

---

# Structural, Morphological and Biotoxicity Studies of Biosynthesized CaO Nanoparticles Via *Cuminum Cyminum*

Sumaira Pervez<sup>1</sup>, Syeda Mona Hassan<sup>1</sup>, Shahzad Sharif Mughal<sup>1, \*</sup>, Alejandro Pando<sup>2</sup>,  
Alvina Rafiq<sup>1</sup>, Nageena Shabbir<sup>1</sup>

<sup>1</sup>Department of Chemistry, Lahore Garrison University, Lahore, Pakistan

<sup>2</sup>Center for Biomedical Engineering, Brown University, Providence, USA

## Email address:

shezi1130@gmail.com (S. S. Mughal)

\*Corresponding author

## To cite this article:

Sumaira Pervez, Syeda Mona Hassan, Shahzad Sharif Mughal, Alejandro Pando, Alvina Rafiq, Nageena Shabbir. Structural, Morphological and Biotoxicity Studies of Biosynthesized CaO Nanoparticles Via *Cuminum Cyminum*. *Advances in Materials*.

Vol. 11, No. 2, 2022, pp. 37-49. doi: 10.11648/j.am.20221102.12

**Received:** June 13, 2021; **Accepted:** May 5, 2022; **Published:** May 12, 2022

---

**Abstract:** Nanotechnology is a branch of science and technology concerned with the design and manipulation of materials and devices with at least one longitudinal dimension of 1100 nanometers. Nanotechnology encompasses not only the study and development of nanoscale materials, but also the study and development of structures, devices, and material architectures that have novel properties and functions due to their nanoscale and dimension components. Plant extracts serve as capping agents and stabilisers in the manufacture of CaO nanoparticles, hence the green synthesis approach was chosen. The CaO nanoparticles were studied using SEM, XRD, FTIR, EDX. The existence of active phytoconstituents was validated by a phytochemical study of *Cuminumcyminum* extract. SEM proved the morphology and monodispersive clusters of CaONPS, while FTIR confirmed the presence of CaONP with defined peaks corresponding to the synthesis of CaO nanoparticles. Following SEM and FTIR, EDS was used to confirm the existence of CaO nanoparticles by displaying the paternity of Ca and O atoms in the form of peaks. The results of the XRD examination were even more conclusive. The current research is cost-effective and can be used to create antifungal reagents in the pharmaceutical and food industries.

**Keywords:** Nanochemistry, *Cuminum Cyminum*, CaO Nanoparticles, Morphology

---

## 1. Introduction

The study of nano materials and their application underpin enormous areas of industrial technology. Therefore, nanotechnology is considered to be, by its nature, a highly interdisciplinary field. In the broad area of laboratory/industrial usage in which chemists develop new nano scale materials, nano scientists and technologists strongly understand that nanotechnology focuses on functional materials. Physicists study their electronic and photonic properties and are intended to be used by engineers in suitable structures and systems and circuits. Nanotechnology deals with engineering and science from 1 to 100 nm at the nano level.

This technology also includes the biological, physical and material sciences. Nanotechnology researchers are seeking to

enhance nano-level materials that first transform into atomic particles and eventually into molecular particles. Because of their small size in comparison to the bulk, nano particles have entirely different properties. Because nanomaterials are distinct from bulk materials and due to their extremely small size, they have unique properties. The explanation for their widespread use in the medicine field is also because of their characteristics. This technology is becoming international because of the varied applications. For study and technology and for the economy of the state and the economy of the state, nano particles are more valuable [1].

Over the past three decades, in nearly every scientific field, including biomedical, chemical, computer, electrical/optical sensing, electronics, and mechanics, and their importance and that of mini atomization in general, nano science and nanotechnology have flourished and

developed. The developments in nano science are mainly focused on the ability to manufacture nano devices with different materials, such as spherical nanoparticles, nano cables, nano rods, nano prisms, and complex architectures of these nanostructures that are efficiently assembled.

Nanoparticle/structure synthesis is technically a well-understood phenomenon, but submicron or nanoscale dimensional materials have been exploited for way longer than anybody might have thought. The "Roman Lycurgus

Cup," a cup made of a unique kind of bronze glass called dichroic glass with coloured lines on the outer surface, is the first known example of silver and gold nanoparticles [2]. According to a study commissioned by the British Museum to exhibit the cup, which is currently on loan to the Chicago Art Institute until August, the extraordinary glass appears green at first but turns into a bright transparent red colour when light passes through it due to the inclusion of colloidal silver and gold nanoparticles on the cup's walls [3].

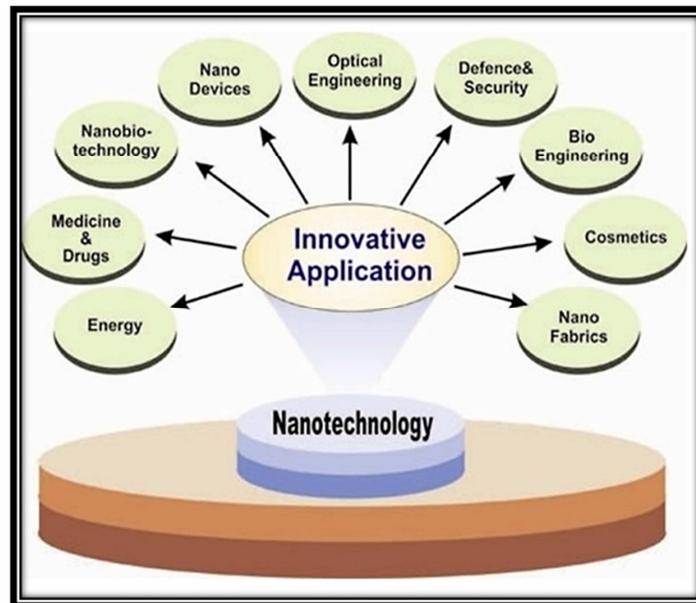


Figure 1. Uses of nanotechnology in different field.

A branch of science concerned with the efficiency of atoms or molecules on nano-scales is the field of nanotechnology and fundamental physical and chemical properties. The synthesis of nanoparticles of metal and metal oxides (1 to 100 nano meter). The materials made up of these components are smaller, lighter, heavier, stronger and have a broad variety of key nanotechnology chemistry applications. The main goal of pursuing a green synthetic approach to synthesis nanoparticles of a range of metal and metal oxide oxide is to eliminate toxic chemicals, reduce pollution, efficiency, low cost and comparatively high product yield [4]. Typical inorganic nanometal oxides include magnesium oxide (MgO), titanium oxide (TiO<sub>2</sub>), copper oxide (CuO), zinc oxide (ZnO) and calcium oxide (CaO). For other human species, these nano metal oxides are safe, stable, antimicrobial agents and have multifunctional properties. The basic antimicrobial properties of calcium oxide (CaO) nanoparticles (CaO NPs) and special structural and optical properties, and are healthy for all living organism. (Ramola et al., 2019). Due to its broad features, nanotechnology is used in most fields. The diagram provides a graphical illustration of nanotechnology.

### 1.1. Top-Down Approach

As the name implies, the smallest artare formed from the largest molecule) dimension, such as copper (Cu), a thick

material that can transform translucent (transparent), unsolvable like gold (Au), into soluble and stable material like aluminium (Al). The top-down approach is used in industry and has a fast turnaround time [5].

### 1.2. Bottom-up Approach

This technique refers to the make up of molecules from smaller bodies. There are many techniques in the advanced synthetic chemistry field to synthesize large molecules used to manufacture many drugs, polymers and Pharmaceutical Goods [6].

Nanoparticles are the tiniest objects on the planet. Furthermore, using plant extracts reduces the expense of microorganism isolation and culture media, making microorganism-produced nanoparticles more cost-competitive. The use of various plants and their extracts to synthesise nanoparticles may be useful in biological synthesis processes that need very complex microbial maintenance [7].

Natural nanomaterials, such as sugars, protein, fat, etc. Inorganic nanomaterials may be toxicology-mediated metals, such as calcium oxide NPs UV visible spectroscopy (UV vis) is the basis for the development of Fourier transform infrared spectroscopy for the purpose of characterization.(FTIR for CaNPs), Xray diffraction (XRD) analysis, Dynamic Light Scattering DLS) and Field scanning Emission [8].

