Chapter 41
Soybean Production in the Americas

Woo-Suk Chang, Hae-In Lee and Mariangela Hungria

Abstract Soybean (*Glycine max* (L.) Merr.) is one of the most important legume crops in the world. Approximately 80 % of the world's soybean is produced by countries in North and South America. Biological nitrogen fixation (BNF) in soybeans, due to the symbiosis with *Bradyrhizobium*, is economically and ecologically beneficial because it reduces the need for synthetic N-fertilizers. This chapter describes history and trends of soybean production, influence of soybean BNF, and development of inoculants to increase the crop yield in North and South America.

41.1 History of Soybean Cultivation in North and South America

Farmers grow soybeans for various purposes such as human food, animal feed, and industrial applications. Since the soybean contains an average of 40 % protein and 20 % oil, it can be a great nutrient source for both humans and animals. A number of soy foods such as tofu, soy sauce, and soymilk are popular in many Asian countries, while in the United States most soybeans for human consumption are used to produce edible oil products such as cooking oils, margarine, mayonnaise, and vegetable shortening. In South America, in addition to the use of oil, soybean-based milk and soups are part of the daily meals of children in public schools. Soybeans are largely added to a variety of processed foods, from meat-derived to crackers. They are

W.-S. Chang (✉) · H.-I. Lee
Department of Biology, University of Texas-Arlington, Arlington, Texas 76019, USA
Tel.: + 1 8172723280
e-mail: wschang@uta.edu

H.-I. Lee
Tel.: + 1 8172723264
e-mail: hilee@uta.edu

M. Hungria
Embrapa Soja, Cx. Postal 231, Londrina, Paraná 86001–970, Brazil
Tel.: + 55 4333716206
e-mail: mariangela.hungria@embrapa.br

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also considered as an excellent protein source for livestock and are used to produce industrial products such as biodiesel, soy-based lubricants, and soy inks.

Soybean originated in East Asia. It was first cultivated in China around 1100 BC and had spread to Japan and many other countries by the first century AD. In the 1980s, the genus *Glycine* was split into the subgenera *Glycine* (wild perennial species) and *Soja* (including both wild-*Glycine soja* Sieb. and cultivated-*Glycine max* genotypes). *G. max* was introduced into Europe (Paris) only in 1740. It was first introduced to North America from China by Samuel Bowen in 1765 (Hymowitz and Harlan 1983). Soybean became a popular crop in the U.S. between the mid-19th to early 20th centuries. The American Soybean Association was founded in 1920 by soybean farmers and extension workers. World War II kindled the prosperity of the soybean farming in the U.S. as the drastic increase in demand for lubricants and oils by the war increased the soybean demand. The U.S. has been the leading country for soybean production in the world. In Canada, soybeans were first cultivated at the Ontario Agricultural College in 1881, and at present Canada is the world's 7th largest soybean producing country.

Glycine spp. were introduced to South America by the end of the 19th century (Argentina, 1880; Brazil, 1882). Seeds from the U.S., Brazil, Argentina, and Japan were taken to Paraguay in the 1920s, and later to Bolivia, Colombia, Uruguay, and Venezuela. Commercial scale production started in the 1940s in Brazil and Argentina, which led to increased production by the 1960s. One major event for soybean expansion in South America was breeding of soybean cultivars with a long juvenile period, which allowed for the production at very low latitudes.

41.2 Soybean Production in North and South America

World soybean production has been rapidly increasing since 1990, mainly due to increased production in North and South America. The top five countries for soybean production include the U.S., Brazil, Argentina, China, and India, which represent more than 90 % of world production (Fig. 41.1).

