Influence of Physiochemical Factors on Size of Gold Nanoparticles Synthesised Using Leaf Extract of Syzygium Cumini

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***Abstract-Syzygium cumini* is well known for its medicinal properties. This plant has shown potential for gold nanoparticles (GNP) synthesis. In the present study, we have varied physiochemical factors (metal ion, leaf extract ratio and incubation temperature) to see the influence on GNP size. GNPs of 13 to 70 nm size were synthesized by varying these parameters. The NPs thus synthesized through greener route and capped by medicinally important constituents can be very useful for applications in which products come in direct human contact.**

Keywords- S. Cumini; GNP; Physiochemical Factors; Greener Route

I. INTRODUCTION

Nanotechnology deals with synthesis of materials/particles having at least one dimension less than 100 nm. It also includes material or devices designed with the help of nanomaterials. GNP applications vary from medicine, drug delivery, catalysis, sensing to water purification [1-7]. Synthesis of GNP with new interesting properties and new arrangement and functionalization of nanomaterials with new diverse molecules has huge applications waiting. It is well known now that size, shape and molecules capping nanoparticles affect their properties. Functionalization/surface covering of GNP along with size and shape plays crucial role in toxicity determination [8]. Plants have been used for GNP synthesis [7, 9-13]. Incubation temperature, metal ion concentration and LE ratio have earlier been used for varying size of GNP. It has been reported that size and stability of metallic nanoparticles depend upon plant used, extract preparation method and reaction conditions [10-14]. Still there is need of biological method which can give GNP of different size with new surface covering with good stability. *S. cumini* is medicinally important plant and its LE has been reported for silver nanoparticles (SNP) and GNP synthesis by our group [15, 16]. Some of medicinally important components from plants have been reported to be involved in SNP and GNP capping. Thus, capping has enhanced the quality of GNPs synthesized [11, 12, 15,16]. In present study, we have synthesized various sized GNPs using *S. cumini* LE. Synthesis of 24 nm GNP by *S. cumini* LE has been reported by us earlier [16]. Here, we have used *S cumini* LE to synthesize various sized GNP by varying physiochemical factors like temperature, metal ion concentration and LE ratio.

II. MATERIALS AND METHODS

A. Materials

S. cumini leaves were collected from the fields of CSIR-IHBT, Palampur, India. $HAuCl_4$ was purchased from Sigma-Aldrich USA. *S. cumini* LE was prepared as described in our previous study [15].

B. Effect of Physiochemical Factors on Gold Nanoparticles Synthesiss

One mM metal ion ($HAuCl_4$), 10% LE ratio and room temperature were considered as standard conditions for GNP synthesis [11, 16]. To see the effect of metal ion concentration on GNP synthesis, 10% (v/v) of LE was incubated with 1, 3, 5, 7, and 9 mM aqueous $HAuCl_4$ solution at RT for 4 h. The effect of temperature on GNP synthesis reaction was studied by incubating 10% (v/v) of LE with 1mM $HAuCl_4$ solution at 40, 60, and 80°C. Effect of LE ratio on GNP synthesis was carried out by incubating 1 mM $HAuCl_4$ with 5, 10, 20 and 40% LE ratio at RT. The absorption spectra for each reaction mixture was recorded by taking 2 µl of solution with UV-visible scan on Nanodrop ND1000 spectrophotometer between 300 to 700 nm. The absorbance near 550 nm in all cases was confirmation of GNP synthesis. The reaction mixtures were centrifuged at 10000 rpm for 10 min to isolate the GNP from unreacted and unbound LE components. The NPs were washed thrice with double distilled water. GNPs were characterized for their morphology using Veeco diNanoscope 3D atomic force microscope (AFM). Water suspended GNPs were spread on the cleaned glass cover slip mounted on AFM stub. Samples were dried completely before characterization. Tapping mode was used for imaging and three images of each sample were obtained to assure reproducible results.

III. RESULTS AND DISSCUSSION

A. Effect of Metal Ion Concentration on GNP Synthesis

$HAuCl_4$ concentration was varied from 1, 3, 5, 7 to 9 mM. At 0 h of incubation, there was no absorbance at 550 nm and color of reaction mixture was light pale yellow. As the incubation time increased absorbance at 550 nm appeared and color of reaction mixture changed to ruby red due to synthesis of GNPs (7, 17). With increase in metal ion concentration from 1 mM to 3 mM, there was increase in peak intensity at 550 nm. This indicates quantitative increase

