Research Paper

Future of Food: Journal on Food, Agriculture and Society
10 (2) April 2022

# The Effect of Transglutaminase on Physicochemical, Microstructural, and Organoleptic Properties of Low-Fat Karish Cheese

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**Data of the article**

First received : 08 February 2022 | Last revision received : 20 March 2022
Accepted : 07 April 2022| Published online :15 April 2022
DOI : 10.17170/kobra-202110144906

**Keywords**

karish cheese;
transglutaminase;
microstructural;
organoleptic properties;
milk proteins

Karish cheese is considered to be one of the most traditional Egyptian cheeses. It is manufactured from low-fat milk. It can be a key part of a healthy diet, especially for the elderly and those suffering from obesity-related illnesses. Low-fat cheeses, such as karish, have a crumbly texture, which is a major issue for cheesemakers. This research aimed to improve the texture of karish cheese using transglutaminase (TGase), and to study the effects on cheese's physical, microstructural, and organoleptic properties at different TGase levels of 1.0, 1.5, and 2.0 U $g^{-1}$ protein. TGase increased the structure of karish cheese at a rate of 1.5–2.0 U $g^{-1}$ protein, compared to the control sample. Scanning electron microscopy images showed that the protein matrices in karish cheese samples that had been treated with TGase were relatively more compact than those in the untreated sample. Proteins in karish cheese samples treated with TGase showed crosslinking in SDS-PAGE scans. Casein fractions corresponding to bands became less intense as the concentration of TGase increased. To summarise, TGase improved the characteristics of karish cheese at the rate of 1.5–2.0 U $g^{-1}$ protein TGase, and this structural modification is recommended for such traditional cheese in order to fulfill customer demand.

## 1. Introduction

Karish cheese is the most popular low-fat soft cheese consumed in Egypt. The coagulating agent of the cheese is a mixed culture of lactic acid bacteria (Delacroix-Buchet et al., 2005). Karish, also known as kariesh or kareish cheese, is a common lactic cheese produced from fresh skimmed milk. It includes the bulk of the components present in skim milk. Defatted milk such as Laban Rayeb, Laban Khad, Laban Zeer, or mechanically skimmed milk are also used in the production of karish cheese (Laban Farz) (S.A., 2008). As a highly demanded low-fat cheese and a healthier dietary option for many in Egypt and other Arab countries (Allam et al., 2017; Topcu et al., 2020), low-fat cheese is still the best choice for obese and diabetic patients' diets (Sayadi et al., 2013). However, low-fat content in cheese results in some structural problems, such as a hard and rubbery texture, and low meltability and stretchability. Furthermore, cheese flavour, colour, and mouthfeel are affected (Pereira et al., 2009).

The competitive dairy industry is trying to improve the quality and usefulness of low-fat dairy products for improved customer acceptance (Lauber et al., 2000; Şanlı et al., 2011). The modification of milk proteins by enzymes to create a cheese with improved output and quality features has been used in the dairy

industry for a long time (Danesh et al., 2018; Soltani et al., 2022; Topcu et al., 2020).

Transglutaminase (TGase) is currently widely used in the food industry because it enhances the functionality, solubility, and emulsification of products by forming links within the protein molecules (Martins et al., 2014). TGase amplifies the textural properties of protein-gel-based dairy products (Monsalve-Atencio et al., 2022; Zhang et al., 2012). TGase is a member of the transferase family, protein-glutamine: amine-glutamyl transferase (TGase, EC 2.3.2.13) (Kieliszek & Misiewicz, 2014; Wang et al., 2018). The crosslinking of milk proteins with TGase is one of the most recent methods for enhancing the biofunctional properties of low-fat cheese products.

TGase has been shown to primarily catalyze protein crosslinking on α-lactalbumin and β-lactoglobulin in whey protein studies. This is primarily because the TGase enzyme can enhance the rheological and physical characteristics of milk-based acid gels by producing intra- and intermolecular ionic crosslinks between two amino acid residues in the milk proteins αs1, αs2, β and κ-caseins, and whey protein (El-Kiyat et al., 2021; Gharibzahedi et al., 2018; Rodriguez-Nogales, 2006). Additionally, TGase is a crosslinking enzyme that can catalyze α -lactoglobulin, β-lactoalbumin, and even caseinomacropeptide (Tolkach & Kulozik, 2005) crosslinking in cheese whey by producing intramolecular and intermolecular ε-(γ-glutamyl) lysine crosslinks, resulting in a high-molecular-weight polymer. However, without the presence of a mediator to enhance enzyme catalysis, TGase may catalyse protein crosslinking.

