

ORIGINAL ARTICLE

Estimation of organic carbon in Nanocrystalline Diamond medicinal product - Heerak Bhasm and its SEM analysis

Gupta Swaroopa Rani N1* and Bhadke Pranay D2

¹Professor, ²Research Scholar, Department of Chemistry, Brijlal Biyani Science College, Amravati, Maharashtra, India *Corresponding author Email- <u>swargupta@yahoo.com</u>

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Abstract

Diamond is a solid form of the element carbon with its atoms arranged in a crystal structure called diamond cubic. Diamond has the highest hardness and thermal conductivity of any natural material, properties that are used in major industrial applications such as cutting and polishing tools. They are also the reason that diamond anvil cells can subject materials to pressures found deep in the Earth. Diamond is chiefly categorized in to four types, mainly, white, red, yellow and black. The white colored diamond is mainly suggested for internal administration, whereas the black and red-colored variety is significant in case of several health conditions and even prevents premature death of babies. The yellow-colored type is mainly used for providing strength.

Heerak Bhasma is an ayurvedic preparation obtained from purified diamond ash. Diamonds are solid form of the element carbon. Under extreme high temperature and pressure far below the earth's surface, the carbon atoms bond in a unique way that results in diamond's beautiful and rare crystalline structure, which is further embellished to make extra-ordinary pieces of jewellery that sparkles with radiance. But apart from being a timeless, elegant, and stunning piece of art, there is more to a diamond than what meets the eye. It used as a beauty ingredient for enhancing skin health or as a herbal compound for promoting health. Heerak Bhasma has been mentioned in several ayurvedic scriptures as an ultimate remedy for several cardiac anomalies, and anemia owing to its enormous therapeutic properties. This incredible herbo-mineral formulation is classified as a potent cardiotonic, aphrodisiac, and immunomodulatory, and hence used for the treatment and management of infertility, insomnia, diabetes, obesity and thus improves overall stamina and body immunity. This paper deals with estimation of organic carbon in nanocrystalline diamond medicinal product - Heerak Bhasm. It also includes Scanning Electron Microscope (SEM) analysis of Heerak Bhasm.

This research, along with better regulation and reporting, will enable consumers to choose products with confidence. This in turn will allow companies to benefit from these novel technologies in the long term while retaining customer confidence.

Keywords: Diamond, Heerak Bhasma, Organic Carbon, Organic matter, SEM Analysis.

1. Introduction

Diamond

Diamond is a solid form of the element carbon with its atoms arranged in a crystal structure called diamond cubic. At room temperature and pressure, another solid form of carbon known as graphite is the chemically stable form of carbon, but diamond converts to it extremely slowly. Diamond has the highest hardness and thermal conductivity of any natural material, properties that are used in major industrial applications such as cutting and polishing tools. They are also the reason that diamond anvil cells can subject materials to pressures found deep in the Earth.

Because the arrangement of atoms in diamond is extremely rigid, few types of impurity can contaminate it (two exceptions are boron and nitrogen). Small numbers of defects or impurities (about one per million of lattice atoms) color diamond blue (boron), yellow (nitrogen), brown (defects), green (radiation exposure), purple, pink, orange, or red. Diamond also has a very high refractive index and a relatively high optical dispersion.

Diamonds have been adopted for many uses because of the material's exceptional physical characteristics. It has the highest thermal conductivity and the highest sound velocity. It has low adhesion and friction, and its coefficient of thermal expansion is extremely low. Its optical transparency extends from the far infrared to the deep ultraviolet and it has high optical dispersion. It also has high electrical resistance. It is chemically inert, not reacting with most corrosive substances, and has excellent biological compatibility. [1]

The most common crystal structure of diamond is called diamond cubic. It is formed of unit cells (Figure 1) stacked together. Although there are 18 atoms in the figure, each corner atom is shared by eight unit cells and each atom in the center of a face is shared by two, so there are a total of eight atoms per unit cell.[2] The length of each side of the unit cell is denoted by a and is 3.567 angstroms.[3] Diamond is chiefly categorized in to four types, mainly, white, red, yellow and black. According to ayurvedic scriptures, the diamond which is more-or-less round in size and possess high gloss is termed as male. The white colored diamond is mainly suggested for internal administration, whereas the black and red-colored variety is significant in case of several health conditions and even prevents premature death of babies. The yellow-colored type is mainly used for providing strength.

