Chapter 2

Review of Literature

Underground stem (rhizome) of *Zingiber officinale* (Ginger) has been used as a medicine in Asian, Indian, and Arabic herbal traditions since ancient times (Altman and Marcussen, 2001). It has been used extensively for more than 2500 years in China for headache, nausea, and colds (Grant and Lutz, 2000) and in Mediterranean (Sharma and Clark, 1988) and Western parts in herbal medicine practice for the treatment of arthritis, rheumatological conditions and muscular discomfort (Bordia *et al.*, 1997, Langner *et al.*, 1998).

During the thirteenth and fourteenth centuries, next to pepper, ginger was the commonest and most precious of spices, costing nearly seven scroling per pound, or about the price of a sheep (Watt, 1872; Mahindru, 1982). The literature also indicates that ginger preserved in syrup (called Green Ginger) was also imported to the Western World during the middle ages and was regarded as a delicacy of the choicest kind. In Africa, ginger is regarded as auspicious, which is absolutely necessary to the Savaras tribe for their religious and marriage functions (Mahindru, 1982).

During the Middle Ages and until the end of the nineteenth century English tavern keepers used to have ground ginger in constant supply for thirsty customer to sprinkle on top of their beer or ale and then stir into the drink with a red-hot poker (Rosengarten, 1969). The Western herbalists and naturalists knew the great qualities of ginger as confirmed by the well-known British herbalist John Gerad. He writes in his herbal
that “ginger is right good with meat in sauces,” and says that this spice is “of an eating and digesting quality (Parry, 1969).

2.1 History of ginger in India

In olden India, ginger was not important as a spice, but it was named as *mahabhesaj, mahaoushadhi*, literally meaning the great cure, the great remedy. For the ancient Indian, ginger was the God given universal remedy for a number of ailments. That may be the reason why ginger found a prime place in the ancient Ayurvedic texts of Charaka (*Charaka samhita*) and Susruth (*Sushrutha samhita*). In *Ashtangahridyam* of Vaghbhatt (a very important ancient Ayurvedic text), ginger is recommended along with other herbs for the cure of elephantiasis, gout, extenuating the juices, and purifying the skin from all spots arising from scorbutic acidities. Ginger is also recommended when exotic faculties were impaired due to indigestion.

Rabbi Benjamin Tudella, who travelled between 1159 and 1173 A.D and gave an account of ginger cultivated on the west coast of India. Tudella gave a vivid description of the place and trade in spices as well as cultivation of spices in and around the port of Quilon in the State of Kerala (Mahindru, 1982). Marco polo (A.D. 1298), in his famous travelogue, writes “good ginger also grows here in known by the name of Quilon ginger. Another traveller, Friar Odoric (A.D. 1322), writes. Quilon is at the extremity of pepper forests towards the south. Ginger is grown here, better than anywhere else in the world and huge quantities.

Linschotten (1596) presented a very interesting description of the spices. He observed that ginger grew in many parts of India, but the best and the most exported grew on the coast of Malabar.
Ridley (1912) gave a detailed description about ginger and turmeric practices prevalent in nineteenth-century India. Buchanan (1807) made many references on the cultivation of various spices, including ginger, during the journey to south India. Later on, ginger cultivation spread from Kerala to various other parts of India, mainly to Bengal and North-eastern states.

2.2 Taxonomical review of ginger

Roscoe (1807) described *Z. officinale* from a plant in the Botanic Garden at Liverpool as “Bracteis ovato-lanceolatis, laciniiis corolla revolutis, nectario trilobato” and referred to *Amomum zingiber* Willd. Willdenow (1797) extended Linnaeus description “Amomum scapo nude, spica ovata” with “squamis ovatis, foliis lanceolatisbad apicem margine ciliates.”

Linnaeus (1753) *Amomum zingiber* is the basionym for the species. The genus *Amomum* of Linnaeus is a nomenclatural synonym of the conserved generic name, *Zingiber* Boehm (Burt and Smith, 1968). The specific epithet *zingiber* could not be used in the genus *Zingiber*. Thus, *Z. officinale* was adopted as the correct name for ginger. The specimen available in most herbaria are without flowers, and it is assumed that Linnaeus based his description on the account and figure given by Rheede in *Hortus Malabaricus*. The figure given by Rheede (Vol., 11, plate 12, 1692) is the designated lecto type of the species *Z. officinale* Rosc. (Jansen, 1981). The species epithet *officinale* was derived from Latin, meaning “work shop,” which in early Latin was used to mean pharmacy, thereby implying that it had a medicinal use.
2.3 Tissue culture work

*In-vitro* plant tissue culture has become more powerful tool after the discovery of “Growth Regulator” auxin (Arteca, 1996) and Cytokinins (Haberlandt, 1913) followed by formulation of artificial nutrient medium (Murashige and Skoog, 1962) which was the breakthrough of plant tissue culture because most of the horticulture crops can be micropropagated in this medium.

The potential of tissue culture in various aspects of plant improvement has already been recognized and attracted the attention of scientists. Rapid multiplication of propagation materials through tissue culture, particularly in the initial selection, assumes importance in plant improvement programmes. The potential of tissue culture in various aspects of plant improvement has already been recognized and attracted the attention of scientists. Rapid multiplication of propagation materials through tissue culture, particularly in the initial selection, assumes importance in plant improvement programmes. It also holds promise in the development of pure lines through haploid technology, production of triploids through endosperm culture, culture of embryos of incompatible crosses and isolation of somaclonal variants (Singh, 1978).

Tissue culture as a tool for vegetative plant propagation is relevant for crop plants that resist conventional asexual propagation (Hackett, 1966). The various explants such as axillary bud, shoot tips, meristem tips, root tips are commonly used. *In-vitro* ginger multiplication, dormant buds on excised rhizomes can be forced to form shoots which can be rooted. This method is rather slow, particularly for plant breeders, as on an average only 20 plants can be produced per year from single, one year old plant (Leffring, 1971).
Literatures on ginger tissue culture are limited. Hosoki and Sagawa (1977) first reported on the induction of maximum of 6 shoots per explant under in-vitro culture of ginger. Later Illahi and Jabeen (1987) conducted an experiment in ginger using different explant materials viz. immature buds, rhizome cutting with shoot bud primordia and juvenile shoots and observed/ obtained efficient plant rejuvenation. Haung (1995) observed that ginger plants were regenerated from the shoot tip explants of 0.2 to 0.9 mm in length were best for in-vitro propagation.

