INFLUENCE OF CPPU ON THE BERRY GROWTH AND COMPOSITION OF SEEDLESS AND SEEDED TABLE GRAPE CULTIVARS

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Summary
Trials conducted in 2003 examined the effects of CPPU on several seedless and seeded table grape cultivars. CPPU increased the berry size of GA treated seedless cultivars (Thompson Seedless, Flame Seedless and Crimson Seedless) an additional 5 to 20%, depending upon rate, compared to the standard GA treatment. CPPU increased the berry size of Redglobe up to 15% when applied alone, but did not improve size when applied in combination with a 40 mg/L GA (cluster directed spray). However, CPPU dramatically increased berry firmness of Redglobe, resulting in a significant quality improvement and market benefit. CPPU changed the shape of cylindrical berries (ex: Thompson Seedless and Crimson Seedless) by increasing their diameter:length ratio; berries of these cultivars appear ovoid to oblate after CPPU treatment. CPPU applied at berry set delayed fruit maturity (sugar accumulation) up to two weeks, depending upon the cultivar and application rate. Titratable acidity, berry firmness and capstem removal force were also increased in most cases. CPPU significantly reduced pigment development in the skin as well as the percentage of clusters with adequate color for harvest in most red cultivars. Crimson Seedless was an exception, as CPPU had no significant detrimental effects on berry color over three years. Ethrel was only partially effective for overcoming detrimental effects on color development, and the greatest problem with the use of CPPU is the reduction of fruit color on Flame Seedless. Autumn Royal did not respond to CPPU or GA sizing treatments. Optimum rates for berry sizing appear to range between 5 and 6 grams for most cultivars. Slightly higher rates, up to 8 grams per acre, are suggested for maturation delay and reduction of ambering on white cultivars such as Thompson Seedless and Princess.

INTRODUCTION
CPPU (forchlorfenuron or N-(2-chloro-4-pyridyl)-N'-phenylurea) is a synthetic cytokinin, which has significant physiological activity on many fruits, including grapes. The compound was originally formulated in the mid-1980's, and has been tested and registered for use on a variety of fruit crops throughout the world during the past two decades. An Experimental Use Permit (EUP) was granted for the use of this compound on table grapes in California in 2001.

The primary physiological effects of CPPU on grapevines involve the regulation of fruit set, berry growth and development. When applied immediately prior to anthesis or bloom, 4 to 8 g/ac CPPU typically increases the fruit set of both seedless and seeded grape cultivars. When applied shortly after fruit set, 2.5 to 15 g/ac CPPU stimulates both cell division and cell elongation, resulting in significant increases in berry size. CPPU applied at fruit set also delays fruit maturation, slowing the accumulation of sugar and color, as well as the respiration of acids. High rates (≥10 g/ac) applied at fruit set may permanently retard color development, particularly on sensitive cultivars such as Flame Seedless and Redglobe, and have also been reported to alter berry flavor and texture. CPPU does not reduce the fruitfulness of either seedless or seeded table grape cultivars the year following its application.

Previous work with CPPU has focused on traditional seedless table grape cultivars, and treatment responses for both Thompson Seedless and Flame Seedless are well described in the literature. Previous studies indicate that CPPU should be used in combination with GA in order to achieve synergistic effects on berry growth and fruit maturation. When used in conjunction with GA, optimum rates for CPPU on Thompson Seedless and Flame Seedless range between 4 and 12 g/ac and 2 and 8 g/ac, respectively. Lower rates are generally suggested for Flame Seedless due to potential detrimental effects on color.
development. Further work is needed to examine treatment timing x rate interactions for other prominent table grape cultivars currently grown in California. A major objective of this work is to evaluate the effects of CPPU berry sizing treatments on seeded cultivars (ex. Redglobe), or GA sensitive seedless cultivars (ex. Crimson Seedless, Autumn Royal), where whole-vine sprays of GA at fruit set are detrimental to either fruit quality or vine fruitfulness. Due to the detrimental effects of this compound on fruit pigment accumulation, work is also needed to examine CPPU x ethephon interactions on colored cultivars.