Heerak Bhasma

Heerak Bhasma is an ayurvedic preparation obtained from purified diamond ash. Diamonds are solid form of the element carbon. Under extreme high temperature and pressure far below the earth's surface, the carbon atoms bond in a unique way that results in diamond's beautiful and rare crystalline structure, which is further embellished to make extra-ordinary pieces of jewellery that sparkles with radiance. But apart from being a timeless, elegant, and stunning piece of art, there is more to a diamond than what meets the eye. It used as a beauty ingredient for enhancing skin health or as a herbal compound for promoting health.

Chemical Composition of Heerak Bhasma

It mainly contains carbon, iron and oxygen. But other essential elements that are present in moderate quantity include sodium, magnesium, potassium, calcium, chromium, aluminum, silicon, phosphorus, and sulfur. This ayurvedic herbo-mineral compound is partly soluble in water and sparingly soluble in organic solvents like chloroform and methanol. [4]

Heerak Bhasm can be formulated by taking finely powdered diamond nano particles and equal quantities of both gandhaka and rasa sindhura. Taking all three in a mortar pestle and grounding well till it becomes fine powder. Heating it in the absence of air and allowing the mixture to cool down on its own. Repeating the above process 14 times to get pure quality of Heerak Bhasma.

Health Benefits of Heerak Bhasma

Promotes Cardiac Functioning: Being a cardiotonic compound, heerak bhasma is extremely effective in treating various heart ailments due to its strong antioxidative nature. It strengthens the heart muscles, prevents the deposition of plaque in the blood vessels and also averts the formation of atherosclerosis (i.e., hardening of the arteries), and hence reduces the risk of heart attacks, heart blocks, blood clots, mitral regurgitation, ischemic cardiomyopathy. Additionally, it improves blood flow to the heart muscles and reduces the sensation of heaviness, pressure, and tightness in the chest, thus treating angina pectoris or any other kind of chest pain.

Treats Metastasis: Cancer is termed as the abnormal growth of healthy body cells. It is extremely lifethreatening and may require extensive medical procedures involving surgery, radiation therapy, chemotherapy and several other treatments. Imbued with powerful anti-cancer, antimetastatic and antimutagenic properties, heerakbhasma offers a potent alternative treatment of cancer. Not only does it increase the immune power and vitality of the patient but also reduce the size of the tumor and prevent further metastasis. However, the treatment with heerak bhasma often takes a long time period from 6 -12 months at a stretch to show positive results.

Remedies Anemia: Heerak bhasma is an absolute remedy for treating and preventing iron deficiency anemia or microcytic anemia. Additionally, it is also quintessential for treating the various symptoms of anemia like general fatigue, physical weakness and yellow discoloration of the skin, vertigo, laziness, shortness of breath, dizziness, swelling and headaches.

Relaxes The Mind: The abundance of adaptogenic, and sedative properties in this mineral formulation not only improves the mood but also relieves stress. It holds high significance in providing mental stability by regulating the stress hormone, i.e., serotonin which in turn reduces various symptoms of anxiety including restlessness, uneasiness, cold hands, and feet, etc. and bestows a fresh and happy feeling.

Reduces Pain and Inflammation: Due to the presence of strong pain-relieving and anti-inflammatory effects, Heerakbhasma is extensively used to provide relief in case of painful conditions like arthritis, muscle spasms, etc. The pain due to Rheumatoid arthritis, also known as Amavata which chiefly stems due to the accumulation of toxins in the joints and vitiation of Vata Doshas, can be alleviated on using this incredible diamond-based formulation.

