EURCAW Poultry SFA

French Reference Centre for Animal Welfare

Depopulation in case of Avian Influenza

Efficacy and welfare consequences of the depopulation methods used in the EU

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Contents

1 Executive summary

This report details the operating procedures of the ten most used depopulation methods in the European Union in case of Avian Influenza, with specific information related to their welfare consequences and efficiency in terms of unconsciousness and death induction. Results are based on answers received to method-specific surveys that were sent out to the competent authorities of the different Member States, and insights obtained from a workshop with depopulation experts. Here are the key findings:

- **Containerized gassing:** various operating procedures are used to perform containerized gassing – owing to different types of existing containers, gas mixtures and filling techniques. The method appears to be efficient but concerns arise related to the necessity of animal handling, and the inability to observe the birds during the procedure due to fogging.
- **House gassing without foam:** two main procedures exist, namely whole-house and partial-house gassing – where only a portion of the house is gassed. Most often, the gas mixture of choice is CO2 only. The method appears to be efficient. Points of caution relate to the filling speed to reach the target concentration (which must be kept as short as possible to minimize exposure to aversive gas concentration) and the difficulty to assess poultry unconsciousness and welfare due to fogging.
- **Lethal injection:** this method is often performed on small flocks of birds, using either Pentobarbital Sodium or T61. Both drugs are mostly injected intraperitoneally or intrapulmonary – rather than intravenously.
- **Cervical dislocation:** this method is performed on small flocks, either manually or using a mechanical equipment. Welfare concerns focus on animal handling and restraint prior to dislocation, and the need for adequate operator training to ensure the efficiency of the procedure.
- **Captive bolt guns:** this method is performed on small flocks of turkeys and ducks, mostly using non-penetrative captive bolt guns. Non-penetrative guns are reported to be efficient. Welfare concerns include the need (and potential difficulty with) animal handling and restraining.

Limited information was retrieved regarding **percussive blow to the head**, **electrical waterbath**, **head-to-body electrical stunning**, **head-only electrical stunning** and **decapitation**.

2 Glossary

Depopulation method	Explanatory variables
Containerized gassing	Birds are killed outside the house by gassing them in containers (EFSA, 2019)
House gassing without foam	Administration of different gases such as CO_2 or gas mixtures into the house – whether the whole house or only part of it is gassed
Lethal injection	Birds are injected with a lethal dose of verterinary medicines (Council regulation, 2009)
Cervical dislocation	The stretching and twist of the neck provoking cerebral ischemia (Council regulation, 2009)
Captive bolt	Birds are individually shot using a bolt that can either be penetrative or non-penetrative and powered by cartridge, compressed air or spring-loaded
Blow to the head	The head of the bird is placed on a hard surface and a blow to the back of the head is delivered with a hard object (e.g. metal pipe, solid wooden stick, bat…) (European Commission, 2018)
Electrical waterbath	The entire body of the bird is exposed to a current generating a generalised epileptic form on the EEG and possibly the fibrillation or the stopping of the heart through a waterbath (Council regulation, 2009)
Head-to-body electrical Stunning	An electrical current is applied using dry electrodes that span the brain and the heart (EFSA, 2019)
Head-only electrical Stunning	An electrical current is applied using two electrodes on either side of the bird's head – such that they span the brain (EFSA, 2019)
Decapitation	Separation of the head from the body by severing the neck, at the level of the first vertebra (EFSA, 2019)fEFSA

3 Introduction

The EURCAW-Poultry-SFA and the French Reference Centre for Animal Welfare have initiated in January 2023 a work aiming at elaborating recommendations on the types of on-farm killing methods to adopt to ensure the most humane and effective killing of poultry during depopulation in case of Avian Influenza. In January 2024, a preliminary report (accessible here) was published to give a general description of the 10 most used depopulation methods in the EU – together with early feedbacks on their welfare issues and efficiency in terms of unconsciousness and death induction.

The present report provides a more detailed and comprehensive description of depopulation processes. It describes the diversity of existing procedures in the application of each of the 10 depopulation methods, and provides specific information regarding their efficiency and welfare consequences.

4 Methods

Method-specific surveys were developed for each of the 10 most used depopulation methods in the EU; and sent out in November 2023 to the MS (Member States) who reported having used said-methods since 2018. The MS who had not specified the methods used in their country received all method-specific surveys. The competent authorities of all MS were given the opportunity to forward the surveys to one or more national experts – which could aggregate (or not) their own response into a single reply. The results obtained and presented in this report thus accurately reflects the field experience of the respondents. Whether this experience is, however, representative of the depopulation procedures used nationwide remains unknown.

When answering the survey, experts were authorized to pass or answer "unknown" for all questions (except the first three questions concerning personal information). In each method-specific survey, some questions were asked multiple times according to different explanatory variables like the species at stake (*Table 1*). Of note, those species included Gallus *gallus* (laying hens and broilers), Ducks and Turkeys for all surveys – except for lethal injection for which Turkeys could not be included due to technical limitations related to Sphinx programming.

The first aim of the surveys was to gather, for each depopulation method, additional information regarding its context of application (e.g., species, housing system) and established operating procedures to be able to provide a more detailed description of its on-field use. The second aim was to collect information regarding the effectiveness (e.g., failure rate) and associated welfare consequences (e.g., cold stress) of each depopulation method. Surveys were closed in January 2024, and data was extracted from Sphinx (Sphinx iQ3 - v8.2.2 version, Le Sphinx Développement) to be compiled in Excel (Microsoft® Excel 2019).

After analyzing the results obtained from the survey, a four-hour workshop was organized on the 20th of June 2024 by the EURCAW-Poultry-SFA with 42 depopulation experts from 13 MS. One of the main objectives of the workshop was to clarify answers given in response to the survey concerning the operating procedures and the welfare consequences. Clarifications were obtained by both oral interventions and the use of Mentimeter (an online audience response system tool). Information received during the workshop are written in violet in the results section below.

Table 1: Overview of the number of MS contacted to answer each method-specific survey (i.e., the MS who reported having used said-method since 2018) together with the chosen explanatory variables

Depopulation method	MS contacted (number)	Explanatory variables		
Containerized gassing	17	Species	Gassing mixture	Container type
House gassing without foam	13	Species	Gassing mixture	
Lethal injection	14	Species	Drug used	Route of injection
Cervical dislocation	15	Species	Type of Cervical Dislocation	
Captive bolt	5	Species	Type of Captive Bolt	Killing method used (if applicable)
Blow to the head	3	Species	Killing method used (if applicable)	
Electrical waterbath	4	Species	Killing method used (if applicable)	
Head-to-body electrical Stunning	2	Species	Number of electric shocks	
Head-only electrical Stunning	2	Species	Killing method used (if applicable)	
Decapitation	1	Species		

5 Data cleaning

Answers from experts that replied to 4 or less questions within one survey were not included in the analysis, which led to the exclusion of answers from two experts in the survey related to House Gassing. Incoherent answers were not considered, except when clarifications were provided by the respondent. Qualitative answers to open questions relative to the main welfare issues encountered were clustered into different categories based on the subjects mentioned. As such, answers to the question *"Please specify how long (in hours) before the start of the containerized gassing is the feed (/water) being removed"* which reported removing feed or water *"immediately"* or *"just before"* the procedure were clustered under the category *"<1h"*. When quantitative answers were expected, textual answers were converted into numerical values of the same unit.

6 Data analysis

The following analysis will be exclusively descriptive, since the data collected cannot be extrapolated nor generalized in order to compare different correlated variables.

7 Structure of the report

For all depopulation methods, the results are presented as follow:

- Results related to the depopulation context in which the method is reported to be used
- Results related to the specific operating procedures used
- Results related to the animal welfare implications
- Results related to the efficiency of the method

At the end of depopulation section, a discussion/conclusion is provided to summarize and put into perspective the main results obtained by confronting them to current scientific knowledge.

For each category of question (e.g., "*how much time does [gas mixture] takes to kill [species]*?"), the number of answers given by the experts is specified and referred to as **n_{ans}**. Some questions were subdivided according to the explanatory variables (e.g., the same question was asked for different species), which sometimes led us to present grouped results (e.g., whichever the species at stake). Grouping levels of one or more explanatory variables often led to a number of answers superior to the number of experts, as one expert could give an answer for each level of the explanatory variables (e.g. one expert gives two answers to the same question, one for Gallus gallus and one for ducks).

To improve the clarity of the results section, a specific approach for presenting how respondents answered the different questions was adopted, using the following index:

- **n_{ans}** (A): number of A answers divided by the total number of informative answers (i.e., excluding missing values or "unknown")
- **n_{unk}**: number of uninformative answers (missing values and "unknown"), divided by the total number of answers received (i.e., including informative answers), missing values and unknown
- **n_{exp}**: total number of experts asked, meaning the ones for whom the question applied (whether they gave an informative answer or not)

Here is an example of this presentation with the following sentence. *"It appears that birds are most often caught* ***manually*** *whichever the species at stake* ***(**n_{ans}**=30/31 according to 15 experts***, ***n_{unk}=9/40 according to 5 experts***, ***n_{exp}=17)***" means that, out of the **total of 40 answers received from 17 experts** (concerning all species), **9 were "unknown" answers from 5 experts**, and among the 31 remaining answers (informative answers, i.e., not "unknown"), **30 answers from 15 experts suggested that birds are caught manually**. *Figure 1* offers a graphical representation of the distribution of the different kind of answers according to the number of respondents for this specific example.

Answers to non-applicable questions were never counted (e.g. a question specific to ducks if the expert had not reported culling ducks).

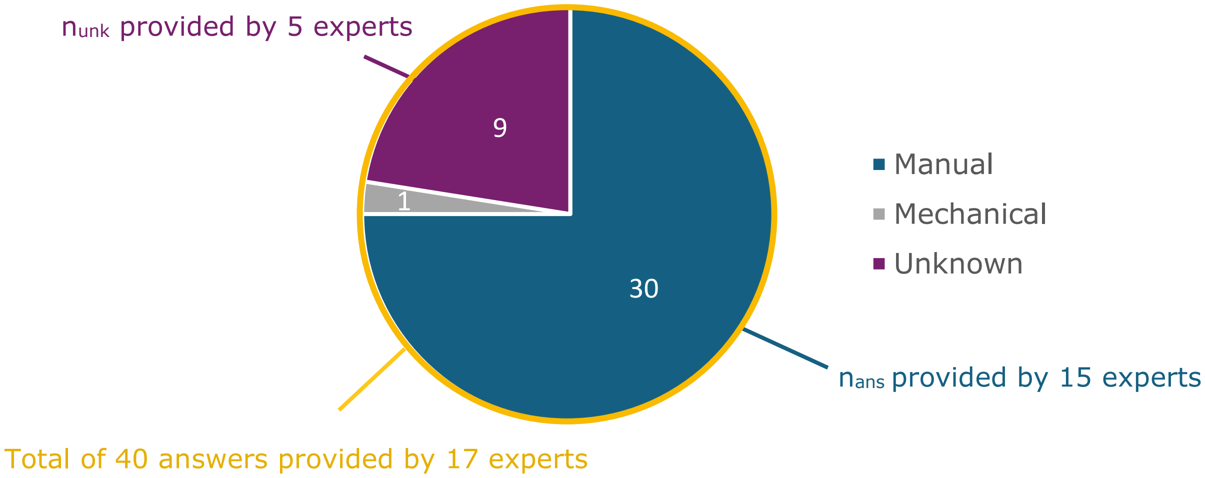


Figure 1: Graphical representation of the answer distribution according to the experts for the following sentence "It appears that birds are most often caught manually whichever the species at stake (n_{ans}=30/31 according to 15 experts, n_{unk}=9/40 according to 5 experts, n_{exp}=17)"

8 Results

Words of caution: Of note, the results presented and discussed here reflect the depopulation procedure used by the experts who answered the survey. Whether these results are representative of the procedure used nationwide or across the European Union is unknown.

Overview of the answers received

The number of answers received for each method-specific survey is detailed in *Table 2*.

Table 2: Overview of the number of MS who answered each method-specific survey together with the total number of answers received

Depopulation method	MS contacted (number)	MS answering (number)	Answers analysed
Containerized gassing	17	16	17
House gassing without foam	13	13[1]	11
Lethal injection	14	11	12
Cervical dislocation	15	10	11
Captive bolt	5	3	4
Blow to the head	3	1	1
Electrical waterbath	4	1	2
Head-to-Body electrical stunning	2	2	2
Head-only electrical Stunning	2	1	1
Decapitation	1	0	0

[1]A MS who initially did not report the use of house gassing as a depopulation method still answered the survey

Containerized gassing

Depopulation context

From the 17 MS who reported the use of Containerized Gassing (**CG**) since 2018, the depopulation experts from 16 MS answered the method-specific survey with a **total of 17 answers** (two experts from one MS replied). The **three most common reasons** given for using CG specifically are 1) the number of birds in the flock, 2) the availability of the equipment and 3) the species at stake.

Across the EU, CG is used in **various types of housing systems** (single-tier mentioned by 10 experts, multi-tier mentioned by 9 experts, and cages mentioned by 6 experts). Two experts specifically reported using CG when whole-house gassing could not be applied due to sealing issues of the building. CG is also used across the EU to depopulate **flocks of various sizes**, from flocks with less than 250 individuals up to flocks with more than 20 000 individuals.

CG has been used on **all the poultry species under study** (Gallus *gallus,* ducks and turkeys). Some MS appear to apply CG on birds regardless of the live weight (n_{ans}=9/15 according to 9 experts, n_{unk}=2/17 according to 2 experts, n_{exp}=17), while others only apply it on relatively light birds (maximum between 7 kg and 15 kg). Above 7 kg, one expert reported using CG with a conveyor (slide type) to help introducing the birds into the container by providing them support. One expert also reported that the weight threshold depends on the dimensions of the container and transport modules available, with no more precision.

Operating procedures

In the EU, three **types of containers** are used: small containers (e.g., bin types), large containers (i.e., those requiring transport by truck), and containerized gassing units (CGU: a gas-tight metal container fitted with a gas delivery system, that is gradually filled after the birds are introduced in the container in crates/modules). During the workshop, experts from various MS specified that their CGU's capacity depends on the size of the birds, and can either contain 10 or 50 tons of birds depending on their design. They also explained that the crates were of different sizes according to the bird species. Also, experts from two MS specified that they used gassing bags in lieu of "small containers".

A total of four **different gas mixtures** were reported being used (listed from the most to the least common used): CO_2 at high concentration[1] (**CO2-High**, reported by 15 experts), CO_2 associated with inert gases[2] (hereafter referred to as **CO2-Inert**, used by 4 experts), CO2 in two phases[3] (hereafter referred to as **CO2-Phase**, used by 3 experts), and carbon monoxide associated with other gases[4] (**CO,** used by 1 expert). Of note, during the workshop, the experts using CO2-Phases underlined the difficulty in fulfilling the requirements of the Annex 1 to the Council Regulation (EC) No 1099/2009 of 24 September 2009 on the protection of animals at the time of killing (hereafter referred to as Council Regulation 1099), since operators struggle to assess the state of consciousness of the birds inside the container between the two. CO2-High,

[1] Depopulation with CO2 in high concentrations refers to a gas mixture containing more than 40% CO2 (COUNCIL REGULATION (EC) No 1099/2009 of 24 September 2009 on the protection of animals at the time of killing, Annex I)

[2] Depopulation with CO_2 associated with inert gases refers to a gas mixture containing at most 40% CO2 associated with inert gases [either nitrogen or argon in practice] (COUNCIL REGULATION(EC) No 1099/2009 of 24 September 2009 on the protection of animals at the time of killing, Annex I)

[3] Depopulation with CO2 in two phases refers to a successive exposure to a gas mixture containing at most 40% CO2, followed, when animals have lost consciousness, by a higher concentration of CO2 (COUNCIL REGULATION(EC) No 1099/2009 of 24 September 2009 on the protection of animals at the time of killing, Annex I)

[4] Depopulation with carbon monoxide associated with other gases refers to a gas mixture containing at least 1% of CO associated with other toxic gasses (COUNCIL REGULATION(EC) No 1099/2009 of 24 September 2009 on the protection of animals at the time of killing, Annex I)

CO2-Inert and CO2-Phase are used to depopulate all poultry species under study, and gas mixture with CO has only been reported to be used on ducks (*Figure 2*). During the workshop, the MS using CO also mentioned the use of the gas mixture on other small birds such as pheasants, but only for small-sized flocks. Whether some gas mixtures are exclusively used in a certain type of container remains unknown. However, during the workshop, no experts mentioned using CO2-Inert (nor inert gases only) in large containers (e.g. skips) or small containers (e.g. gassing bags or small bins).

Regarding the **filling method** of the gas mixtures, containers usually appear to be pre-filled rather than gradually filled, whichever the type of container (n_{ans}=14/20 according to 9 experts, n_{unk}=1/21 according to 1 expert, n_{exp}=14). During the workshop, however, experts specified that CGU are never prefilled (i.e. always gradually filled), contrary to large containers.

Regarding the **target concentration aimed at**, most experts that reported using **CO2-High** mentioned values equal or superior to 70% CO2 for Gallus *gallus* (n_{ans}=8/13 according to 8 experts, n_{unk}=2/15 according to 2 experts, n_{exp}=15) and turkeys (n_{ans}=5/9 according to 5 experts, n_{unk}=2/11 according to 2 experts, n_{exp}=11). Experts using CO2-High on ducks equally reported targeted concentrations equal or superior to 70% CO2, and above 40% CO2 (n_{ans}=3/6 for both, according to 6 experts, n_{unk}=1/7 according to 1 expert, n_{exp}=7).

For **CO2-Inert**, the target concentration was reported to be either of 18% CO2 (n_{ans}=3/6 according to 1 expert, n_{unk}=2/8 according to 1 expert, n_{exp}=4), or less than 5% or 2% O2 (n_{ans}=3/6 according to 2 experts, n_{unk}=2/8 according to 1 expert, n_{exp}=4). During the workshop, experts from one MS specified that when using CO2-Inert in CGU, the gas mixture used is 82% Argon with 18% CO2, since this mixture is commonly available as welding gas. Another MS that had reported using N2 as an inert gas did not know the mixture composition.

