Title

Comparative risk-assessment of spread of highly pathogenic avian influenza H5 viruses spread in French broiler and layer sectors.

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Abstract

Since 2015, French poultry production is threatened almost every year by a reintroduction of highly pathogenic avian influenza H5 viruses. The duck sector was

the most concerned by this crisis but other sectors such as broiler, layer and turkey were also affected by outbreaks. The objective of this work was to assess the risk of highly pathogenic avian influenza H5 virus transmission from one farm to another within the French broiler and layer production network.

This study used the WOAH risk assessment framework. After drawing up a scenario tree of virus transmission from one farm to another, data were collected through a literature review or through experts' elicitation. Three questionnaires were developed according to the experts' field of expertise: avian influenza, broiler and layer sectors. The experts' estimates were combined using a beta distribution weighted by their confidence level. A Monte Carlo iteration process was used to combine the different probabilities of the scenario tree and to assess the transmission risk.

In the broiler sector, the highest transmission probabilities were observed if the exposed farm was an indoor broiler farm and the source a broiler farm (indoor or free-range). The high transmission probability between broiler farm integrated within the same company highlighted the role of integration in this probability. The pathways which generated the highest transmission probabilities between two integrated indoor broiler farms with a good biosecurity level are person movement, transport of feed and manure management. In the layer sector, the highest transmission probabilities were observed if the source farm was a free-range farm and the exposed farm a production farm (indoor or free-range). The pathways with the highest transmission probabilities were egg transport and person movement. The sensitivity analysis showed that the exposed farm's biosecurity had a significant impact on the transmission probability.

Our results provide an insight on the role of each type of farms in the virus spread within the French broiler and layer production sectors and will be useful for the implementation of control measures such as movement restriction or vaccination.

Introduction

Since 2015, France has faced several epizootic waves of highly pathogenic avian influenza (HPAI) viruses [1–3]. The main poultry production sector affected by these waves has been the fattening duck production sector – for the production of duck liver-based delicacies called “foie-gras” [4,5]. During these waves, while the initial introduction of HPAI viruses has likely been due to wild birds [1,6], the large spread of the viruses between poultry farms was linked with the poultry production systems. The structure of the fattening duck sector was one of the main factors facilitating the spread of HPAI [6]. Indeed, in this sector, live birds’ movements occur between the different production stages: rearing, breeding and force-feeding. It has been demonstrated that these movements have played a major role in the spread of HPAI viruses [7,8].

Other poultry production sectors exist in France: the meat sector (broiler, meat duck, turkey, guinea fowl) and layer sector [9], using experts’ elicitation. In the broiler production sector, birds enter the farm as day-old chicks and leave the farm to go to the slaughterhouse. In the layer sector, production is organized in two stages: pullet (from day-old chick to ready-to-lay layers) and layer (from ready-to-lay layers to spent hen). In between these two stages, live bird movements take place to transport ready-to-lay layers from the pullet farm to the layer farm. The same movement pattern is observed for breeders of all species. A live bird movement occurs between

the growth stage farm and the fertile adult stage farm. Even if there are no live bird movements between broiler or layer farms, other avian influenza (AI) virus pathways such, as fomite or human movements, may exist [10]. Identified fomite movements may occur through birds pick-up to slaughter, feed delivery, eggs collection, manure and litter management, dead bird management and shared equipment [11–15]. Movements involving direct contacts between humans and birds are identified as presenting the higher risk of AI virus spread (veterinarian, staff working on multiple premises, company staff, technical advisors etc.) [16,17].

Few risk assessment studies looked at the impact of the integration level (integrated or independent) or farming conditions (indoor vs. free-range) on the risk of AI viruses introduction in a farm [13,15,18]. Moreover, a few modelling studies collected data on the frequency of fomite and human movements between farms [11,14]. Nevertheless, these studies never took a close look at the origin of these movements, i.e. type of farm from which the movement originated.

So far, no study has been conducted on the risk of AI introduction into a chicken farm from another farm within the chicken production sector in France, even though these sectors have also been strongly affected by outbreaks (329 outbreaks in chicken farms during the 2020-22 period) [2]. The aim of this study was to assess the risk of spread of AI virus within two other French poultry production sectors: broiler and layer. This risk assessment study has been conducted considering different types of farms as described in a previous study [9]. These types of farms were defined according to the sector (broiler and layer)), production type (i.e. future breeders, breeders, production), the farming conditions (i.e. indoor and free-range) and the integration level of the farm. For this purpose, this study assesses the risk of transmission from one farm of a defined type to another farm of the same or different

type within the same production sector. It did not consider the transmission risk between farms from different poultry sectors and not the risk of introduction through environmental pathways. The current study focuses on the relative transmission risk, i.e. to identify the type of farms with the highest risk of virus transmission through indirect contacts (human and fomites movements). This study will allow us to identify pathways and farm types most at risk of AI virus transmission.

Materials and methods

Risk assessment model

This study focused the assessment on the risk of introduction described by the WOAH framework [19,20] as the aim of the study was to measure the relative risk of viral spread between different types of farms. We assumed that once the virus is introduced into the flock, the probability of a bird being infected, as well as the risk of exposure, is close to 100%, and we did not assess the consequences of AI spread. The virus introduction through the live bird movement pathway from an infected farm to a free farm was considered very high. Since we have assumed that the introduction of at least one infected bird into a free farm makes the farm infected, our risk assessment study has focused solely on the risk of introduction through indirect pathways within a poultry production network. These pathways were identified in a previous literature scoping review [10]: human with a direct contact with birds; trucks transporting live birds; trucks transporting feeds, manure and dead birds; shared equipment in direct contact with birds (

Figure 1). Two other pathways known to represent a very low risk and thus were not considered: human and shared equipment with no direct contact with birds or manure [11,15,21]. Environmental pathways such as air-born or rodent as mechanical vector

transmission pathways were not assessed as the study was conducted under the hypothesis that these environmental pathways would be similar between all types of farms. Geographical proximity between farms was not considered either.

Figure 1: The transmission pathways considered in this risk-assessment study.

The assessment was conducted for the broiler and layer sectors on several types of farms defined in Table 1 and adapted from the description provided by Hautefeuille et al. [9]. Considering the low number of breeder farms and future layers farms, we did not include the integration level for these types of farms.

Table 1: Listing of the different types of farms studied according to the production type, the farming conditions and the integration level of farms in the French broiler and layer production sector

Farming type	Production type	Integration level
Indoor	Broiler	integrated with an hatchery
		integrated without hatchery
		independent
Free-range	Broiler	integrated with an hatchery
		integrated without hatchery
		independent
Selection	Breeder	-
	Future breeder	-

Data source

The probability of virus introduction from each pathway was calculated using scenario trees as recommended by EFSA [22]. Two scenario trees were built (Figure 2): one to estimate the probability of the non-detection of an infected farm (Figure 2 and Table 2) and one to estimate the probability of viral transmission from the source farm to the exposed farm through a given pathway (Figure 3 and Table 3). The first step of this study was to identify, in the scenario trees, the branches for which estimates of risk probabilities could be obtained from the French legislation. The second step was to generate data for each pathway on the transmission risk, the frequency of occurrence for one type of farm and the probability that this pathway exists between two different types of farms. An expert elicitation survey was conducted to obtain the data not available from the scientific literature.

Figure 2: Scenario tree to assess the risk of not detecting an infected farm

Table 2: Probability that an infected farm (source) is not detected

Node	Parameters estimates	Branch of node	Data sources or justification
Farm routinely tested	Probability that a farm is routinely tested against HPAI	Yes No	French legislation [23]
Time of infection	Estimation of the proportion of farms infected for more or less than 3 days	Less than 3 days More than 3 days	Incubation period [24]

Clinical signs	Probability that a trained farmer detects clinical signs of HPAI infection before 3 days of infection	Yes No	Expert elicitations
	Probability that a trained farmer detects clinical signs of HPAI infection after 3 days of infection	Yes No	Expert elicitations
Farm tested if clinical signs detected	Probability that a farm is tested if clinical signs are detected	Yes No	Expert elicitations
Farm reported if tested positive	Probability that a farm is reported if tested positive	Yes No	French legislation [23]

Figure 3: Scenario tree to assess the risk of virus transmission from an infected farm (source) to a free farm (exposed) through a given pathway.

Table 3: Probability that an infected farm (source farm) not detected transmit the virus to a free farm (exposed farm) for a given pathway

Node	Parameters estimates	Branch of node	Data sources or justification
Biosecurity level of source	Estimation of the proportion of farms of the same type of farms	Very low Low	Expert elicitations

farm (infected farm)	than source farm in the different biosecurity levels	Good High Very High	
Virus release from source farm (infected farm)	Probability that the virus is released from source farm according to the studied pathway	Yes No	Expert elicitations
Link between the source farm and the exposed farm	Probability that the studied pathway occurred in the exposed farm	Yes No	Expert elicitations or production cycle values
	Probability that the studied pathway went during the previous week in the source farm	Yes No	Expert elicitations
Biosecurity level of exposed farm (free of infection)	Estimation of the proportion of farms of the same type of farms than the exposed farm in the different biosecurity levels	Very low Low Good High Very High	Expert elicitations
Virus introduction in the exposed farm	Probability that the virus is introduced in the exposed farm according to the studied pathway	Yes No	Expert elicitations

Fomite (i.e. person, truck or material) disinfection outside farms was considered to be similar for all types of farms and was not sufficient to stop the virus spread.

