National Rural Development Program 2014-2022

Measure 10.2 – Biodiversity

Project: TuBAvI-2 (2021-2024)

REPORT ON THE ACTIVITIES UNDERTAKEN DURING THE THIRD YEAR

PA UniPI

The present report describes the activities carried out from May 1st, 2023, to March 31st, 2024. The activities are described by Action, according to the original programme.

Action 1 – Phenotypical characterization of autochthonous breeds and species

Task 1.1 - Phenotypic characterization of Livorno, Siciliana, Bianca di Saluzzo, Bionda Piemontese, and Millefiori Piemontese chicken breeds.

During the period under consideration, Livorno (L) and Siciliana (S) chicken breeds, hatched in April 2021 and raised at the Poultry Breeding facility of the Department of Veterinary Sciences at the University of Pisa, completed their second laying cycle by October 2023. All animals were raised on sandy soil in outdoor enclosures with partial roofing, under natural light and temperature conditions.

Monitoring of daily egg production continued until the end of October 2023 (see figure 1.1.1), while egg weight evaluation (see figure 1.1.2) and egg morphometric determinations of second-laying eggs were completed by the end of June (data in processing). The assessment of food consumption by second-laying hens was completed by June 2023, and the egg FCE was calculated (see table 1.1.1). Replacement subjects born in April 2023 were monitored for growth, mortality, and morphometric characteristics as outlined in the project (data in progress).

The data concerning the Piedmontese breeds Bianca di Saluzzo (BS), Bionda Piemontese (BP), and Millefiori di Lonigo (ML) preserved at the Poultry Centre for the Conservation of Local Genetic Resources (Carmagnola, TO - supervised by Prof. Achille Schiavone) have been collected and are made available as part of the technical consultancy contract between UniPI and UniTO starting from 05/03/2022 (refer to the technical report by the UniTO consultant).

Task 1.2 - Phenotypic characterization of Ancona (A) breed chickens

Monitoring of daily egg production during the second egg-laying cycle of Ancona hens continued until the end of October 2023 (see figure 1.2.1), while egg weight detection concluded in July (see figure 1.2.2).

Task 1.3 - Characterization of laying hens' adaptation capacity to seasonal environmental stimuli

In 2023, the qualitative characterization of the yolk and albumen of the eggs produced during the second cycle of laying of the L and S breeds was completed in June. The egg components were

evaluated (see table 1.3.1), and chemical analyses of the yolk and egg white were conducted (see table 1.3.2).

Action 7 - Evaluation and identification of genetic resistance traits in livestock animals against diseases

Task 7.1 - Evaluation of resilience for growth under environmental stimuli

In September 2023, the growth test for the Ermellinata di Rovigo (ER), Ancona (ANC), Pepoi (PP), and Robusta Lionata (RL) breeds concluded. This test began with hatching at the end of February and was conducted at the UniPI and UniFI facilities. Figure 7.1.1 shows the body weights and growth rates of the Pisa animals during the first 150 days of life; consumption and ICA data are currently being processed. Total mortality rates (1-150 days of age) were 68.4%, 38.1%, 27.3%, and 25.0% for RL, PP, ER, and ANC, respectively, primarily due to colibacillosis, which manifested itself on the 14th day of the animals' lives. Figure 7.1.2 displays the average body weights of 120-day-old chickens raised at the two sites.

Task 7.2 - Evaluation of resistance/resilience to bacterial and parasitic infections/infestations and resilience of intestinal microbiota

Table 7.2.1 shows final results of the qualitative and quantitative parasitological analyses to detect coccidia (*Eimeria* spp.), nematodes (roundworms, capillaries, *Heterakis* spp.), protozoa (*Giarda* spp. and *Cryptosporidium* spp.) and *Histomonas meleagridis* in stool samples collected from the growing chickens monitored in task 7.1.

At the age of 120 days, four individual cloacal swabs were taken from the same chickens at both sites to identify pathogens such as *Salmonella* spp., *Clostridium perfringens*, *Campylobacter jejuni*, and commensal bacteria such as *Enterococcus* spp., *Lactobacillus* spp., and *Bifidobacterium* spp. (data in progress) and to obtain isolates for evaluating the antibiotic resistance profile. Table 7.2.2 presents the incidence of the different breeds in the antibiotic susceptibility classes found in the Lactobacilli isolated from them.

