BTC3305 BIOPROCESSING AND BIOMANUFACTURING DESIGN

PRODUCTION OF ITACONIC ACID FROM OIL PALM EMPTY FRUIT BUNCH BY ASPERGILLUS TERREUS

GROUP B4

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EXECUTIVE SUMMARY

Itaconic acid (IA) is a chemical product with numerous applications that can be produced on a large scale through fermentative processes. Itaconic acid, also known as methylene succinic acid, is used in a variety of industries, including manufacturing, agriculture, and medicine. Certain characteristics of IA confer a high level of interest in the global market. As a result, itaconic acid has been extensively researched in terms of production methods and polymerization. Due to its widespread application, IA demand is increasing exceeding its production capacity. However, the IA production has a slow rate of development of viable end-use applications and relatively high price. Therefore, seeking cheaper and renewable raw materials is one of the best strategies to decrease the production cost of the IA.

Many researchers have recently attempted to replace the carbon source used for microbial IA production with less expensive alternative substrates. Agricultural wastes such as oil palm empty fruit bunch (OPEFB) is one of the alternatives. OPEFB is an agricultural residue from the palm oil industry in which its extraction process yields huge amounts of biomass residues. They could be used as a raw material for cheap renewable feedstock to produce itaconic acid and many other value-added products. Therefore, this project has the aim to use the efficiency of OPEFB as a cellulosic substrate for IA production by *Aspergillus terreus*, and specifically to produce 1000 t/year of itaconic acid with 99% purity from oil palm empty fruit bunch by *Aspergillus terreus*. Through this project, it is expected to produce 99% purity of itaconic acid with weight of 4572kg per day. This report also provides a techno-economic evaluation for setting up the plant for IA production. The operating cost of this project is expected to be around RM 15,327,360.11 per year with total revenues of RM 80,000,000 annually. This project also has the potential to bring in net profit around RM 64,672,639.90. Therefore, the use of OPEFB as a feedstock can result in a value-added product of itaconic acid and the elimination of residues from communities and industrial plants. The anticipated growth of IA on the appropriate market also depends on the advancement of technologies for producing IA and its derivatives.

DESIGN OBJECTIVE

To produce 1000 t/year of itaconic acid with 99% purity from oil palm empty fruit bunch by *Aspergillus terreus*

LOCATION FOR THE IMPLEMENTATION OF PROCESS DESIGN

The suggested location is at Kinabatangan, Sabah. Sabah is chosen because it is one of the largest palm oil states in Malaysia. In Kinabatangan, there are several palm oil mills such as Felda and FICO, which are the oil palm waste producers. This location is near to our source of oil palm waste and can make the transportation more economical and efficient.

DATABOOK

Product:

Uses	Incorporated into polymers and used as a comonomer in polymers
Form	White crystalline powder
Yield	0.7 kg/kg
Quality	99% purity

Reactants:

Availability	Oil palm empty fruit bunch (oil palm mills) Sodium hydroxide solution Cellulase Aspergillus terreus
Impurities	Cell debris and unfermented sugar

Reactions:

Phases	Solid → liquid → solid
Equilibrium	Isothermic
Kinetics	Glycolysis and tricarboxylic acid cycle
By-products	Succinic, malic, and α-ketoglutaric acid

Physical properties:

Solubility	83g/L in water
Density	1.63 g/cm^3
Vapour pressure	7.11 x 10^{-7} mmHg at 25°C

Chemical properties:

Toxicity	Non-toxic chemical compound
Reactivity	Highly reactive and can be self-polymerized or polymerized with other monomers such as acrylonitrile, and can also be subjected to esterification and addition
Corrosivity	Non-corrosive in presence of glass
Flammability	Non-flammable in presence of open flames and sparks, of heat, of oxidising materials, of reducing materials, of combustible materials, of moisture.

Environmental impact:

Hazards	H315 Causes skin irritation. H319 Causes serious eye irritation. H335 May cause respiratory irritation.
Disposal	Dispose of waste at a licensed waste disposal facility in compliance with local Waste Disposal Authority requirements. This chemical, as well as its container, must be properly disposed of. When managing waste, remember to take the same safety measures you would when handling a product.

PROBLEMS & SOLUTIONS

No.	Problems	Solutions
1	Cake layer formation has a substantial impact on operating performance and is difficult to control.	Carstensen et al. (2013) used “reverse-flow diafiltration” (RFD) to achieve 100% recovery using pure IA solutions. This suggests that the fermentation broth's contents have a negative impact on permeability and product recovery. The RFD technique reduces the hydromechanical stress.
2	A continuous process mode is an excellent option for increasing the volumetric productivity of itaconic acid but product concentrations in continuous processes are frequently lower than in repeated-batch and fed-batch processes.	As a solution, Carstensen et al. (2013) employed an in situ membrane module in a continuously operating stirred tank reactor to improve volumetric activity and produce an itaconic acid-containing permeate stream.
3	Precipitation of IA is a simple and easy process, however, the formation of residues and the need for a considerable amount of precipitant makes it a less attractive option. Because of its high toxicity, lead solutions necessitate chemical processing, which may raise the ultimate cost of the product (Kobayashi and Nakamura., 1971).	Crystallisation can be utilised for both recovery and polishing purposes. It is a reasonably simple and effective recovery technique that produces a pure product. A single crystallisation process is insufficient to recover all of the products in a stream, therefore a recycling phase is always required. Crystallisation is employed as a polishing procedure to crystallise previously concentrated solutions, and in this instance, the size, cost, and

		energy needs are minimal (Magalhães et al., 2017)
4	Itaconic acid production is suppressed during cultivation because itaconic acid produced inhibits *A. terreus* growth significantly (Saha, 2017). This is known as product inhibition.	In-situ product removal should be used to avoid high itaconic acid concentrations (Klement et al., 2012).

BACKGROUND OF PROCESS IDENTIFIED

Malaysia is currently the world's leading producer of palm oil (Gerard, 2017). After Indonesia, it is the world's second-largest producer, together those two countries accounting for more than 80% of global output. A large part of Malaysia's GDP is dependent on the oil palm. In 2020 alone, the gross domestic product from palm oil was estimated to be 36.87 billion Malaysian ringgit. However, several global projects are addressing the challenges concerning the environmental and social harms caused by an unregulated industry of palm oil, which need to undergo positive change. Thus, the Malaysian government is devoted to palm oil sustainability, and it has implemented the Malaysian Sustainable Palm Oil Certification Scheme, which affects all oil palm plantations, independent and organised smallholdings, and palm oil processing facilities throughout the country. "To build, maintain, and administer a fund for the purposes of establishing and administering a sustainable palm oil certification programme in Malaysia," according to the MSPO's mission. The palm oil business is becoming more sustainable and transparent as a result of international standards that are both voluntary and mandatory. This commitment to long-term economic and environmental viability conserves natural resources and biodiversity.

The biomass wastes produced by the palm oil extraction process, particularly OPEFB, account for 20% of the fresh fruit weight. Plantations produce a huge amount of oil palm trunks and fronds. Other biomasses, such as mesocarp fibre, kernel shell, and empty fruit bunches, are produced during the milling of fresh fruit bunches. Oil palm empty fruit bunch (OPEFB) is a by-product of the processing of crude palm oil (CPO) in a palm oil mill in Malaysia, with an annual production of up to 22–23 million tonnes (Padzil, 2020). The OPEFB contains around 65.5 percent holocellulose, which is a rather high percentage and one of the most promising feedstocks for the manufacturing of bio-based components (Faharna, 2010). OPEFB is the cheapest natural fibre with good qualities as a non-wood fibre, and it has a lot of promise as a key raw material to replace woody plants, which are expensive in numerous sectors. 46,000 kilotons in the form of fronds and 11,000 kilotons in the form of trunks were the principal agricultural wastes from the oil palm biomass. OPEFB is an excellent choice as a prospective raw material because of its high cellulose content and widespread availability (Padzil, 2020).

Itaconic acid ($C_5H_6O_4$) is an unsaturated dicarboxylic acid also known as 2-methylene butanedioic acid, propylene dicarboxylic acid, or 2-methylenesuccinic acid. Itaconic acid is highly soluble in water and alcohols, stable at room temperature, and stable in middle-basic, neutral, and acidic conditions due to its weak acidity. It resembles white crystalline powder or crystals and has no odour. Because of the variety of functional groups in itaconic acid, it is an efficient intermediate in the synthesis of various complex organic compounds. It can take part in a wide range of reactions, including esterification with alcohols, metal salt formation, anhydride production, polymerization, and others (Teleky & Vodnar, 2019).

