

## Research Article

# $\beta$ -Mannanase Enzyme Supplementation in Reformulated Grow-Finishing Diets Resulted in Retained Performance and an Economic Benefit

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## Abstract

Many vegetable feed ingredients – such as wheat, corn, barley, palm kernel meal, sun flower meal and others – contain  $\beta$ -mannans, known as strongly antinutritive polysaccharide fibres. The content of soluble  $\beta$ -mannans in swine diets commonly ranges between 0.15% to 0.40%. As little as 0.05% soluble  $\beta$ -mannans in feed can elicit a strong innate immune response. Hemicell HT (Elanco) is a  $\beta$ -mannanase enzyme used to supplement animal feed, breaking down  $\beta$ -mannans and thus preventing economic losses due to the wasteful immune response elicited by these  $\beta$ -mannans. The current field study compared fattening pig performance on a control diet to a reformulated diet, including a  $\beta$ -mannanase enzyme combined with a 45 kcal/kg reduction in net energy (NE). A 130-day feeding trial was conducted on a commercial grow-finishing facility with DanBred x Piétrain pigs starting at 70 days of age. Standard four-phase (0-30 d, 31-60 d, 61-90 d, and 91-130 d) control diets were compared to reformulated diets with a 45 kcal/kg NE reduction along with the inclusion of a  $\beta$ -mannanase enzyme (Hemicell HT; Elanco) at 300 g/tonne. Standard production data were collected and analysed using JMP 17.0 statistical software. Overall, pig performance and carcass quality data did not differ significantly between treatment groups during the entire grow-finishing period. Hemicell HT had an overall benefit of € 5.24 per grow-finishing pig and € 2.38 per tonne of feed due to the 45 kcal/kg NE reduction. The current trial demonstrated that the inclusion of Hemicell HT in reformulated lower-energy diets (- 45 kcal/kg NE) was able to retain production performance in grow-finishing pigs with an additional economic benefit.

**Keywords:**  $\beta$ -mannanase, Grow-finishing pigs, Net energy reduction, Equal performance, Economic benefit

## Introduction

All vegetable feed ingredients commonly used in swine diets contain polysaccharides, which are polymers of monosaccharides linked by glycosidic bonds. Starch, a polymer of glucose units linked by  $\alpha$ -(1-4) with a few  $\alpha$ -(1-6) bonds, is digested in the small intestine of pigs through endogenous enzyme activity. Non-starch polysaccharides (NSPs) are fibrous materials found in the plant cell wall, including celluloses, hemicelluloses, pectins, and oligosaccharides. Monogastric animals like pigs lack the endogenous enzymes required to digest  $\beta$ -linked NSPs like  $\beta$ -mannans [1].  $\beta$ -mannans – an antinutritive factor present in many common feed ingredients [2]– have gained increasing attention in recent years.  $\beta$ -Mannans are linear polysaccharides composed of repeating units of  $\beta$ -1,4-mannose and  $\alpha$ -1,6-galactose and/or glucose attached to the  $\beta$ -mannan backbone [3,4]. In monogastric diets, high concentrations of these  $\beta$ -mannans are considered unsuitable due to their antinutritive properties, mainly due to stimulation of an innate immune response at the level of the intestinal lining. The innate immune cells recognize pathogens through distinct molecules, called pathogen-associated molecular patterns (PAMPs), which are expressed on the pathogen surface [5]. The binding of PAMPs to pathogen recognition receptors (PRR) present on innate immune cells, results in the release of

innate defense molecules such as reactive oxygen and nitrogen species, bacteriolytic enzymes, antimicrobial peptides and complement proteins [6]. These PAMPs include complex polysaccharides that resemble  $\beta$ -mannans [5]. Consequently,  $\beta$ -mannans present in the swine feed may be mistaken by the immune system in the gastrointestinal tract for invading pathogens causing an unwarranted immune activation [7,8], also known as a feed-induced immune response [9]. This misrecognition of  $\beta$ -mannans as invading pathogens results in a futile immune response that wastes energy and nutrients [3]. The hydrolysis of  $\beta$ -mannans through the inclusion of an exogenous  $\beta$ -mannanase enzyme can reduce and potentially eliminate their ability to induce FIIR.