At the end of the monitoring period, individual cloacal faecal sampling was conducted to characterize the intestinal microbiota of the chickens, performed by PA UniFI as part of its research program.

Action 8 - Collection of Biological Material and Germplasm

At the end of 2023, the procurement of biological material from Livorno breed chickens from farms in the Tuscan territory was completed.

To conduct task 7.2, five collections of faecal samples were carried out from the four breeds (ER, ANC, PP, and RL) under study at both sites, totalling 16 sampling pools. Cloacal swabs were performed on subjects bred at both the Pisa and Florence sites, totalling 520 samples (four swabs per animal).

Action 10 – Information and Dissemination

Information Events

- 29 September 2023: BRIGHT Night 2023 - The European Night of Researchers.

- 07-08 October 2023: Urban Nature 2023 – Event: "Nature takes care"

- 24 November 2023: Game of Research (UniPi first edition) – Day to promote research in veterinary sciences, dedicated to non-structured staff. Oral presentation "Italian native poultry breeds: zootechnical and parasitological investigations" (PhD student Marta Raffaelli).

Technical-informative publications for the Pollitaliani website

- Update of sheets relating to the Siciliana breed, the Ancona breed and the White Livorno breed.

- Creation of a sheet on the Black Livorno breed.

Scientific publications

- Physical and chemical characteristics of eggs from eight Italian chicken breeds. Italian Journal of Animal Science (DOI: 10.1080/1828051X.2024.2314149)

FIGURES AND TABLES

Figura 1.1.1 – Egg-laying curve of white Livorno, black Livorno and Siciliana breed hens at second-laying, during the reproductive period from January to December 2023

