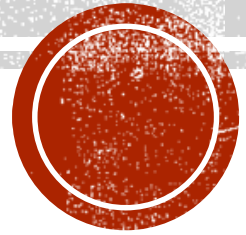
FEEDING ON LARVAE OF BLACK SOLIDER FLIES DOESN'T SUBSTANTIALLY MODIFY CHICKEN CAECAL MICROBIOTA

Moula, N., Cabaraux, J.-F., Dawans, E., Taminiau, B., Detilleux, J.

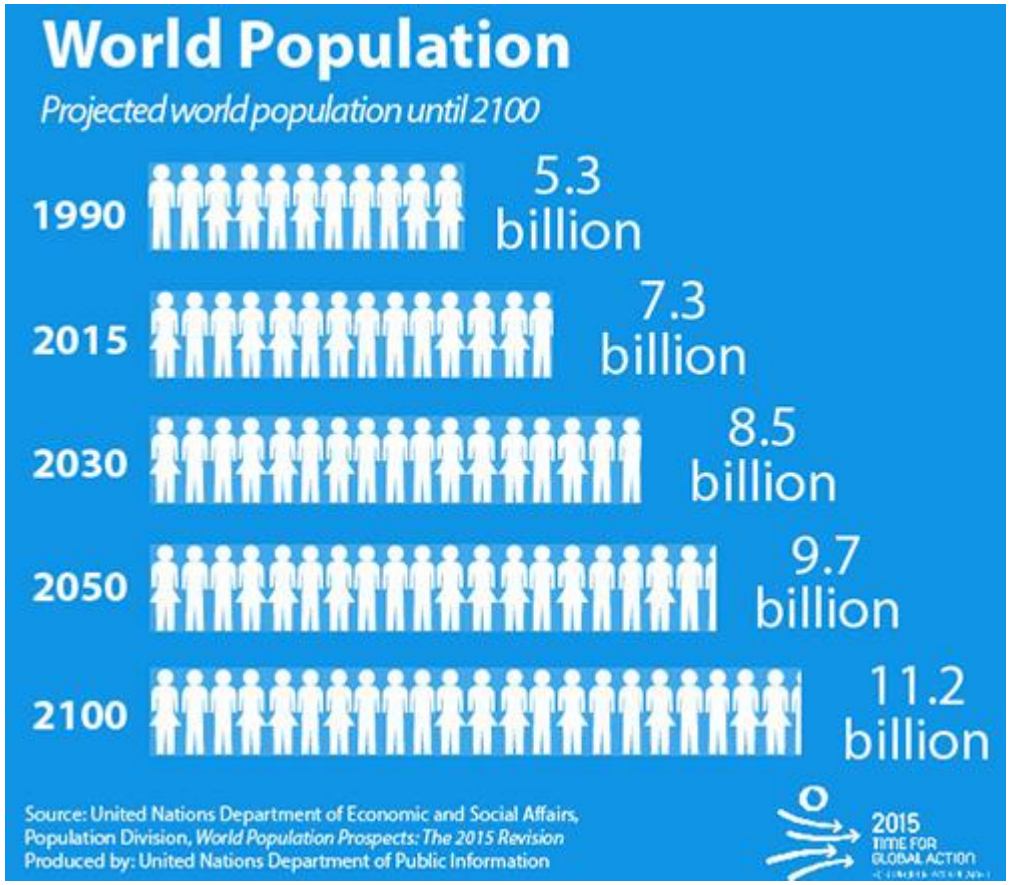
Veterinary management of animal resources



