### Risk assessment of acquiring Avian Influenza from Poultry Products: Exposure assessment

The risk pathway considered in this risk assessment is illustrated in Figure 1. Further detail on what was considered for the three specific products (commercial poultry, game birds, and hen eggs) in scope for this work are provided in the following sections. Since the size of the population of backyard poultry in the UK is not known, nor is it understood how often poultry products from this population is consumed (uncertainty), this pathway was not considered separately. It is highlighted in Section 5 the most related pathways where consideration of backyard poultry might also fall under.

Figure 1: Overall risk pathway from the farm or field through to consumption of poultry products

### 4.1 Commercial poultry

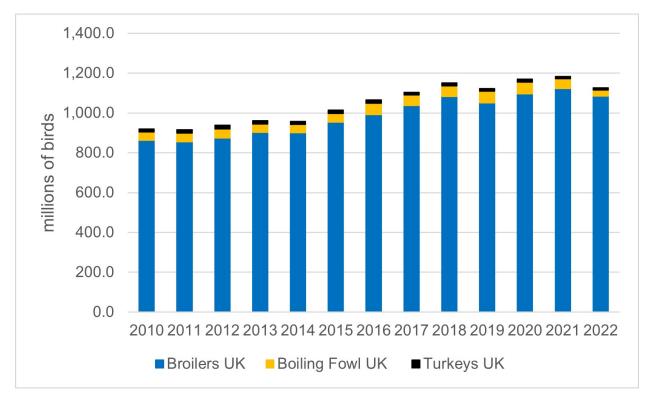
The risk pathway, through to the processing of the poultry meat, considered for AI-infected flocks of commercial poultry is illustrated in Figure 2.

Figure 2: Risk pathway for avian influenza infection from commercial poultry through to the processing stage.

4.1.1 Overview UK poultry meat production

The number of poultry slaughtered each year in the UK (including broilers, turkeys and boiling fowl) has been increasing from 2010 through to 2021, although the effect of the AI outbreaks led to a decrease in 2022 (Defra, 2023b, 2022b). Poultry slaughtering and poultry meat production in the UK is estimated from the number of chicks placed by hatcheries and day old chick imports combined with advice on the life-span and mortality (Defra, 2022b). Total UK poultry meat production went down by 0.8% at 181.7 thousand tonnes between October 2021 and October 2022 (Defra, 2022b). Although the number of poultry availability in the next month is indicated by the number of eggs set each month, consumer demand strongly affects broiler chick and turkey poultry placing in the months running up to Christmas (Defra, 2022b).

Figure 3: Number of Poultry slaughtered per year in the UK. Boiling fowl includes spent hens and spent breeders. Data from (Defra, 2023b)



#### 4.1.1.1 Commercial duck and geese production

Although the commercial poultry industry is dominated by the production of chicken and turkeys, there has been a steady increase in the commercial production of other poultry, such as ducks and geese, in recent years. The duck industry, in particular, has expanded to now produce an estimated 16 million birds a year in the UK. These are mainly ducks of the Pekin variety. Approximately 95% are intensively reared and 5% are free range (Barrow et al., 2021). Although primarily produced for their meat, duck eggs are also consumed, and duck feathers are sought after for the filling of home products such as pillows or duvets.

The commercial production of both ducks and geese is included in the legislation described in section 1.5 for the control of AI (The Avian Influenza and Influenza of Avian Origin in Mammals (England) (No.2) Order 2006, 2006). Therefore, commercial producers of these bird types must follow all measures to reduce the risk of transmission and deal with outbreaks, including the culling of positive flocks. However, as non-indicator species, which may not show signs of infection as clearly as chickens, inspection of the carcass may not be sufficient to rule out disease, particularly for LPAI infection (APHA, 2022a) (uncertainty). There is a lack of evidence regarding the clinical signs of HPAI infection in non-indication species, however, some strains have been reported to cause mortality in domestic ducks (Spackman, 2020) (uncertainty). It is

assumed that commercially reared ducks will act similarly to wild ducks when infected with AI, as described in Section 4.2 (uncertainty).

#### 4.1.2 Probability of AI in commercial poultry sent for slaughter

Initial clinical presentation of HPAI infection in poultry (chicken and turkeys) might be sudden death, with little or no other preceding clinical signs or gross pathology (More et al., 2017). However, there may be cases of acute disease presenting with signs of systemic infection that could include respiratory distress, coughing, sneezing, cyanosis (blue discolouration) of combs and wattle, haemorrhages on the shank, bleeding from the nares, diarrhoea, circling, incoordination, and death (Abd El-Hack et al., 2022).

Other clinical signs described are oedema (swelling) of the head, dullness, a loss of appetite, or depression ("NADIS Animal Health Skills - Avian Influenza"). Nervous system signs would include ataxia, paralysis of the wings and paddling movements of the legs (More et al., 2017).

An investigation by Post at al. (2012) suggested that for LPAI, no clinical signs were observed. Another study found that LPAI infection in chickens and turkeys manifests with mild respiratory distress and reduced egg production; however tracheitis, sinusitis, air sacculitis, nephritis, ovaritis, and oviduct lesions with egg peritonitis in layers can be observed sometimes (Abd El-Hack et al., 2022).

#### 4.1.2.1 Viral distribution and titre in poultry issue

Post et al. (2012) investigated HPAI and LPAI viral load and manifestation in chickens, demonstrating that H7N1 HPAI infected chickens developed illness with depression that led to death. Their qPCR data showed high amounts of viral RNA load in all organs tested (lung, trachea, ileum, and brain). The most significant difference between HPAI and LPAI RNA load was found in lungs, with LPAI detected from the lung for at least a week. It was summarised that, although both HPAI and LPAI could be detected in a broad range of tissues, the pathogenicity and mortality differences between them could originate from differences in virus replication and the resulting host responses in vital organs (Post et al., 2012). However, another study has reported that the amount of LPAI virus in muscles of poultry (including turkeys and quail) experimentally infected ranged from 100.44 to 101.6-102 /g of tissue, as summarised in the EFSA report (EFSA et al., 2018). Another study has detected LPAI H9N2 virus at 103.5/g from trachea (Shibata et al., 2018). These studies provide evidence that LPAI can be isolated from the muscles and organs of experimentally infected poultry; the presence of LPAI in muscle tissue in naturally infected birds has not been demonstrated in published literature.

Experiments performed by Pantin-Jackwood et al. (2017) demonstrated that turkeys infected with dose of 103 EID50 LPAI, have shown clinical sign of respiratory disease, mild lethargy and infraorbital swelling, with no mortality. Additionally, HPAI infection in turkeys have presented differently to chickens, with more neurological signs and no haemorrhagic lesions present (Pantin-Jackwood et al., 2017).

Tumpey at al. (2002) showed that HPAI H5N1 isolated from duck meat was highly pathogenic and able to replicate in chicken following intravenous and intranasal inoculation (Tumpey et al., 2002).

Infection of the liver in chickens is also a concern due to the consumption of chicken liver pate. Several HPAI and LPAI strains have been detected in the liver of experimentally infected chickens (Post et al., 2012; Yamamoto et al., 2017). The initial viral titre in liver was 104.3 to 10 6.5 EID50/ml. Liver samples from birds experimentally infected with HPAI H5N1 have been shown to be positive for virus after 20 days when stored at 4°C and 3 days when stored at 20°C (Yamamoto et al., 2017).

#### 4.1.2.2 Probability of virus present in meat of slaughtered birds

HPAI induces a systemic infection in the host – an infection affecting the whole bird in multiple organs. HPAI viral replication causes high titres of infectious virus in internal organs (for example liver, heart) and also in muscle and other tissues, in addition to respiratory and gastrointestinal tracts. (ACMSF, 2015; Harder et al., 2016).

Golden et al. (2009) have used a state-transition model to investigate the probability that an infected flock with HPAI H5N1 will remain undetected until slaughter, considering three possible states, susceptible (not infected), infected, and dead, and the transition probabilities that would predict the movements between those states (Golden et al., 2009). They predicted a high probability (0.94) that a flock infected with HPAI H5N1 would be detected before being sent to slaughter (Golden et al., 2009). The probability of detection was inversely related to the length of the non-detection window (the time between infection and the flock being identified as infected), that is a function of the effective contact rate, latency, bird mortality rate, and daily mortality threshold (Golden et al., 2009).

