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Passive Surveillance as a Key Tool to African Swine Fever Eradications in Wild Boar: A Standardized Protocol to Find the Carcasses in Mediterranean Area

Elisabetta Coradduzza [1,¥], Federica Loi [2,*,¥], Francesca Porcu [2,¥], Daniela Mandas [2], Fabio Secci [3], Marco Efisio Pisanu [4], Cinzia Pasini [2], Carlo Zuddas [2], Marcella Cherchi [5], Daniele Denurra [5], Ennio Bandino [6], Antonio Pintore [5], Vittorio Guberti [7] and Stefano Cappai [2]

[1] Istituto Zooprofilattico Sperimentale della Sardegna, 07100 Sassari, Italy; elisabetta.coradduzza@izs-sardegna.it (E.C.)
[2] Istituto Zooprofilattico Sperimentale della Sardegna, Osservatorio Epidemiologico Veterinario regionale, 09125 Cagliari, Italy; federica.loi@izs-sardegna.it (F.L.); francescaporcu12@gmail.com (F.P.); daniela.mandas@izs-sardegna.it (D.M.); cinzia.pasini@izs-sardegna.it (C.P.); carlo.zuddas@izs-sardegna.it (C.Z.); stefano.cappai@izs-sardegna.it (S.C.).
[3] Local Sanitary Agency of Sulcis Iglesiente, 09013 Sulcis Iglesiente, Italy; fabiosecci@aslsulcis.it (F.S.)
[4] Associazione CPT – Caccia, pesca e tradizioni Sardegna, 09094 Marrubiu, Italy; marcoefisiopisanu@gmail.com (M.E.P.)
[5] Department of Wildlife, Istituto Zooprofilattico Sperimentale della Sardegna, 07100 Sassari, Italy; marcella.cherchi@izs-sardegna.it (M.C.); danieledenur@virgilio.it (D.D.); antonio.pintore@izs-sardegna.it (A.P.)
[6] Istituto Zooprofilattico Sperimentale della Sardegna, Nuoro, Italy; ennio.bandino@izs-sardegna.it (E.B.)
[7] Institute for Environmental Protection and Research (ISPRA), 00144 Roma, Italy; vittorio.guberti@isprambiente.it (V.G.)
* Correspondence: Correspondence: Federica Loi, federica.loi@izs-sardegna.it
¥ all these authors share first authorship.

Abstract: African Swine Fever (ASF) is one of the most important and serious contagious haemorrhagic viral disease of domestic pigs and wild boar, associated to high mortality rates and great sanitary and socioeconomic impact on international trade in animal and swine products. The early detection of the disease is often hampered by an inadequate surveillance. Among the surveillance strategies, passive surveillance of wild boar was considered the most effective for controlling African swine fever virus (ASFV). Otherwise, the design of a sufficiently sensitive ASF surveillance system requires a solid understanding of the epidemiology related to the local eco-social context, especially in absence of virus detection. Even if the number of carcasses needed to demonstrate the ASF eradication has been established, the scientific context is poor of details on protocols applied to active search the wild boar carcasses. The aim of the study was to describe the strategy applied for passive surveillance implemented in Sardinia, providing detailed information on the protocol applied in the field in term of number of people, dogs, time and space used to active search carcasses in the Mediterranean area. Using a specific tool developed to record, trace, and share on field data (GAIA Observer App), a total of 33 activities for active search the wild boar carcasses were organized during 2021-2023. Most of these activities were planned to find carcasses previously reported by hunters. A total of 24 carcasses were found, only 2 carcasses were not previously reported. The final protocol applied involved 4 people, with an average speed of 1.5 km/hour. When the carcass was reported, about 2 km of distance should be covered in about 1.5 hours to find the carcasses, and even less time was spent when the dog (not trained) was present. In conclusion, it can be stated that for searching for carcasses it is first mandatory a good collaboration with hunters or other forest visitors to report the carcasses, and involve small groups of expert people to active search the carcasses, possible with the use of hunting dogs with no special training.

Keywords: African swine fever; passive surveillance; carcasses; eradication; mobile app; freedom from animal disease; wild boar

1. Introduction

African swine fever (ASF) is a fatal disease infectious to wild and domesticated suids caused by a large double-stranded enveloped DNA virus of the *Asfarviridae* family [1]. The epidemiological characteristics of ASF and the current spread of the disease pose a global threat that can be described as the most serious animal health emergency the world has faced ever [2]. The absence of a vaccine, the rapid spread between continents and the disease's ability to affect both domestic and feral pigs makes its control and eradication even more difficult [3,4]. The declaration of disease extinction from wild boar populations is even more complicated considering their dynamic nature and the not well distributed sampling throughout the territory [5,6].

A key tool of ASF control strategies is the early detection of infected domestic and wild pigs [7]. In most European countries, surveillance in wild populations is mainly based on active surveillance during wild boar hunting. Among the surveillance strategies, passive surveillance of wild boar was considered the most effective for controlling African swine fever virus (ASFV) spread for early detection in domestic and wild pigs [5–9], as well for the virus detection even in endemic areas under conditions of disease endemicity and low prevalence, targeted passive [11], as provided in EU Directive 7113/2015 [10].

