

Action C5.
Extension of the Prevention and
early warning system to other Natural Parks

Deliverable:

Final report on findings in the replication sites

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SUMMARY

Xylosandrus compactus (black twig borer) and *Xylosandrus crassiusculus* (granulate ambrosia beetle) are highly polyphagous parasites that can infest many tree and shrub species. They originated in Asian regions and then spread to other parts of the world, probably through the plants and timber trade. These bark beetles, which host symbiotic fungi, dig tunnels in young branches (*X. compactus*) and trunks (*X. crassiusculus*) of trees. Infested trees show damage such as desiccation, branch dieback and shoots breakage.

Both beetles were introduced in various regions of Italy and showed a significant expansion since about 2014, infesting several places along the Cote D'Azur in France and, as far as *X. crassiusculus* concerned, emerging in 2016 the Spanish Valencia region.

Whereas infestations were mostly known to cause harm to cultivated plants, a massive outbreak in the Italian National Park Circeo in 2016 brought awareness on the risks involved for natural forest stands in protected areas. The alarming damage to the Circeo promontory, the increasing detections of both species of *Xylosandrus* along the Tyrrhenian coast in or near natural parks, and the consequent vulnerability of many species of Mediterranean vegetation, motivated LIFE SAMFIX team to:

- test and implement protocols and tools to deal with the threats;
- to gain knowledge to better understand the pathways of spread of these bark beetles, the causes of damage and the risks to ecosystems;
- and to share the results with other stakeholders.

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Prevention, early warning and rapid response protocols were experimentally implemented in a range of core sites: in Italy in the Circeo National Park (Lazio Region); in Spain in El Tello and its surroundings (Valencia region); and in France at Villa Thuret and Bois de Garoupe in Antibes; Mont Boron and Saint-Jean-Cap-Ferrat extended to the borders of Corniches de la Riviera; and on the Island Ile Ste Margherite (PACA Region).

In order to ensure a rapid and effective response to the possible spread of *Xylosandrus* spp. outside the areas object of project core actions where the insects were found, Action C.5 of the project was focused on the extension of prevention and early warning systems to other natural parks. This was to be done on the one hand by the delivery of training packages and generalised protocols that are broadly usable and that were disseminated. On the other hand, protected areas in the neighbourhood of the core sites were assisted to replicate prevention and early warning protocols. They were provided with protocols adapted to the targeted sites, trainings, traps and lures, and technical assistance in the deployment of the trapping networks, the interpretation of findings and advice on eventual interventions.

This report aims to relate on the findings in these so-called replication sites.

In Italy, the activity was foreseen in 5 Regional Parks located around the Circeo National Park; Riviera di Ulisse, Monti Aurunci, Monti Ausoni e Lago di Fondi, Castelli Romani e Tor Caldara. To those, following requests to participate, the Islands of Ventotene and Zanone, the Presidential Estate of Castelporziano and the Giardino di Ninfa and Pantanello Oasis were added. In all areas, except from the Islands, expansion could be ascertained, though of very limited extend and without relevant damage.

In France, replication was foreseen in Port Cross National Park, but activities were extended to its bounding areas on the island of Porquerolles and nearby continental coastal areas. The entire coastal zone from Menton until Toulon was monitored, with a further extension to the Var Valley, in protection of the Regional Natural Park of Sainte Baume and the Mercantour National Park; to the Regional Parks of Paradou and Croix des Gardes, and to the Regional Park of Corsica, where records were found. Also in France, expansion was acknowledged, except on the Islands of Port Cross and Porquerolles, but limited to isolated spots moving westwards and northwards.

In Spain, the Natural Parks Sierras de Martés y el Ave and Muela de Cortes y el Carroche were focused. Dissemination activities led to several detections and consequent expansion of replication activities in private gardens in Náquera, Banyoles, Vidreres, Vila-Seca, Platja D'Aro and on the Island of Majorca. In Sierras de Martés y el Ave could indeed a limited presence be recorded, while in Muela de Cortes y el Carroche no infestation was found.

The interest of public authorities responsible for natural parks and plant health and from private owners of infected trees was very satisfactory. Early warning protocols could be implemented, visual inspections were performed, some containment trials could be performed, and where needed competent bodies followed up pruning or elimination suggestions.

The findings show that *Xylosandrus spp.* are spreading around the Tyrrhenian coasts, but their spread is generally limited to isolated spots and usually without significant damage.

Overall, it can be concluded that early warning networks should be maintained in urban and landscaped areas bordering natural parks, including in nurseries and biowaste treatment locations, providing pathways for introduction into protected areas, while monitoring networks in areas yet affected together with visual inspection is needed to prevent expansion and damage. Accurate pruning and removal should be evaluated and applied where possible and infestation is limited.

Moreover, broad awareness raising and knowledge transfer result to be pivotal for detection and subsequent protective interventions.

Introduction

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Both beetles were introduced in various regions of Italy and showed a significant expansion since about 2014, infesting several places along the Cote D'Azur in France and, as far as *X. crassiusculus* concerned, emerging in 2016 the Spanish Valencia region.

Whereas infestations were mostly known to cause harm to cultivated plants, a massive outbreak in the Italian National Park Circeo in 2016 brought awareness on the risks involved for natural forest stands in protected areas. The alarming damage to the Circeo promontory, the increasing detections of both species of *Xylosandrus* along the Tyrrhenian coast in or near natural parks, and the consequent vulnerability of many species of Mediterranean vegetation, motivated LIFE SAMFIX participants to combine the scientific expertise that had been acquired to:

- test and implement protocols and tools to deal with the threats;
- to gain knowledge to better understand the pathways of spread of these bark beetles, the causes of damage and the risks to ecosystems;
- and to share the results with other stakeholders.

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Prevention, early warning and rapid response protocols were experimentally implemented in a range of core sites: in Italy in the Circeo National Park (Lazio Region); in Spain in El Tello and its surroundings (Valencia region); and in France at Villa Thuret and Bois de Garoupe in Antibes; Mont Boron and Saint-Jean-Cap-Ferrat extended to the borders of Corniches de la Riviera; and on the Island Ile Ste Margherite (PACA Region).

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This report aims to relate on the findings in the replication sites.

The sites that had been selected for replication of the protocols were the following:

Italy (Lazio Region):

- Regional Park of Riviera di Ulisse - SCI-SAC-SPA IT6040022; SCI-SAC-SPA IT6040023; SCI-SAC IT6040024;
- Regional Park of Monti Aurunci - SPA IT6040043
- Regional Park of Monti Ausoni e Lago di Fondi - SPA IT6040043
- Regional Park of Castelli Romani - SCI IT6030018, IT6030017 and IT6030039
- Regional Natural Reserve of Tor Caldara - SCI-SAC IT6030046.

Spain (Generalitat Valenciana):

- Sierras de Martés y el Ave - SCI ES5233011 and SPA ES0000212
- Muela de Cortes y el Carroche SCI ES5233040 and SPA ES0000212

France (PACA Region):

- Port Cros National Park (SPA FR9310020 and SCI FR9301613)

During the project, following awareness raising on the issues at stake and dissemination of the first project results, several other areas were added to the list, on request of the owners or management bodies:

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Italy (Lazio Region):

- State Natural Reserve of Ventotene and S. Stefano Island – SPA IT6040019
- State Natural Reserve of Castelporziano – SCI IT6030028 and SPA IT6030084
- Natural Monument Giardino di Ninfa Garden and Pantanello Oasis
- Island of Zanone - SPA IT6040019

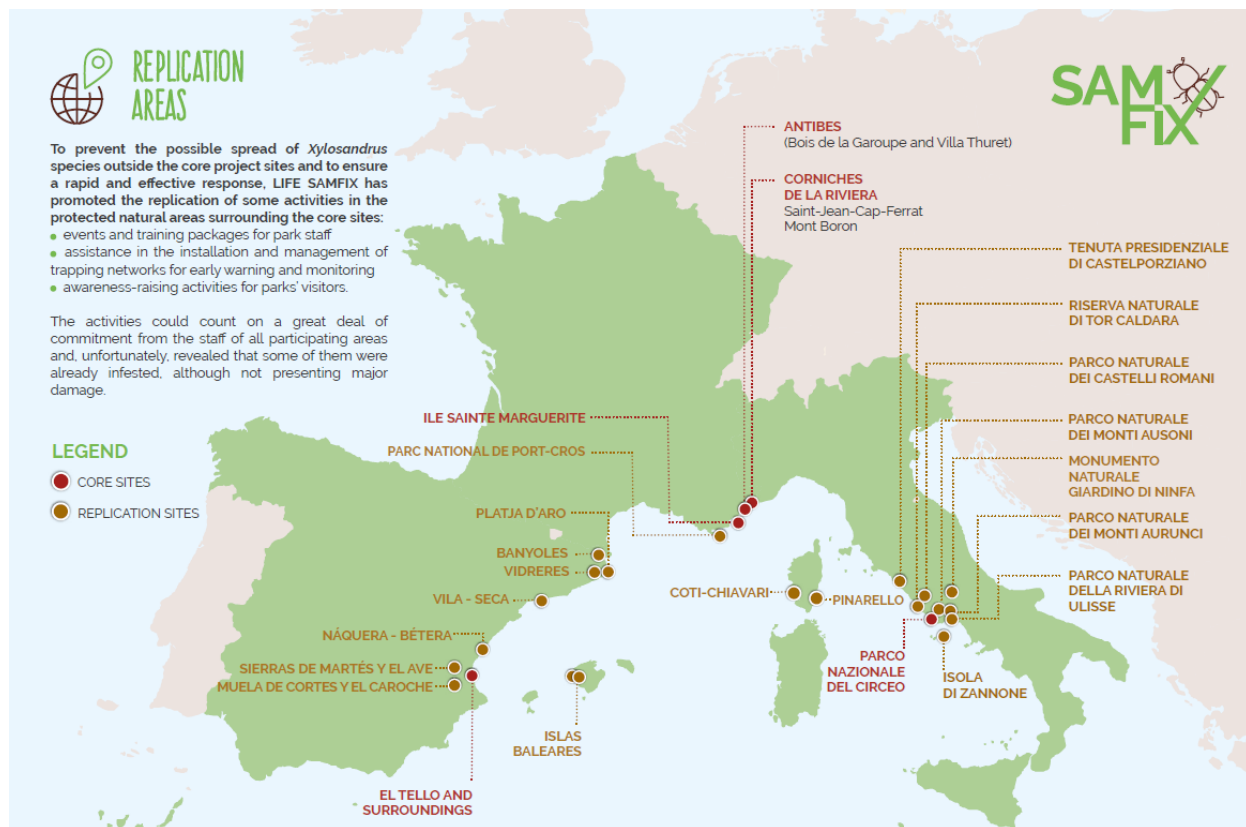
Spain (Generalitat Valenciana):

- Private gardens in Náquera, Banyoles, Vidreres, Vila-Seca, Platja D'Aro
- El Toro, Island of Majorca, bordering Cap de cala Figuera – SPA & SCI ES0000074

France (PACA and Corsica):

- Coastal areas until Toulon, to early warn for expansion towards Regional Natural Park of Sainte Baume (SCI FR9301606)
- Var Valley, to early warn for expansion towards Parc National du Mercantour – SCI FR9301556, FR9301564, FR9301563 and SPA FR9310035
- Regional Parks of Paradou and Croix des Gardes
- Regional Park of Corsica- Pinarellu - FR9400606

The following chapters will be each dedicated to one of the three countries.