Supplementation of  $\beta$ -mannanase to low- and high-mannan diets has the potential to improve the performance of growing pigs [10]. Other studies have concluded that  $\beta$ -mannanase improved growth performance in both weaning and grow-finishing pigs on corn-SBM diets [11-13] with minimal effects on nutrient digestibility [12]. Moreover, innate immune activation is accompanied by downregulation of anabolic functions [14], resulting in a reduced performance capacity. Therefore, supplementation of a  $\beta$ -mannanase enzyme to grow-finishing diets could reduce or eliminate the occurrence of FIIR and increase available energy and proteins for growth.

The objective of the current field study was to evaluate the effects of  $\beta$ -mannanase supplementation of grow-finishing diets with a reduced net energy content of 45 kcal/kg NE of feed containing a high level of  $\beta$ -mannans on grow-finishing pig performance, carcass quality, and economic parameters during the grow-finishing phase.

## Materials and Methods

### Description of Experimental Farm

The field trial was conducted on a conventional 400-pig grow-finishing unit in Belgium. The field study consisted of 7 Control batches and 2 Enzyme-treated batches that were enrolled between April 2023 and November 2024. A total of 3111 grow-finishing pigs were included in the study, of which 2335 grow-finishing pigs were assigned to the Control group and 776 grower pigs to the Enzyme-treated group. Each pen housed 11-12 grower pigs. Compartments were ventilated through mechanical ventilation with an air inlet through side walls. All pens were equipped with partially slatted concrete floors, dry feeders, and water was distributed through nipples in the feeders. Meal feed consumption was registered at group level. Both study groups were randomly allocated in time, eliminating potential seasonal effects throughout the study. No other changes were implemented during the period of the field study that might have impacted on the grow-finishing performance.

## Experimental Design

### Treatment Groups

At the start of the grow-finishing period, the entire batch of pigs was assigned to one of the treatment groups: Control or Enzyme-treated, respectively. A four-phase diet was distributed. The specific treatment was blinded to the farm personnel and only registered by the nutritionist at the feed mill. Grow-finishing pigs in the entire barn were considered one experimental unit and were weighed together at the start and end of the study period.

### Experimental Diets

The pigs were fed a four-phase diet consisting of Phase 1 (0-30 days), Phase 2 (31-60 days), Phase 3 (61-90 days), and Phase 4 (91-130 days) in each of the treatment groups. The main difference between the diets in the Control and Enzyme-treated groups was the reduction in net energy content of 45 kcal/kg NE of feed in all phases (Table 1). The Enzyme-treated group was supplemented with a  $\beta$ -mannanase enzyme (Hemicell HT; Elanco, Indianapolis; IN) at an inclusion rate of 300 g per tonne of feed, according to the manufacturer's instructions for use. All other enzymes (xylanase and phytase) in the diets remained at the same levels in both study groups.

**Table 1:** Feed formulation (expressed as % of total feed) of the different phases (Phase 1-2-3-4) for Control and Enzyme-treated diets with a 63 kcal/kg NE reduction in Enzyme-treated diets. All major feed ingredients in both treatment groups are mentioned. Premix composition is identical in both treatment groups.  $\beta$ -mannan content (expressed as %), proportion of diet that was taken into account for calculation of  $\beta$ -mannan content (expressed as %), and net energy content (calculation 2015; expressed as kcal/kg feed) is given for each of the feed formulations.

