

Microbes from Antarctica as a source for understanding cold adaptive Halal enzymes

Muhammad Asyraf Abd Latip¹, Noor Faizul Hadry Nordin^{2*} and Gomez-Fuentes C³¹Biotechnology Engineering Department, Kulliyah of Engineering, International Islamic University Malaysia, Gombak, 531000, Malaysia²International Institute for Halal Research & Training (INHART), International Islamic University Malaysia, Gombak, 531000, Malaysia³CIMAA, Department of Chemical Engineering, Faculty of Engineering, University of Magallanes, Punta Arenas, Chile

Abstract: Enzymes are widely used in various industries as they exhibit many outstanding benefits. They function to accelerate the reaction process which is more advantageous compared with chemicals as catalysts. However, the halal status of enzymes has been argued especially in food industries. This is because some of the enzymes originate from animal sources. The main concern with this issue is regarding the compatibility of these sources with the Islamic law. Beside animals and plants, microorganisms also play a vital role in producing various types of enzymes naturally. In relation to their halalness, enzymes extracted from microbes are considered as halal. Antarctica is a new frontier with a diverse microbial community that shows a potential for bio prospecting. The extremophiles existing in this region produce enzymes that can function in extreme conditions. Some of these enzymes suit the industrial requirement. The microorganisms obtained from Antarctica are very useful for harnessing and bio prospecting of such enzymes due to their great potential and diverse applications in many industrial fields in the future.

Keywords: Antarctica; cold-active enzyme; psychrophiles; biotechnology; Halal

Received: 18th September 2019

Accepted: 19th October 2019

Published Online: 06th November 2019

*Correspondence:

Noor Faizul Hadry Nordin, International Institute for Halal Research & Training (INHART), International Islamic University Malaysia, Gombak, 531000, Malaysia; faizul@iiu.edu.my

Citation: Abd Latip MA, Hadry Nordin NF and Gomez-Fuentes C. Microbes from Antarctica as a source for understanding cold adaptive halal enzymes. *J Halal Ind Serv* 2019; 2(1): a0000040

Introduction

The contribution of enzymes is very crucial either in different industries or in daily life applications (Choi *et al.*, 2015; Juturu & Wu, 2014; Adrio & Demain, 2014). Generally, an enzyme is a tool to help accelerate the reaction process. It has been used as a biocatalyst to substitute the chemical catalyst (Fersht, 1999). Although the biocatalyst is costlier, it shows more efficiency, selectivity and environmental friendliness compared with the chemical catalyst (Blamey *et al.*, 2017; Nealon *et al.*, 2015).

However, the involvement of this biocatalyst in the food industry has been debated in terms of its halal status (Adapa *et al.*, 2014). The word 'Halal' is a Quranic term that means permissible or lawful by the Islamic law and mostly refers to foods and drinks (Featherstone, 2015; Riaz & Chaudry, 2003). The opposite of Halal is Haram which means prohibited or unlawful. Halal in the food industry is very important especially for Muslim consumers. The awareness about halal food manufacturing or processing has been issued in some publications (Vanany *et al.*, 2018; Ismail *et al.*, 2018; Thadathil & Velappan, 2014). In order for a product to acquire the Halal certificate or be labelled as Halal, there are many aspects to consider, starting from the raw material until the end product including the packaging process (Wahab, 2004). The food ingredients, additives and process aids must fulfill the Halal requirements. As the enzyme is also involved in food processing, it must be certified as Halal as well in order to certify the end products. Many enzymes have been applied in various food industries including dairy, cheese, syrup, starch, baking, brewing, meat, wine and juice (Liu *et al.*, 2015; Merin & Morata, 2015; Ranjan *et al.*, 2016; Dura & Rosell, 2016;

Wang *et al.*, 2016). Some classes of enzymes that are important in the food industry include alpha-amylase, glucoamylase, beta-glucanase, lipase, papain, chymosin, proteases, pectinase, lactase, decarboxylase, glucose oxidase and cellulase, amyloglucosidase and phytase (Wang *et al.*, 2016; Ranjan *et al.*, 2015; Hosseinipour *et al.*, 2015; Tapre & Jain, 2014).

Halal and Non-Halal Enzymes Sources

The most important criteria for the enzyme to be certified as Halal is its source. The sources of enzymes can vary between plants, animals or microorganisms. Since some of the enzymes are extracted from animals, their Halal status has been skepticized. These enzymes could be labelled Haram if the sources are not fully compliant with the Islamic laws. Some of the enzymes that are extracted from animals include catalase, chymotrypsin, lipase, rennet, trypsin, chymosin, reductase and cathepsin (Table 1). The Islamic law has listed the following sources as Haram according to the Quran and Hadith; i) carrion, ii) blood, iii) pig, iv) animal not slaughtered by the name of Allah S.W.T, v) animal killed by beating or fallen, vi) donkey, vii) fangs or claw predators, viii) poisonous animals and ix) animals living both in water and on land (Kashim *et al.*, 2015).

However, if the enzymes are extracted from plants, it is evident that the sources are Halal. But as mentioned earlier, in order to certify an enzyme as Halal, the extracting process, the chemical and the aider used must also be certified as Halal.

