

EARLY INTERVENTIONS DURING INCUBATION AND IMPACT ON A MUSCLE OF LOCOMOTORY RELEVANCE IN BROILERS

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THERKILDSEN

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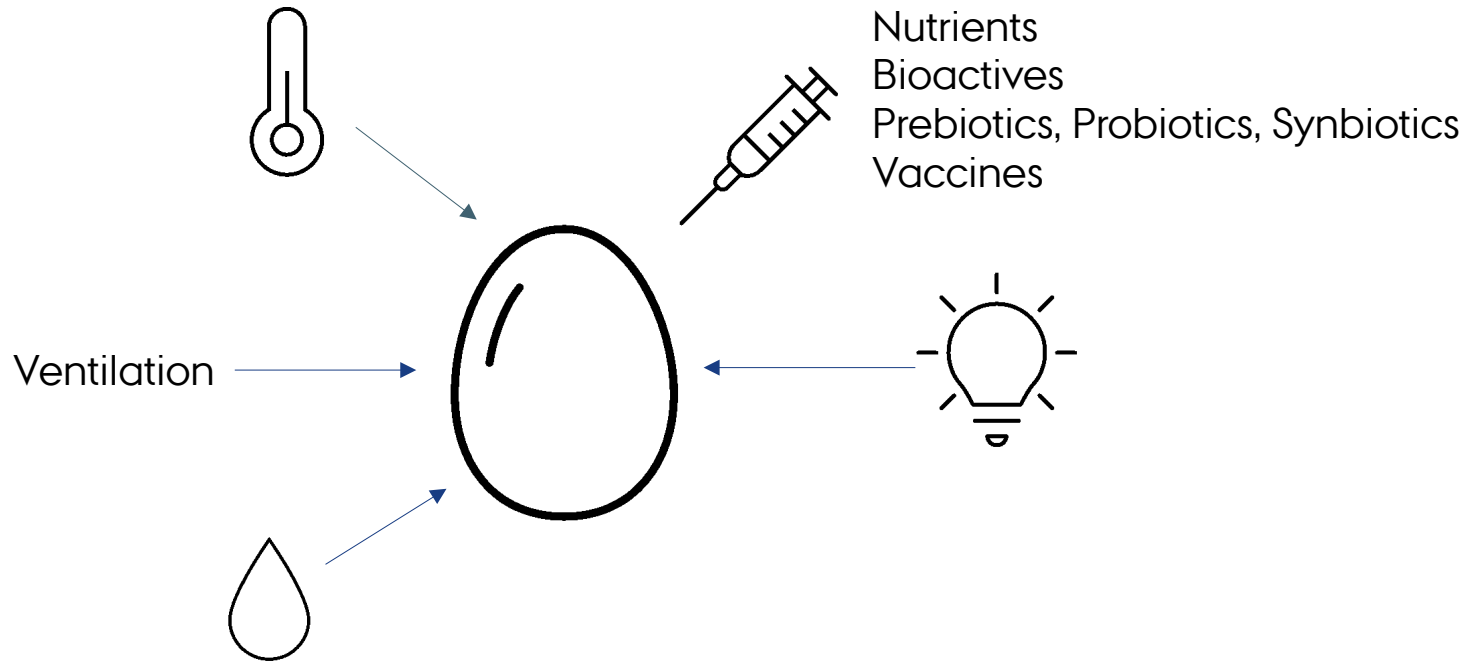
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PRE-HATCH INTERVENTIONS IN BROILER CHICKENS