Most of the earlier work showed that the shoot tips or new emerging buds are the right explants for in-vitro propagation of ginger (Sharma and Singh, 1997; Pandey et al., 1997; Rout. et al., 2001; Kambaska and Santilata, 2009). Lange et al., (1987) reported elimination of nematodes infection from the rhizomes of ginger using in-vitro techniques. Inden et al., (1988) reported that each shoot bud explant produced more than four shoots within nine weeks on MS medium containing BA 5 mg/l and NAA 0.5 mg/l. Balchandran et al., (1990) reported successful in-vitro propagation Curcuma longa using rhizome buds as explants on MS medium with different combinations of BA and kinetin.

Malmug et al., (1991) reported that shoot proliferation of the regenerated shoots was induced with the addition of NAA 1 mg + BA 5 mg/l. Dogra et al., (1994) achieved in-vitro propagation of Zingiber officinale using rhizome buds. The buds produced multiple shoots when cultured on MS medium with BA 2.5 mg/l and NAA 0.5 mg/l.

Palai et al., (1997) observed that when Zingiber officinale cultivars cultured on medium supplemented with increased concentration of BA from 6 to 8 mg/l, there was decreased multiplication of shoots.
Pandey et al., (1997) reported multiple shoot production on MS medium with BA 5 mg/l + NAA 0.5 mg/l using pseudo-stems of ginger as explants. Sharma and Singh (1997) conducted in-vitro culture in rhizomes of ginger cv Himachal local and found 7.7 multiple shoots per bud on MS medium fortified with kinetin 2 mg/l and sucrose 20 g/l after four week of culture and 6.8 cm shoot length and 7.0 cm root length was found in MS medium kinetin 2 mg/l, NAA 2 mg/l and 20 g sucrose per litre. Simultaneously, obtained well developed rhizomes from micropropagated plants which was not affected by Fusarium oxysporum during storage for six months and developed a method in checking storage rot caused by F. oxysporum.

Pandey et al., (1997) obtained disease free ginger plantlets was developed through plant tissue culture protocol with very less cost effective in MS medium with BA 5 mg/l combination with NAA 0.5 mg/l concentration, the highest number of shoots 5.33 shoots per pseudostems after 5 weeks of culture. In same medium highest shoots length and 4.33 numbers of roots were recorded simultaneously.

In 2001 from Orissa (India) an efficient protocol was standardized on (Zingiber officinale cv. V3 S18) ginger in production of (92.2 % of plant show) shoot multiplication in MS medium combination with BAP 26.6 µM, IAA 8.57 µM and adenine sulphate 111.1µM with 3% sucrose, and in-vitro rhizome formation was found in MS medium supplemented with BA 4.44µM, IAA 5.71 µM and sucrose 3-8% after eight week of culture. They observed that successful production of multiple shoots and in-vitro rhizome formation depended on the nutrient medium and the culture environment Rout et al., (2001).

Khatun et al., (2003) studied a rapid shoots multiplication through new shoot tip of ginger rhizome on MS medium with 3 % sucrose and 0.5 % agar fortified with BAP
2.5 mg/l and kinetin 0.5 mg/l. Rooting was also observed on same culture medium after 45 days of culture.

Rha et al., (2007) standardized a systematic protocol for complete plant regeneration using shoot apical meristems culture for *(Zingiber officinale)* ginger. Wanju of Korea. Callus was observed on explant in culture medium MS fortified with a combination of NAA 1 mg/l and kinetin1.0-2.0 mg/l and IAA 0.1 mg/l and BAP 1.0-2.0 mg/l. Most of the shoot differentiation from callus occurred on MS fortified with IAA 0.1 mg/l and BAP 1.0 mg/l. Regenerated plants were exposed to CO2 concentration and observed a 400-4000 ppm increase in atmospheric CO2 concentration leads to increase in adventitious bud and shoot primodium and reduced at the concentration of 8000 ppm.

Zeng et al., (2008) observed in increase in *in-vitro* micro rhizome production of *(Zingiber officinale* Roscoe) ginger on combination of 80 g/l sucrose, 2X MS microelements and 1 X micro element, with a photoperiod of 24 L: 0D (Light/Dark).

Kambaska and Santilata (2009) observed an efficient protocol of *Zingiber officinale* Roscoe, Suprava and Suruchi of Orissa (India) using fresh rhizome bud/sprout culture/micropropagated on semi solid Murashige and Skog's medium with different growth regulator with various concentration and combination of BAP and NAA for shoot and root induction. Multiple shoots of 7.5 numbers found at MS medium supplemented with in BAP 2 mg/l and NAA 0.5 mg/l combination with average shoot length of 6.2 cm and best rooting medium was half MS with NAA 2 mg/l concentration was observed.

Hassan *et al.,* (2009) reported on micropropagation of *(Zingiber officinale* Roscoe) ginger cv. Suruchi and Bari (Ada) using three different explant i.e. leaf, shoot
tip and root, using different combination and concentration of growth regulator in MS medium. Studied on callus induction through five quantative traits i.e. days required for callus induction, size of callus induction, colour of callus and percentage of callus induction. Cultivar variety Suruchi observed 62.64% callus induction, 63.98% shoot induction and 68.76% root induction. Leaf explant found the best explant over the root and shoot tip explant, with the callus, shoot and root induction were 62.64%, 63.98 and 68.76% respectively in MS medium fortified with Dicamba 0.5 mg/l for callusing, MS with kin 1.0 mg/l + BAP 1.0 mg/l for shooting and MS + IBA. 1 mg/l Regenerated in-vitro plants were successfully established in pot and to field.

An efficient in-vitro protocol was developed on Zingiber officinale Roscoe., var-Varda through direct regeneration of vegetative buds on LSBM medium supplemented fortified with BAP (17.76µM) with 96% initiation response (Kavyashree, 2009). Rapid shoot multiplication was observed at the average rate of 4 fold per culture. This efficient protocol for multiple shoots and roots on the same medium after 2-3 subcultures eliminated the steps of in-vitro rooting. The statically analysis pertaining to multiple shoots and roots gave highest mean number of 19.1 and 12.3 respectively. The regenerated plants were successfully established in the field with 86% survival frequency.

