### We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

6,900

186,000

200M

Download

154
Countries delivered to

Our authors are among the

**TOP 1%** 

most cited scientists

12.2%

Contributors from top 500 universities



WEB OF SCIENCE

Selection of our books indexed in the Book Citation Index in Web of Science™ Core Collection (BKCI)

Interested in publishing with us? Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.

For more information visit www.intechopen.com



## Tropical Forage Legumes in India: Status and Scope for Sustaining Livestock Production

Tejveer Singh, Srinivasan Ramakrishnan, Sanat Kumar Mahanta, Vikas C. Tyagi and Ajoy Kumar Roy

Additional information is available at the end of the chapter

http://dx.doi.org/10.5772/intechopen.81186

#### **Abstract**

Livestock contributes enormously in food and nutritional security apart from livelihood security to rural population all over the world. India has the largest number of livestock, representing over 17% of world population. Availability of forage legumes is essential for better animal health, production and increasing the nutritive value of forage-based rations, besides providing a source of biological nitrogen fixation for enriching soil, reducing land degradation and mitigating climate change. However, supply of quality green fodder in India is extremely precarious, and the gap is huge against demand. The major fodder legume crops cultivated in India are Medicago sativa, Trifolium alexandrinum, Vigna unguiculata, Vigna umbellate and range legumes are Stylosanthes spp., Desmanthus virgatus, and Clitoria ternatea. Indian subcontinent represents wide spectrum of eco-climates and reported diversity of 21 forage legumes genera viz., Desmodium, Lablab, Stylosanthes, Vigna, Macroptelium, Centrosema and browse plants Leucaena, Sesbania, Albizia, Bauhinia, Cassia, Grewia, etc. Diversity of forage legumes were collected (>3200 accessions), evaluated and sources for different biotic and abiotic stress tolerance were identified, apart from >50 cultivars developed. Considering these aspects, tropical legumes for livestock production, soil health and ecosystem services, diversity, evaluation and breeding for improved varieties are discussed in this chapter.

**Keywords:** crop wild relatives, gene introgression, germplasm, range legumes, livestock production, N-fixation



#### 1. Introduction

Cultivated forage legumes and range legumes are contributing in sustainable agriculture production apart from nutritional security to the livestock population of India. Cultivated forage legumes and range legumes are also crucial for the nutritional security for mankind as they are integral component for increased availability of animal protein and product which has higher biological value than the plant proteins. The major fodder legumes crops cultivated in India are *Medicago sativa*, *Trifolium alexandrinum*, *Vigna unguiculata*, *Mucuna pruriens*, *Vigna umbellate* and range legumes are *Stylosanthes* spp., *Desmanthus virgatus*, *Clitoria ternatea* and others. Among these, *Medicago sativa*, *Trifolium alexandrinum* and *Vigna unguiculata* are more popular among cultivated legumes and *Stylosanthes* in range legumes because of easy availability of seeds of improved varieties and well developed technology to increase the forage yield and quality. To understand the current status and scope of tropical forage legumes of India for sustaining income through livestock sector, their importance in livestock production, soil health and ecosystem services and diversity among germplasms, evaluation and breeding for improved varieties are discussed in this chapter.

#### 2. Forage legumes in livestock production

India has the largest livestock population in the world with more than 512 million heads. It supports 56.7% of the world's buffaloes, 12.5% of the world's cattle and 20.4% of the world's small ruminants (sheep and goats) [1]. Besides, the country hosts 17% of the world human population [2]. India is also the leading milk producing country in the world but milk productivity per animal basis is very low. Deficiency in quality of fodder is one of the major reasons for the low animal productivity. Although India is very rich in varied flora and fauna but there is deficiency of quality green fodder to the tune of around 35%. The animals need proper feeding to meet their nutrient requirement to express their full genetic production potential.

In fact, the sustenance of Indian rural agricultural economy depends on crop and animal farming, the two key components of a mixed farming system. Although the contribution of agricultural sector in the Indian economy is steadily declining (from 36.4% in 1982–1983 to 14.1% in 2012–2013), it still contributes employment to over 50% of the work force [3]. The contribution of livestock sector to agriculture GDP has increased to more than 28% and is likely to increase further. In the recent past, the lifestyle of people has been changed with a marked shift in food habits towards milk, milk products and meat leading to increase in demand of livestock products. Economic scenario in animal husbandry is also changing with emergence of peri-urban livestock farming and fodder markets. This indicates the huge pressure on available land, most of which, is used for arable farming and food production.

Forages form the main stay of our animal farming to reduce the competition between human beings and animals due to increasing demand for land and other inputs. Sole feeding of green forages to dairy animals is much cheaper than feeding concentrates with crop residues and has the potential of higher level of milk production. Nearly 65% of the total expenditure of milk

production in cows is attributed to the feeding of animals when both concentrates and green fodders are fed as mixed ration. When the milk production is primarily depend upon concentrate based feeding, the cost of feeding towards milk production reaches to 80%, however, in case of forage (legumes) based feeding, it is reduced to only 40% of the total expenditure [4]. Hence, any attempt towards enhancing availability of quality green fodder, and economizing the feed cost would result in better remuneration to livestock farmers/producers.

From an animal perspective, one of the largest benefits provided by legume forages is that they provide a better level of nutrition than cereal forages/grasses at a similar stage of growth, leading to greater forage intake by livestock and increased animal performance. The symbiosis between legumes and Rhizobia provides the plant with an ample supply of N and it is one of the reasons why crude protein (CP) concentrations of legumes are higher than cereals/grasses. In addition to higher concentrations of CP, forage legumes also provide a higher quality protein which may be of equal or greater importance in case of non-ruminant livestock species like equines. Legumes also contain more concentrations of digestible energy than grass/cereal forages due to the structure and development of the legume cell wall. Indeed, the cell wall of legume plants contains fewer hemicelluloses and more pectin compared to that of cereals, thus increasing their digestibility by livestock. However as the cell matures, a secondary cell wall consisting of cellulose and lignin is deposited on the interior of the primary cell wall and reduces the overall availability of the structural carbohydrates in the digestive system. In cereal forages, this phenomenon occurs in all tissues types (i.e. leaves, stems, etc.) while being primarily restricted to the vascular tissues of legume stems. The lignin of non-legumes is also more esterified to hemicelluloses and is more recalcitrant in composition (e.g. higher proportion of syringyl subunits) indicating a more suppressed degradability than in legume species.

#### 3. Forage legumes in soil health and ecosystem services

Forage legumes is essential for providing a source of biological nitrogen fixation (BNF) for enriching soil fertility (15-40 kg fixed N/ha), reduction in land degradation, disease breaks and for mitigating climate change. Estimating biological N<sub>2</sub> fixation of the forage and fodder legumes precisely is challenging because statistics on the areas and productivity of these legumes are highly difficult to obtain. Therefore, N, fixation values of forage and fodder legumes will be less reliable and also estimates of %Ndfa (nitrogen derived from atmosphere) of fodder legumes in those lands. There are very few reports available on forage legumes—BNF in India. But, all works mainly focused on application of Rhizobium inoculants to fodder legumes and testing their potential for enhancing fodder production (fresh and dry weight, crude protein content, forage quality aspects, nodulation properties, etc.). Appreciable amount of atmospheric N (~60–100%) is fixed by forage legumes annually, fixing up to 380 kg N ha<sup>-1</sup> [5]. Quantity of forage residues available for soil incorporation range from 80 to 143 kg N ha<sup>-1</sup> and rice cultivated following forage legumes yields the same as rice with 24–50 kg fertilizer N ha<sup>-1</sup> [6]. About 100–120 Mha of land is under fodder and forage legumes and green manure crops, with assumed average N, fixation rates of 200 kg N/ha/year for alfalfa, 150 kg N/ha/year for clovers (Trifolium spp.), 100 kg N/ha/year for other forages and 50 kg N/ha/year for legume-grass pastures [7]. From this assumption, total nitrogen fixation by forage and fodder legumes was calculated at 12 Tg annually (average of about 110 kg N/ha/year). But fixation by legume-grass mixtures is much more variable, ranging from a just a few kilograms to more than 250 kg N ha<sup>-1</sup>.

