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One Step Nano Selenium Green Synthesis via *Saccharomyces cerevisiae*

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Abstract

With the increasing necessity to synthesis nanoparticles in cheap and efficient methods and also sustain the environment, scientists keen on developing green based methods. Therefore, the present study demonstrated a cheap method to synthesize selenium nanoparticles (SeNP) based on bakery yeast and sodium hydrogen selenite only. The bakery yeast is cheap and abundant in any local market. So, the yeast can be found and applied in swift and easy manner during the experimental procedures that requires nano particle production. The resulted selenium nano-particles were verified by Means of multiple physical and chemical tests, namely Ultra-Violate Visible (UV-Visible) technique, Fourier transform infrared (FTIR) method, scanning electron microscopy (SEM) technique, and X-ray diffraction (XRD) device. The obtained scanning electron microscopy (SEM) results and Fourier transform infrared (FTIR) analysis respectively show 40-80 nm sized selenium nano-particles surrounded by preserving organic molecules. The external organic molecules help protecting and stabilizing the newly formed selenium nano-particles. The internal structure of SeNP was found to be amorphous as it was shown by the figures of the X-ray diffraction (XRD) test. In conclusion, the proposed method reduces the cost of manufacturing NP so that cheap experiments become achievable for researchers and costumers.

Keyword: Cost-effective methods, green synthesis, selenium nanoparticles

Introduction

The role of selenium (Se) in the living tissues was revealed after a century and a half (1) from its discovery as a toxic substance in the 19th century (2). After finding that selenium prevents lesions in animal tissues, intensive studies focusing on its impact on the human tissues and the mechanisms behind were conducted. For example, a link has been established between the levels of selenium and occurrence and development of diseases such as cancer. Furthermore, selenium, as a fundamental micronutrient, adjusts the biological and metabolic processes in the body so that any deficiency of selenium would generate serious disorders namely cancer, neurological disorder, muscular issue, and immune malfunctioning (1). When Selenium used in the synthesis of new derivatives of selenonitrone revealed increase in cell prolipheration (2).The emergence and remedies of the disorders in the human body not only depends on the levels of selenium but also on the size of the selenium particles (3). In general, crystalline or amorphous structures of organic (selenomethionine and selenocysteine) or inorganic (selenite and selenate) Se are essential in biology as well as physics and chemistry (4, 5), because they have cytoprotective and anticancer effects on the tissues (3). Green synthesis of nanoparticles is an eco-friendly by using microorganism (6). Selenium nano-particles SeNPs also preserve the same effects so that it is regarded as a nano-medicine with promising characteristics. Hence, SeNPs can function as reduce toxicity, antimicrobial, anticancer, antidiabetic, antiparasitic, antioxidant, and reduce the impact of reactive oxygen species (ROS) and free radicals (7-9). One of the best reducing candidate for nanoparticle synthesis is the saccharomyces cerevisiae (SC) or well-known as baker's yeast (7). Such choice is based on the saccharomyces cerevisiae's abundant production and well-defined genetic and physiological characteristics. The implementation of the yeast in bioprocesses was reported in the work of Göksungur, who mentioned the absorption role of SC to the metals such as cadmium and lead. Moreover, the SC can participate in the production of cadmium–bisglutathionate complex or vacuoles by anchoring cadmium to glutathione (GSH) (8). Therefore, instead of the traditional chemical techniques to produce nanoparticles, biological based methods are widely emerging (10). For instance, silver and gold nanoparticles have been intensively yielded based on these green methods. However, limits on fabricating selenium nanoparticles, such as the necessity of a large number of bacteria, are still challenging (9, 11, 12). Hence, the present investigation demonstrates an approach to synthesis selenium nanoparticle in a very simple and cheap method based on S. cerevisiae.

Materials and Methods

Materials and chemicals

The current work requires two main materials: yeast (*saccharomyces cerevisiae*), which can be found easily, and sodium selenite were purchased from the local market.

