DOE - GM PARTNERSHIP FOR A

JOINT U.S. - INDIA JATROPHA BIODIESEL RESEARCH PROJECT

Determination of the economic viability & technical feasibility of commercial Jatropha curcas production for generation of Jatropha oil as bio-fuel feedstock from wasteland

FINAL TECHNICAL REPORT

ON LIFE CYCLE IMPACT ASSESMENT OF JATROPHA CULTIVATION

Period:
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A collaborative project between
CSIR-CSMCRI,
General Motors and
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This project was in succession to two important projects with General Motors India, one being to test the biodiesel produced by the Institute in their vehicle and the other for setting up a Jatropha plantation on the premises of GM's Talegaon plant on wasteland. The valuable learning and linkages acquired in the course of these projects are duly acknowledged.

The project undertaken was a team effort wherein scientists from multiple disciplines, technical staff, students, and administrative staff of the Institute coordinated and worked in tandem. The internal team at CSIR-CSMCRI selflessly dedicated themselves towards the successful execution of the project, be it planning, follow up, or action. The Project Investigators, Dr. Arup Ghosh (Scientist & Acting Discipline coordinator, Wasteland Research Discipline) and Dr. Jitendra Chikara (Ex-DC, WLR), sincerely thank the whole team for their valuable contributions:

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Arup Ghosh

Jitendra Chikara

Executive Summary

Ever since it was demonstrated that *Jatropha* seed oil could be converted into a world class biodiesel and could run in unmodified stationary and mobile diesel engines with simultaneous reduction in emissions, it caught the attention of the world. The capability to grow this crop on wastelands added to its attractiveness. However, the single biggest challenge came in the form of the availability of adequate feed stock in the form of the *Jatropha* fruit. Adequacy of feed stock can only be possible if large plantations are cultivated and produce enough fruit. The people, world over, jumped into *Jatropha* cultivation without heeding to the need to first ensure quality germplasm and understand the agronomic requirements of the plants. As a result many plantations failed to give the required yield.

CSIR-CSMCRI had been researching *Jatropha* and had an end-to-end approach, i.e., it developed the best technology to prepare biodiesel and also worked towards the practical problems that it envisaged to be important for raising *Jatropha* productivity. It focused only on cultivation on wastelands as this was the only practical strategy, given the limited arable land India has and the risk of food security for the burgeoning population. While working in this direction, the Institute zeroed-in on a few germplasm, which gave consistently higher seed yield over several years. These germplasm were clonally propagated in large numbers to be raised in experimental plantations at different geographical locations in India. Many agronomic practices were developed as a part of these different projects. It was at this juncture that General Motors and the U.S. Department of Energy joined hands with CSIR-CSMCRI to further the work on *Jatropha*. A center of expertise for *Jatropha* was established and work was initiated to further refine the understanding regarding the best practices. Efforts were to be made to generate primary data, hitherto unavailable for wastelands, on which life cycle assessment studies were to be performed as a part of the project.

New plantations were raised at different places in Gujarat after acquiring wasteland from the concerned authorities, and simultaneously, data were also recorded from the older plantations. New plantations were raised using superior accessions by employing the best agro-practices developed. Efforts were made to find low cost solutions to overcome many significant problems envisaged in cultivating *Jatropha* on wastelands. Among them practical solutions were developed to suitably amend different categories of wasteland soils including shallow and rocky types and land with heavy textured soil. Strategies towards maximizing the use of irrigation water and water harvesting were worked upon. Studies to reduce the requirement of chemical fertilizers having a significant carbon foot print was done, and the *Jatropha* cake as a manure to *Jatropha* plantation itself was extensively studied. It was found that the *Jatropha* cake could improve the photosynthetic and water-use-efficiency in plants, simultaneously raising the yields of the plants. It also improved soil properties and was able to control some important pathogenic microbes. Seed yield >2 t/ha (3t fruits/ha) was realized from the plantation at Neswad, emphasizing that such yields in are practically feasible under wasteland conditions provided proper choice of germplasm and agro-practices are made, even in situations where rainfall is scanty and irrigation facilities are limited. The plantation received very little irrigation (~50 l water/plant/month for 5-6 months in a year) because there was not enough ground water supply. The use of liquid seaweed fertilizer, a patented foliar stimulant that has been proven in many crops, was extensively used throughout the entire plantation. Seed yield improvement of 22-40% was found by using this fertilizer. Although not considered to be important earlier, disease outbreak has turned out to be a major concern. *Jatropha* is very much affected by fungal attack and efforts were made to impart disease resistance in plants through a biotechnological approach.

Intensive data were generated for life cycle inventory creation, and a comprehensive life cycle impact assessment was done for all the steps up to the production of *Jatropha* oil and briquettes from whole fruits. LCA for biodiesel production (based on our patented process) and its use was, however, kept out of the scope of the present investigation on account of the shortened duration of the project, although the impact of its conversion is expected to be minimal. The LCA was done for the production of 510 kg *Jatropha* oil that can be obtained from one hectare plantation annually in addition to realizing 1.53 tonnes of *Jatropha* cake, the majority of which is used to offset chemical fertilizer requirement, although credit for the offset was not taken into consideration. Further, the productivity was assumed by averaging and extrapolating the present day *Jatropha* capsule (fruit) yield (3t/ha) over a period of 20 years, an assumption that seems to be realistic given the reported life span of *Jatropha* to be more than 50 years. Further, a conservative oil extraction efficiency of 25% has been taken, while extracting seed oil in traditional screw type oil expeller, since decentralized model of *Jatropha* biodiesel production is targeted. The production of briquette has also been included, which further is a source of bio-energy in solid form. The carbon sequestered in the standing biomass has been averaged over a period 20 years and has been accounted for. Carbon reduction on account of removal of native biomass before planting of *Jatropha* has also been similarly accounted for. The carbon sequestered by the *Jatropha* fruit (which yields oil, briquettes, and cake) has been excluded since the LCA has been limited to the production of these products, while also excluding their use phase. Life cycle assessment of *Jatropha* oil production reveals that upstream process of fruit production accounts for 64.6% of Global Warming Potential (GWP), whereas, downstream process including deshelling, oil extraction, and production of briquettes accounts for rest of it. The production and use of fertilizer is the single largest contributor in all impact categories (GWP, Acidification Potential, Eutrophication Potential and Ozone Depletion Potential), while electricity was the major contributor for all impact categories in all the downstream processes. The annualized per hectare impact under GWP for land preparation was only 4.9%, thus justifying the generous investment made in the beginning to amend the shallow and rocky land by digging deep trenches. Overall, the *Jatropha* system analyzed had a negative and beneficial balance with respect to GWP.

The significant outcomes and learnings from this project are:

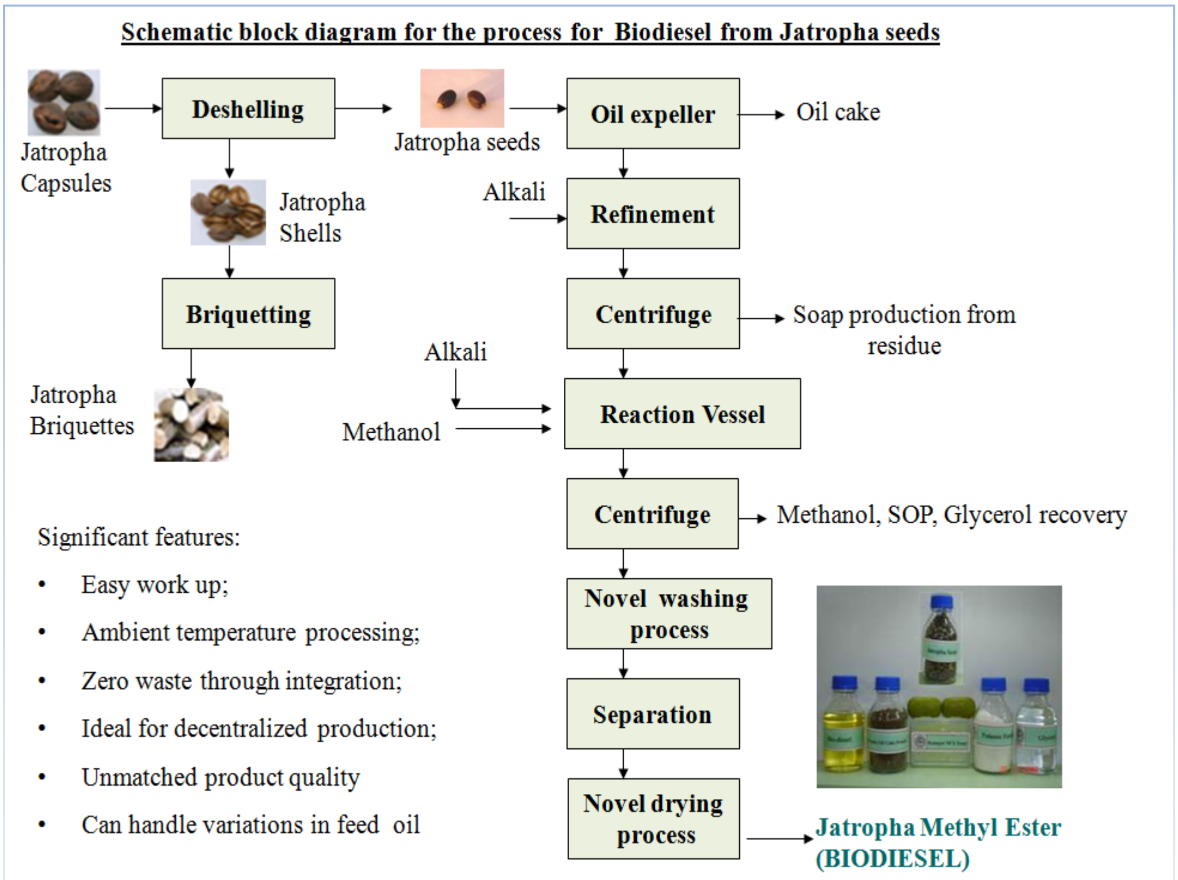
- Fifty hectare plantation was raised using clones of superior accessions and at the end of the project period, over 400,000 elite true-to-type cuttings have been generated, which can, at present, be given to interested stakeholders.
- Innovative ways of making shallow rocky wastelands or water-logged lands amenable to Jatropha cultivation was demonstrated on a large scale.
- Low cost water harvesting and efficient water use techniques were developed.
- Use of *Jatropha* cake as an organic manure not only was able to offset mineral fertilizer requirement but also improved the physiological and bio-chemical parameters in Jatropha, simultaneously raising the yields of the plants.
- Jatropha cake reduced the load of pathogenic fungus in the soil.
- Liquid seaweed fertilizer derived from *Kappaphycus alvarezii* significantly raised fruit productivity.
- Efforts were made to impart disease resistance in plants through biotechnological approach.
- LCA was performed, and an attractive carbon balance computed up to the production of the oil and briquettes from whole dry fruits was observed.

Project background

India is a net energy importing country with around 82% of the demand being met by imports (Petroleum statistics, 2011). Thus, it becomes a necessity to develop alternate sources of energy, both renewable and non-renewable. In order to overcome these issues, the Government of India (through the Planning Commission, 2003; National Policy on Biofuels, 2009) has recommended the use of biofuels either as blends or in neat form especially from *Jatropha curcas* owing to its suitability and potential of growing in marginal wastelands and degraded soils.

CSMCRI embarked on *Jatropha curcas* cultivation on wasteland as early as in 1996 with support from UNDP/TMOP. Thereafter, it undertook many projects with support from various government agencies including CSIR and corporate majors like DaimlerChrysler-DEG, and General Motors (India). During the course of these projects, efforts were made to find solutions to many of the challenging problems that were envisaged to be important in the context of utilization of wasteland, self-reliance, and sustainable mobility. CSMCRI proved the practicality of cultivating the plant on wasteland, which had not been done till that time. An important feat accomplished was forward integration to biodiesel preparation. The aim was to attain biodiesel quality that would allow its neat application in unmodified engines so that rural communities could directly utilize the local produce, besides the use of the fuel in advanced automobiles. The project in collaboration with DaimlerChrysler, University of Hohenheim, and DEG, Germany successfully completed its tenure and eventually, a novel biodiesel process was developed (US Patent No. 7,666,234 dated 23 Feb, 2010) by CSIR-CSMCRI, Bhavnagar with the following features:

- The process can handle variations in free fatty acid content of *Jatropha oil*.
- The process is based on conventional alkali catalysis albeit with innovations to overcome known limitations in post-trans-esterification work up.
- All the operations in the core process are at ambient temperature; heat is required only in the oil expelling process and for the processing of crude glycerol
- No external source of fuel is required for steam generation; instead it is obtained utilizing certain energy containing co-products generated in the process
- Yield of 94-96 % biodiesel with respect to refined oil
- Zero effluent discharge
- Other edible and non-edible oils, including waste vegetable oil, can also be used.



Biodiesel can, in principle, be prepared from any triglyceride such as vegetable oils. The key considerations are the quality of the biodiesel and the cost of manufacture. Another important aspect is the utilization of by-products to distribute costs. By-products of the *Jatropha* seed capsule include: the empty shell having coal-like calorific value, oil cake, soap, and glycerol. Effluent management is yet another important consideration, and, in our case, the spent catalyst is converted into a useful fertilizer.

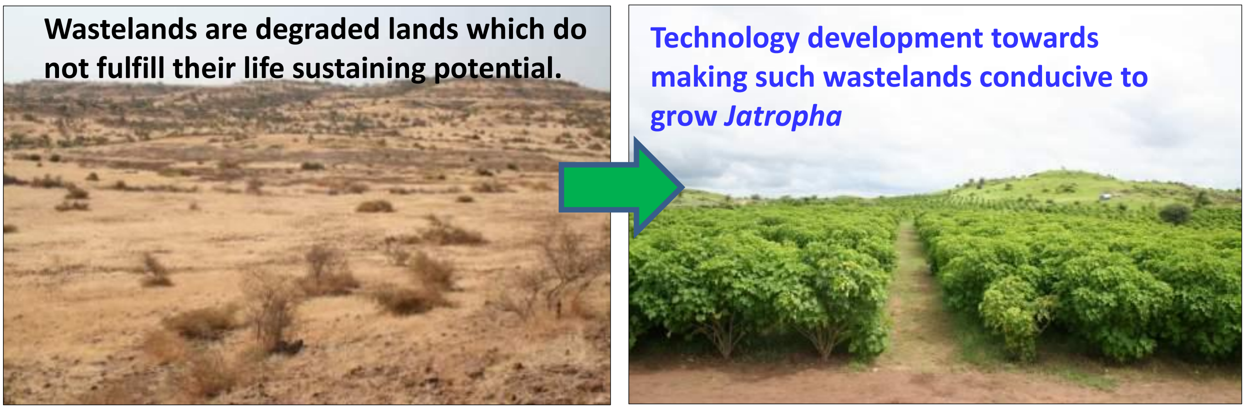
Jatropha biodiesel produced by the CSMCRI process complies with EN14214 specifications and can be used either in blended or neat form in unmodified diesel engines. The *Jatropha* biodiesel yielded the best engine performance among all biodiesels in a study conducted as part of the EU project.

The biodiesel outperformed other EN14214 grade biodiesels from rape seed oil, animal fat, coconut oil, and soybean oil in terms of engine power output, efficiency, and 16-days continuous running tests (tests conducted in all cases with neat biodiesel on HATZ 1D41Z 4 stroke engine; data courtesy Austrian Biofuel Institute). Emission test results (conducted jointly with ARAI, Pune) were also encouraging, particularly the dramatic reduction in hydrocarbon and particulate emissions. Detailed results of the tests have been published by DaimlerChrysler *("On Road Testing of Advanced Common Rail Diesel Vehicles with Biodiesel from the*

Jatropha curcas Plant", S. Mandpe, S. Kadlaskar, W. Degen and S. Keppeler, 2005-26-356, Proceedings of SAEINDIA Conference 2005). Following successful lowering of the pour point of the biodiesel, in collaboration with Lubrizol India, successful test runs of C class Mercedes cars were done by DaimlerChrysler India in Leh in August 2005 to ascertain the performance at high altitude in rarefied atmosphere of Khardungla pass, the highest (18,380 ft.) motorable road in the world.

CSMCRI licensed the know-how to Rajasthan State Mines & Minerals and a demonstration plant is operational near Udaipur since March 2006. RSMML reported excellent performance of the biodiesel in a Tavera van and also in running of mining equipment. CSMCRI too has been running its unmodified Toyota Qualis on neat biodiesel since August 2006 and over 220,000 km of running has been completed with mileage comparable to that with fossil diesel. No modification of any kind was made on the vehicle. Work by VRDE (Defense Research Development Organization, India (DRDO)) using our *Jatropha* methyl ester, points to excellent results on emission control even at a 20% blend. A one TPD zero effluent discharge plant was also installed and commissioned at DRDO in 2008 based on our technology.

A part of the conclusions emerging from our work is discussed in: ***"****Prospects for Jatropha Methyl Ester (Biodiesel) in India"* Ghosh et. al., *Int. J. Environ. Stud.* (Taylor & Francis, U.K.) – a special issue on India's future energy options (in the press). An article has also been published in *Nature ("Biofuel – The little shrub that could – may be," D. Fairless, Nature*, **449**, 2007, pp. 652-655) describing at length CSMCRI's pioneering work on *Jatropha*. The institute's pioneering end-to-end program and accomplishments were featured in a television broadcast by the German Television Channel, ZDF. The Institute's efforts on cultivation were also featured in a climate feedback blog hosted by *Nature* (K. Sanderson, nature.com, 17 Sep, 2009).



CSIR-CSMCRI plantation at Chorvadla raised by transforming wastelands
From: *Nature*, *449*, pp. 652-655, 2007

Ever since it was established that *Jatropha* methyl ester yields biodiesel of an exceptional quality, there has been a surge of interest in *Jatropha* across the globe, more so in view of the potential for avoiding the dilemma of “food vs. fuel.” However, it was emphasized that large scale cultivation of *Jatropha* yielding adequate feedstock remains the single most important issue that will ultimately decide success. As mentioned above, CSMCRI has played a pioneering role in promoting the concept of *Jatropha* cultivation on wasteland, and at the time of this project formulation had 30 hectares of such cultivation with different identified elite accessions. In-depth studies were being undertaken to identify the best agronomy practices including use of *Jatropha* de-oiled cake. CSMCRI also has several proven germplasm for which data existed on field performance both of mother plants and progenies over several years. Accession # IC-565735 was found to fare equally well both in Gujarat & Odisha states of India, and there were other germplasm being tested for their production potential in Gujarat. At Odisha, besides a yield in excess of 2 kg/plant for mature plants, the oil yield touched a maximum of 41% of whole seed weight. It was established that raising a plantation with selected cuttings would lead to a more productive plantation as compared with that by seedlings – even seedlings raised from seeds of elite plants. An important achievement in this direction was the success in raising true-to-type tissue culture plants from shoot tips.

CSIR-CSMCRI also collaborated with the Energy Research Institute, Heidelberg on generating preliminary data towards carbon life cycle analysis. Based on the latest technologies developed at the Institute for *Jatropha* cultivation on marginal lands, the energy output to input ratio was computed to be ca. 6 for production of *Jatropha* biodiesel and shell briquette.

It was in this backdrop that CSIR-CSMCRI began discussions with General Motors in 2005 and, thereafter, entered into an arrangement for supply of B100 *Jatropha* biodiesel to GM India for evaluation in their vehicles. Over 65,000 km of total running of the vehicles with biodiesel blend was carried out without any difficulty. Subsequently, the Institute set up a 2 acre *Jatropha* plantation on rocky terrain at their Talegaon factory in 2008. Thereafter, the US Department of Energy and General Motors, U.S. (GM) joined hands and entered into a partnership for the promotion of *Jatropha curcas*. In 2010, the two organizations entered into collaboration with CSMCRI to explore the highest possible productivity of *Jatropha* on marginal land utilizing the best germplasm and agronomy practices developed by the institute thus far. Through this collaboration, a detailed life cycle assessment of *Jatropha* cultivation and biodiesel based on the primary data was undertaken. Further, the project sought to address possible threats and vulnerabilities.

The project was formulated with the following aims:

Aim: The project aimed to demonstrate that *Jatropha curcas* can be raised on non-arable marginal lands with minimal agronomic inputs and produce significant quantities of *Jatropha* oil for conversion to biodiesel. This renewable fuel solution would go a long way towards realizing and sustaining multiple benefits across a diverse spectrum of interests:

- Offering a sustainable non-food source of energy, thus mitigating food-vs.-fuel concerns
- Enhancing energy security by reducing dependence on fossil fuels and costly oil imports
- Improving environmental quality by reducing auto-related criteria and GHG emissions
- Promoting economic development by creating employment, especially in rural areas

The original project tenure was initially for 5 years, but at a later stage, the time frame of the project was agreed to be shortened to 3 years due to internal reorganization at GM.

Project implementation

The project was operationalized in January 2010 after the agreement was signed between General Motors Corporation and Central Salt & Marine Chemicals Research Institute, Bhavnagar (a constituent lab of Council of Scientific and Industrial Research, New Delhi). The formal joint press announcement of the initiation of this project was made on April, 12th, 2010, which was followed by a visit of the GM officials to the existing plantation and the new sites (Figures 1, 2 and 3).

Figure 1 and 2. Press conference being held at Ahmedabad, Gujarat (left) Officials topping up the Chevrolet Tavera provided by GM with CSMCRI’s B-100 biodiesel (right).

Figure 3. Visit of GM officials to the existing *Jatropha* plantation on 13th April 2010.

During the course of this three year project, a *Jatropha* Center of Expertise was established in Gujarat, wherein 50 hectares plantation was established (20 ha at Kalol and 30 ha in Bhavnagar). In addition, existing plantations were also managed. Land was acquired for the entire duration of the Project (initially for 5 years), namely at Neswad, Bhadrawal (in

Bhavnagar district) and Kanod (in Panchmahal district). Existing *Jatropha* plantations were at Chorvadla in Bhavnagar district. Elite germplasm and best practices were employed to raise the plantations. Sustainable agronomic practices were improved or developed through experimentation to develop the optimum package of inputs necessary for high productivity from the plantation. The results of the project thus provides agronomic data, sustainability information like net energy balance vital to determination of the economic viability and technical feasibility of commercial *Jatropha* production for the generation of *Jatropha* oil as biofuels for feedstock. A life cycle impact assessment (LCIA) was also done for *Jatropha* cultivation and by-product recovery from the fruits in the form of *Jatropha* de-oiled cake (ploughed back into plantation), shell (converted into briquettes), and *Jatropha* oil. The LCA for the preparation of biodiesel and its use was, however, kept out of the scope because of shortened tenure of this project.

The data generated and experimental results inferred from this project have been presented and discussed in a manner that facilitates a comprehensive understanding of the learnings from all the activities carried out in logical sequence and also draw conclusions on the life cycle impact assessment of the best practices. Accordingly, details of the objective, scope, study design, system boundary, data and inventory analysis for the purpose of comprehensive LCA are described in the ensuing sections.

