

Effects of Inclusion Level of Black Soldier Fly Larvae Protein or Oil on Broiler Growth Performance During Heat Stress

T. Veldkamp, A. Rezaei Far, J.J. Mes, S. Naser El Deen, P.G. van Wikselaar, I. Fodor, H. Chen



Introduction (1)

- High environmental temperatures have negative effects on health, welfare and production of poultry.
- Heat stress is major source of systemic oxidative stress.
- Antioxidants can be beneficial to protect against adverse health effects of heat stress.

Introduction (2)

- Insect oil originating from BSFL is a fat source high in MCFA, particularly lauric acid and can decrease the host response to inflammation and are candidates to use in feed to counteract the effects of heat stress (Santos et al. 2019, Chen et al. 2022).
- No *in vivo* trials have been performed yet to test the effect of insect products in broiler diets in heat stress conditions.
- Therefore, insect meal and insect oil was included in a dose response study with 0, medium (5% meal; 2% for oil) and high (10% meal; 4% for oil) inclusion, with and without heat stress challenge in a 2 x 2 x 3 factorial design.

Trial Design

Factorial design

2

x

2

x

3

Treatment	Heat stress Challenge	Insect product	Inclusion Level
1	Control	BSFL protein	0%
2			5%
3			10%
4		BSFL fat	0%
5			2%
6			4%
7	Heat stress	BSFL protein	0%
8			5%
9			10%
10		BSFL fat	0%
11			2%
12			4%



Trial Design



648 birds, 72 Experimental Units, Ross 308 male chicks



Day 0 to day 34



**Cargill Innovation Center
Velddriel, The Netherlands
2 rooms**

Tr	Pen & Block	Pen & Block	Tr
3	9 - A	10 - A	10
9	8 - A	11 - A	7
5	7 - A	12 - A	6
2	6 - A	13 - B	6
1	5 - A	14 - B	12
4	4 - A	15 - B	5
8	3 - A	16 - B	4
12	2 - A	17 - B	10
11	1 - A	18 - B	8

Tr	Pen & Block	Pen & Block	Tr
11	27 - C	28 - C	9
1	26 - C	29 - C	4
6	25 - C	30 - C	10
3	24 - B	31 - C	7
2	23 - B	32 - C	2
9	22 - B	33 - C	3
7	21 - B	34 - C	5
1	20 - B	35 - C	8
11	19 - B	36 - C	12

Tr	Pen & Block	Pen & Block	Tr
2	72 - F	55 - E	4
12	71 - F	56 - E	7
3	70 - F	57 - E	5
5	69 - F	58 - E	8
6	68 - F	59 - E	9
8	67 - F	60 - E	3
10	66 - F	61 - F	1
9	65 - F	62 - F	4
11	64 - F	63 - F	7

Tr	Pen & Block	Pen & Block	Tr
2	54 - E	37 - D	2
11	53 - E	38 - D	11
10	52 - E	39 - D	4
6	51 - E	40 - D	12
12	50 - E	41 - D	3
1	49 - E	42 - D	8
10	48 - D	43 - D	9
7	47 - D	44 - D	6
5	46 - D	45 - D	1



