Series of Crop Specific Biology Documents

# Biology of Zea mays (Maize)





Ministry of Environment and Forests Government of India Department of Biotechnology Ministry of Science & Technology Government of India Series of Crop Specific Biology Documents

# **Biology of** Zea mays (Maize)



Ministry of Environment and Forests Government of India Government of India

Department of Biotechnology Ministry of Science & Technology

#### Biology of Zea mays (Maize)

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#### **Consultation process:**

Circulated to concerned state agricultural universities, research institutions and industry and placed at websites viz. http://www.igmoris.nic.in and http://dbtbiosafety.nic.in. The inputs and comments provided by the following are gratefully acknowledged.

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#### FOREWORD

India is one of the leading countries having an active agricultural biotechnology research and development programmes in diverse crops including cereals, vegetables, oilseeds etc. and traits such as insect, disease, and virus resistance, herbicide tolerance, stress tolerance etc. The successful development and commercialization of biotech derived crops also referred as genetically engineered (GE) or genetically modified (GM) crops requires a science based regulatory process to address the concerns arising out of genetic manipulation to human health and environment.

The Department of Biotechnology (DBT), as one of the implementing agencies for biosafety regulations in India has been providing science based support for evaluating the GM crops by preparing various guidance documents and disseminating information through websites. In continuation with the above efforts, a need was felt to prepare crop specific biology documents to provide relevant baseline information about various crops in a readily accessible format.

I am pleased to note that Dr. K.K. Tripathi, Advisor, DBT and Member Secretary, RCGM has put in considerable efforts in putting together a series of five crops specific biology documents on cotton, brinjal, okra, maize and rice, in association with the Ministry of Environment and Forests (MoEF). The biology documents have been put through a consultative process with various stakeholder viz. agriculture research institutions, state agricultural universities, industry etc. The views have been taken by circulating the documents to relevant institutions as well as by placing them on websites. The documents have also been reviewed by the members of RCGM and GEAC. Biotech Consortium India Limited (BCIL) provided support in compiling the baseline information, as well as the consultative process.

I believe that these crop specific biology documents would be of immense value for both the developers in planning the safety assessment of their products as also the regulators for evaluating the data submitted to them. Scientific developments being advancing at a rapid rate, I hope that these biology documents would be continuously updated from time to time.

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#### PREFACE

Genetically engineered (GE) crops are regulated products in view of various concerns for human and animal health and environment. Extensive evaluation and regulatory approval process take place before any GE crop is introduced for cultivation. The approval for release of a GE crop is given by the Genetic Engineering Approval Committee (GEAC) functioning in the Ministry of Environment and Forests (MoEF) as per "Rules for the manufacture, use, import, export & storage of hazardous microorganisms, genetically engineered organisms or cells, 1989" notified under the Environment (Protection) Act, 1986.

So far, Bt cotton, is the only GE crop approved for commercial cultivation in India. There are several crops under various stages of research, development and field trials. The present set of crop specific biology documents has been prepared jointly by MoEF and the Department of Biotechnology (DBT) to provide scientific baseline information used for safety assessment of GE crops. These biology documents have sections on taxonomy, economic importance, centre of origin, growth and development (vegetative and reproduction biology), ecological interactions, distribution pattern in India etc.

I wish to put on record my appreciation of the sincere efforts put in by Dr. Ranjini Warrier, Director, MoEF who has worked closely with DBT and other stakeholders for this initiative and the consultative approach adopted in finalizing these documents. I also acknowledge the support of members of both GEAC and RCGM for their useful inputs during the review process. The inputs and support provided by Dr O.P. Govila, Former Professor of Genetics, Indian Agricultural Research Institute (IARI) and Dr. Vibha Ahuja, General Manager, Biotech Consortium India Limited (BCIL) has also been extremely valuable.

I am sure that these crop specific biology documents would serve as practical tools for researchers, regulators and industry.

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#### PROLOGUE

Modern biotechnology like any new technology has its associated benefits and risks. Accordingly products of modern biotechnology like biopharma, genetically engineered (GE) crops etc. are regulated for ensuring safety to human and animal health and environment. In case of GE crops, scientific assessments ensure food safety and environmental safety, an integral part of approval process. The whole process of safety assessment is based upon comparison between genetically engineered crop and its unmodified counterpart and thus requires a broad understanding and knowledge of various features of the crop plants. This familiarity with the crops allows both the developers and regulators to draw on previous knowledge and experience to ensure safety of the GE crops.

Keeping in view the above, the Department of Biotechnology (DBT) and the Ministry of Environment and Forests initiated the preparation of a "Series of Crop Specific Biology Documents" to provide information directly relevant to safety assessment in a readily accessible format. The objective of these documents is to make available the information about biology of the crops to applicants as information in applications to regulatory authorities; to regulators as a guide and reference source in their regulatory reviews; and for information sharing, research reference and public information. To start with, crop specific documents for five crops viz. cotton, brinjal, maize, okra and rice have been prepared. In addition to the scientific literature and references, the documents have also taken into account the information available in Consensus documents have been finalized through a consultative process with the concerned research institutions, state agricultural universities and subject experts. The documents were also placed on DBT's biosafety website for public review.

It is proposed to continue this exercise for more crops such as mustard, potato, tomato etc. that are under development. The support from various technology developers from both public and private sector, state agricultural universities, agricultural research institutions and other subject experts in providing information as well as reviewing these documents is acknowledged. We also appreciate the assistance provided by Dr. Vibha Ahuja, General Manager, Biotech Consortium India Limited, Dr. O.P. Govila, Former Professor of Genetics, Indian Agricultural Research Institute and other team members at BCIL for backend support in finalizing these documents.

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# BIOLOGY OF ZEA MAYS (MAIZE)

## 1. GENERAL DESCRIPTION

Maize or corn (*Zea mays*) is a plant belonging to the family of grasses (*Poaceae*). It is cultivated globally being one of the most important cereal crops worldwide. Maize is not only an important food crop for human consumption, but also a basic element of animal feed and raw material for manufacturing of many industrial products. The products include corn starch, maltodextrins, corn oil, corn syrup and products of fermentation and distallaries. It is also being recently used in the production of biofuel.

Maize is a versatile crop grown over a range of agro-climatic zones. In fact the suitability of maize to diverse environments is unmatched by any other crop. It is grown from 58°N to 40°S, from below sea level to altitudes higher than 3000 m, and in areas with 250 mm to more than 5000 mm of rainfall per year (Shaw, 1988; Dowswell *et al.*, 1996) and with a growing cycle ranging from 3 to 13 months (CIMMYT, 2000). However, the major maize production areas are located in temperate regions of the globe. The United States, China, Brazil and Mexico account for 70% of global production. India has 5% of corn acreage and contributes 2% of world production (faostat.fao.org, 2008).

The use of maize varies in different countries. In USA, EU, Canada and other developed countries, maize is used mainly to feed animals directly or sold to feed industries and as raw material for extractive/ fermentation industries (Morris, 1998; Galinat, 1988; Shaw, 1988, Mexico, 1994). In developing countries use of maize is variable. In Latin America and Africa the main use of maize is for food while in Asia it is used for food and animal feed. In fact in many countries it is the basic staple food and an important ingredient in the diets of people. Globally, it has been estimated that approximately 21% of the total grain produced is consumed as food.

Maize is the third most important food grain in India after wheat and rice. In India, about 28% of maize produced is used for food purpose, about 11% as livestock feed, 48% as poultry feed, 12% in wet milling industry (for example starch and oil production) and 1% as seed (AICRP on Maize, 2007). In the last one decade, it has registered the highest growth rate among all food grains including wheat and rice because of newly emerging food habits as well as enhanced industrial requirements.