In India, area under fodder legumes and grasses is about 8 Mha (*Sorghum bicolor*—2.6 Mha, *Trifolium*—1.9 Mha, *Medicago*—1 Mha, other legume forages—1.9 Mha). Mean N uptake by *Trifolium alexandrinum* (240–264 kg/ha), *Medicago sativa* (216–264 kg/ha), *Vigna unguiculata* (161–181 kg/ha), *Sorghum bicolor* (128–160 kg/ha), BN hybrid (*Pennisetum glaucum* × *Pennisetum purpureum*) and *Megathyrsus maximus* (288–360 kg/ha), *Avena sativa* (120–144 kg/ha). Percent nitrogen derived from atmosphere (%Ndfa) is about 0.7 for legumes and 0.1 for cereals/ grasses. Annual contribution of BNF by forage and fodder crops in India is about 0.61 Tg/year which is nearly 5% of world BNF of forage and fodder [8]. However, majority of values available for legume N<sub>2</sub> fixation were based on shoots and above ground parts only. They did not include the fixed N present in roots, nodules and rhizodeposition in general. Published values for below-ground N as a percentage of the total plant N are 22–68% for the pulse and oilseed legumes, *Glycine max*, *Vicia faba*, *Cicer arietinum*, *Vigna radiata*, *Lupinus albus*, *Pisum sativum* and *Cajanus cajan* and 34–68% for the pasture/fodder legumes, subterranean clover, white clover and alfalfa [9–11].

In addition to BNF, many forage legumes have soil-covering growth habit similar to most grasses and deep root system which can contribute to the mitigation of many soil problems, viz., soil conservation by legume cover crops such as Stylosanthes, Crotalaria, Sesbania, Arachis and Desmodium to prevent erosion; contour-hedges with leguminous trees such as Leucaena; rehabilitation of degraded soils by legumes such as Stylosanthes spp., which are deep-rooted and adapted to infertile soils, cycle minerals from deeper soil layers resulting in soil improvement and enhanced concentration of soil organic matter through litter production [12]; the potential of legumes like Stylosanthes hamata can be exploited to ameliorate compacted soil [13]. When used as cover crop forage legumes can also control weed growth, which can be exploited as an attractive alternative to the use of herbicides. They supplement part of N fertilizer application, thus reduce nitrate leaching and eutrophication of water bodies as a consequence of surface runoff as a result of N fertilization in tropical pasture production process. Tropical forage legumes have considerable potential to increase productivity of forage-based livestock systems, while providing benefits to the environment [14]. The environmental benefits, referred as 'ecosystem services', comprise positive effects on: soil conservation and soil chemical, physical and biological properties; mitigation of global warming and of groundwater contamination; saving of fossil energy; and rehabilitation of degraded lands [14]. These features make tropical forage legumes particularly valuable at all levels of the system because of their interaction with plants, soil, animals and the atmosphere.

#### 4. Genetic resources of tropical forage legumes

Plant genetic resources (PGR) are the basic platform for screening, improving and developing fine cultivars, and the important materials for biodiversity studies including

classification, evolution and origin. Therefore, maintenance of enormous genetic diversity is mandatory for broadening the genetic base of the present and future forage improvement programmes to achieve the national goals. Extensive collection, proper evaluation, in depth study of genetic attributes and cataloging of germplasm is prerequisite for its efficient utilization. According to an estimate there are about 650 genera, 18,000 species of legumes (Leguminosae) in the world. Out of these, only about 30 legumes are used to an appreciable extent for forage production [15]. Information regarding the centre of origin of different forage crops is furnished in Table 1.

World-wide, 1500 gene banks are registered in the WIEWS (World Information and Early Warning System on PGR) database [16] and conserve a total of 7.1 million accessions belonging to 53,109 species, including major crops, minor or neglected crop species, as well as trees and wild plants. Out of total germplasms stored, 651,024 accessions belonging to forage

Genus	Species	Centre of origin	Distribution
Atylosia	scarabaeoides	India	
Centrosema	pubescens	South America	South east Asia, Indonesia and Africa
Clitoria	ternatea	Tropical America	Tropical and subtropical parts of the world
Desmanthus	virgatus	Argentina	Florida, throughout the India
Desmodium intortum		Central and South America	Throughout the tropical areas of Africa, Australia and new world
Macroptilium	atropurpureum	Central and South America	Australia, South east Asia, Pacific Islands
Macroptilium	lathyroides	India	Tropical and subtropical world
Macrotyloma	spp.	Africa and Asia	Sri Lanka
Macrotyloma	uniflorum	India	Africa
Stylosanthes	guianensis	Brazil	West Indies, Africa and Pacific Islands
Stylosanthes	hamata	Islands of West Indies	Coastal regions of north and south America
Stylosanthes	humilis	North east Brazil and Venezuela	Tropical parts of world
Stylosanthes	scabra	Tropical America	Kenya, Brazil and Queensland
Stylosanthes	seabrana	Brazil	
Lablab	purpureus	Asia or Africa	India, subtropical areas of Africa, south Asia
Cyamopsis	tetragonoloba	Africa	India (secondary centre of origin)
Trifolium	alexandrinum	Syria	Egypt
Medicago	sativa	Asia Minor	Near East and central Asia

**Table 1.** Centre of origin of different tropical forage legumes.

crops [17]. Among the international organizations major forage germplasm repositories are International Livestock Research Institute (ILRI), Nairobi, CIAT Columbia; ICARDA Syria; CSIRO-Australia, IGER-UK, USDA-Fort Collins. Forage germplasm diversity in these organizations is part of a Consultative Group of International Agricultural Research (CGIAR) coordinated activity in plant genetic resources. The ILRI Gene bank conserves more than 18 thousand accessions of forages from over 1000 species. This is one of the most diverse collections of forage grasses, legumes and fodder tree species held in any gene bank in the world [18]. CIAT gene bank keeps 35,898 accessions of beans, for 44 species of the genus *Phaseolus* from 109 countries, and 23,139 forage accessions belonging to 668 different species of grasses and legumes from 72 countries, that have been introduced over the past 30 years [19]. The IITA gene bank holds the world's largest and most diverse collection of cowpeas, with 15,122 unique samples from 88 countries, representing 70% of African cultivars and nearly half of the global diversity.