Trial timeline



Day 0

Start study
BW



Day 9

Start heat stress
Feed Change
BW
Feed weight



Day 21

BW
Feed weight



Day 34

BW
Feed weight
Carcass Yield

Common Starter diet

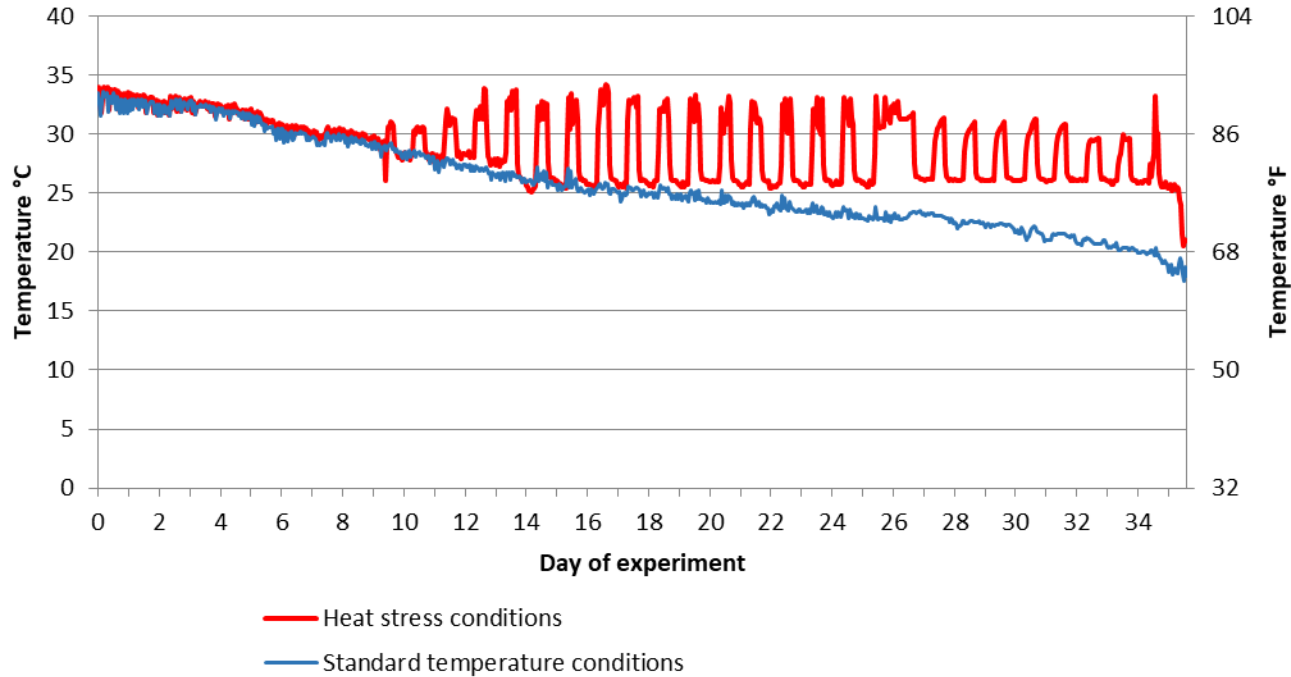
Experimental Grower diets

Basal and two experimental diets with highest inclusion level of BSFL protein and BSFL fat

	Basal	10% BSFL protein	4% BSFL fat
Ingredients (%)			
Soybean meal	34.22	18.49	34.22
Maize	30.82	33.13	30.82
Wheat	25.00	25.00	25.00
Sunflower meal	0.50	1.76	0.50
Rapeseed meal	0.50	5.00	0.50
BSFL protein	0.00	10.00	0.00
Soybean oil	6.00	3.92	2.00
BSFL fat	0.00	0.00	4.00
Limestone	1.38	1.07	1.38
Monocalcium phosphate	0.26	0.33	0.26
Sodium chloride	0.24	0.24	0.24
Sodium bicarbonate	0.22	0.22	0.22
Methionine DL	0.21	0.19	0.21
Lysine HCL	0.10	0.12	0.10
L-Threonine 98	0.03	0.01	0.03
Natugrain enzyme	0.010	0.010	0.010
Natuphos	0.005	0.005	0.005
Premix	0.500	0.500	0.500
Nutrients (g/kg)			
Crude protein	221.2	221.2	221.2
Dry matter	87.7	88.5	87.7
Calcium	6.8	6.8	6.8
Sodium	1.6	1.6	1.6
Chloride	2.0	2.0	2.0
Available phosphorus	3.1	3.1	3.1
ME broiler (kcal)	2950	2950	2950
SID Arginine	13.31	11.91	13.31
SID Isoleucine	8.22	8.14	8.22
SID Leucine	15.53	15.27	15.53
SID Lysine	11.00	11.00	11.00
SID Meth+Cys	8.00	8.00	8.00
SID Threonine	7.00	7.00	7.00
SID Tryptophan	2.35	1.70	2.35
SID Valine	8.95	9.72	8.95

