Chitins: An overview

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Chitin is a relatively recent term used to collectively refer chitin and its derivatives such as chitosan and chitosan oligomers. Chitin is a nitrogenous polysaccharide (poly N-acetyl Amino D- glucose) found in the exoskeleton of insects, shrimps, crabs, lobsters and internal structure of other invertebrates. It is the most abundant organic compound next to cellulose on the earth. Antarctic krill and squid skeleton are typically rich in chitin (~ 40%), while the dry shells of lobster, crab, cray fish, and prawns generally contain around 14-35% chitin. It is also an important structural component of the cell wall of some plant-pathogenic fungi, especially Zygomycets. The average annual production of chitin by arthropods has been estimated to be around 1, 328,000,000 MT from marine ecosystem, 28,000,000 MT from freshwater ecosystem, and 6,000,000 MT from athalassohaline ecosystem. According to Food Agricultural Organisation (FAO), a considerable amount of crustacean shells is discarded as processing waste, which means the raw material for production of this biologically active molecule is readily available at low cost (Barrow and Shahidi, 2007). Chitin is described as a colorless, crystalline or amorphous powder, which is insoluble in water, organic solvents and diluted acid and alkali. Chitin is present as chitin protein complex along with minerals mainly calcium carbonate. So the process of chitin production consists of deproteinisation with dilute alkali and demineralization with dilute acids. Chitin on deacetylation gives chitosan and on hydrolysis with concentrated HCl gives glucosamine hydrochloride. Chitin and chitosan are natural, nontoxic, biodegradable compounds with a broad range of commercial applications. Chitosan is a large, polycationic polymer having degrees of acetylation ranging from 5 to 30%. Depolymerisation of chitosan by chemical and enzymatic hydrolysis yields water-soluble chitosan oligomers or chitooligomers. Generally, chitosan with molecular weight <39 kDa and degree of polymerization <20 are known as chitosan oligomers or chitooligomers (Mourya et al., 2011)

Chitosan

Chitosan and its derivatives have been researched extensively for biomedical applications and unique biological effects such as antioxidant, anti-allergic, anti-inflammatory, anticoagulant, anti-cancer, anti-bacterial, anti-human immuno deficiency virus, anti-hypertensive, anti-Alzheimer's, anti-diabetic, anti-obesity and matrix metalloproteinases inhibitory activities. Among

these, the most extensively covered property in literature is antimicrobial property. Mainly, two different mechanisms are proposed as the cause behind the antimicrobial activity of chitosan; first one, being polycationic in nature, chitosan interfere with bacterial metabolism by electrostatic stacking at the cell surface of bacteria; the other is by way of blocking the transcription of RNA from DNA by adsorption of penetrated chitosan to DNA molecules. However, for the latter one, the molecular weight of chitosan must be small enough (~5000 Da) to be able to permeate into cell.

Structurally, chitosan has three types of reactive functional groups, namely, an amino/acetamido group as well as both primary and secondary hydroxyl groups at the C-2, C-3 and C-6 positions, respectively. The degree of acetylation and/or deacetylation of amino functionality are the main factors which contribute to the differences in chitosan structures and physico-chemical properties. Another important characteristic to consider for chitosan is the molecular weight or chain length and its distribution. Commercial chitosan ranges in molecular weight from around 1,00,000 to 10,00,000 Da. Mild hydrolysis of chitosan yields antibacterial oligomers; however, extensive hydrolysis of chitosan may result in reduced antimicrobial activity (Varun et al., 2017). Chitosan oligomeres with five to seven D-glucosamine units are reported to have good bioactivity. By modulating and improving physiological functions, chitosan and its derivatives may provide novel therapeutic applications for the prevention or treatment of chronic diseases.

Chitin and chitosan, have attracted a great attention of researchers around the world in the past few decades due to their broad range of nutraceutical and healthcare benefits. Currently, chitosan is regarded as a potential marine nutraceutical owing to its superior biological activities, biocompatibility and non-toxic nature. It is the most popular natural food additive used as a preservative in a wide array of products, including snacks and beverages. In the food industry, the hydrolysis of chitosan is aimed to decrease its bitter and astringent-taste and to increase solubility of chtosan at neutral pH. The chitosan hydrolysates exhibiting molecular weights between 30 and 41 kDa were considered to be most suitable as a food additive or functional agent as demonstrated by sensory evaluation.

