Effect of maternal probiotic and/or increased tryptophan supplementation to piglets on the functioning of the stomach at weaning

D.P. Kiernan[a], J.V. O'Doherty[b], M. Ryan[a], T. Sweeney[a]

[a]School of Veterinary Medicine, University College Dublin, Belfield, Dublin 4, Ireland

[b]School of Agriculture and Food Science, University College Dublin, Belfield, Dublin 4, Ireland

An Roinn Talmhaíochta, Bia agus Mara
Department of Agriculture, Food and the Marine

Weaning

- Pigs are weaned prematurely at 3-4 weeks on commercial farms – to maximize pigs produced per sow per year

- Stressful time in the life of a commercial pig

- Issues with early weaning
  - Immature digestive system
  - Immature Immune system

Stomach immaturity at weaning time

- Poorly developed gastric secretions at weaning
    - HCL
    - Pepsin

→

- Activating enzymes involved in protein digestion
- Kill ingested pathogens

- Immediate post-weaning period can be characterized by:
    - Poor protein digestion
    - Increased susceptibility to pathogenic infection

→

- Poor performance
- Infection
- Potential mortality

- Strategies to maintain low gastric pH during post-weaning period:
    - Diet acidification
    - Reduced diet acid-buffering capacity

Promoting the development of acid secretion in the pig pre-weaning?

Target the development of acid secretion in the pre-weaned pig

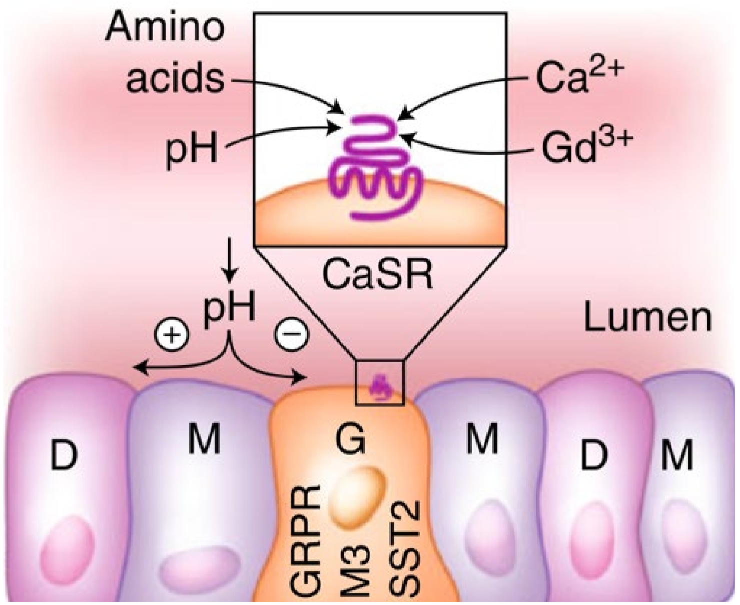
1) Tryptophan

- Suckling pig:
  - Lactose fermentation is the primary sources of acidity in the stomach
  - But alternative source of acidity delays the maturation of acid secretory capacity (Cranwell et al., 1976)

- The calcium-sensing receptor (CaSR) involved in the regulation of acid secretion in response to luminal pH

- CaSR expressed on parietal cells and G-cells

- In *ex-vivo* pig stomach tissue, tryptophan acts via the CaSR to increase the expression of gastrin and acid secretion activity (Xian et al., 2018)



(Goo et al., 2010)

Could increased tryptophan pre-weaning increase stimulation & development of acid-secreting cells?

Target the development of acid secretion in the pre-weaned pig

2) Maternal probiotic

- Probiotic supplementation to suckling piglets increased the number of endocrine cells in the stomach (Babińska et al., 2005)

- Exposure of piglets to a complex microbiota promotes the functional maturation of the fundic gland region (Trevisi et al., 2017)

- The sow is the major microbial source in early life (Chen et al., 2018)

- Maternal *Bacillus subtilis* & *Bacillus amyloliquefaciens* supplementation increased piglet birth and in combination with piglet supplementation it increased weaning weights (Konieczka et al., 2023, Mazur-Kuśnirek et al., 2023)

Could maternal probiotic supplementation promote the functional maturation of the stomach?

