AGRICULTURE AND FOOD DEVELOPMENT AUTHORITY

The Irish Agriculture and Food Development Authority

An overview of the latest research examining the impact of stress on the health and welfare of beef cattle

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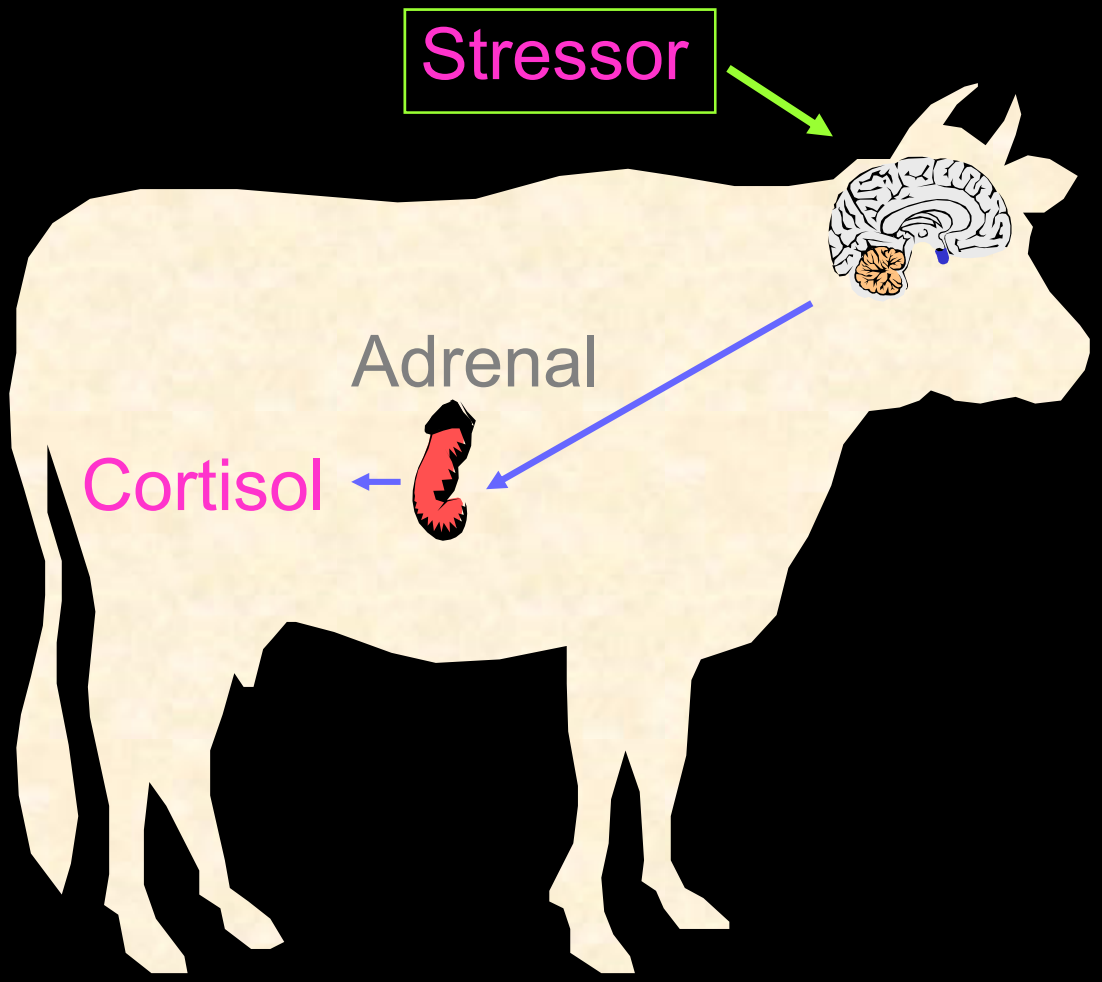
Agriculture and Food Development Authority

The Irish Agriculture and Food Development Authority

The Basics: Generalized responses

The Hypothalamic-Pituitary-Adrenal Axis



Animal Transport studies

http://www.bordbia.ie/industry/farmers/pricetracking/cattle/pages/qualityirishlivestock-video.aspx

Irish cattle exports

(000' head)	2013	2014	% change
Total	**128,342**	**154,286**	**+20%**
NI	17,914	19,135	+7%
GB	4.944	7,041	+42%
Italy	10,667	14,294	+34%
Spain	28,162	36,558	+30%
Netherlands	30,481	37,982	+25%
Belgium	19,119	20,583	+8%
Int. markets	8,464	12,845	+52%

IRELAND (EU)

IRELAND

AUSTRALIA

Table 1: Animal transport – scientific assessment

Journey	Animal type (weanlings)	Study date	Number (N) of animals transported (stocking density/animal)	Number (N) of animals remaining on control farms in Ireland	Regulation
Ireland to Spain	Heifers (270kg)	December 2001	N = 52 (0.9m^2)	N = 28	Directive 91/628/EEC
Ireland Stocking density	Bulls (250kg)	May 2002	N = 16 (0.85m^2) N = 13 (1.27m^2)	N = 16	Stocking density study
Ireland to Italy	Bulls (414 kg)	October 2002	N = 26 (1.2m^2)	N = 22	Directive 91/628/EEC
Ireland to Spain	Heifers (245 kg)	November 2003	N = 40 (0.8m2)	N = 20	COM (2003) 425

Table 2: Animal transport – scientific assessment

Journey Within Ireland	Animal type (weanlings)	Study date 2004	Number (N) of animals transported (stocking density/animal)	Number (N) of control animals	Regulation (91/628)
0	Bulls (370 kg)	Feb.-March	-	N = 24	(91/628)
6 hr	Bulls (371kg)	Feb.-March	N = 24 ($1.0m^2$)	-	(91/628)
9 hr	Bulls (371kg)	Feb.-March	N = 24 ($1.0m^2$)	-	(91/628)
12 hr	Bulls (371kg)	Feb.-March	N = 24 ($1.0m^2$)	-	(91/628)
18 hr	Bulls (385kg)	Feb.-March	N = 24 ($1.0m^2$)	-	(91/628)
24 hr	Bulls (383kg)	Feb.-March	N = 24 ($1.0m^2$)	-	(91/628)
9-12-9+2 hr	Bulls (462kg)	April	N = 15 ($1.2m^2$)	N = 15	9-12-9+2

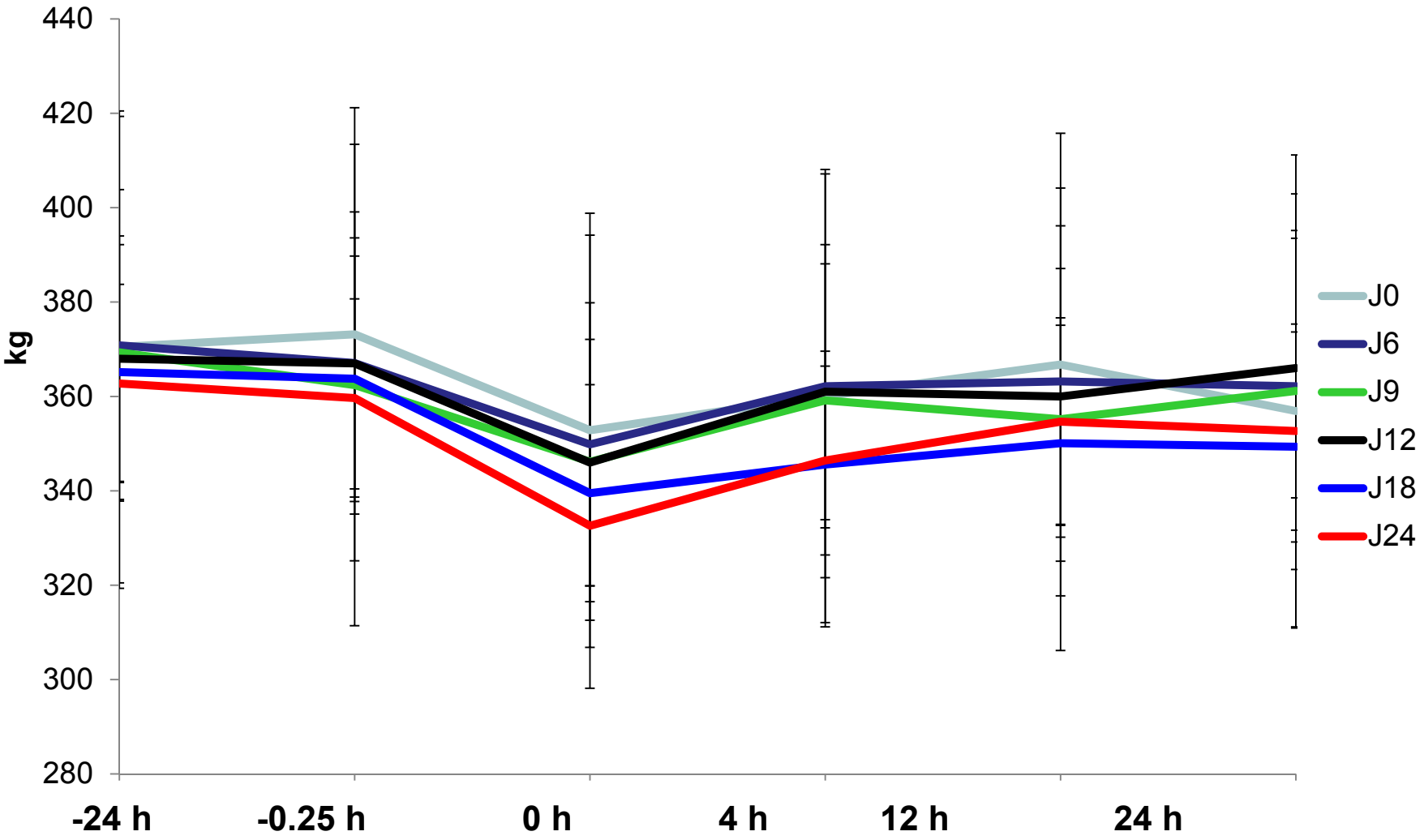
Table 3: Animal transport – scientific assessment

Journey	Animal type	Study date 2005	Number (N) of animals transported (stocking density/ animal)	Number (N) of control animals	Regulation (91/628)
Ireland to Lebanon	Bulls	October	57	54	
Australia	Heifers	July 2006	Pre-transport fasting & journey durations 84		

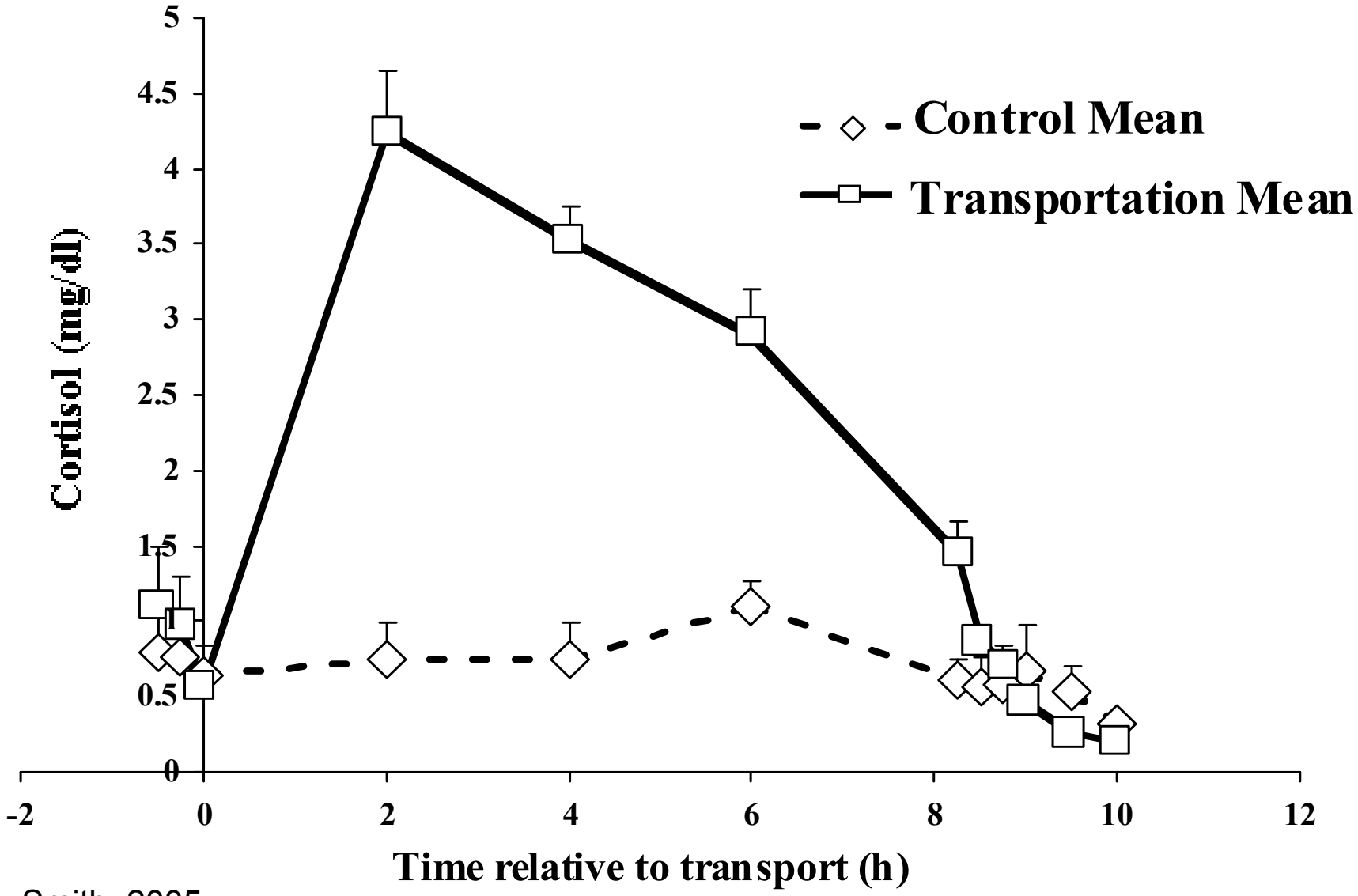
Mean Liveweight loss (%) in control and transported animals prior to and in the 24 hour period after different transport journey durations (J) in hours (0, 6, 9, 12, 18 and 24) (n = 12 bulls per treatment).

Journey (h)	Post-transport	24 hour (h) recovery post-transport journey				Overall weight loss
		0 – 4 h	5 – 12 h	0 - 12 h	13 – 24 h	
J0	-5.4	1.9	1.8	-1.7	-2.7	-3.7
J6	-4.7	3.4	0.2	-1.1	-0.5	-2.3
J9	-4.5	3.7	-1.2	-2.0	1.7	-2.2
J12	-5.7	4.1	-0.3	-2.0	1.7	-0.5
J18	-6.6	1.7	1.3	-3.8	-0.2	-4.3
J24	-7.5	4.0	2.3	-1.4	-0.6	-2.8

Live weight changes (Journeys 0h to 24h)



Changes in Plasma cortisol over a 10-hour journey period in transported animals. Control animals remained on farm of origin.



Smith, 2005

Physiology and Immunology

- **Physiological variables**

Cortisol, glucose, lactate, free fatty acids, beta-hydroxy butyrate, urea, total protein, albumin, creatine phosphokinase (CK), lactate dehydrogenase (LDH), and the acute phase proteins (fibrinogen and haptoglobin).

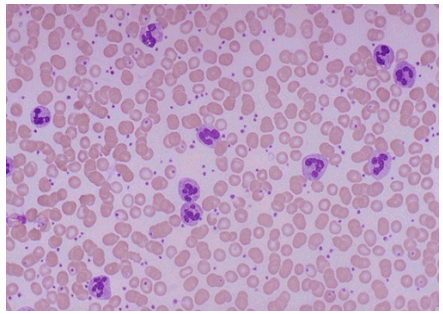
- **Immunological variables**

Stimulated lymphocyte production of interferon gamma in response to keyhole limpet haemocyanin (KLH) and Concanavalin-A (Con-A).

Welfare Indices

- Physiological, haematological and immunological variables were used to determine the welfare status of animals, before, during and after the respective transport journeys.

- Age-matched control animals, retained in Ireland, were blood sampled for physiological and haematological parameters at times corresponding to the transported animals.

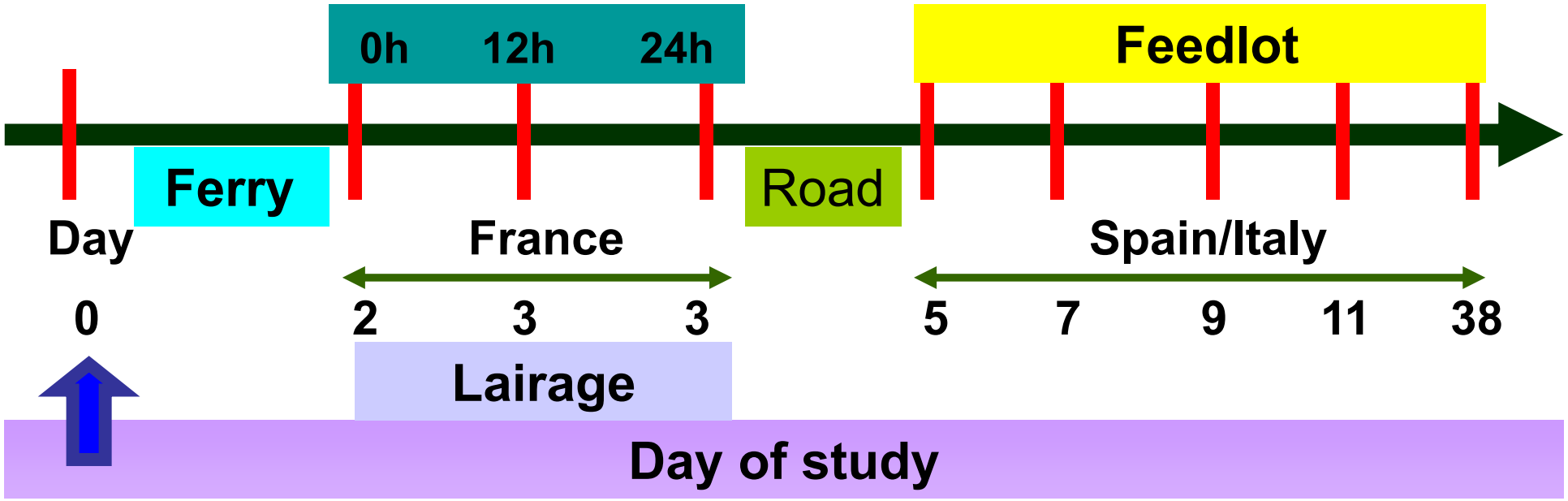


Blood smear

Experimental plan – Spain & Italy

Animals were **blood sampled** on farm of origin to provide baseline physiological, haematological and immunological welfare indices **(day 0)**

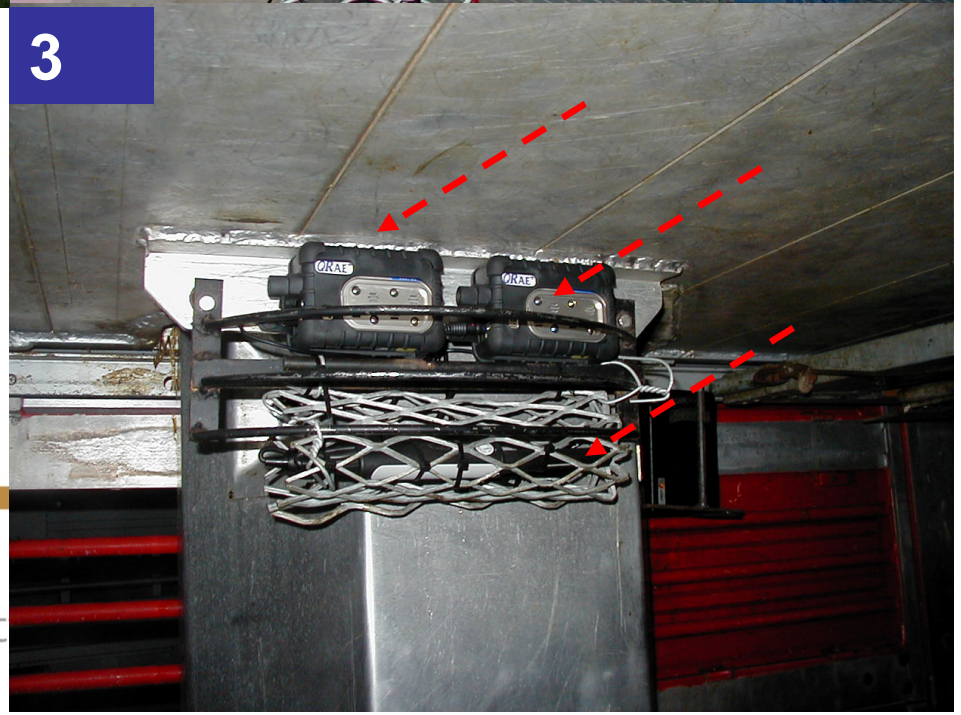
Rectal body temperature prior to transportation (day 0) and on days 2, 3, 5, 7, 9, 11 and 38.
Liveweights prior to transportation (day 0) and on days 2, 5, 11 and 38.