The protein chain of α-lactalbumin consists of 8 glutamine residues and 12 lysine residues, whereas β-lactoglobulin contains 16 glutamine residues and 15 lysine residues (Gauche et al., 2008). The reactions of catalyzed TGase are as follows:

R-Glu-CO-NH2 + H2N- R′ → R-Glu-CO-NH R′ + NH3

R-Glu-CO-NH2 + H2N-Lys- R′ →R-Glu-CO-NH-Lys-R′ + NH3

R-Glu-CO-NH2 + H2O → R-Glu-CO-OH + NH3 (Rachel & Pelletier, 2013).

TGase is utilised to improvise a range of activities, including coagulation and antibacterial immune responses (Kashiwagi et al., 2002). The present goals are to augment the functional qualities of low-fat cheese to meet the demands of cheesemakers and customers, to optimize structure characteristics by developing manufacturing strategies that used TGase, and to identify the optimal concentration of TGase to achieve the goal.

## 2. Materials and Methods

### 2.1. Materials

Low fat pasteurized cow and buffalo milk was used in the production of karish cheese samples. The fat and total solid contents of the pasteurized milk were 0.1% and 12.8%, respectively. Ajinomoto Co. provided the TGase, while Ajinomoto Co., Inc., France provided the food enzyme preparation. The TGase activity was reported to be 120 U g1protein (data supplied by the industry from Merck Chemicals Ltd.). All provided chemicals were of analytical grade (Darmstadt, Germany).

### 2.2. Karish Cheese Manufacturing

Karish cheese was manufactured according to the method of Ezzel-Din (1978). Fresh skimmed cow and buffalo milk (1:1) (1 g fat/100 mL milk) was heat-treated at 85 °C/15 s and left to cool to 40 °C in ice water for 10 minutes. The milk used for karish cheese production was divided into four portions. The first portion was the control karish cheese (C) without TGase addition. The three other portions were treated with 1.0, 1.5, and 2 U $g^{-}1$ protein TGase. The active yoghurt starter culture of Streptococcus thermophilus and Lactobacillus delbrueckii subsp. Bulgaricus (1:1) was obtained from CHR Hansen Lab (Denmark) and was added at the rate of 1.5% of the weight of skimmed milk to complete coagulation. The samples were then treated with 0.02 percent calcium chloride and incubated at 40 °C. After coagulation, 2.5% salt was added to the curd, which was ladled onto mats; the cheese curd was then moved to the refrigerator overnight at 5 ± 2 °C. Cheese samples were taken for chemical and organoleptic measurements after 24 h.

### 2.3. Proximate Analysis


ISSN-Internet 2197-411x OLCL 862804632

The fat, protein, and titratable acidity of the samples were determined using AOAC (2000) methods. The nitrogen content (N) of the samples was estimated using the method described by AOAC (1990), and crude protein was calculated as N × 6.25 (Imran et al., 2008). The ash content was evaluated by dry ashing the samples in a muffle furnace at 550 °C for 6 hours. Before being placed inside the muffle furnace, samples were dried in a 105 °C oven.

### 2.4. Scanning Electron Microscopy (SEM)

Two or three 0.5 to 1 cm samples were collected from the cheese mats and fixed in 5 °C cold buffered gluteraldehyde for two days. Samples were then washed with cacodylate buffer three times for 15 minutes each and postfixed in 1% osmium tetroxide for 2 hours. Next, the samples were again washed in cacodylate buffer three times for 15 minutes each and then dehydrated using an ascending series of ethanol of 30%, 50%, 70%, and 90% for 2 hours each, 100% for two days, and then amyl acetate for two days. Critical-point drying was applied to the samples using liquid carbon dioxide. Each sample was fixed onto metallic blocks using silver paint. By using a golden sputter-coating apparatus, samples were evenly gold-coated to a thickness of 15 nm. Samples were photographed and examined using a JEOL JSM 5400 LV scanning electron microscope 15–25.kv (Bozzola & Russell, 1999).

