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Psychrophilic Microbial Enzymes Implications in Coming Biotechnological Processes

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Abstract

Psychrophilic microorganisms produce a variety of cold-active enzymes which are used as production accelerators at commercial level to cope increasing demands necessitating low temperature conditions. The psychrophilic enzymes are frequently employed in food processing, textile, detergents, feed stocks, bioremediation, cosmetics, paper and pharmaceutical industries. But being extermophilic in nature, psychrophiles have certain pH, ionic strength and temperature limitation. To overcome such issues, their molecular biology and beneficial genetic engineering approaches are current goals of researchers. In this regard, many successful studies have accomplished importance of cold-active enzymes at industrial level. This review summarizes applications of potential psychrozymes.

Keywords: Production accelerators; bioremediation; ionic strength; genetic engineering; cold-active enzyme.

1. Introduction

Microbial flora exist in different habitable locations all around the globe and according to their optimum temperature requirements; microbes are divided into two categories i.e. mesophiles and extermophiles. Among them, mesophiles have been frequently used in various biotechnological processes e.g., from baking to industrial feed stocks' preparation since ascent era. Moreover, potential estimation and utilization of extermophiles are in deliberation to get biotechnological benefits in recent decades. Extermophiles are further diversified into; thermophiles and psychrophiles.

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As in major working areas temperature remains high so thermophiles are easily adjustable there to serve in already running set ups parallel to other products production. With gradual scientific progress, now psychrophiles are center of attention to get biotechnological gains. Their enzymes are **significantly indulged in various processes to serve mankind.**

1.1 Psychrophiles

Psychrophiles are those extermophiles which are inhabitants of cold places. Their survival temperature range tolerance is below zero to even above 20°C [1-3]. They are commonly found in ice dust [4, 5], glaciers, in all Antarctic zones and in artificial cold systems like refrigerators and freezers either at domestic or industrial level [6-11]. However, based on the growth temperature, psychrophilic organisms are again sub-divided in to two categories such as psychrophiles (optimal growth temperature below 15°C) and psychrotrophs or psychrotolerants (breed at optimal temperature of around 20-25°C) [12]. Moreover, cold environment represents an enormous pool of potential microbiota, ranging from Gram negative bacteria (e.g. *Pseudoalteromonas, Moraxella, Psychrobacter, Polaromonas, Psychroflexus, Polaribacter, Moritella, Vibrio* and *Pseudomonas*), to Gram-positive bacteria (e.g. *Arthrobacter, Bacillus* and *Micrococcus*), Archaea (e.g. *Methanogenium,* Methanococcoides and *Halorubrum*), Yeasts (*Candida* and *Cryptococcus*) and Fungi (*Penicillium* and *Cladosporium*). Communally all these organisms have revolutionized the cold biotechnology [13- 17].

1.2 Industrial implication of psychrophiles

There is an increasing demand for the utilization of microbial biocatalysts in industrial application because of their withstanding nature at diverse robust processing conditions. Most of the enzymes which are in use in today's industrial methods are sourced from mesophiles. No doubt mesophiles are of commercial importance but their enzymatic action is limited at peak of different factors like temperature, pH and ionic bonds strength [1]. Moreover, the constrain for cost-cutting lies in the heating or cooling steps of industrial processes and increase in the upturn of the products of enzymatic reaction strength [18] can give a much attention to the use of proteins isolated from cold loving microorganisms. Several psychrozymes and their biotechnological applications are shown in Table 1**.**

The documented data of psychrophiles revealed the capability to degrade an extensive variety of polymeric substances and the producer of various enzymes like amylases, cellulases, pectinases, α- Galactosidase, oxidases, protease and lipase etc. Due to the potential properties, cold-active enzymes have led to accelerate the concern in investment of considerable finance and global effort in the research and expansion for exploring more number of industrially important cold active enzymes in the fields of food commerce (such as pectinase, α-galactosidase), biopolishing and stone washing of textile items and detergent formulation industries (e.g., lipases, amylases, and cellulases). Moreover these psychrozymes effectively take part in bioremediation (such as oxidases) and for biotransformations (methylases and aminotransferases) [52] as well as in biomedical applications [8]. This review covers the microbial cold adapted enzymes and their biotechnologically significant industrial implications.

Table 1: Major psychrophilic enzymes and their biotechnological potential

Cold-active enzyme	Biotechnological implementation
Metalloprotease	Food, detergents, molecular biology [19]
Serine peptidase	Food, detergents, molecular biology [20]
Lipase	Food, detergents, cosmetics [21]
Alkaline phosphatase	Molecular biology [22]
Alcohol dehydrogenase	Asymmetric Chemical synthesis [23]
3-Isopropylmalate dehydrogenase	Asymmetric Chemical synthesis [24]
Lactate dehydrogenase	Biotransformation, biosensor, lactose removal from milk [25]
Valine dehydrogenase	Biotransformation [26]
β-Galactosidase	Dairy industries (e.g. enhancing standards of ice-cream and whey) [27]
RNA polymerase	Molecular biology [28]
DNA polymerase	Molecular biology [29]
DNA ligase	Molecular biology [30]
Uracil-DNA glycosylase	Molecular biology [31]
Restriction Endonuclease *Unb*I	Molecular biology [32]
Triose phosphate isomerase	Biotransformation [33]
Chitobiase	Food, health products [34]
Chitinase A	Food, health products [35]
Cellulase	Animal feed, textiles, detergents [36]
Polygalacturonase (pectinase)	Preparation of cheese, wine and fruit nector [37]
Pectate lyase	Cheese ripening, fruit juice and wine industry [38]
Nitrile hydatase	Low-temperature acrylamide synthesis [39]
Pullulanase	Pullulan hydrolysis [40]
Xylanase	Dough rising, protoplast formation, wine and beverages production [41]
Alanine racemase	Food storage, Antibacterial agent [42]
α-Amylase	Detergents, Dough fermentation, desizing denim jeans, pulp bleach [43]
Glucoamylase	Starch hydrolysis [44]
β-Lactamase	Antibiotic degradation [45]
Phosphoglycerate kinase Catalase	Biotransformation Dairy, water treatment in paper, food, textile, semiconductor industries [46, 47]
Aspartate carbamoyl- transferase	Biotransformation [48]
Chlamysin (lysozyme-like)	Antibacterial agent, food preservation [49]
Isocitrate lyase	Biotransformation [50]
Malate synthase	Biotransformation [51]

2. Food technology

The potent involvement of cold-adapted enzymes in the food trade is significant. For example, in the milk industry, β-galactosidase is introduced at low temperature for lactose reduction [53] accountable for severe intolerances to the milk sugar in world's around two thirds of the population. Moreover, this psychrophilic β-galactosidase can also be used for conversion of cheese byproduct, to quickly fermentable glucose and galactose [54]; whereas commercial addition of pectinases improves the fruit juice extraction, reduction of the viscosity and helps to elucidate the end product; another group of enzymes, proteases facilitate in meat tenderization; moreover, for bakery items enzymes like amylases, proteases and xylanases can be used to optimize the dough fermentation period along with the dough and the crumb quality enhancement, in addition to the retention of aromas and moisture grades. These psychrozymes act directly on starch, gluten and hemicellulases in the flour. Their other important feature is easy activation and this prevents the extended catalysis from altering the crumb which otherwise may become too sticky. That is why, available rheological data indicates efficiency of psychrozymes in the baking industry [55-60]. In addition to that a considerable range of cold active enzymes are regarded as ideal to mesophilic ones in brewing and wine industries, cheese manufacturing and animal feed, and so on [61].