FARAH

LIÈGE université
Médecine Vétérinaire

WE NEED TO FEED THE WORLD

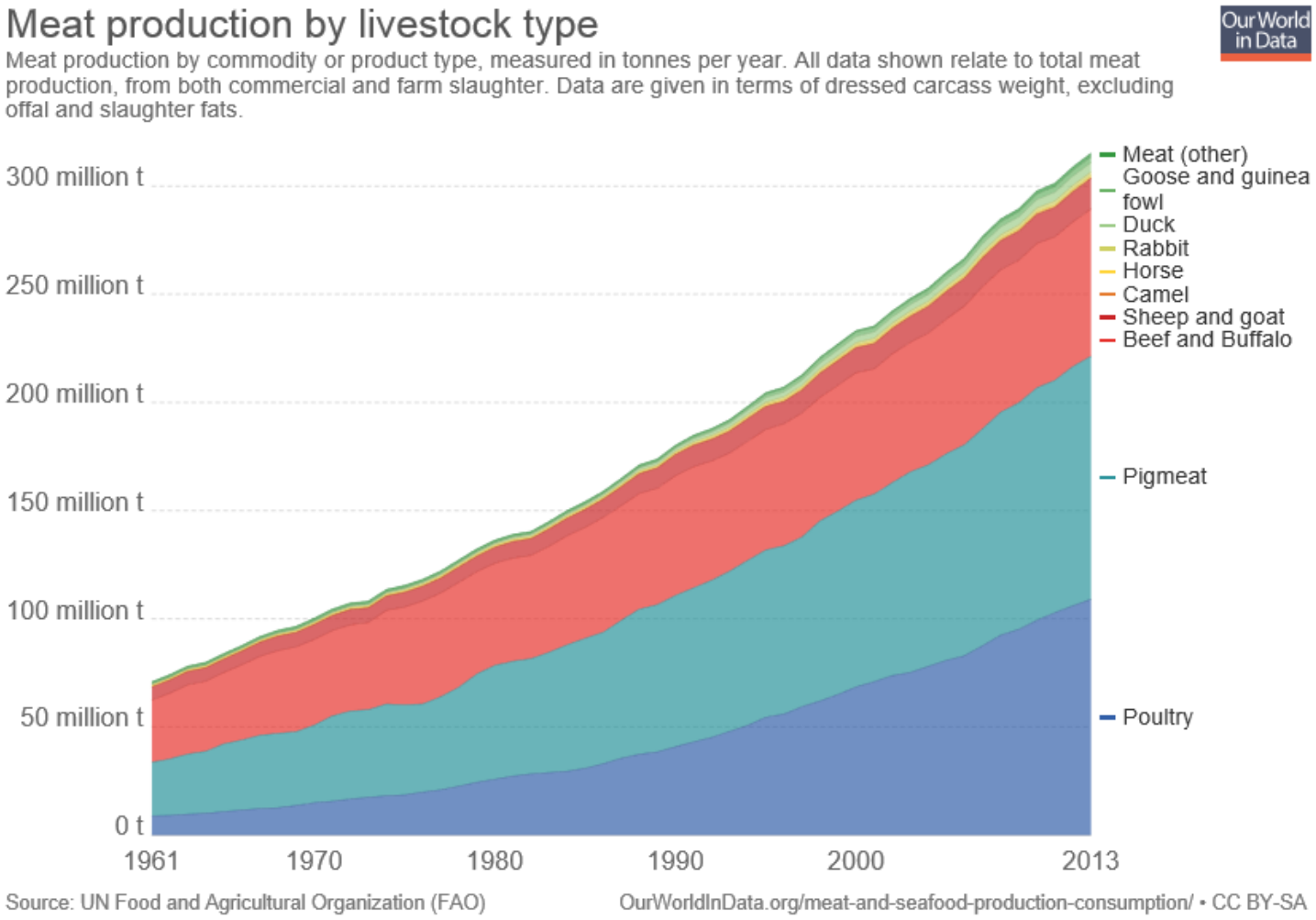


https://www.croatiatraveller.com/southern_dalmatia/Dubrovnik/cruise-schedule.html/

Mostly in developing countries

Food production must increase by 70%

MEAT PRODUCTION AND SUPPLY



CHICKEN CONSUMPTION

Table 3.10: Food consumption of meat

	1964/66	1974/76	1984/86	1994/96	1997/99	2015	2030
	kg per capita, carcass weight equivalent						
World	24.2	27.4	30.7	34.6	36.4	41.3	45.3
Developing countries	10.2	11.4	15.5	22.7	25.5	31.6	36.7
excl. China	11.0	12.1	14.5	17.5	18.2	22.7	28.0
excl. China and Brazil	10.1	11.0	13.1	14.9	15.5	19.8	25.1
Sub-Saharan Africa	9.9	9.6	10.2	9.3	9.4	10.9	13.4
Near East/North Africa	11.9	13.8	20.4	19.7	21.2	28.6	35.0
Latin America and the Caribbean	31.7	35.6	39.7	50.1	53.8	65.3	76.6
excl. Brazil	34.1	37.5	39.6	42.4	45.4	56.4	67.7
South Asia	3.9	3.9	4.4	5.4	5.3	7.6	11.7
East Asia	8.7	10.0	16.9	31.7	37.7	50.0	58.5
excl. China	9.4	10.9	14.7	21.9	22.7	31.0	40.9
Industrial countries	61.5	73.5	80.7	86.2	88.2	95.7	100.1
Transition countries	42.5	60.0	65.8	50.5	46.2	53.8	60.7
Memo item							
World excl. China	28.5	32.6	34.3	34.1	34.2	36.9	40.3
World excl. China and transition countries	26.5	29.0	30.6	32.4	33.0	35.6	39.1
	Meat consumption by type (kg per capita, carcass weight equivalent)						
World							
Bovine meat	10.0	11	10.5	9.8	9.8	10.1	10.6
Ovine and caprine meat	1.8	1.6	1.7	1.8	1.8	2.1	2.4
Pig meat	9.1	10.2	12.1	13.7	14.6	15.3	15.1
excl. China	9.7	10.8	11.3	10.4	10.3	9.9	9.7
Poultry meat	3.2	4.6	6.4	9.3	10.2	13.8	17.2
Developing countries							
Bovine meat	4.2	4.3	4.8	5.7	6.1	7.1	8.1
Ovine and caprine meat	1.2	1.1	1.3	1.6	1.7	2.0	2.4
Pig meat	3.6	4.1	6.4	9.6	10.8	12	12.2
excl. China	2.1	2.4	2.8	3.3	3.4	4.0	4.7
Poultry meat	1.2	1.8	2.9	5.8	6.9	10.5	14.0
excl. China and Brazil	1.2	1.9	3.2	4.8	5.2	8.1	11.6

http://www.fao.org/docrep/005/Y4252E/y4252e05b.htm

DIETARY PROTEIN

Role: growth, egg production, biological functions, …
Quantity: ~ 21 % of diet
Quality: methionine and lysine

Insect as a source of protein ?

