What causes smothering in commercial free-range laying hens? Part 1

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Foreword

This project was conducted to identify risk factors for smothering in Australian free range layer flocks. Smothering in poultry occurs when birds mass together, often on top of each other, resulting in death from suffocation and there are published reports of smothering in loose-housed laying hens in Europe. While the incidence of mortality associated with smothering varies both within and between farms, anecdotal observations by several Australian free-range egg producers report that mortality due to smothering may account for up to 30-40% of total hen mortality in a production cycle.

While smothering has both economic and welfare-related concerns, little is known about smothering in laying hens because the causes appear to be multifactorial, outbreaks are sporadic and unpredictable, and detection is normally after the incident. The Animal Welfare Science Centre and the Veterinary Epidemiology group at the University of Melbourne conducted this prospective observational study at both the flock and farm-level in Australian free-range conditions to identify the major potential risk factors in relation to smothering in laying hens.

This project was funded from industry revenue, which is matched by funds provided by the Australian Government.

This report is an addition to Australian Eggs Limited’s range of peer reviewed research publications and an output of our R&D program, which aims to support improved efficiency, sustainability, product quality, education and technology transfer in the Australian egg industry.

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Abbreviations

ACTH	Adrenocorticotropic hormone
AGP	Alpha-1 acid glycoprotein
APP	Acute phase protein
AV	Aviary
BN	Barn
C	Celsius
CC	Conventional cage
CGB	Corticosteroid- binding globulin
CI	Confidence interval
cm	Centimetre
CNS	Central nervous system
CPM	Counts per minute
CRF	Corticotropin releasing factor
FC	Furnished cage
FR	Free-range
g	Gram
GR	Glucocorticoid receptor
h	Hour
HHP	Hen housed production
H/L	Heterophil:lymphocyte ratio
HPA	Hypothalamic-pituitary-adrenal axis
KMO	Kaiser-Meyer-Olkin
kPa	Kilopascal
m	Metre
min	Minute
mL	Millilitre
mm	Millimetre
MR	Mineralocorticoid receptor
NOT	Novel object test
PBS	Phosphate buffered saline
PCA	Principal component analysis
PVN	Paraventricular nucleus
RIA	Radioimmunoassay
s/sec	Second
SEM	Standard error of the mean
TI	Tonic immobility

Executive Summary

Smothering deaths appear to be caused by suffocation and estimates by Australian free-range egg producers indicate that smothering may account for up to 30 to 40% of total hen mortality in a flock. While smothering has both economic and welfare impacts, little is known about smothering in laying hens. This is because smothering has multiple causes, outbreaks are sporadic and unpredictable, and detection typically occurs well after the smothering incident has occurred. This project was a prospective cohort study of Australian free-range layer hens to identify risk factors for smothering deaths. Identifying and understanding the nature of these risk factors provides the rationale for subsequent experimental studies to confirm that they are, in fact, causally associated with smothering risk.

Three large commercial free-range egg organisations collaborated in the study, with data collection commencing at the organisations' barn and aviary free-range units in January 2019 and concluding in April 2022. A total of 86 flocks were enrolled in the study on the day of bird placement in the production sheds and were followed until the day of depopulation. Risk factors for smothering events were assessed at the bird, flock, shed and organisation level.

Time to event analyses were carried out to document when smothering mortalities occurred in relation to the day of placement. Candidate risk factors were assessed initially using Kaplan-Meier survival curves, grouped at the organisation, shed and flock level. While not adjusting for the effect of confounders, the Kaplan-Meier survival curves indicated a marked difference in smothering risk by several of the candidate risk factors including organisation, shed type, breed, climatic conditions and flock management factors. For example, there were marked differences in smothering risk by organisation with Organisation 1 having the lowest risk (1.28 smothering deaths per 100 birds placed) followed by Organisation 3 (2.09 per 100 birds placed) and Organisation 2 (2.11 per 100 birds placed). Candidate risk factors that were associated with time to smothering event were included in a stratified, multilevel Cox proportional hazards regression model. This allowed us to control for the effect of known confounders, many of which were operating at the organisation and flock-within-organisation level. This provided the opportunity to identify risk factors that showed a consistent relationship with smothering risk across the three organisations.

Smothering risk was associated with shed type (aviary sheds, higher risk), the number of shed walks per day (fewer shed walks per day, higher risk), the number of eggs produced per bird per day (more eggs per bird per day, higher risk), human test score (lower fear/higher curiosity of humans, higher risk), indoor and outdoor novel object test score (higher fear/lower curiosity of novelty, higher risk), outdoor daily average temperature (temperatures outside of the 10-27 °C recommended range, lower risk) and outdoor daily average humidity (higher humidity, higher risk).

The findings from this research provide the necessary first steps to allow producers to implement interventions to reduce smothering risk in their flocks. A profitable area of future work would be to manipulate the modifiable risk factors listed above and to document the change in smothering incidence risk that followed. This would provide free-range egg producers with greater confidence that the risk factors identified in this report are, in fact, causal.

Overall Conclusions

Smothering was a prominent contributor to the total number of bird deaths that occurred on the study farms (44,239 smothering deaths out of a total of 278,634 hen deaths based on 2,427,673 hens placed in the study farms). Throughout the production cycle, 11.5 deaths occurred for every 100 birds placed, with 1.8 deaths per 100 birds placed due to smothering (i.e., 16% of all bird deaths were due to smothering).

There was marked variation in smothering deaths both between organisations (1.28 to 2.11 deaths per 100 birds placed) and between flocks (0 to 3.5 deaths per 100 birds placed). This level of smothering in free-range laying hens raises both welfare and financial concerns.

The Cox proportional hazards regression modelling shows that smothering risk was increased in flocks of hens:

a) housed in aviary sheds;
b) where there were low numbers of shed walks per day;
c) with high levels of egg production;
d) where hens showed lower levels of fear (and higher levels of curiosity) towards a human; and
e) where hens showed higher levels of fear (and lower levels of curiosity) towards a novel object.

Environmental conditions that increased smothering risk included days when:

a) outdoor daily average temperatures were within the 10 to 27°C recommended range; and
b) outside daily average humidity was 70% or greater.

While these results do not demonstrate causality, they provide strong evidence and thus rationale for experimental research. A profitable area of future investigation would be to modify these identified variables in a controlled experiment to determine the subsequent effect on smothering incidence.

1 Introduction

Smothering in poultry occurs when birds group together, often on top of one another, resulting in death from suffocation (Bright and Johnson 2011). Smothering is mainly reported in non-cage systems (see reviews by Lay et al. 2011; Gebhardt-Henrich and Stratmann 2016). There are published reports of smothering in loose-housed laying hens in Europe (Bright and Johnson 2011; Barrett et al. 2014; Defra 2015; Winter et al. 2021), and the problem is understood to occur in other regions where hens are kept in large commercial groupings in non-caged environments (Singh et al. 2017). Smothering is generally considered an unpredictable problem (Bright and Johnson 2011; Gebhardt-Henrich et al., 2016; Rayner et al. 2016) that can cause the death of a large number of birds. For example, smothering accounted for 11% and 16% of total mortality by 40 and 71 weeks of age, respectively in a UK study of 20 flocks that were loose housed in either free-range, barn or aviary (Defra 2015). A survey of 41 Australian free-range flocks found that farmers attributed high mortality rates to predation, grass impaction, heat stress, disease and parasites, and smothering (Singh et al. 2017). While the incidence of mortality associated with smothering varies both within and between farms, anecdotal observations by a number of Australian free-range egg producers suggest that mortality due to smothering may account for up to 30-40% of total hen mortality in a production cycle (P. Szepe, Kinross Farms, personal communication, 5 April 2018; P. Groot, DA Hall & Co Pty Ltd, personal communication, 5 April 2018).

In interviews with ten UK free-range laying hen producers, Bright and Johnson (2011) reported three categories of smother: (1) panic smothers, which were thought to be caused by disturbances due to predators, vermin, sudden noises or light intensity; (2) nest box smothers, which mainly appeared early in lay with a hen in a nest box appearing to attract others to crowd into the same nest box; and (3) creeping/recurring smothers, which appeared to be the most problematic because it was the most common and once started it recurred throughout the laying period. Producers agreed that the triggers for creeping/recurring smothers were age, temperature fluctuations and litter condition. In a questionnaire survey of free-range farm managers (n = 206 respondents), representing 35% of the UK free-range egg supply, Barrett et al. (2014) found that nearly 60% of farm managers reported smothering occurring in their flocks, with an average of 26 birds lost per incident, although mortality due to smothering was low (mean of 2%). Rayner et al. (2016) utilised the results of the questionnaire survey conducted by Barrett et al. (2014) to examine the relationships between, and possible effects of, disease, housing, management practices and smothering on free-range farms. Rayner and colleagues found that breed and nest box manufacturer predicted nest box smothers, while nest box manufacturer, provision of oyster grit or grain on the litter and range use on a sunny day predicted panic and recurring smothers. It is notable that Barrett and colleagues commented that questionnaire studies are retrospective by nature and that it was hoped that their study would provide the stimulus for future on-farm epidemiological studies of smothering issues.

The surveys of laying hen farmers in the UK by Bright and Johnson (2011) and Rayner et al. (2016) suggest that piling behaviour is the behaviour preceding smothering. Piling behaviour in commercial hens has been identified in two studies. In an observational study of two USA flocks housed in barns, Campbell et al. (2016) suggested that laying hens may be socially facilitated to dust bath together on litter and that this flock synchrony could represent an expression of a preference that may lead to piling. Furthermore, the authors suggested that the observed piling might be similar to 'creeping/recurring' piling as defined by Bright and Johnson (2011) as this also occurred spontaneously at any time of day across all time points of the flock cycle and was most frequent on open litter and in corners. The authors also reported that only 7% of the observed piles appeared to result from flock disturbance and that no smothering events were observed in this study. Winter et al. (2021) studied piling behaviour inside the barn and winter garden of 13 commercial Swiss layer farms. Piling behaviour was preceded mainly by single hen activities (78% of piles) and 'non-hysterical' mass movements (8% of piles) and piling behaviour (number of piles) was related to time of day (more at 5-10 hours after the lights came on), flock colour (more in white, and white and brown mixed flocks than brown flocks) and flock age (more in hens at 20 weeks than 30 weeks of age). Only one possible smothering event was observed in this study. During an outbreak of recurrent smothering in a UK commercial free-range flock, video observations revealed 48 piles on 33 of 34 observation days, which resulted in six smothering events and a total 74 deaths (Herbert et al. 2021). These observations were conducted in spring, and the authors reported significant positive associations between piling extent (maximum bird count), water to feed ratio, and the temperature range reported on that day. However, Winter et al. (2021) found that piling behaviour was not related to the temperature in corners of the barn. The paucity of smothering relative to piles in these studies indicates that piling does not often lead to smothering.

Using the existing literature, Gray et al. (2020) proposed four hypotheses relevant to the causes of piling behaviour: (1) it is caused by hens moving toward or away from an attractant/repellent; (2) it is socially influenced; (3) it is influenced by early life experiences and (4) it can be described as a maladaptive collective behaviour.

Smothering deaths appear to be caused by suffocation (Bright and Johnson 2011) and death due to suffocation is not accepted as a humane method for killing animals, including poultry (European Food Standards Agency, 2019). Herbert et al. (2021) estimated that the UK egg industry loses GBP 6.5 million per year due to smothering, based on the mortality rates reported by Bright and Johnson (2011). Thus, hen mortality has welfare (and in turn consumer expectation) implications as well as other important aspects of sustainability such as the environmental footprint and economic viability of the production system (Matthews and Sumner 2015; Weeks et al. 2016).

While smothering obviously has both economic and welfare-related concerns, little is known about smothering in laying hens because it is a problem that appears to be multifactorial, outbreaks are sporadic and unpredictable, and detection often occurs well after the incident itself. Furthermore, smothering is hard to induce experimentally. As Gray and colleagues (2020) suggest, since smothering appears multifactorial, different prevention strategies are necessary for successful mitigation. Therefore, this research project used an epidemiological approach to identify bird, flock- and farm-level characteristics associated with smothering risk in Australian free-range laying hens. Identifying these factors provides the rationale for experiments to confirm that observed effects are, in fact, causal. In addition to the epidemiological study, observations on bird behaviour patterns and circumstances around individual piling events were conducted to characterise piling behaviour and to identify antecedents of piling events.

2 Materials and methods

2.1 Study sites

This was a prospective cohort study carried out over three years on 86 flocks of adult laying hens housed in 47 commercial farms operated by three organisations located in North-East Victoria (n = 20) and South-West Queensland (n = 27). Flock sizes at placement ranged from 16,346 to 45,311 hens. The climate at each location is classified as temperate based on the Köppen classification system.

2.1.1 Rearing systems

Day-old chicks were transferred to indoor rearing facilities at each organisation and transferred to the laying facility at approximately 17 weeks of age. There were three types of rearing facilities at these rearing farms (Figure 1) although the design of each of the three rearing systems varied both within and between organisations.

Floor rearing

The floor rearing system had a fully littered floor, with the drinkers and feeders accessed on a single level. Perching structures and hay bales were also provided. Flock sizes in this system ranged from 19,166 to 38,581.

Jumpstart

The Jumpstart system was an aviary-like structure with multiple levels, which could be raised or lowered to suit the age of the bird. The system was designed to train the young birds to jump to the different levels and perches to access food and water. The Jumpstart system also had a littered-scratch area. Flock sizes in this system ranged from 19,439 to 67,297.

Aviary

The aviary rearing system was a solid structure with multiple levels, which could be closed or opened to suit the age of the birds. Chicks were placed in the central level at one day old and then given access to other levels as they aged. The system incorporated ramps and lighting to encourage the birds to move through the different levels. A scratch area was provided on the floor but was not accessible until the system had been fully opened. Flock sizes in this system ranged from 39,536 to 44,420.

Figure 1 Images of the three types of rearing facilities at 16 weeks of age: Floor rearing (A), Jumpstart (B), Aviary (C).

2.1.2 Production systems

Two types of production (adult hen) facilities were used by the three participant organisations (Table 1).

Flat deck system

The flat deck system was a single tier system with all resources provided on a single level. Flooring was either partially slatted floor with litter or fully slatted. Perches were provided as removable metal structures on the slatted area, and nest boxes were provided either in a central column down the length of the shed or in two rows, and in either single- or double-layer nest boxes. Some flat deck systems also had an outdoor littered area under a veranda (‘winter garden’), which was accessible during the day even when the range was closed (e.g., during inclement weather; Figure 2).

Figure 2 Images of flat deck systems with fully slatted floor (A & B), winter garden (C) and indoor litter area (D).

Aviary system

The aviary system was a multi-tier system that provided nest boxes, perching, food and water in several levels. The design enables more hens to be housed per m^2 ground floor space as it allows the hens to disperse vertically as well as horizontally. There were also two types of aviary system at the study farms: A-frame and terrace aviaries. The A-frame aviary had a single central raised structure with multiple levels, with a scratch area underneath on the entire floor, and aisles on several levels for human passage (Figures 3A and B). The terrace aviary had multiple aviary structures in rows to create channels between the structures down the length of the shed. Human passage was possible only on the ground level, and the entire floor area between and underneath the aviary structures provided a litter area to hens (Figure 3C, D and E).

Figure 3 Images showing an A-frame aviary (A) with stockperson walkway (B), and a terrace aviary (C and D) with ramps to assist hen access to upper levels (E).

Table 1 Types of housing systems in the study farms.

Stage	Housing type		Organisation 1	Organisation 2	Organisation 3
Rearing	Floor rearing			x	
	Jumpstart		x	x	
	Aviary				x
Production	Aviary	A-frame	x	x	
		Terrace			x
	Flat deck	Full slats	x		
		Internal scratch area		x	
		Full slats, winter garden	x	x	

Pullets from the multi-tiered rearing systems (Aviary and Jumpstart) in all organisations were placed in aviary production sheds. Pullets destined for flat-deck production sheds were reared in a floor rearing system in Organisation 2, while in Organisation 1, all pullets were reared in a Jumpstart system regardless of the system used in production.

2.2 Data collection

2.2.1 Farm records and flock manager interviews

Flock managers agreed to provide routinely recorded production details for each flock entering the production cycle. Data collection commenced with the first flock placed in a production shed in January 2019 and concluded in April 2022. The production cycle varied from 64-88 weeks across the study flocks.

Flock-level data included the date on which hens were placed in the production shed (approximately 17 weeks of age), the identity of the shed of placement and details of shed type (aviary, flat deck). Each time the pop holes opened and closed, and the time of scheduled stockperson flock checks were recorded throughout the day for each shed.

Flock-level production data included the total number of birds at placement and daily records of the total number of hen mortalities and the total number of eggs produced. Total egg counts were categorised into oversized, floor eggs, dirty eggs and those suitable for sale. Daily shed-level data included the minimum and maximum temperature and the volume of water, and the weight of feed offered per bird per day.

Interviews were conducted with flock managers at the end of lay to record details of routine management practices, the number of staff that worked with the flock, and some qualitative assessments of hen behaviour.

2.2.2 Bird fear and exploration tests

Behaviour tests were conducted on 49 of the study flocks at 45-55 weeks of age. The two behaviour tests were a human test and a novel object test (NOT) and hen responses to each test were video recorded. The human test (adapted from similar tests used by Cransberg et al*.,* 2000 and Raubek et al., 2007) involved an observer moving through the shed and range in a standard manner stopping every 20 steps for 30 seconds. The number of hens within 1.25 m in front of the observer was recorded at every step when observer was moving, and every 30 seconds whilst the observer was stationary. In the NOT (adapted from a similar test used by Donaldson and O'Connell, 2012), a children's toy was presented in three locations in both the shed and range. The latency for hens to approach (10 or 40 cm radius from the novel object) and interact with the novel object, and the average number of hens within 40 cm of the novel object were recorded every 30 s over 5 mins in the novel object test.

It should be recognised that these two tests were designed to measure the approach and avoidance responses of birds to humans and a novel object. Measures of exploration are often interpreted as providing information about fearfulness and are the basis of some of the standard tests of fear (Bateson and Matheson 2007). Although it is unclear how exactly fear and exploration, particularly curiosity or inquisitive exploration, are related, it is generally agreed that high levels of fear inhibit all other motivational systems including exploration (Feenders et al. 2011). Similar tests to assess fear of humans and novelty have been used previously on laying hens (see Hemsworth and Edwards 2020).

2.2.3 Smothering records

Details of birds that died due to smothering were entered on recording sheets developed for this study. Smothering details included the date and time when the smothering event was detected, the initials of the person recording the event, where the smothering event occurred and a count of the number of birds that died. The location of the smothering event either within the shed itself or outside of the shed on the range was defined using a simple alpha-numeric identifier system developed for each shed. A diagram included at the front of each shed smothering record book provided details of the code to use for each smothering event location. This allowed us to define the location of smothering events down to an accuracy of approximately 10 metres. Additional details included whether or not the smothering event was detected at the time it actually occurred, and free text notes.

Flock data were collected from the time of flock placement in the production system in the study until the end of lay or until the end of the study on 31 March 2022, whichever occurred first. Contact was made with farm staff on a regular basis throughout the study period either in person (before COVID-19 restrictions were applied in March 2020), or by phone or teleconference. Records (production data and smothering logbook data) were transcribed into a relational database by project staff for subsequent analyses. The study variables are presented in Table 2.

There was marked variation in the temporal detail of each of the study variables. Some variables (e.g., indoor and outdoor average temperature) were recorded on a daily basis while others (e.g., presence or absence of a health event) pertained to the entire production cycle. We acknowledge that the variables relating to the entire production cycle were coarse (i.e., insensitive) indicators of smothering risk. Our reasoning for including them (presence or absence of health events, in particular) was to determine if flock manager perceptions of flock characteristics showed any association with the magnitude of smothering incidence risk.

Indoor and outdoor daily average temperatures were re-expressed as dichotomous variables: inside or outside the recommended 10 to 27 °C range for free range layer poultry principally to make it easier for flock managers to interpret the results of the regression model results. This approach was fortuitously convenient because, after explanatory variable adjustment, smothering hazards for temperatures below 10 °C and above 27 °C were of the same sign.

Table 2 Study variables including source of data and description

Variables	Data source	Description
Flock:		
Breed	Farm records	Bird breed - Hy-line Brown, ISA Brown and mixed (ISA Brown and Lohmann or ISA Brown and Hy-line Brown)
Flock number	Farm records	First or second flock within the same shed in the study
Number of birds placed	Farm records	Number of birds in the flock at placement
Weather: [a]		
Temperature	Weather station	Average daily temperature relative to 10-27°C
Humidity	Weather station	Average daily humidity (%)
Barometric pressure	Weather station	Maximum daily atmospheric pressure (kPa)
Precipitation	Weather station	Daily precipitation (mm)
Dew point	Weather station	Lowest temperature at which water vapour starts to condensate (°C)
Staff:		
Staff per shed	Interview	The number of different staff that worked with the flock across the production cycle
Number of shed walks per day	Interview	The average number of daily full floor walks conducted across the production cycle
Egg production:		
Eggs per bird per day	Farm records	Total number of eggs per bird per day, recorded on a daily basis
Floor eggs per bird per day	Farm records	Number of floor eggs per bird per day, recorded daily
Inside shed environment:		
Temperature [b]	Farm records	Average daily shed temperature relative to 10-27 °C
Water	Farm records	Average volume of water (mL) consumed (recorded daily)
Feed	Farm records	Average weight of feed offered (g) per bird recorded daily
Water to feed ratio	Farm records	Feed offered to water consumed ratio calculated daily using farm records
Shed:		
Rearing shed type	Farm records	Type of rearing system (floor, aviary or jumpstart)
Production shed type	Farm records	Type of production system (Flat deck or aviary)
Pop hole length	Farm records	Length of pop hole (cm) per bird
Lux	Interview	Light intensity the shed is maintained at
Health:		
Disease events:	Interview	Occurrence of disease in the flock during production (yes/no)
Behaviour:		
Piling behaviour	Interview	Score of prevalence of piling behaviour as perceived by shed managers
Human test:	Behaviour tests	Score for behavioural responses to a human observer inside the shed and in the range
Novel object test (indoor)	Behaviour tests	Score for behavioural responses to a novel object inside the shed
Novel object test (outdoor)	Behaviour tests	Score for behavioural responses to a novel object in the range

[a] Weather station located outdoors at each of the study farms.

[b] This is the range of temperature outside of which the organisations take protective management measures (e.g., activate foggers or raise curtains).

2.2.4 Video observations on piling behaviour

This study was conducted on a free-range commercial aviary egg farm in Northern Victoria with a study flock of approximately 22,514 Hy-Line brown hens. Hens were housed in an A-frame aviary system and were approximately 17 weeks old when placed in the laying facility.

A brief description of the observations is provided below:

Behaviour observations

An outdoor eyeball dome varifocal IR CCTV camera was installed and recorded continuously to a 16 Channel 400 FPS GeoVision DVR System located on site. The camera was positioned to cover a 5 m × 5 m area in the litter area of an A-frame aviary system inside the shed. The location was identified as a location of frequent piling and smothering, in discussions with the shed supervisors.

Selected observation periods

Observations were conducted on 6 individual days in which a smothering event involving more than 10 hens was reported by shed supervisors in the smothering logbook ('smothering days', n =6), each 'smothering day' observation period was also paired with an observation period in which no smothering event was reported ('non-smothering days', n=6). On the smothering days, the observations were conducted within 24 hours of the smothering event (based on the reported estimated time of the event), whilst a non-smothering day was selected from the same 7-day period as the paired smothering day but not within 24 hours of the smothering day.

Observations of piles

All occurrences of piling within the field of view of the camera between 0700–2000 h on selected days were recorded. Piling was defined as a minimum of 10 hens pressed together with individual movement restricted (i.e., the hens move only as a solid group in response to pressure from other hens unless climbing on top of the others) for a minimum duration of 1 min. The following characteristics of the pile were recorded: start time and end time; maximum head count based on the number of visible combs in the pile at the peak of the pile; pile location (in relation to walls and structures); hen orientation (based on focal hens within the pile); any identifiable sudden changes in the environment (e.g. sudden changes in light intensity, stockperson interactions, pop holes opening, etc.); and identifiable attractants (e.g., shards of light), any discernible reason for the dispersal (e.g., stockperson). Observations were conducted by a single experienced observer with 15 years of experience in developing ethograms and coding behaviour observations. To assess inter-observer reliability in identifying piles, a single day with 12 piles was observed by an additional observer using the same ethogram with excellent agreement (Cohen's kappa = 1.0).

2.3 Statistical methods

2.3.1 Bird fear and exploration tests

A principal component analysis (PCA) with Varimax with Kaiser Normalization was performed in SPSS (version 28) on the data from the behaviour tests to identify sets of components that represent the underlying commonalities (components) in these tests, and regression factor scores were calculated. Since the correlation coefficients were all above the required 0.3, the Kaiser-Meyer-Olkin (KMO) values exceeded the recommended value of 0.6, and Bartlett's Test of Sphericity was statistically significant, the data were considered suitable for the PCA. The human score test PCA results were classified as either less than zero or greater than or equal to zero, with scores greater than zero representing flocks that had a lower fear of humans. Similarly, the novel object test PCA results were classified as either less than zero or greater than or equal to zero, with scores greater than zero representing flocks that had a high fear of novelty.

2.3.2 Epidemiological analyses

The timing of flock-level smothering events in relation to days in lay and calendar date were analysed using a survival analysis approach (Clark et al. 2003). Following placement into the flock at approximately 17 weeks of age, birds were followed until the end of the production cycle or death due to smothering, whichever occurred first. The outcome of interest for our analyses was the calendar date on which a smothering event occurred, and the number of birds that were still present in their respective flock at the end of the production cycle were treated as censored observations (Clark et al. 2003). Due to the large number of birds in each flock, numeric weights representing the number of birds experiencing the event of interest on a given day were used. Similarly, numeric weights were assigned to the number of birds present at the end of the production cycle that were still alive. For birds that were still alive at the end of the production cycle, the weight assigned was equal to the number of birds that entered the shed on the date of placement minus the total number of recorded smothering deaths that occurred throughout the laying period.

The Kaplan-Meier method (Kaplan and Meier 1958) is a commonly used technique to describe time to event (in the context of this study, the date of smothering). The Kaplan-Meier estimate of survival can be plotted graphically with time on the horizontal axis and the corresponding proportion of surviving individuals at each time point on the vertical axis. Kaplan-Meier survival curves are presented as a series of horizontal steps, which is ever-declining in magnitude. The slope of the Kaplan-Meier survival curve provides an indication of how quickly the event of interest is occurring over time. The vertical position of the Kaplan-Meier estimate at the end of the follow-up period quantifies the proportion of the group initially at risk that did not experience the event of interest.