One of the important sources of enzymes is microorganisms. Generally, there are four stages in producing enzymes industrially from microorganisms. There is the selection

of the enzyme, formulating the medium and the process of production and purification of the enzyme. For the production process stage, most industries in advanced countries use the submerged and solid-state fermentation (Pandey et al., 1999). This process is preferable because of its lower cost of production and low contamination compared with others. Besides, the production rate of the enzyme can be increased by optimizing the conditions for the growth of the microorganisms. When dealing with microorganisms that can cause a pathogenic effect on humans, the hygienic production process is an important part of the Halal concept. This is because the concept of Halal Tayyib also refers to the quality of the product and the safety of the consumers (Alzeer et al., 2018). Scholars have discussed and listed the Halal requirements for the fermentation process including the hygienic process, waste control and elimination of contaminants (Riaz & Chaudry, 2003).

Table 1. Enzymes extracted from animals for industries and biotechnology applications

Enzymes	Microorganism	Opt. Temp	References
Catalase	<i>Bubalus bubalis</i>	30	Nadeem et al., 2015
		25–40	
	<i>Camelus dromedarius</i>		Al-Bar, 2012
Chymotrysin	<i>Bos taurus</i>	30	Alptekin et al., 2008
	<i>Bos taurus</i>	25	Wirnt 1965
Lipase	<i>Bos taurus</i>	37	Shahani et al., 1976
	<i>Sus scrofa</i>	37	Wilcox et al., 2014
Chymosin	<i>Bos buffali</i>	37	Malak et al., 1996
	<i>Capra hircus</i>	30	Kumar et al., 2006
Trypsin	<i>Ovis ammon</i>	60	Li et al., 2012
	<i>Sus scrofa</i>	37	Deepthi et al., 2001
	<i>Boops boops</i>	55	
Cathepsin	<i>Sus scrofa</i>	37	Barkia et al., 2010
	<i>Sus scrofa</i>	37	
Carboxylesterase	<i>Bungarus fasciatus</i>		Ramos et al., 2008
Acetylcholines-terase	<i>Crotalus atrox</i>	37	Henke et al., 2003
			Godoy et al., 2005
Phosphodies-terase	<i>Crotalus atrox</i>	55	
	<i>Sus scrofa</i>	37	Bowman et al., 2001
Metalloprotein-ase	<i>Sus scrofa</i>	37	Willis et al., 1988
Lyase			Schauer & Wember, 1996
Syntase			Poyck et al., 2008

*Opt. Temp. – Optimum temperature

Fermentation is the best alternative way to substitute enzymes extracted from animals. For example, rennet is an important enzyme in cheese production. The source of rennet is originally from the calf. As an alternative, scientists have extracted bromelain from pineapples, transformed it into a microorganism and proceeded with the fermentation process for a large-scale production (Arshad et al., 2014).

Some enzymes have very specific characteristics that distinguish them from other common or conventional enzymes. They have the capability to operate under abnormal or extreme conditions such as temperature, salinity, pH etc. These unique features of an enzyme can be very beneficial if it is applied appropriately. Most enzymes that are equipped with these qualities are extracted from extremophiles microorganisms (Siddiqui, 2015; Dalmaso et al., 2015; Urbietta et al., 2015). Various classes of enzymes with these characteristics have been extensively investigated and applied in different fields.

Antarctica as a New Halal Enzymes Source

The Antarctic is located at the South Pole of the Earth and to date, there is no single permanent population on it – Figure 1. The Antarctic is an isolated region that is surrounded by the Southern Oceans. This continent also covers some sub-Antarctic islands like Campbell Island, Heard Island, and South Georgia, some of which are north of the Antarctic Convergence (Anisimov et al., 2001).

After the World War II, there were about seven scientific expeditions to the Antarctic to study microbial diversity. Through the French expedition on 1903–1905, the first microorganism had been discovered on this continent and is isolated from the penguin's intestines (Tsiklinsky, 1908). Besides, an interesting finding that has been discovered from the Blood Falls outflow is a type of active bacteria that lives by harvesting energy from the bedrock or respiring from Fe (III) or SO₄²⁻ (Mikuchi et al., 2009).

This sub glacial environment is part of the Earth's biosphere that remains largely unexplored. The number of scientific studies has been rising in order to enhance the understanding of microorganism communities that survive the harsh Antarctic environment (Matsui et al., 2017).



Figure 1. Antarctica is more pristine than the Arctic because it lacks any human activity. The picture was taken during Chilean Antarctica Expedition 2018 (Photo courtesy of N.F.Hadry Nordin)

Many researchers have switched their focus to the discovery of the cold-active enzyme extracted from psychrophiles because it shows more valuable potentials compared with the

mesophilic enzyme. Generally, the advantages of this cold-active enzyme are many. It has a high efficient catalytic activity at low and moderate temperatures compared with the mesophilic enzyme (Cavicchioli et al., 2011). It also has a thermolabile characteristic which means it yields higher activities at lower temperatures, but deactivates when the temperature is increased. For that reason, its inactivation can be easily controlled, thus minimizing the loss of volatile compounds and undesirable chemical reactions. Furthermore, it confers a lower activation energy and maintains the working efficiency at a low temperature and therefore reduces and saves the energy consumption and production cost. Besides, its specific and selective activity can shorten the processing time and reduce the concentration of the enzyme used (Siddiqui & Cavicchioli, 2006).

