



# Study of the Presence of Pesticide and Insecticide Residues in Fruits and Vegetables using Lassaigne's Nitrogen Test

## *A Qualitative Analytical Investigation*

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### Abstract

Both fruits and vegetables are vital constituents of the human diet owing to the immense nutritional content that they contain. However, the use of pesticides and insecticides in modern-day agricultural activities poses food safety and health concerns. In the current study, the objective is to find out if there are any nitrogenous pesticide residues present in the samples of fruits and vegetables by conducting Lassaigne's test. Twelve samples in total were chosen from the fruit and vegetable section and tested for the presence of any nitrogenous compounds. Except for guava and carrot, all other fruits and vegetables tested positive for the presence of nitrogen.

**Key words:** pesticides, insecticides, Lassaigne's test, health risks, fruits, vegetables

### 1. INTRODUCTION

Fruits and vegetables are very important for keeping people healthy because they are full of vitamins, minerals, and antioxidants. Eating them is strongly linked to a lower risk of chronic diseases like cancer, heart disease, and obesity. But more and more, farmers use pesticides and insecticides to boost crop yields and keep pests and diseases at bay.

In the agricultural sector, pesticides can be defined as chemical or biological materials that are used in an effort to prevent or eliminate pests and include classes of pesticides known as; insecticides, herbicides, fungicides and rodenticides., Due to their effectiveness in increasing agricultural productivity, many of these types of pesticides do not break down in the environment and remain in the environment for long periods of time. After crops are harvested, pesticide residues can remain on and even inside fruits and vegetables. Once pesticides have entered the food chain (e.g., through the consumption of animal products), they can accumulate in animals that are consumed by humans. This accumulation of pesticide residues can lead to a number of different human health concerns, which can include neurological disorders, reproductive toxicity, immune suppression, and tumor formation/cancer. For that reason, monitoring pesticide residues on food items is extremely important to ensure the health of the general public.

## 1.1 Objectives

The specific objectives of this study were:

1. To detect the presence of nitrogen-containing insecticide and pesticide residues in commonly consumed fruits and vegetables using Lassaigne's qualitative test.
2. To assess the potential health hazards posed by pesticide residues in food commodities.
3. To review and propose strategies for reducing pesticide content in fruits and vegetables, including eco-friendly agricultural alternatives.

## 1.2 Classification of Pesticides

The pesticides are categorized depending on the type of pest organism that they target. There are four main groups of pesticides involved in this study:

**Herbicides:** These are the chemicals used to remove unwanted plants. They target specific weeds, hence leaving the required plant untouched by interfering with its growth hormones. An excellent example is 2,4-Dichlorophenoxyacetic acid (2,4-D).

**Insecticides:** Chemical formulations that help to kill, repel, or suppress the growth of insects. They act by interfering with their neurological functions, damaging their external structure, and inhibiting biological functions. Some insecticides include monocrotophos and methyl parathion – all are nitrogenous organophosphorus.

**Rodenticides:** Chemicals used in controlling pests that feed on stored grains, causing spoilage. Also used to curb the transfer of zoonotic diseases. Examples of rodenticides include aluminum phosphide and zinc phosphide.

**Fungicides:** These biocidal substances are used to eliminate pathogens growing in plants. For instance, copper oxychloride (50% WG).

## 2. MATERIALS AND METHODS

### 2.1 Sample Collection

Twelve fresh produce samples-six fruits and six vegetables were collected from local retail markets in the study region. The samples comprised apples, grapes, strawberries, oranges, guavas, and pomegranates (fruits), and tomatoes, spinach, beans, brinjals (eggplants), potatoes, and carrots (vegetables). Samples were selected to represent a diversity of produce types, cultivation practices, and typical consumer purchase patterns. All samples were used within 24 hours of purchase and stored at ambient temperature to prevent microbial degradation prior to analysis.

### 2.2 Apparatus and Equipment

The following laboratory apparatus were employed in this study:

- Mortar and pestle (for mechanical homogenization of samples)
- Glass beakers (250 mL), glass rods, and conical flasks
- Borosilicate fusion tubes and tripod stands
- Buchner funnel with Whatman No. 1 filter paper
- Porcelain China dishes and a thermostatically controlled water bath (80°C)
- Analytical balance (precision  $\pm 0.001$  g)
- Test tubes and test-tube racks