Component	Phase 1 (day 1-30)		Phase 2 (31-60)		Phase 3 (day 61-90)		Phase 4 (day 91-130)	
	Control	Enzyme	Control	Enzyme	Control	Enzyme	Control	Enzyme
Wheat	34.95	25.34	36.72	34.36	38.33	27.27	39.86	37.32
Barley	17.50	23.00	17.50	17.50	17.50	17.50	18.00	17.50
Corn	7.50	10.00	10.00	10.00	5.00	12.50	5.00	7.50
Wheat bran	6.00	6.00	6.00	6.00	6.00	8.00	6.00	8.00
Corn gluten	4.30	5.00	1.50	6.00	3.00	6.00	4.00	6.00
Biscuit by-product	4.00	4.00	4.00	4.00	3.00	4.00	4.00	4.00
Palm kernel meal	3.00	4.00	4.00	2.60	4.00	2.80	3.90	5.00
Corn by-product	3.00	3.00	.	.	2.50	3.00	1.50	3.00
Swine fat	1.00	1.29	1.49	1.51	1.53	1.13	1.49	1.36
CaCO <sub>3</sub>	1.09	1.21	0.92	0.94	0.80	0.84	0.84	0.89
Piglet feed core	0.50	0.50	.	.	.	.	.	.
L-lysine HCl	0.45	0.45	.	0.47	0.46	0.50	0.46	0.45
NaCl	0.37	0.38	0.37	0.33	0.34	0.33	0.24	0.36
CaPO <sub>4</sub>	0.20	0.20	0.10	0.10	.	.	.	.
L-threonine	0.19	0.18	0.17	0.17	0.17	0.18	0.15	0.16
Liquid betaine	0.17	0.17	0.17	0.17	0.27	0.17	0.27	0.27
NaHCO <sub>3</sub>	0.10	0.15	0.10	0.15	0.15	0.15	0.28	0.17
Hydroxymethionine 88	0.14	0.15	0.10	0.10	0.09	0.09	0.07	0.07
L-tryptofaan 25%	0.09	0.09	0.07	0.06	0.02	0.06	0.01	0.01
Enzyme mix 10	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05
Soy products	15.40	14.80	9.50	9.00	6.40	4.90	2.00	3.40
Rapeseed meal	.	.	3.00	3.00	6.00	5.00	6.00	3.20
Beet pulp	.	.	.	.	.	.	1.50	1.50
Sun flower pellets	.	.	3.00	3.00	2.69	5.00	3.87	3.26
Hemicell HT	.	0.03	.	0.03	.	0.03	.	0.03
$\beta$ -mannan content (%)	0.511	0.581	0.552	0.471	0.559	0.466	0.568	0.628
Proportion (%)	88.65	88.14	89.82	91.46	88.92	88.97	94.13	93.08
Net energy 2015 (kcal)	2,427	2,453*	2,412	2,405*	2,388	2,420*	2,389	2,412*

\*Supplementation of Hemicell HT was incorporated into the feed net energy calculation for a total value of 63 kcal per kg feed.

## Experimental Animals

DanBred \* Belgian Piétrain grow-finishing pigs were obtained from a conventional commercial sow farm linked to the grow-finishing facility. The pigs were vaccinated to protect against Mycoplasma hyopneumoniae, Porcine Reproductive and Respiratory Syndrome Virus (PRRSV), and Porcine Circovirus type 2 (PCV-2). A study batch consisted of approximately 389 ( $\pm 1$ ) grow-finishing pigs in each consecutive trial period.

## Performance Data Collection

Pig body weight was measured at group level at 0 and 130 days. Feed provision (ad libitum) was recorded at treatment group level. Average daily weight gain (ADWG; expressed as g/d), average daily feed intake (ADFI; expressed as g/d) and feed conversion rate (FCR; expressed as kg feed per kg of weight gain) were calculated for the entire study period. Mortality was recorded along with the date of death and the number of dead animals.

## Slaughter Parameters

The economically important slaughter parameters were collected from the slaughterhouse carcass database and consisted of the number of underweight (small) pigs, carcass rentability (%), basic price per kg, meat percentage (%), backfat depth (mm), loin depth (mm), AFI (quality index), proportion of pig with a rentability above 82%, quality supplement, and weight supplement.

## Veterinary Treatments

Individual antibiotic treatments were administered as needed due to the critical state of the piglets and in case of a broader health issue in the barn, group treatment could be administered. The same veterinary products and dosages (ml/kg) were used throughout the entire study period. Individual antibiotic treatments or group treatments were recorded daily including the date, product, dose, ID number of treated piglets, presumed cause of treatment, and the number of times the treatment was repeated.