Maize is a crop par excellence for food, feed and industrial utilization. The composition of edible portion of maize (dry) is given in Table 1.

Calories	342.0	Calcium (mg)	10.0
Moisture (g)	14.9	Iron (mg)	2.3
Carbohydrates (g)	66.2	Potassium (mg)	286.0
Protein (g)	11.1	Magnesium (mg)	139.0
Fat (g)	3.6	Copper (mg)	0.14
Fibre (g)	2.7	Amino acids (mg)	1.78
Minerals (g)	1.5	Riboflavin (mg)	0.10
Phosphorus (mg)	348.0	Thiamine (mg)	0.42
Sodium (mg)	15.9	Vitamin C (mg)	0.12
Sulphur (mg)	114.0	Carotene (ug)	90.0

Table 1: Composition per 100 g of edible portion of maize (dry)

Source: Gopalan et al., 2007

However, it is deficit in essential amino acid, lysine and tryptophan. To overcome this deficiency, quality protein maize (QPM) with sufficiently higher quantity of lysine and tryptophan have been developed.

# 2. TAXONOMY, GEOGRAPHIC ORIGIN AND GENOMIC EVOLUTION

#### 2.1 Taxonomy

Maize belongs to the tribe Maydeae of the grass family Poaceae. "Zea" (zela) was derived from an old

Greek name for a food grass. The genus Zea consists of four species of which Zea mays L. is economically important. The other Zea sp., referred to as teosintes, are largely wild grasses native to Mexico and Central America (Doeblay, 1990). The number of chromosomes in Z. mays is 2n = 20.

Name	Maize
Kingdom	Plantae
Division	Magnoliophyta
Class	Liliopsida
Order	Poales
Family	Poaceae
Genus	Zea
Species	mays

Tribe Maydeae comprises seven genera which are recognized, namely Old and New World groups. Old World comprises *Coix* (2n = 10/20), *Chionachne* (2n

= 20), *Sclerachne* (2n = 20), *Trilobachne* (2n = 20) and *Polytoca* (2n = 20), and New World group has *Zea* and *Tripsacum*. It is generally agreed that maize phylogeny was largely determined by the American genera *Zea* and *Tripsacum*, however it is accepted that the genus *Coix* contributed to the phylogenetic development of the species *Z. mays* (Radu *et al.*, 1997).

#### 2.2 Relatives of Maize and Their Distribution

The closest wild relatives of maize are the teosintes which all belong to the genus *Zea*. Outside the *Zea* genus, the closest wild relatives are from the genus *Tripsacum*. Information about wild relatives of *Z. mays* has been elaborated in biology documents prepared by various agencies (CFIA, 1994; OECD,2006; AGBIOS) and is presented below:

#### **Teosintes**

The *Teosintes* are wild grasses native to Mexico and Central America and have limited distribution (Mangelsdorf *et al.*, 1981). *Teosinte* species show little tendency to spread beyond their natural range and distribution is restricted to North, Central and South America. They are not reported to occur in Southeast Asia (Watson & Dallwitz, 1992).

The nearest *Teosinte* relative to *Z. mays* is *Z. mays* ssp. *mexicana* (Schrader). It was previously classified as *Euchlaena mexicana*, *Z. mexicana* (2n = 20). This Central Mexican annual *Teosinte* is a large flowered, mostly weedy grass with a broad distribution across the central highlands of Mexico. It does not spread readily. It has limited use as a forage and green fodder crop, but can be problematic due to weedy tendencies (Doebley, 1990; Watson & Dallwitz, 1992).

*Z. diploperennis* (Iltis *et al.*, 1979) and *Z. perennis* (Hitchcock and Chase, 1951; Reeves and Mangelsdorf, 1939) are diploid (2n = 20) and tetraploid (2n = 40) perennial teosintes, respectively, with narrow distributions in Jalisco, Mexico. *Z. diploperennis* was on the threshold of extinction when found in the late 1970s and has since been used extensively for investigating maize ancestry (Eubanks, 1995). *Z. perennis* is reported to be established in South Carolina, USA (Doebley, 1990; Hitchcock and Chase, 1951).

*Z. luxurians* (Durieu *et* Asch.) is an annual *Teosinte* from southeastern Guatemala (Doebley, 1990; Watson & Dallwitz, 1992).

*Z. mays* ssp. *parviglumis* (Iltis and Doebley, 1980) is a small-flowered, mostly wild teosinte of southern and western Mexico (Doebley, 1990).

*Z. mays* ssp. *huehuetenangensis* (Iltis and Doebley, 1980) is a narrowly distributed teosinte of the western highlands of Guatemala (Doebley, 1990).

#### Tripsacum

The genus *Tripsacum* is comprised of about 12 species that are mostly native to Mexico and Guatemala but are widely distributed throughout warm regions in the USA and South America, with some species present in Asia and Southeast Asia (Watson & Dallwitz, 1992). All species are perennial, warm season grasses.

Species of economic importance to agriculture in Southeast Asia include *Tripsacum dactyloides* (L.)L and *T. laxum* Scrib and Merr. *T. dactyloides* (L.) L. (Eastern gamagrass) is a warm season grass native to

Mexico and Guatemala but now distributed throughout the Western Hemisphere to Malaysia (FAO, 2000b). *T. dactyloides* has a haploid chromosome number of n =18, with ploidy levels ranging from 2n = 2x = 36 up to 2n = 6x = 108 (deWet *et al.*, 1972, 1983). Different accessions exhibit both sexual and apomictic (asexual) reproductive capabilities. *T. dactyloides* has been the focus of extensive breeding work to transfer the apomictic reproductive nature from *Tripsacum* to *Zea mays* (Kindinger *et al.*, 1995; Savidan *et al.*, 1995).

*T. laxum* Scrib and Merr. (Guatemala grass) is a warm season grass native to Central America but now distributed throughout Mexico, South America, Sri Lanka, and Southeast Asia to Fiji. It is used as a fodder grass in Southeast Asia and is reportedly used as a soil binder and organic-matter builder in upland tea estates (FAO, 2000a). It does not flower readily and seed production is unusual except in its native habitat. *T. laxum* has been reported as a significant weed species (Watson & Dallwitz, 1992).

*T. andersonii* Gray (Guatemala) is native to Central America and has 64 chromosomes. It has been suggested to be a hybrid between *Z. luxurians* (n=10) and *T. latifolium* (n=18, 3x=54) (Talbert *et al.*, 1990). It has minor importance as a fodder crop (Watson & Dallwitz, 1992).

*T. lanceolatum* occurs in the Southwest USA and is found along the Sierra Madre Occidental north up to Arizona. *T. floridanum* is native to south Florida and Cuba. *T. manisuroides* is known only from Tuxtla Gutierrez, Chiapas, Mexico (deWet *et al.*,1982, 1983).

#### Coix and other Asiatic Genera

The *Asiatic* genera of the Maydeae tribe are native to an area extending from India to Southeast Asia and the Polynesian islands to Australia (Watson & Dallwitz, 1992). They include *Coix* L. (2n=10, 20 and 40), *Sclerachne* R. Br. (2n=20), *Polytoca* R. Br. (2n=20 and 40), *Chionachne* R. Br. (2n=20) and *Trilobachne Schenk* and *Henrard* (2n=20).

Species from these genera are annuals or perennials and are commonly found in forest margins. Species of *Chionachne* and *Coix* also inhabit streamsides, and open habitats and swamp areas, respectively (Watson & Dallwitz, 1992).

*Coix* sp. is the most familiar genera and includes several species. The species *Coix lacryma-jobi* Linn. (Job's Tears) (2n=20) is native to Southeast Asia and exists in the wild and as cultivated races. It is also found wild in Africa and Asia and warmer parts of the Mediterranean. *Coix* species have been cited as having weediness potential (Watson & Dallwitz, 1992).

