Dataset on the optimization by response surface methodology for the synthesis of silver nanoparticles using Laxitextum bicolor mushroom

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1 Abstract

This study presents an exploratory analysis of the optimization process for synthesizing silver nanoparticles (AgNPs) with an aqueous extract of the Laxitextum bicolor mushroom. Central to this paper is the deconstruction of the Response Surface Methodology (RSM) employed in the original research. By examining the computational procedures and statistical interpretations within the proprietary software STAT-EASE, this work aims to elucidate the specific steps that led from experimental design to the determination of optimal synthesis parameters.

2 Introduction

The optimization of silver nanoparticle (AgNP) synthesis from sources like Laxitextum bicolor is a critical step in advancing green nanotechnology. A foundational study successfully optimized this process using Response Surface Methodology (RSM) in Minitab software (1). This paper undertakes a comparative analysis by re-evaluating their experimental data in a different powerful tool: STAT-EASE. The objective is two-fold. First, we'll assess the consistency of optimization outcomes when processed by different analytical software. Second, we'll critically examine the specific criterion goals set by the original researchers. Their goal was: to maximize the Surface Plasmon Resonance (SPR) intensity at 420nm while simultaneously setting the model to afford sufficient amount of nanometals (minimize Extracts), a crucial consideration for eliminating further experimental trails and errors (1).

In this re-evaluation, we will also discuss the importance of setting the criteria for both the mushroom extract and $AgNO_3$ to "minimize," as this aligns with the principles of green chemistry by reducing cost and conserving resources.

This is based on a prior study (2).

3 Methods

This exploratory study performs a computational replication and comparative analysis of the optimization data from the original research paper using STAT-EASE Software (1). The methodology was conducted as follows:

1. Experimental Design Reconstruction: The Central Composite Design (CCD) used in the source study was reconstructed within the STAT-EASE (Design-Expert) software [@Camacho2022]. The design consisted of 32 experimental runs based on five independent variables (pH, temperature, time, volume of extract, and volume of $AgNO_3$). The experimentally obtained Surface Plasmon Resonance (SPR) values from the original paper were entered as the response data for each corresponding run [@Camacho2022].

2. Model Fitting and Analysis: Analysis of Variance (ANOVA) was performed to assess the statistical significance of a quadratic polynomial model. The model's adequacy was evaluated by examining the coefficient of determination (R^2), adjusted R^2, predicted R^2, and the lack-of-fit test. This process identified the significant linear, quadratic, and interaction terms influencing the SPR response.

3. Optimization and Comparison: Using the numerical optimization feature in STAT-EASE, the optimal conditions were predicted based on the criterion goals established in the original study (i.e., maximizing SPR while conserving reagents) [@Camacho2022]. The resulting statistical model, significant factors, and predicted optimal conditions generated by STAT-EASE were then systematically compared to the findings originally reported using Minitab software [@Camacho2022].

4 Results

This study successfully replicated the Response Surface Methodology (RSM) analysis from the foundational paper by Javier and Camacho (2022) using STAT-EASE software (1). The re-computation of the original 32 experimental runs yielded results that are in strong agreement with the findings originally reported using Minitab (1).

4.1 Model Fit and ANOVA Validation

The statistical validity of the refined quadratic model was confirmed, showing a near-perfect match between the two software analyses. The Analysis of Variance (ANOVA) results, as detailed in Table 2, were identical, confirming that the model is highly significant ($p < 0.001$) and that the Lack of Fit is not significant ($p = 0.223$). This indicates that the model is an excellent fit for the experimental data, irrespective of the software used.

The goodness-of-fit statistics also demonstrated strong congruence:

- R^2: The STAT-EASE analysis produced an R^2 value of 0.9854.
- Adjusted R^2: The adjusted R^2 was 0.9734.
- Predicted R^2: The predicted R^2 was 0.9249.

These values are identical to those reported in the original study, confirming the model's high predictive accuracy (1).