Itaconic acid was found to be a biological metabolite when it was discovered to be produced by *Aspergillus itaconicus*, while *A. terreus* was later discovered to accumulate itaconic acid in even greater amounts than *A. itaconicus* (El-Imam & Du, 2014). Itaconic acid is most commonly found in the plastic and paint industries. It is an unsaturated dicarbonic acid that can be easily incorporated into polymers and used as a comonomer in polymers at a concentration of 1–5% (w/w). Itaconic acid polymerized methyl, ethyl, or vinyl esters are used in plastics, adhesives, elastomers, and coatings. Itaconic acid-containing styrene butadiene copolymers produce rubber-like resins with excellent strength and flexibility, as well as water-proofing coatings with good electrical insulation. Synthetic fibres, lattices, detergents, and cleaners are some other applications. Besides, several mono- and diesters of partially substituted itaconic acid have anti-inflammatory or analgesic properties (Kirimura et al., 2011).

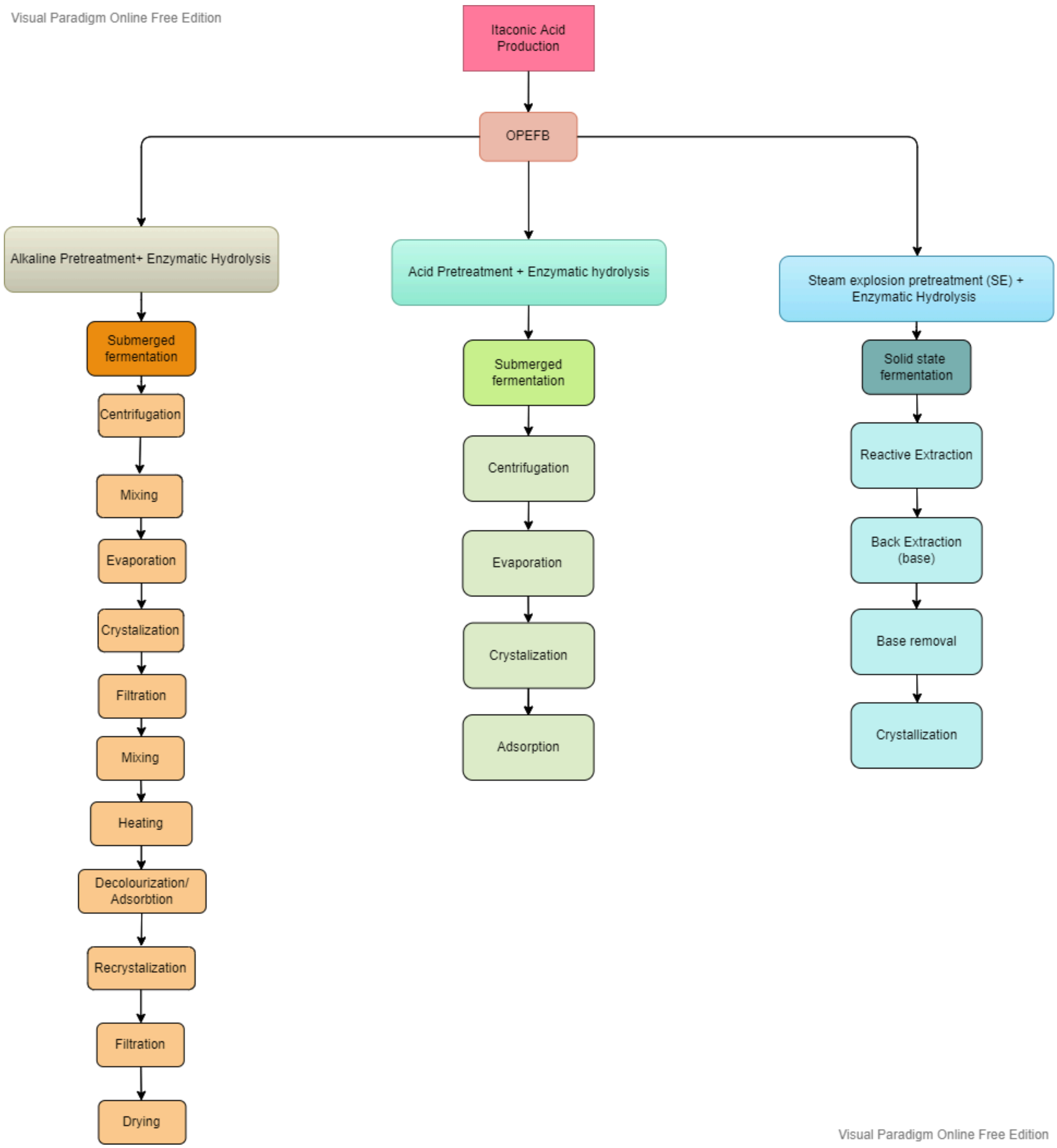
According to Boondaeng et al. (2019), OPEFB is going through pretreatment by soaking in hot water at 80°C, and washed with tap water until it reaches a neutral pH. It is then used for further delignification in a 15% (w/v) sodium hydroxide (NaOH) solution at 90°C. After that, the solid residues are washed with tap water until it reaches a neutral pH. For upstream and downstream processing, it includes culture medium preparation, inoculation, fermentation, centrifugation, evaporation, crystallisation, heating and adsorption (Nieto et al., 2020).

RESEARCH ON SOURCE OF DESIGN INFORMATION

No.	Process	Source
		Literature Research
1	Pre-treatment	Boondaeng, A., U-thai, P., Trakunjae, C., Chuntranuluck, S., & Vaithanomsat, P. (2019). Statistical Optimization of Itaconic Acid Fermentation from Oil Palm Empty Fruit Bunch by Aspergillus terreus K17 for the Application in Textile Industry. *Thai Journal of Agricultural Science*, *52*(2), 119-130. Krull, S., Eidt, L., Hevekerl, A., Kuenz, A., & Prüße, U. (2017). Itaconic acid production from wheat chaff by aspergillus terreus. *Process Biochemistry*, *63*, 169–176. https://doi.org/10.1016/j.procbio.2017.08.010
2	Fermentation	El-Imam, A. A., & Du, C. (2014). Fermentative itaconic acid production. J Biodivers Biopros Dev, 1(1), 1-8. http://dx.doi.org/10.4172/ijbbd.1000119 Yang, J., Xu, H., Jiang C., Zhang, N., Xie, J, Wei, M., & Zhao, J. (2019). Production of itaconic acid through microbiological fermentation of inexpensive materials. Journal of Bioresources and Bioproducts, 4(3), 135-142. https://doi.org/10.12162/jbb.v4i3.001

3	Downstream process	Magalhães, A. I., de Carvalho, J. C., Thoms, J. F., Medina, J. D., & Soccol, C. R. (2019). Techno-economic analysis of downstream processes in itaconic acid production from fermentation broth. *Journal of Cleaner Production*, *206*, 336–348. https://doi.org/10.1016/j.jclepro.2018.09.204 Nieto, L., Rivera, C., & Gelves, G. (2020). Economic assessment of itaconic acid production from aspergillus terreus using superpro designer. *Journal of Physics: Conference Series*, *1655*(1), 012100. https://doi.org/10.1088/1742-6596/1655/1/012100

SYNTHESIS TREE DIAGRAM



COMPARISON ON SYNTHESIS TREE DIAGRAM

The pretreatment process aims to break the lignin bond (delignification), in removing the lignin and some of the hemicellulose contents, damaging the crystalline structure of cellulose and increasing the porosity of the material. Three methods of pre-hydrolysis treatments are proposed and compared; including different chemical pre-treatment which is acid and alkali and steam explosion combined with enzyme hydrolysis treatment.

Comparison	Alkaline pretreatment	Acid pretreatment	Steam explosion pretreatment
Reagents	Sodium hydroxide (NaOH)	Sulphuric acid (H_2SO_4)	High-pressure saturated steam
Temperature	Lower temperatures (120°C)	High temperature (T> 160°C)	High temperatures (190–240°C)
Retention time	Hours to day	Long retention time (50–185 min) at 90 °C Low retention time (7–10 min) at higher temperature (120–130 °C).	(190 °C, 10 min) or (270 °C, 1 min)
Fermentation	Submerged	Submerged	Solid state

Process reaction	Hydrolysis of lignin and the alteration of cellulose structure.	Hydrolysis of hemicelluloses and the alteration of structure.	Hemicellulose degradation and lignin transformation and potential of cellulose hydrolysis.