Analyses were carried out comparing time to smothering for birds with and without a given characteristic thought to influence smothering probability. In the remainder of this report, we call these characteristics 'explanatory variables'. The similarity of the Kaplan-Meier survival curves among levels of a given explanatory variable was tested using the log rank statistic. Explanatory variables where there was evidence of an association with time to event that was statistically significant at the (relatively conservative) alpha level of 0.20 were selected as candidates for inclusion in a multivariable Cox proportional hazards regression model. A Cox proportional hazards regression model allowed us to provide appropriate estimates of the strength of the association between each explanatory variable and smothering risk, adjusted for the effect of variables known to confound (i.e., distort) the association between the two (e.g., organisation and flock).

To start, a Cox model was developed including all explanatory variables identified as influencing smothering time to event at the bivariate level (described above). Analyses were carried out to determine if the association between a given explanatory variable and the logarithm of daily smothering hazard was linear (i.e., as the numeric value of an explanatory variable increased there was a corresponding increase or decrease in log daily hazard that did not depend on the magnitude of numeric value of the explanatory variable). Where a linear relationship was present the explanatory variable was entered into the model in an untransformed state. Explanatory variables where there was a non-linear association with log hazard were categorised, with categorisation choices driven by those that would be meaningful for poultry producers (e.g., indoor daily average temperature within the 10-27 °C recommended range; outdoor daily average temperature outside of the 10-27 °C recommended range).

Explanatory variables that were not statistically significant were removed from the model one at a time, beginning with the least significant, until the estimated regression coefficients for all explanatory variables retained were significant at an alpha level of less than 0.05. Explanatory variables that were judged to be likely confounders were retained in the model, regardless of their statistical significance. Explanatory variables that were excluded at the initial screening stage were tested for inclusion in the final model and were retained in the model if their inclusion changed any of the estimated regression coefficients by more than 20%. Biologically plausible two-way interactions between explanatory variables were assessed.

A key assumption of the Cox model is that of proportionality of hazards. According to this assumption, the effect of an explanatory variable on the outcome of interest does not change over time, i.e., the hazards for each level of an explanatory variable must be proportional at all times. To verify the proportional hazards assumption we plotted the scaled Schoenfeld residuals from the model as a function of follow-up time. Where the proportional hazards assumption holds, the Schoenfeld residuals should be scattered around zero. The Pearson product–moment correlation between the scaled Schoenfeld residuals and time was calculated and the hypothesis of no correlation between the two variables was assessed using a chi-squared test statistic. The proportional hazards assumption held for all explanatory variables included in the final model.

To deal with the confounding effect of flock (within organisation) the Cox model was stratified by organisation and a random effect term included in the model to account for flock (Kleinbaum and Klein 2012). While this eliminated our ability to quantify the association between flock and time to smothering event it had the advantage of providing hazard estimates that were appropriately adjusted for unmeasured organisation-level and flock-level effects.

Our Kaplan-Meier survival curves and Cox proportional hazards models were developed using the contributed survival package (Therneau and Grambsch 2001) implemented in R version 4.1.1 (Team 2022).

3 Results

3.1 Descriptive analysis

3.1.1 Flock enrolment

Table 3 and Figure 4 summarise the number of flocks and bird numbers, and the enrolment of flocks over the study period.

Table 3 Hierarchical structure of the data of laying hens on three free-range organisations in Australia, 2019-2022.

Level	n	Number at next highest level		
		Median	Min	Max
Organisation	3			
Flocks	81	30 [a]	17	34
Birds	2,369,446	20,309	16,346	45,311

[a] Interpretation: the median number of flocks per farm (i.e., the next highest level) was 30.

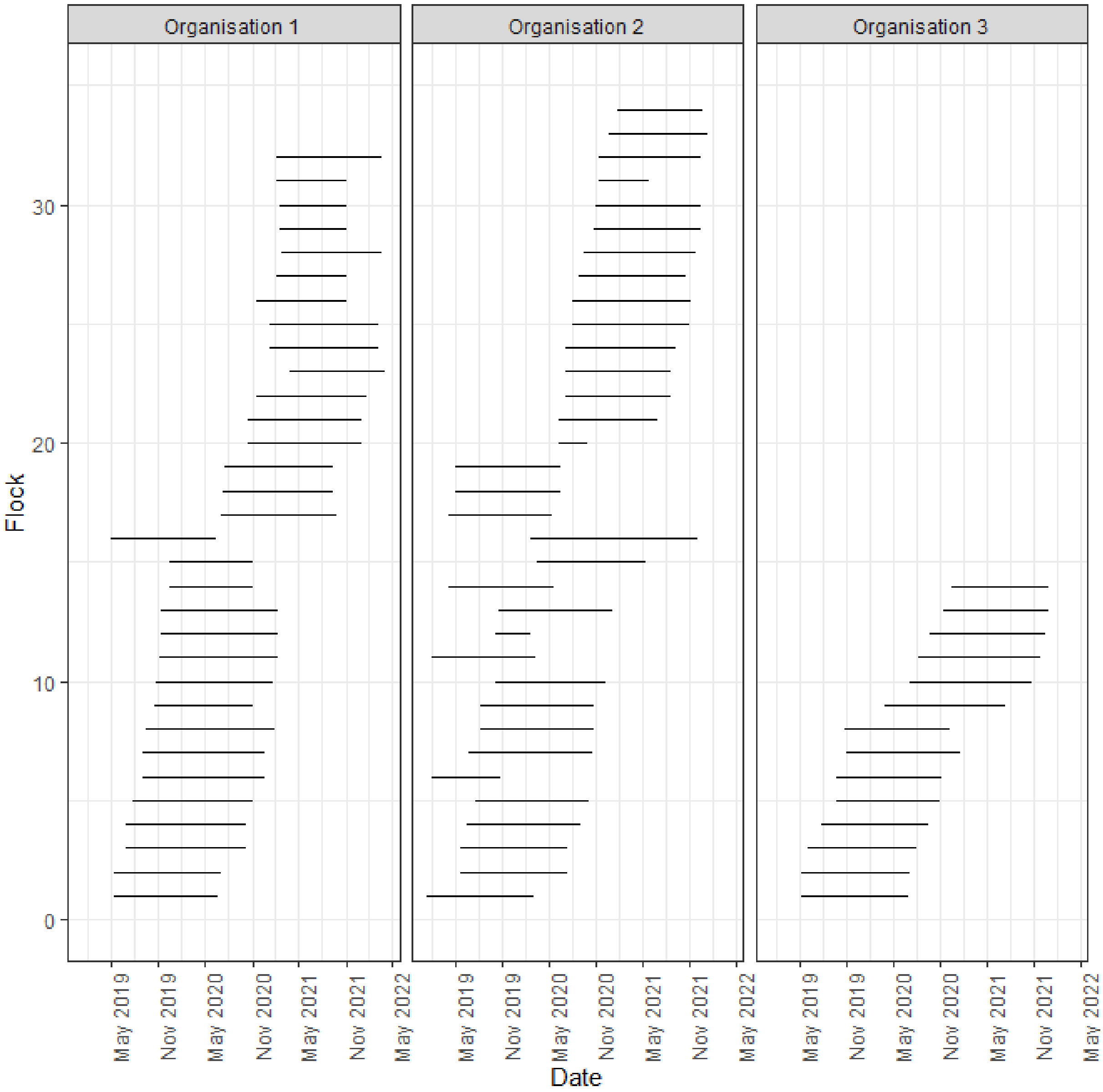

Figure 4 Line plot showing the data collection period for each flock (from timing of flock enrolment into the study to end of lay) by organization.

3.1.2 Smothering related mortality

Tables 4-6 and Figures 5 and 6 summarise smothering mortality in the study birds overall in the three organisations, indoors and outdoors, and in aviary sheds and in flat decks.

Table 4 Counts of the total number of birds placed into the production sheds, counts of the total number of bird-days at risk, counts of smothering deaths and counts of deaths for other reasons, and the incidence risk of smothering deaths and deaths for other reasons, by organisation.

Organisation	Birds	Bird-days [a]	Number			Incidence risk [b]		
			Smother	Other [c]	Total	Smother	Other [c]	Total
1	824,919	279.1	10,546	107,808	118,354	1.28	13.1	14.3
2	910,125	346.6	19,214	73,960	93,174	2.11	8.1	10.2
3	692,629	284.8	14,479	52,608	67,087	2.09	7.6	9.7
Total	2,427,673	910.4	44,239	234,376	278,634	1.82	9.7	11.5

[a] × 1,000,000

[b] Deaths per 100 birds.

[c] ‘Other’ refers to deaths not due to smothering

Table 5 Counts of the total number of birds placed into the production sheds, counts of the total number of bird-days at risk, counts of smothering deaths, the incidence risk of smothering deaths, and the incidence rate of smothering deaths by organisation and location of the smothering event (indoor, outdoor and not recorded locations). The number of birds and the number of bird-days at risk are the same for indoor, outdoor and smothering events where locations were not recorded because it was not possible to determine the proportion of time spent outdoors at the individual bird level.

Location	Birds	Bird-days [a]	Number	Incidence risk [b]
Indoor	2,427,673	910.4	12,745	0.52
Outdoor	2,427,673	910.4	18,925	0.78
Not recorded	2,427,673	910.4	12,569	0.52
Total	2,427,673	910.4	44,239	1.82

[a] × 1,000,000.

[b] Deaths per 100 birds.

Table 6 Counts of the total number of birds placed into the production sheds, counts of the total number of bird-days at risk, counts of smothering deaths and counts of deaths for other reasons, and the incidence risk of smothering deaths and deaths for other reasons, by shed type.

Shed type	Birds	Bird-days [a]	Number			Incidence risk [b]		
			Smother	Other	Total	Smother	Other	Total
Aviary	1,664,844	624.8	32,976	164,787	197,763	1.98	9.90	11.88
Flat deck	762,829	285.6	11,263	69,589	80,852	1.48	9.12	10.60
Total	2,427,673	910.4	44,239	234,376	278,615	1.82	9.7	11.5

[a] × 1,000,000.

[b] Deaths per 100 birds.

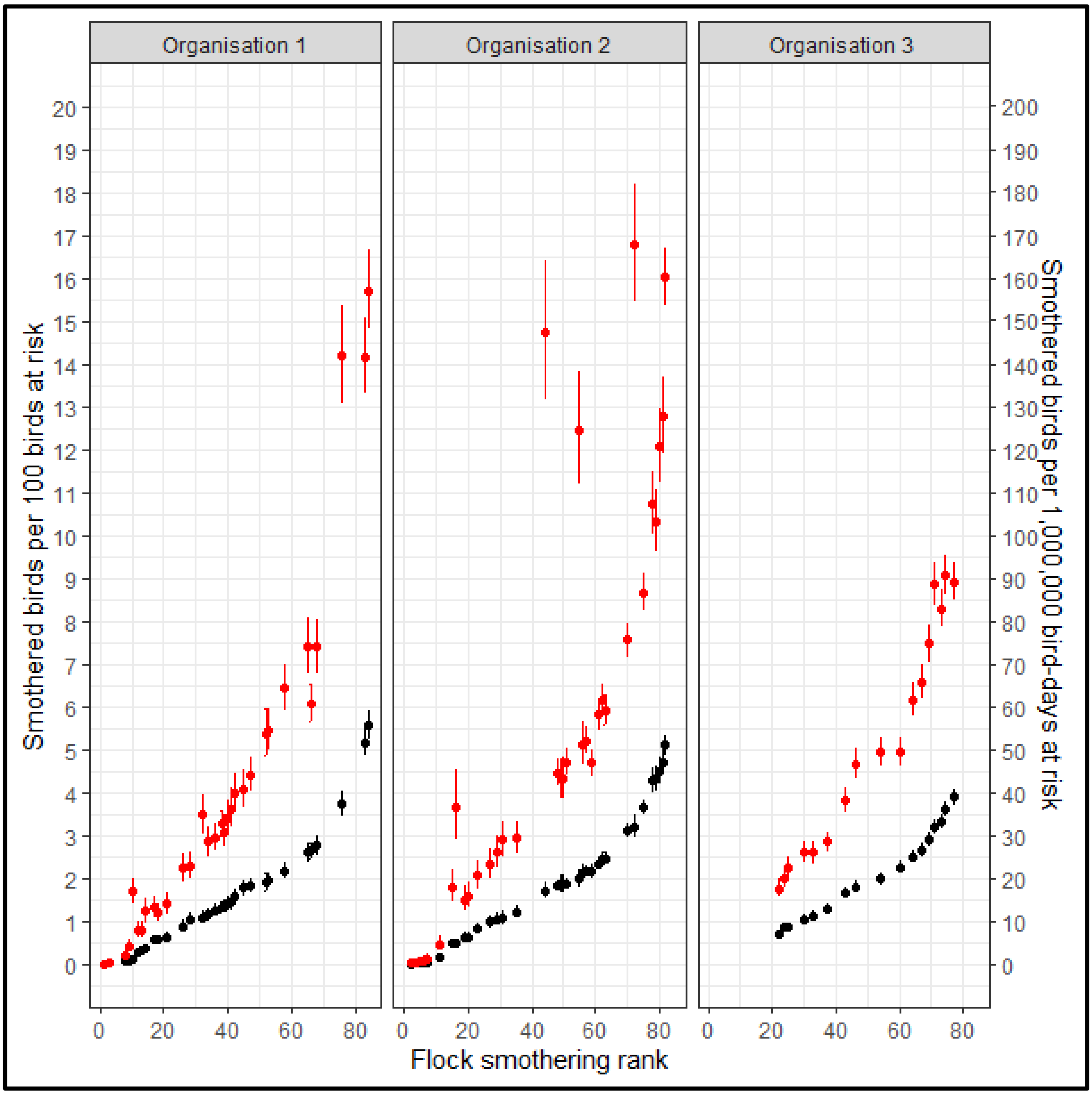

Figure 5 **Error bar plots showing: (a) the number of smothering deaths per 100 birds at placement (and their 95% confidence intervals) (black); and (b) the number of smothering deaths per 1,000,000 bird days at risk (red), by flock and organisation. Flocks within each organisation are sorted in order of smothering incidence risk, across all flocks.**

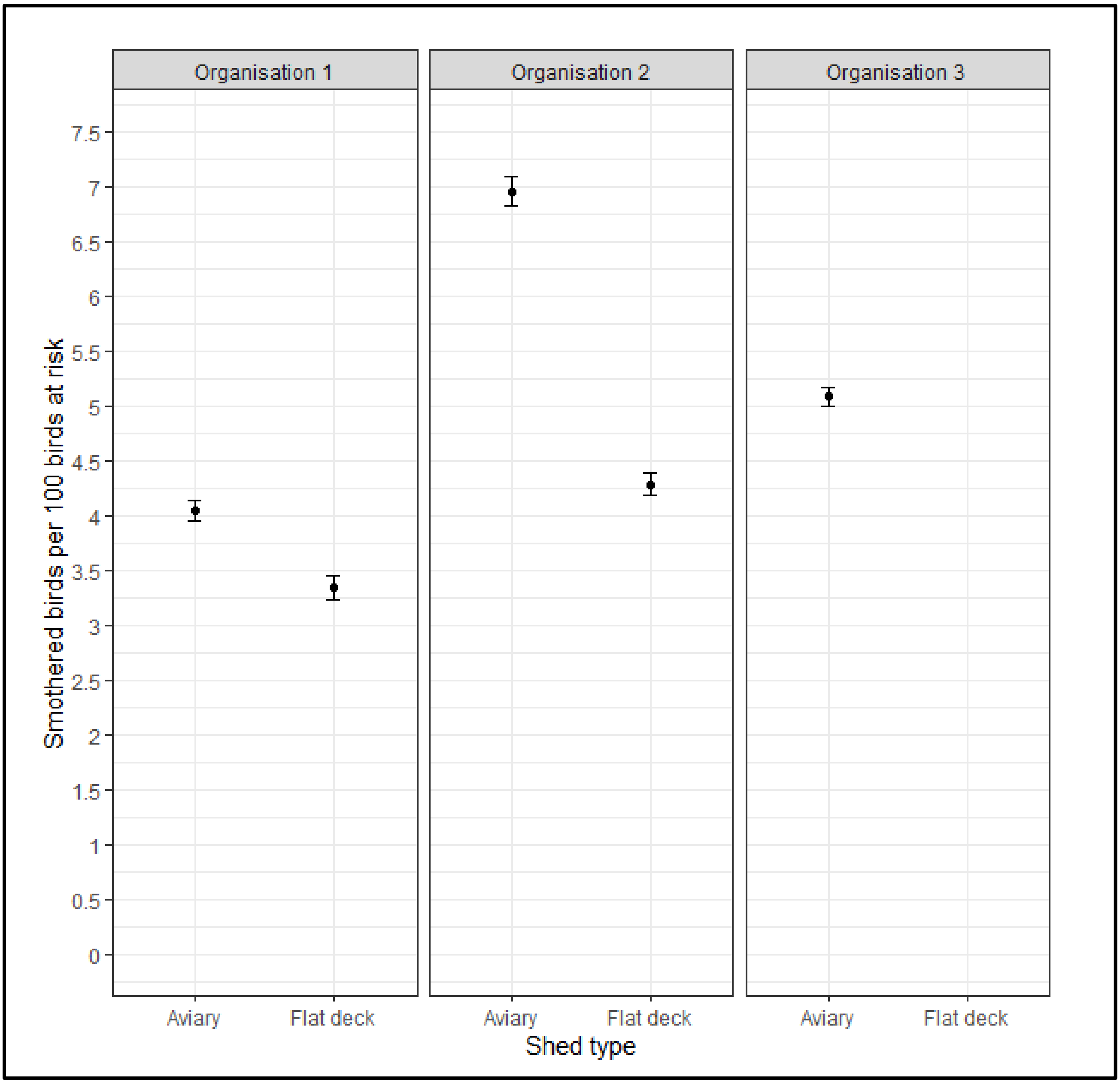

Figure 6 Error bar plots showing the number of smothering deaths per 100 birds at placement (and their 95% confidence intervals) by shed type (aviary, flat deck) and organisation. Organisation 3 had no flat deck sheds.

Differences in smothering risk between organisation, flocks, shed, indoor and outdoor environment was calculated using the Kaplan-Meier survival technique (Appendices 1–4).

3.2 Cox proportional hazard regression analysis

Candidate risk factors identified in the Kaplan-Meier survival analyses (Appendices 1-4) were assessed using a stratified, multilevel Cox proportional hazards regression model. The results from the Cox proportional hazards regression analyses are presented in Table 7. Estimated hazard ratios and their 95% confidence intervals (from Table 7) – that is, the increase or decrease in the daily probability of death by smothering for birds with a given explanatory variable compared with a stated reference group – are presented as an error bar plot in Figure 7.

Table 7 Regression coefficients and their standard errors from a stratified, multilevel Cox proportional hazards regression model of factors associated with the daily hazard ('risk') of smothering

Explanatory variable	Coefficient (SE)	P-value	Hazard ratio (95% CI)
Shed type:			
Flat deck	Reference	-	1.00
Aviary	0.9137 (0.6842)	0.18	2.50 (0.65 to 9.5)
Shed walks per day: [a]			
2	Reference	-	1.00
3	-0.3759 (0.0322)	<0.01	0.69 (0.64 to 0.73)
≥4	-0.5204 (0.0490)	<0.01	0.59 (0.54 to 0.65)
Eggs per bird per day (x 0.1):	0.3067 (0.0098)	<0.01	1.40 (1.30 to 1.40)
Human test score:			
<0	Reference	-	1.00
≥0	0.2894 (0.0975)	<0.01	1.30 (1.10 to 1.60) [b]
Novel object test score (indoor):			
<0	Reference	-	1.00
≥0	0.9524 (0.0619)	<0.01	2.60 (2.30 to 2.90)
Novel object test score (outdoor):			
<0	Reference	-	1.00
≥0	0.1833 (0.0490)	<0.01	1.20 (1.10 to 1.30)
Outdoor daily average temp (°C):			
Within 10–27 °C recommended range	Reference	-	1.00
Outside 10–27 °C recommended range	-0.8020 (0.0252)	<0.01	0.45 (0.43 to 0.47)
Outdoor daily average humidity (%):			
<70	Reference	-	1.00
≥70	0.6614 (0.0157)	<0.01	1.90 (1.90 to 2.00)
Random effect:	Variance		
Shed	2.3226		

[a] The number of times the shed is 'walked' (i.e., checked) by staff each day.

[b] Interpretation: After adjusting for the effect of other variables in the model, birds that had a lower fear of humans (i.e., flocks with high human test scores) had 1.30 (95% CI 1.10 to 1.60) times the daily hazard of smothering compared with birds that had a greater fear of humans.

[c] Interpretation: After adjusting for the effect of other variables in the model, birds that had a higher fear of the novel object (i.e., flocks with high novel object test scores) indoors and outdoors had 2.60 (2.30 to 2.90) and 1.20 (1.10 to 1.30) times the daily hazard of smothering, respectively compared with birds that had a lower fear of the novel object.

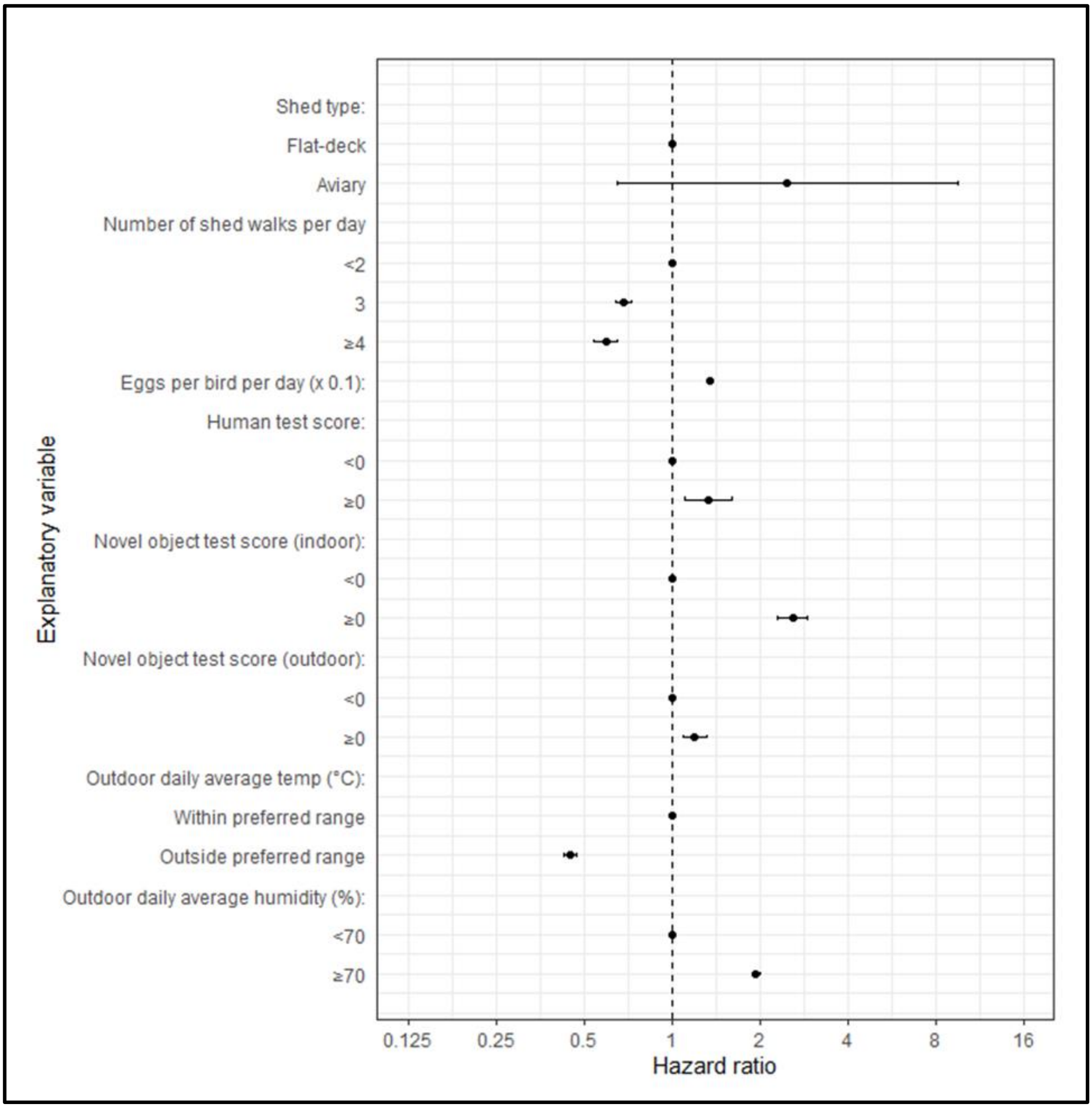

Figure 7 Error bar plot showing the point estimate (and 95% confidence intervals) for each of the hazard ratios for each of the explanatory variables shown in Table 7. A hazard ratio greater than one indicates a positive relationship between the variable and the daily hazard ('risk') of smothering.

3.3 Video observations on piling behaviour

Characteristics of 75 piles were analysed across 12 days of footage. There was only one day in the observations in which piling did not occur. Mean pile duration was 21 min 17 sec (range 1 min 11 sec to 3 h 44 min 28 sec) and the mean maximum number of hens in the pile was 64.4. Piles were observed to occur throughout the day from as early as 0716 h to as late as 1632 h. The number of hens in the pile increased with pile duration (Figure 8); in addition, more hens were recorded in the piles that occurred in the afternoon. Two of the observed 75 piles resulted in smothering of 4 hens in total.

