Research Institute for Farm Animal Biology

Nutrient intake, growth performance, blood metabolites and bone properties of broilers fed a ration containing mineral-enriched black soldier fly larvae

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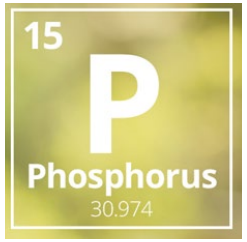
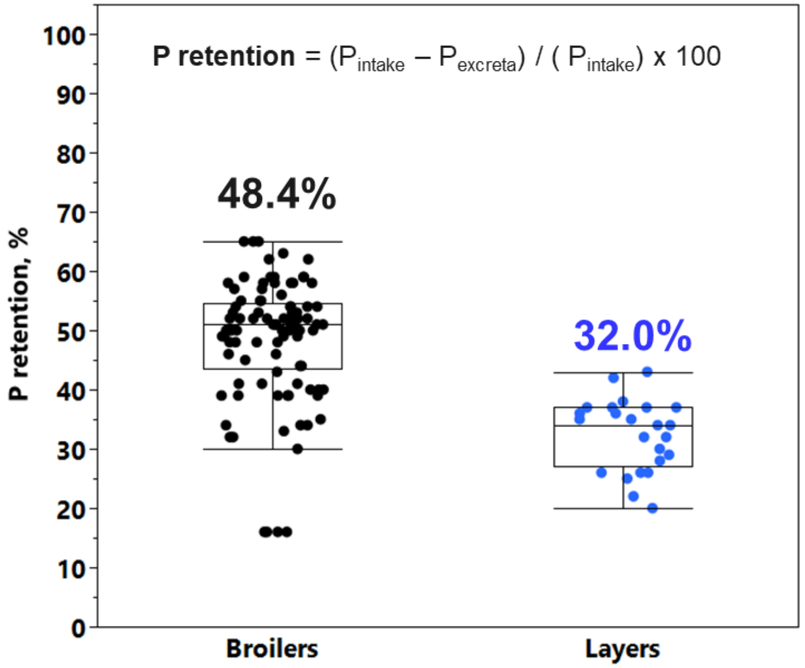
“Critical raw material” used in agriculture



- 90% of P derived from phosphate rock is used as fertilizer or livestock feed[1]
- Global rock phosphate resources are depleting
- Excessive use of P ➔ environmental concerns[2]
- To lower the P-losses through organic waste, P recycling is essential[3]
- Can **mineral recycling technology** and **black soldier fly larvae** help overcome these challenges?

Meta-analysis of P retention by chickens*

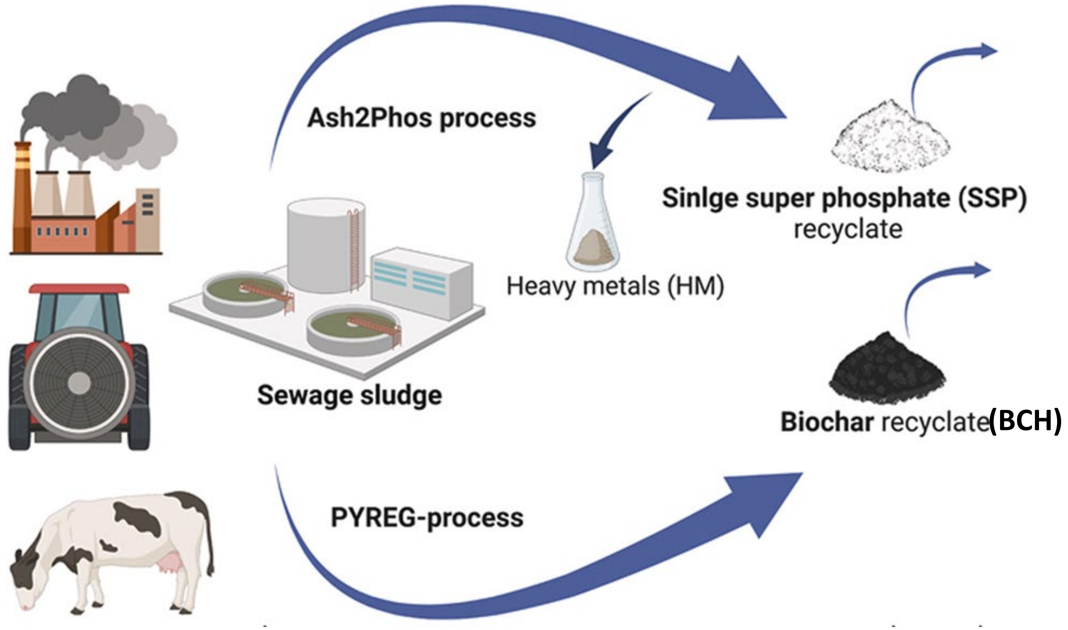


* Own illustration based on data provided by Bougouin et al., 2014

(1) Brunner, 2010.
(2) Liu et al., 2008.
(3) Bergfeldt et al., 2018.

New recycling technologies

Ash2®Phos process: recycling of sewage sludge by incineration[1]



PYREG®-process: pyrolysis of sewage sludge[2]

- An efficient recycling of minerals is possible
- Concerns about heavy metals in sewage sludge recyclates

	Recyclates	
	BCH	SSP
Macrominerals, g/kg DM		
Ca	52.4	208.4
P	65.9	88.8
Mg	7.3	1.8
Na	2.4	14.5
K	4.2	2.1
Heavy metals, mg/kg DM		
Mn	490.0	59.5
Fe	141,064	430
Zn	462.9	8.9
Cu	1,643	23.4
As	3.0	1.3
Cd	<0.02	<0.02
Pb	87.0	1.9
Hg	< 0.02	< 0.02

Color codes
Green : desirable
Red : undesirable

(1) Cohen et al., 2019, International Fertilizer Society
(2) Fesharaki and Rath, 2018, Verkohlungsanlage Pyreg. European Patent Office Patent no. EP 3 358 253 A1

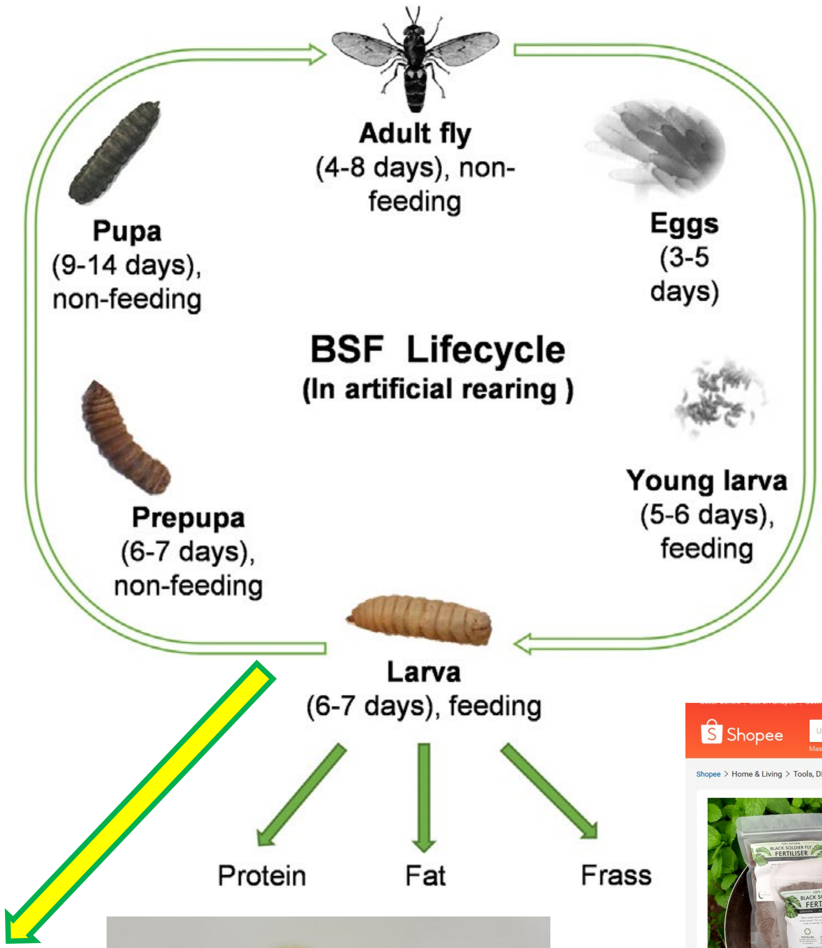
Larvae of black soldier fly (BSFL)[1]


Adult fly

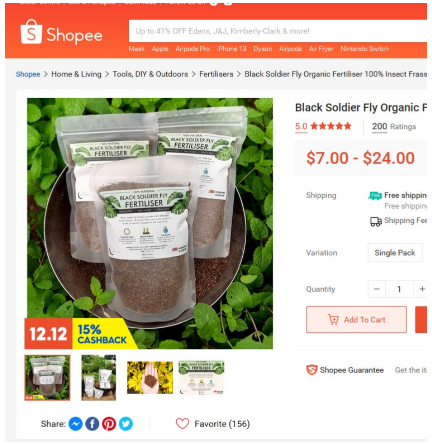

Harvested larvae

- Recycle and upgrade low-grade organic biomass
- Can grow on a large spectrum of feeding substrates
- Classical intention of BSFL rearing:
  - N-recycling: alternative to soya and fishmeal
  - Frass (faeces + residues) as plant-fertilizer
- **Suitable for mineral recycling?**

(1) Seyedalmoosavi et al., 2022. https://doi.org/10.1186/s40104-022-00682-7



Minerals?