Objective:

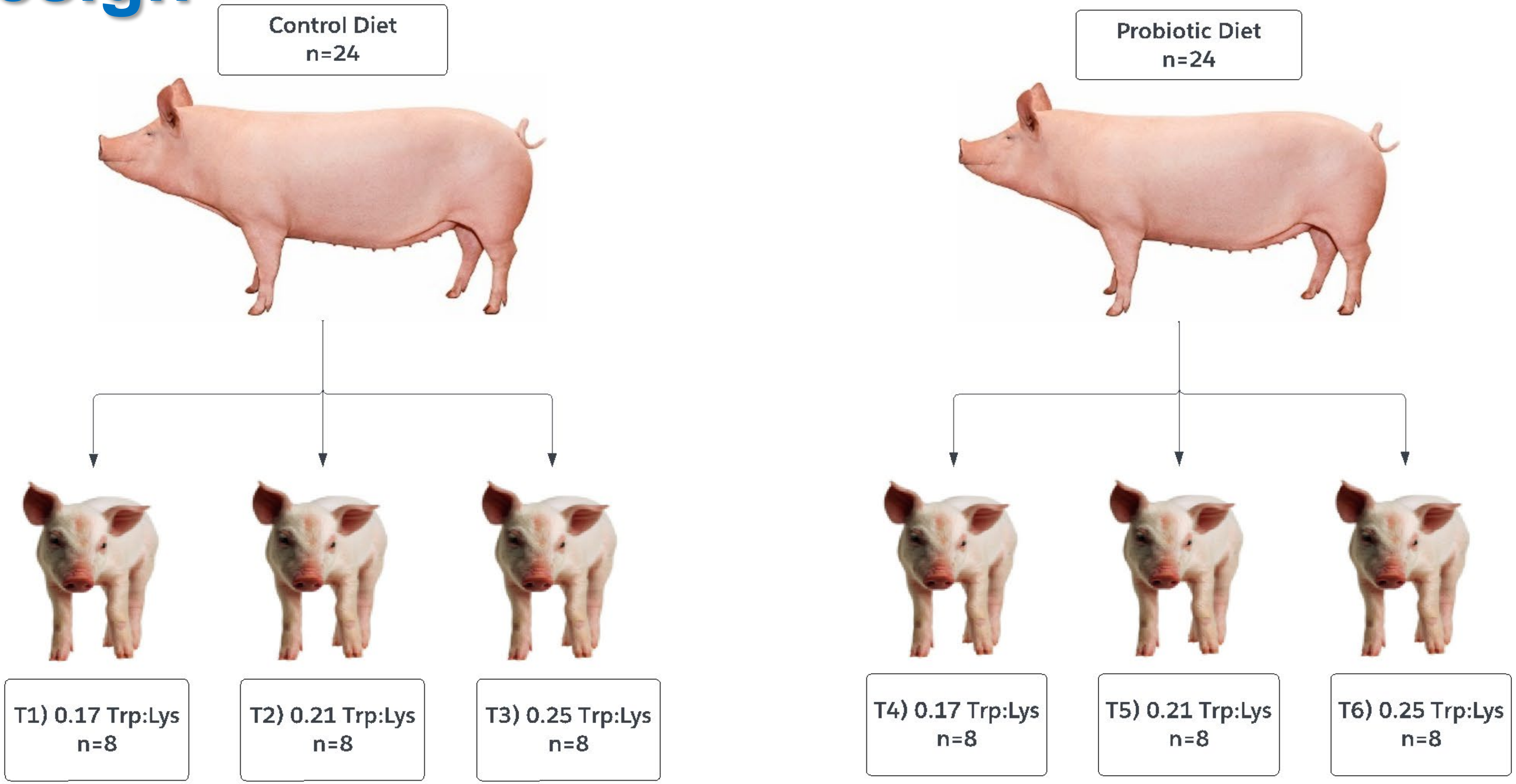
To investigate the effects of maternal probiotic and/or increased tryptophan to piglets on the functioning of the stomach at weaning

Hypotheses:

Increased dietary tryptophan to the piglet would increase the number and/or activity of acid secreting cells in the stomach of piglets at weaning

