Evaluation of biosafety levels in a wildlife hunting farm of Central Italy (Umbria region)

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SUMMARY

The interface between the domestic and wild environment represents a critical point for the transmission of etiological agents from the wild to the domestic world, and consequently to humans. This hypothesis, albeit unconfirmed, has been also suggested in the case of the COVID-19 pandemic, highlighting how an infection of wild animals can lead to a pandemic. Livestock farms, especially extensive or faunistic-hunting ones, with significant contact between wild and domestic animals, pose a risk in this regard if appropriate interventions are not implemented. This study evaluates the infectious risk at a qualitative level of a faunistic-hunting farm in Central Italy, representative of other extensive farms, through visit of the farm and interview of the farm manager using a simplified model of questions adapted from the ClassyFarm checklist. The level of biosecurity of the farm was examined, identifying critical points and strengths. The interview, which provided explanations of the investigated biosecurity actions following each question, also served as a training opportunity. The results emphasize the need to tailor measures to the specific characteristics of the analyzed farm and assess their impact on animal health, as well as from a One Health perspective on human and environmental health, considering the proximity to cattle farms and wild animals. Effective prevention is ensured through the implementation of biosecurity measures that require multidisciplinary skills in both planning and application, actively involving professionals, including within the scope of animal husbandry. Furthermore, the evolution of animal health laws requires an expansion of competencies to address emerging needs, such as training.

KEY WORDS

One Health; biosecurity; infectious emerging diseases; animal farming; wild animals.

INTRODUCTION

The experience of the COVID epidemic and the hypothesized genesis of the SARS-CoV-2 virus, which predicts its passage from wild animal species to humans through intermediate species, have made it clear to everyone the effect that the emergence of an epidemic caused by an emerging infectious agent can have globally [1,2]. Remaining in SARS CoV-2, currently and unexpectedly the only animal species in which there has been widespread intraspecies transmission following human infection are american mink and white-tailed deer, having an efficient animal-to-human transmission [3,4].

It is estimated that 5 new emerging human infectious diseases appear every year, 3 of which are zoonotic, meaning transmitted from animals. In addition, regardless of the nature of the pathogen, it is estimated that an inter-pandemic period of 11-40 years exists between epidemics [5,6]. Countries therefore depend on their ability to prevent and predict these situations. The European Union is moving in this direction, seeking to contain anthropization, which is considered one of the factors favoring the development of emerging infectious diseases [2]. Livestock farming, a sector of particular economic interest in Europe and in Italy country, represents an environment in which the conditions of human-animal and domestic-wild animal contact can facilitate the onset and spread of infectious diseases. The different types of farming, for example intensive or extensive, present different risk profiles. If the weakness of intensive farming is represented by the number of animals per unit of surface area, in extensive farming, now particularly appreciated for its positive effects on animal welfare and environmental sustainability, the risk of contact between domestic and wild animals, and therefore of transmission of contagious diseases, is

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greater [2,7]. The points of contact between these environments, generally the borders, become critical points, as has already emerged for African Swine Fever [8].
Prevention is based on the application of biosecurity measures, intended as all the tools (building works, facilities, devices, operational procedures, etc.) aimed at preventing or reducing the onset of infectious diseases. The farmer has a fundamental role in managing this risk, considering, however, that his role requires tools, both technological and training-related, that can only be defined through a broader and multidisciplinary perspective than that of the main professionals operating in the sector (Agronomists, Animal Scientists, and Veterinarians) [2,9].
The purpose of this work is to improve awareness on the biosecurity and emphasize the possibilities of implementing biosecurity measures tailored to the specific type of farming, through the description of a case study in which a qualitative evaluation of infectious risk was performed and possible actions to increase biosecurity have been proposed in a wildlife-hunting farm.

MATERIALS AND METHODS

Study design

A visit of the farm was conducted by two professionals trained in biosecurity to obtain information and analyze the logistical and structural aspects of the farm; further information on the biosecurity situation was obtained through an interview of the manager of the farm.
The interview had the aim of both acquiring the necessary information but at the same time making the interviewee aware of the concept of biosecurity. During the interview the objectives of the questions were explained to the interviewee, following the answers.