### 2.5. Sodium Dodecyl Sulfate–Polyacrylamide Gel Electrophoresis (SDS-PAGE)

SDS-PAGE of karish cheese samples was performed as described by Fling and Gregerson (1986) using a stacking gel of 4% (wt/vol) and a resolving gel of 12.5% (wt/vol). Electrophoresis was carried out on 9.4 × 8 × 1 mm slabs utilizing a vertical slab unit (Gel electrophoresis equipment, biometra Eco_Mini) and an electrophoresis power supply. Proteins from karish cheese were isolated using SDS electrophoresis, as described by Kalit et al. (2005) according to Laemmli, 1970. A total of 1.2 g of previously ground cheese was extracted for 30 minutes at 40 °C with steady shaking in 15 mL of buffer (0.5 M Tris-HCl, pH 6.8, 2% (wt/vol) SDS, 7% (vol/vol) glycerol, 5% (vol/vol) 2-mercaptoethanol). The obtained suspension was centrifuged at 2.600 g for 15 minutes. The clear solution under the top layer was then carefully removed and diluted with a sample buffer (0.5 M Tris-HCl, pH 6.8, 2% (wt/vol) SDS, 7% (vol/vol) glycerol, 4.3 vol/vol) 2-mercaptoethanol, 0.0025% (wt/vol) bromophenol blue). The ratio of extract to sample buffer was 1:3. Samples that had been diluted and frozen were used. Samples were heated for 5 minutes at 95 °C and then cooled to room temperature before electrophoresis. Each well was filled with 25 µL of the SDS-PAGE samples. To complete the gels, they were operated at 120 volts per gel for 2 hours. Gels were fixed and stained for 3 hours with 0.1% (wt/vol) Coomassie Blue R-250 (dissolved in 10% (vol/vol) glacial acetic acid) and 50% (vol/vol) methanol, and then unstained with 10% glacial acetic acid and 50% (vol/vol) methanol. Data from Pesic et al. (2012) were utilized for casein fraction measurements as well as molecular weight. SDS-PAGE was performed twice.

### 2.6. Organoleptic Properties

Twenty-one panellists (11 male and 10 female panellists, aged between 30 to 52 years) having expertise with white cheese and regular use of its descriptive terminology took part. Cheese samples were rated on colour and appearance (15 points), flavour (50 points), body and texture (35 points), and overall acceptability (100 points) according to Clark et al. (2009). Members of the panel were also instructed to report any flaws or disagreeable flavours.

### 2.7. Statistical Analysis

To assess the statistically significant differences in the experimental data, analysis of variance (ANOVA) was done using IBM SPSS statistics version 21 software. Results were deemed statistically significant when $p \leq 0.05$. Values for the mean ± standard deviation are also presented.

## 3. Results and Discussion

### 3.1. Proximate Analysis

Table. 1 shows the chemical composition of the karish cheese samples. Protein content increased significantly ($p \leq 0.05$) in karish cheese samples at TGase levels of 1.5 and 2.0 U $g^{-1}$ protein. These results agree with those of García-Gómez et al. (2019), who hypothesised that protein growth might be due to covalent bonding created between milk protein molecules (glutamine and lysine). TGase may also bind casein micelles to-

gether by forming crosslinks between different types of casein. β-lactoglobulin and α-lactalbumin are considered to be excellent TGase substrates (Razeghi & Yazdanpanah, 2020). There were no significant variations in ash, acidity, and fat values ($p \geq 0.05$) among the control and TGase-treated karish cheese samples. These findings corresponds with those of Aloğlu and Öner (2013), and Farnsworth et al. (2006).

### 3.2. Microstructural Analysis

TGase-treated and control karish cheese samples were examined with an SEM microscope, and micrographs (1000×) of these samples are shown in Figure 1. The control had an unstructured matrix, with certain areas being well-structured due to the development of thin strands arranged in a way that favoured the production of relatively small pores, while other areas in the control karish cheese had a high degree of compaction and large pores. In contrast, after treating milk with 1, 1.5, and 2 U $g^{-1}$ protein TGase, the protein network significantly changed, displaying a homogeneous and, at a glance, fluffy network with abundant thick strands of protein aggregates that interacted with one another, as well as visible changes in the porosity pattern. The SEM microstructures are shown in Figure 1. The greater strength of karish cheese gels produced from crosslinked milk with increasing TGase in the treated samples with 1.5 and 2 U $g^{-1}$ protein TGase was analyzed, showing a well-organized protein network with smaller pores in the treated product. This more stable structure was the result of the reduced permeability of the gel formed by the action of TGase. These results agree with those of Cadavid et al. (2020), who studied the effect of TGase treatment on the functional properties of semi-skimmed cow milk cheese through its crosslinking activity, where TGase appeared to be able to enhance the cheese microstructure by modifying the protein aggregation pattern.

### 3.3. SDS-PAGE

The electrophoretic separation of karish cheese samples in polyacrylamide gel was performed to confirm changes in the milk proteins in karish cheese produced with the addition of TGase. The SDS-PAGE examination of karish cheese samples treated with TGase indicated the crosslinking of proteins (Figure 2). These results were similar to those of Ozer et al. (2007). Figure 2 shows that as the concentration of TGase increased, the bands corresponding to casein fractions became less intense. According to the results of Cadavid et al. (2020), the increase in the molecular mass of the casein fractions might be attributed to a crosslinking process elicited by the presence of TGase. The number of monomeric whey proteins (α-lactalbumin and β-lactoglobulin) in control karish cheese samples was compared to those in TGase-treated samples. With the addition of TGase, the number of monomeric whey proteins reduced, and several new compounds were formed that did not join the stacking gel due to their molecular weight. The partial deamidation of glutamine and ε-amino groups by TGase treatment reduced the surface hydrophobicity of protein molecules and enhanced electrostatic repulsion. The consequent change in the isoelectric point of whey protein predicted a change in the solution consistency index (Gauche et al., 2008).