Maternal probiotic supplementation would promote the functional maturation of the stomach of piglets at weaning

Trial design

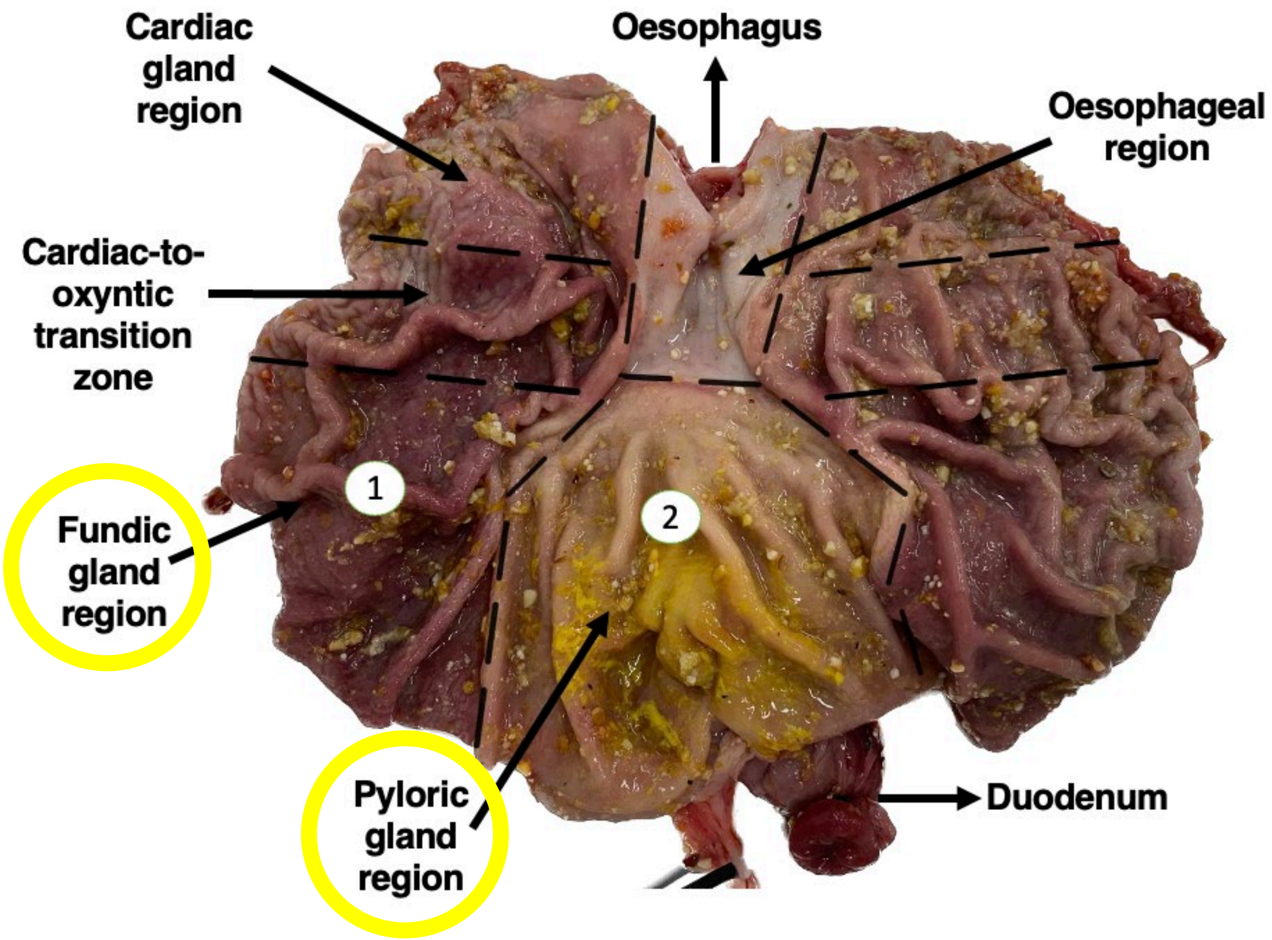


- 2x3 factorial design
- Maternal probiotic (*Bacillus subtilis* & *Bacillus amyloliquefaciens*) supplemented from day 83 of gestation until weaning
- Standard ileal digestible lysine content of the piglet diets was 13 g/kg
- Piglet diets fed from day 8 of lactation until weaning on day 26 of lactation