SID values of BSFL were calculated based on Mwaniki et al. (2019)

Realized room temperature in heat stress conditions and standard temperature conditions

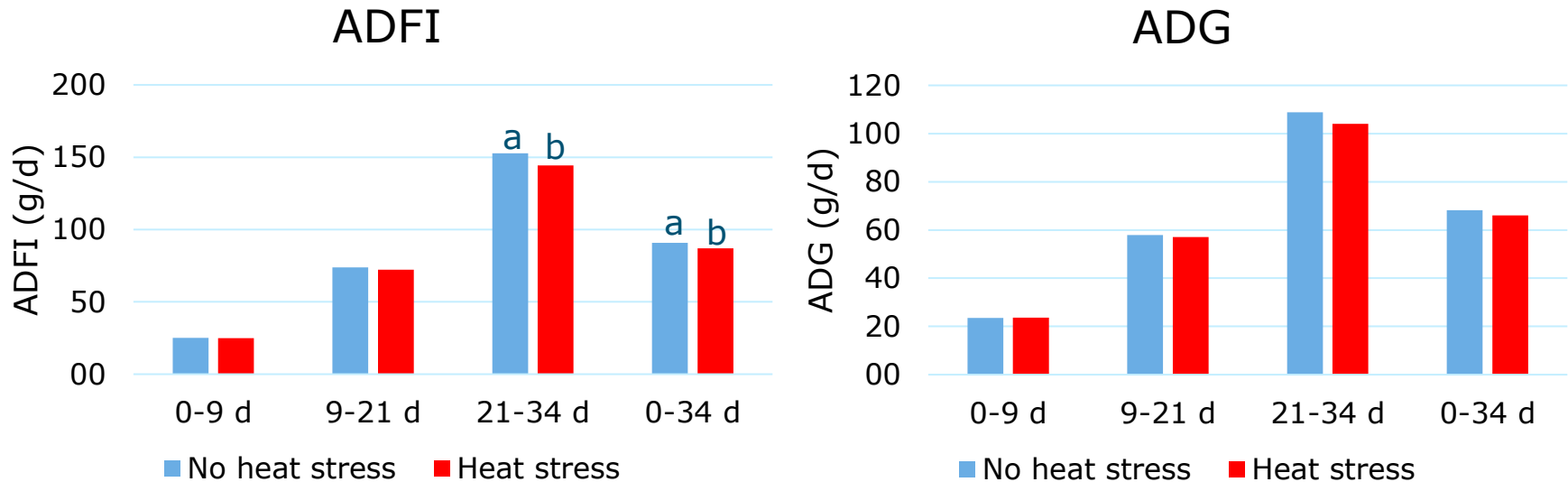


Effects of heat stress challenge on biomarkers in Ross 308 broilers at 35 d of age

Heat stress	No	Yes	SEM	P-value
Heat shock protein-70 (ng/mL)	0.92	0.96	0.31	0.934
Corticosterone (ng/mL)	1.70	0.69	0.31	0.085
C-reactive protein (ng/mL)	1067.1	865.7	236.7	0.580
Malondialdehyde (μ M)	283.1	321.8	43.3	0.562
Calprotectin (ng/mL)	81.9 ^b	124.0 ^a	9.3	0.002

- Ileum calprotectin levels were significantly elevated in broilers exposed to heat stress compared to those in neutral climate conditions
- Serum corticosterone levels showed a tendency to decrease in broilers under heat stress conditions, which contrasts with findings in literature