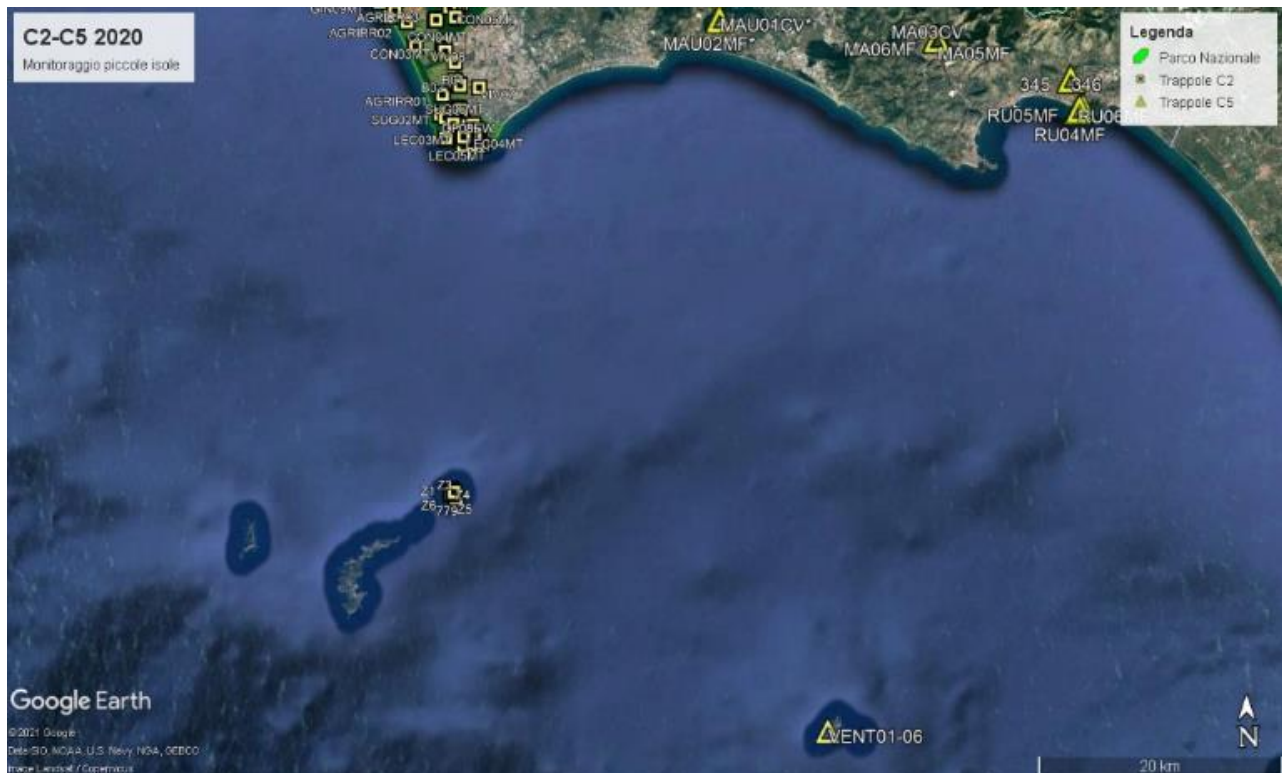
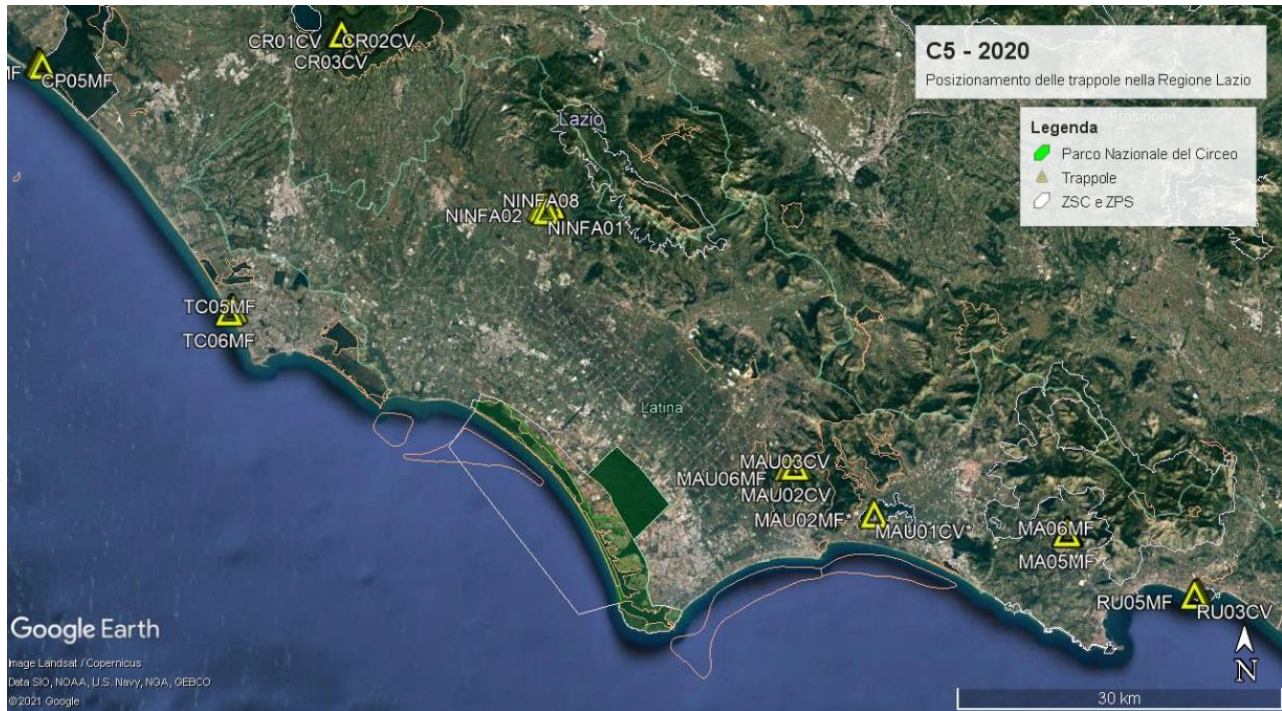
1. Findings in the replication sites in Italy

In Italy, prevention, early warning and rapid response protocols were planned to be carried out in 5 regional natural parks around Circeo Park, that fall under Lazio Region competences:

- 1) Natural Park of Riviera di Ulisse - SAC-SPA IT6040022; SCI-SAC-SPA IT6040023; SAC IT6040024;
- 2) Natural Park of Monti Aurunci - SPA IT6040043
- 3) Natural Park of Monti Ausoni - SPA IT6040043
- 4) Natural Park of Castelli Romani, in particular the SAC IT6030018, IT6030017 and IT6030039
- 5) Natural Reserve of Tor Caldara SAC IT6030046.

To those, the following parks were added during the implementation of the project:

- 6) State Natural Reserve of Ventotene and S. Stefano Island – SPA IT6040019
- 7) State Natural Reserve of Castelporziano – SCI IT6030028 and SPA IT6030084
- 8) Natural Monument Giardino di Ninfa Garden and Pantanello Oasis
- 9) Island of Zannone - SPA IT6040019



2.1 Natural parcs of Riviera di Ulisse, Monti Aurunci, Monti Ausoni and Lago di Fondi, Castelli Romani, Tor Caldara

2019

Although planned to take off in 2020, monitoring activities in replication areas of Lazio Region were started up yet from May 2019, following the advice of project scientists that became aware of the rapid spread acknowledged during 2018 along the Tyrrhenian coast.

During 2019, a limited activity was performed which exclusively served early warning of spreads, using traps baited with α -pinene and ethanol blend.

A training event for park collaborators (foreseen in Action A4) was extended to the staff of all regional parks. It took place on 22-23/03/2019 at Circeo park, and 35 park collaborators from 7 Parks in the Region participated (including Circeo and Castelporziano), as well as rangers from the Carabinieri Forestali and a municipality technician. Prof. M. Faccoli explained *Xylosandrus* spp. characteristics, preferred host plants and the typical damage. He also gave a practical instruction on how to manage the baited traps, collect insects and remove infested plants.

Circeo staff supported the parks' staff in the installation of the trapping network and the start up of surveillance activities. Small plots of 6 traps (3 multifunnel and 3 cross vane) were installed in May in the Regional Parks Monti Aurunci, Castelli Romani and Riviera di Ulisse. In June last traps were placed at Tor Caldara and at Monti Ausoni.

In June Circeo Park and UNITUS staff provided for the first collection of samples and local staff was trained on the job to proceed independently. In September, Circeo Staff had several short meetings with some members of regional Park staff regarding the traps management related to emerging problems. In October-November Circeo staff collected the last samples of all the parks and delivered them for analysis to UNITUS. Hereafter, the parks removed the traps and stored them for the winter.

2020-2021

A first extensive trapping protocol was designed and shared with park staff in April 2020.

The planned installation of the trapping network, foreseen in May, had to be postponed to end of June-July, due to the mobility restrictions imposed by the COVID-19 pandemic. Training was provided on the job by Circeo staff.

In March 2021 an online course was published, to refresh and update general information. Additionally, at the end of March 2021, an online meeting was organised for all the representatives of the regional parks on the trapping and collection protocols to be applied this year, with the support of Circeo Park and the scientific staff.

In 2021 traps could be installed in April.

In each of the protected areas, 12 multifunnel traps were placed, baited with quercivolo, alfa-copaene, ethanol and alfa-pinene. During 2020, the traps were checked every 15 days and lures replaced each 60 days. In 2021 trap control was performed regularly every 3 weeks and attractants replaced every 6 weeks, in accordance with the revised protocol agreed with UNITUS and Circeo. All 2020 and 2021 samples were delivered to UNITUS to be analysed.

In both years, traps were removed by end of October and stored for the next season.

The collected information was useful in defining the voltinism and phenology of *Xylosandrus* spp.: crucial data for implementing adaptative strategies for planning activities and actions of early warning.

The trapping networks were accompanied by territorial phytosanitary surveillance. The presence of damage attributable to *Xylosandrus* spp. was assessed in sensitive areas of the parks such as areas subject to previous infestations of other parasites or pathogens, or suffering from abiotic stress (drought, fires, heat waves ...) by means of a visual surveillance of the territory conducted in the period of maximum insect flight activity (June-October). On a monthly basis, the territory of the parks were explored in search of any outbreaks of infestation represented by trees or groups of trees with reddish foliage and small entrance holes on the main stem and branches. These outbreaks were promptly reported, georeferenced and quantified in terms of number of trees or forest area affected. The temporal trend of any infestation was also checked in the subsequent months.

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Staff had been instructed to apply the following modus operandi in case of detections:

- if specimens are collected in traps.

After confirmation of species by the SAMFIX team, an intensive surveillance of potential host in a radius of 100m around the traps should be performed. The personnel will be further trained and equipped with monitoring tools and a dedicated form, where some information should be reported: trap code and coordinate, date of collection, host tree species presence and, if the case, abundance and distribution of host around the trap, date and staff name that carried out the survey. Damage to trees should also be reported.

- if symptoms in host trees are detected.

After confirmation of the presence of *Xylosandrus* spp. by the SAMFIX team, the following actions should start:

A - filling a specific form that includes trap code and coordinate, date of collection, host tree species presence, and if the case, abundance and distribution of host around the trap, date and staff name that carried out it;

B - pruning or cutting the affected trees, and destruction *in situ* of infested vegetable material before a week since confirmation;

C - intensive surveillance of potential host in a radius of 500m around the affected tree of the potential host. If new attacked plants are detected, the same process of a point should be applied for each affected tree.

Staff members of the Regional Park of Monte Ausoni launched an alert on a potential infestation by *Xylosandrus crassiusculus* of carob trees in the urban park "Villa Placitelli" in Fondi. An inspection followed hereupon, with support of Circeo National Park staff, collecting samples and warning the project's entomologists.

2022

Following the results of monitoring activities in 2019, 2020 and 2021, showing very few occurrences and no damage, the risk assessment led to the decision to provide in 2022 for a very limited monitoring activity, focused on the urban area of Fondi, the only place where a relevant outbreak was ascertained. Meanwhile a long-term regional collaboration is being prepared to assure future monitoring.



Field activities in the 5 Regional Parks

Results

The table below shows the results of the insects caught by the trapping networks in the five replication areas from 2019 till 2021. For PN Monti Ausoni results are presented in two different rows, as the two sampling localities were quite distant from each other.

	2021			2020			2019		
	COMP	CRASS	GERM	COMP	CRASS	GERM	COMP	CRASS	GERM
PN Monti Aurunci	-	4	20	3	1	13	7	36	299
PN Monti Ausoni - Campo Soriano	-	-	1	5	0	8	-	1	-
PN Monti Ausoni – Fondi	1	86	129	1	0	13	-	-	-
PN Castelli Romani	3	1	1144	-	-	-	NA*	NA*	NA*
RN Tor Caldara	2	-	9	-	-	-	15	-	8
PN Riviera di Ulisse	-	5	4	1	9	0	12	28	20

COMP = *X. compactus*; CRASS = *X. crassiusculus*; GERM = *X. germanus*

* The 2019 samples of the Parco dei Castelli Romani were lost due to problems with their conservation, and thus caught insects could not be determined in the laboratory.

During the first recognition in 2019, *X. compactus* was detected in Monti Aurunci, Riviera di Ulisse and Tor Caldara; and *X. crassiusculus* in Riviera di Ulisse, Monti Ausoni and Monti Aurunci. Unexpectedly, also *X. germanus* was found in Monti Aurunci, Riviera di Ulisse and Tor Caldara.

In 2020 very few beetles were caught. It was expected that this was mainly due to having missed the first flight of the beetles, occurring in spring, when the COVID-19 restrictions prohibited to install the trapping network and collect specimens.

