camera and imposes a more regular time step for the surveys.

Generally speaking, the photo mode is preferred to the video mode, which is more cumbersome to process/store and slows down the speed of the cameras. However, this mode can be used for other monitoring purposes (wildlife behaviour in relation to adjustments, etc.).

The setting of the detection sensitivity is normally always set to the maximum sensitivity of the devices. However, this sensitivity can vary between different models without knowing precisely to what extent.

Positionnement et orientation du piège photographique:

In order to observe animals crossing the structures, the camera traps are positioned **inside the structures**. A few exceptions concern the eco-bridges where camera traps can be placed in order to also observe the fauna around the structures (see §8.1.).

The height and direction of the camera traps are two essential parameters that can strongly impact the number of detections obtained. Although instructions were given for optimal positioning of the camera traps (Table 3), the constraints inherent to certain sites and structures led operators to set the cameras at a higher height than recommended, with a corresponding degradation in the detection of passages compromising the quality of monitoring (Meek *et al.*, 2016). The risk of theft of camera traps is thus the primary constraint to their optimal positioning. The concern of not obstructing the passage of fauna, or the avoidance of areas likely to trigger the cameras in an untimely manner (water, vegetation, sun reflections, etc.), or the accessibility of the structure being monitored are also taken into account.

In the case of small structures a metre wide or less (footway, eco-duct, corbel), the majority (70-80%) of the camera traps were placed "above" the pathway and oriented slightly downward (Figure 57) in the axis of the pathway (to also cover the portion of the ground in front of the camera trap). This positioning does not restrict the width of the pathways at ground level and allows for the assessment of animal crossings over longer distances. However, it may momentarily disturb some individuals (see question on p. 73: "Does the presence of a camera trap impact the behaviour of animals using the structure?") who find themselves facing the cameras emitting "black" flashes and electronic noise (WEARN & GLOVER-KAPFER, 2017). This positioning is sometimes not

Figure 57 : Possible orientations of camera traps (in red) with regard to targeted pathways (in green). a) positioned "above", in the axis; b) positioned "at an angle", on one side; c) positioned "perpendicular" to the pathway.

preferred by tracking operators, as the animals then present little lateral movement for the sensors. JUMEAU *et al.* (2017), however, showed that for a Reconyx HC600 infrared detection system, this parameter (vertical or horizontal movement) had no effect on detection efficiency.

To avoid some of these biases, in medium-sized structures (greater than one metre), camera traps were positioned on one side and oriented "at an angle" (20° to 45°) to the path (Figure 57).

In underpasses, at least 2-3 metre wide, structures), the constraint of successfully covering the entire width of the structure according to the maximum range of the cameras sometimes requires positioning the camera on one side, oriented "perpendicular" to the path (Figure 57). This positioning is considered theoretically optimal for motion sensor operation (WEARN & GLOVER-KAPFER, 2017), as it maximises the amount of animal movement the sensor will detect. Because movements too close to the devices are quicker, some passages may then be missed.

In the case of the eco-bridges, there are multiple deployments (see *Rex 1*, p. 119-120) and these vary by concession holder and eco-bridge configuration. Between two and four camera traps (depending on the detection range of the cameras and the width of the scarp) are placed in the middle of the deck and oriented toward the centre of the structure to cover the entire width of the eco-bridge to detect as many taxa as possible (primarily large and medium-sized wildlife). A mounting height of 100 to 130 centimetres for these devices allows to limit the bias related to the growth of vegetation masking the objectives. The choice of positioning the devices in the middle of the deck allows for a better interpretation of the actual crossing of the eco-bridges by the animals. For small mammals (small Mustelids, hedgehogs, etc.), these camera traps are most of the time insufficient, because the detection distance becomes too great compared to the height of the individuals, and the vegetation can be too high. The choice can then be made to direct other camera traps on paths or corridors identified for these smaller and more stealthy species (along windrows, fences, etc.).

Camera traps oriented towards the areas surrounding the structures:

The first monitoring operation by camera traps carried out within the framework of the "Paquet Vert Autoroutier" (2009-2012) by VINCI Autoroutes aimed at positioning a camera trap inside the structure in order to assess their use by fauna. At the same time, other camera traps were placed at the entrance of the structures in order to estimate the share of species/individuals that failed to cross the adaptations. These follow-ups were finally abandoned because of too many technical constraints (e.g. theft of equipment, detection bias due to vegetation).

Indeed, some camera traps positioned at the entrances to the adaptations are then directed at vegetated surrounding areas, and untimely triggering on the vegetation ("false positives") systematically wears out the batteries of the cameras, whose autonomy is sometimes reduced to a few hours/days (Figure 58). Repeated theft of photo traps, more visible at the entrances to the structures, also prevents some of these monitoring operations.

4.4. MODELLING WEEKLY SPECIES OCCURRENCE IN WILDLIFE CROSSINGS

Modelling

To explain weekly occurrences (absence/presence) of species in structures, generalised linear mixed models (GLMM with binomial response and logit linkage; R Core Team, 2021, Bates *et al.*, 2015) were constructed, for underpass and eco-bridges respectively, testing the information provided by:

- descriptive variables of structures (i.e., dimensioning and adaptations of structures; cf. 3.1.3);
- descriptive variables of the structure environment (within a 500 metres, 1 kilometre, 5 kilometres and 10-kilometres radius around the structures; cf. 3.2);
- weather season (autumn, winter, spring, summer);
- biogeographical region (Mediterranean, Atlantic, continental region);
- the co-occurrences of humans and the main domestic species frequenting the structures, i.e. domestic cats and dogs.

To account for pseudo-replication and the spatial non-independence of the occurrence data, the structure identifier and a structure group identifier (defined according to the spatial autocorrelation of the data between structures) were included in the models as random variables.

For each species, all possible combinations of explanatory variables were tested, and the most informative models were selected. A consensus model was then built based on the average of the selected models (Barton, 2009).

For complete details of the analyses refer to Vacher *et al* (2022), available on request from VINCI Autoroutes.

Figure 58: Entrance of an 80 centimetre in diameter eco-duct overgrown with vegetation making monitoring the entrance by camera trap impossible. ©ASF/LPO France.

Selection of taxa

12 species of wild mammals (reduced to 11 taxa, because the data for Pine Marten, Weasels and *Martes sp.* were combined into one under the name "*Martes sp.*") were selected according to their frequency detected in the structures (minimum of 900 passages) and their respective occurrences in the underpasses (8 taxa) and eco-bridges (8 taxa):

- European Badger, *Meles meles*
- Red Fox, *Vulpes vulpes*
- European Roe Deer, *Capreolus capreolus*
- *Martes sp.* (European Beech Marten, *Martes foina* or European Pine Marten, *Martes martes*)
- European Rabbit, *Oryctolagus cuniculus*
- European Hare, *Lepus europaeus*
- European Wild Boar, *Sus scrofa*
- Coypu, *Myocastor coypus*
- Common Genet, *Genetta genetta*
- Red Deer, *Cervus elaphus*
- European Hedgehog, *Erinaceus europaeus*

To these wild species can be added the domestic cat, the domestic dog and humans.

V. INFLUENCE OF THE DESIGN (SIZING) AND ENVIRONMENT (SETUP) OF THE STRUCTURE

In order to answer questions about the influence of sizing or environmental variables on animals' use of wildlife crossings, the statistical model constructed (see §3.1.3., §3.2. and §4.4.) is applied. In the absence of significant results for certain issues, the input of scientific literature will shed light on the most important issues.
Some variables (notably fences, acoustic protection and light shafts) were not found to explain the occurrence of the observed species. They are therefore not discussed in the following chapters.

5.1. INTRODUCTORY FINDINGS: WHICH SPECIES USE THE STRUCTURES?

Table 5 summarises all mammal species detected in the structures.
A very large range of species is found in the monitored structures. Underpasses are more numerous, and logically involve more species than eco-bridges. However, eco-bridges are the only structures where the largest species of the French wildlife are observed (Red Deer, Grey Wolf in particular).

The most observed wildlife species in the structures are not surprisingly all common species throughout national territory, and include: the European Badger (27,375 observations in 133 structures), the Red Fox (23,369 observations in 109 structures) and the European Roe Deer (13,565 observations in 27 structures including 14 eco-bridges).
Rarer species, threatened at the national level, are also found, such as the European Polecat ("Vulnerable" on the IUCN National Red List, France *et al*. (2017), observed in 22 structures), or the Grey Wolf ("Vulnerable", observed on 2 eco-bridges).
Other locally rare species such as the European Otter (observed 55 times in 11 structures), Stoat (observed 19 times in 10 structures) are also present. Also noteworthy is the exceptional presence in an eco-duct of the Bouches-du-Rhône, of the Golden Jackal, a very mobile species coming from Central Europe, but still very rarely observed in France.

ORDER Family	Vernacular name	Scientific name	Total number of crossing	Number of structures used		
				Underpasses	Eco-bridges	Selected
CARNIVORA						
Mustelidae	European Otter	*Lutra lutra*	55	11	-	
Mustelidae	**Beech Marten**	**Martes foina**	**3 569**	**90**	**8**	**X**
Mustelidae	**Pine Marten**	**Martes martes**	**898**	**32**	**5**	**X**
Mustelidae	**Beech / Pine Marten**	**Martes foina/martes**	**3 687**	**93**	**10**	**X**
Mustelidae	**European Badger**	**Meles meles**	**27 375**	**109**	**14**	**X**
Mustelidae	Stoat	*Mustela erminea*	19	10	-	
Mustelidae	Weasel	*Mustela nivalis*	54	15	1	
Mustelidae	Polecat	*Mustela putorius*	170	22	3	
Mustelidae	Polecat/E. Mink/A. Mink	*Mustela putorius/lutreola/ vison*	4	4	-	
Mustelidae	Mustelid ind.	*Mustelidae sp.*	356	18	5	
Viverridae	**Common Genet**	**Genetta genetta**	**1 826**	**47**	**1**	**X**
Canidae	**Red Fox**	**Vulpes vulpes**	**23 369**	**133**	**15**	**X**
Canidae	Grey Wolf	*Canis lupus*	36	0	2	
Canidae	Golden Jackal	*Canis aureus*	2	1	-	
Canidae	**Domestic dog**	**Canis familiaris**	**988**	**65**	**14**	**(*)**
Canidae	Canids ind.	*Canidae sp.*	4	1	1	
Felidae	Wildcat	*Felis silvestris*	687	15	6	
Felidae	Wild /domestic cat	*Felis sp.*	160	16	1	
Felidae	**Domestic cat**	**Felis catus**	**10 614**	**102**	**14**	**(*)**
Procyonidae	Raccoon	*Procyon lotor*	3	3	-	
LAGOMORPHA						
Leporidae	**European Hare**	**Lepus europaeus**	**5 167**	**5 167**	**13**	**X**
Leporidae	Hare ind.	*Lepus sp.*	413	413	1	
Leporidae	**European Rabbit**	**Oryctolagus cuniculus**	**6 223**	**6 223**	**6**	**X**
Leporidae	Rabbit/Hare	*Leporidae sp.*	69	69	2	
	Lagomorph ind.	*Lagomorpha sp.*	3	3	-	
RODENTIA						
Muridae	Wood Mouse	*Apodemus sylvaticus*	184	5	3	
Muridae	Apodemus ind.	*Apodemus sp.*	202	4	-	
Muridae	Grey Mouse	*Mus musculus*	6	2	-	
Muridae	Mouse ind.	*Mus sp.*	4	1	-	
Muridae	Brown Rat	*Rattus norvegicus*	353	27	2	
Muridae	Black Rat	*Rattus rattus*	3	3	-	
Muridae	Rat ind.	*Rattus sp.*	806	37	1	

Table 5 : List of species detected in the structures: total number of crossings of species and structures where the species were detected and species selected for the analysis model.

ORDER Family	Vernacular name	Scientific name	Total number of crossing	Number of structures used		
				Underpasses	Eco-bridges	Selected
Muridae	Murid rodents ind.	*Muridae sp.*	224	25	1	
Cricetidae	Muskrat	*Ondatra zibethicus*	4	3	-	
Cricetidae	Bank Vole	*Clethrionomys glareolus*	14	2	-	
Cricetidae	Vole ind.	*Arvicola sp.*	33	6	-	
Cricetidae	*Arvicolinae* ind.	*Arvicolinae sp.*	10	3	-	
Myocastoridae	**Coypu**	**Myocastor coypus**	**1 902**	**71**	**3**	**X**
Sciuridae	Red Squirrel	*Sciurus vulgaris*	452	20	2	
Gliridae	Garden Dormouse	*Eliomys quercinus*	48	2	-	
Gliridae	Hazel/Garden Dormouse s ind	*Leithiinae sp.*	1	1	-	
	Rodent ind.	*Rodentia sp.*	46	4	-	
EULIPOTYPHLA						
Erinaceidae	**European Hedgehog**	**Erinaceus europaeus**	**1 137**	**56**	**5**	**X**
Soricidae	Shrew ind.	*Soricidae sp.*	27	3	-	
Talpidae	European Mole	*Talpa europaea*	1	1	-	
CETARTIODACTYLA						
Cervidae	**Roe Deer**	**Capreolus capreolus**	**13 565**	**27**	**14**	**X**
Cervidae	**Red Deer**	**Cervus elaphus**	**1 605**	**0**	**5**	**X**
Bovidae	Chamois	*Rupicapra rupicapra*	1	0	1	
Bovidae	Domestic goat	*Capra hircus*	31	2	-	
Bovidae	Domestic sheep	*Ovis aries*	13	2	2	
Suidae	**Wild Boar**	**Sus scrofa**	**5 489**	**31**	**15**	**X**
Equidae	Domestic horse	*Equus caballus*	1	0	1	
CHIROPTERA						
	Bat ind.	*Chiroptera sp.*	165	35	1	
PRIMATES						
Hominidae	**Human**	**Homo sapiens**	**2662**	**48**	**14**	**(*)**
INDÉTERMINÉS						
	Mammal ind.	*Mammalia*	805	65	9	
	Small mammal ind.	*Micromammal sp.*	642	39	7	

(*) Selection as explanatory variables of co-occurrences of wild species in and on the structures.

Figure 59 : Number of crossings by the 16 most detected identified species during monitoring operations (all structures combined)


European Badger: 27 375 ©E. Rondeau


Red Fox: 23 689 ©E. Rondeau


Roe Deer: 13 565 ©E. Rondeau


European Rabbit: 6 223 ©VINCI Autoroutes


Wild Boar: 5 485 ©VINCI Autoroutes


European Hare: 5 167 ©E. Rondeau


Pine and Beech Martens: 3687 ©E. Rondeau


Coypu: 1 902 ©E. Rondeau


Common Genet: 1 826 ©E. Rondeau


Red Deer: 1 605 ©E. Rondeau


European Hedgehog: 1 137 ©VINCI Autoroutes


Wildcat: 687 ©E. Rondeau


Polecat : 170 ©E. Rondeau


European Otter: 55 ©VINCI Autoroutes


Weasel : 54 ©VINCI Autoroutes


Grey Wolf : 36 ©VINCI Autoroutes

Figure 60 : Specific observations

Foxes mating on an eco-bridge

A fox and a Polecat face to face in an eco-duct

Duck and ducklings in an eco-duct

A fox facing a frog

Badger rolling in an eco-duct

Donkey in a 120-cm- diameter eco-duct

Bat in a 120-cm-diameter eco-duct

Raccoon in an eco-duct

Figure 60 : The use of camera traps allowed the observation of remarkable species and behaviours. In total, more than 500,000 photos were taken in the structures

