DWR
Technologies and Techniques

Compiled and edited by
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ICAR - Directorate of Weed Research, Jabalpur
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Initial Date of Certification - 21th December 2016 *Current Date of Certification 21th December 2016*

Date of Expiry :
20 December 2019

1st Surveillance Audit
November 2017
2nd Surveillance Audit
November 2018

Certification Number :
GCS/QMS/0351

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Technical bulletin no. 14

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Citation
ICAR-DWR Technologies and Techniques-2017. Directorate of Weed Research, Jabalpur, 143 p.

Published by
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Published in
November, 2017

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Preamble

Considering the growing infestation of weeds in cropped and non-cropped lands, the Indian Council of Agricultural Research decided to establish the National Research Centre for Weed Science, which came into existence on 22nd April, 1989 with the joining of its first Director, Late Dr. V.M. Bhan. This centre was further upgraded as Directorate of Weed Science Research on 23rd January, 2009. This is a unique institute in the National Agricultural Research System, which is probably the only one of its own kind in the whole world dealing exclusively with weed research. Besides, training, coordination, consultancy and collaborative programmes on weed management are also undertaken with various stakeholders.

Over the last 28 years, the institute has played a significant role in conducting the weed survey and surveillance, development of weed management technologies for diversified cropping systems, herbicide resistance in weeds, biology and management of problem weeds in cropped and non-cropped areas, and environmental impact of herbicides. Adoption of these technologies has been promoted in large areas through on-farm research and demonstrations, which has raised agricultural productivity and livelihood security of the farmers. Training and awareness, and consultancy programmes organized by the Directorate have been found highly beneficial and appreciated by various stakeholders. All these activities have been further geared up to address the emerging challenges in weed management including threats posed by climate change, invasive weeds, herbicide resistance, herbicide hazards, and safety concerns about herbicide tolerant crops.

Since its inception, the Directorate has contributed significantly by developing technologies on weed management and their dissemination to the farming community. Weed management technologies on crops, non-crop situations, problematic weeds, biological control of *Parthenium* and water hyacinth, phytoremediation of pollutants by weeds, and weed utilization have been developed and demonstrated at farmer's fields. Several techniques for undertaking specific studies on weed physiology and biology, climate change and herbicide residues have been developed. Techniques for effective dissemination of technologies to the farming community have also been developed. In the present compilation, all such technologies and techniques generated over the years have been compiled. It is hoped that it will be useful to our stakeholders to get acquainted with these technologies and techniques for better weed management. Any suggestions for future improvement and refinement of the technologies of the Directorate will be appreciated.

Place : Jabalpur

Date : 25 November, 2017

(P.K. Singh)

Director

1.0 Weed management in crops

1.1 Chemical weed control in wheat

The problem: The presence of weeds within the crop may adversely affect production in a number of ways. Weeds compete with crop species for space, water, nutrients and light and ultimately reduce crop yield. As a result, the growth of the crop is adversely suppressed, and the crop productivity is reduced significantly. In general, season long competition from major weeds culminates in yield reduction to an extent of 15-40% in wheat depending upon intensity and type of weed flora under different cropping systems. However, continuous use of the same herbicide may result in development of resistant biotypes. This calls for rotational use of other competitive herbicides for weed management to avoid shift in the weed flora.

The technology: Application of sulfosulfuron + metsulfuron at 30 + 2 g/ha; mesosulfuron + iodosulfuron at 12 + 2.4 g/ha; clodinafop + metsulfuron at 60 + 4 g/ha; sulfosufuron 25 g/ha; carfentrazon 20 g/ha and metribuzin 175 g/ha as post-emergence are most effective to control grassy as well as broad-leaved weeds in wheat whereas, pinoxaden 40 g/ha as post-emergence is most effective to control grasses. This technology increases the grain production, improves quality of grain and results in higher income.

Cost of technology: For the chemical weed control in wheat through above herbicides the cost requires ₹2000-3000/ha.

The benefits: The new technology for weed management in wheat lowers the weed competition from the beginning and saves the loss of nutrients to an extent of 30-50%. The overall net return with the adoption this technology for weed management in wheat is to the tune of ₹ 10,000-12,000/ha. The technology also helps to overcome the labour problem during peak weeding season. This technology relieves pressure on human labour for weeding, avoids loss of nutrients, more cost effective and improves yield.

Effects of weed management treatments in wheat

1.2 Weed management in zero-till wheat

The problem: Wheat is conventionally grown following intensive tillage operations. This results in preparation of good seed-bed and mixing of fertilizers and weeds / stubbles of previous crop, if any, leading to good crop stand and growth as well as early weed control. However, there is greater use of energy, increased cost of cultivation, deterioration of soil physico-chemical and biological properties. To counter these adverse effects, zero-tillage technology has been advocated for growing wheat, but there is possibility of greater infestation of weeds compared with conventional tillage. Besides, the existing weeds of the previous crop may provide greater early crop-weed competition. Unchecked weed growth causes yield losses up to 35-45%.

The technology: Weeds can be managed effectively in zero-till wheat through an integrated approach. Apply paraquat 0.5 kg/ha before sowing to kill the existing foliage of the weeds growing after the harvest of previous crop of rice, maize, soybean, pigeonpea, cotton, blackgram etc. After 2-3 days, sow wheat with specially-designed zero-till seed-cum-fertilizer drill having knife type tynes at 4-5 cm depth using 120 kg seed/ha. Full-dose of P and K along with 50% N should be placed basally after proper calibration of the machine. Apply any of the following herbicides depending on the infestation of the weed flora:

Herbicide	Dose (g/ha)	Time of application	Remarks
Clodinafop propargyl	60	25-30 DAS	Controls annual grasses, especially *Avena* spp., and Phalaris minor
Metribuzin	175-200	30-35 DAS	Controls annual grasses and broad-leaved weeds
Sufosulfuron	25	25-30 DAS	Controls annual broad-leaved weeds and grasses
Sufosulfuron + metsulfuron	30 + 2	25-30 DAS	Controls annual grasses, broad-leaved weeds and sedges
Iodosulfuron + mesosulfuron	12 + 2.4	20-25 DAS	Controls annual grasses, broad-leaved weeds and sedges
Clodinafop + metsulfuron	60 + 4	20-25 DAS	Controls annual grasses, broad-leaved weeds and sedges
Isoproturon + metsulfuron	1000 + 4	20-25 DAS	Controls annual grasses and broad-leaved weeds

Zero-till seed-cum-fertilizer drill with knife-type tynes for sowing of wheat

Good initial crop stand of zero-till sown wheat

Conventional-till wheat

Zero-till wheat

The benefits: Results have shown that the productivity of zero-tilled wheat is almost the same as of conventionally-tilled wheat if sown at the same time. The crop of zero-till wheat can be sown timely (7-10 days early) which can result in higher yield (10-20%) especially when sowing under conventional tillage gets delayed due to late harvesting of *kharif* season crops. The cost of production is reduced by Rs. 2500/- per ha by saving 3-4 ploughing operations in conventional tillage. Infestation of *Phalaris minor* is also less in zero-till crop. There is improvement in soil physico-chemical and biological properties due to little soil disturbance. There may also be less use of irrigation water (5-10%) as the deep percolation losses of water are checked. The practice is eco-friendly, results in less emission of GHGs and thus helps in mitigating adverse effects of climate change.

Precautions: There is required greater care in sowing, crop management and precise conditions in the field for success of this technology. It is essential to ensure proper levelling of the field preferably through laser land leveler so as to place the seed and fertilizer at proper depth. Ensure the seed is properly covered with soil during sowing operation to check damage by ants, birds etc. Seed may be treated with chloropyriphos to check termite damage. There should be adequate moisture in the soil at sowing or else irrigation should be applied after sowing. It is preferable to use 15-20% more seed and fertilizer to compensate for any possible loss in crop stand and growth due to poor seed-soil contact, less germination, damage by birds etc. Recommended herbicide should be applied timely. The technology is suitable for most soil types when the previous crop has been harvested manually and the crop residues except anchored stubbles or fallen leaf litter have been removed from the field.

1.3 Weed management in organically grown wheat

The problem: Wheat crop is severely infested with weeds reducing the grain yield considerably. The weed problems become serious, and their management costly under organic cultivation. Weed management practices other than herbicide-use like hand weeding, mulching, stale- seed bed, intercropping may be useful in management of weeds.

The technology: Apply well rotten FYM 10-20 t/ha in the field. Sow wheat at a row spacing of 30 cm. In between the wheat rows, broadcast berseem seeds at a rate of 20-25 kg/ha. Irrigate the field lightly. The intercrop of berseem will suppress the growth of weeds. Two to three cuts of berseem can be taken during the crop duration of wheat. This will also cut down any weed growth. After the harvest of wheat, apply 1-2 irrigations to the berseem crop which will later come to flower and seed set.

The benefits: This practice may give higher wheat equivalent yields. It is good for crop diversification, fodder availability in land holdings of small farmer . Good quality berseem seed approx. 80-100 kg/ha can also be obtained. Being organic, wheat grain will fetch higher price (30-40%) in market. A benefit : cost ratio of 3.36 could be obtained with this technology as compared to 3.05 obtained under recommended practice.

Precautions: The berseem seeds should be evenly broadcast in between the wheat rows. In berseem, if *Cuscuta* appears then it should be manually removed at the early stage of infestation.

Intercropping of berseem in organic wheat crop for weed suppression

1.4 Weed control in direct-seeded rice (DSR)

The problem: Weeds are a major concern for higher productivity in DSR. Effective weed management in DSR depends on several factors, including the timeliness of the control operations during the early crop growth stages and, in some cases, good control in preceding crops. In transplanted rice, weed control is conducted just before transplanting and the rice has a significant size and competitive advantage over subsequently emerging weeds. The main reasons for high weed pressure in DSR because of simultaneous emergence of rice & weeds, and absence of a weed-suppressive effect of standing water and yield losses to weed competition can approach even up to 100% in poorly managed fields. Integrated approaches to weed management combine multiple tactics and knowledge of site-specific field conditions are essential to increase the efficacy and sustainability of weed management. Judicious use of herbicides and crop competitiveness varieties are the key factors to manage weeds in DSR.

The technology:

Cultural methods

a. Stale seed bed: In this technique, weed seed germination is encouraged by applying light irrigation and then emerged seedlings are killed using a non-selective herbicide before crop sowing. It has been observed that this technique can reduce weed population by 53%.

b. Good crop establishment: Spatially uniform establishment of healthy, vigorous rice seedlings increases crop competitive ability and suppresses weed growth.

c. Brown manuring: This practice involves seeding of rice and *Sesbania* crops together and knockdown the *Sesbania* crop 25- 30 days after sowing with 2, 4-D ester at 0.40- 0.50 kg/ha.

Chemical weed control: The right herbicide for use in DSR depends on the weed flora present in a given field; individual herbicides have strength and also weakness, e.g. bispyribac-sodium is very good on grasses but can't control *Leptochloa*. Rotational use of herbicides with different modes of actions is also desirable to check the possible development of herbicide resistant weed biotypes. Timely use of newly-released effective herbicides in DSR helps in better weed management and results in increase in yield besides increasing profitability. Sequential applications of a pre-emergence herbicide (e.g. pendimethalin 1000 g/ha, pyrazosulfuron 20 g/ha) followed by post-emergence herbicide [e.g., bispyribac-sodium (25 g/ha), penoxsulam (25 g/ha), azimsulfuron (30 g/ha), fenoxaprop (with safener) (60 g/ha), metsulfuron (4 g/ha) + fenoxaprop (60 g/ha)] can provide effective weed control in DSR.


Untreated field of rice


Bispyribac field of rice

Cost of technology : The cost of technology varies with the king of weed management approaches. Among herbicides post-emergence application of bispyribac-sodium requires higher cost than the others. In general the cost of the technology varies from ₹ 2500 - 3500/ha.

The benefits : Improved weed management technologies result in better quality of the produce and higher grain yield. The net income of ₹ 20,000 - 25,000/ha over the traditional practice of weed management may be obtained.

1.5 Integration of *Sesbania* for weed management in direct - seeded rice

The problem: Heavy weed infestation is one of the major constraints limiting productivity of direct - seeded rice. In certain cases the situation becomes worse and results in total crop failure. The traditional methods of hand weeding, because of heavy weed infestation, require comparatively huge man-power which is becoming scarce and costly day by day. Chemical control also sometimes becomes ineffective because of moisture fluctuations in the rice ecologies. Therefore, there is a need to integrate all the possible methods of weed control to obtain optimum yield of direct-seeded rice.

The technology: Sow rice in rows 25 cm apart. In between the rice rows, broadcast seeds of *Sesbania* (*dhaincha*). *Sesbania* will suppress the growth of weeds in the rice crop. After about 30 days of crop growth, spray 2, 4-D at 0.5 kg/ha in the field. This will knock down the *Sesbania* plants and other broadleaf weeds. Further, weeds growth can be checked by manual weeding with considerably less man power. *Sesbania* after decomposition will also add organic matter and nitrogen (20-30 kg/ha) in the soil.

Benefits: The technology effectively controls weeds besides adding organic matter and nitrogen to the soil.

Precautions: The *Sesbania* intercrop should not be continued after 30 days of growth otherwise it will start suppressing rice crop.

Direct- seeded rice (*Sesbania* + 2, 4-D *fb* 1 hand weeding)

Direct-seeded rice (unweeded)

1.6 Chemical weed control in maize

The problem: Maize is one of the most sensitive crop to weed competition during its early growth period usually up to 60 days after sowing, because the growth of maize plants in the first week is very slow and during this period weeds establish rapidly and become competitive. Maximum weed competition in maize occurs during the period of 2 to 6 weeks after sowing. This suggests the importance of maintaining the field weed-free during the critical period of crop weed competition. Incessant rains and dry spells prevent weed control on time by traditional methods. During peak periods of weeding in maize, labour becomes very scarce because of the increasing wage resulting from the migration of rural labour to the cities. The use of herbicide is becoming a necessity to mange weeds in such situations.

The technology: The herbicides *viz*. atrazine 1.0 -1.5 kg/ha; alachlor 2.0-2.5 kg/ha; pendimethalin 1.0-1.5 kg/ha; tank-mix atrazine + pendimethalin 0.75 + 0.75 kg/ha; atrazine + terbutryn 0.53 + 0.57 kg/ha as pre-emergence and atrazine + 2,4-D amine 1.1 + 0.48 kg/ha as early post-emergence. After application of herbicides the soil should not be disturbed. These herbicides are quite effective and show broad-spectrum activity against grasses and broad-leaved weeds, but not effective if applied in dry soil. Sufficient moisture is a pre-requisite for greater herbicidal efficacy. Moreover, integration of mechanical approach at 30 days after sowing with chemical ones enhanced the degree of weed control than the sole chemical approach.

Cost of technology: Lower cost requires for sole chemical method i.e. ₹1200/ha, but integrated approach needs higher cost, upto ₹5000/ha.

The benefits: Weeds and shortage of labour for removal of weeds are two of the most important production constraints in maize causing yield losses upto 40-60%. With the use of herbicides the losses could be minimized and better yield can be achieved with high benefit: cost ratio. By adopting integrated weed management approach, the grain yield of maize can be enhanced up to 60%.


Weed infested field of maize crop


Treated field of maize with atrazine + pendimethalin

1.7 Intercropping cowpea for weed management in maize

The problem: Maize is an important cereal crop in India. The crop grown in *kharif* season suffers abundant growth of wide range of weeds causing severe yield reduction which may vary depending upon the type of weed flora and their population. Yield losses in maize due to weeds in the range of 32 to 42%, and up to as high as 84.3% under season long weed competition in maize have been reported. No single method of weed control can be effective; hence integration of cultural and chemical methods is required to obtain desired weed control. The wide inter row space in maize crop permits growing of short duration intercrops especially of leguminous nature for weed suppression by smothering effect. Their role is likely to be more pronounced when integrated with herbicides and fertilizer nitrogen management.

The technology: Intercrop cowpea for grain or fodder purpose for suppressing the weed growth in maize. In fields, having heavy weed infestation, apply pendimethalin 1.0 kg/ha as pre-emergence followed by one hand weeding at 30 DAS. In this case, nitrogen dose of 50 kg/ha in maize would be sufficient to obtain optimum grain yields.

The benefits: Cowpea can be intercropped with maize at 1:1 row proportion (additive series) which suppressed the weeds by 32-36% with additional yield advantage (maize equivalent yield) from 2969 kg/ha (in sole maize) to 4064 kg/ha (in intercropped cowpea). These helped to achieved B:C ratio from 1.26 to 1.54.

Precautions: The variety of cowpea should be of short stature

Maize + cowpea; pendimethalin 1.0 kg/ha; 50 kg N/ha

1.8 Weed management in chickpea, pea, pigeonpea, greengram and blackgram

Problem: Pulses are major source of protein for vegetarian population in India, and provide significant nutritional and health benefits. Pulses are grown mainly under rainfed situation and weed infestation is the major constraint for increasing productivity of pulses. In general, the loss in crop yield is more in *kharif* pulses (30-40%) as compared with *rabi* pulses (20-25%).

The technology: Due to slow crop growth during initial stages weeds supersede the crop plant. In pulses weeds are generally managed through manual weeding or hoeing, but due to less availability of labour timely inter culture operation becomes a very difficult task. Weed management in pulses requires an integrated approach that utilizes effective preventive, mechanical and chemical methods in a mutually supportive manner into the crop production system.

A. Preventive measures: The entry of weeds from infested areas can be minimized through the following approaches: use of weed free seed, cleaning of tillage and sowing machinery/implements, and use of well decomposed FYM/compost/vermicompost etc.

B. Mechanical measure: Weeding by mechanical methods include the use of hand chisel, hand hoe and wheel hoe etc. during early period of crop growth effectively controlled weeds in pulse crops.

C. Chemical method: In situations where climate does not permit to use mechanical measures, the control of weeds through herbicides have been found more effective and economical also. Among pre-emergence herbicides pendimethalin (750-1000 g/ha) effectively control grassy and broad-leaved weeds as in all pulse crops. Imazethapyr (70-100 g/ha) at 25 days after sowing effectively controlled grassy and broad-leaved weeds in most of the pulses except chickpea, and to control grassy weeds clodinafop-p-propargyl (60 g/ha) and quizalofop-p-eyhyl (50 g/ha) is very effective as post-emergence herbicide. Integration of mechanical and chemical approaches gave higher return as compared to single approach.

Cost of technology: ₹ 2000-3000/ha.

The benefits: Improved weed management technologies result in 30-40% higher yield and better quality of the produce as compared to weedy situation.

Mechanical weed management in chickpea

1.9 Weed management in oilseed crops

Problem: Oilseed crops play vital role in the national economy. Oilseed crops are mostly cultivated in different parts of the country for getting edible as well as non-edible oils. National productivity of oilseed crops is quite low as against their yield potential. Competition by weeds at initial stages, and improper and untimely weed management are the major reasons for low productivity of oilseed crops. Yield losses due to weeds may vary from 20% - 60% depending on nature of crop and magnitude of weed infestation.

The technology: For weed management in oilseed crops, pre-emergence herbicides are available but very limited option for post-emergence herbicides. Pendimethalin (750-1000 g/ha) as a pre-emergence effectively control grassy as well as broad-leaved weeds, in all oilseed crops, and to control grassy weeds clodinafop-p-propargyl (60 g/ha) and quizalofop-p-eyhyl (50 g/ha) is very effective as post-emergence herbicide. In groundnut, imazethapyr (70-100 g/ha) at 25 days after sowing effectively controlled grassy and broad-leaved weeds. For better management and higher sustainability, integrated weed management approach is required. Integration of chemical and mechanical/manual weeding is very effective in almost all oilseed crops in controlling the diverse weed flora. Pendimethalin at 750-1000 g/ha followed by one hand weeding/hoeing at 30-45 DAS is very effective in controlling weeds in rapeseed and mustard, sunflower, groundnut and other oilseed crops.

Cost of technology: ₹ 2000-3000/ha. The cost of weed management practices varies greatly with the methods of weed management. In general ₹ 1500 - 2000/ha is required for chemical approach and ₹ 4500-5000/ha for integrated methods.

The benefits : Improved weed management technologies result in 20-40% higher yield and better quality of the produce as compared to weedy situation. The adoption of integrated method enhanced the seed yield of oilseed crops by 20-30% over sole chemical.

Mechanical weed management in mustard

1.10 Improved weed control in soybean

The problem: Weeds are vigorous competitors with soybean. Weeds usually germinate and emerge with the soybean. Therefore, soybean cannot get ahead of the weeds. Soybean is relatively short and susceptible to shading from taller weeds. Uncontrolled weeds not only reduce soybean yields through their competition for light, nutrients and moisture, but they can also severely reduce harvest efficiency. Since soybean is especially sensitive to moisture deficiencies, a few large weeds can severely reduce yields. Nearly complete weed control must be accomplished within three to four weeks after emergence of the soybean in order to avoid yield losses due to early emerging weeds. Before implementing a weed management plan for soybean, several factors need to be considered including weed species, crop rotation and cost effectivity. The most effective weed management programs in soybean use a combination of cultural, mechanical, and chemical control strategies. Herbicides are the cheapest and often the most efficient weed management tool but it is most successful and economical when all the tools for weed control are utilized in an integrated program.

The technology: The most effective weed management system in soybeans use a combination of cultural, mechanical, and chemical control strategies. Cultural practices include such factors as planting date, seed rate, and row spacing. Cultural practices improve weed control by enhancing the competitive ability of the soybean. Mechanical practices, such as use of hand hoe, cycle hoe, power weeder *etc.* for controlling weeds between rows, are a non-chemical approach for weed management. A multitude of herbicides are used in soybean and can be applied pre-plant incorporated, pre-emergence, post-emergence and post-directed. Application of pedimethalin 750 g/ha oxadiazon 500 g/ha as pre-emergence for controlling many annual and broad-leaved weeds. Imazethapyr 100 g/ha, imazethapyr 14 g/ha + imazemox 14 g/ha, imazethapyr 50 g/ha + pendimethalin 750 g/ha, sodium acifluorfen 16.5 g/ha + clodinafop-propargyl 8 g/ha, chlorimuron 6 g/ha, fenoxaprop 80 g/ha, quizalofop 40 g/ha, as post–emergence are most effective to control grassy as well as broad-leaved weeds in soybean. This technology increases the grain production, improves quality of grain, and results higher income.

Cost of technology: ₹2500-3500/ha

The benefits: The use of herbicides lowers the weed competition at early stage of the crop and saves 40-60% yield loss. The technology also helps to overcome the labour problem during weeding season. This technology relieves pressure on human labour for weeding, avoids loss of nutrients, cost effective and improves yield.

Soybean treated with imazethapyr

Soybean infested with *Euphorbia geniculata*

1.11 Chemical weed control in cotton

The problem: In cotton, weeds cause several direct and/or indirect negative impacts, such as reducing fiber quality, reducing crop yield, increasing production costs, reducing irrigation efficiency, and serving as hosts and habitats for insect pests, disease-causing pathogens, nematodes and rodents. Weeds can directly hinder cotton growth by competing for available resources and in some cases, by releasing allelopathic chemicals. However, the degree of damage from weed competition is related to the weed species, weed densities, and the duration of crop-weed competition. During the early stages of cotton development i.e. first 8 to 10 weeks after planting, weeds can out-compete cotton seedlings and cause serious damage by reducing the plant vigor. This often results in a reduction in formation of squares and bolls however, when the crop has become well-established, the cotton plants will be competitive against weeds and the direct negative impact of the weeds on the crop will be minimal. Therefore, for effective weed management in cotton, growers should concentrate their efforts on weed management in the early part of the growing season.

The technology: Application of pendimethalin 750 g/ha, oxadiazon 500 g/ha as pre-emergence is highly effective for controlling many annual and broad-leaved weeds. Application of pyrithiobac-sodium 75 g/ha, quizalofop 50 g/ha and or directed spray of glyphosate 1000 g/ha as post-emergence are most effective to control grassy as well as broad-leaved weeds in cotton. This technology increases the seed cotton production, improves quality of cotton, and also increases farm income.

Cost of technology: The cost of weed management practice with different pre & post-emergence herbicides may vary from ₹ 2500- 4000/ha. depending on the intensity of weed infestation.

The benefits: To achieve good seed cotton yield, losses due to weed competition can be minimized by using this technology. The weed management technology not only increases the yield but also improves the quality of seed cotton.

Pendimethalin *fb* directed spray of glyphosate (during spraying)

Pendimethalin *fb* directed spray of glyphosate (after spraying)

Unweeded cotton

1.12 Non-chemical weed management in okra

The problem: Vegetable crops like okra are severely infested with weeds reducing their yield considerably. The weed problems become serious and their management costly under organic cultivation. Weed management practices other than herbicide use like hand weeding, mulching, stale-bed, intercropping may be useful in management of weeds.

The technology: Apply FYM at 20 t/ha and mulch the okra crop with black polythene sheet from sowing to harvest for controlling weeds without application of herbicides in okra.

The benefits: Applying FYM and black polythene mulch will effectively reduce weed infestation and produce higher yield of okra (19.46 t/ha) in comparison to 2 hand weeding (15.57 t/ha).

Precautions: FYM should be well-rotten and decomposed to avoid addition of weed seeds in the field. The vegetable beds should be clean and well prepared before spreading the black polythene sheet.

Weed management in okra by black polythene mulch

1.13 Non-chemical weed management in tomato

The problem: Vegetable crops like tomato are severely infested with weeds reducing their yield considerably. The weed problems become serious and their management costly under organic cultivation. Weed management practices other than herbicide use like hand weeding, mulching, stale-bed, intercropping may be useful in management of weeds.

The technology: Apply FYM at 20 tonnes/ha and mulch the tomato crop with black polythene sheet from transplanting to harvest for controlling weeds without application of herbicides in tomato crop.

The benefits: Applying FYM and black polythene mulch will effectively reduce weed infestation and produce higher yield of tomato (32.48 t/ha) in comparison to 2 hand weeding (29.02 t/ha).

Precautions: FYM should be well-rotten and decomposed to avoid addition of weed seeds in the field. The vegetable beds should be clean and well prepared before spreading the black polythene sheet.

Weed management in tomato by polythene mulch

1.14 Weed management in direct seeded onion

The problem: Onion is a poor weed competitive crop. The losses in bulb yield due to weeds may go up to 70%. The weed problem is even more serious in direct-seeded onion. Hand weeding though effective causes damage to the onion plants and is a costly operation. Herbicide recommendations have been made to control weeds, but single application of any herbicide is not effective in controlling weeds throughout the crop growth.

The technology: Apply herbicide oxyfluorfen 200 g/ha at 3 days after sowing (DAS) followed by oxyfluorfen 150 g/ha (30 DAS), and oxyfluorfen 200 g/ha followed by pendimethalin 750 g/ha (30 DAS) to control broad spectrum weed flora and for obtaining higher bulb yields.

The benefits: Weeds in onion come in several flushes which are difficult to control by pre-emergence herbicides alone. Hand weeding in onion would be a costly practice. This technology may give 25-52% higher bulb yield as compared to 2 hand weedings.

Precautions: Care should be taken to apply the herbicides at recommended dosage and stage of crop growth. Herbicides should not be applied later than 30-40 DAS in onion. Pendimethalin at 30 DAS can also be applied by mixing in fine sand and broadcasting it in the crop field.

Unweeded onion crop

Oxyfluorfen 200 g *fb* oxyfluorfen 150 g/ha applied in onion

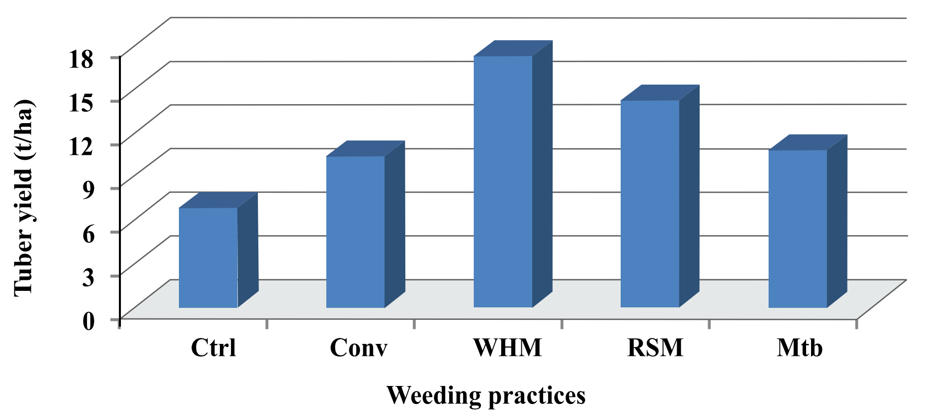
1.15 Non-chemical integrated method of weed management in potato

The problem: Potato is the most important vegetable in India. It is conventionally grown following intensive tillage operation; and subsequently earthing operation is further done to make ridges along the crop row with an intention to fully cover up the crop roots. This takes care of weeds in potato field. However, minimum tillage operation is now-a-days being advocated to counter the deteriorating effects of tillage on soil condition. However, heavy infestation of weeds in absence of secondary tillage operation causes about 50% loss in tuber yield of potato.

The technology: Weeds can be managed effectively in potato through integrated approach. Make shallow furrows and place entire amount of P and K, and 50% of N; mix the fertilizers properly with the soils inside the furrow so that the tubers do not come in contact with the fertilizers during planting. Plant the tubers along the furrow line. Cover the field with thick mulch (12-15 cm) using partially dried water hyacinth biomass or rice straw. Top dress the remaining 50% N at 30-35 days after planting and irrigate the field.

The benefits: The productivity of mulched potato increased hugely over the conventionally-grown potato; and water hyacinth was superior mulch than rice straw. The tuber yield obtained with rice straw was 36% higher than in conventionally-grown potato. The water hyacinth mulch produced 66 and 22% higher tuber yield over conventional practice and rice straw mulch, respectively. Reduction in soil moisture loss, and maintenance of optimum soil temperature and thereby providing tolerance to frost damage are the beneficial effects of mulches, which ultimately increases tuber yield. The release of nutrients from water hyacinth mulch, which is having lower C:N ratio similar to leguminous green manures, during the potato growing period further increases the benefit. Improvement in the aquatic ecosystem as a consequence of removal of water hyacinth for utilization as a mulching material is a priceless benefit.

No mulch **Water hyacinth mulch** **Rice straw mulch**



Ctrl: Control (no weeding); **Conv:** Conventional tillage;
WHM: Water hyacinth mulch; **RSM**: Rice straw mulch; **Mtb:** Metribuzin

Precaution: While putting mulches, care must be taken so that not much obstruction is there on the point of planting. This may cause problem to emergence of potato. Rats may make shelter in the field mulched with rice straw. However, rat problem was not noticed in water hyacinth mulch. Some weeds may come up if the mulches are not placed uniformly and with proper thickness.

