

A risk-based framework for live bird shops (LBSs) in Gujarat, India: Identifying pathogen entry and exposure risk profiles

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ABSTRACT

Live bird shops (LBSs) and markets (LBMs) are frequent venues for chicken trading in Asia. Public-health research commonly explores LBMs across Southeast Asia, emphasizing their relevance in infectious disease epidemiology. However, LBSs' role in pathogen transmission remains understudied, particularly in India, where broiler and indigenous chickens (deshi) are common. This study, conducted in Gujarat, India, described LBSs practices and assessed their potential for pathogen entry and exposure to chickens and humans. Standardized questionnaires and field observations were collected at 86 LBSs across eight major tribal and non-tribal cities, from December 2020 to March 2021. A risk-assessment framework identified three pathogen risk-pathways: LBS pathogen entry, chicken exposure and human exposure. Multivariate analyses assessed LBSs' risk profiles for each risk-pathway, distinguishing between chicken types where applicable. The findings revealed three LBS clusters categorized as lower- or higher-risk for each risk-pathway and respective chicken type sold. LBSs' region was a strong determinant of cluster composition. Higher-risk LBSs were typically located in non-tribal cities and received chicken supplies from farther away and more suppliers compared to other LBSs. The relatively uniform distribution of clusters per city, noticeable in tribal regions, suggests an opportunity for targeted interventions to mitigate pathogen transmission. Higher-risk clusters for chicken exposure were linked to free-roaming rearing conditions for broiler LBSs and a high proportion of surplus for deshi LBSs. Human exposure risks were driven by inadequate individual-protective-equipment use, poor hygiene standards and failure to separate slaughtering from sales areas. Mixing of unsold poultry with newly supplied birds and keeping live birds overnight were observed across all clusters, potentially facilitating spread and persistence of poultry pathogens. This study identified LBS risk-clusters with distinct management and geographical characteristics, highlighting the need for tailored disease control strategies. The presented risk-based framework offers a valuable tool for targeted interventions in similar poultry trade settings.

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1. Background

Poultry production is one of the fastest growing sectors in India's agricultural industry, driven by economic development, human population growth and urbanisation. The vertical integration of broiler production has played a major role in this growth, especially in southern and western India, where Gujarat state is located (Husain, 2014; ICAR-DPR, 2008). While fast-growing, exotic broiler chickens (hereafter referred to as broilers) dominate chicken meat production, indigenous chickens (deshi) (Chatterjee and Rajkumar, 2015; FAO, 2020) still account for approximately 21.7% of Gujarat's poultry population, according to the most recent Livestock Census (DAHDF, 2019a). Extensive deshi production, characterised by low biosecurity (Chatterjee and Rajkumar, 2015; Conan et al., 2012) and scavenging-based rearing, has remained a mainstay of the rural economy in India (Haunshi and Rajkumar et al.; Pal et al., 2020). As observed in other low and middle-income countries, this sector has been driven by high poultry meat demand (Ranasinghe et al., 2024; Bhimraj et al., 2018; Bhuiyan et al., 2005; Haitook et al., 2003; Khan, 2008).

In Asia, live bird markets (LBMs) are common venues where multiple poultry species and breeds are housed together and offered for sale by several vendors (Cardona et al., 2009; Høg et al., 2021). Due to their association with a high burden of avian influenza viruses (AIVs) in several countries, including Bangladesh (Høg et al., 2021; Islam et al., 2023a, 2023b; Chowdhury et al., 2020; Rebecca and Moya, 2019), Vietnam (Fournié et al., 2016; Soares Magalhães et al., 2010), China (Huo et al., 2012; Martin et al., 2011; Zhou et al., 2015) and Indonesia (Indriani et al., 2010; Henning et al., 2019), LBMs have attracted significant attention. Numerous studies have been conducted to characterise the practices and conditions within these settings that promote disease risks. Factors such as the high traffic of vehicles and people, mixing of poultry from different sources, frequent holding of unsold chickens overnight, poor biosecurity and hygiene practices, have been identified as facilitating the maintenance and spread of zoonotic pathogens, including AIV (Soares Magalhães et al., 2010, 2012; Huo et al., 2012; Zhou et al., 2015; Bulaga et al., 2003; Gilbert et al., 2014; Dhingra et al., 2014; Nasreen et al., 2015; Bui et al., 2019) and *Salmonella* spp (Nidaullah et al., 2017; Anbazhagan et al., 2019; Yang et al., 2017). While research extensively focuses on LBMs across Southeast Asia, live bird shops (LBSs) in India may differ in management practices and in the resulting pathogen-related risks, warranting a focused investigation.

LBSs are physical establishments in which a single vendor sells poultry and poultry-related products. In Gujarat, LBSs are the primary outlets for selling chickens to consumers. Given that poultry are kept alive in these settings, they may pose a similar risk for animal and human health as LBMs. However, unlike LBMs, little information exists regarding the practices and conditions in LBSs and their potential in facilitating pathogen spread among birds and to people. The present study aimed to identify practices adopted in LBSs which influence the risk for pathogen transmission amongst poultry and human populations, and to define LBS risk profiles based on these practices.

2. Materials and methods

2.1. Study design

A key informant study conducted in Gujarat identified broiler and deshi as the two main poultry types consumed in the state, and urban and peri-urban LBSs as the main outlets for poultry slaughter and sale to consumers, restaurants, dressed chicken retail shops and caterers. LBSs were selected as the units of interest and stratified by city. Further, stratification of cities was based on their location in tribal and non-tribal areas, as differences in demand and trading practices were anticipated. Tribal areas were defined by a predominance of tribal populations and often characterized by lower economic status (Ahmedabad and Speed, 2018; Desai and Vidyapith, 2018; Gurnam, 2001), and demand for

poultry meat and trading practices were thought to be potentially different from non-tribal areas. Cities were purposively selected for each group: non-tribal (Bhuj, Rajkot, Ahmedabad and Vadodara) and tribal (Godhra, Himatnagar, Bharuch and Surat). These are large cities located in different districts and covering different agro-ecological zones across the state (Fig. 1).

