The value of a retrospective analysis of slaughter records for the welfare of broiler chickens

Xavier Averós,[*,1] Bernardino Balderas,[†] Enrique Cameno,[*] and Inma Estevez[*,‡,1]

[*]Department of Animal Production, NEIKER-Basque Institute for Agricultural Research and Development, Basque Research and Technology Alliance (BRTA), Arkaute, Spain, 01192; [†]Servicio Andaluz de Salud, Granada, Spain, 18016; and [‡]IKERBASQUE, Basque Foundation for Science, Bilbao, Spain, 48013

ABSTRACT The effects of transport risk factors on deaths on arrival (**DOAs**; %) and carcass rejections (%) on broiler chickens transported to a slaughterhouse in Southern Spain were assessed using information routinely collected at the slaughterhouse. A total of 2,284 flocks and 10,198,663 broiler chickens, transported in 2,103 commercial transports from 217 different farms to a single slaughterhouse, were studied using the information of veterinary service and slaughter records. Studied risk factors were transport condition at arrival score (good/bad) and plumage condition score (good/bad) at arrival at the slaughterhouse, mixing loads (yes/no), transport duration (min), and season of the year (spring/summer/autumn/winter). Generalized linear mixed models were used to determine the effects of risk factors and their interactions on DOA and carcass rejections. The interactive nature of risk factors affecting both variables was revealed. Mixing loads interacted with transport duration ($P < 0.0001$), amplifying the negative consequences of long transports on DOA. Mixing loads also interacted with transport conditions at arrival ($P = 0.0005$), with the impact of bad transport conditions at arrival being particularly negative in the case of mixed loads. These facts raise questions about the suitability of mixing loads both from the animal welfare and economic standpoints. Transport duration interacted with season ($P = 0.0003$), with the detrimental effect of long distances on DOA being particularly evident during summer and winter, which highlights the need for alternative management measures when hot or cold temperatures are foreseen during transport. Long transports also increased carcass rejections ($P < 0.0001$). Mixing loads and bad plumage condition exacerbated the overall, detrimental effect of autumn transports on DOA ($P = 0.0084$ and $P = 0.0009$, respectively) with respect to summer transports. Similar results were obtained for the interactions between mixing loads and transport season ($P = 0.0043$) and transport conditions at arrival and transport season ($P = 0.0014$) on carcass rejections. Overall, results highlight the value of slaughter records to produce information useful to reduce the impact of transport risk factors, improve broiler chicken welfare, and improve slaughterhouse economic results.

Key words: broiler chicken, DOA, mixed load, transport, welfare

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INTRODUCTION

World broiler chicken production was estimated at 2.4×10^{10} birds in 2018 (FAOSTAT, 2020), representing 76% of the total live animals in production. This reflects the importance of poultry for world population. Transport to slaughter is a critical step in the chicken production chain that may have negative consequences on bird welfare if associated management and handling procedures are inadequate (Mitchell and Kettlewell, 2009). As mentioned by Schwartzkopf-Genswein et al. (2012), animal welfare concerns include the potential for animals to experience stress, injury, fatigue, mortality, and morbidity due to transportation aspects such as poor handling and exposure to variable climatic conditions or mixing flocks from different farms. Poor transport conditions have obvious negative effects on bird welfare and will, most likely, translate into increased deaths on arrival (DOAs; Bayliss and Hinton, 1990; Warriss et al., 1992; Jacobs et al., 2017a) and increase carcass rejections (Nijdam et al., 2004). Therefore, knowledge on how to control for risk factors will lead to improved welfare conditions and increased economic profit for all actors in the broiler chicken production chain.

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[1]Corresponding authors: xaveros@neiker.eus (XA); iestevez@neiker.eus (IE)

DOA and carcass rejections may be the result of poor transport conditions but may also originate during the pretransport history of broiler chickens (Haslam et al., 2008). A clear relationship exists between on-farm welfare conditions and slaughter problems, shown by the correlation between on-farm mortality and DOA that different studies have described (Haslam et al., 2008; Chauvin et al., 2011; Whiting et al., 2007; BenSassi et al., 2019). Therefore, assessment at slaughter may offer valuable information about prior on-farm welfare conditions (Grilli et al., 2015). Broiler chickens' on-farm health and welfare depends, among others, on bedding conditions and more specifically on bedding water content (Dawkins et al., 2004). Wet litter will result in plumage dirtiness, which is highly correlated with DOA and carcass rejections at slaughter houses (De Jong et al., 2014; Jacobs et al., 2017b; BenSassi et al., 2019). Given that plumage dirtiness is also highly correlated with plumage condition at the end of transport (Jacobs et al., 2017b), the latter visual indicator can be proposed as a reliable estimator of the broiler chicken pretransport welfare status.