1.16 Water hyacinth as green manure to spinach

The problem: The gigantic biomass of water hyacinth, which is otherwise a problem for water bodies, could serve as a resource for agricultural use as the average C:N ratio of this weed (approx 20:1) is comparable to that of legumes. It can be used as green manure in the agricultural lands nearer to water bodies, which are generally cultivated for vegetable crops and frequently irrigated. Low C:N ratio, coupled with the availability of moisture in vegetable fields, favours quick decomposition of this biomass and subsequent release of nutrients, especially of N.

The technology: Chop the water hyacinth biomass into pieces of 2-3 cm length before use. Apply the chopped biomass to field at 4-5 t dry matter/ha and plough the land. This much biomass will release about 100-120 kg N/ha. It should be attempted to use fresh to partially dried biomass for obtaining better result. Following incorporation of the biomass into field, sow spinach seeds. Frequently irrigate the field to keep the soil moist that favours good decomposition of biomass.

Poor growth of spinach in absence of manure

Moderate stand of spinach under FYM addition

Moderate stand of spinach in soil incorporated with dried water hyacinth

Good growth of spinach in soil manured with green (fresh) water hyacinth

The benefits: Compared to Farm yard manure (FYM) treatment, the soil receiving water hyacinth biomass showed similar or higher content of available N, P and K. The spinach yield increased by 25, 42, 85 and 106% over control in the FYM, dry water hyacinth, fresh water hyacinth and urea treatments, respectively. Water hyacinth biomass was inferior to urea in terms of N supplying ability to spinach, but showed better performance than FYM in terms of increasing the nutrient availability in soil and spinach yield.

Treatments	Available nutrient in soil (kg/ha)			Spinach yield (t/ha)	
	Mineral N	Available P	Available K	Fresh weight	Dry weight
Fresh water hyacinth	134	50	512	12.85	1.01
Dry water hyacinth	125	45	522	9.87	0.80
Urea	161	33	384	14.27	1.25
Farm yard manure	127	43	460	8.69	0.74
Control	105	30	362	6.93	0.57

Precautions: Care should be taken that the water hyacinth biomass is not dried up fully before use, as it may cause loss of nutrient. It is also essential to maintain proper moisture condition in the field to facilitate faster decomposition of biomass, and thus proper release of nutrients. Proper care is also needed to manage the weeds in field. Since release of nutrients from organics are relatively slower than from inorganic fertilizers, it is essential to ensure that released nutrients are taken up by crops and not by weeds.

1.17 Weed management through soil solarization

The problem: Innovative approaches to control the pests including weeds are in great demand around the world, particularly those which are cost effective and less harmful to environment. In recent years, there has been increasing concern regarding the hazards of chemicals to the environment, the farmers and the consumers. Therefore, interest in non-chemical approaches which aim to reduce pesticide usage is growing. Manual weeding, though effective and commonly used in India is expensive and time consuming and also is not feasible in all situations. Use of herbicides for controlling weeds is very effective and economical but due to associated residue hazard, evolution of resistant biotypes and polluting the ecosystem have necessitated development of alternate non- hazardous means of weed management. In this light, harvesting of solar energy through soil solarization for controlling soil-borne pests including weeds, pathogens and nematodes will be the key preposition to reduce the dependency on chemicals

The technology: It is based on the principle that light is received by the sun in the form of electromagnetic short waves which would easily pass through TPE. The re-radiation from the earth is, however, through the long waves not permissible through TPE resulting in trapping of heat. The field is irrigated and brought to a fine tilth. The TPE films are laid close to the soil surface and the sides are tucked in the soil to prevent any heat loss. This technique is the best practiced in summer months (April-June) when solar radiation is high; the sky is clear and more importantly the land is vacant. Duration of 4-6 weeks is sufficient to get effective season-long control of many annual weeds.

Soil solarization is a unique method of pest control. It is (i) non-hazardous; (ii) user-friendly; (iii) environmentally- benign; (iv) not dependent on fossil fuel; (v) effective on a wide variety of pests including soil borne fungi, bacteria, nematodes and weeds; (vi) often effective for more than one season or a year, and (vii) stimulatory to crops.

Effect of soil solarization on some major weeds (no./m^2)

Weeds	Non solarized	Solarized
Trianthema portulacastrum	173	3
Digera arvensis	125	3
Acrachne racemosa	943	161
Dactyloctenium aegyptium	139	21
Cyperus rotundus	52	40
Commelina benghalensis	14	0
Avena ludoviciana	9	0
Phalaris minor	41	0
Melilotus indica	88	89
Chenopodium album	30	0
Medicago hispida	35	64

Effect of soil solarization on crop productivity (2 years mean data)

Treatments	Yield (kg/ha)		Total income (₹/ha)
	Soybean	Wheat	
Control	753	2274	25,488
Hand weeding	1470	3741	38,350
Herbicides	1287	2965	37,381
Soil solarization	1952	3738	51,583

The benefits:

- Gives excellent control of many weeds in both *kharif* and *rabi* seasons
- Controls many soil borne pathogens responsible for causing root rot, wilt etc.
- Minimizes plant pathogenic nematodes. By far the most effective treatment for nematodes.
- Enhances crop productivity through control of pests and positive changes in soil biology and chemistry.
- Saves energy as requirement of secondary tillage operations is eliminated.
- Ecologically safe and environment friendly method.

Precautions: It is important to note that, it is a pre-plant treatment and planting or sowing could be undertaken after removing the TPE films. The soil disturbance following solarization be kept to the minimum for the best results. It is a surface phenomenon and hence any tillage (except for opening furrows for placing seeds) would nullify the effect.

View of soil soalrization

2.0 Management of problematic weeds

2.1 Biological control based integrated management of *Parthenium*

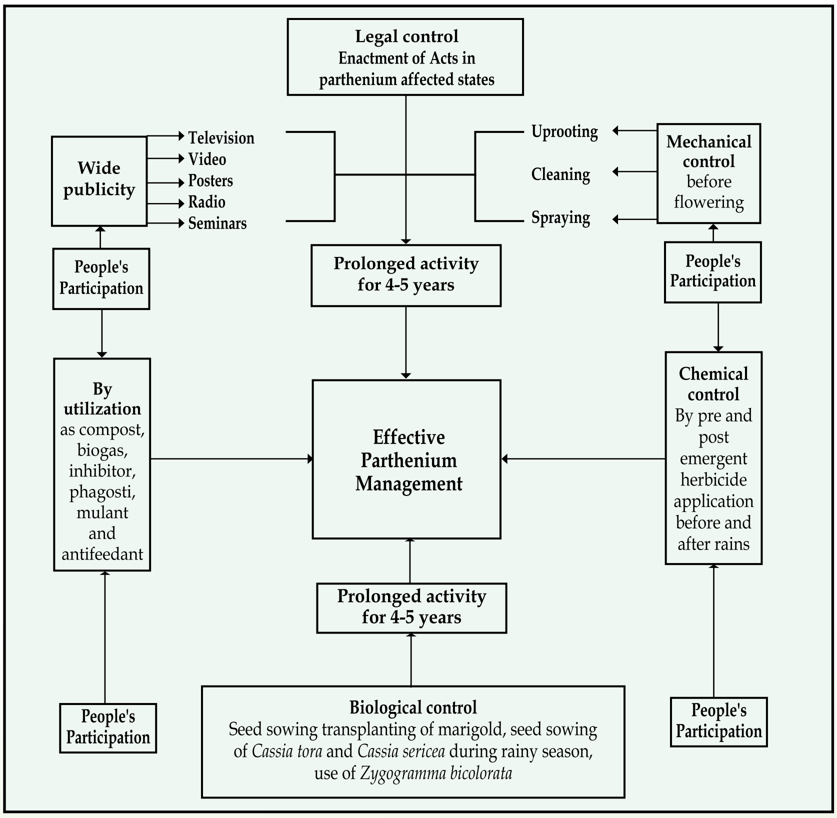
The problem: *Parthenium hysterophorus* L. (Asteraceae) locally called 'Gajarghas' or 'Congress grass' is an alien weed entered into India along with wheat imported from USA in the early fifties. Since then it has spread alarmingly and invaded about 35 million hectares of land in the country. The weed is responsible for causing many diseases like skin allergy, hay fever, breathing problems in man and animals besides depleting crop production and loss of biodiversity. It has the capacity to grow at any time on the availability of little moisture. It has immense power of seed production as up to 25000 seeds can be produced by single plant. The seeds are very tiny and very light hence are easily dispersed in vast area quickly through air, water, vehicle movement *etc*. The management of this dreaded weed is one of the biggest problem for farmers in particular and public in general.

Technology: In non-cropped area during rainy season, host specific exotic bioagent *Zygogramma bicolorata*, a native of Mexico, which is the native of *Parthenium* too, should be released at the rate of 5000/ha at different patches in 50 to 100 numbers, covering the entire area. In addition to Mexican beetle, growing competitive crops (fodder, sorghum, sunflower and maize) in cropped area and or self-perpetuating competitive plant species like *Cassia sericea*, *Cassia tora*, *Tagetus erecta etc.*, in non-crop areas should be used. *Cassia tora* at the rate of 40-60 kg/ha should be broadcasted in the pre-marked *Parthenium* infested sites during March-April. In the protected premises and farms' field, on the road side and bunds in fields, marigold should be grown. By this approach, extra earning can be done by selling the flowers besides beautification of premises. Biological approach is suitable during rainy season, hence integration of herbicide during winter and summer season should be done. The spray of glyphosate 1 and 1.5 kg/ha after pre-monsoon rains during June to early July on small to large growth of *Parthenium* will manage first flush of *Parthenium* effectively. Second flush on the same site will be controlled by the attack of *Z. bicolorata,* if it would have been established earlier in the area. If Mexican beetles have not been released earlier at the given sites, these can be released in patches after pre-monsoon rains for establishment of population.

Manual uprooting of *Parthenium* before flowering during rainy season is very effective hence should be done by individuals in small area and by community participation in large area. The uprooted *Parthenium* biomass should be utilized for compost and vermi-compost making. People of the area should be made aware about the menace and management of *Parthenium* by holding meeting, putting exhibition and showing video films.

The benefits: Integrated management helps to manage the weeds throughout the year as *Parthenium* is able to grow at any time of the year. The use of *Parthenium* for making compost and vermi-compost from the uprooted biomass is also profitable to increase the productivity of the crop.

Precaution: Various methods should be used according to the season to get the maximum benefit. To get maximum benefit of bioagents and competitive plants, they should be used during rainy season. Manual uprooting and compost making from uptrooted biomasss is suitable in rainy season. However, pre-flowered or before flowering plants of *Parthenium* should only be used for compost or vermi-compost making.



2.2 Management of weedy rice

The problem:

- Weedy rice (*Oryza sativa* f. *spontanea* Roshev.) is a menace in direct-seeded rice fields that can cause yield losses between 30-60% depending upon severity of infestation and also reduces harvest quality.
- It is commonly mistaken for wild rice known as '*Sadva, Pasai, Jhara, Jangli dhan*' *etc.*
- Being a rice biosimilar, the weed cannot be distinguished at vegetative stage. But it can be recognized with panicle emergence as they have highly shattering grains which fall to the ground and increase weed seed bank. If not managed timely, the infestations increase largely.
- Hand weeding at vegetative stages is not possible and no selective herbicide is available for its controal. Thus, management based on cultural practices is the only choice.

The technology:

- Use of purple coloured rice cultivars e.g. 'Nagkesar, Shyamla' (as they help distinguish between crop and weed) at higher seed rates in weedy rice infested fields is the basic need of the technology.
- As such, cultivation by puddle transplanting at seed rate of 40 kg/ha *fb* one hand weeding at 40 DAS is the best option to manage weedy rice.
- For DSR, double stale seed bed *fb* sowing at seed rate of 90 kg/ha *fb* one hand weeding at 40 DAS is a better method of choice.

Cultural practice	Dry wt. of weedy rice/m² (60 DAS)	Yield (t/ha)
Stale seed bed (SSB)	28.4 CD	1.71 DE
SSB *fb* hand weeding (40 DAS)	27.9 CD	2.12 BCD
Double stale seed bed (DSSB)	30.6 BC	1.93 CD
DSSB *fb* hand weeding (40 DAS)	22.0 CD	2.21 ABC
Brown manuring (BM)	38.8 AB	1.44 EF
BM *fb* hand weeding (40DAS)	20.5 D	1.86 CDE
Weedy check	40.3 A	1.16 F
Transplanted (TP)	21.5 CD	2.46 AB
TP *fb* hand weeding (40 DAS)	10.2 E	2.69 A
LSD (P= 0.05)	9.3	0.47

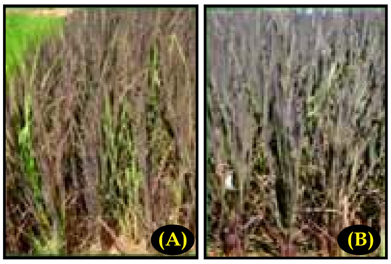


Fig. A) Weedy rice infested plot of purple coloured rice B) after hand weeding

Benefits

- Use of purple coloured rice enables easy recognition of weedy rice due to its green colour.
- Adoption of the technology gradually leads to decrease in weed seed bank over time.

Precautions/Constraints

- The cultural practice adopted needs to be followed every year to gradually eradicate weedy rice from the fields under DSR.
- Use of clean, certified seeds for sowing and clean water for irrigation must be followed to make the technology a success.
- Initially, benefits may very depending on severity of infestation in fields.

2.3 Integrated management of tiger grass (*Saccharum spontaneum* L.)

The problem: Tiger grass (*Saccharum spontaneum* L.) is a serious perennial weed of forage crops, pastures, sugarcane and tea; and common weed of wheat, cotton, jute, maize, peanut, rice and sorghum. It infests nearly 4 million hectares of fertile cultivable land of the country, and often forcing farmers to abandon the entire field. Tiger grass is a pernicious grass which tillers profusely and reproduces rapidly by means of rhizomes. It spreads very fast to turn any neglected areas into a miniature forest in a very short time. Infestation of tiger grass also deteriorates the soil fertility.

The technology: Repeated application of glyphosate 2.0 kg followed by glyphosate 1.0 kg/ha or ammonium glufosinate 1.0 kg *fb* glyphosate 1.0 kg/ha in non-cropped situation after the harvesting of winter season crops probably in the month of April–May. Glyphosate 1.5 kg/ha + summer ploughing followed by *dhaincha*-wheat or soybean-wheat rotations also provide a season long control of tiger grass. This technology should be repeated/adopted for 2-3 times for complete eradication of this perennial noxious weed. Appreciable control of tiger grass can also be achieved with the application of glyphosate 1.5-2.0 kg/ha at 20 days before sowing at active foliage stage of tiger grass followed by fluazifop-p-butyl 300 g/ha or quizalofop-p- ethyl 100-125 g/ha at 20–25 DAS in soybean, chickpea, lentil, mustard and linseed crops.

***Saccharum* infested area**

Effect of fluazifop on *Saccharum* in soybean

Effect of glyphosate and fluazifop-butyl on the growth of *Saccharum spontaneum* L. and productivity of soybean

Treatments	Shoots/m²	Dry matter production (g/m²)	Seed yield (kg/ha)
Glyphosate 2.0 kg/ha	5.80	10.99	1002
Glyphosate 2.0 kg/ha *fb* Fluazifop-butyl 0.3 kg/ha	4.43	9.00	1245
Weedy check	9.46	18.37	563
LSD (P=0.05)	1.32	3.20	253

2.4 Enodon dactylon, convolvulur anvensis and cypurn sotunter is missing

Effect of sequential application of herbicides on tiger grass (*Saccharum spontaneum* L.) in non-cropped situation

Treatments	No. of shoots/m² after 6 months of spraying	Shoot dry weight (g/m²) after 6 months of spraying	Per cent control on dry matter basis
Ammonium glufosinate 1.0 /ha	214	430	17.6
Glyphosate 2.0 kg/ha	155	230	55.9
Ammonium glufosinate 1.0 kg/ha *fb* glyphosate 1.0 kg/ha	38	85	83.7
Glyphosate 2.0 kg/ha *fb* Glyphosate 1.0 kg/ha	52	69	86.8
Control	375	522	-
LSD (P=0.05)	58	69	-

fb - Followed by

The benefits: Results have shown that with the proper adoption of this technology for 2-3 years, the abandoned land could be made free from this pernicious weed and brought under cultivation.

Precautions: After application of glyphosate followed by summer ploughing, the field should not be left unsown. It must be sown with fast growing smother crops and/or must be covered with cropping systems like soybean-wheat, *dhaincha*-wheat for providing season long control.

2.5 Management of *Euphorbia geniculata* in soybean

The problem: Soybean has emerged as one of the major rainy season cash crops in central India. Being the rainy season crop and initial slow growth rate, it suffers heavily due to severe weed infestation, which causes yield loss even up to 80%. Continuous use of a particular herbicide (pendimethalin/fluchloralin/alachlor) resulting in development of sever infestation of non-grassy weeds particularly *Euphorbia geniculata* in Madhya Pradesh. The growth rate of this weed is very high and it suppresses the soybean crop in its early growth stage. Due to longer seed viability and shorter life cycle, controlling this particular weed is a hard task.

The technology: For weed management in soybean various pre- and post-emergence herbicides are available. Among pre-emergence (PRE) oxyfluorfen (200 g/ha) and metribuzin (500 g/ha), and chlorimuron-ethyl (10 g/ha) and imazethapyr (70 g/ha) as post-emergence (POST) herbicide are most effective in controlling the density and dry biomass of diverse weed flora including *E. geniculata* in soybean.

The benefits: Application of chlorimuron-ethyl as POST diminishes the density of *E. geniculata* and total weeds in soybean by 68 and 62%, respectively. The biomass accumulation by total weeds in soybean reduces up to ~70% with PRE application of metribuzin. The application of metribuzin (500 g/ha) as PRE effectively reduces the infestation of predominant weeds, mainly *E. geniculata*; and increases the seed yield of soybean by 2.4 times. The higher additional economic return can be accomplished with application of metribuzin as PRE or chlorimuron-ethyl as POST in soybean.

Treatments	Weed density (no./m^2)		Total weed dry biomass (g/m^2)	Seed Yield (kg/ha)	Treatment cost (Rs/ha)	Additional return over weedy (Rs/ha)
	E. geniculata	Total				
Pendimethalin 1000 g/ha (2 DAS)	10.11	11.88	14.99	1024	1613	4648
Oxyfluorfen 200 g/ha (2 DAS)	6.69	7.55	8.93	1399	2180	9898
Metribuzin 500 g/ha (2 DAS)	4.40	7.48	6.40	1670	1751	13792
Chlorimuron 10 g/ha (21 DAS)	3.57	6.30	7.24	1488	980	11144
Imazethapyr 70 g/ha (21 DAS)	6.86	7.76	8.52	1219	1370	7378
Weedy	11.08	16.69	20.75	692	-	-
LSD (P=0.05)	2.37	3.12	3.15	247	-	-

Metribuzin (500 g/ha) applied in soybean

2.6 Management of parasitic weed-dodder (*Cuscuta campestris* L.)

The problem: *Cuscuta* poses a serious problem in oilseed (niger, linseed) and pulses (blackgram, greengram, lentil, chickpea especially in rice-fallows) and fodder crops (lucerne, berseem) in the states of Andhra Pradesh, Chhattisgarh, Gujarat, Odhisa, West Bengal and parts of Madhya Pradesh under rainfed as well as in irrigated conditions. It is an obligate parasite, which attacks the above ground portions of the host plants and causes a reduction in grain yield to an extent of 35-50% depending on its intensity. The infestation of *Cuscuta* results in heavy loss in terms of quantity and quality of produce. Many times it may cause complete failure of the crops.

The technology: Apply pendimethalin 1.0 kg/ha as pre-emergence for effective control of *Cuscuta* in crops like blackgram, greengram, lentil, chickpea, niger and linseed, except berseem and lucerne. In berseem and lucerne, application of pendimethalin 1.0 kg/ha at 2 week after sowing gives effective control of *Cuscuta*.

The benefits: Very few selective herbicides are available for managing this parasitic weed in pulses and oilseed crops. But the use of pendimethalin could provide effective control of *Cuscuta* for producing optimum yields.

Precautions: Application of pendimethalin in berseem and lucern at 2-3 DAS may be phytotoxic to crop. It should be applied at 2 week after sowing. Proper soil moisture must be available at the time of herbicide application.

Pendimethalin applied in niger

Pendimethalin applied in chickpea

Pendimethalin applied in lucerne

2.7 Chemical control of alligator weed

The problem: Alligator weed, *Alternanthera philoxeroides* (Family: Amaranthaceae) is a native of north-eastern Argentina. This weed is capable of infesting terrestrial, low land and aquatic habitats. It is a problematic weed in 30 countries and has been recognized as an invasive and troublesome weed in rice, corn, cotton and soybean, vegetables and fruit trees in 23 provinces of China. Since its first report from India in 1965, it has spread to 16 more states of India and in some states, it has assumed an alarming situation. This weed has become a nuisance in many aquatic bodies, low land areas' inundated for some time with water and low land terrestrial area. This weed has immense capacity of regeneration from the buried roots after applying herbicides, hence pose challenge for its control. Hitherto, no attempt was made to evaluate herbicides against aquatic or terrestrial form of alligator weed in India in spite of its problem in large area since long time. Therefore, technology was developed for its effective management using herbicides in want of effective biological control.

The Technology: Evaluation of three most used herbicides *viz.* metsulfuron-methyl, glyphosate and 2,4-D was done in ponds, low land plots and naturally infested low land areas. Metsulfuron-methyl was found most effective at 0.016 and 0.024 kg/ha, against the alligator weed in aquatic and terrestrial situations, respectively. It was recommended to control aquatic and terrestrial form of alligator weed for at least six months in Indian situations. If control is required for more than six months, same area should be treated with glyphosate (3.5 kg/ha) followed by metsulfuron-methyl again after six months as repeat application of same herbicides does not control the re-growth of alligator weed effectively.


Alligator weed infestation in pond and low land area

Effect of herbicides on regrowth of terrestrial form of alligator weed at different days after herbicide application (DAA) in naturally infested condition.

Herbicide	Dose kg/ha	Re-growth population (no./m^2)								
		Non-repeat application						Repeat application		
		DAA						DAA		
		30	6 0	90	120	150	180	30	60	90
2,4-D	1.5	1.93*	5.56	6.35	8.27	9.89	13.22	0.71	2.63	9.27
	2.0	0.99	5.13	5.70	7.60	9.17	12.88	0.71	1.40	8.74
	2.5	1.05	3.17	4.30	6.32	7.47	12.16	0.71	0.96	5.23
Glyphosate	2.0	2.83	8.52	8.90	10.46	11.69	12.86	0.71	4.41	8.71
	2.5	2.72	8.18	8.47	9.01	11.05	12.60	0.71	3.45	8.18
	3.0	2.38	7.04	7.66	8.50	9.48	11.47	0.71	2.95	6.16
Metsulfuron-methyl	0.016	0.71	2.77	4.09	6.63	8.62	11.09	0.71	2.91	8.14
	0.020	0.71	2.28	3.17	5.16	6.43	10.48	0.71	2.10	6.84
	0.024	0.71	0.71	2.79	4.05	6.14	9.71	0.71	0.71	4.68
Control	-	26.67	27.82	29.15	35.44	37.25	38.30	35.44	37.25	38.30
LSD (p=0.05)		1.28	1.57	1.50	1.81	1.67	0.76	1.46	1.55	0.93

* Transformed values; DAA: Days after application

The benefits: So far, it is the only effective solution to manage the alligator weed in aquatic and terrestrial situations in India. This method is cost effective than the manual removal.

Precautions: It was observed that if only a fraction of alligator weed population is left uncontrolled, it may again proliferate fast and re-infest the area quickly. Therefore, for long-term control of alligator weed, repeat application is necessary. But repeat application of same herbicides should be avoided owing to resistant nature of alligator weed against the same herbicide in due course. This fact has also been mentioned by other researchers that alligator weed was more resistant to herbicides than other aquatic macrophytes.

2.8 Control of problematic lotus in fish culture pond

The problem: Lotus (*Nelumbo nucifera*) of family Nymphaceae is a rooted emerged weed, grown in ponds from India to Japan and also in Hawaii for ornamental and edible purpose. But in some other aquatic situations like fish culture ponds, lotus may act as a problematic weed. It has also been reported a problem in Dal lake in Srinagar (Jammu & Kashmir). Control of this weed by conventional manual or mechanical method has limitations in addition to expensive cost. There is no suitable biological control options for this weed. Herbicides may provide effective control but people hesitate to use chemicals owing to their unknown effects on non-target organism and water quality. Therefore, work was initiated to find out safe herbicide in context to fish mortality and water quality.

The Technology: In fish culture pond, fishes like *Labio rohita, Cirrhina mrigalla* and *Cyprinus carpio* are cultured. Initially screening of three herbicides namely 2,4-D (ethyl- ester), glyphosate and metsulfuron-methyl was done at 2.0, 2.0 and 0.012 kg/ha, respectively. Among these, glyphosate emerged as an effective herbicide. Therefore, for lotus control in a fish culture pond, glyphosate 2.0 kg/ha should be used safely as it does not harm fish and water quality. The lotus weed is killed within a fortnight after glyphosate spray. No fish mortality occur on using this herbicide in lotus management in the water body. Initially, water quality parameters like pH and dissolved oxygen may be lowered but it become normal in about 15 days.

A pond infested with lotus was controlled with glyphosate

Effect of different herbicides on lotus control (%) and on different parmetres of water quality at 15 days

Herbicides	% control and water quality parameters at 15 days after application					
	% control	pH	Alkalinity (ppm)	Hardness (ppm)	Chloride (ppm)	DO (ppm)
Glyphosate (2.0 kg/ha)	82.7 (97.8)	7.1	251	110	112.4	6.4
2,4-D (2.0 kg/ha)	77.52 (93.5)	7.3	249	120	132.9	5.6
Metsulfuron-methyl (0.012 kg/ha)	59.6 (73.9)	7.4	245	114	115.8	7.2
Control	-	7.8	255	116	105.6	8.6
LSD (P=0.05)	13.2	0.2	6.7	2.9	1.8	0.4

% control values are transformed by arch sign; original values are in parentheses, DO: dissolve oxygen

The benefits: Manual removal of weed is very costly and does not provide satisfactory relief as roots are remain buried in the soil. During removal process of lotus, stems are broken from the roots, which get established and again emerges. Herbicide kill the weed by entering into the system through leaves and also kills roots.

Precaution: Lotus leaves are very smooth hence to enhance the efficacy of herbicides, 1% surfactant like Triton-20 should be added with glyphosate. It was observed that herbicide itself does not lower the pH or dissolved oxygen. Dissolved oxygen is reduced when killed vegetation starts decaying. Therefore, in a severely infested aquatic body, spray of herbicides should be done in different pockets at different time intervals leaving sufficient untreated area for fishes to forage and shelter. Two to three repeated application over a period of 4 months are required for satisfactory control of the weed.

3.0 Mechanical weed management

3.1 Application of non-selective herbicide by DWR wick applicator

The problem: Mechanical hand tools and long handle weeders are adopted for removing weeds at small stages of growth which is time consuming, labour intensive and costly. Weeds grow rapidly and their control by mechanical weeding method is difficult at later stages. The DWR herbicide wick applicator is suitable in these crop situations when weeds are grown up and selective herbicides are not available and recommendations are not there. In these situations, herbicide wick applicator is a better option for application of non-selective herbicides

The technology: DWR herbicide wick applicator has been designed and developed for application of non-selective contact herbicides in between crop rows to kill the weeds. Contact nature of herbicides is applied directly on the weeds, during it's onward movement in between crop rows. The construction details of DWR herbicide wick applicator is detailed below.

- DWR herbicide wick applicator has been designed and developed for application of non-selective, contact herbicides in between crop rows to kill the weeds.
- The herbicide wick applicator consists of cylindrical roller pad, frame, ground wheels, solution tank, and cut off valve.
- The concentrated herbicide solution is stored in chemical tank and flows over to cylindrical rolling pad through cut off valve.
- The cylindrical roller is covered with fibrous clothed pad which is wetted by herbicide solution.
- When the unit is operated in wide spaced crop rows, the wet roller pad/cloth comes in contact with the weed plants. Thus Herbicide solution gets in touch with weed plants and their parts, such as stems, leaves and other parts. Thereby, it kills the weed plants due to herbicidal properties of solution.

Herbicide wick applicator in operation

Methodology: A simple light weight and manually operated herbicide applicator is useful equipment to apply contact herbicide directly on weeds using cylindrical roller with wetting cloth. When operator push the unit, the wheel along with cylindrical roller starts to roll over the weeds in between crop rows and wet cloth with herbicide solution will come in contact with weeds. From chemical tank, solution of herbicide will flow through on/off valve to a tube having small holes lengthwise, which will wet the wetting cloth element on the roller. To have even wetting, herbicide solution will flow through small capillaries to the cylindrical roller. The wet clothed cylindrical roller will roll over during forward movement of the wheel and herbicide will get applied to the weeds. Thus the weeds between crops get killed.

The benefits: The increase in yield of soybean of about 0.5 t/ha has been found in a study at DWR research farm using the herbicide wick applicator resulting in additional income of Rs. 15000/- per ha. The net cost of herbicide application by the wick applicator (two applications) comes to Rs 4200/- taking into consideration that Rs 3000 as cost of herbicide and Rs 1200/- as cost of its application by the herbicide wick applicator (twice). In normal weather conditions, there is saving of Rs 10800/- per ha by using the herbicide wick applicator. Two weeding operations of small hand tool will cost to Rs 9000/- per ha thereby the net benefit comes to Rs 6000/- per ha. In case long handle weeder are used then the cost of mechanical weeding operations is Rs 60000 per ha for two weeding and saving will be Rs 9000/- per ha. Therefore, there is increased saving of Rs 1800 and Rs 4800 in application of herbicide by the use of herbicide wick applicator compared to manual hand weeding by hand tool and mechanical long handle weeder respectively.

Applicability: DWR herbicide wick applicator is a very simple and effective tool for control of weeds in line sown crops by non-selective herbicides. Farmers can easily adopt DWR herbicide wick applicator technology instead of weeding operation by small hand tools or mechanical weeder which involves drudgery and time consuming. The operation of herbicide wick applicator is less tedious as compared to mechanical weeders and sprayers.

Precautions: During operation of herbicide wick applicator, care should be taken that the wet pad should not come in contact with the crop plants which are planted in rows and only the weeds are to be treated sufficiently so that they are killed.

3.2 Development of cycle wheel-hoe weeder

The problem: The wheel hoe is one of the most accepted weeding tool for weeding and intercultural in line sown row crops. The weeding operation by small wheel hoe is tedious and time-consuming requiring more energy particularly in heavy black soil areas. This problem is little solved by development of cycle wheel hoe weeder. It is easy to operated particularly when weeds are small. The weeding operation is faster and comparatively/ easy due to big cycle wheel having bearing and hub in its axle requiring less force for movement of the weeder. The cycle wheel hoe weeder has been developed at DWR and used for weeding in line sown crops in *Kharif* and *Rabi* season.


