PHYTOCHEMICAL ANALYSIS OF BLACK CUMIN OR MIRACLE SEED (*Nigela sativa*)

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Abstract

The seed is traditionally known in the Middle East as habbat al-Baraka (habbat =seed in Arabic; al Baraka = blessing) due to its powerful healing qualities and remarkable therapeutic potentials. The study was carried out to evaluate the Phytochemical analysis of black cumin or miracle seed (Nigela sativa). The Phytochemical screening revealed the presence of tannins, saponins, anthroquinohes, glycosides and terpenoids. The Phytochemical screenings showed that saponins, glycosides and anthroquinones were moderately present in te black cumin seed, steroids, reducing sugar, flavonoids were not detected in the black cumin sed. The results of Phytochemical analysis of black cumin indicate that the samples had good antioxidant activities. The result indicated showed the scientific basis for the traditional uses of black cumin seed or miracle seed (Nigela sativa).

1.0 Introduction

There is an increasing awareness of the therapeutic potential of many natural products and medicinal plants that are frequently considered far less toxic and free from side effects than synthetic drugs (Newman and Cragg, 2007). A vivid example of that is the black cumin (Nigella sativa), which is probably the most extensively studied medicinal plant in recent literature. The medicinal plant N. sativa is an annual spicy herb belonging to the family Ranunculaceae. The plant has long been recognized in the Indian subcontinent as an important element in traditional popular medicine but now it is widely cultivated in many other parts of the world including the Mediterranean regions, the Middle East, Far East and several other countries in Asia as well as in Europe (Chevallier, 1969). Historically, the black cumin has been considered a crop of a great medicinal importance in ancient Egyptian and Greek civilizations where it was used for treatment of various ailments such as headache, nasal congestion, toothache, and intestinal worms (Tembhurne *et al*, 2014).

The seed is traditionally known in the Middle East as habbat al-Baraka (habbat =seed in Arabic; al Baraka = blessing) due to its powerful healing qualities and remarkable therapeutic potentials. It became the most famous and popular medicinal plant among all Muslim communities because it was described by the Prophet Mohammed (PBUH) as a miraculous remedy. The most important active components of the volatile oils of the black seed are thymoquinone (TQ) and dithymoquinoline (DTQ), and both were reported to have anti-inflammatory and antitumor properties (Banerjee *et al.*, 2010).Those two active ingredients (TQ and DTQ) were also reported to have remarkable immunomodulatory and immunotherapeutic potentials (Swamy and Tan, 2000).

In addition to the traditional use the black cumin in popular folk medicine, it was also reported to have some beneficial effects in several aspects of poultry production such as improvement of body weight performance, food intake, feed conversion ratio, dressing percentage, carcass quality and visceral organ weight (Durrani *et al.,* 2007; Miraghaee *et al.,* 2011; Abu-Dieyeh and Abu-Darwish 2008). Moreover, the dietary use of the black cumin was also reported to increase egg production, egg mass and egg shell thickness in laying hens (Akhtar *et al.,* 2003; Nasir *et al.,* 2005).

The black cumin has been regarded since ancient history as a miraculous remedy for various ailments and disease conditions in human patients (Ahmad *et al.,* 2013; Tembhurne *et al.,* 2014). In fact, the seed has long been discovered in preserved crops found with Tutankhamun's tomb in Egypt and it was mentioned as a curative substance in the Holy Bible. The Greek physician Dioskorides used black cumin to treat headache, nasal congestion, toothache and intestinal parasites. Hippocrates regarded the black cumin as a valuable remedy for hepatic and digestive disorders. It has been recorded that the black cumin was prescribed by ancient Egyptian physicians to treat the above mentioned ailments in addition to obesity, back pain, hypertension, gastrointestinal problems and bronchial asthma.

The black cumin was also used in folk medicine for hundreds of years in the Middle East and Far East as a traditional medicine for a wide range of illnesses. It was frequently used as a diuretic agent and to promote menstruation and increase milk production (Goreja, *et al.,* 2003). It is worth mentioning that the black cumin has considerably received a special attention and interest in all Arab and other Muslim communities in which it was extensively used for treatment of various kinds of diseases. The absolute confidence on the curative potential of the black cumin by Muslim communities was due to the fact that the Prophet Muhammad (PBUH) described the black seed as "a remedy for every illness except death". Moreover, the famous Muslim philosopher and physician Avicenna (980-1037A.D.) stated in his famous legendary book "The Canon of Medicine" that the black cumin stimulates the body's energy and helps recovery from fatigue (Chevallier, 1996). He further claimed that the black cumin is good for inner purification of the body by reducing mucous and strengthening lungs, treating fever, coughs, colds, toothache, headache, skin diseases, infected wounds and intestinal parasites in addition to its ability to protect against poisonous bites and stings (Ahmed *et al.,* 2013).

2.0 Methodology

2.1 Sample Collection

The samples of black cumin seeds were purchased from International Market Lokoja, Kogi State, Nigeria and taken to the chemistry laboratory in Kogi State Polytechnic, Lokoja, Kogi State.

2.2 Sample Preparation

Black cumin seeds were purchased from Lokoja International Market Lokoja, Kogi State. The samples were washed with distilled water and thinly sliced, and then treated with chlorine concentrated solution. These samples were oven dried at temperature of 60OC for 24 hours. The samples were ground with a blender (Model No. 205) and stored in air-tight container. Each sample (100g) was weighed and extracted with methanol which was used for the analysis.

2.3 Phytochemical Screening Methods

Phytochemical screening was performed using standard procedures (Ayoola *et al.,* 2008).

