JACUMBREAVÍCOLA LATINOAMERICANA



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In collaboration with:









Introduction

- The impact of heat stress
- Examples of research of phytogenics and heat stress
- Tools for evaluating phytogenic candidates for heat stress management
- Validating those candidates
- Summary

Major growth in demand for poultry meat 2000 – 2030 in (sub)tropical areas



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Source: FAO (September 2011)

2000 – 2030: Increasing global temperature



Heat stress: Acute (short duration) and chronic (long-term) Cyclic (diurnal rhythm) and constant (no diurnal rhythm)

NASA ModelE Climate Simulation (SCo7)

Temperature/Humidity Index (THI) and heat stress



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Broiler signals (2015)

Different mechanisms to restore balance



Effect of heat stress in broilers

- Ross broilers exposed to chronic heat stress day 14-28
- Heat stress:
 - **Cyclic:** 32-24-32°C (32°C for 8h/d)
 - Constant: 32 or 34°C





Heat stress challenges the anti-oxidant defense system

- 180 Quail
- Heat-stress treatments:
 - $-TN(22^{\circ}C)$
 - HS (34°C: 8h/d for 12 wk)
- Oxidative stress biomarkers: Hepatic MDA, SOD, CAT, GSHPx Hepatic transcription factors: Nrf2 and **NF**_KB
- **Results:**
 - Feed intake and egg production reduced by 10% and 14%



Antioxidant defence

Source: Sahin et al. (2010)

Effect of heat stress in laying hens

A meta-analysis of 131 peer-reviewed papers:

	Thermo-neutral (15-29 °C)	Heat stress (30-35 °C)	Change (%)
Feed intake (g/d)	112.8	87.3	-23
Egg production (%)	86.9	77.1	-11
Egg weight (g)	58.1	53.9	-7
Egg mass (g/b/d)	48.5	44.1	-9
Shell strength (g)	3513	3009	-11
Shell thickness (mm)	0.363	0.344	-5

Strategies to reduce the impact of heat stress

Management

Withdraw feed

Adequate feeder space and drinkers

Add salt to water

Cool water

No disturbance of birds during peak of heat

Midnight feed/water

Proper ventilation

Genetics

Formulation

Maintain CP level in feed

Increase synthetic amino acid level in the feed

Energy: Carbohydrates \rightarrow Fat (but keep same energy level)

High digestible raw materials

Maximize salt level (attention litter moisture)

Electrolyte balance

Pellet quality

Feed additives

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Phytogenic feed additives

The world of plant ingredients



Effect of Rosemary on liver histopathology in Japanese quail

- TN: 22°C for 24 h/d. For heat-stress: 34°C for 8 h/d, 22°C 16 h/d
- Relative humidity was approximately 60–65%

	HS		TN			P- Statistical significance			
	Rosemary oil, mg/kg		Rosemary oil, mg/kg			Main effects			
Lesions	o	125	250	o	125	250	SEM	Environmental conditions	Amount of feed additives
Kupffer cell activation	1.71 ^a	0.860 ^b	1.71 ^a	0.148 ^B	0.364 ^{AB}	1.00 ^A	0.216	P < 0.05	P < 0.05
Fatty degeneration	2.45 ^a	0.715 ^c	2.04 ^b	0. 423 ^B	0.712 ^B	1.57 ^A	0.181	P < 0.05	P < 0.05
Apoptotic body	1.71 ^a	0.000 ^b	2.14 ^a	0.000 ^B	0.000 ^B	0.861 ^A	0.100	P < 0.05	P < 0.05
Bile pigment accumulation	1.28 ^a	0.365 ^b	0.862 ^b	0.000 ^B	0.000 ^B	0.713 ^A	0.103	P < 0.05	P < 0.05
Extramedullary hematopoiesis	2.43 ^a	1.00 ^b	2.43 ^a	1.00 ^B	1.00 ^B	1.57 ^A	0.195	P < 0.05	P < 0.05
Hepatocyte apoptotic index	12.5 ^b	7.63 ^c	19.1 ^a	2.57 ^B	3.29 ^B	10.7 ^A	0.681	P < 0.05	P < 0.05

Effect of Ginger powder on laying rate

Parameters	Time/ week	NC	НС	H1
Daily feed intake	1–3 w	89.69 ± 6.29^{ab}	83.21 ± 4.00^{a}	85.05 ± 7.09^{a}
-	4–6 w	95.41 ± 4.08^{a}	77.35 ± 9.84^{b}	88.74 ± 5.95^{a}
	7–9 w	107.94 ± 3.76^{a}	90.41 ± 3.27^{b}	94.84 ± 2.24^{b}
	1–9 w	$97.68 \pm 9.08^{\text{b}}$	83.66 ± 8.15 ^d	89.54 ± 6.63 ^c
Laying rate (%)	1–3 w	$75.97 \pm 2.24^{\rm a}$	66.45 ± 5.95^{b}	$69.05 \pm 6.84^{\mathrm{b}}$
	4–6 w	74.35 ± 1.24^{a}	65.73 ± 2.38^{b}	70.63 ± 2.38^{c}
	7–9 w	$73.65 \pm 9.54^{\rm ac}$	65.62 ± 1.75^{b}	70.03 ± 1.78^{ab}
	1–9 w	74.66 ± 5.26^{a}	$65.93 \pm 3.62^{\text{b}}$	$69.90 \pm 4.30^{\circ}$
Feed/Egg	1–3 w	2.44 ± 0.24	2.51 ± 0.37	2.35 ± 0.33
	4–6 w	2.36 ± 0.06	2.27 ± 0.28	2.49 ± 0.58
	7–9 w	2.60 ± 0.28	2.51 ± 0.21	2.51 ± 0.30
	1–9 w	2.38 ± 0.26^{ab}	2.43 ± 0.30^{ab}	2.45 ± 0.41^{ab}
Average egg weight (g)	1–3 w	54.66 ± 1.55^{a}	$50.75 \pm 1.79^{\circ}$	52.43 ± 2.33^{bc}
	4–6 w	54.81 ± 2.27^{ac}	50.49 ± 2.29^{ab}	49.94 ± 0.75^{b}
	7–9 w	55.41 ± 1.94^{a}	53.85 ± 2.23^{ab}	53.16 ± 1.26^{b}
	1–9 w	56.35 ± 2.77	51.70 ± 2.53^{b}	51.84 ± 2.06^{b}
Egg culling rate (%)	1–3 w	0.84 ± 1.69^{a}	4.25 ± 2.92^{b}	2.22 ± 2.33^{ab}
	4–6 w	2.33 ± 2.57	$1.80~\pm~1.49$	3.89 ± 2.75
	7–9 w	0.43 ± 0.87	0.37 ± 0.74	1.56 ± 2.97
	1–9 w	$1.20 ~\pm~ 1.94^{a}$	$3.68~\pm~3.45^{\rm b}$	2.56 ± 2.73^{ab}

