

# Dedicated to my love for groundnut.

From the author,

This book is written in a format of a ready reckoner for quick consultation by the user. It contains all the essential details of different aspects of groundnut improvement and production. The information contained herein is extracted from various formal and informal publications on groundnut. These sources are gratefully acknowledged. For more details and in-depth information, the reader is advised to consult the additional literature listed at the end.

The book is being published open access so that the groundnut research and development community and students in developing countries could easily access it. Alternately, a pdf version can be, or downloaded from the Feed the Future Innovation Lab for Collaborative Research on Peanut Productivity and Mycotoxin (Peanut & Mycotoxin Innovation Lab) website (pmil.caes.uga.edu).

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# 1. Introduction

<u>Common names</u>: Groundnut or peanut in English, pistache in French, mani in Spanish, amondoim in Portuguese, huasheng in Chinese, mungphali in Hindi, ful sudani in Arabic.

Other not so common names: Monkey nuts, goober nuts, earth nuts.

<u>First written reference to groundnut</u>: Oldest specimens found in Peru dated to 7600 years ago; First written reference in 1535 by Gonzalo Hernandez de Oviedo y Valdes chronicles of travels in Americas; Crop consumed mainly by lowly men and boys and slaves; Used for hogging up to 1930s in the USA; Research efforts of Dr George Washington Carver (credited with inventing 300 different uses) during early 1900 made it an important commercial crop in the southern USA.

Scientific name: Arachis hypogaea L. (Given in 1753).

Life cycle of the cultivated crop: Annual (Legume).

World rank as an oilseed crop: 6<sup>th</sup> in edible oil production among the oilseed crops.

<u>World rank as a protein crop</u>: 3<sup>rd</sup> most important source of vegetable protein.

<u>World rank as a food crop</u>: 13<sup>th</sup> among the food crops (production utilized directly as food or in confections).

(Depending upon the total global production of various crops, these ranks can vary from year to year.)

<u>Utilization of different parts of groundnut plant</u>: <u>Seed (kernel)</u> – Consumed directly raw, roasted and boiled or processed into confections and peanut flour for flavor enhancement or crushed for oil for edible and industrial uses; Source of high quality edible oil (44–56%), easily digestible protein (22–30%), carbohydrates (10–25%), vitamins (E, K and B complex), minerals (Ca, P, Mg, Zn and Fe) and fiber; <u>Shell</u> – Used as fuel, animal feed, cattle litter, filler in feed and fertilizer industry and in making particle boards and alcohol and acetone after fermentation; <u>Haulm</u> (above ground vegetative parts) – Used as animal fodder or in manure; <u>Roots</u> – Being legume add nitrogen (100 – 152 kg/ha N) and organic matter to the soil.

(Groundnut cake obtained after extraction of oil is used in animal feed industry, in making weaning foods for children and invalid foods for aged people and as fertilizer. If groundnut produce is contaminated with aflatoxin, the cake will also be contaminated. However, groundnut oil would be free from aflatoxin, if it is refined. The contaminated cake should not be used as animal or human food.)

<u>Utilization pattern</u>: Varies from region to region (Table 1). Major groundnut oil consuming countries in Asia include India, China, Myanmar and Vietnam; Food use showing an increasing trend over the years.

Region	Sub-region	Food use (%)	Crushed for oil (%)	Other uses <sup>1</sup> (%)
America	North America	74	16	10
	South America	39	53	8
Africa	East Africa	49	46	5
	Southern Africa	71	17	12
	West Africa	55	33	12
Asia	East Asia	40	53	7
	Southeast Asia	69	23	8
	Southwest Asia	9	78	13
Europe	Western Europe	95	4	1
	Eastern Europe	100		***
World		41	49	10

**Table 1.** Utilization pattern of groundnut kernels in different regions of the world.

(Source: Based on USDA data, PS&D database. <sup>1</sup>includes feed, seed and waste.)

<u>Groundnut oil as biofuel</u>: In 1900 World's Fair in Paris, a diesel engine was run on pure groundnut oil; While in developing countries enhanced oil content is needed to meet the ever increasing demand for high quality edible oil, in the USA it is being targeted to produce peanut biodiesel to run farm machinery to reduce the cost of farm operations (USDA-ARS, 2008; Wright, 2012).

<u>Number of countries growing groundnut (2012)</u>: About 114 countries in tropical and warm temperate regions of the world; Commercial production largely confined between 40°N and 40°S latitudes.

<u>Top 10 groundnut growing countries in the world (based on production in 2012)</u>: China (40.7% of the total global production), India (14.0%), USA (7.4%), Nigeria (5.8%), Myanmar (3.3%), Sudan (2.5%), Argentina (1.9%), Indonesia (1.7), Senegal (1.6%) and Cameroon (1.3%); Together these top 10 countries account for about 80.2% of global groundnut production.

World area under groundnut (2012): 24.62 million ha.

World production of groundnut (2012): 41.26 million metric t.

World average productivity of groundnut (2012): 1675.9 kg/ha.

<u>Record yields reported in groundnut</u>: 10.5 t/ha over a small area under intensive cultivation in Shandong province in China (Yanhao *et al.*, 1996), 9.6 t/ha in large plots in Zimbabwe (Hildebrand, 1996), 9.4 t/ha in a 0.2 ha plot in summer season in Maharashtra and 9.5 t/ha in a 3 cent plot in Andhra Pradesh in India (Nigam, 2000).

<u>Trend in global groundnut area, production and productivity (1980-2012)</u>: Both, on a global and regional basis, groundnut showed positive trends in growth in area, yield and production (Table 2).

Region	Annual growth rate (%)		rate (%)	Remarks
Area Yield	Production			
World	0.99	1.72	2.73	
Asia	0.19	2.49	2.68	Limited opportunity for increasing area, gains in production will have to come through gains in yield.
Africa	2.47	1.22	3.72	Large scope to increase both area and yield.

**Table 2.** Annual growth rate in area, yield and production of groundnut during 1980-2012.

With 11.72 million ha area, 10.89 million t in-shell production and an average yield of 928.9 kg/ha in 2012, Africa contributed 47.6% to the global area and 26.4% to total global production of groundnut. Share of Asia in the same year was 47.0% in area (11.59 million ha) and 62.3% in production (25.69 million t). Average groundnut yield in Asia was 2216.8 kg/ha.

Starting 1990, the share of Asia in global groundnut area and production has shown a declining trend (share in area: 1990 = 65.86% and 2012 = 47.0%; share in production: 1990 = 70.57% and 2012 = 62.3%) whereas the reverse was true for Africa (share in area: 1990 = 27.72% and 2012 = 47.6%; share in production: 1990 = 19.34% and 2012 = 26.4%).

<u>Major countries with above world average groundnut productivity</u>: 34 countries reported above world average groundnut yield in 2012 (1675.9 kg/ha) but only countries of consequence in terms of area are China, USA, Argentina, Vietnam, Brazil, Egypt, Mexico and Turkey.

<u>Countries with below world average groundnut productivity</u>: Most countries in Africa and some in Asia and South America led by India, Nigeria, Sudan, Sierra Leone, Tanzania and Myanmar.

<u>Growing ecologies</u>: <u>Rainfed</u> – Sole crop, intercrop and mixed crop; <u>Irrigated</u> – Sole crop; <u>Residual</u> <u>moisture</u> (in rice fallows) – Sole crop.

(Single crop in a year in rainfed and residual soil moisture ecologies; two crops possible in irrigated ecology, if temperatures all year round are favourable for groundnut growth.)

<u>Subsistence farming</u>: Predominant in Asia and Africa; Characterised by low inputs, rainfed cultivation, smallholdings, manual operations using traditional tools, sole crop, mixed or intercropping, low productivity (700 to 1000 kg/ha).

<u>Commercial farming</u>: Prevalent in USA, Australia, Argentina, Brazil, China and South Africa; Characterized by high inputs, availability of irrigation, large holdings, mechanized or semimechanized cultivation, sole crop, high productivity (2 to 4 t/ha).

<u>Share of subsistence farming in total groundnut area</u>: More than 76% groundnut area covered by subsistence or low input rainfed farming globally.

# 2. Cultivated groundnut - Origin, spread and centers of diversity

<u>Groundnut plant</u>: Features of a groundnut plant are shown below (Figure 1); Main stem (upright or prostrate; 12-65 cm in length) develops from a terminal bud of the epicotyl and two cotyledonary laterals (can be prostrate, run along the ground or be upright) grow on opposite sides; Tetrafoliolate leaves, with leaflets on the main stem differing in shape and size from those on lateral branches.



Figure 1. A stylized groundnut plant.

<u>Growth habit</u>: Six classes as per groundnut descriptors (Figure 2); Commonly classified into three groups – Erect/Bunch, Semi-spreading/Semi-runner and Spreading/Runner types.



Figure 2. Growth habits of groundnut.

<u>Branching pattern</u>: Major two types – Alternate and Sequential, a third category of Irregulars also occurs (Figure 3); Presence or absence of flowers on main axis and branching pattern on laterals, although closely linked, are two independent events.



Figure 3. Branching patterns in groundnut.

Alternate branching: Absence of reproductive axes on main stem and presence of alternating pairs of vegetative and reproductive axes on the cotyledonary laterals and other primary branches; First two branches on laterals and primaries always vegetative, e.g., subsp. *hypogaea*.

Sequential branching: Presence of reproductive axes on main stem and presence of continuous series of reproductive axes on the cotyledonary laterals and other primary branches; First node on laterals and primaries always reproductive, e.g., subsp. *fastigiata*.

Irregular branching: Because of extensive inter-subspecific hybridization (i.e., between alternate and sequential branching types), irregular branching habit can arise – presence of flowers on the main stem accompanied with alternate branching on laterals and absence of flowers on main stem accompanied with sequential branching on laterals.

<u>Groundnut flower</u>: Flowers (Figure 4) are aerial but pod development is subterranean due to geotropic movement of the gynophores (pegs); Flowers are borne on inflorescences located in the axils of leaves; Flowering is sensitive to light, temperature and relative humidity; High temperatures and low levels of relative humidity reduce flowering, while temperatures between 22°C and 33°C and soil moisture of 40% are ideal for flowering; A light intensity greater than 45% of full sunlight is necessary for optimum development – flowering is stimulated by light received three days prior to opening of flowers; Generally, only one flower opens at each node at any given time at sunrise.



Figure 4. Longitudinal section of a groundnut flower.



Figure 5. A dissected groundnut flower: a) hypanthium, b) single and fused sepals, c) standard petal, d) wing petal, e) keel petal, f) stamens, and g) style and stigma.

A <u>flower</u> (Figure 5) consists of five petals, ten stamens and a pistil; Five <u>petals</u> are – one yellow to orange standard, two yellow to orange wings, and two petals fused to form a paler yellow keel; Of the ten monadelphous <u>stamens</u>, two are usually not fully developed; Stamens are fused together from one half to two thirds of their length; Eight fertile normally developed anthers consist of four globose, dorsifixed, uniloculate anthers alternating with four adnate, introse, oblong anthers; <u>Pistil</u> consists of an ovary, style and stigma; <u>Ovary</u> is superior, small and conical with a beak-shaped point at the tip and contains a single sessile carpel with 1 to 6 ovules; <u>Style</u> is glabrous throughout its length and covered with bristles near the club-shaped stigma and is enclosed in a filiform hypanthium; <u>Stigma</u> becomes receptive to pollen 24 hours before flower opening; Anthesis and pollination occur at sunrise with self-pollination taking place within the closed keel of the flower; About 40% of the flowers fail to begin pod development and another 40% abort before pod development.

<u>Groundnut pod and seed</u>: Pods (Figure 6) containing seeds are produced below ground; They vary in shape, size and texture and may contain up to five seeds per pod; Only after pods attain full size, the seeds inside start to develop; A seed consists of two large cotyledons (does not contain endosperm), a stem axis and leaf primordial (plumule), hypocotyl and primary root (radicle) and seed coat; Cotyledons comprise about 96% of the seed weight and are the major storage tissue for the developing seedlings.



Figure 6. Groundnut pod and seed components.

<u>Groundnut root</u>: Tap root system has many lateral roots; Roots can go as deep as 135 cm but are generally confined to 5-35 cm zone within a radius of 12-14 cm; Roots lack typical root hairs, instead have tuft of hairs produced in the axil of emerging lateral roots to provide site for nodulation.

<u>Center of origin</u>: Genus *Arachis* is naturally restricted to Argentina, Bolivia, Brazil, Paraguay and Uruguay in South America; No one is certain of the exact origin of cultivated groundnut; Most probably it originated in the region of eastern foothills of the Andes (southern Bolivia and northwestern Argentina).

<u>Centers of diversity</u>: Greatest genetic diversity in *Arachis* occurs in South America; Six recognized gene centers for cultivated groundnut in South America – (i) the Guarani region, (ii) Goias and Minas Gerais (Brazil), (iii) Rondonia and northwest Mato Grosso (Brazil), (iv) the eastern foothills of the Andes in Bolivia, (v) Peru, and (vi) Northeastern Brazil; Secondary center of diversity – Africa.

<u>Spread</u>: Portuguese carried in the late 15<sup>th</sup> century 2-seeded groundnut varieties from the east coast of South America (Brazil) to Africa, to the Malabar coast of southeastern India and possibly to the far east; Spaniards in the early 16<sup>th</sup> century took 3-seeded Peruvian types (including hirsuta types) to Indonesia, China up to Madagascar from the west coast of South America via the western Pacific; By the middle of 16<sup>th</sup> century, groundnut made its way to North America from Africa (through slave trade) as well as from the Caribbean islands, Central America and Mexico and was distributed worldwide; By the 19<sup>th</sup> century, groundnut became an important crop in West Africa, India, China and the USA; More than 114 countries now grow groundnut.

### 3. Wild relatives and their potential in groundnut improvement

<u>Family, tribe, subtribe and genus</u>: Family – Leguminoseae-Papilionoideae, Tribe – Aeschynomeneae, Subtribe – Stylosanthinae, Genus – *Arachis*; genus endemic to South America; About 80 described species reported in nine intrageneric taxonomic sections (Table 3); More than 1300 accessions of wild *Arachis* species collected; Several accessions within a species may show variation; Many species are still awaiting description.

<u>Taxonomic sections of Arachis</u>: Genus Arachis divided into nine taxonomic sections – Section Arachis (32 species described), Section Caulorrhizae (2 species described), Section Erectoides (14 species described), Section Extranervosae (10 species described), Section Heteranthae (6 species described), Section Procumbentes (9 species described), Section Rhizomatosae (4 species described), Section Trierectoides (2 species described) and Section Triseminatae (one species described); Except sections Arachis and Heteranthae, which contain both annual and perennial species, all other sections have perennial species (Table 3).

Section	Major identifiers	Life cycle	Chromosome number	Characteristic species and number of species
Arachis	Leaves tetrafoliolate, plants erect or decumbent, pegs near 45° angle of soil penetration	Annual or perennial	2n=20, 2n=4x=40	Arachis hypogaea L. (32)
Caulorrhizae	Leaves tetrafoliolate, stems with roots/root primordia at the nodes	Perennial	2n=20	Arachis repens Handro (2)
Erectoides	Leaves tetrafoliolate, plants erect or decumbent with flowers and pods grouped generally at the plant base, roots with enlarged laterals in most species, some pegs up to 1 m or longer	Perennial	2n=20	Arachis benthamii Handro (14)
I m or longerExtranervosaeLeaves tetrafoliolate, roots with enlargements (tubers?) of various sizes and shapes (but basically cylindrical), standard petal with red lines on the back side, all flowers 'normal' with an expanded corolla		Perennial	2n=20	Arachis prostrate Benth (10)

Table 3. Characteristics of taxonomic sections of genus Arachis.

Section	Major identifiers	Life cycle	Chromosome number	Characteristic species and number of species
Heteranthae	Leaves tetrafoliolate, root system with tap root but fibrous without enlargements, standard petals with red lines on front only or on both sides, flowers dimorphic, normal and open or very small and closed, small with corolla not exceeding the calyx	Annual or biannual	2n=20	Arachis dardani Krapov. and W.C. Greg. (= ambinervosae Krap. et Greg. nom. nud.) (6)
Procumbentes	Leaves tetrafoliolate, stems with roots occurring in internodes, pegs thickened, horizontal and in many cases long	Perennial	2n=20	Arachis rigonii Krapov. & W.C. Greg. (9)
Rhizomatosae	Leaves tetrafoliolate, plants with rhizomes	Perennial	2n=4x=40	<i>Arachis glabrata</i> Benth. (4)
Trierectoides	Leaves trifoliolate, hypocotyl tuberiform, plants erect, flowers and pods primarily at the base of the main stem, pegs very long, growing horizontal and superficial	Perennial	2n=20	Arachis guaranitica Chodat and Hassl. (2)
Triseminatae	Leaves tetrafoliolate, pod with 2 to 4 segments, lateral branches decumbent with flowers and pods along its length, standard petal with red lines on both sides, cotyledons with ribs on the upper surface (after plant emergence)	Perennial	2n=20	Arachis triseminata Krapov. & W.C. Greg. (1)

(Adapted from Stalker and Simpson, 1995; Bertioli et al., 2011)

<u>Cross compatibilities among sections of *Arachis*</u>: Wild *Arachis* species, a reservoir of several useful genes (diseases and insect pests resistance including genes for yield related traits and high oil content), need to be harnessed for improvement of cultivated groundnut; Interspecific hybridization is possible among most species within a section, whereas hybrids between species of different sections are difficult to produce; Further, intrasectional hybrids range from completely sterile to highly fertile, whereas intersectional hybrids are always sterile.

Fertile hybrids: Rhizomatosae x Erectoides.

<u>Sterile hybrids</u>: Rhizomatosae x Arachis, Erectoides x Arachis, Erectoides x Caulorrhizae, Erectoides x Heteranthae, Heteranthae x Extranervosae.

Less barriers to interspecific hybridization in section *Arachis* as the section is more recently evolved; All species in section *Arachis* compatible with *Arachis hypogaea*; However, hybrids of *Arachis grandulifera* Stalker and *Arachis hypogaea* can be obtained with the aid of embryo rescue and all other species can be used only as male parent; Intersectional hybrids involving section *Arachis* and *Rhizomatosae, Extranervosae, Procumbentes* and *Erectoides* have been successful at ICRISAT.

<u>Genomes</u>: Three genomes designated among section *Arachis* diploid species based on chromosome morphology and crossing relationships; Most diploid species in section *Arachis* have an A genome, *Arachis batizocoi* Krapov. and W.C. Gregory, *Arachis ipaënsis* and four more species of the section *Arachis* have a B genome and *Arachis grandulifera* Stalker has a D genome; Recent studies with FISH mapping of rDNA indicated presence of two more genomes in species earlier considered to belong to B genome – F (*Arachis benensis* and *Arachis trinitensis*) and K (*Arachis batizocoi, Arachis cruziana* and *Arachis krapovickasii*).

<u>Protocols for utilization of wild species</u>: Commonly two pathways: Triploid-Hexaploid pathway and Diploid-Tetraploid (Autotetraploid and Amphidiploid) pathway.

Triploid-Hexaploid Pathway
Arachis hypogaea (4x)
Diploid wild <i>Arachis</i> species of section <i>Arachis</i> A or B genome (2x)
↓ Triploid hybrid (3x, Sterile)
↓ Colchicine treatment Hexaploid (6x, Fertile) X <i>Arachis hypogaea</i> (4x)
Pentaploid (5x Mostly sterile) X Arachis hypogaea (4x)
↓ Selfing or
Population with plants at or near tetraploid level due to loss of chromosomes (Fertile)

Greater recombination between cultivated and wild *Arachis* species in the Triploid-Hexaploid pathway due to selfing at hexaploid level; Interspecific hybrids derived from *Arachis hypogaea* subsp. *fastigiata* var. *fastigiata* x *Arachis cardenasii* following selfing route have shown high levels of resistance to nematodes, early and late leaf spots, leaf hopper, corn earworm and southern corn rootworm including high yield indicating the occurrence of recombination between *Arachis hypogaea* and *Arachis cardenasii*; Recombination between *Arachis hypogaea* and wild *Arachis* 

species in backcrossing through pentaploid route is limited.



Quick introgression of genes (particularly simply inherited dominant genes) in the autotetraploid/amphidiploid route due to circumvention of sterility in triploid, hexaploid or pentaploid hybrids; Seed treatment with colchicine preferred over the same for vegetative cuttings due to higher success rate in doubling the chromosome numbers in the former; However, cytological confirmation necessary to confirm doubling of chromosome number in colchicine treatment; Interspecific hybrids derived from amphidiploid of A and B genome species and *Arachis hypogaea* more fertile than when either genome species used alone; However, the autotetraploid /amphidiploid pathways have disadvantage of polyploidy identification, low vigor in many cases, fewer seeds from hybridization and relatively restricted recombination.

Linkage drag in interspecific hybridization: Wild *Arachis* species harbor both useful genes (disease resistance, insect pest resistance, yield, high oil content, etc.) and several undesirable traits (long duration, poor pod and seed characteristics (shape, shell thickness, reticulation, catenate pods, small pod/seed size, etc.)); Along with desirable genes from wild *Arachis* species, many undesirable traits also get transferred due to tight linkage between them; While getting rid of linkage drag by backcrossing with *Arachis hypogaea*, the resistance to diseases and insect pests is also diluted; Use of molecular markers in selection in breeding populations may reduce the burden of linkage drag; Molecular marker based approaches, Genome Wide Introgression (GWI) and Advanced Backcross-QTL(AB-QTL), enable enhanced utilization of alleles from wild species.

<u>Reported resistance genes in wild species</u>: Table 4 below listing resistant sources in wild *Arachis* species is not exhaustive.

Specific trait	Arachis species	Accession numbers
Early leaf spot (ELS)	Arachis chacoense	PI 276325
	Arachis stenosperma	PI 33820
	Arachis glabrata	PI 262797
Late leaf spot (LLS)	Arachis chacoense	PI 276325
	Arachis cardenasii	PI 262141
	Arachis stenosperma	PI 338280
	Arachis glabrata	PI 262797
Rust	Arachis batizocoi	PI 298639, 338312
	Arachis duranensis	PI 219823
	Arachis spegazzinii	PI 263133
	Arachis correntina	PI 331194
	Arachis stenosperma	PI 338280
	Arachis cardenasii	PI 262141
	Arachis chacoense	PI 276235
	Arachis villosa	PI 210554
Aspergillus flavus (low in vitro seed	Arachis pusilla, Arachis chiquitana, Arachis triseminata,	
aflatoxin production)	Arachis auranensis, Arachis cardenasii	
Groundnut rosette	Arachis annressinila	ICC 8127 8945 14860
disease (GRD)	Arachis docora	
		100 14946
	Arachis diogoi	ICG 4983
	Arachis hoehnei	ICG 8190, 13232
	Arachis kretschmeri	ICG 8191, 8216, 11558, 13224
	Arachis kuhlmannii	ICG 13225, 14862, 14875
	Arachis pintoi	ICG 13222, 14855, 14856, 14888
	Arachis stenosperma	ICG 13171, 13173, 13187, 13210, 14872
	Arachis villosa	ICG 13168
Groundnut rosette virus (GRV) and Groundnut rosette assistor virus (GRAV)	Arachis chacoense	
Peanut bud necrosis	Arachis duranensis	ICG 8199,8956, 11552, 11553, 11555
disease (PBND)	Arachis correntina	ICG 8132
	Arachis monticola	ICG 8189

**Table 4.** Sources of resistance to diseases, insect pests and nematodes and high oil content in wild *Arachis* species.

Specific trait	Arachis species	Accession numbers
Peanut bud necrosis	Arachis cardenasii	ICG 11564, 13164, 13165
	Arachis villosa	ICG 8144, 13168
Tomato spotted wilt	Arachis glabrata	PI 262794, 33864
virus (15wv)	Arachis diogoi	PI 468141
	Arachis helodes	PI 468144
Peanut stripe virus	Arachis glabrata	PI 262801, 262794
(PStV)	Arachis villosa	PI 210555-1
	Arachis correntina	GKP 9530-31
	Arachis diogoi	PI 468141, 468142
	Arachis helodes	PI 468144
Tobacco streak virus (TSV)	Arachis duranensis	ICG 8139, 8195, 8200, 8203, 8205, 11550
	Arachis villosa	ICG 8144
	Arachis stenosperma	ICG 13210
Defoliators (Leaf miner, tobacco caterpillar, cotton bollworm)	Arachis cardenasii, Arachis duranensis, Arachis kempff- mercadoi, Arachis monticola, Arachis stenosperma, Arachis paraguariensis, Arachis pusilla, Arachis triseminata, Arachis ipaensis, Arachis appressipila, Arachis villosa, Arachis batizocoi, Arachis correntina	
Root-knot nematode	Arachis batizocoi, Arachis cardenasii, Arachis diogoi , Arachis duranensis, Arachis helodes, Arachis villosa	
High oil content	Arachis rigoni	WH 10026, 4347, 4377, 10034, 4330, 10025, 4376, 4367 (>57% oil)
	Arachis chacoense, Arachis monticola, Arachis villosa, Arachis cryptopotamica, Arachis oteroi, Arachis chiquitana	

Accessions of wild *Arachis* species in section *Arachis* resistant to cylindrocladium black rot, various nematode species and insect pests (aphids, jassids, thrips, Helicoverpa, etc.) are also reported.

<u>Centers maintaining wild Arachis collections</u>: <u>Argentina</u> – Instituto Botànico del Universidad Nacional de Nordeste (IBONE) (472 accessions); <u>Australia</u> – Australian Tropical Crops and Forages Genetic Resources Centre (65 accessions); <u>Brazil</u> – Embrapa and Recursos Geneticos e Biotecnologia (CENARGEN), Instituto Agronomico de Campinas (1220 accessions); <u>China</u> – Oil Crops Research Institute, Wuhan (246 accessions); <u>Colombia</u> – Centro Internacional de Agricultura Tropical (CIAT) (243 accessions) and Centro de Investigaciones de Nataima, Instituto Colombiano Agropecuario (ICA) (225 accessions); <u>India</u> – International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Patancheru, Telangana (477 accessions); National Bureau of Plant Genetic Resources (NBPGR), ICAR, New Delhi (112); ICAR – Directorate of Groundnut Research (ICAR-DGR), Junagadh, Gujarat (106 accessions); <u>USA</u> – USDA, Griffin, Georgia (607 accessions); Texas A&M University, Stephenville, Texas (1200 accessions); NC State University, Raleigh, North Carolina (406).

*Ex-situ* conservation of wild *Arachis* species: Maintained through both seed multiplication and vegetative cuttings but preservation for species producing few seeds, in general, difficult; Best maintained in large concrete cylinders (75 cm high, 90 cm inner diameter and 5 cm wall thickness) set sufficient apart to stop vines reaching to other cylinders to avoid contamination either in open or preferably inside a glass/screen house; Potting medium sterilized 3 soil : 2 sand : 1 Farm yard manure (FYM) mixture; A cylinder can accommodate 5-6 plants.

<u>Vegetative propagation</u>: All *Arachis* species lend themselves to vegetative propagation, which can be adopted to: i) Multiply species which can be propagated only vegetatively, ii) Multiply limited materials such as  $F_1s$ , iii) Maintain the same plant/genotype generation after generation, and iv) Obtain large number of seeds of the same genotype for experimental purposes.

Procedure: Plant age for taking cuttings – best soon after flowering (ensure stem has sufficient food reserve); Length and preparation of cutting – 10-15 cm or 3-4 internodes long, give a slanting cut at the base of the node, remove all leaves except the first fully opened tetrafolioate and treat the cut end with rooting hormone (Seradix B); Potting and rooting – transplant treated cuttings (4-5 cuttings together) in small plastic pots filled with sterilized sand, place the pots in humidity chambers, control diseases and insect pests if observed, remove flowers and pegs, if they appear; New leaves start appearing soon and the roots appear in 30-40 days under favourable conditions; Condition/harden rooted cuttings before transferring them to bigger pots in the open.

<u>Wild species released as fodder variety</u>: *Arachis glabrata* (section *Rhizomatosae*) – Arb (PI 118457), Arblick (PI 261839), Florigraze (GS 1, 1978) and Arbrook (PI 261817, 1986) in Florida, USA; *Arachis pintoi* (section *Caulorrhizae*) – Amarillo (CPI 58113, 1989) in Tropical Australia, Mani Forrajero and Perenne (1992) in Colombia and Pico Bonito (1993) in Honduras (CIAT 17434); *Arachis* sp. (section *Procumbentes*) – Pantanal (PI 446898) in Florida, USA; *Arachis glabrata* propagated by vegetative cuttings and *Arachis pintoi* by seeds; Maximum dry matter yield recorded is >16 t/ha in Florida; Because of higher levels of protein and resistance to diseases and insect pests, these fodder varieties are used as an alternative to alfalfa in the USA.

<u>Nutritive value of forage Arachis</u>: Nutritive value of Arachis glabrata cv. Florigraze and Arachis pintoi (CIAT 17434) is higher than that of most tropical legumes of commercial importance; They have high levels of digestible energy (60-70% dry matter digestibility) and fermentable nitrogen (13-25% crude protein) and relatively low levels of condensed tannins.

<u>Wild species as a cover and ornamental crop</u>: *Arachis pintoi* and *Arachis repens* used as cover crop for smothering weeds in plantation crops (coffee, banana, oil palm, etc.) and for lawns as ornamental crop because of their intense green foliage color and frequent brilliant yellow flowering.

