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Title: Green Synthesis of Gold and Silver Nanoparticles Using Plant Extracts, Fungal Filtrates, and Bacterial Cultures

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Author Questionnaire

1. We have marked your project as author-provided footage, meaning you film the video yourself and provide JoVE with the footage to edit. JoVE will not send the videographer. Please confirm that this is correct.

☒ Correct

☐ Incorrect

2. Microscopy: Does your protocol require the use of a dissecting or stereomicroscope for performing a complex dissection, microinjection technique, or something similar? **NO**

3. Software: Does the part of your protocol being filmed include step-by-step descriptions of software usage? **Yes, all done!**

4. Proposed filming date: To help JoVE process and publish your video in a timely manner, please indicate the proposed date that your group will film here: **06/22/2026**

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Current Protocol Length

Number of Steps: 17

Number of Shots: 39

Interviews

Authors: Please note that **the questions will not appear on screen**. Please answer in **stand-alone full sentence with sufficient context**.

Note: The statements have been slightly edited for clarity.

INTRODUCTION:

~~REQUIRED: Summarize your research focus and the main questions it aims to answer.~~

- 1.1. **Suanny Mosquera-Romero:** Our research involves the development of reproducible methods for synthesizing metallic nanoparticles using plants, algae, fungi, and bacteria.

- 1.1.1. INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera.

~~Describe the research gap or limitation in existing methods and how this protocol addresses them.~~

- 1.2. **Suanny Mosquera-Romero:** Most nanoparticle synthesis protocols focus on single organisms; our framework enables reproducible comparisons across biological systems.

- 1.2.1. INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera.

CONCLUSION:

~~What does this protocol allow researchers to study or measure?~~

- 1.3. **Joel Vielma-Puente:** This protocol enables reproducible green synthesis and UV–Visible spectroscopy and Transmission Electron Microscopy validation of silver and gold nanoparticles.

- 1.3.1. INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera.

~~Describe the most important consideration or challenge when performing this protocol.~~

- 1.4. **Joel Vielma-Puente:** Reproducible nanoparticle synthesis depends on precise regulation of biological activity and physicochemical parameters such as pH and temperature.

- 1.4.1. INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera.

~~How can future studies build on this work? Describe the questions or research directions that can be investigated next.~~

- 1.5. **Suanny Mosquera-Romero:** Future studies could expand the scalability of these biogenic platforms for diverse environmental, antimicrobial, catalytic, and biomedical applications.
 - 1.5.1. INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera.

Protocol

2. Phytogetic Synthesis of Silver Nanoparticles (AgNPs) Using *Psidium guayaquilensis* Leaves

Demonstrator: María Cecilia Arteaga

- 2.1. To begin, wash the fresh plant material thoroughly with distilled water [1]. Dry the leaves in a tray dryer with forced airflow at 40 degrees Celsius [2]. Use a hand mill to grind the dried leaf material to obtain a fine powder [3].
 - 2.1.1. Talent washing fresh plant material with distilled water.
 - 2.1.2. Talent placing the leaves in a tray dryer for drying.
 - 2.1.3. Talent grinding the dried leaves using a hand mill.
- 2.2. Suspend 10 grams of powdered material in 200 milliliters of distilled water [1]. Heat the suspension at 70 degrees Celsius under continuous stirring for 1 hour [2].
 - 2.2.1. Talent adding 10 grams of powdered material to 200 milliliters of distilled water.
 - 2.2.2. Talent placing the suspension on the heating and stirring equipment.
- 2.3. Allow the extract to cool to a temperature of approximately 28 degrees Celsius [1]. Filter the extract under vacuum through qualitative filter paper with Grade 1 specifications [2-TXT]. Store the filtered extract at 4 degrees Celsius in amber bottles until further use [3].
 - 2.3.1. Talent setting aside the extract to cool.
 - 2.3.2. Talent filtering the extract under vacuum through qualitative filter paper. **TXT: Filter paper: Pore size 11 μ m, Diameter 110 mm**
 - 2.3.3. Talent storing the filtered extract in amber bottles.
- 2.4. For phytogetic fabrication of silver nanoparticles, or AgNPs, dissolve the appropriate mass of silver nitrate in distilled water to prepare a solution, with a concentration between 1 and 5 millimolar [1]. Mix the silver nitrate solution with the plant extract to obtain final concentrations between 10% and 50% volume per volume [2].
 - 2.4.1. Talent dissolving silver nitrate in distilled water to prepare the solution.
 - 2.4.2. Talent mixing the silver nitrate solution with the plant extract.
- 2.5. Heat and stir the reaction mixture at the selected temperature between 25 and 80 degrees Celsius for the desired reaction time between 0.5 and 1 hour [1]. Allow the reaction mixture to stand at room temperature for 24 hours [2].
 - 2.5.1. Talent placing the reaction mixture on the heating and stirring equipment.
 - 2.5.2. Talent setting aside the reaction mixture for standing.
- 2.6. Centrifuge the brown reaction mixture at 1660 g for 15 minutes at room temperature

[1]. Discard the supernatant [2] and collect the solid fraction [3]. Dry the recovered nanoparticles in an oven at 60 degrees Celsius [4].