ELIMINATION OF POSSIBLE SOLUTION

Upstream Process

The highest glucose yield was found in OPEFB treated with NaOH, demonstrating that alkaline pretreatment had a significant impact on glucose yield. By eliminating lignin from lignocellulose, which is linked to both hemicellulose and cellulose, alkaline pretreatment improves enzymatic hydrolysis (Shamsudin et al., 2012). Pretreatment with acid or steam improves enzymatic hydrolysis, however, the glucose yield is lower than pretreatment with alkaline. Most of the hemicellulose was dissolved during acid and steam explosion pretreatment, leaving lignin as a barrier to enzyme penetration, making it difficult for the enzyme to reach the cellulose (Ariffin et al., 2008).

Fermentation

Controlling the amounts of various nutrient elements present in the fermentation medium allows to achieve itaconic acid production that is close to the optimum. The research discovered that particular concentrations of ions of alkaline earth metals, copper, and/or zinc are required for effective production of itaconic acid by submerged fermentation of carbohydrate media. The process of the present invention can be broadly described as the submerged fermentation of a carbohydrate medium such as molasses, corn syrups, beet, OPEFB, or other carbohydrates with a critical concentration of alkaline earth metal ions, as well as copper or zinc ions, in addition to the usual nutrients supplied in this type of fermentation. Any organism capable of generating itaconic acid can be employed. Itaconic acid-producing strains of *Aspergillus terreus*, which have been discovered to produce very excellent yields of itaconic acid, are a particularly preferred class of organisms (Mario & Schweiger, 1963).

Downstream Process

It is proposed that centrifugation is significantly more successful in removing LMW molecules than diafiltration and ion exchange. Centrifugation is thus more effective. The aim of the unit evaporation operation is to concentrate the end product by eliminating water. Separation of itaconic acid by crystallisation was easily recovered by cooling or evaporation crystallisation at low pH values. The filtrate, however, is subjected to another cycle of evaporation, crystallisation, and filtration since it includes a considerable quantity of uncrystallized itaconic acid. The second and third evaporation and filtration processes are carried out in the same way as the first and second evaporation and filtration phases. However, after that, itaconic acid still includes impurities or culture medium residues that must be removed before it reaches its ultimate concentration. For the elimination of contaminants, a granular activated carbon adsorption approach is presented in this study. A considerable number of micropores distinguishes the aforementioned component. These micro-pores are arranged in such a way that the impurity adsorption process will occur. The decolorized solution will be recrystallized and filtered to separate and recover the itaconic acid crystals before being transferred to a rotary drier to remove any leftover moisture using hot air. Finally, since the synthesis technique of producing itaconic acid by catalytic condensation of succinic acid and formalin is unsustainable and costly because it requires massive chemicals, uses more energy, and emits toxic compounds, reactive extraction system (*in situ* product recovery) are not considered (Wasewar, K. L., 2011).

UNIT OPERATION IDENTIFIED

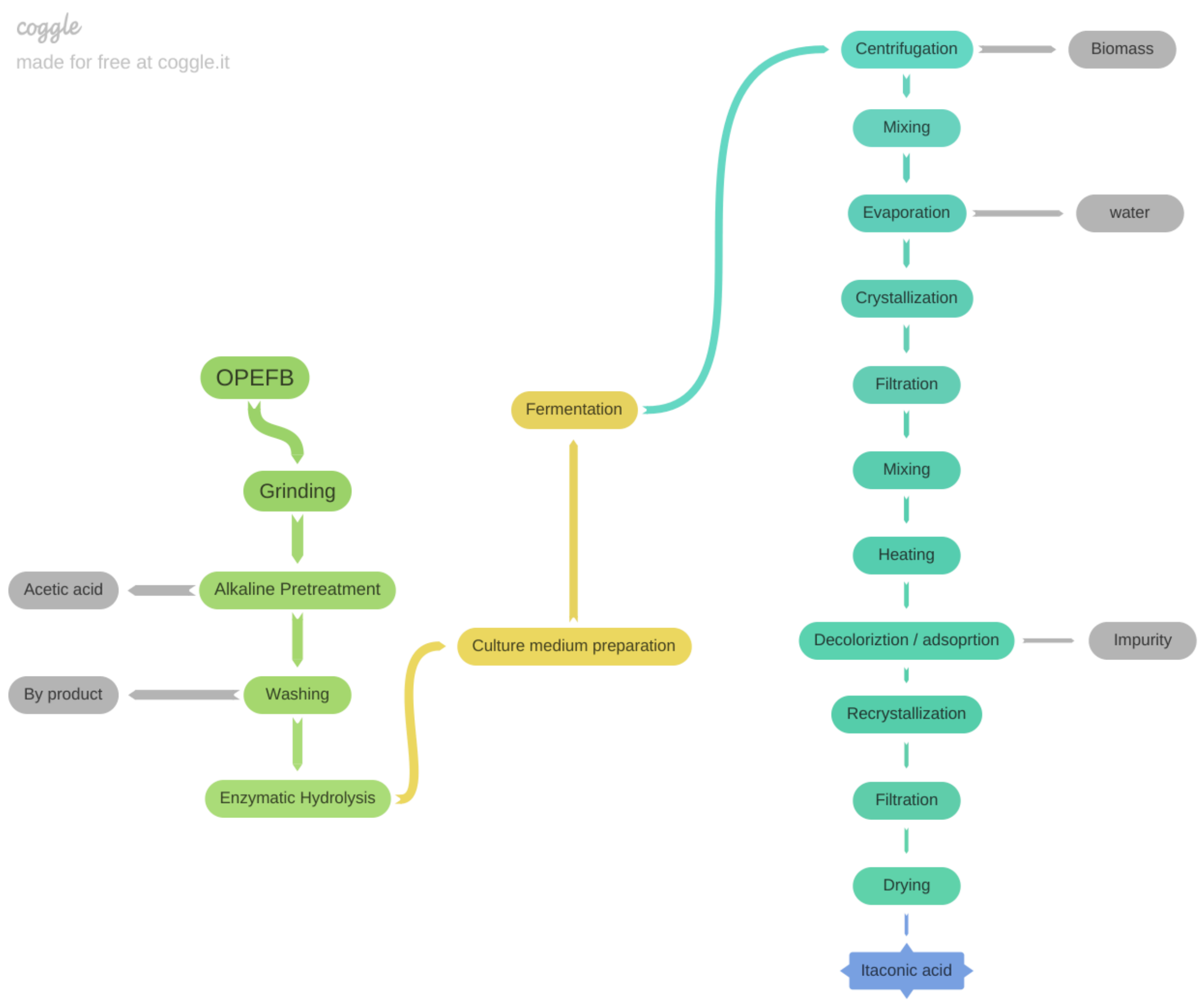
Process	Explanation
<u>Upstream</u> **Grinding**	Impact, collision, friction, and shear forces produced during the grinding process reduce the particle size of OPEFB. This process causes crystal defects and dislocations by altering the cellular structure. Simultaneously, grinding lowers particle size while increasing specific surface areas. The size reduction is improved by raising the milling speed. Milling speed appears to be beneficial to size reduction. OPEFB's structures will be disrupted during grinding, resulting in morphological changes such as an increase in amorphous region ratio and hydrogen bond energy, as well as a decrease in microstructure and size (Herawan et al., 2018). Mechanical grinding treatments were performed using Panjaitan et al., 2017 method. Ethanol and acetone were utilised as solvents. The mechanical grinding activity will be paused for 15 minutes after every 1 hour of grinding (Herawan et al., 2018).
Alkaline pretreatment	An alkaline pretreatment method will be chosen because of the high sensitivity of *A. terreus* towards the sugar degradation products. By dissolving lignin from OPEFB, it is stated as a technique with low generation of inhibitory by-products and an effective pretreatment method for OPEFB at room temperature. For alkaline pretreatment, OPEFB was combined with 0.25 M sodium hydroxide solution at room temperature for 3 days. The pretreated OPEFB was centrifuged for 30 minutes and then rinsed with water (Krull et al., 2017).
Washing	To remove oil residue and debris from the surface of the fibres before biological processing, the OPEFB must be cleaned with tap water and detergent (Arbaain et al.,2019).

