



Contribution of Carbohydrate Binding Modules (CBMs) to decrease the dosage level of recombinant cellulases used to supplement a barley based diet for poultry

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Overview

¢ Introduction

- | What are NSP's?
- | Why enzymatic supplementation?
- | Cellulases and Hemicellulases architecture
- | Role of Carbohydrate Binding Modules (CBM's)

¢ Aim

¢ Experimental procedure

¢ Results

¢ Conclusions



Introduction

Non-Starch Polysaccharides

¢ Important cell wall components

┃ Insoluble in water

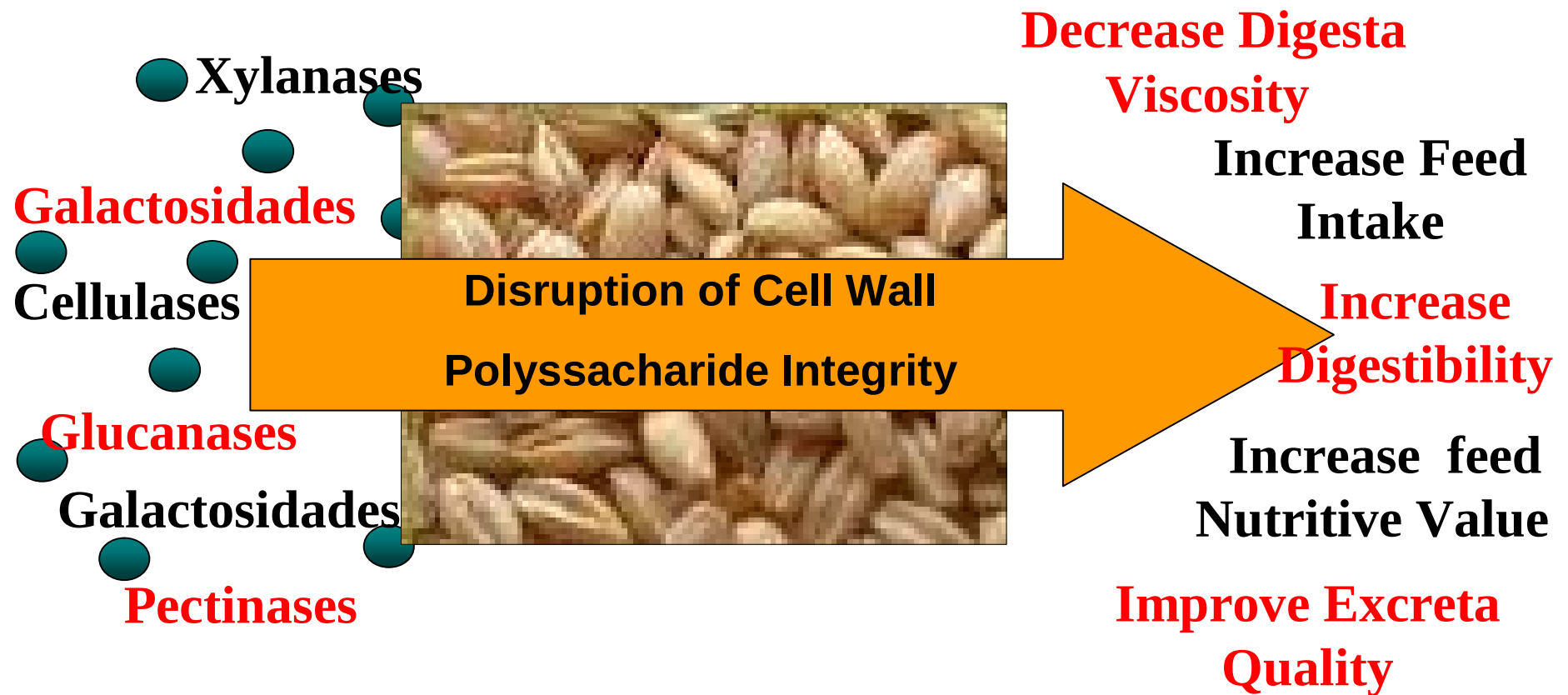
- Absorb water – promote motility
- Physic barrier – decrease digestion efficiency

┃ Soluble in water

- Anti-nutritive effects in monogastric:
 - Increase viscosity → Performance
 - Modification of gut physiology
 - Interaction with microflora



Enzyme Supplementation





Cellulases and Hemicellulases

☼ Modular Structures:

| Catalytic domains

- Glycoside Hydrolases (GH's)
 - GH26 (β -1,3-1,4-glucanase)
 - GH5 (β -1,4-cellulase)

| Non-catalytic domains;

- Carbohydrate Binding Modules (CBM's)
 - CBM11 (Binds both β -1,4-glucan and β -1,3-1,4-mixed linked glucans)