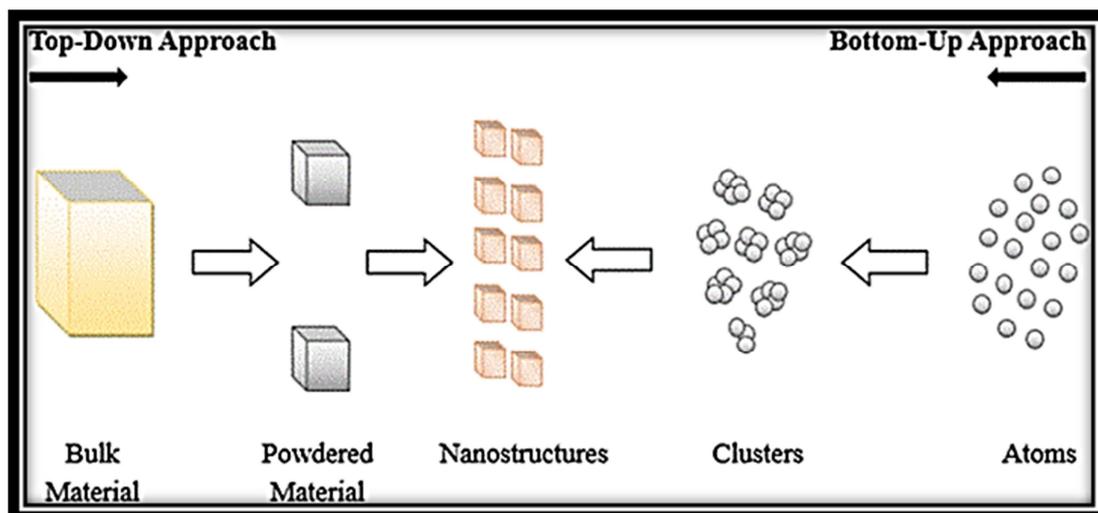


Figure 2. Diagrammatic representation of Top-down and Bottom-up approach.

Bottom-up			Top-down	
Chemical synthesis	Self-assembly	Positional assembly	Lithography	Cutting, etching, grinding
Particles Molecules	Crystals Films Tubes	Experimental atomic or molecular devices	Electronic devices Chip masks	Precision engineered surfaces
Cosmetics Fuel additives	Displays		Quantumwell lasers Computer chips MEMS	High quality optical mirrors

Figure 3. The use of bottom-up and top-down techniques in manufacturing.

Nanotechnology allows scientists and technologists to discover new materials and social gains, but it's still important to think about the negative health and environmental consequences of nanoscale devices. As a result, it's become more important than ever to figure out how modern materials cause negative effects and how to mitigate them. In identifying and discriminating against the risks of nanoscience, nanotechnologists still face obstacles [9].

[10] According to the term, nanotechnology is the field of atomic or molecular processing of substances as a result of controlled synthesis of materials with at least one dimension in the nanoscale size range, i.e. 1-100 nm. Nature has been using molecular manipulation to create its structures, such as plants and animals, since the dawn of life on Earth, so this fascinating science is not new to it. One will be inspired to create nanoscale materials after a thorough examination and understanding of nature's basic design principles. This exciting area of research is

concerned with the study of nanoscale devices and their compatibility with nature.

Traditional techniques have been used for many years, but research has shown that green techniques are more powerful for NP generation because they have a lower chance of failure, are less expensive, and are easier to characterise. Environment change pressures [11]. Plant-based synthesis of NPs is not a difficult procedure; a metal salt is synthesised with plant extract, and the reaction takes minutes to a few hours at room temperature. Over the last decade, this strategy has gotten a lot of interest, particularly for silver (Ag) and gold (Au) NPs, which are safer than silver (Ag) and gold (Au) (Au). It is simple to scale up the production of NPs using green methods, and they are also cost effective. Increased use of chemicals that are detrimental to human health and the atmosphere may increase particle reactivity and toxicity, as well as create unfavourable health effects due to the lack of assurance and variability of composition [12].

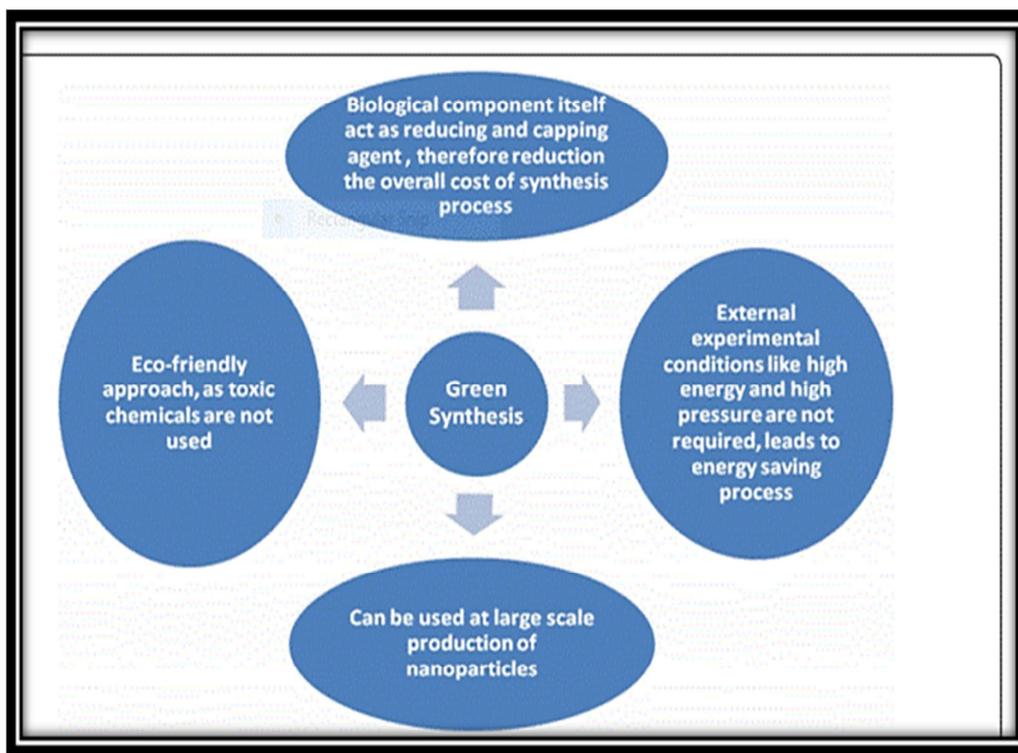


Figure 4. Uses of green synthesis.

The word 'Spices' derived from the word 'species' in Latin, which means explicit form. A closely related phrase, spice, is used to identify plant parts that find similar uses but come from green or delicate parts. For similar plants in which the new leaves are used as spices, the two words could be While other dried bits are used as flavours have different food capacities. They're often used to make food more robust, but they're also used to keep food nutritious and coordinate nutritional and medical benefits. Flavors are dictated by a person's level of growth [13].



Figure 5. *Cuminumcyminum* (Cumin seeds).

Spices are a major bio supplement for food fixings, as well as nutritional enhancements, which is more than that, nourishing changes. Spices have been used as food-added substances since ancient times to enhance the taste and to be a sort of food. Spices also have different medicinal applications in addition to these uses. As an ingredient, they

also go around, which provide healthy and medical benefits as well. Spices are non-green sections of plants (for example, bud, herbal product, seed, bark, rhizome, bulb), used as an improvement or preparation, but many can also be used as a natural medicine [6].

Spices aid us in our daily lives by soothing us, calming us, and energising us. Spices were extensively used by ancient cultures, including the Egyptians, Arabs, and Romans, not only as food and drink additives, but also as drugs, disinfectants, incenses, stimulants, and agents. They are used to store beef, food, and vegetables in Europe, the Middle East, and Asia, which is why they were found in the same manner that gold and precious metals were [14]. Spices help us in our daily lives by healing us, relaxing us, and energising us. Spices were commonly used by ancient cultures including the Egyptians, Arabs, and Romans not only to flavour food and drinks, but also as narcotics, disinfectants, incenses, stimulants, and aphrodisiac agents. They are used to store meat, wheat, and vegetables in Europe, the Middle East, and Asia. No wonder they were looked for in the same way as gold and precious metals [14].