Materials and methods

48 pigs (1 pig per litter) were sacrificed on day of weaning:

- pH of stomach contents measured using pH meter
- Full and empty stomach weights were recorded
- Mucosal samples from the fundic and pyloric gland regions taken for RNA extraction and subsequent QPCR for expression of key genes (location 1 and 2)
- Data was analysed using PROC GLM function in SAS



(Kiernan et al., 2023)

Selection of genes for QPCR analysis

Fundic gland region	
Acid secretion	*ATP4A*
	CLIC6
	KCNE1
	KCNQ1
Acid cell stimulation	*CASR*
	CHRM3
	CCKBR
	HDC
	HRH2
Acid cell inhibition	*SST*
	SSTR2
Digestive enzyme production	*CHIA*
	LCT
	PGA5
Ghrelin production	*GHRL*
Inflammation	*CXCL8*
	IL22
	TNF
Mucosal defense & Immune response	*MUC1*
	MUC5AC
	MUC6
	TLR4
	AHR

Pyloric gland region	
Acid cell stimulation	*CASR*
	GAST
Acid cell inhibition	*SST*
Inflammation	*CXCL8*
	TNF
Mucosal defense & Immune response	*PIGR*
	MUC5AC
	MUC6
	TLR4
	AHR

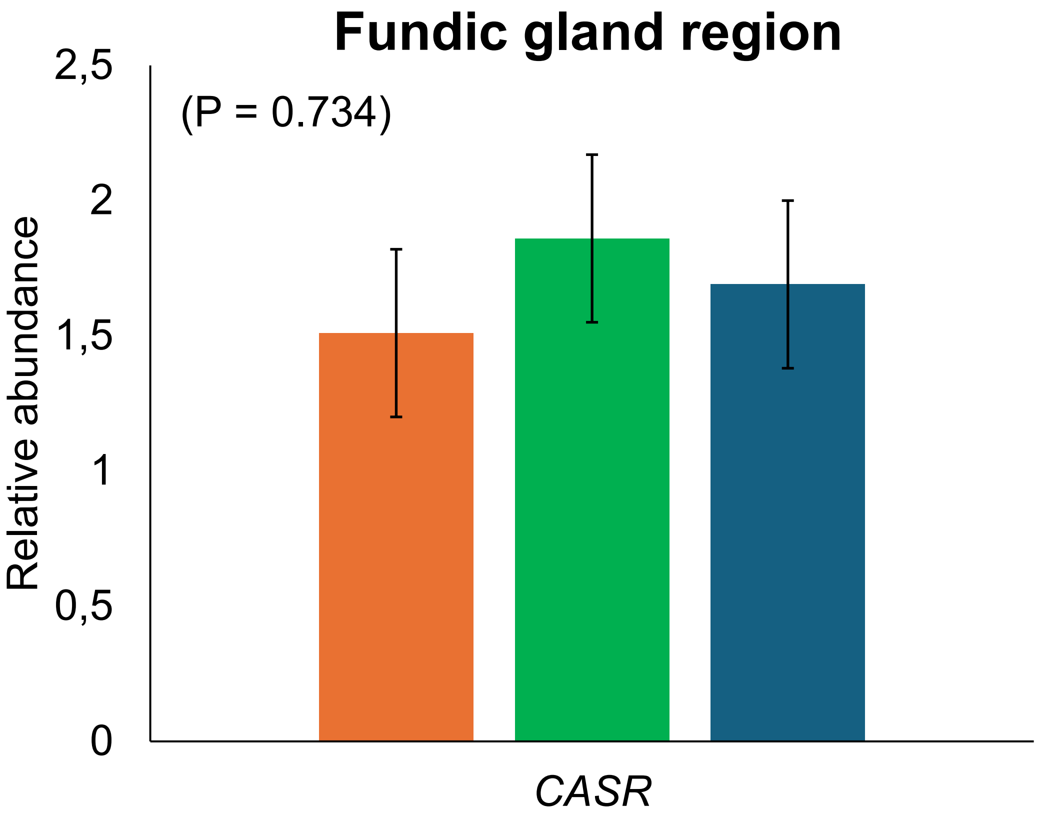
- Genes were selected based on previous work by the research group (Kiernan et al., 2023)

