GROWTH PROMOTION BY DIETARY PEPTIDES

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In previous experiments, Peptiva had utility at improving feed intake and this effect was especially important during the anorexia that accompanied stimulation of the immune system.

Purpose: Examine the growth promoting properties of Vitech's peptide product (Peptiva) during authentic infectious challenges. We will also determine if the improved food intake augments the chick's ability to supply its immune system with nutrients, thereby improving its immune response to the infectious challenges.

EXPERIMENTAL DESIGN

Independent variables (treatments):

6 DIETARY TREATMENTS:

1) Control diet devoid of Peptiva.
2) Control diet plus Peptiva substituted for casein at 0.5%.
3) Control diet devoid of Peptiva + infection with *Eimeria acervulina* (cocci challenge)
4) Control diet plus Peptiva + infection with *Eimeria acervulina* (cocci challenge)
5) Control diet devoid of Peptiva + infection with *Escherichia coli* (*E. coli* challenge)
6) Control diet plus Peptiva + infection with *Escherichia coli* (*E. coli* challenge)

Dependent variables (measurements)

Rate of growth

Feed intake

Immunoglobulin response to *Eimeria* (treatments 3 and 4) or *E. coli* (treatments 5 and 6) at 10 days post challenge.

Delayed type hypersensitivity response to response to *Eimeria* (treatments 3 and 4) or *E. coli* (treatments 5 and 6) at 14 days post challenge.

Experimental procedure:

On the day of hatching, one hundred eighty chickens (Cobb broiler chickens) were assigned to the 2 dietary treatments with 5 pens per treatment group and 6 chicks per pen. Beginning on the first day post-hatch, chickens were fed experimental diets and water ad libitum. Chickens were weighed and feed intake recorded weekly.

A corn-soybean meal diet devoid of animal byproducts that exceeds all requirements suggested by NRC (Nutrient Requirements of Poultry, 1994) was used see below). Peptiva was substituted for an identical amount of casein so that all diets were iso-nitrogenous and iso-energetic.

When chicks were 14 days of age, pathogen challenges were administered. Desporulated *Eimeria acervulina* oocysts (supplied by USDA, Beltsville, MD) were gavaged into the crop of each chick in treatments 3 and 4. 10^5 CFU of a weakly pathogenic strain of *E. coli* (American Type Culture #14283d) were injected i.v. into chicks in treatments 5 and 6.

Seven days following the challenge, chicks were bled and serum levels of immunoglobulin (IgM and IgG) levels were determined by agglutination assay with and without 2ME. Delayed-type hypersensitivity was quantified by examining the infiltration of leukocytes into the wing web following injection of heat killed *Eimeria* (treatments 3 and 4) or *E. coli* (treatments 5 and 6).

The intestines were removed from two chicks per pen. A segment of approximately 1.5 cm in length from the mid-point of the duodenum was flushed with saline and fixed in 10% buffered formalin (pH 7.0). Fixed intestinal samples were embedded with paraffin, sectioned (5 μm), stained with hematoxylin-eosin, and mounted (California Veterinary Services, West Sacramento California). The histological sections were evaluated for: thickness of the lamina propria; villus height – from the base of the lamina propria to the apex of the villus; crypt depth between adjacent villi. Morphometric data was collected on 10 different villi per animal on each of two different serial sections. Measurements were made and analyzed by computer-aided light microscopic analysis at magnifications between 10 to 100x using Image-Pro-Plus analysis software for the PC.

Data from the 6 treatment groups were analyzed using the GLM procedure of SAS for two-way ANOVA and main effects and interactions were examined for significant treatment differences.

BASAL DIET	
Corn Dent Yel grain	553.77
Soybean seeds	346.15
Fat, animal	54.71
Calcium phos.	17.62
Limestone, ground	13.36

salt	4.58
DL-methionine 99%	3.16
Mineral mix - NRC	2.50
Vitamin mix - NRC	2.50
Choline chloride	0.75
ferrous sulfate	0.50
L-lysine 95%	0.40
Total	1000.00

RESULTS

TABLE 1. Effect of Peptiva on performance of healthy broiler chicks (0-7 days and 7-14 days)

Age	Gain		Intake		Efficiency	
	Control	Peptiva	Control	Peptiva	Control	Peptiva
0-7	137.2±2.4	140.0±2.7	169.8±3.3	170.6±2.0	0.81±0.02	0.82±0.05
7-14	262.7±2.6	268.0±12.7	348.4±2.8	351.8±8.4	0.75±0.03	0.76±0.02

Dietary treatment did not significantly affect any parameter (P>.25)

TABLE 2. Effect of Peptiva on performance of broiler chicks challenged with coccidia (14-20 days)

Challenge*	Gain[1]		Intake[2]		Efficiency[3]	
	Control	Peptiva	Control	Peptiva	Control	Peptiva
None	261.3±6.6	269±10.1	410.3±16.0	435.1±7.5	0.64±0.03	0.64±0.03
Coccidia	207.9±17.6	243.4±9.7	371±13.0	380.4±11.2	0.56±0.02	0.63±0.04

*Chicks were challenged with *Eimeria acervulina* on day 14

[1]Significant effect due to Coccidia challenge on rate of gain (P=0.005). Effect of dietary treatment on rate of gain was P=0.09. Interaction between Coccidia challenge and dietary treatment was P=0.28.

