# AFFIDAVIT OF DAVID W. ROGERS

## STATE OF FLORIDA COUNTY OF NASSAU COUNTY

Before me, the undersigned authority, personally appeared, DAVID W. ROGERS, who, being duly sworn, deposes and says:

- 1. I am over the age of eighteen years and have personal knowledge of all facts set forth in this Affidavit.
- 2. I am an environmental scientist currently employed as the Environmental Manager for the Rayonier Performance Fibers, LLC Fernandina plant ("RYAM"). As Environmental Manager, I am responsible for all aspects of environmental compliance for RYAM's existing cellulose manufacturing facility located in the City of Fernandina Beach ("the RPF Plant").
- 3. I also serve as the Biomaterials Sales Manager for RYAM. In my role as Biomaterials Sales Manager, I am responsible for the management and sale of biomaterials generated by the RPF Plant.
- 4. As a result of my current roles, I am familiar with the technical aspects of the 2G-Bioethanol project being proposed at the RPF Plant ("the Project").
- 5. I have also reviewed the scientific and technical information set forth in the Memorandum supplied on RYAM's behalf ("the Memorandum"), a true and correct copy of which is attached to this Affidavit as **Affidavit Attachment #1.**
- 6. Based on my review of the Memorandum, the scientific, regulatory, and technical sources cited therein, my personal knowledge, and my training and experience as an environmental scientist, including my work at the RYAM Plant for more than 20 years, it is my professional opinion that the scientific and technical statements made regarding the proposed Project in the Memorandum are accurate within a reasonable degree of scientific and technical certainty.

## Nature of the Project.

- 7. The RPF Plant's existing operation produces spent sulfite liquor ("SSL") as a byproduct. The SSL contains residual biomass materials that are not part of the specialty cellulose products RYAM sells to its customers. The RPF Plant proposes to use the sugars contained in this biomass by fermenting them to produce ethanol.
- 8. In the existing RPF Plant process, the SSL byproduct is separated from the pulping process and evaporated to concentrate the remaining biomass solids. From there, the SSL is either burned in the sulfite recovery boiler to generate energy and recover valuable raw materials that are reused in the pulping process or is transferred to LignoTech Florida, LLC (co-located at the RPF Plant site), which utilizes some of the SSL to produce wet and dry lignosulfonate products.
- 9. The Project itself consists of a variety of equipment to be installed on the RPF Plant property, which either enable fermentation or support physical separation of water from the bioethanol produced. This equipment includes continuous fermenters, centrifuges to allow yeast recycling, a distillation column for removing water and other liquid components from the ethanol, molecular sieves to further "dry" the ethanol by physically removing water molecules, and storage tanks. The footprint of the RPF Plant will not be expanded by the Project.
- 10. The Project as designed does not pose threats to environmental resources. It will be sited within the existing footprint of the RPF Plant, will be located outside of the floodplain as depicted on the Vicinity and Floodplain Map, and will comply with and incorporate all applicable wetland buffers. It is designed to minimize water usage and wastewater generation, and will comply with the RPF Plant consumptive water use permit and with the wastewater discharge permit. The design will also include primary and secondary containment systems to prevent unintended discharges to the wastewater treatment system and to soil. Yeast used in the process will be recycled with centrifuges or neutralized to eliminate the possibility of yeast leaving the RPF Plant. The Project will require no change in hours of operation, and will not cause fumes,

glare, noise or heat. Visual impacts will remain virtually unchanged with substantial buffering and screening along the current RPF Plant site boundaries. The Project will add a few thousand square feet of impervious surface to the property, insignificant for the existing 660- acre parcel.

- 11. The Project will reduce the overall air emissions from the RPF Plant by reducing emissions from the sulfite recovery boiler. Emissions are reduced because, sugars will be fermented and will not be burned when the SSL is returned to the recovery boiler nor will RYAM replace them with any other type of fuel, like natural gas. To meet market-driven specifications for 2G-BioEthanol and be considered "green fuel," implementation of the Project must be energy neutral to the overall RPF Plant operations. The portion of the fermented SSL that will be sent to LignoTech Florida will continue to be sold and processed as it is now.
  - 12. The Project will generate approximately three truck trips per day.
- 13. The engineering design of the Project was developed in collaboration with local fire officials and the Company's insurance provider to address the potential flammability of ethanol, and includes dedicated fire prevention technologies and fire suppression systems that adhere to applicable federal, state, and local codes, standards, and regulations. As described in detail within the application, the fire safety systems will include concrete containments walls around fermentation, distillation and storage, floating roof tanks, process and ambient monitoring, control system interlocks and alarm systems, fire suppression systems using non-PFAS alcohol foams, and firefighting equipment.
- 14. The location of the process and storage tanks within the footprint of the existing industrial facility were also determined to minimize risk. The proposed location is a safe distance from the pulping operations and related chemical storage, and a safe distance from the community around the facility.

15. RYAM's expert safety consultant conducted an independent evaluation of the potential impacts of a worst-case scenario event. This scenario assumes that none of the safety systems discussed above are in place. The study, which is attached to RYAM's application, concluded that even in that highly unlikely scenario, the area of highest risk extends only 25 meters (0.0155 mile or 82 feet) from the storage tanks and the area of minor risk, such as broken windows, extends only 120 meters (0.0745 mile or 393.7 feet). Both distances are well within the boundaries of the current RPF Plant footprint and well away from the pulping operations and existing chemical storage associated with the pulping operations.

## The 2G-Bioethanol Production Process.

16. RYAM proposes to implement a process that generally includes two stages: 1) fermentation, where sugars are converted into ethanol, and 2) distillation and drying, where the ethanol is separated and purified.

### Fermentation.

- 17. Fermentation is a well-known natural process used by humanity for thousands of years to make beer, liquor, bread, yogurt, cheese, and other similar products. It relies on living organisms, typically microorganisms, to convert organic substrates into the desired product—in this case, ethanol for fuel. Industrial fermentation follows this same natural process and is often referred to within the industry as "bioprocessing" or "biotechnology" rather than chemical manufacturing.
- 18. In forming my opinions regarding the biological nature of the proposed fermentation process, I have reviewed, relied upon, and agree with Glen Fox's expert paper titled *Ethanol production from natural sources of sugar* and the statement of local distiller Roger Morenc who has confirmed that the same type of fermentation proposed by the Project, "is a natural biological process and actually happens in the wild in a large number of instances." A

true and correct copy of Glen Fox's paper and of Roger Morenc's statement is attached hereto as Affidavit Composite Attachment 2.

- 19. Specifically in the Project, sugars contained in the SSL will be converted by yeast to ethanol. This 'conversion' is a simplified way of saying that the structure of the sugar will be changed into a new structure, an alcohol. The sugars are changed over several steps where different enzymes create a new structure, being an ethanol molecule.
- 20. In developing my expert opinion regarding the biologic nature of the SSL conversion process, I have also reviewed, relied upon, and agree with the article, The Scale-Up of Microbial Batch and FedBatch Fermentation Process, a true and correct copy of which is attached hereto as **Affidavit Attachment** 3. This article is the type of scientific statement analyzed and reasonably relied upon by environmental scientists working in my field. This article was also relied on by Medardo Monzon in developing his publicly stated opinions, which further confirm the biologic nature of the SSL conversion process.
- 21. The article explains "[e]thanol can be made synthetically from petroleum or by microbial conversion of biomass materials through fermentation." Thus, there are two distinct ways to make ethanol one a synthetic chemical manufacturing process and one a biologic, fermentation process. This article further identifies the three steps for a natural fermentation process: "The fermentation method generally uses three steps: (1) the formation of a solution of fermentable sugars, (2) the fermentation of these sugars to ethanol, and (3) the separation and purification of the ethanol, usually by distillation." For the Project, step one already takes place in the current existing pulping process when the SSL is created.
- 22. Based on my knowledge of the Project, my technical expertise as an environmental scientist, and my review of the technical literature, it is my expert opinion within a reasonable degree of scientific and technical certainty that the fermentation involved in the production of 2G-

Bioethanol is a biologic, rather than chemical, process and therefore does not constitute "chemical manufacturing" as that phrase is understood within the industry.

## Distillation.

- 23. Just as the fermentation portion of the Project does not fit within a commonly understood usage of the phrase "chemical manufacturing," it is my expert opinion that neither the phrase "chemical manufacturing" nor the phrase "chemical refining," as those terms are commonly understood by the industry, applies to "distillation and drying" proposed by the Project.
- 24. Distillation is a physical separation process based on differences in boiling points of components in a mixture. It involves heating the mixture to convert the more volatile component(s) into vapor, which is then condensed back into a liquid and collected separately. The process of distillation typically involves a distillation column or still, where the mixture is heated, and the vapor is collected and condensed.<sup>1</sup>
- 25. No new chemical is introduced, nor is any new chemical produced. Substances are simply separated from one another. The Project would produce 2G-Bioethanol in a manner identical to the way Marlin & Barrel makes liquor.
  - 26. Roger Morenc owner of Marlin & Barrel describes distilling as follows:

Once all of the potential alcohol is made in a ferment then its [sic] time to take the part you want and discard the rest. Since ferments are a natural process [sic] you'll find that a small amount of other-than ethyl alcohol can be present. Said another way, even though the vast majority of alcohol made is ethyl alcohol, there will be other types present. Each will have its unique molecular weight and that directly corresponds to a unique boiling point. As the ferment is distilled, the liquid is heated and the lighter compounds that require less heat to change from a liquid to vapor state will climb the sill [sic] column first and further then those needing more energy to get to their vapor form. In this way a still organizes all of the different molecule types present in a finished ferment. From that position of stratification a distiller can extract the desired piece of the ferment, cool it to liquid temperature again and then collect

6

<sup>&</sup>lt;sup>1</sup> This definition of distillation is so commonly understood that one can find it recited in general science resources like https://www.britannica.com/science/distillation.

- it. In no part of this process are chemicals being changes [sic]. This is simply a liquid sifter of sorts. (emphasis supplied).
- 27. The same process described by Mr. Morenc applies to the distillation process proposed by the Project.
- 28. Similarly, drying is another way to separate molecules from one another based on size. The Project anticipates using molecular sieves to separate water and ethanol molecules from one another. The molecular sieve will trap water molecules in a bed of adsorbent beads with small pores that small water molecules can pass through and be adsorbed, thereby allowing the ethanol molecules to pass through. This allows water content to be removed without introducing any new chemical or a new process, and no new chemical is produced. Pre-existing substances are simply separated from one another.
- 29. Based on my knowledge of the Project, my technical expertise as an environmental scientist, and my review of the technical literature, it is my expert opinion within a reasonable degree of scientific and technical certainty that the distillation and drying involved in the production of 2G-Bioethanol do not constitute either "chemical manufacturing" or "chemical refining" as those phrases are understood within the industry.
- 30. Other scientific articles that support my expert opinions regarding the nature of the production processes described herein include the following:
  - a. Sergi Maicas. The Role of Yeasts in Fermentation Processes. 2020;
  - b. Ethanol from Cellulose: A General Review by P.C. Badger;
  - c. Fractional Distillation vs. Refining What's the Difference?, This vs. That (Sept. 24, 2023), <a href="https://thisvsthat.io/fractional-distillation-vs-refining">https://thisvsthat.io/fractional-distillation-vs-refining</a>; and
  - d. Institute for Advanced Learning and Research, *Industrial Fermentation Supporting*a Growing Industry, IALR (Feb. 28, 2022), <a href="https://www.ialr.org/industrial-fermentation-supporting-a-growingindustry/#:~:text=At%20its%20basic%">https://www.ialr.org/industrial-fermentation-supporting-a-growingindustry/#:~:text=At%20its%20basic%</a>

20level%2C%20fermentation,then%20harvested%2C%20packaged%20and%20s old.

31. The scientific articles identified in paragraph 30, above, are the type of scientific publications upon which environmental scientists in my field reasonably rely, or which document scientific facts and processes generally understood by environmental scientists in my field. True and correct copies of the articles are attached hereto as **Composite Affidavit Attachment 4.** 

## The Project's Air Permit Application.

- 32. As noted above, my role as Environmental Manager encompasses ensuring the RPF Plant's compliance with applicable environmental permitting and regulatory requirements. My responsibilities will extend to the proposed Project. Thus, I have professional and technical expertise regarding the Federal and state laws and regulations that apply to the type of facilities proposed by the Project, as well as the Federal and state laws and regulations and guidance sources referenced in the Memorandum and their relationship to the Project.
- 33. The RPF Plant and the Project are subject to regulation by the Environmental Protection Agency of the United States (EPA) and the Florida Department of Environmental Protection (FDEP) under several federal and state environmental regulatory schemes, which include the federal Clean Air Act (CAA) and corresponding Florida regulations. The CAA is the comprehensive federal law that regulates air emissions from stationary sources such as the RPF Plant. The main goal of the CAA is to protect the public health and the environment by regulating emissions of certain air pollutants. FDEP acts as the statewide enforcer of the EPA's regulatory framework under the CAA and has authority to issue CAA permits in Florida on behalf of the EPA.

- 34. RYAM was required to apply to the FDEP under Title V of the CAA for an air permit to allow it to construct the Project. The application was prepared to allow FDEP to properly evaluate the issuance of an air permit.
- 35. The CAA is implemented through an extensive list of federal and state administrative regulations. Various regulations implementing the CAA include New Source Performance Standards "NSPS", "Prevention of Significant Deterioration" requirements, and National Emission Standards for Hazardous Air Pollutants "NESHAPS". The specific CAA requirements for the Project are set forth in RYAM's permit application submitted in 2023 to FDEP. None of the application regulations apply to evaluating land use applications.
- 36. On October 31, 2024, FDEP determined that the Project's air permit application was sufficient and issued a construction air permit.

Other Federal and States Laws and Regulations: Purpose and Relationship to the Project's Type of Land Use.

- 37. Certain land uses, including "chemical manufacturing" and "chemical refining" are prohibited under the City of Fernandina Beach's Comprehensive Plan and Land Development Code; however, those terms are not defined and must be interpreted.
- 38. While not definitive, various environmental laws and regulations, including the CAA and Florida's implementing regulations that were included in the Project's air permit application, provide guidance because they demonstrate that the EPA *does* differentiate facilities that manufacture ethanol through chemical synthesis from those that use biological synthesis.
- 39. The EPA has determined that facilities that produce ethanol through fermentation are not considered synthetic organic chemical manufacturing industry (SOCMI) sources subject to regulation under applicable portions of 40 Code Federal Regulations Part 60.

- 40. Pursuant to a series of Applicability Determinations by the EPA analyzing the applicability of various provisions of 40 Code Federal Regulations Part 60 to ethanol manufacturing facilities, the EPA determined that although ethanol is listed as a chemical affected by both "NSPS" Subparts RRR and NNN, background documentation created during the development of both standards indicated that the creation of ethanol by fermentation should properly be excluded from the scope of both NSPS, Subparts RRR and NNN.
- 41. Consistent with this interpretation, the EPA specifically excluded ethanol production by fermentation from its definition of "chemical process plants." For example:

40 CFR § 51.165 provides:

Permit requirements.

- (a) \* \* \*
- (1)\*\*\*
- (iv) \* \* \*
- (C) \* \* \*
- (20) Chemical process plants-The term chemical processing plant shall *not* include ethanol production facilities that produce ethanol by natural fermentation included in NAICS codes 325193 or 312140; (emphasis supplied).
- \* \* \* \*
- (4) \* \* \*
- (xx) Chemical process plants-The term chemical processing plant shall *not* include ethanol production facilities that produce ethanol by natural fermentation included in NAICS codes 325193 or 312140;
- 42. One of the EPA's policy rationales for adopting these exclusions was to correct the disparate treatment related to the production of ethanol for fuel versus the production of ethanol intended for human consumption by applying two different major source thresholds. Because the two manufacturing processes are substantially similar, EPA stated that the process should be treated and regulated identically for purposes of the PSD and Title V regulations under the CAA, regardless of the intended product use. EPA's discussion of NAICS code 325193 in 72 FR 24060 also makes clear that it considers ethanol production distinct from the broader classification of chemical manufacturing in NAICS code 325.

- 43. FDEP has adopted regulations in the Florida Administrative Code that mirror the Code of Federal Regulations, including the exclusion of ethanol production facilities that produce ethanol through natural fermentation processes from the definitions of "chemical processing" and "chemical process plant."
- 44. Rule 62-210.200(156), Florida Administrative Code, provides the definition of "Major Stationary Source" and expressly excludes ethanol production facilities that produce ethanol by natural fermentation:
  - (156) "Major Stationary Source" -
  - (a) A major stationary source is:
  - 1.... chemical process plants (the term "chemical process plants" shall not include ethanol production facilities that produce ethanol by natural fermentation included in North American Industry Classification System (NAICS) codes 325193 or 312140)...
  - (b) ...
  - (c) The fugitive emissions of a stationary source shall not be included in determining for any of the purposes of this definition whether it is a major stationary source, unless the source belongs to one of the following categories of stationary sources:...
    1-19.
  - 20. Chemical process plants (the term "chemical process plants" shall not include ethanol production facilities that produce ethanol by natural fermentation included in North American Industry Classification System (NAICS) codes 325193 or 312140),... 21-27.
- 45. EPA's website includes an information page on "chemical manufacturing" in the context of its Toxic Release Inventory (TRI) Program and how TRI chemical wastes are managed in the chemical manufacturing sector (defined as facilities reporting their primary NAICS code as 325). The page includes a "Chemical Manufacturing Facilities by Subsector" pie chart and provides a summary of operations in the chemical manufacturing sector. According to EPA, operations in the chemical manufacturing sector include:

Basic chemicals facilities produce large quantities of chemicals that are often used to make other chemicals or products. Basic chemicals include petrochemicals, industrial gases, and synthetic dyes and pigments.

Coatings and adhesives facilities mix pigments, solvents, and binders into architectural and industrial paints; manufacture paint products such as paint removers and thinners; and manufacture adhesives, glues, and caulking compounds.

Resins and synthetic rubber facilities manufacture resins, plastic materials, synthetic rubber, and fibers and filaments.

- 46. Facilities in the "Other Chemical Products" subsector make chemicals for a wide variety of applications. These include chemicals used in photography, explosives, inks and toners, and transportation equipment like antifreeze or brake fluid. Facilities that manufacture ethanol through a biological process are not included in the descriptions of chemical manufacturing, even though EPA is clearly aware of the industry's use of biological processes to make ethanol.
- 47. A true and correct copy of the EPA's website page is attached hereto as **Affidavit Attachment 5** to this Affidavit. The EPA's website is the type of information upon which environmental scientists reasonably rely in determining the applicability of federal regulations to specific processes.
- 48. By comparison, the Substance Registry Services (SRS), the List of Lists, and provisions related to Hazardous Air Pollutants ("HAPs") fail to distinguish between ethanol production methods.
- 49. The SRS is the EPA's central system for information about substances that are tracked or regulated by EPA or other sources. It is a resource for basic information about chemicals, biological organisms, and other substances of interest to EPA and its state and tribal partners.
- 50. The SRS makes it possible to identify which EPA data systems, environmental statutes, or other sources have information about a substance and which synonym is used by that system or statute. It becomes possible therefore to map substance data across EPA programs.
- 51. Similarly, the List of Lists is a consolidated list of chemicals subject to: Emergency Planning and Community Right-to-Know Act (EPCRA); Comprehensive Environmental

Response, Compensation and Liability Act (CERCLA); and Section 112(r) of the CAA. The List

of Lists was prepared to help facilities handling chemicals determine, for a specific chemical,

whether they may be subject to certain reporting requirements.

52. These lists are used as a reference tool, not as a definitive source of compliance

information. The industry does not use either the SRS or the List of Lists to categorize its

manufacturing processes and neither purports to categorize a specific method of production.

53. With respect to Hazardous Air Pollutants ("HAPs") EPA does not distinguish

between chemical and biological synthesis of fuel ethanol. If fuel ethanol production facilities

generate HAPs above certain thresholds, the facility will be subject to NESHAP regulations that

include broader definitions. Therefore, NESHAP regulations do not provide guidance regarding

the categorization of a specific method of production for ethanol products. Regardless, the Project

will not produce HAPs at levels to trigger NESHAP requirements.

54. Based on my knowledge of the Project, my technical expertise as an environmental

scientist, and my review of and professional experience with the Federal and state laws and

regulations referenced above, it is my expert opinion within a reasonable degree of scientific and

technical certainty that the Project does not propose "chemical manufacturing" or "chemical

refining" as those terms are understood within the industry or as those terms are applied by

applicable, environmental regulatory bodies.

55. A true and correct copy of my curriculum vitae is attached to this Affidavit as

Affidavit Attachment #6.

FURTHER AFFIANT SAYETH NAUGHT.

DAVID W. ROGERS

STATE OF FLORIDA

# COUNTY OF NASSAU

Sworn to or affirmed and signed before r	me by means of physical presence or
online notarization, this 18th day of December,	, 2024, by DAVID W. ROGERS, who is
personally known to me or produced	as identification and who
did take an oath.	
	Rhoude MBlake
	NOTARY PUBLIC, State of Florida
	NOTARY PUBLIC, State of Florida My Commission expires: 12/28/2026
	My Commission expires: 12/28/2020

Notary Public State of Florida Rhonda M. Blake My Commission HH 308483 Expires 12/28/2026





Reply to: Jacksonville

# **MEMORANDUM**

TO: Sarah Campbell, City Manager, City of Fernandina Beach, Florida

Tammi Bach, City Attorney, Fernandina Beach, Florida

Technical Review Committee Members CC:

Brenna Malouf Durden FROM:

Nicole J. Poot

**Amy Taylor Petrick** 

DATE: December 18, 2024

SUBJECT: RYAM – 2G-Bioethanol Project – Consistency with City's Comprehensive Plan

and Land Development Code

# EXECUTIVE SUMMARY

This Memorandum, written on behalf of RYAM, supplements RYAM's application for its proposed second-generation (2G)-Bioethanol project's ("the Project") site plan submitted to the City of Fernandina Beach (the "City") to demonstrate the Project's consistency with the City's Comprehensive Plan (the "Comprehensive Plan" or "Plan") and Land Development Code ("the LDC.") Rayonier Performance Fibers, LLC ("RYAM") owns and operates an acid-sulfite-based, pulp mill in the City that produces high purity cellulose products sold for commercial use (the "RPF Plant"). The Project, which will simply ferment the byproduct currently generated at the existing RPF Plant as necessary in order to enhance the Plant's energy efficiency, environmental sustainability, and economic viability, is consistent with the Plan and the LDC, and is not a prohibited use in the City as can be ascertained by the City's approval of breweries, bakeries, and the City's own water treatment facility that rely on and use the very same biological process. This memorandum explains the following:

- Application of the traditional rules of statutory construction to the specific facts of I. the Project requires finding the Project to be consistent with the Plan and the LDC.
  - The Project fits squarely within the definition of "Manufacturing and/or A. Assembly - Heavy," which includes "uses involving intensive manufacturing and industrial operations, including the manufacturing, assembly, fabrication, compounding, processing and/or treatment of extracted or raw materials or other industrial products; packaging and freight loading/unloading activities;

245 Riverside Ave. Suite 510 Jacksonville, FL 32202

T: 904.353.6410 F: 904.353.7619

#### ST. PETERSBURG

100 Second Ave. South Suite 501-S St. Petersburg, FL 33701

T: 727.245.0820 F: 727.290.4057

#### **TALLAHASSEE**

106 East College Ave. Suite 1500 Tallahassee, FL 32301 T: 850.222.5702

F: 850.224.9242

#### **TAMPA**

301 West Platt St. Suite A364 Tampa, FL 33606 T: 813.775.2331

#### **WEST PALM BEACH**

360 South Rosemary Ave. Suite 1100 West Palm Beach, FL 33401 T: 561.640.0820 F: 561.640.8202

- utilization, handling and bulk storage of materials including raw materials, chemicals and hazardous materials associated with manufacturing processes; and all other associated or ancillary activities." CITY OF FERNANDINA BEACH, FLA., LAND DEV. CODE § 1.07.00 (2023) (emphasis added).
- B. The Project constitutes an associated and ancillary activity that uses the spent sulfite liquor ("SSL"), a byproduct generated by the existing principal pulp manufacturing process, as the source of sugars for fermentation.
- C. Fermentation of sugars contained in the existing SSL stream is not "chemical manufacturing." Rather, it is a process that occurs in nature through the use of organisms a process that the City has approved in multiple locations throughout the City by permitting breweries, bakeries, and even the City's own water treatment plant, to operate within the City.
- D. Distilling and drying are physical separation processes, rather than "chemical refining," and are the same processes that have also been approved by the City by permitting the operation of distilleries.
- E. The Project is consistent with and furthers many goals and objectives of the City's Comprehensive Plan. These include numerous economic objectives, which are advanced by RYAM's continued economic contribution of more than \$379 million in 2023 in Nassau County alone and nearly \$575 million in the wider wood-procurement region, as well as the \$50 million economic investment represented by the Project itself.
- II. The legal opinion drafted by Weiss, Serota, Helfman, Cole & Bierman, dated May 23, 2024 (the "Weiss Serota Memo"), is fatally flawed in its analysis and is not binding on the City. It is not based on an application made to the City. It rests on an erroneous interpretation of an air permit application and draft construction air permit by persons without the expertise to understand the scientific and industrial context of a highly technical application to ensure compliance with the state of Florida's air regulations and the Federal Clean Air Act – neither of which govern administration of the City's Comprehensive Plan or LDC. Moreover, it failed to apply traditional rules of statutory construction to its interpretation of the phrases "chemical manufacturing" and "chemical refining." Consequently, its absolute conclusion that the Project is not consistent with the Comprehensive Plan is wrong. Finally, the City Manager cannot delegate her interpretive authority to a private law firm. Even if such authority existed, treating Weiss Serota's legal opinion as binding would violate RYAM's due process rights by denying RYAM's right to notice and opportunity to be heard on its application to the City.

## I. INTRODUCTION AND PROJECT DESCRIPTION

RYAM owns and operates an acid-sulfite-based, pulp mill in the City that produces high purity cellulose products sold for commercial use.

RYAM is proposing to install equipment for a second-generation bioethanol (2G-Bioethanol or "bioethanol")) project within the existing boundary of the RPF Plant. 2G-Bioethanol is produced from biomass – not primary food sources like corn. Because it is not a fossil fuel, it reduces greenhouse gas emissions. The Project presents a unique opportunity in the United States to use the sugars contained in the biomass RYAM already uses to make pulp by fermenting the existing SSL byproduct stream from its existing pulping operations.

