Surface Plasmon Resonance of Gold Nano-Sea-Urchins Controlled by Machine-Learning-Based Regulation in Seed-Mediated Growth

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Gold sea-urchin-like nanoparticles (GSNPs) are promising candidates for cancer thermotherapy. Due to the complex growth of GSNPs and the need for a precise prediction of their surface plasmon wavelength, genetic-algorithm-based artificial neural networks (GANNs) are used to determine the relationship between synthesis parameters and the surface plasmon wavelength of GSNPs grown via seed-mediated growth assisted by machine learning. Herein, a low-data test is trained by varying the ratio and concentration of gold seeds, sodium citrate, hydroquinone, and HAuCl₄. Then, a big data confirmation is conducted through massive parameter collection from over 684 samples. The well-trained GANN can guide parameter selection for seed-mediated growth to obtain the desired surface plasmon wavelength. An optimal model can be obtained after big data evolution to assist the growth method screening of the seed-mediated growth of sea-urchin-like gold nanoparticles (SGNPs) to achieve a stronger electromagnetic field of the surface plasmons. Machine learning has an advantage over empirical methods for seed-mediated growth and surface plasmon wavelength prediction, which increases research efficiency and decreases cost. The performance of the grown SGNPs is substantially improved in the visible domain.

1. Introduction

Gold (Au) nanoparticles (NPs) have been extensively studied because of their unique properties in applications related to optics, catalysis, surface-enhanced Raman spectroscopy (SERS), and photonics.¹⁻¹⁰ In Au NP synthesis, the NP size and shape can be controlled to tune the plasmon position.¹⁻¹¹ Au nanomaterials have various shapes and structures, including nanoﬂowers, nano-urchins, nanorods, nanocubes, and nanoplates,¹²⁻¹⁴ when grown via seed-mediated growth. The variation of NP shapes and sizes accommodates the modes and field strength of surface plasmon resonance (SPR) in the visible and near-infrared (IR) range, and thus these NPs have potential applications in plasmonics, catalysis, sensing, and medicine.¹⁵ Sea-urchin-like gold nanoparticles (SGNPs) can be synthesized by Au-seed-mediated growth on the surface of Au NPs; this process is sensitive to the growth solution.¹⁶ The formed morphologies of SGNPs can determine the surface plasmon wavelength.¹⁷ SGNPs are commonly synthesized using a seed-mediated method, such as the polyl method, sonochemical method, microwave hydrothermal method, and surfactant-directed seed-mediated method.¹⁶,¹⁸

The plasmonic heating response serves as a signature of the nanostructure internalization in cells. SGNPs are promising candidates for cancer thermotherapy. The shape effect of SGNPs strongly affects the plasmonic heating response. However, the shape control of SGNPs is difﬁcult in practice. In seed-mediated method, Au seeds are added into a solution consisting of precursors and various reductants to generate seed growth.¹²⁻¹⁹ The reducibility of the reductants is the dominant factor for SGNPs. The reduction of Au¹⁰ to Au⁰ by reductants directly causes an unsuccessful secondary growth in seed-mediated synthesis.²⁰,²¹ Thus, utilizing a moderate reductant, such as ascorbic acid, hydroxylamine, or sodium citrate,²²,²³ is necessary to control the reduction steps during seed-mediated growth. SGNPs have applications in biochemistry and engineering.²⁴ A mechanism for the secondary growth of spines on SGNPs has been proposed; however, the features of the SGNPs vary greatly due to the complex reaction environment. The tunability of the plasmon position using the features of SGNPs is difﬁcult.²⁴ The concentration and volume of the solution strongly affect the structure and thus electromagnetic ﬁeld of surface plasmons.

Materials informatics has been applied to the development of new materials. Artificial neural networks make scientific
research more efficient in terms of image identification, data prediction, and data classification. Genetic-algorithm-based neural networks (GANNs) are suitable for constrained and unconstrained optimization. The deduction process in a genetic algorithm (GA) is derived from biological evolution and natural selection; it repeatedly modifies a population of individual solutions. At each evolution step, the GA selects individuals at random from the current population to be parents and applies them to produce the children for the next generation. Through successive generations, the population generates an optimal solution. GAs can solve a variety of optimization problems that are not suited for standard optimization algorithms. However, GAs have not been applied to solve the fabrication of SGNPs to induce different morphologies.