Regarding transport conditions, duration is a well-known welfare risk factor in all production species. Long transports are stressful for broiler chickens (Zhang et al., 2009) and may lead to increased DOA (Bayliss and Hinton, 1990; Warriss et al., 1992; Vecerek et al., 2006; Whiting et al., 2007; Chauvin et al. 2011; Vieira et al., 2011; Caffrey et al., 2017). Long transport durations will also exacerbate the negative consequences that poor transport departing conditions, such as those caused by inadequate bird catching and loading at the farm, have on DOA (Jacobs et al., 2017b). Climatic conditions during transport are also critical for broiler chickens, as both high temperatures, particularly in combination with high relative humidity, and low temperatures will result in thermal stress and lead to increased DOA and carcass bruising (Nijdam et al., 2004; Schwartzkopf-Genswein et al., 2012). The detrimental consequences of thermal conditions above or below thermoneutrality will additionally be amplified during long transports (Nijdam et al., 2004). Beyond temperature, other adverse weather conditions affecting DOA are wind and rain (Chauvin et al., 2011). In consequence, season of the year, through a combined action of previously mentioned climatic variables, will have a large influence on the welfare of birds as they are transported to slaughter. Different studies have described higher DOA during summer and winter (Petracci et al., 2006; Vecerek et al., 2006; Haslam et al., 2008), although no consensus exists regarding the influence of low temperatures (Warriss et al., 2005), and the same has also been described for carcass bruising (Nijdam et al., 2004). Bird dirtiness differs according to season (Wilkins et al., 2003) and is also associated to weather conditions during transport, with rain particularly (Jacobs et al., 2017b).

Mixing loads is a relatively common practice during commercial transportation to slaughter houses in Spain, that consists in loading animals from different farms into the same truck. This is done to optimize truck occupation and transport costs but may be negative from a health and welfare perspective. In slaughter pigs, mixing loads has been found to increase the percentage of carcass problems and to exacerbate the negative impact of longer distances on mortality at the end of transport (Gosálvez et al., 2006). A similar effect might be expected during broiler chicken transport to slaughter, but no information exists up to date on the impact of mixing loads and how this interacts with other risk factors.

All the aforementioned information is routinely collected during veterinary inspections at the end of transport or can be easily extracted from slaughterhouse records. These data are often overlooked and not sufficiently exploited by industry. Therefore, a retrospective analysis of risk factors contained in slaughter records may be helpful to reveal relationships between them and how they may impact the welfare of broilers assessed on the basis of DOA and carcass rejections. This will result in relying on valuable information to base technical decisions with benefits for both broiler chicken welfare and the economic return of the production chain, ultimately providing these data with an extra added value.

The aim of this study was to determine the influence of transport condition and plumage condition scores at arrival at the slaughterhouse, mixing loads, transport duration, and season of the year on DOA and carcass rejections in broiler chickens transported to a slaughterhouse in Southern Spain, based on the information routinely collected by the official veterinarians at the slaughterhouse.

MATERIALS AND METHODS

Information on 2,103 commercial transports, carrying 2,284 flocks and 10,198,663 broiler chickens from 217 different farms to a single slaughterhouse located in Southern Spain, was collected by the slaughterhouse official veterinary inspection service as part of their routine job. Transports were all carried out during 2017, following the requirements of the Council Regulation (EC) No 1/2005 relative to the protection of animals during transport. At the farm, the loading procedure was similar for all transports. Inside the farm, broilers were caught and introduced into plastic crates. Once complete, plastic crates were placed in metal modules, which were loaded onto the truck. Truck capacity was 10 metal modules and 18 plastic crates/modules. Upper protective canvas toppers were used in all transports, while hauliers were asked to use side protective canvas toppers in case of cold and/or rain. Upon arrival at the slaughterhouse, broiler chickens waited for 4 h on average before slaughter.

Slaughter records were collected manually and recorded on paper. For the purpose of this study, records were transferred to a database. Each transport was individually identified, and farms were given a unique code to be able to identify different transports from a single farm within the year. In case collected information for a specific transport was unclear, missing or erroneous, or reported unusually high DOA and/or carcass rejection values attributable to reasons other than transport,

Table 1. Number of flocks that were finally included in the dead on arrival (DOA) and carcass rejections models, segmented according to each level of each studied risk factor.

Variable	DOA (n = 2,154 flocks)	Carcass rejections (n = 1,649 flocks)
Mixed loads		
Not mixed	1,987	1,514
Mixed	167	135
Transport conditions at arrival		
Good	1,846	1,399
Bad	308	250
Season		
Spring	542	357
Summer	548	471
Autumn	572	564
Winter	492	257
Plumage condition at arrival		
Good	469	396
Bad	1,685	1,253