The carcasses of infected wild boar which gradually degrade seem to play a significant role in the spread of ASFV [4,12–14]. It has recently been showed that the probability of isolating the virus from carcasses is three months if the tissues are kept at a temperature of -20°C, one month at 4°C and none if the carcass is kept at room temperature [15]. Their contacts and consumption by other susceptible individuals have been already demonstrated, and consequently its correlations with disease spread [16]. Thus, rapidly finding and removing carcasses is a crucial measure for effective ASF control [7].

Passive surveillance is often limited to poorly planned sampling, resulting from the activities of inadequately trained volunteers, or citizens, or samples taken following road accidents [11,17]. Furthermore, in conditions of extremely low prevalence of the disease passive surveillance should be implemented also through the active search for carcasses [11,17].

The suspicion of ASF can be formulated following the finding of an increase in the normal mortality of wild boar in a defined territory. The EFSA opinion has defined a mortality of 10% in a wild boar population as 'physiological'; of this percentage, only the 10% may be found in the environment: it follows that the finding of carcasses in the environment in excess of 1% of the estimated resident population may provide the basis for a suspicion of ASF [11]. EFSA has also estimated the need to report, sample and test a number of carcasses (wild boar dead from non-hunting causes) equal to about 1% of the estimated wild boar population each year [11]. Therefore, in Italy, at least 5,000 wild boars per year should be tested for a population of at least 500,000 animals in the pre-reproductive period [18,19].

Even if the EFSA Exit strategy provide guidelines on the number of carcasses to be found as ASFV negative to declare the disease eradicated, the opinion did not provide indication on the practical actions to carry out when the passive surveillance must be put in place [5,10,11]. To be successful, passive surveillance requests the employment of several people for a short period. Thus, and the target approach must consider the social context and the resources needed/available in terms of the associated costs [20]. Sardinia has been recently declared free from the ASFV genotype I in wild boar [21], following the development of passive surveillance based on EFSA Exit Strategy,

The aim of this paper is to describe the strategy applied for passive surveillance implemented in Sardinia, by referring to: 1) the identification of the most suitable areas to find wild boar carcasses, 2) the number of carcasses to be found to provide evidence of absence of disease, 3) the protocol applied

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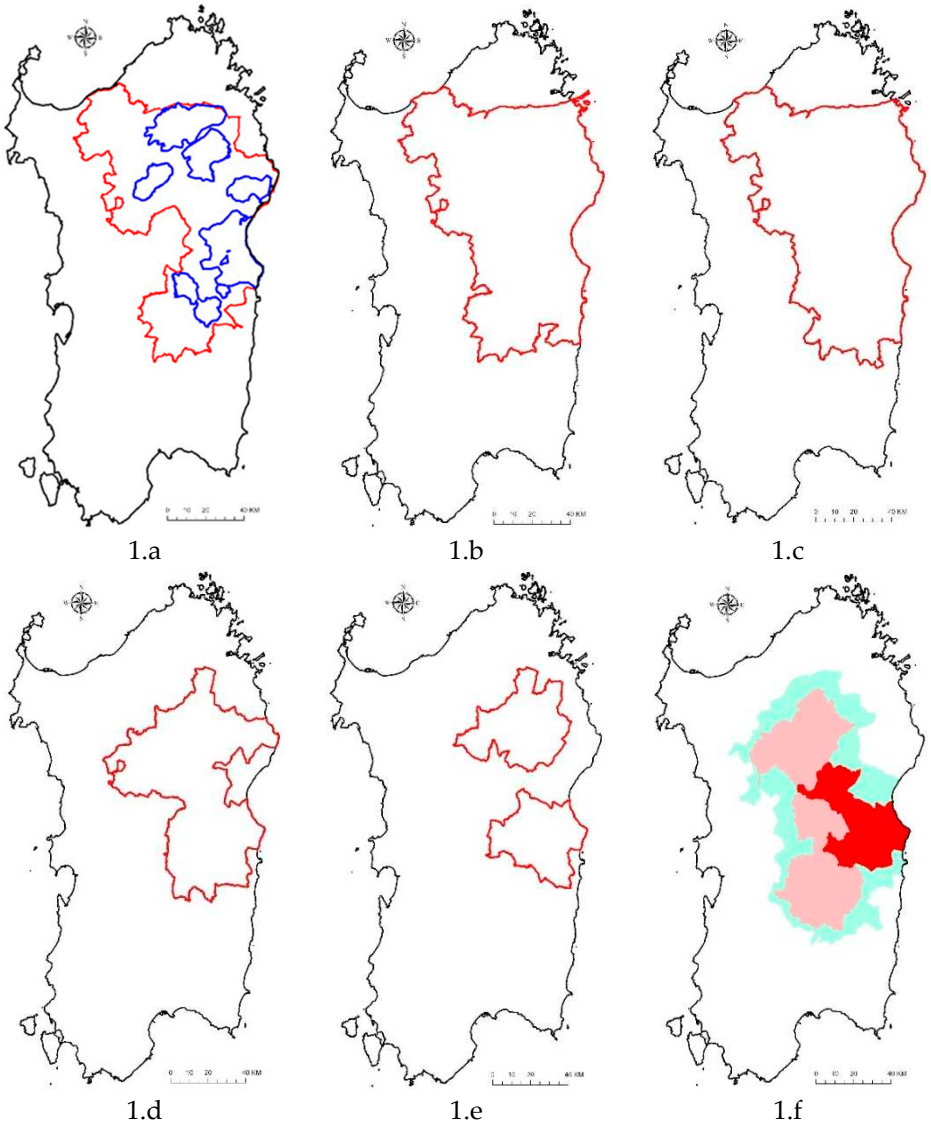
in the field (i.e., number of people, dogs, time and space); 4) the fundamental role of citizen science? and participatory methods, with a focus on the hunters role in find wild boar carcasses.