Improved germplasm/cultivars with wild species genomes released: Some of the improved germplasm /cultivars having genes from wild *Arachis* species are – ICGV 86699, 87165, 99001, 99003, 99004 and 99005, ICGV-SM 85048 and 86715 (ICRISAT, India and ICRIST, Malawi), GPBD 4 (UAS, Dharwad, India), Spancross, Tamnut 74, TxAG 7, COAN, NemaTAM and Tifguard (USA).

<u>Synthetic allotetraploids</u>: Tetraploid groundnuts (amphidiploids and autotetraploids) developed by combining putative genome donors as well as many other closely related A and B genome species; Used to access diploid gene pool to mine for disease resistance, drought tolerance and other traits.

# 4. Cultivated groundnut - Taxonomy and reproductive biology

<u>Putative ancestors</u>: Domesticated species is believed to have originated from a single hybridization event approximately 3500 years ago between putative ancestors (*Arachis duranensis* (A genome), the female progenitor and *Arachis ipaënsis* (B genome), the male progenitor), which was followed by tetraploidization; Subsequently, very little or no introgression from related diploid species occurred, which resulted in only limited DNA polymorphism in cultivated groundnut; However, ample DNA polymorphism is observed within AA and BB genome species; Cultivated groundnut has narrow genetic base due to its allotetraploid origin; However, there is abundant morphological variability present among the varieties in the cultivated species.

<u>Ploidy level</u>: Cultivated groundnut is a segmental allotetraploid consisting of two genomes, A and B, each originating from a different ancestor, but behaves like a diploid; Almost all chromosome pairing during meiosis is bivalent.

### Chromosome number: 2n=4x=40 (x=10; AABB genomes)

<u>Taxonomy</u>: Cultivated groundnut, *Arachis hypogaea*, an annual herb of indeterminate growth habit, is divided into the following two subspecies (subsp.):

- 1. Subsp. *hypogaea*: Key distinguishing features absence of flowers on the central or main axis (stem) and regular alternation of vegetative and reproductive branches on the laterals (alternate ramification), long life cycle.
- 2. Subsp. *fastigiata*: Key distinguishing features presence of flowers on the central or main axis (stem) and no specific order of vegetative and reproductive branches on the laterals (sequential ramification), shorter life cycle.

Subspecies *hypogaea* is divided into two botanical varieties (var.):

- i. Var. *hypogaea*: Key distinguishing features leaflets with glabrous dorsal surface or with some hair along the midrib, short central or main axis, prostrate to erect growth habit, simple inflorescence, 2–3 seeded pods.
- ii. Var. *hirsuta*: Key distinguishing features leaflets with entire dorsal surface hairy (1–2 mm), long central or main axis, prostrate growth habit, 2–4 seeded pods.

Subspecies *fastigiata* is divided into four botanical varieties as follows:

- i. Var. *fastigiata*: Key distinguishing features Leaflets with glabrous dorsal surface and hair only on the midrib, 3-5 seeded pods with smooth or lightly marked reticulation without highlighting of the longitudinal ribs, reproductive branches mostly short and thin, little branched, curved branches, erect growth habit, simple inflorescence.
- ii. Var. *peruviana*: Key distinguishing features Leaflets with glabrous dorsal surface and hair only on the midrib, pods with very marked reticulation and with prominent longitudinal ribs, long and strong reproductive branches (5–10 cm) with strong central axis and lateral branches.

- iii. Var. *aequatoriana*: Key distinguishing features Leaflets with entire dorsal surface hairy (1-2 mm), long reproductive branches, mainly the lateral branches, central axis mostly with short inflorescence and reproductive branches, deep pod reticulation, purple stems, more branched, erect growth habit.
- iv. Var. *vulgaris*: Key distinguishing features Pods mostly 2–seeded, bunched fruits pointing to the base of the plant, erect growth habit, more branched, compound inflorescence.

Var. *hypogaea* is also known as Virginia type (large-seeded) or Runner type (small-seeded), var. *hirsuta* as Peruvian runner, var. *fastigiata* as Valencia type and var. *vulgaris* as Spanish type; Spanish types are likely to have originated due to hybridization between Virginia x Valencia types.

The terms, Virginia, Runner, Valencia and Spanish, used both in botanical sense, defined above, and as market types, which are described below.

U.S. peanut grades:

Seed counts per ounce (28.35 g)

In-shell

- 1. Virginia Jumbo In-shell: 9-11
- 2. Virginia Fancy In-shell: 11-13

#### Kernels

- 1. Virginia Extra Large: 28-32 elongated
- 2. Virginia Medium: 38-42 elongated
- 3. Virginia # 1: 45-55 elongated
- 4. Virginia # 2: Virginia splits
- 5. Runner Jumbo: 38-42
- 6. Runner Medium: 40-50
- 7. Runner # 1: 60-70
- 8. Runner Splits: Splits of Runner kernels
- 9. Spanish Jumbo: 60-70
- 10. Spanish # 1: 70-80
- 11. Spanish Splits: Splits of Spanish kernels
- (Adapted from Anonymous, 1988.)

#### Must ride on screen

#### Kernels

- 1. Virginia Extra Large: 20/64 x 1 inches or 7.94 x 25.40 mm
- 2. Virginia Medium: 18/64 x 1 inches or 7.14 x 25.40 mm
- 3. Virginia # 1: 15/64 x 1 inches or 5.95 x 25.40 mm
- 4. Virginia # 2: 17/64 inches round or 6.75 mm round
- 5. Virginia Splits: 20/64 inches round or 7.94 mm round
- 6. Runner # 1: 16/64 x 3/4 inches or 6.35 x 19.05 mm
- 7. Runner # 2: 17/64 round or 6.75 round
- 8. Runner Splits: 17/64 round or 6.75 mm round
- 9. Spanish # 1: 15/64 x 3/4 inches or 5.94 x 19.05 mm
- 10. Spanish # 2: 16/64 round inches or 6.35 mm round
- 11. Spanish Splits: 16/64 round or 6.35 mm round
- (Adapted from Davidson Jr et al., 1982)

Uses of different types in the USA:

Virginia type: Largest kernels attractive for direct consumption, roasted in-shell, kernels as salted peanuts.

Runner type: Mainly used in peanut butter.

Valencia type: 3-4 seeded pods, sweet in taste, freshly harvested pods used as boiled peanuts, dried pods roasted in-shell.

Spanish type: Smaller kernels, used primarily in candy making, as salted peanuts and in peanut butter.

<u>HPS groundnut in India</u>: Groundnut for edible purposes in trade circles in India is called 'Hand Picked and Selected (HPS)' groundnut as it involves a large measure of hand sorting to ensure high quality; Kernels classified into four classes based on counts per ounce (number of kernels per 28.35 g weight): Small 60-80 counts, Medium 40-60 counts, Large 30-40 counts, and Very Large 20-30 counts; Usually counts have a range of ten for Small kernels, a range of five for Medium and Large kernels and of two for Very Large kernels; Similarly, a range of two is preferred for nuts-in-shell.

<u>Breeding behaviour including cross pollination</u>: Regarded a highly self-pollinated crop with less than 1% cross pollination; However, cross pollination can reach as high as 10% at locations and in seasons where bee activity is high.

<u>Hybridization in groundnut</u>: Depending upon the breeding objective, parents for hybridization should be selected carefully. Normally, female parent should be the locally adaptive variety requiring improvement. Male parent should be the source of desired gene(s). Hybridization can be carried out both in the field and in glass/screen house. Field crossing nursery should be easily accessible and well maintained with intensive protection against diseases and insect pests. It should be near the water source to enable light irrigation every evening during the duration of hybridization program. Parental rows in the nursery should be spaced out to allow emasculation and pollination operations without disturbing the plants. The ratio of female and male rows/plants is variable but enough male flowers should be available in the morning for pollination.



Figure 7. Groundnut hybridization in the field.

Emasculation and pollination: Bud to open next day is selected for emasculation (removal of anthers from buds before their dehiscence to avoid self-pollination), by afternoon or late evening, such buds are well developed and big enough with elongated hypanthium to be handled easily during emasculation; Only one bud is emasculated at each node and all other developing buds are removed from the node causing little or no injury; All the 10 stamens (four stamens with globose, dorsifixed and monothecous anthers and other four with adnate, introse, oblong anthers plus two sterile staminodes) are to be completely removed during emasculation without causing any injury to style and stigma; Pollination (transference of pollen grains from the anthers of one flower to the stigma of another flower) is done next morning soon after sunrise when the buds open into flowers; For pollination, a healthy flower from a predetermined male parent is removed by breaking hypanthium and calyx, standard and wing petals are detached and the keel petal gently pressed between thumb and index finger to squeeze the sticky pollen mass out from the anthers; Sticky lump of pollen is deposited on the tip of the stigma of the emasculated flower; Sometimes low night temperature may delay the dehiscence of anthers and the time of pollination; In delayed pollination, the success rate of hybridization goes down; Stigmatic surface contains enzymes that promote pollen adhesion and within 8 h after anthesis these enzymes apparently degrade; Fertilization occurs about 6 h after pollination; Flower petals wither within 24 h; Fertilization of the egg activates the growth and elongation of the intercalary meristem located at the base of the ovary; Peg with positive geotropic behaviour and a length of up to 15 cm bears the ovary with the fertilized ovules at it tips; Once peg enters the soil and penetrates to a depth of 2-4 cm, the tip becomes diageotropic and the ovary develops into a pod; Pod first attains full size and then the kernel inside starts developing; Under normal growing conditions of temperature, moisture and nutrition, mature seeds formed 40-60 days after fertilization.

Hybridization in groundnut involves working during late evening and early morning hours, which many persons find difficult to observe. By manipulating temperature and day length (extending day length using artificial lights) separately for male and female parents, the bud elongation and development can be achieved in the morning hours to simultaneously allow emasculation and pollination in the morning hours.

<u>Success of hybridization</u>: Successful pollination – indicated by emergence of a peg from the axil of the leaf at the node of emasculated flower after 4–6 days after fertilization under normal growing conditions, the peg emergence may take longer under low temperatures; Unsuccessful pollination – no peg emergence even up to 2–3 weeks after pollination; Success rate, the per cent success of pollination, depends on the skill of operator, timeliness of operations, environmental conditions during hybridization process and the varieties involved in hybridization.

<u>Environmental factors for high success rate</u>: High humidity at the time of pollination and timely pollination soon after sunrise ensure high success. Light irrigation in the evening after emasculation keeps the soil in the field crossing nursery wet in the morning, thus raising the humidity at the time of pollination. If all precautions are taken in hybridization, a success rate as high as 80-90% under screen/glasshouse conditions and 70-80% under field conditions can be obtained.

<u>Confirmation of hybridity</u>: Grow  $F_1$  plants and their parents side by side; Compare carefully each  $F_1$  plants for various morphological traits with male and female parents to confirm their hybridity; If the  $F_1$  plant is exactly similar to the female parent, most likely it is a self, remove all the selfs; If in doubt, compare pod and seed characters after harvest to further confirm hybridity; If doubt still persists, study  $F_2$  generation of such plants for segregation, in the absence of segregation, the plant should be rejected as self.

<u>Genetic markers</u>: These can be used to confirm hybridity of  $F_1$  plants at seedling stage, however, marker traits should have complete penetrance and expressivity at seedling stage and absence of pleiotropism or linkage with sterility; Dominant marker traits – purple stem and leaflet veins, krinkle leaf, narrow leaf, yellow petal, etc.; Recessive marker traits – apetiolated leaf, corduroy leaf, small leaf, imparipinnate leaf, foliaceous stipules, miniature plant, white flower, etc.

### 5. Nutritional quality of groundnut seed and haulm

Groundnut can contribute significantly towards reduction in protein-energy and micronutrients malnutrition; 100 g (3.5 oz.) groundnut seed contributes 585 calories.

Average composition of different parts of pods/seeds:

Shell: 60% crude fiber, 25% cellulose, 8% water, 6% crude protein, 2% ash, 1% fat.

Testa (seed coat or skin): 35% nitrogen-free extracts, 12% fat, 11% ash, 9% water.

<u>Germ (heart)</u>: 42% fat, 20% carbohydrates, 4% nitrogen, 3% ash, 2% crude fiber, 1% or less Ca, Mg, K, P.

<u>Cotyledons</u>: 48% fat, 26% protein, 17% carbohydrates, 2% fiber, 2% ash, 1% or less vitamin E, niacin, folacin, Ca, Mg, Zn, Fe, riboflavin, thiamine, K.

<u>Nutritional value of postharvest haulms</u>: 85-93% dry matter, 6-20% crude protein, 2-13% digestible protein, 47-56% total digestible nutrients (TDN), 8-11 MJ/kg metabolizable energy (ME), 1-3% fat, 42-47% carbohydrate, 22-38% crude fiber, 9-17% minerals, 0.3-0.7% P, 1.5-2.5% Ca.

<u>Oil quality parameters</u>: <u>Iodine Value (IV)</u> = (% oleic acid x 0.8601 + % linoleic acid x 1.7321 + % eicosenoic acid x 0.7854); <u>Oleic (O)/linoleic (L) acid ratio</u> = (% Oleic acid / % linoleic acid); <u>Total Saturated Fatty Acids (TSF) (%)</u> = (% palmitic acid + % stearic acid + % arachidic acid + % behenic acid + % lignoceric acid); <u>Polyunsaturated (P)/saturated (S) acid ratio</u> = (% linoleic acid /% TSF); <u>Total Long Chain Fatty Acids (TLCSF)</u> = (% arachidic acid + % behenic acid + % lignoceric acid).

<u>Oil content</u>: In general, Spanish types have more oil content than other botanical types; 44–50% oil content most common in cultivars; Genotypes with lower than 44% and higher than 50% oil content also available; Recently oil content as high as 60% reported in new breeding lines at ICRISAT and in China.

<u>Oil composition</u>: Saturated fat – about 15%, monounsaturated fat – about 50% and polyunsaturated fat about 34%; Palmitic (16:0, saturated fatty acid) – 7-12%, oleic (18:1, monounsaturated fatty acid) – 40-50% and linoleic (18:2, polyunsaturated fatty acid) – 25-35%, these acids account for approximately 90% of total fatty acids; Longer shelf life than most of the vegetable oils due to high oleic/linoleic ratio (0.76 to 5.5); Cultivars with higher oleic/linoleic acid ratio ( $\approx$ 40, e.g. SunOleic 95R, SunOleic 97R) available in the USA; Most suited for deep frying because of its higher smoking point; Tocopherol (approx. 0.9 mg/g oil), an antioxidant in the oil, prevents development of rancidity.

Oil content and composition influenced by environment, season and location effects, growing conditions, stage of maturity and genotypes.

<u>Protein content and composition</u>: Protein content – 22-30%; Major amino acids present – glutamic acid, aspartic acid and arginine; Deficient in lysine, methionine, threonine, isoleucine and valine essential amino acids; Protein content and composition influenced by environment, season and location effects, growing conditions, stage of maturity and genotypes; 25 g protein contains –

tryptophan 0.2445 g, threonine 0.85 g, isoleucine 0.882 g, leucine 1.627 g, lysine 0.901 g, methionine 0.308 g, cystine 0.322 g, phenylalanine 1.300 g, tyrosine 1.020 g, valine 1.052 g, arginine 3.001 g, histidine 0.634 g, alanine 0.997 g, aspartic acid 3.060 g, glutamic acid 5.243 g, glycine 1.512 g, proline 1.107 g and serine 1.236 g.

<u>Carbohydrates</u>: Content – 20%; Contains water soluble carbohydrates (monosaccharides, disaccharides) and oligosaccharides (including starch, raffinose and stachyose); Sucrose is a major sugar in seeds followed by stachyose and raffinose.

<u>Vitamins</u>: Excellent source of E, K and B group (thiamine (B1), niacin (B3), pantothenic acid (B5), B6 and folate (B9)) vitamins; One of the richest source of thiamin (B1) in plants; Lacks vitamin A and C.

Minerals: Ca, Mg, P, Na, K, Fe, Cu, Zn and Mn; Good source for K, P and Mg, more K than Na.

Other desirable chemicals: Tocopherol, Resveratrol, Beta-sitosterol and Coenzyme Q 10.

<u>Nutrient contents in one ounce of groundnut</u>: Table 5 gives the nutrient contents in one ounce of groundnut kernels.

Nutrient	Amount	% Daily requirement	Functions / Remarks
Calories	161.0	n/a	Energy rich food due to its fat content; A very high proportion of unsaturated fats and high satiety value make groundnut part of a healthy diet.
Protein	7.3 g	14.2%	A powerhouse of less expensive vegetable protein.
Total carbohydrates	4.6 g	1.5%	Good for diabetic diets due to its low Glycaemic Index (a measure of the rate at which carbohydrates from a particular food breakdown and release glucose in blood stream).
Dietary fiber	2.4 g	9.4%	Reduces risk of some types of cancer, helps control blood sugar levels and may help reduce the levels of cholesterol in blood.
Total fat	14.0 g	21.8%	Concentrated source of energy, provides essential fatty acids, carries fat soluble vitamins such as A, D and E and helps maintain healthy skin; Suitable for Indian style of cooking due to its high smoking point (240°C).

**Table 5.** Nutrient contents in one ounce (28.35 g) of raw groundnut kernels(USDA Database for Standard Reference)

Nutrient	Amount	% Daily requirement	Functions / Remarks
Saturated fat	1.9 g	9.5%	A low proportion of saturated fat (bad fat); Saturated fat intake should be less than 10% of the total daily intake of calories.
Monounsaturated fat	6.9 g	n/a	Monounsaturated fats help to remove cholesterol including LDL cholesterol from the blood, thus giving protection from heart attack.
Polyunsaturated fat	4.4 g	n/a	Along with monosaturated fats, polyunsaturated fats are healthy and necessary for the healthy body.
Vitamin E	2.4 mg AT	17.5%	Vital antioxidant which protects Vitamin A and the body's cells and tissues from damage; Important for the immune system and might aid in the prevention of tumor growth; Plays a role in preventing coronary heart disease.
Folate	68 mcg	16.5%	Important for the development of new cells in the body, particularly during growth and pregnancy; Helps to prevent birth defects.
Niacin	3.26 mg	16.3%	Functioning in more than 50 of the body processes, niacin is primarily important in the release of energy from the food that we eat as well as in maintenance of healthy skin, the nervous system and the digestive tract.
Thiamin (B <sub>1</sub> )	0.18 mg	12%	Needed to ensure normal functioning of the nervous system, appetite and digestion.
Riboflavin (B <sub>2</sub> )	0.04 mg	21.3%	Releases energy from the food we eat, helps skin stay healthy and assists in the normal functioning of the eye.
Vitamin B <sub>6</sub>	0.10 mg	5.7%	Makes and breaks down proteins in the body and makes red blood cells used to transport oxygen in the body.

Nutrient	Amount	% Daily requirement	Functions / Remarks
Zinc	0.93 mg	5.9%	Aids in the formation of protein, wound healing, blood formation, taste perception, appetite, night vision and general growth and maintenance of all tissues.
Copper	0.32 mg	15.2%	Important in the formation of haemoglobin, health of bones, blood vessels and nerves.
Selenium	2.0 mcg	2.8%	A trace element required in small quantities for normal functioning of the immune system.
Magnesium	48 mg	12.5%	Important in the building of bones and teeth, creation of protein, transmission of nerve impulses and maintenance of body temperature.
Phosphorus	107 mg	10.6%	Component of all soft tissues that are fundamental to growth, maintenance and repairs of bones and teeth.
Potassium	200 mg	5.3%	Needed to ensure water balance in the body and in the creation of protein; Helps in release of energy from nutrients and aids in nerve impulse transmission.
Calcium	26 mg	3.5%	Needed for development and maintenance of healthy bones and teeth.
Sodium	5 mg	0.22%	Naturally low sodium food.
Iron	1.3 mg	8.1%	Aids in transport and distribution of oxygen in the body's cells.
Boron	1.0 mg	100%	Major factor in the metabolization of calcium in the body and plays significant role in development and maintenance of strong and healthy bones.
Cholesterol	0.0 mg		Free from cholesterol.
Arginine	0.88 g	n/a	Improves wound healing and immunity.
Total Phytosterols	62.4 mg	n/a	These phytochemicals help to prevent diseases and enhance health.

Nutrient	Amount	% Daily requirement	Functions / Remarks
Resveratrol	73 mcg/g without skin	n/a	Ounce for ounce groundnuts have about half of the amount of resveratrol in wine (160 mcg/g); Resveratrol, a phytochemical, has a possible role in reducing cancer and can inhibit build- up of platelets in blood vessels; A potent antioxidant which can reduce the oxidation of LDL cholesterol.
Beta-sitosterol	18.4 mg	n/a	Has anticancer properties and prevents cholesterol uptake.

<u>Allergens</u>: About 0.6% of the population in the USA is allergic to groundnut protein; Seed storage proteins Ara h1, Ara h2 (most important) and Ara h3 cause allergic reaction; PI 261924 and PI 338386 lines have low allergen; No breeding program is addressing the issue of peanut allergy through genetic manipulation; However, research efforts are in progress to develop vaccines to peanut allergens; Refined groundnut oil is allergen free as it does not contain any protein; Dry roasting enhances allergic properties of the proteins in groundnut seeds.

<u>Taste and flavor</u>: Flavor quality – sum of the effects of genetics, production and handling, storage and processing factors; Can be fully perceived by sensory evaluation methods; Gas chromatographic methods used to quantify volatile compounds that contribute to flavor; Carbohydrates and other carbonyls react with amino acids to form flavor compounds during roasting; Roasted flavor is an inherited trait.

# 6. Genetic resources of cultivated groundnut

<u>Accessions</u>: World collection assembled from other agencies with an accession number.

Landraces: Non-uniform material (unselected) collected from farmers' fields or purchased in local markets.

<u>Farmers' variety</u>: A variety that has been traditionally cultivated and evolved by the farmers in their fields.

<u>Breeding lines</u>: Material developed by breeders but not released as cultivars.

<u>Cultivars</u>: A plant or grouping of plants selected for desirable characteristics that can be maintained by propagation and cultivated by farmers.

<u>Genetic stocks</u>: Genotypes identified by special features or sources of resistance to biotic and/or abiotic stresses.

<u>Wild species</u>: Species belonging to the genus of cultivated type and occurring in wild; May or may not be cross compatible with cultivated type.

#### Gene pools in Arachis:

<u>Primary gene pool</u>: Includes landraces, cultivars and breeding lines of cultivated groundnut including *Arachis monticola*.

<u>Secondary and tertiary gene pool</u>: Includes diploid species that are cross compatible with *Arachis hypogaea* and belong to section *Arachis and Arachis monticola*.

<u>Tertiary and fourth gene pool</u>: Consists of species in section *Procumbensae* that can share genes with *Arachis hypogaea* on overcoming postzygotic barriers.

<u>Fourth and fifth gene pool</u>: Consists of the rest of the species that are cross incompatible or weekly cross compatible with section *Arachis*.

<u>Genetic resources of cultivated groundnut</u>: ICRISAT holds the largest collection of 15,445 accessions representing 95 countries and unknown sources of cultivated groundnut in its gene bank with many duplicates in this collection. This germplasm is freely available on request to scientific community and others from ICRISAT after completing certain formalities.

<u>Range of variability</u>: In spite of limited polymorphism at DNA level, there exists tremendous variability for various traits at the morphological level in cultivated groundnut (Table 6).

Character	Minimum	Maximum	Intermediate(s)
Growth habit	Erect	Procumbent	Decumbent
Branching pattern	Sequential	Alternate	Irregular
Stem pigmentation	Absent	Present	
Stem hairiness	Glabrous	Woolly	Hairy, Very hairy
Reproductive branch length	1 cm	10 cm	Continuous
Number of flowers/inflorescence	1	5	2, 3, 4
Peg color	Absent	Present	
Standard petal color	Yellow	Garnet	Lemon yellow, Light orange, Orange, Dark orange
Standard petal markings	Yellow	Garnet	Lemon yellow, Light orange, Orange, Dark orange
Leaf color	Yellowish green	Dark green	Light green, Green, Bottle green
Leaflet length	17 mm	94 mm	Continuous
Leaflet width	7 mm	52 mm	Continuous
Leaflet length/width ratio	1	6	Continuous

**Table 6.** Range of variability in cultivated groundnut observed at ICRISAT, Patancheru, India.

Character	Minimum	Maximum	Intermediate(s)
Leaflet shape	Cuneate	Lanceolate	Obcuneate, Elliptic
Hairiness of leaflet	Subglabrous	Profuse and long	Scarce and short, Scarce and long, Profuse and short
Number of seeds/pod	1	5	2, 3. 4
Pod beak	Absent	Very prominent	Slight, Moderate, Prominent
Pod constriction	Absent	Very deep	Slight, Moderate,
			Deep
Pod length	14 mm	65 mm	Continuous
Pod width	7 mm	20 mm	Continuous
Seed color pattern	One	Variegated	
Seed color	Off white	Dark purple	Yellow, Shades of tan, Rose, Shades of red, Grey-orange, Shades of purple
Seed length	4 mm	23 mm	Continuous
Seed width	5 mm	13 mm	Continuous
100-seed weight	14 g	130 g	Continuous
Days to emergence	4	18	Continuous
Days to 50% flowering	17	54	Continuous
Days to maturity	75	>155	Continuous
Fresh seed dormancy	0 days	>66 days	Continuous
Oil content	31.8%	55.0%	Continuous
Protein content	15.5%	34.2%	Continuous

<u>Resistant sources</u>: Table 7 below listing resistant sources in cultivated groundnut is not exhaustive. For additional information on resistant sources, literature should be consulted. However, it is important to evaluate the reported sources of desirable traits under local conditions and confirm their desirability before their use in a breeding program.

Specific trait	ICRISAT		Texas A & M and other US and non-US institutions	
Specific trait	Number screened	Number identified	Number screened	Number identified
	Dise	ease resistance		
Early leaf spot (ELS)			2500	28
Late leaf spot (LLS)	9400	76	2500	40
Rust	9400	141	1500	12
Web blotch			50	35
Peanut bud necrosis virus (PBNV)	7400	23		
Peanut mottle virus (PMV)	1800	2		
Groundnut rosette disease (GRD) <sup>1</sup>	12,500	150		
Tomato spotted wilt virus (TSWV) <sup>2</sup>			300	3
Peanut stripe virus (PStV) <sup>3</sup>	10,000	0		
Tobacco steak virus (TSV) <sup>4</sup>	150	0		
Indian peanut clump virus	9000	0		
Bacterial wilt <sup>5</sup>	6000	80		
Aspergillus flavus	582	17		
Pod rot	3222	24		
Sclerotinia blight			4100 + 96 <sup>10</sup>	1 + 39
Cylindrocladium black rot (CBR) <sup>6</sup>			1200	A few Virginia and several Spanish types
Sclerotium stem rot <sup>7</sup>			286	4

Table 7. Number of sources resistant to biotic and abiotic stresses
and those with other desirable traits.

Specific trait	ICRISAT		Texas A & M and other US and non-US institutions	
Specific trait	Number screened	Number identified	Number screened	Number identified
	Insect	t pest resistanc	e	
Thrips	5000	14		
Jassids	6500	30		
Termites	520	20		
Aphids	520	20		
Leaf miner	930	18		
Root knot nematode			2331	25
Multiple resistance	9400	85	2500	12
Abiotic stresses				
Drought tolerance	742	38		
Heat tolerance <sup>8</sup>			26	6
Salinity tolerance <sup>9</sup>			127	11
Other desirable traits				
High N fixation	342	4		
Earliness	15,000	21		
High oil	8868	44		
High protein	8868	51		
<sup>1</sup> Tested in Malawi, Nigeria and Mali; <sup>2</sup> Tested at University of Georgia, Georgia; <sup>3</sup> Tested at Muneng, Indonesia; <sup>4</sup> Tested at NBPGR Regional Station, ICAR, Rajendranagar, Hyderabad; <sup>5</sup> Tested in China and Indonesia; <sup>6</sup> Tested at NC State University, North Carolina; <sup>7</sup> Screened at Directorate of Groundnut Research, Junagadh; <sup>8</sup> Screened at University of Reading, UK; <sup>9</sup> Screened at Directorate of Groundnut Research, Junagadh; <sup>10</sup> From US peanut mini-core collection.				

(Adapted from Singh and Simpson, 1994, in The Groundnut Crop: A scientific basis for improvement. Ed. J Smartt, Chapman and Hall, London and other sources)

<u>Global composite collection</u>: Collection of 1000 diverse groundnut accessions jointly formulated by ICRISAT and EMBRAPA, includes 184 accessions from mini core subset, another 184 mini core comparator, 110 from Asia core and mini core, 408 elite germplasm and cultivars including trait specific accessions and 114 wild *Arachis* accessions; This collection has been molecularly profiled.

<u>Reference set</u>: A set of 300 genetically most diverse groundnut accessions, captures 466 (95%) of the 490 composite collection alleles representing diversity from the entire spectrum of composition collection.

<u>Core- and mini-core collections</u>: Concept of core collection promoted to enhance use of diverse germplasm in crop improvement; Found effective in improving efficiency of identifying genes of interest in entire germplasm collection; Core- and mini- core collections may need periodic update if more characters are added or new accessions are included.