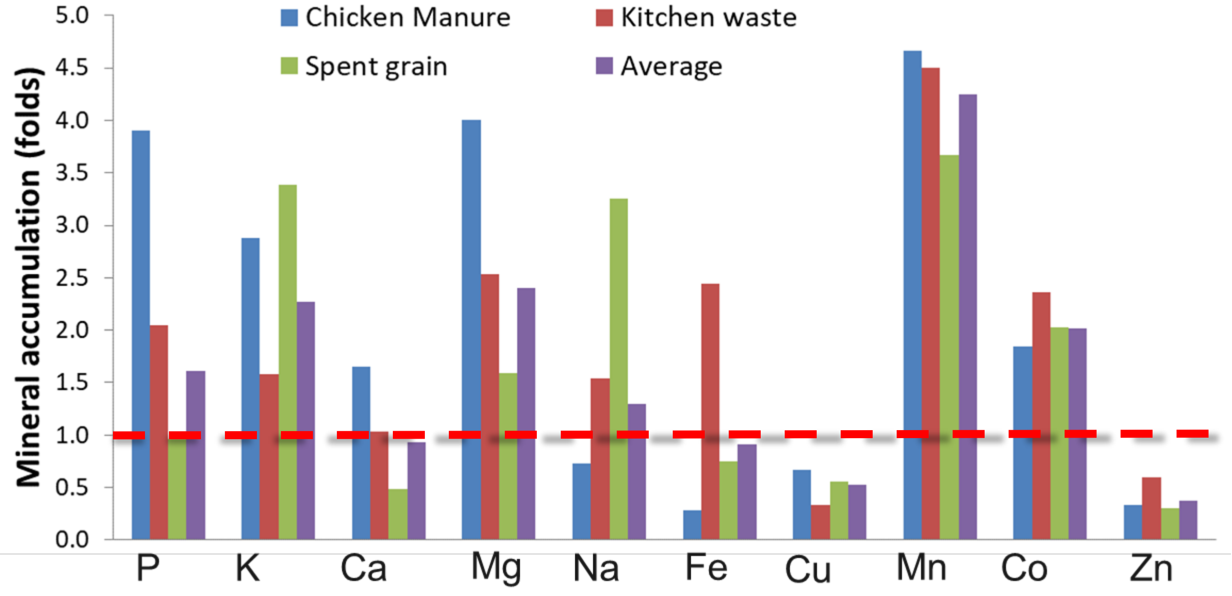


BSFL accumulate certain minerals

>1 = accumulation

1 = no accumulation

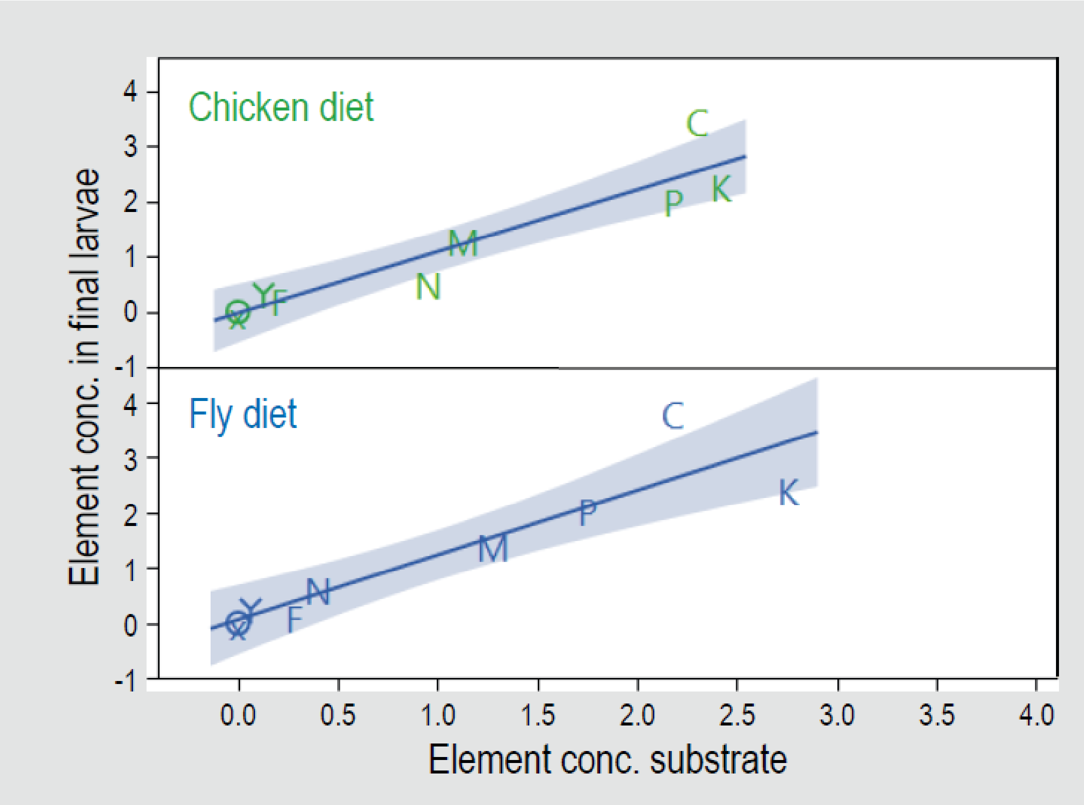
<1 = Depletion



Modified after Shumo et al., 2019, Scientific Reports, 9(1), 1-13.

- Each letter-symbol represents the relationship between average concentrations of a given element in the substrate and in the final larvae.
- Element concentrations shown on both axes are on a log-scale, i.e. [ln (g + 1) / kg dry matter)].

Element	Symbol
Calcium	C
Cadmium	x
Iron	F
Potassium	K
Magnesium	M
Manganese	Y
Sodium	N
Phosphorus	P
Lead	O



Das et al., 2023. *Journal of Insects as Food and Feed*
https://doi.org/10.3920/JIFF2023.0021

Overall hypothesis and objectives

Hypothesis

- ✓ BSFL can accumulate minerals from sewage sludge recylates and mineral-enriched BSFL can be fed to broilers

Objectives

- ✓ How much unprocessed BSFL can be fed to broilers?
- ✓ To what extend BSFL can accumulate minerals from sewage sludge recylates?
- ✓ Impact of mineral-enriched BSFL on broiler performance, blood metabolites and bone properties

Experiments



Experiment 1	Experiment 2	Experiment 3
To what extent whole BSFL can be included in broiler diets?	How much recycled P and minerals can BSFL accumulate from different sources?	What are the effects of feeding broilers with mineral-enriched BSFL?

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Effects of increasing levels of whole Black Soldier Fly (*Hermetia illucens*) larvae in broiler rations on acceptance, nutrient and energy intakes and utilization, and growth performance of broilers

M.M. Seyedalmoosavi *, M. Mielenz *, S. Görs *, P. Wolf †, G. Daş *, C.C. Metges *

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Research article

Upcycling of recycled minerals from sewage sludge through black soldier fly larvae (*Hermetia illucens*): Impact on growth and mineral accumulation

Mohammad M. Seyedalmoosavi [a], Manfred Mielenz [a], Kai Schleifer [a], Solvig Görs [a], Petra Wolf [b], Jens Tränckner [c], Liane Hüther [d], Sven Dänicke [d], Gürbüz Daş [a], Cornelia C. Metges [a,*]

Journal of Insects as Food and Feed, 2023; 9(5): 583-598
Wageningen Academic Publishers

Lipid metabolism, fatty acid composition and meat quality in broilers supplemented with increasing levels of defrosted black soldier fly larvae

M.M. Seyedalmoosavi[1], D. Dannenberger[2], R. Pfuhl[2], S. Görs[1], M. Mielenz[1], S. Maak[2], P. Wolf[3], G. Daş[1*] and C.C. Metges[1]

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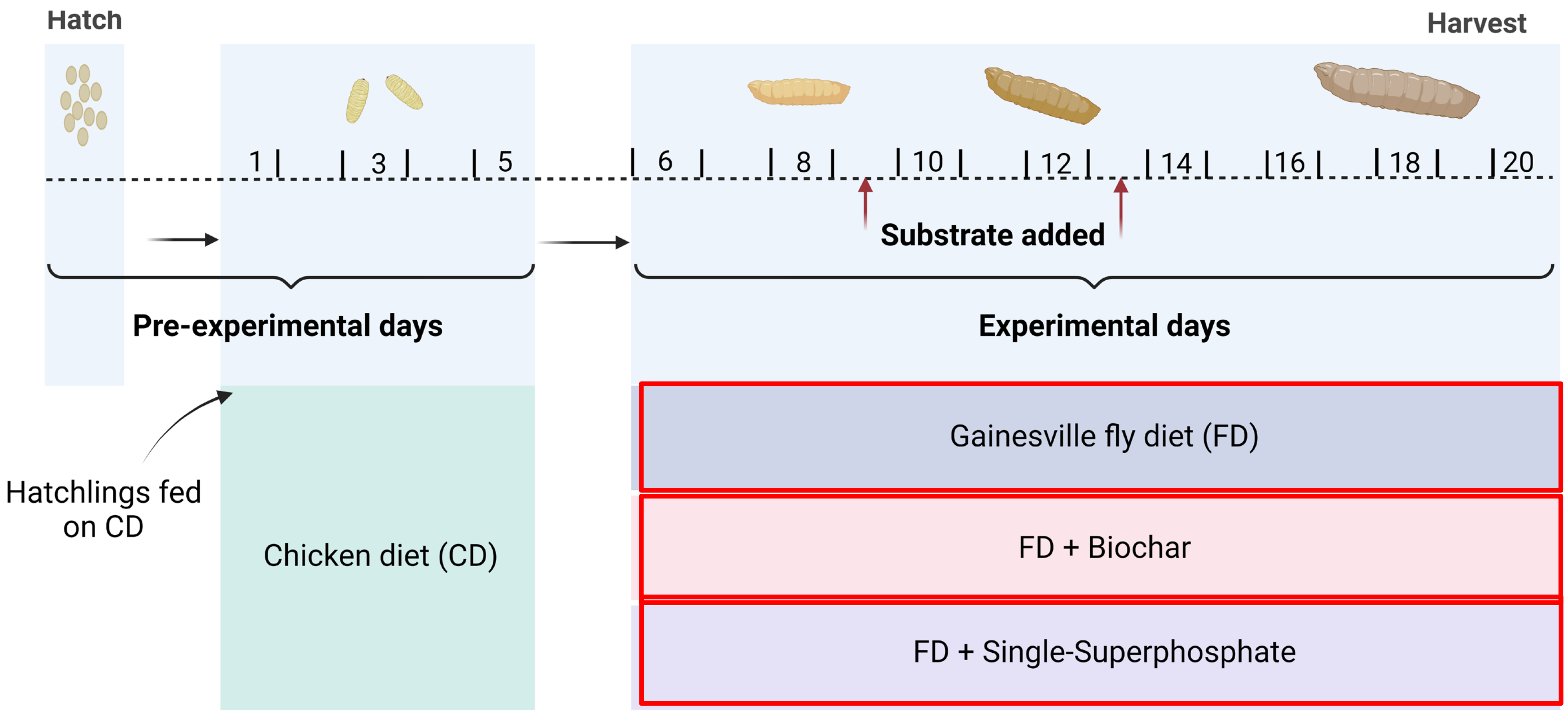
BSFL feeding experiment with sewage sludge recylates (Exp-2)