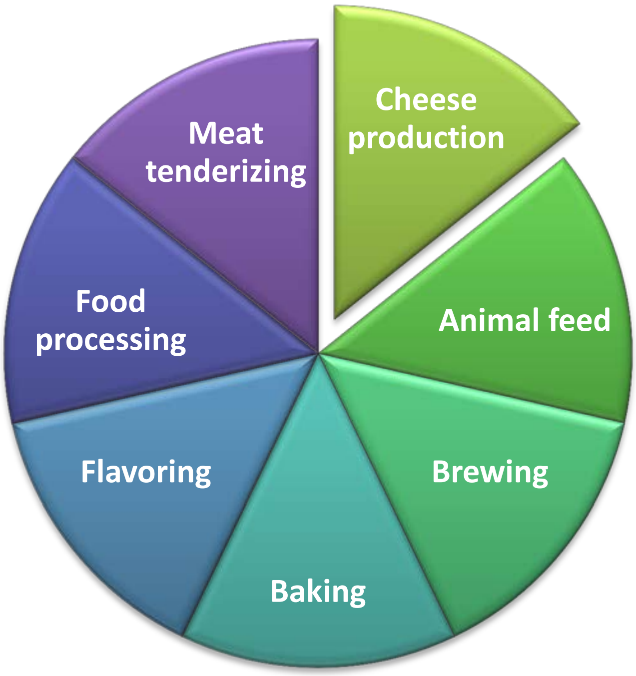

Figure 1: Basic applications of Psychrophilic enzymes in Food Biotechnology

3. Textile

Economically, a cold-adapted cellulase is of great importance in textile industry for biopolishing of fabric and stone-washing of jeans. In fabric production, the pretreatment with it inhibits protrudance of cotton fibers, pill formation and enhances texture durability and appearance of cloth even after several washes [61].

4. Detergents

At industrial level, 30%–40% of psychrozymes are consumed around the globe. The leading one is subtilisin (an alkaline serine protease usually obtained from *Bacillus* species). Whereas at the domestic level, the recent practice is to utilize psychrozymes based detergents are employed for mechanical and financial input reduction as well as to shield texture and brightness of the garments. As an outcome, cold-active subtilisins are well known for best washing at moderate tap water temperatures. In this regard, the first psychrozyme subtilisins isolated from Antarctic *Bacillus* species [62, 63]. However, they lack temperature stability for their shelf-life. Therefore, in current practice, engineered subtilisins are added in cold-active detergents that exhibit storage and alkaline stability along with cold-activity [11]. Moreover, the addition of other psychrophilic enzymes in surfactants like proteases, lipases, α-amylases and cellulases, as additives causes a decrease in energy input and wear and tear reduction. Meanwhile possible drawback is their instability as component of produce and their poor shelf life yet. Thus, still a channelized effort is required to improve the stability psychrozymes along with maintained good catalytic competence at colder temperatures [63]. In this account, one more improvement has mentioned in a recent report that another enzyme group of estrases from Psychrophilic *Rhodococcus sp.* can be employed in detergents for effective stain removal [64].

5. Paper pulp

Various psychrophilic enzymes are currently used in paper industry. The major commercial contributors are xylanases which are capable to stand with the high temperature (55–70°C) and alkaline pH of the pulp proper biobleaching [65, 66]. Moreover, thermo-alkaliphilic or even thermoacidophilic xylanases may also serve as bioconversion agents in hot water and steam explosion, alkaline, solvent or acidic treatments [67, 68]. Moreover, cold active enzymes are involved in the catalysis of wood and other lignocellulosic matter to carbon dioxide, water, and humic substances. Such enzymatic action on cellulose, hemicelluloses, and lignin is required basically in paper and pulp industry. Psychrophilic cellulases are employed in biotechnological processes at commercial level to perform microbial delignification, and to serve as biosensors for pulp fiber surfaces analysis [69].

6. Pharmacy

In recent era, dehalogenases in pharmaceutical industries are indulged for the formation of optically pure drug intermediates including hydroxyalkanoic and haloalkonoic acids [70].In this regard, over the last 20 years, the marine environment has been recognized as a potential reservoir of psychrozymes and metabolites [71]. Oceans contain a vast range of halogens; especially marine algae are the best organohalogen producers [72, 73] and the halogenated terpenes which are obtained from them, exhibit antimicrobial and anticancer properties [74, 75]. Such compounds are composed of vanadium bromoperoxidase and their leading sources are species e.g., *Corallina*, *Laurencia* and *Plocamium* sp. [70]. In another recent study, it has been explored that cold active enzymes producer isolates of caves show good antimicrobial activity and their enzymes are of great pharmaceutical importance. They also exhibit hydrolytic and hemolytic activities at low temperatures [76]. Similarly, among the Antarctic psychrophiles, *Shewanella* and *Colwellia* are ideal producers of PUFA, such as eicosapentaenoic acid or docosahexaenoic acid. The traditional source for long-chain polyunsaturated fatty acids (LPUFA) is effective to fight against numerous disorders like atherosclerosis, diabetes and hypertension. But their purification from fish oil comes with some problems at the risk of large scale production. Moreover, it is expected that a number of economically important fishes are likely to decline in the future. Whereas, oils of algal origin demand comparatively higher level of technology and budget than of psychrophilic ones who also have low lipid profile. Bacterial PUFA are component of some membranous phospholipids, that is why, they are considered as ideal producers of pure PUFA [8].

7. Environmental application

For ecological balance recovery, microbial bioremediation is major contributor since log time [77]. But in temperate zones, steep weather and thermal variations lessen the microbial decomposition of organic pollutants. As in 2014, Bowman and Deming [78] reported that psychrophilic microbes have alkane hydroxylase genes which enable them to degrade alkanes and crude oils in to harmless compounds. However, bioaugmentation and inoculation of pollutants with specific psychrophiles in mixed cultures are expected to enhance the biodegradation of recalcitrant substrates with great catalysis [9, 79]. However, the optimization of such psychrophiles is under process yet. For this purpose, European Union has selected contaminated waste water to estimate the required potential of psychrophiles for a vast range of toxicants along with various biopolymers degradation [77, 80, and 81]. Specifically, the quality of psychrophilic yeasts to catalyze a broad range of phenols and other hydrocarbons is demanding biotechnological perspective [82, 83]. Yeasts isolates from contaminated glacial origins decompose a considerable amount of various hydrocarbons at 10 °C [84]. Likewise, in 2005, Bergauer and coworkers [85] screened a set of psychrophilic yeasts inhabitant of Alpine zones degrade phenol and related monoaromatic compounds at 10 °C. Moreover, they can also assimilate few non- or low volatile aromatic substances which is a strain-related feature. Recently, it has noticed that cold active basidiomycetous yeasts fully decomposed up to 12.5–15 mM phenol at 10 °C under fed-batch cultivation which immobilization may improve yield [83, 86].