In the absence of reliable LBS census data, sampling locations were randomly generated within each city's municipal corporation boundaries, ensuring that the Euclidian distance between any two sampling locations was at least 10% of the distance between the city's furthest extremities. We aimed to survey 50 LBSs selling broiler and 50 selling deshi chickens, with this sample size designed to capture variation in management practices relevant to pathogen introduction and exposure, both within and across cities and chicken types. Seven sampling locations were generated in the two largest cities, Ahmedabad and Surat, and six in the remaining cities. For each sampling location, the nearest LBS were opportunistically identified by asking people who were met in situ. If the LBS only sold one chicken type (e.g., broiler), a second LBS - the closest LBS selling the other type (e.g., deshi) - was also recruited. As a result, the total number of recruited LBSs ranged from 50 (if all LBSs sold both chicken types) to 100 (if the nearest LBSs to each sampling location only sold one chicken type). This procedure was performed using the *sp* package (Pebesma and Bivand, 2024) in R 4.2.2 (R Core Team, 2022).

2.2. Data collection

A structured questionnaire was applied in Gujarati language to either a shop owner or worker (available in Appendix I – Annexes 1–2 for consultation). In addition, observations were recorded using a standardised form. Collection forms (questionnaire and observation) were pilot-tested in a subset of LBSs excluded from the main study. Data collected included demographic and socio-economic characteristics of the interviewee, location and structure of the LBS, species and types of poultry sold, number of chickens sold, management of unsold chickens, types and number of suppliers, hygiene, slaughtering and processing practices. The origin of the poultry was traced through snowball sampling (Johnson, 2014): each LBS identified up to three broiler and/or deshi suppliers, either farms or traders, with the latter being contacted and asked about their suppliers. The approach was repeated to identify the locations of supplying farms.

2.3. Statistical data analysis

2.3.1. Data entry and management

Questionnaire data were entered using ODK (Open Data Kit) (Hartung et al., 2010) and uploaded in a Comma Separated Values format. All data management and analysis were performed using R 4.2.2 (R Core Team, 2022).

2.3.2. Risk assessment framework and descriptive analysis

A risk assessment framework was developed to identify potential pathways for pathogen introduction and exposure in the LBSs. According to WOA's definition (WOAH (OIE), 2022), risk assessment estimates the risks associated with defined hazards – in this case, any avian zoonotic pathogen.

The first step, assessment of pathogen entry, described the main biological pathways for the introduction of pathogens to a LBS. Secondly, the exposure assessment identified the main biological pathways by which chickens and/or humans could be exposed to pathogens following their introduction to the LBS. Pathways were developed separately for chickens and humans.

A descriptive analysis summarised LBS's characteristics, captured by 28 variables (Appendix II – Table A1), likely to influence risk attributed to pathogen entry or exposure pathways. Some variables belonged to both exposure pathways due to a role in exposing both chickens and

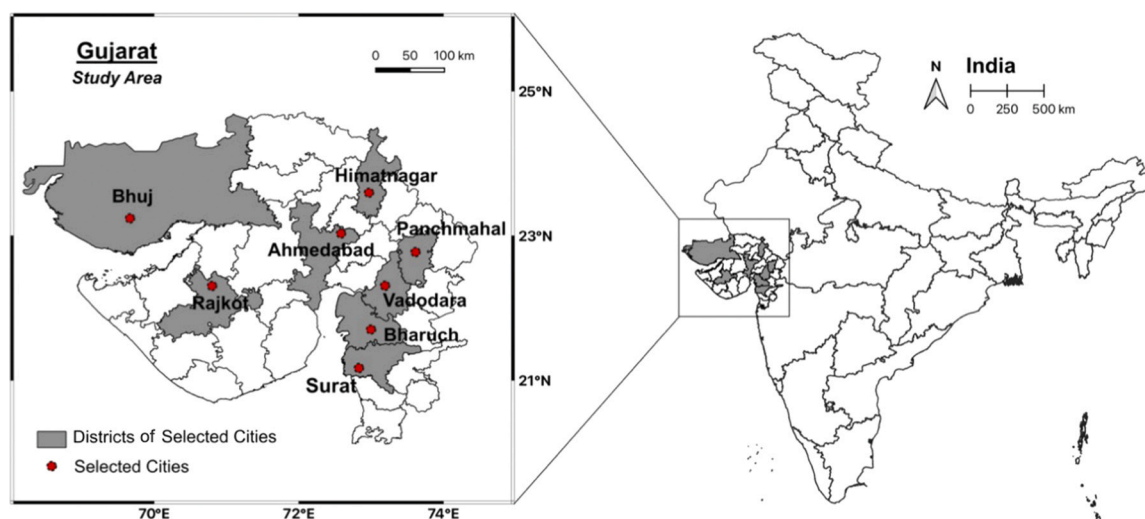


Fig. 1. Representation of the study area in India. A state-boundary map of India (right-side) and a district-boundary map of Gujarat (left), with selected cities and respective districts.

humans to pathogens. Twelve additional variables were supplementary, as they provided valuable socio-demographic context ([Appendix II – Table A2](#)).

2.4. Multivariate analysis and hierarchical clustering

Multivariate analysis and hierarchical clustering were performed to define LBSs' profiles based on the selected variables, with separate analyses for each risk pathway: pathogen entry, chicken exposure and human exposure. Pathogen entry and chicken exposure pathways were analysed separately for LBSs selling broiler and deshi as the related variables were chicken-type specific, while human exposure included all LBSs. LBSs selling both types were included in each respective analysis, based on practices relevant to each chicken type, rather than in a separate "mixed" category; however, to account for potential risks with multi-type sales, the number of poultry species/types sold was included in the analysis.

Two multivariate statistical methods were used to reduce data dimensionality. Given that pathogen entry and chicken exposure assessments included categorical and numerical variables, factor analysis for mixed data (FAMD) was used ([Kassambara, 2017a](#)). Multiple correspondence analysis (MCA) was used for human exposure as all variables were categorical ([Manly et al.,](#)), and the Greenacre's correction was applied ([Lele et al., 2007](#)). These methods transformed a set of correlated variables into fewer synthetic, independent factors that capture a large fraction of the data variability. Factors contributing substantially to the overall variance were retained, determined by a marked eigenvalue drop in scree plots ([Cattell, 1966](#)) and eigenvalues above 1 ([Kaiser, 1960](#)).