2. Materials and Methods

2.1. Study area

Although the island has been affected by ASF for 43 years, the infection in wild boar has always been limited to an infected area located in the north-east [25]. The first wild boar infection area defined in 2012 was composed eight different macro-infected areas (blue areas in Figure 1.a) for a total of 2,521 km², the area was then increased to 8,021 km² in 2015 (red area in Figure 1.a) and subsequently updated every year based on the epidemiological disease trend. In 2017 was established the larger infected area which comprised 124 municipalities and a total of 9,327 km² (Figure 1.b). Until 2017, given the decrease of ASF prevalence (both virological and serological) the infected area was reduced in 2018 to = 9,299 km²) (Figure 1.c), and to 5,322 Km2 in 2021 (Figure 1.d). In 2022 the infected area was split in two different areas, the north, and the south infected areas, based on the cluster of seropositive animals detected during the previous hunting season. These areas were respectively, 1,760 and 1,602 km² (Figure 1.e).Iin May 2023 the European Commission modified the Sardinian status and established three different geographic areas subject to special disease control measures, included in Part I, II and III of ASF risk areas, represented in Figure 1.f. Finally In October 2023, the European Commission declared the eradication of ASFV from Sardinian wild boar population [21].



1.a 1.b 1.c

1.d 1.e 1.f

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Figure 1. Excursus of the Sardinian wild boar infected areas from 2012 to 2022, and restriction zones of 2023. a) infected area instituted in 2012 (blue limits) and subsequent enlarged infected area established in 2015 (red limits); b) infected area of 2017; c) infected area of 2018; d) infected areas of 2021; e) two separated infected areas established in 2022; f) ASF risk areas included as restricted zones in Part I (green area), Part II (pink area) and Part III (red area) of the Annex I.

2.2. Sardinian wild boar population

Sardinia is an Italian island of 24,100 km² characterized by a typical Mediterranean climate and vegetation. The estimated Sardinian wild boar population is of about 100,000 animals, with a mean density of 4.5 wild boar/km² [22].

Wild boar hunting was allowed only under derogation between November-January, with an estimated population tourn-over of about the 45% [17]. All the wild boar hunted inside the infected area have to be tested for ASFV by a Real-Time polymerase chain reaction (RT-PCR), and serological tests are carried out by an Enzyme-Linked Immuno-Sorbent Assay (ELISA) as screening test, with confirmation via immunoblotting [23].

In April 2019 the last wild boar PCR positives were notified followed by a sequel of seropositive cases mainly detected in adult and subadults individuals [17,24]. Until 2019, only active surveillance was in place and prevalence data referred only to hunted or killed by road traffic animals [24]. In 2020, after about one year of virus absence, the passive surveillance was put in place all over the island, applying a context specific EFSA exit strategy [17].

2.3. Wild boar passive surveillance

As illustrated in Figure 2, the passive surveillance included different way of carcasses fundings. For the purposes of this work, the wild boar carcasses falling under the passive surveillance could be clustered as:

1. Carcasses type A: reported by hunters or other forest visitors (i.e., farmers, mushroom gatherers, campers, or common citizens);
2. Carcasses type B: reported by Forest Corp during patrol activities;
3. Carcasses type C: found in the forest during the active search with or without dogs.

Otherwise, wild boar killed by road traffic (carcasses type D) are considered as active surveillance, as suggested by EFSA [11].

