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Assessing The Impact of Transglutaminase and Methylcellulose on Physical Properties of Seitan-Infused Plant-based Meat Analog Patties Compared to Beef Patties

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***Abstract*— The growing consumer shift toward sustainable and health-conscious eating has spurred innovation in plant-based meat alternatives. Among emerging strategies, transglutaminase (TG) is gaining attention as a promising, health-friendly bio-binder that can enhance the structural and functional properties of meat analogs. This study developed plant-based meat analog patties (PBMAP) using textured vegetable protein (TVP) and seitan in three ratios (30:70, 50:50, 70:30), alongside different binder formulations: 2% methylcellulose (MC), 2% TG, and a combination of 1% MC + 1% TG. These patties were benchmarked against a conventional beef patty (control, CON) and evaluated for key physicochemical attributes including visual appearance, cooking loss, color, texture, and surface area. PBMAPs closely resembled CON in appearance but showed significantly reduced shrinkage ($p < 0.05$). Higher seitan content correlated with firmer texture (0.31–3.18 N), although the CON remained substantially tougher (13.77 N, $p < 0.05$). TG-enriched formulations also demonstrated superior moisture retention and increased surface area, with reduced cooking loss ($p < 0.05$). While PBMAPs had lower lightness (L*), their redness (a*) and yellowness (b*) were not significantly affected by protein ratio or binder type. Overall, formulations containing 70% seitan with either 2% TG or a 1% MC + 1% TG blend achieved improved texture and functional performance, making them promising candidates for next-generation meat analogs. Nevertheless, color differences remain a challenge in replicating the full sensory profile of traditional beef. These findings offer valuable insights into the synergistic effects of TG and MC on the quality of plant-based meat products.**

***Keywords*— Meat analog; Patties; TVP; Seitan; Volscan**

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I. INTRODUCTION

Plant-based meat analogues (PBMAs) are designed to mimic the taste, texture, and nutritional value of conventional meat using plant-based proteins [1]. These products are increasingly in demand due to growing concerns over the health risks associated with red and processed meat consumption, such as cardiovascular disease and colorectal cancer, as well as the environmental impacts of livestock production, including greenhouse gas emissions, land use, and water consumption [2,3]. In contrast, plant-based proteins contribute to improved health outcomes by offering dietary fiber, polyunsaturated fatty acids, oligosaccharides and carbohydrates while reducing cardiovascular diseases and cholesterol intake and type II diabetes mellitus [4,5]. Additionally, PBMAs offer a more sustainable alternative with significantly lower greenhouse gas emissions, land use and water consumption compared to animal protein sources [6].

Among various protein sources used in PBMAs, soy-based textured vegetable protein (TVP) and wheat gluten (seitan) are among the most effective due to their high protein content, favourable and cost-effectiveness [7]. TVP, produced through extrusion of defatted soy flour, offers a fibrous texture, high water absorption capacity and a neutral flavour that facilitates diverse sensory profiles [8]. Seitan is composed of hydrated glutenin and gliadin, which form an elastic, meat-like structure upon hydration and heating, which contributes to chewiness and structural integrity [9]. While soy is widely used for its functional versatility, the incorporation of seitan is particularly suitable for achieving desirable meat-like textures in plant-based patties [10].

The choice of binding agents strongly influences the functional quality of PBMAs. Methylcellulose (MC) is a thermally reversible hydrocolloid that is commonly used in commercial formulations due to its ability to form gels upon heating, improving water retention, juiciness, and cohesiveness [3,11]. MC is considered safe for consumption, though it is not digested or absorbed in the human gastrointestinal tract [2]. Transglutaminase (TG), an enzyme employed across various food applications, including meat products, assumes a pivotal role in enhancing the characteristics of food items. It functions as a binding agent, facilitating the creation of isopeptide bonds among proteins. This enzymatic process heightens the texture, firmness, elasticity, and water-binding capacity of food products [12]. TG is particularly useful for overcoming challenges in processing less versatile raw materials like mechanically deboned meat, collagen, and blood proteins. While phosphates in meat processing have raised health concerns [13], the use of transglutaminase in plant-based products is relatively new.