## Economic Benefit per Piglet and per Tonne of Feed

The economic benefit of  $\beta$ -mannanase supplementation combined with a reduction in net energy of approximately 45 kcal/kg NE feed was calculated both at the grow-finishing pig level and at the feed cost level. For the calculation of economic benefit at the grow-finishing pig level, the following parameters were considered: feed cost reduction, pig price correction (standard price for 130 kg slaughter pig), and opportunity costs of mortality. For the calculation of economic benefit at the feed cost level, the following parameters were considered: total feed cost and the total amount of feed consumed.

## Data Management and Statistical Analysis

Data was hand-recorded by the farm personnel and stored in MS Excel on OneDrive at the end of each day. Following the end of the feed trial, the data were extracted from Excel into JMP 17.0 and the blinded treatments were unblinded to reveal the respective treatment groups. Calculations, exploratory data analysis and quality review, and

subsequent statistical analysis were all performed in JMP 17.0. All data were presented as a means with their respective pooled standard error of the mean (SEM). All means were tested for significant differences ( $P < 0.05$ ) using a T-test.

## Results

### Piglet Weight

Data on weight and days in fattening are given in Table 2. The pigs arrived at the grow-finishing facility at an average weight of 22.41 kg. There were no significant differences ( $P > 0.05$ ) observed in the start weight (d0) between both treatment groups. At slaughter (approx. day 130) pigs in the Enzyme-treated group were significantly ( $P < 0.05$ ) heavier as compared to the Control group (130.5 kg  $\pm$  0.9 kg vs. 124.5  $\pm$  2.3 kg, respectively). However, grow-finishing pigs in the Control group were slaughtered on average 6 days earlier as compared to the pigs in the Enzyme-treated group (126 d  $\pm$  2.3 vs. 132 d  $\pm$  2.1, respectively).

### Average Daily Weight Gain, Average Daily Feed Intake and Feed Conversion Rate

Data on ADWG, ADFI, and FCR are given in Table 2. Average daily weight gain was slightly, but not significantly ( $P > 0.05$ ) higher in the Enzyme-treated group as compared to the Control group (822  $\pm$  3 g/d vs. 813  $\pm$  14 g/d, respectively). Average daily feed intake was slightly,

**Table 2:** Performance parameters and slaughter data for both Control and Enzyme-treated groups with a 63 kcal/kg NE reduction in Enzyme-treated diets. Continuous data are given as mean  $\pm$  SEM. *P*-values  $< 0.05$  represent statistically significant differences.

Parameter	Control	Hemicell HT	P-value
<i>Descriptive parameters</i>			
Number of groups	6	2	-
Average number of pigs per group - start	389	388	-
Average number of pigs per group	379	374	-
<i>Production parameters</i>			
Mortality (#)	10 $\pm$ 2.4	14 $\pm$ 8.0	0.347
Mortality (%)	2.5 $\pm$ 0.6	3.6 $\pm$ 2.1	0.346
Days in fattening	126 $\pm$ 2.3	132 $\pm$ 2.1	0.197
Average weight at start (kg)	22.33 $\pm$ 0.34	22.50 $\pm$ 0.00	0.316
Average weight at slaughter (kg)	124.5 $\pm$ 2.3	130.5 $\pm$ 0.9	<b>0.018</b>
Average daily weight gain (g/d)	813 $\pm$ 14	822 $\pm$ 3	0.420
ADFI (g/d)	2,135 $\pm$ 40	2,177 $\pm$ 169	0.423
FCR (kg feed/kg growth)	2.626 $\pm$ 0.028	2.643 $\pm$ 0.093	0.441
Cost per kg growth (€)	0.877 $\pm$ 0.053	0.828 $\pm$ 0.034	0.235
<i>Slaughter parameters</i>			
Number of small pigs	0.33 $\pm$ 0.33	0.50 $\pm$ 0.50	0.404
Rentability (%)	80.7 $\pm$ 0.5	80.8 $\pm$ 0.6	0.456
Basic price (€/kg)	1.79 $\pm$ 0.05	1.83 $\pm$ 0.04	0.320
Meat (%)	64.2 $\pm$ 0.3	64.6 $\pm$ 0.8	0.362
Backfat depth (mm)	12.7 $\pm$ 0.2	12.1 $\pm$ 0.1	0.121
Loin depth (mm)	73.6 $\pm$ 0.6	73.7 $\pm$ 1.3	0.461
AFI	3.2 $\pm$ 0.1	3.1 $\pm$ 0.1	0.293
Supplement >82%	6.4 $\pm$ 1.0	5.8 $\pm$ 1.1	0.336
Quality supplement	0.0270 $\pm$ 0.0034	0.0298 $\pm$ 0.0034	0.301
Weight supplement	-0.0066 $\pm$ 0.0031	-0.0162 $\pm$ 0.0029	<b>0.045</b>

