



## Vacuum Foam-Mat Drying of Red Palm Oil NLC: A Fractional Factorial Screening

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### ABSTRACT

Nanostructured lipid carriers (NLC) derived from red palm oil (RPO) offer promising delivery systems for bioactive compounds, yet their aqueous dispersions remain thermodynamically unstable during extended storage. Vacuum foam-mat drying presents a viable solution for converting liquid NLC into stable powder form while preserving heat-sensitive bioactives at moderate temperatures (50–70°C). This study aimed to systematically identify critical process parameters affecting vacuum foam-mat drying of RPO-based NLC using a Minimum Run Resolution IV fractional factorial design. Five factors were investigated: NLC-to-maltodextrin ratio, maltodextrin concentration, Tween 80 concentration, stirring temperature, and oven temperature, evaluated across 12 experimental runs. Results revealed that the NLC-to-maltodextrin ratio significantly influenced powder yield (66.03–84.95%), while Tween 80 concentration predominantly affected moisture content (8.19–12.87%) and palmitate retention (20.3–110 ppm). Oven temperature emerged as the primary factor controlling particle diameter (400–550 µm) and could induce palmitate degradation at elevated levels. Significant two-factor interactions were observed, particularly between maltodextrin concentration and stirring temperature, affecting palmitate preservation. This screening design effectively reduced the number of experimental runs from 32 to 12 while maintaining a reliable estimation of main effects, providing a foundation for subsequent Response Surface Methodology optimization.

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## **INTRODUCTION**

The application of lipid-based nanoparticles as delivery systems is a significant recent innovation in the food and pharmaceutical sectors. As it stands, the Nanostructured Lipid Carriers (NLC), the second-generation lipid nanoparticles, are the technology of choice (Gordillo-Galeano & Mora-Huertas, 2018; Beloqui et al., 2016). Unlike Solid Lipid Nanoparticles (SLN), NLC systems exhibit a high crystalline structure due to the solid-liquid lipid blends, which also have a superior capacity to retain low levels of actives (Müller et al., 2002; Tamjidi et al., 2013). Of the lipids, red palm oil (RPO) is arguably the most abundant oil to serve as a lipid matrix for NLC emulsification because of its richness as well as its antioxidant components, which include carotenoids (500 - 786 ppm), vitamin E (600 - 1000 ppm), and the metalloids,  $\alpha$ -carotene and  $\beta$ -carotene (82% of total carotenoids) (Tan et al., 2021; Loganathan et al., 2017). NLCs emulsified in RPO oil have good food applications due to RPO's antioxidants.

Previous studies have demonstrated the potential to produce NLCs from blends of RPO and palm stearin, developed via hot homogenization and ultrasonication (Redha et al., 2023; Rohmah et al., 2020). However, it is important to note that NLC aqueous dispersions are thermodynamically unstable. This phenomenon leads to Ostwald ripening and coalescence. This coalescence involved a loss of bioactives during extended storage and reduced storage and coalescence potential for commercialization (Kovacevic et al., 2011).

The possibility of converting liquid nanostructured lipid carriers (NLCs) into powder through drying processes addresses some of the stability issues of liquid carriers. It enables easier storage and transportation, as well as a broader range of applications (Freitas & Muller, 1998). Most traditional drying processes are not suitable for heat-sensitive NLCs. Spray drying is a widely used drying technique, but it operates at a high temperature range of 150-200°C, which is high enough to degrade carotenoids (Mozaffar et al., 2021). Freeze drying is better, but it is prolonged and consumes a large amount of energy (Xia & Kong, 2011). In this case, vacuum foam-mat drying is a better alternative, as it operates at 50-70°C, which is not high enough to cause thermal degradation of the constituents, and it also promotes higher mass transfer rates (Bazyma & Kutovoy, 2005; Hardy & Jideani, 2017). Little work has been done using this technique for NLCs. This technique involves several interacting variables: NLC-to-carrier ratio, surfactant concentration, and foaming and drying temperatures. Each of these variables has a significant effect on the microstructural characteristics of the powder and the balance of the quantity of powder produced, the amount of moisture it contains, the size of the individual particles, and the amount of bioactives it retains. Thus, the high interactivity among the variables permits the adoption of intelligent trial-and-error approaches to pinpoint the crucial variables, rather than conducting exhaustive factorial crossover analyses.

