Renewable Energy Program

North Dakota Industrial Commission

Application

Project Title: Valorization of DDGS

Applicant: North Dakota State University

Principal Investigator: Clairmont Clementson

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Amount of Request: $245,961

Total Amount of Proposed Project: $491,922

Duration of Project: 2 years

Point of Contact (POC): Amy Scott, Assistant Director Sponsored Programs Administration

POC Telephone: (701) 231-8045

POC Email: ndsu.research@ndsu.edu

POC Address: NDSU Dept 4000, PO Box 6050, Fargo, ND 58108-6050

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ABSTRACT

Objective:

Out of about 1.3 million tons of Dried Distiller's Grains with Solubles (DDGS) produced annually by North Dakota ethanol plants only one-tenth (10%) is used for local feeds. The remaining 90% is exported. DDGS exportation is challenged by logistics, flowability, and caking. US Grains Council has identified these issues as major limiting factors for use and profitability derived from DDGS as some railcar operators have limited the use of their railcars for the transport of DDGS. DDGS. Further, in spite of DDGS nutrient richness (30% protein and 40% fiber) it commands a low price. Hence developing technology to increase value and local use will be an opportunity to evade the current logistic challenges and increase the revenue stream. Thereby, improving the viability of North Dakota ethanol plants.

Valorizing DDGS entails adding value for these constituents. This project is intended to develop new process streams, create high value co-products that would increase the profitability-value chain of ethanol plants. The objective of this project is to develop a bioprocessing method aimed at extracting high-value proteins from corn Distiller's Dried Grains with Solubles (DDGS), a byproduct of ethanol production. This initiative is undertaken with the goal of enhancing the sustainability of the ethanol industry.

Expected Results:

The project aims to drive forward the utilization of corn DDGS, marking a substantial transition towards more sustainable and efficient practices within ethanol production and agriculture. By implementing optimized extraction methodologies, a notable increase in protein yields is anticipated. These enhancements are expected to result in statistically significant differences in protein recovery rates compared to current benchmarks, indicating an increase in efficiency that can be quantified.

Moreover, the detailed characterization of the extracted proteins is projected to unveil critical insights into their amino acid profiles, functional properties, and nutritional values. This phase is poised to reveal statistically significant variances in protein quality and functionality, potentially paving the way for new application avenues and influencing market dynamics for DDGS products.

Duration:

Two years

Total Project Cost: $491,922 (NDIC - $245,961, NDSU - $243,461, Tharaldson Ethanol - $2,500)

Participants:

Agricultural Biosystems and Engineering, North Dakota State University:

Clairmont Clementson, Assistant Professor, clairmont.clementson@ndsu.edu, 701-231-1810
Niloy Chandra Sarker, Research Specialist, niloy.sarker@ndsu.edu, 701-231-8602
Ademola Hammed, Research Assistant Professor, ademola.hammad@ndsu.edu, 701-231-8610

PROJECT DESCRIPTION

Objectives:

1. To identify optimized extraction methodologies that maximize protein yield from corn DDGS to reduce waste and enhance the overall sustainability of the ethanol production process.

2. To characterize the amino acid profiles, functional properties, and nutritional values of extracted DDGS proteins, aiming to elucidate the structure-function relationships that govern their utility in various applications.

Methodology

Sample Preparation: DDGS samples will be prepared according to standardized protocols to ensure consistency across all experiments. This will involve milling the DDGS to a uniform particle size of 0.425 – 1 mm and conditioning samples to specific moisture content.

Selection of Extraction Techniques: Extraction techniques will be identified for in-depth investigation following a comprehensive review of literature. This evaluation will encompass traditional methods such as solvent extraction and enzymatic hydrolysis, as well as novel approaches like ultrasound-assisted extraction and pulse electric field processing, which show potential for enhanced efficiency or sustainability. These techniques will be adapted as necessary and explored to identify the most effective methods for extracting high-value proteins from corn DDGS. Our research team, as documented by Bello et al. (2023), has highlighted the promising potential of extracting soybean meal protein using a reusable solvent. Initial experiments have proven successful, significantly increasing the concentrated protein yield. Expanding upon this groundwork, a preliminary study was conducted to DDGS. The simple solvent extraction method, conducted under alkali conditions, facilitates the migration of soluble into the solution in a cost-effective, one-step process. However, protein extraction is currently limited and this project is intended to concentrate the protein content.

For solvent extraction method, DDGS with a particle size of 0.425 – 1 mm will be immersed in varying concentrations of ammonium hydroxide, sodium hydroxide, hydrochloric acid, and water at a ratio of 1:10 (sample-to-solvent). This process will be conducted at different temperatures ranging from 25 - 55 ^{o}C, with continuous shaking for 6 - 12 hours. Subsequently, the mixture will undergo centrifugation at 10,000 g and 25 ^{o}C for 10 minutes to remove insoluble materials. Enzyme aided extraction of protein from DDGS will be carried out following similar process as above except that protease enzymes and 40^{o}C will be used.

