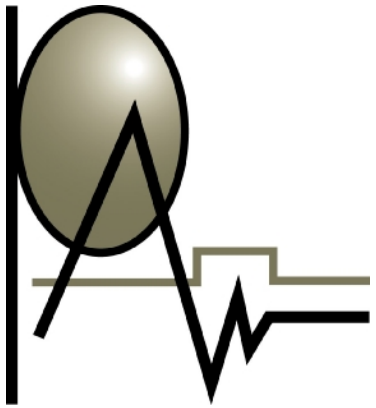
Imprinting of body functions during critical developmental periods

Barbara Tzschentke

Working Group
Perinatal Adaptation

Humboldt-University of Berlin
Institute of Biologie



Lecture Schedule: Embryo Physics Course 2014

I - Imprinting of body functions – the concept

II - Fetal origin of adult disease

III - The bird as model to investigate imprinting of body functions

IIIa - Malprogramming of body functions

IIIb - Pre- and perinatal environment and imprinting of the thermoregulatory system

- Influence of chronic temperature changes (perinatal epigenetic temperature adaptation)

- Influence of short-term temperature changes (perinatal temperature training)

IV - Summary, conclusions

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Konrad Lorenz

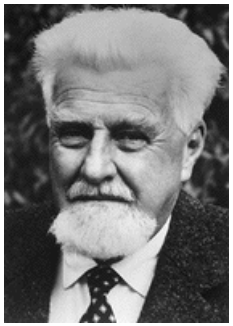
Employed the term **‘imprinting’** – development of social binding

‘imprinting’

⇨ occurs in **‘critical periods’**

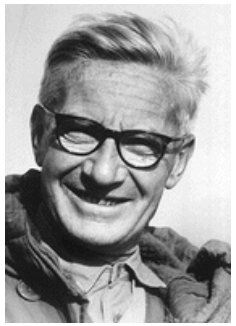
⇨ limited to the animal´s very early life

Nobel prize for medicine 1973



Konrad Lorenz

Karl von Frisch



Nicolaas Tinbergen

Critical and sensitive periods

A **‘critical period’** has to be considered the time during which a certain experience necessarily must occur to enable development to proceed normally. It begins and ends rather abruptly and is limited to the early ontogeny.

A **‘sensitive period’** is a period of “maximal” sensitivity to certain kinds of environmental experiences. It begins and ends rather gradually and may occure during total lifetime.

Bailey et al. (2001), Hensch (2005)

Imprinting of Physiological Control Systems during the Perinatal Period

obviously
realized by

neuronal ‘imprinting’
at the microstructural level
(e.g. in therms of
synaptic plasticity)

+

by lasting environment-induced
modification of the genome

Tzschentke & Plagemann (2006): *WPSJ,* **62**: 626-637.

‘Imprinting’ of physiological control systems

During perinatal ontogeny,
environmental conditions may have a strong influence on the **determination** of the set-point of physiological control systems.

Open loop system

Closed control system

Actual level of the regulated parameter determines the set-point of the system

(determination rule, Dörner (1974). *Acta Biologica and Medica Germanica* **33**: 129-148)

Günter Dörner
Pioneer of developmental
Neuroendocrinology

General etiological concept on
'epigenetic', perinatal programming
of the lifetime function of fundamental
regulatory systems

hormones are
environmental-dependent
organizers of the
neuro-endocrine system

Role of homones, transmitters/ neuropeptides, cytokines (as immune cell hormones) during critical periods

Acting as critical endogenous effectors, which transmit environmental information to the genome

Acting as epigenetic factors

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Perinatal malprogramming

When present in non-physiological concentrations

↓

Hormones transmitters/ neuropeptides, and cytokines can act as ‘endogenous functional teratogens’
by
‘malprogramming’
the ‘ neuro-endocrine-immune network’

↓

developmental disorders and diseases throughout later life

Prenatal hyperglycaemia (e.g. under gestational diabetes)

↓

Leading to fetal hyperinsulinism

↓

May cause long-lasting malprogramming of central regulation of food intake, body weight and metabolism

↓

Leading to obesity, diabetes and related diseases

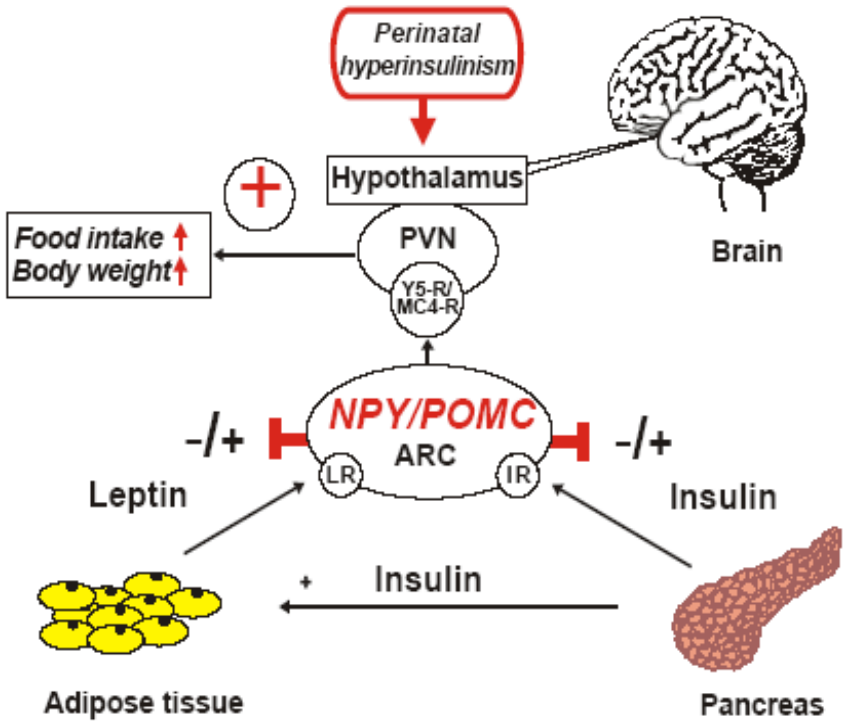


Fig. 3. Exemplary mechanism of perinatal neuroendocrine 'malprogramming'. A temporary hyperinsulinism during critical periods of early development may result in a kind of persisting resistance (increased threshold) to the circulating satiety signals insulin and leptin in orexigenic as well as anorexigenic hypothalamic regulatory systems, leading to a perinatally acquired obesity disposition. NPY—neuropeptide Y; POMC—proopiomelanocortin.

Plagemann (2004): *Journal of Perinatal Medicine* 32: 297-305.

Plagemann (2006): *Hormon Research* 65(Suppl 3): 83-9.

fetal stress
caused by malfunction
of the uterus/placenta

↓

can lead to
chronic moderate hypoxia

↓

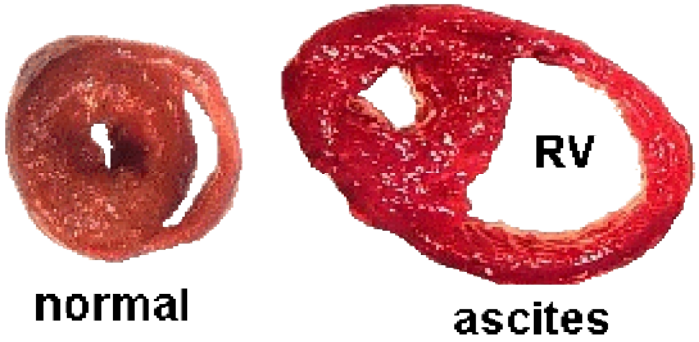
increased risk of adult
cardiovascular diseases

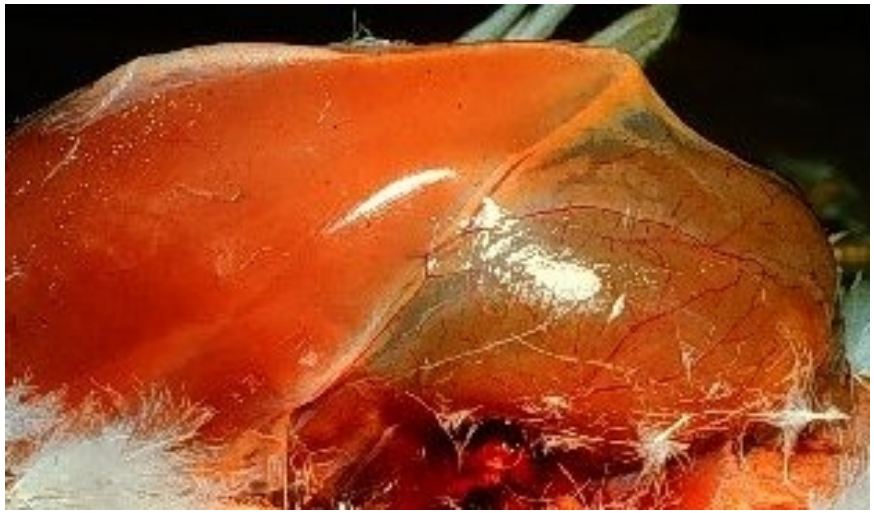
Similar effects can be seen in birds.

