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Series on Harmonization of Regulatory Oversight in Biotechnology No. 15

CONSENSUS DOCUMENT ON THE BIOLOGY OF GLYCINE MAX (L.) MERR. (SOYBEAN)

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OECD Environmental Health and Safety Publications

Series on Harmonization of Regulatory Oversight in Biotechnology

No. 15

Consensus Document on the Biology of *Glycine max* (L.) Merr. (Soybean)

Environment Directorate

Organisation for Economic Co-operation and Development

Paris 2000

About the OECD

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FOREWORD

The OECD's Working[1] Group on Harmonization of Regulatory Oversight in Biotechnology decided at its first session, in June 1995, to focus its work on the development of ***consensus documents*** which are mutually acceptable among Member countries. These consensus documents contain information for use during the regulatory assessment of a particular product. In the area of plant biosafety, consensus documents are being published on the biology of certain plant species, on selected traits that may be introduced into plant species, and on biosafety issues arising from certain general types of modifications made to plants.

This consensus document addresses the biology of *Glycine max* (L.) Merr. (Soybean). It contains a general description as a crop plant, taxonomy, centre of origin/diversity, identification methods, reproductive biology, crosses and ecology. However, its ecology to specific geographic regions is not presented, since this may vary from one geographic region to another. For the intraspecific and interspecific crosses, emphasis has been placed on detailing potential hybridisation between the cultivated species and their close relatives. In this document, only hybridisation events not requiring human intervention are considered, rather than listing all successful crosses, which would be very long and subject to frequent changes.

Canada served as lead country in the preparation of this document.

The Joint Meeting of the Chemicals Committee and the Working Party on Chemicals, Pesticides and Biotechnology has recommended that this document be made available to the public. It is published on the authority of the Secretary-General of the OECD.

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[1] In August 1998, following a decision by OECD Council to rationalise the names of Committees and Working Groups across the OECD, the name of the "Expert Group on Harmonization of Regulatory Oversight in Biotechnology" became the "Working Group on Harmonization of Regulatory Oversight in Biotechnology."

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Preamble

OECD Members are now commercialising and marketing agricultural and industrial products of modern biotechnology. They identified the need for harmonisation of regulatory approaches to the assessment of these products, in order to ensure safety, while avoiding trade barriers.

In 1993, **Commercialisation of Agricultural Products Derived through Modern Biotechnology** was instituted as a joint project of the OECD's Environment Policy Committee and Committee on Agriculture. The objective of this project is to assist countries in their regulatory oversight of agricultural products derived through modern biotechnology – specifically in their efforts to ensure safety, to make oversight policies more transparent and efficient, and to facilitate trade. The project is focused on the review of national policies, with respect to regulatory oversight, that will affect the movement of these products into the marketplace.

The first step in this project was to carry out a survey concentrating on national policies with regard to regulatory oversight of these products. Data requirements for products produced through modern biotechnology, and mechanisms for data assessment, were also surveyed. The results were published in *Commercialisation of Agricultural Products Derived through Modern Biotechnology: Survey Results* (OECD, 1995).

Subsequently, an OECD Workshop was held in June 1994 in Washington, D.C. with the aims of improving awareness and understanding of the various systems of regulatory oversight developed for agricultural products of biotechnology; identifying similarities and differences in various approaches; and identifying the most appropriate role for the OECD in further work towards harmonisation of these approaches. Approximately 80 experts in the areas of environmental biosafety, food safety and varietal seed certification, representing 16 OECD countries, eight non-member countries, the European Commission and several international organisations, participated in the Workshop. *Report of the OECD Workshop on the Commercialisation of Agricultural Products Derived through Modern Biotechnology* was also published by the OECD in 1995.