	The solid residue will be cleaned by soaking it for 60 minutes in hot water at 80°C and then washing it with tap water until it reaches a neutral pH. It was then delignified further at 90°C for 30 minutes in a 15 % (w/v) sodium hydroxide (NaOH) solution (Boondaeng et al., 2019).
Enzymatic hydrolysis	OPEFB has a high percentage of cellulose and hemicellulose, as well as lignin. It is possible to convert cellulose and hemicellulose into fermentable sugars. Cellulase, a multienzyme complex consisting of Carboxymethyl cellulase (CMCase) or endo-β-glucanase, exo-β-glucanase, and β-glucosidase, is used for enzymatic hydrolysis of cellulosic biomass. These enzymes work together to break down cellulose into glucose (Sugiwati et al., 2021). Enzymatic hydrolysis will be performed at optimum temperature of 50 °C with 150 rpm for 24 h. A continuous stirred tank reactor (CSTR) will be used. The pH is adjusted to pH 4.8 with sulphuric acid. 15 FPU/g of raw material is set as the enzyme loading. The produced glucose for 150 g solid loading is 60 g after saccharification (Boondaeng et al., 2019).
Culture medium preparation	Inoculate a *Aspergillus terreus* spore suspension containing 1 x 10^7 spores/ml in 100 ml of growth medium containing (g/L) glucose, 60; KH_2PO_4, 0.88; $MgSO_4 \bullet 7H_2O$, 0.95; NH_4NO_3, 4; and $CuSO_4$, 0.004 (Boondaeng et al., 2019).
Fermentation	It is a continuous fermentation, so continuous stirred tank reactors are used. The fermentation process takes 6 days (Nieto et al., 2020). Since *Aspergillus terreus* are obligate aerobes, ambient air is sterilised through an air filter and compressed by a centrifugal compressor to increase pressure by 2 bar (Petrides & Ferreira, 2020). The enzyme cis-aconitic acid decarboxylase (CAD)

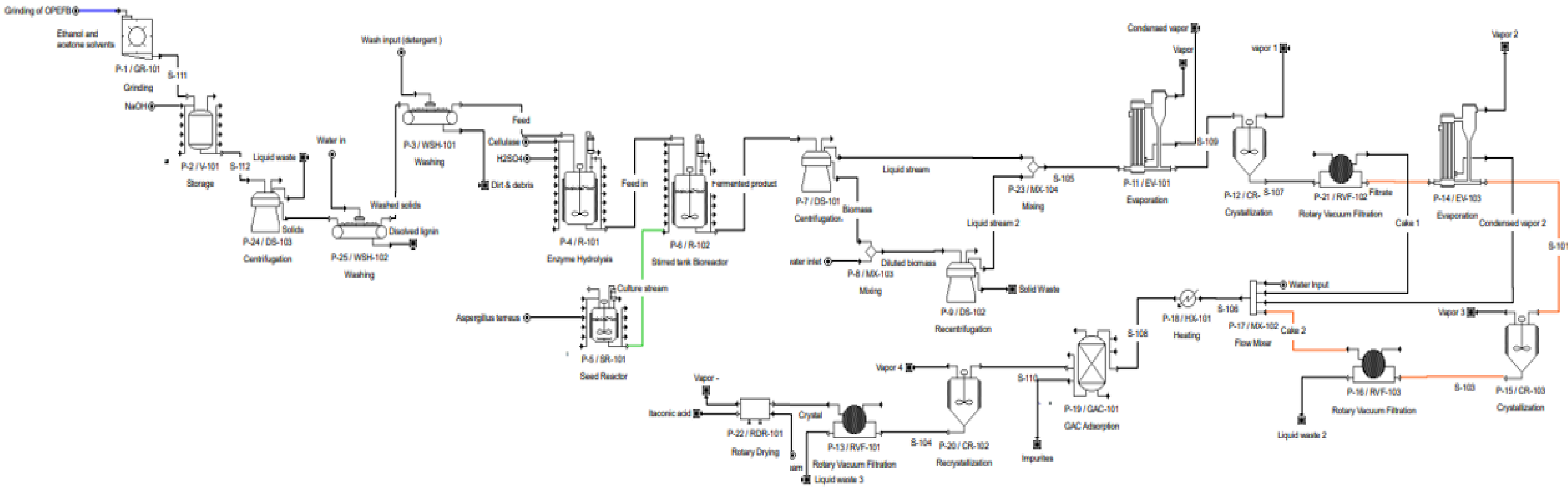
	synthesises itaconic acid from cis-aconitic acid in a single step in *A. terreus* (Petrides & Ferreira, 2020).
<u>**Downstream**</u> **Centrifugation**	The biomass produced by previous fermenters is removed using centrifugation. The liquid stream generated by centrifugation is collected and processed further. A disc-stack centrifuge is used to remove biomass (Nieto et al., 2020). Through centrifugation, 99% of the biomass is removed. Although the majority of itaconic acid is found in the supernatant, some itaconic acid is also found in the centrifuge sludge (Petrides & Ferreira, 2020).
Mixing	The sludge is then re-centrifuged in a second disk-stack centrifuge after being diluted with water using a mixer. A flow mixer is then used to combine the supernatants from both centrifuges (Petrides & Ferreira, 2020).
Evaporation	Triple-stage evaporator is used to remove water to increase itaconic acid concentration to 350 g/L at 80 °C. Saturated steam is used to feed the evaporators. The pressures for the triple effect evaporator are 300, 400, and 500 kPa, while the single effect evaporator is 500 kPa. The evaporator's overall heat transfer coefficient is estimated to be 1.15 kW m^{-2} $°C^{-1}$ (Magalhães et al., 2019).
Crystallisation	Crystallisation is a recovery and polishing technique. Crystallisation is used as a polishing operation to crystallise previously concentrated solutions (Magalhães et al., 2017). A crystallizer receives the concentrated solution. Crystals are formed by cooling the solution to 15 °C, with an expected crystallisation yield of 80%. An itaconic acid crystal is the crystalline form of itaconic acid (Petrides & Ferreira, 2020).
Filtration	The slurry from the crystallizer is then filtered through a rotary

	vacuum filtration procedure, which retains 99 % of the itaconic acid crystals in the filtration cake. However, because the filtrate contains a significant amount of uncrystallized itaconic acid, it is subjected to another round of evaporation, crystallisation, and filtration. The second evaporation and filtration steps are performed in the same manner as the first evaporation and filtration steps. However, the second crystallisation procedure yields 99% itaconic acid crystals (Petrides & Ferreira, 2020).
Mixing	A flow mixing procedure is used to combine the filter cakes from both rotary vacuum filters. In this flow mixer, the condensed steam from the second evaporation procedure, which has a temperature of 79 °C and fresh water are mixed with the filter cakes to produce a suspension with 350 g/L of crystals (Petrides & Ferreira, 2020).
Heating	To re-dissolve the crude itaconic acid crystals, the suspension is heated to 80 °C in a stirred tank (Petrides & Ferreira, 2020).
Decolorization / Adsorption	The itaconic acid solution is then passed through a granular activated carbon column to remove any remaining organic acids or other impurities that might affect the product's colour (Petrides & Ferreira, 2020).
Recrystallisation	The decolorized solution is then transferred to a crystallisation step at 15 °C, where itaconic acid is recrystallized (Petrides & Ferreira, 2020).
Filtration	A rotary vacuum filter is used to separate and recover the itaconic acid crystals (Petrides & Ferreira, 2020).
Drying	The crystals are sent to a rotary dryer, which uses hot air to remove any remaining moisture (Petrides & Ferreira, 2020).

BLOCK FLOW DIAGRAM (BFD)



PROCESS FLOW DIAGRAM (PFD)