Although there are few reports on the assessment of life cycle analysis on the production of biodiesel from *Jatropha*, the base case assumptions on seed yield in most of the reports are based on unsubstantiated yields which were assumed because of a lack of sufficient primary data from mature plantations at that time and based on yields which to our best knowledge can be possible only upon cultivating the plants on fertile and arable lands. However, this would have a serious implication on food security, as the country is facing acute shortage of arable lands for cultivation which are needed to feed the burgeoning population. To the best of our knowledge, data sets that are required for LCA of growing *Jatropha curcas* on marginal and wastelands are lacking. Therefore, we focused our research on *Jatropha curcas* cultivation on wastelands and also modeled the environmental impacts associated under such a situation as a base case scenario.

Results and discussions based on activities undertaken

The *Jatropha* projects around the world started with much fanfare, but later expectations of very high productivity did not match with the reality. The reasons for these ranged from lack of site-specific sound agronomic practices including nutrient, water, pruning, spacing requirement, lack of proven germplasm in sufficient numbers, and lack of consistent data on year-on-year productivity. Besides, there were many myths attached with the *Jatropha* plant, such as the non-requirement of irrigation for its cultivation, ability to produce high productivity in shallow wastelands without any amendment, low gestation period and absence of insect pest and diseases for this plant. All of these myths raised the expectation and many countries in the world hurried into establishing plantations on any land they could grab with any seed material they could lay their hands on.

Suffice it to say that the downstream operations have been fully established including extensive testing in India and overseas of the engine worthiness of *Jatropha* biodiesel. The only way to successfully develop *Jatropha* biofuel in the country is to ensure adequate productivity with optimum level of inputs. CSIR-CSMCRI methodically worked to solve many of these problems and inched towards finding a cost-effective pragmatic solution for *Jatropha* cultivation. This project focused on the following:

1. Ways to make shallow wastelands suitable for cultivation of *Jatropha.*
2. Selection of elite accessions having proven year-on-year seed and oil productivity.
3. Generation of true-to-type quality planting material through cuttings and tissue culture.
4. Effective methodologies to conserve water in the regions where rainfall is scanty and maximize water use.
5. Maximize productivity without deteriorating soil fertility and study the effect as cake as manure in *Jatropha*.
6. Efforts towards finding solution to problem of fungal disease in *Jatropha*.
7. Year-on-year data generation on different accessions/germplasm identified as elite and hybrid production.
8. Data generation for life cycle inventory and life cycle impact assessment.

The activities which were undertaken are described below.

Establishment of new plantations:

Land survey

Gujarat is situated on the west coast of India. The state is bound by the Arabian Sea on the west, Pakistan and Rajasthan in the north and northeast respectively, Madhya Pradesh in the southeast, and Maharashtra in the south. The total geographic area of Gujarat is 196,024.sq. km out of which 21,350.38 sq. km is considered wastelands which accounts for 10.89% of its geographical area (Wasteland Atlas of India 2010, using Resourcesat-1 LISS-III images). Land with dense scrub is the major wasteland category which covers an area of 11,614.83 sq. km. In terms of the change in the wastelands as compared to 2003, there has been an increase in the wastelands of 972.64 sq. km. Land with open scrub is the another type of wasteland which is available in large areas in Gujarat and has been targeted for cultivation of *Jatropha* under the project.

A number of sites falling under the above category in Gujarat were visited to identify potential land for *Jatropha* cultivation on 50 ha area on wastelands. Three sites were found suitable. The required permission from the local village level governing agency (Panchayat) was obtained for all the 50 ha of land at Neswad, Bhadrawal, and Kanod in the state of Gujarat. All the three patches of land were wasteland with scanty cover of perennial as well as annual bushes. Coordinates of these locations where new *Jatropha* plantations were successfully established are as follows:

a. Village Bhadrawal (Bhavnagar district) (Altitude- 280 ft N 21° 29’ 530’’ E 072° 00’ 167’’)
b. Village Kanod (Panchmahal district) (Altitude- 296 ft N 22°41’ 376’’ E 073° 29’ 649’’)
c. Village Neswad (Bhavnagar district) (Altitude-314 ft N 21° 30’ 494’’ E 072° 02’ 185’’)

Estimation of initial biomass in initial vegetation

The sites were visited to gather data regarding native vegetation prior to raising *Jatropha* plantations. The land at all the sites typically had sparse vegetation and the different species of plants found were identified and enumerated **(Figure 4 and 5)**. The initial vegetation was denser in Kanod than that of the Bhadrawal site. Observations on the plant populations were done using eight quadrates of size 20m x 20m at each of the sites. Thereafter, the vegetation was removed along with roots, species-wise, to assess the initial biomass to be replaced by the *Jatropha* plantation. Fresh biomass was collected, weighed **(Figure 6a & b),** and samples of biomass were drawn to be oven dried for further analysis to determine dry matter production and carbon status.

Figure 4 and 5. Initial pictures of the land before taking up cleaning and land preparation work at Bhavnagar site (left) and Kanod, Panchmahal (right) [Picture, 2nd Feb 2010].

Figure 6a and 6b. Species-wise native vegetation being uprooted (left) and weighed to assess the initial biomass, (right).

Bhadrawal, Bhavnagar site:

At the Bhadrawal site of Bhavnagar district, six species, namely *Acacia leucophloea, Acacia nilotica, Prosopis juliflora, Ziziphus mauritiana, Cassia fistula, and Maytenus senegalensis* were prominent. It was found that *Cassia fistula* and *Zizyphus mauritiana* mostly predominated the area with 1034 and 794 plants/ha, respectively **(Figure 7)**, but the bulk of the total dry biomass (above + below ground) of the area (6.98t/ha) was contributed by *Acacia leucophloea* (55.4%) and *Acacia nilotica* (29.3%), respectively **(Figure 10 left)**. At the Bhadrawal site, the total carbon in the native biomass amounted to ca. 3.05 t/ha of which 2.33 t/ha was from the above-ground vegetation **(Figure 11 left)**.

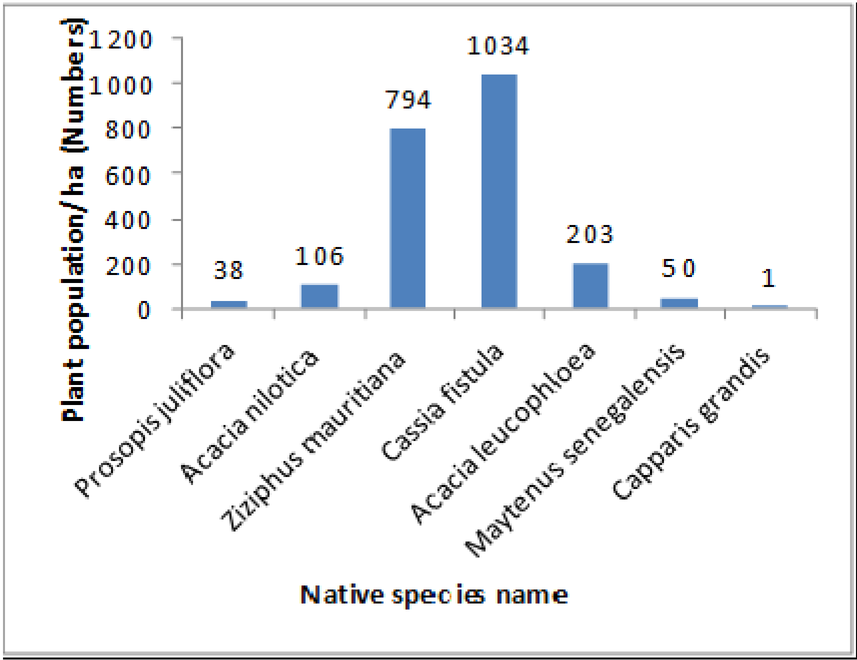


Figure 7. Initial native plant populations predominant in the area prior to clearing off of the plantation at Bhadrawal, Bhavnagar, Gujarat.

Kanod, Panchmahal site:

Summing up the studies on the native biomass of Kanod site, it was found that among the perennials, *Cassia auriculata* and *Jatropha gossypifolia* mostly predominated the area with mean numbers of 1863 and 753 plants/ha, respectively **(Figure 8)**, but the bulk of the total dry biomass (above + below ground) of the area (3.85 t/ha) was contributed by *Prosopis juliflora* (72%) and *Cassia auriculata* (11.9%), respectively **(Figure 10 middle)**. The total amount of carbon in the perennial native biomass displaced amounted to ca. 1.62 t/ha of which 0.77 t/ha was from the above-ground vegetation, while the remaining 0.86 t/ha was attributable to below-ground biomass.

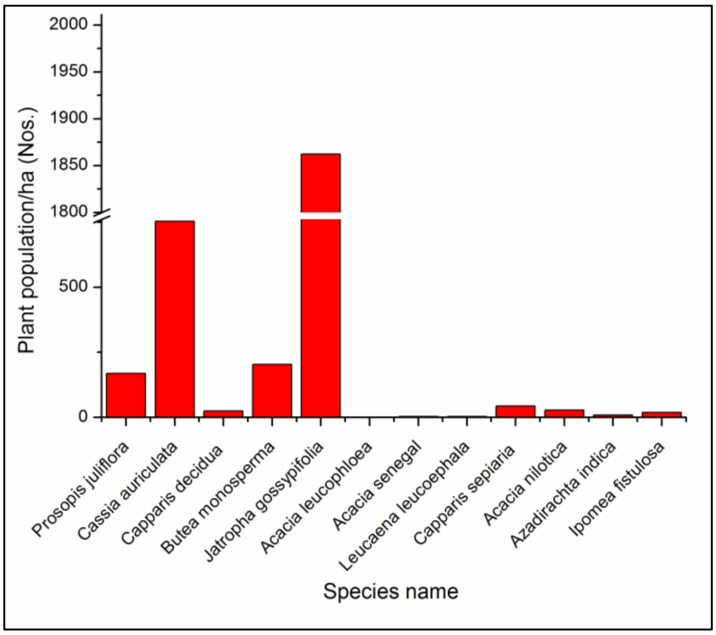


Figure 8. Initial native plant populations predominant in the area prior to clearing off of the plantation at Kanod, Bhavnagar, Gujarat.

Neswad, Bhavnagar site:

Among the prominent species at the Neswad site, *Zizyphus mauritiana* was the most predominant with 726 plants/ha **(Figure 9)**, but the bulk of the total biomass of the area (0.715t/ha) was contributed by *Acacia leucophloea* which weighed 0.35 dry tons per hectare. At the Neswad site, it is apparent that not much loss in terms of total carbon (0.3t/ha) occurred as there was sparse native vegetation.

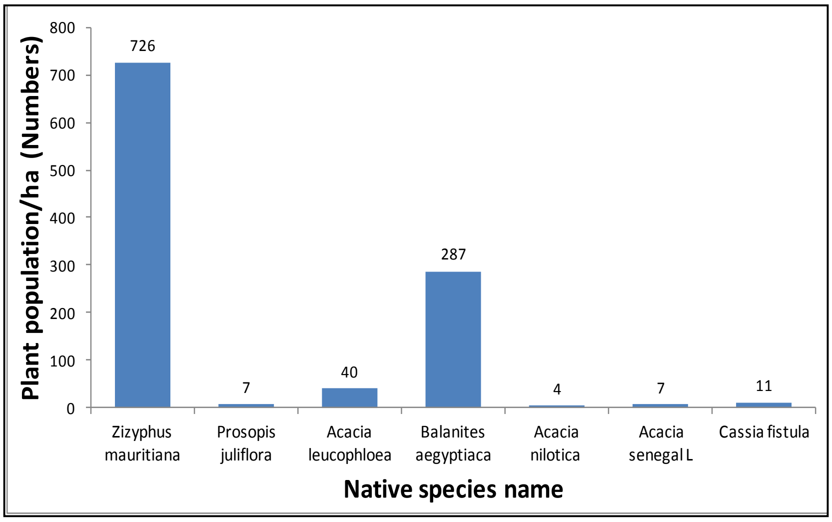
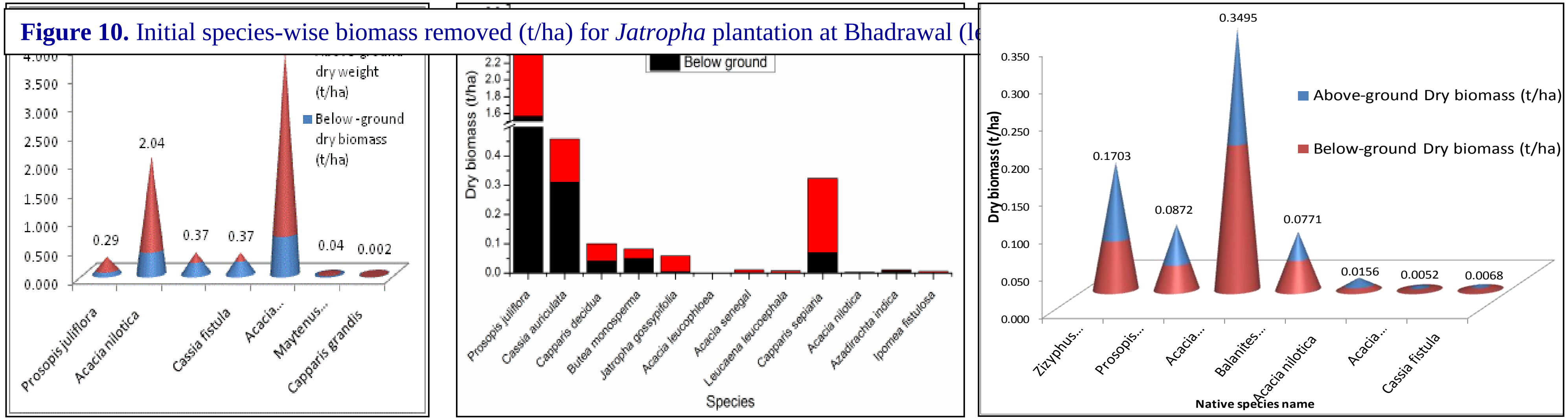


Figure 9. Initial native plant populations predominant in the area prior to clearing off of the plantation at Neswad, Bhavnagar, Gujarat.

Physico-chemical properties of soil

Collection of soil samples: The soil samples were collected from a number of sampling locations at all the sites from different soil depths (0-60cm). These samples were dried and ground to pass through a 2mm sieve. These samples were further analyzed for physico-chemical characteristics. Preliminary studies revealed the soils at Bhadrawal to be calcareous at lower soil strata. Physical and chemical properties of soil including macro-nutrient composition of all the *Jatropha* sites in this project were determined. At Bhadrawal, the soil texture of the upper solum (0-30 cm) varied from sandy loam to sandy clay loam, while the bottom layers comprised of loamy sand to sandy loam. The soils at Kanod were found to be heavier in texture with mostly sandy clay loam to clay up to 60 cm of solum. The soils of Neswad and Chorvadla were mostly sandy loam. The final consolidated results for all these soil parameters are presented in **Tables 1a, b, c**. **Table 2** indicates the critical level of nutrients in soil and plants.

Figure 10. Initial species-wise biomass removed (t/ha) for *Jatropha* plantation at Bhadrawal (le



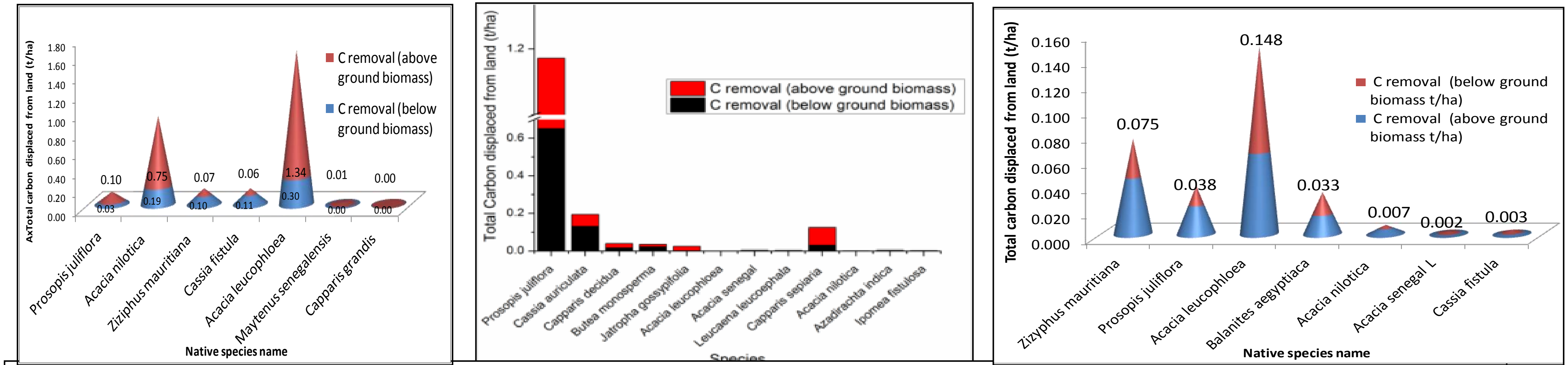


Figure 11. Species-wise total carbon displaced (t/ ha) on account of uprooting the initial native vegetation at Bhadrawal (left), Kanod (center) and Neswad (right) in Gujarat.

Table 1a. Physical properties of soil under *Jatropha* cultivation at different sites

S. No.	Location of the sites	Range, and Mean values of soil physical properties						
		% concretion (in 0-40 cm)	Sand (%)	Silt (%)	Clay (%)	Texture	EC(dS/m)	pH
1	Neswad	16.3-20.9 18.1	71.9-80.3 77.0	11.7-15.8 13.4	7.2-12.3 9.6	Sandy loam	0.09-0.13 0.11	7.0-7.6 7.3
2	Bhadrawal	19.1-22.3 20.7	68.5-76.3 72.4	16.7-21.6 19.1	7.0-9.9 8.5	Sandy loam to Sandy clay loam	0.18-0.34 0.23	7.3-8.1 7.7
3	Kanod	2.1-4.8 3.4	25.8-29.9 27.8	11.9-16.3 14.1	57.9-58.1 58.0	Sandy clay loam	0.07-0.44 0.20	6.8-7.3 6.9
4	Chorvadla	2.2-55 27.4	57.5-76.2 66.8	14.7-30.0 20.7	9.0-17.8 12.4	Sandy loam to sandy clay loam	0.11-0.29 0.17	7.1-7.9 7.5

Table 1b. Chemical properties of soil under *Jatropha* cultivation at different sites

S. No.	Location of the sites	Range, and Mean values of soil chemical properties						
		Organic Carbon %	$CaCO_3$ (%)	CEC (meq/100g)	Total N (%)	Available N (kg ha^{-1})	Available P (kg ha^{-1})	Available K (kg ha^{-1})
1	Neswad	0.22-0.36 0.26	4.0-5.8 4.9	30.5-38.9 34.4	0.014-0.031 0.022	143.1-185.0 172.0	1.24-6.64 2.93	146.7-281.3 176.4
2	Bhadrawal	0.38-0.47 0.43	14.8-19.8 17.3	28.1-43.8 35.9	0.038-0.045 0.041	142.0-200.7 184.1	1.94-3.73 2.42	269.9-325.9 297.5
3	Kanod	0.55-0.81 0.66	2.8-3.8 3.2	37.7-49.3 43.5	0.05-0.069 0.059	103.5-251.1 149.8	2.9-14.5 8.39	185.9-201.6 194.6
4	Chorvadla	0.45-0.89 0.66	4.0-24.0 10.3	25.9-38.0 31.2	0.035-0.104 0.069	159.9-238.3 206.5	0.97-15.21 3.81	161.3-346.1 265.9

The analysis of major and micronutrients of soil was done for the Bhadrawal, Neswad and Chorvadla sites and is represented below. The micronutrient application would be done based on these values, and as can be seen from the **Table 1c,** the Zn content in all the sites is less than the critical level. More importantly, the criticality would now have to be established for *Jatropha* on the basis of its response to zinc fertilization which would be determined by experimentation.

Table 1c. Micronutrient status of soil

Sites	Cu mg/kg	Fe mg/kg	Mn mg/kg	Zn mg/kg
Chorvadla	2.9	9.7	19.6	0.3
Bhadrawal	2.3	8.0	11.4	0.3
Neswad	3.0	15.3	27.7	0.7

Table 2. Usually advocated soil tests and critical levels of nutrients in soils and plants

(Source: Methods, Soil testing in India, Department of Agriculture & Cooperation, Ministry of Agriculture, Government of India)

Element	**Soil Test Method**	**Critical level in soil**	**Critical level in plant**
Sulphur	Hot water, CaCl2 or phosphate	Usual 10 ppm range 8 - 30	< 0.15 – 0.2%
Calcium	Ammonium acetate	<25% of CEC or < 1.5 me Ca/100 g	<0.2%
Magnesium	Ammonium Acetate	<4% of CEC or <1 me Mg/100g	<0.1 – 0.2%
Zinc	DTPA	0.6 (0.4 – 1.2) ppm	<15 – 20 ppm
Manganese	DTPA	2 ppm	<20ppm (10-30)
Copper	DTPA or Ammonium acetate	0.2 pm	<4 ppm(3-10)
Iron	DTPA Ammonium acetate	2.5 – 4.5 ppm 2 ppm	<50 ppm(25– 80)
Boron	Hot water	0.5 ppm	<20 ppm
Molybdenum	Ammonium oxalate	0.2 ppm	<0.1 ppm

Determination of elemental composition of the initially uprooted biomass

The dried biomass samples of native vegetation that were uprooted before establishment of the *Jatropha* plantation were analysed using a CHNS elemental analyser. Their carbon and nitrogen contents are presented below in **Table 3** which would be later used for Life Cycle Assessment (LCA) studies.

Table 3. Carbon and nitrogen content of below and above ground removed plant biomass (moisture content 6-9%) at Bhadrawal

Native Plant *spp. present at* Bhadrawal	%C		%N	
	Above ground	Below ground	Above ground	Below ground
Prosopis juliflora	44.8	45.1	1.4	1.3
Acacia nilotica	45.8	46.1	1.0	0.6
Ziziphus mauritiana	43.6	45.2	1.2	0.8
Cassia fistula	45.1	45.8	1.7	0.7
Acacia leucophloea	42.2	42.7	0.9	1.0
Maytenus senegalensis	41.1	41.6	0.8	0.6

Similarly, the uprooted biomass samples at Neswad were analyzed for the macro and micro-nutrients present in them, and the results are depicted in **Table 4**.

Table 4. Macro and micro-nutrient composition of above- and below- ground parts of the initial native vegetation

	C%		N %		S%		Cu (ppm)		Fe content (ppm)		Mn content (ppm)		Mo content (ppm)		Zn content (ppm)	
Species name	Above-ground biomass	Below-ground biomass	Above-ground biomass	Below-ground biomass	Above-ground biomass	Below-ground biomass	Above-ground biomass	Below-ground biomass	Above-ground biomass	Below-ground biomass	Above-ground biomass	Below-ground biomass	Above-ground biomass	Below-ground biomass	Above-ground biomass	Below-ground biomass
Zizyphus mauritiana	43.50	45.10	1.24	0.79	0.00	0.00	6.7	5.1	158.0	1317.1	12.6	26.0	2.1	2.4	26.5	120.3
Prosopis juliflora	43.95	43.20	1.49	1.19	0.24	0.38	6.9	9.6	96.4	1409.9	11.5	19.6	1.9	2.8	45.1	21.1
Acacia leucophloea	42.01	42.65	0.81	1.00	0.00	0.00	7.0	49.4	260.5	4301.7	19.7	62.7	2.3	2.9	48.9	75.8
Balanites aegyptiaca	43.20	42.50	1.79	1.65			5.4	6.0	141.5	543.8	15.9	14.5	2.2	3.0	28.0	36.6
Acacia nilotica	45.30	44.95	1.12	1.18	0.12	0.12	IP	IP	IP	IP	IP	IP	IP	IP	IP	IP
Acacia senegal L	39.05	43.40	1.72	1.85	0.16	0.90	IP	IP	IP	IP	IP	IP	IP	IP	IP	IP
Cassia fistula	44.16	44.14	1.60	1.27	0.41	0.42	9.1	9.1	314.8	1256.1	19.0	16.4	2.9	2.3	30.1	25.9

Land preparation and arrangements for plantation

After cleaning the land of the existing sparse vegetation, land preparation was done by land shaping to remove undulations by minimal levelling by tractor **(Figure 12a)**. Arrangement for installing the irrigation system and erecting of bio-fencing was done. The layout **(Figure 12b)** for planting was carried out such that *Jatropha* plants would be spaced at a distance of 3m x 3m at both the places.