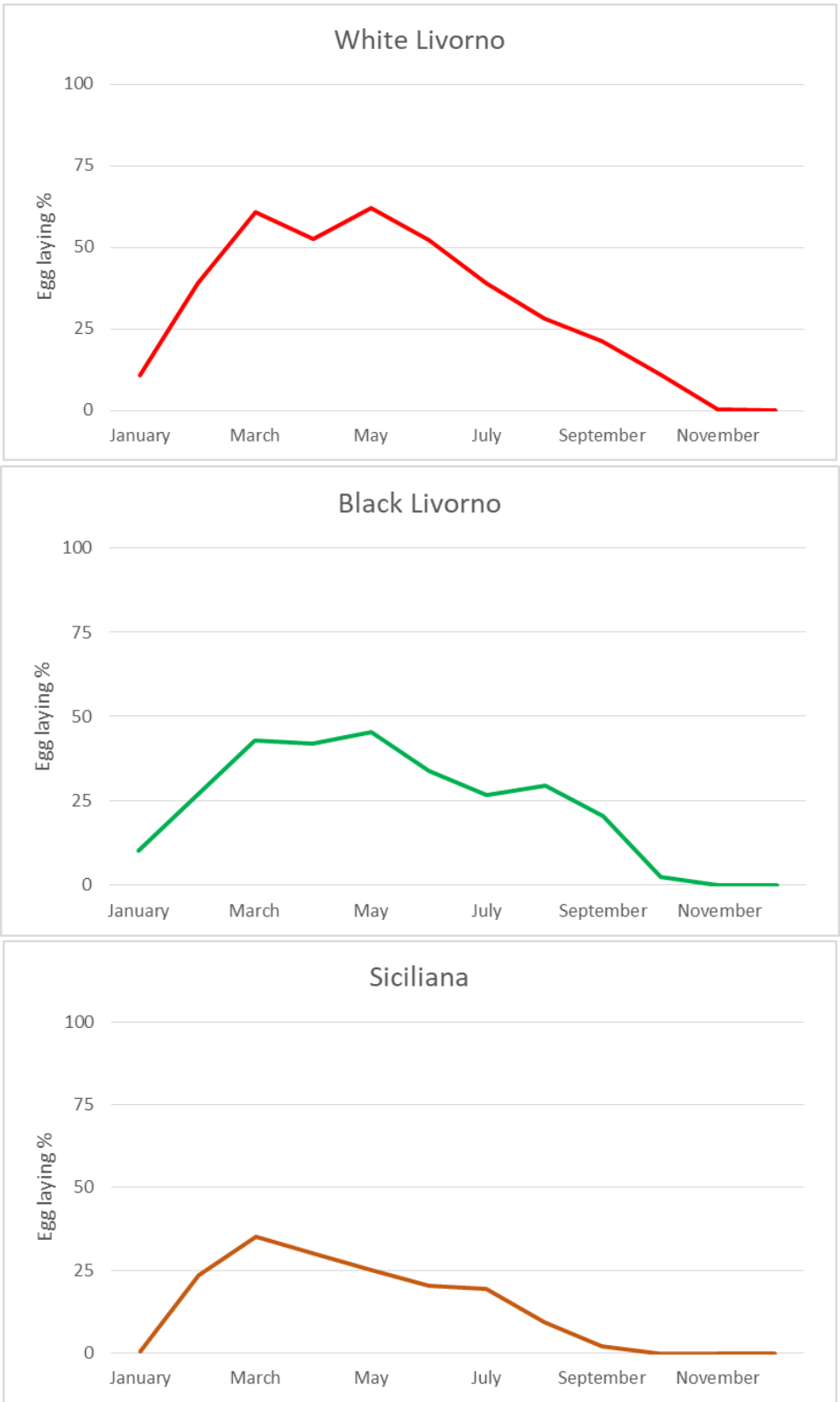
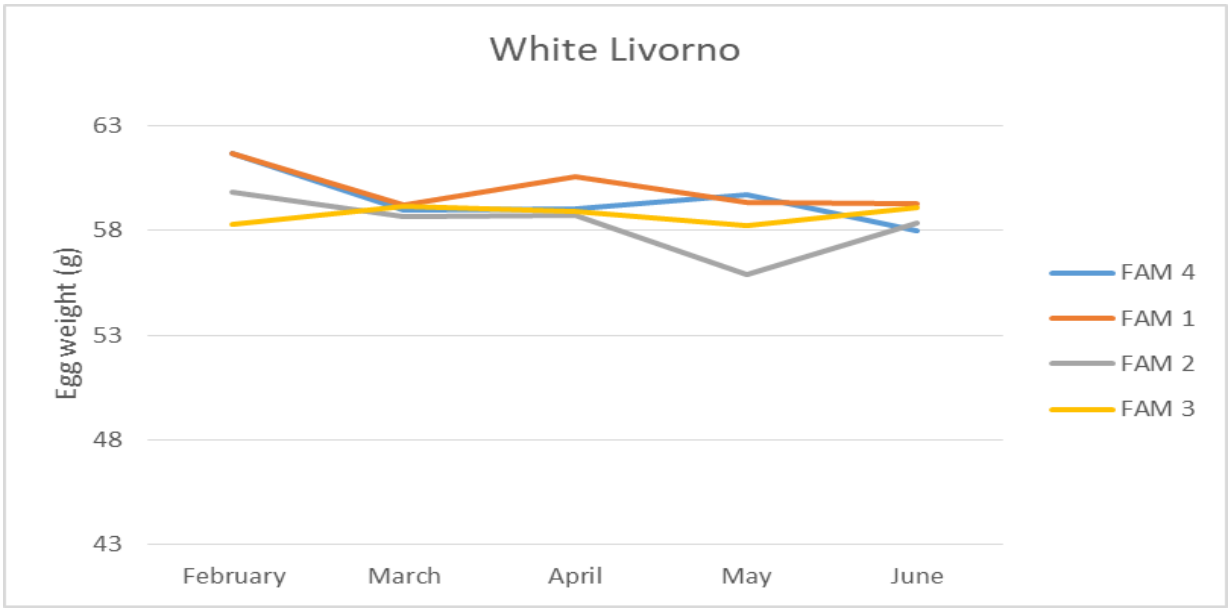


Figura 1.1.2 – Average monthly egg weight from white Livorno and Siciliana breed hens groups at second-laying, recorded from February to June 2023