Day 5 Isolation of larvae from the residual CSF substrate and frass

Distribution of larvae to container (approximately 8,000 larvae/container; n = 6/substrate)

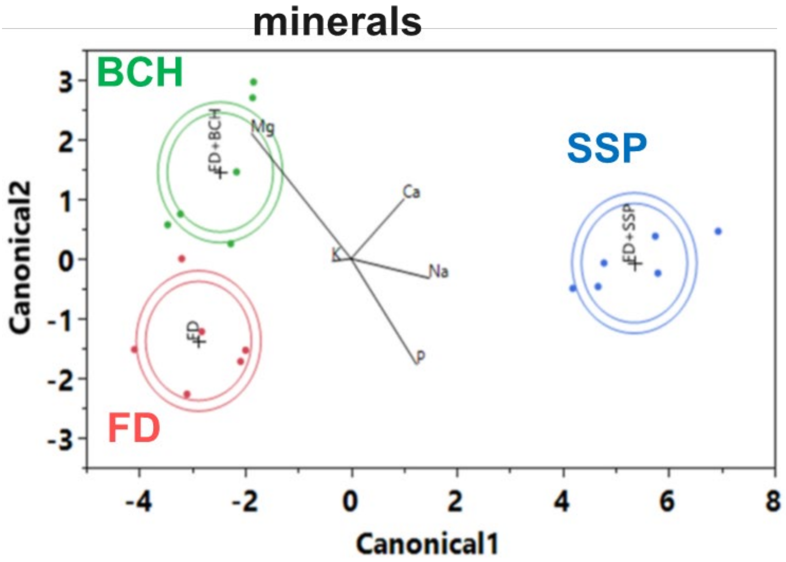
Random allocation of larvae to experimental diets

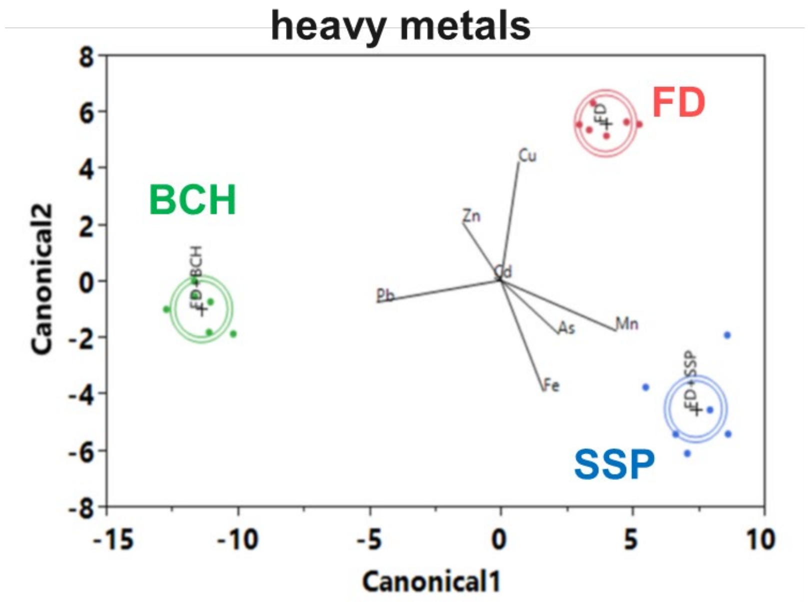
Randomization of boxes on racks

Differentiation of macrominerals and heavy metal patterns in BSFL

	Feeding substrates				P-values , ≤	
	FD	FD + BCH	FD + SSP	SE	T	B
Macrominerals, g/kg DM						
Ca	45.77^{c}	53.24^{b}	64.59^{a}	1.22	0.001	0.022
P	7.14^{b}	7.24^{b}	10.89^{a}	0.39	0.001	0.035
Mg	3.31^{b}	3.63^{ab}	3.96^{a}	0.12	0.005	0.015
Na	0.94^{b}	0.94^{b}	1.27^{a}	0.03	0.001	0.212
K	10.89^{b}	10.01^{b}	14.46^{a}	0.74	0.005	0.071
Heavy metals, mg/kg DM						
Mn*	285	304	303	7.46	0.167	0.108
Fe	150^{b}	557^{a}	339^{ab}	74.4	0.006	0.248
Zn	$95.8^{\dagger}$	$107.4^{\dagger}$	98.5	3.6	0.087	0.047
Cu	$9.92^{\dagger}$	$12.22^{\dagger}$	10.14	0.67	0.052	0.085
As	0.047^{b}	0.066^{b}	0.115^{a}	0.006	0.001	0.340
Cd*	0.63^{b}	0.77^{a}	0.74^{ab}	0.037	0.045	0.147
Pb	0.23^{c}	1.16^{a}	0.54^{b}	0.050	0.001	0.201
Hg	0.000	0.000	0.001	–	–	–

* Above EU limits for livestock feed





<u>Color codes</u>

Green : desirable

Red : undesirable

Broiler feeding experiment with mineral enriched BSFL (Exp-3)

15% of CON feed intake as BSFL

CON no BSFL; ad libitum feed	CON +FD-BSFL **(L-FD)**	CON+BCH-BSFL **(L-BCH)**	CON+SSP-BSFL **(L-SSP)**

N = 80 birds (Ross 308)
N = 20 birds/group (6 pens / group)

d0 | BW wk1 | BW wk2 | BW wk3 | BW wk4 | BW wk5 | BW wk6

Starter | **Grower** | **Finisher**

Pen based measurements

Nutrient intakes

Weekly body weight

Individual measurements

- Plasma
- Bone

Feed intake and growth performance of broilers fed mineral-enriched BSFL

	Dietary treatment groups[1]					P-values[2] (≤)		
	CON	L-FD	L-BCH	L-SSP	SE	G	W	G × W
Intakes (g/wk)								
Feed	629	472	498	469	73.7	0.392	0.001	0.396
Larvae	–	91.9	91.8	91.8	–	–	–	–
Fresh matter	629	564	589	561	73.7	0.905	0.001	0.847
Dry matter	570	454	478	451	66.8	0.563	0.001	0.589
Growth performance								
Initial BW (g)	48.75	49.00	47.88	48.92	0.026	0.750	–	–
Average BW (g)	1011	779	809	780	176	0.750	0.001	0.621
CV of BW (%)	30.07	44.75	34.51	46.78	8.92	0.496	0.001	0.301
Feed conversion ratios								
FCR-1 (FMI/BWG)	1.87	2.26	2.15	2.47	0.320	0.618	0.201	0.668
FCR-2 (DMI/BWG)	1.67	1.79	1.74	1.92	0.220	0.884	0.070	0.599

Energy and nutrient conversion, and mineral and heavy metal intakes of broilers fed mineral-enriched BSFL

	Dietary treatment groups[1]					P-values[2] (≤)		
	CON	L-FD	L-BCH	L-SSP	SE	G	W	G × W
Nutrient and energy intake								
Crude protein (g/wk)	145	121	127	119	16.12	0.669	0.001	0.643
Crude fat (g/wk)	26.43	25.80	26.59	24.03	26.43	0.916	0.001	0.547
Crude ash (g/wk)	46.59	38.94	41.60	39.85	6.01	0.806	0.001	0.796
ME (MJ/wk)	7.24	6.85	7.08	6.61	0.84	0.954	0.001	0.827
CP:ME intake ratio	20.04[a]	17.63[b]	17.89[b]	18.01[b]	0.22	0.001	0.001	0.038
PCR (g CP/100 g BWG)	47.5	53.0	51.3	56.7	6.77	0.812	0.064	0.676
ECR (MJ ME/100 g BWG)	2.17	2.78	2.62	2.95	0.40	0.561	0.168	0.689
Mineral intake (g/wk)								
Ca	9.55	8.42	9.10	8.85	1.33	0.941	0.001	0.815
P	3.63	2.90	3.06	2.98	0.420	0.611	0.001	0.656
Mg	1.29	1.06	1.12	1.07	0.147	0.640	0.001	0.659
Na	1.04	0.81	0.85	0.81	0.132	0.579	0.001	0.531
K	6.36	5.04	5.30	5.10	0.698	0.521	0.001	0.543
Heavy metal intake (mg/wk)								
Mn	83.50	62.94	66.35	62.47	10.31	0.448	0.001	0.422
Fe	199	150	159	149	25.69	0.489	0.001	0.421
Zn	80.81	60.95	64.25	60.48	10.03	0.453	0.001	0.421
Cu	12.81	9.65	10.17	9.57	1.56	0.436	0.001	0.417
As	0.180	0.137	0.146	0.137	0.026	0.621	0.001	0.285
Cd	0.049	0.038	0.039	0.037	0.006	0.428	0.001	0.445
Pb	0.092	0.069	0.073	0.069	0.012	0.496	0.001	0.306

Plasma metabolites, enzymes and immunoglobulins in broilers fed mineral enriched BSFL

	Dietary treatment groups[1]				SE	P-value (≤)
	CON	L-FD	L-BCH	L-SSP		Group
Metabolites						
Albumin (g/L)	12.41	12.37	12.20	13.07	0.347	0.270
Cholesterol (mmol/L)	3.00	3.33	3.26	3.29	0.177	0.492
Glucose (mmol/L)	13.92	14.27	14.40	14.13	0.271	0.596
NEFA (μmol/L)	207.2	276.7	292.6	283.0	28.68	0.117
Triglyceride (mmol/L)	0.826	1.16†	1.00	0.803†	0.108	0.057
Uric acid (μmol/L)	353.1	448.2	415.6	390.7	54.22	0.610
ALP (U/L)	1749	2943	3088	2839	550	0.261
P (mmol/L)	1.94[a]	1.56[ab]	1.42[b]	1.63[ab]	0.123	0.022
Ca (mmol/L)	2.74[b]	3.21[a]	3.22[a]	3.08[ab]	0.129	0.026
Mg (mmol/L)	0.936	0.979	0.989	0.992	0.031	0.508
Immunoglobulins						
IgY (mg/mL)	1.79	1.30	1.33	1.42	0.163	0.107
IgM (mg/mL)	0.201	0.200	0.202	0.196	0.017	0.994
IgA (mg/mL)	0.224	0.220	0.209	0.253	0.021	0.464