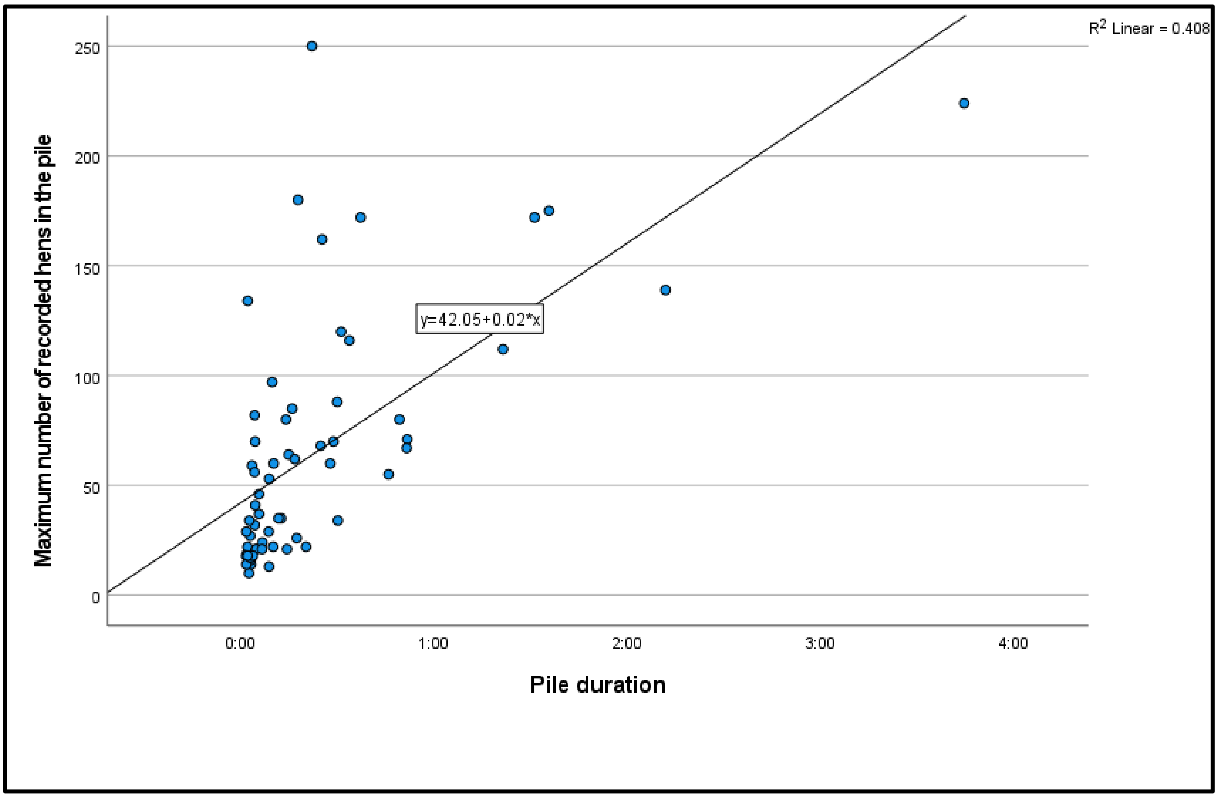

Figure 8 Scatterplot showing the relationship between pile duration and the maximum number of hens in a pile

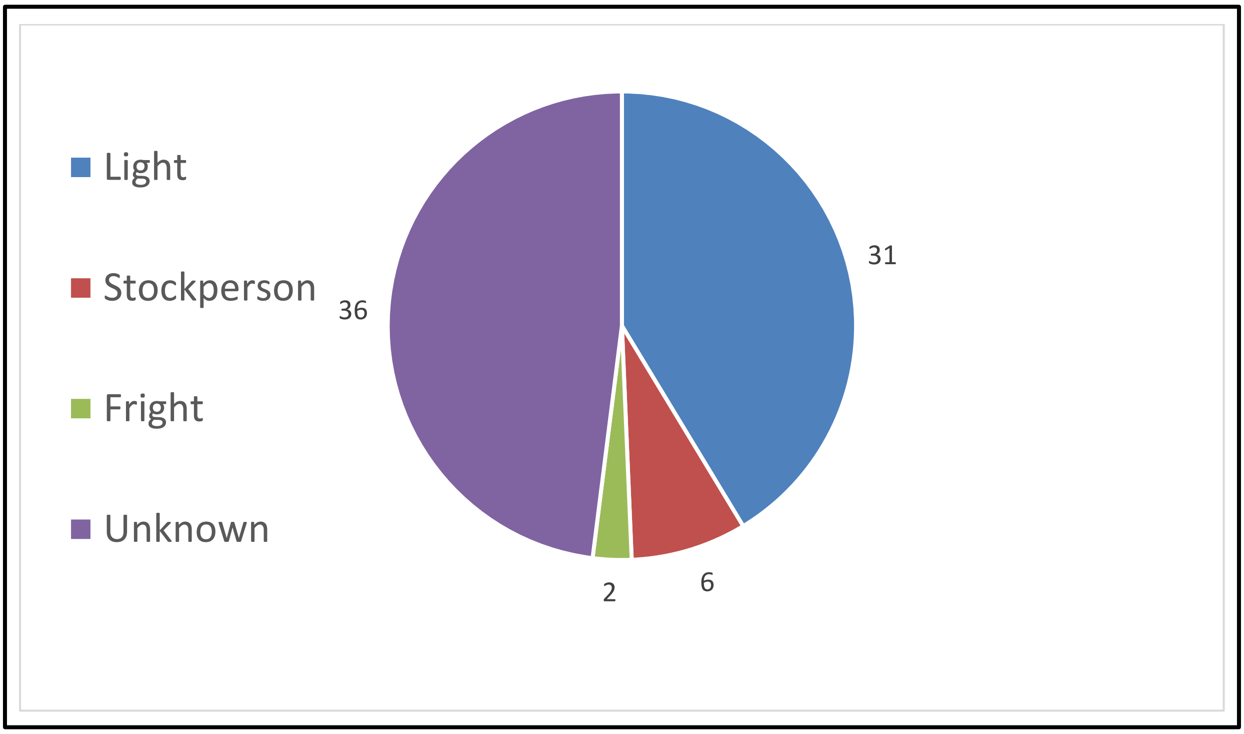

Figure 9 Pie chart showing the relative frequencies of the potential auditory and visual factors associated with piles. The most frequent recognisable factor was shards of light on the litter (41%), however, factors associated with a larger proportion of the piles (48%) could not be determined.

4 Discussion

The results of this study indicate marked differences in smothering risk by organisation (Figure 10, Appendix 1), with Organisation 1 having the lowest risk (1.28 smothering deaths per 100 birds placed) followed by Organisation 3 (2.09 per 100 birds placed) and Organisation 2 (2.11 per 100 birds placed). There was also a marked difference in smothering risk by breed (Figure 11, Appendix 1) with ISA Brown flocks having a lower risk of smothering compared with Hy-Line Brown (HB) flocks and the mixed breed flocks having a higher risk of smothering compared with Hy-Line Brown flocks, however there were very few ISA Brown and mixed breed flocks. It is more than likely that this difference between breeds was confounded by organisation. Organisation 1 had the lowest smothering risk overall and, while all organisations had HB, Organisation 1 was the only organisation that had ISA and mixed breed flocks. Breed differences have been observed in the piling behaviour of laying hens (see Winter et al. 2021) and in the occurrence of nest box smothers reported by flock managers (see Rayner et al. 2016), and while breed in the present study was confounded by organisation, further investigation into genetic strain effects on smothering are warranted.

Smothering risk was associated with shed type (aviary sheds had a higher risk); the number of shed walks per day (fewer shed walks per day, higher risk); the number of eggs produced per bird per day (more eggs per bird per day, higher risk); human test score (lower fear/higher curiosity of humans, higher risk); indoor and outdoor novel object test score (higher fear/lower curiosity of novelty, higher risk); outdoor daily average temperature (temperatures outside the 10-27°C range, lower risk) and outdoor daily average humidity (higher humidity, higher risk).

The shed-level random effect term included in the Cox proportional hazards model is a metric that quantifies unmeasured, shed-level characteristics associated with smothering risk. Used in this way a random effect term (by definition) is centred at zero and the numeric estimate of a shed's random effect term in itself has no inherent meaning. Useful inference from the random effect term comes from its variability. In situations where a random effect term has a large variance, our inference is that there is wide variation in shed-level smothering risk that has not been quantified by the explanatory variables included in the model. In situations where a random effect term has a relatively small variance (i.e., individual shed random effect terms are tightly clustered around zero when plotted as a frequency histogram), our inference is that the contribution of unmeasured shed-level effects on smothering risk is relatively small – that is, the (fixed) explanatory variable effects included in the model account for most of the variation in smothering risk. The shed-level random effect term from the Cox proportional hazards model developed for smothering risk was 2.32 (Table 7). This metric is relatively high and as explained above, this means that unmeasured shed-level effects were determinants of smothering risk, in addition to the explanatory variables listed in Table 7. A potentially profitable area of future investigation will be to compare the management practices and physical characteristics of the 'high smothering risk' sheds (i.e., those with a shed-level random effect term greater than zero) with those of the 'low smothering risk' sheds (those with a shed-level random effect term less than zero) to identify some of the more subtle determinants of smothering risk in free range layer poultry.

Furthermore, in considering the factors identified as risks of smothering (i.e., the results of the Cox proportional hazards model), strong candidates for future randomised experiments on smothering risk include the number of shed walks per day; high levels of egg production and manipulation of shed environment features (e.g., closure of the pop holes when outside daily average humidity is expected to be greater than 70%).

The effect of candidate interventions could be assessed using a classic randomized block design

(Cochran and Cox, 1957) with 'treatments' applied at the block (i.e. flock) level. In such experiments the inclusion of behavioural observations (for example on the distribution and movement of birds indoors and outdoors) may assist in understanding the causal nature between smothering risk and the environmental risk factor studied, which in turn may lead to effective management and environmental design solutions. For example, smothering may be more likely to occur on the range when ranging conditions are optimal (Hugelund et al., 2005; Richards et al., 2012) and there are more birds on the range (i.e. congestion near the pop holes) which may be mitigated by appropriate structures increasing ranging uniformity (Rault et al., 2013).

Outdoor daily average temperatures outside of the recommended range of 10 to 27 °C was (somewhat counterintuitively) associated with 0.45 (95% CI 0.43 to 0.47) times the daily hazard of smothering compared with days when outdoor daily average temperatures were within the 10 to 27 °C range. Only a relatively small proportion of the 32,442 production days monitored in this study (5232 of 32,442, 16%) were designated as days on which average temperatures were outside of the recommended range. It would be of interest to determine details of how flock management was changed on these days (presumably in response to either low or high temperatures) because these management changes (e.g., increasing the number of shed walks, closing pop holes and/or use of misting) may have unintentionally resulted in a reduction in smothering mortality risk.

Our video observations at one free-range aviary farm found that only two of the observed 75 piles indoors resulted in smothering. These results are consistent with the finding reported by Winter et al. (2021) who concluded that only a small proportion of piles result in smothering deaths. The pile which resulted in the only smothering event observed by Winter and colleagues was of a much longer duration than the average pile in the study (128.3 minutes versus an average pile duration of 15.26 minutes) and involved a relatively small number of hens (n = 20). The authors suggested that the number of hens in the pile may not be as important as the duration. In the present study, the two piling events that resulted in smothering were 17.8 minutes and 29.2 minutes in duration, however, the maximum number of hens in the pile was 180. While it is clear that smothering deaths are an outcome of piling behaviour, what remains unclear is why some piles result in smothering while others do not (Winter et al. 2021).

Video observations on piling behaviour in one flock in the present study suggest that high levels of hen curiosity toward shards of light and humans, together with synchronicity of behaviour, may be associated with the formation of piles and thus possibly smothering events. The context in which piles lead to smothering is unknown, but hens that are attracted to shards of light and humans (i.e., are curious about these stimuli) may be susceptible to piling in the presence of these stimuli and thus suffocation in piles. The Cox proportional hazards model in the present study indicated that smothering risk was increased in flocks with less fear (or conversely, more curiosity) of the human stimulus, and more fear (or conversely, less curiosity) of the novel object. It is unclear why smothering risk would be associated with less hen fear of the human stimulus but increased hen fear of the novel stimulus. The novel object test is presumably assessing general or underlying fearfulness (an individual's propensity to be more or less easily frightened; Jones 1986; Boissy 1998), rather than stimulus-specific responses, while the human test aims to assess the human-specific response of poultry (Hemsworth and Coleman 2011). Therefore, a hypothesis that obviously requires testing is that while startling responses of hens to unfamiliar or novel stimuli may result in fleeing towards structures that provide shelter, which may result in piling, curiosity of humans may result in hens approaching and crowding near or as near as possible to humans, which also may result in piling. If this is the case, general fearfulness in hens may be predictive of panic smothers and curiosity of humans could be predictive of recurring smothers. It is also of interest that several authors have proposed that selection on high egg yields may have some side effects on hen behaviour. For example, it has been suggested that selection on high egg yields may have indirectly caused changes in general fearfulness and sociality (Dudde et al., 2018) and reduced motor coordination (Dudde et al., 2018). Therefore, both increases in general fearfulness and sociality associated with high egg production may lead to increased susceptibility to panic smothers, while reduced motor coordination associated with high egg production may lead to hens experiencing difficulty extracting themselves from a pile due to reduced mobility, which could ultimately put them at higher risk of smothering. As indicated earlier such hypotheses require rigorous testing.

While aviary sheds were associated with 2.5- (95% CI 0.65 to 9.5) fold increase in daily smothering hazard, we note ‘shed type’ as an explanatory variable was not statistically significant (at the alpha level of 0.05) as a risk factor for smothering. Our decision to retain shed type in the final model was based the relatively strong point estimate of smothering hazard for shed type and biological plausibility of shed type as a determinant of smothering risk. The relatively large confidence interval around our estimate of the hazard ratio for aviary shed type (0.65 to 9.5) is indicative of marked variation in smothering hazard across aviary sheds. A useful area for further investigation would be to identify possible reasons for this by comparing management and climatic characteristics of ‘high’ and ‘low’ smothering risk aviary sheds. This investigation may identify some common characteristics of high-risk aviary sheds across the three organisations.

In non-experimental research, it is difficult to provide conclusive evidence that a given explanatory variable (e.g. shed type or the number of shed walks per day) is a cause of an outcome of interest (Tabachnick and Fidell 2013), due to issues such as the timing of the occurrence of explanatory variable events in relation to the outcome and unobserved confounding (Tchetgen 2013). This said, the value of the work presented here lies in the fact that we have collected detailed information for a wide range of variables hypothesised to explain smothering risk and have identified a subset associated with smothering, with the probability that the identified associations occurring by chance was unlikely (i.e., less than 0.05) and controlling for the effect of known confounders such as organisation and flock. A profitable area of future investigation would be to modify these identified variables in a controlled experiment to determine the subsequent effect on smothering incidence.

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6 Plain English Summary

Project Title:	**What causes smothering in commercial free range laying hens? Part 1**
Australian Eggs Limited Project No	1HS901UM
Researchers Involved	P.H. Hemsworth, M. Stevenson, A.D. Fisher, P.S. Taylor, P. Chowdhury, M. Rice, and R. Galea
Organisations Involved	Animal Welfare Science Centre, Faculty of Veterinary and Agricultural Sciences The University of Melbourne Parkville VIC 3010 Australia
Phone	+61 3 8344 8383
Fax	-
Email	phh@unimelb.edu.au
Objectives	This prospective observational study at both the flock- and farm-level in Australian free-range conditions was conducted to identify the major potential risk factors in relation to smothering in laying hens.
Background	Smothering in poultry occurs when birds mass together, often on top of each other, resulting in death from suffocation, and estimates by Australian free range egg producers indicate that smothering may account for up to 30 to 40% of total hen mortality. While smothering has both economic and welfare-related concerns, little is known about smothering in laying hens.
Research	Eighty-six commercial barn and aviary free range flocks were studied during lay. Shed and flock data, such as flock size, shed characteristics and environmental conditions and management practices, were used to examine relative smothering risk.
Outcomes	A Cox proportional hazards regression model, which provides appropriate estimates of the strength of the association between each explanatory variable and smothering risk, adjusted for the effect of variables known to confound (distort) the association between the two (e.g. farm and flock), revealed that smothering risk was associated with type of housing; level of egg production; the ratio of water to feed offered; outside temperature and humidity; precipitation; the number of shed walks by staff per day; less fear (or conversely more exploration) of novelty and humans.
Implications	The findings from this research provide the necessary first steps to allow producers to implement interventions to reduce smothering risk in their flocks. A profitable area of future work would be to manipulate the modifiable risk factors listed above and to document the change in smothering incidence risk that followed. This would provide free-range egg producers with greater confidence that the risk factors identified in this report were, in fact, causal.

Key Words	Laying hens, free-range, smothering, risk factors, epidemiology, welfare
Publications	*Maxine Rice, Rutu Acharya, Jessalyn Taylor, Peta Taylor, Andrew Fisher, Paul Hemsworth (2022)* Behaviour of pullets and housing system predicts behaviour of adult laying hens in commercial free-range egg farms, 33rd Annual Australian Poultry Symposium, 7–9 February 2022, University of Sydney *Mark Stevenson, Prabal Chowdhury, Rutu Acharya, Maxine Rice, Jessalyn Taylor, Andrew Fisher, Peta Taylor, Paul Hemsworth (2022)* Smothering in commercial free-range laying hens, 33rd Annual Australian Poultry Symposium, 7-9 February 2022, University of Sydney *Maxine Rice, Rutu Acharya, Peta Taylor, Andrew Fisher and Paul Hemsworth (2021)* A comparison of avoidance and curiosity behaviour of laying hens in two free-range housing systems: aviary vs flat-deck, 8th International Conference 8th International Conference on The Assessment Of Animal Welfare At Farm And Group Level, 16-19 August, 2021, Virtual conference *Rutu Acharya, Maxine Rice, Prabal Chowdhury, Mark Stevenson and Paul Hemsworth (2021),* Ask the Egg-sperts! Flock managers’ views on smothering in free-range laying hens in Australia, 8th International Conference 8th International Conference on The Assessment Of Animal Welfare At Farm And Group Level, 16-19 AUGUST, 2021, Virtual conference *Maxine Rice, Rutu Acharya, Andrew Fisher, Peta Taylor and Paul Hemsworth (2020)* Characterising piling behaviour in Australian free-range commercial laying hens, ISAE 2020 Global Virtual Meeting *Maxine Rice, Rutu Acharya, Peta Taylor, Andrew Fisher and Paul Hemsworth (2020),* Smothers in free-range laying hens, Australasian Veterinary Poultry Association Conference, 20-21st May 2020, Virtual meeting

Appendix 1

Kaplan-Meier survival analyses of smothering mortality stratified by organisation

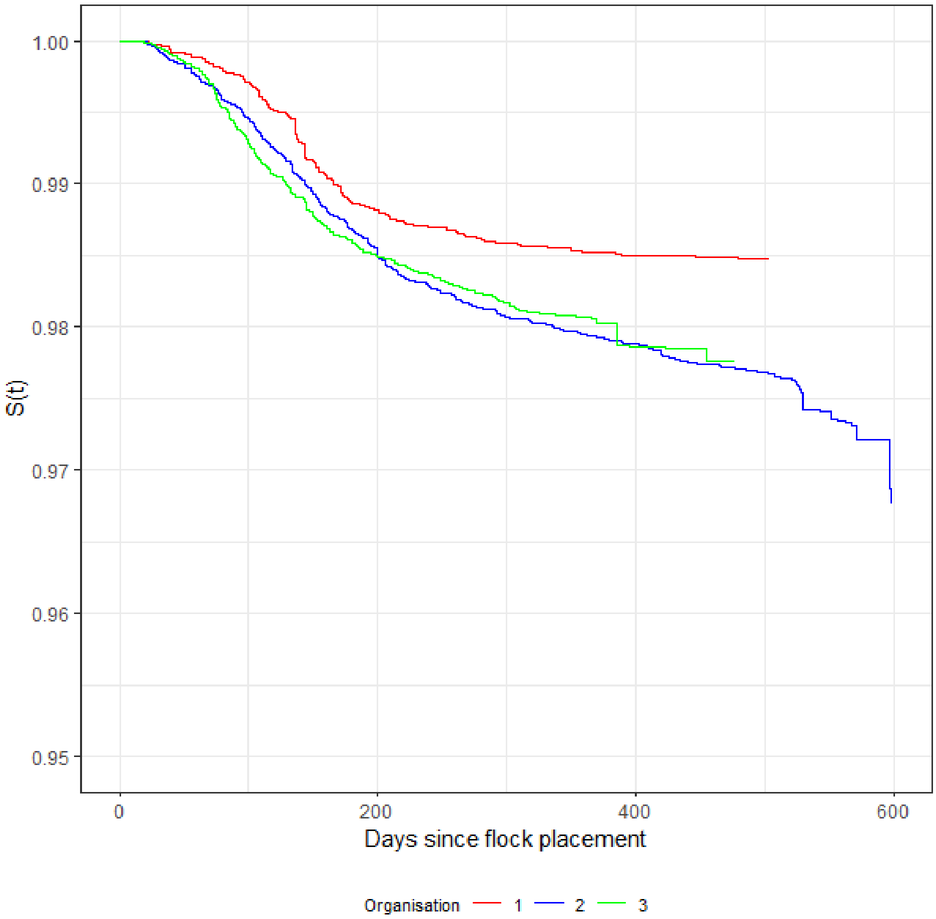

Figure 10 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by organisation. Log rank test statistic 1045; df 2; $p < 0.01$.

Appendix 2

Kaplan-Meier survival analyses of smothering mortality stratified by flock features

Breed

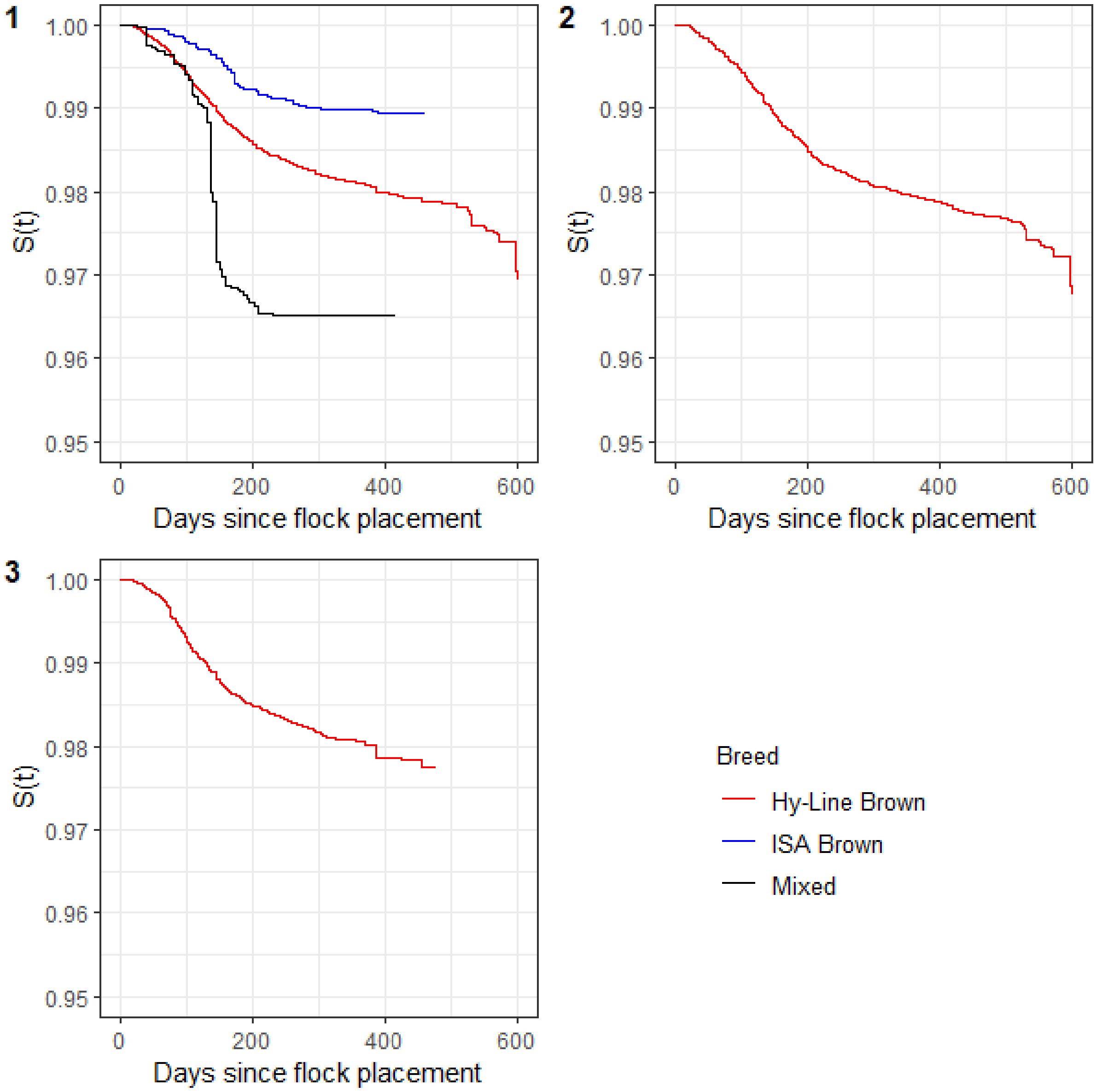

Figure 11 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by flock breed (Hy-Line Brown, ISA Brown and mixed) and organisation. Log rank test statistic 2708; df 2; p <0.01.

Order of flock in the study shed

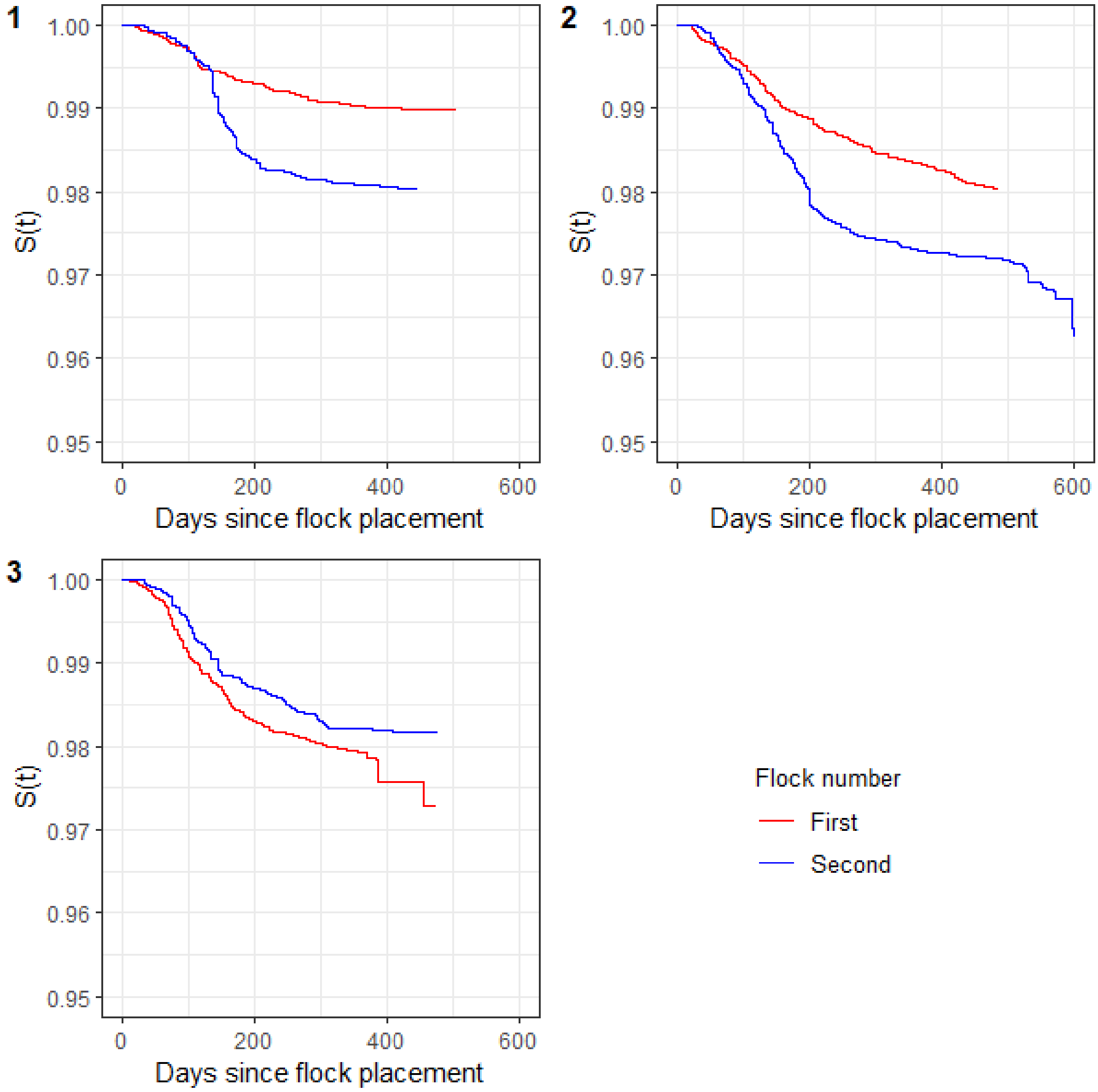

Figure 12 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by the order of flock in each study shed (first, second) and organisation. Log rank test statistic 852; df 1; p = 0.01.

