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A pilot study to develop an assessment tool for sheep welfare after long journey transport

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Abstract

Sheep transport within Europe involves 9.5 million animals yearly, 63% of which travel over long journeys (> 8 h). Livestock transport, particularly over long journeys, gives rise to concern about the welfare of transported animals. The European Commission stimulates the development of market-oriented animal welfare standards for all phases of livestock production, providing an alternative to the 'regulatory approach'. This study aimed to develop and test a new sheep welfare assessment protocol to be used following transport, irrespective of the journey purpose. The protocol included outcome (animal-based measures) and input variables (resource-based and management-based measures), being welfare-relevant aspects of both transport and unloading procedures. Weighted Cohen's Kappa and Fleiss' Kappa index of agreement were calculated to evaluate the raters accuracy and the inter-observer reliability. Overall, good agreement levels were found. The protocol was tested on 40 commercial transports arriving at previously selected assembly centres and slaughterhouses in Italy and Greece. The protocol was found to be feasible when applied to commercial transports, allowing for a comprehensive and quick sheep welfare assessment during unloading, without impairing stockman work. Univariate analysis was carried out to evaluate associations between outcome and input variables. In this study, significant association between outcome measures and risk factors were identified when associated to unloading procedures but not to travel conditions. In collaboration with the relevant stakeholders, this protocol might be developed into a tool for routine checks for certification purposes and could provide direct feedback to all professionals involved in animal transportation on the weaknesses and strengths of their work.

Keywords: *animal welfare, long journey, sheep, transport, welfare assessment, welfare indicators*

Introduction

Road transport of live animals is a large and integral part of today's livestock industry, with the vast majority of animals being transported at least once during their production life. Across European countries, transportation distances have increased in recent decades, mainly due to the removal of customs barriers allowing animals to be sold across larger areas (Gavinelli *et al* 2008). Sheep transport within Europe involves 9.5 million animals every year, 63% of which are transported for more than 8 h (Gebresenbet *et al* 2010). A recent survey identified Italy as the main country of import, accounting for 50–60% of the overall sheep import, followed by Greece (7–13%), whilst the main exporters were eastern European countries (Romania, Poland and Hungary) and Spain (Gebresenbet *et al* 2010). The same study highlighted two main routes for sheep travelling long journeys across the EU: from Poland, Hungary or Romania to Italy and from Spain, Hungary or Romania to Greece.

The process of transportation, including handling, loading, transporting and unloading can have a large impact on the welfare of transported animals, especially if it involves long journeys (Broom 2005; Gavinelli *et al* 2008). The EC Regulation 1/2005 on the protection of animals during transport provides for special requirements for all journeys lasting more than 8 h ('long journeys') (European Council 2004). These requirements aim to minimise any possible negative impacts upon the welfare of transported animals (European Council 2004). Nevertheless, the concern regarding welfare issues associated with animal transport is continually increasing, eg the '8 h' campaign, supported by several non-governmental organisations and by some European Parliament members, proposing an absolute limit of 8 h on all road journeys for animals transported for slaughter (European Parliament 2012). Accordingly, further research is required to develop feasible and reliable tools to ascertain at what level the welfare of animals during long journeys is ensured.

Veissier *et al* (2008) stated that the development of an animal-friendly products' market in Europe is likely to stimulate further developments in animal welfare standards, providing an alternative to the 'regulatory approach' of setting minimum welfare standards through legislation. The development of an easy-to-use tool which allows field evaluation of animal welfare is a fundamental prerequisite for the implementation of such a scheme. Several scientific projects have been conducted on this issue for livestock welfare, with a focus on farm conditions (eg the Welfare Quality® project; Blokhuis *et al* 2010). Some studies were also performed on transport (Maria *et al* 2004; Brown *et al* 2005; Marlin *et al* 2011) but, at present, none provide a feasible protocol to be used to assess the welfare of sheep and lambs after transport over long distances.

The aim of this pilot study was to develop and test the suitability of a newly developed tool for the assessment of sheep and lamb welfare after commercial transport over long distances. Data collected from animals after commercial journeys were used for an exploratory analysis to identify welfare hazards and to assess the current state of live sheep transport in the EU.

Materials and methods

Protocol description

The present protocol was developed to be tested during commercial transports, irrespective of whether the endpoint of the transportation was for slaughter or for breeding of the animals. The protocol was tested under practical conditions and the journeys investigated were often identified just before arrival. This precluded conducting observations on the animals prior to their journey. As a consequence, all animals came from unknown rearing systems and had started their journey with an unknown level of fitness. One of the primary requirements for feasibility of the protocol was for minimal interference of the assessment procedures with the transporters and other operators' work. Furthermore, as previous welfare assessment schemes have often been criticised for being too time consuming for commercial application (Andreasen *et al* 2014), this protocol was designed to be completed solely at the time of the arrival of the truck at the destination. It was also necessary for the assessment to be carried out by recording measures from a distance and without slowing down normal unloading procedures. The assessment was divided into three different phases: before, during and after unloading. On arrival, but before unloading started, the assessor recorded the first set of measures by observing the truck from the outside (Phase 1; P1). The assessment of sheep unloading started when the doors of the truck were opened and the first animal was led towards the ramp, and ended when the last animal had crossed an imaginary line located 3 m after the end of the ramp (Phase 2; P2). Once unloading was completed, a final set of measures was collected on the empty truck and at the resting pens where animals were held (Phase 3; P3). In all cases, the resting pens consisted of a covered, bedded area with feed and water available to the animals. Since the aim of the present work was to evaluate the welfare of sheep and lambs after long journey transportation, irrespective of the housing condition at destination, no further details about these were recorded. Finally, data from the truck driver were acquired to record technical information on the journey itself.