### 3.4. Organoleptic Properties

Enzymes are employed to improve the overall quality of foods (Fernandes, 2010) in a variety of food and beverage industries, including cheese manufacturing

**Table 1.** Mean values[a] for chemical composition of control and TGase-treated (1.0, 1.5, and 2 U $g^{-1}$ protein) karish cheese samples.

| Samples | Ash% | Protein% | Fat% | acidity% |
|---|---|---|---|---|
| Control | 4.50±0.07 [a] | 16.2±0.04 [b] | 0.23±0.05 [a] | 1.92±0.06 [a] |
| 1.0 U | 4.38±0.05 [a] | 16.5±0.07 [b] | 0.25±0.03 [a] | 2.05±0.02 [a] |
| 1.5U | 4.75±0.12 [a] | 17.1±0.05 [a] | 0.26±0.05 [a] | 1.95±0.02 [a] |
| 2.0U | 4.54±0.05[a] | 17.3±0.10 [a] | 0.27±0.02 [a] | 2.13±0.03 [a] |

[a,b,c] Values in the same columns having different superscripts were significantly different ($p \leq 0.05$).
[a]Mean values ± standard deviations.


ISSN-Internet 2197-411x OLCL 862804632

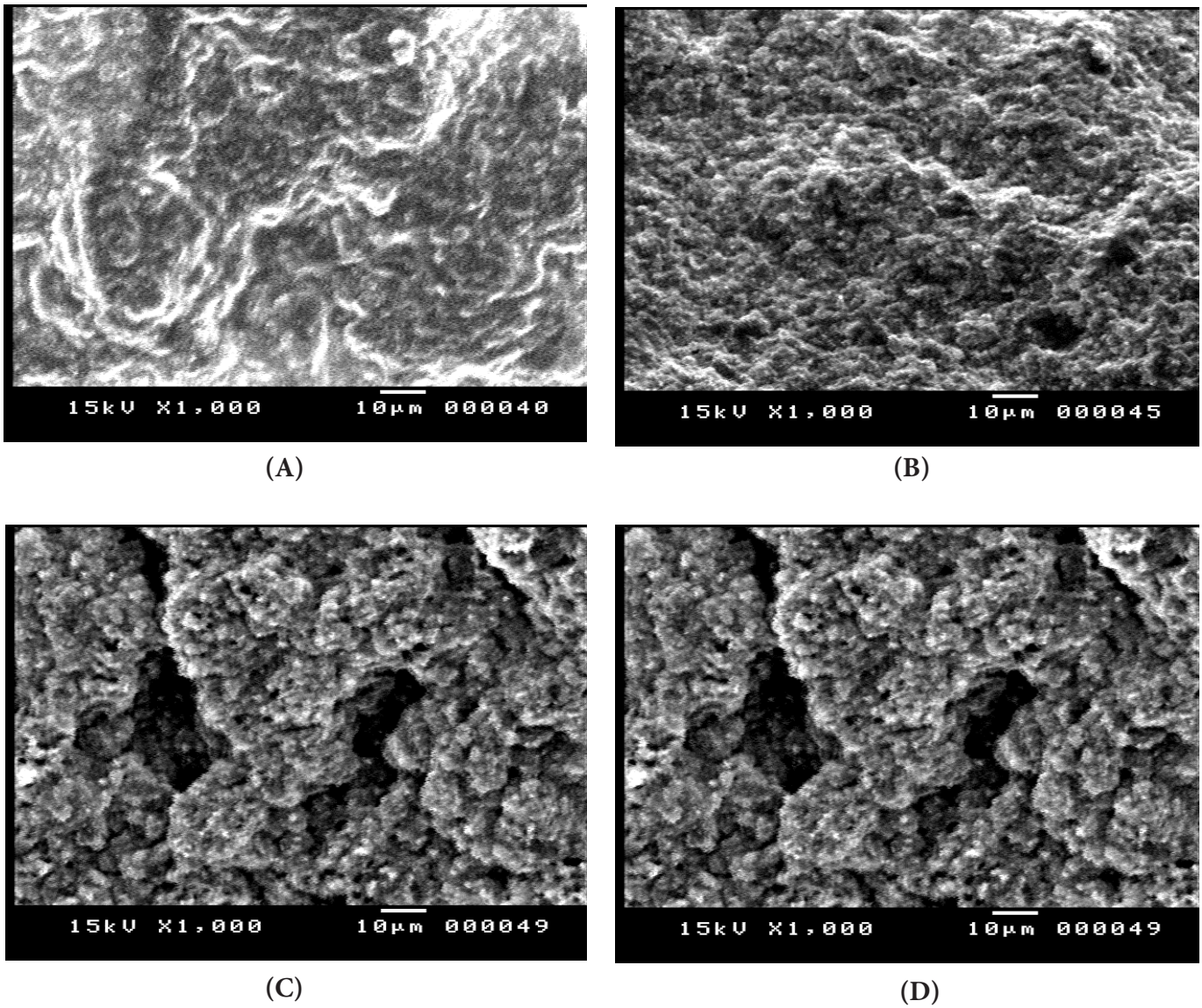


**Figure 1.** Scanning electron micrographs (1000×) of karish cheese samples: (A) control (without TGase treatment), (B) 1.0 U, (C) 1.5 U, and (D) 2 U g–1 protein TGase-treated samples.

(Gurung et al., 2013). The results of the organoleptic properties of the control and TGase-treated karish cheese samples are shown in (Table 2). The results revealed that there were no significant differences ($p \geq 0.05$) among karish cheese samples in colour and appearance and flavour; these results correspond with those of Hovjecki et al. (2021). Vice versa, data on the organoleptic properties revealed