in GNPs synthesis as the peak at 550 nm is characteristic of GNP [7, 18]. Quantitative increase in GNP synthesis with increase in metal ion concentration has been reported earlier. This is due to availability of more and more substrate ions to reducing and stabilizing agents present in the reaction mixture [6, 11, 12]. Peak intensity decreased at 5 mM metal ion and higher (7 and 9 mM) metal ion did not lead to GNP synthesis (Fig. 1a). It is known that, if the concentration of metal ion is more or less than a critical level, no synthesis occurs [19]. Like other metallic nanoparticles, the ratio of metal ion in a synthesis reaction was found to be very crucial for stable GNP synthesis [20, 21]. Within this critical limit GNP size can be varied by varying metal ion concentration [7, 11, 12]. Use of 3 mM metal ion led to uniformly distributed GNPs, while aggregation was induced at 5 mM (Fig. 1b, 1c). Thus 3 mM is optimum metal ion concentration for synthesis of stable GNPs in good quantity. GNP synthesized at 3 and 5 mM metal ion concentrations were 43±3 and 70±6 nm in size, respectively (Fig. 1d). Similar increase in size of NPs with metal ions concentration has been reported earlier [11, 12, 19].

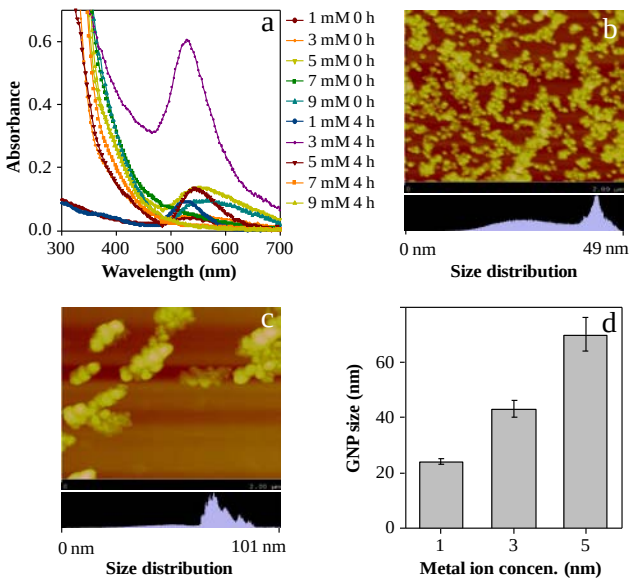

Fig. 1 Effect of metal ion concentration on GNP synthesis by *S. cumini* LE

(a) UV-visible absorption spectra of GNPs between 300 to 700 nm; (b), AFM images of GNPs synthesized at: (b), 3 mM and, (c), 5 mM metal ion concentration; (d), bar diagram shows size of GNPs at various metal ion

B. *Effect of Incubation Temperature on GNP Synthesis*

UV-visible absorption spectroscopy result showed increase in incubation temperature leaded to increase in intensity of GNPs characteristic peak. Increase in peak intensity was many folds as compared to GNP synthesized at RT. However there was less difference in absorption intensity of GNPs synthesized at 40-80°C. So, there was quantitative increase in GNP with incubation temperature (Fig. 2a). Quantitative increase in GNP synthesis with increase in metal ion concentration has also been reported earlier with other systems [10-12, 18]. But incubation temperature has been found to effect size of GNP synthesized with *S. cumini* LE. Size of GNPs was increased from 24±1 nm at RT to 32±3 at 40°C (Fig. 2b, 2e). There was almost no difference in size of GNPs with increase in temperature from 40 to 60°C (Fig. 2b, c, e). However, GNP size increased to 53±4 nm at 80°C (Fig. 2b-e). Such increase in GNP size has also been documented with increase in incubation temperature [12]. Importantly, synthesized GNPs were uniformly distributed in spite of quantitative increase, except little bit of aggregation at 80°C (Fig. 2b-d). This is due to the fact that increase in gold metal particles formed as a result of enhanced reduction of metal ion at higher incubation temperature is balanced by availability of appropriate amount of stabilizer provided by LE in reaction mixture, leading to more quantity of stable GNP [12, 13].

