

# Physiological Stress and Welfare of Broiler Chickens in Transit: Solutions Not Problems!

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**ABSTRACT** The rearing of large numbers of broiler chickens at geographically dispersed sites means that they have to be transported by road, over various distances, to centralized processing plants for slaughter. The birds may be exposed to a variety of stressors during transit, including the thermal demands of the transport microenvironment. The thermal environments experienced by broiler chickens during routine transport in the U.K. on a large number of commercial vehicles under a wide range of external climatic conditions have been characterized using three-dimensional thermal mapping (temperature and water vapor density). Inadequate ventilation results in heterogeneous distributions of temperature and humidity and, thus, thermal loads

within the vehicle, and, therefore, the existence of a “thermal core” in which the risk of heat stress is increased. Relationships between specific physiological indices of stress and quantified thermal loads have been determined in accurate transport simulations in the laboratory. The findings have been employed to establish a predictive model of the induction of heat stress during commercial transportation, as well as to define the acceptable ranges and limits for temperature and humidity within the transport containers. These principles have been utilized in developing a monitoring system to warn of impending heat stress and in improving vehicle design to facilitate the prevention of heat stress during broiler transportation.

(Key words: broiler, stress, thermal load, transportation, welfare)

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## INTRODUCTION

Animal welfare is currently high on the agricultural and political agendas in Europe and North America. The transportation of live animals in particular is subject to close scrutiny. Public concern has been prompted by media attention and open political debate. Attention has been focused on specific transport practices because of extensive reporting of public protests and high profile articles in popular scientific journals (e.g., Birchall, 1990; Bonner, 1995). Failure to reach consensus or agreement on European transport legislation has compounded the situation. In the U.K., the Ministry of Agriculture, Fisheries and Food favor the introduction of national legislation to improve the welfare of animals in transit. They are supported in this aim by the British Veterinary Association (BVA), who also suggest that “veterinarians should be promoted as protectors of animals’ welfare on behalf of the rest of society” (Veterinary Record, 1995). The BVA and various animal welfare organizations

recommend further research as “animal transport occurs now and will continue!”

Despite the recognized difficulties of objectively assessing the welfare of animals in transit and the necessity of separating welfare assessment from moral and ethical judgements (Broom, 1993), scientific analyses of animals’ responses to transport are generally presented as supporting the concept that transportation is inherently detrimental, possibly because one or more of the “five freedoms” is compromised (Savory, 1995). In contrast, it has been claimed that “there is nothing fundamentally wrong about being in a vehicle” (Webster, 1995) and it must be concluded that multiple, interactive stressors associated with other aspects of transportation are responsible for the reported adverse effects of transport on welfare. Confusion exists due to the complexity and uncertainty surrounding the assessment or quantification of “stress”, animal welfare and

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**Abbreviation Key:** AET = apparent equivalent temperature; BVA = British Veterinary Association; CK = creatine kinase; DOA = dead on arrival; GAS = General Adaptation Syndrome; H:L = heterophil:lymphocyte ratio; HPA = hypothalamo-adenohypophyseal axis; pCO<sub>2</sub> = partial pressure of carbon dioxide; RH = relative humidity; SAS = sympatho-adrenal system; TI = tonic immobility; VD = water vapor density; VP = water vapor pressure.

"suffering" (Dantzer and Mormede, 1983; Moberg, 1985; Barnett and Hemsworth, 1990; Mason and Mendl, 1993; Wiepkema and Koolhaas, 1993; Bekoff, 1994). Although stress physiologists and behaviorists may accept that certain physiological and behavioral responses are indicative of disturbances in the predictability and controllability of an animal's environment and thus the animal's well-being, it is also acknowledged that change *per se* in biological variables need not reflect reduced welfare (Moberg, 1985; Cashman *et al.*, 1989; Barnett and Hemsworth, 1990). The highly dynamic patterns of homeostatic response observed during stress make it difficult to deduce any simple relationship between stress and welfare (Wiepkema and Koolhaas, 1993). The concept that there is a specific "cut-off" point in a stress response or physiological variable at which welfare is deemed to be at risk has been criticized (Mendl, 1991), and a more integrated approach to measuring stress reactions has been advocated (Dantzer and Mormede, 1983). The use of a spectrum of different stress indices or the development of stress profiles may represent a more effective approach. This approach can be further strengthened by a complete understanding of the action and functional significance of the physiological and behavioral responses or welfare parameters to be quantified (Mason and Mendl, 1993).

Cognitive ethology has given rise to the proposal that, in addition to the use of empirical data in making decisions concerning animal welfare, subjectivity and common sense have an important role, and that subjective assessments should be viewed in the same critical light as ostensibly objective scientific fact (Bekoff, 1994). Anthropocentric claims about the way in which animals interact in their social and nonsocial worlds are often used to influence decisions on how animals can or should be used by humans (Bekoff, 1994). In the context of animal transportation, it has recently been suggested that "anthropomorphism, used carefully, can be heuristic" (Tudge, 1995). Acceptance of the cognitive capacities and sentience of animals has led to the argument that "if there is uncertainty, even if only slight, about an animal's ability to feel pain or suffer, then the individuals should be given the benefit of the doubt" (Bekoff, 1994). That author, however, acknowledges that it is still necessary to attempt to answer the questions: what are positive grounds for imputing suffering or suspecting that suffering is present but concealed?; what is a significant degree of suffering?; can we measure differences in the degree of suffering?; are these differences morally relevant?

In the face of the complexity of this topic, the apparent conflicts between philosophical, ethical, moral and objective scientific thought, and with the emphasis of research so far tending to favor the identification of problems not the provision of solutions, it is not surprising that an instinctive suspicion exists on the part of animal producers and haulers towards transport welfare research and its applications. Hopefully such fears may be allayed by research that is relevant to the welfare of the animals concerned and to the acceptable

objectives and constraints of commercial animal production. It is proposed that it is reasonable and valid to employ scientific method to identify potential sources of stress and reduced welfare during transportation and identify deficiencies in current practices. The remit of the researcher, however, should not be limited to merely confirming the presence of stress. A more constructive and profitable course is to exploit the knowledge thus gained to alleviate or prevent the stresses by basing improved designs and procedures on a sound scientific foundation. Thus, only through thorough characterization of animals' requirements in transit and the scientific definition of acceptable ranges and limits for environmental variables and exposure to stressors can genuine progress and improvements in transport methods and practices be achieved.