Once the virus is introduced in the exposed farm (free of infection), it was considered that the probability that the virus enters in contact with birds varies according to the pathway. A farm consists of several areas to meet biosecurity criteria as explained in the introduction of the biosecurity grid: public area for visitors, professional area where only the persons and vehicles authorized to work on the farm can go, and the production unit (i.e. the building where the birds are raised with a possible outdoor area). For all studied pathways, the area into which the virus is introduced (Table 4 and Figure 4) and the probability that a virus is transmitted from one area to another (Table 5) were defined.

Table 4: List of the different studied pathways and farm area of virus introduction according to this pathway

Pathways	Farms concerned	Area of the farm where the virus is introduced
Human with a direct contact with birds	All	Direct contact with birds
Chick transport	Broiler Future breeder	
Birds' pick-up transport (pullet and to slaughter)	All	Professional area
Feed delivery	All	
Manure management	All	
Dead bird management	All	Public area

Shared equipment	Broiler, Layer Breeder	Direct contact with birds
Egg	Layer, Breeder	Professional area

Figure 4: Schematic representation of the different areas composing a broiler farm (based on [25])

Table 5: Probability that a virus coming into contact with birds after introduction into a free farm of a given biosecurity level

Node	Parameters estimates	Branch of node	Data sources or justification
Virus presents in public are	Probability that the virus in introduced in the professional area	Yes No	Expert elicitations
Virus presents in professional area	Probability that the virus in introduced in the production unit	Yes No	Expert elicitations
Virus presents in production unit	Probability that the virus enters in contact with birds	Yes No	Expert elicitations

Experts' elicitation survey

The aim of this survey was to collect data on the transmission risk for each type of farms (8 farm types for broiler and 7 for layer production) and for each pathway (9 different pathways). We decided to create a survey with three questionnaires, each of which was a reasonable length for respondents while still collecting all of the necessary data. Each questionnaire was intended for one type of expert profile: one for experts on AI epidemiology and poultry farm biosecurity, one for experts on French broiler sector and one for experts on French layer sector. In our study, experts were defined as persons with relevant experience on one of the different topics studied.

Experts were recruited according to their experience on the different questionnaire topics. Experts requested at first for the questionnaire on AI were scientists from diverse national French research centres currently working and publishing on AI in France. Experts requested at first for the questionnaire on broiler or layer sector are national representatives of the French broiler or layer sector. Following this initial recruitment, a "snowball" recruitment was conducted, in which experts who participated in the survey were asked to recommend other experts to fill out the questionnaire. Experts filled the survey independently without being in contact with each other. The survey was not anonymous in order to contact the experts for clarification, but all their answers were analysed and presented anonymously.

The questionnaire was designed online using the tool Survey Monkey (www.surveymonkey.com). For each questionnaire a pilot survey was sent to two experts working on the questionnaire's field in order to test the survey and optimize it.

In the aim to reduce the length of each questionnaire, some simplifications were decided and validated with these two experts (Table 6).

Table 6: Simplification made according to the probability and the pathway concerned and justification validated by two experts of the poultry production sector.

Probability concerned	Pathway concerned	Simplification made	Justification
Probability of virus introduction and release	Pullet transport Birds to slaughter transport	Merge of the two pathways as pick-up transport	Pick-up of birds is conducted in a similar way for pullet and birds to slaughter.
Probability that the studied pathway went during the previous week in farm A (i.e. Probability of pathway origin)	Manure Dead birds	Merge of the two pathways	Management similar for both pathways
		No consideration of the integration level of farms	Management similar regardless the integration level
	Shared equipment		

The experts were emailed a description of the study objectives and of what was expected for their participation with the link to the online survey. The beginning of the online survey included instructions and contact information of the administering researchers. The questionnaires are available in French (Supplementary file 1).

Each questionnaire was designed to take about one hour to be filled by experts. Questionnaires started with a description of the experts and questions on their expertise level. The first questionnaire on AI epidemiology was divided in four sections: risk of AI virus introduction in a farm, risk of AI virus release from an infected farm, risk associated with wild birds, and risk of an infected farm going undetected. To simplify the questionnaire, risk was assessed per farm biosecurity level rather than farm type. Grids usually used for biosecurity evaluation process are developed to assess the biosecurity at one farm level in a very detail way and in the aim to improve the biosecurity of this one farm. Thus, it was necessary to developed a specific less detailed biosecurity grid to order farms within one farming type between five biosecurity levels defined from very low to very high. This biosecurity grid was built based on previous works on the biosecurity evaluation process [7,26] and guidelines developed by the French poultry technical institute (ITAVI) [25] (Supplementary file 2). This grid was validated by two experts outside of the list of experts identified for the risk assessment and currently working on French poultry farms biosecurity. For the five defined biosecurity levels, experts were asked about the probability of introduction or release of the virus for each risk pathway (poultry production related or wild birds related). At the end, the experts were asked to define the probability that an infected farm would go undetected. The second questionnaire aimed to collect data on broiler sector and had four sections: biosecurity level of poultry farms, frequency of between farm movements and material, and detection level of AI virus infection. To assess the biosecurity level of a studied farm type, experts were asked to distribute 100 farms of this type between the five different levels in order to have the percentage of farms per biosecurity level (Supplementary file 1). The second section aimed to identify for each introduction pathway and for

each type of farm, the frequency that this pathway occurred and the probability that this pathway links the studied farm with another type of farms. The questionnaire concluded on the capacity of a farmer to detect an AI infection based on the type of farm. For question concerning probabilities of virus introduction or release according to the several pathways, experts were asked for the most probable, the minimum and the maximum values. For question concerning probabilities of pathway occurrence, experts were asked for a range of probabilities (Null: 0%, Negligeable: >0 – 20%, Low: 20-40%, Moderate: 40-60%, High: 60-80% and very high (>80%). For question concerning frequencies, experts were asked to select on possibility from a range between “more than seven times a week” and “less than once every two months”. For each question, experts were asked to provide their confidence level with a score from 1 to 5 (Table 7) and they could add comments. Finally, for the three questionnaires, the experts were asked some open-ended questions regarding factors that might have influenced their answers. All the answers provided by the experts are available in Supplementary file 1.

Table 7: Matching between confidence level and score

Level of confidence	Score
Certain	5
Very confident	4
Moderately confident	3
Not confident	2
Uncertain	1

Data analysis

Data collected from the SurveyMonkey platform were saved in a database template developed in Microsoft Excel® version 2007. The data from these two questionnaires were used to elicit Beta distributions characterising the ensemble of experts' assessments on each probability of interest. For each probability of interest, the parameters of the Beta distribution were determined in terms of expected value and variance of the distribution, which were in turn calculated from the expert's responses as follows. For the question asking for the most probable, minimum and maximum values of the target probabilities, the expected value was set at the average of the most probable values weighted by the expert confidence level and the variance was calculated using a formula (1) balancing a maximal variance V_m and an average variance V_i between experts.

$$var = V_i + \frac{(V_m - V_i)^3}{{V_m}^2} \quad (1)$$

The maximal variance V_m corresponds to the variance of a Uniform distribution with support over the maximal interval (i.e. the smallest interval containing all the intervals reported by the experts). The average variance V_i, by contrast, is the average of the variances of the Uniform distributions over each of the reported intervals. This variance calculation allows the representations of divergent opinions without reducing compatible assessments. For questions requiring only one interval of values (i.e. 21% to 40%, 41% to 60%, etc.), such as questions on between farm contact probabilities, the centre of the interval was considered as the most probable, and the boundaries as the minimum and the maximum value for each expert. The analysis was conducted with R (version 4.0.2).

The frequency of occurrence and the origin of some pathways did not need to be assessed by experts as these values depend on the production cycle. The egg transport was considered to enter into a given breeder farm every day and the possible origin of this truck was always breeder farms. The frequency of chick and pullet delivery and slaughter take off was defined by the production cycle of the different types of production. As it appears that experts could not provide an occurrence frequency for shared equipment, it was arbitrarily considered to occur once a month.

A Monte Carlo iterative process was carried out on the scenario trees described above, in order to combine the different probabilities and to calculate the transmission probability from one type of farm to another. For each iteration, the probability of realisation of the event for all branches of the scenario tree were randomly sampled from the Beta distribution previously elicited and all probabilities were multiplied to obtain the transmission probability. The simulation process used 10 000 iterations.

In addition, we estimated the relative transmission probabilities between farm types with respect to the pair that exhibited the highest mean transmission probability. The target pair of source and exposed farm types was deemed to be at very high transmission risk when the sampled transmission probability was greater than that of the reference pair in 40% or more of the Monte Carlo iterations. For percentages between 25 and 40%, the pair was considered to be at high transmission risk. For percentages between 15 and 25%, 5 and 15% and below 5%, the pair was considered to be at medium, low and very low transmission risk respectively.

Sensitivity analysis

A sensitivity analysis was conducted using the one-at-time method applied on the biosecurity level of the source and the exposed farms and on the pathways [27]. This analysis was applied only for two types of farms with the most extreme daily transmission probabilities: one representing the highest and one the lowest. This sensitivity analysis is based on the distribution of the transmission probabilities on 10 000 iterations.

Results

Experts' description

At the end of the process, six experts responded to the questionnaire on AI epidemiology. Among these experts, three had more than ten years of experience on AI, one more than five years and the last two more than three years. Seven experts responded to the questionnaire on the broiler sector and all of them had more than 10 years of experience on the broiler sector. Five experts responded to the questionnaire on the layer sector and all of them had over 10 years of experience on the layer sector.