Figure 12. Land leveling **(a)** and layout **(b)** for *Jatropha* plantation.

Loosening/deepening of soil for planting (digging of trenches)

Since *Jatropha* is a perennial shrub, the soil should be loosened so that the roots find minimum resistance to penetrate down. The soil was loosened to a depth of 70-80 cm mechanically, thereby, breaking the hard pans beneath. The soil loosening was focused in the rows where *Jatropha* would be planted **(Figure 13)**. Drainage channels were dug at the Kanod site to prevent water stagnation and thus drain away the excess rain water as this site suffered from heavy texture.

Figure 13. Mechanical loosening and breaking of hard soil pans for better root penetration.

Effect of cyclone: Exceptionally strong cyclonic winds in the last quarter of 2010 caused damage to many plants. The storage sheds on the older plantation sites of Chorvadla **(Figure 14)** planted in 2005 were blown away.

Figure 14. (left) *Jatropha* plantation at Chorvadla (Site 3) before cyclonic storm **(right)** and blown away *Jatropha* plants wrecked Farm shed due to cyclonic storm.

Thus, it was learned that for even seemingly well-established plantations, the occurrence of a cyclonic storm once in many years can also be a veiled threat and completely wipe out large portions of mature plantations **(Figure 15)**.

Figure 15. Plantations affected by winds.

Ways to make shallow wastelands suitable for cultivation of *Jatropha*

The reason why the plants could not withstand the strong winds was investigated, and it was found that due to the very rocky soil terrain in many portions of the plantation, the roots were not able to penetrate deeper into the soil and thus could not give good anchorage to the soil and lodged due to heavy winds and rains. Replanting at these sites was done with special focus on remedial measures based on this learning. Experience from the earlier project at the premises of General Motors, India at Talegaon (Maharashtra) was utilized to find a solution wherein a *Jatropha* plantation was established in very rocky terrain by digging deep into the stones or overlaying layers of soil from outside. In parallel, CSIR also sponsored a project (EMPOWER) wherein the objective was to devise a strategy of making a deep root zone in rocky terrain without having to bring soil from outside. Accordingly, the rocky zone was dug up very deep (70-80cm) by excavator so as to provide sufficient anchorage to the roots which can withstand strong winds in such soil conditions and windy terrain **(Figure 16, 17)** and soil from an adjacent area was used. The setback to the existing plantation can be seen as a valuable learning with respect to such a category of rocky wastelands prone to strong winds where considerable deepening of land must be ensured at the outset to avoid such an incidence at the later stage. This should constitute the prudent and best practice.

Figure 16. Deep trenching in rocky wasteland (left) and entire deep trenched field (right).

Figure 17. *Jatropha* cuttings in the ameliorated zone.

Comparative study on specific consumption of *Jatropha* biodiesel and fossil diesel to run caterpillar (JCB machine)

As a part of generating data for determining the diesel requirement of the excavator machine for performing different types of operations, a practical study under the rocky terrain was undertaken to find out the efficiency of *Jatropha* biodiesel and fossil diesel to run the caterpillar (JCB machine).

The work details are as follows:

JCB caterpillar specification; 3 DX, 76 HP; Year of manufacture; 2009

Site: Neswad experimental *Jatropha* site

Type of work done by JCB: Shrub uprooting, Leveling, Trench digging up to ~75 cm deep

Type of soil: soil - up to 20cm depth; below 20 cm semi-hard & rocky (but easily broken)

JCB start time (after pouring biodiesel)	JCB – Closing time	Total Hrs. run	Net Biodiesel added in JCB tank before start	Balance of biodiesel in tank at closing time	Total *Jatropha* Biodiesel consumed to dig trenches	Consumption (in litres per hour)
10.45am	**6.00pm**	**7hrs 15 min (7.25 hrs)**	**45 ltrs.**	**8.5 ltr**	**36.5 ltr**	**5.034 ltr**

JCB start time (after pouring fossil-diesel)	JCB – Closing time	Total Hrs. run	Net fossil-diesel added in JCB tank before start	Balance of fossil-diesel in tank at closing time	Total fossil-diesel consumed to dig trenches	Consumption (in litres per hour)
11.30 am	4.45 pm	5hrs 15 min (5.25 hrs)	50 ltrs.	25.5 ltr	24.5 ltr	4.67 ltr

The opinion of the driver was taken and has also been recorded as follows:

Opinion of driver using biodiesel:

- No sort of trouble was encountered while driving/running the JCB caterpillar
- No voice change in engine was noted by the driver
- Exhaust (smoke) was much less in comparison to fossil diesel. The driver checked this many times by pressing lever.
- Constant running for 7.15 hours did not over heat the JCB machine. Further, the machine was run at variable speeds from low to high speed, and no jerks were encountered.

Thus, it was found that the performance of biodiesel in doing the work is absolutely satisfactory. The specific fuel consumption of biodiesel per hour was 7.8% higher than fossil diesel, but this is obvious on account of lower calorific value of biodiesel in comparison with HSD. However, a large reduction in smoke is observed with *Jatropha* biodiesel as was also corroborated by positive feel of the driver.

Creation of high yielding germplasm

Selection of high yielding germplasm has been the main criteria of CSMCRI since the very outset of *Jatropha* research, and it has now several proven accessions which, if multiplied in the millions, could lead to productive plantations throughout India. To date, around the world, most of the plantations have been raised through seeds of unknown pedigree leading to uncertainty in the per unit area production. Our experience shows that plants raised through seeds of even elite parents show wide variability in plant-to-plant seed yield. Hence, there is a need for quality planting material for which the demand is increasing for greater consistency of productivity. This also formed the basis of the project wherein it was decided to set up the new 50 ha plantation using elite germplasm with a proven track record.

Accordingly, about 70,000 true-to-type cuttings/tissue culture plants of elite genotypes were raised in nursery for transplanting in the field and bio-fencing. The cost of cuttings was further brought down by putting up the nursery in raised sand beds **(Figure 18a right)** at Gopalpur, Odisha. This method avoided the requirement of the use of polythene bags **(Figure 18a left)** which leaves a higher carbon foot print.

Figure 18a. Cuttings raised at Bhavnagar (left) and at Berhampur, Orissa (right).

The planting material was to be sent to the respective places of planting **(Figure 18b)**. Later on the cuttings of elite *Jatropha* plants were transplanted **(Figure 18c)** in the field at the new sites after necessary field preparation and nutrient input application **(Figure 18b)**.

Figure 18b. Preparation for transplantation being carried out at Bhadrawal and Kanod.

Figure 18c. Established *Jatropha* plantation at Gujarat (Planted in July-Aug, 2010).

Tissue culture

Vegetative propagation through stem cutting has its own limitations as it will ultimately lead to a depletion of the existing selected genotypes. Hence, tissue culture is one of the solutions for mass propagation of *Jatropha*. The *in vitro* culture generation is the best procedure for large scale propagation of true-to-type elite plants. Micro-propagation can be undertaken from any plant part, but apical and nodal meristems are the best source of tissue for multiplication, as the plants generated this way show lower variability. The optimization of the method was based on trials of explants grown under different conditions, media composition, practical observations of bud formation, optimum number of buds to be left after each subculture, and shoot maturity at the time of harvesting, etc.

Jatropha curcas being perennial in nature has its own limitations even in tissue culture like low culture establishment, slow growth, and a high rate of infection and low rooting percentage. All these problems were simultaneously addressed to improve the protocol while generating tissue culture plants for establishment in the field. A protocol for micro-propagation using nodal explants and shoot apex has been developed to generate true-to-type elite clones of *Jatropha curcas.*

Collection of plant material for culture generation

High yielding plant CP-9 (IC-565735) with good oil content was used for culture generation. In addition, other better performing plants were also used for culture generation. To observe seasonal response, plant material was collected throughout the year at the interval of 30 days and the percent survival rate of the culture was monitored. As cuttings are seasonal, cuttings generated in January could be used from March onwards.

Young shoots of approximately 12 to 15 inch were selected from the mother plant growing in the field **(Figure 19 a)** and in the green house/nursery **(Figure 19 b, c)**. The use of very hard shoots was avoided for culture generation. Leaves along with petiole were removed leaving a small portion of petiole intact. Note that care should be taken to prevent injuries to the stem segment during cutting of materials. Shoots were segmented with the help of a scalpel keeping a single bud in each node (one node/explant). To remove microbes from the surface of explants, they were surface sterilized with Mercuric chloride (0.1% w/v) for 10-15 minutes. The time duration was increased or decreased depending upon juvenility of the explants. Explants were washed 3-5 times with sterile distilled water for one minute per each wash. Explants were air dried under sterile condition and then small portions of the upper and lower end along with petiole attached to the explants, were removed with the help of a sterile surgical blade before inoculation into the medium.

Figure 19. Young shoots were selected from mother plants growing in the field **(Figure 19 a)** and in the green house/nursery **(Figure 19 b, c)**.

Table 5. Plant material collected from field of CSIR-CSMCRI

Month	Shoot bud response (%)	Culture Survival (%)
January	70	20
February	71	20
March	85	90
April	90	90
June	90	90
July	85	65
August	85	65
September	70	65
October	70	50
November	65	40
December	65	20

Table 6. Plant material collected from cutting in green house/nursery of CSIR-CSMCRI

Month	Shoot bud response (%)	Culture Survival (%)
March	90	90
April	90	90
June	90	90
July	85	90
August	85	90
September	85	90
October	70	90
November	65	90
December	55	90

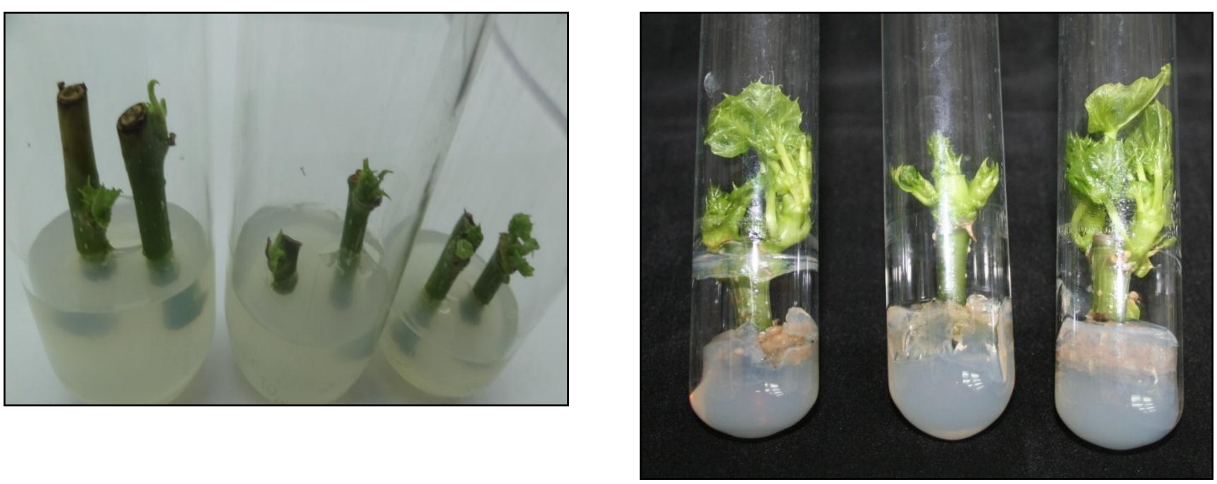


Figure 20. Nodal segment cultured on MS medium BAP and IAA (a) 10 days old and (b) 30 days old.

Shoot bud induction from nodal/shoot tip explants

Explants were cultured vertically in 25 ml/tube (35mm X 200mm) Murashige and Skoog (MS) basal medium (pH5.8) supplemented with BAP and IAA, 3% (w/v) sucrose, solidified with 0.7% (w/v) agar. All cultures were incubated at 25±2 °C under a 16 h light/8 h dark photoperiod by cool-white fluorescent lamps at the intensity of 35 µmol/m^2/s. After 7 days, shoot initiation was observed in both the explants (field and green house/nursery). The best shoot bud inductions were observed in the months of March to September in nursery and March to August in the field but the culture survival was more (90%) in cultures generated through cuttings throughout the year **(Table 5 and 6)**. Culture generation was possible throughout year from cuttings in the greenhouse/nursery.

Shoot proliferation and elongation

After 4-5 weeks, explants were cultured onto subculture medium supplemented with BAP, IAA. During subculture, old mass and dried leaf/tissue of the mother culture was removed and newly generated shoot buds cultured onto the medium. After subsequent subcultures, multiple shoot buds arises from the explant and they need separation as overcrowded buds may hinder their growth. Elongated shoots were used for rooting.

Figure 21. Multiple shoot buds after subsequent sub culture of elite genotype of *J. curcas.*

Rooting of *in vitro* generated shoots

In vitro rooting of the shoots

Elongated shoots were kept for rooting in MS medium supplemented IBA, NAA, and charcoal (0.2%) and the remaining part of the explant was cultured in shoot proliferation and elongation medium further for culture generation. After 15-30 days, root initiation was observed. IBA promoted root regeneration and facilitated root elongation because many lateral roots formed from plantlets cultured on medium with IBA but not in medium without IBA. Later on, the regenerated shoots with well-developed roots were transferred into a polythene bag containing autoclaved gardening soil wetted with sterile water, (0.01%) Bavistin and covered with a transparent poly bag. The polythene bag was gradually opened for acclimatization. After 2 weeks of acclimatization, plantlets were transferred in a glasshouse. However, the regenerated plantlets developed were phenotypically normal and resembled mother plants. Unrooted shoots obtained during in vitro rooting were again used for *ex vitro* rooting.

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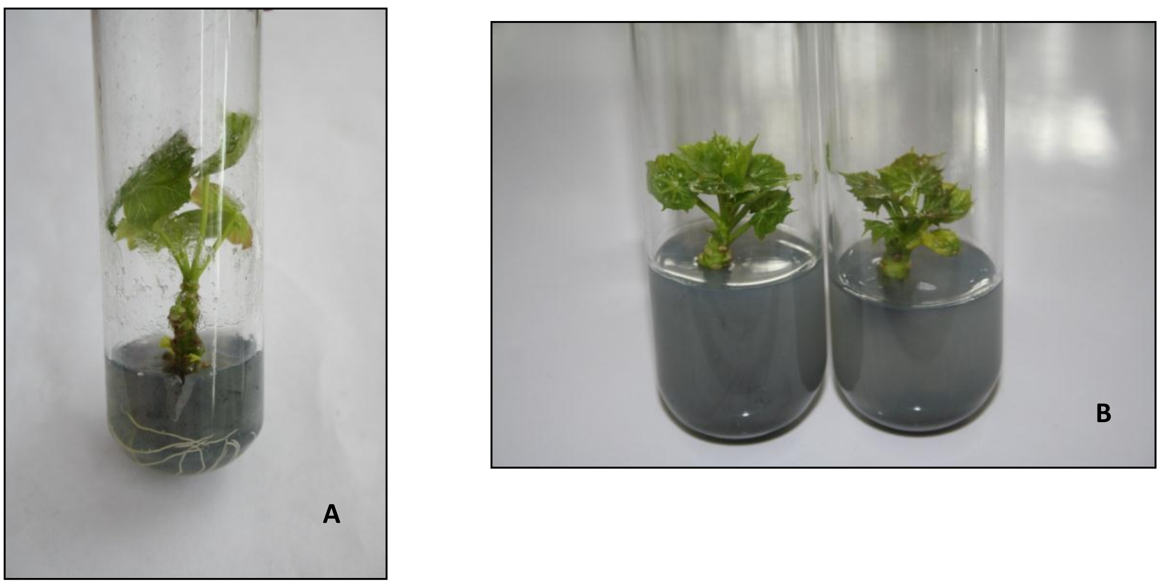


Figure 22. *In vitro* generated shoot (A) *in vitro* rooting in MS medium with Charcoal (B) unrooted shoots obtained during *in vitro* rooting.

Figure 23. *Ex vitro* rooting **(a)** and hardening of the shoots under controlled condition **(b)** *ex vitro* rooted shoots.

Ex vitro rooting of in vitro generated shoot

Elongated and mature shoots harvested from shoot proliferation and elongation medium after giving appropriate treatment were, as was done before, transferred into a polythene bag containing autoclaved garden soil wetted with sterile water with (0.01%) Bavistin and covered with a transparent poly bag. The polythene bag was gradually opened for acclimatization **(Figure 23 a, b)**. After 2 weeks of acclimatization, the plantlets were transplanted to pots and transferred in a glasshouse. However, the regenerated plantlets developed were phenotypically normal and as like mother plants.

US biofuel mission visit to CSMCRI and planting of tissue culture raised plants

On October 25th, 2010, US Biofuel Mission comprising of Scientists/Experts from US DOE and various other US National Laboratories like Argonne National laboratory, Oak Ridge National laboratory, NREL, and Yale University, visited CSIR-CSMCRI, Bhavnagar. The Mission was headed by Dr. Glenn Sonntag, Lead, US-India Biofuels Team, Office of the Biomass Program, United States Department of Energy. Dr. Candace Wheeler, Principal Investigator (General Motors) also accompanied the team. The members of the mission visited the plantations under this project at the sites located at Bhadrawal and Neswad **(Figure 24)**. Incidentally, the members of the delegation transplanted micro-propagated elite *Jatropha* saplings at the newly established plantation site of Bhadrawal **(Figure 25)**.

Figure 24. Visit of US Biofuel mission to *Jatropha* field at Bhadrawal (right).

Figure 25. Transplanting elite *Jatropha* micro-propagated saplings in the field (Right, Dr. Candace Wheeler and left, Dr. Glenn Sonntag).

The same plants which were transplanted by the US Biofuel mission team in 2010 grew up and started yielding fruits in 2012-13 **(Figure 26).**

Figure 26. Tissue culture plants growing in experimental plot Bhadrawal, CSIR-CSMCRI.

Selection of quality germplasm and improvement

A lack of quality germplasm results in lower seed yield and oil. Since the plant is a perennial, consequences of inadequate attention to such details may lead to frustration over poor yields in the subsequent years. Most of the seeds sold currently are collected from forest regions, which have no known pedigree and are used for general plantations. As mentioned earlier, even for plants of known pedigree, plants raised from seeds showed large variability and that is why we focused on raising true-to-type cuttings or tissue cultured plants in large numbers. Thus, it is critical to develop and register varieties that give consistent and high yield.

There is, however, considerable uncertainty so far as the yield of *Jatropha* is concerned. The productivity of *Jatropha* is dependent on a number of complex genetic, edaphic, and climatic factors. We raised a plantation entirely from cuttings of elite plants and its performance was tracked over several years. Performance of progeny plants raised from cuttings of selected mother plants exhibited reasonably high and consistent performance during many years. A total of 10 suitable accessions have now been identified for large scale propagation. Sufficient long term data do not exist on the comparative merits of plantations raised from cuttings versus those raised from seedlings. Until consistency in performance of seedling based plants is established, we are focusing our efforts to ensure that farmers will have access to large volumes of cuttings, for which our success rate is currently >90%.

We have identified a few accessions that have been found to be superior year after year at our various plantations. The plantation at Neswad was established in 2007-08 on shallow-soiled rocky wastelands. Photographs **(Figure 27)** below shows how the plants raised from elite cuttings have drooped due to the weight of the fruits in the fruiting season of 2011. The actual fruits can be seen where the branches were propped up **(Figure 28)**.

Figure 27. Fruiting *Jatropha* plants at Neswad.

Figure 28 Heavy fruiting in plants at Neswad (Picture Date 20th August, 2011).

Figure 29. Heavy fruiting (harvest) at Chorvadla plantation.

An independent assessment of the Neswad plantations was made by an expert nominated by MNRE (Ministry of New and Renewable Energy, Govt. of India, New Delhi) in Sep 2011, who witnessed the high yield obtained on the plants during the fruiting season at Neswad **(Figure 30)**.

Figure 30. Superior yield performances of cuttings of *Jatropha* selections being witnessed by one of the independent experts (Dr. H.M Behl) at the behest of Ministry of New and Renewable Energy, Govt. of India. Picture dated 19th Sept, 2011, CSMCRI *Jatropha* experimental site Neswad, Bhavnagar, Gujarat.

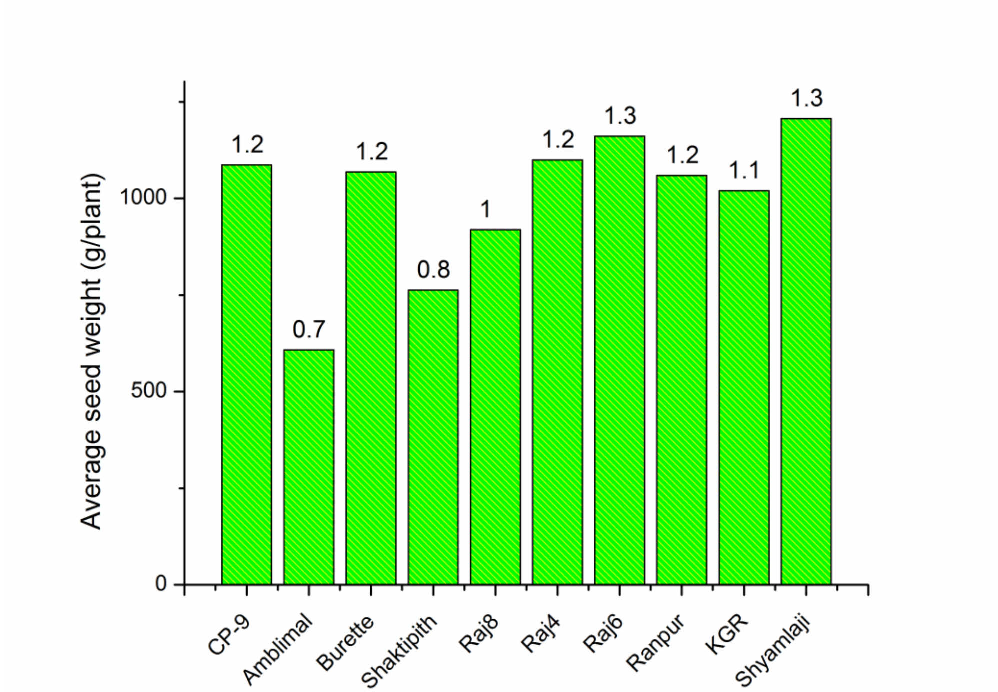


Figure 31. Mean seed yield of selected elite accessions of *Jatropha* at Neswad (2011-12). **(Figures appearing on label are the equivalent seed yield in Metric tonnes/ha)**

In 2011-12, among the different accessions selected for different desirable traits, the accessions Shyamlaji (IC 565733) and Raj-6 recorded the maximum seed yield of ca. 1.3 t/ha. This was closely followed by that obtained ca 1.2t/ha from accessions CP-9 (Accession No. IC-565735, Burette, Raj-4, and Ranpur (IC-565730). **Figure 31** depicts the yield obtained in 2011-12 and also expressed in tonnes per ha.