70-80% of the total costs

Live broilers: Average costs of production at various R$/$ exchange rates, mid-year 2011 ($/kg)

	U.S.	Brazil					
	National	Parana	Goias	Parana	Goias	Parana	Goias
	(actual)	(actual)		(hypothetical)		(hypothetical)	
		R$1.58/$1		R$1.50/$1		R$2.00/$1	
Chicks	0.11	0.19	0.26	0.20	0.27	0.15	0.20
Feed	0.69	0.67	0.70	0.70	0.73	0.53	0.55
Grower payment	0.12	0.13	0.14	0.13	0.15	0.10	0.11
All other	0.08	0.07	0.09	0.07	0.09	0.05	0.07
Total	1.01	1.05	1.19	1.10	1.25	0.83	0.94

Exchange rate based on the average of June and July 2011
Brazil - based on conventional grow-out houses
Brazilan grower's payment is constructed based on producer (grow-out farmer) total costs plus water

http://www.thepoultrysite.com/reports/?id=801

INSECTS AS A SOURCE OF PROTEIN

https://www.standardmedia.co.ke/

Rapid reproductory rates :
1000 eggs/female
egg-adult : 38 days
Able to convert organic waste
Self-harvest capability
Absence of mouthparts in their adult form

Nutrient content of wild black soldier fly (BSF) larvae produced on vegetable and fruit wastes compared to that of fishmeal and soybean.

	Wild BSF larvae	Fishmeal	Soybean
Crude protein (%)	38.98	55.1	47.9
Crude fibre (%, w/w)	12.36	0.013	3.6
Ash (%, w/w)	14.61	18.2	6.40
Fat (%, w/w)	32.62		1.80
Free fatty acids (mh NAOH/g)	42.075	18.7	29.2
Manganese (%)	0.56		
Copper (%)	0.006		
Sodium (%)	3.07		
Iron (%)	0.57	0.82	
Calcium (%)	0.10	1.586	0.35
Potassium (%)	2.27		0.56

https://www.wageningenacademic.com/

MICROBIOTA

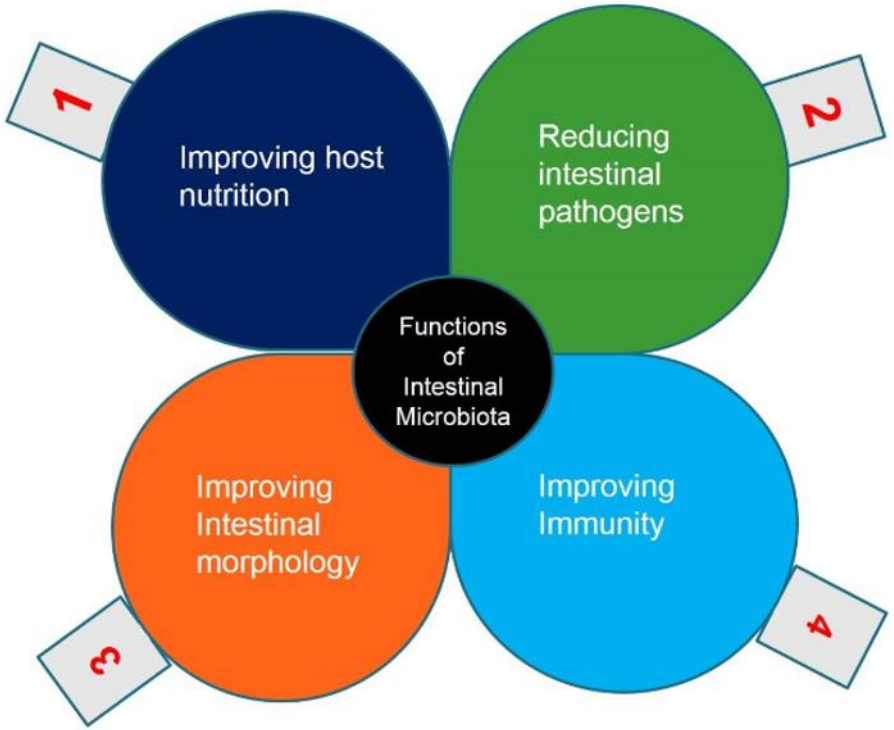
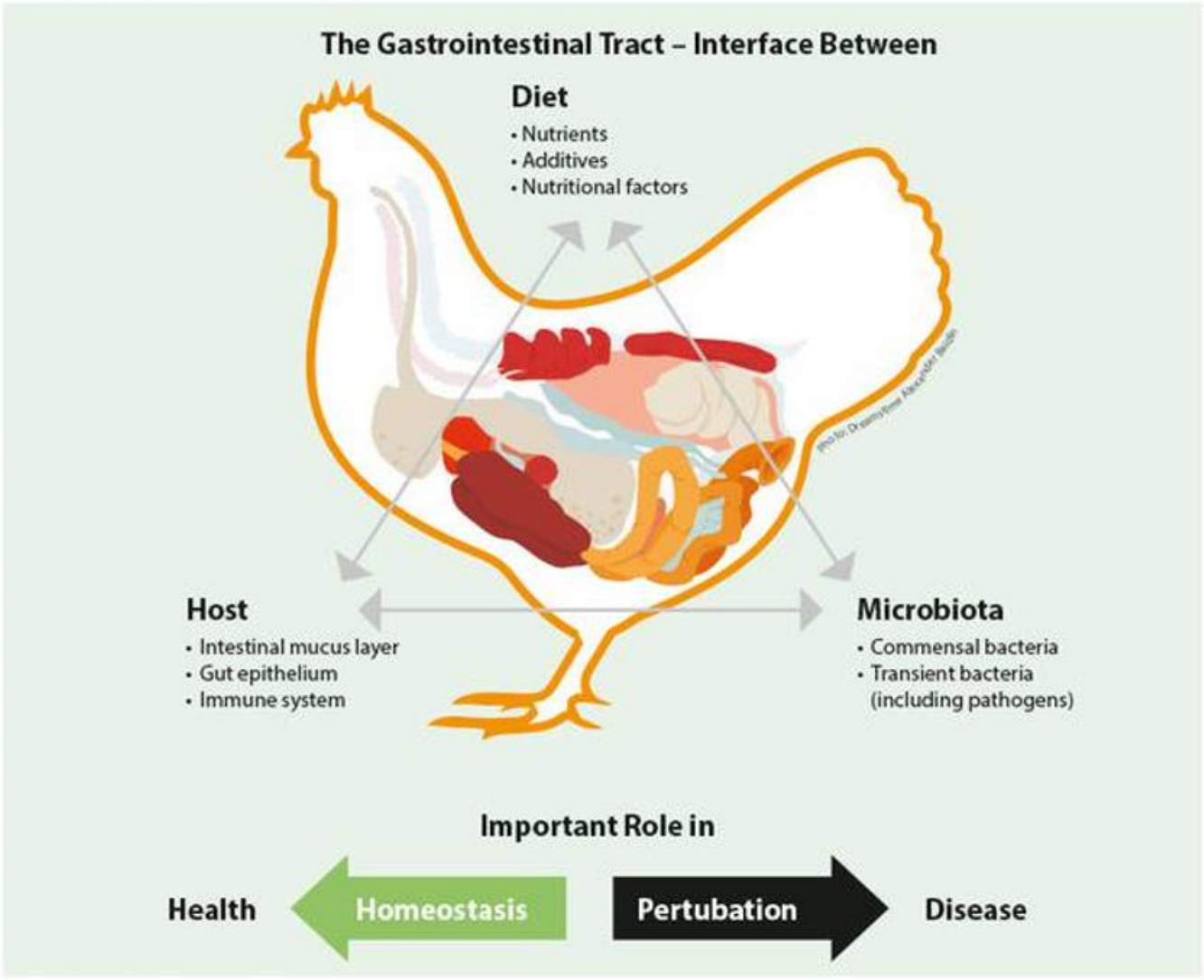


