Novel *Prosopis juliflora* leaf ethanolic extract as natural antifungal agent against *Botrytis cinerea*: application on strawberries shelf-life extension

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1. **ABSTRACT**

**Introduction:** Fresh produces’ spoilage is a worldwide concern that accompanies the global increase in food demand. Adverse human health and environmental effects of commercial pesticides is a major public concern.

**Material and Methods:** In this report, our *Prosopis juliflora* water-soluble leaves ethanolic (PJ-WS-LE) extract will be described as an effective antifungal agent.

**Results:** PJ-WS-LE extract showed total inhibition of *Botrytis cinerea* growth and extend strawberries shelf-life.

**Future directions:** PJ-WS-LE extract will be chemically described and its effectiveness on other fresh produces will be evaluated.

2. **BACKGROUND**

*Botrytis cinerea* is one of the top ten pathogens that affect fresh produce shelf-life around the world. With a global effort to control *B. cinerea* grey mold, countries spend a lot of money on botryticide, wide spectrum fungicides and bio-controllers. *Botrytis cinerea* can progress easily from infected fruits to healthy ones even at low storage temperatures which can lead to entire lots spoilage in few weeks. *Prosopis juliflora* has been explored for its antimicrobial activities. However, studies are not interconnected.

Strawberries are widely consumed raw berries which are famous in their processed forms such as jam and juices. The delicate fruit has a very short postharvest life, it is susceptible to mechanical injuries, fast dehydration and fungal infection. Strawberries’ most common spoiling fungi is *B. cinerea*. Our PJ-WS-LE extract is a promising fungicides alternative that showed in-vitro effectiveness against *B. cinerea*.

3. **OBJECTIVES**

- Identifying a natural plant extract bio-controller.
- Controlling *B. cinerea* fungal postharvest spoilage.
- Extending strawberries shelf-life.
- Maintaining strawberries quality parameters during storage.

4. **METODOLOGY**

4.1 Disk antifungal method

Used to test the effect of 20mg/ml of PJ-WS-LE extract in PDA plate on *B. cinerea* growth compared to normal growth.

4.2 Determination of minimum inhibitory concentration

96-well plate method was used to determine PJ-WS-LE extract MIC against *B. cinerea*.

4.3 Mode of action

The inhibited *B. cinerea* disk was transferred to a clean PDA plate to determine the extract mode of action.

4.4 Effect of PJ-WS-LE extract on the microscopic morphology of *B. cinerea* using SEM

Cultures of *B. cinerea* were prepared in the presence & absence of 8mg/ml of PJ-WS-LE extract. SEM pictures of spores and hyphae were taken.

4.5 Determination of shelf-life of strawberry samples

Treated strawberry samples (8mg/ml PJ-WS-LE extract) and control samples had their shelf-life determined at room temperature and at 4°C.

4.6 Strawberries storage quality parameters monitoring

Quality parameters of treated and control samples were monitored on a weekly basis: weight loss, bacterial and fungal CFU, firmness, pH and TSS.

5. **NOVEL EXTRACT PREPARATION METHOD**

**Collected leaves**

*Prosopis juliflora* leaves are collected from Jordanian desert provinces.

**Washing**

Leaves are washed with running water and then with distilled water.

**Grinding**

Leaves are ground using a grinder and sieved through a 40-μm sieve.

**Extraction**

The sieved leaves are extracted with 80% methanol for 24 hours at room temperature.

**Drying**

Extracts are dried in an oven at 40°C.

**Purification**

Purification is done using ethyl acetate to separate the polar and non-polar fractions.

**Assessment**

The effectiveness of the extract is assessed against *B. cinerea* using in-vitro assays and on strawberries shelf-life.

6. **RESULTS & DISCUSSION**

6.1 Disk antifungal methods results of *B. cinerea*

PJ-WS-LE extract has fungicidal effect on *B. cinerea*.

6.4 Effect of PJ-WS-LE extract on the weight loss percent of strawberries

Fig 5. Weekly weight loss percent (Control (A), coated with 8mg/ml PJ-WS-LE extract (B), coated with 1% chitosan (C)).

Control samples that showed fungal rot were mainly infected with Botrytis which shows the extract efficacy against it.

7. **CONCLUSIONS**

- PJ-WS-LE totally inhibits the growth of *B. cinerea* with fungicidal activity.
- MIC of PJ-WS-LE extract against *B. cinerea* is 1mg/ml.
- Coated strawberries (8mg/ml PJ-WS-LE extract) had their shelf-life at 4°C extended from 4.3 days to 10 days.
- 8mg/ml PJ-WS-LE extract coating is more effective that 1% chitosan.
- PJ-WS-LE extract showed promising fungicidal characteristics that allow it to replace chemical fungicides.

8. **REFERENCES**
