

To Study The Evaluation Of Pea Powder By Comparing Two Different Types Of Extraction.

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Abstract:

Pea powder, derived from the common garden pea (*Pisum sativum*), has gained considerable attention in recent years due to its remarkable antioxidant properties. This abstract provides a concise overview of the research conducted on the antioxidant potential of pea powder and its implications for human health. The antioxidant activity of pea powder can be attributed to its rich composition of bioactive compounds, including phenolic compounds, flavonoids, and carotenoids. These phytochemicals play a crucial role in neutralizing harmful free radicals, thereby preventing oxidative stress and damage to cellular components. Several studies have reported the potent antioxidant capacity of pea powder, as evidenced by its ability to scavenge free radicals, inhibit lipid peroxidation, and increase the activity of antioxidant enzymes. Moreover, the unique combination of antioxidants present in pea powder exhibits synergistic effects, enhancing its overall antioxidant efficacy. Furthermore, the antioxidant properties of pea powder have been linked to various health benefits. Analyse the nutritional content of pea powder extracted by each method. This includes evaluating macronutrients (carbohydrates, proteins, and fats) and micronutrients (vitamins and minerals). By comparing the nutritional profiles, you can determine if any extraction method leads to a higher retention of essential nutrients.

(**Keyword:** Study of Pea powder and extraction, Determine Antioxidant)

Introduction:

Pea plants, scientifically known as *Pisum sativum*, are one of the most well-known and widely cultivated garden plants. They belong to the Fabaceae family, which also includes legumes such as beans, lentils, and peanuts. Pea plants are native to the Mediterranean region and have been cultivated for thousands of years. Pea plants are annual, herbaceous plants that grow as climbing vines or bushy shrubs, depending on the variety. They typically reach a height of 1 to 2 meters (3 to 6 feet) and have compound leaves with multiple leaflets. The leaflets are usually smooth and oval-shaped, and they provide an important role in the process of photosynthesis. Pea plants are known for their distinctive flowers, which are typically white, pink, or purple in colour. The flowers are pea-like in shape and are arranged in clusters on long stalks. Each flower has both male and female reproductive organs and is capable of self-pollination. Pea flowers are not only visually appealing but also attract pollinators like bees and butterflies. The flowers give way to seed pods, known as peas, which are the edible part of the plant. Peas are typically green and round, although there are also yellow and purple varieties available. The peas inside the pods can be consumed fresh, frozen, or dried for later use. Pea plants are highly valued for their nutritional content, as they are rich in vitamins, minerals, fiber, and protein.

Pea powder, derived from dried and ground peas (*Pisum sativum*), has gained considerable popularity as a versatile and nutritious ingredient in the food industry. It offers numerous advantages, such as its high nutritional content, sustainability, and versatility in various culinary applications. However, like any food product, pea powder also presents certain disadvantages and considerations. This introduction aims to provide an overview of the advantages, disadvantages, and properties of pea powder.

Material and Method:

1) Selection and Preparation of Pea powder: The pea was shade dried and crushed using grinder to obtain coarse powder which was further used for extraction.

2) **Materials:** Pea powder, also known as pea protein powder, is a powdered form of pea protein derived from yellow peas (*Pisum sativum*). It has gained popularity as a plant-based protein source due to its nutritional benefits and versatility in various applications.

Method:

Extraction of Pea powder:

A) Soxhlet Extraction Method:

A 25 g of crushed pea powder was extracted using ethanol (125ml) and a Soxhlet extractor for 3 hr at then, the mixture of solvent-oil was filtered through a No.1 paper filter (Whatman). The extract was transferred into a round flask and solvent was evaporated using rotary evaporator.

B) Cold Maceration Method:

The extraction procedure adopted was cold maceration using 80% methanol as the solvent. The extraction mixture was prepared by dissolving weighted amount of the powdered peas in 80% methanol in an air-tight glass container in ratio 2:10 (W/V). The mixture was allowed to soak for 7 days. The mixture was filtered with a funnel plugged with cotton wool and the sample filtrate was collected into a clean glass beaker and the residue was discarded. The filtrate obtained was concentrated on water bath to give a paste.