Cycle wheel-hoe weeder in operation

The Technology: The cycle wheel- hoe weeder consists of main frame, handle assembly, pneumatic cycle wheel (complete with hub, rim and bearings etc.), and fixing arrangement of tine and blade. The wheel diameter is of about 600 mm. The brief specifications of developed cycle wheel hoe is as given below:

Specifications :			
(i)	Length x width x height, cm	=	160 x 55 x 106
(ii)	Wheel diameter, cm	=	72
(iii)	Width of blade, cm	=	25
(iv)	Working depth, cm	=	4-6
(v)	Weight, kg	=	14.5
(vi)	Field capacity, ha/h	=	0.014
(vii)	Material used	=	Medium carbon steel, structural mild steel, M.S. thin wall pipes.

Methodology: The handle assembly has a provision to adjust the height of handle suiting to operator. All the soil working components of the tool are made from medium carbon steel and hardened to 40 – 45 HRC. The other assemblies of the cycle wheel hoe are made from structural mild steel and mild steel thin walled pipes. The working depth of the tools can be adjusted with the help of clamp or through the multiple holes plate provided in the frame and welded to the tool assembly. The tilling depth of the tool and handle height is adjusted and the wheel hoe is operated by repeated push-pull action, which allows the soil working components to penetrate into the soil and cut/uproot the weeds in between the crop rows. Thus the weeds get buried in the soil.

The benefits: The labour requirement for manual hand weeding requires about 40 man-days/ha which requires an investment of Rs. 5000/- per ha. The weeding by cycle hoe requires 10 man – days/ha, which will cost to Rs. 1500/-. The saving of Rs. 3500/- per ha can be achieved with use of the cycle wheel hoe weeder for weeding in crops like soybean, maize, chick pea, mustard etc. The first and second weeding operation will be carried out timely with this mechanical device resulting in getting higher production in field crops.

Applicability: The cycle wheel hoe weeder is a useful weeding implement for farmers to do mechanical weeding operation for field crops, vegetables and other situations. Higher work capacity, weeding efficiencies and profitability can be achieved under light soil or loamy soil conditions.

Precautions: The operation of cycle wheel hoe weeder is tedious under heavy black soils particularly when soil moisture is either low (resulting hardened soil) or high (when soil becomes too sticky). For efficient weeding operation, the cycle wheel hoe weeder is properly adjusted according to height of operator so that the blade is appropriately cutting the weeds and penetrated at shallow depth of soil mass.

3.3 Manually operated rotary push type weeder

The problem : Weeding and mulching is desired in line sown crops like groundnut, safflower, soybean and sorghum having row space between 22.5 to 45 cm. Soil crust is formed in heavy soil due to rain water or irrigation. But it also required to be broken for soil aeration. Keeping, both the objectives, manually operated rotary push type weeder was designed, developed and evaluated at DWR Jabalpur. This weeder is having two row of claw wheels having sharp edged rotors. Which penetrates in to soil while its operation. The operation of rotary push type weeder helps in breaking of soil crust and also preparation of soil mulch after cutting of weeds due to its to-and-fro or push-pull motion.

Rotary push type weeder in operation

The technology: The manually operated rotary push type weeder consists of two rolls of pointed edge disc rotors mounted on two shafts, mounted in a frame in front of cutting blade. The cutting blade is attached to a tine on to main frame for weeding and cutting of weeds. The two rolls of pointed edge disc rotors are moving into the soil so that, it breaks the soil crust as well it mixes the cut weeds during to-and-fro motion of the unit. The weeder frame is fixed on a long pipe frame at one end and a handle on other end. The brief specifications of the unit is described as below:

Specification :			
(i)	Length x width x height, cm	=	197 x 45 x 112
(ii)	Pointed edge star disc dia, cm	=	57
(iii)	Pointed edge star disc circumference, cm	=	180
(iv)	Length of pipe (joining handle and weeder frame), cm	=	145
(v)	Gap between pointed edge star discs, cm	=	3
(vi)	Width of frame holding pointed edge disc rotors, cm	=	10
(vii)	Width of cutting blade, cm	=	15
(viii)	Field capacity of the rotary push type weeder, ha/h	=	0.01
(ix)	Weight, kg	=	12

Methodology : The rotary push type weeder is operated in between crop rows. The pointed edge star rotor disc penetrate in to the soil and break the soil crust and loosened the soil and partly damages the weeds grown. The cutting blade behind the rotor disc cut the weeds in push action. The to-and-fro motion of the unit create the soil mulch in between crop rows to conserve the soil moisture.

The rotary push type weeder is suitable for weeding at 15- 30 DAS during first weeding and 30-40 DAS during second weeding. The weeding operation is tedious as it requires more efforts while weeding, compared to other long handle mechanical weeds. The cutting of weeds and mulch preparation is good and effective.

The benefits: The weeding operation carried out by rotary push type weeder results in higher crop yields. The manual weeding by hand tools "*Khurpi*" needs 40 man-days/ha costing to Rs. 5000/- per ha approximately. The cost of weeding by the rotary push type weeder is Rs. 2250/- per ha approximately taking into account that it required 15 man - days/ha and assuming labour cost to Rs. 150/- per day. There is saving of Rs. 2750/- per ha by adopting the rotary push weeder which amounts to 55% less in cost of operation. The unit gives an added advantage that it breaks the soil crust , mixes the weeds, creates the soil mulch on top of soil in between crop rows.

Applicability: The rotary push type weeder is suitable for weeding and mulching operation in under slightly dry conditions when soil moisture is around 12 to 16%. But, the soil sticks to pointed edge star rotor when soil moisture is higher. Therefore, the weeding operation at higher moisture content is not advisable.

Precautions: The pointed edge star rotor should be kept away from human being as it may cause injury. All nut-bolts should be kept tightened so that rotor blades are kept apart at fixed distance during weeding operation. The unit is slightly heavier at lower portion, therefore, its handling should be done carefully during weeding operation.

3.4 Twin wheel hoe weeder

The problem: In crop season, limited time is available for weeding operation before critical period to the farmers. Therefore, efficient weeding tools are required to achieve maximum output in the limited available time. CIAE twin wheel hoe is one such mechanical weeder which is recommended to farmers. Design modifications were made to improve the work capabilities and weeding efficiency of CIAE twin wheel hoe.

Twin wheel hoe weeder in operation

The technology: In order to increase the work efficiency of CIAE twin wheel hoe weeder, studies has been carried out to make design improvements so that it requires less energy and more economical. Twin wheel hoe weeder with design improvements at DWR has been developed and fabricated. Major improvements made in the design of CIAE twin wheel hoe weeder are as follows-

- The length of long handle is reduced to 157 cm from 190 cm.
- The two ball bearings were incorporated in the hub of twin wheels.
- The length of handle can be adjusted in the new design of the weeder according to the height of the operator. It helps in effective transmission of force and easy weeding operation.
- The cushioned handle grips were provided for easy and comfortable weeding operation in field.

Specifications :		
Length x width x height, cm	:	157 x 58 x 106
Weight, kg	:	6
Number of rows	:	One
Shape of blade	:	V- shaped
Power source	:	One person
Width of coverage, cm	:	20
Depth of tilling, cm	:	2-3
Field capacity, ha/h	:	0.01 – 0.015
Weeding efficiency, %	:	78
Labour requirement, man-days/ha	:	75 – 100

Methodology: The weeding tools are used in standing crops, therefore, they are to be operated with great care so that they do not damage the young crop plants. The working width of twin wheel hoe weeders blade should always be sufficiently less then the crop row spacing. For example, if crop row spacing is 25-30 cm, then 20 cm width of blade of twin wheel hoe weeder is recommended for use. The moisture condition should be such that the twin wheel hoe is easily operated and high weeding efficiency and work capabilities can be achieved. The twin wheel hoe weeder is to be operated in between 15-30 days after sowing (DAS) for first weeding operation and from 35 - 50 DAS for second weeding operation in different *Kharif* and *Rabi* crops sown in lines.

The benefits: The twin wheel hoe weeder is light in weight simple in its weeding operation and at the same time most efficient. The labour required for manual hand weeding is 40 man-days/ha approximately costing Rs. 5000/- per ha. With the use of modified DWR twine wheel hoe weeder, one ha cropped area can be weeded by 12 man-days requiring an investment of Rs. 1800/- per ha. Saving of Rs. 3200/- per ha can be achieved by using DWR twin wheel hoe weeder which amounts to 64%.

Applicability: The DWR twin wheel hoe weeder is a manually operated push type weeder. It uproots and cut the weeds in upland row crops. It is suitable for weeding crops like soybean, maize, gram, mustard, pigeonpea, etc., sown at 25 cm or more row spacing. The DWR, twin wheel hoe weeder is suitable for use in soils from light soil to heavy black soils..

Precautions: The modified DWR, twin wheel hoe weeder has provisions for adjustment in its length of handle according to height of the operator. The modified unit gives higher weeding efficiency when the parts are properly adjusted. The blade needs to sharpened if its edge is blunt or worn out. If required, new blade is to be replaced. All nut-bolts of the unit should be tightly fixed. The unit is to be operated in push pull action of operator. Handle grips are provided, so that it is easier and comfortable to operator for operating the unit for longer durations.

3.5 Engine-powered aquatic weed cutter/harvester

The problem : Large scale weed infestation has been found in ponds, canals and water-bodies in different parts of the country. At present no mechanical aquatic weed cutter / harvester are available or commercially manufactured in India. Some designs of aquatic weed cutters and harvesters either imported or indigenously developed are used in nearby areas of sea-coast. The cutting of aquatic weeds and their collection is a big problem in the country.

The technology: A engine-powered aquatic weed cutter/harvester for small water-bodies and ponds has been designed, developed and evaluated at DWR. The machine consists of main frame, two hull structures, cutting unit, power transmission from engine to cutter bar, a 8 hp engine, steering system, engaging and dis-engaging lever for power to cutter bar, propeller fan, rudder plate, ground transport trolley etc. A boat engine was incorporated for achieving higher forward speed of the machine along with the 8 hp engine in the system. The machine was evaluated and found working satisfactory.

Engine powered aquatic weed cutter in operation

Methodology: The developed weed cutter/ harvester was mounted on the trolley especially designed with pneumatic wheels and trailed behind the tractor for carrying the unit to the pond and aquatic bodies. After reaching the pond shore, the tractor is engaged and the trolley is kept in such a way that just rear half portion of it is on the water and half on the ground. In that position, the machine is pushed into pond or lake. The cylindrical shape hulls are hollow and it allows the machine to half in water. Thereafter, the both engines were made on / start. The boat engine which is mounted at the rear end is powered to drive the unit which is of 9.5 hp through the propeller fan. The 8 hp engine mounted in between the cylindrical hulls on the metallic frame is driving the reciprocating cutter bar fixed on the front end of the machine and cuts the aquatic weeds by its reciprocating movements. The height of the cutter bar can be adjusted according to the cutting requirements of soil-emerging aquatic weeds. The cut aquatic weeds are floating and collected in a net collector especially developed attached at the rear end of the machine. The collected weeds are brought to the shore of lake or pond and front end of the collector is lifted upwards and aquatic weeds are unloaded from the machine's collector unit. After completion of weed cutting and collection at shore, the machine is mounted on the trolley following the same procedure as done for lowering of the unit and finally brought back by towing the machine behind the tractor as tractor trolley.

The benefits: The removal of dense aquatic weeds from pond or water bodies required about 40 man-days per ha costing to Rs. 8000/- per ha. The working width of aquatic weed cutter is 1.2 m having speed of operation 0.1 km/hour during cutting operation. The theoretical work /field capacity of the machine is 0.12 ha/hour. The machine attained the actual field capacity of 0.108 ha/hour taking into account the time loss during turning and idle running. The cost of aquatic weed cutter/harvester is Rs 15 lakhs. The fixed and operating cost of the machine is estimated to Rs 260 and Rs 220 per hour considering 10 years life and 600 hour of use per year. The cost of removal of aquatic weeds of soil emerging and erect nature by the developed aquatic weed cutter/ harvester comes to Rs 5200. If the aquatic weeds are removed by manual labourers, then it costs Rs. 8000/- per ha and by the developed weed cutter/harvester is Rs 5200/- per ha. Therefore, there is net saving of Rs 2800/- per ha. The other benefit is that the weeds removal operation is easy, less tedious and fast.

Applicability: The developed aquatic weed cutter / harvester is suitable for removing aquatic weeds from small ponds, canals and small water bodies. The unit can be owned by Village - Panchayats, irrigation departments and Municipal Corporations.

Constraints / Precautions: The limitations of the developed aquatic weed cutter/ harvester is that the machine is not suitable for removing floating weeds like water hyacinth, *Sylvania* and other floating aquatic weeds which are creating problems in many aquatic lakes, ponds and water bodies. The precaution is to be taken while operating the developed aquatic weed cutter / harvester to ensure and keep balance of the machine level on both the cylindrical hulls, front and rear portions, so that the machine should not sink.

3.6 Spraying technology for herbicide application

The problem: The aim of weed control is to reduce the number of actively growing weeds that interferes with the growth and harvesting of crop. Now a days herbicides are commonly used to prevent, retard and killing the weeds. Herbicidally active chemicals are formulated or prepared so that they are chemically and physically stable during manufacturing, storage, mixing and spraying. All herbicide are capable of being absorbed by plant parts, shoots or roots and moved within the plant to the site of actively growing parts and giving the required response. The effectiveness of the herbicide spray depends on the relationship between the target surface, the spray mixture and the spray droplets cloud. Therefore, it is important to use appropriate spraying nozzles in sprayers to cover uniformly the weed plants in crop field so that required quantity of herbicides reached to target for its response.

The technology: Several types of sprayers are marketed, mostly manually operated one but a few of them have become popular. Hydraulic knapsack sprayer is a low cost machine, easy to maintain and a small holding farmers sprayer. It is particularly suitable for spot and band application of herbicides. For uniform band application, fan, flat-fan and impact flood jet nozzles are recommended for herbicide spraying technology. The fan and flat-fan nozzles are shown in the figure. To avoid drift of spray droplets, nozzles are shielded during herbicide spraying by a protective cover/plastic shield. It helps in uniform deposit of spray droplets in the form of band application of herbicides on the weeds in between crop rows. The plastic shield is quite useful to protect crop plants from herbicide droplets.

Spraying of herbicide using Fan Nozzle with plastic hood.

Methodology: The herbicides are required to be applied precisely and uniformly on weeds in band application. The sprayer along with flat-fan or fan nozzles are calibrated in laboratory for their discharge rate at particular operating pressure, swath width covered, speed of operation. Based on measurement of these parameters, the theoretical application of spray volume is calculated from equation.

$$\text{Application rate (spray volume) (l/ha)} = \frac{\text{Flow rate (l/min) x 10000 (m}^2\text{)}}{\text{Swath width (m) x speed of operation (m)}}$$

The ideal procedure is to select the correct nozzle from the wide range of available nozzles for the desired application rate (l/ha). Most of the herbicides gives higher weeding efficiency at application rates (i.e. spray volumes) varying from 200 to 400 l/ha. Field calibration is also required before field spray application. For this, mark a measured area, in field for spraying, fill the sprayer tank with water to a mark in the spray tank and carryout the spraying using selected nozzle. After spraying, the measured area, find out the volume of water spent to spray the marked area in field by filling the water to the same level marked in the spray tank (in liters). Based on the amount of liquid sprayed (l), calculate the amount of spray volume required to cover one ha area.

The benefits: The correct application of herbicides results in increase in crop yield. The cost of herbicides and its application cost normally varies between Rs. 1500/- per ha in terms of %, because yield of all crop can not be incurred 0.8-1.0 t/ha to Rs. 4000/- per ha. The herbicide application results in increase in crop yield at least 0.8-1.0 t/ha giving economic benefit of approximately Rs. 8000 to 15000/- per ha. If herbicides are not selected properly according to recommendations and their application is faulty, then profit occurred to farmers are reduced and sometimes results in negative outcome.

Applicability: Due to increasing weed problems, the spraying of herbicide is essential which to be adopted by all farmers who are producing grains, pulses, oilseeds, vegetables and horticultural crops. The procedure developed for correct herbicide application consisting of selection of appropriate spraying nozzles, adequate volume of spray liquid, laboratory calibration of nozzle and field calibration, recommended dose of active ingredient, use of plastic shield/cover for spraying if required to avoiding drift of spray droplets, proper safety in handing and spraying. These aspect are essential to be followed by all farmers who are using herbicides in their field crops, vegetables and horticultural crops.

Precautions: The sprayers, nozzles and the gadgets are sometime not easily available in market to meet out the precision/requirements. The training to farmers is essentially required to educate them for proper handling, calibration and application of herbicides to achieve maximum profit.

4.0 Biological weed management

4.1 Biological control of *Parthenium* using Mexican beetle

Skin allergy due to *Parthenium*

The problem: *Parthenium hysterophorus* L. (Asteraceae) locally called Gajar ghas or Congress grass, is an alien weed entered into India along with wheat imported from USA in the early fifties. Since then it has invaded about 35 million hectares of land in the country and has become notorious for causing many diseases in men and animals besides depleting crop production and loss of biodiversity. The invasive nature of weed is due to its high number of tiny, seed production which are able to germinate at any time on the availability of moisture. The management of this weed is one of the biggest problems for farmers in particular and public in general. There are several methods to manage *Parthenium* but among them, biological control has emerged as one of the promising methods.

Action of beetle

The technology: In non-cropped area, *Parthenium* can be managed effectively by using a host specific exotic bioagent *Zygogramma bicolorata* commonly called as Mexican beetle. It is a native of Mexico, which is also the origin of *Parthenium*. The bioagents should be released in the *Parthenium* infested area in 50 to 100 numbers in different patches covering entire area during rainy season when plenty of fresh *Parthenium* plants are available. After establishment, the bioagents will themselves migrate from non-cropped area to cropped area to destroy *Parthenium* amidst the crop. Adults and grubs stages of beetle defoliate and kill the *Parthenium*. Once established, bioagents will work on sustainable basis. On approaching of winter season, a survived population undergoes in diapauses in soil and emerges intermittently in future when conditions become suitable for them. Maximum population emerges during next rainy season but a part may be emerged throughout the year. Augmentative release may be made by collection of adult beetles from area of abundance to area of poor establishment to increase the existing population of the bioagents.

The culture of this bioagents can be procured free of cost from the Directorate. This bioagent has been established in large area, hence can be collected from the established area for further release in non-established area.

Before release | Complete defoliation | Complete defoliation before flowring | Restoration of biodiversity

Fig. Sequence of restoration of biodiversity due to continuous work of bioagents in the given site

The benefits: A study made at Jabalpur based on the herbicide cost, which would have incurred in the area controlled by the beetle, the net economic return was calculated 135% per annum by third year, which increased to 608, 2700, and 12150% per annum for single application of herbicides by 4 , 5 and 6 years, respectively . The total benefit by biological control in six years had been of Rs 62.34 million; 15585% benefit over initial investment. It was concluded that return would have increased many folds if benefits derived in the form of environmental safety and sustainability is taken into consideration.

Precautions: The initial releases must be made on small and succulent growth of *Parthenium* in undisturbed land because in cultivated land, ploughing may disturb the pupation process hence poor survival and subsequent establishment. Low lying areas prone to water logging should also be avoided because pupation takes place in soil and if bioagents are released in such area, there will be no proper population build-up due to death of pupae during inundation of the area. Bioagents should be released during rainy season for fast establishment. They will be no use to release during winter and summer season.

4.2 Biological control of *Parthenium* using competitive plants

The problem: *Parthenium* has become a nuisance in cropped and non-cropped area both due to its immense capacity of reproduction by tiny and very light weight seeds. It is very hard to control this weed by any single method, therefore, we need to think other methods also. There are several methods of its control among which biological control has been considered one of the best methods owing to its environment friendly nature. Many plants like *Xanthium strumarium, Tephrosia purpurea, Achyranthes aspera, Vitex negundo, Cassia sericea, Cassia tora, Cassia* spp. and *Cannabis sativa* were found to be competitive against *P. hysterophorus.* Among these plants, all cannot be used owing to one or other reasons. Studies conducted on *Cassia tora* and *C. serecia* proved them safe botanicals to suppress *Parthenium*.

The technology: In addition to Mexican beetle, growing competitive crops (fodder, sorghum, sunflower and maize) or self-perpetuating competitive plant species like *Cassia sericea*, *C. tora* and *Tagetus erecta* (marigold) in non-crop areas is also effective to manage *Parthenium*. In non-cropped area, *Cassia tora* at the rate of 40-60 kg/ha should be broadcasted in the pre-marked *Parthenium* infested sites during March-April. The plants will grow during rainy season and will suppress *Parthenium*. In due course, sufficient seed bank of *C. tora* will be developed at the given site which will replace the *Parthenium* seeds in coming years (Table 1). In protected premises like offices, institutions and farms, on the road side or bunds where *Parthenium* grows, marigold should be grown. Bunchy red coloured marigold should be used for this purpose. Flowers of marigold offer to god in temples may be collected and dried. Seeds may be separated and these should be broadcasted on the road side in the premises. In the crop field, this may be grown on the bunds.

Replacement of *Parthenium* by *Cassia tora* and marigold on road side

Previously mentioned Population of *Parthenium, C. tora* and other weeds at experimental site after broadcasting of seeds of *C. tora* in 2005

Site	2005				2006				2007			
	Broadcasted		Control		Broadcasted		Control		Broadcasted		Control	
	Parthenium	*C. tora*	*Parthenium*	*C. tora*	*Parthenium*	*C. tora*	*Parthenium*	*C. tora*	*Parthenium*	*C. tora*	*Parthenium*	C. tora
Site 1	57.26	28.8	56.86	11.2	28.2	34.6	11.6	30	12.6	28.0	7	25.4
Site 2	56.73	38.7	58.25	14.6	39.6	10.6	17.2	0	13.6	11.4	11	2.0
Site 3	60.96	45.9	70.68	12.7	5.8	17.4	12.4	1	4.6	19.6	3	7.6

The benefits: Marigold will dominate over *Parthenium* besides to suppress nematode populations in the crop field. By this approach, extra earning can be done by selling flowers besides beautification of premises.

Precautions: Seed coat of *C. tora* is very hard, therefore, the seeds should be broadcasted before 3-4 months of rainy season to get sufficient time to be abraded. There will be very less germination, if seeds are broadcasted on the commencement of rains.

4.3 Biological control of water hyacinth using *Neochetina* weevil

The problem: Water hyacinth, *Eichhornia crassipes* Mart (Solmns.) is a free-floating, annual or perennial exotic aquatic plant of Brazilian origin. It was first introduced into West Bengal, most probably in early 1890s as an ornamental plant but by now it has spread in all types of fresh water bodies throughout the country covering 0.4 million hectares of water surface. It has been categorized as one of the most problematic aquatic weeds of India. Due to its negative impacts, the water hyacinth is popularly known as 'Blue devil'. Water hyacinth is a big menace severely affecting navigation, fishing, recreational use of aquatic bodies and hydroelectric generation besides causing human health hazards by harbouring harmful insects and vectors of diseases like malaria, encephalitis, filariasis *etc.* By increasing the evapo-transpirational losses of water, the weed is responsible for drying up of innumerable water tanks and ponds, which are the mainstay of agriculture in rain fed areas. This weed has received prime attention by the planners and government. Manual or mechanical methods of its control are not cost effective. Use of herbicides may have potential risks on non-target organisms and water quality. In such situations, biological control has been emerged as one of the best methods.

Water hyacinth infested pond

The technology: Exotic bio-control agents, two coleopteran weevils *viz.*, *Neochetina bruchi* and *N. eichhorniae,* commonly called *Neochetina* spp. are effective bioagents for biological control of water hyacinth. The culture of these bioagents can be procured free of cost from Directorate of Weed Research or can be collected from the bioagents' infested water hyacinth from the aquatic bodies. The presence of bioagents can be distinguished easily by observing feeding scarce on the leaves of water. The bioagent at the rate of 500 to 1000 number may be released in the water hyacinth infested aquatic bodies in different places in 50 to 100 numbers in each patch covering entire pond area. Though weevils can be released at any time of the year, but the optimal time for initial releases, especially in tanks and lakes is after the commencement of rains. Once, the bioagents are established in the released area, the

Bioagent *Neochetina* spp.

augmentative release may be made by collection of adult weevils from the area of abundance and to release in the area of poor establishment. After multiplication, the bioagents will themselves migrate from attacked area to non-attacked area of the pond. In general, more number of releases means quicker establishment and control of the weed. The bioagent control water hyacinth in cycles. In general, weevils take about 18-48 months to achieve first cycle of control depending upon the number of initial releases. Second and subsequent cycles of control may occur in 9-20 months depending on the population build-up of the beetle the seed bank of water hyacinth in the pond.

A same pond before (1) release of bioagents; severe infestation of bioagent has dried the weed (2) and clear pond (3) after gradual decay and sinking of attacked water hyacinth inside the pond.

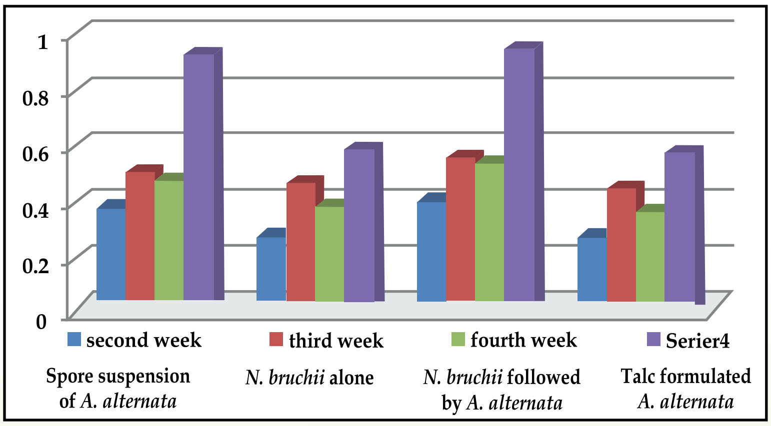
The benefits: In general, manual removal of water hyacinth from one hectare area may cost about 2 to 3 lakh for one time removal while biological control may cost only a few thousands for initial release. Minimum efforts are required for establishment of the bioagents. The benefit is more than the direct estimated cost in terms of environmental safety. Flower production of the weed will be stopped within a year of release.

Precautions: The bioagents work well in perennial water bodies, therefore, release of bioagents should be avoided where water is likely to dry during summer season. In such situations, bioagetns will not be able to multiply due to anchoring of roots in the soil as its pupation takes places amidst the floating roots of water hyacinth. The bioagents will not work effectively in running water like rivers and irrigation canals because bioagents' population will disperse fast in such situations.

4.4 Biological management of water hyacinth using *Alternaria alternata*

The problem: Water hyacinth is freshwater free floating exotic aquatic plant with beautiful large flowers, thick leathery leaves and stolons underwater. It was first recorded in the state of Bengal in 1896 in India. It affects navigation in rivers, blocks irrigation channels and obstructs the flow of water to crop fields in many states. Rapid multiplication occurs by stolons and population is doubled in 10 days. Seeds sink to the bottom of the water body and remain in dormant state in the mud for as long as 20 years. Mechanical management is done using boat harvesters, repeatedly and which is very costly. Chemical management is not recommended in aquatic systems and thus biological control is a viable and effective alternative.

The technology: *Alternaria alternata* isolate DWSR 1 was isolated from the native water hyacinth plants from the aquatic bodies near Jabalpur. Talc formulation was prepared from the pure culture of the fungus. This was sprayed at 1.0 kg talc product in 50 litres of water (filter the contents to remove settled talc) and spray the solution with large nozzle in knapsac sprayer. Recommended dose is 1.0 kg/100 m^2. Before spraying, cold rice (starch) solution (250 ml/kg of talc formulated product) may be added for better sticking of the sprayed solution. Tween 80 at 0.01% (1 ml/10 litres) can be used instead of the rice solution. Spray in the evening to take advantage of the relative humidity for the fungus to establish. The symptoms starts with typical dark brown spots in the leaves, stem and in the air sacs. The individual spots rapidly grow in size and coalesce to form a big spot which soon turns into sunken depression leading to the rotting of the affected portion. The main damage is caused to the destruction of the air sacs which result in the sinking of the plants. The fungus was found to rapidly kill the plants in a period of 3 to 4 weeks and when integrated with the beetles *Neochetina* spp. the efficiency of the fungi was found to increase.



Disease intensity after application of *Alternaria alternata* and *Neochetina bruchii*

Different stages of *Alternaria alternata* infection on Water hyacinth

The benefits : The fungus is host specific and attacks only water hyacinth. It is eco-friendly and safe to fishes and does not cause any toxicity in water. Once sprayed the fungus can sustain itself and causes disease in fresh water hyacinth plants also. It is very cheap and safe when compared to chemical or mechanical harvesting methods.

4.5 Biological management of silk leaf weed *Legascea mollis*

The problem: *Lagascea mollis* (velvet weed or silk leaf weed) is an annual herb and introduced exotic weed in India. It is found on cultivated land and in pastures, along field bunds, roadsides and channels. It grows throughout the year, but appears severe in late *Kharif* and early *Rabi* (August to November). It occurs in maize, mustard, millets and vegetables. *L. mollis* harbors bollworms which survive and proliferates during offseason of cotton. Since they are broad-leaved weeds occurring in broad-leaved crops, chemical management of these weeds is not very easy and effective. Hence biological management is considered a effective method that can be integrated with other management methods.

The technology: Natural infestation of the rust pathogen *Puccinia noccae* was observed on the *L. mollis* in the fields around Jabalpur. The pathogen affects the green leaves and the bracts. Stem, roots and the flowers are not affected. Because of the rapid loss in the green photosynthetic parts, the *P. noccae* causes quick death of the whole plant. Plants when affected at their young stages do not produce flowers and produce less number of tillers. The pathogen generally survives as dormant teliospores and upon favorable climatic conditions, produces uredospores, which are the pathogenic stage of its life cycle. Host range studies indicated that *P. noccae* is highly specific on *L. mollis* and hence may be used as a biocontrol agent against *L. mollis*. At present, since artificial culturing of the obligate rust fungi is not possible, the rust spores can be easily collected from the leaves of naturally infested plants and a spore suspension can be made in water.

P. noccae infection on *L. mollis* photograph showing host specificity of *P. noccae*

P. noccae infection on *L. mollis* photograph showing host specificity of *P. noccae*

This can be applied using garden sprayer on the weeds. Spore suspensions are also available at this Directorate during the season. Once established in a particular area, *P. noccae* occurs regularly in the weed plant in that area in the month of August and continues till the month of February.

The benefits : The rust pathogen causes reduction in the number of productive tillers of the silk leaf weed and also kills the plant in heavy infections. This results in the lesser quantity of weed seed bank for the next season building up of the inoculum. Application of this rust fungus is very simple and easy. It does not require any special machines or laboratory facilities. Farmers need not depend on other agencies, but they themselves can collect the spores from the natural infestations, make spore suspensions in water and spray on the weed plants in the fields.

