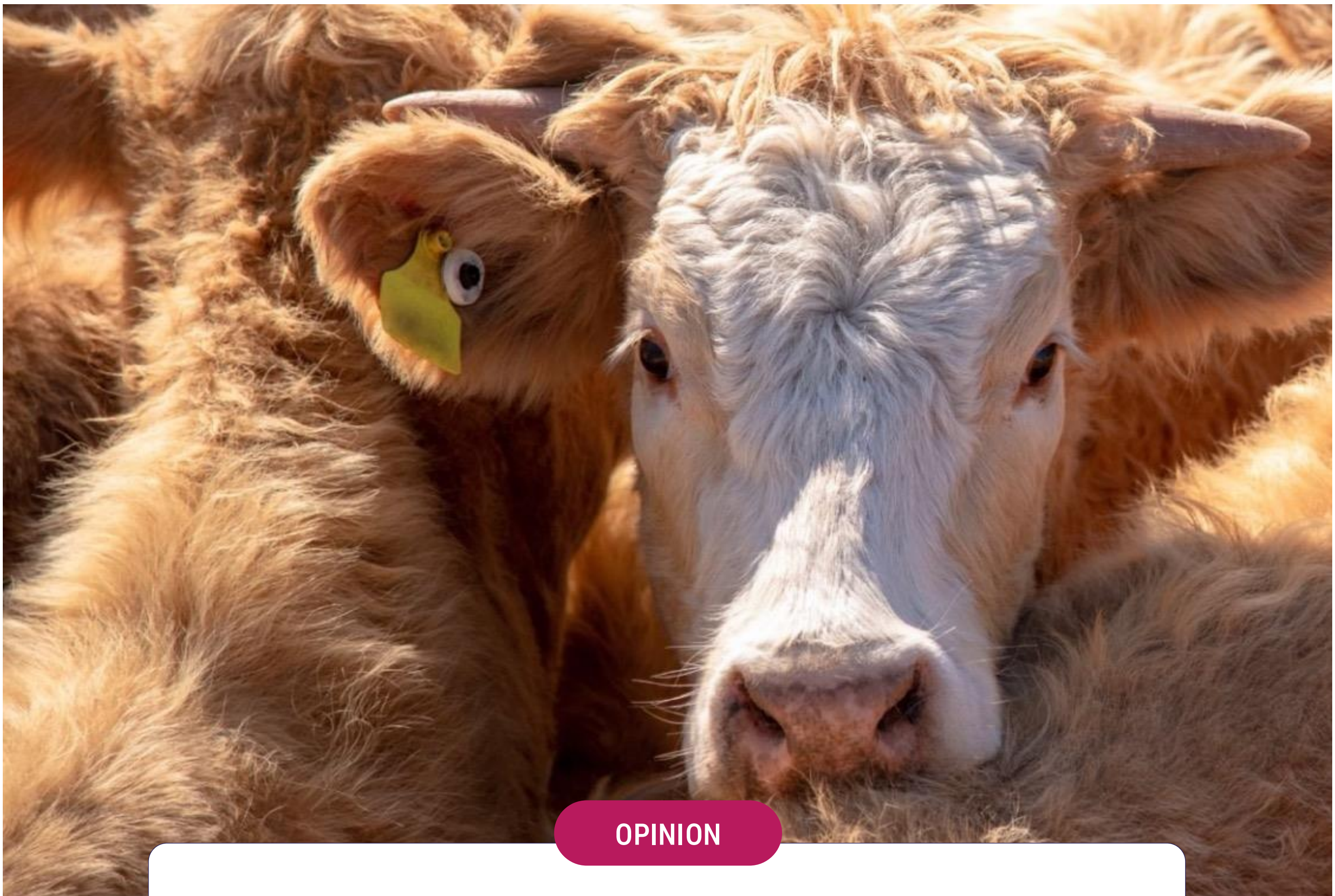
French Reference Centre
for Animal Welfare



OPINION

Impacts of loading density on the risk of falls and injuries in cattle during transport

Original title in French

Impacts de la densité de chargement
sur le risque de chutes et de blessures des bovins
pendant le transport

DECEMBER 2025

Impacts of loading density on the risk of falls and injuries in cattle during transport

Requested by
Animal Welfare Office (BBEA) of the General Directorate for Food (DGAL), French Ministry of Agriculture and Food Sovereignty

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Summary

In 2023, a proposal for a regulation on the protection of animals during transport repealing Council Regulation (EC) No 1/2005 was published. The new text proposes a revision of the rules on space allowances based on the recommendations of the EFSA opinion on the welfare of cattle during transport (EFSA, 2022). The present report from the French Reference Centre for Animal Welfare (FRCAW) summarises the key points of this opinion (EFSA, 2022), focusing exclusively on information relating to falls and injuries associated with loading density during transport. It also provides an in-depth analysis of the literature on the subject, with regard in particular to the behavioural and physiological consequences of different loading densities/space allowances, and to the haematomas observed on carcasses. The report also highlights factors that increase the likelihood of falls and injuries to cattle during transport, and suggests ways to reduce the occurrence of such incidents. Although few existing studies have examined cattle falls and injuries at the space allowances for transport set out in the regulatory proposal, the literature supports the hypothesis that the proposed allowances would reduce falls and injuries during transport. Further research is nevertheless needed to confirm these results, particularly for adult cattle, taking into account the potential aggravating factors discussed in the present report.

Keywords

Transport / Loading density / Falls / Injuries / Cattle

> VIEW ALL WORK CARRIED OUT BY THE FRCAW

Context

The European regulations on farm animal welfare are currently under review. In December 2023, the European Commission published its Proposal for a Regulation of the European Parliament and of the Council on the protection of animals during transport and related operations, amending Council Regulation (EC) No 1255/97 and repealing Council Regulation (EC) No 1/2005, currently in force. The revision was initiated in order to bring the regulatory requirements into line with new scientific knowledge on animal welfare during transport, based on opinions issued by the European Food Safety Authority (EFSA) at the request of the European Commission. Negotiations between the Member States are currently in progress.

Context as defined by the requesting body

The proposed regulation suggests lower densities (higher space allowances) than those in Regulation 1/2005, in order to improve the space available to the animals being transported and hence their welfare.

Professional organisations in the sector argue that the densities laid down in the current regulation mean that animals transported by road would be less likely to fall due to 'mutual support' between animals. They thus suggest that, if lower densities were applied as recommended in the draft regulation, the animals would be more likely to fall, lose their balance and therefore injure themselves, which would have negative impacts in terms of animal welfare.

Request

For the FRCAW to answer the following question as fully as possible:

> Is it the case that [cattle][1] transported by road at the densities set out in the proposed regulation to revise Regulation No 1/2005 are more likely to fall and/or be injured than [cattle] transported by road at the densities laid down in the current regulation (Regulation No 1/2005)?

The FRCAW will address only the transport of cattle by road in this report.

Reference documents

- COUNCIL REGULATION (EC) No 1/2005 of 22 December 2004 on the protection of animals during transport and related operations and amending Directives 64/432/EEC and 93/119/EC and Regulation (EC) No 1255/97
- Proposal for a REGULATION OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL on the protection of animals during transport and related operations and amending Council Regulation (EC) No 1255/97 and repealing Council Regulation (EC) No 1/2005
- EFSA AHAW Panel (2022b). Welfare of cattle during transport. EFSA Journal 2022;20(9):7442, 121 pp. https://doi.org/10.2903/j.efsa.2022.7442

[1] The request as originally worded concerns several species and therefore refers to 'animals' rather than 'cattle'. The present report deals only with the transport of cattle.

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Glossary (English version)

Haematoma

Collection of blood outside the blood vessels, in a body space, organ or tissue, as a result of damage to a vessel. The frequency of haematomas in animals may increase as a consequence of losses of balance and falls caused by rough driving (see EFSA, 2022).

Journey time

The time period during which animals are moved by means of transport, including the time for loading and unloading the animals (European Commission, 2023).

Loading density

Ratio between the number (or preferably live weight) of animals and the surface area available in the vehicle (usually expressed in kg/m^2) (Buckham-Sporer et al., 2023).

Long journey

In the current regulations, a long journey exceeds 8 hours, starting from when the first animal of the consignment is moved (Council of the European Union, 2004).

In the proposed regulation, this is a journey that exceeds 9 hours. A journey starts with the loading of the first animal at the place of departure and ends with the unloading of the last animal at the place of destination (European Commission, 2023).

Road transport vehicle

Means of wheeled transport that is propelled (lorry) or towed (trailer). The characteristics of transport vehicles vary greatly depending on the transporter and the country. They may have 1 to 5 decks, each of which may be divided into 2 to 4 compartments. According to EC regulation 1/2005 (EC Council, 2004), there are two types of transport vehicle: vehicles used under Type 1 transporter authorisation (< 8 hours) and those used under Type 2 authorisation (≥ 8 hours). In addition to the vehicular features required for both lengths of journey (weather protection, non-slip flooring surface, appropriate loading and unloading equipment, etc.), Type 2 vehicles must be equipped with a properly insulated light-colour roof, a specified water supply system, an active ventilation system, a temperature control system and a warning system to alert the driver if maximum or minimum temperature limits are reached. For journeys lasting 8 hours or more, animals of all ages must also be provided with appropriate bedding.

Space allowance

Area available per animal (expressed in m^2/animal), generally calculated on the basis of the weight and body dimensions of the animals (Petherick, 2007).

Stress

Stress, including in animals, refers to the presence of negative affective states. These states occur when the animal feels threatened, whether the threat is real or not. In order to adapt to this threat, the animal responds through its behaviour, with reactions of fight or flight if it is afraid, for example, and through its physiology, with an increase in heart rate and the secretion of certain hormones to enable physical effort, among other things.

Transport[2]

The movement of animals effected by one or more means of transport, and the related operations, including loading, unloading, transfer and rest, until the unloading of the animals at the place of destination is completed (Council of the European Union, 2004) . *This report deals only with road transport by lorry.*

Wound

Any damage to the skin, which may take the form of small superficial punctures, scratches, or larger open lesions that are more than skin deep (adapted from Welfare Quality Network, 2019).

[2] Described as 'journey' in the European Council (2023) proposal.