Bhaskaran et al., (2009) have reported a protocol for indirect and direct somatic embryogenesis from aerial stem explants of ginger (Zingiber officinale Roscoe.) using aerial stem explant of two ginger varieties were cultured on different concentration of 2, 4-D to induced callus. Two types of callus were found, type I callus was observed with hard, nodular and yellowish in colour and type II observed soft, sticky with pale white colour. The somatic embryo was found in the medium MS
supplemented with BAP 2 mg/l. Direct somatic embryo was observed in the medium MS supplemented with thidiazuron alone or combination with IAA. Histological studied found that the somatic embryo in ginger has a distinct single layered epidermis, scutellum, coleoptile, shoot apex and root apex.

Vilamour et al., (2010) studied in-vitro the effects of media strength and source of nitrogen on shoot and root growth of ginger, native variety. Ginger plant was observed significantly proliferation in nitrogen source in the form of KNO3 in full and half strength media.

2.4 Genetic fidelity of micropropagated clones

Tissue culture techniques plays very important role in rapid multiplication of desired clones, simultaneously provide great contribution in conservation of rare and endangered species. These techniques are highly space efficient, minimize disease and pest problems and allow for the manipulation and control of all external variables, which may cause inimitable loss of important mother plants when collections are maintained outdoors.

True-to-type clonal fidelity is one of the most important pre-requisites in micropropagation of crop species (Sharma et al., 2009). Axillary branching or somatic embryogenesis way give rise to genetically uniform and true-to-type plants, as the structured meristems have generally been considered to be immune to genetic changes that might arise during cell division or differentiation under in-vitro conditions (Vasil, 1985; Shenoy and Vasil, 1992).

Somaclonal variation mostly occurs from plantlets derived from in-vitro culture is manifested in the form of DNA methylations, chromosome rearrangements, and point
mutations (N Swkcroft, 1981; Phillips et al., 1994) such variations are heritable too (Breiman, 1987) and is therefore not desirable in clonally propagation. Several studies were conducted to screen somaclonal variations produced in tissue cultured plants such as in turmeric, *Lillium* species, neem, tea and soya bean in case of oil palm, where aberrant flowering patterns were observed among the regenerated plants (Matthes, 2001). Reports of somaclonal variation in tissue culture derived plant material have been described for many species including, horseradish, pecan and alfalfa (Hofmann, 2004; Singh, 2002).

It is very important to detect genetic fidelity at early stage of micropropagated plants because variations may be detected only at late developmental stages, or even in the offspring. Variation can also occur when plants are placed under different culture conditions, which may induce stress like responses. These include media with high sugar concentrations and temperature reduction. Thus it would be very important to monitor these variations quite early in the life of plant to prevent from adverse effect which may prove to be economically disastrous. Researchers tried to assess tissue culture induced variations can be determined at the morphological, cytological, biochemical, and molecular levels with several techniques, but most of the techniques have their own limitations. Cytological analysis cannot study in specific rearrangements of genes in chromosome level (Isabel et al., 1993). Using polymerase chain reaction (PCR), DNA based markers are the best markers which are not influenced by environmental factors and generate reliable, reproducible results. Though Restriction Fragment Length Polymorphism (RFLP) can be used for screening genetic stability of tissue cultured plants, the method involve use of expensive enzymes, radioactive labelling and extensive care, therefore appears unsuitable. Among various methods used
for such determination of genetic fidelity in that Random Amplified Polymorphic DNA (RAPD) is the simplest, cheapest, quick, requires only small amounts of DNA detects rare single base mutations and deletions at the level of primer target and within the amplified fragment and appears to be a useful tool for the analysis of genetic fidelity of in-vitro propagated plants. RAPD analysis used successfully for genetic analysis of in vitro-raised plant materials (Isabel et al., 1993; Rani et al., 1995; Taylor et al., 1995; Munthali et al., 1996; Rani and Raina, 1998; Al-Zahim et al., 1999).

There are reports available on genetic fidelity of micropropagated plants of other species. Sharma et al., (2009) assessed genetic fidelity of in-vitro raised 45 plants of gerbera (Gerbera amesonii Bolus) cultured from three different explants i.e. capitulum, leaf and shoot tip. For investigation 32 ISSR markers was assessed for genetic stability from that 15 markers observed clear, distinct and scorable bands with an average of 5.47 bands per marker. The elite clones form capitulum and shoot tip explants did not observed genetic variation whereas, one of the leaf derived micropropagated clones revealed some degree of variation.

Modgil et al., (2005) reported that RAPD marker detected the genetic similarities and dissimilarities in micropropagated clones of 10 micropropagated clones revive through axillary buds of clonal apple (Malus pumila Mill.). RAPD markers results revealed that 99 were monomorphic and 30 were showed polymorphic with 23.2% polymorphism out of 129 scorable fragments.

Sahoo et al., (2010) investigated genetic fidelity and essential aromatic oil content of rapid regenerated clones of Patchouli, Pogostemon cablin (Blanco) using RAPD marker and Gas Chromatogram. Both markers showed same banding pattern
using. Results ensured that the efficiency of the protocol standardized for the production of this industrially important aromatic plant.

Mathur et al., (2008) experimented biological hardening and genetic fidelity testing of micro-cloned progeny of *chlorophytum borivilianum*. The genetic fidelity testing of micro-cloned, bio hardened progeny based on a RAPD analysis using 40 random decamers DNA primers indicated a strong uniformity in relation to the parent genotype.

Martins et al., 2004 studied genetic stability of micropropagated almond plantlets using RAPD and ISSR markers. Total 22 plantlets was analyzed using 64 RAPD and ISSR primers, 326 distinct and reproducible bands pattern was recorded and all bands found monomorphic exhibiting homogenous patterns for the plant tested.

Joshi et al., (2007) validated the genetic fidelity of *Swertia chirayita* micropropagated clones. Sixteen ISSR markers produced 102 amplicons and homogenous amplification profiles were observed in all micropropagated clones, concluded the safest mode for multiplication of true to type plants. Similarly, Latoo et al., (2006) standardized an organogenetic *in-vitro* protocol for *Chlorophytum arundinaceum* using shoot tip explants. Genetic fidelity RAPD markers, result showed no genomic variation in regenerated plants through shoot bud differentiation and ensures the effectiveness of the protocol developed for the production and conservation of medicinal herb. Gagliardi et al., (2007) concurred with above reports that RAPD markers showed the genetic stability of micropropagated clones of *Archisretusa*. Total 90 bands were recorded from RAPD and 372 from AFLP marker. All amplified homogenous bands showed by both the techniques were monomorphic and results signified that recovered shoots are genetically stable.