In the food industry, enzymes play many important functions in preparing and processing the food. There are some potential cold-active enzymes that can replace the commercial enzymes. This is because a high temperature processing may change the original taste of the foods. By employing enzymes with a lower energy activation and a lower working temperature, this can reduce undesirable chemical reactions while processing the food. Thus, any spoilage in the nutritional value or bacterial contamination can be avoided (Gerday et al., 2000). The most prominent feature of enzymes is that they preserve the original taste and condition of the food. For instance, in cheese making, rennet is very important in the process. Rennet contains more than 90% chymosin and other components like pepsin and lipase which function to solidify the milk (Kumar et al., 2001). The optimum temperature for this enzyme is around 45°C. If a cold-active enzyme can substitute the rennet, the production cost of cheese can be reduced by decreasing the energy used to maintain the enzyme's optimum temperature. Table 2 shows the latest finding of the cold-active enzymes that are extracted from microorganisms which inhibit Antarctica.

Cold-Active Enzyme Applications

Harnessing enzymes for bio prospecting is very beneficial for industrial applications nowadays. Generally, there are six classes of enzymes; Oxidoreductases, Transferases, Hydrolases, Lyases, Isomerases and Ligases.

Oxidoreductases

Alcohol dehydrogenase is the most potential subclass for industrial and biotechnology applications in oxidoreductases. This enzyme has been studied in vinegar production through acetic acid fermentation from ethanol (Zheng et al., 2015), ethanol biosensor (Gomez-Anquela et al., 2015) and lignocellulosic biomass for renewable fuel production (Quaglia et al., 2013). In addition, it is also mostly used as a biocatalyst in enantioselective chemical synthesis for agriculture and pharmaceutical industries (Elleuche et al., 2013). Cold-active alcohol dehydrogenase can possibly be used in synthesizing enantiomer with high vapour pressure in order to minimize the loss of this volatile compound in the processing. Another subclass, oxygenase is widely studied for bioremediation and biodegradation process (Tavakoli & Hamzah, 2017; Al-Alaq et al., 2016). Psychrophiles that exhibit cold active oxygenase have the potential for bioremediation application in countries with a colder environment. Currently, NAD(P)⁺-independent oxidoreductase is studied for microbial fuel cell. However, the currently available oxidoreductases

require an optimum temperature of 30°C and above (Ren et al., 2017; Lin et al., 2017). By using cold-active enzymes in the process, the optimum temperature can be reduced, and the energy can be saved.

Table 2. Latest findings of cold-active enzymes extracted from Antarctica

Enzymes	Microorganism	Opt. Temp.	References
Amino-transferase	<i>Psychrobacter</i> sp	55	Bujacz <i>et al.</i> , 2015
Cellulase	Antarctica bacterium	40	Wang <i>et al.</i> , 2015
Chitinase	<i>Lecanicillium muscarium</i>	40	Fenice, 2016
Glutaredoxin	<i>Pseudoalteromonas</i> sp	30	Wang <i>et al.</i> , 2014
	<i>Pseudoalteromonas</i> sp	40	Shi <i>et al.</i> , 2014
Glutathione-s-transferase	<i>Rhodotorula mucilaginosa</i>	50	Yu <i>et al.</i> , 2015
Phytase	Antarctic microbes	20-40	Matsui <i>et al.</i> , 2017
Protease	<i>Arthrobacter agilis</i>	30	Kim <i>et al.</i> , 2017
Amylase	Antarctic psychrophilic strain	30	Li <i>et al.</i> , 2015
Agarase	<i>Halorubrum lacusprofundi</i>	50	Karan <i>et al.</i> , 2013
	<i>Exiguobacterium antarcticum</i>	30	Crespin <i>et al.</i> , 2016
Galactosidase	<i>Micrococcus antarcticus</i>	25	Miao <i>et al.</i> , 2016
Glucosidase			

*Opt. Temp. – Optimum temperature

Transferases

For the transferases enzyme class, serine hydroxymethyltransferase (SHMT) is important for enantiomers synthesis (Angelaccio et al., 2012). SHMT is involved in the reversible conversion of L-serine and tetrahydropteroylglutamate (H4PteGlu) to glycine and 5,10-methylenetetrahydropteroylglutamate (5,10-CH₂-H4PteGlu) (Florio et al., 2011). Processing in lower temperatures can minimize the retroaldol reaction and maintain synthesis with a maximized capacity. In this situation, cold-active SHMT from the psychrophiles has a lot of advantages. Besides, cold-induced glutathione s-transferase (GST) has been successfully extracted

from the Antarctic bacterium strain and has the potential to be applied in agricultural industries (Wang et al., 2017). Researchers discovered that GST increases the plant tolerance towards salt, stress and lower temperature environments (Seppanen et al., 2000). A potential transgenic plant with a high resistance toward cold temperatures could be valuable for agricultural purposes especially in colder countries. In addition, aspartase is a common enzyme that has been used to produce food additives and artificial sweeteners such as aspartame.