### **Transport Stress**

Approximately 3.5 billion broiler chickens are currently produced yearly in Europe. The major contributors to the output of broiler meat are France (21%), the U.K. (17%), Spain (17%), Italy (13%), the Netherlands (10%) and Germany (7%). Intensive rearing of the birds in large numbers, at geographically dispersed sites necessitates that they be transported by road, often over long distances, to centralized processing plants for slaughter. The birds may be exposed to a variety of potential stressors during transit, including the thermal demands of the transport microenvironment, acceleration, vibration, motion, impacts, fasting, withdrawal of water, social disruption and noise (Nicol and Scott, 1990; Mitchell and Kettlewell, 1993). The adverse effects of these factors and their combinations may range from mild discomfort and aversion to death. Indeed studies in the U.K. have indicated that up to 40% of "dead on arrivals" (DOA) are attributable to "transport stress" (Bayliss and Hinton, 1990) and that mortality increases with journey length (Warriss *et al.*, 1990). The latter investigation reported that broiler journeys in the U.K. were in general 3 h or less, but that occasionally birds might be confined to the vehicle for up to 12 h. It is accepted that transportation conditions and practices may severely compromise both bird welfare and productivity. Transport thus represents an issue of public and political concern and forms a constituent topic of recent European legislation (EC Directive 91/628, 1993). Preslaughter catching and handling of the birds may also have profound effects upon the degree of physiological stress and reduced welfare consequent upon subsequent transportation (Mitchell and Kettlewell, 1993; Scott, 1993; Kettlewell and Mitchell, 1994).

Several previous studies have attempted to characterize the behavioral and physiological responses of birds to transportation in either field or laboratory trials. The findings have been extensively reviewed (Hails, 1978; Freeman *et al.*, 1984; Swarbrick, 1986; Kettlewell, 1989; Nicol and Scott, 1990; Knowles and Broom, 1990; Mitchell and Kettlewell, 1993). Duncan (1989) has suggested that transport on a vehicle represents a severe stressor, based upon measurements of tonic immobility (TI), heart rate,

and plasma corticosterone concentrations. These findings are supported by studies involving measurement of TI following commercial transportation of broilers that indicated that transport *per se* may greatly increase birds "fearfulness" (Cashman *et al.*, 1989, Nicol and Scott, 1990). These responses may be, at least in part, the consequence of motion and noise as proposed by Nicol and Scott (1990), who demonstrated the aversive nature of these factors under laboratory conditions using passive avoidance techniques. Vibration has been studied in commercial transporters by recording the time history of accelerations in three mutually perpendicular axes on the vehicle bed and within the transport containers (Randall *et al.*, 1993). These data have been used to derive power spectral densities and characteristic frequency signatures. The results indicate the presence of vibrations of frequencies and magnitudes that would cause discomfort in humans (Randall, 1992) and their effects upon birds are presently under investigation.

The induction of physiological stress by transportation has been inferred from a number of studies. Plasma corticosterone is elevated following a road journey (Scholtyssek and Ehinger, 1976; Freeman *et al.*, 1984; Satterlee *et al.*, 1989). This apparent activation of the hypothalamo-adenohypophyseal-adrenocortical axis is consistent with the observation of post-transport increases in heterophil:lymphocyte ratios (Satterlee *et al.*, 1989; Mitchell *et al.*, 1992; Maxwell, 1993). Transportation also stimulates glucagon release, which increases lipolysis and raises plasma concentrations of nonesterified fatty acids (Freeman *et al.*, 1984). Transport stress-induced tissue dysfunction and damage are reflected by increased plasma activity of intracellular muscle enzymes, including creatine kinase (Scholtyssek and Ehinger, 1976; Mitchell *et al.*, 1992), although the precise mechanisms involved await elucidation. All such studies utilize the measurement of physiological, endocrine, biochemical, metabolic, or behavioral responses of birds during or after transport, and, from the results, deduce the presence of physiological or psychological stress in transit.

A major factor in the induction of physiological stress during transport is the complex thermal environment to which the birds may be exposed (Mitchell *et al.*, 1992, 1993; Kettlewell *et al.*, 1993; Kettlewell and Mitchell, 1993). Only a few attempts, however, have been made to characterize the thermal conditions experienced by broiler chickens on commercial vehicles (Scholtyssek and Ehinger, 1976; Gschwindt and Ehinger, 1978; Mitchell *et al.*, 1992; Kettlewell *et al.*, 1993; Kettlewell and Mitchell, 1993). Webster *et al.* (1993), using a model chicken to predict metabolic heat loss in transit, suggested that during normal transport in the U.K. well-feathered broiler birds would only experience thermal comfort over a very narrow range of ambient temperatures. It was proposed, however, that stress could be minimized by appropriate control of air movement within the vehicle, whether at rest or in motion. Currently no guidelines are available recommending optimum "on-board" conditions for the transport of broiler chickens or the acceptable ranges and

limits for the thermal loads to which birds might be subjected.

Although examination of the existing literature reveals that thermal stress is acknowledged as a major hazard during animal transportation, it has been poorly characterized under practical conditions. The interactions between the animals and the complex thermal microenvironments clearly require more rigorous analyses. Whereas a combination of air movement and low ambient temperatures may precipitate cold stress during poultry transportation (Webster *et al.*, 1993; Nicol and Saville-Weeks, 1993), this is rarely life-threatening, although welfare may be compromised. Alternatively, heat stress associated with high mortality, decreased productivity and meat quality, and reduced welfare status, is recognized by the industry as the most frequently occurring problem during commercial broiler transportation.

A modern poultry transporter using a modular container system may carry approximately 6,000 birds, at 2 kg BW, each with a metabolic heat production of 10 to 15W and the associated obligatory water loss of about 10.5 g/h. The addition of heat and moisture to the vehicle interior from the birds is thus approximately 90 kW and 63 kg/h respectively, although as temperature rises and the demand for thermoregulatory evaporative cooling through panting increases, the internal water vapor density will be further augmented. This thermal load must be dissipated by the prevailing natural ventilation patterns and the efficiency of this process will obviously be markedly influenced by the external environment. Simple theoretical calculations reveal the magnitude of the ventilation requirement on a typical broiler transporter.