it was discarded. After filtering information, 2,154 and 1,649 flocks were finally available for further analysis and development of DOA and carcass rejections models, respectively. For each transported flock, information on transport duration (minutes between the first chicken was loaded onto the truck until the last chicken was unloaded at the slaughterhouse) and transport season (spring, summer, autumn, winter) was collected. Mean temperature and relative humidity during 2017 in the transport area were 18.9°C and 42.4% in spring, 24.1°C and 40.3% in summer, 11.0°C and 63.3% in autumn, and 8.5°C and 65.7% in winter. Mixed loads were identified by a 2-level variable according to whether the truck transported 1 or 2 flocks. The transport conditions of each flock were visually scored, from outside the truck, by official veterinarians at arrival at the slaughterhouse, using a 2-level scale (Good/Bad) that depended on whether they were able to detect the next visual indicators: trapped birds, signs of hyperthermia (i.e., birds with their head out of the plastic crate and panting) or hypothermia (i.e., birds grouped in the center of the plastic crate to protect from cold), signs of overcrowding, and/or lack of proper trailer canvas toppers to protect birds from adverse climatic conditions. Visual inspection included all transport modules and plastic crates. For a visual indicator to be declared as detected, it had to be observed on at least 50% of inspected birds. Transport condition was scored as bad if at least one of the visual indicators of bad transport conditions was detected. Similarly, plumage condition of each flock was also visually assessed at arrival at the slaughterhouse using a 2-level scale (Good/Bad) according to whether birds presented or not yellowish plumage coloration attributable to wet bedding at the farm. At least 50% of inspected birds had to have a bad plumage condition for the plumage condition of the flock to be declared as bad. If a truck was composed of mixed loads, each flock was scored separately. For each flock, the number of broiler chickens loaded onto the truck, birds that are slaughtered, and rejected carcasses at slaughter were recorded. Deaths on arrival (DOA; n) for each flock were the number of chickens found dead at unloading. Carcass rejections (n) relate to those removed from the production line after slaughter because of emaciation, ascites/edema, septicemia, cellulitis, lesions, trauma, and/or hematomas among others, as established by the Spanish Agency for Food Safety and Nutrition (AECOSAN, 2015).

Statistical analysis of data was carried out with the SAS statistical software (SAS 9.4). To determine whether flock transport condition and plumage condition scores at arrival were affected by transport duration, season, and mixed loads, both variables were treated as binary response variables. With this, the effects of transport duration, season, and mixed loads were modelled using logistic regressions by means of the GLIMMIX procedure, which were obtained separately for flocks contributing to DOA and carcass rejections data sets. The effects of the described risk factors on the welfare of broiler chickens, assessed through DOA and carcass rejections, were evaluated. For this, separate models were developed for DOA and carcass

Table 2. Transport duration, dead on arrival (DOA), and carcass rejections summary statistics for the DOA and carcass rejections models.

Variable	Mean	Median	SD	SE
DOA model (n = 2,154 flocks)				
DOA (%)	0.26	0.16	0.01	0.68
Transport duration for flocks included in the DOA model (min)	178	105.01	211.95	2.26
Carcass rejections model (n = 1,649 flocks)				
Carcass rejections (%)	0.77	0.50	0.04	1.67
Transport duration for flocks included in the carcass rejections model (min)	165	99.65	202.84	2.45

Table 3. Odds ratio estimates of the effects of mixed loads, transport duration, and season on the transport conditions and plumage condition scores at arrival to the slaughterhouse of broiler flocks.[1]

Variable	Odds ratio estimate	95% Confidence limits		P value
		Lower limit	Upper limit	
Dead on arrival (DOA) data set (n = 2,154 flocks)				
Transport conditions at arrival[2]				
Mixed loads				
'Unmixed vs mixed loads'	1.305	0.729	2.338	0.370
Transport duration	1.001	0.999	1.002	0.254
Season				
'Winter vs summer'	0.032	0.012	0.089	<0.001
'Autumn vs summer'	0.201	0.130	0.312	
'Spring vs summer'	1.686	1.267	2.245	
Plumage condition at arrival[3]				
Mixed loads				
'Unmixed vs mixed loads'	1.429	0.977	2.088	0.065
Transport duration	1.001	1.000	1.002	0.063
Season				
'Winter vs summer'	1.370	1.012	1.854	<0.001
'Autumn vs summer'	0.674	0.516	0.881	
'Spring vs summer'	2.551	1.820	3.577	
Carcass rejections data set (n = 1.649 flocks)				
Transport conditions at arrival[2]				
Mixed loads				
'Unmixed vs mixed loads'	1.422	0.757	2.671	0.273
Transport duration	1.001	0.999	1.003	0.198
Season				
'Winter vs summer'	0.029	0.007	0.120	<0.001
'Autumn vs summer'	0.177	0.112	0.280	
'Spring vs summer'	1.934	1.404	2.665	
Plumage condition at arrival[3]				
Mixed loads				
'Unmixed vs mixed loads'	1.412	0.927	2.152	0.108
Transport duration	1.002	1	1.003	0.011
Season				
'Winter vs summer'	1.181	0.817	1.707	<0.001
'Autumn vs summer'	0.685	0.519	0.905	
'Spring vs summer'	3.076	2.041	4.637	

[1]Models are calculated separately for DOA and carcass rejection data sets.
[2]It was modelled the probability of finding a flock with bad transport conditions at arrival.
[3]It was modelled the probability of finding a flock with bad plumage condition at arrival.

rejections using generalized linear mixed models analysis with the GLIMMIX procedure. In both models, flock was considered the experimental unit. DOA and carcass rejections were modelled as count data, which have traditionally been modelled with a Poisson distribution. To avoid data overdispersion generally associated with the Poisson distribution, data were modelled using a negative binomial distribution, which usually provides better estimation models for this type of data (Little et al., 2006) and has been previously applied in similar studies with successful results (Chauvin et al., 2011). The number of loaded and slaughtered broiler chickens were, respectively, offset variables for DOA and carcass rejections. Therefore, DOA and carcass rejections were expressed as percentages.

