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**Isolation, Characterisation and Functional Properties of
Pectin from Gold Kiwifruit (*Actinidia chinensis* cv.
Hort16A)**



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Abstract

This research was concerned with the isolation, physicochemical characterisation and functional properties of pectin from gold kiwifruit cv. Hort16A. The process of extracting pectin from gold kiwifruit was developed by evaluating three different techniques (acid, water and enzyme), four different conditions (time, temperature, puree to solvent ratio and enzyme concentration) and fruit of two different maturities (early-season and main-season fruit). The effects of the extraction techniques and conditions on the physicochemical properties and functional properties of pectin were studied in detail. The effects of the extraction techniques and the fruit maturity on the functional properties of pectin were also investigated. The chemical compositions, physical features and rheological behaviours of the extracted pectins were determined, underlining the importance of these features to the functional properties of gold kiwifruit pectin.

The total non-starch polysaccharide composition, sugar composition, protein and ash contents, degree of esterification (DE) and molecular properties (weight-average molecular weight (M_w), polydispersity index and root mean square radius) of pectin are influenced by the extraction techniques, the extraction conditions and the degree of maturity. Pectin from early-harvested fruit (EHF) (less mature fruit) is more difficult to extract than pectin from main-harvested fruit (MHF) (more mature fruit) when using extraction methods that have been developed for MHF. This is probably because the cell wall network of less mature fruit is more compact/dense and the pectin could still be insoluble, whereas the cell wall network of more mature fruit is less tightly bound because of physiological changes during maturation. Purified pectin from EHF is characterised by a lower yield (1.52 versus 3.64% w/w), a higher DE (90 versus 84%), a lower galacturonic acid (GalA) content (40.21 versus 55.50% w/w), more branching chains (a side chain every 47–57 GalA residues versus every 50–97 GalA residues), a higher protein content (25.94 versus 13.82% w/w), a lower M_w (9.7×10^5 versus 2.52×10^6 g/mol) and a higher viscosity than purified pectin from MHF. Overall, the physicochemical properties of gold kiwifruit pectin are consistent with its rheological behaviour.

Pectins extracted from fruit of different maturities and using different methods have different physicochemical properties. MHF pectin extracted using a water method is characterised by an M_w of 3.75×10^6 g/mol, a GalA content of 51.87% w/w, a degree of branching of 50 and a DE of 84%. MHF pectin extracted using an enzymatic method has a lower M_w (1.65×10^6 g/mol), similar to that of enzyme-extracted EHF pectin (0.21×10^6 g/mol). In contrast, EHF pectin extracted using a water method has low M_w (1.03×10^6 g/mol) and GalA content (42.88%), suggesting that gold kiwifruit pectin with high M_w is recovered from more mature fruit.

The rheological properties of the extracted pectin are affected by its M_w and DE. Purified pectin extracted by enzymatic treatment from both EHF and MHF exhibits the lowest viscosity and M_w . Purified EHF pectin obtained by water treatment exhibits a higher viscosity even though the pectin is characterised by a lower M_w (1.03×10^6 g/mol) with a higher DE (90%) compared to acid-extracted EHF pectin (a higher M_w : 1.66×10^6 g/mol and lower DE: 88%). This is because of low electrostatic repulsion of high DE pectin, which reduces intra- and/or intermolecular distances, resulting in greater molecular association, and the greater degree of branching, which leads to more chain–chain associations, therefore exhibits higher viscosity. A similar trend is observed for MHF pectin. Water-extracted MHF pectin, with a high M_w (3.75×10^6 g/mol) and a low DE (84%), exhibits a lower viscosity than EHF pectin (a low M_w but a high DE, *i.e.* 90%). The viscoelastic properties of the pectin are consistent with its physical properties.

Gold kiwifruit pomace, a by-product from gold kiwifruit juice manufacture, may be a new source of pectin. The physicochemical properties of pomace pectin are quite different from those of whole fruit pectin. Purified pomace pectin is characterised by a higher GalA content (64–68% w/w) but a lower M_w (6.7 – 8.4×10^5 g/mol) and exhibits a lower viscosity.