8. Biocatalyst

Catabolic activity of psychrophilic enzymes is under consideration now days and need optimization to get proper benefit [11]. The significant properties responsible for the three chief privileges of psychrozymes in biotechnology are as under:

- They are cost effective as less amount of enzyme is required to meet activation energy requirement.
- Psychrozymes are proficient without additional thermal aid.
- Due to thermal lability, their selective inactivation can be achieved with less heat input.
- The endemic psychrophiles can be a valuable new source of frequently required catalysts e.g., lipases from the yeast *Candida antarctica* or by the xylanase from the bacterium *Pseudoalteromonas haloplanktis* [13, 61, 87-92].

Moreover, a wide range of the commercially valuable fatty acid esters, peptides, oligosaccharide derivatives and other compounds are gained from substrates having poor aqueous solubility which may be enhanced by introducing less water demanding enzymes [93]. Further investigations have revealed that the hydration level is best managed by limiting the water activity. Here, a conflict arises because in case of less hydration, enzymatic effectiveness usually reduces and it has direct relation with reaction kinetics. That is why, more supporting data is required yet [94]. Psychrozymes might therefore have a possible benefit for implications in less aqueous conditions due to their greater flexibility and tolerance than of mesophilic and thermophilic enzymes. This will permit the use of reduced water content with the consequence of rising yield [61].

9. Industrial usage

The polar yeast *Candida antarctica*, is of well renowned economic importance. It is producer of two types of lipases, A and B. among them, lipase B is involved in wide range of applications related to pharmaceuticals, cosmetics food and animal feed processing [11, 95, 96]. Moreover, in 2013, Novak and coworkers [70] reported that cold active dehalogenases are of industrial importance to achieve protein stability and flexibility in produce. Recently, it has been reported that cold active proteases [97, 98] lipases [99-101] cellulases [102-103] amylases [104, 105] pectinases [106] have found their role into many commercial wings applications like in detergents, food, textiles, cosmetics, beverages and bakery [107].

10. Cosmetics

Psychrophilic bacteria produce various types of enzymes but among them carboxylesterases, lipases and esterases are frequently utilized in cosmetics production. They are capable of hydrolysis of simple partially soluble ester-containing molecules. Similarly, lipases are considered ideal for hydrophobic long-chain triglycerides which are attractive constituents in various surfactants and cosmetic items [108, 109]. Moreover, it has also been observed that cold-active lipases upgrades application results of various beauty products [71]. In current era, cosmetic industry of China is using these psychrophilic enzymes to improve quality of their products [110].

11. Molecular biology

In this regard, cold active alkaline phosphatases have major scope in molecular biology. They are employed for the prevention of recircularization by pre-cloning DNA vector. However, the phosphatase should be vigilantly removed after dephosphorylation to evade disturbances in the later steps. Furthermore, *E. coli* and calf intestinal alkaline phosphatase have found heat-stable and they just require surfactant addition for inactivation. It follows that heat-labile alkaline phosphatases are exceptional alternatives as they are inactivated by moderate heat exposure in same glass ware with minimum amount of nucleic acid [111]. Fifteen years later, a group of Antarctic *V. bouriotis* isolates were explored for alkaline phosphatase and used as gene donor in *E. coli* cloning [112]. This effort solved its crystal configuration [113] and opened a way for further improvement to elevate catalysis and heat-lability [114]. Moreover, now days, the heat-labile alkaline phosphatase is also obtained from the Arctic shrimp *Pandalus borealis* for commercial purpose. In addition, psychrophilic lipase from deep sea inhabitant bacterium, *Psychrobactor sp.* has been used as target for gene cloning to make its utilization fesible in mesophilic conditions effectively [115]. There are two more heat labile psychrophilic enzymes which are available for molecular biology applications. They are shrimp nuclease and heat-labile uracil-DNA *N*-glycosylase both capable of contaminants removal and DNA helix degradation during PCR. The former is used as recombinant form in *Pichia pastoris* which the later one is obtained from Atlantic cod (*Gadus morhua*) and used as recombinant form in *E. coli*. Both psychrozymes can be easily inactivated by moderate thermal treatment [11, 116].

12. Fuel energy

For low cost fuel and energy production, psychrophilic enzymes implication is of great importance. Recently, it has been reported that hydrogen gas can be produced during whey biofermentation process by cold active enzymes [117, 118]. In this regard, majority studies hydrogen gas production during fermentation by employing pure sugars and biomass were addressed [119]. A high carbohydrate content of food waste makes it a probable feedstock for bio-production of hydrogen by psychrophilic microbes [120, 121]. It has been observed that a wide range of psychrophiles inhabit required activity at moderate to low temperatures and from commercial and scientific view, there is a direct relation between enzymatic globular protein structure and thermal stability. So they fulfill this criterion positively for biohydrogen production [16, 122, and 123]. Moreover, the effective biohydrogen production from whey powder by using different psychrophilic anaerobic bacteria like *Rahnella aquatilis, Carnobacterium maltaromaticum, Trichococcus collinsii and Clostridium algidixylanolyticum* has been achieved so far [118]. In addition to that, agriculture and farm wastes are abundant, cost effective and valuable renewable natural resources as biofuels and other commercial chemicals production [124]. Such lingocellulosic waste provides carbonic substrates for anaerobic catalysis. However, this may be affected by thermal variations of surroundings. In this regard, cold temperature is a common stress factor and directly alters enzymatic action. But with scientific progress, a number of reports indicated that psychrozymes have proved themselves for fermentation even in cold milieu [125-127]. So, for proper use of biotechnological potential of cold active agents should be analyzed and obtained for better service in fuel industry [128]. Psychrophilic fermentors considerably requires less energy input required for heating the bioreactor and even work in frozen zones for conversion of agricultural wastes in biogas [125].

Because cellulose and xylan are the most abundant polymers in plants, their microbial transformation is under active research. Similarly, undigested lignocellulose and bedding materials of dairy manure like cellulose containing straws are required to be decomposed to produce simpler hexoses and pentoses, while, lignin is the limiting step in the degradation of lignocellulosic substrates due to extremely recalcitrant in nature [129, 130]. The heterogeneity and composite constitution are a major task for the anaerobic break down of lignocellulosic biomass from plant material [131]. According to reported data, now methane production from cellulose, xylan, cellulose and xylan mixture, cow feces, and wheat straw is possible at psychrophilic state (20°C) by utilizing optimized inoculums [132].

13. Genetic engineering

To enhance microbial biotechnology potential for better industrial output, their genetic engineering is under consideration yet. However, engineered psychrophilic activity having mesophilic enzymes have not been reported to date. The main hurdle is reason complexity of amino acid substitutions and bonding patterns leading to low temperature catabolism, need long period of optimization. As an outcome, engineered psychrozymes currently escape the computational capability. In this regard, laboratory microbial evolutionary analysis has been found successful for production of psychrozymes with or without heat-lability [133]. It is important to mention here, in many cases; due to psychrophilic enzymes structural complications, the after effects of selective random mutations cannot be analyzed yet. For this purpose, the likelihood to introduce a specific property based directed evolutionary study seems to be the best methodology to engineer enzyme cold-activity. Improved cold-activity by chemical alteration has also been reported but still the outcomes of chemical modification remain capricious [134, 135]. So, more research work and estimation is required to achieve better biotechnological gains.