Main fixed effects were the same for both models and included transport duration, season of transport, whether transport was a mixed load, the transport condition score, and the plumage condition score at arrival. Initial models included all possible two-way interactions between main fixed effects. Final models were developed using a stepwise backward procedure, in which nonsignificant interactions were gradually removed from the model using the highest P value as the removal criterion. Final models retained all significant two-way interactions ($P < 0.05$). Both models included an intercept with random variation according to the specific journey to the slaughterhouse. In addition, the DOA model accounted for unequal random transport variance according to season, while the carcass rejections model accounted for unequal random transport variance according to plumage condition. Least square means were computed in case of statistically significant effects ($P < 0.05$), with P values adjusted for multiple comparisons by Tukey range tests. For significant interactions, tests of simple effects (Winer, 1971) were performed to detect differences between the levels of a risk factor within each level of the other risk factor.

RESULTS

Table 1 shows, for the 2,154 and 1,649 flocks finally included in the respective DOA and carcass rejections models, how many flocks contributed to each level of each studied risk factor. This information offers an idea of the amount of information available during the data-modelling step. Flocks transported during mixed transports represented 7.8% of total flocks included in the DOA model and 8.2% of flocks included in the carcass rejection model. Transport conditions of 14.3% and 15.2% flocks were scored as "bad" at arrival in the

Table 4. Parameter estimates and standard error of the estimate for risk factors included in the dead on arrival (DOA; %) model.

Variable	DOA (%)		
	Estimate	SE	P value[1]
Intercept	−5.7574	0.3388	<0.0001
Mixed loads			
Not mixed	−1.3892	0.3366	<0.0001
Mixed	0		
Transport conditions at arrival			
Good	−1.0520	0.2849	0.0006
Bad	0		
Season			
Winter	0.2897	0.2269	<0.0001
Autumn	1.2386	0.2592	
Spring	0.2868	0.2998	
Summer	0		
Plumage condition at arrival			
Good	0.1068	0.0826	0.3884
Bad	0		
Duration	0.0008	0.0007	<0.0001
Duration × Mixed loads			
Not mixed	0.0035	0.0006	<0.0001
Mixed	0		
Duration × Season			
Winter	0.0007	0.0004	0.0003
Autumn	−0.0012	0.0004	
Spring	−0.0004	0.0005	
Summer	0		
Mixed loads × Transport conditions at arrival			
Not mixed × Good	1.0508	0.2896	0.0005
Not mixed × Bad	0		
Mixed × Good	0		
Mixed × Bad	0		
Mixed loads × Season			
Not mixed × Winter	−0.5879	0.2048	0.0084
Not mixed × Autumn	−0.8011	0.2347	
Not mixed × Spring	−0.5009	0.2837	
Not mixed × Summer	0		
Mixed × Winter	0		
Mixed × Autumn	0		
Mixed × Spring	0		
Mixed × Summer	0		
Season × Plumage condition at arrival			
Winter × Good	0.1888	0.1119	0.0009
Winter × Bad	0		
Autumn × Good	−0.1866	0.1053	
Autumn × Bad	0		
Spring × Good	−0.2776	0.1421	
Spring × Bad	0		
Summer × Good	0		
Summer × Bad	0		

[1]*P* value corresponds to the statistical significance of the mai risk factor or interaction.

respective DOA and carcass models. On the other hand, flocks were balanced across the different seasons for DOA (22.8, 26.6, 25.2, and 25.4% during winter, autumn, spring, and summer, respectively) and were slightly more unbalanced for carcass rejections (15.6, 34.2, 21.6, and 28.6% during winter, autumn, spring, and summer, respectively). Plumage condition scored as bad at the slaughterhouse in most of flocks contributing to DOA and carcass rejections models (77.2 and 76.0%, respectively).

Summary statistics for the continuous risk factor transport duration (min), the response variables DOA (%), and carcass rejections (%) are shown in Table 2, distinguishing between DOA and carcass rejection models. Mean transport duration was about 200 min for flocks included in the DOA and carcass rejections models. Values for the rest of transport duration descriptive statistics were also equivalent between models.

The odds ratio estimates of the effects of mixed loads, transport duration, and season on transport condition and plumage condition scores at arrival, obtained separately for the DOA and carcass rejections' data sets, are shown in Table 3. A highly significant seasonal effect was observed in all cases. The risk of finding a flock with bad transport conditions at arrival was much lower during winter and autumn than during summer but was moderately higher during spring than summer, and this effect was found in both DOA and carcass rejections' data sets. On the other hand, the risk of finding a flock with bad plumage condition at arrival was moderately higher during winter and spring than during summer and moderately higher during summer than during