HPAI can be detected in the muscle and other tissues of poultry, in addition to the respiratory and gastrointestinal tract (ACMSF, 2015). However, it is likely that most flocks infected with HPAI will be detected and culled before entering the UK food chain, since clinically affected poultry will be detected and excluded at the farm or upon ante mortem inspection at the processing plant. Post-mortem clinical signs of HPAI infection at post mortem inspection include: lesions that may include cyanosis and oedema of the head, comb, wattle, and snood (turkey); ischemic necrosis of comb, wattles, or snood; oedema and red discolouration of the shanks and feet due to subcutaneous ecchymosis haemorrhages; petechial haemorrhages on visceral organs and in muscle; and blood tinged oral and nasal discharges ("Avian Influenza - Poultry," 2022).

Surveillance data revealed that HPAI H5N1 virus was detected in imported frozen duck meat and on the surface and in internal contents of contaminated eggs. Experimentally, HPAI virus was detected in breast and thigh meat, and blood and bones, as well as in eggs of HPAI infected chickens.

LPAI infections in chickens either do not present or very rarely present any clinical signs. Both experimental infection studies (Bergervoet et al., 2019; Roy Chowdhury et al., 2019) and field observations (Bertran et al., 2018) suggest that LPAI viruses are primarily restricted to the respiratory and gastrointestinal tract. As a result of that, it is agreed with the conclusion of the 2018 EFSA report (EFSA et al., 2018) that the virus is very unlikely to be disseminated systemically, although it should be noted that turkeys are more susceptible than chickens and experimentally-infected turkeys may exhibit some systemic infection (EFSA et al., 2018).

Gonzales et al. (2018), on behalf of EFSA, assessed the probability of exposure to LPAI and subsequent infection of the consumer via raw poultry meat and raw table eggs (EFSA et al., 2018). They concluded that the probability of exposure following an infection of LPAI in birds via raw poultry meat containing this virus is negligible for commercial poultry (EFSA et al., 2018). However, it is unlikely that a bird infected with LPAI will be detected via ante-mortem inspection as diagnosis is dependent on the presence of clinical signs (EFSA et al., 2018). Also, the presence of the LPAI virus in muscle tissues is not always associated with the presence of microscopic lesions, and at the high speed of slaughter lines detection of the lesions and faecal contamination of the carcasses by the visual post-mortem inspection can be unidentified (EFSA et al., 2018).

In birds infected with the LPAI virus, it is rarely found in the blood. Therefore, meat would be unlikely to contain high levels of virus and the likelihood of infection to humans in this case is very

low (More et al., 2017). Similarly, other products of poultry origin, such as processed meat, are not considered as significant sources for human infection, as they are designated for further treatment (for example, heat processing) (More et al., 2017). Commercial turkeys are highly susceptible to LPAI viruses, often showing increased mortality and are more likely to show clinical signs of respiratory distress, sinusitis, and poor performance (Ngunjiri et al., 2021).

In summary, given the veterinary inspection process in slaughterhouses and clinical signs in the flocks (for example reduced egg production), it is unlikely that HPAI-infected birds will be sent for slaughter. The chances are higher for LPAI-infected flocks since clinical signs may be less acute and signs of infection are less likely to be detected by the post-mortem visual inspection.

# 4.1.3 Probability that AI present in poultry products survives the retail and consumer behaviour stages

#### 4.1.3.1 Shelf-life of poultry products

According to the guidance on temperature control in food premises, fresh pre-packed poultry should be labelled and kept at -2°C to +4°C ("Guidance on Temperature Control in Food Premises," 2013). Defra's "Poultry meat Quality Guide" advises that fresh poultry is to be kept at temperatures not below -2°C and not higher than 4°C (Defra, 2011). Their definition of 'frozen poultry' is poultry meat that must be frozen as soon as possible within the constraints of normal slaughtering or cutting procedures and kept at temperatures no higher than -12°C, where this temperature must be stable and maintained (Defra, 2011). 'Quick-frozen' poultry meat is defined as a quick-freezing method using authorised cryonic media (air, nitrogen or carbon dioxide) where the zone of maximum crystallisation is crossed as rapidly as possible and the resulting temperature is no higher than -18°C (Defra, 2011).

For storage at home, fresh poultry, such as chicken and turkey, can be stored at chilled temperatures (recommended -1°C to +2°C, maximum of 8°C) for 4-7 days if packed in air or extended to 10 days if modified atmosphere packaging is used (Air Products, 2018). Fresh poultry can be frozen by the consumer for up to 6 months; this length of time is determined by quality concerns of the meat rather than food safety issues (Love Food Hate Waste, 2022).

Although the recommendation is for poultry meat to be kept at the temperatures ranging between -2°C to +4°C, the national requirements (Temperature Control Requirements) of the Food Safety and Hygiene (England) Regulations 2013 in England and the 2006 Food Hygiene Regulations for Wales and Northern Ireland require food that is likely to support the growth of pathogenic microorganisms or the formation of toxins be held at or below 8°C, or at or above 63°C (Reg (EC) 852/2004, 2004; "The Food Safety and Hygiene (England) Regulations 2013"). Since the regulation require the food to be kept at or below 8°C , it is important to keep the fridge temperature at or lower than 5-6°C to ensure the temperature of the food does not exceed the recommended temperature of 8°C (the air temperatures of the fridge are often few degree lower than a temperature of the food itself) ("Guidance on Temperature Control in Food Premises," 2013).

#### 4.1.3.2 Survival within poultry products

The normal pH range for chicken breast, thighs and drumstick meat is between 5.54 and 6.1, whilst the moisture content is between 70.3% and 72.8% (Beauclercq et al., 2022; Fernandes et al., 2016). Taking into consideration storage conditions, the pH and water content of the poultry (chicken), and the viral persistence in those conditions (described above), it is very likely that AI will persist for at least 100 days in meat.

Although there is no direct evidence that AI has been transmitted to humans via the consumption of contaminated poultry products, there is anecdotal and experimental evidence that the consumption of uncooked poultry blood or meat has transmitted the HPAI H5N1 virus to carnivorous animals, including tigers, leopards, domestic cats, domestic dogs, and a stone marten (Chmielewski and Swayne, 2011).

If food is not thoroughly cooked or properly handled, there could be some contamination of chicken meat with AI virus, and cross contamination during slaughter cannot be excluded. The conclusion of a study carried out by Dai et al., (2022) highlighted the importance of strict inspection and effective disinfection measures in the supply chain of raw poultry worldwide.

#### 4.1.3.3 AI Survival at freezer and refrigeration temperatures

It is well documented from various studies and sources that HPAI tolerates cold and freezing temperatures well. H5N1 virus was shown to be able to survive for 240 days in feather tissue, 160 days in muscle and 20 days in liver at 4°C (Yamamoto et al., 2017). A study carried out in 2012 proved that a high pathogenic strain H7N1 isolated from chicken, turkey and duck meat was still viable after being kept at 4°C for 135, 90, and 75 days, respectively (Beato et al., 2012). Dai et al. (2022) demonstrated that HPAI was able to survive at various temperatures (-20°C, 4°C, and 25°C) for several days (Dai et al., 2022). This study noted that the lower the temperature, the longer the viability of the HPAI. A study carried out in Pakistan showed that the highly pathogenic H5N1 strain remained viable for more than 100 days at 4°C (Shahid et al., 2009). The infectious virus has also been proven to survive in tissues at 20-22°C for up to 6 days post mortem (Busquets et al., 2010).

LPAI is unlikely to be detected in meat, as routine testing is not currently carried out in nonsymptomatic flocks. Some research has demonstrated though that if the LPAI virus is present in the poultry meat, it can survive in this environment with little effect on viability, if stored in chilled or frozen conditions. It has been shown that LPAI virus is able to survive freezing and refrigeration, and those low temperatures do not reduce the concentration or viability of viruses (Gonzales et al., 2018). In the experiment by Ejaz et al., 2007, in broiler-chickens infected with LPAI, H9N2, and frozen at -20°C, virus was detected in: bone marrow and legs after 6 weeks post-storage, neck and wings until 4 weeks post-storage, and breast until 2 weeks post-storage (Ejaz et al., 2007). Nevertheless, lungs and kidney tissues in frozen carcasses infected with LPAI strains, might still harbour infectious virus (More et al., 2017).

It has therefore been widely shown that if AI is present in poultry meat, it can survive chilling and freezing conditions with little effect on the viability of the virus. It has also been shown that in general, low temperatures prolong the survival of the virus in poultry tissue. Therefore, the advice still remains to fully cook poultry (ACMSF, 2015).