If the external conditions are 20 C and 50% relative humidity (RH), then the water vapor density would be 8.6 g/m<sup>3</sup>. If the moisture addition from the birds increased the RH to 90% within the load (but without any change in temperature) it would represent a rise in vapor density of 7 to 15.6 g/m<sup>3</sup>. If the water production rate was 63 kg/h re-establishing the humidity *status quo* would require an air throughout of 2.5 m<sup>3</sup>/s. These theoretical calculations assume no temperature lift and an homogeneity of internal environment and ventilation patterns that may not pertain in commercial vehicles. The exercise demonstrates, however, that if the optimum on-board thermal environment is defined in terms of temperature and humidity with regard to the birds' physiological requirements, then from a knowledge of the temperature lifts and water vapor generation within the load it is possible to calculate minimum ventilation rates for specific conditions and to incorporate these into future design strategies.

The current studies have therefore focused upon the effects of the thermal environment upon broiler chickens in transit and have consisted of three major components: 1) Full characterization of the commercial transport thermal environment. 2) Identification of the most appropriate indices of physiological stress and their

quantitative correlation with defined and graded thermal loads in laboratory-based transport simulations. 3) Construction of predictive models of birds' physiological responses to one or more stressors during transportation.

A multidisciplinary approach has been adopted utilizing the complementary skills from a number of disciplines.

### **Characterization of the Thermal Environment During Transport**

Whereas many models of heat exchange describe the effects of temperature and air movement, the influence of humidity is frequently neglected despite the dependence of evaporative heat loss upon the gradient of water vapor density. This neglect is of particular importance when the reliance on the evaporative avenue of heat loss is increased at elevated environmental temperatures (Richards, 1976; Dawson, 1982). It has been estimated that raising relative humidity from 20 to 80% at 28 C in a transport container would impose a heat load on the birds equivalent to an additional rate of rise of body temperature of 0.42 C/h (Mitchell *et al.*, 1990).

Commercial broiler transporters have therefore been fully instrumented and three dimensional mapping, by multisite recording, of temperature and water vapor density has been undertaken on a large number of journeys of different durations, in each season of the year. The results indicate the existence of a "thermal core" within the "bioload" due to inadequate ventilation and nonhomogeneous air flow distribution (Mitchell *et al.*, 1992; Kettlewell and Mitchell, 1993; Kettlewell *et al.*, 1993). Under certain external climatic conditions and vehicle configurations this inadequacy will precipitate heat stress. On "mild" winter days, temperatures of >30 C and relative humidities of >80% have been recorded in the core resulting in "paradoxical heat stress" in the cold season. These findings are described in more detail below.

In order to characterize the birds' responses to quantified thermal conditions the concept of "apparent equivalent temperature" (AET) has been exploited (Mitchell and Kettlewell, 1993). This parameter is derived from the temperature, water vapor pressure, and the psychrometric constant. It is uniquely related to the wet bulb temperature and describes the total heat exchange between a wetted surface and the environment (Monteith, 1973). AET is the dry bulb temperature that a sample of air of known temperature and water vapor pressure would reach, under ideal conditions, if all the water vapor was condensed without heat being lost from the system. The equations governing this reaction are given in Campbell (1977).

When broiler chickens are placed in a transport container at commercial stocking density and a heat stress is imposed, the constraints upon behavioral thermoregulation and the functional reduction in surface area for sensible heat exchange may increase the reliance upon evaporative heat loss at relatively low dry bulb temperatures. The emphasis on this avenue of thermal exchange may then rise markedly with further increases in

temperature. The influence of water vapor density on heat loss may thus be more pronounced on a crate of birds than on an individual bird under similar conditions. Dependence upon evaporative exchange from the respiratory tract and skin (Richards, 1976; Dawson, 1982) in an increasingly humid environment with the imposed geometric arrangements of the birds in a transport container may represent a circumstance in which total heat loss is best described as if each bird was a single wetted surface. In this case, the heat transfer is proportional to the gradient of AET between the birds' exchange surfaces and the air (Monteith, 1973). The former will tend to remain relatively constant and, as a consequence, heat loss will depend upon the environmental AET, in still air, as in the thermal core of a broiler transporter.

It is therefore proposed that AET may be regarded as an integrated index of the thermal loads imposed upon the birds under the conditions prevailing in commercial transport containers. It should be also emphasized that the "effective temperature" within a transport container may be greater than might be predicted from simple measures of temperature or relative humidity alone and that the thermal loads represented by the appropriate values of AET must be calibrated in relation to the degree of physiological stress induced.

### **Indices of Stress**

In order to determine the extent of stress imposed upon an animal in transit, it is first essential to understand the basis and mechanisms of the physiological stress response and to identify those variables that best reflect the disturbances in homeostatic control induced by the transport environment and practices. This technique constitutes the development of a physiological stress profile for the appropriate age, sex, and species of animal. Stress responses are considered to be essentially adaptive or protective and thus should prevent or minimize detrimental effects of the stressor that was imposed upon the animal. This objective may frequently not be achieved. The two major physiological systems mediating the stress response are the sympatho-adrenal system (SAS) first described as producing the "Emergency syndrome" or "Fight or flight reaction" (Cannon, 1932) and the hypothalamo-adenohypophyseal-adrenocortical axis (HPA) involved in the General Adaptation Syndrome (GAS) recognized by Selye (1936). Other stress responses may be mediated through the more recently identified endocrine functions of the immune system (Blalock and Smith, 1985; Campos *et al.*, 1991). The success of adaptive responses and the resultant effects upon the animal may be categorized in terms of the adequacy of the compensation in the face of a challenge tending to disturb a controlled physiological variable: 1) adequate compensation = successful homeostasis; 2) inadequate compensation = perturbations in controlled variables; 3) decompensation = pathological failure of compensatory mechanisms or direct deleterious effects of compensation.

Physiological changes may be measured in all three types of response. Alterations in the slope of a plot of the appropriate stress variable against the magnitude of the stressor or a "catastrophic" step-change in one or more stress indices may indicate transition from one category to another. Such observations may then be used to determine the severity of the stress imposed.

Employing a range of AET representative of the temperature-humidity combinations encountered in commercial practice, laboratory simulation studies have been used to assess the degree of physiological stress imposed and determine the combinations of temperatures and humidities producing equivalent biological effects. Birds responses have been quantified in accurate simulations, using commercial broiler crates in controlled climate chambers. The physiological measurements employed included deep body temperature, changes in blood chemistry (pH and pCO<sub>2</sub>), differential leucocyte profiles [e.g., heterophil:lymphocyte ratios (H:L) and basophil and eosinophil numbers] and plasma activities of intracellular enzymes (e.g., creatine kinase) whose release is indicative of stress associated tissue dysfunction (Mitchell *et al.*, 1992; Mitchell and Sandercock, 1995). In parallel complementary studies, responses in plasma corticosterone and plasma metabolites (e.g., nonesterified fatty acids, triglyceride, glucose, and lactate) have been employed to further characterize physiological stress and disturbances in substrate supply and utilization and the possible induction of fatigue.