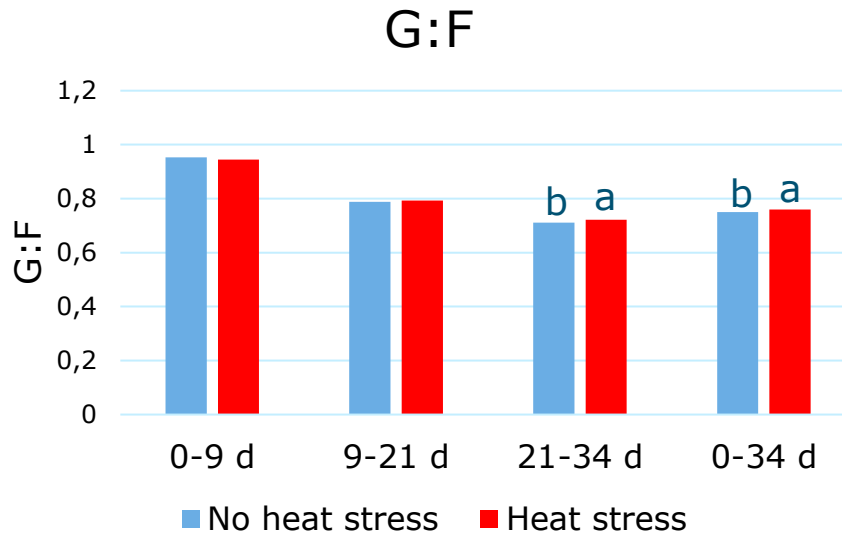
Effect of heat stress on daily feed intake and daily weight gain



- From d 9 to 21, no significant differences in ADFI were observed. Between d 21 and 34, ADFI in the heat stress room was significantly lower.
- ADG remained comparable between the heat stress and control rooms.

Different superscripts show significant difference $P < 0.05$

Effect of heat stress on Gain:Feed ratio



- Broilers in the heat stress room demonstrated a significantly improved G:F ratio between d 21 and 34

Different superscripts show significant difference $P < 0.05$

Effect of heat stress*insect product*inclusion level on daily feed intake and daily weight gain

Heat stress	No	No	No	No	No	No	Yes	Yes	Yes	Yes	Yes	Yes
BSFL product	Protein	Protein	Protein	Oil	Oil	Oil	Protein	Protein	Protein	Oil	Oil	Oil
Inclusion (%)	0	5	10	0	2	4	0	5	10	0	2	4
ADFI (g/d)												
d 9-21	73.0	77.9	71.3	73.6	72.7	74.1	73.1	70.8	73.1	71.8	72.3	72.8
d 21-34	155.3 ^{abc}	157.9 ^a	147.2 ^{abc}	156.4 ^{ab}	145.7 ^{abc}	154.1 ^{abc}	146.7 ^{abc}	142.6 ^c	142.5 ^c	143.1 ^{bc}	145.9 ^{abc}	145.0 ^{abc}
d 9-34	115.8 ^{bc}	119.5 ^b	110.8 ^{bc}	116.6 ^{bc}	110.2 ^{bc}	115.6 ^{bc}	111.4 ^{bc}	108.1 ^c	108.1 ^c	108.8 ^c	110.6 ^{bc}	110.3 ^{bc}
d 0-34	91.6 ^{bc}	94.4 ^b	87.9 ^{bc}	91.8 ^{bc}	87.4 ^{bc}	91.6 ^{bc}	88.6 ^{bc}	85.6 ^c	85.8 ^c	86.2 ^c	88.1 ^{bc}	87.6 ^{bc}
ADG (g/d)												
d 9-21	57.4 ^{ab}	61.1 ^a	54.5 ^b	59.2 ^{ab}	56.0 ^{ab}	59.3 ^{ab}	57.8 ^{ab}	56.5 ^{ab}	55.8 ^{ab}	56.9 ^{ab}	58.0 ^{ab}	57.4 ^{ab}
d 21-34	110.1	111.2	104.9	111.5	106.9	108.5	105.8	102.5	101.2	102.6	106.9	106.0
d 9-34	84.8 ^{ab}	87.1 ^a	80.7 ^{ab}	86.3 ^{ab}	83.2 ^{ab}	84.8 ^{ab}	82.7 ^{ab}	80.4 ^{ab}	79.2 ^b	80.6 ^{ab}	83.4 ^{ab}	82.7 ^{ab}
d 0-34	68.6	70.4	65.5	69.2	67.1	68.5	67.2	65.0	64.3	65.3	67.7	67.0
G:F (g/g)												
d 9-21	0.785	0.784	0.765	0.803	0.796	0.793	0.790	0.799	0.782	0.792	0.803	0.789
d 21-34	0.709	0.705	0.713	0.713	0.722	0.704	0.721	0.718	0.711	0.717	0.732	0.731
d 9-34	0.732	0.729	0.729	0.740	0.745	0.734	0.743	0.744	0.734	0.741	0.755	0.749
d 0-34	0.749	0.745	0.746	0.754	0.760	0.748	0.759	0.759	0.750	0.757	0.769	0.764
Mortality												
d 0-34	0.000	0.019	0.000	0.019	0.000	0.000	0.000	0.037	0.019	0.000	0.000	0.019
BW (g)												
d 9	254.8	257.2	253.4	257.4	252.0	252.4	259.5	254.6	252.3	244.9	258.7	259.9
d 21	943.3	995.5	907.9	963.1	945.8	963.9	953.0	933.2	921.9	934.6	955.3	948.9
d 34	2374.2 ^{ab}	2441.5 ^a	2271.8 ^{ab}	2412.0 ^{ab}	2335.1 ^{ab}	2385.7 ^{ab}	2348.0 ^{ab}	2265.4 ^{ab}	2239.0 ^b	2282.7 ^{ab}	2345.0 ^{ab}	2326.4 ^{ab}