#### 4.1.3.4 Effect of cooking on avian influenza virus

A study to determine the survival of AI subtype H5N1 under various physical and chemical treatments by Wanaratana et al. (2010) showed that all three H5N1 reference viruses used in the study totally lost their infectivity when exposed to a temperature of 70°C for 60 min, or 75°C for at least 45 min. A study by Swayne in 2006 found that the reduction in virus infectivity titres was dependent on virus concentration and no HPAI virus was isolated after 1 s of treatment at 70°C. A change in coloration in the meat from pink-tan to white was associated with a loss in recovery of infectious virus, which is important to note for less than thoroughly cooked (LLTC) poultry products.

Samanta and Bandyopadhyay found that after 30mins exposure at 56°C AI was no longer viable. (Samanta and Bandyopadhyay, 2017). However a study by Zou et al, has shown that H7N9 virus can remain infectious at temperature up to 65°C exposed for 5min, pH levels ?3, and UV light for

20min (Zou et al., 2013). Another study has shown that a HPAI H5N1 strain lost its infectivity after 30min at 56°C and after 1 day at 28°C (Shahid et al., 2009).

Al viruses are considered to be heat labile viruses (Swayne et. al., 2004) so cooking poultry and poultry products thoroughly will destroy them. This indicates that properly cooked poultry meat and eggs do not pose a risk of infection. The evidence from the above studies does indicate that the virus will not persist at higher temperatures. There is a degree of uncertainty here as studies have not been conducted on all the known Al viruses although no information has been found which would suggest that any particular subtype is likely to behave differently in terms of heat stability (uncertainty).

#### 4.1.3.5 Consumption of commercial poultry products

Data from the national diet and nutrition survey (NDNS) on the consumption of chicken and turkey is shown in Table 3 and Table 4 (DHSC, 2013; PHE and FSA, 2020, 2018, 2016, 2014). This data only shows meat consumption without recipes included – that is raw products to be cooked and consumed in the home rather than processed foods such as ready meals, take-aways etc. In the NDNS data, chronic consumption is the amount consumed averaged over four days whereas acute consumption is the amount eaten on one day.

Chicken consumption is by far the most popular poultry eaten with 58.67% of the studied population of 4-64 year olds reporting eating chicken, compared with only 9.08% eating turkey. In infants and children under four, 61.09% are consuming chicken and only 5.70% turkey. A slightly different picture can be seen in the over 65s, although once again chicken is the most popularly consumed poultry meat with 46.49% of respondents consuming it and 8.19% for turkey.

| Table 3: Chicken consum | ption (without | recipes) fror | n NDNS data |
|-------------------------|----------------|---------------|-------------|
|                         |                |               |             |

| Age range                        | % reporting     | Chronic Mean ^ | Chronic Max <sup>^</sup> | Acute Mean^ | Acute Max <sup>^</sup> |
|----------------------------------|-----------------|----------------|--------------------------|-------------|------------------------|
| Infants/children <4<br>(n=3,480) | 61.09% (n=1353) | 9.88           | 78.02                    | 20.99       | 163.73                 |
| 4 to 64 years (n=10,228)         | 58.67% (n=4329) | 32.43          | 233.03                   | 94.26       | 233.03                 |
| 65+ (n=1,538)                    | 46.49% (n=543)  | 32.00          | 240.00                   | 98.00       | 400.00                 |

#### ^: grams/person/day

#### Table 4:Turkey Consumption (without recipes)from NDNS data

| Age range                        | % reporting   | Chronic Mean ^ | Chronic Max <sup>^</sup> | Acute Mean^ | Acute Max <sup>^</sup> |
|----------------------------------|---------------|----------------|--------------------------|-------------|------------------------|
| Infants/children <4<br>(n=3,480) | 5.70% (n=219) | 8.63           | 36.13                    | 30.69       | 110.82                 |
| 4 to 64 years (n=10,228)         | 9.08% (n=934) | 23.65          | 113.60                   | 79.05       | 367.13                 |
| 65+ (n=1,538)                    | 8.19% (n=126) | 80.00          | 150.00                   | 86.00       | 290.00                 |

#### ^: grams/person/day

Figures for the consumption of offal were also requested, however, due to the very low numbers reporting the consumption of offal, the following caveat was implemented, 'Consumption or exposure estimates made with a small number of consumers may not be accurate. The number of consumers is less than 60, this should be treated with caution and may not be representative for a large number of consumers.' The consumption of chicken and turkey offal for adults without recipes (not including processed foods etc) is shown in Table 5. There was no data found for children under 19. Only 0.04% of adults (2 out of 5094 respondents) reported consuming chicken or poultry offal and nobody over 65 years old. There was a slight increase in offal consumption

with recipes included, such as pre-prepared pate and cooked giblets but this was still very low at 0.84% in adults and 0.52% in the over 65yr olds.

| Age range                 | % reporting | Chronic Mean ^ | Chronic Max^ | Acute Mean^ | Acute Max <sup>^</sup> |
|---------------------------|-------------|----------------|--------------|-------------|------------------------|
| Adults 19 to 64 (n=5,094) | 0.04% (n=2) | 42.00          | 63.00        | 140.00      | 3.10                   |
| 65+ (n=1,538)             | 0.00%       | 0.00           | 0.00         | 0.00        | 0.00                   |

#### Table 5: Consumption of Offal -Chicken and Turkey (without recipes) from NDNS data

#### ^: grams/person/day

#### 4.1.3.5.1 LTTC Commercial poultry meat

In the FSA's flagship official statistic survey Food and You 2, 8,672 respondents were asked how often (always, most of the time, about half the time, occasionally, never) they eat either chicken or turkey when it is pink or has pink or red juices (Table 6). In wave 1 (July – October 2020), the data indicated that while a majority (93%) of respondents who eat chicken or turkey report never eating it pink or when it has pink or red juices, 4% report doing this at least occasionally.

### Table 6: How often respondents to the FSA's Food and You 2 survey report eating chicken or turkey meat pink.

| Response            | % of respondents (n=8,672) |
|---------------------|----------------------------|
| Always              | 1%                         |
| Most of the time    | 1%                         |
| About half the time | 1%                         |
| Occasionally        | 4%                         |
| Never               | 93%                        |

### 4.2 Game Birds

The risk pathway considered for AI infection in game birds, through to the cleaning and processing of the carcass, is illustrated in Figure 4.

### Figure 4: Risk pathway for avian influenza infection from game birds through to the processing stage

#### 4.2.1 Overview of the UK game bird industry

Game birds by definition are wild birds that are hunted for human consumption. In the UK common game birds include pheasant, partridge, and grouse as well as waterbirds such as wild geese and wild ducks.

The game bird industry has an estimated worth of over £2 billion a year and continues to grow in popularity. Shoots can no longer rely on wild birds alone, but more often on reared birds. An estimated 83% of shoots use game birds that have been hand reared before being released into the wild.

This is an old practice, with the use of hand reared pheasants and partridges going back over 100 years ("GFA | Game Farming in The UK"). There are approximately 300 game farms in the UK which release an estimated 50 million hand reared pheasants and partridges into the wild for hunting each year (BASC, personal communication). Eggs for hand reared stock are collected in April and hatched in incubators. Although initially reared indoors, chicks are given access to outdoor runs prior to release into the wild to adapt to natural surroundings. In August, at around 8-10 weeks, they are sold to shoots where gamekeepers release them into the countryside ("GFA | Game Farming in The UK"). The shooting season varies depending on the bird but is mainly a winter activity in rural countryside. Not all birds released are shot, some remain in the wild and help to naturally replenish the stock for the following years shooting season.

Wildfowling is similar to game bird shooting; however, it refers specifically to the shooting of geese and ducks and often occurs in marshes or estuaries. Unlike game bird shooting, it does not use reared stocks to supplement the season. Hunted wild geese and ducks mostly migrate from Scandinavia and the Arctic circle in autumn before returning to breeding grounds in Spring (BASC, 2022).

The shooting season for all game birds is tightly regulated and varies depending on species and individual country legislation. Table 7 below summarises the shooting season for each species. The shooting season for most species is in winter which coincides with a higher prevalence of AI in both commercial and wild birds.