Indian sub-continent being one of the world's mega centres of crop origin and crop plant diversity, represents a wide spectrum of eco-climate and reported diversity of 21 forage legumes genera viz., Desmodium, Lablab, Stylosanthes, Vigna, Macroptelium, Centrosema and browse plants including Leucaena, Sesbania, Albizia, Bauhinia, Cassia, Grewia, etc. (Table 2). Diversity of cultivated and range legumes were collected in form of 3261 diverse germplasm accessions through different indigenous and exotic germplasm collection programme. Collected diversity of forage legumes were evaluated and sources for different biotic and abiotic stress tolerance were identified apart from >50 cultivars in different forage legumes for different geographic regions developed. Crop wild relatives (CWR) being the reservoirs of genes for stress tolerance and quality have been utilized for genetic enhancement of forage legumes. The main centre of diversity for tropical legumes viz., Dolichos, Desmodium, Vigna and Crotalaria is peninsular India and subtropical legumes viz. Teramnus, Atylosia, Pueraria and Mucuna are mainly confined to north eastern region. Likewise, rich genetic wealth for the temperate legumes namely Medicago, Melilotus, Trifolium and Hedysarum is distributed in western Himalayan region [20]. Besides, India possesses enormous diversity of minor and under-utilized fodder species such as Agrostis alba, Desmodium parvifolium, Leptochloa fusca, Potentilla fruticosa, Rhynchosia minima and Salvadora persica [21]. The forage genetic wealth of India distributed in 15 agro-climatic zones has been summarized in Table 2.

The National Bureau of Plant Genetic Resources (NBPGR) is the nodal agency for characterization, evaluation, maintenance, conservation, documentation and distribution of germplasm resources in India. Currently a total of 4594 accessions of different forage crops including cereal forages (1167), grasses (11,160, range legumes (1443), forage millets (781) and others [85] are being maintained at long term storage (LTS) module of National Gene Bank at NBPGR, New Delhi [22]. Indian Grassland and Fodder Research Institute (IGFRI) is a unique R&D organization in South Asia for sustainable agriculture through quality forage production for improved animal productivity. IGFRI being the National Active Germplasm Sites (NAGS) on forages works with its three regional stations and All India Coordinated Research Project (AICRP) on forage crops with 18 coordinated centres. At present IGFRI maintains more than 8000 accessions of 19 major forage crops including cereal forages, forage legumes, grasses and fodder tree at midterm storage [23].

S. no.	Agro climatic zone/regions	Subzones/sub regions	Prominent forage genetic resources
1	Western Himalayan Region	Jammu & Kashmir, Himachal Pradesh, Uttarakhand Hills	Medicago spp., Arundinella nepalensis, Chrysopogon, Dactylis glomerata, Eleusine, Echinochloa, Festuca, Zea mays, Kikui grass
2	Eastern Himalayan Region	Sikkim, Arunachal Pradesh, Meghalaya, Nagaland, Manipur, Tripura, Mizoram, Assam, Jalpaiguri and Cooch Bihar district of West Bengal	Rice bean, maize, range grasses, Brachiaria, broom grass and lablab bean
3	Lower Gangetic Plains	Basin plains, central alluvial plains, alluvial coastal plains and <i>Rarh</i> plains	Rice bean, guinea grass, coix and range grasses
4	Middle Gangetic Plains	12 districts of eastern Uttar Pradesh and 27 districts of Bihar plains	Maize, cowpea, rice bean, <i>Pennisetum</i> pedicellatum and coix.
5	Upper Gangetic Plains	central, south-western and northern-western Uttar Pradesh	Maize, sorghum, cowpea Senji, Dichanthium, sehima and Heteropogor
6	Trans-Gangetic Plains	Punjab, Haryana, Delhi, Chandigarh and Sri Ganganagar district of Rajasthan	Guar, maize, bajra, berseem, lucerne, guinea grass, sorghum and cowpea
7	Eastern Plateau and Hills	(i) Sub region of Wainganga, Madhya Pradesh, eastern hills and Orissa inland; (ii) Orissa northern, Madhya Pradesh, eastern hills and plateau; (iii) north and eastern Chota Nagpur hills and plateau; (iv) Chota Nagpur south, West Bengal hills and plateau, and (v) Chhattisgarh and south-western Orissa hills.	Cowpea, rice bean, <i>Pennisetum</i> pedicellatum, guinea grass, <i>Dichanthium</i> spp. and <i>Atylosia</i>
8	Central Plateau Hills	46 districts of Uttar Pradesh, Madhya Pradesh and Rajasthan	Maize, cowpea, rice bean, <i>P. pedicellatum, Coix, Atylosia,</i> sorghum, bajra, guar, <i>Cenchrus,</i> range grasses and legumes
9	Western Plateau and Hills	Maharashtra, parts of Madhya Pradesh and one district of Rajasthan	Maize, sorghum, <i>Dichanthium spp</i> . pearl millet, <i>Dichanthium carzacosum</i> , <i>Vicia</i> , cowpea, rice bean, Cenchrus, range grasses and legumes
10	Southern Plateau and Hills	35 districts of Andhra Pradesh, Karnataka and Tamil Nadu	small millet, <i>Heteropogon</i> , <i>Dichanthium</i> sehima and <i>Stylosanthes</i> sp.
11	East Coast Plains and Hills	(i) Coastal Orissa (ii) North-Coastal Gujarat (iii) South-Coastal Andhra Pradesh, North-Coastal Tamil Nadu (v) Thanjavur and (vi) South Coastal Tamil Nadu.	cowpea, rice bean, guinea grass, coix small millet, sorghum, <i>Heteropogon</i> , <i>Dichanthium</i> and <i>Stylosanthes</i> sp.
12	West Coast Plains and Hills	Western coast of Tamil Nadu, Kerala, Karnataka, Maharashtra and Goa	Congo, signal grass, Paspalum, panicum, Digitaria, Brachiaria, Iseilema laxum, Isilemia and Vicia
13	Gujarat Plains and Hills	19 districts of Gujarat	Lucerne, sorghum, small millet, pearl millet, chioori, range grasses and legumes
14	Western Dry Region	Nine districts of Rajasthan	Guar, moth, cowpea, sorghum, pearl millet and <i>Cenchrus</i> spp.
15	Island Region	Territories of the Andaman and Nicobar Islands and Lakshadweep	

Adopted from Singh et al. [77].

**Table 2.** List of prominent forage genetic resources distributed in 15 agro climatic zones of India.

#### 5. Problems associated with breeding of tropical forage legumes

Tropical forage legumes breeding programmes are associated with certain unique problems. Most of the tropical pasture legumes still possess traits of wild plants that include seed shattering, small seed size, seed dormancy, relatively slow germination rates, etc. In most of the cases we have very little knowledge about the basic biology of the species. Some of the problems include overlapping of vegetative and reproductive growth phases, uneven pod setting, nonsynchronous maturity and seed shattering in forage legumes [24]. Inherent heterozygosity as most forage species are cross pollinated. Self-incompatibility limits the extent to which they may be inbred; small floral parts make artificial hybridization tedious; poor seed producers; or produce seed with low viability as well as inherently low seedling vigor and competitive ability. Many forage species produce weak seedlings and stands are not easily established. Strains may perform differently with different systems of grazing management. Persistence of perennial tropical forage legumes is not as a single trait, but rather as a complex of traits dependent on various factors, such as diseases, insects, abiotic stresses, or management stress. Fertility barriers of one sort or another are very common in tropical forage legume breeding viz., berseem [25], owing to the wild nature of the species and inadequate knowledge of interor intra-specific variation.