As a next step towards harmonisation, the Working Group on Harmonization of Regulatory Oversight of Biotechnology instituted the development of *consensus documents*, which are ***mutually acceptable*** among Member countries. The goal is to identify common elements in the safety assessment of a new plant variety developed through modern biotechnology, to encourage information sharing and prevent duplication effort among countries. These common elements fall into three general categories: the first being the biology of the host species, or crop: the second, the introduced gene and resulting gene product: and the third, environmental biosafety issues arising from certain general types of modification made to plants.

The safety issues identified in the consensus documents on the biology of specific crop plants are intended to address the potential for gene transfer within the crop plant species, and among related species, as well as the potential for weediness. They make no attempt to be definitive in this respect, however, as the many different environments in which the crop species may be grown are not considered individually.

This consensus document is a "snapshot" of current information that may be relevant in a regulatory risk assessment. It is meant to be useful not only to regulatory officials, as a general guide and reference source, but also to industry and others carrying out research.

In using this document and others related to the biology of crop plants, reference to two OECD publications which have appeared in recent years will prove particularly useful. *Traditional Crop Breeding Practices: An Historical Review to Serve as a Baseline for Assessing the Role of Modern Biotechnology* presents information concerning 17 different crop plants. It includes sections on phytosanitary considerations in the movement of germplasm and on current end uses of the crop plants. There is also a detailed section on current breeding practices. *Safety Considerations for Biotechnology: Scale-Up of Crop Plants* provides a background on plant breeding, discusses scale dependency effects, and identifies various safety issues related to the release of plants with "novel traits."

To ensure that scientific and technical developments are taken into account, OECD countries have agreed that consensus documents will be updated regularly. Additional areas relevant to the subject of each consensus document will be considered at the time of updating.

Users of this document are therefore invited to provide the OECD with new scientific and technical information, and to make proposals for additional areas to be considered. ***There is a short, pre-addressed questionnaire for that purpose at the end of this document. The completed questionnaire (or a photocopy) should be returned to the OECD's Environmental Health and Safety Division at the address shown.***

Section I - General Description Including Taxonomy and Morphology, and Use as a Crop Plant

Cultivated soybean, *Glycine max* (L.) Merr., is a diploidized tetraploid (2n=40), in the family Leguminosae, the subfamily Papilionoideae, the tribe Phaseoleae, the genus *Glycine* Willd. and the subgenus *Soja* (Moench). It is an erect, bushy herbaceous annual that can reach a height of 1.5 metres. Three types of growth habit can be found amongst soybean cultivars: determinate, semi-determinate and indeterminate (Bernard and Weiss, 1973). Determinate growth is characterised by the cessation of vegetative activity of the terminal bud when it becomes an inflorescence at both axillary and terminal racemes. Determinate genotypes are primarily grown in the southern United States (Maturity Groups V to X). Indeterminate genotypes continue vegetative activity throughout the flowering period and are grown primarily in central and northern regions of North America (Maturity Groups 000 to IV). Semi-determinate types have indeterminate stems that terminate vegetative growth abruptly after the flowering period. None of the soybean varieties are frost tolerant, and they do not survive freezing winter conditions.

The primary leaves are unifoliate, opposite and ovate, the secondary leaves are trifoliolate and alternate, and compound leaves with four or more leaflets are occasionally present. The nodulated root system consists of a taproot from which emerges a lateral root system. The plants of most cultivars are covered with fine trichomes, but glabrous types also exist. The papilionaceous flower consists of a tubular calyx of five sepals, a corolla of five petals (one banner, two wings and two keels), one pistil and nine fused stamens with a single separate posterior stamen. The stamens form a ring at the base of the stigma and elongate one day before pollination, at which time the elevated anthers form a ring around the stigma. The pod is straight or slightly curved, varies in length from two to seven centimetres, and consists of two halves of a single carpel which are joined by a dorsal and ventral suture. The shape of the seed, usually oval, can vary amongst cultivars from almost spherical to elongate and flattened.

Soybean is grown as a commercial crop in over 35 countries. The major producers of soybeans are the United States, China, Democratic People’s Republic of Korea and Republic of Korea, Argentina and Brazil. Soybean is grown primarily for the production of seed, has a multitude of uses in the food and industrial sectors, and represents one of the major sources of edible vegetable oil and of proteins for livestock feed use.

