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Discovery of New Bioactive Secondary Metabolites from Psychrophilic Microorganisms — Mini Review

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Freezing habitats were long considered as being too harsh to support life. However, it is now known that an abundant variety of psychrophilic microorganisms can survive the harsh environment and these organisms possess amazingly diverse and biologically active secondary metabolites. Psychrophiles have to cope with some challenges, including hypersalinity of sea ice brine channels, the extremely low free water and nutrient availability in permafrost soil, and the unique light conditions. Therefore the survival of these microorganisms requires extraordinary adaptability and resistance against those stressors. This has led to the evolution of new metabolites with unique structures and biological activities. This review is an attempt to present some of the new sources of secondary metabolites produced by psychrophiles.

Key words: Bioactive metabolites; secondary metabolites; psychrophiles

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Many different microorganisms have evolved the ability to adapt and survive at low temperatures even below 0ºC. These organisms known as psychrophiles are small, unable to insulate themselves, and not motile in the sense of being able to relocate to avoid the cold [1]. Psychrophiles have therefore evolved and adopted a variety of adaptive strategies to maintain activity and metabolic function despite the challenging conditions. Cold adaptation has taken place both at the cellular and molecular levels and involves several phenotypic traits. These include changes in low molecular weight compounds, such as carotenoid pigments and compatible solutes, the latter conventionally associated with osmotic stress response [2]. The most obvious expressions of cold adaptation are, however, found in the changed composition of membrane lipids which facilitates the maintenance of membrane fluidity and in the structural changes of enzymes to retain sufficient catalytic capacity at temperatures around 0ºC and producing antifreeze protein [3].

Production of cold-adapted secondary metabolites is one such survival mechanism. One of the proposed explanations for the role of secondary metabolites in nature is its function to defend the habitats of the pschrophiles by inhibiting the growth of its competitors [4–6]. At low and non-inhibitory concentrations, such molecules are believed to function as signalling molecules [7, 8]. This is supported by the assumption that over millions of years the evolution of secondary metabolites happened because psychrophilic microorganisms used them as chemical signals for communication between cells of the same species and defending against different species [9]. As psychrophiles thrive in a different kind of climate, they must have undergone the biological evolution of its active metabolites. Studies on these psychrophilic microorganisms have provided much insight into the identification of new and novel secondary metabolites. Some of these redundant secondary metabolites have shown antibacterial and antifungal activities which will be discussed in this review. These novel metabolites from psychrophiles would be useful for the pharmaceutical industry as a potent antibiotic or as drugs for diseases.

PSYCHROPHILIC BACTERIA

Antagonism is nature's counter action of survival and existence. Bacteria produce some secondary metabolites for their defence against other microorganisms, and these secondary metabolites serve as a source of bioactive compounds. Ivanova *et al*. (2001) isolated and characterised an antimicrobial compound, 2-amino-9,13-dimethyl heptadecanoic acid (1), from Streptomyces strain, a psychrophilic bacteria from the Antarctica. This compound exhibited antibacterial activity against *Micrococcus luteus* (MIC 15 µg/ml) and gram-positive eubacteria *Bacillus subtilis* (MIC 50 µg/ml) but not against *Escherichia coli* [10].

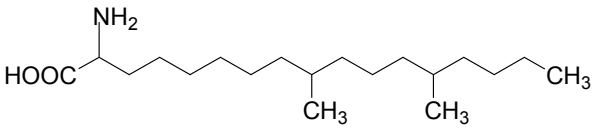
Another report on the antibacterial activity of a psychrophilic microorganism was published in 2005. Brutner *et al*. (2005) observed that frigocyclinone (2), isolated from *Streptomyces griseus* strain NTK 97, showed antibacterial activities against gram-positive bacteria whereas gram-negative bacteria like *E. coli*, *Pseudomonas fluorescens* and *Proteus mirabilis* were not affected [11]. Frigocyclinone is a new angucyclinone compound isolated. Most of the previously reported angucyclinone derivatives have exhibited a wide range of remarkable antibiotic properties. To date, angucyclic quinones are continuously growing more attractive as a class of natural products due to their structural diversity, biomedical potential, and well-defined biosynthetic pathways [12].

Mojib *et al*. (2010) reported that the yellow orange pigment of *Flavobacterium* sp. isolated from landlocked freshwater lakes in East Antarctica has antimycobacterial activity against *Mycobacterium smegmatis*. This pigment is known as flexirubin 7. Another study from the same group reported the isolation of violacein 6 from *Janthinobacterium* sp. (3). Violacein 6 is a purple violet pigment with antimycobacterial activity. Violacein is produced as a defence mechanism by sessile bacteria which are more prone to predation [13].