Butterfly in an eco-duct

Roe Deer sparring on an eco-bridge

Hare grazing on an eco-bridge

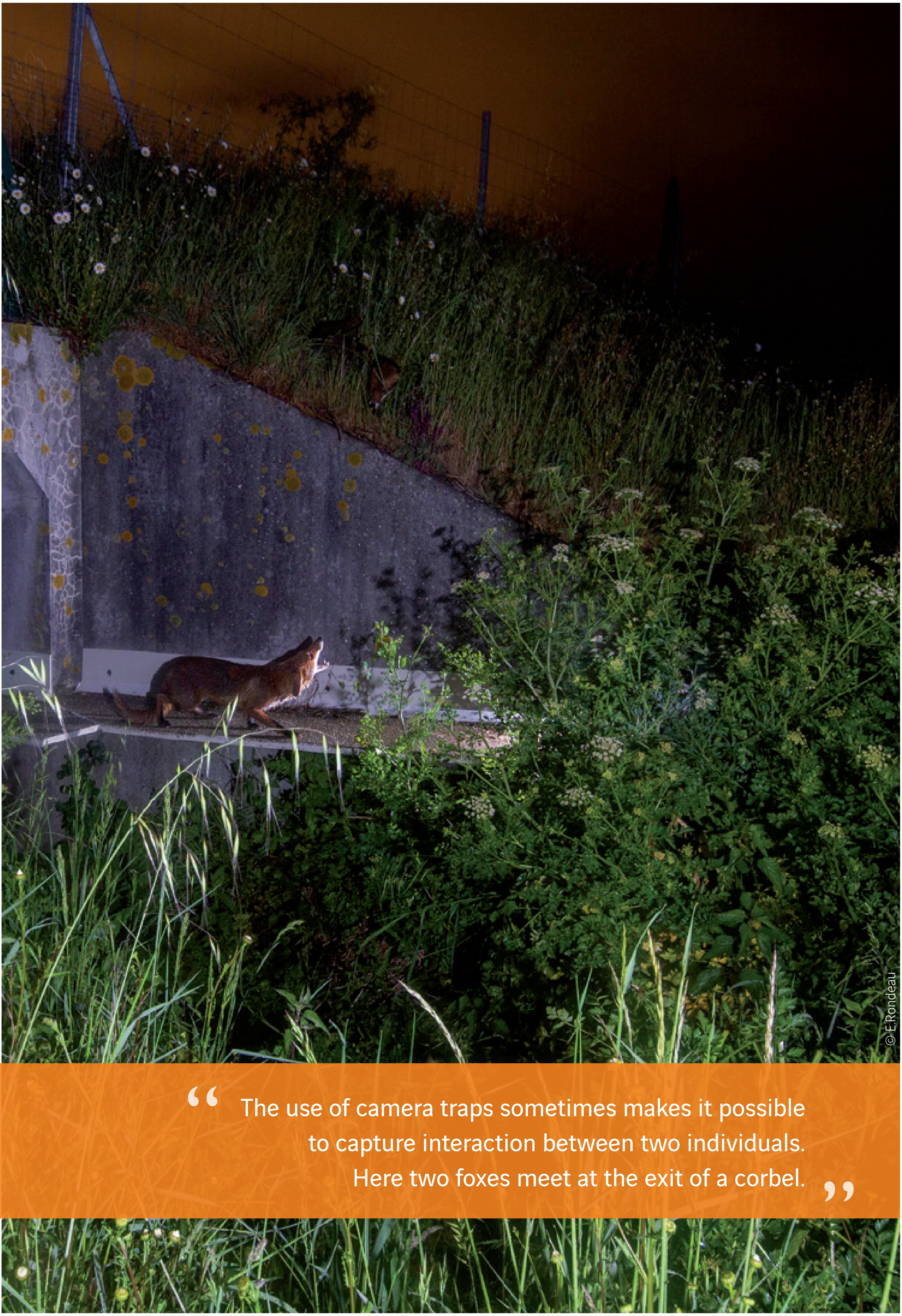
Garden Dormouse on a corbel of a hydraulic structure

Family of Wildcats crossing an eco-bridge

Buzzard hunting on an eco-bridge

Golden Jackal – first observation in an eco-duct in the Bouches-du-Rhône

Beech Marten pulling the corpse of a fellow Beech Marten on the corbel of a hydraulic structure



“ The use of camera traps sometimes makes it possible to capture interaction between two individuals. Here two foxes meet at the exit of a corbel. ”

5.2 DO THE DIMENSIONS OF THE STRUCTURES HAVE AN IMPACT ON FREQUENCY OF WILDLIFE USE?

The model of the present study does not show a significant influence of the variables of the dimensions of the structures (usable height and width for underpasses; width for the eco-bridges) on the frequency of species. One of the limitations of this analysis is the relative homogeneity of the sizing of the structures, most of which have a standardised design. Extreme cases (very large structures such as eco-bridges) are too few in number to enable statistical analysis.

Concerning length, the variable was not retained in the framework of our study because of the lack of a sufficient number of structures of different lengths to perform a statistical analysis.

However, *Rex 1* noted that the 9 longer-than-80-metre underpasses seemed to be less frequented than the rest of the structures and that they were essentially frequented by burrowing species more inclined to use structures with a significant tunnel effect (*Rex 1*, p. 122).

5.3 WHAT SIZING FOR AN ECO-BRIDGE?

Based on analysis of VINCI Autoroutes structures

The analyses carried out within the framework of *Rex 2* do not yield significant results on this question. Based on scientific literature, one of the limitations of the analysis probably lies in the number of eco-bridges monitored (15), which finally have very standardised widths between 11 and 25.8 metres (2/3 of the structures have a usable width between 15 and 25 metres). The average number of crossings detected per day seems to increase with the width of the eco-bridges, but this observation needs to be confirmed by additional analyses.

Based on the scientific literature

The question of the sizing of eco-bridges often remains a central issue for planners who must find a compromise between sufficient passability and at an acceptable cost. This question is therefore developed here, in particular through existing scientific literature.

However, the question of efficiency remains complex and there are few studies conducted on the subject. Moreover, they most often deal with sizing from the point of view of use rather than the efficiency of the structures. However, a structure can be effective while being little used. This is the case for genetic mixing, where the passage of a few individuals (or even a single individual in the case of large species such as deer) may be sufficient to maintain a genetic flow, but insufficient to maintain viable populations on either side of the infrastructure. The question then becomes: how many crossings are needed for a structure to be effective? This will depend on the species involved, the dynamics and the conservation status of its populations in the area where the structure in question is located.

Pfister *et coll.* (1999) in a study carried out in Europe on 21 eco-bridges with widths ranging from 3.4 metres to 186 metres demonstrated that the narrowest eco-bridges (< 20 metres) were significantly less used than the wider ones. On this basis, they recommend a width of at least 50 m to ensure the crossing of large mammals for stress-free daily use of the structures.

COST341 (Iuell *et coll.* 2003) recommends an optimal 40 to 50-meter wide eco-bridges for large wildlife in Europe. This width can be reduced to a minimum of 20 meters if the purpose is only to provide corridor movement for less sensitive species, or when the topography has a channelling effect leading animals directly to the crossing.

In North America, the recommendations are of the same order of magnitude (minimum of 40-50 meters wide for 50-70 meters recommended; Clevenger & Huijser, 2011).

This type of study should now be considered again on a European scale with a larger range of structures, particularly given the greater number of structures and the evolution of monitoring techniques.

In Europe, the majority of eco-bridges are between 25 and 80 metre wide, and the optimal width depends on the species that might use the

The recent CEREMA guide published in 2021 deals with the subject of sizing structures. Pages 95, 102 and 103 of this document recommend the widths of structures according to the level of ecological continuity at stake.
Cerema. *Les passages à faune. Préserver et restaurer les continuités écologiques avec les infrastructures linéaires de transport.* Bron: Cerema, 2021. Collection: références.
ISBN: 978-2-37180-525-5 (PDF)

structure as a major ecological corridor, which is why it is ultimately difficult to determine a generic optimal width. The minimum width of the eco-bridge must therefore be adapted both to the requirements of mobile (large) target species that regularly cross the structure, and to the requirements of less mobile (small) target species that use the wildlife crossing as a habitat continuum, sometimes over several generations (Van der Grift *et al.*, 2011). The identification of the so-called "dimensioning" species, heritage species, green and blue belt species, from the regional to the local level is then a key element to define the sizing of the structure and its surface adaptations.

The Red Deer, the largest wild terrestrial mammal in France, is detected on 5 of the 6 eco-bridges monitored on the VINCI Autoroutes network and located in the immediate vicinity of or within ranges where the species is found (presence data by range from the OFB/FNC/FDC Wild Ungulates network). The occurrence of deer is certainly low (mostly < 5% of the days monitored), but one structure stands out (Figure 61) with a presence detected on 40% of the days monitored. This 18.5-metre-wide structure is certainly used by the species, but what would have been its use if the structure had been wider, within the dimensions recommended in scientific literature? The study by Pfister & Birrir (1991) shows that use by Red Deer increases substantially in structures wider than 30 metres.

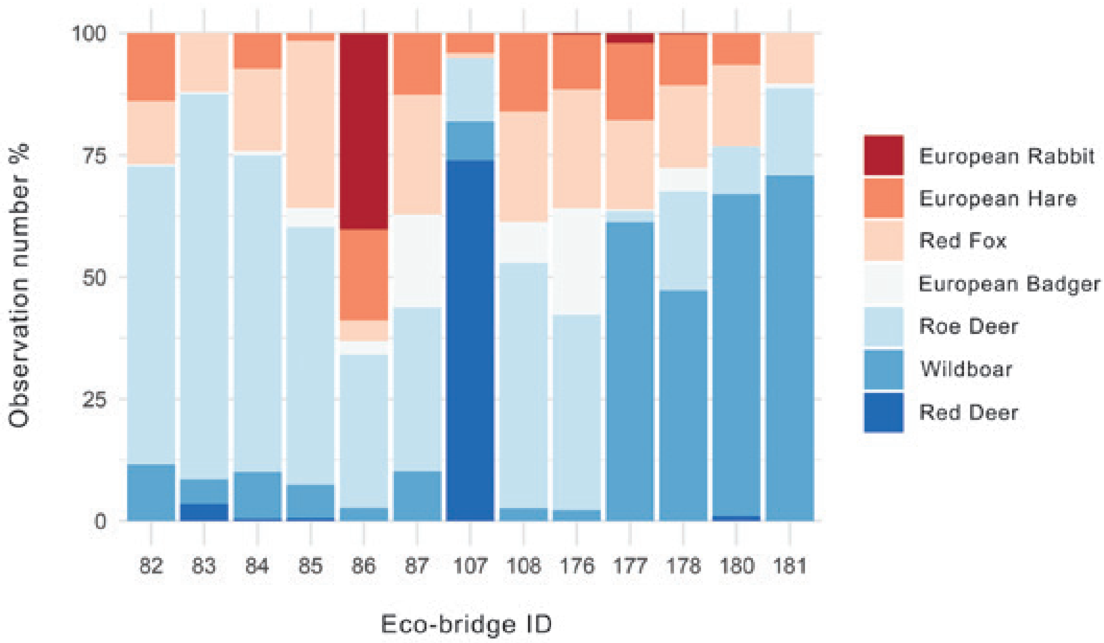


Figure 61 : Proportion of number of observations of the seven most frequent species in all eco-bridges.

Figure 62: Red Deer on an eco-bridge in Charente-Maritime. ©ASF/FDC17.

However, the results of the study by Pfister *et al* (1999) do not demonstrate that it is systematically necessary to build a very wide eco-bridge. The same authors report in a previous study (1997) that in certain circumstances it may be more appropriate to build several smaller (but still sufficiently wide) wildlife crossings, because the area of attraction of a point is "limited by individuals' areas of action". The dilemma remains between whether to have one large or several smaller wildlife crossings (Helldin, 2022), and regional landscape characteristics (e.g., landscape heterogeneity) and fauna characteristics (e.g., specific sizing for target species) make it difficult to generalise.

5.4 DO THE DIFFERENT TYPES OF STRUCTURES INFLUENCE THE FREQUENCY OF FAUNA?

Within the adaptations for small and medium-sized fauna, the higher use of eco-ducts compared to adapted hydraulic structures was already mentioned in Rex 1 (p. 84). The model analyses also point in this direction for certain species, showing for example that the European Badger makes greater use of eco-ducts than hydraulic structures equipped with footways or corbels (Table 6).
This can be explained by the fact that dry footways and corbels overhang a river, most often with water. This proximity on a confined several dozen metre long corridor is not attractive for certain non-aquatic species, such as badgers. On the other hand, it is perfectly suitable and favourable, for example, for semi-aquatic mustelids seeking this proximity.

The type of structure can therefore act as a filter for certain species. The choice of the adaptation must therefore be made according to the target species and their ecology.

It should be noted, however, that there are sometimes surprising behaviours: the monitoring of a metal culvert in the Aude showed that a structure partially and temporarily flooded by an intense rainfall could still be used by the European Badger, which is probably accustomed to using this path (Figure 63), regardless of the conditions.

In addition, occasional crossings of European Roe deer in 120-centimetre culverts or wild boar in an 80-centimetre culvert also seem surprising (*Rex 1*, p. 65).

Table 7 compares the frequency of the 11 species (Beech Marten and Pine Marten combined) most represented in the different types of structures. The trends mentioned for the European Badger, for example, are also verified here by a much higher number of crossings per year in the specific underpasses (eco-ducts type) than in the adapted hydraulic structures.

Coypu also reflects the importance that non-dedicated hydraulic structures (207 crossings detected on average per year by structure) can have for semi-aquatic species. However, in periods of high water, these structures become impassable for terrestrial species and can be very difficult to cross for semi-aquatic species (sometimes strong current).

The figures of eco-bridge frequentation show the importance of this type of adaptation for large fauna (Red Deer, European Roe Deer and European Wild Boar) since the deer is logically exclusively detected on eco-bridges, and the 2 other species are very seldomly (or not at all) detected on the other types of structures. To a lesser extent, this observation is also made for lagomorphs (European Hare and European Rabbit) whose average numbers are higher on eco-bridges than on other types of structures.

Species	Variable	Estimation	ES	Valeur-p
Meles meles European Badger	**OHA - Footway**	**-2.938**	**1.247**	**0.019**
	OHA - Corbel	**-2.998**	**1.240**	**0.016**
	OHA - Rockfill	-0.195	1.600	0.903
	Mixed underpass	-3.771	2.247	0.093
	Non-dedicated underpass	-13.118	349.510	0.970
	Specific fauna underpass	**1.749**	**0.803**	**0.029**
	Usable height	-0.127	0.453	0.779

Table 6 : Variables selected for sizing and design of structures as most explanatory of the occurrence of certain species in underpasses. Results in bold indicate significant effects of variables on species occurrence. According to these results, badgers use eco-ducts significantly more than other hydraulic, footway, or corbel structures. The Estimate, ES, and p-value columns give the coefficient estimates for the variables selected as the most explanatory, their standard errors, and their probabilities in the models. The species not mentioned do not have any design/sizing variable retained as explanatory during the selection of the models.

5.5 DO UNDERPASSES CONTRIBUTE TO BAT CROSSINGS?

The diversity of the protocols implemented and point data obtained during our study could not be statistically analysed to answer this question. However, the acoustic and photographic trap monitoring set up in certain structures allowed us to highlight the use of underpasses, hydraulic structures or dry culverts dedicated to terrestrial fauna, by numerous species of bats. Eight underpasses (gantries, culverts, viaducts) were monitored with ultrasonic detectors by the *Écologistes de l'Euzière* on the project to

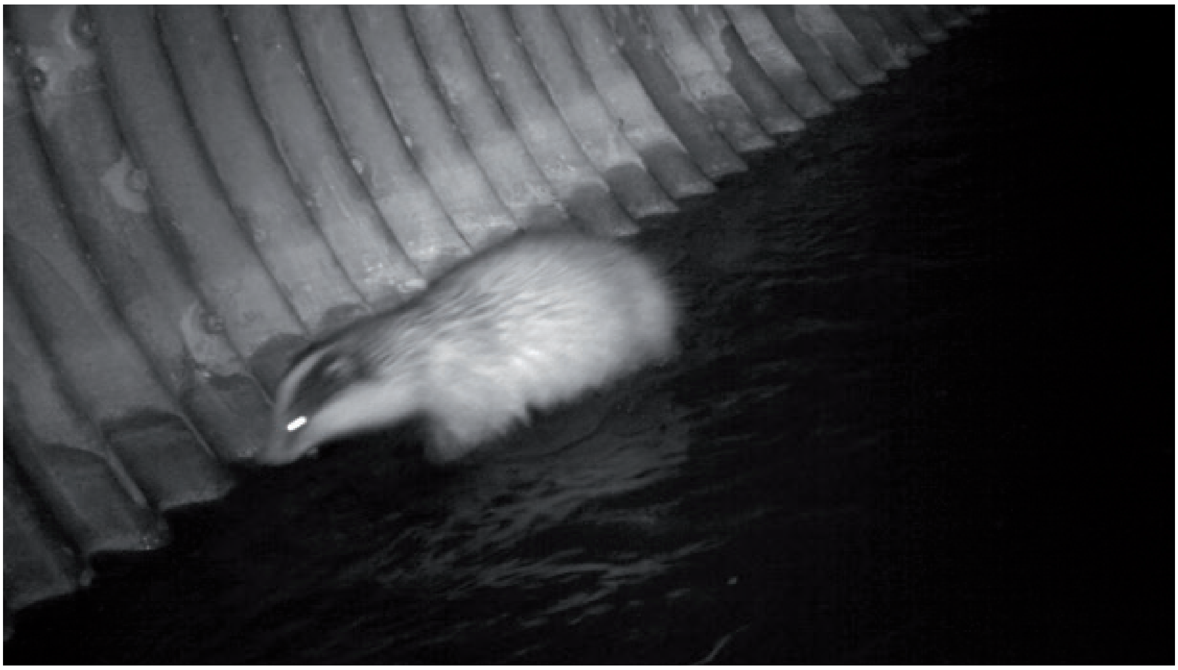
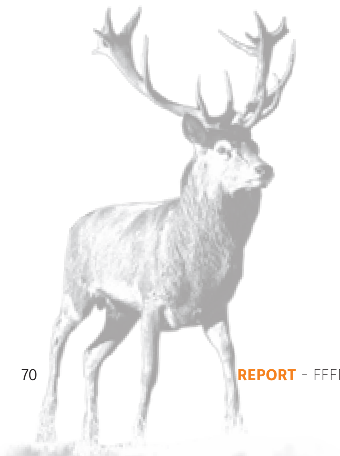


Figure 63 : European Badger crossing a flooded metal culvert. ©Nature en Occitanie - M. BELAUD.