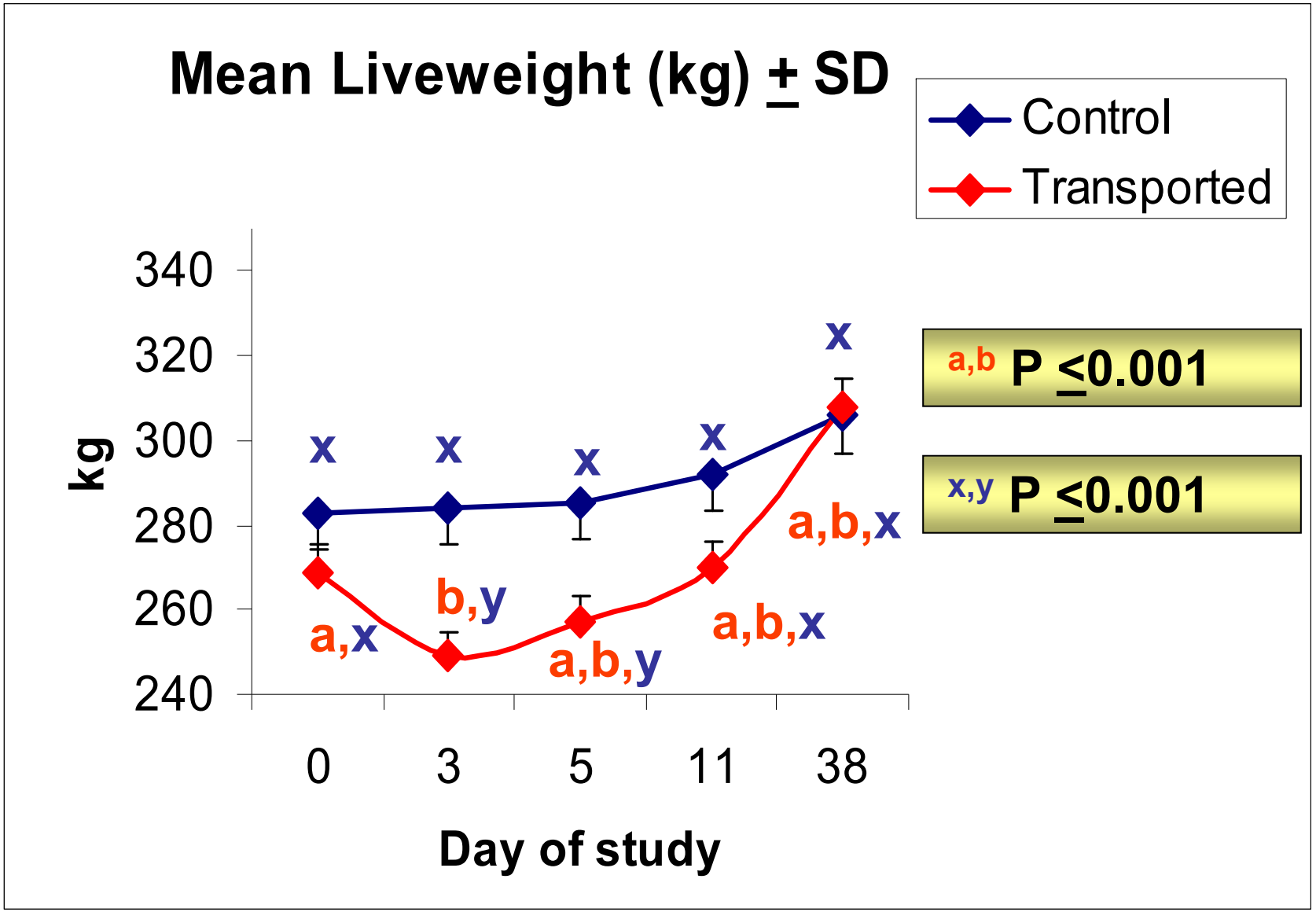


1. Transporter

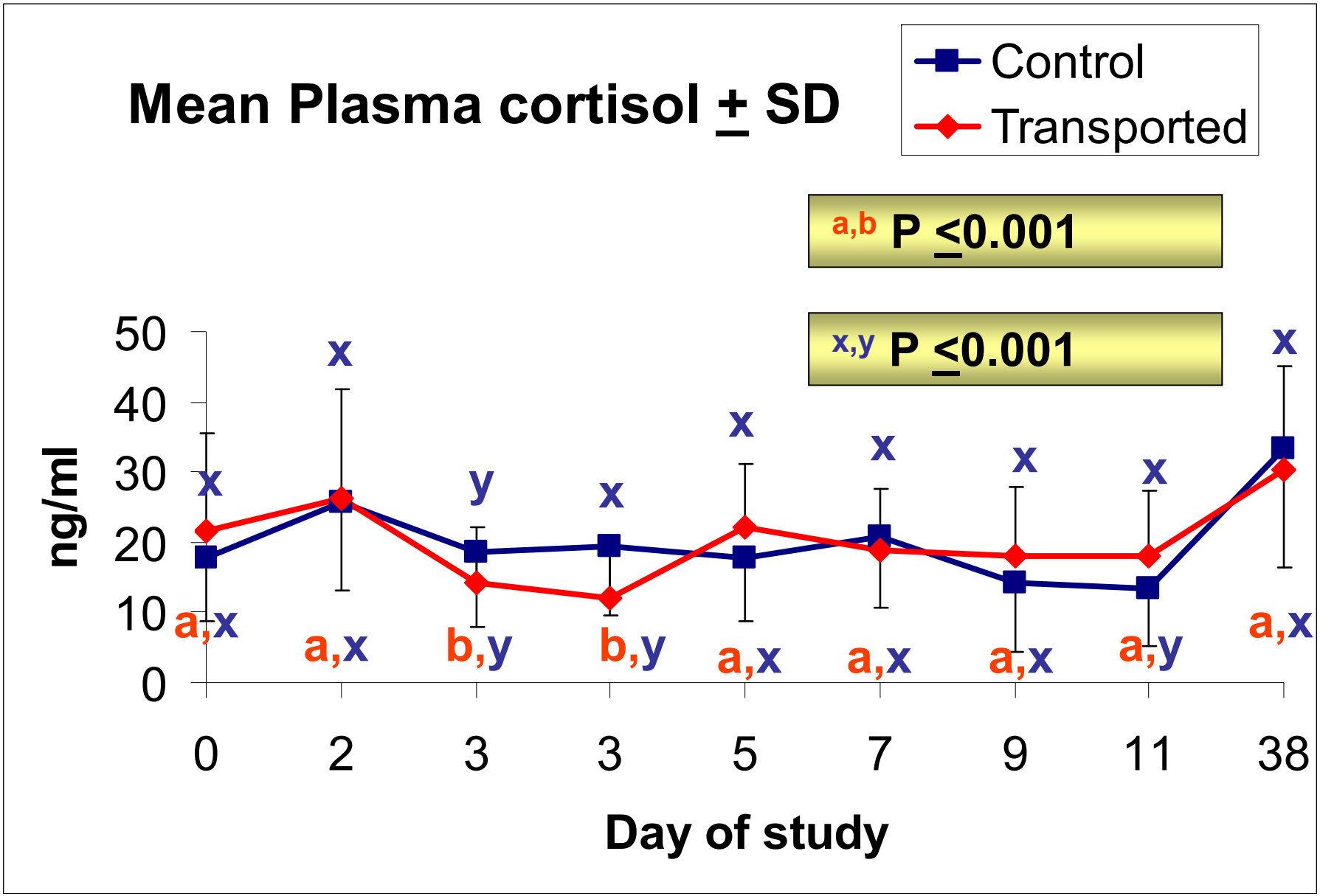
2. Upper deck of transporter with computerised video and environmental monitoring equipment

3. Lower deck of transporter showing the environmental probes

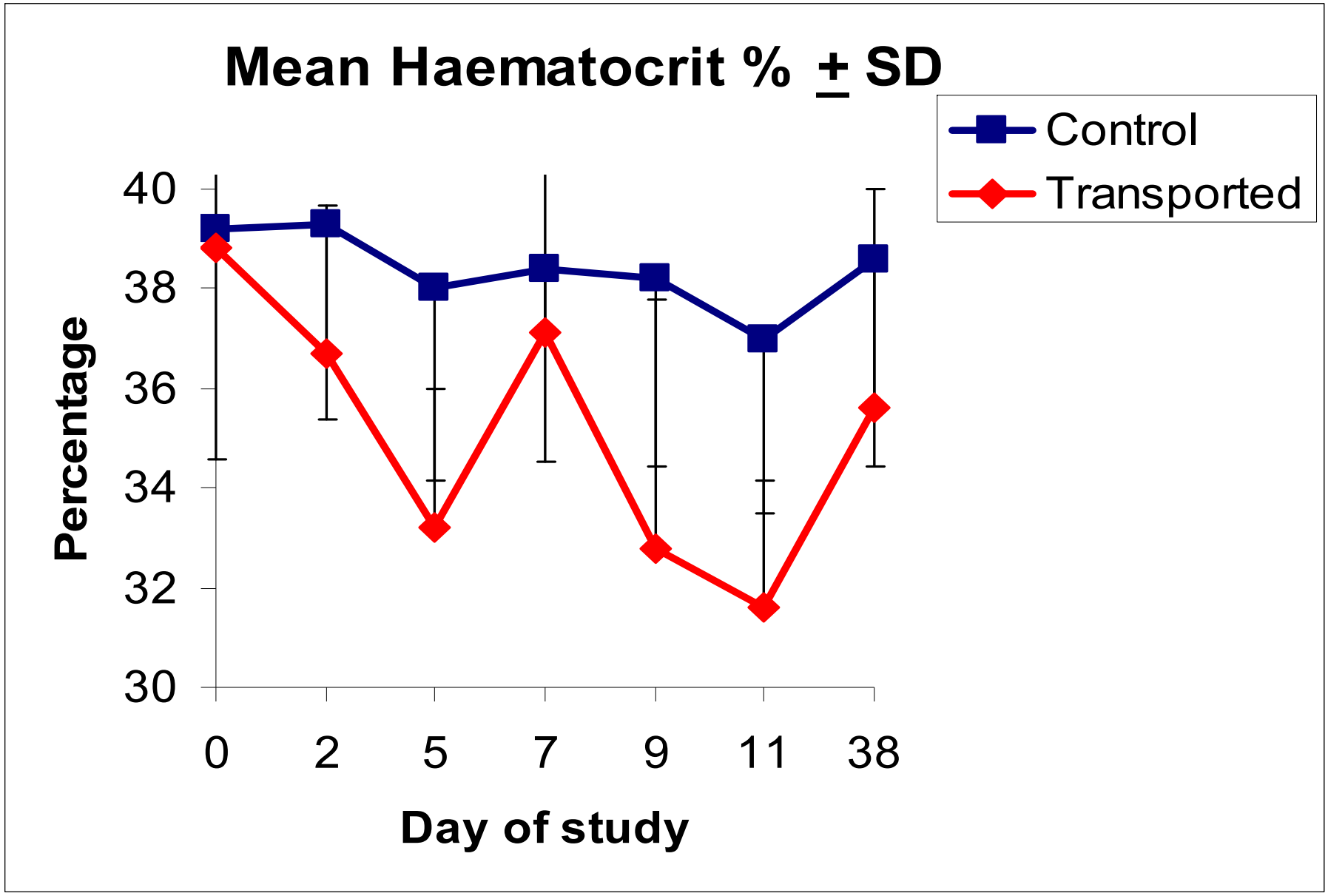




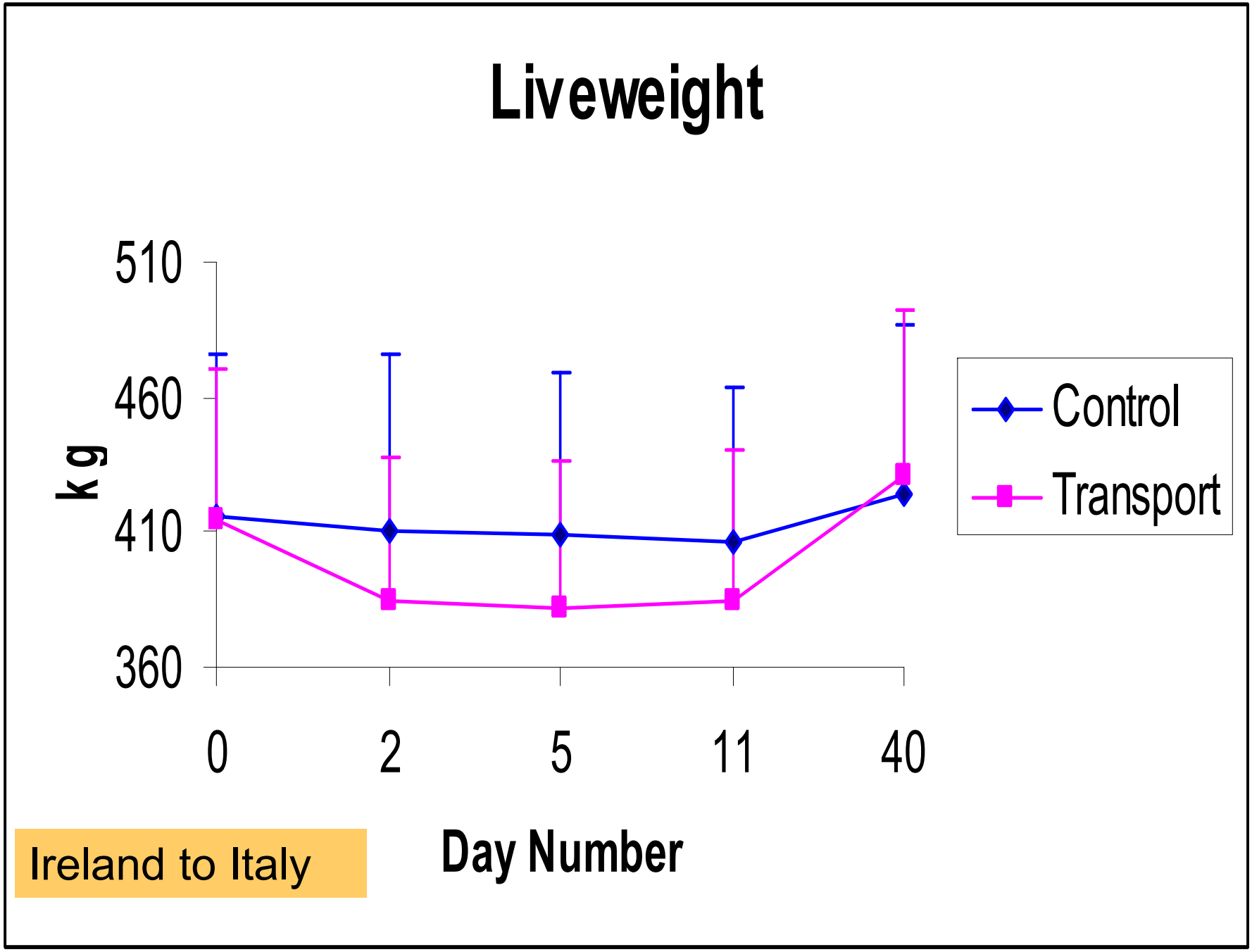
Ireland to Spain



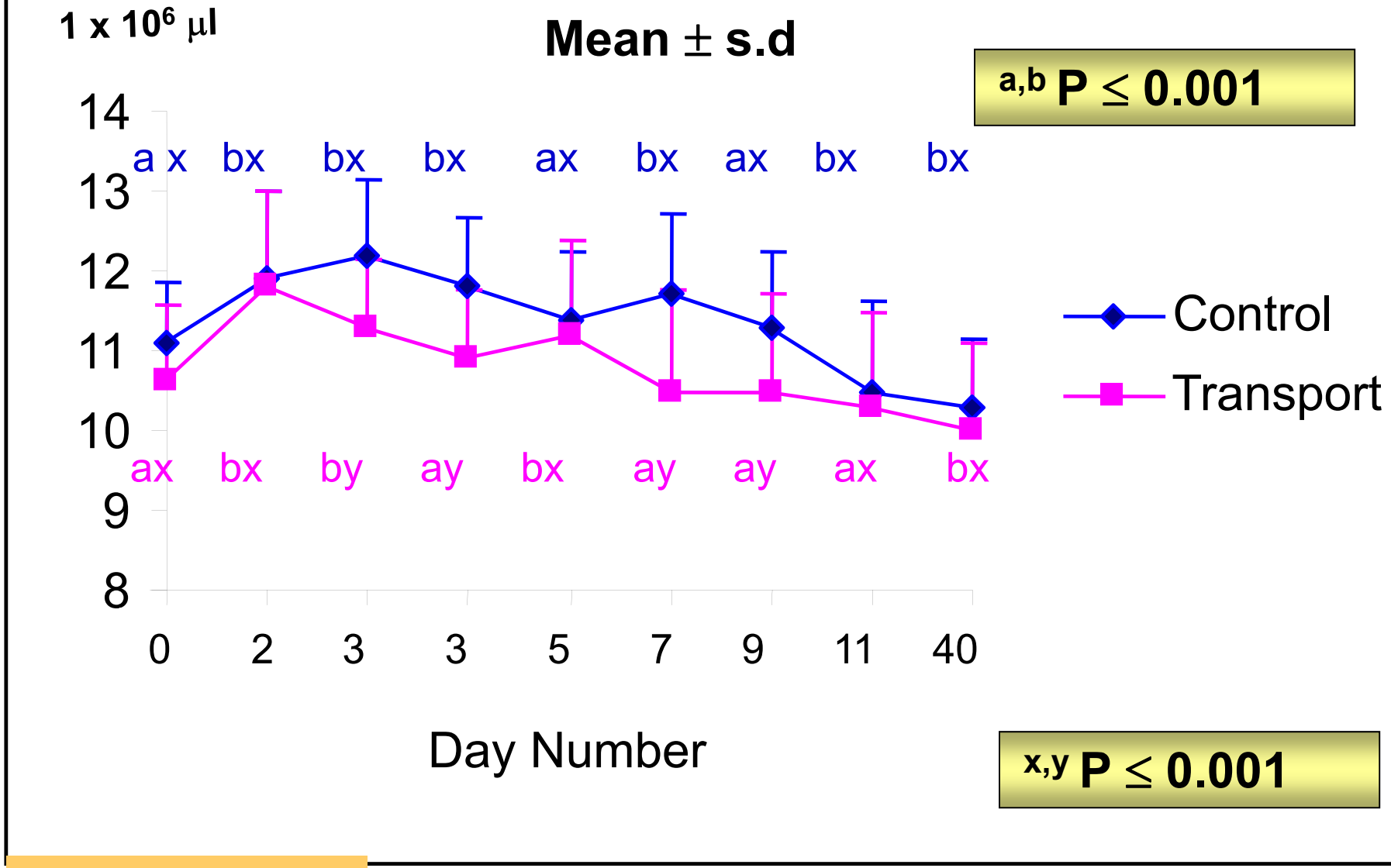
Ireland to Spain



Ireland to Spain

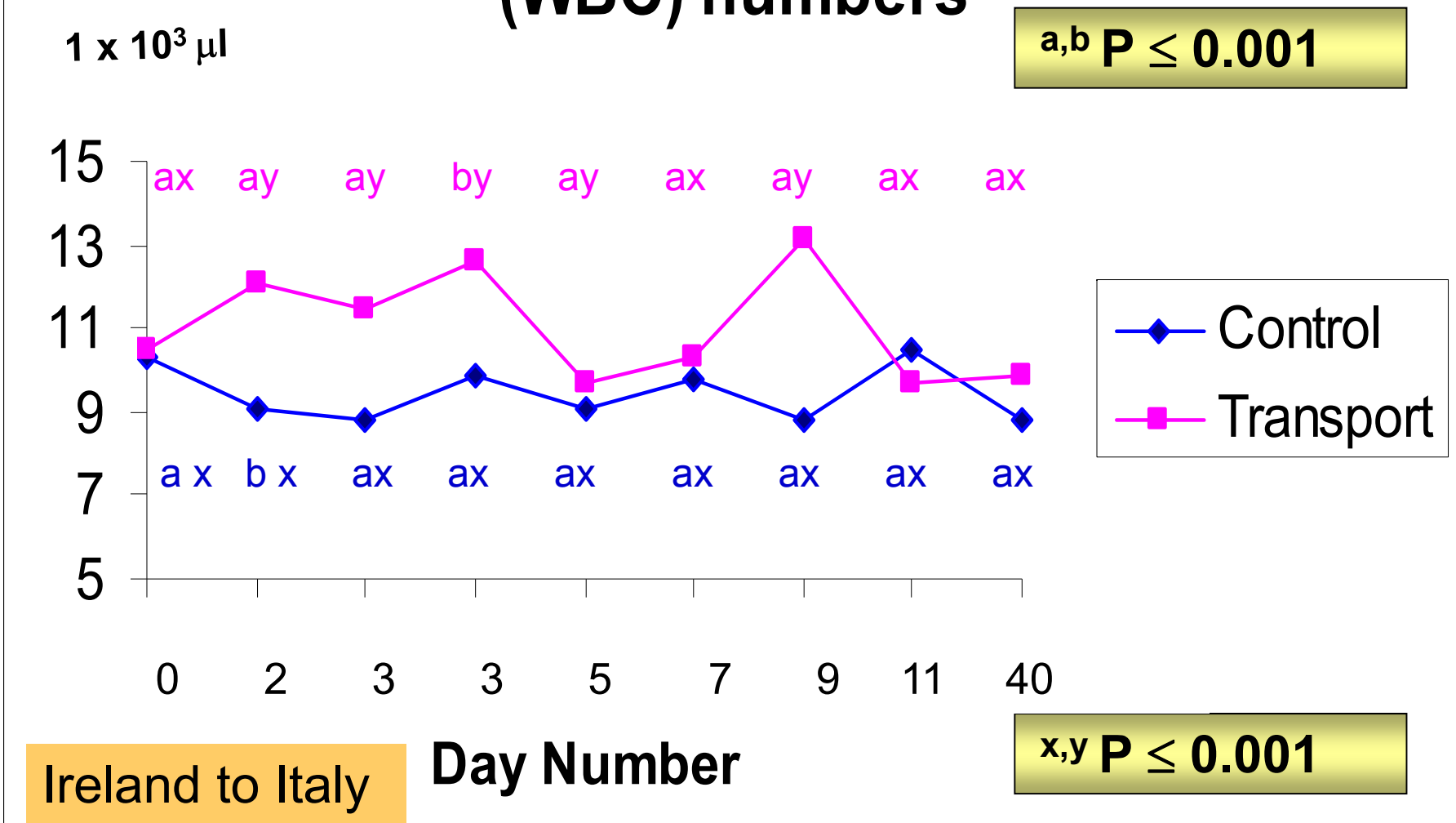

Liveweight
510
460
410
360
kg
0
2
5
11
40
Day Number
Control
Transport
Ireland to Italy