Abbreviations

ALAT

Alanine aminotransferase

AST

Aspartate aminotransferase

BHB

Beta-hydroxybutyrate

CK or CPK

Creatine kinase (CK) or creatine phosphokinase (CPK)

EURCAW

European Reference Centre for Animal Welfare

FRCAW

French Reference Centre for Animal Welfare (CNR BEA)

NEFA

Non-esterified fatty acids

Tables and Figures

Tables

Figures

1 Method

This document provides a synthesis of the information on the risk of falls and injuries associated with transport density set out in the EFSA opinion (2022b) on the welfare of cattle during transport. Further detail has been provided by an in-depth analysis of the literature on this particular subject, including the grey literature and, where possible, materials published since the EFSA opinion was issued.

The bibliographical corpus was established by first conducting a search on the Web of Science(TM) (WOS) platform using the following search parameters:

("loading densit*" OR "stocking densit*" OR "densit*" OR "space allowance*" OR "surface*") AND transport AND (welfare OR "well-being" OR "injur*" OR "wound*" OR "fall*" OR "bruise*" OR "stress*" OR "behavior" OR "behaviour") AND ("calves" OR "calf" OR "cow*" OR "cattle" OR "young bull" OR "young bulls").

From the 364 documents obtained in the search, 58 titles and summaries of interest were selected for further study, including 22 review articles or chapters in books. These documents contained references to a further 8 documents that were deemed relevant to the subject of this report and were added to the corpus.

To obtain materials from the grey literature, we consulted documents relating to animal transport and welfare posted on the IDELE (Institut de l'élevage), EURCAW Ruminants & Equines and European Commission websites. 4 documents from these sources were selected.

In total, the initial corpus thus comprised **70** documents. Of these, **15** were based on experimental research comparing loading densities and are discussed in the review of additional literature (see *3.2*). The other documents in the corpus (review articles, surveys, etc.), some of which are cited in the EFSA opinion, have contributed to the development of the discussion in this report (see *4*).

2 Cattle loading densities and space allowance

The term loading density in this report refers to the live weight of cattle within a lorry compartment, expressed in kg/m^2. The concept of available surface area, which represents the relationship between space and animal in the opposite way, is expressed in m^2/animal (but not in kg). To facilitate comparison between the various scientific studies discussed here the unit of measurement mainly used in this report is surface area in m^2 per 100kg. The EFSA uses the term 'space allowance' to designate the surface area allocated to animals during transport, and this term will therefore be used, where appropriate, in the English-language version of this report.

2.1 The current regulations

Chapter VII (Part B. Bovine Animals) of the technical rules set out in Annex 1 of COUNCIL REGULATION (EC) No 1/2005 of 22 December 2004 on the protection of animals during transport and related operations and amending Directives 64/432/EEC and 93/119/EC and Regulation (EC) No 1255/97 (hereinafter referred to as **Regulation No 1/2005**) details the authorised space allowances for cattle transported by road. The allowances corresponding to each category of animal are summarised in *Table 1.*

Table 1. Space allowances for the transport by road of cattle under the current regulations (Regulation No 1/2005)

Category	*Approximate weight (kg)*	*Area (in m²/animal)*	*Area (in m²/100kg)*
Small calves	50	0.30 to 0.40	0.60 to 0.80
Medium-sized calves	110	0.40 to 0.70	0.36 to 0.64
Heavy calves	200	0.70 to 0.95	0.35 to 0.48
Medium-sized cattle	325	0.95 to 1.30	0.29 to 0.40
Heavy cattle	550	1.30 to 1.60	0.24 to 0.29
Very heavy cattle	> 700	> 1.60	> 0,23

2.2 Proposal for a regulation

Chapter VII (Clause 2) of Annex I of the Proposal for a REGULATION OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL on the protection of animals during transport and related operations and amending Council Regulation (EC) No 1255/97 and repealing Council Regulation (EC) No 1/2005 proposes that the space allowance should be calculated using the following allometric equation:

$A=k*W^{2/3}$, where A = area per animal (in m²), W = live weight (in kg), and where k is a constant specific to a given species (or group of species). For cattle, the value recommended by EFSA is 0.034 (EFSA, 2022).

The minimum space allowances for cattle under the proposed regulations are summarised in *Table 2*.

Table 2. Minimum space allowances for the transport by road of cattle under the proposed regulations

Live weight (kg)	50	75	100	125	150	175	200	225	250	275	300	325	350	375
Area (m²/animal)	0.46	0.60	0.73	0.85	0.96	1.06	1.16	1.26	1.35	1.44	1.52	1.61	1.69	1.77
Area (m²/100kg)	0.92	0.8	0.73	0.68	0.64	0.61	0.58	0.56	0.54	0.52	0.51	0.50	0.48	0.47
Equivalent category	Small calves		Medium-sized calves				Heavy calves					Medium-sized cattle		
Live weight (kg) - continued		400	450	500	550	600	650	700	750	800	850	900	950	1000
Area (m²/animal) - continued		1.85	2.00	2.14	2.28	2.42	2.55	2.68	2.81	2.93	3.05	3.17	3.29	3.40
Area (m²/100kg) - continued		0.46	0.44	0.43	0.41	0.40	0.39	0.38	0.37	0.37	0.36	0.35	0.35	0.34
Equivalent category		Medium-sized cattle			Heavy cattle				Very heavy cattle					

Figure 1, below, shows the minimum space allowance for cattle during transport under the proposed regulations (shown in blue, based on the allometric equation using k = 0.034) compared with the current regulations (upper limits in green and lower limits in red).

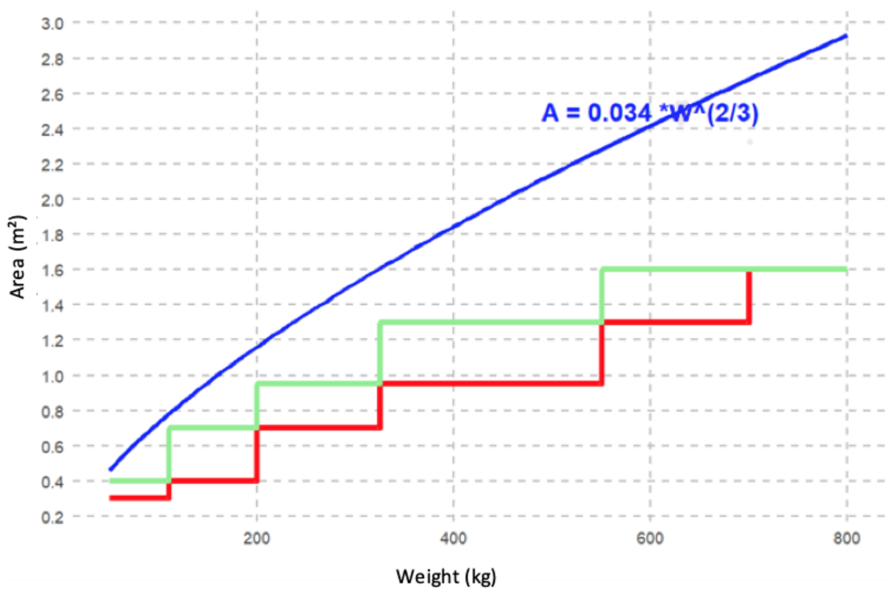

Figure 1. Proposed minimum space allowance for cattle during transport. *Profile of the allometric equation in the new proposal (using constant k = 0.034), shown with the upper (green) and lower (red) limits required under current regulations (Regulation No 1/2005). Produced using RStudio - 2024/2025* [3]

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[3] Inspired by: European Commission (2018) Guide to good practice for the transport of cattle. Animal Transport Guides Project Consortium, revision 1, May 2018. https://op.europa.eu/en/publication-detail/-/publication/f452d88b-7ebd-11ea-aea8-01aa75ed71a1

Box 1. Comparison of loads permitted under the current and proposed regulatory space allowances

By way of example, for a conventional cattle lorry with an area of 30m^2 per deck, containing 2 – 3 decks (for adult cattle or calves respectively), the current regulatory space allowances make it possible to transport up to 300 calves (weighing 50kg), 63 medium-sized cattle (weighing 325kg), or 46 heavy cattle (weighing 600kg), although in practice it is common for lorries to be less heavily loaded, particularly for calves, since load sizes are constrained by the available space at the farms of destination. Under the space allowances in the regulatory proposal, a single lorry could transport up to 195 calves (weighing 50kg), 37 medium-sized cattle (weighing 325kg) or 24 heavy cattle (weighing 600kg).

Details of the calculations are available here in French (modifiable variables = animal category, average weight, surface area per deck and number of decks).

3 Summary of the literature on the risks of falls and injuries as a function of the space allowance

3.1 The EFSA's view

The EFSA takes the view that the space allowance during transport is a determining parameter for the welfare of cattle and recommends the fixing of a minimum space allowance per animal based on the use of an allometric equation ($A = k \times W^{2/3}$), requiring a minimum k-value of 0.034 for cattle to protect them from various negative consequences such as restriction of movement, resting problems, heat stress or distress.

The EFSA's assessment of minimum space requirements takes into account the essential biological functions of cattle during transport, including:

(a) the area required by animals of different physical sizes in a standing posture,

(b) the ability of the animals to adjust their posture in response to the movements of the vehicle (requiring a k-value of at least 0.034),

(c) the possibility for the animals to rest in a lying posture (requiring a k-value of 0.033 to enable all the animals to lie down simultaneously),

(d) the ability of the animals to thermoregulate,

(e) access to feed and water by the animals if provided (requiring a k-value of 0.0315).

Regarding the risk of falls and injuries specifically, the EFSA considers that a space allowance higher than a k-value of 0.034 would benefit cattle by providing them with space to maintain stability during transport, and would therefore reduce the risk of injury and suffocation induced by falls. The EFSA also considers the possibility that, in certain transport situations (poor driving or emergency responses) cattle could benefit from smaller space allowances that would provide mutual support (i.e., enable individual animals to lean on each other to avoid falling). **However, on the basis of the studies analysed, the EFSA considers that, in general, cattle are at greater risk of injury at low space allowances (= higher loading densities) than high allowances (= lower loading densities).**

Last, the EFSA expresses its regret at the limited number of scientific studies to have investigated the effect of loading densities lower than those set by Regulation No 1/2005 on the risk of falls and injuries, and emphasises that even lower densities could have other benefits for the welfare of cattle.

