



IPB UNIVERSITY

**EVALUATION OF COCONUT FATTY ACID DISTILLATE
(CFAD) AS AN ENERGY SUPPLEMENT
FOR DAIRY CATTLE**

DISSERTATION

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**GRADUATE SCHOOL
NUTRITION AND FEED SCIENCE STUDY PROGRAM
BOGOR
2026**



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HIGHLIGHTS

RIKA ZAHERA. Evaluation of Coconut Fatty Acid Distillate (CFAD) as an energy supplement for dairy cattle. Supervised by DESPAL, IDAT GALIH PERMANA, and SATOSHI KOIKE.

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1. This study addresses the utilization of coconut fatty acid distillate (CFAD) as an alternative energy source for dairy cattle, aiming to optimize its protection as calcium soap; evaluate the effects of varying protection ratios and supplementation levels on *in vitro* rumen fermentation, microbial populations, and fatty acid biohydrogenation; and investigate the effects of protected CFAD supplementation on nutrient utilization, blood metabolomic profiles, and lactating dairy cow performance.
2. The research employs three experimental studies, including descriptive analysis of the protection characteristics of CFAD calcium soaps produced using CaO and CaCl₂ at Ca:CFAD mole ratios of 1.0, 1.5, 2.0, and 2.5; an *in vitro* study using a 5 × 4 factorial randomized block design with varying protection ratios (100:0, 75:25, 50:50, 25:75, and 0:100) and supplementation levels (0%, 1%, 2%, and 3%); and an integrated study comprising an *in vitro* experiment using a 3 × 3 factorial randomized block design with different forage:concentrate ratios (60:40, 50:50, and 40:60) and protected CFAD supplementation levels (0%, 2%, and 4%), followed by an *in vivo* evaluation of protected CFAD supplementation (0%, 1%, and 2%) under smallholder dairy farming conditions.
3. Results indicate that CaCl₂ and a 2.5 mole ratio of Ca:CFAD enhanced calcium soap formation, as supported by the lowest acid value and reduced carboxylic acid detected by NIRS and FTIR. Completely protected CFAD maintained rumen fermentation while increasing the relative abundance of the genus *Bacteroides*, nutrient digestibility, and CLA yield during fatty acid biohydrogenation. Supplementation with 2% protected CFAD in dairy cows improved ether extract and crude protein digestibility, enhanced milk production and lactation persistency, and altered serum fatty acids and metabolomic profiles without adversely affecting nutrient intake, milk composition, and energy balance.
4. The findings contribute to dairy nutrition and protected fat technology by providing an integrated understanding of CFAD utilization, from optimization of calcium soap formation to its *in vitro* and *in vivo* evaluation, offering new insights into the potential of CFAD as an alternative energy supplement for dairy cattle.
5. This study demonstrates the importance of optimizing the protection ratio and supplementation level for effective CFAD utilization, with integrated *in vitro* and *in vivo* studies supporting 2% protected CFAD supplementation as a potential alternative energy strategy to improve nutrient utilization and dairy performance.

SOROTAN

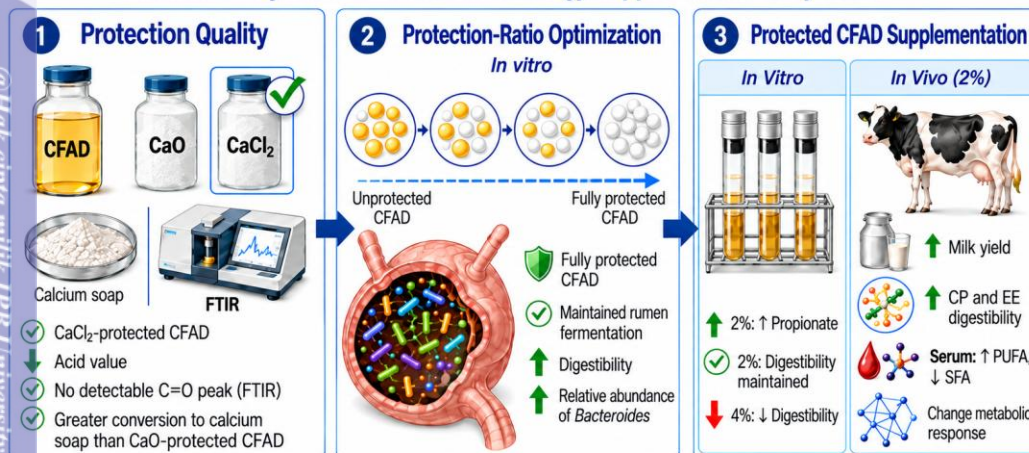
RIKA ZAHERA. *Evaluation of Coconut Fatty Acid Distillate (CFAD) as an energy supplement for dairy cattle*. Dibimbing oleh DESPAL, IDAT GALIH PERMANA, dan SATOSHI KOIKE.