Spices help us in daily life, heal us, calm us and excite us. Ancient cultures, such as the Egyptians, the Arabs and the Romans, used spices extensively, not only to give taste to food and drinks, but even as remedies, disinfectants, incenses, stimulants and sometimes as aphrodisiac agents. It has been used in Europe for the preservation of beef, bread and vegetables in the Middle East and Asia. It was used in the treatment of gentle stomach-related problem as a carminative and eupeptic, as an astringent in horse pneumonic problems and as a hack cure, just as a pain reliever.



Figure 6. Different variety of spices.

In everyday life, spices support us, cure us, relax us, and excite us. Spices were used commonly by ancient civilizations, such as the Egyptians, the Arabs and the Romans, not only as food and drink ingredients, but even as medicines, disinfectants, incenses, stimulants and sometimes as agents. They are used in Europe, the Middle East and Asia to store meat, bread and vegetables no doubt why they were discovered in the same way that they were searching for gold and precious metal.

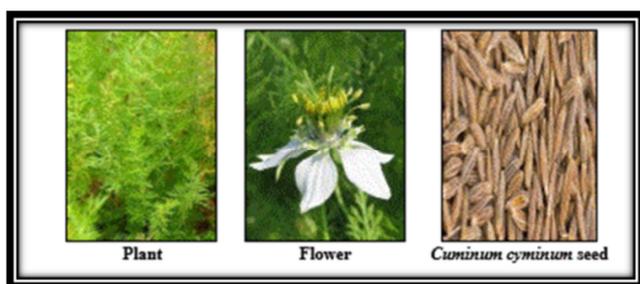


Figure 7. *Cuminumcyminum* (cumin seeds).

In everyday life, spices support us, cure us, relax us, and



Figure 8. Different varieties of cumin.

In traditional Iranian medicine, the cumin product has a specific oil made from terpenoids (such as  $\beta$ -pinene, alphapinene, cumin liquor,  $\beta$  phellandrene) and has been used as an improvement in vitality and resistance, stomach-related, diuretic, bacterial, convulsive and farthostile, and is used to reduce weight in the network [15] Cumin seeds are hand-picked from an annual plant and are thin, boat-shaped seeds

excite us. Spices were used widely by ancient civilizations, such as the Egyptians, and the Romans, not only to give taste to food and beverages, but even as medicines, disinfectants, incenses, stimulants and sometimes as aphrodisiac agents. In Europe, the Middle East and Asia, they are used for storing beef, bread and vegetables. No wonder they were looked for in the same way as gold and precious metals.

Spices help us in daily life, heal us, calm us and excite us. Ancient cultures, such as the Egyptians, the Arabs and the Romans, used spices extensively, not only to give taste to food and drinks, but even as remedies, disinfectants, incenses, stimulants and sometimes as aphrodisiac agents. It has been used in Europe for the preservation of beef, bread and vegetables in the Middle East and Asia. It was used in the treatment of gentle stomach-related problem as a carminative and eupeptic, as an astringent in horse pneumonic problems and as a hack cure, just as a pain reliever [3].

*Cuminumcyminum* is also prevalent as wild or shahijeera in the Persian and Indian subcontinents and is synonymous with natural cumin appearance. Its seeds include long, thin, bent dark brown cases with a particular natural flavor. From Linn's. The herbaceous plant (*Cuminumcyminum*), otherwise known as dark seed or dark cumin, is, The Mediterranean region is used in the Middle East, Central Europe, and Western Asia for food preparation (Kooti, Hasanzadeh-Noohi, Sharafi-Ahvazi, Asadi-Samani, & Ashtary-Larky, 2016). [14] For more than 2000 years. 18-20 other pharmacological features include: hypertensive, nociceptive, aggressive, uricosuric, choleric, anti-fertility, and anti-diabetic, antihistamine, oxidant-hostile, relaxing, anti-microbial, anti-tumor and immune-modulatory. The rich portion of its nigellone and thymol quinine is approximately 27 percent 57 percent and has various therapeutic qualities. Developed in Iran, it is commonly used.

that look like caraway seeds. Cumin is most often present in a brownish-yellow hue, but black cumin, green cumin, and white cumin are also available. While in some patients it induces diarrhoea or constipation, it can track and balance the motility of the GI if used in sufficient doses. A research conducted in Germany in 1996 found that herbal essential oil containing essential oil was found to be [16].



Figure 9. *Cuminum cyminum* extract.

## 2. Materials and Methods

### 2.1. Extract Preparation

Optimized concentration of spice *Cuminum cyminum* about 100g was dissolved in 1000ml distilled water and continue to heating on burner for 1 hour, until the 2/3<sup>rd</sup>

volume of extract was left behind from total amount added distilled water. Extract was cooled and filtered using filter paper (whatman No. 1). Now, extract is ready for use. The extract was freshly prepared for the synthesis of CaONPs.

### 2.2. Green Synthesis of CaONPs

For the preparation of CaONPs, 75g of  $\text{CaCl}_2$  was dissolved in 200ml distilled water and 150g of NaOH in 200ml distilled water in ratio of 1:2. The extract was mixed with  $\text{CaCl}_2$  solution in the beaker at 25°C and was put in funnel. On lower side, NaOH in a flask on hotplate, funnel on upper side and thermometer hang in it. The extract and  $\text{CaCl}_2$  solution fell drop by drop from funnel into NaOH solution which is on hotplate. Both the solutions of  $\text{CaCl}_2$  and NaOH were mixed with magnetic stirrer. The temperature which was recorded at this time between 60-80°C. At this point, the formation of nanoparticles was started. The second step of synthesis is washing. After centrifugation, the mixture was left for 24 hours. Next day, washing was done. During washing small quantity of ethanol was also added for the removal of some extra impurities. Last step is the filtration and drying. Dried NPs were obtained after 72 hours at room temperature, followed by calcinated at 450°C for three hours in muffle furnace and then subjected for the characterization of CaONPs [17].

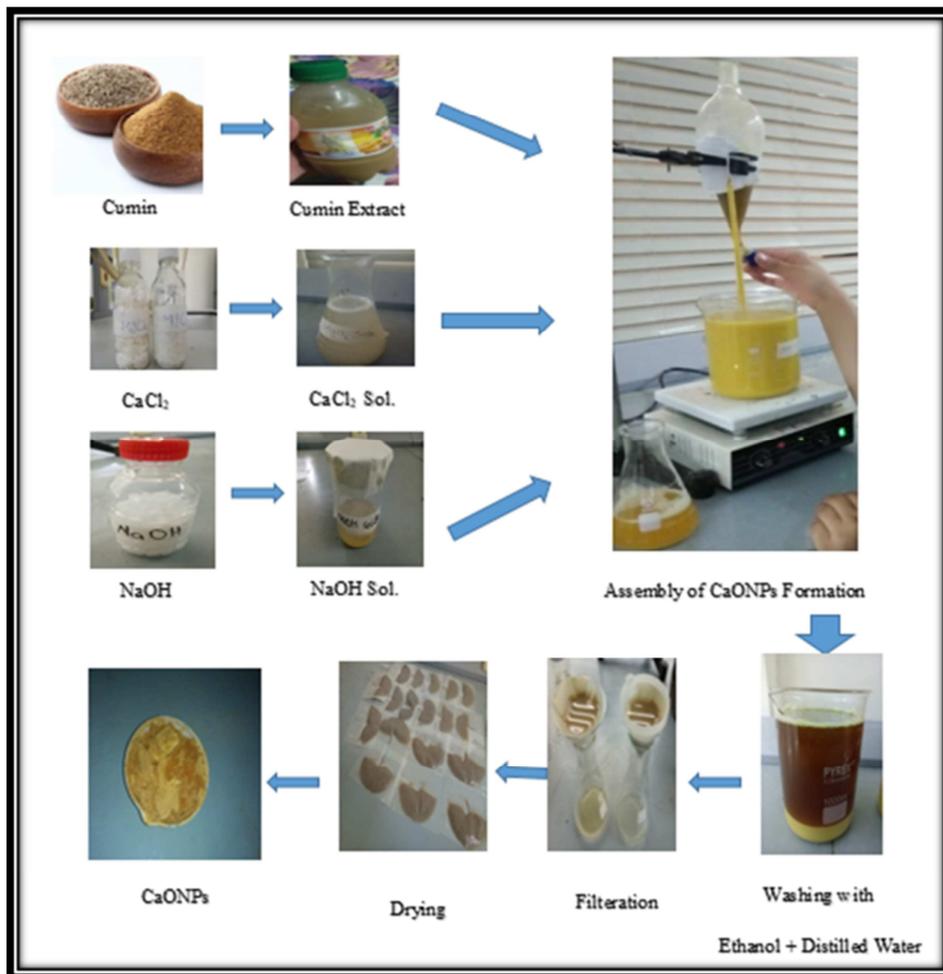


Figure 10. Diagramatic representation of CaONPs.