Protein Isolation and Purification: Proteins will be isolated from DDGS using the optimized extraction methods identified in Objective 1. Subsequent purification steps, including dialysis or

ultrafiltration, will be employed to ensure the proteins are free from extraction residues, facilitating accurate characterization. The supernatant containing crude protein extracts will be collected and its pH will be adjusted to 4.5 – 5. The protein isolate will be allowed to sediment overnight. The sediment will be collected, washed twice with distilled water, and then centrifuged at 10,000g, 25 ^{o}C for 10 minutes. The protein will be collected onto an aluminum plate and dried at 60 ^{o}C for 12 hours. Protein purification will be achieved through repeated washing, precipitation, and membrane filtration.

Experimental Design:

A full factorial design will be implemented, wherein variables such as solvent type, temperature, pH, enzyme concentration (for enzymatic methods), and time will be systematically varied to evaluate their influence on protein yield. Each extraction will be conducted in triplicate to ensure statistical reliability. Subsequently, samples will be collected for the analysis of protein yield and purity.

Amino Acid Profiling: High-Performance Liquid Chromatography (HPLC) will be employed to analyze the amino acid composition of the purified proteins. This analysis will offer crucial insights into the nutritional quality of the proteins and their potential industrial applications. Furthermore, the amino acid profiles of the DDGS proteins will be compared against established reference protein standards to assess their completeness in essential amino acids.

Functional Properties Assessment: A series of analytical tests will be conducted to examine the functional properties of the DDGS proteins, such as solubility, emulsification capacity, foaming ability, and gelation strength. These properties will be assessed under varying environmental conditions (including changes in pH, temperature, and ionic strength) using spectrophotometry for solubility measurements and rheometry for determining gelation characteristics.

Nutritional Value Determination: The nutritional value of the proteins will be evaluated through *in vitro* digestibility tests, which will simulate the digestion process using simulated gastric and intestinal fluids. Furthermore, the bioavailability of essential nutrients will be analyzed to determine how effectively these nutrients can be absorbed and utilized by the body. This evaluation will provide crucial insights into the overall nutritional quality and potential health benefits of the DDGS proteins.

Structure-Function Relationship Analysis: Mass Spectrometry (MS) and Fourier-Transform Infrared Spectroscopy (FTIR) will be employed to elucidate the molecular structure of the proteins extracted from DDGS. These analytical techniques will provide detailed information about the amino acid sequence, post-translational modifications, and overall structural conformation of the proteins. By uncovering the molecular structure of the proteins, MS and FTIR analyses will facilitate the understanding of the relationship between protein structure and functional properties. This knowledge will guide the modification of protein structures to enhance specific functional attributes, thereby optimizing their suitability for various industrial applications.

Analytical Methods: The extracted proteins will undergo analysis using the Kjeldahl method to measure total nitrogen content, providing a basis for determining protein concentration. Additionally, the Bradford assay will be employed to accurately quantify the protein concentration present in the samples. Further characterization of the purity and composition of the extracted proteins will be conducted using Sodium Dodecyl Sulfate Polyacrylamide Gel Electrophoresis (SDS-PAGE) and High-Performance Liquid Chromatography (HPLC). These analytical techniques will provide insights into the molecular weight distribution, purity, and composition of the proteins, allowing for a comprehensive assessment of their quality and suitability for various applications.

Data Analysis: To assess the efficiency of various extraction techniques, analysis of variance (ANOVA) will be employed to evaluate the significance of differences in protein yield and purity under different extraction conditions. This statistical method will enable the comparison of extraction methods and conditions, providing valuable insights into their effectiveness in isolating proteins from corn DDGS.

Optimization: Based on the outcomes of the statistical analysis, the extraction method(s) demonstrating the highest effectiveness will be identified. These method(s) will undergo further optimization through additional experimentation. Subsequently, the scalability of the optimized method(s) will be evaluated for potential industrial application. Factors including cost-effectiveness, environmental impact, and practical feasibility will be taken into consideration during this assessment process.

Anticipated Results

Key expectations for this study include:

1. Identification of highly efficient extraction methods that significantly increase protein yields while minimizing process waste.

2. Establishment of a comprehensive amino acid profile detailing the composition of DDGS proteins, providing insights into their suitability for various applications such as animal nutrition or human dietary supplementation.

3. Elucidation of key functional properties of DDGS proteins, including solubility, emulsion stability, foaming capacity, and gelation properties, essential for optimizing their use in industries like food processing and biomaterials.

4. Nutritional evaluation to assess the proteins' digestibility and bioavailability, crucial for their incorporation into animal feed formulations to improve feed efficiency and support animal health.

5. Investigation of structure-function relationships of DDGS proteins to guide targeted modifications for enhanced functionality.

6. Promotion of diversified applications of DDGS proteins, including novel uses in bioplastics and other bio-based products.

Ultimately, this project aims to advance the utilization of DDGS proteins and encourage innovation in bioenergy byproduct utilization, leading to sustainable solutions and new opportunities in various industries.

FACILITIES and EQUIPMENT

Facilities:

The research will be conducted at the NDSU Pilot Processing Plant (*moving in summer 2024 to the new Peltier Complex under construction*) includes a large general wet/dry processing high bay laboratory (3,500 ft^2) space, an explosion proof laboratory (400 ft^2), quality control labs (480 ft^2), a bioproducts research laboratory (650 ft^2), and 2 offices. The labs contain ample chemical and sterile, laminar flow hood spaces. Standard utilities include 120, 220 and 440 V power, steam, vacuum, compressed air, distilled water, and 18 mΩ NANOpure water. A walk-in cold room and freezer and an explosion-proof freezer are available for sample storage. The Pilot Processing Plant provides space to faculty, staff and, students for both teaching and research in processing. It also provides space for industrial demonstration and services to local farmers who may need space and equipment.