Figure 64 : Point data of an unidentified bat filmed in transit in a 120 cm-diameter culvert. ©ASF/LPO France.

No. crossings detected.an-1.structure-1 (Mean ±Standard deviation [Min-Max] n=number of structures)						
Taxa	**PI specific fauna (ecoduct)**		**OHA-Footpath**		**OHA-Corbel**	
Beech/Pine Marten	35	±ET50 [1-375] n=85	84	±ET136 [0-536] n=20	22	±ET31 [0-93] n=9
Badger	156	±ET209 [0-1154] n=80	11	±ET20 [0-51] n=6	21	±ET41 [0-83] n=4
Common Genet	26	±ET28 [0-105] n=23	5	±ET6 [0-15] n=5	15	±ET18 [1-52] n=8
Red Fox	81	±ET181 [1-1492] n=87	32	±ET49 [1-156] n=17	6	±ET6 [0-16] n=6
Hedgehog	14	±ET27 [0-148] n=41	4	±ET3 [2-8] n=5	0	±ET [0-0] n=1
European Hare	35	±ET85 [0-410] n=45	7	±ET [7-7] n=1	0	±ET [0-0] n=2
European Rabbit	28	±ET36 [0-98] n=27	-	-	1	±ET [1-1] n=1
Coypu	20	±ET65 [0-417] n=43	17	±ET24 [1-61] n=9	6	±ET6 [1-19] n=10
Roe Deer	12	±ET29 [0-117] n=21	1	±ET0 [1-1] n=2	0	±ET [0-0] n=1
Red Deer	-	-	-	-	-	-
Wildboar	5	±ET6 [0-26] n=20	-	-	-	-
Domestic dog	7	±ET11 [0-54] n=36	6	±ET5 [1-15] n=10	1	±ET1 [0-3] n=3
Domestic cat	50	±ET84 [0-395] n=63	94	±ET107 [0-335] n=12	5	±ET8 [0-24] n=7
Man	8	±ET23 [0-110] n=24	25	±ET24 [4-87] n=15	2	±ET2 [0-4] n=5

Table 7: Average number (±standard deviation) of crossings detected per year and per structure, [Minimum and Maximum], n= number of structures in the selected dataset for the analysis of weekly species occurrence in structures.



No. crossings detected.an-1.structure-1 (Mean ±Standard deviation [Min-Max] n=number of structures)								
Taxa	OHA-Notch		OH non-dedicated		Eco-bridge		PI mixed	
Beech/Pine Marten	12	±ET10 [1-21] n=3	37	±ET55 [1-191] n=20	15	±ET21 [0-56] n=12	8	±ET5 [1-13] n=4
Badger	12	±ET17 [1-32] n=3	27	±ET50 [1-161] n=15	17	±ET23 [1-85] n=14	0	±ET [0-0] n=1
Common Genet	-	-	8	±ET14 [1-45] n=9	1	±ET [1-1] n=1	1	±ET1 [0-1] n=2
Red Fox	9	±ET0 [9-9] n=1	83	±ET163 [1-585] n=19	75	±ET74 [4-292] n=15	14	±ET20 [2-37] n=3
Hedgehog	1	±ET [1-1] n=1	8	±ET6 [1-18] n=6	1	±ET1 [0-3] n=5	95	±ET132 [1-188] n=2
European Hare	2	±ET [2-2] n=1	1	±ET2 [1-5] n=6	62	±ET78 [4-232] n=13	21	±ET42 [0-84] n=4
European Rabbit	-	-	2	±ET2 [0-7] n=8	98	±ET233 [1-575] n=6	14	±ET [14-14] n=1
Coypu	5	±ET [5-5] n=1	207	±ET530 [1-1408] n=7	0	±ET0 [0-1] n=3	29	±ET [29-29] n=1
Roe Deer	0	±ET [0-0] n=1	-	-	174	±ET165 [7-498] n=14	12	±ET15 [1-23] n=2
Red Deer	-	-	-	-	92	±ET192 [1-435] n=5	-	-
Wildboar	-	-	15	±ET16 [0-42] n=8	292	±ET467 [1-1466] n=15	1	±ET1 [0-2] n=2
Domestic dog	1	±ET [1-1] n=1	7	±ET6 [1-18] n=12	15	±ET20 [1-74] n=14	3	±ET1 [2-3] n=2
Domestic cat	1	±ET [1-1] n=1	86	±ET90 [1-254] n=16	34	±ET48 [0-155] n=14	41	±ET26 [23-60] n=2
Man	39	±ET [39-39] n=1	23	±ET22 [4-47] n=3	107	±ET113 [5-351] n=14	-	-

relocate the A9 motorway, near Montpellier. The microphones positioned in the structures and outside identified up to 16 bat species and identified the passage of several species under the motorway.

Existing feedback on the subject:

Bat species exhibit different flight behaviours depending on their ecology. For example, the Schreiber's Bent-winged Bat is a rather aerial species, whereas the horseshoe bats move more at low altitude (≤ 2 metres) by following the structural elements of the landscape (Arthur L. & Lemaire M., 2009). They are therefore more likely to encounter the entrances of underpasses at low altitude.

The recommended diameter to facilitate the passage of bats in underpasses therefore depends largely on the issues relating to the species present on site. For bats, the importance of the location of structures in relation to the landscape and to the flight corridors of the species appears to be crucial (LAFORGE *et al.*, 2019).

For species that move more at higher altitudes and are rarely seen in underpasses, specific overpasses (gantries, bat-bridges) can become complementary to underpasses, especially on flight routes known before the implementation of the autoroute. Such structures have been thoroughly studied within the framework of VINCI Autoroutes monitoring (see Chapter 8.2).

5.6 DOES THE LANDSCAPE IN THE VICINITY OF THE ADAPTATIONS INFLUENCE THE USE OF THE STRUCTURES BY WILDLIFE?

Two variables that may reflect the effect of landscape fragmentation around structures have been used for analyses: road and hedge density.

Only two species out of the 12 studied emerged as having an occurrence dependent on this fragmentation. Thus, these two variables influence the weekly occurrence of Wild Boar in underpasses. The higher the road density, the lower the probability of occurrence of the species. Conversely, as the density of hedges increases, the probability of occurrence of the species increases (Figure 66).

Among the other species, only the European Rabbit shows an increasing probability of occurrence with the density of hedges. This effect, although significant, is however low (Figure 67). Rabbits burrow in hedgerow banks. The species is therefore favoured in hedgerow landscapes containing dense hedgerow networks providing it with the two habitat compartments necessary for its life cycle, open habitats for foraging, and hedgerows for digging burrows (Lombardi *et coll.*, 2003, 2007).

Other landscape parameters have a significant influence on certain species. The effects seem to be more pronounced at small scales, i.e., within a radius of 500 metres to 1 kilometre. The probability of occurrence of the Eurasian Hare increases with the percentage of grassland and decreases with the percentage of fodder crops (Figure 69). The European hare is a fan of low vegetation open areas. However, although agricultural environments may be favourable to the hare, the action of mechanical mowing of pastures is detrimental to it (MILANOV, 1996).

The probability of occurrence of the European Hedgehog increases significantly with the percentage of agricultural land within a 1-kilometre radius (Figure 68). This may seem counterintuitive since this is one of the habitats with the least dense hedgehog populations. However, it is probably in these transit agricultural environments that the species makes its greatest

Figure 65: Example A83 (Deux-Sèvres) intersecting an obvious ecological corridor (wooded former railway) in the middle of an intensive agricultural landscape. ©IGN, 2002.

CEN PACA study on monitoring Chiroptera in A8 autoroute underground structures (PICHARD *et al.*, 2012).

The monitoring of 15 underpasses of the A8 motorway with diameters varying between 0.93 to 4 metres has shown a positive correlation between the diameter of the structure and the number of bat contacts (Figure 64) (Pichard *et al.* 2012).

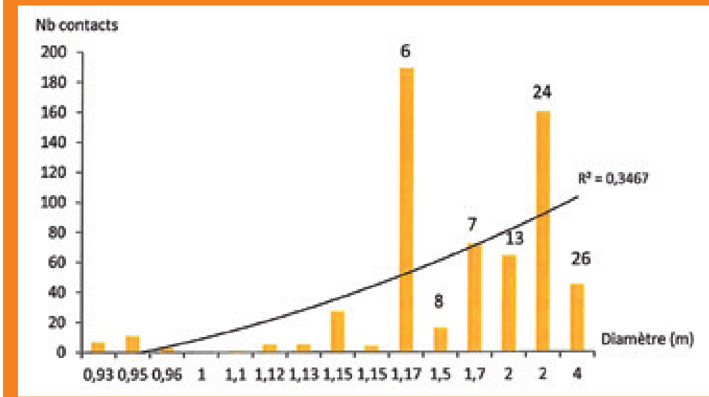


Figure 64 : Graph of total number of contacts in function of structure diameter

In this study, among the species present in the sector, the Lesser Horseshoe Bat, for example, used 14 of the 15 structures, while a species such as the Schreiber's Bent-winged Bat, although definitely present in the vicinity, was not detected in any of the structures.

Only the structures of more than 1.5 metres in diameter were used by other species apart from horseshoe bats.

Biotope/Setra study summarising bat data gathered on over 85 underground passges.

In the CEREMA Chiroptères et infrastructures de transports, guide Nowicki & Rousselle refer to the work of Biotope (Biotope/SETRA, 2011) summarising the data gathered on over 85 underpasses. This work shows that:

- Horseshoe bats may frequent small structures (1 metre). Frequentation reaches its maximum for heights higher than 3 metres.
- Pipistrelles do not use structures less than 3 metres high and maximum frequentation is reached at 6 metres high and 40 metres wide.
- Mouse-eared bats do not use structures less than 2-metres high and maximum frequentation is reached at over 5-metres high.

movement distances, thus increasing its chances of using wildlife crossings.

This observation reinforces the need to also manage degraded territories such as intensive agricultural environments where food resources and refuge areas are less available, but where, as a result, travel distances for wildlife increase.

5.7 DOES THE PRESENCE OF RIVERS HAVE AN EFFECT ON FREQUENCY?

Numerous studies have highlighted the importance of rivers as ecological corridors for terrestrial wildlife (NAIMAN *et al.*, 2005), in addition to aquatic and semi-aquatic wildlife. Rivers associated with riparian vegetation and forests increase the permeability of fragmented landscapes. They are also key elements of ecological functionality to be taken into account in the multi-criteria analysis to guide the selection of the best location for a wildlife crossing prior to its construction.

Therefore, the use of a structure located in a river corridor is directly influenced by the use of this corridor.

In our study, the influence of the river corridor on

Wildboar - Summer

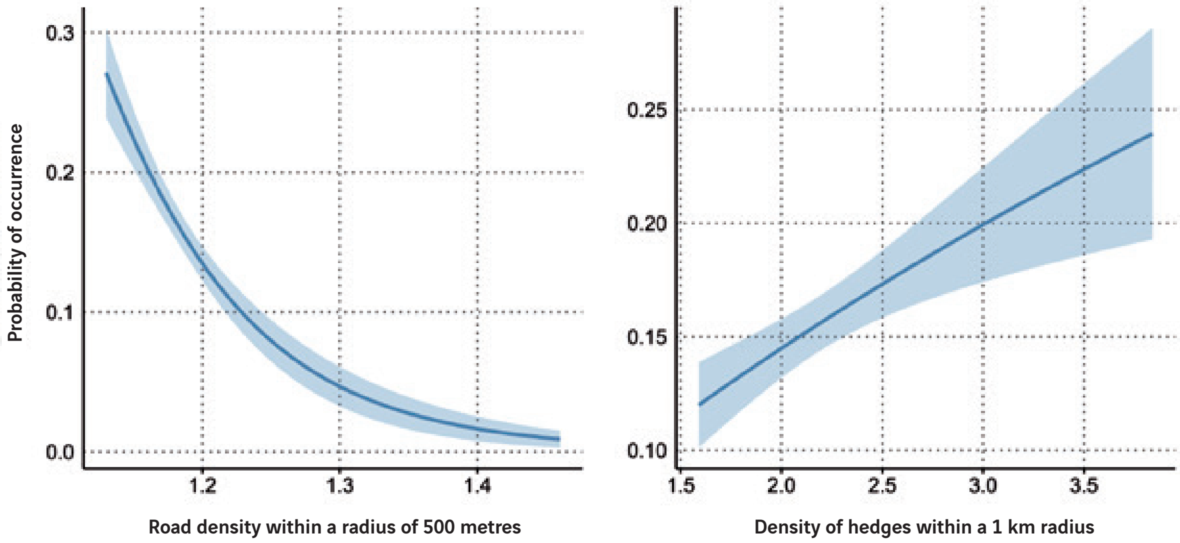


Figure 66: Relation between the occurrence of Wild Boar in the summer on overpass motorway structures (eco-bridges), and "Road density over 500 m" and "Hedge density" over 1 km" variables.

European Rabbit

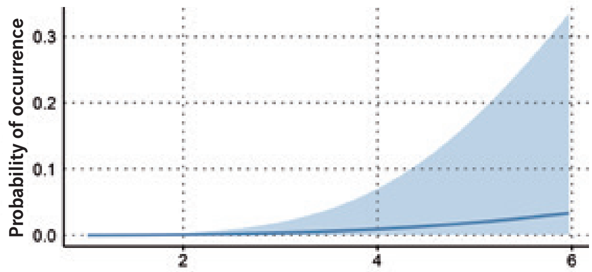


Figure 67: Relation between the occurrence of European Rabbit in motorway underpasses and the "Hedge density within a 10 km radius" variable.

European Hedgehog - Spring

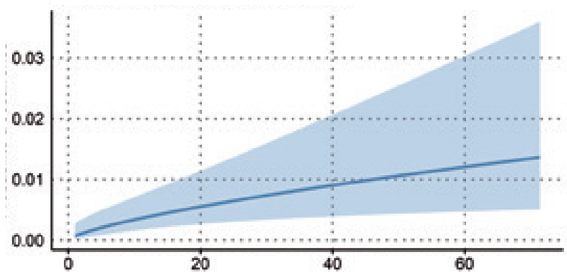


Figure 68 : Relation between the occurrence of European Hedgehog in the spring in motorway underpasses and the "Percentage of agricultural land over 1 km" variable .

European Hare - Summer

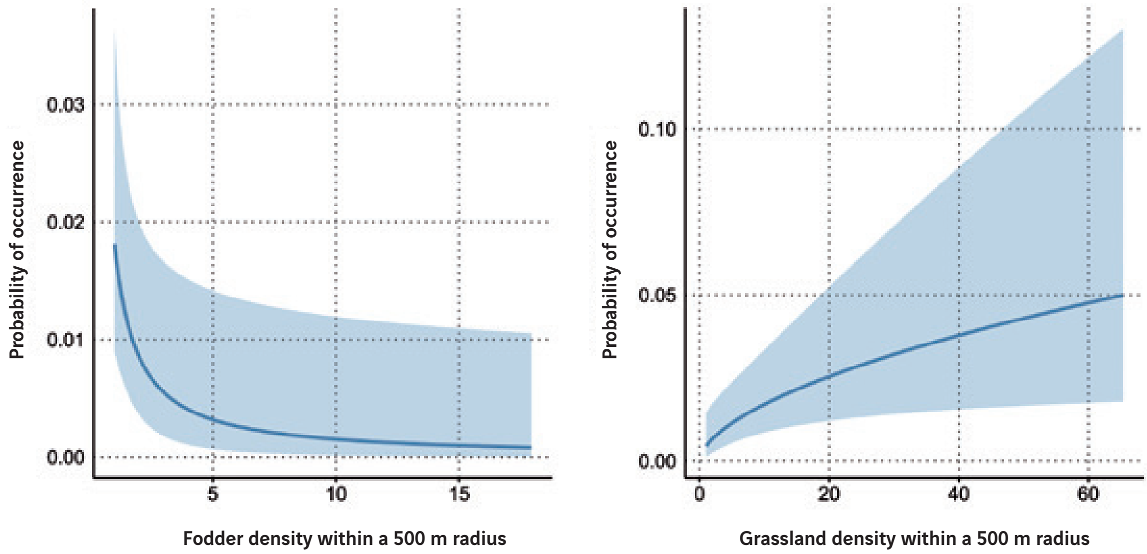


Figure 69 : Relations between the frequency of European Hare in summer and motorway underpass crossings, and "percentage of fodder land over 500 m" and "percentage of pasture over 500 m" variables.