Hierarchical clustering ([Kassambara, 2017b](#); [Manly and Alberto, 2016](#)) was then used to group LBSs into relatively homogeneous clusters according to factors previously selected by the FAMD or MCA. Clusters were formed using Euclidean distances as a measure of dissimilarity, and Ward's criterion was adopted to minimize the total within-cluster variance. The optimal number of clusters was determined by identifying the level at which the gain in within-cluster inertia dropped most sharply in the dendrogram, following recommended methodology ([Manly et al.](#); [Kassambara et al.](#)). For each risk pathway, clusters were interpreted based on variable contributions to retained dimensions ([Appendix II – Table A4](#)). Variables and category levels significantly associated with each cluster ($p < 0.05$) were identified using *FactoMineR* ([Husson et al., 2025](#)) in R 4.2.2 (R Core Team, 2022). Clusters were categorized as lower risk or higher risk based on their main practices.

For each cluster partition, we assessed whether LBSs from the same clusters tended to group within cities using the Shannon diversity index (S_j) ([Zahl, 1977](#); [Dejong, 1975](#)). It was computed for each city as $S_j = -\sum_i p_{ij} \log_2 p_{ij}$, where p_{ij} is the proportion of LBSs belonging to cluster i in city j . It was then averaged over all cities and normalised by its maximum possible value, $\log_2 n$, with n being the number of clusters. An index of 0 means that all LBSs in each city belonged to the same cluster while a normalised index of 1 indicates a high diversity of clusters in each city. Pianka niche overlap indices ([Pianka, 1974](#)) were computed for each cluster partition to assess how similar LBS compositions were between cities. This index was calculated for any two cities j and k as

$$P_{jk} = P_{kj} = \frac{\sum_i p_{ij} p_{ik}}{\sqrt{\sum_i p_{ij}^2 \sum_i p_{ik}^2}}, \text{ where } p_{ij} \text{ is the proportion of shops in city } j$$

belonging to cluster i . An average index of all pairwise city combinations was computed, with an index of 1 indicating identical composition between cities. To test whether the observed similarity was greater or less than expected by chance, we generated 10,000 random permutations of cluster membership and calculated the index for each simulation. P-values were computed as the proportion of simulated indices that were greater than or equal to (or less than or equal to) the observed index, with significance set at $p < 0.05$. FAMD, MCA and hierarchical clustering were performed using *FactoMineR* ([Husson et al., 2025](#)) and *ExPosition* ([Beaton et al., 2014](#)), from R 4.2.2 (R Core Team, 2022).

2.5. Ethics

Ethical approval was obtained from the Royal Veterinary College Clinical Research and Ethical Review Board (CRERB:2020 1983–3). Participation in the questionnaire survey was voluntary, and oral consent was obtained from shop owners, workers and traders at all intervening sites. The present article was written following the STROBE reporting guidelines ([von Elm et al., 2008](#)).

3. Results

A total of 86 LBSs were successfully recruited from December 2020 to March 2021. Only one LBS refused participation, and one did not complete one of the questionnaires. Demographic characteristics of interviewees are summarised in [Appendix II – Table A3](#). Among the 86 recruited LBSs, 72 (83.7 %) sold broilers and 61 (74.4 %) sold deshi, including 47 (54.7 %) selling both types. All but one LBS (98.8 %) were open every day. Most LBSs operated for 11–13 h a day (IQR, median: 12), and employed 1–3 workers (IQR, median: 2). Most LBSs (87.2 %, 75/86) were located in the Ahmedabad district.

n = 75) had a permanent housing structure and were licenced by the city corporation (95.6 %, n = 83) or the Food Safety and Standards Authority of India (1.2 %, n = 1), with only two LBSs, in Surat, being unlicensed (2.3 %).

Variables considered as influencing risks of interest are represented in Fig. 2 and Appendix II – Table A1. Chicken sources (i.e., supplier chain related variables), housing characteristics (e.g., LBSs' neighbourhood and water source) and the presence of multiple animal species in or near the shop were considered as promoting pathogen entry. Risk of exposure was influenced by three groups of variables. The first group included housing characteristics, such as storage of live birds and chicken products, and features of slaughtering facilities (e.g., separation between live chickens and carcasses, level of automatization). The second group covered poultry management practices relevant to chicken exposure (e.g., time held until sale and number of chickens sold). The third group included slaughtering procedures and sanitary conditions relevant for chicken and/or human exposure. Contributions of individual variables to the retained dimensions used in clustering are presented in Appendix II – Table A4, highlighting which variables most strongly influenced LBSs categorization across the three risk pathways.

Following hierarchical clustering, the characteristics of shop clusters were described for each risk pathway: pathogen entry in Table 1 (broiler and deshi LBSs), chicken exposure in Table 2 (broiler and deshi LBSs) and human exposure in Table 3. Three clusters were defined for each pathway and chicken type: B.1–6 for broilers in the entry and exposure pathways, D.1–6 for deshi in the entry and exposure pathways and H.1–3 for the human exposure pathway. Details relative to the results of the cluster analyses are presented in Appendix II – Figures A1–5.

3.1. Pathogen entry risk pathway (Table 1)

Most LBSs in all clusters (B.1–3 and D.1–3) were located in high-traffic areas and sold several chicken types or poultry species. Running water was used in most LBSs, except for D.1. Lower-risk clusters, B.1 and D.1, accounted for 54.2 % (n = 39) of broiler LBSs and 59.0 % (n = 36) of deshi LBSs, respectively. These LBSs exclusively sourced chickens from one supplier, sold only poultry and were less likely to have other LBSs in their vicinity (<100 m). B.2 and D.2 LBSs were defined by selling multiple poultry and animal species, having a single supplier, and being located near other LBSs.

In contrast, clusters B.3 and D.3 included LBSs that also sold multiple chicken types or poultry species, but less frequently sold other animal species compared to B.2 and D.2. These shops had more suppliers than those in other clusters and are often located near other LBSs.