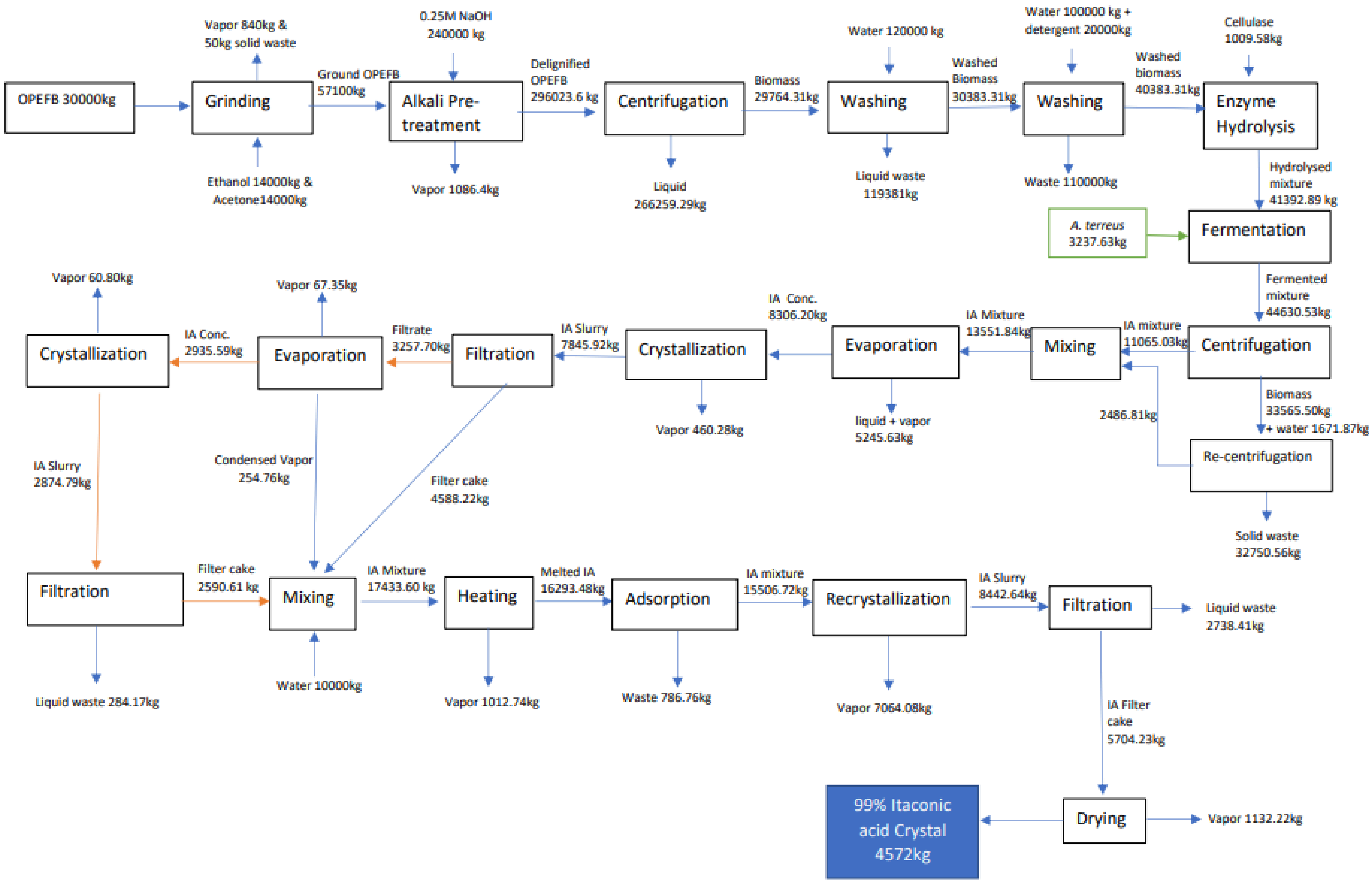


MASS BALANCE OF PRODUCT FORMATION

	Grinding		Alkaline Pretreatment		Centrifugation		Washing		Washing		Enzyme Hydrolysis		Fermentation	
Sections	**In**	**Out**	In	Out	In	Out	In	Out	In	Out	In	Out	In	Out
OPEFB	30000	29950	29950	18569	296023.6	18383.31	18383.31	30383.31	30383.31	40383.31	40383.31	18576.3226	18576.3226	9288.1613
Acetone	14000	13580	13580	13036.8		13036.8								
Ethanol	14000	13580	13580	13036.8		13036.8								
NaOH			240000	228000		228000								
Lignin				11381		11381	11381	11381						
Liquid				12000		12185.69								
IA														
Water							120000	108000	100000	90000		4038.331	4038.331	3634.4979
Detergent									20000	19800				
Cellulase											1009.58275			
H2SO4														
Glucose												13800	13800	
A.Terreus													3237.63226	19425.79356
Biomass												2555.24055	2555.24055	11092.50367
Solid Waste		50								200				
Vapor		840		1086.4										
Condensed V.														
Impurities												2422.9986	2422.9986	1189.568576
TOTAL	58000	58000	297110	297110	296023.6	296023.6	149764.31	149764.31	150383.31	150383.31	41392.89275	41392.89275	44630.52501	44630.52501

	Centrifugation		Re-Centrifugation		Mixing		Evaporation		Crystallization		Filtration		Evaporation	
Sections	In	Out	In	Out	In	Out	In	Out	In	Out	In	Out	In	Out
OPEFB	9288.16	1857.63	1857.63											
Acetone														
Ethanol														
NaOH														
Lignin														
Liquid												3184.47	3184.47	
IA		7430.53		185.76	7616.29	7616.29	7616.29	7616.29	7616.29	7463.97	7463.97	4478.38		2866.02
Water	3634.50	3634.50	1671.87	1337.50	4971.99	4971.99	4971.99	497.20	497.20	198.88	198.88			
Detergent														
Cellulase														
H2SO4														
Glucose														
A.Terreus	19425.79	19231.54	19231.54											
Biomass	11092.50	11405.72	11405.72	30744.32										
Solid Waste				2006.24										
Vapor								4474.79		460.28				67.35
Condensed V.								770.84						254.76
Impurities	1189.57	1070.61	1070.61	963.55	963.55	963.55	963.55	192.71	192.71	183.07	183.07	183.07	73.23	69.57
TOTAL	44630.53	44630.53	35237.37	35237.37	13551.84	13551.84	13551.84	13551.84	8306.20	8306.20	7845.92	7845.92	3257.70	3257.70

	Crystallization		Filtration		Mixing		Heating		Adsorption		Recrystallization		Filtration		Drying	
Sections	In	Out	In	Out	In	Out	In	Out	In	Out	In	Out	In	Out	In	Out
OPEFB																
Acetone																
Ethanol																
NaOH																
Lignin																
Liquid				284.17	10000.00	10127.38	10127.38	9114.64	9114.64	8658.91	8658.91	1731.78	1731.78	2738.41		
IA	2866.02	2808.70	2808.70	2527.83	7006.21	7006.21	7006.21	7006.21	7006.21	6796.02	6796.02	6660.10	6660.10	5661.09	5661.09	4528.87
Detergent																
Cellulase																
H2SO4																
Glucose																
A.Terreus																
Biomass																
Waste										786.76						
Vapor		60.80			254.76	127.38		1012.74				7064.08				1132.22
Condensed V.																
Impurities	69.57	66.09	66.09	62.79	172.63	172.63	172.63	172.63	172.63	51.79	51.79	50.75	50.75	43.14	43.14	43.14
Total	2935.59	2935.59	2874.79	2874.79	17433.60	17433.60	17306.22	17306.22	16293.48	16293.48	15506.72	15506.72	8442.64	8442.64	5704.23	5704.23



OPEFB 30000kg
Vapor 840kg & 50kg solid waste
Grinding
Ethanol 14000kg & Acetone14000kg
Ground OPEFB 57100kg
0.25M NaOH 240000 kg
Alkali Pre-treatment
Vapor 1086.4kg
Delignified OPEFB 296023.6 kg
Centrifugation
Liquid 266259.29kg
Biomass 29764.31kg
Water 120000 kg
Washing
Liquid waste 119381kg
Washed Biomass 30383.31kg
Water 100000 kg + detergent 20000kg
Washing
Waste 110000kg
Washed biomass 40383.31kg
Cellulase 1009.58kg
Enzyme Hydrolysis
Hydrolysed mixture 41392.89 kg
A. terreus 3237.63kg
Fermentation
Fermented mixture 44630.53kg
Centrifugation
Biomass 33565.50kg + water 1671.87kg
Re-centrifugation
Solid waste 32750.56kg
2486.81kg
IA mixture 11065.03kg
Mixing
IA Mixture 13551.84kg
Evaporation
liquid + vapor 5245.63kg
IA Conc. 8306.20kg
Crystallization
Vapor 460.28kg
IA Slurry 7845.92kg
Filtration
Filtrate 3257.70kg
Filter cake 4588.22kg
Evaporation
Vapor 67.35kg
Condensed Vapor 254.76kg
IA Conc. 2935.59kg
Crystallization
Vapor 60.80kg
IA Slurry 2874.79kg
Filtration
Liquid waste 284.17kg
Filter cake 2590.61 kg
Mixing
Water 10000kg
IA Mixture 17433.60 kg
Heating
Vapor 1012.74kg
Melted IA 16293.48kg
Adsorption
Waste 786.76kg
IA mixture 15506.72kg
Recrystallization
Vapor 7064.08kg
IA Slurry 8442.64kg
Filtration
Liquid waste 2738.41kg
IA Filter cake 5704.23kg
Drying
Vapor 1132.22kg
99% Itaconic acid Crystal 4572kg

TECHNOECONOMIC EVALUATION

TECHNICAL EVALUATION

	Our product	**A.B Enterprises**	**Shanghai CRM New Material Technology Co.,Ltd**
Purity (%)	99	98	99
Origin	Malaysia	India	China
Form	White crystalline powder	White to light beige crystalline powder	White crystalline powder
Density (g/cm^3)	1.63	1.63	1.30
Melting point (℃)	163-169	165-168	119-121
Boiling point (℃)	268	268	214.6
Solubility in water (g/L)	83	83	150
Price (RM/kg)	80	25	264

ECONOMY EVALUATION

FINALISED TOOLS AND EQUIPMENT REQUIRED

Equipments/ Materials with Specifications	**Unit Required**	**Net price**	**Function**
Grinder Name: GR-101 Capacity: 60,000.00 kg/day	1	RM 375000	To reduce the particle size of the empty fruit bunch for easy pumping to the next step.