## MATERIALS AND METHODS

### Commercial Transport Environments

Studies were undertaken on commercial broiler transport vehicles consisting of truck and trailer components both fitted with detachable curtain sides. Birds were carried in a modular drawer system at a stocking density of 53 kg/m<sup>2</sup>. Each drawer or crate (1.3 × 0.7 × 0.25 m) was generally loaded with 21 to 22 birds in summer and up to 23 birds in winter. The drawer sides were perforated by vertical slits 10 mm wide at 55 mm center spacing. The capacity of each vehicle was approximately 6,000 birds. The vehicles had a solid headboard and roof but the rear of both truck and trailer components were open. Temperatures and RH were continuously monitored by combined probes<sup>2</sup> at six locations in the load known to be representative of the heterogeneous distribution of the thermal microenvironments within the vehicle as previously described (Mitchell *et al.*, 1992; Kettlewell *et al.*, 1993; Kettlewell and Mitchell, 1993). These recordings were made within the transport containers at the level of the birds and thus reflected the precise thermal conditions

experienced by the broilers in transit. Water vapor pressures and densities (VP and VD, respectively) were automatically calculated for each RH value. Simultaneous measurements of ambient thermal conditions and air speed over the vehicle were made using combined probes and an anemometer mounted on the roof of the truck. Data collection was performed by Grant Squirrel 1201 data loggers<sup>3</sup> with direct down loading to a lap-top PC (CI/LEO LT-920C).<sup>4</sup> These methods allowed three-dimensional thermal mapping of the transport environment in summer and winter seasons when the curtains were open and closed respectively, as well as estimation of thermal gradients between the vehicle and the external environment. Accurate amounts of weather conditions and journey details and events were recorded by an observer travelling on the vehicle.

### Physiological Responses to Thermal Loads

Broiler chickens (42 d of age, approximately 2 kg BW) were placed in transport crates at commercial stocking density (22 birds per crate = approximately 53 kg/m<sup>2</sup>). These crates were fitted with a wire mesh top and located in a climate chamber in which temperature and humidity were controlled to predetermined set points (± 0.2 C and ± 5%). The corresponding "in crate" environmental parameters were measured at 30-s intervals, as described for commercial transport, throughout a standard 3-h exposure. This journey duration was considered representative of the majority of transport times within the U.K. (Warriss *et al.*, 1990, 1992).

The AET or heat loads employed were achieved using "in-crate" temperatures of 22 to 30 C adjusting the VD between 10.5 and 27.0 g/m<sup>3</sup> as required.

Ten birds were studied in each treatment. Body temperatures were measured at the beginning and end of the experimental period, by means of a thermistor probe inserted 5 cm into the rectum. Blood samples (3.0 mL) were obtained from the brachial vein by simple venipuncture from all birds immediately prior to loading into the crate and upon removal. The blood samples, containing anticoagulant (50 μ mL), heparin for general blood chemistry or EDTA for hematology) were placed on ice. Blood pCO<sub>2</sub> and pH were determined using an automated blood gas analyzer<sup>5</sup> within 2 min of withdrawal. Venous blood is acceptable for blood gas or acid base status determination when only pCO<sub>2</sub> and pH are to be considered (Hocking *et al.*, 1994). Blood smears were made for subsequent differential leucocyte analysis following May-Grünwald staining (Robertson and Maxwell, 1990) and H:L ratios were calculated (Mitchell *et al.*, 1992). Plasmas were prepared by centrifugation of blood at 1,500 × g for 5 min and stored at -20 C pending determination of creatine kinase activity (CK) by automated spectrophotometry (Mitchell *et al.*, 1992).

## RESULTS

The environmental data (see Table 1) indicated that when the curtains were open during the summer, only a

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<sup>4</sup>CI/LEO LT-920C-CI, Confidential Ltd., Hampshire International Business Park, Basingstoke, Hants, RG62 0GQ, U.K.

<sup>5</sup>Model 238, Ciba Corning, Halstead, Essex, CO9 2DX, U.K.

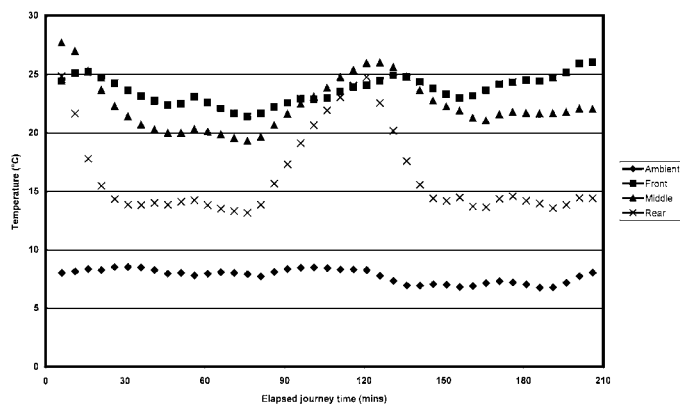
**TABLE 1. Typical mean temperatures and water vapor densities in three locations within the load, during matched summer (curtains open) and winter (curtains closed) commercial journeys of 3.6 h duration. The values are presented as the means  $\pm$  1 SD (n = 218)**

Variable	Curtain configuration	Ambient	Front	Middle	Rear
Temperature, C	Open	21.2 $\pm$ 0.8	26.8 $\pm$ 0.7	25.6 $\pm$ 1.2	24.3 $\pm$ 0.8
	Closed	9.8 $\pm$ 0.7	25.5 $\pm$ 3.7	24.9 $\pm$ 3.3	22.5 $\pm$ 2.9
Water vapor density, g/m <sup>3</sup>	Open	10.4 $\pm$ 1.0	11.2 $\pm$ 1.1	11.0 $\pm$ 1.2	10.4 $\pm$ 1.1
	Closed	8.1 $\pm$ 0.3	16.2 $\pm$ 4.3	14.3 $\pm$ 3.6	12.9 $\pm$ 2.6