## **LITERATURE REVIEW**

Although several papers and publications have discussed the methodological contributions of fractional factorial designs in the efficient simultaneous investigation of several variables and in the ability to identify main effects of the factors, Mason et al. (2003) note that designs are limited in the

number of experimental trials. In designs of Resolution IV, main effects are not confounded with two-factor interactions, so it is unambiguously possible to estimate the main effects of the factors. This condition provides the designer with reliable screening information, which helps identify critical variables while minimizing the experimental load compared to full factorial designs (Montgomery, 2017). Hence, the purpose of this study is to systematically identify vacuum foam-mat drying of RPO-based NLC factors in a Minimum Run Resolution IV fractional factorial design.

## **METHODOLOGY**

### ***Materials***

Red palm oil was purchased from the local market in Pontianak, Indonesia, and received the palm stearin, maltodextrin with a dextrose equivalent of 10 to 12, Tween 80 with a purity of 99%, and fatty acid methyl ester standards from Sigma-Aldrich (St. Louis, MO, USA). Analytical-grade n-hexane was obtained from Merck (Darmstadt, Germany). The nano-lipid carrier was created according to the guidelines in Zhu et al. (2015), but with a modified process.

### ***NLC Preparation***

NLC was prepared using a hot homogenization method (Zhu et al., 2015) with modifications. Briefly, RPO and palm stearin (5:5, w/w) were melted at 60°C and mixed with the aqueous phase containing Tween 80. The mixture was homogenized (Ultra-Turrax IKA T25, 24,000 rpm, 30 min) followed by sonication (20 kHz, 15 min) and cooled to room temperature.

### ***Vacuum Foam-Mat Drying***

10 g of NLC were combined with maltodextrin and diluted to 50 ml with distilled water, and then homogenized at 5000 rpm for 40 minutes at a specified temperature, when preparing the vacuum foam-mat drying dispersion. This mixture was blended with Tween 80 according to the experimental plan and finally poured in layers 3-5 mm thick onto a surface. The layers were dried in a vacuum oven, Memmert VO400, at a pressure of -0.095 MPa for 5 hours.

### ***Analytical Methods***

When analysing the properties of NLC-RPO powder, the yield was defined as the ratio of the dried powder to the initial solids, and the moisture content was determined by oven drying at 70°C until a constant weight, as per Goula and Adamopoulos (2010). GC-MS analyzed the total palmitate content and fatty acid profile with a temperature programme that went from 50 to 250 degrees Celsius at a 4 degrees Celsius per minute rate, using a 30-meter, 0.25 mm diameter, 0.25-micron thick HP-5 MS column and the HP 6890/5973 MSD system, which is in accordance with Hadaruga et al., 2008. Particle diameter was measured with a laser diffraction system, the Shimadzu SALD-2200 in dry mode, using the method outlined by Koyama and Kitamura (2014).

### ***Statistical Analysis***

To identify the factors affecting vacuum foam-mat drying of the NLC-RPO powder, a fractional factorial design with minimum run IV resolution was used. The degrees of the factors are shown in Table 1, and the complete experimental plan is shown in Table 2. The results of 12 runs were scrutinised using ANOVA with  $\alpha = 0.05$  in Design-Expert 13.0 (Stat-Ease Inc., Minneapolis, Minnesota,

USA). The main factors in the drying process were identified using Pareto charts and normal probability plots.

Table 1. Experimental Factors and Coded Levels

Factor	Variable	Low (-1)	High (+1)	Unit
A	NLC:MD ratio	1:50	1:10	w/w
B	MD concentration	40	45	%
C	Tween 80	0	0.5	%
D	Stirring temperature	25	50	°C
E	Oven temperature	50	70	°C

MD: maltodextrin; NLC: nanostructured lipid carrier

Table 2. Experimental Design Matrix (Minimum Run Resolution IV)

Run	A	B	C	D	E
1	-1	-1	-1	-1	+1
2	+1	-1	-1	-1	-1
3	-1	+1	-1	-1	-1
4	+1	+1	-1	-1	+1
5	-1	-1	+1	-1	-1
6	+1	-1	+1	-1	+1
7	-1	+1	+1	-1	+1
8	+1	+1	+1	-1	-1
9	-1	-1	-1	+1	-1
10	+1	-1	-1	+1	+1
11	-1	+1	-1	+1	+1
12	+1	+1	-1	+1	-1

Note: A: NLC: Maltodextrin ratio; B: Maltodextrin concentration; C: Tween 80 concentration; D: Stirring temperature; E: Oven temperature. See Table 1 for coded levels.