CHICKEN MUSCLE GROUPS

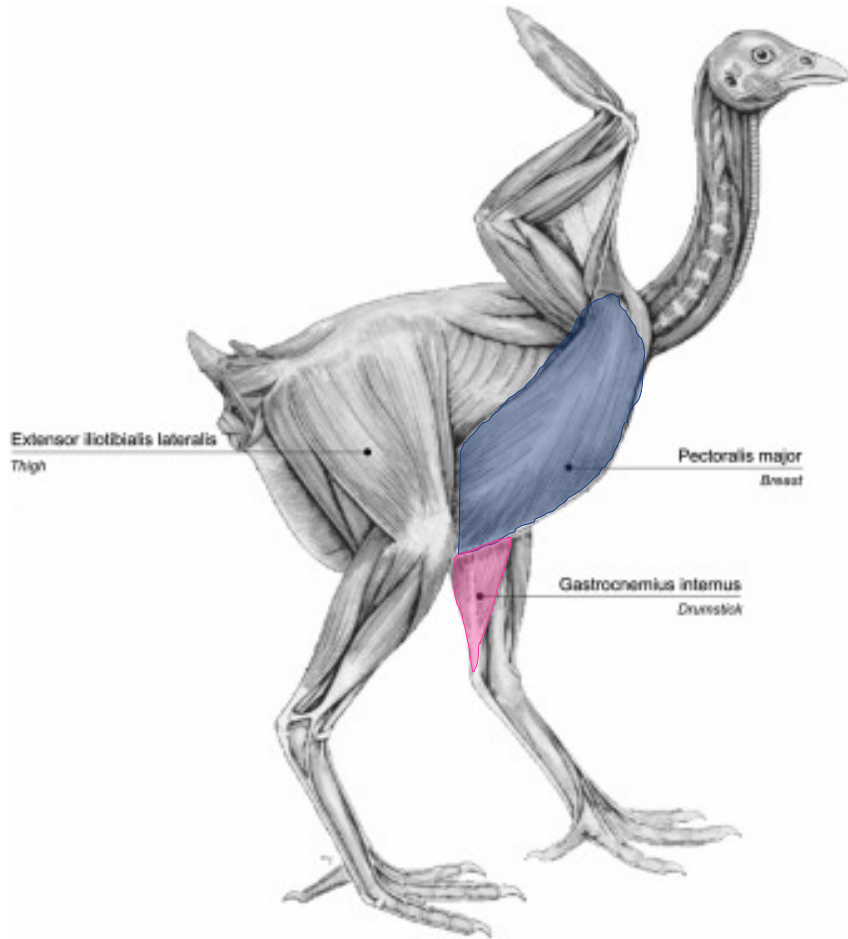


Fig.: Selected muscles in the chicken
(adapted from Baldi et al. 2021)