Probability of avian influenza virus transmission

The survey results for the broiler production network indicated that the probabilities of transmission were the highest either from indoor integrated production farms (i.e. not selection) to indoor broiler farms (regardless of the integration level), or from indoor

or free-range production farms to independent indoor farms. These farm pairs were considered to be at very high transmission risk (Table 8). The highest mean transmission probability was between independent indoor broiler farms, which served as the reference to conduct the relative comparison. The transmission probabilities were the lowest either when the exposed farm was a selection farm or from selection farms to free-range farms (regardless of the level of integration) (farm pairs at very low transmission risk). High transmission risks were found from free-range or independent indoor farms to integrated indoor farms, and between free-range independent farms. Finally, medium transmission risks were found from production to free-range farms, while transmission risks from selection to indoor farms were found to be low.

For the layer production network, the probabilities of transmission between production farms (pullet, indoor or free-range layer) were found to be the highest (farm pairs at very high transmission risk) (Table 9). The highest mean transmission probability was between independent free-range layer farms, which were used as the reference farms for the relative risk comparison. Transmission probabilities between selection farms were the lowest (very low transmission risk). Finally, transmission risks from production to pullet farms, from pullet to production farms and from indoor to production farms (except for independent indoor farms) were found to be medium.

Table 8: Daily transmission probabilities of HPAI H5 virus between an infected source farm and an exposed farm (free of infection) according to the type of farms in the French broiler production network (median, Q25 and Q75 values of 10 000 iterations). Percentage of transmission probabilities below the transmission probability between independent indoor broiler farms: Dark brown: 50-60% (very high transmission risk), brown: 60-75% (high transmission risk), medium brown: 75-85% (medium transmission risk), light brown: 85-95% (low transmission risk), no colour: over 95% (very low transmission risk).

Values in %		**Exposed farm (free of infection)**							
		Indoor broiler			Free-range broiler			Selection	
Source farm (infected)		Integrated with hatchery	Integrated without hatchery	Independent	Integrated with hatchery	Integrated without hatchery	Independent	Breeder	Future breeder
Indoor broiler	Integrated with hatchery	1.2 [0.5 - 3]	0.8[0.3 - 2]	1.0 [0.4 - 2.5]	0.2 [0.1 - 0.4]	0.2 [0.1 - 0.4]	0.3 [0.1 - 0.6]	0.007 [0.0009 - 0.04]	0.005 [0.0005 - 0.04]
	Integrated without hatchery	0.8 [0.3 - 2]	1.2 [0.5 - 3.1]	1.0 [0.4 - 2.5]	0.2 [0.1 - 0.5]	0.2 [0.1 - 0.5]	0.3 [0.1 - 0.6]	0.007 [0.0009 - 0.04]	0.005 [0.0005 - 0.04]
	Independent	0.6 [0.2 - 1.4]	0.6 [0.2 - 1.4]	1.2 [0.5 - 2.8]	0.2 [0.1 - 0.5]	0.3 [0.1 - 0.5]	0.3 [0. 2 - 0.7]	0.008 [0.001 - 0.05]	0.006 [0.0006 - 0.04]
Free-range	Integrated with hatchery	0.6 [0.2 - 1.5]	0.5 [0.2 - 1.5]	1.0 [0.4 - 2.4]	0.4 [0.2 - 0.8]	0.3 [0.1 - 0.6]	0.4 [0.2 - 0.7]	0.008 [0.001 - 0.05]	0.006 [0.0006 - 0.04]

broiler	Integrated without hatchery	0.5 [0.2 - 1.5]	0.5 [0.2 - 1.5]	1.0 [0.4 - 2.4]	0.3 [0.1 - 0.6]	0.4 [0.2 - 0.8]	0.4 [0.2 - 0.7]	0.007 [0.001 - 0.04]	0.006 [0.0006 - 0.04]
	Independent	0.6 [0.3 - 1.7]	0.7 [0.3 - 1.7]	1.2 [0.5 - 2.8]	0.3 [0.1 - 0.6]	0.3 [0.1 - 0.6]	0.5 [0.2 - 1]	0.009 [0.001 - 0.05]	0.007 [0.0007 - 0.05]
Selection	Breeder	0.05 [0.009 - 0.2]	0.05 [0.009 - 0.2]	0.07 [0.02 - 0.3]	0.01 [0.003 - 0.04]	0.01 [0.003 - 0.04]	0.02 [0.005 - 0.05]	0.006 [0.0007 - 0.04]	0.004 [0.0004 - 0.03]
	Future breeder	0.04 [0.008 - 0.2]	0.04 [0.008 - 0.2]	0.06 [0.01 - 0.3]	0.01 [0.002 - 0.04]	0.01 [0.003 - 0.04]	0.01 [0.004 - 0.04]	0.003 [0.0003 - 0.02]	0.006 [0.0006 - 0.04]

Table 9: Daily transmission probabilities of HPAI H5 virus between an infected source farm and an exposed farm (free of infection) according to the type of farms in the French layer production network (median, Q25 and Q75 values of 10 000 iterations). Percentage of transmission probabilities below the transmission probability between independent free-range layer farms: Dark brown: 50-60% (very high transmission risk), brown: 60-75% (high transmission risk), medium brown: 75-85% (medium transmission risk), light brown: 85-95% (low transmission risk), no colour: over 95% (very low transmission risk)

Values in %		**Exposed farm (free of infection)**						
		Pullet	Indoor layer		Free-range layer		Selection	
			Integrated	Independent	Integrated	Independent	Breeder	Future breeder
Source farm (infected)								
Pullet		0.2 [0.06 - 0.5]	0.1 [0.05 - 0.3]	0.1 [0.05 - 0.4]	0.1 [0.05 - 0.3]	0.1 [0.04 - 0.3]	0.007 [0.001 - 0.03]	0.005 [0.0007 - 0.02]
Indoor layer	Integrated	0.1 [0.04 - 0.4]	0.2 [0.09 - 0.6]	0.2 [0.08 - 0.5]	0.2 [0.09 - 0.6]	0.2 [0.09 - 0.6]	0.007 [0.001 - 0.03]	0.005 [0.0008 - 0.02]
	Independent	0.1 [0.04 - 0.4]	0.2 [0.07 - 0.5]	0.3 [0.1 - 0.7]	0.2 [0.09 - 0.6]	0.2 [0.09 - 0.6]	0.007 [0.001 - 0.03]	0.005 [0.0008 - 0.02]
Free-range layer	Integrated	0.2 [0.06 - 0.6]	0.3 [0.1 - 0.8]	0.4 [0.1 - 0.9]	0.4 [0.2 - 1]	0.4 [0.2 - 0.9]	0.009 [0.002 - 0.04]	0.006 [0.001 - 0.03]
	Independent	0.2 [0.06 - 0.6]	0.3 [0.1 - 0.8]	0.4 [0.1 - 0.9]	0.3 [0.1 - 0.8]	0.4 [0.2 - 1]	0.009 [0.00187 - 0.04]	0.006 [0.0011 - 0.03]
Selection	Breeder	0.005 [0.0008 - 0.02]	0.009 [0.002 - 0.03]	0.009 [0.002 - 0.03]	0.008 [0.002 - 0.03]	0.008 [0.002 - 0.03]	0.006 [0.0008 - 0.04]	0.004 [0.0004 - 0.02]
	Future breeder	0.004 [0.0007 - 0.02]	0.007 [0.002 - 0.03]	0.007 [0.002 - 0.03]	0.006 [0.002 - 0.02]	0.006 [0.002 - 0.02]	0.004 [0.0004 - 0.03]	0.004 [0.0005 - 0.03]

Sensitivity analysis

The sensitivity analysis was conducted on two types of farms: one with the highest transmission probability (integrated indoor broiler farm) (Figure 5 and Figure 6 Figure 5) and one with the lowest (future breeder farm for broiler production) as source (infected) and exposed (free) farms (Supplementary file 3).

The distribution of transmission probabilities for all pathways in both cases highlighted the importance of the farm biosecurity level on transmission probability. Indeed, it showed that the transmission probabilities are high for all pathways when biosecurity levels of both source and exposed farms are low, but have a lower impact when the biosecurity levels are high (Figure 5 Figure 5).

The biosecurity level of the source farm has less influence on the probability of transmission than the biosecurity level of the exposed farm on the transmission probability, as showed by the distribution of transmission probabilities. For all pathways, the variation in the biosecurity level of the source farm (infected) had a low impact on the variation of the transmission probabilities (Figure 6 A). On the other hand, the variation in the biosecurity level of the exposed farm had a significant impact on the variation in transmission probabilities (Figure 6 B). Indeed, transmission probabilities were high in exposed farms with biosecurity levels below (3 on 5), while they were very low in farms with high biosecurity level (above 4), for all pathways except person movement for both farm types and material movement for future breeder farms. The distributions of transmission probabilities were very similar between the two types of farms for all pathways except for the shared equipment. As

their respective total transmission probabilities were different, this result reinforces the impact of biosecurity level on the transmission.

Figure 5: Distribution of the transmission probabilities across all **pathways** according to the biosecurity level of the source farm (x axis) and to the biosecurity level of the exposed farm (colours) with integrated indoor broiler farm as exposed and source farms. Threshold at 1/100 (black), 1/1000 (red) and 1/10 000 (orange).

Figure 6: Distribution of the transmission probabilities (A) across all **biosecurity levels of the exposed farm (free)** according to the pathways (x axis) and to the biosecurity level of the source farm (infected) (colours) and (B) across all **biosecurity level of the source farm (infected)** according to the pathways (x axis) and to the biosecurity level of the exposed farm (free) (colours) with integrated indoor broiler farm as exposed and source farms. Threshold at 1/100 (black), 1/1000 (red) and 1/10 000 (orange).