#### 6. Major forage legumes of India

#### 6.1. Egyptian clover (Trifolium alexandrinum L.)

The genus *Trifolium* from the tribe *Trifolieae* of the family Leguminosae (Fabaceae) is important for its agricultural value. A few of the 237 species of this large genus have actually been cultivated [26], out of which 25 species are important as cultivated and pasture crops [27]. Egyptian clover or berseem (*T. alexandrinum* 2n = 16) is commonly cultivated as winter annual in the tropical and subtropical regions. Berseem is popular due to its multicut [4–8] nature, providing fodder for a long duration (November to May), very high quantum of green fodder (85 t/ha) and better quality of fodder (20% crude protein), high digestibility (up to 65%) and palatability. Berseem was introduced in India from Egypt in 1904, and has been established as one of the best *Rabi* (winter season) fodder crop in entire North West Zone, Hill Zone and part of Central and Eastern Zone of the country, occupying more than two million hectare [28].

Berseem being an introduced crop in India, the most important drawback in genetic improvement has been the lack of genetic variability [29, 30]. Variability in the existing gene pool has been induced through mutation, polyploidization and inter-specific hybridization. High biomass production potential along with extended growth period and resistance to biotic stresses specially root rot and stem rot have been the main target traits that were to be improved genetically. Different genetic improvement programmes carried out in various research institutes/universities by utilizing breeding approaches like selection, polyploidy

and mutation resulted in the development of >15 varieties for different berseem growing regions of India. Inter-specific hybridization have been used to improve resistance to biotic and abiotic stresses and extended length of the vegetative period because genes for wide scale adaptability are widely distributed in several wild species of *Trifolium* (**Table 3**). Interspecific hybrids of berseem with *Trifolium apertum* [31], *T. constantinopolitanum* [32], *T. resupinatum* [33] and *T. vesiculosum* [34] were successfully developed and progenies of interspecific hybrids showed introgression of various desirable traits, including late flowering and resistance to root rot and stem rot diseases.

A major breakthrough in berseem breeding in India was achieved through induction of polyploidy. The work on polyploidization of berseem genome was started with the aim to induce greater leaf and stem size [35, 36]. Autotetraploid induced by using colchicine treatment, and selection at tetraploid level resulted in the development of first polyploid variety 'Pusa Giant' with more fodder production and good regeneration capacity, uniform and higher yield throughout the season than diploid varieties released for general cultivation in India [37]. Another big achievement in polyploidy breeding was achieved at IGFRI, Jhansi by developing an autotetraploid variety namely 'Bundel Berseem-3' through colchiploidy followed by recurrent single plant selection followed with mass selection [28]. Major success in Berseem breeding was achieved by induction of longer duration mutant in Mescavi variety through gamma ray treatment which resulted in 'BL-22' a variety released

Species	Chromosome number (2n)	Desirable characters	References [31]	
T. alexandrinum ecotype Mescavi	2n = 16	Annual, multicut, highly productive, crude protein, high digestibility and palatability, basal branching		
T. alexandrinum ecotype Fahli	2n = 16	Annual, single cut, self-compatible, stem branching	[78]	
T. alexandrinum ecotype Saidi	2n = 16	Annual, 2–3 cut, stem and basal branching	[78]	
T. berytheum	2n = 16	Biotic resistance	[79]	
T. salmoneum	2n = 16	Biotic resistance	[79]	
T. apertum	2n = 16	Annual, profuse basal branching, late flowering, resistance against root rot and stem rot, high protein content	[31, 79]	
T. meironense	2n = 16	Biotic resistance	[31]	
T. resupinatum	2n = 16	Root rot and stem rot resistance, soil alkalinity tolerance	[33, 80]	
T. constantinopolitanum	2n = 16	Profuse basal branching, resistance against root rot and stem rot	[32]	
T. vesiculosum	2n = 16	Lateness, disease resistance	[25]	

Table 3. Desirable characters in berseem ecotypes and wild Trifolium species.

in 1988 for temperate and north west zone; and 'BL-180' released in 2006 for cultivation in north-west zone of India [28]. Protocol for in vitro plant regeneration from meristematic tissue and the establishment of regenerable callus culture have been developed in Berseem and related species viz., *Trifolium glomeratum*, *T. apertum*, *T. resupinatum* [38–40]. Embryo rescue technique has been effectively utilized to overcome the problems of post fertilization barriers in interspecific crosses of berseem with *Trifolium apertum*, *T. constantinopolitanum*, *T. resupinatum* and *T. vesiculosum* [31–34]. Recently, SSR based markers were developed for large scale utilization programme in Berseem [30]. Few studies on genetic diversity in Berseem and related *Trifolium* species were reported by using isozymes [29] and molecular markers [41].

#### 6.2. Stylosanthes

The genus *Stylosanthes* comprises approximately 40 species, distributed in the tropical [42], subtropical and temperate regions areas of America, Africa, and Southeast Asia. It can be grouped into two subgeneric sections, Stylosanthes and Stylosanthes. Most species are diploid (2n = 20) but polyploid species (2n = 40 and 2n = 60) also exist. Six species, namely *Stylosanthes scabra*, *S. seabrana*, *S. hamata*, *S. guianensis*, *S. humilis* and *S. viscosa*, are predominantly used as fodder legume in humid to semi-arid tropics of India (**Table 4**). These are very popular and have been widely adapted due to their ability to restore soil fertility, improve soil physical properties, and provide permanent vegetation cover as well as to provide nutritious fodder. The most specific problems associated with Stylosanthes are the limited variations of available germplasm and the susceptibility to anthracnose disease caused by the fungus *Colletotrichum gloeosporioides*. In the past, mainly five species of Stylosanthes

Species	Chromosome	Specific features	
S. scabra	2n = 4x = 40	Adapted in low rainfall areas (325 mm rainfall), suitable for semi-arid areas of Maharashtra, Andhra Pradesh, Karnataka and Tamil Nadu, <i>S. seabrana</i> and <i>S. viscosa</i> are known progenitor of <i>S. scabra</i>	
S. hamata	2n = 2x = 20 $2n = 4x = 40$	Diploid <i>S. hamata</i> and <i>S. humilis</i> are the two progenitors of this species (Curtis et al., 1995), highly palatable, grazing tolerant	
S. viscosa	2n = 2x = 20	Early emergence and highly stickiness of the leaves and stems, drought tolerant, grows on poor soils, some resistance to anthracnose, acaricidal properties	
S. humilis	2n = 2x = 20	Tolerance for salinity, susceptible to anthracnose, hairs on stems and leaves are some of the important features helpful in identifying the species	
S. guianensis	2n = 2x = 20	Suitable for humid and higher rainfall regions, adapted to acid infertile soils, tolerant of Al and Mn	
S. fruticosa	2n = 4x = 40	Allotetraploid, drought tolerant	

Table 4. Important Stylosanthes spp. with specific features.

(*S. hamata, S. scabra, S. humilis, S. viscosa* and *S. guianensis*) have been introduced primarily from Australia and evaluated at different sites in India [43–45]. This was in addition to the native perennial *S. fruticosa* Alston, which is widely distributed throughout the southern peninsular regions [46].