Selection of measures

Factors affecting farm animal welfare include the physical environment and resource availability (Resource-Based Measures; RBM) as well as the management practices employed (Management-Based Measures; MBM). Animal responses to the resource and management inputs depend on animal characteristics (breed, sex, age, etc) and can be assessed directly from the animals, using Animal-Based Measures (ABM) (EFSA 2012). Nevertheless, for some measurements, such as thirst, the available ABM are unsuitable for field application (eg skin test), being too time consuming or requiring specific equipment or expertise to be collected (Keeling 2009). Hence, the collection of MBM and RBM are necessary, in addition to ABM.

The measures were also selected on the basis of their feasibility, ie how practical they are to carry out in different field conditions, including recording time and repeatability.

RBM were recorded during P1 and P3 while MBM and ABM were mainly assessed during P2. Since sheep moved very quickly off the truck, and it was not possible to record all measures at once, some ABM parameters were assessed during P3 on a randomised sample of the unloaded animals (30% of the total on the truck) 20 min after the unloading, once the sheep were all collected in the resting pens.

The selection and definitions of MBM and RBM included in the protocol were mainly derived from the provisions of the EC Regulation 1/2005 (European Council 2004) and from the EFSA Scientific Opinion concerning the welfare of animals during transport (EFSA 2011). The list of selected RBM and MBM can be found in Tables 1 and 2, respectively.

ABM were defined on the basis of publications from the Welfare Quality® project (Blokhuis *et al* 2010). Tables 3 and 4 present the list of selected ABM assessed during P2 and P3, respectively.

The scoring for ABM and MBM included counting the events (eg number of animals falling during unloading) or recording the presence or absence of an event (eg handlers slapping/not slapping animals). The scoring for RBM was binomial (Y/N) or trinomial (total/partial/absent) for most measures, or it required taking surface measures (eg ramp slope) or counting items (eg number of drinkers). All ABM were scored at the individual level, with the exception of slipping, reluctance to move and coughing. This was due to the large number of animals unloaded together in a relatively short space of time, making individual assessment of these measures impossible. Therefore, slipping and reluctance to move were scored at group, ie flock level. A flock was defined as all the animals present in a compartment (ie each sub-section of a deck separated from the rest of the deck by a partition) of the truck being unloaded together. Coughing was assessed at truck level (ie no animals

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Table 1 Resource-based measures for sheep (both P1 and P3).

Measure	Description
Deck height	EC Reg 1/2005 provides that during transport compartments allow ventilation above the animals when they are in a naturally standing position. Record the deck height as adequate if there is a space (min 15 cm) above the heads of the standing animals
Ramp covering	EC Reg 1/2005 provides that ramps are safe for the unloading. Ramp covering prevents light reflection, deadening sounds and reducing the slippery areas. Indicate the type of bedding on the ramp (ie straw, wood-shaving, sand) and the quantity (ie ramp totally covered/partially covered so that it is possible to see the ramp surface/none)
Ramp flooring	Indicate the type of ramp flooring (ie smooth metal, corrugated metal)
Ramp floor conditions	Indicate the presence/absence of harmful surfaces (eg holes, missing battens, sharp edges)
Ramp lateral protections	EC Reg 1/2005 provides that ramps are equipped with lateral protections to prevent animals from escaping. Record the presence of the ramp lateral protections
Gaps or steps	Step and gap are any height difference of more than 10 cm between two surfaces. Indicate the presence of any gap or step being among lorry/ramp/floor or lorry door/lateral protection
Ramp slope	Record the height of the ramp to calculate the slope
Foot battens	Indicate the presence of foot battens on the ramp
Blocking zones	Indicate the presence of any blocking zone (ie shadows or obstacles) on the ramp
Lighting for orientation	Indicate the presence of sufficient lighting to allow the animals to orientate (ie even, diffuse and lightening the ramp up ahead so that the animals are moving from a darker area to a more brightly lit one), and that is not reflecting on the ramp or directly orientated toward the unloading animals
Lighting for handling	Indicate the presence of adequate lighting (ie allowing the assessor to read the scoring sheet) at unloading for handling purposes
Sharp edges in the truck	Indicate the presence of sharp or harmful edges inside the truck
Truck bedding	EC Reg 1/2005 provides that animals are safe during transport. Bedding provides a more comfortable resting surface; it helps absorb urine and faeces, and reduces the risk of slipping. Indicate the type of bedding on the truck (ie straw, wood-shaving, sand) and its quantity (ie totally covers the deck/partially covers the deck so that it is possible to see the ramp surface/none)
Truck hygiene	The decks should be kept clean, minimising the leakage of urine or faeces. Indicate if the amount of manure present covers less or more than 25% of the deck surface
Water supply: drinkers	Indicate the number and type (ie nipples, bowls) of drinkers and if they are functioning
Water supply: tank	Record the amount of water on the truck (ie full/empty/nor full nor empty)
Temperature monitoring and control systems	Record the presence of the temperature monitoring and control system in the truck and and if it is functioning

coughing/at least two animals on the truck coughing). All MBM were assessed at flock level.

Information about the transport itself was collected from the journey log (eg origin and destination, date and hour of departure, number of animals, type of truck), together with the sheep weight category according to Regulation 1/2005 (ie shorn or unshorn/below or over 55 kg) and the temperature at the time of unloading. These data were used to calculate travel duration, distance covered by the truck and space allowance.