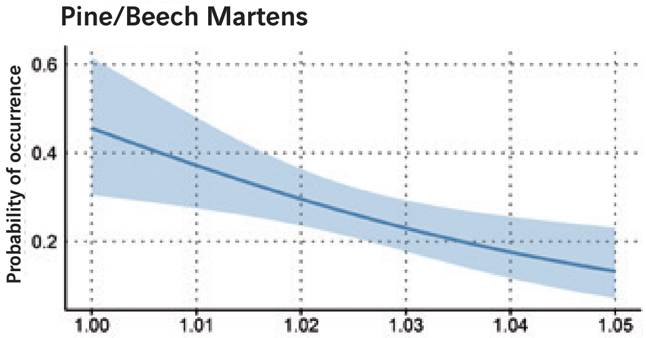


Figure 70: Relation between frequency of Martes sp. In motoway underpass crossings, and "River density over 10 km" variable.

motorway structures designed for wildlife was not tested. There are two major obstacles to the interpretation of these data, separate from the influence of the corridor:

- The distance between the river corridor and the entrances to the adaptations, as well as all the parameters that may influence the connection between the two (difference in level, accessibility, artificial connections, vegetation, etc.).
- The engineered structure itself that can, depending on its nature and/or dimensions, act as a filter for certain species. For example, *Rex* 1 showed that the use of hydraulic structures (dry feet inside the hydraulic structure) was half as high as eco-ducts (*Rex 1*, p. 69).

In our analysis model, however, river density was taken into account. The result is that the probability of occurrence of the genus *Martes* (Beech Marten and Pine Marten) in structures decreases significantly with increasing river density (Figure 70). It can be hypothesised that the greater the density of rivers around a structure, the greater the opportunities to use other corridors and pathways than the structure itself.

5.8 WHAT IS THE AREA OF INFLUENCE OF A WILDLIFE CROSSING?

This is an important issue for planners as it may affect the number and placement of wildlife crossings and could not be addressed in the analyses of this study, but it is well documented in scientific literature.

Individuals potentially use the structures if they encountered them during their movements. It is therefore understandable that the number and spacing of wildlife crossings along the motorway are of critical importance in reducing the barrier effect of linear transportation infrastructures (Karlson *et al.*, 2017).

Studying roadkill locations can help determine wildlife crossing densities in a project. Based on this, Clevenger *et al.* (2002) recommend wildlife underpasses every 150 to 300 metres for new road projects. In Spain, the recommendation in the case of forest environments and other habitats of importance for the conservation of ecological connectivity states a crossing every 500 metres for small fauna (1 kilometre in the case of anthropised habitats, including crops and peri-urban areas), and a crossing every kilometre for large fauna (3 kilometres in the case of anthropised habitats) (Ministry of Agriculture Food and the Environment, 2016). These recommendations should be contextualised according to the species present and the landscape characteristics of each project.

The study of animal movement patterns is also useful for defining the area of influence of wildlife crossings in the area, and to help define their optimal number and spacing. Some authors consider that individuals will not be able to encounter a wildlife crossing if it is located outside their home range. An animal is then supposed to be able to regularly use a wildlife crossing if it is within its daily travel distance.

For Seiler *et al.* (2015), the square root of a species' home range over area gives a good approximation of the daily mobility of individuals. A wildlife crossing in Sweden is thus considered to mitigate the road/rail barrier effect for elk along 2 + 2 kilometres (i.e., the square root of 20 $km^2 \approx 4$) in each direction of the structure. This approximation is then used to determine the number and spacing of wildlife crossings required in the concerned sections. For long-distance dispersal movements, a single efficiently located structure can theoretically produce sufficient connectivity.

Based on scientific literature, the area of influence of a wildlife crossings appears to be directly related to the daily movement capacity of target species.

VI. INFLUENCE OF ECOLOGY OF SPECIES (SEASONALITY, INTERACTION, BEHAVIOUR)

6.1 DOES THE USE OF STRUCTURES BY HUMANS OR DOMESTIC A HAVE AN IMPACT ON THE USE OF STRUCTURES BY WILDLIFE?

Phenology graphs of the 11 most detected species in the structures confirm that the majority of species use the crossings much more frequently at night (Figure 72).

Analyses conducted on the underpasses show the existence of interactions between the weekly occurrences of wildlife and the occurrences of cats, domestic dogs and humans.

The probability of wildlife occurrence decreases with the occurrence of domestic cats (without distinction between house cats and feral cats) for at least three species: the European Rabbit, the European Hare and *Martes sp*. (Figure 71). This interaction had been shown by Mata *et al*. (2020), the avoidance of cats by lagomorphs being explained by the repellent effect of cat odours or droppings for lagomorphs.

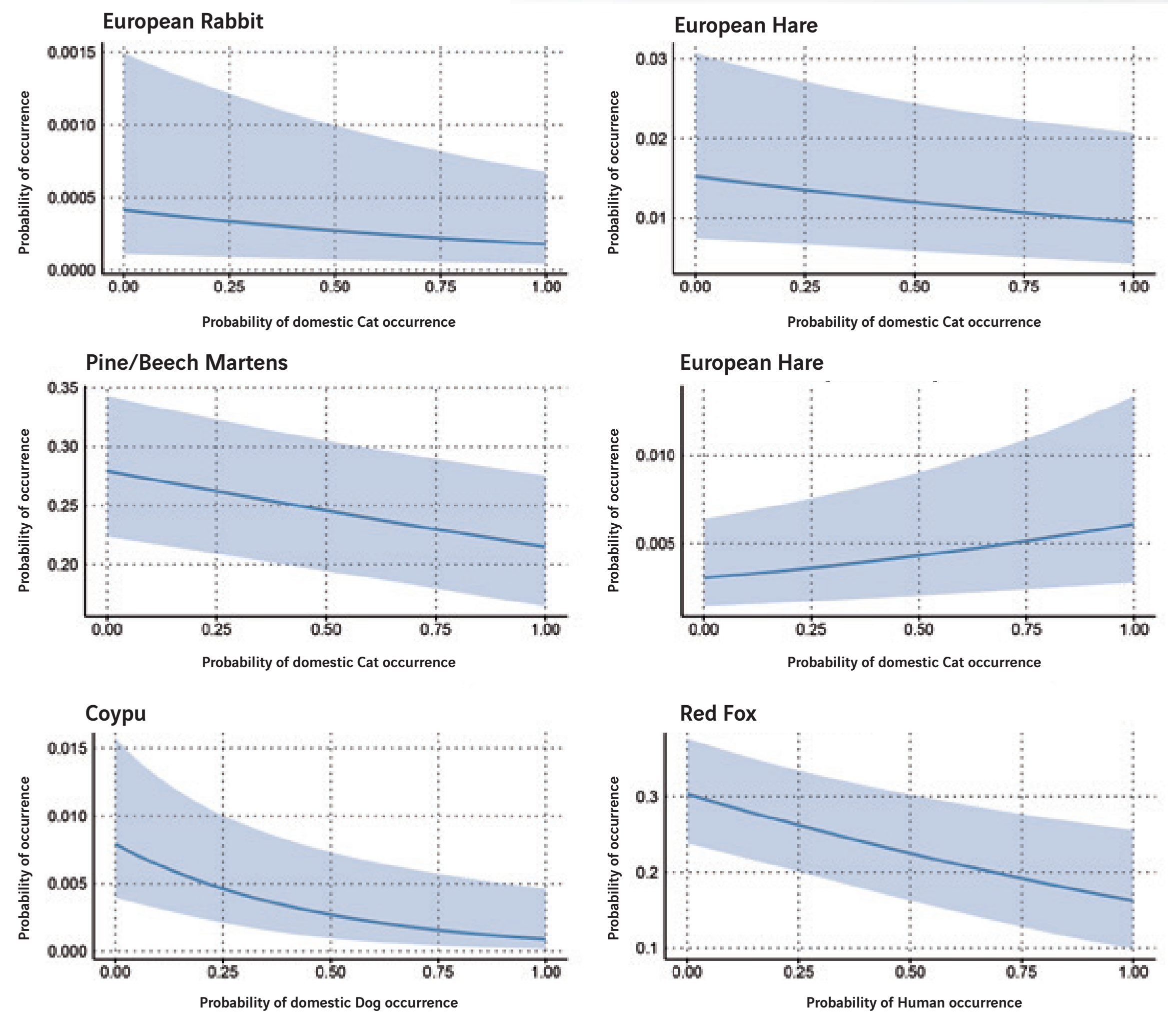


Figure 71: Relations between the occurrence of species in motorway underpass crossings, and the probability of occurrence of domestic cats, dogs or humans.

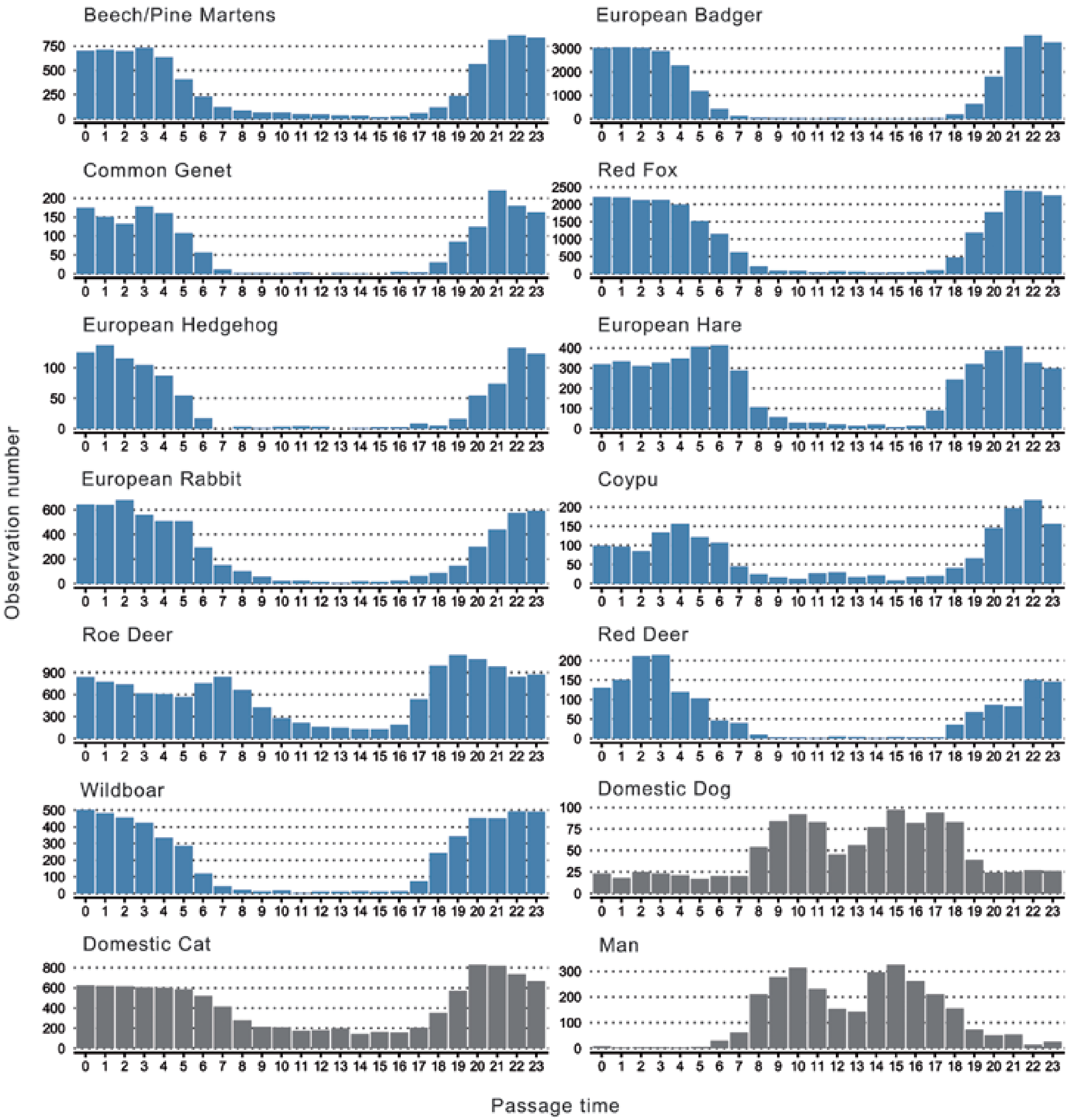


Figure 72 : Graphs of the hourly phenology of the 11 most detected species in the structures, as well as cats, Domestic Dogs, and humans.

It should be noted that also, the probability of occurrence of hedgehogs increases significantly with the occurrence of cats (Figure 71). This effect, which remains low, could be explained by a repulsive effect of cats on potential hedgehog predators such as martens or weasels, whose occurrence decreases with the presence of cats. More likely, these 2 species would simply co-occur in structures located near dwellings.

The occurrence of the domestic dog was only noted as negative for the occurrence of Coypu (Figure 71), and the occurrence of humans only for Red Fox (Figure 71). The asynchrony in the use of the crossings could explain the absence or the little interaction of humans and dogs on the rest of the fauna. Humans and dogs use the wildlife crossings almost exclusively during the day Figure 72), which would probably have less impact on the fauna that uses it mainly at night and at twilight.

6.2 IS SEASONALITY OBSERVED IN THE CROSSINGS?

In fact, the weekly occurrence of fauna in the crossings varies in function of the season and the species. As an examples, some results from the table in **Appendix 3** are interpreted according to the known ecology of the species.

The weekly occurrence of European Badger is higher in spring, then in summer and autumn, and is significantly lower in winter compared to other seasons. This can be explained by the fact that this species is more mobile from spring to summer compared to winter (home range 5 times smaller in winter).

The weekly occurrence of Red Fox is higher in winter, then in spring, periods corresponding respectively to the mating season (active search for partners) and to the rearing of the young (intensive search for prey) causing individuals to move accordingly.

The weekly occurrence of the European Hedgehog is significantly lower in winter than in other seasons. This can be explained simply by the hibernation of the hedgehog during the cold winter season.

The weekly occurrence (Appendix 3) of Red Deer, is significantly higher notably in autumn, during the rut when animals from different groups disperse to find individuals from other groups.

The weekly occurrence (Appendix 3) of Wild Boar, is significantly higher in winter than in other seasons. The greater movements explained by the rut, which occurs mainly in November/ December, and the dispersal of young leaving family groups. The greater hunting pressure at this time could also encourage the animals to move more.

Overall, the seasonality observed can be explained by many factors related to the ecology of the species and the different types of movements described in Chapter 4, page 56. Other local specificities such as the spatio-temporal variability of food resources can locally influence the movement patterns of the fauna.

6.3 DOES THE PRESENCE OF A CAMERA TRAP HAVE AN IMPACT ON THE ANIMALS USING THE STRUCTURE?

The vast majority of data/observations of individuals appearing to turn around in front of a camera trap concern the Red Fox (Figure 73). In the study by Fagart *et al* (2016) where refusals were systematically noted, 77% of observed refusals concerned the Fox. Other species such as the European Badger (15% of refusals observed), or Martes (2% of refusals observed) may display such behaviour, but in much lower proportions. These turn arounds due to a distrustful behaviour seem however to be temporary, with a decrease over time of the rate of refusals observed in the structures(Fagart *et al.* 2016).

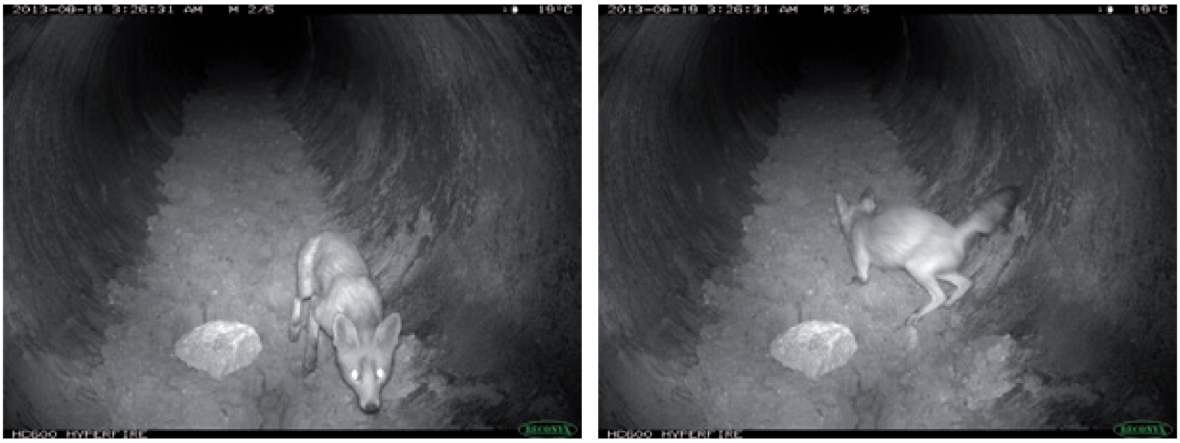


Figure 73: Red Fox surprised by the triggering of the camera trap and immediately turning around in the eco-duct ©ASF/LPO France. ©ASF/LPO France.