<u>Formation of core- and mini-core collections</u>: Entire germplasm collection clustered into different groups by analyzing data on morphological traits following Ward's method; From each cluster, then, 10% accessions randomly selected to formulate core collection; ICRISAT groundnut core collection consists of 1704 accessions and that of the USA 831. Mini core collection developed with further sampling of 10% accessions in the core collection.

<u>Trait specific collection</u>: Subsets selected from the core- or mini-core collection possessing specific traits or after screening full or part of germplasm collection with respect to a particular trait along with other traits and creating a set of germplasm by selecting 10% or minimum of one accession from each cluster after diversity analysis.

<u>Germplasm exchange</u>: Since the Convention on Biological Diversity (CBD) in 1993, the international exchange and field collection of germplasm have become more tedious and restricted, more strict quarantine policies and intellectual property right issues greatly affect international germplasm exchange; Exchange of breeding materials, which was quite common in the past, has now become almost non-existent even within a country; Germplasm obtained prior to 1993 at ICRISAT is freely available but those obtained after are subject to terms of the CBD; ICRISAT is the main source of germplasm and breeding material, where these can be acquired after completing certain formalities such as Material Transfer Agreement (MTA) foregoing claims of ownership and intellectual property rights.

<u>Centers maintaining major collection of groundnut genetic resources</u>: <u>Argentina</u> – IBONE/INTA (3534 accessions); <u>Brazil</u> – CENARGEN/EMBRAPA, Brasilia (3340 accessions); <u>China</u> – Oil Crops Research Institute, Wuhan / National Gene Bank, Chinese Academy of Agricultural Sciences, Beijing (8349 accessions); <u>ICRISAT</u> – RS Paroda Gene Bank, ICRISAT, Patancheru, India (15,418 representing 95 countries); <u>India</u> – ICAR – Directorate of Groundnut Research, ICAR, Junagadh (9024 accessions); National Bureau of Plant Genetic Resources, ICAR, New Delhi (14,585 accessions); <u>USA</u> – Southern Regional Plant Introduction Station, Griffin, GA (USDA collection– 9917 accessions); National Seed Storage Laboratory, Ft. Collins, Co (duplicate collection under long-term storage); NC State University, Raleigh, NC (740 accessions). Other centers are: <u>Senegal</u> – Senegalese Institute of Agricultural Research (ISRA), Bambey and <u>USA</u> – Texas A&M, College Station, TX.

<u>Safe seed storage/conservation</u>: Gene banks dealing with germplasm conservation and distribution should have two stage conservation – i. Base collection, which is used for further multiplication of genotypes, is stored for long term at – 20 °C (preferred) or sub- zero temperatures with seeds having >85% germination and seed moisture ranging between 3 and 7%, a sample containing 1500-2000 seeds for each genotype is to be stored, ii. Active/Working collection, which is used for distribution, is stored for medium term at 4 °C and 20-30% relative humidity with seeds having >80% germination and seed moisture ranging between 7 and 8%, a sample of 1.5 kg is to be stored; The medium-term storage retains >65% seed viability for 10-20 years.

Sum of temperature (in °F) and relative humidity should be less than 100 to have optimal seed storage; Under ideal storage conditions, groundnut seeds remain viable for 15 or more years; Seeds stored at 10 °C and 45% relative humidity had good germination even after 20 years.

# 7. Genetics /inheritance/heritability of various traits of interest

<u>Genetics/inheritance of traits of interest</u>: Most agronomic traits in groundnut are inherited quantitatively and highly influenced by G x E interaction; When both parents belong to the same botanical variety, additive genetic variance is the principal component of genetic variance, and when they belong to different botanical varieties, both additive and non- additive genetic variance may be significant (Table 8).

Trait	Reported inheritance in literature (not an exhaustive review)
Plant type	and associated traits
Growth habit – Erect / Bunch (Valencia and Spanish types), Semi- spreading / Spreading / Runner (Virginia type)	Complex inheritance – monogenic, digenic, trigenic, tetragenic, gene–cytoplasmic and maternal inheritance for growth habit; Generally spreading habit dominant over bunch habit but in some cases, the opposite also found; Semi– spreading habit monogenic dominant to bunch and spreading forms; Spreading dominant to bunch with complementary and duplicate effects of two genes.
Plant height	Both additive and on-additive gene effects.
Dwarf plant stature	Different forms of dwarfism are observed: sterile brachytic, fertile dwarf, etc. Sterile brachytic – Monogenic recessive in natural and induced mutants; Two recessive complementary factors; Trigenic complementary; Tetragenic inheritance with two sets of factors with complementary- duplicate action; Fertile dwarf – Normal monogenic dominant over dwarf with cytoplasmic modifiers; More than one gene involved in plant height determination; Dominant with variable expressivity and penetrance.
Canopy breadth	Additive gene effects; Partial dominance.
Branching vs. non-branching	Branching in Virginia type monogenic dominant over non-branching Valencia type; Monogenically recessive/ double recessive for suppressed primary branches in induced mutants; Quantitative inheritance.
Number of primary branches	Both additive and non-additive gene effects.
Number of secondary branches	Both additive and non-additive gene effects.
Length of primary branches	Both additive and non-additive gene effects.

Trait	Reported inheritance in literature (not an exhaustive review)
Reproductive branches on main stem	Absence is dominant to the presence; Two sets of duplicate loci with epistatis control, when both loci of each set or all four loci are recessive flowering occurs; Two, three or four sets of homozygous recessive loci or modifying factors.
Stem pigmentation	Purple or red pigmentation monogenic dominant/ incompletely dominant over light or green color; Monogenic, digenic duplicate and digenic complementary inheritance for purple color; Two sets of genes of which one responsible for purple and the second for green pigmentation, extra nuclear factors; Duplicate recessive for white stem.
Stem pubescence	Stem pubescence monogenic dominant/ incompletely dominant over absence of pubescence; Monogenic inheritance with overdominance of hairiness.
	Leaf traits
Elliptical shape	Elliptical leaflet shape on the main axis monogenic recessive to elliptical-oblong shape.
Krinkle leaf	Both monogenic dominant and recessive in different krinkle mutants.
Mottled leaf	Monogenic dominant to normal leaf in natural mutants.
Narrow leaf	TMV 2 narrow leaf mutant and Gujarat narrow leaf mutant are genetically different; Partial dominant monogenic; Monogenic dominant in induced mutant.
Cup leaf	Monogenic recessive in induced mutant.
Flop leaf	Monogenic recessive and digenic recessive in induced mutants.
Curly leaf	Monogenic recessive in a natural mutant.
Corduroy leaf	Duplicate recessive in induced mutant.
Puckered leaf	Recessive with 13 normal : 3 puckered leaf ratio.
Trait	Reported inheritance in literature (not an exhaustive review)
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Dark green leaf color	Dark green color monogenic dominant/ incomplete dominant over light green color; Dark green color duplicate recessive in radiation induced mutant; Light green color dominant over dark green color in crosses involving Chico variety with at least two dominant alleles of one of the two loci or one dominant allele each at both loci necessary for regulation of chlorophyll synthesis.
Lutescent leaf color	Two duplicate recessive genes.
Golden yellow color	Two duplicate recessive genes.
Albinism	Duplicate recessive, triplicate recessive, trigenic model in which duplicate loci controlling chlorophyll development epistatic to a third locus governing a zygotic lethal; Cytoplasmic factors influencing expression of nuclear genes governing albinism.
Variegated leaf	Monogenic dominant in induced mutant, maternally inherited.
Leaflet size/ leaf area	Intra-plant variation in leaflet size common in groundnut. Large leaflet size incompletely /completely dominant over small size; Monogenic/duplicate recessive in induced and natural mutants; Predominant additive gene effects for leaf length and both additive and non- additive gene effects for leaf width; Predominantly non-additive gene effects and reciprocal effects for leaf area.
Petiole length	Both additive and non-additive gene effects important; Three recessive genes for short petiole.
Number of stomata	Low number monogenic recessive and digenic recessive.
Number of leaves on main stem	Both additive and non-additive gene effects.
Number of leaves on cotyledonary branches	Predominantly additive gene effects.

Trait	Reported inheritance in literature (not an exhaustive review)	
Iı	nflorescence	
Inflorescence length	Elongated inflorescence dominant over condensed inflorescence with two genes with complementary interaction.	
Type of inflorescence	Two complementary genes with 9 simple: 7 compound inflorescences in Valencia (simple) x Virginia (simple) crosses, however, a ratio of 13 compound: 3 simple inflorescences observed in Valencia (simple) x Spanish (compound) crosses suggesting presence of two genes inhibiting expression of simple inflorescence in Spanish type.	
Color of standard petal	Five different intensities of standard petal color observed; In general deep color dominant over light colors; Orange monogenic dominant /incomplete dominant/co-dominant over white; Faint orange monogenic recessive to orange; Duplicate recessive control of white flower when crossed with lines having yellow flowers; Incomplete monogenic dominance of yellow flower over white flower; Additive effects of two different genes for yellow color; Lemon yellow dominant to orange flower with presence of transposable elements; Two complementary genes for garnet color which is dominant over orange color; Trigenic control.	
Presence vs. absence of standard crescent	Purple crescent dominant to no crescent governed by duplicate genes; Single dominant gene for purple crescent.	
Number of pegs	Both additive and non-additive gene effects.	
Peg strength	Significant SCA.	
Peg pigmentation	Monogenic dominant/digenic inheritance for purple pigmentation of pegs	
Brachytic steriles	One, two, three or more recessive genes; Two sets of factors with complementary-duplicate action condition brachytic character.	
Female sterility	Monogenic and trigenic recessive inheritance.	
Male sterility	Two recessive genes.	

Trait	Reported inheritance in literature (not an exhaustive review)
	Pod traits
Pod size	Large size dominant over small size and monogenic, digenic, trigenic and multigenic control reported; Small size dominant over large size with duplicate gene interaction; Predominantly additive gene effects.
Pod length	Normal length dominant over long length with duplicate genes; Both additive and non- additive gene effects, maternal effects.
Pod width	Both additive and dominance gene effects, maternal effects.
Pod constriction	Absence of constriction or shallow construction dominant over deep constriction and controlled by two independent dominant genes; Trigenic complementary inheritance for shallow constriction; Three unlinked loci and cytoplasmic factor.
Pod reticulation	Monogenic dominant for strong reticulation over shallow or absence of reticulation; At least four factors for deep reticulation; Smooth pods dominant over reticulated pods with digenic inhibitory gene action (13 smooth: 3 reticulated types).
Pod pubescence	Two loci with additive gene action.
Pericarp thickness	Monogenic dominant thin pericarp over thick pericarp; Five factors with thin pericarp dominant over thick pericarp; Three complementary genes for moderately thick pericarp.
Pod beak	Non-beaked pods monogenic dominant over beaked pods; Prominent beak monogenic dominant over non-beaked pods.
Aerial podding	Aerial podding monogenic dominant over normal podding and the opposite also true depending upon the genetic backgrounds of the parents.
One-seed pod	Any two of three duplicate recessive genes control one-seed pod.

Trait	Reported inheritance in literature (not an exhaustive review)	
See	ed characters	
Number of seeds per pod	Three or more seeds dominant over fewer seeds with trigenic/monogenic control; Fewer than three seeds dominant over three or more seeds with monogenic control.	
Seed size	Large seed size monogenic dominant over small size with possible modifiers; Five pairs of genes with four having isodirectional effects.	
Shrivelled seeds	Single recessive gene.	
Seed shape	Round shape monogenic/duplicate genes recessive to elongated seeds; Seed length maternally controlled; Button type seeds dominant over normal seeds (round or elongated) with digenic ratio of 13:3 indicating influence of inhibitory genes on seed shape.	
Seed ends	Flat ends of seed monogenic dominant over smooth ends.	
Seed length	Mainly additive gene effects and significant reciprocal/maternal effects; Both additive and dominance gene effects.	
Seed width	Mainly additive gene effects and significant reciprocal/maternal effects; Both additive and dominance gene effects.	
Seed length and width ratio	Mainly additive gene effects and significant reciprocal/maternal effects.	
Hard seed	Paternal inheritance, both hard and soft seeds are allelic and follow monogenic inheritance in induced mutant, the gene for hard seed has pleiotropic effect.	
Rough testa	Duplicate genes with recessive epistatis with 9 rough:7 smooth testa.	
Testa color	Inheritance of testa color varies from simple to very complex. Seven gene pairs / Nine loci interacting in several ways to produce different testa color.	
i. Flesh (rose/pink/russet or tan) testa	Duplicate dominant loci	
ii. White testa	Recessive to colored tests; Two different dominant genes epistatic to red and flesh testa color in specific genotypes.	

Trait	Reported inheritance in literature (not an exhaustive review)
iii. Red testa	One dominant gene/partial dominance/single recessive gene/two duplicate recessive genes/two complementary recessive genes/polygenic control responsible for red testa color.
iv. Purple testa	Partial/complete dominance over all other colors with inheritance monogenic/digenic epistatic, duplicate, digenic cumulative, trigenic and tetragenic with complex interactions; Duplicate recessive.
v. Wine testa	Monogenic recessive.
vi. Chocolate testa	Monogenic/digenic recessive in an induced mutant.
vii. Variegated testa	Dominant/ partially dominant and recessive to non-variegated testa; Monogenic/digenic/two genes with cumulative effects; Trigenic inheritance with epistatic effect of purple over red.
Seed coat splitting	Monogenic inheritance with additive effects, duplicate additive and digenic complementary.
Yield a	nd related traits
Pod yield	Both additive and non-additive gene effects.
Number of mature pods per plant	Both additive and non-additive gene effects.
Number of immature pods per plant	Both additive and non-additive gene effects.
Shelling outturn	Governed by a pair of genes without dominance; Both additive and non-additive gene effects.
Weight of seed per plant	Both additive and non-additive gene effects.
Seed number per kg	Predominantly additive gene effects.
% sound mature kernels	Predominantly on-additive gene effects.
100-pod weight	Both additive and non-additive gene effects.
100-seed weight	Both additive and non-additive gene effects; Mainly additive gene effects and significant reciprocal/maternal effects.
Haulm yield and quality	
Green weight/haulm yield	Both additive and non-additive gene effects; Both GCA and SCA for biomass.

Trait	Reported inheritance in literature (not an exhaustive review)	
	Life cycle	
Annual vs. perennial	Perennial growth habit dominant over annual habit.	
Ci	rop duration	
Early maturity and its components	Late maturity monogenic dominant/ incompletely dominant over earliness; Four or five genes with complete dominance of lateness over earliness and absence of reciprocal differences; Single gene with additive effect/predominantly additive gene effects for days to flower, three genes with two types of epistatis (dominant recessive, 13 late : 3 early and duplicate dominant, 1 late : 15 early) for days to accumulation of first 25 flowers and absence of reciprocal differences; Two recessive genes acting in an additive manner for days from seedling emergence to first flower; Significantly higher GCA variance than SCA variance for time of emergence (in h), time to first leaf opening on cotyledonary branch (in h), time to first leaf opening on main stem (in h), days to first flower and number flowers per plant (at 32 days) under controlled environment conditions; Significantly higher GCA variance than SCA variance for days to first flower under field conditions; Significant additive genetic variance with bidirectional dominance/ non- additive gene effects for days to first flower; Significant additive genetic variance for pod and seed maturity indices; Both additive and dominance genetic effects for maturity index; Highly significant additive and significant dominance, additive x additive and significant dominance effects with duplicate digenic interaction for days from emergence to first flower, more than two genes or linkage effects between the genes for number of flowers produced during first four days of flowering, highly significant additive and significant dominance effects for percentage of ripe pods at 80 days after sowing; Predominantly non- additive gene effects for days to maturity.	
Biochemic	al/Nutritional traits	
Oil content	Both additive and non-additive gene effects; Epistatis interaction.	

Trait	Reported inheritance in literature (not an exhaustive review)	
Protein content	Both additive and non-additive gene effects; Epistatis interaction.	
Oleic acid content	One/ two recessive genes for high oleic acid.	
Fatty acids (palmitic, stearic, oleic, linoleic, arachidic, eicosenioic, behenic and lignoceric fatty acids, total saturated fatty acids and long chain saturated fatty acids)	Additive gene effects and additive x additive interaction.	
Oleic/linoleic fatty acid (O/L) ratio	Single recessive or two recessive genes and some possible modifiers depending upon the parents involved in the crosses; Additive gene effects and additive x additive interactions.	
Polyunsaturated/saturated fatty acid (PS) ratio	Both additive and dominance gene effects and additive x additive and additive x dominance interactions.	
Arginine content	Two major genes with partial dominance for low arginine.	
Iodine value	Both additive and dominance gene effects and additive x additive and additive x dominance interactions.	
Soluble sugars	Predominantly additive gene effects.	
Physiological traits		
Iron chlorosis	One basic gene and two or four inhibitory complementary genes for expression of iron chlorosis.	
Harvest index	Both GCA and SCA; Predominantly gca; Predominantly additive gene effects with additive x additive epistatis.	
Leaf chlorophyll content	Quasi-quantitative with modifiers either in positive or negative direction in different genetic backgrounds.	
Chlorophyll a, chlorophyll b and total chlorophyll	Non-additive gene effects with significant additive x dominant epistatis interaction.	
Carotenoid content	Dominant gene effects with significant additive x dominance and dominance x dominance interactions.	
SPAD chlorophyll meter reading (SCMR)	Both additive gene effects and dominance gene effects with duplicate and complementary epistatis and additive x additive interactions.	

Trait	Reported inheritance in literature (not an exhaustive review)	
Specific leaf area (SLA)	Predominantly additive gene effects and additive x additive interactions; Predominantly dominance gene effects with duplicate epistatis.	
Specific leaf weight	Predominantly non-additive gene effects.	
Fresh seed dormancy	Complete/ incomplete monogenic dominance for dormancy over non-dormancy; Multigenic control.	
Apparent photosynthesis	Dominance and overdominance effects.	
Response to photoperiod	Mainly gca, additive gene effects in some cases and partial dominance to dominance in others. Maternal effects.	
Nitrogen fixation		
Nodulation and nitrogen fixation	Nodulation dominant to non-nodulation; Monogenic, digenic and trigenic inheritance; A trigenic model proposed - the first two genes produce nodulation while the third one inhibit nodulation when dominant and the former two in homozygous recessive condition. Although the presence or absence of nodulation is governed by a few major genes, the intensity of nodulation appears to be controlled quantitatively; Predominantly additive gene effects for nitrogen fixation; Both additive and non-additive gene effects and additive x additive, additive x dominance and dominance x dominance interactions for nodule number and nodule mass per plant, nitrogenase activity and acetylene reduction; Non-additive gene action and reciprocal effects for nitrogenase activity, nodule number, nodule mass and total nitrogen; Predominantly SCA for nodule number and weight, specific nitrogenase activity, shoot weight and total plant nitrogen, maternal effects also important.	

Trait	Reported inheritance in literature (not an exhaustive review)
Dise	ase resistance
Rust	Sexual stage and races in groundnut rust pathogen not yet observed. Monogenic/ digenic /trigenic inheritance with resistance being recessive; Preponderance of non- additive gene action; Greater dominance variance; Dominant, partial dominant or additive gene action for resistance; Both additive and non-additive gene effects and additive x additive and additive x dominance interactions; Significant GCA and SCA for rust resistance; Resistance dominant/partial dominant in wild <i>Arachis</i> species. The resistance is stable overs years and locations.
Late leaf spot (LLS)	LLS and ELS inherited independently; Duplicate complementary recessive in induced mutants; Resistance recessive and level of resistance controlled by presence of recessive gene(s) at any or all of the five loci; 4-5 duplicate recessive genes; Both additive and non-additive gene effects and additive x dominance epistatis, maternal effect; Additive gene effects for components of resistance (lesion number, lesion area, defoliation, latent period and spore production).
Early leaf spot (ELS)	Chemical induced physiological races observed which may give differential reaction to resistant sources. Additive and non- additive gene effects and additive x additive gene interaction for resistance with involvement of cytoplasmic factors; Duplicate recessive in induced mutants.
Sclerotinia blight	At least two genes involved; Quantitative inheritance with involvement of dominance, epistatis and cytoplasmic factors.
Cylindrocladium black rot	Predominant additive gene effects; Complex inheritance with resistance delaying the onset of disease.

Trait	Reported inheritance in literature (not an exhaustive review)	
Aspergillus flavus/ Aspergillus parasiticus and aflatoxin production	No correlation between aflatoxin content and <i>in</i> <i>vitro</i> seed colonization and the population density of <i>Aspergillus flavus</i> in the soil; Three resistance mechanisms – preharvest resistance, seed coat resistance ( <i>in vitro</i> seed colonization and cotyledon resistance (aflatoxin production)), all inherited independently; Predominance of additive gene effects for seed coat resistance, reciprocal differences observed; Predominantly non-additive gene effects for aflatoxin production.	
Groundnut rosette disease (GRD)- chlorotic rosette and green rosette	Three agents responsible for expression of disease symptoms – groundnut rosette virus (GRV), groundnut rosette assistor virus (GRAV) and satellite RNA (SatRNA); Two independent recessive genes for resistance to GRD (effective against GRV and SatRNA but not against GRAV)	
Peanut bud necrosis disease (PBND)	Three resistance factors inherited additively for reduced disease incidence; Highly significant GCA and significant SCA and reciprocal effects; Nonadditive/ additive/dominance/epistatic and additive x additive gene effects in different crosses.	
Tomato spotted wilt virus disease (TSWV)	Significant GCA and SCA; Transgressive segregation for resistance observed.	
Peanut stripe virus disease (PStV)	Significant GCA and SCA.	
Bacterial wilt	Partially dominant involving three pairs of major genes and some minor genes; Recessive resistance; Nucleo-cytoplasmic interaction and both additive and dominant gene actions.	
Resista	nce to nematodes	
<i>Meloidogyne arenaria</i> (root knot nematode) race 1	Resistance conferred by single/ two dominant genes (one inhibits root galling and the other inhibits egg production).	
Insect pest resistance		
Leaf hopper	Resistance controlled by three recessive genes with additive effects; Predominant additive gene effects for long trichomes on mid-rib and petiole and jassis damage; Predominant non-additive gene effects for short, medium or long trichomes on adaxial leaf surface, margins, mid-rib and petiole.	

Trait	Reported inheritance in literature (not an exhaustive review)
Aphids	Single recessive gene for resistance
Leaf miner	Significant GCA and SCA.

<u>Traits affected by reciprocal/maternal differences</u>: Reciprocal effects due to interactions between nuclear and cytoplasmic factors; Only cytoplasmic factors account for maternal effects indicating superior cytoplasm; Parents with superior cytoplasm used as female parent in hybridization; Significant reciprocal/maternal effects reported for several quantitative traits – nitrogen fixation traits excluding nodule dry weight, pod and seed traits such as pod/seed weight, length and width and number of pods/seeds per plant, yield per plant, resistance to ELS, LLS and *in vitro* seed colonization by *Aspergillus flavus*, calcium concentration in seeds, days from emergence to first flower.

<u>Heritability of various traits</u>: Each heritability estimate is unique and determined by the experimental material and the character under study, experimental design followed, method of estimation, environmental conditions and control. Heritability of various traits of interest is given in Table 9.

Trait	Range (or value) of broad sense heritability (H)	Range (or value) of narrow sense heritability (h <sup>2</sup> )
Height of main axis	33.7 - 97.2	71.0
Branching pattern	90.4	55.3
No. of primary branches	33.5 - 94.5	23.2 - 59.0
No. of secondary branches	12.0 - 98.9	
Length of primary branch	28.0	64.5
Length of secondary branch	38.0	36.2
Shoot dry weight	37.0 - 100.0	
Plant fresh weight	1.0	
Total dry matter	38.8 - 98.0	
No. of nodules per plant	44.3	
Single nodule weight	42.9	
Nodule weight per plant	33.0 - 100.0	
Nitrogenase activity	60.8	
Acetylene reduction	31.0	
Days to emergence	48.0-83.0	51
Days from emergence to first flower	11.9 - 96.9	23.0 - 39.0

**Table 9.** Heritability of various traits reported from different studies in literature.

Trait	Range (or value) of broad sense heritability (H)	Range (or value) of narrow sense heritability (h²)
Days to 50% flowering	66.5 - 96.3	60.0
Days from emergence to accumulation of 25 flowers	17.0 - 61.0	
Number of flowers produced during first four days of flowering	16.0 - 44.0	9.0 - 38.0
Peg number	54.2 - 56.2	11.7 - 45.7
Peg strength	74.1	
Peg length	74.0	37.7
Peg diameter	75.0	
Pod-peg ratio	48.8	
Days to maturity	91.7 - 98.6	
Fruit maturity based on oil pigmentation	69.0 - 95.0	
Maturity based on hull scrap method	71.0	
Percentage of ripe pods 80 days after sowing	13.0 - 41.0	24.0
No. of immature pods per plant	3.7 - 92.7	1.4 - 5.1
Pod size		42.0 - 50.0
No. of mature pods per plant	26.1 - 100.0	31.7 - 32.0
Pod weight	42.0 - 80.0	
Pod length	54.0 - 92.0	
Pod yield per plant	13.2 - 98.0	59.8
100-pod weight	75.0 - 100.0	
Seed yield	38.3 - 83.9	31.5
100-seed weight	28.6 - 100.0	57.3 - 66.8
Harvest index	27.7 - 100.0	
Shelling outturn	33.3 - 100.0	10.5 - 42.0
Oil content in seed	22.0 - 94.4	29.0
Protein content in seed	43.0 - 64.0	14.0
Zn content in seed	92.0	
Fe content in seed	81.0	
Carbon isotope discrimination (Lab)	53.0	

Trait	Range (or value) of broad sense heritability (H)	Range (or value) of narrow sense heritability (h <sup>2</sup> )
Carbon isotope discrimination (Field)	75.0 - 89.0	
Transpiration efficiency	34.0 - 86.0	
Total transpiration	12.0 - 70.0	
SPAD chlorophyll meter reading	9.0 - 98.0	
Specific leaf area	5.0 - 98.0	
Drought tolerance index	54.0 - 98.0	
LLS disease index		22.0 - 27.0
LLS resistance		0 - 13.0
ELS resistance	51.7	
Resistance to CBR	48.0 - 65.0	51.7
Resistance to <i>Sclerotinia miner</i> # of days until plants wilted	41.5 - 50.3	
Disease rating	14 - 23	1.0 - 11.0
Seed coat resistance to Aspergillus flavus	78.5	
Resistance to Arachis parasiticus		
i. Dry seed resistance ii. Aflatoxin production iii. Pre-harvest resistance	30.0 - 65.0 20.0 - 65.0 27.0 - 33.0	
Fodder quality traits		
i. Nitrogen content	0.72	
ii. <i>In vitro</i> organic matter digestibility (OMD)	0.72	
iii. Metabolizable energy content (ME)	0.67	
iv. Digestible haulm yield	0.91	

(Adapted from Murthy and Reddy, 1993 and others.)

# 8. Methods of cultivar development

Standard breeding methods, practiced in a self-pollinated crop, are also followed in groundnut for developing a cultivar; Pedigree and bulk methods are more commonly used by breeders than single seed descent (SSD) or recurrent selection; Some breeders use a combination of breeding methods or make modification in a conventional method; Selection for yield and yield-related traits in groundnut practiced only after harvest because of the subterranean nature of the crop.

(For detailed description of different breeding methods, readers are advised to consult any standard plant breeding book.)

<u>Methods without artificial hybridization</u>: These methods involve selection in existing (land races) or introduced (introductions) variability or variability created without sexual process (mutation breeding and genetic transformation).

(Genetic transformation discussed separately.)

<u>Introduction</u>: Plant introductions useful in bringing in additional genetic diversity in a breeding program; Sometimes they can also be released directly as cultivars if found suitable; Indian cultivars TMV 2 and JL 24 introduced and released under different names in many countries in Southeast Asia and Africa.

<u>Pure line / Mass selection in introduced material</u>: Selection and subsequent seed increase for release as cultivars of single plants (pure line) or group of plants (mass) selected from introduced material and landraces, etc.; Many cultivars in Africa (Makulu Red, Apollo, Egret, Chalimbana, Mani Pintar and Malimba), India (JL 24) and the USA (New Mexico Valencia C) developed through pure line or mass selection in introduced material.

<u>Mutation breeding</u>: X-rays, gamma-rays and various chemicals used to break specific linkages, create variation for specific characters and/or use in conjunction with other breeding methods; Gamma-rays extensively used at BARC, Mumbai to create desired variation for further use in conjunction with other breeding methods; BARC released 15 groundnut varieties, some of these are TG 19, TAG 24, TG 37A, TG 38B, TGB 39, TPG 41, TLG 45, TG 51.

<u>Methods after artificial hybridization</u>: Hybridization provides opportunity to combine genes from different parents; Choice of parents very critical for the success of a breeding program; Parents can be crossed in different manners to access desirable genes from them and recombine them (genes) in a single genotype – single cross, three-way cross, four-way cross, convergent cross, diallel mating, diallel selective mating; It usually takes 12-15 years after hybridization to develop a cultivar; This period can be shortened by taking multiple crops in a year in a glasshouse under controlled conditions (and following single seed descent method) or raising off-season nursery at other locations where environmental conditions are favourable to raise a crop; In crosses between parents having equal or nearly equal number of alleles (k=0.5), selfing provides the greatest probability of recovering the desired plants; As k approaches 1, the probability of recovering desired plants by backcrossing to the better parent is high.

