



## Research article

**Total phenolic, flavonoid content and anti-oxidant potential of *Phaseolus vulgaris***Amit Gupta<sup>1\*</sup>, Simran Srivastava<sup>1</sup>, Arsh Singh<sup>1</sup>, Priya Arya<sup>1</sup>, Atal Bihari Bajpai<sup>2</sup>, Vijay Kumar<sup>3</sup><sup>1</sup> Department of Microbiology and Biotechnology, Graphic Era Deemed to be University, Dehradun.<sup>2</sup> Department of Botany, D.B.S. PG College, Dehradun.<sup>3</sup> Graphic Era Hill University, Dehradun.**Corresponding author:** Amit Gupta ✉ dr.amitgupta.bt@geu.ac.in, **Orcid Id:** <https://orcid.org/0000-0002-2003-8618>

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**ABSTRACT**

Vegetables are parts of plant having several chemical families which belong to the category of antioxidants. In literature, these vegetables and fruits contain a high amounts of antioxidant molecules which may directly benefit to human healthcare. The objective of our study is to determine the antioxidant potential of seed containing fibres aqueous extract and analyzed its phenolic and flavonoid content. For these studies, we procured *Phaseolus vulgaris* (green beans) from the local market of Dehradun and measured its antioxidant activity including phenolic and flavonoid content. The results of these studies showed that seed containing fibres aqueous extract showed higher antioxidant activity at higher doses and also showed enhancement in phenolic and flavonoid content. These studies may indicate that vegetables especially *Phaseolus vulgaris* contain a significant number of antioxidants which is required and acceptable for human healthcare.

**Keywords:** *Phaseolus vulgaris*, Vegetables, Antioxidant, Phenolic, Flavonoids.**INTRODUCTION**

In literature, a plant-based diet with an elevated amount of nutrient rich food having fruits and vegetables may bring down the probability of various infectious diseases. Understanding the complexity of diet especially in chronic diseases is one of the major challenges. In literature, more than twenty thousand bioactive compounds are reported in food. Due to its complexity in food, it is important to understand the role of each bioactive compound and assessed its activity in animal model studies including humans [1,2]. In general, some bioactive compounds are added or an individual alone may show some synergistic effect and showed a positive impact on human healthcare. As per the literature, these bioactive molecules are identified and isolated from plants that collectively formed as phytochemicals and screened for its immunobiological activity.

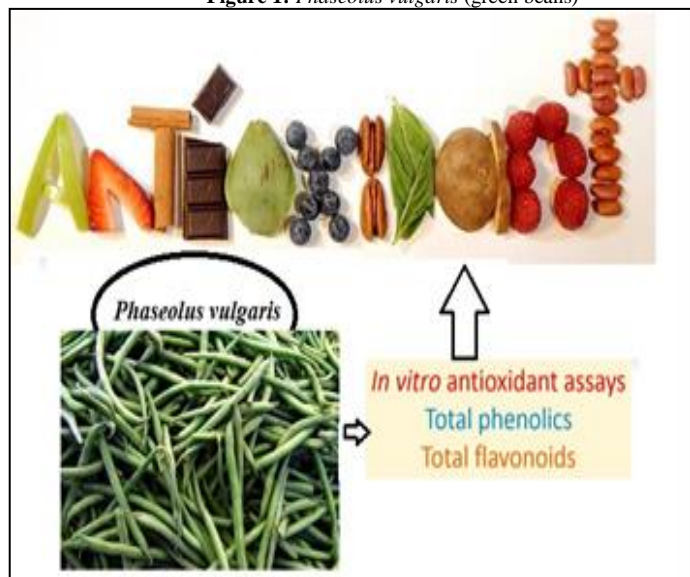
In plants, plant derived chemicals (phytochemicals) are redox active molecules and are therefore called antioxidants. Each antioxidant-containing molecule has a unique role and performs a distinct function. Antioxidant does not specifically relate to a compound, but rather to a category of many diverse compounds that possess antioxidant properties.

Vegetables are plant sections or fragments that are consumed as food by people and other animals. The earliest definition is still frequently used to point out all edible plant material, including the stems, roots, flowers, leaves, and seeds, when applied to plants as a whole [1-3].