Ascites syndrome

associated with

- abnormally high blood pressure (pulmonary hypertension syndrome, PHS) → right ventricle hypertension (RVH)
- increased blood pressure in the veins
- excessive accumulation of fluid in the body cavity





Pulmonary Hypertension Syndrome (PHS)

&

Right Ventricle Hypertension (RVH)

↓

can already be related to causal factors during incubation

prenatal hypoxia or hypoxemia

RUITENBEEK et al. (2000): *Circulation* **102**: 2892-2897.

HASSANZADEH et al. (2004): *Avian Pathology* **33**: 558-564.

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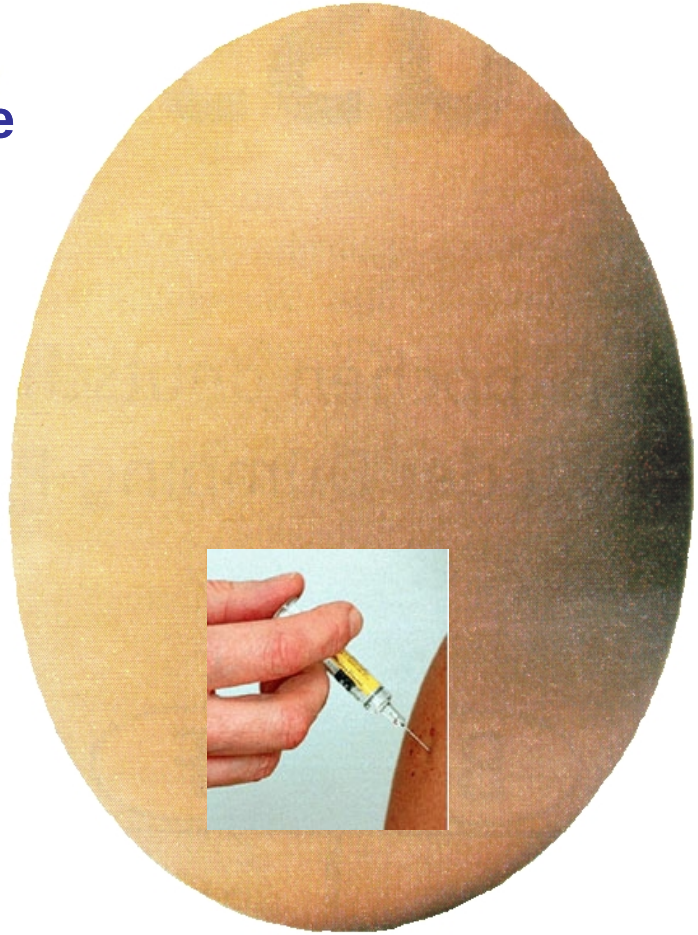
Independent development from the mother allows high standardized and controlled changes in epigenetic (environmental) factors during different time windows of early development.

ambient temperature

O_2 - / CO_2 – content

application of substances

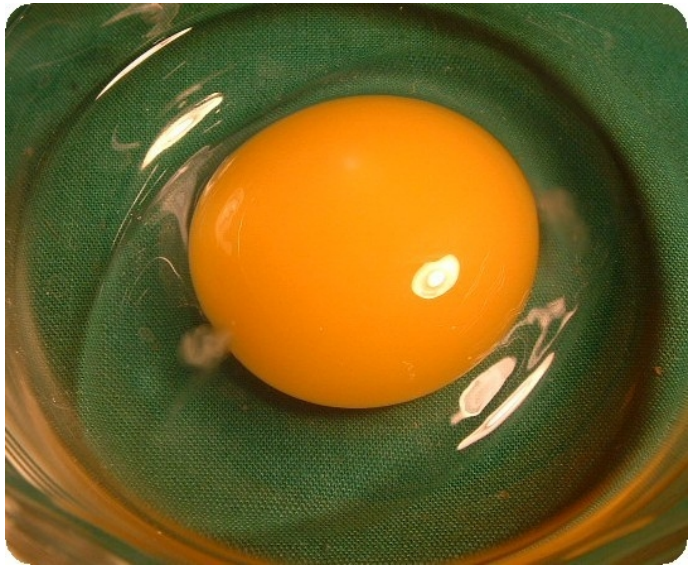


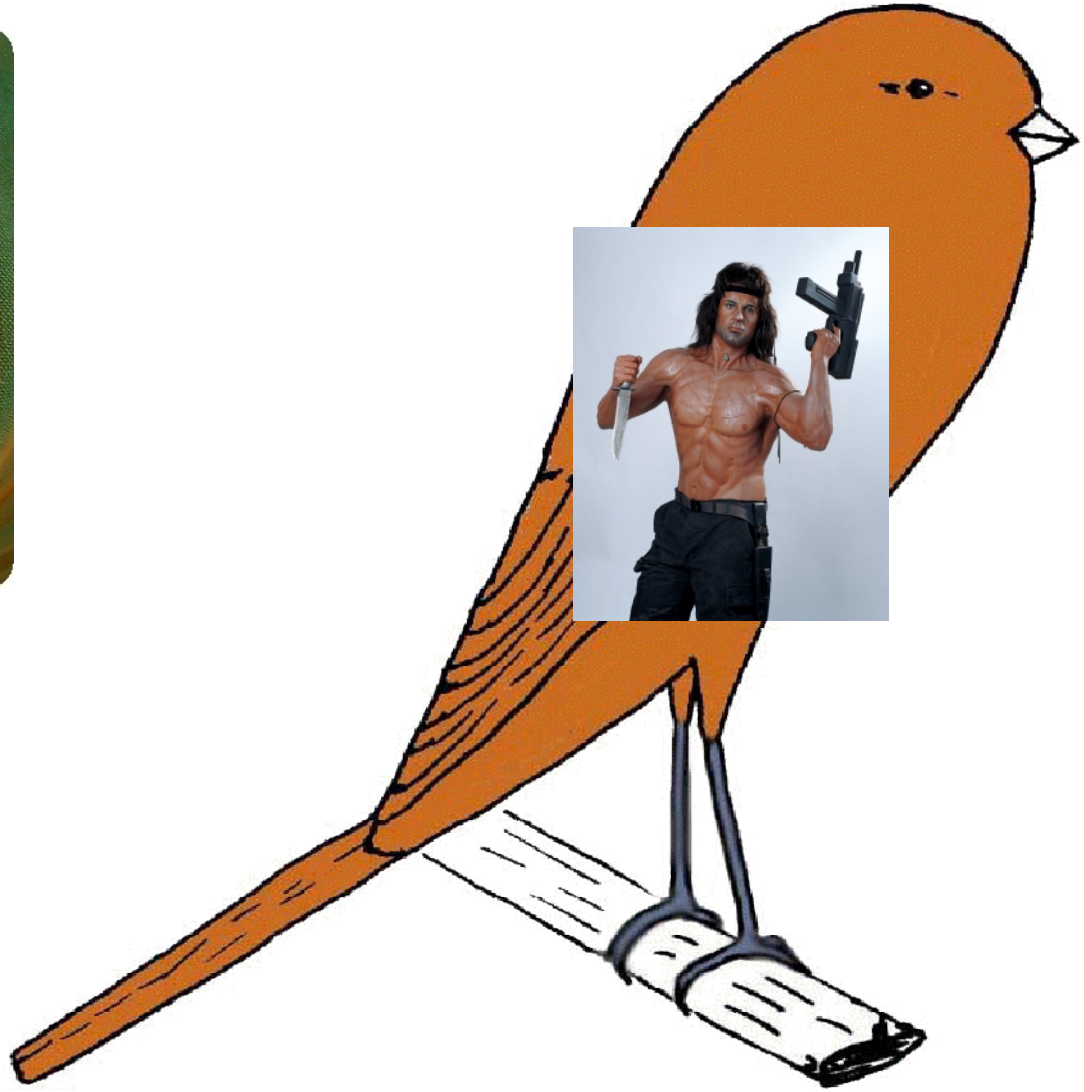
relative humidity

light

acoustic signals

Model for maternal stress





High testosterone level in the yolk changes post-hatching behaviour

Schwabl (1996):*CBP* **114A**: 271 - 276.
Schwabl (1997):. In: Harvey, S., Etches, R.J. (eds.) *Perspectives in avian endocrinology. Bristol; Society for Endocrinology,* pp. 3 – 13.
Groothuis, Schwabl (2008): Phil Trans R Soc B 2008; 363: 1647-61.