Between 2020-2024, there were research projects at the Pilot Plant on soy meal protein extraction and characterization which resulted in approximately 4 peer-reviewed publications and many conference presentations. Hence, there are existing equipment to successfully carryout this research.

Equipment:

- Nicolet 6700 FTIR (smart ark, performer, drift accessories)
- UV Spectrophotometer
- Waters High Performance Liquid Chromatography (HPLC)
- Agilent Gas Chromatography
- Shaker water bath, Thermo MaxQ 7000 (3)
- Ultrasonic water bath
- Incubator Oven, Precision
- Gravity Convection Oven, Precision Scientific
- Convection ovens, Binder
- Vacuum Ovens VWR
- Drying Oven with perforated stainless shelfs
- Mettler AB204-S analytic balance 220g Max
- Mettler LJ16 Moisture analyzer
- VWR hotplate stirrers
- Multiwell Hot Plate/ Stirrer
- Digital Thermometers
- Orion A111 pH meter with probe
- Orion 2 star pH meter with probe
- Orion 230A portable pH meter with probe
- Pipet Washer

- Desiccators /drying cabinets
- Pipettes (fixed volume, pos-displacement)
- Bottle Dispensers/Titrators
- Volumetric flasks and glass beakers
- Separatory funnels
- Plastic tubes, bottles, beakers, cylinders
- Plastic Beakers, pipets, Tubes, Bottles
- Syringe & Syringe Accessories (barrels and filters)
- Filtration Accessories (glass filter,funnels, paper, clamps)
- Refrigerator/freezer

Resources:

Data Management: Primary data generated in this project will be digital. Data will be stored through the Center for Computationally Assisted Science and Technology (CCAST) hub at North Dakota State University. CCAST is an academic supercomputing facility made possible in part by NSF MRI Award No. 2019077. CCAST facilities are housed in the Research 1 and Research 2 buildings, both located in NDSU's Research and Technology Park. These are secure facilities, requiring card key access outside of normal business hours. Doors to hallways and rooms within the building are secured at all hours, and access for each room must be approved for each individual requiring access.

Network infrastructure at NDSU is managed and operated by NDSU Information Technology Services (ITS) which also participates in state and national initiatives to provide state-of-the-art networking capabilities to the entire upper-Midwest region. CCAST has a 100Gbps ScienceDMZ connection to the main NDSU router and then directly onto Internet2 for data transfers beyond the campus boundary. There is a 40 Gbps connection to the main NDSU router for communication with internal NDSU equipment. CCAST HPC systems and storage are:

(1) "Thunder" Cluster: This HPC cluster currently has 118 compute nodes (with 3,668 Intel CPU cores and 18.1 TB aggregate usable RAM), 22 GPUs, 2 login nodes, and several management nodes. The compute nodes include 20-core 64GB RAM Intel Ivy Bridge nodes (including 14x MIC nodes with Intel Phi 5110P accelerators), 32-core 1TB RAM (large-memory) Intel Sandy Bridge nodes, 44-core 128GB RAM Intel Broadwell nodes, and 40-core 192GB Intel Cascade Lake nodes, 36-core 192GB Intel Cascade Lake nodes. The 22 GPU cards include 8x Tesla P100 SMX2, 4x Quadro GP100, and 10x GeForce RTX 2080 Ti.

(2) "Thunder Prime" Cluster: This cluster, housed in the same room with Thunder, is the new flagship HPC cluster in the state of North Dakota. It currently consists of 81 compute nodes (with a total of 8,896 AMD CPU cores and 39.7TB aggregate usable RAM), 48 GPUs, 2 login nodes, and several management nodes. The compute nodes include 64-core 256GB RAM AMD EPYC 7662 nodes, 128-core 512GB RAM GPU-ready AMD EPYC 7662 nodes, and 128-core 1TB RAM GPU-ready AMD EPYC 7662 nodes. The GPUs include 11x NVIDIA A100 40GB PCIe, 12x NVIDIA A40 48GB PCIe, 24x NVIDIA A10 24GB PCIe, and 1x NVIDIA A2 16GB PCIe. The new cluster is growing with more compute nodes and GPUs to be added.

(3) Data storage: CCAST provides home, project, and scratch storage space for users. Home and project directories are backed up at regular intervals. Scratch space is not backed up, as it is intended for temporary storage of data being actively used by calculations. Data is hosted on parallel filesystems of 2.2PB storage capacity. In addition, there are a research data archive of 1.6PB for long-term data storage and an IBM tape archival system of over 6PB capacity for data backup. All these systems are expandable.

The HPC clusters and data storage systems at CCAST are housed in a 1,100 sq. ft. HPC server room equipped with 10 rear door heat exchangers (RDHX's) to remove up to 40 kW per rack. The server room can handle up to 500kW of IT load and is expandable to 1,000kW. All the equipment is fed by power that is conditioned by an uninterruptible power supply (UPS) system and backed up by a 2,000kW diesel generator with fuel onsite for 36 hours of runtime at full capacity. The generator is rated for continuous operation and has advanced emission controls for unlimited hours of operation per year.