Hydrolases

Among all classes of enzymes, hydrolase is the most popular class in terms of its biotechnological and industrial applications. Cold-active protease has been implemented in a wide range of applications such as washing powders, food industries and pharmaceuticals because it can achieve a higher activity level at lower temperatures (Joshi & Satyanarayana, 2013). Cold-active alpha-amylase has been extracted and characterized for a detergent formulation that is suitable for laundry washing with ambient temperature (Ranjan et al., 2016; Roohi et al., 2013; Caf et al., 2014). In the food industry, enzymes play several important functions in the preparing process. There are some potential cold-active enzymes that can replace the current commercial enzymes; for example, β -glucosidase in plant-based food (Mioia et al., 2016), protease for meat tenderizer (Mageswari et al., 2017), and esterase for food fermentation (Esteban-Torres et al., 2014). By using cold-active enzymes, undesirable chemical reactions that may occur at higher temperatures can be reduced while processing the food. Thus, any spoilage in the nutritional value or bacterial contamination can be avoided while preserving the original taste and condition of the food (Gerday et al., 2000). A novel finding of glucosidase from the Antarctic regions showed that this enzyme can directly convert cellulose into glucose and produce ethanol without fermentation (Crespin et al., 2016). Significant to this cellulosic ethanol, the conversion can be made at a lower temperature and the production cost can be reduced. In the molecular study, enzyme phosphatase is absolutely important, especially in DNA studies. Researchers have discovered a cold-active alkaline phosphatase that has been extracted from shrimp in the northern region called *Pandalus borealis* (Nilsen et al., 2001). This shrimp alkaline phosphatase (rSAP) is a heat labile enzyme that can be deactivated at 65°C in 5 minutes. Recently, a metagenomic study has discovered another cold active alkaline phosphatase extracted from psychrophiles (Lee et al., 2015). Therefore, this enzyme can be produced commercially at a lower cost by using bacteria rather than shrimp.

Lyases

From a biotechnological perspective, enzyme decarboxylase from the lyases class has a significant function in the food industry and feedstock production. In cheese manufacturing, decarboxylase has a role in the aroma development in the cheese ripening process (Wang et al., 2017). By using a cold-active enzyme, it can preserve the texture of the cheeses because storing them at a high temperature for a long period will deteriorate their texture. Keto-acid decarboxylase contributes to the production of biofuel. Generally, these long chain branch alcohols like isobutanol, 2-methyl-1-butanol or 3-methyl-1-butanol are synthesized through fermentation processes like ethanol. But through biosynthesis amino acid

pathways, the production can be done without fermentation (Atsumi et al., 2008). The discovery of cold-active 2 keto acid decarboxylase with an optimum activity at 35 °C is advantageous in order to yield large-scale renewable energy at a lower cost (Wei et al., 2013).

Isomerases

In biotechnology applications, the isomerases class is important in the manufacturing of artificial sweeteners. Glucose isomerase is responsible for the conversion of D-glucose to D-fructose. The Thermostable enzyme has been characterized and improved in order to optimize the production of high fructose corn syrup (Liu et al., 2015). In addition, sucrose isomerase has been used in the production of artificial sweeteners such as isomaltulose (Mu et al., 2014). Besides, in the biofuel technology, xylose isomerase is an important enzyme in the production of ethanol as a clean and renewable source of energy. A recent study shows that this enzyme has been engineered in yeast to optimize ethanol production from lignocellulosic hydrolysates (Ko et al., 2016). Nevertheless, cold-active isomerases have many beneficial aspects economically and environmentally. Also, they are energy saving and easier to manoeuvre. There were fewer discoveries of cold-active isomerase, but triose phosphate isomerase (TIM) was among the earliest crystal structures of cold-active enzymes extracted from psychrophilic bacterium to ever be elucidated (Alvarez et al., 1998).

Ligases

The final class are ligases. DNA ligase is important to join two DNA components together, especially in the recombinant process. Cold-adapting DNA ligase has been discovered and works efficiently at a lower temperature of 18°C compared with the commercial DNA ligase which requires a higher temperature of 30°C (Georlette et al., 2000). Besides, its thermolabile property makes the enzyme easier to deactivate without putting the DNA structure at risk of degradation. Another cold active ligase that has been isolated from the same microorganism, *Pseudoalteromonas haloplanktis* is γ -glutamyl-cysteine ligase (Albino et al., 2014).

Conclusion

It is very beneficial to harness, and bio prospect these enzymes due to their remarkable potentials and diverse applications in many industrial fields in the future. However, identifying and extracting specific cold-active enzymes with high stability levels is required. Besides, through genetic strain modification and improvement, these cold-active enzymes would play an important role in various biotechnological industries and applications. In the future, these valuable enzymes in biotechnology applications would have the potential to enhance the enzyme market alongside the thermostable enzymes.

Conflict of Interest

The authors declare that there is no conflict of interest in this work.

Acknowledgement

This work was supported by research grants from Yayasan Penyelidikan Antartika Sultan Mizan (YPASM) 2015 and RIGS 16-332-0496.