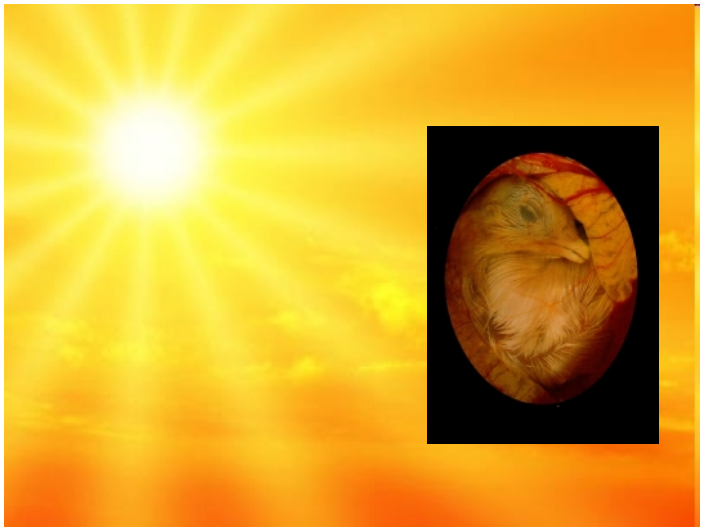
Prenatal influence of light

Light stimulation during final incubation

Development of functional brain assymmetries

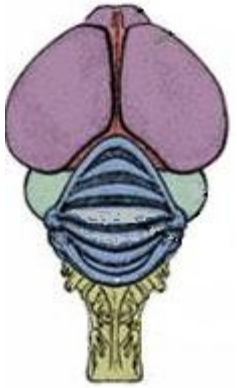
Postnatale behaviour

Roger, L.J. (2012): J. Ornithol., 153: S61-S74



Light during final incubation

Functional brain lateralization



Left hemisphere

learnt and routine behaviour under nonstressful situations

- ability to learn to distinguish between different objects
- attention in training experiments

Right hemisphere

behaviour under emergency or stressful conditions

- attention to novel objects & predators
- social learning
- fear response
- maintenance of social hierarchy
- recognition of face-like stimuli

Incubation in the dark

no or weakly developed anatomical and functional brain asymmetries

post-hatching limitations or losses in behavioural abilities

- learning, unable to discriminate between different conditions
- social behaviour, less stable social hierarchies
- produce more distress calls, higher fearfulness

more vulnerable to post-hatching stress

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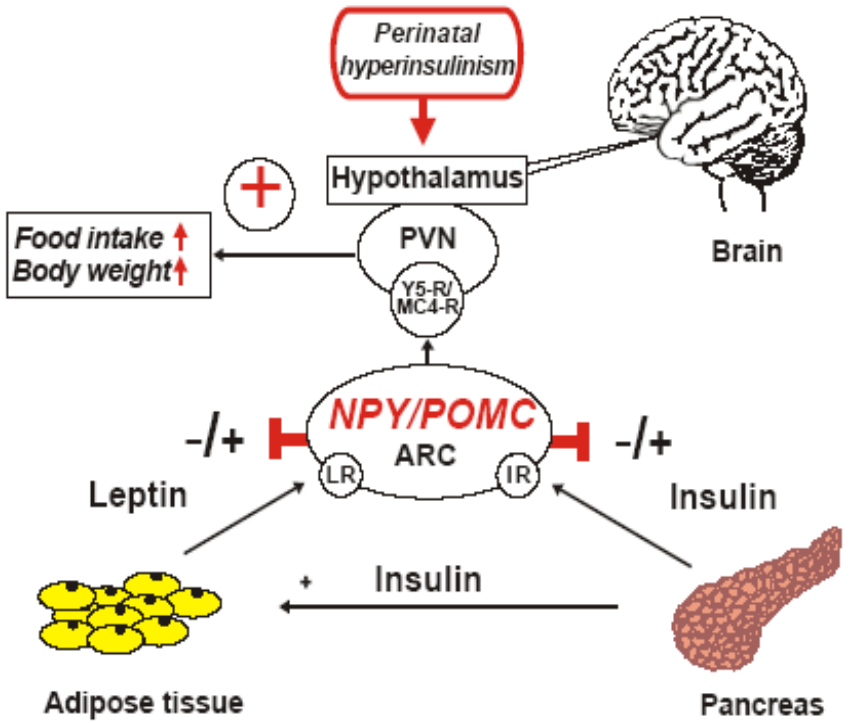

Fig. 3. Exemplary mechanism of perinatal neuroendocrine 'malprogramming'. A temporary hyperinsulinism during critical periods of early development may result in a kind of persisting resistance (increased threshold) to the circulating satiety signals insulin and leptin in orexigenic as well as anorexigenic hypothalamic regulatory systems, leading to a perinatally acquired obesity disposition. NPY—neuropeptide Y; POMC—proopiomelanocortin.

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Plagemann (2006): *Hormon Research* 65(Suppl 3): 83-9.

Epigenetic prenatal malprogramming of metabolism, food intake and body weight regulation using the bird as a model (example: gestational diabetes)

Co-operation between

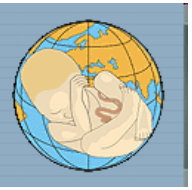
Humboldt-University of Berlin
WG Perinatal Adapatation

Barbara Tzschentke

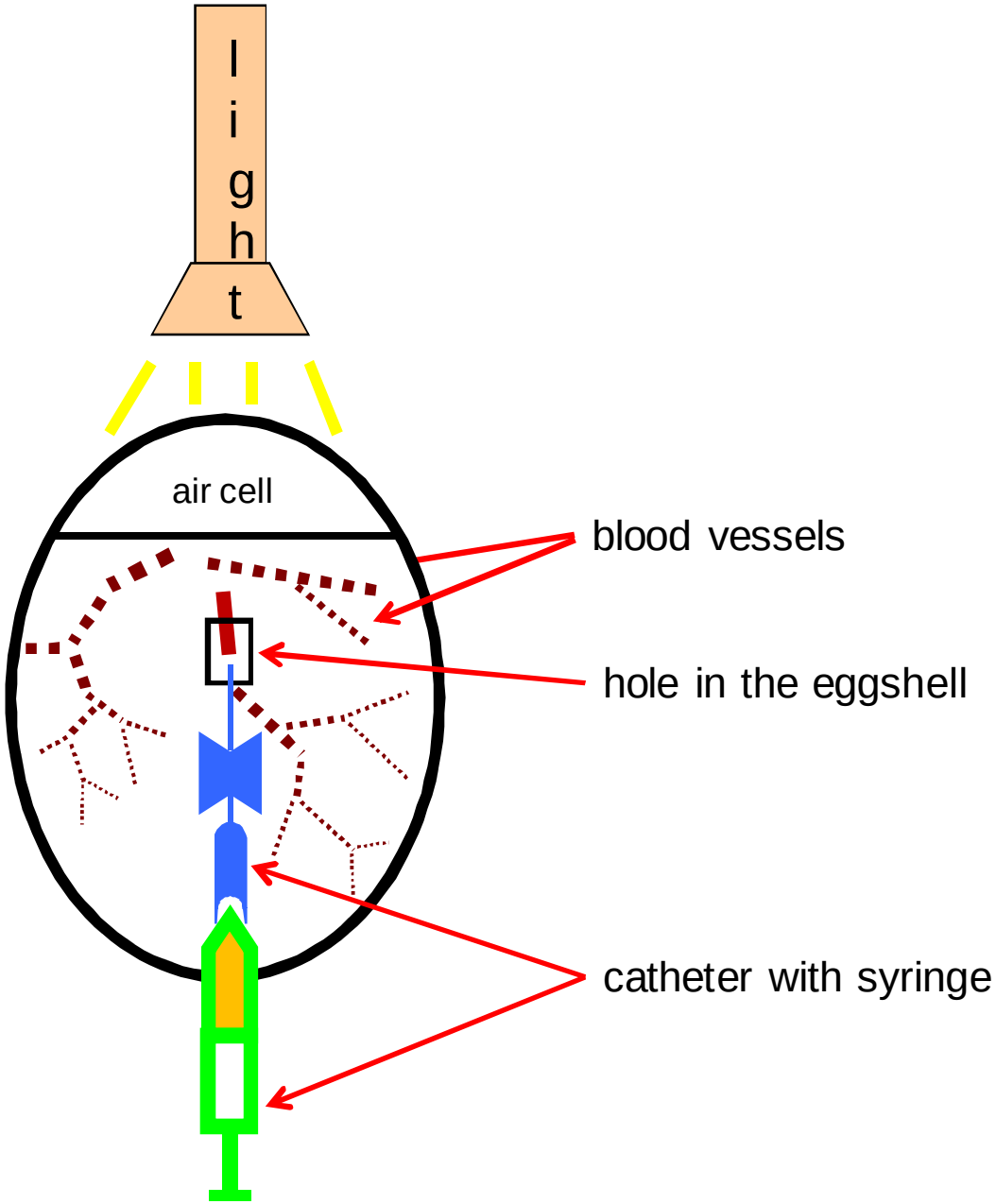
CHARITÉ CAMPUS VIRCHOW-KLINIKUM

Clinic of Obstetrics

Division of 'Experimental Obstetrics'