The soybean cultivation area in the U.S. has rapidly expanded since the mid-20th century and reached about 31 million ha in 2013. Soybeans are produced in more than one third of the states but mostly in the Midwestern states Iowa, Illinois, Minnesota, and Indiana (order of top producing states). The soybean cultivation area declined in 2007, when many farmers turned to the cultivation of corn to supply the growing bioethanol industry (Salvagiotti et al. 2008). Nevertheless, the total production of soybean and the crop yield has been increasing steadily, mostly due to improved varieties and advances in biotechnology. The national average yield in the U.S. has increased from 1581 kg ha^{-1} in 1960 to 2919 kg ha^{-1} in 2013. In Canada, the average crop yield has reached 3300 kg ha^{-1} in 2012. Bioengineered soybeans are one of the most successful crops commercially in North and South America. They account for 93 % of the soybean produced in the U.S., and for about 90 and 100 % in Brazil and Argentina, respectively. Most of these bioengineered soybeans are improved

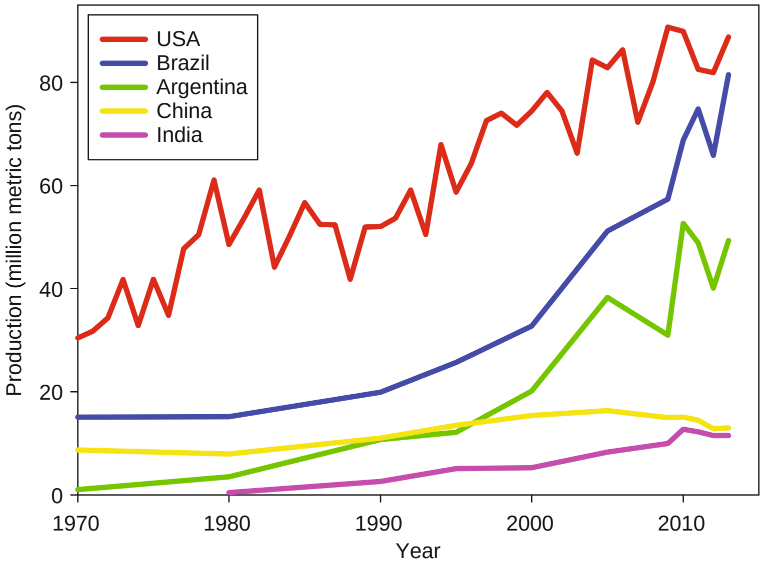

Fig. 41.1 Soybean production of the top 5 countries in the world. The data for the U.S. were retrieved from the USDA statistics for the annual production from 1970 to 2013 (http://www.nass.usda.gov), while the data for the other countries represent occasional years (1970, 1980, 1990, 1995, 2000, 2005, and 2009–2013)

varieties with high resistance against herbicides, and now double resistance to both herbicides and insecticides.

Soybean production continues to grow at impressive rates in the South American countries that account for more than half of the global production (Fig. 41.1); estimates are that soon Brazil may replace the U.S. as the leading producer in the world. National average yield in Brazil increased from 1166 kg ha^{-1} in 1968/1969 to 3115 kg ha^{-1} in 2010/2011. The potential soybean production has been estimated at 8000 kg ha^{-1}, and field trials in North and South America have reported yields of 4000 to 6000 kg ha^{-1}. In addition, while not scientifically proven, there are reports of U.S. farm yields exceeding 10,000 kg ha^{-1} (Hungria and Campo 2004; Hungria et al. 2005, 2006; Hungria and Mendes 2014).

41.3 Nitrogen Fixation Associated with Soybean in North and South America

In 1981, LaRue and Patterson estimated that the average amount of BNF associated with soybean in the U.S. might not exceed 75 kg N ha^{-1} (Larue and Patterson 1981). However, Salvagiotti and colleagues (2008) recently reviewed soybean nitrogen fixation more comprehensively by analyzing 637 data sets from field studies published in international journals between 1966 and 2006. They calculated the average BNF in soybeans to be as high as 111–125 kg N ha^{-1}. Although their analysis was performed with international data sets from many different countries, almost half of the data sets were from studies in the U.S. and Canada.

