



Extraction of pesticide residues from cauliflower collected from cultivators and its nutraceutical analysis

Nayan Talukdar¹, Mafij Uddin Ahmed¹, Indrani Barman¹, Partha Pratim Kalita^{2*}

¹ Faculty of Science, Biotechnology Program, Assam Down Town University, Panikhaiti, Guwahati, Assam, India

² Assistant Professor, Faculty of Science, Biotechnology Program, Assam Down Town University, Panikhaiti, Guwahati, Assam, India

Abstract

Cauliflower is low in fat, high in dietary fiber, contains water and vitamin C, possessing a very high nutritional density. The warm and humid climatic condition of the country, increase use of modern high yielding varieties of crops and more use of chemical fertilizers are highly favorable for development and multiplication of pests and diseases. In Indian context, the cauliflower growers have been using the pesticides frequently to have the higher yield. Profex pesticide is widely used in the cultivation of cauliflowers in Assam. But the overdoses of pesticides make the residue problem, which might pollute our food and be harmful for our health. It has been reported that some of the pesticides are being used on northern part of the country where no pre-harvest time frame after application is maintained.

In the present study, a method employing TLC and UV Spectrophotometry for the separation, identification and quantification of a widely used pesticide on cauliflower was developed and validated. The compound studied was Profenofos. Finally, the method was applied to the determination of these pesticides in commercial samples collected from the cultivators. Therefore, the purpose of this study was to develop an analysis scheme for determination of some pesticides in cauliflower by TLC and UV Spectrophotometry. And this method provides good results for a specific pesticide present in cauliflower i.e. unwashed Sample – 0.084 gm /3ml, H₂O Washed sample – 0.070 gm /3ml and vinegar washed sample – 0.024 gm /3ml of sample. Quantification of protein in the samples were also done and compared and it showed the presence of almost equal amount of protein in both the samples. The amount of protein found in both the samples were 72 µg /ml in vinegar washed cauliflower sample and 70 µg /ml in unwashed cauliflower sample respectively.

Keywords: cauliflower, pesticides, profex, UV spectrophotometry, TLC

Introduction

Vegetables are imperative components of the human food habitat as they are the primary source for essential nutrients that are required for most of the biological reactions happening in the body. Like the other crops, vegetables are also being attacked by pests resulting in huge loss of yield as well as creates a big trouble in post-harvest storage. In order to minimize the loss of vegetables after harvest, pesticides are used together with other pest management techniques during cropping to destroy pests and prevent diseases. However, the use of pesticides during production often leads to the presence of pesticide residues in vegetables after harvest.

The presence of pesticide residues is a concern for consumers because pesticides are known to have potential harmful effects on human health. The major concerns are their toxic effects such as interfering with the reproductive systems and fetal development as well as their capacity to cause cancer and asthma [1]. Pesticides are used worldwide to protect crops before and after harvest in agriculture, gardening, homes and soil treatment. Variety of pesticides is used in current agricultural practice to manage pests and infections that spoil crops [2]. A wide range of pesticides (13–14%) are used for the production of fruits and vegetables in India due to heavy pest infestation throughout the cropping season of horticultural crops whereas cropped area is only 3% [3]. Pesticides have potentially adverse

effects on vegetables, fruits, animal resources resulting serious human health issues [4].

In the year 2000, the vegetable production in India was 92.8 million tones, grown over an area of 6 million hectares, which is about 3% of the gross cropped area of the country. Potato is the most important vegetable crop in India as it occupies 20% of vegetable area and contributes 27% to the total vegetable production. Nevertheless, vegetable production has been diversifying gradually [5]. Globally India occupies first position in the production of cauliflower, brinjal and peas, second in onion and third in cabbage [6]. Since vegetables are an important component of human diet, there is a need to protect these vegetables from agents like aphids, jasssids, diamond moths, caterpillars, etc [7, 8, 9]. *Brassica oleracea* which is commonly known as Cauliflower is an important variety of vegetable with an annual production of 3.39 million tones is heavily affected by various insects, resulting in severe loss of quality and production [9]. Pesticides are widely used to ensure high crop yields. They are used during production and post-harvest treatment of agricultural commodities [10]. However, increased use of chemical pesticides has resulted in contamination of the environment and also caused many associated long term health hazardous impact on human after consumption [11, 12, 13]. The heavy use of pesticides may result in environmental problems like disturbance of the natural balance, widespread pest resistance, environmental

pollution, hazards to non-target organisms and wildlife, and hazards to humans [14-17].

In order to remove residual effect of pesticides, which are toxic, we should know the exact dose which should be recommended to the farmer and the harvest time after spray of each pesticide so that the amount of residual pesticides in vegetables might be lower than the acceptable range [18]. The extraction and quantification of pesticide residues in food matrix mostly involved liquid-liquid extraction with a great variety of solvents and adsorbents for cleanup. Analytical techniques such as Gas Chromatography (GC) and High Performance Liquid Chromatography (HPLC) are widely used to monitor the presence of these compounds in water, soils, foods, fruits and vegetables [19, 20]. Several recent papers have reported advances in this field. With these background knowledge, the present study was designed to extract the pesticide residues from cauliflower samples, identify the pesticides present using TLC [17], to find out the amount of pesticides in each sample by UV Spectrophotometry and to estimate the amount of protein in the samples washed with vinegar to check any loss of quality by Lowry's Method.