Red Blood Cell (RBC) numbers

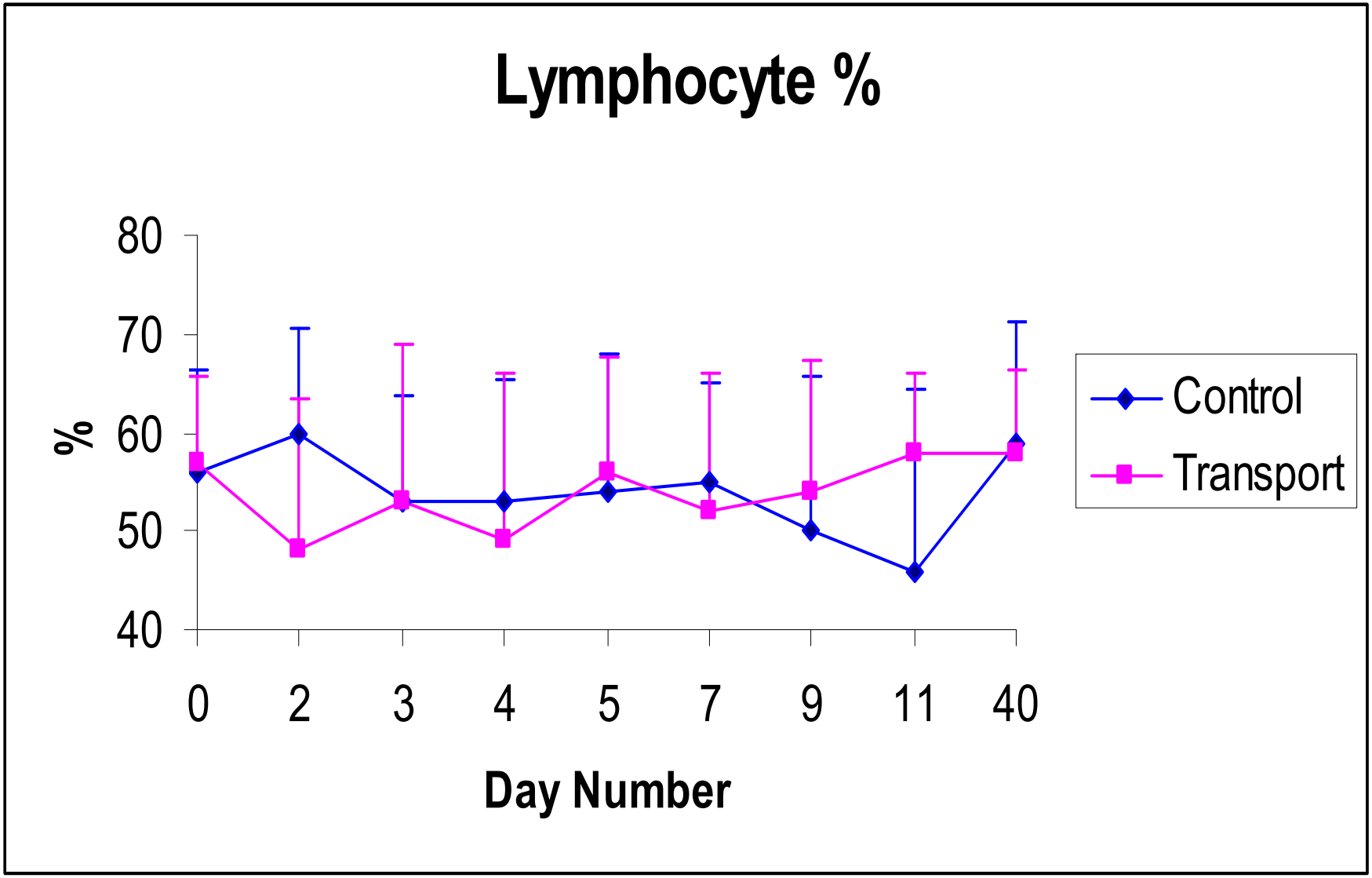


Ireland to Italy

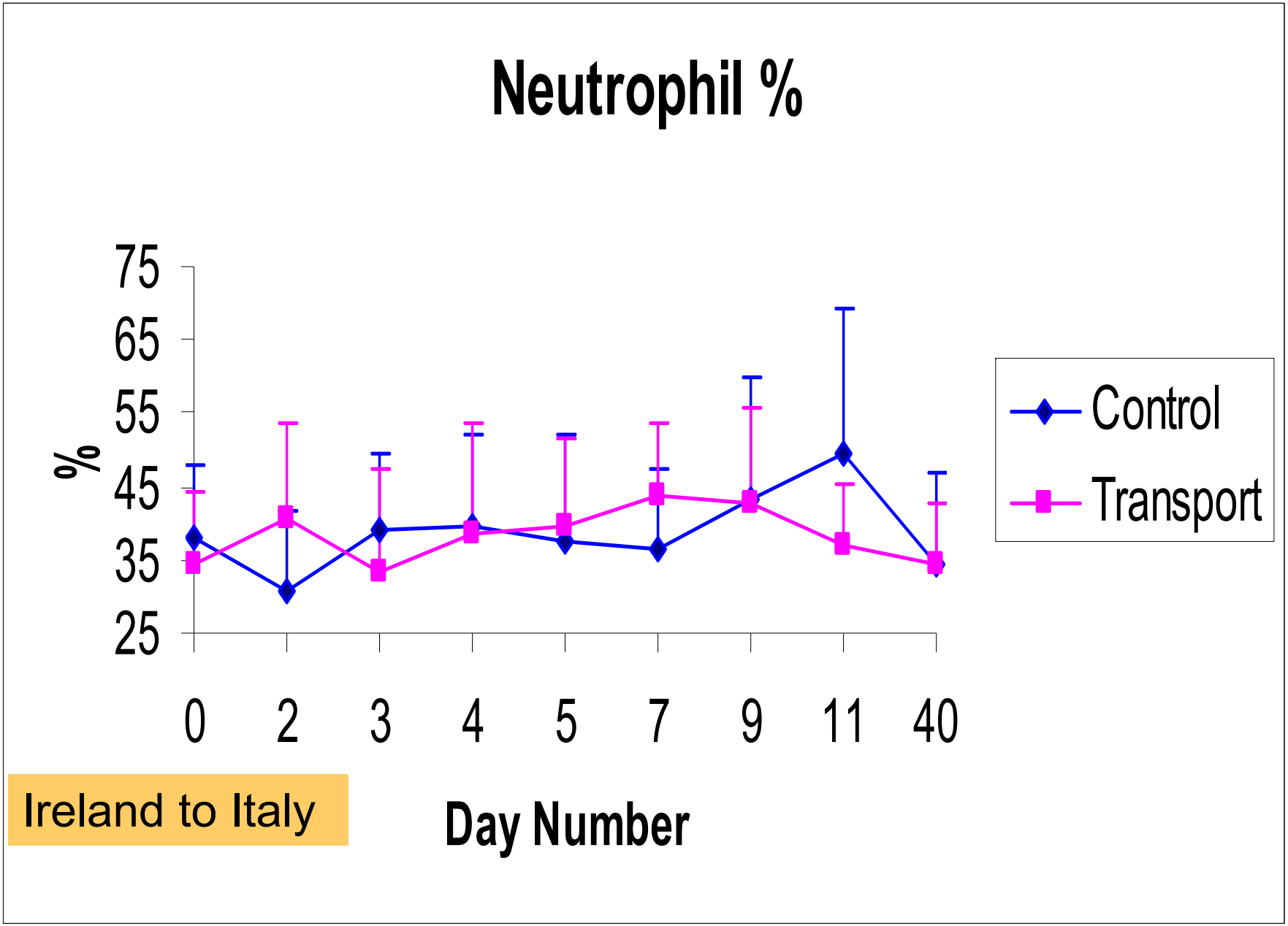
Median White Blood Cell (WBC) numbers



Ireland to Italy



Ireland to Italy


Neutrophil %
75
65
55
45
35
25
%
0
2
3
4
5
7
9
11
40
Day Number
Control
Transport
Ireland to Italy

Conclusions - welfare indices

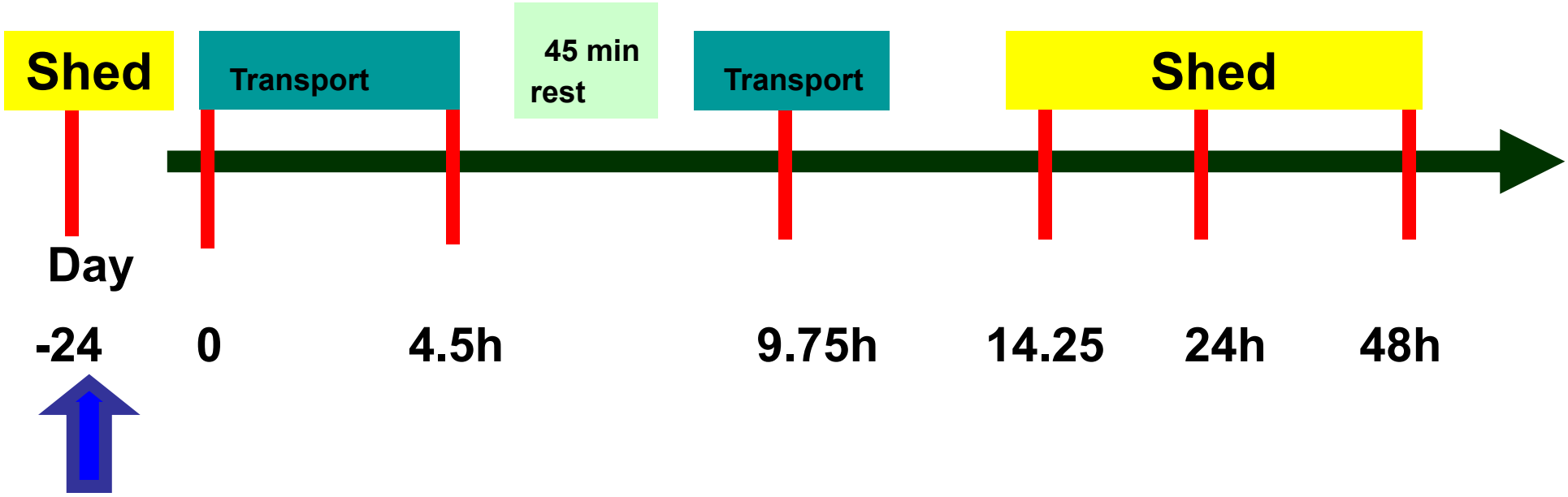
- Transient changes in physiological, haematological and immunological parameters were found in the transported and control animals, the levels that were measured were still within the normal physiological range* for the age and weight of animals studied

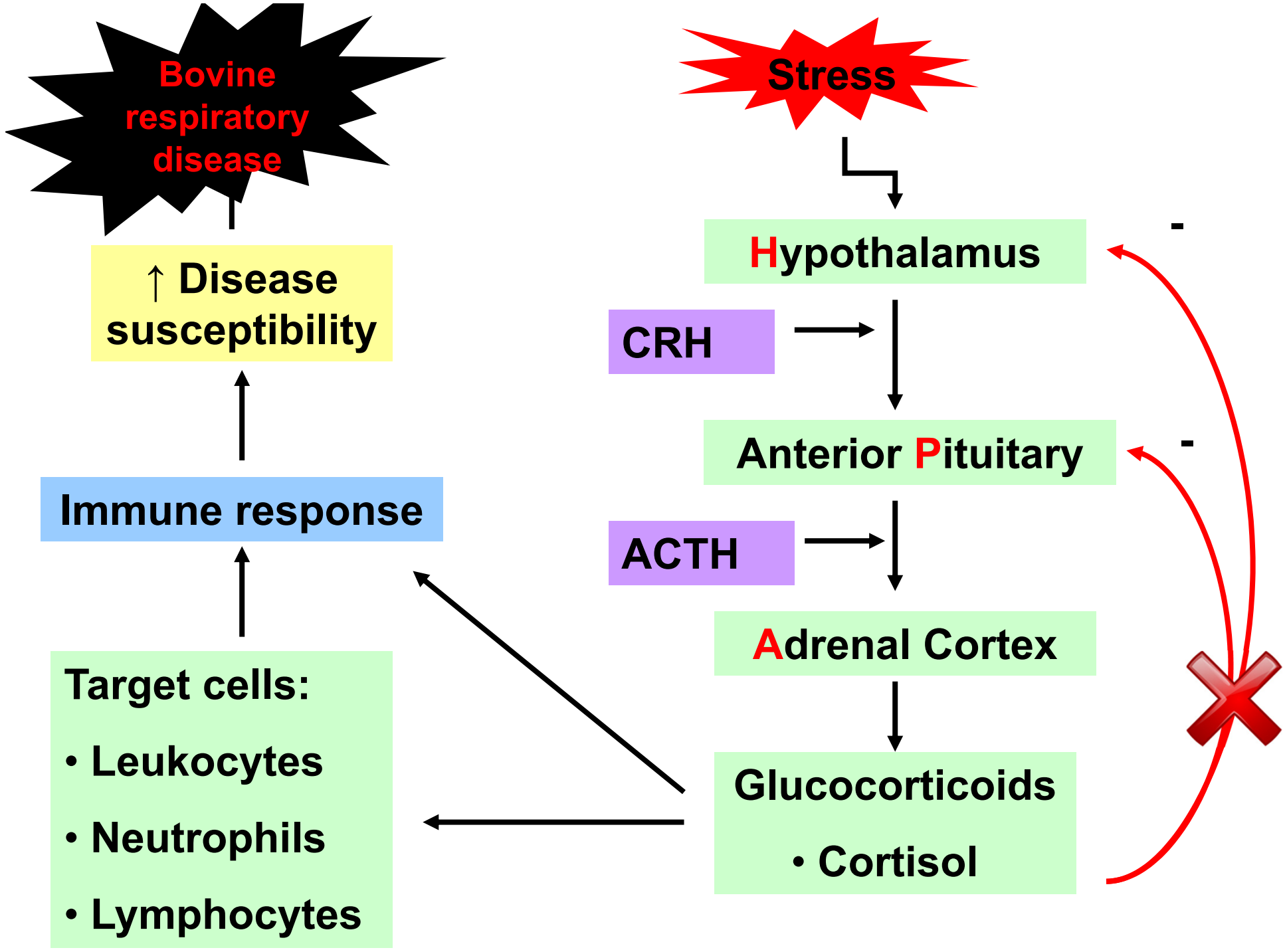
* Reference - Veterinary Laboratories Agency (VLA - UK)

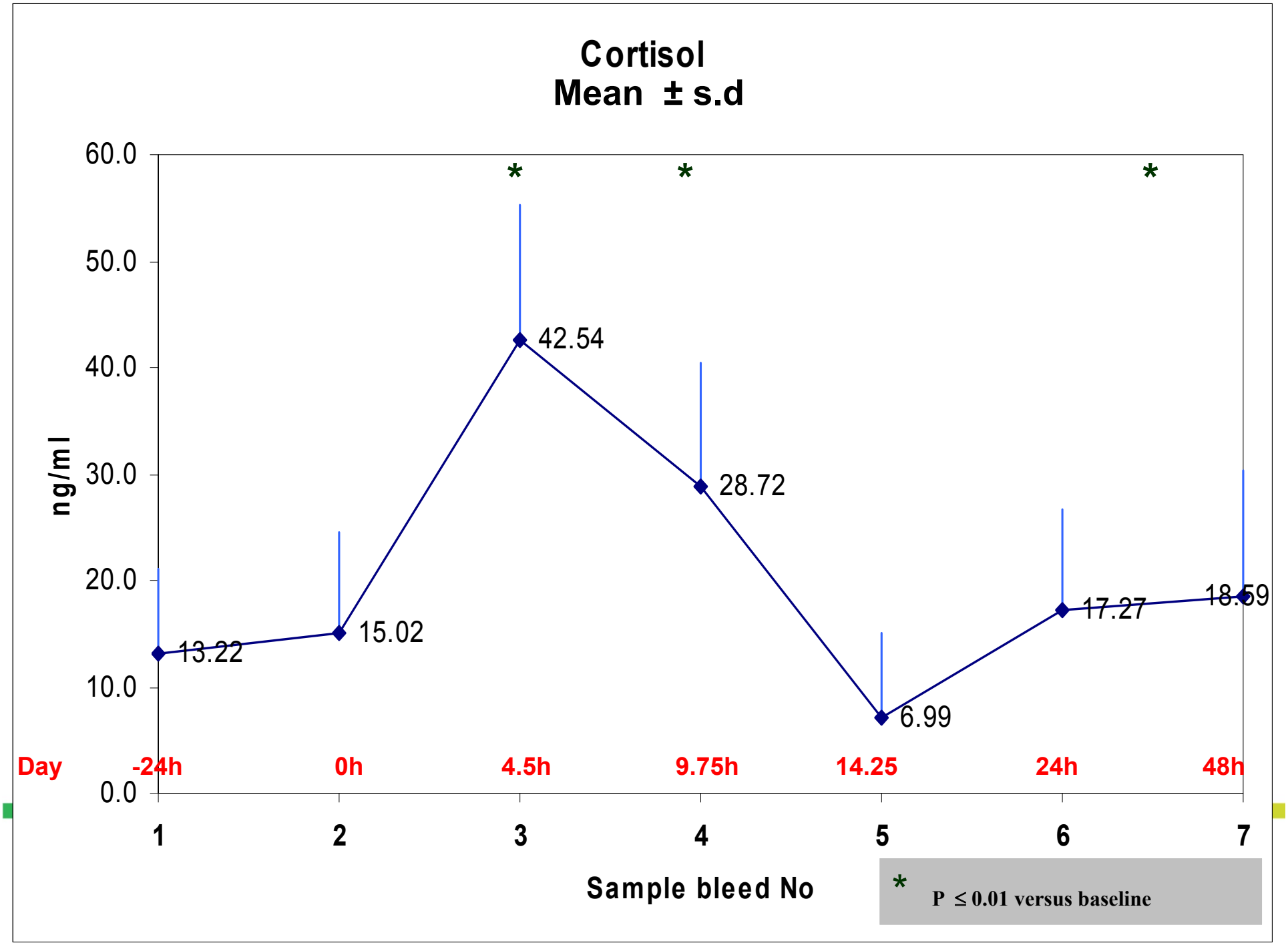
The effect of 9-hour transportation by road on gene expression changes in circulating neutrophils of bulls

Ireland (9 h journey)

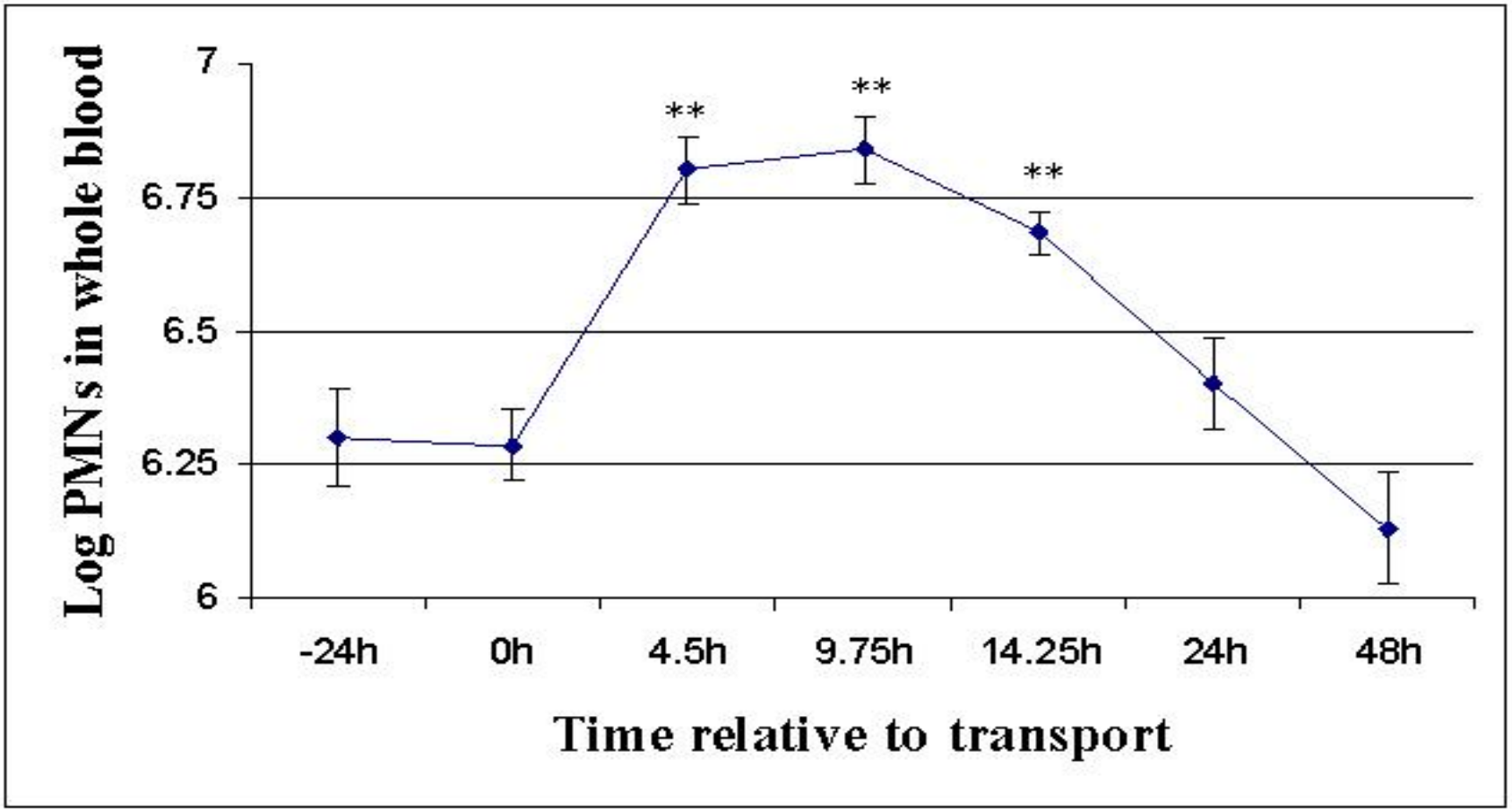
Thirty-six steers, (Belgian Blue x Friesian) approximately 9 months old and 250 kg body weight, were transported for 9 hours at a stocking density of approximately 0.80m^2 by road.