14. Conclusion and future perspective

It can be concluded that psychrophiles have much more to contribute to the field of biotechnology [61]. That is why; the recent increasing interests on psychrophilic microorganisms not only focus on the genomic and proteomic study to establish the relationships but also on production of industrial important substitutions from psychrophiles. Although the psychrophilic enzymes have high specific activity but small half life makes a major drawback in the utilization of these enzymes at commercial aspect. The leading target is to such genetically modified and engineered psychrophilic strains which will be able to cope commercial expectations. Thus it is necessary to identify further key feature of these psychrophilic enzymes to evaluate and upgrade their biotechnological potential [8].

15. Recommendations

It is recommended that further research work is required to develop genetically modified psychrophiles with enhanced enzyme production at achievable temperature range for economical commercial manufacturing.

References

[1]. S. D'Amico, C. Gerday and G. Feller. Structural determinants of cold adaptation and stability in a large protein. J Biol Chem, vol. 276, pp. 25791-25796, 2001.

[2]. A. L. Mascarelli. Geomicrobiology: Low life. Nature, vol. 459, pp. 770–773, 2009.

[3]. A.L. Giudice, V. Bruni, M. De Domenico and L. Michaud. "Psychrophiles - Cold-Adapted Hydrocarbon-Degrading Microorganisms", in Handbook of Hydrocarbon and Lipid Microbiology, T. McGenity, Ed., 2010, pp. 1897-1921.

[4]. A. Hodson, A.M. Anesio, M. Tranter, A. Fountain, M. Osborn, J. Priscu, J. Laybourn-Parry and B. Sattler. Glacial ecosystems. Ecol. Monogr., vol. 78, pp. 41-67, 2008.

[5]. S. MacDonell and S. Fitzsimons. The formation and hydrological significance of cryoconite holes. Prog. Phys. Geogr., vol. 32, pp. 595-610, 2008.

[6]. J.A. Baross and R.Y. Morita. "Microbial life at low temperatures: ecological aspects", in Microbial

Life in Extreme Environments. D. J. Kushner, Ed., Academic Press, 1978, pp. 9 -71.

[7]. E. Willerslev, A.J. Hansen and HN. Poinar. Isolation of nucleic acids and cultures from fossil ice and permafrost. Trends Ecol Evol, vol. 19, pp. 141-147, 2004.

[8]. K.K. Pulicherla, M. Ghosh, P.S. Kumar and K.R.S.S. Rao. Psychrozymes- The Next Generation Industrial Enzymes. J Marine Sci Res Development, vol. 1, pp. 102, 2011.

[9]. P. Buzzini, E. Branda, M. Goretti and B. Turchetti. Psychrophilic yeasts from worldwide glacial habitats: diversity, adaptation strategies and biotechnological potential. FEMS Microbiol Ecol, vol. 82, pp. 217–241, 2012.

[10]. L.Loperena, V. Soria, H. Varela, S. Lupo, A. Bergalli,M. Guigou, A. Pellegrino, A. Bernardo, A. Calviño, F. Rivas and S. Batista. Extracellular enzymes produced by microorganisms isolated from maritime Antarctica. World Journal of Microbiology and Biotechnology, vol. 28 (5), pp. 2249-2256, March 2012.

[11]. C. Struvay and G. Feller. Optimization to Low Temperature Activity in Psychrophilic Enzymes. Int. J. Mol. Sci., vol. 13, pp. 11643-11665, 2012.

[12]. R.Y. Morita. Psychrophilic bacteria. Bacteriol Rev, vol. 39, pp. 144-167, 1975.

[13]. R. Cavicchioli, K.S. Siddiqui, D. Andrewss and K.R. Sowers. Low-temperature extremophiles and their applications. Curr Opin Biotechnol, vol. 13, pp. 253–261, 2002.

[14]. J.W., Deming. Psychrophiles and Polar regions. Curr Opin Microbiol, vol. 5, pp. 301-309, 2002.

[15]. R. Margesin, G. Feller, C. Gerday and N. Russell. “Cold-Adapted Microorganisms: Adaptation Strategies and Biotechnological Potential”, in The Encyclopedia of Environmental Microbiology, vol. 2, G. Bitton, Ed., John Wiley & Sons New York. , 2002, pp. 871-885.

[16]. G. Feller and C. Gerday. Psychrophilic enzymes: hot topics in cold adaptation. Nat Rev Microbiol, vol.1, pp. 200-208, 2003.

[17]. D. Georlette, V. Blaise, T. Collins, S. D’Amico and E. Gratia. Some like it cold: biocatalysis at low temperatures. FEMS Microbiol Rev, vol. 28, pp. 25-42, 2004.

[18]. S. D’Amico, T. Collins, J. C. Marx, G. Feller and C. Gerday. Psychrophilic microorganisms: challenges for life. EMBO Rep., vol. 7, pp. 385–389, 2006.

[19]. M. Turkiewicz, E. Gromek, H. Kalinowska, M. Zielinska. Biosynthesis and properties of an extracellular metalloprotease from the Antarctic marine bacterium Sphingomonas paucimobilis. J Biotechnol, vol. 70, pp. 53-60, 1999.

[20]. J.A. Irwin, G.A. Alfredsson, A.J. Lanzetti, H.M. Gudmundsson and P.C. Engel. Purification and characterization of a serine peptidase from the marine psychrophile strain PA-43. FEMS Microbiol Lett, vol. 201, pp. 285-290, 2001.

[21]. I. Mayordomo, F. Randez-Gil, and J.A., Prieto. Isolation, purification and characterization of a cold-active lipase from Aspergillus nidulans. J Agric Food Chem, vol. 48, pp. 105-109, 2000.

[22]. J.B. Hauksson, O.S. Andresson and B. Asgeirsson. Heat-labile bacterial alkaline phosphatase from a marine Vibrio sp. Enzyme Microb Technol, vol. 27, pp. 66-73, 2000.

[23]. I. Tsigos, K. Velonia, I. Smonou, and S.V. Bourioti. Purification and characterization of an alcohol dehydrogenase from the Antarctic psychrophile Moraxella sp. TAE 123. Eur J Biochem, vol. 254, pp. 356-362, 1998.

[24]. A. Svingor, J. Kardos, I. Hajdu, A. Nemeth, P. Zavodszky. A better enzyme to cope with cold. Comparative flexibility studies on psychrotrophic, mesophilic and thermophilic IPMDHS. J Biol Chem, vol. 276, pp. 28121-28125, 2001.

[25]. G. Feller, J.P. Pauly, A. Smal, P. O'Carra and C. Gerday. The lactate dehydrogenase of the icefish heart: biochemical adaptations to hypoxia tolerance. Biochim Biophys Acta, vol. 1079, pp. 343-347, 1991.

[26]. T. Oikawa, K. Yamanaka, T. Kazuoka, N. Kanzawa and K. Soda. Psychrophilic valine dehydrogenase of the Antarctic psychrophile, Cytophaga sp. KUC-1. Purification, molecular characterization and expression. Eur J Biochem, vol. 268, pp. 4375-4383, 2001.