In 2021, apart from an increase in *X. crassiusculus* occurrence in Monti Ausoni-Fondi (at carob trees in the urban park "Villa Placitelli"), no significant changes were registered in the presence of *X. compactus* and *X. crassiusculus*, appearing limited to isolated spots. Instead, a significant increase of *X. germanus* could be ascertained, especially in Castelli Romani and Monti Ausoni-Fondi.

Results of field activities showed that *Xylosandrus* spp. has spread outside the Circeo National Park. In Castelli Romani and Monti Ausoni all the *Xylosandrus* species were detected. No relevant damage on trees was anyhow spotted and thus there was no need for phytosanitary interventions.

The presence of *Xylosandrus* in the five protected areas identified for the monitoring activities was expected, since there are several apparently suitable areas to assist the dispersion (mainly small woodlots or forest fragments) located between the Circeo National Park and the protected areas where the monitoring took place.

2.2 State Natural Reserve of Ventotene and S. Stefano Islands

After several worrisome reports on deterioration of trees, the State Reserve of Ventotene and S. Stefano Islands was added to the list of sites to be monitored to investigate the situation. The Natural Reserve is located in Ponziene Islands, about 40 km from Southern Latium coast, in front of Circeo National Park.

Since July till October 2020 and from April till October 2021 one transect of six monitoring traps baited with ethanol was put in action in the western part of Ventotene Island, in an area covered with natural scrub and maquis vegetation. Traps were checked according to the monitoring protocol.

Detailed investigations and laboratory analyses did not record any *Xylosandrus* spp. sign of presence.

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2.3 State Natural Reserve of Castelporziano

Yet in May 2019, small plots of 6 traps (3 multifunnel and 3 cross vane) were installed also in Castelporziano Reserve (IT6030084). This reserve, not covered by Lazio Region competences, had not been foreseen in the project, but was considered pivotal by the team and the competent Presidency of the Republic was eager to participate, as it is situated very close to Circeo National Park and thus very sensible to infestation. Staff had been instructed during the training event in April, and was assisted by Circeo park in the installations.

During the first recognition in 2019, *X. compactus* and *X. germanus* were detected in Castel Porziano, while *X. crassiusculus* resulted absent.

During 2020 and 2021 Castelporziano developed the same protocol as applied in the 5 Regional Parks and also participated to the dedicated online meetings.

The total catch achieved in 2020 was extremely low, with only the capture of one adult of *X. germanus*. In 2021, *X. germanus* specimens captured increased to 3 while the other two species were not captured. No damage on trees was found.

2.4 Natural Monument Giardino di Ninfa and Pantanello Oasis

Following a cooperation agreement between the Fondazione Caetani and Circeo National Park, a monitoring campaign for early warning purposes was defined for both the “Giardino di Ninfa” and the “Oasis Pantanello” recognised as Natural Monuments but privately managed and owned by the Fondazione Caetani. They are located in the Latina province, in between the Circeo Natural Park, the Castelli Romani and the for Caldara regional park, and could thus configure perfect stepping stones for dispersion of the infestation.

After the COVID-19 mobility ban was relieved, in July 2020 trapping networks were installed at both sites. They were accompanied by billboards explaining the project and its objectives, to provide for an additional important location of awareness raising.

Park staff was supported by Circeo park staff in the installation of traps and was instructed on the substitution of lures and the emptying of the traps, as well as on the performance of surveillance activities. Circeo park collected regularly the trap contents and delivered it to UNITUS for the identification of captured beetles.

No *Xylosandrus* spp. were found. However, due to the fact that the trapping network could only be installed since July, these data were not considered reliable as the first flight peak was missed..

In 2021 the activity continued: from the beginning of April till mid-October 10 traps were installed and monitored in the area of the Ninfa botanical garden and the Pantanello Oasis. According to the agreed protocol, the traps were monitored every 21 days, while the attractors were changed every 6 weeks.

During the year a total of 13 specimens of *X. compactus* were trapped, 7 of *X. crassiusculus*, while 141 of *X. germanus*.

As no damage was found on the vegetation, the situation was not considered critical or requiring any kind of intervention, apart from the continuation of population monitoring. Therefore, the Fondazione Caetani re-installed the same trapping network also in 2022.

It should be mentioned that in spring 2021 a broader convention was signed with UNITUS and Circeo Park, to proceed with future entomological investigations in the Fondazione Caetani locations in forthcoming years. This convention can also host the maintenance of *Xylosandrus* analyses in the future.

2.5 Island of Zannone

Also at the “Isola di Zannone” traps were installed in June 2020 to provide for a recognition on the presence of the beetles.

No *Xylosandrus* spp. were found.

2.6 Evaluation

Xylosandrus beetles have been detected in all the five replication areas managed by Lazio Region, in Castelporziano and in Giardino di Ninfa, but not on the islands of Ventotene and Zanone. Therefore, the risk of dispersion of these alien species should be assessed. All three *Xylosandrus* species are present (*X. compactus*, *X. crassiusculus*, *X. germanus*). In the Lazio region there are several plant species that can be attacked by this species, although the most important infestations so far occurred on the holm oak (*Quercus ilex*).

Given the presence of many common tree and shrub species amongst the beetles' preferred hosts, they can easily move using gardens, parks and ornamental trees as stepping stones, it is therefore difficult to estimate the possibility that species may expand their range into protected habitats, which are difficult to be surveyed. First attention should thus be posed on surveillance in non protected areas around the natural parks, from where invasion can occur, in particular nurseries, urban parks and private gardens. In these areas, it is also more easy to quickly intervene with pruning and removal operations.

The phytosanitary staff of the Lazio Region has been alerted, and is able to report the possible presence of the species even in semi-natural or suburban contexts.

Nonetheless, should the species be discovered, by direct observation or by detecting its signs of presence, management strategies proposed by the project staff, although difficult to be successfully applied, should be evaluated.

Protocols to continue prevention, early warning and rapid response will have to be updated yearly, on the basis of risk assessments, based on the project findings.

3. Findings in the replication sites in France

In France, *Xylosandrus crassiusculus* has first been detected in 2014 at Mont Boron in the area of Nice whilst *X. compactus* was further detected in 2015 simultaneously at both the nearby town of Antibes and far to the West at Saint Tropez. The first species showed continuous outbreaks at Mont Boron and expanded to the north and to the west during the recent years, reaching Saint Raphael in the West. The second species began to invade the area of Nice but its populations remained rather limited in size. Moreover, it did not seem to expand largely around the western spot of Saint Tropez and Bormes les Mimosas.

In France, Port Cros National Park had been originally targeted for prevention and early warning on *Xylosandrus* spp. infestation. It covers the main parts of the Islands Port-Cros and Porquerolles, which are part of the SPA FR9310020 and SCI FR9301613 Rade d'Hyères. The Islands host, amongst others, 30 Ha of coastal *Olea* and *Ceratonia* forests (Annex I – 9320), while inland

forests are dominated by *Quercus ilex* (Annex 1- 9340), amongst the preferred hosts of *Xylosandrus* spp.

However, it was considered more effective as early warning and protection strategy to broaden the sites to be put under observation, to become aware of the dispersion directions and thus to protect more susceptible sites. This had also been considered prior to the submission of the SAMFIX project, but stakeholders had not been able by then yet to confirm their cooperation, which instead was achieved during the project's start-up.

As such, many places along the coastline, from Menton till Toulon, were included in the replication strategy, providing also for early warning of the Regional Natural Park of Sainte Baume (SCI FR9301606), and parts of Porquerolles Island (SCI FR9301613) outside of the Park were included as well. Following signs of northwards dispersion, also the Var Valley was included, to monitor eventual expansion towards Parc National du Mercantour (SCI FR9301556).

Lastly, after reports on *Xylosandrus* detections on the Island of Corsica, the SAMFIX team provided assistance to the implementation of protocols around Pinarellu (SCI FR9400606) and Coti-Chiavari (close to SCI FR9400616), in protection of the Regional Park of Corsica.

3.1 Year 2019

Like for Italy, The SC decided during its January 2019 meeting to anticipate to 2019 the trapping campaign in the surroundings of the French core sites, due to the ascertained quick spread that had been acknowledged during 2018. A limited activity was performed which exclusively served early warning of spreads, using traps baited with α -pinene and ethanol blend.

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The French team prepared a list of 22 monitoring sites selected in the south-east of France (from Menton to Toulon) and organized all the contacts. At each site, one trap (16 sites) or two traps (6 sites) were deployed, baited with α -pinene and ethanol blend and specifically serving the monitoring and early warning of spread. The covered area included the core sites Mont Boron, Saint-Jean-Cap-Ferrat, Corniches de la Riviera and Ile Ste Marguerite; but also Porquerolles island just close to the limits of the National Park of Port-Cros; and the Western Var to survey the expansion with regard to Natura 2000 sites located there. These latter areas had already been considered during project writing as highly relevant, but by that time were not able to confirm their collaboration. In the end, 30 locations were covered in south-eastern France by the trapping network completed at the 14th of April.

Also in surrounding nurseries traps were installed: one in the Ville de Cannes nursery, one in the Nursery of Rayol (the biggest in this region), 2 by orchard managers at Chateau Leoubé, and ONF installed 4 traps in nurseries in a forest (domaine du Ruscas in the massif des Maures) and one at Ile Ste Marguerite.



2019 Trapping network to survey the western expansion from core sites towards Port Cros Park. Closest record of *X. compactus* to Port-Cros figured in red; closest record of *X. crassiusculus* figured in green.

The competent management bodies of these other sites took part in the placement of the traps and were responsible for their management and collection, to which end they had been trained on the 26th of March (action A4). INRA URZF and UEVT staff assisted them, regularly met for collection and discussion of trapping results, and analysed the collected insects. In October the traps were removed from all the French sites and stored for the winter in the lab at Orleans.

At Porquerolles, possible damage on Carob trees was also surveyed but no damage by ambrosia beetles was noticed.

Results

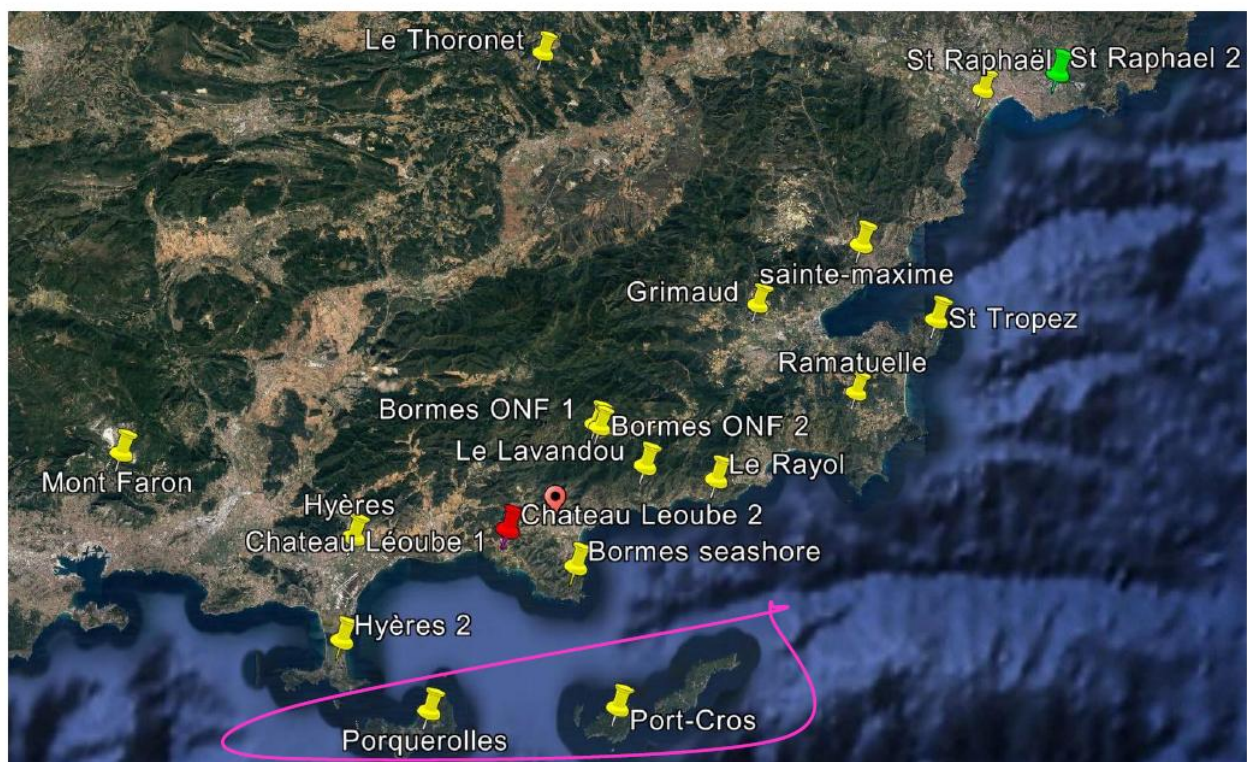
At the island of Porquerolles no records were found, neither at closeby mainland site of Hyères. However, a few specimens of *X. compactus* were trapped in a nearby site at Château Léoube where the beetle was already observed the last year. More generally, a westwards expansion of *X. compactus* was registered with first records in Saint Raphael and Ile Ste Marguerite. *X. crassiusculus* was found in 7 sites, including the Var valley and particularly many specimens in

Mount Boron. *X. germanus* was recorded in 9 plots and especially frequent at the island of Ste Marguerite and in Saint Tropez.