4.6 Mass multiplication of Mexican beetle in tubs

The problem: *Parthenium hysterophorus* popularly known as 'G*ajar ghas*' or 'Congress grass' has been considered one of the most problematic weeds in India. Biological control requires the releases of bioagents. To make introductory releases in new areas and for augmentation in the established site to achieve early defoliation, mass multiplication of the beetle is an imperative step. It has been realized that mass multiplication of *Z. bicolorata* in laboratory conditions is not cost effective due to large number of labours requirement besides electricity and laboratory equipments. In laboratory rearing, higher mortality of grubs is also caused due to diseases. It is also an established fact that continuous mass rearing in laboratory reduce the vigour of bioagent after 3-4 generations. If mass multiplication of biogents is not cost effective, it will not be suitable for mass release and will also not be adopted by the stakeholders. Therefore, cost effective, labour saving mass multiplication techniques of effective bioagent *Zygograma* was developed.

The technology: Beetles are easy to multiply due to its high reproductive capacity. *Z. bicolorata*, is capable of breeding at 20-30 ^{0}C and 50-80%, temperature and relative humidity, respectively. The best development of beetle was observed at 25-28 ^{0}C temperature and 70-75% relative humidity. As per the need, beetles can be mass multiplied in different situations. A single female is capable to lay 500 to 2500 eggs during its life. From these eggs, 50-95% grubs hatch out of which more than 77% are capable to develop into adult. If requirement of beetle is less, these can be multiplied easily in plastic buckets and tubs out side laboratory/house during rainy season. Buckets can be cut and fitted with wire-mesh windows on the sidewall above the bottom level. A six to 10 cm layer of soil should be gently

Multiplication of mexican beetle in plastic buckets and tubs.

compacted in the containers. The mouth of bucket can be tied with the net cloth or with lid, fitted with the wire mesh. Like wise plastic tub or other such containers can be used. As the height of tub remains low, a wooden stick should be inserted in the soil below the net cloth to provide space for growing *Parthenium*. These containers can be kept outside the laboratory or house. In these containers, transplanting of small *Parthenium* should be done. After establishment of plants, 2 to 3 pairs of beetles should be released for egg laying purpose. Old and eaten *Parthenium* plants should be replaced regularly with the fresh plants as and when required. As a thumb rule, while transplanting fresh plants, minimum soil disturbance should be done. This method requires less attention than the laboratory method. From each such container, 100-125 beetles can be obtained in a period of one month.

The benefits: From this method, disease free and robust bioagents can be obtained easily at the cost of 70-80 paise/beetle. This method is particularly beneficial if beetle requirement is less in numbers.

Precautions: After one week of egg laying, adults should be removed to avoid eating of leaves of parthenium. Completely eaten plants should be replaced time to time to provide fresh food to the grubs. After emergence of fresh beetle, these should be collected immediately to avoid eating of transplanted *Parthenium*.

4.7 Mass multiplication of Mexican beetles in cages

The problem: *Parthenium* locally called 'Gajar ghas' has emerged as one of the most problematic weeds in India, responsible for causing harmful effects in men and animals. Biological control by Mexican beetle has been considered one of best approaches for its management. This approach requires the releases of bioagents for which mass multiplication of bioagents is an imperative step. To obtain disease free and robust bioagents at low cost, easy and cheap method of mass multiplication is essential. If mass multiplication of biogents is not cost effective, people will not opt this effectively. Many times, comparatively less number of bioagents is required to release at the given sites for research purpose. In such situation, arrangement of mass multiplication required for huge number of bioagents will not be cost effective and feasible. Therefore, technique were developed to mass rear the bioagent in iron/wooden cages outside the laboratory which is a cost effective, labour saving mass multiplication technique.

The Technology: The cages can be made of wooden or galvanized sheets. The three sides and top should be covered with wire-mesh. A galvanized sheet tray having 8 cm depths should be fitted at the bottom of the cage. A few small holes can be made at the bottom to drain off the excess water. The tray is filled with soil and used for planting *Parthenium*. Iron cage has superiority over wooden cage in durability. These can be kept outside the house or laboratory and maintenance is easy. Iron cages may be made of different size as per need and available space. *Parthenium* can be grown in these cages either from the seeds or by transplanting. Small *Parthenium* plants, uprooted from the infested place can be transplanted in the cage. After establishment of sufficient *Parthenium* plant in the cage, 2 to 4 pairs of *Z. bicolorata* can be released as per cage space. Soon females will start egg laying and life cycle of the beetles start inside the cage. This method requires less attention than rearing in the small plastic cages/jars, tubs and buckets. Rearing in such situation is more hygienic than the plastic cages/containers. From one 2 x 2" size cage, about 200-300 beetles can be obtained in a period of one month.

Multiplication of Mexican beetle in iron cages

The benefits: From this method, disease free and robust bioagents can be obtained easily at the cost of 60-70 paise/beetle. This method is beneficial if beetle requirement is not high and less number of bioagents are required for releasing, augmenting or research purposes.

Precautions: After one week of egg laying, adults should be removed to avoid eating of leaves of *Parthenium*. Completely eaten plants should be replaced time to time to provide fresh food to the grubs. While transplanting fresh *Parthenium* in the cage, minimum disturbance to soil should be made. Sufficient soil moisture has to be maintained during beetles' rearing. However, care should be taken to prevent over watering especially when the insect is under pupation.

4.8 Mass multiplication of Mexican beetle in mosquito nets

The problem: *Parthenium* in India has emerged a notorious weed causing many disease in men and animals besides depleting crop production and biodiversity. Although, it is seen to grow throughout the year on the bank of rivers, water channels, open drainage channels, near hand pumps *etc*. But, maximum germination (70- 80%) of *Parthenium* takes place during rainy season. Among several methods, biological control has been considered an environment friendly method. This method requires cost effective mass multiplication of the bioagents. Several cost effective mass multiplication methods were developed depending on the requirement of the bioagents. It was realized that if one to five thousand beetles are required, mass multiplication in plastic tubs and wooden cages are not cost effective. For such situations, mosquito net method was developed.

The technology: Good number of beetles can be reared in mosquito net method. In this method, standard or big size mosquito net, generally used in house, can be used for multiplication purpose. In market, 1 x 2 m or 2 x 2 m size mosquito nets are easily available. For outdoor multiplication, according to the size of mosquito net, a *Parthenium* bed should be made. The *Parthenium* seeds collected from dry *Parthenium* plants can be broadcasted to germinate. For quick preparation of *Parthenium* bed, small plants of 6-8 inch size should be uprooted from the infested places. These uprooted plants, may be transplanted in the sufficient wet soil. The transplanted *Parthenium* will be well established within 3-6 days. After well establishment of plants, the mosquito net can be erected over the *Parthenium* bed with the help of bamboo/iron poles. In 1 x 2 m mosquito net, 8-10 pairs of beetle should be released. Plants should preferably be in the pre-flowering stage at the time of releasing the adult beetles for egg laying. A cage covering 2 m^2 area, can accommodate up to 300-500 plants of *Parthenium*. Females will start egg laying soon after their release. From one 1 x 2 m mosquito net, about 1000-1200 beetles can be obtained in a period of one month and from 2 x 2 m net, about 2000 to 2500 beetles can be obtained after one month.

Mass multiplication of Mexican bettle in mosquito net cages.

The benefits: This method is very cheap and requires less attention than methods of multiplication in buckets, tubes or cages. The advantage of this method is that the rearing is done almost in natural conditions; therefore, there is no need of pre-acclimatizing the beetle before releasing at other sites. The per beetle cost of multiplication was estimated at 40-50 paisa only. Nylon mosquito net can be used several times for rearing the beetle.

Precautions: As thumb rule, old and eaten *Parthenium* plants should be replaced regularly with the fresh plants as and when required. Watering should be made on need basis. To avoid predation by predators, mosquito net should be buried in the soil from the outer side. A walk in type door arrangement with 'velcro' will be highly beneficial to enable easy entry inside the cages for replacing old plants and collection of emerged beetles. This will also help to check the entry of harmful creatures like snakes *etc.* inside the cages.

4.9 Mass multiplication of Mexican beetle in net house

The problem: Biological control of *Parthenium* requires the cost effective and need based technology of mass multiplication of bioagent Mexican beetle (*Zygogramma bicolorata*). In India, *Parthenium* has also been considered a problematic weed by the government and cities and villages authorities. Many times, government agencies wish to release the bioagents for biological control of *Parthenium* for general benefit of farmers and public. In such situations, large number of releases of bioagents are required during rainy season for which mass multiplication of bioagents is one of the essential prerequisite. It was realized that if several thousand (10,000 to 50,000) of Mexican beetles are required for release in the infested area, mass multiplication in tubs, wooden/iron cages and in mosquito nets are not cost effective. For such situations, mass multiplication technology of Mexican beetle in pre-fabricated big net houses covered with mosquito wire mesh or nylon mosquito net was developed.

The Technology: Large number of beetles can be reared and multiplied in net house conditions. A net house having nylon or wire net of 30-40 meshes size is suitable for this purpose. In open field, desired net house can be made with the help of poles and purlins used to erect tents in marriage function. After erecting poles over the field, nylon mesh may be covered all around the poles to make it a temporary big net house of desired size. An entry door may be made with the help of velcro. *Parthenium* can be grown by seeds or by transplanting method in the net house. Cow dung and DAP should be mixed in the soil as per field practice to grow the crop. This will help for fast germination and good growth of *Parthenium*. The seed collected from the dry *Parthenium* from the infested side should be scattered over the soil in the net house followed by irrigation. Soon, *Parthenium* plants will germinate. When, the size of *Parthenium* plants become 12-15", beetles should be released in pairs for egg laying purpose as per the net house space. As a standard practice, 8-10 pairs of beetles should be released in a 2 x 2 meter space. Watering should be done as and when required. Automation of watering in net house in the form of fogging or drizzling will be highly useful in mass multiplication of the beetle to cut the cost of multiplication. Grubs will hatch from the eggs and will start their development on the plants as occurred in natural conditions. In a net house of about 10 x 20 m size, 15,000 - 20,000 beetles can be obtained within a period of one month. Multiplication may be continued in the net house by sowing fresh seeds or transplanting fresh *Parthenium* plants on depletion of previously grown *Parthenium*.

Mass multiplication of Mexican bettle in mosquito net cages.

The benefits: This method is cost effective and less labor intensive owing to growing of *Parthenium* in comparatively large area. This method may cost only 25 to 30 paisa/beetle. The natural vigour and resistance of bioagent is maintained due to rearing in almost natural conditions.

Precautions: Beetles for egg laying should be released when, plants become sufficient large having good biomass. If these are released on new emerging plants, beetles will soon eat them and will hamper the multiplication process. As thumb rule, new emerging beetles should be collected to avoid over crowding and depletion of food. Sufficient *Parthenium* plants should be maintained in the net house for continuing the mass multiplication of bioagent.

4.10 Mass multiplication of Mexican beetle in open field

The problem: Biological control using Mexican beetle has emerged as one of the most successful methods to lower down the *Parthenium* menace in India. Sometimes, several lakh beetles are required to release on demand by the government against. It was realized that if several lakh Mexican beetles are required to release in the infested area, mass multiplication even in net houses will not be sufficient to meet out the demand. It is well known that Mexican beetle works nicely during rainy season when conditions for its reproduction are most suitable. To meet out such emerging situations, technology of mass multiplication of Mexican beetle during rainy season was developed in open field conditions and on the naturally *Parthenium* infested sites.

The Technology: Large number of beetles can be reared and multiplied in open field conditions during rainy season without using net. A field can be prepared as done for raising the crop by sowing the seeds. Recommended dose of fertilizer should also be added in the field. *Parthenium* seeds should be collected in advance from the *Parthenium* infested sites. These seeds should be broadcasted in the field followed by irrigation. Soon, *Parthenium* plants start germination. On reaching the size of 12-15 inch, Mexican beetle at the rate of 2-4 pairs/m^2 can be released in the field for egg laying purpose. In large number required beetles can be released of 5000/ha. Soon, development of beetles will start from the eggs in the natural conditions. New adult beetles will start to emerge from 28 days onwards after egg laying. Newly emerged beetles should be collected for release in the newer area. From this technique, about 1 to 2 lakh beetles can be multiplied from one hectare area. Mexican beetles obtained from mass multiplication in iron cages or mosquito net in less numbers may be released on the succulent growth of naturally *Parthenium* infested sites after commencement of the rains. These natural *Parthenium* infested sites, will act as breeding sites for multiplication of the bioaget. From these sites, adults may be collected after one months as per need. Such sites may be developed as per the demand of bioagent for further release.

Mass multiplication in open field conditions and close up of the beetle

The benefits: This technology is very cost effective for mass multiplication in natural conditions and may cost only 15-20 paisa/beetle. *Parthenium* may be grown in poor soil or undulated fields for mass multiplication of bioagents. Even natural infested sites on the commencement of rains may be utilized for mass multiplication of beetles. The natural vigour and resistance of bioagent is maintained due to rearing in natural conditions.

Precautions: As a thumb rule, adult beetles for egg laying should be released when plants become sufficient large having good biomass and new emerging beetles should be collected immediately to avoid over crowded and depletion of food. It was seen that if fields are nearby lighted area during night, newly released beetles are attracted towards light resulting poor egg laying on the multiplication sites, hence lighted area should be avoided for multiplication.

4.11 Activity enhancement of Mexican beetle by diapause aversion

The problem: Biological control using exotic bioagent imported from Mexico has been emerged as one of the most economic and practical way of managing *Parthenium*. Diapause is a unique feature in insects to avoid unfavourable conditions by burying themselves amidst the leaves' litter or in soil and to become inactive. Adults of *Z. bicolorata* have been seen to undergo diapause in the soil under field conditions even under favourable conditions by burying themselves over an extended period of time between July and December. It was observed that about 60% adult population of Mexican beetle developed during August died naturally by the end of November. Out of 40%, 30% population enter into soil below 2-6 cm. Remaining 10% population remained without burying themselves into soil but their activity remain very low. Generally, these beetles do not lay eggs and their movement also remain limited. In majority of the cases, such low activity-showing beetles became active with the increasing temperature in February–March when they again start egg laying. Adults having inherent cues may enter into diapause only once during its life time. *Z. bicolorata* is a multivoltine species and generations cannot be differentiated under field conditions. *Parthenium* has the capacity to grow any time with the availability of moisture. This period of absence of the beetles from field becomes a boon for the weed as the weed reaches its flowering and seed production stage by the time the insect population is large enough to cause defoliation of the weed. Therefore, if diapause in *Z. bicolorata* could be averted or broken early, the biocontrol potential of bioagent will be improved appreciably.

The technology: Newly emerged beetles of *Z. bicolorata* aged between 1 to 7 days may be collected from *Parthenium* infested fields during rainy season or during February-March when their population remains high in the field. Laboratory reared newly emerged beetles may also be used to break the diapause. Newly emerged beetles may be easily identified by their soft body and dull brown colour. The diapause chambers may be made of plastic boxes, with 25 to 50 cm diameter with minimum 8 cm depth, with wire mesh lid on top and sides to enable good aeration. These boxes may be filled up with soil (pre-sterilized in autoclave) up to 4 cm deep and the soil should be moistened. Each diapause chamber may house 50 to 100 beetles depending on the size of the chamber. Fresh *Parthenium* twigs should be provided as food. These chambers may be exposed to 35 °C or 10 °C in Biological Oxygen Demand (BOD) incubators at 60-75% RH for 7 and 15 days, respectively to obtain non-diapaused beetle. Beetles that enter into soil may be considered to be in diapauses while those survived beetle remain outside may be considered as non-diapaused beetles.

Effect of temperature on diapause behaviour of *Z. bicolorata*

Beetles stored at temperature			Percentage of beetles
10 °C	35 °C	26 °C	
86.3[a]	41.3[b]	13.3[b]	Non - diapaused
9.8[b]	1.8[c]	63.8[a]	Diapaused
3.5[b]	57.0[a]	23.0[b]	Mortality
6.6	5.9	10.3	LSD (P =0.05)

The benefits: Diapause is a disadvantage to the biocontrol potential of the beetles. Thus diapause aversion or decreasing percentage of adults entering diapause can be of great advantage in enhancing their effectiveness in biological control. These diapause averted beetles can be made active all the year taking into consideration the other ecological factors. These beetles can be used for large scale augmentation purposes at sites of less abundance or when beetle populations remain low due to increasing numbers of beetles entering diapause.

Precautions: Newly emerged beetles should be kept initially at 30 °C for 1 day then the temperature should be gradually increased to 35 °C by the third day. The gradual increase in temperature is done to acclimatize the beetles to high temperature. A constant relative humidity of 60±2% should be maintained as humidity levels above this may be detrimental to the beetles at high temperature. After exposing them to 7 days in high temperature and 15 days to low temperature, they should be brought back to room temperature (26±1 °C). These beetles may be used for augmentation or release purpose during summer or winter season at appropriate place of *Parthenium* infested area. The beetles exposed to low temperature appear soft and white for more than 30 days of age like the newly emerged beetles. There may be delay in hardening of cuticle in these beetles with 86.2% averting diapause.

4.12 Safe and longer storage of Mexican beetle culture

The problem: Biological control by Mexican beetle *Zygogramma bicolorata* has emerged as one of the most successful approaches to contain the menace of the weed in non-cropped area. Many times, being the Directorate of Weed Research, scientists and research scholars working on the research aspects of biological control of *Parthenium* request us for the culture of Mexican beetle to start the work. In nature, Mexican beetle may be abundantly occurring during rainy season but it is hardly noticeable during winter and summer. In this season, only experts may find the beetles at the hidden niche. It was also experienced that there was high mortality of collected adults beetles if kept in room temperature or in BOD at 27 ° C and 70% relative humidity for longer time. In such temperature, they require fresh food to maintain the culture. They also start breeding due to which hatched grubs also feed the foliage and excrete, hence requires more attention. Therefore, it was realized that, availability of healthy culture is necessary on demand for initiating mass multiplication of the bioagent. To overcome this problem, technology was developed to maintain the healthy culture of Mexican beetle in BOD for longer time.

Technology: Newly emerged adults of *Z. bicolorata* aged between 1 to 10 days should be collected from *Parthenium* infested fields during rainy season or during February - March when they are abundantly or sufficiently found in the field. These should be kept in plastic jars having sufficient aeration on the fresh bouquet of *Parthenium* leaves. The jars having adults beetles with bouquet should be kept in BOD maintained at 10 ± 1 °C. The bouquet should be changed once in 10 days only. In this method, there is low mortality (3.6%) of beetles besides less tendency to go under diapause (9.8%). About 86% beetles do not diapause and remained live for long time. No significant difference in the fecundity of the females stored from 0 to 6 months was observed. When these adults are kept at room temperature after taking from the BOD and provided food, they soon become active and start egg laying and breeding. Through this method, bioagents culture may be maintained throughout the year, particularly during winter and summer season when they are not easily available in the nature.

Months	Beetles kept at 108°C (in days)	Beetles kept at 26°C (in days)	Total longevity (in days)	Fecundity (per female)
0	0	122.3^{ab}	122.3^{e}	2333.3^{a}
1	30	125.0^{a}	155.0^{d}	2444.3^{a}
2	60	104.0^{ab}	164.0^{d}	2485.7^{a}
3	90	118.3^{ab}	208.3^{c}	2396.3^{a}
4	120	101.3^{b}	221.3^{c}	2200.7^{a}
5	150	102.0^{b}	252.0^{b}	2263.0^{a}
6	180	74.7^{c}	254.7^{b}	2244.7^{a}
7	210	50.7^{d}	260.7^{ab}	1100.3^{b}
8	240	22.7^{e}	262.7^{ab}	149.0^{c}
9	270	9.3^{f}	279.3^{a}	42.0^{d}
LSD (P=0.05)		1.1	0.6	3.1

The benefits: Low temperature storage reduced the percentage of adults entering diapause with no mortality and after storage, coupled with no reduction in fecundity. This method may provide healthy culture of bioagent throughout the year on demand.

Precautions: It should be ensured that temperature does not fall and rise from the prescribed temperature. If temperature increase, beetle become active and start feeding and excrete. Proper cleaning of jars should be done during changing of leave bouquet. Leaves in bouquet should be wrapped with wet cotton followed by aluminum foil to give moisture to leaves for longer time

4.13 Ideal age and food for transportation of Mexican beetle

The problem: *Parthenium* has been categorized as one of the most notorious terrestrial weeds in India. Among various control methods, biological control through Mexican beetle, *Zygogramma bicolorata* has been emerged as one of the most economical and practical ways to control *Parthenium*. Seeing the damage potential of this bioagent, there is great demand by people from different parts of country to receive the nucleus culture of Mexican beetle to release in their area. During initial phase of country wide introduction of *Z. bicolorata*, we used to send bioagents' consignment to various agencies like Krishi Vigyan Kendra (KVKs) situated all over India, centres of All India Co-ordinated Research Project on Weed Management (AICRP-WM), municipalities, NGOs, farmers and interested persons by private courier and government postal services. During our initial effort of dispatch method, we used to keep green twigs of *Parthenium* with leaves and a wire mesh window on the lid of container, which was packed in the cardboard box having wire mesh lid over the containers. In that method heavy mortality of bioagents (up to 80%) used to be recorded at the consignee end if consignment reached after six days. Therefore, technology was developed to find out the suitable age of the beetle and the food to be kept in the container so that maximum numbers of beetles reach alive at its destination even after 7-8 days journey period.

The technology: If consignment is likely to reach within 2 days, we can keep inside the containers, cuttings of thick *Parthenium* twigs with succulent leaves or without leaves or twigs of upper flowered part as food. In two days period, low mortality occurred in all types of food and resting material but high mortality of beetles starts by 3rd day onwards. Succulent leaves may be kept even if the consignment is likely to reach on 5th day after packing. In case, consignment is likely to take more than 5 days, mixture of cuttings of green thicker *Parthenium* stems and tender *Parthenium* twigs of upper flowered portion should be placed inside the well aerated plastic containers. In the containers, we should keep beetles having age group between 10-15 days because young beetles are highly susceptible to death during transition than the old beetles as up to 30% newly emerged beetles died within 24 hours while 85% died by 4th days. Only 15% beetles survived up to 7th day. In a container of 500 ml volume, 500 adult beetles may be housed by this method.

Cutting and keeping green twigs without leaves in the aerated containers for safe dispatch

Progressive mortality (%) in mixed population of Mexican beetle at different days kept in containers having different type of *Parthenium* twigs with or without leaves.

Treatment	% progressive mortality after days						
	2nd	3rd	4th	5th	6th	7th	8th
Green thick twigs with succulent leaves	4.66	16.66	32.00	32.33	59.00	81.00	89.33
Green thin flowered portion twigs with leaves	6.00	21.00	44.00	47.33	54.66	71.33	78.66
Semi-green twigs without leaves	4.66	27.66	30.33	30.00	66.00	64.66	75.33
Green thick stem and thin flowered part twigs without leaves	12.66	24.00	30.66	29.00	40.66	44.00	51.00
LSD (p=0.05)	6.84	13.57	13.65	7.25	13.73	15.35	4.20

The benefits: Low mortality of beetles occurred following these methods. Age group between 10-15 days will be more fertile to produce eggs than the old beetle.

Precautions: No significant difference is found in mortality bioagent up to 4 days of packing using any type of food representing mixed population of adult beetles. But in general, parcel reach after 4 days to the consignee, therefore, as a thumb rule, we should keep green thick stem and thin flowered part twigs without leaves. Study suggests that only old beetles having age group between 10-15 days should be sent for transition by the postal services. Newly emerged and young beetle can easily be recognised by the dull dusky colour and softness of the body.

4.14 Development of packing boxes for transportation of bioagents

The problem: *Zygogramma bicolorata* has emerged as one of the most economical and practical ways to control *Parthenium*. Seeing the damage potential of this bioagent, there is great demand by stakeholders from different parts of country to receive the nuclear culture of Mexican beetle to release in their area. There used to be high mortality of bioagents during transition sent by the conventional packing (Fig.1). There is no provision in postal department to send such research/bioagent consignment separately with utmost care. All such consignment are packed in smaller bags in postal department. Further, many such smaller bags with tightened mouth are packed in larger bags. Such bags are thrown up and down many times during transportation and are put in heap one upon another. Sometimes, curious people tear the packing box and container inside by inserting sharp object to see what is inside of this peculiar type of packing. A few, we received undelivered consignment parcel back in very shabby conditions. In such situations, there is high probability of mortality of live bioagents, if not packed properly in well aerated strong enough containers and packing box. Therefore, technology was developed to make light, strong and well aerated container and packing box so that maximum number of bioagents reach alive at its destination through postal services even after 7-8 days journey period.

Conventional container having wire mesh on the lid only and small holes on the cardboard box. An undelivered parcel which suffered badly during transportation

The technology: Plastic containers made by light and strong plastic were chosen for this purpose so that they could bear the jerks and pressure during transportation. Two windows opposite to each other were cut on the mid part of the box with the help of soldering unit to facilitate aeration from side also. The fine wire mesh made up of steel or brass may be affixed with the help of soldering unit. The plastic lid is also cut by soldering unit and replaced with wire mesh of iron, steel or aluminium as per the availability. Iron mesh is cheap than the steel and aluminium but get rusted in due course. Thin upper side twigs of green *Parthenium* without leaves can be cut according to the size of container and may be placed inside. Beetles ready for dispatch should be put inside this container. In one kg capacity container, 500 to 700 *Z. bicolorata* may be housed. Beetles soon climb and clinch the twigs and distribute themselves in suitable niche inside the container.

Fitting of brass or soft iron wire mesh windows on side walls of the container and on the lid of the plastic containers with the help of soldering unit. The side and top view of cardboard box with iron wire mesh to prevent the damage to container kept inside with bioagent

This container having beetles/bioagents required to be put inside of a strong, light and well aerated cardboard box which should tolerate the jerks and pressures during transportation of bioagents. Therefore, suitable big holes were made on two side walls of cardboard sheets opposite to each other. A piece of iron wire mesh according to the area of holes was inserted between the layers of cardboard sheets to prevent the insertion of object from the open holes in cardboard sheet.

The benefits: The plastic containers designed in this way are light, strong and allow good aeration from side besides protecting it from direct hit and pressure. The beetles' mortality in these containers was found reduced by 63% than the conventional containers and packing during 7 days' journey. This type of containers may be prepared by ourselves using old/new plastic containers and cardboard sheets of packing in which our household or laboratory material comes.

Precautions: The containers having beetles should be placed inside the cardboard box in such a way that the windows with wire mesh remain opposite to each other. This is very necessary for proper aeration otherwise there will be no benefit to reduce the mortality of biogents. We should keep only green *Parthenium* twigs without leaves inside the containers for keeping healthier environment.

5.0 Weed utilization

5.1 Bio-ethanol production from water hyacinth

The problem: Water hyacinth (*Eichhornia crassipes*) is a fast growing perennial aquatic plant, widely distributed throughout the world. It forms large, dense mats on the surface of water, disrupting the flow of water, reducing the oxygen supply for fishes, hindering the movement of boats. Since herbicides cannot be sprayed in aquatic systems and mechanical removal is costly, their utilization would be an alternate strategy of management. Water hyacinth is a lignocellulosic source for bio ethanol production and can be fermented using yeasts, after conversion of the hemicelluloses to simple sugars.

The technology: With slight modifications in the standard procedures, the technique is described here. Fresh water hyacinth plants are cleaned, chopped into small pieces and dried in hot air oven at 70°C for overnight. The dried material is powdered and 1% sulfuric acid is added to 50 grams of the powder. Final volume is made to 500 ml with water. The mixture is autoclaved and then allowed to cool down. The resulting hemicellulose acid hydrolysate is filtered to remove the unhydrolysed material. The filtrate is again heated to 60 °C and neutralized with solid sodium hydroxide to make pH 9.0. A pinch of calcium hydroxide is added to detoxify the hydrolysate for growth of yeast. 10 grams of neopeptone is added to the above solution to reduce the pH to 5.5. This solution is filled up with 500 ml water and autoclaved. Cool the flask to room temperature and add yeast (*Pichia stipitis)* and the flask incubated at 30 °C for 3 weeks. The solution is then distilled to obtain pure bio-ethanol. Three isolates of lignocellulose decomposing native yeast have been isolated from the aquatic bodies around Jabalpur and maintained at DWR. *Pichia stipitis*, is a very effective isolate of yeast to ferment the detoxified hydrolysate and can be obtained from NCIM, Pune.

Chemical composition of water hyacinth

Sl. No.	Components	% composition
1.	Lignin	12
2.	Cellulose	26
3.	Hemicellulose	37
4.	Ash	21

Matured water hyacinth plant and colonies of yeast isolated from water hyacinth

The benefits: Production of bioethanol from water hyacinth helps in the effective usage of the vast biomass that is generated after clearing the infested water bodies. Bioethanol production is a priority objective for the Government of India, to reach its self-sufficiency in the generation of renewable energy resources. Raw materials other than the food plants would help in preventing the problems of food fuel conflict in the future.

5.2 Compost production from weed biomass and parthenium

The problem: *Parthenium hysterophorus* commonly known as Congress grass, Carrot grass, *Chatak Chandni etc.* is a menace to farm land, human-beings, animals, environment and bio-diversity. During interaction with farmers, it was found that if they use the compost made from the weeds in general and *Parthenium* in particular by recommended NADEP or open pit or heap methods (farmer practice), there was more germination of that type of weeds in the field from which such compost was made. Although, it is always recommended by the experts to collect the *Parthenium* and other weeds' biomass before flowering for making compost either by NADEP or open pit method. But it is not practically possible to collect only flowerless weedy plants in general and *Parthenium* in particular as all the stages of *Parthenium* are available at any time due to non dormancy of seeds which may germinate on the availability of water. It was observed that in NADEP methods, all the weeds seeds are not killed, particularly on the boundary sides of the NADEP pit. Therefore, technology of pit method was developed for the safe use of compost made from the weeds in general and *Parthenium* in particular.

The technology : Make a pit of 3′x 6′x10′ feet (depth x width x length) at a place where water dose not stagnate during rainy season. Pit size can be increased or decreased on the availability of biomass but depth cannot be compromised. If possible, cover the surface and side walls of the pit with stone chips. If stone cheep are not available, make soil surface compact. Arrange about 100 kg dung, 10 kg urea or rock phosphate, soil (1-2 quintals) and one drum of water near the pit. Collect all the *Parthenium* and other uprooted weedy plants during process of weeding from the field, bunds, road side and nearby area. Spread about 100 kg of *Parthenium* on the surface of pit. Over this, sprinkle 500 g urea or 3 kg rock phosphate. Make dung slurry by adding 5-10 kg dung in 25 litter water and sprinkle over the weed biomass. If possible add *Trichoderma viridi* or *Trichoderma harziana* (kind of fungi cultured powder) in the amount of 50 g per layer. All the above constituents will make one layer. Like first layer make several layers till the pit is filled up to one feet above the ground surface. Fill the pit in dome shape. While making layers, apply foot pressure to make weed biomass compact. If there is no sufficient

soil with weeds roots then add 10-12 kg of loamy soil on each layer. When pit is full with above described layers then cover it with mixture of dung, soil and husk. After 3-4 months, we can get well decomposed compost. We can get 37-45% of compost from 37-42 quintals of weed biomass. Sieve that compost with 2x2 cm size mesh. In *Parthenium* weed compost, nutrients were twice more than that of ordinary compost and almost equal to vermi-compost.