The PI's research activities are provided 1TB of space within CCAST however for this project it is estimated to require at most 100GB. The data will be held in the CCAST facility for the duration of the project and for five (5) years thereafter. Subsequently, essential data would be retained on cloud storage. Collaborators on the project will be provided access to the data through the project. Access to the data by external sources will be provided as appropriate upon request.

Working with the leadership of the CCAST facility, the PI will ensure the implementation of the Data Management Plan. This resource is available to the project at no cost to North Dakota Industrial Commission.

Techniques to Be Used, Their Availability and Capability:

Technique	**Purpose/Use**	**Availability**	**Capability**
Solvent Extraction	To extract proteins from DDGS.	Available in our laboratories.	Efficiently separates proteins based on their solubility in various solvents, enabling the recovery of high-purity proteins.
Enzymatic Hydrolysis	To break down proteins into peptides and amino acids, enhancing their functionality and digestibility.	Available, requires specific enzymes that will be purchased.	Capable of targeting specific protein bonds, offering precise control over the breakdown process and the resulting functional properties.
Ultrasound-Assisted Extraction (UAE)	To enhance protein extraction efficiency and	Not available, samples will be sent to other	Utilizes ultrasonic waves to disrupt cell structures, improving the release of

	reduce processing time.	laboratories out of state.	proteins with minimal thermal damage.
Pulsed Electric Field (PEF) Processing	To permeabilize cell membranes and enhance protein yield.	Not available, samples will be sent to other laboratories out of state.	Applies short bursts of high voltage to improve cell membrane permeability, facilitating the extraction of proteins.
High-Performance Liquid Chromatography (HPLC)	For characterizing amino acid profiles and protein purity.	Available in our laboratories.	Offers high-resolution separation and quantification of proteins and amino acids, essential for detailed compositional analysis.
Mass Spectrometry (MS)	To further characterize proteins and peptides, providing molecular weight and structural information.	Not available, samples will be sent to other laboratories.	Provides detailed insights into the molecular structure of proteins, enabling the identification of functional properties and potential applications.
Fourier-Transform Infrared Spectroscopy (FTIR)	To assess the functional groups and bonding structures of proteins.	Available in our laboratories.	Allows for the rapid determination of macromolecular structures, aiding in the understanding of protein functionality.
Biochemical Assays	To evaluate the nutritional values of extracted proteins, including their digestibility and content of essential amino acids.	Available, chemicals will be purchased.	Enables the comprehensive assessment of protein quality and its suitability for various applications, from feed to food.
Spectrophotometric Assays (e.g., Bradford Assay)	To measure protein concentration in samples.	Available in our laboratories.	Allows for quick and accurate determination of protein quantities, critical for evaluating extraction efficiency.
SDS-PAGE			

	To separate proteins by molecular weight for purity and profile analysis.	Available in our laboratories.	Provides a visual representation of protein size distribution, assisting in the assessment of purity and integrity.

Environmental and Economic Impacts while Project is Underway:

Environmental Impacts: This project may encounter certain environmental impacts, including resource consumption, chemical use and waste generation, energy use and associated carbon footprint, laboratory emissions, and transportation impacts. These include the utilization of water, energy, and chemicals, which necessitates careful management to minimize environmental footprints. The project's reliance on solvents and enzymes may lead to waste that requires responsible disposal or recycling efforts to prevent environmental contamination. No significant emissions from laboratory processes are anticipated. By integrating, and adhering to environmental management standards, Ultimately, while the project carries some environmental considerations during its execution, the adoption of sustainable practices throughout the project's duration, such as minimizing waste, adopting energy-saving technologies, combined with the long-term benefits of developing sustainable extraction methods, presents a balanced approach towards achieving environmental sustainability and economic growth.

Economic Impacts: While the project involves substantial costs, the potential for generating high-value products from DDGS offers an opportunity for significant economic return. This includes direct profits from the sale of extracted proteins, as well as long-term benefits such as waste reduction and enhanced sustainability in the ethanol production process.

Ultimate Technological and Economic Impacts:

Technologically, this project aims to advance protein extraction methodologies and characterization by utilizing both traditional and novel techniques for improved efficiency and sustainability. This approach has the potential to transform the utilization of agricultural byproducts, establishing new standards for resource efficiency and environmental responsibility in industrial processes. Economically, the project seeks to transform corn DDGS from a low-value byproduct into a high-value commodity, reshaping the value chain of ethanol production. This transformation is anticipated to create new market opportunities, ranging from nutrition to bioplastics, diversifying revenue streams for the ethanol and agricultural industries and reducing reliance on single outputs.

The broader impacts of these technological advancements and economic shifts are expected to drive further research and investment in sustainable practices, leading to job creation, fostering industrial innovation, and contributing to a more resilient and sustainable economic model for corn-ethanol production. Through this project and similar initiatives, the future of industrial

byproduct management appears to be both sustainable and economically beneficial, demonstrating a model where environmental objectives and economic growth complement each other.

Technological Impacts

Innovation in Bioenergy Byproducts Utilization: The development of efficient extraction methodologies for proteins from DDGS represents a significant technological leap in valorizing bioenergy byproducts. This innovation paves the way for the bioenergy sector to transition from a linear to a circular economy model, where waste is minimized, and byproducts are effectively converted into valuable resources.

