Scale-up simulation and economic evaluation of encapsulated eugenol with casein micelle using spray drying method

A B Wicaksono1, H Hermasnyah1, A Wijanarko1, and M Sahlan1,2*

1 Department of Chemical Engineering, Faculty of Engineering, Universitas Indonesia, Depok, Indonesia
2 Research Center for Biomedical Engineering, Faculty of Engineering, Universitas Indonesia, Depok, Indonesia

Corresponding author: sahlan@che.ui.ac.id

Abstract. Clove or Syzygium Aromaticum is one of Indonesia herbs and spices that have a variety of uses: its oil contained >80% eugenol and was reportedly able to act as anti-microbial, anti-fungal, and anti-inflammatory. Encapsulation was done to protect clove oil from oxidation. Eugenol encapsulation has been studied with casein micelle as its encapsulator. Production of encapsulated eugenol uses 8 unit procedure, with a spray dryer to form a solid product. The economic evaluation shows that the project capital investment is $1,249,000 with the annual operating cost of $1,448,000. Annual production capacity is 6972 kg/year, generating a revenue of $1,634,000 with product price $234.32/kg. Payback period estimated is 6.63 years with IRR 14.30% and NPV $334,000. Sensitivity analysis is conducted using three parameters of payback period, NPV and IRR show that product price fluctuation is the most sensitive variable, followed by capital investment and clove oil price.

1. Introduction
Clove is one of the most popular spices in Indonesia. Clove is an aromatic plant from the Myrtaceae family; a plant originates from Maluku, east Indonesia (1). Clove is commonly used as a flavor enhancer for foods and beverages. Moreover, essential oil from clove is widely used in the pharmacy industry as well as the cosmetic industry. Eugenol mainly dominates the composition of clove essential oil as an active substance. There are several types of research that report the biological activity of eugenol, such as antimicrobial (2), antifungal (3), anti-inflammatory (4), and anti-oxidant (5), which suggest that eugenol, a substance contained in clove essential oil as a beneficial substance and has great potential in its development. Clove essential oil is relatively easy to be obtained on various purity, however, similar to other essential oil, clove essential oil is rather difficult to be handled, because of its properties that are sensitive to sunlight and heat, its also having a short shelf-life if kept under unsuitable condition (6). One of the solutions on handling an essential oil is by the use of encapsulation.

From our previous research regarding encapsulation of eugenol are done in the Universitas Indonesia, has shown good results of encapsulation efficiency through the usage of casein micelle as an encapsulator with 63.86% - 87.99% efficiency. Casein is a protein which dominates the protein composition in milk (±80%). Together with calcium phosphate, casein can form a dispersed colloidal particle that is known as a micelle. The presence of k-casein on casein micelle has an important function to maintain other casein (a and b) from the precipitate and making casein micelle more stable. Casein micelle also has some hydrophobic residue that can strongly adsorb a surface of a droplet, thus making
this characteristic as one of the reasons for high efficiency in the encapsulation of hydrophobic compound. However, research studied by other researcher was done on a laboratory scale, thus giving no information on commercialization.

The computer-based simulation could facilitate the development of process design to scale-up a project as well as to evaluate its economic value. Unlike a petrochemical or other industry that operate with big throughputs and utilizing a continuous process, most bio-processes or pharmaceutical industries operate on a batch or semi-continuous processes (7). During process development, simulation software allows the user to perform several tasks, mainly to represent the entire process, estimate equipment size and perform material and energy balance which contribute massively to the understanding of the entire process (8).

Knowing the potential of clove oil and its eugenol component on many applications that was also supported by various results from previous studies, the objective of this research will focus on the scale-up simulation process design and its economic evaluation to estimate whether production of encapsulated eugenol is economically feasible or not. This research could be used in the future as consideration for encapsulated eugenol commercialization.

2. Material and Methods

2.1. Material

Clove oil was purchased from PT. Natura essential oil, aquadest (PT. Brataco), CaCl₂, NaH₂PO₄, and Na₂HPO₄ were purchased from Merck. HCl and NaOH were purchased from Sygma Aldrich. Cow's milk (Diamond) was purchased from Giant supermarket.

2.2. Casein isolation and eugenol encapsulation

Casein isolation and eugenol encapsulation were prepared as described by Wijayanto et al. (6).