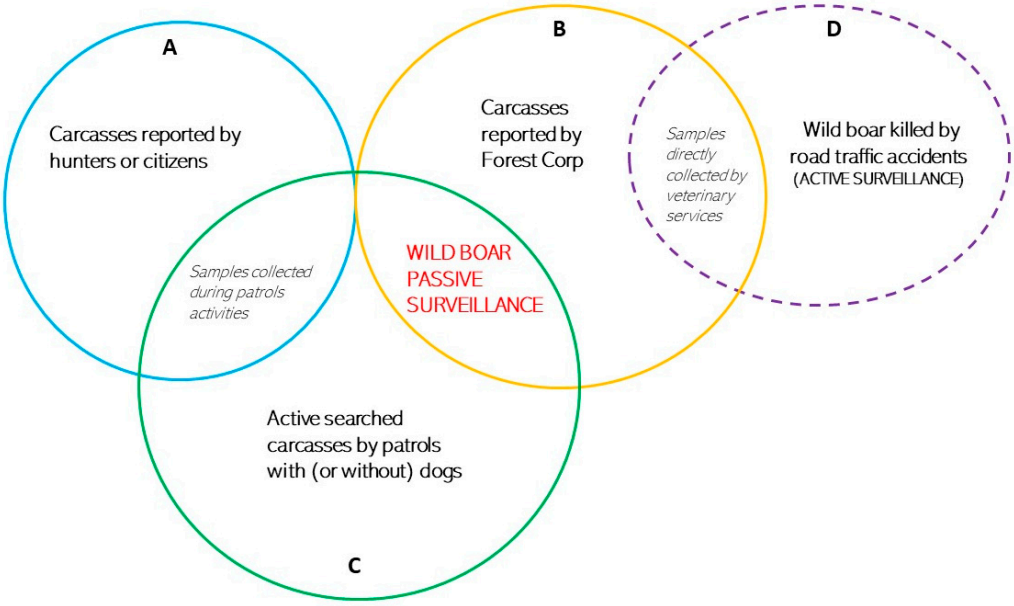

Figure 2. Graph representing the passive surveillance components: carcasses reported by hunters or citizens (A), carcasses reported by Forest Corp during patrol activities (B) and carcasses found by

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organized active search with or without dogs (C). Wild boar killed by road traffic (D) even if dead animals are not considered as passive surveillance but active surveillance.

Carcasses reported by Forest Corp during patrol activities, as well as wild boar carcasses killed by road traffic accidents, are immediately sampled by the veterinary services. If some carcass is notified by hunters or citizens during hunt or forest visits, these are reported to Epidemiological Observatory of IZS-Sardegna which organized the active search of these carcasses. During these, these carcasses are collected and sampled. Furthermore, the activities of active search on the field are also organized to find and sample wild boar carcasses not previously reported.

2.4. How many carcasses and where?

As previously defined by Cappai et al., 2022 [17], the protocol put in place for passive surveillance was based on the number of carcasses to be collected and tested in Sardinia to provide evidence of ASFV absence,: considering a surface of about 5,000 km^2, a total of 10 and 20 carcasses/year during the screening and the confirmation phases should have been detected and tested as ASFV negative to declare the area free from ASF.

Active searches of carcasses (carcasses type A or C) were planned in specific areas, starting from June 2021 till June 2023. As carried out in several countries, a specific distribution model was applied to investigate whether the fine-scale distribution of ASF-infected animals can be predicted and support wild boar carcass searches [26–28]. The results of the models led to the creation of a graphic tool providing specific indications about areas where active search of carcasses was a priority [17,24].

2.5. The GAIA observer App

A specific mobile application (App) named "GAIA observer" was developed by Xvalue SRL®, compatible with Android, iOS and Windows. The platform combined with the mobility app makes it possible to collect several georeferenced information on the environment. Gaia observer only requires GPS position to function and allows all maps of Italian regions to be downloaded free of charge (Figure 3).

Figure 3. GAIA Mobile App screen.

Given that the App was created purposely for the data collection of ASF passive surveillance project activities, particularly for the reporting of carcasses type A or type C and it was developed in Italian language to facilitate the users in the real-time findings and sightings reporting.

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The license was provided to IZS-Sardinia. The online app gives the possibility to download the overall data previously uploaded by the users, or to monitoring the real-time data forwarded by the users on-field.

The platform reserved for ASF passive surveillance includes:

i. *Report database*. The section includes carcasses code, date of founding, the name of the operator who notified the founding, municipality, province, region, latitude, longitude, altimetry, track code (unique code), and photos of the carcasses;
ii. *Realtime monitor report*. The section provides a real-time view of carcass findings with the date of founding, carcasses code and its coordinates;
iii. *Route database*. The database contains the reports of each on-field patrol: name of the operator, region, province, common, date, start time, end time, duration, distance (m) of the route;
iv. *Realtime routes monitor*. The section shows the geographical map of Sardinia where the operator in action in the field can be monitored in real time.

An interface of the App in English language is reported in Figure 4.