7.1. OPTIMISED MONITORING BY CAMERA TRAPS

7.1.1. SETTING CAMERA TRAPS: DETAILED RECOMMENDATIONS

Rex 1, published in 2016, provided instructions for camera traps. This new study encourages us to give more precise recommendations in order to guide the operators towards an optimal and standardised installation of the camera traps for monitoring the use of the structures by wildlife (Table 8).

According to Wearn & Glover-Kapfer (2017), the manufacturers of camera traps often recommend camera heights for large fauna (1.5 metres for example in the *Reconyx Hyperfire* manual). However, in order to detect a broader range, notably small and medium-sized fauna, it is important to place the cameras lower. Various authors (for example O'Brien *et al.*, 2003; Tobler *et al.*, 2008; Kays *et al.*, 2011; Wearn *et al.*, 2016) recommend heights between 20 and 50 centimetres. To target only small animals (for example small mammals and birds), it may be recommended to position the sensor even lower, at 10-20 centimetres from the ground (Thornton *et al.* 2012).

	Interdependence parameters	**Micromammals** (experimental, See §7.1.2.)	**Small and medium-sized fauna or multi-species**[1]	**Large fauna**
Target species	-	Voles, shrews, dormice*, etc.	Small carnivores, lagomorphs, hedgehogs, weasel, etc.	Ungulates, large carnivores, etc.
Detection distance [min-max] (beyond that, provide additional devices)	Parameter fixing the monitoring	~ 0,7 m [0.4-1 m]	~ 2 m [1-3 m]	~ 3 m [2-7 m]
Height of mounting [min-max] (in relation to path)	The shorter the detection distance, the closer the mounting height must be to the minimum height	[5-20 cm] Be careful with minimum focusing distance	[30-70 cm]	[100-130 cm]
Orientation [min-max] (angle in relation to path axis)	The larger the width of the crossing (adapted structure), the closer the chosen orientation can be to the maximum.	+ [20-45°]	+ [20-45°]	+ [20-90°] > 45° pour D > 4m

[1] **Multi-species monitoring**: for multi-species monitoring, it is essential to place the camera to photograph the most difficult-to-detect species, therefore the smallest species targeted (except micromammals). In a set-up targeting small and medium-sized fauna, if the large fauna is detected in the immediate vicinity of the camera, only the lower part or the legs of the animals will be visible, but this does not generally pose any particular difficulties for identifying these species.

Table 8: Recommendations for setting infrared mode camera traps for models offering detection performance comparable to recent models (Ex. Reconyx HP2X), depending on species targeted in the structures.

7.1.2. SOME EXPERIMENTS TO BE CARRIED OUT

Micromammals, an experimental case:

In 2016, in *Rex 1*, camera trap monitoring of micromammals was not addressed. Today, the progress made in terms of detection sensitivity and image definition enable, under certain conditions and for certain species, the development of such monitoring to be consideed. One of the conditions to consider it is that the path should be sufficiently narrow so that the detection field of a close-up camera trap (< 1 metre) can cover the entire width of the path (Figure 74). It is also necessary to use cameras with very short focal lengths (close to 50 centimetres) to limit the blurred images and enable the identification of target species. It should be noted that accessory or fixed short-focal-length lenses enabling closer focusing distances (Figure 75) are very recent technological developments for the next evolution in camera traps (Glover-Kapfer *et al.* 2019; Ortmann & Johnson 2020).

The identification of micromammals by camera trap is often not possible. In function of the objectives sought, groups by size or groups of species may however suffice to respond to the requested assessment.

It should be noted that in a counting approach, the failure rates of the equipment used must be calibrated. This requires having another reliable means of detection (video, cell) enabling to evaluate the failure rate of the camera trap.

If species approach is required, additional monitoring methods such as hair traps or faeces collection combined with genetic identification can be deployed (see §9.2.)

In all cases, and particularly for small target species, it is recommended to estimate the detection rate in function of the distance before deploying camera traps in the field (Hofmeester *et al.*, 2017). These detection rates of camera traps are known to vary depending on the location and orientation of the camera, the triggering and detection modes, the camera settings, the temperature differentials, the target species, and their sizes and behaviours (Meek *et al.* 2015; Apps & McNutt 2018).

Detection failures of passive infrared sensors can be reduced by adding active systems such as a photocell (Figure 76) (Meek & Pittet, 2012), or a vibrating cloth (*Rex 1,* p. 122). It should be noted that these two processes create additional constraints: less autonomy (to be calculated in function of the consumption in ampere/hours of the cell, usually 12 to 25 days of autonomy with a 9 Ah battery), additional battery, system to be sealed if necessary. The cost of these systems remains reasonable (around 1000 to 1200 € excluding VAT), especially since nowadays some new models of camera traps offer an external port for connecting these detection systems directly.

Reptiles et Amphibians

For reptiles and amphibians, infrared motion detection is totally dependent on the ambient temperature conditions, which are therefore not controlled. It is essential to set up a dedicated monitoring system: vibrating cloth, Time-Lapse* (Figure 77), infrared barrier, etc.

The modification of the thermal background of the active detection area of the camera trap with a material that "stays cold" (Welbourne, 2013) may maximise temperature differences and thus to improve detection. It should however be noted that in underpasses, often sheltered from the sun, this optimisation is already more or less naturally present at French latitudes.

Figure 74: Camera trap for monitoring a hollowed out micromammal pathway. ©ASF/GREGE.

Figure 75 : A Bank Vole photographed using a camera trap fitted with a close-up focus lens. ©Jean Chevallier.

Figure 76 : Infrared barrier detection system triggering a camera trap on the corbel of a motorway hydraulic structure. ©LIFE VISON/LPO France.

Figure 77: Fire Salamander, photographed in Time-Lapse mode. ©ASF/LPO Drôme.

7.1.3. TOWARDS A STANDARDISATION OF SEMI-AUTOMATIC DATA ENTRY AND AN OPTIMAL MANAGEMENT OF THE COLLECTED DATA

Good management of camera trap monitoring data is important to avoid loss of resolution and achieve optimal data banking. Scotson *et al.* (2017) list nine recommendations on the best practices for managing all camera trap monitoring data (study site metadata, camera trap deployment metadata, image classification data and derived products:

1. Adopt a standardised, non-exclusive and transferable data storage format to store all camera trap data;
2. Accompany all entry sheets with formatted metadata;
3. Record data at the highest possible resolution;
4. Use a clearly documented and consistent geographic coordinate system;
5. Maintain consistent date-time format;
6. Record covariate data that could be used to assess the probability of detection;
7. Plan for the eventual identification of all non-target species and human data;
8. Manage the data as an authoritative group, on which several users can act consistently and simultaneously;
9. Archive data and make it available to other researchers under defined reuse conditions.

During the consolidation of the Rex 2 camera trap database, several problems in the way crossing data were entered were identified: loss of data (omission of certain crossings), data entry errors (duplication, error in the date/time), ambiguous date and time format, poorly informed observation pressure, incomplete metadata and ambiguous or even unintelligible data entry/formatting, inconsistent treatment according to the operator of the independence of successive crossings of individuals of the same species, etc. These errors greatly complicate the creation and consolidation of the database. Moreover, the photos, while they are banked by VINCI Autoroutes, are not directly linked to the crossing data, complicating the verification of the data or identifications.

Image analysis remains a time-consuming task in camera trap monitoring. In connection with the repetitive nature of this step, input errors are common.

In order to optimise this data entry, there are software programs to assist in the processing of camera trap data, enabling the direct use of the EXIF data of the photos. They enable an automatic retrieval of many parameters (e.g., date, time, temperature, phase of the moon, camera trap settings data), and to help with dedicated interfaces to the processing and banking of images and identification and crossing data.

Comparative studies of recent software for data and metadata management of wildlife monitoring by camera traps have been conducted by Young *et al.* (2018), Scotson *et al.* (2017), Wearn & Glover-Kapfer (2017) and also the WildCam Canada group in 2020. After having tested and compared eight software programmes that do not require programming knowledge, for use by the largest number of people, three software programmes (that can be installed locally) appear to be of interest for the wildlife monitoring carried out on wildlife crossings: **Wild Id[1], Camelot[2] and Timelapse[3]**. None of them were implemented during the monitoring of this Rex.

Timelapse software (Greenberg *et al.*, 2019) has the advantage, among others, of being in a constant state of development, of enabling the free addition of formatted fields (list of species, ticks, list of choices, etc.), of proposing setting up parameters of the independence of successive crossings and of presenting a user-friendly

1 https://www.wildid.app/
2 https://camelotproject.org/about-us
3 https://saul.cpsc.ucalgary.ca/timelapse/

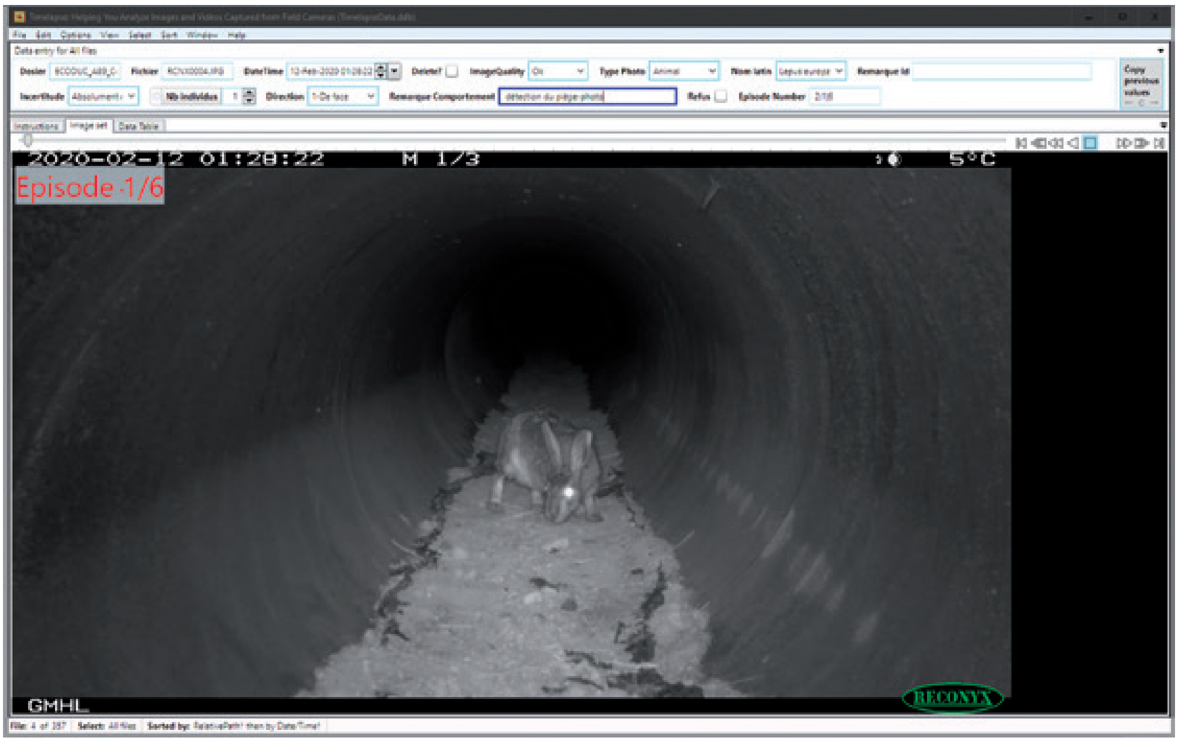


Figure 78 : Interface of software for Time-Lapse camera trap data processing.

access. On the other hand, this software does not manage the monitoring metadata and use of camera traps. It is a purely data entry and standardisation software, standardised metadata on the use of camera traps and monitoring must be entered in parallel.
The processed data then enables the images to be linked directly to the passage data in the images database, enabling a faster verification of the determinations and passage data. This should also facilitate the development of future software for automatic image detection and classification.

7.2. SIZING OF CAPTURE-MARK-RECAPTURE (CMR) PROTOCOLS FOR MONITORING ADAPTATIONS FOR SMALL FAUNA

7.2.1. ISSUE

It was seen above that various protocols have been considered for small fauna in certain configurations: time-lapse camera traps in underpasses, for example. Other configurations such as overpasses or eco-bridges do not always allow the implementation of these protocols. Other methods could conceivably measure the flow of small fauna through the crossings. One of these methods, capture-mark-recapture (CMR), involves marking or tagging captured individuals in order to highlight possible movements by recapturing (or detecting) the tagged individuals. The results obtained enable the estimation of demographic rates (such as the survival rate) and transition rates (such as emigration or recruitment). Usually, CMR protocols for small fauna are carried out by scheduling field visits to capture and mark the individuals. This type of protocol is nevertheless very dependent on the number of individuals, the probability of detection and the number of visits. In order to evaluate the parameters necessary for such a CMR protocol, a statistical simulation was performed.
The objective of using simulated data is to choose a sampling strategy that results in a sufficiently reliable statistical estimate. In the case of assessing the use of crossings by small fauna with a CMR protocol, this approach will make it possible to evaluate how many capture sessions are required to correctly assess the transition rate, i.e., the "crossing events" of the structure studied.
The elements describing the development of the calculation model are presented in **Appendix 4**.

7.2.2. RECOMMENDATIONS FOR THE DEVELOPMENT OF MONITORING OF CROSSINGS BY IDENTIFICATION OF INDIVIDUALS (CMR, RFID, CAMERA TRAPS)

The Figure 79 shows the variation in the number of times the CMR model fails to detect a transition when it should, under the different scenarios considered. The results of the simulations show that in order to detect a transition rate of around 1%, the CMR method seems appropriate only for populations of more than 90 individuals with at least 6 visits per year.
The results of the monitoring of amphibians, reptiles and micromammals carried out on six eco-bridges over three years show that the transition rates (crossing rates) are low, with no transitions observed for amphibians and reptiles, and only two transitions for micromammals (a transition being considered as such when an individual completely crosses the adapted eco-bridge). These observations indicate that the 1% transition per year scenario appears realistic. The results show that using the CMR technique to

detect such a transition rate does not seem to be suitable for most species.

Indeed, for amphibians and reptiles, the probabilities of capture and recapture are generally low, and the effort needed to detect such a transition rate would require more than 10 crossings per year, sometimes even more than 20 crossings in the case of small populations (Figure 79). The results show that to detect a transition rate of 2%, the CMR effort to be deployed would be similar.

For micromammals, whose numbers seem to be higher according to the feedback, CMR could be appropriate if the capture effort involved at least 5 sessions per year. Nevertheless, this possibility seems difficult to implement in view of the effort required for capture sessions with traps.

In the case of higher transition rates, from 5% transitions onwards, CMR would be appropriate for populations of 60 or more with 4 visits per year (Figure 79).

Thus, before adopting a CMR protocol to evaluate crossing events for small fauna on a crossing structure, it is recommended to have minimal indications concerning population density and the number of individuals potentially accessible for tagging, in function of the target species. This will assess the protocol required (number of traps, number of crossings, etc.) to obtain reliable results on the crossings of the structure.

Other protocols could conceivably be used to evaluate the use of a crossing by small fauna. For example, tagging with passive transponders (PIT-tags), and setting up an antenna on the ground that enables the recording of each individual that crosses it. This technique requires the capture and implantation of transponders, which is carried out once at the beginning of the survey[4], and then the installation of monitoring equipment in the field (electrically powered antenna). This nevertheless imposes a

4 This intervention requires authorisations: capture and marking of animals for scientific purposes, approval from the ethics committee and the ministry in charge of research.