Flock size at placement

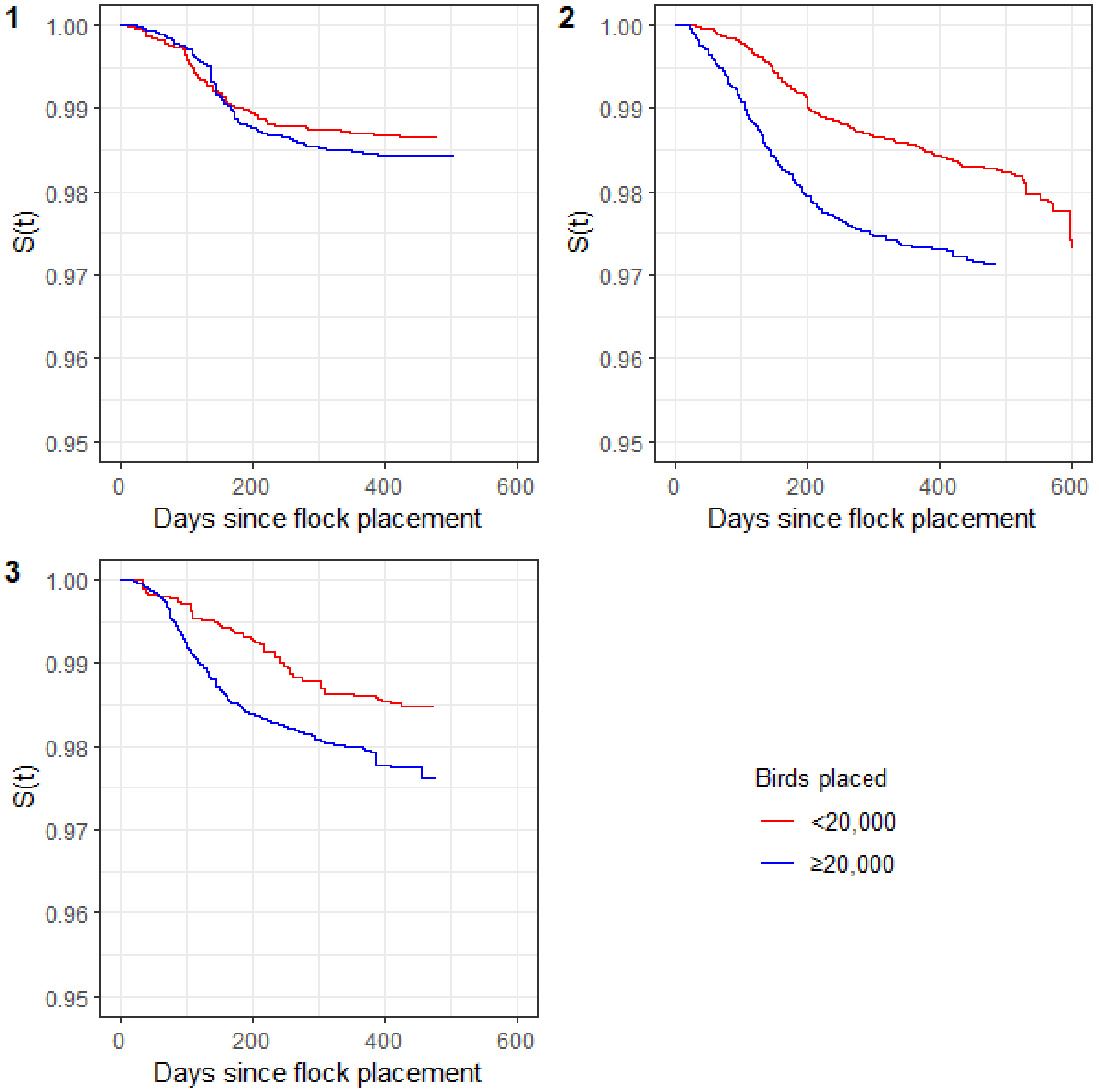

Figure 13 Kaplan-Meier survival curves showing the cumulative proportion of birds to yet experience death by smothering as a function of days since flock placement, stratified by flock size at placement (<20,000 birds and ≥20,000 birds) and organisation. Log rank test statistic 1169; df 1; $p < 0.01$.

Number of birds placed per square metre of floor space

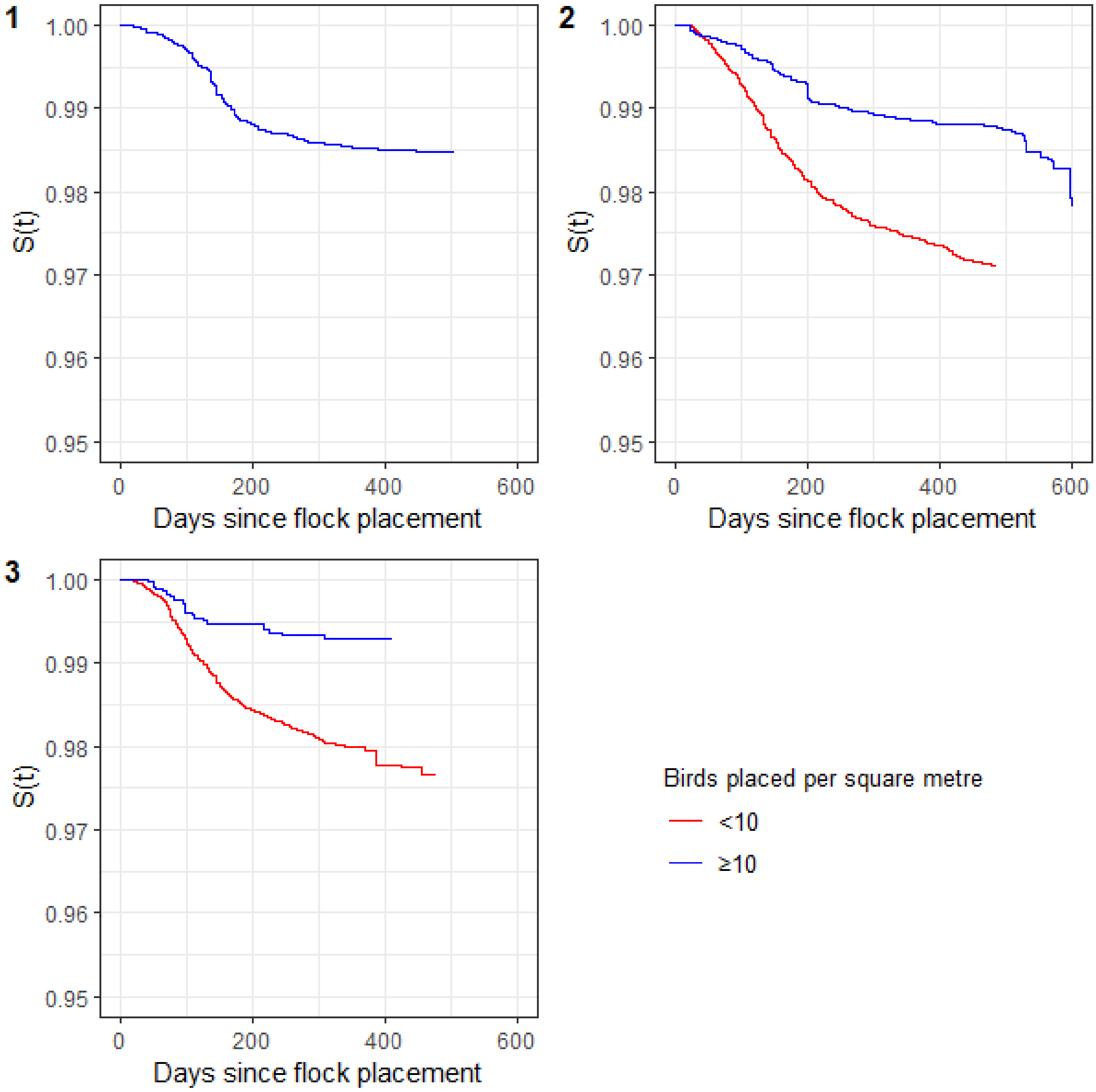

Figure 14 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by the number of birds placed per square metre of shed floor space at placement (< 10 and ≥10) and organisation. Log rank test statistic 2914; df 1; p <0.01.

Flock behaviour

Human test

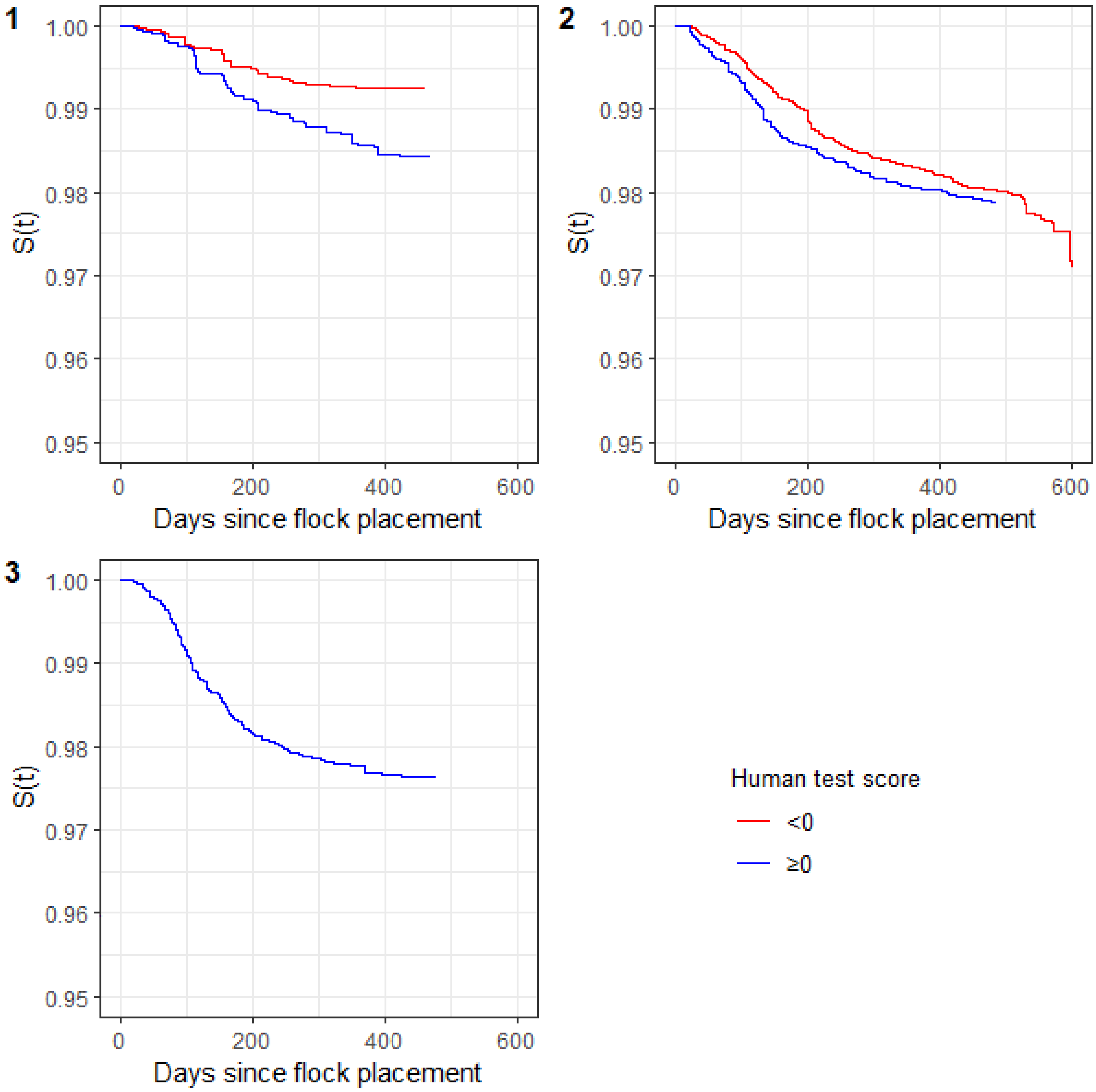

Figure 15 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by flight distance ‘human test score’ (<0 units and ≥0 units) and organisation. Log rank test statistic 261.4; df 1; p <0.01.

Novel object test (outdoors)

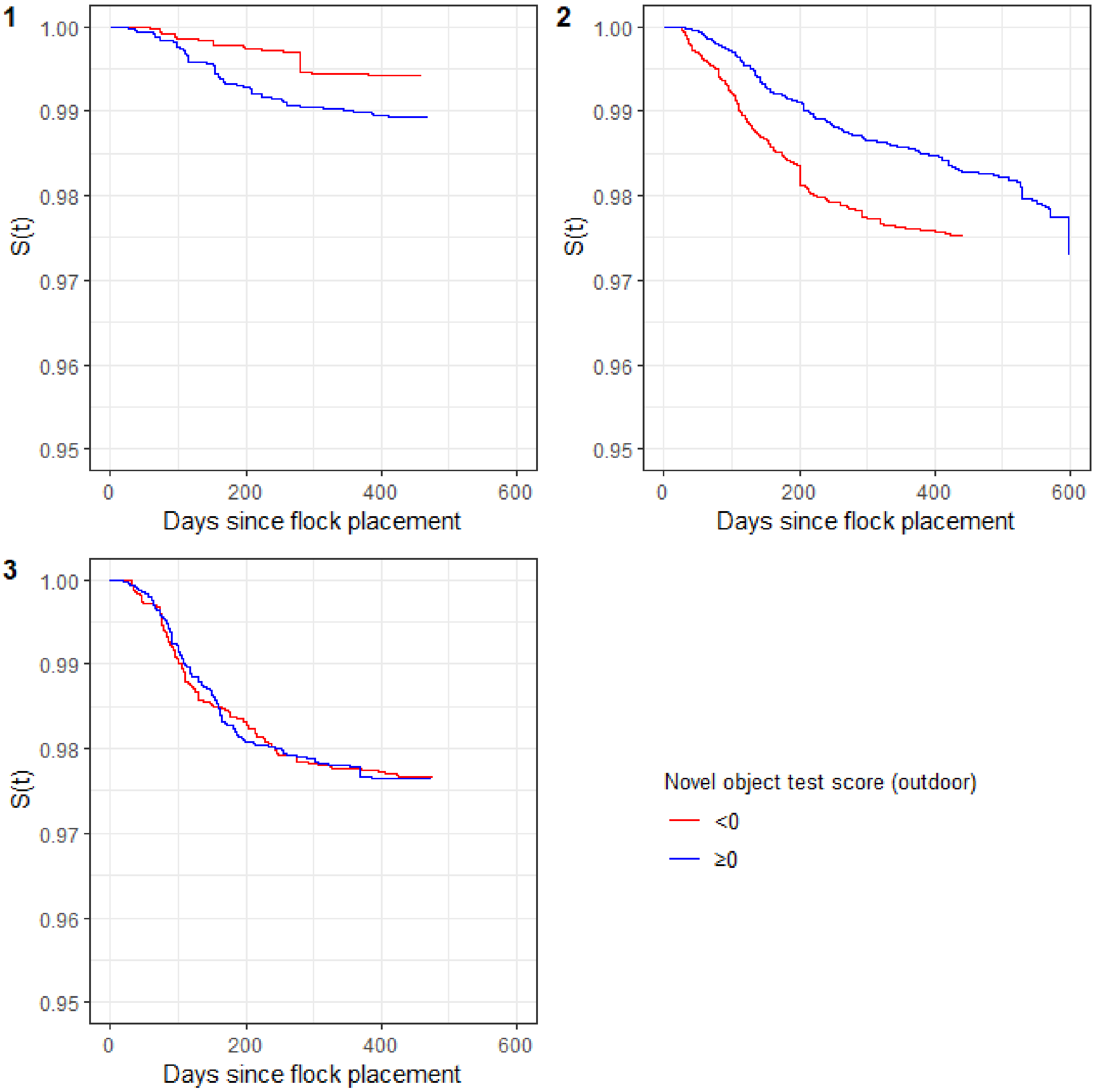

Figure 16 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by novel object test PCA score in range (<0 units and ≥0 units) and organisation. Log rank test statistic 663.1; df 1; p <0.01.

Novel object test (indoors)

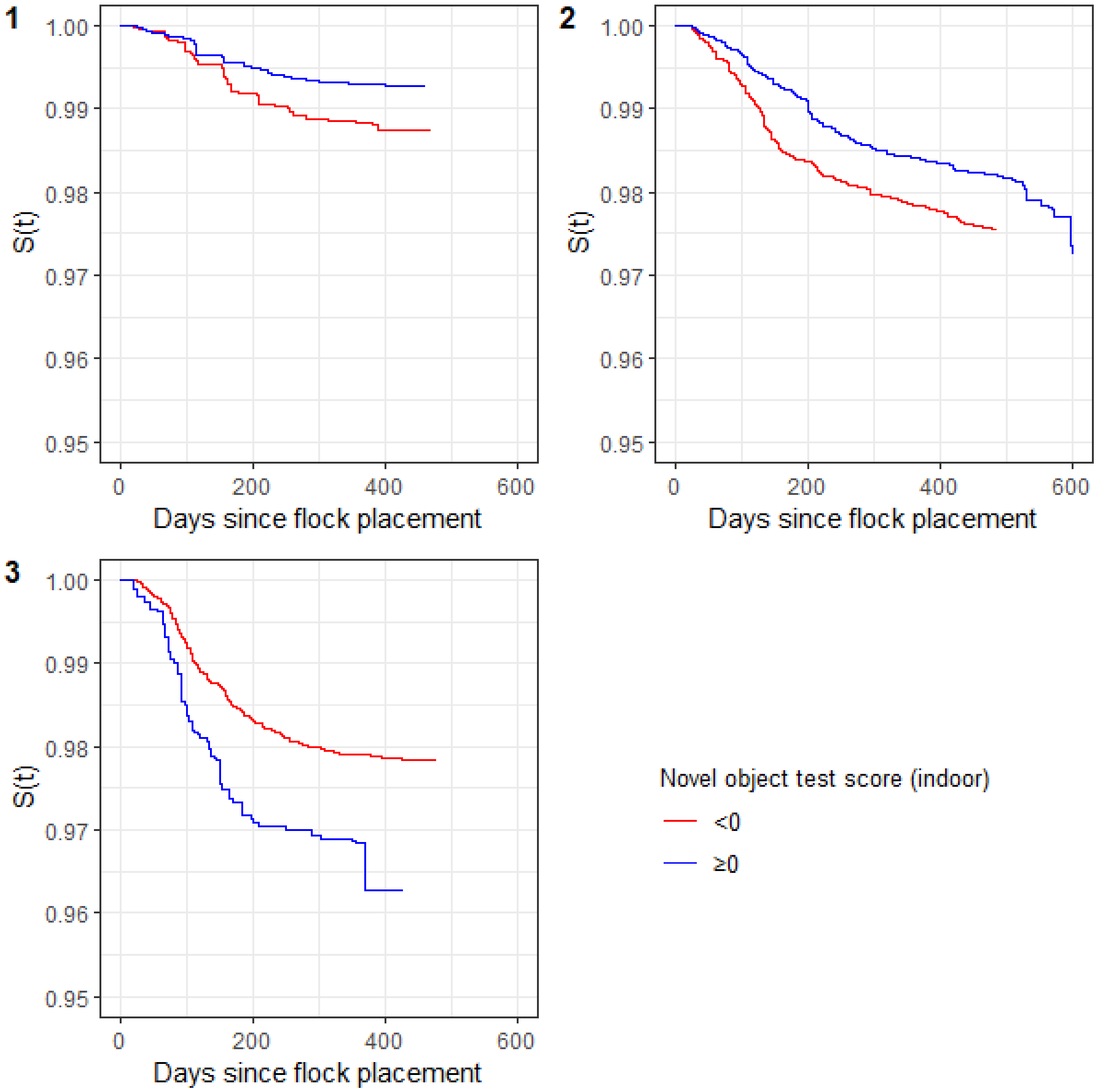

Figure 17 Kaplan-Meier survival curves showing the cumulative proportion of birds to yet experience death by smothering as a function of days since flock placement, stratified by novel object test PCA score in shed (<0 units and ≥0 units) and organisation. Log rank test statistic 64.3; df 1; p <0.01.

Shed manager’s perception of piling behaviour

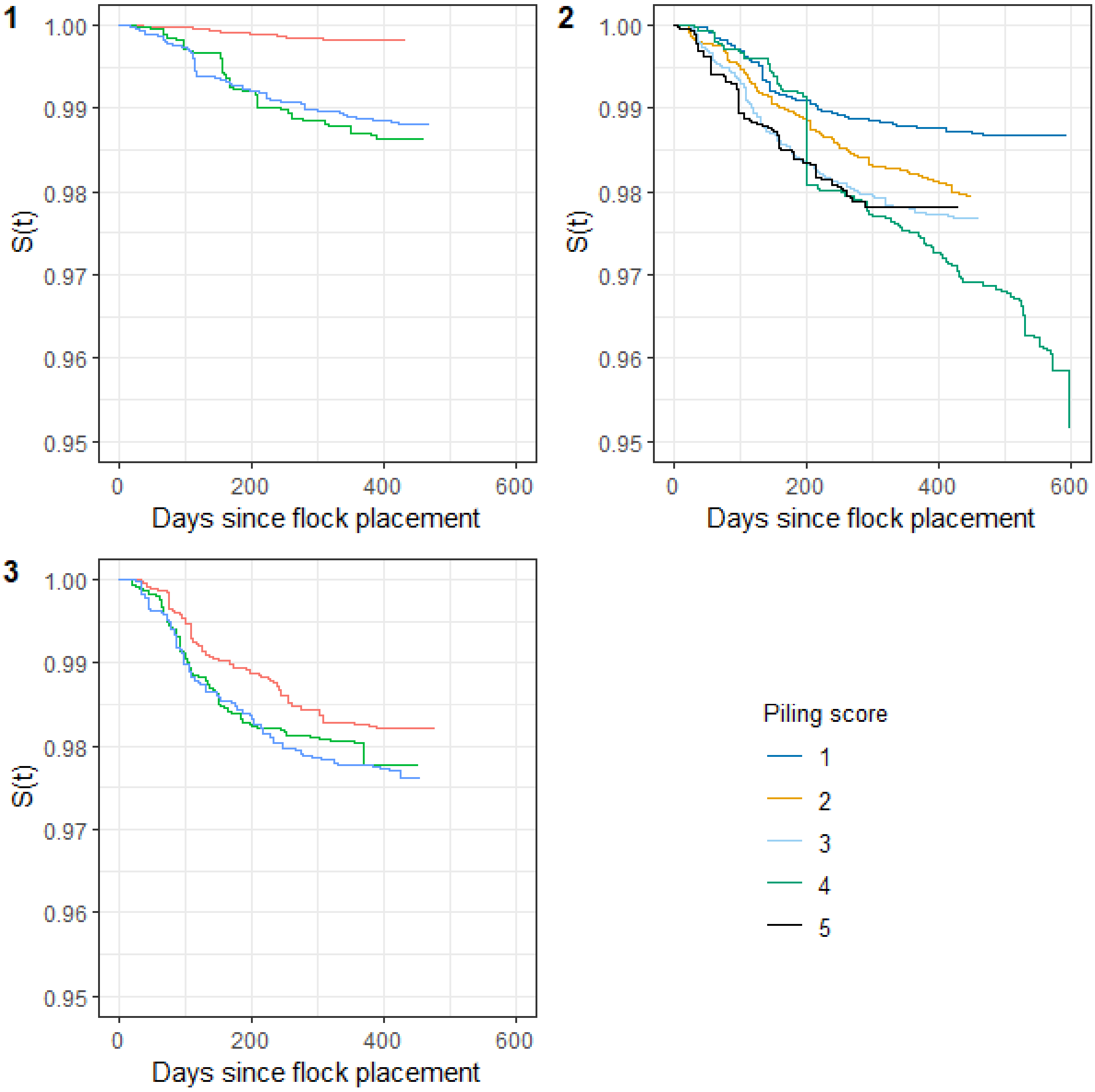

Figure 18 Kaplan-Meier survival curves showing the cumulative proportion of birds to yet experience death by smothering as a function of days since flock placement, stratified by shed manager’s perception of piling behaviour (1-5) and organisation. Log rank test statistic 1794; df 4; p <0.01.