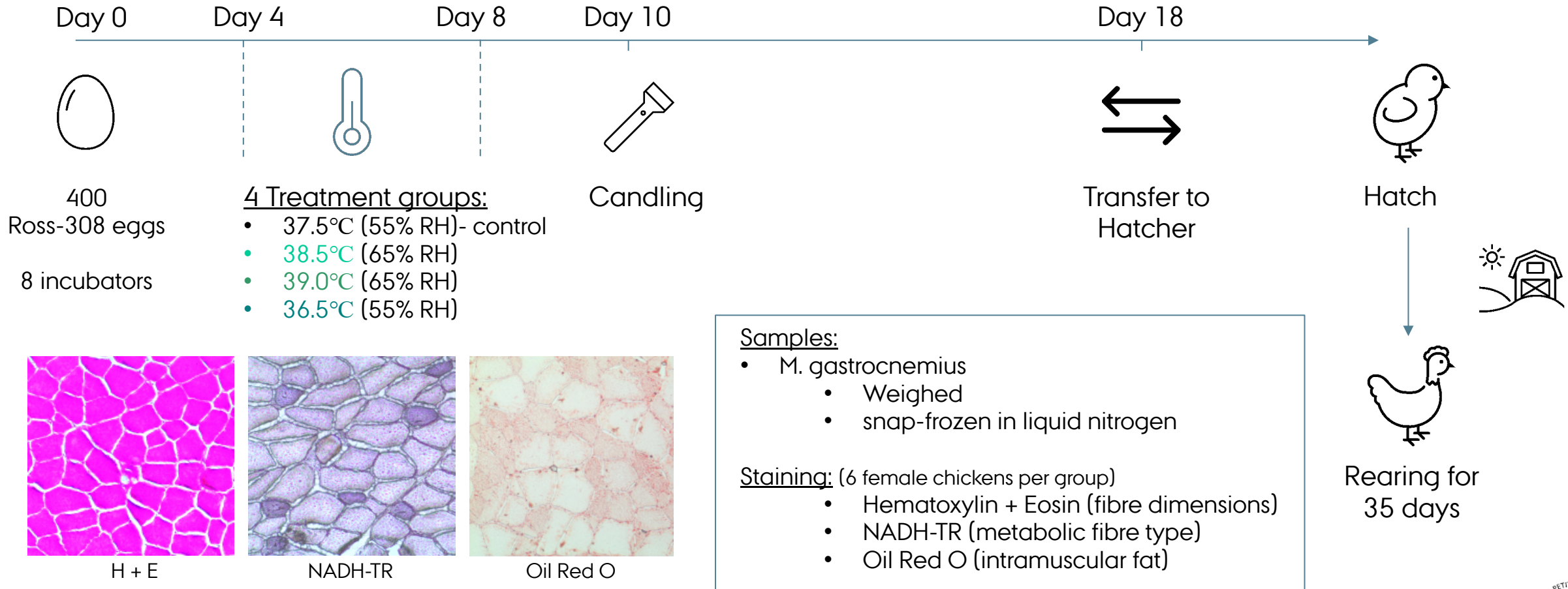
M. Pectoralis major

- Main target for genetic selection
 - Originally for flying
- Higher temperatures in incubation trigger proliferation (Yalcin et al. 2022, Piestun et al. 2015, Piestun et al. 2011)
- Fibre traits affected by *in ovo* injection of pre-, pro-, and synbiotics on day 12 (Bogucka et al. 2022, Dankowiakowska et al. 2019, Maiorano et al. 2017, 2012)

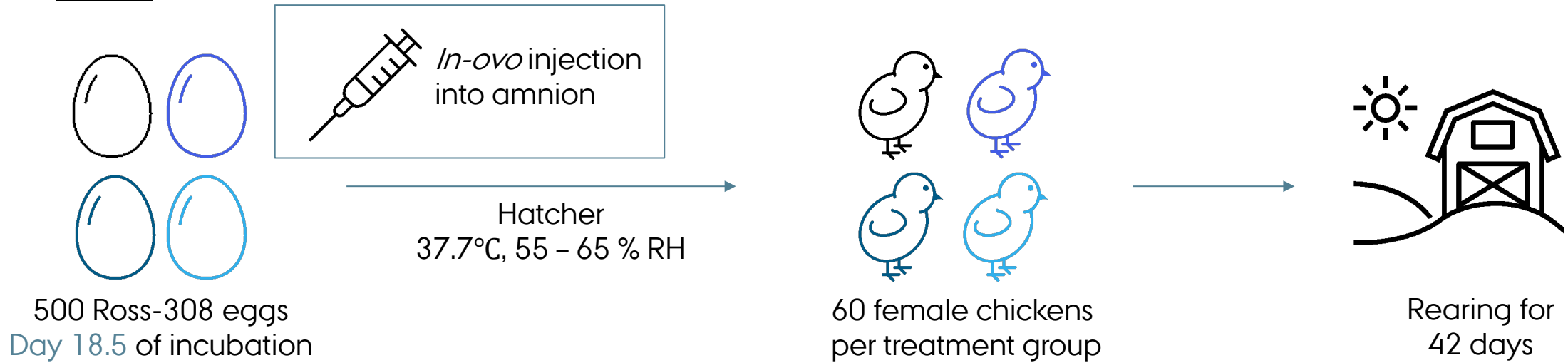
M. Gastrocnemius

- No specific selection
- Functional relevance for walking (flexor of the knee)
- Proliferation in layer chicken embryos exposed to higher incubation temperatures (Hammond et al. 2007), effect in broiler embryos not clear (Al-Musawi et al. 2012)
- Shift in ratio between M. gastrocnemius and M. pectoralis in response to incubation temperature change (Oksbjerg et al. 2019)
- Not investigated in relation to *in ovo* programming

EXPERIMENT 1: EARLY INCUBATION TEMPERATURE



EXPERIMENT 2: IN OVO MICROBIAL PROGRAMMING



4 Treatment Groups:

A _{vac}	B _{pro}	C _{pre}	D _{pro+pre}
Vaccine only	Vaccine + Probiotic	Vaccine + Prebiotic	Vaccine + Probiotic + Prebiotic
	Bifidobacterium lactis	Astragalus polysaccharides	Astragalus polysaccharides + B. lactis