3.2. Chicken exposure risk pathway (Table 2)

All clusters (B.4–6 and D.4–6) shared common features, such as retaining chickens for a median of 12 h before sale, keeping unsold birds overnight in the LBS and mixing them with newly supplied birds the following day, although this was less frequent in clusters B.6 and D.6. However, clusters B.6 and D.6 exhibited a heightened risk of chicken exposure due to the relatively higher proportion of shops holding birds in free-roaming conditions (i.e., legs untied). The proportion of LBS storing waste near live poultry and appearing dirty during field visits (i.e., large amount of faeces observed) was generally low but highest in B.6 and D.6, with cleaning occurring less frequently (i.e., generally once a day), and with birds often slaughtered away from open drains in these LBSs. In D.4, slaughtering is uniquely avoided in-shop.

B.5 and D.5 have the highest proportion of days with unsold birds, with D.5 also showing a high proportion of unsold deshi at the end of such days. Both clusters also have the highest proportion of LBSs where slaughtering occurs near live birds, and bird mixing was more frequent. In B.5, poultry were usually free roaming in fenced areas, with segregation of species. Conversely, D.5 LBSs typically cage their poultry but frequently mix different species. Additionally, B.5 also accounted for the

largest shops in terms of daily broiler sales.

3.3. Human exposure risk pathway (Table 3)

Most LBSs across clusters lack defeathering machines and do not sell waste products (including feathers and offal). Slaughtering mainly occurs indoor, with a higher proportion of H.3 LBSs slaughtering near sales areas compared to other clusters. Despite more frequent cleaning, birds in H.3 were slaughtered near open drains, carcasses were stored on non-protected surfaces, and IPE were not used. In contrast, H.1 LBSs avoid open drains for slaughtering and store carcasses on protected counters or fridges, though often close to live poultry. H.2 LBSs did not sell dressed poultry and employees, like those in H.1, were more likely to wear IPE.

3.4. Socio-demographic characteristics of clusters

For most risk pathways, the diversity of LBS types per city was lower than expected by chance as indicated by low average Shannon indices, with tribal cities being generally less diverse than non-tribal cities. Pianka indices showed that, overall, cities were more dissimilar in their LBS composition than expected by chance, although tribal cities were more similar to one another than non-tribal cities (Table 4). Lower-risk B.1 and D.1 LBSs were mainly located in tribal cities (59.0 % and 66.7 %, respectively), whereas higher-risk B2–3 and D2–3 LBSs were mostly in non-tribal cities (Table 5). LBSs that did not sell dressed chickens (H.2) were predominantly located in non-tribal cities.

LBSs in tribal and non-tribal cities were predominantly supplied in broilers by farms located within the same type of region (tribal vs. non-tribal). However, most deshi supplies came from tribal regions. LBSs in non-tribal cities sourced deshis from non-contiguous, tribal regions (Table 5).

Among the 86 investigated LBSs, 27.8 % (n = 15) consistently clustered in low-risk groups across all three risk pathways – pathogen entry, chicken exposure and human exposure (Table 6). Conversely, 25.9 % (n = 14) LBSs were consistently categorized as high-risk across all pathways.

4. Discussion

Our study revealed heterogeneity in risk profiles of LBSs in Gujarat, India, highlighting various factors contributing to higher risk of pathogen entry and exposure for both chickens and humans. Of the 86 LBSs surveyed, nearly three-quarters (72 %) of LBSs were classified as high-risk in at least one risk pathway, with many exhibiting mixed-risk profiles. A quarter of LBSs (26 %) fell into the highest-risk category, simultaneously classified as high-risk for entry, animal and human exposures.

Notably, geographic location was a strong determinant of LBS profiles, consistent with previous findings from LBMs in Northern Vietnam and Cambodia (Fournié et al., 2012). Higher-risk clusters included LBSs that were more consistently located in non-tribal regions (B2–3; D2–3) than in tribal regions (D1). This difference may be attributed to specific characteristics typical of these LBSs, such as fewer supply sources as observed D.1, or other characteristics not accounted in this study. Moreover, deshi chicken supplies predominantly originated from tribal and farther distances (i.e., non-contiguous cities), reflecting the heavy reliance of LBSs on chickens sourced from tribal regions, where backyard systems are more prevalent and the mainstay of indigenous breeds. The reliance on tribal areas as supply sources likely reflects the lower availability of backyard chickens in more urbanized regions, leading traders to source from rural and less densely populated areas. These patterns align with observations from Bangladesh (Moyen et al., 2021) and Cambodia (Van Kerkhove et al., 2009), where indigenous chickens, while often sold locally, may enter larger trade networks and be transported far from their point of origin. Similarly, a recent study by Awais et al. (2022), in Pakistan highlighted how varied sourcing patterns and