The performances of some accessions had been consistently good over a period of years, at Gujarat and other places (Orissa state) and formed the basis of the selections in the ongoing research project for large scale testing. **Figure 32** depicts the year-on-year seed yield at the Neswad field (planted in 2007-08 at 3m x 3m spacing) of 5 such shortlisted accessions found to yield consistently higher. As can be seen, accessions IC-565733 (Shyamlaji), IC-565735 (CP-9), and Raj-6 performed well in all the years. It may be noted that the seed yields may not show increasing trend with each passing year till the plantation attains stability in yield. There may be setbacks due to unfavorable environmental conditions any time during the life period. Such conditions prevailed in 2010-11, when relatively higher rainfall intensity coupled with an unseasonal downpour beyond the normal monsoon season (in November 2010), adversely affected flowering and fruit set, probably due to the washing away of the pollen. Therefore, little yield was realized as compared to earlier years. Similarly, disease outbreak (fungal or viral) or pest attack during a season can also affect the productivity.

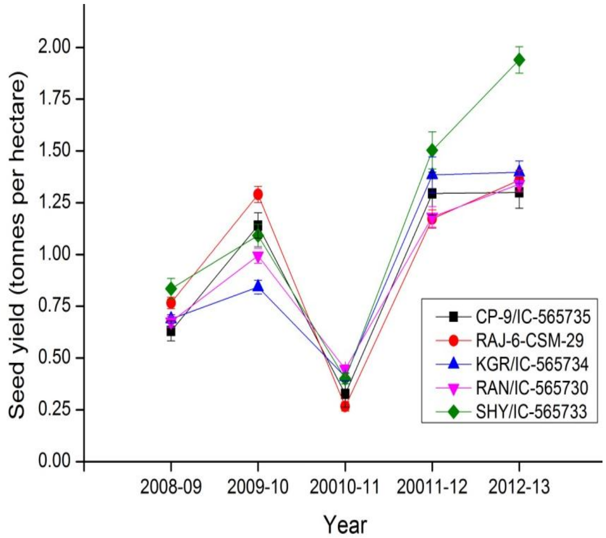


Figure 32. Year on year seed yield of elite accessions of *Jatropha* at Neswad, Gujarat.

Rainfall data is given in **Figure 33**. However, as reported earlier, the yields recovered in the subsequent year (2011-12).

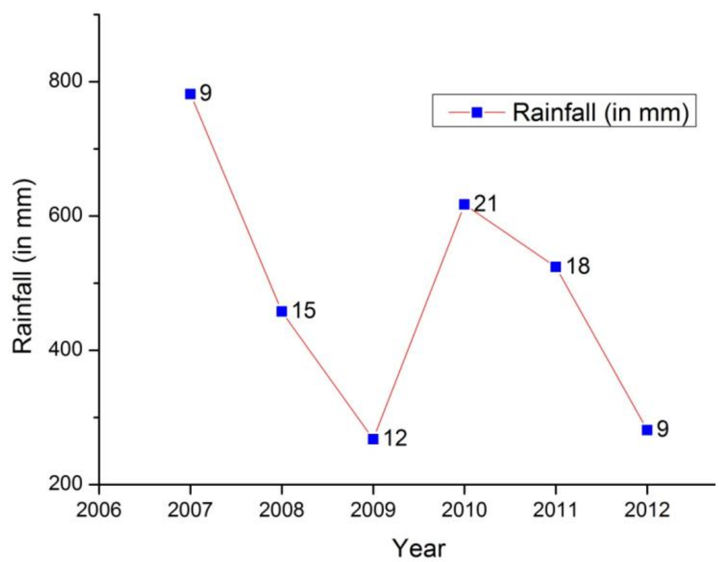


Figure 33. Annual rainfall (mm) in Bhavnagar for the last 6 years. Labels show number of rainy days (own observations in field).

The months of July to September are characterized by monsoon season during which Gujarat receives most of its rainfall. It is interesting to note that the fruiting season of 2012-13 was characterized by scanty rainfall in Bhavnagar district, which was lower than many previous years and nearly 280 mm of rainfall was received in a total of 9 rainy days during this wet season (**Figure 33**). Although, there was much less rainfall during this season, this did not drastically effect the plant growth, signifying that *Jatropha* can tolerate scanty rainfall conditions. Such a drought is a characteristic of the semi-arid areas of the country, and hence, *Jatropha* is a fit crop in such situations. The number of flushes of flowering was, however, reduced to one or two. This may have been an advantage since the harvesting was synchronised for the entire plantation and was concentrated within a short period. The same trend can be seen in 2009 when rainfall was less but did not drastically affect the seed yields. The intensity of rainfall, rather than amount of rainfall is more important. However, it is cautioned that *Jatropha* is prone to *Macrophomina phaseolina* fungus causing collar rot, and it affects in dry conditions when summer irrigation cannot be provided. Collar rot can affect even well grown plants.

Compared to 2011-12, the seed yield in 2012-13 increased by 13.6% for the accession IC-565730 (Ranpur), while in the accession IC-565733 (Shyamlaji) the seed yield increased by 29.3%, recording the highest seed yield of 1.94 tonnes per ha (~2 tonnes/ha) (**Figure 32 and 34**). Other accessions which performed consistently in the last few years, i.e., IC-565739 or CP-9, IC-565734 or KGR and Raj-6 also yielded relatively high in the range of 1.3 to 1.4 t/ha.

Figure 34. *Jatropha* plants heavily laden with green fruits at Neswad, Bhavnagar, Gujarat (Picture: October 2012).

In addition to these elite accessions, one genotype namely CP-9 (IC- 565735 was assessed for its performance for several years at our Berhampur site (Odisha state). Evaluation of the performance of this accession is presented in the table below. However, the seed yield dipped in 2011-12 due to climatic reasons. The analysis of the mean oil content of the seeds of this accession was also undertaken in the six harvest years and depicted in **Table 7** below. **Figure 35** depicts the seed oil content.

Table 7. Year-on-Year performance (seed yield) of plants raised from cuttings of IC-565735 (CP-9) identified as the best performing provenance at Mahuda, Berhampur, Orissa

Mean seed yield in kg/plant (N=10) at Berhampur							
Year	2005-06	2006-07	2007-08	2008-09	2009-10	2010-11	2011-12
Seed yield	0.30	0.70±0.2	1.40±0.8	2.00±0.8	2.04±0.6	2.2±0.5	1.3±0.4

NB: Date of planting- Jan 2003; Planting material- cuttings of IC-565735; Irrigation- once a month with 30 L/plant during non-rainy months; Fertilizer: NPK along with *Jatropha* cake

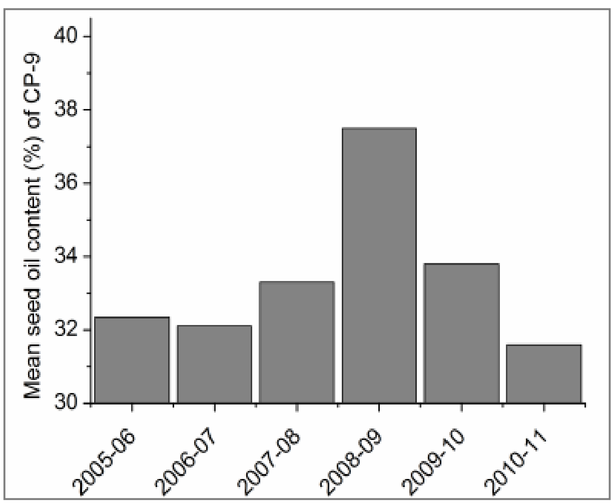


Figure 35. Mean oil content in accession IC-565735 for six years

Another very promising accession IC-565739 from our Zanjmer site also performed well and proved high yielding under sand dunes of Gujarat and yielded between 3.5 to 5.5 kg/plant from year 2007-08 to 2009-10. However, during 2010-11, the yield declined to 3.74 kg/plant (yields were uniformly low across all places in Gujarat). In 2011-2012 the plants were affected by termite attack leading to poor yield **(Table 8)**. However, control measures were taken thereafter, but the plant recovery is expected to take some time. Micro-propagation offers opportunity to conserve the germplasm and propagate true-to-type plants from such individual elite plants that may be vulnerable to pest attack.

Table 8. Mean seed yield of accession IC-565739 in different years at Zanjmer site

Year	**2007-08**	**2008-09**	**2009-10**	**2010-11**	**2011-12**
Seed yield (kg/plant)	4.14	5.20	5.55	3.74	2.01

Figure 36. Elite germplasm (IC-565739) on sand dunes of Zanjmer **(a)**; fruiting **(b).**

The fatty acid composition of the seed samples shows considerably higher unsaturated fatty acids (oleic and linoleic) than saturated fatty acids (palmitic and stearic acids). With much higher proportion of unsaturated fatty acids, *Jatropha* biodiesel has a lower melting point and a high flash point. The fatty acid compositions of some of the elite accessions found to be good are depicted in **Table 9** below:

Table 9. Fatty acid composition of *Jatropha* seed oil

			Fatty acid composition %					
Sr. No.	Elite accessions	Oil yield (%) (from kernels)	Palmitic acid	Stearic acid	Palmitoleic acid	Elaidic acid	Oleic acid	Linoleic acid
1	CSMCRI-GUJ-Banas-1205-C-1	48.5	14.26	7.69	0.32	0.63	45.4	32.33
2	CSMCRI-GUJ-Panch-0106-C-3	44.45	17.54	5.39	0.69	0.93	42.52	33.69
3	CRIDA-AP-Adila-0904-JL-06	43.6	18.07	8.43	0.65	0.62	45.45	26.15

4	CRIDA-MP-Jhabua-02-03-JJ-06	47.88	18.26	8.45	0.63	0.76	45.26	26.36
6	NBPGR-GUJ-SKN-0605-Hansraj	45.66	18.57	8.37	0.72	1.01	44.27	27.93
7	NBPGR-GUJ-SKN-0605-Urlikanchan	45.19	17.34	9.53	0.72	0.52	45.64	26.92
8	NBPGR-GUJ-SKN-0605-Chhatrapati	47.44	20.11	9.39	0.72	0.52	44.14	24.7

Hybridization

In order to develop *Jatropha* hybrids from single crosses, hybrid lines of *Jatropha curcas* were derived from identified *Jatropha* accessions and their performance was observed for two successive generations. Promising hybrid lines were multiplied through stem cuttings and transplanted in an isolated plot at Bhadrawal during monsoon. Early seedling growth was assessed on approx. 1500 stem cuttings raised from various promising hybrid lines in the nursery. Out of eight cross combinations, cuttings raised from hybrids lines showed better growth and more branches as compared to the other six combinations. Around 74% F_1 plants showed an increase in fruit set over their respective parents. Up to 21 fruits per inflorescence and 180 g seed yield per plant were recorded in this experiment. During the first year, plants could yield between 40 g to 90 g of seeds; however, in a few F_1s, it ranged between 110 to 180 g whereas, for parents it was between 90 to 105 g. During the second year, the seed yield increased considerably with an average of 175 g to the maximum 300 g. These hybrids are being monitored for selecting desirable hybrids based on higher productivity and oil content.

Confirmation of hybridity:

Hybrids were generated among the selected parents, and F_1 and F_2 were raised from the seeds. These were planted in the CSIR-CSMCRI field, Bhadrawal. They were planted as sets such that each set represent particular crossed parents and their respective offspring. The first set of cross was done between **IC 565735 X IC 565739,** the second set includes **IC 565730 X IC 565731**, the third set includes **IC 565736 X IC 565737** and the fourth set of crosses was done between **IC 565733 X IC 565734.** These were subjected to hybrid confirmation using RAPD marker systems. Among the plants in the first set, 18 out of 26 putative hybrids were confirmed to be hybrids through RAPD finger printing. This was given by the primers opL1, opL4, opL5, opL7, opP2 and opP6. In the second set 22 out of 40 putative plants generated were found to be hybrids. Among the primers studied opL4, opL5 and opL7 has given the results for hybrid confirmation. In the third set, 10 out of 40 hybrids were found to be hybrids of their respective parents. Among the primers studied, opL7, opL14 and opP2 have given the results of hybrid confirmation of 10 hybrids. In the fourth set, 22 among the 40 hybrids were found to be hybrids of their respective parents. Among the primers studied, opL4, opL5, opL14, opL7, opN4, opL18 has given the results of hybrid confirmation.

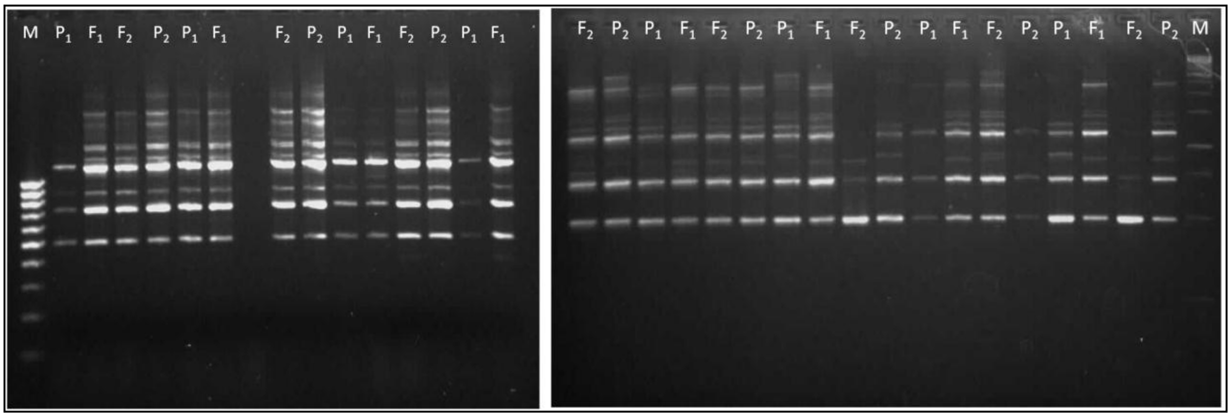


Figure 37. Agarose gel photos showing the hybrid confirmation of offspring and their respective parents using opL- 7 RAPD primer. M- DNA ladder; P_1 and P_2 are parents; F_1 and F_2 are their respective off springs.

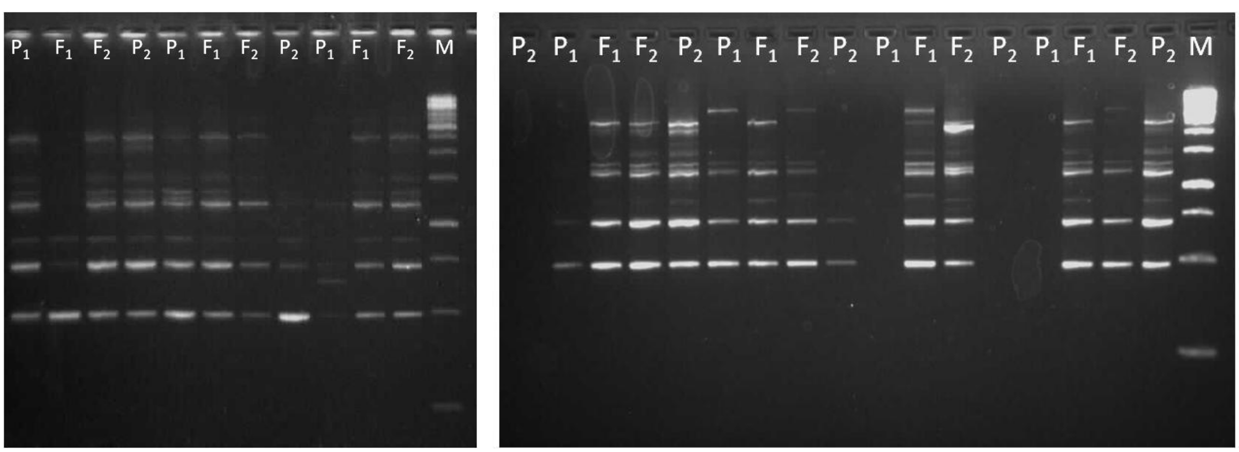


Figure 38. Agarose gel photos showing the hybrid confirmation of offspring and their respective parents using opL- 7 RAPD primer. M- DNA ladder; P_1 and P_2 are parents; F_1 and F_2 are their respective off springs.

Development of good agro-practices on wastelands and learnings

Land amendment

Wastelands are degraded lands that do not fulfill their life sustaining potential. Wastelands can result from a number of inherent or imposed disabilities. The wastelands we have worked on during this project were non-saline lands which were either very shallow and had rocky strata beneath or were prone to situational constraints like being water-logged due to heavy soil texture, impeding any agricultural crops to be taken up in these lands. The primary limitation at Kanod site was that parts of it remained water-logged **(Figure 39)** for a considerable length of time during the rainy season. This was due to the low permeability of the heavy-textured soil coupled with a site situation which did not allow drainage of water. Subsequently, in the dry season, the soil cakes up and becomes very hard. We stress that being a perennial plant, *Jatropha* is expected to have a long productive life. It is, therefore, prudent to invest generously in the initial phase so that the plantations are productive and remain so. Rushing ahead with *Jatropha* cultivation on a large scale, without proven germplasm and agronomic practices and without an understanding of plant performance under different edaphic conditions, will inevitably lead to future disappointment.

Figure 39. Stagnation of water at Kanod site for considerable lengths of time during monsoon season (June to September).

Figure 40. Deep drainage channels diverting excess run-off out of the field.

Efforts were made to address the problem by incorporating coarse sand in the root zone of *Jatropha* to allow proper penetration and aeration of the roots at least during the juvenile stage of the plant. A suitable deep drainage provision **(Figure 40)** was given to divert substantial run-off from the root zone of *Jatropha*. Subsequently, measures were also taken to manipulate the planting and root zone to ward off the situation of water stagnation and planting was done on ridges. The plants established after 3 years are shown in **Figure 41a-c** below and have also started flowering and fruiting.

Figure 41a&b. Established plantation at Kanod**; c** fruiting in plants at Kanod.

Addressing scarcity of water

As can be seen from the rainfall data given in the previous section, occasionally, the region growing *Jatropha* might experience periods of very low rainfall. Although, the mature plantations are capable of handling such stresses, it becomes very necessary to support the young plants with life-saving irrigation. A bund for check dam was repaired at Chorvadla site which enabled recharging of the ground water reservoir in the rainy season, thus allowing for life saving irrigation in the ensuing harsh summer **(Figure 42)**.

Figure 42. Excess run off was made to recharge the ground water by a check dam which helped the water in the well to last longer.

Efforts were also made to make maximum use of available water for above and below ground plant growth in shallow and rocky types of wastelands where water availability is scanty and different water conservation methods were adopted including the use of mulch (**Figure 43**).

Development of innovative agronomic techniques for higher water use efficiency

The use of drip irrigation allows efficient use of irrigation scanty water resource, but the disadvantage of drip irrigation is that the roots tend to remain shallow, while the limitation of surface irrigation includes application and evaporation losses. Therefore, an innovative agro-technique (drip-in-sand technique) for higher water use efficiency was developed on the stretch of land where soil was shallow and stony (**Figure 44**). In this technique, water was let in through a dripper into a vertical column, dug from the surface to the bedrock, filled with coarse sand to enable water to percolate into deeper layers of the soil at a faster pace before lateral and upwards redistribution occurs. This prompted the roots to penetrate deeper to explore water and nutrients.

In all cases, the improvised techniques fared better. There was marked improvement in overall height, canopy, girth, secondary branching, biomass weight, and total root length (**Figure 45**) by the drip-in-sand technique as compared to the surface and normal irrigation techniques **(Table 10)**.

Figure 43. Mulching in Jatropha.

Figure 44. Low cost drip-in-sand technique.

Figure 45 Measurement of increased root growth due to efficient water management techniques.

Notable changes in biochemical properties were observed (**Table 11**) and the enzymes related to nutrient cycling in soil were favorably increased with this technique, enabling enhanced nutrient availability to plants, hence corroborating the observed improvement in plant growth.

Use of Liquid Seaweed Fertilizer from *Kappaphycus alvarezii*

The use of sap from *Kappaphycus alvarezii* (a marine seaweed or macroalgae) – a commercialized and patented CSIR-CSMCRI product, which has shown significant improvement in yield of several agricultural crops, was used in the *Jatropha* plantation. Seed yield improvement, ranging from 22-40%, was observed by using *Kappaphycus* sap foliar spray @ 10-15% concentration, 3 times during the growing season.

Table 10. Growth characteristics of *Jatropha* under different irrigation techniques

Treatment	**Height (cm)**	**Canopy (cm)**	**Girth (cm)**	**No. of branches per plant: Primary**	**No. of branches per plant: Secondary**	**No. of branches per plant: Tertiary**	**Fresh biomass (kg/plant)**	**Root parameters: Total length (cm)**
T1 Drip-in-sand technique	109.4	210.3	4.5	2.6	9.2	0.6	3.2	285.7
T 2 Normal drip irrigation	73.2	79.6	3.0	2.1	4.8	0.1	0.6	127.0
T3 Surface irrigation	58.2	67.1	2.4	2.1	2.8	0.0	0.2	79.7

Table 11. Biochemical properties of soil under different irrigation techniques

Treatments	**Soil enzyme activity (mean ± sd of 3 replicates)**			
	Aryl sulphatase activity (measured as µg of nitrophenol released/h/g of soil)	Acid Phosphatase activity (measured as µg of nitrophenol released/h/g of soil)	Alkaline phosphatase activity (measured as µg of nitrophenol released/h/g of soil)	Glucosidase activity (measured as µg of nitrophenol released/h/g of soil)
T1 Drip-in-sand technique	62.75 ± 10.52	206.08 ± 20.20	494.42 ± 74.78	276.24 ± 38.52
T2 Normal drip irrigation	48.68 ± 18.25	164.86 ± 10.78	342.32 ± 7.33	137.40 ± 4.95
T3 Surface irrigation	40.20 ± 18.32	115.38 ± 3.84	268.24 ± 3.89	138.52 ± 11.30
CD at 5%	NS	35.0	95.7	48.6
CD at 1%	NS	58.1	158.6	80.6
C.V. %	18.89	9.53	11.46	11.65

Figure 46. Making of reservoir for gravity irrigation system:

1. Potential hillock selection and excavation; 2. Excavated hillock; 3&4. Strengthening the walls of reservoir by stones obtained from hillock; 5&6. Making of reservoir for gravity irrigation downstream; 7. Irrigation network through gravity irrigation; 8. Reservoir filled with water and gravity irrigation being done.