All experts using **CO2-Phase** reported values inferior or equal to 40% CO2 for unconsciousness induction (1^{st} phase) whichever the species at stake (n_{ans}=9/9 according to 3 experts, n_{unk}=0/9, n_{exp}=3). The exact target concentration for death induction with CO2-Phase (2^{nd} phase) is not known (n_{ans}=6/9 according to 2 experts, n_{unk}=0/9, n_{exp}=3) but one expert reported a target concentration of 70% for all species under study. During the workshop, one expert explained his country's operating procedures when using CO2-Phase: the operators insert conscious birds within the large container which is filled with 25% CO2. They do so for each layer of birds as soon as the previous layer is deemed unconscious and until the container is full of unconscious (but supposedly alive) birds. Then, CO2 concentration is raised to 70% CO2 to kill all the birds. Another expert specified that in his MS, a national legislation prohibits the use of CO2-Phase in large containers, by enforcing a constant concentration of 80% CO2 within large containers. The expert that reported using CO specified that the target concentration was at least 1% CO.

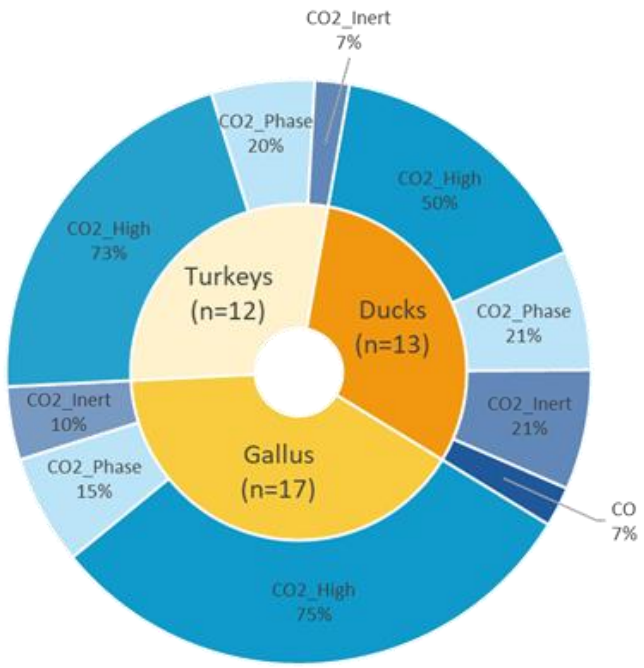


Figure 2: Gas mixtures used according to the depopulated species. CO2_High: CO_2 at high concentration, CO2_Phase: CO_2 in two phases, CO2_Inert: CO_2 associated with inert gases, CO: carbon monoxid associated with other gases

The question of the **time taken to reach the targeted concentration** once the birds were inserted into the containers was addressed during the workshop. Four experts reported reaching 40% CO2 within 2 minutes and 70% CO2 within 3 minutes when using CO2-Phase in CGU. When using CO2-Inert in CGU, four experts reported reaching less than 5% O2 within 5 min.

Once the target concentration is reached, the reported **durations for which it is maintained** in the container were very heterogenous. For CO2-High, the durations reported by 7 experts are shown in *Figure 3*. They were all inferior to 30 min after having reached the target concentration, with a mean of approximately 11 min (± 10 min) for Gallus *gallus,* 16 min (± 12 min) for ducks and 12 min (± 6 min) for turkeys. Of note, two experts reported times relative to events different from the moment when the target concentration was reached: one specified a duration "once [Gallus gallus and turkeys were] dead", and the other "15 min after the last bird [was] dropped". For CO2-Inert, one expert reported a duration of 15 min for Gallus *gallus* and ducks, and another reported a duration of "5 min after the birds stopped moving" – whichever the species at stake. During the workshop, four experts reported maintaining less than 5% O2 for 10 min when using CO2-Inert in containers. For CO2-Phase, one expert reported waiting 5 min after reaching both 40% CO2 and 70% CO2 – whichever the species at stake. Without giving a specific duration, another expert mentioned maintaining the target concentration of CO2-Phase until the death of the birds whichever the species at stake. For CO, the expert concerned reported maintaining the target concentration for 10 min.

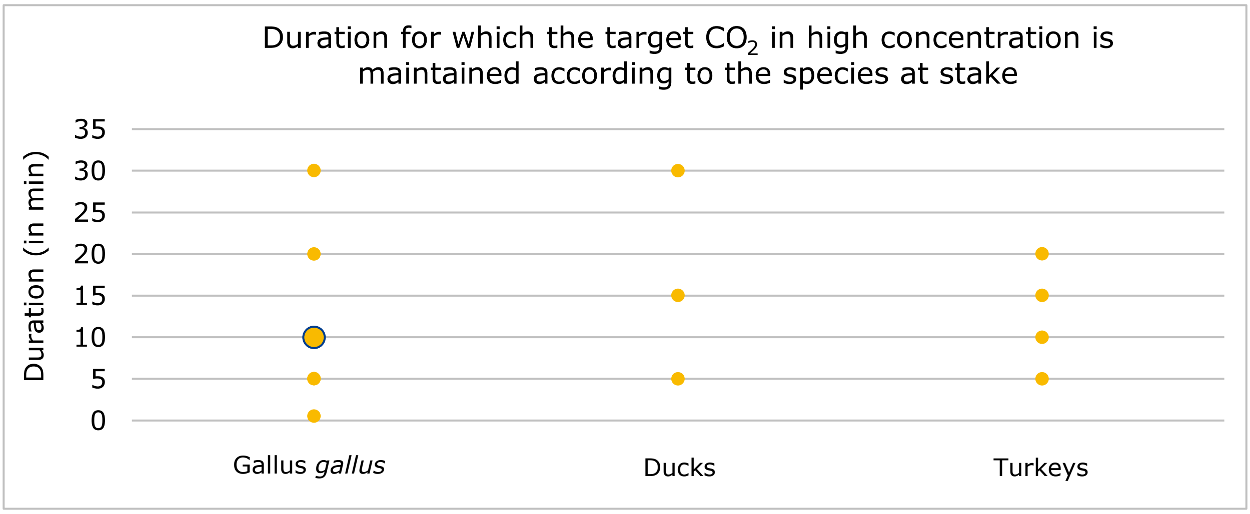


Figure 3: Reported durations (min) for which the CO_2 in high concentration is maintained, for Gallus gallus (left), ducks (center) and turkeys (right). Each dot corresponds to one answer, except for the bigger dot circled in blue for which two identical answers were obtained (10 min for Gallus gallus)

Welfare implications

The **main welfare consequences** associated with CG reported by the respondents were (from the most to the least often cited):

- The animal handling, mentioned by 6 experts
- The aversive reaction to high CO_2 concentration, mentioned by 3 experts
- Bird smothering, mentioned by 3 experts
- The difficulty to reach the targeted concentrations, mentioned by 2 experts

Other welfare aspects that were mentioned by single experts: the difficulty to assess death, the cold stress induced to birds and the dropping of the birds within the containers.

Welfare implications associated with the preparation of the killing

Food and water are seldom removed before the start of CG (food: n_{ans}=5/17 according to 5 experts, n_{unk}=0/17, n_{exp}=17 and water: n_{ans}=4/17 according to 4 experts, n_{unk}=0/17, n_{exp}=17). When water is removed, feed is also systematically removed; from 12 h to immediately before the start of the depopulation.

Most answers suggest that the **injection points are positioned in a way to prevent the birds from freezing**, whichever the gas mixtures used (n_{ans}=12/13 according to 9 experts and n_{unk}=10/23 according to 8 experts, n_{exp}=17).

Welfare implications associated with animal handling

Regarding the **means of catching** the birds, it appears that birds are most often caught manually whichever the species at stake (n_{ans}=30/31 according to 15 experts, n_{unk}=9/40 according to 5 experts, n_{exp}=17). When the operators catch the birds manually, they don't hold more than two turkeys or ducks per hand, but sometimes do so for Gallus *gallus*. For more details regarding the number of birds held by hand by one operator according to the depopulated species, see *Figure 4*.

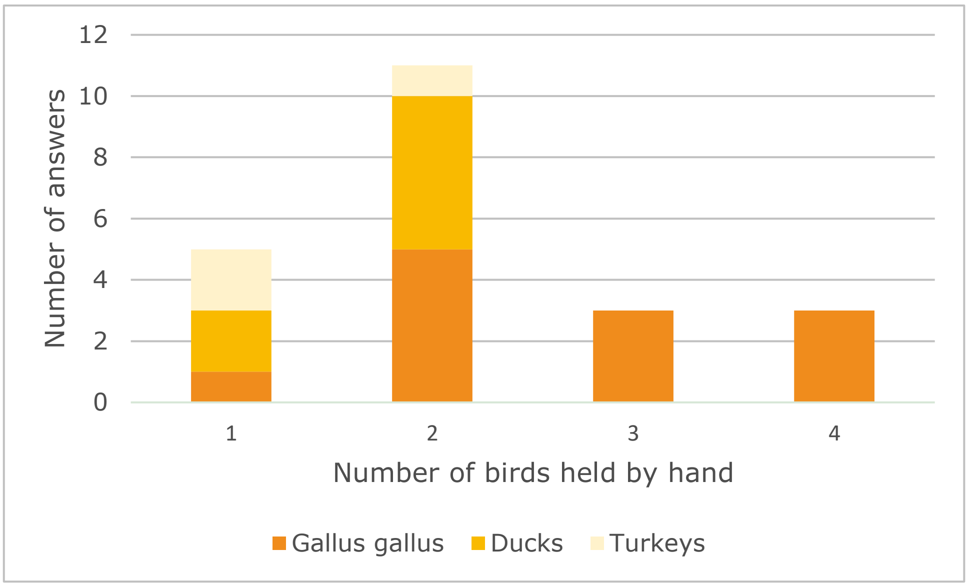


Figure 4: Number of birds held in one operator's hand according to the species at stake

Birds are mostly **transported** manually (n_{ans}=12/19 according to 12 experts, n_{unk}=3/22 according to 3 experts, n_{exp}=16) and less frequently via crates (n_{ans}=4/19 according to 4 experts n_{unk}=3/22 according to 3 experts, n_{exp}=16) or using mechanical devices (n_{ans}=3/19 according to 3 experts n_{unk}=3/22 according to 3 experts, n_{exp}=16). One expert reported that the killing of the birds occurs directly "on the spot". Whether some means of transport are used exclusively for certain species remain unknown. During the workshop, one MS specified that when culling turkeys using CGU, the operators would unpile the crates in the barn and let the turkeys walk in, before forklifting each layer of crates to the container. Also, experts were asked if they thought that it was possible to walk ducks and laying hens to large containers. Three experts answered "yes", four "no" and seven did not know.

Birds transported manually are equally reported to be **held in an inverted position and in an upward position** (n_{ans}=6/12 for both, according to 12 experts, n_{unk}=0/12, n_{exp}=12).

When transported in crates, Gallus *gallus* are grouped in **sets of 6 to 10 individuals** (n_{ans}=2/2 according to 2 experts, n_{unk}=2/4 according to 2 experts, n_{exp}=4). No data relative to the size of the groups within the crates for ducks or turkeys was obtained. Birds remain inside their crates during the depopulation procedure when CGU are used (n_{Ans}=2/2 according to 2 experts, n_{unk}=0/2 n_{exp}=2) but not when other types of containers are used (n_{Ans}=3/3 according to 2 experts, n_{unk}=0/3 n_{exp}=2).

The **transport duration** between the barn and the container is always made the shortest possible (n_{ans}=15/15 according to 15 experts, n_{unk}=2 according to 2 experts, n_{exp}=17).

Specific equipment is rarely used **to drop birds inside the container** (n_{ans}=4/15 according to 4 experts, n_{unk}=2/17, n_{exp}=17), but conveyor belts (slide types) are sometimes installed (n_{ans}=2/2 according to 2 experts, n_{unk}=2/4 according to 2 experts, n_{exp}=4).

Welfare issues occurring during the depopulation procedure

All experts with an informative answer reported that the **birds are evenly distributed inside the containers** regardless of the type of container used (n_{ans}=17/17 according to 10 experts, n_{unk}=12/39 according to 6 experts, n_{exp}=16). The experts also reported that the **death of all birds introduced in the container is usually ensured before another layer of birds is added** (n_{ans}=6/8 according to 6 experts, n_{unk}=4/12 according to 4 experts, n_{exp}=12). Most experts reported that **birds are observed during the procedure** when small containers and CGU are used (n_{ans}=10/15 according to 9 experts, n_{unk}=1/16, n_{exp}=16). When big containers are used, only half of the experts who gave an informative answer reported that the birds are observed during the procedure (n_{ans}=3/6 according to 3 experts, n_{unk}=1/7, n_{exp}=7). During the workshop, experts from the two MS that specified using gassing bags (comprised in the "small containers" category), explained that, in their experience, it was not possible to avoid birds smothering in these bags. One MS shared its negative experience with gassing bags by explaining that it was difficult to manage the birds flow from the barn to the gassing bags.

Regarding the **percentage of hyperventilating and deep-breathing birds** observed within the container during the gassing procedure, answers were only obtained for CO2-High used on Gallus *gallus*, ducks and turkeys (reported by 5 experts) and CO2-Inert used on Gallus *gallus* and ducks (reported by 1 expert). Overall, results were very heterogenous and ranged from "less than 0.1%" to "100%" for ducks, and from 0.1% to 100% for Gallus gallus and turkeys when using CO2-High. The only answer obtained for Gallus *gallus* and ducks subjected to CO2-Inert indicate that 100% of the birds hyperventilate. The percentage of hyperventilating birds according to the species at stake and the type of gas mixtures is detailed in *Figure 5A*. During the workshop, experts from the MS that had reported observing 100% birds (whichever the species) hyperventilating when CO2-High and CO2-Inert are used specified that they did not agree with the answers filled in the survey, since it is impossible in practice to assess the proportion of hyperventilating birds because of the fog. Experts from the MS that had reported 0.1-1% of Gallus *gallus* and less than 0.1% of ducks hyperventilating with CO2-High rectified these percentages to 10%, without giving further information relative to how hyperventilating is assessed. The corrected percentage of hyperventilating birds according to the species at stake and the type of gas mixtures is detailed in *Figure 5B*.

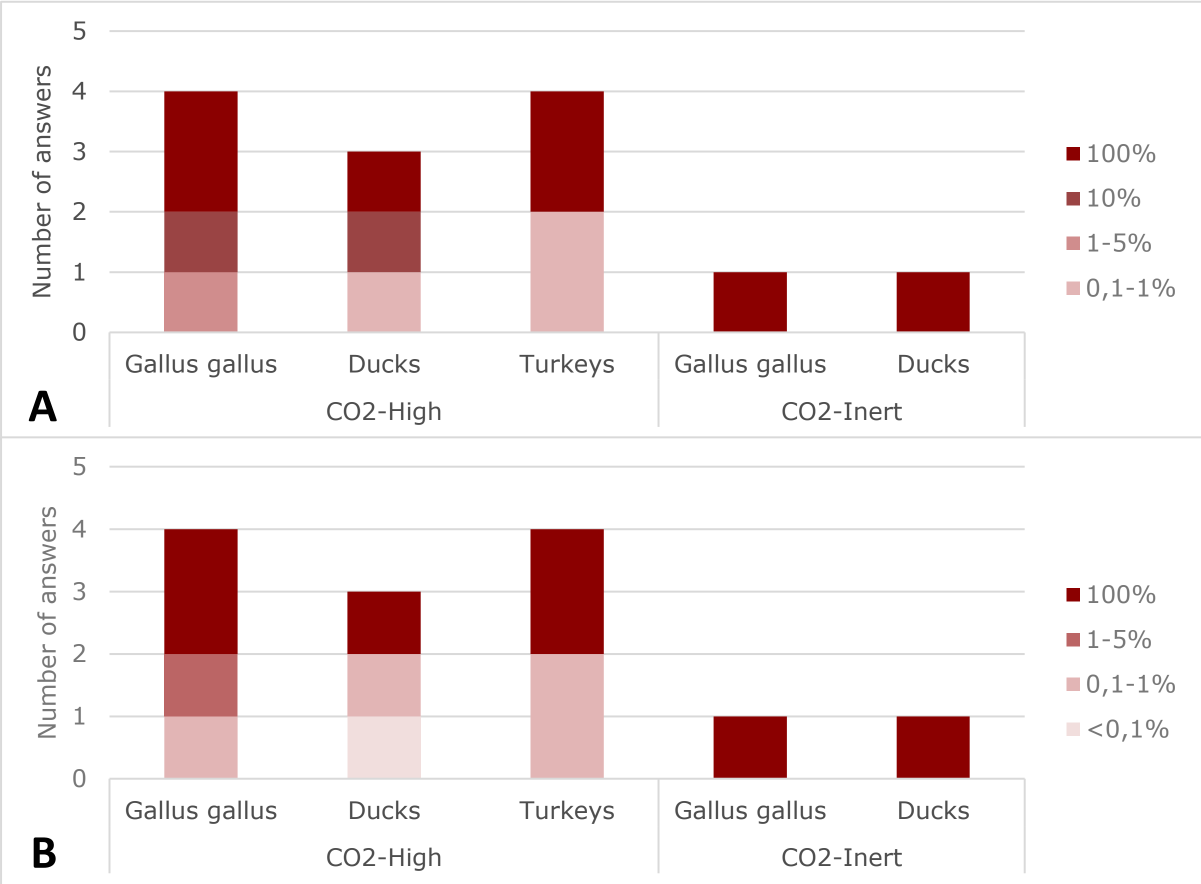


Figure 5: Number of answers (A) and of corrected answers after the workshop (B) obtained for different classes of percentage of hyperventilating and deep-breathing birds according to the species at stake and type of gas used. CO2-High: CO_2 in high concentrations, CO2-Inert: CO_2 associated with inert gases

Regarding the **percentage of birds with frostburns**, answers suggest that less than 0.1% of the birds are affected regardless of the species at stake and the type of gas mixture used (n_{ans}=16/18 according to 7 experts, n_{unk}=29/47 according to 9 experts, n_{exp}=17). Contrary to others though, one expert indicated that between 1% and 5% of Gallus *gallus* and turkeys have frostburns when gassed with CO2-High. No answer was obtained for CO2-Phase nor CO.

Efficiency

Regarding the **percentage of birds remaining conscious or regaining consciousness**, it appears that, overall, less than 0.1% of the birds remain conscious (n_{ans}=20/22 according to 8 experts and n_{unk}=10/32 according to 6 experts, n_{exp}=15), or regain consciousness (n_{ans}=19/19 according to 8 experts and n_{unk}=13/32 according to 8 experts, n_{exp}=15) at the end of CG when CO2-High or CO are used – whichever the species at stake. One expert, though, indicated that between 1% and 5% of Gallus *gallus* and turkeys remain conscious when gassed with CO2-High. No answer was obtained regarding the percentage of birds remaining conscious or regaining consciousness when CO2-Inert or CO2-Phase are used.

