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Monitoring of Carbendazim residues in post-harvest treated Red Delicious apples

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ABSTRACT

This research paper aims to provide a comprehensive analysis of the presence and levels of Carbendazim residues in post-harvest treated Red Delicious apples in two condition i.e. ambient stored condition and cold stored condition. Carbendazim, a benzimidazole fungicide, has gained prominence as an effective means of controlling fungal diseases in apple orchards, including the commonly encountered apple scab and powdery mildew. The residue levels of Carbendazim in post-harvest treated Red Delicious apples, however, raise concerns about consumer safety and environmental impact. The investigation encompasses residue analysis methodologies, factors influencing residue levels, and potential mitigation strategies to ensure food safety and sustainability. The study showed that Carbendazim residue persistence was more in cold stored apple compared to ambient stored apple and to reduce its concentration to less than 5ppm it took 60 days, if pesticide applied ate recommended dose. It indicates, that the apple can be produced residue free and safe for consumption if judicious use of pesticides with respect to concentration and time is followed. Pesticides applied properly and scientifically avoiding drift to environment can reduce the impact on human health.

Key words : *Carbendazim, Apples, Pesticide residues*

Introduction

Apples (*Malus domestica*) are globally popular fruits, valued for their nutritional content and versatility in cooking. Despite their popularity, apple cultivation faces consistent challenges, with fungal diseases posing a significant threat to crop yield and fruit quality. Among these, apple scab (*Venturia inaequalis*) and powdery mildew (*Podosphaera* spp.) are particularly concerning. Apples, like several other fruits, can be stored for extended periods before being sold and consumed. Disease prevention is crucial in this process to maintain the produce's quality during storage. To address these diseases and ensure a plentiful supply of high-quality apples, farmers often turn to fungicides, with Carbendazim being a common choice due to its effectiveness against a wide range of fungal pathogens. However, the use of Carbendazim in apple orchards, especially in post-harvest treatments, has raised worries about potential residues in harvested fruit. These residues can have implications for consumer health, food safety, and environmental well-being.

The presence of carbendazim residues, a systemic fungicide, in treated apples is a matter of concern for both researchers and consumers (Kepczynska *et al.*, 1979; Rondelli *et al.*, 1985; Cano *et al.*, 1987; Papadopoulou-Mourkidou, 1991; Sharma *et al.*,

1997). Research has shown that these residues tend to be concentrated mainly in the peel and decrease towards the core of apples that were treated post-harvest using the immersion method (Cano *et al.,* 1987). The dissipation rate of carbendazim in fruit is influenced by factors such as variety, storage conditions, and formulation (Cano *et al.,* 1987; Kiigemagi *et al*., 1991; Awasthi *et al.,* 1997; Soudamini Mohapatra *et al.,* 1998). Studies have reported that carbendazim tends to persist for a longer period in apples that are stored in cold conditions (Kepczynska *et al.,* 1979; Rondelli *et al*., 1985; Cano *et al*., 1987). Sharma *et al.* (1997) suggested that when carbendazim is used at the recommended dose, it poses a low risk to health. Therefore, it is crucial to have a comprehensive understanding of carbendazim residues in post-harvest treated Red Delicious apples. This knowledge is essential for evaluating the safety of these apples for consumption and for developing strategies that strike a balance between effective disease control and considerations for consumer health and environmental sustainability.

This research paper aims to provide a comprehensive analysis of the presence and levels of Carbendazim residues in post-harvest treated Red Delicious apples. By shedding light on the intricacies of Carbendazim residue dynamics in this specific apple variety, this study contributes to the broader discussion on pesticide management, food safety, and sustainable agriculture. This study focuses on quantifying and understanding Carbendazim residues in post-harvest treated Red Delicious apples.