Eggs per bird per day

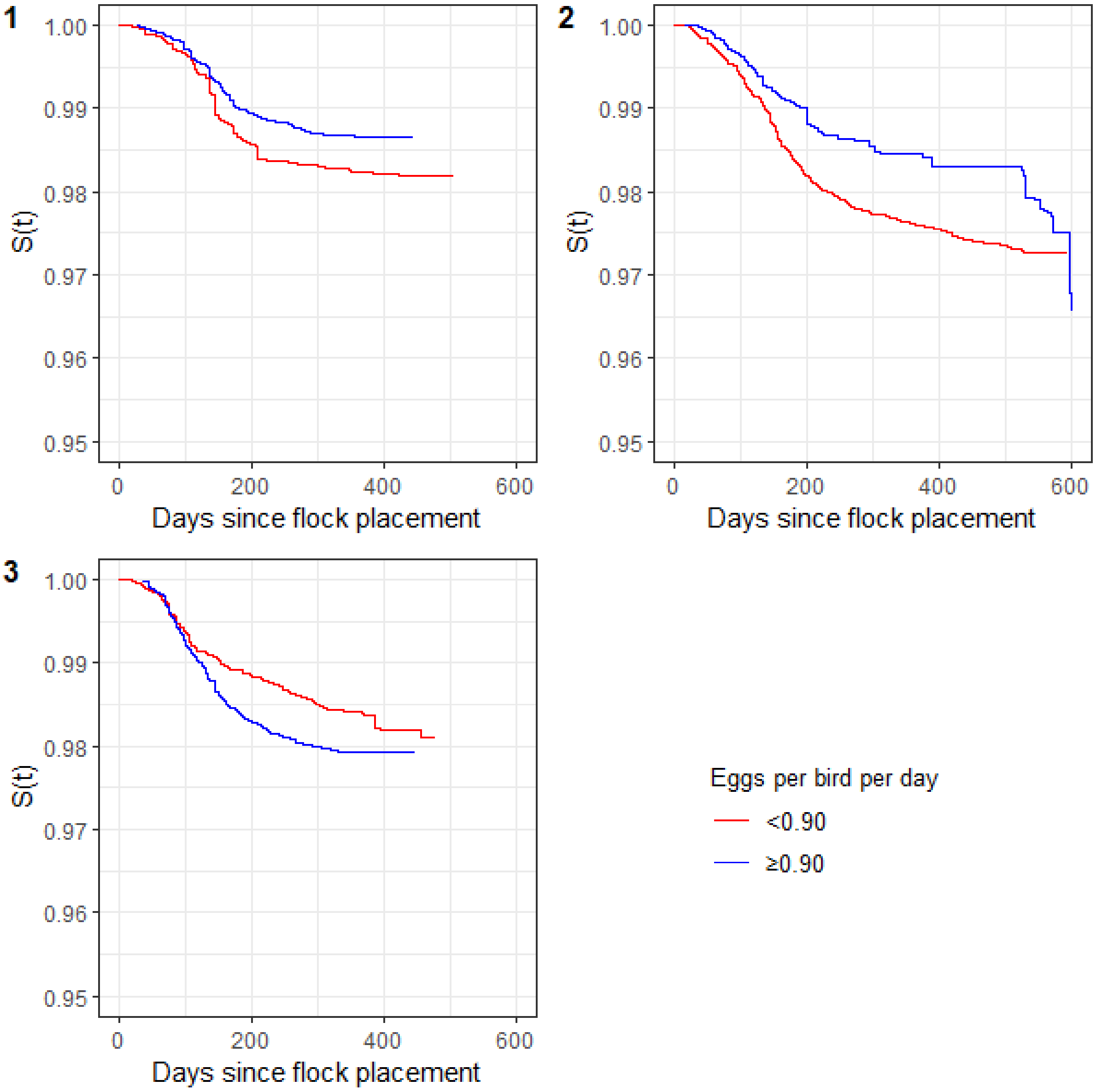

Figure 19 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by number of eggs per bird per day (<0.90 and ≥0.90) and organisation. Log rank test statistic 3834; df 1; p <0.01.

Waste eggs per bird per day

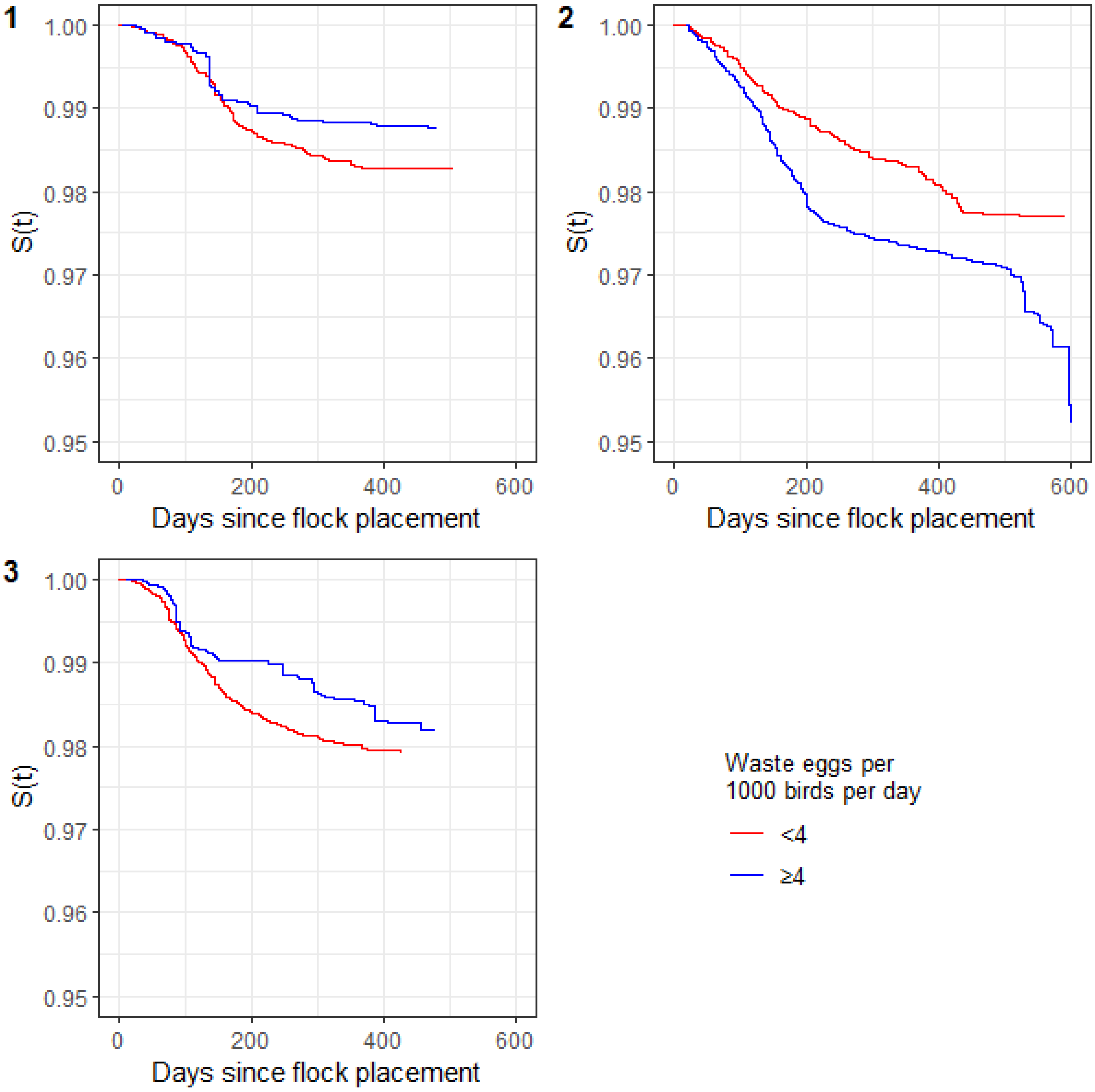

Figure 20 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by number of waste eggs per 1000 birds per day (<4 and ≥4) and organisation. Log rank test statistic 7197; df 1; p <0.01.

Dirty eggs per bird per day

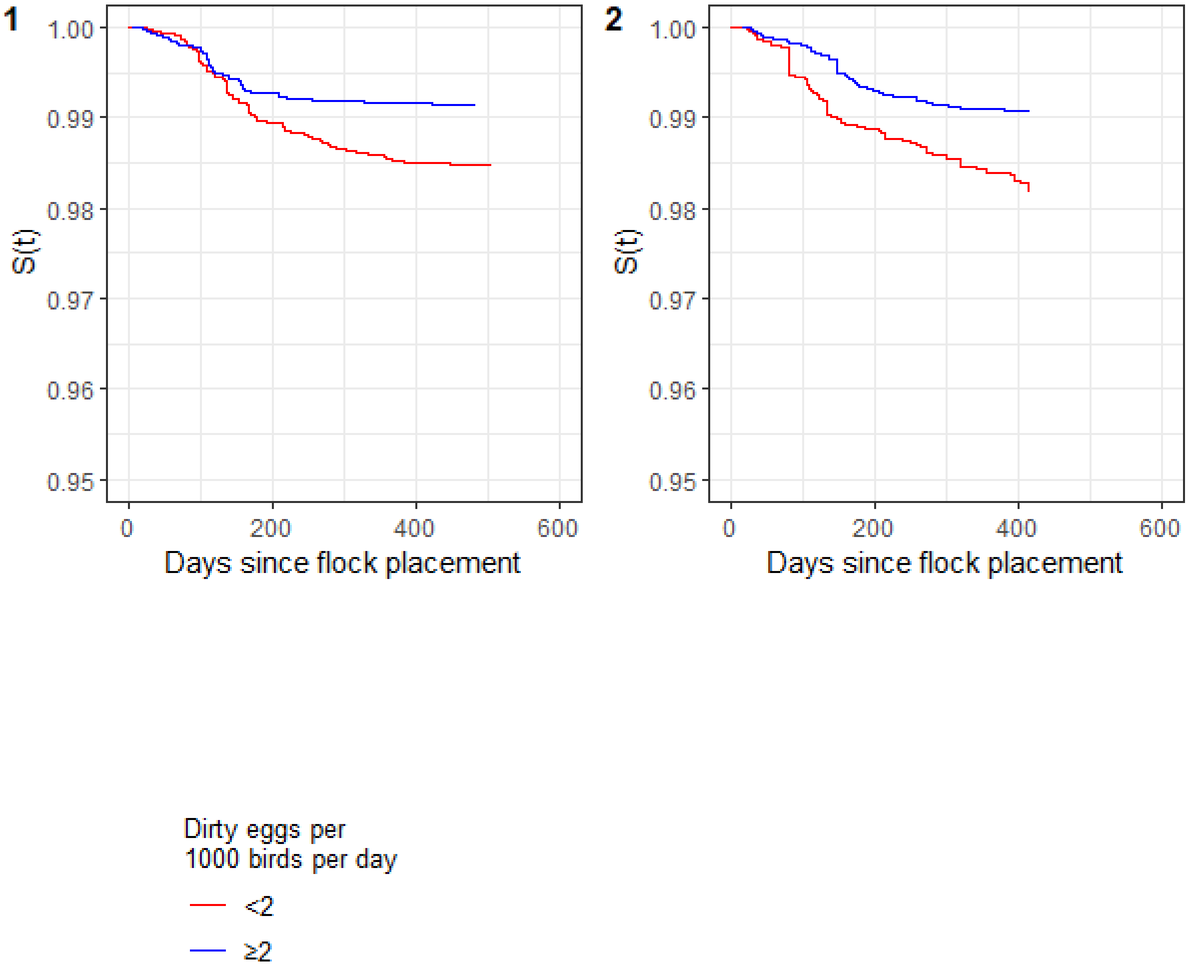

Figure 21 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by number of dirty eggs per 1000 birds per day (<2 and ≥2) and organisations 1 and 2. Organisation 3 did not record dirty eggs). Log rank test statistic 940.2; df 1; p <0.01.

Floor eggs per bird per day

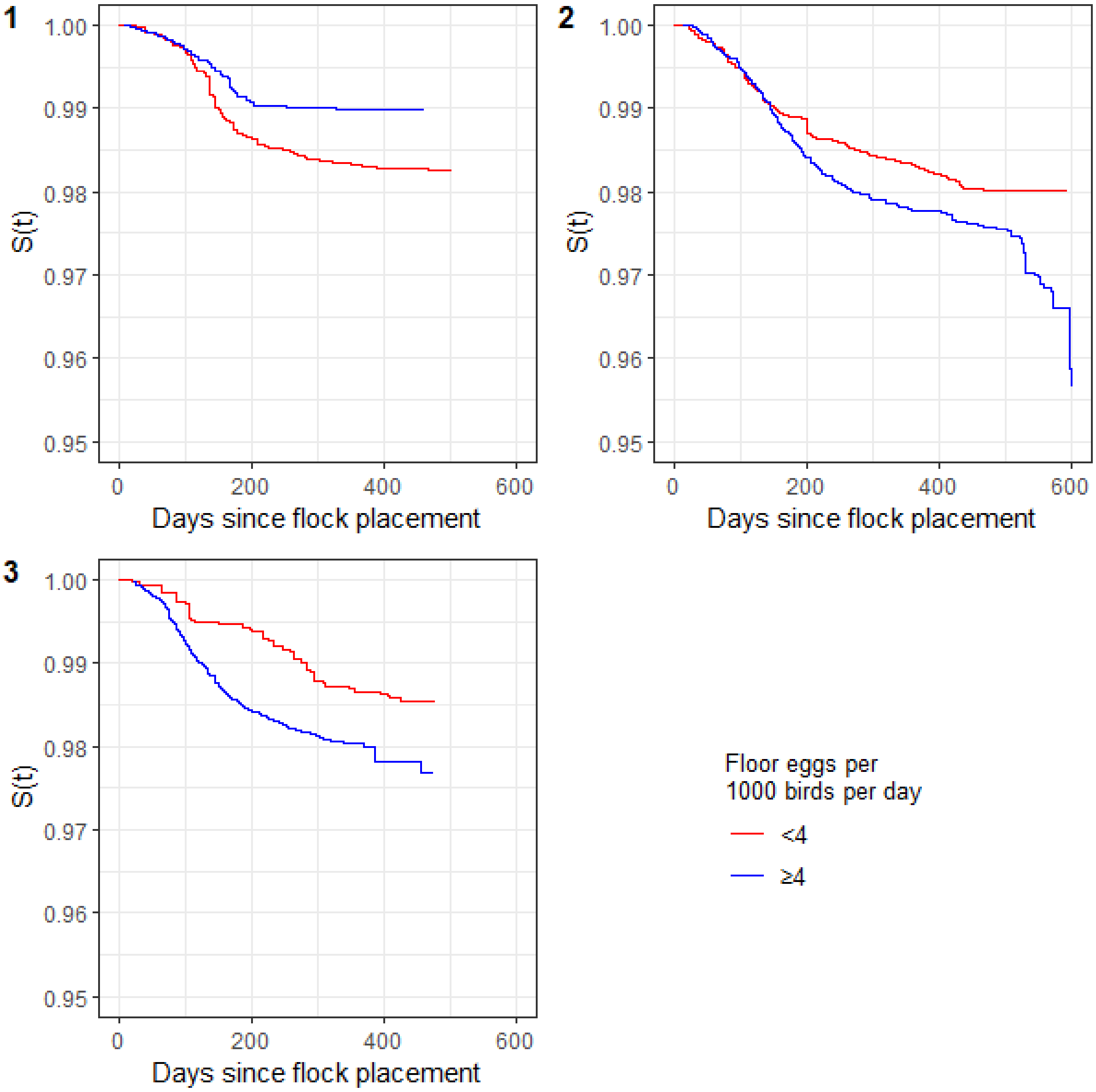

Figure 22 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by number of floor eggs per 1000 birds per day (<4 and ≥4) and organisation. Log rank test statistic 1282; df 1; p <0.01.

Water

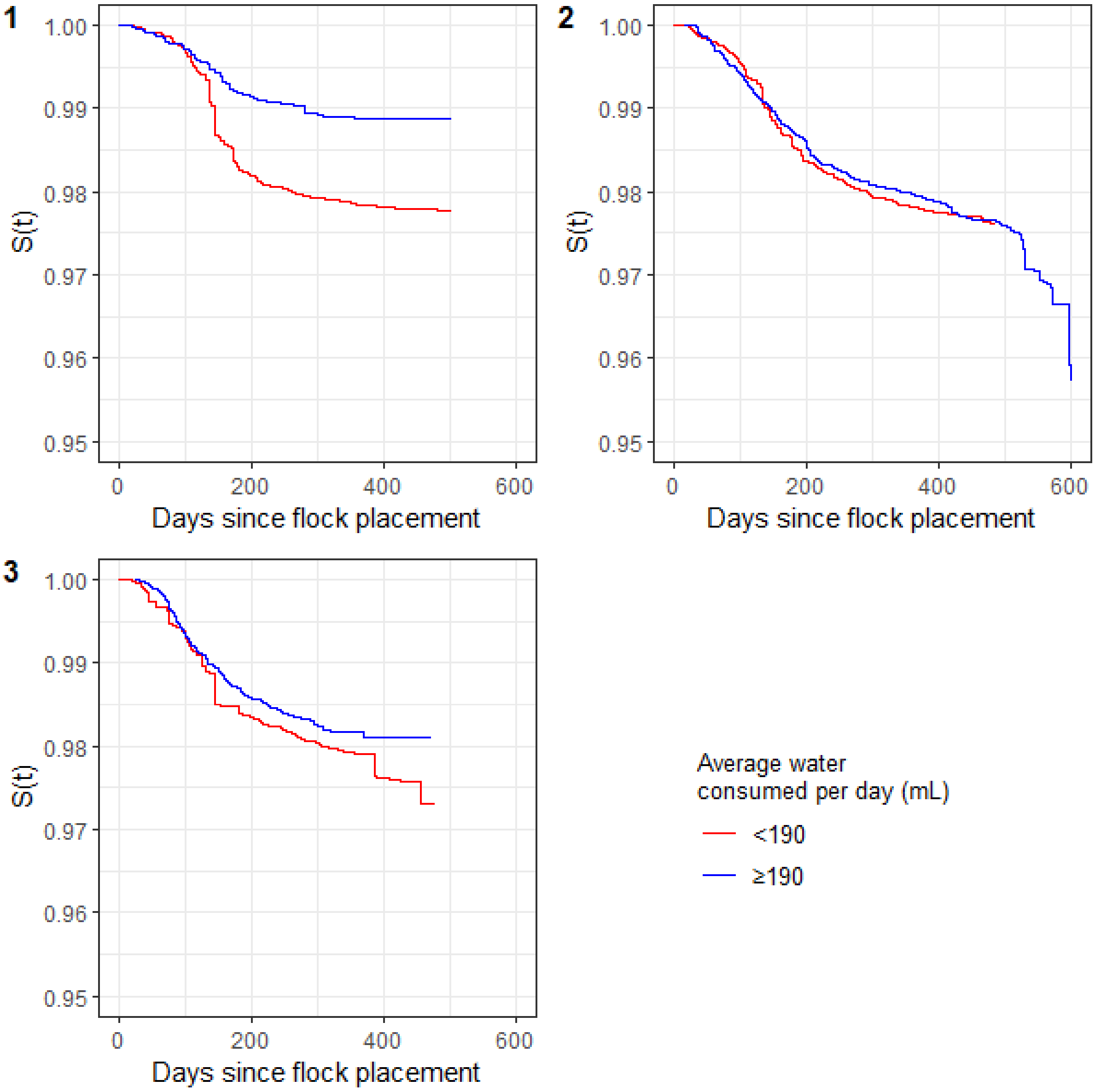

Figure 23 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by average amount of water consumed per bird per day (<190 mL and ≥190 mL) and organisation. Log rank test statistic 289.1; df 1; p <0.01.

Feed

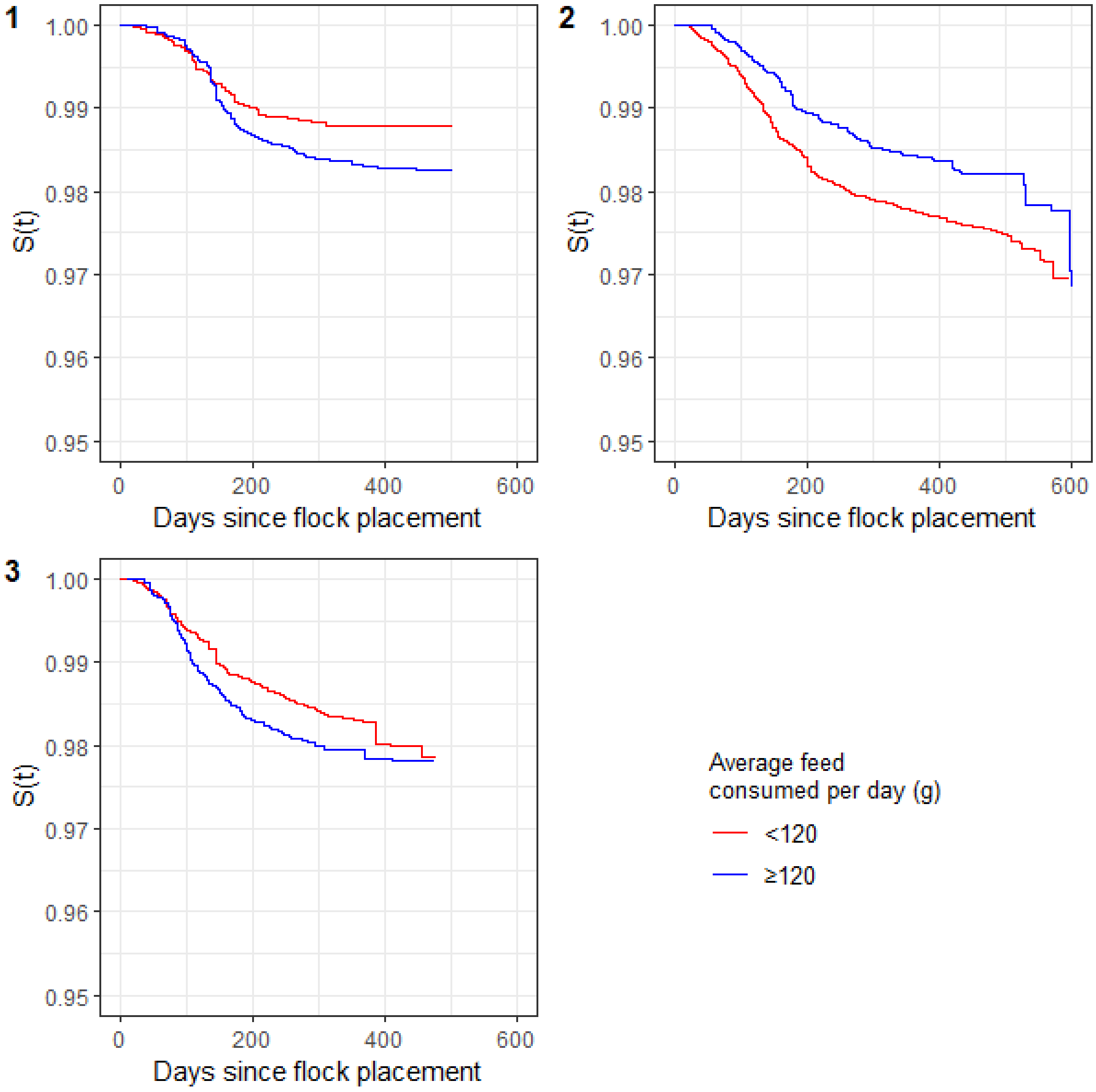

Figure 24 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by average weight of feed consumed per bird per day (<120 g and ≥120 g) and organisation. Log rank test statistic 24.98; df 1; p <0.01.

Water to feed ratio

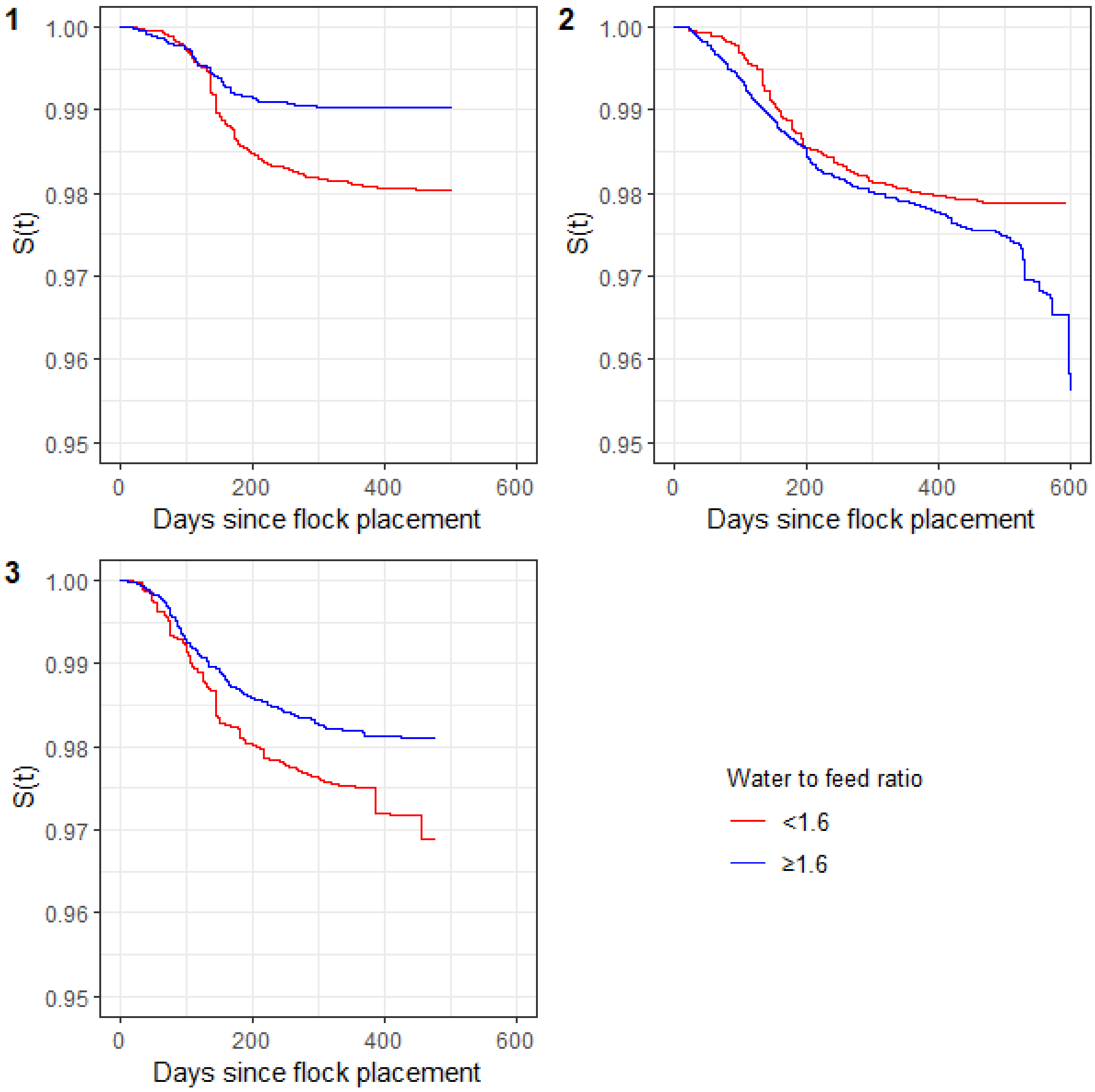

Figure 25 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by water to feed ratio per day (<1.6 mL/g and ≥1.6 mL/g) and organisation. Log rank test statistic 924.4; df 1; p <0.01.