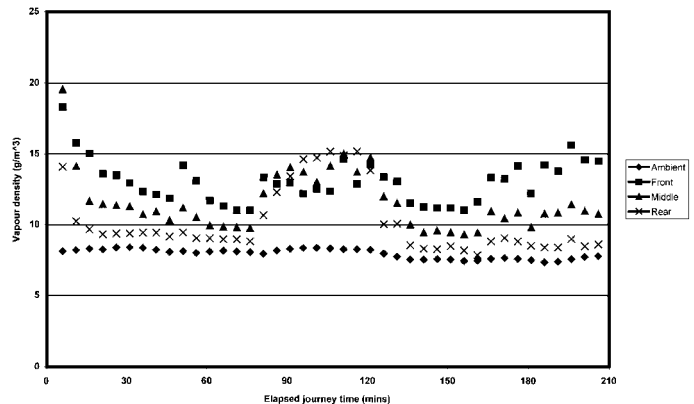
small gradient of temperature (2 to 5 C) existed between the transport containers and ambient conditions. Heat loads were slightly higher in the trailer than in the truck section. No discernible difference in VD between the vehicle interior and the surroundings was observed. The “on-board” environment was thus relatively homogeneous in this curtain configuration when the vehicle was moving due to the prevailing ventilation regimen. In contrast, during the winter journeys with the curtain sides closed, substantial increases in both temperature and VD above ambient values were recorded. Elevations of mean temperature and VD of 14.5 C and 6.3 g/m<sup>3</sup>, respectively, were observed. Gradients of both parameters existed from front to rear of truck and trailer. A “thermal core” was thus identified towards the upper front of the two compartments where ventilation was minimal and the consequent risk of heat stress was proportionately greater. In both open and closed configurations, temperature and RH increased when the vehicle was stationary although the relative effect was greater during winter transport. The temperature and VD profiles during a typical journey with curtains closed are presented in Figures 1 and 2. The data describe the spatio-temporal distribution of thermal loads within the vehicle. It is clear that both temperature and VD are highest and relatively constant in the thermal core throughout the journey, but that during periods when the vehicle is stationary and ventilation consequently reduced there is a relative thermal

equilibration of the different locations within the load. The microenvironments in those sites that previously exhibited the minimal thermal loads approach conditions similar to those in the thermal core.

Examples of the ranges of temperatures and VD occurring in the three locations within the truck in the closed configuration are presented in Table 2. In general, in the reduced ventilation regime with the curtains closed in cold weather, container temperatures in the “thermal core” will approach those observed in summer but accompanied by higher VD, thus constituting higher thermal loads upon the birds. In the thermal core birds may be exposed to temperatures approaching 30 C and VD exceeding 20 g/m<sup>3</sup>, which may impose extreme demands upon the thermoregulatory capacity of the birds when the constraints upon behavior and heat loss associated with the transport container and stocking density are considered. The AET describing the observed “on-board” environments are presented in Table 3. In the front and middle of the load with curtains open, mean values of approximately 50 C were found with maxima of about 60 C. In the curtains closed configuration, the equivalent heat loads ranged from 60 to 80 C. It should be noted that these data were derived from “typical” journeys and therefore the practical range of AET under commercial conditions may greatly exceed that reported here, particularly during “warm” periods in spring and autumn when vehicles may run in the winter configuration with curtains closed.



**FIGURE 1.** Variations in temperature in three locations in a poultry transporter with elapsed time during a typical winter journey (curtains closed configuration). The vehicle was stationary during a mandatory “driver break” between 78 to 118 min.



**FIGURE 2.** Variations in water vapor density in three locations in a poultry transporter with elapsed time during a typical winter journey (curtains closed configuration). The vehicle was stationary during a mandatory “driver break” between 78 to 118 min.

**TABLE 2. Ranges of temperature and water vapor density in three locations within the vehicle trailer during typical summer (curtains open) and winter (curtains closed) journeys**

Variable	Curtain configuration	Ambient	Front	Middle	Rear
Temperature, C	Open				
	Maximum	23.5	29.0	30.4	28.5
	Minimum	19.3	23.1	23.4	22.5
	Closed				
	Maximum	9.1	28.1	26.8	25.1
	Minimum	6.5	21.3	19.2	13.0
Water vapor density g/m <sup>3</sup>	Open				
	Maximum	13.2	14.4	14.1	14.3
	Minimum	8.6	8.6	8.4	8.8
	Closed				
	Maximum	8.4	23.8	21.6	18.2
	Minimum	8.3	10.2	8.9	7.7

Calibration of AET in terms of physiological response or stress profiles was therefore conducted as described using a range of AET consistent with the findings on commercial vehicles but with due consideration of elevated climatic thermal loads. The values of AET employed were 45, 58, 70.4, 81.1 and 91.5 C. These were achieved at "in-crate" temperatures between 22 and 30 C and VD 10.5 to 27.0 g/m<sup>3</sup>.

The physiological responses of broiler chickens to these heat loads appear in Tables 4 to 8. Confinement of the birds in the crates tends to induce hyperthermia at all the heat loads employed (Table 4). At an AET of about 70 C, the hyperthermia becomes marked (>1.0 C) and at an AET of 80 C or more, the hyperthermia is profound and may become life threatening. Disturbances in acid-base balance and pCO<sub>2</sub> due to thermal panting, followed generally similar patterns in relation to heat load as body temperature (Tables 5 and 6). A marked degree of physiological stress may be present at AET >60 C indicated by the marked respiratory hypocapnia and alkalosis occurring at the two higher values of AET (>70 C). Plasma CK activities increased in all groups of birds confined to transport containers, perhaps reflecting the physical stress of this procedure (Table 7). Plasma CK of 1,000 IU/L or greater resulted from exposure to all AET above 45 C although the response tended to be proportional to AET. The elevations of CK of 45 to 50% associated with exposure

to AET of 81.1 and 91.5 C, suggest the presence of extensive disruption of sarcolemmal integrity at these heat loads and therefore significant muscle damage (Mitchell and Sandercock, 1995). Increases in corticosteroid secretion, as reflected by the H:L ratio (Gross and Siegel, 1993) were significant at AET values of 70 C and above and were proportional to heat load (Table 8).

The responses in all the physiological parameters measured are presented in Table 9. The values represent the mean changes in each variable during a 3-h exposure period to each temperature-humidity combination (AET). The lowest heat load consisted of a temperature of 22 C and a VD of 10.5 g/m<sup>3</sup>. The higher heat loads were achieved at two dry bulb temperatures of 25 and 30 C by adjusting the VD to values in the range 14.8 to 27.0 g/m<sup>3</sup>.