Farm biosecurity and pathways in the broiler sector

The majority of integrated broiler farms (indoor and free-range) were scored with a good biosecurity level (3 in 5) (53% of indoor and 45% of free range) (Supplementary file 4). Independent indoor broiler farms were scored with a higher biosecurity level (43% at good level) than independent free-range broiler farms (42% at low level). For both indoor and free-range farms, independent broiler farms were estimated to have a lower biosecurity level than integrated farms. The pathways which generated the highest transmission probabilities between two integrated indoor broiler farms with a

good biosecurity level are person movement, transport of feed and manure management (respective median value of 0.23%, 0.03% and 0.03%). The most frequent transmission pathways for integrated broiler farms were person movement (median occurrence frequency of 21%) and transport of feed (median occurrence frequency of 30%). When considering an integrated indoor broiler farm as an exposed farm, the probability of contact through person and feed movement was high if the source farm was an indoor broiler farm integrated in the same farmers' association, medium if the source farm was an indoor broiler farm integrated in another farmers' association and low if the source farm was an independent indoor broiler farm, a free range broiler farm or a selection farm (

Figure 7).

The production farms with the lowest transmission probability were integrated free-range broiler farms as exposed farm even if their biosecurity levels were equivalent to biosecurity levels of integrated indoor broiler farms. The pathways with the highest transmission probability between integrated free-range broiler farm at good biosecurity level were material, feed and manure movement (respectively median value of 0.02% and 0.01%). Person movement had a lower impact on the probability of transmission in free-range farms than in indoor farms. The occurrence frequency of person movement between integrated free-range broiler farms (median value of 62%) was lower than between integrated indoor broiler farms (median value of 78%). For both types of farms (integrated indoor and free-range), transport of birds from hatchery (chicks) or to slaughter and dead bird management were the pathways with the lowest impact on the transmission probabilities. Movement of birds from hatchery and to slaughter had the lowest frequency of occurrence (associated with the production cycle). Dead bird management was considered to have a high frequency

of occurrence, but this pathway is the only one that stops in the public area of the farm.

Figure 7: Probability of pathway occurrence in integrated indoor broiler farm according to the different types of pathways and the different type of farms as pathway source (Broiler I int SA: Indoor farm integrated in the same farmer association, Broiler I int OA: Indoor farm integrated in another farmer association, Broiler I ind: Indoor farm independent, Broiler FR: Free-range broiler farm)

Farm biosecurity and pathways in the layer sector

Indoor layer farms were estimated to have between very good biosecurity level and good biosecurity level. Free-range farms were estimated to have between good biosecurity level. Pullet farms were considered with a good biosecurity level (3 in 5) (50%) (Supplementary file 4).

Due to their production, layer birds transport occurs at a lower frequency than broiler production. Moreover, the pathway with the highest impact on transmission probabilities was egg transport. When comparing the layer production farm with the highest transmission probability (independent free-range layer farms with a low biosecurity level) and the farm with the lowest transmission probability (integrated indoor layer farms with a good biosecurity level), the pathways with the highest transmission probabilities were for both egg transport, person movement and shared equipment. Nonetheless, independent free-range farms had higher transmission probabilities than integrated indoor farms (median transmission probability for egg transport of 0.09% and 0.59% respectively). According to the five experts of the layer

sector, the transport of eggs was the pathway with the highest frequency of occurrence for both indoor and free-range layer farms (median values of 45% and 30% respectively). Person movements and shared equipment were just as frequent as the other pathways but they are both associated with direct contact with birds.

Farm biosecurity and pathways in selection farms

For both broiler and layer sectors, experts considered the biosecurity levels of selection farms to be high to very high (4 to 5 out of 5) (Supplementary file 4), implying very low transmission probabilities, except for person movement. As with future breeder farms (Supplementary file 3 Figure 3), shared equipment has no impact on transmission probability. If the results for breeder farms in both productions were similar, eggs transport and manure management appeared to have higher transmission probabilities in broiler production than in layer production. If the transmission probabilities are very low at high and very high biosecurity levels, they are closed to those observed in production farm for person movement, transport of feed and eggs, and manure management if the biosecurity level of the breeder farms reached a good level (3 in 5).

Discussion

This study analyzed the risk of virus transmission within French production networks, i.e. the risk that a virus released by an infected farm is introduced into a free farm by all possible indirect pathways and including all the different types of farms identified in the network analysis. Most assessments of the risk of avian influenza (AI) virus being introduced into a poultry farm have been based on risk factors related to the

production network and to wild birds [28,29]. Several assessments have been conducted on the risk of introduction or release of AI viruses in farms engaged in different types of production [14,15,18,21], and a few of these evaluated the risk of virus transmission between farms depending on the type of farm [18,30]. However, none considered the level of integration of the farms in the network and of the risk of virus transmission from one type of farm to another (e.g. from an indoor broiler farm to a breeder farm).

This study was based on a risk assessment method using scenario trees and data from the literature and experts' elicitation when data were not available. The experts' elicitation was inspired by the Delphi method even though the method was not used entirely, as the objective of the Delphi method is to reach a consensus between different experts whereas in the present study, the difference of opinion among experts was used to estimate uncertainty about transmission probabilities. Hence, our study is subject to potential bias. Six different tools were used to limit bias in this study: (1) different questionnaires to segment questions, (2) the level of confidence for each question provided by the experts and included in the statistical analysis, (3) the possibility for the experts to not answer questions, (4) a comment section for each group of questions in which experts could comment on their response, (5) a guide to help the experts understand the questions and (6) a biosecurity grid to ensure the experts had the same level of understanding.

The epidemiological situation concerning avian influenza in France and Europe differed between 2020 and 2025. Between 2021-22 and 2022-23, the number of outbreaks increased drastically and led to the culling of a large number of birds worldwide [31]. France started to vaccinate against HPAI viruses in October 2023, previous to this date, this was not an option for the control of bird flu. The increase in

the number of outbreaks mainly occurred in the duck sector and vaccination was consequently conducted in this sector. The avian flu situation in the broiler and layer sectors has not changed significantly since 2020. Farmers and practising veterinarians may be more sensitive to the disease, even if they were already aware of the threat in 2020 because France had already faced epizootics in 2015-16 and 2017-18. Repeating the same use of experts' elicitation in 2025 would almost certainly produce very similar results to those obtained in 2020.

One of the original features of the present study is the inclusion of biosecurity levels in the risk assessment process. Biosecurity classifications developed for poultry farms generally provide a farm-level biosecurity score [26,32,33], while few studies have assessed the level of biosecurity based on the type of farm (breeder, broiler or layer) [34] or on the level of farm integration in the network [35,36] and, to date, none have used this classification to assess the risk of disease transmission within a given farm type. Our description of biosecurity levels was largely based on previous work done in France [7,25] and was adapted and validated in collaboration with poultry farm biosecurity specialists in France. The biosecurity criteria are illustrated in a schematic diagram (Supplementary file 2) that was shown to the experts who expressed no difficulty in understanding the grid and the biosecurity classification. Using an approach based on farm biosecurity enabled us to study a large number of farm types. The risk assessment applied to biosecurity can now be used to assess the risk of virus transmission between farms in any other French poultry production network. Likewise, the biosecurity grid can now be used to assess the biosecurity level of any type of French poultry farm.

The farm biosecurity level of an exposed farm (i.e. a farm that is free of infection) has a major impact on the risk of transmission of the AI virus between farms regardless of

the pathway and of the biosecurity level of the source farm (i.e. a farm that is already infected). The importance of farm biosecurity in the risk of introduction of the AI virus has been highlighted in several studies [37–40]. In the present study, farms dedicated to producing birds destined for reproduction (future breeders and breeder farms) mainly received a score of 5, indicating very high biosecurity, while production farms were scored 3, indicating good biosecurity. Fewer than 5% of the production farms were considered to have a very high level of biosecurity, in line with the results of a study conducted in New Zealand [32]. Another study in South Korea that measured the risk of HPAI for farms linked by livestock transport vehicles reported a low risk for 73% of breeder chicken farms [18]. In the present study both free-range and indoor broiler farms that belong to the network were judged to have a generally good biosecurity level. Indoor layer farms were judged to have a higher biosecurity (level 3 (good) to level 4 (high)) than free-range layer farms (level 2 (low) to level 3 (good)). As biosecurity measures to limit contact between domestic birds and wild birds were included in the biosecurity grid used to assess the farm biosecurity level, the impact of being free-range was included in the assessment. This result is interesting because risk assessment studies in other countries have shown that free-range farms are more at risk of the virus being introduced by wild birds than indoor farms [29,41]. Wild birds are known to play a role in the introduction and long-distance spread of HPAI viruses, but their role in short distance dissemination is reported to be limited [42,43]. In the present study the proportion of broiler farms with a low biosecurity level (level 2) was higher among independent farms (40% of indoor and 42% of free-range) than in farms that belong to the network (15% of indoor and 22% of free-range). This result is difficult to compare with that of other studies because the studies concerned were conducted in countries with very different

production contexts (Indonesia and Thailand) [35,36]. Moreover, the differences observed need to be interpreted with caution as expert confidence was higher for farms that belong to the network (mean of 4.2 and 3.9 out of 5 for indoor and free-range farms respectively) than for independent farms (mean of 3 and 3.4 out of 5 for indoor and free-range farms respectively). Similar results were observed for independent free-range layer farms but not for independent indoor layer farms for which the proportion of farms with a high biosecurity level (mean value of 43%) was higher than the proportion with a good biosecurity level (mean value of 37%). However, only three experts out of five provided data for this type of farm (mean confidence of 3.7 out of 5) illustrating the lack of knowledge concerning this minority farm type. As the present study was based on expert opinion, it highlights the need for more detailed data on farm biosecurity. Another study carried out in France provided information on biosecurity as a function of farm type (including broiler and layer) and location, but did not investigate independent farms [44]. Moreover, in the present study the biosecurity level assessment was conducted at the national level and did not consider the spatial heterogeneity of farms in France.