### 2.3. Characterization of CaONPs

The presence of nanoparticles in the solution mixture was verified using a UV-VIS spectrometer. Furthermore, characterization of the CaONPs were carried out. In detail, the purity of the phase was determined by X-ray diffraction (XRD) analysis using an XPert PRO diffractometer (Holland) with a detector operating under a voltage of 45.0 kV and a current of 20.0 mA with Cu-K $\alpha$  radiation. The recording range of  $2\theta$  was 20° to 80°. The average crystallite size from XRD was calculated using Scherrer equation.

Fourier-transform infrared (FTIR) spectra were recorded in the range of 500-4000 cm<sup>-1</sup>. The morphology was examined with a TM-1000 scanning electron microscope (SEM) (Hitachi, Japan) and a JEM-1230 Electron dispersive x-ray spectrometer (Japan).

### 2.4. Biological Potential of Nanoparticles

The extract of *Cuminumcyminum* against a panel of microorganisms which included five (*Aspergillusparasiticus*, *Aspergillusflavus*, *Fusariumoryzae*, *Fusariumtritichum*, *Aspergillusoryzae*) collected from the Fungal Bank, University of Punjab, Lahore. Fungal strains were grown in the PDA (Potato Dextrose agar) overnight at 28°C [18]. Slants of microbial strains were preserved at 4°C. The antimicrobial ability of plant extracts was determined by the use of disc diffusion assay.

### 2.5. Disc Diffusion Assay

The antifungal activity of CaO nanoparticles was determined by disc diffusion assay against *Aspergillus flavus*, *Aspergillus parasiticus*, *Fusarium oryzae*, *Fusarium tritichum* and *Aspergillus oryzae* by previously adopted method [19] with little amendments. PDA (Potato dextrose agar) solution prepared, autoclaved. In sterilized Petri plate under laminar air flow, about 20mL PDA (Potato dextrose agar) solution was poured. On the agar plates sterilized discs (6mm) of wicks sheet impregnated with nanoparticles. Fluconazol (30  $\mu$ g/disc) (Oxoid), Rifampicin (30  $\mu$ g/disc) (Oxoid) used as positive reference for fungal strains. As a negative control disc without samples were used. Standard disc and test disc were placed in separate Petri dishes. The plates were incubated at 37°C for 48h for fungal growth. By measuring the diameter of inhibition zones (mm) by zone reader, antifungal activity was evaluated.

### 2.6. Statistical Analysis

By performing the experiment in singlet (n=1), the data was described, mean  $\pm$  SD. Data analyzed at 5% significant level by using the mathematical package Minitab 2000 Version 13.2 (Minitab Inc. Pennsylvania, USA). As mean values at 95% confidence interval data of antifungal activity presented. Significant differences of mean were calculated by using LSD. Onstipation [20]. In the present study, three tests and their color reveals the prsence of carbohydrates in extract of *Cuminum cyminum*. The result of present study is in

agreement with the previous study of [21] who reported the prsence of Carbohydrates in *Cuminum cyminum*.

## 3. Results

### 3.1. UV-Vis Spectroscopy

The current results match those of a previous study [22], which discovered phenolic compounds in *Cuminum cyminum* is the most valid confirmatory tool for the detection of CaO NPs. UV-Visible spectroscopy is very common and useful tool in analytical chemistry. This tool is basically used for the identification of chemical products and their qualitative analysis. The molecules or atoms absorb UV-Visible radiation and their electrons excited as they progress from lower to higher energy levels. In the present study, we have obtained the UV-Visible spectra in the range of wavelength 260-410 nm. The broad peaks have obtained at 272 and 350 nm; this indicating the formation of calcium oxide nanoparticles at such wavelength.

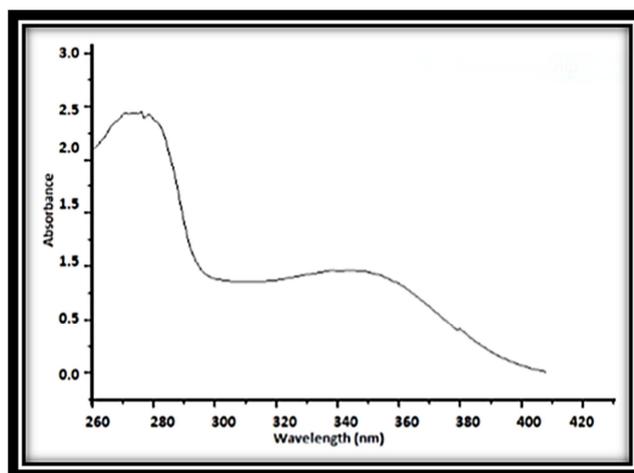


Figure 11. UV-Vis spectrum of CaONPs.

### 3.2. Characterization of CaO NPs by XRD Analysis

The XRD techniques are widely used to determine the size of particles and the structure of nanoparticles. The crystalline nature of the synthesized CaO nanoparticles was further verified by X-ray diffraction pattern (XRD). XRD measurement of biologically synthesized CaO NPs from CaCl<sub>2</sub> and NaOH done. Using Cu K radiation and an X-ray diffraction method (Philips PAN analytical, The Netherlands), the origin and phase evolution of calcined powder and sintered samples is investigated. The voltage and current of the generator were set to 35 KV and 25 mA, respectively. In continuous scan mode, the CaO sample was scanned in two ranges: 10 to 90. The scan speed was 0.04 seconds per second. The search match feature of X'pert high score programme was used to identify the phases present in the study. The Scherrer's equation was used to calculate the crystallite size of the calcined powders based on X-ray line broadening:

CaO NPs show peaks corresponding to the planes at  $2\theta =$

23.01° (012), 2θ = 29.36° (104), 2θ = 35.90° (110), 2θ = 39.37° (113), 2θ = 43.12° (202), 2θ = 47.49° (018), 2θ = 48.46° (116), 2θ = 57.34° (122). The diffraction patterns for the calcium oxide nanoparticles contain diffraction peaks for cubic CaO (JCPDS2-1095) at 23.01°, 29.36°, 35.90°, 39.37°, 43.12°, 47.49°, 48.46 and 57.34°. Compatible with (012), (104), (110), (113) and (202), (018), (116) and (122) crystal planes, respectively, and no other impurity peaks were detected. From the diffraction peaks using the Debye-Scherrer equations, the average crystallite size was determined.

$$d = k \lambda / (\beta \cos \theta)$$

where d is the crystal particle dimension, k is the Scherrer constant (0.9), λ is the X-ray wavelength (0.154178 nm), β is the XRD peak half-height range, and θ is the Bragg diffraction angle. Where D denotes the crystal size; λ is the X-ray wavelength (= 0.15406 nm) for CuK; K denotes the line width at half-maximum height; and β is the line width at half-maximum

height. All the reflection peaks in the Figure can be conveniently indexed with a cell parameter of 4.22700 Å. to the pure cubic process of CaO (JCDPS No. 75- 0447). The density of CaO NPs measured was found to be 2.71g/cm<sup>3</sup> [23].