Heerak Bhasma has been mentioned in several ayurvedic scriptures as an ultimate remedy for several cardiac anomalies, and anemia owing to its enormous therapeutic properties. This incredible herbo-mineral formulation is classified as a potent cardiotonic, aphrodisiac, and immunomodulatory, and hence used for the treatment and management of infertility, insomnia, diabetes, obesity and thus improves overall stamina and body immunity.

Total organic carbon (TOC)

Total organic carbon (TOC) is the amount of carbon found in an organic compound and is often used as a non-specific indicator of water quality or cleanliness of pharmaceutical manufacturing equipment. TOC may also refer to the amount of organic carbon in soil, or in a geological formation, particularly the source rock for a petroleum play; 2% is a rough minimum. [5] For marine surface sediments average TOC content is 0.5% in the deep ocean, and 2% along the eastern margins. [6]

A typical analysis for total carbon (TC) measures both the total organic carbon (TOC) present and the complementing total inorganic carbon (TIC), the latter representing the amount of non-organic carbon, like carbon in carbonate minerals. Subtracting the inorganic carbon from the total carbon yields TOC. Another common variant of TOC analysis involves removing the TIC portion first and then measuring the leftover carbon. This method involves purging an acidified sample with carbon-free air or nitrogen prior to measurement, and so is more accurately called nonpurgeable organic carbon (NPOC). [7]

Since all TOC analyzers only actually measure total carbon, TOC analysis always requires some accounting

for the inorganic carbon that is always present. One analysis technique involves a two-stage process commonly referred to as TC-IC. It measures the amount of inorganic carbon (IC) evolved from an acidified aliquot of a sample and also the amount of total carbon (TC) present in the sample. TOC is calculated by subtraction of the IC value from the TC of the sample. Another variant employs acidification of the sample to evolve carbon dioxide and measuring it as inorganic carbon (IC), then oxidizing and measuring the remaining non-purgeable organic carbon (NPOC). This is called TIC-NPOC analysis. A more common method directly measures TOC in the sample by again acidifying the sample to a pH value of two or less to release the IC gas but in this case to the air not for measurement. The remaining non-purgeable CO2 gas (NPOC) contained in the liquid aliquot is then oxidized releasing the gases. These gases are then sent to the detector for measurement.

Whether the analysis of TOC is by TC-IC or NPOC methods, it may be broken into three main stages:

Acidification

The first stage is acidification of the sample for the removal of the IC and POC gases. The release of these gases to the detector for measurement or to the air is dependent upon which type of analysis is of interest, the former for TC-IC and the latter for TOC (NPOC). Addition of acid and inert-gas sparging allows all bicarbonate and carbonate ions to be converted to carbon dioxide, and this IC product vented along with any purgeable organic carbon (POC) that was present.

Oxidation

The second stage is the oxidation of the carbon in the remaining sample in the form of carbon dioxide (CO2) and other gases. Modern TOC analyzers perform this oxidation step by several processes: High temperature combustion [8, 9] High temperature catalytic oxidation (HTCO) [10-13] Photo-oxidation alone Thermo-chemical oxidation Photo-chemical oxidation Electrolytic oxidation

Detection and Quantification

Accurate detection and quantification are the most vital components of the TOC analysis process. Conductivity and non-dispersive infrared (NDIR) are the two common detection methods used in modern TOC analyzers. [14]

Diamonds, the most precious gemstones, have been widely recognized for their exceptional powers and miraculous therapeutic abilities. Various micro and nanocrystalline diamond skin care products and their applications are studied with a particular focus on Diamond Cellular Anti-Ageing Cream which deals with Scanning Electron Microscope (SEM) images, Transmission Electron Microscope (TEM) images and FTIR spectra of Diamond Cellular Anti-Ageing Cream. [15].

This paper deals with estimation of organic carbon in nanocrystalline diamond medicinal product - Heerak Bhasm. It also includes Scanning Electron Microscope (SEM) analysis of Heerak Bhasm. This research, along with better regulation and reporting, will enable consumers to choose products with confidence. This in turn will allow companies to benefit from these novel technologies in the long term while retaining customer confidence.