that there were significant differences ($p \leq 0.05$) among karish cheese samples in body and texture; these results correspond with those of Darwish et al. (2019). The (2.0 U) TGase-treated karish cheese sample had the highest body and texture of (34.9) whereas the control karish cheese sample had the lowest overall acceptability of (28.1). Furthermore, when the general acceptability of the sensory qualities was examined, there was a significant difference ($p \leq 0.05$) between the (1.5 U and 2.0 U) TGase treated karish cheese samples and control and (1 U). The overall acceptance of the control karish cheese sample was (87.1), whereas the 2.0 U TGase-treated karish cheese sample had the highest overall acceptability of (97.3). These results agree with those of Kumazawa and Miwa (2009), who discovered that using transglutaminase in cheese manufacturing had a substantial influence on texture and hardness. However, there was no significant difference in the colour, appearance, and flavour of karish cheese that was treated with TGase and the control samples; these findings coincide with those of Şanlı et al. (2011). Lastly, the addition of TGase to karish cheese

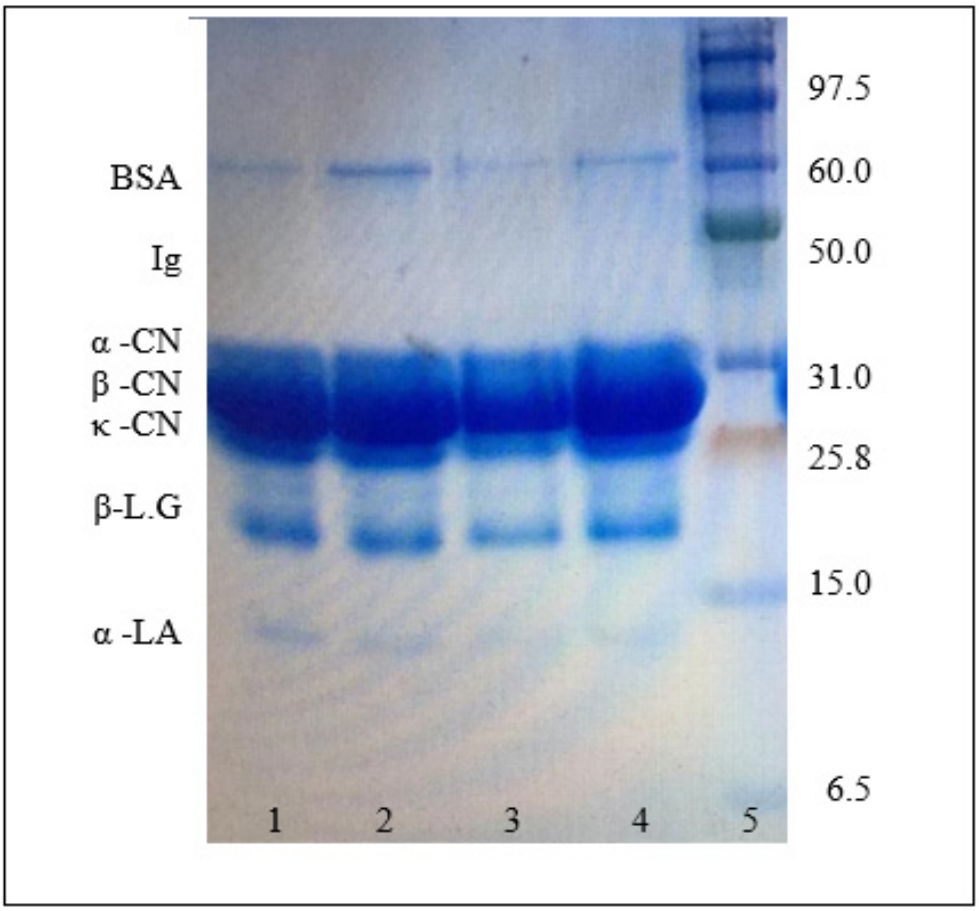


**Figure 2.** SDS-PAGE separation of proteins in karish cheese samples. Lane 1—control; lane 2—1 U; lane 3—1.5 U; lane 4—2 U; lane 5—molecular weight standard. BSA: Bovine serum albumin; Ig: Immunoglobulins; α-CN: α-casein; β-CN: β-casein; κ-CN: κ-casein; β-LG: β-lactoglobulin; α-LA: α-lactalbumin.