2.3.1 Determination of Saponins in Black Cumin Seeds

0.4g of sample was added to 4ml of distilled water in test tubes and the solutions were shaken vigorously and observed for a stable persistent froth. The frothing was mixed with 3 drops of olive oil and shaken vigorously after which it was observed for the formation of an emulsion.

2.3.2 Determination of Triterpenoids in Black Cumin Seeds

0.4g of the sample was dissolved in 1ml of chloroform. 1ml of acetic anhydride was added, followed by the addition of 2ml of concentrated H_2SO_4. Formation of reddish violet colour indicates the presence of triterpenoids.

2.3.3 Determination of Tannins in Black Cumin Seeds

Two methods were used to test for tannins:

(a) To 10ml of freshly prepared 10% KOH in a beaker, 0.5g of extract was added and shaken to dissolve. A dirty precipitate observed indicated the presence of tannin.

(b) About 0.5g of each extract was boiled in 10ml of water in a test tube and then filtered. A few drops of 0.1% ferric chloride was added and the solution observed for brownish green or a blue-black colouration

2.3.4 Determination of Reducing Sugar (Fehling's test) in Black Cumin Seeds

0.5g of sample was dissolved in 5ml distilled water and filtered. The filtrate was hydrolysed with dilute HCl, neutralized with alkali (NaOH) and heated with Ferling`s A and B solutions. Formation of red precipitate indicated the presence of reducing sugars.

2.3.5 Determination of Anthraquinones in Black Cumin Seeds

0.5g of sample was boiled with 10ml of H_2SO_4 and filtered while hot. The filtrate was shaken with 5ml of chloroform, the chloroform layer was pipette into another test tube and 1ml of dilute ammonia was added. The resulting solution was observed for colour changes.

2.3.6 Determination of Steroids in Black Cumin Seeds

0.5g of extract was dissolved in 10ml of chloroform and equal volume of concentrated H_2SO_4 was added by the sides of the test tubes. Reddish upper layer and yellowish sulphuric acid layer with green fluorescence indicate the presence of steroids.

2.3.7 Determination of Cardiac Glycosides (Keller-Killiani Test) in Black Cumin Seeds

0.5g of extract dissolved, 5ml of water was added and 2ml of glacial acetic acid solution containing one drop of ferric chloride solution. This was underlayed with 1ml of concentrated H_2SO_4. A brown ring at the interface indicated the presence of a deoxysugar characteristics of cardenolides. A violet ring may appear below the brown ring while in the acetic acid layer a greenish ring may form just above the brown ring and gradually spread throughout this layer.

2.3.8 Determination of Flavonoids in Black Cumin Seeds

Two methods were used to test for flavonoids:

(a) A portion of the extracts were heated with 10ml of ethyl acetate over a steam bath for 3 minutes, the mixture was filtered and 4ml of the filtrate was shaken with 1ml of dilute ammonia solution. A yellow colouration indicated the presence of flavonoids.

(b) Dilute ammonia (5ml) was added to a portion of an aqueous filtrate of the extract. Then, concentrated sulphuric acid (1ml) was added. A yellow colouration indicated the

presence of flavonoids. Test for Alkaloids Extracts were dissolved individually in dilute HCl and filtered.

(a) Filtrates were treated with Mayer`s reagent (potassium mercuric iodide). Formation of a yellow coloured precipitate indicates the presence of alkaloids.

(b) Filtrate was treated with Dragendroff's reagent (solution of potassiumbismuthiodide). Formation of red precipitate indicates the presence of alkaloid. Filtrate was treated with Hager`s reagent (saturated picric acid solution). Presence of alkaloid is confirmed by the formation of yellow coloured precipitate.

2.4 Statistical Analysis

All determinations were carried out in triplicates. The results generated from the analysis were subjected to statistical package for social science (SPSS) version 16. Descriptive statistic was used to interpret the result. A P>value less than 0.05 was considered statistically significant.

3.0 Results and Discussion

3.1 Results

Table 1: Phytochemcial Composition of Black Cumin

Phytochemical compound	Ethanol extracts
Tannins	++
Saponins	+++
Anthraquinones	+++
Steroids	_
Glycosides	+++
Terpenoids	++
Reducing Sugar	_
Flavonoids	_

Key:
+++ = Highly present
++ = Moderately present
+ = Slightly
- = Not detected

3.2 Discussion

The results of the phytochemical screening of black cumin revealed the presence of Tannins, Saponins, Glycosides, Anthraquinones and Terpenoids. Their presence shows the medicinal properties of black cumin. Tannins for example are known to be made up of phenolic compounds Tannins possess homeostatic properties and therefore widely used as tropical application on superficial wounds and infections.

Black cumin is a widely observed plant which has a tremendous latent in terms of medicinal uses. Constantly, it contains different chemical constituents. This review comprises the pharmacological aspects of Black cumin as a rich source of phytonutrients. Numerous studies have shown that Black cumin exhibit numerous properties like Anti-flammatory Ayoola *et al.*, 2008).

In the present study which is found, one hand to be better for the extraction of tannins (Ahmed *et al.,* 2013) Black cumin seeds extract is a natural food additive which is used as food in

comparing the phytochemistry of the seeds to that of fruits for their possible use as natural food preservative in the food industries.

4.0 Conclusion

The results of phytochemical analysis of the Black cumin shown that ethanol seeds extracts of Black cumin revealed the presence of tannins, terpenoids, Saponins, glycosides & Anthraquinones, but no steroids, reducing sugar & flavonoids. And Black cumin seeds can be used for air freshener because the seed is made of eucalyptol which mostly contains terpene and cymene, which are organic aromatic compound.

Extract from the seeds of Black cumin seeds can be used in synergy other compounds as food additive. Several studies reports direct addition of aromatic plants essential oils and extracts to foodstuffs to exert an antimicrobial (Tembhurne *et al.,* 2014).

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