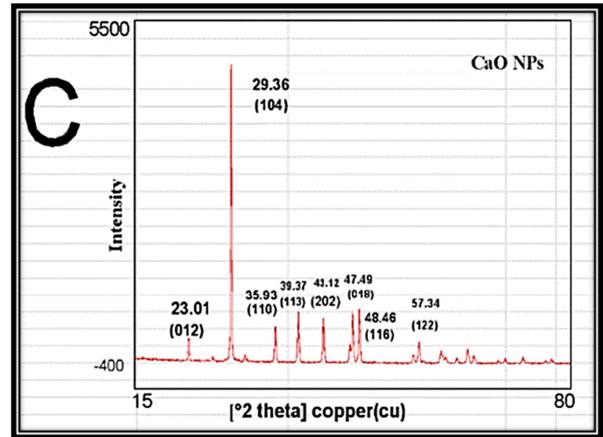


Figure 12. XRD spectrum of CaONPs.

	A	B	C	D	E	F	G	H	I	J
1	K	λ	Peak Position 2θ	β (FWHM)	crystallite size (L)					
2		(Å)	θ	θ	nm					
3	0.94	1.54178	23.015	0.197	43.23502					
4	0.94	1.54178	29.361	0.197	43.57338					
5	0.94	1.54178	35.361	0.246	35.57305					
6	0.94	1.54178	39.377	0.246	35.85088					
7	0.94	1.54178	43.125	0.246	36.14806					
8	0.94	1.54178	47.466	0.197	46.04485					
9	0.94	1.54178	48.466	0.246	37.01675					
10	0.94	1.54178	57.364	0.295	32.08516					
11					<b>Average size = 309.52715/8 = 38.690 nm</b>					

Figure 13. Determination of particle size from XRD data.

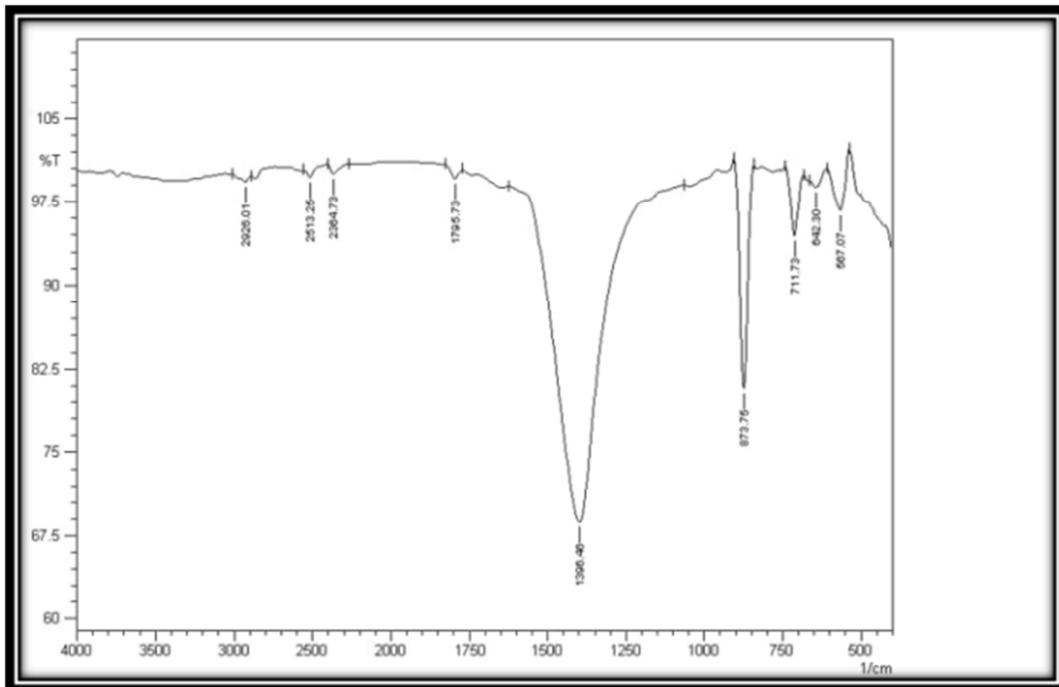


Figure 14. FTIR spectra of green synthesized CaO nanoparticles.

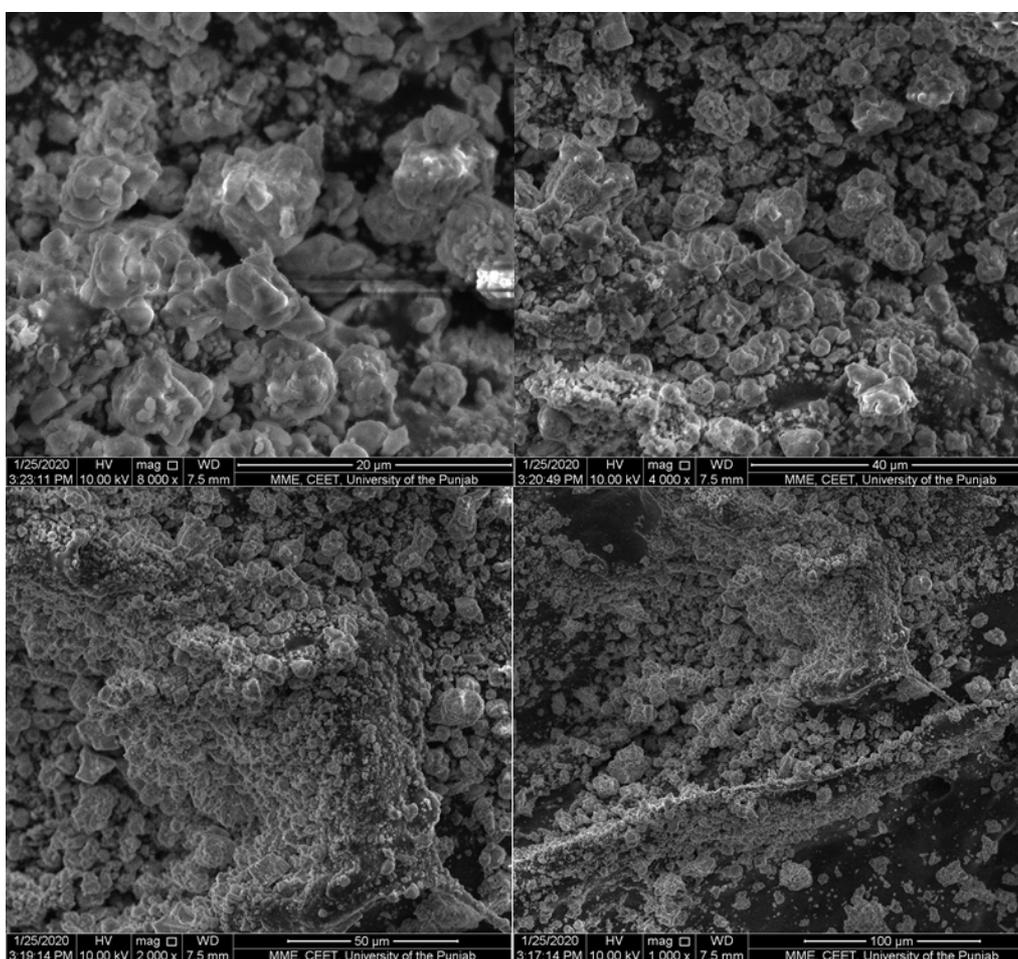
### 3.3. Characterization of CaONPs by FTIR Analysis

Biomolecules which are responsible for the capping and reduction of CaO NPs are confirmed by Fourier transform infrared spectroscopy (FTIR). In order to support that Ca ions reacted with the OH functional group of the secondary compound existing in the system, the FTIR analysis was also applied in this experiment. The FTIR analysis was measured in the range of 500-4000  $\text{cm}^{-1}$ , where it indicated vibration peak at 567.07, 642.30  $\text{cm}^{-1}$ , 711.73  $\text{cm}^{-1}$ , 873.75  $\text{cm}^{-1}$ , 1396.46  $\text{cm}^{-1}$ , 1795.3  $\text{cm}^{-1}$ , 2364.73  $\text{cm}^{-1}$ , 2513.25  $\text{cm}^{-1}$  and 2926.01  $\text{cm}^{-1}$ . The peak observed at 2926  $\text{cm}^{-1}$  is corresponding to the C-H stretching vibration depicting the presence of an alkane. The peak obtained at 2513  $\text{cm}^{-1}$  is confirming the presence of S-H Thiol, peak at 2364  $\text{cm}^{-1}$  corresponds to C=O stretching  $\text{CO}_2$ , peak at 1795  $\text{cm}^{-1}$  is

confirming the presence of C=O stretching, peak at 1396  $\text{cm}^{-1}$  corresponds to the presence of C-H Bending and the peak at 873.75  $\text{cm}^{-1}$  confirms the presence of C=C bending. The observed peaks at 642.30  $\text{cm}^{-1}$  and 567.07  $\text{cm}^{-1}$  confirm the presence of CaO NPs [24].