<u>Bulk selection</u>:  $F_1$  plants space-planted to produce as much  $F_2$  seed as possible, which is planted en masse the next generation;  $F_3$  seed harvested in bulk (limited selection for easily identifiable traits (high heritability) such as plant type or seed size can be practiced on  $F_2$  plants) and a sample planted the next generation with the process being repeated until  $F_6$  generation; At this stage a portion of the seed space-planted, and individual plants selected for evaluation in progeny test, undesirable plants discarded from the  $F_6$  derived progenies to make them uniform for replicated on-station and multilocation evaluation; Because of its low cost and little efforts required in generation advance, the method continues to be used in breeding programs; Can be used to select for desirable traits in segregating populations originating from inter-subspecific crosses between adapted and unadapted germplasm; But less effective in selection in intra-subspecific crosses; However, bulk selection may cause genetic shifts in segregating populations; Should be performed for few generations only.

<u>Pedigree method</u>: Very commonly used method in spite of extensive record keeping of line descent or pedigree of each plant; Individual plants selected based on desired traits in space-planted  $F_2$ 

populations; Selected plants then space-planted in progeny-rows in  $F_3$  generation and again the best plants selected and progeny-rowed in  $F_4$ ; Selection process repeated in  $F_5$  and the selected plants advanced to  $F_6$  or  $F_7$  generation; When progenies become uniform (with each generation of selfing, the homozygosity increases), selection is made on plot basis; Selected genotypes (uniform experimental line) then evaluated in replicated trials on- station and at multilocation; Selection can also be carried out in a sequential manner at different locations to enhance adaptability and stability of the selected populations; First used in the USA in Florida in 1928; Most of the cultivars developed by pedigree method are  $F_5$ -derived lines.

<u>Bulk pedigree</u>: This method, a combination of pedigree and bulk methods and often not described in breeding books, practiced in groundnut breeding program at ICRISAT; Differs from bulk method that selected individual plants in  $F_2$  population are bulked into different bulks based on morphological traits including pod and seed characteristics and their record of descent kept; Method allows retention of enough subtle variation in the resultant uniform looking bulk populations for recipient breeders to exploit it while evaluating them under local conditions.

Single seed descent (SSD): A modification of the bulk method that can be used to inbreed populations in a shorter length of time;  $F_2$  seeds obtained from space-planted  $F_1$  plants dense planted and a single seed or pod or small quantity of  $F_3$  seed collected from each  $F_2$  plant and bulked which is again grown densely and the process of selection repeated until the desired number of inbreeding generations achieved (generally  $F_6$ ); At this stage the final selected population space-planted for selection of individual plants for evaluation in progeny test, undesirable plants are discarded from the  $F_6$  derived progenies to make them uniform for replicated on-station and multilocation evaluation; At NC State University peanut breeding program, they could raise three generations in 14 months following this method in a glasshouse and breaking seed dormancy before planting, if required.

<u>Backcross</u>: Mostly used for transferring desired traits from a donor source to existing cultivar; Occurrence of small number of simply inherited traits of economic importance in groundnut limits the use of this method; However, with increasing emphasis on disease resistance breeding, this method is gaining popularity among breeders; First used in Malawi (Southern Africa) to incorporate groundnut rosette virus disease resistance governed by two recessive genes into highyielding cultivar Makulu Red; In the USA, used to transfer high oleic trait into acceptable agronomic backgrounds; For more details on two or three stages of crossing and selfing per cycle of selection, minimum number of plants required in each stage and cost effectiveness, consult Isleib, 1997 (Crop Science 37: 139-144).

<u>Recurrent selection</u>: Recurrent selection received little attention in groundnut breeding because of efforts needed to make a large number of crosses; It allows incorporation of diversity in breeding programs while providing opportunities for recombination; It enhances the probability of incorporation of several desirable traits from different sources in a single genotype; Peanut breeding program at NC State University, NC, USA extensively used this breeding method.

<u>Diallel selective mating (DSM)</u>: A form of recurrent selection, which allows use of a selected group of parents and provides opportunity for multiparental gene combination; Offspring of two-way crosses can be crossed to each other or can be grown out and examined for important characteristics and only selected plants are intercrossed; At any stage in the program, material can be intercrossed or carried through a pedigree, bulk or SSD selection program to obtain potentially new cultivars.

<u>Single cross vs. Complex crosses</u>: Complex crosses (three-way, double cross, convergent crosses, etc.) recommended for obtaining desirable combinations of traits from different sources; Some

researchers found three-way crosses superior to single crosses, while others did not find any difference between pedigree selections from single and double crosses.

<u>Inter-subspecific vs. Intra-subspecific crosses</u>: In addition to releasing greater diversity, intersubspecific hybridization also results in deleterious albino, brachytic, dwarf and sterile plants in segregating generations; Inter-subspecific crosses generally show more heterosis than intrasubspecific crosses.

<u>Reciprocal cross differences</u>: Direction of the cross has a bearing on the types of segregants obtained; Reciprocal differences observed due to cytoplasmic effects and maternal effects; These two need to be separated in reciprocal effects as cytoplasmic effects have a significant bearing and the maternal effects have very little bearing on breeding programs; Reciprocal differences often occur in inter-subspecific crosses (Spanish/ Valencia types x Virginia types).

<u>Maternal effects</u>: Observed particularly when studying seed characteristics (such as dormancy and fatty acid composition, etc.) as they are influenced by the mother plants on which seeds are borne; However, such maternal effects last for only one generation and have little consequence in breeding programs.

<u>Early generation testing</u>: Normally early generation testing is done to identify superior crosses and eliminate inferior crosses early in the breeding program, thus enhancing breeding efficiency; No clear and significant advantage in early generation testing observed in groundnut as most breeding efforts deal with quantitatively inherited traits; Traits with higher heritability can only be selected effectively in early generations.

<u>Index selection</u>: Index selection allows concurrent improvement of several traits in early generations which go into the making of selection index; A selection index may contain traits with higher heritability, which are related with other economically important desirable traits that have low heritability.

<u>Natural hybridization</u>: Depending upon the level of bee activity, variety and weather conditions, the outcrossing rate may vary from <1% to 10%; If natural hybridization occurs between the genotypes of two subspecies, it is easy to identify the natural hybrids in an otherwise uniform parental population; If it occurs between the genotypes within a subspecies, the identification becomes difficult; Natural hybridization causes genetic instability in the mother genotype as the resultant hybrids and their progenies genetically contaminate the mother genotype; Natural hybridization remains unknown; Exploited as early as in 1923 to develop groundnut cultivars in Indonesia; At ICRISAT, cultivars ICGS 11, ICGS 44, ICGS 37, etc. originated from natural hybrid population of Robut 33–1.

<u>Genotype x environment interaction (G x E)</u>: Desirable quantitative traits usually have both genetic and environmental components, separation of these components necessary to achieve maximum efficiency in breeding; Significant G x E interaction has been observed for most of the quantitatively inherited traits confirming the need of extensive multiyear and multilocation testing prior to cultivar release.

<u>Multiline variety</u>: Bulking of phenotypically similar but genotypically dissimilar sister lines to form a cultivar; Sister lines maintained individually and bulked prior to each seed increase of Foundation seed; This method allows the breeder to improve the cultivar after release by adding or dropping component lines; Main advantage – cultivar with wide genotypic variability and stability, phenotype satisfies market and growers' requirements; Main disadvantage – less uniform, seed stocks difficult to maintain, multiline cultivar lower yielding than the best component line; Successfully used in Florida breeding program in the USA to develop Florunner and Florigiant multiline cultivars.

<u>Variety release procedures</u>: Procedures vary from country to country. The procedure followed in India is given below; New varieties in India can be released either at the central level by the Central Sub-committee on Crop standards, Notification and Release of Varieties (CVRC) or at the state level by the respective State Variety Release Committees (SVRC).

Variety release at the central level: Three steps involved in release of a potential advanced breeding line as a variety by the CVRC: (1) Evaluation, (2) Identification, and (3) Release and Notification followed by seed multiplication.

During the Annual All India Coordinated Research Project (AICRP) Workshops, a breeder, with supporting evidence of superior performance in station trials, may propose his new breeding line(s) for inclusion in multilocation AICRP trials; For each crop, based on climatic and edaphic factors, the country has been divided into various agroecological zones and in each zone are several testing locations; Each genotype has to be evaluated for at least three years in a 3-tier system of AICRP trials (Initial Varietal Trial (IVT), Advanced Varietal Trial I (AVT I) and Advanced Varietal Trial II (AVT II) before it is considered for Identification for release by the Variety Identification Committee (VIC) constituted by ICAR each year during the Annual Workshop of the respective crops.

An IVT is conducted at all the locations across all the zones for a particular crop; Entries tested in IVT for one year only and those found promising (at least 10% advantage in yield) promoted in respective zones to the Advanced Varietal Trial I (AVT I) for further evaluation; At this stage, the entries evaluated in a specific zone (s) and simultaneously, also screened for reaction to diseases and insect pests in screening nurseries; On the basis of yield superiority, the entries promoted to AVT II for further evaluation in respective zones; In AVT II, the promoted entries again evaluated for yield and other agronomic traits at all pests and also evaluated in separate agronomic trials (varieties x date of sowing, variety x fertilizer doses, varieties x number of irrigations, variety x spacing, etc.) to work out suitable agronomic practices for the new entry and also to test its performance in bigger plots.

Based on 3-year AICRP trials data, the concerned breeders submit proposal to the Variety Identification Committee (VIC), which meets during the Annual Workshop of the AICRP; VIC identifies suitable varieties for potential release based on 3-year data on performance of the variety as compared to check, seed quality and 2-year data on resistance to major diseases and insect pests; Variety should have at least 10% higher yield than the best local check or should have superiority in resistance to any major biotic/abiotic stress if it is at par with best local check in yield performance; After a variety is identified for release, the concerned breeder submits a proposal in a prescribed format for consideration of CVRC for its release and notification; Breeder of the new variety should ensure availability of sufficient quantity of Breeder seed of the proposed new variety for conducting front line demonstrations in farmers' fields and for further seed increase by public sector seed agencies.

The CVRC consists of Deputy Director General (DDG) (Crop Science), ICAR as chairman of the Committee, Production Commissioner, Government of India (GoI); Project Director and Project Coordinator (of respective crops); Principal Investigators; Directors of Agriculture of all the states; a representative of National Seeds Corporation (NSC); Director (High Yielding Varieties), Ministry of Agriculture; and Deputy Commissioner (Seeds); Once a variety is released by the CVRC, the Director (High Yielding Varieties) notifies the concerned authorities for its seed multiplication and distribution; Notification of a variety appears in the Gazette of India.

A variety must be notified by the Ministry of Agriculture to qualify for seed certification by the Seed Certification Agency of different states; After its notification, Foundation seed of the newly released variety is produced and the Certified seed in the next season; In the third crop season the Certified seed of the new improved variety is available for commercial cultivation; Nucleus and Breeder seed production is the responsibility of the concerned breeder/institute; Foundation and Certified seed production is generally undertaken by the public sector seed agencies.

Variety release at the state level: Procedure for release of varieties at the state level may slightly vary from one state to another; State agricultural universities (SAUs) follow their own system of multilocation evaluation including adaptive trials within the state.

If a genotype performs well in adaptive trials in a particular state for 3 consecutive years, it can be identified by the State Variety Release Committee (SVRC) for release in that particular state provided that the concerned genotype has been evaluated under AICRP for at least one year.

Breeder of the new variety first submits the proposal to the University level varietal release committee which evaluates the proposal and recommends only selected proposals to the SVRC. SVRC consists of (1) Director of Agriculture of the state as its chairman, (2) Director, State Seed Corporation, (3) Director, State Seed Certification Agency, (4) Additional, (5) Directors of Research of all agricultural universities of the state; In addition, Deputy Directors of Agriculture of the state, Officer-in-charge of all Regional Agricultural Testing and Demonstration Centres and Breeders, Agronomists, Pathologists and Entomologists of all agricultural universities and institutes in the state are also invited though they are not the members of the SVRC.

Once a variety is released at the state level by SVRC, a proposal for its notification is submitted by the concerned breeder with all requisite documentation to CVRC. This notification is essential if the variety has to enter formal seed production program.

The seed of a variety/hybrid developed by the private sector can be produced and marketed by the concerned seed company as 'Truthful Labelled' seed after its initial evaluation in AICRP trials.

<u>Farmer-participatory varietal selection (FPVS) and plant breeding (FPPB)</u>: These two activities empower farmers to influence the outcome of the plant breeding research to suit their requirements.

FPVS: Allows farmers to select among advanced breeding lines or genetically stable populations through on-farm farmer-participatory varietal selection trials; Various models of FPVS trials are being pursued in on-farm trials (researcher-designed and researcher- managed, researcher- designed and farmer-managed and farmer-designed and farmer- managed); Trials can follow various experimental designs based on the purpose of the trials and availability of seed material, etc.; Mother-Baby trials appear to be most popular design in FPVS; FPVS meets immediate need of better germpalsm of the farmers as it identifies the material best suited to the farmers' need and socio-economic conditions and agroecological conditions prevailing in the locality; Also allows multiplication and wide distribution of seed of farmer-preferred varieties through on-farm FPVS trials; FPVS also helps to identify varieties which should be used as parents in future hybridization program; Recent example of FPVS in groundnut – release of ICGV 91114 in 2006 in Andhra Pradesh, in 2008 in Odisha and in 2009 in Karnataka, of ICGV 87846 in 2010 as CO 6 and of ICGV 00351 in 2013 as CO 7 in Tamil Nadu in a short-period of time in India.

FPPB: Allows farmers' participation in a crop improvement program at various levels, which is non-existent in conventional breeding; In complete participatory breeding model, farmers' involvement at all stages – selection of source germplasm, trait development (pre- breeding), cultivar development and varietal evaluation; In an efficient participatory breeding, farmers involved at the selection of source germplasm and varietal evaluation stages; In traditional farmer breeding, all stages of crop improvement conducted by the farmers themselves without any input from the scientists; FPPB consumes more resources if selection activities carried out in farmers' fields; It should be used when the possibilities of FPVS have been exhausted; FPPB can be conducted at several locations with the same breeding populations to identify the best material suited to local conditions by exploiting G x E for the benefit of farmers; However, cultivation of several varieties enhances genetic diversity in farmers' fields but may pose problem in seed production and certification program; FPPB is a 'demand driven' process whereas the conventional breeding is 'supply driven'; Desirable to involve market players and processors also at appropriate stage in the evaluation of the varieties/breeding materials.

## 9. Genomics

<u>Genomics</u>: Understanding the highly complex structures and processes that make up a phenotype by integrating knowledge of organization, regulation and interaction of genome to create structures products and activities; Groundnut has lagged behind in use of molecular genetic technology due to the shortage of genome infrastructure, tools and resources and low levels of molecular polymorphism observed in cultivated species.

<u>Marker-assisted selection (MAS)</u>: A plant breeding tool proposed in 1923, based on principle – 'use of easy-to-score phenotypes to select difficult-to score or low heritability traits that are linked to them'; A good marker should i. permit separation of homozygotes from heterozygotes to allow more genetic gain per generation, <u>ii</u>. have early expression in the plant to save time on waiting for the desired phenotype to develop, and <u>iii</u> not have interaction with other markers; Initially breeders depended on morphological markers but these have either dominance effects, late expression of phenotype, epistatic relationship or have deleterious effect on the plant and as such do not qualify as good markers; Efficiency of MAS is dependent on quality of mapping process which requires substantially contrasting parents for target trait, precise phenotyping technique and large mapping population; Cost effective application of MAS requires one marker very close (or preferably within) to the gene of interest and two markers closely flanking either side and that these markers are based on simple robust PCR-based marker-assays; Construction of a genetic linkage map is necessary to facilitate QTL analysis and gene tagging to apply marker- assisted selection in a crop.

<u>Polymorphism</u>: Natural variation in a gene, DNA sequence, or chromosome that has no adverse effects on the individuals and occurs with fairly high frequency in general variation; Involves one of two or more variants of a particular DNA sequence; Most common polymorphism involves variation at a single base pair, can also be much longer in size and involve long stretches of DNA; Essential for marker-assisted molecular breeding; Abundant polymorphism in wild *Arachis* species but only limited or moderate polymorphism in cultivated groundnut.

<u>Reasons for abundant morphological variation but limited polymorphism</u>: Morphological traits are altered by one or a few major genes with expression influenced by modifiers and epistatic interactions and intense selection pressure under cultivation results in diversification; Variation for biochemical and molecular markers, which are not subject to direct selection, often decreases during domestication; Apparent lack of variation at the molecular level could be due to isolation of cultivated groundnut from other *Arachis* species soon after polyploidization of the single hybridization event between *Arachis duranensis* and *Arachis ipaënsis* followed by successive selection during breeding efforts.

<u>Quantitative trait loci (QTLs)</u>: Stretches of DNA containing or linked to the genes that underline a quantitative trait; Identification and mapping of QTLs required for marker-assisted breeding.

<u>Molecular markers</u>: Have great potential for increasing breeding efficiency because they have a) large numbers of polymorphisms, b) alternate alleles rarely deleterious at the molecular or whole plant level, c) often co-dominant, d) allow all genotypes to be distinguished in each generation, e) rarely segregate in epistatic ratio, f) scoring of molecular markers not dependent upon gene expression, g) not affected by environment, h) an accurate genotype can be established using any plant tissue at any development stage, i) are useful in pyramidizing desirable genes and in tracking them through generations of backcrossings, and j) reduced requirement of the time and space necessary to evaluate plant population; However, a large number of markers must be evaluated for association with various traits and the linkage between markers and desired traits must be known for effective breeding. Different types of markers are described below.

<u>Isozymes</u>: Any two proteins that catalyse the same biochemical reaction but differ in chemical composition; Earliest molecular marker systems used for plant analyses based on polymorphisms in enzyme mobility; Found not useful in characterizing polymorphism in cultivated groundnut.

<u>Restriction fragment length polymorphisms (RFLPs)</u>: First marker system that had a large number of polymorphisms; Widely used both to create linkage maps and implement indirect selection strategies; Can be used to study both recessive genes and multiple alleles; Being co-dominant, rapidly identifies homozygous individuals; RFLPs produced by digesting DNA with restriction endonucleases that recognize specific DNA sequence and then cleave the DNA strand in or near the sequence; Radioactivity is used to label the probes and bands are visualized in an auto radiograph; Disadvantage – cost and time involved in the assay; Little variation detected by RFLPs in cultivated groundnut but significant variation observed among wild *Arachis* species.

<u>Random amplified polymorphic DNA (RAPD)</u>: A polymerase chain reaction (PCR) – based marker system that detects only dominant markers; Requires only small amount of DNA to screen large number of markers, does not require radioactivity, very sensitive to polymorphism and easy and rapid to perform; Very little variation detected in cultivated groundnut but large amount of genetic variation detected among wild *Arachis* species.

<u>Sequence characterized amplified regions (SCARs)</u>: Also a PCR-based marker system, SCAR is a genomic DNA fragment at a single genetically defined locus that is identified by PCR amplification using a pair of specific oligonucleotide primers; Superior to RADP markers as it is less sensitive to reaction conditions, does not require radioactivity, can be used as co- dominant genetic markers; Dominant SCARs may be used as a quick plus/minus assay for a particular product.

<u>Amplified fragment length polymorphisms (AFLPs)</u>: Also a PCR-based dominant marker system that combines benefits of RFLPs and RAPDs; Like RAPDs no knowledge of nucleotide sequence required; About 10-fold increase over RAPDs in number of loci screened; Showed less variation in cultivated groundnut.

<u>Diversity arrays technology (DArT) markers</u>: A DNA segment, present or absent in a defined genomic representation, depending upon the individual genotype; Many methods available to produce a genomic representation; Not very useful in genetics and breeding applications.

<u>Simple sequence repeats (SSR) or microsatellite markers</u>: More variable than other marker systems; Co-dominant and easily detected from relatively little amount of DNA after PCR amplification; Higher levels of polymorphism particularly with longer TC motif repeats; Transferable among related species; Currently preferred markers of choice in groundnut as they work easily in the tetraploids; More than 3000 SSR markers available, many of these SSR markers developed from ESTs.

<u>Single nucleotide polymorphism (SNP) markers</u>: Most common type of genetic variation, each SNP represents a difference in a single nucleotide, highly amenable to high-throughput genotyping

approaches.

Linkage maps: Dense genetic linkage maps essential for wide spectrum of genetics and breeding applications such as linkage mapping or association analysis based trait mapping, marker-assisted breeding, map-based cloning and physical map alignment; <u>Reference genetic maps</u> developed both in diploid AA and BB and in tetraploid genomes, created enabling comparison of different groundnut maps and even allowing alignment of maps with other legume species; Marker density, however, is not very satisfactory especially in the context of large genome size (2800Mb/1C) and 20 linkage groups (LGs); Using marker segregation data for 10 RILs and one BC population from the international groundnut community, with the help of common markers across different populations, a reference consensus genetic map of cultivated groundnut developed: Map is comprised of 913 marker loci including 911 simple sequence repeats (SSRs) and 2 cleaved amplified polymorphic sequence (CAPS) loci distributed on 20 LGs spanning a map distance of 3,607.97 cM with an average map density of 3.94 cM; This reference consensus map will serve as a reliable reference for aligning new genetic and physical maps, performing QTL analysis in a multi- populations design, evaluating the genetic background effect on QTL expression, and several other genetic and molecular breeding activities in groundnut. As more research groups are getting involved in genomics of groundnut, the linkage map is getting saturated fast.

<u>Targeting Induced Local Lesions IN Genomes (TILLING)</u>: A reverse genetic technique that requires knowledge of gene sequences since mutants are detected by screening for DNA sequence changes rather than phenotypic differences (forward genetics); Used to find genes of interest in a mutant population of a species, to enhance genetic diversity available for exploitation and in functional genomic studies; A tilling population of over 3400 mutant lines from Tifrunner groundnut cultivar generated using chemical mutagenesis and screened for mutation in six genes.

<u>Expressed Sequence Tags (EST)</u>: Important transcriptome resources for major crops including crops with large genomes (such as groundnut), where genome sequence is not available, to enable gene discovery, microarray gene expression analysis and molecular marker development and genetic map construction; Many plant EST libraries sequenced as an alternative to whole genome sequencing where genome is complex and large; NCBI EST database contains 225,264 ESTs from groundnut as of Nov 2011; ESTs will complement the whole genome sequence.

<u>BAC libraries</u>: Large insert genomic DNA libraries such as bacterial artificial chromosome (BAC) libraries provide a platform for physical mapping, map-based cloning of the genes for traits of interest, analysis of gene structure and function and genome sequencing; First BAC library with 182,784 clones developed for tetraploid groundnut cultivar Florunner; Recently two BAC libraries for AA and BB genomes.

<u>Genome sequence database</u>: Peanut Genome Project (PGP) has fully sequenced the groundnut genome in 2014 and is now engaged in the development genomic resources for use in groundnut improvement (http://www.peanutbioscience.com/peanutgenomeproject.html).

<u>Mapping populations</u>: Three primary types of populations used for molecular mapping –  $F_2$ , backcross, and recombinant inbreds; These are created from  $F_1$  lines that are derived from two parents that show differing phenotypes for a target trait; Members of a fixed mapping population contain differing amount of recombination and linkage disequilibrium between loci; Population is genotyped with molecular markers and linkage analysis performed to estimate linkage map for population.  $F_2$  and backcross populations are not eternal and will have to be created each time when DNA is needed; Recombinant inbreds are eternal as they are maintained by single seed descent method and are available any time to harvest DNA.

<u>Recombinant inbred lines (RILs)</u>: RILs are the offspring of two genetically distant parents (biparental population) that are inbred either through self-fertilization or by sibling mating; They have a little bit of each parent genome (due to recombination) but are homozygous at any given point in the genome so dominance effects do not need to be considered in subsequent genetic and phenotypic analyses.

<u>Multi-parents advanced generation intercross (MAGIC) population</u>: Population consisting of RILs originating from multi-parents crossing; Magic populations provide a useful germplasm resource with diverse allelic combinations for precise QTL mapping and for direct and indirect use in variety development; They allow parallel high resolution mapping of different complex traits in the same population.

<u>Chromosome segment substitution lines (CSSLs)</u>: CSSLs, which carry a particular chromosome segment from a donor line in the genetic background of another line, are useful populations for detection and mapping of QTLs for target traits; These can be used for detection of QTLs with small additive effects that are masked by QTLs with larger effects in primary populations such as  $F_2$  and RIL populations; These are powerful QTL mapping populations to elucidate the molecular basis of interesting traits of wild species. These are developed by recurrent backcrossing followed by selfing (e.g. BC<sub>3</sub>F<sub>4</sub>).

<u>Comparative mapping</u>: Comparative genomics is an useful approach to define common attributes among species showing close phylogenetic relationship; Knowledge of genome structure and gene function gained from one species can be used to study the other related species; In legumes the knowledge of genome structure and gene function gained from *Glycine, Medicago* and *Lotus* can be applied to other lesser studied legume species; This will allow breeders to mine desirable genes from germplasm collection rapidly and cost effectively.

<u>Foreground and background selections</u>: Foreground selection refers to using markers that are tightly linked to the gene of interest in order to select for the target allele or gene. Background selection refers to using markers that are not tightly linked to the gene of interest in order to select against other DNA from the donor parent (i.e., to select for recurrent parent alleles at other loci than the target).

<u>Status of marker-assisted breeding in groundnut</u>: Molecular markers associated with resistance to rust, early leaf spot, late leaf spot, Cylindrocladium black rot, tomato spotted wilt virus (TSWV), nematode, aphids and drought, yield parameters, high oleic acid and other seed biochemical traits identified; Many of these QTLs not major (account for <10% phenotypic variation); Major QTLs identified for rust, late leaf spot and nematode resistance may be of wild species origin; Genomic selection (GS) now preferred over marker-assisted backcrossing (MABC) and marker-assisted recurrent selection (MARS) approaches for improving complex traits.

<u>Root knot nematode</u>: First example of MAS in groundnut breeding – transfer of a single dominant root knot-nematode resistance gene from *Arachis cardenasii* into a variety named Nema TAM in the USA; However, Nema TAM found not suitable for cultivation in Southeastern USA due to its susceptibility to TSWV; Tifguard, which combines resistance to root-knot nematode and TSWV, was developed following conventional breeding approach; Tifguard High O/L, which combines resistance to root-knot nematode and TSWV and high O/L, was developed following three cycles of backcrossing using as pollen donors  $BC_nF_1$  progenies selected with molecular markers for these traits; Use of co-dominant marker allowed backcrossing with heterozygous lines for high O/L ratio, which is a recessive trait, selfed  $BC_3F_2$  plants yielded marker-homozygous individuals identified as Tifguard High O/L; Use of MAS backcross breeding compressed hybridization and selection phases to less than 3 years. <u>Foliar diseases</u>: With markers (RAPD, EST-SSR and SSR) and QTLs associated with rust and late leaf spot resistance already identified, marker-assisted breeding efforts are in progress; Seven markers linked to LLS severity score, which explain 32–59% phenotypic variations, identified; Two EST-SSR markers closely linked to rust resistance identified; Soon leaf spot and rust resistant groundnut cultivars developed through MAS would become available for cultivation by farmers.

<u>Aflatoxin contamination</u>: Six QTLs, each located on a different linkage group, for resistance to *Aspergillus flavus* invasion explaining 6.2-22.7% phenotypic variation reported.

<u>Drought</u>: Identification of few major, many minor M-QTLs and QTL x QTL interactions indicates the complex and quantitative nature of drought tolerance in groundnut and suggest use of marker-assisted recurrent selection (MARS) or genomic selection (GS) instead of marker-assisted backcrossing approach in breeding to introgress a large number of QTLs associated with drought resistance.

<u>Tomato spotted wilt virus</u>: Two major QTLs for resistance to TSWV identified.

<u>Insect pests</u>: Three QTLs for component traits associated with bruchid resistance explaining 14–39% phenotypic variation reported.

<u>High oil/oleic acid content</u>: Three SSR alleles associated with high oil content in wild *Arachis* species are absent in cultivated groundnut; Using wild *Arachis* species the oil content in cultivated groundnut can be increased; One tightly linked marker each for high and low oil content reported.

Tifguard High O/L cultivar developed after three accelerated backcrossing combining nematode resistance from Tifguard and high O/L trait from Georgia-02C and Florida-07 following marker-assisted selection.

## **10. Genetic transformation**

Creates new genetic diversity for exploitation in cultivar development and provides opportunity to combine conventional resistance with nonconventional resistance harnessed from unrelated organism; Genetic transformation adopted only when a reliable tissue culture system to regenerate plants exists; Regeneration highly influenced by genotype, media, light, temperature and growth regulators; Now reliable regeneration protocols for groundnut available; Gene promoters are the most critical element for obtaining gene expression in groundnut; Till date (2014) no groundnut transgenic cultivar released in the world; Public resistance to transgenic food crops, particularly in Europe, heavy cost of meeting regulatory requirements for the release of transgenic cultivars including food safety and issues related to patented technologies come in the way of field testing and release of transgenic cultivars; Product of all breeding approaches is a genetically modified genotype when compared to its predecessor genotype (GMO), the better term for product of genetic transformation is transgenics; The first transgenic food came in the market in 1996; In 2012, transgenic crops covered 1.5 billion ha globally; Most of soybean, cotton, maize and canola in the USA and other countries are transgenic.