Training of the assessors

The measures included in the protocol were designed to be simple and descriptive. Nevertheless, it was necessary to investigate whether systematic errors in interpretation by multiple observers were present (Whay 2007). Research on inter-observer agreement for animal-based measures confirm that a high Kappa index or correlation between observers can be reached through training (Kristensen *et al* 2006; March *et al* 2007; Laister *et al* 2009). Therefore, training of the three observers was conducted to obtain repeatable assessments and to ensure uniform interpretation of the protocols.

Video and photographic material were collected during two commercial transports and scored by a team of veterinarians experienced in animal welfare assessment (ie having received dedicated training in livestock animal welfare assessment and having several years of experience in animal welfare research). This material was then used to train the assessors and to prepare a series of 'gold standard' assessments to refer to. Three assessors (two veterinarians and one ethologist) were trained in a two-day course. The first day was dedicated to classroom training, where the protocol was illustrated and scoring systems explained through the use of video and photographic material. On the second day repeatability assessments were performed, both in the classroom using multimedia, and through field exercises in which the protocol was practiced on commercial transports at an assembly centre.

Table 2 Management-based measures for sheep in P2.

Measure	Description
Handler moving excitedly*	Assess whether or not the handler is moving the animals excitedly (ie arm-waving, making quick or sudden changes of direction)
Handler making loud noises*	Assess whether or not the handler is making loud noises (ie shouting or clanging and banging metal equipment together or on the truck/ramp sides) to induce the animals to move
Handler slapping*	Assess whether or not the handler is slapping the animal without reasons or to make them move when they are not in the condition to do it
Appropriate handling*	Assess whether or not the handler gives the sheep the opportunity to slow down and inspect start of the ramp and if the handler is able to control the speed of movement
Handler performing forbidden practice*	Assess whether or not the handler is performing forbidden practices, according to Regulation EC 1/2005: • hit the animals or kick them; • press sensible areas to cause unnecessary pain; • lift animals with mechanical devices; • lift or pull animals by the head, the ears, the horns, the legs, the tail or the fleece; • use sharp devices; • tie animals by the horns, using nose devices or tie their legs together; • use instruments which administer electric shocks

* Group assessment.

Table 3 Animal-based measures for sheep at P2.

Measure	Description
Dead on arrival	Record the number of animals found dead on arrival, carefully inspecting the truck during unloading operations
Non-ambulatory	Record the number of animals that cannot rise or are unable to walk unaided
Slipping*	Measure taken at flock level (0 to 1 animal slipping/more than 1 animal slipping within a flock). A slip is defined as occurring when an animal shows a loss of balance during unloading without a non-limbic part of the body touching the ground
Falling	Record the number of animals showing a loss of balance during unloading causing other part(s) of the body (beside legs) to touch the floor
Reluctance to move*	Record the number of flocks where animals show unwillingness to go forward or suddenly stopping just before the beginning or during the unloading for at least 3 s
Lameness	Record the number of animals showing lameness. Lameness describes an abnormality of movement and can vary in severity from reduced mobility to inability to bear weight. Only animals showing signs of severe lameness (ie marked flicking of the head while moving, reluctance to bear weight on a foot, difficulty in rising up, reluctance to move when standing) are to be recorded
Injury	Record the number of injured animals. To assess injuries, a technique inspired by the one developed by Jørgensen *et al* (2009) is to be used. For the purpose of this protocol, an animal is to be considered injured only if presenting unhealed injuries scoring 4 to 5 in the above mentioned reference, that is to say: • A wound through the skin which involves damage to deeper tissue (muscles, tendons) or a cut through the skin which is so big that it would normally be stitched; • An extensive and serious injury that may cause loss of function over a long period of time, eg (serious damage to a tendon or joint, fracture)
Coughing*	Record the presence or absence of coughing events on the truck. Coughing is defined as a sudden and noisy expulsion of air from the lungs

* Group assessment.

Table 4 Animal-based measures for sheep in P3.

Measure	Description
Dead in pen	Record the number of animals found dead in the resting pen (20 min after arrival)
Hampered respiration	Record the number of animals presenting dyspnoea: respiration is deep and overtly difficult and expiration is supported by the muscles of the trunk, mostly accompanied by pronounced sound. Breathing rate may only be slightly increased
Exhaustion	Record the number of animals showing severe fatigue or exhaustion (ie chin or limbs resting against partitions or troughs, closed eyes, high drive to rest in recumbent position)
High respiratory rate (HH)	Record the number of animals presenting polypnoea: breathing in short gasps carried out with the mouth and with increased frequency

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Observer agreement evaluation

To quantify the level of accuracy (agreement of the observer with the 'gold standard') and inter-observer reliability (agreement between two or more independent assessors), a final test was performed by the observers. This test consisted of showing a number of videos (n = 20) to the participants, who were then asked to score the selected measures.

To evaluate accuracy, each assessor was compared to the 'gold standard' by calculating a weighted Kappa value (Cohen 1968). To evaluate the inter-observer reliability, Fleiss' Kappa index of agreement between raters was computed (Fleiss 1971). According to Landis and Kock (1977), agreement levels for Kappa values are as follows: 0.00, less than chance agreement; 0.01–0.20, slight agreement; 0.21–0.40, fair agreement; 0.41–0.60, moderate agreement; 0.61–0.80, substantial agreement; and 0.81–0.99, almost perfect agreement; 1, perfect agreement. For all analyses, a *P*-value was given indicating whether agreement was more than could be expected by chance alone ($\alpha \leq 0.016$, applying Bonferroni correction).