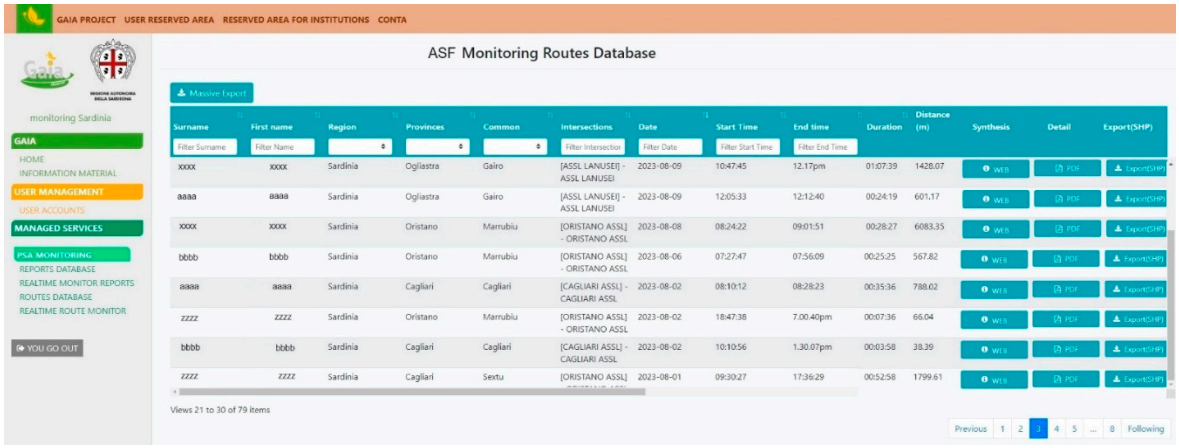

Figure 4. GAIA operator App online interface, with a focus on the routes database. Data collected were referred to the user (name and surname encrypted to guarantee the people privacy), location information (region, province, common, ASL intersection in the territory), time spent, distance travelled.

2.6. The active carcasses search protocol

The following protocol was applied to find carcasses ype A (reported carcasses) or type C (carcasses not reported found by active search).

Each involved team was composed by a veterinarian of the IZS-Sardegna, the experts of the field area from the local hunting companies, and the veterinarians of the Local Veterinary Service (ASL). Trained dog and his owner could or not participate to the search.

Each on-field activity was planned and organized based on a specific calendar, shared, and approved by each team category coordinator. Furthermore, after the identification of the target areas, a report was sent to each coordinator. For carcasses type A (Figure 2, bundle A), the area was selected according to previously notification from hunters or forest visitors; for carcasses type C (Figure 2, bundle C) the areas were selected according to the distribution model. The report included the indications about areas (i.e., shape files, coordinates, vegetation, density of wild boar, presence of farms or private properties).

Before the activities, each person must give consent to use his name, surname, affiliation, and contact (i.e., telephone number). This was essential to contact the user in case of data inconsistency, or doubts or errors of the data collected. Subsequently, the user had to download the GAIA observer App on its personal mobile for real-time data collection (routes, times, patrolled areas, and carcasses found).

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At the spot, the team was informed about the area to be covered, possible carcasses already reported, the sub-division of the territory, presence of private properties, expected duration of the search and final the meeting point. People were equipped with high visibility caps or jackets, transceivers, gloves, and material for sampling (Figure 5).

Figure 5. People involved in patrol activities during the carcasses founding.

When a wild boar carcass was found, the person who found it had to register the GPS point of the finding, inform the veterinarians of the team by phone or transceivers, and upload the data on the GAIA observer App. The veterinarians were responsible to collect samples from carcasses, in compliance with the biosecurity measures, storage the samples and send them to the IZS-Sardinia laboratories. The samples collected from carcasses were subjected to the same procedure applied to all samples collected by hunted wild boars and subject to the viral DNA searched by Real Time PCR, as established by the OIE diagnostic manual [23].

The conservation status of the carcass (i.e., fresh, in decomposition, or mummified) was assessed during sample collection [29]. Furthermore, the veterinarians were responsible to upload the data related to location (region, province, municipality, latitude, and longitude), date of sampling, age, and sex of the carcasses (observed or hypothesized) and the type of sample (i.e., spleen, blood, tonsil, kidney, or lymph node) in the national animal disease database for passive surveillance (SINVSA).

2.7. Data collection and analyses

A specific database was created including the data recorded by the GAIA observer App and available for downloading in Excel .csv format. Each record was checked to evaluate its validity (i.e., switch on of the app at the time of activities and switch off when the activity ended, use the app only walking, stop the app when the user moves with car). Any incongruency was corrected after consultation with the user.

Correspondence analysis was performed to evaluate data collected by GAIA observer and those recorded in SINVSA by the veterinarians.

Simple descriptive statistics were carried out to summarize the number of on-field session, number people with or without dogs, time spent, distance travelled, and number of carcasses found. Name and surname of the operator, municipality and date were used as key to match the carcasses database (Report database) and the route database (Realtime routes monitor).

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3. Results

From 2021 to 2023, a total of 148 wild boar carcasses were found and samples in Sardinia, both inside (33, 22%) and outside (115, 78%) the 2021 infected area, as reported in Table 1.

Table 1. Passive surveillance data of 2021-2023 years from the whole Sardinian region. Data are presented as number (percentage), distinguishing from infected and not infected area of 2021.

Year	Infected area	Not infected area	Total
2021	11 (24%)	34 (75%)	45 (30%)
2022	13 (19%)	54 (81%)	67 (45%)
Jan - Jun 2023	9 (25%)	27 (75%)	36 (24%)
Total 2021-2023	33 (22%)	115 (78%)	148

Specifically, 45 carcasses were collected in 2021 (11, 24% inside and 34, 75% outside the infected area), 67 in 2022 (13, 19% inside and 54, 81% outside the infected area), and 36 in 2023 (9, 25% inside and 27, 75% outside the infected area).