In the broiler sector, our results showed that the risk of transmission was higher when exposed farms (free of infection) were indoor farms compared to other types of farms. This is mainly due to the fact that transmission routes occur more frequently in this type of farms than on free-range farms, as previously identified [34]. The risk of transmission was highest between broiler farms that belong to the same farmers' association. The probability of contact via such a pathway was highest between such farms, particularly for people in direct contact with birds, and people involved in feed truck pathways. This result is easily explained by the structure of the poultry network in France. Indeed, poultry farmers' associations are based on a feed manufacturer

and technicians, and veterinarians are often employed by the farmers' association. The role of the poultry network in the spread of AI between farms was already highlighted in previous studies [11,13,16]. The frequency of contact was higher between farms of the same type (i.e. indoor, free-range and breeder) than between farms of different types. According to the experts, fewer contacts occurred between farms that are members of an association or cooperative and independent farms, especially indoor broiler farms. In the layer sector, the probability of transmission was higher for free-range layer farms as source (farms that are already infected), linked to the biosecurity level of the farm. This result confirms that egg transport plays a major role in the probability of transmission [18,30]. Even if breeder farms have a very high level of biosecurity, the existence of many of pathways between breeder farms increases? the frequent passages between breeder farms increase? the risk of transmission of the virus. Indeed, our results show that if the biosecurity level of a breeder farm that is an exposed farm is only 'good', the probability of transmission is similar to that of production farms. The risk of transmission in breeder farms was confirmed by previous studies [34,37]. A study of HPAI H5 outbreaks in France between 2020 and 2022 showed that at least 9% of outbreaks occurred in a breeder farm [2]. The aim of the present risk assessment study was to investigate all the links between all types of farm in French broiler and layer production networks to identify the type of farms and links associated with the highest risk of AI transmission. The links we studied only concerned indirect transmission pathways (i.e., humans or fomites) and not direct transmission pathways (i.e., live domestic birds). Live domestic bird movement is known to present high risk of virus transmission, especially in the duck sector where living ducks are moved at different stages of growth [8]. However, this particular pathway is absent in broiler farms and only used

at very infrequent intervals in layer and breeder farms (about once every 12-13 months), which is why the present study focused on indirect contact, which is much more likely to occur during an AI epidemic. In the present study, we considered that management of fomites (i.e. a person, truck or equipment) outside farms is similar in all types of farms and is not sufficient to affect viral transmission. Disinfection of fomites inside the farm is included in the biosecurity classification. If the impact of fomite disinfection outside farms is the same for all types of farms, this impact will not affect relative risk. Nonetheless, a good cleaning and disinfection process at the truck level would considerably reduce the risk of transmission between farms. Studies conducted in France detected the presence of the viral genome in half (in 2017) and in one third (in 2021) of trucks tested after disinfection [45,46].

The movement of people was found to be the pathway with the highest transmission probabilities. This pathway is associated with direct contact with birds and with a high frequency of occurrence. The other pathways including direct contact with potentially infectious substances such as manure or dead birds or shared equipment that has been in direct contact with birds were also associated with high transmission probabilities. Similar results were obtained in previous studies [11,15,21]. The transport of feed was also associated with high transmission probabilities. The frequency of occurrence of shared equipment was set at once a month for all farms, as this occurrence does not depend on the type of farm but on management and location of the farm. This choice was validated by a poultry production expert and have an impact on the relative role of shared equipment in the viral transmission compared to other pathways, especially in farms with a low biosecurity level. But it does not on the comparison of the viral transmission between the different types of farms. Our study focused on the transmission pathways that were described as being

the most at risk of transmission in the literature [10], but other transmission pathways, such as wind or vermin (i.e. mechanical vectors such as rodents or insects) are also mentioned in the literature. Air-borne transmission has been found to have a limited role in AI virus transmission in France [47], even though this type of transmission is possible over short distances, as some virus genome was found in samples of air collected in infected farms [48]. Dust also appears to play a role in viral dispersal [49]. Other studies conducted in the USA suggest that airborne transmission might play a role in the spread of AI viruses [50,51]. Virus transmission through the movement of vermin between farms is possible but difficult to distinguish from other environmental factors [28,30,52]. These last pathways can be included in an environmental transmission pathway mainly due to close proximity between farms. High poultry density is known to have an impact in the spread of AI viruses [53]. Our study compared the relative risk of transmission from one farm to another under the hypothesis that environmental transmission occurs in the same way for all types of farms regardless of their level of biosecurity. Environmental transmission could have an impact on short distance spread and should thus be included in studies integrating geographical information, which was not the case in the present work.

Finally, this work provides a complete picture of the risk of AI viruses spreading within French broiler and layer production networks including the risk of transmission of the virus between any type of farms. To mitigate this risk, management measures should focus on biosecurity, especially any movements that enable entry into the farm. Careful attention should be paid to movements that occur very frequently, including the movement of people and the transport of feed, manure and dead birds. Network managers should be aware of the increased risk between farms belonging

to the same association and take the necessary actions, such as cleaning and disinfection, between visits to farms, to reduce this risk. The typology of farms based on the transmission risk developed for this study will be useful for the implementation of control measures such as restriction of movement or vaccination.

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Conflict of interest disclosure

The authors declare that they have no financial conflict of interest with the content of this article.