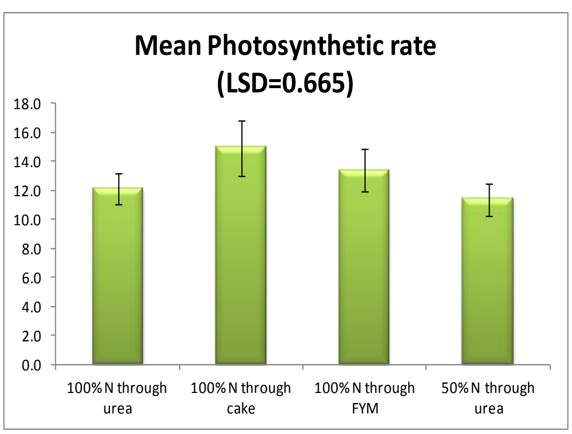
Use of natural heights in hillocks for making reservoirs for gravity irrigation

Wastelands existing in Bhavnagar possess numerous hillocks which were advantageously used to create a reservoir through which it was possible to carry out gravity irrigation. Even the construction of it was made with the stones obtained by the excavation of hillock, thus making it cost effective. It can be used for life saving irrigation by connecting it to irrigation pipeline **(Figure 46)**.

Utilization of *Jatropha* cake in *Jatropha* plantation

An experiment to evaluate the effect of different inorganic and organic sources of nutrients was also continued through the project period from 2008 onwards. Observations on photosynthetic parameters **(Figure 47)** were taken, and it was observed that the use of cake could significantly improve the net photosynthetic rate in plants and also improve the water use efficiency **(Figure 48 a&b)**.

Figure 47. Gas exchange measurements like photosynthesis, transpiration, stomatal conductance being taken with the help of Infra-red gas analyser system (LI-COR, USA).



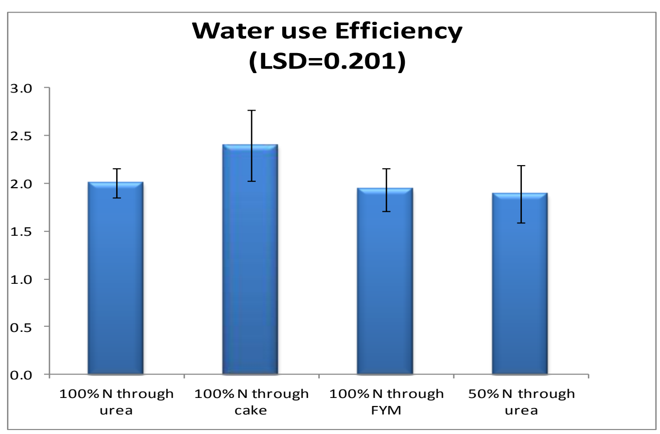
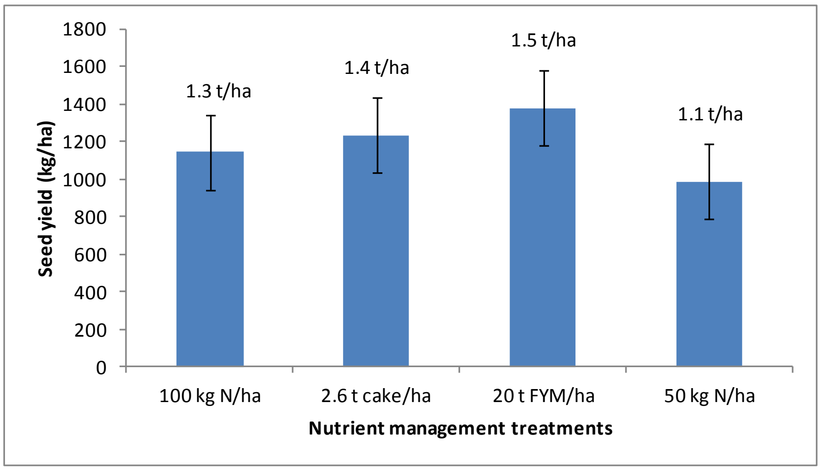


Fig 48 a. Net photosynthetic rate; (P_N) (µmol CO_2 $m^{-2}s^{-1}$) ; **b.** Water use efficiency (µmol CO_2 $m^{-2}s^{-1}$ CO_2/mol $m^{-2}s^{-1}$ water) due to different nutrient management.

The results reveal that the *Jatropha* cake was found to be statistically at par with the traditional Farm Yard Manure (FYM) and was a better fertilizer over inorganic sources of nutrients. In 2011-12, the highest seed yield was found to be on the order of 1.5 t/ha with 20 t FYM per ha being applied as compared to that (1.4 t/ha) obtained by the application of 2.6 t *Jatropha* cake per hectare in the fourth year of the plantation **(Figure 49)**. The experiment used a high rate of fertilizers to determine the highest potential yield that may be obtained and to promote an early advantage with respect to growth (above- and below- ground) and hence does not reflect the general dose which is applied to the plantations which is lower than that applied in this experiment and is usually a combination of inorganic and organic sources.



Year of planting: 2008; Harvesting period: Sep 2010 to Feb 2011, n=25x 4= 100 plants for this experiment; Site: Neswad

Figure 49. Effect of different sources of nutrients and doses on *Jatropha* yield in 2011-12.

In the subsequent year (2012-13), it was found that the *Jatropha* cake application (@ 100 kg N/ha) led to the highest seed yield of 2.08 tonnes per hectare which was significantly different and higher than the other treatments viz. application of 100% recommended dose of nitrogen through farmyard manure or inorganic source (urea). 50% dose of nitrogen (50 kg N/ha) gave significantly lower seed yield of 1.33 t/ha **(Figure 50)**. Thus, it may be seen that a seed yield of 2t/ha or more is definitely realizable after 5-6 years provided agronomic requirements are met and a good choice of germplasm is made with emphasis on a true-to-type plantation.

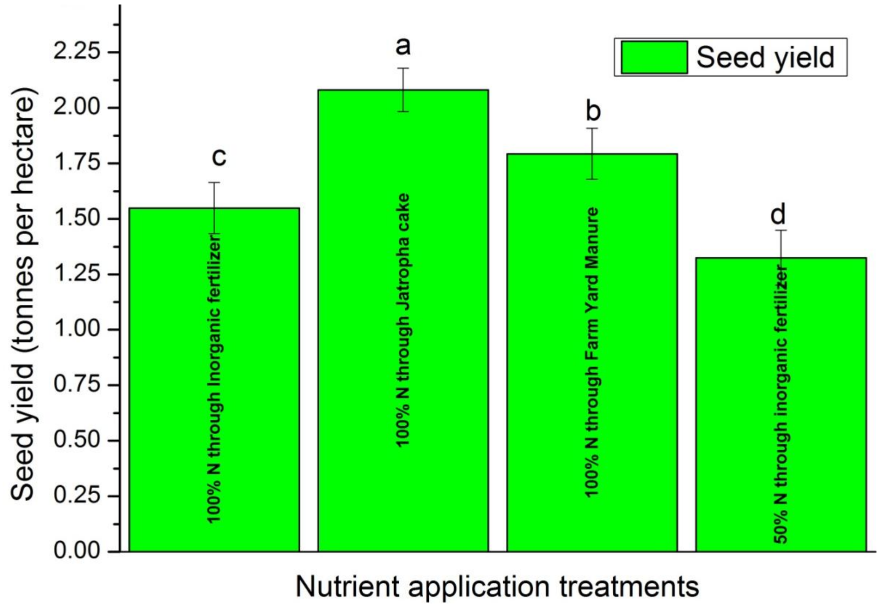


Figure 50. Effect of *Jatropha* cake, FYM and inorganic fertilizers on seed yield of *Jatropha* in harvesting season of 2012-13 (Date of planting 2007-08).

To further understand the correlation with the yield behavior, a systematic soil bio-chemical study was undertaken for this experiment, wherein, different soil enzymes playing important roles in nutrient recycling were analyzed for the soil sampled in Feb 2012 (8 months after nutrient application). It can be seen that the enzymatic activity in the cake treated soils remains higher than that of the other treatments for a prolonged period of time **(Table 12)**. Further, aryl sulphatase was found positively and significantly correlated ($p<0.05$) with both alkaline phosphatase and glucosidase.

Table 12. Effect of organic and inorganic amendments on soil enzymatic activities in *Jatropha* plantation at Neswad

Treatments	**Soil enzyme activity**				
	Dehydrogenase activity (measured as the amount of TPF formed/ 100ml)	Aryl sulphatase activity (measured as µg of nitrophenol released/h/g of soil)	Acid Phosphatase activity (measured as µg of nitrophenol released/h/g of soil)	Alkaline phosphatase activity (measured as µg of nitrophenol released/h/g of soil)	Glucosidase activity (measured as µg of nitrophenol released/h/g of soil)
100 kg N/ha	170.5 ± 5.4	16.0 ± 1.3	210.4 ± 13.1	111.0 ± 14.7	196.7 ± 16.1
2.6 tonne Cake/ha	236.2 ± 20.4	20.2 ± 1.5	309.3 ± 21.0	168.8 ± 25.1	217.5 ± 14.9
20 tonne FYM/ha	171.6 ± 20.1	24.9 ± 1.2	202.1 ± 9.4	190.7 ± 6.2	145.2 ± 8.3
50 kg N/ha	134.0 ± 11.0	15.5 ± 1.8	236.3 ± 18.2	138.3 ± 17.2	104.1 ± 6.0
CD at 5%	NS	NS	NS	NS	NS

Values represented are mean ± SEM of 5 replicates; Soil collection period: Feb 2012; Year of planting: 2007-08; n=25x 4= 100 plants for this experiment; Site: Neswad

Effect of cake on soil biological properties

Apart from assessing the effect of *Jatropha* cake on seed productivity and soil biochemical properties, its effect on soil micro-biological diversity was also carried out. The soil microbial community plays a fundamental role in decomposition, nutrient cycling, and energy flow. The beneficial effects of soil microorganisms range from nitrogen fixation and organic matter decomposition to the breakdown of metabolic by-products and agrochemicals, enhancing the bioavailability of nitrates, sulphates, phosphates, and essential metals. Plant health and development is maintained by supplementing nutrients through nutrient cycling generated by microbes in the rhizosphere. The rhizosphere microbial community is influenced by the culture conditions and plant species because of micro-climate variations, differences in root exudation and rhizo-deposition in different root zones. Thus, soil microbial diversity analysis was carried out as a part of this study to assess whether there is any shift in the microbial community due to the suggested management techniques for growing *Jatropha*.

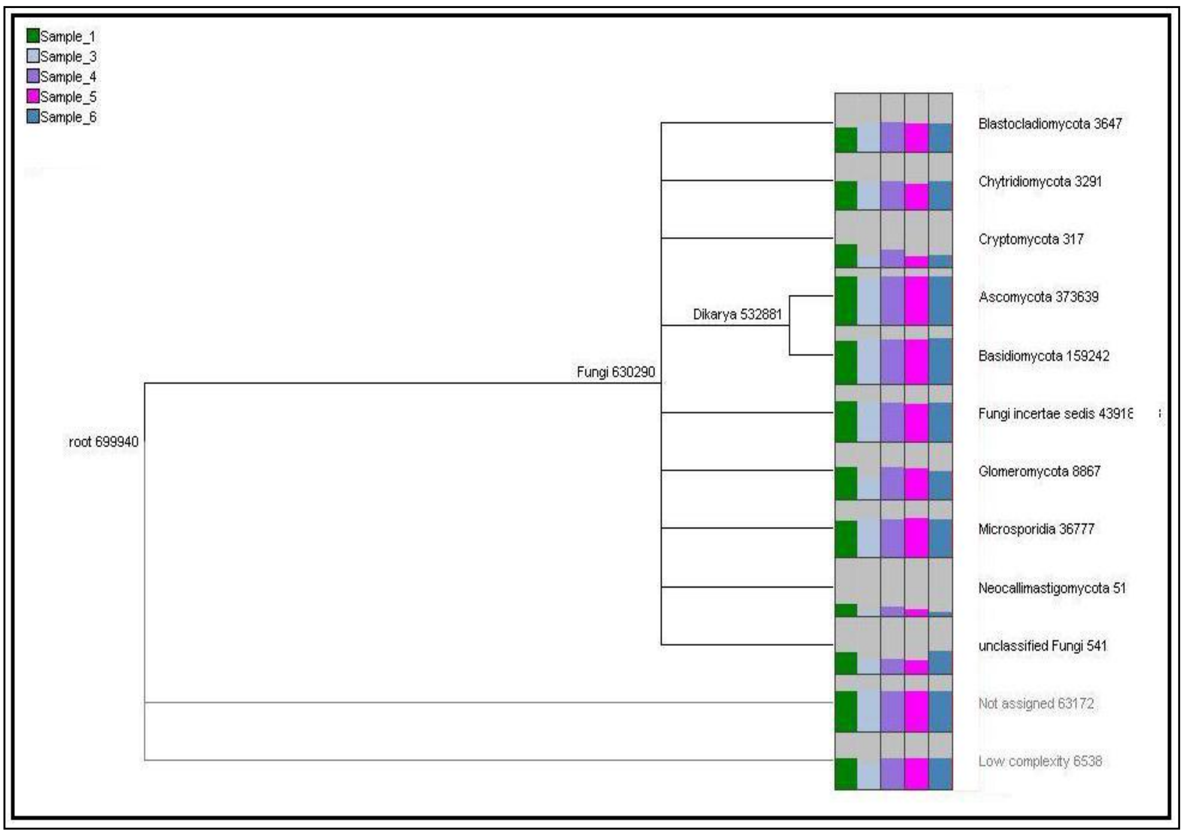


Figure 51. Comparative analyses of metagenomes of 5 soil samples for fungus in the nutrient management experiment

NB: Sample 1: 50% N through inorganic; Sample 3: 100% N through inorganic; Sample 4: 100% N through FYM; Sample 5: wasteland soil with no *Jatropha*; Sample 6: 100% N through cake

A comparison was drawn on the soil bacterial and fungal populations ascertained by a metagenomic approach between native (pre-land amendment) and *Jatropha* rhizosphere with different inorganic and organic treatments.

The study of bacterial and fungal diversity in the soil was carried out through r-DNA analysis using 16S and 18S rDNA analysis. 18S rDNA analysis was used to study the fungal diversity of the soil and 16S rDNA was used for bacterial diversity analysis. The land was amended following the earlier described approach and planted with *Jatropha*. The soil samples were then collected from rhizosphere of *Jatropha curcas* with different treatments in November 2012. Comparative analysis of metagenomes of 5 soil samples for fungus in the nutrient management experiment are presented in **Figure 51,** and it can be seen that there were shifts in soil fungal community in conditions when barren soil was converted to grow *Jatropha*. Fungal community shifts were also observed between inorganic and organic nutrient management treatments. Importantly, the data revealed that *Macrophomina phaseolina*, a fungus, that causes deadly collar rot disease in *Jatropha*, aggravates when *Jatropha* is cultivated with only inorganic amendments while the population goes down if *Jatropha* cake is amended in the soil **(Figure 52)**.

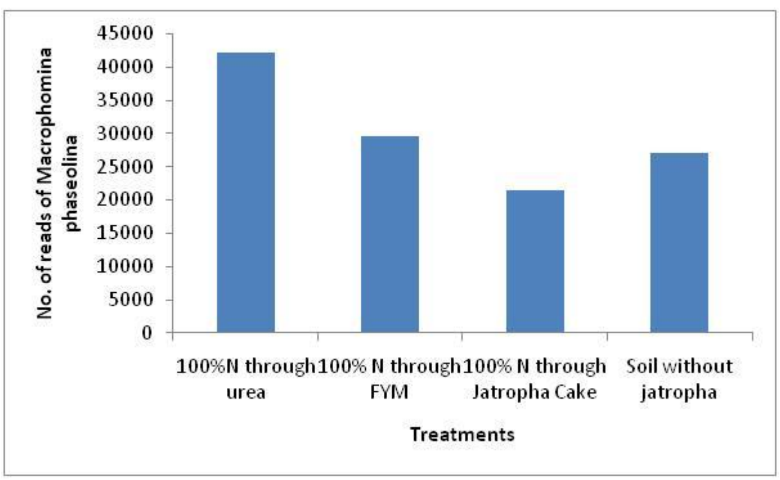


Figure 52. Comparison table of *Macrophomina phaseolina* reads with 18S metagenome.

Similarly, soil a metagenome analysis of bacteria was also done and changes were recorded by different treatment application (**Figure 53**).

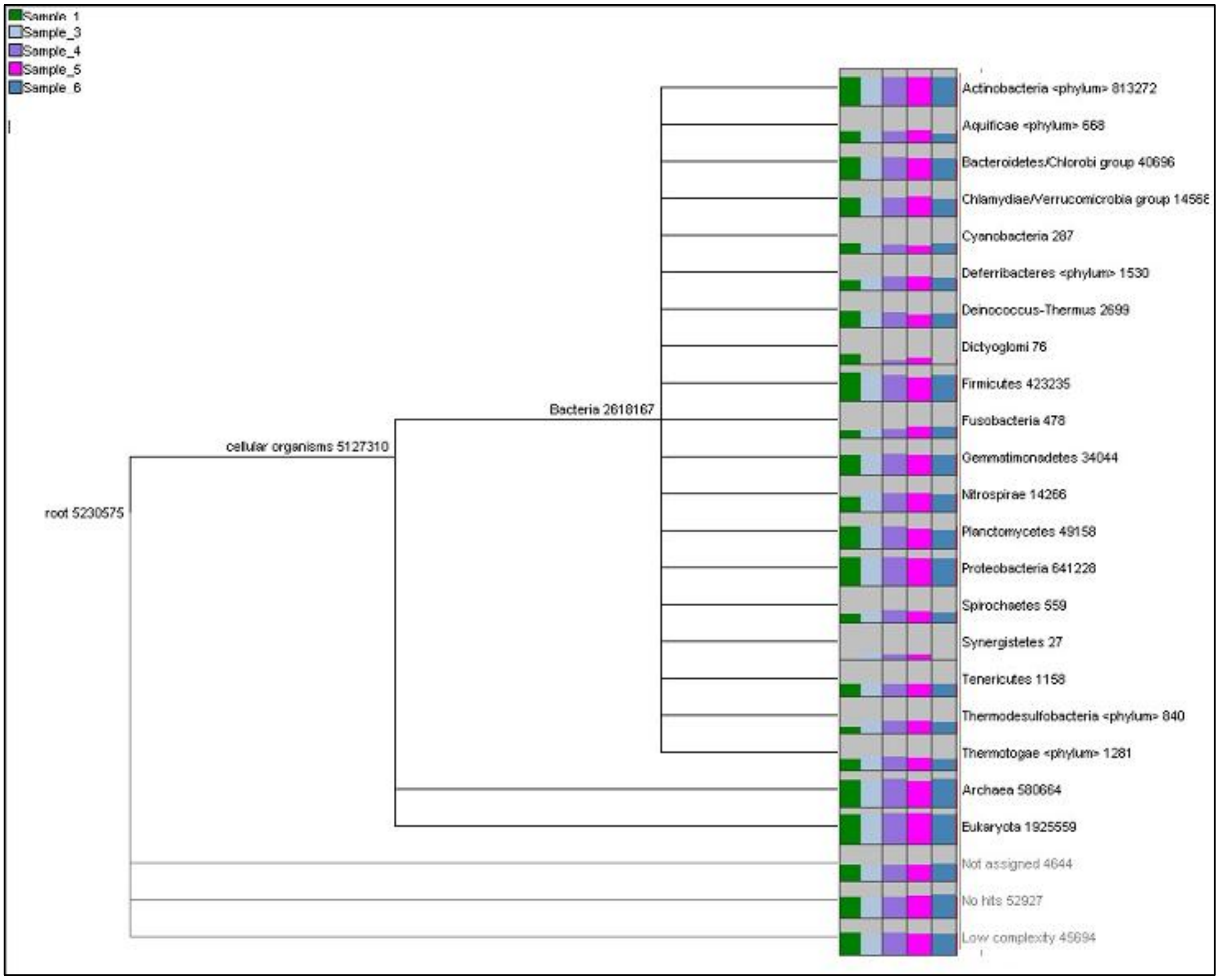


Figure 53. Comparative analysis of metagenomes of 5 soil samples for bacteria in the nutrient management experiment.

NB: Sample 1: 50% N through inorganic; Sample 3: 100% N through inorganic; Sample 4: 100% N through FYM; Sample 5: wasteland soil with no *Jatropha*; Sample 6: 100% N through cake

There was shift in the gram negative bacteria population with a less proportional gram negative bacteria diversity to the total in samples treated with *Jatropha* cake as compared to soil without *Jatropha* or with 100% inorganic fertilizer application. There was a marked increase in gram positive bacterial reads in either inorganic or cake amended soil under *Jatropha* cultivation when compared to soil where no *Jatropha* **(Figure 54).**

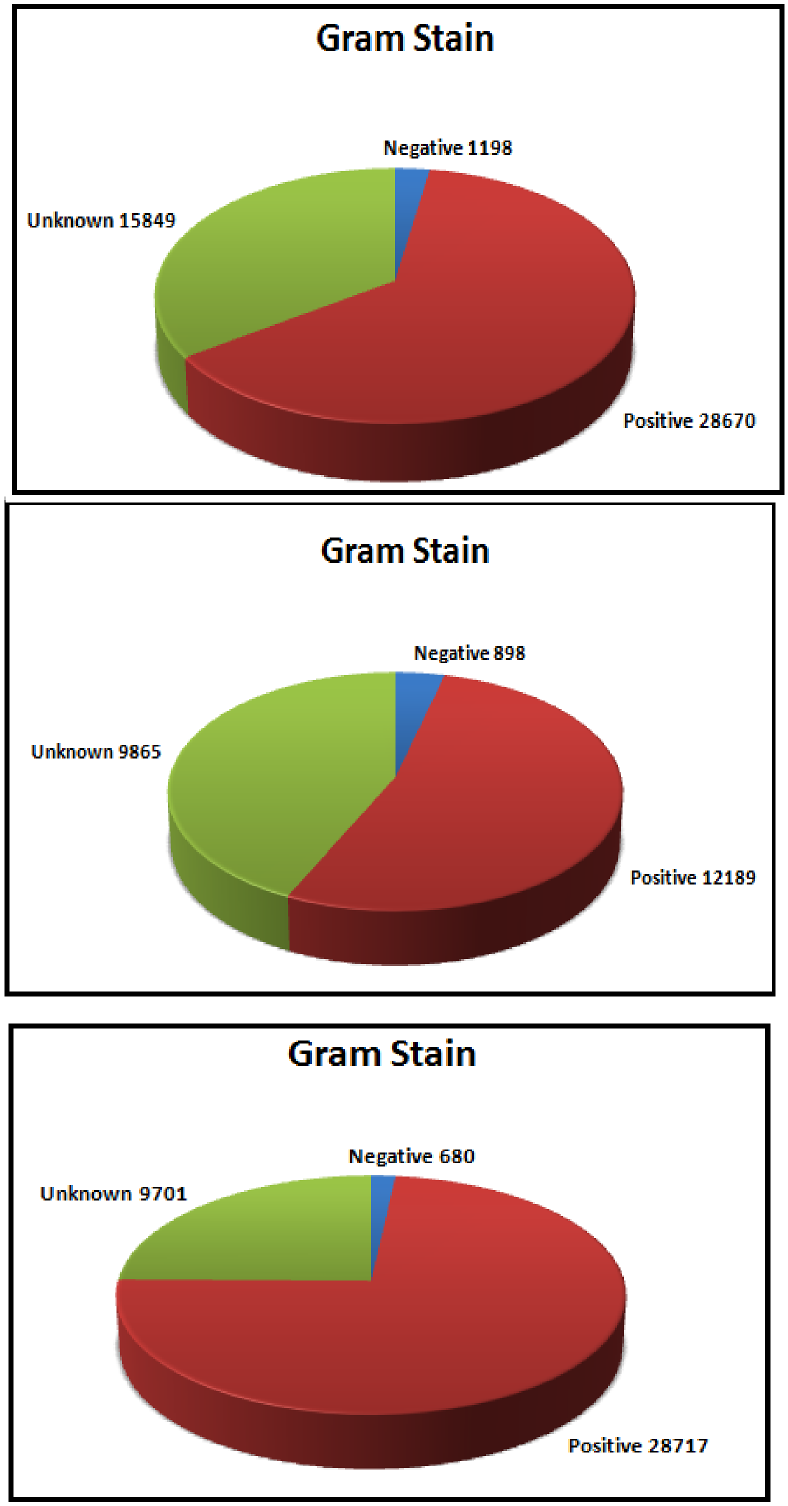


Figure 54. Bacterial read distribution in treatments 100% N to *Jatropha* through inorganic source (top), *Jatropha* cake (middle) and outside soil without *Jatropha* (bottom) according to gram stain.