They are essential to human nutrition as they accommodate water- or fat-soluble vitamins, essential minerals, plant derived

chemicals, and dietary fiber, vegetables are pivotal for human healthcare. In other words, these antioxidants-based vitamins (especially A, C, and E) and dietary fiber levels, in precise, play a pivotal character in maintaining or prolonging human health.

**Figure 1:** *Phaseolus vulgaris* (green beans)



Most of them are substantial and full but low in fat and calories. The amount of food generated from plants that cannot be entirely broken down by digestive enzymes in humans is known as dietary fiber or roughage. We must ensure that consume adequate amounts of fiber because it supports good digestion, body composition, blood sugar regulation, and many other things. It has even been connected to increased longevity and a lower chance of developing cancer<sup>[3,4]</sup>.

*Phaseolus vulgaris*, popularly familiar as the common bean or French bean, is an herbaceous annual plant that is reported and grown around the world for its palatable dry seeds or unripe fruit. This is one such vegetable with high fiber content (both commonly called green beans)<sup>[5,6]</sup>. Green beans have a lot of fiber, which is a crucial nutrient for a variety of reasons. Each cup of fresh green beans contains 2.7 g of fiber. By reducing our levels of LDL cholesterol (bad cholesterol), the soluble fiber in particular may assist to boost the health of your heart. Green beans' fiber keeps our digestive system functioning normally. By reducing blood pressure and limiting inflammation, it may also benefit heart health. Green beans, like many other vegetables, are a nutritious complement to practically any diet since they are a low-calorie, low-fat food. They also include a lot of healthy vitamins, antioxidants, and minerals without consuming a lot of calories. Their combination makes them the perfect food for a diet that encourages a healthy weight<sup>[7,8]</sup>. In this paper, we focused on fiber content of *Phaseolus vulgaris* and determined its antioxidant and anti-inflammatory potential.

## MATERIALS AND METHODS

### Procurement of chemicals and reagents

Chemicals and reagents were obtained from HI media, SRL and CDH, India.

### Sample Collection and preparation

Common beans (*Phaseolus vulgaris*) seeds were collected from the local market of Dehradun (authenticated by Dr. Atal Bihari Bajpai, Botanist with number PGDBS1) and these seeds were macerated in mortar and pestle using phosphate buffered saline (PBS). Briefly, the seed samples (5 g) were collected, macerated and dissolved in PBS (40 ml) and then incubated (37 °C, 60 min) in the water bath. The seed sample residues were filtered (Whatman No. 4), and prepare serial dilutions for analyzing its phytochemical (qualitative based) using standard methods<sup>[9-11]</sup>, phenolic and flavonoid content including antioxidant activity.

### Estimation of Phenolic and flavonoid content

Aqueous extract from seed containing fibers was prepared and estimated its total phenolic compounds were determined using the Folin-Ciocalteu method. Serial dilutions of seed containing fibers were placed in a test tube (200 µl) mixed with Folin-Ciocalteu reagent (250 µl) and sodium carbonate (20%, 750 µl) and distilled water to reach the final volume. For these studies, gallic acid was pre-owned as a standard for quantifying the phenolic amalgam. The results of these studies were expressed as µg gallic acid equivalent (GAE)/g and experimental analysis was performed in triplicates and measured its absorbance at 570 nm<sup>[9]</sup>.

For measuring the total flavonoid content in the aqueous extract from seed containing fibers was measured using aluminum chloride method. In this experiment, seed containing fibers diluted extract (500 µl) was added in a test tube having distilled water (2 ml) and sodium nitrite (150 µl, 5% NaNO<sub>2</sub>). After 5 min, AlCl<sub>3</sub>·6H<sub>2</sub>O (10%, 150 µl) was added and mixed with 1 M NaOH (1 ml) and distilled water (1.2 ml). The absorbance of these samples was measured at 570 nm using water as blank. Quercetin is utilized as a standard for the computation of total flavonoid content. Results of these studies were indicated as µg Quercetin equivalent (CE)/g. This study was performed and analyzed in triplicates<sup>[10]</sup>.