Type of bio-fertilizer	Nutrient (%)				
	N	P	K	Ca	Mg
Parthenium compost	1.05	0.84	1.11	0.90	0.55
Vermic ompost	1.61	0.68	1.31	0.65	0.43
Farm yard manure	0.45	0.30	0.54	0.59	0.28

The benefits: Weed compost including of *Parthenium* is a type of bio-fertilizer which has no harmful effects on crops, human-beings and environment. Weed compost is an eco-friendly biofertilizer that can be made by low cost inputs and application of it in crop fields will increase the fertility of soil. During the process, the temperature rises 60-70 °C due to which seeds are killed.

Precautions: Avoid using flowered plants having seeds. Pit should be in open and shady upland. If fresh germination of weeds near the pit is spotted where weed was collected to fill up the pit, destroy them otherwise they may contaminate the compost after flowering. Check the moisture level of compost. If there is dryness in the pit, make a few holes and pour water in the pit and close the holes. While it may take about three to four months to prepare the compost in a warm climate, in cold regions it can take more time.

5.3 Accelerated composting of weed biomass

The problem: Productivity of Indian soils has started declining in their nutrient status and organic matter, which is one of the major reason for the declining fertility of crops. This lack of organic matter is a result of over exploitation of soils through intense cultivation and without recycling of organic matter back to soil over the last many decades. Green manuring is an important method of recycling organic content to the soil. However addition of fresh green manure does not provide the full benefits to the soil and also carries the risk of spreading the plant pathogens. Composting of green biomass using the cellulose degrading fungus *Trichoderma viride* is an effective way to fasten the process of decomposition, enrich the compost with beneficial microbes and decrease the population of pathogenic microbes. Currently farmers burn the weeds collected from their field or feed a part of them to cattle or apply as fresh in the fields which takes longer time to decompose.

The technology: The technology described here explains the accelerated composting of weed biomass with the help of *T. viride* under the field conditions. *T. viride* was multiplied in 2 kg broken boiled rice for about 10 days. Entire mat of fungi along the rice grains was dried and grinded. This was used as fungal inoculant for application in the compost pit. A shaded, elevated area of size 5 m X 2.5 m in the corner of the field is selected. Weeds are segregated into cellulosic (example *Parthenium, Alternanthera*, water hyacinth) and leguminous (example *Medicago*). Ten kg of weeds are finely chopped and placed in alternating layers of cellulosic and leguminous weeds one over the other in the form of a pile. In between two layers of weeds, cow dung slurry and *T. viride* were gently and uniformly sprinkled. Up to six such layers can be made and the top layer was covered with a gunny bag to maintain moisture. Water is sprinkled gently and there should not be excess moisture. The pile was maintained for 45 to 60 days and turned upside down after 15 to 30 days to provide aeration. More

Compost under preparation in the field

composting period is required for fibrous woody weeds. At the end of the period the compost material becomes dark brown in colour. *Trichoderma viride* is available with the Directorate of Weed Research, Jabalpur.

Nutrient status of the compost of weed biomass

Sl. No	Particular	% composition
1	Organic carbon	50-55
2	Total nitrogen	1.87-2.50
3	Phosphorus	0.61-0.40
4	C: N ratio	14-16:1

The benefits: Getting compost materials earliest is pre-requisit, by using *T. viride* fungus. The decomposition time can be minimized and compost can be used in the field to increase the organic matter content. The technology is cheaper and can be practiced easily by farmers.

tanks. In order to discharge the filtered water through gravity flow on the irrigating field, each pair of tank is constructed at different height. The inlet and outlet pipes of the each pair of tank is connected in such a way that the water flow from sedimentation tank to the first treatment tank followed by second and third tank by gravity flow.

Changes in contaminants in drain water treated in constructed wetland system using *Arundo* sp.

Treatments	pH	TSS mg/l	NO_3 ppm	PO_4 mg/l	Cu ppm	Ni ppm	Zn ppm	Mn ppm	Bacteria Cfu/ml
Sedimentation	7.35	238	19.50	2.72	0.388	0.269	0.43	1.78	6.5×10^4
Tank I (Hydroponics)	7.25	152	9.40	1.90	0.340	0.192	0.41	1.17	24×10^3
Tank II (Surface)	7.21	134	8.79	1.71	0.187	0.156	0.35	0.93	14.7×10^2
Tank III (Sub-surface)	7.16	89	2.29	1.56	0.141	0.112	0.12	0.69	8.1×10^2
Waste water (Untreated)	7.50	249	20.30	2.91	0.468	0.305	0.54	1.83	18.7×10^4
Efficiency (%)	-	64.0	88.4	46.3	69.4	62.4	78.0	61.7	

Treated and untreated water

Treated water being used for irrigation

The benefits : This technique is applicable for remediation of contaminants such as heavy metals (Cu, Mn, Zn, Pb) in low lying waste carrying drain. Plant assisted bioremediation system has an application for irrigation use in agriculture industry. The system retain P in filtered water with less hydraulic retention time of 1-2 days. The system saves energy by basket flowing treated water from outlet of treatment tanks through gravity flow discharging directly on the field plots for irrigation. In the long run system is useful for protection of field soil from heavy metal contamination. The system can treat 10000 litres of water daily. It time and energy saving system and further no risk of clogging of the treatment zone as usually occurs in case of earlier water purification system.

The harvested wetland plants can be used for basket, mat making and also for fuel purpose. Clear and odorless water has no risk of human contact while irrigation.

6.2 Phyto remediation technique to decontaminate sulfosulfuron and its metabolites

The problem : No doubt herbicides are boon to control weeds to a great extent but residual herbicides may persist longer than desired and harm the rotational crops or may enter the food chain relentlessly and pose a serious threat to the human health and the integrity of ecosystems worldwide. Sulfosulfuron and its metabolite such as sulfonamide and desmethyl sulfosulfuron were found persisting in soil and affected growth of rotational pea and lentil in our study conducted for 3 years in a sandy loam soil under the project 'Behaviors of sulfosulfuron in sub soil under the influence of wheat cropping system-identification and quantification of potential transformation products responsible for the phytotoxicity and their bioaccumulation in fish'. Thus a phytoremediation techniques was developed to phytoremediate toxic level of sulfosulfuron and its metabolites in the soil.

The technology: A phytoremediation is a set of innovative technology for environ-mental cleanup that involves advantage of the unique extractive and metabolic ability of plants. This phytoremediation techniques involved deep plouging of the affected field followed by growing *Sesbania* (*Dhaincha*) plants as green manuring and of FYM at 10 ton/ha rate. This technique was validated by detoxing sulfosulfuron and its metabolite, sulfonamide and desmethyl sulfosulfuron which were found most persisting in the soil of wheat field and affected growth of pea and lentil at higher doses. Pea was found more sensitive towards the sulfosulfuron residues followed by maize, barley and sorghum. Deep ploughing, followed by growing of *Sesbania* plants in affected field along with application of farm yard manure enhanced the degradation of persisting sulfosulfuron residues and its metabolites. In deeper soil profile, once the sulfosulfuron had been taken up by growing *Sesbania*, plant biomass was disposed off in the soil. Disposing off and mixing of the grown up *Sesbania* plants in the soil improved the soil health also.

The possible mechanism in the phytoremediation technique involved the uptake, transportation and detoxification of sulfosulfuron from soil by growing green *Sesbania* which absorbed sulfosulfuron primarily by root. Penetration to roots occurred mainly by simple diffusion through unsuberized cell wall, from which herbicide reach the xylem stream. Upon entering to the symplast sulfosulfuron was modified by, reduction or hydrolysis reactions followed by conjugation. Conjugated molecule then sequested as part of insoluble cell wall polymers or in cellular compartments such as vacuole, where they can be metabolized further to simple molecules and further ideally to CO_2.

The benefits: This technique presents clear benefits such as wide applicability, environmental value and cost-effectiveness. While herbicides or other organic pollutants can be degraded to less toxic forms by plants, or even mineralized. Deep plouging of contaminated soil along with application of *Sesbania* and FYM was found effective to decontaminate sulfosulfuron and its metabolites under field conditions. This phytoremediation technique may also be equally useful to mitigate adverse effects of other ALS inhibitors.

crop without phytoremediation;

crop after phytoremediation technique

6.3 EDTA assisted phyto-extraction of cadmium

The problem: Soil contaminated by cadmium which mainly originate from non-ferrous mineral processing and smelting activities, has become one of the major environmental concerns and has profound effects on health of human beings. Numerous struggles have been done to invent processes for the restoration of affected soils, comprising *ex-situ* method of washing of soil with physiochemical technique, and immobilization of heavy metal toxins as *ex situ* method. In recent times, the phyto-extraction of metal contaminants from disturbed soils has been fascinated due to its lesser cost of implementation than other techniques. Usually, it is a determining factor for phytoremediation that the plant has the ability to cultivate a large biomass with high contents of toxic metals in its shoots. However, for certain metals such as cadmium, lead which are less bio-available in soil and have limited translocation ability from root to shoots are major obstacles in the phyto-extraction process experienced by most of plant species.

Large proportion of environmentally hazardous heavy metals, such as Cd, Pb, Cu, Zn remains sorbed to solid soil constituents. To acquire these 'soil-bound' metals, phyto-extracting plants have to mobilize them into the soil solution. This so-called mobilization of 'soil-bound' metal can be accomplished by way metal-chelating molecules which can be secreted into the rhizosphere to chelate and solubilize 'soil-bound' metal.

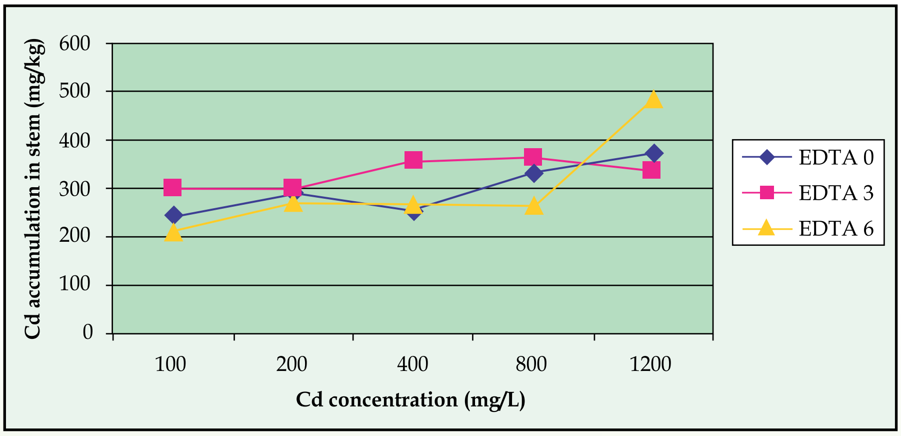
The technology: *Arundo donax* was exposed to different concentrations of cadmium as 0, 100, 200, 400, 800 and 1200 mg/L with ethylene diamine tetra-acetic acid (EDTA) applied at 0, 3 and 6 ppm found no adverse effect on growth of the weedy plant. Further, results are as follows:

- The cadmium accumulation in shoot was increased with increased concentration of cadmium. The higher cadmium accumulation was observed in root than shoot part of *Arundo donax*.
- The addition of EDTA at 3-6 mg/L further enhanced the cadmium accumulation in shoot. However, the EDTA at 6 mg/L enhanced 2- 3 times higher cadmium accumulation in root as compared to control.
- The order of cadmium accumulation was root > stem > leaf. As far as plant parameters is concerned no adverse effect of cadmium levels were observed on height and chlorophyll reflectance by leaves of *Arundo donax*.

- The concentrations of Cd in giant reed shoots were significantly increased when applying EDTA at lower levels. Combined use of EDTA alongwith high biomass producing *Arundo* has implication for phyto-extraction of cadmium contaminated sites.
- Use of EDTA along-with potential plant species may be applied for increasing bioavailability of heavy metals like Pb, Cd, Fe, Mn.
- EDTA should be used at optimum range of 3 mg/L. Avoid excess use of EDTA in soil for prevention of its loss in soil due to excess leaching especially in sandy soils.

Treatments	Cd (mg/kg DW) in different parts of *Arundo donax*	
	Stem	Root
Cd (mg/L)		
100	249.9	340.1
200	285.6	1626.5
400	292.2	2319.1
800	320.7	3481.1
1200	398.5	2485.7
LSD (P=0.05)	56.18	475.0
EDTA (mg/L)		
0	297.7	1095.7
3	331.9	2128.3
6	298.4	2927.5
LSD (P=0.05)	NS	445.4

Cadmium accumulation in different parts of *Arundo donax*



Enhancement of cadmium accumulation in shoot of *Arundo donax* by EDTA

6.4 Electro-bioremediation technique for heavy metal removal from polluted water. Electro-bioremediation technique has not been explained ?

The problem: Arsenic has been recognized as one of the most hazardous contaminant on Earth for over a decade. In some parts of West Bengal and Chhattisgarh, elevated arsenic concentrations are recently reported in ground water and soil. On continuous irrigation, the deposition of arsenic, cadmium, lead, chromium in soil add to the plant uptake giving vent for heavy metal entry into the food chain.

The technology: What is Electro-remediation ? Electro-remediation is the potential technique for arsenic removal. In our study, As was depleted from surface water by 90% by electrodes which was at par with combined effect of *Lemna minor*. *L. minor* is tolerant to arsenic. Besides *Lemna, Vetiveria zizinoides* and *Arundo donax* are other potential weed species screened recently for lead and manganese. The highest lead and manganese accumulation ratio shown by *Vetiveria zizinoides* (17.9, 6.38 respectively) and *Arundo donax* (12.5, 6.06 respectively) indicate its suitability for heavy metal phytoremediation.

The benefit: As far as higher content of arsenic, lead and manganese in ground and surface water is concerned, these fast growing high biomass plants combined with the electro-remediation can be beneficial for the metal contamination for the deeper layer of soils and for large volume of water.

Future outlook:

- Electro-bioremediation works for arsenic removal from the water.
- In electricity intensive areas, use of solar run batteries requires to be explored for running the electro-bioremediation.
- This technique along with deep rooted plant like *Arundo donax*, the roots of which grow more than 3-4 meters can work for the *in-situ* phyto-remediation of the ground water.
- Use of some bio-surfectants needs to be explored so that contaminants are transported to be made available for uptake by plant roots. The contaminants are concentrated within the plants, which can then be harvested and subsequently destroyed or treated to extract the absorbed chemicals.

6.5 Water weeds: toxicants extraction potential from polluted water

The problem: The environmental contamination by heavy metals is serious concern for surface water and ultimately animal and human health. Heavy metals, unlike organic pollutants cannot be destroyed or changed to forms that are harmless. Treatments for remediation of polluted waters, should therefore aim at extracting these substances from water and concentrating them before final disposal. Among macrophytes, weeds are more suited to remove heavy metals from water due to its fast growth resulting high biomass.

The technology: Phytoremediation of heavy metals using locally available potential weeds got more attention as remedial measures in present context. *Eichhornia crassipes*, the water hyacinth is a common aquatic weed in India which has the ability to take up and accumulate elements from water. On testing, the heavy metal uptake potential, *Eichhornia crassipes* accumulates higher concentration of cadmium, nickel, iron and manganese in its roots. *Alternanthera philoxeroides* accumulates higher concentration of heavy metals in shoots than root part of the plants. The extent of heavy metal accumulation ratio is given in the following table .

Heavy metal ratio between water and plant parts of aquatic weeds

Name of Weeds	Plant part	Heavy metals (mg/kg dw)				
		Ni	Cd	Cu	Fe	Mn
Alternanthera philoxeroides	Shoot	441	16.4	2448	3586	1666
	Root	149	12.0	2839	569	1196
Eichhornia crassipes	Shoot	56.8	1.44	552	2645	1014
	Root	253	21.8	2868	6576	6624

The benefits: Both weeds are suitable for the pytoremediation of multi-metal contaminated water. Due to fast growth of the weed species water hyacinth has utility for exploiting its root for removal of toxicants from lower layer of water, whereas alligator shoot can remove the contaminats from top layer of water. Thus both the plants species can be utilized for phytoremediation of multi-metal contaminated water.

7.0 Herbicide decontamination by microbes

7.1 *Aspergillus niger* as a potential bio-degradation agent of penoxsulam in soil

The problem: Herbicides account for the largest part of overall pesticide use in the world. Penoxsulam is a new post-emergence triazolopyrimidine sulphonamide herbicide registered for weed control in rice crop. Some studies demonstrated that sulfonamides in the soil may act as potential environmental hazards. The residues of sulfonamide herbicides have also been detected frequently in surface and ground water due to runoff and leaching after their application in /or near the agricultural fields. Like other types of ALS inhibitors, there is a possibility that penoxsulam or its transformation/degradation products may persist in the soil for a relatively longer period, and cause significant damage to rotational crops, and enhanced risk of environmental contamination. Microbial degradation is a one of the main route for ALS inhibitors detoxification in the soils. Thus the soil fungi was isolated and evaluated for degradation of unintended high spray of penoxsulam in soil.

The technology: *Aspergillus niger* was isolated and identified from rhizosphere of rice field where penoxsulam was applied to control annual and broad-leaved weeds. Degradation potential of *Aspergillus niger* was evaluated against duifferent doses of penoxsulam in soil and transformation products was identified by LC-MS/MS.

Degradation kinetics showed that degradation of penoxsulam in the soil followed by formation of several degradation products/metabolites by the fungus. Cleavage of the sulfonamide was found to be a major degrading pathway, yielded the corresponding sulfonamide and heterocyclic amine. *A. niger* was able to degrade penoxsulam and yielded five degradation products which were identified as 2-(2.2-difluoroethoxy)-*N*-(5-hydroxy-8-methoxy[1, 2, 4] triazolo[1, 5-c]pyrimidin-2-yl)-6-(trifluoromethyl) benzene sulfonamide; 5-8-dimethoxy[1, 2, 4] triazolo[1, 5-*c*]pyrimidin-2-amine; 2-(2.2-difluoro ethoxy)-*N*-(1*H*-1, 2, 4-trizolo-3-yl)-6-difluoro methyl benzene sulfonamide; 3-({[2-(2.2-difluoroethoxy)-6-(trifluoromethyl) phenyl] sulfonyl} amino)-1*H*-1,2,4-trizolo -5-carboxylic acid ; 5-8-dimethoxy[1, 2, 4] triazole [1, 5-c] pyrimidin-2-yl) sulfuric acid.

Macroscopic and microscopic view of *Aspergillus flavus*, and *A. niger*

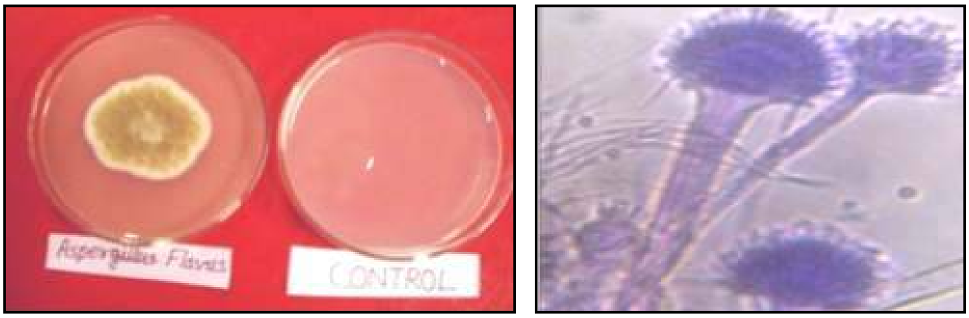
The benefits: This showed that *A. niger* is useful for degradation of penoxsulam in soils. Penoxsulam was also mineralized in soil inoculated with pure culture of fungus. The results showed that, either *A. niger* or the enriched mixed fungal consortium can be effectively used for the enhanced biodegradation of penoxsulam in agricultural soil.

7.2 *Aspergillus flavus* as a potential bio-degradation agent of penoxsulam in soil

The problem: In Asia, sulfonamide group of herbicide are being used on a wide range of crops, because of their very high herbicidal activity on broadleaf and grassy weeds at low use-rates, and low mammalian toxicities. Due to high herbicidal activity of sulfonamide, some crops such as legumes and pastures are highly sensitive to trace-level residues of sulfonamide in soils which may cause severe economic loss to agricultural production, enhanced risk of environmental contamination, and ultimate adverse effect to human. Herbicide residues of sulfonamide have frequently been detected in surface water and groundwater as a result of runoff and leaching after their application. Microbial degradation is a one of the main route for ALS inhibitors detoxification in the soils. Thus the soil fungi *Aspergillus flavus* was isolated and evaluated for degradation of unintended high spray of penoxsulam in soil.

The technology: *Aspergillus flavus* was isolated from rhizosphere of penoxsulam treated rice field. Degradation potential of fungus was evaluated against various doses of penoxsulam in soil and residues were analyzed by reverse phase HPLC with RF Detector and transformation products was identified by LC-MS/MS.

A. flavus was found able to degrade penoxsulam and five degradation products which were characterized as 2-(2.2-difluoroethoxy)-*N*-(5-hydroxy-8-methoxy[1, 2, 4] triazolo[1,5-c] pyrimidin-2-yl)-6-(trifluoromethyl) benzene sulfonamide; 5-8-dimethoxy [1, 2, 4] triazolo[1, 5-*c*] pyrimidin-2-amine; 2-(2.2-difluoro ethoxy) -*N*- (1*H*-1, 2, 4-trizolo-3-yl)-6-difluoro methyl benzene sulfonamide.



Macroscopic and microscopic view of *Aspergillus flavus*

The benefits: Analysis of soil samples containing *A. flavus* indicated that it was able to degrade penoxsulam. Penoxsulam was also mineralized in soil inoculated with pure culture of *A. flavus*. The results showed that, either *A. flavus* or the enriched mixed fungal consortium can be effectively used for the enhanced biodegradation of penoxsulam in agricultural soil.

7.3 Decontamination of sulfosulfuron-contaminated soil by *fungus*

The problem: Sulfosulfuron is a selective and systemic herbicide. It controls weeds in wheat like *Phalaris minor*, *Avena ludoviciana*, *Rumex dentatus*, etc. resulting in higher grain yield. But its residues in soil may cause phytotoxicity to the crops of next season. Barley (*Hordeum vulgare*), oats (*Avena sativa*), maize (*Zea mays*), sunflower (*Helianthus annuus* L.) and sorghum (*Sorghum bicolor* L.) are among those crops which are sensitive to sulfosulfuron. The present investigation was undertaken in order to isolate soil borne sulfosulfuron-degrading fungi, which can decontaminate soil from sulfosulfuron.

The technology:

Isolation and characterization of sulfosulfuron degrading fungus

Fungi isolated from rice rhizospheric soil were allowed to grow in the minimal media having sulfosulfuron as carbon and nitrogen source. *Trichoderma viride,* a widely distributed fungus in soil, survived and grew in that media enriched with sulfosulfuron at the level as high as 200 mg/L. This organism was characterized on the basis of morphological characters.

Degradation of sulfosulfuron by *Trichoderma viride* in sterilised soil

The incubation of *Trichoderma* sp. in minimal media and soil led to major degradation of the compound. Five key metabolites, which give the direction of degradation pathway, are confirmed by mass spectra and synthesis of the metabolites, and related literature.

Details of application methodology:

- **Isolation and identification of sulfosulfuron degrading fungus:** The collected soil was enriched with sulfosulfuron (5 mg in 100 g of soil) and incubated for a week at 30^{0}C. For selection of fungi as a suitable sulfosulfuron degrading agent, serial dilution following agar plating of incubated soil was done. Fungi that appeared on PDA plates (prepared from 200 g of potato, 20 g of dextrose, 20 g of agar and 1000 mL of water) after 5 days of incubation were further plated for obtaining pure cultures. The fungi screened from sulfosulfuron enriched soil were again incubated for 7 days in minimal PDA broth (prepared from 10 g of potato, 20 g of dextrose, and 1000 mL of water) containing different levels of sulfosulfuron, viz. 25, 50, 100, and 200 mg per 100 mL of broth. The most efficient fungus was screened out on the basis of their growth and was further inoculated on potato dextrose agar (PDA) plates. After two days of incubation colony morphology of the isolate was examined. The fungus was characterized on the basis of colony morphology and microscopy of spore structures.

- **Degradation of sulfosulfuron by *Trichoderma viride*:** For the degradation studies, 25 mg of sulfosulfuron was added to 100 mL of sterile dextrose-minimal broth (prepared from 100 g of potato, 10 g of dextrose, and 1000 mL of water) in 250 mL flask. The sulfosulfuron was allowed to dissolve overnight on shaker before use. Twenty such flasks were incubated with isolated *Trichoderma viride* in the dark at 25^0C for 27 days in BOD incubator. Three flasks with minimal broth and sulfosulfuron, and without the incubation with *Trichoderma viride* were kept in dark as control. Degraded products were extracted by partitioning in chloroform from the broth sampled in different time intervals, viz. 3, 9, 16, and 27 days of incubation. Solvent was then evaporated under low pressure in rotary vacuum evaporator to obtain a crude mixture of products. Products were purified by preparative thin-layer chromatography and characterized by the spectroscopic techniques.

The benefits: Sulfosulfuron seems to have the ability to inhibit the growth of some fungi present in soil as it did not allow many of them to grow in media and soil incubated with the herbicide. But its biodegradation, both in soil and in media by *Trichoderma* indicates that the application of this fungus can decontaminate soil from sulfosulfuron. Moreover, *Trichoderma viride* is beneficial for agricultural soil, as it restricts the growth of many phytopathogens.

7.4 Fungi-assisted decontamination of sulfosulfuron contaminated water

The problem: The use of herbicide in weed management is an essential component of the modern day agriculture. In our country, area under herbicide applied weed management is increasing rapidly. This increasing use of herbicides may contaminate our environment and food chain. Persistant herbicides may cause health hazards to us and domestic animals. Sulfosulfuron is a moderately persistent herbicides. It is used in wheat to manage its major weeds. It may risk contaminating water by runoff and seepage through soil. To avoid this risk, the herbicide is required to be degraded in water. Microbial processes can be useful to contaminate the water from pollutants. A technique is developed here by isolating efficient fungi that can degrade the herbicide into non-toxic components.

The technology:

Isolation and identification of sulfosulfuron degrading fungi from irrigation water : Two fungi, *Aspergillus flavus* and *Mucor piriformis* were isolated as sulfosulfuron degrading agents. These two fungi could survive in minimal media containing sulfosulfuron at the level of 200 mg/L.

Degradation of sulfosulfuron by *Mucor piriformis* : The incubation of *Mucor piriformis* in irrigation water led to major degradation of the compound. Five metabolites, which give the direction of degradation pathway, were confirmed by mass spectra, synthesis of metabolites, and related literatures.

Details of application methodology:

- **Isolation and identification of sulfosulfuron-degrading fungus :** The collected water sample was enriched with sulfosulfuron (5 mg in 100 ml of water) and incubated for a week at 30^{0}C. For selection of fungi as a suitable sulfosulfuron-degrading agent, serial dilution following agar plating of incubated water was done. Fungi that appeared on potato dextrose agar (PDA) plates (prepared from 200 g of potato, 20 g of dextrose, 20 g of agar and 1000 mL of water) after 5 days of incubation were further plated for obtaining pure cultures. The fungi screened from sulfosulfuron-enriched water were again incubated for 7 days in minimal PDA broth (prepared from 10 g of potato, 20 g of dextrose, and

1000 mL of water) containing different levels of sulfosulfuron, viz. 25, 50, 100, and 200 mg per 100 mL of broth. The most efficient fungus was screened out on the basis of their growth and was further inoculated on PDA plates. After two to three days of incubation colony morphology of the isolate was examined. On the basis of colony morphology and microscopy of spore structures the fungus was characterized.

- **Degradation of sulfosulfuron by *Mucor piriformis* :** For the degradation studies, 25 mg of sulfosulfuron was added to 100 mL of sterile dextrose-minimal broth (prepared from 100 g of potato, 10 g of dextrose, and 1000 mL of water) in 250 mL flask. The sulfosulfuron was allowed to dissolve overnight on shaker before use. Twenty such flasks were incubated with isolated *Mucor piriformis* in the dark at 25^0C for 27 days in BOD incubator. Degraded products were extracted by partitioning in chloroform from the broth sampled in different time intervals, viz. 3, 9, 16, and 27 days of incubation. Solvent was then evaporated in rotary vacuum evaporator to obtain a crude mixture of products, which were purified by preparative thin-layer chromatography and characterized by the spectroscopic techniques.

The benefits: Sulfosulfuron seems to have the ability to inhibit the growth of some fungi present in water as it did not allow many of them to grow in media and soil incubated with the herbicide. But its biodegradation, both in water and in media by *Aspergillus flavus* and *Mucor piriformis* indicates that the application of this fungus can decontaminate water from sulfosulfuron.

7.5 Decontamination of orthosulfamuron contaminated soil by fungi

The problem: Orthosulfamuron has been recently introduced in Indian agriculture. It is a systemic herbicide belonging to the family of pyrimidinyl sulfonylureas and controls broadleaf weeds and sedges in the transplanted and direct seeded rice. The interaction between this new herbicide and microbes, particularly under Indian condition is yet not established, but this information is imperative for the its safe use. From this point of view, the present investigation was carried out to reveal the effect of this herbicide on fungi present in the rhizosphere of rice and the metabolism of it therein.

The technology:

Isolation and identification of orthosulfamuron degrading fungi from rice rhizospheric soil: From the rice rhizospheric soil, ten orthosulfamuron-degrading fungi, viz. *Aspergillus niger, A. flavus, A. fumigatus, A. terreus, Curvularia* sp., *Cladosporium herbarum, Penicillium chrysogenum, Trichoderma viride, Trichoderma* sp. and *Penicillium* sp. were isolated. They could survive in the soil contiaining the same herbicide at the level of 4 g/kg.

Metabolism of orthosulfamuron by *Aspergillus niger*: *Aspergillus niger* isolated from the soil easily metabolised orthosulfamuron into relatively less toxic components. The degraded products were isolated, characterized and confirmed by the mass spectra, synthesis of metabolites, and related literatures.

Details of application methodology:

- **Isolation and identification of orthosulfamuron-degrading fungus from soil:** The collected soil sample was enriched with orthosulfamuron (5 mg in 100 g of soil) and incubated for a week at 30 ± 2^0C. For a selection of fungi as a suitable orthosulfamuron-degrading agent, serial dilution following agar plating of incubated soil was done. Fungi that appeared on PDA plates (prepared from 200 g of potato, 20 g of dextrose, 20 g of agar and 1000 mL of water) after 5 days of incubation were further plated for obtaining pure cultures. The fungi screened from orthosulfamuron-enriched soil were again incubated for 7 days in minimal PDA broth (prepared from 10 g of potato, 20 g of dextrose, and 1000 mL of water) containing different levels of orthosulfamuron, viz. 25, 50, 100, and 200 mg per 100 mL of broth. Efficient fungi were screened out on the basis of their growth and was further inoculated on PDA plates. After two to three days of incubation, colony morphology of each of the isolates was examined. On the basis of colony morphology and microscopy of spore structures the fungi were characterized.