Although both MC and TG have been individually employed in plan-based systems, limited studies have investigated their combined effects in a dual-protein base, such as seitan and TVP. The present study addresses this gap by examining the physicochemical and structural properties of plant-based patties formulated with different TVP to seitan ratios and binder combinations. The aim are to evaluate and compare the visual appearance and physicochemical properties, including cooking loss, color, texture, shear force, and surface area, with conventional beef patties to determine the effectiveness of binder interactions and protein synergies.

II. MATERIAL AND METHODS

A. Raw Materials

The plant-based meat analog patties (PBMAPs) were developed using wheat flour and high-quality textured vegetable protein (TVP) as the primary protein sources. Methylcellulose (high viscosity, Modernist Pantry, Eliot, ME, USA), transglutaminase (Meat Glue, Germany), and isolated soy protein 90% (ISP) (Green Spring, China) were used as binders. Other ingredients, including red yeast rice, garlic powder, mushroom powder, cajun spices, smoked liquid, cocoa butter, palm oil, white pepper, and black pepper, were sourced locally, as described in **Table 1**. Fresh beef loins, aged less than 10 hours postmortem, were obtained from a regional market in Kampung Raja, Terengganu, Malaysia, for control patties (CON) preparation.

B. Sample Preparation and Processing

The PBMAPs were prepared following a previously established method with slight modifications [14]. The formulations included three different ratios of TVP-to-seitan (30:70, 50:50, and 70:30). Seitan was made by kneading wheat flour and water until smooth, then soaking it in cold water for 2 hours and washing it multiple times until clear. The resulting gluten was filtered and sieved to remove excess starch and water. TVP was rehydrated in 150 ml warm water at 60°C for 2 h. The prepared seitan and TVP were mixed with the remaining ingredients using a food processor (MK-5087M, Osaka, Japan). CON were made using minced beef and isolated soy protein (ISP) as a binder, using the same procedure and equipment. Both patties were shaped using a mold and weighed to 60 g each. However, only PBMAPs were steamed for 40 minutes before being chilled at 4°C for subsequent cooking and analysis.

C. Visible Appearance and Warner-Batzler Shear Force Analysis

The visual aspect of the patties was documented through photography, employing a digital camera (EOS 700D, Canon, Tokyo, Japan). This process facilitated the observation and comparison of the external appearance before and after the cooking phase [15]. Patty's firmness was measured using a stable microsystem texture analyzer (Instron 4443, CA, USA) and the Warner-Bratzler shear force method. A 1 mm thick Warner-Bratzler shear blade was used to cut a 2 × 2 × 2 cm sample at a speed of 1.5 mm/s, ensuring complete separation (100% cutting percentage) [16]. The sample was then sheared with a triangular slotted blade perpendicular to the muscle fiber orientation. The force needed to shear each sample was plotted against time in a force-deformation graph, with the highest point indicating the maximum shear force. The resulting force was recorded in Newtons (N).

D. Textural Profile and Volscan Profile Analysis

A double arm (Stable Micro Systems Ltd, England) was employed to evaluate the texture profile of the cooked patty samples. This texture analysis encompassed attributes such as hardness, chewiness, cohesiveness, springiness, and gumminess. The patty samples were sectioned into cubes measuring 1 × 1 × 1 cm and subjected to examination using a cylindrical probe (P/75, 7 mm Compression Platen). The probe operation adhered to a meat-specific default configuration, maintaining constant speeds of 1.00 mm, 5.00 mm, and 5.00 mm for the pre-test, test, and post-test stages, respectively. Employing a two-cycle sequence along with a 30 kg load cell, the samples were compressed at a crosshead speed of 5.0 mm, ultimately reaching a final strain of 50 [17]. The measurement of the patties' surface area was conducted using the Volscan Profiler (VSP300, Stable Micro Systems, Godalming, UK). The system calibration followed the manual procedure, using tare weight and manual weight settings. A vertical step of 10.0 mm was specified, alongside a standard rotation speed, while the shape parameter was configured as round. To commence the measurement, the sample was positioned on the profiler's base and centred accordingly.

TABLE 1
FORMULATION OF CONVENTIONAL AND PLANT-BASED MEAT ANALOG PATTIES.

