

## Post harvest treatment of crops using emulsified petroleum

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**Abstract:** The present invention relates to the post-harvest treatment method for eradication of biological infestation in crops including pests, parasites, funguses, moulds, etc. comprises the step of applying post-harvest to the crop by petroleum oil that has a density at 15° C. in the range of 0.75 to 0.83 g/ml.

**Keywords-** Emulsion, Efficacious, Aliphatic, Calyx, Eradication, Larvae, Inoculums, Infestations

### Introduction

The present invention relates to the post-harvest treatment of crops. More particularly, the invention relates to the application of petroleum derived oils and emulsions With specified characteristics to crops such as fruit and vegetables for treating biological infestations including pests, parasites, funguses, moulds, etc. Often become biologically contaminated post-harvest. Contamination can be initiated pre-harvest (e.g. by parasitic presence at the time of picking/harvesting), during harvesting (e.g. Where contaminants are introduced by mechanical harvesters or human intervention) and post-harvest (e.g. Where parasites and spore settle on post-harvested produce). Regardless of the time of contamination, it is desirable to treat harvested fruit, vegetables and plants prior to transportation and storage to eradicate any such contamination. For example, international quarantine regulations and inspection require fruit to be free of live pests. Typically, the oil is applied in the form of oil in water formulation, most preferably as an emulsion he oil is emulsified in the Water to be present in a range of 1—6%, more typically at about 3% by total volume. Such oils have been found to have a capacity to efficiently cover, spread and/or penetrate various crops (especially fruit) to which they are applied so as to penetrate and thus exterminate pests, parasites, funguses, mould, bacteria etc. Light paraffinic oils have the advantage of low viscosity which can aid in penetrating difficult to access regions on crop surfaces (e.g. under the calyx of citrus fruit).

### Mode of carrying out the invention

Notwithstanding any other forms which may fall within the scope of the present invention, various preferred forms of the invention will now be described with reference to the following non-limiting examples like Post-Harvest Treatment of Citrus Fruit for the elimination of Mealybug and Light Brown Applemoth

The need for the elimination of these pathogens arose because the use of post-harvest broad spectrum pesticides (other than

fungicide and Wax baths) was (and is currently) not allowed with export fruit (e.g. to the USA) due to concerns over residues. Many growers were moving back to liberal applications of broad spectrum pesticides in the field (i.e. pre-harvest) in the hope of delivering pest free fruit to processors. In accordance with the present invention, various formulations were applied post-harvest to citrus fruit to test the efficiency against insect pests infecting the same. These formulations are tabulated in the examples below. The calyx of citrus fruit was observed to provide a protective shelter for a range of small arthropods, including light brown apple moth (LBAM) (epiphyas postvittana). LBAM spins a silken domicile that is hydrophobic and thus many LBAM larvae survive prior art washing processes prior to packing so that the larvae develop in storage. Experiments were conducted to identify a treatment which ensured 100% eradication of mealybug and light brown apple moth from citrus fruit in post-harvest processing facilities. Treatments were applied using a dipping bath (mean volume=2,500 L) at a rate of 5% (i.e. 250 L) Where the bath was renewed after treatment of 80 tone of citrus. The expected citrus mass to be treated each growing season was assumed to be 10,000 tones (Which thus equated to an estimated 15,000 L of product).

Example 1

Component	PCM no.	CAS no.	Volume %
Light solvent extracted, dewaxed, hydrotreated paraffinic oil	1022	74742-56-9	92.3
Polyethylene glycol dioleate	2564	9005-07-6	5.56
Sorbitan mono-oleate water	2023	1338-43-8	1.28

Example 2

Food grade oil plus surfactants graded for incidental contact with food

Component	PAS no.	Volume %
White mineral oil (medicinal paraffin)	8042-47-5	94
Cetyl-oleyl alcohol condensed	68439-49-6	4.8
Polyethylene sorbitan trioleate	9005-70-3	1.2

Example 3

Food grade oil plus food grade surfactants

Component	CAS no.	Volume%
White mineral oil	8042-47-5	90
Mono oleate polyoxyethylene sorbitan	9005-65-6	6.2
Sorbitan mono-oleate	1338-43-8	3.8