A. General Tests For Preliminary Phytochemicals Screening:

- Carbohydrates:- Molisch's test:- Mix 1 ml reagent in 2ml of test solution. Add 1 ml of concentrated sulphuric acid. Red to violet ring depending on the amount of sugar appears at the junctions of the two liquids.
- Iodine test: - Mix 0.5ml of iodine solution with 1ml of the test solution. Starch gives deep blue colour.
- Fehling's test: - Mix 1ml of Fehling's solution 'A' with 1ml of Fehling's solution 'B' and 1ml of test solution. Boil. Yellow to red precipitate.
- Benedict's test: - Mix 2ml of Benedict's reagent with 2ml test solution. Boil in a water bath. Formation of red, yellow or green colour / precipitate depending on the sugar concentration.
- Non-reducing sugars: - Mix 5ml of test solution with 5 drops of concentrated HCL. Boil. Add 10% sodium hydroxide solutions. Perform Benedict's test. Red or yellow

B) PROTEINS:-

- Million's test:- Mix 2ml test solution with 2ml Million's reagent. Boil. Red colour.
- Xanthoprotein test: - Mix 2ml test solution with 2ml conc.H₂SO₄. White precipitate
- Lead acetate test: - Mix 2ml test solution with 2ml 40% HgCl₂ or 5% ammonium sulphate. Black to brown colour

C) AMINO ACIDS:-

- Ninhydrin test: - Mix 2ml test solution with 1 ml 5% ninhydrin solution. Boil for 5 min.in water bath. Blue-purple colour.
- Tyrosine test: - Mix 2ml test solution with 1ml Million's test reagent boil. Dark red colour.
- Cysteine test: - Mix 3ml test solution with 0.5 ml 40% NaOH. Add 10% lead acetate . Boil. Black precipitate.

D) ALKALOIDS:-

- Dragendorff's reagent: - Mix 2 ml of reagent with 2 ml filtrate of plant drug extract. Reddish brown precipitate.
- Modified Dragendorff's reagent: - Mix 2 ml of reagent with 2 ml filtrate of plant drug extract. Precipitation.

E) GLYCOSIDES:-

- General Test: - Solution A: Extract sample powder with alcohol or water and then add Fehling Solution. Solution B: to the water or alcoholic extract add sulphuric acid and then add Fehling Solution. If solution B has darker colour than solution A or if sugar content is high in solution B than solution A indicates presence of glycosides.

F) CARDIAC GLYCOSIDES: -

- Keller-kiliani test for digitoxin sugar: To the alcoholic extract of sample add 5 ml of water and 0.5 ml of strong solution of lead acetate. Filter and treat the clear filtrate with equal volume of chloroform and evaporate to yield dry residue. Add glacial acetic acid, 0.5 ml of ferric chloride solution and 2 ml of concentrated sulphuric acid. Initially the red brown layer changes to blue green.

G) FLAVONOIDS:-

- Shinoda test:- Add magnesium powder and a few drops of concentrated HCl or H₂SO₄ to 2 ml of sample solution
Flavones, flavanols and xanthenes: Orange, pink, red, purple.
- Flavones and flavanols: weak pink to magenta colours or no colour at all.
- Sulphuric acid:- Add H₂SO₄ in sample. Flavones and flavanols Deep yellow colour. Chalcones and aurones Red or red-bluish.
- Flavones: Orange red colours
- Lead acetate. Mix test solution with lead acetate yellow precipitate.

H) TANNINS:-

Ferric chloride (5%) Mix 2 ml of test solution with ferric chloride solution Blue, blue-black, or blue. green colour reaction.

Lead acetate test : Mix test solution with lead acetate solution White precipitate Dilute iodine test Mix test solution with dilute iodine solution Red colour

I) STEROIDS OR TRITERPENOIDS: -

- Salkowski reaction:- Dissolve 1-2 mg of the sample in 1 ml of CHCl₃ and add 1ml concentrated H₂SO₄. Chloroform layer shows red colour and acid layer shows green fluorescence.

J) SAPONINS: -

Foam test:- Shake aqueous solution of a saponin containing sample producing foam, which is stable for 15 min or more. Foam lasts for more than 15 seconds.