Testing and evaluation of wide germplasms carried out at IGFRI on acid and saline soil which contribute major part of the soils of India, indicated better adaptation of S. hamata and S. seabrana lines over other species in salinity. The potential of S. seabrana for tropical and subtropical regions of the country with clay and heavy soils, cool winters and distinct wet-dry seasonal conditions directed the use of this species in developing new breeding approach. The one could be based on the finding that it is the second progenitor of S. scabra which in turn elucidated the evolution of one of the most important Stylosanthes species, S. scabra may lead to important impacts on the efforts of improving S. scabra [47]. It may be possible to artificially synthesize *S. scabra* using pre-selected *S. viscosa* and *S.* seabrana accessions [48]. These artificial S. scabra genotypes could be used directly or more likely, be used in breeding programs. By doing so the genetic variation existing in the two diploid progenitor species would become available in improving the allotetraploid S. scabra. So far developed map and linked markers with anthracnose resistance also provide the opportunity to use them after converting them in sequence tagged sites (STS) or sequence characterized amplified region (SCAR) and then using them in direct breeding programs.

#### 6.3. Alfalfa (Medicago sativa L.)

Genus Medicago is one of the oldest forage legume comprising 60 perennial and 35 annual species, distributed mainly around the Mediterranean basin, cultivated throughout the world in diverse environments ranging both temperate and tropical environments [49]. It is generally agreed that the basic chromosome number for the genus *Medicago* are x = 7 and x = 8. Its ploidy varies from diploid (2n = 16) to polyploid (2n = 32, 48, 64). Perennial species are mainly tetraploids (2n = 4x = 32) and allogamous, however diploid (2n = 2x = 16) and hexaploid (2n = 6x = 48) cytotypes have also been reported [50]. Medicago sativa (alfalfa or lucerne) is widely cultivated as the most important forage legume in the temperate areas of the world. Lucerne is native to South West Asia as indicated by occurrence of wild types in the Cancasus and in mountainous region of Afghanistan, Iran. M. sativa complex, comprises of several members at the same ploidy level e.g., M. falcata, M. media and M. glutinosa, which freely intercross, without any hybrid sterility in the F<sub>1</sub> or later generations [51]. In India, it is grown in Maharashtra, Gujarat, Andhra Pradesh, Karnataka, Tamil Nadu, Haryana, Madhya Pradesh, Rajasthan, Punjab. The major breeding objectives in the crop include vigorous tall growing plants, better branching, quick regeneration, and balance between seed and forage yield and persistence.

Genetic resources for alfalfa improvement are limited and restricted to the *M. sativa* complex but tolerant sources for biotic and abiotic constraints are lacking in the complex [52].

The annual and perennial species of the genus *Medicago* are the reservoir of several useful agronomic traits, including disease and insect resistance and potential salt and drought tolerance having direct implication in cultivated alfalfa improvement (**Table 5**). Most of the lucerne cultivars grown in the country and worldwide are susceptible to many diseases and insect pests and the most serious constraint is the alfalfa weevil (*Hypera postica* Gyll.) [53]. Resistance to weevil has been reported in several annual species such as *M. scutellata*, *M. prostrata*, *M. turbinata* and *M. intertexta* [54–57]. Genes conferring resistance to aphid have been identified in *M. rugosa*, *M. scutellata* and *M. littoralis* [58]. Similarly, three woody species *viz. M. arborea*, *M. strasseri* and *M. citrine* of the section *Dendrotelis* have been reported as excellent sources for incorporating drought and salt tolerance in *M. sativa* [59–61]. However, due to post fertilization barrier, interspecific hybridization is difficult, so we may need to use biotechnological tools like ovule-embryo culture and electroporation.

Inter specific hybrids of *M. sativa* with some of the perennial species viz. *M. cancellata*, *M. glomerata*, *M. papillosa*, *M. prostrata*, *M. rhodopea* and *M. saxatilis* have been recovered by conventional crosses [51]. However, pollen and embryological studies demonstrated that there exist strong post fertilization barriers for recovering hybrids between *M. sativa* and annual species [62]. Utilizing embryo culture and fertilized pod culture techniques interspecific hybrids were obtained between *M. sativa* and many other annual species however, no hybrids were produced between *M. sativa* and weevil resistant *M. scutellata* [63, 64]. Bauchan and Elgin [65] reported chromosomal incompatibility and presence of two SAT chromosomes in *M. scutellata* as the major barriers for getting interspecific hybrids between *M. sativa* and *M. scutellata*. Utilizing protoplast fusion technique S<sub>1</sub> plants were obtained between *M. sativa* and *M. rugosa* and it was confirmed by genomic *in situ* hybridization (GISH) that small portions of *M. rugosa* chromosomes were present in the hybrid however, it is not clear that in which chromosome the resistance genes are present [50].

A lot of molecular information has been generated across species. However, information from M. truncatula on marker-trait association is unlikely to be exploitable in lucerne, considering the large differences between annual and perennial [66]; in addition to the differences due to the ploidy level which may further contribute to the inconsistent genetic control of some morpho-physiological traits between the two species [67]. Some breeding goals such as region-specific adaptation; drought-tolerance; improvement for forage quality should be considered [68]. Attempts have been made to produce transgenic alfalfa containing fungal chitinase gene for resistance against fungal pathogens [69], tolerance to abiotic stresses such as salt and cold [70, 71], improved forage quality [72], and sulfur-containing amino acids [73], value addition by making it an edible forage vaccine [74]. In recent years the breeding strategies for Lucerne are more towards utilizing potential of polycross methods followed with phenotypic selection. It has resulted in development of a few cultivars in recent years. The future strategies should include development of cold and drought hardy lucerne with degree of persistence for pasture and meadows, increasing genetic base, high seed production, stress tolerance, diseases and pest resistance etc.

Species	Annual/ perennial	Chromosome number (2n)	Distribution	Desirable traits	References
M. dzhawakhetica Bordz.	Perennial	32	Western Mediterranean region	Cold tolerance and resistance to <i>Phoma</i> medicaginis	[81]
M. suffruticosa Ram.	Perennial	<u>-</u>		Resistance to <i>Phoma</i> medicaginis, deep taproot system and high palatability	[81]
M. cancellata M.B.	Perennial	48	Russia	Resistance to <i>Stemphyllium</i> leaf spot	[82]
M. prostrata Jacq.	Perennial			Resistance to alfalfa weevil and potato leafhopper	[54]
M. scutellata (L.) Miller	Annual	30	Mediterranean Basin, Southern Ukraine	High biomass production, Resistance to alfalfa weevil and aphid	[83]
M. turbinata (L.) All.	Annual	_	Mediterranean Basin	Resistance to alfalfa weevil	[54, 56]
M. intertexta (L.) Miller	Annual	16	West Mediterranean Basin	Resistance to alfalfa weevil	[54, 57]
M. rugosa Desr.	Annual	30	Mediterranean Basin	Resistance to aphid	[58]
M. littoralis Rohde ex Lois	Annual	_	Mediterranean Basin, East Europe, Caucasus	Resistance to aphid	[58, 83]
M. polymorpha L	Annual	14	Europe, North Africa, Middle East, Ukraine, Georgia, Central Asia	Plant height, high seed production potential	[83]
M. lupulina L	Annual			Excellent species for sustainable agriculture, reported to improve soil health, reduce diseases and save moisture	
M. arborea Hutch.	Perennial	32	Mediterranean region	Woody species, ornamental value, drought and salt tolerant	[59, 61]
<i>M. strasseri</i> Greuter et al.	Perennial	32	Crete Iceland	Woody species, drought and salt tolerance	[60]
Medicago citrine (Font Quer) Greuter	Perennial	48	Balearic Islands	Highly drought and salt tolerant species within the section <i>Dendrotelis</i>	[84, 85]
M. truncatula Gaertner	Annual	16	Mediterranean Basin, East Europe, Russia	Genes possessing broad spectrum resistance to anthracnose, stay green genes	[86]

 Table 5. Annual and perennial Medicago species and their desirable characters.