- **Degradation of orthosulfamuron by *Aspergillus niger* in soil:** For the degradation studies, 25 mg of orthosulfamuron was added to 100 mL of sterile dextrose-minimal broth (prepared from 100 g of potato, 10 g of dextrose, and 1000 mL of water) in 250 mL flask. The orthosulfamuron was allowed to be dissolved overnight on shaker before use. Twenty such flasks were incubated with isolated *Aspergillus niger* in the dark at 25^{0}C for 27 days in BOD incubator. Similarly, 20 sets of 100 g of sterilized soil containing 100 mg of orthosulfamuron were incubated with *A. niger*. Degraded products were extracted by partitioning in chloroform from the broth sampled at different time intervals, viz. 3, 9, 16, and 27 days of incubation. The soil samples in triplicate for each day (10, 20, 30 and 40 days after inoculation) were extracted in dichloromethane and cleaned up. The solvent was then evaporated under low pressure in the rotary vacuum evaporator to obtain a crude mixture of degraded products. The products were purified by preparative thin-layer chromatography and characterized by the spectroscopic techniques as (i) *o*-amino-*N*,*N*-dimethylbenzamide, (ii) *N*-(4,6-dimethoxypyrimidin-2-yl)urea, (iii) 2-amino-4,6-dimethoxypyrimidine, (iv) *N*-[2-(*N*,*N*-dimethyl carbamoyl)phenyl] sulfamic acid, and (v) 2-(*N*,*N*-dimethyl carbamoyl)phenyl sulfamoyl urea.

The benefits: The herbicide orthosulfamuron does not have an inhibitory effect on the isolated fungi from rice rhizosphere of the recommended dose. On the contrary, fungus like *Aspergillus niger* can metabolize the herbicide through the cleavage of sulfonyl-amide linkage of urea side leaving behind relatively less toxic degraded products.

7.6 *Penicillium chrysogenum* as potential biodegradation fungi of pyrazosulfuron-ethyl in soil

The problem: Sulfonylureas inhibit acetolactate synthase (ALS) in the sensitive plants. Some crops such as legumes and pastures are highly sensitive to trace-level residues of sulfonylurea herbicides in soils. In the neutral or alkaline soil, the persistence of some sulfonylurea is very long, which poses risk of leaching or damage to successive crops. Biological degradation mediated by microbial enzymes is the main route for detoxification of sulfosulfuron in the soil. Owing to reported phytotoxicity of some sulfonylurea class of herbicides in number of sensitive crops and higher persistence in soil, present technique was develop to isolate and identify pyrazosulfuron-ethyl degrading fungi from soil of rice field.

The technology: The fungi, *Penicillium chrysogenum* can be used as a microbial consortium, to enhance biodegradation of pyrazosulfuron-ethyl in soil. In this techniques, *P. chrysogenum* was isolated and identified from rhizosphere of rice field. Degradation potential of *P. chrysogenum* was assessed in sterilized and unsterilized soil, and degradation products were identified by LC-MS/MS.

P. chrysogenum, was found capable to degrade pyrazosulfuron-ethyl by cleavage and hydrolysis of the sulfonylurea bridge. Degradation products of sulfosulfuron were identified as 4,6-dimethoxypyrimidin-2-amine; ethyl-5-[(4,6-dimethoxypyrimidin-2-ylcarbamoyl) sulfamoyl]-1-methylpyrazole-4-carboxylic acid; ethyl 1-methyl-5-sulfamyl-1H-pyazole-4-carboxylate and and 4,6 dimethoxypyrimidin -2-amine.

Pure culture of *Penicillium chrysogenum* and *Aspergillus niger* on PDA media along with spores, sporangiophore and mycelia

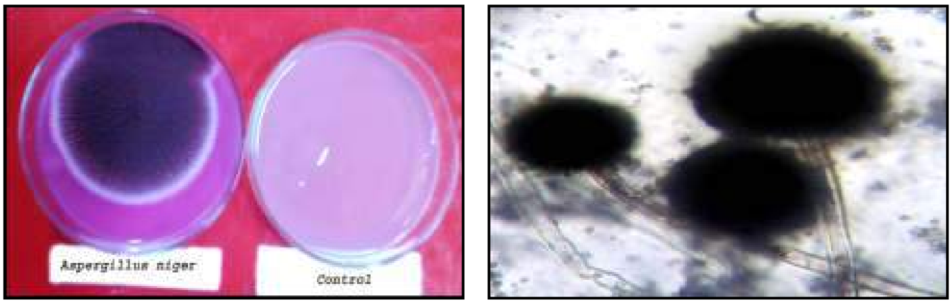
The benefits : *Penicillium chrysogenum* was found effective to degrade pyrazosulfuron-ethyl in the soil. This showed that, either *P. chrysogenum* or the enriched mixed fungal consortium can be effectively used for the enhanced degradation of pyrazosulfuron-ethyl in the agricultural field suspecting sulfonylureas residues. This technique is also important for bioremediation of agricultural fields having alkaline soil and loaded with pyrazosulfuron-ethyl or similar group of herbicide.

7.7 *Aspergillus niger* as potential biodegradation fungi of pyrazosulfuron-ethyl in soil

The problem: The sulfonylureas are used on a wide range of crops, because of their very high herbicidal activity on numerous broadleaf and grassy weeds at low use-rates. Widespread use of pyrazosulfuron-ethyl has raised increasing concerns on soil contamination and phytotoxicity problems in some direct seeded rice fields, which may affect agricultural production, and pose risk of environmental contamination, and human health. Biological degradation mediated by microbial enzymes is the main route for sulfonylurea herbicides detoxification in soils. Thus an *Aspergillus niger* fungal mechanism was identified to degrade pyrazosulfuron-ethyl in the soil by isolating and identifying pyrazosulfuron-ethyl degrading fungi from soil of rice field.

The technology: The fungus *Aspergillus niger* can be used separately to enhance degradation of unintended high spray of pyrazosulfuron-ethyl in soil. In this technique, *A. niger* was isolated from rhizosphere of rice field where pyrazosulfuron-ethyl was applied continuously to control annual and broad leaved weeds. Degradation potential of *A. niger* was assessed in sterilized and unsterilized soil, and degradation products were identified by LC-MS/MS.

A. niger were found capable to degrade pyrazosulfuron-ethyl. Transformation products were identified as ethyl-5-[(4,6-dimethoxypyrimidin-2-ylcarbamoyl) sulfamoyl]-1-methylpyrazole-4-carboxylic acid; ethyl 1-methyl-5-sulfamyl-1H-pyazole-4-carboxylate and 4,6-dimethoxypyrimidin-2-amine. Identification of degradation products suggested that the herbicide had undergone cleavage and hydrolysis of the sulfonylurea bridge.



Pure culture of *Aspergillus niger* on PDA media along with spores, sporangiophore and mycelia

The benefits: *Aspergillus niger* was found effective to degrade pyrazosulfuron-ethyl in the soil. This showed *A. niger* or mixed fungal consortia can be effectively used for the enhanced degradation of pyrazosulfuron-ethyl in the agricultural field suspecting sulfonylureas residues. This technique is also important for bioremediation of soil having neutral or alkaline soil and contaminated with pyrazosulfuron-ethyl or similar group of herbicide.

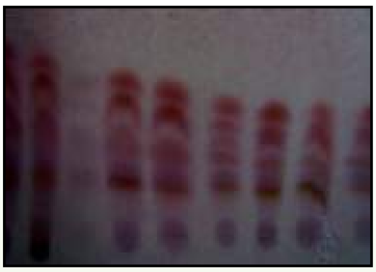
7.8 An ameliorative technique for mitigation of sulfosulfuron toxicity in plants

The problem: Sulfosulfuron is a unique herbicide, due to its wide weed control efficiency, low use rates, and low mammalian toxicity. Sulfosulfuron translocates throughout the plant and inhibits the action of acetolactate synthetase (ALS) enzyme responsible for formation of branched chain amino acid biosynthesis, stopping plant growth, and eventually killing the plant. Several secondary plant products are derived from these amino acids. Also, the intermediates of the branched-chain amino acid biosynthetic pathway are used in the biosynthesis of pantothenate and propionyl-CoA. Because mammals do not synthesize these amino acids, their biosynthesis in plants is of special interest. Sulfosulfuron is reported to persist in soil at phytotoxic concentrations for more than one year after application due to formation of secondary metabolites, especially at low temperatures and high pH, and can cause damage to sensitive crops such as pea (*Pisum sativum* L.), barley (*Hordeum vulgare*), lentil (*Lens culinaris Medik*.), sorghum (*Sorghum bicolor* L.), and sunflower (*Helianthus annuus* L.) grown as rotational crops after winter wheat. Thus a technique was developed to detoxify harmful level of sulfosulfuron in soil/plants.

The technology: Effect of sulfosulfuron phytotoxicity on plants up to sulfosulfuron 10 x dose and ameliorative effect of dextrose a most common reducing sugar were evaluated on biosynthesis of valine, leucine and isoleucine in crop and weed plants. To validate the technique, phytotoxicity of sulfosulfuron in wheat and *Vicia sativa* was evaluated. Healthy seeds of Common Vetch (*Vicia Sativa)* and wheat were in grown and treated with sulfosulfuron alone and along with dextrose using different doses. Dextrose was used at 0.5 and 1.0 kg/ha doses to detoxify sulfosulfuron phytotoxicity. Root, shoot growth and amino acid contents of weed and wheat plants were recorded at different days interval using standard protocols. Higher amino acid content, shoot and root growth were observed in treatment of glucose at 0.5 kg/ha dose.

The benefits: Results of this technology indicated that phytotoxicity of sulfosulfuron due to unintended higher doses can be mitigated by applying 0.5kg/ha dose of dextrose. This technology can also be applicable to deal with the toxic level of other sulfonylurea group of herbicides. It can also be used along with other phytoremediation techniques to expedite detoxification process.


Adverse effect of sulfosulfuron on plants amino acids


Detoxification of sulfosulfuron by formation of sulfosulfuron-dextrose conjugation

8.0 Expert system on weeds

8.1 Knowledge Based System (KBS) for weed seed identification

The problem: On occasions, the agricultural producers or other professionals will be challenged to identify a weed by the features of its seed. Accurate identification of weed seeds is necessary for the correct labelling of seeds moving in commercial channels which require skill and knowledge. The task of identifying weed seeds is difficult due to smaller size of many seeds, the differences in maturity, the loss of certain parts and the changes in surface appearance caused by processing or handling and difference in size and colour which occur under different soil and climate conditions. Some of the difficulties in identifying the weed seeds are lack of familiarity with the terms, failure to observe closely important features of the seed, lack of general knowledge in that area, and the fact that description of one observer does not always convey the intended information to another observer.

The technology: Knowledge Based System **(**KBS) on weed seed identification was developed with the combined effort of domain experts and software professionals. Based on the information collected on different weed seeds, a datasheet has been prepared with 11 characters and 'Scientific name' is considered as a primary key for managing the records. Using this datasheet, a knowledge base was developed which contains information on seeds of about 120 weeds with 11 parameters each. This system was developed using Visual Basic. Net as front-end applications and MS Access as back-end applications with user-friendly menus.

To use the system easily, the user friendly interface was developed with Graphical User Interface (GUI). The three major activities considered in designing the system are 'Weed-seed information, Search–Query, Weed thumbnail'. These three options allow the user to identify the weed seed characteristics by selecting family name (or) scientific name (or) seed photo. These fields were created with text boxes for data entry/modification and label boxes for title of the text. Image(s) of seed and plant photograph are embedded in the knowledge base itself. Based on the knowledge base, application software has been developed which consists of 4 modules. The multiple document interface (MDI) form of the software consists of 4 options, viz 'Masters', 'Details', 'Reports', and 'Help'. At present, data related to 120 weed seeds were fed into the software system, for storage and accessing. Open Data Base Connectivity (ODBC) has been provided to access the data from the database with the developed application.

Main menu

Weed seed information menu

Search by family name

8.2 Expert system for identification of weed seedlings

The problem: Identifying weeds at seedling stage is very much important for planning right management strategy. Also identification of weed seedlings is technically important because it would be helpful to identify the seed bank of the expected competitor weeds so that management can be planned in advance. Identifying the seedling stages of weeds is important because they are most susceptible to chemical or mechanical control at this stage. Also, accurate identification of these seedling weeds often is necessary to select the best herbicide or other method of weed control. Weed seedling identification is important to timely implement the appropriate weed control practice. Controlling the weeds in the early stages of growth not only increase the effectiveness of the control measures, but also reduces the crop losses. Identifying of a weed at seedling stage is very difficult compared to identifying a grown up weed sometimes even for a specialist also. Assistance in the form of a manual or software is very much needed for effective identification of weed seedlings.

Main menu

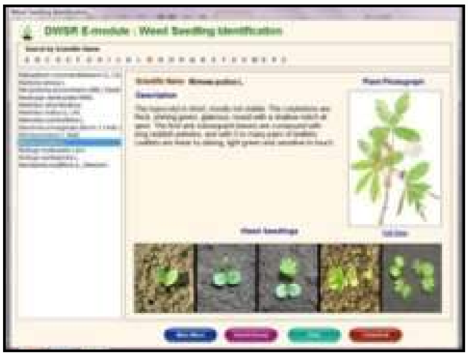
The technology: An expert system on weed seedling identification was developed with the combined efforts of specialists from the concerned subject, software professionals and other technical experts.

The main menu of this software consists of 4 modules, viz. Weed Information, Search Query, About software and Exit. The scientific names of plants are considered as primary key for identifying the characteristics of weed seedling.

Weed Seedling Information: It is a simple search mechanism which allows the user to search for a particular weed seedling by its scientific name.

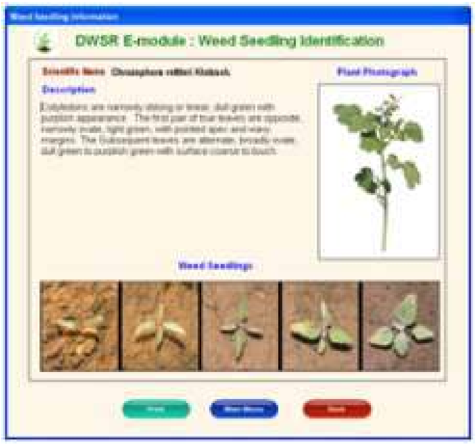
For easy searching, all the names are arranged alphabetically viz. A ….. Z. For example if the user is searching for a particular weed seedling and its name starts with 'M' like '*Mimosa pudica* L.', the user has to select 'M', then a list of scientific names gets displayed. By selecting '*Mimosa pudica* L.' from the list, the information viz. seedling description, image of the weed (grown up) and images of weed seedling at 5 different stages gets displayed. The user can also view the full image of the plant by selecting the option 'Full View'.

Search /Query: It is a powerful tool through which the user can make a query for a particular weed seedling by selecting Cotyledon shapes. 19 Cotyledon shapes are defined and designed in such a way with its image followed by the name and all the plants scientific names are classified into these 19 shapes. Once any one of the above shape is selected, a list of scientific names of weed seedlings that have the opted characteristics gets displayed and selecting one among the list displays the information of a particular weed seedling having the characteristics opted. For example, if the user selects the 'oblong' in Cotyledon shape option, a list of scientific names of weed seedlings having oblong cotyledons gets displayed. The user can select one of the scientific names from that list to display the characteristics of that particular weed seedling viz. description, plant photo and five different stages of weed seedlings. Hard copy of the report can be taken by selecting the 'Print' option. Navigating tools allows the user to move back and forth between the screens easily.



Weed Seedling Information menu

Search by leaf type



Weed seedling identification

About software: This is a portable software, which makes possible to execute this software in any system. For this, a 'SETUP' program is created (executable file) including all the files and data. Any user can install this software by running this 'SETUP' program and the execution of the software is self-explanatory.

8.3 Expert system for identification of weeds

The problem: The first step for effective weed management is accurate identification and basic understanding of the weeds' life cycle. It is said that identifying the weeds is half way to control. Correct identification is an important step so that new weeds can be eradicated before they become established. Proper weed identification helps in selecting right herbicide and time of application to control a particular weed. Sometimes identified plant may not be a troublesome weed and help in protecting conserving the biodiversity. However, identifying weeds is a difficult task unless one has experience gained over a period of time. Unfortunately specialist assistance is not always available everywhere for weed identification.

The technology: In order to overcome this problem, expert systems have been developed. The primary goal of expert systems research is to enable decision makers and technicians to do the job more efficiently. The expert system technology is a new approach for weed identification. An expert system is a computer program that contains formally encoded knowledge of experts in a given problem area or domain and is able to use this knowledge to provide help to a non-specialist in problem solving in that domain. The expert system on weeds allows their identification by specifying characteristics from a variety of categories viz., flowers, leaves and stem and sub-categories viz., flower colour, leaf shape, and stem cross-section. One can simply examine a weed and make choices based on its traits. There is an option to choose more obvious characteristics such as flower colour, plant height or leaf shape. All characteristics are displayed via detailed botanical glossary, which is as useful to novices as it is to professionals. With each choice you make, the list of possible plants shrinks. It is easy to confirm the identity of a weed by comparing the sample specimen to the many colored photos. This application is as useful to the amateur enthusiast as it is for professionals in the field of weed science. An attempt was made to develop expert system for identification of 337 commonly found weed species of cropped and non-cropped areas in the country.

This is portable software, which makes possible to execute this software in any system. For this, a 'SETUP' program is created (executable file) including all the files and data. Any user can install this software by running this 'SETUP' program and the execution of the software is self-explanatory.

Main menu


Weed information menu


Search query by leaf shape


Search query by flower colour


Advance search menu

9.0 Techniques on weed physiology and biology, climate change and herbicide residue studies

9.1 Screening of plants and their constituents for allelopathic potential

The problem: Allelopathy, interactions among plants including microbes through release of chemicals, called allelochemicals, into the environment, is a complex phenomenon of profound importance in agriculture, forestry and natural and managed ecosystems. The plants exhibiting allelopathy may affect growth and development and survival of species around them affect crop production. The allelochemcals (mostly secondary metabolites) present in the allelopathic plants offer feat for their use as pesticides including herbicides as such or after chemical modification and also they may provide lead for the development of pesticides with newer site of action or provide lead for the development of newer pesticide formulations. It is imperative before hand to know allelopathic potential of plants, plant parts, and their constituents. This may require a simple, easy to practice and comprehensive protocol.

However, allelopathic potential, capability to exert allelopathy, of a plant in a simplistic laboratory evaluation using effect on germination and early seedling growth and on aquatic weed can be best tested by bringing the active principle in solution or in culture medium since competition for nutrients and space can be minimized and monitored under these conditions. Any change in response of the species growing in the medium can be, under these conditions, attributed to applied allelochemical species or the allelochemical treatment. The method may facilitate evaluation of allelopathic potential of a species, plant parts and allelochemical crude or an isolated constituent. The technique may help in rapid evaluation of the allelopathic potential.

The technique:

1. **Preparation of allelochemical crude from whole plant:** Whole plants or plant parts can be collected, washed briskly, blot dried, and plunged into water and allowed to stand for specified time e.g., 24 hours at ambient or cold temperatures (e.g. 10°C) depending on

materials and type of the constituents. The leachate is allowed to stand, decanted and coarse particles removed through straining through a pre-washed and dried muslin cloth. The clear solution or leachate is spread over glass or metal surface into a thin film and dried under fans preferably at cooler temperatures. The allelochemical crude is scraped, collected into glass containers and stored in cool and dry place until use.

2. **Preparation of plant materials, plant parts residues and allelochemical crude:**

2.1. **Preparation of plant materials and plant parts residues:** Allelopathic plant or suspected allelopathic plant is harvested, rapidly washed and blot dried, further drying is done in shade preferably under fans, ground to pass through an 80 mesh sieve and the powder (residue) is stored in cool and dry place until use.

2.2. **Preparation of allelochemical crude:** The plant or plant parts powders (residues) prepared as described above is suspended in water with occasional or continuous stirring for 24 hours preferably at around 10°C. The contents are allowed to settle and the clear solution is decanted. Coarse particles if any are removed through straining through a pre-washed and dried muslin cloth. The solution or leachate is spread into a thin layer over plastic or glass surface and allowed to dry under fans. Drying can be done in a better way *in vacuo*, or at lower temperature or under nitrogen or carbon dioxide at lower temperatures to prevent possible changes in the constituents. The allelochemical crude is scraped, collected in glass containers and stored in a cool and dry place until use.

3. **Concentration of the allelochemical constituents:** Further fractionation of active constituents involves finding a suitable solvent that extracts the bioactive fractions most efficiently. This step is to facilitate concentration of the active constituents. The scheme developed and employed in the laboratory is primarily based on the fact that since the allelochemicals have to move through water in nature, they should be extracted first with water. The allelochemical crude can be extracted with 13 solvents covering a range of polarity spectrum from polar to non-polar involving acetone, carbon tetrachloride, chloroform, cyclohexane, dichloromethane, diethyl ether, ethanol, ethyl acetate, hexane, methanol, pentane, toluene and

petroleum ether, and the constituents solubilized in each of them can be dried and collected. These can be bioassyed using standard test materials to evaluate yield and bioactivity. The solvents that give maximum yields of biologically active materials are used for concentration of the active principles for further isolation of the constituents.

4. **Isolation of allelochemcials from the mixture:** Specific methods are required for isolation of allelochemical constituents. Harborne's (1995) scheme for fractionation of constituents to different chemical classes viz., phenolics, terpenoids, alkaloids, etc. may be employed to facilitate isolation and characterization of the allelochemicals employing analytical and preparative thin layer- and column-chromatography, and physicochemical techniques involving group specific detection reagents and spectral details using standards and/or published details.

5. **Evaluation of allelopathic/inhibitory/herbicidal potential:**

5.1. **On seed germination and early seedling growth:** The plant parts, plant parts residues, allelochemical crude and allelochemical molecules can be evaluated for allelopathic potential on weed and crop seed germination and early seedling growth by using leachate obtained by soaking the plant materials in water for specified periods or by using solutions of the crude or isolated constituents in a series of concentrations. The materials to be evaluated can be suspended in water at different levels e.g. at 0.01, 0.025, 0.05, 0.1, 0.25, 0.50, 0.75 and 1.0% (w/v) for specified periods or continuously retained in the medium depending on objectives of the study, or molecules can be prepared in the concentration range of 1-500 ppm in water. Water alone may serve as controls. Germination, early seedling growth of crop and weed seeds under standard conditions (following ISTA recommendations), seedling growth in the dark etc. are monitored. Comparison of response over control will show positive or negative allelopathic potential of the plant, plant parts, allelochemical constituents and isolated molecules.

5.2. **On floating and submerged aquatic weeds:** For evaluation of allelopathic potential on floating and submerged aquatic weeds, the materials are soaked in water to obtain leachates or solutions or are retained in the medium as necessitated, keeping in view objectives of the experimentation, and to these pre-weighed floating weeds viz., water hyacinth (*Eichhornia crassipes* Mart Solmns.), pistia (*Pistia stratiotes* L.), spirodella (*Spirodella polyrhiza* L.), lemna (*Lemna pausicostata* Hegelm.) and azolla (*Azolla nilotica* Decne) and submerged weeds viz., hydrilla (*Hydrilla verticillata* L. f. Royle), ceratophylum (*Ceratophyllum demersum* L.), potamogeton (*Potamogeton crispus* L.) and green musk chara (*Chara zeylanica* Willd.) are loaded. The medium is supplemented with standard nutrient medium. The test is incubated outdoors replenishing evapotranspiratory loss of water twice daily. The treatments along with nutrient mediums as controls are evaluated at intervals for change in biomass or appearance of toxicity with time and allelopathic potential is computed.

6. **Salient findings about the technique:** The technique facilitates complete evaluation of allelopathic and herbicidal potential of plant, plant parts, constituents and allelochemicals suitable for basic research work as well as applied explorations.

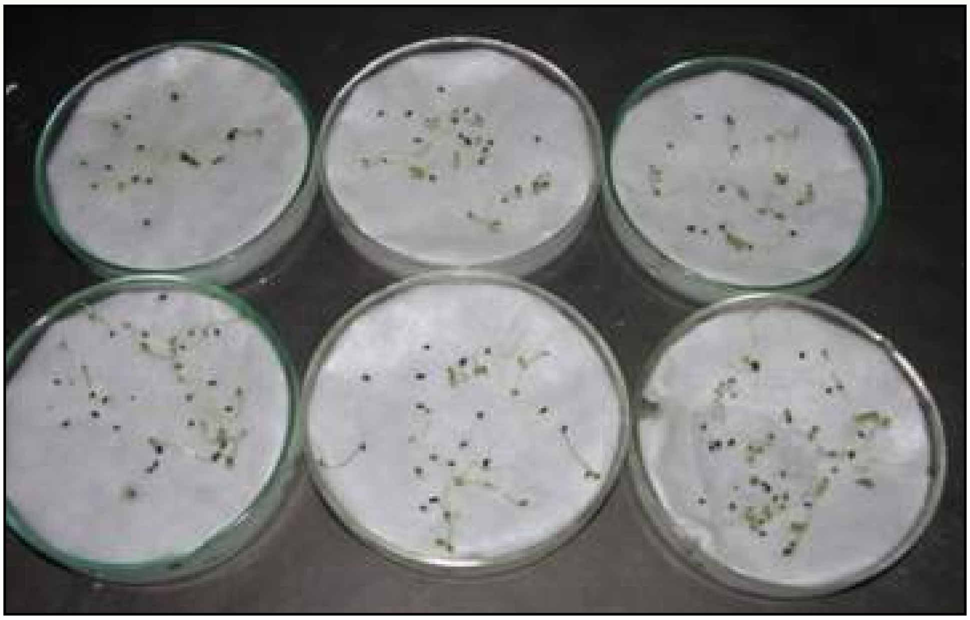
9.2 Technique for quick germination of *Parthenium hysterophorus* seeds

The problem: Fresh seeds of *Parthenium hysterophorus* do not germinate. They should remain in the soil for a considerable period of time before they get germinated. This is because of the dormancy which is mostly due to seed born toxicants. The dormancy will lead to the enrichment of *Parthenium* seed bank in the soil. For conducting research on *Parthenium* in the laboratory or pot culture or in the field involving seeds the difficult task is getting its germination. Understanding the factors responsible for keeping the *Parthenium* seeds dormant is essential for breaking the dormancy with a suitable technique.

The technique: The fresh seeds of *Parthenium* were kept in nylon net bags and were subjected to running water for different durations of time i.e. 0 'hour (fresh seeds), 4 hours, 8 hours, 24 hours, 28 hours, 32 hours and 48 hours. The germination was tested on filter paper in petridishes and in sandy and clayey soils in pots. The first observation from this study was that the fresh *Parthenium* seeds when placed on moist filter paper for germination left a dark brown stain on the paper before they got germinated. In the fresh seeds the % germination was very low and the time taken for initiation of germination was more when compared to the washed seeds. Increased duration of time under running water flow enhanced the % germination and reduced the duration for initiation of germination. The % germination recorded on 5th day after sowing was 17%, 41%, 50%, 73% 83% and 100% in fresh seeds, 4 hours, 8 hours, 24 hours, 28 hours, and 32 hours of running water treatment respectively. In case of 48 hours of running water treatment, germinated seeds were found within the nylon bags at the end of the treatment period.

Germination of *Parthenium* seeds on filter paper after subjecting to continuous running water treatment for different durations of time

Duration of running water treatment (Hours)	% Germination on filter paper
0 (Fresh seeds)	17
4	41
8	50
24	73
28	83
32	100
S.E	1.61
LSD (P=0.05)	4.96

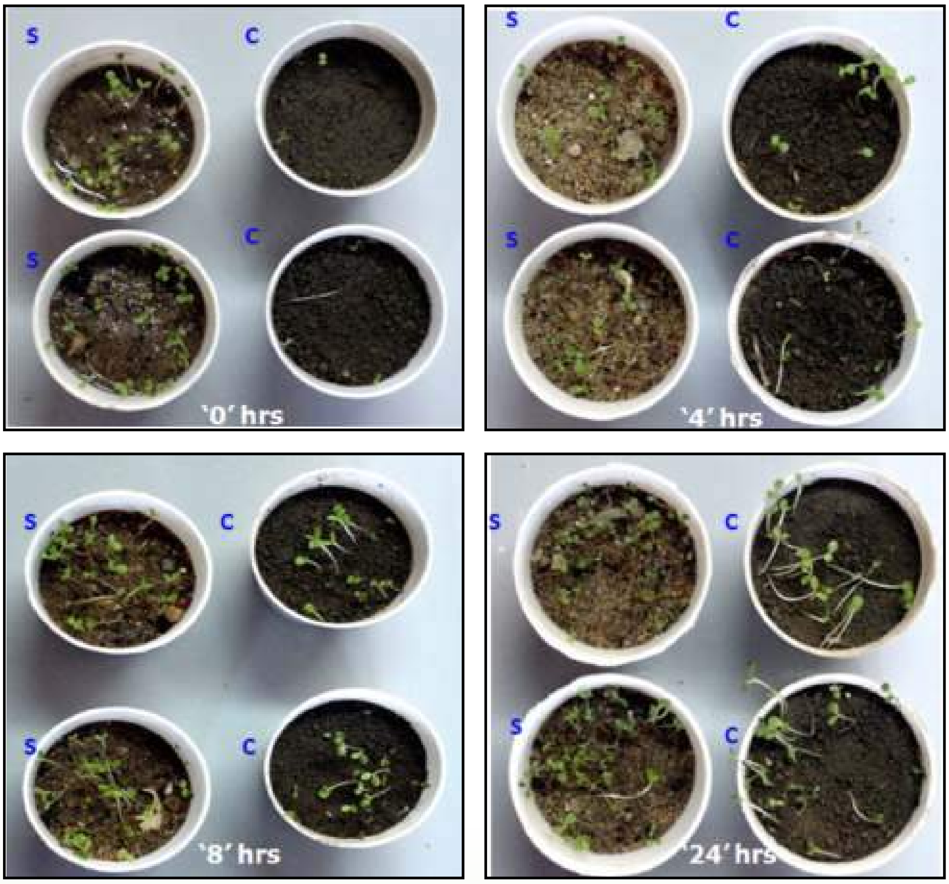


The seeds germinated within the nylon net bags after 48 hours of continuous running water flow.

Testing the germination in sandy and clayey soils, which differ in moisture holding capacity has also confirmed above results. Very early germination was observed in sandy soil and the % germination of fresh seeds was very higher in sandy soils (76%) than in clayey soil (5%). The seeds that were subjected to running water have shown enhanced germination both in sandy and clayey soils but % germination was higher in sandy soils in all the treatments.

Germination of *Parthenium* seeds on clayey and sandy soils after subjecting to continuous running water treatment for different durations of time

Duration of running water treatment (Hours)	% germination on Clay soil	% germination on Sandy soil
0 (Fresh seeds)	5	76
4	45	80
8	46	83
24	66	96
28	73	100
32	80	100
S.E	2.52	1.8
LSD (P=0.05)	7.78	5.82



Germination of *Parthenium* seeds on clayey and sandy soils after subjecting to continuous running water treatment for different durations of time.