Four ABM were tested for repeatability. The level of accuracy of each assessor with the 'gold standard' was high, ranging between substantial to perfect agreement for all comparisons ($k > 0.61$). Inter-observer reliability was perfect, as expected, for the measure 'dead on arrival' ($k = 1$). It was almost perfect for two other measures: reluctance to move ($k = 0.85$) and slipping ($k = 0.84$), and was substantial for the last one 'falling' ($k = 0.70$). The reliability was thus considered acceptable for all these measures.

Initially, all ABM were to be included in the training exercises. However, the material collected for training purposes was found to be too homogenous for some of the measures, which did not allow for statistical assessment of reliability for these measures (eg non-ambulatory animals).

Welfare assessment of commercial transports

Field assessment

Forty assessments were carried out between October 2012 and June 2013. Agreements were made with the veterinary services in both central Italy and Greece to be kept informed of any ovine transports travelling over long journeys (trips > 8 h). Assessments were carried out with the authorisation of the food business operator at the place of arrival of the truck. No additional selection criteria were applied and all transports which the research team were informed of, within the aforementioned time-frame, were assessed. Of this convenient sample, 15 evaluations were carried out in Greece (one at control post, six at slaughterhouses and six at farms) and 25 in Italy (20 at slaughterhouses and five at assembly centres). Once informed of a suitable transport arrival to be assessed, the assessors ensured they arrived at the destination prior to the appearance of the truck. During assessments, assessors dressed in dark clothing and all observations were performed from a position which afforded a good view of the animals without interfering with their movements or those of the stockmen.

Statistical analysis

A descriptive exploratory analysis was carried out to summarise the main characteristics of the assessments performed.

Univariate analysis was carried out in order to examine associations between outcome welfare variables (ABM) and input factors (RBM and MBM) with the aim of facilitating the identification of animal welfare hazards for transported sheep.

It was decided to split the dataset into two groups. The first group of ABM provided information on the impact of the journey on the animals while the second group included measures mainly linked to the quality of the unloading from the truck. RBM and MBM recorded in the protocol were associated either to the travel group or the unloading group.

Transport-related measures included death, injured and non-ambulatory animals assessed upon truck arrival, and animals with signs of exhaustion, coughing, hampered respiration, high respiratory rate and death recorded in resting pens. RBM potentially affecting the quality of the journey were the following: travel duration, stocking density and deck height, drinker type and number of drinkers available per animal, quantity and type of bedding, truck hygiene and presence of sharp edges on the truck.

Unloading-related measures included the percentage of flocks per truck in which animals were recorded slipping or showing reluctance to move, and the percentage of animals per truck falling during unloading. These were investigated in relation to MBM (ie moving excitedly, producing loud noise, slapping/hitting the animals, performing forbidden practices) and RBM (ie ramp slope, ramp covering type and quantity, gaps between lorry-ramp-floor).

Spearman correlation tests were carried out to investigate correlations between continuous variables. In particular, slipping, falling and reluctance to move were investigated in relation to each of the five MBM (as percentage of flocks per truck where stockmen performed each of the MBM) and to ramp slope. *P*-level was set at 0.003 applying Bonferroni correction for 15 comparisons. Also, the percentages of dead, injured or non-ambulatory animals were correlated to stocking density and travel duration. After applying the Bonferroni correction for six comparisons, *P*-level was set at 0.008.

ABM included in the transport-related group (ie percentages of dead, injured or non-ambulatory animals) were also investigated in relation to categorical variables using non-parametric tests: the Mann-Whitney *U* test was used for dichotomous variables (ie deck height, truck hygiene, presence of sharp edges on the truck) while the Kruskal-Wallis test was used for variables with more than two categories (ie quantity and type of bedding). ABM included in the unloading group (ie falling and slipping behaviours) were tested in relation to dichotomous variables (ie type of ramp coverage, presence of sharp edges or slipping areas on the ramp) or other categorical variables (ie quantity of ramp coverage) using Mann-Whitney *U* tests and Kruskal-Wallis tests, respectively. The Mann-Whitney *U* test was also used as *post hoc* test where appropriate, applying Bonferroni correction. All analyses were carried out using R version 2.15.3 software package for Windows 7, and hypotheses were tested at the 0.05 level of significance, unless specified otherwise.

Results

Welfare assessment: descriptive analysis

The countries of departure for the 25 Italian assessments were represented by Hungary (n = 14), Romania (n = 6), France (n = 4) and Poland (n = 1), while those arriving in Greece came from Romania (n = 12), Greece (n = 2) and Spain (n = 1). On average, the overall travelled distance between the site of departure and the destination was 1,236 km (ranging from 471 to 1,867) whilst the average durations of transport were 23.2 h (ranging from 14 to 101 h). The total number of animals unloaded was 19,080 (mean 477, ranging from 90 to 880 sheep per truck). The outside temperature, at the time of unloading, ranged from –1 to 23°C. Most of the transported animals (over 92% including both adults and lambs) were within the weight category 'less than 55 kg'; the remaining 8% were 'over 55 kg'. Since no further information about the age class of the sheep was recorded, this parameter was not included in the following analysis.

The average duration of the unloading was 17.6 min (ranging from 5 to 45 min), and, overall, the assessments took approximately 50 min per truck, including the post-unloading assessment in the resting pens (P3).