Goto et al., (1998) determined genetic stability of more than 10 years micropropagated shoots of Japanese black pine (Pinus thunbergii Parl.). Total 36 shoots consisting three morphotypes (short, medium and long needles) were randomly chosen from 4000 micropropagated clones. Out of 126 primers, 30 primers gave 134 clear scorable bands. Total 4824 bands were obtained from this experiment, no aberration was observed in RAPD banding pattern between the experienced shoots.

Rout et al., (1997) evaluated the genetic stability of micropropagated plants of Zingiber officinale (V3S18) using RAPD marker. Fifteen arbitrary decamers were used to assess the genetic fidelity. All RAPD banding profile were monomorphic and similar to those of field grown plants, no aberration were observed within the micropropagated clones.

Nayak et al., (2007) assessed the genetic stability of micropropagated replica of Cucurma longa L. by Cytophotmetry and RAPD of 26 months old culture plants. Both assessed methods showed uniformity among micropropagated clones of C.longa. This result induces the production of disease free planting material of turmeric for commercial utilization.
Tanwar et al., (2008) used RAPD markers for determination of somaclonal variation in micro-propagated plants of Sugarcane varieties (Co94012) and VSI434. The RAPD banding patterns of both varieties were monomorphic and similar banding pattern was detected in all bands. Studies indicated that somaclonal derived clones showed high genetic fidelity with no genetic variation among the plantlets produced in vitro. The results revealed that RAPD analysis can be efficiently to assess the genetic purity of sugarcane clones derived from tissue culture.

2.5 Antioxidant activity of Zingiber officinale and other medicinal plants

A large amount of work is available particularly on evaluation of antimicrobial and antioxidant properties of various plants. Antioxidants have been reported to prevent from oxidative damage during generation of free radicals in an integral part of normal metabolism Shahidi et al., (1992). The potential reactive oxygen species (ROS) is generated in vivo, through various physiological and biochemical processes such as mitochondrial respiration, activation of phagocytes, enzymatic oxidation, UV and ionizing radiations. Reactive oxygen species attack various biomolecules like protein, lipids, DNA etc., and inflict damage breaking down various cellular processes (Farber, 1994). Although, natural antioxidant defences are generally adequate to neutralize the radicals, their concentration and rate of generation decreases with age, as well as under inflammation. Under these conditions, exogenous addition of antioxidant becomes necessary to prevent the oxidative stress that has been implicated for various pathological condition and degenerative diseases. As a result antioxidants have assumed great interest in medical science (Buyukokuroglu et al., 2001).

Ginger has a high content of antioxidants activity and has been grouped as one of the spices with good antioxidant rating (Chipault et al., 1952). Sethi and
Agarwal (1957) observed that dried ginger has low antioxidant properties and increase in concentration of crude gingerol increases the antioxidant activity but the gingerol constituent at 165°C for 30 min indicated the withholding of the antioxidant activity only to 10 percent.

Fugio et al., (1969) investigated the antioxidant activity of the chemical constituents of many spices and observed that the shogaol and zingiberene found in ginger revealed strong antioxidant activities. The free radical scavenger activity of ginger is fully dependent on the side chain structures and substitution patterns on the benzene ring.

Nakatani and Kikuzaki (2002) observed zingerone, 6-gingerol, and 6-shogaol found moderate antioxidant activity and antioxidant activity will decreases with the increasing chain length. Tsushida et al., (1994) observed that antioxidant activity is mostly exerted by gingerol and hexahydro curcumen. Due to this it is a free radical scavenger (Lee and Ahn, 1985).

Ahmed et al., (2001) investigation diet containing ginger showed a more protective effect against the malathion-induced oxidative damage exhibiting the antioxidant activity and incorporation of salt and ginger extract to precooked lean beef retarded rancidity during storage, increased the tenderness, and extended shelf life (Kim and Lee, 1995).

Stoyanova et al., (2006) studied the antioxidant effect and total phenols of Zingiber officinale from Vietnam. The carbon dioxide extract of ginger was analyzed with DPPH (2, 2- diphenyl-1-picryl hydrazyl), lipid oxidation and pro-oxidant activity with regards to hydroxyl radicals at body temperature (37°C). The total phenol obtained
from the alcohol extract was 870.1 mg/g from dry extract, DPPH scavenging activity found/reached 90.1% exceeds that of butylated hydroxytoluene (BHT), IC50 concentration was found for inhibition of DPPH was 0.64 µg/ml. The antioxidant activity in a linoleic acid/water was highest at 37°C–73.2%, and 71.6% was found when the formation of conjugated dienes was inhibited. The ginger extract inhibited the hydroxyl radicals 79.6% at 37ºC and 74.8% at 80ºC190 and 2.78 µg/ml.

Jaafar et al., (2010) reported the antioxidant activities, total phenol and flavonoids content in two varieties of Malaysian young ginger (Zingiber officinale) Halia-Bentong and Halia-Bara. The antioxidant activities of methanol extract from leaves, stem, and rhizomes were analyzed to compare validates the medicinal potential of the young ginger. The antioxidant activity and total phenol content of leaves reported higher than stem and rhizome. Analysis of FRAP activity found higher in rhizome than leaves and stem. All analysis report concludes that Halia Bara variety contain higher antioxidant activities than Halia Bentong.

Bhattacharya et al., (2009) studied the antioxidant activities of different solvent fractions of ginger (Zingiber officinale Rosc). Out of 34 solvent fractions, 10 fractions found free radical scavenging activity ranging from 5.88 % to 80 %. Diethyl ether and ethyl acetate (1:1) showed maximum inhibition percent (80 %) of antiradical activity and liver protective, solvent chloroform fraction observed maximum hydroxyl radical activity, the maximum nitric oxide (NO) generation activity found at benzene fraction (27.27 %) and concluded that ginger flavonoids have some contributory roles in scavenging free radical activity.

Mishra et al., (2011) studied correlation of phytochemical characteristics and antioxidative properties have been studied in hot and cold extracts of Canna edulis
rhizome. Total phenol and flavonoid in hot extracts (42.71 mg GAE /g and 21.92 mg QE /g) and in cold extracts (33.7 mg GAE /g and 15.12 mg QE /g). IC50 value of DPPH and H2O2 and electron donation ability (EDA) observed higher in hot extracts than cold extracts. Analysis observed that hot extracts of C. edulis exerts more effective antioxidant property as compared to cold extract.