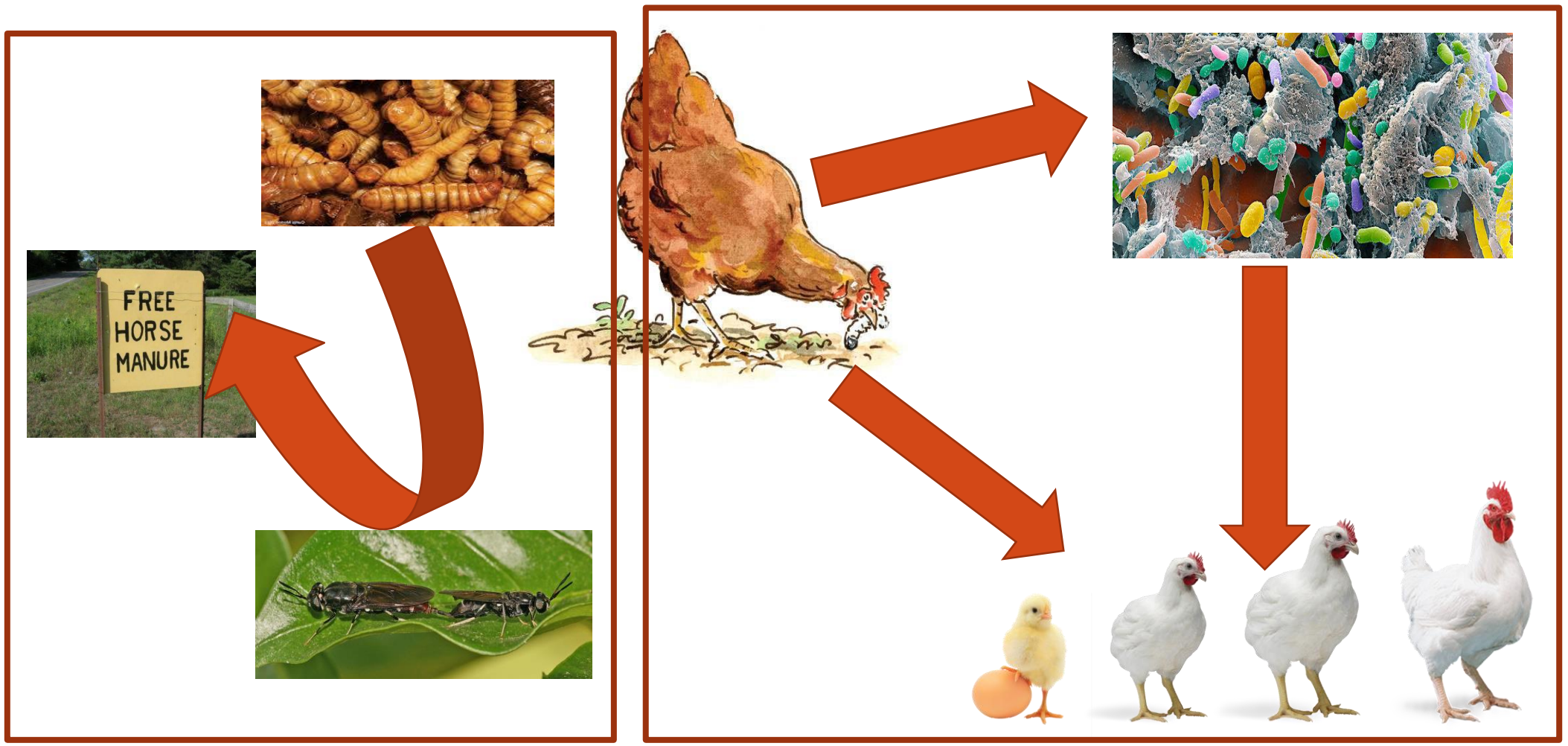
Figure 1. Interplay between diet, host, and gut microbiota - factors influencing the gut ecosystem and contributing to health and disease.