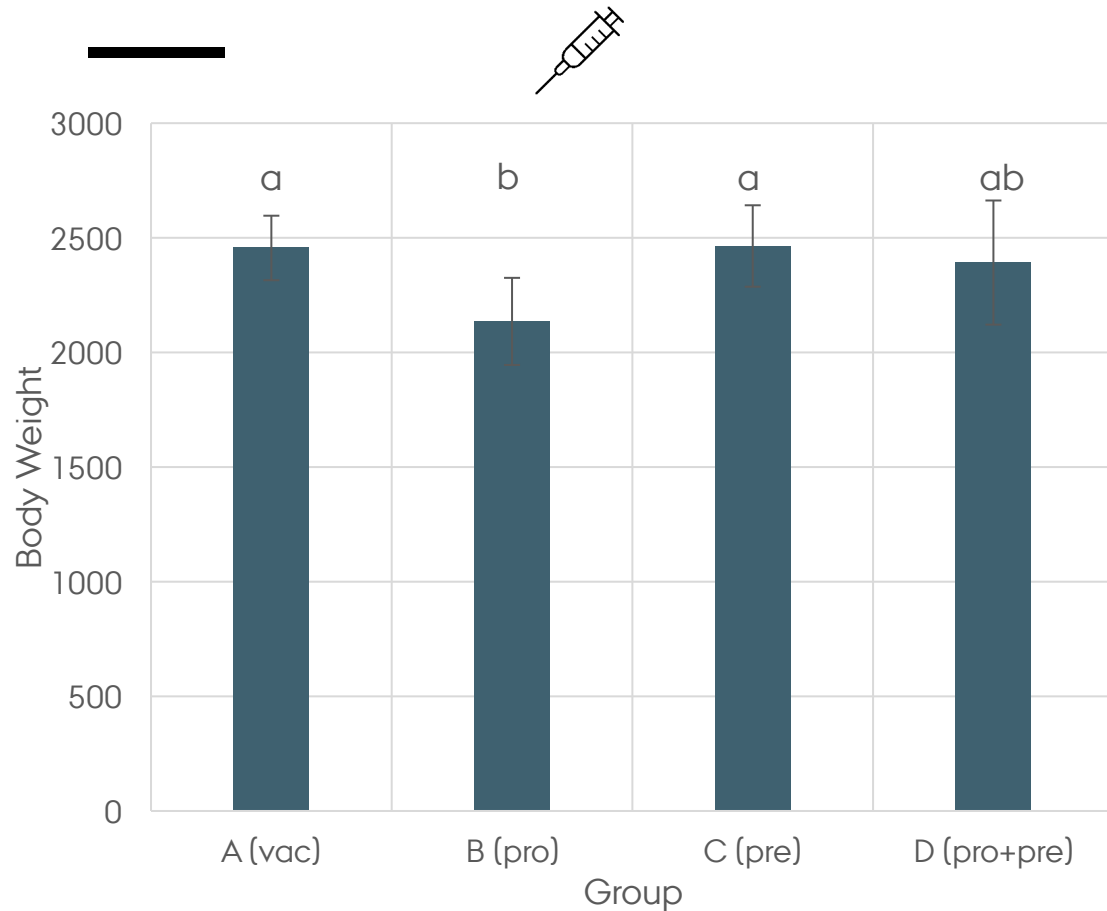
Samples:

- M. gastrocnemius from 6 chickens per group
 - Weighed
 - snap-frozen in liquid nitrogen