Lic26A

Cel5E

CBM 11



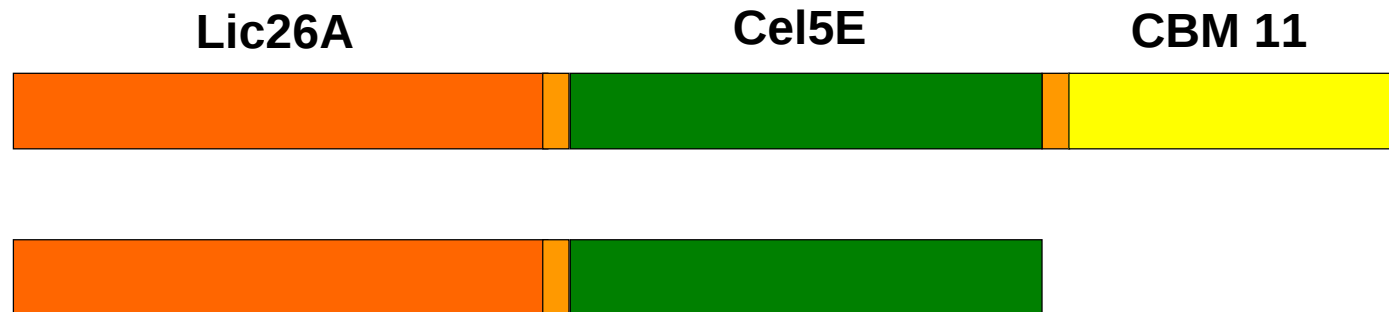
Role of CBMs

- ¢ Promote a close and prolonged interactions enzyme – substrate
- ¢ Proximity effect potentiates catalysis
- ¢ Targeting effect important in the complex structure of cell walls



AIM of this project

- ¢ To evaluate the role of CBMs in the function of cellulases used to supplement diets for broiler chicks

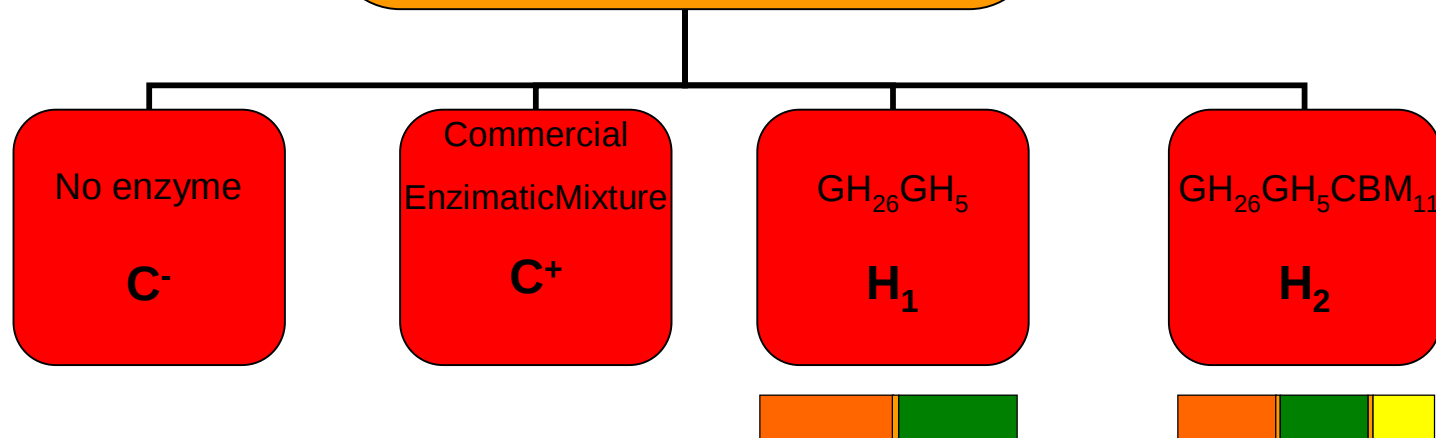




Experimental Design



**Barley based diet (55%)
160 broiler chicks
Ad Libitum (feed and water)**





Results - High dosage

Performance

Treatment	Body Weight	Weight Gain	Feed Ratio
C-	1158.0 ^b	1115.7 ^b	0.618
C+ (10U/kg)	1243.4 ^a	1200.8 ^a	0.602
H ₁ (30U/kg)	1240.4 ^a	1198.2 ^a	0.619
H ₂ (30U/kg)	1247.9 ^a	1205.9 ^a	0.623



Results - High dosage

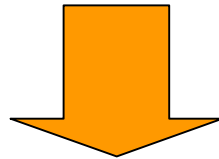
¢ Digesta Viscosity

Treatment	Duodenum + Jejunum
C ⁻	11.3 ^a
C ⁺ (10U/kg)	5.53 ^b
H ₁ (30U/kg)	7.10 ^b
H ₂ (30U/kg)	7.24 ^b



CBM effect redundant

- ¢ Recombinant enzyme as effective as commercial preparation
- ¢ CBM₁₁ was unable to promote the efficiency of the recombinant enzyme