Description of the wildlife hunting farm

The farm examined covers approximately 700 hectares and is located in a hilly area in Central Italy, from 334 to 670 meters above sea level. The entrance to the farm is through a gate which is the only form of access to the farm. The destination is both for agricultural land use and as a hunting reserve, therefore different activities are practiced there. The farm borders two beef cattle farms, one on each side, in an area at low density of domestic animal farms. The farm has approximately 350-380 roe deer, over 200 wild boars, 25-35 red deer, 25-30 fallow deer, 300 pheasant, around 200 hares, some wolves and red foxes, all estimated annually by census.
From a vegetation point of view, the area is mainly characterized by Mediterranean bush. The reserve encloses within its perimeters several natural water sources: a reservoir, a large river, and many smaller streams and canals (flowing mostly in winter) around the perimeters, serving as natural boundaries.
An area of the farm has been dedicated to the processing of hunted animals, according to the regional guidelines relating to food safety [10]. There is a disposal system for animal by-products (part of carcasses, digestive tract content, ans do on) that are taken away by a company that deals with the disposal of this type of waste [11].

Data collection

The manager of the farm was interviewed using a simplified model of the questions present in the ClassyFarm checklist, paying particular attention to the aspects of biosecurity, i.e. the set of actions, tools and procedures useful for containing the risk of introduction and transmission of infectious diseases, as currently there is no specific system referring to extensive farming [12]. The objective of the interview was to raise awareness of culture of the biosecurity and to identify the critical points present in the farm for the introduction and spread of infectious diseases.
Additional specific literature on biosecurity and infectious risk for wildlife hunting farms has been gathered from international sources [13,14].
The 35 selected questions were divided into the following macro-areas: general section, management, hunting activity, external inputs, staff training, and biosecurity.
The questions asked during the interview were extensively reported in a degree thesis on this topic [15]. The main questions and topics carried out are reported in the Tables 1-5.

RESULTS

Based on the visit on the farm and the information acquired by the interview, a qualitative analysis of the biosecurity of the farm was performed. The primary infectious risks on the farm were associated with the transmission of infections between external wildlife and internal animals, or vice versa, involving both wild species (e.g., wild boars) and domestic animals (e.g., cattle).

Table 1 - Questions, answers and comments based on biosecurity point of view for general section.

Questions	Answers	Comments	Possible actions of improvement
General section			
Is this farm used for agricultural activities? If so, what kind of crops are there?	Yes, this land is used to cultivate crops like pasturage and hay. At times it is used to produce legumes along with 2nd cut alfalfa.	-	-
What are the internal activities and how do they work?	Mainly logging by third-party of woodland companies (by both local and foreign workers). We also practice sow and hay harvest and road and building maintenance	The uncontrolled entrance of people can lead to the introduction and spread of infectious diseases	Checking the entrances and increasing the biosecurity measures to limit the spread of infectious diseases such as “no entry” signs, change of clothes and shoes, etc.

The main points that emerged during the interview are reported in the Tables 1-5.
About the location and natural structure, the farm has many positive aspects that contribute to biosecurity:
- the numerous water sources make it possible to dilute the animals in the area for watering purposes, reducing the possibility of direct contact. In fact, water sources are recognized as a risk for infectious diseases due to the potential proximity of several animals while drinking and the possibility that water may protect some etiological agents from inactivation.
- The farm is located in an area with very low livestock density (two beef cattle farms), generating a low level of risk of transmission of infectious diseases between domestic and wild animals. The animals of the hunting farm represent populations that live in a controlled environment but belong to species that occur naturally in the wildlife and that have the same susceptibility spectrum of other external wild and domestic animals, representing a possible bridge for infectious diseases. The multi-species nature of the farm is intrinsic to the wildlife-hunting type of farm, which predisposes the farm to a number of

Table 2 - Questions, answers and comments based on biosecurity point of view for management of the farm.