Synthesis of selenium nanoparticles

The first step of the experiment includes mixing 100 gm of dried yeast in 1 L of deionized distilled water at 45°C in conical flask. Secondly, 50 gm of sugar was added to the mixture, which was next lain on pre-adjusted magnetic stirrer with 500 rpm stirring power at 45°C. Then, the yeast solution was mixed with 0.1 M of sodium hydrogen selenite ($NaHSeO_3$) solution in 2:1 ratio. The resulted solution was kept in darkness by covering the

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flask with aluminum foil to avoid the impact of the light on SeNPs composition. Finally, the flask was stirred again on the magnetic stirrer at room temperature until the formation of the red brick color, which is the initial sign of the generation of SeNPs (13).

Characterization of SeNPs

The formation of SeNPs was checked by UV–Visible spectroscopy (Shimadzu, UV–1600, Japan) (14) with wavelength range 190.00 to 1100.00 nm. The next step focused on the purity of the surface of SeNPs. Hence, Fourier transform infrared (FTIR) spectroscopy (ABB, spectro-lab, MB3000, UK) was applied to determine the chemical nature of the associated functional groups and biological extracts attached to the SeNPs in the range 400–4000 cm^{-1} with the framework of classic KBr pellet technique that measures infrared intensity against the wavelength of light. After that, the inner structure of SeNPs was detected by X-ray diffraction (XRD) technique (Shemadzu, XRD-6000, Japan) with CuK_{α} radiation with wavelength equals to 1.5406 A°. Finally, the morphology and distribution of SeNPs are determined by scanning electron microscopy (SEM) (Tescan Vega lll, Czech).

Results

The first result of the experiment is the change of the color throughout the whole biosynthesis process. Fig. 1 illustrates different color codes corresponding to different solutions of the work. Tubes A, B, C, and D refer transparent sodium hydrogen selenite solution, beige solution of the yeast, light-red selenium nanoparticles mixed with yeast, and dark-red selenium nanoparticles solution, respectively.

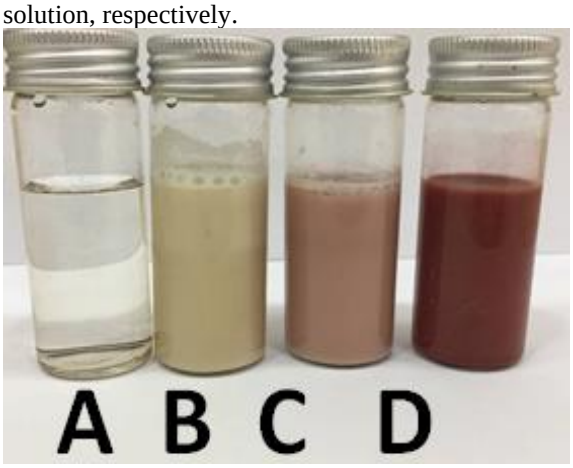

Figure 1: Color changes during biosynthesis of selenium nanoparticles (SeNPs) using yeast solution (B) mixed with sodium hydrogen selenite solution in 1:2 ratio. The color code is (A) sodium hydrogen selenite, (B) yeast, (C) SeNPs with yeast, and (D) SeNPs.

After color change, SeNPs, $NaHSeO_3$, and yeast solutions were tested by the UV-Visible spectroscopy to verify the presence of the nanoparticles. These finding were depicted in Fig.2. A prominent peak at approximately 300 nm wavelength has been obtained for the nanoparticle containing solution, SeNPs (Fig.2a). However, no-peak was noticed for the sodium selenite solution, Fig.2b, and low intensity peak was recorded for yeast, Fig. 2c.The Fourier transform infrared spectroscopy (FTIR) of the final SeNPs hydrosol was shown in Fig.3. The FTIR analysis was conducted to investigate the nature of the bonds so that one can recognize the biological compounds responsible for the synthesis, stability of the particles, and any purity of the synthesized particles. The major output of the present FTIR is the multiple absorption valleys at different wavelengths. Secondly, he absorption values aggregate in a band-like structure with high and low bands. The third point is that the absorption values are high with amount reaches approximately to 75%.