- BSFL protein at 5 and 10% inclusion level and BSFL oil at 2 and 4% inclusion level did not exhibit notable benefits under mild cyclic heat stress conditions in Ross 308 broilers.
- Significant three-way interaction effects were detected between heat stress, insect product, and inclusion level after 21 d of age.
- Under thermoneutral conditions, ADFI and ADG decreased numerically at 2% BSFL oil inclusion and increased numerically at 4% BSFL oil inclusion relative to 2% BSFL oil inclusion. In contrast also under thermoneutral conditions, 5% BSFL protein inclusion led to a numeric increase in ADFI and ADG and led a numerically reduction in ADFI and ADG at 10% BSFL protein inclusion relative to 5% BSFL inclusion.

Effect of heat stress*insect product*inclusion level on carcass yields

Heat stress	No	No	No	No	No	No	Yes	Yes	Yes	Yes	Yes	Yes
Product	Protein	Protein	Protein	Oil	Oil	Oil	Protein	Protein	Protein	Oil	Oil	Oil
Inclusion (%)	0	5	10	0	2	4	0	5	10	0	2	4
BW (kg)	2.51	2.62	2.49	2.53	2.49	2.56	2.48	2.43	2.40	2.47	2.53	2.50
Carcass (%)	76.19	76.36	76.43	76.19	76.21	76.38	78.24	77.36	77.21	77.34	78.41	77.59
Breast (%)	33.48	33.04	32.90	33.81	32.95	33.41	31.78	32.12	31.84	31.96	32.26	32.31
Fat pad (%)	0.89	0.99	1.08	0.83	0.96	0.99	1.22	1.29	1.29	1.17	1.36	1.21

Carcass yields presented as percentage of live body weight and breast and fat pad presented as percentage of carcass weight.

- Heat stress resulted in a numerical increase in relative carcass yield, a numerical decrease in breast meat yield, and an increase in relative fat pad.

Different superscripts show significant difference $P < 0.05$

Conclusion

- Heat stress challenge was successfully applied (reduced ADFI, ADG, and final BW).
- Replacing traditional ingredients with BSFL-based products is feasible under both neutral and heat stress climate conditions.
- BSFL protein at 5 and 10% inclusion level and BSFL oil at 2 and 4% inclusion level did not exhibit notable benefits under mild cyclic heat stress conditions in Ross 308 broilers and no heat stress * insect products interaction effects have been observed for stress biomarkers.

Acknowledgement

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