The pulp mill's existing operation produces SSL as a byproduct. The use of this byproduct, as the Project proposes, fits within the descriptions of permissible uses in the Industrial land use category and the I-2 Heavy Industrial zoning district. The Project will ferment the sugars contained in the SSL to create the 2G-Bioethanol. The fermentation relied upon by the Project is a natural metabolic process using biologic organisms, usually micro-organisms found in yeast, in which sugar molecules are converted to substances like the lactic acid found in the human body, alcohols, such as beer and wine, and carbon dioxide. The Project also constitutes an associated and ancillary activity to the current principal pulp mill use, subordinate to the RPF Plant's efficiency, sustainability and economic viability and likewise consistent with the referenced Plan and LDC provisions.

In the existing RPF Plant process, the SSL byproduct is not fully utilized. Currently, after the SSL is separated from the pulping process, it is evaporated to concentrate the remaining biomass solids. From there, the SSL is either burned in the sulfite recovery boiler to generate energy and recover valuable raw materials that are reused in the pulping process or is transferred to LignoTech Florida, LLC (co-located at the RPF Plant site), which utilizes some of the SSL to produce wet and dry lignosulfonate products.

The Project itself consists of a variety of equipment to be installed on the RPF Plant property, which support fermentation and allow concentration of the 2G-Bioethanol through physical separation. This equipment includes continuous fermenters, centrifuges to allow yeast recycling, a distillation column for removing water and other liquid components from the ethanol, molecular sieves to further "dry" the ethanol by physically removing water molecules, and storage tanks. Importantly, the footprint of the RPF Plant will not be expanded by the Project.

The Project does not pose threats to environmental resources. It will be sited within the existing footprint of the RPF Plant, will be located outside of the floodplain, and will comply with and incorporate all applicable wetland buffers. It is designed to minimize water usage and wastewater generation, and will comply with the RPF Plant consumptive water use permit and wastewater discharge permit. The design will also include primary and secondary containment systems to prevent unintended discharges to the wastewater treatment system and to soil. Importantly, yeast used in the process will be recycled with centrifuges or neutralized to eliminate the possibility of yeast leaving the RPF Plant. The Project will require no change in hours of

operation or cause any changes in fumes, glare, noise or heat. Visual impacts will remain virtually unchanged because of the existing and substantial buffering and screening along the current RPF Plant site boundaries. The Project will add a few thousand square feet of impervious surface to the RPF Plant property, which is insignificant for a 660-acre parcel. In fact, overall, the Project's characteristics will actually enhance the environmental sensitivity of the Heavy Industrial ("I-2") zoned land with the surrounding land uses by reducing the overall air emissions from the RPF Plant.

The Project allows RYAM to use the RPF Plant property for its highest and best use and ensures the RPF Plant's continuing economic contribution to the City, which furthers two important Goals contained in the Comprehensive Plan, in addition to the many other ways in which the Project is consistent with the Comprehensive Plan. Moreover, the Project will also reduce emissions from the sulfite recovery boiler. Emissions are reduced because, once fermented, almost no sugars will remain in the SSL that is returned to the recovery boiler to be burned, nor will RYAM replace the de-sugared SSL with any other type of fuel, like natural gas. This is because, in order to meet market-driven specifications for 2G-Bioethanol and be considered "green fuel," implementation of the Project must be energy neutral to the overall RPF Plant operations, further ensuring the environmental benefit of the Project. The portion of the fermented SSL that will be sent to LignoTech Florida will continue to be sold and processed as it is now.

RYAM intends to invest \$50 million to establish the Project. This investment will increase the City's tax base and support the RPF Plant's long-term economic viability by protecting against economic downturns and ensuring the RPF Plant's sustainability. By growing and investing, RYAM is protecting hundreds of local jobs, the local economy, and the environment.

## II. CONSISTENCY WITH APPLICABLE PLAN AND LDC PROVISIONS

In Florida, comprehensive plans are often referred to as "constitutions," and future development orders issued by a local government must be consistent with the provisions of its comprehensive plan. *Nassau Cnty. v. Willis*, 41 So. 3d 270, 276 (Fla. 1st DCA 2010); *Citrus Cnty. v. Halls River Dev., Inc.*, 8 So. 3d 413, 420-21 (Fla. 5th DCA 2009). Fernandina Beach's LDC implements and is consistent with the City's Plan. *See* Sections 1.00.01 and 1.00.02, LDC.

As discussed more fully below, RYAM's application is consistent with and furthers the applicable Plan and LDC provisions, which are addressed in turn.

<sup>&</sup>lt;sup>1</sup> The City is not tasked with evaluating or permitting based on the air emissions from the Project. That regulatory authority is exclusively reserved for the Florida Department of Environmental Protection ("FDEP") acting on behalf of the federal Environmental Protection Agency ("EPA"). However, as explained herein, the City's expressed goals, policies, and objectives set forth in its Comprehensive Plan include reducing climate change, encouraging the production and use of energy generated from renewable resources, integrating green/sustainable development, and striving to meet air quality standards established by the EPA and FDEP. As such, the reduction in emissions from the Project provides additional support for a consistency determination in favor of the Project.

- A. The Industrial Land Use Category Description and the LDC Definition of Manufacturing and/or Assembly-Heavy.
  - 1. Policy 1.07.12 Industrial (IN), Future Land Use Plan.

The RPF Plant property has an Industrial (IN) Future Land Use Map ("FLUM") designation. The City's Comprehensive Plan Policy 1.07.12 sets forth the Plan's description for the "IN" land use category as follows:

Policy 1.07.12. Industrial (IN)

- a. The industrial land use category is intended to recognize existing industrial development, appropriate open air recreation activities and animal shelter, and to ensure the availability of land for industrial and airport purposes.
- b. The intensity of industrial development shall not exceed a FAR of. 0.75.
- c. Industrial sites should have transportation access by air, rail, or highway.
- d. Industrial uses include: airport dependent uses, manufacturing, assembling and distribution activities; warehousing and storage activities; green technologies, general commercial activities; integral airport related support services such as rental car facilities, parking facilities; and other similar land uses.

CITY OF FERNANDINA BEACH, FLA., 2030 COMPREHENSIVE PLAN, Goal 1, Objective 1.06, Pol'y 1.07.12 (emphasis added).

The general policy set forth above makes clear the City's intent to recognize existing industrial development and to ensure the availability of land for industrial purposes. It also expressly includes manufacturing, assembly and distribution activities, storage activities, green technologies, and other similar land uses. The Project clearly falls within this list of permissible authorized industrial uses. In addition, the Site Plan meets the FAR limitation of 0.75, and shows that the RPF Plant site has transportation access, as required by this Policy 1.07.12. See Proposed Site Plan, dated December 4, 2024. Thus, the Project is consistent with these portions of the Policy.

However, the Policy also contains the following prohibition:

g. Heavy metal fabrication, batch plants, salvage yards, **chemical** or petroleum **manufacturing or refining**, rubber or plastics manufacturing, or other uses generating potentially harmful environmental or nuisance impacts shall be prohibited.

CITY OF FERNANDINA BEACH, FLA., 2030 COMPREHENSIVE PLAN, Goal 1, Objective 1.06, Pol'y 1.07.12 (emphasis added).

Due to this limiting provision, the only interpretive issue for demonstrating consistency with the Industrial land use category description is whether the biological process being proposed constitutes "chemical . . . manufacturing or refining." "Chemical . . . manufacturing or refining" is not defined in the City's Comprehensive Plan.

# 2. LDC Definition of Manufacturing and/or Assembly-Heavy.

The RPF Plant property is zoned Heavy Industrial, I-2, as shown on the City's Zoning Map. In addition to the description of the IN land use category contained in the Plan, the definition of Manufacturing and/or Assembly-Heavy in the LDC (hereafter the "Heavy Manufacturing" definition) describes the numerous types of manufacturing uses and activities authorized in the Heavy Industrial I-2 zoning district. The term as defined means:

[U]ses involving intensive manufacturing and industrial operations, including the manufacturing, assembly, fabrication, compounding, processing and/or treatment of extracted or raw materials or other industrial products; packaging and freight loading/unloading activities; utilization, handling and bulk storage of materials including raw materials, chemicals and hazardous materials associated with manufacturing processes; and all other associated or ancillary activities.

CITY OF FERNANDINA BEACH, FLA., LAND DEV. CODE § 1.07.00 (2023) (emphasis added).

The Project fits squarely within the description of the types of uses and activities included in this definition. The Project proposes the processing and treatment of SSL, an extracted material, and the utilization, handling and bulk storage of chemicals associated with the pulp manufacturing process.

The definition further specifies that "all other associated or ancillary activities" are also included in the definition and authorized in the Heavy Industrial I-2 zoning district. Here, the Project again fits squarely within this phrase. The Project is an "associated or ancillary activity" to the existing and permitted pulp manufacturing use. The terms "associated" and "ancillary" are not defined in the LDC. When terms in laws or regulations are not defined, plain and ordinary meanings using dictionary definitions to determine the meaning of the terms is appropriate. See Roldan v. City of Hallandale Beach, 361 So. 3d 348, 354 (Fla. 4th DCA 2023) and statutory construction discussion at Section II.B. below. "Associated" means closely connected (as in function or office) with another; having secondary or subordinate status. Associated, MERRIAM-WEBSTER DICTIONARY, https://www.merriam-webster.com/dictionary/associated (last visited November 25, 2024). The term "ancillary" means having a subordinate, subsidiary or secondary MERRIAM-WEBSTER DICTIONARY, https://www.merriamnature. Ancillary, webster.com/dictionary/associated (last visited November 25, 2024). "Ancillary" is also defined as "providing necessary support to primary activities or operation of an organization, institution,

<sup>&</sup>lt;sup>2</sup> This memo does not address petroleum refining because the starting material for such refining operations would be a petroleum product such as crude oil. See How Refineries Work, AM. FUEL & PETROCHEMICAL MFRS., <a href="https://afpm.org/industries/operations/how-refineries-work">https://afpm.org/industries/operations/how-refineries-work</a> (last visited November 22, 2024). The Project does not involve petroleum products.

industry or system; the development of ancillary services to support its products." Ancillary, Oxford Learner's Dictionary <a href="https://www.oxfordlearnersdictionaries.com/us/definition/english/ancillary">https://www.oxfordlearnersdictionaries.com/us/definition/english/ancillary</a> (last visited November 25, 2024). The SSL is a byproduct of the existing manufacturing process. Using the SSL, as proposed by the Project, is closely connected and subordinate to the pulp manufacturing process because without the pulping operations, the sugars in the biomass would not be available to ferment. Given these plain and ordinary meanings, the Project constitutes both an associated and ancillary activity.

Similar to the description of the IN-Industrial land use category in the Plan, the Heavy Manufacturing definition also includes the following sentence:

Such use does not include heavy metal fabrication, batch plants, salvage yards, chemical or petroleum manufacturing or refining, rubber or plastics manufacturing, or other uses generating potentially harmful environmental or nuisance impacts.

CITY OF FERNANDINA BEACH, FLA., LAND DEV. CODE § 1.07.00 (2023).

Just as the case is with the Industrial land use category, the Project clearly fits within the portion of the LDC definition describing all of the permissible and allowable uses and activities included in the Heavy Manufacturing definition. Thus, the only remaining interpretive issue for demonstrating compliance with the Heavy Manufacturing definition as well as the Industrial land use description, is whether the Project falls within the "chemical . . . manufacturing or refining" prohibition. The LDC, like the Comprehensive Plan, contains no definition for "chemical manufacturing or refining."

# B. Pursuant to the Rules of Statutory Construction, the Project is Consistent with and Furthers Future Land Use Plan Policies, Goals and Objectives.

When the terms in a law or regulation are not defined, then the language of the regulation should usually be given its plain and ordinary meaning "unless the context indicates they bear a technical sense." Roldan at 354, quoting Antonin Scalia and Bryan A. Gardner, Reading Law: The Interpretation of Legal Texts 69 (2012); see also Fla. Birth-Related Neurological Injury Comp. Ass'n v. Fla. Div. of Admin. Hearings, 686 So. 2d 1349, 1354 (Fla. 1997); Holly v. Auld, 450 So. 2d 217, 219 (Fla. 1984) (quoting A.R. Douglass, Inc. v. McRainey, 102 Fla. 1141, 137 So. 157, 159 (1931) (..."[W]hen the language of the statute is clear and unambiguous and conveys a clear and definite meaning, there is no occasion for resorting to the rules of statutory interpretation and construction."). However, where a term is subject to different meanings and therefore ambiguous courts will resort to the rules of statutory construction. See e.g. Forsythe v. Longboat Key Beach Erosion Control Dist., 604 So. 2d 452, 455 (Fla. 1992); Katherine's Bay, LLC. v. Fagan, 52 So. 3d 19, 28 (Fla. 1st DCA 2010) (explaining that rules of statutory construction apply to comprehensive plans). In addition to the long-standing common law principle that comprehensive plans be read as a whole, section 163.3187(4), Florida Statutes, requires that comprehensive plans be internally consistent. Therefore, interpretations that create internal inconsistency within the Comprehensive Plan must be rejected, along with interpretations that are irrational or render words meaningless. Katherine's Bay, LLC., 52 So. 3d at 28. Where the exact meaning of a term is not defined in a statute itself, courts have concluded that terms could be defined by "industry custom." State v.

Brake, 796 So. 2d 522, 528 (Fla. 2001), citing State v. Fuchs, 769 So. 2d 1006, 1009 (Fla. 2000) and State v. Hagan, 387 So. 2d 943, 945-46 (Fla. 1980). Courts also recognize that "zoning regulations are in derogation of private rights of ownership" and therefore give "words used in a zoning ordinance" the meaning that offers the broadest range of uses for the property owner, under the principle that "when there is no definition or clear intent to the contrary, the ordinance should be interpreted in favor of the property owner." Rinker Materials Corp. v. City of N. Miami, 286 So. 2d 552, 553 (Fla.1973).

## 1. The Term "Chemical".

The modifier "chemical" in the phrase "chemical . . . manufacturing or refining" is ambiguous if for no other reason than the word "chemical" is often used both as an adjective, defining the nature of a process, or as a noun, defining the output of a process. The EPA determined that the phrase "chemical process plant" was not subject to a plain meaning interpretation, concluding:

We do not believe that the term "chemical process plant" is subject to a "plain meaning interpretation." There is not a universally accepted definition of chemical process, and accepted definitions differ depending on whether you view the term from a purely scientific sense or from an engineering sense, or for economic purposes.

Prevention of Significant Deterioration, Nonattainment New Source Review, and Title V: Treatment of Certain Ethanol Production Facilities Under the "Major Emitting Facility" Definition, 72 Fed. Reg. 24060, 24063 (May 1, 2007).

Since the phrase "chemical manufacturing or refining" is ambiguous, the language must be read consistent with the rules of statutory interpretation considering the context of the phrase's usage, both within the Comprehensive Plan as a whole and considering the technical and scientific background in which it applies. *Conage v. U. S.*, 346 So. 3d 594, 598 (Fla. 2022).

Both the Plan and the LDC clearly authorize various types of manufacturing in the Heavy Industrial I-2 zoning district, distinguishing between types by process. The definition of "Manufacturing and/or Assembly – Heavy" includes "uses involving intensive manufacturing and industrial operations, including the manufacturing, assembly, fabrication, compounding, processing and /or treatment of extracted or raw materials or other industrial products; packaging and freight loading/unloading activities; utilization, handling and bulk storage of materials including raw materials, chemicals and hazardous materials associated with manufacturing processes; and all other associated or ancillary activities." (emphasis added)

Likewise, Merriam-Webster offers the following definitions for the word "chemical," both of which focus on the involvement of either chemistry or chemicals in the *process*:

1: of, relating to, used in, or produced by <u>chemistry</u> or the phenomena of chemistry chemical reactions

2a: acting or operated *or produced by <u>chemicals</u>* a *chemical* fire extinguisher.

Chemical, MERRIAM-WEBSTER DICTIONARY, <a href="https://www.merriam-webster.com/dictionary/chemical">https://www.merriam-webster.com/dictionary/chemical</a> (emphasis added) (last visited Nov. 24, 2024); see Barco v. Sch. Bd. of Pinellas Cnty., 975 So. 2d 1116, 1122 (Fla. 2008) (referring to Merriam-Webster and holding "[i]t is appropriate to refer to dictionary definitions when construing statutes or rules.")

Thus, both the dictionary definition and the context of the Plan and LDC provisions call for the word "chemical" in the phrase "chemical manufacturing or refining" to refer to a manufacturing process that uses chemistry or chemicals in a chemical reaction process to create manufactured or refined items. As explained below, the Project does not use a chemical reaction process to create 2G-, but rather uses biological and physical processes and thus, the Project is not prohibited.

2. The Proposed Production of 2G-Bioethanol at the RPF Plant is a Natural Fermentation and Physical Separation Process that is not "Chemical Manufacturing" or "Chemical Refining".

RYAM proposes to implement a process that generally includes two stages: 1) fermentation, where sugars are converted into ethanol, and 2) distillation, where the ethanol is separated and purified.

# a. Fermentation is Not "Chemical Manufacturing".

The Project will produced bioethanol through the natural process of fermentation. Fermentation is a process that happens in nature. It relies on living organisms, typically microorganisms, to convert organic substrates into the desired product—in this case, ethanol for fuel. <sup>3</sup> Industrial fermentation follows this same natural process and is often referred to as "bioprocessing" or "biotechnology" rather than chemical manufacturing.<sup>4</sup>

Fermentation is a well-known natural process used by humanity for thousands of years to make beer, liquor, bread, yogurt, cheese, and other similar products.<sup>5</sup> Local distiller Roger Morenc

<sup>&</sup>lt;sup>3</sup> Industrial Fermentation Supporting a Growing Industry, *Institute for Advanced Learning and Research*, (Aug. 20, 2024), <a href="https://www.ialr.org/industrial-fermentation-supporting-a-growing-industry/#:~:text=At%20its%20basic%20level%2C%20fermentation,then%20harvested%2C%20packaged%20and%20sold.">https://www.ialr.org/industrial-fermentation-supporting-a-growing-industry/#:~:text=At%20its%20basic%20level%2C%20fermentation,then%20harvested%2C%20packaged%20and%20sold.</a>

<sup>&</sup>lt;sup>4</sup> Id.; see also Christopher J. Hewitt et al., The Scale-Up of Microbial Batch and FedBatch Fermentation Processes, 62 ADVANCES IN APPLIED MICROBIOLOGY 105-135 (2007) (article relied on by Medardo Monzon in forming his opinions stated publicly).

<sup>&</sup>lt;sup>5</sup> Sergi Maicas, *The Role of Yeasts in Fermentation Processes*, NATIONAL LIBRARY OF MEDICINE (2020), <a href="https://pmc.ncbi.nlm.nih.gov/articles/PMC7466055/">https://pmc.ncbi.nlm.nih.gov/articles/PMC7466055/</a>; see also Memorandum from Glen P. Fox, Anheuser-Busch Endowed Professor in Department of Food Science & Technology at University of California, on Ethanol Production from Natural Sources of Sugar (Oct. 10, 2024) (Attached hereto and made part hereof as Exhibit 1).

confirms this, stating, "[t]his is a natural biological process and actually happens in the wild in a large number of instances." Specifically for the Project, sugars contained in the SSL will be converted by yeast to ethanol.<sup>7</sup>

An article relied on by Medardo Monzon in developing his publicly stated opinions (in opposition to the Project) confirms the biologic nature of the sugar conversion process. The article explains, "[e]thanol can be made synthetically from petroleum or by microbial conversion of biomass materials through fermentation." Notably, this reference acknowledges there are two distinct ways to make ethanol – one a synthetic chemical manufacturing process and one a fermentation process. This article further identifies the three steps for a natural fermentation process: "The fermentation method generally uses three steps: (1) the formation of a solution of fermentable sugars, (2) the fermentation of these sugars to ethanol, and (3) the separation and purification of the ethanol, usually by distillation." For the Project, step one already takes place in the RPF Plant's current existing pulping process when the SSL is created. Finally, this article describes fermentation as "involv[ing] microorganisms that use the fermentable sugars for food and in the process produces ethanol and other byproducts."

By contrast, chemical manufacturing refers to the industrial production of chemicals through chemical reactions involving non-living substances. These processes often include synthesis, catalysis, and other chemical transformations of raw materials such as petroleum, natural gas, or minerals. In fact, the Weiss Serota Memo, which will be discussed more fully below, compares biochemical fermentation to a thermochemical conversion process, which it describes as "involving adding heat *and chemicals* to a biomass feedstock to produce syngas, which is a mixture of carbon monoxide and hydrogen...[which is then] mixed with a catalyst and reformed into Ethanol and other liquid coproducts." Weiss Serota Memo, p. 4 (emphasis added). The difference between the two processes, and one of the failings of the Weiss Serota Memo's flawed logic, is clear: on the one hand, the Project uses biological agents in the manufacturing process in the form of yeast while the thermochemical manufacturing process uses chemicals and non-renewable raw materials.

There is a clear distinction between a biological process like fermentation and chemical manufacturing. To conclude otherwise would mean that the production of beer, liquor, yogurt, bread, and even the operation at the City's water treatment plant, which uses living organisms to breakdown waste, would be prohibited in the City. Since the City expressly allows fermentation

<sup>&</sup>lt;sup>6</sup> Memorandum from Roger Morenc, Owner of Marlin & Barrel Distillery (Oct. 15, 2024) (Attached hereto and made part hereof as Exhibit 2).

<sup>&</sup>lt;sup>7</sup> Memorandum from Glen P. Fox, *supra* note 5.

<sup>&</sup>lt;sup>8</sup> P.C. Badger, *Ethanol from Cellulose: A General Review*, reprinted from TRENDS IN NEW CROPS AND NEW USES (J. Janick and A. Whipkey eds., 2002).

<sup>&</sup>lt;sup>9</sup> *Id.* at 17.

<sup>&</sup>lt;sup>10</sup> *Id*.

<sup>11</sup> Id.

<sup>&</sup>lt;sup>12</sup> *Id.* (stating that ethanol can be made synthetically).

within its boundaries in the same Industrial future land use designated area and Heavy Industrial I-2 zoning district as the RPF Plant, as shown in Table 2.03.02, Table of Land Uses in the City's LDC, the phrase "chemical manufacturing" cannot be interpreted as encompassing fermentation and remain internally consistent. As noted above, Florida Statutes requires both that comprehensive plans be internally consistent, and that zoning regulations be consistent with comprehensive plans. § 163.3187(4), Fla. Stat. (2023); § 163.3194, Fla. Stat. (2002). Therefore, the express allowance of breweries and distilleries, which create alcohol through a fermentation process, means that the Project must likewise be allowed since it also creates alcohol through a fermentation process. *Fla. Dept. of Env't. Prot. v. ContractPoint Fla. Parks, LLC*, 986 So. 2d 1260, 1270 (Fla. 2008) (A basic rule of statutory construction requires the court to avoid a literal interpretation that would result in an absurd result, ridiculous conclusion or untenable consequence.)

## b. Drying and Distillation is Not "Chemical Refining".

Just as chemical manufacturing does not encompass the production of alcohol through fermentation, the phrase "chemical refining" does not encompass the processes of "drying and distillation."

Distillation is a physical separation process based on differences in boiling points of components in a mixture. It involves heating the mixture to convert the more volatile component(s) into vapor, which is then condensed back into a liquid and collected separately. The process of distillation typically involves a distillation column or still, where the mixture is heated, and the vapor is collected and condensed.<sup>13</sup> No new chemical is introduced, nor is any new chemical produced. Substances are simply separated from one another. The Project would produce 2G-Bioethanol in a manner identical to the way Marlin & Barrel makes liquor.

Roger Morenc owner of Marlin & Barrel describes distilling as follows:

Once all of the potential alcohol is made in a ferment then its [sic] time to take the part you want and discard the rest. Since ferments are a natural process [sic] you'll find that a small amount of other-than ethyl alcohol can be present. Said another way, even though the vast majority of alcohol made is ethyl alcohol, there will be other types present. Each will have its unique molecular weight and that directly corresponds to a unique boiling point. As the ferment is distilled, the liquid is heated and the lighter compounds that require less heat to change from a liquid to vapor state will climb the sill [sic] column first and further then those needing more energy to get to their vapor form. In this way a still organizes all of the different molecule types present in a finished ferment. From that position of stratification a distiller can extract the desired piece of the ferment, cool it to liquid temperature again and then collect it. In no part of this process are chemicals being changes [sic]. This is simply a liquid sifter of sorts.

<sup>&</sup>lt;sup>13</sup> Distillation, BRITANNICA, <a href="https://www.britannica.com/science/distillation">https://www.britannica.com/science/distillation</a> (last updated Oct. 24, 2024).

Memorandum from Roger Morenc, Owner of Marlin & Barrel Distillery (Oct. 15, 2024) (Exhibit 2, attached) (emphasis added).

Similarly, drying is another way to separate molecules from one another based on size. The Project anticipates using molecular sieves to separate water and ethanol molecules from one another. The molecular sieve will trap water molecules in a bed of adsorbent beads with small pores that small water molecules can pass through and be adsorbed, thereby allowing the ethanol molecules to pass through. This allows water content to be removed without introducing any new chemical or a new process, and no new chemical is produced. Pre-existing substances are simply separated from one another.

Conversely, "chemical refining" typically involves chemical reactions to remove impurities or unwanted components from a substance. It may involve adding chemical agents that react with specific impurities, turning them into removable substances. Chemical refining is also commonly used in the purification of oils (like vegetable oils) or metals (such as the refining of crude metals into purer forms). <sup>14</sup> For example, in vegetable oil refining, caustic soda is added to the mixture to neutralize free fatty acids. <sup>15</sup> In metal refining, chemicals are added to remove impurities like sulfur or phosphorus.

The critical distinction between the two processes is that distillation and drying are physical processes, relying on temperature differences and mechanical separation, while chemical refining is a process that involves the use of chemicals to cause reactions. And most important, the City has approved distillation of ethanol intended for drinking within its City limits. As confirmed by its owner, distillation operations like those Marlin and Barrel performs are the same as the distillation operation that is proposed by the Project. Thus, the City cannot define the processes proposed by the Project as "chemical refining," while maintaining internal consistency within the Plan and between the Plan and the LDC, as well.