3.2 Year 2020

Given the expansion ranges observed in south-eastern France for each of the beetle species during 2019, we proposed to settle a trapping network extending from Antibes to Hyères (the closest continental site to the islands of Port-Cros) in order to monitor the 2020 expansion towards the national park Port-Cros. The network included a total of 20 sites to be surveyed, including 2 traps that were planned to be settled within the National Park of Port-Cros, on each of the islands Port-Cros and Porquerolles.

Moreover, monitoring around the core sites was to be enforced, among which the regional natural parks of Croix des Gardes and Vallauris Paradou in Cannes, but also extending the surroundings of Corniches de la Riviera Park, with 4 traps in Nice and the upper Var Valley.



Suggested 2020 trapping network for early warning of expansion of *Xylosandrus* spp. and possible incursion to the National park of Port-Cros (delimited by the purple line). Closest record of *X. compactus* to Port-Cros figured in red; closest record of *X. crassiusculus* figured in green.



Suggested trapping network 2020 in and around the core sites

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The trappings were planned to start from early April on. The delivery of the material to partners was planned for 24-25 March in Antibes. Unfortunately, the general confinement in France due to COVID-19 since 16 March prevented this delivery, and thus the settlings of the traps in the field were postponed until the restrictions would be lifted.

Since late June, the traps around the core sites could be installed, featuring 33 traps at 18 sites.



19

Installed 2020 trapping network in and around the core sites Antibes, Isle Ste Marguerite and Corniches de la Riviera Park: (in purple the new traps added in 2020)

Since July, a network of traps could be settled in 17 sites from the closest site of presence of each species of *Xylosandrus* (Chateau Léoube for *X. compactus* and Saint Raphael for *X. crassiusculus*) until the seashore nearby to Port-Cros Park in order to be capable of detecting as early as possible natural spread of each species towards the park. A provisional protocol was delivered mid May to the Port-Cros Natural Park authority, but in the end we did not get any reply from the park managers, and thus no trappings could be implemented in this park this year. However, with the support of a private company, 2 traps could be deployed from early July onwards at Porquerolles island, just close to the limits of the Park areas on this island.



Installed 2020 trapping network for early warning of expansion of *Xylosandrus* spp. and possible incursion to the National park of Port-Cros (delimited by the blue circle).

20

Multifunnel black traps were used, baited with the attractive lure that had been defined. Simultaneously, it was proposed to park rangers to look for any symptoms of tree and shrub damage on the islands of the National Park, especially surveying the species considered as sensitive to *Xylosandrus* spp. such as Carob trees.

Stakeholders were instructed, in case of captures of *Xylosandrus* on the continent in traps located elsewhere than in the areas where the beetles have already been recorded, to check all sensitive plant species for damage symptoms within a radius of 100m. Hereupon, possible pruning or removal of infested plants was to be discussed with officers of the Forest Health Department.

On request of the Corsican authorities, that suspected an infestation on the island, additional traps were settled by mid-June 2020 on the western coast at Coti-Chiavari, south of Ajaccio, and at Sainte-Lucie de Porto-Vecchio on the eastern coast.

Results

Around the core sites, the following was found:

At Corniches de la Riviera a first record was found of *X. crassiusculus*, although we cannot ascertain if this implied an expansion or a previously existing presence. In the nearby Nice, pre-existing *X. compactus* presence appeared low.

X. crassiusculus was also found in Levens, northwards in the upper Var Valley, close to Brec d'Utelle (SCI FR9301563) and Monte Férian (FR9301564).

In the regional parks of Cannes Vallauris Paradou and Croix des Gardes a limited presence of *X. crassiusculus* was found but a massive presence of *X. compactus*.

A new invader, *Amasa truncata*, was trapped in numbers in all areas with Eucalypts (Thuret, Ile Ste Marguerite, Croix des Gardes, Paradou).

Additional visual surveys performed in private gardens and nurseries near the core areas showed a very high density of *X. compactus* at Villefranche and Vallauris (Cannes).

Westwards from Antibes to Hyères the following was observed:

No *Xylosandrus* spp. were trapped at the Porquerolles Island, nor at the seashore near the Park of Port-Cros. The network revealed the presence of a few *X. compactus* in two sites of the Var seashore, rather far (>50 km) from Port-Cros: at Agay- St Raphaël (3 individuals) and Roquebrune sur Argens (1 individual). The other traps did not catch any *Xylosandrus*. Conversely to 2019, no *Xylosandrus* were trapped at Chateau Léoube vineyards nor at Le Rayol nursery, which are both located closer (<5km as the crow flies) to the National park.

However, no conclusions could be drawn, as it is likely that we missed the first beetle generation because of the delayed settlement of the traps due to COVID-19's lockdown. Herewith we missed probably the peak of emergence, which occurs usually mid-May to early-June for *X. crassiusculus*; and late May- late June for *X. compactus*.

A repetition of the networks in 2021 was thus necessary, in order to be capable of appreciating the population dynamics of both beetles.

The additional traps settled in Corsica revealed the presence of *X. compactus* on the western coast at Coti-Chiavari, south of Ajaccio but no *Xylosandrus* were trapped at Sainte Lucie de Porto Vecchio on the east side of the Island. However, visual inspections revealed damage on a carob tree in Pinarello, located just nearby (10 km) Sainte Lucie, which required further inspections next year.

3.3 Year 2021

A meeting with all stakeholders was organised on the 9th of February in city council of Antibes. Taking into account the problems occurred in 2020, specific arrangements were adopted in order to guarantee the beginning of the trappings by early April until October, and thus allowing a reliable comparison of the numbers of trapped beetles between 2019 and 2021. A detailed design for the 2021 network was delivered on 26th of February.

The trapping on the seashore was maintained at the same sites as in 2020 in order to appreciate the differences with the 2020 captures. A special attention was put on the two sites of former presence of *X. compactus* of Chateau Leoubé and Le Rayol where survey of damaged shoots was also organized in order to confirm or not the absence of the beetles.

During the first week of April, 15 multifunnel traps were deployed at 9 sites along the seashore, baited with the combination of 4 attractive lures previously defined. Collection of the trapped insects was performed every 3 weeks and the attractants were changed every 6 weeks by the collaborating stakeholders with support of INRAE.

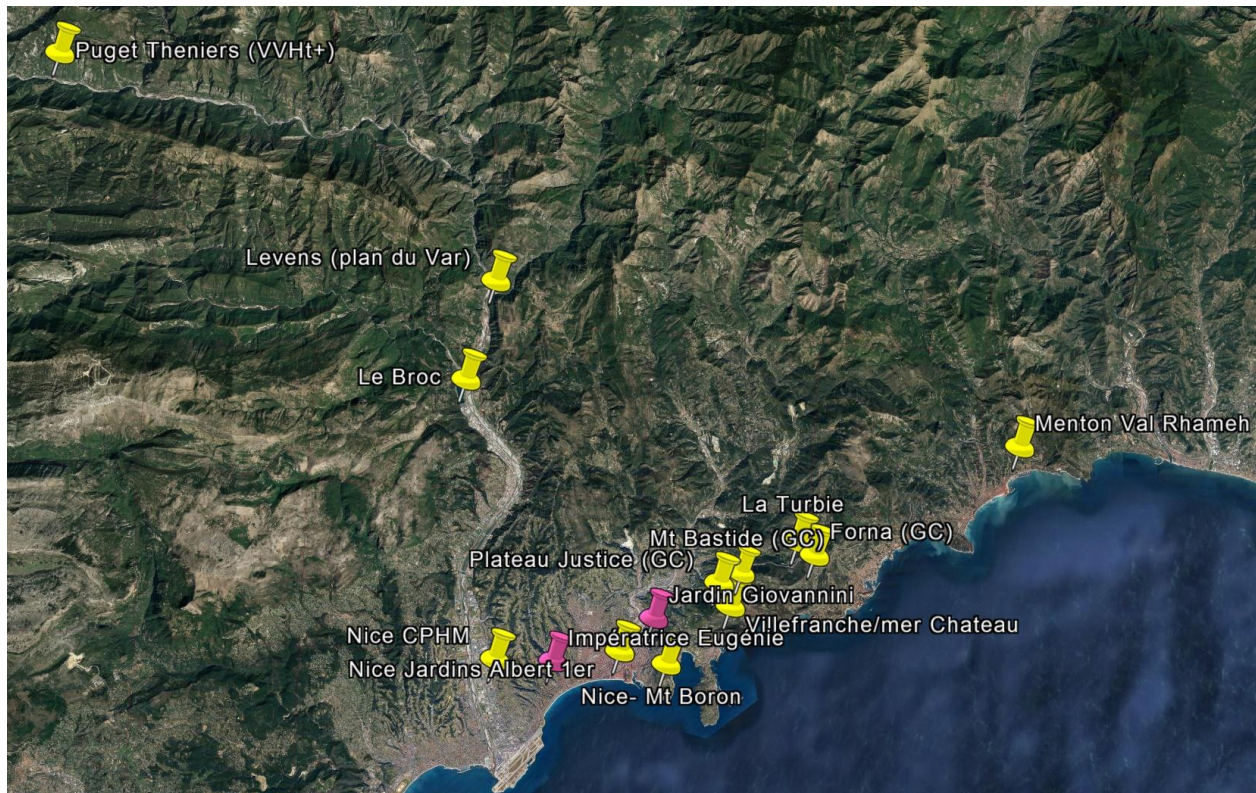
After a long round of discussions, INRAE finally obtained on 16 May 2021 from the Park Scientific Council the permission of installing one trap in the core of the Port Cros National Park, within the area of Porquerolles belonging to the Park. The trap was immediately installed within the fig tree collection of the botanical conservatory, which also hosts major collections of mulberry and carob trees. Two additional traps were previously deployed on 1 April in other parts of the Porquerolles island, which are not part of the National Park. INRAE also performed a survey on the Island in July to search for any symptoms of tree and shrub damage caused by *Xylosandrus*.



Map of the 2021 trapping network for early warning of expansion of *Xylosandrus* spp. and possible incursion to the National Park of Port-Cros. New trap in Porquerolles island in purple.

The network around the core sites was extended northwards in the Var Valley, with a site at Puget Theniers, to assess expansion towards the Park Mercantour. 11 sites were covered with 11 traps, and 3 sites were surveyed for damage.

Also in the seashore near Cannes and Sainte Marguerite, and in the areas around Antibes the network was enforced, in particular with traps at Mandelieu and Théoule/Mer.