Reference

- Adapa, V., Ramya, L. N., Pulicherla, K. K., et al. (2014). Cold active pectinases: Advancing the food industry to the next generation. *Applied Biochemistry And Biotechnology*, 172(5): 2324–2337.
- Adrio, J., & Demain, A. (2014). Microbial enzymes: Tools for biotechnological processes. *Biomolecules*, 4(1): 117–139.
- Al-Bar, O. A., (2012). Characterization of partially purified catalase from camel (*Camelus dromedarius*) liver. *African Journal of Biotechnology*, 11(40): 9633–9640.
- Al-Alaq, F. T., Abdulazeem, L., Al-Dahmoshi, H. O. M., et al. (2016). PCR-based investigation of oxygenase among crude oil degrading bacteria in Hilla city, Iraq. *International Journal of Pharm Tech Research*, 9(5): 284–291.
- Albino, A., De Angelis, A., Marco, S., et al. (2014). The cold-adapted γ -glutamyl-cysteine ligase from the psychrophile *Pseudoalteromonas haloplanktis*. *Biochimie*, 104: 50–60.
- Alptekin, Ö., Tükel, S.S. & Yildirim, D., (2008). Immobilization and characterization of bovine liver catalase on eggshell. *Journal of the Serbian Chemical Society*, 73(6): 609–618.
- Alvarez, M., Zeelen, J.P., Mainfroid, V., et al. (1998). Triose-phosphate Isomerase (TIM) of the psychrophilic bacterium *Vibrio marinus* kinetic and structural properties. *Journal of Biological Chemistry*, 273(4): 2199–2206.
- Alzeer, J., Rieder, U., & Hadeed, K. A. (2018). Rational and practical aspects of Halal and Tayyib in the context of food safety. *Trends in Food Science & Technology*, 71: 264–267.
- Angelaccio, S., Florio, R., Consalvi, et al. (2012). Serine hydroxymethyltransferase from the cold adapted microorganism *Psychromonas ingrahamii*: A low temperature active enzyme with broad substrate specificity. *International Journal of Molecular Sciences*, 13(2): 1314–1326.
- Anisimov, O., Fitzharris, B., Hagen, J. O., et al. (2001). Polar regions (Arctic and Antarctic). *Climate Change*, 801–841.
- Arshad, Z. I. M., Amid, A., Yusof, F., et al. (2014). Bromelain: An overview of industrial application and purification strategies. *Applied Microbiology and Biotechnology*, 98(17): 7283–7297.
- Atsumi, S., Hanai, T., & Liao, J. C. (2008). Non-fermentative pathways for synthesis of branched-chain higher alcohols as biofuels. *Nature*, 451(7174): 86.
- Barkia, A., Bougatef, A., Nasri, R., et al. (2010). Trypsin from the viscera of Bogue (Boops boops): Isolation and characterisation. *Fish Physiology and Biochemistry*, 36(4): 893–902.
- Bernbäck, S., Hernell, O., & Bläckberg, L. (1985). Purification and molecular characterization of bovine pregastric lipase. *European Journal of Biochemistry*, 148(2): 233–238.
- Blamey, J. M., Fischer, F., Meyer, H. P., et al. (2017). Enzymatic biocatalysis in chemical transformations: A promising and emerging field in green chemistry practice. In G. Brahmachari (Ed.), *Biotechnology of microbial enzymes* (pp. 347–403). Elsevier.
- Bowman, K. J., Pla, R. L., Guichard, Y., et al. (2001). Evaluation of phosphodiesterase I-based protocols for the detection of multiply damaged sites in DNA: The detection of abasic, oxidative and alkylative tandem damage in DNA oligonucleotides. *Nucleic Acids Research*, 29(20): e101–e101.
- Bujacz, A., Rutkiewicz-Krotewicz, M., Nowakowska-Sapota, K., et al. (2015). Crystal structure and enzymatic properties of a broad substrate-specificity psychrophilic aminotransferase from the Antarctic soil bacterium *Psychrobacter* sp. B6. *Acta Crystallographica Section D: Biological Crystallography*, 71(3): 632–645.
- CAF, Y., Valipour, E., & Arikan, B. (2014). Isolation and characterization of alkaline, halotolerant, detergent-stable and cold-adaptive-amylase from a novel isolate *Bacillus* sp. Calp12-7. *International Journal of Current Microbiology and Applied Sciences*, 3(4): 950–960.
- Cavicchioli, R., Charlton, T., Ertan, H., et al. (2011). Biotechnological uses of enzymes from psychrophiles. *Microbial Biotechnology*, 4(4): 449–460.
- Choi, J. M., Han, S. S., & Kim, H. S. (2015). Industrial applications of enzyme biocatalysis: Current status and future aspects. *Biotechnology Advances*, 33(7): 1443–1454.
- Crespim, E., Zanphorlin, L. M., de Souza, F. H., et al. (2016). A novel cold-adapted and glucose-tolerant GH1 β -glucosidase from *Exiguobacterium antarcticum* B7. *International Journal of Biological Macromolecules*, 82: 375–380.
- Dalmaso, G., Ferreira, D., & Vermelho, A. (2015). Marine extremophiles: A source of hydrolases for biotechnological applications. *Marine Drugs*, 13(4): 1925–1965.
- Deepthi, S., Johnson, A., & Pattabhi, V. (2001). Structures of porcine β -trypsin–detergent complexes: The stabilization of proteins through hydrophilic binding of polydocanol. *Acta Crystallographica Section D: Biological Crystallography*, 57(11): 1506–1512.
- Dura, A., & Rosell, C. M. (2016). Enzymes in baking (pp. 295–314). Boca Raton, FL: CRC Press (Taylor & Francis Group).
- Elleuche, S., Fodor, K., Klippel, B., et al. (2013). Structural and biochemical characterisation of a NAD⁺-dependent alcohol dehydrogenase from *Oenococcus oeni* as a new model molecule for industrial biotechnology applications. *Applied Microbiology and Biotechnology*, 97(20): 8963–8975.
- Esteban-Torres, M., Mancheño, J. M., de las Rivas, B. et al. (2014). Characterization of a cold-active esterase from *Lactobacillus plantarum* suitable for food fermentations.