*Chionachne* includes several species native to Southeast Asia. The species *C. semiteres* is cultivated as a fodder crop (Watson & Dallwitz, 1992).

*Polytoca* includes a few species, none of which are commonly cultivated. One species has been described for *Trilobachne* and is not known to be cultivated. Both genera are native to Southeast Asia (Watson & Dallwitz, 1992). *Sclerachne* has previously been cited to include one species (Soreng *et al.*, 2000).

#### 2.3 Geographic Origin and Distribution

The center of origin for *Z. mays* has been established as the Mesoamerican region, now Mexico and Central America (Watson & Dallwitz, 1992). Archaeological records suggest that domestication of maize began atleast 6000 years ago, occurring independently in regions of the southwestern United States, Mexico, and Central America (Mangelsdorf, 1974). The Portuguese introduced maize to Southeast-Asia from the America in the 16<sup>th</sup> century. The maize was introduced into Spain after the return of Columbus from America and from Spain it went to France, Italy and Turkey. In India, Portuguese introduced maize during the seventeenth century. From India, it went to China and later it was introduced in Philippines and the East Indies. Corn now is being grown in USA, China, Brazil, Argentina, Mexico, South Africa, Rumania, Yugoslavia and India.

Various hypothesis have been proposed on the origin/domestication of maize (OECD, 2006). *Teosintes* (*Z. diploperennis* and *Z. mays* ssp. *mexicana*) and *Tripsacum* species are often described as having roles in the domestication process of maize (Mangelsdorf, 1974; Galinat, 1988). An early hypothesis proposed that *Z. mays* ssp. mexicana was the product of a natural hybridization of *Tripsacum* and *Zea* (Mangelsdorf, 1974). Further crossings of *Teosinte* with wild maize are thought to have produced the modern races of maize. The possibility of intergeneric hybridization of either *Z. diploperennis* or *Tripsacum* with an extinct wild maize has also been proposed as the ancestral origin of *Z. mays* (Radu *et al.*, 1997; Purseglove, 1972). Eubanks (1993, 1997a) suggested that domesticated maize may have arisen via human selection of natural hybrids between *Tripsacum* and perennial *Teosinte*.

#### 2.4 Germplasm Diversity

Maize is a cultivated crop throughout the world and accordingly germplasm resources are preserved *ex situ* in many parts of the world. However, only in the Meso-American region there still exists, *in situ*, the original ancient maize that gave rise to improved varieties that are grown in all regions of the world. Most of the maize variation can be found in the Meso-American region and the northern part of South America. The great diversity of environments and conditions have created the basis for the development of maize varieties well adapted to harsh conditions of soil and climate as well as to biotic stresses. There is a close correlation among community culture, production system and the type of consumption of maize, with the diversification and variation of maize (Aguirre *et al.*, 1998; Louette and Smale, 1998).

There is a growing trend in developing countries to adopt improved maize varieties, primarily to meet market demand. In Mexico, only 20% of the corn varieties grown 50 years ago remain in cultivation (World Watch Institute, 2000). The narrowing of genetic diversity in modern maize varieties emphasizes the importance of conserving genetic traits for future plant breeding. CIMMYT (International Maize and Wheat Improvement Centre) has taken the lead in preserving maize germplasm. It has the world's largest collection of maize accessions, with over 17,000 lines (CIMMYT, 2000).

India also harbours diverse maize germplasm (Singh, 1977; Wilkes, 1981). Landraces with primitive characteristics exist in the North-Eastern Himalayan (NEH) region and are called "Sikkim Primitives"

(Dhawan, 1964). An extensive collection of germplasm from the entire NEH region has been made by researchers at the Indian Agricultural Research Institute (IARI), New Delhi. It has been shown that the two primitive Sikkim maize strains (Sikkim Primitive 1 and Sikkim Primitive 2) were different from the primitive Mexican races (Mukherjee *et al.*, 1971). Indian maize races have been classified under four categories i.e. primitive group, advanced or derived group, recent introduction and hybrid races (Singh, 1977). The National Gene Bank at New Delhi houses about 6,000 indigenous accessions. Systematic and comprehensive evaluation of this germplasm is being attempted for agronomically useful traits (Prasanna *et al.*, 2009).

In addition to the races, there are several local varieties in India. The genetic variability has resulted by crossing of Indian germplasm with strains imported from other countries particularly USA (Mukherjee, 1989). It has been reported that crosses of Indian x Indian germplasm gave yield superiority of 24-43 per cent, whereas Indian x US dent germplasm out yielded local varieties by 58 per cent (Dhawan and Singh, 1961). Highest yielding single cross hybrids were obtained from crosses between Indian x USA germplasm followed by USA x USA and Indian x Indian germplasm, thus highlighting the significance of genetic divergence for obtaining higher yields (Ahloowalia and Dhawan, 1963). Dent x Flint crosses involving Indian and Caribbean, and Indian and US germplasm showed highest expression of heterosis over better parent (47-54%) (Mukherjee and Dhawan, 1970).

# 3. **REPRODUCTIVE BIOLOGY**

#### 3.1 Growth and Development

Maize is a tall, determinate, monoecious, annual plant. It produces large, narrow, opposite leaves, borne alternatively along the length of stem. All maize varieties follow same general pattern of development, although specific time and interval between stages and total number of leaves developed may vary between different hybrids, seasons, time of planting and location. The botanical features of various plant parts are detailed in Annexure-I. The various stages of maize growth are broadly divided into the vegetative and reproductive stages as follows:

#### Vegetative Stages

- Seedling/Sprouting stage comes about one week after sowing, and the plants have about 2-4 leaves at this stage.
- Grand growth stage also called knee height stage of plant, arrives about 35-45 days after sowing.
- Tasseling/Flower initiation stage is the stage at which the tassels or male flowers appear. Generally the maize plant would have attained its full height by this stage.

#### **Reproductive Stages**

• Silking stage involving the formation of the female flowers or cobs is the first reproductive stage and occurs 2-3 days after tasseling stage. This stage begins when any silks are visible outside the husk. These are auxillary flowers unlike tassels that are terminal ones. Pollination occurs when these new moist silks catch the falling pollen grains.

- Soft-dough/Milky stage commences after pollination and fertilization is over. Grains start developing but they do not become hard. This soft dough stage is noticed by the silks on the top of the cob which remain partially green at this stage. The covering of the cobs also remains green.
- Hard dough/Maturity stage shows that the leaves get dried; silks get dried completely and become very brittle. Harvesting is done at this stage.

#### 3.2 Floral Biology

Maize is a monoecious plant, i.e. the sexes are partitioned into separate pistillate (ear), the female flower and staminate (tassel), the male flower (Figure 1). It has determinate growth habit and the shoot terminates into the inflorescences bearing staminate or pistillate flowers (Dhillon and Prasanna, 2001). The main shoot terminates in a staminate tassel. Maize is generally protandrous, i.e. the male flower matures earlier than the female flower. Within each male flower spikelet, there are usually two functional florets, although development of the lower floret may be delayed slightly in comparison to the upper floret. Each floret contains a pair of thin scales i.e. lemma and palea, three anthers, two lodicules and rudimentary pistil. Pollen grains per anther have been reported to range from 2000 to 7500 (Kiesselbach, 1949). Within an average of 7000 anthers per tassel and 2000 grains per anther, each tassel could produce 14 x 6-10 pollen grains. Kiesselbach (1949) estimated that 42,500 pollen grains are produced per square inch of corn field. In terms of the ratio of pollen grains produced per ovules fertilized, it appears that since each ear requires about 1000 pollen grains for fertilization, there are about 20,000 pollen grains per kernel in excess of what is actually needed if pollination were 100 percent efficient. The pollen grains are very small, barely visible to the naked eye, light in weight, and easily carried by wind. The wind borne nature of the pollen and protandry lead to cross-pollination, but there may be about 5% self-pollination.



