Effect of Calcium and Boron Nutrition on Grey Mould of Capsicum (*Capsicum annuum* L.) and Fruit Quality

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Abstract

Capsicum (*Capsicum annuum* L.) is mostly cultivated in humid and warm conditions, which increases disease development, particularly grey mould caused by *Botrytis cinerea*. Infection of capsicum fruit by *B. cinerea* often occurs preharvest but symptoms of grey mould are not usually visible until after harvest making the pathogen difficult to control. Appropriate fertilisation that ensures calcium (Ca) and boron (B) is sufficient in plant tissues, especially in fruit, has been suggested as an alternative to fungicides for disease management. This research studied the infection pathway of *B. cinerea* and the effect of Ca and B on grey mould development and quality of fruit in two capsicum cultivars (cv. Aries and cv. Papri Queen).

Botrytis cinerea infected capsicum preharvest and flowers often died when inoculated at anthesis. The number of dead flowers increased when inoculum concentration increased. The extent of grey mould development on fruit inoculated preharvest was not affected by timing of inoculation [at anthesis, 3 days after anthesis (DAA) or 6 DAA], but was dependent on inoculum concentration and cultivar. When capsicum fruit were inoculated after harvest, grey mould developed most rapidly in red (R) fruit from cv. Aries and breaker red (BR) fruit from cv. Papri Queen. An inoculation of 10^6 conidia mL^{-1} caused more disease on fruit than 10^4 or 10^5 conidia mL^{-1}. Cv. Aries was more susceptible to *B. cinerea* than cv. Papri Queen regardless of whether inoculation occurred before or after harvest.

The effect of both soil and foliar application of boron (B), at different concentrations, on grey mould development and fruit quality of capsicum was examined. Preharvest B application, from transplanting to harvest when fruit were mature and red, using 0.05 or 0.1 mM H_3BO_3 via soil amendment or 2.0 or 7.0 mM H_3BO_3 as a foliar spray increased B concentration in leaves and fruit of both cultivars. However, soil application was more effective than foliar application in increasing B concentration in plant tissues. Foliar application of B at low concentrations (0.025 or 0.075 mM H_3BO_3) did not increase B concentration in plant tissue. Increasing B concentration in leaf and fruit tissue reduced grey mould

development on fruit inoculated with *B. cinerea* preharvest compared to the control, but did not affect grey mould development on red fruit inoculated with *B. cinerea* postharvest. Preharvest soil application of B increased shelf life of fruit, but did not affect quality of fruit including water content, firmness, total soluble solid content (TSSC) and titratable acidity (TA) at harvest or during storage. Symptoms of B toxicity were observed on leaves from plants that received high B concentration (0.1 mM H_3BO_3) in the soil, but no effect was observed on fruit.

Preharvest application of calcium (Ca) via soil amendment [1.5, 4.0 or 8.0 mM $Ca(NO_3)_2$] or as a foliar spray [0.5 or 1.0 % w/v mM $Ca(NO_3)_2$] increased Ca concentration in leaves, but did not increase Ca concentration in fruit, regardless of cultivar. Soil Ca application appeared to increase Ca concentration in leaf tissue more effectively than the Ca foliar spray. Ca concentration in leaf tissue from cv. Aries was significantly higher than in leaf tissue from cv. Papri Queen when plants received the same amount of Ca, regardless of application method. Ca treatment did not affect quality of fruit at harvest or during storage. Preharvest application of Ca reduced grey mould development on fruit that had been inoculated with *B. cinerea* preharvest, but did not reduce grey mould in fruit inoculated postharvest. Symptoms of Ca deficiency were observed on plants that received no Ca or low Ca concentration [1.5 mM $Ca(NO_3)_2$] from transplant to fruiting.

Dipping and vacuum infiltration with calcium chloride ($CaCl_2.2H_2O$) did not increase Ca concentration in flesh after treatment, but vacuum infiltration did increase Ca concentration in flesh after 10 days of cool storage (10°C). Ca treatment after harvest did reduce grey mould development on fruit, but did not affect the quality of fruit during storage. A directly inhibitory effect of Ca on fungal growth was responsible for reducing grey mould development on fruit.

In conclusion, capsicum was most sensitive to infection by *B. cinerea* at anthesis and high inoculum concentrations caused a greater disease incidence in capsicum fruit, regardless of whether inoculation occurred preharvest or after harvest. Reducing inoculum concentration, especially during flowering, is therefore recommended to reduce losses in capsicum. Preharvest application of Ca or B

may be used as an alternative method to reduce grey mould on capsicum fruit, but they had no effect on fruit quality. Postharvest application of Ca could also be recommended for cv. Aries fruit before or during storage for controlling grey mould on fruit. Findings in this research may therefore provide basic knowledge for management of *B. cinerea* in the capsicum industry.

Declaration

I certify that this work contains no material which has been accepted for the award of any other degree or diploma in my name, in any university or other tertiary institution and, to the best of my knowledge and belief, contains no material previously published or written by another person, except where due reference has been made in the text. In addition, I certify that no part of this work will, in the future, be used in a submission in my name, for any other degree or diploma in any university or other tertiary institution without the prior approval of the University of Adelaide and where applicable, any partner institution responsible for the joint-award of this degree.

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Abbreviation

Abbreviation	Full term
ANOVA	Analysis of Variance
AOAC	Association of Official Analytical Communities
ASTA	American Spice Trade Association
B	boron
BR	breaker red
°Bx	°Brix
°C	Degrees Celsius
Ca	calcium
$CaCl_2$	calcium chloride
$Ca(NO_3)_2$	calcium nitrate
$CuSO_4$	copper sulphate
cv.	cultivar
DAA	days after anthesis
DAH	days after harvest
DPI	days post-inoculation
DG	deep green
DW	dry weight
EDTA	ethylenediaminetetraacetic acid
et al.	and others
e.g.	for example
FW	fresh weight
FAO	Food and Agricultural Organisation
Fe^{3+}-EDTA	Ethylenediaminetetraacetic acid iron (III)
Fig	Figure
g	gram
h	hour
H_3BO_3	boric acid
ICP-OES	Inductively Coupled Plasma Optical Emission Spectrometer
kg	kilogram

kgf	kilogram force
kGy	kiloGray
KCl	potassium chloride
KNO_3	potassium nitrate
KH_2PO_4	potassium dihydrogen orthophosphate
KOH	potassium hydroxide
K_2SO_4	potassium sulphate
L	litre
LSD	Least Significant Difference
$MgSO_4$	magnesium sulphate
$MnSO_4$	manganese sulphate
mg	milligram
min	minute
mL	millilitre
mm	millimetre
mm^2	square millimetre
mt	million tones
N	Newton
NaH_2PO_4	sodium phosphate dibasic
NH_4NO_3	ammonium nitrate
$(NH_4)_6Mo_7O_{24}$	ammonium molybdate tetrahydrate
PDA	Potato dextrose agar
PE	pectinesterase
PG	polygalacturonase
pH	power of hydrogen (negative log of H^+ concentration)
ppm	parts per million
RH	relative humidity
RO	reserve osmosis
SE	standard error
sec	seconds
TSSC	total soluble solid content
TA	titratable acidity

UC	University of California
UV	ultraviolet
$ZnSO_4$	zinc sulphate
w/v	weight by volume
µM	micromoles per litre
µL	microlitre
%	percentage