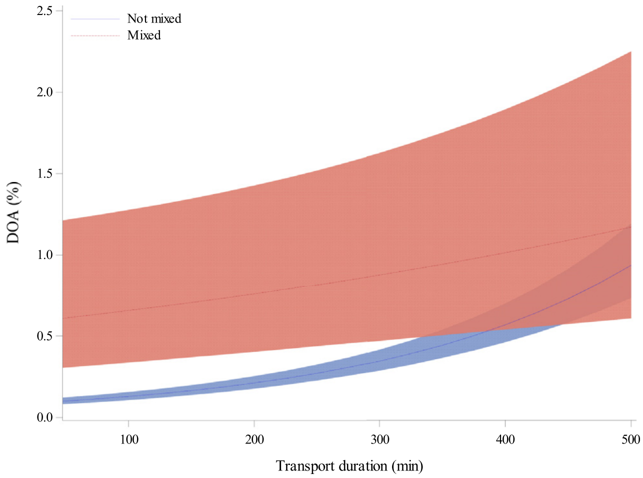

Figure 1. Interaction between the effect of transport duration and of mixing loads on the DOA (%) of 2,154 broiler chicken flocks transported to slaughter in Southern Spain during 2017. Figure shows mean predicted DOA values for not mixed transports (solid line) and for mixed transports (dashed line), as well as 95% confidence intervals (semi-transparent bands).

winter. This also occurred for both DOA and carcass rejections data sets. In addition, a slight but significant positive association between transport duration and the risk of finding a flock with bad plumage condition at arrival was found in the carcass rejection data set.

The DOA model and results are shown in Table 4. Figure 1 illustrates the interaction of transport duration and mixed loads on DOA and reflects that increasing transport duration generally resulted in increased DOA, although values were higher in mixed loads, particularly during shorter transports. The results of the interaction between transport duration and season are presented in Figure 2, showing that the detrimental effects of transport duration were particularly evident during winter transports. The interaction between mixing loads and bad transport conditions at arrival resulted in a significantly higher DOA (Table 5). DOA also differed according to season and whether transports were mixed or not (Table 6). Differences corresponding to the interaction between transport season and plumage condition score at arrival on DOA are also shown in Table 6.

The carcass rejection model and the statistical significance of included risk factors and interactions are shown in Table 7. Figure 3 illustrates the effect of transport duration on carcass rejection. The interaction between transport season and mixed loads shows that mixed loads in autumn and spring resulted in a higher percentage of carcass rejection than summer transports (Table 8). The interaction between transport season and transport conditions at arrival score on carcass rejections (Table 8) shows that a high percentage of carcass rejections were detected in winter and spring in flocks scored with good transport conditions at arrival, while for flocks with bad transport conditions at arrival, this percentage was higher during autumn and spring.

DISCUSSION

Data on 1-year records of broiler chicken transports to one slaughterhouse in Southern Spain were collected, and with this, the interactive nature of risk factors affecting the welfare of broiler chickens was determined.

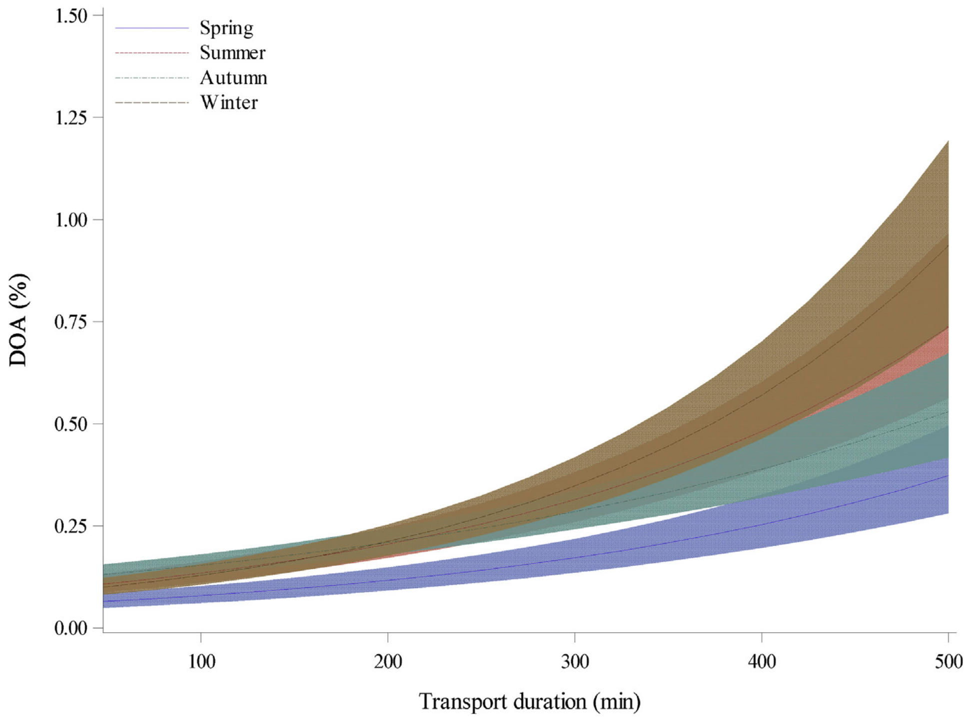

Figure 2. Interaction between transport duration and season of the year on DOA (%) of 2,154 broiler chicken flocks transported to slaughter in Southern Spain during 2017. Figure shows mean predicted DOA values for spring (solid line), summer (short-dashed line), autumn (dash-dotted line), and winter (long-dashed line), as we as 95% confidence intervals (semi-transparent bands).