Interestingly, Herridge et al. (2008) reported that the percentage of N derived from air (%Ndfa) by soybean in the U.S. is on average 60 % of the total Ndfa, which indeed gives around 140 kg N ha^{-1}. Estimates of soybean-associated BNF contribution to the total Ndfa in South America are greater, around 80 % in both Brazil and Argentina (Herridge et al. 2008). This would correspond with 190 kg N ha^{-1}; however, there are reports of contributions higher than 300 kg N ha^{-1} (Hungria et al. 2006). The lower contribution of BNF in the U.S. might be attributed to heavy applications of N-fertilizers in agriculture, with leftovers for the soybean, as well as to low adoption of inoculation by the farmers.

41.4 Inoculants and N-fertilizers

Bradyrhizobium In the Americas all inoculant strains belong to the genus Bradyrhizobium, including the species *B. japonicum*, *B. diazoefficiens*, and *B. elkanii* (Delamuta et al. 2013). The *Bradyrhizobium*-soybean symbiosis requires specific signal exchange between the two partners. Soybean secretes isoflavonoids, such as genistein and daidzein, into the rhizosphere and these substances trigger nodulation (*nod*) genes in *Bradyrhizobium*. These *nod* genes encode Nod factors, which initiate root hair curling and formation of infection threads. *Bradyrhizobium* cells invade the host plant root cells through the infection threads and subsequently form a new organelle (i.e., the nodule), in which bacteria develop into bacteroids. Within the nodule, the bacteroids can express oxygen-sensitive nitrogenase (*nif*) genes due to the microoxic condition (see Chap. 23).

The Agricultural Research Service (ARS) of the U.S. Department of Agriculture (USDA) has collected *Rhizobium* isolates, including *Bradyrhizobium*, since the early 1900's, and the collection is well known internationally. As of March 2014, the Germplasm Resources Information Network (GRIN) database of the USDA/ARS reports 534 strains isolated from soybeans. Many of the strains were isolated by research programs in the 1930s and 1940s, and the USDA began to produce inoculants for the small research field in the 1950s and 1960s. The need for a *Rhizobium* culture collection was emphasized as the importance of BNF was recognized as a means to supplement hydrocarbon-based fertilizers.

Inoculants The first U.S. patent for pure cultures of rhizobia to be used in conjunction with artificial inoculation was issued in 1896. Two years later the first inoculant company, Nitragin, was established in the U.S. Inoculants for soybean were commercialized by the company in the early 1900s. The search for an adequate medium to carry rhizobia focused on peat which ultimately became globally established as the "gold standard". By the 1950s, inoculant industries expanded into South America.

Particularly, in the last two decades there has been a shift away from peat towards liquid formulations, as they are easily applied to the seeds and preferred by the farmers. However, concerns about liquid inoculants were raised long ago; Burton and Curley (1965) reported inferior performance of liquid-based inoculants even though 2.5 times as many rhizobia as in peat-based inocula were applied to the seeds. Several decades later, although protective molecules, adhesives and several polymers have been introduced into inoculants, innovation in liquid, gel or other non-solid formulation has been modest. Therefore, a need for a second generation of inoculants has arisen in the soybean industry (more information in section 41.5).

Most farmers in South America are convinced of the benefits of soybean reinoculation with elite strains selected through research programs and registered in governmental agencies. This results in a market size estimated at over 50 million doses of inoculants applied by about 60 % of the farmers (some farmers use multiple doses, especially in first-year planting areas). In contrast, the use of inoculants in the U.S. is a common practice for only about 15 % of the soybean production area, and is perhaps due to low prices of N-fertilizers or a perception of their insignificant performance. An evaluation of the effect of inoculants used in Midwestern states, including Indiana, Iowa, Minnesota, Nebraska, and Wisconsin, between 2000 and 2008 revealed that the use of inoculants was not successful in enhancing crop yield or economic return when used in soils that already had a history of soybean cultivation (De Bruin et al. 2010). Rising prices of N-fertilizers and concerns about the resultant nitrate contamination of the environment may shift this inoculation panorama.