Lim et al., (2008) screened 26 ginger species belonging to nine genera. Analysis was performed with total phenolics content (TPC) and ascorbic acid equivalent antioxidant capacity (AEAC). Out of 26 species, leaves of Etlingera species found the highest TPC and AEAC. Ferrous ion chelating (FIC) abilities of leaves and rhizomes of eight species were compared and six of the eight species showed higher values in leaves than rhizome. Analyzed value of FIC showed Alpinia galanga leaves which were more than 20 times higher than that of rhizomes. From five species of Etlingera, leaves of E. elatior showed strongest tryrosinase inhibition activity than E. flugens and E. maingayi and this three also found high antioxidant activity and antibacterial properties.

Goyal et al., (2010) studied the antioxidant activity with spectrophotometrically analyze the ability of the plant extracts to scavenging activity of DPPH, TPC and flavonoid content from methanolic leaf extract of Bambusa vulgaris. Analyzed results showed the presence of carbohydrates, reducing sugars, flavonoids, steroids, sapionins, alkaloids, tannins, anthraquinones and glycosides. The antioxidant activity of the investigated extract found a scavenging ability of hydroxyl peroxide radicals (421.74 ± 25.61 mg/ml) and DPPH radical activity (95%). The TPC and flavonoid content was measured (GAE22.69 ± 0.084 mg/g of dry extract) and (Quercetin 159.80 ± 0.047 mg/g of dry extract) indicated that these compounds contribute to the antioxidative activity.
Fagbenro and Jauncey (1994) observed that during study of the chemical and nutritional quality of fermented fish silage containing potato extracts, formalin, and ginger extracts, it was observed that ginger extract showed to be effective as an antioxidant in fermented tilapia silage (*Oreochromis niloticus*). Nakatani and Kikuzaki (2002) observed zingerone, 6-gingerol and 6-shogaol found moderate antioxidant activity and antioxidant activity will decreases with the increasing chain length.

Nishimura (1995) investigated the volatile compounds for the aroma of fresh rhizomes of ginger and the compounds with high dilution factor were linalool, geraniol, geranial, neral, isobor-neol, borneol, 18-cineol, 2-pinenol, geranyl acetate, 2-octenal, 2-decenal, and 2-dodecenal. The pungent principle of ginger, 6-gingerol, has been reported to be a potential antioxidant among 10 phenolics compounds separated by TLC.

2.6 Antimicrobial activity

Medicinal plants have been used since antiquity to treat common diseases because they contain bioactive constituent for remedial value to treat various diseases. This plant-based, traditional medicine system continues to play an essential role in health care, with about 80% of the world’s inhabitants relying mainly on traditional medicines for their primary health care (Owolabi *et al.*, 2007). According to World Health Organization, medicinal plants would be the best source to obtain a variety of drugs. Therefore, such plants should be investigated to better understand their properties, safety and efficacy (Nascimento *et al.*, 2000). Many researchers reported that some medicinal plants contains many components such as peptides, unsaturated long chain aldehydes, alkaloidal constituents, some essential oils, phenols and water, ethanol, chloroform, methanol and butanol soluble compounds. These plants have
potential in therapeutic application against human pathogens, including bacteria, fungi or virus (Elastal et al., 2005). Some application were used by others researchers are the use of bearberry (*Arctostaphylos uva-ursi*) and cranberry juice (*Vaccinium macrocarpon*) to treat urinary tract infections is reported in different manuals of phytotherapy, while species such as lemon balm (*Melissa officinalis*), garlic (*Allium sativum*) and tea tree (*Melaleuca alternifolia*) are described as broad spectrum antimicrobial agents (Rio and Recio, 2005).

Nguanpuag et al., (2011) investigated antimicrobial activity both in-vitro and in-vivo using ginger oil extracted by hydro distillation and solvent extraction methods. The bioactive compounds obtained from hydro distillation were camphene, 1, 8-cineol and a-pinene and from solvent extraction were ß-phellandrene and 1, 8-cineol. Both extraction oils inhibited *Bacillus subtilis, Bacillus nutto, Pseudomonas aerogenosa, Rhodoturola* sp., *Samonella newport DMST 15675, Samonella enteritidis DMST 15676* and *Fusarium* sp. No inhibition was found on *Escherichia coli, Campylobactor coli NTCT 11353* and *Campylobacter jejuni ATCC 33291*. In vivo growth of microorganism was suppressed significantly in shredded green papaya with 5 and 10 5 and 10 µL. Major volatile ginger oils detected were a-pinene, camphene, ß-phellandrene and 1, 8-cineolin shredded package papaya and observed that ginger oils can be used for reducing population of microorganism in shredded papaya and other fresh produce processed products.

Adeshina et al., (2011) reported antibacterial activity of fresh red and white *Allium cepa* (Onion) and *Zingiber officinale* (Ginger) juice against multidrug resistant bacteria viz *Pseudomonas aeruginosa, Staphylococcus aureus, Escherichia coli and*
Salmonella typhi isolated from salad was using agar well diffusion and agar dilution methods.

Ayse et al., (2008) reported an antimicrobial and cytotoxic activities of chloroform and ethanol extracts of Zingiber officinale against Klebsiella pneumoniae, Salmonella thyphimurium, Bacillus cereus, Enterococcus fecalis and Staphylococcus aureus and cytotoxic effects on human cervical cancer (HeLa) and mouse fibroblast (L929) cell lines. Schnitzler et al., (2001) investigated acyclovir resistant clinical isolates of herpes simplex virus type 1 (HSV-1) were analyzed in-vitro for their susceptibilities to essential oils of ginger, thyme, hyssop, and sandalwood. Similarly, Patel et al., (2011) observed that aqueous ginger extracts which was used to check antimicrobial activity found to be efficient.

Anjan et al., (2012) experimented recorded that in-vitro antimicrobial potential of 10 % ginger extract against Streptococcus mutans (S.mutans), Candida albicans (C. albicans) and Enterococcus faecalis (E.faecalis) are mostly causation of oral infections microorganism, during study results revealed that 10% ethanolic ginger extract possess high antimicrobial potential.