PROCEDURES
Growing conditions and cultural practices
Studies were established on mature Flame Seedless and Thompson Seedless grapevines planted in a fine sandy loam soil at the Kearney Agricultural Center in Parlier. The vines were spaced 8' (between vines) x 12' (between vines). The Flame Seedless was bilateral cordon trained, spur pruned and trellised to the standard "T" system. The Thompson Seedless was head trained, cane pruned and grown on the open gable trellis system. The Redglobe, Crimson Seedless and Autumn Royal experiments were conducted in a commercial vineyard near Fowler, CA. The vines are grafted to Freedom rootstock, planted in a fine sandy loam soil, and spaced 8' (between vines) x 12' (between rows). The Redglobe and Autumn Royal vines were quadrilateral cordon trained and trellised to the open gable system. The Crimson Seedless vines head trained and cane pruned. All vines were drip irrigated, and cultural operations were performed in accordance with standard commercial practices for each cultivar.

Experimental treatments
Flame Seedless
The Flame Seedless experiment examined GA x CPPU rate interactions on berry size and color development. GA was applied to the vines at 20, 40 or 60 g/ac alone or in combination with 5 g/ac CPPU. Treatments were applied to both fruit and foliage using a hand-held spray wand and 200 gallon of spray solution per acre. Treatments were applied at fruit set, when mean berry diameter was approximately 6 to 7 mm. All vines were adjusted to approximately 28 clusters following berry set.

Thompson Seedless
The Thompson Seedless experiment examined GA x CPPU rate interactions on berry size and fruit maturation rate. GA was applied to the vines at 20, 40 or 80 g/ac alone or in combination with 10 g/ac CPPU. Treatments were applied to both fruit and foliage using a hand-held spray wand and 200 gallon of spray solution per acre. Treatments were applied at fruit set, when mean berry diameter was approximately 7 to 8 mm. All vines were adjusted to approximately 30 clusters following berry set.

Redglobe
This experiment examined the effects of CPPU and GA on the berry growth and color development of Redglobe table grapes. The following treatments were evaluated at fruit set (7 to 8 mm berry diameter) and fruit set + 2 weeks (10 to 11 mm berry diameter):

1. 40 mg/L GA applied as a cluster directed spray
2. 8 g/ac CPPU applied as a whole-vine application
3. 40 mg/L GA applied as a cluster directed spray + 8 g/ac CPPU
4. Untreated control

CPPU solutions were applied to both fruit and foliage using a hand-held spray wand and 200 gallons of spray solution per acre. Crop loads adjusted to similar levels (25 clusters per vine) on all vines following
Crimson Seedless
This experiment examined the effects of CPPU and GA on the berry growth and color development of Crimson Seedless table grapes. The following treatments were evaluated at fruit set (7 to 8 mm berry diameter) and fruit set + 2 weeks (10 to 11 mm berry diameter):

1. 4 g/ac GA
2. 4 g/ac CPPU
3. 4 g/ac GA + 4 g/ac CPPU
4. Untreated control

An additional experiment was initiated to examine the effect of CPPU application rate (4, 8 and 12 g/ac) and timing (fruit set and fruit set + 2 weeks).

Autumn Royal
This experiment examined the effects of CPPU and GA on the berry growth and color development of Autumn Royal table grapes. The following treatments were evaluated at fruit set (7 to 8 mm berry diameter) and fruit set + 2 weeks (10 to 11 mm berry diameter):

1. 8 g/ac GA
2. 8 g/ac CPPU
3. 8 g/ac GA + 8 g/ac CPPU
4. Untreated control

Experimental solutions were applied to both fruit and foliage using a hand-held spray wand and 200 gallons of spray solution per acre. Crop loads adjusted to similar levels (35 clusters per vine) on all vines following berry set.

Experimental design
All treatments in each experiment were replicated 8 times, using three vine plots arranged in a randomized complete block design. The middle vine in each replicate was used for data collection.