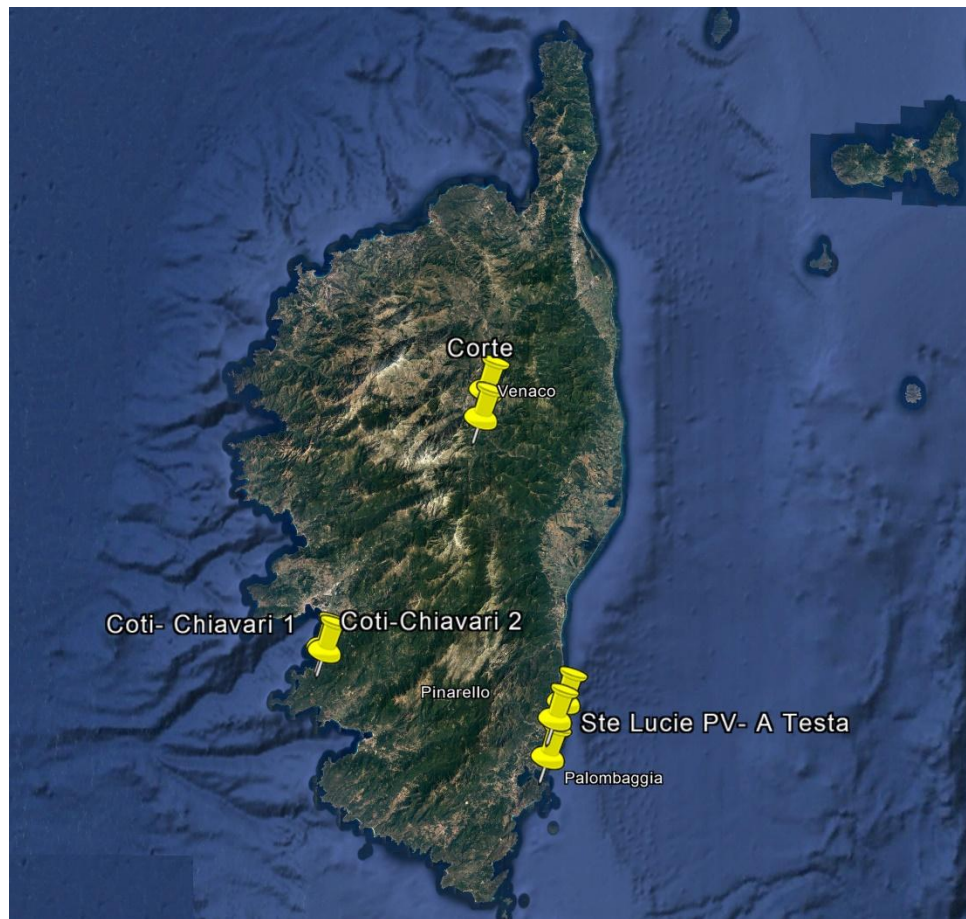


Trapping network 2021 in and around the core sites Corniches de la Riviera and Mont Boron: (in purple the new traps added in 2021); 11 sites, 11 traps and 3 sites surveyed for damage



Trapping network 2021 in and around the core sites Antibes and Ste Marguerite: 13 sites, 38 traps and 1 site surveyed for damage

In addition, following the discovery of *X. compactus* in Corsica in 2020 and the discussions at the meeting of the EU ALIEM project where Alain Roques presented SAMFIX, 13 traps were installed in Corsica on both sides of the island, including the core of the Parc naturel régional de Corse in order to survey a possible expansion on the Island.



Trapping network 2021 in Corsica

In case of captures of *Xylosandrus* on the continent and in Corsica in traps located elsewhere than in the areas where the beetles have already been recorded, all sensitive plant species were to be checked for damage symptoms within a radius of 100m. Discussions were undertaken with officers of the Forest Health Departement about pruning or removal of the infested plants.

In case *Xylosandrus* would be captured on the Park islands, park administration is suggested to check all sensitive plant species for damage symptoms within a radius of 100m of the trap with the help of INRAE. If the damage is limited, pruning can be recommended, but if it is generalized immediate removal of the entire plant is necessary to eradicate the beetles from the park.

Results

The 3 traps settled within the Port-Cros National Park did not reveal any captures of *Xylosandrus* for the moment, suggesting that the National Park is still free of beetle presence. However, a few *X. compactus* were trapped again on the close-by seashore, at Chateau Léoube- Bormes Les

Mimosas (ca 11 km of the islands on a birdwing) as well as at Manjastre (ca 21 km on a birdwing). In this last place large damage was also observed on plane trees.

At Chateau Léoube also 1 capture of *X. crassiusculus* was recorded.

Anyhow, no beetle was captured by any of the other traps deployed along this seashore, thus suggesting that the captures correspond to isolated populations within the Bormes les Mimosas area, maybe facilitated by a local recycling center at Manjastre.

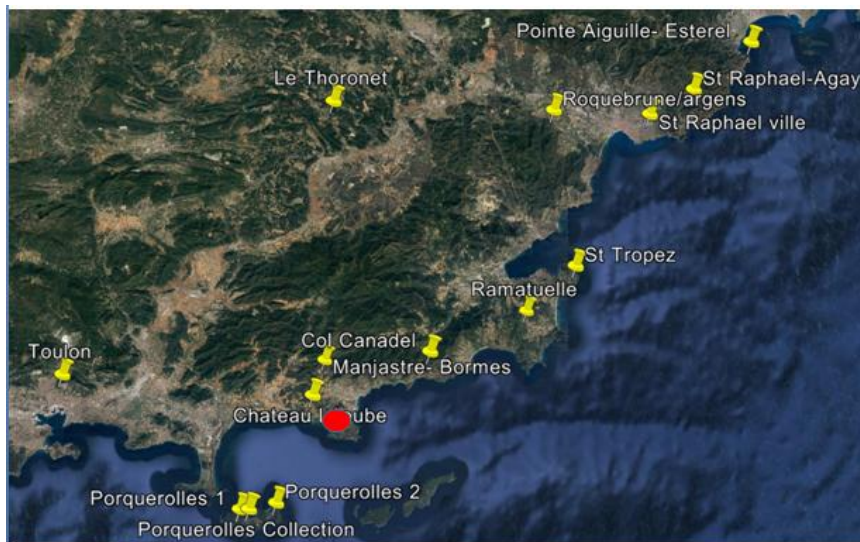
Instead, *X. germanus* was found, still in very low density, in 6 places along the seashore, but, again without any capture at the Island of Porquerolles in or close to the Port-Cros park.



• Captures at Manjastre and Château- Léoube

Agay	0
Château Léoube	2
Col Canadel	0
Le Thoronet	0
Manjastre	5
Porquerolles 1	0
Porquerolles 2	0
Porquerolles- Conservatoire	0
Roquebrune/Argens	0
Saint Tropez	0
St Raphaël	0
Toulon	0

X. compactus captures from the 2021 trapping network for early warning of expansion of *Xylosandrus* spp. and possible incursion to the National Park of Port-Cros



- Only 1 capture at Château-Léoubé

Agay	0
Château Léoubé	1
Col Canadel	0
Le Thoronet	0
Manjastre	0
Porquerolles 1	0
Porquerolles 2	0
Porquerolles- Conservatoire	0
Roquebrune/Argens	0
Saint Tropez	0
St Raphaël	0
Toulon	0

X. crassiusculus captures from the 2021 trapping network for early warning of expansion of *Xylosandrus* spp. and possible incursion to the National Park of Port-Cros



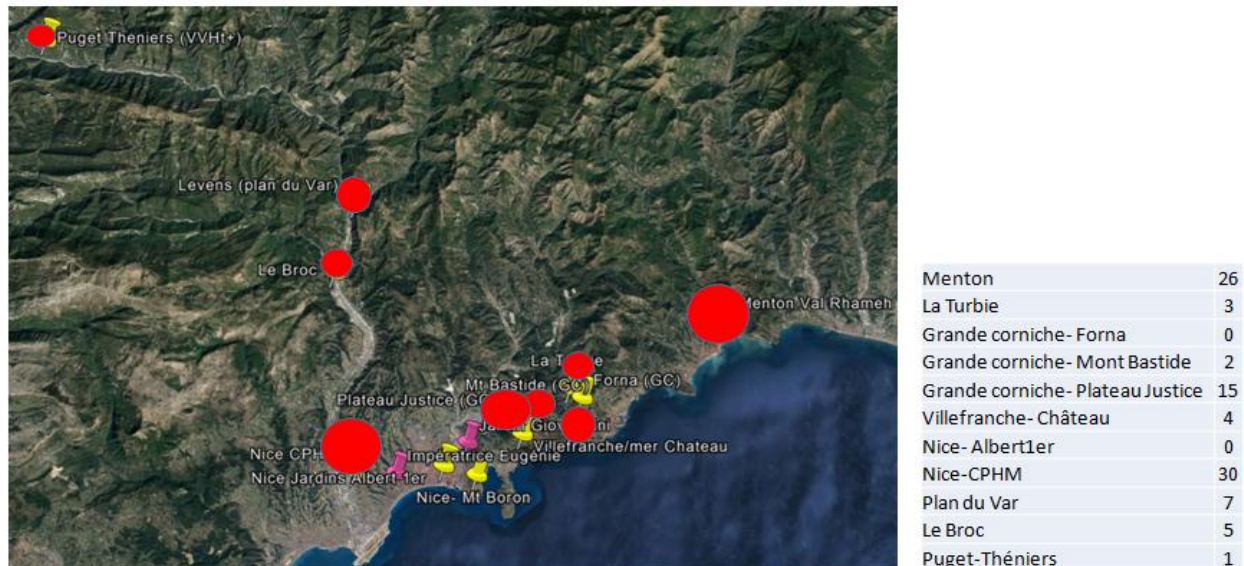
- 6 sites with low captures

Agay	8
Château Léoubé	1
Col Canadel	1
Le Thoronet	0
Manjastre	0
Porquerolles 1	0
Porquerolles 2	0
Porquerolles- Conservatoire	0
Roquebrune/Argens	2
Saint Tropez	8
St Raphaël	0
Toulon	1

X. germanus captures from the 2021 trapping network for early warning of expansion of *Xylosandrus* spp. and possible incursion to the National Park of Port-Cros

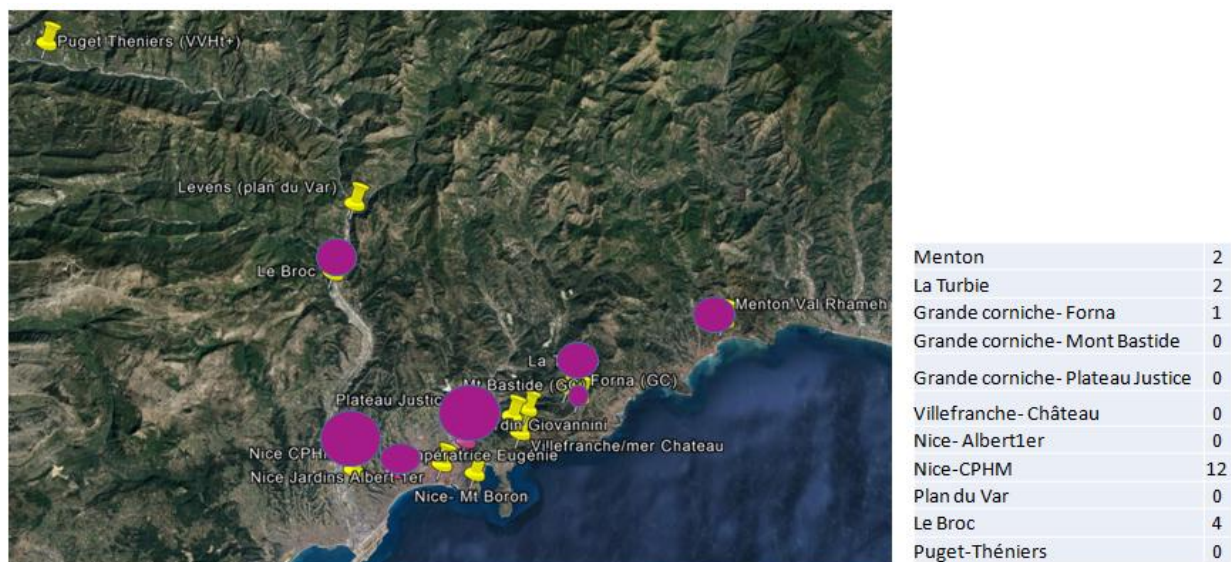
Around the Corniches de la Riviera and up to the high Var Valley, a widespread presence of *Xylosandrus crassiusculus* was ascertained, although density in the Corniches de la Riviera itself was low. Whereas *X. crassiusculus* expanded northwards in the Var Valley until Levens and Puget Théniers, *X. compactus* was so far not found above Le Broc. Nurseries (at Nice) and botanical

gardens (at Menton) seem to be important origins of *X. crassiusculus* expansion. Presence of *X. compactus* was very limited in this area, although density is high in Nice nursery (CPHM) where visual inspections revealed large attacks, even on olive trees. There are strong reasons to presume that the Nice nursery may be the source of both species of *Xylosandrus* dispersion in the region.



2021 *Xylosandrus crassiusculus* captures in and around Corniches de la Riviera and in Var Valley

28



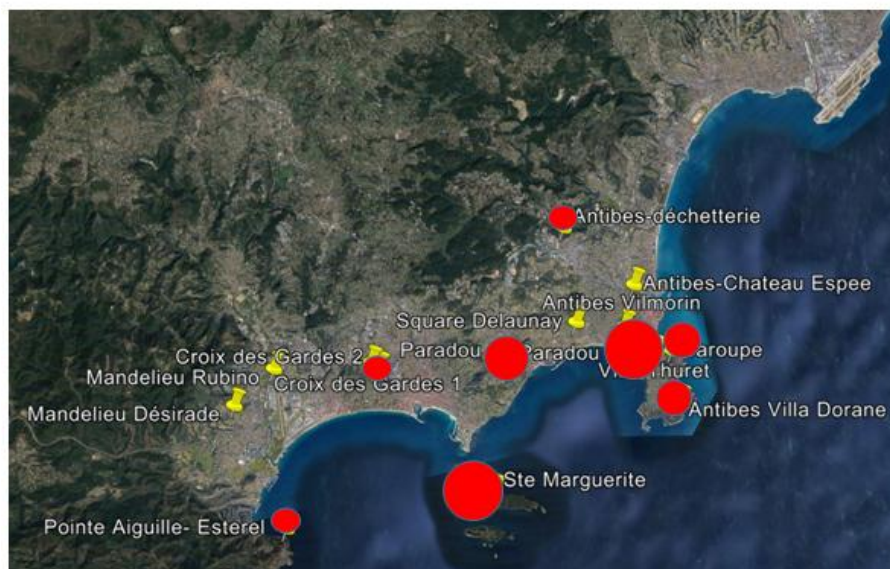
2021 *Xylosandrus compactus* captures in and around Corniches de la Riviera and in Var Valley

Differently from *X. crassiusculus* and *X. compactus*, *X. germanus* was not trapped in the CPHM nursery in Nice, but a large presence was ascertained in the Var Valley.