4.2 Diagnostic Plot Confirmation

The diagnostic plots generated by STAT-EASE (Figures 1 and 2) validate the assumptions of the regression model. The Normal Probability Plot of Residuals shows the data points aligning closely along a straight line, confirming the normal distribution of residuals. The Residuals vs. Predicted values plot displays a random scatter, indicating no systematic errors or underlying patterns. These plots are visually consistent with those presented in the original Minitab analysis (1).

4.3 Predicted Optimal Conditions and Response

The primary outcome of the optimization process was successfully replicated. The optimal conditions for the synthesis of silver nanoparticles predicted by STAT-EASE were identical to the original findings (1):

- pH: 10
- Temperature: 55°C
- Time: 180 min
- Volume of Extract: 1.5 mL
- Volume of $AgNO_3$: 20 mL

Under these conditions, the STAT-EASE model predicted a maximum Surface Plasmon Resonance (SPR) absorbance of 3.45. This value is in very close agreement with the experimentally verified absorbance of 3.40 reported by Javier and Camacho, representing a negligible difference of only 1.4% (1). The 3D surface plots (Figure 3) generated by STAT-EASE also visually correspond to those from the original study, illustrating the same interaction effects between variables (1).

Table 1: Summary of experiment runs and peak intensity response at 420 nm absorbance for *L. bicolor* optimization.

Sci Order	**Run Order**	**pH**	**Temp** ($^{\circ}$C)	**Time** (min)	**Extract (mL)**	**$AgNO_3$ (mL)**	**Peak Intensity**
23	1	8.5	20	120	0.5	25	0.16569
12	2	10.0	55	60	3.5	20	2.88199
31	3	8.5	40	120	2.5	25	0.85780
13	4	7.0	25	180	3.5	30	6.54736
25	5	8.5	40	120	2.5	15	1.37705
26	6	8.5	40	120	2.5	35	0.61652
8	7	10.0	55	180	1.5	20	3.44737
10	8	10.0	25	60	3.5	30	1.16447
28	9	8.5	40	120	2.5	25	0.99561
27	10	8.5	40	120	2.5	25	1.12450
7	11	7.0	55	180	1.5	30	0.21446
17	12	5.5	40	120	2.5	25	0.56311
1	13	7.0	25	60	1.5	30	0.19605
15	14	7.0	55	180	3.5	20	1.00559
16	15	8.5	40	120	2.5	25	0.63072
11	16	7.0	55	60	3.5	30	0.32032
4	17	10.0	55	60	1.5	30	1.62778
24	18	8.5	40	120	4.5	25	0.60132
32	19	8.5	40	120	2.5	25	0.94162
20	20	8.5	70	120	2.5	25	2.13783
22	21	8.5	40	240	2.5	25	1.37880
16	22	10.0	55	60	3.5	30	4.08624
21	23	8.5	40	120	2.5	25	0.69836
30	24	8.5	40	120	2.5	25	0.93115
14	25	10.0	25	180	3.5	20	2.05509
4	26	10.0	25	60	1.5	20	1.17515
18	27	11.5	40	120	2.5	25	4.23401
5	28	7.0	25	180	1.5	20	0.34080
6	29	7.0	25	60	3.5	20	0.19605
6	30	10.0	25	180	1.5	30	0.98364
3	31	7.0	55	60	1.5	20	0.57730
29	32	8.5	40	120	2.5	25	1.21265

Table 2: Range and Levels of 5 level Half-FDractional Central Composite Design

Variable	**-2**	**-1**	**0**	**+1**	**+2**
pH	5.5	7.0	8.5	10.0	11.5
Temp. ($^{\circ}$C)	10	25	40	55	70
Time (min)	0	60	120	180	240
Extract (mL)	0.5	1.5	2.5	3.5	4.5
$AgNO_3$ (mL)	15	20	25	30	35

Table 3: ANALYSIS OF VARIANCE (ANOVA)