A major food use in North America and Europe is as purified oil, utilised in margarines, shortenings and cooking and salad oils. It is also used in various food products, including tofu, soya sauce, simulated milk and meat products. Soybean meal is used as a supplement in feed rations for livestock. Industrial use of soybeans ranges from the production of yeasts and antibodies to the manufacture of soaps and disinfectants.

Soybean is commonly considered one of the oldest cultivated crops, native to North and Central China (Hymowitz, 1970). The first recording of soybeans was in a series of books known as Pen Ts'ao Kong Mu written by the emperor Sheng Nung in the year 2838 B.C., in which the various plants of China are described. Historical and geographical evidence suggests that soybeans were first domesticated in the eastern half of China between the 17th and 11th century B.C. (Hymowitz, 1970). Soybeans were first introduced into the United States, now a major producer, in 1765 (Hymowitz and Harlan, 1983).

Section II - Agronomic Practices

Soybean is a quantitative short day plant and hence flowers more quickly under short days (Garner and Allard, 1920). As a result, photoperiodism and temperature response is important in determining areas of cultivar adaptation. Soybean cultivars are identified based on bands of adaptation that run east-west, determined by latitude and day length. In North America, there are thirteen maturity groups (MG), from MG 000 in the north (45° latitude) to MG X near the equator. Within each maturity group, cultivars are described as early, medium or late maturing.

The seed will germinate when the soil temperature reaches 10°C and will emerge in a 5-7 day period under favourable conditions. In new areas of soybean production an inoculation with *Bradyrhizobium japonicum* is necessary, for optimum efficiency of the nodulated root system. Soybeans do not yield well on acid soils and the addition of limestone may be required. Soybeans are often rotated with such crops as corn, winter wheat, spring cereals, and dry beans.

Section III - Centres of Origin of the Species

Glycine max belongs to the subgenus *Soja*, which also contains *G. soja* and *G. gracilis*. *Glycine soja*, a wild species of soybean, grows in fields, hedgerows, roadsides and riverbanks in many Asian countries. Wild soybean species are endemic in China, Korea, Japan, Taiwan and the former USSR, but do not exist naturally in North America. Cytological, morphological and molecular evidence suggest that *G. soja* is the ancestor of *G. max*. *Glycine gracilis* is considered to be a weedy or semi-wild form of *G. max*, with some phenotypic characteristics intermediate to those of *G. max* and *G. soja*. *Glycine gracilis* may be an intermediate in the speciation of *G. max* from *G. soja* (Fekuda, 1933) or a hybrid between *G. soja* and *G. max* (Hymowitz, 1970).

Section IV - Reproductive Biology

Soybean is considered a self-pollinated species, propogated commercially by seed. Artificial hybridisation is used for cultivar breeding.

The soybean flower stigma is receptive to pollen approximately 24 hours before anthesis and remains receptive 48 hours after anthesis. The anthers mature in the bud and directly pollinate the stigma of the same flower. As a result, soybeans exhibit a high percentage of self-fertilisation, and cross pollination is usually less than one percent (Caviness, 1966).

A soybean plant can produce as many as 400 pods, with two to twenty pods at a single node. Each pod contains one to five seeds. Neither the seedpod, nor the seed, has morphological characteristics that would encourage animal transportation.

Section V - Cultivated *Glycine max* as a Volunteer Weed

Cultivated soybean seed rarely displays any dormancy characteristics and only under certain environmental conditions grows as a volunteer in the year following cultivation. If this should occur, volunteers do not compete well with the succeeding crop, and can easily be controlled mechanically or chemically. The soybean plant is not weedy in character. In North America, *Glycine max* is not found outside of cultivation. In managed ecosystems, soybean does not effectively compete with other cultivated plants or primary colonisers.