2.6.1. Talent placing the brown reaction mixture in the centrifuge.

2.6.2. Talent discarding the supernatant.

2.6.3. Talent collecting the solid fraction.

2.6.4. Talent placing the recovered nanoparticles in the oven.

3. Mycogenic Synthesis of Silver Nanoparticles (AgNPs) Using *Earliella* sp

3.1. Transfer actively growing mycelial fragments of *Earliella* species onto Potato Dextrose Agar plates [1]. Incubate the plates at 28 degrees Celsius for 48 hours [2].

3.1.1. WIDE: Talent transferring mycelial fragments onto Potato Dextrose Agar plates.

3.1.2. Talent placing the plates in the incubator.

3.2. Transfer a mycelial fragment into Potato Dextrose Broth [1]. Incubate the culture under constant agitation at 150 revolutions per minute for 5 days [2].

3.2.1. Talent transferring a mycelial fragment into Potato Dextrose Broth.

3.2.2. Talent placing the culture on the agitation system.

3.3. Filter the fungal culture through sterile filter paper to separate the mycelial biomass from the extracellular broth [1]. Collect the filtrate for subsequent nanoparticle synthesis [2].

3.3.1. Talent filtering the fungal culture through sterile filter paper.

3.3.2. Talent collecting the filtrate.

3.4. For mycogenic fabrication of AgNPs, mix 2.5% to 10% volume per volume fungal broth with a 1 to 10 millimolar silver nitrate solution to a final volume of 50 milliliters [1]. Adjust the final reaction volume according to the experimental condition being tested [2].

3.4.1. Talent mixing the fungal extracellular broth with the silver nitrate solution.

3.4.2. Talent adjusting the final reaction volume to 500 mL.

3.5. Stir the reaction mixture at the selected temperature between 25 and 70 degrees Celsius for the desired reaction time between 1 and 4 hours [1]. Allow the solution to cool to room temperature for 24 hours [2].

3.5.1. Talent placing the reaction mixture on the stirring equipment.

3.5.2. Talent setting aside the solution to cool.

3.6. After centrifugation, collect the solid fraction [1] and dry in an oven at 60 degrees Celsius [2].

- 3.6.1. Talent collecting the solid fraction after centrifugation.
- 3.6.2. Talent placing the collected material in the oven.

4. Characterization of Phytogenic and Mycogenic Silver Nanoparticles by UV Spectroscopy

- 4.1. Turn on the ultraviolet-visible spectrophotometer [1]. Allow the instrument to warm up for at least 15 minutes to ensure signal stability [2].
 - 4.1.1. WIDE: Talent turning on the ultraviolet-visible spectrophotometer.
 - 4.1.2. SCREEN: 4.1.2.mp4. 00:00–00:02; 00:30–00:34; 01:03–01:04; 01:14–01:16
- 4.2. Select spectral scanning mode [1]. Configure the wavelength range from 300 to 800 nanometers using a data interval of 1 nanometer [2].
 - 4.2.1. SCREEN: 4.2.1.mp4. 00:02–00:04
 - 4.2.2. SCREEN: 4.2.2.mp4. 00:03–00:11
- 4.3. Collect aliquots of freshly synthesized silver nanoparticle suspensions [1]. Dilute the samples with distilled water as needed to maintain absorbance within the linear detection range [2].
 - 4.3.1. Talent collecting aliquots of freshly synthesized nanoparticle suspensions.
 - 4.3.2. Talent diluting the samples with distilled water.
- 4.4. Perform baseline correction using distilled water as the blank solution [1]. Transfer the prepared sample into a cuvette [2]. Place the cuvette in the sample holder [3]. Record the ultraviolet-visible spectrum over the 300 to 800 nanometer range [4].
 - 4.4.1. SCREEN: 4.4.1.mp4. 00:12–00:17; 01:17–01:21
 - 4.4.2. Talent transferring the prepared sample into a cuvette.
 - 4.4.3. Talent placing the cuvette in the sample holder.
 - 4.4.4. SCREEN: 4.4.4.mp4. 00:01–00:03; 00:06–00:09; 00:25–00:29; 00:58–01:02
- 4.5. Identify the surface plasmon resonance, or SPR (*S-P-R*) band corresponding to nanoparticle formation [1].
 - 4.5.1. SCREEN: 4.5.1.mp4. 00:04–00:06; 00:15–00:18