Ingredient %	**Formulation %**									
	CON	**MC3070**	**MC5050**	**MC7030**	**TG3070**	**TG5050**	**TG7030**	**MCTG3070**	**MCTG5050**	**MCTG7030**
Transglutaminase (TG)		2	2	2				1	1	1
Methylcellulose (MC)					2	2	2	1	1	1
Textured soy protein (TVP)		19	32	45	19	32	45	19	32	45
Seitan		45	32	19	45	32	19	45	32	19
Isolated soy protein	2	0	0	0	0	0	0	0	0	0
Minced meat	68	0	0	0	0	0	0	0	0	0
Garlic powder	3	3	3	3	3	3	3	3	3	3
Mushroom powder	3	3	3	3	3	3	3	3	3	3
Cajun spices	3	3	3	3	3	3	3	3	3	3
Smoke seasoning powder	3	3	3	3	3	3	3	3	3	3
Black pepper	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
White pepper	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
Red yeast rice	0	3	3	3	3	3	3	3	3	3
Salt	1	1	1	1	1	1	1	1	1	1
Sugar	3	3	3	3	3	3	3	3	3	3
Cocoa butter	5	5	5	5	5	5	5	5	5	5
Palm oil	5	5	5	5	5	5	5	5	5	5

Note: Control (CON), Methylcellulose with 30:70 seitan and TVP (MC3070), Methylcellulose with 50:50 seitan and TVP (MC5050), Methylcellulose with 70:30 seitan and TVP (MC7030), Transglutaminase with 30:70 seitan and TVP (TG3070), Transglutaminase with 50:50 seitan and TVP (TG5050), Transglutaminase with 70:30 seitan and TVP (TG7030), Methylcellulose and Transglutaminase with 30:70 seitan and TVP (MCTG3070), Methylcellulose and Transglutaminase with 50:50 seitan and TVP (MCTG5050), Methylcellulose and Transglutaminase with 70:30 seitan and TVP (MCTG7030).

E. Cooking Loss

The cooking loss was taken to be the percentage of fluids lost (which may contain water, proteins, fats, and minerals) after cooking a sample for a specific time, which was calculated using the following formula [18]. The patties were cooked on a hot pan with a low flame for 5 min on each side. The calculation of the cooking loss was based on this formula:

Cooking loss % = [Initial Weight (g) – Weight after cooking (g)/ initial weight x 100%] eq (1)

F. Color Properties Analysis

The color of cooked patties was assessed using a tristimulus colorimeter, the Konica–Minolta Chromameter (CR-400 with 8-mm aperture and 0° viewing angle, Konica-Minolta, Inc., Tokyo, Japan). This evaluation was conducted following the CIE 1976 L*a*b* and CIEL*C*h* color spaces. To ensure accuracy, the instrument was calibrated against a standard white reference tile provided by the instrument manufacturers. The CIE 1976 L*a*b* color space comprises three dimensions: L* (lightness), which pertains to the white-to-black axis, *a (redness) signifying the red-to-green axis, and b* (yellowness) representing the yellow-to-blue axis.

G. Statistical Analysis

Each experimental measurement was conducted in triplicate. The data were analyzed using a one-way analysis of variance, followed by the Duncan test for multiple mean comparisons ($p < 0.05$). Statistical analyses performed on measurement means and standard deviations, conducted using SPSS version 23 (IBM Corp., SPSS, Statistic, Armonk, NY, USA).