These stress profiles suggest that at heat loads associated with AET of 45 to 50 C, physiological stress would be minimal or mild. At values of AET between 50 and 70 C, moderate to severe and increasing physiological stress associated with failure of several homeostatic systems, tissue dysfunction and damage, metabolic derangements and activation of the HPA will be observed with increasing mortality. If AET reaches 90 C or more, then extremely severe stress will occur with catastrophic failure of thermoregulation, profound hyperthermia, collapse and death in large numbers of birds in commercial practice. It is suggested that temperature-humidity combinations constituting AET of

**TABLE 3. Mean and maximum apparent equivalent temperatures at three locations in the vehicle trailer during typical summer (curtains open) and winter (curtains closed) journeys**

Curtain configuration	Apparent equivalent temperature		
	Front	Middle	Rear
	(C)		
Open			
Mean	51.9	50.1	47.5
Maximum	61.4	62.3	60.6
Closed			
Mean	61.8	56.9	51.1
Maximum	81.7	75.4	66.0

**TABLE 4. Body temperature in broiler chickens exposed to a range of thermal loads ( $\theta^*_{app}$ ) for 3 h under simulated transport conditions. Values are given as the mean  $\pm$  1 SD (n = 10)**

$\theta^*_{app}$	Body temperature	
	T <sub>0</sub>	T <sub>1</sub>
	(C)	
45.0	41.6 $\pm$ 0.13	42.0 $\pm$ 0.25
58.0	41.7 $\pm$ 0.17	42.4 $\pm$ 0.29
70.4	41.5 $\pm$ 0.17	42.5 $\pm$ 0.30
81.1	41.5 $\pm$ 0.11	43.0 $\pm$ 0.33
91.5	41.3 $\pm$ 0.23	44.6 $\pm$ 0.33

TABLE 5. Venous blood pH in broiler chickens exposed to a range of thermal loads ( $\theta^*_{app.}$ ) for 3 h under simulated transport conditions. Values are given as the mean  $\pm$  1 SD (n = 10)

$\theta^*_{app.}$	Venous Blood pH	
	T <sub>0</sub>	T <sub>1</sub>
45.0	7.28 $\pm$ 0.04	7.29 $\pm$ 0.04
58.0	7.34 $\pm$ 0.04	7.36 $\pm$ 0.05
70.4	7.32 $\pm$ 0.05	7.36 $\pm$ 0.06
81.1	7.29 $\pm$ 0.05	7.37 $\pm$ 0.04
91.5	7.32 $\pm$ 0.04	7.41 $\pm$ 0.03

65 C or greater should always be avoided in commercial practice by means of the appropriate preventive strategies. Thermal loads associated with AET of 40 to 50 C may represent an acceptable "in-crate" microenvironment for broiler transportation.

These experimental data relate to standard journey simulations of 3 h duration and with constant conditions. During commercial transportation, journey length and periods of low ventilation and radiant gain during unavoidable stationary periods may exacerbate the imposition of physiological stress. These factors should be considered when interpreting the likelihood of moderate or severe stress and the associated occurrence of potential failure of compensatory mechanisms, decompensation, and important perturbations in homeostasis and welfare.

The determination of physiological stress profiles related to specific thermal loads that may occur in the thermal core of commercial transporters, facilitates definition of acceptable ranges and limits for temperatures and humidities within the "bio-load". In turn, strategies for matching the "on-board" micro-environment to the birds' biological requirements or "safe-zone" can be developed from a knowledge of the appropriate thermal envelope.

## DISCUSSION

The thermal data pertaining to the conditions within the broiler crates serve to emphasize the complexity of the commercial transport microenvironment. If the average metabolic rate of a 2-kg bird is 15 W, then in

TABLE 6. Venous blood pCO<sub>2</sub> in broiler chickens exposed to a range of thermal loads ( $\theta^*_{app.}$ ) for 3 h under simulated transport conditions. Values are given as the mean  $\pm$  1 SD (n = 10)

$\theta^*_{app.}$	Venous Blood pCO <sub>2</sub>	
	T <sub>0</sub>	T <sub>1</sub>
45.0	52.5 $\pm$ 6.2	48.7 $\pm$ 4.6
58.0	45.3 $\pm$ 5.8	45.8 $\pm$ 7.3
70.4	49.9 $\pm$ 6.6	43.8 $\pm$ 7.9
81.1	48.0 $\pm$ 4.4	38.6 $\pm$ 3.5
91.5	50.1 $\pm$ 5.0	36.6 $\pm$ 3.1

TABLE 7. Plasma creatine kinase (CK) activity in broiler chickens exposed to a range of thermal loads ( $\theta^*_{app.}$ ) for 3 h under simulated transport conditions. Values are given as the mean  $\pm$  1 SD (n = 10)

$\theta^*_{app.}$	Plasma CK activity	
	T <sub>0</sub>	T <sub>1</sub>
45.0	693 $\pm$ 257	888 $\pm$ 324
58.0	696 $\pm$ 296	1,043 $\pm$ 432
70.4	652 $\pm$ 178	996 $\pm$ 241
81.1	830 $\pm$ 323	1,205 $\pm$ 393
91.5	810 $\pm$ 215	1,239 $\pm$ 410

excess of 90 kW of heat and the associated obligatory metabolic water production (10.5 g/h) must be dissipated by the ventilation of a typical broiler transporter. Any impairment of airflow through the structure will result in the accumulation of heat and moisture that in combination will impose heat stress upon the birds. The consequent stimulation of thermal panting will increase evaporative water loss from the animals, adding to the moisture load, further precipitating heat stress and creating a vicious spiralling cycle of hyperthermia. The installation of curtain sides will create conditions of low ventilation that will exacerbate this process and will lead to the presence of a "thermal core" in which the risk of heat stress is greatest. The core is located immediately behind the headboards and towards the top of the compartment in both truck and trailer. The thermal conditions in the core may deteriorate further when the vehicle is stationary. The detrimental effects of such heat loads will be exacerbated by the constraints imposed upon the birds behavioral thermoregulatory capacity by the high stocking density within the crates or containers. These observations will be generally applicable to many types of poultry transport vehicle currently employed in the U.K., Europe and North America.

It is apparent that whereas heat stress may occur in the open configuration when external temperature and humidity are elevated or the vehicle is stationary, the risk will be reduced in all but extreme conditions if the VD is minimized by adequate airflow through the load. In the winter with the curtains closed, however, a "paradoxical heat stress" may be induced in the thermal core even in temperate or cool climatic conditions.