K) ANTHRAQUINONES: -

Bontrager's test:- Take little quantity of a aqueous solution of sample; add H₂SO₄, then add CCl₄ or ether. Separate the organic layer and shake with dilute ammonia Rose pink colour of ammonia layer.

L) COUMARIN GLYCOSIDES: -

- Odour test:- Take the odour of powder or extract Aromatic smell Alkali test:- Mix the test solution with alkali Blue green fluorescence

M) MUCILAGE:-

- Ruthenium red test:- Treat the powder with ruthenium red. Red colour.
- Swelling test:- Dissolve the powder in water. Powder swells.

N) FATTY OIL:-

- Filter paper test Press the powder between filter paper. Permanent oily spot.

O) ESSENTIAL OIL:-

- Sudan Red 3 test: - Treat the test solution with Sudan Red 3. Red colour.

Result and Discussion:

1. Microscopic Evaluation:- In this shows Epidermis , Endosperm, Testa, Sclerenchyma



Figure 01. Pea powder microscopy.

2. Physical Appearance:

SR.NO.	TEST	OBSERVATION
1	Colour	Light green
2	Odour	Odourless
3	Texture	Smooth
4.	Taste	Bitter

Table no.01- Physical Appearance pea powder

➤ **Solubility:-**

- A) Water:- Insoluble in water.
- B) Methanol:- Slightly soluble in methanol.
- C) Boiling water:- Completely soluble in boiling water

POWDER TEST:-

A) CARBOHYDRATES:-

Sr. No.	Test	Observation	Test
1.	Molisch test:-	Red to violet ring at junction of two liquids	Absent
2.	Iodine test:-	Deep blue colour	Present
3.	Fehling’s test:-	Yellow to red ppt	Present
4.	Benedict’s test:-	Red Yellow, green colour ppt.	Present
5.	Non-reducing sugars:-	Red/Yellow colour	Present

B) PROTEINS:-

Sr. No.	Test	Observation	Test
1.	Million's Test	Red colour	Present
2.	Xanthoprotein	White ppt	Absent
3.	Lead acetate test	Black to brown colour	Absent

C) AMINO ACIDS:-

Sr. No	Test	Observation	Result
1.	Ninhydrin Test	Blue-purple colour	Present

D) ALKALOIDS:-

Sr. No	Test	Observation	Result
1.	Drangendroff's test:-	Reddish brown ppt	Absent
2.	Modified drangendroff's test:-	Precipitation	Present

E) FLAVONOIDS:-

Sr. No	Test	Observation	Result
1.	Shinoda test	No colour at all	Flavanones and flavanols present
2.	Lead acetate test	Yellow ppt	Present
3.	Sulphuric acid	Yellow red, orange colour	Present

F) CARDIAC GLYCOSIDES:-

Sr. No	Test	Observation	Result
1.	Keller-kill ani test	Reddish brown colour acquiring bluish green.	Present

G) TANNINS:-

Sr. No	Test	Observation	Result
1.	Ferric chloride 5%	Blue, blue-black or blue green colour	Absent
2.	Lead acetate test	White ppt	Absent
3.	Dilute iodine test	Red colour	Absent

H) SAPONINS:-

Sr. No	Test	Observation	Result
1.	Ferric chloride 5%	Blue, blue-black or blue green colour	Absent
2.	Lead acetate test	White ppt	Absent
3.	Dilute iodine test	Red colour	Absent

I) GLYCOSIDES:-

Sr. No	i. Test	Observation	Result
1.	General Test	Solution B has darker colour than A	Absent

J) STEROIDS OR TRITERPENOIDS:-

Sr. No	Test	Observation	Result
1.	Salkowski reaction	Acid layer show green fluorescence and chloroform layer shows red colour.	Present

K) ANTHRAQUINONES:-

Sr. No	Test	Observation	Result
1.	Bontrager's Test	Rose pink colour of ammonia layer	Absent

L) COUMARIN GLYCOSIDES:-

Sr. No	Test	Observation	Result
1.	Odour Test	Aromatic smell	Absent
2.	Alkali Test	Blue green fluorescence	Absent

M) MUCILAGE:-

Sr. No	i. Test	Observation	Result
1.	Ruthenium red test	Red colour	Present
2.	Swelling test	Powder swells	Present