Figure 1: Maize plant with male and female flowers Source: www.openlearn.openacuk/

The female flower initially is smooth but protuberances soon form in rows. The basal protuberances are formed first and development advances towards the tip of the ears. The part above the attachment of the carpel develops a single sessile ovule, which consists of a nucellus with two integuments or rudimentary seed coats. The united carpel's, which will form the ovary wall or pericarp of the mature kernel, grow upward until they completely enclose the ovule. The two anterior carpels, which face the ear tip, form outgrowths, which develop into the style i.e into long thread, known as silks. Silks are covered with numerous hairs, trichomes which form an angle with the silk where pollen grains are harboured. The base of the silk is unique, as it elongates continuously until fertilization occurs. The cobs bear many rows of ovules that are always even in number.

The female inflorescence or ear develops from one or more lateral branches (shanks) usually borne about half-way up the main stalk from auxillary shoot buds. As the internodes of the shanks are condensed, the ear remains permanently enclosed in a mantle of many husk leaves. Thus the plant is unable to disperse its seeds in the manner of a wild plant and instead it depends upon human intervention for seed shelling and propagation.

#### 3.3 Pollination and Fertilization

In maize, the pollen shed is not a continuous process and usually begins two to three days prior to silk emergence and continues for five to eight days. The silks are covered with fine, sticky hairs which serve to catch and anchor the pollen grains. Pollen shed stops when the tassel is too wet or too dry and begins again when temperature conditions are favourable. Under favourable conditions, pollen grain remains viable for only 18 to 24 hours. Cool temperatures and high humidity favour pollen longevity. Under optimal conditions the interval between anthesis and silking is one to two days. Under any stress situation this interval increases. Fertilization occurs after the pollen grain is caught by the silk and germinates by growth of the pollen tube down the silk channel. Within minutes of coming in contact with a silk and the pollen tube grows the length of the silk and enters the embryo sac in 12 to 28 hours. Pollen is light and is often carried considerable distances by the wind. However, most of it settles within 20 to 50 feet. Pollen of a given plant rarely fertilizes the silks of the same plant. Under field conditions, 97% or more of the kernels produced by each plant are pollinated by other plants in the field. Fertilization of ovules begins about one third of the way up from the base of the ear.

#### 3.4 Seed Dispersal

Seed dispersal of individual kernels naturally does not occur because of the structure of the ears of maize. Maize, as a thoroughly domesticated plant, has lost all ability to disseminate its seeds and relies entirely on the aid of man for its distribution (Stoskopf, 1985). The kernels are tightly held on the cobs. In case ears fall to the ground, so many competing seedlings emerge that the likelihood that any will grow to maturity is extremely low.

#### 3.5 Mating Systems

Under natural conditions, maize reproduces only by seed production. Pollination occurs with the

transfer of pollen from the tassels to the silks of the ear. About 95% of the ovules are cross-pollinated and about 5% are self-pollinated (Poehlman, 1959). Although plants are completely self-compatible. Rhoades (1931) and many workers have reported cytoplasmic male sterility (CMS) in maize. Based on restorer/ maintainer reaction patterns, Duvick (1965) and Beckett (1971) defined three CMS types: CMS-T (Taxas), CMS-S (USDA) and CMS-C (Charrua).

#### 3.6 Methods of Reproductive Isolation

Maize, being a cross-pollinated crop, various reproductive isolation methods are used by plant breeders and by seed producer to produce genetically pure seed. The isolation of crops using separation distances and physical barriers are common techniques for restricting gene flow and ensuring seed purity for maize seed production. Various experimental practices used to maintain reproductive isolation maize are:

- i) Maintaining isolation distance: Cross-pollination is controlled in seed lots by separating different lines. The Minimum Seed Certification Standards require 200m 600m isolation distance for maize seed production of various varieties and hybrids (Tunwar and Singh, 1988).
- ii) **Detasseling:** Mechanical removal of tassels is another effective method in corn. By remaining the tassel containing the pollen produce male flowers, it is possible to eliminate entirely the source of genetic material from the male flower that can be transferred via pollen.
- iii) Use of barrier crops: The use of barrier crops have been recommended to decrease the distance, but at the same time achieve the required separation. Barrier crop provides a physical barriers for pollinators. The African tall maize has been used as a barrier crop, as it is taller than maize crop, and is dense and thus provides an effective barrier in preventing cross pollination outside the experimental plot.

In line with the above, requirements of appropriate isolation distance along with rows of African tall maize has been adopted for conducting confined field trial of genetically engineered maize.

# 4. CROSSABILITY BETWEEN ZEA SPP. AND HYBRIDIZATION

#### 4.1 Intra Specific Crosses

Maize essentially being nearly 100% cross pollinated crop species, all varieties of maize freely cross pollinate forming fertile hybrids (Purseglove, 1972).

### 4.2 Interspecific Crosses

There is also great sexual compatibility between maize and annual teosinte and it is known that they produce fertile hybrid (Wilkes, 1977). It has been reported that all teosintes can be crossed to maize and form fertile hybrids, except for the tetraploid *Z. perennis*. However, maize teosinte hybrids exhibit low fitness and have little impact on gene introgression in subsequent generations (Galinat, 1988). The tendency

to form natural hybrids differs among *Teosintes*: *Z. luxurians* rarely hybridizes with maize, wheras *Z. mays ssp mexicana* frequently forms hybrids (Wilkes, 1997). Molecular data confirms that there is gene flow between maize and *Teosintes* and suggests that introgression of maize and *Teosintes* occurs in both directions but at low levels( Doebley, 1990).

#### 4.3 Intergeneric Hybridization

Although, it is difficult, *Tripsacum* species (*T. dactyloides, T. floridanum, T. lanceolatum*, and *T. pilosum*) have been successfully hand crossed with maize to form hybrids. However these hybrids have a high degree of sterility and are generally unstable. This infertility is common in such wide crosses because of differences in chromosome number and lack of pairing between chromosomes (Eubanks, 1997b). The maize-*Tripasacum* hybrids generally have 28 chromosomes, 10 from maize and 18 from *Tripsacum*, and are pollen sterile with limited female fertility (Mangelsdorf & Reeves, 1939; Mangelsdorf 1974).

Though studies on the Asiatic Maydeae (*Coix, Sclerachne, Polytoca, Chionachne, Trilobachne*) are very limited, no reports have been found on the ability of these genera to cross with *Z. mays*. Genetic studies using isozyme analyses indicate that the Asiatic genera are very distinct from both maize and *Teosintes* (Katiyar & Sachan, 1992). Chromosomal studies between maize and *Coix* sp. revealed strong structural differences between these genomes although the number of chromosomes (2n = 20) is the same for both genera (Katiyar *et al.*, 1992). The similarity in chromosome number suggest that there may be potential for crossing to occur between maize and the *Asiatic* genera. The genera *Trilobachne*, *Chionachne* and *Coix* have been studied to screen germplasm for disease resistant genes for potential use in cultivated maize (Sharma *et al.*, 1995).

Maize readily crosses with hexaploid wheat (*Triticum aestivum*) with high frequencies of fertilization and embryo formation (plant breeders use maize pollen to develop double haploids of hexaploid wheat). However, maize chromosomes are eliminated from the genome during the initial stages of meiosis and result in haploid embryos (Laurie & Bennett, 1986). There is little evidence to suggest fertile hybrids between maize and hexaploid wheat could be produced in nature.