## RESULTS AND DISCUSSION

### *Identification of Significant Process Factors*

To understand the factors affecting the vacuum-foam-mat-dried NLC-RPO powder quality, we used a fractional factorial design. The first round of experiments in Table 3 included 12 combinations of experimental parameters, yielding % yield values ranging from 66.03 to 84.95, moisture levels of 8.19 to 12.87%, and total palmitate content ranging from 20.3 to 110 ppm. The evidence of variability presented in Table 3 justified the complete design. These results formed the basis for the next round of factorial design, involving fewer variables, for the first response surface contours optimization study.

Table 3. Yield, Moisture Content, and Total Palmitate Content of NLC Powder

Run	NLC:MD	Tween 80 Conc. (%)	Oven Temp. (°C)	Stirring Temp. (°C)	Maltodextrin Conc. (%)	Yield (%)	Moisture Content (%)	Total Palmitate (ppm)
1	1:50	0.5	50	25	45	70.06	10.09	76.5
2	1:20	0.5	50	50	45	72.67	11.89	80.4
3	1:20	0.5	50	25	45	76.45	12.87	110.0
4	1:20	0.5	70	50	40	66.67	8.41	68.0
5	1:20	0	50	25	40	68.78	11.49	40.7
6	1:50	0	70	50	40	68.90	8.66	72.0
7	1:50	0.5	50	50	40	67.88	8.19	57.9
8	1:50	0.5	70	50	45	75.52	10.75	47.9
9	1:50	0	70	25	40	66.03	8.76	20.3
10	1:50	0	50	50	45	75.69	11.04	49.0
11	1:20	0.5	70	25	40	84.95	9.14	66.8
12	1:20	0	70	25	45	84.22	9.31	83.0

Note: Ratio NLC:MD = Oleoresin:Maltodextrin Ratio; Tween 80 Conc. = Tween 80 Concentration; Oven Temp. = Oven Temperature; Stirring Temp. = Stirring Temperature; Maltodextrin Conc. = Maltodextrin Concentration

Four of the statistical models presented had meaningful, statistically significant p-values ( $\leq 0.05$ ) and, thus, could be considered reasonable empirical models (Table 4). Regarding yield response, NLC: maltodextrin model with a 0.0011 p-value is statistically significant due to model pedal coupling with A, D, E factors and two-factor interactions: BC, BD, BE, and DE. This task is relatively complex because, while the foam is being dried, it is also being foamed, a phenomenon further complicated by the vacuum.

Table 4. Statistical Models and Significant Factors for NLC Powder Response Variables

No	Response Variable	Model	Significant Factor	P-value model
1.	Yield	Yield=A+C+D+E+BD+BE+DE	Maltodextrin:NLC ratio, Tween 80 concentration, stirring temperature and oven temperature	0.0011
2.	Moisture content	Moisture content=C+BC+CD	Tween 80 concentration	0.0025
3.	Particle diameter	Particle diameter=E+AD	Oven temperature	0.0119
4.	Total palmitate content	Total palmitate=C+E+BC+BD+BE	Tween 80 concentration and oven temperature	0.0384

Looking at the simplified model for Tween 80 concentration (C), found a significant correlation with moisture content, with contributions from the BC and CD interactions. Since Tween 80 plays a significant role in the interfacial properties and the distribution of water in the foam matrix, this is no surprise.

The particle diameter, however, is primarily controlled by oven temperature, E, and the interaction with the adhesion parameter, AD, at a significance level of  $p = 0.0119$ . This phenomenon suggests that the rate of thermal kinetics determines the particle formation and collapse in the foam. The

palmitate retention model provided insight into the protective nature of the formula in bioactive systems, thanks in part to the carrier ratio (A), surfactant concentration (C), and heat (E), all of which are fundamental for preserving bioactivity.