TABLE 8. Heterophil: lymphocyte(H:L) ratios in broiler chickens exposed to a range of thermal loads ( $\theta^*_{app.}$ ) for 3 h under simulated transport conditions. Values are given as the mean  $\pm$  1 SD (n = 10)

$\theta^*_{app.}$	H:L ratio	
	T <sub>0</sub>	T <sub>1</sub>
45.0	0.45 $\pm$ 0.13	0.47 $\pm$ 0.19
58.0	0.35 $\pm$ 0.18	0.39 $\pm$ 0.20
70.4	0.48 $\pm$ 0.26	0.63 $\pm$ 0.10
81.1	0.23 $\pm$ 0.10	0.69 $\pm$ 0.18
91.5	0.48 $\pm$ 0.26	1.35 $\pm$ 0.23



**TABLE 9. Mean changes in body temperature, blood pH, pCO<sub>2</sub>,<sup>1</sup> creatine kinase activity, and heterophil:lymphocyte ratio (H:L) in response to a 3 h exposure to different apparent equivalent temperatures ( $\theta^*_{app}$ ) during broiler transport simulations<sup>1</sup>**

$\theta^*_{app}$	BT	pH	pCO <sub>2</sub>	CK	H:L
	(C)		(torr)	(IU/L)	
45	+0.4	+0.02	+3.8	+195	+0.02
58	+0.7	+0.02	+0.5	+347	+0.03
70	+1.0	+0.04	-6.1	+344	+0.21
81	+1.5	+0.08	-9.4	+375	+0.46
92	+3.3	+0.10	-13.5	+429	+0.87

<sup>1</sup>pCO<sub>2</sub> = partial pressure of carbon dioxide.

Dissipation of the temperature and humidity gradients and the heterogeneous distribution of thermal loads within the vehicle should therefore be the primary objective of improved designs of ventilation systems for broiler transporters.

The physiological responses characterized in the laboratory simulation experiments indicate that AET is a useful predictor of thermal stress during exposure to different temperature-humidity combinations. The results also underline the importance of VD in determining the thermal loads imposed upon the birds. The influence of humidity upon evaporative heat exchange is well recognized (Richards, 1976; Dawson, 1982). It has previously been predicted, on theoretical grounds, that increasing RH from 20 to 80% in a transport crate at a temperature of 28 C would result in a thermal load upon the birds equivalent to an additional rate of rise of body temperature of 0.42 C/h (Mitchell *et al.*, 1990). In the present study, raising RH at elevated temperature exerted marked effects upon the degree of physiological stress experienced by the birds as indicated by the relationships between the physiological variables and AET. The rise in deep body temperature with increasing heat load accelerated sharply at values of AET above 70.

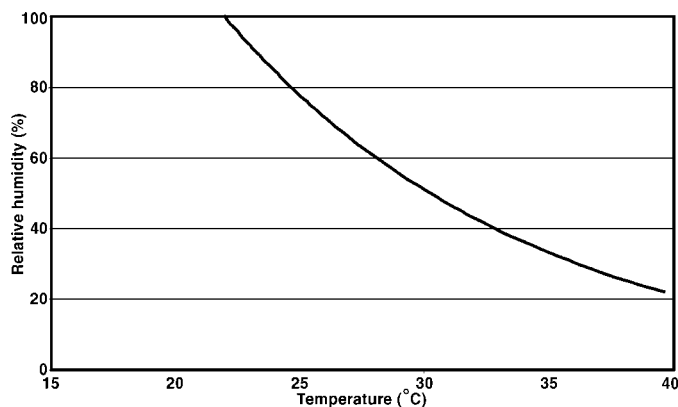
Similarly, the thermoregulatory effort expended by the birds at such thermal loads is reflected in the extent of the hypocapnia and alkalosis resulting from the induced thermal panting. These effects are also proportional to thermal load. Respiratory overventilation during panting produces excessive elimination of carbon dioxide and thus increases blood pH (Mather *et al.*, 1980; Arad, 1983), as observed in this study. At high thermal loads, thermoregulatory and acid-base homeostatic mechanisms may become antagonistic and severe acid-base disturbances may be superimposed upon the direct effects of hyperthermia. The proportionality of such perturbations in blood chemistry and the respiratory response to the imposed thermal loads are consistent with previous reports (Mitchell, 1986) and are further supported by measurements of respiratory frequency in complementary transport simulation studies (Mitchell and Kettlewell, unpublished data). An increase in CK in the circulation due to disturbances in muscle membrane

integrity has been described in broilers in response to a number of environmental challenges, including heat stress and transportation (Ostrowski-Meissner, 1981; Mitchell *et al.*, 1992). The present study demonstrates that this phenomenon is also related to the degree of stress imposed.

The H:L ratio is an indicator of the activity of the HPA and increases with the magnitude of the heat load upon the birds, more profound elevation of H:L being observed at AET values >70 C. The predictive nature of AET in relation to the physiological stress indices employed, might be expected as it incorporates a measure of temperature *per se* and of the VD gradient between the evaporative surfaces of the bird and the environment and, thus, the efficiency of this avenue of heat exchange. Increased physiological stress will be consequent upon the compromise and inadequacy of the thermoregulatory response at high temperatures and VD.

A composite profile of all the measured physiological responses reflects the degree of physiological stress imposed by each temperature-humidity combination and has therefore been employed to identify acceptable ranges and limits. Values of AET of <45 C may be regarded as presenting no risk to the welfare or survival of the birds under normal transport conditions. Between 45 and 65 C, the physiological stress is designated as moderate and is associated with disturbances in blood chemistry, metabolism, and tissue function. At AET >65 C, stress is severe, tissue damage will occur, and mortalities will increase. It is suggested that bird welfare will be compromised under the conditions precipitating both moderate and severe physiological stress and that the temperature-humidity combinations resulting in the latter state should be avoided at all times. The temperature-humidity combinations yielding an AET value of 65 C are shown in Figure 3. For example, the extreme theoretical values producing the equivalent thermal load are 65 C in completely dry air and 22.2 C at an RH of 100%. Air at 40 C and 21% RH would result in the same AET. The studies described here have indicated that, realistically, a RH of less than 50% will rarely occur in the thermal core under commercial transport conditions regardless of temperature. Therefore, even if water vapor could be controlled to this level, the absolute maximum permissible dry bulb temperature would be 30 C. The range of RH more commonly encountered in the thermal core is of the order of 70 to 80% and thus the onset of severe physiological stress may be expected at 25 to 26 C. Ventilation should thus be adequate to maintain core temperature below this level and be sufficient to minimize the concurrent water vapor load.