[27]. J.M. Coombos and J.E. Brenchley. Biochemical and phylogenetic analyses of a cold-active β-galactosidase from the lactic acid bacterium Carnobacterium piscicola BA. Appl Environ Microbiol, vol. 65, pp. 5443-5450, 1999.

[28]. S. Uma, R.S. Jadhav, G. Seshu-Kumar, S. Shivaji, and M.K. Ray. An RNA polymerase with transcriptional activity at 0°C from the Antarctic bacterium Pseudomonas syringae. FEBS Lett, vol. 453, pp. 313-317, 1999.

[29]. C. Schleper, R.V. Swanson, E.J. Mathur, and E.F. Delong. Characterization of a DNA polymerase from the uncultivated psychrophilic archaeon Cenarchaeum symbiosum. J Bacteriol, vol. 179, pp. 7803-7811, 1997.

[30]. D. Georlette, Z.O. Jonsson, F.V. Petegem, J.P. Chessa, J.V. Beeumen, U. Hubscher and C. Gerday. A DNA ligase from the psychrophile Pseudoalteromonas haloplanktis gives insight into the adaptation of proteins to low temperature. Eur J Biochem, vol. 267, pp. 3502-3512, 2000.

[31]. O. Lanes, P.H. Guddal, D.R. Gjellesvik and N.P. Willassen. Purification and characterization of a cold-adapted uracil-DNA glycosylase from Atlantic cod (Gadus morhua). Comp Biochem Physiol B, vol. 127, pp. 399-410, 2000.

[32]. M. Kawalec, P. Borsuk, S. Piechula and P.P. Stepien. A novel restriction endonuclease UnbI, a neoschizomer of Sau96I from an unidentified psychrofilic bacterium from Antarctica is inhibited by phosphate ions. Acta Biochim Pol, vol. 44, pp. 849-852, 1997.

[33]. M. Alvarez, J.P. Zeelen, V. Mainfroid, F. Rentier-Delrue, J.A. Martial, L. Wyns, R.K. Wierenga and D. Maes. Triose-phosphate isomerase (TIM) of the psychrophilic bacterium Vibrio marinus. J Biol Chem, vol. 273, pp. 2199-2206, 1999.

[34]. T. Lonheinne, J. Zoidakis, C.E. Vorgias, G. Feller, C. Gerday and V. Bouriotis. Modular structure, local flexibility and cold-activity of a novel chitobiase from a psychrophilic Antarctic bacterium. J Mol Biol., vol. 310, pp. 291-297, 2001.

[35]. T. Lonhienne, E. Baise, G. Feller, V. Bouriotis and C. Gerday. Enzyme activity determination on macromolecular substrates by isothermal titration calorimetry: application to mesophilic and psychrophilic chitinases. Biochim Biophys Acta, vol. 1545, pp. 349-356, 2001.

[36]. A.H. Iyo and C.W. Forsberg. A cold-active glucanase from the ruminal bacterium Fibrobacter succinogenes S85. Appl Environ Microbiol, vol. 65, pp. 995-998, 1999.

[37]. T. Takasawa, K. Sagisaka, K. Yagi, K. Uchiyama, A. Aoki, K. Takaoka, K. Yamamato. Polygalacturonase isolated from the culture of the psychrophilic fungus Sclerotinia borealis. Can J

Microbiol, vol. 43, pp. 417-424, 1997.

[38]. L.V. Truong, H. Tuyen, E. Helmke, L.T. Binh and T. Schweder. Cloning of two pectate lyase genes from the marine Antarctic bacterium Pseudoalteromonas haloplanktis strain ANT/505 and characterization of the enzymes. Extremophiles, vol. 5, pp. 35-44, 2001.

[39]. I. Watanabe, Y. Satoh, K. Enomoto, S. Seki and K. Sakashita. Optimal conditions for cultivation of Rhodococcus sp. N-774 and for conversion of acrylonitrile to acrylamide by resting cells. Agric Biol Chem, vol. 51, pp 3201-3206, 1987.

[40]. T. Kimura and K. Horikoshi. Characterization of Pullalan-hydrolysing enzyme from an alkalopsychrotrophic Micrococcus sp. Appl Microbiol Biotechnol, vol. 34, pp. 52-56, 1990.

[41]. I. Petrescu, J.L. Brasseur, J.P. Chessa, P. Ntarima, M. Claeyssens, B. Devreese, G. Marino and C. Gerday. Xylanase from the psychrophilic yeast Cryptococcus adeliae. Extremophiles, vol. 4, pp. 137-144, 2000.

[42]. K. Yokoigawa, Y. Okubo, H. Kawai, N. Esaki and K. Soda. Structure and function of Psychrophilic alanine racemase. J Mol Catal B Enzym, vol. 12, pp. 27-35, 2001.

[43]. G. Feller, F. Payan, F. Theys, M. Qian, R. Haser and C. Gerday. Stability and structural analysis of α-amylase from the Antarctic psychrophile Alteromonas haloplanctis A23. Eur J Biochem, vol. 222, pp. 441-447, 1994.

[44]. R. Demot and H. Verachtert. Purification and characterization of extracellular α-amylase and glucoamylase from the yeast Candida antarctica CBS 6678. Eur J Biochem, vol. 164, pp. 643-645, 1987.

[45]. G. Feller, Z. Zekhnini, J. Lamotte-Brasseur and C. Gerday. Enzymes from cold-adapted microorganisms: the class C β-lactamase from the antarctic psychrophile Psychrobacter immobilis A5. Eur J Biochem, vol. 244, pp. 186-191, 1997.

[46]. M. Bentahir, G. Feller, M. Aittaleb, J. Lamotte-Brasseur, T. Himri, J.P. Chessa and C. Gerday. Structural, kinetic and calorimetric characterization of the cold-active phosphoglycerate kinase from the Antarctic Pseudomonas sp. TAC II 18. J Biol Chem, vol. 275, pp. 11147-11153, 2000.

[47]. I. Yumoto, H. Iwata, T. Sawabe, K. Ueno, N. Ichise, H. Matsuyama, H. Okuyama and K. Kawasaki. Characterization of a facultatively psychrophilic bacterium, Vibrio rumoiensis sp. that exhibits high catalase activity. Appl Environ Microbiol, vol. 65, pp. 67-72, 1999.

[48]. Y. Xu, Y. Zhang, Z. Liang, M.V. de-Casteele, C. Legrain, and N. Glansdorff. Aspartate carbamoyltransferase from a psychrophilic deep-sea bacterium, Vibrio strain 2693: properties of the enzyme, genetic organization and synthesis in Escherichia coli. Microbiology, vol. 144, pp. 1435-1441, 1998.

[49]. I.W. Nilsen, K. Overbo, E. Sandsdalen, E. Sandaker, K. Sletten, B. Myrnes. Protein purification and gene isolation of chlamysin, a cold-active lysozyme-like enzyme with antibacterial activity. FEBS Lett, vol. 464, pp. 153-158, 1999.

[50]. S. Ohgiya, T. Hoshino, H. Okuyama, S. Tanaka and K. Ishizaki. "Biotechnology of enzymes from cold-adapted microorganisms", in Biotechnological Applications of Cold-Adapted Organisms. R. Margesin and F. Schinner, Ed., Heidelberg: Springer-Verlag, 1999, pp. 17-34.