Indu and Nirmala (2010) reported that Zingiberene was the main chemicals constituents for an antimicrobial activity of fresh and dry ginger against Bacillus subtilis, Pseudomonas aeruginosa, Candida albicans, Trichoderma spp, Aspergillus niger, Pencillium spp. and Saccharomyces cescerevisiae microorganism. Furthermore, Tagoe et al., (2011) compared the antifungal properties of Onion, Ginger and Garlic against Aspergillus flavans, Aspergillus niger and Cladosporium herbarum using pour plate technique in PDA medium and results found that ginger showed highest antifungal activity on all test fungi with a mean diameter 1.40 cm followed by garlic
and onion (1.80 cm). Likewise, Ekwenye and Elegalam, (2005) recorded aqueous extracts found high antimicrobial activity against *E. coli* and *S. typhi* microorganism.

Nader *et al.*, (2009) reported antimicrobial activity of ginger extracts of cold-water, hot-water and ethanolic and essential oil against pathogenic bacteria *Escherichia coli*, *Salmonella* sp, *Klebsiella* sp, *Serratia marcescens*, *Vibrio cholerae*, *Staphylococcus aureus*, *Streptococcus* sp. was examined disc diffusion method, results showed that ginger extracts were more effective on gram positive bacteria than gram negative bacteria and ethanolic extract observed highest antibacterial activity (11 to 28mm) than other extracts. Phytochemical analysis of ethanolic extracts revealed the presence of glycosides, terpenoids, flavonoids and phenolic compounds.

Kumar *et al.*, (2011) studied antibacterial potential of natural food preservatives against *Staphylococcus aureus* isolated from different food samples. Six different spices were used for study such as *Cucurma longa* (Turmeric), *Zingiber officinale* (ginger), *Piper nigrum* (black pepper), *Trigonella foenum graecum* (methi), *Syzygium aromaticum* (clove) and *Ferula assafoetida* (hinge). All sample showed significant antibacterial activity against *S.aureus* isolated from food samples. It was observed that, ethanolic extracts of all the spices showed highest inhibited effect against *S.aureus* followed by methanolic and aqueous extracts. Pedgee *et al.*, (2012) recorded chloroform extracts of both sample of turmeric and ginger rhizome found potent antimicrobial activity against *Pseudomonas* and *E. coli* than distil water extracts.

However, Premlata *et al.*, (2011) recorded chloroform and water extracts of *Withania somnifera* (RUBL-20668) and *Cenchrus setigerus* (CAZRI-76) showed high antimicrobial activity against *Bacillus subtilis*, *Pseudomonas aeruginosa*, *Enterobacter*
aerogens and Aspergillus flavus. Water extract of stems of C. setigerus (IZ-21.83 ± 0.24 mm, ai- 0.780) and chloroform extract of calyx of W. somnifera (iZ-16.17 ± 0.24 mm, ai 1.078) showed highest activity against B. subtilis. Auta et al., (2011) reported that ethanol extract of Zingiber officinale at the concentration of 20 mg/ml showed high antimicrobial activity against E.coli and Pseudomonas aeruginosa pathogens for gastrointestinal tract. Similar results was concurred by Chand (2013) found antibacterial effect of garlic and ginger against bacteria Escherichia coli, Salmonella typhi, Staphylococcus aureus and Bacillus cereus.

2.7 Molecular studies

Morphological markers such as leaf area, dry matter production, length and weight of secondary rhizomes, tiller number and leaf number differed significantly (Yadav, 1999). Biochemical markers such as total gingerol, shogaol, polyphenol content etc. are used to identify the superior ginger plant (Ravindran et al., (1994); Manmohan das et al., (2000). However, ginger breeders are often unable to use these markers effectively because they are greatly influenced by environmental factors and show a continuous variation with a high degree of plasticity. A lucid knowledge of genetic variability is essential for formulating a meaningful breeding strategy. Nybe and Nair (1982) reported that morphological characters are not reliable to classify the types, although some of the types can be distinguished to a certain extent from rhizome characters. Hence, to overcome these problems, research has shifted to using more sensitive DNA marker technology. Molecular markers very efficiently enhance morphological, cytological, and biochemical characters in germplasm characterization, varietal identification, clonal fidelity testing, assessment of genetic diversity, validation

### 2.7.1 Morphological marker

Generally ginger has been classified into different species or varieties by morphological characters. Most of the vegetatively propagated crop species the amount of genetic variations will be limited unless samples are drawn from distinctly different agro-ecological situations (Ravindran et al., 1994). Minute variability exists among the genotypes that are grown in the same area; however, good variability has been reported among cultivars that came from widely divergent areas (Ravindran et al., 1994). Most of studies were carried out in morphological characters, rhizome yield and quality parameter.

Ravindran et al., 1994 reported characterization of 100 accessions of ginger germplasm based on morphological, yield, and quality parameters. They observed moderate variability for many yield and quality traits, reported tiller number per plant had the highest variability, followed by rhizome yield/plant and in biochemical trait, the shogaol content traced the highest variability, followed by crude fiber and oleoresin.

Yadav (1999) reported a high genotypic coefficient of variation for length and weight of secondary rhizomes, weight of primary rhizomes, number of secondary and primary rhizomes, and rhizome yield/plant. Furthermore, Sasikumar et al., (1992) studied 100 accessions of ginger germplasm for variability, correlation, and path analysis. They found that rhizome yield was positively correlated with plant height, tiller number, leaf number, leaf length and leaf width.
Nybe and Nair (1982) studied the morphological characters of ginger accessions and reported that phenotypic studies are not reliable to classify the types, although some of the types can be distinguished to a certain extent from rhizome characters. All the morphological characters were found to vary among types except for breadth of leaf, leaf area index, and number of primary fingers. Quality parameters such as dry recovery and oleoresin and fiber contents are known to vary with the soil type, cultural conditions, and climate (Ravindran et al., 1994).

2.7.2 Biochemical marker

Biochemical markers were widely used for characterization of different plant germplasm (Das et al., 2002). The bioactive constituent of gingers plays a very important role in characterization of different ginger germplasm. Oleoresin of ginger is the total extract of ginger containing all the bioactive principles as well as the pungent constituents. The oleoresin contains two important compounds gingerol and shogaol that contribute to the ginger pungency (Ravindran et al., 2005). The quality and characterization of ginger was classified by amount of gingerol and shogaol present in the extract of ginger.

