ANTIMICROBIAL FACTOR FROM GRAPES

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Abstract

Food-borne pathogens and in particular the emergence of new food-borne bacterial pathogens, which includes *Listeria monocytogenes*, are a significant threat to public health. This thesis investigated a commercial red grape juice, derived from *Vitis vinifera* variety Ribier grapes, with a strong inhibitory action against *Listeria monocytogenes*. The commercial grape juice caused a 6-log reduction in *Listeria monocytogenes* numbers within 10 minutes exposure to the grape juice. The inhibitory effect was specific to all *Listeria* species. The grape juice was bactericidal and inhibitory at pH $\leq$ 5.0. The addition of protein (BSA), PVPP, and metal ions (Fe^{3+} and Mg^{2+}) suppressed the antilisterial activity of the grape juice. The antilisterial factor in the commercial grape juice was identified as pigmented condensed tannin (proanthocyanidin oligomers). The antilisterial pigmented condensed tannin in the commercial grape juice was likely derived from the skin of Ribier grapes. Fractionation of the commercial grape juice pigmented condensed tannin by molecular weight revealed that the antilisterial activity was distributed over a range (mean degree of polymerisation 2.8 through 8.5) of tannin fractions. Condensed tannin fraction (G) had a mean degree of polymerisation of 5.9 and was found to have the strongest antilisterial activity. The molecular weight of oligomers identified in fraction G ranged from MW 867 to MW 2905. Fraction G contained the highest percentage of polymeric colour with 90% the pigmented moieties (anthocyanins) in the fraction incorporated into the polymer structure. Fraction G was comprised of the flavonoid subunits catechin, epicatechin, epigallocatechin, and epicatechin-3-*O*-gallate.

Ribier grape seed condensed tannin had strong antilisterial activity. The antilisterial condensed tannin derived from Ribier grape seed was comprised of the flavonoid subunits catechin, epicatechin, and epicatechin-3-*O*-gallate and increasing antilisterial activity was associated with increasing mean degree of polymerisation of the tannin polymers. The efficacy of the pigmented condensed fraction of the commercial grape juice and Ribier grape seed condensed tannin as natural food preservatives were assessed. In the presence of *Listeria monocytogenes* contaminated cabbage the inhibitory activity of the pigmented condensed tannin from commercial grape juice was suppressed. The antilisterial activity of the Ribier grape seed condensed tannin was reduced but not eliminated when *Listeria monocytogenes* was associated with cabbage. The grape seed tannin may be effective at eliminating *Listeria monocytogenes* from food.

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API	Analytical Profile Index
AWRI	Australian Wine Research Institute
BHI	Brain Heart Infusion broth
BSA	Bovine Serum Albumin
CFU	Colony Forming Units
ESI-MS	Electrospray Ionising Mass Spectroscopy
FTIR	Fourier Transform Infrared Spectroscopy
HPLC	High Performance Liquid Chromatography
IES-MS	Infusion Electrospray Spectroscopy Mass Spectroscopy
LC-MS	Liquid Chromatography Mass Spectroscopy
mDP	Mean Degree of Polymerisation
MGJ	Model Grape Juice solution
MIC	Minimum Inhibitory Concentration
MPN	Most Probable Number
MW	Molecular Weight
MWCO	Molecular Weight Cut Off
MWS	Model Wine Solution
PVPP	Polyvinylpolypyrrolidone
OD	Optical Density
TEM	Transmission Electron Microscopy
TSA	Tryptic Soy Agar
TSB	Tryptic Soy Broth