Example 4

Food grade solvent plus surfactants graded for incidental contact with food

Component	CAS no.	Volume %
Paraffinic(c15)solvent<5 ppm sulphur	64771-72-8	80
Cetyl-oleyl alcohol condensed	68439-49-6	7.2
Polyoxyethylene sorbitan trioleate	9005-70-3	4.8

Example 5

Food grade solvent plus food grade surfactants

Component	CAS no.	Volume %
Paraffinic(c15)solvent<5 ppm sulphur	64771-72-8	80
Mono oleate polyoxyethylene	90058-65-6	10.4
Sorbitan mono-oleate	1338-43-8	9.6

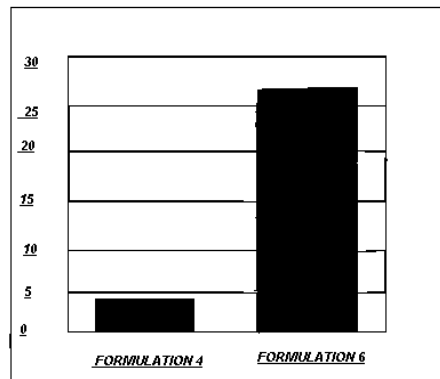
Example 6

Component	CAS no.	Volume%	Mass%
C15 aliphatic hydrocarbon	64771-72-8	87.12	85.2
Food grade, paraffinic base oil	NA	9.16	10
Glycerol mono-oleate	NA	0.99	1.2
Polyoxyethylene sorbitan trioleate surfactant	973005-70-3	2.73	3.6

For Examples 1 to 3, initial mortality studies on exposed light brown apple moth were promising when the product was used at 5% in Water (Table 1). However, latter Work on insects hidden under the calyx of oranges identified that Examples 4, 5 and 6 were superior. This was attributed to the lower density of the base oil used in Examples 4 to 6 as opposed to the oil used in the first three formulations. Formulation 4 was appealing, being a very stable emulsion but was not very efficacious unless used at very high treat rates, Formulation 5 provided a very quick breaking emulsion which was surmised to have applications With other crops (i.e. other than citrus dipping). Formulation 6 was the most suitable as a post-harvest spray and post-harvest dip for citrus fruit, and analysis confirmed that Formulation 6 had the best oil deposition behavior (FIG. 1).

Acceptable oil-in-Water volume % ranges in the formulations were found to be from about 1% to about 6%.

Fig1:%o



il

deposited on test strips

Field trial

A confidential field trial of formulations 5 and 6 was carried out in a 1,000 L bath and each formulation was used to treat one crate of citrus fruit. The trials were successful in that 100% control of insect pests was obtained and no detrimental effect on fruit quality was perceived. Further, the fruit was let stand for 24 hours post dip and was then processed in the normal Way. Oil dipped fruit gave a better appearance after waxing than did control fruit and initial long term storage tests indicated that treated oranges had a better “orange color” than control fruit. This was surmised to be due to enhanced ethylene production in the treated fruit. Also, no adverse taste perception was obtained.

Details of formulation components

The preferred use of the formulations was in dipping baths with continual mixing. Thus, the emulsion characteristic of “good strike” was most important. An emulsion with good strike was one that displayed easy mixing and an even milky appearance. A desirable quality of the formulation when used for citrus treatment was its capacity for enhanced ingress under the fruit calyx. It was seen to be an advantage if the emulsion did not break between treatments of crates of citrus fruit. The choice of surfactants was confined to those which were either food grade quality or graded safe for incidental contact with food. All formulations were observed to be stable With respect to phase separation at 0° C, 20° C. and 43° C. Blending of Formulations 4, 5 and 6 required no special techniques and was simply a matter of adding the appropriate amount of surfactant to the C15 oil and mixing until a clear solution was obtained as noticed. The biocontrol properties of various emulsions (primary formulations 4 5 and 6) were tested against green mould (Penczllium dzgzatum) in oranges. Two experiments were conducted to test these properties. Experiment No. 1 tested for localized biocontrol where the suspensions were directly pipette into Wounds. In experiment No. 2, fruit were dipped into the proposed biocontrol solution.

Experiments performed

Experiment no. 1

Localised Biocontrol

Aim: Assessing biocontrol activity of the oil by directly pipetting oil and spores into Wounds.