III. RESULT AND DISCUSSION

A. Visual Appearance

Digital images of both beef (CON) and plant-based meat analog patties (PBMAP), featuring varying percentages of seitan, TVP, and binders, were captured under uniform lighting conditions (**Figure 1**). Employing image processing techniques, the background of the samples was removed, and subsequent categorization of the images was based on their respective formulations. Before cooking, the color of the PBMAP differed between treatments, displaying a red hue that transformed into a brown color post-cooking. Notably, the raw PBMAP with a higher seitan ratio (70%) exhibited a lighter coloration. Following the cooking process, no evident disparities were observed in terms of formulation, whether in terms of base ingredients or binders added. Cooking caused both CON and PBMAP to undergo shrinkage. The CON demonstrated a visible reduction in size, while the PBMAP counterparts maintained their original diameter. This phenomenon can be attributed to the development of gluten aggregates during the cooking of PBMAP, leading to enhanced network integrity and reduced protein solubility [19]. The shrinkage observed in the CON during cooking is likely due to the natural denaturation of meat proteins such as actin and myosin. This process causes the proteins to aggregate and release moisture [18]. This process helps develop the expected texture and flavour in beef patties [20]. However, if denaturation happens before cooking, it usually indicates a decline in quality. The structural difference between meat proteins and gluten also affected the texture and appearance of beef and plant-based patties [21]. **Figure 1** presents volscan profiler images, offering a 2D sectional view of the patties' internal structure. In the case of PBMAP, the interval line displayed a uniform and consistent network pattern. In contrast, the interval matrix of the CON appeared heterogeneous, indicative of intermuscular connective tissue and myofibrillar protein fibrous structure. This interval line serves as a visual representation of protein networks and muscle tissue arrangement [22]. Consequently, these differences contribute to the distinct appearances between beef and PBMAP.

B. Warner-Batzler Shear Force and Texture profile Analaysis

The texture properties of patties are outlined in **Table 2**. The CON exhibited the highest shear force value at 13.77 N, contrasting with the range of 0.31-3.18 N observed in the PBMAP samples, with the differences being statistically significant ($p<0.05$). Shear force serves as an effective indicator of initial bite tenderness/toughness in meat, a trait influenced by cooking methods that cause modifications in myofibrillar protein and connective tissues due to heat. These alterations can significantly affect the overall texture and tenderness of the meat. The denaturation of myofibrillar proteins and the subsequent reduction in connective tissue size can lead to beef firmness, thereby resulting in heightened toughness [15]. Within the PBMAP group, TG5050 registered the highest shear force value (3.18 N), trailed by MCTG5050 (2.64 N), and MC3070 (2.22 N). The amalgamation of well-balanced base ingredients (TVP and seitan) along with a 2% TG infusion has contributed to the denser texture of the TG5050. This density imparts increased toughness to this plant-based patty in comparison to the other treatments. The incorporation of transglutaminase as a binding agent also contributes to the patty's hardness, due to the formation of a more robust and compact protein network [23]. Despite these variations, the shear force values for the PBMAP samples remained lower than CON ($p<0.05$), likely due to the differences in protein types and forms used. This outcome aligns with the findings of Vu, Zhou [18], who observed lower shear force values in plant-based burgers when contrasted with their beef patty samples.