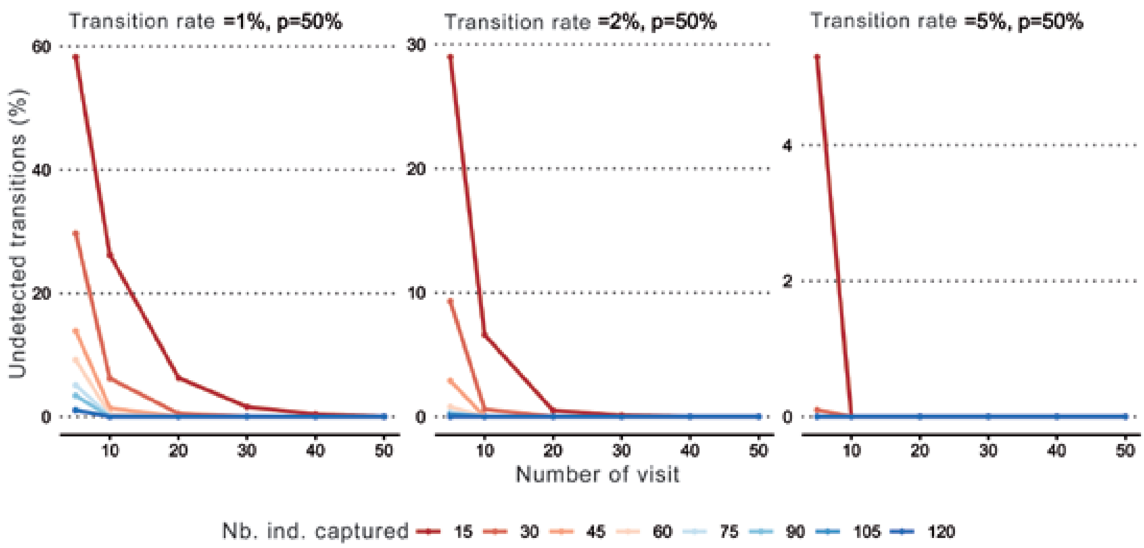


Figure 79: Variation in the number of non-detections of transitions in function of scenarios of number of visits and population sizes. The results presented in this figure correspond to a detection probability (p) of 50%. The results for the other values of p are not presented, as they are similar.

significant rate of population tagging, which is often difficult to obtain in function of the group. In some cases, the setting up of camera traps is envisaged for monitoring amphibians in narrow underpasses but has the disadvantage of having technical limitations (width of passages, only individually recognised species, such as the Fire Salamander, can be studied).

7.3. QUESTIONS/ANSWERS RELATED TO PROTOCOLS

7.3.1 DO CAMERAS UNDERESTIMATE THE NUMBER OF CROSSINGS IN THE STRUCTURE?

The study by Jumeau *et al* (2017) shows that a HC600 camera trap, compared to continuous video recording in underpasses, misses 17% of medium-sized mammals crossings and 43.6% of micromammal crossings (mice, voles, shrews).
As mentioned in paragraph 7.1.1, the continuous development of camera traps is improving their detection capabilities.
In the present feedback, one operator changed camera trap models during the monitoring of 14 structures (13 in the South-West and one in Auvergne), about halfway through the monitoring, from the Reconyx HC600 to the Reconyx HP2X, which was considered more sensitive by users. The quantity of data collected with the more recent model (HP2X) is significantly higher (Wilcoxon V = 14, p = 0.01; Figure 80).
Although confusing effects (meteorology, phenology and/or species habituation) could explain this difference, it would appear that the use of an HP2X model is more effective for detecting certain crossings by fauna.
Furthermore, comparative tests (LIFE Vison, 2022) aimed at evaluating the detection rate of small and medium-sized fauna were recently carried out between a latest generation infrared camera trap (Reconyx HP2X), and the same model triggered by an infrared barrier, a reference device with a theoretical detection rate of 100%. The results show a detection rate of 99.2% for the HP2X infrared camera trap alone for 4 medium-sized species and small wildlife (genet, beech marten, squirrel and weasel) with 1 false negative (shot without animal) out of 127 crossings. The number of false negatives then logically increased with the reduction in the size of the animals, notably micromammals which were not targeted during this test (85% detection rate for the Brown Rat, for example).

Interpretation of eco-bridge crossings by small fauna s: *Carabidae* beetles, potentially a good indicator.

In order to ensure that small fauna crosses the eco-bridge, it is necessary to collect geo-referenced observations on both sides of the eco-bridge and to set up a monitoring system that makes it possible to recognise individuals. The interpretation of the crossings of small mammals, due to their ecology (forest and woodland edge species) and their small home ranges, is sometimes complicated.
Carabidae beetles are non-flying, highly mobile terrestrial insects that move mostly at night and can cover fairly long distances in their home range in search of prey or mates. They therefore seem to be good models for measuring the effectiveness of eco-bridges as corridors for small wildlife.
The CEFE therefore proposes a protocol for monitoring *Carabidae* beetles on eco-bridges (Appendix 5).

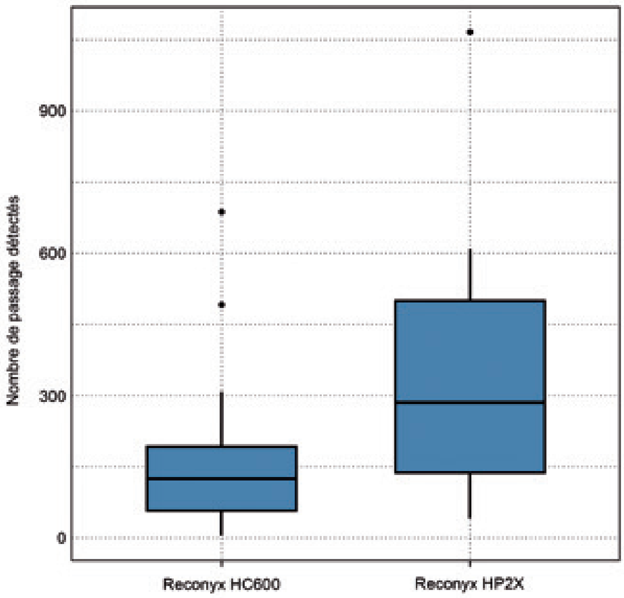


Figure 80: Comparison of the evolution of data collected during identical monitoring (n = 14 structures), following the change from Reconyx HC600 to Reconyx HP2X devices.

7.3.2 IMPACT OF USING MARKING RELIEF IN FRONT OF THE CAMERA TRAPS

Placing a stone/branch in front of the camera trap is a technique indirectly used by naturalists/photographers who are looking for marking points of animals to place their camera/shooting equipment. The objective here is to create relief (topography) (as natural as possible) in front of the camera trap (Figure 81), to encourage the animals to mark their territory while crossing. The animals sniffing or marking the relief stay a little longer in front of the camera trap and are thus more likely to be detected and identified.

This hypothesis has never been verified by comparing monitoring with and without "marking relief", but it does not appear to be a possible source of reduced detection or avoidance of crossing by wildlife.

7.3.3 HOW TO DEAL WITH THE INDEPENDENCE OF SUCCESSIVE CROSSINGS?

It is impossible to clearly answer this question since there is no way of individualising with camera traps (except in rare cases of recognisable individuals). On the other hand, there is a real and necessary requirement to standardise the independence of crossings for the different data entry operators. VINCI Autoroutes' latest recommendation on this question was to enter all

Figure 81 : Marking relief (stone) widely used by wildlife, positioned on the floating pontoon of a hydraulic road structure. ©LPO France.

the data independently when U-turn behaviour was not clearly observed. Thus, post-entry processing to manage these successive and/or repeated crossings in a global way could help to standardise the dataset.
Example: a fox observed in one direction and then a fox observed in the other direction 40 seconds later, without a U-turn observed on the photos, must be considered and entered as two different lines of data.

7.3.4 WHAT PERIOD, DURATION AND FREQUENCY FOR MONITORING ADAPTED WILDLIFE CROSSINGS?

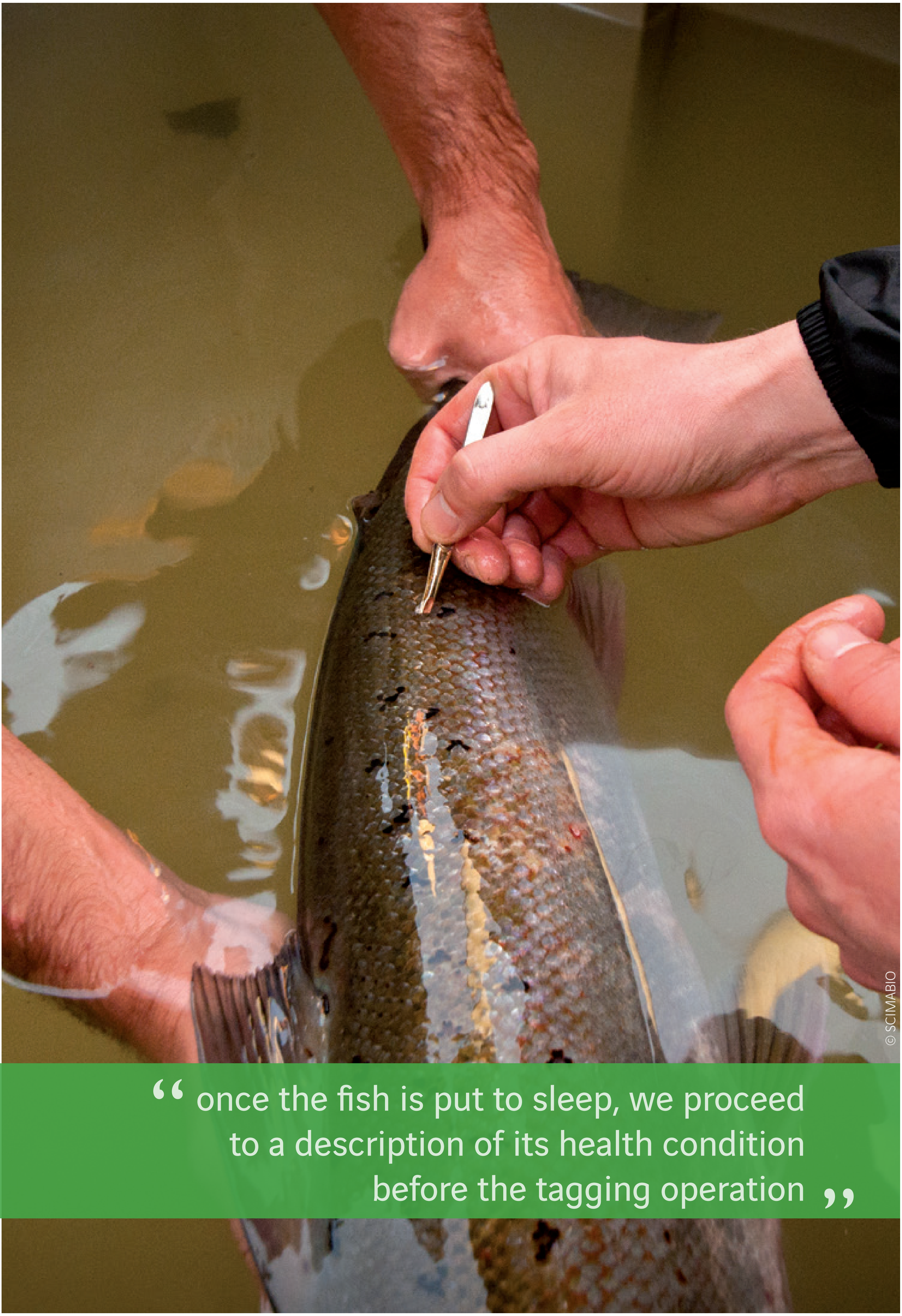
The **period of monitoring** depends primarily on the objective of the monitoring and the recovery objective that the structure meets (Van der Grift & Van der Ree, 2015). If the objective is, for example, to allow the spring migration of amphibians to their egg-laying sites, then the monitoring will focus on this period. Similarly, if the target species is absent at a given time (migratory or hibernating species), the monitoring should focus on the period of presence of the species or the maximum period of known activity of the species. If the objective is on the other hand to estimate the annual frequentation of a structure by a range of species, such as medium-sized and large fauna, then continuous monitoring covering at least a whole year is recommended to follow the different movement phenologies (in the home range, dispersion or migration) of the species.

The **frequency of monitoring** should also be adapted in close relation to the monitoring period (Van der Grift & Van der Ree, 2015). If the monitoring is monthly and, for example, on the migration period of a species, then daily monitoring may be necessary. Conversely, if the objective is to monitor movements on an annual cycle, weekly monitoring may be sufficient. The monitoring technique also strongly influences the monitoring frequency. Camera traps enable 24-7 monitoring with a limited number of rotations, for example once every 2 months, whereas footprint traps require much more frequent visits, once a week or even every 2 to 3 days for continuous monitoring.

The **duration of monitoring** is known to strongly influence the estimation of wildlife crossings. Monitoring over several years is thus recommended in relation to the possible inter-annual variations in the use of the crossings, in particular when the population size of the target species is known to vary significantly from one year to the next (e.g., cyclical nature of the abundance of micromammal populations) or when temporal trends used are sought. The use of crossings is also known to increase over time after their construction, as individuals need time to discover and become accustomed to the crossings. The frequentation of crossings by wildlife after their installation then progresses during the monitoring. The monitoring of several eco-ducts initiated just after their installation in the South-west of France on the VINCI Autoroutes network showed that the use of the structures had increased eightfold during the first three years of monitoring (Fagart *et al.*, 2016). In Canada, where long-term monitoring (> 15 years) is carried out, the time taken for large wildlife to become accustomed to the use of structures has been shown to be between 3 and 9 years, depending on the species (Clevenger & Barrueto, 2014).

In COST 341, Iuell and his colleagues (Iuell *et al.* (2003) recommend monitoring the frequency of crossings for at least 3 years and not evaluating the effectiveness of crossings just after their construction.

Thus, the monitoring of structures carried out just after their construction needs to be renewed over time (e.g., 5 years, 10 years) to assess possible changes in fauna frequentation over the long term. The parameters influencing the attractiveness of crossings may also evolve over time (e.g., development of vegetation, land use surrounding the crossing).



“ once the fish is put to sleep, we proceed to a description of its health condition before the tagging operation ”

Monitoring of structures specific to fish, bats and micromammals were carried out by specialised environmental consultancies. They are covered by specific reports published in parallel with the present document. A summary of these monitoring operations is presented below.

8.1. FISH MONITORING

This monitoring font is covered by specific feedback published in the report: *Use of technology in the framework of assessing fish passage to provide the knowledge required for restoring ecological continuity in rivers,* SCIMABIO Interface, 2023.

8.1.1. MONITORING BY RADIOTELEMETRY – ASSESSMENT OF THE CROSSING OF ADAPTATIONS TO WEIR A89 ON THE RIVER ALLIER BY ATLANTIC SALMON (PUY-DE-DÔME,63)

The Atlantic Salmon is an emblematic species of aquatic biodiversity que in the Loire-Allier drainage basin. Ecological continuity issues are central to the threats facing the species, in particular due to the length pf the stretch of river that the genitors must swim up to reach the first high-quality spawning grounds. In this context and in order to respond to reglementary obligations, VINCI Autoroutes undertook works on its "A89" weir on the River Allier, which were completed in October 2017. Fish monitoring was then programmed in order to measure the gains for the spawning migration of the salmon in the Allier.

This fish monitoring was carried out in 2019 and consisted in trapping, then capturing the salmon in the Allier at the Vichy and dam and the intragastric implanting of a radio-emitter. In total, 24 salmon were tagged as from 26 March 2019.

The monitoring set up in the study area consisted above all of 7 fixed stations managed by SCIMABIO Interface and FDPPMA63, to which should be added the downstream fixed station fixe at Vichy set up and managed by LOGRAMI (Figure 82). Each fixed station consisted of a radio receiver/recorder programmed to scan the frequencies of the emitters implanted in the salmon. It should be noted that in addition to fixed monitoring systems, mobile surveys were conducted (by vehicle, on foot or by canoe) so as to monitor the movements of the fish between the areas covered by the fixed antennae.

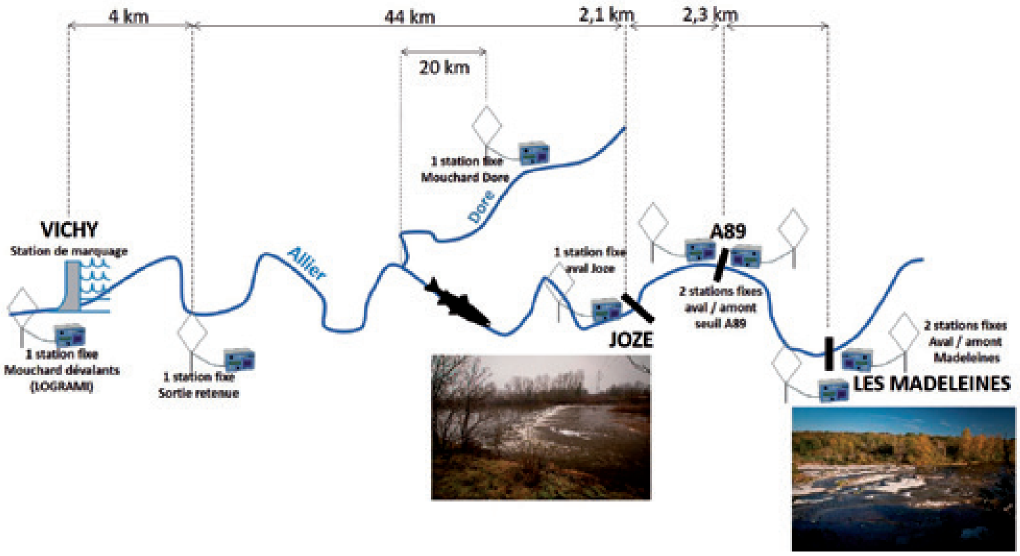


Figure 82: Diagram of the locations of the 7 fixed monitoring stations in the "VINCI Autoroutes" study area + the LOGRAMI downstream station at Vichy.

