

**UNIVERSIDADE ESTADUAL PAULISTA -UNESP
CAMPUS DE JABOTICABAL**

**EFEITO DA INCLUSÃO DE CARBOIDRASES SOBRE A
EFICIÊNCIA ENERGÉTICA EM FRANGOS DE CORTE E
POEDEIRAS COMERCIAIS**

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RESSALVA

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POEDEIRAS COMERCIAIS**

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Coorientadora: Profa. Dra. Michele Bernardino de Lima**

Tese apresentada à Faculdade de Ciências Agrárias e Veterinárias – Unesp, Campus de Jaboticabal, como parte das exigências para a obtenção do título de Doutor em Ciência Animal.

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DADOS CURRICULARES DO AUTOR

RITA BRITO VIEIRA- Filha de Pedro Almeida Vieira e Rosa da Silva Brito, nasceu no dia 22 de março de 1993, na cidade de Parintins, Amazonas. Em outubro de 2013 ingressou no curso de Zootecnia da Universidade Federal do Amazonas – Campus Parintins, no Instituto de Ciências Sociais, Educação e Zootecnia, cidade de Parintins, Amazonas, graduando-se em julho de 2018. Em março de 2019 ingressou no curso de Mestrado em Zootecnia na Universidade Federal Rural de Pernambuco - Campus Dois Irmãos – Recife – Pernambuco, Sob orientação da prof. Dra. Maria do Carmo Mohaupt Marques Ludke, defendendo sua dissertação junho de 2021. Em agosto do mesmo ano iniciou o curso de Doutorado em Ciência Animal na Faculdade de Ciências Agrárias e Veterinárias da Universidade Estadual Paulista – Campus de Jaboticabal, São Paulo, sob orientação do Prof. Dr. Edney Pereira da Silva. Em outubro de 2025 defendeu sua tese de doutorado para obtenção do título.

DEDICATÓRIA

Dedico, aos meus pais, meus irmãos e meu companheiro Hélio, por todo apoio, incentivo, amor, persistência, carinho e vitória.

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CEUA – COMISSÃO DE ÉTICA NO USO DE ANIMAIS

CERTIFICADO

Certificamos que o projeto intitulado "**Evaluation of organic acids and carbohydrases in diets for pullets and layers**", protocolo nº 003761/23, sob a responsabilidade do Prof. Dr. EDNEY PEREIRA DA SILVA, que envolve a produção, manutenção e/ou utilização de animais pertencentes ao Filo Chordata, subfilo Vertebrata (exceto o homem), para fins de pesquisa científica (ou ensino) - encontra-se de acordo com os preceitos da lei nº 11.794, de 08 de outubro de 2008, no decreto 6.899, de 15 de julho de 2009, e com as normas editadas pelo Conselho Nacional de Controle de Experimentação Animal (CONCEA), e foi aprovado pela COMISSÃO DE ÉTICA NO USO DE ANIMAIS (CEUA), da FACULDADE DE CIÊNCIAS AGRÁRIAS E VETERINÁRIAS, UNESP - CÂMPUS DE JABOTICABAL-SP.

Vigência do Projeto	01/07/2023 a 31/03/2024
Espécie / Linhagem	Gallus gallus domesticus / Poedeiras comerciais / Lohmann LSL-Lite
Nº de animais	480
Peso / Idade	1.110 – 1.600 kg / 15 – 42 semanas
Sexo	Fêmeas
Origem	A definir

Jaboticabal, 16 de maio de 2023.