Source: Adapted from Conway, 1994

http://www.primeanimalhealth.com/microbiota.html

GOAL OF THE STUDY



Change in caecal microbiota of chicken fed BSF larvae raised on horse manure

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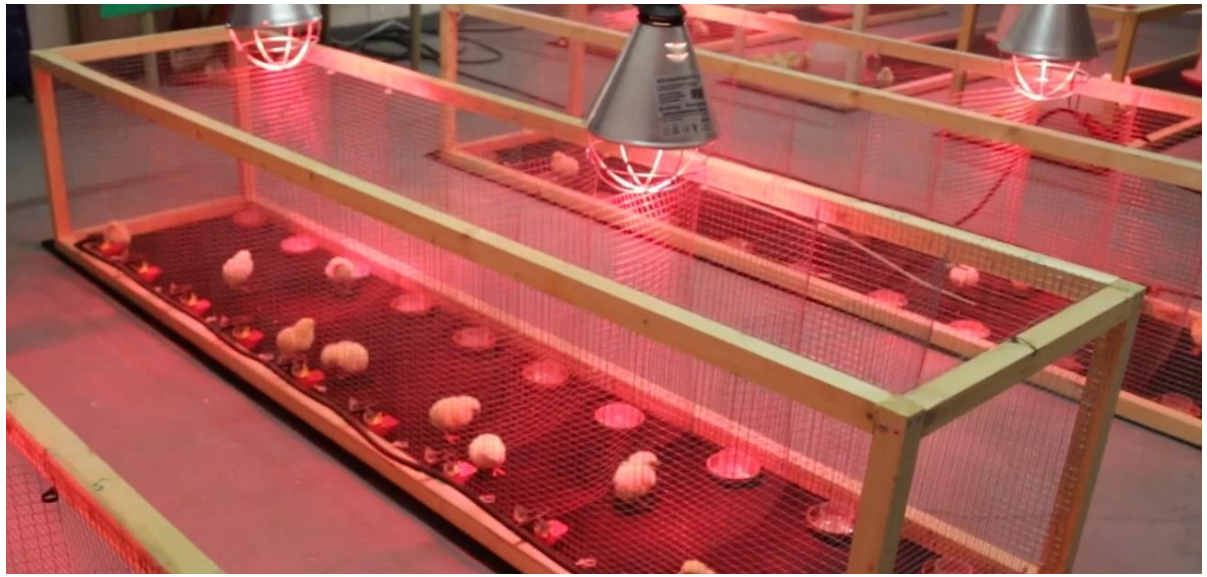
http://www.fmv-multimedia.ulg.ac.be

MATERIAL AND METHODS

40 Ross chicks: 0-13 days
2 groups; 2 replicates; 10 birds
Individual cages



http://www.fmv-multimedia.ulg.ac.be

MATERIAL AND METHODS

Iso-nitrogenous and iso-caloric	Control	Experimental
Dry matter (g/kg)	149	147
Crude protein (g/kg)	201	201
Crude fat (g/kg)	47	47
Nitrogen-free extract (g/kg)	509	507
Crude fiber (g/kg)	29	31
Ash (g/kg)	65	67
Starch (g/kg)	468	453
Gross energy (MJ/kg)	15.6	15.6

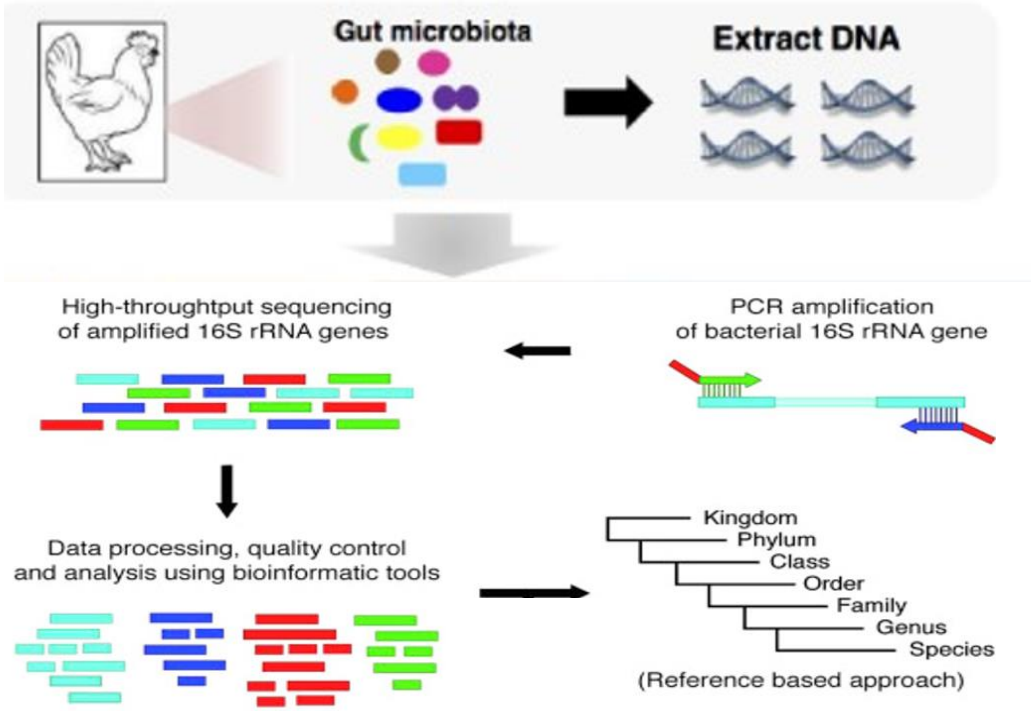
Feed supplied *ad libitum*
Diet as mash
Starter commercial feed
Iso-nitrogenous and iso-caloric