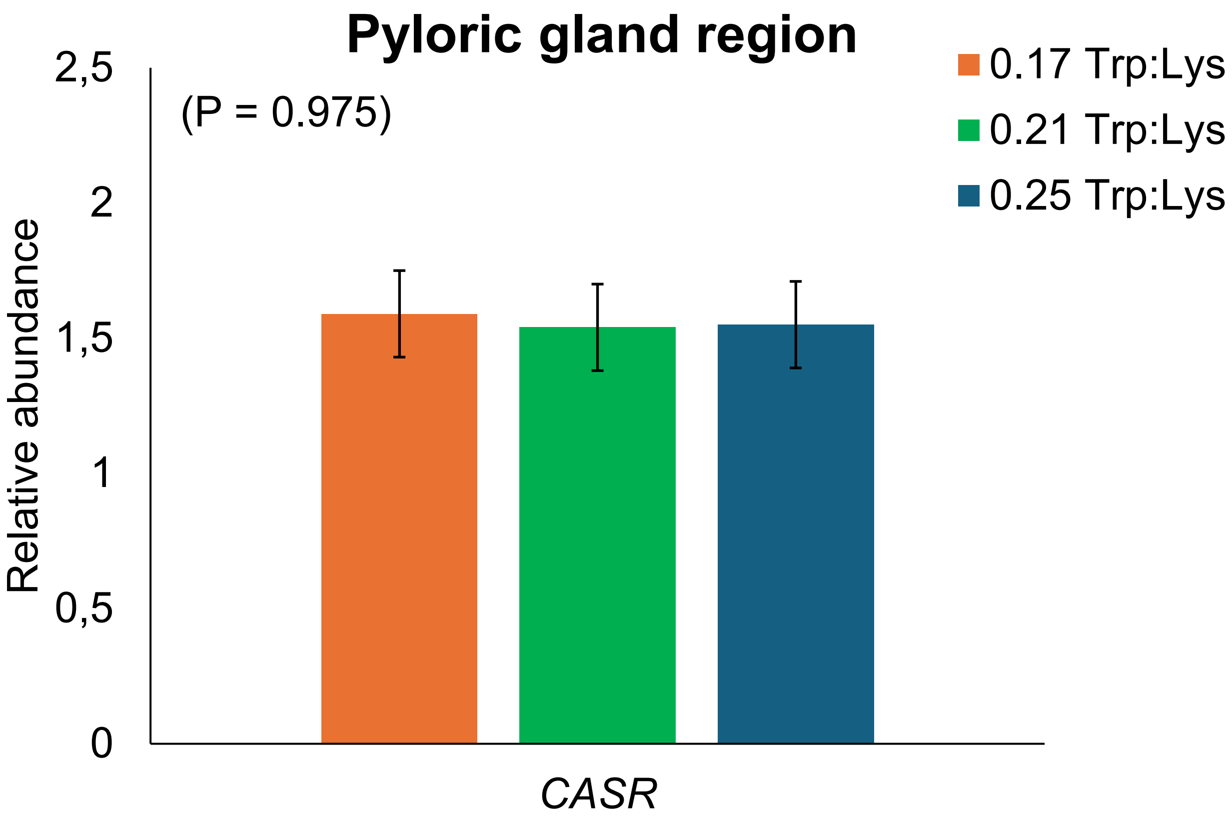
Results – bodyweight, pH, stomach weight, creep intake