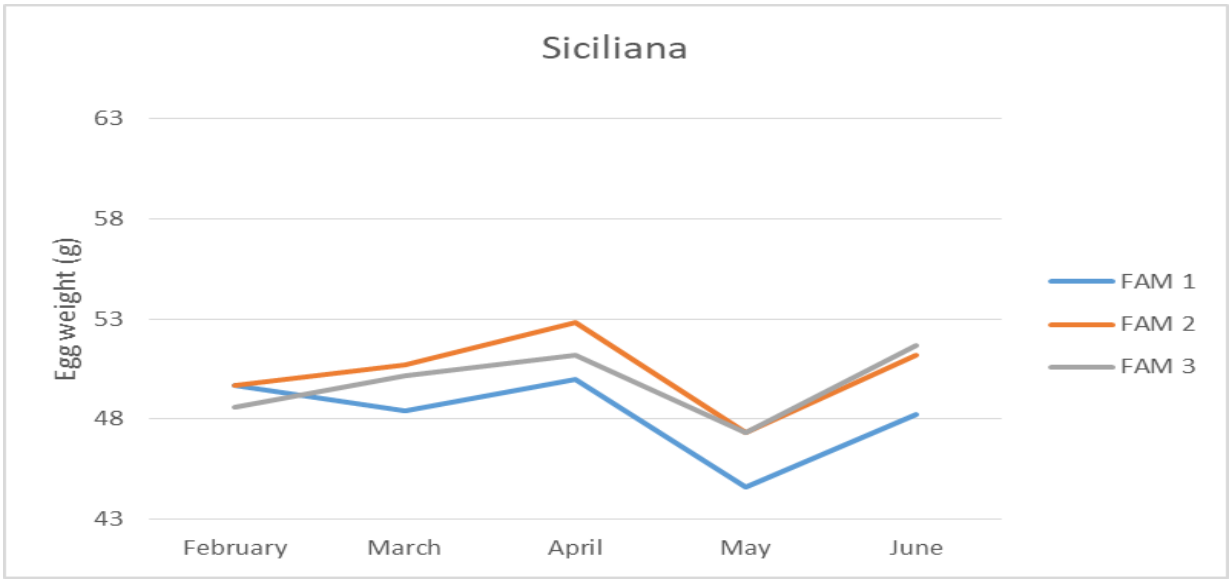


Table 1.1.1 – Feed consumption and FCE for eggs in the different production periods of white Livorno (LB), black Livorno (LN), and Siciliana (SIC) breed hens at second-laying

BREED	average bird/day feed consumption (g)			
	March	April	May	June
LB	128.5 ± 1.4	148.4 ± 4.8	75.1 ± 5.3	93.4 ± 3.0
LN	182.7 ± 33.0	173.0 ± 0.7	125.9 ± 6.2	152.9 ± 13.0
SIC	121.8 ± 36.5	114.3 ± 17.3	99.8 ± 13.8	100.1 ± 19.9
	average FCE (g)			
	March	April	May	June
LB	3.4 ± 0.2	4.5 ± 0.1	3.5 ± 1.7	3.0 ± 0.4
LN	4.3 ± *	3.7 ± 1.0	3.3 ± 1.2	4.2 ± *
SIC	4.8 ± *	5.5 ± 2.9	7.9 ± 3.1	5.8 ± *

* referring only to the group in production

Figure 1.2.1 - Egg-laying curve of Ancona breed hens at second-laying, recorded in the reproductive period from January to December 2023

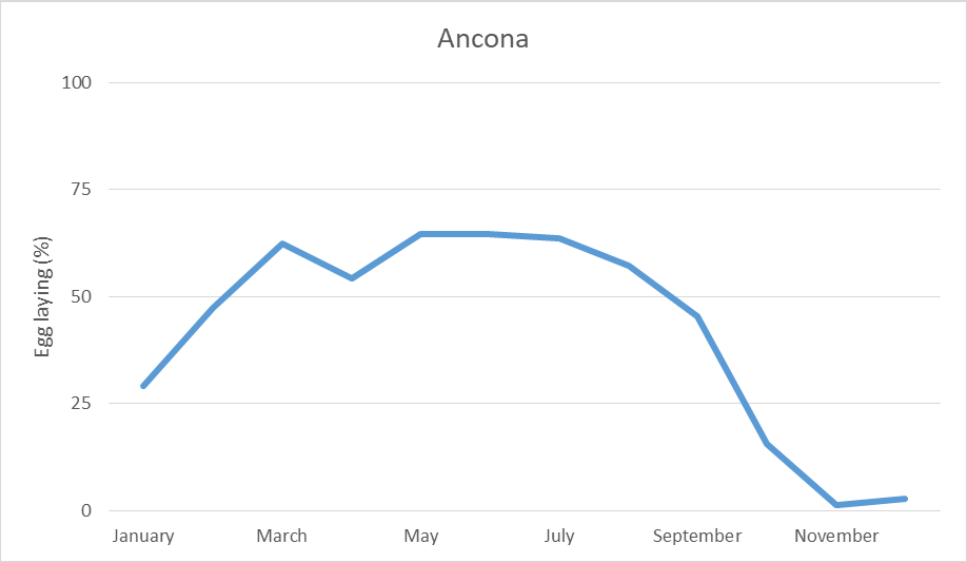


Figure 1.2.2 – Average monthly egg weight from three Ancona breed hens groups in the second-laying, recorded from February to June 2023