The Pareto charts and half-normal plots in Table 5 basically told the same story, with some factors well above the Bonferroni limit. The fractional factorial approach we took cut down the number of necessary runs to a manageable 12. Compared to the 32 that would have been required for a complete factorial Design, this still gives us a perfectly reliable capability to calculate main effects and two-factor interactions without any overlap.

Table 5. Recapitulation of Significant Factors Identification

Response Variable	Normal Plot Indication	Pareto Chart Result	Significant Factor (ANOVA)	Effect Direction
Yield (%)	A, C, D, E, DE deviated from normal line	D, A, DE exceeded Bonferroni limit	A (Ratio NLC:MD)	1:20 → Higher
Moisture Content (%)	C, BC, CD deviated from normal line	Only C exceeded Bonferroni limit	C (Tween 80)	0.5% → Lower
Total Palmitate Content (ppm)	A, C, E, BC, BD, BE deviated from normal line	BD, E, C, BC exceeded Bonferroni limit	A (Ratio NLC:MD)	1:20 → Higher
			C (Tween 80)	0.5% → Higher
			E (Oven Temp.)	70°C → Lower
			BD (Interaction)	Complex
Particle Diameter (µm)	E, AD deviated from normal line	Only E exceeded Bonferroni limit	E (Oven Temp.)	70°C → Smaller

Note: A = NLC:Maltodextrin Ratio; B = Maltodextrin Concentration; C = Tween 80 Concentration; D = Stirring Temperature; E = Drying Temperature; BD = Interaction between B and D

### Yield

The response surface plots in Figure 1 are informative for examining the effect of the NLC: maltodextrin ratio on the yield of a particular product. From a relatively low NLC: maltodextrin ratio perspective, Figure 1a shows a highly curved response surface, with yields between 60 and 90%. The maximum yield is achieved when the maltodextrin concentration is high, and the Tween 80 concentration is low. Well-known as a sign of a strong interaction between variables, the elliptical contours in this case are a feature of the main effect of the NLC: maltodextrin ratio on the product’s yield. On the other hand, at a higher maltodextrin: NLC ratio, the response surface, as shown in Figure 1b, is much flatter, suggesting reduced interaction effects between the formulation variables.

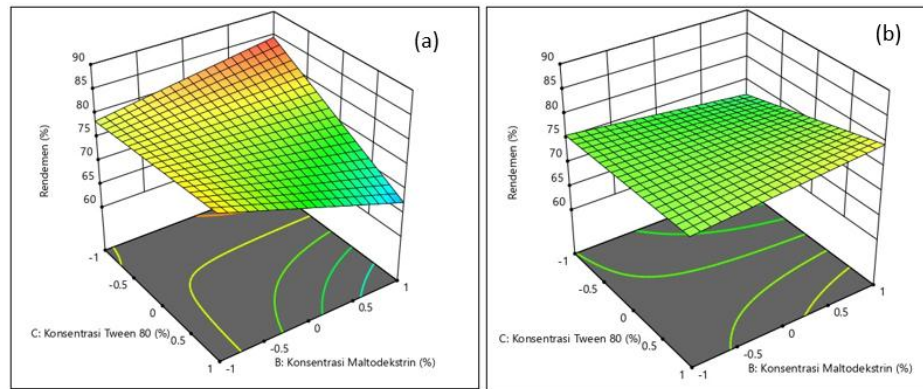


Figure 1. Response surface of the main effect of NLC: Maltodextrin ratio on NLC powder yield at (a) low NLC: Maltodextrin ratio (1:50) and (b) high maltodextrin: NLC ratio (1:20)

In the interaction between maltodextrin and NLCs, maltodextrin's role as a carrier is key to the observed response patterns. Higher levels of carrier material, as evidenced by lower maltodextrin: NLC ratios, bolster the foam structure, leading to more efficient moisture removal and, as a result, higher powder recovery. Tween 80's interaction with maltodextrin and NLCs is quite remarkable, as it stabilizes the foam via steric stabilization. The response patterns are best encapsulated by Ekpong et al. (2016). The issue, however, is that at higher maltodextrin: NLC ratios, the excess carrier matrix clogs everything, resulting in decreased performance. In general, according to the study by Tamjidi et al. (2013), adding more maltodextrin to the mixture at a particular ratio leads to the maltodextrin effects becoming less pronounced as the mixture becomes more viscous.