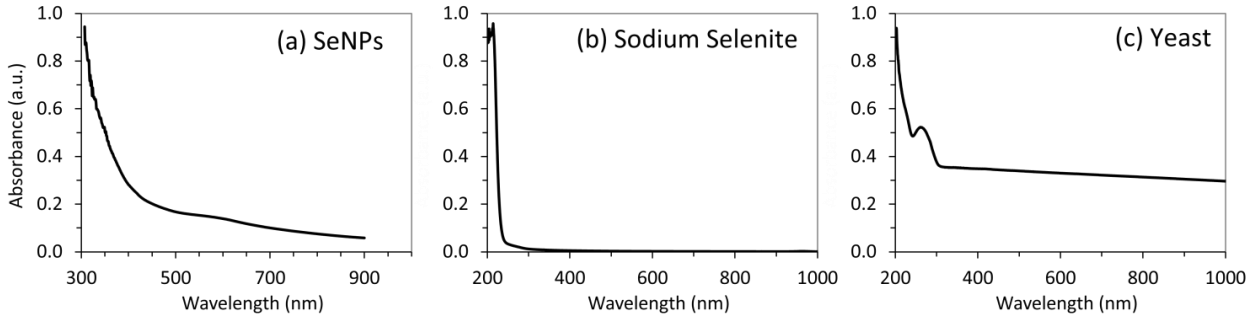

Figure 2: The UV-visible spectroscopy analysis of (a) SeNPs , (b) $NaHSeO_3$ and (c) yeast.

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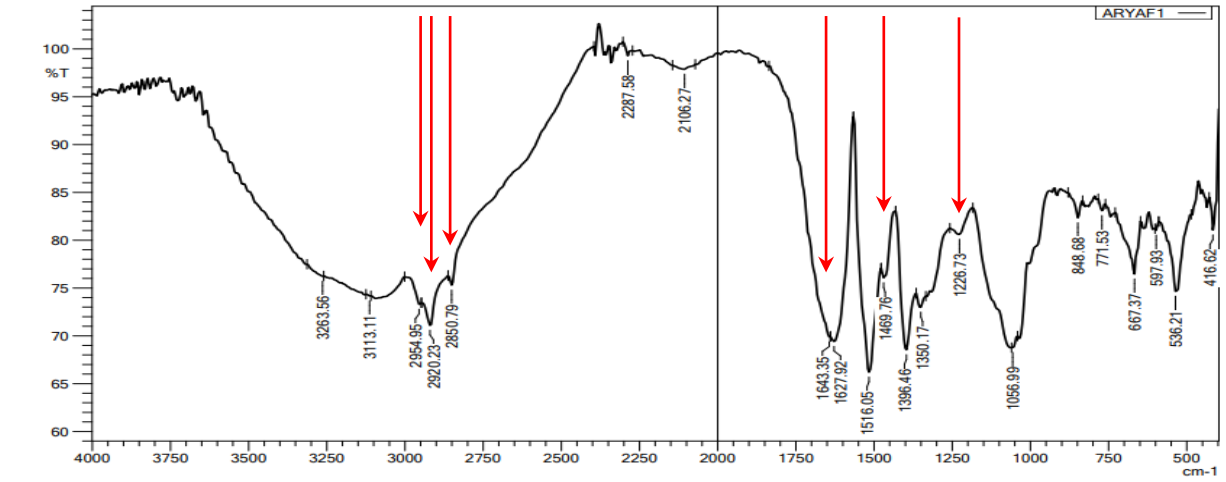

Figure 3: FTIR spectrum of functional groups attached to SeNPs

In addition, we have performed the scanning electron microscopy (SEM) technique to envisage the size of the particles, as shown in Fig. 4. This figure approves the existence of nanoparticles with size range from 40 nm and above. Further, the particles exhibit two distinguished areas: the dark inner region and the light outer sector.