	Maternal		SEM	Creep			SEM	*p*-value	
	Control	Probiotic		0.17 Trp:Lys	0.21 Trp:Lys	0.25 Trp:Lys		Maternal	Creep
Bodyweight (kg)	8.48	8.40	0.33	8.15	8.94	8.22	0.42	0.864	0.337
Gastric pH	3.7	3.5	0.2	3.5	3.9	3.4	0.2	0.397	0.304
Full stomach weight (g)	143.4	160.0	8.8	155.9	153.2	146.0	10.6	0.188	0.790
Empty stomach weight (g)	74.0	79.0	1.9	77.8	74.7	76.9	2.5	0.082	0.651
Empty stomach to body weight ratio (%)	0.89	0.98	0.04	0.98	0.87	0.96	0.05	0.127	0.242
Total litter creep intake (kg)	0.76	0.96	0.14	0.97	0.90	0.72	0.17	0.227	0.536

- No maternal x creep interaction
- Low level of creep intake across all groups: <1kg per litter

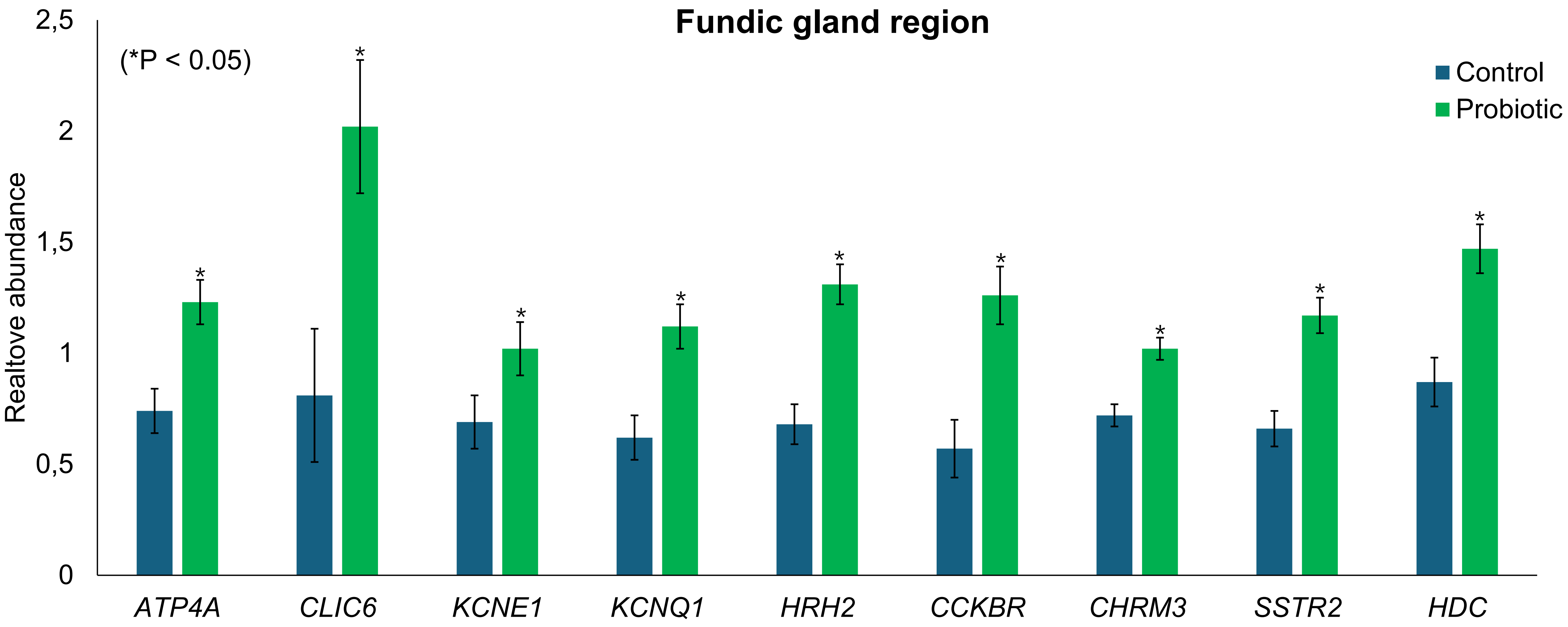
Creep tryptophan effect on *CASR* expression





