INTRODUCTION

Medicinal and aromatic plants have always been intimately linked with human health and culture. Medicines, derived from plants, constitute about 80% of the present day human healthcare systems in developing countries (WHO 2008). Secondary plant products (bioactive constituents), produced by the medicinal and aromatic plants, are derivatives of plant secondary metabolism and are involved in many other aspects of a plant’s interaction with its immediate environment. Since the times immemorial, humans have been using the active compounds derived from plants for treating diseases. Some of the important medicinal products obtained from plants include morphine (analgesic), codeine (anti-tussive), reserpine (antihypertensive), digoxin (cardiotonic), vincristine and vinblastine (anticancer) and artemisinin (antimalarial).

Diabetes mellitus is one of the top 5 significant diseases of the developed nations. Diabetes mellitus is the fastest growing metabolic disease of the world and the number of diabetic patients is likely to increase to 439 million by the year 2030 (Shaw et al. 2010). According to International Diabetes Federation (IDF) (2013) there are 382 million people living with diabetes and by 2035 this will rise to 592 million. By 2030, the number of diabetics in India will cross the 100 million mark (IDF 2013). Therefore, efforts are being made to look for the anti-diabetic drugs, which are safe and have no side effects. Diabetes mellitus is a heterogeneous metabolic disorder characterized by hyperglycaemia resulting in defective insulin secretion, resistance to insulin action or both (Jacobsen et al. 2009). Although the drugs biguanides and sulphonylurea are valuable in the treatment of diabetes mellitus, their use is restricted by their limited action, pharmaco-kinetic properties, secondary failure rates and accompanying side effects (Krentz and Bailey, 2005). Moreover, these therapies only partially compensate for metabolic derangements seen in diabetes and do not necessarily correct the fundamental biochemical lesion (Taylor and Agius. 1988; Bailey et al. 1989). Therefore efforts are being made to look for the natural and safe anti-diabetic drugs. There are many plants which show hypoglycaemic activity and one such plant is fenugreek (*Trigonella foenum-graecum* L.), which is recommended for diabetic patients to take as vegetable. Fenugreek has been extensively used in different systems of medicine as a source of anti diabetic compounds obtained from its seeds, leaves and extracts (Raju et al. 2001; Srinivasan 2006; Khalki et al. 2010).
Fenugreek, a native to Southern Europe, the Mediterranean region and Western Asia, is an annual herb of family Fabaceae. Fenugreek is locally known as Methi and is cultivated throughout India for culinary and medicinal purposes. It is also grown as fodder. This plant is well known for its miraculous medicinal properties. The plant extract is anti-allergic (Thiel 1997), antipyretic (Ahmadiani et al. 2001), anti-diabetic (Gupta and Prakash. 2001; Kumar et al. 2005), anti-cholesterolemic (Hannan et al. 2003) anti-malarial, antioxidant (Madhava et al. 2011), and anti-cancerous (Khoja et al. 2011; Alsemari et al. 2014); it cures anaemia and respiratory disorders (Kaviarasalan et al. 2004), relieves skin irritation and is used in the treatment of indigestion and flatulence (Sauvare et al. 1991). It is an immunomodulatory agent and is also used as galactagoguc (Gaby 2002). Medicinal properties of fenugreek can be attributed to its bioactive constituents like 4-hydroxyisoleucine (4-HI), trigonelline, galactomannans, diosgenin, flavonoids, carotenoids, coumarins, proteins, saponins, and lipids. Fenugreek seeds act as dietary supplement and significantly reduce the symptoms of diabetes mellitus (DM) such as polydipsia, polyuria, urine sugar, renal hypertrophy and glomerular filtration rate (Ulbricht et al. 2007). Fenugreek seed-alkaloid, known as trigonelline (Figure 1) controls diabetes mellitus through operation of the mechanism of insulin secretion, modulation of β-cell regeneration and stimulation of activity of glucose metabolism related enzymes (Hamza et al. 2012; Zhou et al. 2012).

Plant growth regulators enhance the production of secondary metabolites in plants. It has been well documented that the foliar application of plant growth regulators augment the productivity of plants and enhance the active constituents of medicinal and aromatic plants (Alamet et al. 2012; Nacem et al. 2012, 2014; Ahmad Dar et al. 2015; Idrees et al. 2015). The application of GA₃ exerts numerous growth effects on plants such as those belonging to cell elongation, accumulation of flavonoids, anthocyanins and glutathione, and modulation of the activities of enzymes such as peroxidise, chlorophyllase and phenylalanine ammonia lyase (PAL) (Sharaf-Eldin 2007). Exogenously applied GA₃ improved the accumulation of steroidal alkaloids in Solanum aviculare (Subroto and Doran. 1994) and enhanced the alkaloids content in stem, leaves and roots of Catharanthus roseus (Srivastava and Srivastava, 2007). Similarly, significant effect of kinetin application was observed on growth, productivity, and yield in Nigella sativa (Shah 2007a). Besides, kinetin influenced
Figure 1. Structure of Trigonelline
diverse plant processes like plant cell division, delay of leaf senescence, stimulation of leaf area expansion, chlorophyll accumulation (Pospílova et al. 2000) and dry matter production (Davies 1995; Shah 2007a).