- ¢ Repeat experiment with lower enzyme levels



Results – Low Dosage

Performance

Treatment	Body Weight	Weight Gain	Feed Ratio
C-	1146.5 ^a	1100.9 ^a	0.569
C+ (10U/kg)	1205.6 ^b	1160.3 ^b	0.596
H ₁ (10U/kg)	1191.7 ^{ab}	1145.8 ^{ab}	0.574
H ₂ (10U/kg)	1237.6 ^b	1192.7 ^b	0.601



Results – Low Dosage

¢ Digesta Viscosity

Treatment	Duodenum + Jejunum
C ⁻	7.450 ^a
C ⁺ (10U/kg)	4.955 ^b
H ₁ (10U/kg)	6.962 ^a
H ₂ (10U/kg)	6.149 ^{ab}

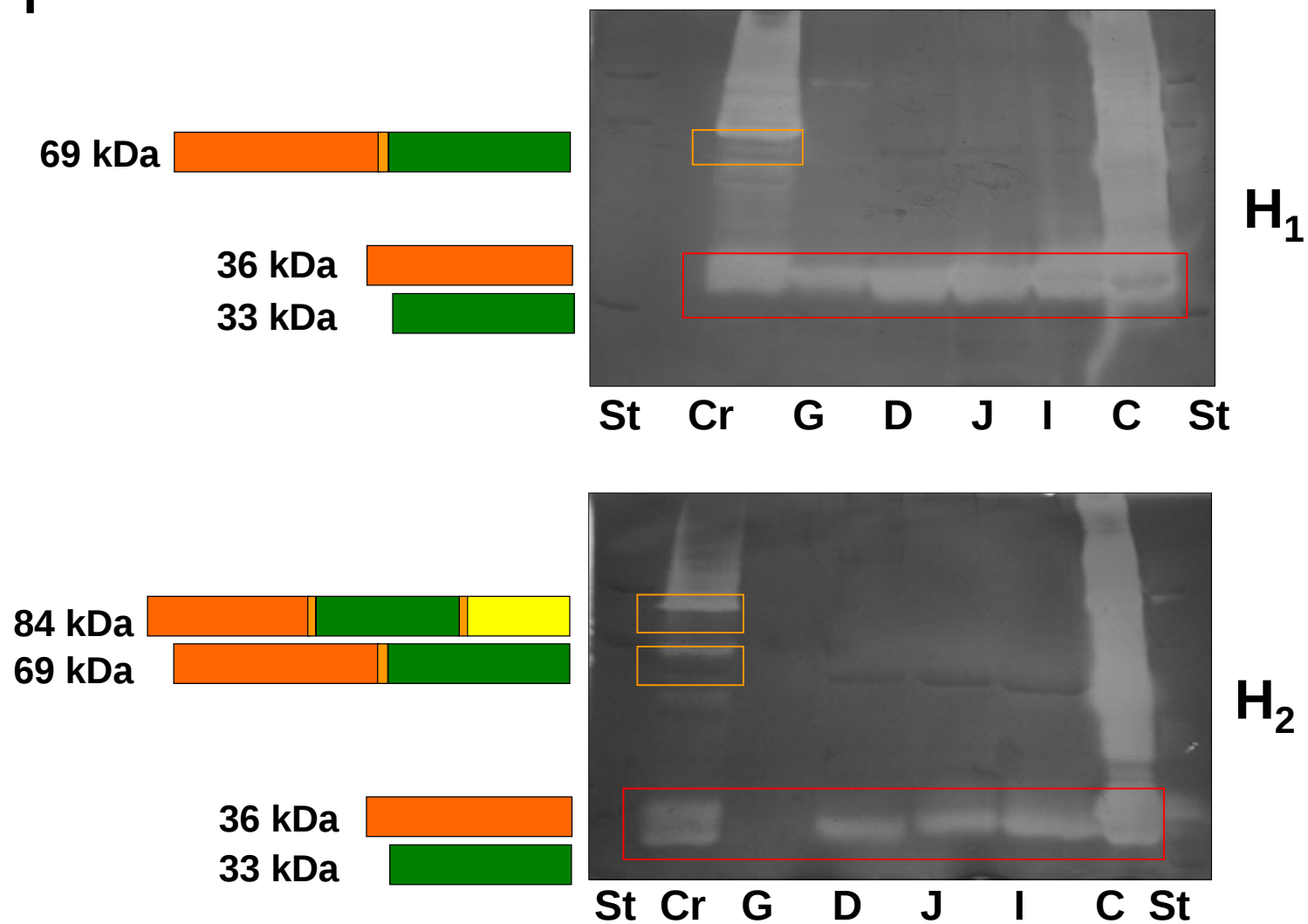


Integrity of enzymes

- ¢ Samples of the various GI compartments were recovered
- ¢ Cellulase activity was measured through plate assays and zymograme analysis



Results





Conclusions

- ¢ Individual recombinant cellulases are as effective as commercial to improve the nutritive value of barley based diets for broilers;
- ¢ CBMs improve the efficacy of feed cellulases used at lower dosage rates;
- ¢ Together the data suggest that cellulases act primarily in initial portions of the GI tract.



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