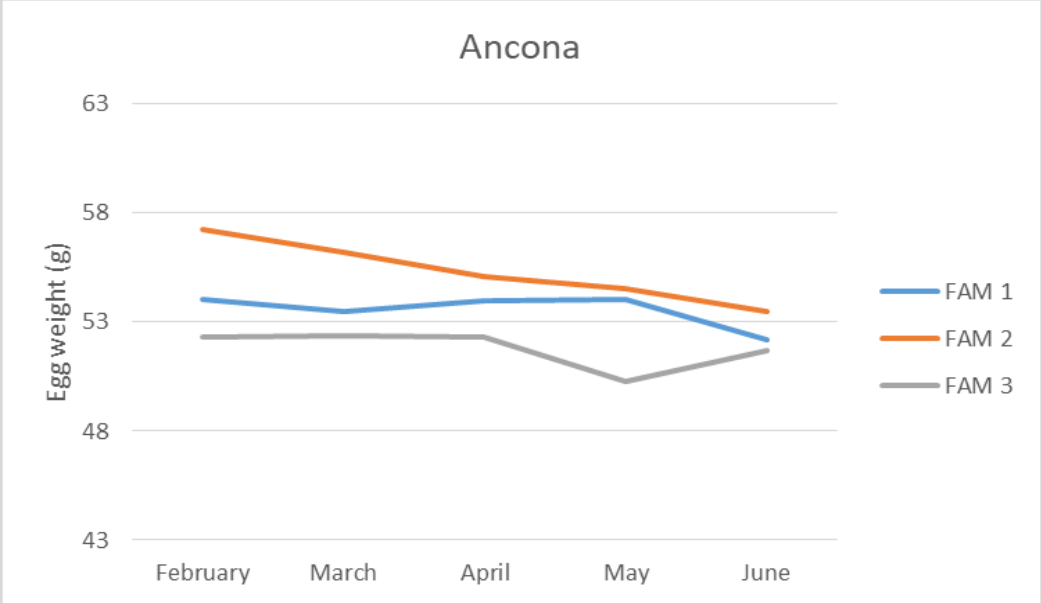


Table 1.3.1 – Physical characterisation of eggs from white Livorno (LB) and Siciliana (SIC) hens at second-laying

Breed		Weight (g)	Components weight (g)			Shell thickness (mm)			Components (%)		
		Egg	Yolk	Albumen	Shell	Polo Acuto	Polo Ottuso	Equatore	Yolk	Albumen	Shell
LB	Feb.	60.40 ± 3.26	17.40 ± 1.12	37.42 ± 2.68	6.31 ± 0.50	0.41 ± 0.04	0.40 ± 0.04	0.41 ± 0.03	28.51 ± 2.13	61.16 ± 2.24	10.32 ± 0.68
	March	59.02 ± 3.49	16.73 ± 1.06	36.41 ± 2.68	6.14 ± 0.52	0.41 ± 0.03	0.40 ± 0.02	0.41 ± 0.02	28.26 ± 1.89	61.38 ± 1.94	10.36 ± 0.63
	April	58.99 ± 3.75	17.01 ± 1.41	36.66 ± 2.56	5.94 ± 0.66	0.42 ± 0.04	0.39 ± 0.04	0.40 ± 0.03	28.55 ± 1.82	61.50 ± 2.16	9.95 ± 0.93
	May	58.13 ± 4.86	17.02 ± 1.52	35.49 ± 3.72	5.62 ± 0.51	0.40 ± 0.02	0.39 ± 0.03	0.38 ± 0.02	29.35 ± 2.28	60.96 ± 2.24	9.69 ± 0.62
	June	58.60 ± 2.90	16.89 ± 1.21	35.97 ± 2.35	5.80 ± 0.48	0.41 ± 0.03	0.39 ± 0.03	0.39 ± 0.02	28.81 ± 1.84	61.30 ± 1.95	9.98 ± 0.75
SIC	Feb.	49.49 ± 1.75	15.53 ± 0.93	28.49 ± 1.39	5.50 ± 0.33	0.41 ± 0.02	0.38 ± 0.03	0.41 ± 0.02	31.37 ± 1.60	57.52 ± 1.54	11.11 ± 0.58
	March	50.03 ± 3.58	15.78 ± 1.11	29.23 ± 2.84	5.64 ± 0.82	0.42 ± 0.04	0.40 ± 0.03	0.41 ± 0.05	31.23 ± 2.09	57.65 ± 1.89	11.11 ± 1.32
	April	51.53 ± 3.95	15.75 ± 1.09	30.63 ± 3.21	5.63 ± 0.75	0.43 ± 0.06	0.40 ± 0.05	0.41 ± 0.05	30.47 ± 1.59	59.08 ± 2.76	10.90 ± 1.41
	May	45.36 ± 2.43	15.57 ± 1.25	26.47 ± 2.68	5.39 ± 0.36	0.40 ± 0.03	0.41 ± 0.03	0.40 ± 0.02	33.57 ± 1.44	57.49 ± 4.81	11.30 ± 0.91
	June	50.68 ± 3.90	15.94 ± 1.26	29.68 ± 2.97	5.05 ± 0.85	0.39 ± 0.05	0.37 ± 0.04	0.38 ± 0.05	31.51 ± 2.12	58.53 ± 2.76	9.96 ± 1.49