[51]. S. Watanabe, Y. Takada and N. Fukunaga. Purification and characterization of a cold-adapted isocitrate

lyase and a malate synthase from Colwellia maris, a psychrophilic bacterium. Biosci Biotechnol Biochem, vol. 65, pp. 1095-1103, 2001.

[52]. H. Okuyama, S. Ohgiya, T. Hoshino, S. Tanaka and K. Ishizaki. "Cold-adapted microorganisms for use in food biotechnology", in Biotechnological Applications of Cold-Adapted Organisms. R. Margesin and F. Schinner, Eds., Berlin, 1998, pp. 101-117.

[53]. A. Hoyoux, I. Jennes, P. Dubois, S. Genicot, F. Dubail, J.M. Francois, E. Baise, G. Feller and C. Gerday. Cold-adapted beta-galactosidase from the Antarctic psychrophile Pseudoalteromonas haloplanktis. Appl. Environ. Microbiol., vol. 67, pp. 1529–1535, 2001.

[54]. E. S. Nam, Y. H. Kim, K. H. Shon and J. K. Ahn. Isolation and characterization of a psychrophilic bacterium producing cold active lactose hydrolyzing enzyme from soil of Mt. Himalaya in Nepal. African Journal of Microbiology Research, vol. 5(16), pp. 2198-2206, 2011.

[55]. B.G. Sproessler. "Milling and baking", in Enzymes in Food Processing, T. Nagodawithana and G. Reed, Eds., Academic Press, 1993, pp. 293–320.

[56]. T. Collins, M.A. Meuwis, I. Stals, M. Claeyssens, G. Feller and C. Gerday. A novel family 8 xylanase, functional and physicochemical characterization. J. Biol. Chem., vol. 277, pp. 35133–35139, 2002.

[57]. T. Collins, M.A. Meuwis, C. Gerday and G. Feller. Activity, stability and flexibility in glycosidases adapted to extreme thermal environments. J. Mol. Biol., vol. 328, pp. 419–428, 2003.

[58]. V.F. Petegem, T. Collins, Meuwis, M.A. C. Gerday, G. Feller, J V. Beeumen. The structure of a cold-adapted family 8 xylanase at 1.3 Å resolution. Structural adaptations to cold and investigation of the active site. J. Biol. Chem., vol. 278, pp. 7531–7539, 2003.

[59]. T. Collins, D. de Vos, A. Hoyoux, S.N. Savvides, C. Gerday, J. Van Beeumen and G. Feller. Study of the active site residues of a glycoside hydrolase family 8 xylanase. J. Mol. Biol., vol. 354, pp. 425–435, 2005.

[60]. D. De Vos, T. Collins, W. Nerinckx, S.N. Savvides, M. Claeyssens, C. Gerday, G. Feller and J.V. Beeumen. Oligosaccharide binding in family 8 glycosidases: Crystal structures of active-site mutants of the beta-1, 4-xylanase pXyl from Pseudoaltermonas haloplanktis TAH3a in complex with substrate and product. Biochemistry, vol. 45, pp. 4797–4807, 2006.

[61]. C. Gerday, M. Aittaleb, M. Bentahir, J.P. Chessa, P. Claverie, T. Collins, S. D'Amico, J. Dumont, G. Garsoux, D. Georlette, A. Hoyoux, T. Lonhienne, M. A. Meuwis and G. Feller. Cold-adapted enzymes: from fundamentals to biotechnology, TIBTECH MARCH, vol. 18, pp. 103-107, 2000.

[62]. S. Davail, G. Feller, E. Narinx and C. Gerday. Cold adaptation of proteins. Purification, characterization, and sequence of the heat-labile subtilisin from the Antarctic psychrophile Bacillus TA41. J. Biol. Chem., vol. 269, pp. 17448–17453, 1994.

[63]. E. Narinx, E. Baise and C. Gerday. Subtilisin from psychrophilic Antarctic bacteria: characterization and site-directed mutagenesis of residues possibly involved in the adaptation to cold. Protein Eng., vol. 11, pp. 1271–1279, 1997.

[64]. C.D. Santi, P. Tedesco, L. Ambrosino, B. Altermark, N.P. Willassen and D. de Pascale. A New Alkaliphilic Cold-Active Esterase from the Psychrophilic Marine Bacterium Rhodococcus sp.: Functional and Structural Studies and Biotechnological Potential. Appl Biochem Biotechnol, vol. 172, pp. 3054–3068, 2014.

[65]. Q.K. Beg, M. Kapoor, L. Mahajan and G.S. Hoondal. Microbial xylanases and their industrial applications: a review. Appl. Microbiol. Biotechnol., vol. 56, pp. 326–338, 2001.

[66]. L. Viikari. Xylanases in bleaching: from an idea to the industry. FEMS Microbiol. Rev., vol. 13, pp. 335–350, 1994.

[67]. J.R. Mielenz. Ethanol production from biomass: technology and commercialization status. Curr. Opin. Microbiol., vol. 4, pp. 324–329, 2001.

[68]. B.C. Saha. Hemicellulose bioconversion. J. Ind. Microbiol. Biotechnol., vol. 30, pp. 279–291, 2003.

[69]. K.E.L. Eriksson. Biotechnology in the pulp and paper industry. Wood Science and Technology, 24 (1): 79-101, 1990.

[70]. H. R. Novak, C. Sayer, J. Panning and J.A. Littlechild. Characterisation of an L-Haloacid Dehalogenase from the Marine Psychrophile Psychromonas ingrahamii with Potential Industrial Application. Mar Biotechnol, vol. 15, pp. 695–705, 2013.

[71]. A. Trincone. Marine Biocatalysts: Enzymatic Features and Applications. Marine Drugs, vol. 9, pp. 478–499, 2011.

[72]. J.A. Field, F.J.M. Verhagen and E.D. Jong. Natural organohalogen production by Basidiomycetes. Trends Biotechnol, vol. 13, pp. 451–456, 1995.

[73]. C. Valverde, A. Orozco, A. Becerra, M.C. Jeziorszi, P. Villalobos, J.C. Solis. Halometabolites and cellular dehalogenase systems: an evolutionary perspective. Int Rev Cytol, vol. 234, pp. 143–199, 2004.

[74]. K. Kurata, K. Taniguchi, Y. Agatsuma and M. Suzuki. Diterpenoid feeding-deterrents from Laurencia saitoi. Phytochemistry, vol. 47, pp. 363–369, 1998.

[75]. M.T. Cabrita, C. Vale and A.P. Rauter. Halogenated compounds from marine algae. Marine Drugs, vol. 8, pp. 2301–2317, 2010.

[76]. I. Tomova, I. Lazarkevich, A. Tomova, M. Kambourova and E.V. Tonkova. Diversity and biosynthetic potential of culturable aerobic heterotrophic bacteria isolated from Magura Cave, Bulgaria. International Journal of Speleology, vol. 42 (1), pp. 65-76, 2013.

[77]. K.N. Timmis and D.H. Pieper. Bacteria designed for bioremediation. Trends Biotechnol., vol. 17, pp. 201–204, 1999.

[78]. J. S. Bowman and J. W. Deming. Alkane hydroxylase genes in psychrophile genomes and the potential for cold active catalysis. BMC Genomics, vol. 15, pp. 1120, 2014.