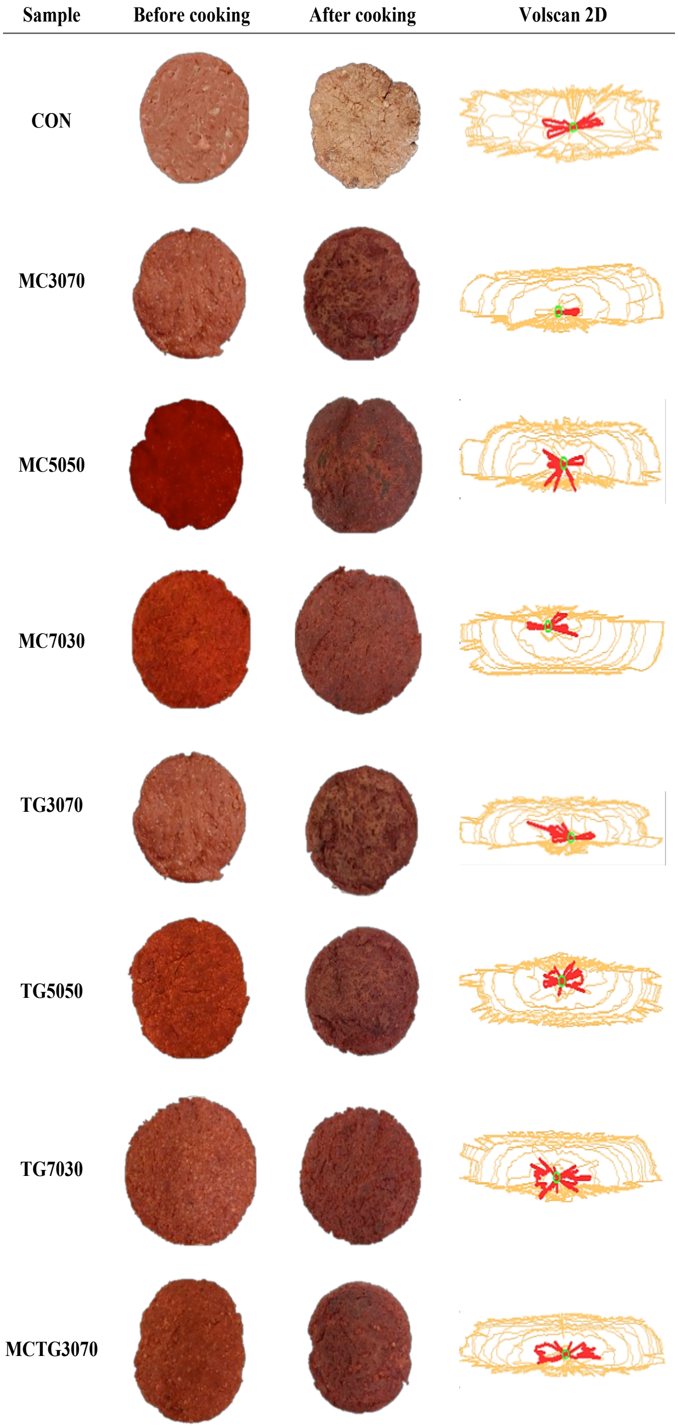

Fig.1 Visible appearance of beef and plant-based meat analog patties before and after cooking and 2D image from volscan profile.

TABLE 2
THE WARNER-BRATZLER SHEAR FORCE AND TEXTURE PROFILE ANALYSIS

Sample	Shear force (N)	Hardness (kg)	Springiness (mm)	Cohesiveness (%)	Gumminess (kg)	Chewiness (kg)
CON	13.77±1.87^{a}	9.21±2.62^{a}	0.86±0.02^{a}	0.72±0.02^{a}	6.60± 0.01^{a}	5.69±1.48^{a}
MC3070	2.22±1.22^{bc}	4.80±0.84^{b}	0.64±0.04^{b}	0.58±0.02^{bc}	2.77±0.05^{b}	1.76±0.23^{b}
MC5050	1.74±0.26^{bcd}	3.60±1.18^{bc}	0.65±0.12^{b}	0.60±0.08^{b}	2.23±1.04^{bc}	1.53±0.97^{b}
MC7030	1.84±0.05^{bc}	2.34±0.19^{c}	0.39±0.01^{d}	0.40±0.01^{d}	0.93±0.07^{c}	0.36±0.04^{c}
TG3070	1.64±0.32^{cd}	4.02±0.82^{bc}	0.63±0.02^{b}	0.53±0.03^{c}	2.13±0.05^{bc}	1.34±0.35^{bc}
TG5050	3.18±0.41^{b}	2.15±0.41^{c}	0.65±0.03^{b}	0.54±0.02^{c}	1.16±0.20^{c}	0.76±0.15^{bc}
TG7030	1.92±0.64^{bc}	3.34±0.24^{bc}	0.50±0.05^{c}	0.41±0.02^{d}	1.36±0.15^{c}	0.67±0.08^{bc}
MCTG3070	0.31±0.49^{d}	4.02±0.98^{bc}	0.64±0.02^{b}	0.54±0.01^{bc}	2.19±0.57^{bc}	1.40±0.39^{bc}
MCTG5050	2.64±0.35^{bc}	2.46±0.17^{c}	0.59±0.02^{b}	0.55±0.01^{bc}	1.35±0.07^{c}	0.79±0.069^{bc}
MCTG7030	1.39±0.81^{cd}	2.78±0.23^{c}	0.38±0.04^{d}	0.34±0.03^{e}	0.94±0.15^{c}	0.36±0.09^{c}

The results comprise mean ± standard deviation. $^{a\text{-}e}$ Means with different letters within a column are statistically significant ($p < 0.05$).