There have been unsubstantiated reports of hybridization between maize and sugar cane (Saccaharum sp.).

#### 4.4 Wild Relatives in India

There are no sexually compatible wild or weedy relatives of *Zea* in proximity to the corn producing areas in India. The *American* genera are not native to India. All of the Oriental genera *Chinoachne* species are widely distributed in India, particularly in dry regions of Eastern and Western Ghats, Tamil Nadu and other parts of the country.

# 5. ECOLOGICAL INTERACTIONS

#### 5.1 Outcrossing and Gene Flow

Gene flow from maize can occur by two means: pollen transfer and seed dispersal. Seed dispersal can be readily controlled in maize as domestication has all but eliminated any seed dispersal mechanisms that ancestral maize may have previously used (Purseglove, 1972). As mentioned earlier, the kernels are held tightly on the cobs and if the ear falls to the ground, competing seedlings limit growth to maturity (Gould, 1968).

Pollen movement is the only effective means of gene escape from maize plants. As maize is mainly cross pollinated, wind speed and direction affects pollen distribution. Maize pollen measuring about 0.1 mm in diameter and largest pollen among members of the grass family, has been reported to be disseminated by wind from a comparatively low level of elevation. Further, due to its large size, maize pollen settles at a rate that is approximately 10 times faster than pollen from other wind-pollinated plants (Di-Giovanni *et al.*, 1995). Raynor *et al.* (1972) showed maize pollen is not transported as far by the wind as smaller pollen grain; does not disperse as widely horizontally or vertically; and settles to earth more quickly, much of it within the source itself.

Insects, such as bees, have been observed to collect pollen from maize tassels, but they do not play a significant role in cross-pollination as there is no incentive to visit the female flowers (Rayor *et al.*, 1972).

However, in the cultivation of commercial maize varieties, differences in flowering dates are small and thus cross-pollination between varieties may occur if grown in adjacent fields. The limited viability of maize pollen reduces the risk of cross-pollination, as a receptive host must be found within the 30 minutes that the pollen remains biologically active (Luna *et al.*, 2001). Cross-pollination is also affected by the concentration of maize pollen released; pollen produced by a maize crop will successfully compete with foreign pollen sources when present in higher concentrations (Rayor *et al.*, 1972).

Gene flow from maize (*Zea mays*) to other species in the same genus (interspecific) and between genera (intergeneric) first requires the formation of a viable intermediate hybrid that is capable of producing fertile progeny that can survive into the next generation. Assuming sexual compatibility exists, other factors also contribute to the likelihood of hybridization: proximity of the crop and related species to each other; environmental conditions; and overlapping flowering periods. The introgression of genes from maize to other plant species may require several generations of recurrent backcrossing.

#### 5.2 Potential for Gene Transfer from Maize

#### 5.2.1 Gene transfer between different maize species

Based on the available information, *Z. mays* can be crossed with teosintes to form fertile hybrids which exhibit low fitness and have little impact on gene introgression in subsequent generations. The similarity in chromosome number suggests that there may be potential for crossing to occur between *Z. mays* and wild species from the Asiatic genera (*Coix, Sclerachne, Polytoca, Chinachne, Trilobachne*). However, studies on Asiatic genera are limited and there are no reports found on ability of these to cross with *Z. mays*.

#### 5.2.2 Gene transfer from maize to other plants

The introgression of genetic information from one plant to another is only significant if the two plants are sexually compatible and if their hybrid offspring are viable. This is not applicable in maize.

#### 5.2.3 Gene transfer from maize to other organisms

Horizontal gene transfer from plants to animals (including humans) or microorganisms is extremely unlikely. No evidence has been identified for any mechanism by which maize genes could be transferred to humans or animals, nor any evidence that such gene transfer has occurred for any plant species during evolutionary history, despite animals and humans eating large quantities of plant DNA. The likelihood of maize genes transferring to humans and other animals is therefore effectively zero. Similarly gene transfer from maize, or any other plant, to microorganisms is extremely unlikely.

Horizontal gene transfer from plants to bacteria has not been demonstrated experimentally under natural conditions (Nielsen *et al.*, 1997; Nielsen *et al.*, 1998; Syvanen, 1999) and deliberate attempts to induce such transfers have so far failed (Schlüter *et al.*, 1995; Coghlan, 2000).

#### 5.3 Free Living Populations of Maize

The term "free living" is assigned to plant populations that are able to survive, without direct human assistance, over long term in competition with the native flora. This is a general ecological category that includes plants that colonize open, disturbed prime habitat that is either under human control (weedy populations) or natural disturbed areas such as river banks and sand bars (wild populations). There are no such free living populations of maize in India.

#### 5.4 Volunteers and Weediness

As already mentioned, maize has gained agronomically significant attributes and depends on human intervention to disseminate its seed. It has become so domesticated that seeds cannot be separated from the cob and disseminated without human intervention.

Although maize from previous crop year can sometimes germinate the following year, it cannot persist as a weed. Volunteers are common in many agronomic systems, but they are easily controlled because maize is incapable of sustained reproduction outside of domestic cultivation. Maize plants are non-invasive in natural habitats (Gould, 1968).

## 6. HUMAN HEALTH CONSIDERATIONS

In maize no endogenous toxins or significant levels of antinutritional factors have been found till date. It is not considered a pathogen and is not capable of causing any disease in humans, animals or plants. Maize allergy can occur due to the ingestion of maize or maize derivatives, or to the inhalation of maize flour or maize pollen. Although some cases of maize allergy have been reported but no proteins responsible for allergy have yet been identified.

## 7. MAIZE CULTIVATION IN INDIA

#### 7.1 Climate and Soil Requirements

Maize crop is primarily a warm weather crop and it is grown in wide range of climatic conditions (ICAR, 2006). Maize can successfully be grown in areas receiving an annual rainfall of 60 cm, which should be well distributed throughout its growing stage. It needs more than 50% of its total water requirements in about 30 to 35 days after tasseling and inadequate soil moisture at grain filling stage results in a poor yield and shriveled grains. It cannot withstand frost at any stage.

Prolonged cloudy period is harmful for the crop but an intermittent sunlight and cloud of rain is the most ideal for its growth. It needs bright sunny days for its accelerated photosynthetic activity and rapid growth of plants.

In India, maize is traditionally grown in monsoon (Kharif) season, which is accompanied by high temperature ( $<35^{\circ}$  C) and rains. However, with the development of new cultivars and appropriate production technology, winter cultivation of maize has emerged as a viable alternative.

Soil texture is a foremost requirement as it controls moisture and nutrient capacity. Loam or silt loam surface soil and brown silt clay loam having fairly permeable sub soil are the ideal soil types for cultivation of maize. Deep fertile soils rich in organic matter and well-drained soils are the most preferred ones, however, maize can be grown on variety of soils. Soil pH in the range of 7.5 to 8.5 supports good crop growth, however, at pH beyond these extremes, problems of toxicity are found with certain elements and essential nutrients. Since about 85% of maize in India is grown during monsoon season, where over 80% of the total annual precipitation is received, it thus is very imperative for the soil to have adequate water holding capacity as also the proper drainage to minimize damage due to water logging and seed and seedling diseases.

#### 7.2 Zonalization of Varietal Testing System

The Directorate of Maize Research (DMR), upgraded in January 1994 from its earlier status as All India Coordinated Maize Improvement Project (AICMIP), has the mandate to conduct and coordinate maize research, generate improved technology for continuous enhancement in productivity of maize, and

promote the diversified uses of its products. The DMR has demarcated 5 zones for varietal testing as indicated below:

- i) Zone 1 Northern India Hills Zone: This is further subdivided into 1-A comprising of Jammu & Kashmir, Himachal Pradesh and Uttaranchal. 1-B comprising of North eastern India and Assam.
- ii) Zone 2 Northern Plains Zone: This comprises of Punjab, Haryana, Delhi and western Uttar Pradesh.
- iii) Zone 3 Comprising of Rajasthan, Gujarat and Madhya Pradesh
- iv) Zone 4 Comprising of Peninsular India.
- v) Zone 5 New Maize growing areas like Orissa, Jharkhand, West Bengal etc.