2. Materials and Method

1. Estimation of Organic Carbon in Heerak Bhasm by Walkley-Black method

The estimation of Organic Carbon in Heerak Bhasm is based on the Walkley & Black chromic acid wet oxidation method. Oxidizable organic carbon in the Heerak Bhasm is oxidised by potassium dichromate $(K_2Cr_2O_7)$ solution in concentrated sulfuric acid. The heat of reaction raises the temperature which is sufficient to induce substantial oxidation.

Chemical reaction is as follows: 2 K₂Cr₂O₇ + 8 H₂SO₄ + 3 C \rightarrow 2 K₂SO₄ + 2 Cr₂(SO₄)₃ + 3 CO₂ + 8 H₂O The K₂Cr₂O₇ reduced during the reaction with Heerak Bhasm is proportional to the oxidisable organic C present in the sample. The organic carbon can then be estimated by measuring the remaining unreduced Potassium dichromate by back-titrating with ferrous sulphate or ammonium ferrous sulphate using diphenylamine or o-phenanthroline-ferrous complex as an indicator.

Chemical reaction is as follows:

 $2FeSO_4$ (NH₄)₂SO₄. $6H_2O$ + $K_2Cr_2O_7$ + $5H_2SO_4 \rightarrow 2$ $(NH_4)_2SO_4 + Fe_2(SO_4)_3 + K_2SO_4 + Cr_2(SO_4)_3 + O_2 + 17 H_2O$

Alternately the organic carbon can be calculated from the amount of chromic ion (Cr3+) formed, using a colorimetric procedure measuring absorbance at 588 nm (after Sims and Haby 1971).

Heerak Bhasm Titration: 0.1 g of Heerak Bhasm was weighed with electronic balance and transferred to 100 ml conical flask. To it 1 ml of 1N K₂Cr₂O₇ and 2 ml concentrated H₂SO₄ was added then kept it aside for 30 minutes. Then 10 ml distilled water, 1 ml of 85% pure Ortho phosphoric Acid (H₃PO₄) and 2-3 drops of 0.5 % di-phenyl amine indicator was added. And titrated it with 0.5 N ferrous ammonium sulphate and end point was noted. Colour changes from violet blue to dark green was observed.

Blank Titration: In 100 ml conical flask 1 ml of 1N K₂Cr₂O₇ and 2 ml concentrated H₂SO₄ was taken then kept it aside for 30 minutes. Then 10 ml distilled water, 1 ml of 85% pure Ortho phosphoric Acid (H₃PO₄) and 2-3 drops of 0.5 % di-phenyl amine indicator was added. And titrated it with 0.5 N ferrous ammonium sulphate and end point was noted. Colour changes from violet blue to dark green was observed.

2. SEM Analysis of Heerak Bhasm

The Electron Microscope is an essential component for scientific analysis of a variety of materials. Scanning Electron Microscope (SEM) comprises a powerful tool in studying (cell and molecular biology, anatomy, science) microbiology, pathology and forensic

biological specimens, food stuffs and several other areas of material sciences (electronics, metallurgy, polymer and surface science).

Morphological graphs of the Heerak Bhasm sample is provided by scanning electron microscopy (Digital Scanning Electron Microscope - JSM 6100 - JEOL) with a Link analytical system operating at 10 KV (acceleration voltage).

Scanning Electron Microscope (SEM) - Digital Scanning Electron Microscope - JSM 6100 (JEOL)

SEM facilitates the observation of very fine details (high resolution) of biological materials and good focus over a wide range of specimen surface (large depth of field). It also produces clear image of specimen ranging from object visible to the naked eye to a structure spanning few nanometers. Besides its use in studying soils, sedimentary particles and rock materials, it also helps to elucidate the architecture and evolution of microfossils.

Digital Scanning Electron Microscope - JSM 6100 (JEOL) is used with a digital image processor. It has a large specimen chamber that allows observation of the entire surface of a specimen upto 150 mm and a tilt of -5 to 90°. A special feature of this SEM is a cryostage attached to it to study the low melting point specimens. The image processing function permits image averaging and storage, filling of acquired still images and comparison of two/four images displayed simultaneously on the 12 inch CRT. This function makes it possible to observe specimens without causing damage to them.