### Antioxidant Activity

The free radical scavenging ability of the aqueous extract from seed containing fibers was tested by DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay. The samples (seed containing fibers) were proceeded with the steady DPPH radical in an ethanol solution. The reaction concoction incorporates seed containing fibres aqueous extract (500 µl), absolute ethanol (3000 µl) and DPPH radical solution (300 µl, 0.5 mM in ethanol). In general, DPPH responds with a compound as an antioxidant in aqueous extract of seed containing fibres, which can release hydrogen, it is reduced. The changes in color (from deep violet to light yellow) were

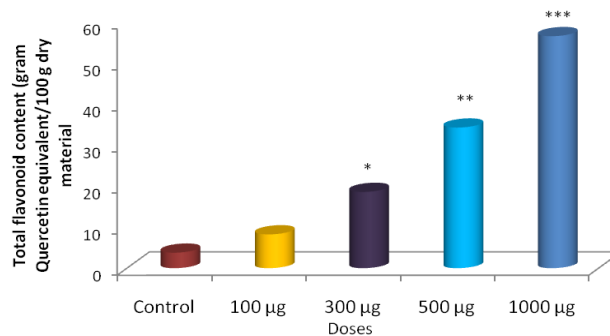
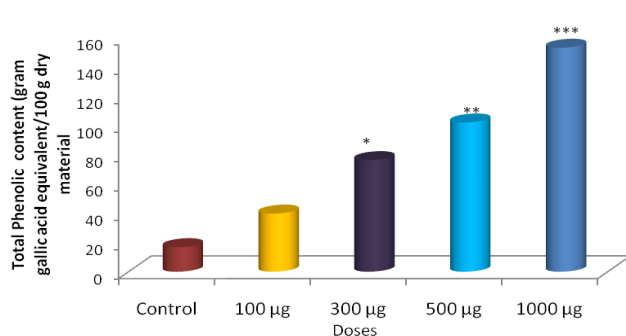
read and measured its absorbance at 517 nm after 100 min of reaction using a UV-VIS spectrophotometer (Shimadzu, Model no. UV-1900). In this study, a combination of ethanol (3300 µl) and seed containing fibres aqueous extract (500 µl) was set out as blank [11]. The control samples were put together by a combination of ethanol (3500 µl) and DPPH radical solution (300 µl) and measured its scavenging activity percentage (AA%) as shown below-

$$\% \text{ DPPH radical scavenging activity} = \{(A_0 - A_1)/A_0\} \times 100$$

### Statistical analysis

Values were indicated in the form of Mean  $\pm$  S.E. and analyzed its statistical data using a one-way ANOVA test.

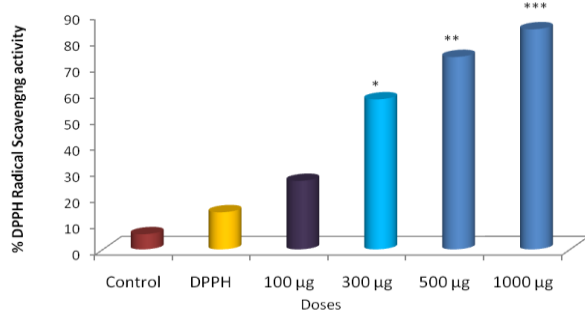
**Figure 2:** Estimation of phenolic and flavonoid content from *Phaseolus vulgaris*. Statistical analysis was performed,  $P < 0.05$ ;  $P < 0.01$  and  $***P < 0.001$



### Antioxidant Activity

The results of seed containing fibres aqueous extract for measuring its antioxidant activity using DPPH as shown in Figure 3. The assay is based on the evaluation of the antioxidants' ability to scavenge it. By obtaining a hydrogen atom from antioxidants and converting it to the equivalent hydrazine, the odd electron of the nitrogen atom in DPPH is lowered. From these studies, it may have been revealed that seed containing fibres aqueous extract showed antioxidant activity at higher doses.