improved its manufacturing because the crosslinking proteins of TGase use the acyl exchange process to crosslink cysteine and lysine residues in a protein to create a proteinous network structure (Kumazawa & Miwa, 2009), which was visible in the organoleptic properties.

## 4. Conclusion

The use of TGase to crosslink proteins appears to be an efficient method of improving the techno functional features of karish cheese. TGase can synthesise high-molecular-weight polymers from milk proteins while preserving the chemical properties of karish cheese. The treatment of karish cheese with TGase increased gel firmness, which had a positive effect on karish cheese structure at the level 1.5–2.0 U $g^{-1}$ protein. These techniques have the potential to solve a wide range of production issues. In general, TGase treatment may be a suitable method for making modified karish cheese. The goal of this study was to improve the structure of karish cheese by utilizing a transglutaminase treatment. Physicochemical, microstructural, and organoleptic properties of karish cheese treated with TGase were studied and compared to cheese control samples that had not been treated with TGase. Comparison of results showed that the optimized cheese had a good structure. Lastly, our results are expected to pique dairy technologists and the cheese manufacturing industry in using TGase to enhance the structure of low-fat cheese.

## Conflict of Interests

The authors declare no conflict of interest.

## Acknowledgements

The present study is the result of independent research and has not been done with organizational financial support.


ISSN-Internet 2197-411x OLCL 862804632

**Table 2.** Organoleptic properties of karish cheese samples.

| | Color and Appearance | Flavor | Body and Texture | Overall Acceptability |
|---|---|---|---|---|
| | **15** | **50** | **35** | **100** |
| Control | 13.8 [a] | 45.9 [a] | 28.1 [d] | 87.1 [c] |
| 1.0 U | 13.9 [a] | 45.7 [a] | 33.5 [c] | 94.9 [b] |
| 1.5 U | 14.3 [a] | 46.3 [a] | 34.7 [b] | 96.9 [a] |
| 2.0 U | 14.1 [a] | 46.1 [a] | 34.9 [a] | 97.1 [a] |

[a,b,c,d] Values in the same columns having different superscripts were significantly different ($p \leq 0.05$).

## References


S.A., A. D. (2008). Origin, history and manufacturing process of Egyptian dairy products: an overview. Alexandria Journal of Food Science and Technology, 5(1), 51-62. doi: 10.21608/AJFS.2008.20141

Allam, M. G. M., Darwish, A. M. G., Ayad, E. H. E., Shokery, E. S., & Darwish, S. M. (2017). Lactococcus species for conventional Karish cheese conservation. LWT-Food science and Technology, 79, 625-631. doi: 10.1016/j.lwt.2016.11.032

Aloğlu, H. Ş., & Öner, Z. (2013). The effect of treating goat's milk with transglutaminase on chemical, structural, and sensory properties of labneh. Small Ruminant Research, 109(1), 31-37. doi: 10.1016/j.smallrumres.2012.10.005

Helrich, K. (1990). Official methods of analysis, 15th Edition. USA: Association of Official Analytical Chemists.

Horwitz. W. (2000). Official method of analysis of AOAC international, 17th Edition. USA: Association of Official Analytical Chemists.

Bozzola, J. J., & Russell, L. D. (1999). Electron microscopy: principles and techniques for biologists. USA: Jones and Bartlett Publishers.

Cadavid, A. M., Bohigas, L., Toldrà, M., Carretero, C., Parés, D., & Saguer, E. (2020). Improving quark-type cheese yield and quality by treating semi-skimmed cow milk with microbial transglutaminase. LWT, 131, 109756. doi: 10.1016/j.lwt.2020.109756

Danesh, E., Goudarzi, M., & Jooyandeh, H. (2018). Transglutaminase-mediated incorporation of whey protein as fat replacer into the formulation of reduced-fat Iranian white cheese: physicochemical, rheological and microstructural characterization. Journal of Food Measurement and Characterization, 12(3). doi: 10.1007/s11694-018-9858-5