MATERIAL AND METHODS

http://www.fmv-multimedia.ulg.ac.be

MATERIAL AND METHODS

16s ribosomal *RNA* sequencing techniques



MOTHUR

Reference alignment and taxonomical assignation

silva
high quality ribosomal RNA databases

RESULTS (1)

17,616 OTU, 37 families and 5,275 species

Firmicutes: 90.83%
- *Ruminococcaceae* (33.01%)
- *Lachnospiraceae* (46.17%)

Bacteroidetes : 6.93%
- *Bacteroidaceae* (6.65%)

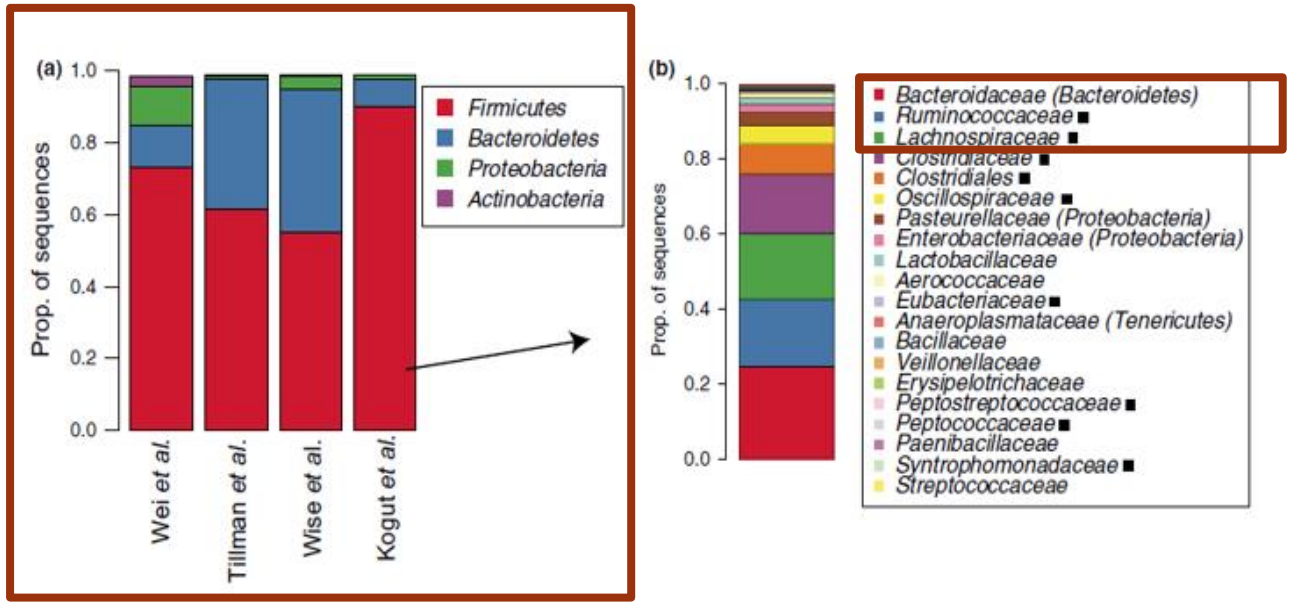


Fig. 1. Relative proportions of bacterial phyla (a) and families (b) found in chicken ceca. Data from Wei *et al.* (2013) represent publically available sequences retrieved as described. Data from Tillman *et al.* (2011) and Wise & Siragusa (2007) are re-analyzed from data included in (Oakley *et al.*, 2013) representing 8 and 10 birds, respectively. Kogut *et al.* data are unpublished, collected, and analyzed as previously described (Oakley *et al.*, 2012 b, 2013) representing 20 birds and c. 20 000 sequencing reads. Data for each of these three flocks are from 3 weeks posthatch. Sequences from Wei *et al.* were additionally screened by removing sequences with ambiguous base calls, and all sequences were classified against a reference database of type strains from SILVA v115 (Pruesse *et al.*, 2007). Many of the sequences reviewed in (Wei *et al.*, 2013) do not contain metadata regarding bird age, which can have strong effects on community composition and structure. For (b) families belong to the phylum Firmicutes unless otherwise noted; families followed by black squares belong to the Clostridiales.