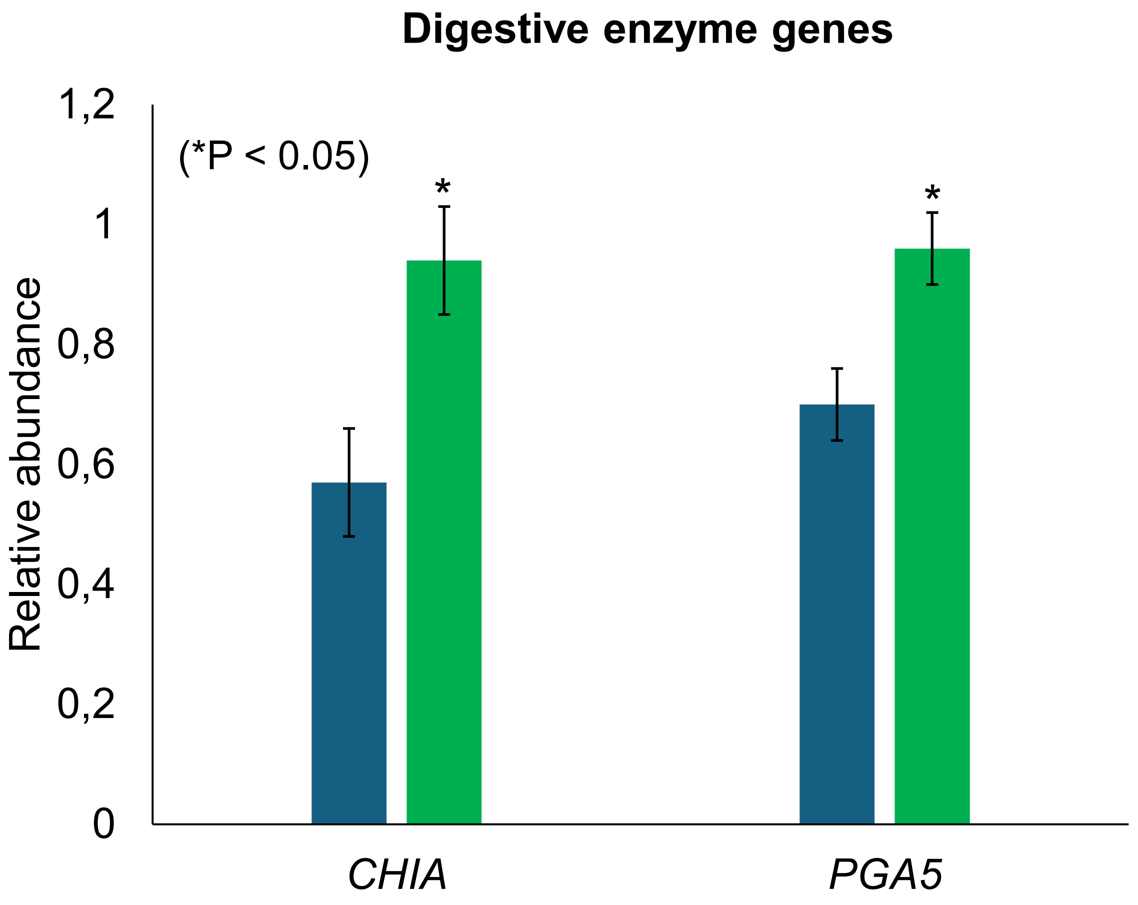
- No effect of tryptophan level on gene expression
- Creep intakes were very low and may have impacted potential effects