Chitosan is a popular dietary fibre often used to treat obesity and high cholesterol (Schiller et al., 2001; Sumiyoshi et al., 2006; Trivedi et al., 2016). There are a number of *in vitro* and *in vivo* reports demonstrating the dietary lipid and bile acid binding activities of chitosan (Zhang et al., 2015) The cationic nature of chitosan enables it to bind to the negatively charged lipids, thereby reducing their gastrointestinal uptake and potentially lowering

serum cholesterol level. Chitosan absorb many times their weight of fat and cholesterol (. A very recent report on the clinical investigation of chitosan supplementation for 8 weeks indicated lower blood lipid level, simultaneously maintaining the normal calcium, magnesium, and iron status in elderly hyperlipidemic patients. Apart from that chitosan is reported as effective against the complications that kidney failure patients on dialysis often face, including high cholesterol, anemia, loss of strength and appetite, and disturbed sleeping (insomnia). Choi et al. (2012) have demonstrated the effect of chitosan oligomers on body weight gain, adipocyte size, adipokine level, lipid profile, and adipose tissue gene expression profile in high-fat diet-induced obese mice. Mice fed with high fat diet supplemented with 3% chitosan oligomers had gained 15% less weight but did not display any change in food and energy intake. Apart from that, chitosan supplementation markedly improved the serum and hepatic lipid profiles.

Chitosan has been used as an antioxidant for protection of oils and fats against oxidation. It was reported that chitosan at concentration of 0.02% (w/v) had antioxidant activities in lard and crude rapeseed oil. However the activity was less than ascorbic acid. Investigations by Xie et al., (2001) have demonstrated that the scavenging mechanism of chitosan is related to the fact that the free radicals can react with the hydrogen ion from the ammonium ions to form a stable molecule. Chitosan could also significantly reduce serum free fatty acid and malondialdehyde concentrations and increase antioxidant enzymes activities such as superoxide dismutase, catalase and glutathione peroxidase, indicating antioxidant enzyme regulating activities and decreased lipid peroxidation. However, the mechanism of antioxidant activity of chitosan is still disputable. Many studies have clearly shown low or no antioxidant activity of native chitosan, although the activity significantly increased with appropriate chemical modifications of the biopolymer. Studies by Je et al., (2004) have shown that chitosan may eliminate various free radicals by the action of nitrogen on the C-2 position of the chitosan. Further, the effects of dietary chitosan supplementation on lipid peroxidation and cardiac antioxidant defence system in isoprenaline-induced myocardial infarction in rats was reported (Anandan et al., 2012). Similarly, anti-aging effect of dietary chitosan supplementation on glutathione-dependent antioxidant system in young and aged rats was demonstrated (Anadan et al., 2013).

The anti-inflammatory activity of chitosan and chitosan oligomers is well documented in literature (Azuma et al., 2015). Fernandes et al. (2010) have demonstrated that the anti-inflammatory activity of chitosan oligomers in carrageenan-induced paw oedema method was not only dose-dependent

but also molecular weight-dependent at higher doses. Apart from that oral administration of chitosan oligomers was found to be effective against intestinal inflammation and mortality in mouse model of acute colitis.

Chemically modified biofunctional chitosan derivatives is a new addition to the industry, and may open up new applications in functional food and nutraceutical development. Two new conjugates, namely vanillic acid and coumaric acid grafted chitosan derivatives with superior antioxidant and antimicrobial activities compared to native chitosan was developed by Chatterjee et al. (2016). Besides, chitosan has been successfully used in controlled or targeted delivery systems of nutrients and bioactive compounds. Chitosan has several advantages in encapsulation over other biopolymers, namely; ability to adhere to the gastric mucosa, lack of allergic or irritant reaction, pH dependent controlled release of the encapsulated bioactive material etc. It has been successfully used for encapsulation of cashew apple extract, olive leaf extract, tuna oil, enzymes, lactose and various antioxidants. The mounting research data published in this area every year evidently indicate the growing interest in application of chitosan and chitosan derivatives in health and nutrition.

Glucosamine

Glucosamine (2-amino-2-deoxy-D-glucose) is an amino-monosaccharide derived by the hydrolysis chitin. Glucosmaine is primarily a component of articular cartilage, intervertebral disc and synovial fluid. Glucosamine is chemically glucose in which a hydroxyl group on the second carbon atom is substituted with an amino group. It crystalizes as glucosamine hydrochloride during purification under acidic conditions. Among the various derivatives, glucosamine hydrochloride and sulfate are the most commercialized forms worldwide. Glucosamine, is classified as a 'safe dietary supplement' and is widely marketed for pain relief in osteoarthritis.

With age, the body's ability to produce glucosamine may become impaired, resulting in considerable dysfunction and pain. As a "building block" of cartilage, glucosamine appears to have an ability to treat osteoarthritis by protecting and strengthening cartilage, allowing it to retain its cushioning effects and lubricate the joints. It also plays a role in preventing further joint damage, helps to reduce inflammation and supports pain-free movement of the joints by enhancing cartilage synthesis and inhibiting cartilage break down.