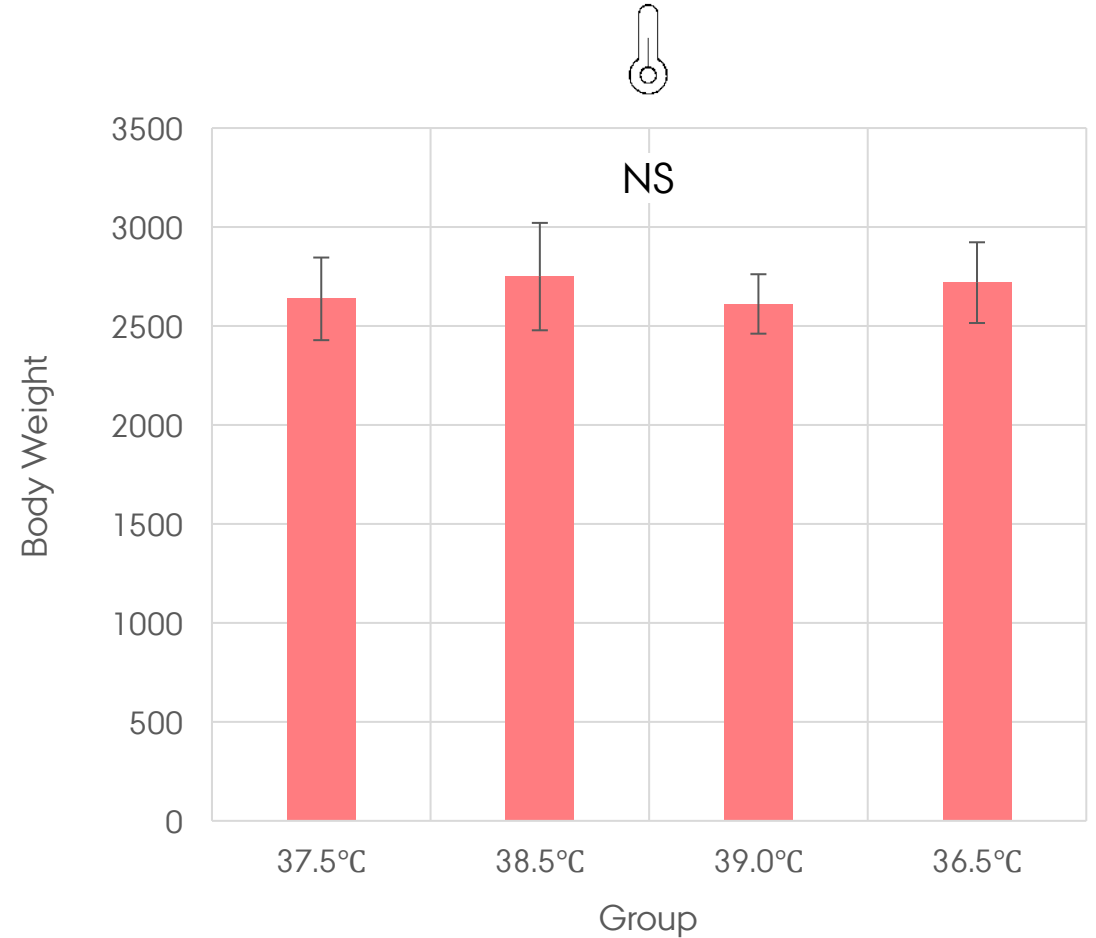
Staining:

- Hematoxylin + Eosin (fibre dimensions)
- NADH-TR (metabolic fibre type)
- Oil Red O (intramuscular fat)