2.3. Simulation

The simulation of the casein isolation and eugenol encapsulation were done with SuperPro Designer Simulator 9.0 (Intelligent, Inc., Scotch Plains, New Jersey).

2.4. Economic Evaluation

2.4.1. Internal Rate of Return. The internal rate of return (IRR) is a rate corresponding to the after-tax cash inflow and outflow on present value. Determination of IRR require a tool and rather difficult to be calculated. IRR is a rate of percentage chosen that will make a value of NPV equal or greater than zero. Calculation of IRR is a trial and error; therefore, if a percentage does not give an NPV of zero or greater, then the percentage will be adjusted down or up. Once an IRR value has been determined, then it will be compared to the required rate of return. If IRR value is equal to or greater than the required rate of return, then the project is acceptable (9).

2.4.2. Net Present Value. Net present value (NPV) is the difference between the present value of inflow and outflow cash over a period. Net present value accounts for the time value of money, thus making it more appropriate when selecting project as it would make the calculation more realistic. By calculating NPV, one can determine whether the present value of a project is greater, equal to or less than zero. If NPV is greater or equal to zero, then a project will be considered meaning that yield is greater or equal than the required rate. However, if its value is less than zero then it would be considered as unfavorable (9).

2.4.3. Payback Period. The payback period (PBP) is some the year required to completely repay the initial capital investment cost that normally disregards the salvage value. PBP could be calculated in two ways; the first one is when the annual cash flows are equal in value for a certain period (9).

2
3. Simulation and Results

3.1. Simulation

SuperPro has been used widely by many known companies such as Bayet Technology Services (Europe and USA), Biogen Idec, Bio-rad Laboratories, Bio-Springer (France) and Chugai Pharmaceuticals (Japan). Aside from companies, there are also over 300 universities worldwide that are using Superpro Designer as a tool for engineering process simulation.

SuperPro Designer is best known for its use in the bioprocessing field, in which it allows its users to simulate a petrochemical or bio-related processes through batch, semi-batch or continuous process, in which account for time dependency and sequencing of events. The system is facilitating the user with broad features starting from end-of-pipe treatment processes, waste minimization, scheduling and economic evaluation. Therefore, SuperPro Designer opens a potential to develop small-scale processes into a bigger one further. The potential application of commercial batch process scheduling and simulation tool throughout the life-cycle of development and commercialization of pharmaceutical products such as tablets was demonstrated by Papavasileiou et al. (8), other SuperPro based research was done by Kawachale N et al. (7) about a scale-up and economics evaluation for the recovery process.

The simulation of this research is divided into two main processes, namely the casein isolation and eugenol encapsulation. Casein isolation process is consisting of procedures to produce solid casein from milk. During this process, the content of milk is precipitated using a rennet which consists of enzymes such as a protease. In the simulation, the first procedure for this process is happening in a stirred reactor tank (R-101). The tank consists of several operations, namely; charge; heat; agitate; react; hold; and CIP (in-place-cleaning). Each batch starts with charging 100L milk into the tank. Inside, the milk was heated to raise the temperature from 25°C to 35°C for 10 minutes, and once the temperature reaches 35°C, HCl was added with the amount of 5L to reduce the pH of milk to 6.4 from ±7.21 while being agitated for 15 minutes. Reducing the temperature and pH of milk will increase the enzymatic activity of rennet (6).

Once the condition reaches 35°C and pH 7.21, 1L of aqueous rennet was added to the system to precipitate casein. The precipitation process was carried out at 35°C, 1 bar pressure and agitated for 45 minutes. After agitation for 45 minutes, 50L of water at 70°C was introduced to deactivate chymosin enzyme, followed by incubation (hold) for 45 minutes to increase the particle size (10). Each charging operation of materials was assumed to take 3minutes. Once incubation is complete, the casein content is separated by using a plate and frame filter (PFF-101), and the cake (casein) was washed with water to eliminate whey content. Separated casein, which still contains some degree of water content was then dried using a freeze dryer (FDR-101), drying process proceeds for 6 hours, and was set for 100 water removal content. The usage of freeze dryer benefits this production process as a micelle in casein.
molecule are vulnerable to a high temperature >100°C to the point it cannot maintain its micellar integrity (Beliciu., 2011). Dried casein was reduced in size by using a grinder (GR-101) to a powder form before entering the next process. Every equipment in this process was included with CIP to clean the residue/waste left in the equipment after it completed its operation.