- Journal of Agricultural and Food Chemistry*, 62(22): 5126–5132.
- Featherstone, S. (Ed.). (2015). *A complete course in canning and related processes: Volume 3 Processing Procedures for Canned Food Products*. Woodhead Publishing.
- Fenice, M. (2016). The psychrotolerant Antarctic fungus *Lecanicillium muscarium* CCFEE 5003: A powerful producer of cold-tolerant chitinolytic enzymes. *Molecules*, 21(4): 447.
- Fersht, A. (1999). *Structure and mechanism in protein science: A guide to enzyme catalysis and protein folding*. Macmillan.
- Florio, R., di Salvo, M.L., Vivoli, M. et al. (2011). Serine hydroxymethyltransferase: A model enzyme for mechanistic, structural, and evolutionary studies. *Biochimica et Biophysica Acta (BBA)-Proteins and Proteomics*, 1814(11): 1489–1496.
- Georlette, D., Jonsson, Z. O., Van Petegem, F., et al. (2000). A DNA ligase from the psychrophile *Pseudoalteromonas haloplanktis* gives insights into the adaptation of proteins to low temperatures. *European Journal of Biochemistry*, 267(12): 3502–3512.
- Gerday, C., Aittaleb, M., Bentahir, M., et al. (2000). Cold-adapted enzymes: From fundamentals to biotechnology. *Trends in Biotechnology*, 18(3): 103–107.
- Godoy, S., Violot, S., Boullanger, P., et al. (2005). Kinetics study of *Bungarus fasciatus* venom acetylcholinesterase immobilised on a langmuir–blodgett proteo-glycolipidic bilayer. *ChemBioChem*, 6(2): 395–404.
- Gómez-Anquela, C., García-Mendiola, T., Abad, J. M., et al. (2015). Scaffold electrodes based on thioctic acid-capped gold nanoparticles coordinated Alcohol Dehydrogenase and Azure A films for high performance biosensor. *Bioelectrochemistry*, 106: 335–342.
- Henke, E., Bornscheuer, U. T., Schmid, R. D., et al. (2003). A molecular mechanism of enantio-recognition of tertiary alcohols by carboxylesterases. *ChemBioChem*, 4(6): 485–493.
- Hosseinipour, S. L., Khiabani, M. S., Hamishehkar, H., et al. (2015). Enhanced stability and catalytic activity of immobilized α -amylase on modified Fe_3O_4 nanoparticles for potential application in food industries. *Journal of Nanoparticle Research*, 17(9): 382.
- Houen, G., Madsen, M. T., Harlow, K. W., et al. (1996). The primary structure and enzymic properties of porcine prochymosin and chymosin. *International Journal of Biochemistry and Cell Biology*, 28(6): 667–676.
- Ismail, I., Abdullah, N. A. N., Ahmad, Z., et al. (2018). Halal principles and Halal purchase intention among muslim consumers. In *Proceedings of the 3rd International Halal Conference (INHAC 2016)* (pp. 131–138). Springer, Singapore.
- Joshi, S., & Satyanarayana, T. (2013). Biotechnology of cold-active proteases. *Biology*, 2(2): 755–783.
- Juturu, V., & Wu, J. C. (2014). Microbial cellulases: Engineering, production and applications. *Renewable and Sustainable Energy Reviews*, 33: 188–203.
- Karan, R., Capes, M. D., DasSarma, P., et al. (2013). Cloning, overexpression, purification, and characterization of a polyextremophilic β -galactosidase from the Antarctic haloarchaeon *Halorubrum lacusprofundi*. *BMC Biotechnology*, 13(1): 3.
- Kashim, M. I. A. M., Majid, L. A., Adnan, A. H. M., et al. (2015). Principles regarding the use of haram (forbidden) sources in food processing: a critical Islamic analysis. *Asian Social Science*, 11(22): 17.
- Kim, S. M., Park, H., & Choi, J. I. (2017). Cloning and characterization of cold-adapted α -amylase from Antarctic *Arthrobacter agilis*. *Applied Biochemistry and Biotechnology*, 181(3): 1048–1059.
- Ko, J. K., Um, Y., Woo, H. M., et al. (2016). Ethanol production from lignocellulosic hydrolysates using engineered *Saccharomyces cerevisiae* harboring xylose isomerase-based pathway. *Bioresource Technology*, 209: 290–296.
- Kumar, A., Grover, S., Sharma, J., et al. (2010). Chymosin and other milk coagulants: Sources and biotechnological interventions. *Critical Reviews in Biotechnology*, 30(4): 243–258.
- Kumar, A., Sharma, J., Mohanty, A. K., et al. (2006). Purification and characterization of milk clotting enzyme from goat (*Capra hircus*). *Comparative Biochemistry and Physiology Part B: Biochemistry and Molecular Biology*, 145(1): 108–113.
- Lee, D. H., Choi, S. L., Rha, E., et al. (2015). A novel psychrophilic alkaline phosphatase from the metagenome of tidal flat sediments. *BMC Biotechnology*, 15(1): 1.
- Li, J., Hu, Q., Li, Y., & Xu, Y. (2015). Purification and characterization of cold-adapted beta-agarase from an Antarctic psychrophilic strain. *Brazilian Journal of Microbiology*, 46(3): 683–690.
- Li, J.L., Yu, Q.L., Zhang, L., et al. (2012). Purification and characteristics of trypsin from the pancreas of tibetan sheep. *Journal of Food Biochemistry*, 36(1): 122–128.
- Lin, T., Bai, X., Hu, Y., et al. (2017). Synthetic *Saccharomyces cerevisiae*-*Shewanella oneidensis* consortium enables glucose-fed high-performance microbial fuel cell. *AIChE Journal*, 63(6): 1830–1838.
- Liu, Z. Q., Zheng, W., Huang, J. F., et al. (2015). Improvement and characterization of a hyperthermophilic glucose isomerase from *Thermoanaerobacter ethanolicus* and its application in production of high fructose corn syrup. *Journal of Industrial Microbiology & Biotechnology*, 42(8): 1091–1103.
- Ma, G., & Su, Z. G. (2013). *Microspheres and microcapsules in biotechnology: Design, preparation and applications*. Pan Stanford.
- Mageswari, A., Subramanian, P., Chandrasekaran, S., et al. (2017). Systematic functional analysis and application of a cold-active serine protease from a novel *Chryseobacterium* sp. *Food Chemistry*, 217: 18–27.
- Malak, C. A. A., El Adab, I. F. A., Vukashinovic, V., et al. (1996).