Regarding the **percentage of birds being unconscious but still alive** at the end of CG, answers suggest that less than 0.1% of birds are unconscious but alive at the end of the procedure (n_{ans}=25/25 according to 10 experts and n_{unk}=22/47 according to 8 experts, n_{exp}=17), whichever the species at stake and the gassing mixture used.

Regarding the **latency for unconsciousness induction of the first bird** once the targeted concentration has been reached within the container, results obtained for CO2-High are heterogeneous: durations for unconsciousness induction range from immediate (n_{ans}=2/13 according to 1 expert, n_{unk}=11/24 according to 8 experts, n_{exp}=15) to 1-5 minutes (n_{ans}=4/13 according to 2 expert, n_{unk}=11/24 according to 8 experts, n_{exp}=15) for Gallus *gallus* and turkeys, and from less than 1 minute (n_{ans}=3/5 according to 3 expert, n_{unk}=2/7 according to 2 experts, n_{exp}=7) to 1-5 minutes (n_{ans}=2/5 according to 2 experts, n_{unk}=2/7 according to 2 experts, n_{exp}=7) for ducks. When CO2-Inert is used, durations range from less than 1 minute ((n_{ans}=1/2 according to 1 expert, n_{unk}=0/1, n_{exp}=2) to 1-5 minutes (n_{ans}=1/2 according to 1 expert, n_{unk}=0/1, n_{exp}=2) for Gallus *gallus*, and only one duration of 1-5 minutes was reported for ducks (n_{ans}=2/2 according to 2 experts, n_{unk}=1/3 according to 1 expert, n_{exp}=3). No duration was indicated for turkeys. When CO2-Phase is used, only "immediate" durations have been reported whichever the species at stake (n_{ans}=3/3 according to 1 expert, n_{unk}=6/9 according to 2 experts, n_{exp}=3). When CO is used on ducks, the only expert concerned reported a duration inferior to 1 min. *Figure 6A* details the time for unconsciousness induction for the first bird according to the species at stake and the type of gas mixture used. Of note, during the workshop, experts emphasized that assessing unconsciousness was very complicated in all types of containers because of the fog created by gas vaporization.

Regarding the **latency for unconsciousness induction of the last bird** once the targeted concentration has been reached within the container, results obtained for CO2-High are heterogeneous: durations for unconsciousness induction range from less than 1 minute (n_{ans}=5/11 according to 3 experts, n_{unk}=9/20 according to 8 experts, n_{exp}=15) to 5-15 minutes (n_{ans}=2/11 according to 1 expert, n_{unk}=9/20 according to 8 experts, n_{exp}=15) for Gallus *gallus* and ducks, and from immediate (n_{ans}=1/5 according to 1 expert, n_{unk}=6/11 according to 6 experts, n_{exp}=11) to 5-15 minutes for turkeys (n_{ans}=1/5 according to 1 expert, n_{unk}=6/11 according to 6 experts, n_{exp}=11). When CO2-Inert is used, durations range from 1-5 minutes (n_{ans}=2/4 according to 1 expert, n_{unk}=1/5 according to 1 expert, n_{exp}=3) to 5-15 minutes (n_{ans}=2/4 according to 1 expert, n_{unk}=1/5 according to 1 expert, n_{exp}=3) for Gallus *gallus* and ducks. No duration was indicated for turkeys. When CO2-Phase is used, the only expert who gave an informative answer reported "immediate" durations whichever the species at stake (n_{ans}=3/3 according to 1 expert, n_{unk}=6/9 according to 2 experts, n_{exp}=3). When CO is used on ducks, the only expert concerned reported a duration between 1-5 minutes. *Figure 6B* details the time for unconsciousness induction for the last bird according to the species at stake and the type of gas mixture used.

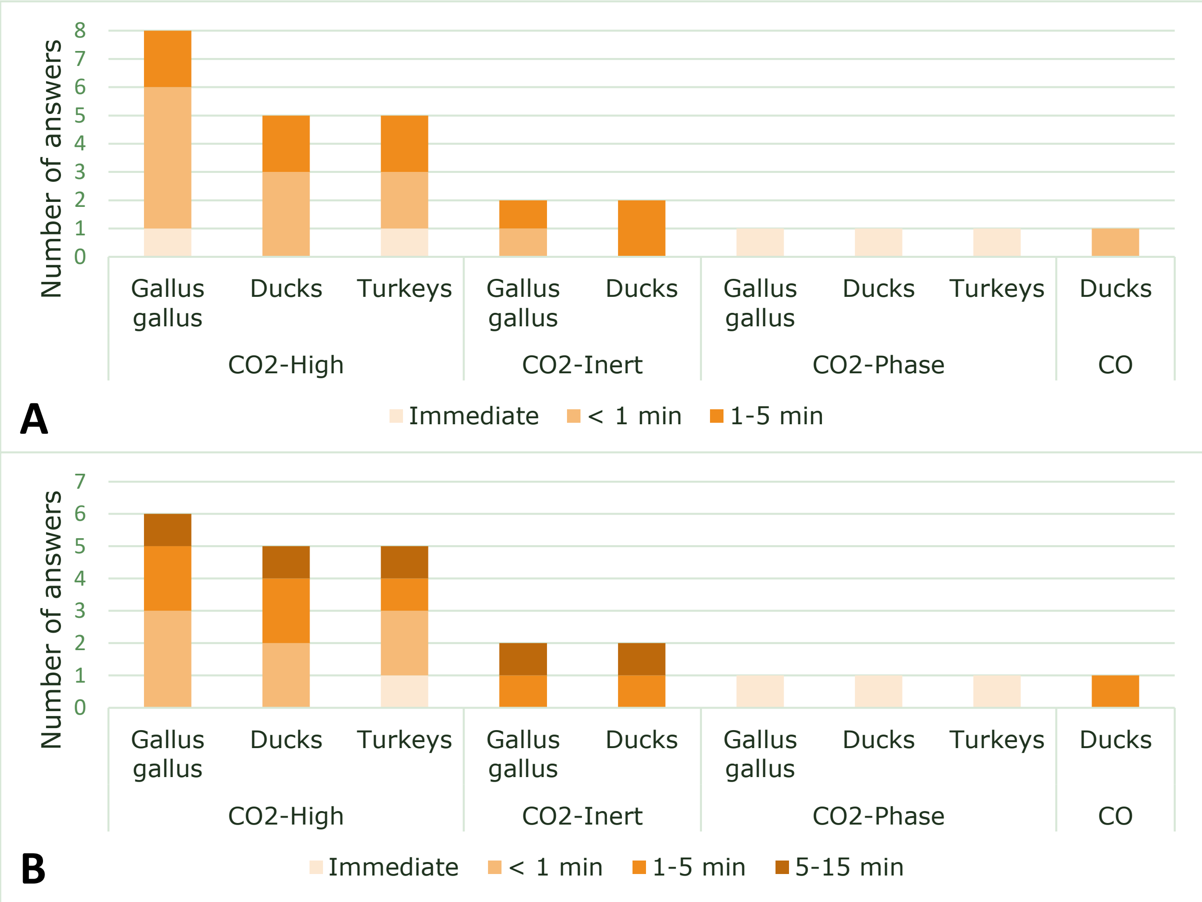


Figure 6: Number of answers obtained for different classes of latency for unconsciousness induction of the first (A) and last bird (B) of the same batch according to the species at stake and type of gas used. CO2-High: CO_2 in high concentrations, CO2-Inert: CO_2 associated with inert gases, CO2-Phase: CO_2 in two phases, CO: carbon monoxide associated with other gases

Regarding the **time of death induction of the first bird** once the targeted concentration has been reached within the container, results obtained for CO2-High are heterogeneous: durations for death induction range from less than 1 minute (n_{ans}=4/12 according to 3 experts and n_{unk}=12/24 according to 8 experts, n_{exp}=15) to 5-15 minutes (n_{ans}=2/12 according to 1 expert and n_{unk}=12/24 according to 8 experts, n_{exp}=15) for Gallus *gallus* and turkeys, and from less than 1 minute (n_{ans}=1/5 according to 1 expert and n_{unk}=2/7 according to 2 experts, n_{exp}=7) to 1-5 minutes for ducks (n_{ans}=4/5 according to 4 expert and n_{unk}=2/7 according to 2 experts, n_{exp}=7). When CO2-Inert is used, the only expert who gave an informative answer reported a duration of 1-5 minutes for Gallus *gallus* and ducks (n_{ans}=2/2 according to 1 expert and n_{unk}=3/5 according to 2 experts, n_{exp}=3). No duration was indicated for turkeys. No duration was indicated when CO2-Phase is used. When CO is used on ducks, the only expert concerned reported a duration inferior to 1 minute. *Figure 7A* details the time for death induction for the first bird according to the species at stake and the type of gas mixture used.

Regarding the **time of death induction of the last bird** once the targeted concentration has been reached within the container, results obtained for CO2-High are heterogeneous: durations for death induction range from 1-5 minutes (n_{ans}=11/16 according to 4 experts and n_{unk}=15/31 according to 9 experts, n_{exp}=15) to 5-15 minutes (n_{ans}=5/16 according to 2 experts and n_{unk}=15/31 according to 9 experts, n_{exp}=15) for all species under study. When CO2-Inert is used, the only expert who gave an informative answer reported a duration of 5-15 minutes for Gallus *gallus* and ducks (n_{ans}=2/2 according to 1 expert and n_{unk}=3/5 according to 2 experts, n_{exp}=3). No duration was indicated for turkeys. No duration was indicated when CO2-Phase is used. When CO is used on ducks, the only expert concerned reported a duration between 1-5 minutes. *Figure 7B* details the time for death induction for the last bird according to the species at stake and the type of gas mixture used.

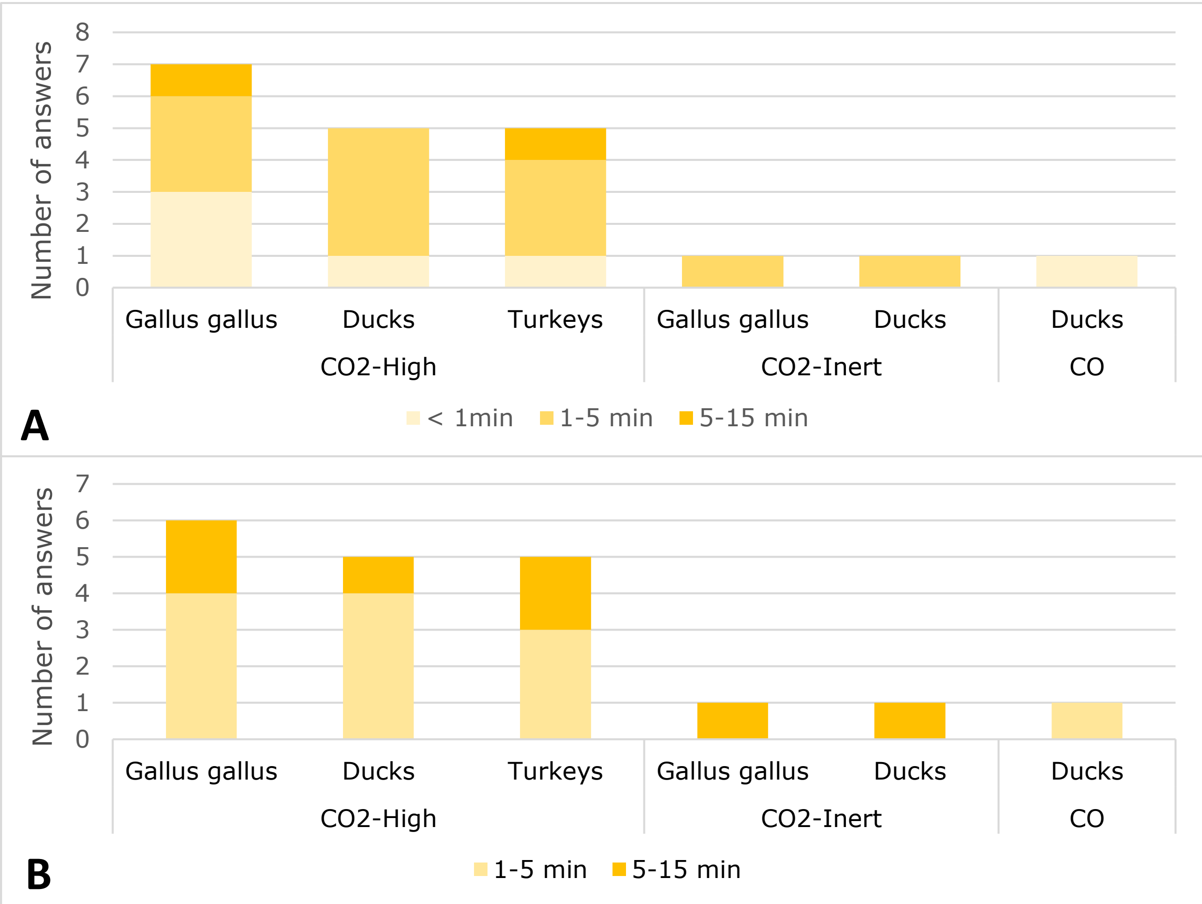


Figure 7: Number of answers obtained for different classes of latency for death induction of the first (A) and last bird (B) of the same batch according to the species at stake and the type of gas used. CO2-High: CO2 in high concentrations, CO2-Inert: CO2 associated with inert gases, CO: carbon monoxide associated with other gases

When birds are still alive at the end of CG, different **back-up methods** are used to ensure their death. These methods include (from the most reported to the least reported): cervical dislocation (reported by 11 experts), lethal injection (reported by 7 experts), and additional gassing (by increasing the concentration or starting over the procedure, reported by 5 experts). Decapitation (reported by 2 experts), gunshot (reported by 2 experts), and electrical waterbath (reported by 1 expert) were also mentioned.

Discussion and conclusion

Various operating procedures are used to perform CG. This diversity partially owes to the fact that:

- Various resources (type of container, gas mixture) may be used to conduct CG
- Distinct filling techniques exist
- CG can be, and *is*, used on farms with different kinds of housing systems and flocks of all sizes

In practice, **three types of containers** are used on depopulation sites: CGU, large containers and small containers. All types of containers were almost equally reported to be used in the survey, but whether countries use one type more than the other is not known. The use of each type of container has its pros and cons. CGU, which are designed specifically for depopulation purpose, are completely hermetic and thus allows for a more controlled environment compared with other types of containers. Building such containers nonetheless requires economic investment, which may hinder their acquisition by some MS. Large containers constitute a cheaper and more practical alternative to CGUs, as common structure like farm dumpsters can easily be found and used for depopulation purposes. However, the use of large containers poses a threat to animal welfare, since birds are often dropped from high heights in the container and may die from smothering. Also, owing to the design of the large containers (skips, frequently opened from the top to add birds), gas concentration can be very fluctuant and difficult to maintain at the proper target concentration. Scarce information was obtained concerning small containers (bags and bins). Overall, it still appears that the main advantage of small containers are their availability and practicality, provided a sufficient number of operators is present on site to transport the animals from the barn to the container. In practice, however, gassing bags often lead to bird smothering.

In practice, **various types of gas mixtures** (CO2-High, CO2-Inert, CO2-Phase, CO) appear to be used throughout the EU. CO2 is predominantly used, probably in all types of containers. The use of CO2-Inert and CO appears to be restricted to depopulation sites where CGU are available. CO is never used in large containers, and its use appears furthermore limited to small flocks. The reluctance to use CO could be explained by its dangerous properties: the storage of CO is associated with a risk of explosion and flammability. Additionally, CO is highly toxic for humans as it binds to hemoglobin with a much higher affinity than oxygen (New Jersey Department of Health, 2016). Further research could be led on gassing mixtures containing argon (eventually associated to CO2) in large containers.

The choice of the **best gassing mixture** to use in relation to their animal welfare repercussions **remains undetermined**. The literature leaves no doubt about the fact that all gassing mixtures reported here are at least mildly aversive (Sandilands, Raj, Baker, & Sparks, 2011; Rucinque et al., 2024); especially CO2 at high concentration, the most used gassing mixture, which has been proved to be aversive to poultry (EFSA 2005). Papers have highlighted that the addition of inert gases to CO2 shortens the duration to lose consciousness and to die (and with less variability) (Rucinque et al., 2024). Of note, as underlined by the EURCAW-Poultry-SFA (2022), death by anoxia that occurs when inert gas is added in the gas mixture leads to more intense convulsions, which may cause self-inflicted injuries (wing fractures) or injuries and pain to the other birds that have not yet lost consciousness (McKeagan et al., 2007, Berg & Raj, 2015). It is not entirely clear if 1) the convulsions are a reflexive reaction occurring after the bird loses consciousness or 2) if the birds are still conscious and trying to escape from this modified atmosphere when they convulse (McKeagan et al., 2007, Coenen, Lankhaar, Lowe, & McKeegan, 2009). Therefore, it is difficult to determine which gassing mixture should be used when gassing in containers. The choice of gassing mixture also depends on its availability at local and national level, which may vary - especially during outbreaks. For this reason, it is important that MS assess and plan for the sufficient quantities of gas beforehand (EURCAW-Poultry-SFA, 2024). The welfare consequences of the gas mixture used also depend on the filling technique adopted.

Various filling techniques with yet-to-be determined welfare impacts on birds are used on the field, depending on the container used. Containers may be gradually filled, if the mixture is progressively administered in the container while the birds are present; or prefilled, if the birds are placed in the container once a certain concentration is achieved. CGU are gradually filled and never prefilled. When reviewing existing literature, a lack of knowledge on the optimal filling kinetic (with regard to animal welfare) to adopt in a CGU has been brought to light, as experimental studies use prefilled pits (Rucinque et al., 2024) or unrepresentative small-size version of CGUs. Large containers are often prefilled to a certain concentration, and then gradually filled to a higher concentration once the birds are inserted. It is unsure how small containers (gassing bags, small bins) are filled in practice. It is uncertain which type of filling (gradual filling, prefilled containers, or combination of both) is best in regard to animal welfare in large and small containers, but the decision has to be made with regards to the type of gas mixture used. For example, prefilling with CO2 at very high concentrations (e.g. 80% CO2) implies exposing conscious birds to highly aversive gas concentrations. When using CO2 in large containers, to avoid inserting birds at highly aversive concentrations, other MS use a ‘two phase’ procedure, in which birds are added continuously within the large container filled with 25% CO2 until it is full, and then concentration is raised to lethal levels. Description of this protocol brought to light a potential risk of birds smothering before losing consciousness. Moreover, this protocol does not fulfil as Council Regulation 1099 requirements as there is no possibility of visually assessing if birds have lost consciousness before raising CO2 concentration to high levels (i.e. higher than 40% CO2).