Treatments:

1. Unwounded control
2. Wounded only control
3. Wounded, oil emulsion, spore suspension
4. Wounded, spore suspension, oil emulsion
5. Wounded, oil emulsion, Wait 2 hrs, spore suspension
6. Wounded, spore suspension, Wait 2 hrs, oil emulsion
7. Wounded, oil emulsion, Wait 24 hrs, spore suspension
8. Wounded, spore suspension, Wait 24 hrs, oil emulsion
9. Wounded, spore suspension
10. Wounded, Wait 2 hrs, spore suspension
11. Wounded, Wait 24 hrs, spore suspension

Method: 110 (10 reps of 1 fruit/treatment; 2 wounds/fruit) oranges with normal post-harvest treatments were surface sterilized With 70% ethanol, allowed to dry, wounded with a sterile nail to make a wound size of (3mm Wide\*5 mm deep), and inoculated with the oil and Penicillium spores. The oil treatment was 3% in water to make an emulsion, which was stirred constantly. 5 microlitre of this was pipette/Wound. The spore inoculum was washed spores of Penicillium digitatum at a concentration of (1\*10<sup>6</sup> spores/ml). 5 microlitre of this suspension was pipette into each Wound. Fruit were assessed every day for incidence and severity of disease. (Severity being defined as the average diameter of the lesion in mm.

Table 3

Results-incidence=% of wounds with disease

Treatment	4 days %	5 days %	6 days %	7 days %
1.unwounded	0	0	0	0
2.wounded only	0	0	0	0
3.wound,oil,spores	0	0	0	0
4.wound,spores,oil	0	0	0	0
5.wound,oil,2 hrs, spores	0	0	0	0
6.wound,spores,2 hrs, oil	0	0	0	0
7.wound,oil,24 hrs, spores	25	50	50	55
8.wound,spores,24 hrs, oil	15	40	50	50
9.wound,spores	20	40	50	50
10.wound,24 hrs, spores	15	45	50	50

Table 4

Severity –avg. diameter of lesions in mm

Treatment	4 days %	5 days %	6 days %	7 days %
1.unwounded	0	0	0	0
2.wounded only	0	0	0	0
3.wound,oil,spores	0	0	0	0
4.wound,spores,oil	0	0	0	0
5.wound,oil,2 hrs, spores	0	0	0	0
6.wound,spores,2 hrs, oil	0	0	0	0
7.wound,oil,24 hrs, spores	2.650+-1.440	10.200+-3.890	23.4+-8.04	33+-10.7
8.wound,spores,24 hrs, oil	1.500+-0.764	7.050+-3.04	19.9+-7.66	30.25+-10.2
9.wound,spores	1.500+-0.764	7.050+-3.04	19.9+-7.66	30.25+-10.2
10.wound,24 hrs, spores	2.000+-1.070	8.450+-3.77	20.8+-8.04	30+-10.3

1.unwounded	0	0	0	0
2.wounded only	0	0	0	0
3.wound,oil,spores	0	0	0	0
4.wound,spores,oil	0	0	0	0
5.wound,oil,2 hrs, spores	0	0	0	0
6.wound,spores,2 hrs, oil	0	0	0	0
7.wound,oil,24 hrs, spores	0	0	0	0
8.wound,spores,24 hrs, oil	2.650+-1.440	10.200+-3.890	23.4+-8.04	33+-10.7
9.wound,spores	1.500+-0.764	7.050+-3.04	19.9+-7.66	30.25+-10.2
10.wound,24 hrs, spores	2.000+-1.070	8.450+-3.77	20.8+-8.04	30+-10.3

It can be seen that no disease was recorded for was pipetted into each wound the control treatments (unwounded and wounded only); for Fruit were assessed every second day for incidence and for severity of disease. Severity being the average diameter of wounded+oil+wait 2 hrs then spores; for wounded+spores+25 the lesion in mm, wait 2 hrs then oil and for wounded+oil+wait 24 hrs then spores. The treatments where disease did occur Were in wounded+spores+Wait 24 hrs then oil; wounded+spores; wounded+wait 2 hrs then spores and in wounded+Wait 24 W hrs then spores. From table 3, the percentage incidence did not vary significantly for the treatments Where Penzclllzum digitatum produced green mould in the oranges. Table 4 shows the severity, giving the average diameter of the lesion recorded for each treatment. The average diameter of the lesions did not vary significantly between those spores treatments where disease occurred.