Analysis of migration behaviours in the study sector and the crossing of Weir A89 was carried out on the basis of the 13 individuals out of 24 tagged that continued their migration after tagging as far as Weir A89.

Among the principal results of this study, we can cite that the mean estimated crossing speeds for Weir A89 did not show any retarding effect, thus providing some clear elements of a response to the reglementary obligation. Moreover, no blockages were detected at the foot of the structure.

8.1.2. MONITORING BY RFID TECHNOLOGY - ASSESSMENT OF FISH PASSAGE THROUGH CULVERT OH 448 OF THE A89 MOTORWAY ON THE DUROLLE RIVER (PUY-DE-DÔME, 63)

The Durolle is a salmonid river in the Puy de Dôme department, situated at the head of a drainage basin, which is home to a functional and abundant trout population. Hydraulic structure OH 448 enables the A89 motorway to cross the Durolle near the village of St Rémy sur Durolle. It is an old structure, dating from the construction of the A72 in 1978, which used to be an uncrossable obstacle for trout due to the flow conditions inside the culvert. In 2016, this culvert was adapted with a unique hydraulic system in order to restore free fish circulation (Figure 83).

The real effectiveness of this system was assessed between 2017 and 2019 using RFID (Radio Frequency Identification) technology and monitoring the movements of the tagged trout by leans of transponders. In total, 815 trout were tagged (Figure 84) on a 1680-metre stretch downstream of the culvert. In parallel, a double-antenna fixed RFID device located immediately upstream of the culvert detected throughout the duration of the monitoring the tagged individuals that had passed through the culvert. In addition, 7 mobile surveys were conducted to characterise the movements of the tagged trout downstream of the culvert. The passage data were analysed with respect to the characteristics of individuals,

Figure 83: Presentation of the culvert before (left) and after works (centre and right). Source ASF.

Figure 84: Steps in the tagging of the trout monitored during the study, from capture to putting them back in the water.

together with the hydrology and the water temperature.
The results of the study showed that the adapted hydraulic system developed in 2016 provided good passability through the culvert for the trout of the Durolle, whatever the hydraulic or thermal conditions recorded during the study.

8.2. BAT MONITORING

This monitoring is covered by specific feedback published in the report: *Monitoring of experimental structures for bats, Naturalia-Environnement, 2023*.
In recent years, VINCI Autoroutes has been interested in the effectiveness of road crossing structures concerning the biological group of bats, whether or not the structures are specific to them.
Dual carriageways have a negative effect on bats' hunting and/or transit activity, over distances of up to more than 5 kilometres from the road (Claireau *et coll.*, 2019 a). To reduce this negative effect (called the "road-effect zone"), crossing structures are arranged on the motorway network which can be of various types: above the road (eco-bridges, bat bridges…), below the road (hydraulic structures, wildlife crossings…) or intermediate (green springboards/hop-overs …).
Since 2014, ASF has notably been interested in two types of structures: structures specific to bat crossings, referred to below as "bat-bridges" (Figure 85), and eco-bridges (a new generation of wildlife crossing). To this end, ASF commissioned the environmental consultancy Naturalia-Environnement, in collaboration with CESCO-MNHN, in order better to understand how these reduction measures can be effective for reducing the impact of the motorway network on bats.
In total, 5 structures were studied between 2014 and 2019: 3 bat bridges (two on the A89 and one on the A83) and 2 eco-bridges (one on the A64 and the other on the A89). Each site was monitored by means of a scientific process as close as possible to a Before-After-Control-Impact (BACI) experimental design. All these structures have been (or are being) covered

by publications in international peer-reviewed journals or conferences, whose references are listed in the box below.

To assess the effectiveness of these structures, two parameters were studied. The first was the capacity of the structures to enable species to cross safely (i.e., at a height of more than metres (Berthinussen & Altringham, 2012)). The second was to find out whether the structure was able to improve ecological connectivity. Two types of innovative, published monitoring were implemented. For the first parameter, visual monitoring using a thermal imaging camera was carried out and, for the second, acoustic monitoring. These methods enabled the trajectography of the bats' flightpaths to be plotted (Claireau *et coll.*, 2021, 2019 b).

This new feedback on experience, by monitoring these structures, demonstrated that, provided the specific structure is correctly placed in an ecological corridor, it is used by bats. This utilisation is greater when the ecological the corridor is narrow (e.g., a hedge); conversely, in the configuration where a structure is placed in a broader corridor (e.g., a wood), it will be less frequented.

Moreover, the capacity of these structures to raise the bats' flight altitude seems to be more favourable when the road is embanked, and tall trees are planted either side of the structure.

Figure 85: Bat-bridge (cradle structure) at Moulin-Paris on the A89.

8.3. MICROMAMMAL MONITORING

This monitoring is covered by specific feedback published in the report: *Evaluation of frequentation by micromammals of a footway under a hydraulic structure fitted with a hollow section*, GREGE, 2023.

8.3.1. MONITORING OF THE UTILISATION OF A HOLLOWED-OUT SECTION FOR MICROMAMMALS INTEGRATED INTO A FOOTWAY ON THE A89.

In the framework of the adaptations carried out by ASF in the Deiro structure (A89, commune of Soudeilles en Corrèze) to re-establish ecological continuities for fauna, a specific 120-centimetre-wide footway was created in the structure. This adaptation subsequently led to the creation of stone spurs running along the footway in the riverbed. On this occasion, ASF wanted to test the setting up of a hollowed-out section for micromammals developed by the GREGE originally for water shrews and water voles. This thirteen-centimetre-wide and ten-centimetre-high “notch” was hollowed in the wall of the "small wildlife" footway to create a covered pathway for micromammals.
In order to assess the efficacy of this innovative arrangement, the GREGE joined forces with GMHL to evaluate the frequentation of the hollowed-out section by micromammals, using experimental protocols appropriate for censusing and identifying micromammals. Four techniques were combined to census the passages and identify the species: footprint trackers, camera traps, sample collection tubes (fur and faeces), and the gathering of environmental DNA in the hollow section with the genetic identification of the species detected.

Out of the 935 nights when the camera traps were operational, 1330 crossings by five species or species-groups were recorded in the (923 crossings by the group of two wood mice in the genus *Apodemus*; 361 crossings by the group of two water shrews in the genus *Neomys*; 8 crossings by Red Squirrels *Sciurus vulgaris*; 5 crossings by the group of small voles and 1 crossing by the group of large voles in the genus *Arvicola*). Therefore, over the two years of monitoring, the circulation of micromammals in the hollow section was close to 36 crossings per month. These results are globally higher than in the very rare bibliographical references and confirm high frequentation by micromammals of the adapted structure. This monitoring demonstrates the utilisation of the hollow section for micromammal crossings.
The numerous crossings attributed to the genus *Neomy*, with a notable peak between July and October, are particularly remarkable (no other reference on the subject to our knowledge whether in terms of frequentation or even monitoring) and show the great interest of this adapted structure for this group of protected species. Moreover, the protocol specifically developed to collect DNA in the hollow section or samples in the dedicated collection tubes, and the genetic identification of species, confirmed the circulation de la European Water Shrew (*Neomys fodiens*) and the Wood Mouse (*Apodemus sylvaticus*).

Figure 86: General view of the Deiro structure and its re-adaptation for small fauna (photos taken in the worksite phase). ©GREGE.

Figure 87 : Neomys sp. *Circulating in the hollow section for micromammals. ©GREGE/GMHL/ASF.*

IX. PERSPECTIVES: TOWARDS NEW OBJECTIVES AND APPROPRIATE MONITORING

9.1. THE VARIOUS MONITORING ISSUES

As previously seen, the vast majority of the monitoring carried out in this feedback concerns monitoring by camera traps, making it possible to evaluate the species inventory and the frequency with which they cross the wildlife crossings, i.e., the use of the structures by wildlife and not really their effectiveness in relation to clearly specified objectives.

While the regulations only concern monitoring methods (L 122-1-1 I, R 122-5 II of the Environment Code) or monitoring devices (R 122-13 II of the Environment Code), the doctrine and national guidelines on "Avoiding, reducing and compensating for impacts on the natural environment" concern the effectiveness of measures and result indicators.

It should be noted that that these monitoring methods may be imposed as part of project authorisations (e.g., prefectural decrees on protected species)

This monitoring obligation is covered in the latest CEREMA guide published in 2021: *Wildlife crossings. Preserving and restoring ecological continuity with linear transportation infrastructures.*

File 23 (pages 260 to 265) of the "How to monitor wildlife crossings" guide states that:

"*Firstly, the objectives of the measure must be specified. For example, for a given target species:*

- *the crossing must allow daily movements between resting and feeding habitats;*
- *the crossing must allow seasonal movements between resting and breeding sites;*
- *the crossing must allow occasional movements so that genetic mixing between sub-populations living in metapopulations can occur;*
- *more generally at project scale, the overall permeability of the infrastructure must allow for population maintenance".*

Monitoring the effectiveness of a structure therefore requires:

- carrying out an initial assessment before adapting the structure in order to propose an adaptation appropriate to the issues and relevant analysis before/after adaptation;
- specifying the objectives of the adapted structure, proportionate to the issues at stake;
- implementing a monitoring protocol that is appropriate to the adapted structure and the set objectives (techniques, time periods, frequency, survey durations).

Nevertheless, as specified in File 23 on page 162 of the CEREMA guide, assessing the effectiveness of a crossing for maintaining populations falls into the field of scientific research, requiring a much more advanced level of investigation with notably the implementation of a BDACI* (Before-During-After-Control-Impact) study.

However, it should be noted that this method has its limitations:

- In the case of existing motorways, it will only be possible to assess the initial status with the structure already set up before adaptation.
- In the case of new projects, it appears complex to assess the free movement of wildlife prior to the creation of the infrastructure, and then compare it to crossings concentrated at a single point on the adapted structure.

Furthermore, for both of these cases, the evaluation of population numbers present before, during and over several years after the construction of a structure and on a control site (ideal BDACI: Before-During-After-Control-Intervention protocol) would make it possible to robustly evaluate the use and effectiveness of the structure for the populations of one or more animal species. However, this requires complex protocols and monitoring which often prove difficult to carry out.

Here we see the difficulty in meeting certain

objectives, notably those concerned with population dynamics.
Three frequently raised questions are addressed below:

Effectiveness of an adapted structure setup: "what crosses and what does not cross":

This issue aims to locally compare "what crosses" to "what does not cross" or "what should cross" the structure.
In the framework of monitoring motorway structures, this type of monitoring can for example be established on the model of B(D) ACI (Before-(During)-After-Control-Impact) protocols, which aim to compare flows Before, (During) and After adaptation, or even with a Control area that is identical to the Impacted area (Andis *et coll.*, 2017) thus eliminating the location effect. Such protocols were fully implemented during bat bridge monitoring by Naturalia Environnement (see Chapter 8.2)
This question can also be dealt with more simply, for example by comparing the fauna detected inside the structures to the fauna detected at structure entrances using camera traps. However, this simple methodology, which has already been implemented in the framework of monitoring VINCI Autoroutes, does involve numerous technical constraints as described in §4.3.3. on page 52. Solutions to these constraints should be found to propose a protocol adapted to the reliable collection of such data.
Concerning the monitoring of frequentation inside the structures, it is sometimes possible to note the refusal of certain individuals to cross structures (turn back). This has made it possible to address the subject of the familiarisation of animals to a structure over time (Fagart *et al.*, 2016), but without taking into account the proportion of animals that do not enter a structure at all. The monitoring of crossing refusals remains experimental and within the field of research-innovation.

Movement of individuals in relation to infrastructure:

The study of the movement of individuals in relation to infrastructure seeks to determine the extent to which roads inhibit or change wildlife movements and the extent to which wildlife crossings can reduce these effects (Soanes *et coll.*, 2018).
The methods used to monitor the movements of individuals are mainly CMR (either by direct trapping or using hair or faeces traps with the genotyping of individuals), passive monitoring using passive transponders (PIT-tag), telemetric monitoring (radio/GPS) or camera traps coupled with automatic recognition of individuals by Artificial Intelligence for species with visible, characteristic phenotypes. The latter making it possible to avoid direct trapping for the fitting of a transponder (e.g., the Newtrap method developed by the Luxembourg Institute of Science and Technology for the monitoring of newts).
In the case of new construction projects, this identification of individuals can make it possible to observe changes in their movements in relation to the development of a new infrastructure and associated wildlife crossings. In the case of existing infrastructures, the monitoring of individuals allows for the study of their behaviour and movements with respect to a wildlife crossing. In both cases, a Before/After design allows for the comparison of movements and the quantification of impacts/benefits.
The monitoring of fish using RFID transponders and radio telemetry carried out by Scimabio on the A89 is an example of the monitoring of individuals to evaluate the crossing of adapted hydraulic structures (see Chapter 8.1).

The use of genetics for motorway infrastructure impact assessment studies:

Genetics can help answer some important questions, including the impact of roads on populations through various material collection techniques: sample gathering, fur or faeces tubes, trapping, environmental DNA, etc. Depending on the specific developments and the quality of the collected samples, the sample can theoretically identify, amongst other aspects (O'Brien, 2018):

- the origin of individuals arriving at a wildlife crossing;
- the characteristics (e.g., sex) of individuals;
- how many individuals use the same wildlife crossing;
- the frequency of crossing use by specific individuals;
- the paths of individuals moving through the landscape;
- the level of exchange of individuals between populations;
- a measure of whether dispersal results in gene flow;
- the species killed on roads, etc.

Genetic sampling can be time-consuming with high analysis costs depending on the study. These protocols may therefore need to be developed in the initial stages by conducting pre-project baseline sampling within the study population potentially impacted by a project. Repeating the study several years after the implementation of the project and its crossings should make it possible by comparison to assess the absence of genetic drift or population partitioning caused. For example, GREGE *et al.* (2012) showed the genetic partitioning of Red Deer populations in the Landes region caused by the A63 motorway between Bayonne and Bordeaux.
The number of study/research programmes related to the conservation/ecology of species using these DNA sampling techniques is increasing. The possibilities of incorporating questions on road ecology into existing programmes are undeniable, given the vast range of possibilities for studying via DNA sampling.

9.2. WHICH METHODOLOGY FOR WHICH OBJECTIVES?

A multitude of monitoring measures can be implemented depending on the project framework (upgrading of existing infrastructure *versus* new construction project), the objective pursued, the type of structure or the target species. In order to set up a monitoring system that enables the desired results to be achieved, several steps are required to identify the precise performance evaluation plan for an impact mitigation measure (Obrien *et al.* 2018):

- Identification of the target species and mitigation objectives;
- Selection of the most relevant indicators;
- Selection of the monitoring method;
- Selection of spatial and temporal scales for data sampling;
- Assessment of the duration and frequency of sampling required for the monitoring;
- Selection of explanatory variables.

A case-by-case analysis is therefore required. Table 9, taken from the study by O'Brien (2018), lists the different methods and their relevance for different taxonomic groups. Three columns have been added in order to specify whether monitoring can identify individual animals or not, and to roughly assess whether a technique requires a rather high or low investment in time and equipment.
The development of new experimental protocols should undergo bias assessment and a measured comparison of the advantages/disadvantages of different techniques in order to best guide protocol choice. For example, in

Target groups / Methods	Large mammals (deer, large carnivores)	Medium-sized mammals (carnivores, mustelids, lagomorphs, large rodents)	Small Mammals (Micromammals)	Bats	Walking birds	Flying birds
Footprint trap (coarse sand)	++	++	0/-[1]	-	++	-
Footprint trap (fine sand)	++	++	-	-	++	-
Footprint sensor	-	++	-	-	-	-
Footprint in snow	+	+	-	-	-	-
Photo/video capture	++	++	+/-[2]	-	++	?
Infrared detector	0	0	0	0	0	-
Artificial shelters (panels, etc.)	-	-	+	-	-	-
Bat detector	-	-	-	++	-	-
Direct observations (visual or acoustic)	-	-	-	+	-	++
Indirect observations (faeces, droppings, fur)	+	+	+	-	+	+
Fur trap - identification	+	+	+	-	-	-
Fur trap - DNA analysis	+	+	+	-	-	-
Capture Marking Recapture	-	+/-[2]	++	-	-	-
Capture Marking passive Monitoring (PIT transponder)	+	+	+	-	+	-
Capture + telemetric Monitoring (radio/GPS/ satellite)	+	+	-	+	+	-
Capture/release (trap, net)	-	-	++	++	+	-
Lethal capture (traps)	-	-	-	-	-	-

Table 9: The different wildlife monitoring techniques for wildlife crossings and their suitability for the target wildlife groups, according to O'Brien et al. (2018), table supplemented with details on the sizes of mammals and some details on investment costs (last 3 columns). Legend: ++ Highly appropriate; + Appropriate; 0 Identification of taxonomic groups, but not of the species; - Inappropriate; ? Unknown.