Source	**dF**		**Sum of Squares**		**Mean Square**		**F-value**		**P-value**	
	Orig.	Rec.	Orig.	Rec.	Orig.	Rec.	Orig.	Rec.	Orig.	Rec.
Model	14	14	35.28	35.28	2.52	2.52	82.13	82.01	<0.0001	<0.0001
A-pH	1	1	18.99	18.99	18.99	18.99	619.00	618.12	<0.0001	<0.0001
B-Temperature	1	1	4.62	4.62	4.62	4.62	150.70	150.43	<0.0001	<0.0001
C-Time	1	1	1.46	1.46	1.46	1.46	47.56	47.70	<0.0001	<0.0001
D-Extract Vol	1	1	0.79	0.7876	0.79	0.7876	25.67	25.63	<0.0001	<0.0001
E-$AgNO_3$ Vol	1	1	0.68	0.6814	0.68	0.6814	22.21	22.18	<0.0001	0.0002
AB	1	1	2.11	2.11	2.11	2.11	68.76	68.66	<0.0001	<0.0001
AC	1	1	0.52	0.5208	0.52	0.5208	16.97	16.95	<0.0001	0.0007
AD	1	1	0.31	0.3106	0.31	0.3106	10.12	10.11	0.005	0.0055
BC	1	1	0.29	0.2940	0.29	0.2940	9.58	9.57	0.007	0.0066
CD	1	1	0.18	0.1817	0.18	0.1817	5.92	5.92	0.026	0.0264
DE	1	1	0.39	0.3852	0.39	0.3852	12.55	12.54	0.002	0.0025
A^2	1	1	3.60	3.63	3.60	3.63	118.20	118.10	<0.0001	<0.0001
B^2	1	1	0.27	0.2732	0.27	0.2732	8.89	8.89	0.008	0.0084
D^2	1	1	0.81	0.8268	0.81	0.8268	26.97	26.91	<0.0001	<0.0001
Residual	17	17	0.52	0.5223	0.03	0.0307				
Lack of Fit	12	12	0.44	0.4338	0.036	0.0361	2.04	2.04	0.223	0.223
Pure Error	5	5	0.09	0.0886	0.018	0.0177				
Cor Total	31	31	35.80	35.80						
R^2	0.9854									
Adj. R^2	0.9734									
Pred. R^2	0.9249									
Adeq Precision	35.0510									

Table 4: Solutions table on a 100 max solutions.

Number	pH	Temperature (°C)	Time (min)	Extract Vol (mL)	$AgNO_3$ Vol (mL)	SPR band	Desirability	Status
1	**10.00000**	**55.0000**	**180.00**	**1.50000**	**20.0000**	**20.0000**	**0.931**	**Selected**
2	9.99214	54.9999	180.00	1.50000	20.0672	3.43030	0.927	
3	9.99999	54.8889	175.55	1.50004	20.0000	3.40302	0.927	
...	...	...	...	...	...	....	...	
98	9.97941	28.4446	180	2.27426	20	2.09524	0.662	
99	10	30.7503	180	2.42722	20	2.28806	0.654	
100	9.999956	25.063	60	2.13075	20.0606	1.38089	0.588	

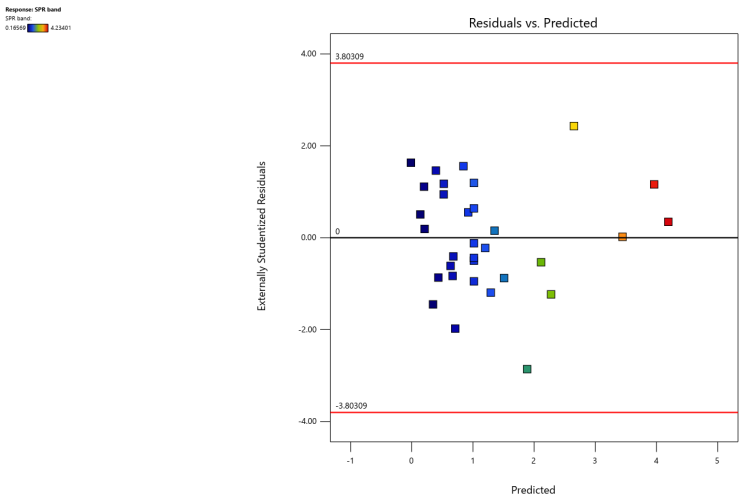

Figure 1: . Standard residual vs fitted value of SPR for the refined model.