Effect of ginger powder, Chinese herbal medicine on production performance of laying hens.

Note: Values with different lowercase letters significant difference indicate significant difference (P < 0.05).

Effect of Artemisia on blood pH in broilers

• Cyclic heat stress: 34°C for 8 h/day; 22 °C for 16 h/day)



Effect of Artemisia on performance

Table 2 Effects of enz	zymatically treated	Artemisia annua	L. on growth perform	nance of broilers rea	red under heat stre	S
Item	Treatment+					
	Control	HS	HS-EA _{0.75}	HS-EA _{1.00}	HS-EA _{1.25}	
BWG, g	1452 ^a	1285 ^c	1339 ^{bc}	1389 ^{ab}	1375 ^{ab}	19.04
FI, g C:E. ala	2094 0.540 ^a	2041 0.506 ^b	2282 0.510 ^{ab}	2030 0.527 ^{ab}	202 <i>5</i> 0.524 ^{ab}	19.30 0.007
Carcass yield, %	75.28 ^a	70.51 ^c	72.02 ^{bc}	73.46 ^{ab}	73.23 ^{ab}	0.660
Abdominal lat, %	1.389	1.434	1.397	1.410	1.345	0.037

Effect on Thyme and Squaw Mint on Antioxidant status

• Cyclic heat stress: 32°C for 8 h/day



Pirmohamammadi et al. (2016)

Selecting phytogenics candidates for heat stress evaluation

Evaluation and validation of candidates

- Step 1: Survival rate of nematode *C. elegans*
- Step 2: Gene expression in *C. elegans* for HSP-transcription factors
- Step 3: Gene expression in Caco-2 cells for HSP
- Step 4: *In vivo* validation

In vitro evaluation of heat stress: C. elegans survival rate

- Free-living nematode, soil dwelling
- Easy to manage



Length: 1 mm, Diameter: 65 µm

1 day	3 days	2 days	7-10 days	
Egg	4 Larval stages	Adult		
	Incubated at 20°C	Add test substance 37°C		
			Survival rate evaluated every 30 min until all worms dead	
			Measured via increase in SYTOX Green fluorescense	

Step 1: EO Blend and C. elegans survival



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Increased survival rate calculated from surface area:

- 7975 minutes
- = 15.8% increase relative to control

Step 1: Screening of test substances and *C. elegans* survival

Product	Dose (mg/L)	Survival rate increase (%)
Vitamin C	250	10.9
Vitamin E	150	5.1
Betaine	500	11.3
EO Blend	25	15.8
Flavonoid Blend	250	4.2
Capsaicin oleoresin	500	0.6

Step 2: *C. elegans* and HSP transcription factors

Unstressed: DAF-16 in in the cytosol

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Heat stress: DAF-16 in nucleus where genes are up-regulated for synthesis of HPS70





Delacon Internal Study

Step 3: Heat-stressed Caco-2 cells



Step 4: In vivo validation phytogenic combinations

- Heat stress: cyclic (8h at 32°C; 12h at 26°C); from d21-42 of age
- Diet: Corn/Wheat based diet Betaine positive control
- Birds: 576 broilers (Ross 308)
- Duration: 42 days
- Design: 6 reps, 32 birds each
- Performance negative control: BWG: 2543 g; FCR: 1.767; Mort.: 3.4%



* Essential oil, flavonoids & pungent substances (Biostrong® Comfort) Delacon internal study

Step 4: In vivo validation phytogenic combinations

- Heat stress: cyclic (12h at 35°C; 12h at 24°C) from d21-42 of age
- Diet: 3-Phase corn/soy-based diet
- Birds: 600 broilers (Cobb 500)
- Duration: 42 days

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Design: 4 reps, 25 birds each



* Essential oil, flavonoids & pungent substances

Delacon internal study

Summary

- Thermal stress can have deleterious impacts upon production performance
- Phytogenic feed additives are one strategy to manage heat stress
- Significant body of research supports phytogenics efficacy
- Screening and validation methodologies deliver targeted, efficacious products
- Phytogenic combinations can prove highly effective e.g. combinations of essential oils, flavonoid and pungent substances
- Globally, it's only going to get hotter!



Keeping cool: mitigating the impact of heat stress in poultry using phytogenic feed additives