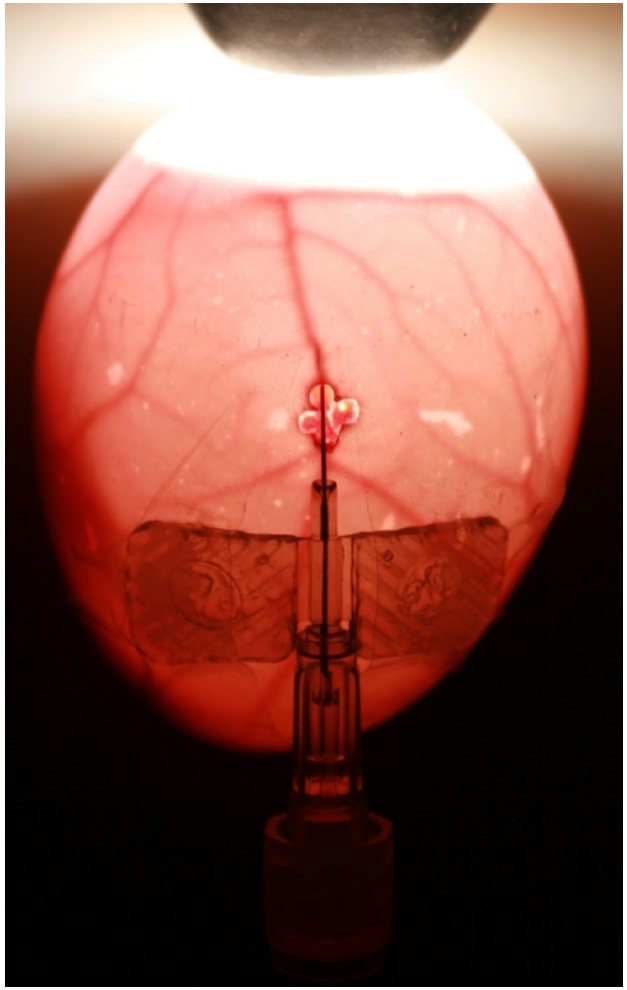


Andreas Plagemann

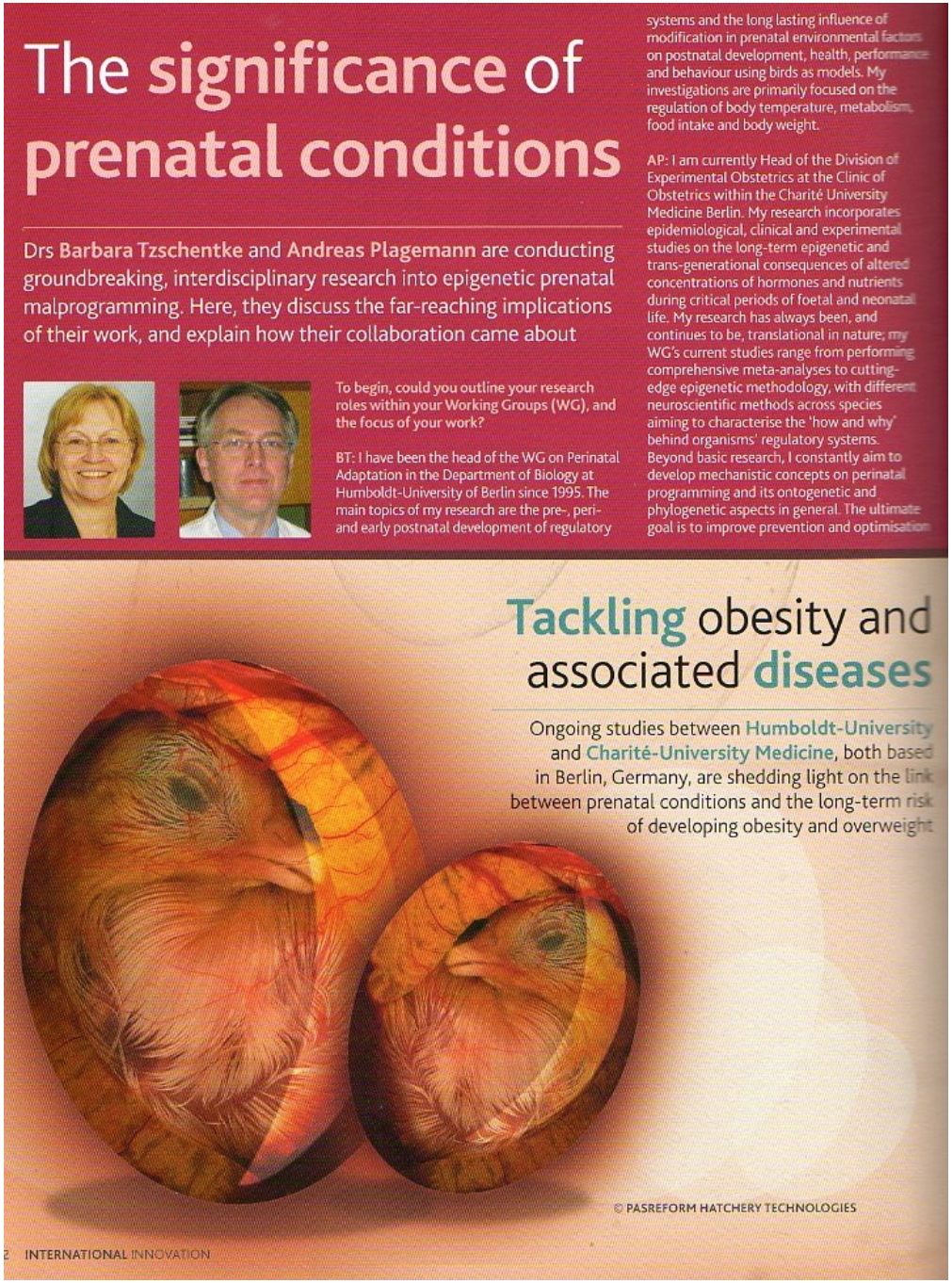


Daily glucose injection between day 14 and 17 of incubation

(in chicken total incubation time 21 days)



International Innovation, Healthcare Issue 21, 2013



The significance of prenatal conditions

Drs Barbara Tzschentke and Andreas Plagemann are conducting groundbreaking, interdisciplinary research into epigenetic prenatal malprogramming. Here, they discuss the far-reaching implications of their work, and explain how their collaboration came about

To begin, could you outline your research roles within your Working Groups (WG), and the focus of your work?

BT: I have been the head of the WG on Perinatal Adaptation in the Department of Biology at Humboldt-University of Berlin since 1995. The main topics of my research are the pre-, peri- and early postnatal development of regulatory systems and the long lasting influence of modification in prenatal environmental factors on postnatal development, health, performance and behaviour using birds as models. My investigations are primarily focused on the regulation of body temperature, metabolism, food intake and body weight.

AP: I am currently Head of the Division of Experimental Obstetrics at the Clinic of Obstetrics within the Charité University Medicine Berlin. My research incorporates epidemiological, clinical and experimental studies on the long-term epigenetic and trans-generational consequences of altered concentrations of hormones and nutrients during critical periods of foetal and neonatal life. My research has always been, and continues to be, translational in nature; my WG's current studies range from performing comprehensive meta-analyses to cutting-edge epigenetic methodology, with different neuroscientific methods across species aiming to characterise the 'how and why' behind organisms' regulatory systems. Beyond basic research, I constantly aim to develop mechanistic concepts on perinatal programming and its ontogenetic and phylogenetic aspects in general. The ultimate goal is to improve prevention and optimisation

Tackling obesity and associated diseases

Ongoing studies between Humboldt-University and Charité-University Medicine, both based in Berlin, Germany, are shedding light on the link between prenatal conditions and the long-term risk of developing obesity and overweight

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During a critical perinatal period, already short-term experiences of hyperglycemia can lead to long-lasting changes in neuronal glucose sensitivity on both a cellular and a molecular level, affecting neuronal plasticity and gene expression respectively.

These hyperglycemia-induced changes could potentially point to an increase in the long-term risk of developing diabesity.

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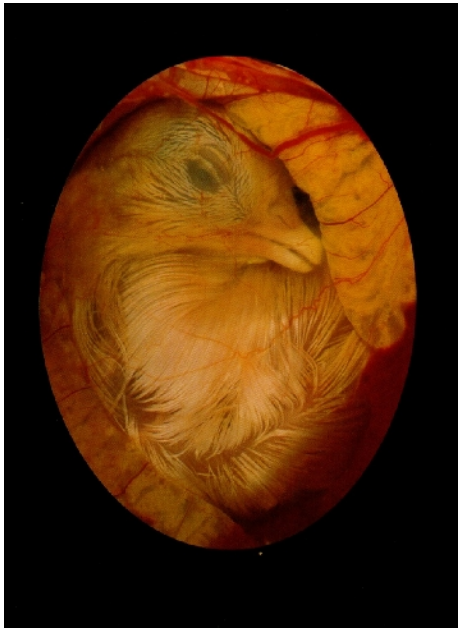
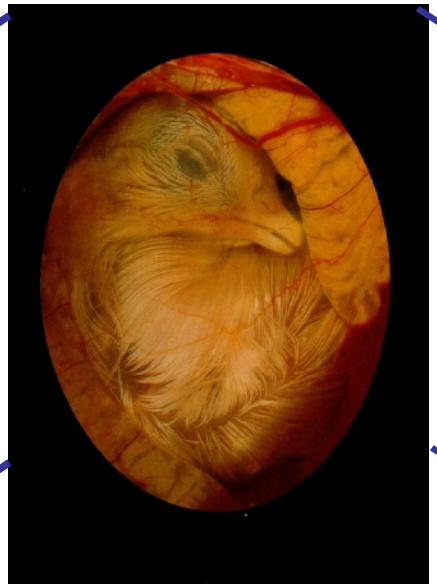
Optimal time frame for temperature manipulations with long-lasting implications

During final incubation the bird embryo has all prerequisites to react on changes in incubation temperature



Tzschentke, B. (2007): *Poultry Science,* **86**: 1025-1036.