*Table 1. Elemental ratio of CaO nanoparticles.*

S. No	Absorption	Identified functional group
	2926 $\text{cm}^{-1}$	-CH stretching (alkanes)
	2513 $\text{cm}^{-1}$	S-H Thiol
	2364 $\text{cm}^{-1}$	C=O stretching $\text{CO}_2$
	1798 $\text{cm}^{-1}$	C=O stretching Anhydride
	1396 $\text{cm}^{-1}$	C-H Bending (Aldehydes)
	873 $\text{cm}^{-1}$	C=C bending (Alkene)
	711 $\text{cm}^{-1}$	Ca-O stretching vibrations
	642 $\text{cm}^{-1}$	Ca-O stretching vibrations



*Figure 15. SEM images of CaONPs Showing shape, morphology, size and monodispersive clusters.*

### 3.4. Scanning Electron Microscopy (SEM)

The surface morphologies of nanoparticles, which are circular and irregular, are seen. The SEM photographs of the CaO nanoparticles collected reveal that the powders are made up of a combination of large grains and fine substance particles. The scale, shape, and position of individual nanoparticles can be determined using scanning electron

microscopy techniques. The Jeol JSM-6480 LV SEM machine was used to classify mean particle size and morphology of nanoparticles in this study. A small drop of the CaO powder sample NP solution was sonicated with purified water and put on a glass slide to rinse. The nanoparticle size is shown in the SEM micrographs by the bar at 10m and 4,000x. SEM studies show that the calcinated product brings more quick grain growth and larger values of particle size.

### 3.5. Characterization of CaONPs by EDX

Energy dispersive X-ray spectroscopy (EDX or EDS) spectrum of the relative counts of the detected X-rays versus energy is determined. It is for quantitative and qualitative determinations of the elements as shown in Figure (Rezaei, Khajenoori, & Nematollahi, 2011). EDX divulge the presence of Mg and O. Graph of EDX analysis indicates the growing Mg and O nanoparticles composition. In Graph, the peaks other than Ca and O indicate the impurity (Jhansi et al., 2017). It is apparent that prepared nanoparticles are the

composition of Ca and O alone and other elements e.g. Sodium, calcium etc. in very minute quantities have been found in the EDX spectrum (Somanathan, Krishna, Saravanan, Kumar, & Kumar, 2016). Four elements including Oxygen (O), Sodium (Na) and Calcium (Ca) were found in the EDX spectrum of nanoparticles of CaO with percentage weight of, 46.53, 53.47, 35.58 and 64.42 and atomic percentage of 25.78, 74.22, 58.04 and 41.96 respectively. The observed a couple of intense peaks correspond to the composition of Ca and O and rest of the peaks arises by carbon tape or the sample holder as evident from the Figure.

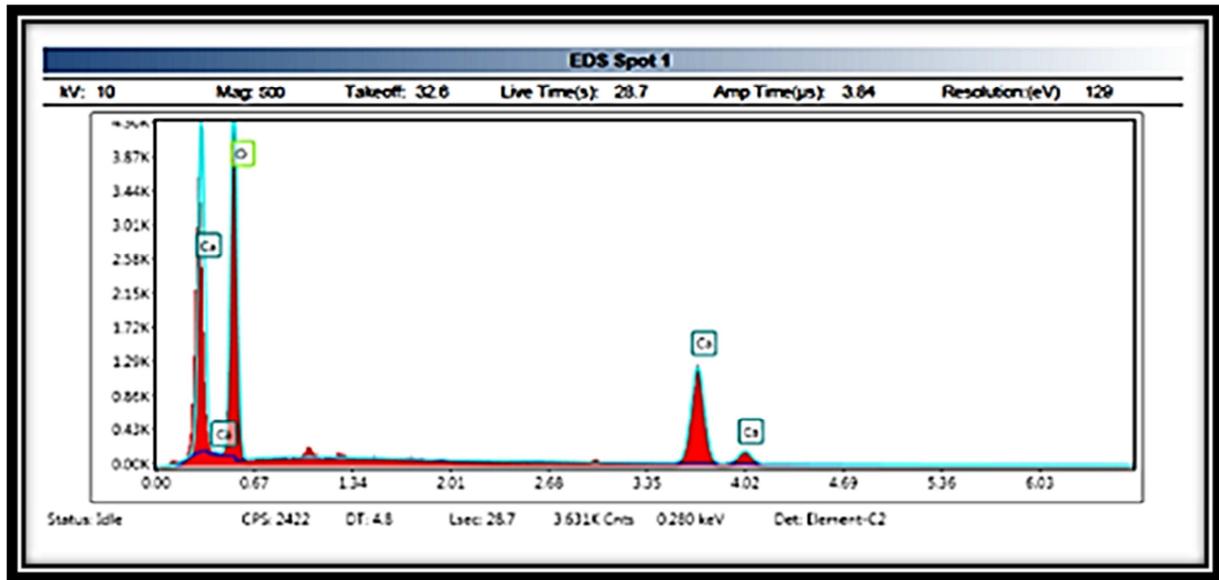


Figure 16. EDX spectra of green synthesized CaO nanoparticle.

Table 2. Elemental ratio of CaO nanoparticles.

Element	Weight %	Atomic %
O	53.47	74.22
Ca	46.53	26.78
100	100.0	100.0

### 3.6. Antifungal Activity of CaONPS

The antifungal activity of CaO nanoparticles was determined by disc diffusion assay against *Aspergillusflavus*, *Aspergillusparasiticus*, *Fusariumoryzae*, *Fusariumtritichum* and *Aspergillusoryzae*. The zones of inhibitions are presented in table 3. It was concluded that from the data that CaO nanoparticles showed strong activity against the *Aspergillusparasiticus* (24 mm zone of inhibition). It was observed that CaO nanoparticles are least fungal source respectively. The Fluconazole showed D/Z (23mm, 19mm,

15mm, 18mm and 13mm) against *Aspergillusflavus*, *Aspergillusparaciticus*, *Fusariumoryzae*, *Fusariumtritichum* and *Aspergillusoryzae* respectively.



Figure 17. Antifungal activity of CaO NPs.

Table 3. Antifungal activity of CaO NPs.