Of these 148 carcasses, only 22 were collected during the active search with the use of GAIA observer App. The other 126 were found by Forest Corp during the patrol activities, or by citizens that voluntary informed the Forest Corps, and sampled by the veterinary services (carcasses type B, Figure 2, bundle B).

To obtain the free status, the organized patrol activities were mainly focused inside the infected area, as represented in Figure 6.

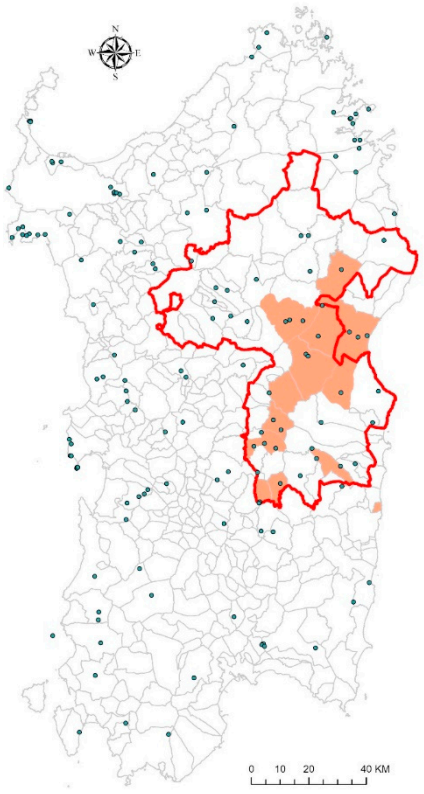

Figure 6. Sardinian map summarized the geographical distribution of wild boar carcasses and patrol activities. The points indicate the carcasses found during 2021-2023, inside and outside the 2021 infected area (red line). The orange areas indicate the municipalities where the patrol activities with the use of GAIA observer App were put in place.

A total of 235 records were downloaded by GAIA observer App related to each user during the 6, 11 and 16 on-field activities carried out in 2021, 2022 and 2023, respectively. The 21% of the records

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(35 records) presented anomalies: distance coverage equal to zero, duration lower than ten minutes, proportion between distance coverage and duration higher than 4 km/hours (human average speed). Few of these records (9, 5% of the total) were correct after consultation with the user, while the other 26 (15% of the total) were excluded by the analysis. Summarized data on carcasses collected during the 33 active searches are reported in Table 2.

Table 2. Detailed data on patrol activities carried out in 2021-2023 years with the use of GAIA observer App. Data are presented as data of activity, number of humans involved, number of dogs eventually involved, surface explored by humans and dogs (kilometers), duration of the session (hours), speed of humans (kilometers for hours), and number of wild boar carcasses eventually found, and type of carcasses based on 2.3 categories.

Data	Humans involved	Dogs involved	Surface explored by humans (km)	Surface explored by dogs (km)	Total duration of the field sessions (hours)	Average human speed (km/hours)	Number of carcasses detected	Type of carcasses*
13.05.2021	22	0	17.15	0	2.78	0.28	0	-
27.05.2021	15	0	22.75	0	3.59	0.42	0	-
16.06.2021	14	0	9.90	0	3.41	0.21	0	-
29.06.2021	8	0	23.75	0	4.23	0.70	0	-
14.10.2021	4	0	13.03	0	3.25	1.00	0	-
21.10.2021	11	0	22.99	0	2.88	0.73	0	-
17.05.2022	7	2	13.15	26.15	3.31	0.57	4	A
28.05.2022	5	0	11.11	0	3.23	0.69	0	-
07.06.2022	4	0	10.92	0	2.37	1.15	1	A
15.06.2022	4	0	7.14	0	2.45	0.73	2	A
16.06.2022	5	0	9.42	0	2.78	0.68	0	-
05.08.2022	8	0	15.16	0	4.10	0.46	0	-
13.08.2022	11	1	17.99	18.51	3.06	0.53	0	-
15.09.2022	8	0	12.85	0	2.15	0.75	0	-
16.09.2022	8	0	31.57	0	4.38	0.90	0	-
10.10.2022	5	0	5.10	0	0.55	1.86	1	A
28.10.2022	3	0	1.50	0	1.10	0.45	4	A
16.03.2023	4	0	9.47	0	1.59	1.49	2	A
04.05.2023	4	1	5.74	7.41	1.28	1.12	0	-
03.06.2023	7	0	38.01	0	3.18	1.71	0	-
01.08.2023	6	1	21.68	12.12	1.22	2.96	2	A
02.08.2023	2	0	3.15	0	0.55	2.86	0	-
06.08.2023	10	0	25.79	0	2.59	1.00	0	-
09.08.2023	3	0	7.46	0	1.32	1.88	2	A
10.08.2023	5	0	13.68	0	2.23	1.23	0	-
13.08.2023	4	0	9.56	0	1.75	1.36	0	-
16.08.2023	3	0	15.02	0	3.15	1.59	0	-
22.08.2023	4	0	8.12	0	1.48	1.37	1	A
01.09.2023	18	1	51.90	22.76	3.25	0.89	0	-
18.09.2023	12	1	28.00	14.55	3.02	0.77	0	-
25.09.2023	3	0	2.06	0	0.45	1.52	2	A
29.09.2023	3	0	5.90	0	1.23	1.60	1	C
07.10.2023	1	1	1.43	5.26	1.14	1.25	0	-

* type A refers to previously reported carcasses, type C refers to carcasses never reported and found during the patrols activities.