Resource-based measures

With the exception of one truck, all ramps observed during the assessments were made of anti-slip corrugated metal and had lateral protections. Foot battens were present in all cases. No dangerous holes were recorded on the ramps, while sharp edges were present in 15% of cases. Straw was used to cover the ramp during unloading in 55% of cases, whereas no cover was provided for the remainder. Of these, 1/5 were only partially covered, exposing the ramp surface. Steps or gaps between the truck floor and the ramp were rarely observed (3% of cases) while a step between the end of the ramp and the floor was observed in 25% of the trucks. Ramp slope ranged from 10 to 70%, complying with the Regulation 1/2005 (ie less than 50% of the horizontal, corresponding to a 26.34° angle) in 85% of cases. In all but two transports, lighting condition during unloading was sufficient (according to the protocol definition; for a reference, see Grandin 2003) for the animals to orientate, and for the staff to safely perform the handling procedures.

Space allowance was, on average, 0.27 m^2 per animal, with a wide range of variability (between 0.09 and 0.7 m^2 per animal). In 58% of cases this was over 0.2 m^2 per animal, thus complying with Regulation 1/2005. Recording of RBM inside the truck showed that the most prevalent drinker type were nipple drinkers (60%), followed by water bowls (20.5%) or both systems together (15%). Absence of drinkers was recorded in one case. The water storage tank was empty in 35% of cases and in 40% of cases drinkers were not functioning at the arrival of the truck. The assessors reported difficulties in assessing the level of filling of some water tanks. Deck height was sufficient to allow the animals to stand in their natural position and allow air ventilation above them in the majority of transports (85%). Bedding material, straw in almost all cases and sawdust as an alternative, was present in all but one transport and the quantity was enough to cover the whole surface in 85% of cases. The deck floor appeared clean (less than 25% of the deck covered in manure) on 80% of trucks. Temperature-monitoring systems were not functioning in half the lorries.

Management-based measures

From the assessment of the unloading management procedures it emerged that in approximately 60% of flocks the stockmen performed appropriate handling (as defined in Table 2). In 20% of the assessments, both forbidden practices and loud noises were performed. Handlers moving excitedly or slapping the animals were observed on 3 and 7% of unloadings, respectively.

Animal-based measures

The recording of ABM during unloading showed that the percentages of animals dead on arrival and those that were unable to ambulate unassisted (non-ambulatory) were very low (about 0.2%; n = 24). Overall, 12 severely lame animals (0.09%) were observed. Falling was observed on most of the unloadings, ranging between 0.3 and 61.6% of the total animals per truck.

Slipping was recorded for most of the unloaded flocks (70.7%). Reluctance to move was observed on one-third of the unloaded flocks. Coughing events were recorded during seven unloadings.

None of the assessments carried out in the resting pens 20 min after unloading highlighted particular issues. Of the 7,483 sheep inspected (32% of the total), one was found dead in the pen, 17 were exhausted and four showed high respiratory rate. No signs of hampered respiration or coughing were recorded in any of the assessments.

Welfare assessment: identification of predictors

Transport-related measures

As the prevalence of ABM included in this group was very low (below 2%), statistical analysis could not be performed.

Unloading-related measures

A significant positive correlation emerged between the ramp slope and the percentage of falling events per truck (R_S = 0.49; P = 0.001, alpha ≤ 0.003 applying Bonferroni correction). Although not significant, similar trends were found comparing the performance of forbidden practices with falling events (R_S = 0.39; P = 0.01) and the ramp slope with slipping events (R_S = 0.37; P = 0.019).

Kruskal-Wallis tests revealed that the amount of ramp covering affected the percentage of falling and slipping behaviours (falling: H = 18.65; $P \leq 0.0001$; slipping: H = 11.03; P = 0.004). For both comparisons, *post hoc* analysis revealed that there was significantly less falling and slipping events when the ramp surface was totally covered compared to when it was partially or completely uncovered by straw (Figure 1).

No other significant relationships between measures emerged for variables included in the unloading-related group.

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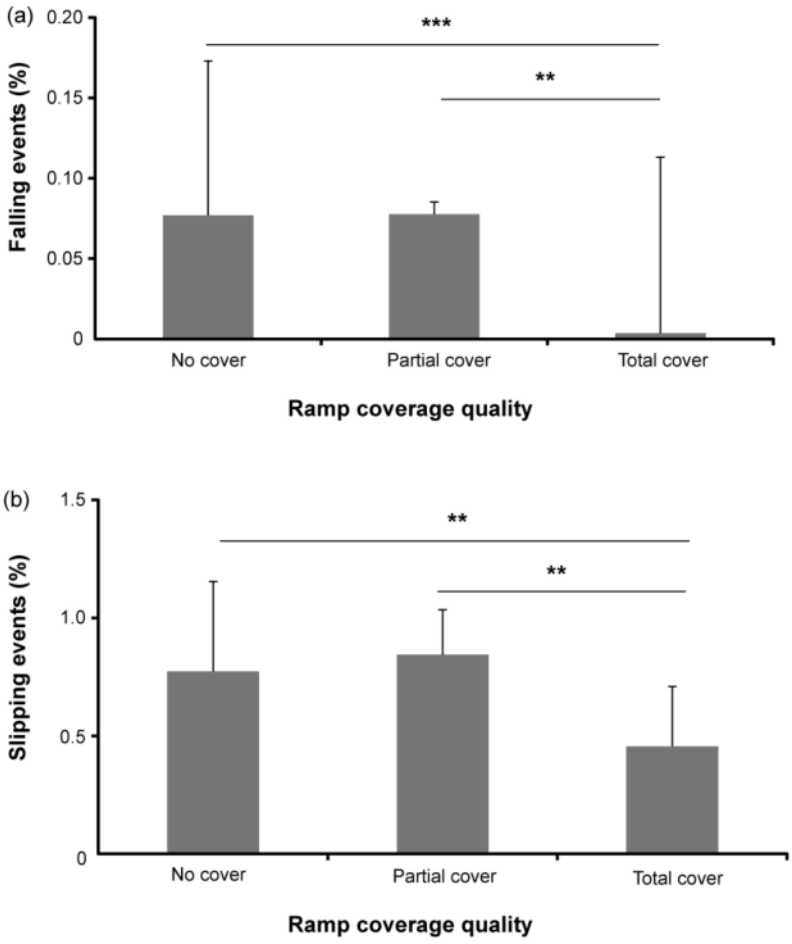
Figure 1