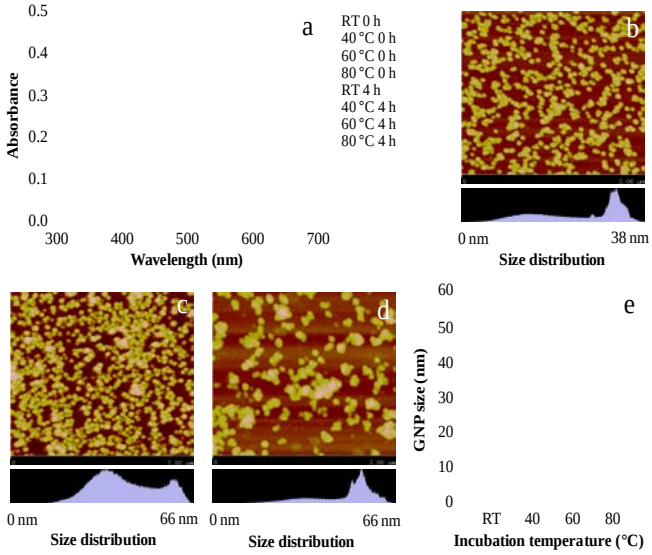

Fig. 2 Effect of incubation temperature on GNP synthesis by *S. cumini* LE

(a) UV-visible absorption spectra of GNPs between 300 to 700 nm. AFM images of GNPs synthesized at different incubation temperatures: (b), 40°C (c), 60°C; (d),80 °C; and (e), bar diagram shows size of GNPs at different incubation temperature

C. *Effect of Leaf Extract Ratio on GNP Synthesis*

Increase in LE ratio also had a positive effect on GNP synthesis. As LE ratio was increased from 5% to 40%, there was increase in peak intensity at 550 nm. Such increase thus indicated quantitative increase in GNP synthesis [19]. There was less difference in peak intensity of GNP at 5% and 10% LE ratio. However, 20% and 40% LE led to many folds increase in peak intensity (Fig. 3a). Increase in peak intensity with increase in LE could be due to availability of more reducing and stabilizing agent, thus increasing quantity of synthesized GNP. Higher LE ratio has been reported previously for quantitatively more GNPs [10, 12, 19]. All LE ratios from 5-40% led to stable GNP synthesis. However, size of GNPs was decreased with increase in LE ratio (Fig. 3b-e). 5% LE led to synthesis of 38±7 nm GNPs. Size of GNP decreased to 24±1 nm at 10% LE. Further increase in LE ratio to 20% and 40% led to synthesis of 22±1 and 13±2 nm GNP, respectively (Fig. 3e). The appropriate ratio of LE in reaction mixture was found to be required for stable GNP

synthesis [22, 23]. Since LE contains both reducing and capping agent, its ratio is very important factor for controlled sized GNPs synthesis [24]. Further the ratio of reducing to capping agents within a particular LE depends upon plant used and method of extract prepared. Good amount of capping agents at higher LE ratio may be responsible for decrease in size of GNP with increase in LE ratio [10, 12, 18, 19].

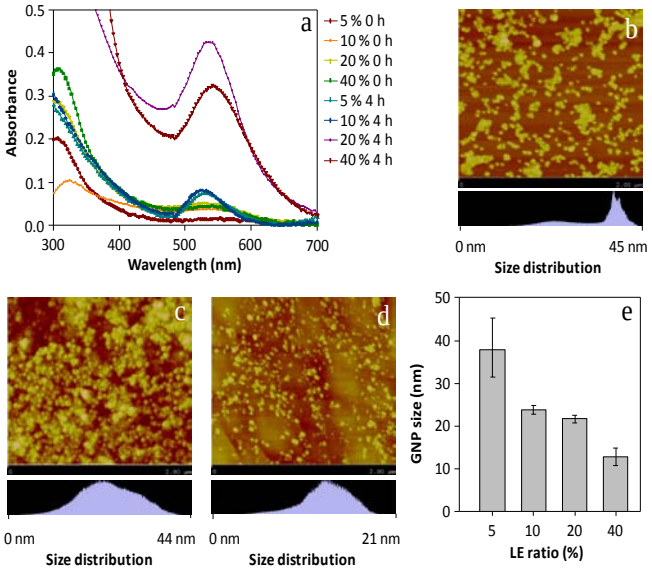

Fig. 3 Effect of LE ratio on GNP synthesis by *S. cumini* LE

(a) UV-visible absorption spectra of GNPs between 300 to 700 nm. AFM images of GNPs synthesized at different LE ratio: (b), 5 %; (c), 20 %; (d), 40 %; and (e), bar diagram shows size of GNPs at various LE ratio

IV. CONCLUSION

In plant leaf extract (LE)-mediated synthesis, constituents present in LE act as reducing and capping agent. Involvement of medicinally important component adds quality to the synthesized NPs. *S. cumini* having medicinally important constituents has been explored for SNPs and GNPs synthesis in our previous studies. Here effort was made to synthesize various sizes GNP and to quantitatively increase GNP synthesis. Incubation temperature and LE ratio showed positive effect on GNP synthesis. Metal ion concentration up to 3 mM led to quantitative increase in GNP, but concentration above this led to aggregation. So, for quantitative increase in GNP synthesis 3 mM metal ion, higher LE ratio and incubation temperature were recommended. GNP synthesized by this greener approach can find applications in drug delivery, biosensor, agriculture and biomedical fields.

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