### 2.3 Chemicals and Reagents

- Absolute alcohol (ethanol, 99.9%) — for extraction
- Freshly cut sodium metal — for Lassaigne fusion
- Ferrous sulphate crystals ( $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ ) — freshly prepared aqueous solution
- Ferric chloride solution ( $\text{FeCl}_3$ , 5% w/v)
- Dilute hydrochloric acid ( $\text{HCl}$ , 10% v/v)
- Double-distilled water (DDW)

### 2.4 Chemical Principle

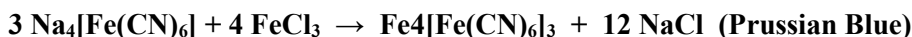
The qualitative detection of nitrogen in organic compounds is based on Lassaigne's Test (sodium fusion test). The principle relies on the conversion of covalently bonded nitrogen in the organic pesticide compound to the ionic form (cyanide,  $\text{CN}^-$ ) by fusion with metallic sodium at high temperature. The key reactions are:

**Na + C + N → NaCN (Sodium Cyanide)**

The cyanide ion in the sodium fusion extract subsequently reacts with a freshly prepared ferrous sulphate solution to form sodium hexacyanoferrate (II) (sodium ferrocyanide):



On acidification with dilute HCl and addition of ferric chloride (FeCl<sub>3</sub>), the ferrocyanide complex reacts to form Prussian Blue (iron (III) hexacyanoferrate(II)):



The formation of a characteristic blue or blue-green precipitate or coloration is interpreted as a positive test for the presence of nitrogen-containing pesticide residues in the sample.

## 2.5 Experimental Procedure

1. Each fruit/vegetable sample was washed under tap water and surface-dried. The samples were peeled where applicable and cut into small uniform pieces (approximately 1 cm<sup>3</sup>) using a clean stainless-steel knife.
2. Cut pieces (approximately 50 g per sample) were transferred individually into a mortar and thoroughly crushed to a homogeneous paste using a pestle.
3. Each homogenized sample was transferred to a separate 250 mL beaker and treated with 100 mL of absolute alcohol. Contents were stirred vigorously for 5 minutes using a glass rod and left to macerate at room temperature for 30 minutes.
4. The alcoholic extract was vacuum-filtered through Whatman No. 1 filter paper. The filtrate was collected in a pre-weighed china dish.
5. Filtrates in china dishes were evaporated to dryness on a water bath maintained at 80°C. The resulting dry residues were oven-dried at 60°C for 30 minutes to remove residual moisture.
6. A small piece of sodium metal (~50 mg) was placed in a clean fusion tube and gently heated over a flame until it melted.
7. A spatula of each dry residue was added to the molten sodium and the tube was further heated to red heat (approximately 800°C) for 2–3 minutes.
8. The hot fusion tube was immediately dropped into a china dish containing 10 mL of distilled water, shattering the tube. The contents were boiled for 5 minutes to extract the ionic fusion products.
9. The cooled solution was filtered to remove glass fragments and carbon residues. The clear filtrate (sodium fusion extract) was collected in a test tube.
10. To the sodium fusion extract, 1 mL of freshly prepared ferrous sulphate solution was added and the mixture was gently warmed. Subsequently, 2–3 drops of ferric chloride solution were added and the mixture was acidified with dilute HCl.
11. Formation of a blue or blue-green precipitate or coloration (Prussian Blue) was recorded as a positive result, indicating the presence of nitrogen-containing pesticide residues. Absence of coloration was recorded as negative. The entire procedure was repeated for all 12 samples.

## 3. RESULTS AND DISCUSSION

### 3.1 Observations

Table 1 summarizes the results of Lassaigne's nitrogen test applied to all 12 fruit and vegetable samples collected from local markets:

**Table 1:** Results of Lassaigne's Test for Nitrogen-Containing Pesticide Residues in Fruits and Vegetables

| S. No | Name of Fruit/Vegetable | Test for Nitrogen (Lassaigne's Test) | Presence of Insecticide/Pesticide Residues |
|-------|-------------------------|--------------------------------------|--------------------------------------------|
| 1     | Apple                   | Positive                             | Yes                                        |
| 2     | Grapes                  | Positive                             | Yes                                        |
| 3     | Strawberries            | Positive                             | Yes                                        |
| 4     | Orange                  | Positive                             | Yes                                        |
| 5     | Guava                   | Negative                             | No                                         |

|    |                    |          |     |
|----|--------------------|----------|-----|
| 6  | Pomegranate        | Positive | Yes |
| 7  | Tomato             | Positive | Yes |
| 8  | Spinach            | Positive | Yes |
| 9  | Beans              | Positive | Yes |
| 10 | Brinjal (Eggplant) | Positive | Yes |
| 11 | Potato             | Positive | Yes |
| 12 | Carrot             | Negative | No  |