<u>Cisgenesis</u>: The process of engineering a genotype with desired genes transferred artificially between organisms that could otherwise be conventionally bred; Unlike transgenics, genes are transferred between closely related organisms; Gene belongs to conventional gene pool, a cis plant contains no foreign gene; Has been used to transfer natural resistance genes to blight in potato and scab in apple.

<u>Binary vector</u>: A pair of plasmids consisting of a binary plasmid and a helper plasmid – the two plasmids used together to produce genetically modified plants; Both artificial vectors created from

T1 Plasmid found in *Agrobacterium tumefaciens*.

<u>Agrobacterium-mediated transformation</u>: Groundnut tissues susceptible to infection by *Agrobacterium tumefaciens* include – leaf sections, cotyledonary nodes, longitudinal cotyledon halves, embryo axes, embryo leaflets, and hypocotyls; A target gene of interest engineered into the T-DNA region of a disarmed plasmid and introduced into *Agrobacterium tumefaciens*; Transgene within the T-DNA borders further transferred into plant cells by co-cultivation of *Agrobacterium tumefaciens* and wounded plant tissue in appropriate medium; PCR, which can amplify a few molecules of the gene from residual Agrobacterium because of its very sensitive nature, Southern Blot assay, where the digesting enzyme cuts at only one site or not at all within the introduced plasmid, should be used to confirm gene integration and stable transformation; It is an efficient and rapid technique for transformation due to circumvention of tissue culture step (4–5 months to obtain transgenic plants); However, technology is genotype specific and of limited use in groundnut.

<u>Microprojectile bombardment-mediated transformation</u>: Directly transfers target genes into plant cells by delivering coated microprojectiles at high velocity; Less dependent on host genotype; Microprojectile bombardment can be followed as long as groundnut can be generated from somatic tissues; It is low in efficiency, results in frequent infertility among tissue culture regenerants and takes longer (12-14 months) from induction of transformation event to plant maturity; Successfully used in transferring multiple genes for protein and RNAi-mediated gene silencing.

Steps involved and other requirements in field testing and release of transgenics in India: Department of Biotechnology, Ministry of Science and Technology, Government of India has issued two publications – Recombinants DNA Safety Guidelines and Regulations in 1990 and Revised Guidelines for Research in Transgenic Plants and Guidelines for Toxicity Evaluation of Transgenic Seeds, Plants and Plant Parts in 1998 – which give guidelines and regulatory requirements to be followed by all those engaged in recombinant DNA research and product development; Department of Biotechnology provides an oversight to all DNA recombinant research in the country; There are different levels of transgenic research; Transgenic research can be carried out only in laboratories following good practices and in specified greenhouse/glasshouse meeting prescribed standards of environmental safety; In addition to standard agronomic evaluation of transgenic material under controlled conditions on-station and in fields, biosafety tests are also required; Toxicity evaluation of transgenic seeds, vegetables and leaves involves oral toxicity tests using rats and goats and skin irritation tests involving rabbits and guinea pigs; Allergenicity tests are to be carried out in animal model.

<u>Status of transgenic material</u>: First successful transformation and accompanying plant generation using microprojectile bombardment technique in groundnut was achieved in 1993 in the USA; In most cases, the level of resistance achieved through transgenic is more or less similar to that achieved through conventional breeding; However, transgenics do provide opportunity to combine conventional resistance with that of nonconventional resistance to improve the level of protection against pathogen or stress factors.

<u>Virus diseases</u>: Received more attention than fungal diseases in transgenic research; Coat proteinmediated resistance, in general, offered only moderate protection; RNAi-mediated resistance expected to offer higher levels of protection.

Peanut bud necrosis disease (PBND): Both *Agrobacterium tumefaciens*-- and microprojectile-mediated genetic transformation approaches using PBNV nucleocapsid gene encoding for viral coat protein pursued at ICRISAT; Transgenic events with PBNVnp gene showed lower incidence and delayed onset of disease and also recovery from disease suggesting only a modest tolerance to PBNV; Currently, RNAi-mediated approach being followed to counter the effect of non-- structural silencing suppressor gene (NSS gene) in the PBNV genome.

Tomato spotted wilt virus disease (TSWV): Protection of transgenic plants against TSWV under control of both RNA – and protein-mediated control; Nucleocapsid protein gene (NP) introduced via microprojectile bombardment into New Mexico Valencia A cultivar and a runner cultivar; *Agrobacterium tumefaciens*-mediated transformation also followed; AT 120 (with antisense nucleocapsid gene) and Marc 1 (with coat protein gene) cultivars also transformed; Expression of sense or antisense NP gene from TSWV delayed expression of symptoms and prevented systemic virus infection but did not provide complete resistance to the disease; This single gene resistance may be short-lived because of highly heterogeneous population of the virus.

Peanut stripe virus disease (PStV): Transgenic plants of Gajah and NC 7 cultivars containing one of the two forms of PStV coat protein gene (cp 2 or cp 4) exhibited high levels of resistance to PStV; Mechanism of resistance appears to be RNA-mediated.

Peanut stem necrosis disease (PSND): Transgenics produced following *Agrobacterium tumefaciens*mediated transformation with TSV coat protein gene (TSVcp gene) showed three symptoms – blockage of systemic movement of TSV within the plants, recovery from an initial infection and subsequent new growth devoid of TSV symptoms and susceptible reaction; Transgenic lines cv. JL 24 containing sense and antisense coat protein gene of TSV using *Agrobacterium tumefaciens*mediated transformation developed; However, these lines yet to be tested for disease reaction at hotspot locations under field conditions.

Peanut clump disease: Transgenic lines having  $IPCV_{cp}$  and  $IPCV_{rep}$  genes of Indian peanut clump following *Agrobacterium tumefaciens*-mediated transformation produced and tested under containment facilities at ICRISAT; Some events showed resistant phenotype where the virus titre declined with maturity.

Groundnut rosette disease (GRD): Pathogen-derived resistance (introduction of GRAV or GRV genomic sequences or genes or SatRNA-derived sequences that down regulate GRV replication) a potential strategy for controlling GRD through the generation of transgenic plants; Groundnut transgenics having  $\text{GRAV}_{cp}$  gene developed at ICRISAT and currently being tested in South Africa (as the disease does not occur in India).

<u>Fungal diseases</u>: Several genes (glucanase, chitinase, SniOLP and Rs-AFP2 for late and early leaf spots, chitinase for rust, oxalate, glucanase and chitinase for sclerotinia blight and Stilbene synthase, glucanase, chitinase, mod 1, anionicperoxidase, synthetic peptide D4E1, LOX 1, Nonheme chloroperoxidase (cpo) and Pn LOX 3 for *Aspergillus flavus* infection and aflatoxin biosynthesis) used in genetic transformation; These genes suppressed the disease, delayed the onset of disease, enhanced resistance and decreased disease incidence; In the case of sclerotinia blight, reduced lesion area and in the case of *Aspergillus flavus*, reduced aflatoxin production also noticed.

<u>Insect pests</u>: Synthetic genes, cry1 EC against *Spodotera litura*, cry1 X against *Heliothis armigera* and *Spodotera litura*, and cry1 Ac against lesser cornstalk borer showed good promise.

<u>Drought and salinity tolerance</u>: a) A cis-acting transcription factor that binds to dehydration responsive element (DRE) from Arabidopsis thaliana, AtDREB1 A, under the control of a stress inducible promoter from the rd29A gene used to develop transgenic lines with drought tolerance; One transgenic line showed 40% increase in transpiration efficiency; b) Transgenic lines of groundnut transformed with AtNHX1 showed drought and salt tolerance; c) Transgenic lines transformed with isopentenyltransferase (IPT) gene driven by SARK promoter showed improved biomass retention and an average of 58% yield increase.

### Nutritional quality:

Vitamin A: Zmpsy 1 gene from maize and beta-lycopene cyclase gene from tomato used to enrich groundnut seeds with vitamin A; Second generation transgenic events showed many fold increase in vitamin A content.

Oleic/Linoleic fatty acid ratio: Transgenic event with an FAD2 gene RNAi construct showed reduced content of linoleic acid and increased stability of groundnut oil.

Allergens: Endogenous allergens, Ara h 2 and Ara h 6, silenced by introducing RNAi construct targeting homologous coding sequence; Human IgE binding to these proteins was greatly reduced.

<u>Herbicide tolerance</u>: Transgenic groundnut expressing human Bcl-xL gene showed improved tolerance to paraquat; Transgenic groundnut overexpressing pEGAD-EPSPS with altered kinetics of enzyme showed improved tolerance to glyphosate (Roundup<sup>TM</sup>).

## **11. Current status and future needs in groundnut breeding**

In the past, market forces, changing climate and economic considerations have caused shift in varietal scenario in a given region; Prior to 1940, Spanish types covered about 90% area in Georgia in the USA, now 100% area is covered by Runner types due to their good flavor, better roasting characteristics (suitable for peanut butter) and higher yields; On the other hand, Runner types have been replaced by the Spanish types due to frequent and unpredictable droughts and difficulty in their harvesting in Southern India.

From a large collection of more than 15,000 accessions of cultivated groundnut germplasm, only a limited number, mostly sources of resistance to diseases and insect pests, has been utilized in crop improvement; Main reasons for this limited utilization of resistance sources are their poor agronomic background causing linkage drag, absence of information on their reaction to other biotic and abiotic stresses and lack of information on their allelic relationship and combining ability; Keeping the requirement of consumers and processors with respect to taste and flavor and seed size and color, breeders in most applied breeding programs prefer to re-use improved breeding lines in hybridization.

<u>Yield and yield related traits</u>: Newer high-yielding cultivars allocate a great proportion of dry matter to reproductive tissue early in the growth cycle with greater reproductive efficiency than the older cultivars; Newer releases in the USA also have more spreading growth habit and greater seed and pod weight than the earlier releases; In India also, the increase in yield in newer Spanish releases was largely due to gain in seed size, seed weight and number of pods.

Selection either for higher pod yield or for greater harvest index essential for improvement of yield potential in future cultivars; Remobilization of reserves from vegetative dry matter to pods under conditions of source limitations (falling temperature, defoliation by pathogens or water stress) likely to be significant in maintaining yields but may limit response to improved conditions specially with high partitioning.

Un-constricted and deeply constricted pods – commercially unacceptable, the former with flat seed ends make blanching difficult and their seeds tend to split during shelling out operation which may damage embryo, in the case of latter, harvesting and shelling out is difficult; Strongly reticulated pods also not preferred as they do not come out clean at the time of harvesting.

<u>Genetic gains in groundnut breeding</u>: Between 1944 and 1987, the average yearly genetic gain for yield in Virginia market type cultivars in the USA – 14.7 kg/ha; When the emphasis in groundnut breeding in the USA shifted to pest resistance, earliness and quality, the new cultivars improved

upon these traits while maintaining the yield but failed to combine them with genes for increased yield potential; During 1980s and mid-1990s, the groundnut yield in India increased by 1.4% per annum; Increase of 0.43% per year in seed yield, of 0.29% in seed weight and of 0.52% in pod growth rate during 1948-2004 obtained in Argentina.

There still remains a huge gap between realized and potential yield of groundnut in most developing countries; Breeding programs in developing countries should aim at bridging this gap in yield through resistance breeding against biotic and abiotic stresses.

<u>Resistance to foliar fungal diseases</u>: Early leaf spot (caused by *Cercospora arachidicola*, ELS), late leaf spot (caused by *Phaeoisariopsis personata*, LLS), and rust (caused by *Puccinia arachidis*) are most widely distributed and economically important foliar diseases of groundnut; Only one leaf spot dominates in a region, however, both pathogens can be observed in the same field; Infector-row technique used to screen germplasm and breeding materials in the field; Combining high levels of resistance to early and late leaf spots into high yielding cultivars with acceptable market traits continues to be difficult; Resistance to leaf spots generally associated with late maturity, longer incubation and latent periods, reduced sporulation, smaller lesion diameter, lower infection frequency and less defoliation in resistance sources; Level of resistance to foliar diseases very high in wild *Arachis* species, interspecific derivatives confer higher levels of resistance to ELS and LLS inherited independently in wild species.

ELS: More serious in southern Africa and the USA; Tolerant *Arachis hypogaea* genotypes identified in Malawi (4), western Africa (18) and India (15) with a disease score between 3.6 and 6.3 on a 1-9 scale where 1=no disease and 9=more than 81% foliage destroyed; Resistant sources reported from the USA (NC 3033, PI 270806, PI 259747 and PI 350680) found susceptible in India and Malawi; Genotypes ICG 6284, ICG 6902, ICG 7878, ICG 10000, ICG 10948 and ICG 13917 show some resistance at more than one location; Rate-reducing resistance quantitative in nature and controlled predominantly by additive gene effects; Narrow sense heritability varies from low to high; Resistant wild *Arachis* species among others include – *Arachis cardenasii, Arachis chacoense, Arachis stenosperma, Arachis appressipila, Arachis pusila, Arachis kempff-mercadoi, Arachis batizocoi, Arachis diogoi, Arachis hagenbeckii, Arachis glabrata and Arachis repens; ELS tolerant cultivars reported, among others, in India – ICGS 44, ICGS 76, M 335, BG 3, Somnath, CSMG 84–1, M 522, Prutha and GG 7 and in the USA – Georgia-01R and Georgia-05E.* 

LLS: Sixty-nine *Arachis hypogaea* genotypes tolerant to LLS with disease score ranging between 3 and 5 on a 1-9 scaleidentified – 49 of these landraces belong to var. *peruviana* with low yield and poor shelling outturn; High levels of resistance/immunity in wild *Arachis* species – *Arachis cardenasii, Arachis chacoense, Arachis stenosperma, Arachis repens, Arachis appressipila, Arachis hagenbeckii, Arachis glabrata, Arachis batizocoi, Arachis duranensis, Arachis correntina, Arachis villosa, Arachis pusila, Arachis diogoi, Arachis kempff-mercadoi and Arachis paraguariensis; Some tolerant cultivars released: India – RG 141, ICG(FDRS) 4, ICGV 86590, ICGV 86325, K 134, Girnar 1, GBPD 4, R 8808, ALR 1, ALR 2, ALR 3,BSR 1, VRI 5 and CSMG 84-1 among others, USA – Southern Runner, Florida MDR 98, C-99R, Georgia-01R and Georgia-05E.* 

Rust: One hundred and sixty-nine *Arachis hypogaea* genotypes reported resistant (a score of five or less on a 1-9 scale); Of these 135 landraces belong to var. *peruviana* – many (ICG 7896, ICG 7897, ICG 7899, ICG 10014, ICG 10030, ICG 10052, ICG 10053, ICG 10067, ICG 10933, ICG 10939, ICG 10940, and ICG 10943) have a disease score of <3 but are agronomically poor (low shelling outturn, thick pod shell, strong pod reticulation and unacceptable seed coat color); New sources of resistance – ICG 10056, ICG 10567, ICG 10925, ICG 10932, ICG 11108, ICG 12059, ICG 12112, and ICG 12113 and the interspecific derivatives involving *Arachis batizocoi* and *Arachis duranensis* – have high levels of resistance with good agronomic potential and resistance to other biotic

stresses; Several other species belonging to section *Arachis* are either immune or highly resistant – *Arachis cardenasii, Arachis chacoense, Arachis stenosperma, Arachis repens, Arachis appressipila, Arachis hagenbeckii, Arachis glabrata, Arachis batizocoi, Arachis duranensis, Arachis correntina, Arachis villosa, Arachis pusila, Arachis diogoi, Arachis kempff-mercadoi, Arachis paraguariensis, Arachis villosulicarpa, and Arachis spegazzinii; Some resistant cultivars released in India – ICG(FDRS) 4, ICG(FDRS) 10, ICGV 86590, GBPD 4.* 

Multiple resistance to foliar diseases: Resistance to rust and LLS is correlated (r=0.48-0.60); Fortytwo LLS resistant genotypes also resistant to rust – ICG 1703, ICG 4995, ICG 10920 and interspecific derivative ICG 13917 [259-2 (red)] useful in multiple resistance breeding, the last one being resistant to all three pathogens; Other useful sources of resistance to both LLS and rust with agronomic potential: *Arachis hypogaea* genotypes – ICG 6330, ICG 7884, ICG 10023, ICG 10035 and ICG 11182, interspecific derivatives – ICG 11312, ICG 11317 (also resistant to ELS), ICG 11321, ICG 11325, ICG 11337, ICG 13916, ICG 13917, ICG 13919, ICG 13920 and ICG 13922; Several wild *Arachis* species have very high levels of resistance or immunity to all the three foliar diseases.

Future needs: Levels of resistance to leaf spots (both ELS and LLS) in cultivars need further improvement; As LLS and rust often occur together, a combined resistance to these pathogens required; However, often with higher levels of resistance, crop duration is enhanced; Higher levels of resistance to foliar fungal diseases required in short-duration cultivars without affecting their duration; Similarly, linkage drags (poor pod shape, unattractive seed coat color, thick shell, low partitioning, etc.) need to be eliminated completely in resistant cultivars; Desirable to intermate second or third generation advanced breeding lines originating from different parents including interspecific derivatives to improve the level of resistance against foliar diseases without bringing in linkage drags.

<u>Resistance to soil-borne diseases</u>: Breeding for resistance to soil-borne fungal diseases continues to be a difficult task as creating uniform disease pressure in disease screening nursery remains challenging; Among the soil-borne diseases, *Aspergillus flavus* infection and aflatoxin contamination receives the maximum attention; Other important soil-borne diseases include white mold, cylindrocladium black rot (CBR), sclerotinia blight, collar rot and dry root rot/dry wilt.

Aspergillus flavus/ Aspergillus parasiticus seed infection and aflatoxin production: Three barriers to Aspergillus flavus/ Aspergillus parasiticus seed infection and aflatoxin production - pod wall, seed coat and cotyledons; Resistance to pod infection attributed to shell wall structure and that of seed coat to thickness and density of palisade layers, absence of fissures and cavities and presence of wax and cutin layers on the seed coat; Three resistance mechanisms - preharvest resistance, seed coat resistance (*in vitro* seed colonization (IVSC)) and cotyledon resistance (aflatoxin production), their independent inheritance provides opportunity for gene pyramiding; Sources resistant to preharvest infection (≤ 2%; 21 genotypes -- ICG 1122, ICG 1173, ICG 1323, ICG 1326 (J 11)\*, ICG 1859, ICG 1994, ICG 3263 (U 4-47-7)\*, ICG 3267, ICG 3336\*, ICG 3700\*, ICG 4589, ICG 4749 (PI 337394 F)\*, ICG 4888, ICG 7633 (UF 71513)\*, etc.; \*consistent across locations), IVSC (≤ 15% seed colonized; PI 337394F\*, PI 337409\*, UF 71513, Ah 78223, J 11\*, U \$-47-7, Var 27, Faizpur, Monir 240-30 etc.; \*consistent across locations and pathogen pressure) and aflatoxin production (<0.7 μg per kg: ICG 10609, ICG 11682, ICG 10615, ICG 6760, ICG 9610, etc.) available, but none of these is completely free from infection or aflatoxin production; Containment of preharvest infection essential as once infected, the seed cannot be disinfected and the infection is carried forward, seed coat resistance provides postharvest protection in storage, the ultimate aim is freedom from aflatoxin production; Recommended for use in breeding because of their multiple resistance - ICG 1326, ICG 1859, ICG 3263, ICG 3336, ICG 3700, ICG 4749, ICG 7633, ICG 9407, ICG 9610, ICG 10094, etc.; Drought predisposes groundnut to aflatoxin contamination, some drought tolerant lines also show low preharvest seed infection and aflatoxin production; Fatty acid composition

and N<sub>2</sub> fixation and related traits also reported to influence directly or indirectly aflatoxin contamination; Aspergillus flavus sick plot with imposed end-of-season drought during the dry season to avoid interference from rains used for screening germplasm and advanced breeding lines at ICRISAT; In the USA, a large-scale field system at a desert location (Yuma, Arizona) with subsurface irrigation, which allows extended drought in pod zone without causing any plant mortality, developed and used for screening against *Aspergillus flavus* infection and aflatoxin production; Important to have more number of replications as plot to plot/ plant to plant variation in preharvest infection could be large in spite of sufficient number of fungal propagules being present in the soil throughout the disease screening nursery; Screening for resistance to in vitro seed colonization and aflatoxin production done in laboratory following protocols prescribed by various researchers; During field and laboratory screening, not unusual to find nil preharvest infection but presence of aflatoxin in the same genotype, the reverse also observed; Conventional breeding alone does not ensure complete freedom from aflatoxin contamination, at best it is able to combine the level of resistance available in resistant parents with high yield and other agronomic characters; Level of resistance in breeding line similar to that of resistance sources, gene pyramiding has also not changed the situation; Elite breeding lines giving good performance in India and Mali / Niger include - ICGV 88145, ICGV 89104, ICGV 91278, ICGV 91283, ICGV 91284, ICGV 87084, ICGV 87094 and ICGV 87110 and in China include ICGV 95440, ICGV 95422, ICGV 94435, ICGV 95435 and UF 71315.

Future needs: Sampling procedures and screening methods including development of a sick plot need to be further refined to improve uniformity of infection, characterization and precision of estimation of infection and aflatoxin production in a genotype in a consistent manner; Allelic relationship among resistant sources to ascertain if they possess different resistance genes; Marker-assisted selection likely to improve efficiency of breeding; Similarly, genetic transformation also has potential to produce aflatoxin free genotypes.

White mold: White mold or stem rot caused by *Sclerotium rolfsii* is wide spread in major groundnut growing areas in the world; Genetic variation for resistance to white mold exists in cultivated groundnut; Sources of moderate resistance – NC 2, NC Ac 18016, NC Ac 18416, ICG 15233, ICG 15234, ICG 15235, ICG 15236, ICGV 96590 and ICGV 87160 identified at ICRISAT and NRCGCS 47, NRCGCS 99, NRCGCS 131 and NRCGCS 319 identified at the Directorate of Groundnut Research, Junagadh in India; Cultivars with moderate resistance released in the USA- Southern Runner, Toalson, Pronto, Georgia Browne, Sunbelt Runner, Tamrun 96, Georgia-01R and Georgia-05E; Field screening more consistent than greenhouse screening; Uniformity and level of inoculum in the sick plot enhanced by adding sterilized oat seed inoculum of *Sclerotium rolfsii*, but individual plants may still escape, 'agar disk technique' used to screen individual plants.

Cylindrocladium black rot (CBR): CBR, caused by *Cylindrocladium parasiticum*, largely reported from the USA; Screening done at naturally infested hot spot locations; Genetic variation for resistance to CBR – in general, Spanish cultivars most resistant, Valencia cultivars most susceptible and Virginia cultivars moderately susceptible (NC 8C, NC 10C, NC 12C); Complex inheritance-resistance delays the onset of epidemics rather than the rate of disease progress.

Sclerotinia blight: Sclerotinia blight, caused by *Sclerotinia miner* Jagger, important in Virginia and Oklahoma in the USA; Genetic variation for resistance to Sclerotinia blight – in general, cultivars with Spanish ancestry more resistant than those with Valencia and Virginia ancestries; Complex inheritance of resistance (dominance, epistatis and cytoplasmic factors); 'Detached shoot technique', which rely on rate of lesion growth and development, and disease infected fields (hot spot) used for screening; Interspecific lines derived from *Arachis hypogaea* x *Arachis cardenasii* cross highly resistant; Cultivars with Spanish ancestry more resistant than those with Valencia or Virginia ancestries; Spanish cultivars Toalson and Tamspan 90 have good resistance.

Collar rot: Also known as crown rot or seedling blight is caused by *Aspergillus niger*; No breeding program in progress.

Dry root rot/Dry wilt: It is caused by *Macrophomina phaseolina* (sclerotial stage known as *Rhizoctonia bataticola*); No breeding program in progress.

Future needs: Soil-borne diseases are becoming increasingly important and they require more research attention; Higher levels of genetic resistance to all soil-borne diseases needed in released cultivars; Resistance of wild *Arachis* species, particularly *Arachis* cardenasii, to Sclerotinia blight needs to be exploited to develop cultivars with higher levels of resistance.

<u>Resistance to virus diseases</u>: Of the 31 viruses from 14 genera naturally infecting groundnut, only nine are economically important in different parts of the world – peanut mottle potyvirus (PMV), peanut stripe potyvirus (PStV), peanut clump furovirus (PCV), groundnut rosette disease (GRD) complex, tomato spotted wilt tospovirus (TSWV), peanut stunt cucumovirus (PSV),cowpea mild mottle carlavirus (CMMV), cucumber mosaic cucumovirus (CMV), peanut bud necrosis tospovirus (PBNV) and tobaco streak ilavirus (TSV); However, the breeding attention is focused on PBNV, TSWV and GRD.

Peanut bud necrosis disease (PBND): PBND, caused by peanut bud necrosis virus (PBNV), is economically important in south and southeast Asia; Transmitted by Thrips palmi - virus acquired by the larvae but transmission is exclusively by adults; Not seed transmitted; Sources of resistance in cultivated groundnut - Several germplasm accessions with consistently low disease incidence in the field (ICG 848, ICG 851, ICG 852, ICG 862, ICG 869, ICG 885, ICG 2271, ICG 2306, ICG 2307, ICG 2323, ICG 2741, ICG 3042, ICG 3806, ICG 3873, ICG 5030, ICG 5024, ICG 5043, ICG 5044, ICG 6135, ICG 6317, ICG 6323, ICG 7676, ICG 7892 and others) and breeding lines, ICGV 86031 and ICGV 86388, resistance to both vector and PBNV, Other lines with resistance to PBND under field conditions include DRG 18, ICG 7812, ICG (FDRS) 10, ICGV 80325, ISSP 3, KNG 22 and PI 393516; Highly resistant wild species in section Arachis free from disease or virus in the field - Arachis duranensis (ICG 8199 (PI 468200), ICG 8956 (PI468201), ICG 11552 (PI 475882), ICG 11553 (PI 475883) and ICG 11555 (PI 475885), Arachis correntina (ICG 8132 (PI 262808)), Arachis monticola (ICG 8189 (PI 468199)), Arachis cardenasii (ICG 11564, ICG 13164 and ICG 13165), Arachis villosa (ICG 8144 and ICG 13168); Genetics of resistance - Three factors with additive gene effects responsible for low disease incidence, Resistance stable across environments, Significant GCA, SCA and reciprocal effects observed but GCA predominant, Because of significant reciprocal effects, the resistant source should be used as female parent in hybridization, Non-additive gene effects also reported for low PBND incidence, In an another study additive gene effects were found major contributors to PBND resistance besides add x add and dominance gene effects; Resistant cultivars released -- CO 3, ICGS 11, ICGS 44 (ICGV 87128), ICGS 37 (ICGV 87187), R 8808 (KRG 2), R 9251, K 134, DRG 12, RSHY 1, Kadiri 4 among others in India and Khon Kaen 6 in Thailand; Sources of resistance to vector - NC Ac 2242, NC Ac 2214, NC Ac 2243, NC Ac 2240, NC Ac 2232, NC Ac 2230 and others.

Tomato spotted wilt virus (TSWV) disease: TSWV is a major disease problem in groundnut producing areas of the USA; Transmitted by thrips species *Frankliniella fusca* (Hinds) (tobacco thrips) and *Frankliniella occidentalis* (Pergande) (western flower thrips), *Frankliniella intonsa*, *Frankliniella schultzei*, *Scirtothrips dorsalis*, *Thrips tabaci*, *Thrips palmi* and *Thrips setosus* in a persistent manner- the first two primary vectors; Not seed or pollen borne; Sources of resistance in cultivated groundnut – PI 203396 (also resistant to LLS), PI 196621, PI 339967, PI 341267; Highly resistant wild species in section Arachis – Arachis diogoi (PI 468141), Arachis helodes (PI 468144), Arachis sp. (PI 468345, PI 468370, and PI 468371) Arachis correntina; Genetics of resistance – Significant general and specific combining abilities and transgressive segregation for

TSWV reported; however, genetic mechanism of resistance not elucidated; Breeding lines derived from varieties *hypogaea* and *hirsuta* have higher resistance to TSWV; TSWV resistant/tolerant cultivars in the USA – Southern Runner, Georgia Browne, Georgia Green, Tamrun 96, Georgia Bold, Georgia Hi-O/L, Georgia 01R, Georgia-05E, Georgia-10T, C-99R, Florida MDR 98, Tifguard, Georganic (highest level of field tolerance among cultivars) and others – however, they may suffer significant damage during extreme epidemics.