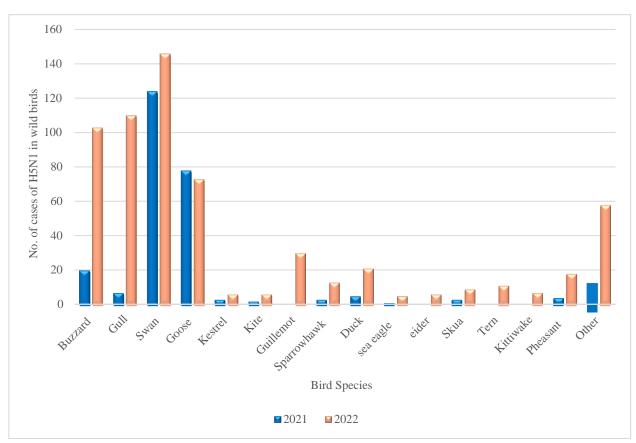
| Species                                 | England and Wales  | Scotland                  | Northern Ireland          | Isle of Man  |
|---|--|---------------------------|---------------------------|--|
| Pheasant                                | October 1 to February 1  | October 1 to February 1   | October 1 to January 31   | October to January 31  |
| Grey Partridge                          | September 1 to February 1  | September 1 to February 1 | September 1 to January 31 | Protected (ban in force)   |
| Red-legged Partridge                    | September 1 to February 1  | September 1 to February 1 | September 1 to January 31 | September 13 to January 31   |
| Red Grouse                              | August 12 to December 10   | August 12 to December 10  | August 12 to November 30  | August 25 to October 31*   |
| Black Grouse                            | August 20 to December 10<br>(Somerset, Devon and New<br>Forest: September 1 to<br>December 10) | August 20 to December 10  | -                         | -  |
| Ptarmigan                               | -  | August 12 to January 31   | -                         | -  |
| Duck and Goose inland                   | September 1 to January 31  | September 1 to January 31 | September 1 to January 31 | September 1 to January 31 -<br>Ducks<br>July 1 to March 31 - Geese |
| Duck and Goose below HWM<br>(see below) | August 12 to January 31  | August 12 to January 31   | September 1 to January 31 | September 1 to January 31 -<br>Ducks<br>July 1 to March 31 - Geese |
| Common Snipe                            | August 12 to January 31  | August 12 to January 31   | September 1 to January 31 | September 1 to January 31  |
| Jack Snipe                              | Protected  | Protected                 | September 1 to January 31 | Protected  |
| Woodcock                                | October 1 to January 31  | September 1 to January 31 | October 1 to January 31   | October1 to January 31   |
| Golden Plover                           | September 1 to January 31  | September 1 to January 31 | September 1 to January 31 | Protected  |

# Table 7: Summary of shooting season in the UK by species and region (BASC, QuarrySpecies and Shooting Seasons)

| Species      | England and Wales         | Scotland                  | Northern Ireland | Isle of Man |
|--------------|---------------------------|---------------------------|------------------|-------------|
| Coot/Moorhen | September 1 to January 31 | September 1 to January 31 | Protected        | Protected   |

### 4.2.2 AI in game birds on the farm or field

There is currently no active surveillance or testing of wild game birds for either HPAI or LPAI in the UK (uncertainty). However, some inference can be made from the increasing number of cases of infection detected in reported dead wild birds. Surveillance of wild birds is primarily carried out by patrols by wardens on wild bird reserves, who collect dead birds for testing, and by reports of dead wild birds by members of the public. The numbers of positive findings have increased significantly from 278 in 2021 to 945 in 2022 (APHA, 2022b). The total numbers of positive birds from these cases have increased from 459 in 2021 to 1513 in 2022 (APHA, 2022b). The breakdown of species affected is shown in Figure 5.



# Figure 5: The number of avian influenza positive findings in wild birds in 2021 and 2022 broken down by species. Figure generate from data in (APHA, 2022b)

#### 4.2.2.1 Likelihood of detection of clinical signs of HPAI and LPAI in game birds

Clinical signs of HPAI in gallinaceous species of game birds (for example pheasants and partridges) include lethargy, ruffled feathers, bright green faeces and sudden mortality. Several species of birds also show neurological signs such as ataxia (Brookes et al., 2022). LPAI infection in gallinaceous species varies from subclinical to clinical signs of conjunctivitis, and/or mild respiratory and neurological clinical signs. Severe neurological signs are unique to HPAI infection and with LPAI under experimental conditions rarely causing mortality. This makes LPAI infected birds harder to detect (Bertran et al., 2014).

Wild aquatic birds such as ducks and geese are a natural reservoir for AI. Some subtypes, especially LPAI, are highly adapted to these species of birds and therefore infection tends to be

subclinical (APHA, 2022a). There are some knowledge gaps with regard to HPAI in these birds. Some subtypes also do not show clinical signs of infection, however, some strains of HPAI have been reported to cause mortality in domestic ducks (Spackman, 2020). Clinical signs have also been observed in wild Anatidae infected with the current circulating strains of AI (APHA, unpublished) (uncertainty).

The severity of clinical signs in gallinaceous species caused by HPAI make infected birds less likely to be shot, either due to the rapid mortality or the unlikelihood that they will fly during the shoot. However, LPAI in these birds and both HPAI and LPAI in aquatic birds is less likely to cause obvious clinical signs (uncertainty).

#### 4.2.2.2 Restrictions on hunting/capture in AI Prevention Zones

Shooting of game birds is not affected by the controls of AI outbreaks within commercial poultry premises. There have however been exceptions during large outbreaks of H5N1 in wild birds, where shooting has been stopped within controlled zones to reduce the movement of infected birds (GOV.UK, 2022b). The housing order for an Avian Influenza Prevention Zone, introduced on November 7 2022 for poultry and captive birds in the whole of England, was amended on January 9, 2023 to restrict the movement of game birds until 21 days after they've been "caught up", referring to the practice of gathering together wild game birds to be held in captivity for the purpose of restocking supplies of game or any breeding programme for the production of such birds (GOV.UK, 2022c).

For HPAI there is usually a Protection Zone (PZ) out to 3km from the IP and a further Surveillance Zone (SZ) out to 10km. Hunting is permitted within these zones, however, movements of birds and bird products are prevented except under licence and gamebird releasing is specifically banned (APHA, 2022a). This has resulted in an estimated 40% reduction of reared game birds being released into the wild for the 2022 shooting season (BASC, unpublished).

#### 4.2.3 AI in game birds during processing stage

#### 4.2.3.1 Likelihood of AI detection before meat released

The likelihood of detection of AI in captive game birds depends on the size of the game establishment and the age of the birds. All holdings keeping 50 or more birds, including game birds that are only housed for part of the year, must register with the government in England, Wales, and Scotland. In Northern Ireland, flocks of all sizes must be registered. Gamebirds, in pens awaiting release into the wild will therefore be required to adhere to regulations in control areas and can be monitored for signs of disease (GFA, 2021). Once old enough to be released, there is no surveillance of game bird populations in the wild so detection at this point is possible only if disease has caused mortality or clinical signs severe enough to see (for example birds too sick to fly).

Those who shoot game birds for human consumption, such as hunters, are recognised as primary producers according to EU food hygiene regulations. Primary wild game products are infeather game that has not undergone food processing. There are three main routes through which these products enter the food chain for human consumption, summarised in Table 8.

### Table 8: Regulations for the sale of game birds in the UK for main routes into food chain for human consumption

|                  | Route 1              | Route 2   | Route 3   |
|------------------|----------------------|---|---|
| Use of game bird | Private domestic use | Direct supply of small quantities to the final consumer or local retailer | Supply to retailers through Approved Game Handling Establishment (AGHE) |

|                     | Route 1  | Route 2   | Route 3  |
|---------------------|--|---|--|
| Source of game brid | Primary Producer (for example, hunter)                       | Primary producer (for example, hunter)                    | Primary Producer (for example, hunter)   |
| Regulations         | Does not require adherence to EU food<br>hygiene regulations | Does not require adherence to EU food hygiene regulations | Approval by FSA or equivalent.<br>Adherence to EU regulations. Carcass<br>subject to specialist and veterinary<br>examination. Records for traceability. |

Firstly, they can be kept for domestic use, where they are only used for domestic consumption by the primary producer or given to friends and family of the primary producer. Domestic use does not require adherence to EU food hygiene regulations as it is not being consumed as part of a food business operation (FSA, 2015). Secondly, the primary producer can directly supply small quantities of in-feather game to final consumers or local retailers, which also does not require adherence to EU food hygiene regulations (FSA, 2015).

Finally, the producer can supply in-feather game indirectly to consumers or local retailers, but this must be through an Approved Game Handling Establishment (AGHE) (FSA, 2015). To sell to an AGHE, primary producers must register with their local authority, comply with the food business operator's responsibilities under both the general hygiene requirements for primary production, Regulation 852/2004 (Reg (EC) 852/2004, 2004), and the specific provisions in Regulation 853/2004 for the initial handling of wild game intended for subsequent supply to an AGHE (Reg (EC) 853/2004, 2004). This includes the requirement that a trained person is present to inspect the game when it is shot, and that temperature controls and hygienic transport is used for storage and transport.