Prof. Dra. Paola Castro Moraes
Vice-coordenadora em exercício – CEUA FCAV

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EFEITO DA INCLUSÃO DE CARBOIDRASES SOBRE A EFICIÊNCIA ENERGÉTICA EM FRANGOS DE CORTE E POEDEIRAS COMERCIAIS

RESUMO - A utilização de carboidrases na alimentação de aves é uma prática muito empregada na nutrição animal, pois promove melhorias no aproveitamento dos nutrientes provenientes da dieta. Dessa forma, o presente estudo teve por objetivo avaliar o desempenho, a digestibilidade e a eficiência energética em frangos de corte e poedeiras na fase de produção. No primeiro experimento, objetivou-se determinar os valores de energia metabolizável (EM) em dietas com níveis reduzidos de energia, suplementadas com carboidrases, visando quantificar a liberação de energia promovida por essas enzimas e seus efeitos sobre o desempenho zootécnico de frangos de corte alimentados com dietas à base de milho e farelo de soja. Foram utilizados 2.400 pintos de corte da linhagem Ross 308 para avaliação do desempenho. Foram conduzidos dois ensaios de digestibilidade e um de desempenho, utilizando delineamento inteiramente casualizado, com seis tratamentos e dez repetições. As dietas experimentais consistiram em uma dieta basal (D1), três níveis de redução energética (D2, D3, D4) e duas dietas com suplementação enzimática (D5 e D6), contendo β -mananase isolada ou associada a β -xilanase e β -glucanase. A digestibilidade dos nutrientes foi avaliada nas fases de 7 a 17 dias e de 28 a 37 dias de idade, utilizando 360 e 240 frangos de corte, respectivamente. A análise estatística incluiu o teste de Dunnett e regressão linear. A suplementação com carboidrases aumentou os coeficientes de digestibilidade da matéria seca, do nitrogênio, da energia bruta e da energia metabolizável. A liberação energética estimada variou entre 52 e 98 kcal/kg, dependendo da combinação enzimática. As dietas suplementadas resultaram em melhor conversão alimentar, maior eficiência energética e maior retenção de energia na forma de proteína, além de menor deposição de gordura na carcaça, em relação às dietas com redução de energia sem suplementação. No segundo experimento, avaliou-se o desempenho produtivo, a qualidade dos ovos e a composição corporal de poedeiras na fase de produção, utilizando delineamento inteiramente ao acaso, com cinco tratamentos e 12 repetições de seis aves. Os tratamentos consistiram em: T1 – controle positivo (CP), 2.869 kcal/kg; T2 – controle negativo (CN), 2.769 kcal/kg; T3 – CN mais mananase; T4 – CN mais xilanase e glucanase; T5 – CN mais mananase, xilanase e glucanase. A suplementação enzimática promoveu efeitos significativos sobre a massa de ovos, a eficiência alimentar e a conversão alimentar por massa e por dúzia de ovos. Também foram observadas melhorias na qualidade dos ovos, especialmente na altura do albúmen e na Unidade Haugh. A composição corporal das aves foi influenciada, com alterações na deposição de proteína e gordura em relação às dietas sem suplementação. Dessa forma, a utilização de carboidrases mostrou-se eficiente na hidrólise de substratos específicos presentes na dieta, evidenciando seu potencial para melhorar o aproveitamento de nutrientes e o desempenho produtivo de frangos de corte e poedeiras comerciais.

Palavras-chave: Carboidratos na nutrição animal, desempenho, enzimas digestivas, frango de corte, postura.

Effect of Carbohydrase Inclusion on Energy Efficiency in Broiler Chickens and Commercial Laying Hens