Here, we fabricate SGNPs with different morphologies by tuning parameters. The optical and morphological properties of the SGNPs are characterized using UV–vis spectroscopy, field-emission scanning electron microscopy (FESEM), and transmission electron microscopy (TEM) to examine the plasmon position. The characteristics of the fabricated SGNPs are analyzed in line with information gained from the peak position and full width at half maximum (FWHM) of the surface plasmon wavelength obtained based on an analysis of the profile of the peak to determine NP shape. The experimental parameters and measurement results are input into a GANN for training to build an accurate, predictable, and controllable model. Then, the optimization of machine learning (ML) is conducted based on the tunable parameters in hidden layers, able to execute linear division, neuron number, representing an activate function, mating ratio, indicating the difficulty to conjugate with another data, and learning cycle in deep learning of GANN. These parameters can guarantee the accuracy of the model, making inferential data more dependable. Assisted by empirical results, we established a model based on the artificial neural network to predict material properties that may guide NP selection to obtain an anticipated material function.

2. Results and Discussion

2.1. Decision Flow

The design and arrangement of the experimental process and parameters influence the accuracy of the trained model providing prediction and optimization and also effect on data distribution. As shown in Figure 1, the development of SGNP synthesis was studied in advance and the function of each reagent was evaluated. The designed framework-like parameters in GANN workflow are helpful for exploring unknown experimental fields and predicting data in interpolation increase the precision. ML is used for calculation, including the operation and setting of the rules of the GANN, which can be accomplished in four steps. The first step is data evaluation, which ranks the data and unconstrained optimization. The deduction process in a genetic algorithm (GA) is derived from biological evolution and natural selection; it repeatedly modifies a population of individual solutions. At each evolution step, the GA selects individuals at random from the current population to be parents and applies them to produce the children for the next generation. Through successive generations, the population generates an optimal solution. GAs can solve a variety of optimization problems that are not suited for standard optimization algorithms. However, GAs have not been applied to solve the fabrication of SGNPs to induce different morphologies.

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2.2. Low-Data Test

Figure 2a shows a 3D scatter plot of the data distribution, where the X-axis is the amount of gold colloid, the Y-axis is the amount of hydroquinone aqueous solution, and the Z-axis is the plasmon position. The information in Figure 2a was collected from more than 100 samples, as shown in Figure S4, Supporting Information. The concentration of chloroauric acid for all samples was fixed at $5 \times 10^{-4}$ M. After stirring for 30 min at room temperature, the reductants, sodium citrate, and hydroquinone were used to grow SGNPs using a seed-mediated method. The liquid color changed from transparent to red, pink, blue, and purple (Figure S4, Supporting Information). After the reaction, the plasmon position was measured. More than 100 data points were collected and input to the ML to develop the model for the prediction of SGNP characteristics. In Figure 2, two algorithms were used for the calculation, namely, a back-propagation neural network (Figure 2b) and a GANN (Figure 2c). The X-axis is the prediction value calculated by the trained model and the Y-axis is the observation value from the experiment. The distribution of dots and the distance from a data point to the regression line show that the error of the trained model is related to prediction accuracy. To get better ML performance, the lower error ($<0.01$) evaluated by RMSE is taken into consideration. For the GANN (RMSE = 0.025), the data distribution is closer to the regression line and the RMSE is lower compared with those for the back-propagation neural network (RMSE = 0.041). Thus, based on the experiment data and prediction model, the GANN was applied.

