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Low molecular weight thiols: glutathione (GSH), mycothiol (MSH) potenail antioxidant compound from actinobacteria

Anita Garg[1*], Bhavana soni[1], Balwant Singh Paliya[2], Smarti Verma[2], Vishwjeet Jadaun[2]
Madhav Institute of Technology & Science, Gwalior – 474005, India.

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ABSTRACT

The class of *Actinobacteria* is a large group of gram-positive bacteria and having high G+C content. *Actinobacteria* also produced curious compound like thiols mainly Mycothiol (MSH) and Glutathione (GSH). MSH is also known as mycothiol and comprised of a cysteine residue with an acetylated amino group which is linked to glucosamine to inositol but Glutathione (GSH) having gamma peptide linkage between cysteine group which is attached by normal peptide linkage to a glycine and the carboxyl group of the glutamate side-chain. These thiols play a key role in maintaining a reducing environment in the cell, which is necessary for regular metabolic activities and represent adaptation under stress condition for survival of organisms. Both Mycothiol and glutathione (GSH) having property to protect cells against oxygen toxicity but MSH shows 7 fold slower ability to resistance of autoxidation compare to GSH but GSH is absent in archaebacterium and rarely found in *Streptomycetes* strains (*Streptomyces lactamdurans*). In this review article we discussed about the GSH and MSH structure, properties and how GSH is better than MSH in the case of antioxidant production.

INTRODUCTION

The class of *Actinobacteria* is a very large and diverse group of gram positive bacteria having high G and C content and producing variety of morphologies, which is start from small cocci to branched mycelia (Ventura *et al.,* 2007). *Actinobacteria* found in a wide range of ecosystems, from soil and sea water, skin, lungs, and gastrointestinal tract of humans. They are responsible for the production of commercial products, including amino acid, vitamins and antibiotics (Kalinowski *et al.,* 2003; Weber *et al.,* 2003). Members of *Actinobacteria* are causing important human disease, such as leprosy, tuberculosis, diphtheria and they are used in the biodegradation of organic compounds during bioremediation (Larkin *et al.,* 2005). Most of the *Actinobacteria* are produced Mycothiol (MSH) which is a small thiol that is often present in millimolar amounts and have analogous function to glutathione (GSH), which play a major role in protecting cells against oxygen toxicity, but its production among prokaryotes appears to be restricted to the cyanobacteria and the purple bacteria (Fahey *et al.,* 1987; Newton and Fahey, 1989; Newton *et al.,* 1993). Many strictly aerobic bacterial species have lack glutathione and produce other low-molecular-weight thiols which have analogous role of GSH. G-glutamylcysteine was found to be the major low-molecular-weight thiol in the halobacteria that is an aerobic subgroup of the archebacteria (Newton and Javor, 1985) and appears to function similarly to glutathione (Sundquist and Fahey, 1989). We about low molecular weight thiols and its properties of antioxidant activity to protect the cell and primary role of mycothiol is to maintain the intracellular redox homeostasis which acts as an electron acceptor/donor and serves as a cofactor in detoxification reactions for alkylating agents, free radicals and xenobiotics (Rawat *et al.,* 2006).

** Corresponding Author*
Anita Garg, Address: Biotechnology Department, Madhav Institute of Technology & Science, Gwalior – 474005, India. Phone +91-9074687706

LOW MOLECULAR WEIGHT THIOLS

Glutathione (GSH)

Glutathione (GSH) is a tripeptide with a gamma peptide linkage between the amine group of cysteine which is attached by normal peptide linkage to a glycine and the carboxyl group of the glutamate side-chain. It has an antioxidant activity for preventing damage to important cellular components caused by reactive oxygen species such as free radicals and peroxides (Pompella *et al.,* 2003).