**Moisture Content**

As shown in Figure 2a, flat response surfaces were observed when no Tween 80 was used, with moisture levels ranging from 7% to 9%. However, with a higher concentration of Tween 80 at 0.5% in Figure 1b, the results showed consistently lower levels of moisture content, between 5% and 7%, with minimal curvature on the response surface, signifying that the effect of maltodextrin on NLC ratio and maltodextrin itself was not nearly as strong as that of Tween 80 concentration.

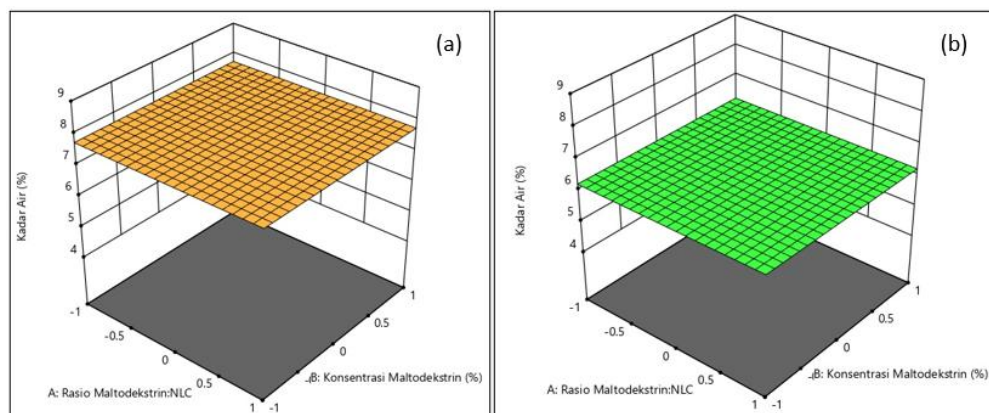


Figure 2. Response surface of the main effect of Tween 80 concentration on moisture content at (a) low Tween 80 concentration (0%) and (b) high Tween 80 concentration (0,5%)

The moisture level decreases as the Tween 80 concentration increases when using Tween 80. This emulsifier is famous for its ability to control the properties of foam that forms during the drying process. Smoothing the air-water interface, Tween 80 produces smaller, more uniform bubbles that facilitate more efficient evaporation. Thanks to its ability to stabilize the foam, almost all the water can be removed, leaving the final powders with very little moisture. This advantage is especially true for lipid-based formulations, which are sensitive to moisture levels and can be rendered unstable if they contain too much water.

### Particle Diameter

From the particle diameters at different oven temperatures, it is clear that the response surface is strongly influenced by temperature, as shown in Figure 3. Coming fast on the heels of the calibration, at the lower oven temperature of 50 degrees Celsius, the surface had a very flat appearance, with particles measuring 500-550 micrometres (Figure 3a). In contrast, the surface at the higher oven temperature of 70 degrees Celsius had a similar flat shape, but over a smaller scale, at around 400 to 500 micrometres (Figure 3b). The almost horizontal gradient of both surfaces suggests that particle size is not affected by the NLC: maltodextrin ratio or maltodextrin concentration, but is primarily controlled by the oven temperature, which is why this is a crucial factor in determining the physical properties of NLC powders.