2.3. Prediction Section

ML is used to establish a suitable model, which can calculate the relationship between all parameters and the measurement results to make accurate predictions. The prediction data calculated by the GANN are shown in Figure 2d,e and Figure S5, Supporting Information. The Z-axis is the position of the activated plasmon. When the size of SGNPs increases, the activated plasmon redshifts. Because each position has a unique color, the color distribution in the activated plasmon position in Figure 2d,e and Figure S5, Supporting Information, indicates plasmon shifts. To understand the influence of chloroauric acid on the SGNPs, the concentration of chloroauric acid was varied from $10^{-4}$ to $5 \times 10^{-4}$ M, as shown in Figure 2d,e and Figure S5, Supporting Information. The Z-axis is also the reciprocal of FWHM, which indicates the morphology and shape of the measured peaks (Figure 2b,c). An increase in 1/FWHM means that morphology and shape of the peak are more condensed or sharper, referring to the resonant quality.

It was found that when the amount of gold colloid (hydroquinone) was increased, the plasmon position...
blueshifted (redshifted). When the chloroauric acid concentration was increased to $5 \times 10^{-4}$ M, the plasmon position blueshifted for given amounts of gold colloid and hydroquinone. The blue and green areas are larger at higher concentration of gold colloid and hydroquinone, which indicates that the average diameter of Au NPs is smaller at lower chloroauric acid concentration, as shown in Figure S5, Supporting Information. The reciprocal of FWHM increased, when the amount of gold colloid raised. In contrast, when the amount of hydroquinone was increased, $1/\text{FWHM}$ decreased at a rate that depended on the amount of gold colloid. For a small amount of gold colloid, hydroquinone only slightly influences $1/\text{FWHM}$.

2.4. Big Data Confirmation

As shown in Figure 2f, the low-data test was used to build the model. RMSE was used to evaluate the match between the prediction data and observation data. To check the performance of the trained model, data of over 600 experimental samples were utilized to confirm whether the experimental results obtained for $10^{-4}$ M chloroauric acid were suitable for the trained model. Figure 2f shows the accuracy for excess over 600 samples experimental data input into previous trained model to check the trained model. The results reveal that the trained model for $10^{-4}$ M chloroauric acid is also suitable for $5 \times 10^{-4}$ M chloroauric acid.
Figure 2. a) 3D scatter plot showing the distribution of samples for various amounts of gold colloid and hydroquinone and the measured plasmon position. The concentration of chloroauric acid was $5 \times 10^{-4}$ M and that of sodium citrate was 0.2 wt%. Leave-one-out cross-validation results for b) back-propagation neural network (RMSE = 0.041) and c) GANN (RMSE = 0.025) predicting the synthesis of SGNPs. The $Y = X$ line represents ideal prediction. d) Plasmon position and e) reciprocal of FWHM of SGNPs calculated by GANN. The concentration of chloroauric acid was $10^{-4}$ M and that of sodium citrate was 0.2 wt%. f) Leave-one-out cross-validation results for GANN models predicting the synthesis of SGNPs. The $Y = X$ line represents ideal prediction. g) Plasmon position and h) reciprocal of FWHM of SGNPs in experiments. The concentration of chloroauric acid was $10^{-4}$ M and that of sodium citrate was 0.2 wt%.
A comparison of the prediction data (Figure 2b,c) obtained using the GANN with experimental data (Figure 2f) indicates that the experimental tendency from low amount to high amount is consistent with the results of predictions because experimental data are close to prediction data. This tendency can be observed from the color change. The tendency in color change is consistent with the predictions of the well-trained model.

The experimental results including more than 684 parameters are shown in Figure 2g,h. The X, Y, and Z axes are the amount of gold colloid, amount of hydroquinone, and plasmon position/reciprocal of FWHM, respectively. Here, the concentration of HAuCl₄ was fixed at 10⁻⁴ or 5 x 10⁻⁴ M. The amounts of gold colloid and hydroquinone were varied. In Figure 2g,h, the maximum plasmon position was obtained when the amount of gold colloid was close to zero and the amount of hydroquinone was around 250–300 μL. When the amount of added hydroquinone was over 250 μL, the plasmon position decreased. The influence of hydroquinone was small. The green area, which represents the SGNP plasmon position, is around 630 nm. A comparison of Figure S5, Supporting Information, and Figure 2g,h indicates that the reciprocal of FWHM for 5 x 10⁻⁴ M HAuCl₄ is higher than that for 10⁻⁴ M HAuCl₄, which means that the intensity of absorption increases and the shape becomes sharp for given amounts of gold colloid and hydroquinone. The variation of the reciprocal of FWHM can explain the morphology and intensity. The NP's have similar sizes. The precursor amount and Au seed density affect the distribution of NP size.