3.2 Additional literature

Table 3 provides a summary of the results of the 14 experimental studies found in the literature. Each line of the table corresponds to a study (ranging from 1988 to 2024) that evaluates, for different types of cattle (beef cattle, bulls, calves, etc.), the effects on various indicators of the space allowance during transport. References shown in bold were taken into account in the EFSA opinion. It should be noted that the discussion of space allowances in the EFSA opinion does not specifically examine the risk of injuries and falls as a function of transport densities.

The table shows, for each weight of animal studied, the relevant space allowance (in m^2/100kg) as set out the proposed regulations (green column) and the current regulations (blue column). It provides details of the different space allowances for the animals in each study (m^2/100kg), based on the densities or space allowances (m^2/animal) used in each article, along with the average weight of the animals studied.

Given that falls and injuries are rarely observed directly in the studies reviewed, the FRCAW has chosen to consider in its analysis all the behaviours observed, as well as the meat quality parameters studied, the latter being able to provide indirect clues relating to injuries (observed on the carcasses), but also relating to the stress felt by the cattle during transport. To complete this analysis, physiological indicators were also considered in order to obtain additional information on the stress experienced by the cattle during transport. *Figure 2* summarises the various links between the parameters considered in this report.

Table 3. Summary of the results of experimental studies on the impact of the space allowance for cattle during transport on their risk of injuries and falls, through analysis of behaviour, physiology and meat quality

The space allowances shown in the green boxes correspond to those in the new regulatory proposal, those in the blue boxes to the current regulatory requirements, and those in the red boxes are lower than the current regulatory requirements. Conclusion column: "+" the study concludes that there is an improvement in the welfare of the cattle with an increase in the space allowance (or a decrease in stocking density) / "=" the study concludes that an increase in the space allowance has no significant negative impact on welfare / "-" the study concludes that there is a deterioration in the welfare of the animals with an increase in the space allowance. Unless otherwise indicated, all results are significant. NS: not significant. Abbreviations: []°: concentration, AST: aspartate aminotransferase, ALT: alanine aminotransferase, BHB: beta-hydroxy-butyrate, CK: creatine kinase, CPK: creatine phosphokinase, LDH : lactate dehydrogenase, NEFA: non-esterified fatty acids, PCV: Packed Cell Volume (or haematocrit), L: luminosity, a*: red colour, b*: yellow colour, pH_{mus}: muscle pH, pm: post-mortem. "/": parameter not studied/mentioned in the article.*

Type of cattle	Reference (in bold when cited in EFSA opinion)	Journey duration (h)	Average weight (kg)	Minimum space allowance to be respected according to the proposed regulation (m^2/100kg)	Current regulatory minimum area (m^2/100kg)	Space allowance studied (m^2/100kg)			Behaviour	Physiology	Injuries/meat quality	Conclusion
Calves	(Grigor et al., 2001)	18 (2 x 9h, separated by a break of 1h or 12h)	48	0.94	0.6	**0.98**	**0.77**		Loss of balance, falls, contact with other calves, time spent lying down, water consumption: NS **- At the highest space allowance:** orientation both perpendicular and parallel to the direction of travel **- At the lowest space allowance:** ⇧ perpendicular orientation	Plasma parameters (CK activity, osmolarity, []° potassium, []° glucose, []° free fatty acids, []° cortisol, []° albumin, []° sodium, []° chloride), PCV, heart rate: NS	/	=
	(Jongman & Butler, 2014)	12	38	1.01	0.79	**1.32**	**0.79**	**0.53**	**At the lowest space allowance:** - ⇩ changes in posture (difficulty getting up and lying down) - ⇧ agitation - ⇧ rest on arrival **At the highest space allowance:** - ⇧ resting time in the presence of straw bedding - ⇧ ability to reposition - No loss of balance observed	**At the lowest space allowance:** ⇧ CK activity **At the intermediate space allowance:** ⇧ of CK (vs lowest allowance) Other parameters (PCV: indicator of dehydration BHB: indicator of metabolic state): NS	/	+
	(Todd et al., 2000)	12	~40	1	0.75	**1**		**0.5**	/	PCV, BHB, urea, total plasma proteins: NS - ⇩ space allowance: - []° plasma glucose higher from 16h after feeding - ⇧ []° plasma lactate - ⇧ plasma CPK	/	+
	(Uetake et al., 2011)	1 or 1.7 or 2.3	51	0.91	0.58	**0.88**	**0.68**	**0.49**	**At the lowest space allowance:** ⇩ number of animals lying down **At the highest space allowance:** ⇧ change of direction	Holstein calves: **⇧ space allowance:** ⇩ []° plasma noradrenalin, ⇧ []° blood lactic acid []° total protein: NS Crossbred calves: **⇧ space allowance:** ⇧ []° plasma noradrenalin, ⇩ []° total protein, ⇩ []° blood lactic acid	/	+

Type of cattle	Reference (in bold when cited in EFSA opinion)	Journey duration (h)	Average weight (kg)	Minimum space allowance to be respected according to the proposed regulation (m²/100kg)	Current regulatory minimum area (m²/100kg)	Space allowance studied (m²/100kg)	Behaviour	Physiology	Injuries/meat quality	Conclusion
Heavy calves	(Abubakar et al., 2024) and **(Abubakar et al., 2021)**[4]	9 or 17	290	0.51	0.24-0.33	**0.5** **0.25** **0.17**	/	/	**⇩ space allowance:** - ⇧ L*, a*, b* at D1, D7 and D14 pm - ⇩ muscle glycogen at D0, D1, D7, D14 pm - ⇧ pH_{mus} at D0, D1 and D14 pm - ⇧ cooking losses at D1, D7, and D14 pm	+
Heavy calves	**(Earley & O'Riordan, 2006)**	12	250	0.54	0.28-0.38	**0.51** **0.34**	/	Haematological variables: NS - Plasma albumin, AST, BHB, NEFA glucose, LDH, CPK, cortisol: NS	/	=
Heavy calves	**(Grigor et al., 2004)**	3	234	0.55	0.30-0.41	**0.41** **0.30**	Behaviours involving loss of balance, falls or collisions between calves: NS **- ⇧ space allowance:** ⇧ mounts and movements Latency before lying down on arrival and time spent lying down on arrival: NS	Heart rate, [ ]° plasma CK, [ ]° plasma cortisol: NS	L*, a*, b*, pH_{mus} at D1, and shear strength: NS Tenderness, juiciness, meat flavour: NS **- ⇧ space allowance:** ⇩ cooking losses	=
Medium-sized cattle	(Brennecke et al., 2021)	1	~400-500	0.44	0.21-0.28	**0.24** **0.22** **0.20**	/	/	**At the lowest space allowance:** ⇧ weight of lesions on carcasses compared with higher space allowances	=
Medium-sized cattle	(Eldridge et al., 1988)	1.5 or 0.5 or 6	~350	0.48	0.27-0.37	**0.33** **0.31** **0.29** **0.26** **0.25** **0.23**	Orientation of animals in the truck, number of movements: NS	⇩ space allowance: ⇩ heart rate	/	-
Medium-sized cattle	(Eldridge & Winfield, 1988)	6	400	0.46	0.24-0.33	**0.35** **0.29** **0.22**	**At the highest space allowance (compared to the lowest):** - More movements > 1m **At the intermediate space allowance (compared to the lowest and highest):** - More animals aligned with the direction of the truck Movements associated with aggression: NS	/	**At the intermediate space allowance (compared to lowest and highest):** - Lower haematoma score - Lower total number of haematomas on carcasses	-
Medium-sized cattle	(Mounaix et al., 2009)	29 (2 times 14, separated by 1h break)	396	0.46	0.24-0.33	**0.34** **0.29** **0.26**	**At the highest space allowance (compared to the intermediate space allowance) before the break:** - ⇧ aggressive behaviour - ⇧ loss of balance in standing animals - ⇩ perpendicular positions - ⇧ diagonal positions - ⇧ changes of position - ⇩ constrained movements **At the highest space allowance (compared to the intermediate allowance) before and after the break:** - ⇧ overlaps - ⇧ free movements	Biochemical indicators of fatigue or stress: NS Amount of feed consumed and weight loss: NS	/	-

[4] Two articles based on the same study, but describing complementary results

Type of cattle	Reference (in bold when cited in EFSA opinion)	Journey duration (h)	Average weight (kg)	Minimum space allowance to be respected according to the proposed regulation (m²/100kg)	Current regulatory minimum area (m²/100kg)	Space allowance studied (m²/100kg)						Behaviour	Physiology	Injuries/meat quality	Conclusion
Heavy cattle	(Ferreira et al., 2020)	?	Males: 511.4 (Straight Truck), 618.4 (Livestock Trailer), 544.0 (Straight truck + Trailer) Females: 337.4 (Straight Truck), 357.9 (Livestock Trailer)	Males: 0.43 (ST)/ 0.4 (LT)/ 0.42 (STT) Females: 0.49 (ST)/ 0.48 (LT)	Males: 0.24-0.29 Females: 0.29 - 0.4	**0.36 (F - ST)**	**0.35 (F - LT)**	**0.25 (M - ST)**			**0.21 (M - LT + STT)**	/	/	Males: **STT (lower space allowance):** highest percentage of haematomas on the ribs and lowest on the front of the body **ST (highest space allowance):** lowest percentage of haematomas on the ribs Females: **LT (lowest space allowance):** higher percentage of haematomas at the front of the body	+
	(Tarrant et al., 1988)	4	603	0.4	0.22-0.27	**0.5**	**0.33**				**0.17**	**⇩ space allowance:** - ⇧ falls - ⇧ loss of balance - ⇩ exploratory behaviour - ⇧ aggressive behaviour - ⇩ sexual interactions - ⇩ ability to adopt a preferred position	**⇩ space allowance**: - ⇧ plasma cortisol - ⇧ plasma glucose - ⇧ plasma CK	**⇩ space allowance:** - ⇧ carcass haematoma score	+
	(Tarrant et al., 1992)	24	618	0.4	0.21-0.26				**[0.22 – 0.23]**	**[0.19 – 0.2]**	**[0.17 – 0.18]**	**At the lowest space allowance**: - ⇧ falls - ⇧ difficulty maintaining balance - ⇩ shifts of footing - ⇧ mounting behaviour	**At the lowest space allowance:** - ⇧ plasma cortisol - ⇧ plasma glucose - ⇧ plasma CK - ⇧ red blood cells, haemoglobin and cell volume	**At the lowest space allowance:** - ⇧-haematoma score on carcasses	+

3.2.1 Calves

Of the 14 studies, 4 investigated young calves (around 50kg) and 3 at heifers or bull calves, classified as "heavy calves" (200 to 300kg) under current regulations (Council of the European Union, 2004). A total of 7 studies were therefore concerned with calves, 4 of which compared an allowance corresponding to the space allowances in the proposed regulation with allowances in line with current requirements, or even lower. Of these 7 studies, 4 concluded that transporting calves at the higher experimental space allowance improved their welfare compared with transporting them at lower allowances. The last 3 studies found no negative impact on calf welfare (and therefore no increase in the risk of injury or falls) from the increase in space allowance (*Table 4*).