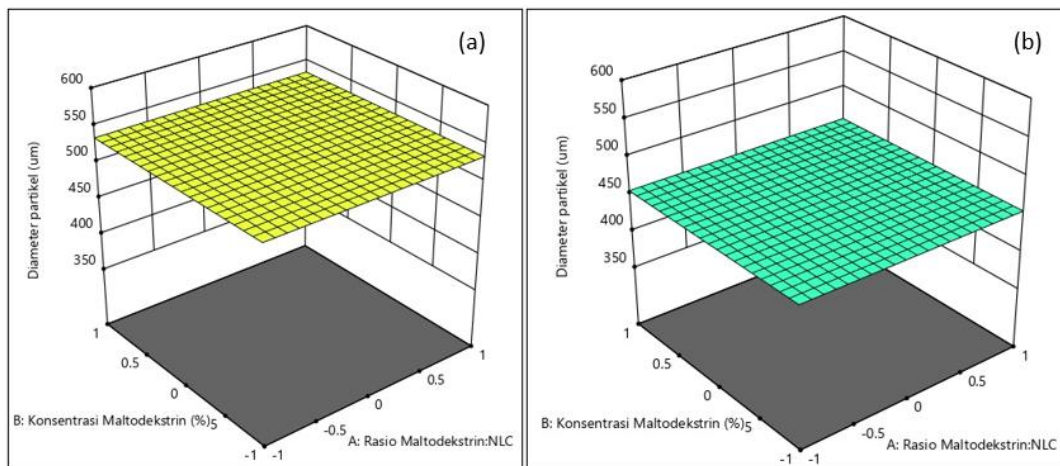


Figure 3. Response surface of the main effect of oven temperature on particle diameter at (a) low oven temperature (50°C) and (b) high oven temperature (70°C)

When increasing the oven temperature, the smaller particle diameters observed can be attributed to the drying process speeding up at higher temperatures, causing the foam to shrink rapidly and form denser, more compact powder particles. Upon emerging from the oven, this foam matrix is also more tightly packed, thanks to the increased heat, as Ekpong et al. (2016) found, thereby reducing porosity and the overall size of the dried particles. Well-known particle surfaces do not change much at either temperature, indicating that once the oven temperature stabilises, other ingredients in the formula have little impact on the final particle size.

### Total Palmitate Content

Tween 80 concentration and oven temperature were the leading contributors to the process, according to a fractional factorial design, in the study of NLC formulation (Figure 5). The response surface plots depicted two very different patterns. At a Tween 80 concentration of 0%, the surface slope is sharp. Within this range, palmitate levels can vary from 120 ppm to 20 ppm and are heavily influenced by the NLC: maltodextrin ratio and maltodextrin concentration. However, at a Tween 80 concentration of 0.5%, the surface took on a much smoother appearance, and the levels of palmitate remained in the 60-80 ppm range, largely unaffected by changes in the other formulation components.

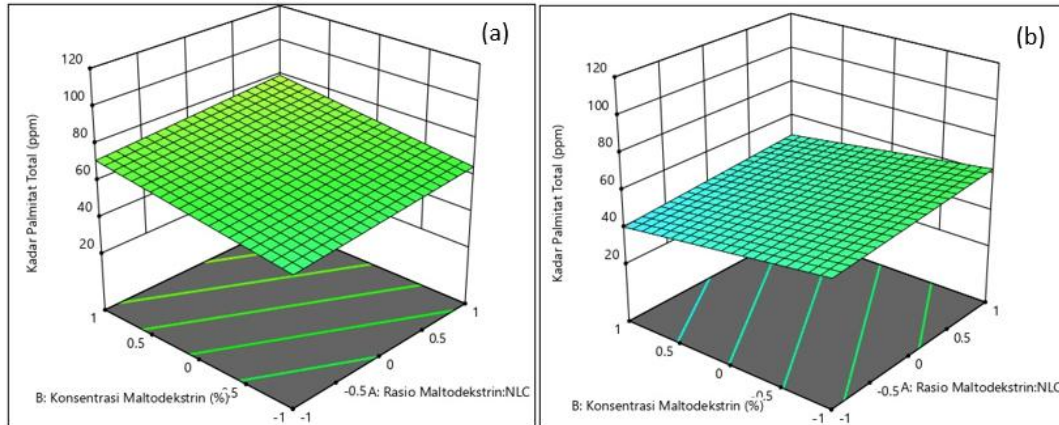


Figure 4. Response surface of the main effect of Tween 80 concentration on total palmitic content at (a) low Tween 80 concentration (0%) and (b) high Tween 80 concentration (0,5%)

Regarding palmitate retention, higher concentrations of Tween 80 form a protective interfacial film around lipid nanoparticles, acting as a steric stabilizer to prevent oxidation and thermal degradation. This has been observed in Loo et al. (2020). Moreover, the surfactant reduces interfacial tension during foam formation, creating uniform bubble structures that rapidly eliminate moisture while limiting the exposure of the encapsulated palmitate to the oven temperature.