Competitiveness and effectiveness of inoculants have been considered key properties to guarantee the success of nitrogen fixation with the soybean crop. There are long time concerns, however, about highly competitive but low effective indigenous/naturalized *Bradyrhizobium* population in the soybean rhizosphere (Baldwin and Fred 1929). One main example is of the USDA 123 serocluster in the Midwestern U.S., given rise to 60 to 80 % of the nodules, while more effective inoculant strains result in only 10 to 20 % of the nodules formed. This serogroup is also a concern in Canada, Korea, and Brazil (Hungria and Mendes 2014). Several laboratories have tried to identify bacterial properties implicated in competitiveness, such as mobility, chemotaxis, exopolysaccharides, bacteriocins, capacity to respond to several substrates. Interestingly, molecular approaches have also been considered (Hungria et al. 2006); however, application of new strategies or genetically engineered strains to agriculture has been limited.

Reports of the impossibility of displacing competitive strains established in the soils (Thies et al. 1991) have probably discouraged research to select elite strains, and farmers to adopt inoculation in the U.S. Currently, in South America probably more than 90 % of the areas cropped to soybean have been previously inoculated, showing naturalized bradyrhizobial populations ranging from 10^3 to up to 10^6 cells g^{-1} of soil. Dozens of field experiments performed in the last 20 years have consistently shown that reinoculation of soybean results in yield increases. The analysis of sets of experiments indicates that the average increase in yield due to annual reinoculation is 8 % and 14 % in Brazil and Argentina, respectively, compared to the non-inoculated control (Hungria et al. 2006; Hungria and Mendes, 2014). In addition, it is worth noting that massive reinoculation reported in South America may cause the replacement of persistent strains with more efficient strains (Hungria et al. 2006; Hungria and Mendes 2014).

N-fertilizers Less than 40 % of the soybean cultivation area in the U.S. is supplemented with chemically synthesized fertilizers. Nevertheless, as soybean represents a high profit crop, there is also increasing pressure for farmers to use N-fertilizers. Soybeans apparently do not require additional N-fertilizers in normal soil conditions due to BNF with its symbiotic partner *Bradyrhizobium*. Additionally, one of the factors confounding our understanding of both the efficiency of inoculants and the use of N-fertilizers is the effect of crop rotation. High residual soil N as a result of corn cultivation, usually in the Midwest states (i.e., Corn Belt area), may influence interpretation of soybean yield when soybean is planted behind corn (Stewart Smith, *personal communication*). In South America, studies performed over the last two decades demonstrated that soil N availability as low as 20–40 kg of N ha^{-1} may decrease nodulation and N_2 fixation in soybeans with no benefits to crop yield. In addition, no benefits have been reported with the addition of 30 to 50 kg of N ha^{-1} at flowering, early or late pod filling stages. Indeed, applications of up to 400 kg N ha^{-1}, split across ten applications did not result in higher yields in comparison to inoculation with elite strains (Hungria et al. 2006; Hungria and Mendes 2014).