Section VI - Crosses

A. Inter-Species/Genus

In considering the potential environmental impact following the unconfined release of genetically modified *Glycine max*, it is important to have an understanding of the possible development of hybrids through interspecific and intergeneric crosses with related species. The development of hybrids could result in the introgression of the novel traits into these related species and result in:

– the related species becoming more weedy

– the introduction of a novel trait, with potential for ecosystem disruption, into related species.

For a trait to become incorporated into a species genome, recurrent backcrossing of plants of that species by hybrid intermediaries, and survival and fertility of the resulting offspring, is necessary.

The subgenus *Soja*, to which *G. max* belongs, also includes *G. soja* Sieb. and Zucc. (2n=40) and *G. gracilis* Skvortz. (2n=40), wild and semi-wild annual soybean relatives from Asia. *Glycine soja* (2n=40) is a wild viny annual with small and narrow trifoliate leaves, purple flowers and small round brown-black seeds. It grows wild in Korea, Taiwan, Japan, Yangtze Valley, N.E. China and areas around the border of the former USSR. *Glycine gracilis*, an intermediate in form between *G. soja* and *G. max*, has been observed in Northeast China (Skvortzow, 1927). Interspecific, fertile hybrids between *G. max*. and *G. soja* (Sieb and Zucc.) (Ahmad et al., 1977; Hadley and Hymowitz, 1973; Broich, 1978), and between *G. max* and *G. gracilis* (Karasawa, 1952) have been easily obtained.

In addition to the subgenus *Soja*, the genus *Glycine* contains the subgenus *Glycine*. The subgenus *Glycine* consists of twelve wild perennial species, including *G. clandestina* Wendl., *G. falcata* Benth, *G. latifolia* Benth., *G. latrobeana* Meissn. Benth., *G. canescens* F.J. Herm., *G. tabacina* Labill. Benth., and *G. tomentella* Hayata. These species are indigenous to Australia, South Pacific Islands, China, Papua New Guinea, Philippines, and Taiwan (Hymowitz and Newell, 1981; Hermann, 1962; Newell and Hymowitz, 1978; Grant, 1984; Tindale, 1984,1986). Hybrids between diploid perennial *Glycine* species show normal meiosis and are fertile.

Early attempts to hybridise annual (subgenus *Soja*) and perennial (subgenus *Glycine*) species were unsuccessful. Although pod development was initiated, these eventually aborted and abscised (Palmer, 1965; Hood and Allen, 1980; Ladizinsky et al., 1979). Intersubgeneric hybrids were later obtained in vitro through embryo rescue, between *G. max* and *G. clandestina* Wendl; *G. max* and *G. tomentella* Hayata (Singh and Hymowitz, 1985; Singh et al., 1987); and *G. max* and *G. canescens*, using transplanted endosperm as a nurse layer (Broué et al., 1982). In all cases, the progeny of such intersubgeneric hybrids was sterile and obtained with great difficulty.

B. Introgression into Relatives

Soybean can only cross with other members of *Glycine* subgenus *Soja*. The potential for such gene flow is limited by geographic isolation. Wild soybean species are endemic in China, Korea, Japan, Taiwan and the former USSR. These species are not naturalised in North America, and although they could occasionally be grown in research plots, there are no reports of their escape from such plots to unmanaged habitats.

C. Interactions with Other Organisms

The table in the Appendix is intended as an identification guide for categories of organisms, which interact with *Glycine max*. This table, representative of North America, is intended to serve as an example only. Environmental safety assessors should, on a country-by-country basis, draw up their own lists as a guide for assessing potential effects of the release of genetically modified plants on interacting organisms in their country.