Tibia characteristics and bone mineral status in broilers fed mineral enriched BSFL

	Dietary treatment groups[1]					*P*-value (≤)
	CON	L-FD	L-BCH	L-SSP	SE	Group
Diameter (cm)	0.907	0.792	0.887	0.883	0.055	0.462
Length (cm)	9.44	8.44	9.08	9.25	0.380	0.282
Weight (g)	11.33	9.14	10.19	11.26	1.18	0.511
Strength (N)	281.5	226.0	258.1	267.3	32.19	0.654
Ash (% DM)	52.49	49.02	50.64	47.94	1.82	0.324
P (in ash) (% DM)	18.59	18.50	18.40	17.83	0.542	0.739
Ca (in ash) (% DM)	36.34	36.89	34.01	34.23	2.19	0.728

Conclusion (Exp-3)

- ✓ 15% of CON feed intake can be provided broilers as whole BSFL
- ✓ 15% mineral-enriched BSFL in broiler diets did not affect nutrient intakes, tibia statues and tibia mineral composition
- ✓ SSP recycled-mineral enriched whole BSFL may be included in broiler diets
  - ✓(P and C replacement instead of supplementation?)

Caution

- ✓ Large within group variations
- ✓ Potential heavy metal accumulation in broiler meat was not investigated

Growth, nutrient uptake, blood metabolites and bone properties in broilers consuming feed with mineral-enriched whole black soldier fly larvae

in Journal of Insects as Food and Feed

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Thank you for your attention!

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Chemical analysis of experimental substrates and recyclates

Exp- 2

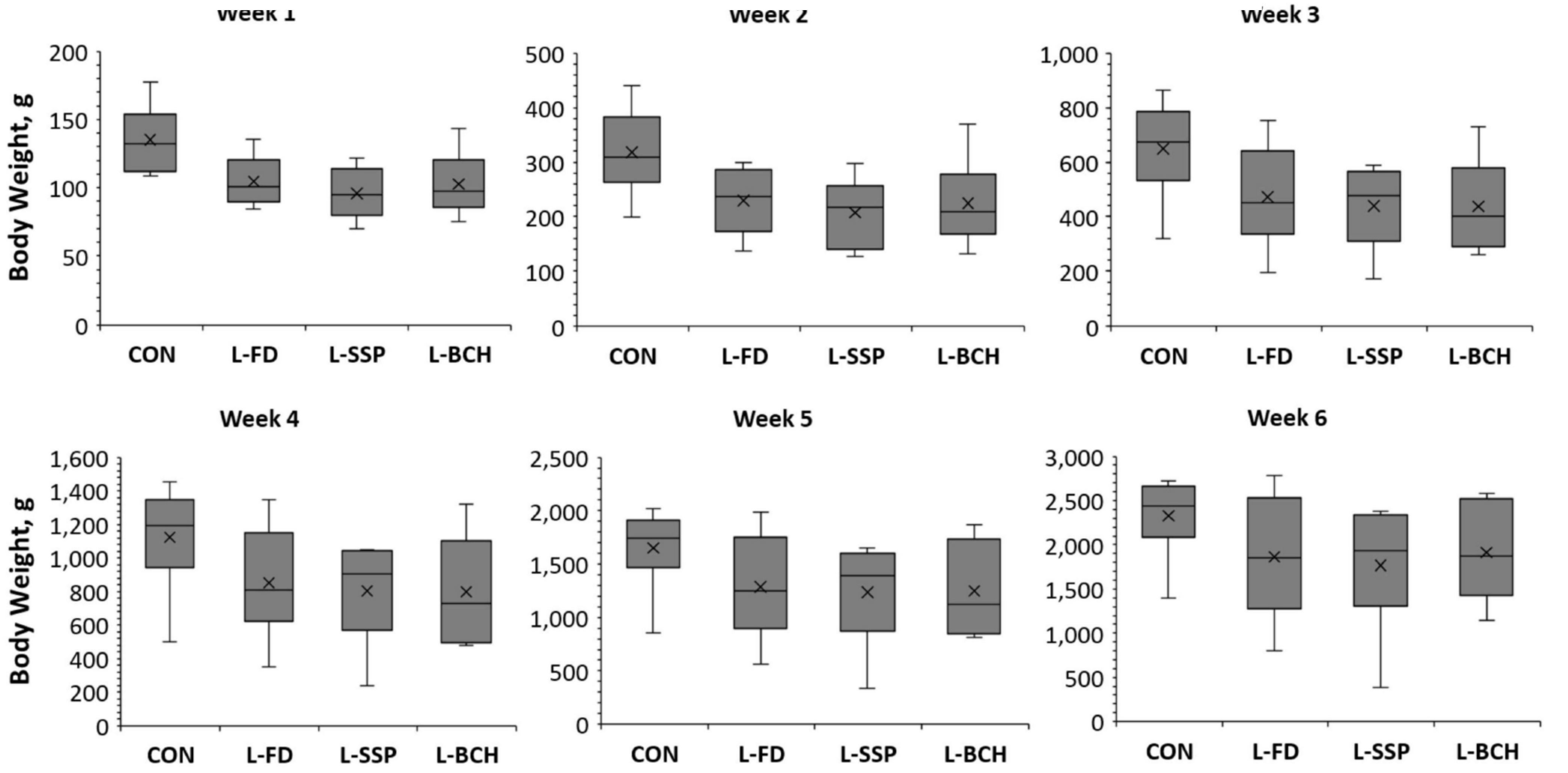
Ingredients	Feeding substrates				Recyclates	
	FD	FD+BCH	FD+SSP		BCH	SSP
Modified Gainesville diet, % FM						
Lucerne	9	9	9		-	-
Sugar beet pulp	7.5	7.5	7.5		-	-
Corn meal	6	5.4	5.4		-	-
Wheat bran	7.5	6.9	6.9		-	-
Biochar (BCH)	0	1.2	0		-	-
Single Superphosphate (SSP)	0	0	1.08		-	-
Water	70	70	70.12			
Chemical composition, g/kg DM						
Dry matter	854	856	856		-	-
Crude ash	77.5	75.9	75.9		-	-
Crude protein	153.7	147.3	147.3		-	-
Crude fat	31.8	29.7	29.7		-	-
Crude fibre	174.6	171.1	171.1		-	-
Starch	201.0	182.0	182.0		-	-
NDF	395.0	382.0	382.0		-	-
ADF	216.0	211.3	211.3		-	-
ME, MJ/kg DM	7.9	7.4	7.4		-	-
Gross energy, MJ/kg DM	10.9	10.2	10.2		-	-
Macro minerals, g/kg DM						
Ca	7.9	9.9	16.9		52.4	208.4
P	4.8	7.0	8.3		65.9	88.8
Mg	2.6	2.8	2.6		7.3	1.8
K	14.9	14.6	14.5		4.2	2.1
Na	0.5	0.6	1.1		2.4	14.5
Heavy metals, mg/kg DM						
Mn	65.8	82.3	65.7		490.0	59.5
Fe	337.0	5,852.0	351.0		141,064	430
As	0.12	0.23	0.17		3.0	1.3
Cd	0.13	0.14	0.13		<0.02	<0.02
Pb	0.25	3.65	0.33		87.0	1.9
Hg	0.02	0.02	0.02		< 0.02	< 0.02

Chemical analysis of experimental feed

Exp- 2

	Basal diets		
	Starter (d 1-14)	Grower (d 15-28)	Finisher (d 29-42)
Ingredients, %			
Soybean meal 48%	36	34	35
Wheat	31	28	28
Maize	21.5	28	26.5
Barley	5	4	5
Linseed oil	3	3	3
Vit-Min. Premix	2.5	2.5	2.5
Oyster shells	1.0	0.5	0.0
Chemical analysis, g / kg DM			
Dry matter, g / kg	908	905	906
Crude ash	69.4	71.8	90.5
Crude protein	254.4	275.1	241.7
Crude fat	55.1	48.6	43.1
Crude fiber	30.8	25.4	25.4
Starch	383.3	369.1	406.2
aNDFom	11.0	12.2	11.0
ADFom	37.4	38.7	33.1
ME, MJ / kg DM	13.0	12.8	12.6
Minerals, g / kg DM			
Ca	12.44	12.27	20.42
P	6.61	6.41	6.29
Mg	2.20	2.43	2.21
Na	1.21	1.77	1.99
K	11.45	12.15	10.49
Heavy metals, mg/kg DM			
Mn	117.7	144.2	154.4
Fe	287.3	311.7	386.5
Zn	110.6	139.6	150.2
Cu	18.17	22.54	23.40
As	0.160	0.222	0.410
Cd	0.095	0.064	0.080
Pb	0.159	0.126	0.190
Hg	<0.003	<LLD	<0.003

Body weight development in broilers fed mineral enriched BSFL



Background

- ✓ Growing human population requires the sustainable use of resources
- ✓ Food security treats; climate change, biodiversity loss, plant and animal diseases (1)
- ✓ Insects have a great potential to substitute conventional protein sources (2)
- ✓ BSFL meal in broiler diets without adverse effect on growth performance (3, 4)
- ✓ BSFL meal production requires expensive feed processing technology
- ✓ Instead of meal form, whole BSFL could be included in broiler diets (5, 6, 7)
- ✓ Diets containing whole larvae are highly interesting for poultry (5)

1) IPCC AR6, 2022
2) Dörper et al., 2020. J. Insects as Food Feed.
3) Cullere et al., 2016. Animal
4) de Souza Vilela et al., 2021. Foods.
5) Moula et al., 2018. Anim. Nutr.
6) Ipema et al., 2020. Appl. Anim. Behav. Sci.
7) Bellezza Oddon et al., 2021. J. Anim. Physiol. Anim.

