

Chapter 1

Introduction

1.1 Background

Meat analogue are food products with similar nutritional and sensory qualities to meat products that use ingredients derived from sources such as plants, fungi, and microbial-derivatives (Kumar et al., 2019). Plant-based proteins are currently the most common ingredients used for meat analogue development with examples include isolated and textured soy protein, wheat gluten, and pea proteins (Kyriakopoulou et al., 2021). However, these plant protein sources need unfolding and cross-linking processes to turn their globular protein structure into a fibrous structure found in meat (Dinali et al., 2024). Another meat analogue alternative with the advantage of having a filamentous structure is mycoprotein. Mycoprotein is a protein source derived from fungi that is produced from agroindustrial waste fermentation (Finnigan et al., 2019). Fungi strains such as *Aspergillus oryzae* produce filamentous mycelium in the biomass which can mimic the functional and textural properties of fibrous structure from traditional meat (Gamarra-Castillo et al., 2022). Furthermore, mycoprotein has chitin and β -glucan as dietary fibers as well as more essential amino acids with lower concentration of anti-nutrition compounds compared to soy proteins which means higher protein digestibility (Montebello et al., 2025). Because of this, mycoprotein can be a suitable alternative in developing a meat analogue product with high nutritional value (Lee et al., 2026). However, application of mycoprotein into meatballs has not been fully investigated compared to sausage and patties.

Meatball is a processed meat product made from minced meat, commonly beef and fish, mixed with seasonings, binders, and fillers (Ahmadi et al., 2021). It has a round shape and allows for consumption of meat at cheaper cost which makes it a versatile and convenient food. Meatballs have a firm texture, strong savory flavor, meaty aroma, and cooked meat-like appearance, defining its appeal for meat and non-meat consumers (Su et al., 2025; Zor et al., 2025). Meat

analogues often come in meatball type products to attract more consumers (Andreani et al., 2023).

Currently, literature sources on the development of mycoprotein as meat analogue products such as meatball were limited to *Fusarium venenatum* and other strains while *Aspergillus oryzae* strain had not been fully explored yet (Shahid et al., 2024; Shahbazpour et al., 2021; Hashempour-Baltork et al., 2023; Li et al., 2025; Stephan et al., 2017). While mycoprotein has fungal filaments similar to fibrous proteins of meat, it lacks the ability to form a three-dimensional gel network through protein interactions (Colosimo et al., 2020). Thus, mycoprotein requires egg white albumin and hydrocolloids to form more cohesive gel networks that are essential for meatball processing (Muhedaner et al., 2025). Hydrocolloids such as xanthan gum, sodium alginate, and kappa-carrageenan have been found to improve the gel strength and network of meatball through thickening, gelling, and synergistic interactions with the protein (Arora et al., 2016; Zhang et al., 2023; Peñaranda et al., 2025; Pan et al., 2017).

A study by Majzoobi et al. (2017) used xanthan gum and kappa-carrageenan to improve the color, water holding capacity, and texture profile of meat-free sausage made from soy protein isolate and texturized soy protein. It was found that kappa-carrageenan at 0.3-1.5% concentration significantly increased the hardness and water holding capacity of the sausage. On the other hand, xanthan gum was found to improve the lightness in increasing concentrations to 1.5% compared to kappa-carrageenan. Because of this, xanthan gum was better used by combining with other hydrocolloids to complement the desirable effects. Sodium alginate is commonly used for its ion-induced gelation effect to simulate elasticity found in meat-based products (Guo et al., 2025; Lee et al., 2023). Another study by Zhang et al. (2020) found that addition of sodium alginate up to 1.0% to peanut protein meat analogue had improved elasticity with highest hardness reached in 0.5% concentration.

Other researches related to mycoprotein-based analogues are limited to specific strains such as *Fusarium venenatum* while studies on using *Aspergillus oryzae* strain for mycoprotein as meat analogue base still has not been expanded yet (Mariano et al., 2024). Moreover, studies such as Hashempour-Baltork et al. (2023), Gamarra-Castillo et al. (2022), Choi et al. (2025), Li et al. (2025), and Shahbazzpour et al. (2021) only analyzed the physical properties of mycoprotein-based meat analogue using various hydrocolloids individually and compared with controls. Besides using underexplored *Aspergillus oryzae* strain, this presented a gap where the effect of combined hydrocolloids in mycoprotein matrix as well as optimization formulation study using RSM are not investigated yet. Therefore, this research aimed to fill that gap as the objective.

1.2 Objective

The objective of this research is to determine the influence of xanthan gum, sodium alginate, and kappa-carrageenan to the physical properties and interactions within mycoprotein meatball and find the optimized formulation in physical properties (texture profile, color, WHC, Cooking yield) for mycoprotein-based meatball using response surface methodology according to commercial benchmark

1.3 Hypothesis

The null hypothesis (H_0) is that there is not any significant difference between each hydrocolloid to the texture profile, color, water holding capacity, and cooking yield of mycoprotein meatball. The alternative hypothesis (H_1) is that there is at least one significant effect from each hydrocolloid to the texture profile, color, water holding capacity, and cooking yield of mycoprotein meatball.