Table 4. Number of experimental studies on calves showing the authors' conclusions relating to an increase in space allowance, and indicating whether or not space allowances corresponding to those in the regulatory proposal were studied

Category	Number of studies	Number of studies including space allowances corresponding to those in the regulatory proposal	Number of studies concluding that an increase in space allowance during transport ...		
			...would be beneficial to welfare (including space allowances corresponding to those in the regulatory proposal)	...would not have a negative effect on welfare (including space allowances corresponding to those in the regulatory proposal)	...would be detrimental to welfare (including space allowances corresponding to those in the regulatory proposal)
Calves	4	3	3 (2)	1 (1)	0 (0)
Heavy calves	3	1	1 (1)	2 (0)	0 (0)
Total	7	4	4 (3)	3 (1)	0 (0)

With regard to **falls** specifically, only 3 studies observed losses of balance and potential falls in calves, including 2 studies that compared an allowance corresponding to the regulatory proposal with an allowance in line with the current regulation. **Neither of these 2 studies observed an increased number of losses of balance or falls at space allowances corresponding to the new regulatory proposal compared with space allowances in line with the current regulations**.

With regard to **injuries**, none of the studies directly observed physical injuries to calves during transport. The studies looked at other parameters, such as indicators of fatigue, agitation or stress. Nevertheless, **5 studies measured creatine kinase (CK, or creatine phosphokinase - CPK) concentrations in the blood as an indirect indicator of potential haematomas**. The concentration of creatine kinase in the blood increases following damage to muscle cells resulting from stress or direct trauma to the muscle. An increase in CK activity may therefore indicate that standing animals have muscular fatigue or that lying animals are trampled (leading to haematomas) (Jongman & Butler, 2014). Of these studies, two compared two space allowances above the current regulatory threshold but below the allowances in the regulatory proposal. The authors did not observe any significant difference in CK concentrations between calves transported at the different allowances. The other three studies compared allowances meeting the requirements of the proposed regulations with lower allowances (in line with the allowances in the current regulations or even lower). Of these, one study showed no significant difference in plasma CK activity as a function of surface area (Grigor et al., 2001). The other two observed respectively a lower CK activity and a lower plasma CK concentration in calves transported at the highest allowances[5], thus indicating a potentially lower number of haematomas in these calves and, at the very least, reduced muscular fatigue.

It should be noted that one study highlighted a significantly higher number of mounting behaviours in calves weighing around 230kg transported at 0.41m²/100kg compared with those transported at 0.3m²/100kg[6] (median, first and third quartile of respectively 5, 3, 7 events per hour compared with 0, 0, 0 events per hour, $p < 0.001$; Grigor et al., 2004). Such overlapping in a confined space may increase the risk of injury. However, the authors of the study did not find this to be the case and stated that this mounting activity did not contribute to any increase in useful muscle pH, measured on the meat at the abattoir.

In conclusion:

In view of the information available in the literature, it would appear that calves transported at space allowances corresponding to those in the proposed regulations are no more likely to fall or be injured than calves transported at allowances in line with the current regulations. On the contrary, it would appear that calves transported at these new space allowances have a reduced risk of injury.

3.2.2 Young and adult cattle

Out of 14 studies, 4 looked at medium-sized cattle (350 to 500kg) and 3 looked at large cattle (500 to 700kg). In total, therefore, 7 studies looked at young cattle or adult cattle, only one of which compared an allowance corresponding to the space allowances in the regulatory proposal with allowances in line with current requirements, or even lower. Of these studies, three concluded that transporting cattle at the highest experimental allowance improved their welfare compared with transporting them at lower allowances (fewer haematomas on carcasses, fewer losses of balance and falls, less stress and muscle fatigue in large cattle at higher allowance). Three other studies concluded, conversely, that transporting cattle at the highest experimental allowance was likely to have a negative impact on their welfare compared with one or more lower experimental allowances (increased heart rate, more haematomas on carcasses, and more losses of balance and mounting behaviours in young

[5] $Log_{(10)}$ (CK) giving values of 2.33, 2.40, and 2.16 respectively, depending on flooring type at a space allowance of 0.79m²/100kg, and 2.13, 2.29, and 2.07 at a space allowance of 1.32m²/100kg, $p \leq 0.001$ (Jongman & Butler, 2014). Post-transport CK concentrations were approximately 460 U/L at a space allowance of 0.5m²/100kg versus approximately 100 U/L at 1m²/100kg, $p < 0.05$ (Todd et al., 2000).

[6] Two space allowances lower than the regulatory proposal.

cattle transported at the highest space allowance were linked to possible stress). Last, one study found no significant negative impact on the welfare of cattle transported on the highest experimental allowance compared with the lowest experimental allowance *(Table 5)*.

Table 5. Number of experimental studies on young and adult cattle showing the authors' conclusions relating to an increase in space allowance, and indicating whether or not allowances corresponding to those in the regulatory proposal were studied

Category	Number of studies	Number of studies including space allowances corresponding to those in the regulatory proposal	Number of studies concluding that an increase in space allowance during transport...		
			...would be beneficial to welfare (including space allowances corresponding to those in the regulatory proposal)	... would not have a negative effect on welfare (including space allowances corresponding to those in the regulatory proposal)	... would be detrimental to welfare (including space allowances corresponding to those in the regulatory proposal)
Medium-sized cattle	4	0	0 (0)	1 (0)	3 (0)
Heavy cattle	3	1	3 (1)	0 (0)	0 (0)
Total	7	1	3 (1)	1 (0)	3 (0)

In these studies, falls were generally observed via video recordings, while injuries were generally assessed through the presence and/or severity of lesions and haematomas (often characterised by a score), observed either on live animals on arrival at the destination, or on carcasses at the abattoir, the latter being the most frequent method. It should be noted that haematomas observed on carcasses may be linked to events during transport in the vehicle, but also to the loading phase and the unloading and waiting phases at the abattoir, during which the animals may also fall and/or injure themselves.

Only one of the seven studies compared a space allowance corresponding to those in the regulatory proposal with an allowance in line with the current regulations. The authors of this article observed a decrease in the haematoma score[7] on carcasses with an increase in the space allowance, with scores averaging 1.6 and 3.1 respectively (depending on the type of journey) with a space allowance of 0.5m^2/100kg of cattle (600kg cattle on average) compared with scores averaging 3.2 and 3.6 with a space allowance of 0.33m^2/100kg of cattle (allowance already slightly higher than the current regulations) ($p < 0,01$). With regard to the risk of falls, the authors noted that, at a space allowance of 0.5m^2/100kg of cattle, the animals adopted their preferred position significantly more, i.e. in line with the direction of travel, making it easier to maintain their balance. Nevertheless, they did not observe any significant difference in the number of falls between the two space allowances of interest. They did, however, observe significantly more falls on the lower space allowance (below the current regulatory requirements compared with the two space allowances of interest) (Tarrant et al., 1988).

[7] Score awarded at the abattoir by a panel of 3 judges, ranging from 0 (no haematomas) to 7.

In conclusion:

The available literature (a single study) on the transport of adult cattle seems to suggest that an increase in space allowance (and therefore a decrease in density) to match the regulatory proposal does not increase the risk of falls and injuries for heavy cattle compared with the current regulatory space allowances. On the contrary, this increase in space allowance would reduce the risk of injury during transport. **However, further studies comparing these two space allowance ranges would be needed to confirm this hypothesis**.

Of the other studies to examine the impact of the space allowance during transport on the risk of cattle falls and injuries, a distinction has been made between studies of heavy cattle (over 500kg) and those concerned with lighter adult cattle.

The other two studies on **heavy cattle**, which compared the regulatory space allowances with smaller areas, both concluded that the lower space allowances (0.21m^2/100kg and 0.17 to 0.18m^2/100kg respectively) resulted in significantly more haematomas on the carcasses[8] than the higher allowances (8.5[9] versus 3.7[8] ($p < 0.01$) and 30.5%[10] versus 7.2%[9] ($p < 0.001$) respectively). Tarrant et al (1992) also observed a higher number of losses of balance and falls at a space allowance of between 0.17 and 0.18m^2/100kg compared with allowances of between 0.19 and 0.23m^2/100kg (respectively 10 losses of balance compared with 4 and 5, and 8 falls compared with 1 and 1[11]).

Figure 2 shows the number of losses of balance and falls observed during transport as a function of the space allowance (in m^2/100kg) reported by the only two articles in the corpus to include this indicator in their results (Tarrant et al., 1988, 1992). According to these two articles, the relationship between losses of balance or falls and the space allowance during transport weakened, although the number of falls actually observed was very low at space allowances above around 0.2m^2/100kg. It should be noted that these articles concerned only heavy cattle weighing over 600kg.

[8] Specifically at rib level for the study by Ferreira et al. (2020).
[9] Score awarded at the abattoir by a panel of 3 judges, ranging from 0 (no haematomas) to 7.
[10] Percentage of haematomas on carcasses in the rib region.
[11] Data corresponding to the sum of losses of balance or falls observed over periods of 10 minutes per hour over 24 hours of transport (averaged over 3 journeys).