Zachariah et al., (1993) also classified 86 ginger accessions into high, medium, and low quality types of oleoresin based on the relative contents of the quality components. There are many ginger cultivars with high oleoresin, a few them, such as Rio de Janeiro, Ernad Chernad, Wynad, Kunnamangalam, and Meppayyur, also had high gingerol content. The inter character association showed a positive correlation with oleoresin, gingerol, and shogoal.
Shamina et al., (1997) investigated the variability in total free amino acids, proteins, total phenols, and isozymes, using 25 cultivars. Moderate variations were recorded for total free amino acids, proteins, and total phenols. Isozyme variability for polyphenol oxidase, peroxidase, and SOD was reported to below, indicating only a low level of polymorphism. Jiang et al., (2005) experimented metabolic profiling analysis from different origin. They observed that no qualitative difference among major volatile bioactive compounds whereas low variations was observed in non-volatile composition, particularly regarding the content of 6, 8 and 10 gingerols. Singh et al., (1999) grouped 18 cultivars into three clusters under Nagaland conditions based on $D^2$ analysis. The major forces influencing divergences of cultivars were rhizome yield per plant and oleoresin and fiber contents.

2.7.3 Molecular Marker

Williams et al., (1990) described a novel type of genetic marker based on DNA amplification, which does not require prior information of target DNA sequences. These markers called RAPD (Random Amplified Polymorphic DNA) markers are generated by the amplification of random DNA segments with single primers of arbitrary chosen primers. Use of random amplified polymorphic DNA (RAPD) markers, detected by PCR amplification of small inverted repeats scattered throughout the genome, adds a new technology of DNA fingerprinting to the molecular analysis of relatedness between genotypes. The PCR based RAPD technique (Williams et al., 1990) is an attractive complement to conventional DNA fingerprinting. RAPD analysis is conceptually simple. Nano gram amounts of total genomic DNA are subjected to PCR using short synthetic oligonucleotide of random sequence. The amplification protocol differs from the standard PCR conditions (Erlich, 1989) in that only a single random
oligonucleotide primer is employed and no prior knowledge of the genome subjected to analysis is required. When the primer is short (e.g. 10 mer), there is a high probability that the genome contains several priming sites close to one another that are in an inverted orientation. The technique essentially scans a genome for these small inverted repeats and amplifies intervening DNA segments of variable length. The profile of amplification products depends on the template-primer combination and is reproducible for any given combination. The amplification products are resolved on agarose gels and polymorphisms serve as dominant genetic markers, which are inherited in a Mendelian fashion (Williams et al., 1990; Carlson et al., 1991; Welsh, Peterson and McClelland, 1991). Amplification of non-nuclear RAPD markers is negligible because of the relatively small non-nuclear genome sizes. Since discovered, random amplified polymorphic DNA (RAPD) assay (Williams et al, 1990) is being used for a number of areas in plant taxonomy. At the present it is the most preferred DNA markers due to greater speed, easy-to-perform and non-requirement of radioactive materials etc. In ginger and other species of Zingiber, a considerable amount of work has been carried out which are summarized below.

Rout et al., (2007) analyzed genetic fingerprinting among eight varieties of Zingiber officinale using RAPD markers, the investigation showed the distant variation within the varieties, similar result was obtained by (Harisaranraj et al., 2009) within the eight varieties of ginger of Orissa.

Pattanayak et al., (2010) assayed forty nine ginger clones cultivated in North East India using RAPD markers. The high polymorphism detected in a cultivated species in this study exhibit aptness the resolving power of the RAPD markers selected for genetic diversity investigation. Similarly, Wahyuni et al., (2003) investigated
genetic diversity of morphological distinct (big and small) Indonesian gingers using AFLP markers and analysis of genetic variation reported that there was no clear genetic variation between small and big form gingers. Results also showed there was higher genetic diversity in small size ginger than big size variant.

Nayak et al., (2005) characterized 16 elite cultivars of gingers using cytological and RAPD markers, and assayed result reported that significant genetic variations were detected in gingers variant.

Jaing et al., (2006) studied genetic diversity using phylogenetic analysis and metabolic profiling among and within ginger species and result found that gingers variant from different geographical origins were indistinguishable.

In a study of Jatoi et al., (2008) genetic diversity in gingers and relationships among the Zingiber species using rice ISSR markers as RAPD markers. They observed significant allelic diversity in ginger from Myanmar. Result showed higher genetic variability were observed in gingers collected from farmers’ fields in comparison with gene bank accession and market collection. Similar study was carried out by Watanabe et al., (2006) using rice SSR marker as RAPD marker for genetic diversity analysis in Zingiberaceae. They reported that high variation was found among ginger, turmeric and galangal species.

Kizhakkayil and Sasikumar (2010) investigated Indian gingers diversity using RAPD and ISSR markers. They found gingers diversity are geographical bias and significant similarity among the clones. Subramanian et al., (2007) successfully identified using RAPD markers for characterization of diseases resistant to Fusarium oxysporium f.sp Zingiberi and susceptible varieties of gingers.
2.7.4 Cytological marker

Studying the structural properties and spatial organization of chromosomes is important for the understanding and evaluation of the regulation of gene expression, DNA replication and repair, and recombination. Different facets of chromosomal research are gaining significance for the analysis of genomes in plant taxonomy (Bennett, 1987; Mowforth and Grome, 1989; Ceccarelli et al., 1992; Das et al., 1995; Roser et al., 1997).

Cytological markers of the genus *Zingiber* were elaborately studied in the early 1920s with many interesting features. The somatic chromosome number of *Zingiber officinale* 2n=22 was first studied by Sugiura 1928 followed by Sharma and Bhattacharyya, (1959); Ramachandran, (1969); Rai et al., (1997); Kihara et al., (1931) and 2n=66 Bisson et al., (1968) were reported. The nuclear DNA content varies considerably not only among species, but also among and within populations of species Bennett and Leitch (1995). Das et al., (1998) examined karyotype and estimated 4C DNA in ginger. They found significant differences of 4C DNA between the cultivars.

2.7.5 Morphological study

Knowing morphological characters of horticultural crops is very necessary for formulating a fruitful breeding strategy. The most easily obtained assessment of genetic variation is that of measuring morphological or phenotypic variation. The sharing of phenotypic characters is interpreted as an indication of relatedness. Morphological characters are however, often influenced by environmental conditions Jasienski, (1997); Kercher and Sytsma (2000), which in turn may influence the estimation of genetic
variation and relatedness. Morphological variation studies were carried out by many researchers on ginger and other plants, few are of them are as follows.

Mohanty and Sarma (1979) reported on morphological studies of ginger that expected genetic advance and heritability estimates were high for the number of secondary rhizome and total root weight.