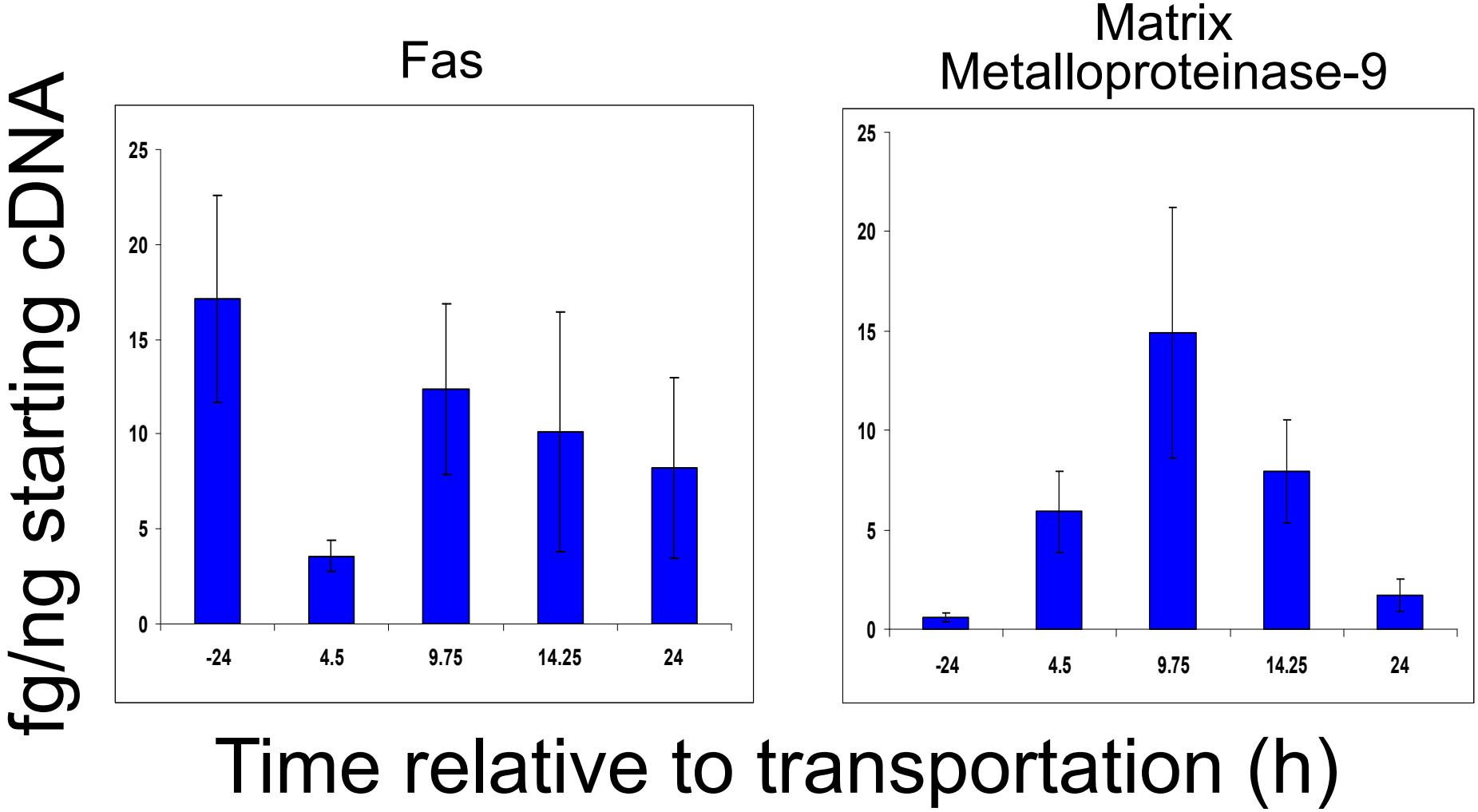

Bovine respiratory disease
↑ Disease susceptibility
Immune response
Target cells:
• Leukocytes
• Neutrophils
• Lymphocytes
Stress
Hypothalamus
CRH
Anterior Pituitary
ACTH
Adrenal Cortex
Glucocorticoids
• Cortisol
-
-


Cortisol
Mean ± s.d
60.0
50.0
40.0
30.0
20.0
10.0
0.0
ng/ml
*
*
*
42.54
28.72
13.22
15.02
6.99
17.27
18.59
Day
-24h
0h
4.5h
9.75h
14.25
24h
48h
1
2
3
4
5
6
7
Sample bleed No
* P ≤ 0.01 versus baseline

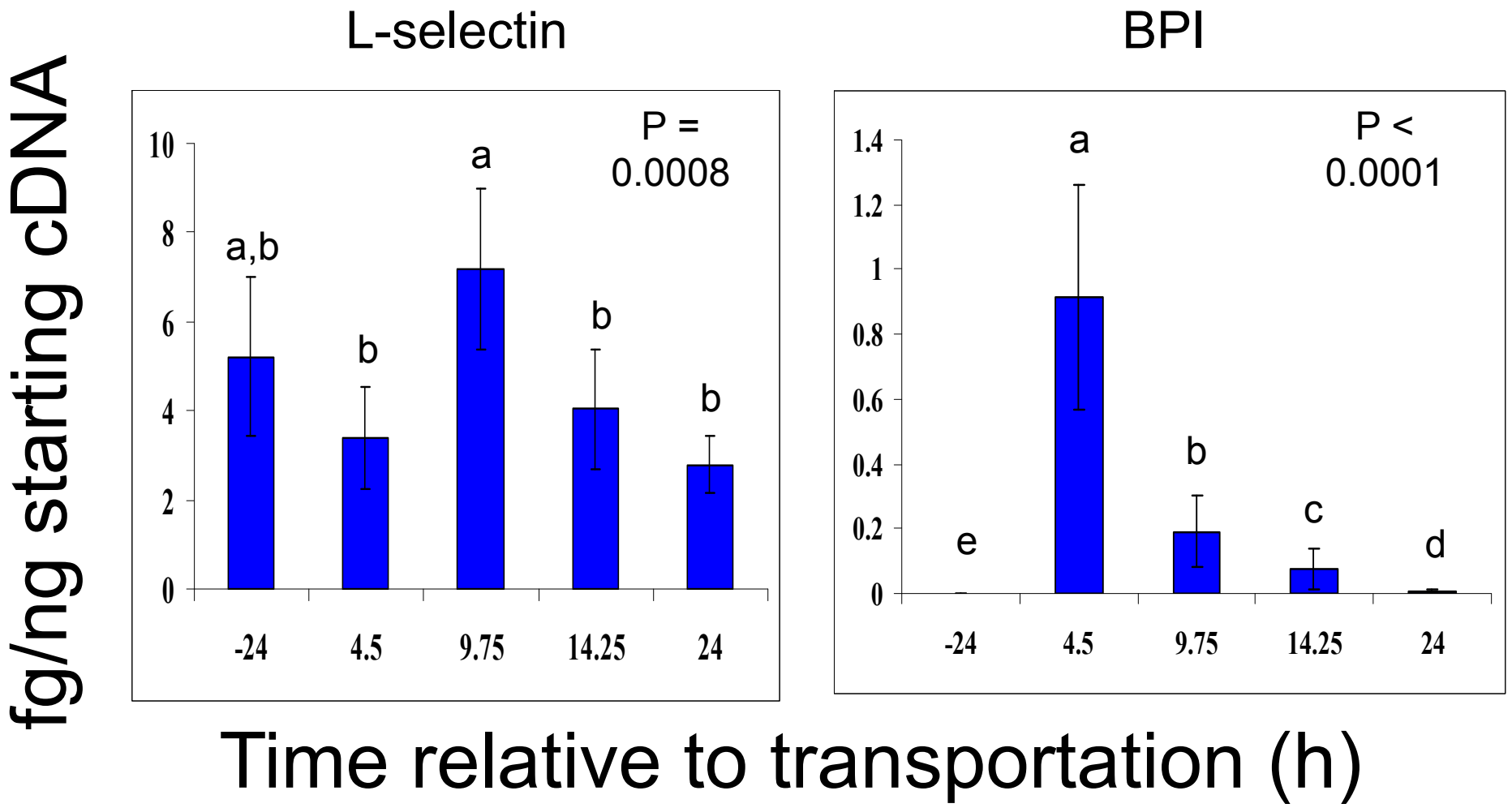
Absolute Whole Blood Neutrophil Counts in Transport Stressed Steers



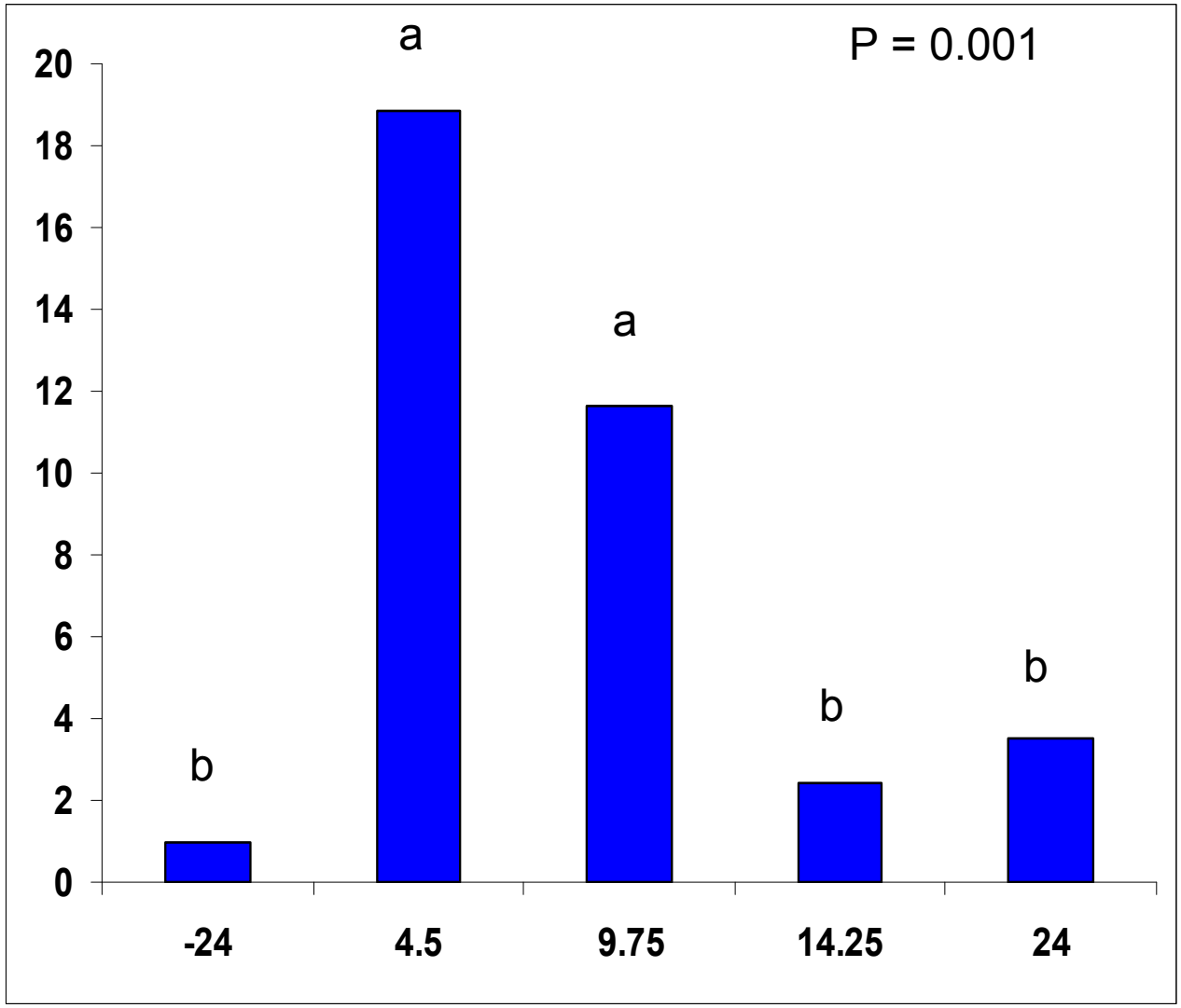
Pronounced changes in neutrophil gene expression at 4.5 and 9.75 h



Pronounced changes in neutrophil gene expression at 4.5 and 9.75 h

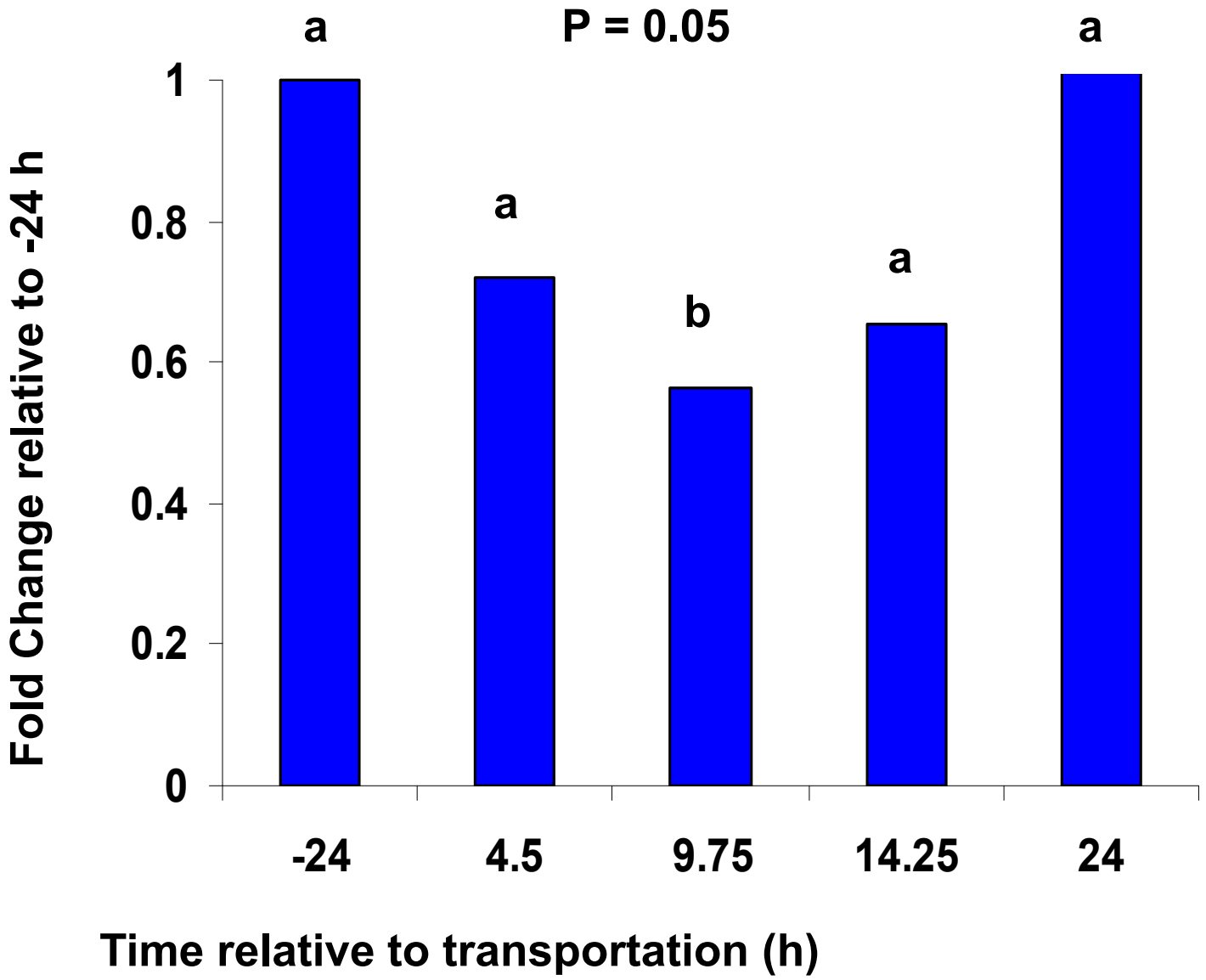


Representative immune function gene: IL-8 precursor



- **Potent neutrophil chemoattractant and activator**
- **Associated with neutrophil infiltration and tissue damage in rheumatoid arthritis and lung reperfusion syndrome**
- **(Harada et al., 1994)**