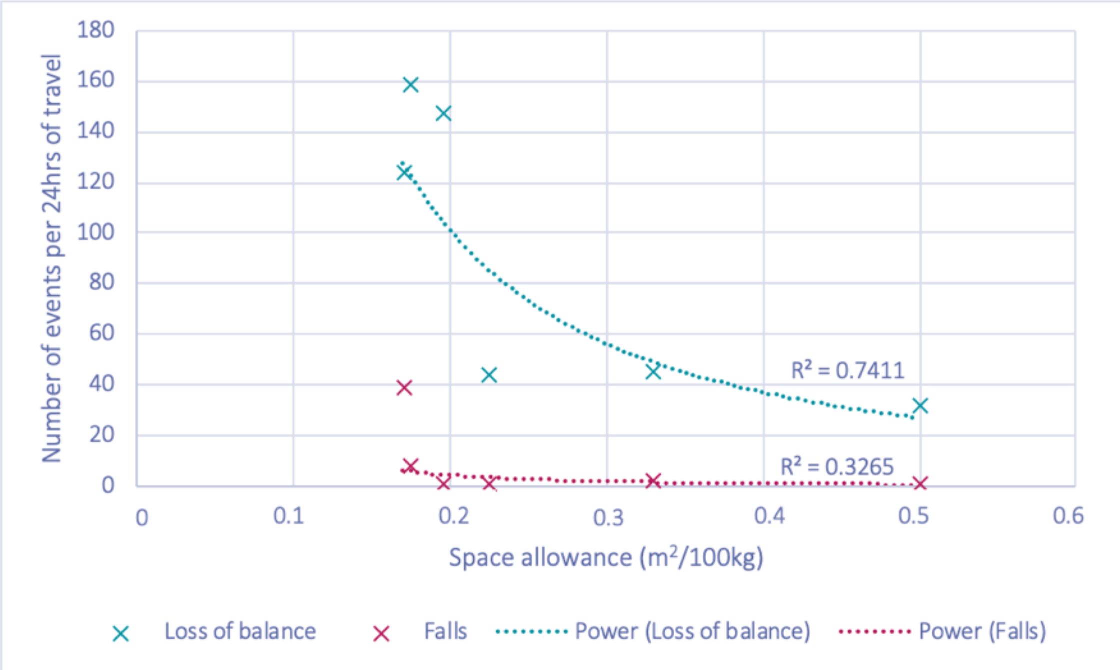

Figure 2. Number of losses of balance (blue-green) and falls (pink) as a function of the space allowance during transport.

Graph based on the two articles in the corpus showing the number of losses of balance and falls observed during transport (Tarrant et al., 1988, 1992)

The four studies to examine **medium-sized cattle** (300 to 500kg) allow no conclusions to be drawn regarding the impact of an increase in the space allowance on the risk of falls and injuries, as they did not look specifically at falls during transport. Nevertheless, two articles considered lesions and haematomas on the carcasses of medium-sized cattle after transport. The first noted a significantly lower average lesion weight (0.30± 0.40kg) for cattle weighing 400 to 500kg transported at a space allowance of 0.22m^2/100kg compared with the average lesion weight for cattle transported at a space allowance of 0.20m^2/100kg (0.80± 1.30kg, p > 0.0001). The lesion weight was not significantly different between space allowances of 0.22m^2/100kg and 0.24m^2/100kg (Brennecke et al., 2021). The second study observed both a lower haematoma score and a lower number of haematomas on carcasses for cattle weighing around 400kg transported at a space allowance of 0.29m^2/100kg (haematoma score = 1.9, average number of haematomas on carcasses = 1.2) compared with space allowances of 0.22m^2/100kg and 0.35m^2/100kg (haematoma scores were 8.2 and 4.6 respectively (p < 0.01) and mean numbers of haematomas on carcasses were 3.2 and 2.3 respectively (p < 0.01)) (Eldridge & Winfield, 1988). It is therefore possible that the optimum transport space allowance would be around 0.29m^2/100kg of cattle, with a greater chance of limiting animal injuries than higher or lower space allowances. Nevertheless, further studies are needed to either confirm or refute this hypothesis, particularly with regard to space allowances that correspond to those in the regulatory proposal.

Figure 3 shows the haematoma score observed at the abattoir on carcasses as a function of the space allowance during transport (in m^2/100kg) as reported by the only three articles in the corpus to use indicators that allowed comparison (Eldridge & Winfield, 1988; Tarrant et al., 1988, 1992). All three articles used the haematoma score established by Anderson & Horder (1979) in observing injuries. The three articles concluded that the relationship between injuries and the space allowance during transport tends to weaken as the area increases.

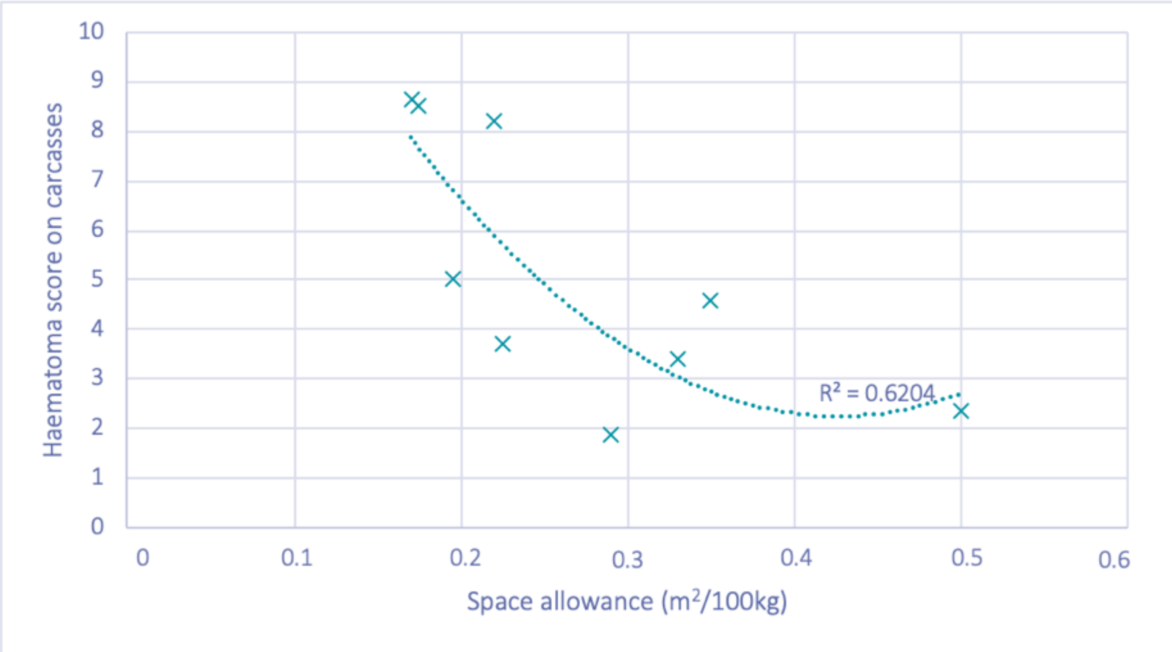

Figure 3. Haematoma scores recorded on carcasses as a function of the space allowance during transport

Graph based on the three articles in the corpus that used a comparable indicator, i.e., the carcass haematoma score established by Anderson & Horder (1979) (Eldridge & Winfield, 1988; Tarrant et al., 1988, 1992).

Several additional studies have also attempted to establish a link between the space allowance for cattle during transport and the risk of injury by conducting *a posteriori* analysis of large quantities of data supplied by abattoirs or cattle transporters (Bethancourt-Garcia, Vaz, Vaz, Restle, et al., 2019; Bethancourt-Garcia, Vaz, Vaz, Silva, et al., 2019; González et al., 2012; Mendonça et al., 2019; Romero et al., 2012, 2013; Vaz, Dutra, et al., 2023; Vaz, Mendonça, et al., 2023; Vimiso et al., 2013; Zanardi et al., 2022). The analysis in these articles is less precise for two reasons. First, several different categories of cattle were included in each study, resulting in extensive variations in animal weights and the surface areas available to each animal. Second, the variables for the different transport routes analysed were not standardised. Of these studies, **five concluded that the risk of injury increases when the space allowance decreases**, particularly when the latter is less than 0.31 or 0.33m^2/100kg (Romero et al., 2013; Vaz, Mendonça, et al., 2023; Vimiso et al, 2013). In other studies, experimental space allowances were set even lower, at less than 0.23m^2/100kg or 0.25m^2/100kg compared with allowances of up to 0.27m^2/100kg or more (Bethancourt-Garcia, Vaz, Vaz, Restle, et al., 2019; Bethancourt-Garcia, Vaz, Vaz, Silva, et al., 2019; Mendonça et al., 2019). One hypothesis put forward by some authors is that a lower space allowance makes cattle that fall more likely to be trampled by their fellows, who seek to occupy as much of the available space as possible. **Conversely, two studies have established a positive linear relationship between space allowance and the probability of haematomas on carcasses** (Romero et al., 2012; Zanardi et al., 2022). These authors assume that the excess available space available could encourage losses of balance. It should be noted that one of these studies expresses density in terms of animals/m^2 without specifying the weight of the individual animals involved, making it impossible to know the available experimental surface areas per 100kg of cattle.

In conclusion:

The literature does not provide an answer to the question posed[12] for medium-sized cattle, due to the lack of studies comparing space allowances corresponding to the regulatory proposal with lower space allowances, and the lack of studies containing conclusions on cattle falls during transport. The studies that have compared regulatory space allowances with each other or with lower space allowances do not allow conclusions to be drawn as to the cause of injuries observed on cattle carcasses. The dates at which haematomas observed on the carcasses occurred were not systematically recorded, making it difficult to determine whether these injuries were exclusively the result of travel in the vehicle (and, in particular, falls during transport) or whether they also occurred during handling before or after transport. **Nevertheless, at present, there is no scientific evidence to suggest that the transport of medium-sized cattle at space allowances corresponding to the regulatory proposal make them more likely to fall and/or be injured than cattle transported at current regulatory space allowances.**

It should also be noted that the lack of consensus between studies on the impacts of an increase in the available transport surface area on cattle welfare can be largely explained by the presence of multiple co-factors that may contribute to the risk of falls and injuries in cattle during transport. These include quality of driving, the configuration of the lorry, the length and distance of the journey, the type of road, etc. (see *4.2*).