Although the core sites Antibes Villa Thuret and Ile Ste Marguerite continue to show a notable infestation of *Xylosandrus crassiusculus*, its presence in outer areas of Antibes, in Bois de la Garoupe and in Cannes was limited to isolated spots where only a few beetles were captured. More westwards, in Mandelieu, no captures occurred, but in Théoule sur Mer 2 were found, which is in between Cannes and Saint-Raphael, confirming a further spread along the coast.

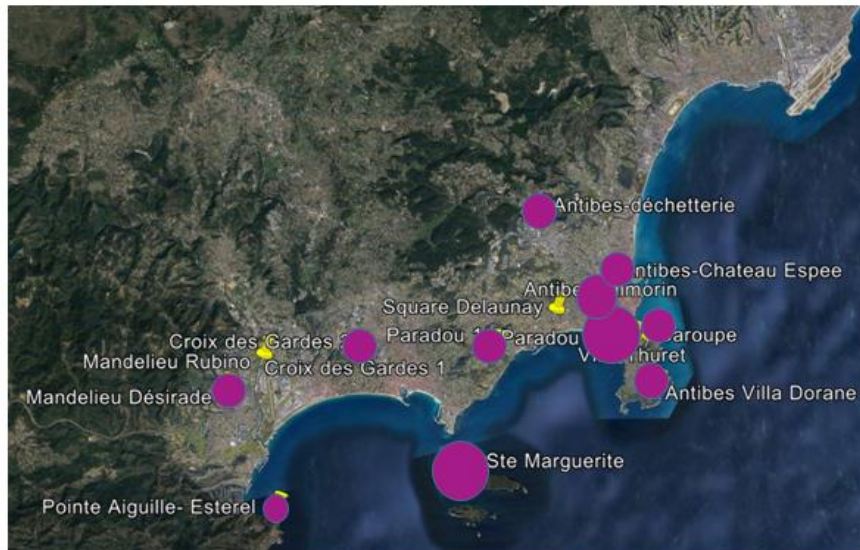
As far as *X. compactus* concern, the presence results generalized in the entire area, but at low density. The presence in the Antibes woody recycling center gives rise to the suspicion that this might be a hub for transportation.

Very large captures occurred of *Xylosandrus germanus* in Ile Ste Marguerite and nearly all its surroundings. Only a few of them were found in the recycling center.



Antibes Déchetterie	1
Antibes Delaunay	0
Antibes Villa Daurane	5
Antibes Vilmorin	0
Antibe Villa Thuret	51
Antibes Garoupe	6
Croix des Gardes	3
Parc du Paradou	10
Ste Marguerite	52
Mandelieu- Désirade	0
Mandelieu- Pép Rubino	0
Estérel	2

2021 *Xylosandrus crassiusculus* captures in Antibes and towards the west until Estérel (Théoule sur Mer)



• Captures on most sites

Antibes Déchetterie	4
Antibes Delaunay	0
Antibes Villa Daurane	2
Antibes Vilmorin	5
Antibes Villa Thuret	21
Antibes Garoupe	7
Croix des Gardes	4
Parc du Paradou	3
Ste Marguerite	20
Mandelieu- Désirade	2
Mandelieu- Pép Rubino	0
Estérel	1

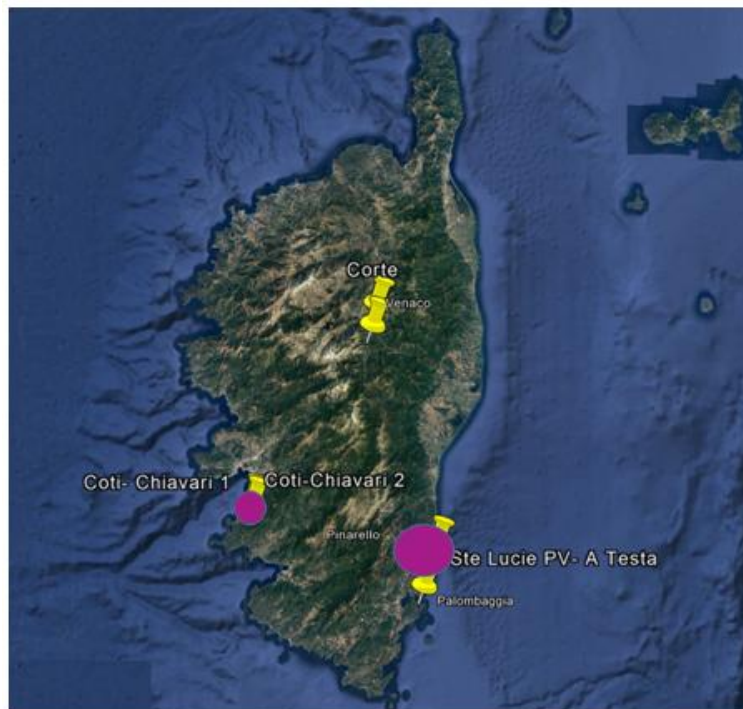
2021 *Xylosandrus compactus* captures in Antibes and towards the west until Estérel (Théoule sur Mer)

The trapping network revealed, unexpectedly, that *Amasa nr. truncata* seems present in all areas with eucalyptus trees along the French Riviera. As this species had never been noticed prior to the start of SAMFIX, it is so far unknown if it was already present but simply not discovered, or if it is in expansion.

30

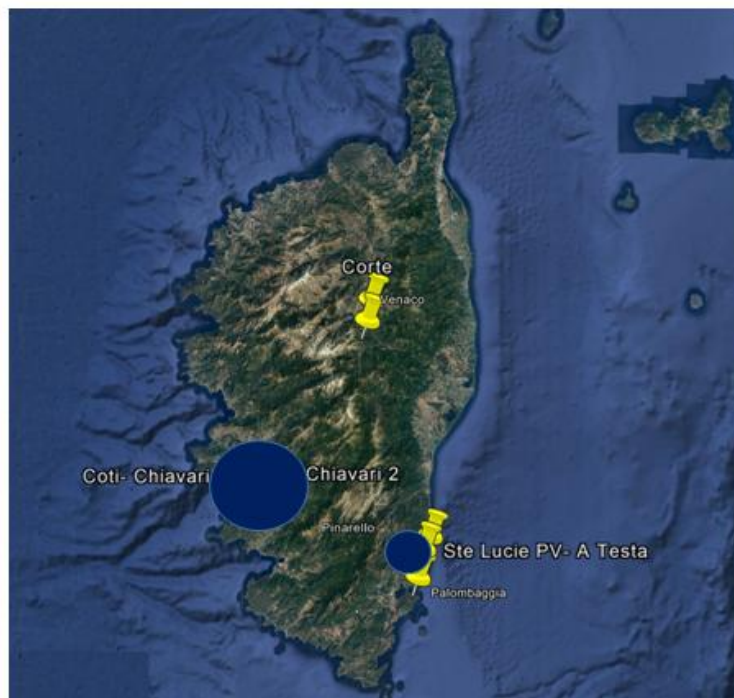
The trappings deployed in Corsica confirmed the presence of *X. compactus*, with abundant captures and noticeable damage in a private property in Pinarello, a site on the eastern coast near Porto Vecchio. Damage was found on shoots of *Arbutus*, Carob tree, Cycas and Banksia but also on trunks of *Phyllirea* and Carob trees with associated fungi. However, no damage was observed in the close environment of the infested property and no insects were trapped in the nearby site of Sainte Lucie de Porto Vecchio, suggesting the beetle is not widespread in the area, yet. Only one insect was trapped on the western coast (Coti-Chiavari), confirming its presence but not revealing any expansion. Both sites are surrounded by Natura2000 protected areas and located close (<15km) to the Regional Natural Park of Corsica, thus may represent important threats to the natural heritage of the island.

Also *X. germanus* was detected, with apparently inverse population densities.



Corte	0
Coti-Chiavari	1
Pinarello	12
Porto Vecchio	0
Ste Lucie PV	0
Venaco	0

2021 *Xylosandrus compactus* captures in Corsica



Corte	0
Coti-Chiavari	105
Pinarello	3
Porto Vecchio	0
Venaco	0

2021 *Xylosandrus germanus* captures in Corsica

3.4 Year 2022

A meeting with the Port-Cros National Park managers and scientific council has taken place on March 28th to discuss the results of 2021 activities and plan the 2022 experiments on which the Park Scientific Council agreed.

Following the ascertained presence of *X. compactus* along the close-by seashore, future monitoring of the situation in the Port-Cros National Park is pivotal and was happily agreed.

During the meeting, the Port-Cros park has agreed to manage 3 traps in 2022 at different, new locations on the Porquerolles island within the borders of the Natural Park. These traps will be baited with lures similar as those in 2021. The lures have been provided by INRAE on March 31 and the trappings were installed on 11th of April. They cover the botanical collection of fig and carob trees; a planted row of carob trees where end of March potential *Xylosandrus* damage has been noticed; and an old carob tree in the wild.

Unfortunately, only 5 traps were deployed on the close seashore since we got only a few positive responses for continuing the surveys there. However, these traps concern the sites where *Xylosandrus* spp. have previously been observed (Chateau-Léoube, St Tropez, St Raphaël). They took off since early April.

In case *Xylosandrus* would be captured on the Park islands, park administration is suggested to check all sensitive plant species for damage symptoms within a radius of 100m of the trap with the help of INRAE. If the damage is limited, pruning can be recommended, but if it is generalized immediate removal of the entire plant is necessary to eradicate the beetles from the park.

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Also in and around the core sites surveillance and trapping networks were confirmed and implemented in 2022, according to the same protocol as in 2021: one black multifunnel trap baited with the 4 attractive compounds (Ethanol/ [-] α-pinene/ + quercivorol/ + α-copaene) were deployed per site, instructions on the verification of damage symptoms remained the same, as well as those on pruning and removal. A total of 19 sites are being surveyed, amongst which:

- 5 traps within the Corniches de la Riviera, and 5 in its surroundings
- 3 traps will be placed in Antibes core area and 2 in the surroundings (besides 2 X-traps at Villa Thuret)
- 2 traps will be installed at Ile Sainte Marguerite with 2 additional traps on the close seashore (Cannes).

The network was installed by half April and remains in place until October, with a collection of trapped beetles every 3 weeks and change of lures every 6 weeks.

In Corsica, the managers of the Environmental Council (OEC) appreciated the results achieved with the SAMFIX activities, that were presented to them during a meeting of the INTERREG ALIEM project. The recent launch of the ALIEM II project, again funded by INTERREG, will allow to maintain the 6 sites surveyed in at least 2022.

3.5 Evaluation

Nurseries, botanical gardens and urban parks confirm to be important pathways for introduction and spread of *Xylosandrus* spp. It is likely that a major pathway of dissemination is the planting along the roads and in parks of infested plants coming from the administrative nursery in Nice. To prevent further dispersion, it is thus of utmost importance to strengthen and preserve awareness on the species amongst their managers, in order to provide for a careful survey of the symptoms on apparently healthy plants prior to shipment and planting. Also, because here pruning and removal of limited numbers of infested plants are much more feasible than if to be applied in large-scale protected areas.

Wood recycling centers are identified as another major stakeholder in the prevention of *Xylosandrus* expansion, and may represent an additional hub for transportation.

The Parc National de Port-Cros, yet indemn of damage, has to be continuously surveyed, especially regarding possible transportation of infested plants. Similarly, it is important and still possible to prevent the expansion of the infested spot in southeastern Corsica towards natural areas.

The defined lure blend appears to be very effective as well for the capture of *Xylosandrus germanus*, which was found unpurposely but included in the analyses.