<sup>&</sup>lt;sup>14</sup> Chemical Refining, BRITANNICA, <a href="https://www.britannica.com/technology/chemical-refining">https://www.britannica.com/technology/chemical-refining</a> (last visited Nov. 22, 2024); Chemical Refining, FEDIOL,

https://www.fediol.eu/web/chemical%20refining/1011306087/list1187970098/f1.html (last visited Nov. 22, 2024); Fractional Distillation vs. Refining, THIS VS. THAT, https://thisvsthat.io/fractional-distillation-vs-refining (last visited Nov. 26, 2024); Said Gharby, Refining Vegetable Oils: Chemical and Physical Refining, NAT'L LIBR. MED. (Jan. 11, 2022), https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8767382/#:~:text=However%2C%20the%20chemical%20refining%20has,the%20release%20of%20polluting%20effluents.

<sup>&</sup>lt;sup>15</sup> Said Gharby, Refining Vegetable Oils: Chemical and Physical Refining, NAT'L LIBR. MED. (Jan. 11, 2022),

https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8767382/#:~:text=However%2C%20the%20chemical%20refining%20has,the%20release%20of%20polluting%20effluents.

3. The Weiss Serta Memo Incorrectly Interprets Chemical Manufacturing and Chemical Refining Based on Flawed Reasoning, Insufficient Data, and a Lack of Understanding Regarding the Science and the Industry.

On May 23, 2024, the City received the Weiss Serota Memo that considers whether the production of bioethanol at the RPF Plant would violate the prohibition against chemical manufacturing and refining found in the description of the Industrial land use category (Policy 1.07.12, Land Use Element) and the definition of "Heavy Manufacturing" in the LDC. It cannot be stressed enough that there was no application pending before the City when the Weiss Serota opinion was requested. The request was made because of an application submitted by RYAM to the FDEP.

The Weiss Serota Memo, authored by attorneys, without apparent input from scientific or technical consultants, was based on the attorneys' flawed understanding of information gleaned from an air permit application submitted to FDEP by RYAM, and on a draft construction air permit issued by FDEP -- both of which have never been submitted to the City nor made a part of any application to the City by RYAM. These documents contained only the information necessary for FDEP to conduct a highly technical "prevention of significant deterioration" ("PSD") applicability review and evaluate whether and under what conditions a construction air permit could properly be issued under the Federal Clean Air Act ("CAA") and applicable Florida law.

Importantly, the FDEP documents did not contain information required by the City when submitting a site plan for approval and did not address how words used in the City's Plan, a regulatory document created for an entirely different purpose, apply to the Project. Nevertheless, the Weiss Serota Memo improperly attempted to correlate these extra-record documents produced in an entirely different context to the question of whether the Project is consistent with the Comprehensive Plan and LDC with insufficient information about the Project, with limited or no expertise in the industry or in the areas of air permitting and critically PSD applicability review, and with little to no analysis of the Comprehensive Plan as a whole. As a result, the conclusions in the Weiss Serota Memo are legally and factually unsupported and do not represent competent, substantial evidence upon which the City could deny the Project.

While the Weiss Serota Memo correctly identifies that the Project is in an area zoned "Heavy Industrial (I-2)" and that chemical manufacturing and refining are not permitted uses under this zoning category, it does not accurately analyze whether the specific processes involved in the Project, namely fermentation and distillation, fit within the subject prohibitions.

Instead, Weiss Serota's Memo summarily interprets the word "chemical" as a noun in isolation, with no apparent cross-reference to other uses permitted in the I-2 district, or the remaining language in the Plan as a whole, and concludes that any operation that results in a chemical being produced is prohibited chemical manufacturing and refining. As a result, the interpretation advanced by the Weiss Serota Memo would include within the category of prohibited manufacturing virtually all substances. For example, water (H<sub>2</sub>O) can be considered a chemical.

Rather than analyzing the phrase "chemical manufacturing" in context, the Weiss Serota Memo focuses on the end product "ethanol" and simply asserts that because ethanol is a chemical, its production automatically qualifies the Project as one that constitutes prohibited "chemical manufacturing or refining". This oversimplification neglects to differentiate between different types of processing operations to create chemicals and ignores the fact that production of alcohol is already expressly allowed and permitted under the same Plan and LDC provisions through the same processes proposed by the Project. This narrow view also leads to results that are inconsistent with the City's existing application of the Plan and ignores well-established cannons of statutory construction.

Where the Weiss Serota Memo *does* attempt to consider industry regulation for context, it gets it demonstrably wrong. The Weiss Serota Memo attempts to support its argument that the Project constitutes "chemical manufacturing" by noting references to ethanol in the Toxic Substances Control Act ("TSCA"), ethanol being listed in EPA's Substance Registry Services ("SRS") online web database, ethanol being listed in EPA's Consolidated List of Chemicals, and the existence of a NAICS code, (NAICS 325-193), for "Ethyl Alcohol Manufacturing." As described herein, none of these bases serve to establish what is and is not "chemical manufacturing and refining" under the Plan, which functions in the land use, rather than in any federal regulatory protection context. However, the Weiss Serota Memo fails to recognize that, even in the regulatory context it relies on, the EPA has determined that the alcohol made by and the production processes used by breweries and distilleries is functionally equivalent to the production of bioethanol.

Specifically, while purportedly relying on the information contained in the air permit application, the Weiss Serota Memo noticeably omits from its analysis guidance documents from EPA that distinguish ethanol production by fermentation from chemical manufacturing. EPA's guidance is reviewed below.

The RPF Plant and the Project are subject to regulation by the EPA and FDEP under the Clean Air Act. The CAA is the comprehensive federal law that regulates air emissions from stationary sources such as the RPF Plant. The main goal of the CAA is to protect the public health and the environment by regulating emissions of certain air pollutants. The CAA is implemented through an extensive list of federal and state administrative regulations. Various regulations implementing the CAA include New Source Performance Standards "NSPS", "Prevention of Significant Deterioration" requirements, and National Emission Standards for Hazardous Air Pollutants "NESHAPS". The specific CAA requirements for the Project are set forth in RYAM's permit application submitted in 2023 to FDEP.

Generally, the EPA differentiates facilities that manufacture ethanol through chemical synthesis from those that use biological synthesis. For example, the EPA has determined that facilities that produce ethanol through fermentation are *not considered* synthetic organic chemical manufacturing industry (SOCMI) sources subject to regulation under applicable portions of 40 Code Federal Regulations Part 60. Pursuant to a series of Applicability Determinations by the EPA analyzing the applicability of various provisions of 40 Code Federal Regulations Part 60 to ethanol manufacturing facilities, the EPA determined that although ethanol is listed as a chemical affected by both "NSPS" Subparts RRR and NNN, background documentation created during the

development of both standards indicated that the creation of ethanol by fermentation should properly be excluded from the scope of both NSPS, Subparts RRR and NNN.

Consistent with this interpretation, the EPA specifically excluded ethanol production by fermentation from its definition of "chemical process plants." For example:

## 40 CFR § 51.165 provides:

Permit requirements.

- (a) \* \* \*
- (1)\*\*\*
- (iv) \* \* \*
- (C) \* \* \*
- (20) Chemical process plants—The term chemical processing plant shall not include ethanol production facilities that produce ethanol by natural fermentation included in NAICS codes 325193 or 312140;
- \* \* \* \* \*
- (4) \* \* \*
- (xx) Chemical process plants—The term chemical processing plant shall not include ethanol production facilities that produce ethanol by natural fermentation included in NAICS codes 325193 or 312140.

### 40 CFR § 51.165 (emphasis added).

The EPA expressed that one of its policy rationales for adopting the exclusion was to correct the disparate treatment related to the production of ethanol for fuel versus the production of ethanol intended for human consumption by applying two different major source thresholds. Because the two manufacturing processes are substantially similar, EPA stated that the process should be treated and regulated identically for purposes of the PSD and Title V regulations under the CAA, regardless of the intended product use. It is also clear from EPA's discussion of NAICS code 325193 in the Federal Register, that it considers ethanol production distinct from the broader classification of chemical manufacturing in NAICS code 325. <sup>16</sup>

<sup>&</sup>lt;sup>16</sup>Prevention of Significant Deterioration, Nonattainment New Source Review, and Title V: Treatment of Certain Ethanol Production Facilities Under the "Major Emitting Facility" Definition, 72 Fed. Reg. 24060, 24062 (May 1, 2007) ("Ethanol fuel and industrial ethanol fall within NAICS 325193 (Ethyl Alcohol Manufacturing) which includes denatured alcohol, nonpotable ethanol, and nonpotable grain alcohol. NAICS 312140 (Distilleries) includes potable ethyl alcohol and grain alcohol beverages. Even though NAICS 325193 (ethyl alcohol manufacturing) has been classified under NAICS Chemical Manufacturing subsector, unlike under the SIC classification of 2869 (Industrial Organic Chemicals, Not Elsewhere Classified), ethyl alcohol manufacturing is within its own narrowly defined category.")

Consistent with EPA's "chemical process plant" exclusion, FDEP, which acts as the statewide enforcer of the EPA's regulatory framework under the CAA and has authority to issue CAA permits in Florida on behalf of the EPA, has adopted regulations in the Florida Administrative Code that mirror the Code of Federal Regulations. For example, in Rule 62-210.200(156) of the Florida Administrative Code the definition of "Major Stationary Source" expressly excludes ethanol production facilities that produce ethanol by natural fermentation:

- (a) A major stationary source is:
- 1..... chemical process plants (the term "chemical process plants" shall not include ethanol production facilities that produce ethanol by natural fermentation included in North American Industry Classification System (NAICS) codes 325193 or 312140)....
- (c) The fugitive emissions of a stationary source shall not be included in determining for any of the purposes of this definition whether it is a major stationary source, unless the source belongs to one of the following categories of stationary sources: ....
- 20. Chemical process plants (the term "chemical process plants" shall not include ethanol production facilities that produce ethanol by natural fermentation included in North American Industry Classification System (NAICS) codes 325193 or 312140)

Fla. Admin. Code R. 62-210.200(156) (2020) (emphasis added).

The National Resource Defense Council ("NRDC") challenged a similarly adopted exclusion in the State of Indiana. Specifically, the Supreme Court of Indiana determined that its state environmental agency, which adopted a mirror definition of "chemical process plant" to exclude fuel ethanol plants for air permitting purposes was reasonable as a matter of regulatory interpretation. *Nat'l Res. Def. Council v. Poet Biorefining-N. Manchester*, LLC, 15 N.E. 3d 555, 565 (Ind. 2014). In that case, the NRDC argued that fuel ethanol plants should be classified as "chemical process plants" based on a plain reading of the term. However, the Court rejected this interpretation. The Court noted that prior to the EPA's revision to its definition of chemical process plant, ethanol production by fermentation for human consumption was treated differently than ethanol production for fuel, despite both involving the same biological processes. Moreover, the Court found that fermentation, a process used in ethanol production, is common across industries, many of which are not classified as "chemical process plants."

The Court emphasized that the NRDC did not justify why its interpretation of "chemical process plant" should apply only to fuel ethanol plants and not to food-grade ethanol production or other industries using fermentation. As such, the Court held that such an interpretation would improperly broaden the definition and stated "[w]e will not interpret a regulatory phrase in a way that both produces absurd results and vitiates other regulatory provisions for the sake of strictly applying the "plain meaning" canon of regulatory interpretation. *Id.* at 565. We give words their common and ordinary meaning without unduly emphasizing a strict literal or selective reading of the individual words." *Id.* The Court concluded that the Indiana environmental agency's exclusion of fuel ethanol plants from the "chemical process plant" category was reasonable, as it was consistent with EPA's stance and did not conflict with the regulatory framework.

Thus, the EPA's application of the Clean Air Act and judicial review of those decisions support RYAM's position (and common sense) here: the Project is like brewing beer, distilling alcohol for human consumption, or even making yoghurt or bread, not "chemical manufacturing" as that term is used by the industry or its environmental regulators. The Weiss Serota Memo's failure to evaluate or even discuss the CAA when the EPA and FDEP have clearly offered the above guidance further indicates that the Weiss Serota Memo is incomplete, based on incompetent evidence and analysis, and should be disregarded by the City in making its consistency determination.

Further, the Weiss Serota Memo selectively analyzes other Federal and state laws and regulations and ignores any of the references identified below to the various exclusions found throughout EPA's and FDEPs regulations for facilities producing ethanol by natural fermentation like the Project. Moreover, none of the regulatory provisions cited in the Weiss Serota Memo define or take into account the process of "chemical manufacturing." For example, TSCA establishes certain reporting, testing and recordkeeping requirements. The main goal of the TSCA is to ensure that chemicals used in commerce do not pose an unreasonable risk to human health or the environment. TSCA does not define any actual manufacturing processes. Nonetheless, the Weiss Serota Memo concludes that because ethanol falls within TSCA's broad definition of "chemical substance," its production must constitute "chemical manufacturing," as that term is used in an entirely different regulatory context in a different jurisdiction and under a document with no express reference to the TSCA or its purposes.

Similarly, the Weiss Serota Memo relies on NAICS definitions without considering that NAICS are used by federal agencies for statistical purposes only. While they may help identify applicable regulations, they are not determinative of what constitutes "chemical manufacturing or refining" in any context much less the land use regulations of a local government in Florida. Furthermore, there is no requirement that a local government must utilize NAICS, nor is there any indication that the City considered the NAICS code in adopting the Heavy Manufacturing definition included in the LDC.

<sup>&</sup>lt;sup>17</sup> See e.g., Learn About the Toxic Substances Control Act (TSCA), U.S. ENV'T PROT. AG., <a href="https://www.epa.gov/assessing-and-managing-chemicals-under-tsca/learn-about-toxic-substances-control-act-tsca">https://www.epa.gov/assessing-and-managing-chemicals-under-tsca/learn-about-toxic-substances-control-act-tsca</a> (last updated Oct. 4, 2024).

Additionally, while the EPA's Substance Registry Services ("SRS")<sup>18</sup> and the Consolidated List of Chemicals ("CLC")<sup>19</sup> cited to in the Weiss Serota Memo, may be useful tools for identifying regulated substances and tracking chemical inventories, their listing of ethanol does not automatically determine that the production of ethanol through fermentation is classified as "chemical manufacturing." The SRS and CLC identify substances that are regulated under various environmental statutes, such as the CAA and TSCA. While ethanol may appear on these lists, this is primarily for regulatory tracking, safety, or reporting purposes. The presence of a substance in these databases does not automatically dictate how the process of producing that substance is categorized -- especially by the industry. These databases ensure that producers are aware of regulations related to the handling, reporting, and usage of substances, but they do not define how

Substance Registry Services (SRS) is the Environmental Protection Agency's (EPA) central system for information about substances that are tracked or regulated by EPA or other sources. It is the authoritative resource for basic information about chemicals, biological organisms, and other substances of interest to EPA and its state and tribal partners.

The SRS makes it possible to identify which EPA data systems, environmental statutes, or other sources have information about a substance and which synonym is used by that system or statute. It becomes possible therefore to map substance data across EPA programs regardless of synonym.

The system provides a common basis for identification of, and information about:

- Chemicals
- Biological organisms
- Physical properties
- Miscellaneous objects

About Substance Registry Services (SRS), U.S. ENV'T PROT. AG., <a href="https://cdxapps.epa.gov/oms-substance-registry-services/about-srs">https://cdxapps.epa.gov/oms-substance-registry-services/about-srs</a> (last visited Nov. 26, 2024).

<sup>19</sup>The United States Environmental Protection Agency explains the list as follows:

The List of Lists is a consolidated list of chemicals subject to:

- Emergency Planning and Community Right-to-Know Act (EPCRA);
- Comprehensive Environmental Response, Compensation and Liability Act (CERCLA); and
- Section 112(r) of the Clean Air Act (CAA).

It was prepared to help facilities handling chemicals determine, for a specific chemical, whether they may be subject to certain reporting requirements. These lists should be used as a reference tool, not as a definitive source of compliance information.

Consolidated List of Lists, U.S. ENV'T PROT. AG., <a href="https://www.epa.gov/epcra/consolidated-list-lists">https://www.epa.gov/epcra/consolidated-list-lists</a> (last updated Nov. 12, 2024).

<sup>&</sup>lt;sup>18</sup>The United States Environmental Protection Agency describes the SRS as follows:

the process of producing said substance is categorized. As explained above, this distinction matters when evaluating consistency with the Comprehensive Plan and was not something considered by the Weiss Serota Memo.

This distinction between the processes is further emphasized by examining EPA's website, which includes an information page on "chemical manufacturing" in the context of its Toxic Release Inventory ("TRI") Program and how TRI chemical wastes are managed in the chemical manufacturing sector (defined as facilities reporting their primary NAICS code as 325).<sup>20</sup> The page includes a "Chemical Manufacturing Facilities by Subsector" pie chart that provides a summary of operations in the chemical manufacturing sector. According to EPA, operations in the chemical manufacturing sector include:

- o Basic chemicals facilities produce large quantities of chemicals that are often used to make other chemicals or products. Basic chemicals include petrochemicals, industrial gases, and synthetic dyes and pigments.
- Coatings and adhesives facilities mix pigments, solvents, and binders into architectural and industrial paints; manufacture paint products such as paint removers and thinners; and manufacture adhesives, glues, and caulking compounds.
- o Resins and synthetic rubber facilities manufacture resins, plastic materials, synthetic rubber, and fibers and filaments.
- Facilities in the "Other Chemical Products" subsector make chemicals for a
  wide variety of applications. These include chemicals used in photography,
  explosives, inks and toners, and transportation equipment like antifreeze or
  brake fluid.

Chemical Manufacturing, U.S. ENV'T PROT. AG., <a href="https://www.epa.gov/trinationalanalysis/chemical-manufacturing">https://www.epa.gov/trinationalanalysis/chemical-manufacturing</a> (last updated Aug. 20, 2024).

Notably absent from its descriptions are facilities that manufacture ethanol through a biological process.

Similarly, another glaring issue with the Weiss Serota Memo is its conclusion (without citation) that distillation is equivalent to "chemical refining." As explained above, these processes are distinct and have very distinct technical meanings. Thus, simply stating refining and distilling are the same is not a supportable position.

# 4. The Weiss Serota Memo Fails to Analyze the Comprehensive Plan as a Whole.

The Weiss Serota Memo also fails to analyze the Plan holistically, as required by Florida law. For example, the Weiss Serota Memo does not recognize that the prohibition against "chemical manufacturing or refining" is aimed at preventing the types of environmental and public safety hazards posed by traditional, stand-alone chemical and refining plants rather than existing

<sup>&</sup>lt;sup>20</sup> Chemical Manufacturing, U.S. ENV'T PROT. AG., <a href="https://www.epa.gov/trinationalanalysis/chemical-manufacturing">https://www.epa.gov/trinationalanalysis/chemical-manufacturing</a> (last updated Aug. 20, 2024).

facilities wishing to augment their existing production process with renewable energy production as a means to utilize its byproducts in a more environmentally sensitive manner. This point is supported by the following language that was included as part of the prohibition language when it was initially adopted by the City in October, 2000, and describes characteristics of the targeted uses that will be avoided as a result of the prohibitions:

"These uses typically generate heavy truck traffic, require significant acreage, are difficult to screen and buffer from residential areas, and therefore should be located in more sparsely developed unincorporated areas."<sup>21</sup>

Even though this language has since been deleted from the current version of Policy 1.07.12 describing the Industrial future land use category, it nevertheless serves as a clear indication of the original intent and rationale behind the prohibitions. As the Florida Supreme Court in *Ham v. Portfolio Recovery Assocs, LLC*, 308 So. 3d 942, 947 (Fla. 2020) stated, "[w]e thus recognize that the goal of interpretation is to arrive at a 'fair reading' of the text by 'determining the application of [the] text to given facts on the basis of how a reasonable reader, fully competent in the language, would have understood the text at the time it was issued." quoting *Scalia & Gardner, Reading Law* at 33. The Project has none of the characteristics of the uses that were the original drafter's target. Rather, the Project will generate only three truck trips per day, will require only about 2.5 acres located within the current RFP Plant property, and will be easily screened and buffered by trees and other natural buffers that already exist.

In addition, the Weiss Serota Memo does not consider the broader goals and objectives of the Comprehensive Plan. Principles of statutory construction demand that consideration be afforded "not only to the literal and usual meaning of the words, but also to their meaning and effect on the objectives and purposes of the statute's enactment." Florida Birth-Related Neurological Inj. Compen. Assoc. v. Fla. Div. of Admin. Hrg., 686 So. 2d 1349, 1354 (Fla. 1997). In this case, when viewed in its entirety, the Plan's emphasis on sustainability, economic growth, and industrial development actually supports, rather than conflicts with, approval of the Project.

# 5. The Project is Consistent with and Furthers Future Land Use Provisions Related to Safety.

For example, the Project supports Goal 1 of the Future Land Use Element, which encourages land uses that maintain the City's economic base while minimizing threats to health, safety, welfare, and environmental resources. The Project will contribute to a diverse economic base by introducing renewable energy production, a key industry that supports job creation and economic growth without compromising public safety or natural resources. The Project's focus on sustainability also furthers the Goal's stated intent to protect environmental resources.<sup>22</sup>

The city shall encourage and accommodate land uses which maintain the city as a viable community, enhance the city's economic base, and offer diverse opportunities for a wide

<sup>&</sup>lt;sup>21</sup>City of Fernandina Beach, Fla., Ordinance 99 – 49, (Oct. 17, 2000).

<sup>&</sup>lt;sup>22</sup> GOAL 1 of the City's Comprehensive Plan provides the following:

With respect to the second part of Goal 1, the Project is engineered to comply with rigorous safety and environmental protocols and regulations to protect health, safety, welfare, and environmental resources. The safety of RYAM's employees and the community is RYAM's highest priority. From a safety perspective, the primary concern of the Project is the flammability of ethanol, a fact of life whether you are brewing beer or making ethanol for fuel. The engineering design of the Project was developed in collaboration with local fire officials and the Company's insurance provider and includes dedicated fire prevention technologies and fire suppression systems that adhere to the latest federal, state, and local codes, standards, and regulations. As described in detail within the application, the fire safety systems will include concrete containments walls around fermentation, distillation and storage, floating roof tanks, process and ambient monitoring, control system interlocks and alarm systems, fire suppression systems using non-PFAS alcohol foams, and firefighting equipment.

The location of the process and storage tanks within the footprint of the RPF Plant is also an important consideration for risk minimization. The proposed location is a safe distance from the pulping operations and related chemical storage, and a safe distance from the community around the facility. RYAM's expert safety consultant conducted an independent evaluation of the potential impacts of a worst-case scenario event. This scenario assumes that none of the safety systems discussed above are in place. The study concluded that even in that highly unlikely scenario, the area of highest risk extends only 25 meters (0.0155 mile or 82 feet) from the storage tanks and the area of minor risk, such as broken windows, extends only 120 meters (0.0745 mile or 393.7 feet). Both distances are well within the boundaries of the current RPF Plant footprint and well away from the pulping operations and chemical storage.

Notwithstanding the actual impact radius for the worst-case event, the 2024 Emergency Response Guidebook<sup>23</sup> recommends an 800-meter (0.497 mile or 2,624.7 feet) evacuation radius for incidents involving flammable liquids. For the proposed process, that evacuation radius would not reach any residences or other private properties. It should be noted that, by contrast, a gasoline fire at the gas stations located on State Road 200 and on 8<sup>th</sup> Street are also subject to an 800-meter evacuation radius, and an incident at one of these *would* impact numerous residences and businesses in the community.

Finally, the Project will not create nuisances for the community. There will be no increase in raw materials usage. Finished product shipments will require up to three trucks per day. In recent years, the existing RPF Plant has reduced truck traffic by about six trucks per day by reducing the use of fuel oil at the facility.

variety of living, working, shopping, and leisure activities, while minimizing any threats to health, safety and welfare posed by hazards, nuisances, incompatible land uses and without adverse impact on its natural environment and cultural resources.

CITY OF FERNANDINA BEACH, FLA., 2030 COMPREHENSIVE PLAN, Goal 1, Objective 1.02. <sup>23</sup> U.S. Department of Transportation, Pipeline and Hazardous Materials Safety Administration, 2024 Emergency Response Guidebook, <a href="https://www.phmsa.dot.gov/sites/phmsa.dot.gov/files/2024-04/ERG2024-Eng-Web-a.pdf">https://www.phmsa.dot.gov/sites/phmsa.dot.gov/files/2024-04/ERG2024-Eng-Web-a.pdf</a> (last visited December 3, 2024).

# C. The Project is Consistent with Numerous LDC Provisions, Which Implement the Applicable Future Land Use Plan Policies.

For the same reasons that it is consistent with the Plan, the Project is consistent with the LDC, which implements the Plan. Pursuant to the LDC, the RPF Plant is zoned Heavy Industrial (I-2). Section 2.01.00 of the LDC, provides for the establishment and purpose of the City's Zoning Districts. Section 2.01.17 specifies the intent of the Heavy Industrial zoning district as follows:

The I-2 District is intended for the development of warehousing, fabrication, storage, and commercial services which are likely to produce adverse physical and environmental impacts such as noise, land, air, and water pollution and transportation conflicts. The Heavy Industrial District recognizes existing heavy manufacturing development with locations that have access to major highways. Residential development, with exception of a caretaker's unit, is not permissible within the zoning district. The designation of land for the I-2 District shall be based on compatibility with surrounding land uses, considering environmental sensitivity, intensity of use, hours of operation, heat, glare, fumes, noise, and visual impacts.

Table 2.03.02, Table of Land Uses in the LDC, lists numerous uses authorized as "permissible" or "permissible subject to supplemental standards," in the Heavy Industrial I-2 zoning district. For example, permissible uses include bakery plants, craft breweries, wineries, and distilleries, bulk storage yards for liquids and solids, bottling plants, wastewater treatment plants, and heavy manufacturing and assembly. Section 1.07.00 of the LDC defines several allowable uses for property zoned "Heavy Industrial," including "Heavy Manufacturing and Assembly," which contains language similar to that in the Plan regarding the prohibition of "chemical or petroleum manufacturing or refining." <sup>24</sup> As with the Plan, the terms "chemical or petroleum manufacturing or refining" are not defined in the LDC. Nonetheless, since the LDC implements the Plan and does not differ substantively from the Plan in terms of the specific "chemical manufacturing or refining" prohibition, the analysis set forth above demonstrating compliance with the Industrial future land use category also demonstrates compliance with the LDC's corresponding zoning requirements.