A welfare issue of CG is the **necessity of animal handling** when birds are transported to the container. Birds are reported to be transported mostly manually to the containers, which can affect their welfare depending on the procedure used. When transported manually, experts reported that operators transported them equally in an inverted and upright position. Transporting birds in an inverted position causes stress, fear and wing flapping behavior (EFSA, 2022). This may be avoided by crating the birds or transporting them in an upright position. However, crating and uncrating the birds can cause damage (e.g. broken wings, injuries to the back and thigh, bruises, etc.) (EFSA, 2022). If birds are to be gassed uncrated in containers, some scientists hence recommend to transport them without crates (i.e. manually or mechanically with conveyor belts for e.g.) from the barn to the container, over crating them in the barn and then uncrating them at the container (as two experts reported doing) in order to limit suffering and stress.

Another major welfare issue is the **inability for operators to observe the birds**, and consequently to observe their reaction to the gassing mixtures and assess their welfare during the procedure. The welfare assessment is hampered by the fog created by gas vaporization. As a result, onset of unconsciousness, hyperventilation and deep-breathing cannot be reliably assessed.

Despite its potential welfare concerns, **containerized gassing remains an effective method** to kill birds for all sizes of flocks, with less than 0.1% of failure (birds regaining consciousness, being unconscious but alive or remaining conscious at the end of the procedure). It is a complex task to determine a best practice as there are many possible arrangements (type of gas, of container, of filling technique, …). However, it seems that overall, from a welfare point of view and concerning the type of container to use, CGU seems to be the best option for containerized gassing. It avoids excessive handling and smothering by using crates, avoids exposing conscious birds to highly aversive concentrations of gas by gradually (albeit quickly, within minutes) filling the container, allows monitoring gas concentration continuously, and allows inert gases to be used.

House gassing without foam

Depopulation context

A total of at least 13 MSs reported to have used House Gassing without foam (**HG**) since 2018 in the EU. In total, answers from 10 MS were obtained (three experts from one MS, and two experts from another MS replied), and two answers had to be discarded. Only **11 answers** were thus analysed. The **three most common reasons** given for using HG specifically are 1) the animal welfare consequences, 2) the number of birds in the flock, and 3) the species at stake.

Across the EU, HG is used in **various types of housing systems** (single-tier mentioned by all 11 experts, multi-tier mentioned by 10 experts, and cages mentioned by 7 experts). Most experts reported that HG can be applied to housing with outdoor access (n_{ans}=9/11 according to 9 experts, n_{unk}=0/11, n_{exp}=11). During the workshop, four experts mentioned the procedure can also be used for barns with slatted floors as well. Of note, one expert specified that house-gassing was difficult to use in barns where the floor had a strong slope.

Whichever gas is used, HG is mostly used **exclusively on flocks of more than 500 birds** (n_{ans}=10/11 according to 9 experts, n_{unk}=1/12, n_{exp}=11), although one expert also reported using HG for smaller flocks.

HG appears to be used to depopulate **all poultry species under study** (from most to least often cited): turkeys, Gallus *gallus*, and ducks (reported by 11, 10 and 4 experts, respectively). The method is applied regardless of the bird weight (n_{ans}=10/11 according to 10 experts, n_{unk}=0/11, n_{exp}=11), although one expert reported using it "especially [with] heavy turkeys".

Operating procedures

Two types of procedures were (unexpectedly) reported to be used when performing HG: whole-house gassing and, less frequently, partial house gassing (at least 2 MS). Partial house gassing differs from whole house gassing in that only a subpart of the house is gassed. This subpart is typically delineated using a tarp. In the survey, questions on the type of HG used were not asked. As such, procedures, welfare consequences and efficiency specific to each procedure could not be determined.

Two types of gas mixtures were reported being used across the EU. All experts reported using CO_2 at high concentration[5] (hereafter referred to as **CO2-High**), and one of them also reported using nitrogen alone[6] (hereafter referred to as **N2**). CO2-High has been reported to be used on all species under study, contrary to N2 for which the expert only mentioned using it on Gallus *gallus* and turkeys. Among the two countries that specified carrying out partial house gassing, one specified scattering CO_2 pellets on the floor while birds were restrained within their transport crates, whereas the other country mentioned scattering the pellets on the ground while birds were moving freely in a subpart of the barn.

Before the start of the depopulation procedure, most experts reported **shutting down the ventilation immediately** before injecting the gas within the house (n_{ans}=10/11 according to 10 experts, n_{unk}=0/11, n_{exp}=11). The gas mixture appears to be rarely pre-heated before being injected (n_{ans}=3/10 according to 3 experts, n_{unk}=2/12 according to 2 experts and n_{exp}=11). During the workshop, experts explained that the main reason for which they do not preheat the gas before injection is that they do not have (or are not aware of) specifically designed equipment available for this purpose. Experts also mentioned that preheating can slow down the depopulation procedure and one expert believed that pre-heating was "unnecessary". The approximate cost of preheating the gas in an average barn was unknown by the experts present the day of the workshop.

When injected, **the gas is delivered through one or more injection points**, depending on the house size. These injection points are generally positioned or designed in a way to ensure CO2-High is not directly orientated toward the birds (n_{ans}=7/10 according to 7 experts, n_{unk}=1/11 according to 1 expert and n_{exp}=11). For N2, the expert specified that the injection points were not

[5] i.e., a gas mixture containing more than 40% CO2
[6] i.e., a gas mixture containing nitrogen leading to anoxia

positioned or designed in a way to ensure that the gassing mixture is directly oriented toward the birds (n_{ans}=1/1 according to 1 expert, n_{unk}=0/1, n_{exp}=1).

The **target concentrations** of CO_2 reported by experts when using CO2-High were heterogenous, varying between 40% and 90% of CO_2. That being said, half of the target concentrations were reported to be equal or above 70% CO_2 (n_{ans}=4/8 according to 4 experts, n_{unk}=3/11 according to 3 experts, n_{exp}=11). During the workshop, most experts reported that for whole house gassing (as opposed to partial house gassing) with CO2-High, the CO_2 concentration achieved and maintained in the building was around 40-50%. In the survey, experts reported that CO2-High target concentrations are reached within an hour (n_{ans}=7/8 according to 7 experts, n_{unk}=3/11 according to 3 experts, n_{exp}=11), except for one expert who mentioned a three-hour filling duration. During the workshop, experts were asked about the shortest and longest durations to reach the target concentration that they had experienced in a barn of intermediate size (2 000 - 10 000 birds). For the shortest duration, three experts indicated durations inferior to 6 minutes. For the longest duration, three experts indicated durations between 15 and 20 minutes, and another expert indicated a duration of 30-40 minutes due to improper sealing.
The target concentration reported for N2 was of 97-98%, and said to be reached in 60 min.

The **volume of gas** required to perform whole house gassing is calculated based on different equations across the EU, which are dependent on the size of the barn. The equations used with CO2-High are synthesized in *Table 3*. For whole house gassing with N2, the expert only mentioned the calculation depended on the barn volume.

Table 2. Equations used to calculate the volume of CO_2 required when performing house gassing, together with the number of experts reporting using said equation

Experts (nb)	Equation
2	$V_{gas} = V_{barn} * 1.8 * 0.8$
1	$V_{gas} = L_{barn} * W_{barn} * (H_{barn}+1) * 1.75$
1	$V_{gas} = V_{barn} * 2.5$
1	$V_{gas} = V_{barn} * 2$

Where, V_{gas} = Volume of gas (m^3), V_{barn} = Volume of the barn (m^3), L_{barn} = Length of the barn (m), W_{barn} = Width of the barn (m), H_{barn} = Height of the barn (m)

Welfare implications

The main welfare issues associated with HG reported by the respondents were (from the most to the least often cited):

- The temperature drop and the cold temperature in the barn, due to gas expansion (mentioned by 3 experts)
- The risks of overheating and accumulating noxious gases before the procedure, because of ventilation shutdown (mentioned by two experts)
- The difficulty to control and maintain the targeted gas concentration in the barn or under the plastic sheets in case of partial house gassing (mentioned by two experts)
- The excessive time to induce unconsciousness (mentioned by two experts)
- The difficulty to visually check unconsciousness and death induction (mentioned by two experts)
- The aversive reaction to high CO_2 concentrations (mentioned by two experts)

Other welfare issues were mentioned once: the risk of birds getting frostburns from contact with the gas stream, and the necessity for animal handling with partial house gassing – which can trigger aggressive behaviors especially in turkeys.

Welfare implications associated with the preparation of the killing

Food is seldom removed before the start of the procedure (n_{ans}=5/11 according to 1 expert, n_{unk}=0/11, n_{exp}=11), contrary to **water which is often removed** (n_{ans}=9/11 according to 9 experts, n_{unk}=0/11, n_{exp}=11) to avoid freezing the pipes and to facilitate movement in the barn. Both resources appear to be generally removed within 2 hours before the start of the procedure (n_{ans}=8/9 according to 6 experts, n_{unk}=5/14 according to 3 experts).

Welfare issues occurring during the depopulation procedure

Regarding the **percentage of hyperventilating and deep-breathing birds** observed within the barn during the gassing procedure, answers were only obtained for CO2-High. Overall, results were very heterogeneous (ranging from less than 0.1% to 100%) as each expert gave a different answer for all species at stake. The percentage of hyperventilating and deep-breathing birds according to the species at stake is detailed in *Figure 8*. During the workshop, experts from the MS who had reported extreme values (either <0.1% or 100%) were asked to clarify the reasons for which they observed these proportions of birds. The answer was that it was not possible to know the percentage of birds hyperventilating during the procedure as the fog makes visual inspection hardly feasible.

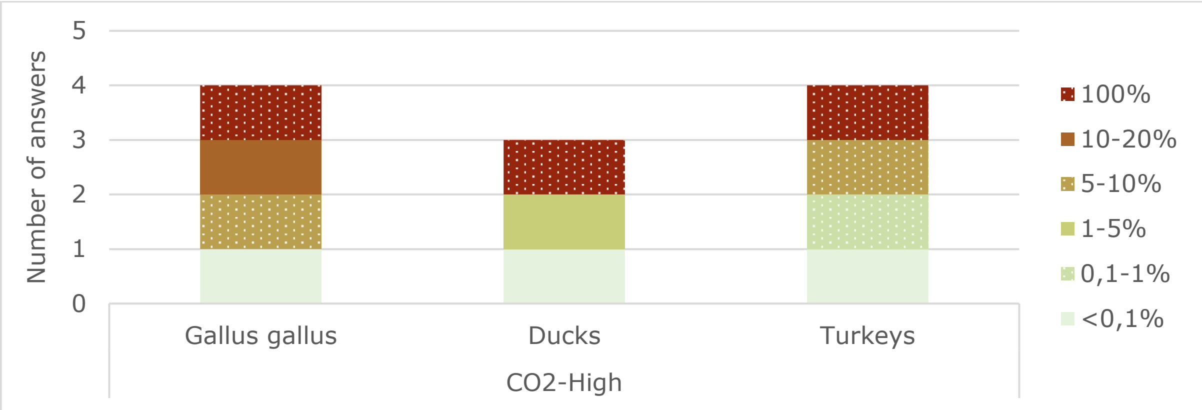


Figure 8: Number of answers obtained for different classes of percentage of hyperventilating and deep-breathing birds according to the species at stake, when CO2 in high concentration is used.

Regarding the **percentage of birds with frostburns**, most answers indicate that less than 0.1% of birds are affected when CO2-High is used, whichever the species at stake (n_{ans}=17/22 according to 7 experts, n_{unk}=3/25 according to 2 experts, n_{exp}=11). The highest percentage intervals of birds with frostburns reported for Gallus *gallus* and turkeys were comprised between 1 and 5%, and between 0.1 and 1% for ducks. Those intervals were cited by one expert only. During the workshop, an expert from one of the MS using partial house gassing specified that if the pellets are small enough, the birds show no aversive reactions nor frostburns.

When using N2, the expert concerned reported that between 1 and 5% of the birds (Gallus gallus and turkeys) present frostburns at the end of HG (n_{ans}=2/2 according to 1 expert, n_{unk}=0/2, n_{exp}=1). The percentage of birds presenting frostburns according to the species at stake and the type of gas mixture used is detailed in *Figure 9*.

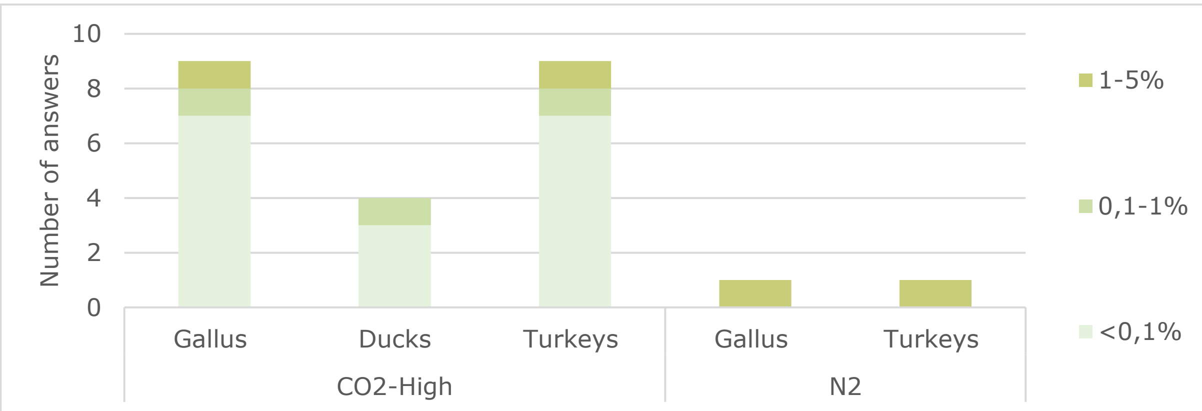


Figure 9: Number of answers obtained for different classes of percentage of birds that presented frostburns at the end of HG, according to the species at stake and type of gas mixture used. CO2-High: CO_2 in high concentration, N2: Nitrogen alone

During the workshop, experts were asked for an estimation of the temperature in the barn when the birds start losing consciousness – in cases where the gas is not preheated when injected.

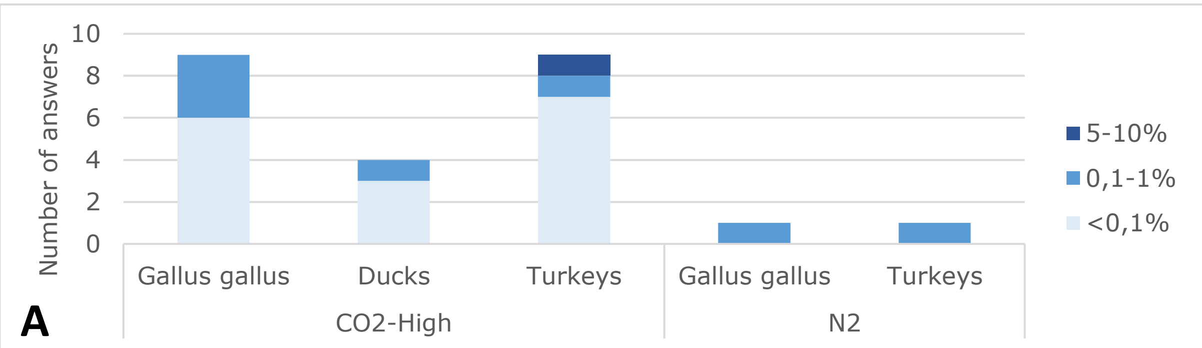
Most experts could not answer the question and specified not using thermometers inside the barn. That being said, two experts gave informative answers and indicated temperatures around 10°C, with a minimum of 5°C when the CO_2 concentration is quickly raised.

Efficiency

Regarding the **percentage of birds remaining conscious**, most answers indicate that less than 0.1% of the birds remain conscious at the end of the procedure when CO2-High is used, whichever the species at stake (n_{ans}=16/22 according to 7 experts and n_{unk}=3/25 according to 2 experts, n_{exp}=11). Of note, one expert still reported that between 5 and 10% of the turkeys would remain conscious at the end of the procedure. When using N2, the only concerned expert reported that between 0.1 and 1% of Gallus *gallus* and turkeys remain conscious at the end of the procedure. *Figure 10A* details the percentage of birds remaining conscious at the end of the procedure, according to the species at stake and the type of gas mixture used.

Regarding the **percentage of birds regaining consciousness**, most answers indicate that less than 0.1% of the birds regain consciousness at the end of the procedure when CO2-High is used, whichever the species at stake (n_{ans}=16/22 according to 7 experts and n_{unk}=3/25 according to 2 experts, n_{exp}=11). Of note, one expert still reported that between 5 and 10% of the turkeys would regain consciousness at the end of the procedure. When using N2, the only concerned expert reported that less than 0.1% of Gallus *gallus* and turkeys regain consciousness at the end of the procedure. *Figure 10B* details the percentage of birds regaining consciousness at the end of the procedure, according to the species at stake and the type of gas mixture used.

Regarding the **percentage of birds remaining unconscious but still alive**, most answers suggest that less than 0.1% of the birds remain unconscious but alive at the end of the procedure, whichever the species at stake and the gas mixture used (n_{ans}=21/24 according to 8 experts and n_{unk}=3/27 according to 2 experts, n_{exp}=11). When CO2-High is used, the highest percentage intervals reported for Gallus *gallus* and ducks were comprised between 1 and 5%, and between 5 and 10% for turkeys. *Figure 10C* details the percentage of unconscious but alive birds at the end of the procedure, according to the species at stake and the type of gas mixture used.