41.5 A Second Generation of Inoculants

Promising results have been reported with the use of other microorganisms and molecules as inoculants for soybean. Co-inoculation with plant-growth-promoting rhizobacteria (PGPR) such as *Azospirillum brasilense* may increase yield (Hungria et al. 2013). One of the most exciting new concepts is to take advantage of the molecular dialogue between plant and bacterium. Spaink and colleagues (Spaink et al. 1992) separated Nod metabolites from several *Rhizobium* and *Bradyrhizobium* strains using thin-layer chromatography (TLC) and found that common *nod* genes, such as *nodABC*, play a key role in Nod metabolite production. The Nod metabolite produced by *Bradyrhizobium* strain USDA 110 was subsequently identified as a lipochitooligosaccharide (LCO) signaling molecule, similar to those from *Rhizobium* species (Sanjuan et al. 1992). The LCO from USDA 110 was able to promote the growth of soybean at a low concentration (100 nM) in hydroponic conditions (Souleimanov et al. 2002). The Novozymes' patented LCO molecule product, Optimize®, has been commercially available both in North and South America. Apparently, field responses with LCO have shown slight but consistent increases in soybean yields by an average of ca. 2–3 %, although the increases may depend on specific conditions (Leibovitch et al. 2002). Additionally, it can bring other benefits such as promoting root growth (Souleimanov et al. 2002). The positive effects of Nod factors or bacterial metabolites can be also observed in non-legumes such as corn (Liang et al. 2013; Marks et al. 2013).

41.6 Perspectives on Economical and Ecological Benefits of BNF with Soybeans

Enhancement of soybean BNF has profound economical and ecological benefits by reducing the use of chemical N-fertilizers that are a significant source of greenhouse gas emissions and cause degradation of water sources ranging from groundwater, where derivatives of nitrogen cause the "blue-baby" syndrome (Knobeloch et al. 2000), to oceanic systems where increased nitrogen leads to hypoxia (lack of oxygen) and large scale "dead zones" in the Northern Gulf of Mexico (Burkart and James 1999).

The enormous demand for N by the soybean crop results from the need of about 80 kg of N per 1000 kg of grains; considering that the efficiency of N-fertilizers is rarely more than 50 %, it is calculated that there is a need of 480 kg N-fertilizer ha^{-1}, or about 700 kg of urea, the most broadly used N-source (Hungria et al. 2006). In an exercise to quantify the global contribution of BNF with the soybean crop, Herridge et al. (2008) concluded that 16.4 Tg of N is fixed annually, representing 77 % of the N fixed by all crop legumes. Considering the high price of N-fertilizers in Brazil—70 % of which is imported—in combination with the area cropped, the average BNF rate, and the national average yield, BNF is estimated to save about US$ 15 billion yearly. Not least important, by using a conservative rate of 4.5 kg of e-CO_2 kg^{-1} (CO_2 equivalent) of N-fertilizer, the replacement of BNF by N-fertilizers in Brazil would result in the emission of about 45 million tons of e-CO_2(Hungria and Mendes, 2014). The importance of this approach, specifically in South America, with a strong partnership between plant breeders and microbiologists towards improving the contribution of BNF should be highlighted. Outstanding symbiotic performances as those reported in South America can be lost in a few years if plant breeders and microbiologists do not continue the long-term and successful partnership towards increasing BNF contribution. A continuous pressure from companies to supply N-fertilizers to the soybean crop claiming higher yields can also have profound impacts in BNF contribution. Another important consideration is the increasing use of pesticides and other chemicals in the seed treatment, which can drastically affect *Bradyrhizobium* survival and impair BNF (Mendes and Hungria 2014). On the contrary, disclosure and dissemination of the successful results such as those achieved in South America can stimulate more farmers to use inoculants.

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References

Baldwin IL, Fred EB (1929) Strain variation in the root nodule bacteria of clover, *Rhizobium trifolii*. J Bacteriol 17:17–18

Burkart MR, James, DE (1999) Agricultural-nitrogen contributions to hypoxia in the Gulf of Mexico. J Environ Qual 28: 850–859.