Plants necessitate macro- and micro-nutrients, which are essential for a plant to complete its life cycle (Maathuis and Diatloff, 2013). Phosphorus (P) is one of the essential macronutrients for plant growth and development, constituting about 0.2% of plant dry weight (Harrison 2002). Moreover, it is an important component of biomolecules such as nucleic acids, phospholipids and adenosine triphosphate (ATP). The amount of P present in the soil may be high, but it may be present in unusable (fixed) forms, which cannot be taken up by plants as such; it limits the soil-P availability to plants, resulting in phosphorus deficiency in plants (Kanwar and Grewal, 1990). In India the agricultural soils are mostly deficient in phosphorus (Hasan 1996). To overcome the P deficiency, different kinds of phosphate fertilizers are applied to the soil mainly in the case of legumes, which carry inherent potential of phosphorus utilization compared to other crops (Gentili et al. 2006; Rotaru and Sinclair, 2009).

Radiation processed natural polysaccharides can be utilized in the field of agriculture after converting them to more useful forms (Sabharwal et al. 2004). According to this technique, the natural polymers, like chitosan, carrageenan and sodium alginate, are subjected to gamma rays irradiation, which breaks them down to lower molecular weight oligomers. The oligomers, obtained by the radiation-processing of natural polysaccharides, when applied to the plants as foliar sprays, or added to the tissue culture medium and hydroponics solutions as supplements, positively influenced various biological and physiological activities of plants including seed germination, shoot elongation, root growth, flower production, phytoalexin induction, stimulation of antimicrobial activity, amelioration of heavy metal stress, reduction of the use of insecticides and chemical fertilizers, shortening the harvesting period of various crops and enhancement inplant growth in general (Kume et al. 2002; Lee et al. 2003; Luan et al. 2003; Hu et al. 2004; Sabharwal et al. 2004; Hegazy et al. 2009; Aftab et al. 2011; Khan et al. 2011; Sarfaraz et al. 2011).

Alginates are prominent natural polysaccharides available in nature. They are present in the cell wall of marine brown-algae like Laminaria hyperborean, Macrocystis pyrifera, Laminaria digitata, Scophyllum nodosum, Laminaria japonica,
*Edonia maxima*, *Lessonianigrescens* and *Sargassum* spp. Sodium alginate is the sodium salt of alginic acid, derived from brown algae and consists of residues of homopolymeric poly-β-(1, 4) D-mannuronic acid and poly-α-(1, 4) L-guluronic acid. Irradiated sodium alginate (ISA) enhanced the growth and productivity of *Zea mays* (Hu et al. 2004; Hegazy et al. 2009). Irradiated sodium alginate is also known to enhance the alkaloid production and amount of active constituents in different medicinal and aromatic plants like *Papaver somniferum*, *Foeniculum vulgare*, *Mentha arvensis*, *Eucalyptus* spp., *Artemisia annua* and *Cymbopogon flexuosus* (Idrees et al. 2011; Khan et al. 2011; Sarfaraz et al. 2011; Aftab et al. 2014; Ali et al. 2014; Naeem et al. 2014; Idrees et al. 2015).

Chitosan is one of the natural marine polysaccharides consisting of randomly distributed β-(1-4)-linked D-glucosamine (deacetylated) and N-acetyl-D-glucosamine (acetylated) units. It is extracted from crustaceous shells, exoskeletons of insects and shrimps and cell walls of fungi (Shahidi et al. 1999). Using gamma irradiation, chitosan is degraded by into lower molecular weight units (oligomers), which are soluble in water after initially treating with acetic acid. The chitosan-oligomers have special biological, chemical and physical properties, different from that of the unirradiated chitosan. Foliar application of gamma-irradiated chitosan (IC) showed useful biological activities such as antibacterial activity (Zheng and Zhu, 2003), antifungal activity (Jeon et al. 2001) and antitumor activity (Qin et al. 2002). Irradiated chitosan has been reported to induce various other biological activities in plants (Nge et al. 2006; Chmielewski et al. 2007; Hossain et al. 2013), which resulted in promotion of growth in plants such as rice and wheat (Tham et al. 2001), orchids (Nge et al. 2006), barley and soybean (Luan et al. 2006) and *Eucalyptus citriodora* Hook (Ali 2014). Therefore, it was hypothesized whether the irradiated sodium alginate (ISA) and irradiated chitosan (IC) could be used as plant growth promoters in the present study on fenugreek.

Keeping in mind the immense medicinal importance of fenugreek and the role of plant growth regulators, mineral nutrients and depolymerised form of natural polysaccharides in increasing productivity and secondary metabolite production of plants, five pot experiments were conducted on fenugreek. The aim of the experiments conducted was to investigate whether foliar application of the selected plant growth regulators (gibberelllic acid and kinetin) and irradiated natural
polysaccharides (sodium alginate and chitosan), applied alone and in combination with phosphorus soil-dressing, could enhance the growth, yield and alkaloid content of fenugreek (the anti-diabetic plant). It is need of the hour because the demand for anti-diabetic drugs is far more than that of supply and researchers are working round the world towards improving anti-diabetic active constituents of plants by various means. So far, different strategies have been used for enhancing the growth, yield and active constituents of fenugreek by different workers (Khan et al. 2005; Jagdale and Dalve, 2010; Singh 2010; Bairva et al. 2012; Jat et al. 2012; Danesh-Talab 2014).