Background

Positive effects of using insect in poultry diet

- ✓ Functional feed additive for poultry; chitin and antimicrobial peptides [1, 2]
- ✓ Improvement of growth performance, intake and efficiency [3]
- ✓ Microbial community improvement [3]
- ✓ No detrimental effect on meat quality and sensory traits [4, 5]
- ✓ Animal behavior and welfare [6, 7]

1) Gasco et al., 2020. J Insects as Food Feed.
2) Józefiak and Engberg, 2017. J Anim Feed Sci.
3) Kierończyk et al., 2022. Anim. Nutr.
4) De Souza Vilela et al., 2021. Foods .
5) Popova et al., 2020. J Insects as Food Feed.
6) Ipema et al., 2020. Sci. Rep.
7) Pichova et al., 2016. Appl. Anim. Behav. Sci.

Background

Potential risks associated with using BSFL as feed

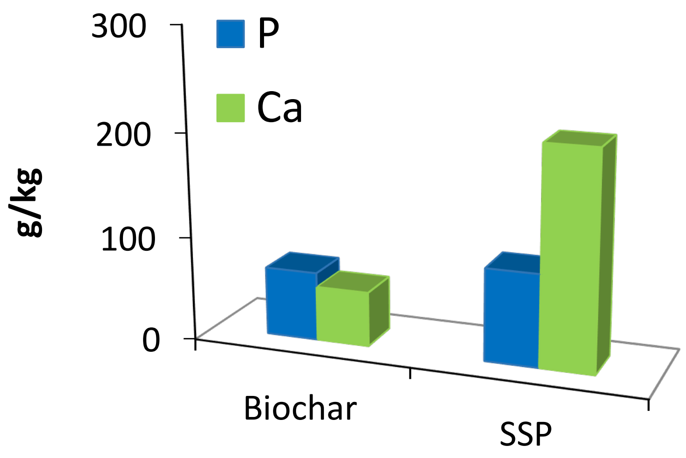
- ✓ BSFL convert **waste** into valuable biomass to improve nutrient cycle (6)
- ✓ Restrictions on BSFL farming and use of BSFL as animal feed in the EU (6, 7)
- ✓ European legislation prohibited waste for feeding BSFL (8)

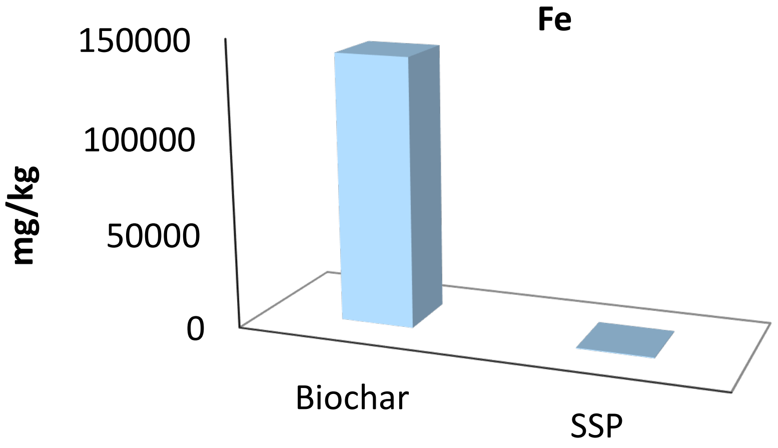
1) Lievens et al., 2021. J. Insects as Food Feed.
2) Lalander and Vinnerås, 2022. J. Insects as Food Feed.
3) European Commission, 2009.

Recyclates

✓ **Single Superphosphate (SSP)** originated from the recycling of sewage sludge produced by incineration using the Ash2®Phos process [1]

✓ **Biochar** derived from pyrolysis of sewage sludge produced by the PYREG®-process [2]





(1) Cohen et al., 2019, International Fertilizer Society (30 May 2019).
(2) Fesharaki and Rath, 2018, Verkohlungsanlage Pyreg. European Patent Office Patent no. EP 3 358 253 A1.

Publication

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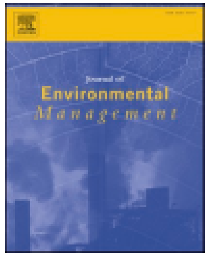

Research article

Upcycling of recycled minerals from sewage sludge through black soldier fly larvae (*Hermetia illucens*): Impact on growth and mineral accumulation

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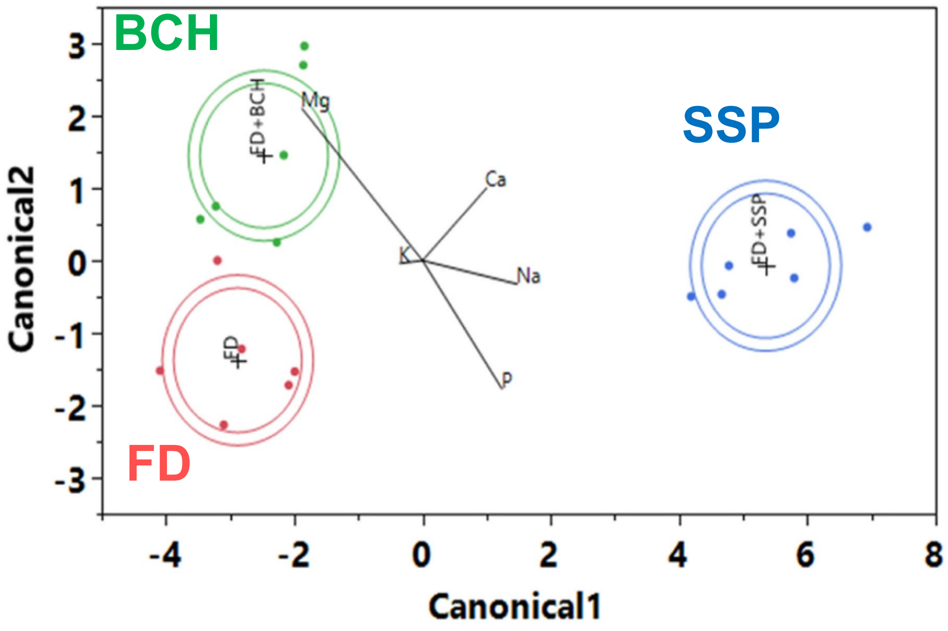
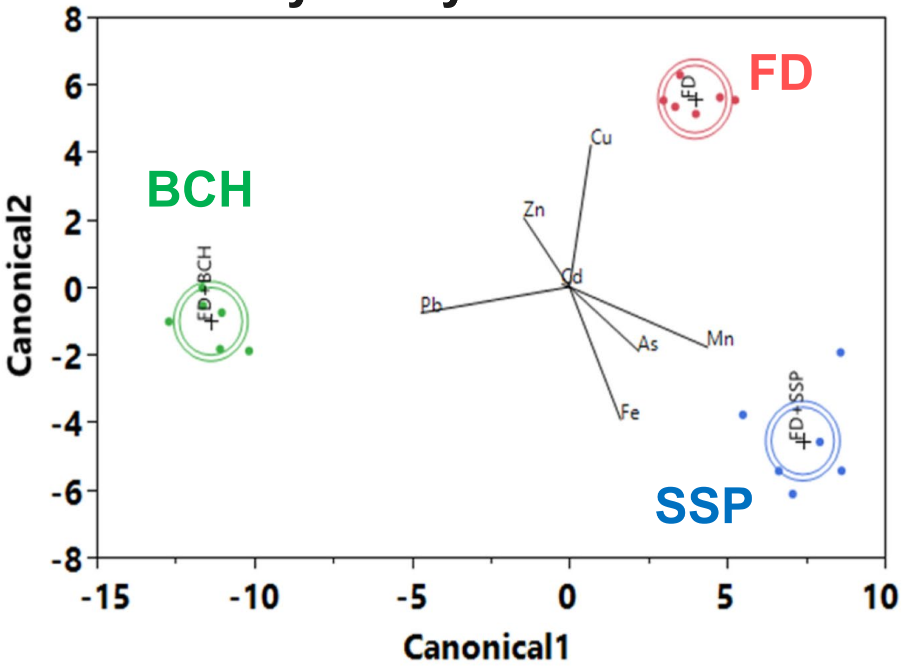
Scan Me

Linear discriminant analysis for overall differentiation in BSFL fed recyclate supplemented substrates