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4. Findings in the replication sites in Spain

In Spain, *Xylosandrus crassiusculus* was detected for the first time in 2016 in carob trees at El Pla de les Clotxes, Benifalló in the Spanish Valencia region, located at 3 km from the Municipal Natural Park El Tello, that hosts two public forests, Monte de Picassent (v3044) and Monte Aledua (V036), the SAC Sima del Águila (ES5234005) and the Plant Micro-Reserve Lloma del Tramussar. *Xylosandrus compactus* was never detected in Spain prior to the start of the SAMFIX project. The project's core activities were planned therefore in and around the El Tello park, while for replication 2 protected areas were targeted:

- Sierras de Martés y el Ave - SCI ES5233011 and SPA ES0000212
- Muela de Cortes y el Caroché - SCI ES5233040 and SPA ES0000212

Sierras de Martés y el Ave distances 13 km from the *X. crassiusculus* outbreak and Muela de Cortes y el Carroche 20 km. Both includes the habitats *Quercus ilex* and *Quercus rotundifolia* forests (Annex I – 9340), Thermo-Mediterranean and pre-desert scrub (Annex I – 5330) and *Arborescent matorral* (sclerophyllous scrub) with *Juniperus* spp. (Annex I - 5210), which are amongst the ecosystems that result at risk from the data gathered in previous European detection areas. The Generalitat Valenciana considered thus of utmost importance that early warning and prevention protocols would be implemented in these two parks.

During the project, other replication activities were developed upon reported suspected detections that reached the project team, especially after several public events.

Amongst those, the most relevant concerned:

- several private gardens where *X. crassiusculus*, *X. compactus* and *X. germanus* were detected along the coast of Valencia Region (Náquera) and Catalonia (Banyoles, Vidreres, Vila-Seca and Platja D'Aro);
- a private garden in El Toro, Island of Majorca, bordering Cap de cala Figuera – SPA & SCI ES0000074, where the first detection in Spain of *X. compactus* occurred.

4.1 Year 2019

Differently from the areas in Italy and France, no anticipation to 2019 was decided for Spanish replications, as no evidence of quick expansion from the original detection site had been perceived. The SAMFIX team used this year to raise awareness amongst managers, phytosanitary inspectors and environmental agents of the Generalitat Valenciana; amongst nurseries, paying individual visits to several of them located in Picassent, close to El Tello; to technicians of the local authorities of the municipalities in the El Tello area, Picassent and Benifaió; and to forestry and agricultural technicians of the Consellería of Alicante in Plant Health.

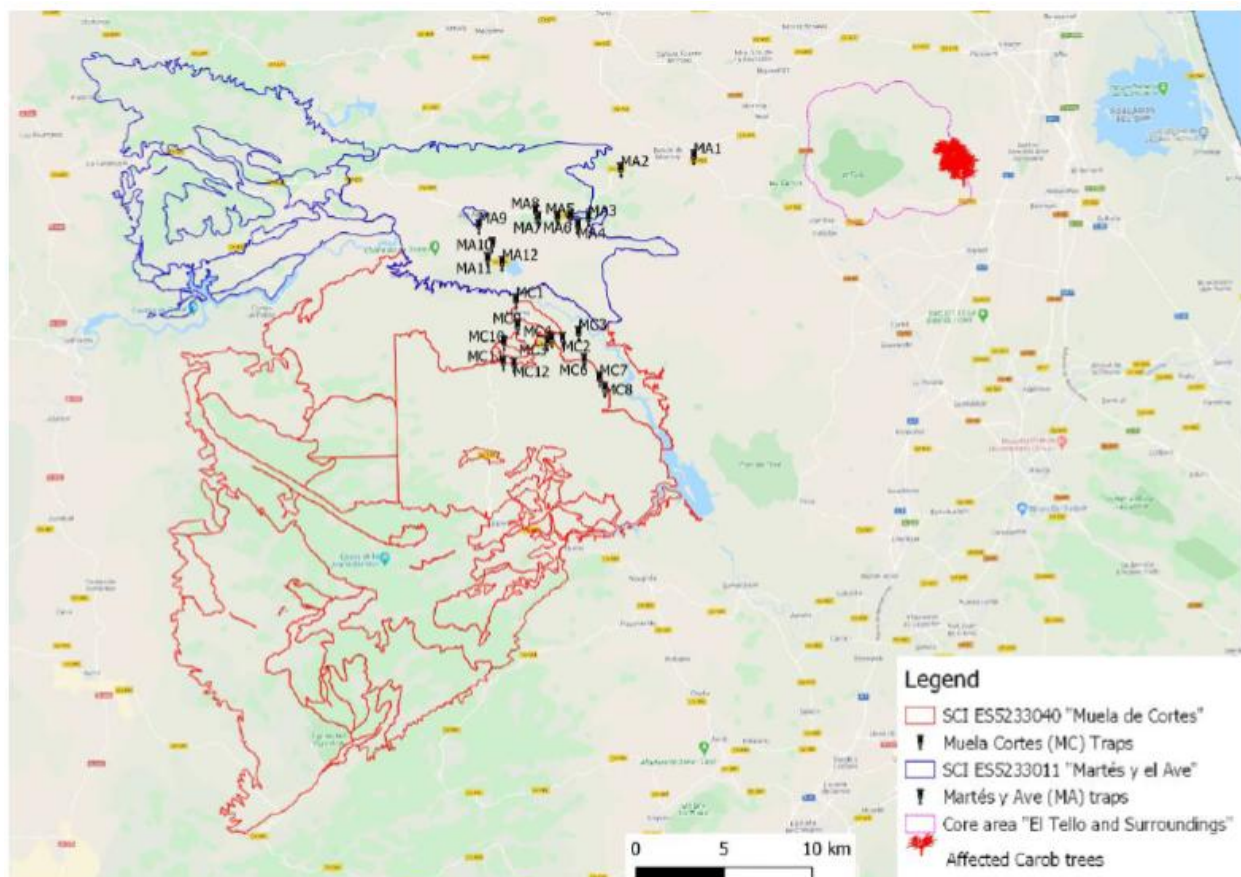
4.2 Year 2020

UA impaired a training to managers of “Muela de Cortes y El Carroche” in Requena (Valencia) on the 10th of March 2020. A similar activity to Sierra de Martés y el Ave managers was programmed for the 12th of March in Xàtiva (Valencia) but the meeting had to be suspended due to COVID-19 restrictions.

Trapping installation in both replications areas was planned to be performed within end of March 2020, after both training activities in Requena and Xàtiva, but the quarantine imposed in Spain on 15/03/2020 blocked these activities.

On the 9th of July, 12 traps could be installed in “Muela de Cortes y el Carroche”, according to the protocol defined and shared just prior to the lock down. Meanwhile, the protocol was defined with the “Sierras del Martés y del Ave” management and on the 23rd of July also here 12 traps were installed. Both areas feature a very rough terrain and were affected by a great wildfire in 1994 that devastated massively the afforested pine forests. At present, the ancient broadleaf tree forests (*Quercus rotundifolia*) are in regeneration.

Traps were installed at the nearest distance from the outbreak site in El Pla de les Clotxes, in accessible areas (close to roads) and where susceptible trees were found. The distance between traps was about 500 m and they were baited with a-pinene and ethanol. Traps were monitored biweekly until mid-November.



2020 Trap locations in Muela de Cortes y el Carroche and Sierras del Martés y del Ave

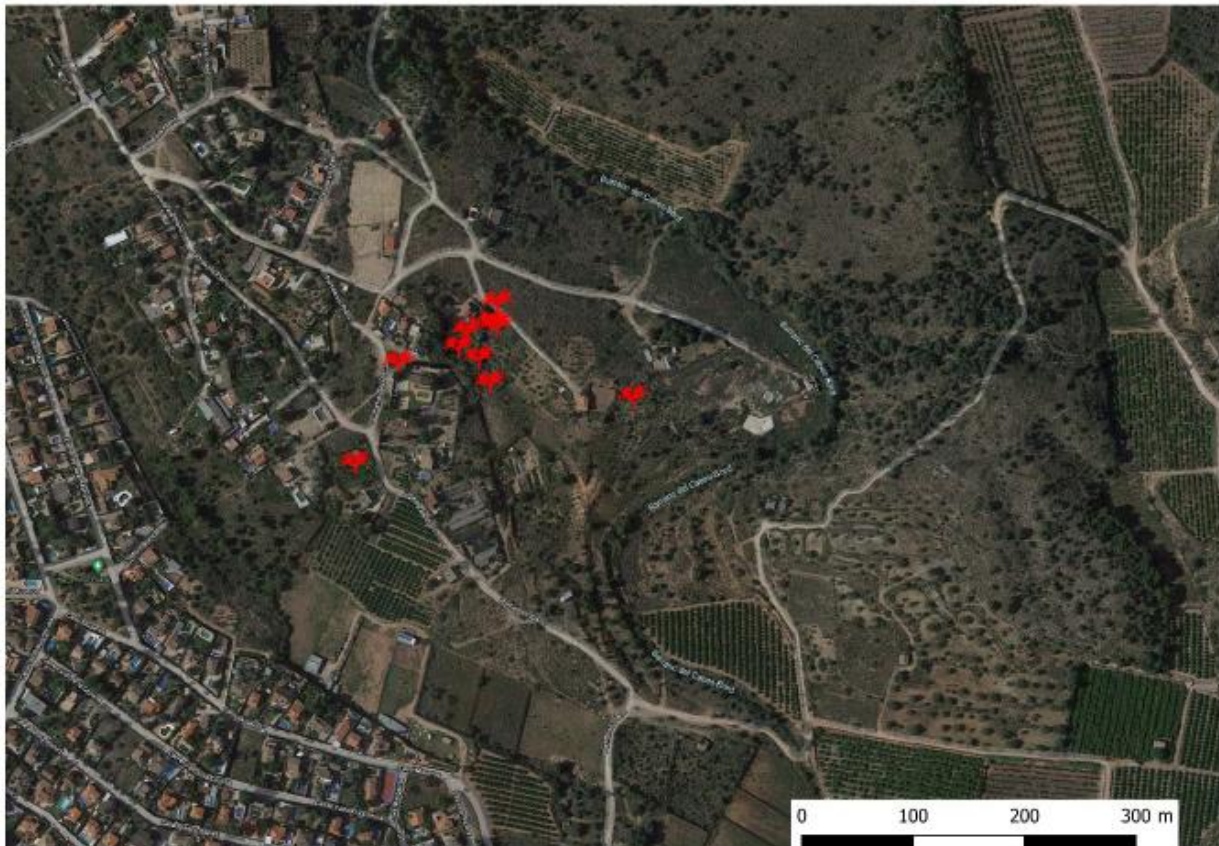


2020 Some traps in Muela de Cortes y el Caroché



2020 Some traps in Sierras del Martés y del Ave

A new outbreak of *X. crassiusculus* was found in Náquera (Valencia), at 35 km of the first outbreak near El Tello, affecting a dozen ornamental carob trees. It was referred to the team by a participant of the project symposium organised in Valencia, that had seen a picture of these trees in a private Facebook group of friends of trees.



2020 *Xylosandrus crassiusculus* detected in Náquera (Valencia)

Like at El Tello, all attacks occurred on trees located in gardens or in neighbourhoods of houses of a residential area adjacent to a Natural Area, the Parc Natural de la Serra Calderona. This new area was visited at 23 July, inspecting all carob trees and other potential hosts of the area, and installing 2 traps baited with α -pinene, ethanol, quercivorol and α -copaene. Both traps have been located into the private garden of the person that alerted us of *Xylosandrus* presence. Captures were collected biweekly by the house owner. The site was visited for a second time at 10th of September, refilling the traps by new mass trapping lures and inspecting the area for new attacks.