Table 5. Interaction between the transport conditions score at arrival and mixed loads on the dead on arrival (DOA; %) of broilers transported to slaughter.[1]

Variable	DOA (%)		P value
	Good transport conditions at arrival	Bad transport conditions at arrival	
Not mixed	0.186 ± 0.005	0.186 ± 0.010	0.0005
Mixed	0.199 ± 0.019[b]	0.569 ± 0.147[a]	

[1]Within each line (i.e., for not mixed and mixed transports independently), different letters indicate the existence of significant differences between good and bad transport conditions at arrival ($P < 0.05$). P value refers to the overall interaction effect.

Mixing loads amplified the negative consequences of long transports and bad transport conditions at arrival on DOA. The detrimental effect of long transports on DOA was remarkable during summer and winter, and long transports also increased carcass rejections. Seasonal differences were particularly remarkable when transport conditions were suboptimal (mixing loads, transport conditions, and plumage condition scored as bad at arrival at the slaughterhouse) and pointed out to a negative influence of autumn transports with respect to other seasons.

The number of flocks and transports finally used in the analysis differed between DOA and carcass rejection models. This is due to the filtering process that resulted in information availability being different between both models. Literature shows a large degree of variability regarding DOA, reporting either higher (Warriss et al., 1992, 2005; Haslam et al., 2008; Chauvin et al., 2011), similar (Vecerek et al., 2006), or lower (Petracci et al., 2006; Vieira et al., 2011; Caffrey et al., 2017; Jacobs et al., 2017a; Whiting et al., 2007) DOA values than the ones obtained in this study. Many risk factors can simultaneously affect broiler chickens during transport (Schwartzkopf-Genswein et al., 2012), which ultimately determines the resulting DOA for one specific transport. Thus, resulting DOA is likely to reflect differences according to such risks. Mean DOA values in our study (Table 2) were mostly below 0.3%, except for mixed flocks during autumn and winter. The threshold set by the Spanish Agency for Food Safety and Nutrition (AECOSAN, 2015) is 2%. The Welfare certification scheme sets a minimum target value to be achieved of 0.5% (Welfair Certification, 2020), which is the same standard of acceptable threshold established by the National Chicken Council (2017) in the United States. In any case, values indicate that there is still room for improvement if risk factors are better determined and controlled. References reporting carcass rejections in broiler chickens are much scarcer, but studies in the UK (Haslam et al., 2008) and Canada (Herenda and Jakel, 1994) report higher values of 1.11% and 1.48%, respectively, which are substantially higher than those of the present study. These values are well below the acceptable 5% threshold set by the Spanish authorities when carcass rejection causes are assessed together (AECOSAN, 2015).

Our results confirm the interactive nature of transport risk factors affecting broiler chicken welfare (Mitchell and Kettlewell, 2009; Schwartzkopf-Genswein et al., 2012) and, in particular, the impact of transport duration (e.g., Warriss et al., 1992; Vecerek et al., 2006; Chauvin et al., 2011; Caffrey et al., 2017). Our results reveal the important impact that mixing loads has on DOA and carcass rejections, effects that are amplified for mixed transports. No studies have to date evaluated the impact of mixing loads on broiler chickens. This is a relatively common commercial practice in Spain that intends to optimize transport costs and is also common in other species (Gosálvez et al., 2006). Results, however, suggest that mixing has a negative impact on broiler chickens' welfare and ultimately leads to increased DOA and carcass rejections, similar to the results already described for pigs (Gosálvez et al., 2006). Indeed, mixing loads will increase DOA even during short transports (Figure 1), which highlights the negative consequences of this practice. All these negative consequences raise concerns about the suitability of mixing loads from both the welfare and economic perspectives.

Increased transport duration also resulted in a higher percentage of carcass rejections (Figure 3), although in this case, no interaction was detected with other risk

Table 6. Interaction between the season of the year and mixed loads and between season of the year and plumage condition at arrival on the DOA (%) of broilers transported to slaughter.[1]

Variable	DOA (%)				P value
	Spring	Summer	Autumn	Winter	
Mixed loads					
Not mixed	0.133 ± 0.008[c]	0.205 ± 0.010[a,b]	0.226 ± 0.010[a]	0.194 ± 0.010[b]	0.0084
Mixed	0.247 ± 0.054[y,z]	0.231 ± 0.046[z]	0.568 ± 0.118[x]	0.393 ± 0.073[xy]	
Plumage condition at arrival					
Good condition at arrival	0.166 ± 0.025[c]	0.229 ± 0.028[b,c]	0.344 ± 0.039[a]	0.320 ± 0.036[ab]	0.0009
Bad condition at arrival	0.197 ± 0.021[y]	0.206 ± 0.021[y]	0.373 ± 0.040[x]	0.238 ± 0.023[y]	

[1]Within each line (i.e., for not mixed and mixed transports independently in the case of mixed loads, and for good and bad condition at arrival independently in the case of plumage condition) different letters indicate the existence of significant differences between transport seasons ($P < 0.05$). P value refers to the overall interaction effect.