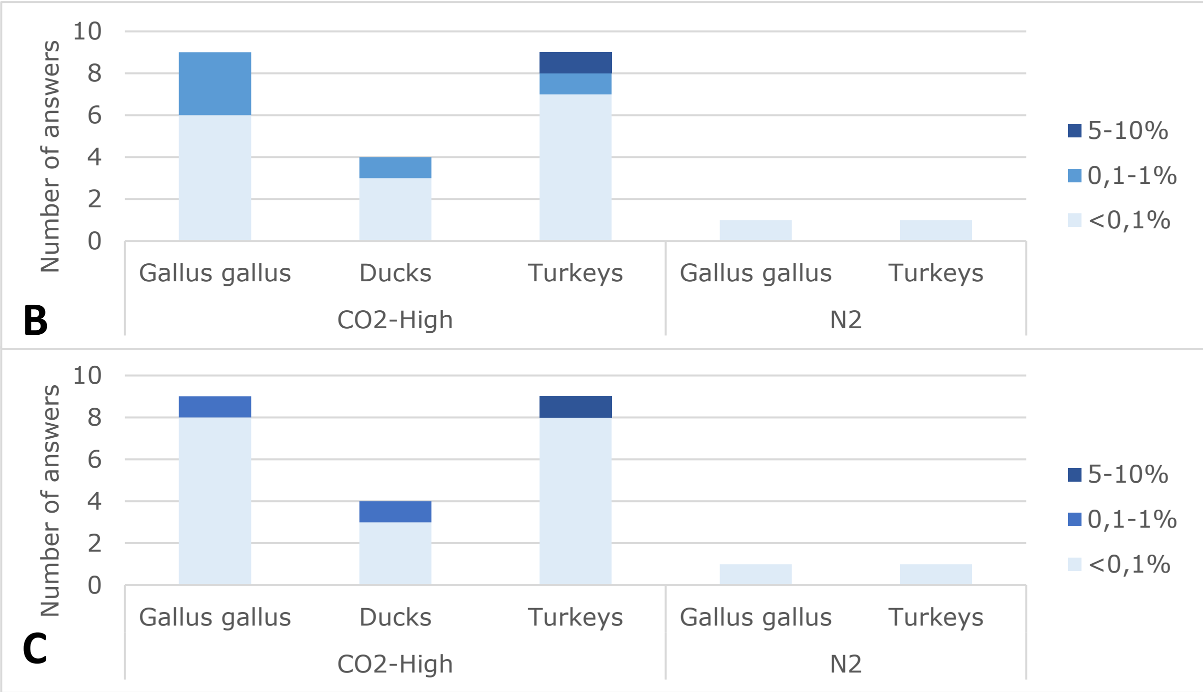


Figure 10: Number of answers obtained for different classes of percentage of birds that are conscious (A), have regained consciousness (B) or are unconscious but alive (C) at the end of the procedure according to the species at stake and type of gas mixture used. CO2-High: CO_2 in high concentration, N2: Nitrogen alone

Regarding the **latency for unconsciousness induction of the first and last bird** once the gassing mixture is injected, only one expert gave informative answers (the 10 others only replied "unknown" whichever the species at stake and the gas used). This expert reported the same durations for the first and the last bird to become unconscious, with an induction time inferior to 1 min for Gallus *gallus* and ducks, and comprised between 1 and 5 min for turkeys. During the workshop, experts were asked for which reasons unconsciousness induction was difficult to assess in practice. All experts who replied indicated that it was not possible to visually inspect the birds during the procedure because of the fog.

Regarding the **latency for death induction for the first bird** once the gas started to be injected, most answers indicate a death induction within 15 min when CO2-High is used, whichever the species at stake (n_{ans}=18/20 according to 7 experts, n_{unk}=5/25 according to 3 experts, n_{exp}=11). At the species level, durations range between 1-5 min (n_{ans}=8/16 according to 4 experts, n_{unk}=5/21 according to 3 experts, n_{exp}=11) and more than 45 min (n_{ans}=2/16 according to one expert , n_{unk}=5/21 according to 3 experts, n_{exp}=11) for Gallus *gallus* and turkeys, and between 1-5 min (n_{ans}=1/4 according to 1 expert, n_{unk}=0/4, n_{exp}=4) and 5-15 min (n_{ans}=3/4 according to 3 experts, n_{unk}=0/4, n_{exp}=4) for ducks. When N2 is used, the only concerned expert reported a death induction time superior to 45 min for Gallus *gallus* and turkeys.

Regarding the **latency for death induction for the last bird** once the gas is inserted within the house, most answers indicate that the death induction occurs within 30 min when CO2-High is used, whichever the species at stake (n_{ans}=16/18 according to 6 experts, n_{unk}=7/25 according to 4 experts, n_{exp}=11). At the species level, durations range between 1-5 min (n_{ans}=2/14 according to 1 expert, n_{unk}=7/21 according to 4 experts, n_{exp}=11) and more than 45min (n_{ans}=2/14 according to one expert , n_{unk}=7/21 according to 4 experts, n_{exp}=11) for Gallus *gallus* and turkeys, and between 1-5 min (n_{ans}=1/4 according to 1 expert, n_{unk}=0/4, n_{exp}=4) and 15-30 min (n_{ans}=2/4 according to 2 experts, n_{unk}=0/4, n_{exp}=4) for ducks. When N2 is used, the only concerned expert reported a death induction time above 45 min for Gallus *gallus* and turkeys. Of note, it is the same expert who mentioned a time above 45 min for both CO2-High and N2. *Figure 11* **details the time for death induction** for the **first (A**) and the **last** bird (**B**) according to the species at stake and the type of gas mixture used.

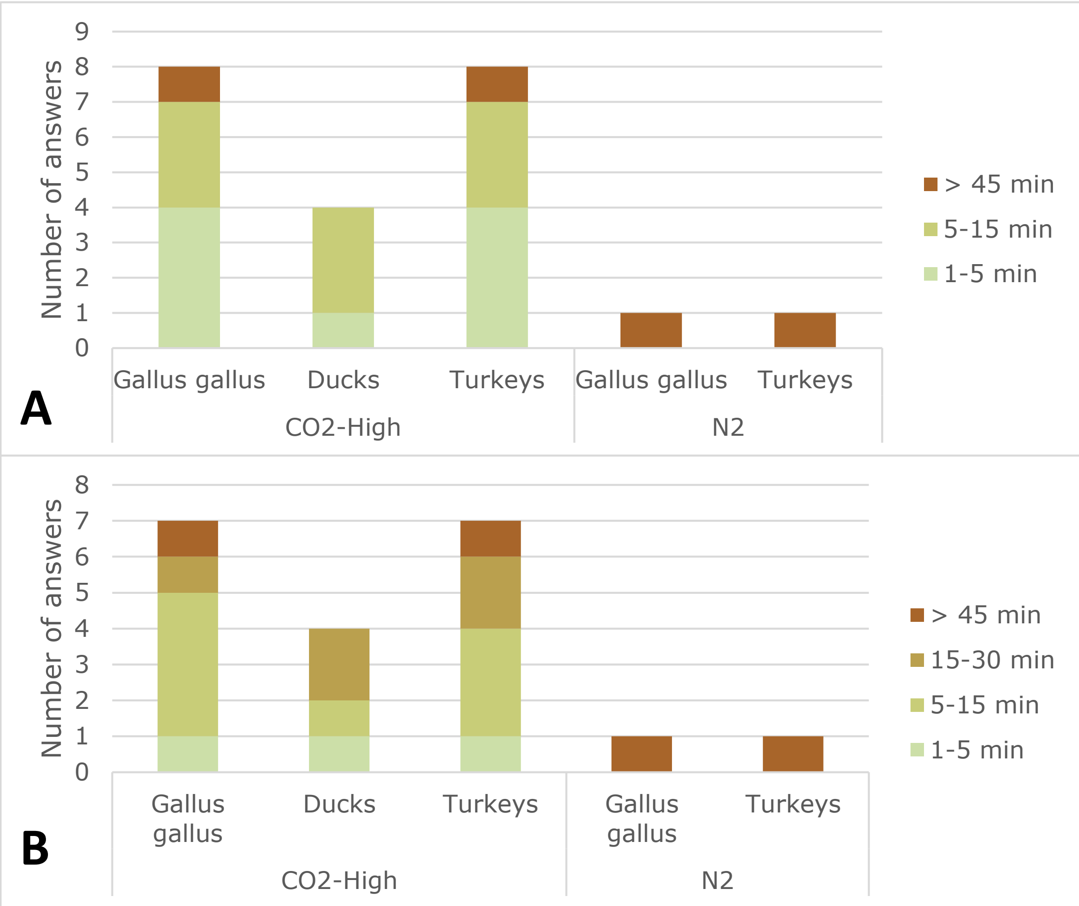


Figure 11. Number of answers obtained for different classes of latency for death induction of the first (A) and last bird (B) of the same batch according to the species at stake and type of gas used. CO2-High: CO_2 in high concentration, N2: Nitrogen alone

Regarding the **percentage of birds piled up** at the end of the procedure, most answers indicate that less than 1% of Gallus *gallus* and turkeys (n_{ans}=13/18 according to 7 experts, n_{unk}=3/21 according to 2 experts, n_{exp}=11), and less than 5% of ducks (n_{ans}=3/4 according to 3 expert, n_{unk}=0/4, n_{exp}=4) were observed piled-up at the end of the procedure when using CO2-

High. The highest percentage intervals reported are comprised between 10 and 20% for Gallus *gallus* and ducks, and between 30 and 35% for turkeys. During the workshop, clarifications were asked to experts from the MS who had reported the extreme values for turkeys. The expert who reported observing 30 to 35% of turkeys piled-up after using a CO2-High explained that this high value was due to the use of CO_2 pellets (in partial house gassing), with a longer time to reach target concentration at head level of turkeys than other animals due to their height. The expert who reported observing less than 0.1% of turkeys piled-up explained that this low value was due to turkeys being kept in smaller flocks than other species, hence less piling-up. When using N2, the only concerned expert reported that between 0.1 and 1% of Gallus *gallus* and turkeys were observed piled-up at the end of the procedure. The *Figure 12* details the percentage of piled-up birds according to the species at stake and the gas mixture used.

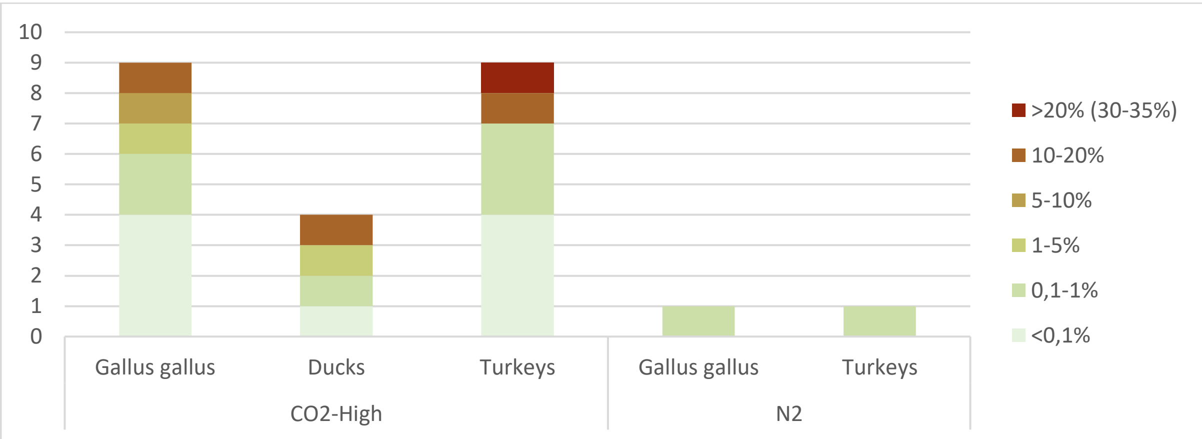


Figure 12: Number of answers obtained for different classes of percentage of birds observed piled-up at the end of the procedure according to the species at stake and type of gas used. CO2-High: CO_2 in high concentration, N2: Nitrogen alone

During the workshop, experts were asked to list the main factors that have an impact on the piling-up of birds. Most answers mentioned the injection speed (i.e. the quicker the flow the lower the proportion of birds piling-up because less panic movements have time to arise) and other answers mentioned environmental parameters (lights on, improper sealing, flock size, orientation of the gas flow towards the bird).

When birds are still alive at the end of HG, different **back-up methods** are used to ensure their death; mostly cervical dislocation (reported by 11 experts, with or without the use of a stunning method first) and lethal injection (reported by 3 experts). The use of a gunshot (reported by 1 expert), has also been mentioned.

Discussion and conclusion

Results indicate that HG is mostly performed on large flocks of birds, on any species or housing systems at stake. Two main types of HG have been reported to be used in the EU: whole house gassing (predominantly used) and partial house gassing (used by at least 2 MS), for which only a part of the barn is gassed. Scarce information was obtained on partial house gassing, specifically.

Concerning the **gas mixtures used**, CO_2 is generally administered – often in a liquid form with no pre-heating treatment. CO_2 is more often used than N2 as the sole constituent of the gas mixture administered. Although gas mixtures can be injected in various forms, most experts reported that they injected it without pre-heating, mainly owing to their MS lacking adequate preheating equipment and savoir-faire. Preheating the gas may delay the start of the procedure, but speeds up gas delivery because outlet pipes will not have to be closed to avoid freezing (personal communication from Gerritzen, 2024). The lack of pre-heating treatment goes against some researchers' recommendation as it results in chilling and nasal damage in birds when they inhale the cold gas (e.g., Raj, Sandilands, & Sparks, 2006). In line with this observation, one expert observed up to 5% of birds suffering from frostburns following whole-house gassing without preheating. In the survey, however, most experts reported that less than 0.1% of birds suffered from frostburns, whether the gas was pre-heated or not. CO_2 administered in pellets did not appear to cause frostburns either. There is debate about the welfare implications of using liquid CO_2 (Raj, Sandilands, & Sparks, 2006, Sparks et al., 2010), mostly because it is unsure whether the birds are conscious while experiencing very low temperatures during whole house gassing.

The **target concentrations** aimed at within the barn and the **time taken to reach said concentrations** vary on-field. This observed heterogeneity can stem from the fact that various factors (and their modalities) influence the speed at which the gas reaches the target concentration. Besides the injection, the filling speed can be influenced by many factors – besides the injection rate and the potential gas leakage – such as the state of the injected gas, the number of injection points, and the configuration of the barn. Injecting vaporized gas compared to liquid gas speeds up gas delivery (personal communication from Gerritzen, 2024). Likewise, using multiple injection points accelerates gas delivery and furthermore helps homogenize gas concentration in the gassed volume. The configuration of the barn may also influence the filling speed: one expert specified that when the ground of the building showed a great slope, the gas accumulates at the lowest point of the slope – making it apparently harder to reach the targeted concentration at the highest point of the barn. That being said, other experts specified successfully reaching their target concentration at any point in space, even in multi-tier systems.

To prepare an effective depopulation operation, some experts reported to **shut the ventilation down immediately** before injecting the gas mixture. This prevents the building from overheating (which happens when shutting the ventilation down earlier), as recommended by Raj et al. (2006) and additionally prevents ammonia and CO_2 from reacting together, thereby slowing down the CO_2 concentration increase.

One of the **greatest animal-welfare challenges** associated with the use of HG **is reaching the target gas concentration inside the house** in the shortest possible duration. Filling speed of gas, which appears to vary greatly from one depopulation site to another, is linked to animal welfare as it controls the duration for which conscious birds are exposed to variably aversive concentrations of gas. Increasing the speed of gas delivery (as well as turning off the lights) was reported to reduce the number of piled-up birds observed by some MS.

The main **limiting factor for the assessment of animal welfare** during HC **is fogging**. In fact, gas vaporization creates a fog which prevents operators from observing the birds during the procedure. As a result, operators are unable to assess unconsciousness and death onset, as well as the welfare issues occurring during the procedure, such as tachypnea and deep-breathing. The heterogeneity in the answers obtained from the survey probably reflects a high uncertainty of the respondents in their own answers. Alternative ways of assessing (un)consciousness despite fog should be investigated (e.g., by using infrared videography as used by Turner et al., 2012 for whole-house gassing with liquid CO_2, or by putting sensors on some birds).

Despite the aforementioned welfare concerns, **house gassing without foam remains an effective method**. Almost all experts reported less than 0.1% of birds regaining consciousness, being unconscious but alive or remaining conscious at the end of the procedure.

Lethal injection

Depopulation context

From the 14 MS who reported the use of Lethal Injection (**LI**) since 2018, the depopulation experts from 11 MS answered the method-specific survey with a **total of 12 answers** (two experts from one MS replied). The **three most common reasons given** for using LI specifically are 1) the (relatively low) number of birds in the flock, 2) the practicality of the method and 3) the species at stake. LI is sometimes used as a routine method (n_{ans}=4/12 according to 4 experts, n_{unk}=0/12, n_{exp}=12).

Across the EU, LI is used in **various types of housing systems**: single-tier (as reported by 7 experts), multi-tier (as reported by 3 experts) and cages (also reported by 3 experts). Most experts reported using LI **exclusively on small flocks** (i.e., of less than 250 birds) (n_{ans}=8/12 according to 8 experts, n_{unk}=0/12, n_{exp}=12) – or exclusively on small flocks used for recreational purposes (n_{ans}=2/12 according to 2 experts, n_{unk}=0/12, n_{exp}=12). Incidentally, two experts reported using LI to depopulate flocks of 250 to 5000 birds.

The same number of experts (9) have reported LI to be used on **Gallus *gallus* and ducks**. As a reminder, turkeys were not included in the scope of this method-specific survey.

Operating procedures

In the EU, two **types of drugs** were reported to be used to perform LI on both Gallus *gallus* and ducks: Pentobarbital Sodium (hereafter referred to as **Pento**, n_{ans}=11/18 according to 6 experts, n_{unk}=6/24 according to 3 experts, n_{exp}=12), and **T61** (Tanax®) a mixture of embutramide, mebezonium, and tetracaine (n_{ans}=7/18 according to 4 experts, n_{unk}=6/24 according to 3 experts, n_{exp}=12). Of note, one expert indicated that any kind of authorized drugs could be used – and that the choice was made at the veterinarian's discretion. Also, one expert specified injecting Torbugesic, Xylazin, and a combination of Tilatemin with Zolacepam as anesthetic before intraperitoneal injection with T61 or Sodium Pentobarbital. *Figure 13* details the type of drug used according to the depopulated species. During the workshop, one expert specified that the product used for pentobarbital injection in their MS is “Realease®”, which is both used on turkeys and geese. Only veterinarians appear to be authorized to perform LI on birds.