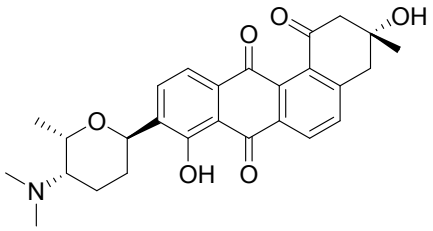
PSYCHROPHILE FUNGI

Several psychrophilic fungi have been studied for their bioactive metabolites and have been reported to be producers of new secondary metabolites.

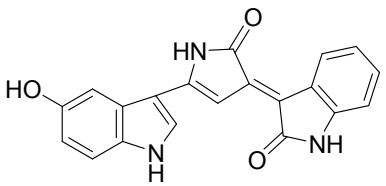
In 2004, the psychrophilic fungus *Penicillium reibeum* reported producing two new metabolites psychrophilin A (4) and cycoloaspeptide D (5) [14]. Psychrophilin A is a cyclic peptide (cyclic protein)



(1) 2-amino-9,13-dimethyl heptadecanoic acid



(2) Frigocyclinone



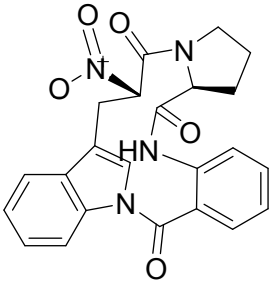
(3) Violacein 6

Figure 1. Structure of secondary metabolites isolated from psychrophilic bacteria.

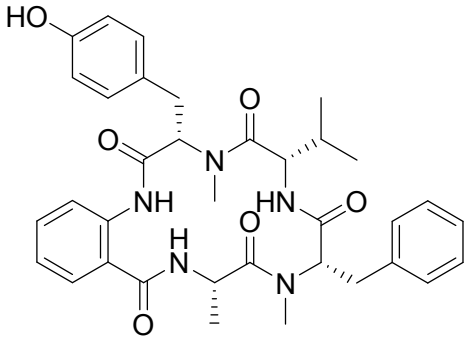
whose amino and carboxy ends are linked together with a peptide bond forming a circular chain. Some of the known cyclic peptide compounds are used as an anti-infective agent in medicines [15]. Cycloaspeptide D which is categorised as pentapeptide group have been reported to have antibiotic properties and can act as antimalarial agent [16].

Dalsgaard *et al*. (2004) isolated new psychrophilins, psychrophilin B **(6)** and C **(7)** from psycrophile fungus *Penicillium rivulum*. Further investigation on the fungus reported production of complex alkaloids communesins G **(8)** and H **(9)** [17]. However, no bioactivity has been reported for these compounds [18]. *Penicillium algidium* a psychrophilic fungus isolated from Greenland reported to produce new cyclic nitropeptide, Psychrophilin D **(10)** together with two known cyclic peptides cycloaspeptide A and cycloaspeptide D. These two known compounds were reported to be isolated from psychrophiles for the first time. Psychrophilin D exhibited cytotoxic activity against murine leukaemia cells. But, Psychrophilin D did not show inhibition in antimicrobial, antivirus and antiplasmodial activities [19].

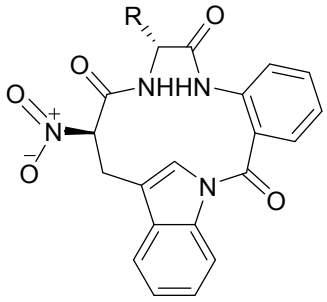
Recently Li and co-workers (2012) isolated two new epipolythiodioxopiperazines (ETPs), chetracins B **(11)** and C **(12)**, and five new diketopiperaines, chetracin D **(13)**, and oidioperaines A-D from the psychrophilic fungus *Oidiodendron truncatum* collected from the soil under lichens in the Antarctica. Oidioperaines A-D reported being an intermediate in the biosynthesis of the chetracins. Chetracins B and C have been found in metabolites with cytotoxic, immunomodulatory, antiviral, antimicrobial, and antiproliferative activity [20, 21]. ETPs, characterised by a unique bridged disulphide or polysulphide dioxopiperazine six-membered ring,