by minerals



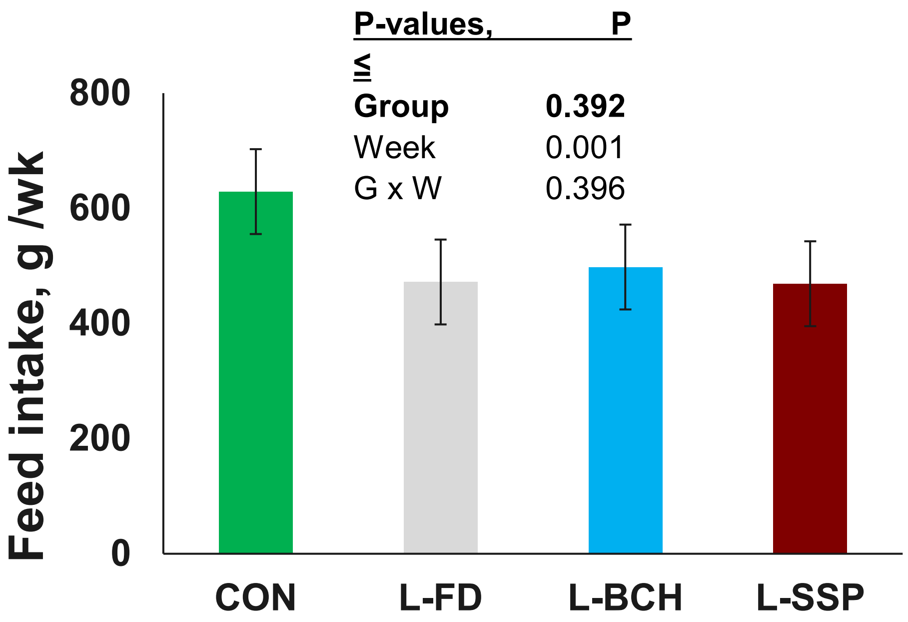
by heavy metals

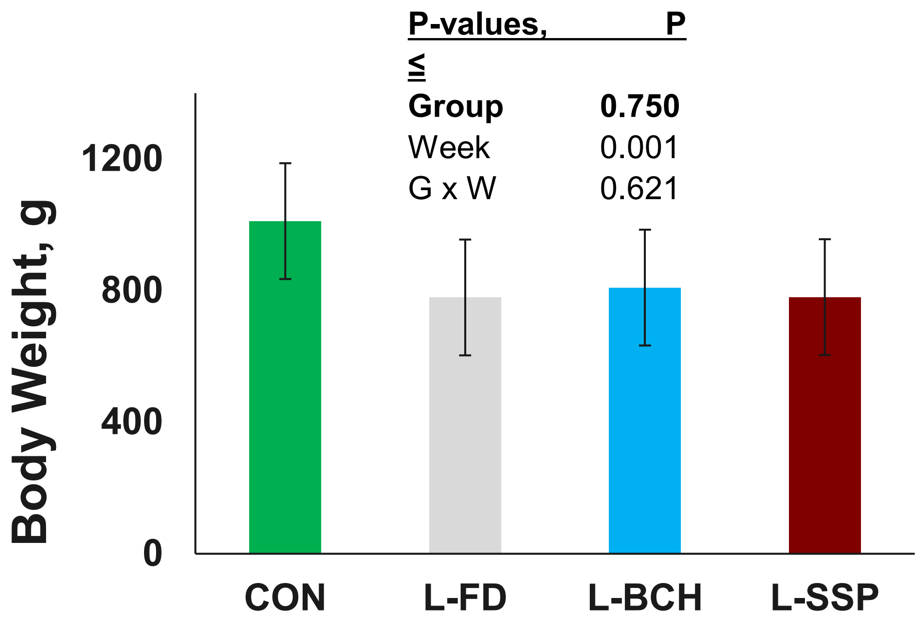


- SSP differentiated from FD mainly via P, Ca and Na
- BCH differentiated from FD mainly via Mg and P
- Except for Cd all heavy metals differentiated BCH and SSP from FD

Details of the analysis: Canonical plot of points and means (+) from linear discriminant analysis of larval concentrations of the macro-minerals with groups
All variables had the same units of measurement (i.e. mg/ kg DM).
Each dot represents the overall response of one container of BSFL fed on a specific substrate.
The inner circles represent the 95% confidence region for containing the true overall mean of the group, and the outer circles are the 50%

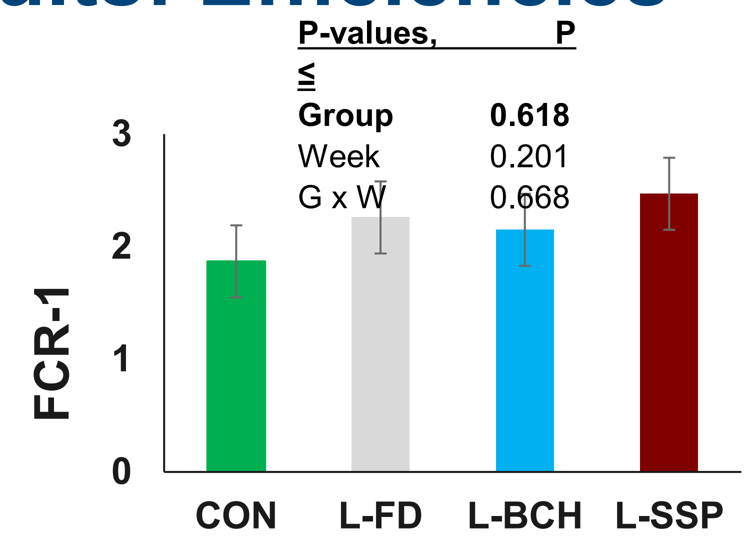
Results: Intake and Growth

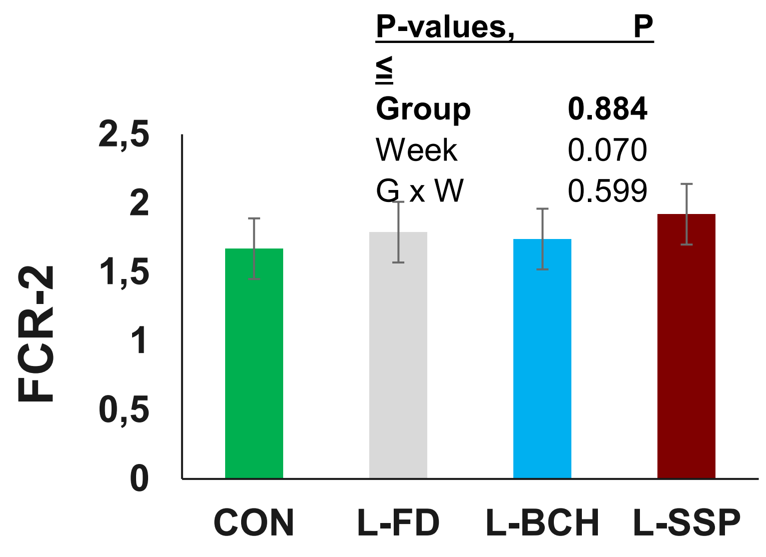


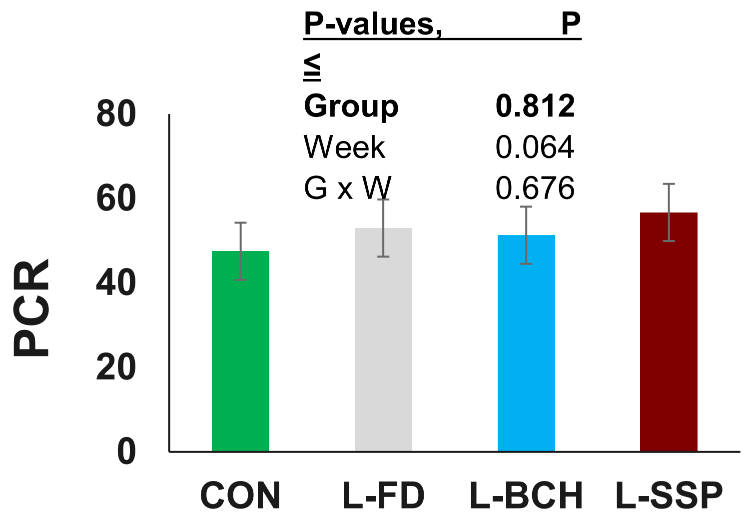


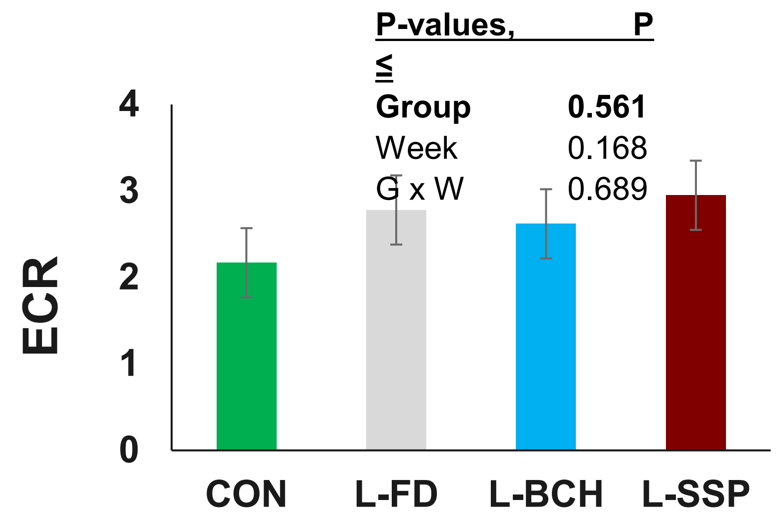
- Feed intake and growth of birds were not affected by the groups