Effect of long term N and P fertilization on soil nutrient availability

Effect of long term application (since 2004) of different combinations of N and P fertilizer to *Jatropha* soil was determined in existing experimental plantation at Chorvadla, Bhavnagar. Particularly, the effect on available soil nutrients including carbon, N, P and K were determined.

Treatment details

Factorial Randomized Block Experiment
3 replications
Nitrogen (N0=0; N1=40, N2=80, N3=120 kg N/ha)
Phosphorus (P0= 0, P1=15, P2=30, P3=45 kg P_2O_5/ha)

Table 13. Effect of N and P on organic carbon in soil

N&P level	P0	P1	P2	P3	Mean
N0	0.70	0.55	0.52	0.57	0.58
N1	0.61	0.53	0.57	0.62	0.58
N2	0.54	0.56	0.55	0.58	0.56
N3	0.60	0.55	0.61	0.57	0.58
Mean	0.61	0.54	0.56	0.58	

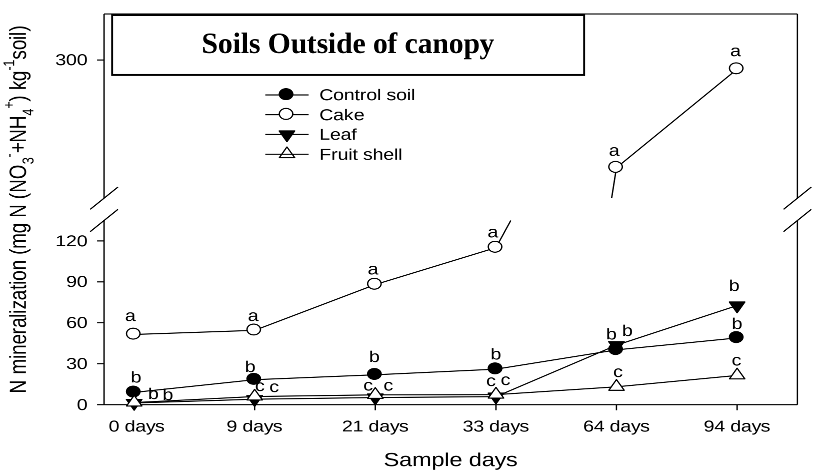
Table 14. Effect of N and P on available P kg/ha in soil

N&P level	P0	P1	P2	P3	Mean
N0	11.28	12.10	16.39	24.51	16.07
N1	6.94	10.13	12.24	12.19	10.38
N2	5.49	7.89	16.57	21.71	12.91
N3	6.00	8.41	9.60	13.40	9.35
Mean	7.43	9.63	13.70	17.95	

Results of the analysis of soil samples from N and P fertilizer experiments showed that there was no significant effect of nitrogen and phosphorus application on organic carbon (because of high temperature in tropical condition) **(Table 13)**, total nitrogen and available potassium. However, available P was significantly affected. There was a significant independent effect of N and P application on available P but the interaction (NXP) effect was non-significant **(Table 14)**. The availability of P was significantly decreased by N application over control (N0) however, the availability of P was significantly increased by P application at 30 kg and above levels of P_2O_5.

Studies to characterize the nitrogen (N) mineralization of *Jatropha* residue during decomposition in soil beneath or outside *Jatropha* canopy.

A study was done with the objective to characterize the nitrogen (N) mineralization of *Jatropha* residue during the decomposition in soil beneath or outside the *Jatropha* canopy. Two laboratory incubation (94 days) studies (beneath and outside *Jatropha* canopy soil) had four *Jatropha* residue soil amendments (fruit shell, leaf, cake or control of soil only). The results showed that only the *Jatropha* cake caused net N mineralization. Soil amended with leaf and fruit shell residues immobilized N during the first 64 and up to 94 days respectively **(Figure 55 a&b)**.



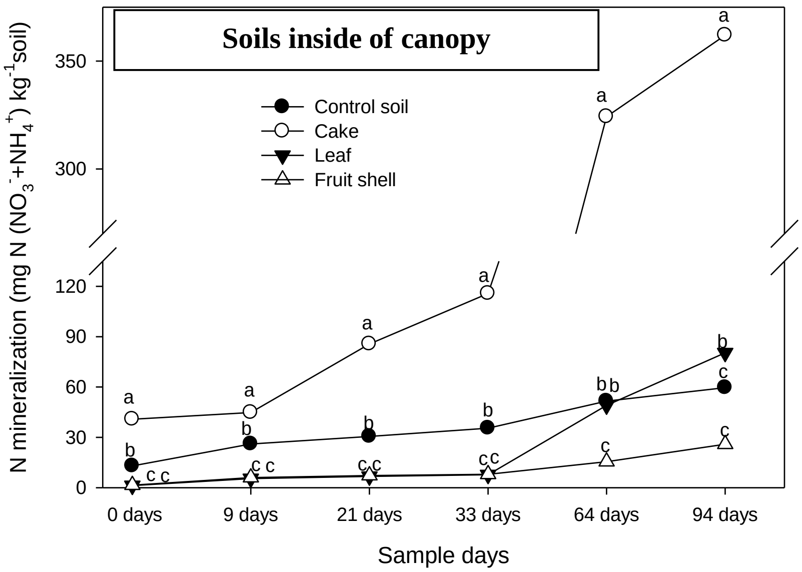


Figure 55 a&b. Nitrogen mineralization in *Jatropha* residue amended soil.

Efforts towards control of fungal disease

The collar rot caused by *Macrophomina* causes heavy losses on the *Jatropha curcas* plantations. *Macrophomina* is soil borne, anamorphic, ascomycetes fungi. Microsclerotia in the soil, seed, or host tissue serve as inoculum and plants die/decay due to the production of fungal toxin phaseolinone and the production of fungal tissue that plug the host xylem vessels. This disease can play havoc and can wipe out an entire plantation at one go, if not controlled. The disease onset is during summer and the symptoms are apparent when most of the damage is already done to the plant. The symptoms are visible just after the onsets of rainy season when the healthy plants sendoff prolific new shoots while the affected plants fail to produce green leaves and shoots **(Figure 56)**.

Figure 56. Photograph of 7 year old *Jatropha* tree affected by collar rot (plant situated in middle showing pale green wilted leaves) flanked by healthy plants on both sides which show dark green leaves, photograph 2013 **(left);** Clear demarcation between two accession with one accession CSM U-1 showing severe susceptibility to collar rot disease (c.o. *Macrophomina phaseolina*) photograph 2008-09 **(right).**

Systematic work was initiated to impart fungal tolerance in the *Jatropha* plant. Salicylic acid is known to enhance systemic acquired resistance in plants. A number of pathogenesis related genes, playing an important role in imparting disease tolerance are also induced by salicylic acid.

Towards, developing tolerance in *J. curcas*, we have isolated the cDNA of a stress responsive transcription factor JcWRKY and pathogenesis related gene (JcPR-10a, NCBI accession number JN566052) from *Jatropha curcas* with an open reading frame of 483 bp. The deduced amino acid of JcPR-10a on blast analysis shows homology to plant PR10 proteins, with 43%, 42% and 39% identity to Mal d (PR10), MtPR10-1 and CaPR-10/VvPR10.2, respectively. Phylogenetic analysis shows that JcPR-10a gets clustered with dicots, close to CaPR-10, which is found to play an important role in both fungal and viral resistance. The 568 bp genomic clone (NCBI accession number JN639632) shows the presence of an intron at 185 bp, similar to other PR-10 genes. Transcript expression of *JcPR*-10a was upregulated in response to different stimuli such as NaCl, salicylic acid, methyl jasmonate and *M. phaseolina* **(Figure 57)**. We are the first to report transcript accumulation of a pathogenesis-related gene *JcPR-10a* with response to *Macrophomina.*

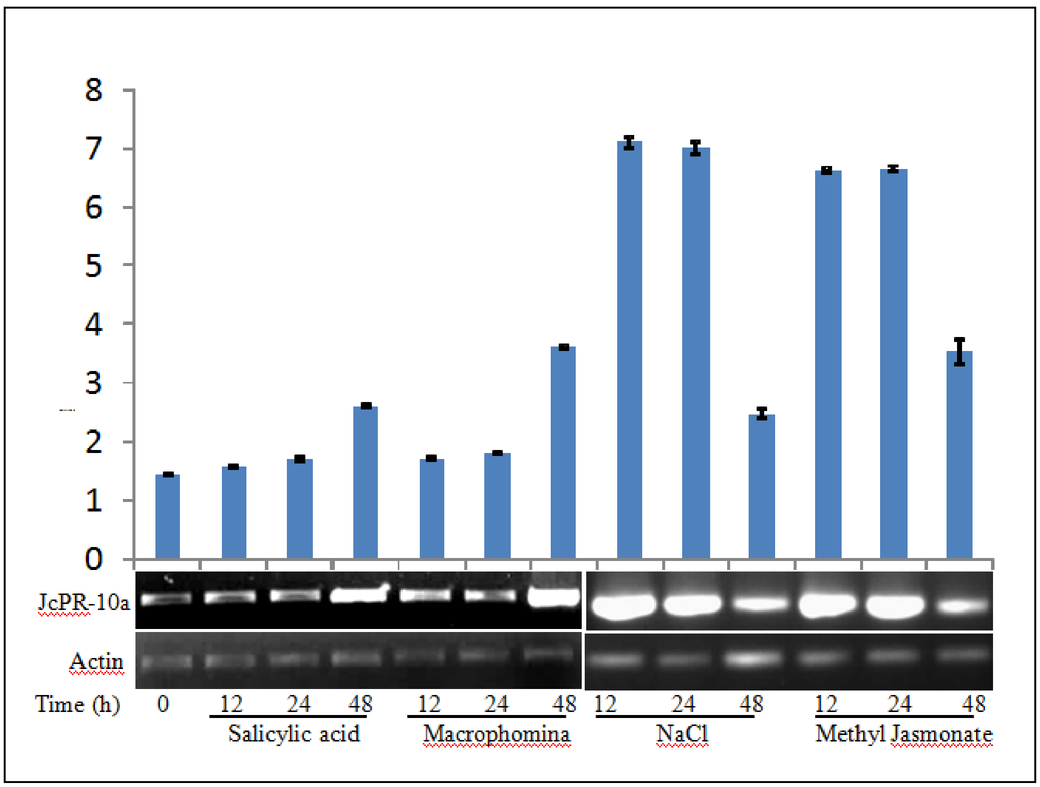


Figure 57. Transcript analysis of JcPR-10a gene under different stress treatments, 2.5 mM salicylic acid, Macrophomina fungus, 250 mM NaCl, 100 µM methyl jasmonate. Graphs over each radiograph show the relative value of JcPR- 10a against the actin gene to show the quantitative increase in the expression levels.

The JcPR-10a recombinant *E. coli* cells showed better growth in LB medium supplemented with NaCl, whereas growth of the recombinant cells was inhibited in LB medium supplemented with KCl, mannitol, sorbitol, methyl jasmonate, and salicylic acid **(Figure 58)**. The JcPR-10a protein was over expressed in *E.coli* cells and was purified to homogeneity. The purified protein exhibited RNase at a wide range of pH (4-9) and DNase activity. JcPR-10a recombinant protein inhibits the growth of fungus *Macrophomina*. The fungal growth was found inhibited in the 2-3 mm zone in proximity to the discs **(Figure 59a)**. However, with boiled recombinant protein and only protein elution buffer, no growth inhibition was observed **(Figure 59b)**. Microscopic observations show that the untreated hyphae are thin with pointed tips **(Figure 59c, e)** whereas, on treatment with JcPR10a protein, the tips get more rounded **(Figure 59d, f)** and it appears that the hyphal growth gets stunted. The protein showed antifungal activity against *Macrophomina*, indicating that JcPR-10a can serve as an important candidate to engineer stress tolerance in *Jatropha* as well as other plants susceptible to collar rot by *Macrophomina.*

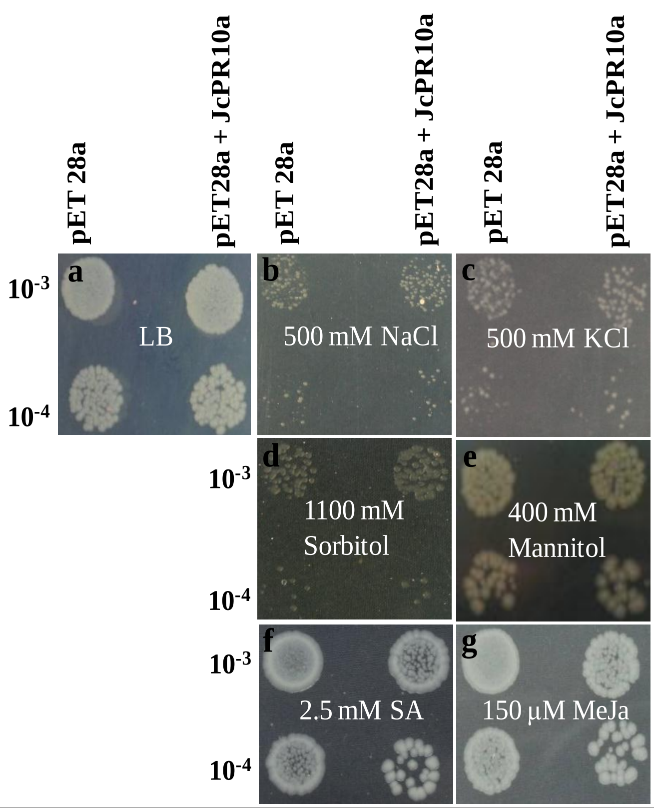


Figure 58 The growth analysis of *E.coli* cells containing JcPR-10a recombinant plasmid and pET28a alone in LB medium (solid) with different supplements. (a) LB only (b) 500 mM NaCl. (c) 500 KCl. (d) 1,100 mM sorbitol. (e) 400 mM mannitol. (f) 2.5 mM salicylic acid. (g) 150 µM methyl jasmonate.

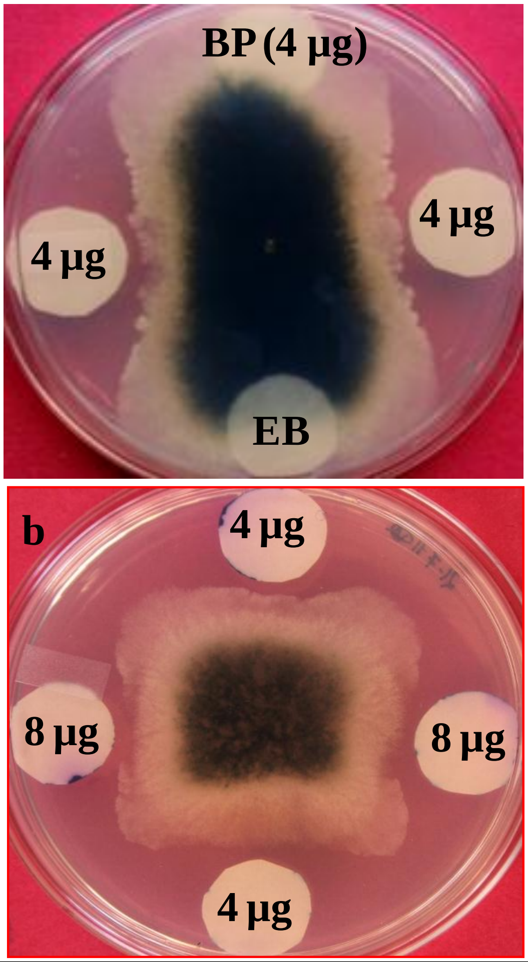
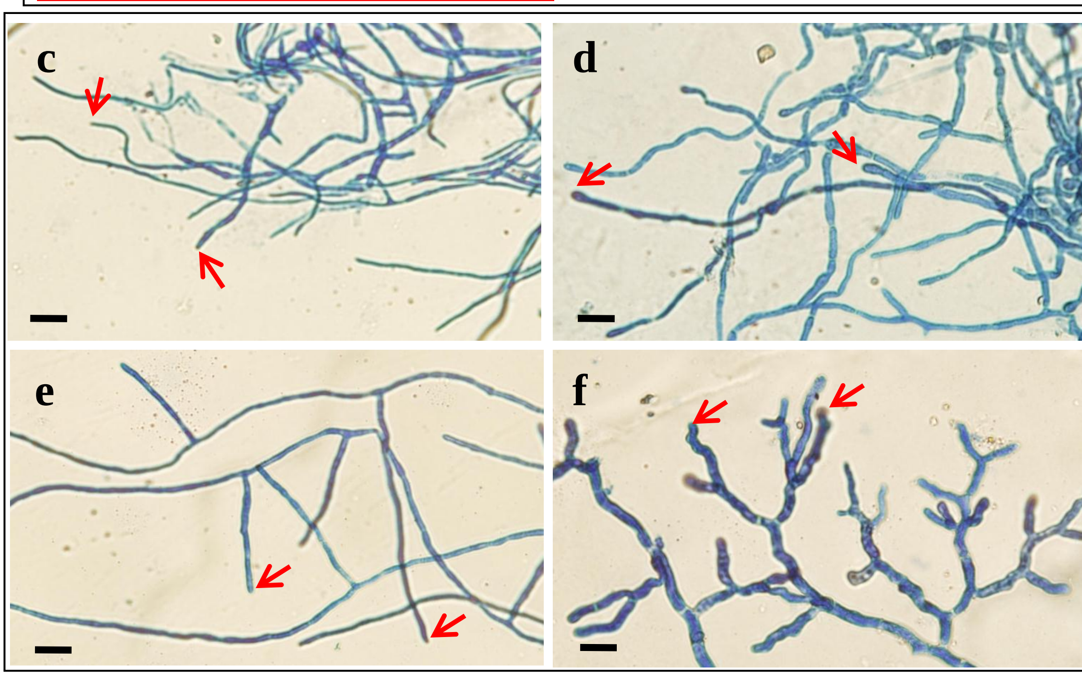


Figure 59 JcPR-10a protein inhibits the growth of fungus *Macrophomina*. (a) 4 µg of JcPR10-a recombinant protein, boiled protein (BP) and protein elution buffer (EB) was used. (b) 4 and 8 µg of JcPR10-a recombinant protein was used. (c, e) Hyphal morphology of *Macrophomina* under control conditions at 24 and 48 h, respectively. (d, f) Hyphal morphology of *Macrophomina* on treatment with 8 µg recombinant protein at 24 and 48 h, respectively. Scale bar 100 µm.



Year-on-Year dry matter accumulation in *Jatropha* plants

Jatropha sequesters atmospheric carbon in its biomass as it grows over a period of years. Accordingly, dry biomass was recorded for the plantations which were established 5 to 7 years ago in Gujarat. It was found that *Jatropha* accumulated 7.7 kg/plant or 8.6 t of dry biomass per hectare (3m x 3m spacing) in its leaves, stem and branches, while excluding fruits at Neswad, Bhavnagar after 5 years of planting. The *Jatropha* plantation established in 2004 similarly accumulated 14.6 t/plant or 24.3 t of dry biomass per hectare (at 3m x 2 m spacing; **Figure 61**). The year on-year biomass accumulation data is depicted in **Figure 60**.

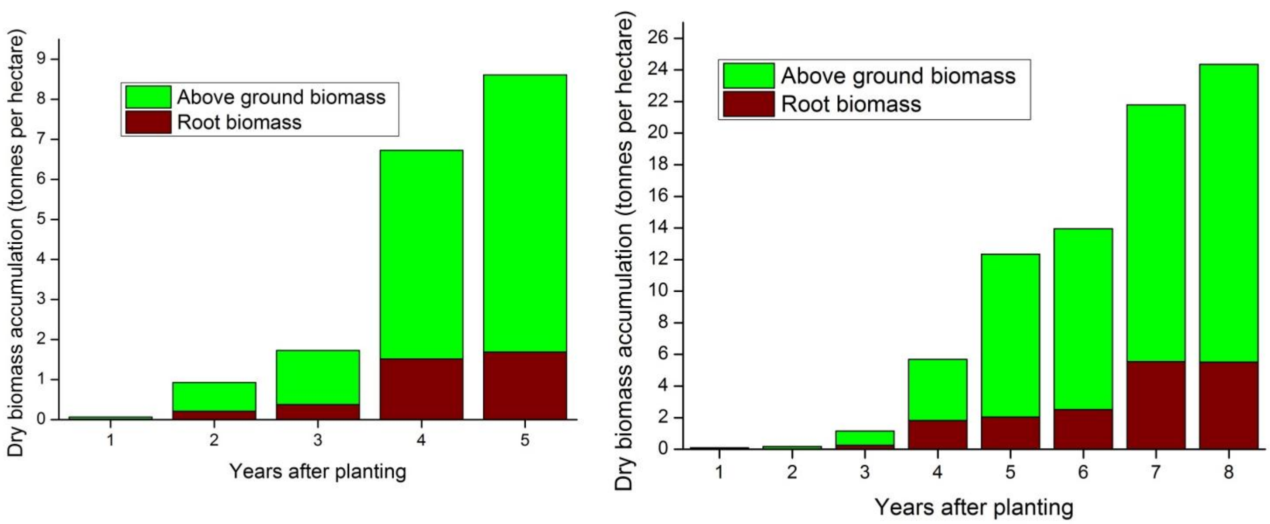


Figure 60. Dry biomass accumulation in *Jatropha* over the years (excluding fruit weight) at Neswad (left) and Chorvadla (right).

Figure 61. Older plantation at Chorvadla, Bhavnagar district, Gujarat showing huge biomass build up.

Life cycle assessment

Jatropha curcas has been actively promoted as a biofuel crop for wastelands and as a likely solution to energy scarcity without compromising food and environmental security. It is desirable to prove this hypothesis by evaluating the sustainability of existing *Jatropha* plantations with emphasis on carbon and energy budgets using the Life Cycle Assessment (LCA). Both environmental and social impacts of many biofuels including ethanol are being contended now days. A few studies on *Jatropha* have been carried out by some researchers. However, it is felt necessary to quantify and verify the environmental impacts of *Jatropha* cultivation on marginal lands based on the primary data generated by us from the life cycle point of view. Thus life cycle assessment was done within the purview of this project.