Darwish, M. S., Elawady, A., & Mostafa, M. S. (2019). Extraction, Purification and Characterization of Transglutaminase from Silver Beet Leaves and its Effect on Sensory, Chemical and Rheological Properties of Kareish Cheese. Journal of Food and Dairy Sciences, 10(12), 495-501. doi: 10.21608/jfds.2019.71368

El-Baradei, G., Delacroix-Buchet, A., Pery, P., & Ogier, J.-C. (2005). Identification of bacterial communities of Egyptian Karish cheese using molecular footprinting tools. Journal of Dairy Science, 33(1), 25-34. Retrieved from https://www.researchgate.net/publication/259233401_Identification_of_Bacterial_Communities_of_Egyptian_Karish_Cheese_Using_Molecular_Fingerprinting_Tools

El-Kiyat, W., Laurenthia, E., Michaela, J., & Pari, R. F. (2021). Recent Advances in the Use of Transglutaminase in Cheese Production. ASEAN Journal on Science and Technology for Development, 38(2), 83–88. doi: 10.29037/ajstd.675.

Ezzel-Din, A. (1978). Studies on Kareish cheese making. Egypt: Cairo University.

Farnsworth, J. P., Li, J., Hendricks, G. M., & Guo, M. R. (2006). Effects of transglutaminase treatment on functional properties and probiotic culture survivability of goat milk yogurt. Small Ruminant Research, 65(1-2),

113-121. doi: 10.1016/j.smallrumres.2005.05.036

Fernandes, P. (2010). Enzymes in food processing: a condensed overview on strategies for better biocatalysts. Enzyme Research, 2010. doi: 10.4061/2010/862537

Fling, S. P., & Gregerson, D. S. (1986). Peptide and protein molecular weight determination by electrophoresis using a high-molarity tris buffer system without urea. Analytical Biochemistry, 155(1), 83-88. doi: 10.1016/0003-2697(86)90228-9

García-Gómez, B., Vázquez-Odériz, M. L., Muñoz-Ferreiro, N., Romero-Rodríguez, M. Á., & Vázquez, M. (2019). Interaction between rennet source and transglutaminase in white fresh cheese production: Effect on physicochemical and textural properties. LWT, 113, 108279. doi: 10.1016/j.lwt.2019.108279

Gauche, C., Vieira, J. T. C., Ogliari, P. J., & Bordignon-Luiz, M. T. (2008). Crosslinking of milk whey proteins by transglutaminase. Process Biochemistry, 43(7), 788-794. doi: 10.1016/j.procbio.2008.04.004

Gharibzahedi, S. M. T., Koubaa, M., Barba, F. J., Greiner, R., George, S., & Roohinejad, S. (2018). Recent advances in the application of microbial transglutaminase crosslinking in cheese and ice cream products: A review. International Journal of Biological Macromolecules, 107, 2364-2374. doi: 10.1016/j.ijbiomac.2017.10.115

Gurung, N., Ray, S., Bose, S., & Rai, V. (2013). A broader view: microbial enzymes and their relevance in industries, medicine, and beyond. BioMed Research International, 2013. doi: 10.1155/2013/329121

Hovjecki, M., Miloradovic, Z., Mirkovic, N., Radulovic, A., Pudja, P., & Miocinovic, J. (2021). Rheological and textural properties of goat's milk set type yoghurt as affected by heat treatment, transglutaminase addition and storage. Journal of the Science of Food and Agriculture, 101(14). doi: 10.1002/jsfa.11242

Clark., S., Costello., M., Drake., M.-A., & Bodyfelt., F. (2009). The Sensory Evaluation of Dairy Products. USA: Springer.

Imran, M., Khan, H., Hassan, S. S., & Khan, R. (2008). Physicochemical characteristics of various milk samples available in Pakistan. Journal of Zhejiang University Science B, 9(7), 546-551. doi: 10.1631/jzus.B0820052

Kalit, S., Havranek, J. J., Kaps, M., Perko, B., & Curik, V. C. (2005). Proteolysis and the optimal ripening time of Tounj cheese. International Dairy Journal, 15(6-9), 619-624. doi: 10.1016/j.idairyj.2004.09.010

Kashiwagi, T., Yokoyama, K.-I., Ishikawa, K., Ono, K., Ejima, D., Matsui, H., & Suzuki, E.-I. (2002). Crystal Structure of Microbial Transglutaminase from Streptoverticillium mobaraense. Journal of Biological Chemistry, 277(46), 44252-44260. doi: 10.1074/jbc.M203933200

Kieliszek, M., & Misiewicz, A. (2014). Microbial transglutaminase and its application in the food industry. A review. Folia Microbiologica, 59(3), 241-250. doi: 10.1007/s12223-013-0287-x

Kumazawa, Y., & Miwa, N. (2009). Process for producing cheese curd. Retrieved from https://patentimages.storage.googleapis.com/55/8d/c2/411724801d1172/US7504119.pdf