During the encapsulation process, eugenol will be encapsulated with casein using buffer phosphate pH 10 and CaCl₂ 10% to support the encapsulation process. Similar to the previous tank on casein isolation, the tank in the process consist of transfer; charge; agitate; react, and CIP operation. Each batch starts with 2.3 kg of casein powder resulted from the previous process of entering the tank (R-102), followed by the addition of 30L of buffer phosphate and was agitated for 15 minutes. Buffer phosphate will act as a medium for the encapsulation. After agitation have been completed, 4.9L of clove oil and 8.9L of CaCl₂ 10% were charged into the tank so that encapsulation reaction would proceed. Encapsulation reaction was modeled using a stoichiometry based on the loading capacity of 57%, meaning for 1 gram of encapsulated eugenol, there will be 0.57 grams of eugenol and 43 grams of casein. Reaction happened on 25°C, for 60 minutes. HCl 0.1N and NaOH were also added to the system to keeps the pH at 7. The mixture resulted which already contain encapsulated eugenol was then homogenized for 15 minutes using a high-pressure homogenizer (HG-101) with a pressure drop of 250 bar. Homogenous mixture was then dried using a spray dryer at 180°C; drying process takes 2 hours to be completed. The dried product was then stored on drum storage before being transported to another party. Every equipment in this process was also included with CIP to clean the residue/waste left in the equipment after it completed its operation.

3.2. Operating Cost

Operating cost was the total cost compounded from several annual expenditures needed on a project. The total annual operating cost was calculated to be $1,445,000 with most of the expenditure goes to raw materials with $1,236,000 (85.59%), followed by facility-dependent with $175,000 (12.15%), labour $26,000 (1.77%), laboratory $4000 (0.26%), utilities $3000 (0.22%) and waste treatment/disposal.

3.3. Capital Investment

Most of the cost listed (Figure 4) on the direct fixed capital cost table are derived from the equipment purchase cost multiplied with a factor according to each expenditure. The total direct fixed capital cost is $1,125,000 that consist of the total plant direct cost $440,000 (TPDC), total plant indirect cost $290,000 (TPIC) and contractor’s fee and contingency $394,000 (CFC).
3.4. **Overall economic Evaluation**

The resulting economic evaluation (Table 1) were all summarized in the table above. The project cost a $1,249,000 capital cost with an annual operating cost of $1,448,000 and a unit production cost of $207.66/kg. The product price was calculated to be $234.32/kg thus generating revenues of $1,634,000. The payback period for this project was 6.63 years with IRR of 14.30% thus making this project a feasible project since the WACC calculated was 8.25%.

**Table 1. Overall economical evaluation**

| Items                                | Value   | Unit    |
|--------------------------------------|---------|---------|
| Total Capital Investment             | 1,249,000 | $       |
| Capital Investment Charged to Project| 1,249,000 | $       |
| Operating Cost                       | 1,448,000 | $/yr   |
| Revenues                             | 1,634,000 | $/yr   |
| Cost Basis Annual Rate               | 6.972   | kg MP/yr|
| Unit Production Cost                 | 207.67  | $/kg MP |
| Net Unit Procutin Cost               | 207.67  | $/kg MP |
| Unit Production Revenue              | 234.32  | %       |
| Gross Margin                         | 11.38   | %       |
| Return On Investment                 | 15.09   | years   |
| Payback Time                         | 6.63    | years   |
| IRR (After Taxes)                    | 14.3    | %       |
| NPV (at 8.5% Interest)               | 334000  | $       |
| Total Capital Investment             | 1,249,000 | $       |
| Capital Investment Charged to Project| 1,249,000 | $       |
| Operating Cost                       | 1,448,000 | $/yr   |
| Return On Investment                 | 15.09   | years   |
| Payback Time                         | 6.63    | years   |

3.5. **Sensitivity Analysis**
After simulating the whole production and obtaining the overall economic evaluation, a sensitivity analysis was done to analyze an effect and change (sensitivity) caused by some uncertain variables. Parameters to be evaluated in this research are payback period (PBP), net present value (NPV), and internal rate of return (IRR).