#### 7.3 Pests and Diseases of Maize

Of the 130 insect pests that affect maize crop, stem borers, shootfly, armyworm, jassids, thrips, white ants, pyrilla, grasshoppers, grey weevil, hairy caterpillars, root worms, earworms and leaf miner are more serious, though the spectrum varies in different agro-ecological regions. Most of the research efforts have gone into breeding for resistance to European corn borer (*Ostrinia nubilalis*), a pest of maize in the USA and Europe. Fall armyworm (*Spodoptera frugiperda*) is another very important pest in tropical and subtropical areas. In India, spotted stem borer (*Chilo partellus*) is the most serious pest. The major stalk-borer (*Chilo partellus*) is the major pest throughout the country, particularly during the Kharif season. The details of various insects pests affecting maize crop are placed in Annexure-II.

Maize suffers from about 110 diseases on a global basis caused by fungi, bacteria and viruses. The disease spectrum varies in different agro-climatic zones. Several diseases such as seed and seedling delights, foliar disease, downy mildews, stalk rots and leaf as sheath blight occur in various parts of the country. It has been indicated that diseases in Rabi maize are comparatively lesser than the Kharif maize. Details of diseases of maize are placed in Annexure-III. It has been reported that about 13.2% of the economic product of maize is estimated to be lost annually due to diseases (Dhillon and Prasanna, 2001). The information has been collected on distribution and appearance of diseases in different maize-growing regions in the country and different protection measures have been worked out (Sharma and Lal, 1998).

#### 7.4 Breeding Objectives, Milestones in Breeding Advances and Challenges

Maize breeding in India received great impetus with the establishment of the All India Coordinated Maize Improvement Project (AICMIP) in 1957. In the AICMIP though initial emphasis was on development of double cross hybrids using inbred lines but later on focus shifted to composite breeding and to early maturing composites. Significant progress has also been made in breeding single cross hybrids.

Breeding objectives depend upon various factors like the requirements of the farmers, market forces,

production level, constraints and cropping patterns in different climatic regions. The major objectives of maize breeding with reference to the Indian maize progarmme have been appropriate maturity, grain yield, tolerance to biotic stresses viz. diseases resistance, insect pest resistance, drought tolerance, cold tolerance.

Grain yield is the most important and complex trait. The salient components of maize yield are number of ears, kernel rows, kernels per row, test-weight, and shelling percentage. Stability of the performance is also very important to ensure high stable returns particularly in India where maize is cultivated during monsoon season characterized by erratic rains. The cultivars should possess an ability to perform well over a range of environment. Efforts have also been made to undertake breeding of high yielding corn hybrids which are stable across locations for both wet (Kharif) and dry (Rabi) seasons.

Breeding for appropriate maturity is another important objective. Maize is a short day plant. The time of flowering is influenced by temperature and photo period. In maize, various traits are used to measure maturity traits. Biotic and abiotic stress is important as considerable yield losses occur due to disease and pest-insects. Besides drought, cold, water logging, low nitrogen acidity are also important stresses. Maize breeders have also been quite successful in developing new cultivars having increased content of starch, oil and protein in the kernel.

Indian maize improvement programme has developed a large number of hybrids and composites. Ganga Safed-2, Ganga-5, Ganga 11, Histarch, Deccan 103 and Deccan 105 and composites, Vijay, Kiran, Ageti 76, navjot merit particular mention. Release of single cross hybrids i.e. Paras, PEHMI, PEHM2, PEHM3, BN2187 etc. are important landmark in heterosis breeding in India. Significant progress has been made in developing speciality maize types such as QPM (Quality protein maize), sweet corn, pop corn and baby corn.

Developing hybrids of suitable maturity and for marginal lands for unpredictable monsoon are the major challenges in breeding of maize. Insect pests like *Chilo partellus* in wet season and *Sesamia inferens* in dry season is a major breeding objective. Diseases like, post flowering stalk rots and ear rots pose a major problem. However, the progress in breeding for insect resistance is slower in comparison to disease resistance, due to lack of effective insect pest rearing methods and germplasm screening technique.

#### 7.5 Status of Maize Cultivation

Maize occupies an important place in Indian Agriculture. It is the third most important cereal in India after wheat and rice. The major maize growing states are Uttar Pradesh, Bihar, Rajasthan, Madhya Pradesh, Punjab, Andhra Pradesh, Himachal Pradesh, West Bengal, Karnataka and Jammu & Kashmir, jointly accounting for over 95% of the national maize production. The estimate of maize production in 2007 was 13-14 million tonnes from an area of 7.2 million hectares with an average productivity of 2 tons/hectare as shown below in Figure 2.

As mentioned earlier, maize is traditionally grown during the summer (monsoon) season, which is accompanied by high temperatures (<35° C) and rains. Rabi (winter) cultivation of maize is a relatively new introduction started in mid sixties in some pockets of Bihar and South India, but now in the

country as a whole. Rabi maize has comparative advantage of low incidence of diseases and insect pests, crops do not suffer on account of heavy rainfall, slow growth of weeds, etc. and hence, preferred by the farmers.



Though, the area under maize has shown an increasing trend, with maize emerging as a competitive crop,

*Figure 2:* Area, production and yield of maize in India *Source:* FAOSTAT, 2009 (faostat.fao.org)

the level of production has to be substantially raised to meet growing demand of maize for human food, animal feed, poultry feed, as well as industrial processing to produce value added products. Use of biotechnology has emerged as one of the important techniques for increasing the maize productivity by controlling the major insect pests as well as herbicide tolerance (James, 2008). Various other traits such as high lysine content, amylase enzyme, phytase enzyme, drought tolerance etc. are also being incorporated in maize (Stein and Rodriguez-Cerezo, 2009). The status of biotech interventions successfully applied in maize globally is presented in Annexure-IV. The confined field trials of transgenic maize containing traits such as insect resistance and herbicide tolerance are also underway in India.

# ANNEXURE - I

## **BOTANICAL FEATURES**

Maize is a tall, determinate annual  $C_4$  plant varying in height from 1 to 4 metres producing large, narrow, opposit leaves (about a tenth as wide as they are long), borne alternately along the length of a solid stem. The botanical features of various plant parts are as follows:

**Root:** Normally maize plants have three types of roots, i) seminal roots - which develop from radicle and persist for long period, ii) adventitious roots - fibrous roots developing from the lower nodes of stem below ground level which are the effective and active roots of plant and iii) brace or prop roots - produced by lower two nodes. The roots grow very rapidly and almost equally outwards and downwards. Favourable soils may allow corn root growth up to 60 cm laterally and in depth.

**Stem:** The stem generally attains a thickness of three to four centimeters. The inter nodes are short and fairly thick at the base of the plant; become longer and thicker higher up the stem, and then taper again. The ear bearing inter node is longitudinally grooved to allow proper positioning of the ear head (cob). The upper leaves in corn are more responsible for light interception and are major contributors of photosynthate to grain.

Flower: The apex of the stem ends in the tassel, an inflorescence of male flowers and the female inflorescences (cobs or ears) are borne at the apex of condensed, lateral branches known as shanks protruding from leaf axils. The male (staminate) inflorescence, a loose panicle, produces pairs of free spikelets each enclosing a fertile and a sterile floret (Figure 3). The female (pistillate) inflorescence, a spike, produces pairs of spikelets on the surface of a highly condensed rachis (central axis, or "cob"). The female flower is tightly covered over by several layers of leaves, and so closed in by them to the stem that they don't show themselves easily until emergence of the pale yellow silks from the leaf whorl at the end of the ear (Figure 4). The silks are the elongated stigmas that look like tufts of hair initially and later turn green or purple in colour.