CITY OF FERNANDINA BEACH, FLA., LAND DEV. CODE § 1.07.00 (2023).

<sup>&</sup>lt;sup>24</sup> Section 1.07.00 of the LDC defines Manufacturing and/or Assembly – Heavy as follows:

<sup>&</sup>quot;[U]ses involving intensive manufacturing and industrial operations, including the manufacturing, assembly, fabrication, compounding, processing and/or treatment of extracted or raw materials or other industrial products; packaging and freight loading/unloading activities; utilization, handling and bulk storage of materials including raw materials, chemicals and hazardous materials associated with manufacturing processes; and all other associated or ancillary activities. Such use does not include heavy metal fabrication, batch plants, salvage yards, chemical or petroleum manufacturing or refining, rubber or plastics manufacturing, or other uses generating potentially harmful environmental or nuisance impacts.

# D. The Project is Consistent with and Furthers Plan Provisions Related to Economic Development.

The Plan aims to balance and harmonize economic development with environmental protection and community needs. The production of bioethanol through fermentation in the heavy industrial zoning district furthers the City's stated goal to harmonize these respective goals. On the one hand, the Project allows RYAM to realize the highest and best use of its private property through the utilization of its existing, unique SSL stream to produce another saleable product within its existing Heavy Industrial-designated footprint, which in turn supports and encourages millions of dollars of continued investment in the community, increasing the tax base and job opportunities in the City. On the other hand, the Project creates a renewable energy source that reduces greenhouse gas emissions without compromising safety or causing compatibility issues for neighboring properties.

Importantly, the Project contributes significantly to the City's economic development strategies outlined in Goal 12 of the Economic Development Element. <sup>25</sup> First, it ensures RYAM's continued contribution to the economic needs of the City. A preliminary economic study prepared by the University of Florida, *Economic Contributions of Forest Product Manufacturing in Fernandina Beach, Florida* (Dec. 10, 2024), concludes that RYAM's total economic contribution in 2023 in Nassau County alone was \$379.5 million and nearly \$575 million in the wider wood-procurement region. The Project on its own represents a \$50 Million capital investment by RYAM within the City limits, which increases the City's tax base. Moreover, by maintaining skilled labor jobs and expanding those jobs into the renewable energy sector and reducing reliance on imported fuels, the Project aids in diversifying the City's tax and employment base, contributing to long-term economic sustainability.

Not only does the Project expand the economic contribution of the RPF Plant, but it also supports its continued existence by introducing economic efficiencies and diversifying revenue streams that are vital to the RPF Plant's continued existence. Every business has to continue to invest, innovate, and diversify to remain competitive. The Project allows RYAM to do this and as a result, RYAM will be in a better position to protect local jobs and local operations for the long-term. Further the Project will support other existing businesses in the City by creating new jobs

CITY OF FERNANDINA BEACH, FLA., 2030 COMPREHENSIVE PLAN, Goal 12.

<sup>&</sup>lt;sup>25</sup> GOAL 12 of the City's Comprehensive Plan, the Economic Development Element, provides the following:

The City shall create and implement an economic development strategy focused on the retention, expansion, and relocation of high wage jobs and targeted businesses, while seeking to diversify the city's tax and employment base to lessen the tax burden for existing residents and businesses. This shall be accomplished through the preservation of the City's unique character, historical, cultural, and environmental assets and through promotion of sustainable development, redevelopment, and rehabilitation of properties with existing infrastructure and public services.

and additional demand for support services for the Project. This aligns with Objective 12.03<sup>26</sup> which aims to promote the expansion and sustainability of local businesses.

The Project also furthers Objective 12.07,<sup>27</sup> which emphasizes workforce development and retention. The Project creates opportunities for workforce training in green technology and renewable energy, while ensuring the continuation of existing jobs at the RPF Plant.

The Project is also consistent with and furthers Policy 1.01.01<sup>28</sup>, which directs the City to develop strategies to reduce greenhouse gas (GHG) emissions. The Project will reduce reliance on fossil fuels by not burning additional natural gas to replace the loss of sugars in the SSL returned to RYAM's recovery boiler, thereby contributing to the City's climate change mitigation goals. It will also assist the City in establishing a GHG baseline by providing data on emissions reductions through renewable energy production.

The Project directly aligns with Policy 1.01.04,<sup>29</sup> which encourages the production and use of energy from renewable resources. The Project's role in converting biomass into bioethanol exemplifies the City's policy for renewable energy, reducing its carbon footprint, and dependence on non-renewable energy sources.

The Project furthers Objective 1.02<sup>30</sup>, which focuses on managing growth through sustainable land use by facilitating industrial growth that is both economically beneficial and

The City of Fernandina Beach shall implement growth management techniques to ensure that land use decisions are consistent with the Fernandina Beach Comprehensive Plan, to provide land development regulations consistent with accepted planning principles and practices, to ensure that public services and facilities are provided when needed by

<sup>&</sup>lt;sup>26</sup> Objective 12.03 of the Comprehensive Plan's Economic Development Element, which details support of existing businesses, provides that "[t]he City shall develop and maintain strategies that support and promote the expansion of existing businesses within the City, including its small businesses." CITY OF FERNANDINA BEACH, FLA., 2030 COMPREHENSIVE PLAN, Goal 12, Objective 12.03.

<sup>&</sup>lt;sup>27</sup> Objective 12.07 of the Comprehensive Plan's Economic Development Element, which details workforce development and retention, provides that "[t]he City shall develop and maintain a strategy to provide training for and retain a qualified workforce to support targeted industries and to better prepare its local students for future careers relating to employment within its identified target industries and businesses." CITY OF FERNANDINA BEACH, FLA., 2030 COMPREHENSIVE PLAN, Goal 12, Objective 12.07.

<sup>&</sup>lt;sup>28</sup> Policy 1.01.01 of the Comprehensive Plan's Future Land Use Element provides that "The City shall explore various funding opportunities to assist in developing City GHGs emissions baseline data, which will support setting GHG emission goals, developing strategies to reduce climate change and mitigating and adapting to its impacts throughout the City." CITY OF FERNANDINA BEACH, FLA., 2030 COMPREHENSIVE PLAN, Goal 1, Objective 1.07, Pol'y 1.01.01.

<sup>&</sup>lt;sup>29</sup> Policy 1.01.04 of the Comprehensive Plan's Future Land Use Element provides that "The City shall encourage the production and use of energy generated from renewable resources." CITY OF FERNANDINA BEACH, FLA., 2030 COMPREHENSIVE PLAN, Goal 1, Objective 1.07, Pol'y 1.01.04.

<sup>&</sup>lt;sup>30</sup> Objective 1.02 of the Comprehensive Plan's Future Land Use Element, which details growth management, provides:

environmentally sustainable. It also demonstrates creative land use, as it repurposes SSL that is created by the existing RPF Plant for energy production within the RPF Plant current footprint, thereby preventing sprawl while ensuring the land is used efficiently.

# E. The Project is Consistent with and Furthers Plan Provisions Related to Conservation and Coastal Management.

The Project furthers Objective 5.12<sup>31</sup>, which commits the City to maintaining air quality standards as established by the EPA and FDEP. Bioethanol production, particularly from second-generation feedstocks, results in lower GHG and pollutant emissions, helping the City and region to meet air quality goals. The City's coordination with major industrial operators regarding air quality, as set forth in Policy 5.12.03<sup>32</sup>, will extend to the Project to ensure compliance with these standards.

The Project is a prime example of green development/sustainable development, as defined in the Comprehensive Plan.<sup>33</sup> Not only will the Project's design and operation integrate environmentally responsive technologies, use resources efficiently, and promote sustainability, the use of SSL as a feedstock minimizes waste and reduces environmental impact from the RPF Plant, fostering and furthering resource efficiency, as contemplated in Policy 5.13.02.

The Project qualifies as clean technology, supporting renewable energy and improving energy efficiency. By producing bioethanol from SSL and reducing carbon emissions, it embodies the principles outlined under green technology in the City's Comprehensive Plan.<sup>34</sup>

development, to control instances of sprawl, to support sustainability and to encourage creativity in land use and design.

CITY OF FERNANDINA BEACH, FLA., 2030 COMPREHENSIVE PLAN, Goal 1, Objective 1.02.

A development approach that integrates the following elements: environmental responsiveness, which benefits the surrounding environment; resource efficiency, which involves using resources in the construction and development and operations of buildings and/or communities in ways that are not wasteful; and sensitivity to culture and community, which is to foster a sense of community in design, construction, and operations.

CITY OF FERNANDINA BEACH, FLA., 2030 COMPREHENSIVE PLAN, Acronyms & Definitions (noting policy references to "5.13.02 (pg. 5-23), 11.06.03 (pg. 11-7), [and] 11.09 (pg. 11-10)").

<sup>&</sup>lt;sup>31</sup> Objective 5.12 of the Comprehensive Plan, which details air quality, provides that "[t]he City will continue to strive to meet air quality standards established by the EPA and the DEP." CITY OF FERNANDINA BEACH, FLA., 2030 COMPREHENSIVE PLAN, Goal 5, Objective 5.12.

<sup>&</sup>lt;sup>32</sup> Policy 5.12.03 of the Comprehensive Plan provides that "[t]he City will coordinate with major industrial operators within the City such as Rayonier, Smurfit Stone, and the Port regarding air quality information." CITY OF FERNANDINA BEACH, FLA., 2030 COMPREHENSIVE PLAN, Goal 5, Objective 5.12, Pol'y 5.12.03.

<sup>&</sup>lt;sup>33</sup> The Comprehensive Plan defines "Green Development/Sustainable Development" as:

<sup>&</sup>lt;sup>34</sup> The Comprehensive Plan defines "Green (or Clean) Technology" as: "[r]enewable energy and energy efficiency technologies plus other technologies that make use of resources more environmentally benign

# F. The Project is Consistent with and Furthers Plan Provisions Related to Property Rights.

Under Policy 1.02.07<sup>35</sup> and Goal 9<sup>36</sup>, the City must consider the rights of property owners to develop their land with broader community objectives. The establishment of the Project respects these rights while furthering the City's vision for sustainable growth. The Project supports RYAM's ability to utilize its property by realizing the highest and best use of the property in a manner consistent with the City's growth and environmental policies. Without the ability to modernize its treatment of SSL, RYAM's use of its property will be substantially diminished and the continued viability of its RPF Plant will be threatened. A finding that the Project furthers this Comprehensive Plan policy is consistent with the Court's instruction to expansively interpret land use regulations as needed to minimize impacts on "private rights of ownership" in *Rinker Materials Corp. v. City of N. Miami*, 286 So. 2d 552, 553 (Fla.1973).

## III. THE WEISS SEROTA MEMO CANNOT BE CONSIDERED BINDING

In the sections above, the factual and logical flaws of the Weiss Serota Memo are addressed. However, there are additional reasons why the Weiss Serota Memo cannot be treated as binding the City's decision regarding the interpretation of Plan and LDC provisions as applied to the Project.

and/or reduce carbon emissions." CITY OF FERNANDINA BEACH, FLA., 2030 COMPREHENSIVE PLAN, Acronyms & Definitions.

Pursuant to Section 163.3177(6)(i), Florida Statutes the following rights will be considered in local decision making:

- 1. The right of a property owner to physically possess and control his or her interests in the property, including easements, leases, or mineral rights.
- 2. The right of a property owner to use, maintain, develop, and improve his or her property for personal use or for the use of any other person, subject to state law and local ordinances.
- 3. The right of the property owner to privacy and to exclude others from the property to protect the owner's possessions and property.
- 4. The right of a property owner to dispose of his or her property through sale or gift.

CITY OF FERNANDINA BEACH, FLA, 2030 COMPREHENSIVE PLAN, Goal 9.

<sup>&</sup>lt;sup>35</sup> Policy 1.02.07 of the Comprehensive Plan provides that "[t]he City shall implement the Fernandina Beach Comprehensive Plan in a manner that acknowledges private property rights." CITY OF FERNANDINA BEACH, FLA., 2030 COMPREHENSIVE PLAN, Goal 1, Objective 1.02, Pol'y 1.02.07.

<sup>&</sup>lt;sup>36</sup> Goal 9 of the Comprehensive Plan, which details private property rights, provides the following:

## A. The City Manager Cannot Delegate Her Interpretive Authority to Outside Law Firm.

Section 1.04.00, LDC, identifies the City Manager as the person responsible to "administer, interpret and implement the standards, criteria and procedures of this LDC." This same section also expressly provides that "[t]he City Manager may delegate such responsibilities to City staff." By its very terms, this provision expressly limits the City Manager's ability to delegate interpretive authority to City staff only. The City Manager has no authority to either exercise or delegate power in a manner that is inconsistent with the powers expressly delegated to her. See Barry v. Garcia, 573 So.2d 932 (Fla. 3d DCA 1991). Since the LDC expressly limits the potential recipients of delegated authority from the City Manager to City staff, the City Manager has no authority to delegate interpretive authority to outside counsel like Weiss Serota. Notably, neither does the LDC grant the City Attorney any power to delegate interpretive power to an outside law firm.

While local government officials may obtain advisory opinions from outside law firms, the final interpretive responsibility must be exercised consistent with the express provisions of the applicable governing documents. *Id.* A decision to treat the Weiss Serota Memo as binding effectively converts it from advisory material to an exercise of delegated authority not contemplated by the LDC. Thus, the LDC does not permit the City to treat the Weiss Serota Memo as binding on the City Manager in her final interpretive role.

B. Treating the Weiss Serota Memo as Binding Violates Procedural Due Process By Depriving RYAM of Notice and an Opportunity to Be Heard and By Relying on the Consideration of Extra-Record Materials By Persons Not Competent to Evaluate Such Materials.

Even if the LDC did contemplate the delegation of interpretive authority to an outside law firm, which it does not, the exercise of that delegated authority would still be required to meet state and federal constitutional standards for procedural due process. While the specific requirements for procedural due process are context specific, the cornerstone of procedural due process is a fundamental right to be notified of a decision implicating a property interest and a fundamental right to be heard on that decision within a meaningful time and in a meaningful manner. *Goldberg* v. *Kelly*, 397 U.S. 254, 267-68 (1970).

Treatment of the Weiss Serota Memo as binding in this case would represent a shocking deprivation of RYAM's rights to notice and an opportunity to be heard. First and foremost, the Weiss Serota Memo was sought before RYAM had submitted any type of application to the City, and there is no reference in the LDC or the City's Charter to any type of pre-application interpretive decision-making process that would put RYAM on notice that its rights were being determined by this extra-administrative process.

Since RYAM had not submitted an application, or even a pre-application, before Weiss Serota issued its Memo, RYAM necessarily had no opportunity to be heard in the process. To the contrary, the only documents that Weiss Serota chose to consider were materials developed and submitted as part of the Air Permit Application and a Draft Permit issued by FDEP. These materials were provided to and by an entirely different agency under entirely different regulatory

criteria and thus have no relevance to the Site Plan Application requirements or the approval process used by the City. Weiss Serota itself has no scientific or technical expertise sufficient to correlate the two very different regulatory processes and never offered RYAM, who does have the scientific and technical expertise, the opportunity to provide information in the form of evidence or legal analysis to Weiss Serota in the development of its opinion. The Weiss Serota Memo cannot be considered "evidence" because it was written by persons unfamiliar with the Project, relied on irrelevant materials and was not otherwise authenticated. As a result, it is of dubious interpretive value.

Application of comprehensive plan and development code definitions to a specific application is inherently context specific. For that reason, local government decisions pertaining to site plans and other similar development orders typically provide notice and opportunity to be heard in the form of a quasi-judicial hearing, where the competency of evidence can be established and the grounds for decision-making can be adequately disclosed. See e.g. Park of Com. Assoc. v. City of Delray Beach, 636 So. 2d 12, 15 (Fla. 1994); Board of Cnty. Comm'r v. Snyder, 627 So. 2d 469, 474 (Fla. 1993); Grace v. Town of Palm Beach, 656 So. 2d 945, 946 (Fla. 4th DCA 1995); City of St. Petersburg v. Cardinal Industries Dev. Corp., 493 So. 2d 535, 537 (Fla. 2d DCA 1986).

The LDC provisions governing the site plan approval process, which is not well delineated in process by the LDC and does not appear to allow for a quasi-judicial hearing, already fall short of the typical regulatory standard in this regard. The decision to delegate contextual analysis to an outside agency with no expertise in the scientific or technical intricacies of the proposed add-on process before an application is even made and with no notice or opportunity to be heard to the would-be applicant is unprecedented. It is worth noting that the City itself does not appear to have taken similar action for other site plan approval applicants, raising the question of whether RYAM is being singled out in this regard.

If the City decides that the Weiss Serota Memo is binding, it will have effectively delegated its interpretive authority to a private law firm without the requisite expertise in a manner not contemplated by the LDC or other City Code, in a manner that to our knowledge it has never done before, and without having advised RYAM or given RYAM any opportunity to provide input. Accordingly, a decision that the City is bound by the Weiss Serota Memo would be a rank violation of RYAM's constitutionally protected due process rights.

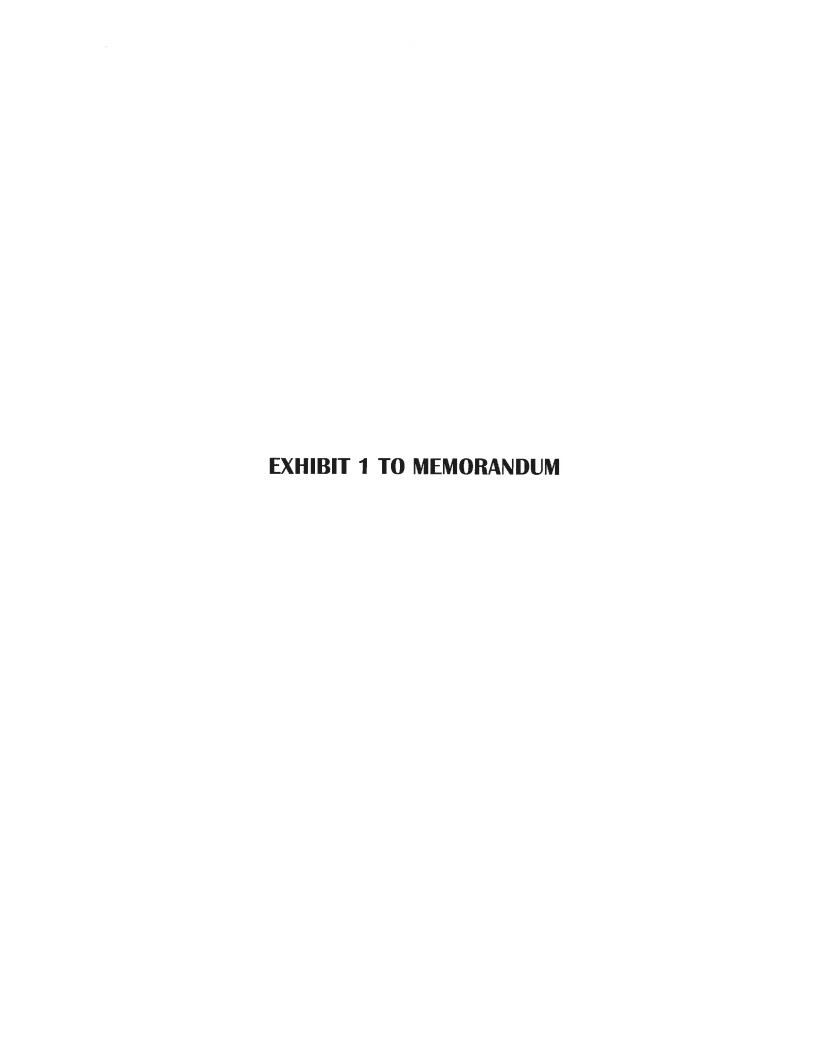
### IV. CONCLUSION

The Project is consistent with the Comprehensive Plan and LDC. The Project fits squarely within the description of permissible industrial uses contained in both Policy 1.07.12 of the Plan and in Sections 2.01.17, 1.07.00 and the Table of Land Uses, Table 2.03.02 of the LDC. In addition, the Project qualifies as a bona fide "associated or ancillary activity" to the existing pulp manufacturing process because it uses the SSL, a byproduct of the current manufacturing process. It constitutes an "add-on" process that is closely connected to and subordinate to the existing manufacturing process. Moreover, the Project does not constitute "chemical manufacturing" or "chemical refining." In fact, it is consistent with and furthers many of the goals in the City's Comprehensive Plan.

The Project consists of fermentation of sugars contained in SSL, a byproduct of the current pulp production process, and the physical separation process of drying and distillation. Technical literature explains that fermentation is not considered "chemical manufacturing or refining," but is instead a biological process occurring in nature that has been expressly excluded from other regulations that apply to chemical processing plants. Furthermore, the same biological process is utilized by many operations in Fernandina Beach and those operations, such as breweries, bakery plants, and water treatment plants, are all shown as permissible uses in Table 2.03.02 in the Heavy Industrial I-2 zoning district. At no time has this same process ever been classified as "chemical manufacturing and refining" by the City. Similarly, physical separation processes like distillation and drying are also allowed by the City in the Industrial land use category and Heavy Industrial I-2 zoning district and do not constitute "chemical refining." Accordingly, it would be arbitrary and capricious and render the Comprehensive Plan internally inconsistent to conclude that the Project is "chemical manufacturing" but breweries and distilleries, bakeries, and even the City's water treatment plant, are not.

The Weiss Serota Memo failed to undertake the required, thorough review and analysis to determine that the Project as is being presented to the City with RYAM's site plan approval application is inconsistent with the Comprehensive Plan. In reality, it simply determined that because ethanol is a chemical in the generic sense, the fact that it will be the resulting product of the Project must constitute chemical manufacturing and refining. This overly simplistic approach to the interpretive inquiry ignores the context provided by other language in the Plan and LDC, as well as other regulations and interpretive cases impacting the bioethanol production industry. As a result, the Weiss Serota Memo is deficient and its conclusion should not be considered persuasive by the City. In addition, by the very terms of the LDC, the Weiss Serota Memo has no binding effect on the City Manager in her final interpretive role. Moreover, the procedure used to procure the Weiss Serota Memo failed to adhere to basic principles of procedural due process in administrative land use decision-making and therefore cannot be binding on the City as to RYAM's application.

We look forward to the City's full consideration of RYAM's application and are available if you have any questions or need additional information.



## Ethanol production from natural sources of sugar

Glen P Fox

Anheuser-Busch Endowed Professor Malting and Brewing Science

University of California,

Department of Food Science & Technology

Davis, California

### **Executive summary**

For millions of years, a range of microorganisms have survived and evolved to change simple sugars like glucose and xylose into alcohol. Both fungi and bacteria are capable of undertaking such biochemical processes<sup>1</sup>. Around 10,000 years ago, humans through trial and error discovered the fermentation process when grains like barley and wheat sprouted and the liquid left over that cause the sprouting, absorbed some of the sugars released form the grains, and wild yeast changed the sugars to alcohol. It wasn't until the 1800's that we began to understand the science behind this process, but from those very early days of brewing with grains or fermenting grapes, societies have developed and major fermentation industries have been established.

It is typically thought that fermentation is used only in the production of alcohol for human consumption, but the same fermentation process, i.e., a series of natural biochemical processes, can also be used in the production of industrial ethanol (bioethanol). Bioethanol is produced from various sources of plant and wood materials or byproducts from industrial processing of these materials, using the normal fermentation process.

The use of such fermentation process is one way to reduce human dependance on fossil fuels as well as upcycle waste streams from industrial processing of organic materials.

<sup>&</sup>lt;sup>1</sup> Biochemical processes occur within living organisms and cells and involve the alteration of biomolecules – in fermentation, the conversion/transformation of sugars to ethanol. These types of processes can generally be referred to as metabolism. There are many examples of biochemical processes that occur in nature and within the human body. Examples other than fermentation include digestion and photosynthesis.

## History of fermentation

Alcohol, and specifically ethanol, is produced from the biochemical (enzymic) modification of sugar by several species. Typically, ethanol is produced by fungi and bacteria.

## Pre-cursors of fermented beverages

This natural process, that has been going on for tens of thousands of years, could be witnessed where ripe fruit will drop to the ground, and wild fungi will inoculate the open fruit and convert the sugars into alcohol. Animals would eat the sweet fruits with trace amounts of alcohol and get a good feeling from the low volume of alcohol produced. Our ancestors followed suit in eating these fermented fruits from the ground and enjoyed the residual sweetness but also feeling the effects of alcohol.

## Fermented beverages

Possibly, the very earliest fermented beverage was mead, where honey may have dripped from a hive onto the ground. The simple sugars in honey, (sucrose, fructose and glucose) are highly fermentable by fungi and a natural fermentation would have occurred. Animals and our human ancestor would have consumed this and found it very flavorsome with some nice side-effects. Over time, humans worked out a way to control this 'fermentation' process so they had a readily available supply of mead.

The next alcoholic beverage created by nature was beer. Around 10,000 years ago, in the now Middle East, people started to farm the cereal grains, barley and wheat, for food production. It is thought that some harvested grains got wet, and started to germinate. The natural enzymes in the germinating grains, the same enzymes that help grains germinate when a farmer plants seed for a new crop such as corn, hydrolyze starch (long polymers of glucose), into small chains forming the simple sugars, glucose and maltose (glucose + glucose), which are readily fermentable by fungi. Over the millennia, humans industrialized the brewing process to be the global multibillion dollar industry it is today.

The final major fermented beverage created by nature was wine, where wild grapes would have also split open on the vine or dropped to the ground when ripe, and the natural sugars (glucose and fructose) in grapes were converted to alcohol. Humans domesticated grapes, like the cereals, to making these fermented beverages a key part of life, with harvest celebrations, worshipping the ancient gods, as well as the very first taxation systems being based on alcohol production.

#### Fermentation in the human body

For a very small number of people, they go through the fermentation process and naturally produce alcohol in their bodies. This rare condition is called Auto-brewery syndrome, where gut bacteria produce the same alcohol (ethanol) as per a normal beer, or wine fermentation.