Groundnut rosette disease: Groundnut rosette diseases (GRD) is confined to African continent and its surrounding islands; it is a complex of three causal agents - groundnut rosette assistor virus (GRAV), groundnut rosette virus (GRV) and a satellite RNA (SatRNA); These three agents synergistically act with each other for survival and spread; GRV is dependent on GRAV for transmission by aphid vector Aphis craccivora and SatRNA, which is responsible for rosette symptoms, is itself dependent on GRV for replication; GRV and SatRNA alone do not produce GRD symptoms; GRAV on its own can cause mild vellowing/chlorosis of leaves and can cause reduction in plant growth and yield; GRV and SatRNA must be packaged within GRAV coat protein to be aphid transmissible; GRV is dependent on its SatRNA for encapsidation in coat protein; GRV on its own produces transient symptoms only; GRV and SatRNA always found together in nature; the three agents are not seed borne; there are two variants GRD symptoms - chlorotic rosette and green rosette; Chlorotic rosette throughout sub-Saharan Africa and green rosette, which earlier was largely confined to West Africa, is now also reported from southern and eastern Africa; SatRNA responsible for symptoms variation; Resistant sources to GRD in Arachis hypogaea- several pure line selections (48-7, 48-14, 48-15A, 48-21, 48-34, 48-35, 48-36, 48-37, 48-44, 48-45 and 48-70A) from late maturing Virginia landraces in Burkina Faso; Subsequently several other sources of resistance to GRD in cultivated groundnut found including short duration Spanish types from outside Africa; All the sources of GRD resistance are resistant to GRV and its SatRNA but they are susceptible to GRAV; Resistance to GRD (effective against GRV and its SatRNA) in cultivated types is governed by two independent recessive genes which are effective against both chlorotic and green rosette; Resistant cultivars released in Africa - RMP 12, RMP 91, 69-101, KH 241D, KH 149A, RG 1, Nyanda (ICGV 93437), ICGV-SM 90704, ICG 12991, ICGV-SM 99568, ICGV-SM 99555, ICGV-SM 99557, ICGV-SM 01711, ICGV-SM 01721, Samnut 23 (ICGV-IS 96894), Samnut 21 (UGA 2) and Samnut 22 (M572.801); Resistant sources to GRD in wild Arachis species - 25 accessions in nine wild Arachis species (Arachis appressipila (ICG 8127, ICG 8945 and ICG 14860), Arachis decora (ICG 14946), Arachis diogoi (ICG 4983), Arachis hoehnei (ICG 8190 and ICG 13232), Arachis kretschmeri (ICG 8191, ICG 8216, ICG 11558 and ICG 13224), Arachis kuhlmannii (ICG 13225, ICG 14862 and ICG 14875), Arachis pintoi (ICG 13222, ICG 14855, ICG 14856 and ICG 14888), Arachis stenosperma (ICG 13171, ICG 13173, ICG 13187, ICG 13210 and ICG 14872) and Arachis villosa (ICG 13168); Arachis chacoense immune to both GRV and GRAV; Wild Arachis species may have different genes foe resistance; Sources resistant to aphid vector - EC 36892 (ICG 5240) and ICG 12991 (released as Baka in Malawi and as Serenut 4T in Uganda)- show less GRD but they are susceptible to all the three agents of GRD.

Future needs: Combining virus resistance with vector resistance to improve the levels of resistance to virus diseases; As coat protein gene in transgenics does not give complete protection against virus diseases, there is need to pursue RNAi approach to provide better protection against virus diseases; In the case GRD, unraveling the mystery of off-season survival of disease causing agents, transfer of resistance to GRAV from wild *Arachis* species to cultivated groundnut, study of allelic relationship among various sources of resistance and development of short-duration GRD resistant cultivars.

Resistance to bacterial disease:

Bacterial wilt: Bacterial wilt caused by *Ralstonia solanacearum* is a major production constraint in groundnut in southeast and East Asia; Schwarz 21, a bacterial wilt resistant variety selected from a local population, was the first diseases resistant variety released in 1925 in Indonesia; Several sources of resistance, mostly belonging to subspecies *fastigiata*, reported from Indonesia and China, some available with ICRISAT; Many bacterial wilt resistant cultivars released in China (Xiekongchung, Teishansanliyue, Yue You 13, Yue You 589, Yue You 92, Yue You 256, Yue You 200, Yue You 256, Yue You 79, Wu You 4, Gui You 28, E Hua 5, Zhong Hua 2, Lu Hua 3, Yuanza 9307) and Indonesia (Schwarz 21, Gajah, Matjan, Kidang, Banteng, Macan); ICG 1703, ICG 1705, ICG 7893, ICG 7894 and an interspecific derivative ICG 11325 resistant not only to bacterial wilt but also to rust and LLS; Resistance reported to be partially dominant involving three pairs of major genes and some minor genes, recessive resistance also reported, nucleo-cytoplasmic interaction also observed, both additive and dominant gene actions observed.

Future needs: Important to broaden the genetic base of resistance and improve the level of stable resistance by harnessing resistance genes from different sources including wild *Arachis* species; Combine resistance to bacterial wilt with resistance to rust and leaf spots; More studies on genetics and mechanisms of resistance.

<u>Insect pests resistance</u>: Breeding for resistance to insect pests lagging behind due to difficulty in large scale screening of germplasm lines and breeding populations under sporadic and variable natural insect pests pressure; Only limited screening under laboratory and field (infester rows with seeding/release of cultured insect pests population) or controlled conditions (caged/forced feeding) leading to identification of sources of resistance to insect pests, several with multiple resistances.

Sucking pests: Include mainly thrips (*Scirtothrips dorsalis, Thrips palmi, Frankliniella schultzei, Caliothrips indicus*), jassids (*Empoasca kerri*) and aphids (*Aphis craccivora*); Thrips and aphids also vector of virus diseases; Genotypes resistant to thrips and jassids reported and some are listed in sections on PBND and TSWV; High density, distribution and length of trichomes (NC Ac 2214, NC Ac 2230, NC Ac 2240) and thick leaf cuticle (NC Ac 2242, NC Ac 2243) associated with resistance to thrips and jassids; Genotypes tolerant to jassid damage: Breeding lines – ICGV 86388, ICGV 86252, ICGV 86393 and ICGV 85455, Cultivars – Georgia-01R and Georgia-05E; Antibiosis (reduced growth and fecundity) operates in the case of aphids resistance in NC Ac 343, ICG 5240 (EC 36892), ICG 12991and ICGV 86030 genotypes; Several wild *Arachis* species have very high levels of resistance to insect pests (thrips – *Arachis chacoense*; jassids – *Arachis cardenasii, Arachis duranensis, Arachis kempff-mercadoi, Arachis monticola, Arachis stenosperma, Arachis paraguariensis, Arachis pusila, Arachis triseminata, Arachis batizocoi, Arachis chacoense*); Non-additive genetic variance for all trichome characters, additive genetic variance for trichome length, jassid damage and resistance to complex of insect pests (thrips, jassids and *Helicoverpa*).

Defoliators: Major defoliators in groundnut – Leaf miner (*Aproaerema modicella*), tobacco caterpillar/ armyworm (*Spodoptera litura*) and gram pod borer (*Helicoverpa armigera*); Other defoliators include hairy caterpillars (*Amsacta albistriga* and *Arachis moori*) and Bihar hairy caterpillar (*Spilosoma oblique*); Screening for resistance to defoliators under natural field conditions difficult because of variation in infestation in space and time; No-choice cage technique is used to screen for *Spodoptera litura*, nuclear insect culture is maintained on artificial diet, a known number of first- or third-instar larvae are released for varying period of time on 15-day old greenhouse grown plants which are kept inside a plastic jar cage with wire-mesh screen windows, observations on insect survival (number of surviving larvae and larval weight) and leaf area damage are recorded; For leaf miner, natural infestation is relied upon, which can be enhanced

by planting soybean as an infester crop and creating prolonged drought, it is difficult to devise nochoice cage screening for leaf miner.

Breeding lines ICGV 86031, ICGV 87154 and ICGV 87160 and germplasm accessions ICG 2271 and ICG 1697 show resistance to both tobacco armyworm and leaf miner; Other genotypes which show promise against leaf miner – NC Ac 343, NC Ac 17090 and ICG (FDRS) 4; Several wild Arachis species also show resistance to these two insect pests – Arachis chacoense, Arachis repens, Arachis appressipila, Arachis hagenbeckii, Arachis glabrata, Arachis batizocoi, Arachis correntina, Arachis villosa, Arachis diogoi.

Future needs: Resistance genes from wild *Arachis* species should be harnessed to improve resistance to insect pests in cultivated groundnut; Field screening techniques need to be improved to create desired and uniform insect pressure.

#### Nematode resistance:

Root knot nematode: Screening involves growing plants in glasshouse and inoculating them with *Meloidogyne arenaria* race 1 inoculum cultured on roots of tomato or eggplants and scoring the roots of 70 day old harvested groundnut plants for root galls and egg masses; In contrast to moderate level of resistance in cultivated groundnut, wild *Arachis* have a high level of resistance against this nematode; Accessions supporting less egg production of *Meloidogyne arenaria* race 1 identified both in cultivated groundnut and wild species *Arachis cardenasii, Arachis batizocoi* and *Arachis diogoi*; Resistance in *Arachis cardenasii* conferred by two dominant genes – one gene inhibiting root galling and the other gene inhibiting egg production; In the USA, root knot nematode resistant variety Nema TAM was the first variety developed using MAS but it was susceptible to TSWV, subsequently Tifguard, resistant to both root knot nematode and TSWV was developed following conventional breeding; Similar to Nema TAM, COAN, the first release with resistance to M. arenaria is also susceptible to TSWV, it carries the resistance gene from TxAG 7, which is a backcross derivative of TxAG-6, a complex interspecific derivative involving *Arachis cardenasii, Arachis batizocoi* and *Arachis diogoi*.

Kalahasti malady: 'Kalahasti disease' caused by stunt nematode (*Tylenchorhynchus brevilineatus*) first noticed in 1975/76 in Kalahasti area in Andhra Pradesh, India; Disease characterized by reduction in pod size and presence of small, brownish yellow lesions on pegs and young developing pods, later pod surface and roots become completely discolored; From replicated screening of 1599 genotypes in a hot spot location in a farmer's field in Kalahasti during 1985/86 – 1986/87, 14 resistant genotypes identified; Most of these genotypes had undesirable pod/seed characteristics with the exception of TCG 1518, an advanced Virginia bunch breeding line, which was later released as Tirupati 3 for cultivation in disease-affected areas; In another screening of 39 genotypes during 1992–94, TCGS 307, TCGS 313 and TCGS 320 (released as Kalahasti) were also identified as resistant to the disease with the last two having pod yield exceeding 3 t/ha.

Future needs: The resistance to nematodes needs to be combined with resistance to other major biotic stresses operating in a region to derive larger benefits from the resistance breeding efforts.

### Physiological traits:

Drought: A majority of groundnut in developing countries is grown under rainfed conditions; Although considered a drought tolerant legume, it can suffer early-season, mid-season, end-ofseason or intermittent droughts impacting adversely on yield and yield- related characters including quality of the produce due to reduced photosynthesis,  $N_2$  fixation and calcium uptake by developing pods; Impact depends upon the timing of occurrence, duration and intensity of drought; A 20 to 26 day moisture stress soon after crop emergence is beneficial to the crop as it forces roots to go to deeper layers of soil and when the moisture stress is released it induces profuse flowering resulting in synchronized and uniform maturity and increased yield; The adverse effect of end-ofseason drought can be overcome by developing short-duration varieties with their life cycle matching the period of soil moisture availability; Mid-season drought is the cause of concern as insufficient water at the time of flowering and fruiting reduces the yield significantly.

Direct selection for yield under drought is effective but is resource consuming and lacks repeatability across different environments; Passioura (1977) gave the following equation for vield under water-limited conditions: Pod vield = Transpiration (T) x Transpiration Efficiency (TE) x Harvest Index (HI); Drought tolerance can be enhanced by improvements in soil water extraction ability (T) or improvements in water-use efficiency (TE); Genetic variation for root system and transpiration efficiency (amount of dry matter produced for each unit of water transpired (g dry matter per kg of water transpired) is reported, but these traits are difficult to measure; Easily measurable surrogates for these traits are needed for use in a large scale breeding programs; Transpiration efficiency is negatively correlated with  $\Delta^{13}C$  (carbon isotope discrimination) in leaves (r= -0.88 to -0.92), which is rapid but expensive to measure;  $\Delta^{13}$ C is highly positively correlated with specific leaf area (SLA, ratio of leaf area to leaf dry weight), which is easy and inexpensive to measure; SLA has inverse relationship with relative leaf water content (RWC) and the low SLA types are drought tolerant as they are able to maintain higher RWC; However, SLA is influenced by the time of sampling and age of the leaf; SLA is inversely correlated with SPAD chlorophyll meter reading (SCMR) (r= 0.77), which, in turn, is positively correlated with TE; SCMR is measured by a hand held device, which is easy to operate and can rapidly record observations; Thus, for rapid screening, SCMR can be used in a large scale breeding programs aiming to improve drought tolerance in groundnut; SLA and SCMR can be recorded any time after 60 days of crop growth, preferably under moisture deficit conditions; However, the utility of SLA and SCMR in screening for drought tolerance has been questioned in some studies.

Sufficient variation for physiological traits such as SLA, WUE, T, TE and HI and in tolerance to midseason and/ or terminal droughts is reported; High heritability for HI, SCMR and  $\Delta^{13}$ C and medium to high heritability for SLA are reported; Both additive and additive x additive gene effects for SLA and HI and additive gene effects for  $\Delta^{13}$ C are reported. Field screening for tolerance to different droughts is done in dry season by withholding irrigation as follows: for early-season drought by withholding irrigation for 40 days after the first irrigation for germination and subsequently following normal irrigation schedule, for mid-season drought withholding irrigation after 40 days up to 80 days and before and after drought period following regular irrigation schedule, and for end-of-season drought withholding irrigation from 80 days onward till maturity and following normal irrigation schedule prior to imposition of drought; For germplasm screening, line-source sprinkler system of irrigation is used to create a gradient of soil moisture deficit and the genotypes are selected based on biomass and HI; Segregating populations are screened in the field under imposed drought conditions and selections are based on pod yield, pod number and pod filling; Surrogates SCMR or SLA can also be used along with pod yield and other characters in selection of plants/populations; Both empirical (yield-based) and trait-based approaches are equally effective in selecting for drought tolerance; In the case of trait-based approach, TE is the major contributor to pod yield, which indicates more efficient utilization of available water; However, in the case of empirical approach, it is T which is a major contributor to pod yield, which indicates better mining of water from soil layers; However, better mining does not necessarily mean better utilization of water; In case of limited water availability, enough T may not occur thus impacting on pod vield; It is advisable to integrate surrogates of TE in the selection scheme for drought tolerance.

Some of the drought tolerant cultivars/genotypes are ICGS 11, ICGS 44, ICGS 76 and ICG(FDRS) 10 for mid-season drought and TPT 25 for both early- and mid-season drought in India and 55-437 (also tolerant to aflatoxin contamination), GC 8-35, 55-21, 55-33, TS 32-1, SRV 1-3, SR 1-96, ICGV

86024 (also tolerant to leaf spots), ICGV 86124 and ICGV- SM 87003 in West Africa.

When drought occurs, the temperature also rises; Drought and heat tolerance appear to be correlated; Heat tolerant genotypes include 796, 55-437, TMV 2, ICGS 11, ICG 1236, ICGV 86021, ICGV 87281, ICGV 92121, etc.; In vitro pollen germination, pollen tube growth, membrane thermostability, growth rates, fruit set and partitioning are the traits used to record response of groundnut genotypes to high temperature.

Future needs: Drought tolerant cultivars that are also resistant/tolerant to aflatoxin contamination in different botanical backgrounds are needed; Adoption of marker-assisted recurrent selection (MARS) to accumulate several QTLs with small effects in a single genotype when the dense linkage map in groundnut becomes available.

Salinity: No targeted program in progress to breed salinity tolerant groundnut; A pot culture technique standardized for screening for tolerance to salinity in groundnut; In limited studies, tolerant genotypes (cultivars and breeding and germplasm lines) based on plant survival and seed yield per plant identified – NRCG 2588, NRCG 4659, NRCG 5513, NRCG 6131, NRCG 6450, NRCG 6820, NRCG 6919, NRCG 7206, TMV 2 NLM, TG 33, JNDS-2004-15, VRI 3, UF 70-103, TKG 19 A, S 206, Tirupati 4, M 522, Punjab 1, BG 3, Somnath and ICGV 86590.

Seed dormancy: When rainfed groundnut is caught in rains at the time of harvest, it results in *insitu* germination in Spanish and Valencia cultivars, thus causing significant losses in yield and quality of the produce; Incorporation of 2-3 weeks fresh seed dormancy in Spanish and Valencia cultivars will help to avoid these losses, which could reach up to 40%.

Different seed parts – seed coat, cotyledons and embryo – play a role in imparting dormancy; Fresh seed dormancy is more under control of testa than cotyledons; Complexity arises in studying inheritance of seed dormancy when both maternal (testa) and zygotic (cotyledons) tissues are involved in its control; From monogenic control with seed dormancy dominant over non-dormancy to quantitative inheritance with additive, dominance and digenic epistatic effects are reported in literature; Several Spanish breeding lines/cultivars with fresh seed dormancy are available now; Most of these originate from Virginia x Spanish/Valencia crosses; Instead of screening for seed dormancy in early generations, the advanced generation Spanish breeding lines are screened for fresh seed dormancy in laboratory and under field conditions.

Nitrogen fixation: Nodulation and  $N_2$  fixation are dependent on the interaction between the host cultivar and the nodulating *Bradyrhizobium* strain; Virginia types are superior to Spanish and Valencia types in  $N_2$  fixation; Considerable variation exists for ability to nodulate and  $N_2$  fixation, which are under genetic control of the host and are heritable; No directed breeding program currently in progress to improve  $N_2$  fixation in groundnut; Selection for high yield also indirectly improves  $N_2$  fixation.

Adaptation traits: Growth and development in groundnut is largely driven by temperature; Optimum temperature  $(T_o)$  for growth and development in groundnut ranges between 27°C and 33°C; Base temperature  $(T_b)$  in groundnut ranges between 9°C and 13°C below which the growth ceases;  $T_b$  varies for different phenological stages and among genotypes; Growth is slowed down at lower temperatures and it takes longer for crop to mature; Reverse is observed at relatively higher temperatures, but at temperatures exceeding 45°C, the growth stops as protein gets denatured; At temperatures above the optimum, significant reduction in dry matter production and partitioning of dry matter to pods are observed but flower production is not affected; Photoperiod does not affect flowering in groundnut but it affects partitioning; However, these effects are genotypic specific; Irradiance, together with temperature, also plays a role in determining the crop duration.

Short-duration: Early maturity is a relative term – in India early maturing varieties are less than 100-day duration whereas in China and USA a variety of 120-day duration will qualify as early maturing variety; Selection based on days to first flower alone is ineffective in identifying early maturing lines as other processes also involved in reaching to maturity; Instead of calendar days, use of cumulative thermal time (CTT) measured in day degrees (°Cd) is recommended for selecting for early maturity at a given location; CTT is measured in day-degrees (°Cd) above the base temperature and is calculated on successive days by subtracting the base temperature from the mean daily temperature and adding each value to the sub-total accumulated since the seed was sown (CTT (°Cd) =  $\mathcal{E}$  ((Tmax+Tmin)/2 – Tb) ); In photoperiod-insensitive genotypes the CTT for maturity does not differ across environments barring the influence of environmental factors other than photoperiod; For photoperiod-sensitive genotypes, the CTT will vary with photoperiod over the photoperiod-sensitive range.

Based on the past records of temperatures (15-20 years), CTT for the desired crop duration at a given location should be worked out; When the desired CTT is achieved the segregating breeding populations are uprooted and plants selected visually in the field based on number of matured pods (internal pericarp color) and high pod yield for further laboratory evaluations for seed appearance, uniformity, and maturity, and plants, finally selected, are advanced to the next generation; Advanced breeding lines are evaluated for 2–3 years in on- station replicated trials with staggered harvesting starting from a predetermined date based on the desired CTT; In addition to pod yield and pod and seed maturity, extent of further gains in pod yield, seed weight and shelling turnover in the following staggered harvesting are taken into account while selecting advanced breeding lines; Those with high pod yield and pod and seed maturity and minimum gains in pod yield, seed weight and shelling turnover over the preceding harvest are selected for further multilocation evaluation and selection.

Incorporating large seed size in short duration cultivars is unlikely to succeed as large seeds take more time to emerge on sowing, and to develop and mature; Similarly, combining higher levels of resistance to foliar diseases and short-duration will be difficult to achieve; On the other hand, a moderate level of resistance will have only limited influence on crop duration and would also stabilize productivity in a cropping system.

In breeding for early maturity, it is helpful to partition crop duration into different segments/stages and examine the possibility of shortening their duration individually and collectively with an overall aim to reduce crop duration; These segments/stages include days to germination and emergence, days to first flower after emergence, days from opening of first flower to opening of a given number of flowers per plant and days from opening a flower to maturation of seeds that develops from that flower; Based on the botanical characteristics and physiological behaviour of the crop, the following characteristics could be visualized for attaining short duration of the crop: short plant stature (plant height in case of subspecies fastigiata and plant spread in case of subspecies hypogaea) with smaller internodal length, faster germination and emergence, fewer days to first flowering, and accumulation of a maximum number of early flowers, more flowers per node, absence of late flowers, fewer days after fertilization for a peg to enter soil, faster pod and seed growth, high seed partitioning, and high shelling turnover; To capitalize on the full potential of the genotypes with aforementioned traits, it would be essential to modify crop husbandry to accommodate larger numbers of plants per unit area to provide quick ground cover and to provide plant with required nutrients and other inputs; The following considerations in breeding strategy will help to achieve the objective of early maturity along with high yield: (i) Selection for low Tb and CTT for various phenological stages (ii) Selection for tolerance to high temperature (iii) Selection for photoperiod-insensitive genotypes (iv) Selection for high crop growth rate and partitioning (v) Selection for high water-use efficiency, and (vi) Evaluation in
target environments/cropping systems.

Sources of early maturity: Chico, Gangapuri, JL 24, ICG 3540, ICG 3631, ICG 4558, ICG 4729, ICG 4890, ICG 9427, ICG 9930, ICG 11605, 91776, 91176, Dh 40, ALG(E) 57, TG 1E, TG 2E, TG 3E, etc.

Short-duration cultivars: Pronto and Spanco in the USA, Dh 40, TNAU 97, ALG(E) 57, GG 3, GG 5, GG 12, TG 26, R 9251, M 522, RS 138, VRI 3 and Co 4 in India and 55-437, TS 32-1, 73-30, KH 149A, KH 241D, Te 3 and Fleur 11 in Africa, among others, in different habit groups.

Methods of estimation of maturity: Determination of optimum maturity critical as it affects marketable yield, quality, and flavor of the produce; Indeterminate fruiting characteristic of groundnut results in seed of varying maturities on the same plant as harvest time approaches; Maturity estimation methods include indirect methods (days after planting, heat units), relative color evaluations (internal pericarp color, oil color, methanol extract, pod maturity profile), weight and weight relationships (kernel density, seed to hull ratio maturity index) and quantification of a specific component (arginine maturity index and protein markers); Some methods are more complex than others to evaluate and each method has deficiencies; However, the most commonly used method by smallholder farmers in developing countries is internal pericarp color because of its simplicity; When 75 to 80% pods in cultivars belonging to subspecies *fastigiata* and 70 to 75% pods in cultivars belonging to subspecies *hypogaea* show internal pericarp darkening, the crop is ready for harvest.

Future needs: Diversification of sources of earliness and studies on their genetic constitution and allelic relationships are needed to identify different genes for earliness that could be accumulated in a desired genotype.

### Quality traits:

Oil quality: Oxidative stability and shelf life of groundnut and its products can be enhanced by improving oleic to linoleic fatty acid ratio, which normally ranges between 0.8 and 2.5 in old commercial cultivars; these two fatty acids constitute about 80% (55% oleic acid (18:1) and 25% linoleic acid (18:2)) of the oil content of groundnut; Of these two fatty acids, linoleic fatty acid is less saturated and less stable than oleic acid; In peanut breeding program at the University of Florida in 1987, two breeding lines originating from F 435, a high oleic acid spontaneous mutant, with 80% oleic and 2% linoleic fatty acid composition were identified; With simple inheritance (single recessive or two recessive genes and some possible modifiers depending upon the parents involved in the crosses), it is easy to transfer high-oleate trait to other genotypes through backcross breeding program.

Varieties developed with high O/L ratio in the USA through conventional breeding are SunOleic 95R, SunOleic 97R, Tamrun OL01, Georgia 04S, Andru II, Florida-07 and Hull, through chemical mutagenesis are Mycogen-Flavorunner and M 2-225 and through Gamma radiation are Georgia-02 C and Georgia Hi-high; Varieties with high levels of oleic fatty acid, when consumed, have beneficial effect on human and animal health.

Improved flavour: Since 1980, the flavor of roasted groundnut has become an important consideration in breeding programs engaged in developing Virginia varieties for direct consumption as it influences consumers' acceptance; Several roasted groundnut quality sensory attributes are heritable; Choice of parents critical in ensuring good flavor of roasted groundnut in breeding lines; Jenkins Jumbo, one of the ancestors of USA-bred Virginia varieties, found responsible for their poor roasted flavour; Parents selected for crossing programs should have at least acceptable roasted flavour to ensure consumers' acceptability for new varieties; During the selection process all plants with off-type flavour should be rejected.

Biofortification: Large genetic variation exists for Zn (44–95 mg/kg) and Fe (33–68 mg/kg) contents in seeds among groundnut genotypes; Both Zn and Fe could be improved simultaneously as they are positively correlated; No trade-off between Zn and Fe contents and pod yield and oil content; High broad sense heritability: Zn – 92% and Fe – 81%; High yielding and stable genotypes across environments with high Zn and Fe contents – ICGV 06099 and ICGV 06040.

Fodder quality: As groundnut fodder (haulms) is highly valued, dual purpose groundnut cultivars are preferred in developing countries; In spite of preference for dual-purpose cultivars, farmers not willing to sacrifice pod yield for fodder yield; Studies have shown large variation in haulm yield, haulm nitrogen content, *in vitro* organic matter digestibility, metabolizing energy content and digestible haulm yield, high heritability for these traits and absence of any negative relationship between fodder quality traits and pod or haulm yield, which provide ample opportunities to improve fodder quality traits; In spite of the need to improve fodder quality and yield, the primary target trait in groundnut improvement remains pod yield; Due consideration to haulm yield and its quality parameters in the selection scheme of cultivars as haulm is an economic component of produce of groundnut crop.

## 12. Non-genetic management of major production constraints

a. <u>Biotic constraints</u>: Available non-genetic options to manage diseases and insect pests include – cultural, chemical and biological/botanical control measures; No single option including resistant cultivars for a given insect pest or disease may be adequate for maximizing the economic returns; An integrated insect pest and disease management (IPM and IDM) encompassing various approaches is required to effectively and safely contain the damage; Chemical sprays should be need-based and appropriate safety precautions taken while applying them; For high volume sprayers, 450-500 L and for low volume sprayers 225-250 L water is required to cover 1 ha; While using chemicals, protective clothing should be borne and proper care should be taken to dispose of empty bottles/cartons of chemicals in a safe manner.

Foliar fungal diseases: ELS, LLS and rust are the major foliar fungal diseases distributed worldwide.

ELS: ELS lesions, often surrounded by a yellow hallow, are sub-circular, dark brown on the upper leaflet surface where most sporulation occurs and lighter shade of brown on the lower leaflet surface; Yield losses in southern Africa and the USA can exceed 50%.

LLS: LLS lesions are nearly circular and darker than those of ELS; On the lower leaflet surface, where most of the sporulation occurs, the lesions are black and slightly rough in appearance; It can cause up to 55% yield loss.



Figure 8. Late leaf spot.



Figure 9. Defoliation caused by leaf spots.



Figure 10. Early leaf spot.

When the attack is severe, both the diseases cause severe premature defoliation; Spores of ELS and LLS causing fungi can survive for years in soil. These diseases can also spread through airborne inoculum coming from other infected fields.

Rust: Orange colored rust pustules appear on the lower surface of leaflets; On rupturing, they release masses of reddish-brown spores; In contrast to the rapid defoliation associated with leaf spots, leaves infected with rust become necrotic and dry up, but tend to remain attached to the plant; It can cause yield losses in excess of 50% and when it occurs in combination with LLS, the losses can reach 60-70%; As rust spores do not survive for long under ambient conditions (not more than 30 days under ambient conditions in South India), its inoculum is mostly airborne and comes from other infected fields.



Figure 11. Rust 'hot spot' in the field.



Figure 12. Groundnut rust.

Management: Generally, ELS, LLS and rust start appearing on the lower leaves from 30-35 days after sowing in humid regions and spread rapidly in favorable weather conditions; If not managed properly at the initial stage, the crop losses could be severe; Effective chemical control measures are available for these diseases (Chlorothalonil 75 WP 1kg/ha for combined occurrence of leaf spots and rust, Carbendazim 50 WP 500 g/ha or Mancozeb 50WP 1kg/ha for both leaf spots and Calixin 250 ml/ha for rust alone); Chemical control measures should start soon after the appearance of disease symptoms; Spray schedule varies depending upon the agroclimatic conditions and severity of the diseases; Number of sprays can be reduced by following integrated disease management practices; In the USA and other developed countries, farmers follow weatherbased disease management advisory to optimize chemical use and increase its effectiveness; In many countries, rust resistant and leaf spots tolerant cultivars are now released; In disease endemic areas, these cultivars together with limited chemical control provide best economic returns; In the case of severe premature defoliation, it is advisable to harvest the crop early; Cultural management practices include removal of debris from the field and volunteer plants in the

vicinity, intercropping with cereals (pearl millet, sorghum, maize, etc.), deep plowing and crop rotation, particularly for leaf spots to reduce inoculum in the soil.

<u>Soil-borne diseases</u>: Difficult to control once they appear; But maintaining soil health is very important for good agriculture.