BODY WEIGHTS AT 5 AND 6 WEEKS

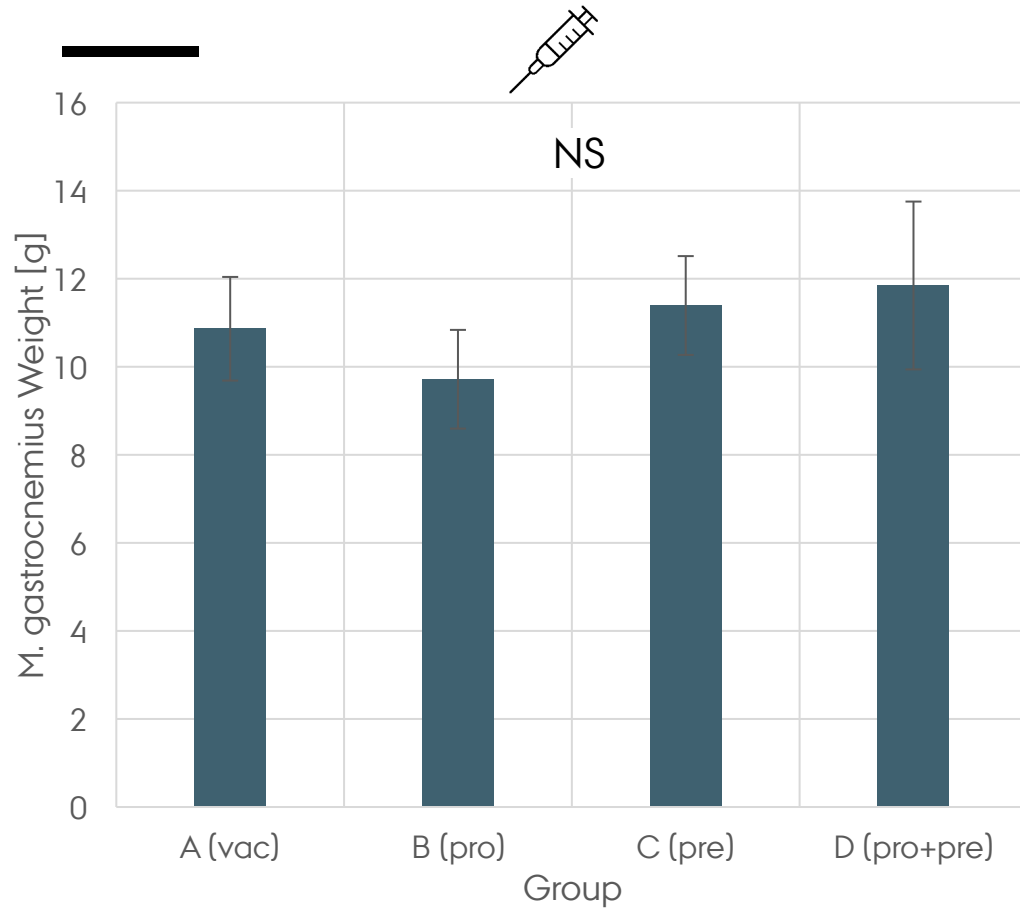


Body weight on day 42 post-hatch by treatment (A: vaccine only, B: vaccine + probiotic, C: vaccine + prebiotic, D: vaccine + probiotic + prebiotic), $p = 0.03$



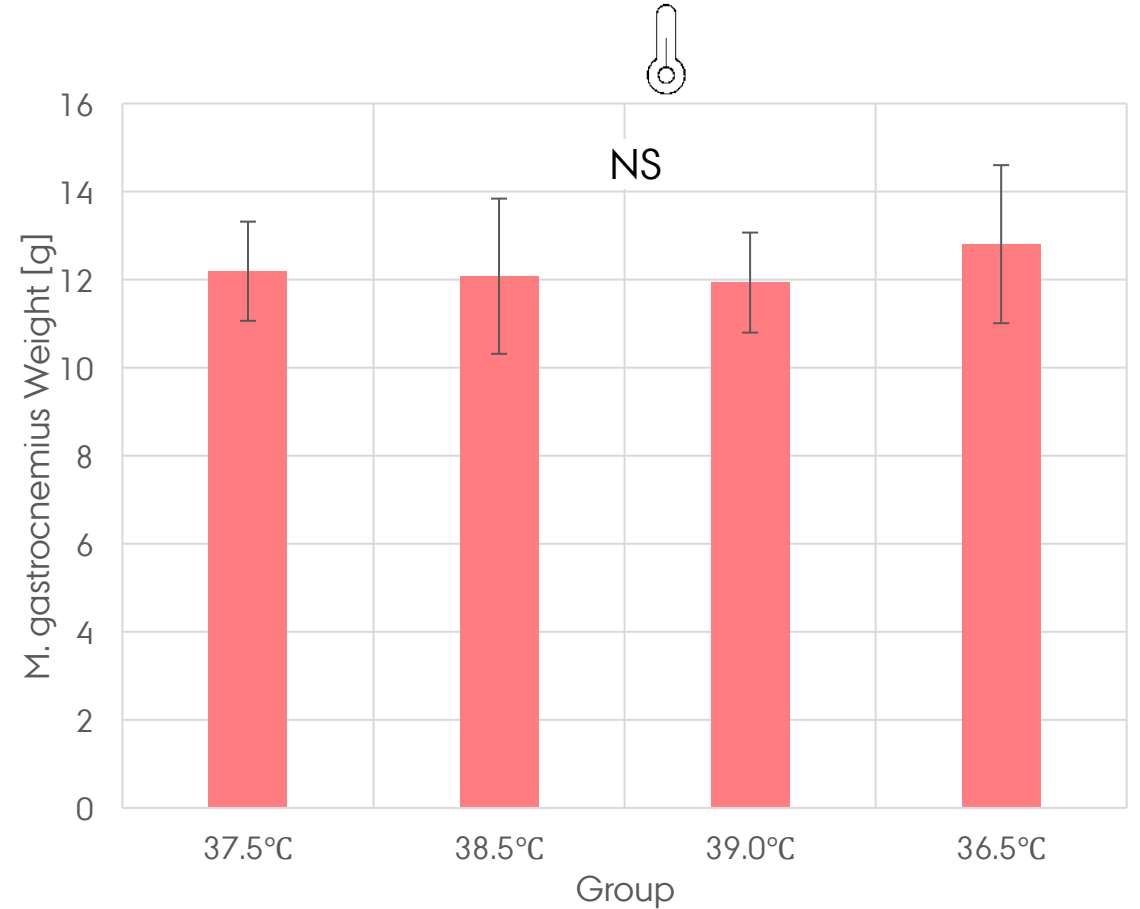
Body weight on day 35 post-hatch by temperature treatment