Association between ramp covering and falling and slipping behaviours. Comparison among different quantities of ramp covering (ie no cover, partial cover with straw, total cover with straw) during unloading associated with (a) falling and (b) slipping behaviours. Falling was measured at animal level (% of animals falling per truck) while slipping was calculated at group level (% of flocks slipping per truck). *Post hoc* comparisons showed that for both behaviours there was no significant difference when the ramp had either none or partial coverage, while there are significantly less falling and slipping when the ramp had total coverage in comparison to when it was partial (falling: $W = 110$; $P = 0.0002$; slipping: $W = 99$; $P = 0.003$) or when there was none (falling: $W = 221$; $P = 0.0002$; slipping: $W = 196$; $P = 0.007$).
** Significant level for $P < 0.01$;
*** Significant level for $P < 0.001$.

Discussion

Feasibility and reliability of the measures

The protocol presented in this paper was designed to assess the welfare of sheep (adults and lambs) travelling over long distances across the EU. This pilot study presents, to our knowledge, the first independent and practical tool to assess the welfare of transported sheep, which includes animal-, resource- and management-based measures. The measures were chosen to be relatively easy to assess for non-veterinary trained personnel, thus increasing the likelihood of it being implemented during various transport situations.

Recent developments in welfare science suggest that preference should be given to ABM in welfare assessment protocols (EFSA 2012). The European Commission also favours this approach as the way forward for regulatory and commercial purposes (European Commission 2012). The use of ABM for welfare assessment purposes has the advantage of being applicable in different situations, not being restricted to a given structure or facility. The animal-based measures assessed in this project were found to be feasible and repeatable across different observers. MBM, such as stockmanship skills, were assessed directly by observing the behaviour of the personnel, since indirect assessment of stockmanship via the animals has been shown to be too complex and inefficient in a previous study (de Passillé & Rushen 2005). The management-based criteria were selected from the Regulation 1/2005 requirements, all of them considered relevant for animal welfare and also allowing for the evaluation of the compliance with the legislation.

Four ABM were evaluated for inter-observer agreement and accuracy with a 'gold standard'. Accuracy in scoring the measures and inter-observer reliability were high for all measures assessed, highlighting their reproducibility. For some ABM, it was not possible to assess observer agreement because of their low prevalence in the dataset. This is not uncommon, as similar problems were also reported in other studies involving assessor training on animal based welfare measures (eg Burn *et al* 2009; Muri *et al* 2013).

Description of assessed transports

The assessments were carried out in the two European countries, Italy and Greece, which are the main sheep importers in the EU (73% of the overall in 2009) (Gebresenbet *et al* 2010).

Drinking devices were present in the compartments of all trucks but one. The presence of drinkers alone does not completely satisfy requirements which also stipulate that

water should be continuously available to the animals during travel. Some drivers, for example, admitted that the pipes were closed to avoid undesirable splashing and spilling of water during transportation, and opened only during the 1 h rest stop for the driver. For this reason, it was decided to include an evaluation of drinker functioning and water levels in the tank at arrival. However, these measures were difficult to assess and did not provide useful information on the actual availability of water during the journey. Further efforts should be made in order to define an operational measure to assess the degree of hydration in the animals following transport.

Space allowance can have major impact on the welfare of transported sheep (Knowles *et al* 1998), and adequate space allowance should be provided to animals travelling over long journeys (SCAHAW 2002). The EC 1/2005 Regulation minimum space requirement for shorn sheep weighing between 26 and 55 kg is 0.2 m^2 per animal, although lower space allowances may be permitted for small lambs. In this study, space allowance on the trucks was below 0.2 m^2 per animal in 40% of cases. However, in this study, the different stocking densities appeared not to affect any of the ABM.

One truck had several problems, eg an inappropriate ramp (made of wood) without lateral protections and no drinkers inside the compartments. Interestingly, this truck was used for a domestic transportation. This finding is in line with previous studies which have reported that truck specifications for long journey transports were better for international than domestic journeys (Baltussen *et al* 2011).

Management practices were often recorded as non-compliant with the Regulation. Although no animal appeared to be seriously harmed (eg no wounds or injuries were observed as a result of these actions) and no inappropriate tools were used to stimulate animal movements (eg no pointed implements or electric prods), sheep were often unloaded by dragging or lifting by the fleece or legs. These practices are likely to cause pain (Goddard 2008) and are also listed among the practices forbidden in the EC Regulation 1/2005. Moreover, in several instances, sheep were slapped or shouted at to make them move even though their movement was impaired by other animals in the flock. It is possible that the presence of the observer affected stockman behaviour, but not to the extent that inappropriate handling practices were completely avoided or concealed.

Welfare assessment: toward the identification of predictors

Resource- and management-based measures were found to be associated with animal-based measures, and these are thought to reflect the direct consequences of inappropriate travel conditions and unloading procedures (EFSA 2012). Links between predictors (RBM and MBM) and outcomes (ABM) were investigated during travel and unloading.

