Physical and biological synthesis of GNPs and keratin nanoparticles from chicken's feather and applications

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ABSTRACT: In this paper, we aim to synthesize keratin nanoparticles from chicken's feather. Crude keratin was extracted by chemical method. We obtained results for keratin nanoparticles using glutaraldehyde as a cross-linking agent. In this study, gold nanoparticles (GNps prepared by way Laser Ablation) with keratin nanoparticles for the production of antibacterial materials. NPs samples were characterized using a Transmission Electron Microscope (TEM). The absorption spectra were measure by UV-Visible, double beam spectrophotometer. Three types of pathogenic bacteria were used in this study (Pseudomonas aeruginosa, Staphylococcus aureus, and E. coli) in addition to one opportunistic yeast (Candida albicans). Nano-creatine particles can be used for wound dressing as well as cosmetic preparation.

Keywords: chicken's feather, Laser Ablation, Keratin nanoparticles, GNps

Introduction:

Keratin (kεrɔtɪn)[1][2]) is one of a family of fibrous structural proteins known as scleroproteins. α-Keratin is a type of keratin found in vertebrates. It is the key structural material making up hair, nails, feathers, horns, claws, hooves, calluses, and the outer layer of skin among vertebrates. The feathers are the major waste from poultry industry and are economically feasible material that can be used for various applications. Chicken feathers are composed of 91% protein,
8% moisture and 1% lipid in total combination [3]. In general, the synthesis of NPs is broadly divided into two main classes; Bottom-up approach and top-down approach [1]. Ablative-pulsed laser (APL) is an example of a top-down approach. It is an efficient physical technique for nanomaterial synthesis, particularly ablation of solids (including metals, semiconductors, ceramics, and alloys) in liquid environments (pure water or a water solution of a stabilizing agent). This method is much simpler than chemical methods, producing highly purified nanoparticles with weak agglomeration effects [4][5][6].

Recently, nanoparticles have attracted the interest of many researchers due to their high surface area, low diffusion resistance, and more active sites [7][8][9]. Considering the advantages of nanoparticles and keratin biosorbents, keratin nanoparticles were synthesized in this study [10][14].

METHODS:-

Preparation of GNPs nanoparticles
The colloidal solutions of the gold nanoparticles (GNPs) were prepared with double deionized water (DDDW), (pH=7.2), using pulsed laser ablation in the liquid phase (PLAL), with 1064 nm Nd-YAG laser, and energies (600) mJ and the laser spot intensity of laser (2.23) J.cm⁻². The number of applied pulses associated with these energies were 300 pulses. The repetition rate and the pulse duration were (10 ns) and (6Hz), respectively for the solutions.

Extraction of keratin:-

Feathers Chicken were obtained from home slaughter, washing was carried out using detergent solution Triton-X100, a chicken's feather were dissolved in 5% of sodium hydroxide solution kept in shaker (Remi) at 70 rpm for 4 h of incubation. This mixture was then filtered using Whitman No. 1 filter paper. This crude extract was deep-frozen and lyophilized in Lyophilized (Remi) at −40°C, from which the powder form of crude keratin protein was obtained and stored at 4°C[11][12].

Preparation of Keratin nanoparticles:-

Add 100 mg of the isolated keratin in 2ml of DDW, for 8ml absolute ethanol it under constant stirring. Next, 1µl of 8% glutaraldehyde was added to conjugate the protein molecules and the formation of nanoparticles. This mixture was stirred using magnetic stirrer at 40 rpm for 24 h. After that, stirred content was centrifuged by
cooling centrifuge at 20,000 rpm for 20 min. The supernatant was discarded, the pellet was collected, and lyophilization process was performed to obtain keratin nanoparticles.

**The Results and Discussion:**

**Characterization of gold nanoparticles:**

Fig.1 represent SPRs of GNPs in DDDW, The prepared GNP colloidal solution by 600 mJ with 100 pulses was neglect due to lack of concentration. SRPs were measured at room temperature, the peaks of absorption were be found at (526) nm for GNPs in DDDW. The size measurements were achieved by the calculation and the analysis of the transmission electron microscope (TEM). The prepared GNPs with DDDW were found between (10 and 11) nm diameters.

![Graph showing SPR spectra of GNPs](image1)

**Characterization of keratin nanoparticles:**

Fig.2 represent SPRs of keratin in DDW, The prepared KeNPs for 8ml absolute ethanol it under constant stirring. Next, 1µl of 8% glutaraldehyde [13]. SRPs were measured at room temperature, the peaks of absorption were be found at (271) nm for KeNPs in DDW. The size measurements were achieved by the calculation and the analysis of the transmission electron microscope (TEM).

![Graph showing SPR spectra of keratin nanoparticles](image2)
microscope (TEM). The prepared KeNPs with DDW were found between (10 and 9) nm diameters.

Fig 2: SPR spectra of KeNPs in DDW (right) and TEM of prepared KeNPs with DDW were found between (10 and 9) nm diameters (left).

**Antimicrobial activity of GNPs and KeNPs.**

Figure (3) and table (1) show the antibacterial activity of GNPs and KeNPs. These NPs showed no toxicity against *E. coli* and *S. aureus* but it showed significant toxicity toward *P. aeruginosa*. Smaller particles show more toxicity.

Fig. 3: Antibacterial activity of GNPs and keratin nanoparticles
Table (1) show the antibacterial activity of GNPs and KeNPs.

| NPs size/concentrations(ug/ml) | Microorganisms | P. aeruginosa | S. aureus | E. coli | C. albicans |
|-------------------------------|----------------|---------------|-----------|---------|-------------|
| Keratin NPs nm               |                |               |           |         |             |
| 10                            |                | 17            | -         | -       | 24          |
| 20                            |                | 18            | -         | -       | 38          |
| 50                            |                | 20            | -         | -       | 55          |
| GNPs nm                      |                |               |           |         |             |
| 10                            |                | -             | -         | -       | 14          |
| 20                            |                | -             | -         | -       | 32          |
| 50                            |                | 17            | -         | -       | 38          |

Table (1) and Figure (4) show the effect of KeNPs and GNPs on *candida albicans* growth. There were inhibitory effects seen for both KeNPs and PtNPs sizes at different concentrations.

![Figure (4): Effect of KeNPs and GNPs on *candida albicans* growth](image-url)
Discussion

High-purity gold and keratin nanoparticles with two particle sizes (10 and 9 nm) respectively, have been successfully synthesized in pure water by using the laser ablation method and method Lowry’s, respectively.

The antimicrobial assay showed high anti-pseudomonas activity of gold and keratin nanoparticles while it showed no effects on E. coli and S. aureus. KeNPs with a particle size of (9 nm) showed higher toxicity than GNPs with a particle size of (10 nm) at the same concentrations used.

Effect of KeNPs and GNPs on candida albicans growth of the antimicrobial assay showed high activity of gold and keratin nanoparticles as well as keratin nanoparticle showed higher toxicity than GNPs.

Conclusions

In this paper, keratin nanoparticles were synthesis to dressing wound as well as an antibacterial. Use of feathers may lead to decrease in waste management also will economically advance the industries. These GNPs and KeNPs can be used to treat complicated pseudomonas infections. Effect of KeNPs and GNPs on candida albicans growth of the antimicrobial assay showed high activity of gold and keratin nanoparticles.

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