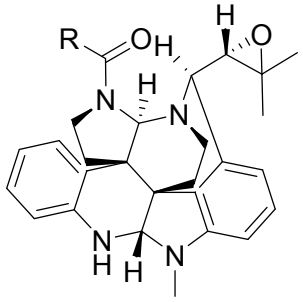
(4) Psychrophilin A



(5) Cycoloaspeptide D

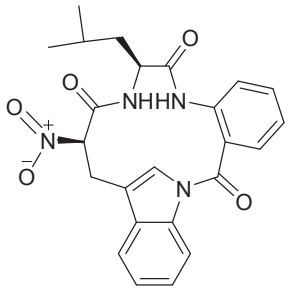


(6) Psychrophilin B: R = $CH(CH_3)_2$
(7) Psychrophilin B: R = CH_3

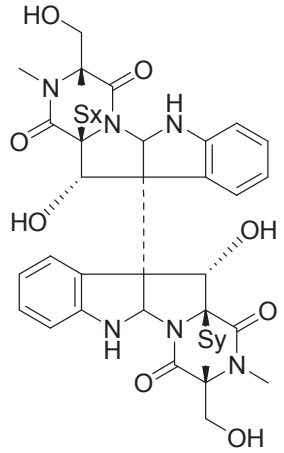


(8) Communesins G: R = CH_2CH_3
(9) Communesins H: R = $CH_2CH_2CH_3$

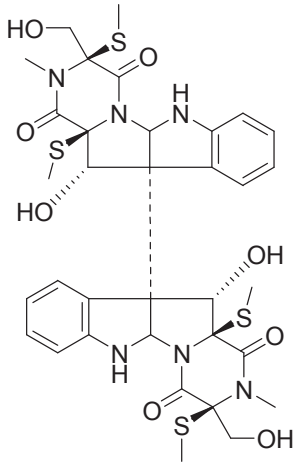
Figure 2. Structure of secondary metabolites isolated from psychrophilic fungi.



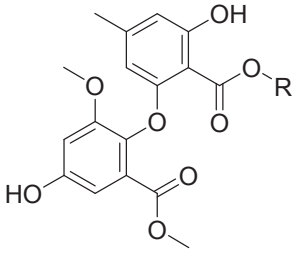
(10) Psychrophilin D



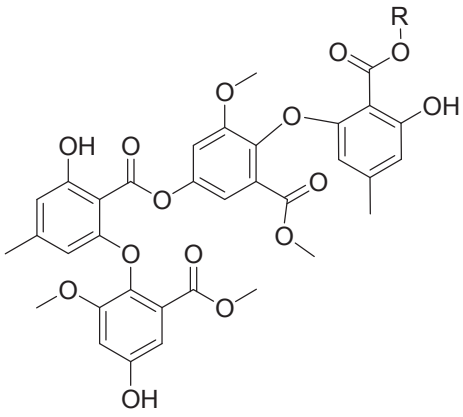
(11) Chetracins B: x = 2, y = 3
(12) Chetracins C: x = 3, y = 2



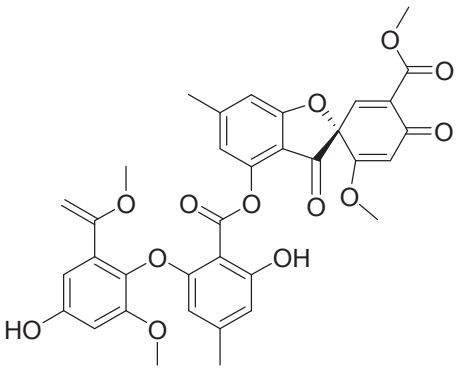
(13) Chetracin D



(14) Ethyl asterrate: R = CH_2CH_3
(15) n-Butyl asterrate: R = $(CH_2)3CH_3$



(16) Geomycin A: R = CH_3
(17) Geomycin B: R = H



(18) Geomycin C

Figure 2. (Cont.) Structure of secondary metabolites isolated from psychrophilic fungi.

occur in many fungi. Due to its broad spectra of bioactivities, ETPs have drawn wide attention in recent years [22].

Some novel compounds and metabolites with bioactive potential continue to be isolated and characterised from psychrophilic fungi which are capable of various bioactivities. In 2008, Li *et al*. isolated five new asterric acid derivatives, ethyl asterrate **(14)**, n-butyl asterrate **(15)** and geomycins A-C **(16–18)**, from cultures of an isolate of the Antarctic ascomycete fungus *Geomyces* sp. The isolate, geomycin B effectively inhibited antifungal activity against *Aspergillus fumigatus*. Also, geomycin C/bisdechlorogeodin showed antimicrobial activity against gram-positive *S. aureus* and gram-negative *E. coli* [21, 23]. It is reported that asterric acid is an antibiotic and a fungal metabolite [24]. Also, studies proposed that asterric derivatives could be useful as anti-angiogenic agents [25].

CONCLUSION

The search for natural bioactive compounds from psychrophiles has begun only in the recent years. From past and on-going research, psychrophilic organisms have shown to produce a diversified range of bioactive compounds. Therefore, it is worth giving a broader and focussed approach to the exploitation of psychrophilic organisms and the associated secondary metabolites aided by genomic analyses, applying metabolic approach and employing combined biomedical and biotechnology efforts which would lead to the discovery of some novel compounds with varying degree of bioactivity. The secondary metabolites isolated and characterised from psychrophilic microorganism would be useful in drug development for diseases or might be beneficial to the environment.

ACKNOWLEDGEMENTS

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