Final Incubation



Neuro-endocrine system
is well developed
(e.g. hypothalamic-pitatury-thyroidal-axis)

Physiological systems
and
sensory capacity, e.g. for hearing and vision,
are functional

Behavioural mechanisms
(acoustic communication, motility)

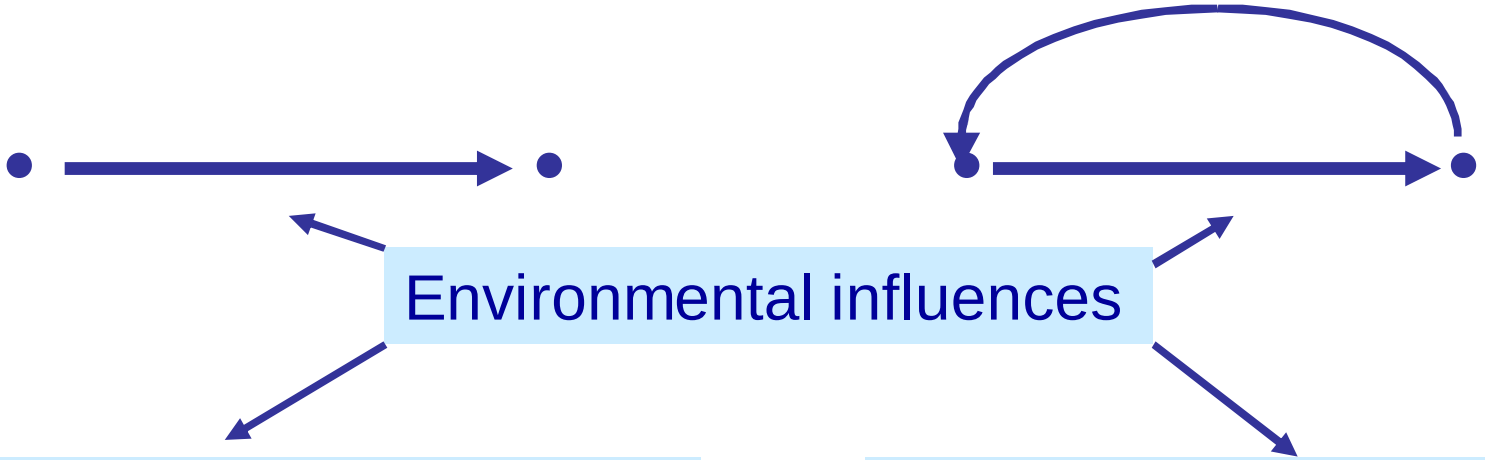
Regulatory systems develop
feed back mechanisms
(e.g. thermoregulation)

Critical period

Development of feed back mechanisms: a „critical period“ in the development of physiological control systems

Open loop system

Closed control system



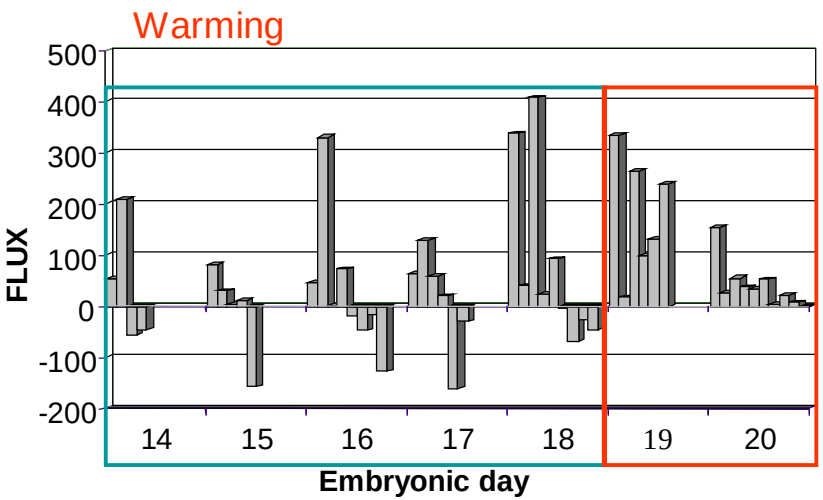
Environmental influences

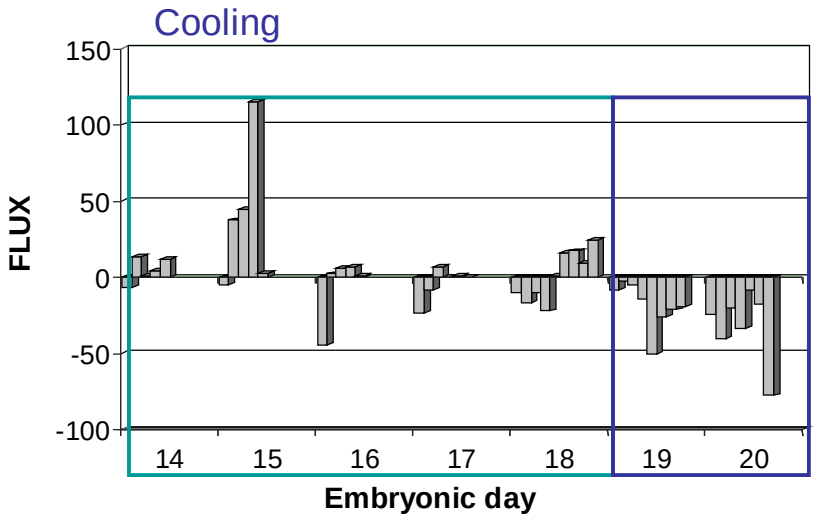
Proximate non-adaptive reactions

Coordinated and adaptive reactions

Tzschentke & Plagemann (2006): WPSJ, **62**: 626-637.

Characterization of "critical periods" by environmental manipulation of immature physiological mechanisms in chicken embryos





Laser-Doppler-Probe

measurement of blood flow in the chorioallantois membrane

uncoordinated, proximate non-adaptive reactions
blood flow is increasing or decreasing while warming or cooling