Abstract - The use of carbohydrases in poultry feeding is a widely employed practice in animal nutrition, as it promotes improvements in the utilization of nutrients from the diet. Thus, the present study aimed to evaluate performance, digestibility, and energy efficiency in broiler chickens and laying hens during the production phase. In the first experiment, the objective was to determine metabolizable energy (ME) values in diets with reduced energy levels, supplemented with carbohydrases, aiming to quantify the energy release promoted by these enzymes and their effects on the zootechnical performance of broiler chickens fed corn- and soybean meal-based diets. A total of 2,400 male Ross 308 broiler chicks were used for performance evaluation. Two digestibility assays and one performance experiment were conducted, using a completely randomized design with six treatments and ten replicates. The experimental diets consisted of a basal diet (D1), three levels of energy reduction (D2, D3, D4), and two diets with enzyme supplementation (D5 and D6), containing β -mannanase alone or combined with β -xylanase and β -glucanase. Nutrient digestibility was evaluated during the 7- to 17-day and 28- to 37-day phases, using 360 and 240 broiler chickens, respectively. Statistical analysis included Dunnett's test and linear regression. Carbohydrase supplementation increased the digestibility coefficients of dry matter, nitrogen, gross energy, and metabolizable energy. The estimated energy release ranged from 52 to 98 kcal/kg, depending on the enzyme combination. Supplemented diets resulted in better feed conversion, higher energy efficiency, and greater energy retention as protein, in addition to lower carcass fat deposition, compared with energy-reduced diets without supplementation. In the second experiment, productive performance, egg quality, and body composition of laying hens during the production phase were evaluated, using a completely randomized design with five treatments and 12 replicates of six birds. The treatments consisted of: T1 – positive control (PC), 2,869 kcal/kg; T2 – negative control (NC), 2,769 kcal/kg; T3 – NC plus mannanase; T4 – NC plus xylanase and glucanase; T5 – NC plus mannanase, xylanase, and glucanase. Enzyme supplementation promoted significant effects on egg mass, feed efficiency, and feed conversion per egg mass and per dozen eggs. Improvements were also observed in egg quality, particularly in albumen height and Haugh unit. Body composition of the birds was affected, with changes in protein and fat deposition compared with diets without supplementation. Thus, the use of carbohydrases proved efficient in the hydrolysis of specific substrates present in the diet, demonstrating their potential to improve nutrient utilization and the productive performance of broiler chickens and commercial laying hens.

Keywords: Carbohydrates in animal nutrition, performance, digestive enzymes, broiler chickens, laying hens.

LIST OF ABBREVIATIONS

ADFI – Average daily feed intake
ADG – Average daily gain
AH – Albumen height
AIC – Akaike information criterion
ANOVA – Analysis of variance
AP – Albumen percentage
AW – Albumen weight
BMC – Bone mineral content
BMD – Bone mineral density
BW – Average body weight
C – Contrasts
c – Chemical composition
cAsh – Ash composition
CEUA – Ethics Committee on Animal Use
CONCEA – National Council for the Control of Animal Experimentation
CV – Coefficient of variation
d – Chemical component deposition
dAsh – Ash deposition
dWater – Water deposition
D1 – Diet 1
D2 – Diet 2
D3 – Diet 3
D4 – Diet 4
D5 – Diet 5
D6 – Diet 6
DM – Dry matter
DXA – Dual-energy X-ray absorptiometry
EEf – Energy Efficiency
EI – Energy intake
EP – Egg production

EPEF – European Production Efficiency Factor
ERFat – Energy retained as fat
ERProtein – Energy retained as protein
ERTotal – Total energy retained
ESP – Eggshell percentage
ESS – Eggshell strength
EST – Eggshell thickness
ESW – Eggshell weight
EW – Egg weight
EU – Experimental units
FCR – Feed conversion ratio corrected for mortality
FI – Feed intake
FM – Fat mass
FTU – Phytase units per kilogram
GEI – Gross energy intake
HU – Haugh unit
LD – Linear dichroism
L-Met – L-methionine
L-Thr – L-threonine
LM – Lean mass
ME – Metabolizable energy
MEI – Metabolizable energy intake
ME_n – Nitrogen-corrected apparent metabolizable energy
ME_w – Metabolizable energy for growth
M_{en} – Body weight
N – Nitrogen
NB – Nitrogen balance
NC – Negative control
NE – Net energy
NSPs – Non-starch polysaccharides
PC – Positive control
SEM – Standard error of mean