Table 7. Parameter estimates and standard error of the estimate for risk factors included in the carcass rejections (%) model.

Variable	Carcass rejections (%)		
	Estimate	SE	P value[1]
Intercept	−5.7905	0.1601	<0.0001
Mixed loads			
Not mixed	0.0815	0.1410	<0.0001
Mixed	0		
Transport conditions at arrival			
Good	0.3367	0.0820	0.0982
Bad	0		
Season			
Winter	0.1329	0.5443	0.0004
Autumn	1.2026	0.2497	
Spring	0.9302	0.2490	
Summer	0		
Plumage condition at arrival			
Good	−0.0715	0.0418	0.0875
Bad	0		
Duration			
Mixed loads × Season	0.0012	0.0002	<0.0001
Not mixed × Winter	−0.4414	0.1963	0.0043
Not mixed × Autumn	−0.7471	0.2025	
Not mixed × Spring	−0.6540	0.2384	
Not mixed × Summer	0		
Mixed × Winter	0		
Mixed × Autumn	0		
Mixed × Spring	0		
Mixed × Summer	0		
Transport conditions at arrival × Season			
Good × Winter	0.4753	0.5125	0.0014
Good × Autumn	−0.6489	0.1610	
Good × Spring	−0.2634	0.1134	
Good × Summer	0		
Bad × Winter	0		
Bad × Autumn	0		
Bad × Spring	0		
Bad × Summer	0		

[1] P value corresponds to the statistical significance of the main risk factor or interaction.

factors. To our knowledge, no other study has dealt with the effects of transport duration on carcass rejection in broiler chickens, although Caffrey et al. (2017) suggested that longer transports might be particularly problematic for birds suffering some disease or with injuries caused during catching and loading. Long journeys are more likely to result in increased occurrence of lesions, trauma, and hematomas, which are direct causes of carcass rejection, what would explain our finding.

Considering the effect of season alone, DOA was higher during autumn, followed by winter, spring, and summer, thus matching the results obtained by Caffrey et al. (2017). Nevertheless, DOA was particularly high during winter and summer as compared to spring for long transports. This result evidences the negative impact of more extreme outdoor temperatures on broiler chickens when they are transported to slaughter over long distances. Some disagreement exists in literature regarding the effect of season on DOA, so that Vecerek et al. (2006) described higher DOA during both summer and winter months, but Warriss et al. (2005), Petracci et al. (2006), and Haslam et al. (2008) reported increased DOA during summer months as compared to the rest of seasons, but of course this will depend on the location of the study and weather patterns of the year of the study. On the other hand, when including external temperature in their models, Caffrey et al. (2017) found higher DOA as temperatures were colder; with this inclusion, these authors found that DOA were higher in autumn than in spring. It should be however noticed that the mentioned study was carried out in Canada. Fast-growing broiler chickens may experience reduced thermotolerance caused by intensive genetic selection (Sandercock et al., 1995; 2006). Therefore, both high and low outdoor temperatures during long transports are detrimental for broiler chicken DOA, due to hyperthermia or hypothermia. Our results confirm that, when climatic conditions go beyond the chicken thermoneutral range such as the cases of summer and winter transports, particular attention should be paid to longer transports as they will incur in a higher risk of DOA. Alternative management strategies should be taken in these cases to maintain transport conditions as close as possible to bird thermoneutrality. This would result in a reduction of welfare problems originated by thermal stress and reduce DOA.

As expected, transports that were scored with bad conditions at arrival (chickens trapped in cages, showing signs of hyperthermia or hypothermia, overcrowded, and/or lacking a proper trailer canvas topper) resulted