LCA objective and scope

The study was carried out to evaluate the environmental impacts during cultivation of *Jatropha curcas* on marginal wastelands in Gujarat, India for the production of *Jatropha* oil which can be converted into biodiesel. Cultivation of *Jatropha* on wastelands is a challenge owing to their poor soil quality and lack of water especially during the summer season. Further, the depth of top soil in these regions was only ca. 20-30 cm beneath which were bed rocks that prevented sustainable growth and establishment of the plantation as the rocks prevented the deep penetration of the roots and better anchorage. Thus, a method was developed (described in earlier sections) for planting *Jatropha curcas* in such wastelands so as to make a conducive zone for the roots to penetrate deeper down. Such a model also allows better anchorage of the plants. Further, the optimal material inputs and agro-technology that is required for cultivation were determined which would enable the plantation to sustain for period of time (20 years base case assumption). In this model, we propose irrigating the plants for 5-6 months especially during the later parts of winter through summer till the onset of monsoon. The rest of the period is rain fed. Further, this model also envisages the use of de-oiled cake as organic amendment to offset the use of mineral fertilizers which would otherwise be required to fulfill the nutrient requirements of the plant in order to obtain a sustainable average yield of 2 tons per hectare per year for period of 20 years. We have already demonstrated the feasibility of mature Jatropha plantations yielding fruit @ 3t/ha (seed @ 2t/ha) in the previous sections using good germplasm and following proper agro-practices. The model also proposes the generation of briquettes from the shells (having calorific value similar to coal) which can serve as a source of fuel in traditional Indian villages. Thus the model proposes a decentralized scheme for promoting the cause of "local produce for local use", the concept mooted by us (Ghosh *et al* 2007). Accordingly, the oil extraction is proposed through mechanical screw expeller available at villages and envisages a cooperative model. The various impact categories that were assessed for the model include 100 year Global Warming Potential (GWP), Acidification Potential (AP), Eutrophication Potential (EP) and Ozone Depletion Potential (ODP) and net carbon footprint. All the analysis was carried out using GaBi software (version 6). The proposed model is based on all the inputs that are required on a per hectare basis and annualized and hence the amount of oil (510 kg) extracted from one hectare of plantation under the given conditions would serve as one functional unit. The GaBi tool of LCA offers flexibility to scale up the functional unit to any extent as desired and since the life cycle inventories are based on averaged data over period of years and areas, estimation of Life Cycle Impact Assessment is possible for any number of hectares and any number of years by scaling up the functional unit.

Study design and system boundary

The geographic boundary of the study is Gujarat, India. The life cycle system boundary of *Jatropha* oil production from wasteland that is being proposed here includes land preparation, nursery raising, irrigation, fertilizer and its application, pesticide production and its use, de-husking of *Jatropha* capsules, briquetting of shell, and using part of shells to produce steam required for softening of seeds during *Jatropha* oil extraction. The system boundary expansions considered in the study also includes use of de-oiled cake as organic amendments to the soil and net generation of biomass carbon in the standing crop over a period of time after offsetting the biomass carbon removed prior to planting on wastelands. LCA for biodiesel production (based on our patented process) and its use was, however, kept out of the scope of the present investigation on account of shortened duration of the project, although the impact is expected to be minimal. The LCA was done for the production of 510 kg *Jatropha* oil annually from 1 hectare in addition to realizing 1.53 tonnes of *Jatropha* cake, the majority of which is used to offset chemical fertilizer requirement, although credit on account of offset was not taken into consideration. Fruit yield per hectare is the most important parameter while determining the sustainability of *Jatropha* oil production and its subsequent biodiesel conversion. The productivity was assumed by averaging and extrapolating the present day *Jatropha* capsule (fruit) yield (3t/ha) over a period of 20 years, an assumption that seems to be realistic given the reported life span of *Jatropha* to be more than 50 years. Further, a conservative oil extraction efficiency of 25% has been taken, while extracting seed oil in traditional screw type oil expeller, since decentralized model of *Jatropha* biodiesel production is targeted. The production of briquettes has also been included which is also a source of solid form of bio-energy. The carbon sequestered in the standing biomass has been extrapolated and averaged over 20 years period and has been accounted for while carrying out the LCA and the carbon outgo on account of removal of native biomass before planting of *Jatropha* has also been similarly accounted for. The inputs used for the present LCA are based on learning and data obtained from older plantations at Chorvadla and Neswad villages of Gujarat. While new plantations were also established at Kanod and Bhadrawal sites as a part of the project, they were too young at the end of the project for their yield and other generated data to be included in the analysis. The carbon sequestered by *Jatropha* fruit (which yields oil, briquettes and cake) has been excluded since the LCA has been limited to the production of these products, while also excluding their use phase.

Data and inventory analysis

Jatropha cultivation on wastelands was modeled on GaBi 6.0 product sustainability software (PE International, Germany). Custom modules were built for the approximation to meet India specific conditions. Wherever India specific data sets were not available the European datasets which closely approximated Indian conditions were used from Eco-invent integrated LCA software. India specific diesel and electricity data sets provided by PE sustainability solutions as part of the integrated eco-invent database were used in the model. The detailed life cycle inventory for different activities carried out based on experience and best practices identified so far are described in the following sections. The base case inputs for the model and their life cycle impact assessment of each of the individual modules have also been described in detail in the appendix section.

Results, discussion and data Interpretation of LCA

Four different parameters were employed in the present study to evaluate the various environmental impacts on the entire life cycle analysis of *Jatropha* oil production. These are GHG emissions as measured by Greenhouse Warming Potential (Kg CO_2 eq), Acidification Potential (Kg SO_2 eq), Eutrophication Potential (Kg Phosphate eq) and Ozone Depleting Potential (Kg R11 eq). The LCIA-CML 2001(Nov, 2011) procedure was employed for all the balance calculations analysis. For a comprehensive understanding, we discuss the results in two parts:

Plantation model: Life Cycle Impact Assessment of *Jatropha* plantation in 1 ha area (1 year) having an annual average *Jatropha* fruit production of 3 Metric tonnes. (Upstream component only)

For this, a plan was modeled, wherein the inventory values for activities like land clearing, burning of biomass for energy and nursery, which are to be carried out once in a life time and thus have cumulative impact, was distributed across the life cycle duration assumed in this study (20 years) and calculated for one year. Similarly, total biomass built up over 20 years was also distributed and allocation for one year was made. All other inventory values having an annual nature of inflow and outflow, like fertilization, irrigation, inter-culture operation etc. were used as such for this model that represents LCA for 1 ha and 1 year. Manpower was not included in the model as it is not customary to include manpower while conducting LCA and hence no impacts were assumed due to activities carried out manually, e.g., harvesting, manual fertilizer and pesticide applications etc. The model thus developed represents annualized input and output flows for 1 ha plantation yielding 3 t fruits from it. The products such as *Jatropha* oil, cake and shell/briquettes can be obtained from fruits and have been just indicated pictorially, however, the impacts attributed to their conversion is not included in this plantation model **(Figure C8)**.

Aggregate model: LCIA of products from 1 ha *Jatropha* plantation viz., oil, briquette and cake production (impacts attributed to upstream and downstream components):

This aggregated module is an extension of the plantation model and includes all input and output flows due to the downstream operations, *viz*., decortication, oil expulsion, steam generation, briquetting to yield de-oiled cake (including its application back in field), briquette production and *Jatropha* oil **(Figure F)**.

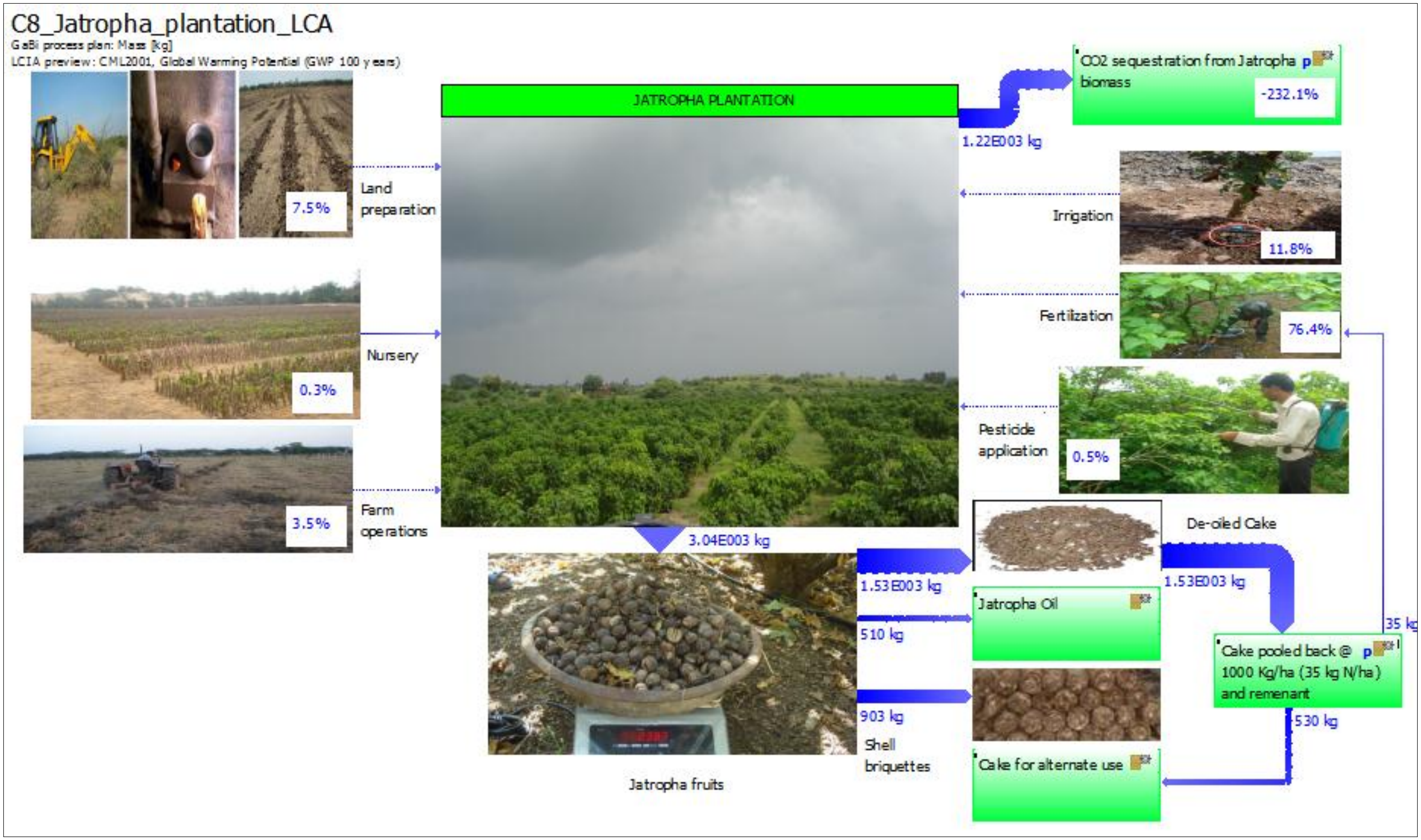


Figure C8. Life cycle modeling and life cycle impact assessment preview for cultivation of *Jatropha* and production of *Jatropha* whole fruits (or capsules) from 1 ha area (1 year).

NB: Figures in % represent GWP 100 years (Global warming potential) based on CML 2001

Plantation model: Both the models discussed above if scaled up 20 times, as the case may be, would reveal the life cycle impact of *Jatropha* fruit production or deriving the co-products, i.e., oil, cake and briquettes from 1 hectare over the assumed functional life of 20 years. However, for the ease of comprehension, the functional unit has been kept as products (fruit or its processed products) derived from 1 ha in 1 year. The assumptions and input values for individual modules used for modeling fruit production from 1 ha is given in **Table 15 and 16**.

Table 15. GaBi plan for *Jatropha* Plantation module

Process name	**Module purpose**	**Comments**
Jatropha plantation	This custom module aggregates and calculates the impacts of all the sub-processes involved in the cultivation of *Jatropha curcas* in one hectare for one year	
Land preparation	This custom module aggregates and calculates the impacts of all sub-processes involved in land use change for one hectare apportioned to one year	
Nursery	This custom module aggregates and calculates the impacts of all the sub-processes involved in raising of cuttings in nursery required for transplanting in one hectare area	
Farm operations	This custom module aggregates and calculates the impacts of emissions resulting from weeding, transport of fertilizers and other miscellaneous items into and within the field for one hectare over a period of one year	
Irrigation	This custom module aggregates and calculates the impacts of all the sub-processes involved in irrigating one hectare of the plantation using a drip system for a period of one year	
Fertilization	This custom module aggregates and calculates the impacts of all fertilizer production and their use in one hectare of the plantation for a period of one year	
Pesticide application	This custom module aggregates and calculates the impacts of pesticide application and their use in one hectare of the plantation for a period of one year	

Table 16. Inputs of *Jatropha* Plantation module

Parameter	Value	Unit	Documentation	Source
Diesel	102.3729	Kg	Amount of diesel utilized in various agricultural machinery for one hectare of *Jatropha* plantation per annum	
Transport	23.1	tkm	Transport of cargo using a 20-28 t lorry	
Urea	119.5652	Kg	Amount of Urea applied to one hectare in one year	Own data
Jatropha cake	1000	Kg	Amount of cake used in the field as a substitute for fertilizer	Own data
Direct N_2O emission factor	0.0065		Emission factor for Indian conditions (N_2O-N/kg N input)	Garg et al., 2012
N_2O leaching Factor	0		No leaching of N_2O is assumed due to use of drip and rocky soil	
Indirect N_2O volatilization emission factor	0.005		Emission factor for N volatilization and re-deposition, kg N_2O–N (kg NH_3–N + NO_X–N Volatilized)	Garg et al., 2012
NOx Fraction	0.21		Total NOx Fraction	IPCC 2006
Leaf residue	1332.2	Kg	Total amount of dry leaf residue per hectare @1.2 kg leaf residue/ plant	Own data
N content in leaf	0.02		2% N content in leaves	Own data
Wood/stem residue	555.5	Kg	Total amount of dead wood and stem residue per hectare per year @ 0.5 kg/ plant	Own data
N content in stem	0.004		0.4 % N content in stems	Own data
N crop residue	28.9	Kg	Total N supplied per year through crop residues (kg/ha)	
NH_3 volatilization factor inorganic	0.08		8% lost as N- NH_3 volatilization of total N supplied as inorganic fertilizer in wheat. Hence a similar value is assumed for *Jatropha*	Sharma et al., 2008
Carbon emission factor	0.2		Carbon emission factor from urea	IPCC 2006
NH_3 volatilization factor organic	0.2		Default Fraction for Volatilization from all organic N fertilizers applied	IPCC 2006
Single Super Phosphate	187.5	Kg	Amount of P_2O_5 supplied as SSP for one hectare	Own data
P leaching fraction	0		Phosphorus leaching fraction is assumed as zero due combination of factors such as scanty rainfall, drip irrigation and presence of rock below	
P content factor	0.00095		The average P content for top soil is assumed to be 0.00095 kg/kg soil	Eco-invent report 15a
P run off fraction	0.175		Refers to the average quantity of P lost through run-off for arable land	Eco-invent report 15a
Correction factor for run off	1.14		Calculated according to formula in Eco-invent report	Eco-invent report 15a

Quantity soil eroded	5000	t/ha/yr	Quantity of soil that is subjected to erosion in the state of Gujarat	Sharda et al., 2013
Enrichment factor for P	1.86		Correction factor that takes into account the higher content of P in eroded soils than average soil	Wilke and Schaub 1996
Fraction to river	0.2		This is the fraction of eroded soil that reaches river	Eco-invent report 15a
Muriate of Potash	33.33333	Kg	Amount of MOP used in one hectare of plantation	
Oceanic transport	203720	Kg km	Transport of Potassic fertilizer from Belarus to India in bulk oceanic transport carrier	
Polyvinyl chloride	22.8	Kg	Amount of PVC granules that is apportioned for one year out of a total of 456.3 kg with an assumed life span of 20 yrs	Polyvinyl chloride
Explosives	0.002	Kg	Apportioned amount of explosive that is used in the well which is assumed to last for 100 yrs and sufficient for 25 ha irrigation	Own data
Number of cuttings	1111		Total number of cuttings planted in one hectare of plantation with 3m X 3m spacing	
Manure	70.4	Kg	Total amount of compost applied to cuttings in nursery.	Own data
Lubricant	1.5755334	Kg	Amount of lubricant used in various agricultural machinery in one hectare of *Jatropha* plantation	Assumed
Water	285080	Kg	Amount of water required for irrigation in one hectare plantation along with that used in nursery	
Fungicide	0.424	Kg	Amount of fungicide that was used per hectare of plantation per annum (active ingredient at 75%)	
Insecticide	0.282	Kg	Amount of insecticide that was used per hectare of plantation per annum active ingredient at 50%	
Jatropha fruits	3044.776	Kg	Assumption on the quantity of fruits that can be harvested from one hectare of a mature plantation in one year (3m x 3m spacing)	
Biomass	1215	Kg	Amount of apportioned biomass generated in one hectare of mature *Jatropha* plantation in one year. Vegetative biomass data of 24.3 t/ha achieved in 8 years was assumed to be the upper limit, and thereafter the values were assumed to constant till 20 years.	Based on own Experimental data and conservative assumption thereof
CO_2 Sequestered	2300	Kg	Amount of CO_2 that is sequestered in the biomass of 1215 kg containing 51.6 % average carbon content in one hectare	Own data

The life cycle impact across all the categories pertaining to overall land preparation is given in **Figure 62a-d.**

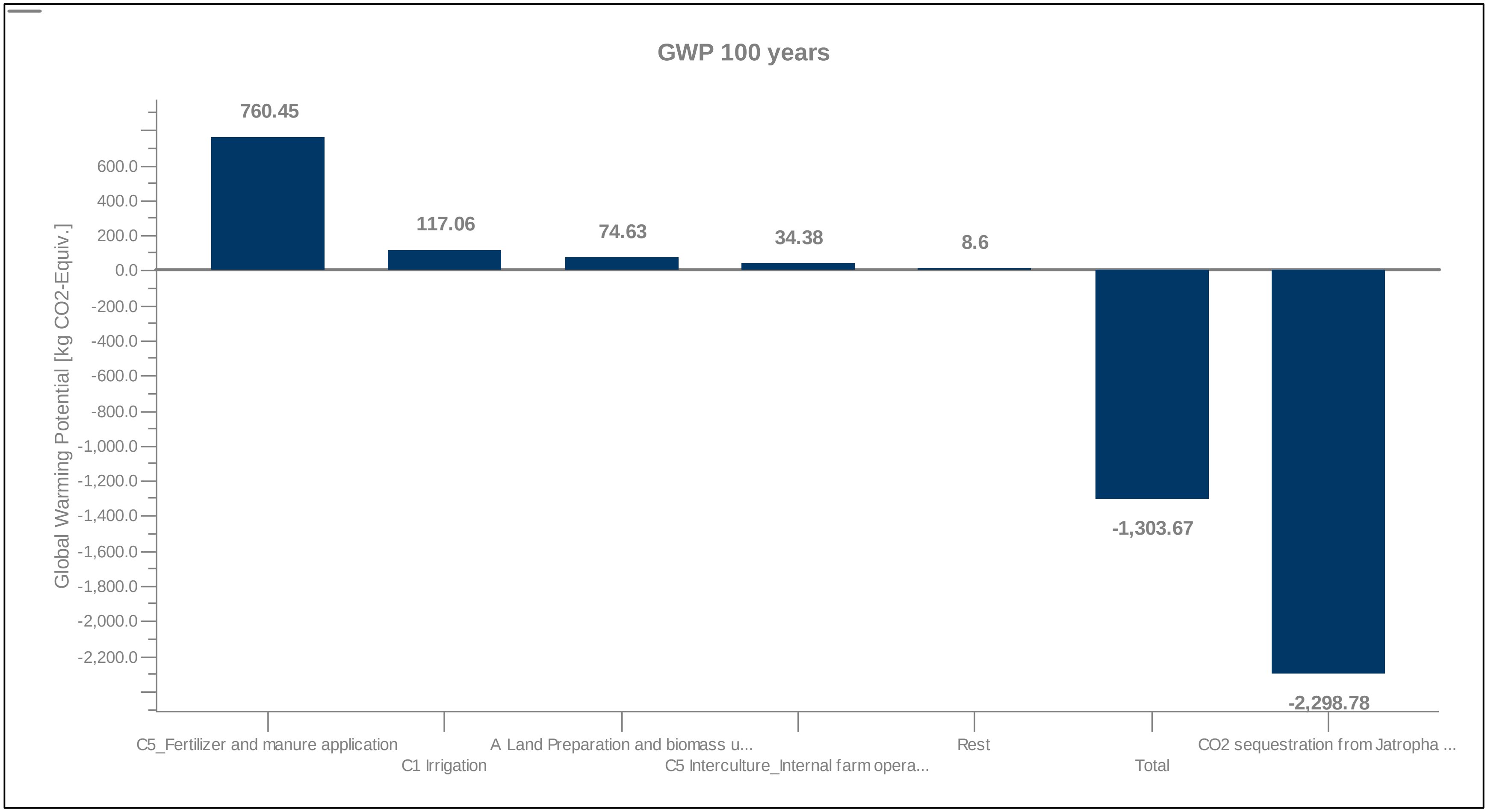


Figure 62a. Life cycle impact of cultivation of *Jatropha* in 1 ha area (1 year): GWP (Global warming potential).