Results: Efficiencies



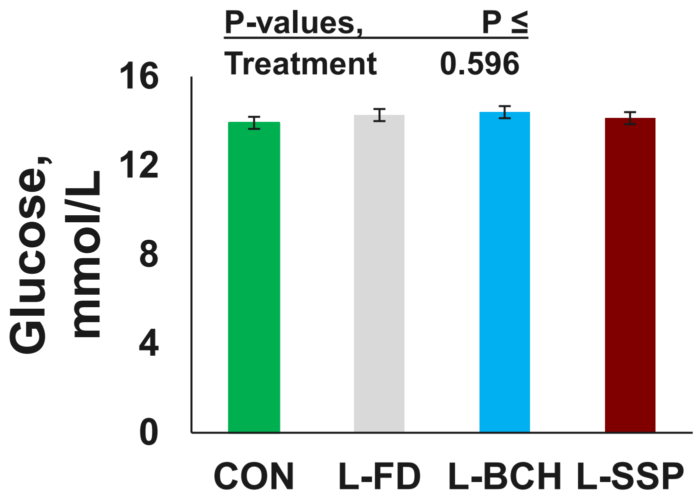


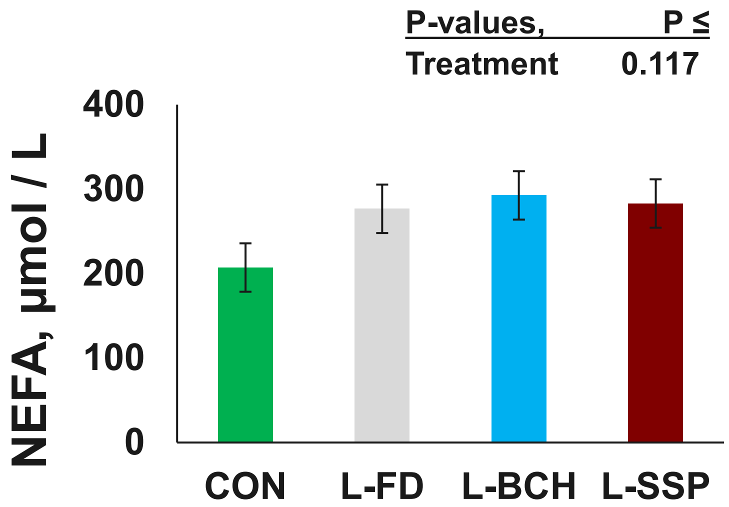


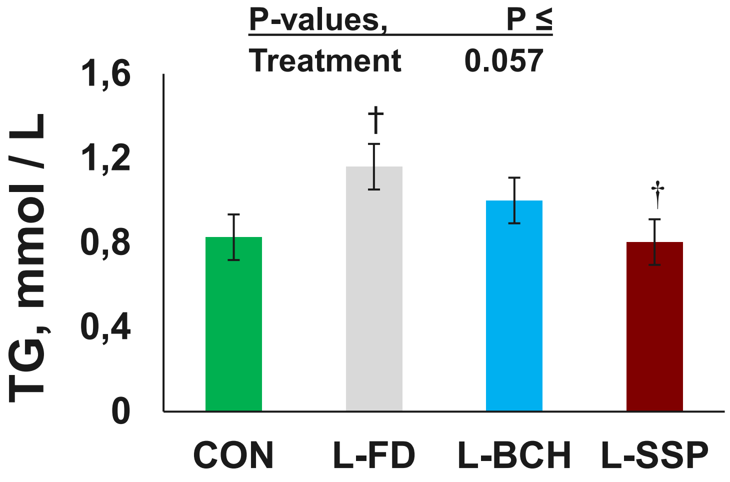


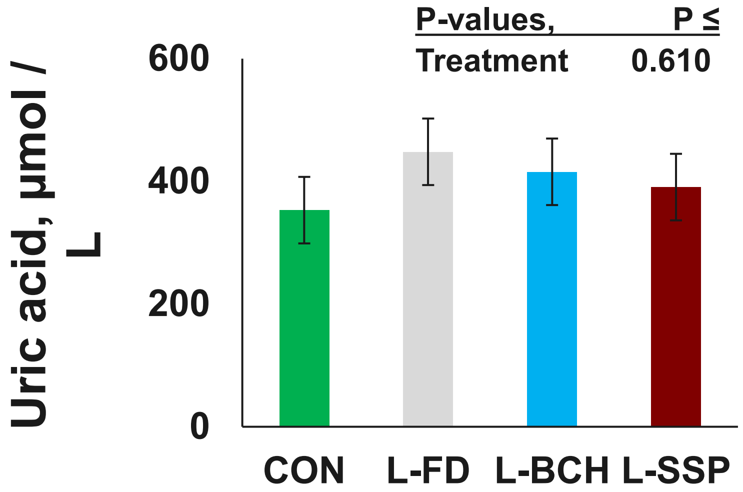
- **Feed, protein and energy conversion ratios were not affected**

Results: Metabolites



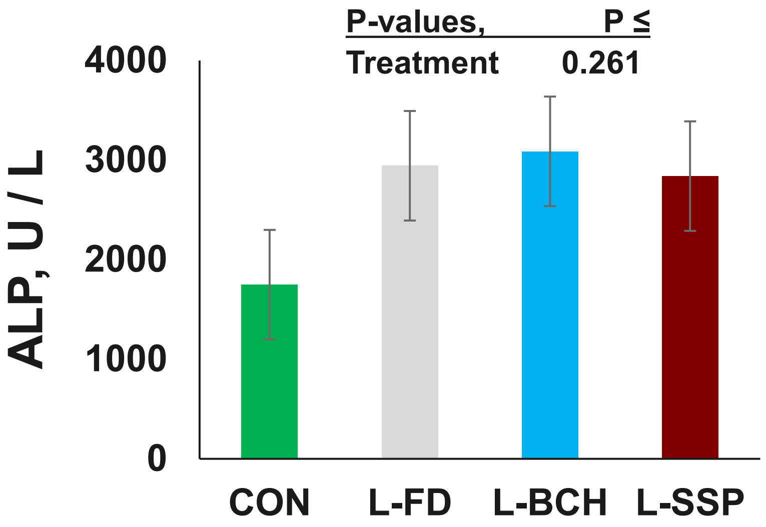


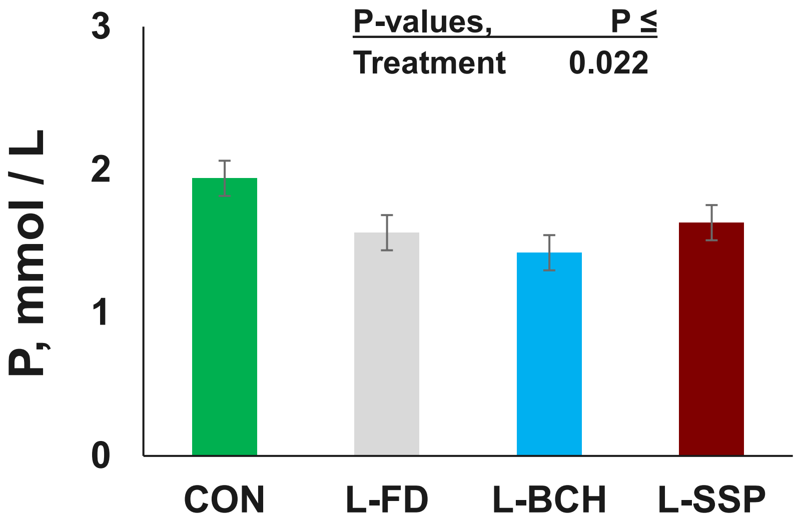


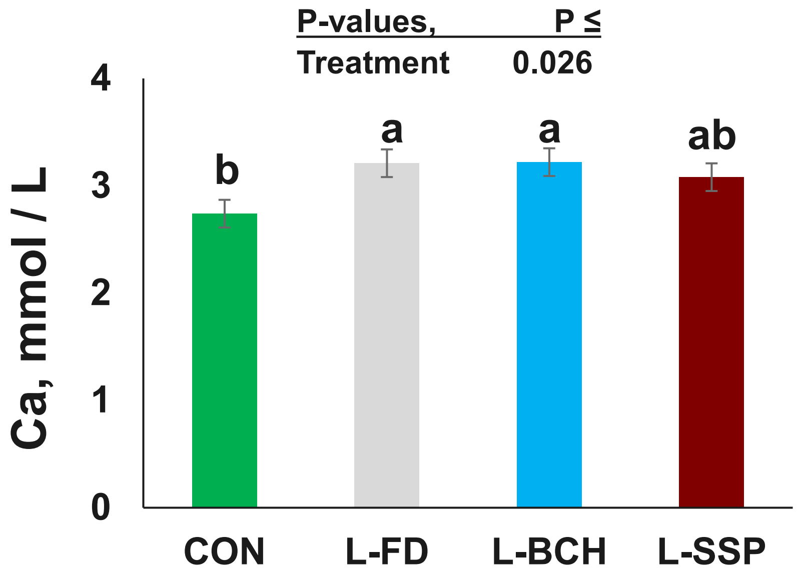


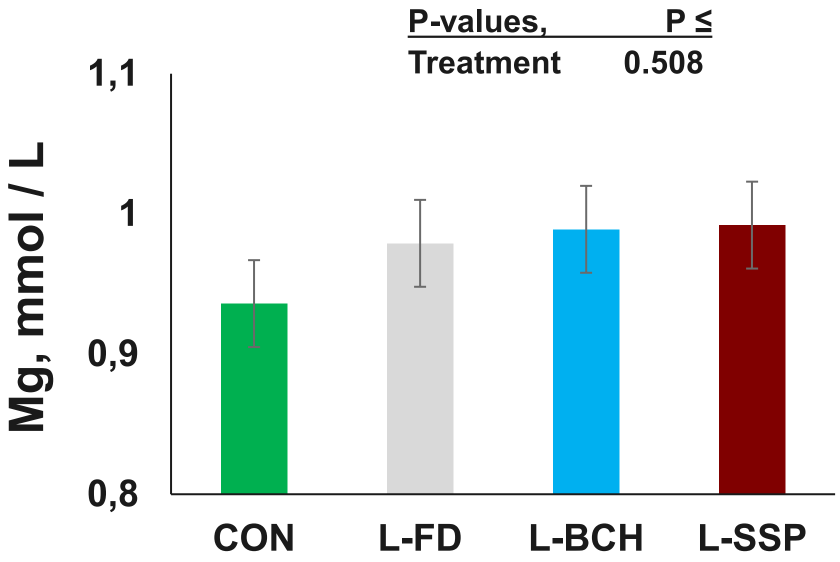
- Plasma glucose, NEFA and uric acid were not affected by groups
- L-FD tended to have higher triglyceride than L-SSP