Advancement in Biofertilizer Production: Converting DDGS proteins into other bio-products such as biofertilizers will introduce a novel, sustainable agricultural input that can significantly reduce the dependency on petroleum. Biofertilizer, for example, has the potential to revolutionize farming practices by offering an environmentally friendly, cost-effective alternative that enhances soil health and fertility.

Cross-sectoral Technological Synergies: This research facilitates cross-sectoral technological synergies between the bioenergy and agriculture sectors. By interconnecting these sectors through innovative utilization of byproducts, it generates new opportunities for integrated technological solutions aimed at addressing sustainability challenges.

Economic Impacts

Creation of New Markets and Revenue Streams: The commercialization of technologies for extracting proteins from DDGS and producing bioproducts from the extracted proteins can unlock new markets and generate additional revenue streams for both the bioenergy and agricultural sectors. This diversification is crucial for enhancing economic resilience and fostering growth.

Job Creation and Economic Development: Implementing the research findings is expected to lead to job creation across multiple stages of the value chain, from research and development to manufacturing, marketing, and application. These jobs contribute to local and regional economic development, particularly in rural areas where bioenergy production and agriculture are key economic activities.

Cost Savings for Farmers: By extracting high-quality proteins from DDGS and its subsequent transformation into valuable products, farmers can significantly reduce their expenditures on conventional feed and fertilizer inputs. Enhanced feed quality from these proteins can lead to improved livestock health and productivity, translating into higher profitability. Additionally, the potential use of extracted proteins in agronomic practices could improve soil health and crop yields, further contributing to the economic and sustainable operations of farms.

Attracting Investment: The technological innovations and market opportunities generated by this research can attract investment from both the public and private sectors. Investment in sustainable technologies and products is critical for accelerating the transition to a green economy and stimulating economic growth.

Leadership and Export Opportunities: Establishing leadership in the technologies developed through this research can position our region and country as global leaders in sustainable bioenergy and agricultural solutions. This leadership can translate into export opportunities for technologies, products, and expertise, further expanding economic impact.

Why the Project is Needed:

North Dakota plays a significant role in the U.S. corn market, contributing to both national corn production and ethanol manufacturing. Ethanol production from corn is a thriving industry in the state, offering an alternative fuel source and bolstering the agricultural sector by creating a demand for surplus corn. However, this process also yields Distillers Dried Grains with Solubles (DDGS), a valuable byproduct primarily utilized as high-protein livestock feed. Approximately 1.3 million tons of DDGS are produced annually in North Dakota's ethanol plants, with a significant portion being exported (ND Ethanol Council, 2024).

To diversify the applications of DDGS and generate additional revenue for the state, while promoting biorenewable systems and sustainable agricultural practices, this study aims to develop a bioprocessing method for extracting high-value proteins from DDGS and characterizing these proteins. The global market for plant-based proteins is expected to witness substantial growth, underscoring the economic potential of enhancing the value of DDGS beyond its conventional use as animal feed. Moreover, optimizing the utilization of DDGS through bioprocessing technologies contributes to waste reduction and resource efficiency, aligning with environmental sustainability goals.

Overall, the development of bioprocessing methods to extract high-quality proteins from DDGS offers numerous benefits, including economic diversification, environmental sustainability, and the harnessing of emerging market opportunities in the plant-based protein sector. The following are broader rationale for developing bioprocessing methods to extract high-quality proteins from DDGS:

i. **Sustainability in Ethanol Production**: the ethanol industry generates vast amounts of DDGS as a byproduct. Currently, the potential of DDGS, particularly its protein content, is not fully utilized. This project will address the urgent need to enhance the sustainability of ethanol production by developing optimized methodologies for extracting proteins from DDGS. This approach aims to reduce waste and create value from byproducts, aligning with global sustainability goals.

ii. **Economic Diversification and Value Creation**: North Dakota's economy, with its strong emphasis on agriculture and energy, stands to benefit significantly from the diversification and value addition provided by this project. By extracting and characterizing high-quality proteins from DDGS, the project can open new markets, create jobs, and stimulate economic growth. This diversification is particularly critical in the face of fluctuating global oil prices and the increasing competitiveness of the agricultural sector.

iii. **Research and Educational Opportunities**: This project offers numerous opportunities for research and education, providing a platform for advancing knowledge in environmental sciences and agricultural technologies. It can foster partnerships between academic institutions, industry, and government agencies, creating a collaborative environment for innovation. Furthermore, the project will serve as a valuable educational tool, preparing students and researchers to tackle future challenges in renewable energy and sustainable agriculture.

iv. **Alignment with Policy and Environmental Goals**: The project aligns with both national and international goals for renewable energy production, waste reduction, and environmental protection. By demonstrating an economically viable model for converting ethanol production byproducts into valuable resources, the project supports policy objectives aimed at promoting sustainability and reducing environmental impacts. This alignment is crucial for securing funding, regulatory support, and public acceptance.

v. **Response to Climate Change**: Addressing climate change requires innovative approaches to reducing greenhouse gas emissions and enhancing carbon sequestration. The project's focus on reusing waste materials (DDGS) to produce valuable products represent an effort to provide an alternative to carbon-intensive agricultural inputs and promoting more sustainable land-use practices. By leveraging the untapped potential of DDGS, the project offers a practical solution to some of the challenges posed by climate change.