coordinated and adaptive reactions

blood flow is increasing during warming

and decreasing during cooling

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Chronic temperature changes

- Adapation to high or low temperatures -

Via imprinting of the thermoregulatory sytem

Normal incubation temperature 37.5 ° C

Day of Incubation

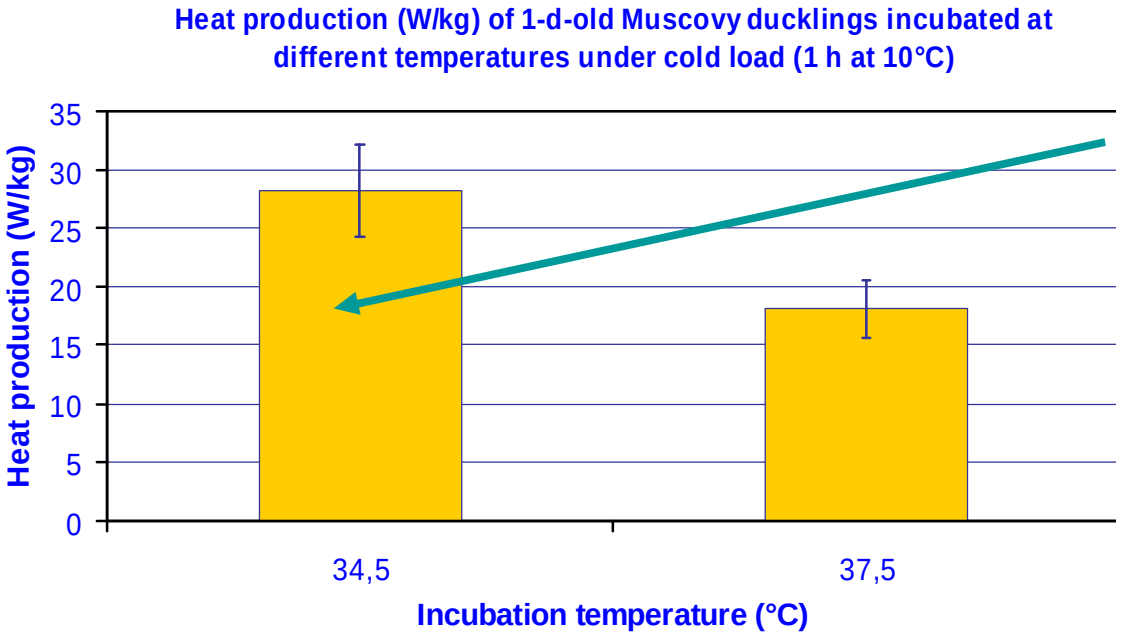
0-17 | 18-21

Chronic temperature increase (38.5 ° C) → Heat adaptation

Chronic temperature decrease (34.5 ° C) → Cold adaptation

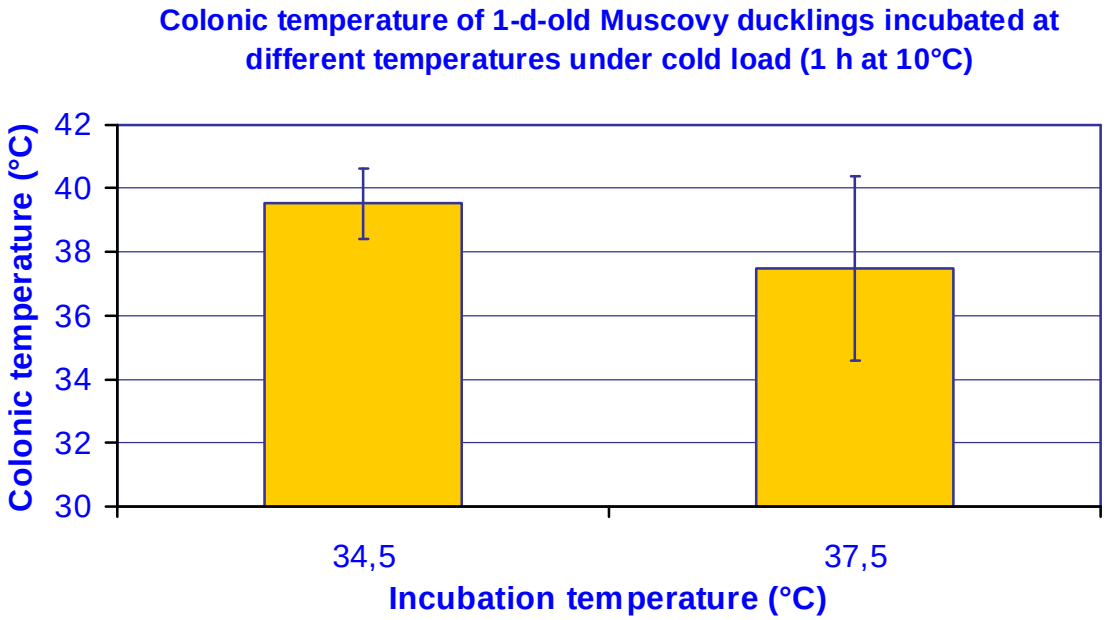
Post-hatching Changes

Physiological parameters
Neuro-endocrine parameters
Thermoregulatory behaviour
feed intake, body weight



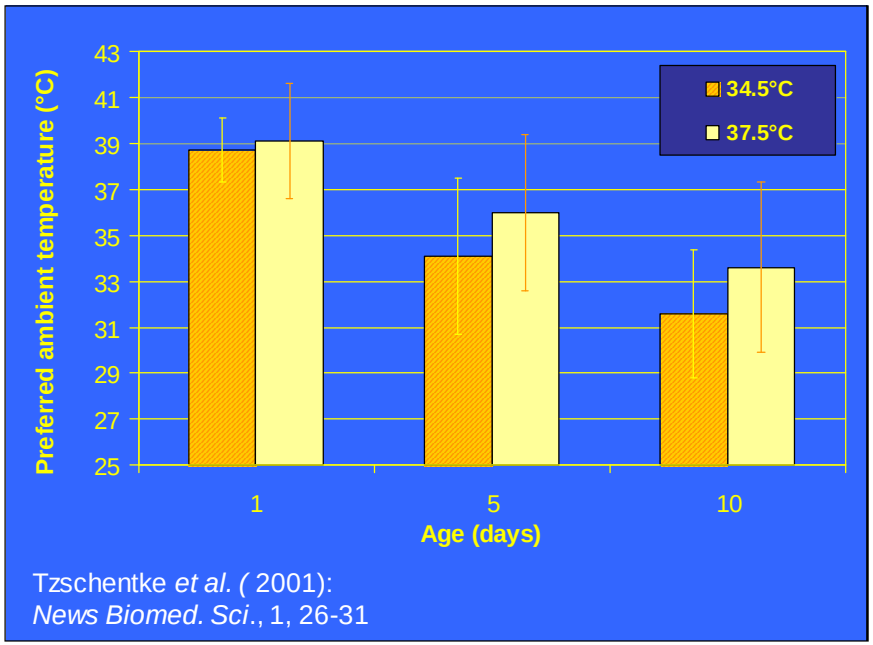
In the cold-incubated chicken postnatal increased T_3/T_4 concentration

***Decuypere (1984) Arch. Exp. Vet.-Med*. 38, 439-449**

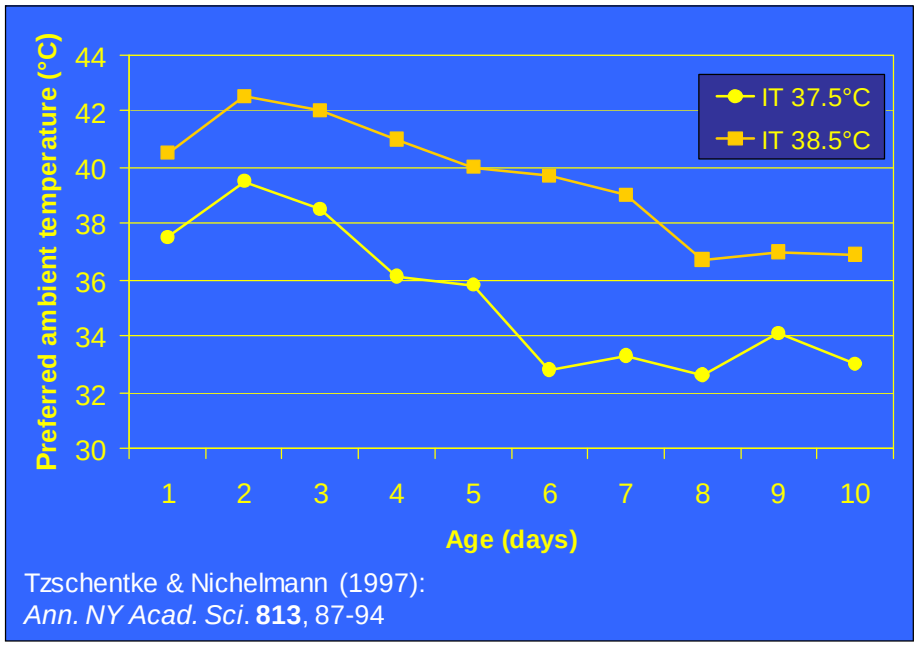


Tzschentke & Nichelmann (1999):*Ornis Fennica* 76: 189-198 .

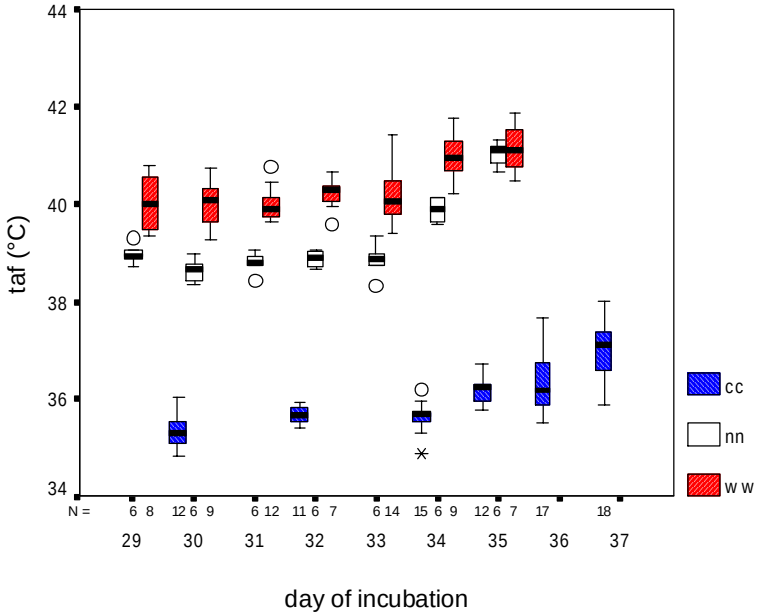
Epigenetic Temperature Adaptation



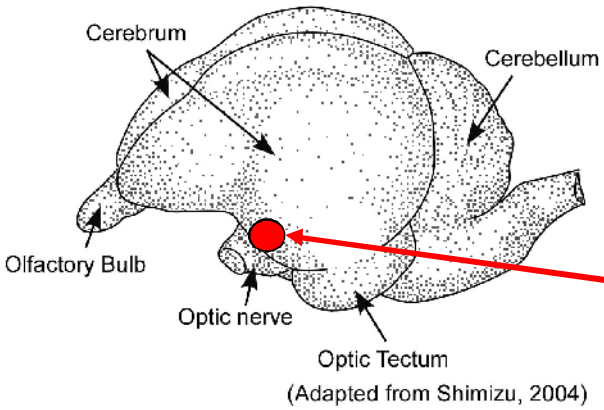
Tzschentke *et al. (* 2001):
News Biomed. Sci., 1, 26-31