In the work on the functional properties of gold kiwifruit pectin, its gelation was investigated and was compared with that of other commercial pectins (apple and citrus). Gold kiwifruit pectin, which is classified as a high methoxyl pectin (HMP), has “weak gel” properties and gels at high temperature. Because of low electrostatic repulsion among the

pectin chains, the gelation properties are not markedly affected by varying the gelation conditions such as pH and sugar concentration. However, increasing the concentration of pectin leads to an increase in the gel strength. The viscoelasticity of the gel is strongly influenced by the DE; the gel strength increases as the DE decreases, because high charge density (or low DE) pectins have numerous active sites with high probability for hydrogen bonding, and sufficient hydrophobic interactions, thus resulting in more stabilised molecular networks.

A common defect in acidified milk drinks (AMDs) is sedimentation caused by protein aggregation. HMP is commonly added to prevent this separation. In this part of the work, the influence of gold kiwifruit pectins (DE 84, 85 and 90%) on the stability of AMDs (10% w/w low heat skim milk powder) was evaluated and was compared with that of pectins from other sources (apple and citrus). The stabilities, in terms of serum separation, amount of adsorbed and non-adsorbed pectin, viscosity and particle size, were compared at different pectin concentrations (0.1–1.0% w/w). High M_w gold kiwifruit pectin (DE 84%) stabilises an AMD at lower concentrations (0.3% w/w) than other commercial HMPs. In addition, HMPs with very low charge density and very low M_w do not have the ability to stabilise AMDs at the pectin concentrations used in this study. These results suggest that M_w has a very important role in stabilising an AMD. In addition, the viscosity effect created by high M_w apparently supports the dispersion of casein–pectin complexes in the drink.

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List of Abbreviations

AMD	: Acidified Milk Drink
ANOVA	: Analysis of Variance
AOAC	: Association of Official Analytical Chemists
AR	: Arabinase
Ara	: Arabinose
CA	: Citric Acid
CCP	: Colloid Calcium Phosphate
CE	: Capillary Electrophoresis
CDTA	: Cyclohexane Diamino Tetra Acetic Acid
CMC	: Carboxymethylcellulose
CM	: Carboxymethylcellulase
Cv	: Cultivar
GalA	: Galacturonic Acid
Gal	: Galactose
GCP	: Grinsted Citrus Pectin
GDL	: Glucono- δ -Lactose
GKP	: Gold Kiwifruit Pectin
GLC	: Gas Liquid Chromatography
Glc	: Glucose
GOPOD	: Glucose Oxidase Peroxidase Colour Reagent
DF	: Dietary Fiber
DE	: Degree of Esterification
DLS	: Dynamic Light Scattering
DM	: Dry Matter
DMSO	: Dimethyl Sulfoxide
DMP	: 3-5-dimethylphenol
DRI	: Differential Refractive Index

Dwb	: Dry Weight Basis
EDTA	: Ethylene Diamino Tetra Acetic Acid
EH-GKP	: Early-Harvested Gold Kiwifruit
E-MHGKP	: Enzyme-Extracted Pectin from Main Harvested Fruit
Fuc	: Fucose
HMP	: High Methoxyl Pectin
HPLC	: High Performance Liquid Chromatography
IU	: International Unit
IEC	: Ion-Exchanged Chromatography
LA	: Amidated Low Ester Pectin
LC	: Conventional Low Ester Pectin
LHSMP	: Low Heat Skim Milk Powder
LMP	: Low Methoxyl Pectin
LS	: Light Scattering
MALLS	: Multi-Angle Laser Light Scattering
Man	: Mannose
M_w	: Molecular Weight
NI	: No Information
NSP	: Non-Starch Polysaccharide
PAW	: Phenol:Acetic:Acid:Water
PGalA	: Polygalacturonic Acid
PG	: Polygalacturonase
PME	: Pectin Methyl Esterase
PMEI	: Pectin Methyl Esterase Inhibitor
PS	: Pure Serum
Rha	: Rhamnose
RMS	: Root Mean Square
RS	: Resistant Starch
SEC	: Size-Exclusion Chromatography
SI	: System International
TPA	: Texture Profile Analysis

TSS	: Total Soluble Solid
USDA	: United State Department of Agriculture
UV	: Ultraviolet
XTH	: Xyloglucan Endotransglucosylase/Hydrolase
Xyl	: Xylose
W-EHGKP	: Water-Extracted Pectin from Early Harvested Fruit
W-MHGKP	: Water-Extracted Pectin from Main Harvested Fruit
WSP	: Water Soluble Polysaccharide
Wwb	: Wet Weight Basis