Washer Name: WSH-101 Capacity: 1000 kg/h	2	RM 35,000	To clean OPEFB by removing oil residue and debris
Storage Tank Name: ST-101 Capacity: 58000 L	2	RM 1227000	One of the storage tanks will be used to collect waste and another one will be to soak the OPEFB in NaOH.
Stirred reactor Name: R-101 Capacity: 45000 L	8	RM 30,49,000	2 of them for the enzyme hydrolysis and 6 of them are for the fermentation
Seed Fermenter Name: SR-101 Capacity: 850 L	4	RM 30,19,000	For pre-culture of *Aspergillus terreus*
Disk stack centrifuge Name: DS-101 Capacity: 100,000 L/h	3	RM 191,24,000	To remove solid-liquid stream biomass from the culture media
Evaporator Name: EV-101 Capacity: 500.00 m^2	2	RM 650,01,000	To remove water to increase itaconic acid concentration

Crystalliser Name: CR-101 Capacity: 10000 L	3	RM 3,295,000	To crystallise previously concentrated solutions and recover itaconic acid in crystal form
Heat exchanger Name: HX-101 Capacity: 90.00 m^2	1	RM 547,000	To transfer heat
Rotary dryer Name RDR-101 Capacity: 47.12 m^2	1	RM 1,427,000	To removes the residual moisture with hot air
GAC column Name: GAC-101 Capacity: 35342.92 L	1	RM 1,960,000	To remove impurities or residues of culture media from itaconic acid
Mixer Name: MX-101 Capacity: 360,000 kg/h	3	RM 390,000	To mix all the components
Rotary vacuum filter Name: RVF-101 Capacity: 75 m^2	3	RM 1,021,000	To filter and recover the itaconic acid crystals
Sodium hydroxide (0.25 M)	240 kg	RM 0.84/ kg	For alkaline pretreatment process

Detergent	20000 kg	RM 1.1 / kg	To remove oil residue and debris
Ethanol	1106 kg	RM 3.4 / kg	As solvent
Acetone	700 kg	RM 3.7 / kg	As solvent
Cellulase	1009.58 kg	RM12.34 / kg	For enzymatic hydrolysis to break down cellulose to glucose

ESTIMATION OF COSTS & EQUIPMENT PROCUREMENT

CALCULATION OF CAPEX

Total cost required for equipment

Equipment	Unit	Unit Cost (RM)	Cost (RM)
Grinder	1	37,500	37,500
Washer	2	35,000	70,000
Stirred tank bioreactor	8	3,049,000	243,920,00
Disk stack centrifuge	3	19,124,000	57,372,000
Seed reactor	4	3,019,000	12,076,000
Evaporator	2	6,501,000	13,002,000
Crystalliser	3	3,295,000	9,885,000
Heat exchanger	1	547,000	547,000
Rotary dryer	1	14,270,000	14,270,000
GAC column	1	1,960,000	1,960,000
Mixer	3	390,000	1,170,000
Rotary vacuum filter	3	1,021,000	3,063,000
Storage tank	2	1,227,000	2,454,00
		TOTAL	**127,793,000**

CALCULATION OF OPEX

The operation expenditure (OPEX) is divided into fixed costs and variable costs. The fixed costs are yearly costs which are not directly dependent on the running hours of the plant. The variable costs depend on the actual operational hours of the plant, and the electricity and water consumption.

Overall material cost

Bulk Material	**Unit Cost (RM)**	**Per Entry (kg)**	**Annual Amount**	**Annual Cost (RM)**
Acetone (5%)	3.7	700	182,000	673,400
Ammonia	1.2	1010	262,600	315,120
Ethanol (79 g/L)	3.4	1106	287,560	977,704
Cellulase	12.34	1009.58	262,490.80	3,239,136.47
Detergent	1.1	20000	5,200,000	5,720,000
NaOH (0.25M)	0.84	240	62,400	52,416
Discrete Materials	**(RM/Entity)**	**Per Entry**	**Entities**	**Cost (RM)**
OPEFB	88	30000	330	29,040
			TOTAL	**11,006,816.47**

Electric consumption in production of itaconic acid

Equipment	**Unit operation required**	**Power (kW)**	**Operation hours**	**Total kW/h**	**Electric consumption (RM)**
Grinder	1	3	8	28	6.21
Washer	1	2.2	8	17.6	3.91

Stirred reactor	8	75	24	1,800	3,196.80
Disk stack centrifuge	3	4	4	16	10.65
Seed reactor	4	3	24	72	63.36
Evaporator	2	2	8	16	7.10
Crystalliser	3	25	8	200	133.20
Heat exchanger	1	-	8	-	-
Rotary dryer	1	0.6	8	4.8	1.07
GAC column	1	35	8	280	62.16
Mixer	3	0.75	8	6	4.00
Rotary vacuum filter	3	0.6	8	4.8	3.21
				Total	**RM 3,491.67**

*Price of electricity per kilowatt hour = RM0.222

Water consumption for production of 4572 kg itaconic acid

Total water used (m^3)	**Water consumption (RM)**
220 per batch	440

*Price of water per m^3 = RM2.00

Management and staff cost

Staff	**No. of staff required**	**Monthly salary (RM)**	**Total (RM)**
Operator (full time)	10	2100	21,000

Engineer	3	3000	9000
Administrative officers	5	2500	12,500
		Total	**42,500**

Financial economic assumption

Parameter	**Assumption**
Plant capacity	4572 kg/day
Time required to produce 1000 tonnes itaconic acid	260 days
CAPEX	RM 127,793,000
OPEX	RM 11,053,248.14

ANNUAL OPERATING COST SUMMARY TABLE

Cost Item	**Annual cost (MYR per Year)**	**%**
Raw material	11,210,360	78.12228236
Operator & engineer	510000	3.554066382
Maintenance	200000	1.393751522
Research & Development	1000000	6.968757611
Transportation	5000	0.034843788
Water Bill	114400	0.797225871
Electric Bill	910000	6.341569426
Licensing Fee	100000	0.696875761
Royalty	300000	2.090627283
TOTAL	**14,349,760**	**100**

FIXED CAPITAL ESTIMATE SUMMARY

Total Plant Direct Cost (TPDC)		**Cost (RM)**
Equipment Purchase Cost		12779300
Installation		65,979,000
Process Piping		52,750,000
Instrumentation		60,286,000
Insulation		4,521,000
Electrical		15,072,000
Buildings		67,822,000
Yard Improvement		22,607,000
Auxiliary Facilities		60,286,000
	TPDC	**477,116,000**
Total Plant Indirect Cost (TPIC)		
Engineering		125,009,000
Construction		175,013,000
	TPIC	**300,023,000**
Total Plant Cost (TPC = TPDC + TPIC)		
	TPC	**777,139,000**
Contractor’s Fee & Contingency (CFC)		
Contractor’s Fee		40,003,000
Contingency		80,006,000
	CFC	**120,009,000**
Direct Fixed Capital Cost (DFC = TPC + CFC)		
	DFC	**897,148,000**