Tzschentke & Nichelmann (1997):
Ann. NY Acad. Sci. **813**, 87-94

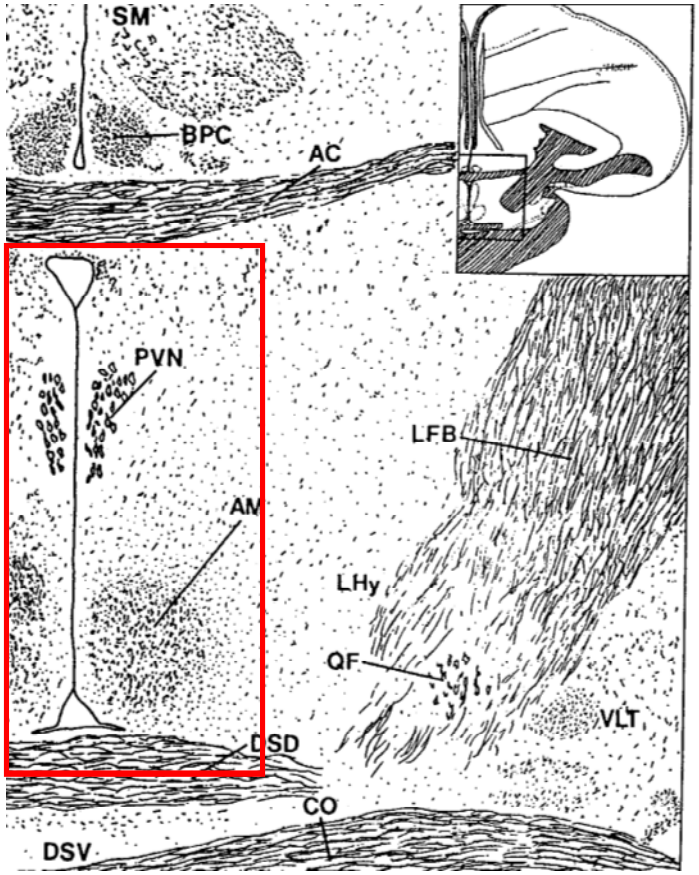


Loh *et al*. (2004): *Avian Poult. Biol. Rev*, **15:3/4**, 119-128



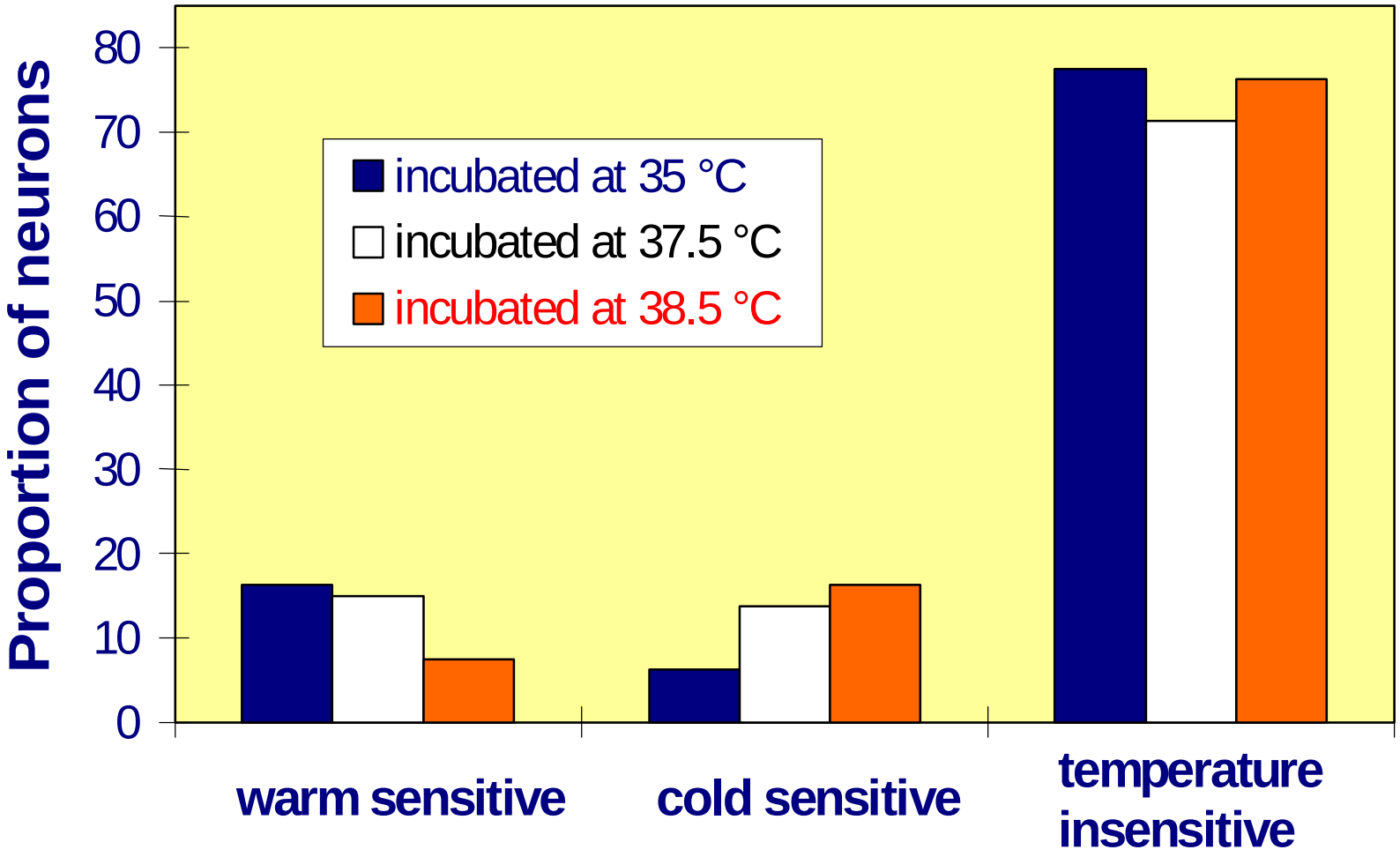
(Adapted from Shimizu, 2004)

Hypothalamus
(thermoregulatory center)

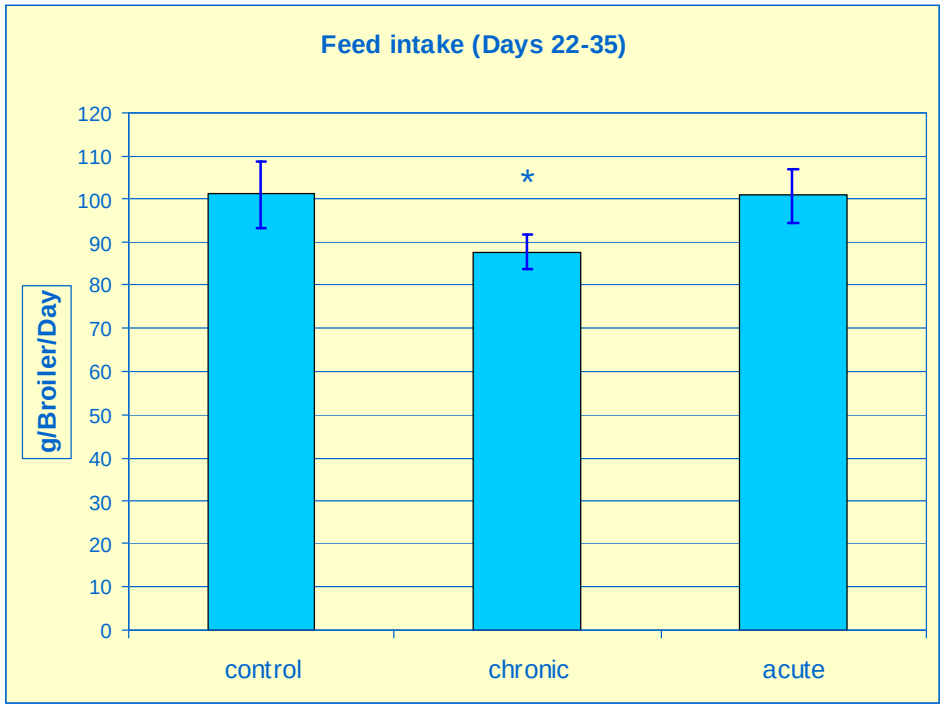


after Kuenzel and Tienhoven (1986)

10-d-old Muscovy ducklings



Tzschentke & Basta (2002): *CBP Part A* 131, 825-832



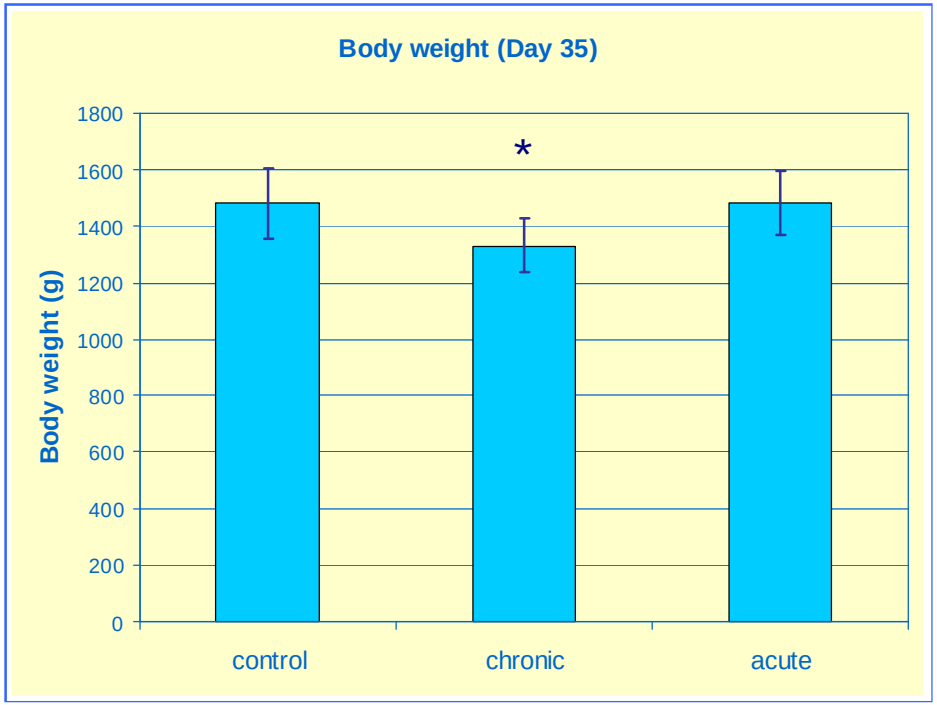
Growing until slaugher age (35 days) at 32°C

♂

Decrease in

Feed intake

Body weight at slaughter



Incubation from day 18 until hatch

1) normal (control)
2) + 1°C (chronic)
3) 2 h/day + 1°C (acute)

Halle & Tzschentke, 2011: *J. Poult. Sci*. 48, 97-105.