but not significantly ( $P > 0.05$ ) higher in the Enzyme-treated group as compared to the Control group ( $2,177 \pm 196$  g/d vs.  $2,135 \pm 40$  g/d, respectively). Feed conversion rate was slightly, but not significantly ( $P > 0.05$ ) higher in the Enzyme-treated group as compared to the Control group ( $2.643 \pm 0.043$  kg feed/kg gain vs.  $2.626 \pm 0.028$  kg feed/kg gain, respectively).

### Antimicrobial Treatment

No significant differences were observed at either the level of individual treatment or at the level of group treatment between both treatment groups during both feed trials.

### Mortality

Data on mortality are given in Table 2. Overall, mortality was slightly, but not significantly ( $P > 0.05$ ) higher in the Enzyme-treated group as compared to the Control group (3.6 % vs. 2.5 %, respectively).

### Slaughter Parameters

Overall, slaughter parameters were not significantly ( $P > 0.05$ ) different between both treatment groups. Only the weight supplement was significantly ( $P < 0.05$ ) lower in the Enzyme-treated group as compared to the Control group ( $-0.0162 \pm 0.0029$  vs.  $-0.0066 \pm 0.0031$ , respectively). This might be the result of the average higher slaughter weight in the Enzyme-treated group.

### Economic Benefit per Piglet and per Tonne of Feed

The detailed calculation of economic benefit per grow-finishing pig is given in Table 3. Overall, supplementation of a  $\beta$ -mannanase enzyme combined with a reduction of net energy by 45 kcal/kg feed over the four phases resulted in an economic benefit per piglet of € 5.24. The detailed calculation of economic benefit per tonne of feed is given in Table 4. Overall, supplementation of a  $\beta$ -mannanase enzyme resulted in a feed cost reduction of € 2.38 per tonne of feed.

**Table 3:** Detailed calculation of economic benefit per piglet considering a reduction in feed cost, piglet price corrections (130 kg; % slaughter rentability) and the opportunity cost of mortality for a reduction of 63 kcal/kg NE in Enzyme-treated diets.

Parameter	Control	Hemicell HT
Feed cost per fattening pig (0-130 d)	€ 92.18	€ 88.24
Benefit feed cost reduction		+€ 3.94
Pig price corrections (130 kg, % rentability)	€ 180.44	€ 182.38
Benefit technical results		+€ 1.94
Mortality (#)	5	6
Total opportunity cost due to mortality (€)	€ 1,170	€ 1,410
Opportunity cost per marketed piglet (€/piglet)	€ 3.05	€ 3.69
Benefits mortality		- € 0.64
<b>Overall benefit per piglet</b>		<b>+€ 5.24</b>

**Table 4:** Detailed calculation of economic benefit of feed cost per tonne of feed considering total feed costs and total amount of feed consumed for a reduction of 63 kcal/kg NE in Enzyme-treated diets.

Parameter	Control	Hemicell HT
Total feed costs (0-130 d)	€ 30,843	€ 27,667
Total amount of feed consumed (tonne)	110.244	99.742
Feed cost per unit (€/tonne)	€ 279.77	€ 277.39
<b>Overall benefit per tonne of feed</b>		<b>- € 2.38</b>