The TEM morphology of samples with various plasmon positions was analyzed (Figure 3). The color diagram can be divided into seven color areas, as shown in Figure 3a–g. As shown in Figure 3h, the plasmon position is related to morphology (i.e., particle size and shape). There is a high-intensity electric field around the thorns or branches of SGNPs, especially at thorn tips. The intensity of plasmonic enhancement is influenced by the material, number of thorns, and core diameter. The plasmon position at around 547 nm (navy blue) is contributed by irregularly shaped SGNPs, as shown in Figure 3a. The plasmon position at around 573 nm (light blue) color is contributed by smooth SGNPs, as shown in Figure 3b. The light green

![Figure 3](image-url)

Figure 3. The comparison of morphology-plasmon positions of SGNPs through a–g) empirical analysis and h) prediction. The seven regions represent different activated plasmon positions and the TEM images show the corresponding profiles of SGNPs. The concentration of chloroauric acid was 10⁻⁴ M and that of sodium citrate was 0.2 wt%.
Table 1. Prediction results of SGNPs for various concentrations of chloroauric acid (HAuCl₄). The concentration of chloroauric acid was varied from $10^{-4}$ to $5 \times 10^{-4}$ M and that of sodium citrate was 0.2 wt%. The addition of gold colloid is varied from 0 to 300 μL. The addition of hydroquinone is varied from 250 to 3500 μL. The $R_2/R_1$ ratio and number of thorns are shown, respectively. $R_1$ and $R_2$ are the diameters of SGNPs without and with long thorns, respectively.

| HAuCl₄   | $R_2/R_1$ | Thorns amount |
|----------|-----------|---------------|
| $1 \times 10^{-4}$ M | ![Image](177x549 to 312x654) | ![Image](408x552 to 544x654) |
| $2 \times 10^{-4}$ M | ![Image](177x439 to 312x543) | ![Image](408x443 to 544x443) |
| $3 \times 10^{-4}$ M | ![Image](177x329 to 312x433) | ![Image](408x334 to 544x334) |
| $4 \times 10^{-4}$ M | ![Image](177x219 to 312x323) | ![Image](408x223 to 544x223) |
| $5 \times 10^{-4}$ M | ![Image](177x108 to 312x213) | ![Image](408x113 to 544x113) |
color represents sharp SGNPs, which grow to around 80 nm in size. A large number of sharp thorns appear on the particle surface. The green color represents rounded SGNPs with sharp thorns on the surface. The diameter of these SGNPs is around 95 nm. The plasmon position is at around 635 nm.

The TEM observations (Figure 3) can partially explain the profile and formation mechanism of SGNPs. The relationship, containing the reaction space, reaction concentration, and reaction kinetics influenced by the synthesis process, has been generalized and concluded. The influence factors especially were trained by the GANN prediction system.

As shown in Figure 3a–g, the supply of Au\textsuperscript{0} reduced by hydroquinone in the synthesis reaction affects SGNP generation and is determined by the morphology of urchin thorns. Gold colloid, as the core of mature SGNPs or premature Au NPs, allows ionic Au in the solution to accumulate, stack, and grow sharp thorns on the surface. Added sodium citrate as a reductant has two major functions, namely, it reduces Au\textsuperscript{III} to Au\textsuperscript{I} at room temperature and it acts as a surfactant that forms micelles and sustains the morphology of SGNPs. Sodium citrate can reduce Au\textsuperscript{III} to Au\textsuperscript{I} and Au\textsuperscript{I} to Au\textsuperscript{0} in high temperature. The experiment on the concentration and amount of sodium citrate (Figure S1, Supporting Information) indicated that they slightly shift absorption.