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Short-term temperature stimulation

Perinatal activation of regulatory systems may have a

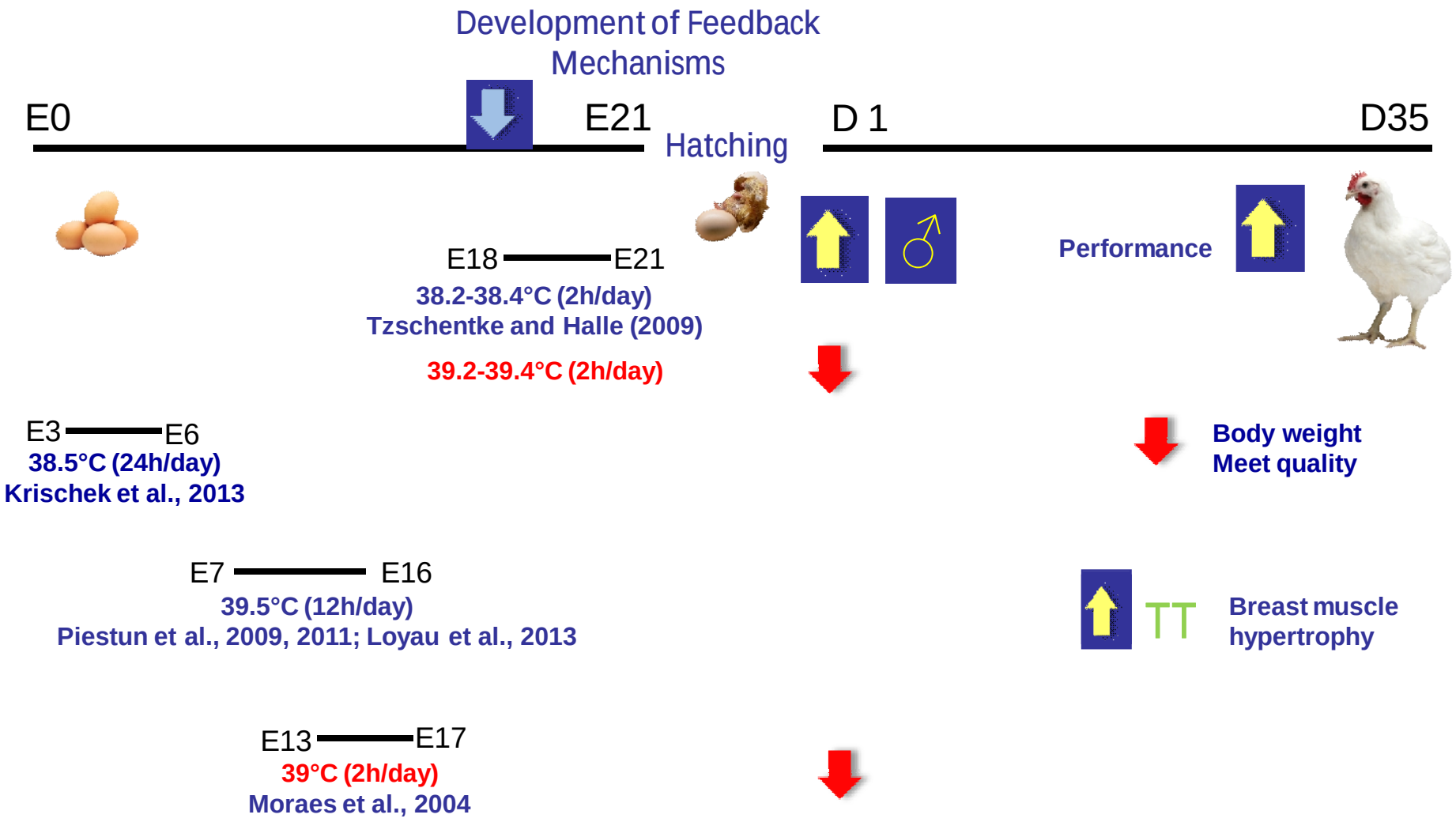
training effect

on the postnatal efficiency of body functions

improved robustness

Timing (as well as amplitude and duration) of embryonic temperature stimulation is critical for achieving chicken robustness

Long-lasting improvement in performance and adaptability are determinants



Day of incubation
0-17

Temperature stimulation
+ 1°C for 2 hrs per day

Decreased embryonivc heat production during stimulation

Decreased embryonic temperature

Increased hatching rate

Increased proportion in male hatched chicks

Increase in body weight at slaughter age in male broilers

Improved feed conversion in male and female broilers

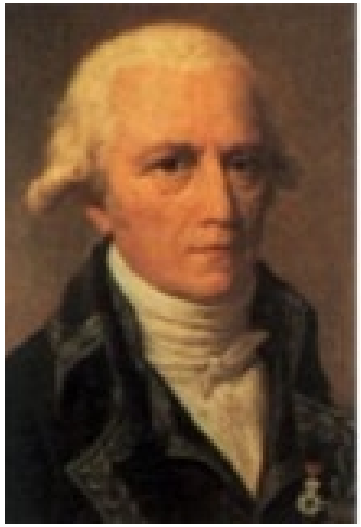
Changed behaviour
? less fearful ?
better immune response
Impact on reproduction

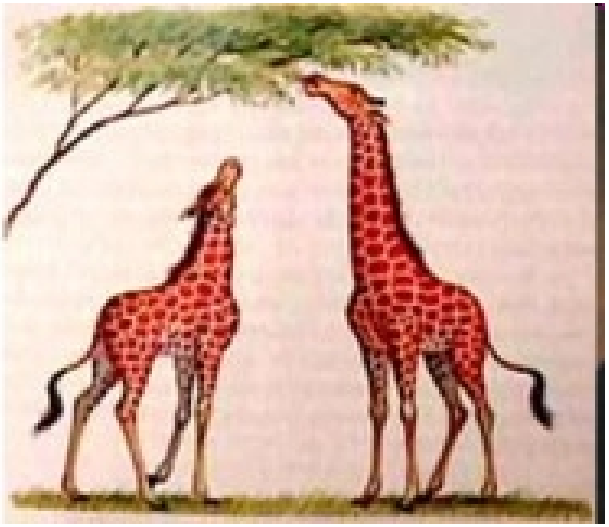
Changed phenotype

Environmental induced phenotypic changes may be even passed on the succeeding generations in an epigenetic fashion

Epigenetics
Transgenerational transfer of phenotypic characters without modification of gene sequence

(Ho and Burggren (2010): Epigenetics and transgenerational transfer: a physiological perspective. J. Exper. Biol. 213: 3-16.)





Lamarck, 1809

Dutch-Hunger-Winter 1944

400-800 calories/d

Malnutrition of pregnant womans

Epigenetic changes in DNA in the offsprings
(e.g. IGF2 methylation)
Take effects up to the third generation

Offsprings

- reduced growth
- higher rates of obesity, diabetes, cardiovascular disease
- age-associated decline of cognitive functions

Sources:

- University Leiden, NL
- University Göttingen, Germany, Department of Epigenetic and Neurodegenerative Disease, Prof. André Fischer (IGF2 methylation, anxiety disorder, Alzheimer)

- Rooij et al. (2010): PNAS 39: 16881–16886.
- Schulz (2010): PNAS, 39, 16757–16758

Transgenerational effects of perinatal temperature stimulation?

F 0

F 1

Quails

Hatching results

F 1

F 2

	F 1		F 2	
	Control	T-Stimulated F 1	Control	T-Stimulated F 1
Eggs/ Incubator	279	279	244	246
Hatching rate %	64.2	71.3	68.0	74.8
Body weight g/chicken	8.1	8.3	7.2	8.0

I - Imprinting of body functions – the concept

II - Fetal origin of adult disease

III - The bird as model to investigate imprinting of body functions

IIIa - Malprogramming of body functions

IIIb - Pre- and perinatal environment and imprinting of the thermoregulatory system

- Influence of chronic temperature changes (perinatal epigenetic temperature adaptation)

- Influence of short-term temperature changes (perinatal temperature training)

IV - Summary, conclusions

Summary & Conclusions

Imprinting of body functions during the critical period in the development of regulatory systems is a fundamental process of life, which improves adaptability to the prenatal and with it to the expected postnatal environment.

Under suboptimum environmental conditions (or mismatch between the pre- and postnatal environment), 'imprinting' of physiological control systems may be a basis for perinatal malprogramming of the respective systems, which causes disorders and diseases during later life.

Already short-term and moderate changes in environmental conditions during critical periods in the early development may modify the phenotype. The effects are different in males and females of the respective species.

Imprinting of body functions has long-lasting effects and may be even passed on the succeeding generations in an epigenetic fashion.

Thank you!