Table 1.3.2 – Chemical analysis of the yolk and albumen of eggs from white Livorno (LB) and Siciliana (SIC) hens at second-laying

Breed		Yolk (%)				Albumen (%)		
		Lipids	Proteins	Dry matter	Sterins	Lipids	Proteins	Dry matter
LB	February	41.18 ± 0.86	9.04 ± 0.65	54.09 ± 1.21	1.89 ± 0.04	0.70 ± 0.10	13.28 ± 0.77	13.13 ± 0.95
	March	42.24 ± 1.68	8.52 ± 0.80	54.61 ± 1.71	1.95 ± 0.08	0.74 ± 0.09	13.35 ± 0.59	13.11 ± 0.70
	April	43.10 ± 2.09	8.66 ± 0.73	55.82 ± 2.54	2.01 ± 0.13	0.79 ± 0.12	13.38 ± 0.68	13.06 ± 0.76
	May	42.64 ± 1.58	8.46 ± 0.80	54.90 ± 1.74	1.97 ± 0.08	0.77 ± 0.11	12.98 ± 0.70	12.53 ± 0.72
	June	42.07 ± 1.17	8.24 ± 0.64	54.05 ± 1.59	1.95 ± 0.06	0.76 ± 0.10	13.12 ± 0.66	13.72 ± 0.68
SIC	February	42.01 ± 1.53	8.17 ± 0.33	53.74 ± 1.41	1.94 ± 0.07	0.75 ± 0.11	12.40 ± 0.37	12.04 ± 0.48
	March	44.97 ± 2.03	8.44 ± 0.53	57.02 ± 2.73	2.11 ± 0.13	0.67 ± 0.05	12.31 ± 0.54	11.88 ± 0.56
	April	43.72 ± 2.63	8.69 ± 0.56	56.58 ± 3.61	2.06 ± 0.18	0.68 ± 0.08	12.36 ± 0.63	11.93 ± 0.69
	May	43.00 ± 1.62	8.17 ± 0.54	55.76 ± 2.57	2.00 ± 0.10	0.65 ± 0.06	12.03 ± 0.84	11.56 ± 0.99
	June	42.61 ± 1.76	8.19 ± 0.49	54.78 ± 2.25	1.98 ± 0.10	0.71 ± 0.08	11.94 ± 0.48	11.47 ± 0.52

Figure 7.1.1 - Trend of average body weight and weight gain in the four breeds (ER, PP, RL and ANC) bred at the Pisa location