Reference

North Dakota Ethanol Council. (2024). Distillers Grains. Accessed: March 29, 2024. https://www.ndethanol.org/distillers-grains#:~:text=North%20Dakota's%20ethanol%20plants%20produce,tons%20of%20distillers%20grain%20annually.

Bello, I., Adeniyi, A., Mukaila, T., Hammed, A. 2023. Optimization of Soybean Protein Extraction with Ammonium Hydroxide (NH4OH) Using Response Surface Methodology. Foods, 12(7), 1515. https://doi.org/10.3390/foods12071515

STANDARDS OF SUCCESS

The Standards of Success for this project are defined by a set of integrated and strategic deliverables aimed at maximizing the value and sustainability of DDGS within North Dakota's ethanol and agricultural sectors. Success will be measured through the development and implementation of efficient, scalable extraction protocols that enhance the quality and yield of proteins from DDGS, alongside a comprehensive characterization of these proteins in terms of amino acid profiles, functional properties, and nutritional values, to uncover their potential applications.

Value to North Dakota: Enhancing the utility of DDGS, this project is poised to significantly boost revenue streams for local farmers and ethanol producers by reducing waste and promoting sustainability practices. By optimizing extraction methodologies to maximize protein yield from DDGS, the proposed study aligns with North Dakota's commitment to renewable energy and agricultural innovation. This effort will contribute to environmental sustainability and cements the state's position as a pioneering leader in sustainable agricultural practices and bioenergy resource management, offering a model for integrating agricultural byproducts into the circular economy.

Public and Private Sector Utilization: The utilization of project findings by both the public and private sectors underscores its broad relevance and potential impact. State and local government agencies with a focus on agriculture and energy are poised to integrate these insights into their policymaking, regulatory frameworks, and educational initiatives, enhancing the governance and support structure for sustainable practices. Meanwhile, in the private sector, ethanol producers, agricultural businesses, and biofertilizer manufacturers stand to directly benefit from the optimized extraction protocols developed through this project. This practical application is anticipated to yield immediate advantages, including cost savings, product diversification, and enhanced sustainability practices, showcasing the project's comprehensive approach to fostering innovation and efficiency across key industry sectors.

Commercialization Potential: The project's commercialization potential extends far beyond the immediate technological advancements in protein extraction from DDGS. These protocols are not just scientific achievements; they represent pivotal opportunities for businesses within and related to the agricultural and bioenergy sectors to innovate and expand their market presence. By developing new product lines or enhancing existing offerings, companies can tap into emerging markets and consumer demands for more sustainable and environmentally friendly products. This could lead to a broader adoption of DDGS-derived products across various industries, including but not limited to, animal feed, agriculture, and bioenergy, driving competitiveness and fostering economic growth.

Advancement of Education, Research and Development (R&D), and Marketing: The project's emphasis on enhancing education, research, development, and marketing within North Dakota represents a multifaceted approach to advancing sustainable energy and agricultural practices while also to cultivating a robust knowledge economy. By facilitating partnerships between academic institutions, research bodies, and industry stakeholders, this initiative serves as a vital conduit for knowledge transfer and innovation. Engaging students in hands-on research projects

directly related to the project will enrich their educational experience and prepare a new generation of professionals equipped with the skills and insights needed to tackle future challenges in renewable energy and sustainable agriculture.

The collaborative nature of these efforts fosters an environment where theoretical knowledge is integrated with practical application, ensuring that the findings and technologies developed through this project have a tangible impact on the industry. Furthermore, the active dissemination of research outcomes through conferences, publications, and workshops plays a crucial role in amplifying North Dakota's contributions to the renewable energy and sustainable agriculture sectors on a national and global scale. This will enhance the state's reputation as a leader in these critical areas and attract investment, talent, and interest nationally, thereby stimulating economic growth.

Job Creation and Preservation: The transformative potential of this project extends deeply into the economic fabric of North Dakota, particularly through its capacity to preserve existing jobs and catalyze the creation of new ones. By introducing new technologies and methodologies for extracting proteins from DDGS and their subsequent conversion into valuable products, the project directly supports the ethanol and agriculture sectors—key pillars of North Dakota's economy.

In the realm of job preservation, the project's advancements ensure that the ethanol production facilities and agricultural operations remain competitive in an increasingly sustainability-conscious market. This competitiveness is crucial for maintaining the viability of these sectors and the jobs they provide, from technicians and engineers in ethanol plants to agronomists and farm workers in the agricultural sector. The introduction of sustainable practices and technologies can lead to increased demand for North Dakota's bioenergy and agricultural products, further securing these jobs.

As the project moves from research and development into the commercialization phase, there will be a need for a wide range of professionals. Scientists and researchers will be essential for continuous innovation, while manufacturing roles will grow to produce the new technologies and products developed through the project. Furthermore, marketing and sales professionals will play a critical role in promoting these innovations, both domestically and internationally, expanding North Dakota's agricultural and energy sectors to new markets and opportunities.