Figure 3: Male flower, the tassel Figure 4: Female flower, the silk
Source: http://search.com/reference/Maize

Each of the female spikelets encloses two fertile florets, one of whose ovaries will mature into a maize kernel once sexually fertilized by wind-blown pollen.

Grain: The individual maize grain is botanically a caryopsis, a dry fruit containing a single seed fused to the inner tissues of the fruit case. The seed contains two sister structures, a germ which includes the plumule and radical from which a new plant will develop, and an endosperm which will provide nutrients for that germinating seedling until the seedling establishes sufficient leaf area to become autotrophy (Figure 5).



*Figure 5 : Maize Grain Source:* http://cerealsdb.uk.net/zeastar4.jpg

The germ is the source of maize "vegetable oil" (total oil content of maize grain is 4% by weight). The endosperm occupies about two - third of a maize kernel's volume and accounts for approximately 86% of its dry weight. The endosperm of maize kernels can be yellow or white. The primary component of endosperm is starch, together with 10% bound protein (gluten), and this stored starch is the basis of the maize kernel's nutritional uses.

# **ANNEXURE - II**

### KEY INSECT PESTS OF MAIZE

Over 130 insect pests have been reported to infect but only few are serious and require management. Among these, the most serious pests are the stem borers. The important pests affecting different stages of maize are listed below followed by brief explanation.

#### i) Stem Borer (Chilo partellus)

Stem borer a very serious pest of maize found throughout in India. The damaging stage of the pest is larvae. The eggs hatch in about two to five days. The freshly hatched caterpillars migrate towards the central shoot where they first feed on the tender leaves for sometime. Later on they bore into top internode and move downwards.

In case of younger plants, the growing point and base of central whorl gets badly damaged resulting into the drying up of the central shoot. It is commonly known as 'dead heart' (Figure 6). This condition, however, does not appear when the plant is attacked in the later stages.



*Figure 6: Maize plant infected with* Stem Borer (*Chilo partellus*) *Source: http://ipmworld.umn.edu/* 

#### ii) Pink Borer (Sesamia inferens)

It is a polyphagous pest and is not so serious in north as in south India. It generally attacks the crop in the late stage when cob formation starts in the field. Full grown larvae are stout, smooth bodied, 25-30 mm in length, 3 mm in width. Purplish pink on the dorsal side, ventral side white, head capsule reddish brown, larval period 3-4 weeks, migratory. Newly hatched larvae remain in group behind the

leaf sheath and begin chewing on the stem and epidermal layer of the sheath. Some migrate to neighbouring leaf sheaths, while others penetrate the stem. Whorl feeding of larvae results in rows of oblong holes in unfolding leaves unlike round shot holes produced by *Chilo partellus*. Later they bore into central shoot resulting in the drying up of the growing point and formation of dead heart in young maize plant. As a result of larval feeding some times the bottom internodes show circular ring like cuts. Severe damage causes the stem to break.

#### iii) Shoot Fly (Atherigona soccata, A.naquii)

It is a very serious pest of maize in South India but also severally damages spring and summer maize crop in North India. The attack is maximum when the crop is in seedling stage. The tiny maggots creep down under the leaf sheaths till they reach the base of the seedlings. After this they cut the growing point or central shoot which results in the formation of characteristic dead hearts.

#### iv) Shoot Bug (Peregrinus maidis)

It is occasionally a serious pest on maize. It is found attacking maize in South India and Madhya Pradesh with peak activity during August-October. Adult is yellowish-brown to dark brown with translucent wings in macropterous forms while brachypterous forms have underdeveloped wings, nymphs are yellowish and soft bodied. Adults and nymphs suck sap, resulting in unhealthy, stunted and yellow plants. Leaves wither from top downwards and plants die if attack is severe (Figure 7). Honey dew excreted by the insect causes growth of sooty mold on leaves. Mid ribs of leaves turn red due to egg laying and may dry subsequently.



Figure 7: Shoot Bug (Peregrinus maidis) infection in maize Source: http://ikisan.com

### v) Corn Leaf Aphid (Rhopalosiphum maidis)

The aphid is widely distributed and appears in serious form occasionally during drought years. Maize plants at the end of mid whorl stage are usually attacked. Aphids suck sap from plants and cause yellowing and mottling. Diseased plants may become stunted and turn reddish as they mature. If young plants infected they seldom produce ears. The aphid colony may some time cover completely the emerging tassels and the surrounding leaves preventing the emergence. Ears and shoots are also infested and seed set may be affected. Honey dew excreted by aphids favours the development of sooty molds.

# **ANNEXURE - III**

## MAJOR DISEASES OF MAIZE

Maize is subjected to as many as 112 diseases on a global basis. In India, there is record of about 35 of them. The major diseases prevalent in India are as under:

#### i) Maydis Leaf Blight (Helminthosporium maydis)

The disease is prevalent in northern states. Leaves show greyish tan, parallel straight sided or diamond shaped 1-4 cm long, lesions with buff or brown borders or with prominent colour banding or irregular zonation. Symptoms may be confined to leaves or may develop on sheaths, stalks, husks, ears and cobs.

#### ii) Downy Mildews

Downy mildews are group of fungi which attacks many economically important crop plants. Some of the important ones affecting maize are:

- Sorghum Downy Mildew (*Perenosclerospora sorghi*) Systemic interaction, usually localized in late planted areas, malformation of tassels (Figure 8).
- Brown Stripe Downy Mildew (*Sclerophthora rayssiae*) Symptoms observed only on leaves that show chlorotic strips; generally start from top leaves present a burnt appearance in advance stages (Figure 9).
- Crazy Top Downy Mildew (*Sclerophthora macrospore*) Partial or complete malformation of the tassel continues until it resembles a mass of narrow twisted leafy structures; stunted growth of plant (Figure 10).
- Sugarcane Downy Mildew (*Peronosclerospora sacchari*) Characterized by local lesions that initially are small, round, chlorotic spots and systemic infection which appears as pale yellow to white streaks on leaves (Figure 11). Downy growth on the both leaf surfaces and the plants may be distorted with small, poorly filled ears with mis-shapen tassels.



Figure 8: Sorghum Downy Mildew (Perenosclerospora sorghi)



Figure 9: Brown Stripe Downy Mildew (Sclerophthora rayssiae)



Figure 10: Crazy Top Downy Mildew (Sclerophthora macrospore)

Source: http://ikisan.com



Figure 11: Sugarcane Downy Mildew (Peronosclerospora sacchari)

#### iii) Pythium Stalk Rot (Pythium aphanidermatum)

The lower internodes near the ground level show brown elliptic lesions (Figure 12). In the affected part of the internode the pith is destroyed but not the fibre vascular bundles. The stalk as a result becomes weak and breaks leading to lodging of the plant. Such lodged plants continue to remain green for some days.

#### iv) Bacterial Stalk Rot (Erwinia carotovora, Erwinia chrysanthemi)

This disease occurs in many states where high temperatures coupled with high humidity develop during the pre-flowering stage of the crop. The organism is soil borne and makes its entry through wounds and injuries on the host surface. The organism survives saprophytically on debris of infected materials and serves primary inoculum in the next season. Ears and shank may also show rot. They fail to develop further and the ears hang down simply from the plant.

#### v) Common Rust (Puccinia sorghi)

This disease is prevalent in cooler parts of the country. It is very common in Himalayan region during Kharif season and in South India during Rabi season.