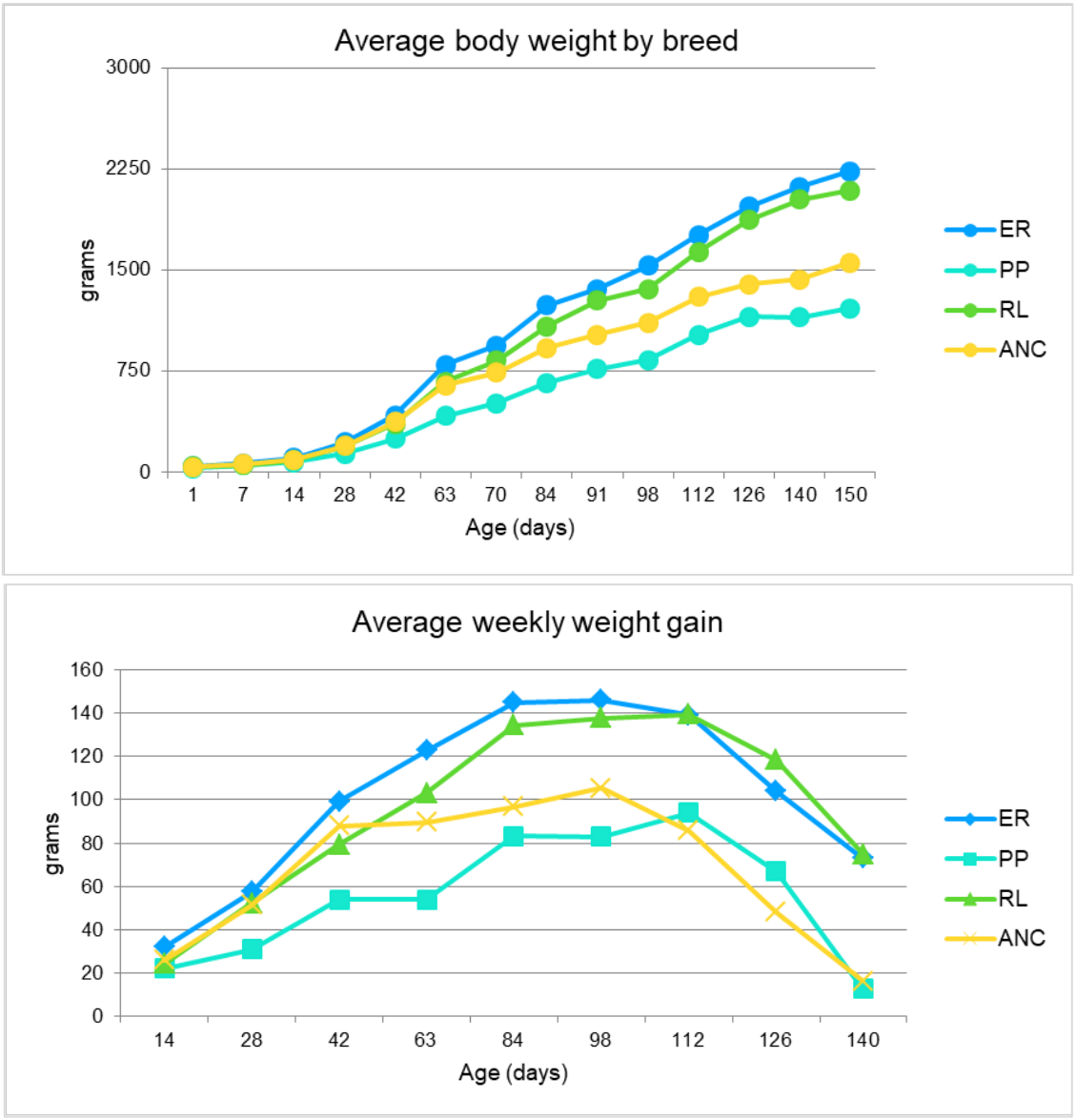


Figure 7.1.2 – Males and females average body weight at 120 days in ER, PP, RL and ANC breeds reared at the two locations

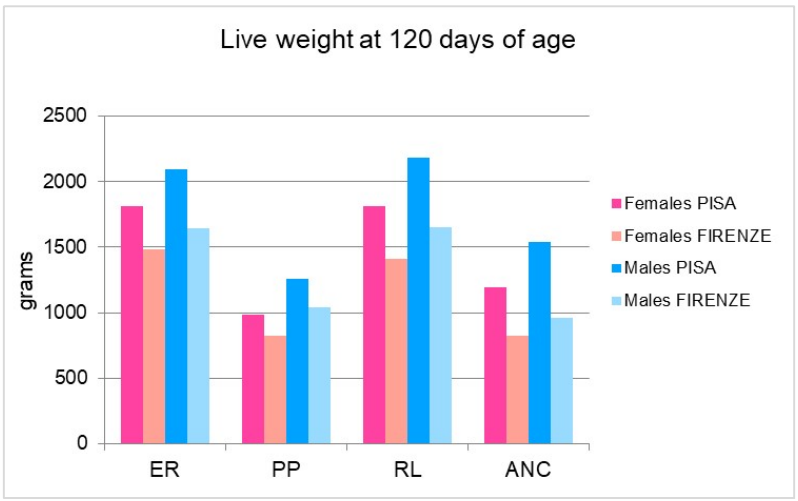


Table 7.2.1 - Results of qualitative and quantitative parasitological analysis in the four breeds and in the two locations (Pisa and Florence): year 2023