EXECUTIVE SUMMARY

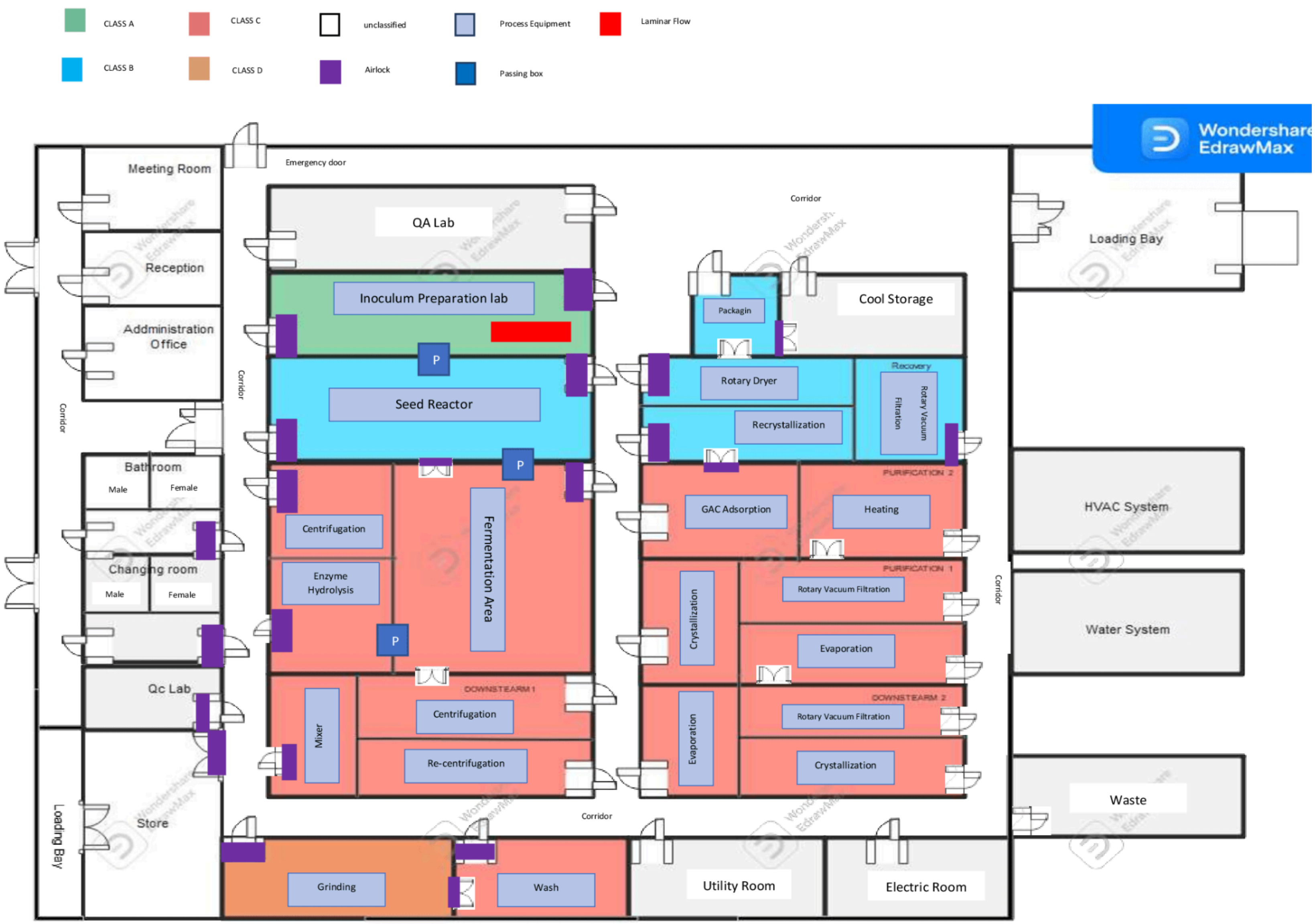
Economics		
Total Investment	1024941000	MYR
Total Revenues	80000000	MYR/yr
Operating Cost	15327360.11	MYR/yr
Production Ref.	1000.00	Ton/yr
Unit Production Cost	15.3273601	MYR/Kg
Net Profit	64672639.9	MYR
Project Indices		
Gross Margin	80.84	%
Gross Profit	64.68	MYR/Kg
Markup	405.37	%
Return On Investment (ROI)	-93.69	%
Annualized ROI	-45.74	%
NPV at (7.00 %)	-755,472,168.26	MYR
Payback Time	4.42	yr

PLANT LAYOUT

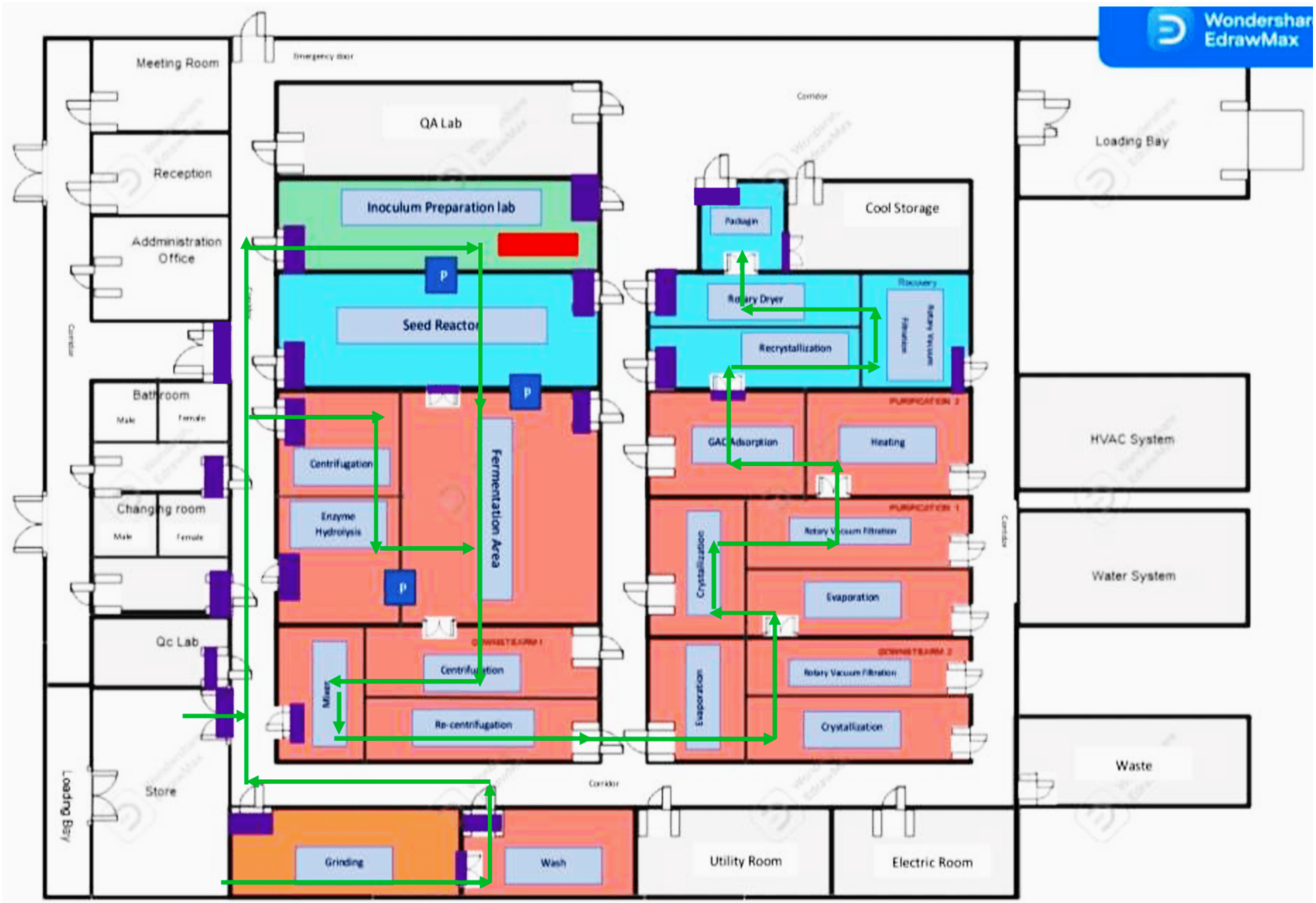
Total Area (m) = 30m x 20m = 600m^2 = 6458.35 ft^2