[2]Significant effect due to Coccidia challenge on feed intake (P=0.003). Dietary treatment did not affect feed intake (P=0.18).

[3]Significant interaction between dietary treatment and coccidia challenge (P=0.05). This interaction indicates that Peptiva increased the efficiency of feed conversion in the coccidia challenged chicks but not in the health control chicks.

TABLE 3. Effect of Peptiva on performance of broiler chicks challenged with *E. coli* (14-20 days)

Challenge*	Gain[1]		Intake[2]		Efficiency[3]	
	Control	Peptiva	Control	Peptiva	Control	Peptiva
None	261.3±6.6	269±10.1	410.3±13.0	435.1±7.5	0.64±0.03	0.64±0.03
E. coli	237.4±11.1	245.8±9.0	402.1±8.6	417.0±7.4	0.59±0.02	0.59±0.04

*Chicks were challenged with *E. coli* on day 14

[1]Significant effect due to *E. coli* challenge on gain (P=0.01). Effect due to dietary treatment was P=0.11.

[2]Significant effect due to *E. coli* challenge on feed intake (P=0.05). Significant effect due to dietary treatment on feed intake (P=0.04).

[3]Significant effect due to E. coli on efficiency of feed utilization (P=0.04). Dietary treatment did not affect feed efficiency (P>0.25).

TABLE 4. Effect of Peptiva on antibody response and delayed-type hypersensitivity to coccidia (day 20)

Challenge*	Anti-cocci IgG[1]		Anti-cocci IgM[2]		DTH to cocci [3]	
	Control	Peptiva	Control	Peptiva	Control	Peptiva
None	ND	ND	2.2±0.5	1.9±1.0	2.1±0.4	2.3±0.6
Coccidia	4.5±0.6	4.7±0.9	6.4±1.1	6.2±1.2	3.7±0.8	3.9±0.9

*Chicks were challenged with *Eimeria acervulina* on day 14 and blood was taken on day 20.
[1]ND means non-detectable. There were no significant effects due to diet (P>0.05)
[2]Significant effect due to Coccidia challenge on anti-cocci IgM (P=0.001). Dietary treatment did not affect anti-cocci IgM (P>0.25).
[3] Significant effect due to Coccidia challenge on DTH to cocci (P=0.02). Dietary treatment did not affect DTH (P>0.25).

TABLE 5. Effect of Peptiva on morphometrics of the intestinal duodenum of chicks.

Treatment	lamina propria (µm)	villus height (µm)	villus width (µm)	crypt depth (µm)	intra-epithelial lymphocytes (#/villi)	lamina propria leukocytes (#/villi)
Control diet	69	778	133	129	9	27
Control + Peptiva	72	785	141	128	11	33
Control + Cocci	89	611	155	152	18	120
Control Peptiva + Cocci	92	657	151	140	21	115
Pooled SEM	4.3	18	15	16	3.9	23
ANOVA P value; Diet	0.17	0.09	0.37	0.62	0.24	0.50
ANOVA P value; Cocci	<0.01	<0.01	0.07	0.04	<0.01	<0.01
Diet x Cocci	0.82	0.05	0.11	0.15	0.72	0.27

TABLE 6. Effect of Peptiva on antibody response and delayed-type hypersensitivity to *E. coli* (day 20)

Challenge*	Anti-coli IgG[1]		Anti coli IgM[2]		DTH to E. coli[3]	
	Control	Peptiva	Control	Peptiva	Control	Peptiva
None	2.7±0.7	2.5±0.5	3.7±1.2	3.7±1.0	3.2±0.4	3.5±0.5
E. coli	6.6±1.1	6.3±0.8	8.7±0.9	8.1±1.1	5.5±1.3	5.8±1.9

*Chicks were challenged with *E. coli* on day 14 and blood was taken on day 20.
[1] Significant effect due to E. coli challenge on anti-E. coli IgG (P<0.001). Dietary treatment did not affect anti-E. coli IgG (P>0.25).
[2]Significant effect due to E. coli challenge on anti-E.coli IgM (P<0.001). Dietary treatment did not affect anti- E. coli IgM (P>0.25).

[3] Significant effect due to E. coli challenge on DTH to E. coli (P=0.03). Dietary treatment did not affect DTH (P>0.25).

CONCLUSIONS

- Peptiva did not affect the performance of healthy chicks.
- Peptiva improved the efficiency of feed utilization in chicks challenged with *Eimeria acervulina*.
- Peptiva improved feed intake in chicks challenged with *E. coli.*
- Peptiva did not affect the adaptive immune response to *E. coli* or *Eimeria acervulina*.
- Peptiva prevented part of the loss in villi length caused by Eimeria. This effect was apparently not related to an effect on the immune response to the Eimeria because the numbers of leukocytes in the epithelium and lamina propria were not affected.
- Improved villi length may indicate better nutrient absorption, which would explain the improved efficiency of feed utilization.