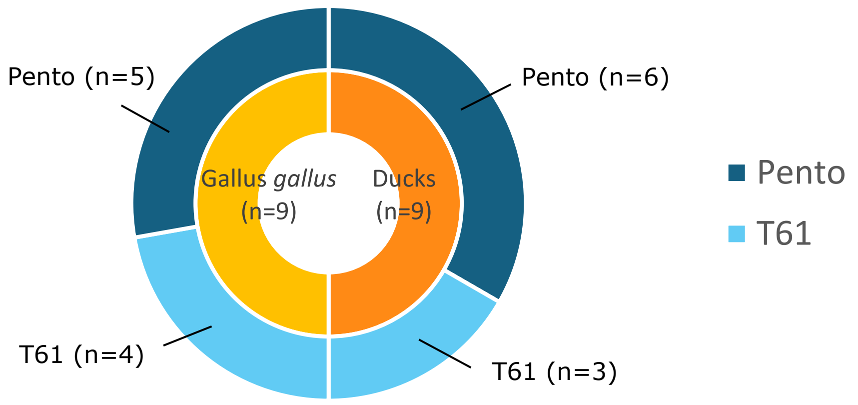


Figure 13: Drug used according to the depopulated species. Pento: Pentobarbital sodium

Concerning the **routes used**, Pento and T61 are most often injected intraperitoneally, whichever the species at stake (n_{ans}=11/30 according to 4 experts, n_{unk}=1/31 according to 1 expert, n_{exp}=8), but they can also be injected intracardially (n_{ans}=6/30 according to 2 experts, n_{unk}=1/31 , according to 1 expert, n_{exp}=8), intrapulmonary (n_{ans}=7/30 according to 3 experts, n_{unk}=1/31 according to 1 expert, n_{exp}=8) and intravenously (n_{ans}=4/30 according to 1 expert, n_{unk}=1/31 according to 1 expert, n_{exp}=8). One expert also reported that T61 is injected through the sinus occipital (n_{ans}=2/2 according to 1 expert, n_{unk}=0/2 n_{exp}=1). Of note, the expert that mentioned injecting Xylazine as an anaesthetic specified injecting it intramuscularly on both Gallus *gallus* and ducks (n_{ans}=2/2 according to 1 expert, n_{unk}=0/2 n_{exp}=1)). For more details on administration routes of these drugs, please report to *Figure 14*.

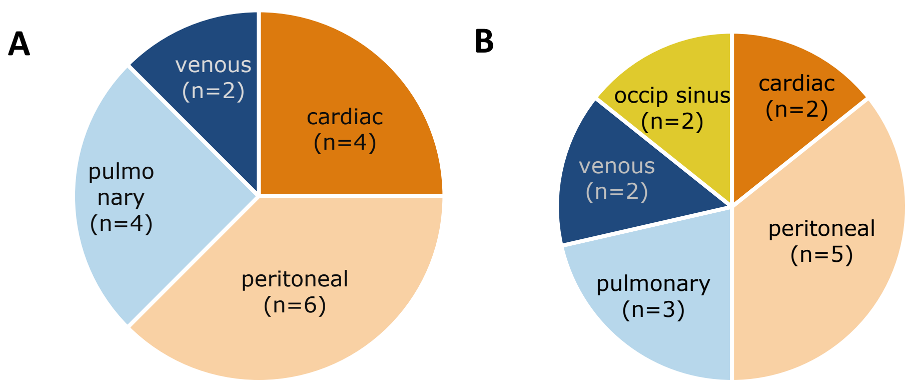


Figure 14: Administration routes reported when Sodium Pentobarbital is used (A), and when T61 is used (B), for Gallus gallus and ducks. peritoneal: intraperitoneal injection, cardiac: intracardiac injection, pulmonary: intrapulmonary injection, venous: intravenous injection, occip sinus: injection in occipital sinus

Regarding the **lethal dose used**, only one answer was obtained for each drug administered in a specific way on a given species. For Pento, 3 mL where reported to be used when injected intracardially (unreported dosage), and 750 mg/kg when injected intrapulmonary – whichever the species at stake. For T61, 1 mL was reported to be used on Gallus *gallus* (no value reported for ducks*)* when administered intraperitoneally or intrapulmonary (unreported dosage). When T61 is administered via the occipital sinus, 0,3 mL and 0,4 mL were reported for Gallus *gallus* and ducks, respectively (unreported dosage). No dosage was specified for Pento administered intraperitoneally or intravenously, T61 administered intracardially or intravenously.

Welfare implications

The main welfare consequences associated with LI reported by the respondents were the handling and restraint of the birds (by 6 experts) and the correct needle placement in order to hit the vein and avoid hitting the air sacs (by 5 experts). Other welfare aspects were mentioned by single experts: a too low dosage, or the absence of an anesthetic use before an intraperitoneal injection of Pento or T61.

Welfare implications associated with the preparation of the killing

Food and water were never reported to be removed before the procedure (n_{ans}=0/7, n_{unk}=5/12 according to 5 expert, n_{exp}=12 for both food and water). Most experts reported that **birds were brought to a more confined space** (e.g., specific part of the barn or put in crates/boxes) before the start of the procedure (n_{ans}=6/9 according to 6 experts, n_{unk}=3/12 according to 3 expert, n_{exp}=12). However, only one expert reported that the birds were **transported to a specific location to be killed** (n_{ans}=1/9, n_{unk}=3/12, n_{exp}=12).

Welfare issues occurring during the depopulation procedure

In practice, Pento is injected on conscious (n_{ans}=4/8 according to 2 experts, n_{unk}=2/6 according to 1 expert, n_{exp}=6) and unconscious birds (n_{ans}=4/8 according to 2 experts) whichever the species at stake. When T61 is used, it is reported to be mostly injected (although not systematically) on unconscious birds, whichever the species at stake (n_{ans}=4/6 according to 2 experts, n_{unk}=1/7 according to 1 expert, n_{exp}=4).

Efficiency

Regarding the **latency for unconsciousness induction**, only one answer was obtained for each drug administered in a specific way on a given species. Whichever the species at stake, experts reported immediate loss of consciousness when Pento is injected intracardially, (n_{ans}=2/2 according to 1 expert, n_{unk}=2/4 according to 1 expert, n_{exp}=2) and loss of consciousness within

30 s when injected intrapulmonary (n_{ans}=2/2 according to 1 expert, n_{unk}=2/4 according to 1 expert, n_{exp}=2). When T61 is injected intraperitoneally, the expert reported that the loss of consciousness of Gallus *gallus* (no answer given for ducks) occurs between 30 and 60 s. When injected intrapulmonary in Gallus *gallus* (no answer for ducks) or via the occipital sinus in ducks (no answer for Gallus *gallus*), experts reported loss of consciousness within 30 s (n_{ans}=2/2 according to 2 experts, n_{unk}=1/3 according to 1 expert, n_{exp}=3). No answer was obtained for Pento administered intraperitoneally or intravenously, T61 administered intracardially or intravenously.

Regarding the **latency of death induction**, experts exclusively reported that the death occurs within 30 s when Pento and T61 are injected intrapulmonary, intracardially, and intravenously – whichever the species at stake (n_{ans}=19/19 according to 5 experts, n_{unk}=0/19, n_{exp}=5) or for T61 injected in the occipital sinus (n_{ans}=2/2 according to 1 expert, n_{unk}=0/2, n_{exp}=1). When injected intraperitoneally, answers reported that death induction occurs within 30 s (n_{ans}=4/9 according to 1 expert, n_{unk}=2/11 according to 1 expert, n_{exp}=4) or between 30 to 60 s (n_{ans}=5/9 according to 2 experts, n_{unk}=2/11 according to 1 expert, n_{exp}=4), whichever the species at stake, for both Pento and T61.

Regarding the **percentage of birds remaining conscious** at the end of the procedure, only one answer was obtained for each drug administered in a specific way on a given species. When Pento is injected intrapulmonary and intracardially, experts reported that less than 0.1% of the birds remain conscious at the end of the procedure, whichever the species at stake (n_{ans}=4/4 according to 2 experts, n_{unk}=4/8 according to 1 expert, n_{exp}=3). When T61 is injected intraperitoneally or intrapulmonary on Gallus *gallus* (no result for ducks), the expert reported that 1 to 5% of the birds remain conscious at the end of the procedure (n_{ans}=2/2 according to 1 expert, n_{unk}=3/5 according to 2 experts, n_{exp}=3). No result was obtained for Pento administered intraperitoneally, intravenously, intracardially or intrapulmonary, nor for T61 administered intracardially, intrapulmonary, intraperitoneally, intravenously or via the occipital sinus.

Regarding the **percentage of birds regaining consciousness** after LI, only one answer was obtained for each drug administered in a specific way on a given species. When Pento is injected intracardially and intrapulmonary, the experts reported that less than 0.1% of birds regain consciousness after the injection, whichever the species at stake (n_{ans}=4/4 according to 2 experts, n_{unk}=4/8 according to 1 expert, n_{exp}=3). When T61 is injected intracardially or intrapulmonary on Gallus *gallus* (no answer obtained for ducks), the experts also reported that less than 0.1% of birds regain consciousness after the injection – whichever the species (n_{ans}=1/1 according to 1 expert, n_{unk}=2/3 according to 1 expert, n_{exp}=2). No answer was obtained for Pento administered intravenously or intraperitoneally, nor for T61 administered intracardially, intravenously or via the occipital sinus.

Regarding **the percentage of birds that are being unconscious but still alive** at the end of the procedure, all experts reported that less than 0.1% of the birds were unconscious but alive at the end of procedure when Pento is used – whichever the species at stake and the route of administration (n_{ans}=16/16 according to 5 experts, n_{unk}=0/16, n_{exp}=5). When T61 is injected intracardially, intrapulmonary and intravenously, experts also reported that less than 0.1% of the birds were unconscious but alive at the end of procedure – whichever the species at stake (n_{ans}=7/7 according to 2 experts, n_{unk}=0/7, n_{exp}=2). For T61 injected intraperitoneally, most answers suggested that less than 0.1% of the birds are unconscious but alive at the end of the procedure (n_{ans}=4/5 according to 2 experts, n_{unk}=0/5, n_{exp}=3), although one expert reported that 0.1 to 1% of Gallus *gallus* remain alive. When T61 is injected in the occipital sinus, the expert reported that 0.1 to 1% of the birds remain unconscious but alive at the end of the procedure whichever the species at stake (n_{ans}=2/2 according to 1 expert, n_{unk}=0/2, n_{exp}=1).

Regarding the **killing rate**, answers indicate throughputs comprised between 15 and 60 birds per hour when Pento is used – whichever the species or administration route (n_{ans}=16/16 according to 5 experts, n_{unk}=0/16, n_{exp}=5). The throughput is comprised in the upper range of the interval (40-60 birds per hour) when Pento is administered in intracardiac (n_{ans}=4/4 according to 2 experts, n_{unk}=0/4, n_{exp}=2), and in the lower range of the interval (15-40 birds per hour) when Pento is intrapulmonary injected (n_{ans}=4/4 according to 2 experts, n_{unk}=0/4, n_{exp}=2). When T61 is used, throughputs of 40 birds per hour were reported for intracardiac and intravenous injections on birds – whichever the species (n_{ans}=4/4 according to 1 expert, n_{unk}=0/4, n_{exp}=1). When T61 is intraperitoneally administered, reported throughputs range between 20-100 Gallus *gallus* per hour (n_{ans}=3/3 according to 3 experts, n_{unk}=0/3, n_{exp}=3) and 20-40 ducks per hour (n_{ans}=2/2 according to 2 experts, n_{unk}=0/2, n_{exp}=2). When T61 is intrapulmonary administered, answers indicate throughputs comprised between 40 and 100 Gallus *gallus* per hour (n_{ans}=2/2 according to 2 experts, n_{unk}=0/2, n_{exp}=2) and a throughput of 40 ducks per hour (n_{ans}=1/1 according to 1 expert, n_{unk}=0/1, n_{exp}=1). When T61 is administered in the occipital sinus, throughputs range from 200 Gallus *gallus* to 400 ducks per operator per hour ((n_{ans}=2/2 according to 2 experts, n_{unk}=0/2, n_{exp}=2). *Figure 15* details the number of birds killed per hour according to the drug and the route used.

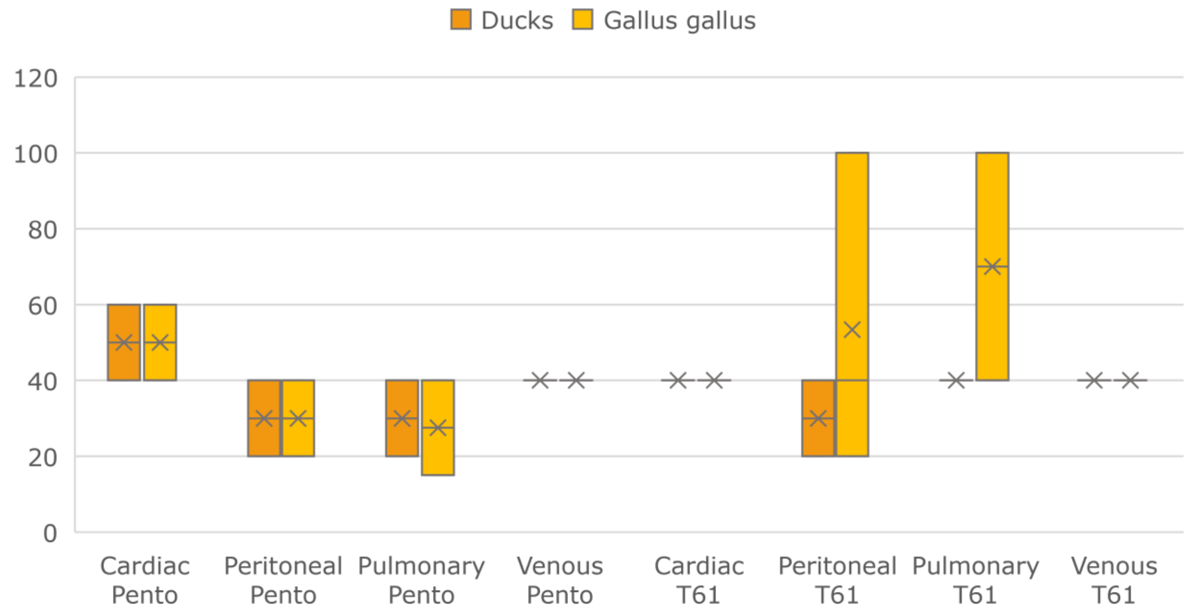


Figure 15: Boxplot of the number of birds killed per hour according to the route and drug used. Each box represents the extreme values (minimum and maximum) reported. The mean value is represented by a

cross, and the median (exclusive) by a horizontal line. Occipital sinus injection of T61 was not represented to prevent scale compression. Cardiac: intracardiac injection, Peritoneal: intraperitoneal injection, Pulmonary: intrapulmonary injection, Venous: intravenous injection. Pento: Pentobarbital Sodium

When birds are still alive at the end of LI, different **back-up methods** are reported to be used to ensure their death. These methods include (from the most reported to the least reported): cervical dislocation (reported by 7 experts), an additional lethal injection (reported by 6 experts), and decapitation (reported by 2 experts).

Discussion and conclusion

Results indicate that LI is used on all categories of birds housed in any type of housing systems, and mostly on very small flocks (< 250 birds).

Two drugs – Pentobarbital Sodium (Pento) and T61 – are typically injected to cull birds, conscious or not. From the results obtained in response to the survey (which may not be representative to all practices used throughout the EU), Pentobarbital Sodium appears to be as often injected on conscious as unconscious birds, whereas T61 appears to be mostly used on unconscious birds. As a reminder, **T61 should not be used on conscious animals** (ANSM, 2014, EFSA 2019) as it triggers pain and irritation in birds with a rapid injection, and the cessation of breathing may occur before the loss of consciousness (EFSA 2004). The administration route can also greatly influence the welfare impact of lethal injection, depending on the drug used.

Various routes of administration are used for both drugs, which have different consequences on animal welfare. Pento and T61 have mostly been reported to be administered intraperitoneally and intrapulmonary, although scientific evidence advises against these administration routes on conscious animals (AVMA, 2020; Animal Health Australia, 2015). Whatever the drug used, the induction of **death is delayed with intraperitoneal and intrapulmonary injections** compared to intravenous methods (USDA APHIS, 2015, AVMA 2020, AAVAC, n.d.). In line with the preceding idea, results from the survey indicate that unconsciousness and death induction latencies were the longest when intraperitoneal route was used. We thus do not recommend injecting Pentobarbital intraperitoneally. Incidentally, intracardiac injection was also reported to be used by 2 MS, sometimes on conscious animals. However, intracardiac injections should only be used on unconscious or anesthetized birds (AVMA, 2020, McGill University Animal Policy and Welfare Oversight Committee, 2021) because of the difficulty and unpredictability of accurately performing the injection (AVMA, 2007). In practice, intravenous injections and injections via the occipital sinus were rarely reported to be used. In literature, the intravenous route is, yet, the recommended one because of its efficacy and the lack of pain response that it triggers (EFSA, 2019, AVMA, 2020).

Concerning the **efficiency of the drugs**, both Pento and T61 seem effective. In fact, most experts reported less than 0.1% of birds remaining conscious, regaining consciousness or being unconscious but alive at the end of the procedure. That being said, when T61 is injected intraperitoneally or intrapulmonary on Gallus *gallus*, one expert still reported that 1 to 5% of the birds remained conscious at the end of the procedure, which can be considered a high proportion of birds.

Most injection routes presented similar throughputs, except for injection in the occipital sinus. Throughputs are inferior to 100 birds per operator per hour for intracardiac, intraperitoneal, intrapulmonary and intravenous injection. Throughputs for injection in the occipital sinus stand out with reported killing rates of 200 Gallus *gallus* and 400 ducks per operator per hour (when T61 is used). Injection via the occipital sinus hence appears very effective. Interestingly, however, no expert reported injecting Pento via the occipital sinus. It can, however, be hypothesized that injecting Pento via the occipital sinus might trigger less pain reaction than T61 when applied on conscious birds. Field knowledge on the practical requirements for lethal injection in the occipital sinus during a large-scale depopulation procedure should be looked for and analyzed, to determine if this method can be recommended as a good practice.

Cervical dislocation

Depopulation context

From the 15 MS who reported the use of Cervical Dislocation (**CD**) since 2018, the depopulation experts from 10 MS answered the method-specific survey with a total of **11 answers** (two experts from one MS replied). The **three most common reasons** given for using CD specifically are 1) the number of birds in the flock, 2) practicality and 3) the species at stake. CD is systematically reported to be a non-routine method (n_{ans}=10/10 according to 11 experts, n_{unk}=1/11, n_{exp}=11).

Across the EU, CD is reported to be used in **various types of housing system**: cages and multi-tier systems (as reported by 4 experts, each), single-tier systems (as reported by 3 experts). Of note, four experts reported using CD on "other" systems – namely "backyard" flocks. CD is reported to be used **exclusively on small flocks** (< 250 birds) by almost all experts (n_{ans}=10/11 according to 11 experts, n_{unk}=0/11, n_{exp}=11) with one expert reporting its use for flocks of 251-500 birds as well.

CD has been reported to be used on **all the animal species under study** (from most to least often): Gallus *gallus (*i.e. broilers and laying hens, by 8 experts), ducks (by 6 experts) and turkeys (by 4 experts).