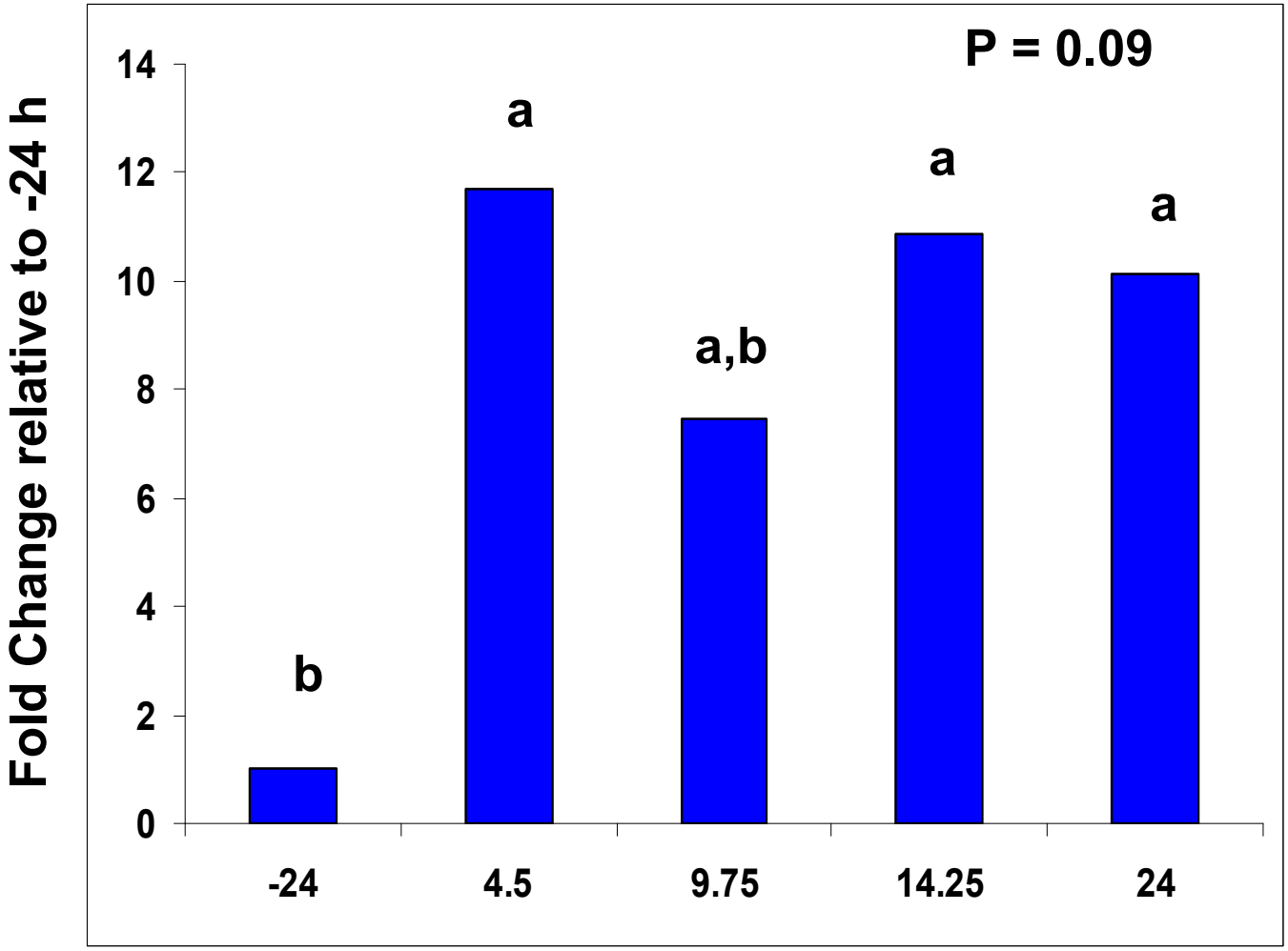
Representative apoptosis gene: Caspase-13



- Member of proteases involved in apoptosis cascade
- Activated by caspase-8

(Humke et al., 1998)

Representative wound healing gene: Peroxisome Proliferator Activated Receptor γ



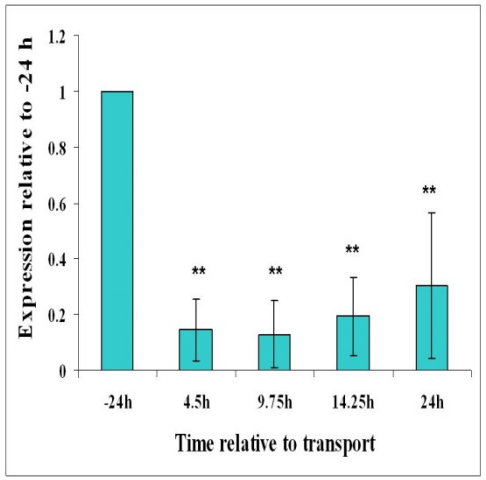
- Transcriptional regulator of many biological processes
- Anti-apoptotic effects on neutrophils; involved in wound healing

(Standiford et al., 2005)

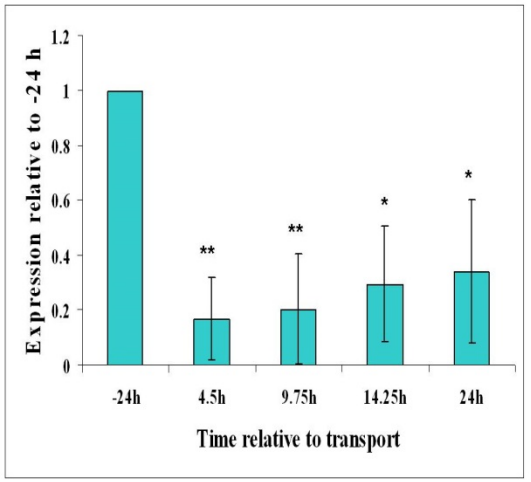
Pro-apoptotic

death inducing

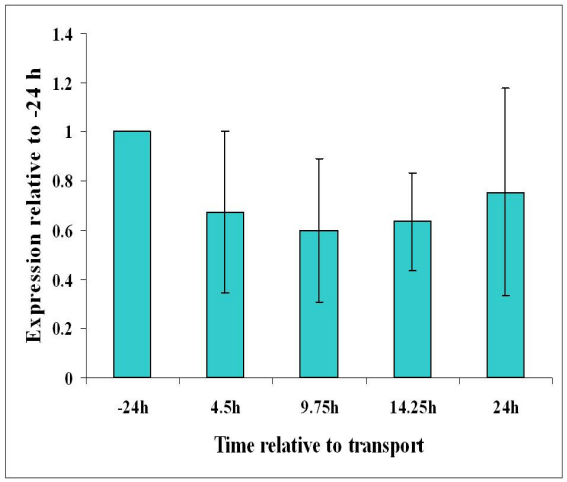
Caspase 13



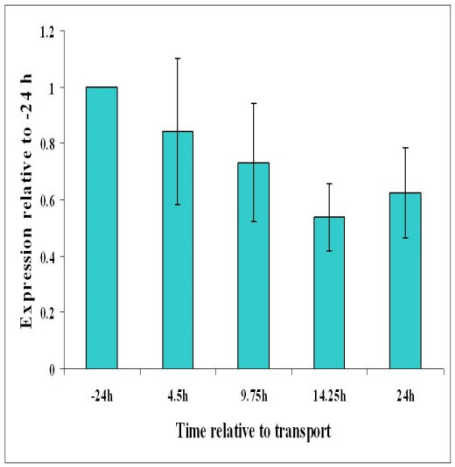
Traf 6



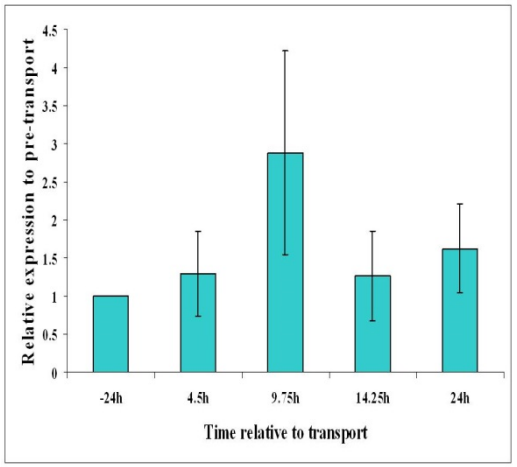
DAPK



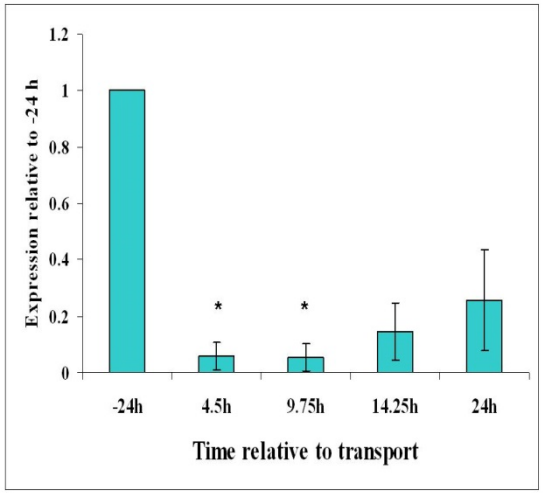
Mcl-1



Gro gamma



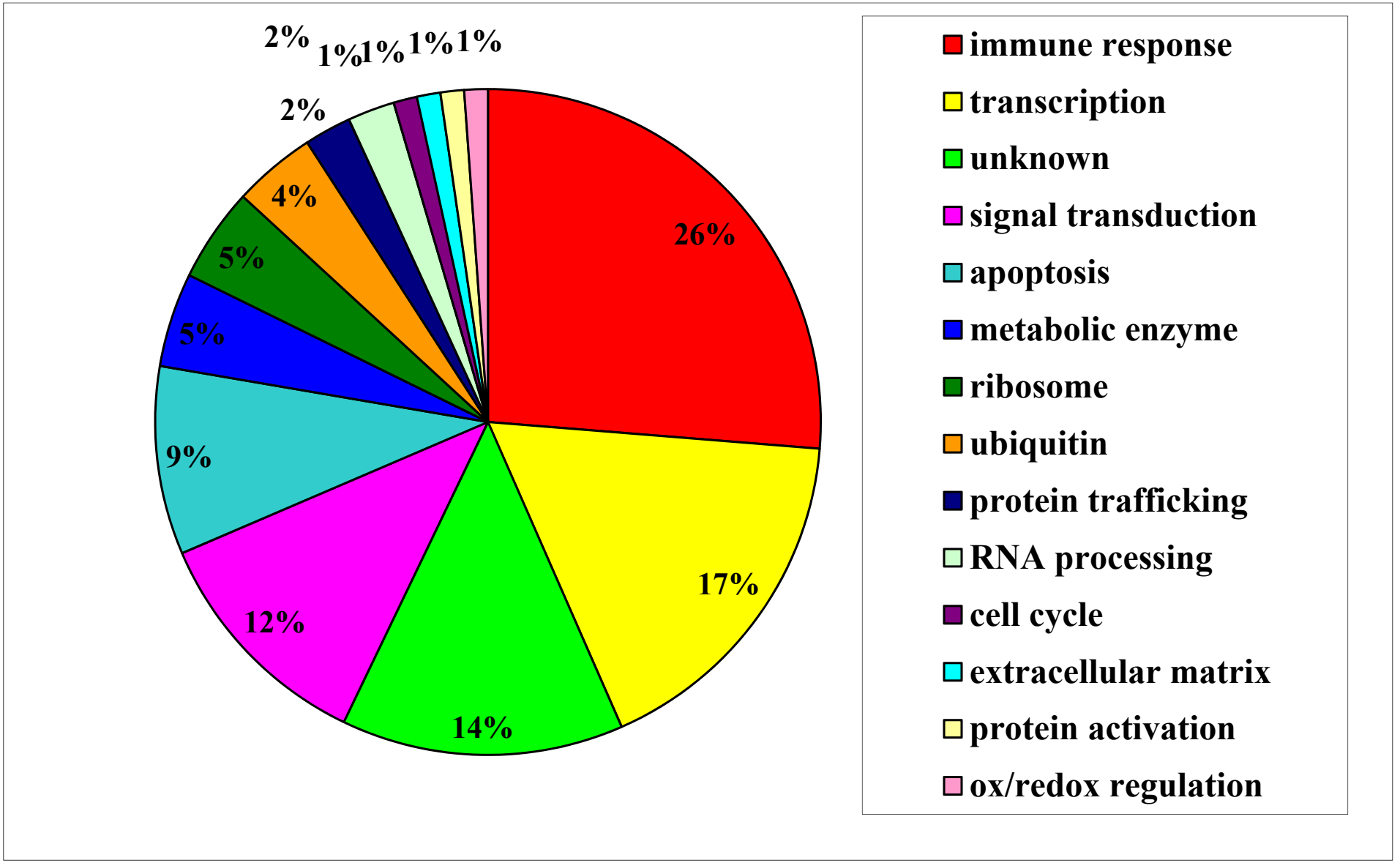
TGFR-3



Anti-apoptotic

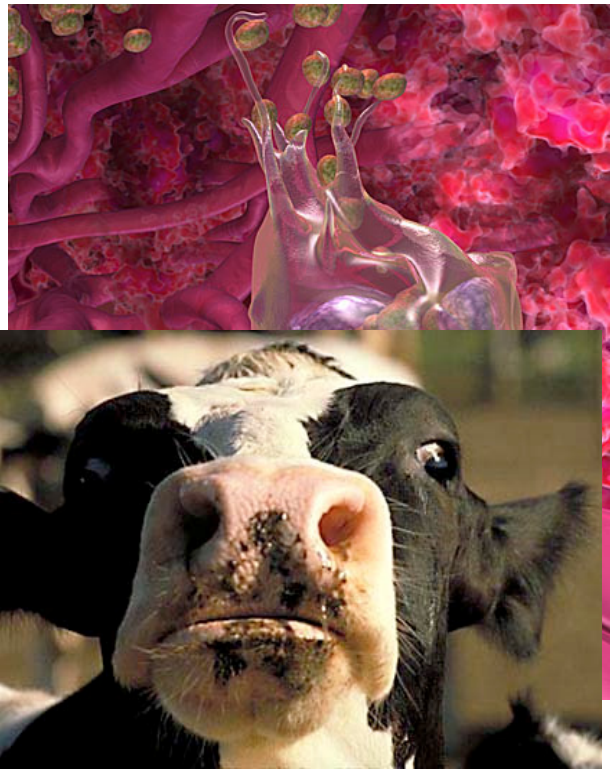
Neutrophil chemotractant

Betaglycan



Ontological clusters of genes affected by transport stress. Genes in the largest cluster, “immune response,” represent those involved in activation, chemotaxis, cell adhesion, bactericidal activities, and wound healing functions of neutrophils.

Conclusions



← Increased neutrophil activation and recruitment

← Decreased neutrophil apoptosis + increased circulating neutrophils

← Increased wound healing and tissue breakdown by neutrophils

Increased inflammatory potential

Protect against respiratory disease

The effect of road transport and a 12 hour mid-journey rest period on the physiological responses of bulls

There is limited scientific data on the physiological recovery of bulls after long durations of transport and in particular when given access to a 12 hour (h) mid-journey rest period during transport.

Transport conditions have the potential to alter the biological responses of animals.

Study objective

To investigate effect of a **9 h road transport** followed by a **12 h** mid-journey **rest** period and a subsequent **9 h road transport** on the welfare of bulls as assessed through physiological concentrations of blood variables.

Animals

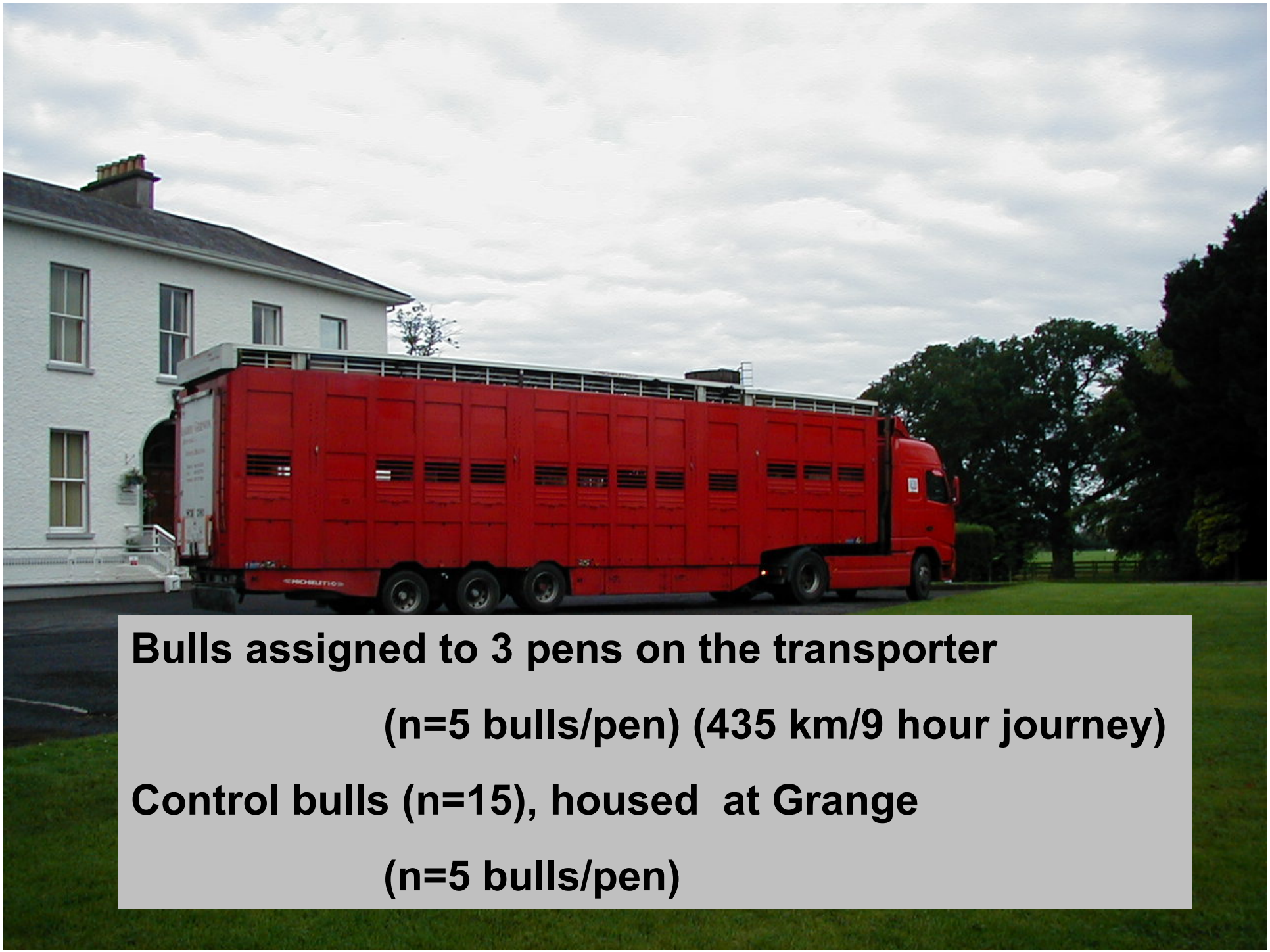
Thirty **bulls**, continental breed crosses from crossbred suckler dams:

mean live weight **486**, s.d. 57.0 **kg**

Assigned by live weight to one of two treatments:

Transport (T) (n=15)

Control (C) (n=15)

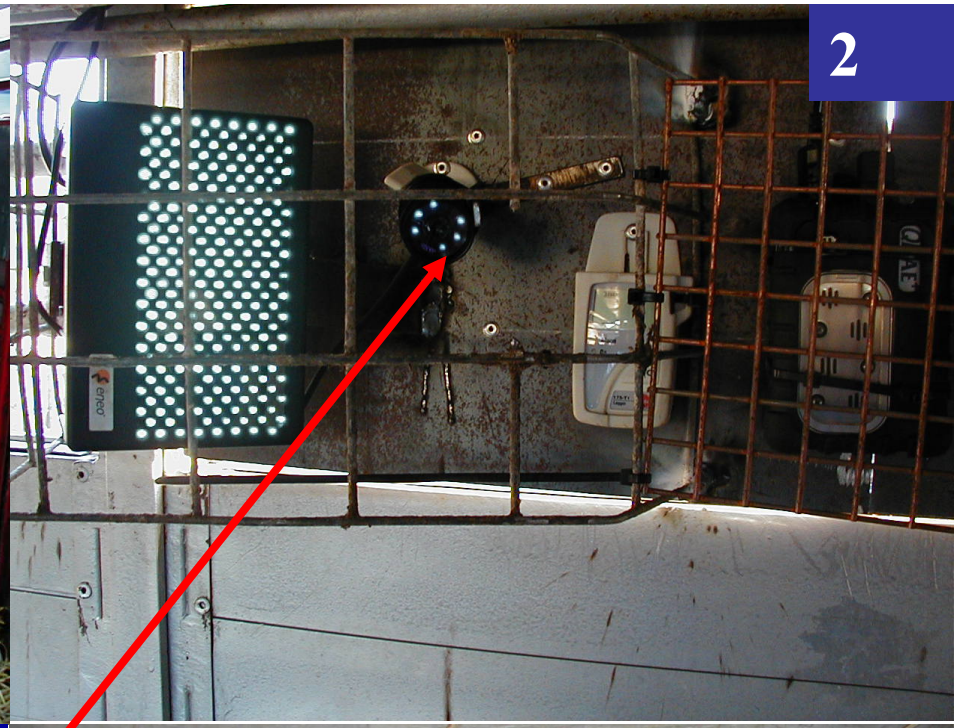


Bulls assigned to 3 pens on the transporter

(n=5 bulls/pen) (435 km/9 hour journey)