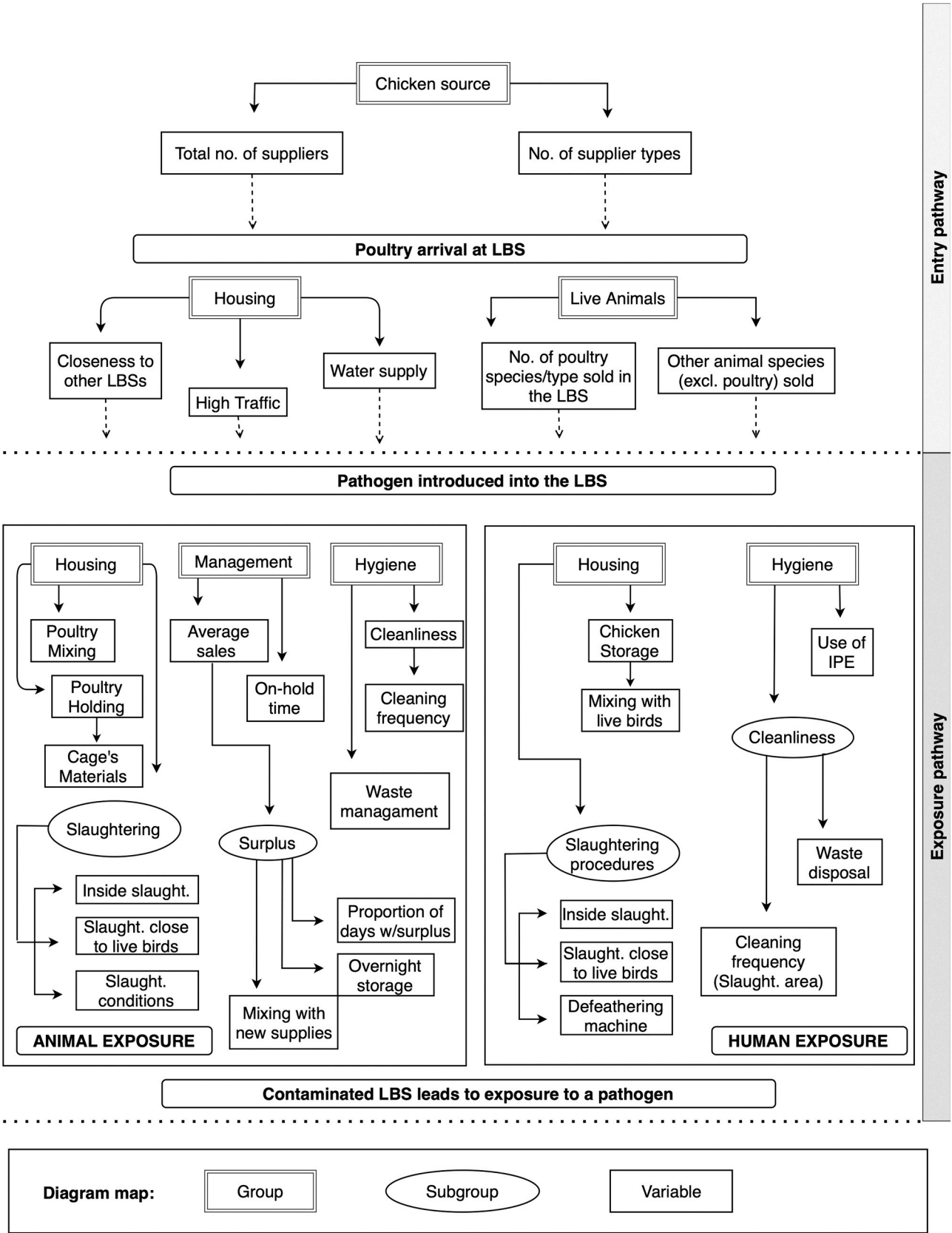


Fig. 2. Risk pathways for pathogen introduction and exposure in a LBS. Variables were associated with different pathways, if they influenced the risk of pathogen introduction into the shop (Entry pathway) or the risk of exposure following introduction (Exposure pathway). The exposure pathway was subdivided into exposure to animals (left side) and exposure to humans (right side). Note: Slaughter = slaughtering; LBS = Live Bird Shop; IPE = Individual Protective Equipment.

Table 1

Features of LBS clusters for pathogen entry risk. Clusters were categorised as lower or higher risk based on the distribution of relevant variables, separately for broiler and deshi LBSs. Categorical variables are reported as no. (%), while numerical are summarized as median (IQR).

	Broiler shops (n = 72)			Desi shops (n = 61)		
	B.1 Lower-risk shops (n = 39)	B.2 Higher-risk shops, mixing of species (n = 17)	B.3 Higher-risk shops, multiple suppliers (n = 16)	D.1 Lower-risk shops (n = 36)	D.2 Higher-risk shops, mixing of species (n = 13)	D.3 Higher-risk shops, multiple suppliers (n = 12)
Supplies (chicken source)						
No. of supplier types						
1 supplier type	39 (100.0)	17 (100.0)	1 (6.2)	36 (100.0)	13 (100.0)	1 (8.3)
> 1 supplier type	0 (0.0)	0 (0.0)	15 (93.8)	0 (0.0)	0 (0.0)	11 (91.7)
Total number of suppliers	1 (1–2)	1 (1–1)	2 (2–3)	1 (1–1)	1 (1–1)	2 (2–2)
Housing characteristics						
High traffic location						
Yes	31 (79.5)	13 (76.5)	15 (93.8)	31 (86.1)	9 (69.2)	10 (83.3)
No	8 (20.5)	4 (23.5)	1 (6.2)	5 (13.9)	4 (30.8)	2 (16.7)
Closeness to other LBSs						
Direct neighbour	2 (5.1)	9 (52.9)	7 (43.8)	5 (13.9)	8 (61.5)	6 (50.0)
< 100 m	18 (46.2)	7 (41.2)	5 (31.2)	17 (47.2)	4 (30.8)	4 (33.3)
No LBS < 100 m	19 (48.7)	1 (5.9)	4 (25.0)	14 (38.9)	1 (7.7)	2 (16.7)
Water supply						
Tap in the shop only	23 (59.0)	11 (64.7)	12 (75.0)	16 (44.4)	10 (76.9)	11 (91.7)
Water source other than shop	16 (41.0)	6 (35.3)	4 (25.0)	20 (55.6)	3 (23.1)	1 (8.3)
Presence of live animals						
Other animal species (excl. poultry) sold						
Yes	2 (5.1)	14 (82.4)	7 (43.8)	0 (0.0)	12 (92.3)	7 (58.3)
No	37 (94.9)	3 (17.6)	9 (56.2)	36 (100.0)	1 (7.7)	5 (41.7)
No. of poultry species / type sold in the LBS						
Only 1 chicken type	18 (46.2)	2 (11.8)	5 (31.2)	9 (25.0)	0 (0.0)	4 (33.3)
More than 1 chicken type/poultry species	21 (53.8)	15 (88.2)	11 (68.8)	27 (75.0)	13 (100.0)	8 (66.7)

market environments contribute to disease risk. These sourcing patterns create opportunities for mixing of birds from various origins, raising concerns about the introduction and spread of pathogens. The greater number of supply sources and the proximity to other LBSs observed in clusters B.3 and D.3 further contribute to their susceptibility to pathogen introduction (Hog et al., 2021; Fournié et al., 2016). For these LBSs, biosecurity efforts should focus on preventing the introduction of pathogens into LBSs, by limiting the mixing of newly arrived birds from different sources, particularly clinically ill birds, and enforcing routine sanitation protocols for cages to limit cross-transmission.