Operating procedures

Five experts reported using only manual CD, and five reported using **both mechanical and manual CD**. It is unsure if manual or mechanical CD is used exclusively on certain species. The five experts who reported using mechanical CD specified using "mechanical tongs". Among the ten experts who reported using manual CD, nine experts specified applying it on birds below a certain weight limit (n_{ans}=9/9 according to 9 experts, n_{unk}=1/10 according to 1 expert, n_{exp}=10): 3 kg (5 answers), 4 kg (1 answer), and 5 kg (3 answers). The expert reporting using manual CD on birds below 5 kg mentioned using CD solely as a back-up method for HG, and in accordance with a specific national derogation.

Among the five experts who reported using mechanical CD, three experts specified applying it on birds below a certain weight limit (5 kg) (n_{ans}=3/5 according to 3 experts, n_{unk}=0/5, n_{exp}=5). That being said, one of the three experts specifically reported using mechanical CD on some birds with weight superior to 5 kg, in accordance with a granted derogation.

All experts reported that **no anesthetics are administered before** the procedure to induce unconsciousness, whichever the species at stake (n_{ans}=18/18 according to 8 experts, n_{unk}=0/18, n_{exp}=8).

Welfare implications

For most experts, the main welfare consequences associated with CD is the need to handle the birds. Three experts emphasized the necessity of having capable staff available (i.e. trained staff, and not fatigued by an extensive number of birds to deal with). Among these three experts, two of them specified that there would be no animal welfare issue at all (if the procedure was correctly done) if capable staff were to kill the birds.

Welfare implications associated with the preparation of the killing

Only one expert (out of the 11 experts who answered the survey) stated that **food and water were removed** before the beginning of the procedure, within 2 hours before its start.

Welfare implications associated with animal handling

Birds are not often transported from their barn to a specific location to be killed (n_{ans}=3/9 according to 3 experts, n_{unk}=2/11 according to 2 experts, n_{exp}=11). When it occurs, birds are caught and transported manually, in an upward position. One expert specified holding three Gallus *gallus* or two ducks per hand when transporting birds manually.

Most often, it appears that birds are restrained immediately before CD, whichever the species at stake (n_{ans}=11/18 according to 5 experts, n_{unk}=0/18, n_{exp}=8). Still, for each species, some experts reported that the birds could be restrained for up to 5 min (n_{ans}=7/18 according to 3 experts, n_{unk}=0/18, n_{exp}=8) prior to CD. Whether differences exist in terms of restraining duration between manual and mechanical cervical dislocation remains unknown.

Efficiency

Regarding the **latency of unconsciousness induction**, most experts reported that both manual and mechanical CD induce immediate loss of consciousness, whichever the species at stake (n_{ans}=15/17 according to 5 experts, n_{unk}=10/27 according to 2 experts, n_{exp}=8). That being said, one expert reported a duration between 30 to 60 s for Gallus *gallus* and within 30 s for turkeys when using manual CD.

Regarding **the latency of death induction**, most experts using manual CD reported an immediate induction of death for Gallus *gallus* and ducks (n_{ans}=9/10 according to 5 experts, n_{unk}=4/14 according to 2 experts, n_{exp}=8). Of note, one expert still reported that Gallus *gallus* were to die between 30 to 60 s after the application of CD. When using manual CD on turkeys, most experts reported that turkeys die within 30 s (n_{ans}=2/3 according to 2 experts, n_{unk}=1/4 according to 1 expert, n_{exp}=4), and one expert reported immediate death upon CD application. All experts using mechanical CD reported an "immediate" death of the birds upon CD application, whichever the species at stake (n_{ans}=4/4 according to 2 experts, n_{unk}=5/9 according to 2 experts, n_{exp}=4).

All experts reported that less than 0.1% of birds **remain conscious, regain consciousness or remain unconscious but still alive** once the procedure has been applied – whichever the type of CD performed (i.e., manual or mechanical) and the species at stake (n_{ans}=17/17 according to 6 experts, n_{unk}=10/27 according to 2 experts, n_{exp}=8). Of note, 8 out of 13 answers from experts familiar with manual CD were informative answers (i.e. not "unknown"), compared to only 4 out of 9 answers from experts familiar with mechanical CD.

When birds are still alive at the end of CD, different **back-up methods** are reported to be used to ensure their death. These methods include (from the most reported to the least reported): additional cervical dislocation (reported by 10 experts), lethal injection (reported by 3 experts), and gunshot (reported by 1 expert). Neck-cutting, decapitation, electrical waterbath, and gassing were never reported as a backup method for CD.

Discussion and conclusion

Results from the survey indicate that CD is used to kill all species under studies – although apparently less frequently turkeys than Gallus *gallus* and ducks. CD is used almost exclusively on very small flocks (<250 birds), in all types of housing system.

Manual CD was reported more frequently to be used than mechanical CD, with distinct weight thresholds. All ten experts reported using manual CD, generally on birds lighter than 3 kg. Among those 10 experts, five also reported using mechanical CD (with mechanical tongs). However, the specific tools used for mechanical CD could not be determined in the survey. Contrary to Council Regulation 1099, some experts reported culling birds weighing more than 3 kg (between 3 and 5 kg) by manual CD, and birds weighing more than 5 kg by mechanical CD – but they reported doing so in accordance with national derogations. Handling heavier birds is likely to cause more fatigue, which renders operators more prone to making mistakes.

Whichever the type of CD used, **birds need to be restrained during the procedure** which may have welfare consequences also. Most often, birds are restrained immediately before CD but three experts reported restraining them for up to 5 min before the procedure. When restrained in cones, birds are inverted and compressed. Therefore, restraining them for such a long time may cause welfare issues such as fear or pain, especially as they may suffocate (Poultry Industry Council, 2016). When birds are held in a cone, the Human Slaughter Association - hereafter referred to as HSA - (2013) recommends a maximum restraining time of 1 min for poultry, with an extension to 2 min for turkeys, geese and ducks.

Both manual and mechanical CD appear to be **very efficient** without notable differences between both techniques. Concerning the onset of unconsciousness and death, most experts reported in the survey that both types of CD induce immediate loss of consciousness and death (except when manual CD was applied on turkeys, which generally took longer to die). These answers contrast with Jacobs et al. (2019) who found that brain death occurs later when mechanical CD (using Koechner Euthanizing Device KED) is used instead of manual CD. This discrepancy could arise from the fact that mechanical tools other than KED are used in practice (e.g., Livetec Nex). Concerning failure rates, experts reported that less than 0.1% of the birds would remain conscious, regain consciousness, or be unconscious but alive at the end of the procedure, whichever the species at stake and the type of CD used. These failure rates are lower than what Martin *et al*. (2018) found. In their study, trained and experienced operators had a success rate of 96.9% when using manual CD on eight hundred Gallus *gallus*, and a success rate of 98.3% when using mechanical CD with a cone and a nylon cord on four hundred turkeys. Differences between field and experimental results may stem from the difficulty to properly assess unconsciousness in high-throughput depopulation operations.

When CD fails, the **most used back-up method is an additional cervical dislocation**, of which welfare consequences are uncertain. Although a second application will cause further damage, and hence reduce blood flow and swiftly dispatch a bird in a compromised welfare state, it is very likely that it could also stimulate more nociceptors, causing additional pain if the bird remains conscious, even briefly (personal communication from Martin, 2024). Efficient backup methods after CD failures should be investigated. Due to the complexity of the technique required to successfully complete CD, operators must be properly trained to perform this depopulation method. This training should be conducted whichever the type of CD used (manual, or using any kind of mechanical device), as there is variability in the application of any type of CD method by different operators (Martin et al., 2018). To this end, dummies may be used in order to avoid training inexperienced personnel with live birds. In any case, CD should be used as a last-resource depopulation method since loss of consciousness may not occur immediately upon application in birds (AVMA, 2020).

Captive bolt

Depopulation context

From the 5 MS who reported the use of a Captive Bolt (**CB**) since 2018, the depopulation experts from 3 MS answered the method-specific survey with a total of **4 answers** (two experts replied from one MS). The **three most common reasons** given for using CB are 1) the number of birds in the flock, 2) the species at stake, and 3) the animal welfare consequences.

Across the EU, CB seems to be used **only in single-tier systems** (3 experts reported using CB for single-tier systems only), or on "small herds [as a] replacement procedure only".

No expert reported using CB **on flocks larger than 5 000 birds**. All experts reported that CB was used on very small flocks (<250 birds). Among these experts, three also reported it was used on flocks between 251 and 500 birds, and two between 501 and 5000 birds.

All four experts reported to have used CB **on turkeys**, and one expert also reported using CB **on ducks**. No expert mentioned using CB on laying hens nor broilers.

Concerning a possible weight threshold regarding the use of CB, one expert mentioned that only birds weighing 7 kg and more were culled using CB (n_{ans}=1/2 according to 1 expert, n_{unk}=2/4 according to 2 experts, n_{exp}=4).

Operating procedures

In total, **two types of captive bolts guns** were reported to be used in the survey: non-penetrative captive bolts (**NPCB,** reported by three experts) and penetrative captive bolts (**PCB**, reported by one expert) guns. Whichever the types of bolt guns used, all experts reported always having a backup device in case the gun overheats (n_{ans}=3/3 according to 3 experts, n_{unk}=1/4 according to 1 expert, n_{exp}=4), and half of the experts reported having a specific operator dedicated to reloading the captive bolt between each bird (n_{ans}=1/2 according to 1 expert, n_{unk}=2/4 according to 2 experts, n_{exp}=4).

Both types of guns are only used for **stunning purposes** (NPCB: n_{ans}=2/3 according to 2 experts, n_{unk}=1/5 according to 1 expert, n_{exp}=4, PCB: n_{ans}=1/1), although one expert reported that a killing method was not systematically applied on birds after using NPCB. The killing methods reported to be used after NPCB stunning were either cervical dislocation or neck-cutting.

In terms of gun characteristics, **NPCB are exclusively reported to be cartridge-powered** (n_{ans}=3/3 according to 3 experts, n_{unk}=0/3, n_{exp}=3), whereas PCB are reported to be either cartridge-powered (n_{ans}=1/2 according to 1 expert, n_{exp}=1), or spring-loaded (n_{ans}=1/2 according to 1 expert, n_{exp}=1). When using NPCB guns, experts reported employing a bolt with a convex head (n_{ans}=5/5 according to 3 experts, n_{unk}=0/3, n_{exp}=3) and a length below 10 mm (n_{ans}=4/4 according to 2 experts, n_{unk}=1/5 according to 1 expert, n_{exp}=3). Only one of the three experts knew the diameter of the NPCB used, and reported a diameter superior or equal to 6 mm for its use on ducks and turkeys. No answer was obtained regarding the diameter, shape or length of PCB used.

Welfare implications

The main welfare consequences associated with CB reported by the respondents were the handling of the birds and the correct positioning of the bolt, both mentioned by two experts.

Welfare implications associated with the preparation of the killing

Food and water were never reported to be removed before starting the procedure (n_{ans}=0/3, n_{unk}=1/4 according to 1 expert, n_{exp}=4 for both food and water). Most experts reported that **birds were caught and brought to a more confined space** (e.g., specific part of the barn or put in crates/boxes) before the start of the procedure (n_{ans}=2/3 according to 2 experts, n_{unk}=1/4 according to 1 expert, n_{exp}=4).

Welfare implications associated with animal handling

Birds are mostly restrained in a cone, rather than manually, to receive the shot (n_{ans}=2/3 according to 2 experts, n_{unk}=1/4 according to 1 expert, n_{exp}=4). The expert who reported that the birds were held manually did not know in which position the birds were held nor if a specific operator was dedicated solely to the bird restrain, but they specified that the birds (turkeys) were restrained for 30 to 60 seconds. When restrained in a cone, the birds are reported to be restrained for less than 30 s before being shot, whichever the species at stake (n_{ans}=3/3 according to 2 experts, n_{unk}=1/4 according to 1 expert, n_{exp}=3).

Efficiency

Regarding the **percentage of conscious birds observed immediately after having been shot** (i.e. % of failed stunning), answers varied according to the species at stake when NPCB is used: for turkeys, one expert reported a percentage of ineffective stunning inferior to 0.1% (n_{ans}=1/2, n_{unk}=1/3 according to 1 expert, n_{exp}=3, for both values), while another expert reported a percentage comprised between 0.1 et 1%. For ducks, the only expert concerned reported that less than 0.1% of ducks were observed conscious right after the shot. No answer was obtained for PCB.

Regarding the **percentage of birds that regained consciousness** after the shot, all experts indicated a percentage inferior to 0.1% when NPCB is used – whichever the species at stake (n_{ans}=3/3 according to 2 experts, n_{unk}=1/4 according to 1 expert, n_{exp}=3). No answer was obtained for PCB.

Regarding **the percentage of birds being unconscious but alive** after having been shot, all experts reported a percentage inferior to 0.1% when NCPB is used – whichever the species at stake (n_{ans}=2/2 according to 1 expert, n_{unk}=3/5 according to 2 experts, n_{exp}=3). No answer was obtained for PCB.

When a killing method is applied on birds following the shot, all experts reported **a time interval** below one minute **between the stunning and the killing** of the birds – whichever the type of captive bolt used, the species at stake, and the method of killing used (n_{ans}=6/6 according to 3 experts, n_{unk}=0/6, n_{exp}=3).

Regarding **the duration of death induction**, the expert who reported using NPCB as a killing method specified that birds died within 30 seconds after having been shot– whichever the species at stake (n_{ans}=2/2 according to 1 expert, n_{unk}=0/2, n_{exp}=1). No answer was obtained for PCB.

In terms of **throughputs**, when NPCB is used as a stunning method and followed by neck cutting, the expert reported that 30 birds were killed per operator per hour whichever the species at stake (n_{ans}=2/2 according to 1 expert, n_{unk}=0/2, n_{exp}=1). The expert who specified using NPCB as a killing method reported throughputs of 50 birds per operator per hour when CD is used as a backup method and 60 birds per operator per hour when lethal injection is used as a backup method. No throughput was indicated for PCB.

When birds are still alive at the end of the procedure, two **back-up methods** are reported to be used to ensure their death. These methods are (from the most reported to the least reported): cervical dislocation and lethal injection (each reported by 2 experts).

Discussion and conclusion

CB is apparently used to cull flocks of **less than 5 000 birds** – mostly turkeys, and more rarely ducks. However, this observation may not be representative of the use of CB across the EU, since as very few MS replied to this survey.

Two types of CB are used in practice. Non-Penetrative Captive Bolt (NPCB) is reported to be more frequently used than Penetrative Captive Bolt (PCB). Both types of captive bolts are mainly used as a stunning method, in accordance with Council Regulation 1099, although one expert reported not systematically using a killing method after NPCB. Overall, very little information could be obtained concerning PCB throughout the survey.

NPCB appears to be applied using the **same parameters in most depopulation sites**. The design is a cartridge-powered with convex heads, whichever the species at stake. This choice of head shape is in line with the recommendations of the HSA (n.d.). In contrast, the European Commission recommends using flat heads for small birds such as chickens (Preparation of best practices on the protection of animals at the time of killing).

Concerning the **efficiency** of the tools, reported values for NPCB were **satisfactory**. The values reported were less than 0.1 or 1% of birds remaining conscious, and less than 0.1% regaining consciousness or being unconscious but alive. The latter parameter suggests that NPCB can be a killing method, as already highlighted by literature (EFSA 2019), and even though Council Regulation 1099 refers to CB as a simple stunning method. That being said, in accordance with legislation, all but one expert reported that a killing method (cervical dislocation or neck-cutting) was systematically applied within 1 min after the bird got shot.

Experts pointed out **animal handling as one major welfare consequence** associated with CB. Inappropriate handling can result in inadequate placement of the CB and hinder its efficiency.

Birds are either restrained manually for 30-60 s, or restrained in a cone for less than 30 s – which is a duration deemed acceptable in terms of animal welfare (HSA, 2013).

Percussive blow to the head

Depopulation context

From the 3 MS who reported the use of Percussive Blow to the Head (**PBH**) since 2018, only the depopulation expert from one MS answered the method-specific survey, with a total of **1 answer**. The **three main reasons given** for using PBH specifically are 1) the animal welfare consequences, 2) the species at stake, and 3) the practicality of the method.

Regarding the species at stake, the housing systems and the size of the flocks on which PBH can be applied, the expert provided no informative answer. Nonetheless, they specified that PBH was used to cull the whole flock of the farm, not only a subgroup. The expert did not specify if a weight range is respected to apply PBH.

Operating procedures

The expert mentioned the use of **different tools to perform PBH**: a metal pipe, a bat, and a solid wooden stick. No information was provided as to whether the head of the bird is placed on a hard surface to be hit.

The expert specified that the purpose of the procedure was **to stun birds**, and that decapitation was performed following PBH to ensure the death of the bird.

Welfare implications

No main welfare consequence was raised by the expert concerning PBH.

Welfare implications associated with the preparation of the killing

The expert did not know if food and water are removed before the procedure.

Welfare implications associated with animal handling

The expert did not know if the birds are placed in a confined space before the implementation of the procedure or if they are transported to a specific location. They reported that each bird is restrained manually, by a specific operator dedicated to its sole restrain. However, the expert did not know for how long the birds are restrained.

Efficiency

No information relative to the efficiency of the procedure was retrieved from the survey.

Discussion and conclusion

Results related to the use of Percussive Blow to the Head (PBH) **are very scarce**. This lack of information may partially stem from the fact that PBH is not widely spread as a depopulation method since Council Regulation 1099 specifies that PBH "shall not be used as [a] routine method but only where there are no other methods available for stunning". When PBH is performed, the tools used are metal pipes, bats, and solid wooden sticks.

Uncertainty remains around the operating procedures effectively used to perform PBH. From the answers obtained, it remains unknown whether the bird's head is placed against a hard surface when the blow is applied – a position recommended by the European Commission (2017) and EFSA (2019). Whether PBH is only applied on small birds remains also unknown. In theory, PBH induces effective loss of consciousness in birds weighing up to 16 kg (Cors et al., 2015). However, in practice, Council Regulation 1099 forbids the use of PBH on birds weighing more than 5 kg. Little information regarding the handling procedure of the birds was also obtained. The only respondent of the survey reported that the birds are manually restrained by an operator dedicated to this purpose solely, but for an unknown duration.

The only respondent to the survey reported using PBH as a **stunning method**, and applying a killing method (decapitation) afterwards. This is in accordance with EURCAW-Poultry-SFA (2021). No information concerning the method efficiency was obtained. For this reason, field observations are welcome to assess the method's efficiency in depopulation conditions.

Electrical waterbath

Depopulation context

From the 4 MS who reported the use of Electrical Waterbath (**EW**) since 2018, only **2 depopulation experts from 1 MS** answered the method-specific survey. Regarding the **main reasons given** for using EW specifically, both experts mentioned the number of birds in the flock and the species at stake. One expert also reported the animal welfare consequences, while the other indicated the farm configuration (open vs sealed barn).