[12] "Is it the case that [cattle] transported by road at the densities set out in the proposed regulation to revise Regulation No 1/2005 are more likely to fall and/or be injured than [cattle] transported by road at the densities laid down in the current regulation (Regulation No 1/2005)?"

4 Additional analysis and discussion

4.1 The impact of different space allowances on cattle stress during transport

Falls during transport can be a source of stress for cattle. To gain a better understanding of the mechanisms involved in losses of balance and the additional consequences this may have on cattle, two lines of analysis of the literature are proposed to complement the content of the previous section. These address the consequences of variations in space allowance on **1) behavioural responses and 2) physiological responses**. Physiological responses alone are not specific to stress, as they can also be linked to an increase in physical activity or vigilance. It is therefore essential to analyse physiological responses in relation to the environmental context and to animals' behavioural responses in order to be able to interpret them in terms of stress. It should be noted that physiological and behavioural responses influence meat quality, affecting post-mortem temperature and pH reduction for example, which in turn have an impact on meat colour, tenderness and other carcass quality characteristics (Terlouw & Bourguet, 2022).

4.1.1 Behavioural responses

4.1.1.1 Calves

The studies in the corpus that examined the behaviour of calves during transport essentially looked at two parameters: changes of position between standing and lying down, and the orientation adopted in the lorry.

Cattle are known to lie down very little during transport, particularly during the first few hours of transport and/or when temperatures are high. However, cattle will lie down after a few hours as they become accustomed to transport conditions and/or start to feel tired (Grigor et al., 2001; Knowles, 1999). It would nevertheless appear that young calves are more likely to lie down during transport than adult cattle. Indeed, a study on the lying behaviour of calves during transport found that they spent more than 40% of the journey time lying down (Jongman & Butler, 2014). Jongman and Butler (2014) also noted that the younger the calves, the more likely they were to lie down during transport (3-day-old calves lay down 59% of the time compared with 42% of the time for 10-day-old calves). Further, it would appear that the time spent by calves lying down also depends on the type of bedding provided in the lorry. In particular, these same authors observed that calves transported on a space allowance of 1.32m^2/100kg remained lying down for 71% of the journey when straw bedding was provided (compared with 42% on bare flooring).

All studies in the corpus that observed the lying behaviour of calves during transport concluded that calves lie down more when they have more space, particularly on space allowances that correspond to those in the regulatory proposal (1.32m^2/100kg - Jongman & Butler, 2014 ; 1m^2/100kg - Todd et al, 2000[13] ; 0.88m^2/100kg - Uetake et al., 2011)[14] compared with a lower space allowance.

With regard to the orientation of calves in the lorry, it would appear that they orient themselves in line with the direction of travel when they have the opportunity to do so, particularly when they are provided with a space allowance corresponding to those in the regulatory proposal (0.98m^2/100kg, Grigor et al., 2001). Several studies of young and adult cattle suggest that this position, as well as a position perpendicular to the direction of the road, is preferred by these animals and enables them to maintain their balance more easily than diagonal positions (Grigor et al., 2001; Tarrant et al., 1992).

Last, several studies found that calves transported at space allowances in line with current regulations spent more time resting on arrival (0.79m^2/100kg for 38kg calves and 0.30m^2/100kg for heavy 234kg calves) compared with calves transported at higher space allowances (1.32m^2/100kg for 38kg calves and 0.41m^2/100kg for heavy 234kg calves, Grigor et al., 2004[15]; Jongman & Butler, 2014). The authors attributed this increase in resting time on arrival to greater fatigue in the calves transported at the current regulatory space allowances.

In conclusion:

The study of calf behaviour during transport suggests that space allowances corresponding to those in the regulatory proposal would enable calves i) to adopt a position that allows them to maintain better balance, ii) to lie down, particularly when supplied with straw bedding and iii) to become less generally fatigued, all of which would help to limit falls and potential injuries.

4.1.1.2 Young and adult cattle

With regard to young and adult cattle, the studies in the corpus observed several types of behaviour that make it possible to fill in gaps in the analysis of falls and injuries during transport, in particular interactions between conspecifics (sexual interactions and/or

[13] Descriptive analysis only
[14] Only in the presence of straw bedding, in Jongman & Butler (2014)
[15] Descriptive analysis only

aggression), changes in position and/or orientation within the vehicle and, last, losses of balance.

Sexual interactions between conspecifics can be a source of excitement inside the vehicle, and therefore of potential falls and/or injuries. Two studies by Tarrant et al. (1988, 1992) on heavy cattle (over 600kg) concluded that mounting behaviours were infrequent during transport regardless of space allowance and observed no significant differences in the number of mounts at the different experimental space allowances (between $0.5m^2$/100kg, $0.33m^2$/100kg and $0.17m^2$/100kg respectively, and between [0.22 - $0.23m^2$/100kg], [0.19 - $0.2m^2$/100kg] and [0.17 - $0.18m^2$/100kg]). These authors nevertheless recorded an increase in typical pre-mount behaviour (resting the chin on the hindquarters of a conspecific) between a space allowance of $0.33m^2$/100kg and a space allowance of $0.5m^2$/100kg (respectively 3 and 10 for the first journeys and 8 and 13 for the second, (χ^2, $p < 0.01$)) (Tarrant et al., 1988). Conversely, a study of grazers (around 400kg) noted a significant increase in mounting behaviour in animals transported at $0.34m^2$/100kg compared with those transported at $0.29m^2$/100kg (+ 1300% before the break, Dunnet test $p = 0.004$; and +757% after the break, Dunnet test $p = 0.003$) (Mounaix et al., 2009). It should be noted that, in this study, in order to be able to install cameras to observe the animals, the upper deck that would usually be present during transport had been removed. This certainly explains the differences in results between this study and those of Tarrant et al. (1988, 1992). Under conventional transport conditions, the height of the compartments prevents the cattle from overlapping.

With regard to **aggressive behaviour**, contradictory results have been observed in the literature. Two studies in the corpus concerned the transport of the same type of cattle (male grazers[16] weighing around 400kg) at similar space allowances (0.35 and $0.34m^2$/100kg versus $0.29m^2$/100kg). Of these, one observed a significant increase in the number of aggressions with the increase in space allowance[17] (+ 250%, Dunnet test $p = 0.04$; Mounaix et al, 2009), while the other observed no significant difference in this parameter (Eldridge & Winfield, 1988). Breed could possibly play a role in these differences since the first study used Charolais grazers while the second studied Hereford grazers. For heavy cattle (over 600kg), one study showed a greater number of aggressive behaviours and horn blows in cattle transported at $0.5m^2$/100kg compared with those transported at $0.33m^2$/100kg (aggressive behaviours: 45 versus 30 for the first set of journeys and 15 versus 10 for the second, (χ^2, $p < 0.01$) / horn blows: 46 versus 25 for the first set of journeys and 27 versus 24 for the second, (χ^2, $p < 0.01$)). The authors noted, however, that aggressive behaviours remained relatively infrequent overall on all routes (Tarrant et al., 1988). **It would therefore seem that an increase in the space allowance during transport may increase the aggressive behaviour of adult cattle and hence the risk of injury, or even falls, in certain contexts which have yet to be determined**.

[16] In the study by (Mounaix et al., 2009), one of the three journeys was made carrying Limousin/Charolais crossbred females because the Charolais males were not available for this journey.
[17] Only before the break, i.e. during the first 14 hours of travel.

Animal orientation was used as a parameter by certain authors to analyse the preferred positions of the cattle and to estimate possible discomfort or even loss of balance related to restrictions on freedom of movement and forced positions where the space allowance was limited. **It would appear that, whenever possible, cattle prefer positions perpendicular and parallel to the direction of travel, thus avoiding diagonal positioning** (Eldridge & Winfield, 1988; Mounaix et al., 2009; Tarrant et al., 1992). Several articles highlighted **a significant reduction in freedom of movement and orientation at space allowances corresponding to the regulations or lower** (reduction in movements of more than one metre below $0.29m^2$/100kg (Eldridge & Winfield, 1988); 36% reduction in changes of position, 166% reduction in free movements and 45% increase in constrained movements at $0.29m^2$/100kg (Mounaix et al., 2009); less freedom in the choice of orientation below $0.18m^2$/100kg (Tarrant et al., 1992)). However, one study showed an increase in diagonal positions at a space allowance of $0.35m^2$/100kg compared with an allowance of $0.29m^2$/100kg during the first 14 hours of the journey (before the break). After the break, the same authors found no significant difference in the orientation of the cattle between the space allowances, due to the greater number of animals lying down (Mounaix et al., 2009).

Last, loss of balance was studied as an indicator of the risk of falls. Once again, **the literature is divided on this indicator**, with one study observing no significant difference in the number of losses of balance in heavy cattle between surface areas of 0.5 and $0.33m^2$/100kg. Nevertheless, at these two surface areas, cattle lost their balance significantly less than at a space allowance of $0.17m^2$/100kg (Tarrant et al., 1988). Conversely, a study of grazers showed a significant 145% increase in losses of balance in cattle at a space allowance of $0.34m^2$/100kg, compared with an allowance of $0.29m^2$/100kg during the first 14 hours of the journey (before the break). After the break, as with the previous indicator, these authors found no significant difference between the space allowances, due to the greater number of animals lying down (Mounaix et al., 2009).

In conclusion:

Studies of the behaviour of young and adult cattle during transport suggest that an increase in space allowance (particularly above $0.29m^2$/100kg of cattle) enables the animals to move around the lorry better and gives them greater freedom of movement. On the other hand, this could be a source of increased sexual behaviour such as mounting and overlapping if the height of the compartment allows, and of aggressive behaviours that could lead to falls and/or injuries. These potential consequences have yet to be confirmed, as there is no consensus in the literature. In particular, these consequences should be studied for space allowances corresponding to those in the regulatory proposal. Last, the only study to compare a space allowance corresponding to those in the regulatory proposal with a space allowance in line with current regulations found no significant difference in the number of losses of balance in the cattle transported.