**Figure 3:** Estimation its antioxidant activity of *Phaseolus vulgaris*. Statistical analysis was performed,  $P < 0.05$ ;  $P < 0.01$  and  $***P < 0.001$



### DISCUSSION

Over the past year's fruits and vegetables have shown a tremendous performance in the anti-oxidant activity. Various antioxidant derived foods including tomatoes, green leafy vegetables, nuts, and so on are reported. One such vegetable that is green beans also known as *P. vulgaris* acted as an immunomodulator. In literature, an antioxidant is a chemical that, in small amounts, delays or stops a substrate from oxidizing. Chemical

### RESULTS

#### Phenolic and flavonoid content

The results of seed containing fibres aqueous extract for measuring the phenolic and flavonoid content as shown in Figure 2. These studies revealed that aqueous extract at higher concentrations revealed a higher amount of phenolic and flavonoid content.

#### Phytochemical analysis

This immunopharmacological activity of seed extract is mainly due to the presence of phytochemicals (flavonoids, saponins and alkaloids) that are present in seed containing fibres aqueous extract.

processes such as hydrogen atom transfer (HAT), single electron transfer (SET), and the capacity to bind transition metals are all used by antioxidant chemicals to exert their effects. Understanding the biological significance of antioxidants, their potential applications, their manufacturing via chemical synthesis or biotechnological techniques or the standardized assessment of antioxidant activity is

all reasons why understanding antioxidant processes are important. Antioxidant compounds often react either through several processes or through a single, predominant one. Any material that hampers or intercepts the oxidative devastation to a desired molecule can be classified as an antioxidant. The potential of an antioxidant to catch out free radicals is its most predominant hallmark [12, 13]. Antioxidant substances such as phenolic acids, polyphenols, and flavonoids scavenge free radicals including peroxide, hydro peroxide, and lipid peroxy, obstructing the oxidative track that generates degenerative diseases. In this study, we worked on aqueous extract from seed containing fibres and showed enhancement in phenolic and flavonoid content. Both flavonoids and many other phenolic components have been outlined and disclosed its effective antioxidant activity including anticancer, antimicrobial, cardio protective agents, anti-inflammation, immuno-stimulant and skin protection from UV radiation and engrossing molecules for biopharmaceutical and medical relevance.

The complicated process of oxidative load in biological systems is described by an imbalance between the body's capacity to

remove free radicals (FR) through the employment of exogenous and endogenous antioxidants. Reactive oxygen species (ROS), including hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) and the superoxide radical anion, among others, operate as catalysts for a wide range of events that occur throughout metabolic processes. When there is an excess of ROS in a biological system, it can cause a variety of pathologies, such as the development of cancer and cardiovascular illnesses<sup>[14]</sup>. Antioxidant defenses in biological systems regulate enzymatic and nonenzymatic alterations that result in ROS inactivation. Enzymes like catalase, glutathione peroxidase, and superoxide dismutase, as well as non-enzymatic substances like bilirubin and albumin, are examples of endogenous antioxidants. The endogenous antioxidant system is damaged when an organism is exposed to a lot of ROS, which prevents it from providing the organism with full protection. The body can employ exogenous antioxidants obtained from food, dietary supplements, or medications to make up for this antioxidant deficiency. The phenolic molecules carotenoids, vitamins C, and certain minerals like selenium and zinc are among the most significant exogenous antioxidants. Because the pi electrons of the aromatic systems existing in the molecule can make up for the absence of one electron, the 1,1-diphenyl-2-picrylhydrazyl radical (DPPH) is classified as a stable free radical. Using DPPH in solution, we assessed the antioxidant activity of particular compounds or extracts. The reduced forms of DPPH-H are created when a solution of DPPH comes in contact with a material that can provide a hydrogen atom or with some other radical, which results in the loss of color and a reduction in absorbance. As a result, the decrease in DPPH offers a measure to determine how well the test drug can capture radicals. In short, herbal plants have been recognized and investigated as satisfactory and acceptable antioxidants<sup>[15]</sup>. Phenolic components and flavonoids are the secondary plant metabolite that contains the aromatic ring having at least one hydroxyl group. Additionally, phenolic compounds (n >80,000) have been identified as naturally materialize substances in plants.

## CONCLUSION

In fruits like watermelon, strawberries, and grapefruit as well as vegetables like broccoli, cauliflower, tomatoes, boiled spinach, pumpkin, etc., there is naturally occurring antioxidant activity. The health of people may be endangered in a number of ways by excessive antioxidant supplementation.

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## Declaration of competing interest

Authors declare that they have no conflicts of interest to disclose.

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