Reptiles	Amphibians	Non-flying insects	Flying insects	Other invertebrates	Able to identify individual animals	Investment: field time/ reading frequency	Investment: equipment/cost of processing/ analysis
0/-[1]	-	-	-	-	No	High	Low
-	-	-	-	-	No	High	Low
-	-	-	-	-	No	High	Low
-	-	-	-	-	No	High	Low
?	+/-[3]	-	-	-	No[2]	Low	Intermediate
-	-	-	-	-	No	Intermediate	Intermediate
++	++	+	-	+	Yes/No	High	Low
-	-	++	++	-	No	Intermediate	Intermediate
++	++	++	++	-	Yes/No	High	Low
-	-	+	+	-	No	High	Low
-	-	-	-	-	No	Intermediate	Low
-	-	-	-	-	Yes	Intermediate	High
++	++	++	++	+	Yes	High	Intermediate
+	+	+/-[2]	+/-[2]	-	Yes	High	High
+/-[2]	+/-[2]	-	-	-	Yes	High	High
+	++	++	++	++	Yes	High	Intermediate
-	-	++	+	+	Yes/No	High	Intermediate

[1] Registration, but not at species level and only for certain species in the group.
[2] Suitable only for certain species in the group.
[3] Suitable for use in small structures.

a study comparing the effectiveness of a camera trap and continuous video recording, Jumeau *et al*. (2017) found that continuous video recording was more effective than camera trap monitoring in terms of both quantification and data accuracy. However, they finally recommend camera trap monitoring for underpasses given the equipment costs and the time-consuming video analysis.

APPENDIX 1

Typology of structures	Nb	Length (m)				Usefull width (m)				Usefull height (m)		
		mean	±ET	min.	max.	mean	±ET	min.	max	mean	±ET	min.
Wildlife-specific underpass crossing	96	49,2	14,0	30,0	100,0	1,2	1,9	0,5	12	1,1	0,8	0,7
Adapted hydraulic structure - Footway	22	67,1	33,5	24,9	148,7	1,2	0,7	0,5	4	1,6	0,6	0,8
Adapted hydraulic structure - Corbel	13	50,8	16,4	26,0	86,0	0,5	0,1	0,5	0,7	1,3	0,7	0,5
Adapted hydraulic structure - hollow	3	62,9	29,2	30,0	85,8	1,5	1,2	0,8	2,8	2,1	0,6	1,5
Mixed underpass	4	32,8	2,6	30,0	35,0	5,1	3,3	2,5	10	5,3	0,9	4,3
Non-dedicated underpass	1	71,0				7,5				4,5		
Non-dedicated hydraulic structure	24	68,7	25,4	35,0	140,0	5,6	9,9	0,7	36	2,3	1,3	0,8
Eco-bridge	15	56,9	12,3	34,5	75,0	18,6	5,0	11,0	25,8			

Table 10 : Number of monitoring structures by type of structure and average size of the different types of structures

APPENDIX 2

Organisation in charge of monitoring	Number of structures monitored
Association d'Etudes, de Protection et d'Aménagement de la Nature en Touraine (SEPANT)	2
Biotope & Ligue pour la Protection des Oiseaux (LPO)	1
CERA Environnement	3
Cistude Nature	8
COFIROUTE	9
Cofiroute & CPIE Loire Anjou	13
Conservatoire d'espaces naturels de Provence-Alpes-Côte d'Azur (CEN PACA)	2
Ecologistes de l'Euzière	22
ECO-MED & LPO	1
Ecosphère & LPO	1
Egis & LPO	1
Eure-et-Loir Nature	3
Fédération Départementale de Chasse (FDC) de Charente-Maritime	1
FDC de Dordogne	1
FDC de la Corrèze	1
FDC de la Drôme	1
FDC de la Loire	1
FDC de Vendée	3
FDC des Landes	1
FDC des Pyrénées-Atlantiques	5
FDC d'Eure et Loir	1
FDC d'Indre-et-Loire	1
FDC du Puy-de-Dôme	3
FDC de l'Aude	1
France Nature Environnement (FNE) - Loire & FDC de la Loire	26
FNE - Rhône	8
FNE - Rhône & FDC du Rhône	8
Groupe Mammalogique et Herpétologique Limousin (GMHL)	8
LPO Anjou	2
LPO AuRA Drôme-Ardèche	2
LPO Auvergne	1
LPO France	10
LPO Loire	3
LPO PACA	3
LPO Rhône	1
LPO Sarthe	3
Nature en Occitanie	13
Nature Midi-Pyrénées	5
Sarthe Nature Environnement	8

Table 11: List of 40 organisations in charge of monitoring wildlife by camera trap.

APPENDIX 3

Species	Season	Underpasses			Eco-bridges		
		Estimate	Std. Error	p-value	Estimate	Std. Error	p-value
Meles meles							
	Spring	**1.048**	**0.095**	**0.000**	**1.202**	**0.159**	**0.000**
	Summer	**0.636**	**0.093**	**0.000**	**0.497**	**0.161**	**0.002**
	Autumn	**0.502**	**0.094**	**0.000**	0.193	0.170	0.258
Vulpes vulpes							
	Spring	**-0.182**	**0.084**	**0.029**	–	–	–
	Summer	**-0.829**	**0.083**	**0.000**	–	–	–
	Autumn	**-0.848**	**0.084**	**0.000**	–	–	–
Lepus europaeus							
	Spring	0.398	0.213	0.062	**0.302**	**0.136**	**0.026**
	Summer	**0.649**	**0.208**	**0.002**	0.063	0.126	0.615
	Autumn	-0.113	0.235	0.629	**-0.368**	**0.131**	**0.005**
Erinaceus europaeus							
	Spring	**2.255**	**0.353**	**0.000**	–	–	–
	Summer	**3.344**	**0.339**	**0.000**	–	–	–
	Autumn	**2.396**	**0.347**	**0.000**	–	–	–
Capreolus capreolus							
	Spring	–	–	–	**-0.725**	**0.124**	**0.000**
	Summer	–	–	–	0.161	0.120	0.182
	Autumn	–	–	–	0.032	0.120	0.789
Sus scrofa							
	Spring	–	–	–	**-0.373**	**0.122**	**0.002**
	Summer	–	–	–	**-0.713**	**0.119**	**0.000**
	Autumn	–	–	–	**-0.304**	**0.113**	**0.007**
Cervus elaphus							
	Spring	–	–	–	**-1.389**	**0.359**	**0.000**
	Summer	–	–	–	**0.518**	**0.263**	**0.049**
	Autumn	–	–	–	-0.042	0.282	0.881

Table 12 : Influence of season on the occurrence of species in underpasses and eco-bridges. The results in bold indicate significant impacts.

APPENDIX 4: CONSTRUCTION OF A SIMULATION MODEL

Two strata can be defined which correspond to the spaces on either side of the crossing device. Stratum A and B in Figure 88.

There are four transition states that can be achieved between two capture sessions:

- Probability that an individual from stratum A transits toward stratum B φAB
- Probability that an individual from stratum B transits toward stratum A φBA
- Probability that an individual from stratum A remains in stratum A φAA
- Probability that an individual from stratum B remains in stratum B φBB

The principle of the simulation is to generate a realistic mock dataset that is integrated into a multi-state CMR model, which will estimate the number of times the model fails to detect transitions when they exist.

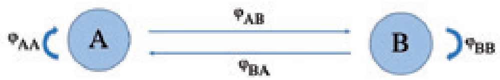


Figure 88 : Schematic representation of the possibilities of transits between two strata. φAB = probability that an individual transits from stratum A to stratum B. φBA = probability that an individual transits from stratum B to stratum A. φAA = probability that an individual remains in stratum A. φBB = probability that an individual remains in stratum B.

It is expected that the number of non-detections of transits decreases when the number of visits, the number of individuals and the probability of detection increases.

Thus, the power test will enable to choose a threshold value of the number of capture sessions to integrate in the sampling protocol in function of the detection probability and the estimated number of individuals.

Material et methods

- Building scenarios: Several hundred scenarios are built for a multi-state CMR model by varying the following parameters:
  - Number of individuals: from 15 to 120 by increments of 15
  - Number of visits: from 3 to 10, then up to 50 by increments of 10
  - Probability of detection: from 10 to 60 %
  - Transition rate: 1 %, 2 % and 5 %. The transition rate from stratum A to stratum B and from stratum B to stratum A are always kept equal (no unbalanced transitions).

The transition percentage values are chosen in function of the CMR monitoring results compiled in the eco-bridge monitoring reports. These reports show that over three years of monitoring, the number of transitions detected is very low, always less than 1% per year, even for populations greater than 100 individuals (micromammals).

The total number of simulations concern 1152 scenarios.

- Building of a simulated data set: For each scenario, a mock dataset is generated from the parameters of the different scenarios. A multi-state CMR model is then applied, taking into account transition rates. For each scenario, 1000 model simulations are evaluated, enabling to compute a mean variance of the transition rates and to evaluate the number of times the model does not detect transitions when they exist. It is this last parameter that is used to evaluate the reliability of the scenarios. Thus, a scenario with a large number of non-detections of transitions will be considered as not very reliable; conversely, a scenario with no non-detection of transitions will be considered as reliable.

These simulations were conducted using the RMark package (LAAKE, 2013) implemented in the R v.4.1.1 software.

APPENDIX 5: PROPOSITION OF A PROTOCOL FOR MONITORING *CARABIDAE* BEETLES ON THE ECO-BRIDGES USED AS CORRIDORS

CEFE-CNRS - Jean-Pierre VACHER, Claude MIAUD, Aurélien BESNARD.

Context

The objective of this monitoring is to highlight the effectiveness of eco-bridges used as a corridor for wildlife crossing. In order to ensure that small fauna crosses the eco-bridge, and in a particular direction, it is necessary to collect geo-referenced observations on both sides of the eco-bridge and to set up a monitoring system that can recognise the individuals. Carabidae beetles are highly mobile terrestrial insects (they do not fly) that move mostly at night and can travel quite long distances in their home range in search of prey or partners. They therefore seem to be good models to measure the effectiveness of eco-bridges used as corridors for small fauna. We therefore propose a protocol based on capture-recapture that enables recognition of individuals and thus modelling of their movements and directions.

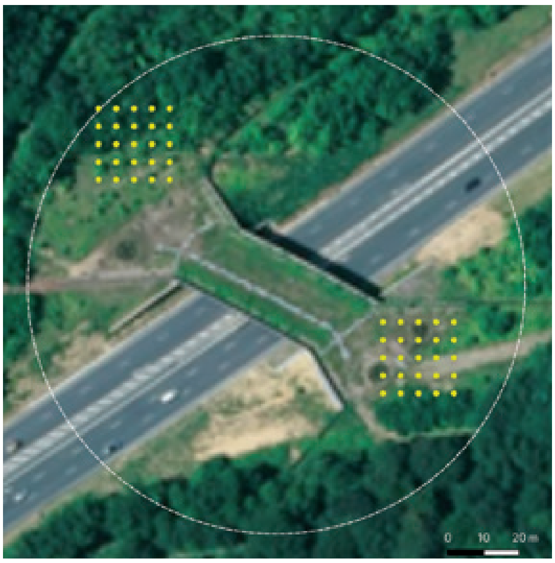


Figure 89 : Schematic representation of the layout of two arrays of pot traps for the interception of beetles.

Protocol

- Definition of the reference landscape unit: The landscape unit of reference will be the eco-bridge with an approximately 50-metre buffer zone surrounding it, calculated from the bridge ends (Figure 89).
- Definition of the sample: Two arrays of 25 pot traps placed on both sides of the eco-bridge (Figure 89). The pots will be 5 metres apart (Figure 89), numbered, and each array will be GPS geo-referenced.

Sampling

Capture-recapture type sampling is carried out on *Carabidae* beetles measuring more than 15 millimetres, i.e., mainly the genera *Calosoma, Carabus, Cychrus, Eurynebria, Broscus, Pterostichus, Abax, Sphodrus*. Marking will be carried out with a combination of dots affixed on the elytra (upper and lower) and the pronotum (upper part of the thorax) with a coloured marker (uni-PAINT© non-toxic paint marker), on the left and right sides.

If you choose to use only one colour, the number of possible combinations is 26-1 per species. If you decide to use two colours, the number of combinations leaves more room to manœuvre, 212-1 per species Figure 90). Nevertheless, the first solution with 63 combinations seems reasonable considering the time and the number of trap pots, but the second option is possible if the number of captures is higher. The colours will be different on each side of the eco-bridge in order to easily recognise the origin of the individuals. The proposed tagging lasts about one month.

Frequency and timing of surveys

Monitoring will include two one-month sessions at two periods of the year, in spring (April-May) and autumn (September-October), because two cohorts of *Carabidae* are likely to use the eco-bridge. The choice of the dates is to be adapted according to the regions. The survey of the pots will be done every morning during three days at the beginning of the monitoring, then the pots will be closed, and opened again for three days (surveyed every morning) the 15th day, then closed again, and finally opened for three days at

Figure 90 : Examples of unique marking with a combination of two colours and six positions on the pronotum (2 positions) and on the elytra (4 positions), here on a Pterostichus niger *individual.*

the end of the monitoring (day 28 until day 30) with a daily morning survey.

Data recorded

All the individuals of the species concerned found in the trap pots will be marked, with the number of the pot in which it was found and photographed once the marking has been applied.

Covariates to be measured at each session

- Air temperature (ground level) to be measured in the field
- Recording date
- Recording time
- Cloud conditions (cloudy/variable/sunny).

Duration and frequency of monitoring

This protocol is intended for one monitoring session, i.e. one month, as the tagging does not last longer. In some contexts of early implementation of the eco-bridge, one year of monitoring could be sufficient to obtain information on the movements of *Carabidae.* Nevertheless, the monitoring could be renewed annually for three years, notably in the case of newly built eco-bridges.

GLOSSARY

APPB: Prefectural order for biotope protection.

ASF: Autoroutes du Sud de la France.

Brushwood (brushwood panels): Natural fencing made from heather twigs collected from brushwood habitats.

B(D)ACI: Before-(During)-After-Control-Impact. Method for assessing the differences between "pre-project"/(during project)/"post-project" statuses in one or more treated area(s), using one or more control site(s).

CEFE: Centre of Functional and Evolutionary Ecology (Joint Research Centre at the University of Montpellier).

CEREMA: Centre for Studies and Expertise on Risks, the Environment, Mobility and Urban Planning.

Cesco - MNHN: Centre for Ecology and Conservation Sciences - National Museum of Natural History.

CMR: Capture – Mark – Recapture. A statistical method for estimating the size of an animal population.

COST 341 Report: Habitat fragmentation due to transport infrastructures, SETRA, September 2007.

DDT: Departmental Territories Direction.

DREAL: Regional Environment, Planning and Housing Directorate.

DRJSCS: Regional Directorate for Youth, Sports and Social Cohesion. Since 2021, divided into Regional Academic Delegations for Youth, Commitment and Sports (DRAJES) and Regional Directorates for the Economy, Employment, Work and Solidarities (DREETS).

Environmental DNA: DNA extracted from samples collected in an environment (water, soil, faeces…) without directly targeting an organism.

False negative: A false negative (false negative test) is when the test result is not true.

FDC/FNC: Départemental and national Hunters' Federation.

Feral cat: Domestic cat that has returned to the wild.

Glirids: Family of medium-sized rodents (dormice, muskrats…).

Hibernaculum: Artificial shelter, refuge used by small fauna during hibernation or as a regular shelter the rest of the year.

ICE: Information on Ecological Continuity.

Mustelids: Family of carnivorous mammals (European Badger, Weasel, Stoat, Polecat…)

NNR: National Nature Reserve.

OFB: French Biodiversity Agency.

Rex 1: First VINCI-Motorways feedback published in 2016.

Rex 2: Second VINCI-Motorways feedback published in 2023 (this document).

RFID: Radio Frequency Identification. Méthode de mémorisation et de récupération de données à distance par transfert d'énergie électromagnétique.

Rhopalocera: taxon used for the *Papilionoidea* superfamily. Familiarly but inaccurately used to refer to "butterflies".

RNCFS: National Hunting and Wildlife Reserve.

RNR: Regional Nature Reserve.

SETRA: Service for Studies of Transports and Roads and their Development (now CEREMA).

SCI: Site of Community Importance.

SIPAF: Wildlife Crossing Information System.

SPA: Special Protection Area.

SRADDET: Regional Plan for Planning, Sustainable Development and Equality of Territories.

Time-lapse: A technique used in photography to automatically take photographs at a defined regular frequency and over a defined period of time.

ZNIEFF1: Natural Area of Ecological, Floristic and Faunistic Interest Type 1.

ZNIEFF2: Natural Area of Ecological, Floristic and Faunistic Interest Type 2.

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