Berry sampling and analyses
Berry samples were collected from each replicate at harvest. Samples were taken by removing berries from the top, middle, and bottom of each cluster on the vine (approximately 100 berries removed per vine). Fruit was transported to the laboratory immediately after sampling, and the berry number and fresh weight of each sample recorded. Fifty berries were ground in an electric blender until macerated, and the juice and pulp filtered through a paper towel. The filtrate was allowed to settle, and the juice used to determine juice soluble solids content and titratable acidity. Soluble solids were determined using a hand-held, temperature compensated refractometer. Acidity was determined by titrating a 5 ml aliquot of juice with 0.1 N NaOH to a pH endpoint of 8.2 using a automatic titrator. Berry length and diameter were determined by placing the remaining 50 berries of each sample in a specially designed trough so that their ends (berry length) or equators (berry diameter) gently touched. The cumulative length and diameter of the berries were recorded and used to calculate mean values for each parameter.
Fruit color development.
Ten randomly selected clusters on each vine were tagged prior to berry softening and used to monitor the rate of color development during ripening. The number of clusters with adequate color for harvest was recorded at regular intervals during fruit ripening. Clusters were considered harvestable when 80% of their surface has adequate color for harvest.

Anthocyanin analysis
Twenty-five berries were randomly selected from each vine at harvest, placed in sealed plastic bags, and stored at -15 °C until analyzed. Two, 10 mm skin disks were removed from the equator of each frozen berry using a cork borer and forceps. The disks were placed in clear polystyrene tubes containing 50 ml of acidified methanol (1% HCl by volume), and extracted in darkness at 25 °C. After 48 hours the samples were removed from the darkness, mixed for 5 sec using a vortex mixer, and settled for 30 min. The absorbance of a 5-ml aliquot from each sample was determined at 520 nm using a spectrophotometer.

Vine yield components
Clusters were removed from the vine at harvest, counted, and weighed. Clusters were rated cull if less than 80% of their surface had adequate color for harvest. Clusters were rated as cull if other defects, such as bunch rot, berry cracking, or excessive compactness, could not be easily removed from the cluster by trimming.

Postharvest evaluations
Fruit from each experiment (5 to 10 representative clusters per replicate, depending upon the experiment and availability of fruit) was packed in Styrofoam boxes and placed in storage at 32 F and 90% RH. SO₂ gas was applied weekly. Clusters were weighed at 10 to 14 day intervals to determine moisture loss rate. On each measurement date, 3 to 5 berries were removed from each cluster and used to determine berry firmness using the UC Fruit Firmness Tester.

RESULTS
Flame Seedless
GA x CPPU rate interactions on Flame Seedless table grapes are presented in Figure 1. The addition of 5 g/ac CPPU significantly increased the berry weight (berry size) of all GA treatments. However, these differences diminished as GA rate per acre was increased. Vines treated with 20 g/ac GA + 5 g/ac CPPU produced similar sized berries as vines receiving 60 g/ac GA alone. However, the addition of 5 g/ac CPPU in the treatment solution also significantly reduced berry color and the percentage of harvestable clusters (based on color) per vine for all GA rates (Figure 2). The reduction of berry color is a critical consideration regarding the use of CPPU on Flame Seedless table grapes.

Thompson Seedless
GA x CPPU rate interactions on Thompson Seedless table grapes are presented in Figure 3. The addition of 10 g/ac CPPU significantly increased the berry weight (berry size) of all GA treatments. As observed with Flame Seedless, however, these differences diminished as GA rate per acre was increased. Vines treated with 40 g/ac GA + 10 g/ac CPPU produced larger berries than vines receiving 80 g/ac GA alone.

Redglobe
CPPU x GA interactions on the berry weight of Redglobe are presented in Figure 4. GA treatments were most effective for improving berry size when applied at fruit set + 2 weeks. Combined applications of CPPU and GA also produced the greatest effects on berry size when applied at fruit set + 2 weeks. In contrast, CPPU treatments were most effective when applied at berry set. Although CPPU slowed color
development, the total number of clusters removed from each treatment by the completion of the harvest period was similar (Figure 5). While CPPU did not improve berry firmness when applied alone, the combined application of CPPU + GA at fruit set + 2 weeks significantly improved berry firmness compared to the control (Figure 6). The combined treatment of CPPU + GA also improved capstem removal force (Figure 7), but had no effect on the rate of fruit water loss during storage (Figure 8).