Location	Breed		Age (days)				
			15	30	60	90	120
UNIFI	ANC	*Eimeria* spp.	Neg	11012.5 OPG	2775 OPG	1350.0 OPG	150.0 OPG
	ER		Neg	Neg	11900 OPG	1187.5 OPG	125.0 OPG
	PP		Neg	Neg	136550 OPG	2650.0 OPG	225.0 OPG
	RL		Neg	Neg	15750 OPG	2412.5 OPG	175.0 OPG
	ANC	*Capillaria* spp.	Neg	Neg	Neg	Neg	327.3 EPG
	ER		Neg	Neg	Neg	Neg	202.3 EPG
	PP		Neg	Neg	Neg	Neg	387.5 EPG
	RL		Neg	Neg	Neg	Neg	187.5 EPG
UNIPI	ANC	*Eimeria* spp.	Neg	Neg	187.5 OPG	1612.5 OPG	337.5 OPG
	ER		Neg	Neg	200.0 OPG	1687.5 OPG	1087.5 OPG
	PP		Neg	Neg	112.5 OPG	3775.0 OPG	1550.0 OPG
	RL		Neg	Neg	50.0 OPG	465.0 OPG	12.5 OPG

OPG: Oocysts per Gram of feces; EPG: Eggs per Gram of feces

Table 7.2.2 – Evaluation of the four breeds by susceptibility classes of Lactobacilli towards the following antibiotics: ampicillin (AMP), tetracycline (TE), gentamicin (CN), linezolid (LZN), erythromycin (E), streptomycin (S)

		AMP			TE			CN			LZD			E			S		
LOCATION	BREED	R	MS	S	R	MS	S	R	MS	S	R	MS	S	R	MS	S	R	MS	S
Firenze	Robusta Lionata	0%	0%	100%	59%	0%	41%	12%	0%	88%	0%	0%	100%	18%	0%	82%	88%	12%	0%
Pisa	Robusta Lionata	0%	0%	100%	0%	0%	100%	33%	0%	67%	0%	0%	100%	0%	0%	100%	67%	33%	0%
Firenze	Ermellinata di Rovigo	0%	0%	100%	20%	5%	75%	35%	0%	65%	0%	0%	100%	10%	0%	90%	95%	5%	0%
Pisa	Ermellinata di Rovigo	0%	0%	100%	8%	0%	92%	0%	0%	100%	0%	0%	100%	0%	0%	100%	58%	42%	0%
Firenze	Pepoi	0%	0%	100%	12%	0%	88%	0%	0%	100%	0%	0%	100%	12%	0%	88%	88%	12%	0%
Pisa	Pepoi	0%	0%	100%	23%	15%	62%	0%	0%	100%	0%	0%	100%	0%	0%	100%	46%	46%	8%
Firenze	Ancona	0%	0%	100%	0%	0%	95%	0%	0%	100%	0%	0%	100%	0%	0%	100%	90%	0%	5%
Pisa	Ancona	0%	0%	100%	18%	0%	82%	0%	0%	100%	0%	0%	100%	0%	0%	100%	55%	36%	9%
	Robusta Lionata n.20	0%	0%	100%	50%	0%	50%	15%	0%	85%	0%	0%	100%	15%	0%	85%	85%	15%	0%
	Ermellinata di Rovigo n.32	0%	0%	100%	16%	3%	81%	22%	0%	78%	0%	0%	100%	6%	0%	94%	81%	19%	0%
	Pepoi n.30	0%	0%	100%	17%	7%	77%	0%	0%	100%	0%	0%	100%	7%	0%	93%	70%	27%	3%
	Ancona n.31	0%	0%	100%	6%	0%	90%	0%	0%	100%	0%	0%	100%	0%	0%	100%	77%	13%	6%
Firenze		0%	0%	100%	22%	1%	76%	12%	0%	88%	0%	0%	100%	9%	0%	91%	91%	7%	1%
Pisa		0%	0%	100%	15%	5%	79%	3%	0%	97%	0%	0%	100%	0%	0%	100%	54%	41%	5%

R: resistant; MS: medium susceptible; S: sensitive