## Discussion

In current field trial, involving the 45 kcal/kg NE reduction, the  $\beta$ -mannan content ranged from 0.466% to 0.628% in all four phases. This  $\beta$ -mannan content was sufficiently high to maintain the standard feed composition without the need for additional protein substitutions as previously reported [15]. Since high levels of  $\beta$ -mannans are known to be an antinutritive factor [2], this may stimulate an innate immune response mainly due to their resemblance with PAMPs [5]. This innate immune response, known as FIIR [9] can lead to unnecessary immune activation, causing energy and nutrients to be wasted [3]. Therefore, 300 g/tonne of an exogenous  $\beta$ -mannanase enzyme (Hemicell HT; Elanco, Greenfield, IA) was added to the diet to hydrolyze these antinutritive  $\beta$ -mannans. The overall results demonstrated no significant differences between treatment groups in the measured (pig weight, feed provision) or calculated (ADWG, ADFI, FCR) performance parameters. Despite some minor numerical differences, the overall result confirmed that the addition of an exogenous  $\beta$ -mannanase to adapted formulations, with a reduction in net energy content of 45 kcal/kg of feed and the presence of a sufficient level of  $\beta$ -mannans, allowed grow-finishing pigs to perform equally to those fed the standard Control diets. These results are consistent with other recent studies in low- and high-mannan diets [10,15-18]. Slaughter data, including carcass quality parameters and economically important traits, did not differ significantly between treatment groups. These observations are in accordance with another study with inclusion of  $\beta$ -mannanase [16].

In addition to similar results in production performance, a substantial economic benefit of supplementation of a  $\beta$ -mannanase enzyme could be calculated. Based on the actual feed prices and measured feed intake, we obtained a 4.27% reduction in the feed cost (€ 88.24 vs. € 92.18 in the Enzyme-treated vs. the Control group, respectively) per grow-finishing pig produced and a 0.85% reduction in feed cost per tonne of feed (€ 277.39 vs. € 279.77, in Enzyme-treated vs. Control group, respectively). Considering all costs (feed cost, pig price correction at 130 kg, and opportunity costs for mortality) the income per produced piglet was € 5.24 higher for the Enzyme-treated group. Others concluded that  $\beta$ -mannanase improved growth performance in both weanling and grow-finishing pigs on corn-SBM diets [11-13]. A diet with a 150 kcal/kg reduction in digestible energy supplemented with  $\beta$ -mannanase outperformed in weight gain and feed efficiency [11].

Overall, the results from the current grow-finishing trial demonstrated that in the presence of a sufficient amount of  $\beta$ -mannans in the diet formulations, the addition of a  $\beta$ -mannanase enzyme (Hemicell HT; Elanco) could support grow-finishing pig performances under field conditions with formulations adapted towards net energy reduction. The adapted diet formulation resulted in improved economic benefits at the individual grow-finishing pig level and the cost per tonne of feed level.

## Conclusions

The current trial demonstrated that the inclusion of Hemicell HT in reformulated diets with a lower energy content (45 kcal/kg NE) was

able to retain production performance in grow-finishing pigs with an economic benefit. The inclusion of Hemicell HT provided an overall benefit of € 5.24 per piglet and € 2.38 per tonne of feed attributable to the 45 kcal/kg NE reduction. There was no significant difference in the overall quality of the slaughtered pigs.

## Declarations

## Ethics Approval and Consent to Participate

Field trial with an EFSA approved feed supplement for use in swine. No additional ethical approval was needed. Consent to participate was obtained following full information of the farmer on the protocol to be carried out.

## Availability of Data and Material

The datasets analyzed during the current study are available from the corresponding author on reasonable request.

## Competing Interests

The authors declare that they have no other competing interests.

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## Author's Contributions

FV and AdB were both involved in study design, data collection, data analysis and manuscript preparation. Both authors read and approved the final manuscript.

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## Author's Information

FV is currently a Principal Technical Advisor Swine & Nutritional Health for Benelux / UK&ROI within Elanco Animal Health. He holds a DVM, a Master in Veterinary Public Health and Food Safety, a PhD in Veterinary Sciences, a PhD in Applied Biological Sciences, an EBVSTM European Specialist in Porcine Health Management and a Diplomate of the American Board of Veterinary Practitioners – Swine Health Management. He has an interest in swine intestinal health and specific approaches to improve intestinal health through non-antibiotic solutions.

## Abbreviations

ADFI: Average Daily Feed Intake; ADWG: Average Daily Weight Gain; FCR: Feed Conversion Rate; NSP: Non-Starch Polysaccharide; PCV-2: Porcine Circo Virus - type 2; PRRSV: Porcine Reproductive and Respiratory Syndrome Virus; SBM: Soybean Meal

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