Crimson Seedless
The effects of CPPU on the berry growth of Crimson Seedless are presented in Figure 9. While all rates of CPPU improved berry weight compared to the control, and applications performed at fruit set generally provided similar efficacy as applications performed following fruit set. In contrast to other seedless cultivars, CPPU seems equally effective for improving berry size compared to GA and there appears to be no synergistic effects of combining the two materials (Figure 10). In addition, the total number of clusters removed from each treatment by the completion of the harvest period was similar (Figure 11).

Autumn Royal
The effects of CPPU and GA treatment combinations on the berry growth of Autumn Royal are presented in Figure 12. None of the chemical treatments included in the trial significantly increased berry weight compared to the untreated control. The portion of harvestable fruit (based on color) removed from each treatment was similar (Figure 13).

CONCLUSIONS
• CPPU increases the berry size of seedless cultivars an additional 5 to 20% compared to the use of GA alone. The berry size of most seedless cultivars can be increased 30% or more when CPPU is applied alone. CPPU increases the berry size of the seeded cultivar Redglobe 5 to 10% when applied alone, but typically does not improve berry size when applied in combination with GA. CPPU changes the shape of cylindrical berries (ex: Thompson Seedless) by increasing their diameter:length ratio. Optimum rates for berry sizing appear to range between 5 and 6 g/ac for most seedless cultivars.

• CPPU applied at berry set delays fruit maturity (sugar accumulation) up to two weeks, depending upon cultivar and rate. Titratable acidity, berry firmness and capstem removal force are increased. CPPU also reduces the rate of skin ambering or yellowing on Thompson Seedless and Princess. Rates ranging between 8 and 10 g/ac at fruit set + 2 weeks are suggested for this purpose.

• CPPU slows the accumulation of pigment in the skin, and can also reduce the percentage of clusters with adequate color for harvest. Ethrel is only partially effective for overcoming these detrimental effects on color development. Flame Seedless is the most sensitive cultivar. Redglobe is intermediate, and Crimson Seedless appears to be the least affected. CPPU is not generally recommended for Flame Seedless at this time. Rates ranging between 6 and 8 g/ac at fruit set + 2 weeks are suggested for improving the berry firmness of Redglobe, while rates between 5 and 6 g/ac at fruit set are suggested for experimentation and observation on Crimson Seedless.
Flame Seedless

Harvestable clusters (%)

GA$_3$ (g/AC)

- CPPU

+ CPPU (5g/ac)
Thompson Seedless

Berry weight (g)

0 20 40 60 80
GA₃ (g/ac)

+CPPU (10g/ac)

-CPPU
Capstem removal force (g)

- Control
- 40ppm GA$_3$ dip @ FS + 2 weeks
- 40ppm GA$_3$ + 8ppm CPPU dip @ FS + 2 weeks
- 8ppm CPPU (WVS) @ FS + 2 weeks

Redglobe

Force values:
- Control: 1021 g
- 40ppm GA$_3$ dip: 1054 g
- 40ppm GA$_3$ + 8ppm CPPU dip: 1112 g
- 8ppm CPPU (WVS): 1059 g
Redglobe
- ○ Control
- ▽ 40 ppm GA₃ dip @ FS + 2 weeks
- □ 40 ppm GA₃ + 8 ppm CPPU dip @ FS + 2 weeks
- ◇ 8 ppm CPPU (WVS) @ FS + 2 weeks

Water loss (%)

Days in storage
Crimson Seedless

Berry weight (g)

Control | 4  | 8  | 12  | 4  | 8  | 12
---------|----|----|-----|----|----|-----
Fruit set| 6.1| 6.9| 7.1 | 7.3| 7.0| 6.9 | 6.7
Fruit set + 2 weeks |
Crimson Seedless

Harvestable clusters (%)

- Control: 84%
- 4 gl/acz GA3: 82%
- 4 gl/acz CPPU: 79%
- 4 gl/acz GA3 + 2 weeks: 81%
- 4 gl/acz CPPU: 77%
- 4 gl/acz CPPU: 78%

Legend:
- White: Fruit set
- Shaded: Fruit set + 2 weeks
Berry weight (g)

Control

8 g/ac GA3

8 g/ac CPPU

8 g GA3 + 8 g/ac CPPU

Autumn Royal

Fruit set + 2 weeks