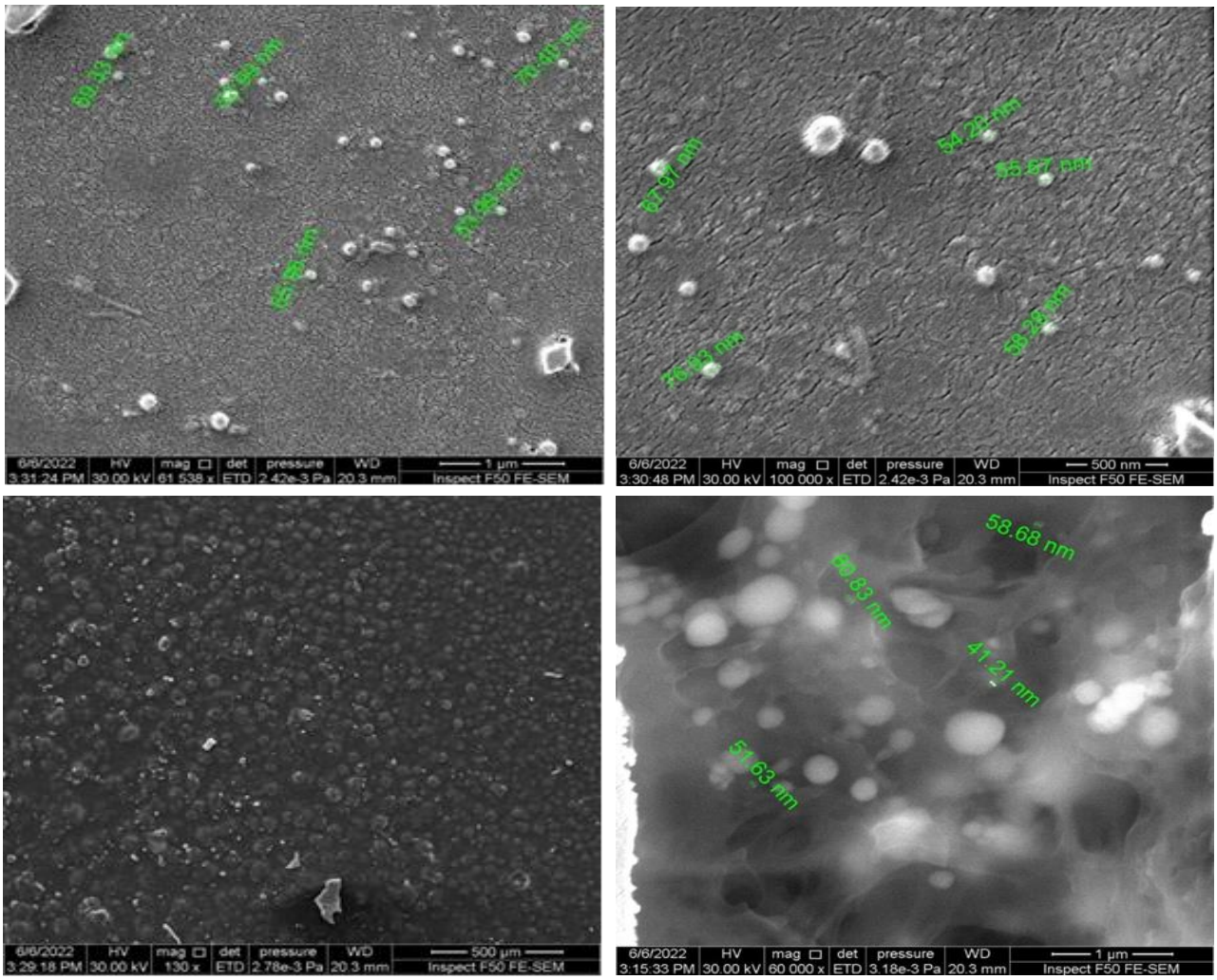

Figure 4; Morphology and size characterization of selenium nanoparticles.