Texture profile analysis encompasses several sensory attributes, including hardness, springiness, gumminess, cohesiveness, and chewiness. As outlined in **Table 2**, the CON exhibited the highest values across all these texture profile attributes. Specifically, hardness, springiness, cohesiveness, gumminess, and chewiness were quantified at 9.21 kg, 0.86 mm, 0.72%, 6.60 kg, and 5.69 kg, respectively. In contrast, the PBMAP samples showed significantly lower values ($p<0.05$), with hardness ranging from 2.15-4.80 kg, springiness from 0.38-0.65, cohesiveness from 0.34-0.60%, gumminess from 0.93-2.77 kg, and chewiness from 13.43-16.43 kg. these differences can be attributed to variations in protein quality and contrast between the animal-based and plant-based matrices. Recent studies have demonstrated that PBMAs often exhibit lower hardness and cohesiveness due to the absence of myofibrillar protein structures and reduced protein-protein interactions during thermal processing [24,25]. Furthermore, the process of protein denaturation during cooking predominantly contributes to the enhanced hardness observed in the CON sample, where heat-induced triggers changes in muscle proteins, leading to cross-linking and the formation of a rigid protein network. This transformation significantly impacts the meat's texture, rendering it firmer and tougher [26,27]. Among PBMAP samples, MC3070 exhibited higher values for hardness, gumminess, and chewiness, whereas MC5050 showcased heightened cohesiveness and springiness values. Formulations with higher seitan content, particularly those containing 70%, displayed significantly greater values for these texture attributes ($p <0.05$). Seitan, a hydrated and structured product derived from wheat gluten, is known for its fibrous and meat-like texture due to the formation of an elastic protein network. This network arises from glutenin and gliadin interactions that facilitate covalent (disulfide) and non-covalent bonding. Contributing to structural strength and elasticity [28,29]. Higher gluten content has been shown to enhance the fibrous structure and mechanical resiliency of PBMAs, which likely explains the denser, chewier texture in high-seitan formulation [28]. Moreover, Somayeh, Yunyu [9] emphasized that wheat gluten reinforces texture, binding capacity and moisture retention through disulfide crosslinking and protein aggration. Conversely, the impact of 2% binders on cohesiveness did not exhibit substantial variation across formulations, indicating that the influence of base ingredients is more pronounced in shaping the textural attributes of PBMAP samples.

C. Surface Area and Cooking Loss

As discussed above, the alteration in the shape observed in the CON sample after cooking is reflective of changes in surface area, a finding corroborated by the data in **Table 3**. Specifically, the CON sample exhibited a smaller surface area than the PBMAP samples ($p <005$). This reduction in surface area can be attributed to the shrinkage of myosin within the temperature range of 50 to 65°C and actin within 70 to 75°C. Such shrinkage leads to fluid loss within the protein matrix due to decreased surface area and a reduction in pore number [30]. Our findings align with those of Vu, Zhou [30], who similarly observed a reduction in surface area during the cooking process. Interestingly, the inclusion of MC alone in the formulation decreased surface area (measuring 13.43-14.91 m^2), albeit without a significant difference compared to the CON sample. Conversely, formulations involving TG and MCTG displayed surface area values comparable to each other and exceeding those of the MC binder. We posit that the creation of a protein-protein network by transglutaminase at lower temperatures (pretreatment) holds greater significance than MC, as MC forms a gel at higher temperatures. Transglutaminase facilitates protein chain extension and enhances the gel network, thereby aiding in maintaining the structural integrity and surface area during cooking. A study by Li, Fu [31], supports the fact that low-temperature pretreatment notably promotes transglutaminase cross-linking, amplifying emulsifying properties and water-holding capacity. In contrast, higher cooking temperatures cause protein denaturation, where MC primarily mitigates losses by forming a thermoreversible gel upon heating [32]. Remarkably, the combination of MC (1%) and TG (1%) yields a pronounced impact on surface area even at lower concentrations, indicative of a synergistic effect.