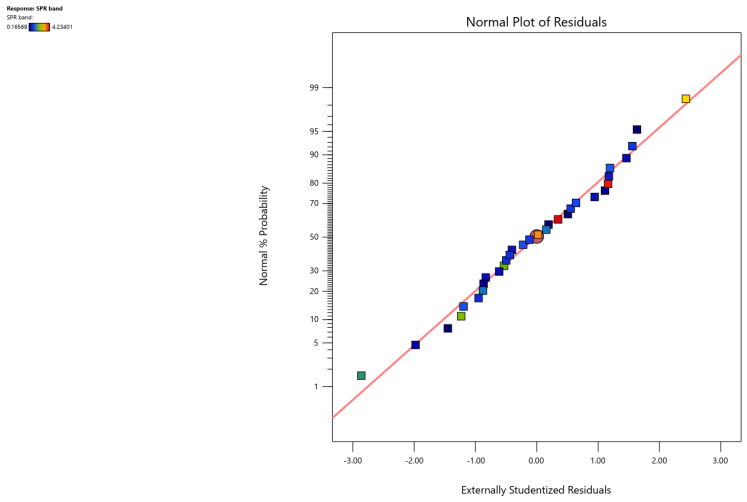

Figure 2: Normal probability plot of standardized residuals for the refined model.

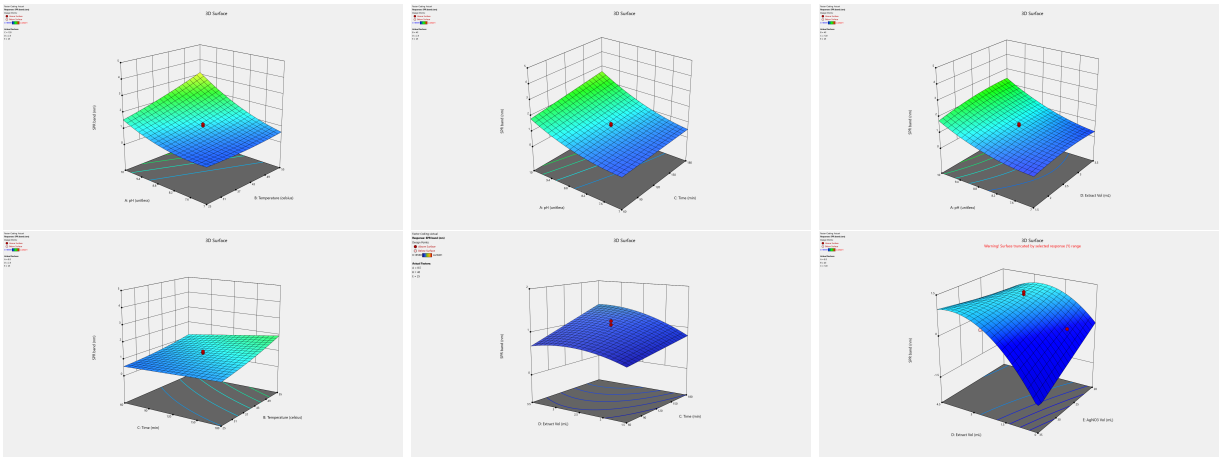

Figure 3: Surface plots of significant model terms: (Top-Left) pH∗Temperature, (Top-Center) pH∗Time, (Top-Right) pH∗Volume Extract, (Bottom-Left) Temperature∗Time, (Bottom-Center) Time∗Volume Extract, (Bottom-Right) Volume Extract*AgNO3 Volume.