1. Penelitian ini membahas pemanfaatan coconut fatty acid distillate (CFAD) sebagai sumber energi alternatif untuk sapi perah, dengan tujuan mengoptimalkan proteksinya dalam bentuk sabun kalsium; mengevaluasi pengaruh berbagai rasio proteksi dan level suplementasi terhadap fermentasi rumen secara *in vitro*, populasi mikroba, dan biohidrogenasi asam lemak; serta mengkaji pengaruh suplementasi CFAD terproteksi terhadap pemanfaatan nutrisi, profil metabolomik darah, dan performa sapi perah laktasi.
2. Penelitian ini menggunakan tiga tahap penelitian, meliputi analisis deskriptif karakteristik proteksi sabun kalsium CFAD yang diproduksi menggunakan CaO dan CaCl₂ pada rasio mole Ca:CFAD 1,0; 1,5; 2,0; dan 2,5; studi *in vitro* menggunakan rancangan acak kelompok faktorial 5 × 4 dengan berbagai rasio proteksi (100:0, 75:25, 50:50, 25:75, dan 0:100) dan level suplementasi (0%, 1%, 2%, dan 3%); serta studi terintegrasi yang terdiri atas studi *in vitro* menggunakan rancangan acak kelompok faktorial 3 × 3 dengan rasio hijauan:konsentrat yang berbeda (60:40, 50:50, dan 40:60) dan level suplementasi CFAD terproteksi (0%, 2%, dan 4%), yang dilanjutkan dengan evaluasi *in vivo* suplementasi CFAD terproteksi (0%, 1%, dan 2%) pada kondisi peternakan sapi perah rakyat.
3. Hasil penelitian menunjukkan penggunaan CaCl₂ dan rasio mole Ca:CFAD sebesar 2,5 meningkatkan konversi pembentukan sabun kalsium, dengan bilangan asam terendah dan berkurangnya gugus asam karboksilat yang terdeteksi melalui NIRS dan FTIR. CFAD terproteksi penuh mampu mempertahankan fermentasi rumen sekaligus meningkatkan kelimpahan relatif genus *Bacteroides*, pencernaan nutrisi, dan produksi CLA selama proses biohidrogenasi asam lemak. Suplementasi 2% CFAD terproteksi pada sapi perah meningkatkan pencernaan lemak kasar dan protein kasar, meningkatkan produksi dan persistensi susu, serta mengubah profil asam lemak dan metabolomik darah tanpa memberikan pengaruh negatif terhadap konsumsi nutrisi, komposisi susu, maupun keseimbangan energi.
4. Temuan penelitian ini berkontribusi terhadap bidang nutrisi ternak perah dan teknologi lemak terproteksi dengan memberikan pemahaman yang terintegrasi mengenai pemanfaatan CFAD, mulai dari optimasi pembentukan sabun kalsium hingga evaluasinya secara *in vitro* dan *in vivo*, serta memberikan wawasan baru mengenai potensi CFAD sebagai suplemen energi alternatif untuk sapi perah.
5. Penelitian ini menunjukkan pentingnya optimasi rasio proteksi dan level suplementasi untuk meningkatkan efektivitas pemanfaatan CFAD, dengan hasil studi *in vitro* dan *in vivo* yang terintegrasi mendukung suplementasi 2% CFAD terproteksi sebagai strategi sumber energi alternatif yang potensial untuk meningkatkan pemanfaatan nutrisi dan performa sapi perah.