SI – Shape index
SPH – Sphericity
SRC – The standard response curve
SCFAs – Short-chain fatty acids
SGLT – 1-Sodium-dependent cotransporter
tEI – Total gross energy intake
T1 – Treatment one
T2 – Treatment two
T3 – Treatment three
T4 – Treatment four
T5 – Treatment five
TGU – Unit of glucanase activity
TLA – Three letter acronym
TM – Total mass
TXU – Unit of xylanase activity
YP – Yolk percentage
YW – Yolk weight

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CHAPTER 1 – GENERAL CONSIDERATIONS

1.1. Introduction

Broiler chicken production reached 14.97 million tons in 2025, resulting in a per capita consumption of 45.5 kg/inhabitant. In the same period, egg production from commercial laying hens reached 57 billion units, with a consumption of 269 eggs/habitant (ABPA, 2025). These figures evidence the continuous growth of Brazilian poultry farming, driven by the increasing demand for nutritious, quality food at accessible prices for the population.

Given the constant need for gains in productivity and efficiency in poultry farming, bird nutrition plays a fundamental role in maximizing nutrient utilization and reducing production costs. In this context, the cereals commonly used in the diets of these broiler and laying birds are sources of fiber that may present moderate or low digestibility (Adebowale et al., 2019; Valente Junior et al., 2024), since birds do not secrete sufficient endogenous enzymes for the degradation of β -glucans (soluble and insoluble) present in these ingredients. The presence of these structures can increase digesta viscosity through water retention, hindering the digestion and absorption of nutrients in the gastrointestinal tract, in addition to limiting enzyme access to cellular content, reducing starch digestibility (Classen et al., 1985; Viana et al., 2011).

Given this physiological limitation, the use of zootechnical additives, such as exogenous enzymes, specifically carbohydrases, has been incorporated into bird feeding. These enzymes act by hydrolyzing the glycosidic bonds between the monomeric units of non-starch polysaccharides (Choct et al., 1996; Cowieson et al., 2006). The presence of high concentrations of soluble NSPs (such as arabinoxylans, β -glucans, and β -mannans) and insoluble NSPs impairs the diffusion and transport of nutrients (Daskiran et al., 2004), compromising the performance of broiler chickens (Morgan et al., 2021a; Kim et al., 2025) and laying hens (Nguyen et al., 2021a; Nguyen et al., 2021b; Nguyen et al., 2022).

The inclusion of carbohydrases (isolated or in association), such as β -xylanase, β -glucanase, and β -mannanase, acts to mitigate these antinutritional effects by promoting the rupture of the plant cell wall and eliminating the nutrient encapsulation effect (Olukosi et al., 2008; Olukosi et al., 2015; Bedford and

Apajalahti, 2022; Wang et al., 2022; Valente Junior et al., 2024). This action results in reduced digesta viscosity, nutrient release, and the consequent improvement of weight gain, feed intake, and dietary energy efficiency (Zhang et al., 2014; Munyaka et al., 2016; White et al., 2021; Shuaib et al., 2025). Additionally, the hydrolysis of undigested carbohydrates generates substrates for fermentation by the intestinal microbiota in the cecum, stimulating the production of short-chain fatty acids (such as acetate, propionate, and butyrate), which play a relevant role in the intestinal health of birds (van der Wielen Paul et al., 2000; Hooper et al., 2002).

The magnitude of the responses to enzyme supplementation is influenced by diet composition. For example, corn-based ingredients contain a higher proportion of insoluble NSPs, whereas wheat-based diets have a higher concentration of soluble NSPs, such that the degradation of both fractions is determinant for optimizing performance (Kiarie et al., 2014). Although broiler chickens and laying hens present distinct nutritional requirements due to their different production purposes (body mass gain and egg production, respectively), carbohydrase supplementation is advantageous for both categories (Zhang et al., 2014; White et al., 2021)). However, despite the volume of existing studies (Wu et al., 2004; Liu et al., 2017), gaps still persist regarding the efficacy of these enzymes under different formulation conditions, energy levels, ages, and commercial strains.