Control bulls (n=15), housed at Grange

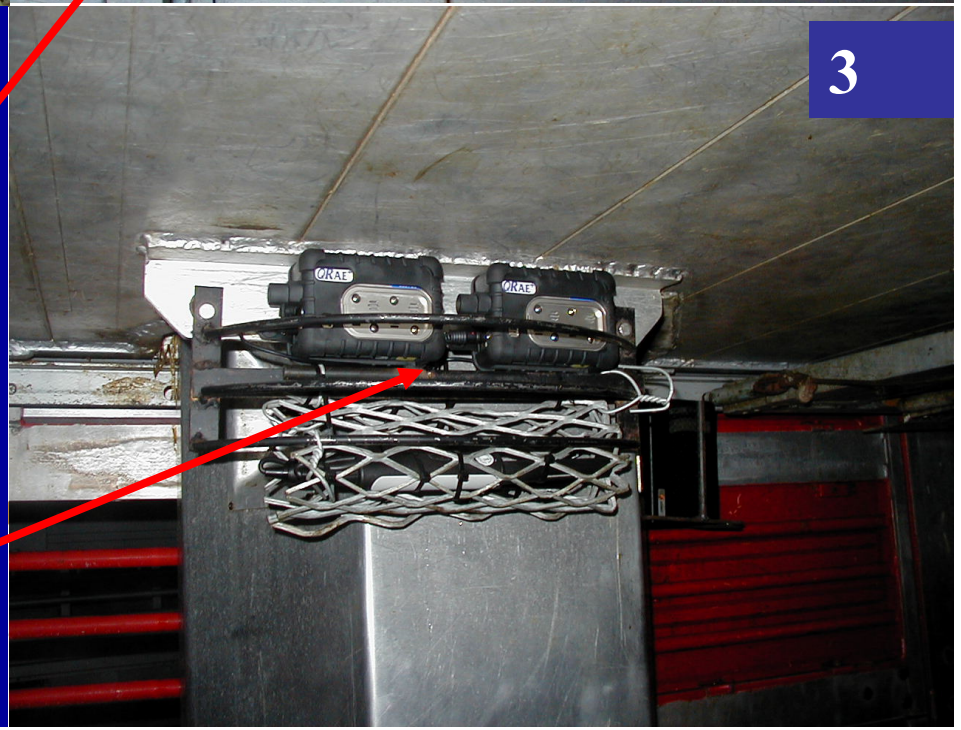
(n=5 bulls/pen)



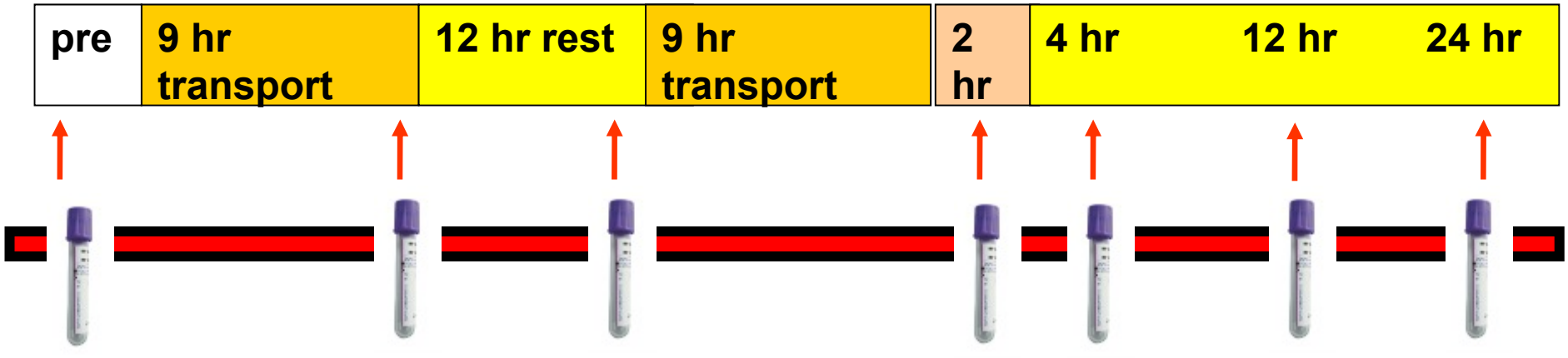
1. Lower Deck of Transporter

2. Lower deck of Transporter showing the CCTV camera

3. Environmental loggers



Transport by road



Bulls - blood sampled (jugular venipuncture)

Haematological variables neutrophil, lymphocyte, a haematology analyser)

Metabolites (Creatine kinase, Glucose, ßHB)

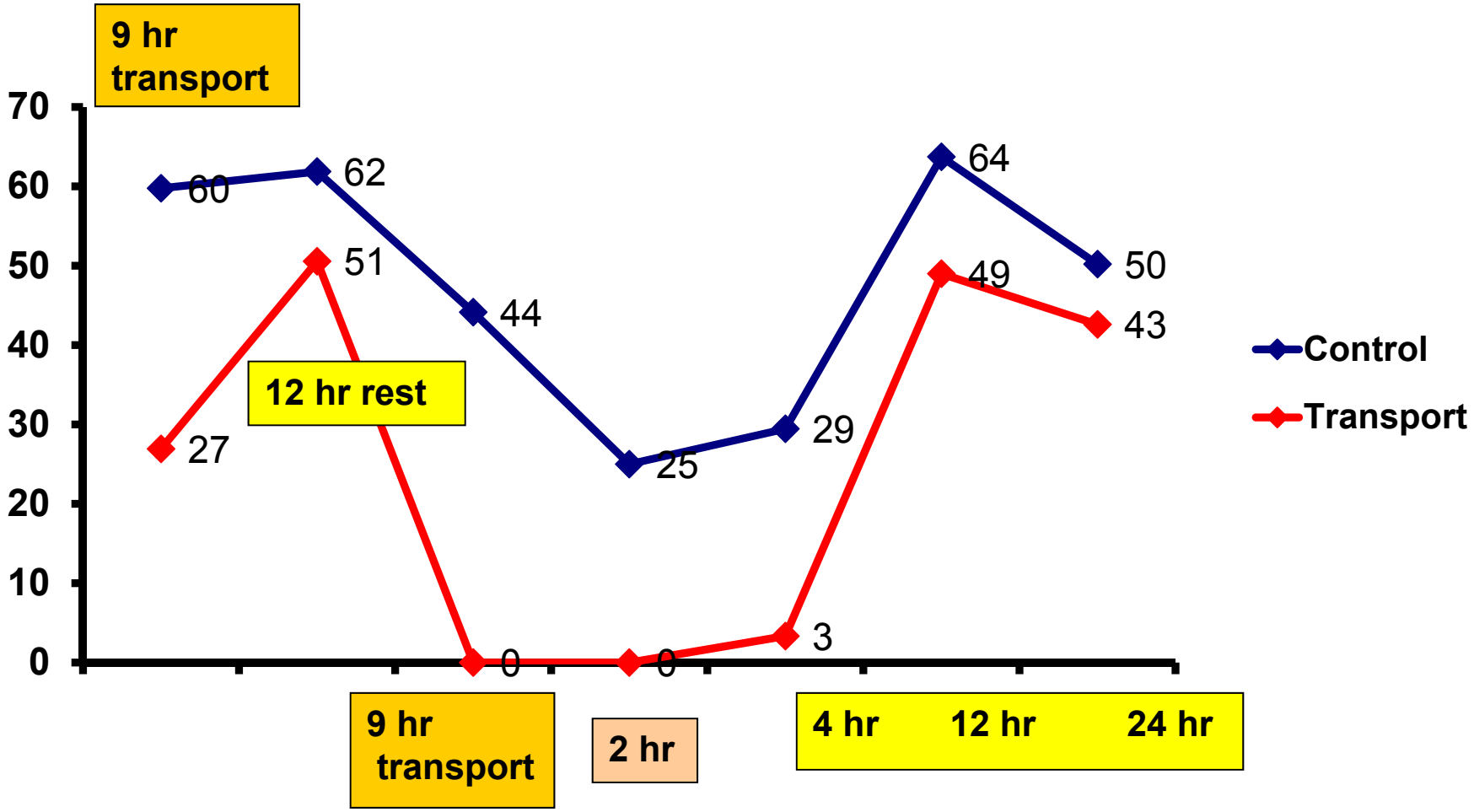
Environmental conditions during transport study

Transport phase	CO_2 ppm	Temp °C	Wind speed m/s	Vapour density td °C	NH_3 ppm
1 st 9 h Journey	475	10.7	1.0	9.5	0.62
12 h rest (Shed)	602	8.4	0.0	8.3	0.39
2 nd 9 h Journey	826	12.2	0.6	9.4	0.95
2 h rest on transporter	749	14.2	0.1	8.6	0.17

Live weight loss %

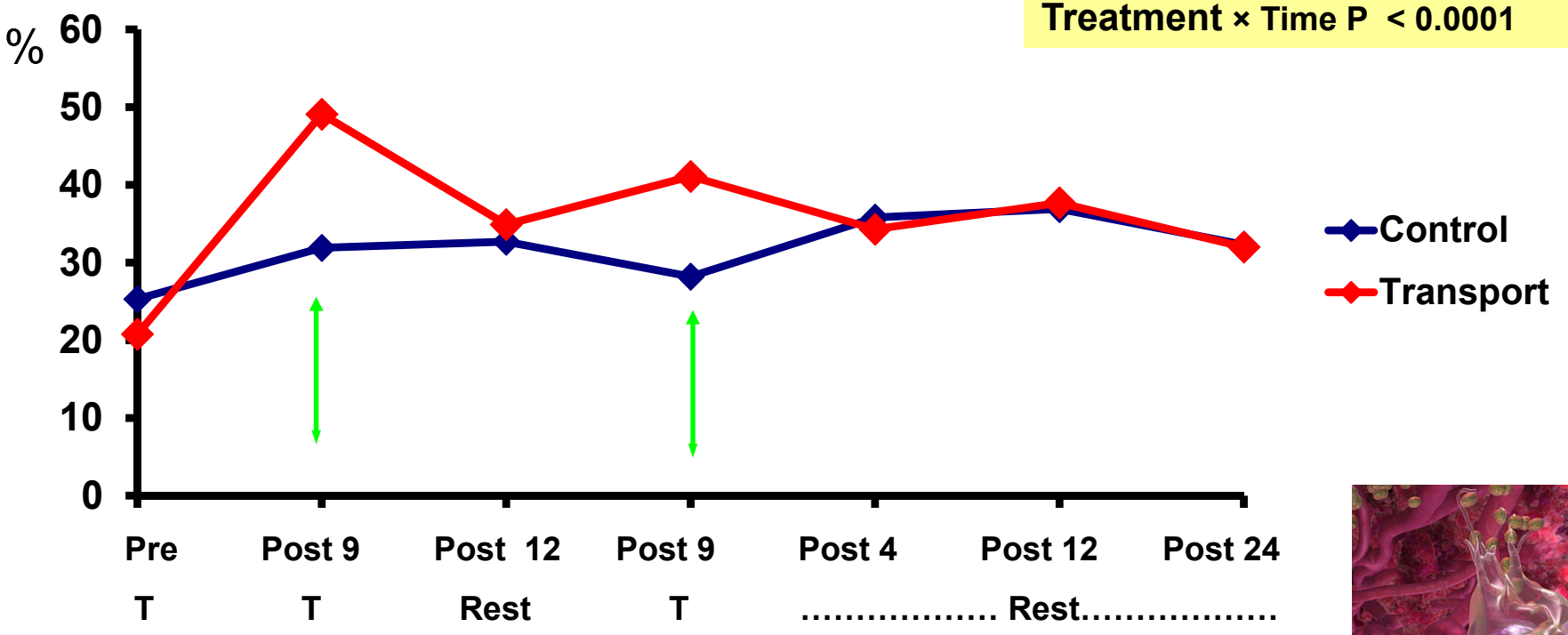
	9 Transport	12 Rest	9 Transport	Start to Finish
Transport	-6.2 %	+ 3.1 %	- 4.0 %	[Overall 4.0%]
Control	-2.7 %	+1.2 %	-1.1 %	[Overall 1.6%]

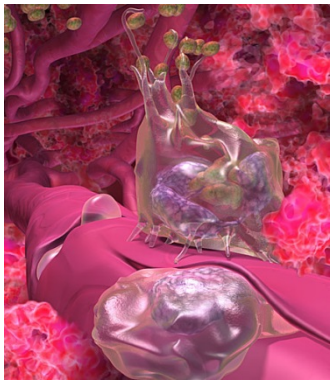
Percentage time lying



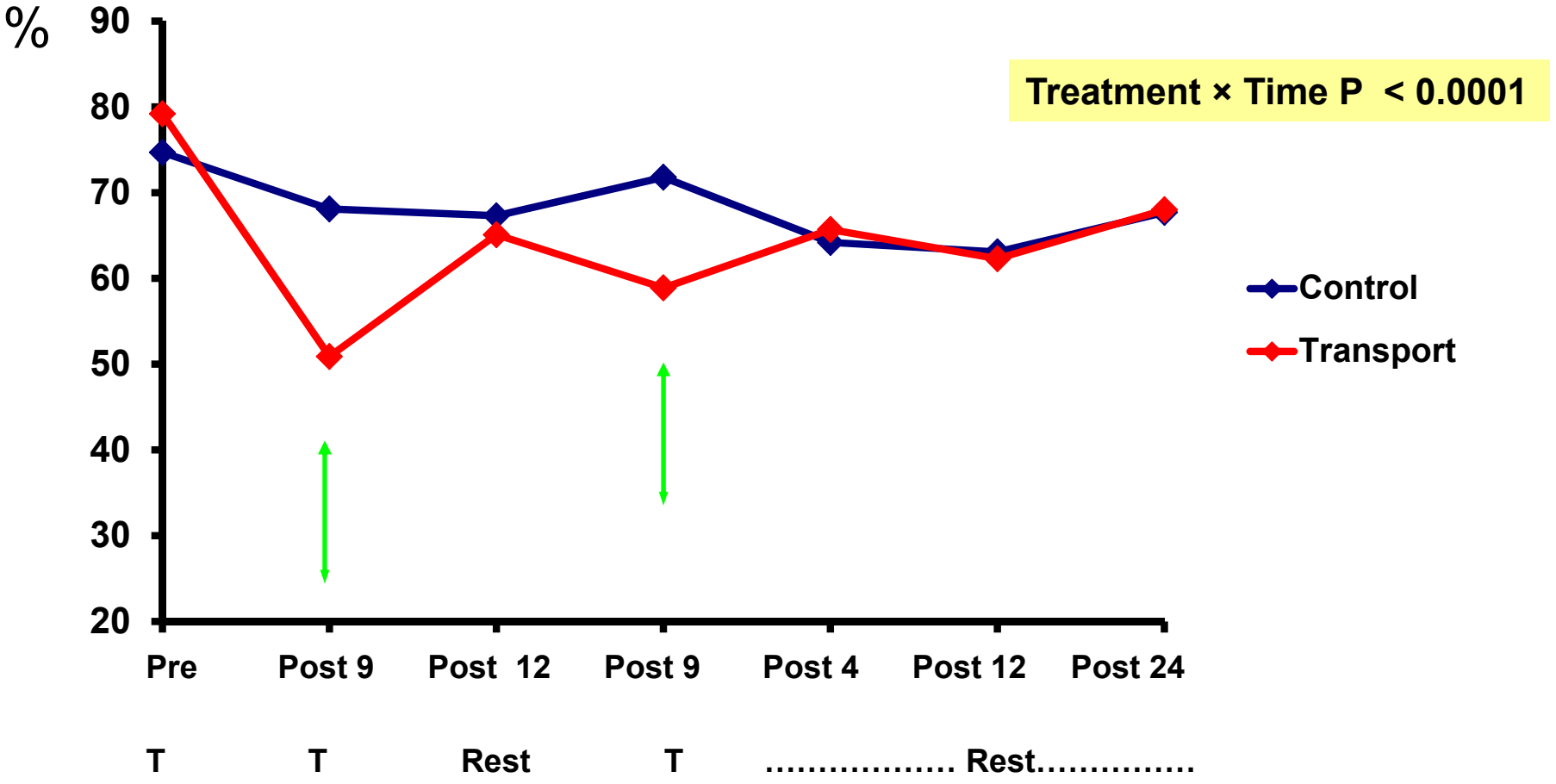
Neutrophil %

Treatment × Time P < 0.0001



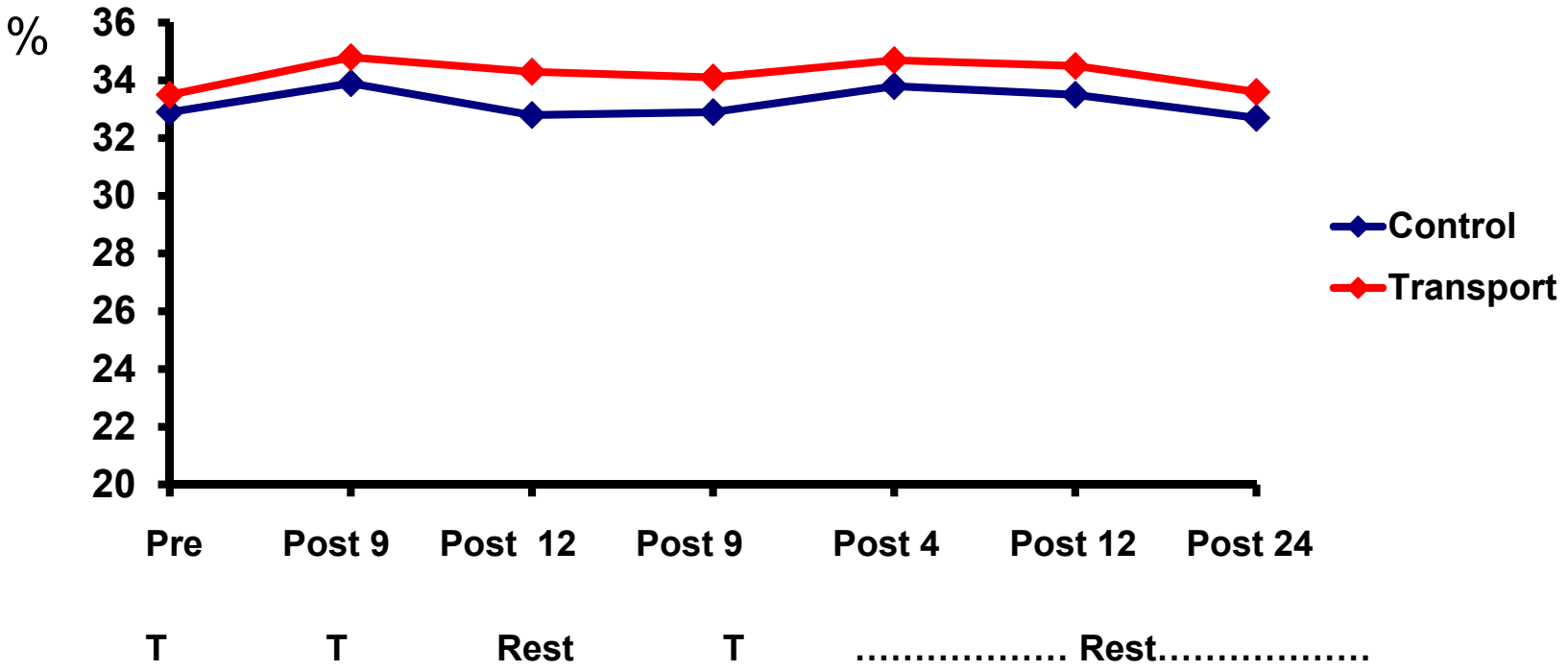


Lymphocyte %

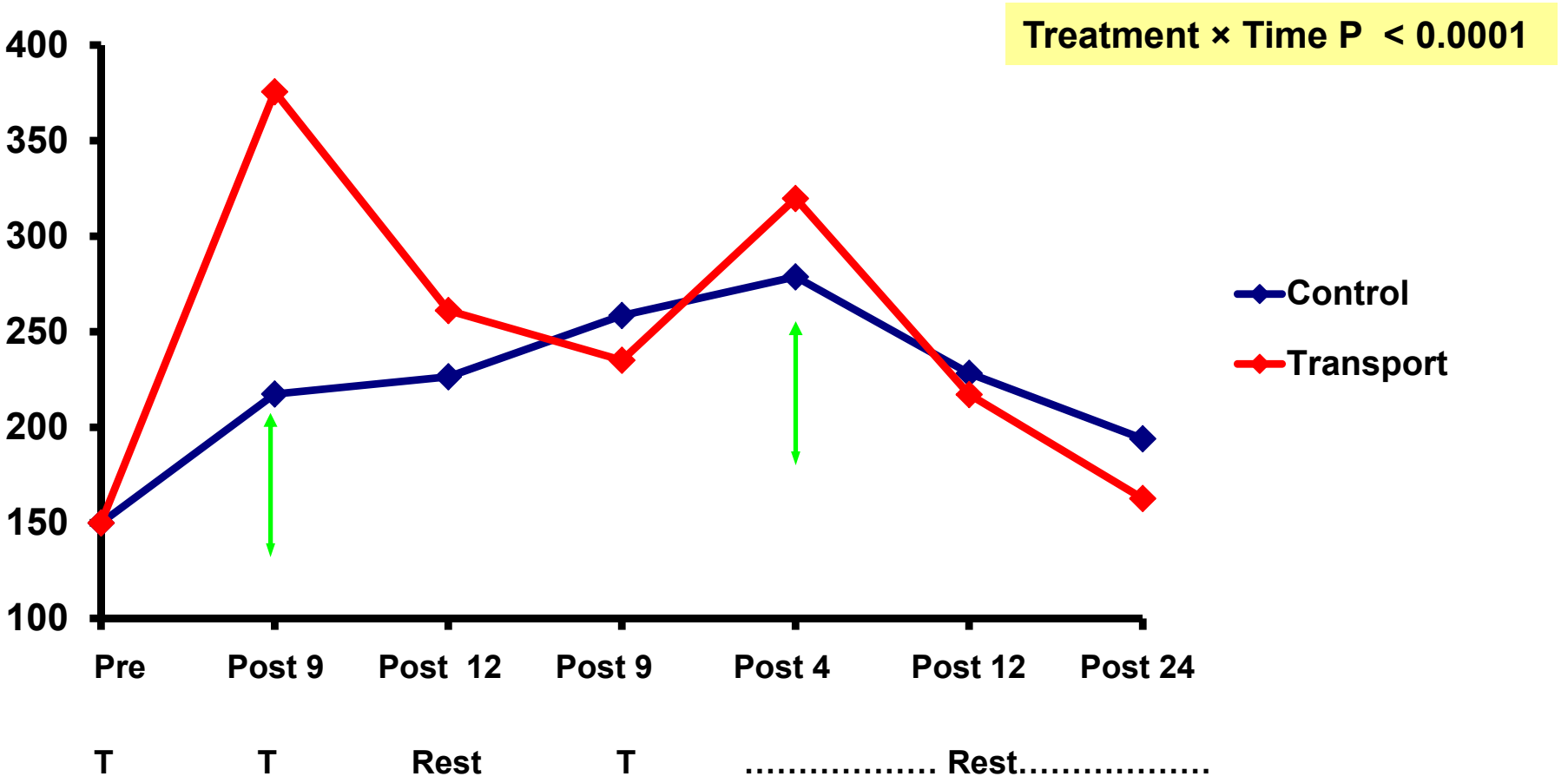

%
90
80
70
60
50
40
30
20
Treatment × Time P < 0.0001
Control
Transport
Pre
Post 9
Post 12
Post 9
Post 4
Post 12
Post 24
T
T
Rest
T
..... Rest.....