Materials and Methods

Sample preparation

40 kg of Red delicious variety apples were randomly selected from a pallet of apple. The samples were stored in the cold store maintaining at 4 degree Celsius with a relative humidity of 90-95% in order to avoid any degradation of residues between sampling and analysis. To a certain extent, the amounts of residues found in pesticide- treated fruit depend on the surface area of the fruit and on contact time (Norman *et al*., 1972; Ottender *et al*., 1975). Accordingly, the apples used in this study were previously calibrated for size; diameters ranged between 60 and 70 mm (measured perpendicular to the petiole). Selected apples were divided into four group consisting 10 kg in each group. Apples were treated with four different concentration. Carbendazim (50% WP), a broad-spectrum fungicide was applied at 500 ppm, 1000 ppm and 2000 ppm. The apples were rapidly plunged into a bath containing the fungicides and were left for 5 min. After applying pesticide at different concentration, samples were kept for 24 hours at room temperature. Half of the samples of each concentration were kept in cold storage and another half at room temperature for further study of degradation rate of Carbendazim. The treatments, including the untreated control were replicated 3 times. Apples were peeled and residue quantification was done separately for both peel and pulp.

Extraction and clean up

The sample preparation technique employed for analyzing pesticide residues in apples was the QuEChERS method. First, the apple samples were weighed using a digital weighing balance and blended to ensure a uniform mixture. Subsequently, 5 g of the homogenized sample was placed in an Oakridge centrifuge tube. Then, 5 g of acetonitrile solvent was introduced, and the mixture was vortexed for 2 minutes. Following this, 2 g of sodium sulphate and 0.75 g of NaCl were added to the mixture, which was vortexed for an additional 2 minutes. The mixture was then centrifuged at 5000 rpm for 5 minutes. A portion of 1 ml of the resulting supernatant was transferred to a microcentrifuge tube for subsequent cleaning steps. To this microcentrifuge tube, 150 mg of anhydrous $MgSO_4$ and 50 mg of PSA were added. The mixture was vortexed for 2 minutes, and centrifuged at 5000 rpm for 5 minutes. The resulting supernatant (0.5 ml) obtained after the cleaning step was filtered through a nylon syringe filter with a pore size of 0.22 μm into a 2 mL glass auto-sampler vial. This filtered extract was then prepared for further analysis using LC-MS/MS.

Recovery of Carbendazim

Before analyzing actual sample for quantification of pesticide, the efficiency of the method was evaluated in recovery experiments by spiking untreated samples of fruits (collected from control plots) and fortified with Carbendazim pesticide at four different levels (0.01, 0.05, 0.1, 1 ppm). A 50 g well homogenized sample of Carbendazim was spiked with known amount of standard pesticides at the 4 con-

centrations each replicated thrice. The fortified samples were extracted and cleaned with the method followed for analyzing actual samples.

Results and Discussion

The recoveries for the four levels fell within the acceptable tolerance of 70 to 120 per cent range (SANCO/12571/2014) indicating good performance of extraction, clean up and chromatographic parameters for Carbendazim residue determination in Red delicious apple (Table 1). The relative standard deviations (RSDs) were less than 20 per cent for all the levels.

Table 1. Recovery of Carbendazim residues from the fortified samples of Red Delicious variety ofapple fruit

Amount fortified μgl^{-1})	Average recovery (%)	Relative standard deviation(%)
0.01	82.34	15.24
0.05	85.17	14.36
0.1	86.82	10.45
1	90.31	11.17

The average recoveries of Carbendazim in apple fruit fortified at four different levels (0.01, 0.05, 0.1, 1 ppm) varied from 82.34 – 90.31 percent.