Materials and Methods

Study Area

The study was conducted at Department of Biotechnology, Assam Down Town University and Central Instrumentation Facility Laboratory, Assam down town University, Assam.

Plant Material Collection

The cauliflowers were collected from farmers of Barpeta district, Assam, India. Samples were then transported to Department of biotechnology, Assam Down Town University in plastic bags.

Preparation of Different Extract

Cauliflowers were chopped and then divided them into three equal parts. First part was unwashed, second part was washed with water and third part was washed with a solution of vinegar and water. The samples were later chopped, grinded with 20 g of sodium sulphate, 2.5 g NaCl and 30 ml of chloroform and were shaken for 30 minutes. The aqueous materials were then filtered through whatman no.1 filter paper.

Preparation of Standards and Analysis

1 % standard solution of Profex pesticide was prepared in chloroform.

U.V Spectrophotometry

The prepared samples were tested in U.V Spectrophotometer at 323 nm wavelength to check the concentration of pesticide in each of the samples by plotting a graph.

Thin Layer Chromatography

TLC plate was activated by keeping at 105°C for 30 minutes and then cooled and the samples (standard, unwashed, washed and vinegar washed) were loaded on the plate. Then the plate was placed in the chromatography tank with air tight lid for the development of the spots.

Protein Estimation in the Samples

Standard Lowry's method was used to quantify the proteins in the samples.

Results and Discussion

U.V Spectrophotometry for Pesticide

Each of the samples was tested in U.V Spectrophotometer at 323 nm to find out the concentration of pesticides in each of them. The following results were seen from the graph.....

Unwashed Sample – 0.084 gm /3ml

Washed sample – 0.070 gm /3ml

Vinegar washed sample – 0.024 gm /3ml

The amount of pesticide is the lowest in the sample washed with vinegar, a little higher in the washed sample and highest in the sample which is unwashed.

Thin Layer Chromatography

The presence of pesticide in cauliflower sample was confirmed by TLC method and quantification was done by U.V Spectrophotometry which reveals the presence of pesticide in unwashed cauliflower Sample as higher, then in the washed sample and it was present least in the vinegar washed sample which developed very light spots. After investigating about the pesticide used in the cauliflowers, the farmers told that a pesticide named Profex is used during the cultivation of cauliflowers. This was quite shocking because a pesticide intended to kill bollworms in cotton is widely used in edible vegetables like cauliflowers, which is quiet dangerous in scientific view.

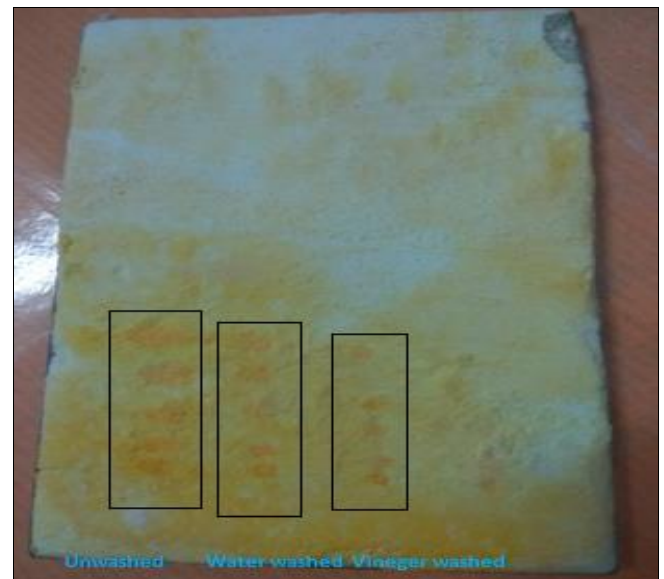


Fig 1

Washing the vegetables with vinegar prior cooking reduces a considerable amount of pesticides. This practice should be widely followed all over the world as pesticide consumption can cause many allergic reactions, diseases, biomagnifications etc [17].

Protein Estimation

The amount of protein present in the unwashed sample and vinegar washed sample was found by Lowry's Method and it was found that there was no change in the amount of protein content in both the samples as both samples gave almost the same values after plotting the graph which are as follows –

Amount of protein present in vinegar washed cauliflower sample: 72 µg /ml

Amount of protein present in unwashed cauliflower sample: 70 µg /ml

Conclusion

In the current study the pesticides were identified and also quantified. Comparative analysis of different cauliflower samples washed with water and vinegar were done based on which we can conclude that the pesticide residues remain in the vegetables long after harvesting and cannot be removed easily only by washing them with water. But we can reduce their concentration by washing them with a solution and water which does not interfere with the overall nutritive value of the cauliflower.

No doubt, the pesticides degraded within days but their degraded elements remain in vegetables. Maximum residue limit is the lowest limit at which a pesticide can cause disease to human beings. Residue limit of pesticides should not exceed from maximum residue limit. Regular checking of vegetables collected from different markets should be done to analyze the concentration of different pesticides in vegetable. Different techniques can be used for detection and quantification of pesticides but Thin Layer Chromatography (TLC) is a cheap method.

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