Beyond the direct creation of jobs, this project aligns with the broader mission of promoting sustainable development and energy independence in North Dakota. By doing so, it ensures the long-term prosperity of the state, preserving existing jobs and creating a dynamic, innovative environment that attracts new businesses and talent to North Dakota. This holistic approach to economic development, grounded in sustainability and innovation, positions North Dakota as a forward-thinking leader, ready to meet the challenges and opportunities of the future.

Alignment with the Program's Mission: This project uniquely aligns with the mission of promoting renewable energy development and sustainability in North Dakota as it focuses on leveraging ND's rich agricultural and energy resources in a manner that sets a precedent for the rest of the country. By focusing on the extractability and utility of proteins from DDGS, the project

exemplifies a commitment to resource efficiency and the circular economy, principles that are increasingly becoming vital to environmental sustainability and economic resilience.

This commitment extends beyond the immediate environmental benefits of reducing waste and optimizing resource use. It embraces a holistic approach to sustainability that encompasses economic development, technological innovation, and societal well-being. In doing so, the project directly contributes to North Dakota's goal of becoming a leader in renewable energy, demonstrating how agricultural byproducts can be repurposed in ways that benefit the environment, economy, and communities.

In essence, the project embodies a forward-looking approach to renewable energy development, one that integrates economic development with environmental sustainability and social well-being. It is a testament to North Dakota's commitment to leading by example in the transition to a more sustainable and resilient energy future, ensuring that the state remains a vibrant, prosperous, and sustainable place for the next generations.

BACKGROUND/QUALIFICIATIONS

Our research team, as documented by Bello et al. (2023), has highlighted the promising potential of extracting soybean meal protein using a reusable solvent. Initial experiments have proven successful, yielding approximately 70% protein concentrated from the initial 40%.

Summary of qualification and experience

Clairmont Clementson

Clairmont Clementson, the Project Director, is a registered professional engineer (PE) and an Assistant Professor at North Dakota State University (NDSU). His initial professional training was in Mechanical Engineering, focused on mechanized agricultural systems. His graduate study provided a deeper understanding of biological systems and agricultural processes. Coupling these insights provided the foundation for post-graduate studies in grain handling, storage, processing and co-product utilization. The diversity of cereal and grains grown in the Northern Plains provides immense opportunities to contribute to the body of knowledge in this area.

His research is directed towards addressing the effects of climate change on post-harvest operations, and value addition for co-products. During his time at NDSU, he has completed a comprehensive study of the physical characteristics of corn varieties, indicating variances in corn varieties during post-harvest operations. Additionally, he is involved in improving the protein concentration extracted from soybean meal.

He is an active member of the American Society of Agricultural and Biological Engineering (ASABE), currently serving as chair of the PRS 707 – Food and Agricultural Waste Management and Utilization committee. He is also a certified Project Management Professional (PMP) with years of project management experience. These skills will be leveraged to effectively manage this project, ensuring the timely accomplishment of its objectives.

Niloy Chandra Sarker

The Co-PI, Niloy Chandra Sarker is a research specialist and affiliated graduate at North Dakota State University (NDSU). He has worked for the past 10 years at different level of his career on value added agriculture, agricultural product development including renewable energy. He is managing the NDSU pilot plant and extensively involved in bioprocessing research group supervision at NDSU. He has been working with the PD and Co-PDs for last 3.5 years on similar projects and presented research outcome in the form of presentation and peer reviewed journal article. Dr. Sarker is well experienced in every aspect of project management that includes but not limited to experimental design, purchasing supplies though procurement department, experimental, data collection, report writing, and manuscript preparation.

Ademola Hammed

Dr. Ademola Hammed, a research assistant professor and Co-Principal Investigator, holds dual PhDs in biotechnology engineering and bioprocess engineering. His academic and professional journey is marked by significant contributions to the field of protein extraction from various sources such as soybean, soymeal, and fish skin. Dr. Hammed possesses a profound expertise in the physicochemical characterization of proteins, encompassing their biological properties like antioxidant, anti-inflammatory effects, and digestibility.

His research activities extend into exploring and enhancing the functional attributes of proteins derived from both common and unconventional sources. Through his work, Dr. Hammed aims to address critical issues related to food sustainability and nutritional quality, focusing on the efficient utilization of bioresources. His multidisciplinary approach in engineering and biotechnology underpins his innovative strategies in protein science, contributing substantially to the academic community and industry standards. Dr. Hammed's role in his current project involves applying his extensive knowledge and skills to pioneer advancements in protein extraction methodologies and applications, ensuring significant impacts on health and nutrition sectors.

MANAGEMENT

This project will be managed using the five phases of project management: initiation, planning, execution, monitoring and control, and closure. During the initiation phase, the Co-PIs will review institutional procedures and guidelines related to procurement, accounting, and reporting. In the planning phase, the project requirements will be carefully assessed to identify the necessary skill set among graduate students, and an evaluation rubric will be developed for assessing potential candidates. Additionally, the equipment and materials required for experimentation will be determined, and a timeline for procurement and research assistant onboarding will be established.

During the execution phase, experimentation will be conducted according to the outlined methodology. In the monitoring and control phase, the Co-PIs will meet bi-weekly to assess the progress of the project, address any challenges, and explore opportunities to overcome them. Any extraneous circumstances that arise will be resolved during this phase.