Concerning travel itself, statistical analysis of the data was not possible due to the low prevalence of the recorded ABM (eg non-ambulatory and dead animals). In future, it may be advantageous for assessment protocols to identify and include measures that have higher prevalences. Several different factors could have impacted our results. On the one hand, since the majority of trucks were found to be in compliance with the EC Regulation, negative effects on the animals will have been minimised, thus leading to a rather homogenous sample in the present study. In addition, since all assessments were performed after the authorisation of the food business operator at the place of arrival of the truck, there may have been a bias in the sample toward the higher quality end of the market (ie only transportations with a relatively low incidence or likelihood of poor welfare of the transported animals were included in the study). However, the stratification of results, which requires a larger sample and more systematic data collection, for example, by country of origin and destination or age of the animals, might provide more insight and should be considered to further improve the protocol which was developed in this pilot study. Previous studies have shown that age can have an impact on the ability of sheep to cope with long journeys (Zhong *et al* 2011). Future assessments should include an age classification in addition to the weight classification provided by the EC Reg 1/2005 (less or over 55 kg).

The analysis of measures assessed during unloading provided some indications about predictive factors associated with adverse effects on animals. In accordance with what has previously been described (Knowles *et al* 1998), the present results showed that a higher ramp slope increased the percentage of falling events and a similar trend was found when considering slipping events during descent from the truck. Although significant, the correlation was not very strong, which may be due to the fact that multiple factors may affect falling and slipping events during unloading. The quantity of ramp covering, for example, influenced the percentage of falling and slipping events which were more frequent when the straw on the ramp was either absent or quantitatively low, compared to ramps covered with straw. Slipping events agitate animals and tend to make unloading management harder (Grandin 2013). Limiting ramp slope and providing the appropriate quantity of bedding on the ramp may help to reduce these events, thereby improving both the safety of sheep and hastening unloading procedures, since animals will be less likely to stop or trip over each other. A trend in the results suggests that forbidden practices may increase the likelihood of falling events, entailing a higher injury risk. In addition, using positive stimuli (eg using a tame sheep to lead flocks down from the truck at arrival) has previously been reported to be more effective for moving sheep than using repeated fear-inducing stimuli (eg shouting or moving excitedly) (Eldridge *et al* 1982).

Animal welfare implications and conclusion

This paper describes a new protocol for the assessment of sheep and lamb welfare after long journeys, and its subsequent testing on 40 trucks arriving in two major importing countries in the EU (Italy and Greece). The protocol was comprehensive (ie including resource-, management- and animal-based measures) and easy to use: assessors only reported minor issues related to the practical applicability of the protocol.

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In contrast with current welfare assessment systems for long journey transportation, which are based on resource checklists only, the present tool is based on welfare outcome indicators, allowing for the identification of welfare consequences associated with unloading procedures. Further development of the protocol will be needed in order to refine the detection of hazards related to the travel itself and thus provide a proper risk analysis. Nevertheless, at the present stage of development, the protocol already represents a valuable tool to assess sheep welfare after long journeys, being able to provide direct feedback to transporters on the weaknesses and strengths of their work practices. The European Commission stimulates the development of market-oriented animal welfare standards for all phases of livestock production, thus providing an alternative to the 'regulatory approach'. The authors believe that this tool should be further developed and incorporated into a routine checking instrument for certification purposes in collaboration with the transport industry.

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References

Andreasen SN, Sandøe P and Forkman B 2014 Can animal-based welfare assessment be simplified? A comparison of the Welfare Quality® protocol for dairy cattle and the simpler and less time consuming protocol developed by the Danish Cattle Federation. *Animal Welfare 23*: 81-94. http://dx.doi.org/10.7120/09627286.23.1.081

Baltussen WHM, Gebrensbet G and de Roest K 2011 *Study on the impact of regulation (EC) no 1/2005 on the protection of animals during transport.* http://ec.europa.eu/food/animal/welfare/transport/docs/study_report_en.pdf

Blokhuis HJ, Veissier I, Miele M and Jones RB 2010 The Welfare Quality project and beyond: safeguarding farm animal well-being. *Acta Agriculurae Scandavica A, Animal Science 6*: 129-140

Broom DM 2005 The effects of land transport on animal welfare. *Revue Scientifique et Technique de l`Office International des Epizooties 24*: 683-691

Brown SN, Knowles TG, Wilkins LJ, Chadd SA and Warris PD 2005 The response of pigs to being loaded or unloaded onto commercial animal transporters using three systems. *The Veterinary Journal 170(1)*: 91-100. http://dx.doi.org/10.1016/j.tvjl.2004.05.003

Burn CC, Pritchard JC and Whay HR 2009 Observer reliability for working equine welfare assessment: problems with high prevalences of certain results. *Animal Welfare 18*: 177-187

Cohen J 1968 Weighted kappa: nominal scale agreement with provision for scaled disagreement or partial credit. *Psychological Bulletin 70*: 213-220. http://dx.doi.org/10.1037/h0026256

de Passillé AM and Rushen J 2005 Can we measure human–animal interactions in on-farm welfare assessment? Some unresolved issues. *Applied Animal Behaviour Science 92*: 193-209. http://dx.doi.org/10.1016/j.applanim.2005.05.006

EFSA Panel on Animal Health and Welfare (AHAW) 2011 Scientific Opinion concerning the welfare of animals during transport. *EFSA Journal 9(1)*: 1966

EFSA Panel on Animal Health and Welfare (AHAW) 2012 Statement on the use of animal-based measures to assess the welfare of animals. *EFSA Journal 10(6)*: 2767

Eldridge GA, Wythes JR, Arthur RJ, Meischke HRC, Vowles WJ, Hutson GD, Barnett JL, Hemsworth PH and Davies GT 1982 Handling and transport of meat animals in relation to efficiency, meat quality and welfare. *Animal Production in Australia 15*: 116-129