GSH is absent in the aerobic archaebacterium, Halobacterium halobium (Newton and Javor, 1985), in this organism, -y-glutamylcysteine is produced at millimolar concentrations and appears to have an antioxidant role (Newton and Javor, 1985; Sundquist and Fahey, 1989). In eukaryotes the thiol is glutathione (GSH) and the glutathione *S*-transferases (GSTs) constitute a superfamily of enzymes that catalyze the reaction of GSH with a wide range of electron deficient substrates (Fahey, 2001; Jothivasan, 2008; Newton, 2008; Patel and Blanchard, 1999). Chlorinated hydrocarbons are one important class of GST substrates and 1-chloro-2,4-dinitrobenzene (CDNB) is a convenient, but not universal (Patel and Blanchard, 2001). GSTs also catalyze the reduction of organic hydroperoxides by GSH such as the addition of GSH to enals, enones and thiocarbamates, the conjugation of GSH with epoxides, and the GSH-dependent isomerization at carbon-carbon double bonds (Newton *et al.,* 2008). The GST superfamily includes at least 15 different enzyme classes (Newton *et al.*, 2008; Rawat and Av-Gay, 2007) and these enzymes are currently a common subject of research. Metal toxicity increases both factors in order to meet the elevated GSH demand in cells to ensure detoxification and survival. Sulfur is taken up by plants from the soil as sulfate. After reduction, it is assimilated into bio-organic compounds, with cysteine. Generally, this pathway is regulated by demand for reduced sulfur. Since GSH is an important storage form of reduced sulfur in cells, high demands for GSH due to metal stress stimulate sulfate uptake, reduction and assimilation in order to meet the needs of cysteine for GSH and PC biosynthesis (Davidian and Kopriva, 2010; Queval *et al.,* 2009). GSH is the more important thiol in eucaryotic organisms and play an important role in protecting cells against oxygen toxicity (Fahey RC and Sundquist AR, 1991). It presents in most gram-negative bacteria but has not been found in several major classes of gram-positive bacteria (Fahey RC et al, 1978; Fahey RC and Newton GL, 1983). According to Conflicting results presence GSH could not be found in cell extracts of Streptomyces griseus (Fahey and Newton, 1983) were reported but according to Castro JM, low levels of GSH were found in Streptomyces lactamdurans (syn. Nocardia lactamdurans) in 1983 and show streptomycetes are able to produce GSH (Fahey and Sundquist, 1991). There is one complementary compound δ-(L-oa-aminoadipyl)-L-cysteinyl- D-valine (ACV) or other low molecular-weight thiols are found which play a similar role of y-L-glutamyl-L-cysteinyl-glycine (GSH) in streptomycetes in antioxidant activity in other organisms (Gerald *et al.,* 1993). GSH and other thiols groups are reducing agents, existing at a concentration of approximately 5 mM in animal cells. One role of GSH is to serve as a slowly autoxidizable reservoir of cysteine in cells (Fahey RC and Sundquist, 1991) and converted to its oxidized form glutathione disulfide (GSSG). Multiple studies have indicated that plants exposed to any of a diverse array of metals elicit oxidative stress, a process in which the cellular redox balance between pro- and antioxidants is disturbed in favour of the former (Horemans *et al.,* 2009; Cuypers *et al.,* 2009; Cuypers *et al.,* 2011; Cuypers *et al.,* 2011; Penugonda and Ercal, 2004). Uncontrolled increases in the steady-state concentrations of these pro-oxidants lead to free radical-mediated chain reactions that target proteins, lipids, polysaccharides and DNA. It has been suggested that metal-induced oxidative stress in cells is partially responsible for the toxic effects of metals (Ercal et al 2001). In order to cope with this oxidative damage, small fluctuations in pro-oxidant concentrations play an important role in signaling processes that regulate cellular responses in cellular protection and/or acclimation to Cd or excess Cu (Cuypers *et al.,* 2011).

GSTs (glutathione *S*-transferases)

The first bacterial GST was detected in *Escherichia coli* using the spectrophotometric assay with CDNB as substrate (Shishido, 1981) it have been less extensively studied but already involved in diverse chemical processes, including metabolism of xenobioic compounds (Fahey *et al.,* 1978; Vuilleumier and Pagni, 1981). Consequently bacterial GSTs associated with the chi, theta, beta and zeta classes and determined MAPEG (membrane-associated proteins involved in ecosanoid and glutathione metabolism) classes of GSTs (Allocati *et al.,* 2009) and found mostly in cyanobacteria, proteobacteria and phylagenerally to produce GSH (Fahey *et al.,* 1987; Fahey and Newton, 1983; Newton and Fahey, 1989). Bacterial GSTs have shown a wide range of GSH-dependentactivities to catalyze which are includes detoxification of antibiotics (such as fosfomycin) (Piccolomini *et al.,* 1990), double bond isomerization of maleylpyruvate (Fang *et al.,* 2011), maleylacetoacetate (Anandarajah *et al.,* 2000), reductive and hydrolytic dechlorination of organic chlorides (Federici *et al.,* 2010; Kiefer et al.,2002; Vuilleumier *et al.,* 2001), and reduction of disulfides (Xun *et al.,* 2010). The rates for bacterial GSTs are substantially lower (5-200-fold) than those of mammalian liver GSTs (Piccolomini *et al.,* 1989).