To act as an AGHE, establishments must be approved by FSA, Food Standards Scotland (FSS), or by the Department of Agriculture and Rural Development (DARD) depending on geographical location. They must adhere to various other procedures and retained EU regulations such as 852/2004 and 853/2004 and waste disposal Regulation (EC) No.1069/2009. EU Regulation 852/2004 includes the requirement of storage of in-feather game in a regulated game larder which is required to have appropriate ventilation and temperature control (Reg (EC) 852/2004, 2004). It must be regularly cleaned and protected against animal and pest contamination. EU regulation 852/2004, stipulates that wild game must be cooled to no more 4?C within a reasonable time after killing on a continual cooling curve and maintained at these temperatures or below. Chillers must be cleaned regularly and not overfilled. Furthermore, they are subject to official veterinary controls and inspections and can only process game that has been initially examined and approved by a trained specialist after a shoot. Finally, they are required to keep detailed records for traceability to identity where the game has come from (geographical location) and verify the specialist has been properly trained to inspect the products (FSA, 2015).

Due to the regulations to be an official AGHE or sell to an AGHE, this route of game birds into the food chain for human consumption has the lowest possibility for AI transmission. Examination of carcasses by trained specialists should reduce the likelihood of HPAI infected birds being sold, due to the clear pathology caused by this type of infection - such as inflammation of the sinuses, trachea and air sacs. Oedema of the head and discolouration of the skin may also be seen, and birds will likely show signs of dehydration and their muscles may be congested. Haemorrhages on the lining of the proventriculus, the glandular stomach, can also occur (NADIS, 2016). Similarly, the refrigeration and hygiene controls should reduce cross contamination and the requirement for traceability means carcasses can be traced back to infected areas.

Game birds, however, used for private domestic use or sold directly in small quantities to local retailers will not have undergone these checks and there is no regulation on the processing stages, such as defeathering in the home, so potentially have higher likelihood of transmission (uncertainty).

The exact number of game birds shot each year and consumed through unregulated routes is unknown (uncertainty). However, estimates provided by The British Association for Shooting and

Conservation (BASC), suggest 37.5% of the 50 million game birds released each year are shot, 97% of which are consumed. 35% of those consumed will go to restaurants and shops through an AGHE and 62% are consumed domestically or are sold through non-AGHE's (therefore non-regulated) (BASC, personal communication). Due to the restrictions on release of game birds in PZ's BASC also estimate that 40% less fewer birds were released into the wild for shoots throughout 2022. Based on these estimates the numbers of game birds consumed in the UK through various routes are calculated in Table 9, for a typical year and 2022. In 2022, an estimated 3.94 million game birds were sold though as AGHE while 6.77 million game birds were sold through non-AGHEs or used for home consumption.

# Table 9: Estimate of number of game birds consumed in the UK on a typical year versus2022 through various routes of sale

| Year                          | Released for shooting (million) | Number shot (million) | Sold to<br>Restaurants/shop<br>through AGHE (million) | Non-AGHE sale and<br>home consumption<br>(million) | Not consumed (million) |
|-------------------------------|---------------------------------|-----------------------|---|--|------------------------|
| Typical year                  | 50                              | 18.75                 | 6.56  | 11.28  | 0.56                   |
| 2022 (due to Al restrictions) | 30                              | 11.25                 | 3.94  | 6.77   | 0.34                   |

To conclude, the likelihood of AI infected game birds being detected before reaching the home for processing and consumption is greater if acquired thought the AGHE route. The regulations, including carcase inspection, storage, transport, and processing of carcases as well as product traceability all further reduce the likelihood of cross contamination and infected material reaching the consumer home. However, due to the lack of data collected on game birds acquired through hunting the number of birds consumed through the non-AGHE route can only be estimated (uncertainty).

#### 4.2.3.2 Viral distribution and titre in game birds

As AI is a respiratory pathogen the transmission routes of AI from birds to humans is believed to be through direct, close contact with respiratory secretions. However, the virus has been found in the feathers, blood, organs, bones and skin of infected game birds making these other possible routes of transmission.

#### 4.2.3.2.1 Feathers

In ducks, the titre of HPAI H5N1 in the feather follicle has been shown to be high enough to be infective to other birds and was shown to persist for days to weeks, at various temperatures, after the bird has been dispatched and the feather plucked, increasing the risk feathers may function as fomites (Yamamoto et al., 2010). A possible reservoir of the virus in the feather of water birds such as ducks or geese is preen oil, a secretion from the preen gland in these birds which they spread across their feathers during preening. Preen oil has been shown to accumulate various pathogens, including high levels of H5N1 (Karunakaran et al., 2019).

The levels of H5N8 in feathers and cloacal swabs were compared in positive flocks of ducks and geese. The viral loads were found to be 103 higher in the feather samples compared to the cloacal swabs. This result was consistent across all flocks (Gaide et al., 2021).

No publications could be found through extensive literature search that looked for levels of LPAI in game bird feathers (uncertainty).

What these publications demonstrate is that a range of AI strains persist in the feathers of various game birds at high levels, highlighting a potential risk of transmission to humans (uncertainty). The defeathering process requires close contact with the bird and the process of defeathering can

also lead to airborne infectious particles of AI (Bertran et al., 2011).

#### 4.2.3.2.2 Organs and blood

HPAI has been found in a range of organs in game birds including the cloacal bursa, spleen, liver, kidney, heart, brain, lung, pancreas and skeletal muscle (Bertran et al., 2014). The presence in the organs is believed to be due to the ability of HPAI to infect endothelial cells resulting in viremia and subsequent systemic spread (Vreman et al., 2022). In contrast, as discussed in 4.1.2.1, LPAI infections typically are restricted to the respiratory and intestinal systems and are rarely seen in other organs (Vreman et al., 2022).

#### 4.2.3.2.3 Meat

It is well established that HPAI, but not LPAI, can be found in the meat of chickens. Experimental infections have been have carried out to directly compare HPAI and LPAI dissemination in chicken meat. Whereas HPAI caused systemic spread to the breast and thigh meat, LPAI infection was found to be localised to the respiratory and GI tract (Swayne and Beck, 2005).

Although similar studies could not be identified in the literature for game birds, there have been reports of HPAI found in the meat of various game birds such as ducks (Wu et al., 2020).

In summary, the presence of HPAI in the feathers and organs of game birds is of particular concern in carcasses used for domestic purposes. Without regulations necessitating a properly functioning game larder, with good ventilation and hygiene practices, there may be a greater risk of transmission from the processing of these birds (uncertainty).

#### 4.2.4 AI in game birds during retail and consumer stages

#### 4.2.4.1 Survival in meat from game birds

There is limited research into the survival of avian influenza in meat from game birds, although a 2012 study by Beato et al. determined that HPAI H7N1 could survive for up to 75 days in duck meat kept at 4°C (Beato et al., 2012). It is assumed that AI survival within meat from game birds will behave similarly to its behaviour in poultry meat (Section 4.1.3.2).

#### 4.2.4.2 Effect of cooking game meat on AI

There is limited research into the effect of cooking on the inactivation of game bird meat, although it is considered likely that the this will be similar to the effect described for poultry meat discussed in Section 4.1.3.4.

#### 4.2.4.3 Consumption of game birds

Data from the national diet and nutrition survey (NDNS) on the consumption of duck and other game birds are provided in Table 10 and Table 11(DHSC, 2013; PHE and FSA, 2020, 2018, 2016, 2014). The majority of duck consumption relates to commercially reared poultry as described in section 4.1.1.1. We are using this data as there is limited equivalent data available on wild game duck. This data only shows meat consumption without recipes included – that is raw products to be cooked and consumed in the home rather than processed foods such as ready meals, take-aways etc. In the population ranging in age from 4-64 years old, 1.28% report eating duck. Other game is very rarely reported being consumed by only 0.17% of the participants. In infants, duck is rarely consumed with only 0.26% reporting consuming it and a negligible 0.08% reported consumption of other game. A slightly different picture can be seen in the over 65s, as this is the most popular age group for consuming duck and game, with 1.82% of participants

consuming duck and 0.46% consuming other game. With the figures for duck and game being so low, we should note that consumption or exposure estimates made with a small number of consumers may not be accurate. When the number of consumers is less than 60, this should be treated with caution and may not be representative for a large number of consumers.