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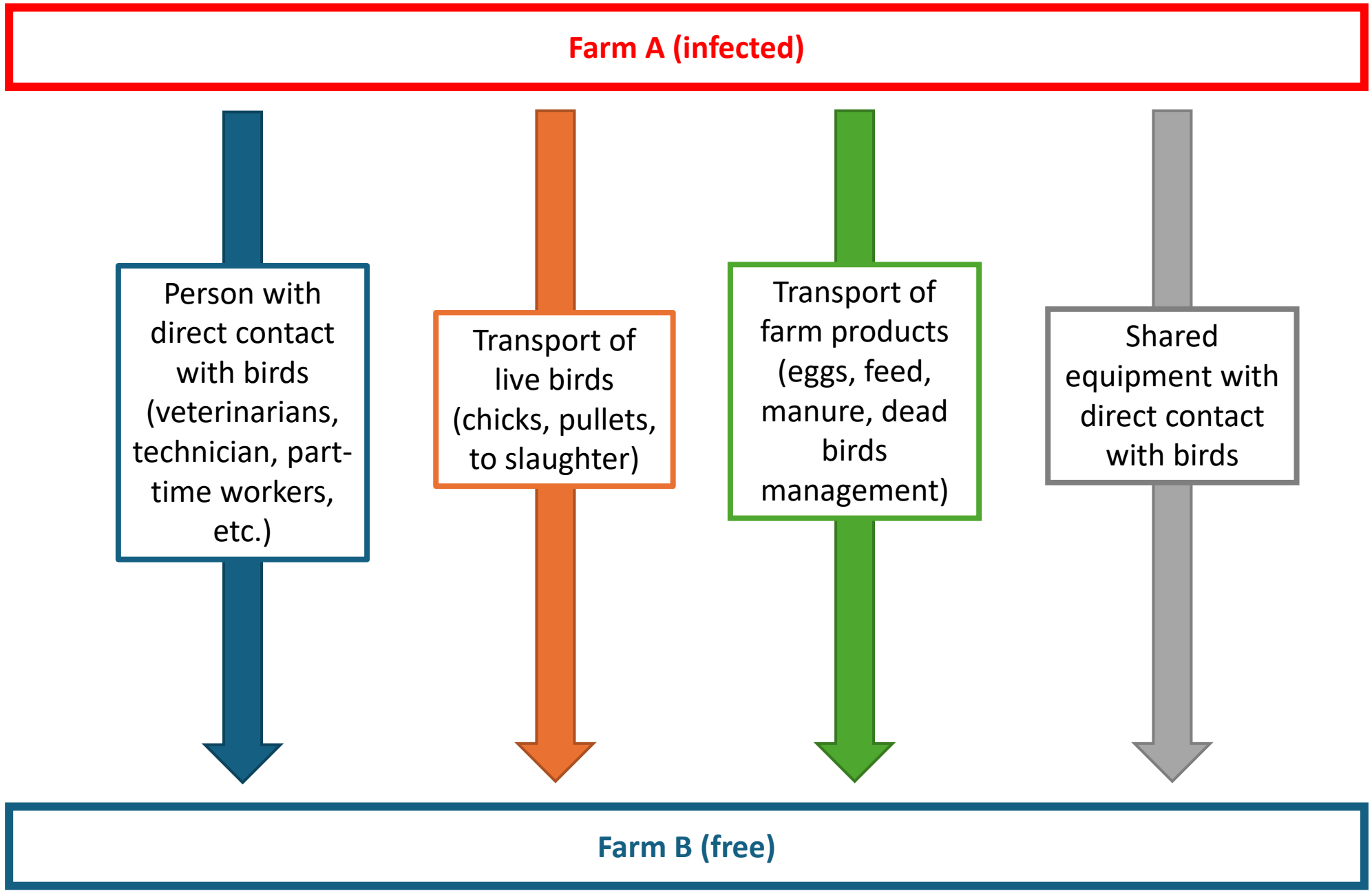
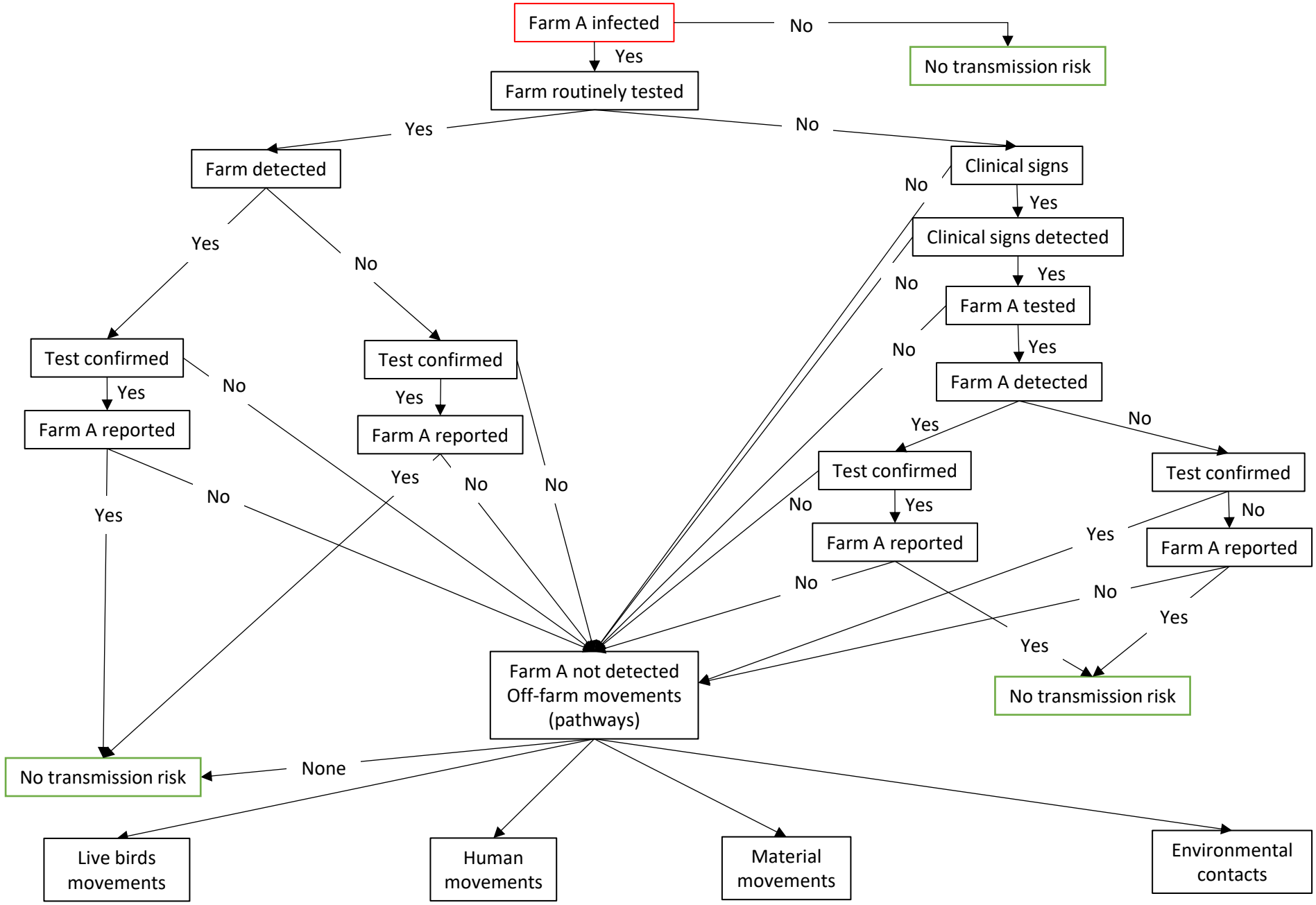
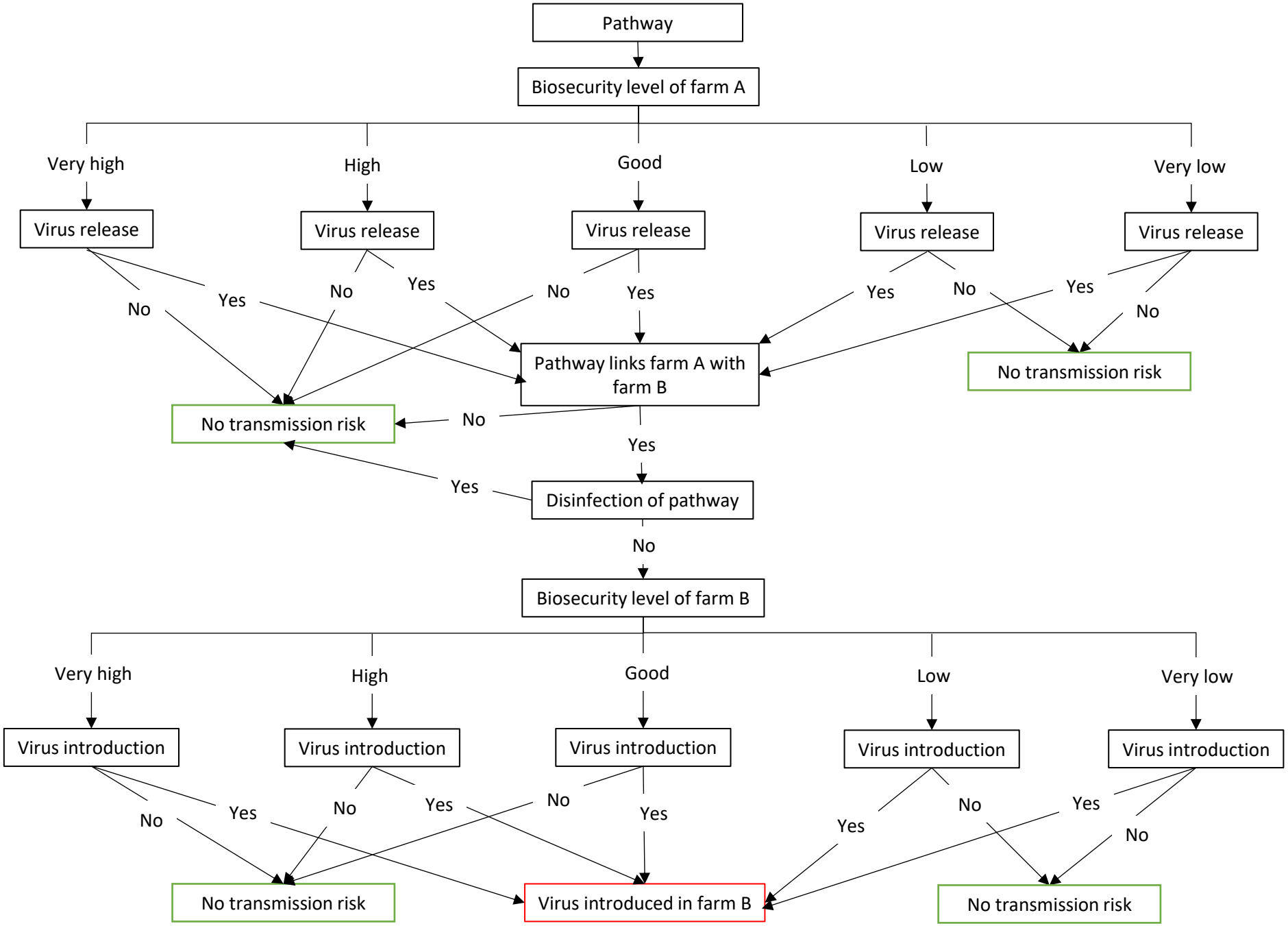
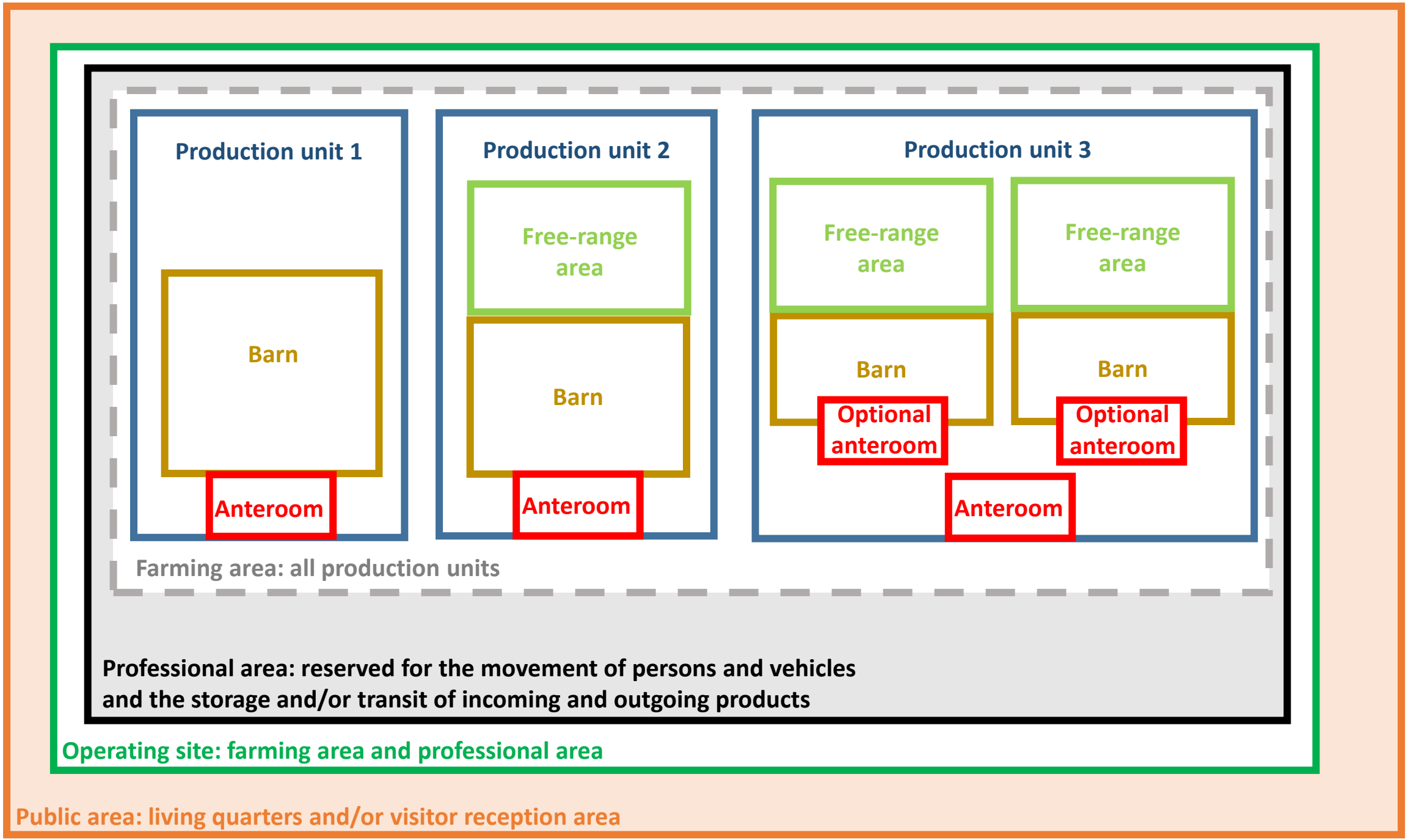
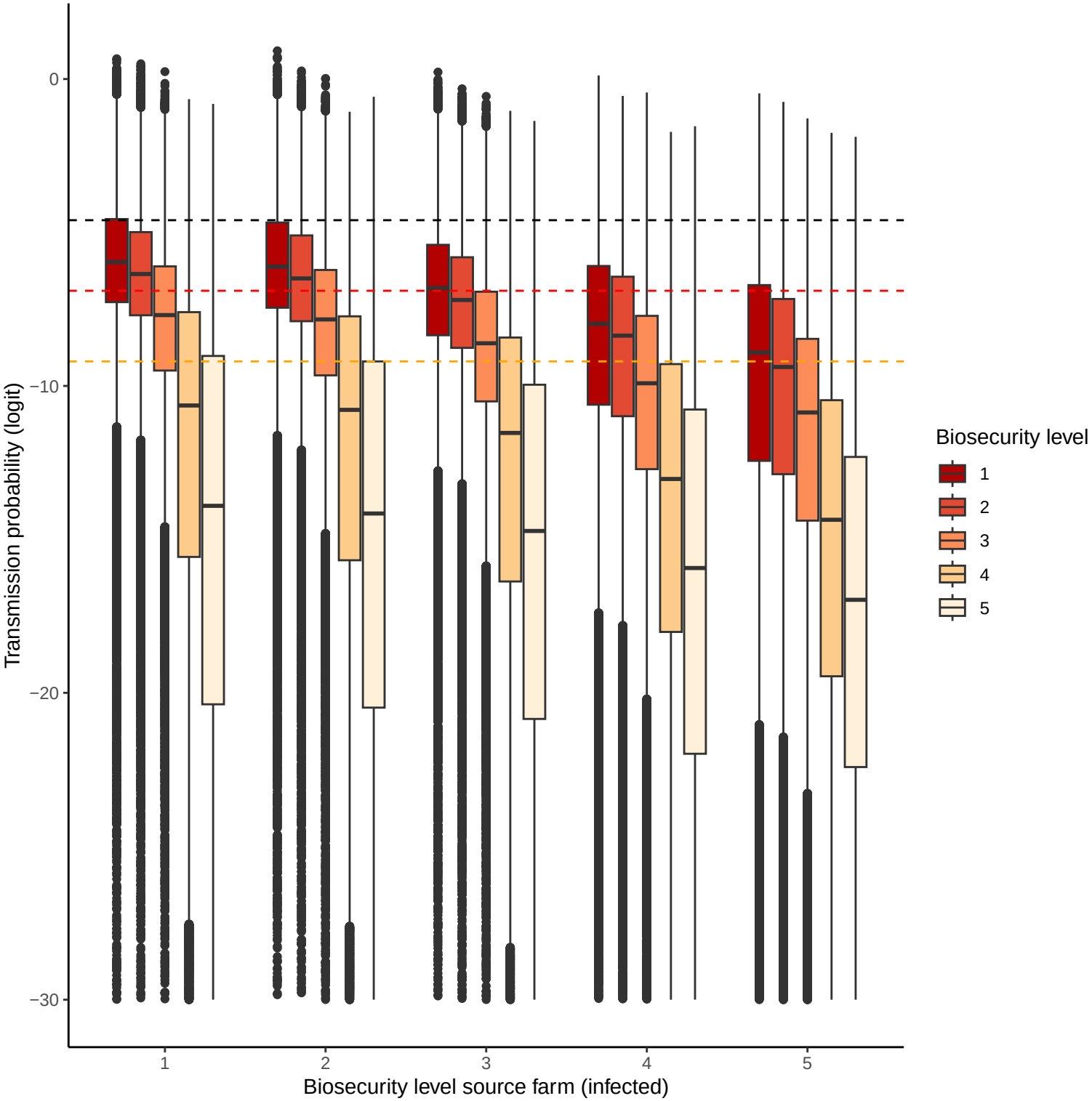