Results: Metabolites



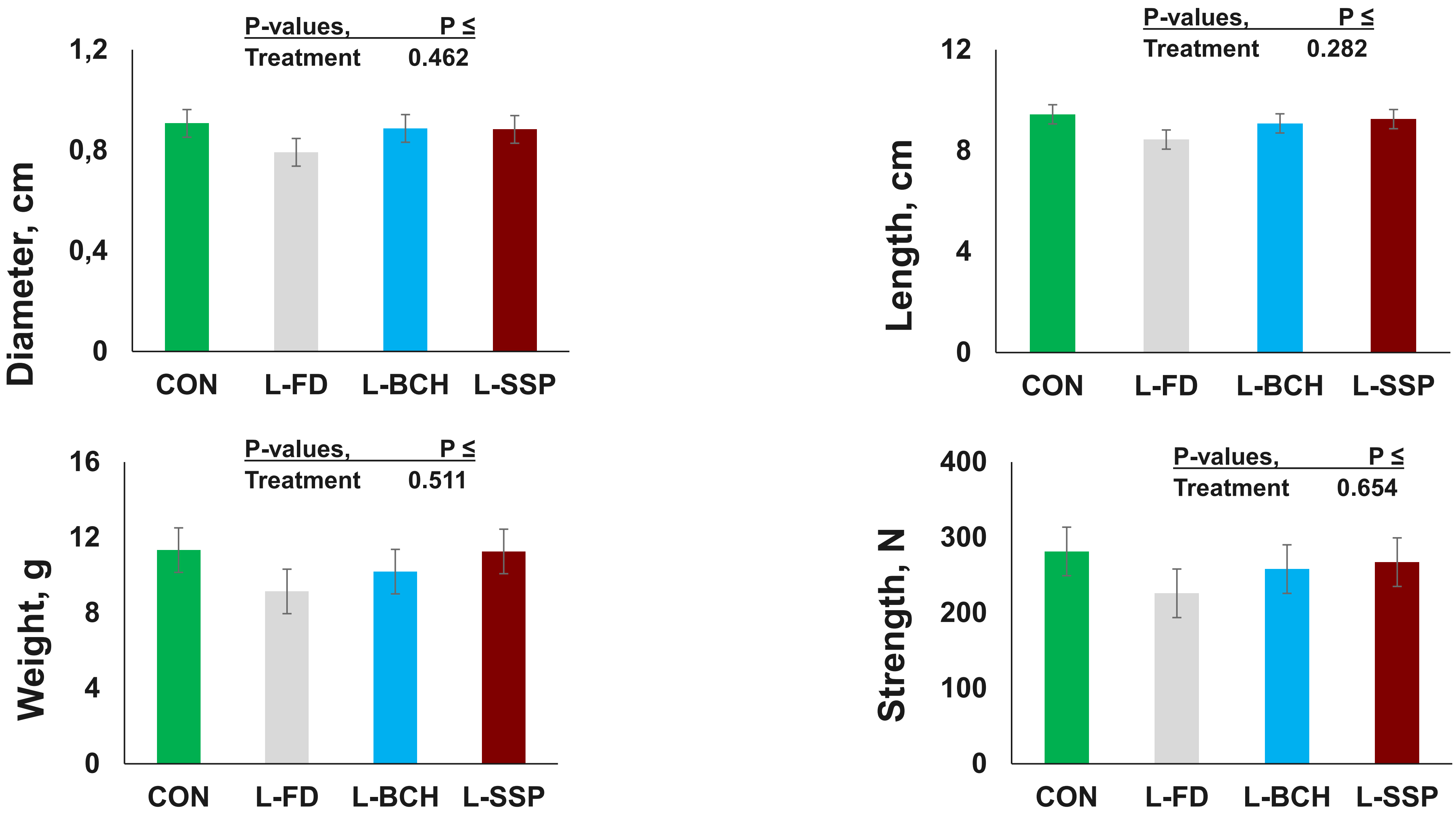






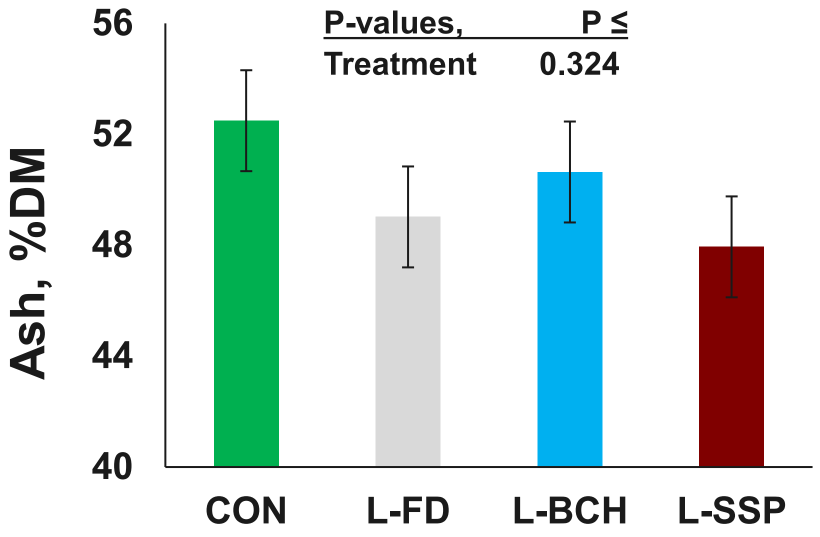
- Serum ALP, P and Ca were not affected by groups
- L-FD and L-BCH had higher serum Ca than CON

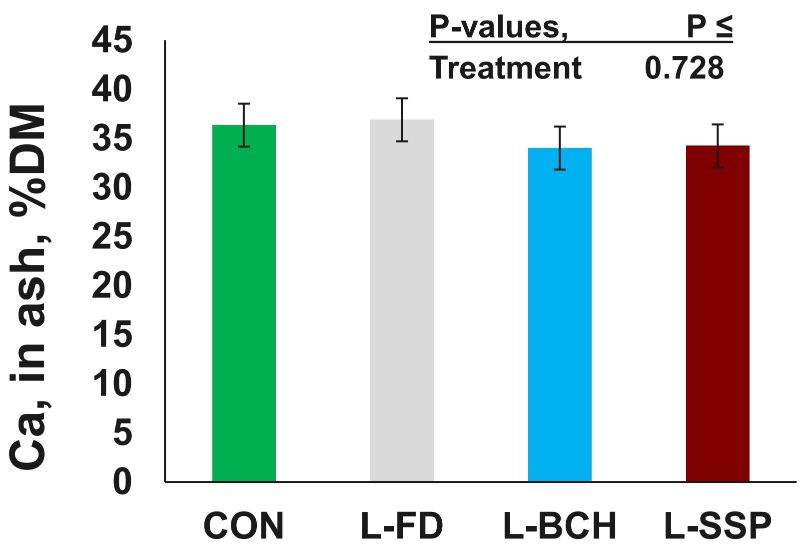
Results: Bone Characteristics

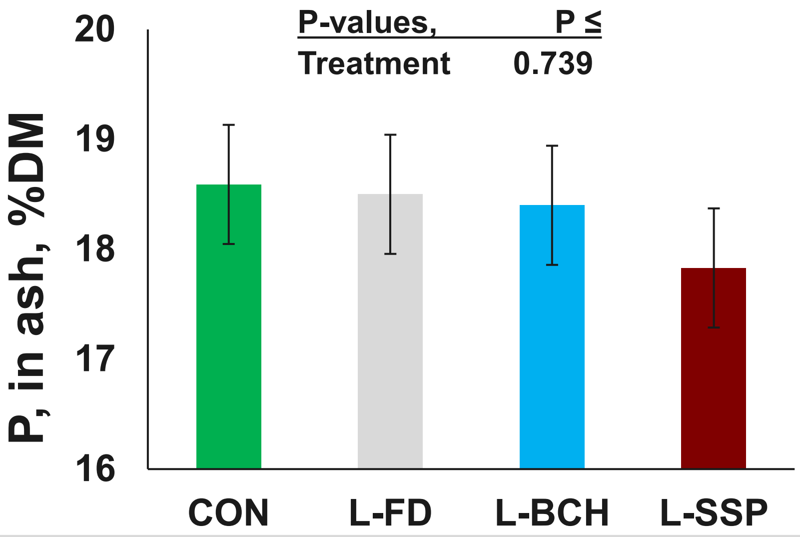


- Bone characteristics were not affected by groups

Results: Bone Mineralization







- Bone mineralization were not affected by groups

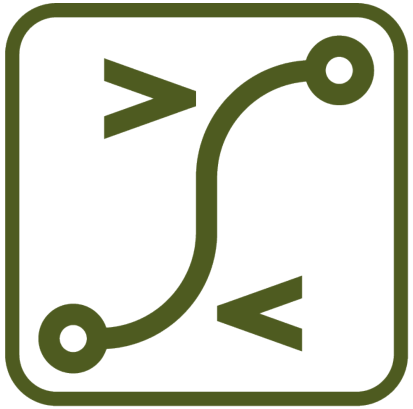
Our four focus topics

01

Individualising farm animal husbandry

02

Farming animals in sustainable resource cycles

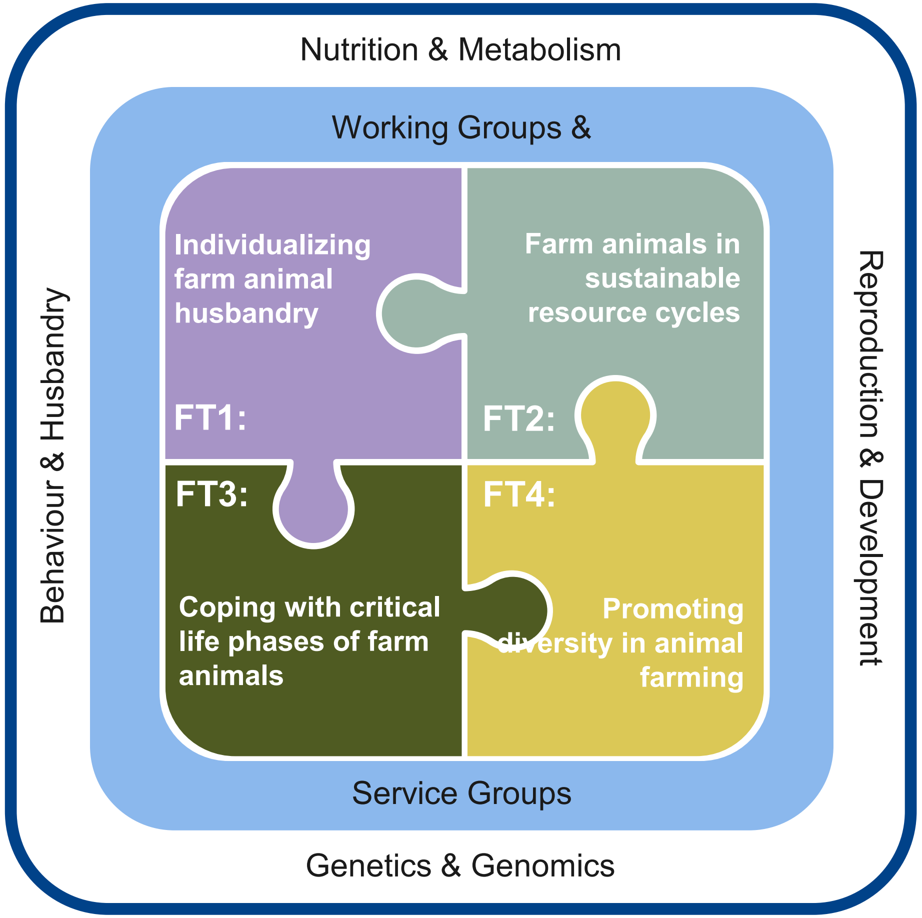


03

Coping with critical life phases of farm animals

04

Promoting diversity in animal farming


Nutrition & Metabolism
Working Groups &
Individualizing farm animal husbandry
FT1:
Farm animals in sustainable resource cycles
FT2:
FT3:
FT4:
Coping with critical life phases of farm animals
Promoting diversity in animal farming
Service Groups
Genetics & Genomics
Behaviour & Husbandry
Reproduction & Development

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