| Table 10: Duck consumption (w | vithout recipes) from NDNS data |
|-------------------------------|---------------------------------|
|-------------------------------|---------------------------------|

| Age range                              | % reporting   | Chronic Mean^ | Chronic Max^ | Acute Mean^ | Acute Max^ |
|--|---------------|---------------|--------------|-------------|------------|
| Infants/children less than 4 (n=3,480) | 0.26% (n=10)  | 11.10         | 34.00        | 34.00       | 74.50      |
| 4 to 64 years old<br>(n=10,228)        | 1.28% (n=132) | 24.34         | 119.77       | 89.42       | 286.74     |
| 65+ (n=1,538)                          | 1.82% (n=28)  | 20.00         | 58.00        | 79.00       | 230.00     |

#### ^: grams/person/day

#### Table 11: Other game birds (not duck) (without recipes) consumption from NDNS data

| Age range                              | % reporting  | Chronic Mean^ | Chronic Max^ | Acute Mean^ | Acute Max <sup>^</sup> |
|--|--------------|---------------|--------------|-------------|------------------------|
| Infants/children less than 4 (n=3,480) | 0.08% (n=3)  | 9.20          | 10.20        | 33.33       | 33.33                  |
| 4 to 64 years old<br>(n=10,228)        | 0.17% (n=18) | 27.61         | 44.06        | 103.61      | 172.22                 |
| 65+ (n=1,538)                          | 0.46% (n=7)  | 47.00         | 79.00        | 150.00      | 260.00                 |

^: grams/person/day, \*: n is total number of respondents.

#### 4.2.4.3.1 LTTC game bird meat

In the FSA's flagship official statistic survey Food and You 2, 4,227 respondents were asked how often (always, most of the time, about half the time, occasionally, never) they eat duck meat when it is pink or has pink or red juices (Table 12). In wave 1 (July – October 2020), the survey found that around two thirds (67%) of respondents who eat duck report never eating it pink or when it has pink or red juices, yet almost a third (31%) eat pink duck at least occasionally; 3% report always eating duck pink, 8% do this most of the time, 4% about half the time, and 16% occasionally.

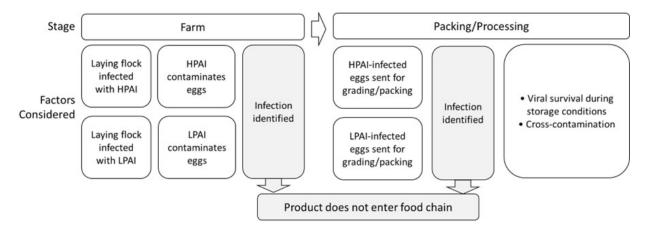
### Table 12: How often respondents to the FSA's Food and You 2 survey reported eating duck poultry meat pink

| Response            | % of respondents (n=4,227) |
|---------------------|----------------------------|
| Always              | 3%                         |
| Most of the time    | 8%                         |
| About half the time | 4%                         |
| Occasionally        | 16%                        |
| Never               | 67%                        |

### 4.3 UK Egg Production

The risk pathway, through to packing and processing step, considered for eggs infected with AI reaching the consumer is illustrated in Figure 6.

### Figure 6: Risk pathway for avian influenza infection from eggs through to the processing stage.



#### 4.3.1 Overview of Egg production in the UK

Based on 2021 industry estimates, the UK consumes 13.5 billion eggs a year. A majority of these eggs are produced within the UK, based on industry estimates of UK egg production at 11.3 billion eggs, with 1.4 billion eggs imported and 405 million eggs exported (British Lion Eggs, 2022). A majority of these eggs, 65%, enter the market at retail as shell eggs while the remainder enter as egg products (17%) or the food service industry and would thus need to adhere to food safety regulations (British Lion Eggs, 2022).

Hen laying flocks may be kept in cages, barns or be free-range; however, housing orders introduced during AI outbreaks to help reduce transmission would affect the amount of free-range eggs that are on the market; the last housing order for all poultry and captive birds in England was introduced on November 7th, 2022. (GOV.UK, 2022c). Typically, eggs are transported from farms to packing centres, where the eggs are graded and packed. Nearly 90% of UK eggs are produced following assurances required by the Lion Code scheme (British Lion Eggs); this assurance scheme introduces extra measures producers must adhere to in order to reduce the levels of Salmonella reaching consumers through table eggs. Eggs that don't meet Grade A requirements (which may be due to cracks in the shell or dirt on the shell) are classed Grade B and heat treated to remove any potential pathogens before further use (British Lion Eggs); these eggs are therefore out of scope for this risk assessment. Transport times and storage times and the temperatures associated with them are variable. These values were estimated by an expert elicitation exercise which suggested eggs are likely to encounter a temperature of about 15°C during the processing and supply chain and may take over 30 days to reach retail from the farm ( EFSA, 2014).

An additional route to market would be small producers selling eggs directly to consumers, either through on-farm sales or at local market stalls. The number of laying hens from small flocks compared to those large enough to be registered with APHA is estimated to be a very small percentage of the egg market. Additionally, these eggs would only reach a local market. The handling of these eggs in terms of storage times and temperatures is not known (uncertainty).

The FSA recommends that eggs be stored in a cool, dry place and ideally in the fridge (FSA, 2022a). Grade A eggs are required to carry a best before date (a maximum of 28 days after laying). Smaller producers that do not need to be registered with APHA must still provide best before dates for their eggs and recommend consumers keep them chilled ("The Eggs and Chicks (England) Regulations 2009"). As best before dates are indicators of quality and not safety, the FSA suggests eggs past their best before date are safe to eat provided they are thoroughly cooked (FSA, 2022a). It is not known how long consumers may keep eggs before consuming (uncertainty).

### 4.3.2 AI Infections in laying hens on the farm

#### 4.3.2.1 Effect of HPAI infection on laying hen flocks and their eggs

Infection with HPAI in commercial poultry will lead rapidly to presentation of clinical signs, typically within 2-3 days post-infection (see Section 324.1.2). There is often a drop in egg production, which ranged from 10-55% when studied in flocks naturally infected (Lu et al., 2004); in another study, this reduction reached a drop of about 30% in egg production (down from 79% egg production, based on a three day average of percentage of eggs/hen/day) within 3 days of infection in chickens infected experimentally (Swayne et al., 2012). Additionally, HPAI infection may lead to changes in the egg, such as malformed eggs (Qi et al., 2016; Swayne et al., 2012). During an outbreak, researchers found that up to 10% of eggs were malformed, including being "thin-shelled, soft-shelled or abnormally small" (Cappucci et al., 1985).

During HPAI infections, the reproductive tissue of laying hens can be infected (Harder et al., 2016). If the ovary is infected, this can lead to infectious virus being present in the yolk. If the oviduct is infected, virus will also be present in the albumin (egg whites). The virus can be present on the shell of the egg, although it is likely this is due to faecal contamination from passage through the cloaca (EFSA, 2006).

As chickens will likely stop laying eggs around 4 days post-infection due to either clinical progression of disease or death, there is the possibility that eggs laid in the period between infection and clinical detection could contain virus. Experimental infections determined that eggs laid early in the infection did not exhibit virus in or on the egg, but it was found in the last eggs laid (Bauer et al., 2010; Cappucci et al., 1985). Studies from outbreaks of naturally infected flocks estimated 7-57% of eggs may be HPAI positive (Bauer et al., 2010; Cappucci et al., 1985). Another study found that 53% of eggs from hens experimentally infected with HPAI H5N2 contained the virus (Swayne et al., 2012). Available data on viral titres isolated from HPAI-infected eggs and the time of collection post-infection are summarised in Table 13.

| Reference                 | Bird species and HPAI strain | Egg product and titre (EID <b>50</b> /ml)                                | Time taken dpi^             | Type of infection |
|---------------------------|------------------------------|--|-----------------------------|-------------------|
| (Bean et al., 1985)       | Hens H5N2                    | White: 10 <b>5.6</b><br>Yolk: 10 <b>3.6</b>                              | Last eggs laid before death | experimental      |
| (Promkuntod et al., 2006) | Quails H5N1                  | Egg internal contents: 10 <b>4.6</b><br>- 10 <b>6.2</b>                  | Not reported                | natural           |
| (Swayne et al., 2012)     | Hens H5N2                    | Shell: 10 <b>2.99</b><br>Albumin: 10 <b>3.01</b><br>Yolk: 10 <b>2.17</b> | 1-4 dpi                     | experimental      |
| (Uchida et al., 2016)     | Hens H5N8                    | Shell: 102.7-5.5<br>Albumin: 102.2-5.3<br>Yolk: 101.5-4.4                | 3-4 dpi                     | experimental      |

# Table 13: Avian Influenza viral titres isolated from egg internal contents. Table adapted and updated from Bauer et al., 2010

Given the clinical signs of HPAI in infected flocks, it is likely that the infection would be identified, and eggs stopped from reaching the market. However, there is a small window between initial infection and the presentation of clinical signs where contaminated eggs may be released from the farm containing viral loads between 101.5 - 106.2 EID50/mI.