3.5.1. **Capital Investment Fluctuation.** Capital Investment was an initial expenditure expected to launch a project. Capital investment will affect the length of the payback period as well as IRR and NPV thus affecting investor decision in funding this project. The capital investment fluctuation is shown in Table 2.

| Deviation | Product Price per Unit USD | IRR (%) | NPV (USD) | PP (Years) |
|-----------|----------------------------|---------|-----------|------------|
| -20%      | $ 899,200.00               | 20.7%   | $ 606,000.00 | 5.28       |
| -10%      | $ 1,011,600.00             | 17.3%   | $ 474,000.00 | 5.92       |
| -5%       | $ 1,067,800.00             | 15.7%   | $ 408,000.00 | 6.25       |
| 0%        | $ 1,124,000.00             | 14.3%   | $ 334,000.00 | 6.63       |
| 5%        | $ 1,180,200.00             | 12.9%   | $ 264,000.00 | 6.99       |
| 10%       | $ 1,236,400.00             | 11.6%   | $ 194,000.00 | 7.33       |
| 20%       | $ 1,348,800.00             | 9.0%    | $ 36,000.00  | 8.10       |

3.5.2. **Clove Oil Price Fluctuation.** Clove oil is one of the main raw material used. Clove price is one of the components of the operational cost. Even though clove oil cost was only 14.26% of the total raw material cost, its price by far is the most expensive with a current price of $27.24. Unlike any other material listed for this project, clove oil was not very popular, and not many industries are producing clove oil. Hence the increase or decrease might be caused by public demand, and these changes will be taken into account for sensitivity analysis. Clove oil fluctuation as is shown in Table 3.

| Deviation | Clove Oil per Unit USD | IRR (%) | NPV (USD) | PP (Years) |
|-----------|------------------------|---------|-----------|------------|
| -20%      | $ 21.79                | 17.1%   | $ 508,000.00 | 5.94       |
| -10%      | $ 24.52                | 15.7%   | $ 421,000.00 | 6.27       |
| -5%       | $ 25.88                | 14.9%   | $ 377,000.00 | 6.44       |
| 0%        | $ 27.24                | 14.3%   | $ 334,000.00 | 6.63       |
| 5%        | $ 28.60                | 13.5%   | $ 291,000.00 | 6.82       |
| 10%       | $ 29.96                | 12.7%   | $ 247,000.00 | 7.03       |
| 20%       | $ 32.69                | 11.7%   | $ 155,000.00 | 7.48       |

3.5.3. **Product Price Fluctuation.** Product price plays a significant role in the result of all the economic parameters since this project only generating a single product. Encapsulated eugenol product is a new product in public, so it is expected rather to increase or decrease based on markets demand. Product price fluctuation as is shown in Table 4.

| Deviation | Product Price per Unit USD | IRR (%) | NPV (USD) | PP (Years) |
|-----------|-----------------------------|---------|-----------|------------|
| -20%      | $ 187.46                    | 1.2%    | $ -1,828,000.00 | 17.60      |
| -10%      | $ 210.89                    | 3.2%    | $ -614,000.00  | 13.81      |
| -5%       | $ 222.60                    | 6.8%    | $ -96,000.00    | 8.96       |
| 0%        | $ 234.32                    | 14.3%   | $ 334,000.00    | 6.63       |
| 5%        | $ 246.04                    | 20.7%   | $ 733,000.00    | 5.27       |
| 10%       | $ 257.75                    | 27.0%   | $ 1,131,000.00  | 4.36       |
| 20%       | $ 281.18                    | 38.2%   | $ 1,903,000.00  | 3.25       |

3.6. **Sensitivity Analysis Graph**
3.6.1. **Internal Rate of Return Sensitivity.** The result of IRR sensitivity in Figure 5 shows that product price was the most sensitive variable with a range of IRR being 1%-38%, a decrease of -20% from the initial price would cause the IRR to be 1.2% while the maximum increase of 20% results in a 38.2% of IRR. Clove oil price and capital investment as a variable only has a slight effect on the fluctuation of IRR with the minimum IRR (20%) being 11.2% and 9% while the maximum IRR (-20%) 17.11% and 20.17%, both are respectively to the clove oil price and capital investment cost fluctuation. An increase of IRR would make this project more favorable to investors thus making the project more feasible to be executed.