Questions	Answers	Comments	Possible actions of improvement
Management			
Is the animals' nutrition natural or integrated? Is it completely natural?	It is completely natural.	This is positive because the introduction of external feed could be a carrier of infectious disease	-
Are the water sources shared by a variety of animals?	Yes, the water sources are shared by different animals. Luckily though, there are many different water sources like a reservoir, the (Chiascio) river, the stream and many canals spread throughout the area.	This is a great advantage for the farm. Being rich in water sources allows a reduction of the possibility of contact between the animals, reducing the possibility of spread of infectious diseases.	-
How does the fence system work? What's its height?	The fence is 1.80 meters high. The metallic fences are placed in the perimeters where there are no water sources. So, the perimeters are defined whether by a metallic fence or by watercourses 6 meters wide.	The fence system is useful to reduce the possibility of contact with external animals. Their damage is a risk because it makes it easier for the animals to enter or exit the perimeter of the farm.	The regular maintenance of the fencing system is an important investment that needs to be organised but that leads to a significantly lower risk of infection.
How deep is the fence?	The fence is 10-20 centimetres deep and it was built in 1980, so it needs to be renovated. Moreover, hunters periodically damage the fence in different spots to encourage wild boar to get outside of the perimeters.	-	-
How often is the fence checked? Are the damaged parts restored?	The fence is checked once a year for 15 days, just before the hunting season begins. The damaged parts are restored.	-	-
Is there closed-circuit television (CCTV) in the wildlife hunting farm?	No, just camera traps for counting wild animals installed by other institutions.	-	-
Are the animals that are introduced in the farm identified? If so, how does the load and unload of the animal work?	The introduction of external animals is not allowed. Those that enter the perimeters of the reserve get in spontaneously, they are mainly wild boars. In this sense the fram has made hunted and dead wild boars available for analysis for African Swine Fever.	This is a good point in terms of the limitation of infectious disease.	It would be useful to regularly test the population of wild boar, mainly in regard to the African Swine Fever, so that we can be sure to have a healthy population.
Is reproduction internal in the farm?	The reproduction is completely natural and internal.	This is a good point in terms of the limitation of infectious disease.	-
Are there dogs on the farm? Can they get out of the farm perimeters?	Yes, there are around 15 dogs but only one has the tendency to escape from the farm.	They could potentially act as carriers of infectious diseases.	-

multi-species infectious diseases (i.e. leptospirosis, salmonellosis, coxiellosis, etc.).
-The animal diet administered in the farm does not require supplementation; this is an important aspect for biosecurity as it reduces the entry of vehicles and external people. Food introduced on farms or means of transport have often been the cause of the spread of highly diffusive diseases, such as highly pathogenic avian influenza, African Swine Fever, swine coronavirus and so on [16-18].
From a managerial and structural point of view, the farm is visited for various activities and some aspects were considered deficient and to be implemented for biosecurity:

- lack of no-entry signs for unauthorized personnel at the entrance and along the fences; these signs could be easily placed and act as a deterrent, at least for occasional visitors. Information posters could be used to summarize all the good practices useful for avoiding the spread of infectious diseases.
- Absence of a specific disinfection area, which could be designed at the entrance to disinfect incoming vehicles.
- The creation of a reserved parking area would be useful for

Table 3 - Questions, answers and comments based on biosecurity point of view for hunting activity and external entries of the farm.

Questions	Answers	Comments	Possible actions of improvement
Hunting activity			
Are visitors checked when entering and leaving the farm? Is there a specific assiduity of entrances over the year?	Most of the entries happen during the hunting season. This, for the wild boars, lasts for three months, usually from the 30th of October to the 31st of January, with possible variations. For roe deer, some selection hunting plans are organised based on the surplus of counted animals.	-	It might be useful, in the busiest periods, to pay more attention than usual to the measures of biosecurity normally applied, so that we can be sure that the risk is lowered.
What is the meaning of selection hunting?	Cervids in Umbria are hunted following specific selective plans structured on the base of gender and age. These plans are annually revised in accordance with the results of the population censuses.	-	-
How is the surplus of hunted animals calculated?	The number of counted/censused animals is then sent to the Regione Umbria institution that will calculate whether there is a surplus or not.	-	-
External entries			
How many people, over the year, enter the hunting reserve?	Approximately 10 people during the hunting season for the selection hunting, plus another 5 special hunters who are allowed here all over the year for the containment hunting (reserved to roe deer).	It is good that the numbers of people entering the farm are not too high, but that does not mean that they must be less careful in terms of biosecurity measures.	-
Is there a specific parking area in the farm? If not, would it be possible to identify one?	No, there is not. We could try to identify a parking area, but its success would be limited since hunter's roam around with their own cars.	Not having a specific parking area means that whoever gets in the farm can freely roam around, potentially carrying around etiological agents.	It would be a useful biosecurity measure to identify a parking area or install a designated area for disinfecting the tyres of vehicles entering the farm, reducing the possibility of spreading infectious agents.
Do people such as producers of feed, zootechnical specialists, or people that could have had contact with other animals come to the farm?	Yes, they do. Mainly farmers who work the land of the farm (and work with their tractor) and contractors who cut the wood.	These are all potential risks for the spread of infectious diseases.	Correctly applying some biosecurity measures, such as change of clothes, change of shoes and checking the entrances, would be a very useful tool to keep the wildlife healthy.
For the hunters coming from elsewhere, is the accommodation within the farms' perimeters or somewhere else?	No, they usually stay in hotels nearby.	-	-

gathering visitors, including hunters, limiting the potential introduction and spread of infectious diseases, when people entering the perimeters go to visit other agricultural land, maybe with livestock, and potentially carriers of infected fomites. Internal vehicles could be provided for the internal movements of the hunters.