The six active searches of carcasses carried out in 2021 were useful to understand that several non-expert persons cover a too small area and take too long to cover it: up to a maximum of 22 not expert persons were involved for a total surface explored of about 17 km, (average speed in 2021 = 0.56 km/hour). No wild boar carcasses were found in 2021 by these activities.

Since 2022, the protocol involved smaller teams (about 5-7 expert people), with one or two trained dogs. An average speed of 1.3 km/hour was reported by the participants. Furthermore, considering that the activities organized in 2021 to find carcasses based on the distribution model yielded no results, the activities were focused on area where the possible presence of wild boar

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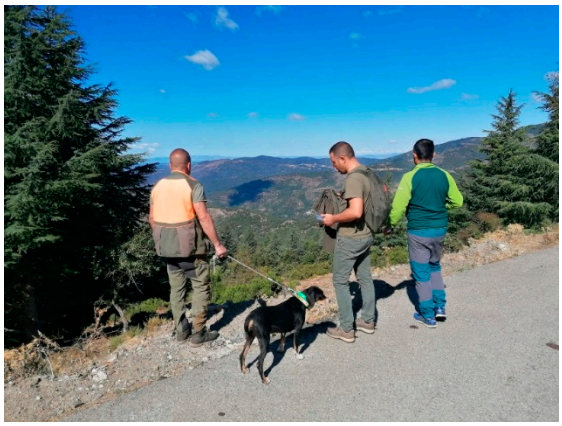
carcasses were notified by hunters. A total of 12 carcasses were found in 2022 during 5 carcasses active searches, all previously notified by hunters (carcasses type A, Figure 2, bundle A).

In 2023, the protocol was further refined: only 4 people were generally involved in active searches of carcasses (Figure 7.a) to catch up the previously reported carcasses, with an average speed of 1.5 km/hour. When the carcass was reported, about 2 km of distance should be covered in about 1.5 hours to find the carcasses. Even less time was spent when the dog was present as reported for the activity of 01.08.2023 when 1.2 hours were employed (Figure 7.b). A total of 10 carcasses were found in 2023 during six active searches of carcasses. Only two carcasses not previously reported were found during this year, in 22.08.2023 (Figure 7.c) and 29.09.2023 (Figure 7.d) and classified as carcasses type C (Figure 2, bundle C).



7.a

7.b

7.c

7.d

Figure 7. Patrol activities results; a) the expert team with trained dog; b) the trained dog found a wild boar carcass previously notified by hunter; c) the wild boar carcasses found in 22.08.2023 (not previously reported); d) the wild boar carcasses found in 29.09.2023 (not previously reported.

4. Discussion

The international spread of the ASF virus is adversely affecting animal health and welfare, resulting in economic losses of tens of billions of Euros per year, and potentially affecting livelihoods [29,30]. The situation was also complicated by the absence of any validated strategy to define an ASF free an area following the detection of the virus in wild boar population and where the virus is not anymore detectable under the usual surveillance condition (hunting and opportunistic finding of wild boar carcasses) [31].

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At present, through the EFSA Exit strategy, is possible to define a free ASF wild boar population despite the sporadic presence of seropositive adult wild boar that could represents the normal evolution of an infection which agent has been eradicated [17,32].

The Italian National Surveillance Plan for ASF (PNSE PSA 2023), in compliance with the latest recommendations of the European Commission and the scientific opinions of the EFSA, defines the passive surveillance of ASF in wild boars as a key means of epidemiological surveillance and the most important basic tool for the eradication of the ASF virus from the natural environment in the areas where this disease occurs, especially in the early stage of its development [17,33]

The design of a sufficiently sensitive ASF surveillance system requires a solid understanding of the epidemiology related to the local eco-social context, which allows it to be risk-based and thus make optimal use of the usually limited financial and human resources [7,34]. If stakeholders understand the value of the data collected and the impact of surveillance on animal and human health, this should lead to a better design of surveillance activities, greater compliance with data collection, a greater probability of investment and achieving the overall goal of surveillance to enable the development of the 'one health' approach [35].

Community involvement in scientific processes has the potential to combine useful data collection with awareness-raising and education, helping to bridge the knowledge gap between academia and the general public [36] and well-designed citizen science projects can usefully inform research, decision making and policy formation [37].

Surveillance of wildlife diseases poses considerable logistical challenges compared to that of humans or livestock [38]. In contrast to traditional epidemiology, citizen science, a passive surveillance model and innovative participatory approach, engages law enforcement, farmers, hunters, trekkers, birdwatchers, and ordinary citizens in scientific research, facilitating the collection of large data sets and increasing public awareness [39].