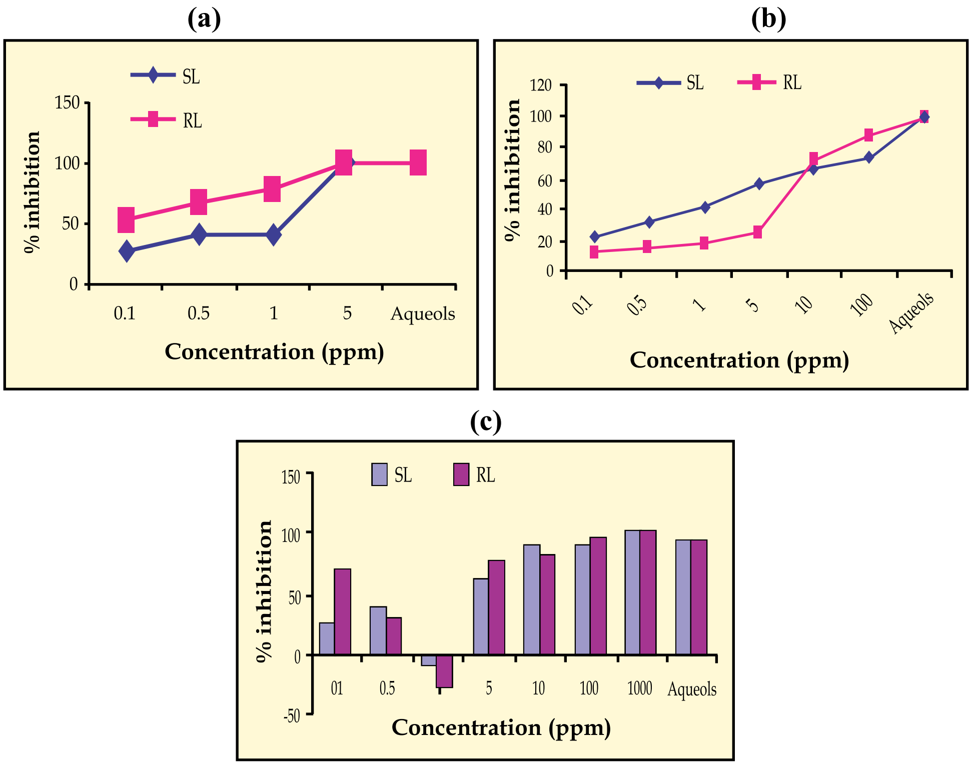
S: Sandy soil C: Clayey soil

9.3 Technique for the isolation of allelochemicals from root exudates of linseed

The problem: While herbicides are very important to agriculture, under certain circumstances they may act as pollutants that can deteriorate soils, ground and surface waters, and may have deleterious effects on environment and non-targeted organisms. As a result farmers and grower are looking for less harmful, non-persisting biopesticides. Allelopathic substances, if present in crop varieties, may reduce the need for weed management, particularly herbicide use. Many plant chemical often called as allelochemicals in natural or processed forms have potential for development as viable components of biopesticides. Allelo chemicals have wide range of activity against crop pests such as weeds, nematodes, insects, microbial and fungal diseases etc. and thus can be used as biopesticides. Allelopathy alone may not be a perfect weed management technology but it may be a supplementary tool for weed control. Hence a technique was developed to isolate root exudates from linseed plants.

The technology: Water soluble allelochemicals may be released through leaching in response of action of rain or watering by exudation of water soluble toxins from below ground parts. Hence an efficient, and simple technique was developed to isolate, allelochemicals from root exudates of linseed. Linseed plants were grown on plastic pots in a medium of sand and soil and small holes were made in the bottom of plastic pots to allow leaching of allelochemicals from roots along with leachates. After two weeks root entered to the below attached pots through the small holes. Root exudates of linseed were collected at every 4-5 days interval up to the plant maturity. Linseed allelochemicals were isolated form concentrated extract through solvent partioning method. Two compounds were isolated from methanol fraction of linseed at Rf of 0.64 and 0.35 which were further purified and identified by HPLC using PDA detector. In ethyl acetate, fraction two peaks were identified at RT of 2.6 and 4.5. Pure compounds obtained from different fractions were further identified by HPLC using RF detector at RT of 10.92 (14.8%) from aqueous fraction, RT of 7.6 (41.5%) from petroleum ether fraction, RT of 18.13 (45.3%) from chloroform fraction, RT of 19.23 (21.9 %) from n-hexane fraction and RT of 13.89 (14.8%) from methanol fraction of linseed. This demonstrated success of this technique for the isolation of trace amount of allelochemicals from linseed/marigold plants roots.

Growth inhibitory activity of viscous powder isolated from linseed was evaluated on *Lathyrus sativa, Phalaris minor, Convolvulus arvensis, Parthenium hysterophorus, Vicia sativa*, etc. The phytotoxic effect of root leachates was in the order *P. minor* > *L. sativa* > *P. hysterophorus* > *Vicia sativa* > *C. arvensis.* Stimulatory effect was observed on *Euphorbia* sp. growth at 1.0 ppm. The aqueous extract was found more phytotoxic in reducing the growth of all the weeds as compared to isolated allelochemicals alone. Linseed allelochemicals reduced the growth of weeds by inhibiting root growth.



Effect of isolated compound from linseed root leachates on the growth of (a) *Phlaris minor,* (b)*Vicia sativa* and (c) *Euphorbia hirta*

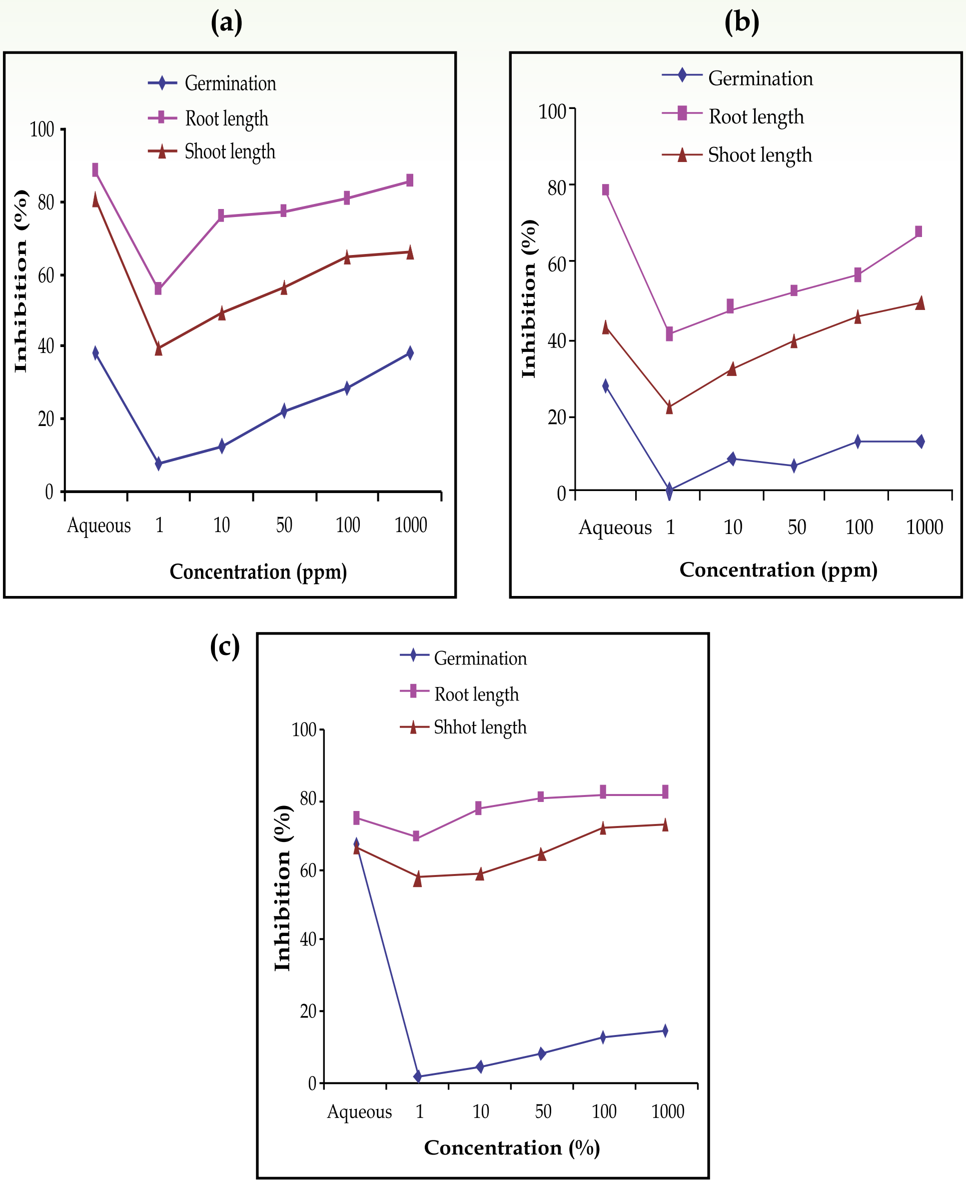
The benefits: This technology was found successful to isolate allelochemicals from linseed without altering plant growth. Aqueous extract and allelochemicals were found phytotoxic against a number of weeds under laboratory studies. This technology can be used to isolate allelochemicals from other plants.

9.4 Technique for the isolation of allelochemicals from root exudates of marigold

The problem: Allelochemicals are gaining popularity throughout the globe due to public awareness about environment, growing knowledge of pollution and health hazards due to synthetic chemicals as a result farmers and grower are looking for less harmful, non-persisting biopesticides. Allelopathic substances, if present in crop varieties, may reduce the need for weed management, particularly herbicide use. Allelochemicals in natural or processed forms have potential for development as viable components of biopesticides. Allelochemicals have wide range of activity against crop pests such as weeds, nematodes, insects, microbial and fungal diseases etc. and thus can be used as biopesticides. In general, these compounds are less toxic to non-target species, and less persistent in soil than synthetic pesticides.

The technology: An efficient, simple and rapid new technique was developed to isolate, allelochemicals from root exudates of marigold without affecting plant natural growth. Marigold plants were grown on plastic pots and small holes were made in the bottom of pots to allow leaching of allelochemicals from roots. Root exudates were collected weakly up to four months. Leachates were filtered and concentrated to approximately 500 mL. Allelochemicals were isolated in form concentrated extract through solvent partioning method. Three compounds were isolated from methanol fraction with Rf of 0.36, 0.42 and 0.82 as orangish brown colour spots. Pure aqueous leachates yielded one compound at Rf of 0.82. These isolated compounds on further purification by HPLC using PDA detector yielded allelochemicals at RT of 14.05 (26.01 %) from aqueous fraction, RT of 27.2 (20.6%) from petroleum ether fraction, RT of 20.2 (53.5 %) from acetone fraction, RT of 17.9 (52.6 %) from ethyl acetate fraction and RT of 18.26 (27.3%) from methanol fraction of marigold. This demonstrated success of this technique for the isolation of trace amount of allelochemicals from linseed/marigold plants roots.

Phytotoxic ability of viscous powder isolated from marigold was found in order *P. hysterophorus* >*P. minor* >*L. sativa*> *C. arvensis* > *A. ludoviciana* > *V. sativa.* The aqueous extract was found more phytotoxic in reducing the growth of all the weeds as compared to isolated allelochemicals alone. In general weed root growth was affected more as compared to shoot growth by the allelochemicals and aqueous extract.



Effect of isolated compound from marigold root leachates on the growth of (a) *Lathyrus sativa,* (b) *Phalaris minor*, (c) *Parthenium hysterophorus*

The benefits: This technology was found successful to isolate allelochemicals from marigold in trace quantity without disturbing plant natural growth. Aqueous extract and trace level of allelochemicals were found phytotoxic against a number of weeds under laboratory studies. Allelochemicals may be used as a supplementary means for control of these or other weeds under diverse situations.

9.5 Quantification of *Orobanche* seed bank in soil

The problem: *Orobanche* is a root parasitic plant that depends fully on the host plants for its nutrients and water requirements. They attack several agriculturally important plants including mustard, tomato, brinjal, potato, sunflower and tobacco causing loss in both quality and yield of the crops. They attach and enter into the host roots and develop underground for about 45-60 days to form fleshy tubercles. When the conditions are favorable, flower stalks emerge above ground and produce several flowers with lakhs of very minute seeds. The seeds remain viable for about 25 years and the quantity of seeds in the soil gives a clue about the infection in the next cropping season. Quantification of *Orobanche* seeds in soil is difficult because of their very minute size, similarity with other weed seeds and their inherent nature to germinate only in the presence of stimulants from the host. Thus instead of conventional germination tests special procedures using costly synthetic germination stimulants are used.

The technology: Surface sterilized seeds, about 100 in numbers were placed on clean sterile moist filter paper kept in Petriplates. The plates were covered with black polythene sheets and aluminum foil to provide complete darkness and incubated at $28\pm2^{0}C$ for 10 days for pre-germination conditioning. The conditioned seeds with filter paper were transferred aseptically on a bed made of sterilized sand, cotton and filter paper kept in 1000 ml beaker. Sand is used for maintaining moisture for longer periods, cotton and filter paper were used to keep the fine sand particles separated from the seeds. In a parallel setup, surface sterilized seeds of host plants (mustard or tomato) are germinated on germination papers in separate Petriplates. After germination, about 10 seedlings were transferred to the beaker containing the preconditioned *Orobanche* seeds. The beakers provided sufficient vertical space for the growth of host plant seedlings. A mild fungicide such as bavistin was sprayed to avoid fungal contamination. The beakers were kept at room temperature for 10 days and the filter paper with *Orobanche* seeds were observed under stereo binocular microscope for their germination.

Photographs showing *Orobanche* infestation in a) tomato, b) tobacco, c) brinjal and d) mustard crops and set up for germination of *Orobanche cernua* under controlled conditions

The benefits: The germination setup explained above is used for estimation of the weed seed bank in soil, which is essential to predict the intensity of infection in the cropping season and apply appropriate management strategies. This technique is useful in quarantine and export of the food grains from the infested areas. The technique is simple and use of live host seedlings removes the dependency on use of synthetic germination stimulants which is costly and not available commercially in many countries including India.

9.6 Weed associated rhizobacteria for metal tolerance and plant growth promoting traits

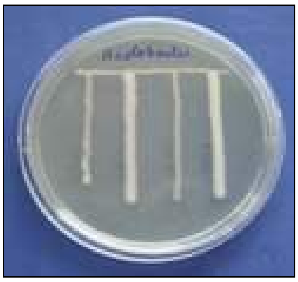
The problem: Although heavy metals are naturally present in the soil, geologic and anthropogenic activities increase the concentration of these elements to amounts that are harmful to both plants and animals. Plants growing on these soils show a reduction in growth, performance, and yield. Since, the present study investigated the metal under tolerance of plant growth promoting bacteria colonizing in the rhizosphere of naturally growing weedy grass species metal contaminated areas.

The technique: Plant growth promoting rhizobacteria were isolated using serial dilution technique on King's B medium from the selected weed rhizosphere in different heavy metal contaminated areas of Jabalpur district. The isolates were screened based on maximum tolerance concentration (MTC) of various heavy metals *viz.*, Cd^{2+}, Hg^{2+}, Co^{2+}, Ni^{2+} and Zn^{2+}. The 16S rRNA gene sequence analysis revealed these elite isolates; which belong to genus of *Bacillus* sp., *B. subtilis* and *B. licheniformis.* Plant growth promoting traits of all the three selected isolates were analysed and results recorded that the isolates were found to produce phytohormones, siderophores, solubilized minerals, isolates also produced enzyme such as ACC deaminase that can modulate plant growth and development. The experimental results indicated that the isolates were highly compatible with each other bio-inoculants. The pathogenicity experiment was conducted these selected strains do not cause any disease symptoms to tomato plants. This study recognized the microorganisms inhabiting metal-contaminated sites are important material for both study and applications of bioremediation.

The benefits: Bioremediation is an effective and economical method of treating heavy metal polluted soils. It is a widely accepted method that is mostly carried out *in situ*; hence it is suitable for the establishment/re-establishment of crops on treated soils. Microorganisms and plants employ different mechanisms for the bioremediation of polluted soils. Combining both microorganisms and plants is an approach to bioremediation that ensures a more efficient clean-up of heavy metal polluted soils.

The limitation: The success of the technique depends on the species of microbe and weed plants involved and to some extent on the concentration of the heavy metal in soil.

Photographs showing the plant growth promoting traits of the isolates



Compatability

Mineral solubilization

Siderophore production

Some of these activities include mining and smelting of metals, burning of fossil fuels, use of fertilizers and pesticides in agriculture, production of batteries and other metal products in industries, sewage sludge, and municipal waste disposal.

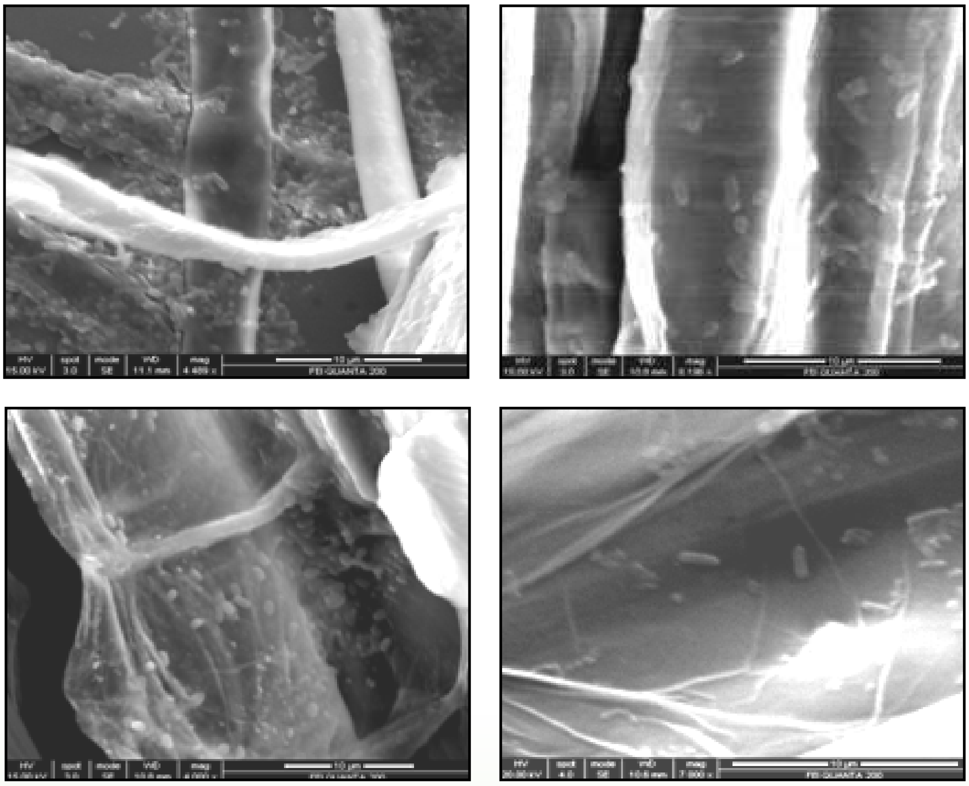
9.7 Identification of diazotrophic endophytes from the weedy grasses

The problem: Molecular nitrogen or dinitrogen (N_2) makes up four-fifths of the atmosphere, but is metabolically unavailable directly to higher plants or animals. Advances in agricultural sustainability will require an increase in the utilization of biological nitrogen fixation as a major source of nitrogen for plants. However, biofertilizers lose their effectiveness if the soil is too hot or dry and excessively acidic or alkaline soils. Moreover, they are less effective if the soil contains an excess of their natural microbiological enemies. Since the search for natural association and endophytic interaction of diazotrophs with weed grass species is considered very promising, especially in grasses that grows naturally with adverse environmental conditions. Therefore the present investigation is focused to identify and characterize the unexplored culturable endophytic diazotrophic bacterial diversity of weed grasses.

The technique: Weedy grass roots containing rhizospheric soil were washed with sterile distilled water, disinfected with 70% ethanol, rinsed, disinfected superficially with 3% sodium hypochlorite, rinsed again to eliminate hypochlorite, and spread on nutrient agar to confirm root surface sterility at 30°C for 5 days. Finally, roots were added with 0.9% NaCl (1:10) and macerated with mortar and pestle. One gram of macerated tissue was placed in a tube containing 9 ml sterile 0.9% NaCl. Specimen of washed roots, stems and leaves were macerated and 0.1 ml of serial dilutions in 4% (commercial) sugar solution up to 10^{-6} and there dilutions were inoculated into vials containing selective N-free semi solid media. The elite isolate were screened and further analysed for their potential of endophytic colonization. Dehulled seeds of rice (cultivar- ADT 43) were surface sterilized and treated with the selected bacterial inoculant for 15 min. The roots and culms from 15 days old fresh rice seedlings were cut, fixed with 3% (v/v) glutaraldehyde, for 2 h at 4°C and washed with 0.1M phosphate buffer (pH 7.2) at room temperature for 10 min. The samples again post fixed in 1% (w/v) osmium tetroxide in the same buffer for 2 h at 4°C. The fixed samples were dehydrated in a graded ethanol series for 5 minutes in each concentration. Then the samples were treated with CO_2 and mounted on an aluminum cylinder with silver paste, and finally covered with a steam of carbon and ionized gold. Localization and distribution of endophytic bacteria on roots and culms of rice seedlings were analyzed by scanning electron microscopy (SEM) (ICON, anaytical FEI Quanta 200, USA) operated at 15 kV at an 8-10 mm distance. Colonizing ability of the endophytic isolates in rice seedlings were documented as microphotographs.

The benefits: Elite diazotrophic strains from weeds having multiple plant growth promoting traits, as bioinoculants for nutrient management, biotic, abiotic stress mitigation and sustainable crop production with fewer chemical inputs.

The limitations: These endophytes from weed grasses may be either effective or ineffective, this depends on: (i) whether the microbes living inside are capable of fixing N; (ii) whether the microbes and the host plant can form a highly harmonious symbiotic N-fixing system, and (iii) the environmental condition in which the host plant is growing. Relationships of a plant with microbes, sunlight, temperature, air and moisture, etc. are factors to be considered.



Colonization of diazotrophic bacterial strain (*Serratia* sp. CRE2) on rice seedlings.
A- Longitudinal section of culm; **B**- Surface of the root;
C and **D**- Longitudinal section of root.

9.8 Screening of weedy rice for germination under anaerobic conditions

The problem: Poor seed germination and delayed plant establishment in rice in flood-prone lowland areas is major concern for rice production in such areas, especially in South and Southeast Asia where rice is a major crop. As such, rice is unique in being capable of growing well in waterlogged and submerged soils because of its well-developed aerenchyma system but is also extremely sensitive to anaerobic conditions during germination (anaerobic germination) and early growth of the embryo. Soil water logging or flooding can be encountered when it rains immediately after seeding or when the land is not well- levelled in irrigated areas or due to improper drainage. Genetic variation in the ability to germinate and establish in flooded soils has been observed in some rice cultivars and landraces and these can be used for developing cultivars with ability to germinate well under anaerobic conditions.

Weedy rice, a natural hybrid of cultivated and wild rice, possess properties of both rices. It has capacity to grow in standing waters, though the ability to do so may vary across germplasm. Being a rich source of varied gene pool due to natural hybridization events, it may apparently have ability to germinate under anaerobic conditions too. The method facilitates evaluation of germination under anaerobic conditions and hence screens weedy rice germplasm collected from different agro-ecosystems across the Nation. The technique may help in rapid screening of plants for anaerobic germination.

The technique: Seeds of weedy rice (along with awn) were placed one cm below surface of autoclaved soil in a single line in a plastic container and placed in containment facility. Each container had ten accessions. After sowing of seeds of different accessions, it was covered with 6 cm standing water and maintained so throughout the experiment. Percent germination was observed after every seven days to assess ability to germination. After 15 days, root length and vigor index were assessed. Seeds of accessions that did not germinate were put to standard tetrazolium test to assess their viability.

Germplasm of weedy rice collected from different states of India were evaluated in aforesaid manner and the germplasm revealed variation in the ability for anaerobic germination. Among the sixty five samples studied, only twenty seven germinated which included four rice cultivars and 23 weedy rice samples. Highest germination (100%) was recorded in an accession from Madhya Pradesh (MP) and Kerala (100%).

Germination of rice cultivars and weedy rice accessions under anaerobic conditions

Accessions T- weedy rice C=cultivated rice	Parameters studied		
	Germination (%)	**Root length (cm)**	**Vigour index**
T3: UP	40^{ABC}	4.53^{A}	888^{ABC}
T4: UP	20^{ABC}	4.00^{A}	614^{ABC}
T5: UP	53^{ABC}	3.72^{A}	1263^{ABC}
T8: UP	66^{ABC}	4.85^{A}	1686^{ABC}
T1: MP	20^{ABC}	0.50^{A}	40^{ABC}
T3: MP	60^{ABC}	5.90^{A}	1390^{ABC}
T6: MP	**100^{A}**	4.98^{A}	**2928^{A}**
T1: Kerala	60^{ABC}	2.02^{A}	575^{C}
T2: Kerala	93^{AB}	3.67^{A}	2306^{ABC}
T3: Kerala	**100^{A}**	3.10^{A}	**2805^{AB}**
T1:CGH	46^{ABC}	2.63^{A}	775^{BC}
T1:JKD	93^{AB}	3.72^{A}	2392^{ABC}
T1: WB	20^{ABC}	6.00^{A}	198^{ABC}
T1:Bihar	73^{ABC}	2.70^{A}	2014^{ABC}
C:Jyothi	40^{ABC}	2.52^{A}	805^{BC}
C: Sahbhagi	73^{ABC}	4.29^{A}	1718^{ABC}
C: Jaya	20^{ABC}	3.61^{A}	658^{ABC}
C: IR64	20^{ABC}	4.40^{A}	51^{ABC}
C: P. Basmati	40^{ABC}	2.75^{A}	1276^{ABC}
SE(d)	**20.01**	**1.77**	**590.7**
LSD (P=0.01)	**54.04**	**NS**	**1597**

(a) (b) (c)

Weedy rice accessions germinating under anaerobic conditions

(a) Ten accessions in a container having 6 cm standing water, (b) seeds that grew under anaerobic germination, (c) seeds that did not germinate were viable

The benefits:

1. The setup can be used for evaluating germplasm of plant species for ability to germinate under anaerobic conditions and help pick potential candidates for further molecular studies.
2. The technique is a representation of *in situ* conditions and hence reflects field conditions.
3. A large number of germplasm can be screened in a relatively less area.

9.9 A standardised protocol for the investigation on the role of leaf cutin in the photolysis of herbicides

The problem: After application, herbicides deposit on leaf surface. Leaf surface, made up of polymerised esters of higher fatty acids, is the first reaction environment for those herbicides. These fatty substances generate reactive radicals or ions in presence of sunlight. These radicals or ions may interact with herbicide molecules leading towards degradation. This sunlight assisted degradation, known as photolysis results in the loss of bioefficacy of herbicides and the formation of metabolites of unknown toxicity. It is very difficult and improper to account photolysis of any herbicide on foliar surface of live plant because some more factors other than sunlight also act upon the herbicide. Therefore, it has been attempted to develop a technique for the investigation on the role of leaf surface in the photo-transformation of herbicides.

The technology: A technique or protocol has been developed to simulate the condition of photolysis of any compound on leaf surface. The cutin substances were extracted from the leaf of crop or weeds. To simulate the leaf surface, a thin film of required thickness was made with the extracted cutin on glass surface. This thin film was then coated with herbicides to be tested and was irradiated under artificial UV-light and under sunlight, separately to study the rate of photolysis and the formation of photoproducts, respectively. Here, 2,4-D ethyl ester (2,4-D EE) and isoproturon were taken as test herbicides. Using this technique it was found that the cutin of rice slowed down the degradation process of 2,4-D EE by quenching the photolysis and the cutin of *Phalaris* and *Avena* imparted a similar effect on the photolysis of isoproturon. Photoproducts isolated from the photolysis of 2,4-D EE on different surfaces are: 2,4-D, 2,4-dichlorophenoxy acetyl chloride, chlorophenoxy acetic acid ethyl ester, and 2,4-dichlorophenol. Photoproducts isolated from the photolysis of isoproturon on different surfaces are: 2-amino-5-isopropyl-*N*,*N*-dimethyl benzamide, 4-isopropyl phenyl isocyanate, and 1,1-dimethyl-3-methyl-3-isopropyl urea. All these degradation products are herbicidally less toxic.

Details of application methodology

- **Extraction of leaf cuticle :** The process of extraction of epicuticular waxes from rice, wheat, *Echinochloa colona, Phalaris minor* and *Avena ludoviciana* was standardized. Chloroform and dichloromethane were found to be the most suitable solvents for the extrtaction of epicuticular wax. The extracts were further treated with activated charcoal to expel the co-extractives (pigments, etc) to obtain purified cuticular wax.

- **Preparation of thin cutin surface :** Three mL of the wax solution in chloroform (1 mg/mL) was placed on glass surface, and the solvent was allowed to evaporate to obtain an uneven thin film. Film was then placed in the oven (110 ^{0}C) for a few minutes to allow a better homogenization of the wax. Such thin-films of cutin of different sources were utilized in the study of photolysis of 2,4-D EE and isoproturon.
- **Coating of herbicides on thin-films :** The processes of coating the thin-film of cutin with 2,4-D EE and isoproturon were standardized. In this case, water could be the best solvent. But the solubility of 2,4-D EE and isoproturon in water is negligible. Other solvents like hexane, chloroform, and acetone dissolve the cuticular waxt to various extents. Methanol was found to be suitable for this purpose, as it had minimum interference with waxes.
- **Exposure of thin film under UV and sunlight :** The rate kinetic studies on the photolysis of 2,4-D EE and isoproturon on the extracted wax were carried out under UV-light and sunlight. Irradiation of the herbicides on cutin surfaces and on glass surface was also carried out under sunlight for the study on photoproduct formation.
- **Extraction of irradiated samples at different time intervals :** The irradiated samples of different time-interval were extracted in chloroform; cleaned-up by silica gel-based column chromatography; dried; and diluted to required volume in suitable solvent (hexane for 2,4-D EE and methanol for isoproturon).
- **Isolation and purification of degradation-products :** Major degradation products were isolated by preparative thin-layer chromatography and further purified by crystalisation.
- **Analysis of samples for rate kinetic :** For rate kinetic study, the samples of 2,4-D EE were analyzed by GLC method and of isoproturon by HPLC method. For structural elucidation, the purified products and products in extract were finally characterized by LC-MS/MS.

The benefits: This technique may be useful to understand the photochemical behaviour of any herbicide on leaf surface.