Collar rot: Caused by *Aspergillus niger*, symptoms include preemergence rotting of seeds, rotting of cotyledons, rotting in the collar region, rotting regions covered with black conidia of the fungus, post emergence seedling blight with dark brown and shredded collar region and occasional wilting of plants at later stage; Sometimes mature plants also attacked – lesions develop on the stem below soil and they spread upwards, dead and



Figure 13. Crown or collar rot.

dried branches easily detach from the disintegrating collar region.

White mold/stem rot: Caused by *Sclerotium rolfsii* is widely distributed in most of the groundnutgrowing countries; First symptoms - yellowing and wilting of a lateral branch or, if the main

stem is attacked, the whole plant; Leaves on affected branches become chlorotic and then turn brown as they rapidly dry out; Sheaths of white mycelium of the fungus seen around affected plants at or near the soil surface imparting a 'white-washed' appearance to the base of the affected plants.

Management: Management practices include growing tolerant cultivars, seed treatment with thiram or captan or mancozeb @ 3 g/kg of seed or carbendazim @ 2 g/kg of seed or tebuconazole (Raxil 2% DS) @ 1.25 g/kg seed or *Trichoderma viride* or *Pseudomonas flurescens* @ 10 g/kg seed, soil application of



Figure 14. Advanced symptoms of white mold/stem rot.

*Trichoderma viride* or *Trichoderma harzianum* @ 2.5 kg/ha in conjunction with neem or castor cake @ 300–500 kg/ha, crop sanitation, crop rotation with wheat or gram and avoiding injury to seedlings during intercultivation.

Aflatoxin contamination: Aflatoxins are secondary metabolites produced in groundnut seeds by *Aspergillus flavus* and *Arachis parasiticus* and are carcinogenic to human beings, poultry and livestock; These fungi (soil and air borne) get access to seeds through microscopic cracks in pod wall and seed coat, mechanical injury to pods during inter cultural operations and harvesting and insect and nematode damage to pods; In international trade, very strict upper limits set for aflatoxins – 2 µg/kg of seed for aflatoxin B<sub>1</sub> by European Union; In Asia, *Aspergillus flavus* infection is more prevalent; Fungus infection can occur in the field when the pods and seeds are developing (preharvest infection), during drying and curing after harvest and in storage (postharvest infection); Moisture stress at pod and seed development stage predisposes them for *Aspergillus flavus* infection in the field; Rainfed groundnut is more vulnerable to fungal infection in the field; Postharvest infection is serious under wet and humid condition



Figure 15. Groundnut kernels naturally inoculated with Aspergillus flavus.



Figure 16. Groundnut kernels artificially inoculated with Aspergillus flavus.

Management: In addition to the above mentioned management practices for soil-borne diseases, the following additional measures should be adopted to contain the aflatoxin contamination in groundnut.

- §□ Application of FYM/compost @ 5-10 t/ha
- §□ Application of gypsum @ 400-500 kg/ha at flowering
- $\square$  Provide light but frequent irrigation, if available, during pod and seed development stages
- $\$  Avoid mechanical damage to the pods during weeding, harvesting and curing, threshing and storage
- $\square$  Harvest the crop as soon as it matures (in case of severe drought, the crop should be harvested early)
- $\$  Dry pods to lower moisture content below 8%
- $\square$  Do not mix gleanings (pods left into soil at the time of harvesting) with main produce
- §□ Remove damaged and under developed pods from the produce
- §□ Store groundnut in-shell and at low temperature, low humidity and in moisture-free conditions and protect it from the damage by storage insect pests
- §□ Remove immature pods from the haulm before feeding them to animals

<u>Virus diseases</u>: Field identification of virus diseases based on symptoms alone often difficult and misleading; Important to confirm the identity of a disease after serological and other laboratory tests.

PBND: <u>Primary symptoms</u> – appearance of faint chlorotic spots or mottling on young leaflets that may develop into chlorotic and necrotic rings and streaks, occasionally, the leaflets may show a general chlorosis with green islands, necrosis of the terminal soon follows; In very young plants, total necrosis of the plants can happen; Necrosis on older plants usually spreads only to the petiole or to the portion of the stem immediately below the terminal bud; <u>Secondary symptoms</u> – stunting and proliferation of axillary shoots, leaflets on these axillary shoots show a wide range of symptoms including reduction in size, distortion of the lamina, mosaic mottling, and general chlorosis.



Figure 17. Terminal necrosis caused by peanut bud necrosis virus.



Figure 18. Groundnut plants with advanced symptoms of peanut bud necrosis virus.

TSWV: Wide array of symptoms include concentric ring spots on leaflets, various patterns of chlorosis on leaflets, stunting of all above ground parts of the plant, small or misshapen geocarpophores, pods and kernels, and reddish discoloration and cracking of seed coats; Roots of the affected plants typically show varying degree of necrosis that can result in the death of the plants; General yellowing and wilting of the plants without typical above ground symptoms.



Figure 19. TSWV symptoms - concentric ring spots on a groundnut leaf.

PStV: Characteristics symptoms – dark green stripes and discontinuous banding along the lateral veins of young leaves and an okra leaf or blotched pattern of dark green on older leaves with infected plants showing stunting; Eight strains of PStV reported, which cause different symptoms; Yield losses vary with strain type and can reach as high as 55%.



Figure 20. Stripe and blotch symptoms caused by different strains of PStV.



Figure 21. Oak leaf symptoms occasionally caused by PStV.

PSND: Caused by tobaco streak ilavirus; Symptoms - first appear on young leaves as necrotic lesions

and veinal necrosis, later necrosis spreads to petiole and stem; Necrotic lesions on the stem later spread upward killing the buds; Majority of the plants infected within a month after sowing die due to necrosis, which also spreads downwards in case of early infection; In some cultivars, surviving plants produce axillary shoots with small leaves with general chlorosis.



Figure 22. PSND necrosis of terminal leaflets and stems.



Figure 23. PSND infected groundnut pods.

Management: Management options – growing field tolerant varieties (for PBND, TSWV and PSND; no tolerance available for PStV), field sanitation and cultural management (removal of volunteer plants in the off-season, timely sowing (sowing dates should be adjusted to avoid migration peak of the vector at the location), careful removal and destruction of infected seedlings, optimum plant population, effective control of vectors and intercropping and border cropping with fast growing tall cereal crops, removal of alternate weed hosts of the virus (particularly *Parthenium* in and around groundnut fields in case of PSND) and the vector during off-season); Seed treatment with imidacloprid offers protection against sucking pests up to 30 days and helps to reduce disease incidence in early stages of the crop; Intercropping and border cropping act as barrier to the vector.

PCV: Wide spread in West Africa, also found in Indian subcontinent (known as ICPV) in isolated patches particularly in sandy and sandy loam soils; Disease is soil borne and transmitted by soil inhabiting fungus *Polymixa graminis*; Virus is seed borne (6-50%); Diseased plants severely stunted, dark green and bushy with young leaves showing mosaic mottling with chlorotic rings; Infected plants produce flowers but a few poorly developed pods.



Figure 24. Leaf symptoms caused by IPCV.



Figure 25. An IPCV infected field.

Management: Available cultural management options to minimize the disease in the long run – use of disease free seed, growing of trap crop (pearl millet and other cereals), early sowing before the onset of monsoon, soil solarisation and continuous rotation of dicotyledonous crops.

GRD: Two symptom variants in GRD – chlorotic rosette and green rosette; Both forms of the disease cause severe stunting with shortened internodes and reduced leaf size giving the plants a bushy appearance; In chlorotic rosette, leaves are usually bright yellow with a few green islands and curved lamina; In green rosette, leaves appear dark green with light green to dark mosaic; GRD is present in small proportion every growing season but it is more severe in late planted crop causing 5-30% yield losses; Epidemics cause significant yield losses reaching up to 100% in larger areas, sometimes even affecting the seed availability for the next season crop.



Figure 26. Chorotic rosette symptoms.



Figure 27. Green rosette symptoms.

Management: Management options – Use of resistant cultivars (the best option, several are released in Africa), removal of off-season volunteer groundnut plants that serve as inoculum source for the main season crop, timely/early planting and ensuring optimum plant population, seed treatment with imidacloprid and control of aphid vector with systemic insecticide; Long acquisition access

feeding period required by the vector provides an opportunity to control aphids before they can spread the disease.

For further refining management strategies, the research on the following aspects should be intensified: identification of alternate host(s) of the components of the virus complex for off-season survival, genetic diversification of resistance and combining resistance to GRAV, GRV and vector in a single cultivar.

<u>Bacterial wilt disease (BW)</u>: A soil borne disease, caused by *Ralstonia solanacearum* in groundnut, mostly confined to East and Southeast Asia but also reported from Uganda and South Africa in Africa; Disease spread by infested soil and water and infected seed; Bacterium has five races, only race 1 (biovar 3 and 4) infect groundnut; First sign of BW – slight drooping or curling of one or more leaves; In advanced stages, the plants may bend at the tip appear dry and eventually turn brown, wither and die; Roots and pods discolored and rotten; Diagnostic characteristics – dark brown discoloration in xylem and pith and streaming of bacterial ooze when roots placed in water; In heavily infested fields, disease incidence could reach over 50%, in some cases, a total crop failure can also happen.



Figure 28. A wilted groundnut plant due to bacterial wilt disease.

Figure 29. A bacterial wilt infected groundnut field.

Management: Management options - growing resistant varieties (many are available), crop rotation with non-host crops (rice in lowlands and sweet potato, jute, maize, sugarcane, soybean, blackgram, and mungbean, etc.) and flooding the field; Good field drainage and field sanitation also help in reducing the disease.

<u>Nematode diseases</u>: <u>Symptoms of root knot disease</u> caused by *Meloidogyne arenaria* and *Meloidogyne hapla* – stunted plant growth and chlorotic leaves, poor root system with dense bushy appearance and knots and warts (galls) on pods; <u>Symptoms of root lesion disease</u> caused by *Pratylenchus brachyurus* – stunted plant growth with poor and discolored root growth, tan to brown colored pin-point areas on the pod surface with older lesions giving a blotchy appearance and indistinct margins; <u>Symptoms of 'Kalahasti malady</u>' caused by *Tylenchorhynchus brevilineatus* in India – stunted plant growth with foliage greener than normal, small brownish yellow lesions on pegs and developing pods with lesion margins slightly elevated and complete discoloration of pods in advanced stage.

Management: Management options - growing resistant varieties (most effective and practical), crop rotation with non-host crops or trap crops.



Figure 30. Pods infected with *Tylenchorhynchus brevilineatus*.



Figure 31. Galls on roots and pods caused by *Meloidogyne arenaria*.

<u>Major field insect pests</u>: Major sucking pests – thrips, jassids and aphids; Major defoliators – leaf miner, tobacco caterpillar, gram pod borer and hairy caterpillars; Major soil insect pests – white grubs, termites, earwig and jewel beetle.

Thrips: Small insects, present any time of the year, live in flowers and folded leaves and have wide host range; Nymphs pass through four instars before becoming adults; Damage symptoms –white patches on the upper and necrotic patches on the lower surface of the leaves which distort their shape particularly in seedlings.



Figure 32. Leaflets scarified by thrips.



Figure 33. An adult thrips.

Jassids: Both nymphs and adults suck sap from lower surface of young leaves; Damage symptoms – whitening of the veins and appearance of chlorotic (yellow) patches at the tips of leaflets with typical 'V' appearance.



Figure 34. Jassid damage ("V" shape hopper burn on leaflet tips).



Figure 35. An adult jassid.

Aphids: Dark brown nymphs develop into shiny black adults which congregate on young leaves, leaf buds, flowers and aerial pegs and desap through phloem vessels; Plant becomes chlorotic and leaves curl; They secrete a sticky fluid on the plant which is turned black by fungus.

Leaf miner: Young larvae mine into the leaves as soon as they hatch, presence of small brown blotches on the leaf can be seen; Mines are about 1mm long and enlarge as the larvae grow inside them; Larvae complete different instars, emerge out and web the adjacent leaflet's together, and continuously feed on leaf tissue from inside the webbed leaves; Severely attacked fields give burnt appearance from a distance.



Figure 36. An adult leaf miner.



Figure 37. Brown blotches (mines) on leaflets.

Tobacco caterpillar: Adults lay eggs on the leaves in clusters; First instars feed by scrapping under the surface of the leaves leaving the vein and upper epidermis giving a fabric surface; Young larvae are light green but later instars are dark green to brown on their backs, lighter underneath and have prominent black spots on the thorax; They defoliate totally leaving only the stems; 2<sup>nd</sup> and 3<sup>rd</sup> instar larvae feed by making small holes on the leaf; Subsequent instars feed voraciously on entire lamina, petioles and on the tender twigs, sometimes they feed on the flower and bore the tender groundnut pods; Pupation takes place in soil; Older larvae are night feeders.



Figure 38. Tobacco caterpillar larvae.



Figure 39. An adult moth of the tobacco caterpillar.

Gram pod borer: A polyphagous pest, adults lay eggs singly on tender plant parts; Larvae – dark greenish yellow/brown with variation in color; Unlike tobacco caterpillar, they do not have black spots on thorax; Damage to foliage similar to tobacco and hairy caterpillar, larvae prefer to feed on the flowers and buds; They do not hide in the soil during the day.



Figure 40. Gram pod borer larvae.



Figure 41. Gram pod borer moth.

Hairy caterpillars (*Amsacta albistriga, Arachis moori* and *Spilosoma obliqua*): Hairy caterpillars devastating but highly sporadic; Larvae – initially light brown but turn reddish when they grow, their hairs make them conspicuous and they often migrate from field to field in search of food; Young larvae feed gregariously on the under surface of the leaves by scrapping them; Grown up larvae are voracious feeders, feed on leaves, flowers and growing points and defoliate the crop

presenting a cattle grazed field; Young larvae of *Spilosoma obliqua* scrap leaves, some leaves become papery thin and dry, grown up larvae devour the foliage completely.



Figure 42. Red hairy caterpillar larvae.

Figure 43. Red hariy catepillar moth.

White grubs (*Lachnosterna consanguinea, Lachnosterna serrate*): Pupation takes place in the soil and the larvae remain there until the next year, adult beetles emerge with first monsoon showers; Damage to roots starts in the early first instar and maximum damage occurs during third instar larvae; Plants show varying degrees of wilting, and ultimately die; Damaged plants can be easily pulled out; Grubs also damage the pod by scratching the nut shell and roots show a sharp cut; Patches of dead plants seen throughout the field, which later coalesce and produce intensive areas of damage.



Figure 44. White grub adults.



Figure 45. White grub larvae.

Termites (*Microtermes* spp., *Odontotermes* spp.): Termites gnaw and hollow out taproot causing wilting and premature death of plants especially in sandy and red soils; They also feed on pod shell by removing the corky material between the strands of vascular tissues, which is called 'scarification'.



Figure 46. Hollowing out damage of tap root by termites.



Figure 47. Termites.



Figure 48. Termite scarification on pods.

Earwig (*Demaptera stali*): Both nymphs and adults bore into the tender and matured pods and feed on kernels; Bored holes are mostly plugged inside with sand particles, excreta and decayed pulp; Infested pods either empty or with partly fed kernels and rarely with sand and faecal materials.

Management: Integrated pest management should be followed; Chemical control measures should be need based, guided by economic threshold levels and used as a last resort.

<u>For control of sucking insect pests</u> - grow resistant varieties, treat seeds with imidacloprid (Gaucho 70 WS) @ 2 ml/kg seed, spray dimethoate @ 200-250 mL a.i./ha when the crop is young and all terminal buds are infested with aphids or more than five thrips per terminal (folded) leaf are observed up to 30 days after emergence (DAE) or more than 10% of all leaves have characteristic 'hopper burn' up to 30 DAE.

<u>For control of defoliators</u>, the following steps should be taken.

Leaf miner – Spray dimethoate @ 200–250 mL a.i. or monocrotophos @ 150–200 mL a.i. per ha if 5 or more active larvae per plant up to 30 DAE, 10 larvae per plant up to 50 DAE or 15 larvae up to 75 DAE or later,

install pheromone traps @ 25 traps/ha, setup light



Figure 49. Pod damage by earwig.



Figure 50. Earwig with eggs.

traps for destroying the moths; Economic threshold studies indicated that chemical control should be adopted when the larval population reaches 61–70 per 100 leaflets.

Tobacco and hairy caterpillars and gram pod borer – For bird predators provide perches (bird perches @ 10-15/ha) in the field; Plant sunflower and castor bean trap crops on borders and in the groundnut field (1 plant per 20 m<sup>2</sup>) and destroy egg masses on trap crops and groundnut plants manually; Tobacco caterpillar and gram pod borer nocturnal in habit and hence control

measures to be taken up either in the early morning hours or in the late evening hours; Apply NPV @ 250 LE/ha, obtained from a reliable source and neem seed kernel extract @ extract obtained from 10 kg neem fruit powder ha<sup>-1</sup>; Spray endosulfan @ 350 mL a.i. or monocrotophos @ 300 mL a.i. or fenvalerate @ 100 mL a.i. or indoxacarb @ 70 mL a.i. or spinosad @ 45 mL a.i. per ha if defoliation exceeds 25% during the first 50 days or one or more larva per plant is observed or pheromone trap (10 traps/ha for tobacco caterpillar and gram pod borer) catches exceed 100 moths per night averaged over a week or 1-2 tobacco caterpillar egg masses per meter of crop row (7-12 plants).

For red hairy caterpillars, digging 15–20 cm deep trench all around the field and placing Ipomea or some other plant twigs in them can restrict the migrating caterpillars in the trench where they could be destroyed manually or by spraying fenvalerate @ 1mL/L water; Alternately, a short barricade of polythene fence (10 cm high) across the migrating route can prevent their entry in the field and they can be collected manually and destroyed.

<u>Management of soil insect pests</u> – as prophylactic measures apply carbofuran 3G granules in the furrow @ 1 kg a.i./ha and treat seeds with chlorpyriphos 20 EC @ 12.5 mL/kg seed for protection against white grubs in the initial stages; Spray feeding trees of adult white grubs with carbaryl 50 WP @ 2 g/L of water 3-4 times until mid-July ideally using community approach; Destroying the termite mounds in the vicinity of field, removal of plant residues and debris from field and timely harvest can help to minimize the termite damage.

### Major storage insect pests:

Groundnut bruchid (*Caryedon serratus*): Both pods and kernels are damaged; Eggs laid on the pods and kernels; Young larvae cut through shell and burrow into the kernel and feed; Cocoon is papery white and tough; Pupation takes place on the pods or kernels.



Figure 51. Eggs of groundnut bruchid.



Figure 52. Adult groundnut bruchid.

Red flour beetle (*Tribolium castaneum*) and rice moth (*Corcyra cephalonica*): These are considered secondary pests as they are not able to infest intact pods; However, with a lot of mechanical damage caused to pods due to poor intercultivation and harvesting and curing practices, their damage in storage is quite common; Signs of damage – webbing in the case of rice moth and powdery remnants without webbing in the case of red flour beetle; Red flour beetles are oblong in shape and brown in color; Rice moth has grayish brown forewings.

Management: Development of storage insect pests can be arrested if seed moisture content is kept low (not more than 5%); Storage sanitary measures include spray of malathion 1.25% or deltamethrin 0.04% on the walls, floor and roof of the warehouses or godowns before storage; Fumigation with celphos (aluminium phosphide) 3 g tablet per sack of groundnut (40 g) and covering the sacks with a polythene sheet for five days can effectively control storage pests.



Figure 53. An adult red flower beetle.



Figure 54. An adult rice moth.



Figure 55. Powdery remnants resulting from red kernel flour damage to kernels.



Figure 56. Webbing resulting from damage by rice moth.

### b. Abiotic constraints

Drought: All rain water conservation practices should be followed and harvested water should be used to give life-saving irrigation and irrigation at critical stages (podding and seed development) of plant life cycle.

Salinity: Saline/sodic fields should be reclaimed by leaching salts with excess water (sprinkling or flooding) and adequate drainage; Soil amendment (gypsum) should also be added to the soil; Salt tolerant varieties and crops should be grown; Some of the crops with varying degree of salt tolerant are maize, sorghum, wheat, alfalfa, crested and tall wheatgrass and sorghum-sudan hybrids.

Low pH: Maintaining soil pH essential as it affects the availability of nutrients to plants; In case the pH is <5, lime in appropriate form and quantity should be mixed thoroughly into the soil before land preparation or at the time of land preparation so as to bring it in the optimal range; Rate of application of lime depends up on the type of lime, soil type and depth of application; As a general recommendation, it would require 1.5 t/ha of lime (CaCO<sub>3</sub>) to raise soil pH from 5.0 to 6.5.

Nutrient deficiencies and their management: Most micronutrients more likely to be deficient at high pH, particularly in calcareous soils; Most soils in India are deficient in micronutrients – Zn, B and S; If soil test shows deficiency of these micronutrients, the remedial measures should be taken as follows:

Boron (B): Boron, an essential micronutrient, necessary for proper flowering and podding; Boron deficiency causes less intense flowering, prolongs flowering period and affects seed development resulting in a discolored cavity (hollow heart) on the inner face of the cotyledons; Severe Boron deficiency causes leaf to turn deep green, terminal leaves to become small and deformed, reduced internodal length and production of secondary branches making the plant appear stumpy and short.

Application of borax to the soil at the time of land preparation at a rate of 3-4 kg/ha usually overcomes groundnut seed due to Boron deficiency. the symptoms with a good residual effect that lasts for

several seasons; Deficiency can also be corrected by spraying 0.1% borax early in the season to assure uptake before flowering; Over application of Boron can cause toxicity in plants.

Zinc (Zn): Zinc deficiency, observed when the crop grown in high pH soils, is wide spread in sandy and sandy-loam soils in India; Zinc-deficiency causes stunted plant growth with reduced internodal length, new leaves develop slowly and terminal leaflets are small, thickened and dark green in color; Basal application of 10-20 kg/ha zinc sulphate once in three years to the soil where groundnut is grown continuously can rectify the deficiency.

Figure 57. Discolored cavities in



Figure 58. Zinc deficiency symptoms in groundnut.

Sulphur (S): Deficiency of sulphur, common in sandy soils, affects terminal growth, restricts root growth and new leaves become pale green or yellow; Application of gypsum provides adequate sulphur to the crop.



Figure 59. Sulphur deficiency symptoms in groundnut.



Figure 60. Iron deficiency symptoms in groundnut.

Iron (Fe): Iron deficiency serious problem in calcareous soils; Plants suffering from Iron-deficiency show interveinal chlorosis (starting in the youngest leaves) followed by chlorosis of the entire leaf (whitish-yellow) and brown spots leading to marginal chlorosis, slows down plant growth and results in poor nodulation; Can be alleviated by applying 10 kg/ha ferrous sulphate to the soil or spraying the affected crop with 0.5% ferrous sulphate + 0.2% urea, if required, the spray can be repeated at 10-14 days interval.

Manganese (Mn): Manganese deficiency occurs on soils inherently low in this nutrient, especially if they have been over limed; Manganese deficiency causes interveinal chlorosis with symptoms ranging from mild (light green leaves with the regions immediately adjacent to the veins and the veins themselves remaining green) to severe (entire interveinal area chlorotic); Foliar Manganese application can correct Manganese deficiency more rapidly than soil Manganese application.



Figure 61. Manganese deficiency symptoms in groundnut.

## **13. Cultural practices**

<u>Selection of field and land preparation</u>: Groundnut after groundnut in the same field not advisable due to build-up of diseases and insect pests in the soil; Groundnut should be rotated with a well-fertilized cereal crop; A proper crop rotation can result in higher yields and in substantial savings in disease control and fertilizer requirements; Ideal groundnut soil – well drained, light colored, with either sand, loamy sand or sandy loam texture and pH ranging between 6.0 and 6.3.

Field should be cleared off all stubbles and plant residues of the previous crop (un- decomposed plant residues promote growth of disease causing soil borne fungi); Land preparation – plow to a depth of 15-20 cm (very deep plowing should be avoided) and give several passes of harrow to obtain a fine tilth; Conservation tillage (no-tillage, minimum tillage, reduced tillage and strip tillage) does not consistently produce yields equal to the conventional tillage; Very deep plowing encourages pod formation in deeper layers of soil rendering harvest more difficult; Groundnut

can be sown on flat bed or ridges or raised beds separated by furrows; Sowing on raised beds with 0.4–0.8% slope allows easy drainage of excess water, avoids compaction of seed beds and facilitates field operations as all movements are restricted to furrows; Width of the raised bed varies depending up on soil type, irrigation system in use (in case of irrigated crop) and available equipment for land preparation and bed formation.

<u>Manure and macro-nutrient requirement</u>: Groundnut responds to residual soil fertility better than the direct application of fertilizers; Crop(s) preceding groundnut should be well fertilized to build up soil fertility particularly for P and K.

Basics of the fertilization strategy are the following.

Soil test: Soils from fields to be sown with groundnut must be analysed for determining the status of macronutrients and micronutrients; Soil samples should be collected from several places in the field at plow depth; Samples should be bulked and mixed thoroughly to draw a representative sample of the field for analysis.

Build soil phosphorus (P) and potassium (K) to medium or high level: Groundnut responds to residual soil fertility better than the direct application of fertilizers; Building soil P and K levels to at least the medium range should assure adequate P and K nutrition; If P and K fertilizers are needed, they should be turned deep before sowing to keep them out of the pegging zone where they can interfere with calcium uptake; Better to fertilize the preceding crop well to build up residual fertility for the following groundnut crop.

Provide calcium (Ca) to pegging zone: Calcium, essential for pod and seed development, is most commonly deficient for groundnut in non-calcareous soils; Having adequate quantities of Ca present in the top 8 cm -10 cm of soil, available for direct absorption by developing pegs and pods, is an absolute must for producing high quality groundnut yields; Adequate Ca supply to the pods also helps to reduce infection by Aspergillus flavus and other pod rot causing fungi.

Nutrient requirement for high yields: The amounts of nutrients removed from the field by groundnut pods and vines are presented in Table 10.

Quantity (kg/ha) <sup>1</sup>										
Pod yield (t/ha)	Ν	Р	K	Ca	Mg	S	Fe	Mn	Zn	В
1	58	5	18	11	9	4	2	0.09	0.08	0.05
2	117	10	36	23	18	9	4	0.19	0.16	0.11
3	174	15	54	34	27	13	6	0.29	0.24	0.16
4	232	20	73	45	36	18	8	0.38	0.32	0.22
5	290	25	91	56	45	22	10	0.48	0.41	0.27
6	348	30	109	68	54	26	12	0.58	0.49	0.33
7	406	35	126	77	63	30	14	0.68	0.56	0.38
8	464	40	144	88	72	34	16	0.78	0.64	0.44
9	522	45	162	99	81	38	18	0.88	0.72	0.49
10	580	50	180	110	90	42	20	0.98	0.80	0.54
<sup>1</sup> Calculation based on Sahrawat, Srinivas Rao, and Nambiar. 1988. Plant and Soil 109:291-293.										

Table 10. Estimated nutrients required to produce selected pod yields of groundnut.

Nitrogen (N): Under most conditions enough N fixed through symbiotic relations with the native Bradyrhizobium spp. to avoid deficiency throughout the plant's life cycle; However, under certain conditions, N deficiency can occur – poor and eroded soils, lack of or inefficient Bradyrhizobium in the soil, drought and high temperatures; In many countries, N is applied to the crop and the rate ranges between 10–30 kg/ha N; In areas of high intensity rains, split application of N beneficial in light soils; In some studies, foliar application of N at the time of podding also resulted in increased yield; However, N application not recommended in the USA.

Phosphorus (P): On a global scale, P may be the most deficient element for groundnut; In general, P deficiency easily corrected by application of P fertilizers since the crop is grown in sandy soils with low amounts of clays, in such soils P fixation is not a problem; But in calcareous sandy soils, P fixation can lead to P deficiency in spite of P fertilization; Soil P levels required for groundnut are often lower than those required for other crops.

Potassium (K): K needed by groundnut from early stage of its growth to maturity; Considerable amount of K is taken by the crop but most of the Indian soils are rich in K; Groundnut does not respond to K fertilization unless soil available K is very low; Mutual antagonistic effect on the uptake of K, Ca, and Mg; Ratio of K:Ca:Mg more important than the total amount of any one of them; A high concentration of K in the pod zone is harmful because it affects pod quality, especially at low levels of Ca; Suggested ratios in the literature are 4:4:2 and 4:2:0; Latter was reported from studies on sandy loam soils under rainfed and irrigated conditions in Tirupati, Andhra Pradesh, India.

Calcium (Ca): 30-day period following pegging most critical for Ca supply; If soil pH adjustment is recommended by the soil test, lime may be used to provide both pH adjustment and Ca to the pegging zone; However, lime must be applied and incorporated to a shallow depth immediately after deep plowing to allow enough time for Ca in lime to dissolve and get into soil solution; Where lime is not needed, calcium sulphate (gypsum or land plaster) should be applied when needed; Need for additional Ca best determined by taking a 'pegging zone soil test'; Regardless of soil test Ca level, gypsum should be applied to all large-seeded groundnut and groundnut seed crop; Peak flowering stage of the crop best time to apply gypsum to the soil; Gypsum should not fall on the foliage as it may cause scorching of leaves; Gypsum application should be followed by light inter cultivation or last hand weeding to ensure its incorporation into the soil; Groundnut produced under Ca- deficient conditions exhibits poorer germination than those produced with an adequate Ca supply; Lack of sufficient soil moisture in the top 8-10 cm zone during the pod and seed developmental period can result in Ca deficiency as the Ca is not replenished in the soil solution; It is important to ensure sufficient moisture supply in the podding zone for the development of well-filled pods.