Results

5. Results

- 5.1. Representative ultraviolet-visible absorption spectra of AgNPs synthesized using *P. guayaquilensis* leaf extract showed a well-defined SPR band centered between 416 and 425 nanometers, indicating successful NP formation and stable colloidal dispersion [1]. Subsequent transmission electron microscopy confirmed the formation of predominantly quasi-spherical AgNPs with an average size of approximately 24 nanometers and a relatively narrow size distribution [2].
 - 5.1.1. LAB MEDIA: Figure 2A. *Video Editor: Please emphasize the value 425 in the image.*
 - 5.1.2. LAB MEDIA: Figure 3A. *Video Editor: Please emphasize the spherical shaped particles.*
- 5.2. Similarly, ultraviolet-visible absorption spectra of AgNPs synthesized using *Earliella* sp. extracellular fungal filtrate exhibited SPR maxima between 405 and 416 nanometers, confirming effective bioreduction of silver ions and AgNP formation [1].
 - 5.2.1. LAB MEDIA: Figure 2C. *Video Editor: Please emphasize the value 403 in the image.*

Pronunciation Guide:

1. Phytogenic
Pronunciation link: <https://www.merriam-webster.com/dictionary/phytogenic>
IPA: /ˌfætoʊˈdʒenɪk/
Phonetic Spelling: fy·toh·jen·ik
2. Synthesis
Pronunciation link: <https://www.merriam-webster.com/dictionary/synthesis>
IPA: /ˈsɪnθəsəs/
Phonetic Spelling: sin·thuh·suhs
3. Nanoparticles
Pronunciation link: <https://www.merriam-webster.com/dictionary/nanoparticle>
IPA: /ˌnænoʊˈpɑːrtɪkəl/
Phonetic Spelling: nan·oh·par·tuh·kuhl
4. Psidium
Pronunciation link: <https://www.howtopronounce.com/psidium>
IPA: /ˈsɪdiəm/
Phonetic Spelling: SID·ee·uhm
5. guayaquilensis
Pronunciation link: <https://www.howtopronounce.com/guayaquilensis>