Fungal strain	CaO-NPs DIZ (mm)	Flucanazol DIZ (mm)
<i>Aspergillusparasiticus</i>	23 ± 1.36 <sup>a</sup>	30 ± 1.20
<i>Aspergillusflavus</i>	19 ± 2.74 <sup>bc</sup>	29 ± 2.10
<i>Fusariumoryzae</i>	18 ± 3.24 <sup>de</sup>	25 ± 2.44
<i>Fusariumtritichum</i>	13 ± 1.23 <sup>f</sup>	23 ± 1.90
<i>Aspergillusoryzae</i>	15 ± 3.24 <sup>ef</sup>	18 ± 3.10

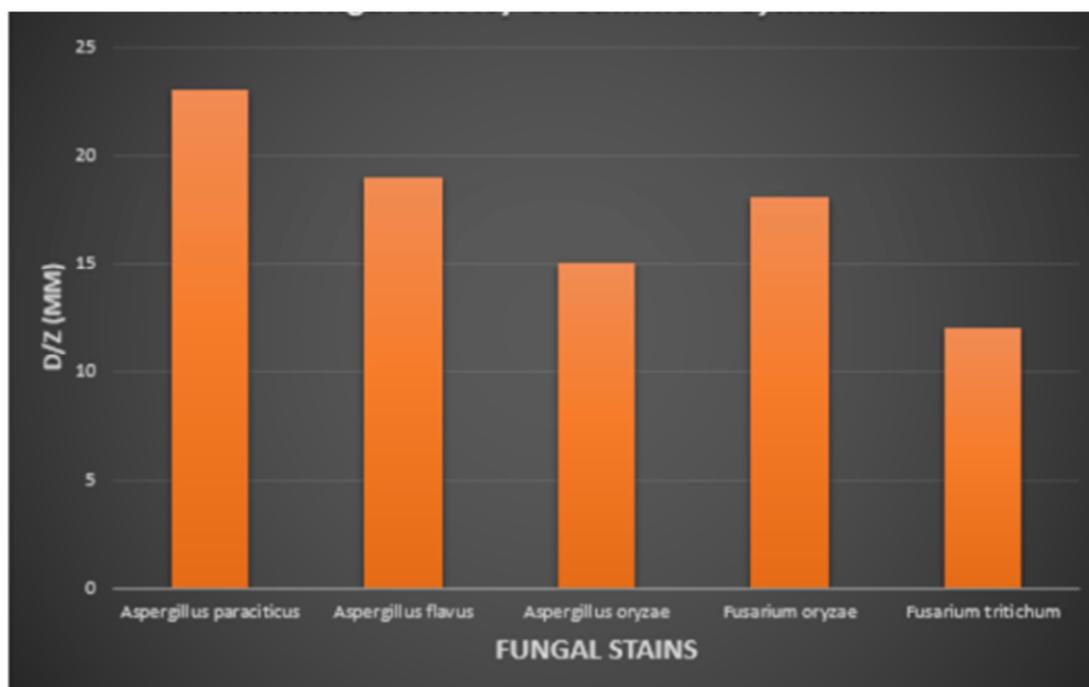


Figure 18. Zone of inhibition of CaO NPs against *Aspergillus paraciticus*.

## 4. Phytochemical Screening of *Cuminum Cyminum*

Tests were performed in order to evaluate the presence of certain phytochemicals including Alkaloids, Flavonoids, Phenols, Tannins, Glycoside, Saponins, Quinone and Terpenoids. The symbol + indicates presence while the symbol – indicates absence. The result of phytochemical analysis reveals the presence of Carbohydrates, proteins, amino acid, flavonoids, phenolic compounds and Alkaloids and absence of Steroids [24].

### 4.1. Alkaloids

These are secondary metabolites of plants. Defence mechanism of the plant is due to these metabolites Alkaloid shows effectiveness against malaria and used as allopathic [25]. Alkaloid shows activity like cytotoxicity [26]. In the present study, reddish brown color in the test tube, reveals the presence of alkaloids in the extract of *Cuminum cyminum*. The result of present study is in agreement with the previous study of [21] who reported the presence of alkaloids in *Cuminum cyminum*.

### 4.2. Carbohydrates

Carbohydrates are most commonly called sugars, are the medium for energy metabolism, they can influence lipid metabolism insulin and blood glucose through fermentation. They help to control of diabetes, body weight, cardiovascular disease, bone mineral density, resistance to gut infection and constipation [20]. In the present study, three tests and their

color reveals the presence of carbohydrates in extract of *Cuminum cyminum*. The result of present study is in agreement with the previous study of Sawant [21] who reported the presence of carbohydrates in *Cuminum cyminum*.

### 4.3. Proteins

Protein is biomolecules which comprised from long chain of amino acid. Protein in *Cuminum cyminum* supplement in maintains of tissue strength, repair, hyperglycemic response, reduced cancer growth and producing neurotransmitters. In the present study, yellow color in the test tube, reveals the presence of proteins in the extract of *Cuminum cyminum*. The result of present study is in agreement with the previous study of [19] who reported the presence of proteins in *Cuminum cyminum*.

### 4.4. Amino Acids

In the present study, purple color in the test tube, reveals the presence of amino acids in the extract of *Cuminum cyminum*. The result of present study is in agreement with the previous study of [24] who reported the presence of amino acids in *Cuminum cyminum*.

### 4.5. Phenols

Phenol is a crystalline substance, soluble in organic solvents and in water, colourless, characteristic odour [27]. This derivative has certain biological benefits such as antioxidant, anti-diabetic antibacterial, antifungal, insecticidal, anti-carcinogenic capacities [28]. In the present study, dark green color in the test tube, reveals the presence of phenolic compounds in the extract of *Cuminum cyminum*. The result of present study is in agreement with the previous

study of [22] who reported the presence of phenolic compounds in *Cuminum cyminum*.

#### 4.6. Flavonoids

Flavonoids are polyphenolic secondary metabolites present in plants. Flavonoids show biological activities, include anti-inflammatory, vasodilating actions, antiallergenic, antiviral anti-carcinogenic activities [29] In the present study, yellow color in the test tube, reveals the presence of flavonoids in the extract of *Cuminum cyminum*. The result of present study is in agreement with the previous study of [15] who reported the presence of flavonoids in *Cuminum cyminum*.

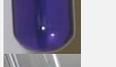
#### 4.7. Anthraquinones

Anthraquinones (9,10-dioxoanthracenes) comprised an imperative class of synthetic and natural and these compounds have variety of applications. Therapeutic application includes arthritis, constipation, cancer and multiple sclerosis [30]. In the present study, rose pink coloration, reveals the presence of anthraquinones in the extract of *Cuminum cyminum*. The result of present study is in agreement with the previous study of [18] who reported the presence of anthraquinones in *Cuminum cyminum*.

#### 4.8. Steroid

Steroid are hormones that perform biological activity. They are necessary for reproduction and growth. Steroids are used in the treatment of diseases. It is also used in lowering of blood pressure. It has different biological activities like cardiovascular protection, anti carcinogen and anti inflammation [31]. In the present study, white color in test tube, reveals the absence of steroid in the extract of *Cuminum cyminum*. The result of present study is not in agreement with the previous study of [32] who reported the presence of steroid in *Cuminum cyminum*.

**Table 4.** Screening of phytochemicals from extract of *Cuminumcyminum*.

Sr. no.	Phytochemical Analysis	Results	
1.	Alkaloids (by Wagner test)	(+)	
2.	Carbohydrates (by Iodine Test)	(+)	
	Carbohydrates (by Fehling Test)	(+)	
	Carbohydrates (by Molisch's Test)	(+)	
3.	Protein	(+)	
4.	Amino acids	(+)	
5.	Phenol (Ferric chloride test)	(+)	

Sr. no.	Phytochemical Analysis	Results	
6.	Flavonoids (Alkaline reagent test)	(+)	
7.	Anthraquinones	(+)	
8.	Steroid	(+)	

## 5. Conclusion

Calcium oxide nanoparticles were successfully manufactured by green methods employing *Cuminumcyminum* and  $\text{CaCl}_2$  salt in NaOH, according to the findings. Plant extracts serve as capping agents and stabilisers in the manufacture of CaO nanoparticles, hence the green synthesis approach was chosen. The CaO nanoparticles were studied using scanning electron microscopy (SEM), X-Ray Diffraction (XRD), Fourier Transform Infrared Spectroscopy (FTIR), and Electron Dispersive Spectroscopy (EDX). The existence of active phytoconstituents was validated by a phytochemical study of *Cuminumcyminum* extract. SEM proved the morphology and monodispersive clusters of CaONPS, while FTIR confirmed the presence of CaONP with defined peaks corresponding to the synthesis of CaO nanoparticles. Following SEM and FTIR, EDS was used to confirm the existence of CaO nanoparticles by displaying the paternity of Ca and O atoms in the form of peaks. The results of the XRD examination were even more conclusive. The current research is cost-effective and can be used to create antifungal reagents in the pharmaceutical and food industries.