EW is reported to be used in **all types of housing systems** by both experts: single-tier, multi-tier and cages. EW is only reported to be used **on flocks smaller than 5 000 birds** (n_{ans}=1/1 according to 1 expert, n_{unk}=1/2 according to 1 expert, n_{exp}=2). However, during the workshop, experts from the same MS that reported using EW on flocks up to 5 000 narrowed this number down to 500 birds.

EW is reported to be used to cull **all poultry species under study**: Gallus *gallus* and ducks (according to both experts); and turkeys (according to one expert). This expert reported that there was no weight range to use EW, but he specified that the method was only applied if "the animals are large enough to be hung on the hooks and reach deep enough into the water" (n_{ans}=1/1 according to 1 expert, n_{unk}=1/2 according to 1 expert, n_{exp}=2). During the workshop, both experts specified that they were not aware of any equipment specifically designed for ducks, but stated that the shackles hang sufficiently low for the ducks to be immerged in the waterbath almost up to their breast (apparently including head and neck). For turkeys, one expert clearly mentioned that pre-stun shocks could not be avoided, hence their choice to use method other than EW whenever possible: "it is our last choice for sure".

Operating procedures

EW is reported to be used as a **killing method** rather than a simple stunning method (n_{ans}=1/1 according to 1 expert, n_{unk}=1/2 according to 1 expert, n_{exp}=2).

Regarding the electrical parameters, **different current values** appear to be used depending on the species at stake: both experts reported values of 0.16 A/bird minimum for Gallus *gallus*, 0.2 A/bird minimum for ducks, and the expert who reported using EW on turkeys specified a minimal current of 0.25 A/bird. Both experts did not know the electrical frequency used for EW nor the frequency at which the equipment was calibrated – although one expert specified that the electrical frequency did not depend on the species at stake (n_{ans}=1/1 according to 1 expert, n_{unk}=1/2 according to 1 expert, n_{exp}=2). During the workshop, an expert from the MS that had reported the use of EW specified that the frequency of the device used is 50 Hz. In the survey, experts did not indicate the duration of immersion of the birds neither.

Welfare implications

One expert reported several welfare consequences associated with the use of EW: the difficulty of culling birds of heterogenous sizes (which cannot all be immersed sufficiently deep in the waterbath), insufficient amount of current flowing through each bird, and the risk of pre-stun shocks – particularly in the case of waterfowl.

Welfare implications associated with the preparation of the killing

One expert reported that neither food nor water is removed before the start of the procedure, whereas the other reported their removal in cases where the feeding/water systems interfere with the catching. In the latter case, resources are removed within the hour preceding the catching of the birds.

Welfare implications associated with animal handling

Both experts reported that the birds are placed in a confined space before the procedure, but not transported to a specific location to be killed (n_{ans}=1/1 according to 1 expert, n_{unk}=1/2 according to 1 expert, n_{exp}=2).

Welfare issues occurring during the depopulation procedure

Both experts reported that birds are shackled by two legs on wet shackles, provided they are not injured, for an unknown duration before entering the waterbath. One expert reported that the width of the shackles could not be adjusted to the size of the legs of the birds (n_{ans}=1/1 according to 1 expert, n_{unk}=1/2 according to 1 expert, n_{exp}=2). During the workshop, one expert who filled the survey nonetheless specified that electrical waterbath specifically designed for turkeys exist in their country. With these devices, the shackles size and the space between the shackles are adapted to turkeys.

No answer was obtained concerning the percentage of pre-shocked birds.

Efficiency

Regarding the **percentages of conscious birds**, or **birds regaining consciousness**, or **birds unconscious but still alive** at the end of the procedure, no answer was obtained.

No answer was obtained concerning the number of birds killed per operator per hour. However, during the workshop, experts from the MS who use EW on turkeys mentioned that the equipment used has a throughput of around 700 turkeys per hour, but specified that other mobile models exist and allow much higher throughputs. They specified that they did not use these other models because of the difficulty to shackle birds at these high speeds and the need for personnel.

When birds are still alive at the end of EW, only cervical dislocation was reported as a **back-up method** to ensure the death of the birds (n_{ans}=1/1 according to 1 expert, n_{unk}=1/2 according to 1 expert, n_{exp}=2).

Discussion and conclusion

Results from the survey (2 experts from one MS) indicate that EW is used on all species under study. EW is used as a killing method and in all types of housing systems, preferably in relatively small flocks.

In practice, **EW appears to be performed in accordance to law** - with electrical parameters defined per regulation to ensure the death of all birds. The reported frequency was 50 Hz, in accordance with Council Regulation 1099. The minimum currents used are 0.16 A/bird for Gallus *gallus,* 0.2 A/bird for ducks and 0.25 A/bird for turkeys. Assuming these currents correspond to the average values per animal, these are in line with the values requested by Council Regulation 1099 – with even higher currents than those required for Gallus *gallus* and ducks. In case of depopulation, HSA recommends to use current amplitudes even higher than those actually used on field "to increase the probability that all birds will die" (HSA, 2016), as it reduces the risk of stunning failure, especially for birds with high resistance. When several birds are immerged at the same time when using mobile devices, a bird with a higher-than-average resistance will receive less current than the other birds, and it may not be adequately stunned if the amount is lower than intended (Berry, Meeks, Tinke & Frost, 2002). HSA (2013) recommends for on farm killing for disease control purposes to use the minimum current of 400 mA and waveform of 50 Hz (AC) to induce effective cardiac arrest (killing) in chicken, guinea fowl, duck and geese. Lower currents have been shown to be effective in inducing cardiac arrest in chickens (Gregory and Wotton, 1990). For on-farm killing, these minimum currents should be delivered using electrical waterbath stunners supplied with 50 Hz sine wave AC, which is more effective in inducing cardiac ventricular fibrillation at stunning leading to death.

Various reservations concerning welfare issues linked to the use of EW have emerged. First, birds are most often hung in shackles of non-adjustable size. This practice may harm the birds as shackles can compress the tissues of the shank, including the innervated periosteum and the tarsometatarsal bone (HSA, n.d.a). Moreover, involuntary inversion causes stress and fear to poultry as viscera compress their heart and lungs (because they lack diaphragms) (EFSA, 2022, HSA, n.d.a). EW also causes frequent pre-shocks on turkeys. This observation is in line with the existing scientific literature (Wooton & Gregory, 1991; Gregory, 1994). In their review, Sparks et al. (2010), explained that "turkeys are especially prone to pre-stun shocks […] because their wings hang lower than their heads when hung inverted on a shackle". Additionally, as turkeys may be much heavier than Gallus *gallus* or other categories of poultry (e.g., weighing more than 15 kg), the load on their leg is therefore greater. As a result, HSA recommends not to shackle turkeys of more than 15 kg (HSA, n.d.a).

Uncertainty persists surrounding the efficacy of the method. No answer was obtained with regard to the percentages of conscious birds, birds regaining consciousness, unconscious but alive birds at the end of the procedure, or associated throughputs. Likewise, the immersion duration of the birds was not reported in the survey – despite its influence on unconsciousness and death induction (HSA, 2016). Therefore, more information should be gathered on the aforementioned variables through field data analysis before providing an opinion on the use of EW in depopulation context with specific procedures (such as high current intensity).

Head-to-body electrical stunning

Depopulation context

From the 2 MS who reported the use of Head-to-Body Electrical Stunning (**HBES**) since 2018, the depopulation experts from 2 MS answered the method-specific survey with a total of **2 answers**. The **main reasons given** for using HBES specifically are, for the first expert 1) the species at stake, 2) the animal welfare consequences, and 3) the number of animals in the flock; and for the second expert 1) the availability of the equipment, and 2) the possibility of using HBES on heavy and aggressive birds that are difficult to handle.

Both experts reported HBES to be used in **single-tier housing systems** only. HBES is only reported to be used on turkeys (n_{ans}=2/2 according to 2 experts, n_{unk}=0/2, n_{exp}=2) - the experts did not report its use on Gallus *gallus* nor ducks - and **on flocks of 250-500 birds** (n_{ans}=1/1 according to 1 expert, n_{unk}=1/2 according to 1 expert, n_{exp}=2), and.

The experts specified there was a weight range to the use of HBES: one expert specified that HBES is only used on turkeys weighing more than 7 kg, while the other wrote that HBES is only performed on male turkeys at the end of the fattening period. During the workshop, an expert from one of the two MS that reported using HBES explained that this method has been used on turkeys (especially big turkeys) due to a lack of adequate alternatives to deal with such birds.

Operating procedures

HBES is reported to be used as a **killing method** by both experts. Killing occurs after either **one electrical cycle** (on head and sternum), **or two cycles** with tongs separated by less than one minute (on the head first to stun, and then on the heart/breast of the birds to kill). During the workshop, an expert from the MS that reported the use of the tongs specified that these tongs were initially designed for piglets, applying different electrical parameters on birds ("higher values").

The expert using the one-cycle technique specified that the size of the electrodes was adaptable, contrary to the electrodes (tongs) used for the two-cycle technique.

Concerning electrical parameters, both experts reported that the current and the frequency values used depend on the species at stake. The expert that reported the use of the one-cycle technique did not give a current nor a frequency value. The expert that reported the use of two-shock technique mentioned applying a current of 1.3 mA for both shocks, but did not specify a frequency value. The expert using the one-cycle technique indicated that the equipment is calibrated on several occasions, i.e., in-between depopulation sites and between individuals of the same flock (n_{ans}=1/1 according to 1 expert, n_{unk}=1/2 according to 1 expert, n_{exp}=2).

Welfare implications

Only one expert reported issues related to unconsciousness induction and animal welfare. According to them, the main issues are "Difficult[y] to restraint the animals, apply[ing] correctly the electrodes and hav[ing] a good source of energy".

Welfare implications associated with the preparation of the killing

One expert reported that both food and water are removed immediately before the start of the procedure to "facilitate the handling of animals and movement of people" (n_{ans}=1/1 according to 1 experts, n_{unk}=1/2 according to 1 expert, n_{exp}=2).

Welfare implications associated with animal handling

One expert reported that the birds are placed in a confined space before the procedure, but are not transported to a specific location to be killed (n_{ans}=1/1 according to 1 expert, n_{unk}=1/2 according to 1 expert, n_{exp}=2). Both experts reported that birds are restrained manually to apply the electrical current, for a duration of either "less than 30 seconds" (one-cycle technique) or "between 1 and 5 minutes" (two-cycle technique).

Welfare issues occurring during the depopulation procedure

Regarding accidental pre-shocks, experts gave very different values: less than 0.1% of turkeys are pre-shocked with the one-cycle technique, whereas more than 20% of turkeys are pre-shocked with the two-cycle technique. Several factors were mentioned by the expert using the two-cycle technique to justify the amount of pre-shocks: the difficulty to handle and restrain the animals, the lack of staff training, and the not fit for purpose equipment.

Efficiency

Regarding the **percentage of birds remaining conscious**, less than 0.1% of turkeys are reported to be conscious after the one-cycle technique, while more than 20% of turkeys are reported to be conscious after the two-shock technique.

Regarding the **percentage of birds having regained consciousness** or **being unconscious but still alive at the end of the procedure**, less than 0.1% of turkeys are reported to have regained consciousness or to be unconscious but alive after the one-cycle technique. No value was obtained from the expert who reported the use of the two-shock technique, after neither shocks.

Regarding the **number of turkeys killed per operator per hour**, no value was obtained from either expert.

When birds are still alive at the end of HBES, different **back-up methods** are reported to be used to ensure their death: an additional electrical current (reported by both experts) or a lethal injection (reported by one expert).

Discussion and conclusion

Results from the survey indicate that HBES is used as a **killing method to depopulate only turkeys, especially heavy ones** (weighing 7 kg and more). HBES is used as a kind of last resort depopulation method for turkeys, when no other method is available, and in in single-tier housing systems only. Flocks of 250-500 birds were reported to be culled using this method.

Two devices appear to be used to perform HBES, with contrasting level of efficacy. The first device ('two-cycle' technique) consists in tongs originally designed to cull piglets, the size of which could not be adjusted to their application on poultry. The two-shock technique was used by delivering a first shock on the head to stun the bird, followed by a second shock on the breast/heart to kill the already unconscious bird. Both shocks had a reported current value of "1.3 mA", (which is most likely a typo for "1.3 A") and the frequency was unknown. To apply the shocks, birds were reported to be restrained manually for up to five minutes, which may be prejudicial to their welfare. The use of this device resulted in more than 20% of birds undergoing preshocks and more than 20% of birds remaining conscious after the first shock. The failure rate could potentially be reduced by applying the second shock immediately following the first shock (rather than within one minute). In light with these results, the EURCAW-PSFA concludes that this device is inadequate for turkeys and does not recommend its use to cull turkeys. Alternative device should be used. The second device ('one-cycle' technique) allows for the application of a single current from head to sternum. The size of the electrodes is adaptable to the species, and the birds are restrained for less than 30 seconds. The use of the device resulted in less than 0.1% of birds undergoing pre-shocks and failures (birds remaining conscious, regaining consciousness or being unconscious but alive at the end of the procedure). This device and the protocol used hence appear very efficient at successfully killing birds without triggering substantial aversive reactions. However, very scant specific information could be retrieved about this device and its electrical parameters, both during the workshop and in the survey answers. Therefore, more information should be gathered before expressing an opinion on its use for depopulation of poultry.

Head-only electrical stunning

Depopulation context

From the 2 MS who reported the use of Head Only Electrical Stunning (**HES**) since 2018, the depopulation expert from 1 MS answered the method-specific survey with a total of **1 answer**. The main reasons given for using HES specifically are 1) the species at stake, 2) the animal welfare consequences, and 3) the number of animals in the flock.

The expert reported HES to be used **in single-tier housing systems** only, and solely on turkeys weighing 7 kg and more. No value was obtained for the size of the flocks on which HES is used.

Operating procedures

HES is reported to be used as **a killing method**, with electrical tongs of size that can be adapted to the species at stake.

Concerning the electrical parameters (current and frequency), no value was obtained. The expert mentioned that the equipment is calibrated on several occasions, i.e. "In-between depopulation sites; In-between species; During the procedure (i.e., between certain individuals)".

Welfare implications

No main welfare consequence was raised by the expert concerning HES.

Welfare implications associated with the preparation of the killing

The expert did not know if food and water are removed before the start of the procedure.

Welfare implications associated with animal handling

The expert did not know if the birds are placed in a confined space before the procedure, but they reported that the birds are not transported to a specific location to be killed. He also reported that the birds are restrained manually to receive the electric shock, for a duration of "less than 30 seconds".

Welfare issues occurring during the depopulation procedure

Regarding accidental pre-shocks, the expert reported that less than 0.1% of turkeys are pre-shocked.

Efficiency

Regarding the **percentages of birds remaining conscious**, **having regained consciousness or being unconscious but alive** at the end of the procedure, the expert reported the same values of less than 0.1% of turkeys in each case.

Regarding the **number of turkeys killed per operator per hour**, no value was obtained.

When birds are still alive at the end of HES, the expert did not know what **back-up methods** are used to ensure their death.

Discussion and conclusion

Results from the survey (one answer) seem to indicate that **HES is rarely used across the EU**, but the fact that other MS potentially use this method and did not answer the survey cannot be overruled. The only respondent reported using HES on turkeys only, especially heavy turkeys (weighing 7 kg and more). HES is reported to be used as a killing method, which goes against Council Regulation 1099 – where HES is referred to as a stunning method only. The reason behind the specific use of HES for heavy turkeys has not been brought to light.

Based on the information retrieved, **only one general operating procedure has been identified**. When HES is used, turkeys are manually restrained for a relatively short duration (less than 30 s) before the application of the electrical current via tongs. No additional step appears to be taken to ensure the animal's death (e.g., via the application of another current, or the use of a killing method such as cervical dislocation).

No value was obtained regarding the electrical parameters of the electrical tongs used, but note that the current used should at least be equal to 400 mA when HES is performed on turkeys – as per requirement from Council Regulation 1099. Current intensity higher than the legal requirement can also be used to reduce the risks of failures (HSA, 2016) since no meat quality constraints apply in this depopulation context.

Concerning the welfare indicators, **results reported in the survey are – albeit scarce – highly satisfactory**. Less than 0.1% of turkeys appear to be pre-shocked, to remain conscious, to regain consciousness or to be unconscious but alive at the end of the procedure. More detailed information about the procedure and tools used would be welcome before recommending the use of HES as a depopulation method.

Decapitation

No MS answered the survey on decapitation.

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About EURCAW-Poultry-SFA

EURCAW-Poultry-SFA is one of the four European Union Reference Centres for Animal Welfare. It focuses on poultry and other small farmed animals welfare and legislation, and covers the entire life cycle from hatch/birth to the end of life. EURCAW-Poultry-SFA's main objective is to scientifically and technically support the European Commission and Member States for implementation of welfare legislation. This includes:

- Directive 98/58/EC concerning the protection of animals kept on farms;
- Regulations 1/2005/EC and 1099/2009/EC concerning their protection during transport and slaughter;
- Directive 1999/74/EC laying down minimum standards for the protection of laying hens;
- Directive 2007/43/EC laying down minimum rules for the protection of chickens kept for meat production.

Partners

EURCAW-Poultry-SFA receives funding from DG SANTE of the European Commission and represents a collaboration between the following four partner institutions:

- ANSES, France
- IRTA, Spain
- ANIVET, AU, Denmark
- IZSLER, Italy

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Activities of EURCAW-Poultry-SFA

- Coordinated Assistance
  Providing support, networking and Questions to EURCAW;
- Welfare indicators, Assessment & Good Practices
  Identifying animal welfare indicators, including animal based, management based and resource-based indicators, that can be used to verify compliance with the EU legislation;
- Scientific and technical studies
  Preparing Scientific Reviews of knowledge on welfare topics, identify research needs and perform scientific and technical studies to fill the gaps of knowledge;
- Training
  Reviewing existing training activities and developing new training materials, webinars and knowledge pills for official inspectors and competent authorities;
- Communication and Dissemination
  Increasing awareness of our outputs via the website, and newsletter.

Website and contact

EURCAW-Poultry-SFA's website offers relevant and actual information to support enforcement of poultry and other small farmed animals' welfare legislation.

We offer a 'Questions to EURCAW' service for official inspectors, policy workers, and other personnel providing advice or support for official controls of poultry and other small farmed animals welfare in the EU. For more information go to the Q2E webform available online here or https://survey.anses.fr/SurveyServer/s/DSL/Queryw. All Q2E answers are available online

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