Presence of disease in flock

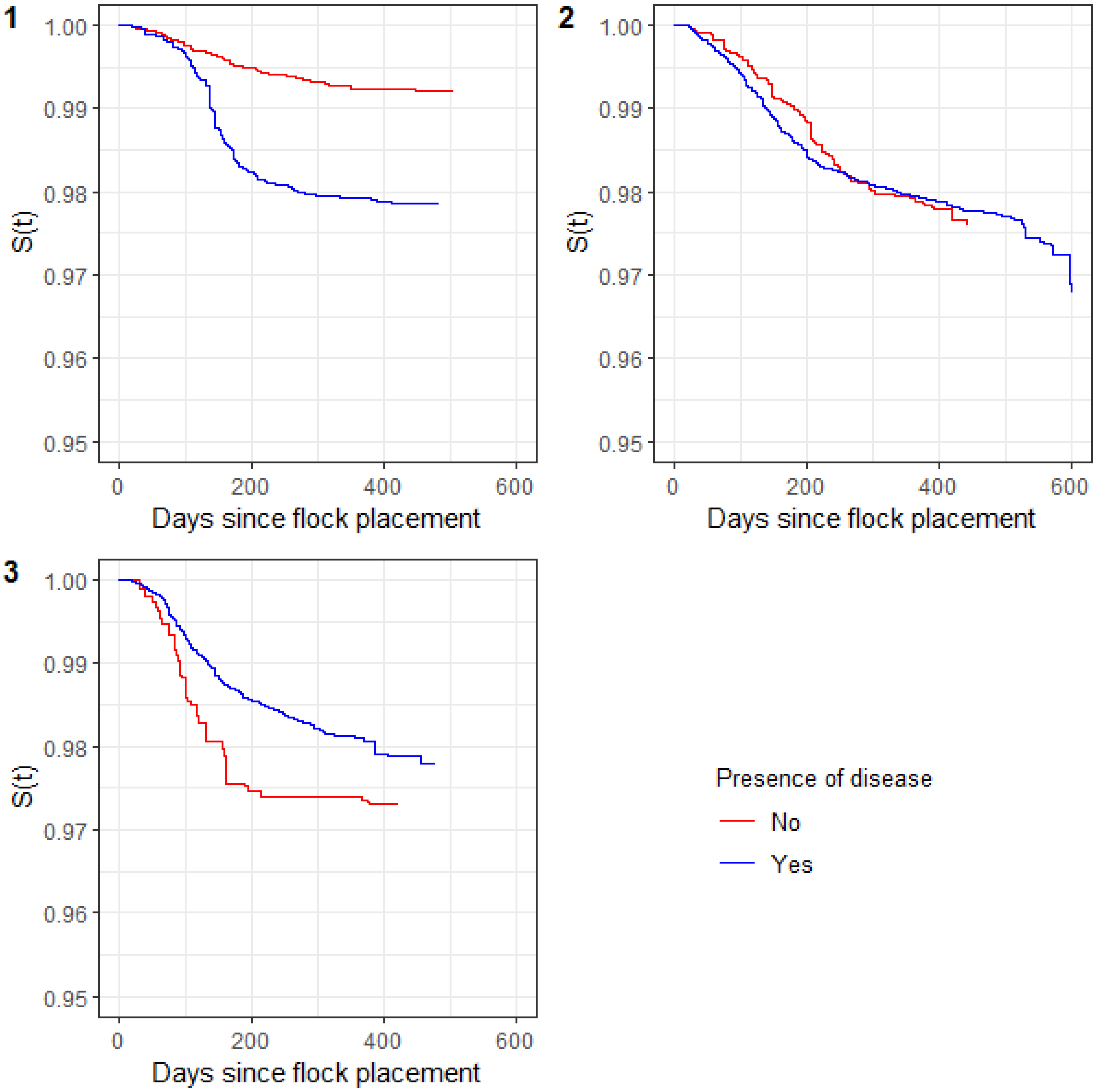

Figure 26 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by presence or absence of disease (no, yes) and organisation. Log rank test statistic 1387; df 1; p <0.01.

Staff per shed

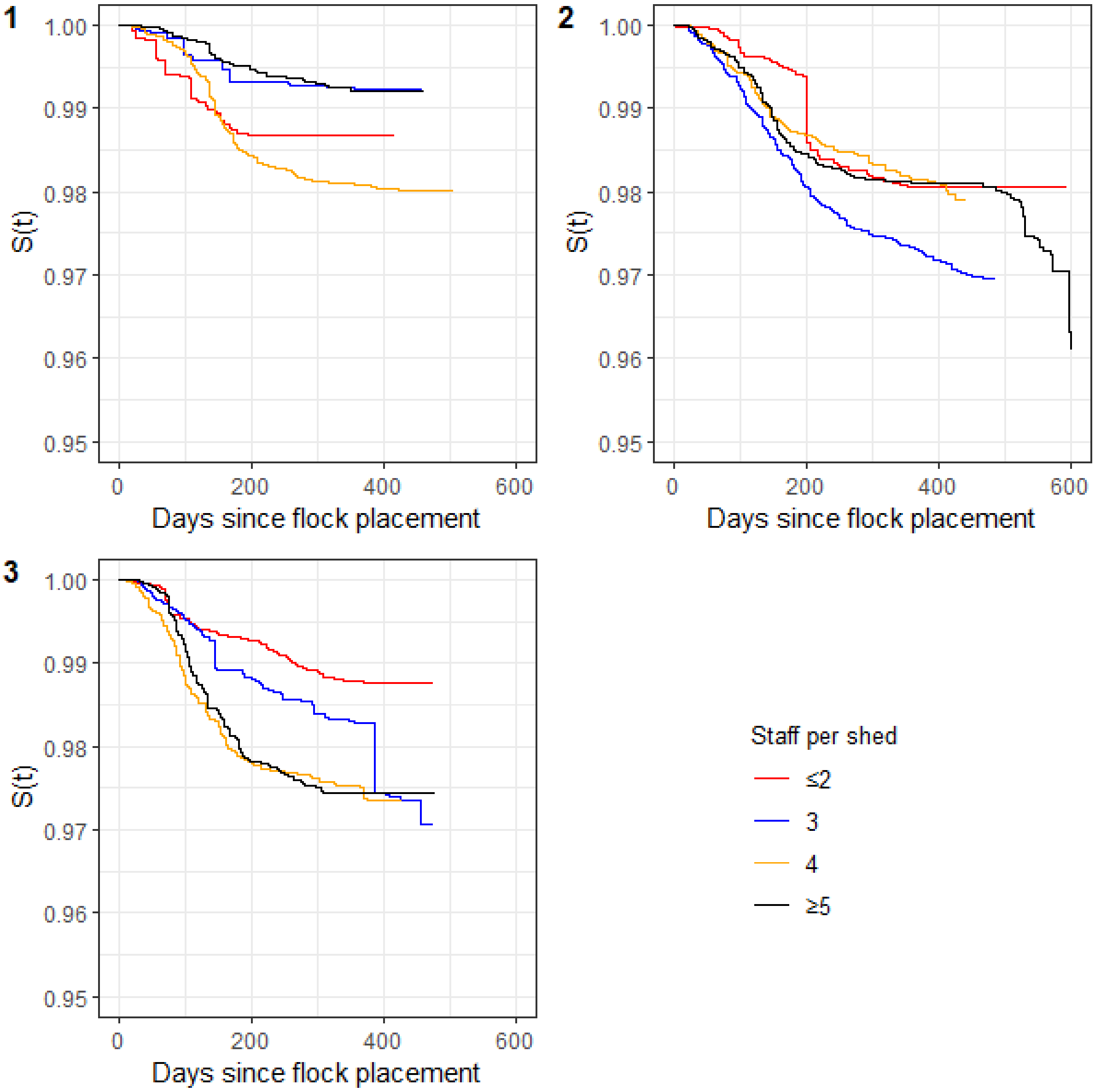

Figure 27 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by number of shed staff (≤2, 3, 4, and ≥5) and organisation. Log rank test statistic 1973; df 3; p <0.01.

Shed walks per day

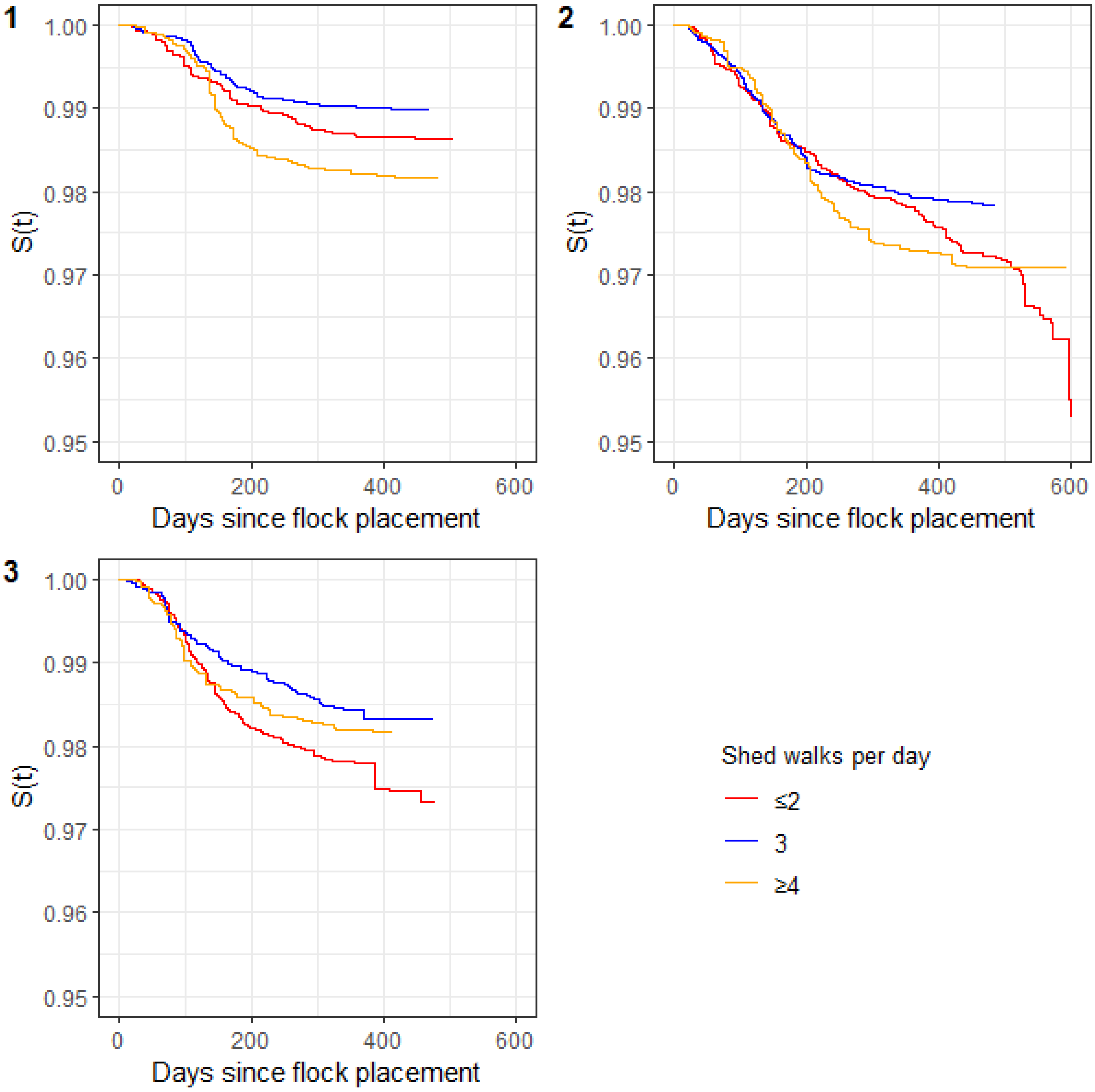

Figure 28 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by number of shed walks per day (≤2, 3 and ≥4) and organisation. Log rank test statistic 544.2; df 2; p <0.01.

Appendix 3

Kaplan-Meier survival analyses of smothering mortality stratified by shed features

Rearing shed type

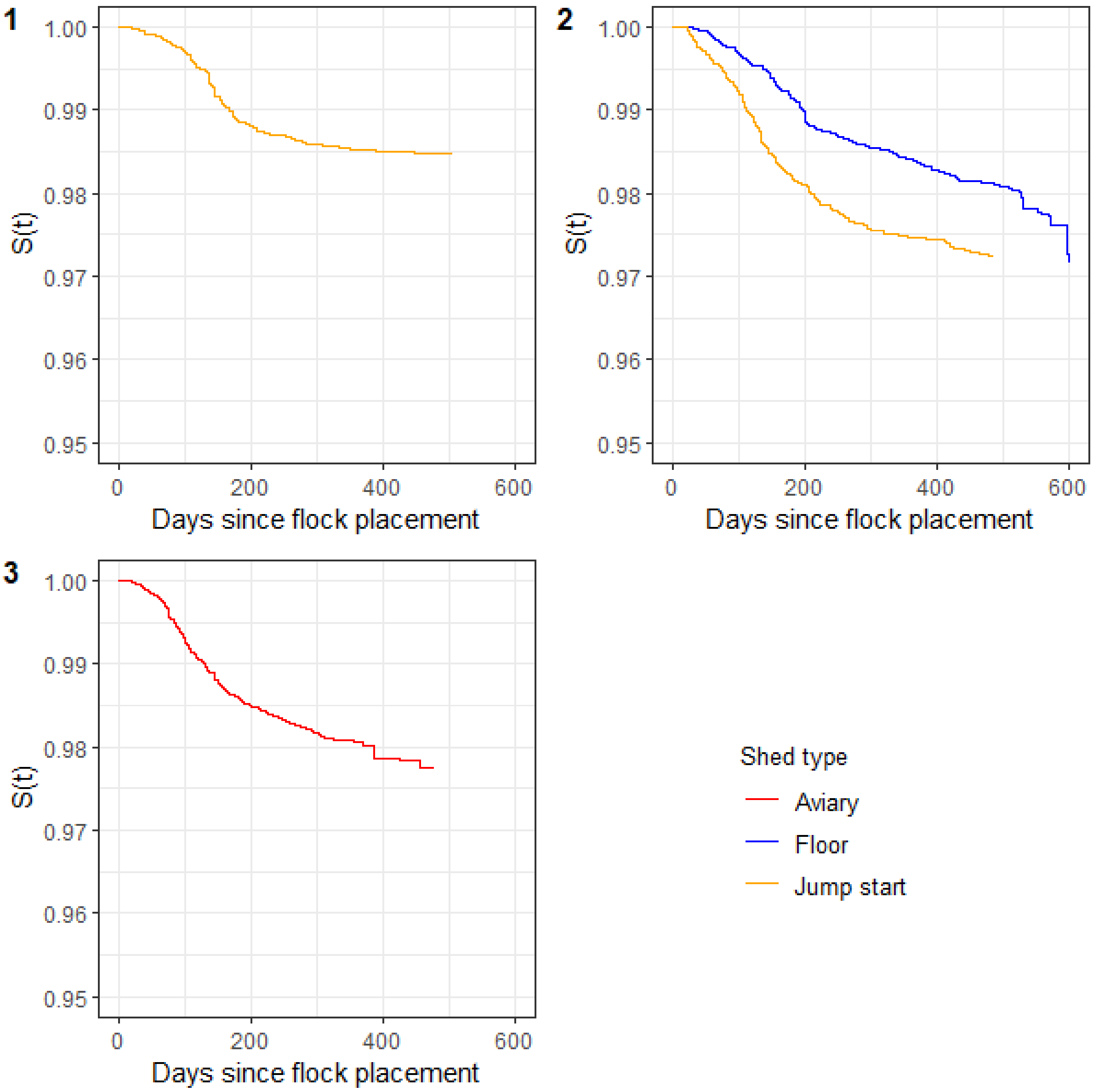

Figure 29 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by rearing shed type (aviary, floor, jump start) and organisation. Log rank test statistic 39.13; df 2; p <0.01.

Production shed type

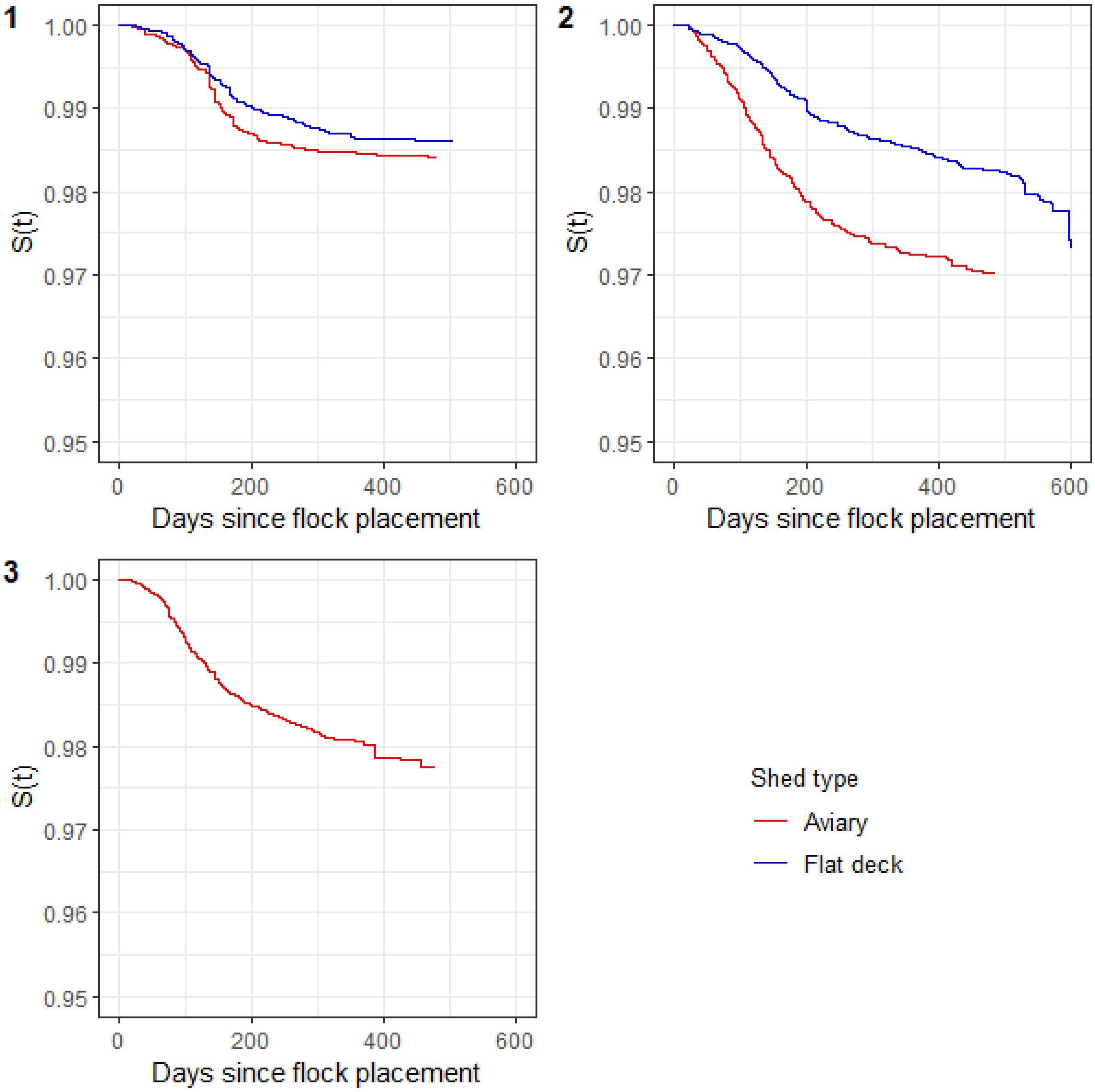

Figure 30 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by production shed type (aviary, flat deck) and organisation. Log rank test statistic 748.8; df 1; p <0.01.

Pop hole length per bird

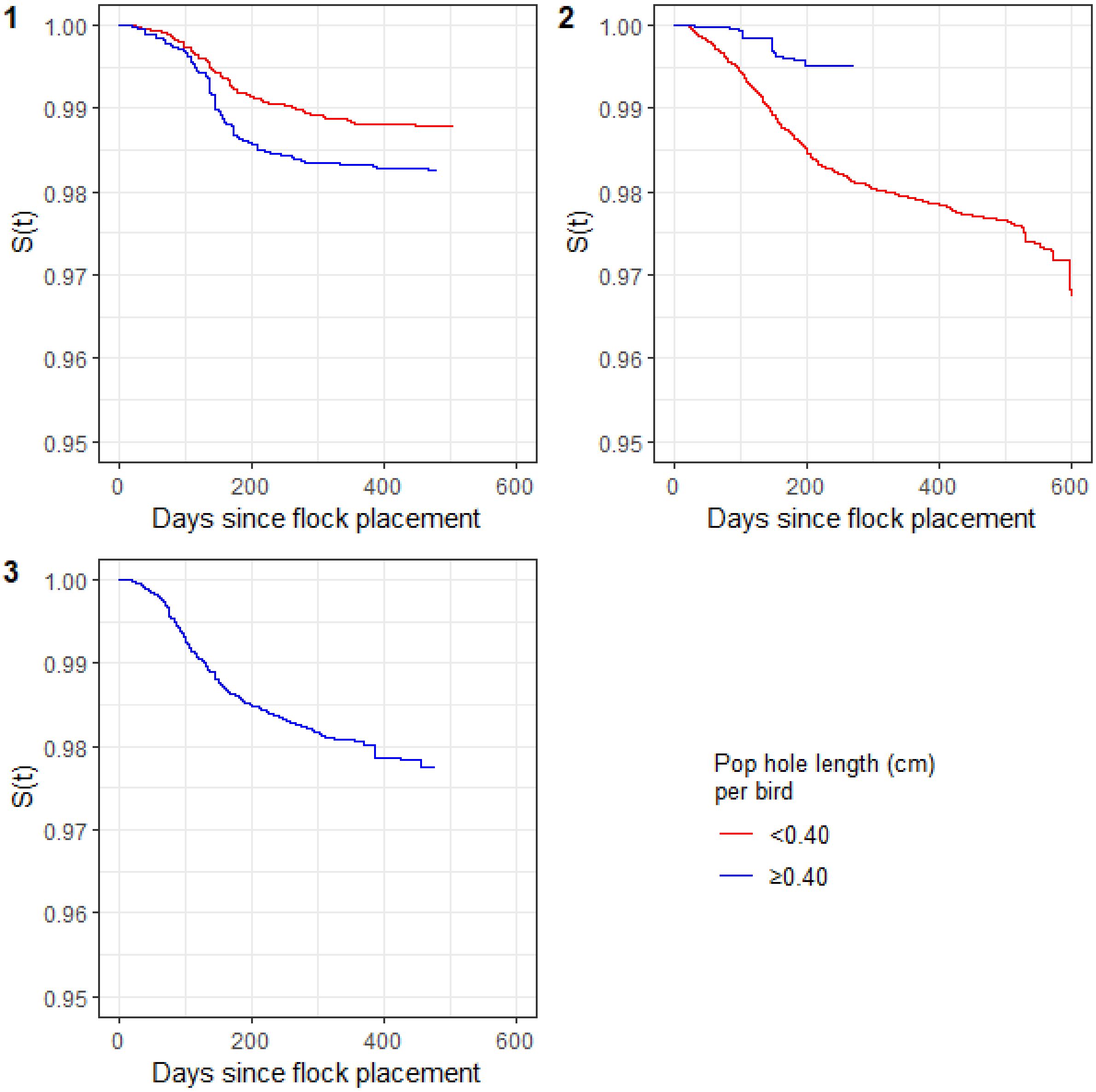

Figure 31 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by pop hole length per bird (<0.40 cm and ≥0.40 cm) and organisation. Log rank test statistic 3.11; df 1; p =0.08.

Light intensity (indoors)

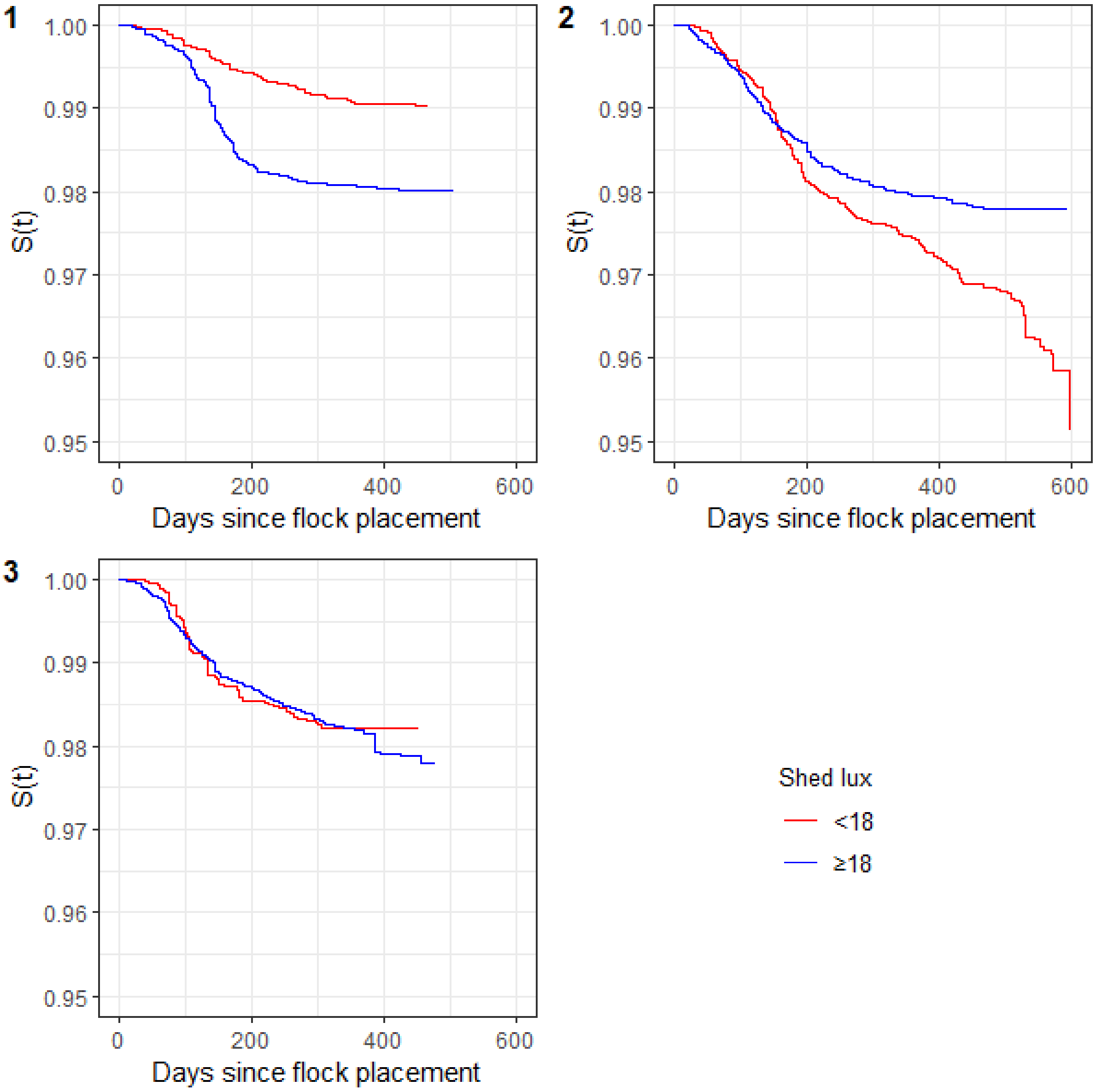

Figure 32 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by shed light intensity (<18 lux and ≥18 lux) and organisation Log rank test statistic 23.03; df 1; p <0.01.

