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BARD

FINAL REPORT

PROJECT NO. IS-2710-96

**The Physiological Bases for Impaired
Photosynthetic Performance of Chilling-
Sensitive Fruit Trees**

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Project Title: The Physiological Bases for Impaired Photosynthetic Performance of
Chilling-Sensitive Fruit Trees

Investigators

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Institutions

ARO, The Volcani Center
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USDA/ARS
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Keywords *not* appearing in the title and in order of importance. Avoid abbreviations.

Abbreviations commonly used in the report, in alphabetical order:

Budget: IS: \$206,000

US: \$94,000

Total: \$300,000

Signature
Principal Investigator

Signature
Authorizing Official, Principal Institution

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Publication Summary (numbers)

	Joint IS/US authorship	US Authors only	Israeli Authors only	Total
Refereed (published, in press, accepted)	1		2	3
Submitted, in review, in preparation	1			1
Invited review papers		1		1
Book chapters				
Books				
Master theses				
Ph.D. theses				
Abstracts	5		4	9
Not refereed (proceedings, reports, etc.)		1	2	3

Postdoctoral Training: List the names and social security/identity numbers of all postdocs who received more than 50% of their funding by the grant.

Damian Allen (346-94-4067)

Cooperation Summary (numbers)

	>From US to Israel	From Israel to US	Together, elsewhere	Total
Short Visits & Meetings	3	1	2	6
Longer Visits (Sabbaticals)				

Description of Cooperation:

Visit by YS to Urbana for joint discussions (year one), and DRO reciprocated the visit in year two. Three week visits by Damian Allen, the BARD supported post-doc in the US lab, in Bet-Dagan, to conduct collaborative experiments using sophisticated gas-exchange equipment under field conditions in both year 2 and year 3. To allow this high level of cooperation budgeted travel funds were augmented by non-BARD sources.

Patent Summary (numbers)

	Israeli inventor (s) only	US inventor (s) only	Joint IS/US inventors	Total
Submitted				
Issued (allowed)				
Licensed				

Abstract

Chilling-sensitivity is an important agricultural problem in both the U.S. and Israel. Most research attention has focused so far on herbaceous crop plants, even though the problem is also acute in the fruit tree industry. Under BARD funding we made substantial progress in identifying the mechanisms involved in the disruption of photosynthesis following a chill in mango. Our investigation with fruit trees has been substantially accelerated by drawing on our knowledge and experience with herbaceous crops. The four original research objectives, focused on discovering the underlying mechanisms of chill-induced inhibition of photosynthesis in fruit trees, and the main achievements are listed below.

[1] Separating stomatal from non-stomatal components of chilling on photosynthesis in fruit trees. We found evidence that the dark chill-induced inhibition of photosynthesis in mango was a combination of both stomatal and mesophyll components.

[2] Differentiating photodamage from light-induced photoprotection of photosystem II (PSII). Dark chilling exacerbate high light photoinhibition, as a result of primary inhibition in the carbon reduction cycle. Nevertheless, in Israeli orchards we observed chronic photoinhibition of PSII photochemistry in the winter. This photodamage was reversible over a few days if sunlight was attenuated with filters or night temperature rose. Practical implications of this finding deserve further investment. Additional achievement was the development of a new biophysical tool to study macro-structural changes of LHCII particles in intact, attached leaves.

[3] Determine the role of oxidative stress in the dark-chilling-induced inhibition, with emphasis on oxygen radical scavenging, lipid peroxidation and redox-controlled carbon-cycle enzymes. We found an increase in lipid peroxidation following a dark chill, and partial protective effects of an antioxidant. However, the photoinhibition observed in mango orchards in Israel during the winter did not appear to be a general oxidative stress.

[4] Investigate whether chilling interferes with the diurnal and circadian rhythm of gene expression of key photosynthetic proteins as has been shown for chilling-sensitive crop plants.

The results indicated that most of the circadian rhythm in photosynthesis was due to reduced leaf internal CO₂ concentrations during the subjective night, as a result of rhythmic stomatal closure. Chilling-induced interference with circadian timing in mango, does not play the central role in chilling inhibition of photosynthesis that has previously been demonstrated in certain chilling-sensitive herbaceous plants. Practical implications of the research achievements are feasible, but require few more years of research.

Achievements

Significance of main scientific achievements or innovations.

[1] *Separating stomatal from non-stomatal components of chilling on photosynthesis in fruit trees.* Chilling the aerial parts of potted mango plants overnight, produced no significant inhibition of photosynthesis during the first hours after rewarming on the following day under controlled conditions, or under near-field conditions in Israel. By midday, however, there was a substantial decline in leaf light-saturated CO₂ assimilation and stomatal conductance. The midday decline in photosynthesis, following a 7°C night, occurred concurrently with a rise in stomatal limitation of *A* and a decline in Rubisco activity. These factors demonstrate that the dark chill-induced inhibition of photosynthesis in mango, was a combination of both stomatal and mesophyll components.