RESULTS (1)

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- *Ruminococcaceae* (33.01%)
- *Lachnospiraceae* (46.17%)

Bacteroidetes : 6.93%
- *Bacteroidaceae* (6.65%)

Degradation of cellulose and other highly recalcitrant polysaccharides

Degradation of less recalcitrant non-starch polysaccharides and starch

<u>Butyric acid</u> :
- source of energy
- anti-inflammatory properties
  production of mucins and antimicrobial peptides
  inhibition of the transcription factor NFκB activity
  inhibitory activity against *Salmonella* and *Clostridium perfringens*

RESULTS (2)

Phylum	Control group	Experimental group
Actinobacteria	0.02 (0.00 - 0.04)	0.04 (0.00 - 0.06)
Bacteroidetes	7.61 (0.15 - 15.07)	5.70 (0.13 - 11.27)
Firmicutes	89.92 (84.83 - 95.00)	92.39 (86.91 - 97.86)
Proteobacteria	0.78 (0.69 - 0.87)	0.20 (0.17 - 0.24)
Other and unclassified bacteria	1.67 (0.01 - 2.75)	1.67 (0.01 - 2.75)

Means (and 95% confidence intervals) for the relative abundance (%) of bacterial phyla in the caeca of young Ross broilers fed (experimental diet) or not (control diet) larvae of black soldier flies.

Bacillaceae: 0.04% vs 0.00%

Rhodobacteraceae: 0.31% vs 0.02%

RESULTS (2)

Key genera	Species	SD (%)	ID (%)
Bacteroides	*Bacteroides coprocola*	2.97 ± 1.01	1.20 ± 0.25
	Bacteroides heparinolyticus	2.47 ± 0.85	1.38 ± 0.19
	Bacteroides plebeius	1.00 ± 0.24	3.00 ± 0.58*
	Bacteroides salanitronis	4.37 ± 0.58	0.45 ± 0.15**
Parabacteroides	*Parabacteroides merdae*	3.69 ± 0.80	0.82 ± 0.51*
Elusimicrobiaceae	*Elusimicrobium minutum*	0.23 ± 0.06	1.51 ± 0.52*
Clostridiales	*Alkaliphilus crotonatoxidans*	1.74 ± 0.34	3.04 ± 0.77
	Alkaliphilus transvaalensis	0.12 ± 0.04	1.25 ± 0.32**
	Christensenella minuta	0.66 ± 0.10	3.39 ± 0.35**
	Vallitalea guaymasensis	0.40 ± 0.06	1.03 ± 0.17*
Oscillospira	*Flavonifractor plautii*	1.36 ± 0.16	4.61 ± 0.71**
	Intestinimonas butyriciproducens	3.13 ± 0.29	4.61 ± 0.61
	Oscillibacter valericigenes	2.61 ± 0.41	2.70 ± 0.09
Ruminococcus	*Clostridium leptum*	0.53 ± 0.04	1.06 ± 0.20
Veillonellaceae	*Unclussified sp*	1.25 ± 0.15	0.41 ± 0.07**
Fusobacterium	*Unclussified sp*	6.02 ± 2.77	0.26 ± 0.08**
Succinivibrionaceae	*Succinatimonas hippei*	1.61 ± 0.21	0.25 ± 0.08**

SCIENTIFIC REPORTS

OPEN

: 14 June 2017
l: 14 November 2017
l online: 24 November 2017

Insect-based diet, a promising nutritional source, modulates gut microbiota composition and SCFAs production in laying hens

Luca Borrelli[1,6], Lorena Coretti[2,3,6], Ludovico Dipineto[1,6], Fulvia Bovera[1], Francesca Menna[1,6], Lorenzo Chiariotti[2,4,6], Antonio Nizza[1], Francesca Lembo[5,6] & Alessandro Fioretti[1,6]

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RESULTS (2)

Table 8. Selected microbiota counts in the caecal digesta determined by DAPI staining and fluorescent *in situ* hybridization, log cell number · g^{-1} of digesta (Experiment 3)

Indices	Treatments[1]				SEM[2]	*P*-value
	NC	SL20	TM20	HI20		
total number of bacteria[3]	9.77	9.73	9.56	9.84	0.04	<0.0001
Bacteroides–Prevotella cluster	8.61^{c}	8.81^{b}	8.60^{c}	9.05^{a}	0.09	<0.0001
Clostridium leptum subgroup	8.28	8.32	8.43	8.45	0.07	0.061
Streptococcus spp./ *Lactococcus* spp.	8.17^{b}	8.33^{b}	8.21^{b}	8.93^{a}	0.20	<0.0001
Clostridium coccoides–Eubacterium rectale cluster	8.22^{c}	8.39^{bc}	8.48^{b}	8.87^{a}	0.16	<0.0001
Lactobacillus spp./ *Enterococcus* spp.	8.27^{b}	8.54^{ab}	8.17^{c}	8.60^{a}	0.05	0.007

Journal of Animal and Feed Sciences, 27, 2018, 131–139 https://doi.org/10.22358/jafs/91967/2018
The Kielanowski Institute of Animal Physiology and Nutrition, Polish Academy of Sciences, Jabłonna

Full-fat insect meals as feed additive – the effect on broiler chicken growth performance and gastrointestinal tract microbiota

