Preparation, characterisation and functionality of kafirin microparticles

by

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DECLARATION

I hereby declare that this thesis submitted at the University of Pretoria for the award of PhD degree is my work and has not been submitted by me for a degree at any other University or Institution of Higher Education.

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April 2008

ABSTRACT

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Janet Taylor

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Whilst working on a Masters degree on alternative solvents and extractants for the sorghum prolamin protein, kafirin, the author serendipitously found an ethanol-free method of making kafirin microparticles in dilute organic acid. Further, on drying a suspension of kafirin microparticles in dilute organic acid, a clear, transparent film was found to be formed. Microparticles from zein, the maize prolamin protein, have shown potential for food and pharmaceutical applications. Kafirin is more hydrophobic and less digestible than zein so it was hypothesised that it may form microparticles with superior properties. However, the structural and functional characteristics of kafirin microparticles and films made from them needed to be known before any potential applications could be exploited.

Kafirin microparticles were made by dissolution of kafirin in glacial acetic acid followed by precipitation on addition of water. They were characterized by Light microscopy (LM), Scanning Electron Microscopy (SEM) and Transmission Electron Microscopy (TEM) and were found to be mainly spherical, porous and between 1-10 μm in diameter. The kafirin microparticles had very large internal surface area due to the presence of many smooth walled holes or vacuoles of variable sizes, probably caused by entrapment of air during microparticle formation. Increasing the final acetic acid concentration resulted in kafirin microparticles of increased size, with an increasing number of internal holes. At 40% acetic acid the spherical microparticle structures completely disappeared and were replaced by an open matrix which resembled an expanded foam.

The kafirin microparticles were found to form very thin (<15 μm) free standing films and coatings. A minimum concentration of organic acid (10.8 percent) is required to form a cohesive kafirin microparticle film relative to the concentration of protein (1 percent for acetic acid). Some functional properties, e.g. smooth film surface properties, low water

vapour permeability (WVP) and low protein digestibility of these films are superior to those of similar conventionally cast kafirin films.

With the aim of exploiting the porous nature of kafirin microparticles for encapsulation of nutrient additives, several factors were examined for their influence on retarding protein digestibility. Retardation of digestibility of kafirin microparticles would allow controlled release of the encapsulated agent in the stomach and gastrointestinal tract. The importance of disulphide cross-linking and sorghum condensed tannin protein interactions were confirmed as major causal factors of the poor protein digestibility of sorghum. Gamma-kafirin was found to bind the most condensed tannins compared to the α-and β-kafirins, probably due to its high proline content. As expected, the protein digestibility of kafirin-tannin complexes was much lower than unbound kafirins. This seems to slow the biodegradation of kafirin films made with bound tannins.

The antioxidants, catechin and sorghum condensed tannins were encapsulated within kafirin microparticles and the antioxidant release profiles investigated under simulated gastric conditions. Over a period of four hours, catechin and condensed tannin encapsulated kafirin microparticles showed virtually no protein digestion but released approximately 70% and 50% respectively total antioxidant activity.

The mechanism for the formation of kafirin microparticles and films formed from them seems to involve controlled aggregation of kafirin molecules. Models for the formation of both were proposed based on an analogy with protein body formation and the potential ability of γ-kafirin to undergo a structural inversion exposing either hydrophilic or hydrophobic ends depending on the prevailing conditions.

Research into cross-linking by physical or chemical agents is needed before practical applications can be exploited. However, encapsulation of catechin and sorghum condensed tannins within kafirin microparticles seems to be an effective way to use the binding properties of polyphenols with protein to enhance potential health benefits by controlled release of antioxidant activity within the stomach and gastrointestinal tract.

DEDICATION

To my family, with love. Without you all I would not have succeeded.

'The outcome of any serious research can only be to make two questions grow where one question grew before'

Thorstein Veblen

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Think where man's glory most begins and ends, and say my glory was I had such friends
William Butler Yates

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