GRAPHICAL SUMMARY

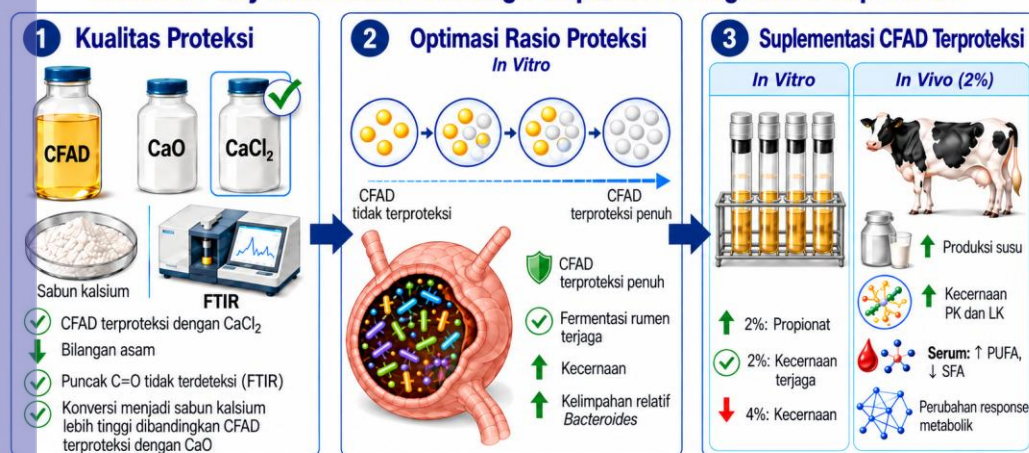
Coconut Fatty Acid Distillate as an Energy Supplement for Dairy Cattle



2% protected CFAD was the optimal supplementation level for dairy cattle.

RINGKASAN VISUAL

Coconut Fatty Acid Distillate sebagai Suplemen Energi untuk Sapi Perah



CFAD terproteksi 2% merupakan level suplementasi optimal untuk sapi perah.

SUMMARY

RIKA ZAHERA. Evaluation of Coconut Fatty Acid Distillate (CFAD) as an energy supplement for dairy cattle. Supervised by DESPAL, IDAT GALIH PERMANA, and SATOSHI KOIKE

Dairy cows require a high energy supply to support milk production. Insufficient energy intake can lead to a negative energy balance, mobilization of body reserves, and reduced productive performance. Dietary fat supplementation can increase the energy density of the ration without increasing the proportion of rapidly fermentable carbohydrates. Coconut fatty acid distillate (CFAD), a by-product of coconut oil refining, has the potential to serve as an alternative energy source because of its high medium-chain fatty acids (MCFAs). However, the antimicrobial properties of MCFAs may suppress rumen microorganisms, impair fiber degradation, and alter ruminal fermentation when CFAD is supplied in an unprotected form. Therefore, CFAD needs to be protected to minimize its adverse effects in the rumen while maintaining its potential as an energy source.

The first study was descriptively analyzed to evaluate the protection strength of CFAD calcium soaps produced using CaO and CaCl₂ at Ca:CFAD mole ratios of 1.0, 1.5, 2.0, and 2.5. The observed parameters included acid value determined by chemical analysis and carboxylic acid functional groups assessed using NIRS and FTIR. Increasing the calcium-to-CFAD mole ratio reduced the acid value of the calcium soap, indicating a greater degree of saponification conversion. Calcium soaps produced at a 2.5 mole ratio of Ca:CFAD were further evaluated using NIRS and FTIR. The NIRS results showed that CFAD calcium soap produced using CaCl₂ through the *double decomposition process* had lower reflectance than raw CFAD and CFAD calcium soap produced using CaO through the *modified fusion method*. This finding was supported by the FTIR results, which showed the absence of a C=O transmittance peak associated with carboxylic acids in the CFAD calcium soap produced using CaCl₂. These findings indicate that protection using CaCl₂ with a 2.5 mole ratio of Ca:CFAD achieved a higher conversion rate, thereby providing greater protection of fatty acids from early ruminal degradation, potentially reducing their antimicrobial effects, and improving energy utilization efficiency.

The second study evaluated five unprotected-to-protected CFAD ratios (100:0, 75:25, 50:50, 25:75, and 0:100) and supplementation levels of 0%, 1%, 2%, and 3% under *in vitro* conditions. The study used a 5 × 4 factorial randomized block design with three rumen fluid sources as blocks. The observed parameters included ruminal fermentability (pH, ammonia concentration, total volatile fatty acid concentration, and partial volatile fatty acid proportions), dry matter and organic matter digestibility, microbial populations, estimated methane production, and fatty acid biohydrogenation. Completely protected CFAD (0:100) resulted in the highest dry matter digestibility ($P < 0.05$). Increasing CFAD supplementation levels reduced dry matter and organic matter digestibility, as well as methanogen and protozoal populations ($P < 0.05$). Increasing the proportion of protected CFAD enhanced the relative abundance of *Bacteroides* and conjugated linoleic acid (CLA) production. CLA production was positively correlated with the C18:2 isomerization index and total C18 recovery. Based on these results, completely protected CFAD



(0:100) showed optimal rumen function, as indicated by higher dry matter digestibility and *Bacteroides* abundance, and was therefore selected for the third study.