## References

- [1] Zhang, L., Gu, F., Chan, J., Wang, A., Langer, R., & Farokhzad, O. (2008). Nanoparticles in medicine: therapeutic applications and developments. *Clinical pharmacology & therapeutics*, 83 (5), 761-769.
- [2] Rowel, R., Sheikhattari, P., Barber, T. M., & Evans-Holland, M. (2012). Introduction of a guide to enhance risk communication among low-income and minority populations: a grassroots community engagement approach. *Health promotion practice*, 13 (1), 124-132.
- [3] Evanoff Jr, D. D., & Chumanov, G. (2005). Synthesis and optical properties of silver nanoparticles and arrays. *ChemPhysChem*, 6 (7), 1221-1231.
- [4] Ramola, B., Joshi, N. C., Ramola, M., Chhabra, J., & Singh, A. (2019). Green synthesis, characterisations and antimicrobial activities of CaO nanoparticles. *Oriental Journal of Chemistry*, 35 (3), 1154-1157.
- [5] Kiang, C.-H., Goddard III, W. A., Beyers, R., Salem, J. R., & Bethune, D. S. (1996). Catalytic effects of heavy metals on the growth of carbon nanotubes and nanoparticles. *Journal of Physics and Chemistry of Solids*, 57 (1), 35-39.

- [6] Adams, W. W., & Baughman, R. H. (2005). Richard E. Smalley (1943-2005). *Science*, 310 (5756), 1916-1916.
- [7] Roy, S., & Das, T. K. (2015). Plant mediated green synthesis of silver nanoparticles. *A Review. Int. J. Plant Biol. Res*, 3 (3), 1044-1055.
- [8] Madhusudhana, N., Yogendra, K., & Mahadevan, K. (2012). A comparative study on photocatalytic degradation of Violet GL2B azo dye using CaO and TiO<sub>2</sub> nanoparticles. *Int. J. Eng. Res. Appl*, 2 (5), 1300-1307.
- [9] Hernandez, Y., Nicolosi, V., Lotya, M., Blighe, F. M., Sun, Z., De, S.,... Gun'Ko, Y. K. (2008). High-yield production of graphene by liquid-phase exfoliation of graphite. *Nature nanotechnology*, 3 (9), 563-568.
- [10] Sheeparamatti, B., Sheeparamatti, R., & Kadadevaramath, J. (2007). Nanotechnology: Inspiration from nature. *IETE Technical Review*, 24 (1), 5-8.
- [11] Abdelghany, T., Al-Rajhi, A. M., Al Abboud, M. A., Alawlaqi, M., Magdah, A. G., Helmy, E. A., & Mabrouk, A. S. (2018). Recent advances in green synthesis of silver nanoparticles and their applications: about future directions. A review. *BioNanoScience*, 8 (1), 5-16.
- [12] Shao, Y., Wu, C., Wu, T., Yuan, C., Chen, S., Ding, T.,... Hu, Y. (2018). Green synthesis of sodium alginate-silver nanoparticles and their antibacterial activity. *International journal of biological macromolecules*, 111, 1281-1292.
- [13] Raghavan, S. (2007). Handbook of spices, seasonings, and flavorings. Boca Raton: CRC: Taylor & Francis Group.
- [14] Singh, R. P., Gangadharappa, H., & Mruthunjaya, K. (2017). *Cuminum cyminum*—A popular spice: An updated review. *Pharmacognosy Journal*, 9 (3).
- [15] Kooti, W., Hasanzadeh-Noohi, Z., Sharafi-Ahvazi, N., Asadi-Samani, M., & Ashtary-Larky, D. (2016). Phytochemistry, pharmacology, and therapeutic uses of black seed (*Nigella sativa*). *Chinese journal of natural medicines*, 14 (10), 732-745.
- [16] Agarwal, U., Pathak, D. P., Kapoor, G., Bhutani, R., Roper, R., Gupta, V., & Kant, R. (2017). Review on *Cuminum Cyminum*—Nature's Magical Seeds. *Journal of Chemical and Pharmaceutical Research*, 9 (9), 180-187.
- [17] Sundrarajan, M., Suresh, J., & Gandhi, R. R. (2012). A comparative study on antibacterial properties of MgO nanoparticles prepared under different calcination temperature. *Digest journal of nanomaterials and biostructures*, 7 (3), 983-989.
- [18] Klančnik, A., Piskernik, S., Jeršek, B., & Možina, S. S. (2010). Evaluation of diffusion and dilution methods to determine the antibacterial activity of plant extracts. *Journal of microbiological methods*, 81 (2), 121-126.
- [19] Swenson, J. M., Killgore, G. E., & Tenover, F. C. (2004). Antimicrobial susceptibility testing of *Acinetobacter* spp. by NCCLS broth microdilution and disk diffusion methods. *Journal of clinical microbiology*, 42 (11), 5102-5108.
- [20] Cummings, J., & Stephen, A. (2007). Carbohydrate terminology and classification. *European journal of clinical nutrition*, 61 (1), S5-S18.
- [21] Sawant, R., & Godghate, A. (2013). Qualitative phytochemical screening of rhizomes of *Curcuma longa* Linn. *International Journal of Science, Environment and Technology*, 2 (4), 634-641.
- [22] Amin, M., Jassal, M., & Tygi, S. (2013). Phytochemical screening and isolation of eugenol from *Syzygium aromaticum* by gas chromatography. *International Journal of Research in Phytochemistry and Pharmacology*, 3 (1), 74-77.
- [23] Walkley, B., Page, S. J., Rees, G. J., Provis, J. L., & Hanna, J. V. (2019). Nanostructure of CaO-(Na<sub>2</sub>O)-Al<sub>2</sub>O<sub>3</sub>-SiO<sub>2</sub>-H<sub>2</sub>O Gels Revealed by Multinuclear Solid-State Magic Angle Spinning and Multiple Quantum Magic Angle Spinning Nuclear Magnetic Resonance Spectroscopy. *The Journal of Physical Chemistry C*, 124 (2), 1681-1694.
- [24] Anantharaman, A., Ramalakshmi, S., & George, M. (2016). Green synthesis of calcium oxide nanoparticles and its applications. *Int. J. Eng. Res. Appl*, 6 (10), 27-31.
- [25] Fischer, D. C. H., de Amorim Gualda, N. C., Bachiega, D., Carvalho, C. S., Lupo, F. N., Bonotto, S. V.,... Avila, P. E. (2004). In vitro screening for antiparasitic activity of isoquinoline alkaloids from Brazilian plant species. *Acta Tropica*, 92 (3), 261-266.
- [26] Evans, W. (2005). Trease and Evans Pharmacognosy 15 edn. Division of Reed Elsevier India Pvt. Ltd., New Delhi, India, ISBN-13, 978-981.
- [27] Michałowicz, J., & Duda, W. (2007). Phenols--Sources and Toxicity. *Polish Journal of Environmental Studies*, 16 (3).
- [28] Zachariah, T., Krishnamoorthy, B., Rema, J., & Mathew, P. (2005). Oil constituents in bud and pedicel of clove (*Syzygium aromaticum*). *Indian Perfumer*, 49 (3), 313.
- [29] Pietta, P.-G. (2000). Flavonoids as antioxidants. *Journal of natural products*, 63 (7), 1035-1042.
- [30] Malik, E. M., & Müller, C. E. (2016). Anthraquinones as pharmacological tools and drugs. *Medicinal research reviews*, 36 (4), 705-748.
- [31] Nyarko, A. A., & Addy, M. E. (1990). Effect of aqueous extract of *Adenia cissampeloides* on blood pressure and serum analytes of hypertensive patients. *Phytotherapy Research*, 4 (1), 25-28.
- [32] Vasantharaj, S., Sathiyavimal, S., & Hemashenpagam, N. (2013). Antimicrobial activity of silver nanoparticle synthesized by using *Costeus igneus*. *Research Journal of Pharmaceutical, Biological and Chemical Sciences*, 4 (2), 733-738.