Burton JC, Curley RL (1965) Comparative efficiency of liquid and peat-base inoculants on field-grown soybean (*Glycine max*). Agron J 57:379–381

De Bruin JL, Pedersen P, Conley SP et al (2010) Probability of yield response to inoculants in fields with a history of soybean. Crop Sci 50:265–272

Delamuta JR, Ribeiro RA, Ormeno-Orrillo E et al (2013) Polyphasic evidence supporting the reclassification of *Bradyrhizobium japonicum* group Ia strains as *Bradyrhizobium diazoefficiens* sp. nov. Int J Syst Evol Microbiol 63:3342–3351

Herridge DF, Peoples MB, Boddey RM (2008) Global inputs of biological nitrogen fixation in agricultural systems. Plant Soil 311:1–18

Hungria M, Campo RJ (2004) Economical and environmental benefits of inoculation and biological nitrogen fixation with soybean: situation in South America. In: World Soybean Research Conference, 7., International Soybean Processing and Utilization Conference, 4., Brazilian Soybean Congress, 3. Proceedings, Embrapa Soja, Londrina, Brazil, pp 488–498

Hungria M, Mendes IC (2014) Nitrogen fixation with soybean: the perfect symbiosis?. In: de Bruijn F (ed) Biological nitrogen fixation. Wiley-Blackwell, New Jersey. (Wiley Publisher, Hoboken)

Hungria M, Franchini JC, Campo RJ et al (2005) The importance of nitrogen fixation to soybean cropping in South American. In: Werner D, Newton WE (eds) Nitrogen fixation in agriculture, forestry, ecology, and the environment. Springer, Dordrecht, pp 25–42

Hungria M, Campo RJ, Mendes IC et al (2006) Contribution of biological nitrogen fixation to the N nutrition of grain crops in the tropics: the success of soybean (*Glycine max* L. Merr.) in South America. In: Singh RP, Shankar N, Jaiwal PK (eds) Nitrogen nutrition and sustainable plant productivity. Studium Press/LCC, Houston, pp 43–93

Hungria M, Nogueira MA, Araujo RS (2013) Co-inoculation of soybeans and common beans with rhizobia and azospirilla: strategies to improve sustainability. Biol Fertil Soils 49:791–801

Hymowitz T and Harlan JR (1983) Introduction of soybean to North America by Bowen, Samuel in 1765. Econ Bot 37:371–379

Knobeloch L, Salna B, Hogan A et al (2000) Blue babies and nitrate-contaminated well water. Environ Health Perspect. 108:675–678.

Larue TA, Patterson TG (1981) How much nitrogen do legumes fix. Adv Agron 34:15–38

Leibovitch S, Migner P, Zhang F et al (2002) Evaluation of the effect of SoyaSignal technology on soybean yield [*Glycine max* (L.) Merr.] under field conditions over 6 years in eastern Canada and northern United States. J Agron Crop Sci 187(4):281–292

Liang Y, Cao Y, Tanaka K et al (2013) Nonlegumes respond to rhizobial Nod factors by suppressing the innate immune response. Science 341:1384–1387

Marks BB, Megías M, Nogueira MA et al (2013) Biotechnological potential of rhizobial metabolites to enhance the performance of *Bradyrhizobium japonicum* and *Azospirillum brasilense* inoculants with the soybean and maize crops. Appl Microbiol Biotechnol Express 3:21

Salvagiotti F, Cassman KG, Specht JE et al (2008) Nitrogen uptake, fixation and response to fertilizer N in soybeans. A review. Field Crops Res 108:1–13

Sanjuan J, Carlson RW, Spaink HP et al (1992) A 2-O-methylfucose moiety is present in the lipo-oligosaccharide nodulation signal of *Bradyrhizobium japonicum*. Proc Natl Acad Sci U S A 89:8789–8793

Souleimanov A, Prithiviraj B, Smith DL (2002) The major Nod factor of *Bradyrhizobium japonicum* promotes early growth of soybean and corn. J Exp Bot 53:1929–1934

Spaink HP, Aarts A, Stacey G et al (1992) Detection and separation of *Rhizobium* and *Bradyrhizobium* Nod metabolites using thin-layer chromatography. Mol Plant Microbe Interact 5:72–80

Thies JE, Singleton PW, Bohlool BB (1991) Influence of size of indigenous rhizobial populations on establishment and symbiotic performance of introduced rhizobia on field-grown legumes. Appl Environ Microbiol 57:19–28