Following dipping for 5 minutes in 500, 1000 and 2000ppm the apple contained initial deposits of 4.8, 11 and 20.8 ppm respectively at the corresponding concentration. The concentration of carbendazim residues in post-harvest treated apples and its degradation when the samples are kept in ambient and cold storage condition is shown in Fig. 1, 2 and 3. It was observed from the Fig. 1, 2 and 3 that persistence of Carbendazim in red delicious apple was more in cold storage condition than compare to ambient condition. Similar results also reported by Kepczynska *et al*. (1979). When the apples are stored in ambient temperature around 50 percent of the residues were degraded in 10 days in both peeland pulp samples. The residue levels were less were found to be less than 1 ppm on 30th day and the similar results have been reported by Jain and Agnihotri (1987). When apples were kept in cold store it takes more time to degrades carbendazim residue in apple. In cold stored apple it takes more than 60 days to lost 50% of the Carbendazim residue in both peel and pulp. Similar type of results also reported by Monico and Xiram (1987). In cold stored peel samples, it took 120 days of treatment to reduce the residues to less than 1ppm level. In cold stored pulp samples residues gradually lowers down to about 1ppm within 60 days of treatment. Most of the pesticide residues are concentrated in peel of the apple. The maximum residue level (MRL) fo rcarbendazim in apple is 2 mg kg^{-1} (Pesticides Regulations, 2002). According to World Health Organization (1974), the acceptable limit of Carbendazim was (5ppm). Most of the carbendazim residue are concentrated on peel compared to pulp, so it took more time to degrade. Treatment and time had effect for the concentration of carbendazim residue in both apple flesh and peel. For cold storage condition residues degraded to less than 5 ppm in 60 days.

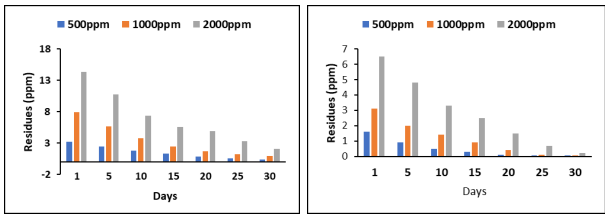

Fig. 1a. Residue of Carbendazim at ambient condition for peel (b) Residue of Carbendazim at ambient condition for pulp

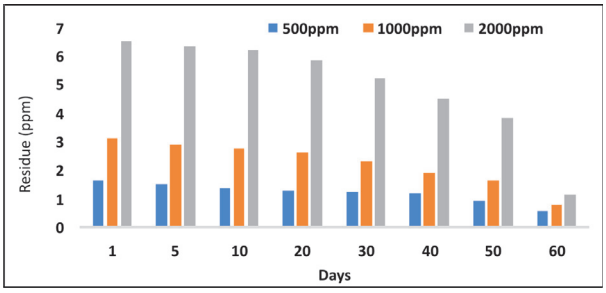

Fig. 2. Residue of Carbendazim at Cold storage for peel

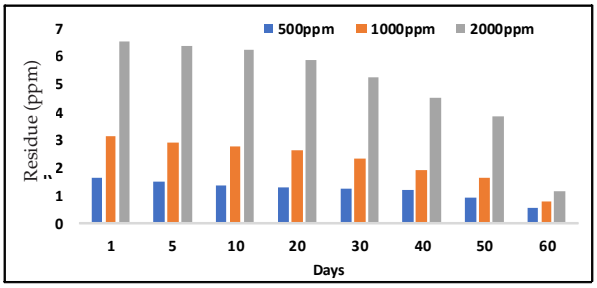

Fig. 3. Residue of Carbendazim at Cold storefor pulp

Conclusion

The fruit samples of Red Delicious variety of apple

treated with Carbendazim 50% WP. Carbendazim 50% WP was 10 applied at 500ppm, 1000ppm and 2000ppm concentration on Red Delicious apple after harvesting. If applied at recommended dose of 1000 ppm then after 60 days residues isreduced in cold stored sample and decreased down to below MRL as set by World Health Organization (1974). It indicates, that the apple can be produced residue free and safe for consumption if judicious use of pesticides with respect to concentration and time is followed. However, waiting period based on the prescribed maximum residue limits needs be adopted strictly as index of safety to consumers. Pesticides applied properly and scientifically avoiding drift to environment can reduce the impact on human health.

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