The third study evaluated protected CFAD as an energy supplement for dairy cows through *in vitro* and *in vivo* experiments. The *in vitro* experiment evaluated forage-to-concentrate ratios of 60:40, 50:50, and 40:60 and protected CFAD supplementation levels of 0%, 2%, and 4%. The experiment used a 3 × 3 factorial randomized block design with three rumen fluid sources as blocks. The observed parameters included ruminal fermentability (pH, ammonia concentration, total volatile fatty acid concentration, and partial volatile fatty acid proportions), nutrient digestibility (dry matter, organic matter, neutral detergent fiber, and acid detergent fiber digestibility), and estimated methane production. Supplementation with 2% protected CFAD decreased the acetate proportion and acetate to propionate ratio while increasing the propionate proportion ($P < 0.05$). Supplementation at 4% reduced dry matter, organic matter, neutral detergent fiber, and acid detergent fiber digestibility ($P < 0.05$). These findings indicate that 2% protected CFAD was more effective in optimizing ruminal fermentation without reducing fiber digestibility. The *in vivo* experiment involved 18 lactating dairy cows supplemented with 0%, 1%, or 2% protected CFAD. The experiment used a randomized block design with three treatments and six dairy farmers as blocks. Supplementation with 1% and 2% protected CFAD increased crude protein digestibility from 60.18% in the control group to 68.75% and 67.63%, respectively ($P < 0.05$), and increased ether extract digestibility from 84.60% to 90.44% and 91.45%, respectively ($P < 0.01$). Supplementation with 2% protected CFAD resulted in the highest final milk yield of 13.40 kg d⁻¹ ($P < 0.01$), the smallest decline in milk yield of 0.64 kg d⁻¹ ($P < 0.01$), and the highest milk production persistency of 95.30% ($P < 0.05$). Protected CFAD supplementation also altered serum fatty acids. Metabolomic analysis identified cholesterol, 2-hydroxybutyric acid, phosphate, urea, 3-hydroxybutyric acid, and 1,5-anhydrosorbitol as the key metabolites discriminating among treatments. In conclusion, CFAD can be effectively protected in the form of calcium soap using CaCl₂ at a Ca:CFAD mole ratio of 2.5. Completely protected CFAD produced the optimal results in terms of dry matter digestibility and CLA production. Supplementation with 2% protected CFAD improved crude protein and ether extract digestibility, milk production, lactation persistency, and modified serum fatty acid composition and metabolomics. Therefore, protected CFAD at 2% supplementation can be used as an alternative energy supplement for lactating dairy cows.

Keywords: blood metabolomics, calcium soap, coconut fatty acid distillate, dairy cows, medium-chain fatty acids, rumen fermentation

RINGKASAN

RIKA ZAHERA. *Evaluation of Coconut Fatty Acid Distillate (CFAD) as an energy supplement for dairy cattle*. Dibimbing oleh DESPAL, IDAT GALIH PERMANA, dan SATOSHI KOIKE.

Sapi perah membutuhkan pasokan energi yang tinggi untuk mendukung produksi susu. Kurangnya konsumsi energi dapat menyebabkan neraca energi negatif, mobilisasi cadangan tubuh, dan penurunan performa produksi. Suplementasi lemak dalam ransum dapat meningkatkan densitas energi tanpa meningkatkan proporsi karbohidrat yang cepat difermentasi. Coconut fatty acid distillate (CFAD) merupakan hasil samping pemurnian minyak kelapa, berpotensi menjadi sumber energi alternatif karena mengandung tinggi asam lemak rantai menengah. Akan tetapi, adanya sifat antimikroba asam lemak rantai menengah dapat menekan mikroorganisme rumen, mengganggu degradasi serat, dan mengubah fermentasi rumen apabila diberikan dalam bentuk tidak terproteksi. Oleh karena itu, CFAD perlu diproteksi untuk mengurangi pengaruh negatifnya di dalam rumen sekaligus mempertahankan potensinya sebagai sumber energi.