Farm A (infected)
Person with direct contact with birds (veterinarians, technician, part-time workers, etc.)
Transport of live birds (chicks, pullets, to slaughter)
Transport of farm products (eggs, feed, manure, dead birds management)
Shared equipment with direct contact with birds
Farm B (free)

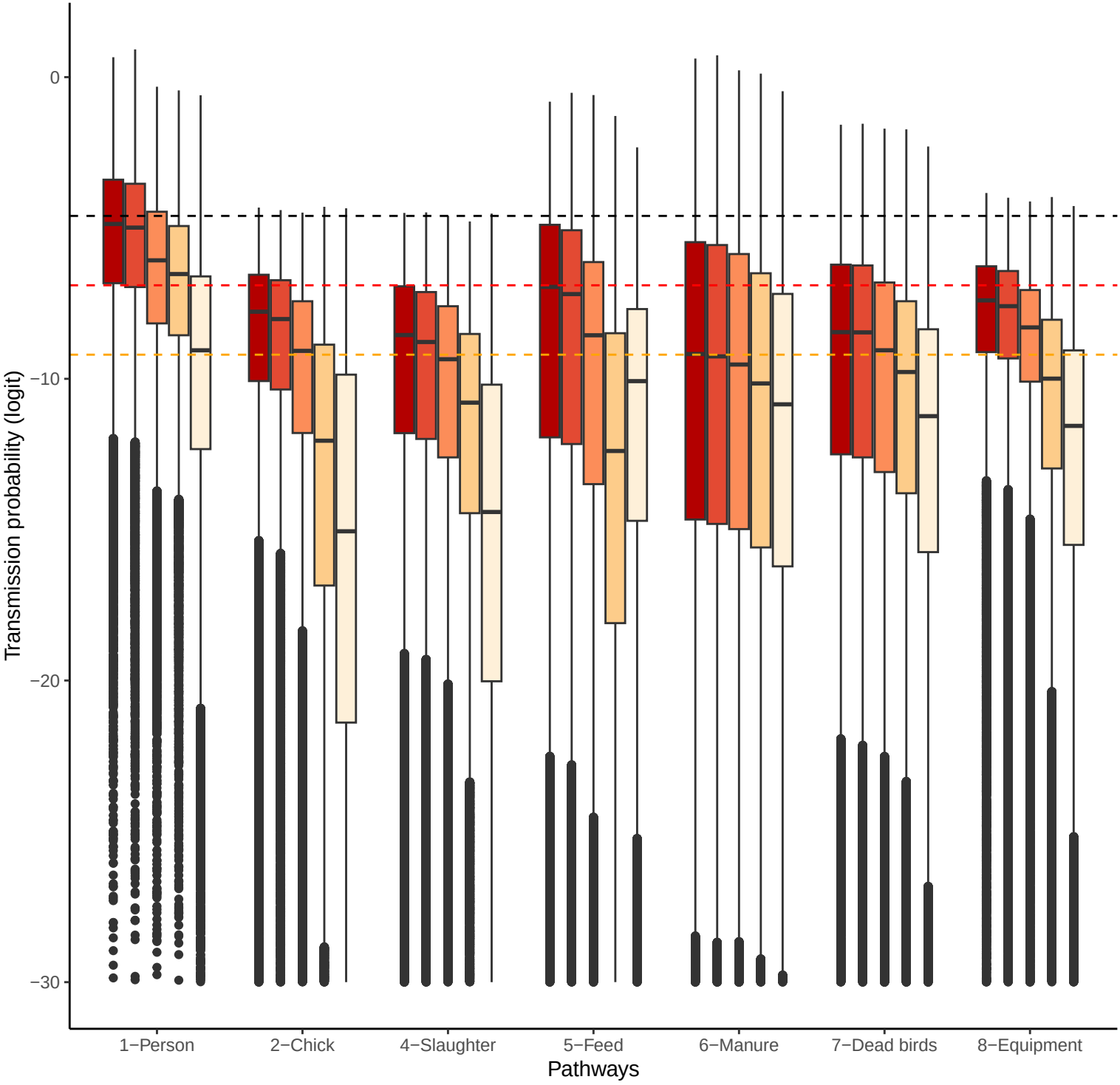

Farm A infected
No
No transmission risk
Yes
Farm routinely tested
Yes
No
Farm detected
Clinical signs
No
Yes
Clinical signs detected
Yes
No
Farm A tested
Yes
No
Farm A detected
Yes
No
Test confirmed
Test confirmed
Test confirmed
Yes
No
Yes
Farm A reported
Farm A reported
Farm A reported
Farm A reported
Yes
No
No
Yes
No
Yes
No
Yes
No
Yes
No
Yes
No transmission risk
Farm A not detected
Off-farm movements
(pathways)
No transmission risk
None
Live birds
movements
Human
movements
Material
movements
Environmental
contacts