#### 4.3.2.2 Effect on LPAI infection on laying hen flocks and their eggs

Flocks infected with LPAI viruses do not present with as many clinical signs as those infected with HPAI viruses. The only evidence may be ruffled feathers and a slight drop in egg production; it is likely it may not be detected at all (Center for Food Security and Public Health, 2022). LPAI viruses have not been found replicating in as many tissues in birds and are typically restricted to the respiratory and gastrointestinal tract (Harder et al., 2016). This means eggs are less likely to contain active LPAI virus as the reproductive tract is not infected. While some studies have not found LPAI in or on eggs from LPAI-infected flocks (Lu et al., 2004), other evidence suggests that LPAI can be found in the internal contents of eggs, although this has been reported less frequently than finding HPAI in egg internal contents (Cappucci et al., 1985; Pillai et al., 2010). Evidence also suggests that LPAI is found less frequently in eggs from LPAI-infected flocks, with only 1.67% (2/120) of eggs from commercial layers positive for H5N2 LPAI compared to 24.6% (37/156) of eggs positive for the high pathogenicity version of the strain during a natural outbreak (Cappucci et al., 1985). There is still the possibility for faecal cross contamination of the eggshell surface from passage through the cloaca. How the different LPAI viruses may behave in different bird species as far as their ability to contaminate eggs internal contents and the shell is unknown (uncertainty).

#### 4.3.3 AI contamination in eggs during the grading and packing process

As noted above, up to 10% of eggs produced by hens infected with HPAI were malformed, including being "thin-shelled, soft-shelled or abnormally small" (Cappucci et al., 1985). These eggs would be removed from the food chain during the grading process (APHA, 2018). Class A eggs are not allowed to have foreign matter on them, which means eggs with visible faecal contamination, which may carry the AI virus (see discussion in Section 4.4.1), will also be removed from the food chain (APHA, 2018).

#### 4.3.4 AI in eggs during the retail and consumer stages

#### 4.3.4.1 Survival in eggs

Experimental infection with HPAI strain H5N2 in and on eggs provided some estimation of the viral survival during storage times of different lengths and temperatures. When virus was applied on the eggshell and allowed to dry, it could not be re-isolated from the surface after 3-4 hours, regardless of the temperature exposure. The same held true even if the virus was mixed with "droppings" (assumed to be bird faeces). However, active virus could be isolated from the internal contents of the eggs. Virus survived better in the yolk compared to the albumin, and viral titres in both dropped off as either length of storage or temperature increased. At 4°C, virus was still detected after 17 days in both the yolk and albumin, whereas it was only found in the yolk after 17 days at both 15°C and 20°C (de Wit et al., 2004).

#### 4.3.4.2 Evidence for heat inactivation in eggs

Inactivation of AI in eggs at pasteurisation temperatures has been studied using experimentally inoculated egg samples. Swayne and Beck (2004) found that for both HPAI and LPAI H5N2, viral

titre was reduced by 90% within 20 seconds at 61°C in both homogenised whole egg and in egg whites. Another study found there was a 5-log reduction in H5N2 HPAI in 34 seconds for homogenised whole egg and in 4 seconds for plain egg yolk at 60°C (Chmielewski et al., 2013). Viral reduction could also be achieved at lower temperatures for longer treatments, ranging from 4.3 minutes for reduction of HPAI in liquid egg at 55°C to 11 minutes for HPAI in whole homogenised egg at the same temperature (Swayne and Beck, 2004).

#### 4.3.4.3 Consumption of eggs

Data from the national diet and nutrition survey (NDNS) on the consumption of eggs by different age ranges are presented in Table 14 - Table 16 (DHSC, 2013; PHE and FSA, 2020, 2018, 2016, 2014). These data include foods consumed "with recipes", meaning that data is included both when the egg is eaten as an individual item and when it forms part of a recipe. Since AI can be inactivated by thorough cooking of eggs, data on eggs that would be LTTC was also included. In the tables below, "all egg consumption" would include raw, poached, and thoroughly cooked eggs.

#### Table 14: Egg consumption data for infants and children (less than 5) n=4,227)

| Egg consumption     | % reporting     | Chronic Mean^ | Chronic Max <sup>^</sup> | Acute Mean^ | Acute Max <sup>^</sup> |
|---------------------|-----------------|---------------|--------------------------|-------------|------------------------|
| Raw eggs            | 1.4% (n=60)     | 2.4           | 34.4                     | 4.2         | 30.1                   |
| Poached eggs        | 1.2% (n=49)     | 16            | 43                       | 58.6        | 112.1                  |
| All egg consumption | 72.5% (n=3,065) | 9.9           | 100.3                    | 29.5        | 232.6                  |

#### ^: grams/person/day

#### Table 15: Egg consumption data for people aged 5 to 64 years. n=14,995

| Egg consumption     | % reporting      | Chronic Mean^ | Chronic Max <sup>^</sup> | Acute Mean^ | Acute Max <sup>^</sup> |
|---------------------|------------------|---------------|--------------------------|-------------|------------------------|
| Raw eggs            | 3.6% (n=541)     | 1.2           | 28                       | 4.3         | 80.7                   |
| Poached eggs        | 5.4% (n=815)     | 30.4          | 210                      | 99          | 400                    |
| All egg consumption | 94.6% (n=14,189) | 22.7          | 870                      | 59.5        | 870                    |

#### ^: grams/person/day

#### Table 16: Egg consumption data for UK people aged 65+. n=1,538

| Egg consumption     | % reporting    | Chronic Mean^ | Chronic Max <sup>^</sup> | Acute Mean^ | Acute Max^ |
|---------------------|----------------|---------------|--------------------------|-------------|------------|
| Raw eggs            | 4.8% (n=74     | 1.2           | 10                       | 3.2         | 24         |
| Poached eggs        | 9.1% (n=140)   | 24            | 130                      | 78          | 150        |
| All egg consumption | 94.5% (n=1454) | 23            | 150                      | 60          | 280        |

#### ^: grams/person/day

The data reveal that a vast majority of the UK population, in all age groups, consumes eggs. The amount that is eaten raw or poached is much smaller, with the elderly reporting the highest percentage of consumers eating raw and poached eggs. While those aged 5 to 64 years report eating raw and poached eggs less frequently than those that are 65+, they report eating larger servings of these products.

Data from the FSA's Food and You survey was also reviewed to support evidence on egg consumption in the UK (Table 17; (FSA, 2021)). Self-reported rates of consumption of eggs eaten

raw, LTTC and thoroughly cooked indicated that, of the respondents that consume eggs at home, only a small percentage eat raw eggs once a week or more. A higher percentage eat LTTC eggs and thoroughly cooked eggs. 61% of respondents never eat raw eggs, 22% never eat LTTC eggs and 8% never eat thoroughly cooked eggs. Both data sets indicate that when eggs are consumed in the UK, they are most frequently eaten thoroughly cooked.

#### Table 17: Frequency of eggs eaten raw, LTTC or thoroughly cooked in the UK.

Data from Food and You 2 (Wave 2) survey on the eating habits of UK consumers (FSA, 2021).

| Occurrence             | Raw (a) | LTTC (b) | Cooked thoroughly |
|------------------------|---------|----------|-------------------|
| Every day              | 1%      | 1%       | 2%                |
| Most days              | 2%      | 4%       | 5%                |
| 2 to 3 times a week    | 4%      | 12%      | 15%               |
| About once a week      | 7%      | 24%      | 27%               |
| 2 to 3 times a month   | 4%      | 15%      | 16%               |
| About once a month     | 4%      | 11%      | 15%               |
| Less than once a month | 14%     | 9%       | 1%                |
| Never                  | 61%     | 22%      | 8%                |

a: eggs that are uncooked for example, in homemade mayonnaise or homemade desserts like mousse or soft meringues

b: eggs that have a runny yolk for example, soft boiled

c: eggs that have a firm yolk for example, hard boiled

# 4.4 Cross-contamination of AI from birds and poultry products

#### 4.4.1 Survival in faeces

Both HPAI and LPAI is shed in the faeces of both symptomatic and asymptomatic carriers of the disease. It is quite often associated with the further spread and infectivity of the disease – particularly with the wild bird population contaminating water and feed sources.