Penelitian tahap 1 dianalisis secara deskriptif untuk mengevaluasi kekuatan proteksi sabun kalsium yang dihasilkan dari CaO dan CaCl₂ pada berbagai rasio mol Ca:CFAD (1; 1,5; 2; 2,5). Parameter yang diamati meliputi nilai asam secara kimia dan ikatan karboksilat menggunakan NIRS dan FTIR. Peningkatan rasio mol kalsium terhadap CFAD menurunkan bilangan asam sabun kalsium yang menunjukkan tingginya konversi saponifikasi. Sabun kalsium dengan rasio Ca:CFAD 2,5 diuji lebih lanjut menggunakan NIRS dan FTIR. Berdasarkan hasil NIRS, sabun kalsium CFAD dengan CaCl₂ (double decomposition process) menunjukkan nilai reflektansi lebih rendah dibandingkan CFAD dan sabun kalsium CFAD dengan CaO (modified fusion method). Hal ini juga didukung hasil dari FTIR yang menunjukkan tidak adanya puncak transmitansi C=O dari asam karboksilat pada sabun kalsium CFAD dengan CaCl₂. Hal ini menunjukkan bahwa proteksi menggunakan CaCl₂ dengan rasio mol 2,5 menghasilkan tingkat konversi yang lebih tinggi, sehingga asam lemak lebih terlindungi dari degradasi awal di rumen, berpotensi mengurangi efek antimikroba, serta meningkatkan efisiensi pemanfaatan energi.

Penelitian tahap 2 mengevaluasi lima rasio CFAD tidak terproteksi terhadap CFAD terproteksi, yaitu 100:0, 75:25, 50:50, 25:75, dan 0:100, pada level suplementasi 0%, 1%, 2%, dan 3% secara in vitro. Penelitian ini menggunakan rancangan acak kelompok faktorial 5 × 4 dengan 3 kelompok cairan rumen. Parameter yang diamati, antara lain fermentabilitas (pH, total amonia, VFA total dan parsial), pencernaan bahan kering dan organik, populasi mikroba, estimasi produksi metan, dan biohidrogenasi asam lemak. Hasil menunjukkan proteksi CFAD secara penuh (0:100) menghasilkan pencernaan bahan kering tertinggi ($P < 0,05$). Peningkatan level suplementasi CFAD menurunkan pencernaan bahan kering dan bahan organik serta menurunkan populasi metanogen dan protozoa ($P < 0,05$). Peningkatan proporsi CFAD terproteksi meningkatkan kelimpahan relatif *Bacteroides* dan produksi Conjugated Linoleic Acid (CLA). Produksi CLA berkorelasi positif dengan indeks isomerisasi C18:2 dan total recovery C18. Berdasarkan hasil tersebut, proteksi penuh CFAD (0:100) menghasilkan fungsi



rumen yang optimal dengan peningkatan pencernaan nutrisi dan kelimpahan genus *Bacteroides*, sehingga digunakan untuk penelitian tahap 3.