Mycothiol

Actinobacteria produced a curious thiol compound, which is known as Mycothiol (MSH or AcCys-GlcN-Ins) (Fahey 2001; Jothivasan and Hamilton, 2008) and proposed by Spies and Steenkamp (Spies and Steenkamp, 1994) and mainly contained a cysteine residue with an acetylated amino group which is linked to glucosamine to inositol (Newton *et al.,* 2008). Mycothione is an oxidized disulfide form of mycothiol, and by the help of flavoprotein mycothione it reduced to mycothiol (Patel and Blanchard, 1999; Patel and Blanchard, 2001). Enzymes which are used in the biosynthesis of mycothiol have been anticipated to be a good target for the treatments of tuberculosis; such as mycothiol-dependent formaldehyde dehydrogenase and mycothione reductase (Newton and Fahey, 2002; Rawat and Av-Gay, 2007). According to recent reviews majority of work performed on mycobacteria and covered all perspective of MSH i.e. biochemistry, metabolism, MSH dependent enzyme and comparison with GSH- dependant enzyme (Bhave *et al.,* 2007; Rawat and Av-Gay, 2007) and it has been isolated with bioinformatics analysis from *Streptomycetes* stains and *Mycobacterium bovis* (Newton *et al.,* 1995; Sakuda *et*

al., 1994; Steenkamp, 1994). Resistance to autoxidation is a main property of MSH and it is more rapid compare to GSH (Hicks *et al.,* 2007; Meyer, 2008), autooxidation is produced hydrogen peroxide that was lethal to cells (Held and Biaglow, 1994) cysteine has an accountability to aerobic cells and maintained at a low concentration which is derivatives of amino and carboxyl groups that is present in high amount in cell to slow autooxidation that is 7-fold slower than GSH (Newton *et al.,* 1995). MSH is more resistant than GSH to auto oxidation due to absence of metal chelating residue other than thiol group.

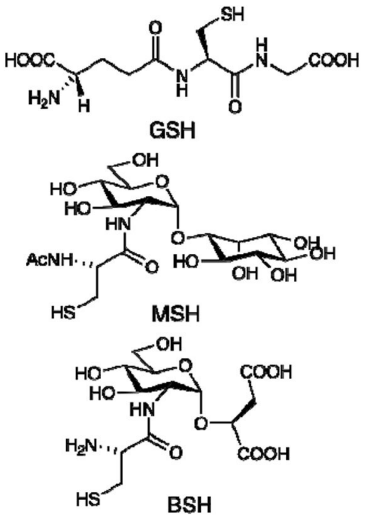

Fig. 1: Structures for glutathione (GSH), Mycothiol (MSH) and bacillithiol (BSH) (Newton *et al.,* 2011).

The structure comprised of a cysteine residue with amino group acetylation and carboxyl group which is linked D-glucosamine to α (1-1) linked myo-inositol and MSH also have an unique property that is amide linkage AcCys to GlcN-Ins which is cleaved only by specialized enzymes, such as the MSH *S*-conjugate amidase (Mca). The affinities of the substituent for heavy metals decrease in the order in cysteine residue is –S-> NH2 >COO- (Newton *et al.,* 2008).

CONCLUSION

MSH and GSH are the major low-molecular-weight thiol and act like an antioxidant in the *Actinobacteria* but GSH is rarely found in *Actinobacteria*. Both MSH and GSH are stabilized form of cysteine which is required by all organisms for protein and CoA synthesis. But MSH is 7 fold slower than GSH in case of resistance of auto oxidation which occurs in presence of oxygen and UV-radiation to form peroxides and hydro peroxides and block amino group of cysteine that is the major importance of MSH.

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