The symptoms are appearance of circular to elongate golden brown or cinnamon brown, powdery, erumpent pustules on both leaf surfaces (Figure 13). As the crop matures brownish black pustules containing dark thick walled two celled teliospores develop. In severe cases infection spreads to sheaths and other plant parts.

#### vi) Charcoal-Rot (Macrophamina phaseolina)

It is prevalent in comparatively drier maize growing areas, particularly Andhra Pradesh, Karnataka, West Bengal, Bihar, Uttar Pradesh and Delhi. The disease development is maximum during grain filling stage and is favoured by warm temperature (30-40°C) and low soil moisture. The fungus infects through roots and proceeds towards stem and plants show evidence of pre-mature ripening. The out sides of lower internodes become straw colored and the pith becomes badly disintegrated (Figure 14).



Figure 14 : Maize plant having Charcoal Rot (Macrophamina phaseolina) Source: http://ikisan.com



Figure 12: Pythium Stalk Rot (Pythium aphanidermatum) in maize Source: http://ikisan.com



Figure 13: Maize plant having Common Rust (Puccinia sorghi) infection Source: http:// bio.mq.edu.au/

#### vii) Brown Spot (Physoderma maydis)

The disease normally occurs in areas of high rainfall and high mean temperatures. It attacks leaves, leaf sheaths, stalks, and sometimes outer husks. The first noticeable symptoms develop on leaf blades and consist of small chlorotic spots, arranged as alternate bands of diseased and healthy tissue. Spots on the mid-ribs are circular and dark brown, while lesions on the laminae continue as chlorotic spots (Figure 15). Nodes and internodes also show brown lesions. In severe infections, these may coalesce and induce stalk rotting and lodging.



Figure 15: Brown Spot (Physoderma maydis) in maize Source: http://fao.org/

#### viii) Turcicum Leaf Blight (Exserohilum turcicum)

This is a fungal disease of maize prevalent in South India. It is seen both in Kharif and Rabi seasons. The early symptoms of the disease are oval, water-soaked spots on leaves and the later diseased stage shows

characteristic cigar shaped lesions that are 3 to 15cm long. These elliptical, long cigar-shaped gray-green or tan color lesions develop into distinct dark areas as they mature and become associated with fungal sporulation (Figure 16). Lesions typically first appear on lower leaves, spreading to upper leaves and the ear sheaths as the crop matures. Under severe infection, lesions may coalesce, blighting the entire leaf. Yield losses as high as 70% are attributed to Turcicum leaf blight.



Figure 16: Turcicum Leaf Blight (Exserohilum turcicum) of Maize Source: http://btny.purdue.edu/

# ANNEXURE - IV

## BIOTECH INTERVENTIONS IN MAIZE

Maize has been genetically engineered to insert agronomically desirable traits i.e. incorporation of a gene that codes for the *Bacillus thuringiensis* (Bt) toxin, protecting plants from insect pests and resistance to herbicides. Both pest resistant and herbicide tolerant genes/events have also been stacked. As of now several events of the two traits have been approved in different countries and are being extensively cultivated. In early 2009, there were nine different event of GM maize in the varieties cultivated globally and three additional maize events have been authorized in atleast one country worldwide, but not yet commercialized anywhere (Table 2).

Developer	Product name	Event name/ genes	Trait	Unique identifier
Commercialised ma	ize events			
Monsanto	YieldGardCorn Borer	MON810	Insect resistance (to lepidopterans)	MON-ØØ81Ø-6
Monsanto	RoundupReady Corn 2	NK603	Herbicide tolerance (to glyphosate)	MON-ØØ6Ø3-6
Monsanto	YieldGardRootworm	MON863	Insect resistance (to coleopterans)	MON-ØØ863-5
Monsanto	YieldGard VT	MON88017	Insect resistance (to coleopterans)	MON-88Ø17-3
Dow Agro Sciences and Pioneer Hi-Bred	Herculex I	1507	Insect resistance (to lepidopterans)	DAS-Ø15Ø7-1
Dow AgroSciences andPioneer Hi-Bred	Herculex RW	59122	Insect resistance (to coleopterans)	DAS-59122-7
Syngenta	Agrisure CB	Bt11	Insect resistance (to lepidopterans)	SYN-BTØ11-1
Syngenta	Agrisure GT	GA21	Herbicide tolerance (to glyphosate)	MON-ØØØ21-9
Syngenta	Agrisure RW	MIR604	Insect resistance (to coleopterans)	SYN-IR6Ø4-5
Maize events authorised in at least one country but not yet commercialised anywhere				
Monsanto	YieldGard VTPRO	MON89034	Insect resistance (to lepidopterans)	MON-89Ø34-3
Monsanto	High lysine	LY038	Crop composition (high lysine content)	REN-ØØØ38-3
Syngenta	n/a	3272	Crop composition (amylase content)	SYN-E3272-5

#### Table 2: Commercial GM maize and GM maize in the commercial pipeline worldwide

Source: Stein and Rodriguez-Cerezo, 2009

In addition, there are five more GM maize events that have entered the regulatory system in at least one country but that are not yet authorised anywhere in the world, namely Syngenta's new lepidopteranresistant maize, Pioneer's Optimum GAT maize and three GM maize events from China (Table 3).

Developer	Product name	Event name / genes	Trait	Unique identifier
Syngenta	Agrisure Viptera	MIR162	Insect resistance (to lepidopterans)	SYN-IR162-4
Pioneer Hi-Bred	Optimum GAT	98140	Herbicide tolerance (to ALS inhibitors and glyphosate)	DP-Ø9814Ø-6
n/a (China)	n/a	Cry1A	Insect resistance	n/a
n/a (China)	n/a	n/a	Crop composition (high lysine content)	n/a
n/a (China)	n/a	n/a	Crop composition (phytase enzyme)	n/a

Table 3: GM maize in the regulatory pipeline worldwide

In addition to the above seven new events containing traits regarding crop composition and drought tolerance are at advanced stages of research and development, as given in Table 4.

Developer	Product name	Event name / genes	Trait
Monsanto	n/a	MON87754	Crop composition (high oleic content)
Pioneer Hi-Bred	Optimum Acre Max 1	n/a	Insect resistance (to coleopterans)
Monsanto and BASF	n/a	MON87460	Abiotic stress tolerance (to drought)
Dow AgroSciences	DHT	n/a	Herbicide tolerance
n/a (India)	n/a	cry1Ac + cp4epsp4	Insect resistance
Syngenta	n/a	n/a	Abiotic stress tolerance (to drought)
BASF Plant Science	NutriDense	n/a	Crop composition (protein, amino acid
			and phytase content)

#### Table 4: GM maize in the advanced R&D pipeline worldwide

On a global basis, in 2008, genetically engineered maize occupied 37.3 million hectares equivalent to 24% of the global maize area of 157 million hectares (James, 2008). There have been a substantial increase in the deployment of stacked traits of Bt and herbicide tolerance, particularly in USA. The triple gene products in GM maize, featuring two Bt genes (one to control the European corn borer complex and the other to control root worm) and one herbicide trait have been widely adopted. The area occupied by maize with insect resistant and herbicide tolerant genes and the two characters stacked together is as follows (Table 5).

S. No.	GM maize containing	Area in million hectares
1.	Insect resistant gene	7.1
2.	Herbicide tolerant gene	5.7
3.	Stacked traits	24.5
	Total	37.3

Table 5: Global area of GM maize in 2008

Source: James, 2008

In total maize was grown in 17 countries worldwide. Major countries growing GE maize are USA, Argentina, Canada, Brazil and South Africa. Other countries include Uruguay, Philippines, Chile, Egypt, Honduras and seven EU countries.

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