Lauber, S., Henle, T., & Klostermeyer, H. (2000). Relationship between the crosslinking of caseins by transglutaminase and the gel strength of yoghurt. European Food Research and Technology, 210(5), 305-309. doi: 10.1007/s002170050554

Martins, I. M., Matos, M., Costa, R., Silva, F., Pascoal, A., Estevinho, L. M., & Choupina, A. B. (2014). Transglutaminases: recent achievements and new sources. Applied Microbiology and Biotechnology, 98(16), 6957-6964. doi: 10.1007/s00253-014-5894-1

Monsalve-Atencio, R., Sanchez-Soto, K., Chica, J., Echavarría, J. A. C., & Vega-Castro, O. (2022). Interaction between phospholipase and transglutaminase in the production of semi-soft fresh cheese and its effect on the yield, composition, microstructure and textural properties. LWT, 154, 112722. doi: 10.1016/j.lwt.2021.112722

Ozer, B., Kirmaci, H. A., Oztekin, S., Hayaloglu, A.


ISSN-Internet 2197-411x OLCL 862804632

A., & Atamer, M. (2007). Incorporation of microbial transglutaminase into non-fat yogurt production. International Dairy Journal, 17(3), 199-207. doi: 10.1016/j.idairyj.2006.02.007

Pereira, C. I., Gomes, A. M. P., & Malcata, F. X. (2009). Microstructure of cheese: Processing, technological and microbiological considerations. Trends in Food Science & Technology, 20(5), 213-219. doi: 10.1016/j.tifs.2009.02.006

Pesic, M. B., Barac, M. B., Stanojevic, S. P., Ristic, N. M., Macej, O. D., & Vrvic, M. M. (2012). Heat induced casein–whey protein interactions at natural pH of milk: A comparison between caprine and bovine milk. Small Ruminant Research, 108(1-3), 77-86. doi: 10.1016/j.smallrumres.2012.06.013

Rachel, N. M., & Pelletier, J. N. (2013). Biotechnological applications of transglutaminases. Biomolecules, 3(4), 870-888. doi: 10.3390/biom3040870

Razeghi, F., & Yazdanpanah, S. (2020). Effects of free and encapsulated transglutaminase on the physicochemical, textural, microbial, sensorial, and microstructural properties of white cheese. Food Science & Nutrition, 8(7), 3750-3758. doi: 10.1002/fsn3.1663

Rodriguez-Nogales, J. M. (2006). Enhancement of transglutaminase-induced protein cross-linking by preheat treatment of cows' milk: a statistical approach. International Dairy Journal, 16(1), 26-32. doi: 10.1016/j.idairyj.2005.01.003

Şanlı, T., Sezgin, E., Deveci, O., Şenel, E., & Benli, M. (2011). Effect of using transglutaminase on physical, chemical and sensory properties of set-type yoghurt. Food Hydrocolloids, 25(6), 1477-1481. doi: 10.1016/j.foodhyd.2010.09.028

Sayadi, A., Madadlou, A., & Khosrowshahi, A. (2013). Enzymatic cross-linking of whey proteins in low fat Iranian white cheese. International Dairy Journal, 29(2), 88-92. doi: 10.1016/j.idairyj.2012.10.006

Soltani, M., Saremnezhad, S., Faraji, A. R., & Hayaloglu, A. A. (2022). Perspectives and recent innovations on white cheese produced by conventional methods or ultrafiltration technique. International Dairy Journal, 125, 105232. doi: 10.1016/j.idairyj.2021.105232

Tolkach, A., & Kulozik, U. (2005). Fractionation of whey proteins and caseinomacropeptide by means of enzymatic crosslinking and membrane separation techniques. Journal of Food Engineering, 67(1-2), 13-20. doi: 10.1016/j.jfoodeng.2004.05.058

Topcu, A., Bulat, T., & Özer, B. (2020). Process design for processed Kashar cheese (a pasta-filata cheese) by means of microbial transglutaminase: Effect on physical properties, yield and proteolysis. LWT, 125, 109226. doi: 10.1016/j.lwt.2020.109226

Wang, L., Yu, B., Wang, R., & Xie, J. (2018). Biotechnological routes for transglutaminase production: recent achievements, perspectives and limits. Trends in Food Science & Technology, 81(1), 116-120. doi: 10.1016/j.tifs.2018.09.015

Zhang, L., Zhang, L., Yi, H., Du, M., Ma, C., Han, X., Feng, Z., Jiao, Y., & Zhang, Y. (2012). Enzymatic characterization of transglutaminase from Streptomyces mobaraensis DSM 40587 in high salt and effect of enzymatic cross-linking of yak milk proteins on functional properties of stirred yogurt. Journal of Dairy Science, 95(7), 3559-3568. doi: 10.3168/jds.2011-5125