The intention is not to require comparison data between a plant with novel traits and its *G. max* counterpart(s) for all interactions. Depending on the novel traits, applicants might decide to submit data for only some of the interactions. Sound scientific rationale will be required to justify the decision that data would be irrelevant for the remaining interactions. For example, the applicant might choose not to provide data on the weediness potential of a plant with novel traits if it can be clearly shown that the novel trait will not affect reproductive or survival characteristics of *G. max*, either directly or indirectly. Some of the life forms are listed as categories (i.e., pollinators, mychorrhizal fungi, animal browsers, birds, soil microbes, and soil insects). When, because of the novel traits, a concern is perceived for these specific categories, applicants will be required to provide detailed information on interactions with indicator species in each category. Where the impact of a plant with novel traits on another life form (target or non-target organism) is significant, secondary effects may need to be considered.

This section will be revised to include relevant new data as they become available.

Section VII - Summary of Ecology of *Glycine max*

1. *Glycine max* (L.) Merr., the cultivated soybean, is a summer annual herb that has never been found in the wild (Hymonitz, 1970). This domesticate is in fact extremely variable, due primarily to the development of soybean “land races” in East Asia. The subgenus *Soja* contains, in addition to *G. max* and *G. soja*, the form known as *G. gracils*, a form morphologically intermediate between the two. This is a semi-cultivated or weedy form, and is known only from Northeast China.

2. *Glycine soja*, considered the ancestor of cultivated soybean, is an annual procumbent or slender twiner that is distributed throughout China, the adjacent areas of the former USSR, Korea, Japan and Taiwan. It grows in fields and hedgerows, along roadsides and riverbanks.

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Appendix

Examples of Potential Interactions of *G. max* with Other Life Forms During its Life Cycle

Other life forms Common Name	Interaction with *G. max* Pathogen; Symbiont or Beneficial Organism; Consumer; Gene Transfer
Brown spot (*Septoria glycines*)	Pathogen
Downy mildew (*Peronospora trifoliorum var. manshurica*)	Pathogen
Brown stem rot (*Phialophora gregata* or *Acremonium strictum*)	Pathogen
Phytophthora root and stalk rot (*Phytophthora megasperma*)	Pathogen
Stem canker (*Diaporthe phaseolorum var caulivora*)	Pathogen
Rhizoctonia stem and root rot (*Rhizoctonia solani*)	Pathogen
Pythium root rot (*Pythium spp*.)	Pathogen
Fusarium wilt, blight, and root rot (*Fusarium spp*.)	Pathogen
Sclerotinia stem rot (*Sclerotinia sclerotiorum*)	Pathogen
Pod and stem blight (*Diaporthe phaseolorum var. sojae*)	Pathogen
Bacterial Blight (*Pseudomonas syringae*)	Pathogen
Soybean mosaic virus (SMV)	Pathogen
Anthracnose (*Colletotrichum truncatum*)	Pathogen
Purple seed stain (*Cercospora kikuchii*)	Pathogen
Powdery mildew (*Microsphaera diffusa*)	Pathogen
Root knot (*Meloidogyne spp*.)	Pathogen
Spider mite (*Acari: Terranychidae*)	Consumer
Soybean cyst nematode (*Heterodera glycines*)	Consumer
Soybean looper, white fly (*Lepidopterans*)	Consumer
Soil insects	Consumer
Birds	Consumer
Animal browsers	Consumer
Pollinators	Symbiont or Beneficial Organism; Consumer
Mychorrhizal fungi	Symbiont or Beneficial Organism
Soil microbes	Symbiont or Beneficial Organism
Earthworms	Gene Transfer
Other *G. max*	Symbiont or Beneficial Organism
Others	---

QUESTIONNAIRE TO RETURN TO THE OECD

This is one of a series of OECD Consensus Documents that provide information for use during regulatory assessment of particular micro-organisms, or plants, developed through modern biotechnology. The Consensus Documents have been produced with the intention that they will be updated regularly to reflect scientific and technical developments.

Users of Consensus Documents are invited to submit relevant new scientific and technical information, and to suggest additional related areas that might be considered in the future.

The questionnaire is already addressed (see reverse). **Please mail or fax this page (or a copy) to the OECD, or send the requested information by E-mail:**

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