![Graphs showing absorption intensity and FWHM](image-url)

Figure 4. The comparison of morphologically influenced absorption intensity (a–g) and the predicted reciprocal FWHM (h). a–g) Morphology of absorption peak under various conditions. h) The intensity increases with the increasing reciprocal of FWHM. The peak shape influences the FWHM. The concentration of chloroauric acid was 10^{-4} M and that of sodium citrate was 0.2 wt%.
Excessive citrate does not benefit the generation of SGNPs. The varying amount of sodium citrate was thus not taken into consideration. The concentration of HAuCl₄ affects the formation of SGNPs, including the growing kinetics and molecular movement in different reaction parameters. The increasing of HAuCl₄ indicates that the supplement of Au⁺ is raised, which provides the opportunity to form the branch growth or another atomic stacking.

As shown in Figure 4a–g, the concentration of HAuCl₄ affects the performance of SGNPs in terms of absorption intensity, activated plasmon position, particle size, particle profile, and reciprocal of FWHM (Figure 4h). The smooth absorption peak is related to the nonuniformity of thorns on the SGNPs, including thorn length, sharpness, and symmetry. The nonuniformity of thorns influences the interaction of the electromagnetic fields and the field fluctuation activates the resonance of electron sea. The aforementioned responses cause plasmon enhancement.

The smooth absorption peak is also related to the particle size distribution, which means that SGNPs of different sizes contribute to different activated plasmon positions, creating a smooth extinction peak. The uniformity of thorns and the particle size distribution for the off-white color are better than those for the off-gray color in Figure 4h. The profile and morphology of the SGNPs vary (in terms of thorns and particle size) with the amount of added reagents, as determined by a comparison of Figure 3 and 2b,c. Based on the prediction results containing the variation of chloroauric acid from 10⁻⁴ to 5 × 10⁻⁴ M, the ratio of R₁ and R₂ and the thorn amount of SGNPs are shown in Table 1. R₁ and R₂ are the diameters of SGNPs without and with long thorns, respectively. When the R₂/R₁ ratio is large, the thorns of SGNPs become sharp. The maximum R₂/R₁ ratio was obtained when the amount of added gold colloid was less than 100 μL and the amount of added hydroquinone was more than 3000 μL. The number of thorns strongly depends on the amount of added gold colloid. When the amount of gold colloid was above 200 μL and the amount of hydroquinone was insufficient, the number of thorns was close to zero and the SGNPs were spherical. The prediction by GANN from high concentration to low concentration of chloroauric acid is convenient and intuitive to analyze the tendency and variation when the synthesized condition is altering.

SGNPs are sensitive to growth conditions, which influence the structure and morphology and thus change the electromagnetic field of SPR. To discover the tendency of the gold NP, low data is built and put into calculation to build the prediction model. After the prediction model was optimized by a feedback mechanism, the big data for chloroauric acid of low concentrations was built and put into calculation to build the prediction model. After the designed GA, we could easily select an efficient experimental process.

4. Experimental Section

Materials: Chloroauric acid (HAuCl₄), sodium citrate (Na₃C₆H₅O₇·2H₂O), and hydroquinone (C₆H₄(OH)₂) were purchased from Sigma-Aldrich. The chemical reagents were of analytical grade. Deionized water (DI) was used throughout the experiment.

Preparation of Gold Colloid: Chloroauric acid (HAuCl₄) (1 g) was added into 100 mL of DI water to form a clear solution under ultrasonic oscillation for 5 min. The aforementioned solution (2.059 mL) was added into 97.94 mL of DI water. The concentration of the solution was 0.5 mM. Sodium citrate (0.228 g) was added into 20 mL of DI water under ultrasonic oscillation for 5 min to obtain 38.3 mM sodium citrate. Then, 50 mL of 0.5 mM chloroauric acid (HAuCl₄) solution was mixed with 5 mL of 38.8 mM sodium citrate. The mixed solution was heated to 100 °C and stirred magnetically at 600 rpm for 1 h. The color of the mixed solution changed from light yellow to wine red.