Maternal probiotic effect – acid secretion genes



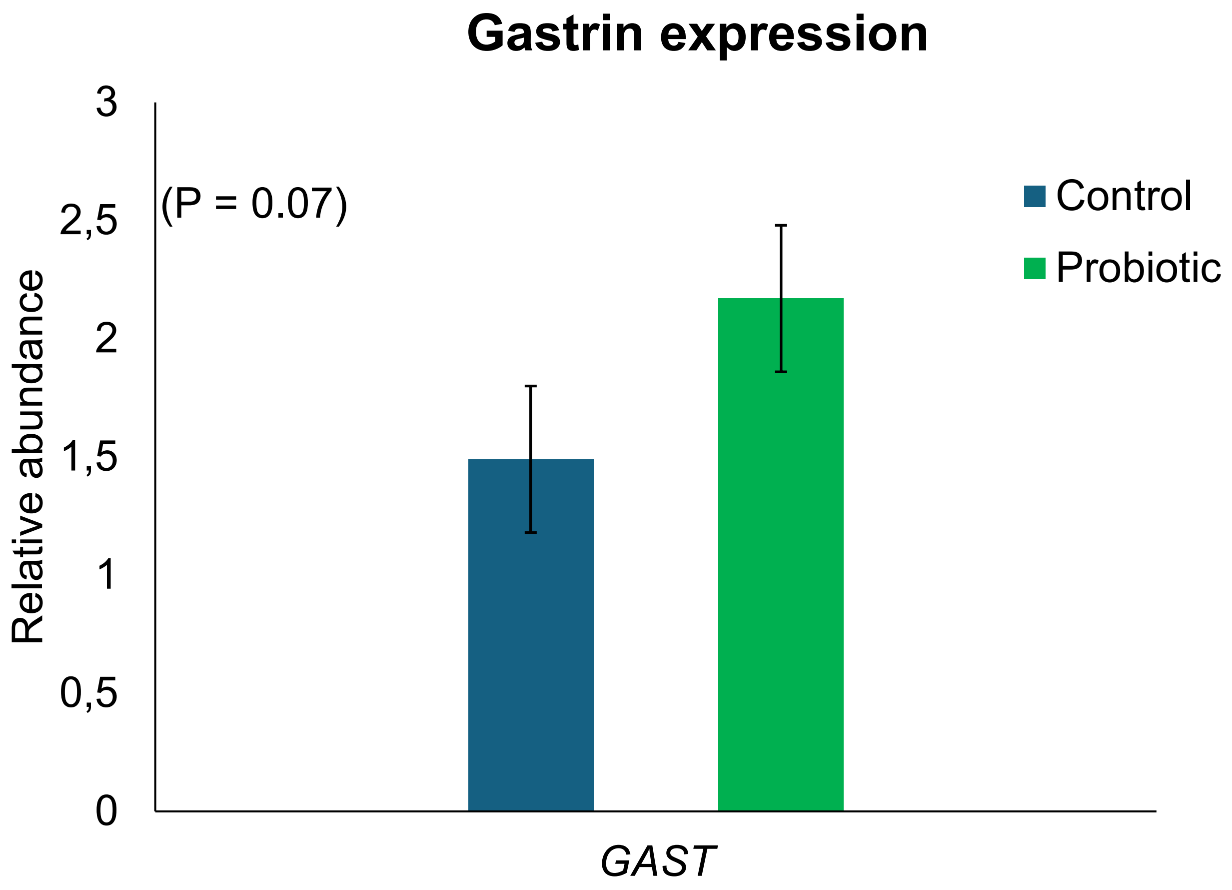
- 9 of the 11 genes related to the regulation and process of acid secretion were increased in offspring from probiotic sows

Maternal probiotic effect – fundic gland region



- No differential expression of the mucins, *MUC1* or *MUC5AC* (P > 0.05)
- No differential expression of inflammation related genes *CXCL8*, *IL22*, *TNF* or *AHR* (P > 0.05)

Maternal probiotic effect – pyloric gland region



- Maternal probiotic supplementation tended to increase expression of gastrin in the pyloric gland region (P = 0.07)
- No differential expression of somatostatin, inflammation or mucosal defense genes in the pyloric gland region (P > 0.05)

Conclusion

Maternal probiotic supplementation:

- Gene expression indicates a more functionally mature stomach in terms of acid and enzyme secretion
- Tended to increase empty stomach weight
- No effect on gastric pH
- This enhanced stomach maturity could improve health and performance in the post-weaning period

Level of tryptophan in the pre-weaning creep diet:

- Did not affect gastric pH, stomach weights or genes expression in the stomach
- Limited creep intake could be a confounding factor in the current study

No maternal probiotic × creep tryptophan interaction on any measured parameters

THANK YOU

Email: Dillon.Kiernan@ucdconnect.ie

UCD DUBLIN

SETU Ollscoil Teicneolaíochta an Oirdheiscirt South East Technological University

Teagasc Agriculture and Food Development Authority

An Roinn Talmhaíochta, Bia agus Mara Department of Agriculture, Food and the Marine