Penelitian tahap 3 mengevaluasi CFAD terproteksi sebagai suplemen energi pada sapi perah dengan pengujian *in vitro* dan *in vivo*. Penelitian *in vitro* mengevaluasi rasio hijauan terhadap konsentrat sebesar 60:40, 50:50, dan 40:60 serta suplementasi CFAD terproteksi sebesar 0%, 2%, dan 4%. Pengujian *in vitro* menggunakan rancangan acak kelompok faktorial 3×3 dengan 3 kelompok cairan rumen. Parameter yang diamati, antara lain fermentabilitas (pH, total ammonia, VFA total dan parsial), pencernaan nutrisi (bahan kering, bahan organik, NDF, dan ADF), populasi mikroba, estimasi produksi metan. Hasil menunjukkan suplementasi CFAD terproteksi sebesar 2% menurunkan proporsi asetat dan rasio asetat terhadap propionat serta meningkatkan proporsi propionat. Suplementasi sebesar 4% menurunkan pencernaan bahan kering, bahan organik, NDF, dan ADF. Hal ini menunjukkan CFAD terproteksi sebesar 2% lebih optimal dalam fermentasi rumen tanpa menurunkan pencernaan serat. Penelitian *in vivo* menggunakan 18 ekor sapi perah laktasi yang diberikan suplementasi CFAD terproteksi sebesar 0%, 1%, atau 2%. Penelitian ini menggunakan rancangan acak kelompok dengan 3 perlakuan dan 6 peternak sapi sebagai kelompok. Suplementasi sebesar 1% dan 2% meningkatkan pencernaan protein kasar dari 60,18% menjadi masing-masing 68,75% dan 67,63% ($P < 0,05$), serta meningkatkan pencernaan lemak kasar dari 84,60% menjadi 90,44% dan 91,45% ($P < 0,01$). Suplementasi sebesar 2% menghasilkan produksi susu akhir tertinggi, yaitu 13,40 kg hari⁻¹ ($P < 0,01$), penurunan produksi susu terkecil sebesar 0,64 kg hari⁻¹ ($P < 0,01$), dan persistensi produksi susu tertinggi sebesar 95,30% ($P < 0,05$). Suplementasi CFAD terproteksi juga mengubah proporsi asam lemak serum darah. Analisis metabolomik mengidentifikasi kolesterol, asam 2-hidroksibutirat, fosfat, urea, asam 3-hidroksibutirat, dan 1,5-anhidrosorbitol sebagai metabolit utama yang membedakan perlakuan. Oleh karena itu, dapat disimpulkan bahwa CFAD dapat diproteksi secara efektif dalam bentuk sabun kalsium menggunakan CaCl_2 pada rasio mol kalsium terhadap CFAD sebesar 2,5. Proteksi CFAD secara penuh memberikan hasil terbaik berdasarkan pencernaan bahan kering dan produksi CLA. Suplementasi CFAD terproteksi sebesar 2% meningkatkan pencernaan protein dan lemak, produksi susu, persistensi laktasi, serta memodifikasi komposisi asam lemak dan metabolomik serum. Dengan demikian, CFAD terproteksi sebesar 2% dapat digunakan sebagai suplemen energi alternatif untuk sapi perah laktasi.

Kata kunci: asam lemak rantai menengah, coconut fatty acid distillate, fermentasi rumen, metabolomik darah, sabun kalsium, sapi perah



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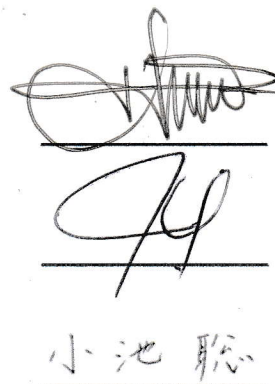
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PREFACE

All praise and gratitude are due to Allah subhanahu wa ta'ala for the grace and blessings, which enabled the author to complete this dissertation. The research, conducted from July 2023 to April 2026, focused on the development and evaluation of protected coconut fatty acid distillate as an alternative energy supplement for dairy cattle and is entitled "Evaluation of Coconut Fatty Acid Distillate (CFAD) as an energy supplement for dairy cattle."

I would like to express my sincere gratitude to my supervisors, Prof. Dr. Despal, S.Pt., M.Sc.Agr.; Prof. Dr. Ir. Idat Galih Permana, M.Sc.Agr.; and Prof. Satoshi Koike, Ph.D., for their invaluable guidance, encouragement, constructive comments, and thoughtful suggestions throughout the research and preparation of this dissertation. I also extend my sincere appreciation to Prof. Dr. Ir. Asep Sudarman, M.Rur.Sc., and Dr. Ir. Lilis Khodijah, M.Si., who served as examiners at my oral qualifying examination, for their constructive feedback and valuable suggestions for improving my research proposal. My sincere appreciation also goes to Dr. drh. Anita Esfandiari, M.Si., who served as the seminar moderator, as well as to the external examiners for their valuable comments, suggestions, and evaluation of this dissertation. I also thank Prof. Irma Isnafia Arief, as Chair of the Examination, and Prof. Dewi Apri Astuti, as the study program representative, for their time, attention, and support during this doctoral promotion examination. My sincere thanks also go to Dr. Dilla Mareistia Fassah, S.Pt., M.Sc., and Dr. Wulansih Dwi Astuti, S.Pt., M.Si., who served as examiners at my dissertation examination, for their constructive feedback and valuable suggestions for improving this dissertation. I gratefully acknowledge the Graduate School of IPB University and Hokkaido University for their institutional support in the implementation of the double-degree program. Special appreciation is extended to Prof. Hanny Wijaya and Assist. Prof. Maria Stefanie Dwiyanti for their invaluable guidance and assistance throughout the program.

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I sincerely hope that this dissertation will be beneficial to those who need it and will contribute to the advancement of knowledge, particularly in the fields of dairy cattle nutrition and feed technology.

Bogor, October 2026

Rika Zahera



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