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Figure 5 shows the XRD pattern of the nano-selenium powder synthesized by yeast. There is a specific diffraction peak in the range of 19–25° at the angle of 2θ, as shown in the dashed black rectangle in Fig. 5. The peak also illustrate a wide energy range and high absorption intensity.

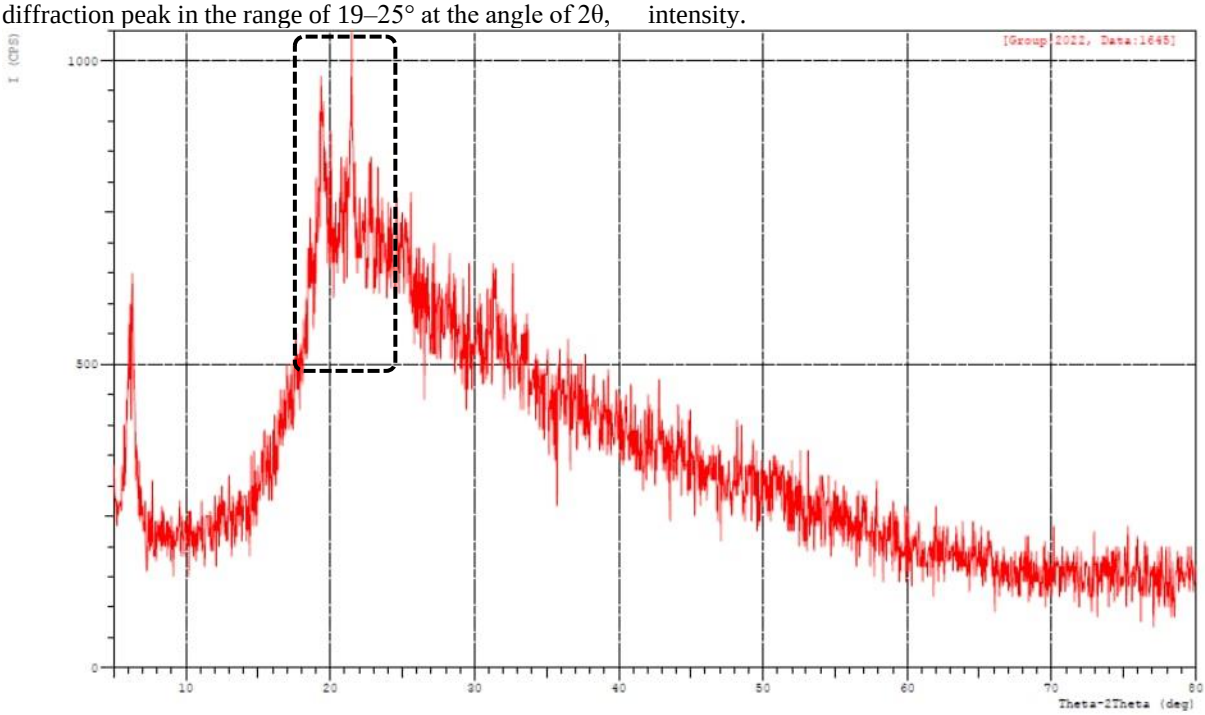

Figure 5: XRD pattern of nano-selenium synthesized from yeast.