By controlling the thermal environment in accordance with these prescribed ranges, it is possible to minimize the incidence of heat stress and improve the welfare of poultry in transit. It should be emphasized that the ranges have been defined for an exposure period of 3 h



**FIGURE 3.** Dry bulb temperature and relative humidity combinations yielding an Apparent Equivalent Temperature of 64 C, the recommended acceptable upper limit for the thermal load in the core of a commercial poultry transporter.

and at stocking densities of approximately 53 to 58 kg/m<sup>2</sup>, which compare very favorably with standards proposed by the EU (EC 91/628, 1993). Whereas the resulting recommendations will be appropriate for the majority of journeys in commercial practice in the U.K., the thermal limits for those of much longer duration will require appropriate adjustment. It should also be noted that the current study has specifically addressed the problem of heat stress in transit but the approaches and philosophies employed may be applied to cold stress in future investigations.

In a more recent complementary research program, a method for on-line routine monitoring of the environment within the thermal core of commercial vehicles has been developed. This program utilizes the physiological response model data to provide a warning system indicating the risk of impending heat stress during journeys (Mitchell, 1992). From temperature and humidity measurements within the transport space, a predictive computer program calculates the AET within the thermal core and from this value indicates the potential degree of physiological stress. Simple modifications of the side-curtains of existing vehicles to facilitate opening during a journey allow the driver to respond to such a warning by increasing ventilation.

Integration of the above microenvironment analysis and physiological stress response models with computational fluid dynamic modelling (Baker, 1994) and measurements of surface pressure profiles on moving transporters has resulted in recommendations for improvements in vehicle and ventilation system design that are currently being evaluated under commercial conditions.

The pressure fields surrounding commercial transporters have been employed to identify the locations of pressure gradients driving natural ventilation when the vehicle is in motion. Through mathematical and physical modelling, design modifications that exploit favorable gradients and that should increase ventilation of the

thermal core have been developed (Baker, 1994). A vehicle modified in this manner and capable of carrying an additional two modules (approximately 500 birds) has been compared with one of its predecessors with a conventional ventilation system. In matched and well-controlled, back-to-back, alternate journeys over identical routes on 4 consecutive d, the internal thermal core environments have been characterized in addition to the assessment of the physiological stress experienced by the birds and productivity on each of the two vehicles. The results are presented in Table 10. In the thermal core of the standard vehicle, the average temperature on these journeys was 28.7 C accompanied by a RH of 63%, i.e., a VD of 17 g/m<sup>3</sup>, and thus an AET of 68.8 C (above the recommended limit previously prescribed). Associated with this environment were clear indications of physiological stress, in particular a marked elevation of the H:L ratio. In the modified, better ventilated vehicle, under the same external climatic conditions, the average core temperature was only 22.5 C and the corresponding RH 51%, yielding a VD of 10.2 g/m<sup>3</sup> and an AET of 45.1 C. The improved ventilation reduces the thermal load in the core as evidenced by the lower AET and, as a consequence, physiological stress as measured by CK and H:L responses, may be decreased on the modified vehicle. The results of this preliminary study suggest that a relatively simple modification of vehicle design intended to match thermal environments to the birds' biological requirements may provide significant improvements in welfare during transit concomitant with an increase of approximately 10% in the number of birds delivered per journey and a reduced mortality.

The success of this research program has led to the application of the approaches and philosophies employed to stressors encountered during the transportation of day-old chicks, turkeys, and other species, including pigs, sheep, and cattle. New studies are addressing journey durations and limits for each species, optimum stocking densities, vehicle ventilation require-

**TABLE 10.** Comparison of environmental conditions (thermal core), physiological stress responses and productivity on two broiler transporters on four matched journeys on consecutive days

Variable	Vehicle A	Vehicle B
Core temperature, C	22.5	28.7
Core vapor density, g/m <sup>3</sup>	10.2	17.8
CK, IU/L	310	407
H:L	0.30	0.97
Mean load, (no. of birds <sup>3</sup> )	6,120	5,580
Mean dead-on-arrival, %	0.43	0.49
Total birds delivered	24,375	22,210

<sup>1</sup>Each journey lasted 3 h, the mean external environmental temperature was 11.7 C, with a water vapor density of 6.6 g/m<sup>3</sup>. Vehicle A had a modified ventilation system aimed at improving thermal homogeneity within the load, whereas Vehicle B was unmodified. The modified vehicle had an increased capacity of two extra modules

<sup>3</sup>Birds were loaded at 21 to 22 per crate.

ments, feeding and watering intervals, frequency and duration of rest stops, improvements in handling and transport procedures and the design of protective strategies (e.g., pretransport husbandry and nutrition and "anti-stress" treatments). The problems of heat stress on stationary vehicles and in lairage, and the design and specification of forced ventilation systems for application in such circumstances, are also being investigated using a definition of physiologically optimum environments.

Finally, in addition, it is proposed that the results of these studies should, at least in part, provide the informed, relevant and scientifically sound basis for codes of practice and legislation relating to the transportation of live animals. In this context, it is proposed that a "10 point plan" be adopted as the foundation for future projects in this field. 1) Characterize the transport process and environment under practical conditions. 2) Identify the major stressors imposed. 3) Identify appropriate indices of physiological stress. 4) Produce integrative, predictive models relating physiological stress response to quantified stressors ("dose-response curves"). 5) From the relevant physiological stress profiles determine the acceptable ranges and limits for the stressor. 6) Examine interactions among concurrent multiple stressors including summation or facilitation of effects. 7) Test laboratory derived models under "field conditions". 8) Design appropriate strategies for the alleviation or prevention of stress including the control of stressors within the prescribed ranges and limits. 9) Use the model to evaluate the effectiveness or success of the strategies. 10) Use this philosophy and approach as the scientific basis for recommendations for improvements in future vehicle and container design, transport practices and legislation.

The techniques and findings reported in this study facilitate both the prediction and prevention of thermal stress in broilers in transit. It is suggested that such exploitation of the symbiotic relationship between the scientific disciplines of biology, engineering, and physics represents an important strategy for progress in the study of broiler transportation and the compatible optimization of bird welfare and productivity in modern agricultural systems.

It is proposed that this is an example of the effective application of fundamental biological and engineering research to provide practical solutions to welfare problems.

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