- Buffalo (*Bos buffali* L.) chymosin purification and properties. *Comparative Biochemistry and Physiology Part B: Biochemistry and Molecular Biology*, 113(1): 57–62.
- Matsui, M., Kawamata, A., Kosugi, M., et al. (2017). Diversity of proteolytic microbes isolated from Antarctic freshwater lakes and characteristics of their cold-active proteases. *Polar Science*, 13: 82–90.
- Merín, M. G., & Morata de Ambrosini, V. I. (2015). Highly cold-active pectinases under wine-like conditions from non-Saccharomyces yeasts for enzymatic production during winemaking. *Letters in Applied Microbiology*, 60(5): 467–474.
- Miao, L. L., Hou, Y. J., Fan, H. X., et al. (2016). Molecular structural basis for the cold-adaptedness of psychrophilic β -glucosidase BglU in *Micrococcus antarcticus*. *Applied and Environmental Microbiology*, 82(7): 2021–2030.
- Mikucki, J. A., Pearson, A., Johnston, D. T., et al. (2009). A contemporary microbially maintained subglacial ferrous" ocean". *Science*, 324(5925): 397–400.
- Mitsuda, H., & Yasumatsu, K. (1955). Crystallization of animal catalase and studies on its optimum temperature. *Journal of the Agricultural Chemical Society of Japan*, 19(3): 200–207.
- Mu, W., Li, W., Wang, X., et al. (2014). Current studies on sucrose isomerase and biological isomaltulose production using sucrose isomerase. *Applied Microbiology and Biotechnology*, 98(15): 6569–6582.
- Nadeem, S. M. S., Khan, J. A., Murtaza, B. N., et al. (2015). Purification and properties of liver catalase from water buffalo (*Bubalus bubalis*). *South Asian Journal of Life Sciences*, 3(2): 51–55.
- Nealon, C. M., Musa, M. M., Patel, J. M., et al. (2015). Controlling substrate specificity and stereospecificity of alcohol dehydrogenases. *Acs Catalysis*, 5(4): 2100–2114.
- Nilsen, I. W., Øverbø, K., & Olsen, R. L. (2001). Thermolabile alkaline phosphatase from Northern shrimp (*Pandalus borealis*): Protein and cDNA sequence analyses. *Comparative Biochemistry and Physiology Part B: Biochemistry and Molecular Biology*, 129(4): 853–861.
- Pandey, A., Selvakumar, P., Soccol, C. R., et al. (1999). Solid state fermentation for the production of industrial enzymes. *Current science*, 149–162.
- Poyck, P. P., Hoekstra, R., Vermeulen, J. L., et al. (2008). Expression of glutamine synthetase and carbamoylphosphate synthetase I in a bioartificial liver: Markers for the development of zonation in vitro. *Cells Tissues Organs*, 188(3): 259–269.
- Quaglia, D., Pori, M., Galletti, P., et al. (2013). His-tagged horse liver alcohol dehydrogenase: Immobilization and application in the bio-based enantioselective synthesis of (S)-arylpropanols. *Process Biochemistry*, 48(5–6): 810–818.
- Ramos, A. M., Glenn, K. L., Serenius, T. V., et al. (2008). Genetic markers for the production of US country hams. *Journal of Animal Breeding and Genetics*, 125(4): 248–257.
- Ranjan, B., Singh, B., & Satyanarayana, T. (2015). Characteristics of Recombinant Phytase (rSt-Phy) of the thermophilic mold *Sporotrichum thermophile* and its applicability in dephytinizing foods. *Applied Biochemistry and Biotechnology*, 177(8): 1753–1766.
- Ranjan, K., Lone, M. A., & Sahay, S. (2016). Detergent compatible cold-active alkaline amylases from *Clavispora lusitanae* CB13. *The Journal of Microbiology, Biotechnology and Food Sciences*, 5(4): 306.
- Ren, H., Jiang, C., & Chae, J. (2017). Effect of temperature on a miniaturized microbial fuel cell (MFC). *Micro and Nano Systems Letters*, 5(1): 13.
- Riaz, M. N., & Chaudry, M. M. (2003). *Halal food production*. CRC press.
- Roohi, R., Kuddus, M., & Saima, S. (2013). Cold-active detergent-stable extracellular α -amylase from *Bacillus cereus* GA6&58; Biochemical characteristics and its perspectives in laundry detergent formulation. *Journal of Biochemical Technology*, 4(4): 636–644.
- Schauer, R. & Wember, M., 1996. Isolation and characterization of sialate lyase from pig kidney. *Biological Chemistry*, 377(5): 293-300.
- Seppänen, M. M., Cardi, T., Hyökki, M. B., et al. (2000). Characterization and expression of cold-induced glutathione S-transferase in freezing tolerant *Solanum commersonii*, sensitive *S. tuberosum* and their interspecific somatic hybrids. *Plant Science*, 153(2): 125–133.
- Shahani, K. M., Khan, I. M. & Chandan, R. C., (1976). Bovine Pancreatic Lipase I. I. isolation, homogeneity, and characterization. *Journal of Dairy Science*, 59(3): 369–375.
- Shi, Y., Wang, Q., Hou, Y., et al. (2014). Molecular cloning, expression and enzymatic characterization of glutathione S-transferase from Antarctic sea-ice bacteria *Pseudoalteromonas* sp. ANT506. *Microbiological Research*, 169(2–3): 179–184.
- Siddiqui, K. S. (2015). Some like it hot, some like it cold: Temperature dependent biotechnological applications and improvements in extremophilic enzymes. *Biotechnology Advances*, 33(8), 1912–1922.
- Siddiqui, K. S., & Cavicchioli, R. (2006). Cold-adapted enzymes. *Annual Review of Biochemistry*, 75: 403–433.
- Tapre, A. R., & Jain, R. K. (2014). Pectinases: Enzymes for fruit processing industry. *International Food Research Journal*, 21(2).
- Tavakoli, A., & Hamzah, A. (2017). Characterization and evaluation of catechol oxygenases by twelve bacteria, isolated from oil contaminated soils in Malaysia. *Biological Journal of Microorganism*, 5(20).
- Thadathil, N., & Velappan, S. P. (2014). Recent developments in chitosanase research and its biotechnological applications: A review. *Food Chemistry*, 150: 392–399.
- Tsiklinsky, M. (1908). *Flore microbienne: Expédition antarctique française (1903-1905)*. Par Mlle. Tsiklinsky. Masson & Cie.
- Urbietta, M. S., Donati, E. R., Chan, K. G., et al. (2015).