Concentrations of other cations in the soil, particularly K and Mg, can affect Ca uptake and thereby affect groundnut yield and quality; Optimum Ca/K ratio in the groundnut pegging zone topsoil is about 10; Optimum Ca/Mg ratio for obtaining maximum percentage of sound mature kernels is 24 to 28.

Application of fertilizers and their dose should be based on the nutrient status of the soil as determined by the soil test and the targeted yield; Achieving very high yields under rainfed conditions may not be feasible due to soil moisture limitations;

General recommendations for groundnut:

FYM or Compost: 10-12 t/ha, 25-30 days before sowing

Introducing green manure in crop rotation also helps in increasing the organic matter content of the soil and improving its structure. It also improves nodulation and enhances the availability of P to the crop. Farmers in India also apply tank silt 15-25 t/ha once in three years mainly to

increase water-holding capacity of the soil.

Macronutrients: N, P and K: 8-20 N, 16-80 P205, 0-75 K20 kg/ha as basal application

Ca: 200–400 kg/ha of gypsum at the peak flowering stage as side placement (Calcium is essential for good seed development)

<u>Seed and seed treatments</u>: Certified seed purchased from a reliable source or own saved seed which is pure (true to type), graded (medium-size), undamaged, fully developed and healthy (free from discoloration and fungal infection) with germination above 90% should be used; Germination test on seeds should be carried out one week before sowing and the seed rate should be adjusted accordingly; Gap filling after 8-10 days of sowing does not help much.

*Rhizobium* inoculation: *Rhizobium* inoculation beneficial in newly cleared fields, rice fallows, fields with eroded soils and low fertility; Seeds should be treated just before sowing with *Rhizobium* culture, obtained from a reliable source, following instructions of the manufacturer.

For soil borne diseases: Seeds should be treated with captan (1.5 g) + thiram (1.5 g), carbendazim (2.0 g) or mancozeb (3.0 g) per kg of seed or other locally recommended fungicide(s); Seed treatment with *Trichoderma viride* or *Trichoderma harzianum* @ 4-5 g/kg of seed also helps in managing seed and soilborne diseases.

For insect pests: Imidacloprid 2 ml/kg of seed for sucking pests and chlorpyriphos 20 EC 12.5 ml kg/ seed for white grubs in endemic areas.

Seed treatment with fungicide/insecticide gives protection up to 30 days after sowing; It should be carried out one or two days prior to sowing; Seeds should be treated first with liquid chemicals and after drying with powder/dust chemicals; If the *Rhizobium* strain is not compatible with fungicide/insecticide (see manufacturer's instructions), the culture can be applied in sowing rows following slurry method.

For breaking seed dormancy: Virginia varieties have postharvest seed dormancy, which may last up to 5-6 months in some cases; If such varieties are to be sown immediately after harvest, the postharvest seed dormancy must be broken; Seeds should be thinly spread over a tarpaulin or plastic sheet and sprayed thoroughly 2-3 times with etherel (5 ml in 1 L water) and air dried just before sowing.

Pre-germination and seed priming: In North Vietnam, farmers sow pre-germinated (with protruding radicle) seeds for the spring season crop; In spite of prevailing low temperatures and cold winds at the time of sowing, this practice results in quicker seedling emergence and a near perfect plant stand; Maturity is also hastened due to early seedling emergence; However, it is essential to have a good tilth and enough soil moisture or irrigation soon after sowing to support seedling growth; Seeds are pre-germinated by soaking them for 4–6 h in lukewarm water and then, storing them in cloth bags overnight at a warm place in the house (near the oven in the kitchen); Next morning, seeds have emerging radicle and are ready for sowing, non-germinating seeds are discarded; Sprouted seeds are hand sown and covered with soil immediately after sowing; Recently, seed priming is receiving attention in India and Vietnam; In seed priming, seeds are soaked for 8 h in water and then dried to their original water content; A primed seed will germinate only if it takes up additional moisture from the soil after sowing; Apart from swelling slightly and weighing more, primed seed can be treated in the same way as non-primed seed.

<u>Plant density, sowing depth and sowing methods</u>: Optimum plant population varies from country to country and from region to region within a country; Optimum row spacing depends on the variety, nature of crop cultivation and machinery available for sowing; However, the aim should be to obtain full ground cover as quickly as possible; A wider row spacing leaving uncovered ground

proliferates thrips/ aphids, which can carry virus inoculum (peanut bud necrosis, peanut stem necrosis, peanut mottle and peanut stripe diseases); Row spacing varies from as low as 20 cm to as high as 100 cm, and within row spacing from 7.5 cm to 15 cm in different countries; In most countries in Asia, row spacing varies between 30 cm and 45 cm and within the row spacing between 10 cm and 15 cm depending up on the growth habit of the variety; In many countries, 2-3 seeds are dropped at each hill in manual sowing; If optimum plant stand is assured with high germination of seed lot, this practice may not be necessary as it increases the quantity of seed required for sowing; A closer spacing for Spanish/Valencia (bunch) cultivars and a wider spacing for Virginia (semi-spreading or spreading) cultivars are recommended; In India, 'paired row sowing' in Gujarat and 'cris-cross sowing' in rice fallows have sown yield advantage over the conventional pattern of sowing.

Sowing depth: Optimum sowing depth – 5 cm; Deeper sowing results in delayed emergence, elongated hypocotyl, poor root and shoot development, poor nodulation and decreased nitrogen fixation and consequently lower yields; In the case of shallow sowing, the field should be frequently irrigated to avoid drying of germinating seeds to ensure optimum germination and plant stand.

Optimum plant population: Optimum plant population in India – 330,000 plants/ha for Spanish/Valencia cultivars and 148,000 for Virginia cultivars.

Seed rate: Seed rate depends up on seed weight, germination% and row to row and seed to seed spacing; It may range from 100 to 160 kg/ha in India.

For sowing, use of seed drill (bullock-drawn or tractor-mounted) is recommended as it results in faster sowing, quicker emergence and uniform plant stand; There should be enough moisture in the soil before sowing to ensure quick and uniform germination; In case of dry sowing, irrigation should be provided soon after, preferably with sprinklers.

Intercropping/Crop rotation: Intercropping/ mixed cropping is often practiced in rain fed (semicommercial) and subsistence agriculture as an assurance against crop failures; Crops of differing duration grown simultaneously in a field in an appropriate row arrangement can often produce more than the individual crop grown separately; Besides better utilization of light, water, and nutrient resources, it also helps to minimize disease and pest incidence; Because of their differing duration, if one crop fails due to drought at critical stages, the other crop has a chance to survive and contribute to the productivity of the system; Groundnut is intercropped generally with cereals such as maize, sorghum and pearl millet; Lately, groundnut and pigeon pea (both legumes) intercropping has become popular in Gujarat in India as this cropping system, besides adding to the stability and productivity of the system, also provides pigeon pea stalks which are used as fuel; It is essential to grow suitable combination of varieties of intercrops to derive the maximum benefit of the system; Similarly, the management practices should be cropping system-based instead of sole crops.

When manuring and fertilization is done regularly and insect pests and diseases are effectively controlled, continuous cropping of groundnut has not led to reduction in yield; but good rotation of crops helps to maintain the soil fertility, improves organic matter and physical structure of soil and reduces disease inoculum and insect pest population in the soil; As groundnut responds well to residual fertility, it is desirable to rotate it with other well fertilized crops particularly cereals; In Southeast Asia, groundnut-rice rotation is very common; In North India, efforts are being made to popularize groundnut cultivation in the spring/summer season after harvest of potato to increase cropping intensity and productivity of the high input cropping system.

<u>Intercultivation and Weed management</u>: It is essential to keep groundnut fields weed free up to 45 days after crop emergence; Even at later stages it is desirable not to have weeds in the field as

they interfere with harvesting and field inspection in case of seed production; Application of preemergence herbicides such as pendimethalin @ 1.0 -1.5 kg a.i./ha as spray or fluchloralin @ 1.0 -1.5 kg a.i./ha as pre-plant soil incorporation followed by 1-2 hand weeding, as and when needed, effectively reduces weed competition; Last hand weeding can be done along with gypsum application so as to incorporate it in the soil; Once pegs enter soil, the plant should not be disturbed; Interculture in a rainfed crop helps to reduce weeds and also encourages infiltration of rainwater.

Many farmers practice earthing up (mounting soil around the plant) to allow pegs from higher nodes to enter soil; This practice may promote growth of stem rot causing fungus (Sclerotium rolfsii) and also deteriorate the quality of earlier set mature pods while waiting for the later set pods to mature.

<u>Water management</u>: Although groundnut requires relatively less water, it cannot tolerate moisture stress at flowering, pegging, pod and seed formation stages; Exact water requirement for groundnut crop depends on the soil type and climatic conditions of a given locality; Water requirement of groundnut in different types of soil varies from 420 mm to 820 mm in India; Generally, 600-650 mm of water is sufficient; Quality of irrigation water can affect groundnut productivity; Limits for saline water for groundnut are EC (Electrical Conductivity) <4.0 mmhos/cm and RSC (Residual Sodium Carbonate) <2 meq/L.

<u>Rainfed crop</u>: Proper arrangements for drainage should be made so that excess rain water does not stagnate in the field as groundnut does not tolerate standing water in the field; Standing water even for 4–6 hours in the field can damage the crop; Planting on raised beds or ridges facilitates drainage; If supplementary irrigation is available, it should be given at critical stages such as flowering, pegging and pod and seed development; Besides increasing yield, the absence of moisture stress at the time of pod and seed development will also discourage pod and seed invasion by Aspergillus flavus and subsequent aflatoxin production.

<u>Irrigated crop</u>: Except for crop grown on residual moisture, rabi/summer/spring season crops are fully irrigated; A 2-3 week moisture stress soon after crop emergence followed by regular irrigation, often helps in inducing profuse flowering and uniform pod maturity; At pegging and pod and seed development stages, light but frequent irrigation is required; Excessive irrigation at later stages of crop growth may promote pod and seed diseases at maturity; Preferred method of irrigation is sprinkler irrigation; Flood irrigation, often practiced in flat sowing in south Asia, is not a good method of irrigation as it wastes water, results in over watering and trampling of plants in the field by workers engaged in irrigation.

<u>Use of growth regulators</u>: Use of chemical growth regulators on groundnut to suppress vegetative growth, achieve higher yield and improve pod quality have met with varying degree of success; But their use in groundnut is very common in China and Vietnam.

The following are based on the recommendations in the Chinese literature.

Dinocap (DPC): It inhibits stem growth, enhances root development and branching and increases pod yield by increasing pod number per plant; Early flowering stage is the best time to apply 80 parts per million (ppm) DPC as foliar spray.

Paclobutrazol (P 333): If groundnut plants in highly fertile soils grow more vigorously than expected, foliar application of P 333 at 60 ppm is recommended around 25-30 days after the first flowering; Its application reduces stem growth and increases pod number per plant, pod and seed mass and pod yield.

Fosamine: Fosamine strongly inhibits the growth of aerial parts and flowering of groundnut; Its effect lasts for longer duration; Foliar application of 500 ppm is recommended at late pod

forming stage to reduce late forming flowers; However, seeds from fosamine-treated groundnut have poor germination and produce abnormal seedlings; Therefore, they should not be used as seeds to grow the next crop.

<u>2</u>,3,6 trichlorobenzoic acid (TCBA): It inhibits growth of the aerial parts and late ineffective flowers; It should be sprayed (250 ppm) at peak or late peak flowering stage; Once the effect of the chemical wanes, the plant may grow more rapidly; Response to TCBA application (increase in yield) varies with the genotypes; It gives good results with large-seeded varieties of medium- and long-duration; Its effect on Spanish varieties is very little.

P 333 is the more commonly used growth regulator; It has given positive response in Vietnam also; In the USA, kylar 85 (1.2 kg/ha of powder formulation, 85% active ingredient (a.i.) in 38 L of water) is recommended for arresting excessive vegetative growth; It can also be applied in split doses; However, growth promoters are no substitute for good crop husbandry; Their maximum positive response is realized only when all other factors contributing to crop production are optimum.

# 14. Seed production in India

<u>Acts and regulations related to seeds in India</u>: Acts/Regulations, which control all seed related activities in India, are:

<u>Indian Seed Act 1966 and Seed Rules 1968 with Seeds (Control) Order 1983</u>: These are legal instruments regulating production, distribution and quality of seeds for sale and their related matters; Quality of seed is regulated through compulsory labelling and/or voluntary certification.

<u>Plant Varieties and Farmers' Rights Act (PPV&FR Act)</u>: The act, which was passed in 2001, aims to provide for establishment of effective system for protection of plant varieties and for the rights of the farmers and plant breeders, to stimulate investment in research and development and facilitate growth of seed industry and to ensure availability of high quality seeds and planting materials of improved varieties to farmers; The Protection of Plant Varieties and Farmers' Rights Authority (PPV&FR Authority), India, operational since 2005, accepts applications for 57 notified crop species.

<u>Farmers' Rights:</u> India's sui generis law recognises the farmer as a cultivator, as a conservator of agricultural gene pool and as breeder of several successful varieties; Under this act, a farmer can register and protect a variety bred/developed by him/her; This act also allows farmers to save, use, sow, re-sow, exchange, share or sell his/her farm produce including seed of a protected variety without branding it with its original registered name; In case of spurious seed supplied by a breeder/seed company, the farmer is entitled for compensation.

<u>Breeders' Rights:</u> A breeder has complete rights of commercialization of his/her registered variety either in person or anyone he/she designates; Infringement of Breeders' Rights is punishable with fines including jail term.

<u>Varietal/seed replacement</u>: Adoption rate of improved groundnut cultivars in most developing countries remains low primarily because of non-availability of their seeds in required quantities; Old cultivars, which have outlived their utility, continue to dominate groundnut cultivation affecting groundnut productivity in many countries; Normally, seed stock should be replaced by farmers every 3<sup>rd</sup> or 4<sup>th</sup> year to maintain the quality and varietal integrity of the seed and its productivity, but it rarely happens.

<u>Status of groundnut seed production</u>: In general, groundnut seed production has failed to attract participation of private seed sector due to its self-pollinating nature (absence of hybrids), low seed

multiplication ratio (1: 8-10), large seed mass/volume and quick loss of seed viability; Most of the formal seed production remains under the domain of public sector seed producing agencies, which often fail to meet their expected obligations; A majority of the groundnut seed production occurs in informal seed sector by well-off farmers often passing off the commercial produce after grading it as seed; Local traders also indulge in groundnut seed trade by procuring local commercial produce and selling it as seed in local markets after cleaning and grading the produce; This seed sold in the local markets is often a mixture of several cultivars; A majority of the farmers use either self-saved seed or buy it from local traders.

<u>Formal seed system</u>: Formal seed production in groundnut is largely undertaken by public sector seed agencies in India; Seed of only notified varieties is eligible for certification; Farmers, who want to produce Foundation/Certified seed for these agencies, must register with the State Seed Certification Agency; Parental seed should be obtained from authorised sources only and the attached seed certification label should be preserved till the seed crop is harvested; Prescribed standards for field selection and other agronomic practices including monitoring should be followed.

Formal seed production chain: Nucleus seed (produced by institution/breeder who developed the variety)  $\rightarrow$  Breeder seed Stage I (produced by institution/breeder who developed the variety or authorised sponsored breeder)  $\rightarrow$  Breeder seed Stage II (produced by institution/breeder who developed the variety or authorised sponsored breeder)  $\rightarrow$  Foundation seed Stage I (Public/Private seed sector agencies or Dept. of Agriculture)  $\rightarrow$  Foundation seed Stage II (Public/Private seed sector agencies or Dept. of Agriculture)  $\rightarrow$  Certified seed.

Two stages of seed production of Breeder seed and Foundation seed allowed in India due to low seed multiplication in groundnut; In some countries, in between Foundation seed and Certified seed, Registered seed stage is also observed.

Color of labels used in tagging the bags: Breeder seed – Golden Yellow, Foundation seed – White, Certifies seed – Azure.



Figure 62. Different color labels used for tagging different classes of seed.

Seed certification standard: Seed certification standards vary from country to country. The standards prescribed for groundnut seed production in India are given in Table 11.

Factor/Requirements	Class of seed			
r uctor / requirements	Foundation	Certified		
I. Land requirement				
No groundnut variety grown in the selected field	or the past two	or the past two		
	seasons	seasons		
II. Field standards				
lsolation distance required from the fields of other groundnut varieties (m)	3	3		
Isolation distance required from the fields of the same variety not conforming to the varietal purity requirements for certification (m)	3	3		
Presence of off-types at final inspection (maximum) in the field (%)	0.1	0.2		
Minimum number of field inspections (anytime from the initiation of flowering till maturity)	2	2		
III. Seed standards				
Pure seed (minimum) (%)	96	96		
Inert matter (maximum) (%)	4	4		
Other crop seeds (maximum)	Nil	Nil		
Weed seeds (maximum)	Nil	Nil		
Germination of the hand-shelled seeds (minimum) (%)	70	70		
Moisture content in the hand-shelled seeds (maximum) (%)	9	9		
Moisture content under vapour proof containers for hand shelled seeds (maximum) (%)	5	5		

**Table 11.** Standards for production of different classes of seed in groundnut.

No prescribed standards for Breeder seed except that the Breeder seed should be so pure as to guarantee that in the subsequent generation, i.e., certified Foundation seed conform to the prescribed standards.

Postharvest tests: Postharvest tests conducted by the State Seed Certification Agency on seed produced under seed certification program include examination of morphological characters of the seed, color reaction to certain chemicals, properties of seedlings and response of seedlings to controlled environment, growth stimulants and stable plant characters to confirm varietal trueness; Other tests include field plot tests or grow out test and DNA finger printing.

<u>Informal seed systems</u>: Enterprising farmers and local traders produce/procure seed of varieties released in public domain and sell it to the farming community as 'Truthful seed'; Selling of 'Truthful seed' indicating quality parameters through their opal green color label without certification is legally allowed in India; Seed laws regulate the quality mentioned on the label; To enhance fast and wider diffusion of improved cultivars and build seed sufficiency at local level, several schemes under informal seed sector are in operation – village seed program, farmer-participatory seed production, etc.; Local traders often procure seed through contract farming.



Figure 63. Schematic presentation of PKV methods of seed production by farmers.

<u>Management of seed crop</u>: A seed production crop requires more care and attention than that required by a commercial crop; In addition to standard cultural practices, seed crop requires some special attention, some salient points are listed below.

Agronomy: Select a well-drained-field having sandy, loamy sand or sandy loam soil which is free from soil-borne diseases and insect pests; Seed production should be undertaken under assured growing conditions; Optimum soil pH (6.0-6.3) should be maintained to ensure nutrient availability to the plants; Groundnut responds to residual soil fertility better than direct fertilization; Seed production field should have adequate levels of Ca to ensure proper seed development and high yield; Micronutrient deficiencies of Mg, B, Zn and S should always be rectified without delay; Only one seed per hill should be sown to facilitate rouging; Gap filling, if required, must be done within 10 days after sowing; Groundnut does not tolerate standing water in the field; Sprinkler or drip irrigation methods should be preferred over flood or furrow methods of irrigation.

Crop health and sanitation: Seed crop should be free from diseases and insect pests damage; Appropriate steps should be taken to keep the crop healthy; Diseased plants in the field, if any, should be rouged; Seed crop should be weed free till harvest as weeds interfere with inspection and monitoring of the field and with harvest.

### 15. Harvesting, drying and curing and storage

Several biochemical (Optical density of oil, Arginine maturity index, Methanol extract and maturity protein marker) and physical (Kernel density, Internal pericarp color, Seed hull maturity index and Hull scrap) methods to estimate maturity in groundnut available; Easiest and most practical method of assessing optimum time of harvesting is by evaluating internal pericarp color of the pods collected from a few representative plants in the field; If more than 75-80% pods in case of Spanish/Valencia cultivars and 70-75% pods in case of Virginia cultivars show internal pericarp darkening, the crop is ready for harvest; If sprouting of seeds is observed in Spanish/Valencia cultivars (due to rains at harvest time in cultivars lacking fresh seed dormancy), the crop should be harvested as soon as the conditions permit without waiting 75-80% pods to mature; In the case of end-of-season drought or under severe disease pressure, sometimes, forced premature harvesting may be required to salvage the crop; However, premature/forced harvesting may result in low shelling turnout which may disqualify the produce for seed certification; Over maturity or delay in harvesting can result greater pod loss in the soil and deterioration in pod quality.

Although commercial groundnut combines are available, the crop in Asia is harvested generally by uprooting the plants manually; In some cases, animal drawn digger, which cuts the roots of the plants below the podding zone, is also used and is followed by manual lifting of the plants.

Proper drying (removal of moisture from the produce to a point at which the moisture content of the produce comes into equilibrium with the moisture of the surrounding air) and curing (the total process of moisture removal and flavor and texture development in bringing the produce into storable condition) of the harvested produce ensure good quality of the produce; Harvested plants are kept inverted with pods facing upwards in windrows for about 2-3 days in the field for initial curing of plants and pods under ambient temperatures; If day temperatures are high, the plants should be dried in circular heaps avoiding direct exposure of pods to the sun; Exposure to high temperatures adversely affects the seed quality and germination; Groundnut pods at harvest contain moisture ranging between 35-60%; Plants should be dried till the moisture content in pods is brought down to 18-20% for mechanical threshing and 15% for hand threshing; Pods should be further dried, preferably under shade, to <10%; Groundnut stores better in pods than in seeds: It is important to remove all damaged, discolored, rotted, immature and sprouted pods. other plant materials and soil from the produce before storage; Only sound, mature, clean and well filled pods from 'true to type' plants should be selected for seed purpose; Well-dried pods with about 5% moisture content should be stored to avoid fungal and insect pests attack in storage; Pods should be stored in polythene-lined gunny bags or in some other safe storage structure in a well-ventilated and rodent free room which is not in general use and out of bound for children; Bags should be placed on wooden planks (not more than five in a stack) and away from walls to avoid damage from dampness and should be protected from damage by storage pests by dusting the bags with 5% lindane or 5% malathion dust; In case of pest outbreak in storage, seed bags after covering with polythene sheet should be fumigated with celphos (tablets) @ 3 g/bag (40 kg bag); Fumigator should use protective clothing, hand gloves, nose mask and goggles; Fresh air should be allowed in for some time by keeping windows and door open before entering the store 4-5 days after fumigation; Storage temperature should be low, temperatures below 13°C inactivate most insects and arrest growth and influence of other quality deteriorating factors; Seed quality can be maintained for at least a year at temperatures ranging from 1 to 5°C and moisture content of 7% or lower; Relative humidity in storage should be between 65-70%; Under unfavorable conditions, groundnut seeds lose viability quickly. High moisture in groundnut seeds causes more deterioration than any other single factor.



Figure 64. Harvested plants in circular heaps for field drying.



Figure 65. Tagged and stored seed pods in gunny bags.

## 16. Groundnut cultivation under polythene mulch

(The following information is adapted from literature published by the Chinese scientists.) Introduced from Japan in 1978, polythene mulch revolutionized groundnut cultivation in China by increasing its yield by 20-50%; It now covers more than 20% of the groundnut area in the country; With adoption of new high yielding varieties and improved production technology with polythene mulch, the average groundnut productivity in China increased from 1.21 t/ha in the 1970s to 2.9 t/ha in 2000s; Now becoming popular in southeast Asian countries, particularly in Vietnam.

### <u>Advantages</u>

- §□ Suitable for both cold and semi-tropical regions in all kinds of soils and in all seasons, thus extending the crop cultivation into non-traditional areas
- §□ Increases pod yield, proportion of well-filled pods and oil and protein contents and oleic to linoleic fatty acid ratio in seeds
- §□ Crop can be sown under lower temperatures (mean soil temperature over five days in the top 5 cm soil 12.5°C and above)
- §□ Requires fewer irrigations and suffers less insect pest damage as compared with nonmulched crop
- $\square$  Provides better weed control with pre-emergence herbicide application

### **Disadvantages**

- §□ Labor intensive and requires high initial investment; may not be suitable under rainfed conditions due to risks of crop failure involved
- §□ Full retrieval of polythene film at harvest is difficult film sticking to haulms leaves them unfit for fodder use and that left in the soil may cause environmental pollution (now biodegradable films are available in China but their economics needs to be worked out)
- $\$  Uiability of seeds produced under polythene mulch is lower than that of produced without mulch

### Factors that result in high yield under polythene mulch

Soil temperature: Because of above 80% sunlight transmittance, transparent polythene film raises soil temperature due to heat entrapment; Increased accumulated temperature shortens crop duration and increases pod yield; On the other hand, during the hot season, polythene film protects soil from direct sunlight, and its impermeability to hot air ensures optimum temperature for the middle growth phase of groundnut.

Soil moisture: Polythene mulch prevents soil evaporation, which accounts for 25–50% of the total quantity of water used in crop production; During heavy rains, polythene film retards soil erosion and rapid infiltration of rainwater into the soil; Optimum soil moisture ensures good emergence and seedling growth.

Soil structure: As the soil under polythene mulch remains undisturbed during the cropping period, it maintains higher porosity resulting in better root growth and nodulation.

Soil microorganisms: Population of microorganisms (fungi, actinomycetes, ammonifiers, nitrogen fixing bacteria and nutrient solubilizing bacteria) in the soil goes up significantly under polythene mulch, which accelerates the decomposition and transformation of organic matter leading to increasing levels of available nutrients in the soil.

Microclimate: During the mid-growing stage, reflection of sunlight by polythene film increases illumination between rows; Accumulated soil temperature is more and the wind speed between the rows is faster; Faster wind speeds favor air exchange and CO<sub>2</sub> movement; All these factors increase photosynthetic efficiency.

### Cultivation practices

Selection of polythene film: Polythene film with thickness varying between 0.004 mm and 0.014 mm can be used for mulching, but the thickness of 0.007 mm is optimum and economic; Film should have light transmittance  $\geq$ 70% and elasticity  $\geq$ 100%; Width of the film will depend on the cropping season and the type of variety to be grown (small seeded or large-seeded); It can vary

between 750 mm and 900 mm; Preference should be given to biodegradable film, if found economic.

Land preparation and fertilizer application: Deep tillage must be carried out for polythene mulching; Field should be prepared thoroughly before seedbed formation; Fertilizer requirement under mulched conditions is higher than that of the non-mulched conditions because of higher yields obtained under the former; FYM should be applied at the time of land preparation and the inorganic fertilizers at the time of seedbed formation.

Seedbed formation: 50-60 cm wide beds (for planting two rows) alternated with 30 cm furrows are made 4-6 days before sowing, either by a bed-former drawn by tractor or manually; Beds should be 11-12 cm high with both sides vertical and the surface smooth without clods or pebbles so that the film hugs the soil surface and is not dislodged by strong winds.



Figure 66. Seed beds for polythene mulching.

Polythene mulching: Mulching can be done either before or after sowing; Before laying polythene film over the bed, appropriate herbicide should be sprayed over the bed to kill weeds; Film can be laid manually or by a mulching machine drawn by a power tiller; Polythene film should be well stretched over the bed surface and buried on either side of the bed.



Figure 67. Manual mulching.

Figure 68. Machine mulching.

Sowing after mulching: Holes, 3-4 cm deep, are made at desired spacing with a hole-maker; Two seeds are placed in each hole and covered with soil; If soil moisture is low, some water is poured in the hole and seeds are covered with moist soil; Some additional soil is also placed over the hole to cover the edges of polythene.







Sowing before mulching: Normal sowing with two seeds per hill is done in rows 30-40 cm apart on bed surface followed by mulching;

As the soil cracks due to emerging seedlings, holes of 4–5 cm diameter are made over the crack in the film and the hole is covered with moist soil to facilitate emergence of seedlings through the holes in polythene film.

Good quality, uniform sized seeds with >98% germination should be used in sowing after appropriate chemical seed treatment; Seedlings and young plants should be inspected regularly to ensure that all emerging lateral branches are over the film; In case some seeds fail to germinate, the gaps should be filled in early, preferably with sprouted seeds.

Cultural management: Shallow hoeing should be done in furrows as and when required to keep them weed free and encourage water infiltration and moisture conservation; Irrigation should be given in furrows when long dry spell occurs at moisture sensitive stages such as peak flowering, pegging and podding; Appropriate plant protection measures and other cultural treatments should be followed as and when required.



Figure 71. Making holes at seed cracks.

Harvesting: Crop under polythene mulch matures 7–10 days earlier than the non-mulched crop, pod maturity is relatively uniform under the former; When 90% pods are mature, the crop should be harvested; During harvesting, all film should be retrieved from the soil and plants for recycling to avoid environmental pollution.

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He graduated in 1974 with a PhD degree in plant breeding from GB Pant University of Agriculture and Technology, Pantnagar, Uttarakhand. He has more than 365 publications to his credit, which include journal articles, books/book chapters, Information/Research bulletins. conference papers and newsletter articles. More than 130 groundnut cultivars developed by him and his team are released jointly with NARS in 35 countries in Asia and Africa. His services in promoting the cause of groundnut in Asia have been recognised by the national programs in Bangladesh, Nepal, India, Philippines, Sri Lanka and Vietnam. Other special recognitions include Medal for Agriculture and Rural Development,



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