After 7 days, the average lesion diameter for fruit not treated with the oil emulsion was 30.17+-15 .71 mm. The average diameter of the lesions for fruit that were treated with the oil was 5.50:2.34 mm. This result of 5.50+-2.34 mm was due solely to the treatment of wound+spores+Wait 24 hrs then oil.

Conclusion

From these results, the various oil emulsions were shown to have biocontrol properties against green mould of orange. When pipetting the oil emulsion and spore suspension directly into the wounds .The only treatment where the oil did not control the disease was for treatment 8—wound+ 7. Wound, spores, oil dip spores+Wait 24 hrs then oil. This was because the disease already had a hold on the fruit during the 24 hrs before the oil was applied.

Experiment no. 2

Biocontrol by dipping fruit in the oil emulsion

Aim: Assessing biocontrol activity of various formulations by dipping

Treatments:

1. wounded only
2. wounded ,oil dip
3. wounded,spore suspension
4. wounded,oil suspension
5. oil dip wounded
6. oil dip,wounded,spore suspension

Method: 110 (8 reps of 2 fruit/treatment;2 wounds/fruit)oranges with no post harvest treatment were surface sterilized with 70% of ethanol and allowed to dry. Dipping was done for 30 seconds in a shallow bucket on a magnetic stirrer;2 fruit were dipped at one time. Dipped fruit were then left to dry. Wounding was done with a sterile nail to make a wound size of 3mm wide)\*5mm deep. The spore inoculum was washed spores of *penicillium digitatum* at a concentration of  $3 \times 10^5$  spores/ml.5 microlitre of his suspension was pipetted into each wound. Fruit were assessed every second day for incidence and severity of disease

Table 6

Incidence-%of wounds with disease

Treatment	3 days %	5 days %	7 days %
Wounded only	0	0	0
Wound ,oil dip	0	0	0
Wound spores	100	100	100
Wound,oildip,spores	53	100	100
Oil dip,wound	0	0	0
Oil dip,wound,spores	84	100	100

Table 7

Severity –diameter of lesions in mm

Treatment	3 days mm	5 days mm	7 days mm
Wounded only	0	0	0
Wounded,oil dip	0	0	0
Wound,spores	11.09+-0.65	28.69+-1.42	44.87+-1.79
Wound ,oil dip,spores	3.81+-1.09	23.62+-0.61	39.19+-1.31
Oil dip	0	0	0
Oil dip,wound ,spores	4.84+-0.86	26.12+-1.85	40.62+-0.90

From table 6, it can be seen that no disease was recorded for the control treatments (wounded only; Wound+oil dip & oil dip+Wound). For the wound+spores treatment, all wounds had disease (100%) at just 3 days after inoculating. For 3 treatments with the Oil (Wound+Oil dip+Wound+spores and Wound+spores+oil dip) and after 3 days the incidence was 53, 84 and 62.5% respectively. All treatments causing disease with the oil dipping and spores had significantly less disease than the Wound+spores treatment. Table 7 verifies the results already discussed giving the average diameter of the lesions. At 7 days the fruit with the treatment Wounded+spores gave the diameter of lesions of 4487+-1.79 mm.

### Conclusion

From these results, it was concluded that the preferred oil emulsions did have biocontrol properties against green mould of orange when the oranges were dipped into the oil emulsion. The

treatments with oil dipping and spores slowed down the disease by up to 2 days, compared with the spores alone.

### Comparison of Experiments

In experiment 1, (pipetting directly into Wounds), reached up to 55%, whereas in experiment 2, (fruit dipped in oil emulsion), the incidence reached up to 100% 3 days after inoculating. This difference was surmised to be due to a difference in quality of the fruit and the fruit from experiment 2 had no post-harvest treatments of fungicides etc, thus making it more susceptible to disease. Greater control by the oil was obtained in experiment 1 compared with experiment 2. In experiment 1, the oil emulsion and spores were directly pipetted into a small Wound 3 mm wide\*5 mm deep. In experiment 2, the fruit were only dipped in the oil emulsion. In experiment 1, the oil was in closer proximity to spores and possibly caused a barrier in the wound to not allow the spores to infect the fruit. This may account for the greater control recorded for experiment 1.

### General Conclusion on Treatment of Green Mould

From the results of these two experiments, it can be seen that the preferred oil emulsions of the invention protected oranges against green mould caused by *Penicillium digitatum* when spores and oil were pipetted into wounds or when fruit was dipped into the oil emulsion.

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