Control Experiments: In the SGNP synthesis process, sodium citrate and hydroquinone were used as reductants in different steps. The former reduced Au⁺ into Au⁻ and the latter reduced Au⁻ to Au²⁺. Gold colloid was added to serve as a nucleation center.(19) The rate-determining step was the reduction from Au⁺ to Au²⁺. To investigate the effect of each reagent, the concentration and volume of the reagents were varied. The amount of hydroquinone was varied from 150 to 3500 μL and that of gold colloid was varied from 0 to 300 μL. The concentration of sodium citrate was varied from 0.1 to 0.2 wt%. This concentration is more insensitive to the rate-determining step than the addition of hydroquinone. The concentration of chloroauric acid was varied from 10⁻⁴ to 5 × 10⁻⁴ M.

ML Approach: In materials science, predictions can be used to clarify the properties, characteristics, and functions of materials. Advanced artificial neural networks can be used as prediction models. Back-propagation neural networks are a popular type of artificial neural network. Back-propagation neural networks do not use evolutionary algorithms such as GA and the fitness function. Therefore, we applied a GANN to model and tune the multiple variables in the experimental process.

The ML approach used for the GANN framework in this study is shown in Figure 1a. There are two hidden layers and six neurons in the network. Data separation can be determined by the type of collected data. The hidden layer can be considered to divide data into several parts. In two dimensions, hidden layers are represented as lines or dots, and in three or more dimensions, they are represented as planes or curved planes. The number of layers depends on the data. The input data consist of experimental parameters, including gold colloid amount, hydroquinone amount, and sodium citrate concentration. After matrix calculation and validation with training data and testing data, the output layer generates prediction results.

3. Conclusion

GANN prediction can guide the selection of experimental methods for fabricating SGNPs. The amounts of added sodium citrate, hydroquinone, gold colloid, and chloroauric acid were varied to synthesize SGNPs with different shapes to confirm the designed ML model. We analyzed the morphology of the synthesized NPs using TEM and measured the optical characteristics using UV–vis spectroscopy. The SPR, usually generated on the surfaces of Au NPs, was studied. The results reveal that the plasmon wavelength depends on NP size and shape as well as number of thorns. The hidden layers in ML separate data into different regions. We found that four factors influence the precision of the training model: 1) the convergence of testing data and training data; 2) neuron number in the hidden layer (neuron number increases the complexity of the training process, and here the amount of neurons is considered into the independent variables); 3) the moderate mating ratio, helpful for establishing well-training model to reach the solution optimization; and 4) learning cycle length. Moreover, a desired prediction can be achieved by reducing the RMSE. With the designed GA, we could easily select an efficient experimental process.
Characterization: The surface morphology of the samples was measured using FESEM (Hitachi SU8200). TEM images were also taken (JEOL 3010, The Netherlands, Philips). X-ray powder diffraction (XRD) patterns were obtained using a D8 Advance diffractometer with Cu Kα radiation (wavelength = 0.111 nm). The absorption spectra of samples were measured using a Lambda 950 UV–vis spectrophotometer (PerkinElmer, New York, USA).

Supporting Information
Supporting Information is available from the Wiley Online Library or from the author.

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Conflict of Interest
The authors declare no conflict of interest.

Author Contributions
C.-C.W. and F.P. contributed equally to this study. C.-C.W. synthesized and analyzed all samples. C.-C.W., F.P., and Y.H.S. set up the ML parameters and ran the models. C.-C.W., F.P., and Y.H.S. wrote the manuscript.

Data Availability Statement
The data that support the findings of this study are available at Bionic Heterojunction Energy Transfer Material Digital Database [http://140.116.33.118/).

Keywords
- Genetic-algorithm-based artificial neural networks, gold nano-sea-urchins, machine-learning-based regulation, seed-mediated growth, surface plasmon resonance

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