MUSCLE WEIGHTS



M. gastrocnemius weight at hatch and on day 42 post-hatch by treatment

(A: vaccine only, B: vaccine + probiotic, C: vaccine + prebiotic, D: vaccine + probiotic + prebiotic)

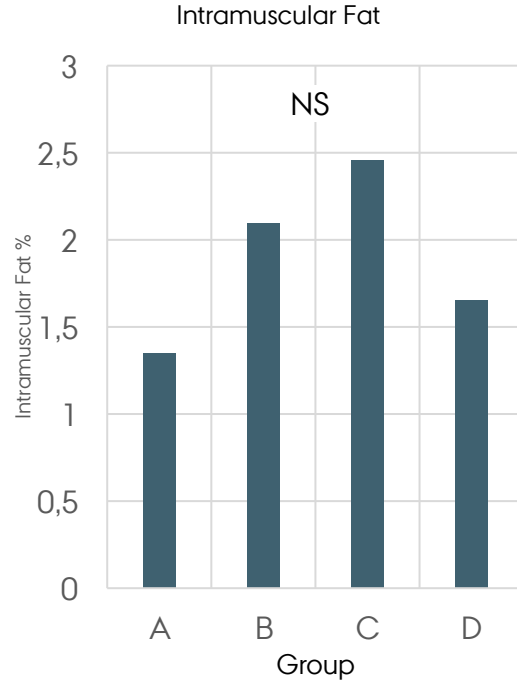
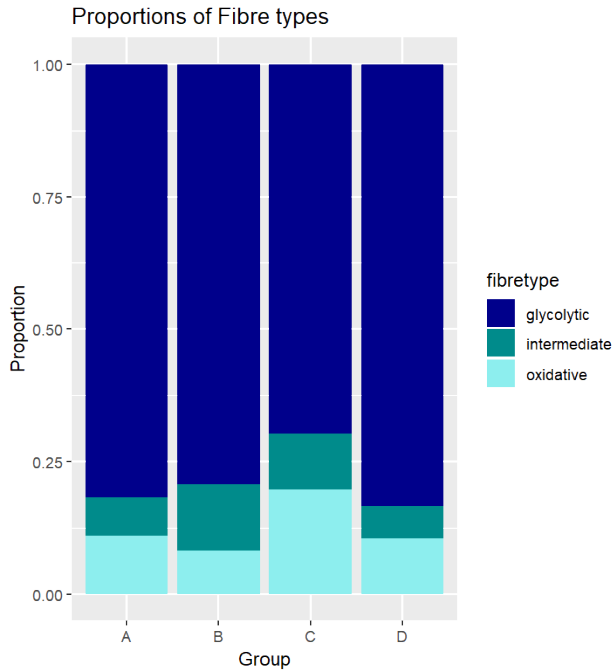