Appendix 4

Kaplan-Meier survival analyses of smothering mortality stratified by environmental factors

Average temperature (outdoors)

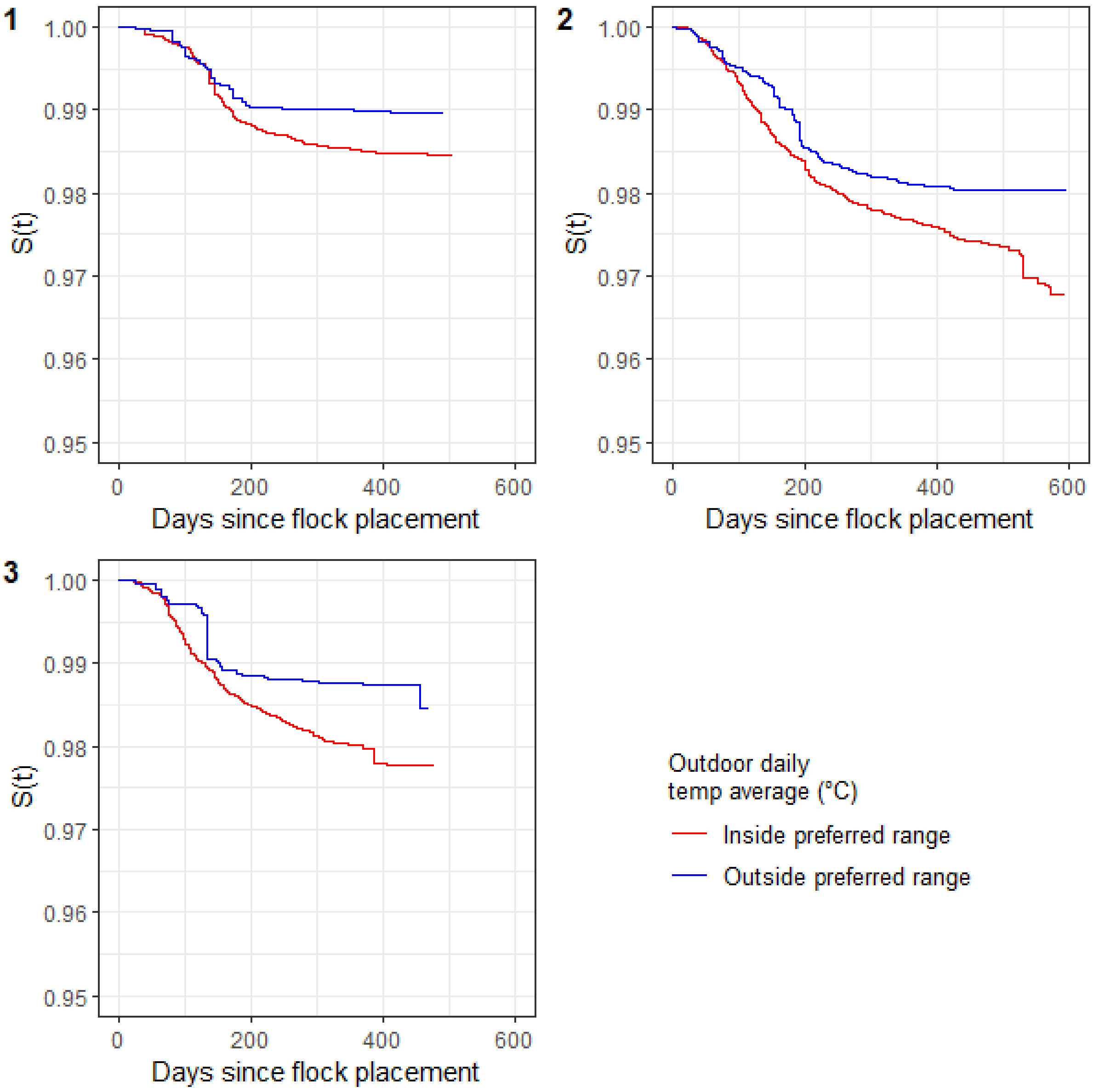

Figure 33 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by average outdoor daily temperature average inside or outside the preferred range and organisation. 'Preferred' temperature range for free range poultry is defined as 10 to 27 degrees Celsius. Log rank test statistic 616; df 1; p <0.01.

Minimum temperature (outdoors)

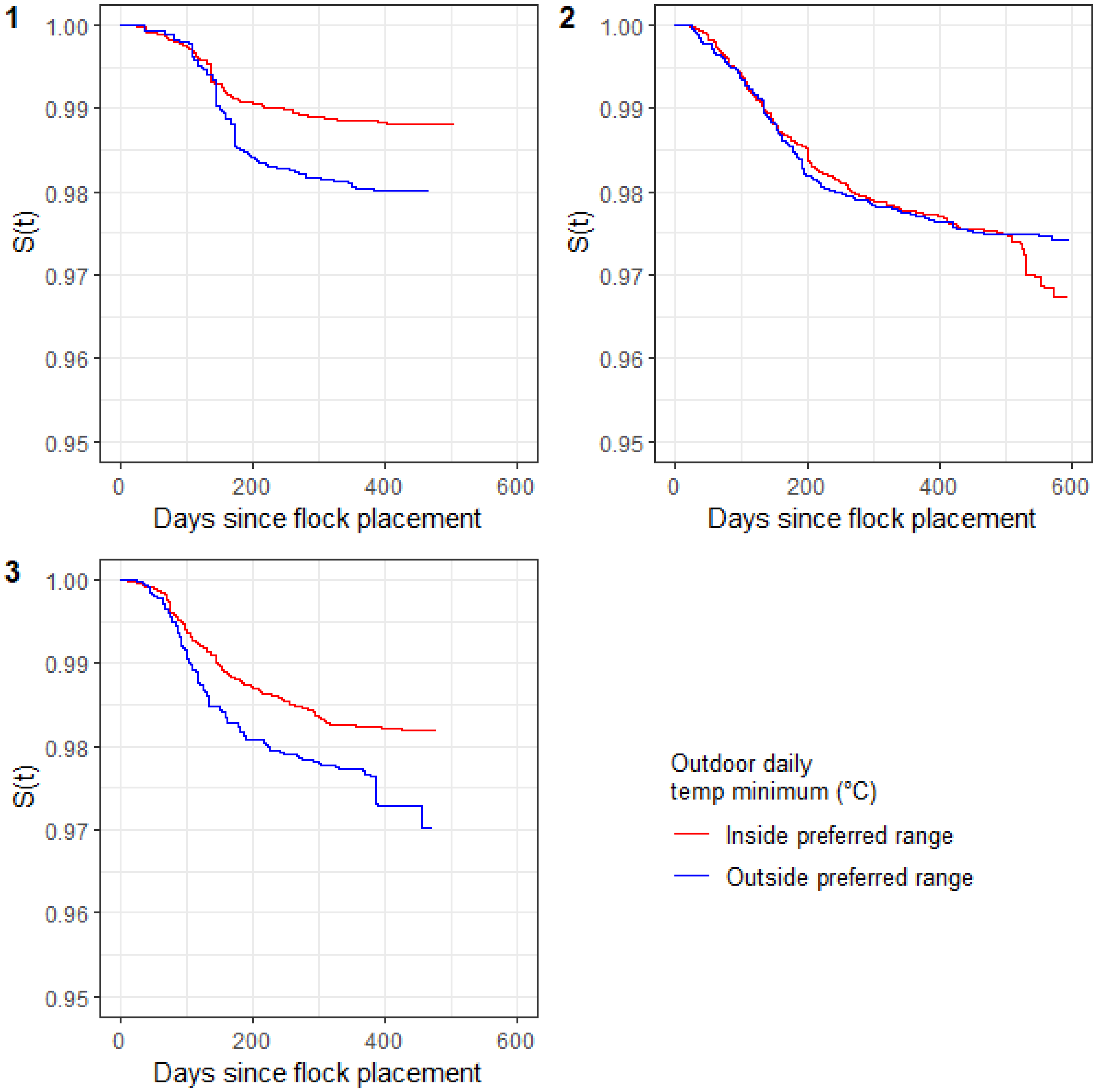

Figure 34 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by outdoor daily minimum temperature inside or outside the preferred range and organisation. 'Preferred' temperature range for free range poultry is defined as 10 to 27 degrees Celsius. Log rank test statistic 2.33; df 1; p = 0.10.

Maximum temperature (outdoors)

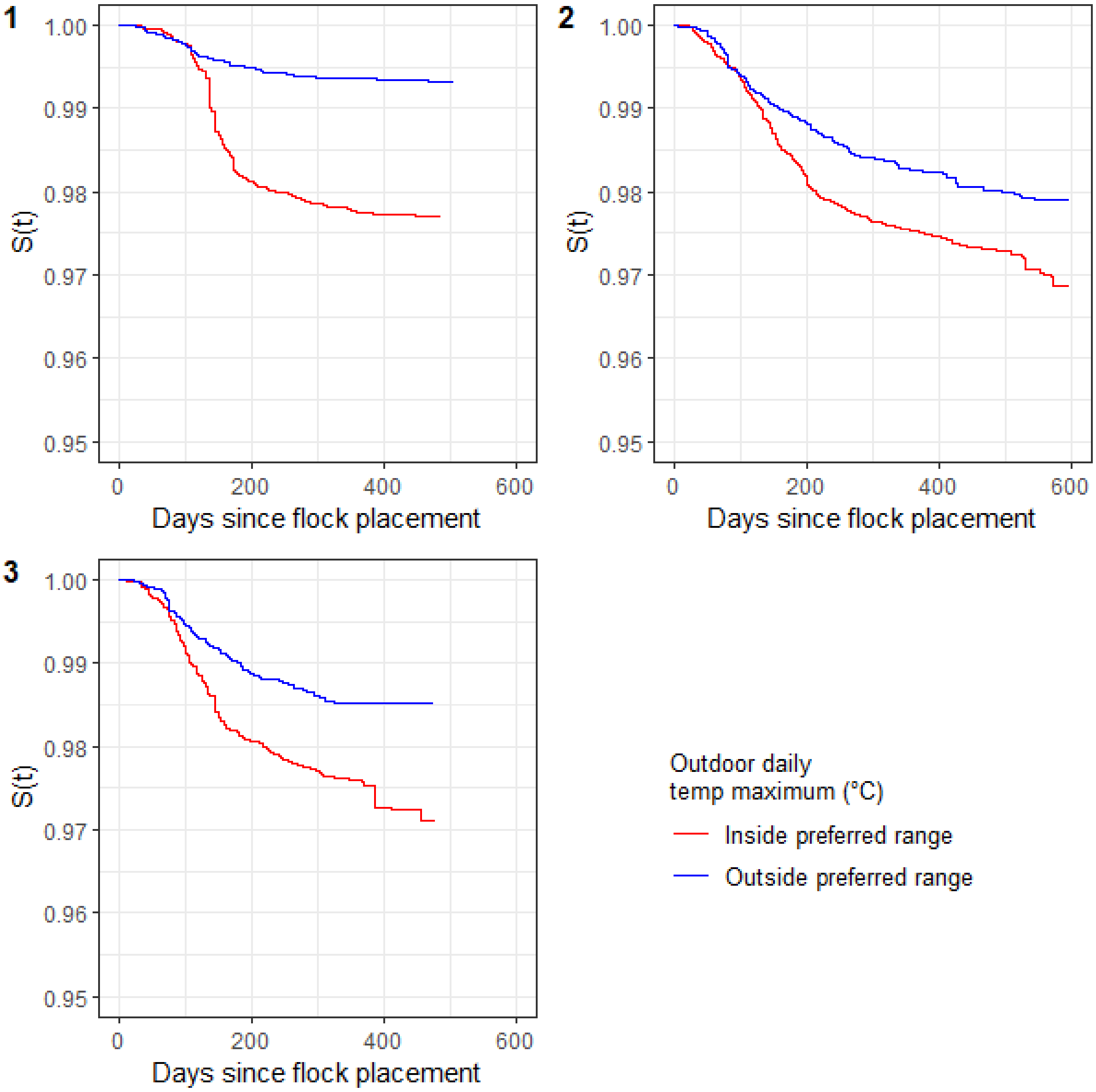

Figure 35 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by outdoor daily maximum temperature inside or outside the preferred range and organisation. 'Preferred' temperature range for free range poultry is defined as 10 to 27 degrees Celsius. Log rank test statistic 1081; df 1; p <0.01.

Temperature range (outdoors)

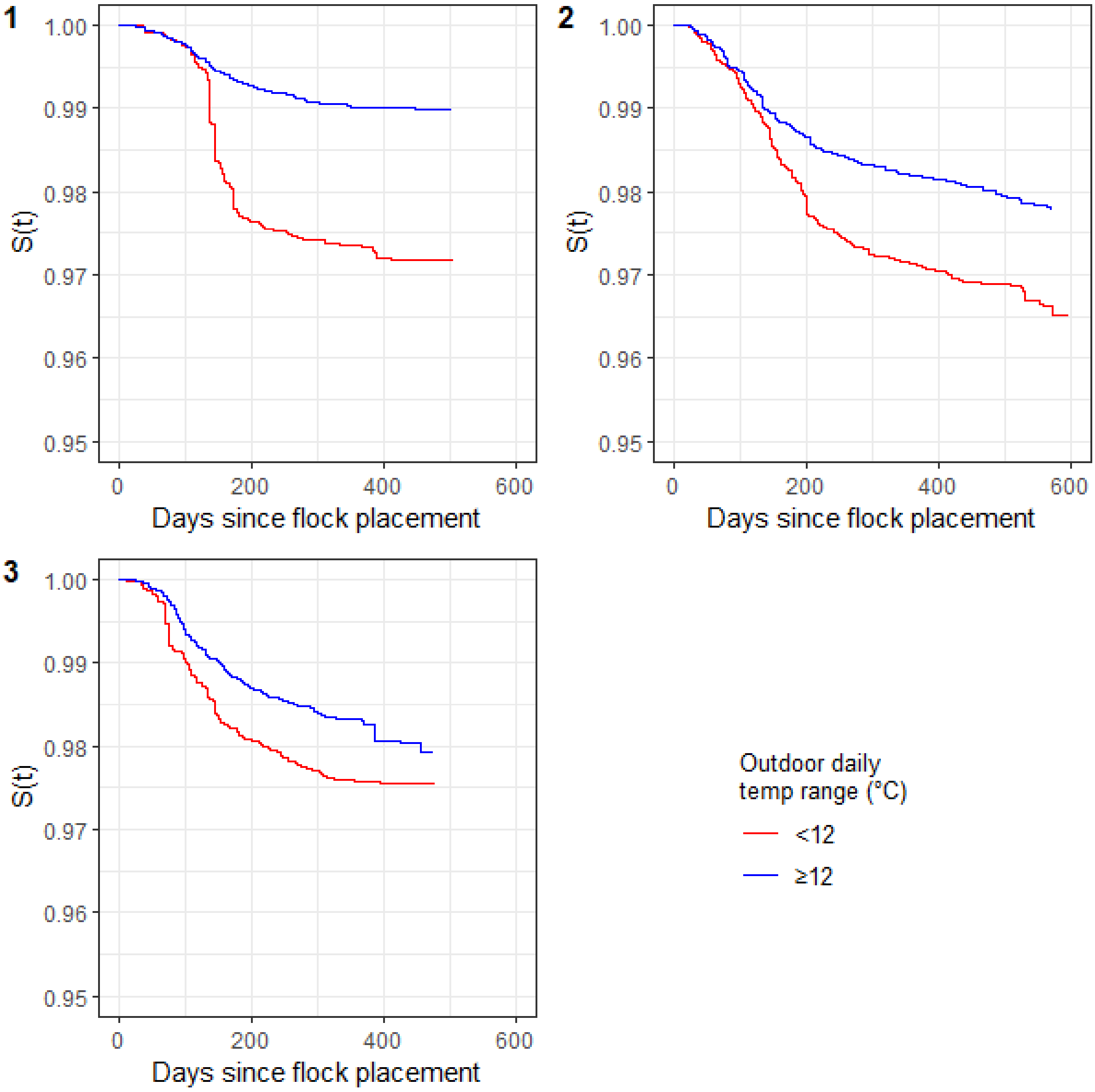

Figure 36 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by outdoor daily temperature range (<12 degrees and ≥12 degrees) and organisation. Log rank test statistic 1212; df 1; p <0.01.

Dew point (outdoors)

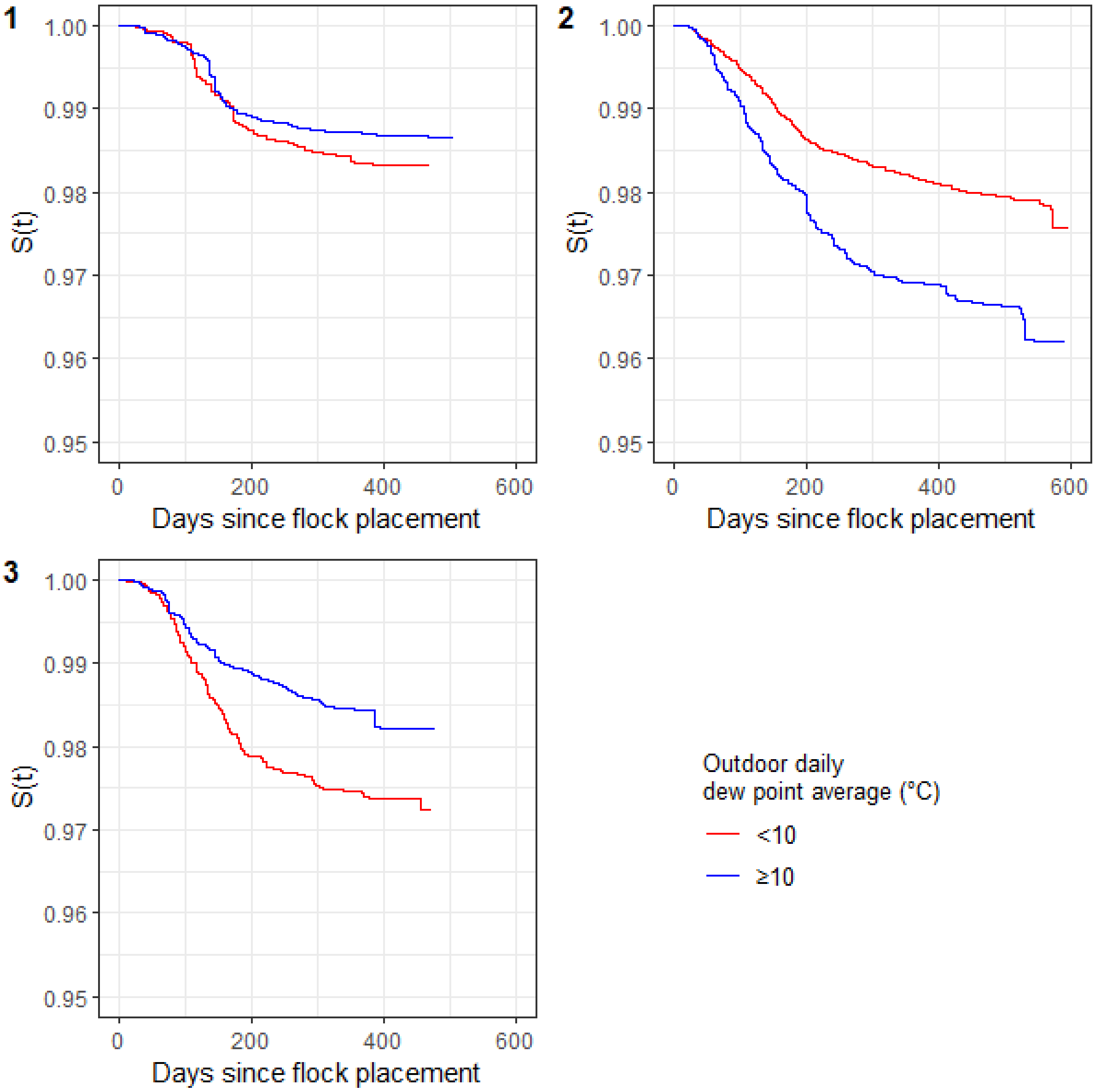

Figure 37 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by outdoor daily dew point average (<10 degrees and ≥10 degrees) and organisation Log rank test statistic 39.06; df 1; p <0.01.

Humidity (outdoors)

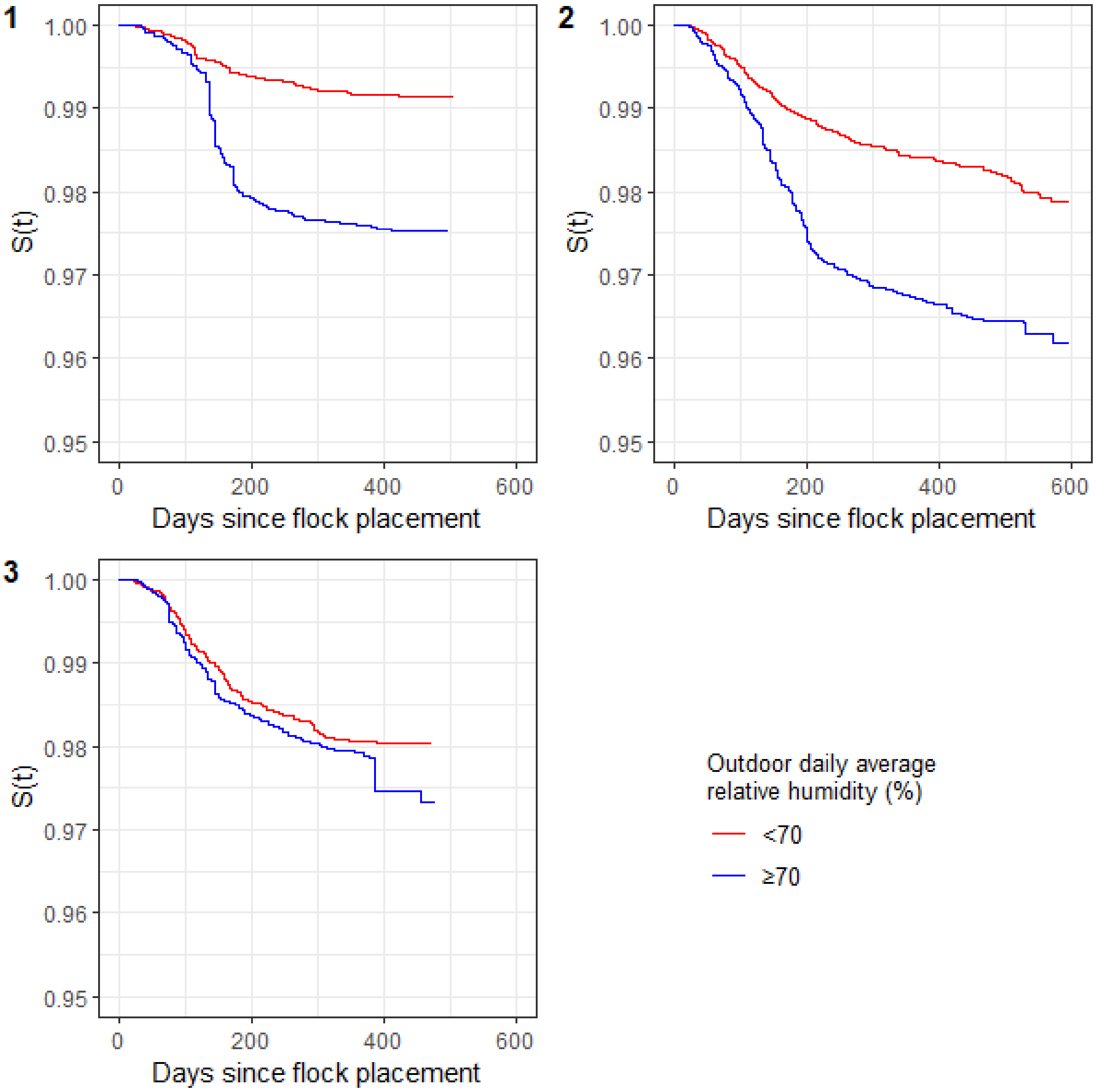

Figure 38 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by outdoor daily average relative humidity (< 70% and ≥70%) and organisation. Log rank test statistic 994.3; df 1; p <0.01.