4.1.2 Physiological responses

In the sections that follow, only studies comparing space allowances above the current regulatory threshold are considered. A description of the physiological measurements analysed in the literature can be found in *Appendix 1.*

4.1.2.1 Calves

In addition to CK concentrations, which were discussed in *3.2.1*, many physiological parameters have been analysed to assess possible stress in calves during transport, including plasma concentrations of glucose, cortisol, beta-hydroxy-butyrate (BHB), aspartate aminotransferase (AST), non-esterified fatty acids (NEFA), etc.

With the exception of a single article on calves weighing around 40kg indicated higher post-transport plasma glucose concentrations in calves transported at a space allowance of 0.5m^2/100kg compared with those transported at a space allowance of 1m^2/100kg (Todd et al, 2000), no study in the literature has observed any significant difference in physiological responses in calves between the different experimental space allowances (Earley & O'Riordan, 2006; Grigor et al., 2004; Jongman & Butler, 2014). Todd et al (2000) attributed the higher plasma glucose concentrations they observed at the lowest space allowance to greater muscular activity in the calves under these conditions. They linked this muscular activity to the calves' struggles to counteract the movements of the vehicle, which would not be necessary in calves transported at the higher allowance as the latter would be able to lie down.

Authors whose results suggested no significant difference in the physiological responses of calves to different space allowances during transport have questioned their own analytical methods (blood was sampled after transport, sometimes without specifying the time lapse between unloading and sampling) or their choice of space allowances for comparison (the space allowances being potentially too similar to observe a significant difference between treatments).

In conclusion:

Analysis (single study only) of the physiological responses of calves during transport suggests that an increase in the space allowance corresponding to the space allowances in the regulatory proposal would reduce muscular activity and even fatigue in calves compared with the current regulatory space allowances. This increase in space allowance would not, a priori, have any negative consequences in terms of potential stress or muscular fatigue in calves, since those studies that do not demonstrate any benefit from an increase in space allowance observe no significant negative impact either.

4.1.2.2 Young and adult cattle

Of the articles in the corpus, only two compared the physiological reactions of young and adult cattle at more than one experimental space allowance higher than the current regulatory threshold:

- Mounaix et al. (2009) compared grazers weighing around 400kg transported at space allowances of 0.29 and 0.34m^2/100kg (both allowances below the regulatory proposal threshold) for 29 hours.
- Tarrant et al. (1988) compared cattle weighing around 600kg transported on space allowances of 0.33m^2/100kg (allowance below the regulatory proposal threshold) and 0.5m^2/100kg (allowance above the regulatory proposal threshold) for 4 hours.

Mounaix et al. observed no significant difference in physiological parameters (blood concentrations of CK, AST, glucose, urea, alanine aminotransferase (ALAT), haptoglobin, etc.) between the different experimental space allowances. The concentrations measured in the animals were within normal ranges, with the exception of CK, urea, glucose and haptoglobin, for which concentrations were above normal ranges at all experimental space allowances. The authors also noted an increase in CK concentrations after transport in cattle transported at the lowest allowance (654.79 U/L at 0.29m^2/100kg, compared with 544.64 U/L at 0.34m^2/100kg with an a priori normal range of 66-120 U/L), although this difference was not significant. The authors associated these high values (CK, urea, glucose and haptoglobin) with a state of stress and moderate and reversible muscle fatigue linked to transport.

Tarrant and colleagues observed a significant decrease in plasma cortisol and plasma glucose concentrations at the highest transport space allowance (15.7 ng/mL and 5.24 mmol/L respectively at 0.5m^2/100kg, versus 30.3 ng/mL and 5.81 mmol/L at 0.33m^2/100kg; $p < 0.001$). They observed similar plasma CK activity at these two space allowances (34 U/L at 0.5m^2/100kg and 32 U/L at 0.33m^2/100kg), but this was nevertheless significantly lower than that in cattle transported at a space allowance of 0.17m^2/100kg (allowance below the current regulatory threshold, with a CK value of 200 U/L). On the basis of these results, the authors concluded that a reduction in space allowance during transport increases the stress response in cattle.

It should be noted that, in view of the CK values observed in the two studies cited, it can be assumed that cattle require space allowances in excess of 0.34m^2/100kg to achieve a significant reduction in plasma CK activity.

In conclusion:

As for calves, analysis of the physiological responses of young and adult cattle during transport suggests that an increase in space allowance corresponding to those in the regulatory proposal would reduce cattle stress compared with the current regulatory allowances. This increase in space allowance would not a priori have any negative consequences in terms of potential stress. However, further studies are needed to confirm these results.

4.2 Factors increasing the risk of falls and/or injuries during the transport of cattle

4.2.1 Factors potentially increasing the risk of falls and/or injuries associated with the space allowance

4.2.1.1 Quality of driving

The careful driving of transport vehicles is essential in preventing falls and injuries to animals. Violent braking, rapid acceleration, speed changes and cornering sharply can lead to loss of balance, increasing the risk of falls, injuries and stress for the animals (Meat Institute Animal Welfare Committee, 2024; Tarrant et al., 1988, 1992).

According to the EFSA (2022), **some debate has existed in the literature as to whether a lower space allowance could benefit cattle in the event of rough driving, due to 'mutual support' between closely spaced animals. In the light of current knowledge, this does not appear to be the case**. On the contrary, with a lower space allowance, cattle could be more likely to suffer from the "domino effect" (Strappini et al., 2009), making falls more likely to occur when nearby cattle lose their footing and animals find themselves unable to adjust their position.

4.2.1.2 The route

Routes that minimise the time spent on unsurfaced roads and avoid potholes would reduce the risk of falls and injuries regardless of the transport space allowance (Meat Institute Animal Welfare Committee, 2024; Mendonça et al., 2019). **As with the quality of driving, it may be assumed that this factor has a greater impact on the risk of loss of balance when the space allowance is greater, but there are no articles in the literature to support this argument**.

4.2.1.3 The design of transport vehicles

The **type of flooring and floor covering** used during transport can also have an impact on the number of falls and injuries suffered by cattle. Indeed, a floor that is poorly maintained or too smooth could increase the risk of slips and, consequently, of falls and injuries (Mendonça *et al.,* 2019). According to Mendonça et al., (2019), this risk could be reduced by the use of rubber mats. In addition, other authors who have analysed the respective impacts of the space allowance and type of flooring during transport have noted interactions between these two factors: **the negative consequences of a reduced space allowance (less than 0.79m²/100kg for calves) would be less pronounced with the addition of straw bedding, particularly for calves** (a higher percentage of calves lie down, CK activity and BHB concentrations after transport are reduced) (Jongman & Butler, 2014).

Vehicle size and configuration can also influence falls and injuries. Long vehicles, with a greater distance between the animal compartment and traction points, generate more vibrations in the floor than do smaller lorries. Vibrations could be a source of discomfort and greater movement in cattle, potentially leading to loss of balance (Mendonça et al., 2019). Additionally, long and/or articulated lorries are subject to greater centrifugal force at their centres, increasing the risk of falls (Bethancourt-Garcia, Vaz, Vaz, Silva, et al., 2019; Mendonça et al., 2019).

Last, the location of the **vehicle compartment** in which cattle are transported could also play an important role in the risk of injuries and falls at certain space allowances. Indeed, the results reported by González et al (2012) suggest that **cattle transported in the "belly" and "deck" compartments (Figure 4) would be at greater risk of falling and injuring themselves when transported at a space allowance calculated using an allometric equation with a k value less than or equal to 0.015 or greater than 0.035**[18]. The authors nevertheless indicate that further study is needed to test this hypothesis.

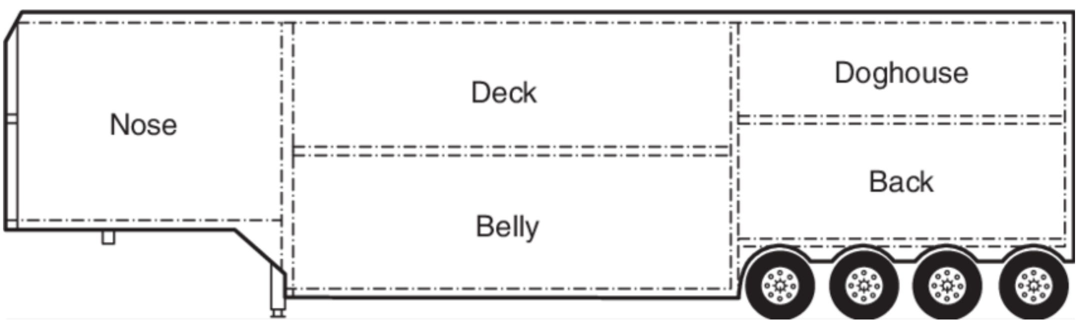

Figure 4. Example of compartment layout in a trailer (Schwartzkopf-Genswein & Grandin, 2014, adapted from an image by Merrit Equipment Co., Denver, Colorado)

[18] Calculation of the available surface area per 100kg is impossible given the variability in the weight of the animals considered in this study.

4.2.2 Other factors

Other factors that are a priori independent of the space allowance can influence the risk of injury or even falls during transport. These include factors intrinsic to cattle such as **breed**, **sex** and **age**, which influence the reactivity of animals and their response to different environmental conditions (Bethancourt-Garcia, Vaz, Vaz, Restle, et al, 2019; Bethancourt-Garcia, Vaz, Vaz, Silva, et al., 2019; Uetake et al., 2011), whether or not animals **are horned** (transporting horned cattle has been reported to present an increased risk of injury; Alende, 2010). Environmental factors that may increase the risk of injuries and/or falls include the **distance travelled** (the greater the distance, the greater the risk of injury; Bethancourt-Garcia, Vaz, Vaz, Silva, et al., 2019) and the **transport of cattle unfamiliar to each other** (transporting cattle from different farms may increase aggression between cattle due to social stress, thus increasing injuries during transport; Mendonça et al., 2019).