The Pianka and Shannon indices revealed homogeneity in cluster distribution within cities, emphasizing the need for interventions tailored to specific risk profiles of LBSs. LBSs in tribal cities exhibited more uniform profiles than those in non-tribal cities, suggesting that disease control measures tailored to specific LBS types could have broader applicability. For instance, interventions designed for a particular city's LBSs profile may be transferable to other locations with comparable structure or management practices, allowing for more efficient implementation of mitigation strategies. Similar urban-rural contrasts were highlighted in a study in Mali (Molia et al., 2016), where biosecurity practices in the capital city of Bamako differed from other regions, reflecting how infrastructure and market region can shape disease management practices. Our findings underscore the need for a nuanced approach to biosecurity interventions, addressing both regional (tribal versus non-tribal) and operational differences (e.g., deshi versus broiler) based on the identified risk profiles of LBS. As shown for Bangladesh (Rebecca and Moyon, 2019), poultry trading networks differ between chicken types and city supplied making standardized approaches less effective.

The cluster analysis identified key poultry management practices that distinguished higher-risk clusters on pathogen exposure risk (B.5, B.6, D.5 and D.6) from lower-risk ones. These clusters were characterised by practices such as keeping birds roaming in an enclosure and

insufficient segregation of poultry species, both of which are established risk factors for AIV transmission (Sayeed et al., 2017a). Additionally, clusters B.5 and D.5 had the highest proportions of unsold birds and a low turnover rate, increasing exposure risks due to prolonged bird co-mingling (Yu et al., 2014). Despite consistent recommendations in the literature to segregate sick from healthy birds (Islam et al., 2023b; Sayeed et al., 2017a), this study identified a lack of clear separation, emphasizing the need for stricter biosecurity enforcement.

Another concerning pattern observed in higher-risk clusters was inadequate cleaning practices, especially in clusters B.6 and D.6, where cleaning was reported as infrequent or inconsistent. Overall, LBSs selling dressed chickens in tribal cities exhibited worse sanitary conditions than those in non-tribal cities, aligning with expected trends (Chatterjee and Rajkumar, 2015; Conan et al., 2012; DAHDF, 2019b). This lack of routine sanitation considerably increases environmental exposure risks, allowing pathogens to persist in contaminated environments (Van Kerkhove et al., 2011). Regular detergent use has been proven effective in reducing environmental contamination (Chowdhury et al., 2020; Martin et al., 2011; Fournie et al., 2013) and inactivating pathogens that pose both animal and zoonotic risks (Islam et al., 2023b; Sayeed et al., 2017b). While LBSs in B.5 and D.5 clusters reported conducting slaughter near an open drain, which may facilitate cleaning and improve sanitary conditions, this practice can also serve as a source of contamination. Strengthening routine sanitation protocols, particularly in high-risk clusters, could substantially mitigate pathogen exposure risks for both animals and humans.

The research pinpointed a higher-risk profile for pathogen exposure to humans. LBSs in cluster H.3 were more commonly located in tribal cities and received supplies from neighboring and distant regions. These shops were especially vulnerable due to infrequent cleaning of slaughtering and processing areas, which increased the accumulation of waste near open drains (where slaughter is performed) and heightened cross-contamination risks during defeathering (Nidaullah et al., 2017; Cason

Table 2

Features of LBS clusters for chicken exposure. Same legend as Table 1.

	Broiler shops (n = 72)			Desi shops (n = 61)		
	B.4 Lower-risk shops (n = 46)	B.5 Higher-risk shops, high surplus, poultry species mixing (n = 13)	B.6 Higher-risk shops, lower hygiene (n = 13)	D.4 Lower-risk shops (n = 18)	D.5 Higher-risk shops, high surplus, poultry species mixing (n = 33)	D.6 Higher-risk shops, lower hygiene (n = 10)
<i>Housing characteristics</i>						
Poultry mixing						
Yes	22 (47.8)	2 (15.4)	4 (30.8)	6 (33.3)	20 (60.60)	3 (30.0)
No	24 (52.2)	11 (84.6)	9 (69.2)	12 (66.7)	13 (39.4)	7 (70.0)
Poultry holding						
Cage or limited mobility	39 (84.8)	1 (7.7)	8 (61.5)	16 (88.9)	23 (69.7)	4 (40.0)
Free roaming, legs untied	0 (0.0)	10 (76.9)	1 (7.7)	2 (11.1)	3 (9.1)	1 (10.0)
Both	7 (15.2)	2 (15.4)	4 (30.8)	0 (0.0)	7 (21.2)	5 (50.0)
Cages' materials						
Steel or Metal	46 (100.0)	3 (23.1)	4 (30.8)	14 (77.8)	30 (90.9)	4 (40.0)
Plastic or Wood	0 (0.0)	0 (0.0)	3 (23.0)	2 (11.1)	0 (0.0)	2 (20.0)
No cages	0 (0.0)	10 (76.9)	6 (46.2)	2 (11.1)	3 (9.1)	5 (40.0)
Inside						
slaughtering						
Yes	41 (89.1)	12 (92.3)	11 (84.6)	6 (33.3)	33 (100.0)	10 (100.0)
No	5 (10.9)	1 (7.7)	2 (15.4)	12 (66.7)	0 (0.0)	0 (0.0)
Slaughtering close to live birds						
Yes	13 (28.3)	6 (46.2)	2 (15.4)	3 (16.7)	13 (39.4)	1 (10.0)
No	33 (71.7)	7 (53.8)	11 (84.6)	15 (83.3)	20 (60.6)	9 (90.0)
Slaughtering conditions						
Near open drain	37 (80.4)	11 (84.6)	4 (30.8)	1 (5.6)	33 (100.0)	4 (40.0)
Other	9 (19.6)	2 (15.4)	9 (69.2)	17 (94.4)	0 (0.0)	6 (60.0)
<i>Management features</i>						
Average weekly sales	29.5 (20–66.25)	60 (14.5–125)	29 (21–28)	3.3 (2.5–6)	3 (2.5–6.5)	8 (4.3–12.1)
On-hold time (hours)	12 (11–13)	12 (5–12)	12 (12–13.5)	12 (11.3–13.4)	12 (11–13)	12 (6.8–13.5)
Proportion of days with surplus	0.4 (0.3–0.5)	0.5 (0.5–0.5)	0.2 (0.2–0.3)	0.3 (0–0.5)	0.5 (0.4–0.5)	0.2 (0.1–0.3)
Overnight storage						
Shops	45 (97.8)	8 (61.5)	13 (100.0)	15 (83.3)	29 (87.9)	10 (100.0)
Vendors	1 (2.2)	2 (15.4)	0 (0.0)	3 (16.7)	2 (6.1)	0 (0.0)
Shops and Vendors	0 (0.0)	3 (23.1)	0 (0.0)	0 (0.0)	2 (6.1)	0 (0.0)
Mixing with new supplies						
Yes	46 (100.0)	11 (84.6)	7 (53.8)	17 (94.4)	33 (100.0)	5 (50.0)
No	0 (0.0)	2 (15.4)	6 (46.2)	1 (5.6)	0 (0.0)	5 (50.0)
<i>Hygiene characteristics</i>						
Cleanliness						
Large amount of feces	11 (23.9)	0 (0.0)	5 (38.5)	3 (16.7)	4 (12.1)	5 (50.0)
Small amount of feces	35 (76.1)	13 (100.0)	8 (61.5)	15 (83.3)	29 (87.9)	5 (50.0)
Cleaning frequency						
1 time daily	1 (2.2)	1 (7.7)	11 (84.6)	0 (0.0)	0 (0.0)	10 (100.0)
> 1 time daily	45 (97.8)	12 (92.3)	2 (15.4)	18 (100.0)	33 (100.0)	0 (0.0)
Waste management						
Kept away from poultry	33 (71.7)	11 (84.6)	8 (61.5)	14 (77.8)	26 (78.8)	7 (70.0)
Next to live poultry	13 (28.3)	2 (15.4)	5 (38.5)	4 (22.2)	7 (21.2)	4 (30.0)