Pathway
Biosecurity level of farm A
Very high
High
Good
Low
Very low
Virus release
Virus release
Virus release
Virus release
Virus release
No
Yes
No
Yes
No
Yes
Yes
No
Yes
No
Pathway links farm A with farm B
No transmission risk
No transmission risk
No
Yes
Yes
Disinfection of pathway
No
Biosecurity level of farm B
Very high
High
Good
Low
Very low
Virus introduction
Virus introduction
Virus introduction
Virus introduction
Virus introduction
No
Yes
No
Yes
No
Yes
Yes
No
Yes
No
No transmission risk
Virus introduced in farm B
No transmission risk

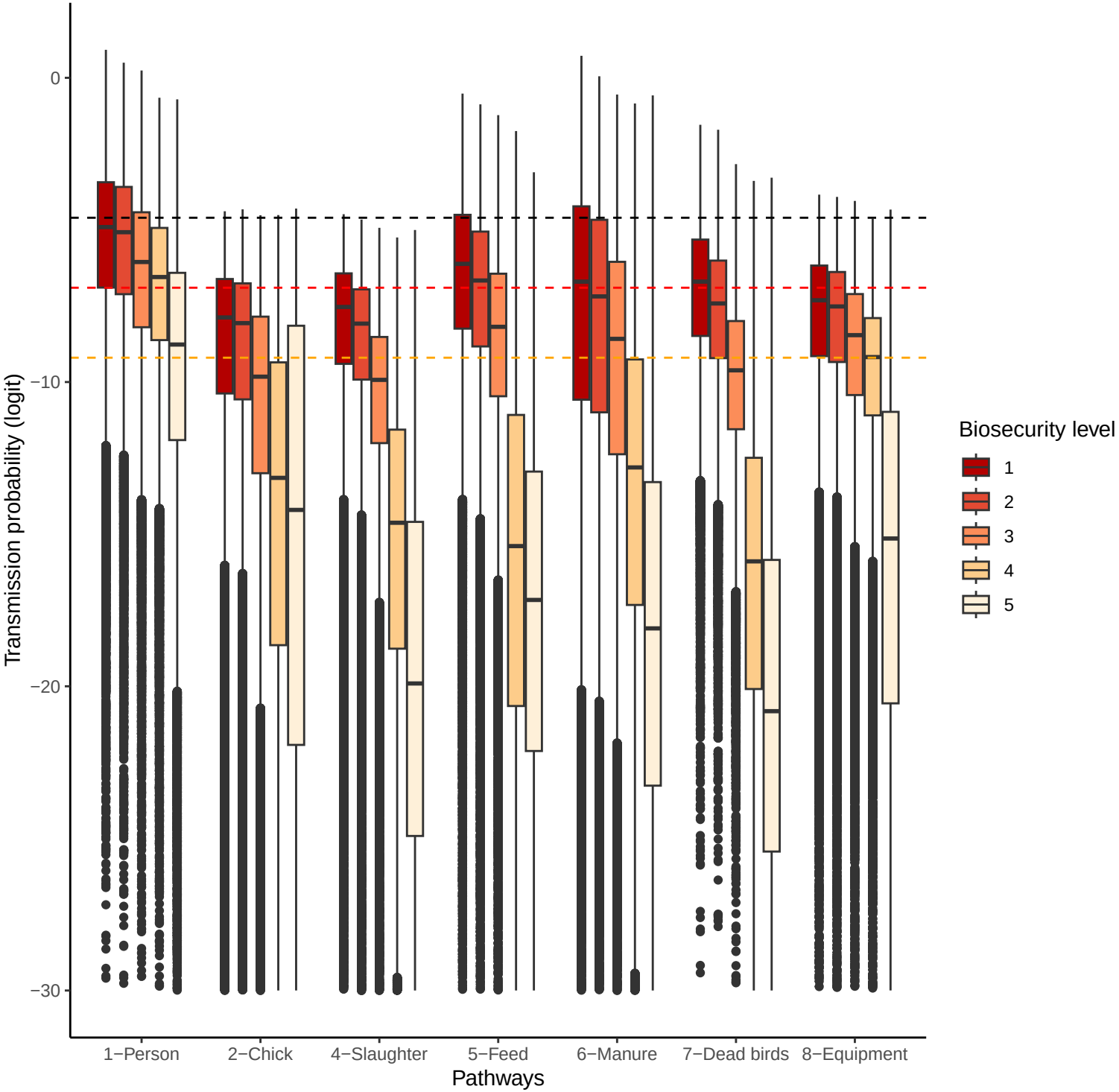

Production unit 1
Barn
Anteroom
Production unit 2
Free-range area
Barn
Anteroom
Production unit 3
Free-range area
Free-range area
Barn
Barn
Optional anteroom
Optional anteroom
Anteroom
Farming area: all production units
Professional area: reserved for the movement of persons and vehicles and the storage and/or transit of incoming and outgoing products
Operating site: farming area and professional area
Public area: living quarters and/or visitor reception area

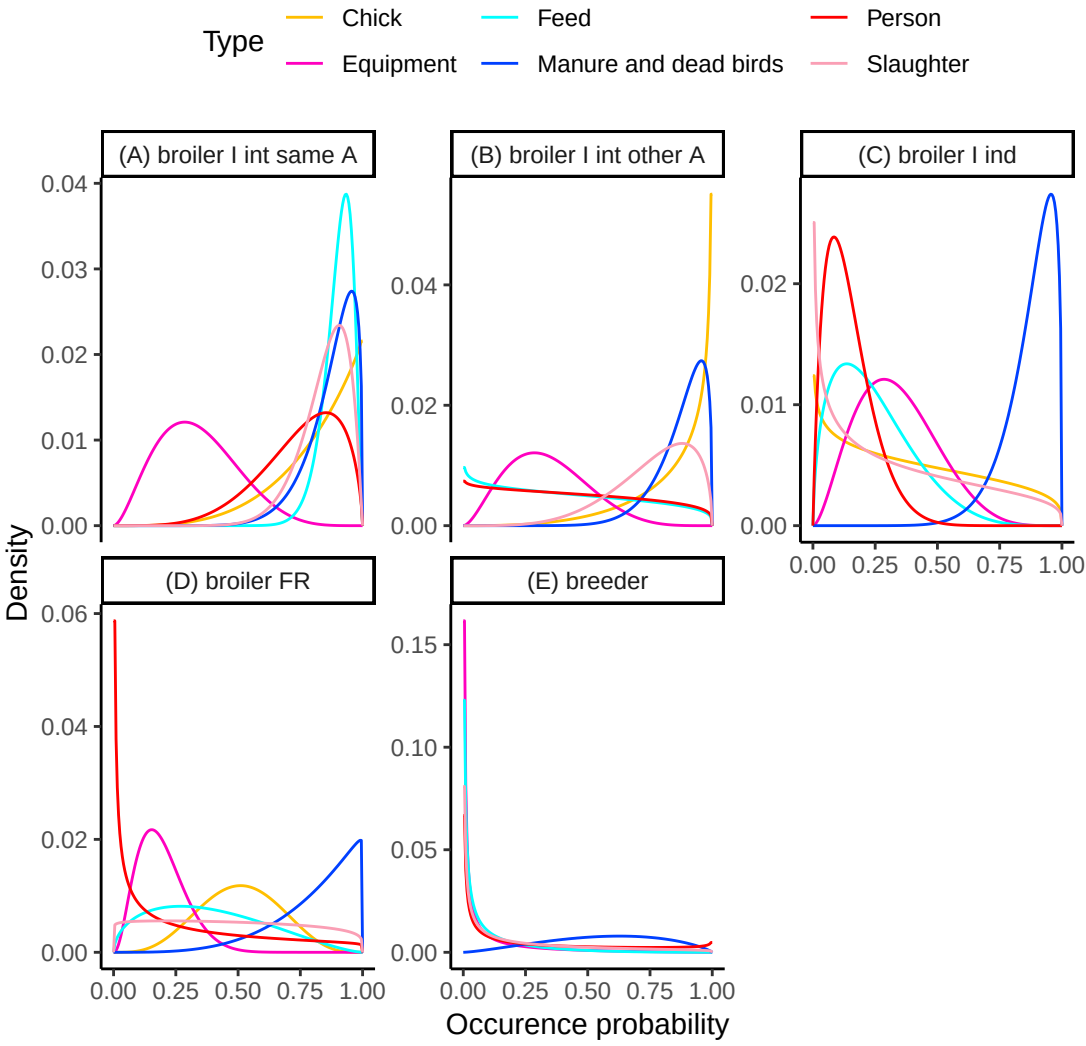

0
−10
−20
−30
Transmission probability (logit)
1
2
3
4
5
Biosecurity level source farm (infected)
Biosecurity level
1
2
3
4
5

(A) Variation on biosecurity levels of the exposed farm (free)



(B) Variation on biosecurity levels of the source farm (infected)




Type
Chick
Feed
Person
Equipment
Manure and dead birds
Slaughter
(A) broiler I int same A
(B) broiler I int other A
(C) broiler I ind
(D) broiler FR
(E) breeder
Density
Occurence probability
0.00
0.01
0.02
0.03
0.04
0.05
0.06
0.10
0.15
0.25
0.50
0.75
1.00