M. Gastrocnemius weight at hatch and on day 35 by temperature treatment

MUSCLE FIBRE TRAITS

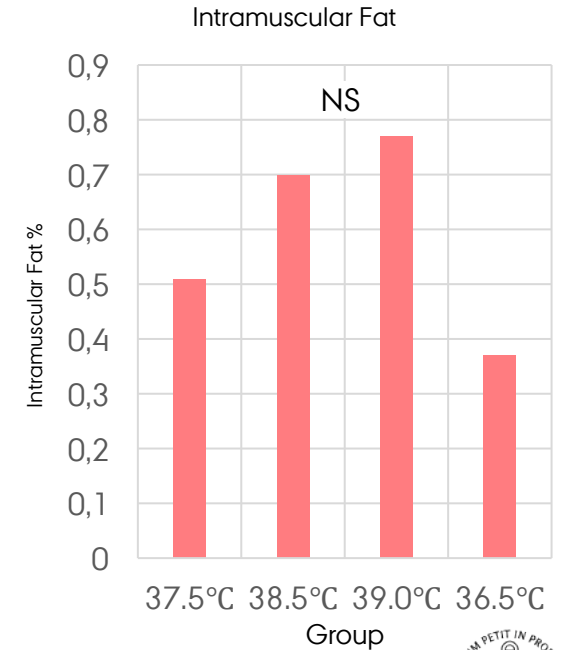
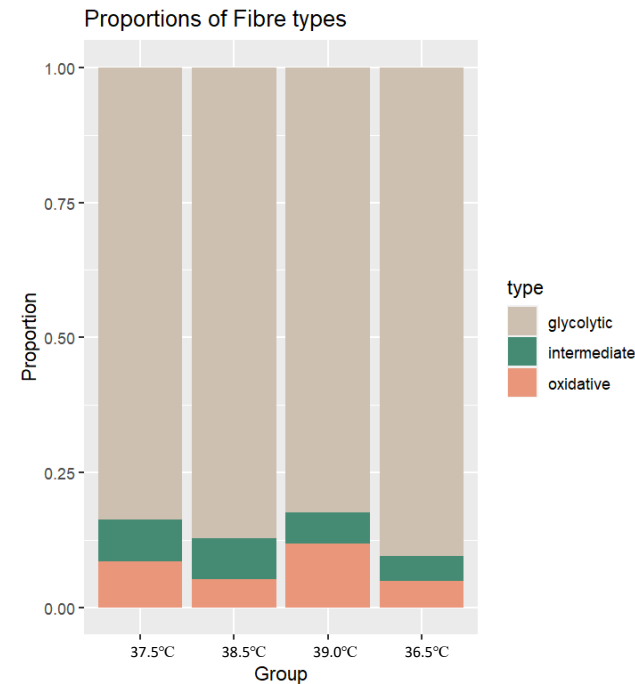
M. Gastrocnemius fibre traits by group
(A: vaccine only, B: vaccine + probiotic, C: vaccine + prebiotic, D: vaccine + probiotic + prebiotic)

	Treatment				P-value
	A _{vac}	B _{pro}	C _{pre}	D _{pro+pre}	
Fibre Diameter (µm)	38 ± 4.8	37 ± 2.4	40 ± 5.9	42 ± 3.9	0.25
Fibre number /mm ²	517 ± 102.7	537 ± 61.1	478 ± 123.3	442 ± 83.8	0.34



M. Gastrocnemius fibre traits by temperature treatment

	Treatment				P-value
	36.5°C	37.5°C	38.5°C	39.0°C	
Fibre CSA (µm ²)	2684 ± 501	2819 ± 400	2591 ± 532	2608 ± 313	0.80
Fibre number /mm ²	206 ± 37.6	228 ± 52.1	220 ± 51.5	230 ± 51.9	0.85



DISCUSSION AND CONCLUSION

- Muscle microstructure of M. gastrocnemius after in-ovo microbial programming has not been investigated before
- Observations from the breast muscle (injection day 12) have not been found in M. gastrocnemius
- The present results do not indicate changes of functional relevance in the microstructure after application on day 18
- M. gastrocnemius has been shown to proliferate in embryos subjected to higher temperature in early incubation (Hammond et al. 2007, Al-Musawi et al. 2012) – this does not seem to last until slaughter age
- Differences in metabolism after incubation temperature changes (Krischek et al. 2016, 2018) seem to be transient as well
- no changes in microstructure that indicate functional advantage or disadvantage

TAKE-HOME MESSAGE

Neither in-ovo injection with prebiotics, probiotics or synbiotics, nor early incubation temperature manipulation have disadvantageous effects on M. gastrocnemius, a muscle of postural relevance for the chicken. From that point of view, they are suitable for other applications, like gut programming, thermoregulation etc.

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