- Thermophiles in the genomic era: Biodiversity, science, and applications. *Biotechnology Advances*, 33(6): 633–647.
- Vanany, I., Maarif, G. A., & Soon, J. M. (2018). Application of multi-based quality function deployment (QFD) model to improve halal meat industry. *Journal of Islamic Marketing*.
- Wahab, A. R. (2004). *Guidelines for the preparation of halal food and goods for the Muslim consumers* [PDF file]. Retrieved February, 28, 2012, from <http://www.halalrc.org/images/Research%20Material/Literature/halal%20Guidelines.pdf>.
- Wang, B., Bai, Y., Fan, T., *et al.* (2017). Characterisation of a thiamine diphosphate-dependent alpha-keto acid decarboxylase from *Proteus mirabilis* JN458. *Food Chemistry*, 232: 19–24.
- Wang, J., Liu, H., Wang, H., *et al.* (2016). Isolation and characterization of a protease from the *Actinidia arguta* fruit for improving meat tenderness. *Food Science and Biotechnology*, 25(4): 1059–1064.
- Wang, Q., Hou, Y., Shi, Y., *et al.* (2014). Cloning, expression, purification, and characterization of glutaredoxin from Antarctic sea-ice bacterium *Pseudoalteromonas* sp. AN178. *BioMed Research International*, 2014.
- Wang, Y. B., Gao, C., Zheng, Z., *et al.* (2015). Immobilization of cold-active cellulase from antarctic bacterium and its use for kelp cellulose ethanol fermentation. *BioResources*, 10(1): 1757–1772.
- Wang, Y., Han, H., Cui, B., *et al.* (2017). A glutathione peroxidase from Antarctic psychrotrophic bacterium *Pseudoalteromonas* sp. ANT506: Cloning and heterologous expression of the gene and characterization of recombinant enzyme. *Bioengineered*, 8(6): 742–749.
- Wei, J., Timler, J. G., Knutson, C. M., *et al.* (2013). Branched-chain 2-keto acid decarboxylases derived from *Psychrobacter*. *FEMS Microbiology Letters*, 346(2): 105–112.
- Willis, T. W. & Tu, A. T., (1988). Purification and biochemical characterization of atroxase, a nonhemorrhagic fibrinolytic protease from western diamondback rattlesnake venom. *Biochemistry*, 27(13): 4769–4777.
- Wilcox, M. D., Brownlee, I. A., Richardson, J. C., *et al.* (2014). The modulation of pancreatic lipase activity by alginates. *Food Chemistry*, 146: 479–484.
- Wirnt, R., (1965). Chymotrypsin. In *Methods of enzymatic analysis* (pp. 800–806).
- Yu, P., Wang, X. T., & Liu, J. W. (2015). Purification and characterization of a novel cold-adapted phytase from *Rhodotorula mucilaginosa* strain JMUY14 isolated from Antarctic. *Journal of Basic Microbiology*, 55(8): 1029–1039.
- Zheng, Y., Zhang, K., Su, G., *et al.* (2015). The evolutionary response of alcohol dehydrogenase and aldehyde dehydrogenases of *Acetobacter pasteurianus* CGMCC 3089 to ethanol adaptation. *Food Science and Biotechnology*, 24(1): 133–140.