Haematocrit %

Treatment × Time P > 0.05



Creatine Kinase activity (IU)



Conclusions

Control bulls showed similar changes in physiological responses to transported bulls.

Twenty four hours in recovery with access to feed and water was sufficient for many of the blood variables to recover to pre-transport concentrations.

The effect of pre-transport periods (0, 12 and 24 h) of food and water deprivation on the response to 12 and 24 h transport in yearling heifers

Teagasc and CSIRO Australia
collaborative study

Teagasc
Agriculture and Food Development Authority

CSIRO
Livestock Industries

Introduction

The combination of 8 h of fasting (feed) and 8 h of transport - no negative impact on animal welfare

Longer duration of pre-transport food and water deprivation ?

Australia - Transport

Objective

To quantify the effect of pre-transport food and water deprivation on the response to transport in cattle.

84 *Bos Taurus* beef breed yearling heifers (262 ± (s.e.) 12.4 kg)

0 h Restricted (A) + 12 h Transport (T)

12 h Restricted (A) + 12 h Transport (T)

24 h Restricted (A) + 12 h Transport (T)

0 h Restricted (A) + 24 h Transport (T)

12 h Restricted (A) + 24 h Transport (T)

24 h Restricted (A) + 24 h Transport (T)

Jugular blood & urine samples - (6 time points)
prior to, on completion of the restricted period and 0, 24, 48 and 72 h post-transport.

Study - repeated twice – over a 4 week period

Transport duration (distance)

12 h (960 km)

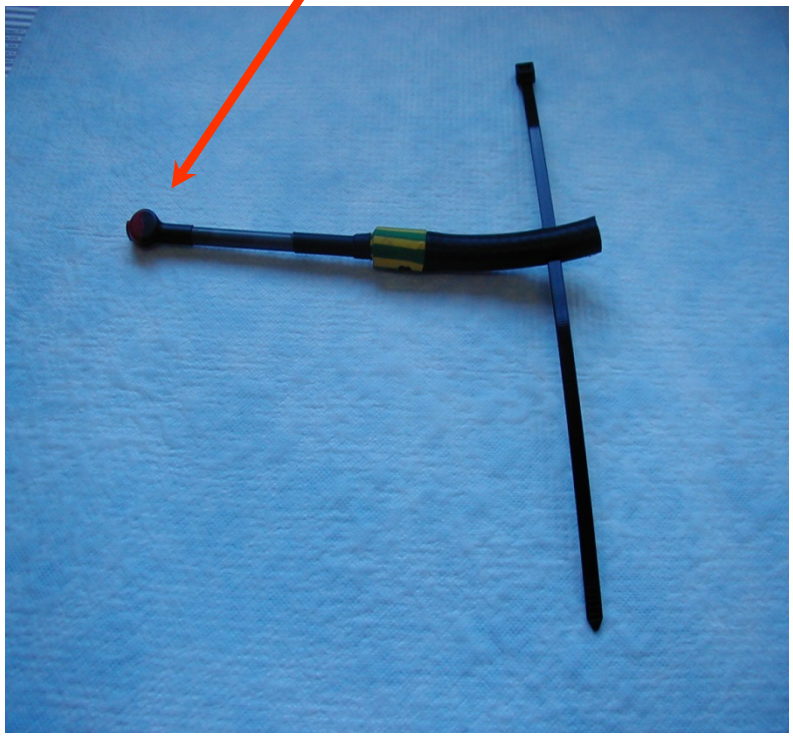
24 h (1920 km)

Spatial allowance of 0.82 m^2/head

An animal fitted with IceTag logger

- **Lying and standing behaviour**

An animal fitted with rectal temperature logger (Thermochron iButton)



Physiological Measurements

Haematology

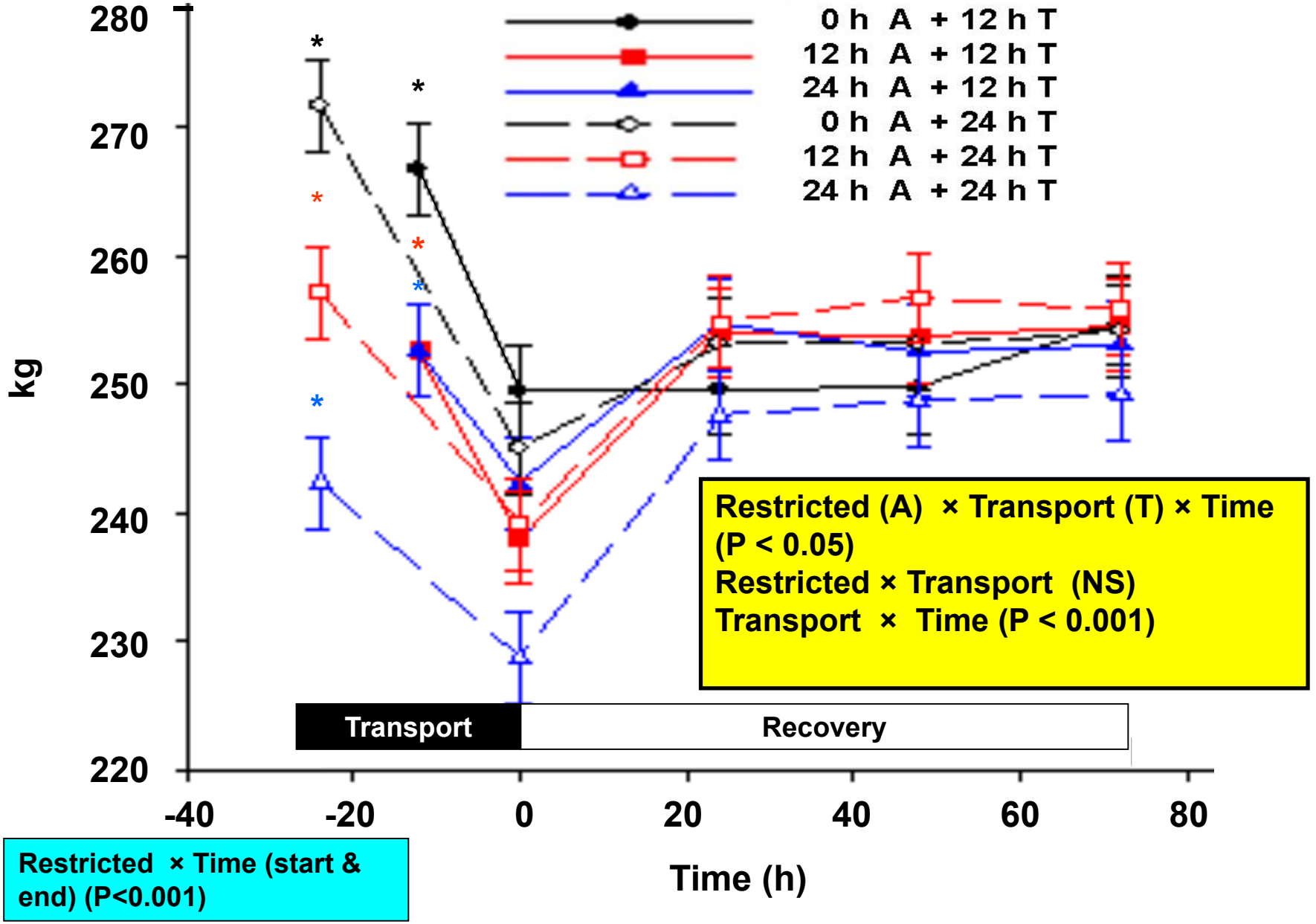
Red blood cells (RBC) and white blood cells (WBC) count, differential cell counts of neutrophils (NEU) and lymphocytes (LYM) and haematocrit (HCT) %.

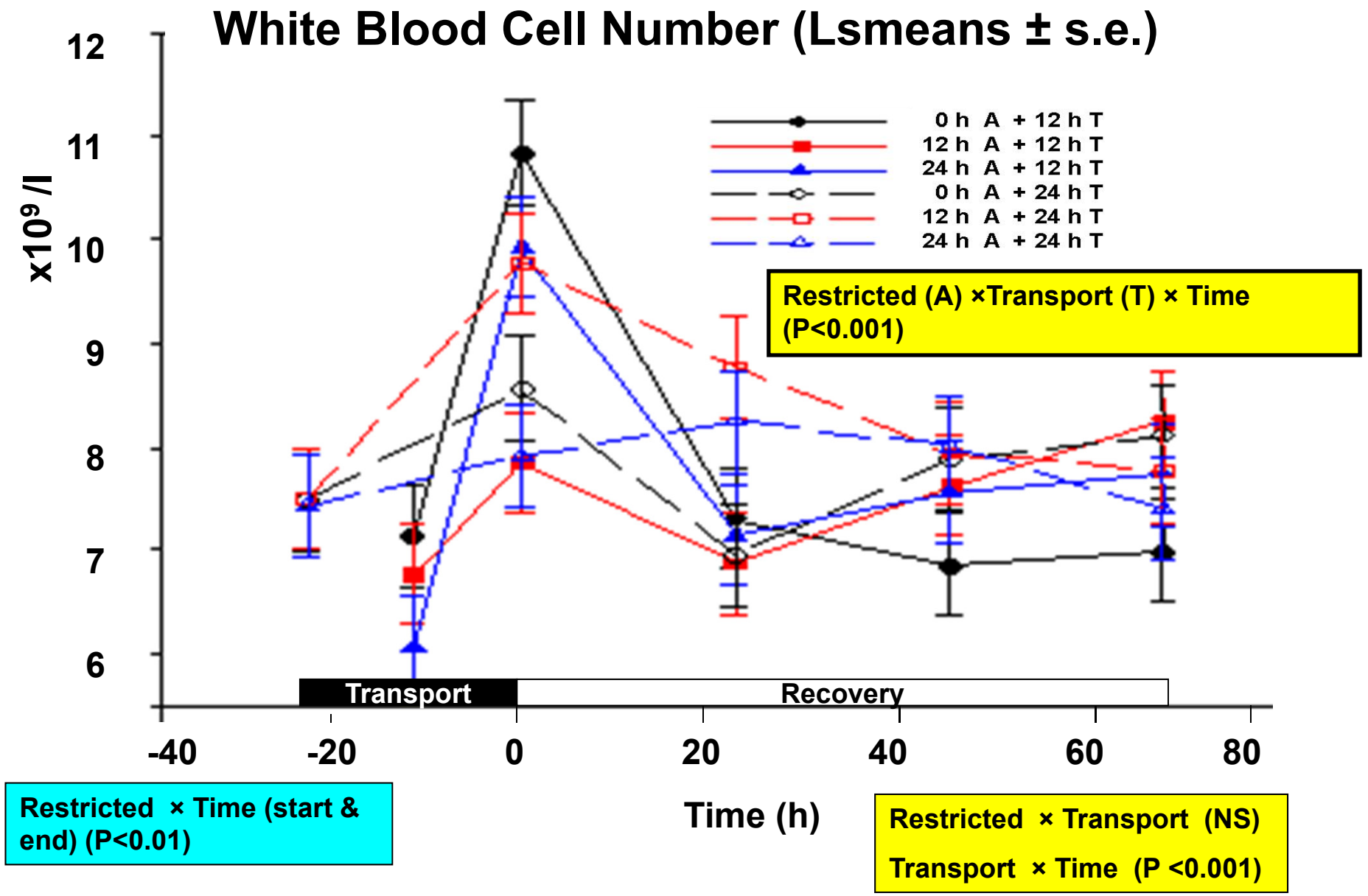
Serum concentrations of cortisol, total protein, albumin, creatine kinase (CK), blood urea nitrogen (BUN)

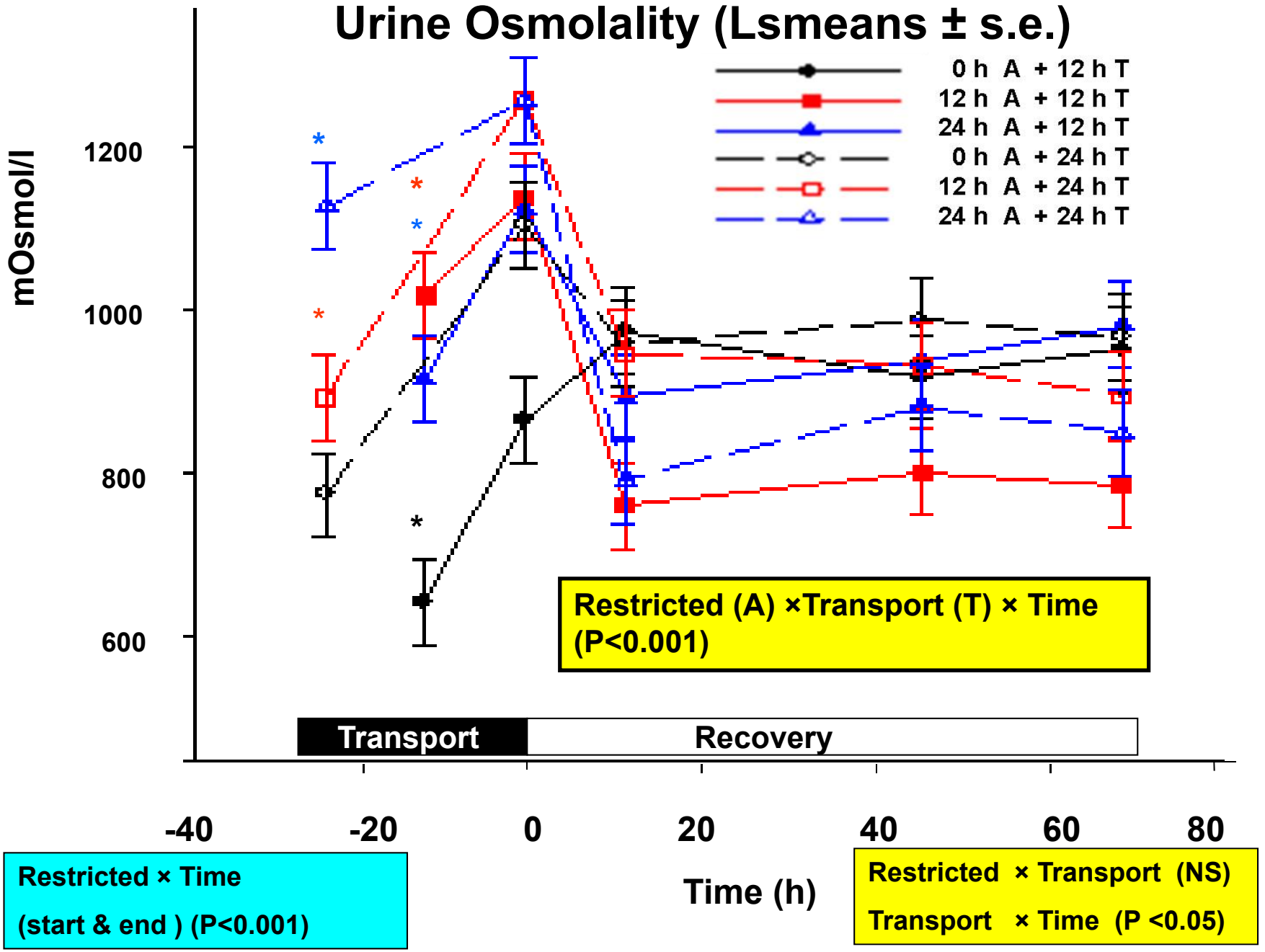
Urine - specific gravity & osmolality

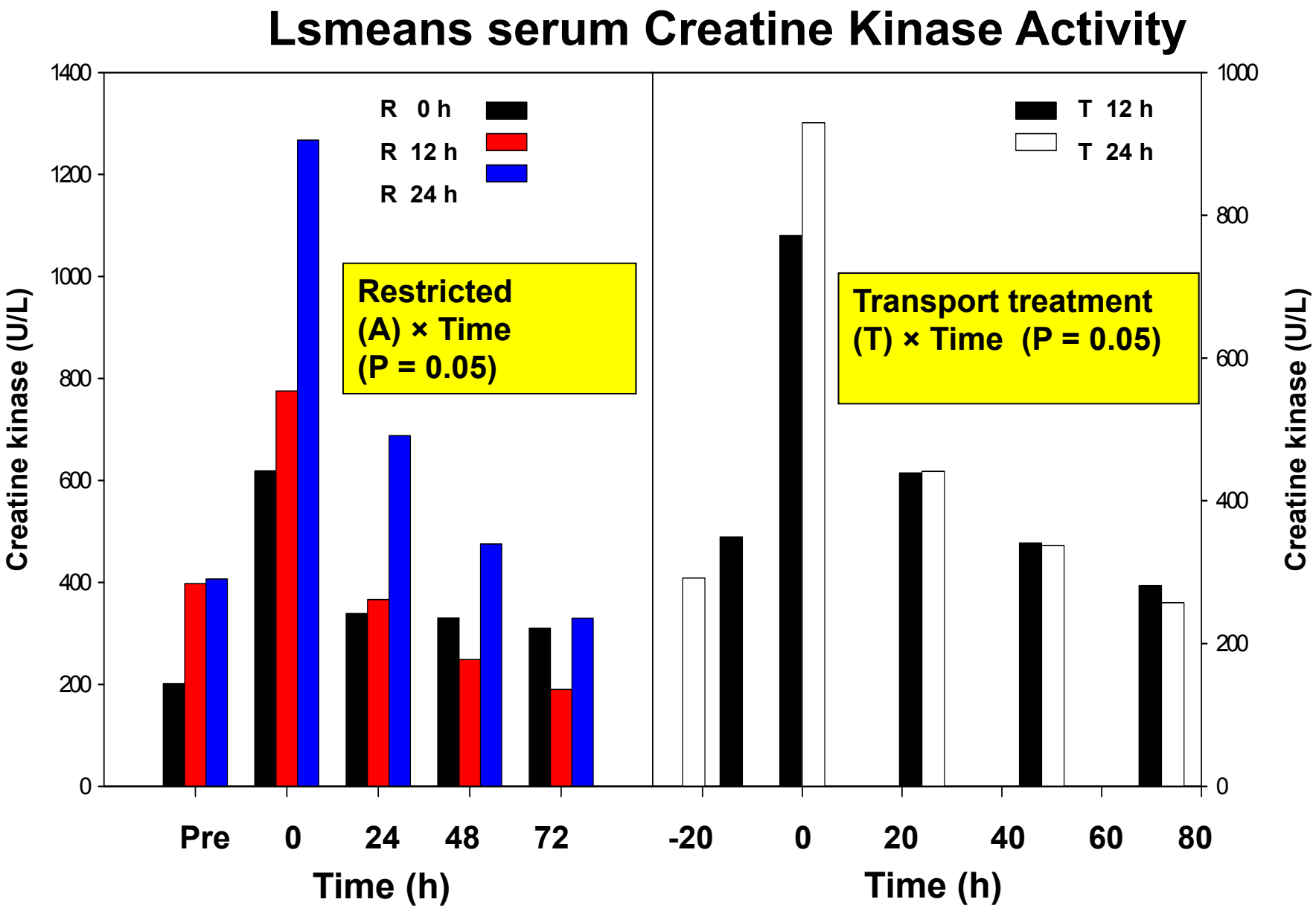
Water intake – flow meters – recovery period