#### 6.4. Cowpea (Vigna unguiculata (L.) Walpers)

Cowpea (2n = 2x = 22, genome size = 620 Mb) also known as 'black eye pea' or 'hungry-season crop' is an annual food and forage crop mostly grown throughout the semi-arid tropics in parts of Asia, Africa, Southern Europe, Southern United States, and Central and South America (Singh 2005). It can be grown throughout the year due to its short duration and fast growing nature. It is suitable for inter, mixed and relay cropping system. Cultivated cowpea, which is in subspecies *unguiculata*, is divided into five cultivar groups namely *Unguiculata*, *Sesquipedalis* (yard-long-bean), *Textilis*, *Biflora* and *Melanophthalmus* [75]. The commonly cultivated cowpea belongs to cultivar group *Unguiculata* the most widespread and economically important group of the species. They are pulse and vegetable and forage types. Other cultivar group *Biflora* also known as 'catjang cowpea' mainly cultivated in South Asia (India, Sri Lanka) as a pulse or as forage for hay and silage, and as a green manure crop. In Australia and Asia cowpea is primarily a fodder crop, but is also used for green manure or as a cover crop [76]. In India, the crop is cultivated around 6.5 lakh ha with 3 lakh as fodder crop in Rajasthan, Gujarat, Maharashtra, Karnataka and Tamil Nadu [24].

Cowpea was first introduced to India 1000–1500 years ago and now Indian-subcontinent appears to be a secondary centre of diversity. In India a large numbers of varieties for vegetable, pulse and fodder purpose have been developed. The breeding objectives have focused around developing lines with terminal drought tolerance, early maturity, erect growth to fit in cropping systems and enabling improved radiation use efficiency, high harvest index and resistance to diseases. The desirable traits in forage cowpea varieties are leafiness with indeterminate growth to get green fodder for a longer period. International Institute of Tropical Agriculture (IITA) has developed several dual purpose cultivars of cowpea with high grain and biomass yields and erects habit for intercropping/mixed farming purposes. In future development of cowpea lines against various forms of root-knot nematode, cowpea aphids and *Fusarium* wilt, is required. Further, development of transgenic cowpea lines with resistance to major insect pests can also be a breakthrough in cowpea breeding.

#### 7. Conclusion

Tropical forage legumes were promoted in the past with the major focus on livestock production in India. This has led to a substantial decrease in research on tropical forage legumes. In view of current climate change problems and environmental concerns, research on forage legumes should be resumed with adequate funding support at national and international levels. Newer biotic and abiotic stress tolerant varieties should be developed for the changing environmental conditions. Forage legumes have potential to contribute significantly to environment-friendly agricultural land use and sustainable livestock production in the tropics.

#### **Author details**

Tejveer Singh, Srinivasan Ramakrishnan\*, Sanat Kumar Mahanta, Vikas C. Tyagi and Ajoy Kumar Roy

\*Address all correspondence to: srinivasmic@gmail.com

ICAR-Indian Grassland and Fodder Research Institute, Jhansi, India

#### References

- [1] BAHS. Basic Animal Husbandry Statistics. New Delhi: Department of Animal Husbandry, Dairying and Fisheries, Ministry of Agriculture, Government of India; 2012. p. 130
- [2] Ghosh PK, Mahanta SK. Forage resource development in India: Looking ahead. In: Agriculture Year Book. New Delhi, India: Agriculture Today; 2014. pp. 134-140
- [3] NAAS. Augmenting Forage Resources in Rural India: Policy Issues and Strategies. Policy Paper No. 80. New Delhi: National Academy of Agricultural Sciences; 2016. pp. 1-16
- [4] Mahanta SK, Singh KK, Das MM, Das N. Forage based feeding of livestock. In: Das N, Misra AK, Maity SB, Singh KK, Das MM, editors. Forage for Sustainable Livestock Production. Delhi: Satish Serial Publishing House; 2009. pp. 407-426
- [5] Peoples MB, Ladha JK. Herridge DF enhancing legume N<sub>2</sub> fixation through plant and soil management. Plant and Soil. 1995;**174**:83-101
- [6] Ladha JK, Kundu DK, Angelo V, Coppenolle MG, Peoples MB, VR C, et al. Legume productivity and soil nitrogen dynamics in lowland rice-based cropping systems. Soil Science Society of America Journal. 1996;60:183-192
- [7] Smil V. Nitrogen in crop production: An account of global flows. Global Biogeochem Cycles. 1999;13:647-662
- [8] Rao DLN, Balachandar D. Nitrogen inputs from biological nitrogen fixation in Indian agriculture. In: Abrol et al, editors. The Indian Nitrogen Assessment: Sources of Reactive Nitrogen, Environmental and Climate Effects, Management Options, and Policies. Elsevier Inc. 2017. pp. 117-132. DOI: 10.1016/b978-0-12-811836-8.00008-2
- [9] Herridge DF, Peoples MB, Boddey RM. Global inputs of biological nitrogen fixation in agricultural systems. Plant and Soil. 2008;**311**:1-18
- [10] Yasmin K, Cadisch G, Baggs EM. Comparing <sup>15</sup>N labelling techniques for enriching above- and below ground components of the plant–soil system. Soil Biology and Biochemistry. 2006;**38**:397-400
- [11] Mahieu S, Fustec J, Faure M-L, Corre-Hellou G, Crozat Y. Comparison of two 15N labelling methods for assessing nitrogen rhizodeposition of pea. Plant and Soil. 2007;**295**: 193-205

- [12] Boddey RM, Carvalho INO, Rezende CP, Cantarutti RB, Pereira JM, Macedo R, et al. The benefit and contribution of legumes and biological N, fixation to productivity and sustainability of mixed pastures. In: Evangelista AR, CLS A, Casagrande DR, Lara MAS, Bernardes TF, editors. Proceedings of the 1st International Conference on 'Forages in Warm Climates'. Lavras, MG, Brazil: Universidade Federal de Lavras; 2015. pp. 103-140
- [13] Lesturguez G, Poss R, Hartmann C, Bourdon E, Noble A, Ratana-Anupap S. Roots of Stylosanthes hamata create macropores in the compact layer of a sandy soil. Plant and Soil. 2004;**260**:101-109
- [14] Schultze-Kraft R, Rao IM, Peters M, Clements RJ, Bai C, Liu G. Tropical forage legumes for environmental benefits: An overview. Tropical Grasslands-Forrajes Tropicales. 2018;6:1-14
- [15] Trivedi BK. Grasses and Legumes for Tropical Pasture. Jhansi: Indian Grassland and Fodder Research Institute; 2010
- [16] http://apps3.fao.org/wiews/
- [17] FAO. The State of ex situ Conservation: The Second Report on the State of the world's PGRFA. 2010. p. 351. http://www.fao.org/docrep/013/i1500e/i1500e03.pdf
- [18] www.ilri.org
- [19] www.ciat.cgiar.org
- [20] Arora RK, Chandel KPS. Botanical source areas of wild herbage legumes in India. Tropical Grasslands. 1972;6:213-221
- [21] Bhag M. Neglected and underutilized crop genetic resources for sustainable agriculture. Indian Journal of Plant Genetic Resources. 2007;20:1-14
- [22] Joshi V, Tyagi V, Kak A, Lal A. Status of genetic resources of forage crops in India: A review. Indian Journal of Plant Genetic Resources. 2009;22(3):243-252
- [23] Sahay G, Sevanayak D, Tyagi VC, Bhardwaj N, Shmed S. Tropical and subtropical forage genetic resources of India: Their conservation and utilization. Forage for Future. 2018;6:3
- [24] Roy AK, Malaviya DR, Kaushal P. Genetic improvement of fodder legumes especially dual purpose pulses. Indian Journal of Genetics and Plant Breeding. 2016;76:608-625
- [25] Malaviya DR, Roy AK, Kaushal P, Bhaskar RB, Kumar B. Evaluation of Trifolium species for defining multiple use gene pool for tropical Trifolium species. Indian Journal of Genetics. 2004;64(3):251-252
- [26] Zohary M, Heller D. The Genus Trifolium. Jerusalem: The Israel Academy of Sciences and Humanities; 1984
- [27] Lange O, Schifino-Wittmann MT. Isozyme vtriation in wild and cultivated species of the genus Trifolium L. (Leguminosae). Annals of Botany. 2000;86:339-345