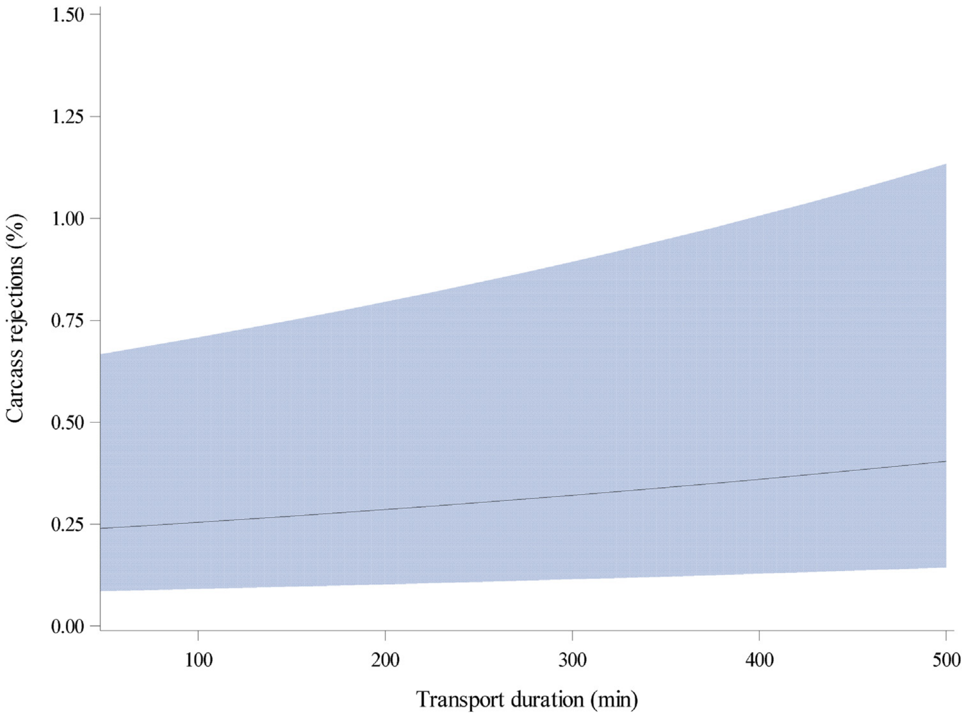

Figure 3. Effect of transport duration on carcass rejections (%) of 1,649 flocks broiler chicken flocks transported to slaughter in Southern Spain during 2017. Figure shows mean predicted values (solid line) and 95% confidence intervals (semi-transparent bands).

in higher DOA (Table 4). Hyperthermia and hypothermia have already been discussed, and regarding these, the absence of a proper trailer canvas topper will expose birds to adverse weather. Trapped birds and cage overcrowding are the result of inadequate catching, crating, and loading practices, with consequences over subsequent transport and slaughter, and even leading to elevated DOA (Nijdam et al., 2005). Given the link between bad transport conditions scored at arrival and increased DOA, the use of these scores to identify problems and trigger targeted, corrective measures during future transports appears advisable to reduce DOA, and lead to improved welfare and higher economic turnover. These results also confirm that the impact of poor transport conditions scored at arrival will be further aggravated when loads are mixed (Table 5), suggesting a synergistic interaction between them, and the need to avoid them as much as possible through a careful planning of transport logistics. Mixing loads will exacerbate the detrimental effect of autumn transports on DOA and carcass rejections with respect to summer transports (see tables 6 and 8). This may appear surprising but has already been described for slaughter pigs (Guardia et al., 1996) and has been attributed to the fact that companies try to adapt their transport logistics when weather conditions are expected to be extreme, such as the case of summer conditions. With the exception of longer transports that are in any case problematic, this logistics adaptation may explain the overall good results found during summer with respect to autumn.

Bad plumage condition score at arrival, attributable to poor bedding condition (de Jong et al., 2014), also tended to increase the percentage of carcass rejections. Wet bedding, that depends among others on the litter material, relates to the development of pathogenic microorganisms and disease in broiler chickens' farms (Dunlop et al., 2016) and on-farm disease is, in its turn, strongly associated to increased carcass rejection at slaughter (Haslam et al., 2008). Birds with bad

Table 8. Interaction between the season of the year and mixing loads on carcass rejections at slaughter (%).[1]

Variable	Carcass rejections (%)				P value
	Spring	Summer	Autumn	Winter	
Mixed loads					
Not mixed	0.553 ± 0.024	0.479 ± 0.021	0.546 ± 0.038	0.446 ± 0.113	0.0043
Mixed	0.980 ± 0.185[a]	0.441 ± 0.062[b]	1.062 ± 0.167[a]	0.639 ± 0.181[ab]	
Transport conditions at arrival					
Good transport conditions at arrival	0.764 ± 0.077[a]	0.544 ± 0.03[b]	0.651 ± 0.048[ab]	0.801 ± 0.056[a]	0.0014
Bad transport conditions at arrival	0.710 ± 0.078[x]	0.388 ± 0.039[y]	0.890 ± 0.135[x]	0.356 ± 0.181[xy]	

[1]Within each line (i.e., for not mixed and mixed transports independently in the case of mixed loads, and for good and bad transport conditions at arrival independently in the case of transport conditions at arrival) different letters indicate the existence of significant differences between transport seasons ($P < 0.05$). P value refers to the overall interaction effect.

plumage condition may be those more sensitive to the action of additional stressors and therefore those with a higher probability of dying during autumn transports, as reflected by DOA concentrating during autumn with respect to other seasons (Table 6). The interaction between transport conditions score at arrival and transport season on carcass rejections does not offer clear results (Table 8) but also points toward a modulation of the seasonal effect of transport according to journey conditions.

In conclusion, this study confirms the relevance of transport risk factors on the DOA and carcass rejections and their interactive nature. The detrimental impact of mixing loads on DOA and carcass rejections makes this practice questionable both in animal welfare and economic terms. Mixing loads amplify the negative consequences of other risk factors such as long journey duration and bad transport conditions scored at arrival, the latter being originated during the loading process and/or transport. The detrimental effect of long journeys on DOA was particularly evident during summer and winter, when more extreme outdoor temperatures were more probable. This highlights the need for alternative management practices during transport during more extreme weather conditions, to maintain birds as close as possible to thermoneutrality and prevent their negative impact over welfare and economic returns. Mixing loads, as a stressful procedure, and bad plumage condition scores, reflecting suboptimal farm departing conditions, would exacerbate the overall, detrimental effect that autumn transports have on broiler chickens with respect to summer transports.

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