et al., 2004). Additionally, H.3 LBSs often stored carcasses on non-protected surfaces, heightening human exposure risk, whereas H.1 LBSs demonstrated better practices, avoiding open drains and using protected surfaces. To mitigate these risks, increased surveillance of LBSs in cluster H.3 and tribal cities is recommended, with a focus on monitoring, and addressing, sanitation practices and waste management to prevent potential public health threats. A notable gap across all clusters was the limited adoption of Individual protective equipment

(IPE). Employees in cluster H.1 were more likely to use at least one type of IPE, such as gloves or masks, compared to those in H.3; however, overall adoption remained low. Addressing these gaps requires affordable, accessible and easy-to-use IPE options particularly for poultry workers (Huo et al., 2012). Given the challenges in IPE adoption, environmental controls (e.g., handwashing stations, scalding pots, improved drainage) could be provide practical alternatives.

Despite the novelty of the results, the study has several limitations

Table 3

Features of LBS clusters for human exposure. Same legend as Table 1.

	All shops (n = 86)		
	H.1 Lower-risk shops, with dressed chickens (n = 17)	H.2 Lower-risk shops, no dressed chickens (n = 25)	H.3 Higher-risk shops, dressed chickens and poorer sanitary conditions (n = 44)
Housing characteristics			
Mixing with live birds			
Yes	9 (52.9)	0 (0.0)	15 (34.1)
No	8 (47.1)	25 (100.0)	29 (65.9)
Chicken storage			
Non-protected counter	5 (29.4)	0 (0.0)	25 (56.9)
Protected counter or fridge	8 (47.1)	0 (0.0)	17 (38.6)
Not stored	4 (23.5)	0 (0.0)	2 (4.5)
No dressed chickens	0 (0.0)	25 (100.0)	0 (0.0)
Defeathering machine			
Yes	0 (0.0)	0 (0.0)	5 (11.4)
No	17 (100.0)	25 (100.0)	39 (88.6)
Slaughtering close to live birds			
Yes	1 (5.9)	4 (16.0)	18 (40.9)
No	16 (94.1)	21 (84.0)	26 (59.1)
Inside slaughtering			
Yes	10 (58.8)	19 (76.0)	44 (100.0)
No	7 (41.2)	6 (24.0)	0 (0.0)
Slaughtering conditions			
Near open drain	3 (17.6)	12 (48.0)	44 (100.0)
Other (bucket, outside shop...)	14 (82.4)	13 (52.0)	0 (0.0)
Hygiene			
Use of IPE			
None	7 (41.2)	8 (32.0)	42 (95.5)
>50 %	8 (47.1)	11 (44.0)	0 (0.0)
<=50 %	2 (11.7)	6 (24.0)	2 (4.5)
Waste disposal			
Yes	0 (0.0)	0 (0.0)	5 (11.4)
No	17 (100.0)	25 (100.0)	39 (88.6)
Cleaning frequency (Slaught. area)			
Every hour	6 (35.3)	5 (20.0)	42 (95.5)
1,2 or 3 times daily	11 (64.7)	20 (80.0)	2 (4.5)

that should be acknowledged. Although a random sampling procedure was used (Kumar, 2007; Kondo et al., 2014), the lack of detailed socio-economic data for different neighbourhoods (e.g., income levels, market demand, local preferences for broiler vs. deshi) limited our ability to assess how these factors might have influenced LBS risk profiles. Additionally, we focused on large cities. While our results are generalisable to urban centres in Gujarat, further investigation should explore practices in smaller cities and rural areas as well as other Indian

states and other countries. Moreover, the lack of prior knowledge on pathogen prevalence and diversity made it more difficult to define a formal sample size calculation a priori. Nevertheless, post-hoc power calculations using a chi-square test with three groups, a moderate effect size (Cohen's $w = 0.4$), and a significance level of 0.05 indicate that our sample size approximately achieved 94.5 % power, supporting its adequacy for detecting meaningful differences across clusters. Furthermore, the cross-sectional design did not take for seasonal variations,

Table 4

Average Shannon (S) and Pianka (P) indices for each risk pathway. Shannon index is averaged over all 8 cities, the 4 tribal cities and the 4 non-tribal cities; Pianka index is averaged between all possible pairs of cities among all 8 cities, tribal cities and non-tribal cities; in brackets: median and IQR of simulated values obtained through permutations of cluster membership.