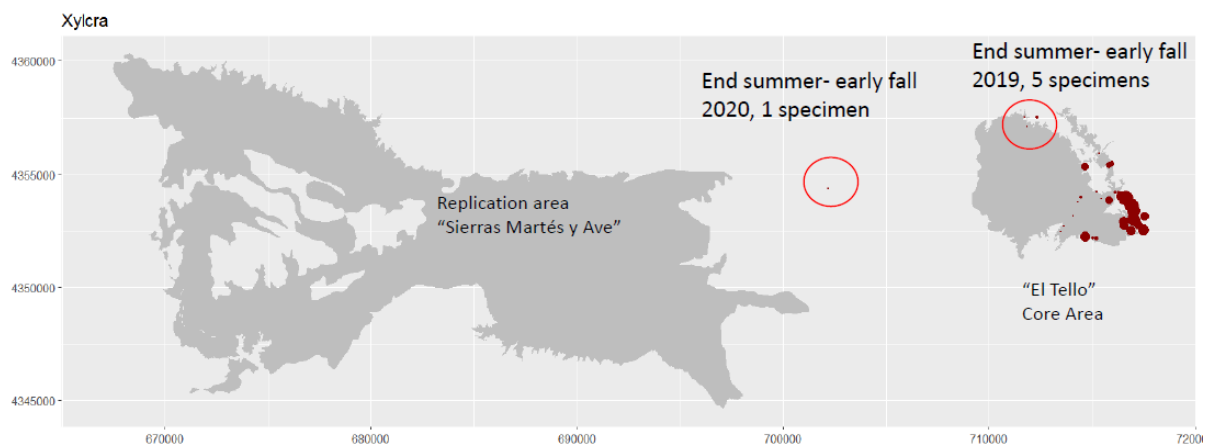
In June 2019, unusual symptoms such as necrosis of the leaves and withered and dried twigs were observed in a carob tree (*Ceratonia siliqua* L.) in a private garden in "El Toro" Residential Area in the Municipality of Calvià, Majorca, Balearic Islands, bordering the protected area of Cap de cala Figuera (SPA & SCI ES0000074). The area features Aleppo pines (*Pinus halepensis* Miller) and mixed vegetation of Mediterranean garriga, with presence of *Pistacia lentiscus* L., *Rosmarinus officinalis* L., *Erica multiflora* L. and *Olea europaea* var. *sylvestris* L. Multiple small holes were located in both small and large branches and in twigs. During August, a sap

exudation from holes was observed by the University of the Balearic Islands. In October 2019, its Laboratory of Zoology received some insect samples, and the collected specimens were then identified as *Xylosandrus compactus* (Eichhoff). The SAMFIX spanish team provided support through the process of collection and identification. This represented the first infestation in Spain of this species. The observed damage in large branches (as opposed to small branches and twigs) of carob trees concerns a rare behavior, previously reported by Gugliuzzo *et al.* (2019) for first time in Sicily (Italy). The Spanish SAMFIX team got involved in the analyses and the definition of management protocols. In 2020, it supported three actions in the area of the outbreak, implemented in collaboration with the Plant Health authorities of the Balearic Islands Government and the University of the Balearic Islands:

- (1) an intensive survey campaign;
- (2) a trapping network using 20 crossvane traps baited with alpha-pinene and ethanol;
- (3) an information campaigns to stakeholders and citizens.

Results

No attacked plants were detected and no *Xylosandrus* spp. have been captured in both the selected protected areas during 2020. However, one specimen of *X. crassiusculus* was collected in Trap 1 of the trapping network of Sierras del Martés y Ave, although outside of the protected area boundaries. The trap is located in an orange crop area with carob trees. Possibly, this specimen has been collected during its dispersive flight, because the far captures have been always reported in early fall.



Sole capture of *Xylosandrus crassiusculus* in 2020 between El Tello and Sierras del Martés y Ave, compared to expansion found in El Tello in 2019

Instead, in Náquera we have found the first case of death of a carob tree caused by *X. crassiusculus* attacks. The big and old carob tree resulted massively attacked in July at its lower

trunk, and died early September. However, a very quick and vigorous resprouting response was observed on the ground.



Massive attack in lower trunk in July 2020.



Left: dead tree early September 2020 – right: resprouting in early September 2020

More than a thousand *X. crassiusculus* specimens have been collected in both traps from 23 July to 6 September.

Due to this high amount, the site was considered very suitable to experiment the push & pull experiment in 2021.

As far as the Island of Maiorca concerns, only 6 specimens were captured in traps and no damage was detected to plants.

4.3 Year 2021

Traps were not removed but refilled from April onwards in both replication sites of “Muela de Cortes y el Carroche” and “Sierra del Martés y el Ave”. They were monitored biweekly until mid-November.

Responsibles and environmental agents of the replication site “Sierra del Martés y el Ave” attended the 2nd SAMFIX Spanish Conference, delivered on line on 29 April 2021. Herewith the missed training course in 2020, due to COVID-19 restrictions, could be covered up, providing updated information on the protocols.

Due to the assured presence of insects in Náquera, trials with X-trap and with push-and-pull were performed not only in El Tello, but also here. 2 X-traps were placed, in private areas so protected by fences. One of the two had issues, and was removed on 30 november for repair.

One push-and-pull square was installed in April 2021.



X-trap at Náquera

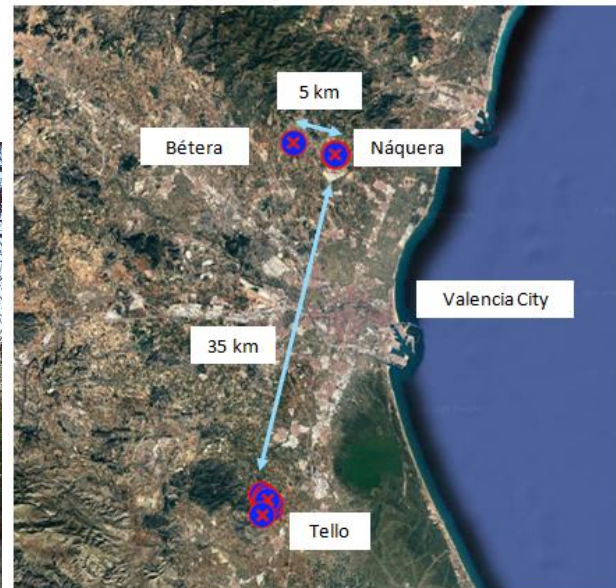


Pink: push traps with verbenone; green: pull traps with the 4 components lure; yellow: control traps

A new outbreak of *X. crassiusculus* was found in Bétera, at about 5 km of the outbreak in Náquera. Also here a dead carob tree was found, with similar characteristics as the one in Náquera in 2020. No damage was detected on surrounding nor other sampled insects.



Dead carob tree in Bétera 2021



Locations of ascertained outbreaks in Valencia Region until 2021

In El Toro at the Island of Majorca, the trapping network installed in 2020 was maintained in 2021 and the SAMFIX team continued providing support for species identification.

The very first *Xylosandrus compactus* infestation on the peninsula was detected in Banyoles (Girona) in July 2020. In 2021 responsables of Generalitat of Catalonia carried out an inspection campaign, detecting *X. compactus* in Vidreres, Vila-seca and Platja D'Aro.

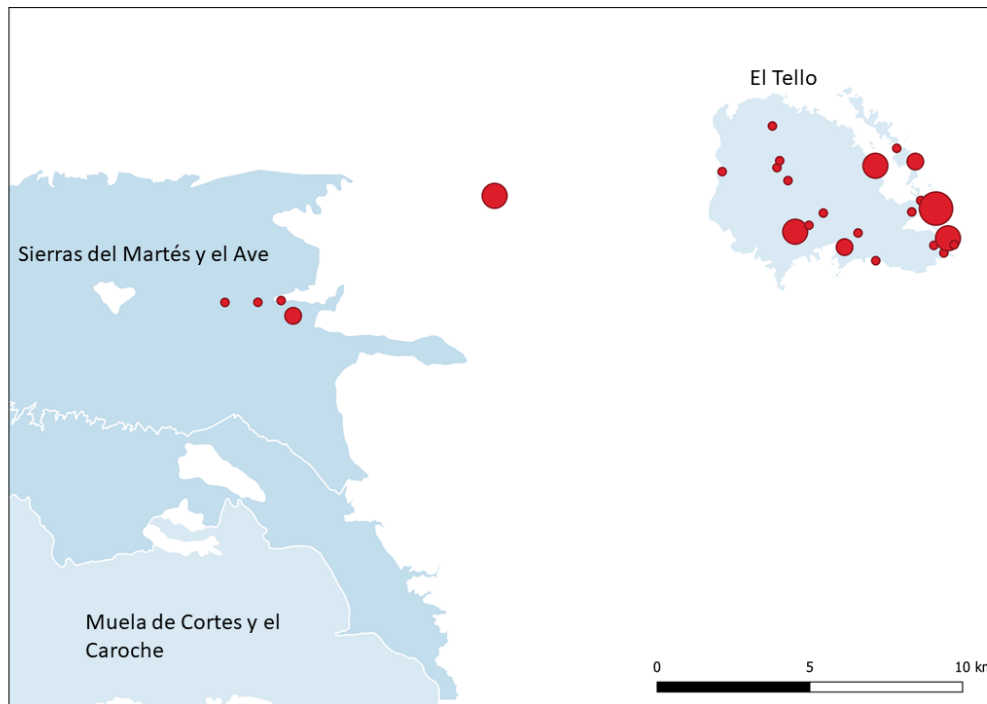
Results

In Sierras del Martés y del Ave the presence of *Xylosandrus crassiusculus* was confirmed in 4 traps, collecting 11 specimens at late summer and early autumn of 2021, possibly due to the dispersive flight of the species. No damage was detected to trees.

In Muela de Cortes y el Carroche no detections occurred, neither by the trapping network nor by visual observations.

In El Toro at the Island of Majorca no damage to plants has been detected, although 46 specimens of *X. compactus* were collected in the sampling network in 2021, also in late summer and early autumn, possibly related with the dispersive flight from a not detected sources.

In Catalonia (Banyoles, Vidreres, Vila-Seca, Platja D'Aro), no trapping networks were installed in 2021, so no data of captures in traps are available. Instead, authorities reported a wide detection of *X. compactus* in private gardens, natural vegetation and, for first time in Spain, damage to carob and hazelnut crops in Vila-seca.



Locations of captures of *X. crassiusculus* in the replication site "Sierras del Martés y el Ave" in 2021.

4.4 Year 2022

The management of the parks Sierras del Martés y del Ave and Muela de Cortes y el Caroche agreed to keep the trapping network in place until the end of 2022. Surveillance of attacked trees will also continue until end of 2022.

In Náquera, the push and pull experiment and the X-traps were reinstalled in the first week of April and remain in place until the end of 2022.

UA also continues to support and advice private owners on surveillance and pruning.

UA and the Forest Health Service of the Generalitat Valenciana, responsible for the trapping networks "Muffet" and "RAT", have agreed to include the early detection of *Xylosandrus* spp. in these yet existing networks for forest health monitoring during 2022 and thus mainstream the surveillance for the forthcoming years, as long as the need is felt, within their ordinary forest health monitoring services.

Responsibles of Balearic Island continued the trapping and surveillance in El Toro at the Island of Majorca. No data were reported yet.

In Banyoles, Vidreres and Platja D'Aro responsables continue the surveillance campaign. In Vilaseca, a push and pull experiment was installed (20/05/2022) in a private carob tree crop, supported and advised by the Spanish SAMFIX team. The periodical surveys of the experience were carried out by the Plant Health staff of Generalitat of Catalonia. No results were reported yet.

4.5 Evaluation

Xylosandrus crassiusculus has so far mainly infested carob trees in Spain. No recurrent infestation of the same tree is ever found, which confirms the hypothesis that after one colonisation the beetles search for a new host. Trees generally regenerate well, not showing evidences of previous attacks after three years, except from the trees that died from the infestation.

An Early Warning Network using baited traps should be permanently active in forest areas and especially in urban and landscaped areas, aimed to detect either existing species or introduced species (with special attention to invasive alien species). The Muffet and RAT networks of the Generalitat Valenciana provide for such, but it is also of utmost importance for insular territories, like the Balearic Islands.

However, an Early Warning Network is not enough to assure effective detection. Dissemination actions of SAMFIX in Spain helped the detections of *Xylosandrus* spp. in Catalonia and Balearic Islands. The promotion of citizen science by informative campaigns with new technologies support is an excellent complementary tool for prevention of non-native species introduction, as we learned from the records received from people after they had been informed.

5. Conclusions

Replication efforts were very successful: in Italy, additional territories adhered to the implementation of early warning protocols (Ventotene Island, Monumento Oasi di Pantanello and Giardini di Ninfa of Fondazione Caetani, Presidential estate of Castel Porziano); in France protocols could be extended to the Corniches de la Riviera park where indeed *X. crassiusculus* was detected, and a collaboration on Porquerolles could be obtained. Found records led to the involvement of the regional parks of Paradou and Croix des Gardes. In Spain, early warning protocols were implemented in both parks foreseen in the project, while activities were also extended to Náquera, at 43 km from the outbreak close to El Tello, where *X. crassiusculus* was detected following a report received from a participant to the national conference. Also, support was given to the Island of Mallorca where the first *X. compactus* infestation in Spain was detected in 2019, to Corsica, and to Plant Health officers engaged in several localities in Catalonia.

The findings show that *Xylosandrus* spp. are spreading around the Tyrrhenian coasts, but their spread is generally limited to isolated spots and usually without significant damage.

Overall, it can be concluded that early warning networks should be maintained in urban and landscaped areas bordering natural parks, including in nurseries and biowaste treatment locations, providing pathways for introduction in protected areas, while monitoring networks in areas yet affected together with visual inspection is needed to prevent expansion and damage. Accurate pruning and removal should be evaluated and applied where possible and infestation is limited.

Moreover, broad awareness raising and knowledge transfer resulted pivotal for detection and subsequent protective interventions.