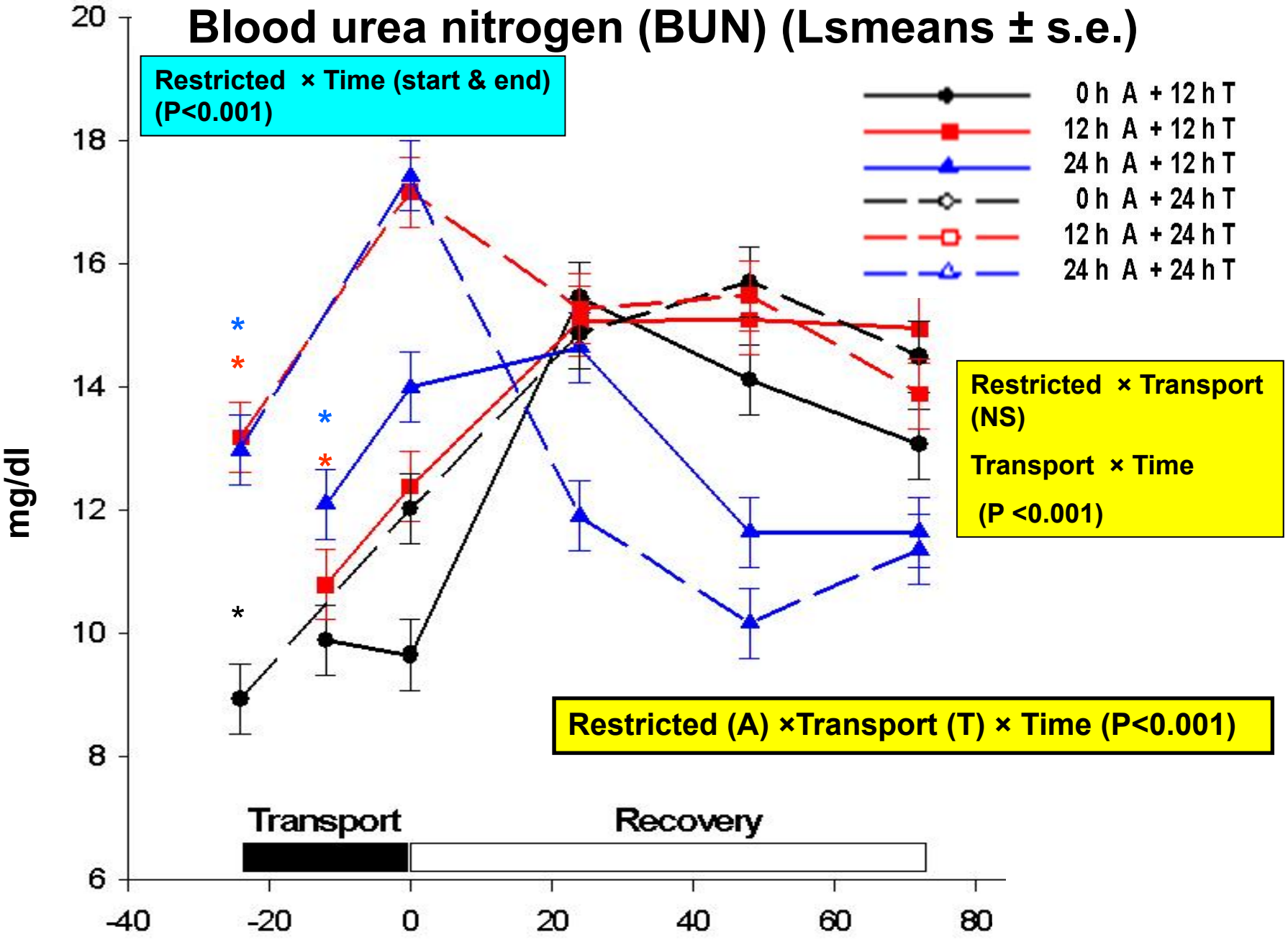
Liveweight (Lsmeans ± s.e.) interactions



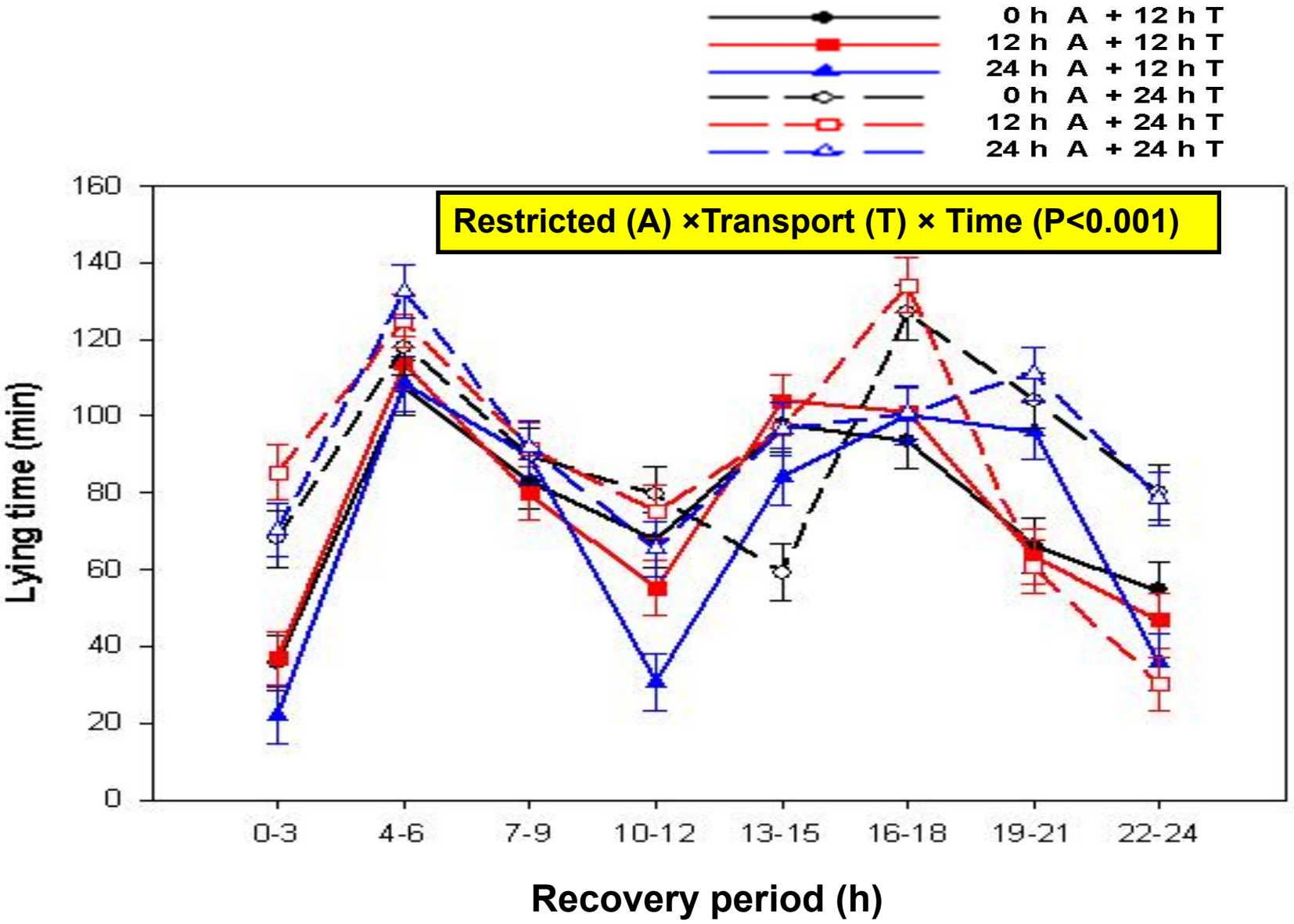

White Blood Cell Number (Lsmeans ± s.e.)
x10⁹ /l
12
11
10
9
8
7
6
0 h A + 12 h T
12 h A + 12 h T
24 h A + 12 h T
0 h A + 24 h T
12 h A + 24 h T
24 h A + 24 h T
Restricted (A) ×Transport (T) × Time (P<0.001)
Transport
Recovery
-40
-20
0
20
40
60
80
Time (h)
Restricted × Time (start & end) (P<0.01)
Restricted × Transport (NS)
Transport × Time (P <0.001)


Urine Osmolality (Lsmeans ± s.e.)
0 h A + 12 h T
12 h A + 12 h T
24 h A + 12 h T
0 h A + 24 h T
12 h A + 24 h T
24 h A + 24 h T
mOsmol/l
1200
1000
800
600
Restricted (A) ×Transport (T) × Time
(P<0.001)
Transport
Recovery
-40
-20
0
20
40
60
80
Time (h)
Restricted × Time
(start & end) (P<0.001)
Restricted × Transport (NS)
Transport × Time (P <0.05)

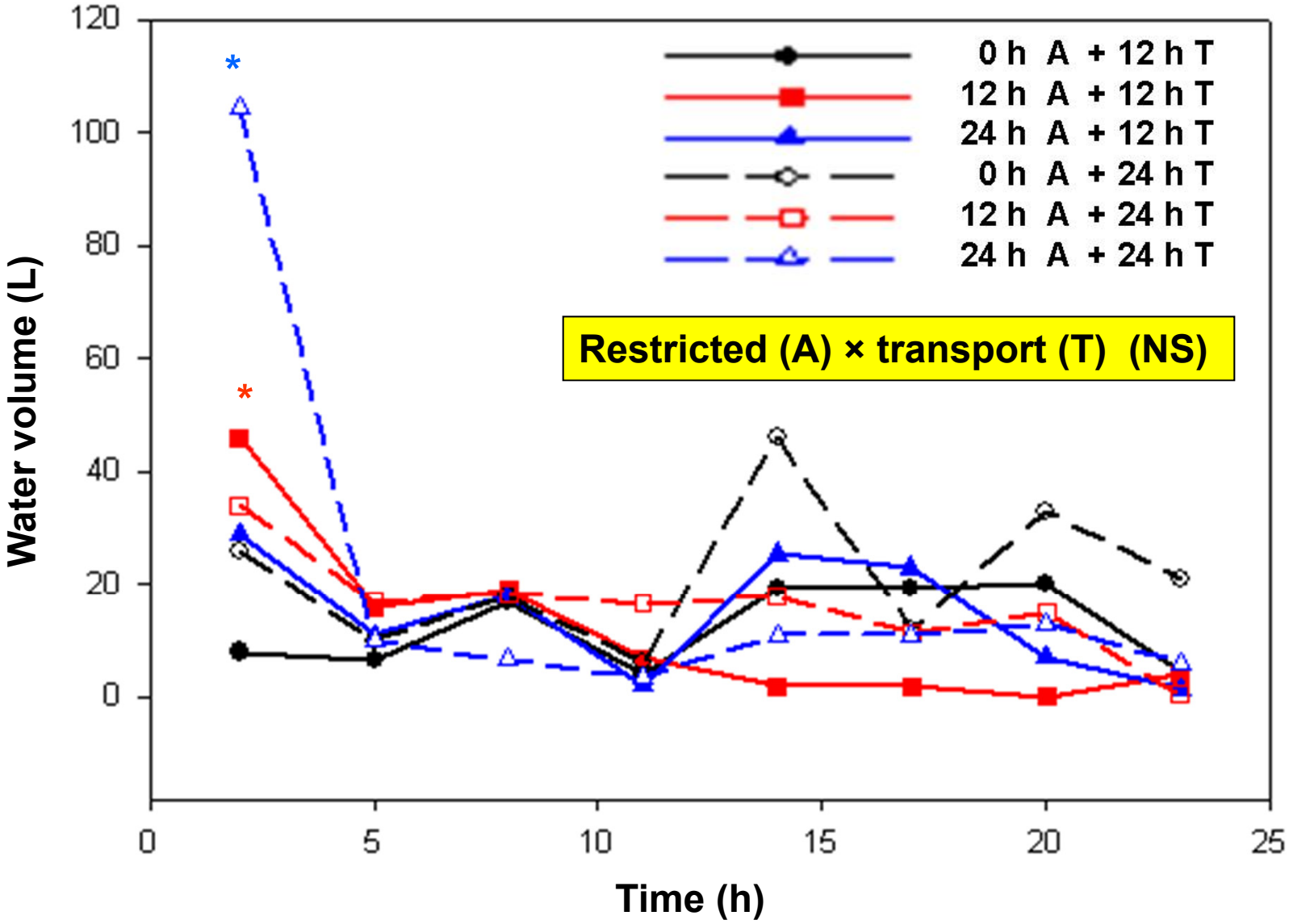

Lsmeans serum Creatine Kinase Activity
Creatine kinase (U/L)
1400
1200
1000
800
600
400
200
0
R 0 h
R 12 h
R 24 h
Restricted
(A) × Time
(P = 0.05)
Pre
0
24
48
72
Time (h)
T 12 h
T 24 h
Transport treatment
(T) × Time (P = 0.05)
-20
0
20
40
60
80
Time (h)
1000
800
600
400
200
0
Creatine kinase (U/L)


Blood urea nitrogen (BUN) (Lsmeans ± s.e.)
Restricted × Time (start & end) (P<0.001)
0 h A + 12 h T
12 h A + 12 h T
24 h A + 12 h T
0 h A + 24 h T
12 h A + 24 h T
24 h A + 24 h T
Restricted × Transport (NS)
Transport × Time
(P <0.001)
Restricted (A) ×Transport (T) × Time (P<0.001)
mg/dl
20
18
16
14
12
10
8
6
-40
-20
0
20
40
60
80
Transport
Recovery

Lying time (Lsmeans ± s.e.) initial 24 h recovery period



Mean water volume consumed (initial 24 h recovery period)



Excreta levels following transport on the floor of the transporter

1). 0 h Restricted + 12 h Transport

6). 24 h Restricted + 24 h Transport

Ambient temperature ranged from -7.6 – 15.9 °C
Relative humidity ranged from 44 – 99 %

Conclusions

- The interaction between restricted treatment × transport treatment × time (sampling) was significant for many of the blood and urine measures, liveweight and behaviour.

- Pre-transport periods of food and water deprivation prior to transport did not enhance the capacity of the animals to cope with transport.

Acknowledgements

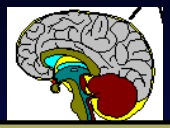
Post-graduate students; Kelly Buckham (MSU), Sandeep Gupta,
Research Scientists; Dr. Andrew Fisher (University of Melbourne)

Thank you for your attention

Teagasc
Agriculture and Food Development Authority

The Irish Agriculture and Food Development Authority

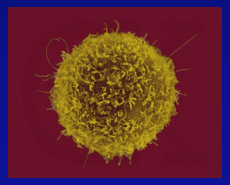
Husbandry Stressors



Stressor → PAIN ↑ → BEHAVIOR

PAIN → STRESS (CORTISOL) ↑

STRESS (CORTISOL) → IMMUNITY ↓



IMMUNITY → MORBIDITY ↑

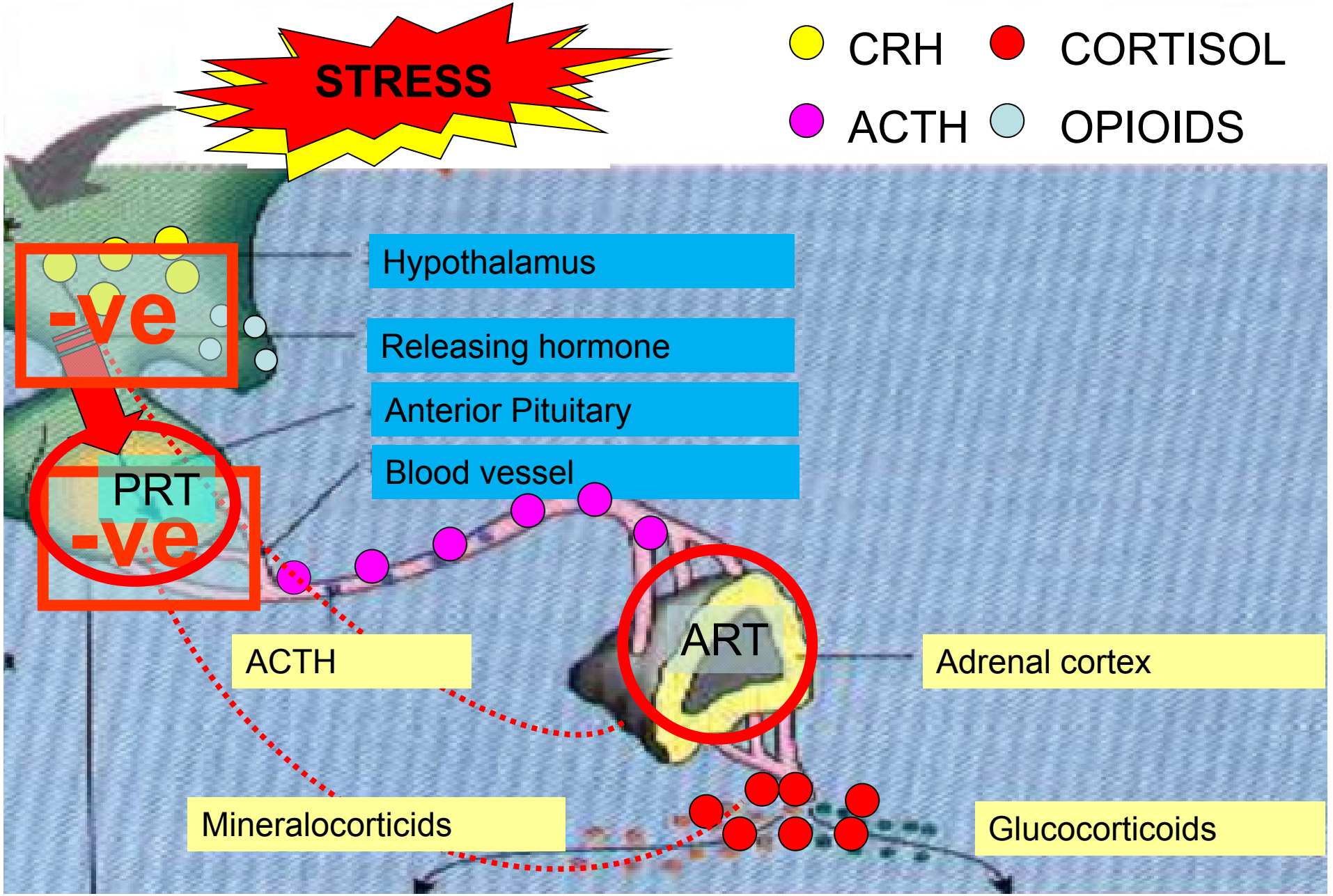
STRESS (CORTISOL) → INFLAMMATION ↑

INFLAMMATION → PERFORMANCE (feed intakes and Growth) ↓

(Teagasc, Research)
Earley, Fisher, Ting

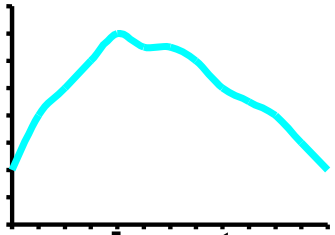
	Australia	EU
Journey duration	36 hr (48 h permissible)	14 h transport 1 h rest (on truck) 14 h transport
Subsequent rest period	12 to 24 hr (36 h if journey was 48 hr)	24 h
Watering interval	36 hr (48 h permissible)	Vehicles fitted with drinkers
Feeding interval	Not stipulated	29 h (for journeys > 8 h)
Space allowance (300 kg cattle)	0.86 m^2 *	0.95 to 1.3 m^2 (325 kg) Trucks must be fitted with humidity.temp loggers & GPS

HPA- axis response



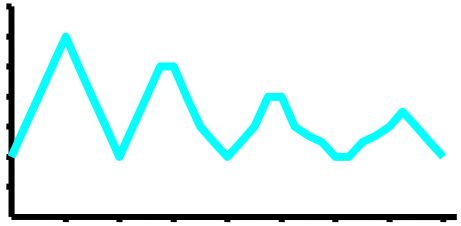
- **Short- to medium-term stress**: Activation of HPA axis

Cortisol



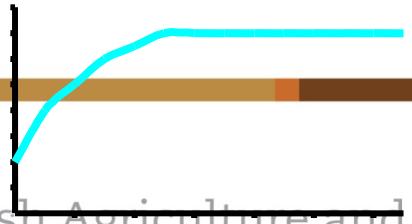
- **Medium-term to chronic stress**: change in HPA axis regulation

Cortisol



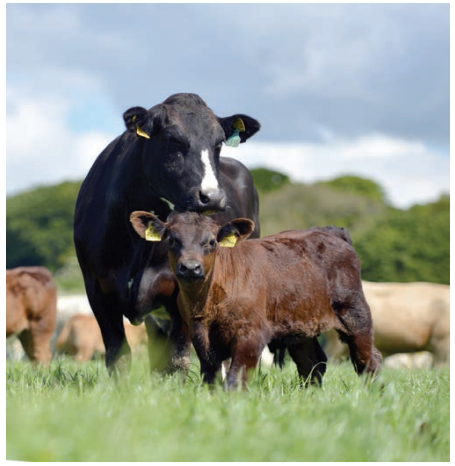
- **Severe stress** + **failure to cope**: HPA axis dysregulation

Cortisol



Galway

Meath



Cork

It is our pleasure to inform you that your presentation entitled 'Research overview - **the impact of animal transport on the health and welfare of beef cattle' with abstract number 18557,** that you have submitted for EAAP Annual Meeting 2014, Copenhagen, Denmark has been accepted by the Scientific Committee.

You are scheduled for session 33 "Interactions between stress, metabolism and immunity" that will be held on 27 August 2014 from 14:00 - 18:00. Your presentation is scheduled as a Theatre presentation. You have 45 minutes for your presentation, **including discussion**.