5 Discussion

This exploratory study successfully validated the statistical findings of Javier and Camacho (2022) on the optimization of silver nanoparticle (AgNP) synthesis by replicating their analysis using STAT-EASE software (1). The original research employed Minitab 17 to process data from a central composite design (CCD) (1). Our re-evaluation confirms that the results generated in STAT-EASE strongly agree with the original Minitab calculations, demonstrating the software-independent validity of the findings. The analysis in STAT-EASE successfully replicated the refined reduced quadratic model developed by the original authors (1). However, beyond simple confirmation, this analysis provides a deeper insight into the model's implications for scientific reproducibility and green chemistry.

A central outcome of this re-evaluation is the powerful confirmation of scientific reproducibility. The fact that two distinct proprietary software packages yielded identical optimal conditions (pH: 10, Temperature: 55°C, Time: 180 min, Volume of Extract: 1.5 mL, and Volume of $AgNO_3$: 20 mL) underscores the robustness of the

original experimental design and conclusions (1). This software independence is critical, as it demonstrates that the scientific findings are not an artifact of a specific program's algorithm. It reinforces the integrity of the Response Surface Methodology (RSM) approach used and builds confidence that the predicted outcomes are a true function of the experimental variables, not the computational tool.

Furthermore, this analysis fulfills the objective to critically examine the optimization criteria, particularly the goal of minimizing reagents as a principle of green chemistry. The model's prediction of an optimal extract volume of 1.5 mL, the lowest level tested in the design is highly significant (1). It proves that maximizing the Surface Plasmon Resonance (SPR) at peak intensity 420nm does not require a large volume of the biological extract (1). This is a crucial finding for sustainable nanotechnology, as it promotes resource conservation and dramatically lowers the potential cost of synthesis, making the process more economically viable and environmentally friendly. This outcome validates that the goals of high yield and green chemistry are not mutually exclusive in this process.

Finally, the model's predictive accuracy is noteworthy for its practical applications. The STAT-EASE model predicted a maximum SPR absorbance of 3.45 , which represents a negligible difference of only 1.4% from the experimentally verified absorbance of 3.40 reported by Javier and Camacho (1). This close correlation is not merely a statistical success; it signifies that the computational model is a highly reliable proxy for real-world laboratory experiments. Researchers can confidently use this validated model to predict outcomes, thereby reducing the need for costly and time-consuming trial-and-error experiments, which was a key consideration of the original study (1). The minor variation is easily attributable to the slight differences in regression algorithms or rounding protocols between software packages, further strengthening the conclusion that the model itself is fundamentally sound.

6 Conclusion

This exploratory study successfully validated the findings of Javier and Camacho (2022) by demonstrating that the Response Surface Methodology analysis is reproducible and robust across different statistical software packages (1). The re-computation of the experimental data using STAT-EASE confirmed that the core outcomes are consistent with the original analysis performed in Minitab 17, leading to the same scientific conclusions (1).

The key findings, including the identical optimal synthesis conditions (pH 10, 55°C, 180 min, 1.5 mL extract, 20 mL $AgNO_3$) and the validation of the models high significance and fit ($R^2 = 0.9854$), were successfully replicated. However, this comparative analysis also highlighted that while the final interpretations align, each software package has distinct characteristics. Minor differences were observed in the presentation of graphical outputs and in the final decimal places of some calculated values, such as the predicted optimal response (3.45 in STAT-EASE vs. the verified 3.40) (1).

These slight numerical variations are considered negligible and can be confidently attributed to the different proprietary algorithms and internal rounding-off protocols inherent to each software. Ultimately, this study concludes that despite these minor computational and presentational differences, the results are fundamentally the same. STAT-EASE is proven to be an equally effective and reliable tool for performing and validating this type of optimization analysis, reinforcing the integrity and reproducibility of the original research (1).

7 Acknowledgement

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8 References

(1) (2) (3) (4) (5) (6) (7) (8)

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