Wind (outdoors)

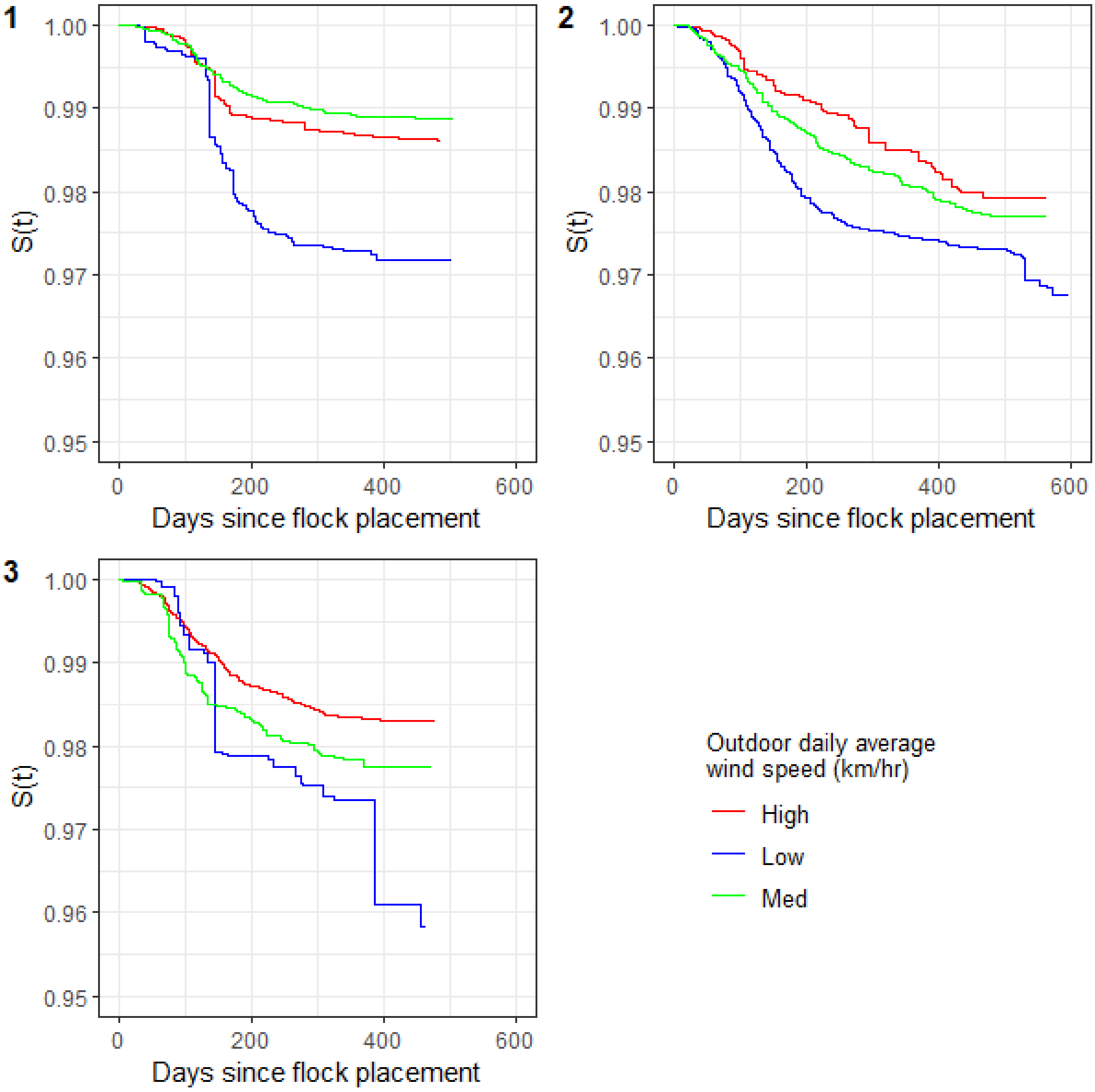

Figure 39 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by outdoor daily average wind speed tercile (low: 0 to 2.4 km/hr; med: 2.4 to 5.4 km/hr; high: > 5.4 hm/hr) and organisation. Log rank test statistic 1029; df 1; p <0.01.

Barometric pressure (outdoors)

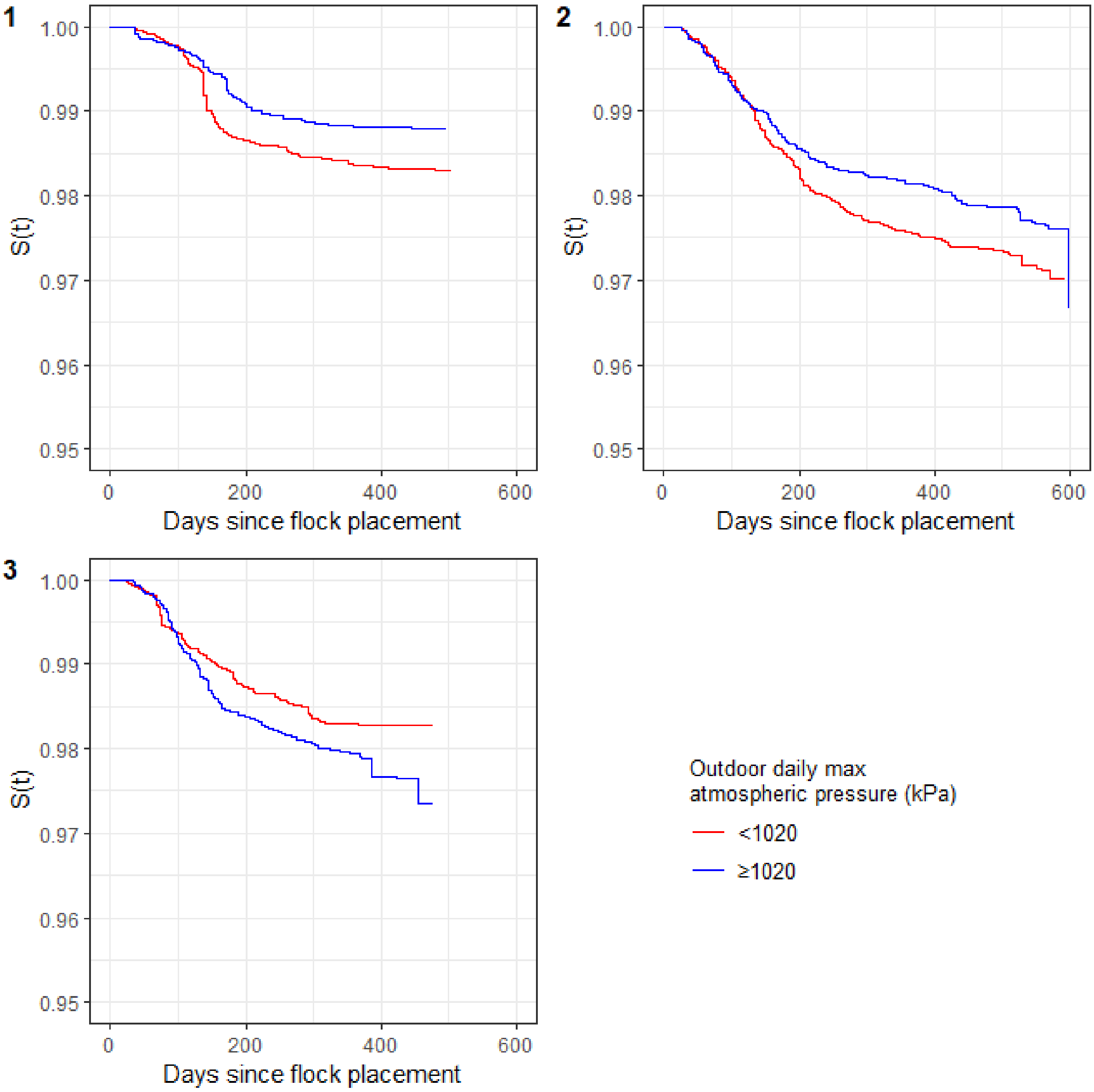

Figure 40 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by outdoor daily maximum atmospheric pressure (<1020 kPa and ≥1020 kPa) and organisation. Log rank test statistic 129.8; df 1; p <0.01.

Rainfall

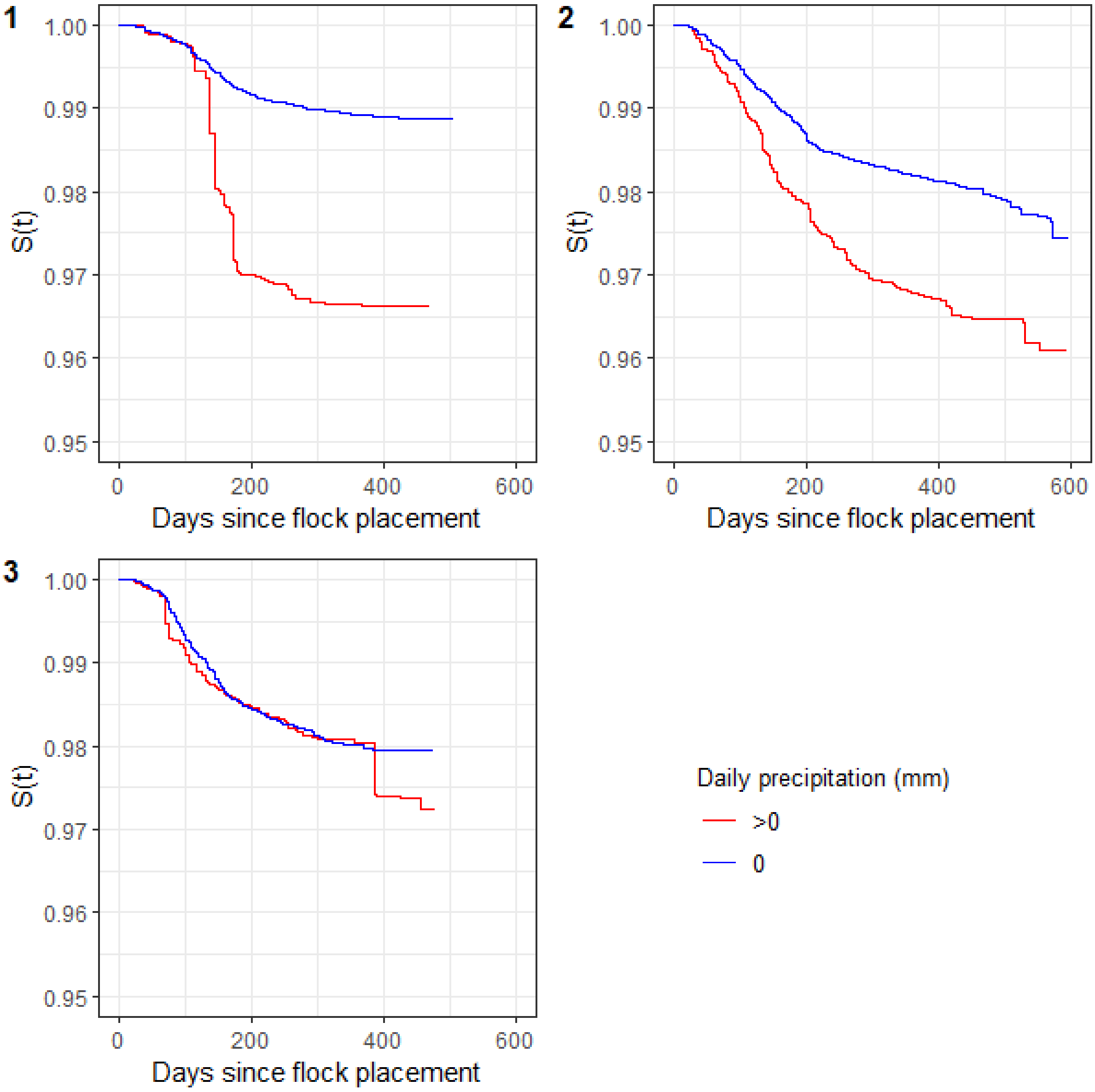

Figure 41 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by total daily rainfall (0 mm and >0 mm) and organisation. Log rank test statistic 2898; df 1; p <0.01.

Average temperature (indoor)

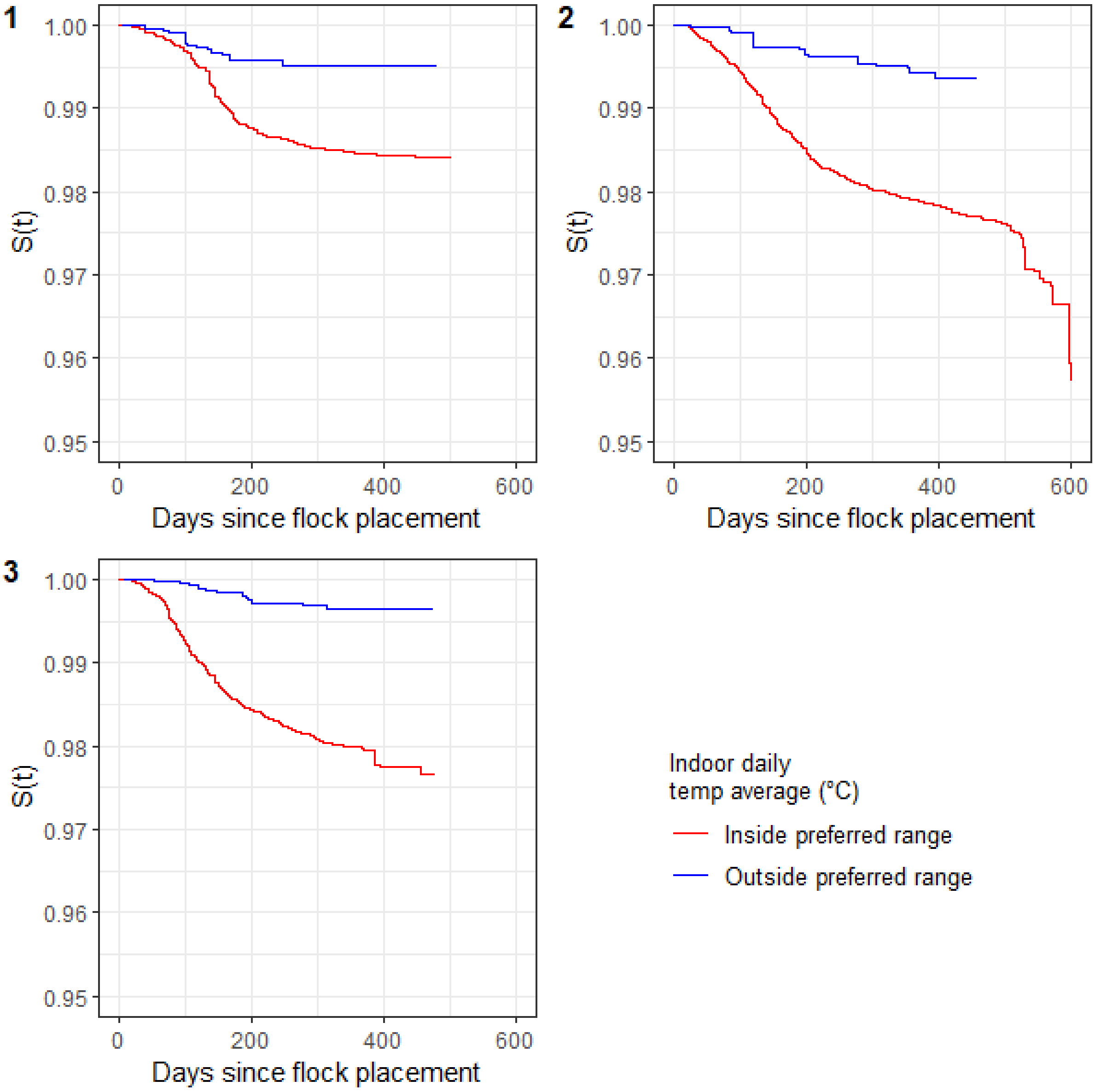

Figure 42 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by indoor daily average temperature inside or outside the preferred range and organisation. 'Preferred' temperature range for free range poultry is defined as 10 to 27 degrees Celsius. Log rank test statistic 189.7; df 1; p <0.01.

Minimum temperature (indoor)

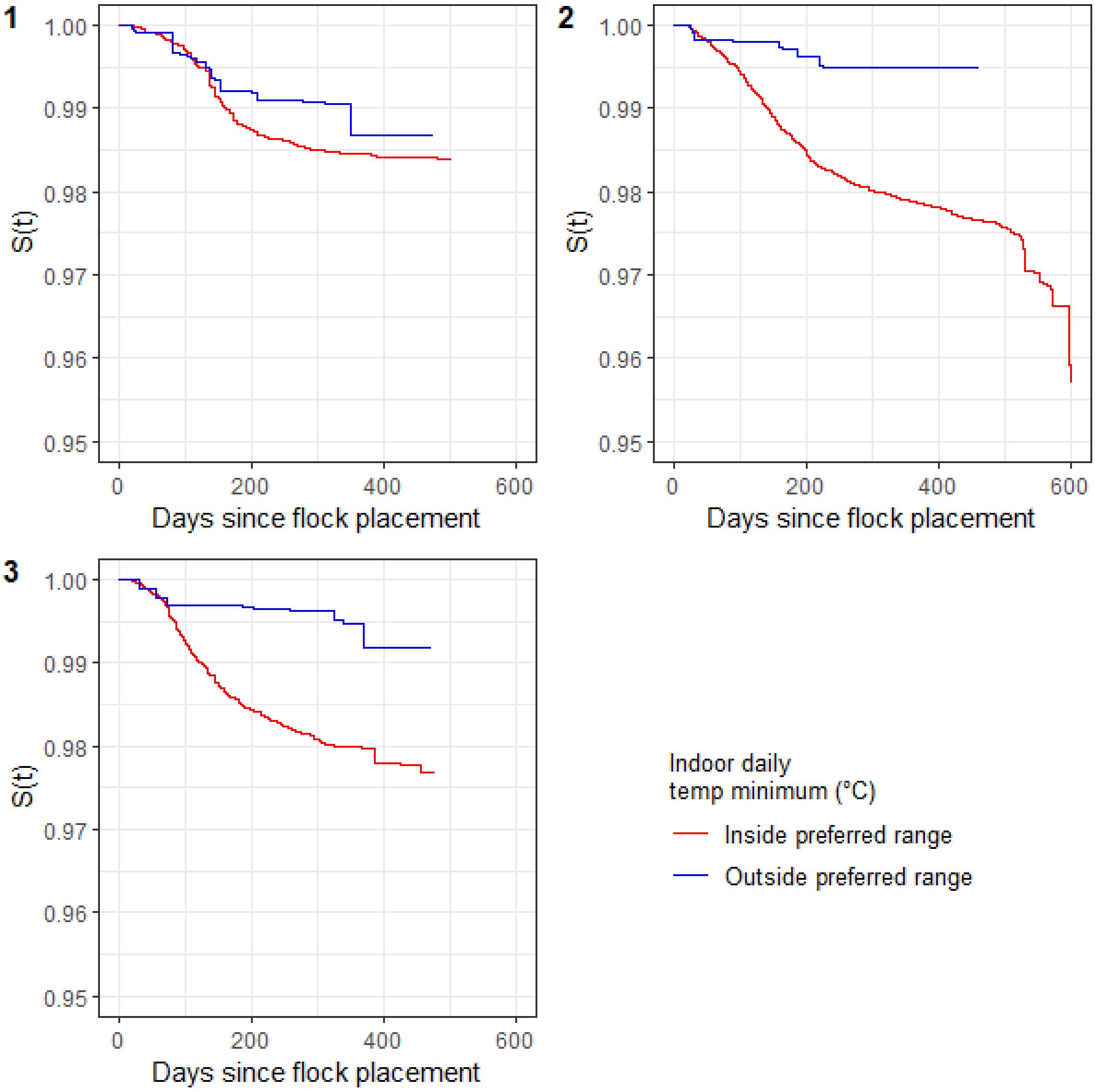

Figure 43 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by indoor minimum daily temperature inside or outside the preferred range and organisation. 'Preferred' temperature range for free range poultry is defined as 10 to 27 degrees Celsius. Log rank test statistic 706.2; df 1; p <0.01.

Maximum temperature (indoor)

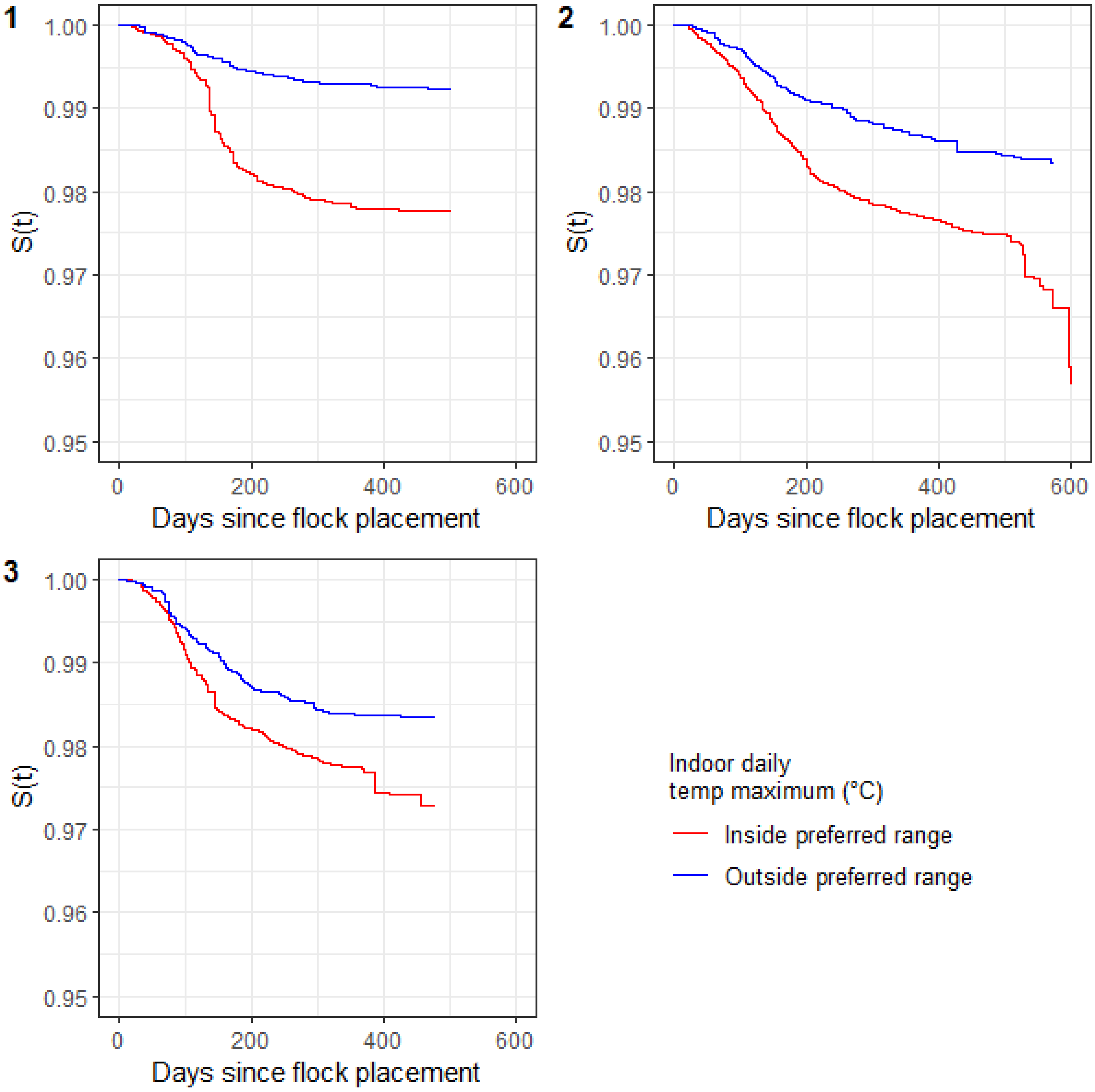

Figure 44 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by indoor daily maximum temperature inside or outside the preferred range and organisation. 'Preferred' temperature range for free range poultry is defined as 10 to 27 degrees Celsius. Log rank test statistic 1430; df 1; p <0.01.

Temperature range (indoor)

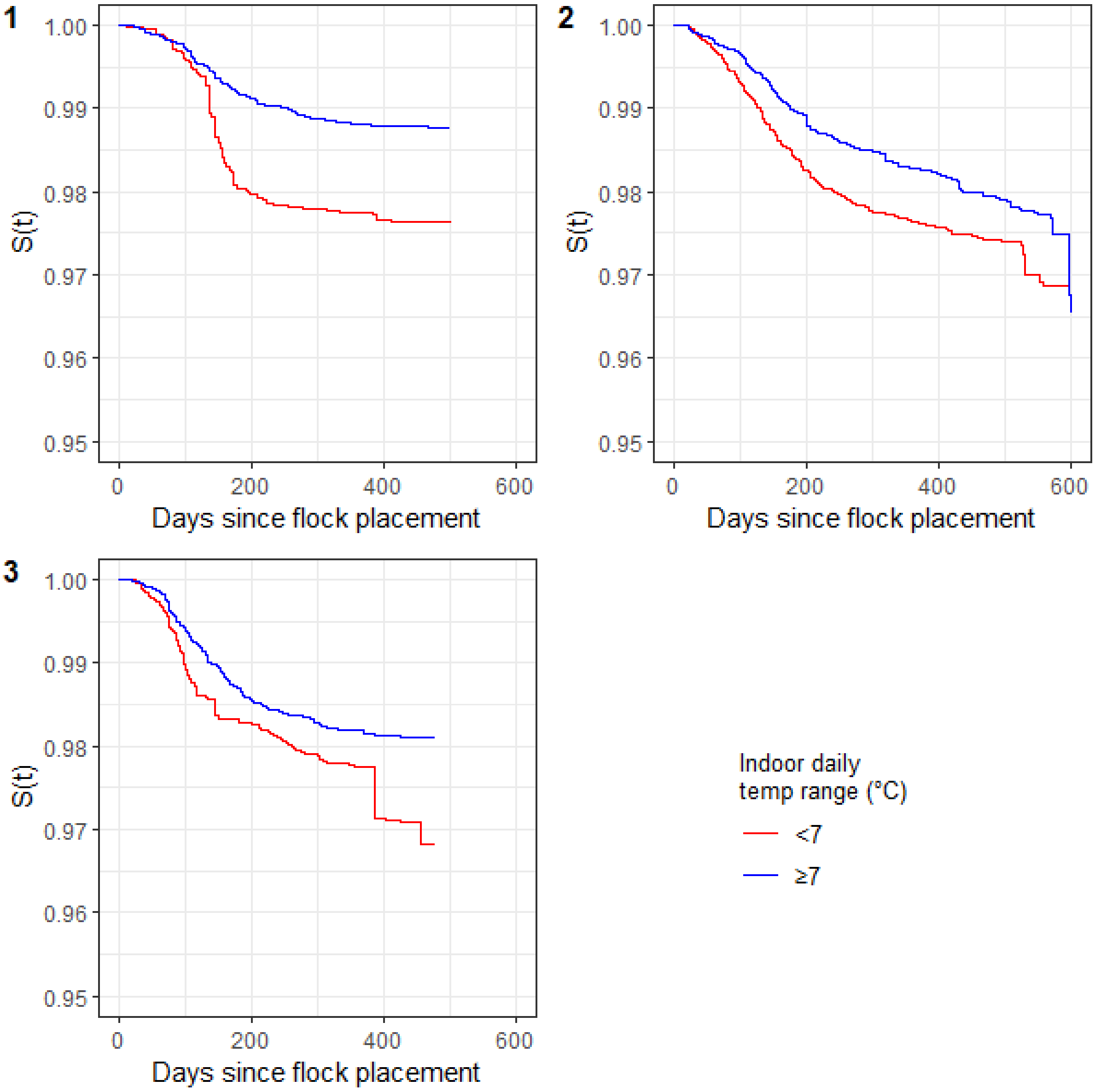

Figure 45 Kaplan-Meier survival curves showing the cumulative proportion of birds yet to experience death by smothering as a function of days since flock placement, stratified by indoor daily temperature range (<7 degrees and ≥7 degrees) and organisation. Log rank test statistic 1963; df 1; p <0.01.