[79]. R. Margesin and F. Schinner. "Biotechnological Applications of Cold-adapted Organisms", in Springer-Verlag: Berlin/Heidelberg, Germany, 1999.

[80]. R. Margesin and F. Schinner. Efficiency of indigenous and inoculated cold-adapted soil microorganisms for biodegradation of diesel oil in alpine soils. Appl. Environ. Microbiol., vol. 63, pp. 2660–2664, 1997.

[81]. R. Margesin and F. Schinner. Low-temperature bioremediation of a waste water contaminated with anionic surfactant and fuel oil. Appl. Microbiol. Biotechnol., vol. 49, pp. 482–486, 1998.

[82]. R. Margesin. Alpine microorganisms: useful tools for low-temperature bioremediation. J Microbiol, vol. 45, pp. 281–285, 2007.

[83]. R. Margesin and G. Feller. Biotechnological applications of psychrophiles. Environ Technol, vol. 31,

pp. 835–844, 2010.

[84]. R. Margesin, S. Gander, G. Zacke, A.M. Gounot, and F. Schinner. Hydrocarbon degradation and enzyme activities of cold-adapted bacteria and yeasts. Extremophiles, vol. 7, pp. 451–458, 2003.

[85]. P. Bergauer, P.A. Fonteyne, N. Nolard, F. Schinner and R. Margesin. Biodegradation of phenol and phenol-related compounds by psychrophilic and cold-tolerant alpine yeasts. Chemosphere, vol. 59, pp. 909–918, 2005.

[86]. I. Krallish, S. Gonta, L. Savenkova, P. Bergauer and R. Margesin. Phenol degradation by immobilized cold-adapted yeast strains of Cryptococcus terreus and Rhodotorula creatinivora. Extremophiles, vol. 10, pp. 441–449, 2006.

[87]. N.J. Russell. Molecular adaptations in psychrophilic bacteria: Potential for biotechnological applications. Adv. Biochem. Eng. Biotechnol., vol. 61, pp. 1–21, 1998.

[88]. R. Margesin and F. Schinner. "Biodegradation of organic pollutants at low temperatures", in Biotechnological Applications of Cold adapted Organisms, R. Margesin and F. Schinner, Eds Springer, 1999, pp. 271–289.

[89]. D. Allen, A.L. Huston, L.E. Weels and J.W. Deming. "Biotechnological use of psychrophiles", in Encyclopedia of Environmental Microbiology G. Bitton, Ed., John Wiley and Sons: New York, NY, USA, 2002, pp. 1–17.

[90]. J.C. Marx, T. Collins, S. D'Amico, G. Feller and C. Gerday. Cold-adapted enzymes from marine Antarctic microorganisms. Mar. Biotechnol., vol. 9, pp. 293–304, 2007.

[91]. R. Margesin, F. Schinner, J.C. Marx and C. "Gerday. Psychrophiles, from Biodiversity to Biotechnology", Springer-Verlag: Berlin/Heidelberg, Germany, 2008.

[92]. R. Cavicchioli, T. Charlton, H. Ertan, M.S. Omar, K.S. Siddiqui and T.J. Williams. Biotechnological uses of enzymes from psychrophiles. Microb. Biotechnol., vol. 4, pp. 449–460, 2011.

[93]. G. Bell, P.J. Halling, B.D. Moore, J. Partridge and D.G. Rees. Biocatalyst behavior in low-water systems. Trends Biotechnol., vol. 13, pp. 468–473, 1995.

[94]. J. Partridge, P.J. Halling and B.D. Moore. Practical route to high activity enzyme preparations for synthesis in organic media. Chem. Commun., vol. , pp. 841–842, 1998. ???

[95]. Lohan, D. and Johnston, S., UNU-IAS Report: Bioprospecting in Antarctica, 2005. Available online: http://www.ias.unu.edu/binaries2/antarctic_bioprospecting.pdf (accessed on 14 September 2014).

[96]. J. Babu, P.W. Ramteke and G. Thomas. Cold active microbial lipases: Some hot issues and recent developments. Biotechnol. Adv., vol. 26, pp. 457–470, 2008.

[97]. T. Hamamoto, M. Kaneda, K. Horikoshi and T. Kudo. Characterization of protease from a psychrotroph, Pseudomanas fluorescenes 114. Appl. Environ. Microbiol., vol. 60(10), pp. 3878-3880, 1994.

[98]. V.S. Baghel, R.D. Tripathi, P.W. Ramteke, K. Gopal, S. Dwivedi, R.K. Jain, U.N. Rai and S.N. Singh. Psychrotrophic proteolytic bacteria from cold environment of Gangotri glacier, Western Himalaya, India. Enzyme Microb. Technol., vol. 36, pp. 654–659, 2005.

[99]. A. M. Abdou. Purification and partial characterization of psychrotrophic Serratia marcescens Lipase. J. Dairy Sci., vol. 86, pp. 127– 132, 2003.

[100]. H.K. Lee, M.J. Ahn, S.H. Kwak, W.H. Song and B.C. Jeong. Purification and characterization of cold

active lipase from Psychrotrophic Aeromonas sp. LPB 4. J. Microbiol., vol. 41 (1), pp. 22-27, 2003.

[101]. B. Joseph, P.W. Ramteke and P.A. Kumar. Studies on the enhanced production of extracellular lipase by Staphylococcus epidermidis. J. Gen. Appl. Microbiol., vol. 52, pp. 315-320, 2006.

[102]. J. Loveland, K. Gutshall, J. Kasmir, P. Prema and J.E. Brenchley. Characterization of psychrotrophic microorganisms producing betagalactosidase activities. Appl. Environ. Microbiol., vol. 60, pp. 12–18, 1994.

[103]. S. Shipkowski and J.E. Brenchley. Characterization of an unusual cold-active beta-glucosidase belonging to family 3 of the glycoside hydrolases from the psychrophilic isolate Paenibacillus sp. strain C7. Appl. Environ. Micro-biol., vol. 71, pp. 4225–4232, 2005.

[104]. M.R. Kuddus, J.M. Arif and P.W. Ramteke. Structural adaptation and biocatalytic prospective of microbial cold-active á-amylase. AFR. J. Microbiol. Res., vol. 6 (2), pp. 206-213, 2012.

[105]. M. Cotârleþ, T. Negoitã, G. Bahrim and P. Stougaard. Cold adapted amylase and protease from new Streptomyces 4 Alga Antarctic strain. Inn. Romanian Food Biotechnol., vol. 5, pp. 23- 30, 2009.

[106]. M.S. Cabeza, F. L. Baca, E.M. Puntes, F. Loto, M.D. Baigorí and I. M. Vilma. Cold-Active Pectinases from Psychrotolerant Microorganisms. Food Technol. Biotechnol., vol. 49 (2), pp. 187–195, 2011.

[107]. A. K. Maharana and P. Ray. Isolation and Screening of Cold Active Extracellular Enzymes Producing Psychrotrophic Bacteria from Soil of Jammu City. Biosciences Biotechnology Research Asia, vol. 10(1), pp. 267-273, 2013.

[108]. J.L. Arpigny and K.E. Jaeger. Bacterial lipolytic enzymes: classification and properties. Biochem J, vol. 343, pp. 177–183, 1999.