[2] *Differentiating photodamage from light-induced photoprotection of photosystem II (PSII).*

As previously shown for herbaceous crops, such as tomato, the inhibition of CO₂ assimilation in mango leaves following a cold night, was not due primarily to photoinhibition (i.e., photodamage or light-induced down regulation of PSII). The inhibition of photosynthesis was not accompanied by any significant reductions in the dark-adapted maximum, or light-adapted steady-state quantum efficiencies of PSII electron transport. Dark chilling did exacerbate high light (1800 $\mu\text{mol m}^{-2} \text{s}^{-1}$ PPFD) photoinhibition, but this was likely the result of a primary inhibition in the carbon reduction cycle. Nevertheless, in Israeli orchards we observed necrosis of mango leaves in the field following a cold night, which was most apparent on branches exposed to the highest sunlight intensities. Chronic photoinhibition of PSII photochemistry was observed in the winter in Israeli mango orchards (manuscript in preparation). This photodamage was reversible over a few days if sunlight was attenuated with filters or night temperature rose. This photoinhibition cannot fully account for the low photosynthesis in these leaves during the winter, however. Unlike PSII photoinhibition, substantial winter declines in CO₂ assimilation could not be ameliorated by transient warmer ambient temperatures or shading of sunlight. The protection from photoinhibition that shading provided in the orchard suggests that this may be a useful strategy in protecting mango trees during the winter. Further work is underway testing this management practice for protection of mango fruit yield in Israel.

A new, non-destructive biophysical tool to study the macro-organization of LHCII was developed during this project. This novel approach, which is based on circularly polarized luminescence (CPL) of chlorophyll, is not directly related to photoinhibition, but rather to slower

responses of PSII reaction centers to environmental changes. Initial results on mango leaves and chill responses indicated the potential of this approach to provide a new insight into the processes under study.

[3] Determine the role of oxidative stress in the dark-chilling-induced inhibition, with emphasis on oxygen radical scavenging, lipid peroxidation and redox-controlled carbon-cycle enzymes.

We obtained very convincing data demonstrating an increase in lipid peroxidation following a dark chill. An additional, very intriguing finding was that the lipid peroxidation could be prevented by foliar application of antioxidants prior to the chill. However, the photoinhibition observed in mango orchards in Israel during the winter did not appear to be general oxidative stress, as lipid peroxidation did not decline during 17 days of shading, as the leaves recovered from photoinhibition.

[4] Investigate whether chilling interferes with the diurnal and circadian rhythm of gene expression of key photosynthetic proteins as has been shown for chilling-sensitive crop plants.

Mango leaves exhibit very strong rhythms in CO₂ assimilation with an interval of *ca.* 24 h, even when maintained under constant conditions of light, leaf temperature, humidity and CO₂ concentration. These circadian rhythms in photosynthesis (*ca.* 35%) were synchronized with a larger (*ca.* 65%) rhythm in stomatal conductance and consequently with a rhythm in the intercellular CO₂ concentration. This synchronization suggests that most of the circadian rhythm in photosynthesis was due to reduced leaf internal CO₂ concentrations during the subjective night, as a result of rhythmic stomatal closure. It has not been possible to determine whether there is also a smaller circadian rhythm in mesophyll photosynthesis that is independent of the stomatal rhythm. However, it is clear that no rhythmic changes in the rate of PSII electron transport occur in mango. An overnight dark chill at 10°C, immediately prior to the free-running period of continuous light, while inhibiting CO₂ assimilation and stomatal conductance, did not upset the circadian rise in these parameters during the subsequent subjective day and night. The maintenance of a circadian pattern after a dark chill indicates that a low temperature disruption of the circadian rhythm in stomatal opening, was not the cause of the midday rise in stomatal limitation and depression of photosynthesis. This does not exclude however, the possibility that an independent, smaller, mesophyll photosynthetic rhythm was impacted. It is important to appreciate that a central conclusion from this work is that chilling induced interference with circadian timing in mango, does not play the central role in chilling inhibition of photosynthesis that has been demonstrated in certain chilling-sensitive herbaceous plants.

Potential for application:

Medium term, depending on continuation of the research.

Agricultural and/or economic impacts of the research findings

The research findings, including the shading effects, the involvement of peroxidative responses, and the effects of foliar application of antioxidants might have practical/economical impacts on the performance and profitability of chill-sensitive fruit trees in many parts of the world, Israel and the US included. Turning the potential into practice would require few more years of research.

Details of cooperation

The collaboration is based on our mutual interest in chilling and high light effects on the photosynthetic performance of agricultural crops. We exchanged and discussed results frequently via the internet and by fax, collaboratively planning new experiments based on our results as they emerged. We have had a visit by YS to Urbana for joint discussions (year one), and DRØ reciprocated the visit in year two. Travel funds supported two 3-week stays by Damian Allen, the BARD supported post-doc in the US lab, in Bet-Dagan, to conduct collaborative experiments. The first visit (year 2) facilitated the use of sophisticated gas-exchange equipment under field conditions to enable validation of controlled environment studies in Urbana and resulted in significant aspects of the data contained in a joint publication (Allen *et al.*, 2000). The relevance of this work on potted trees to mango orchards in Israel was tested when DA returned to Bet-Dagan during the winter of year 3. These studies were substantially extended after DA's visit and a joint manuscript arising from this work is in preparation. Clearly, the project objectives were promoted as a result of this cooperation.

List of Reviewed Publications

- Allen DJ, Ratner K, Giller YE, Gussakovsky EE, Shahak Y, Ort DR (2000) An overnight chill induces a delayed inhibition of photosynthesis at midday in mango (*Mangifera indica* L.). *J Exp Bot* 51: 1893-1902
- Gussakovsky EE, Shahak Y, van Amerongen H and Barzda V (2000) Circular polarized luminescence reflects the macroorganization of grana in pea chloroplasts. *Photosynth Res* 65: 83-92.
- Allen DJ and Ort DR (2001) Impacts of chilling temperatures on photosynthesis in warm-climate plants. *Trends in Plant Science* 6: 37-42.
- Michaeli R, Philosoph-Hadas S, Riov J, Shahak Y, Ratner K. and Meir S. (2001) Chilling-induced leaf abscission of *Ixora coccinea* plants: III. Enhancement by high light via increased oxidative processes. *Physiol Plantarum*, in press.