![Figure 4. IRR sensitivity graph](image)

3.6.2. **Net Present Value Sensitivity.** The result of NPV sensitivity analysis in Figure 6 shows that both capital investment cost and clove oil price only has a slight effect to the NPV, it can be seen by comparing the normal NPV (0%) which is $334,000 to the maximum (-20%) and minimum (20%). From the graph above, the difference of NPV value between capital investment and clove oil price fluctuation is very thin, with capital investment being a little more sensitive than clove oil price fluctuation with difference of ±$50,000-$100,000, the value was $606,000 and $508,000 for the maximum, while the minimum is $35,000 and $155,000, for capital investment and clove oil price fluctuations respectively. The most sensitive variable for NPV is product price and decreasing the price by -20% would result in a negative $-1,828,000, while increasing its value by 20% resulting in $1,903,000.

3.6.3. **Payback Period Sensitivity.** The result of PBP sensitivity analysis in Figure 7 shows an almost similar pattern to the NPV analysis, with a slight difference of effect between capital investment and clove oil fluctuation to the length of PBP. The result shows that product price fluctuation is the most sensitive variable compared to the other two with a maximum PBP of 17.60 years and minimum PBP of 3.25 years. PBP resulted from the fluctuation of capital investment and clove oil only differ to a magnitude ±0.7 years for maximum and minimum PBP. The minimum PBP for capital investment and clove oil price fluctuation are 5.28 years and 5.98 years, while the maximum is 8.10 years and 7.48 years, thus making the capital investment as a variable slightly more sensitive than clove oil price.
4. Conclusion
The production simulation of this encapsulated eugenol research consists of 8 unit procedures, with total production time for one batch is 17.35 hours. The economic value for this project is $1,249,000 with annual operating cost of $1,448,000 generating a revenue of $1,634,000. IRR resulted from this project is 14.30% with NPV $334,000 and payback period of 6.63 years. The most sensitive variable based on sensitivity analysis is product price fluctuation, followed by capital investment cost and clove oil price fluctuation.

References
[1] Cortés-Rojas DF, de Souza CRF, Oliveira WP. Clove (Syzygium aromaticum): a precious spice. Asian Pac J Trop Biomed. 2014 Feb 26;4(2):90–6.
[2] Devi KP, Nisha SA, Sakthivel R, Pandian SK. Eugenol (an essential oil of clove) acts as an antibacterial agent against Salmonella typhi by disrupting the cellular membrane. J Ethnopharmacol. 2010;130(1):107–15.
[3] Carrasco H, Raimondi M, Svetaz L, Liberto M Di, Rodriguez M V., Espinoza L, et al. Antifungal Activity of Eugenol Analogues. Influence of Different Substituents and Studies on
Mechanism of Action. *Molecules*. 2012 Jan 19 [cited 2018 Jul 16];17(1):1002–24.

[4] Magalhães CB, Riva DR, DePaula LJ, Brando-Lima A, Koatz VLG, Leal-Cardoso JH, et al. *In vivo anti-inflammatory action of eugenol on lipopolysaccharide-induced lung injury. J Appl Physiol*. 2010 Jan 14;108(4):845–51.

[5] Marília d’AF, Souto OP, Pereira DFS, Jacobsen FT, P. PCM, Quintana OS, et al. *Eugenol derivatives as potential anti-oxidants: is phenolic hydroxyl necessary to obtain an effect? J Pharm Pharmacol*. 2013 Dec 27;66(5):733–46.

[6] Wijayanto A, Putri YRP, Hermansyah H, Sahlan M. *Encapsulation of eugenol from clove oil using casein micelle for solid preparation. AIP Conf Proc*. 2017;1817.

[7] Kawachale N, Kumar A. *Simulation, scale-up and economics of adsorption and membrane-based processes for isoflavones recovery. Chem Eng Res Des*. 2011;89(4):428–35.

[8] Papavasileiou V, Koulouris A, Siletti C, Petrides D. *Optimize Manufacturing of Pharmaceutical Products with Process Simulation and Production Scheduling Tools. Chem Eng Res Des*. 2007;85(7):1086–97.

[9] Donohue S. *Capital budgeting: do private sector methods of budgeting for capital assets have applicability to the Department of Defense*. Naval Postgraduate School; 2005.

[10] Sahlan M, Supardi T. *Encapsulation of Indonesian propolis by Casein micelle. Int J Pharma Bio Sci*. 2013;4(1):297–305.