- [28] Pandey KC, Roy AK. Forage Crops Varieties. Jhansi, India: IGFRI; 2011
- [29] Malaviya DR, Kumar B, Roy AK, Kaushal P, Tiwari A. Estimation of variability of five enzyme systems among wild and cultivated species of *Trifolium*. Genetic Resources and Crop Evolution. 2005;**52**:967-976
- [30] Verma P, Chandra A, Roy AK, Malaviya DR, Kaushal P, Pandey D, et al. Characterization and cross-species transferability of genomic SSR markers in berseem (*Trifolium alexandrinum* L.), an important multi-cut annual forage legume. Molecular Breeding. 2015;**35**:23
- [31] Malaviya DR, Roy AK, Kaushal P, Kumar B, Tiwari A, Lorenzoni C. Development and characterization of interspecific hybrids of *Trifolium alexandrinum* × *T. apertum* using embryo rescue. Plant Breeding. 2004;**123**:536-542
- [32] Roy AK, Malaviya DR, Kaushal P, Kumar B, Tiwari A. Inter-specific hybridization of *Trifolium alexandrinum* with *T. constantinopolitanum* using embryo rescue. Plant Cell Reports. 2004;**22**:705-710
- [33] Kaushal P, Malaviya DR, Roy AK, Tiwari A, Kumar B. *Trifolium alexandrinum* × *T. resupinatum*—Interspecific hybrids developed through embryo rescue. Plant Cell, Tissue and Organ Culture. 2005;83(2):137-144
- [34] Kaur A, Kaur KP, Kalia A. Generation of inter-specific hybrids between *Trifolium vesicu-losum* and T. *alexandrinum* using embryo rescue. Euphytica. 2017;**213**:253
- [35] Mehta R, Swaminathan M. Studies on induced polyploids in forage crops. 1. Survey of previous work. Indian Journal of Genetics. 1957;17:27-57
- [36] Sikka SM, Mehta RK, Swaminathan MS. Studies on induced polyploids in forage crops II. Colchicine treatment methods for berseem and senji. Indian Journal of Genetics. 1959;19(1):90-97
- [37] Mehta R, Swaminathan M. 'Pusa Giant' berseem. Indian Farming. 1965;15(6):4-5
- [38] Kaushal P, Malaviya DR, Roy AK, Kumar B. In vitro response of immature zygotic embryos of *Trifolium alexandrinum* (Egyptian clover). Plant Cell Biotechnology and Molecular Biology. 2004;**5**(3-4):109-114
- [39] Kaushal P, Tiwari A, Roy AK, Malaviya DR, Kumar B. *In vitro* regeneration of *Trifolium glomeratum*. Biologia Plantarum. 2006;**50**(4):693-696
- [40] Malaviya DR, Roy AK, Tiwari A, Kaushal P, Kumar B. *In vitro* callusing and regeneration in *Trifolium resupinatum*—A fodder legume. Cytologia. 2006;71(3):229-235
- [41] Kaila V, Sood VK, Chaudhary HK, Bhandari JC, Sood A, Mittal RK. Morphological and RAPD markers-mediated assessment of genetic diversity amongst various *Trifolium* species and identification of potential ideotypes for genetic upgradation of berseem under changed climate in mid-hills of north-west Himalayas. Indian Journal of Genetics. 2009; 69(4):1-6

- [42] Kirkbide JH, De Kirkbride CG. Typification of Stylosanthes (Leguminosae) and its sections. Taxon. 1985;36:455-458
- [43] Rai P, Patil BD. Production potential of Stylosanthes in pure and mixed swards. Indian Journal of Range Management and Agroforestry. 1985;6:35-57
- [44] Chatterjee BN, Singh RD, Matti S. An agronomical appraisal of Stylosanthes in eastern region of India. Indian Journal of Range Management and Agroforestry. 1985;6:27-33
- [45] Ramesh CR, Mal B, Hazara CR, Sukanya DH, Ramamurthy V, Chakraborty S. Status of Stylosanthes development in other countries. III. Stylosanthes development and utilization in India. Tropical Grasslands. 1997;31:467-475
- [46] Hooker JD. Flora of British India. In: Sabiaceae to Cornaceae. London: L. Reeve and Co. Ltd; 1879. p. 148
- [47] Tewari S, Chandra A. Genetical assessment of diploid progenitors of S. scabra by isozyme, RAPD and STS markers: A possible strategy for improvement of drought tolerant allo-tetraploid S. scabra species. Euphytica. 2008;162(1):39-50
- [48] Liu CJ, Musial JM. Stylosanthes sp. aff. S. scabra: A putative diploid progenitor of Stylosanthes scabra (Fabaceae). Plant Systematics and Evolution. 1997;208:99-105
- [49] Small E, Jomphe M. A synopsis of the genus Medicago. Canadian Journal of Botany. 1989;67:3260-3294
- [50] Mizukami Y, Kato M, Takamizo T, Kanbe M, Inami S, Hattori K. Interspecific hybrids between Medicago sativa L. and annual Medicago containing Alfalfa weevil resistance. Plant Cell Tissue and Organ Culture. 2006;84:79-88
- [51] Lesins KA, Lesins I. Genus Medicago (Leguminosae) a Taxonomic Study. The Hague, The Netherlands: Junk by Publishers; 1979. p. 228
- [52] Barnes DK, Bingham ET, Murphy RP, Hunt OJ, Beard DF, Skrdla WH, et al. Alfalfa germplasm in the United States: Genetic vulnerability, use, and maintenance. In: USDA-ARS Tech. Bull. Hyattsville, MD: USDA-ARS; 1977. p. 1571
- [53] Chandra A, Pandey KC. Assessment of genetic variation in lucerne (Medicago sativa L.) using protease inhibitor activities and RAPD markers. Journal of Environmental Biology. 2011;32:559-565
- [54] Sorensen EL, Horber EK. Glandular trichomes on Medicago: Building self defense against insects. In: Rep. of the Twenty-Ninth Alfalfa Improvement Conf. 1984. p. 135
- [55] Kleitner GL, Sorensen EL. Erect glandular trichomes of Medicago scutellata (L.) Mill: Gland deveropment and early secretion. Botanical Gazette. 1983;144(2):165-174
- [56] Chandra A, Pandey KC, Singh UP. Medicago scutellata—A possible source of weevil resistance for lucerne improvement. Indian Journal of Plant Genetic Resources. 2006; **19**:291-293