The cooking loss of PBMAP and CON is shown in **Table 3**. Notably, the cooking loss of CON exhibited a significantly higher value (36%) compared to PBMAP (12-19%). As discussed above, the higher losses in meat protein are largely attributed to protein denaturation. The application of binding agents and base ingredients played a pivotal role in influencing the cooking loss results in PBMAP samples. The impact was particularly pronounced when incorporating higher amounts of TVP at 70% and utilizing MC as a binder. Upon examining the outcomes in Table 3, it becomes evident that substitutions involving a high TVP content at 70% (MC7030, TG7030, and MCTG7030) consistently exhibited higher cooking losses. Similarly, samples exclusively containing MC (MC3070, MC5050, and MC7030) also demonstrated elevated cooking loss percentages. In contrast, the inclusion of seitan at a higher proportion, specifically at 70% (TG3070 and MCTG3070), and the inclusion of TG and a combination of MCTG appeared to mitigate these losses. The incorporation of seitan prevents excessive cooking loss due to the formation of a three-dimensional network of disulfide protein links. This network creates a fibrous structure during high-moisture extrusion, functioning as a binder and structuring agent within gluten [33]. The reduction in cooking loss observed in samples treated with TG can be attributed to its catalytic activity, which facilitates the creation of covalent cross-links involving ε-(γ-glutamyl) lysine interaction between γ-glutamyl residues and ε-amino groups of lysine residues. This process extends protein chains and forms high molecular-weight polymers [34], inadvertently enhancing the stability (gelation, elasticity, water-holding capacity, solubility, and functional properties) of plant-based protein products [35]. According to Forghani, Eskandari [23] The ε-(γ-glutamyl) lysine bond is approximately 20 times stronger than hydrogen bonds and hydrophobic forces, thus contributing to improved mechanical and rheological properties of proteins. Comparatively, the cooking loss in PBMAP containing MC is slightly higher than in patties containing TG or a combination of MCTG as binders. This distinction arises from the differing polymer chains of these substances. MC polymers are surrounded by water molecules in a cage-like structure, accompanied by hydrophobic methyl groups that gradually lose their effectiveness during cooking. In contrast, TG catalyzes protein molecules to form extensive polymeric structures through a cross-linking process. This aggregation enhances the patty's water retention capacity, preventing water shrinkage during processing and ultimately improving the patty's cooking yield [23, 30].

TABLE 3
THE SURFACE AREA AND COOKING LOSS ANALYSIS

Sample	Surface area (m^2)	Cooking loss (%)
CON	13.70 ± 1.18^{bc}	36.13 ± 0.57^{a}
MC3070	14.91 ± 0.84^{abc}	18.91 ± 1.47^{b}
MC5050	13.43 ± 1.71^{c}	17.21 ± 0.76^{c}
MC7030	14.87 ± 0.86^{abc}	19.09 ± 0.14^{b}
TG3070	16.06 ± 0.75^{a}	12.38 ± 0.87^{d}
TG5050	15.93 ± 0.92^{a}	16.98 ± 0.36^{c}
TG7030	16.43 ± 0.42^{a}	18.93 ± 1.09^{b}
MCTG3070	16.10 ± 0.96^{a}	12.40 ± 0.57^{d}
MCTG5050	15.73 ± 0.96^{a}	18.36 ± 0.14^{bc}
MCTG7030	15.40 ± 1.32^{ab}	17.94 ± 0.51^{bc}

The results comprise mean ± standard deviation. $^{a\text{-}d}$ Means with different letters within a column are statistically significant ($p < 0.05$).

D. Color Properties

The color properties of the patties are outlined in **Table 4**. The changes in color properties of the cooked patties can be attributed to alterations in microstructures and compositions. Notably, the CON sample exhibited a higher lightness (L*) value of 35.27, distinct from the range of 23.65–27.67 observed in the PBMAP samples. The L* values are generally influenced by the scattering and absorption effects of light waves [18]. The heterogeneous structure of meat leads to lighter color attributes due to the dispersion of light waves [18, 36, 37]. Conversely, the homogeneous structure of PBMAP might be responsible for the lower L* values due to their propensity for light wave absorption. Among the PBMAP samples, the lowest L* value was recorded in the samples with higher seitan incorporation (MC3070, TG3070, and MCTG3070 with 23.65, 24.52, and 24.02, respectively). Despite seitan being produced from wheat flour and initially having a whiter color, the process of forming gluten dough and undergoing multiple washing cycles to remove soluble and dispersible components contributes to its opacity. Consequently, its color deepens upon cooking [38]. Similar observations were found by Zhang, Wang [39], who noted decreased L* values in crust batter with the addition of wheat gluten. However, the lower lightness values in the PBMAP were offset by a well-balanced integration of TVP and seitan. This balance led to an increase in L* values, evident in samples like MC5050, TG5050, and MCTG5050, which displayed values of 27.67, 27.51, and 27.15, respectively.