European Commission 2012 *Communication from the Commission to the European Parliament, the Council and the European Economic and Social Committee on the European Union Strategy for the Protection and Welfare of Animals 2012-2015*. EU: Brussels, Belgium

European Council 2004 *Council Regulation (EC) 1/2005 on the protection of animals during transport and related operations and amending and amending Directives 64/432/EEC and 93/119/EC and Regulation (EC) No 1255/97*. EU: Brussels, Belgium

European Parliament 2012 *Written declaration on the establishment of a maximum 8-hour journey limit for animals transported in the European Union for the purpose of being slaughtered*. http://www.europarl.europa.eu/sides/getDoc.do?pubRef=-//EP//TEXT+TA+P7-TA-2012-0096+0+DOC+XML+V0//EN

Fleiss JL 1971 Measuring nominal scale agreement among many raters. *Psychological Bulletin 76*: 378-382. http://dx.doi.org/10.1037/h0031619

Gavinelli A, Ferrara M and Simonin D 2008 Formulating policies for the welfare of animals during long distance transportation. *Veterinaria Italiana 44(1)*: 71-86

Gebresenbet G, Baltussen W, de Roest K, Neilson K and Sterrenburg P 2010 *Evaluation of the Feasibility of a Certification Scheme for High Quality Control Posts.* http://ec.europa.eu/food/animal/welfare/financing/docs/call_10753-2010__feasability_report_cepost_en.pdf

Goddard PJ 2008 The management of sheep. In: Dwyer K (ed) *The Welfare of Sheep* pp 291-323. Springer Science Business Media BV: The Netherlands. http://dx.doi.org/10.1007/978-1-4020-8553-6_8

Grandin T 2013 Recommended animal handling guidelines. In: Grandin T (ed) *Recommended Animal Handling Guidelines and Audit Guide: A Systematic Approach to Animal Welfare* pp 11-31. AMI Foundation: Washington, DC, USA

Keeling L 2009 How did we design the welfare measures? In: Butterworth A, Blokhuis H, Jones B and Veissier I (eds) *Delivering Animal Welfare and Quality: Transparency in the Food Production Chain* pp 23-25. 8–9 October 2009, Uppsala, Sweden

Knowles TG 1998 A review of the road transport of slaughter sheep. *Veterinary Record 143*: 212-219. http://dx.doi.org/10.1136/vr.143.8.212

Kristensen E, Dueholm L, Vink D, Andersen JE, Jakobsen EB, Illum-Nielsen S, Petersen FA and Enevoldsen C 2006 Within- and across-person uniformity of body condition scoring in Danish Holstein Cattle. *Journal of Dairy Science 89*: 3721-3728. http://dx.doi.org/10.3168/jds.S0022-0302(06)72413-4

Laister S, Brörkens N, Lolli S, Zucca D, Knierim U, Minero M, Canali E and Winckler C 2009 Reliability of measures of agonistic behaviour in dairy and beef cattle. In: Forkman B and Keeling L (eds) *Assessment of Animal Welfare Measures for Dairy Cattle, Beef Bulls and Veal Calves* pp 95-112. Welfare Quality reports: Cardiff University, UK

Landis JR and Kock GG 1977 The measurement of observer agreement for categorical data. *Biometrics 33*: 159-174. http://dx.doi.org/10.2307/2529310

March S, Brinkmann C and Winckler C 2007 Effect of training on the interobserver reliability of lameness scoring in the dairy cattle. *Animal Welfare 16*: 131-134

María GA, Villaroel M, Chacón G and Gebresenbet G 2004 Scoring system for evaluating the stress to cattle of commercial loading and unloading. *Veterinary Record 154(26)*: 818-821. http://dx.doi.org/10.1136/vr.154.26.818

Marlin D, Kettlewell P, Parkin T, Kennedy M, Broom D and Wood J 2011 Welfare and health of horses transported for slaughter within the European Union Part 1: Methodology and descriptive data. *Equine Veterinary Journal 43(1)*: 78-87. http://dx.doi.org/10.1111/j.2042-3306.2010.00124.x

Muri K, Stubsjøen SM and Valle PS 2013 Development and testing of an on-farm welfare assessment protocol for dairy goats. *Animal Welfare 22(3)*: 385-400. http://dx.doi.org/10.7120/09627286.22.3.385

Nielsen BL, Dybkjaer L and Herskin MS 2011 Road transport of farm animals: effects of journey duration on animal welfare. *Animal 5(3)*: 415-427. http://dx.doi.org/10.1017/S1751731110001989

SCAHAW 2002 *The Welfare of Animals During Transport (Details for Horses, Pigs, Sheep and Cattle)*. Scientific Committee on Animal Health and Animal Welfare, European Commission Health and Consumer Protection Directorate General: Brussels, Belgium

Veissier I, Butterworth A, Bettina B and Roe E 2008 European approaches to ensure good animal welfare. *Applied Animal Behaviour Science 113*: 279-297. http://dx.doi.org/10.1016/j.applanim.2008.01.008

Whay HR 2007 The journey to animal welfare improvement. *Animal Welfare 16*: 117-122

Zhong RZ, Liu HW, Zhou DW, Sun HX and Zhao CS 2011 The effects of road transportation on physiological responses and meat quality in sheep differing in age. *Journal of Animal Science 89*: 3742-3751. http://dx.doi.org/10.2527/jas.2010-3693

https://doi.org/10.7120/09627286.24.4.407 Published online by Cambridge University Press