There is now a large convergent documental evidence that glucosamine sulfate, given at a daily oral dose of 1,500 mg, is able to significantly reduce the symptoms of osteoarthritis (Reginster et al., 2012). It is one of the amino sugars used by biological systems for bringing modification to the

functions of proteins. Although glucosamine was discovered long back, the interest in neutraceutical use received great attention since last two decades. The rationale in using glucosamine for arthritis is that in the joint and synovial fluid glucosamine will stimulate the synthesis of proteoglycans that help in repair of damaged cartilage, such as hyaluronic acid, heparin sulphate, and keratan sulphate. In the cartilage system, proteoglycans are intertwined with the collagen network. Due to the net negative charge of the proteoglycans, a large amount of water is enclosed in the cartilage mass. This water content is important for the resilient and elastic properties of collagen fibrils as well as for the lubrication of the joint system. Also, as a "building block" of cartilage, glucosamine appears to have an ability to treat osteoarthritis by protecting and strengthening cartilage, allowing it to retain its cushioning effects and lubricate the joints. It also plays a role in preventing further joint damage, helps to reduce inflammation and supports pain-free movement of the joints by enhancing cartilage synthesis and inhibiting cartilage break down.

Glucosamine has been designated as an 'over the counter' dietary supplement by the US Food and Drug Administration. Although there are contradictory reports on the effectiveness glucosamine in the treatment of osteoarthritis, there are more than 150 generic preparations of glucosamine alone or in combination with similar supplements in the global market. Taniguchi et al. (2012) reported that long-term oral administration of glucosamine sulfate reduced the destruction of cartilage and upregulation of MMP-3 mRNA in a model of spontaneous osteoarthritis in Harley guinea pigs. Oral administration of glucosamine sulfate for at least 12 months may prevent the need for knee arthroplasty, revealing the profound extent of the disease-modifying power of this compound. Collagen peptide is also reported to have synergistic effects with glucosamine. Inspite of all promisive results, the use of glucosamine in the management of osteoarthritis remains controversial and its specific mechanism of action in pain relief and function modification are still unclear. Animal experiments have shown that, glucosamine is having good peptic ulcer healing properties. Oral administration of glucosamine helps in the synthesis of gastric mucosa to repair the ulcer and provides pain relief. Latest research has claimed that glucosamine supplementation mimics low calorie diet in rats and increased the life span compared to control animals. Calorie restriction was proven in animals to improve the life span in laboratory studies. Gliucosamie was shown to reduce the amount of glucose metabolized through the glycolytic pathway thus mimics low calorie diet.

Glucosamine is a more recent entry to the nutracosmetic category. It can increase the skin's content of hyaluronic acid to increase moisturization,

leading to enhanced skin barrier properties and reduced dryness. Glucosamine has also been reported to have potential to inhibit skin melanin production. Glucosamine has been shown to inhibit glycosylation, the addition of polysaccharide units to proteins in in-vitro melanocyte cell culture. Glycosylation is a required step in the conversion of certain inactive pro-enzymes to their active forms. Active tyrosinase, a key enzyme in the pathway for melanin production, is glycosylated. Thus, glucosamine inhibits the production of melanin in melanocytes.

Apart from the well-known antiarthritic and antiaging potential, recent evidences suggest a potential beneficial effect of glucosamine against cancer risk . In these studies, the anti-cancer activity was more correlated with either one of, a decreased DNA synthesis, cell cycle arrest in G1 phase, induction of apoptosis, or inhibition of protein N-glycosylation. Based on a few other observations, glucosamine has been found to be useful for ameliorating inflammamtory bowel disease, migraine, and viral infections. In addition to that, there have been a number of studies suggesting the anti-inflammatory, antioxidant, antifibrotic, neuroprotective and cardioprotective activities of this aminosugar, enlisting it as an ideal neutrceutical supplement for meeting many of the dietary requirements.

Glucosamine is a natural component of the human body, hence, more compatible and does not impose any side effects. Glucosamine is normally synthesised in our body from its precursors however, in instants of osteoarthritis, glucosamine supplementation may be beneficial. An observed safety level (OSL) of up to 2000 mg/day reported for glucosamine, supports a confident conclusion of their long-term safety. Hence, it may be wiser option to consider the use of glucosamine as a combination therapy with other dietary supplements for better and promisive results. It is available commercially in different forms in market, including in combination with herbs, vitamins, creatine, chondroitin sulphate, ascorbic acid, manganese or dimethylsulfone. Of the various available forms of commercially available glucosamine, glucosamine sulfate is found to be more effective for treating osteoarthritis.

Suggested Readings

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