Drawing Scale = 1:100

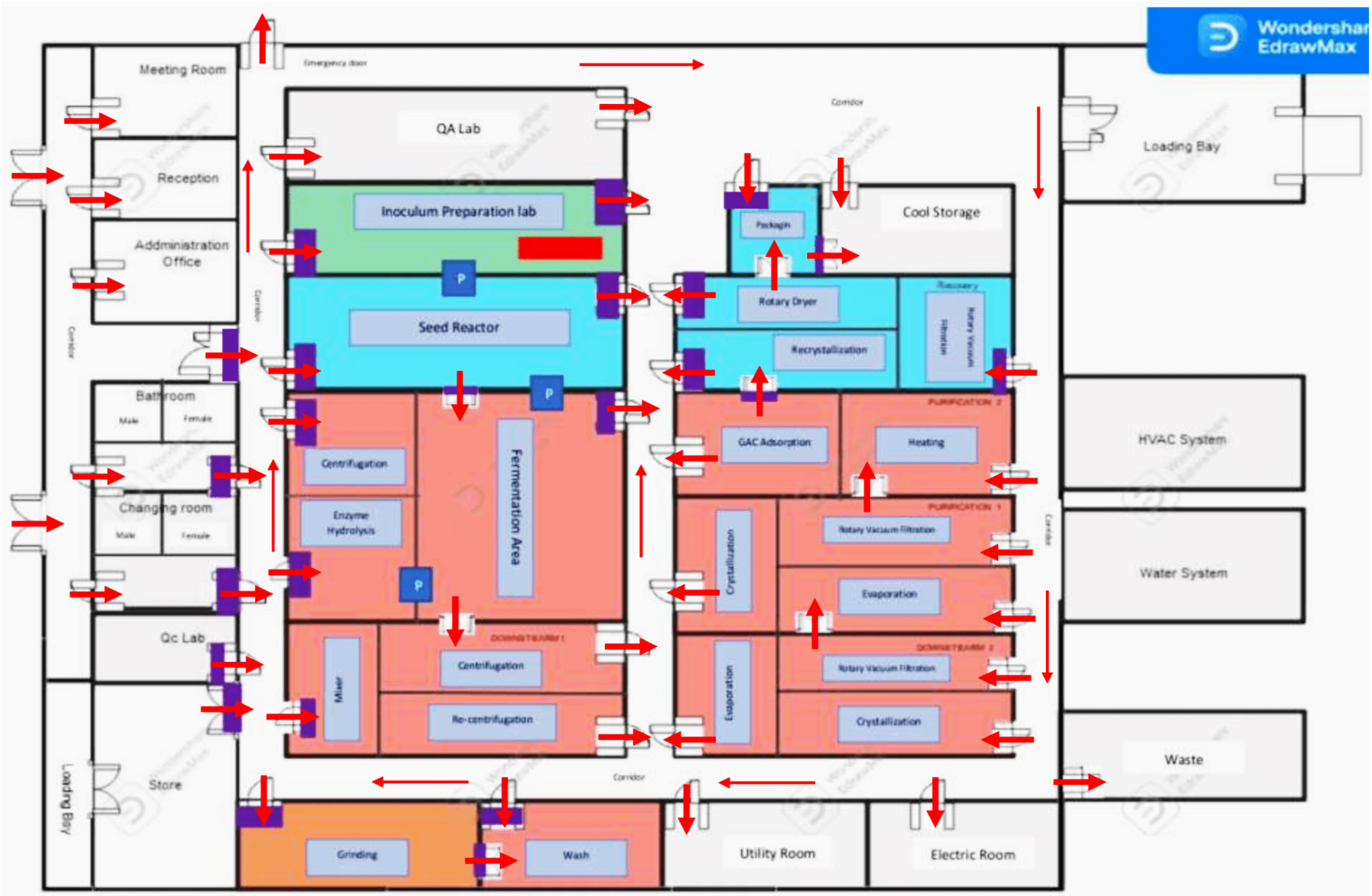
General Plant Layout



Process Flow



Personnel Flow



Material Flow

SAFETY MEASURES

General Workplace Safety Measures

a. Notify the supervisor of any permanent or temporary impairment before starting work.
b. Protect ourselves from potential hazards by wearing personal protective equipment (PPE).
c. Only operate equipment if the workers have been properly trained and authorised.
d. Regardless of severity, report all accidents, near misses, and property damage to the supervisor right away.
e. All tools and equipment should be kept in a designated area.

First Aid Measures

a. Swallowed
    i. Don't make someone vomit if they swallow.
    ii. If vomiting occurs, tilt the patient forward or position them on their left side to keep their airways open and avoid aspiration.
b. Eye
    i. Hold the eyelids apart and continuously flush the eye with running water if this product gets into the eyes.
    ii. Keeping the eyelids apart and away from the eye, as well as occasionally lifting the upper and lower lids, will ensure complete irrigation of the eye.
c. Skin
    i. Remove all contaminated clothing, including footwear, as soon as skin contact occurs.
    ii. Use running water to wash the skin and hair.
d. Inhaled
    i. Remove the patient from the contaminated area if fumes or combustion products are inhaled, and then lay them down.
    ii. Rest and stay warm.

Fire Fighting Measures

a. Suitable extinguishing media: Use water spray, alcohol-resistant foam, dry chemical, or carbon dioxide.
b. Advice for firefighters: Wear self-contained breathing apparatus and protective gloves for firefighting.

Accidental Release Measures

a. Minor spills
    i. Eliminate every ignition source.
    ii. Immediately clean up any spills.
    iii. Avoid contact with skin and eyes.
    iv. Use protective gear to limit personal contact.
    v. Avoid creating dust and use dry cleaning techniques.
    vi. Put in a suitable, labelled container for disposal of waste.
b. Major spills
    i. Inform emergency personnel of the location and nature of hazard.

Safety Measures for Unit Operations

Unit Operation	Safety Measure
Grinder	● Do not use the tool if feeling tired or under the influence of drugs, alcohol, or medication, as this could result in serious personal injury. ● Keep loose clothing, gloves, jewellery, and long hair away from moving parts as they can get caught (Gribbins Insulation, 2016).
Continuous stirred tank reactor	● When the temperature or pressure exceeds the safety limit, use the temperature or pressure relief valve to prevent it from exploding. ● Check the electric system regularly.

Disk-stack centrifuge	● Operate at above 0℃ as below it will make the liquid freeze and lead to machine damage. ● Do not repair the worn-out bowl of a disk-stack centrifuge by welding because welding heat can change the bowl steel's microstructure, making it brittle and reducing its strength (Prabhu, 2022).
Mixer	● To avoid accidental starting and possible injury, locate and lock the power out. ● Check carefully because when tools or bolts are left on moving parts, they can fly off and injure people (Myers Mixers, n.d.).
Evaporator	● Check the condition of the pressure gauges and other indicators on a regular basis to ensure that the boilers and other metallic devices are free of corrosion and that their rubber gaskets are in good working order. ● To avoid air contamination and heat stress, install efficient exhaust ventilation and air conditioning (IIOSH, 2000).
Crystalliser	● Only qualified service personnel should examine and correct problems that necessitate opening the crystallizer's control panel or diagnosing the cause with electrical wires. ● Wear gloves and safety glasses while operating.
Rotary vacuum filter	● Work on the filter should be done by a team of at least two people. ● Before touching the filter or its ancillaries for any reason, always turn it off. ● Never work on the filter when it is hot (Techniplant, n.d.).
Heat exchanger	● To ensure a smooth start-up and shutdown, keep the heat exchanger from being over-pressurized.

	• Stay away from hot surfaces to avoid skin burns (Alaqua, 2022).
Granular activated carbon column	• An oxygen analyzer, combustible gas detector, and hydrogen sulphide detector must all be used to check the atmosphere inside the vessel. • Before entering the vessel, the carbon bed must be completely drained (George et al., 1980).
Rotary dryer	• The fan must be checked to make sure it rotates clockwise, or the wind direction will be incorrect. • In order to avoid harsh vibrations and rain corrosion during transportation, the storage location must be dry and well ventilated indoor rooms (ABC Machinery, n.d.).

ENVIRONMENTAL IMPACT

Global Warming

With 40% of the total impacts, the inoculum and fermentation section is the main contributor, followed by the pretreatment section with 35%. Electricity consumption accounts for 86% of total greenhouse gas emissions during the inoculum and fermentation stages. The breakdown of the greenhouse gas emissions from itaconic acid production: 85% for CO_2, 9% for N_2O, and 5% for CH_4.

Eutrophication

The inoculum and fermentation section is responsible for 55% of the impacts, with the recovery section accounting for 22%. Furthermore, these effects are caused by phosphate in water, which accounts for about 98% of the total (Rebolledo-Leiva et al., 2022).

CONCLUSION

From the above justifications, it successfully demonstrated the design and manufacturing plant for production of itaconic acid (IA) from oil palm empty fruit bunch by *Aspergillus terreus*. This report demonstrated the importance of predicting a large-scale bioprocess focused on improving the overall productivity of a biotechnological product. The plant will produce 4572 kg/day of itaconic acid at a unit cost of RM80/kg which means in a year, it will produce about 1,188,720 kg of itaconic acid. The CAPEX required is estimated at RM 127,793,000 and the OPEX around RM 11,053,248.14. IA current niche market may increase with innovation and specific market targeting, as well as the use of low-cost feedstock. Utilisation of the OPEFB to produce itaconic acid is possible and can reduce the cost to produce useful products of itaconic acid and thus lower the market price.

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