Mohanty et al., (1981) observed significantly varietal differences for all the characters except for the number of tillers per plant and number of leaves per plant. Nybe and Nair (1982) suggested that morphological characters are not reliable to classify the types, although some of the types can be distinguished to a certain extent from rhizome characters. All the morphological characters were found to vary among types except for breadth of leaf, leaf area index, and number of primary fingers.

Rattan et al., (1988) reported on ginger that plant height was positively and significantly correlated with number of leaves, leaf length, rhizome length, rhizome breadth, and yield per plot.

Pandey and Dobhal, (1993) observed a wide range of variability for most of the characters studied by them. Rhizome yield per plant was positively associated with plant height, number of fingers per plant, weight of fingers, and primary rhizome.

Ravindran et al., (1994) characterized 100 accessions of ginger germplasm based on morphological, yield, and quality parameters. Moderate variability was observed for many yield and quality traits. Tiller number per plant had the highest variability, followed by rhizome yield/plant.

At IISR, Sasikumar et al., (1992) studied 100 accessions of ginger germplasm for variability, correlation, and path analysis. They found that rhizome yield was
positively correlated with plant height, tiller and leaf number, and leaf length and width. Manmohandas et al., (2000) found that all the cultivars differed significantly in tiller number and leaf number.

Ravindran et al., (2005) found little variability among the ginger genotypes that are grown in the same area and high variability was estimated among cultivars that came from widely divergent areas. Terratas et al., (2007) observed that stem trait was identification characteristics of 21 pithaya genotypes. Yadav, (1999) found high genotypic variation for length and weight of secondary rhizomes, weight of primary rhizomes, number of secondary and primary rhizomes, and rhizome yield/plant of gingers. Singh, (2001) observed during morphological studies of ginger, plant height, number of tiller per plant and leaf length had a maximum direct effect on rhizome yield.

2.8 Remote sensing and GIS in agriculture

Food Security is a big bang question for all developed or developing countries. Agricultural sustainability has the uppermost priority in all countries. Remote Sensing and GIS technology are gaining importance as useful tools in sustainable agricultural management and development. The sustainable agriculture is to maintain the natural land resources with crop requirement towards achievement of sustained productivity over a long period Lal and Pierce, (1991). The key for providing food security to all people of the world without affecting the agro ecological balance lies in the adaptation of new research tools, particularly from aerospace Remote Sensing, and combining them with conventional as well as frontier technologies like Geographic Information Systems (GIS).
In 1971 first time remote sensing technology was used for large area crop survey in USA under Corn Blight Watch Experiment (CBWE). In 1973 Crop Identification Technology Assessment for Remote sensing (CITARS) was started for to quantify the Crop Identification Performance (CIP). From 1974 to 1977 (NASA) major wheat growing areas of the world was forecast under Large Area Crop Inventory Experiment (LACIE). The Monitoring Agriculture through Remote Sensing (MARS) project was developed for crop growth and monitoring system (CGMS), which helps into crop simulation models, agro meteorological models, and real time data for crop predicting and assessment. Lepoutre (1991) used remote sensing satellite imagery for monitoring crop production, for estimating losses due to drought in France in 1998 to 1991 and to monitor potential crop yields throughout Europe.

In context of Indian scenario pioneer application of remote sensing and GIS technology was carried out by Dakshinamurti et al., (1971) experimented on coconut root wilt disease using colour infrared aerial photography. Later mega project of agriculture was experimented about agricultural resources under Agriculture Resources Inventory and Survey Experiment (ARISE) in Anantpur (1974-75) and Patiala (1975-77). Subsequently identification and classification of paddy and sugarcane crops in Madhya Pradesh (1975-77) were experimented.

Systematic crop productions are of vital importance to country such as India, where the agricultural production is highly susceptible to the vagaries of monsoon. Acreage estimation using RS technology was first demonstrated by Mc Donald (1984) and Renondo et al., (1985) in various parts of world. In India Dadhwal and Parihar (1985) estimated wheat acreage estimation of Karnal district using Land sat MSS digital data and supervised classification. Rai et al., (2004) experimented on land use statistics
through integrated modelling using GIS. Mishra et al., (2005), developed an integrated approach for estimation of crop acreage using remote sensing data, GIS and field survey of hilly region.


Utpala et al., (2006) analyzed the environmentally most site suitable for ginger cultivation in India using Eco-crop model of DIVA-GIS. Three parameters were studied i.e. growing periods, temperature and rainfall. Study found that most suitable areas were Orissa, West Bengal, Kerala, Western Ghat region of Karnataka and Maharashtra for ginger cultivation in India. In north-eastern states Assam, Mizoram, Tripura, Western Ghat of Meghalaya showed most site suitability for ginger cultivation.

Utpala et al., (2007) studied the site suitability location of turmeric cultivation in India, researcher found that site suitability is an important factor to determine the productivity of the crop, determine suitable areas which are useful to have the greatest success for growing a particular crop in a region. Land suitable analysis was carried out in such a way that local needs and condition. Experiment found that most suitable were Andra Pradhesh for turmeric cultivation leaving small patch in central and western part
of Andra Pradesh, Assam due to high suitable environmentally for turmeric cultivation, Bihar, Kerala is most suitable for turmeric cultivation.

Kris Sunato, (2011) analyzed the site suitability for cultivation of ginger (*Zingiber officinale* Roscoe) in Indonesia, using geographical data, analysis tools and overlay technique on Geographic Information System (GIS). Analyzed data were produced and displayed in maps, graphs, images and tables. The study recorded about 11.36% of total area comprising about 33874 Ha found high suitable for ginger cultivation.

Utpala *et al.*, (2008) reports on identification of suitable areas and effect of climate change on ginger cultivation in India. Reports suggest that increase in area is not always directly proportional to the increase in production. Records of data for thirty years on area and production were compared with Eco-crop suitability model, which showed that suitability has direct impact on production. The most suitable areas are Orissa, West Bengal, Mizoram and Kerala. North western states like Gujarat, Rajasthan, Uttar Pradesh and Madhya Pradesh are marginally suitable or unsuitable. The north-eastern states of Assam, Mizoram, Tripura, and Meghalaya showed most site suitability for ginger cultivation.

Analysis of future prediction of Eco-crop model showed that the temperature increases by 1.5 to 2°C will reduce drastically from high suitability to marginally suitable, showed the effect of climate change.