Given this, the objective was to evaluate the role of carbohydrases used in poultry feed in the degradation of carbohydrates present in feed ingredients, their effects on dietary nutrient utilization, and their influence on the digestibility of specific dietary substrates and the subsequent performance and nutrient digestibility of broiler chickens and laying hens.

5. Conclusions

The use of carbohydrases proved effective in recovering the performance and egg quality of laying hens fed diets reduced in metabolizable energy.

Supplementary Materials: The following supporting information can be downloaded at: <https://www.mdpi.com/xxx/s1>. Table S1: Akaike's information criterion (AIC) estimated to each covariance structures for variables measured in performance. egg quality and body composition assay of the layers.

Author Contributions: For research articles with several authors. a short paragraph specifying their individual contributions must be provided. The following statements should be used "Conceptualization, R.B.V., J.A.P., G.H.N.d.S. and E.P.d.S. methodology, R.B.V., R.C.M., R.K.G.M., M.B.d.L., L.O.d.S. and L.R.R.d.O. software, R.B.V., M.B.d.L. and E.P.d.S. validation, R.B.V., R.C.M., R.K.G.M., M.B.d.L. and E.P.d.S. formal analysis, R.B.V., M.B.d.L. and E.P.d.S. investigation, R.B.V. and E.P.d.S. resources, J.A.P., R.C.M., R.K.G.M. and E.P.d.S. data curation, R.B.V., M.B.d.L., J.A.P., G.H.N.d.S., L.O.d.S. and L.R.R.d.O. writing—original draft preparation, R.B.V., M.B.d.L., L.O.d.S., L.R.R.d.O. and E.P.d.S., writing—review and editing, R.B.V., J.A.P. and E.P.d.S. visualization, R.B.V., G.H.N.d.S., M.B.d.L. and E.P.d.S. supervision. E.P.d.S. project administration, R.B.V., M.B.d.L. and E.P.d.S. funding acquisition, R.C.M., R.K.G.M. and E.P.d.S. All authors have read and agreed to the published version of the manuscript.

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Institutional Review Board Statement: This study was conducted using white laying hens at the experimental facility of São Paulo State University (UNESP, Jaboticabal City—State of São Paulo). All procedures were approved by the Ethics Committee for the Use of Animals (CEUA) under Protocol number 3761/23.

Informed Consent Statement: Not applicable.

Data Availability Statement: The datasets generated and analyzed during the current study are available from the corresponding author upon request.

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Conflicts of Interest: Author Rosana Cardoso Maia. Rodrigo Knop Guazzy Messias were employed by the company BASF S.A. South America. The other authors declare that they have no conflict of interest.

Abbreviations

The following abbreviations are used in this manuscript:

AH	Albumen height
AIC	Akaike information criterion
ANOVA	Analysis of variance
AP	Albumen percentage
AW	Albumen weight
BMD	Bone mineral density
BW	Body weight
C	Contrasts
c	Chemical composition
cAsh	Ash composition
CEUA	Ethics Committee on Animal Use
CONCEA	National Council for the Control of Animal Experimentation
CV	Coefficient of variation
d	Chemical component deposition
dAsh	Ash deposition
dWater	Water deposition
DXA	Dual-energy X-ray absorptiometry
EBW	Empty body weight
EP	Egg production
ESP	Eggshell percentage
ESS	Eggshell strength
EST	Eggshell thickness
ESW	Eggshell weight

EW	Egg weight
FCR	Feed conversion ratio
FI	Feed intake
HU	Haugh unit
LD	Linear dichroism
LD	Linear dichroism
L-Met	L-methionine
L-Thr	L- threonine
Men	body weight
ME _n	Metabolizable energy
NC	Negative control
NSP	Non-starch polysaccharides
PC	Positive control
SEM	Standard error of mean
SI	Shape index
SPH	Sphericity
T1	Treatment one
T2	Treatment two
T3	Treatment three
T4	Treatment four
T5	Treatment five
TLA	Three letter acronym
TLA	Three letter acronym
YP	Yolk per centage
YW	Yolk weight

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