- [57] Chandra A, Pandey KC. Effect of proteinase inhibitors on Indian alfalfa weevil (Hypera postica Gyll.) growth and development. Acta Physiologiae Plantarum. 2008;30:501-505
- [58] Verma JS, Mishra SN. Advances in forage plant improvement in upper Gangetic Plains. In: Hazra CR, Mishri B, editors. New Vistas in Forage Production. Jhansi-284003, India: Indian Grassland and Fodder Research Institute. 1995. pp. 83-96
- [59] Olives G. La alfalfa arbórea. In: Pesca y Alimentación. Madrid: Ministerio de Agricultura; 1969
- [60] Greuter W, Matthäs U, Risse H. Notes on Cardaegan plants. 3. Medicago strasseri, a new leguminous shrub from Kriti. Wild. 1982;12:201-206
- [61] Rosato M, Castro M, Rosselló JA. Relationships of the Woody Medicago species (section Dendrotelis) assessed by molecular cytogenetic analyses. Annals of Botany. 2008;102(1):15-22
- [62] MCCOY T. Interspecific hybridization of *Medicago sativa* L. and *M. rupestris* M.B. using ovule-embryo-culture. Canadian Journal of Genetics and Cytology. 1985;**27**(2):238-245
- [63] Wang JW, Sorensen EL, Liang GH. In vitro culture of pods from annual and perennial *Medicago species*. Plant Cell Reports. 1984;**3**:146-148
- [64] McCoy TJ, Smith LY. Interspecifi c hybridization of perennial *Medicago* species using ovule-embryo culture. Theoretical and Applied Genetics. 1986;**71**:772-783
- [65] Bauchan GR, Elgin JH Jr. A new chromosome number for the genus *Medicago*. Crop Science. 1984;**24**:193-195
- [66] Volenec JJ, Cunningham SM, Haagenson DM, Berg WK, BC J, Wiersma DW. Physiological genetics improvement: Past failures, future prospects. Field Crops Research. 2002;75:97-110
- [67] Bingham ET, Groose RW, Woodfield DR, Kidwell KK. Complementary gene interactions in alfalfa are greater in autotetraploids than diploids. Crop Science. 1994;34:823-829
- [68] Annicchiarico P, Scotti C, Carelli M, Pecetti L. Questions and avenues for lucerne improvement. Czech Journal of Genetics and Plant Breeding. 2010;46:1-13
- [69] Hipskind JD, Paiva NL. Constitutive accumulation of a resveratrol-glucoside in transgenic alfalfa increases resistance to *Phoma medicaginis*. Molecular Plant-Microbe Interactions. 2000;**13**(5):551-562
- [70] Winicov I. Alfin1 transcription factor over expression enhances plant root growth under normal and saline conditions and improves salt tolerance in alfalfa. Planta. 2000;**210**: 416-422
- [71] McKersie BD, Murnaghan J, Jones KS, Bowley SR. Iron-superoxide dismutase expression in transgenic alfalfa increases winter survival without a detectable increase in photosynthetic oxidative stress tolerance. Plant Physiology. 2000;**1224**:1427-1437

- [72] Xie DY, Sharma SB, Paiva NL, Ferreira D, Dixon RA. Role of anthocyanidin reductase, encoded by BANYULS in plant flavonoid biosynthesis. Science. 2003;**299**:396-399
- [73] Tabe LM, Wardley-Richardson T, Ceriotti A, Aryan A, McNabb W, Moore A, et al. A biotechnological approach to improving the nutritive value of alfalfa. Journal of Animal Science. 1995;73:2752-2759
- [74] Carrillo C, Wigdorovitz A, Trono K, Dus Santos MJ, Castañón S, Sadir AM. Induction of a virus-specific antibody response to foot and mouth disease virus using the structural protein VP1 expressed in transgenic potato plants. Viral Immunology. 2001;14:49-57
- [75] Pasquet RS. Cultivated cowpea (Vigna unguiculata): Genetic organization and domestication. In: Pickersgill B, Lock JM, editors. Advances in Legume Systematics 8: Legumes of Economic Importance. Kew, UK: Royal Botanic Gardens; 1996. pp. 101-108
- [76] Tarawali SA, Singh BB, Peters M, Blade SF. Cowpea haulms as fodder. In: Singh BB, Mohan Raj DR, Dashiell KE, Jackai LEN, editors. Advances in Cowpea Research. Sayce, Devon, UK: Co-publication Intl Inst Tropical Agric (IITA) and Japan Intl Res Center Agric Sci (JIRCAS); 1997. pp. 313-325
- [77] Singh AK, Varma N, Yadav SK, Mohanty A, Singh S, Singh S. Indian forage genetic resources: Perspectives and strategies. Progressive Agriculture. 2009;9(2):250-256
- [78] Singh T, Malaviya DR, Kaushal P. Genetic analysis of some morphological traits in Egyptian clover (Trifolium alexandrinum L.) sustainable use of grassland resources for forage production, biodiversity and environmental protection. In: Roy AK, Kumar RV, Agrawal RK, Mahanta SK, Singh JB, Das MM, Dwivedi KK, Prabhu G, Shah NK, editors. Extended Abstracts 23rd International Grassland Congress. 2015. Paper ID: 1467
- [79] Putiyevsky E, Katznelson J. Cytogenetic studies in *Trifolium* spp. related to berseem. I. Intra- and inter-specific hybrid seed formation. Theoretical and Applied Genetics. 1973;**43**:351-358
- [80] Bhaskar RB, Malviya DR, Roy AK, Kaushal P. Evaluation of exotic Trifolium accessions for disease incidence and resistance. In: Abstr. Nat. Symp. on 'Grassland and Fodder Research in the New Millennium'; IGFRI, Jhansi. 2002. pp. 31-32
- [81] Renfro BL, Sprague EW. Reaction of Medicago species to eight alfalfa pathogens. Agronomy Journal. 1959;51:481-483
- [82] Borges OL, Stanford EH, Webster RK. Selection for resistance to Stemphylium botryosum in alfalfa. Crop Science. 1976;**16**:456-458
- [83] Chandra A. Screening global Medicago germplasm for weevil (Hypera postica Gyll.) tolerance and estimation of genetic variability using molecular markers. Euphytica. 2009;1693:363-374
- [84] Boscaiu M, Riera J, Estrelles E, Güemes J. Números cromosomáticos de plantas occidentales 751-776. Anales del Jardín Botánico de Madrid. 1997;55:430-431

- [85] Alomar G, Mus M, Rosselló JA. Flora endèmica de les Balears. Palma de Mallorca: Consell Insular de Mallorca; 1997
- [86] Zhou C, Han L, Pislariu C, Nakashima J, Fu C, Jiang Q, et al. From model to crop: Functional analysis of a STAY-GREEN gene in the model legume *Medicago truncatula* and effective use of the gene for alfalfa improvement. Plant Physiology. 2011;157:1483-1496





## IntechOpen

# IntechOpen