EFFECT OF NITRIC OXIDE ON METABOLISM OF FRESH-CUT APPLES AND LETTUCES IN RELATION TO SURFACE BROWNING

By

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DECLARATION

I hereby certify that the work embodied in this thesis is the result of original research and has not been submitted for a higher degree to any other University or Institution

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PUBLICATIONS

Part of this thesis have been published and presented in conference.

Journal paper

1. Huque, R., Wills, R. B. H. and Golding, J. B. (2011). Nitric oxide inhibits cut-surface browning in four lettuce types. *Journal of Horticultural Science & Biotechnology* 86 (2) , 97–100

Conference papers

1. Roksana Huque, John Golding and Ron Wills. (2009). Metabolic effects of nitric oxide on apple slices. Fifth International Symposium on Managing Quality in Chains in collaboration with the Australasian Postharvest Horticultural Conference, Napier, New Zealand. Programme and Abstract book – S13.2, p 83.

LIST OF ABBREVIATIONS

AAP	ascorbic acid-2-phospate
AATP	ascorbic acid-3-triphosphate
ABA	abscisic acid
ACC	1-aminocyclopropane-1-carboxylic acid
DETA	diethylenetriamine
DETANO	diethylenetriamine nitric oxide
DNA	deoxyribonucleic acid
EDRF	endothelium-derived relaxing factor
EDTA	ethylenediamine tetraacetic acid
FAD	flavin adenine dinucleotide
FMN	flavin mononucleotide
$HO^{\bullet}$	hydroxyl radical
H_2O_2	hydrogen peroxide
HR	hypersensitivity response
IFPA	International Fresh-cut Produce Association
Inos	inducible NOS
LOX	lipoxygenase
mRNA	messenger RNA
MDA	Malondialdehyde
NiR	nitrite reductase
NO	nitric oxide
$NO^{\bullet}$	free radical nitric oxide
NO^{+}	nitrosonium cation
NO^{-}	nitroxyl anion
NO_2	nitrogen dioxide, nitrogen peroxide
N_2O	nitrous oxide
NOS	nitric oxide synthase
NR	nitrate reductase

1-MCP	1-methylcyclopropene
O_2	oxygen
$O_2^{\bullet-}$	superoxide anion
$OONO^-$	peroxynitrite ion
PAL	phenylalanine ammonia lyase
PBN	N-*tert*-butyl-α-phenylnitrone
POD	peroxidase
PPO	polyphenol oxidase
ROS	reactive oxygen species
RNA	ribonucleic acid
SNAP	S-nitroso-N-acetylpenicillamine
Sin^{-1}	3-morpholinosyl-nonomone
SNP	sodium nitroprusside

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ABSTRACT

Surface browning is an important cause of deterioration of fresh-cut produce during postharvest handling. Nitric oxide (NO) has recently been found to delay the onset of surface browning in fresh-cut apples and iceberg lettuce.

Effectiveness of NO applied as NO gas and the NO donor compound 2,2'-(hydroxynitrosohydrazino)-bisethanamine (diethylenetriamine nitric oxide, DETANO) dissolved in phosphate buffer (pH 6.5) solution on the physiological parameters of ethylene production, respiration and water loss, and biochemical parameters of total phenol content, PPO activity, ion leakage and lipid peroxide level were investigated. Granny Smith apple slices treated with 10 $\mu l.l^{-1}$ NO gas and 10 $mg.l^{-1}$ DETANO showed delayed development of surface browning and also resulted in a lower total phenol content, inhibition of PPO activity, reduced ion leakage and reduced rate of respiration but had no significant effect on ethylene production, water loss or lipid peroxide level as measured by malondialdehyde and hydrogen peroxide levels. The two control treatments of phosphate buffer (pH 6.5) and water dips also had significant effects on all parameters compared to untreated slices. The relative effectiveness treatments on postharvest life, apple physiology and biochemistry was DETANO > NO gas > phosphate buffer > water > untreated. The NO donors, sodium nitroprusside (SNP) and Piloty's acid dissolved in water also inhibited development of surface browning but were not as effective as DETANO.

Apple slices dipped in chlorogenic acid dissolved in water showed surface browning within an hour of treatment. Dipping in DETANO solution negated the effect of chlorogenic acid whether applied before or after dipping in chlorogenic acid solution while the buffer and NO

gas were also effective. A UV-scan of chlorogenic acid dissolved in water showed a marked decreased in absorbance over the eight day storage period suggesting that chlorogenic acid was oxidised by aerial oxygen. The addition of NO gas and DETANO accelerated the loss of chlorogenic acid.

It is suggested that browning development of fresh-cut produce can be inhibited by action taken soon after cutting. The concentration of phenols on the surface could be the rate limiting steps in browning development with non-enzymatic oxidation of phenols by atmospheric oxygen a contributor to browning.

NO gas, DETANO and SNP inhibited the surface browning of green oak lettuce slices. The optimum concentration of DETANO or SNP (500 $mg.l^{-1}$) and NO gas (100 $\mu l.l^{-1}$) resulted in approximately 60% and 30% increase in postharvest life over untreated slices respectively.