Other features of this microscope are:

- Resolution: 4.0 nm at 8mm working distance
- Working distance: 6 to 48 mm •
- Accelerating Voltage: 0.3 to 30 KV
- Magnification: x 10 to x 300,000 •
- Image Recording: on 120 B&W Roll Film (100 • ASA) or 35mm B &W roll (25 ASA)
- Instant Print: an instant print is also possible on a Thermal Video Printer (8x10.5).

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3. Results and Discussion

1. Estimation of Organic Carbon in Heerak Bhasm by Walkley-Black method

% Organic Carbon = Blank Reading – Normality of ferrous ammonium sulphate X meq. Weight of C Weight of sample
X100

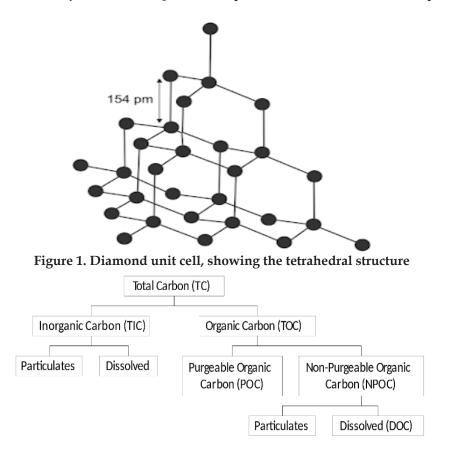
% Organic Carbon = $\frac{(6-2) \times 0.2 \times 0.03}{0.1} \times 100$

% Organic Carbon = = 6

Hence 6 % of Organic Carbon was found in sample of Heerak Bhasm.
% Organic Matter = % Organic Carbon X 1.724 (1.724 Van Bemmelen factor)
% Organic Matter = 6 × 1.724 = 10.34
Hence 13.34 % of Organic Matter was found in sample of Heerak Bhasm.

2. SEM Analysis of Heerak Bhasm

Figure 2 (A) – (E) shows Scanning Electron Microscope images of Heerak Bhasm. We can learn from Figure 2 (A) – (E) that the material mainly consisted of spherical particles with $1-3 \,\mu\text{m}$ in diameter, and has a smaller aggregated particle size. Although the majority of material consists of micrometer or grains, smaller particles with nanoscale are also present.



Heerak Bhasm Titration

Heerak Bhasm taken	Volume of 0.5 N ferrous ammonium sulphate required	Constant End Point
0.1 g	2.1 ml	
0.1 g	2.0 ml	2.0 ml
0.1 g	2.0 ml	

Blank Titration:

Н	eerak Bhasm taken	Volume of 0.5 N ferrous ammonium sulphate required	Constant End Point
	0.0 g	6.1 ml	
	0.0 g	6.0 ml	6.0 ml
	0.0 g	6.0 ml	

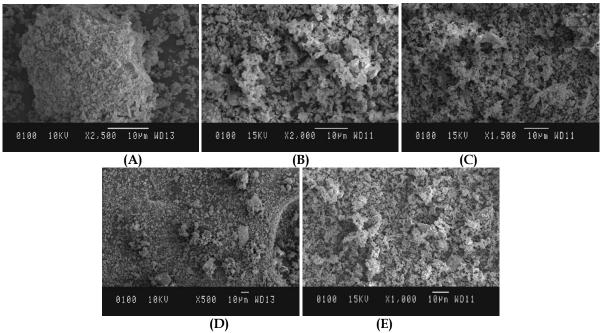


Figure 2. (A) - (E) Scanning Electron Microscope images of Heerak Bhasma

4. Conclusion

Heerak Bhasm, contains 6 % of Organic Carbon and 10.34 % of Organic Matter Scanning Electron Microscope image of Heerak Bhasma shows that the material mainly consisted of spherical particles with 1-3 μ m in diameter, and has a smaller aggregated particle size. Although the majority of material consists of micrometer or grains, smaller particles with nanoscale are also present.

Conflicts of interest: The author stated that no conflicts of interest.

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