N) FATTY OILS:-

Sr. No	Test	Observation	Result
1.	Filter paper test	Permanent oily spot	Absent

O) ESSENTIAL OIL:-

Sr. No	Test	Observation	Result
1.	Sudan red 3 test	Red colour	Absent

➤ **Extraction Test:-1.Cold Maceration Test:-**

Sr. No.	Test	Observation	Inference
1.	Test for tannins	Blue black or blue green ppt	Absent
2.	Test for steroids and Triterpenoids	Red colour and Yellow colour	Present
3.	Test for glycosides	Brick red ppt	Absent
4.	Test For Anthraquinones	Pink, red or violet colour	Absent
5.	Test For saponins	Warming	Present
6.	Test for phenols	Deep bluish green ppt	Absent
7.	Test for cardenolide	Turbid brown colour ppt.	Absent
8.	Test for terpenoids	Bluish green Ppt.	
9.	Test for carbohydrate(Fehling's test for Reducing Sugar)	Brick red ppt.	Present
10.	Test for flavonoids	Yellow colour turned colourless	Present
11.	Test for cardiac Glycosides (keller-killani Test)	Reddish burn layer bluish green	Present
12.	Test for the phlobatannin	Reddish ppt	Absent
13.	Test for Balsams	Park green colour	Absent
14.	Test For Volatile oils	White Ppt	Absent

Extraction Test:-

A) CARBOHYDRATES:-

Sr.no.	Test	Observation	Result
1.	Molisch test	Red to violet ring at junction of two liquids	Absent
2.	Iodine test	Deep blue colour	Absent
3.	Fehling's test	Yellow to red ppt	Present
4.	Benedict's test	Red Yellow, green colour ppt.	Present
5.	Non-reducing sugars	Red/Yellow colour	Present

B) PROTEINS:-

Sr. No.	Test	Observation	Test
1.	Million's Test	Red colour	Absent
2.	Xanthoprotein	White ppt	Absent
3.	Lead acetate test	Black to brown colour	Absent

C) AMINO ACIDS:-

Sr. No.	Test	Observation	Test
1.	Ninhydrin Test	Blue-purple colour	Absent
2.	Tyrosine Test	Dark red colour	Absent
3.	Cysteine Test	Black Ppt	Absent

D) ALKALOIDS:-

Sr. No.	Test	Observation	Result
1.	Sample + 1ml Picric acid	Yellow ppt	Present
2.	Drongendroff's test	Reddish brown ppt	Present

E) FLAVONOIDS:-

Sr. No.	Test	Observation	Result
1.	Sample + NaOH yellow colour + dilute acid	colourless mixture	Present

F) CARDIAC GLYCOSIDES:-

Sr. No.	Test	Observation	Result
1.	Keller-kill ani test	Reddish brown colour bluish green.	Present

G) TANNINS:-

Sr. No.	Test	Observation	Result
1.	Ferric chloride 5%	Blue, blue-black or blue green colour	Absent

H) SAPONINS:-

Sr. No.	Test	Observation	Result
1.	Foam test	Foam last for 15 seconds	Present

I) GLYCOSIDES:-

Sr.No.	Test	Observation	Result
1.	General Test	Solution B has darker colour than A	Absent

J) STEROIDS OR TRITERPENOID:-

Sr. No.	Test	Observation	Result
1.	Salkowski reaction	Acid layer show green fluorescence and chloroform layer shows Red color	Present

K) ANTHRAQUINONES:-

Sr. No.	Test	Observation	Result
1.	Bontrager's Test	Rose pink color of ammonia Layer	Absent

L) COUMARIN GLYCOSIDES:-

Sr. No.	Test	Observation	Result
1.	Odour Test	Aromatic smell	Present

Conclusion:

The pea would be useful as an antioxidant and free radical scavenging agent, can serve as important tool in treatment of many diseases mediated by reactive oxygen species. Thus, it can be concluded that Ethanolic extract and water extract of Pea powder are easily available natural antioxidants with consequent health benefits. The results of the present study indicate that, the Pea powder and its extraction evaluated and can be used as a potent source of natural antioxidants

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