The flattened response surface at 0.5% Tween 80 (Figure 5) suggests that stabilisation of palmitate becomes the dominant factor in its preservation, completely masking the effects of any changes in the other processing parameters. The reaction surface maps showed a very different picture of the total palmitate level at different oven temperatures. From a relatively flat base at 50 degrees Celsius, the surface of the palmitate content appeared consistent across various NLC-to-maltodextrin ratios and maltodextrin levels. However, it showed a more pronounced slope at 70 degrees Celsius, where palmitate levels varied greatly and were sensitive to the other formulation factors.

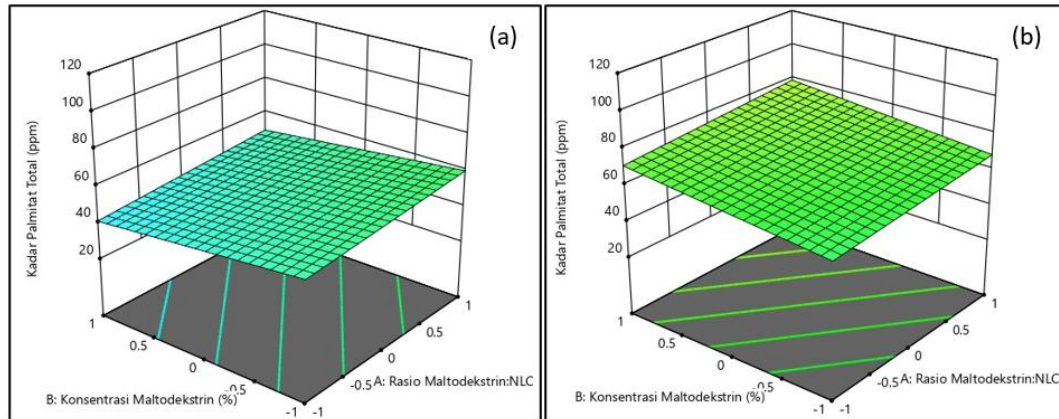


Figure 5. Response surface of the main effect of oven temperature on total palmitic content at (a) low oven temperature (50°C) and (b) high oven temperature (70°C)

The level of palmitate retention drops off rapidly as the temperature rises, due to the destructive effects of heat on lipid-based compounds during drying. Elevated temperatures trigger oxidative reactions in fatty acids, producing peroxides and, in turn, shattering the molecular structure of palmitate. The high heat can also damage the intricate structure of the NLC matrix, reducing its ability to shield palmitate from the stresses of drying, as per Tamjidi et al. (2013).

A study by Pezeshki et al. (2019) shows that a flattened response at 50 degrees Celsius indicates that running the drying process at a relatively low temperature effectively eliminates thermal degradation and removes just the right amount of moisture. Vacuum foam-mat drying at reduced temperature is thus found to be more efficient in retaining palmitate content. However, it is a trade-off, and we have to weigh bioactive retention against drying time.

## CONCLUSIONS AND RECOMMENDATIONS

The Minimum Run Resolution IV fractional factorial design provided a clear picture of the five key factors influencing the vacuum foam-mat drying of NLC-RPO powder. The established NLC-to-maltodextrin ratio determines the final product yield, while increasing Tween 80 led to higher moisture content and greater preservation of palmitate in the finished product. The particle size of the NLC powder was found to be influenced by oven temperature and could also cause palmitate degradation at higher levels. There is a beautiful interplay between maltodextrin concentration and stirring temperature that affects the palmitate levels. The existence of significant two-factor interactions is clear-cut and particularly evident in the relationship between maltodextrin concentration and stirring temperature, as both variables affect palmitate levels.

Since we are using a 5-factor model, we will now delve deeper into the conditions required to meet all our requirements, including yield, palmitate retention, moisture content, and particle size. We will then use Response Surface Methodology to fine-tune and check the storage stability of our final NLC powder formulation.

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