9.10 Technique to study crop-weeds interaction at elevated CO_2

The problem: Rapid global industrialization during current and last century resulted in production of greenhouse gases. CO_2 is major contributor of greenhouse gases which, being a primary substrate for photosynthesis, may have a significant impact on plant metabolism. Changes in any climate factor influence not only the performance of individual organism but also can impact interactions with other species i.e. weeds at different growth stages, hence, alter inter-species competition. There is a broad agreement that higher atmospheric CO_2 concentration stimulates photosynthesis more in C_3 than C_4 plants. The argument that rising atmospheric CO_2 concentration will reduce weedy competition because most of the weeds possessed C_4 photosynthetic pathway seems to be unrealistic as number of C_3 weed species are already present within agronomic system and posing a serious threat to production. In most of the studies in past, either open top chambers (OTCs), environmental chambers/greenhouses or gradient tunnels have been used to study the effect of high CO_2. Advent of the Free Air CO_2 Enrichment (FACE) technology enabled us to study the weed-crop interaction with precision and will provide some definite insights towards the physiology of crop-weed interactions.

The technique: In FACE the simulation of elevated level of CO_2 is achieved in the open fields by artificially injecting CO_2 gas in a controlled manner. FACE facility is designed and installed for the purpose of CO_2 enrichment to study crop-weeds interactions to elevated CO_2. Basically, FACE unit at DWR consists of three octagonal structures (each having area $\approx 50\ m^2$) out of which one can be used for ambient CO_2 (395 ± 5 ppm) while remaining two can be used for CO_2 enrichment (550 ± 50 ppm). Small weather station is also installed at the site which provides every second data on various parameters like wind speed wind direction, temperature, radiation and relative humidity. Based on input data from weather station, CO_2 supply is controlled by the master control system and CO_2 concentration is precisely maintained within the rings which can be measured by infra red gas analyzer (IRGA) installed at the centre of experimental area of each treatment.

Different components of Free Air CO_2 Enrichment (FACE) facility

Control and precision of this system is based on very fast feedback (every second) from weather station and CO_2 analyzer to master control system as input which analyze data every second and send the signal to PID valve system which is responsible to control the direction of flow and concentration of CO_2 into the experimental rings. To study crop-weeds interaction in FACE facility, mixed population of crop and weeds species must be maintained at uniform level (similar to the actual field conditions) in each in rings meant for ambient and elevated CO_2 exposure. Usually, enrichment of CO_2 needs to be started from emergence of sowing of crop continued upto its maturity of during light hours. Depending upon the objectives and target parameters of the study, exposure of the CO_2 can be modified by including night hours as well as by altering the duration of exposure (short-term/long-term). In addition to CO_2, secondary treatments like water stress, and nutrient level and herbicides can also be imposed. Unlike other techniques like open top chambers and growth chambers, gradient tunnels etc., this technique do not require any enclosure and provide absolutely open environment similar to the field conditions (except CO_2 concentration), hence, artifacts like change in temperature and humidity can be easily avoided in experimental area.

The benefits: FACE studies are fully open air and have many advantages over experiments conducted in controlled environment chambers, open-top chambers (OTCs) and gradient tunnels. FACE allows the study of ecosystem (almost similar to the natural conditions) and does not modify the plants' interaction with light, temperature, wind, precipitation, pathogens and insects. Large size of FACE plots, allows the integrated measurement of many plant and ecosystem processes simultaneously in the same plot, and avoids many of the problems associated with edge effects prevalent in pots and OTCs. It enables more experimental plant materials to be harvested without compromising the experiment, hence allows carrying out studies throughout life cycle. In addition to crop-weeds performance, interactions with soil microbes, pathogens and insects also possible to study even at micro level.

Limitations: FACE facility provide the most realistic mimic of a future elevated CO_2 atmosphere, it nevertheless has its limitations too. Maintenance of FACE facility is cumbersome task as it involves lots of electronics and electronic components which need expertise at high precision. Further, problem complicated during rainy season as atmospheric humidity rise which create problem with electronic components of the FACE facility. Operational expertise is required to conduct experiments in FACE facility. In facility available at Directorate, experiments can be conducted only at two CO_2 concentrations (ambient and 550 ppm). Future FACE experiments should also consider multiple levels of elevated CO_2, ranging 250 to 600 ppm. This would allow more accurate scaling of physiological results and validation of ecosystem models. In addition to CO_2, provision to alter level of temperature and ozone would further provide deeper and realistic insights as far as crop-weeds interaction is concern. Input cost in terms of CO_2 enrichment is high; hence some cheaper technique for elevation of CO_2 is definitely on demand.

Open Top Chambers (OTCs) for exposure of crop and weeds to elevated CO_2 and temperature

9.11 Technique to study combined effect of elevated CO_2 and temperature on crop-weeds interaction

The problem: Global climate change is expected to alter many elements of the future crop production. Atmospheric CO_2 concentration and average temperature will be higher in coming years. Different climate change drivers, their inter-relatedness and possible impacts on crop production need to be evaluated. Several studies have been carried out on the effect of individual factor (either elevated temperature or elevated CO_2), however not much information are available on how these factors in combination will impact on crops, weeds and interaction between these two. A modified version of Open Top Chambers (OTCs) is developed at Directorate which enabled us to study individual and combine effect of elevated CO_2 and temperature on crop-weed interactions.

The technique: OTCs consist of metallic structure covered with transparent vertical side-walls of polycarbonate. Top of the chambers kept open to avoid any build of temperature due to enclosure. With the help of timer device, CO_2 is infused from cylinders into the chambers which is circulated with the help air blower. All the supply is controlled and maintained by controller and with timer device. CO_2 concentration inside the chamber is continuously monitored by CO_2 analyzer. To elevate temperature, a heating device is fixed with temperature regulator. Again this device is connected with timer device which by on/off mechanism help in maintaining the desired temperature inside chamber. Chamber is of 3 m diameter which is sufficient to conduct experiment normally in pots. Whole unit consists of four uniform circular chambers. One chamber maintained at ambient condition, second and third chamber are modified for elevated temperature (ambient + 3 °C) and elevated CO_2 (550 ± 50 ppm), respectively. In fourth chamber, combination of elevated temperature (ambient + 3 °C) + elevated CO_2 (550 ± 50 ppm) is maintained. Some of the components of OTC unit are illustrated in on-site photograph as shown below:

The benefits: OTC is good for conducting pot experiment with elevated CO_2 and temperature. Cost of inputs (CO_2 and electricity) is less as compared to FACE facility. Secondary experiment like drought stress also can be maintained. In this design, combined effect of more than one factor (CO_2, temperature, drought, salinity etc.) can be studied.

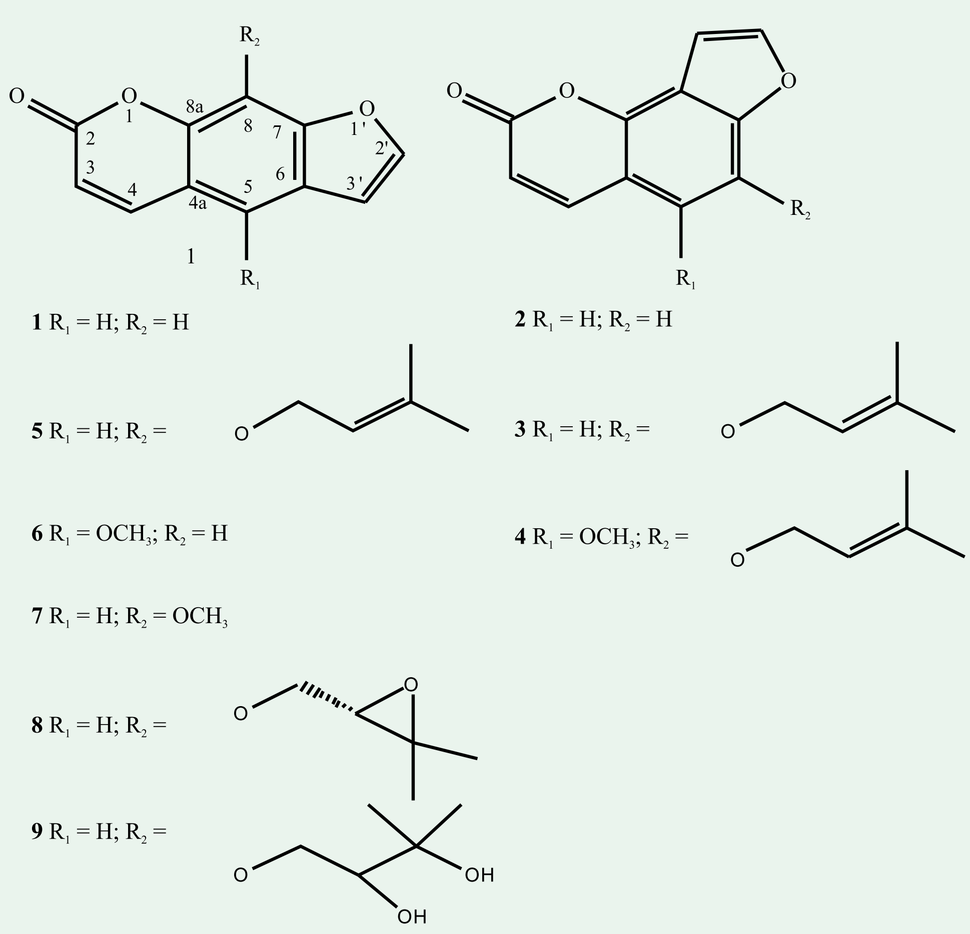
Limitations: It is small facility in which only small scale experiments (most in pots or trays) can be conducted. Further this facility lacks automation which required manual interventions. Due to lack of space, crop-weeds interaction study is difficult to conduct.

9.12 A state of art bioassay guided technique for the isolation of phytotoxic furanocoumarins

The problem: Worldwide efforts are being made to reduce the heavy reliance on synthetic herbicides because of environmental and human health concerns. The decline in the number of new synthetic herbicides derived from traditional screening programs and the increasing spread of weed resistance to synthetic herbicides provides impetus for natural products as important alternatives for herbicide development. Some plant secondary metabolites possess a wide diversity of bioactivity, including phytotoxicity. These phytochemicals might be used directly or as a template for new synthetic pesticides.

The technology: In this technique, a state of art phytotoxic bioassay guided technique for the isolation of phytotoxic furanocoumarins from *Semenovia transiliensis* plants was demonstrated. Air dried powder of shoots of *S. transiliensis* were extracted with MeOH / CH_2Cl_2 to get extractable. Silica gel column chromatographic fractionation of the MeOH/CH_2Cl_2 extract guided by *Lactuca sativa* and *Agrostis stolonifera* bioassays were used to identify and isolate the phytotoxic fractions. Phytotoxicity was qualitatively evaluated by visually comparing the amount of germination of the seeds in each well with the untreated controls after 7 days. The qualitative estimate of phytotoxicity was evaluated by using a rating scale of 0–5, where 0 is no effect and 5 is no germination or growth.

Purification yielded psolaren (m/z 187 $[M+H]^+$, $C_{11}H_6O_3$), isopsolaren (m/z 187 $[M+H]^+$, $C_{11}H_6O_3$), heratomin (m/z 271 $[M+H]^+$ $C_{16}H_{14}O_4$), isopentenyloxyisobergapten (m/z 301.31$[M+H]^+$, $C_{17}H_{16}O_5$), imperatorin (m/z 271 $[M+H]^+$, $C_{16}H_{14}O_4$), bergapten (m/z 217 $[M+H]^+$ $C_{12}H_8O_4$), xanthotoxin (m/z 217 $[M+H]^+$, $C_{12}H_8O_4$ 216.04), heraclenin (m/z 287 $[M+H]^+$, $C_{16}H_{14}O_5$) and heraclenol (m/z 305 $[M+H]^+$, $C_{16}H_{16}O_6$), respectively as a pure white powdery compounds. Mode of action of xanthotoxin and bergapten is found to be associated with inhibition of cell division.



Phytotoxic compounds isolated from *Semenovia transiliensis*

The benefits: The state of art phytotoxic bioassay guided technique was found effective and robust for the isolation of phytotoxic planar furanocoumarins which can also be equally useful to isolate furanocoumarins from other candidate plants. These compounds are well known to have biological activity as fungicides, antifeedants, and insecticides. These compounds are known to bind DNA and can be exploited to use as non-selective herbicides.

9.13 A specific design of lysimeters to determine herbicide leaching risk

The problem : Though agrochemicals perform an important role in managing pest and disease, their indiscriminative use may result in filtering of toxic chemicals deep into the soil that contaminate ground water. To evaluate downward movement of targeted chemicals/ in soil, results of short-term laboratory and field experiment with simulated rain models are commonly used. However, the environmental conditions in these experiments are reasonably different from natural field conditions in which biological, chemical, and physical processes occur in a complex soil structure, mostly under no equilibrium conditions apart from problems to dig or fix in the soil profile and hard to remove and these even do not have provision to demonstrate leaching of chemicals at particular rain. These lysimeters have problem of collecting infiltrating water (leachates) and soil sampling at various depths. Therefore one way to get around all this complexities is to use a more specified feasible and applicable lysimeter design, through which not only calculate total rain received during entire experiment and describe accurately affects of leaching on soil physico-chemical properties, but correlate climate factors, rainfall intensity on leaching of chemicals through soil profile as well. Therefore a specified new design of lysemeter was made and constructed under field conditions to assess leaching and risk of ground water contamination due to herbicide use.

The technology: The technology relates to a specified design of lysimeters of RCC cemented which are fixed vertically to assess precise downward movement of targeted herbicides at various soil depths under field conditions receiving natural rains. In this technology, a specified provision was made to each 25 cm distance up to the depth of 225 cm to draw and collect soil samples with the help of long spatula. To validate this technology, various herbicides were applied individually to the surface of lysimeters in *kharif* and allowed to receive natural rains and evaluated for leaching potential. A specified sampling site of 5 x 5 cm size was made in each lysimeter which were covered with compact iron cover. A special provision was made to collect leachates in the plastic canes of 25 L capacity attached on-site to the drain of each lysimeters. Provision was made to evaluate various physico-chemical parameters and leaching index in relation to herbicide application using these lysimeters.

The benefits: Field lysimeter are found good alternatives to PVC columns/small lysimeters used in laboratory/field experiments. This technology has an application for evaluating leaching and transport of targeted herbicides as well as its applicability may be explored for other chemicals, which are considered to be the most likely to contaminate soil and leach to groundwater. Using these lysemeters, leaching potential of many herbicides such as cyhalofop-butyl, tribenuron, imazethapyr, oxyfluorfen, pyrazosulfuron-ethyl, pretilachlor etc. was characterized. These lysimeters provided strength, durability and easy soil and rain water/leachates sampling options based on demand of experiments without disturbing soil profile. The same design can also be used for laboratory and yard conditions for small scale applications.

An overview of field lysimeter having provision for collection of soil samples and leachates.

9.14 A field design for evaluating unintended spray of herbicides on non-targeted organisms

The problem: The persistence of herbicides in the aquatic body is important as well as their ultimate fate. A side effect of herbicide use may result in unfortunate consequences to the non-target organisms. Many studies demonstrated ailing effect of herbicides on fishes and other aquatic organisms. Herbicides can enter water sources mostly through drift, runoff, soil erosion, leaching. Therefore, due to environmental concern there is an obvious need to develop a technology which can evaluate adverse effect of herbicides on non-target organisms as a result of unintended spray. Therefore a high thought off, field specific plot-pond design for evaluating suspected effects of herbicides on non-targeted organisms and water quality was developed and constructed at fixed site at Directorate of Weed Research, Jabalpur.

The technology: The technology relates to a high thought off specific field plot-pond design at fixed site at Directorate of Weed Research, Jabalpur to assess any detrimental effect of herbicides due to runoff on soil biota, water quality and fishes under field conditions. In this technique, herbicides are applied to the crops at recommended doses and through runoff and irrigation, they are allowed to enter to the adjacent ponds. This allows to detect herbicide residues in soil, plants, water and fishes and any detrimental effect of herbicides on non-targeted organism such as fishes, microbes, earthworm etc. Fingerlings of freshwater fishes were released in the ponds one month before application of herbicide in wheat and rice crop in *Rabi* and *Kharif*, respectively. Carfentrazone, fenoxaprop-p-ethyl, pinoxaden, metsulfuron, sulfosulfuron, and clodinafop-propargyl were applied in *Rabi* whereas pinoxsulam, pyrazosulfuron-ethyl, pretilachlor, butachlor, anilofos and oxyfluorfen were applied in *Kharif* at recommended doses in consecutive years to control grassy and broad leaved weeds. Soil, plants, water and fishes samples were analysed to evaluate fish mortality, water quality and residues of applied herbicides.

Dissipation of herbicides in wheat and rice soil followed first order rate kinetics. Fish mortality and toxicity symptoms were recorded initially in the pond where herbicides entered through runoff water in *Kharif* season. Several metabolites/transformation products of pyrazosulfuron, pretilachlor, fenoxaprop-p-ethyl and penoxsulam were detected from soil and pond water which were identified by LC/MS/MS.

The benefits: This technology was found effective to evaluate detrimental effects of several herbicides on non-targeted organisms. This technology has an application for evaluating transport of targeted herbicides or other class of pesticides, as-well-as its applicability may be explored for other chemicals, which may contaminate soil, and reaches to aquatic sources through runoff and leaching. Using this technique of plot-pond design, suspected detrimental effects of many herbicides such as, carfentrazone, fenoxaprop-p-ethyl, pinoxaden, metsulfuron, sulfosulfuron, clodinafop-propargyl, pinoxsulam, pyrazosulfuron-ethyl, pretilachlor, butachlor, anilofos, almix, pendimethalin, oxyfluorfen etc. were characterized on fishes. This technique also provided a noble way to assess fate of the herbicides in a paddy based ecosystems.

An overview of a specific pond-plot design at experimental field

10 Technique for dissemination of weed management technologies

10.1 Kisan mobile sandesh through SMS

The problem: Information about an existing innovation or a technology is pre-requisite for adoption of that innovation or technology by the farmers. There is no dearth of innovations in the field of agriculture but their timely transfer to the needy farmer is very important. Several technical information or innovations on weed management have been generated by our research system for increasing agricultural production and productivity. The farmers have adopted some of them but still there are many, which have not reached to our farmers. To increase production and productivity of crops, it is necessary that developed technologies are made available to the farmers. Since mobile service is widely being used by the farming community, SMS through mobile telephone network, termed as 'Kisan Mobile Sandesh' was used as an ICT tool for rapid and wide spread dissemination of improved weed management technologies.

The technique: The Directorate of Weed Research, Jabalpur has started 'Kisan Mobile Sandesh' service through computer linked software in the year 2013 with the objective to disseminate the knowledge- based information on weed management to the stakeholders in no time. Location-specific and crop-specific advisory containing real time agricultural information on weed management is directly delivered to about 10000 registered stake holders through SMS in local language. It delivers customized knowledge during the initial period of cropping seasons to improve farmer's decision making ability so that they may make a strategy to timely manage weeds with suitable low cost technology to increase their productivity and profitability. Registration is free for all interested stakeholders of the country and can be done by sending an e-mail to dirdwsr@icar.org.in

The benefits: Farmers were highly benefited by this service, as it is evidenced by the fact that they expressed favourable attitude towards the timely information on weed management, and said that the messages were of much use to them. Several farmers have given feedback that the recommendations were applicable to their farm, and they also advised other fellow farmers to adopt the recommendations.

Limitations :

1. Frequent network breakdown
2. Unavailability of hindi / regional language font in software of some mobile instrument
3. Incapability of receiving long messages in some mobile instrument
4. It is not useful for the farmers who cannot handle mobile phone.

10.2 Dissemination and evaluation of weed management technologies through on-farm research cum demonstration

The problem: Improved weed management technologies are in great demand by the farmers due to the acute labour scarcity and high cost of manual weeding throughout the country. Unfortunately, there is not enough awareness among the farmers about technical know-how of improved weed management practices, even in areas which are not for away from the research institutions. Since, the conventional top - down extension system does not have much relevance with the felt need of farmers in their prevailing farming situations, it is necessary to transfer improved weed management technologies by following community based participatory approaches.

The techniques: Appropriate technologies are roots of agrarian development. However, any weed management technology cannot perform equally in every agro - climatic situation. On farm research cum demonstration of weed management technologies are mainly focused to test / evaluate already developed technologies in terms of location specific and need based sustainable crop and land use system, which is helpful to solve the most important and widely spread weed problems of farmers in a defined area within their farming system. The OFR cum demonstration on weed management was initiated by the Directorate of Weed Research, Jabalpur in the year 2012 in six identified localities to understand farmers' problems and under take necessary interventions through farmer

participatory approach to develop, test and evaluate cost - effective solutions to their weed related problems.

The benefits: This OFR cum demonstrated programme on weed management has given very encouraging results, due to this a large numbers of farmers in the adopted and surrounded area have started using IWM technologies especially in rice, soybean, maize, greengram and wheat. It was also noticed that weed control in these crop were a highly cumbersome operation, and the traditional manual weedings requires large number of labourers, which were mostly not available at the time of need and was capital intensive. Compared to the traditional manual weeding that requires around Rs. 5000/ha, the cost of IWM practices was only Rs. 2000/ha and also showed an additional income of Rs.12000-15000/ha. These techniques were successful in convincing farmers that utilization of IWM technologies was essential to increase yield and economic benefit.

Precaution: Farmers input or opinions should be given due consideration while planning the OFR. While executing the plan, the extension personnel/scientists should exchange ideas with the farmers', exhibit sensitivity to the social/cultural aspects of the locality, and work with them as a team with dedication and honesty.

10.3 Dissemination of weed management technologies through farmer fair cum exhibition

The problem: In order to increase and sustain the food grain production it is essential to adopt intensive agricultural system with advanced technologies along with modern crop protection concepts. Weeds are major biological constraints and crop yield reduced even higher than those are caused by impact and diseases. Traditionally, weeds had been controlled manually, which is labour intensive and costlier. Further, it is time-consuming; as a result it is not possible to complete weeding operation within a desired time limit. This causes substantial loss in crop yield.

The technique farmer fair cum exhibition is an organized educational activity for involving and educating farmers by bringing together the farmers, scientists, extension workers, input agencies, developmental departments and non-governmental agencies on agriculture or allied aspects at a research institution, where the farmers can see, interact and gain first hand knowledge about the latest technologies and developments in agriculture and allied aspects. It enable the farmers to directly discuss with the specialists about the problems relating to agriculture and allied aspects, and also provide an opportunity to practically witness the new production technologies, to directly come in contact with input manufacturers, dealerof agricultural machinery and implements, and to know about the latest agricultural inputs, machinery, equipment etc. available in the market.

Twelve farmer fair cum exhibitions were organized by this Directorate during last 12 years. A large number of farmers from Jabalpur and adjoining districts and states participated every year in this programme. *Krishak Sangosthi* and field visits were also arranged. During the event, 50-55 stalls of various organizations demonstrated their latest technologies and products. Technical calendar on weed management, extension folders and related materials were distributed to the farmers, stakeholders and other visitors. Farmers always showed keen interest in different improved weed management technologies for cropped and noncropped situation, bio-control agent of *Parthenium*, weeding tools, spraying techniques, nozzle, spray machine, conservation agriculture technologies and were impressed with the performance of zero-till sown wheat, maize, chickpea and mustard at the experimental farm of the Directorate. During the programme, every year some progressive/achiever farmers were honoured.

Apart from the Former fair cum exhibition at DWR, Jabalpur, Directorate was actively participated in *Kishan Mela* (Farmers fair) (organized by other implikate), conference, seminar, 'symposium' and workshop and various societies. On the occassion recent development in used management and integrated weed management approach in different agro-ecosystems were advocated. Time to time recent weed management techniques and technologies were updted in official website of the Directorate for wide dessimination, accessibility and popularization among various stackholders.

If this is indicated that snapshot of website screen as photo may be inserted before hits will be visible.

The benefits: The farmers fair conducted in this Directorate:

- Developed a habit among large number of farmers to visit this Directorate frequently to learn about latest weed management technologies.
- Motivated the farmers to adopt the improved technologies by seeing its performance and profitability under field conditions.
- Clear hesitations doubts, superstitions and unfavourable attitude about the new practices like herbicides, conservation agriculture etc.
- It also helped the scientists to get feedback on recommended technologies.

Limitation:

- Farmer fair-cum-exhibition involves considerable expenditure on the part of the organizing institution. It needs a meticulous planning and involvement of different agencies, input firms and the research organizations.
- Fixing the farmer fair cum exhibition date based on convenience of farmers, facilities, climatic conditions and a good standing crop is an important aspect to be given due care.

10.4 Weed Manager : A mobile App for weed management

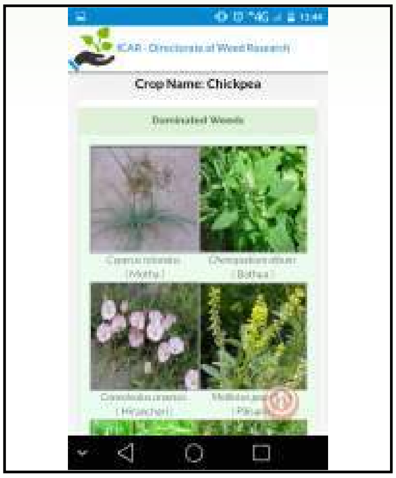
The problem: India is an agriculture based developing country. Agriculture is the primary occupation for 65-70% of Indian population who depends on agriculture for their living. In this competitive world, farmers need to produce more and more. To achieve higher agricultural production, farmers should be well equipped with latest technology and current information on day-to-day agricultural affairs. The data regarding farming are available from many sources such as print media, audio and visual aids, newspaper, TV, internet, mobile etc., but the formats and structures of data are dissimilar. Information dissemination to the knowledge intensive agriculture sector is upgraded by mobile-enabled information services and rapid use of mobile.

The technology: This mobile App named as 'Weed Manager' was developed by the ICAR-Directorate of Weed Research, Jabalpur. It is a user-friendly mobile app for farmers and agricultural industry professionals. This app allows users to scout crop name and identify common dominated weeds with possible control recommendations. The basic requirement to operate this app is have an Android device with net connectivity, and the software of 'Weed Manager' to be downloaded from Google Play Store. After completion of download, run the setup file for the installation 'Weed Manager' in a device. After complete installation a 'Weed Manager' icon will appear on mobile screen.

Features: It is totally menu-driven app, crops are grouped by season. User can select crop by selecting the season either rainy or winter or summer (i.e. *Kharif, Rabi* and Zayad). After selecting season, user can select the crop for weed management options in a particular crop. A screen will appear with dominated weeds with recommendations for a particular crop. Main features of the ICAR-DWR developed mobile app is as follows.

- The App is easily accessible and simple in use.
- It is designed for the end user
- From installed App, user can select its crop by selecting the season (i.e. *Kharif, Rabi* and Zayad)
- It provides multiple, high quality photos of dominated weeds along with recommendations for weed management in selected crop.

Icon of DWR WEED RECOMMENDED mobile application



Cost of technology: Free of cost

The benefits: It bridges the gap between the available knowledge of weed management and recent available / recommended weed management options. Mobile communications technology has quickly become the world's most common way of transmitting voice, data, images and services in the developing world. Mobile phones have many key advantages: affordability, wide ownership, voice communications, and instant and convenient service delivery. The cost of acquisition of a typical mobile phone is lower than PC. It is also easy to learn how to use a mobile phone, even for computer-illiterate people. This fact makes a mobile device the most appropriate medium to introduce latest agricultural technologies to users who are not computer savvy.

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Name	Formulation(s)	$LD_{50, \text{acute, oral,rat}}$ (mg/kg)	Toxicity code
Propaquizafop	10% EC	5000	Blue
Pyrazosulfuron-ethyl	10% WP	>5000	Blue
Pyrithiobac-sodium	10% EC	4000	Blue
Quizalofop-ethyl	5% EC	1670	Blue
Quizalofop-P-tefuryl	4.41% EC	1012	Blue
Sirmate	38% WP, 4% GR	1620	Blue
Sulfosulfuron	75% WDG	>5000	Green
Tembotrione	34.4% SC	>2000	Blue
Topramezone	33.6% SC	>2000	Blue
Thiobencarb	50% EC, 10% GR	1300	Blue
Triallate	50% EC	1100	Blue
Tribenuron	10% EC	>5000	Green
Trifluralin	48% EC	>10000	Green
Triasulfuron	20% WG	>5000	Blue
Chlorimuron + Metsulfuron	10+10% WP	>4000	Blue
2,4-D ethyl ester + Anilophos	32 + 24% EC	>500	Yellow
Pretilachlor + Bensulfuron	6.0+0.6% WDG	>5000	Green
Sulfosulfuron+Metsulfuron-methyl	75 +5% WG	NA	Green
Clomazone + 2,4-D ethyl ester	20% + 30% EC	>500	Yellow
Fenoxaprop -P-ethyl +Metribuzin	7.77+ 13.6% EC	NA	NA
Hexazinone +Diuron	13.2 + 46.8% WP	4888	Blue
Mesosulfuron -methyl + Iodosulfuron-methylsodium	3.6% WDG	>5000	Blue
Imazamox + Imazethapyr	35 + 35% WG	NA	Green
Pendimethalin +Imazethapyr	30 + 2% EC	>5000	Blue
Clodinafop-propargyl + Metsulfuron-methyl	15 + 1% WP	>5000	Blue
Carfentrazone -ethyl + Sulfosulfuron	20 + 25% WG	NA	NA
Sodium acefluorfen + Clodinafop-propargyl	16.5 + 8% EC	NA	Blue
Clodinafop-propargyl + Metribuzin	9 + 20% WP	NA	NA
Clodinafop-propargyl + Metribuzin	12 + 42% WG	NA	NA
Pretilachlor + Pyrazosulfuron-ethyl	6 + 0.15% GR	NA	NA

Notes:- LD_{50} acute, oral : Acute oral median lethal dose (LD_{50}) is the dose of a substance (here herbicide) or mixture of substances, in milligrams per kilogram of test animal body weight, which, when administered orally as a single dose, produces death of 50 percent of test animals (here rats) within 14 days.

- Red: Extremely toxic
- Yellow: Highly toxic
- Blue: Moderately hazardous
- Green: Slightly hazardous

Formulations

- CS : Capsule Suspension
- DF : Dry Flowable
- EC : Emulsifiable Concentrate
- EW : Emulsion, Oil in Water
- GR : Granule
- HN : Hot Fogging Concentrate
- SC : Suspension Concentrate
- SG : Water Soluble Granule
- SL : Soluble Concentrate
- WP : Wettable Powder
- WSC : Water Soluble Concentrate
- WG/ WDG : Water Dispersible Granules

Herbicides banned in India
Nitrofen, Paraquat dimethyl sulphate, Metoxuron

Herbicides withdrawn in India
Dalapon, Simazine

Herbicides with refused registration
2,4,5-T, Ammonium sulphamate, Calcium arsenate, TCA

Herbicide restricted in use
Dazomet, (The use of dazomet is not permitted on tea)

Source: Insecticides / Pesticides Registered under section 9(3) of the Insecticides Act, 1968 for use in the Country (as on 20/10/2015), Central Insecticides Board and Registration Committee, Ministry of Agriculture, Government of India.

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