Ethanol is also the reason civilizations grew in regions with extreme population growth as the alcohol consumed allowed humans to survive the quality of toxic water supplies, prior to having any water treatment facilities.

## Discovery of the fermentation process

Despite productive industrialized alcohol processes growing in different societies globally, there was no understanding of this mystical, and magical conversion of the content of the fruit, or of the water used in germinating grains, into better tasting, safer and mind-altering products. It wasn't until the 1800s, through the efforts of scientists working at breweries, that alcohol was characterized.

It was also discovered what was responsible for this change from sugar to alcohol. In the 1700s, using one of the first microscopes, an 'animal' was first identified in a beer sample. This 'animal' was a wild yeast (fungus). Thus, our understanding of this very natural process of turning sugar into alcohol was born. From this study, researchers (Louis Pasteur and Emil Hansen) at Carlsberg Research Laboratory in Denmark in the 1800s purified the first beer yeast strain as well as identified the first spoilage bacteria.

Many 1000's of different type of fungi can carry out this natural fermentation process. But, for most alcohol beverages (and for bioethanol), we use yeast. For sour beer and kombucha, we use yeast and bacteria. For fermented food, such as sauerkraut, specific strains of bacteria can be used. For sour dough breads, it would be a mix of yeast and bacteria.

A recent article describes the value of yeast alone to the global economy is worth around \$900 billion. The screen shot below gives the key industries benefiting from our fungal friends.

One important point is that the bioethanol industry, like the alcoholic beverage industry, uses yeasts to convert starch from grains, and sugars from plant and wood materials to bioethanol.

## The fungus that's worth \$900 billion a year

BY NICHOLAS P MONEY FEBRUARY 25TH 2018

Brewing: \$311 billion, 2.23 million jobs

· Wine: \$220 billion, 1.7 million jobs

· Baking: \$311 billion, 1.8 million jobs

Bioethanol: \$44 billion estimated value

Yeast insulin: \$15 billion estimated value

Other yeast products: \$1 billion estimated value (Report FB2233)

https://blog.oup.com/2018/02/fungus-worth-900-billion/

## Relationship between bacteria and yeast

It is also important to understand the relationship between bacteria and yeast because some industries, like bioethanol, use only yeasts, some use bacteria, and some used mixed cultures of both. The figure below (Figure 1) shows the family tree of life.

Fungi, which includes yeasts, are in between plant species and animal species. Whereas, bacteria are not so related.

However, of specific interest are some common genes amongst all these different life forms. An example is a key enzyme, alpha-amylase, which is responsible for hydrolysis starch into its simple glucose units. It is present in plants, grains, and animals including humans (saliva and pancreas). Another example is alpha glucosidase, which is also involved in hydrolyzing starch in germination seeds and also used by fungi to break down maltose (glucose + glucose) from starch into two glucose molecules, which become of the first sugars required in the production of ethanol in any alcohol production.

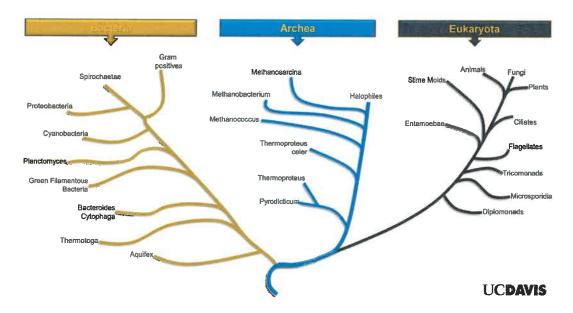
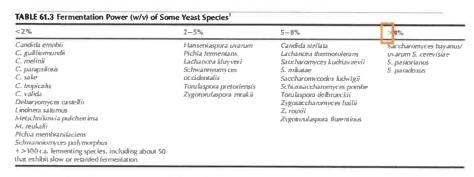


Figure 1. The tree of life.

As mentioned previously, there are 1000s of different fungi and each have different characteristics including different tolerances to ethanol production. The table below shows the different fungal species, including the most common in ethanol production, *Saccharomyces cerevisiae*.

## YEASTS & ALCOHOLIC FERMENTATIONS



- Saccharomyces Species most commonly in the production of alcohol
  - o saccharo means sweet
  - o myces means fungi
- All ferment carbohydrates (simple sugars) to alcohol
- Hundreds of specialized strains are used industrially, with some very specific applications.

**UCDAVIS** 

### Conversion of sugars to ethanol

As mentioned several times above, the fermentation process is a natural process where a sugar is converted to ethanol.

This 'conversion' is a simplified way of saying, the structure of the sugar is changed into a new structure, making it an alcohol (see Figures 2, 3, 4 & 5).

This multiple step conversion (Figure 4) is controlled by specific natural proteins, namely enzymes. What are the mechanics of this conversion? The simplest analysis is a lock and key mechanism. You require a specific key (enzyme) to open (modify) the lock (specific substrate). The stepwise glycolysis process follows foundational biochemistry, where the substrate (a sugar) is changed over several steps by different enzymes into a new chemical structure. The final structure being an ethanol molecule.

#### Details of the fermentation process to convert sugars to alcohol

Regardless of the type of sugar, the same metabolic pathway converts that sugar to alcohol and CO2. Figure 2 shows the different types of sugars and their assimilation into a yeast cell for beer fermentation. For a simple monosaccharide sugar like glucose or fructose, the transport into the cell is straight forward. But for disaccharides, like sucrose and maltose, then an enzyme splits the disaccharide into the two base units before these are transported into the cell.

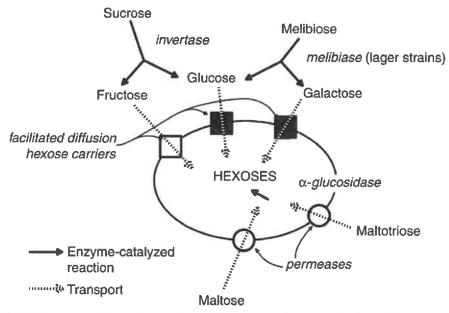


FIG. 15.1. Access of sugars to yeast cells. (Courtesy C. W. Bamforth—© ASBC.)

**Figure 2.** Transport of different sugar molecules into a yeast cell. From Scientific Principles of Malting and Brewing (Bamforth & Fox (2023)).

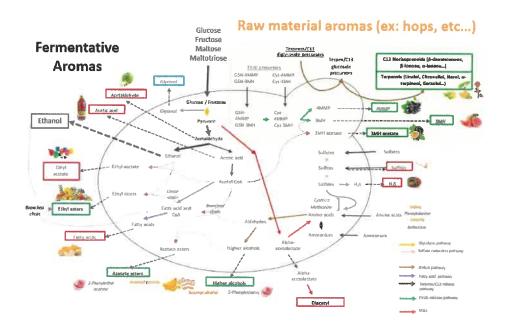


Figure 3. Different pathways within the yeast cell.

Figure 4 shows the structure of sucrose, the most abundant sugar in plants. Sucrose is made of two other monosaccharides, glucose and fructose. During the growth of the plant, sucrose is made, split into glucose and fructose and then remade during the daily diurnal cycle (light/dark or day/night) as these sugars are used in other metabolic processes. Other plant sugars are used in different metabolic process such as xylose which is used in building cell walls of plants.

In a liquid matrix where there are different sugars, yeast have a preference of sugars so they metabolism sugars in specific orders, i.e., monosaccharides (one sugar) before disaccharides (two bonded sugars). The three examples provided show glucose and fructose being converted to pyruvate via the glycolysis process or the Embden Meyerhof-Parnas (EMP) pathway (Figure 5). Pyruvate is important because it is the first step for several pathways to ethanol, depending upon fermentation conditions.

## Pyruvate conversion during fermentation

For alcohol production, pyruvate is enzymically changed in two steps to acetaldehyde, then ethanol (Figure 6). The overall mass balance from one sugar molecule is two molecules of ethanol and two molecules of CO<sub>2</sub>.

Pyruvate is also the molecule used in other fermentation conversions, but these are dictated by the amount of oxygen available or of another organisms involved in the process. More relatable examples of pyruvate being converted into other molecules from the natural glycolysis process are the production or sour bread or yogurt. In these cases, pyruvate is converted to lactate (lactic acid) via a single enzymic reaction (lactate dehydrogenase). This is called lactic acid fermentation.

To bring another example, which almost very human can relate to when exercising, is sore muscles. Under aerobic conditions when exercising, you have a lot of air (oxygen) coming into your cells via blood transport, and you can exercise for a long time. However, when you start to run out of air through intense exercise, your system goes anaerobic (low oxygen), then your body converts pyruvate to lactic acid. Your muscles are crying out for relief. Only when you regain your normal breathing then you reduced the production of lactic acid from pyruvate.

FIG. 15.7. The action of invertase. (Courtesy C. W. Bamforth and G. P. Fox—⊕ ASBC. Drawing by L. Watson.)

**Figure 4**. The structure of sucrose, glucose and fructose. From Scientific Principles of Malting and Brewing (Bamforth & Fox (2023)).

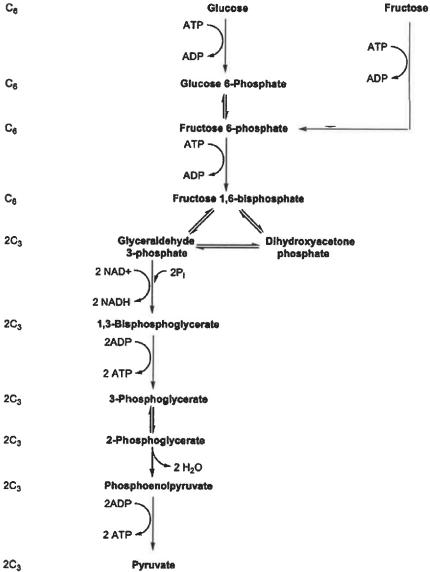


FIG. 15.8. The Embden-Meyerhof-Parnas pathway. (Reproduced by permission from Bamforth, C. W., and Cook, D. J. (2019) Food, Fermentation and Microorganisms, 2nd ed. Wiley Blackwell, Chichester, U.K.)

**Figure 5.** Enzymic conversion of glucose and fructose to pyruvate. Modified from Figure 15.8 in Scientific Principles of malting and Brewing (Bamforth & Fox (2023)).

The C on the left of the Figure 5 indicates the number of carbons in each molecule, e.g., pyruvate is a 3 carbon molecule ( $C_3$ ). Additionally, the changes in ATP (adenosine triphosphate), and ADP (adenosine diphosphate) shown in Figure 5 show the change in energy for the specific enzyme reactions.

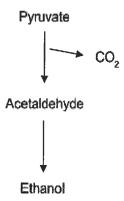
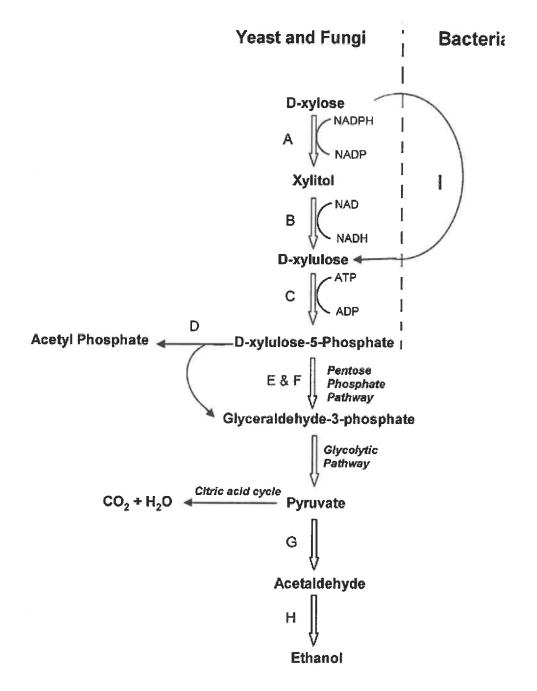


Figure 6. Enzymic conversion of pyruvate to ethanol.

## Conversion of C5 sugars

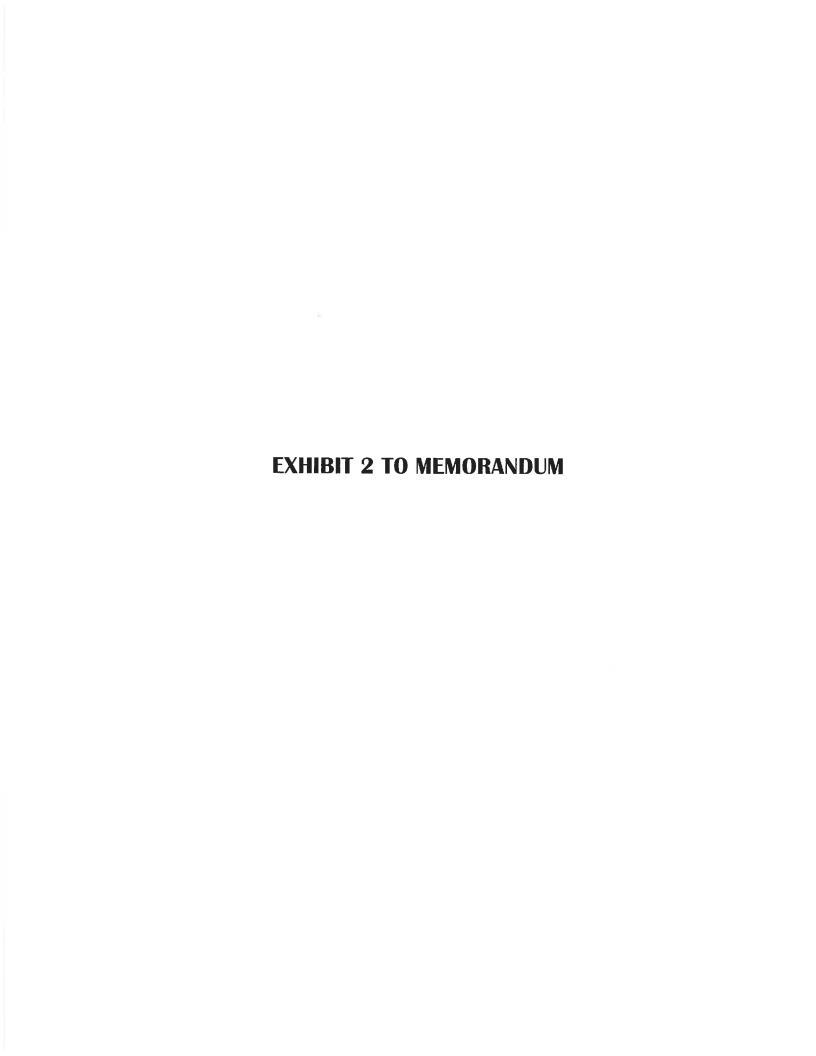
The enzymic pathway for converting a  $C_5$  molecule like xylose to ethanol involves several steps prior to the last 9 steps of the EMP pathway. Not all yeasts are capable of doing this; so, specific yeast strains are required for the conversion of C5 sugars into ethanol. Figure 7 shows the more complex pathway for the natural conversion of the C5 sugar xylose to pyruvate, then pyruvate conversion to ethanol follows the same steps shown in Figure 6.



**Figure 7.** The natural enzymic conversion of the five carbon sugar, xylose, to pyruvate following the pentose phosphate pathway, then the Embden Meyerhof-Parnas pathway. Pyruvate is converted to ethanol as per the normal acetaldehyde steps.

#### Conclusion

The production of alcohol from sugars is a natural molecular conversion process, regardless of the source of sugars or the end product. In recent decades, there have been specific industrial processes successfully established to use specific plant materials for the production of bioethanol. These processes use plant materials, with the addition of specific yeast strains to produce alcohol through the natural fermentation process under controlled conditions.



### Marlin & Barrel Distillery

A prefacing comment; at the Federal level we are primarily regulated by the Trade & Tax Bureau (TTB). This is the governing agency that authorizes all distillation plants in the nation.

#### Distillation & Fermentation

I tell anyone who's toured our facility that most people think that still make alcohol. That's a misnomer. All alcohol is made through the process of fermentation. This is true for beer, wine and distilled spirits. The still is simply an extraction tool.

#### Fermentation

Fermentation starts with a sugar source. Sugar is a generic term, but for simplicity's sake, any fermentable sugar is the basis for alcohol. A ferment is the combination of sugar, water and the necessary ingredients to create a healthy environment for yeast. Yeast are added to a ferment as the last step and, at first, they will propagate. That activity lasts until the right oxygen depleted anaerobic conditions occur. At that point the yeast find sugar molecules and break sugar into CO2 (which escapes the tank) and ethyl alcohol. That lasts until all sugars have been consumed.

This is a natural biological process and actually happens in the wild in a large number of instances. For example, rum made in Caribbean islands once controlled by the French still follow their original process. The juice of sugar cane is fresh pressed and immediately will see wild yeast begin the process of fermentation. This is the nature of yeast and sugar.

#### Distillation

Once all of the potential alcohol is made in a ferment then its time to take the part you want and discard the rest. Since ferments are a natural process you'll find that a small amount of other-than ethyl alcohol can be present. Said another way, even though the vast majority of alcohol made is ethyl alcohol, there will be other types present. Each will have its unique molecular weight and that directly corresponds to a unique boiling point. As the ferment is distilled, the liquid is heated and the lighter compounds that require less heat to change from a liquid to vapor state will climb the sill column first and further then those needing more energy to get to their vapor form. In this way a still organizes all of the different molecule types present in a finished ferment. From that position of stratification a distiller can extract the desired piece of the ferment, cool it to liquid temperature again and then collect it. In no part of this process are chemicals being changes. This is simply a liquid sifter of sorts.



## Ethanol production from natural sources of sugar

Glen P Fox

Anheuser-Busch Endowed Professor Malting and Brewing Science

University of California,

Department of Food Science & Technology

Davis, California

## **Executive summary**

For millions of years, a range of microorganisms have survived and evolved to change simple sugars like glucose and xylose into alcohol. Both fungi and bacteria are capable of undertaking such biochemical processes<sup>1</sup>. Around 10,000 years ago, humans through trial and error discovered the fermentation process when grains like barley and wheat sprouted and the liquid left over that cause the sprouting, absorbed some of the sugars released form the grains, and wild yeast changed the sugars to alcohol. It wasn't until the 1800's that we began to understand the science behind this process, but from those very early days of brewing with grains or fermenting grapes, societies have developed and major fermentation industries have been established.

It is typically thought that fermentation is used only in the production of alcohol for human consumption, but the same fermentation process, i.e., a series of natural biochemical processes, can also be used in the production of industrial ethanol (bioethanol). Bioethanol is produced from various sources of plant and wood materials or byproducts from industrial processing of these materials, using the normal fermentation process.

The use of such fermentation process is one way to reduce human dependance on fossil fuels as well as upcycle waste streams from industrial processing of organic materials.

<sup>&</sup>lt;sup>1</sup> Biochemical processes occur within living organisms and cells and involve the alteration of biomolecules – in fermentation, the conversion/transformation of sugars to ethanol. These types of processes can generally be referred to as metabolism. There are many examples of biochemical processes that occur in nature and within the human body. Examples other than fermentation include digestion and photosynthesis.

## **History of fermentation**

Alcohol, and specifically ethanol, is produced from the biochemical (enzymic) modification of sugar by several species. Typically, ethanol is produced by fungi and bacteria.

## Pre-cursors of fermented beverages

This natural process, that has been going on for tens of thousands of years, could be witnessed where ripe fruit will drop to the ground, and wild fungi will inoculate the open fruit and convert the sugars into alcohol. Animals would eat the sweet fruits with trace amounts of alcohol and get a good feeling from the low volume of alcohol produced. Our ancestors followed suit in eating these fermented fruits from the ground and enjoyed the residual sweetness but also feeling the effects of alcohol.

## Fermented beverages

Possibly, the very earliest fermented beverage was mead, where honey may have dripped from a hive onto the ground. The simple sugars in honey, (sucrose, fructose and glucose) are highly fermentable by fungi and a natural fermentation would have occurred. Animals and our human ancestor would have consumed this and found it very flavorsome with some nice side-effects. Over time, humans worked out a way to control this 'fermentation' process so they had a readily available supply of mead.

The next alcoholic beverage created by nature was beer. Around 10,000 years ago, in the now Middle East, people started to farm the cereal grains, barley and wheat, for food production. It is thought that some harvested grains got wet, and started to germinate. The natural enzymes in the germinating grains, the same enzymes that help grains germinate when a farmer plants seed for a new crop such as corn, hydrolyze starch (long polymers of glucose), into small chains forming the simple sugars, glucose and maltose (glucose + glucose), which are readily fermentable by fungi. Over the millennia, humans industrialized the brewing process to be the global multibillion dollar industry it is today.

The final major fermented beverage created by nature was wine, where wild grapes would have also split open on the vine or dropped to the ground when ripe, and the natural sugars (glucose and fructose) in grapes were converted to alcohol. Humans domesticated grapes, like the cereals, to making these fermented beverages a key part of life, with harvest celebrations, worshipping the ancient gods, as well as the very first taxation systems being based on alcohol production.

Fermentation in the human body

For a very small number of people, they go through the fermentation process and naturally produce alcohol in their bodies. This rare condition is called Auto-brewery syndrome, where gut bacteria produce the same alcohol (ethanol) as per a normal beer, or wine fermentation.

Ethanol is also the reason civilizations grew in regions with extreme population growth as the alcohol consumed allowed humans to survive the quality of toxic water supplies, prior to having any water treatment facilities.

## Discovery of the fermentation process

Despite productive industrialized alcohol processes growing in different societies globally, there was no understanding of this mystical, and magical conversion of the content of the fruit, or of the water used in germinating grains, into better tasting, safer and mind-altering products. It wasn't until the 1800s, through the efforts of scientists working at breweries, that alcohol was characterized.

It was also discovered what was responsible for this change from sugar to alcohol. In the 1700s, using one of the first microscopes, an 'animal' was first identified in a beer sample. This 'animal' was a wild yeast (fungus). Thus, our understanding of this very natural process of turning sugar into alcohol was born. From this study, researchers (Louis Pasteur and Emil Hansen) at Carlsberg Research Laboratory in Denmark in the 1800s purified the first beer yeast strain as well as identified the first spoilage bacteria.

Many 1000's of different type of fungi can carry out this natural fermentation process. But, for most alcohol beverages (and for bioethanol), we use yeast. For sour beer and kombucha, we use yeast and bacteria. For fermented food, such as sauerkraut, specific strains of bacteria can be used. For sour dough breads, it would be a mix of yeast and bacteria.

A recent article describes the value of yeast alone to the global economy is worth around \$900 billion. The screen shot below gives the key industries benefiting from our fungal friends.

One important point is that the bioethanol industry, like the alcoholic beverage industry, uses yeasts to convert starch from grains, and sugars from plant and wood materials to bioethanol.

## The fungus that's worth \$900 billion a year

BY NICHOLAS P. MONEY FEBRUARY 25TH 2018

Brewing: \$311 billion, 2.23 million jobs

Wine: \$220 billion, 1.7 million jobs

Baking: \$311 billion, 1.8 million jobs

Bioethanol: \$44 billion estimated value

· Yeast insulin: \$15 billion estimated value

Other yeast products: \$1 billion estimated value (Report FB2233)

https://blog.oup.com/2018/02/fungus-worth-900-billion/

## Relationship between bacteria and yeast

It is also important to understand the relationship between bacteria and yeast because some industries, like bioethanol, use only yeasts, some use bacteria, and some used mixed cultures of both. The figure below (Figure 1) shows the family tree of life.

Fungi, which includes yeasts, are in between plant species and animal species. Whereas, bacteria are not so related.

However, of specific interest are some common genes amongst all these different life forms. An example is a key enzyme, alpha-amylase, which is responsible for hydrolysis starch into its simple glucose units. It is present in plants, grains, and animals including humans (saliva and pancreas). Another example is alpha glucosidase, which is also involved in hydrolyzing starch in germination seeds and also used by fungi to break down maltose (glucose + glucose) from starch into two glucose molecules, which become of the first sugars required in the production of ethanol in any alcohol production.

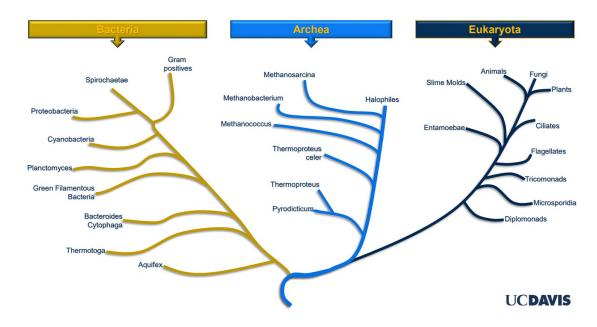
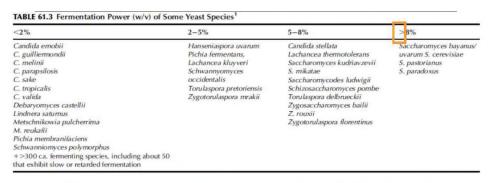


Figure 1. The tree of life.

As mentioned previously, there are 1000s of different fungi and each have different characteristics including different tolerances to ethanol production. The table below shows the different fungal species, including the most common in ethanol production, *Saccharomyces cerevisiae*.

## YEASTS & ALCOHOLIC FERMENTATIONS



- o Saccharomyces Species most commonly in the production of alcohol
  - o saccharo means sweet
  - o myces means fungi
- o All ferment carbohydrates (simple sugars) to alcohol
- Hundreds of specialized strains are used industrially, with some very specific applications.



### Conversion of sugars to ethanol

As mentioned several times above, the fermentation process is a natural process where a sugar is converted to ethanol.

This 'conversion' is a simplified way of saying, the structure of the sugar is changed into a new structure, making it an alcohol (see Figures 2, 3, 4 & 5).

This multiple step conversion (Figure 4) is controlled by specific natural proteins, namely enzymes. What are the mechanics of this conversion? The simplest analysis is a lock and key mechanism. You require a specific key (enzyme) to open (modify) the lock (specific substrate). The stepwise glycolysis process follows foundational biochemistry, where the substrate (a sugar) is changed over several steps by different enzymes into a new chemical structure. The final structure being an ethanol molecule.

## Details of the fermentation process to convert sugars to alcohol

Regardless of the type of sugar, the same metabolic pathway converts that sugar to alcohol and CO2. Figure 2 shows the different types of sugars and their assimilation into a yeast cell for beer fermentation. For a simple monosaccharide sugar like glucose or fructose, the transport into the cell is straight forward. But for disaccharides, like sucrose and maltose, then an enzyme splits the disaccharide into the two base units before these are transported into the cell.