The redness (a*) values of the CON, measuring lower at 8.45, stand in contrast to the range of PBMAP, which exhibits values from 12.61 to 17.96. This discrepancy is anticipated, as beef patties inherently contain myoglobin, contributing to their natural red color. The diminished a* value in CON can be elucidated by the denaturation of myoglobin and the alteration of the redox state [18]. Meanwhile, the augmented a* values in PBMAP can be attributed to the inclusion of red yeast rice within the formulation. In terms of yellowness (b*) values, CON showcases a value of 13.77, aligning with the spectrum of values found in PBMAP samples ranging from 10.38 to 14.06. The denaturation of myoglobin leads to a shift in the redox state within beef, and the formation of new pigments in cooked patties contributes to a brownish hue, stemming from the Maillard reaction . However, the brown coloration observed in PBMAP is primarily ascribed to the foundational ingredients employed. Our findings are parallel to the conclusions of Vu, Zhou [18], who found that plant-based meat analog patties displayed lower L* values and higher a* values, connecting these color variations to the divergent composition and structure between beef and plant-based meat analog patties. A similar pattern was also documented by Forghani, Eskandari [23], who attributed the color divergence in plant-based meat analog patties to disparities in formulations and cooking processes.

TABLE 4
THE COLOR PROPERTIES

Sample	**L***	**a***	**b***
CON	35.27±0.24^{a}	8.45±0.26^{f}	13.77±0.99^{ab}
MC3070	23.65±0.37^{e}	12.61±0.30^{e}	6.38±3.40^{d}
MC5050	27.67±0.19^{b}	16.99±0.39^{abc}	12.27±0.47^{abc}
MC7030	26.15±0.22^{c}	16.40±0.13^{c}	11.38±0.42^{bc}
TG3070	24.52±0.03^{d}	15.37±0.04^{d}	11.29±0.07^{c}
TG5050	27.51±0.69^{b}	17.96±1.39^{a}	14.06±1.06^{a}
TG7030	25.74±0.40^{c}	16.91±0.42^{bc}	12.41±0.68^{abc}
MCTG3070	24.02±0.72^{de}	16.06±0.60^{cd}	12.41±0.86^{abc}
MCTG5050	27.15±0.29^{b}	17.55±0.17^{ab}	13.62±0.53^{abc}
MCTG7030	25.79±0.11^{c}	16.80±0.41^{bc}	11.95±0.67^{abc}

The results comprise mean ± standard deviation. $^{a-f}$ Means with different letters within a column are statistically significant ($p < 0.05$).

CONCLUSION

This study shows that plant-based meat analog patties (PBMAP), developed using textured vegetable protein (TVP), seitan, and functional binders, showed noticeable cooking behavior and textural properties compared to conventional beef patties (CON). PBMAP exhibited significantly less cooking shrinkage and a more uniform internal structure. While softer in overall texture, higher seitan content increased hardness and chewiness, aligning with seitan's role in contributing to fibrous structure and mechanical strength. Transglutaminase enhanced structural integrity and water retention, while methylcellulose facilitated gel formation during the cooking process. Although protein denaturation likely contributes to cooking loss in CON, this was inferred from texture and moisture loss and further molecular validation is recommended. In contrast, TG-treated, seitan-rich PBMAP showed reduced cooking loss and improved structural stability. In terms of appearance, PBMAP samples are darker with redness and yellowness varying depending on formulation. These findings demonstrate how targeted protein and binder combinations can be used to optimize the texture, functionality, and cooking performance of plant-based meat analogs.

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CONFLICT OF INTEREST

No potential conflicts of interest are disclosed by the authors. The authors declare that no generative AI or AI-assisted technologies were used in the writing or data processing of this manuscript.

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