IPA: /gwaɪ,jɑ:kɪ'lɛnsɪs/

Phonetic Spelling: gwy-yah-kee-LEN-sis

6. Distilled

Pronunciation link: <https://www.merriam-webster.com/dictionary/distilled>

IPA: /dɪ'stɪld/

Phonetic Spelling: dih-STILD

7. Celsius

Pronunciation link: <https://www.merriam-webster.com/dictionary/Celsius>

IPA: /'selsiəs/

Phonetic Spelling: SEL-see-uhs

8. Suspension

Pronunciation link: <https://www.merriam-webster.com/dictionary/suspension>

IPA: /sə'spenʃən/

Phonetic Spelling: suh-SPEN-shuhn

9. Qualitative

Pronunciation link: <https://www.merriam-webster.com/dictionary/qualitative>

IPA: /'kwɒlə'teɪtɪv/

Phonetic Spelling: KWAH-luh-tay-tiv

10. Specifications

Pronunciation link: <https://www.merriam-webster.com/dictionary/specification>

IPA: /,spesəfə'keɪʃən/

Phonetic Spelling: spes-uh-fuh-KAY-shuhn

11. Amber

Pronunciation link: <https://www.merriam-webster.com/dictionary/amber>

IPA: /'æmbər/

Phonetic Spelling: AM-ber

12. Fabrication

Pronunciation link: <https://www.merriam-webster.com/dictionary/fabrication>

IPA: /,fæbrɪ'keɪʃən/

Phonetic Spelling: fab-ruh-KAY-shuhn

13. Silver

Pronunciation link: <https://www.merriam-webster.com/dictionary/silver>

IPA: /'sɪlvər/

Phonetic Spelling: SIL-ver

14. Nitrate

Pronunciation link: <https://www.merriam-webster.com/dictionary/nitrate>

IPA: /'naɪ,treɪt/

Phonetic Spelling: NY-trayt

15. Millimolar

Pronunciation link: <https://www.merriam-webster.com/dictionary/millimolar>

IPA: /,mɪlə'moʊləər/

Phonetic Spelling: mil-uh-MOH-ler

16. Centrifuge
Pronunciation link: <https://www.merriam-webster.com/dictionary/centrifuge>
IPA: /ˈsentrəˌfjuːdʒ/
Phonetic Spelling: SEN·truh·fyooj
17. Supernatant
Pronunciation link: <https://www.merriam-webster.com/dictionary/supernatant>
IPA: /ˌsuːpərˈneɪtənt/
Phonetic Spelling: soo·per·NAY·tuht
18. Mycogenic
Pronunciation link: <https://www.howtopronounce.com/mycogenic>
IPA: /ˌmaɪkəˈdʒenɪk/
Phonetic Spelling: my·kuh·JEN·ik
19. Earliella
Pronunciation link: <https://www.howtopronounce.com/earliella>
IPA: /ˌɪərliˈɛlə/
Phonetic Spelling: eer·lee·EL·uh
20. Mycelial
Pronunciation link: <https://www.merriam-webster.com/dictionary/mycelial>
IPA: /maɪˈsiːliəl/
Phonetic Spelling: my·SEE·lee·uhl
21. Potato
Pronunciation link: <https://www.merriam-webster.com/dictionary/potato>
IPA: /pəˈteɪtəʊ/
Phonetic Spelling: puh·TAY·toh
22. Dextrose
Pronunciation link: <https://www.merriam-webster.com/dictionary/dextrose>
IPA: /ˈdekˌstroʊs/
Phonetic Spelling: DEK·strohs
23. Agar
Pronunciation link: <https://www.merriam-webster.com/dictionary/agar>
IPA: /ˈeɪɡər/
Phonetic Spelling: AY·gar
24. Incubate
Pronunciation link: <https://www.merriam-webster.com/dictionary/incubate>
IPA: /ˈɪŋkjəˌbeɪt/
Phonetic Spelling: IN·kyuh·bayt
25. Agitation
Pronunciation link: <https://www.merriam-webster.com/dictionary/agitation>
IPA: /ˌædʒəˈteɪʃən/
Phonetic Spelling: aj·uh·TAY·shuhn
26. Biomass
Pronunciation link: <https://www.merriam-webster.com/dictionary/biomass>

IPA: /'baɪoʊˌmæs/

Phonetic Spelling: BY·oh·mass

27. Extracellular

Pronunciation link: <https://www.merriam-webster.com/dictionary/extracellular>

IPA: /ˌɛkstrə'seljələr/

Phonetic Spelling: ek·struh·SEL·yuh·ler

28. Filtrate

Pronunciation link: <https://www.merriam-webster.com/dictionary/filtrate>

IPA: /'fɪlˌtreɪt/

Phonetic Spelling: FIL·trayt

29. Spectrophotometer

Pronunciation link: <https://www.merriam-webster.com/dictionary/spectrophotometer>

IPA: /ˌspektrəfoʊ'tɑːmətər/

Phonetic Spelling: SPEK·truh·foh·TAH·muh·ter

30. Ultraviolet-visible

Pronunciation link: <https://www.merriam-webster.com/dictionary/ultraviolet>

IPA: /ˌʌltrə'vaɪələt/

Phonetic Spelling: ul·truh·VY·uh·luht

31. Spectral

Pronunciation link: <https://www.merriam-webster.com/dictionary/spectral>

IPA: /'spektrəl/

Phonetic Spelling: SPEK·truhl

32. Wavelength

Pronunciation link: <https://www.merriam-webster.com/dictionary/wavelength>

IPA: /'weɪvˌleŋθ/

Phonetic Spelling: WAYV·length

33. Aliquots

Pronunciation link: <https://www.merriam-webster.com/dictionary/aliquot>

IPA: /'ælɪˌkwɒt/

Phonetic Spelling: AL·ih·kwaht

34. Absorbance

Pronunciation link: <https://www.merriam-webster.com/dictionary/absorbance>

IPA: /əb'zɔːrbəns/

Phonetic Spelling: ub·ZOR·buhns

35. Baseline

Pronunciation link: <https://www.merriam-webster.com/dictionary/baseline>

IPA: /'beɪsˌlaɪn/

Phonetic Spelling: BAYS·line

36. Cuvette

Pronunciation link: <https://www.merriam-webster.com/dictionary/cuvette>

IPA: /kjuː'vet/

Phonetic Spelling: kyoo·VET

37. Plasmon

Pronunciation link: <https://www.howtopronounce.com/plasmon>

IPA: /'plæzmɑ:n/

Phonetic Spelling: PLAZ·mon

38. Resonance

Pronunciation link: <https://www.merriam-webster.com/dictionary/resonance>

IPA: /'rezənəns/

Phonetic Spelling: REZ·uh·nuhns