Discussion

During the chemical interaction between sodium selenite (precursor) and yeast (reducing agent), the color of the solution gradually starts to dye red-brick. Such pigment represents the first characteristic feature of the formation of SeNPs (15), as shown in Fig.1D. In other words, the precursor is reduced to selenium atoms that, first, attach to each other to form small clusters, and secondly, resist dissolving back in the solution. Then, the newly formed and distributed Se clusters aggregate as nanoparticles. Simultaneously, as the size and number of NPs grows, they interact differently with light that excites the surface of the NPs leading to collective vibrations of the electrons, so-called surface plasmons, on the surface of the NP. Hence, the solution shows as red-brick color. The UV technique depends on the interaction between light, in the ultraviolet-visible region, and nanoparticles in a solution. Hence, the interaction of the incident light with NP dyes the solution with a special color (wavelength) due to the nature of the elements at the nano-scale that seriously relies on their type and size. Hence, NP solution depicts a special absorption spectrum with a distinguished peak for each element. In the current study, the absorption spectrum of SeNPs, as shown in Fig 2a, demonstrates a peak at 307 nm, which coincides with the plasmon energy of SeNPs as it was previously reported by Ullah et al. (5, 16, 17). The former and later teams mentioned different absorption bands, 200-300 nm and 266.5-353 nm respectively, depending on the reducing material. But both teams confirmed the emergence of SeNPs around 300 nm wavelength. Furthermore, the UV spectrum was also obtained for the sodium selenite, Fig 2b, and yeast, Fig 2c. The last two UV spectra were performed to benchmark the work and clarify the interference between the red of the yeast with SeNPs solution and the red of the SeNPs alone solution. The FTIR data revealed the appearance of several IR adsorption valleys related to different bonding vibrations. These valleys confirm the formation of bonds that refer to the existence of biological molecules alongside the SeNPs. Such finding was also approved by the images of the SEM, where each particle illustrates two regions (dark inner core surrounded by light external corona), Fig. 4. The IR spectral fingerprint of the solution includes the bands at ~1643 cm^{-1} probably due to amid I (18), ~2960 cm^{-1} and ~2920 cm^{-1} for antisymmetric stretching of the CH_3 and CH_2 in the protein, respectively (19), while 2850 cm^{-1} is for symmetric CH_2. The bands at ~1470cm^{-1} represents the deformation in C-H bonding, ~1226 cm^{-1} is for C=O stretching (20). All these organic bonds prove the attachment of the organic functional groups to the SeNPs and they may result from the yeast of the absorbance of water to the yeast molecules. The SEM technique not only gives images of the nanostructure but also represents an essential tool to probe the morphology,

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density and size NPs in the hydrosol. By applying the SEM on our samples, Fig. 4, we found an accumulation of unequally distributed spherical nanoparticles but with different sizes ranging from 41 nm to 80 nm. Moreover, the produced SeNPs seem to be coated with an organic membrane, as mentioned previously, so that the structure would be composed of an inorganic core surrounded by organic shell, so-called core-shell NP. This nano-capsule offers tremendous applications especially in the bioapplications. For instance, they can be used in cosmetics, sensors, and ink industry. The wide diffraction peak indicates that the synthesized nano-selenium particles are small in size, poor in crystallinity, and amorphous. The newly formed micro nano-selenium is a simple substance selenium, its valence electron structure is $4s^2 4p^4$, 4d orbital is all empty, and nano-selenium particles have very high surface free energy, showing the surface effect of nanomaterials. They adsorb the biological macromolecules on their surface, blocking and hindering the growth of crystals, and hence, amorphous substances are produced. Fig. 5 shows the XRD spectrum of the SeNPs. It can be clearly seen that the synthesized particles have an amorphous structure. The broad peak appearing in the figure is for the background of the glass slide.

Conclusions

The main conclusion of our work is the production of selenium nanoparticle (SENPs) via very cheap green synthetic method that depends on bakery yeast and sodium selenite. The occurrence of SeNPs was approved by mean of UV-Visible, SEM, FTIR, and XRD techniques. These particles possess an amorphous internal structure with 60 nm average size, and they are shielded organic molecules. Such Se-core and organic-shell structure insures more stability of the particles by preventing them from direct interaction with external solutions. Our results are beneficiary in academic and technological level because SeNPs can be produced by less than 10 USD.

Author's declaration

The authors declare no conflict of interests. We hereby confirm that all figure and tables in the manuscript are ours.

Author's contribution

Majida A. J. Al-Qayim designed the study. Aryaf Mahmood Sabea and Ahmed Thamer Wali did the experiments. Aryaf Mahmood Sabea wrote the paper and analyzed the data.

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