Hunters, a special and often overlooked group of citizen scientist [40], offer unique advantages giving their knowledge of the territory, facilitating the collection of long-term data over large areas that otherwise would not be possible to investigate [41]. Thus, should be considered a standard partnership [45]. Hunters play an important role in wildlife management and thus also in the implementation of disease control measures in the wild boar population, especially in the enforcement of ASF control measures and passive disease surveillance [42].

In Sardinia several training courses were carried out for hunters giving information on ASF, the way to report wild boar carcasses, the importance of passive surveillance aimed at ASF eradication from wild boar and finally their paramount role in collecting or reporting the highest number of carcasses. The awareness process includes by social media (i.e., Facebook), leaflets posted in the location frequented by hunters (i.e., wild boar handling points) or sent to dedicated mailing lists (Supplementary materials Figure 1.s).

The active search of wild boar carcasses (carcasses type A or C) requires the involvement of several persons and huge economic resources [43–45]. The results of this study confirmed the key role of active search carcasses and their removal in eradicating ASF, in combination with hunting activities [46] and the importance of implementing, even in the long term, the application of user-friendly apps for efficient data collection. Furthermore, it demonstrates that in the Mediterranean habitats the dense vegetation makes difficult to search carcasses and only expert and motivated persons are able to carry out it. Among expert persons, hunters are the easiest to involve. They are the most regular wood visitors given that often they are also farmers and mushroom gatherers. Moreover, in Sardinia the hunters not only know very well the landscape, but are censed for the authorization of the under-derogation hunting season, thus they are easy to contact and gather [47].

During the active search of carcasses, several problems were faced given the rough vegetation, the availability of people, and the typical Sardinian climate. Specifically, the possible presence of carcasses type A in a more or less large area was notified by hunters or citizens and the active search of carcasses were organized in this area. Otherwise, persons who searched the carcasses were not the same persons who notified the carcasses. Thus, when the precise location of the reported carcasses was available the patrols needed very short time to find them, supported by the georeferenced data

via GAIA observer app. Otherwise, the session took longer time due to large area to be explored including the impossibility to detect any carcass.

Finally, even if the presence of the dogs was not always guaranteed, the results confirm that for searching for carcasses hunting dogs with not special training can be applied with good results, as previously stated [44].

In the final stage of ASF eradication process there is shadowed time-period during which the virus is not anymore detected because of its extremely low prevalence and the number of found carcasses is also very low due to the possible absence of the virus and related mortalities. In such epidemiological situations the probability to detect an ASF-positive wild boar is much higher in animals found dead than in hunted animals [11,17,48]. Thus, the willingness and the motivation of hunters to support passive surveillance is of utmost importance using participatory methods [42]. Citizen science and participatory methods can enable a wider coverage, but building an efficient disease monitoring system that relies on hunters is the major challenge [38].

Supplementary Materials: The following supporting information can be downloaded at the website of this paper posted on Preprints.org.

Author Contributions: Conceptualization, Federica Loi, Marco Pisanu, Vittorio Guberti and Stefano Cappai; Data curation, Federica Loi, Francesca Porcu, Daniela Mandas, Vittorio Guberti and Stefano Cappai; Formal analysis, Elisabetta Coradduzza, Federica Loi and Stefano Cappai; Funding acquisition, Federica Loi; Investigation, Elisabetta Coradduzza, Daniela Mandas, Fabio Secci, Marco Pisanu, Cinzia Pasini, Carlo Zuddas, Marcella Cherchi, Daniele Denurra, Ennio Bandino, Antonio Pintore and Stefano Cappai; Methodology, Elisabetta Coradduzza, Federica Loi, Fabio Secci, Marco Pisanu, Antonio Pintore, Vittorio Guberti and Stefano Cappai; Project administration, Federica Loi; Resources, Federica Loi, Marco Pisanu, Antonio Pintore and Stefano Cappai; Software, Federica Loi and Stefano Cappai; Supervision, Federica Loi, Marco Pisanu, Antonio Pintore, Vittorio Guberti and Stefano Cappai; Validation, Federica Loi, Francesca Porcu, Daniela Mandas, Daniele Denurra, Ennio Bandino, Antonio Pintore, Vittorio Guberti and Stefano Cappai; Visualization, Elisabetta Coradduzza, Federica Loi, Francesca Porcu, Daniela Mandas, Carlo Zuddas, Marcella Cherchi, Daniele Denurra, Ennio Bandino, Antonio Pintore, Vittorio Guberti and Stefano Cappai; Writing – original draft, Elisabetta Coradduzza, Federica Loi, Francesca Porcu, Vittorio Guberti and Stefano Cappai; Writing – review & editing, Elisabetta Coradduzza, Federica Loi, Francesca Porcu, Vittorio Guberti and Stefano Cappai.

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Institutional Review Board Statement: Ethical review and approval were waived for this study due to not include experiment with animals but only results from samples of died pigs, that must be tested for African swine fever, as established by the OIE manual and by the European Commission decision for the infectious disease. These samples are not under regulation of the ethical committed.

Informed Consent Statement: Not applicable.

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