Risk Pathway	Index	All cities	Region	
			Tribal	Non-Tribal
Entry				
Broiler	S	0.56 (0.88 [0.78–0.96])**	0.40	0.71
	P	0.57 (0.86 [0.76–0.94])**	0.71	0.56
Deshi	S	0.43 (0.85 [0.72–0.94])**	0.38	0.49
	P	0.55 (0.86 [0.74–0.94])**	0.95	0.39
Chicken Exposure				
Broiler	S	0.61 (0.88 [0.78–0.95])**	0.70	0.52
	P	0.64 (0.87 [0.77–0.94])**	0.87	0.61
Deshi	S	0.43 (0.85 [0.73–0.94])**	0.35	0.52
	P	0.50 (0.85 [0.73–0.93])**	0.83	0.36
Human exposure				
	S	0.81 (0.91 [0.82–0.97])*	0.77	0.84
	P	0.73 (0.88 [0.79–0.95])*	0.65	0.65

* p-value < 0.05;

** p-value < 0.001.

Table 5

LBS location and characteristics of supply sources across clusters.

			Location LBSs (%)		Supply source (%)		Distance to supply source (%)		
	Risk-level	Cluster	Tribal	Non-tribal	Tribal	Non-tribal	Same-city	Contiguous	Non-contiguous
Disease Entry	Lower-risk	B1	59.0	41.0	61.5	38.5	69.2	26.9	3.8
		D1	66.7	33.3	66.7	33.3	12.1	33.3	54.5
		B2	41.2	58.8	50.0	50.0	28.6	50.0	21.4
	Higher-risk	D2	30.8	69.2	38.9	61.1	6.7	40.0	53.3
		B3	41.2	58.8	16.7	83.3	64.3	28.6	7.1
		D3	8.3	91.7	100	0	8.3	8.3	83.3
Chicken Exposure	Lower-risk	B4	75.0	25.0	83.3	33.3	13.8	31.0	55.2
		D4	38.9	61.1	40.0	60.0	6.7	33.3	60.0
		B5	51.3	48.7	66.7	43.3	61.3	35.5	3.2
	Higher-risk	D5	66.7	33.3	87.9	45.5	12.1	33.3	54.5
		B6	14.3	85.7	25.0	75.0	41.2	41.2	17.6
		D6	0	100	75.0	25.0	8.3	8.3	83.3
Human Exposure	Lower-risk	H1	35.3	64.7	58.3	41.7	8.3	33.3	58.3
		H2	20.0	80.0	46.7	60.0	6.7	33.3	60.0
	Higher-risk	H3	68.2	31.8	89.7	44.8	13.8	31.0	55.2

Table 6

LBSs distribution across the identified higher and lower-risk clusters. The table described the number and percentage (%) of LBSs that were consistently present at high-risk clusters across three, two, one risk-pathway (entry) or none.

Risk category	N (%)
High-risk for all three risk-pathways (Entry, Animal, Human exposure)	14 (25.9)
High-risk for entry and animal exposure	12 (22.2)
High-risk for entry and human exposure	2 (3.7)
High-risk for entry	5 (9.3)
Low-risk across all risk-pathways	15 (27.8)

potentially missing seasonal shifts in practices (Soares Magalhães et al., 2012; Van Kerkhove et al., 2009; Delabougliuse et al., 2017). Finally, the study was conducted during the COVID-19 pandemic. Since disruptions related to the COVID-19 pandemic may have impacted poultry supply chains and consumer demand in Gujarat, primarily due to potential decline in consumer demand and human movement restrictions as highlighted by recent research (Mishra et al., 2023; Sattar et al., 2021; Chapot et al., 2021). Despite its limitations, this study offered unprecedented insights into epidemiologically-relevant characteristics of LBSs in Gujarat, India, and establishes a risk-based framework that is transferable to other settings where LBSs are key for poultry procurement by consumers.

5. Conclusions

The study highlighted the diversity of LBS management practices in Gujarat and identified an LBS typology based on risk pathways related to pathogen entry and exposure for both chickens and humans. While management practices varied across clusters, overarching trends emerged, revealing widespread hygiene deficiencies and differences between cities and tribal and non-tribal regions. These findings underscore the need for context-specific prevention and control strategies that specifically address the detected biosecurity shortcomings and local practices. Future research should apply the developed risk-based framework to other areas in India and other countries and incorporate biological sampling to validate these epidemiological risk profiles and enhance disease control measures in these settings.

CRediT authorship contribution statement

Guillaume Fournié: Writing – review & editing, Visualization, Resources, Project administration, Methodology, Investigation, Conceptualization. **Anne Conan:** Writing – review & editing, Visualization, Supervision, Resources, Project administration, Methodology, Investigation, Funding acquisition, Conceptualization. **Damer Blake:** Writing –

review & editing, Investigation, Funding acquisition. **Fiona Tomley:** Writing – review & editing, Investigation, Funding acquisition. **Lorraine Chapot:** Writing – review & editing, Investigation. **Dirk U. Pfeiffer:** Resources, Project administration, Investigation, Funding acquisition, Conceptualization. **Ketankumar J. Panchal:** Writing – review & editing, Investigation. **Prakash Koringa:** Writing – review & editing, Investigation. **Akash Golaviya:** Writing – review & editing, Investigation. **Sequeira Sara:** Writing – review & editing, Writing – original draft, Visualization, Software, Project administration, Methodology, Investigation, Formal analysis, Data curation. **Haidaruliman Paleja:** Writing – review & editing, Resources, Methodology, Investigation, Conceptualization.

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Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at [doi:10.1016/j.prevetmed.2025.106661](https://doi.org/10.1016/j.prevetmed.2025.106661).

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