- The fence is a critical point for a farm. In this case, external hunters break the fences to allow farm animals to escape and be hunted; this represents a problem in terms of biosecurity, especially if the escaping animals then return within the perimeters of the hunting reserve, bringing with them microorganisms that they have taken from outside. The fence is therefore important to limit and control the movement of animals. In this regard, it could be useful to educate local farmers on the biosecurity and risks involved and favors cooperation to promote the existence and control the status of fences. It may also be helpful to install security cameras along perimeters to control them.
- Given the relevance of infectious diseases and their spread among different animal populations, even if it was noted during the interview that employees are aware of the basic principles of hygiene and infectious diseases, it is important to maintain an appropriate level of training. In this sense, it would be important to define and follow some basic biosecurity rules for internal and external personnel, such as changing shoes, wearing clean and sanitized clothes and dedicating a special area to this sanitization to reduce the probability of introducing potential pathogens. Moreover, the personel operating in the farm demonstrated knowledge of the main infectious diseases of ungulates.
- It should not be allowed to leave food or waste on the holding by personnel to avoid the risk of introducing pathogens like African Swine Fever. Waste containers could easily be installed within the area but should be made inaccessible to animals. At the moment workers who work on the ground or in the wood of the farm or in the fields during the day do not have this possibility. Moreover, the farmers who work the land often come from their farms, where they have livestock, and bring in undisinfected clothes, shoes, tools and their tractor, posing a high risk of introducing numerous infectious diseases.
- Another advantage of the farm was the sensitivity of the management to the health situation of the animals and the prompt attention to surveillance, participating in voluntary surveillance plans of the national health system. In fact, when requested by the national veterinary service, the farm has promptly and voluntarily made the animals available, in particular the wild boars, dead or killed, for the surveillance of the African Swine Fever, participating to a national plan. At the same time, this analysis of dead animals is useful to detect the presence of any other etiological agent.

In the interview, the animal welfare parameter was not mentioned, even if it is a fundamental element to be analyzed in accordance with national and European laws; however, in this farm, animal welfare was clearly expressed as the ethology of the animals respected the natural rhythms and the density of the animals was controlled.

A key qualitative aspect to highlight is that, following the interview, the farm manager independently recognized critical biosecurity issues that had previously been overlooked. Specifically, the manager identified concerns regarding personnel who regularly enter the farm, especially for work reason, after having contact with external animals, which could serve as infection vectors. Additionally, it is important to note that this type of farm is not currently classified as a target by the ClassyFarm system.

DISCUSSION

This study represents an attempt to analyze the risk of infectious diseases in the management of a faunistic-hunting farm, hosting wild animal species in a controlled context and underlining how from a One Health perspective there are always links between different species. The proposed model of analysis is very easy and can be applied to perform a prelim-

Table 4 - Questions, answers and comments based on biosecurity point of view for staff training of the farm.

Questions	Answers	Comments	Possible actions of improvement
Staff training			
How many people work and manage the wildlife hunting farm?	Two people who are security guards (Guardie Particolari Giurate GPG) authorised by the prefecture, live within the wildlife hunting farm and are also trained and authorised to handle fresh meat.	-	-
What are their responsibilities?	They check and survey the area in general, also collaborating on the internal activities. Mainly, during the hunting season, they oversee the evisceration of the carcasses. The evisceration is operated in the dedicated room and after that the carcasses can remain for up to five days in the cold room, before being sent to the slaughterhouse.	Therefore, it is very important that the staff knows the basic rules of biosecurity and food safety when it comes to managing the carcasses and the eviscerated bodies, not to contaminate what will become meat or take zoonotic infections.	-

inary infection risk analysis and to begin to improve the biosecurity level of a farm. In fact, this model can be applied and shared with other agricultural contexts comparable with the one studied, which allows to highlight the importance of the application of processes and methods of biosecurity and all the economic, environmental and social benefits deriving from them. A higher level of biosecurity increases animal welfare by reducing disease status from infectious agents, that is another relevant objective to breed animal in European Union [9]. Moreover, increased awareness of the concept of biosecurity improves acceptability of current policy and facilitates a transition to sustainable production.

An immediate impact of the interview and analysis was to increase awareness of manager and staff on the risks of infectious diseases and the potential factors that can cause them and that could be better managed to reduce this risk. The interview itself served as a training opportunity. This translates into greater attention to actions and procedures that were not previously placed in attention. This underlines how much training is a tool to make farms autonomous in the planning of biosecurity plans, as required by European regulation [9] and recently also at national level [19]. However, personalized and multidisciplinary advice would be appropriate for each farm, considering the typical characteristics of a specific area, the attitude of the farm and the type of production, focusing on protection of the environment and the territory. Often many operator habits are considered normal while on the contrary they represent a risk of introducing infectious agents.