Experimentally it has been shown that AI might remain infectious in duck faeces within a wide range of days to several weeks depending on temperature conditions. A conservatively estimated expected viral titre in 1g of duck faeces was replicated by spiking duck faeces with tissue culture infective dose (TCID) of 106 TCID50/ml in 1g. The study found that influenza viruses may remain infectious in duck faeces for periods of time ranging from a few days (at 30°C and 20°C) or a few weeks (at 10°C) to several months (at 0°C). The study took into account their previous work where a replicated egg infective dose per g of fresh duck faeces became undetectable after the faeces were dried overnight at room temperature (20°C), while in wet faeces, the virus remained viable for 4 to 6 days at 37°C leading to uncertainty about the virus' persistence in dried faecal matter (Nazir et al., 2011).

A study held in India showed that the survival time of the virus in faeces was up to 30 days at 4°C and for 7 days at 20°C, however it only persisted for up to 24h and 18h at 37°C and 42°C respectively, in both wet and dry faeces (Kurmi et al., 2013).

When spiking faeces with HPAI H5N8 and LPAI H6N2, differences in persistence were seen between the strains. HPAI persisted until the end of the experimental period, 96 hours, while the LPAI strain was only detected until 24 hours after the material was spiked. In both cases, temperatures ranged from 19 to 22.5 °C in the room where the experiment was performed (Hauck

et al., 2017).

As noted in Section 4.3.3, eggs with visible contamination on the shell will be removed during the grading process, reducing the amount of faecal contamination consumers may encounter when handling table eggs. Insufficient cleaning of in-feather poultry or game birds to remove any faecal contamination could be a route of transmission during slaughter and processing of the carcass, particularly in a domestic setting (uncertainty). There is strong evidence that the virus will persist in faecal matter and without thorough cooking and hygienic handling of contaminated products, cross-contamination could occur.

#### 4.4.2 Survival in the environment

Environmental persistence of AI virus has been determined as an important factor for the epidemiology of the virus within wild bird populations and within aquatic habitats. The wild bird population is proven to be an important vector in the spread of the virus to the domestic and commercial poultry industries. Surface water is considered to be a major site of environmental contamination (Keeler et al., 2014).

The majority of studies that have investigated the persistence of AI in the environment have tended to concentrate on the persistence of the virus in faeces. Wood et al., (2010) carried out a study to investigate how the virus persisted in the environment on different surfaces, in different humidity and temperature conditions. They tested the persistence on porous surfaces (soil and faeces) and non-porous surfaces (glass and galvanised metal) (Wood et al., 2010). Using test results and prediction modelling they estimated that HPAI could survive up to 57 days on galvanised metal and 72 days on glass surfaces. HPAI persisted longer without exposure to both UV-A and UV-B simulated sunlight than with exposure. The detrimental effect of UV was markedly higher on the HPAI on the non-porous surfaces. Low temperatures lead to a longer persistence in all cases. Overall, the study concluded that the HPAI H5N1 virus used in the study tends to survive longer in the environment (outside a host) in areas that are relatively colder and have less sunlight.

Brown et al., (2009) investigated AI virus persistence in water at different pH, and temperatures. They found that all the AI viruses tested in the study were, in general, most stable at pH 7.4-8.2 at lower temperatures of <17°C, and in fresh to brackish salinities (salinity ranging from 0 to 20,000 ppm) (Brown et al., 2009). The study by Keeler et al., (2014) showed similar results in that persistence of the virus was longest in waters at temperatures as low as <17°C, neutral to basic pH of 7.0-8.5, and a low salinity of less than 0.5 ppt (Keeler et al., 2014)

#### 4.4.3 Effect of disinfectants on avian influenza

Al can be inactivated by a range of disinfectants at the recommended concentrations (Shahid et al., 2009):

- Formalin (0.2, 0.4 and 0.6% after 15 minutes)
- lodine crystals (0.4 and 0.6% after 15 minutes)
- Phenol crystals (0.4 and 0.6% after 15 minutes
- Virkon®-S (0.2% after 45 minutes, 0.5 and 1.0% after 15 minutes)
- Surf Excel (soap), Life Buoy (detergent) and Caustic soda (alkali) inactivated the virus at 0.1, 0.2 and 0.3% concentration, respectively, after 5 minutes contact time.

Guan et al., (2015) investigated an effectivity of disinfectant against avian influenza AI during winter months. They have shown that commercial disinfectants, such as Virkon and Accel, when supplemented with antifreeze agent (propylene glycol-PG, methanol-MeOH, or calcium chloride-CaCl2) inactivated AI within 5min at -20°C or 21°C by 106. However, the PG and MeOH did not kill AI alone at those temperatures, but 20% CaCl2 inactivated 105 AI within 10 min. Similarly,

Kabir at al. (2021) tested the microbicidal activity of a mixture of quaternary ammonium compounds (QACs) and food grade additive grade calcium hydroxide (FdCa(OH)2) at -20°C using anti-freeze agent (AFA) containing methanol against a variety of viruses, including AI (Kabir et al., 2021). They found that avian influenza was inactivated within 30min of the treatment at -20°C, and within 10min at 1°C (Kabir et al., 2021). Another study investigated survival of HPAI H5N1 against chlorination, and found that free chlorine concentrations (0.52–1.08 mg/L) typically used in drinking water are sufficient to inactivate the virus by >3 orders of magnitude, after 1 min exposure (Rice et al., 2007). An acidic agent, potassium monopersulfate (PMPS) was investigated against AI, and showed that it is able to inactivate the virus at 5000, 2,500, and 1,250 ppm within 30sec, 5min and 15min, respectively (Sonthipet et al., 2018). Ota et al. (2016) investigated the antiviral properties of calcinated egg shell (Egg-CaO) in the form of powder and aqueous solutions against pathogens, including AI (Ota et al., 2016). They showed that AI was inactivated after 1 h exposure, and that the powder was effective against AI after both exposure to sunlight for 2 weeks or re-suspension of the powder up to 7 times with water - simulating the harsher conditions in the field rather than the laboratory. (Ota et al., 2016).

### 4.5 Infectivity of avian influenza in the human GI tract

Influenza virus hemagglutinin (HA) proteins binding to host cell sialic acid-based receptors is known to be a first step in viral invasion of the host, as well as considered to be the first barrier to cross-species transmission (Zhao and Pu, 2022). Avian and human influenza viruses have a different sialic acid (SA)-binding preferences, but only a few amino acid changes in the HA protein are needed to cause a switch from avian to human receptor specificity (de Graaf and Fouchier, 2014). Al viruses have a preference for SA linked to the galactose in an ?-2,3 format, whereas human influenza viruses (H1, H2, and H3) prefer SA that are linked in an ?-2,6 format (de Graaf and Fouchier, 2014). The human upper respiratory tract lacks SA ?-2,3-Gal, which may help explain the species barrier seen between humans and birds in becoming infected with AI.

One component of saliva that can have anti-influenza virus activity is sialic acid-containing molecules. Limsuwat et al. (2014) investigated the role of human saliva against influenza viruses by hemagglutination inhibition (HI) and neutralization (NT) assays (Limsuwat et al., 2014). They showed that human saliva had higher antiviral activity against human influenza viruses compared to an H5N1 avian influenza strain (Limsuwat et al., 2014). They then went on the characterise the sialic acids present in human saliva, identifying that SA ?-2,6-Gal was more abundant than SA ?-2,3-Gal (Limsuwat et al., 2016). This supports the evidence that human saliva will have lower antiviral activity against AI viruses due to the presence of a greater amount of SA with preferential binding to human-adapted influenza viruses.

Since human cases of AI also report gastrointestinal symptoms, one study has investigated whether AI H5N1 virus can infect and replicate in the human GI tract (Shu et al., 2010). While SA ?-2,6-Gal was abundantly expressed along the entire GI tract, they did identify expression of SA ? -2,3-Gal, which increased traveling from the ileum to the rectum. Using human *ex vivo* colon samples, the research group was able to demonstrate infection of the tissue by the AI virus. Increases in viral titre in the supernatant of the *ex vivo* cells suggested the virus was also capable of replication (Shu et al., 2010). For evidence of active AI infection in the human GI tract, colonic samples of an H5N1 deceased patient were stained for presence of the nucleoprotein viral antigen, which was detected in the tissue. Furthermore, in two other H5N1 patients experiencing diarrhoea as a symptom, viral RNA was recovered from faecal samples. However, this was the only study identified during literature searches specifically investigating expression of AI viral receptors in the human GI tract. While epidemiological evidence does not indicate that the GI tract is a common route for human infection by AI viruses, these results highlight that it remains a possibility.