Finally, in the closure phase, reports and financial statements will be prepared and presented as necessary for the project.

Milestones	2024				2025							
	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug
Initiation												
Planning												
Execution												
Monitoring and control												
Closure												

TIMETABLE

Please provide a project schedule setting forth the starting and completion dates, dates for completing major project activities, and proposed dates upon which the interim reports will be submitted.

Milestones	2024				2025							
	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug
Develop initial protein extraction methodologies from DDGS.												
Select and optimize the best protein extraction techniques.												
Characterization of extracted proteins												
Reporting and presentation												

BUDGET

Project Associated Expense	NDIC's Share	Applicant's Share (Cash)	Applicant's Share (In-Kind)	Other Project Sponsor's Share
Personnel	91,600	102,621		
Fringe Benefits	2,748	46,685		
Supplies	24,280			2,500
Fees	51,000			
Tuition		26,968		
Indirect Costs	76,333	67,187		
Total	$245,961	$243,461		$2,500

NDIC Share

Two graduate research assistants will be hired for two years at a salary of $22,900/year. The students will conduct research on the bioprocessing of DDGS protein as described in the proposal. Total $91,600.

Fringe benefits for the graduate research assistants is calculated at 3%. Total $2,748.

Supplies needed to carryout the proposed work includes citrate buffer solutions ($750), ultra-filtration membrane ($2,500), size exclusion gel ($2,500), consumables, such as flask, beakers, pipette tips, etc ($4,500), ion exchange gel ($8,880 for six units), NaOH ($1,250), Braford reagent ($750), HCL ($650), and Enzymes ($2,500). Total $24,280.

Fees include: 1) Ultrasound-Assisted Extraction; cost estimated for 75 samples at $150/sample based on current rates at external labs ($11,250). 2) Pulsed Electric Field processing; cost estimated for 75 samples at $130/sample based on current rates at external labs ($9,750). 3) Mass Spectometry performed at University of North Dakota; cost for 250 samples at $120/sample ($30,000). Total $51,000.

Indirect costs are calculated using NDSU's negotiated rate of 45% of modified total direct costs (MTDC). The MTDC is $169,628 x 45%. Total $76,333.

Applicants' share

Clairmont Clementson will lead this effort and devote 20% effort both years for a total salary of $40,289. He will lead the project, coordinate communication with stakeholders, lead Bi-monthly project meetings, and coordinate communication of research findings through reports and briefings. He will also assist with protocol development and evaluation of project data.

Niloy Chandra Sarker will devote 30% effort both years for a total of $36,119. He will procure and maintain material supplies. He will also ensure the maintenance and proper functioning of equipment, provide daily support and equipment training as necessary.

Ademola Hammad will devote 20% effort both years for a total of $26,213. He will conduct laboratory training and protocol guidance to graduate students. He will also coordinate laboratory activities and perform preliminary evaluation of data.

Fringe benefits are calculated at 36% of salary for PI Clementson, 55% for Co-PI Sarker, and 52% for Co-PI Hammed. Total $46,685.

Tuition for graduate students is calculated at $421.39/credit x 16 credits/year for $6,742 per student, per year. Total $26,968.

Indirect costs are calculated using NDSU's negotiated rate of 45% of modified total direct costs (MTDC). The MTDC is $149,306 x 45%. Total $67,187.

Other Sponsor's Share

Tharaldson Ethanol will provide the DDGS at a value of $2,500. See attached letter.

If Less Funding is Available

If 12-18% less funding is available than requested, the project's objectives will still be attainable. If the funding available is reduced by more than 18% of the requested amount, further assessments will be made with the intent of scaling down the project to achieve sufficient insights from the project objectives.

May 21, 2024

RE: Agricultural and Biosystem Engineering, NDSU's Project "Valorization of DDGS"

Dear North Dakota Industrial Commission:

I am writing this in support of the "Valorization of DDGS" for the North Dakota Industrial Commission. My name is Ryan Carter and I am the Chief Operating Officer of Tharaldson Ethanol, Casselton, North Dakota.

This study seeks to diversify the applications of DDGS and generate additional revenue while promoting biorenewable systems and sustainable agricultural practices, offering significant economic return. The broader impacts of these technological advancements and economic shifts are expected to drive further research and investment in sustainable practices, leading to job creation, fostering industrial innovation, and contributing to a more resilient and sustainable economic model for corn-ethanol production. This project aligns with the broader mission of promoting sustainable development and energy independence in North Dakota.

For these reasons, Tharaldson Ethanol is supportive of this project. Should this proposal be funded, we will provide the DDGS for this project, which is estimated at $2,500. This project is intended to develop new process streams and create high-value co-products, thereby increasing the profitability-value chain of ethanol plants.

Please contact me if you have questions about this letter of support.

Sincerely,

Ryan Carter
Chief Operating Officer, Tharaldson Ethanol
Telephone: 7014374000 | 7013473314
Email: rcarter@tharaldsonethanol.com

CONFIDENTIAL INFORMATION

Applicant is not requesting confidentiality.

PATENTS/RIGHTS TO TECHNICAL DATA

Applicant does not have any related patents. NDSU requests a level of confidentiality to safeguard the novelty of the idea and to allow NDSU to publish on the findings.

STATE PROGRAMS AND INCENTIVES

None.