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Emulsifying Properties of Interfacial Components of Coconut Oil Bodies

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Abstract

Oil bodies are organelles in plant that store triacylglycerol (TAG) in plants. Some oil bodies exhibit remarkable physical and chemical stability to against coalescence and lipid oxidation due to their unique interfacial layer. These interfacial biomaterials present themselves as ideal materials for encapsulation and have potential applications in food, pharmaceutical and cosmetic formulations. This study compared the emulsifying properties of biomaterials obtained from coconut oil body membrane (COBM) and coconut skimmed milk extracts (CSME) and investigated the structure of resulting COBM and CSME emulsions.

Coconut oil body membrane (COBM) and coconut skimmed milk extracts (CSME) were extracted from freshly prepared coconut milk (around 20%, w/w, fat). The properties of CSME and COBM such as solubility, isoelectric pH and their ability to reduce the interfacial tension between water and soybean oil was characterized by tensionmeter. The CSME and COBM (0.2, 0.4, 0.6 and 0.8%, w/w, final protein concentration) were used for preparing soybean oil-in-water emulsions (20%, w/w) by microfluidizer. The physicochemical characteristics of the CSME and COBM emulsions at different pH (2-8) and NaCl (0-500 mM) conditions were investigated by using dynamic light scattering techniques and confocal laser scanning microscopy. Sodium dodecyl sulphate polyacrylamide electrophoresis (SDS-PAGE) was used to characterize the proteins composition of the 2 extracts (CSME and COBM) and the proteins composition of droplet interfacial proteins of CSME and COBM emulsions. To investigate the surface composition of CSME and COBM emulsions, the surface of the droplet of CSME and COBM emulsions was perturbed by the enzymes pepsin, trypsin and phospholipase A_2 (PLA_2), separately.

The SDS-PAGE analysis of two extracts showed distinct differences in the protein composition of CSME and COBM. The isoelectric points of CSME and COBM solutions were between pH 4 and 5. Both extracts lowered the interfacial tension between water and oil but the extent of decrease in surface tension was greater for COBM than that for CSME, indicating that COBM was more surface active than CSME.

The particle size of CSME emulsion decreased with an increase in protein concentration, while the effect of protein concentration on particle size was less pronounced in COBM emulsions. Compared with CSME emulsion, COBM emulsion had a smaller particle size with less degree of flocculation and was more stable during storage. These results suggest that COBM had the better emulsifying capacity than CSME. The CLSM images revealed that the droplet surface of CSME and COBM emulsions consisted of both protein and phospholipids. Both pepsin and phospholipase A_2 treatment of CSME and COBM emulsions lead to the coalescence, which indicates the possible droplet interfacial layer structures of CSME and COBM emulsions are similar, that both phospholipids and protein sequences with aromatic and hydrophobic residues present at the interface. The SDS-PAGE analysis of the droplet surface proteins of CSME and COBM emulsions revealed that not all proteins in CSME and COBM were adsorbed on the droplet surface of CSME and COBM emulsions. The surface protein composition of COBM emulsion was similar to that of natural coconut oil body.

The effect of pH on CSME and COBM emulsions revealed that the isoelectric points of both emulsions were close to each other (around pH 4.7 and 4.5 for COBM and CSME respectively). In addition, both COBM and CSME emulsions were stable at high pH (pH > pI) but had different behaviour below pI. While the COBM emulsion flocculated at pH near pI and the CSME emulsion showed coalescence at pH ≤ pI.

In the presence of salt, CSME and COBM emulsions were still negatively charged, even at 500 mM NaCl, indicating that 500 mM NaCl was unable to screen all the charges on the droplet surface. Flocculation occurred in COBM emulsion with the increase in NaCl concentration. No coalescence was observed in CSME emulsion at all given salt concentrations. This work shows that COBM emulsion has better stability against changes in pH than CSME emulsion.

Both COBM and CSME were successfully stabilized emulsions, which indicates that COBM and CSME may be suitable for use as a food emulsifier. COBM seems to be a better emulsifier material, since it can form emulsions with smaller droplet size and the flocculation occurred at low pH and high salt conditions may not be a major problem in some food, such as sauces and yoghurts.

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Table of Contents

List of Abbreviations

CCP1	Coconut cream protein (isoelectric precipitation)
CCP2	Coconut cream protein (freeze-thaw)
CLSM	Confocal laser scanning microscopy
COBM	Coconut oil body membrane
CSME	Coconut skimmed milk extracts
CSPC	Coconut skimmed milk protein concentrate
CSPI	Coconut skimmed milk protein isolate
$d_{4,3}$	Average volume-weighted diameter
$d_{3,2}$	Average surface-weighted diameter
FG	Fast Green
HLB	Hydrophilic-lipophilic balance
MFGM	Milk fat globular membrane
NR	Nile Red
OB	Optical density
pI	Isoelectric point
PLA_2	Phospholipase A2
POB	Protein of oil body
Rd-DHPE	Lissamine^{TM} rhodamine B
SDS	Sodium dodecyl sulfate
SDS-PAGE	Sodium dodecyl sulfate polyacrylamide electrophoresis
SGF	Simulated gastric fluid
SIF	Simulated intestinal fluid
TAG	Triacylglycerol
w/w	Weight/weight
w/v	Weight/volume
v/v	Volume/volume

List of Tables

List of Figures