5 Conclusion and levers for action

5.1 Conclusion

With regard to the transport of calves, **the available literature highlights that the space allowances set out in the proposed regulation (European Commission, 2023) would enable calves to sustain fewer injuries than the current regulatory space allowances (Council of the European Union, 2004)**, in particular because these new space allowances would enable them to achieve better balance, lie down more (particularly when provided with of straw bedding), and generally become less fatigued. The literature on these factors suggests that calves are no more likely to fall at space allowances corresponding to the proposed regulation than at the current regulatory allowances. Furthermore, no additional stress has been demonstrated in calves transported at space allowances corresponding to the proposed regulations.

For young and adult cattle, a single article compares fall and injury numbers between a space allowance corresponding to the regulatory proposal and an allowance in line with the current regulations, for cattle weighing around 600kg. **According to this article, cattle are injured less at the new regulatory space allowances than at current allowances, and the number of falls does not differ between the two**. This would appear to be due to the fact that, as for calves, provision of space corresponding to the allowances in the regulatory proposal would allow cattle to adopt their preferred positions and hence maintain better balance during transport. More studies are required to confirm these results for the different categories of young and adult cattle. However, **at present, no studies suggest that cattle would not suffer greater falls and injuries at the space allowances set out in the regulatory proposal than at the current regulatory allowances**[19]. Indeed, the few articles to have observed greater losses of balance at higher space allowances during transport used experimental allowances lower than those set out in the proposed regulation. It is possible that, at these space allowances, the cattle did not have sufficient space to maintain optimal balance.

The results of this assessment are therefore in line with those set out in EFSA (2022), i.e., that a priori cattle would be less likely to be injured at the space allowances recommended in the proposed regulation than at those in the current regulation. In addition, as indicated in EFSA (2022), the reduction of transport densities offers other advantages: less exposure

[19] Question as asked: "Is it the case that [cattle] transported by road at the densities set out in the proposed regulation to revise Regulation No 1/2005 are more likely to fall and/or be injured than [cattle] transported by road at the densities laid down in the current regulation (Regulation No 1/2005)?"

to heat stress[20] and improved resting capacity, both of which are essential factors for animal welfare during transport.

Last, it should be noted that it is essential to consider other parameters in order to minimise falls and injuries during transport. These include the quality of driving, the route taken, the design of vehicles, the presence and type of bedding, and the duration and distance of the journey. A priori, these factors do not appear to contribute to increased falls and injuries when the space allowance is increased; the opposite is more widely observed in the literature. Nevertheless, it would appear important to carry out a more detailed assessment of the interactions between these factors and space allowances for cattle during transport.

5.2 Actions to mitigate the risk of injuries and falls during transport

For researchers

Given the limited literature available to answer the question asked, **it is essential to continue research into falls and injuries in cattle at transport space allowances/loading densities corresponding to the regulatory proposal compared with current transport space allowances/loading densities**. In particular, it would be appropriate to conduct trials involving more behavioural analysis (e.g. via video recordings) to allow direct quantification of the number of falls, and identify the extent to which the injuries observed are linked to falls in the course of the journey. It would also be of interest to model the risk of falls and injuries at different densities for each category of cattle, incorporating co-factors such as quality of driving or journey duration.

In addition, a meta-analysis is needed to provide a more precise assessment of the methodologies used in the various studies referred to in this report, in order to establish the reliability of the results they report.

For training organisations

To guarantee the protection of animals during transport, the factors that cause falls and/or injuries in cattle must be controlled. It is therefore essential to include good cattle handling practices (calm approach, safe movements, etc.), the recognition of behavioural indicators of stress, and good road driving practices (progressive braking, anticipation of bends, etc.) in livestock driver training courses.

For livestock transport companies

In order to minimise animal falls and injuries, it is essential that drivers take particular care in driving, in particular by avoiding sudden acceleration and braking and anticipating bends

[20] Detailed analysis in another report from the FRCAW (CNR BEA, 2025)

in order to limit the centrifugal force exerted. Experienced drivers could also be given preference for the transport of live animals. The routes taken should be optimised to avoid unsurfaced and twisting roads as much as possible.

The design of the vehicles used to transport animals should be optimised, with air suspension systems designed to reduce shocks, anti-slip flooring, and modular internal partitions to allow animals to be separated by size, age or category (young or pregnant animals, etc.). The use of straw bedding should be encouraged. In addition, the condition of vehicles (brakes, tyres, ventilation system) should be checked before each journey (if this has not already been carried out) to reduce sudden movements and ensure safer, more comfortable transport for the animals.

Last, the loading density should be adjusted to suit the category, sex, breed and origin of the animals, the journey duration, and the weather, in order to reduce falls, crushing and trampling, and to limit stress.

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Appendix 1. Definitions of physiological indicators used in the assessment of stress and injury during cattle transport

Term	Definition	Use for stress or injury assessment	Sampling
ALAT (Alanine Aminotransferase)	An enzyme mainly found in the liver, measured in blood to detect liver damage.	Elevated levels of ALT may indicate liver damage or metabolic stress affecting the liver.	Blood
Albumin	The main plasma protein, essential for maintaining oncotic pressure and transporting various molecules.	A drop in albumin can be observed in cases of chronic inflammation or prolonged stress, affecting nutritional status and liver function.	Blood
AST (Aspartate Aminotransferase)	Enzyme present in various tissues (liver, muscles, heart), used to detect tissue damage.	An increase in AST can indicate muscle or liver damage, often associated with physiological stress or trauma.	Blood
BOHB/ BHB (Beta-hydroxybutyrate)	A ketone produced by the liver when fat is broken down, particularly when carbohydrate intake is insufficient, indicating that the body is drawing on its energy reserves. Measuring the level of BOHB in the blood enables us to assess the overall energy state and determine whether the body is in ketosis, i.e. a metabolic state in which fats are mainly mobilised to provide energy.	A rise in BOHB can indicate an energy imbalance (ketosis) often associated with metabolic stress, particularly in ruminants.	Blood
CK (Creatine Kinase)	Enzyme present in muscles and other tissues, involved in energy production.	Elevated CK often indicates muscle damage or intense exercise, which can be induced by severe physical stress.	Blood
Cortisol	Cortisol is a steroid hormone produced by the cortex of the adrenal glands that regulates metabolism, inflammation and the immune response. It is secreted in response to activation of the hypothalamic-pituitary-adrenal axis during stressful situations.	In stressful situations, increased cortisol rapidly mobilises the energy needed to activate the 'fight or flight' response. However, prolonged exposure to high levels of cortisol can upset the physiological balance and adversely affect overall health.	Blood but also saliva or hair
Fibrinogen	A protein in blood plasma that turns into fibrin during coagulation.	Their increase can be a marker of an inflammatory reaction or acute stress, particularly during infections or trauma.	Blood (plasma)
Glucose	A simple sugar molecule present in the blood and serving as a source of energy for the cells.	In times of stress, the release of hormones (adrenalin, cortisol) can lead to hyperglycaemia, indicating increased energy mobilisation.	Blood
Haptoglobin	An acute-phase protein that binds to haemoglobin released when red blood cells are lysed.	Its increase is an indicator of inflammation or acute stress, often in response to infection or trauma.	Blood
Haematocrit	Ratio of blood cell volume to total blood volume. It is obtained by centrifugation, which allows the separation of blood cells and plasma.	An increase in haematocrit may reflect haemoconcentration, often due to dehydration or physiological stress.	Blood
Haematology	Study of the cellular components of blood (red blood cells, white blood cells, platelets) and their proportions.	Changes in haematological parameters (e.g. leucocytosis or changes in red blood cell count) may be responses to stress.	Blood
Haemoglobin	Red hetero protein present in red blood cells	An increase in haemoglobin levels may be associated with stress or dehydration.	Blood
IgM (Immunoglobulin M)	First class of antibodies produced during an immune response, essential for the initial defence against infections.	Changes in IgM levels can reflect the state of the immune response, often affected by chronic or acute stress.	Blood (serum)
Lactate	Salt of lactic acid, often measured to assess the intensity of anaerobic glycolysis.	High levels of lactate indicate recourse to anaerobic glycolysis, typical of intense physical effort or metabolic stress.	Blood and muscle
Lymphocyte	A mononuclear blood cell classified according to its diameter into large (9 to 15 µm) and small (6 to 9 µm) lymphocytes which play a fundamental role in the body's immunological response, and are generally found in the circulatory system and in the "lymphoid organs" (lymph nodes, spleen, thymus).	A decrease in the percentage of lymphocytes is often interpreted as a sign of chronic or acute stress.	Blood
NEFA (Non-Esterified Fatty Acids)	Free fatty acids circulating in the blood, indicators of the mobilisation of lipid reserves.	An increase in NEFA suggests increased mobilisation of fats in response to an energy deficit, often observed in situations of prolonged stress.	Blood (plasma)

Term	Definition	Use for stress or injury assessment	Sampling
NEFA (non-esterified fatty acids)	Fraction of free fatty acids circulating in blood	High levels of post-transport NEFA indicate increased mobilisation of fat reserves, which is often associated with a state of metabolic stress and increased energy demand.	Blood
Neutrophil	Effector cells of innate immunity, involved in particular in the inflammatory response	An increase, both in percentage and absolute number, may reflect a stress response or transport-induced inflammation.	Blood
Noradrenaline	A precursor neuromediator of adrenalin, secreted by the adrenal medulla.	Its release, like that of adrenaline, marks the activation of the sympathetic system during acute stress.	Blood
PCV (Packed Cell Volume)	Packed Cell Volume or haematocrit: percentage of blood volume occupied by red blood cells.	Variations in PCV may reflect hydric changes (dehydration or overhydration) or stress responses (changes in blood volume).	Blood (haematocrit)
Proteins	All plasma proteins such as albumin and globulin.	Variations (increase or decrease) may reflect nutritional imbalances, dehydration or a stress-related inflammatory reaction.	Blood
RBC (Red blood cell count)	This indicator corresponds to the number of red blood cells per unit of blood volume.	An increase in RBC count in transported animals may indicate an adaptive response to stress or increased blood concentration due to dehydration.	Blood
Red blood cells	A less common term, it generally refers to the red blood cells (erythrocytes) responsible for transporting oxygen.	A change in the number of erythrocytes can be linked to states of stress (for example, in the case of haemoconcentration due to dehydration).	Blood
Heart rate (bpm)	Number of heartbeats per minute.	An increase in heart rate is a physiological response to stress (activation of the sympathetic system).	Measured using devices
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