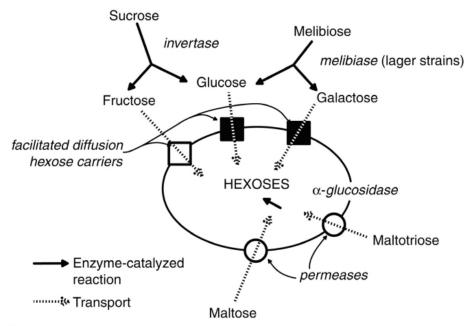


FIG. 15.1. Access of sugars to yeast cells. (Courtesy C. W. Bamforth—© ASBC.)

**Figure 2.** Transport of different sugar molecules into a yeast cell. From Scientific Principles of Malting and Brewing (Bamforth & Fox (2023)).

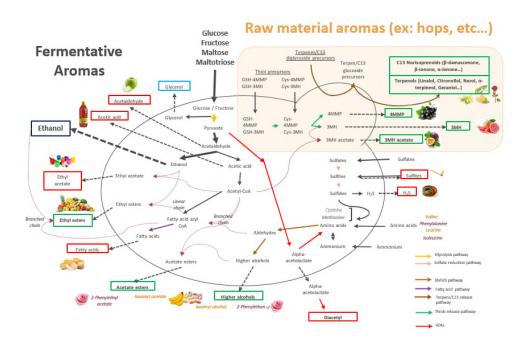


Figure 3. Different pathways within the yeast cell.

Figure 4 shows the structure of sucrose, the most abundant sugar in plants. Sucrose is made of two other monosaccharides, glucose and fructose. During the growth of the plant, sucrose is made, split into glucose and fructose and then remade during the daily diurnal cycle (light/dark or day/night) as these sugars are used in other metabolic processes. Other plant sugars are used in different metabolic process such as xylose which is used in building cell walls of plants.

In a liquid matrix where there are different sugars, yeast have a preference of sugars so they metabolism sugars in specific orders, i.e., monosaccharides (one sugar) before disaccharides (two bonded sugars). The three examples provided show glucose and fructose being converted to pyruvate via the glycolysis process or the Embden Meyerhof-Parnas (EMP) pathway (Figure 5). Pyruvate is important because it is the first step for several pathways to ethanol, depending upon fermentation conditions.

## Pyruvate conversion during fermentation

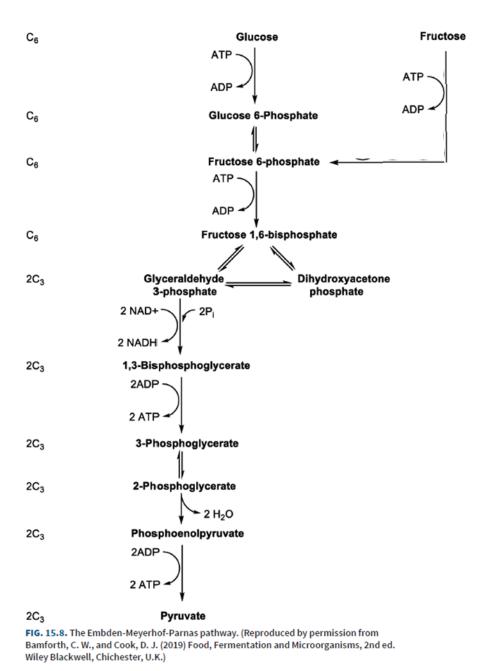
For alcohol production, pyruvate is enzymically changed in two steps to acetaldehyde, then ethanol (Figure 6). The overall mass balance from one sugar molecule is two molecules of ethanol and two molecules of CO<sub>2</sub>.

Pyruvate is also the molecule used in other fermentation conversions, but these are dictated by the amount of oxygen available or of another organisms involved in the process. More relatable examples of pyruvate being converted into other molecules from the natural glycolysis process are the production or sour bread or yogurt. In these cases, pyruvate is converted to lactate (lactic acid) via a single enzymic reaction (lactate dehydrogenase). This is called lactic acid fermentation.

To bring another example, which almost very human can relate to when exercising, is sore muscles. Under aerobic conditions when exercising, you have a lot of air (oxygen) coming into your cells via blood transport, and you can exercise for a long time. However, when you start to run out of air through intense exercise, your system goes anaerobic (low oxygen), then your body converts pyruvate to lactic acid. Your muscles are crying out for relief. Only when you regain your normal breathing then you reduced the production of lactic acid from pyruvate.

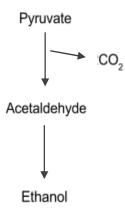
FIG. 15.7. The action of invertase. (Courtesy C. W. Bamforth and G. P. Fox—© ASBC. Drawing by L. Watson.)

**Figure 4**. The structure of sucrose, glucose and fructose. From Scientific Principles of Malting and Brewing (Bamforth & Fox (2023)).



**Figure 5.** Enzymic conversion of glucose and fructose to pyruvate. Modified from Figure 15.8 in Scientific Principles of malting and Brewing (Bamforth & Fox (2023)).

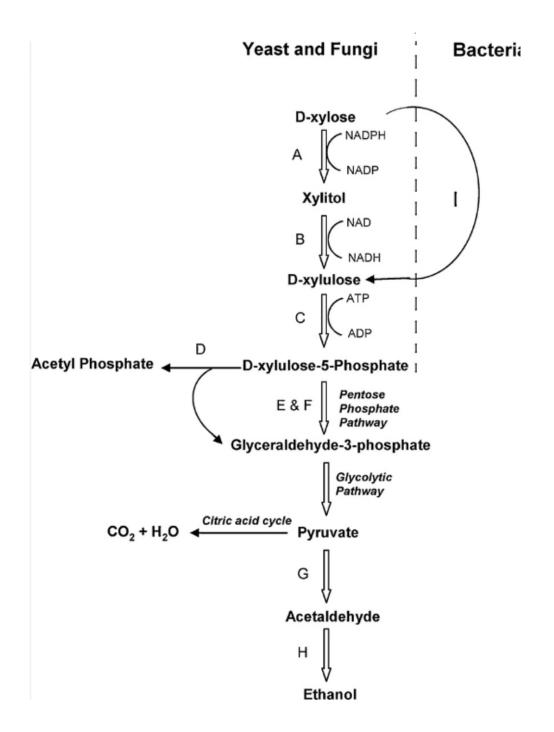
The C on the left of the Figure 5 indicates the number of carbons in each molecule, e.g., pyruvate is a 3 carbon molecule (C<sub>3</sub>). Additionally, the changes in ATP (adenosine triphosphate), and ADP (adenosine diphosphate) shown in Figure 5 show the change in energy for the specific enzyme reactions.



**Figure 6.** Enzymic conversion of pyruvate to ethanol.

## Conversion of C5 sugars

The enzymic pathway for converting a  $C_5$  molecule like xylose to ethanol involves several steps prior to the last 9 steps of the EMP pathway. Not all yeasts are capable of doing this; so, specific yeast strains are required for the conversion of C5 sugars into ethanol. Figure 7 shows the more complex pathway for the natural conversion of the C5 sugar xylose to pyruvate, then pyruvate conversion to ethanol follows the same steps shown in Figure 6.



**Figure 7.** The natural enzymic conversion of the five carbon sugar, xylose, to pyruvate following the pentose phosphate pathway, then the Embden Meyerhof-Parnas pathway. Pyruvate is converted to ethanol as per the normal acetaldehyde steps.

#### Conclusion

The production of alcohol from sugars is a natural molecular conversion process, regardless of the source of sugars or the end product. In recent decades, there have been specific industrial processes successfully established to use specific plant materials for the production of bioethanol. These processes use plant materials, with the addition of specific yeast strains to produce alcohol through the natural fermentation process under controlled conditions.

#### Marlin & Barrel Distillery

A prefacing comment; at the Federal level we are primarily regulated by the Trade & Tax Bureau (TTB). This is the governing agency that authorizes all distillation plants in the nation.

#### Distillation & Fermentation

I tell anyone who's toured our facility that most people think that still make alcohol. That's a misnomer. All alcohol is made through the process of fermentation. This is true for beer, wine and distilled spirits. The still is simply an extraction tool.

#### Fermentation

Fermentation starts with a sugar source. Sugar is a generic term, but for simplicity's sake, any fermentable sugar is the basis for alcohol. A ferment is the combination of sugar, water and the necessary ingredients to create a healthy environment for yeast. Yeast are added to a ferment as the last step and, at first, they will propagate. That activity lasts until the right oxygen depleted anaerobic conditions occur. At that point the yeast find sugar molecules and break sugar into CO2 (which escapes the tank) and ethyl alcohol. That lasts until all sugars have been consumed.

This is a natural biological process and actually happens in the wild in a large number of instances. For example, rum made in Caribbean islands once controlled by the French still follow their original process. The juice of sugar cane is fresh pressed and immediately will see wild yeast begin the process of fermentation. This is the nature of yeast and sugar.

#### Distillation

Once all of the potential alcohol is made in a ferment then its time to take the part you want and discard the rest. Since ferments are a natural process you'll find that a small amount of other-than ethyl alcohol can be present. Said another way, even though the vast majority of alcohol made is ethyl alcohol, there will be other types present. Each will have its unique molecular weight and that directly corresponds to a unique boiling point. As the ferment is distilled, the liquid is heated and the lighter compounds that require less heat to change from a liquid to vapor state will climb the sill column first and further then those needing more energy to get to their vapor form. In this way a still organizes all of the different molecule types present in a finished ferment. From that position of stratification a distiller can extract the desired piece of the ferment, cool it to liquid temperature again and then collect it. In no part of this process are chemicals being changes. This is simply a liquid sifter of sorts.





# Advances in Applied Microbiology

Volume 62, 2007, Pages 105-135

# The Scale-Up of Microbial Batch and Fed-**Batch Fermentation Processes**

Christopher J. Hewitt \*, Alvin W. Nienow †

Show more V

Share 55 Cite

https://doi.org/10.1016/S0065-2164(07)62005-X 7 Get rights and content ↗

# **Publisher Summary**

The scale-up of single-celled aerobic microbial fermentation processes is complicated that can lead to unpredictable process performance. However, this is not due to the introduction of fluid dynamic generated stresses (or so-called "shear damage"), whether arising from agitator generated turbulence or bursting bubbles, rather it is because the large-scale fed-batch bioreactor provides a very dynamic environment with large spatial and temporal heterogeneities. Such environmental heterogeneities can induce multiple physiological responses in cells. These responses consume energy and resources such that biomass concentration and product yields can be reduced. These phenomena are not observed in well-mixed homogeneous laboratoryscale reactors where much process development is done and their effects are difficult to model mathematically. Therefore, the ability to obtain data on how a recombinant laboratory process may perform at the large scale, dependent on feeding regime

employed or controlling action taken is invaluable for any detailed and informed development program.

# Introduction

Microorganisms are important both for human health and to industry, so the fedbatch cultivation of microbial strains, often overexpressing recombinant or natural proteins, to high cell density has become an increasingly important technique throughout the field of biotechnology, from basic research programs to large-scale pharmaceutical production processes (Hewitt et al., 1999). The scale-up of such a process is usually the final step in any research and development program leading to the large-scale industrial manufacture of such products by fermentation (Einsele, 1978). It is important to understand that the process of scaling-up a fermentation system is frequently governed by a number of important engineering considerations and not simply a matter of increasing culture and vessel volume. Therefore, it is perhaps surprising when the large scale does not perform as the small-scale laboratory process does. It is often observed that the biomass yield and any growthassociated products are often decreased on the scale-up of an aerobic process (Enfors et al., 2001). For Saccharomyces cerevisiae, the biomass yield on molasses increased by 7% when the process was scaled-down from 120 m<sup>3</sup> to 10 liter even when a seemingly identical strain, medium, and process were employed (George et al., 1993). In an Escherichia coli fed-batch recombinant protein process, the maximum cell density reached was found to be 20% lower when scaling-up from 3 liter to 9 m<sup>3</sup> and the pattern of acetic acid formation had changed. (Bylund et al., 1998). During another study (Enfors et al., 2001), the performance of a recombinant strain of E. coli during fed-batch culture was found to vary on scale-up from the laboratory-scale to 10-30 m<sup>3</sup> industrial bioreactors. This included lower biomass yields, recombinant protein accumulation, and surprisingly perhaps a higher cell viability. These findings are typical of those found when scaling-up most fermentation processes, yet only a few mechanisms have been presented that can satisfactorily explain these phenomena.

In this chapter, we will briefly discuss the main engineering considerations involved in fermentation scale-up and then critically review those mechanisms thought to be responsible for any detrimental change in bioprocessing at the larger-scale. Although it addresses mainly *E. coli* fed-batch fermentations, much of the discussion also applies to batch and other single-celled aerobic microbial fermentations too.

# Access through your organization

Check access to the full text by signing in through your organization.

Access through your institution

# Section snippets

# Agitator tasks in the bioreactor

The agitation system in the bioreactor provides the liquid motion that enables many different tasks to be fulfilled. An example of a typical stirred bioreactor is shown in diagrammatic form in Fig. 5.1. It is important to understand the interaction between the fluid motion, the agitator speed, and the power input into the bioreactor and these tasks. It is also necessary to know how a change of scale affects these relationships. Many of these aspects can be studied without carrying out a...

# Fluid mechanical stress or so-called "shear damage"

Anecdotal reference to the damaging effects on cells of fluid mechanical stress or so-called "shear damage" is frequently made to explain poor process performance when mechanical agitation and aeration are introduced into a bioreactor as compared to the nonagitated and nonsparged conditions in a shake flask or microtitre plate (Thomas, 1990). Thomas (1990) suggested that cells might be considered to be unaffected by fluid dynamic stresses if they were of a size smaller than the Kolmogoroff...

# Conclusions and Future Perspective

The scale-up of single-celled aerobic microbial fermentation processes is complicated, and unpredictable process performance can result. However, this is not due to the introduction of fluid dynamic generated stresses (or so-called "shear damage"), whether arising from agitator generated turbulence or bursting bubbles, rather it is because the large-scale fed-batch bioreactor provides a very dynamic environment with large spatial and temporal heterogeneities. Such environmental heterogeneities...

Recommended articles

# References (53)

L.A. Boon et al.

Comparing a range of impellers for "stirring as foam disruption."

Biochem. Eng. J. (2002)

J. Buchs et al.

Out of phase operating conditions, a hitherto unknown phenomenon in shaking bioreactors

Biochem. Eng. J. (2001)

S.O. Enfors et al.

Physiological responses to mixing in large bioreactors

J. Biotechnol. (2001)

S.Y. Lee

High cell-density culture of E. coli

**TIBTECH (1996)** 

B.J. Minihane et al.

Fed-batch culture technology

Biotechnol. Adv. (1986)

A.W. Nienow et al.

The versatility of up-pumping hydrofoil agitators

Trans. Inst. Chem. Eng. A (2004)

P. Vrabel et al.

Mixing in large-scale vessels, stirred with multiple radial or radial and axial pumping up impellers: Modelling and measurements

Chem. Eng. Sci. (2000)

A. Amanullah et al.

Mixing in the fermentation and cell culture industries

L.C. Andersson et al.

Cell segregation and lysis have profound effects on growth of *Escherichia coli* in high cell density fed batch cultures

Biotechnol. Prog. (1996)

N. Borth et al.

Flow cytometric analysis of bacterial physiology during induction of foreign protein synthesis in recombinant *Escherichia coli* cells

Cytometry (1998)



View more references

Cited by (108)

Fermentation utilizing engineered microbes: revolutionizing the production of commercial products from plant-derived bioactive compounds

2024, Scale-up and Chemical Process for Microbial Production of Plant-Derived Bioactive Compounds: From Laboratory to Industry

# Digital models in biotechnology: Towards multi-scale integration and implementation

2022, Biotechnology Advances

#### Citation Excerpt:

...Other considerations, including the bioreactor design, shear-force limitations (cell dependent), and culture conditions, mean that following such a general rule of thumb may not ensure homogeneous conditions, particularly at larger scales. Challenged by the heterogeneous environment, the production rate may differ considerably from an ideal mixing situation at bench scales, as reported for several different strains and processes (Bylund et al., 1998; Enfors et al., 2001; George et al., 1998; Hewitt and Nienow, 2007; Junker, 2004; Takors, 2012; Xu et al., 1999). Combined CFD and kinetic models can be used to understand better the effect of environmental fluctuations a given organism may become exposed to in an industrial scale bioreactor....

Show abstract ✓

# A bench-scale rotating bioreactor with improved oxygen transfer and cell growth

2022, Chemical Engineering Science

Show abstract ✓

# Fermentation and downstream processing: Part 1

2022, Integration and Optimization of Unit Operations: Review of Unit Operations from R&D to Production: Impacts of Upstream and Downstream Process Decisions

Show abstract ✓

Understanding the scale-up of fermentation processes from the viewpoint of the flow field in bioreactors and the physiological response of strains

2021, Chinese Journal of Chemical Engineering

Show abstract ∨

# Expression, purification, and characterization of pneumococcal PsaA-PspA fusion protein

2021, Protein Expression and Purifica	ion
---------------------------------------	-----

Show abstract ✓



View all citing articles on Scopus ↗

View full text

Copyright © 2007 Elsevier Inc. All rights reserved.

All content on this site: Copyright © 2024 Elsevier B.V., its licensors, and contributors. All rights are reserved, including those for text and data mining, AI training, and similar technologies. For all open access content, the Creative Commons licensing terms apply.

Composite Affidavit Attachr	ment 4

As a library, NLM provides access to scientific literature. Inclusion in an NLM database does not imply endorsement of, or agreement with, the contents by NLM or the National Institutes of Health.

Learn more: PMC Disclaimer | PMC Copyright Notice



Microorganisms. 2020 Aug; 8(8): 1142.

Published online 2020 Jul 28. doi: 10.3390/microorganisms8081142

PMCID: PMC7466055

PMID: 32731589

#### The Role of Yeasts in Fermentation Processes

Sergi Maicas

#### **Abstract**

In recent years, vessels have been discovered that contain the remains of wine with an age close to 7000 years. It is unclear whether, in ancient times, humans accidentally stumbled across fermented beverages like wine or beer, or was it a product intended as such. What is a fact is that since then, alcoholic beverages have been part of the diet and culture of many of the civilizations that have preceded us. The typical examples of beer and wine are an example of many other drinks resulting from the action of yeasts. In addition to these two beverages, various companies have developed other types of fermented foods and non-alcoholic beverages prepared in a traditional or commercial manner. The climatic conditions, the availability of raw material and the preferences of each region have conditioned and favored the maintenance of some of these products. In addition to the aforementioned traditional alcoholic beverages produced from fruits, berries, or grains, humans use yeast in the production of chemical precursors, global food processing such as coffee and chocolate, or even wastewater processing. Yeast fermentation is not only useful in food manufacturing. Its uses extend to other products of high interest such as the generation of fuel from vegetable sources.

**Keywords:** yeast, non-*Saccharomyces* yeast, wine, beer, beverages

#### 1. Introduction

Fermentation is a well-known natural process used by humanity for thousands of years well undamental purpose of making alcoholic beverages, as well as bread and by-products. Upon a strictly biochemical point of view, fermentation is a process of central metabolism in which an organism converts a carbohydrate, such as starch or sugar, into an alcohol or an acid. For example,

Feedback

yeast performs fermentation to obtain energy by converting sugar into alcohol. Fermentation processes were spontaneously carried out before the biochemical process was fully understood. In the 1850s and 1860s, the French chemist and microbiologist Louis Pasteur became the first scientist to study fermentation, when he demonstrated that this process was performed by living cells. Fermentation processes to produce wines, beers and ciders are traditionally carried out with *Saccharomyces cerevisiae* strains, the most common and commercially available yeast. They are well known for their fermentative behavior and technological characteristics which allow obtaining products of uniform and standard quality. Many other important industrial products are the result of fermentation, such as yogurt, cheese, bread, coffee. Yeasts also play a key role in wastewater treatment or biofuel production. Upon a biochemical point of view, fermentation is carried out by yeasts (and some bacteria) when pyruvate generated from glucose metabolism is broken into ethanol and carbon dioxide (Figure 1).

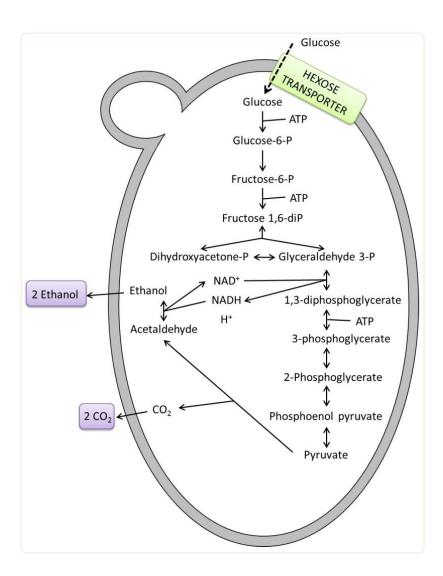


Figure 1

 $Central\ metabolism\ of\ fermentation\ in\ yeasts.$ 

The schematic chemical equation for the production of ethanol from glucose is as follows:

$$C_6H_{12}O_6(glucose) \longrightarrow 2C_2H_5 OH(ethanol) + CO_2(carbon dioxide)$$

Under absence or oxygen-limited conditions, ethanol is produced from acetaldehyde, and two moles of ATP are generated. This is not a fully satisfactory reaction for cells, as they have to consume high amounts of glucose to deliver enough ATP to the system. As a consequence, ethanol is accumulated and when this occurs the fermentative activity is stopped [1].

#### 1.1. Yeasts

Yeasts are eukaryotic microorganisms that live in a wide variety of ecological niches, mainly in water, soil, air and on plant and fruit surfaces. Perhaps the most interesting habitat at this point is the latter, since they directly intervene in the decomposition of ripe fruit and participate in the fermentation process. In this natural environment, yeasts can carry out their metabolism and fermentation activity satisfactorily as they have the necessary nutrients and substrates [2]. On a nutritional level, yeasts are not particularly demanding compared to other microorganisms such as lactic acid bacteria. However, their growth is supported by the existence of basic compounds such as fermentable sugars, amino acids, vitamins, minerals and also oxygen. Upon a morphological point of view, yeasts present a high morphological divergence, with round, ellipsoidal and oval shapes being the most common. In fact, in the identification processes, microscopic evaluation is the first resource followed by other more discriminatory tests such as microbiological and biochemical ones. In a next stage, the classical classification includes other more laborious tests such as those of sugar fermentation and amino acid assimilation [2]. The production and tolerance to ethanol, organic acids and  $SO_2$  are also important tools to differentiate among species. The reproduction of yeasts is mainly by budding, which results in a new and genetically identical cell. Budding is the most common type of asexual reproduction, although cell fission is a characteristic of yeasts belonging to the genus Schizosaccharomyces. Growing conditions that lead to nutrient starvation, such as lack of amino acids, induce sporulation, which is a mechanism used by yeasts to survive in adverse conditions. As a result of sporulation, yeast cells suffer from genetic variability. In industrial fermentation processes, the asexual reproduction of yeasts is advisable to ensure the preservation of the genotype and to maintain stable fermentation behaviour that does not derive from it for as long as possible. At the metabolic level, yeasts are characterised by their capacity to ferment a high spectrum of sugars, among which glucose, fructose, sucrose, maltose and maltotriose predominate, found both in ripe fruit and in processed cereals. In addition, yeasts tolerate acidic environments with pH values around 3.5 or even less. According to technological convenience, yeasts are divided into two large groups namely Saccharomyces and non-Saccharomyces. Morphologically, Saccharomyces yeasts can be round or ellipsoidal in shape depending on the growth phase and cultivation conditions. *S. cerevisiae* is the most studied species and the most utilized in the fermentation of wines and beers due to its satisfactory fermentative capacity, rapid growth and easy adaptation. They tolerate concentrations of SO<sub>2</sub> that normally most non-Saccharomyces yeasts do not survive. However, despite these advantages, it is possible to find in the nature representatives of *S. cerevisiae* that do not necessarily have these characteristics.

#### 1.2. Non-Saccharomyces Yeasts

Non-Saccharomyces yeasts are a group of microorganisms used in numerous fermentation processes, since their high metabolic differences allow the synthesis of different final products. Generally, many of these yeasts capable of modifying the sensory quality of wines are considered as contaminants, so eliminating them or keeping them at low levels was a basic objective in the past [3]. In order to eliminate their activity in wine fermentation, it is usual to disinfect the tanks and fermentation containers using sulfite. This perception has been modified year after year, gaining relevance the action of these yeasts in the spontaneous fermentation, since they contribute positively in the final sensory quality of the wine. These yeasts are the majority in the initial phase of spontaneous fermentation to the point where the concentration of ethanol reaches 4 and 5% v/v. At that point, between alcohol and the exhaustion of dissolved oxygen, their growth is inhibited [4]. When the process is completed, *Saccharomyces* yeasts, the most resistant to ethanol, predominate and complete the fermentation. It has been reported that some non-Saccharomyces yeasts are able to survive toward the end of the spontaneous fermentation and exert their metabolic activity, thus contributing positively to the sensory quality of wines. Based on this evidence, in recent years, many researchers have focused their studies in understanding the nature and fermentative activity of the non-Saccharomyces yeasts [5]. The findings demonstrated the enormous potential of these yeasts for use in the fermentation of traditional and nontraditional beverages. Despite the fact that most non-Saccharomyces yeasts show some technological disadvantages compared to *S. cerevisiae* such as lower fermentative power and production of ethanol, non-Saccharomyces yeasts possess characteristics that in S. cerevisiae are absent, for instance, production of high levels of aromatic compounds such as esters, higher alcohols and fatty acids [6]. In addition, it has been reported that the fermentative activity of these yeasts is manifested in the presence of small amounts of oxygen which leads to an increase in cell biomass and the decrease in ethanol yield, a strategy that can be used to reduce the ethanol content of wines produced in coculture with *S. cerevisiae* [7]. With the aim of exploiting the positive characteristics of non-Saccharomyces yeasts and reducing their negative impact, fermentations with mixed and sequential cultures with *S. cerevisiae* can be performed to produce fermented beverages with different sensory profiles [8]. The most important fact is related to the potential for producing a broad variety of compounds of sensory importance necessary to improve the organoleptic quality of wines and beers. The findings reported so far in literature have led to rethink the role of these yeasts in fermentative processes and to evaluate their use in the development of new products. Among the most studied non-Saccharomyces yeasts that reached special importance for researchers include Candida, Kloeckera, Hanseniaspora, Brettanomyces, Pichia, Lanchacea and Kluyveromyces, among others.