[109]. E.Y. Yu, M.A. Kwon, M. Lee, J.Y. Oh, J. Choi, J.Y. Lee, B. K. Song, D. Hahm, and J. K. Song. Isolation and characterization of cold-active family VIII esterases from an arctic soil metagenome. Applied Microbiology and Biotechnology, vol. 90 (2), pp. 573-581, 2011.

[110]. S. Li, X. Yang, S. Yang, M. Zhu and X. Wang. Technology Prospecting on Enzymes: Application, Marketing and Engineering. Comput Struct Biotechnol J., vol. 2 (3), pp. 1-11, 2012.

[111]. H. Kobori, C.W. Sullivan and H. Shizuya. Heat-labile alkaline phosphatase from Antarctic bacteria: Rapid 5' end labeling of nucleic acids. Proc. Natl. Acad. Sci., vol. 81, pp. 6691–6695, 1984.

[112]. M. Rina, C. Pozidis, K. Mavromatis, M. Tzanodaskalaki, M. Kokkinidis and V. Bouriotis. Alkaline phosphatase from the Antarctic strain TAB5. Properties and psychrophilic adaptations. Eur. J. Biochem., vol. 267, pp. 1230–1238, 2000.

[113]. E. Wang, D. Koutsioulis, H.K. Leiros, O.A. Andersen, V. Bouriotis, E. Hough and P. Heikinheimo. Crystal structure of alkaline phosphatase from the Antarctic bacterium TAB5. J. Mol. Biol., vol. 366, pp. 1318–1331, 2007.

[114]. D. Koutsioulis, E. Wang, M. Tzanodaskalaki, D. Nikiforaki, A. Deli, G. Feller, P. Heikinheimo and V. Bouriotis. Directed evolution on the cold adapted properties of TAB5 alkaline phosphatase. Protein Eng. Des. Sel., vol. 21, pp. 319–327, 2008.

[115]. R. Chen, L. Guo and H. Dang. Gene cloning, expression and characterization of a cold-adapted lipase from a psychrophilic deep-sea bacterium Psychrobacter sp. C18. World J Microbiol Biotechnol, vol. 27, pp. 431–441, 2011.

[116]. I. Leiros, E. Moe, O. Lanes, A.O. Smalas and N.P. Willassen. The structure of uracil-DNA glycosylase

from Atlantic cod (Gadus morhua) reveals cold-adaptation features. Acta Crystallogr. D Biol. Crystallogr., vol. 59, pp. 1357–1365, 2003.

[117]. Z. D. Alvarado-Cuevas, A. M. L. Hidalgo, L. G. Ordonez, E. Oceguera-Contreras, J. T. Ornelas-Salas, and A. D. Leon-Rodrý´guez. Biohydrogen production using psychrophilic bacteria isolated from Antarctica. International journal of hydrogen energy, vol. 30, pp. 1-7, 2014.

[118]. M. Debowski, E. Korzeniewska, Z. Filipkowska, M. Zielifinski and R. Kwiatkowski. Possibility of hydrogen production during cheese whey fermentation process by different strains of psychrophilic bacteria. International journal o f hydrogen energy, vol. 39, pp. 1972- 1978, 2014.

[119]. S. Manish, Rangan Banerjee. Comparison of biohydrogen production processes. International Journal of Hydrogen Energy, vol. 33, pp. 279 – 286, 2008.

[120]. I.K. Kapdan and F. Kargi. Biohydrogen production from waste materials. Enzyme Microb Technol, vol. 38, pp. 569-82, 2006.

[121]. F. Kargi, N.S. Eren and S. Ozmihci. Hydrogen gas production from cheese whey powder (CWP) solution by thermophilic dark fermentation. Int J Hydrogen Energy, vol. 37, pp. 2260-6, 2012.

[122]. L. Lu, N. Ren, X. Zhao, H. Wang, D. Wu and D. Xing. Hydrogen production, methanogen inhibition and microbial community structures in psychrophilic single-chamber microbial electrolysis cells. Energy Environ Sci, vol. 4, pp. 1329-36, 2011.

[123]. L. Lu, D. Xing, N. Ren and B.E. Logan. Syntrophic interactions drive the hydrogen production from glucose at low temperature in microbial electrolysis cells. Bioresour Technol, vol. 124, pp. 68-76, 2012.

[124]. L.R. Lynd, M.S. Laser, D. Bransby, B.E. Dale, B. Davison, R. Hamilton, M. Himmel, M. Keller, J.D. McMillan, J. Sheehan and C.E. Wyman. How biotech can transform biofuels. Nature Biotechnology, vol. 26 (2), pp. 169–172, 2008.

[125]. D.I. Massé, L. Masse and F. Croteau. The effect of temperature fluctuations on psychrophilic anaerobic sequencing batch reactors treating swine manure. Bioresource Technology, vol. 89 (1), pp. 57–62, 2003.

[126]. D.I. Massé, L. Masse, Y. Xia and Y. Gilbert. Potential of low-temperature anaerobic digestion to address current environmental concerns on swine production. Journal of Animal Science, vol. 88 (13), pp. 112–120, 2010.

[127]. L.M. Safley and P.W. Westerman. Low-temperature digestion of dairy and swine manure. Bioresource Technology, vol. 47 (2), pp. 165–171, 1994.

[128]. R.C. Kasana and A. Gulati. Cellulases from psychrophilic microorganisms: A review. Journal of Basic Microbiology, vol. 51 (6), pp 572–579, 2011.

[129]. L. Neves, R. Oliveira and M.M. Alves. Anaerobic co-digestion of coffee waste and sewage sludge. Waste Management, vol. 26 (2), pp. 176–181, 2006.

[130]. Z. Yue, C. Teater, J. MacLellan, Y. Liu and W. Liao. Development of a new bioethanol feedstock – anaerobically digested fiber from confined dairy operations using different digestion configurations. Biomass and Bioenergy, vol. 35 (5), pp. 1946–1953, 2011.

[131]. S. Weiß, M. Tauber, W. Somitsch, R. Meincke, H. Müller, G. Berg, and G.M. Guebitz. Enhancement of biogas production by addition of hemicellulolytic bacteria immobilized on activated zeolite. Water

Research, vol. 44 (6), pp. 1970–1980, 2010.

[132]. N.M.C. Saady and D.I. Massé. Psychrophilic anaerobic digestion of lignocellulosic biomass: A characterization study. Bioresource Technology, vol. 142, pp. 663–671, 2013.

[133]. P.L. Wintrode and F.H. Arnold. Temperature adaptation of enzymes: Lessons from laboratory evolution. Adv. Protein Chem., vol. 55, pp. 161–225, 2000.

[134]. K.S. Siddiqui, A. Poljak, and R. Cavicchioli. Improved activity and stability of alkaline phosphatases from psychrophilic and mesophilic organisms by chemically modifying aliphatic or amino groups using tetracarboxy-benzophenone derivatives. Cell. Mol. Biol., vol. 50, pp. 657–667, 2004.

[135]. K.S. Siddiqui and R. Cavicchioli. Improved thermal stability and activity in the cold-adapted lipase B from Candida Antarctica following chemical modification with oxidized polysaccharides. Extremophiles, vol. 9, pp. 471–476, 2005.