Structural aspects should be considered at the beginning of the planning of a farm and maintained to ensure high levels of biosecurity, but they are often not present because they were not considered for farms created years ago or reconverted. Structural interventions are often expensive and in some cases technologies can help to better approach these aspects (choice of new construction materials, use of biosensors, use of surveillance cameras, etc.), also from the perspective of environmental sustainability in a One Health approach. In this case, fences represent the barrier between the farm and the wildlife and Eu-

Table 5 - Questions, answers and comments based on biosecurity point of view for biosecurity of the farm.

Questions	Answers	Comments	Possible actions of improvement
Biosecurity			
Do the staff know about transmissible infectious diseases between species?	Yes, they know the basic of it.	This is a good starting point, but constant development and improvement can be when it comes to the application of biosecurity measures.	Through training
Do the staff adopt biosecurity measures like the change of shoes or the change of clothes?	Not really.	This can be improved to lower the level of risk.	Through training
Are the biosecurity measures applied by people coming from outside of the wildlife hunting farm? Like change of shoes, change of clothes, restricted access areas, etc...	No, not at all.	This can be improved to lower the level of risk.	Through signs and regulations
Is the contact of wild animals possible with animals of neighbouring farms, maybe sharing the same space? For the transmission of diseases like tubercolosis, paratubercolosis.	Yes, it is possible but just outside the perimeters of the wildlife hunting farm. It is possible, in fact, that wild boar and red deer could go grazing in the fields where cattle have grazed before. Also, roe deer are subject to sporadic migration, but they are generally quite stationary animals. For this reason, if they get out of the perimeters it is due to a population's surplus and it is unlikely that they are going to return.	This might be a risk for the spreading of infectious diseases.	It could be useful to monitor the flow of animals to have a clearer idea of the possible risk of infection.
Do the hunters or the people who come into the farm for any other activities, eat in the area? If so, do they take their waste back home, or do they just leave it there?	Yes, it is possible that they eat a sandwich, or some lunch brought from home. In case of leftovers, they might throw them away in the surrounding nature.	This can be dangerous nowadays, considering how quickly African Swine Fever can spread.	Taking the leftovers back home and throwing them away in the right bins, is a small action that can make a big difference in terms of the spread of infectious disease.
Are there any bins in the area?	There are no bins in the area.	-	This is something that could be easily fixed providing bins with lids that can not be opened by animals.

ropean and national laws even impose some criteria on this, as happens for the prevention of African Swine Fever [20].
Many other aspects could be enhanced in different farms and new tools are needed to design and improve existing structures and new technologies to facilitate the prevention and control of infectious diseases, with an integrated approach that considers as many parts as possible, in accordance with the One Health perspective, respecting human, animal and environmental health. The integration of multiple skills and the active collaboration between breeders and technicians fully responds to the approach suggested by the One Health system, towards the integration of all the actors in the supply chain, from producers, to animals, to the consumer, confirming itself as a driving force of human, environmental, and animal health.

CONCLUSION

This study highlights the importance of implementing robust biosecurity measures in wildlife hunting farms to mitigate the risk of disease transmission between wildlife and domestic animals. Our findings underscore the need for continuous monitoring and improvement of biosecurity practices, which are essential for protecting animal and public health in the Umbria region, as such as other similar lands.
Notably, the interview process significantly increased the awareness of biosecurity in the farm manager, emphasizing the crucial role of education and engagement in enhancing biosecurity measures. This is particularly important in the context of wildlife hunting farms, a type of farm that has not yet been fully considered from a biosecurity perspective. Moreover, the challenges encountered by this hunting farm may be similar to those observed in extensive animal farming systems. Further research and policy development are critical to ensuring the effectiveness of these measures and safeguarding against potential infectious and zoonotic diseases.

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Author contributions
Chiara Poeta: Conceptualization, Methodology, Investigation, Data Curation, Writing - Original Draft; Silva Costarelli: Methodology, Investigation, Data Analysis, Writing - Review & Editing; Alessandro Monacelli: Investigation, Data Analysis, Supervision. Emiliano Lasagna: Data Analysis, Supervision, Writing - Review & Editing; Raffaella Franceschini: Conceptualization, Supervision, Writing - Review & Editing; Maria Luisa Marenzoni: Conceptualization, Methodology, Investigation, Supervision, Writing - Review & Editing.

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