A. Józefiak[1,6], B. Kierończyk[2], M. Rawski[2,3], J. Mazurkiewicz[3,4], A. Benzertiha[4], P. Gobbi[4], S. Nogales-Mérida[4], S. Świątkiewicz[5] and D. Józefiak[2]

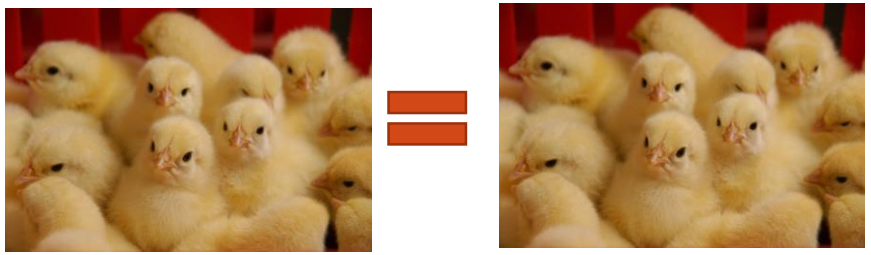
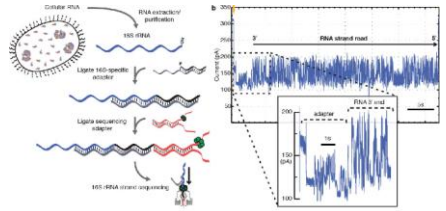


Table 3. Oligonucleotide probes used for intestinal microbiota analyses using fluorescent *in situ* hybridization (FISH) (Józefiak et al. (2016))

Target	Probe	Sequence (from 5' to 3')
Bacteroides–Prevotella cluster	Bacto303	CCAATGTGGGGGACCTT
Enterobacteriaceae	Enter1432	CTTTTGCAACCCACT
Lactobacillus spp./ *Enterococcus* spp.	Lab158	GGTATTAGCAYCTGTTTCCA
Clostridium coccoides–Eubacterium rectale cluster	Erec482	GCTTCTTAGTCARGTACCG
Clostridium leptum subgroup	Clept1240	GTTTTRTCAACGGCAGTC
Streptococcus spp./ *Lactococcus* spp.	Strc493	GTTAGCCGTCCCTTTCTGG



RESULTS (3)

Presence of BSF larvae in the diet

decreases the *relative abundance of Bacillaceae*

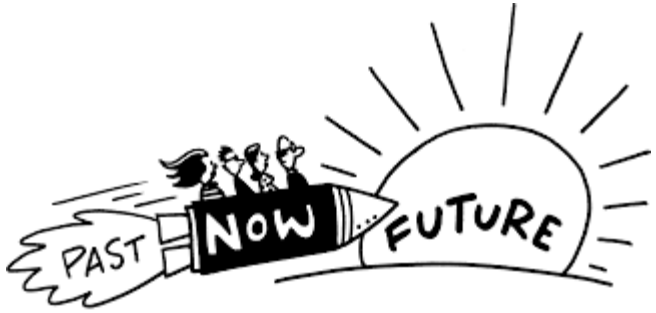
which increases → Average daily gain

which decreases → Feed conversion ratio

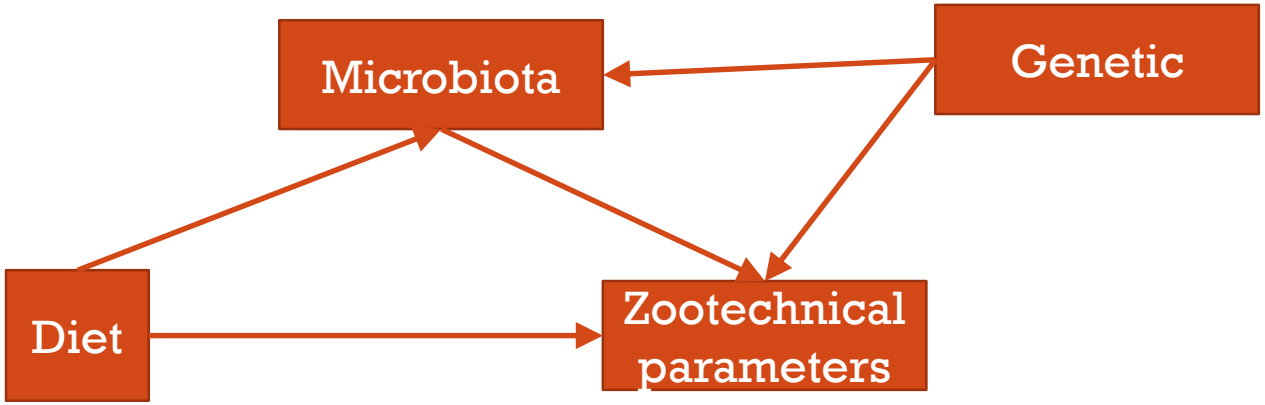
Abdominal fat score

increases the *relative abundance of Rhodobacteraceae* → Tibia ash percentage

NEXT ?



- Preliminary findings need to be confirmed
  - Higher number of animals
  - Different percentages of BSF in the feed
  - Improved statistical methods
    - Design : non-genetic effects (gender, diet, replication, ….)
    - Mixed structural equation model



NEXT ?

- Preliminary findings need to be confirmed
  - Higher number of animals
  - Percentage of BSF in the feed
  - Statistical methods (SEM)
- Niger
  - BSF and/or other insects
  - Imported chicken “without head” *vs* local chicken “bicycle”