

UNIVERSITI TEKNOLOGI MARA

**METHOD VALIDATION FOR
QUANTITATIVE ANALYSIS OF
CHOLESTEROL IN COOKED MEAT
PRODUCTS AND OPTIMISATION
OF SC-CO₂ EXTRACTION OF FAT
AND CHOLESTEROL FROM BEEF
FLOSS USING RESPONSE SURFACE
METHODOLOGY**

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MSc

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AUTHOR'S DECLARATION

I declare that the work in this thesis was carried out in accordance with the regulations of Universiti Teknologi MARA. It is original and is the results of my own work, unless otherwise indicated or acknowledged as referenced work. This thesis has not been submitted to any other academic institution or non-academic institution for any degree or qualification.

I, hereby, acknowledge that I have been supplied with the Academic Rules and Regulations for Post Graduate, Universiti Teknologi MARA, regulating the conduct of my study and research.

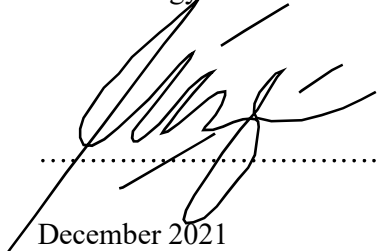
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ABSTRACT

The analytical methods for the determination of cholesterol in food has been extensively studied for years, especially on raw food products. The study aims to validate the method for cholesterol extraction from cooked meat products and its quantification by gas chromatography-flame ionization detector (GC-FID). This study showed that the limit of detection (LOD) and limit of quantification (LOQ) were 4.37 and 15 mg/100 g, respectively, showing high sensitivity of the GC-FID with the correlation coefficient (R^2) greater than 0.9905. The calibration curve exhibited good linearity over the tested cholesterol concentration ranging from 15 to 300 mg/100 g with R^2 of 0.9991. The validation for precision (e.g., repeatability and within-lab reproducibility) and trueness test was performed using cooked meat samples such as beef meatball, beef floss, and Quality Reference Material (QRM) made of dried beef. The results indicated that both repeatability and within-lab reproducibility were highly precise with acceptable relative standard deviations (RSD) ranging from 0.51 to 0.58% and within the acceptable Horrat values of 0.5 to 2.0 for all samples. The method was also found to be accurate, with 100.33%, 100.13% and 100.21% of cholesterol recovery from beef meatball, beef floss and QRM, respectively. This study indicated that the method was successfully validated and can be potentially adopted as an appropriate technique for determining cholesterol in cooked meat products. This validated method was then applied to determine the cholesterol content of untreated beef floss and Sc-CO₂-treated beef floss. Supercritical carbon dioxide (Sc-CO₂) was applied to extract fat and cholesterol from beef floss (BF). A response surface methodology (RSM) based on central composite design (CCD) was employed to optimize the extraction conditions of temperature (30 - 62 °C), pressure (7 - 35 MPa), and extraction time (0 - 40 min). The optimum conditions were estimated to be at 51.0 °C and 32.8 MPa for a duration of 32.7 min. Under such conditions, the percentage of fat and cholesterol reduction plus lightness of Sc-CO₂ treated BF (STBF) were 81.12%, 86.17%, and 57.60, respectively. There were no significant differences ($p > 0.05$) between experimental and predicted values, indicating the adequacy of the well-fitting models. Furthermore, the protein and ash content of STBF were increased significantly ($p < 0.05$) as a result of the extraction. This study indicated that mild Sc-CO₂ conditions can be potentially employed in maximizing the extraction of fat and cholesterol from BF.

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