#### 2. Yeast Fermentation Processes

#### 2.1. Alcoholic Fermentations

The production of alcoholic beverages from fermentable carbon sources by yeast is the oldest and most economically important of all biotechnologies. Yeast plays a vital role in the production of all alcoholic beverages. Yeast plays a vital role in the production of all alcoholic beverages and the se-

lection of suitable yeast strains is essential not only to maximise alcohol yield, but also to maintain beverage sensory quality [2].

2.1.1. Wine Fermentation In wine fermentation, strains with specific characteristics are needed, for instance, highly producers of ethanol to reach values of 11-13% v/v, typically found in this beverage. On the other hand, beers and ciders contain less amounts of ethanol with a balanced and distinctive sensory profile characteristic of each one. In recent years, new consuming trends and requirements for new and innovative products have emerged. This situation led to rethink about the existing fermented beverages and to meet the demands of consumers. Yeasts are largely responsible for the complexity and sensory quality of fermented beverages. Based on this, current studies are mainly focused on the search of new type of yeasts with technological application. Non-Saccharomyces yeasts have always been considered contaminants in the manufacture of wine and beer. Therefore, procedures for eliminating them are routinely utilized such as must pasteurization, addition of sulfite and sanitization of equipment and processing halls. In recent years, the negative perception about non-Saccharomyces yeasts has been changing due to the fact that several studies have shown that during spontaneous fermentations of wine, these yeasts play an important role in the definition of the sensory quality of the final product. Based on this evidence, the fermentative behavior of some non-Saccharomyces yeasts is being studied in deep with the purpose of finding the most adequate conditions and the most suitable strain to be utilized in the production of fermented beverages.

2.1.2. Beer Fermentation Beer is the most consumed alcoholic beverage worldwide. It is traditionally made from four key ingredients: malted cereals (barley or other), water, hops, and yeast. Each of these ingredients contributes to the final taste and aroma of beer. During fermentation, yeast cells convert cereal-derived sugars into ethanol and  $CO_2$ . At the same time, hundreds of secondary metabolites that influence the aroma and taste of beer are produced. Variation in these metabolites across different yeast strains is what allows yeast to so uniquely influence beer flavor [9]. Although most breweries use pure yeast cultures for fermentation, spontaneous or mixed fermentation is nowadays used for some specialty beers. These fermentation procedures involve a mix of different yeast species (and bacteria as well) that contribute to the final product sequentially, giving the beer a high degree of complexity. Commonly, breweries have their own stock of selected yeasts for their specific beers. As it is well-known, two types of yeast are used in brewing: *S. cerevisiae* as the top-fermenting yeast to make ales while *S. pastorianus* is a bottom-fermenting yeast used in lager brewing processes [10].

2.1.3. Cider Fermentation Cider is another alcoholic beverage derived from the apple fruit industry, very popular in different countries in the world, mainly Europe, North America, and Australia [11]. Although traditional ciders are produced from spontaneous fermentation of juice carried out by autochthonous yeasts, selected *S. cerevisiae* strains are also commonly used to carry out alcoholic fermentation. This ensures a consistent quality of the finished products [12]. Some other non-*Saccharomyces* yeast species are involved in spontaneous fermentation of apple juice for cider production. However, these yeasts contribute at a lesser extent than *Saccharomyces* and can be producers of off-flavours [13]. Research articles on this type of product are scarce compared to wine, especially in phenomena associated with microbial activities. The microbiome of wine fermentation and its dynamics, the organoleptic improvement of healthy and pleasant products and

the development of starters are now extensively studied. Although the two beverages seem close in terms of microbiome and process (with both alcoholic and malolactic fermentations), the inherent properties of the raw materials and different production and environmental parameters make it worthwhile research on the specificities of apple fermentation. An excellent review of the microbial implications associated with cider production, from ecosystem considerations to associated activities and the influence of process parameters [11].

In addition to these three worldwide-famous fermented beverages, there are many others made from fruit in various countries in Africa, Asia, and Latin America. Although its consumption is local or regional, in some countries drinks made using fruits such as bananas or grapes as raw materials are very popular. The most widespread alcoholic fruit drink in Eastern Africa is banana beer, which in addition to gastronomic interest is especially culturally relevant. Banana beer is a mixed beverage made from bananas and a cereal flour (often sorghum flour) [14]. Dates in North Africa, pineapples and cashew fruits in Latin America and jack fruits in Asia are other of the most relevant products.

#### 2.2. Non-Alcoholic Fermentations

Moreover, yeast can act in the fermentation of global non-alcoholic products (bread, chocolate or coffee, beverages such as kefir, sodas, lemonades, and vinegar or even biofuels and other chemicals.

2.2.1. Bread Fermentation The fermentation of the dough made by the yeasts is the most critical phase in the making of bread. The fermentative yield of yeast cells during this fermentation is crucial and determines the final quality of the bread. Yeasts not only produce  $CO_2$  and other metabolites that influence the final appearance of the dough, volume, and texture, and of course, the taste of the bread. The yeast strain, pregrowth conditions, its activity during the dough fermentation process, the fermentation conditions, as well as the dough ingredients are basic to control the process. The fermentation rate is also conditioned by the ingredients of the dough, including the amounts of sugar and salt used in its preparation. Commercial bread producers currently produce various types of dough such as lean, sweet or frozen dough. Depending on the type of dough, and to obtain optimal fermentation rates, it is recommended to use suitable yeast strains with specific phenotypic traits  $[\underline{15}]$ .

2.2.2. Coffee Fermentation Yeasts play an important role in coffee production, in the post-harvest phase. Its performance can be done in two phases. On the one hand, aerobically, in which the berries just collected are deposited in a tank and the yeasts are allowed to act. This process is carried out under control of basic parameters, such as time and temperature. Alternatively, coffee berries are deposited in a container mixed with water and microorganisms are allowed to act anaerobically (in the absence of oxygen). This second process is more homogeneous and easy to control than the aerobic. Sometimes, coffee beans are even fermented in a mixed process, first in an aerobic and finally anaerobic manner [16]. To develop these processes in a satisfactory manner, and to preserve/improve the organoleptic properties of coffee, refine its sweetness, control acidity, give them body or add sensory notes (chocolate, caramel, fruits) mucilage should be re-

moved. The process is naturally carried out by the yeasts present in the mixture, although the process can be improved by the addition of appropriate enzymes (polygalacturonase, pectin lyase, pectin methylesterase) [17].

2.2.3. Chocolate Fermentation Raw cacao beans have a bitter and astringent taste, because of high phenolic content. Anthocyanins are one group of these polyphenols, and it both contributes to astringency and provide the reddish-purple color. Fermentation allows the enzymatic breakdown of proteins and carbohydrates inside the bean, creating flavor development. This is aided by microbial fermentation, which create the perfect environment through the fermentation of the cacao pulp surrounding the beans. This processing step enables the extraction of flavor from cacao and contributes to the final acidity of the final product. Yeasts (and also bacteria) ferment the juicy pulp among the cacao beans by different methods, generally following a an anaerobic phase and an aerobic phase. During the anaerobic phase, the sugars of the pulp (sucrose, glucose, fructose) are consumed by yeasts using anaerobic respiration to yield carbon dioxide, ethanol, and low amounts of energy [18,19]. The aerobic stage is dominated by lactic and acetic-acid-producing bacteria [20].

#### 2.3. Not Only Food: Biofuels and Other Chemicals

The fermentation processes of substrates such as xylose are also of high interest on an industrial level. In addition to expanding the range of substrates that can be used for this purpose, they allow the environmental cost of efficient production of biofuels and other advanced chemicals to be reduced. Some interesting approaches have been made in biorefinery to reprogram yeast for use in these bioprocesses [21,22,23].

#### 3. Special Issue on "Yeast Fermentation"

This issue in *Microorganisms* aims to contribute to the update of knowledge regarding yeasts, regarding both basic and also applied aspects. Among the great contributions to this issue we have a manuscript devoted to the brewing industry and the recent isolation of the yeast *Saccharomyces* eubayanus [24]. The use of headspace solid-phase microextraction followed by gas chromatography-mass spectrometry (HS-SPME-GC-MS) has contributed to the production of volatile compounds in wild strains and to compare them to a commercial yeast. All these findings highlight the potentiality of this yeast to produce new varieties of beers. Haile et al. [17] have explored the possibility to identify and select pectinolytic yeasts that have potential use as a starter culture for coffee fermentation. Almost 30 isolates, eight of them with the ability to produce pectinase enzymes were identified and confirmed by using molecular biology techniques. A helpful bioinformatics tool (MEGA 6) was also used to generate phylogenetic trees able to determine the evolutionary relationship of yeasts obtained from their experiments. Biofuel production by recombinant Saccharomyces cerevisiae strains with essential genes and metabolic networks for xylose metabolism has been also reported [23]. The authors have shown that the deletion of cAMP phosphodiesterase genes PDE1 and PDE2 can increase xylose utilization. Moreover, the door is opened to provide new targets for engineering other xylose-fermenting strains. The utilization of xylose, the second most abundant sugar component in the hydrolysates of lignocellulosic materials, is a relevant issue. Understanding the relationship between xylose and the metabolic regulatory systems

in yeasts is a crucial aspects where hexokinase 2 (Hxk2p) is involved [25]. All of these processes can be damaged if contaminated. Because most fermentation substrates are not sterile, contamination is always a factor to consider. With a very interesting approach, a genetically modified strain of *Komagataella phaffii* yeast was used for the use of glycerol as a base substance in lactate production. Polyactide, a bioplastic widely used in the pharmaceutical, automotive, packaging and food industries was produced. The disruption of the gene encoding arabitol dehydrogenase (ArDH) was achieved, which improves the production of lactic acid by *K. phaffii* as a biocatalyst [26]. Seo et al. [27] have developed and proposed alternative solutions to control contamination. This review includes information on industrial uses of yeast fermentation, microbial contamination and its effects on yeast fermentations. Finally, they describe strategies for controlling microbial contamination.

# Acknowledgments

Thanks to all the authors and reviewers for their excellent contributions to this Special Issue. Additional thanks to the Microorganisms Editorial Office for their professional assistance and continuous support.

#### Conflicts of Interest

The editors declares no conflict of interest.

#### References

- 1. Puligundla P., Smogrovicova D., Obulam V.S.R., Ko S. Very high gravity (VHG) ethanolic brewing and fermentation: A research update. *J. Ind. Microbiol. Biotechnol.* 2011;38:1133–1144. doi: 10.1007/s10295-011-0999-3. [PubMed] [CrossRef] [Google Scholar]
- 2. Walker G.M., Stewart G.G. Saccharomyces cerevisiae in the Production of Fermented Beverages. *Beverages*. 2016;2:30. doi: 10.3390/beverages2040030. [CrossRef] [Google Scholar]
- 3. Ciani M., Maccarelli F. Oenological properties of non-Saccharomyces yeasts associated with wine-making. *World J. Microbiol. Biotechnol.* 1997;14:199–203. doi: 10.1023/A:1008825928354. [CrossRef] [Google Scholar]
- 4. Grangeteau C., Gerhards D., Terrat S., Dequiedt S., Alexandre H., Guilloux-Benatier M., von Wallbrunn C., Rousseaux S. FT-IR spectroscopy: A powerful tool for studying the inter- and intraspecific biodiversity of cultivable non-Saccharomyces yeasts isolated from grape must. *J. Microbiol. Methods.* 2016;121:50–58. doi: 10.1016/j.mimet.2015.12.009. [PubMed] [CrossRef] [Google Scholar]
- 5. Estela-Escalante W., Moscosa-Santillán M., González-RamÃrez J., Rosales-Mendoza S. Evaluation of the potential production of ethanol by Candida zemplinina yeast with regard to beer fermentation. *J. Am. Soc. Brew. Chem.* 2017;75:130–135. doi: 10.1094/ASBCJ-2017-2532-01. [CrossRef] [Google Scholar]
- 6. Cordero-Bueso G., Esteve-Zarzoso B., Cabellos J., Gil-DÃaz M., Arroyo T. Biotechnological potential of non-Saccharomyces yeasts isolated during spontaneous fermentations of Malvar (*Vitis vinifera* cv. L.) *Eur. Food Res. Technol.* 2013;236:193–207. doi: 10.1007/s00217-012-1874-9. [CrossRef] [Google Scholar]

- 7. Ciani M., Capece A., Comitini F., Canonico L., Siesto G., Romano P. Yeast interactions in inoculated wine fermentation. *Front. Microbiol.* 2016;7 doi: 10.3389/fmicb.2016.00555. [PMC free article] [PubMed] [CrossRef] [Google Scholar]
- 8. Canonico L., Agarbati A., Comitini F., Ciani M. Torulaspora delbrueckii in the brewing process: A new approach to enhance bioflavour and to reduce ethanol content. *Food Microbiol.* 2016;56:45–51. doi: 10.1016/j.fm.2015.12.005. [PubMed] [CrossRef] [Google Scholar]
- 9. Dzialo M.C., Park R., Steensels J., Lievens B., Verstrepen K.J. Physiology, ecology and industrial applications of aroma formation in yeast. *FEMS Microbiol. Rev.* 2017;41:S95–S128. doi: 10.1093/femsre/fux031. [PMC free article] [PubMed] [CrossRef] [Google Scholar]
- 10. Libkind D., Hittinger C.T., Valério E., Gonçalves C., Dover J., Johnston M., Gonçalves P., Sampaio J.P. Microbe domestication and the identification of the wild genetic stock of lager-brewing yeast. *Proc. Natl. Acad. Sci. USA.* 2011;108:14539–14544. doi: 10.1073/pnas.1105430108. [PMC free article] [PubMed] [CrossRef] [Google Scholar]
- 11. Cousin F.J., Le Guellec R., Schlusselhuber M., Dalmasso M., Laplace J.M., Cretenet M. Microorganisms in fermented apple beverages: Current knowledge and future directions. *Microorganisms*. 2017;5:39. doi: 10.3390/microorganisms5030039.

  [PMC free article] [PubMed] [CrossRef] [Google Scholar]
- 12. Lorenzini M., Simonato B., Slaghenaufi D., Ugliano M., Zapparoli G. Assessment of yeasts for apple juice fermentation and production of cider volatile compounds. *LWT.* 2019;99:224–230. doi: 10.1016/j.lwt.2018.09.075. [CrossRef] [Google Scholar]
- 13. Pando Bedriñana R., Querol Simón A., Suárez Valles B. Genetic and phenotypic diversity of autochthonous cider yeasts in a cellar from Asturias. *Food Microbiol.* 2010;27:503–508. doi: 10.1016/j.fm.2009.11.018. [PubMed] [CrossRef] [Google Scholar]
- 14. Gensi R., Kyamuhangire W., Carasco J. Traditional production method and storage characteristics for banana beer (tonto) in Uganda. *Acta Hortic*. 2000;540:569–574. doi: 10.17660/ActaHortic.2000.540.62. [CrossRef] [Google Scholar]
- 15. Struyf N., Van der Maelen E., Hemdane S., Verspreet J., Verstrepen K., Courtin C. Bread Dough and Baker's Yeast: An Uplifting Synergy. *Compr. Rev. Food Sci. Food Saf.* 2017;16:850–867. doi: 10.1111/1541-4337.12282. [PubMed] [CrossRef] [Google Scholar]
- 16. de Melo Pereira G., Soccol V., Pandey A., Medeiros A., Andrade Lara J., Gollo A., Soccol C. Isolation, selection and evaluation of yeasts for use in fermentation of coffee beans by the wet process. *Int. J. Food Microbiol.* 2014;188:60–66. doi: 10.1016/j.ijfoodmicro.2014.07.008. [PubMed] [CrossRef] [Google Scholar]
- 17. Haile M., Kang W.H. Isolation, Identification, and Characterization of Pectinolytic Yeasts for Starter Culture in Coffee Fermentation. *Microorganisms*. 2019;7:401. doi: 10.3390/microorganisms7100401. [PMC free article] [PubMed] [CrossRef] [Google Scholar]
- 18. Ho V., Zhao J., Fleet G. Yeasts are essential for cocoa bean fermentation. *Int. J. Food Microbiol.* 2014;174:72–87. doi: 10.1016/j.ijfoodmicro.2013.12.014. [PubMed] [CrossRef] [Google Scholar]
- 19. Papalexandratou Z., Nielsen D. It's gettin' hot in here: Breeding robust yeast starter cultures for cocoa fermentation. *Trends Microbiol.* 2016;24:168–170. doi: 10.1016/j.tim.2016.01.003. [PubMed] [CrossRef] [Google Scholar]
- 20. Ho V., Fleet G., Zhao J. Unravelling the contribution of lactic acid bacteria and acetic acid bacteria to cocoa fermentation using inoculated organisms. *Int. J. Food Microbiol.* 2018;279:43–56. doi: 10.1016/j.ijfoodmicro.2018.04.040. [PubMed] [CrossRef] [Google Scholar]

- 21. Kwak S., Jo J.H., Yun E.J., Jin Y.S., Seo J.H. Production of biofuels and chemicals from xylose using native and engineered yeast strains. *Biotechnol. Adv.* 2019;37:271–283. doi: 10.1016/j.biotechadv.2018.12.003. [PubMed] [CrossRef] [Google Scholar]
- 22. Jansen M., Bracher J., Papapetridis I., Verhoeven M., de Bruijn H., de Waal P., van Maris A., Klaassen P., Pronk J. *Saccharomyces cerevisiae* strains for second-generation ethanol production: From academic exploration to industrial implementation. *FEMS Yeast Res.* 2017;17 doi: 10.1093/femsyr/fox044. [PMC free article] [PubMed] [CrossRef] [Google Scholar]
- 23. Wu M., Li H., Wei S., Wu H., Wu X., Bao X., Hou J., Liu W., Shen Y. Simulating Extracellular Glucose Signals Enhances Xylose Metabolism in Recombinant Saccharomyces cerevisiae. *Microorganisms*. 2020;8:100. doi: 10.3390/microorganisms8010100. [PMC free article] [PubMed] [CrossRef] [Google Scholar]
- 24. Urbina K., Villarreal P., Nespolo R.F., Salazar R., Santander R., Cubillos F.A. Volatile Compound Screening Using HS-SPME-GC/MS on Saccharomyces eubayanus Strains under Low-Temperature Pilsner Wort Fermentation. *Microorganisms*. 2020;8:755. doi: 10.3390/microorganisms8050755. [PMC free article] [PubMed] [CrossRef] [Google Scholar]
- 25. Zheng L., Wei S., Wu M., Zhu X., Bao X., Hou J., Liu W., Shen Y. Improving Xylose Fermentation in Saccharomyces cerevisiae by Expressing Nuclear-Localized Hexokinase 2. *Microorganisms*. 2020;8:856. doi: 10.3390/microorganisms8060856. [PMC free article] [PubMed] [CrossRef] [Google Scholar]
- 26. Tamires Moreira Melo N., Pontes G.C., Procópio D.P., Cunha G., Eliodório K.P., Paes H.C., Basso T.O., Parachin N.S. Evaluation of Product Distribution in Chemostat and Batch Fermentation in Lactic Acid-Producing Komagataella phaffii Strains Utilizing Glycerol as Substrate. *Microorganisms*. 2020;8:781. doi: 10.3390/microorganisms8050781. [PMC free article] [PubMed] [CrossRef] [Google Scholar]
- 27. Seo S.O., Park S.K., Jung S.C., Ryu C.M., Kim J.S. Anti-Contamination Strategies for Yeast Fermentations. *Microorganisms*. 2020;8:274. doi: 10.3390/microorganisms8020274. [PMC free article] [PubMed] [CrossRef] [Google Scholar]

# **Ethanol From Cellulose: A General Review**

P.C. Badger

#### INTRODUCTION

The use of ethanol as an alternative motor fuel has been steadily increasing around the world for a number of reasons. Domestic production and use of ethanol for fuel can decrease dependence on foreign oil, reduce trade deficits, create jobs in rural areas, reduce air pollution, and reduce global climate change carbon dioxide buildup. Ethanol, unlike gasoline, is an oxygenated fuel that contains 35% oxygen, which reduces particulate and NOx emissions from combustion.

Ethanol can be made synthetically from petroleum or by microbial conversion of biomass materials through fermentation. In 1995, about 93% of the ethanol in the world was produced by the fermentation method and about 7% by the synthetic method. The fermentation method generally uses three steps: (1) the formation of a solution of fermentable sugars, (2) the fermentation of these sugars to ethanol, and (3) the separation and purification of the ethanol, usually by distillation.

#### SUGAR FEEDSTOCKS

Fermentation involves microorganisms that use the fermentable sugars for food and in the process produces ethanol and other byproducts. These microorganisms can typically use the 6-carbon sugars, one of the most common being glucose. Therefore, biomass materials containing high levels of glucose or precursors to glucose are the easiest to convert to ethanol. However, since sugar materials are in the human food chain, these materials are usually too expensive to use for ethanol production.

One example of a sugar feedstock is sugarcane. Brazil developed a successful fuel ethanol program from sugarcane for a number of reasons: (1) Brazil traditionally relied heavily on imported oil for transportation fuels, which caused a severe economic drain on the country; (2) Brazil can attain very high yields of sugarcane; and (3) Brazil has also experienced periods of poor sugar markets. As a result, the Brazilian government established programs supportive of the industry with the result that Brazil has been able to successfully produce and use sugarcane for fuel ethanol production.

Although fungi, bacteria, and yeast microorganisms can be used for fermentation, a specific yeast (*Saccharomyces cerevisiae* also known as Bakers' yeast, since it is commonly used in the baking industry) is frequently used to ferment glucose to ethanol. Theoretically, 100 grams of glucose will produce 51.4 g of ethanol and 48.8 g of carbon dioxide. However, in practice, the microorganisms use some of the glucose for growth and the actual yield is less than 100%.

Other biomass feedstocks rich in sugars (materials known as saccharides) include sugar beet, sweet sorghum, and various fruits. However, these materials are all in the human food chain and, except for some processing residues are generally too expensive to use for fuel ethanol production.

#### STARCHY FEEDSTOCKS

Another potential ethanol feedstock is starch. Starch molecules are made up of long chains of glucose molecules. Thus, starchy materials can also be fermented after breaking starch molecules into simple glucose molecules. Examples of starchy materials commonly used around the world for ethanol production include cereal grains, potato, sweet potato, and cassava. Cereal grains commonly used in the US for ethanol production include maize and wheat.

Approximately 475 million tonnes of maize were produced in the world in 1990 with about 200 million t produced in the US. Approximately 8 to 9 million t, or 4% of US maize grain went into ethanol in 1990. A bushel of maize grain (25.3 kg or 56 lb. at 15% moisture) can produce from 9.4 to 10.9 L (2.5 to 2.9 gallons) of pure ethanol, depending on the technology used.

Starchy materials require a reaction of starch with water (hydrolysis) to break down the starch into fermentable sugars (saccharification). Typically, hydrolysis is performed by mixing the starch with water to

Trends in New Crops and New Uses

form a slurry which is then stirred and heated to rupture the cell walls. Specific enzymes that will break the chemical bonds are added at various times during the heating cycle.

#### CELLULOSIC FEEDSTOCKS

Like sugar materials, starchy materials are also in the human food chain and are thus expensive. Fortunately, a third alternative exists—cellulosic materials. Examples of cellulosic materials are paper, cardboard, wood, and other fibrous plant material.

Cellulosic resources are in general very widespread and abundant. For example, forests comprise about 80% of the world's biomass. Being abundant and outside the human food chain makes cellulosic materials relatively inexpensive feedstocks for ethanol production.

Cellulosic materials are comprised of lignin, hemicellulose, and cellulose and are thus sometimes called lignocellulosic materials. One of the primary functions of lignin is to provide structural support for the plant. Thus, in general, trees have higher lignin contents then grasses. Unfortunately, lignin which contains no sugars, encloses the cellulose and hemicellulose molecules, making them difficult to reach.

Cellulose molecules consist of long chains of glucose molecules as do starch molecules, but have a different structural configuration. These structural characteristics plus the encapsulation by lignin makes cellulosic materials more difficult to hydrolyze than starchy materials.

Hemicellulose is also comprised of long chains of sugar molecules; but contains, in addition to glucose (a 6-carbon or hexose sugar), contains pentoses (5-carbon sugars). To complicate matters, the exact sugar composition of hemicellulose can vary depending on the type of plant.

Since 5-carbon sugars comprise a high percentage of the available sugars, the ability to recover and ferment them into ethanol is important for the efficiency and economics of the process. Recently, special microorganisms have been genetically engineered which can ferment 5-carbon sugars into ethanol with relatively high efficiency.

One example is a genetically engineered microorganism developed by the University of Florida that has the ability to ferment both 5- and 6-carbon sugars. This microorganism was issued US patent 5,000,000. Other researchers have developed microorganisms with the ability to efficiently ferment at least part of the sugars present.

Bacteria have drawn special attention from researchers because of their speed of fermentation. In general, bacteria can ferment in minutes as compared to hours for yeast.

#### ETHANOL-FROM-CELLULOSE

In times of fuel shortages, fermentation ethanol has been commercially manufactured in the US from cellulosic biomass feedstocks using acid hydrolysis techniques. Currently, some countries in locations with higher ethanol and fuel prices, are producing ethanol from cellulosic feedstocks. However, it is only recently that cost-effective technologies for producing ethanol-from-cellulose (EFC) in the US have started to emerge.

There are three basic types of EFC processes—acid hydrolysis, enzymatic hydrolysis, and thermochemical—with variations for each. The most common is acid hydrolysis. Virtually any acid can be used; however, sulfuric acid is most commonly used since it is usually the least expensive.

#### ACID HYDROLYSIS

There are two basic types of acid processes: dilute acid and concentrated acid, each with variations. Dilute acid processes are conducted under high temperature and pressure, and have reaction times in the range of seconds or minutes, which facilitates continuous processing.