### 3.2 Discussion

The experiment utilized 12 samples, comprising six fruits and six vegetables procured from local retail outlets. Ten samples (83.3%) showed evidence of pesticides or insecticides that contained nitrogen, identified through the formation of blue-green color (Prussian Blue) using Lassaigne's test. The positive samples included apple, grapes, strawberries, orange, and pomegranate, whereas guava showed negative results. The observation follows worldwide agricultural trends where strawberries, grapes, and apples require regular spraying of pesticides because they are vulnerable to infestations. Guava, on the other hand, possesses natural resistance against pests, explaining its negative results.

In the case of vegetables too, tomatoes, spinach, beans, brinjal, and potatoes gave positive results, whereas carrots tested negative. The prevalence of pesticides among vegetables can be attributed to their extensive use, especially in vegetables such as spinach and brinjal, which are extremely susceptible to insect attacks and infections. Since carrots are root vegetables, they may not have direct exposure to pesticide spray, and hence may even benefit from the filtering action of the soil. In summary, the presence of nitrile compounds indicates a high level of pesticide pollution in common vegetables. Though Lassaigne's test can only provide qualitative data, these results are consistent with those obtained through chromatography, indicating pesticide residue levels of organophosphates and carbamates often surpassing the permissible levels.

### 3.3 Health Effects of Pesticide Residues

The health implications of chronic exposure to pesticide residues in food are well-documented and multifaceted:

**Neurotoxicity and Nervous System Disorders:** Organophosphate compounds inhibit acetylcholinesterase, causing excessive nerve stimulation. Symptoms of acute poisoning include excess salivation, abdominal pain, vomiting, and diarrhea. Chronic low-dose exposure has been associated with Parkinson's disease, cognitive impairment, and peripheral neuropathy.

**Child Neurodevelopment and ADHD:** Children are disproportionately vulnerable due to higher food intake relative to body weight, greater gastrointestinal absorption and a more permeable blood-brain barrier. Epidemiological studies have linked elevated urinary organophosphate metabolites in children to attention deficit hyperactivity disorder (ADHD), autism spectrum disorder, and developmental delays.

**Carcinogenesis:** Several pesticides are classified as probable or possible human carcinogens by the International Agency for Research on Cancer (IARC). Evidence links chronic pesticide exposure to elevated risks of breast cancer, non-Hodgkin lymphoma, leukemia, and prostate cancer.

**Immunosuppression:** Pesticides have been found to reduce lymphocyte counts and impair white blood cell function, compromising the body's ability to mount effective immune responses against pathogens.

**Endocrine Disruption and Reproductive Toxicity:** Certain organochlorine and organophosphate pesticides interfere with hormonal signaling, potentially causing reproductive disorders, miscarriage, birth defects, and developmental abnormalities in the fetus and infant.

## 4. CONCLUSION

According to this study, nitrogen-based pesticide and insecticide residues are found in practically every fruit and vegetable sold in stores, with only two examples providing no positive results for nitrogen nitrates (i.e., guava and carrots). The large number of positive results (83.3% of tested fruits/vegetables) suggests levels of pesticide contamination in fresh produce are unacceptably high. In addition to raising significant public health concerns, this poses particular concern for at-risk populations such as children, pregnant females, and those who have weakened immune systems. The significant reliance on synthetics in conventional agriculture is responsible for the increase in pesticide residues in food, and while a benefit of using synthetics is improved crop yields, it also creates a significant risk to the safety of the food supply and the sustainability of the environment. Long-term exposure through diet to residues of these chemicals has been linked to numerous adverse health outcomes such as neurotoxicity, cancer, hormonal disruptions, and impaired immune function.

The following strategies would be instrumental in mitigating these challenges. Organic farming and regenerative agricultural methods can minimize the dependence on synthetic chemicals. Application of IPM practices can help optimize pesticide usage and still retain plant yields. There is a need for regulatory bodies to monitor pesticide residues in market foods and comply with set MRLs. Consumers can adopt decontamination procedures such as washing fruits and vegetables in a diluted vinegar solution (1:4) for fifteen minutes before rinsing thoroughly. Information dissemination programs must be developed to sensitize people about pesticides' health impacts and proper food handling practices. Research on pesticide quantification will play a significant role in future analysis of residue amounts. Advanced analytical approaches like GC-ECD and LC-MS will be utilized alongside QuEChERS extraction methods to determine individual pesticide molecules and assess compliance with national and international safety standards.

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