As an example, using a dilute acid process with 1% sulfuric acid in a continuous flow reactor at a residence time of 0.22 minutes and a temperature of 237°C (458°F) with pure cellulose provided a yield over 50% sugars. In this case, 0.9 t (1 ton) of dry wood would yield about 189 L (50 gallons) of pure ethanol. The combination of acid and high temperature and pressure dictate special reactor materials, which can make the reactor expensive.

Most dilute acid processes are limited to a sugar recovery efficiency of around 50%. The reason for this is that at least two reactions are part of this process. The first reaction converts the cellulosic materials to

sugar and the second reaction converts the sugars to other chemicals. Unfortunately, the conditions that cause the first reaction to occur also are the right conditions for the second to occur. Thus, once the cellulosic molecules are broken apart, the reaction proceeds rapidly to break down the sugars into other products—most notably furfural, a chemical used in the plastics industry. Not only does sugar degradation reduce sugar yield, but the furfural and other degradation products can be poisonous to the fermentation microorganisms.

The biggest advantage of dilute acid processes is their fast rate of reaction, which facilitates continuous processing. Their biggest disadvantage is their low sugar yield. For rapid continuous processes, in order to allow adequate acid penetration, feedstocks must also be reduced in size so that the maximum particle dimension is in the range of a few millimeters.

Since 5-carbon sugars degrade more rapidly than 6-carbon sugars, one way to decrease sugar degradation is to have a two-stage process. The first stage is conducted under mild process conditions to recover the 5-carbon sugars while the second stage is conducted under harsher conditions to recover the 6-carbon sugars. Unfortunately, sugar degradation is still a problem and yields are limited to around 272 L/t (80 gallons of ethanol/ton) of dry wood.

The concentrated acid process uses relatively mild temperatures and the only pressures involved are usually only those created by pumping materials from vessel to vessel. One concentrated acid process was first developed by USDA and further refined by Purdue University and the Tennessee Valley Authority.

In the TVA concentrated acid process, corn stover is mixed with dilute (10%) sulfuric acid, and heated to 100°C for 2 to 6 hours in the first (or hemicellulose) hydrolysis reactor. The low temperatures and pressures minimize the degradation of sugars. To recover the sugars, the hydrolyzed material in the first reactor is soaked in water and drained several times.

The solid residue from the first stage is then dewatered and soaked in a 30% to 40% concentration of sulfuric acid for 1 to 4 hr as a pre-cellulose hydrolysis step. This material is then dewatered and dried with the effect that the acid concentration in the material is increased to about 70%. After reacting in another vessel for 1 to 4 hr at 100°C, the reactor contents are filtered to remove solids and recover the sugar and acid. The sugar/acid solution from the second stage is recycled to the first stage to provide the acid for the first stage hydrolysis. The sugars from the second stage hydrolysis are thus recovered in the liquid from the first stage hydrolysis.

The primary advantage of the concentrated process is the high sugar recovery efficiency, which can be on the order of over 90% of both hemicellulose and cellulose sugars. The low temperatures and pressures employed also allow the use of relatively low cost materials such as fiberglass tanks and piping. Unfortunately, it is a relatively slow process and cost effective acid recovery systems have been difficult to develop. Without acid recovery, large quantities of lime must be used to neutralize the acid in the sugar solution. This neutralization forms large quantities of calcium sulfate, which requires disposal and creates additional expense.

Using some assumed cellulose conversion and fermentation efficiencies, ethanol yields from glucose can be calculated for corn stover (the above-ground part of the corn plant less the ears) as shown in Table 1 showing ethanol yield from glucose. Similarly, ethanol yields from the xylose can be calculated as shown in Table 2.

Thus, in this example, the total yield/t of dry stover is about 227 L (60 gallons) of ethanol. These numbers also show how critical sugar conversion and recovery efficiencies and fermentation efficiencies are. If one could attain 95% for both efficiencies, then the yield would be approximately 350 L/t (103 gallons of ethanol/ton).

**Table 1.** Ethanol yield from glucose.

Dry stover	1 tonne (1000 kg)
Cellulose content	× 0.45
Cellulose conversion and recovery efficiency	$\times 0.76$
Ethanol stoichiometric yield	× 0.51
Glucose fermentation efficiency	$\times 0.75$
Yield from glucose	131 kg ethanol = 151 L (40 gallons)

**Table 2.** Ethanol yield from xylose.

Dry stover	1 tonne (1000 kg)
Hemicellulose content	× 0.29
Hemicellulose conversion and recovery efficiency	× 0.90
Ethanol stoichiometric yield	× 0.51
Xylose fermentation efficiency	× 0.50
Yield from xylose	66  kg ethanol = 76  L  (20  gallons)

#### **ENZYMATIC HYDROLYSIS**

Another basic method of hydrolysis is enzymatic hydrolysis. Enzymes are naturally occurring plant proteins that cause certain chemical reactions to occur. However, for enzymes to work, they must obtain access to the molecules to be hydrolyzed. For enzymatic processes to be effective, some kind of pretreatment process is thus needed to break the crystalline structure of the lignocellulose and remove the lignin to expose the cellulose and hemicellulose molecules. Depending on the biomass material, either physical or chemical pretreatment methods may be used.

Physical methods may use high temperature and pressure, milling, radiation, or freezing—all of which require high-energy consumption. The chemical method uses a solvent to break apart and dissolve the crystal-line structure.

An example of an enzymatic hydrolysis-based process is under development by the National Renewable Energy Laboratory (NREL). After a dilute acid pretreatment, the slurry is detoxified to remove materials that would be poisonous to the microorganisms used in the process. A small part of this slurry is sent to a separate vessel that is used to grow microorganisms that produce the cellulase enzyme for the process. Another part of the slurry is sent to another vessel to maintain and grow a yeast culture for fermentation. In the NREL process, both enzymes and the fermentation microorganisms are added at the same time to the slurry, and sugar conversion and fermentation occur simultaneously in a process called simultaneous saccharification and cofermentation (SSCF).

Due to the tough crystalline structure, the enzymes currently available require several days to achieve good results. Since long process times tie up reactor vessels for long periods, these vessels have to either be quite large or many of them must be used. Either option is expensive. Currently the cost of enzymes is also too high and research is continuing to bring down the cost of enzymes.

However, if less expensive enzymes can be developed enzymatic processes hold several advantages: (1) their efficiency is quite high and their byproduct production can be controlled; (2) their mild process conditions do not require expensive materials of construction; and (3) their process energy requirements are relatively low.

#### THERMOCHEMICAL PROCESSES

There are two ethanol production processes that currently employ thermochemical reactions in their processes. The first system is actually a hybrid thermochemical and biological system. An example is a process under development by Bioengineering Resources in Fayetteville, Arkansas. Biomass materials are first thermochemically gasified and the synthesis gas (a mixture of hydrogen and carbon oxides) bubbled through specially designed fermenters. A microorganism that is capable of converting the synthesis gas is introduced into the fermenters under specific process conditions to cause fermentation to ethanol.

The second thermochemical ethanol production process does not use any microorganisms. In this process, biomass materials are first thermochemically gasified and the synthesis gas passed through a reactor containing catalysts, which cause the gas to be converted into ethanol. An intensive effort was made by Germany in World War II to develop these processes for fuel. Numerous efforts have been made since then to develop commercially viable thermochemical-to-ethanol processes.

Ethanol yields up to 50% have been obtained using synthesis gas-to-ethanol processes. Some processes that first produce methanol and then use catalytic shifts to produce ethanol have obtained ethanol yields in the

**Table 3.** A partial listing of companies developing ethanol-from-cellulose technologies.

Company & headquarters location	Technology	Primary feedstock	Ethanol capacity	Comments
BCI, Dedham, MA	Dilute acid	Bagasse	7560 million L/yr (20 million gpy²)	Plant to break ground in 2002
Bioengineering Resources, Fayetteville, AR	Thermochemical gasification with fermentation			Pilot plant operating
Ethxx International, Aurora, ON	Thermochemical gasification with catalytic conversion	Wood		Pilot plant operating
Fuel Cell Energy, Lakewood, CO	Thermochemical gasification with catalytic conversion	Wood		Pilot plant operating
Iogen, Ottawa, ON	Enzymatic	Oat hulls, switchgrass, wheat straw, and corn stover	378 million L/yr (1 million gpy)	Experimental plant operating
Masada, Birmingham, AL	Concentrated acid	MSW	3780 million L/yr (10 million gpy)	Plant to break ground early 2002
Paszner Technologies, Inc, Surrey, BC	Acidified aqueous acetone process	Wood		Commercial plants under construction
PureVision Technology, Ft. Lupton, CO	Enzymatic	Wood		Constructing pilot plant

<sup>&</sup>lt;sup>z</sup>gpy=gallons per year

range of 80%. Unfortunately, like the other processes, finding a cost-effective all-thermochemical process has been difficult.

#### **COMMERCIALIZATION EFFORTS**

Several EFC plants were built and operated in various countries in World War II, when wartime conditions changed economic conditions and priorities. These countries included Germany, Russia, China, Korea, Switzerland, the United States, and other countries. Today, due to competition from synthetically produced ethanol, only a few of these plants are still operating with virtually all of them in Russia.

A paper manufacturing plant in Temi-schammaig, Quebec, operates off of byproduct sugars contained in "sulfite liquor," which contains about 2% fermentable sugars. This is the only facility of its kind in North America. This facility is operated by Tembec, Inc., and produces 4 million gallons per year of industrial grade ethanol.

Several efforts are underway in North America to commercially produce ethanol from wood and other cellulosic materials as a primary product. Table 3 partially summarizes these companies and their activities, which are in various states of progress.

#### **SUMMARY**

Ethanol-from-cellulose (EFC) holds great potential due to the widespread availability, abundance, and relatively low cost of cellulosic materials. However, although several EFC processes are technically feasible, cost-effective processes have been difficult to achieve. Only recently have cost-effective EFC technologies begun to emerge.

This	vs.	That	Compare
			/

# Fractional Distillation vs. Refining

# What's the Difference?

Fractional distillation and refining are both processes used in the petroleum industry to separate and purify crude oil. Fractional distillation is a physical separation technique that utilizes the different boiling points of the components in crude oil to separate them. It involves heating the crude oil and collecting the vapor at different temperature ranges. On the other hand, refining is a chemical process that involves treating the crude oil to remove impurities and improve the quality of the end products. It includes processes like desalting, catalytic cracking, and hydrotreating. While fractional distillation focuses on separating the different components of crude oil, refining aims to enhance the quality and value of the final petroleum products.

Copy This URL

# Comparison

Attribute	Fractional Distillation	Refining
Process	Separation of a mixture into its components based on boiling points	Purification of crude oil to obtain usable products
Application	Used in the petroleum industry to separate crude oil into different fractions	Used in the oil industry to purify crude oil and obtain valuable products like gasoline, diesel, etc.
Objective	To separate a mixture into its individual components based on boiling points	To remove impurities and obtain usable products from crude oil
Boiling Points	Components are separated based on their different boiling points	Impurities and different hydrocarbon fractions have different boiling points
Process Complexity	Relatively simple process	Complex process involving multiple steps and techniques

Equipment	Uses a fractionating column and condensers	Requires various equipment such as distillation towers, heat exchangers, etc.
Products	Produces different fractions of hydrocarbons like gasoline, diesel, kerosene, etc.	Produces usable products like gasoline, diesel, jet fuel, lubricants, etc.
Impurities	Does not focus on removing impurities	Focuses on removing impurities like sulfur, nitrogen, metals, etc.

# **Further Detail**

#### Introduction

Fractional distillation and refining are two essential processes in the petroleum industry. Both techniques play a crucial role in separating and purifying crude oil to obtain valuable products such as gasoline, diesel, and various petrochemicals. While they share similarities in terms of their objectives, there are distinct differences in their methods and applications. This article aims to explore and compare the attributes of fractional distillation and refining, highlighting their significance in the oil refining process.

#### **Fractional Distillation**

Fractional distillation is a physical separation process used to separate a mixture of different components based on their boiling points. It is primarily employed in the initial stages of crude oil refining. The process begins by heating the crude oil in a distillation column, which consists of several trays or plates. As the temperature increases, the crude oil vaporizes, and the vapors rise through the column. The column is designed in such a way that the temperature decreases gradually from the bottom to the top.

As the vapors rise, they encounter trays or plates at different temperatures. Each tray or plate represents a specific temperature range, allowing the separation of different components. The components with lower boiling points, such as gasoline and other light hydrocarbons, vaporize at lower temperatures and rise to the top of the column. In contrast, components with higher boiling points, such as heavy oils and bitumen, remain in the liquid state and collect at the bottom of the column.

The separated components are then collected and further processed to obtain various petroleum products. For instance, the gasoline vapors are condensed and collected as a liquid, while the heavier fractions are sent for additional refining processes. Fractional distillation is a crucial step in the refining process as it enables the initial separation of crude oil into its different components, which can then be further processed to obtain valuable products.

# Refining

Refining, also known as secondary processing, involves the conversion of the separated crude oil components obtained through fractional distillation into more valuable products. This process aims to remove impurities, improve the quality, and enhance the performance characteristics of the petroleum products. Refining encompasses various techniques such as catalytic cracking, hydrocracking, hydrotreating, and reforming.

Catalytic cracking is a refining process that breaks down heavy hydrocarbon molecules into lighter ones by using a catalyst. This technique is employed to produce gasoline and other light products from heavier fractions. Hydrocracking, on the other hand, involves the use of hydrogen and a catalyst to break down heavy hydrocarbons into lighter ones. It is particularly useful in converting heavy oils into diesel and jet fuel.

Hydrotreating is a refining process that removes impurities, such as sulfur and nitrogen compounds, from petroleum products. This technique helps in reducing the environmental impact of the fuels and improving their combustion properties. Reforming is another refining process that involves rearranging the molecular structure of hydrocarbons to enhance their octane rating, making them suitable for gasoline production.

Overall, refining plays a crucial role in transforming the separated crude oil components into valuable products with improved quality, performance, and environmental characteristics. It complements the initial separation achieved through fractional distillation and ensures that the final petroleum products meet the required specifications and standards.

## Comparison

While both fractional distillation and refining are integral parts of the petroleum refining process, they differ in terms of their objectives, methods, and applications. Fractional distillation focuses on the initial separation of crude oil into its different components based on their boiling points. It is a physical separation process that relies on the differences in boiling points to achieve separation.

On the other hand, refining encompasses various chemical processes that aim to convert the separated crude oil components into more valuable products. It involves techniques such as catalytic cracking, hydrocracking, hydrotreating, and reforming, which modify the molecular structure of the hydrocarbons to improve their quality and performance characteristics.

While fractional distillation is primarily employed in the early stages of crude oil refining, refining processes occur after the initial separation to further process the separated components. Fractional distillation separates the crude oil into different fractions, such as gasoline, diesel, and heavy oils, while refining processes convert these fractions into specific products with desired properties.

Another difference lies in the equipment used for each process. Fractional distillation relies on a distillation column with trays or plates at different temperatures to achieve separation. The column is designed to gradually decrease the temperature from the bottom to the top, allowing the separation of components based on their boiling points.

In contrast, refining processes involve various reactors, catalysts, and other equipment specific to each technique. For example, catalytic cracking requires a reactor vessel and a catalyst to break down heavy hydrocarbons, while hydrotreating involves reactors and hydrogen gas to remove impurities from the petroleum products.

#### Conclusion

Fractional distillation and refining are two essential processes in the petroleum industry. Fractional distillation enables the initial separation of crude oil into its different components based on their boiling points, while refining processes further convert these components into valuable products with improved quality and performance characteristics.

While fractional distillation relies on physical separation, refining involves various chemical processes such as catalytic cracking, hydrocracking, hydrotreating, and reforming. These processes

modify the molecular structure of the hydrocarbons to enhance their properties and meet the required specifications.

Both processes are crucial in the overall refining of crude oil and play complementary roles in obtaining the desired petroleum products. Fractional distillation sets the foundation by separating the crude oil into fractions, while refining processes add value by converting these fractions into specific products suitable for various applications.

Understanding the attributes and differences between fractional distillation and refining is essential for the efficient and effective production of high-quality petroleum products that meet the demands of various industries and consumers.

Comparisons may contain inaccurate information about people, places, or facts. Please report any issues.

# **Featured**

Female Bones vs. Male Bones

Conservative vs. Labour

Eastern Culture vs. Western Culture

Conservative vs. Liberal

Communism vs. Marxism

KFC vs. McDonald's

EDT vs. EST

Academic Text vs. Non-Academic Text

Germ Theory vs. Terrain Theory

Development vs. Growth

More...

This vs. That

Contact
Terms Of Service
Privacy Policy

© 2023 Bit Flip LLC. All Rights Reserved.

August 20, 2024

# INDUSTRIAL FERMENTATION: SUPPORTING A GROWING INDUSTRY

At its basic level, fermentation is a biological process in which microorganisms, such as bacteria or yeast, are used to create a product of value. The products are then harvested, packaged and sold.

Fermentation has been around for centuries and is commonly known for the creation of food and beverage products like wine and cheese. Modern biotechnology is changing this, and products can range from renewable plastics, fuels, food and much more. While the term fermentation scientifically refers to anaerobic processes, it is used broadly in industry, covering all large-scale industrial microbiological processes and supported by the disciplines of industrial microbiology, fermentation technology and biochemical engineering.

In a laboratory setting, these processes can be conducted in small flasks. With the right technology and expertise, however, they can be executed on an industrial scale to produce various products.

The Applied Research team at the Institute for Advanced Learning and Research (IALR) is leading and hosting a five-day, hands-on workshop (Jan. 13-17) on the science, technology and engineering of

instructors from industry.



"This workshop, and our growing work in the industrial fermentation space, reflects IALR's commitment to facilitating industry-driven, collaborative workforce development programs for the careers of today and tomorrow." – Telly Tucker, IALR President

# What Is Industrial Fermentation?

Biotechnology is the use of living organisms, cells and biological processes to develop products and technologies for various applications. Healthcare, agriculture and environmental management are a few industries that utilize biotechnology processes.

Fermentation is a fundamental process in biotechnology. It uses microorganisms' metabolic activities to produce different products.

chemical engineering and fermentation technology. All of these are involved. This is highly interdisciplinary. It's both science and engineering together." – Dr. Biswarup Mukhopadhyay, Professor, Virginia Tech Department of Biochemistry

Industrial fermentation processes are used in many different industries, including:

Pharmaceuticals: Fermentation produces many antibiotics, vaccines and therapeutic proteins.

Food and Beverages: Fermentation processes are used to produce alcoholic beverages like beer and wine and foods like yogurt and cheese.

Agricultural Products: Fermentation processes are used in the development of agricultural products, including biostimulants, biopesticides and biofertilizers. The Plant Endophyte Research Center at IALR studies how these naturally occurring biostimulants and biocontrol agents can lead to more sustainable agriculture.



26:19

# Preparing the Workforce

IALR is committed to supporting and managing programs that equip the workforce with the skills and certifications for successful work. Just some of the many collaborative efforts include a systematic work-based learning framework, hands-on career introductions for Virginia middle schoolers, and accelerated training for adults looking to start a career in the advanced manufacturing sector.

IALR's Vice President of Applied Research, Dr. Scott Lowman, has been exploring options to provide fermentation training for years.

"We wanted to partner to provide training and opportunities to people who may want to change careers or get into the biotech industry at a technician level." – Dr. Scott Lowman, Vice President of Applied Research, IALR

This five-day workshop will introduce microbiology and engineering aspects of industrial microbial cultures, also known as fermentation, emphasizing technology development and scale-up.



Participants will generate, analyze and discuss their own data in a group setting.

#### The workshop will include:

Anaerobic systems relevant to industry focused on human and animal gut microbiome and biofuel

Theoretical lectures alternating with practical experiments

Lessons relating to the day-to-day operation of a fermentation plant

"This class is focused on how to take a small-scale process that has been developed in the lab and take it into the production stage," Dr. Mukhopadhyay said.

Companies interested in having employees participate in this workshop should contact Dr. Scott Lowman.

Dr. Lowman and Dr. Mukhopadhyay plan to make this a recurring workshop. And offering occasional workshops is not the end of the support that IALR will provide for the growth of industrial fermentation and the biotechnology industry as a whole.

"It's an industry we could support beyond just workforce training, but we're entering it through the workforce training opportunity. As we work with these companies, we may be able to support them with research services as well." – Dr. Scott Lowman, Vice President of Applied Research, IALR

Filed Under: Featured, IALR, Research Tagged With: applied research, Dr. Biswarup Mukhopadhyay, Dr. Scott Lowman, fermentation, Telly Tucker

#### **OUR STORY**

About Us

**Our Focus** 

#### **LEADERSHIP**

Leadership Team

Strategic Plan

**Board of Trustees Meetings** 

**Foundation Board** 

## **CONNECT WITH IALR**

Contact Us

Newsletters

**Podcast** 

**Tours** 

Social Media

Speakers Bureau

## **QUICK LINKS**

Megabytes Cafe

Make a Payment

FOIA

Current Bids & Proposals

Accessibility

Privacy Policy

150 Slayton Avenue Danville, VA 24540

info@ialr.org (434) 766-6700



Copyright © 2024 The Institute For Advanced Learning And Research. All rights reserved.

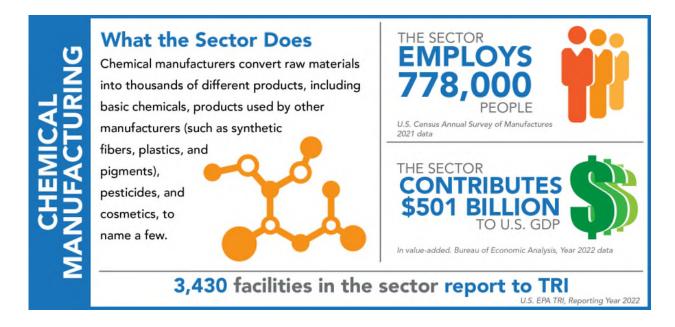




Home <a href="https://epa.gov/">https://epa.gov/trinationalanalysis">https://epa.gov/trinationalanalysis</a> / Comparing Industry Sectors <a href="https://epa.gov/trinationalanalysis/comparing-industry-sectors">https://epa.gov/trinationalanalysis/comparing-industry-sectors</a>

# **Chemical Manufacturing**

This section examines how TRI chemical wastes are managed in the chemical manufacturing sector (defined as facilities reporting their primary NAICS code as 325).



This map shows the locations of the chemical manufacturing facilities that reported to TRI for 2022, sized by their releases. Click on a facility for details on its TRI reporting.

Chemical Manufacturing Facilities Reporting to TRI, 2022 View Larger Map

For 2022, more facilities reported to TRI from the chemical manufacturing sector than from any other industry sector (3,430 facilities; 16% of all facilities that reported to TRI for 2022). This sector reported 54% of all waste managed, more than any other sector.

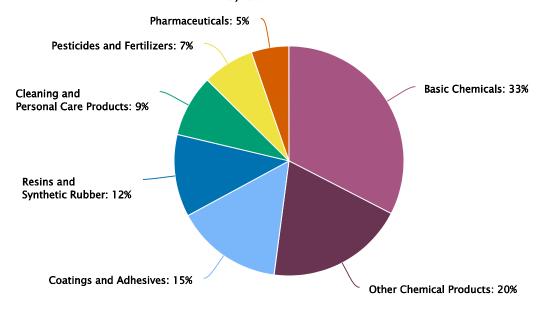
This large and diverse sector includes facilities producing basic chemicals and those that manufacture products through further processing of chemicals. The chart below shows the number of facilities by chemical manufacturing subsectors that reported to TRI for 2022.

## Chemical Manufacturing Facilities by Subsector, 2022





#### 3,430 total facilities



Operations in the chemical manufacturing sector include:

- Basic chemicals facilities produce large quantities of chemicals that are often used to make other chemicals or products. Basic chemicals include petrochemicals, industrial gases, and synthetic dyes and pigments.
- Coatings and adhesives facilities mix pigments, solvents, and binders into architectural and industrial paints; manufacture paint products such as paint removers and thinners; and manufacture adhesives, glues, and caulking compounds.
- Resins and synthetic rubber facilities manufacture resins, plastic materials, synthetic rubber, and fibers and filaments.
- Facilities in the "Other Chemical Products" subsector make chemicals for a wide variety of applications. These include chemicals used in photography, explosives, inks and toners, and transportation equipment like antifreeze or brake fluid.

**Previous** <a href="https://epa.gov/trinationalanalysis/manufacturing-waste-management-trend">https://epa.gov/trinationalanalysis/manufacturing-waste-management-trend</a>

**Next** <a href="https://epa.gov/trinationalanalysis/chemical-manufacturing-waste-management-trend">https://epa.gov/trinationalanalysis/chemical-manufacturing-waste-management-trend</a>

This page was published in March 2024 and uses the 2022 TRI National Analysis dataset made public in TRI Explorer in October 2023.

Last updated on August 20, 2024



## David W. Rogers 4625 Glynwoods Court Fernandina Beach, FL 32034

#### **EDUCATION:**

North Carolina State University 1989-1993 BS in Chemistry BS in Biochemistry

#### **WORK HISTORY:**

#### **Duke University** 1994-1996

Master of Environmental Management in Environmental Toxicology and Chemistry

#### International Paper, Erie, PA 1996-2001

Environmental, Health and Safety Coordinator 1996-1998

Responsible for safety in the machines and finishing operation as well as mill wide Env compliance management systems development.

Project Engineer 1998-2000

Responsible for project engineering and implementation of projects in the Pulpmill. Focus on Cluster Rule strategy, engineering, and implementation.

Pulp Mill Shift Supervisor 2000-2001

Responsible for all aspects of shift management with a priority on health and safety, environmental compliance, and production management of a 24/7 rotating schedule operation.

#### Rayonier Performance Fibers Jesup Mill 2001-2007

Environmental Engineer 2001-2004

Assistant to the Environmental Manager, was responsible for installing management systems for all aspects of compliance at the mill with a focus on the mill's first Title V permit.

Manager, Environmental Operations 2004-2007

Expanded role of responsibility including personnel management for all employees in the Environmental department.

#### Rayonier Performance Fibers/RYAM Fernandina Mill 2007-Present

Environmental Manager, 2007-2022

Responsible for all aspects of Environmental compliance at the Fernandina mill.

Manager, Biomaterials Sales and Environmental 2022-2024

Incorporated responsibility for byproduct sales and management for RYAM's Jesup and Fernandina mills into the Environmental manager role.