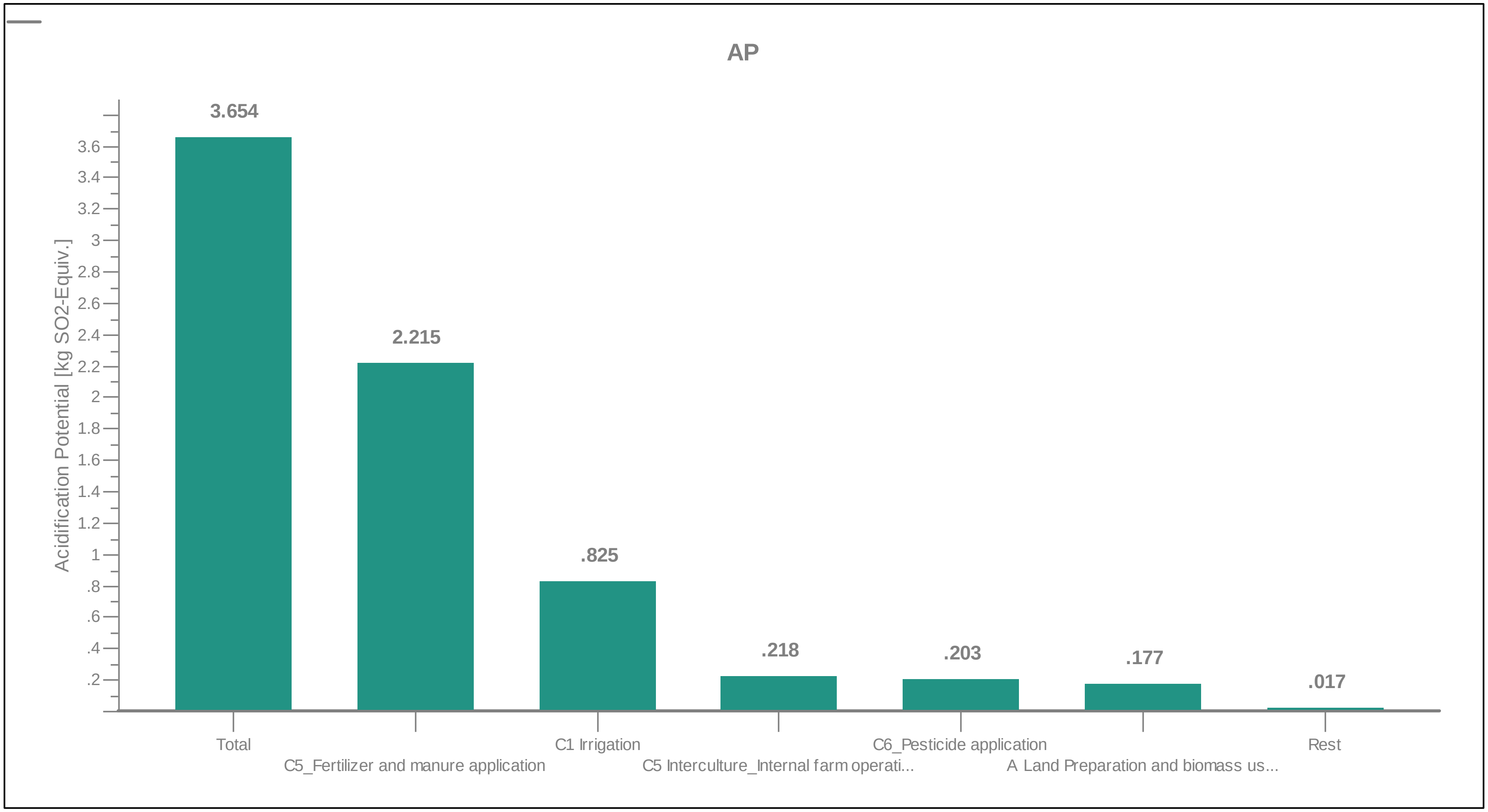


Figure 62b. Life cycle impact of cultivation of *Jatropha* in 1 ha area (1 year): AP (Acidification Potential).

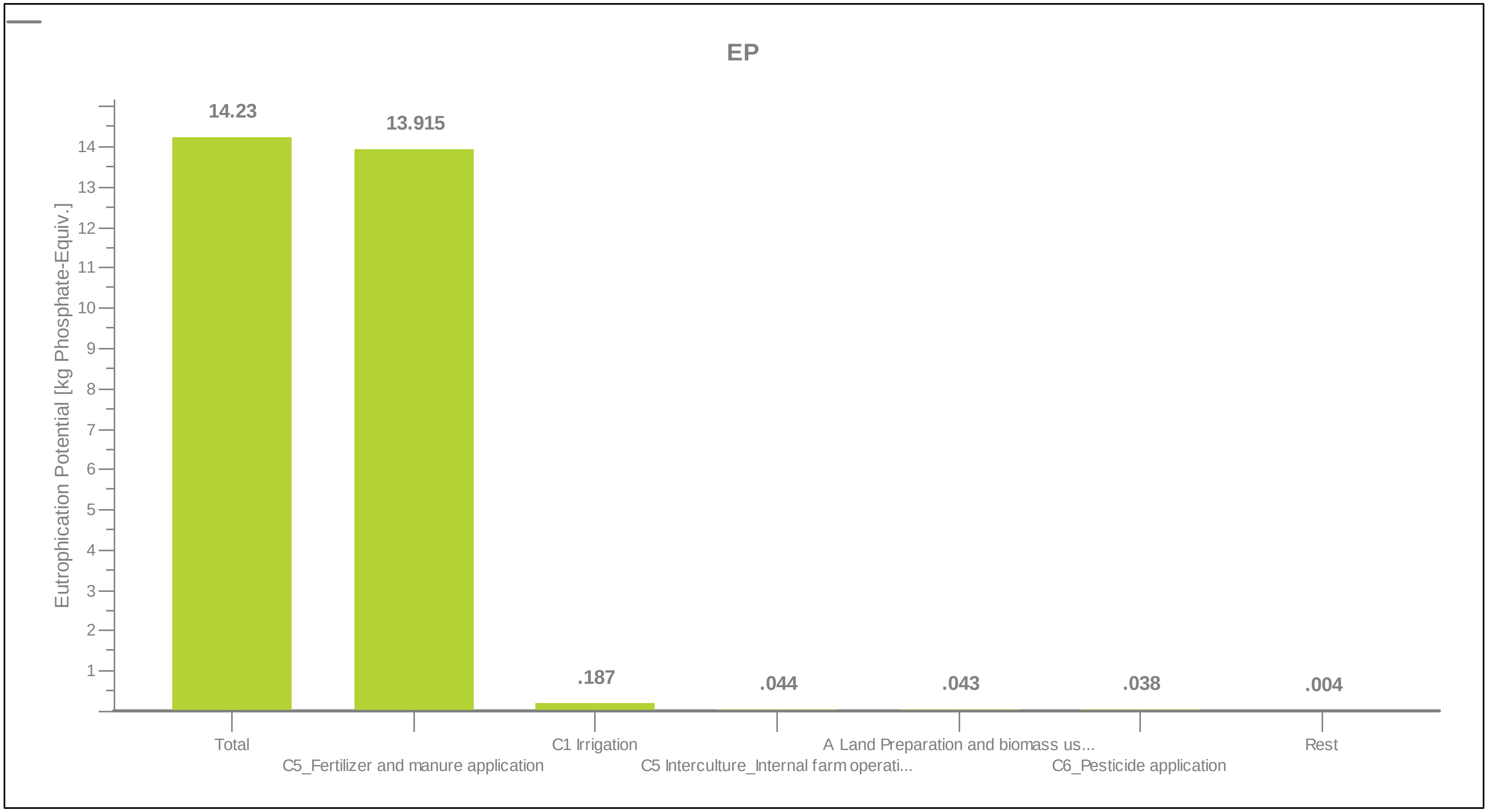


Figure 62c. Life cycle impact of cultivation of *Jatropha* in 1 ha area (1 year): EP (Eutrophication potential).

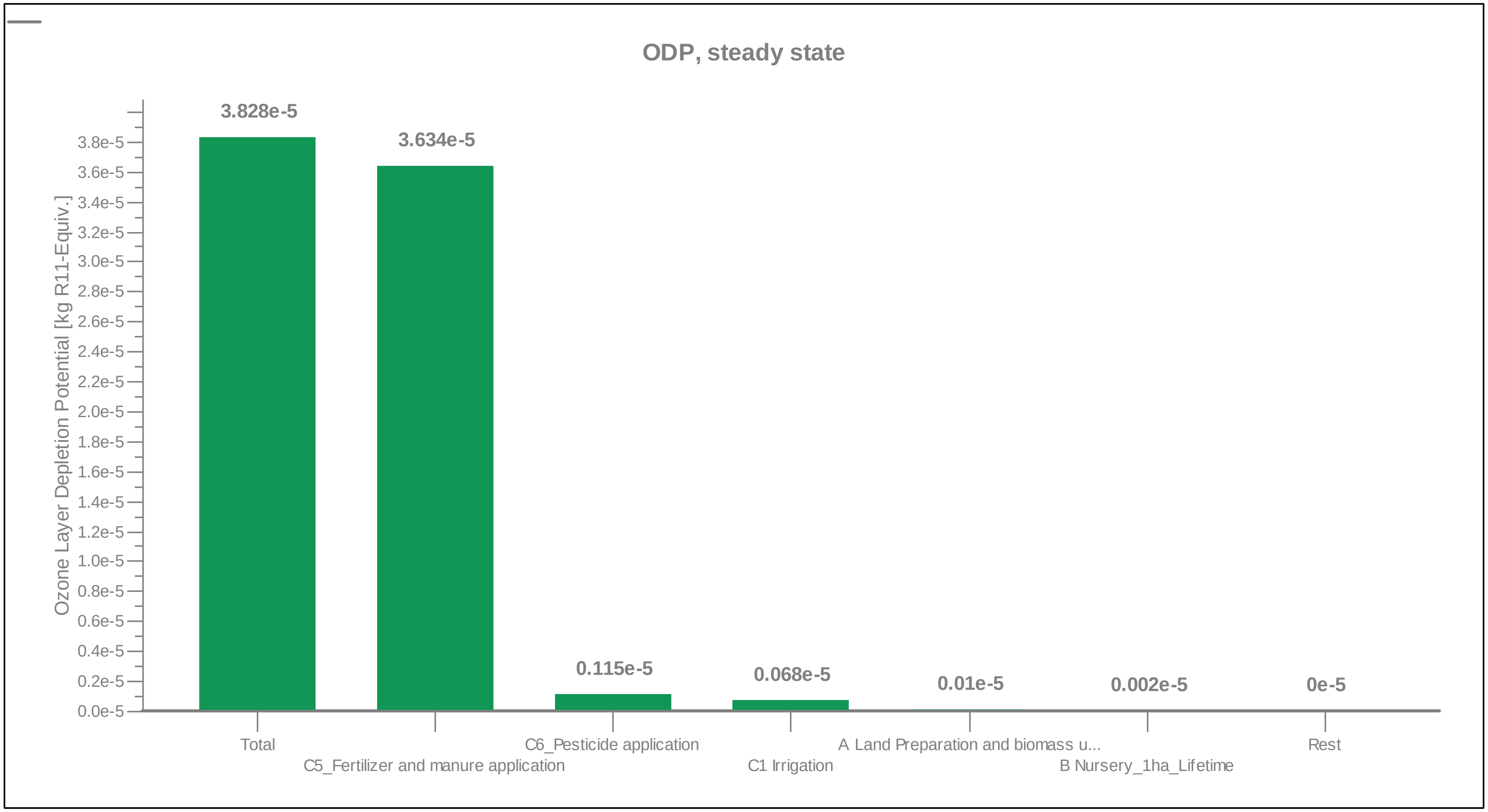


Figure 62d. Life cycle impact of cultivation of *Jatropha* in 1 ha area (1 year): ODP (Ozone depletion potential).

If one considers the life cycle assessment for the cultivation process alone, then fertilization and manuring accounts for 76.4% of the total GWP **(Figure C8)**. The percentage of contributions from other processes to the total GWP from 1 ha *Jatropha* plantation were as follows: irrigation (11.8%), land preparation (7.5%), weeding and farm operations (3.5%). Similarly, fertilization and irrigation also accounted for the major portion of impacts resulting in acidification potential, eutrophication potential and ozone depleting potential **(Figure 62b, c, d).** Thus, it is quite clearly evident that fertilizer production and its application in the field resulted in major impacts in all the environmental categories that were measured.

Considering fertilization process alone, one can quite clearly see that the major source of GWP impact lies in the production (in this case urea) and use of nitrogen fertilizer (including organic manure) contributing nearly 88.3% while phosphorus fertilization accounted for 10.3% of the total GWP due to overall fertilizer and manure application. Potassium fertilizers have a very low GWP (1.4%) even after considering their trans-oceanic transport **(Figure C5)**. A similar trend was observed in the other impact category of eutrophication potential and ozone depletion potential, wherein, the use of N fertilization and phosphorus fertilization resulted in 55.32%, 40.29% and 76.4%, 19.9% of the total impacts, respectively, within these categories **(Figures A7 c, d in appendix)**. The major contribution to this impact category in nitrogen fertilization was primarily due to the production of urea. Production of phosphorus and nitrogen fertilizers contributed to 60.68% and 36.79% in the impact category of acidification potential.

The nursery accounted for only 0.3% of the total GWP calculated for the entire life cycle analysis for *Jatropha* cultivation. This was primarily on account of innovative nursery raising in sand beds as against the conventional use of polybags which significantly increases GHG emissions. Within the nursery process, composting accounted for more than 95% of GWP and was also the significant contributor for the other impact categories **(Figures B2)**. Pumping of water, PVC production and extrusion into pipes contributed to the major chunk of GHG emissions in the irrigation module as well as in other impact categories **(Figures A5a in appendix)**.

Pesticide application contributed to 0.5% of the total GWP for the entire life cycle analysis.

ii) Aggregate LCA model for production of oil, briquette and cake from 1 ha (1 year).

Since this model is an extension of the plantation model which additionally includes modules for downstream processing of fruits into seed oil, *Jatropha* de-oiled cake, steam generation from a portion of *Jatropha* shells used for softening of seeds during expelling of oil through a screw type oil expeller and briquetting of remaining shells, all the assumptions and input values used for plantation model in the previous section as given in **Table 15 and 16** remain valid for this module as well. The carbon sequestered by fruits or its bio-energy derivatives, i.e., shell, oil or cake has not been accounted for. The overall assumption and additional input values are presented in **Table 17 and 18.**

Table 17. GaBi aggregate module for *Jatropha* oil production:

Process name	Module purpose	Comments
Jatropha plantation	This custom module aggregates and calculates the impacts of all the sub-processes involved in the cultivation of *Jatropha curcas* in one hectare for one year	
Land preparation	This custom module aggregates and calculates the impacts of all sub-processes involved in land use change for one hectare apportioned to one year	
Nursery	This custom module aggregates and calculates the impacts of all the sub-processes involved in raising of cuttings in nursery required for one hectare	
Farm operations	This custom module aggregates and calculates the impacts of emissions resulting from weeding, transport of fertilizers and other miscellaneous items into and within the field for one hectare over a period of one year	
Irrigation	This custom module aggregates and calculates the impacts of all the sub-processes involved in irrigating one hectare of the plantation using a drip system for a period of one year	
Fertilization	This custom module aggregates and calculates the impacts of all fertilizer production and their use in one hectare of the plantation for a period of one year	
Pesticide application	This custom module aggregates and calculates the impacts of pesticide application and their use in one hectare of the plantation for a period of one year	
Decorticating/Deshelling	This custom module calculates the impacts during the deshelling of ca.3 tons of fruits	
Steam generation	This custom module aggregates and calculates the impacts during generation of 480 kg of steam	*Jatropha* shells are used as source of energy for heating
Briquette production	This module calculates the impacts during 903kg of briquette production	
Oil expulsion	This custom module calculates the impacts during expulsion of 510 kg *Jatropha* oil as well as 1530 kg of deoiled seed cake from 2040 kg of seeds	The yield of 2040 kg from a mature plantation is an assumption derived from extrapolation of the present yield

Table 18. Inputs of aggregate plan for *Jatropha* oil production

Parameter	Value	Unit	Documentation	Source
All inputs of Table 34 and in addition the following				
Steam	490	Kg	Amount of steam that can be produced shells obtained from one hectare of plantation	Own data
Oil content	0.25		Assumption of 25% extractable oil from seeds	Conservative data; can achieve even higher as obtained in our case
Jatropha seeds	2040	Kg	Amount of seeds that is produced from one hectare of *Jatropha* plantation	Based on own data (66.7% of fruit)
Electricity	1227.75714	MJ	Total amount of electricity consumed during deshelling, steam generation, briquette production and Oil expulsion processes	Calculated data
Jatropha Oil	510	Kg	Amount of *Jatropha* oil produced from ca. 2 ton seeds	Assumption of 25% extractable oil
Jatropha deoiled cake	1530	Kg	Amount of Cake produced as a by-product during oil expulsion	Estimated
Water	612	/kg	Softened water required for 490 Kg steam generation	Own data

The life cycle impact across all the categories pertaining to overall land preparation is given in **Figure 63a-d.**

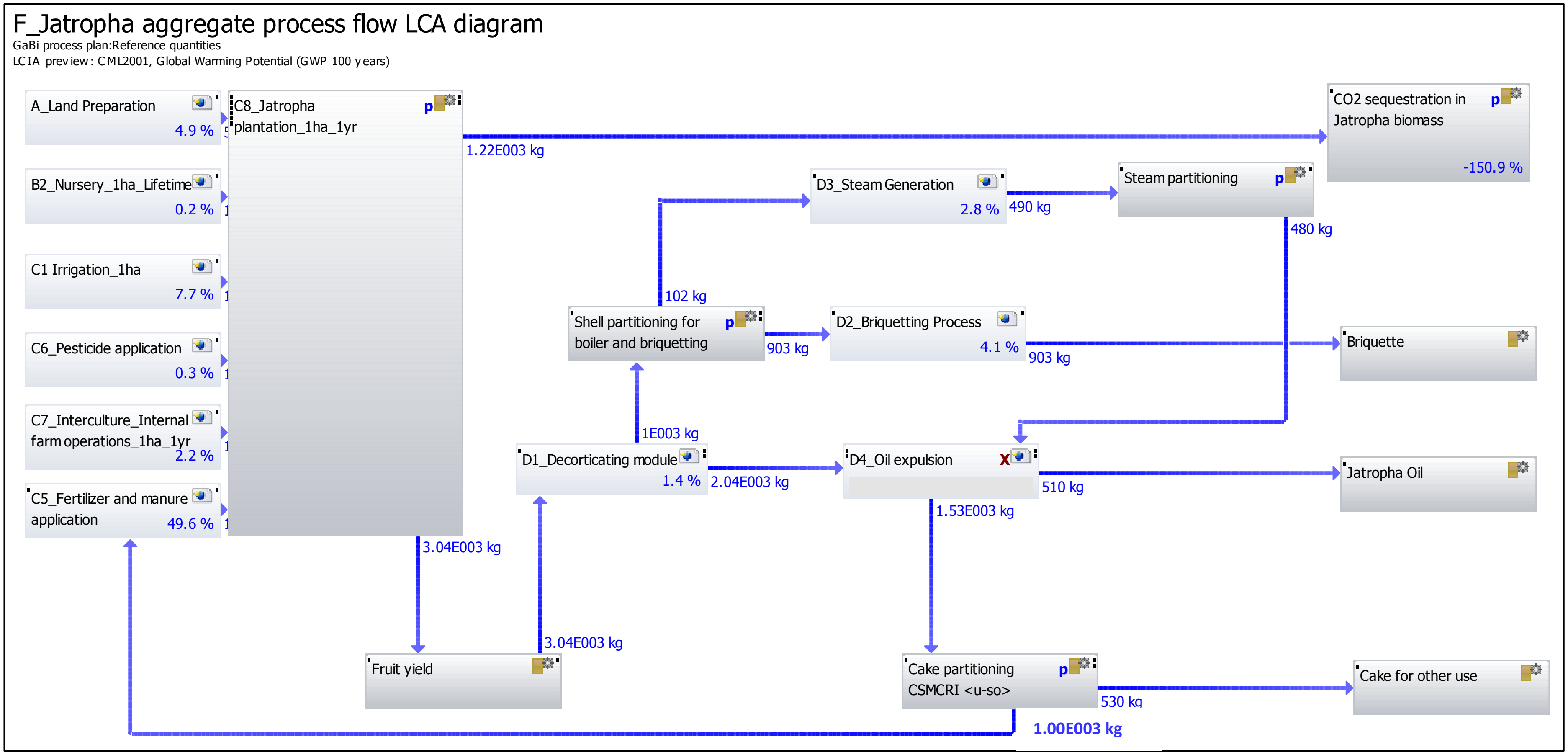


Figure F. Life cycle modeling and life cycle impact assessment preview for cultivation of *Jatropha* and generation of co-products (*Jatropha* oil, *Jatropha* shell briquettes, *Jatropha* cake) from 1 ha area (1 year).

NB: Figures in % represent GWP 100 years (Global warming potential) based on CML 2001

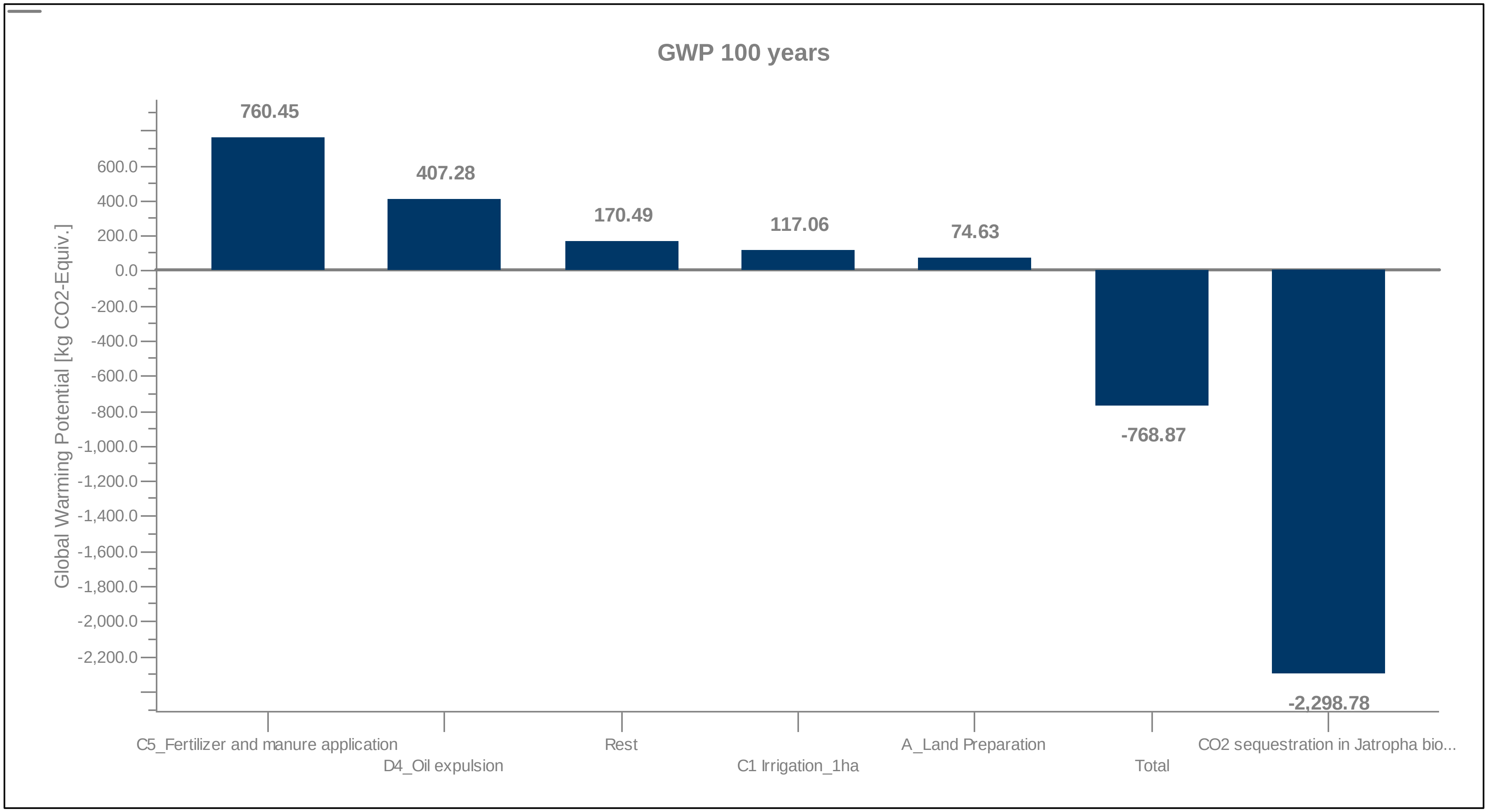


Figure 63a. Life cycle impact of cultivation of *Jatropha* and generation of co-products (*Jatropha* oil, *Jatropha* shell briquettes, *Jatropha* cake) from 1 ha area (1 year): GWP (Global warming potential).

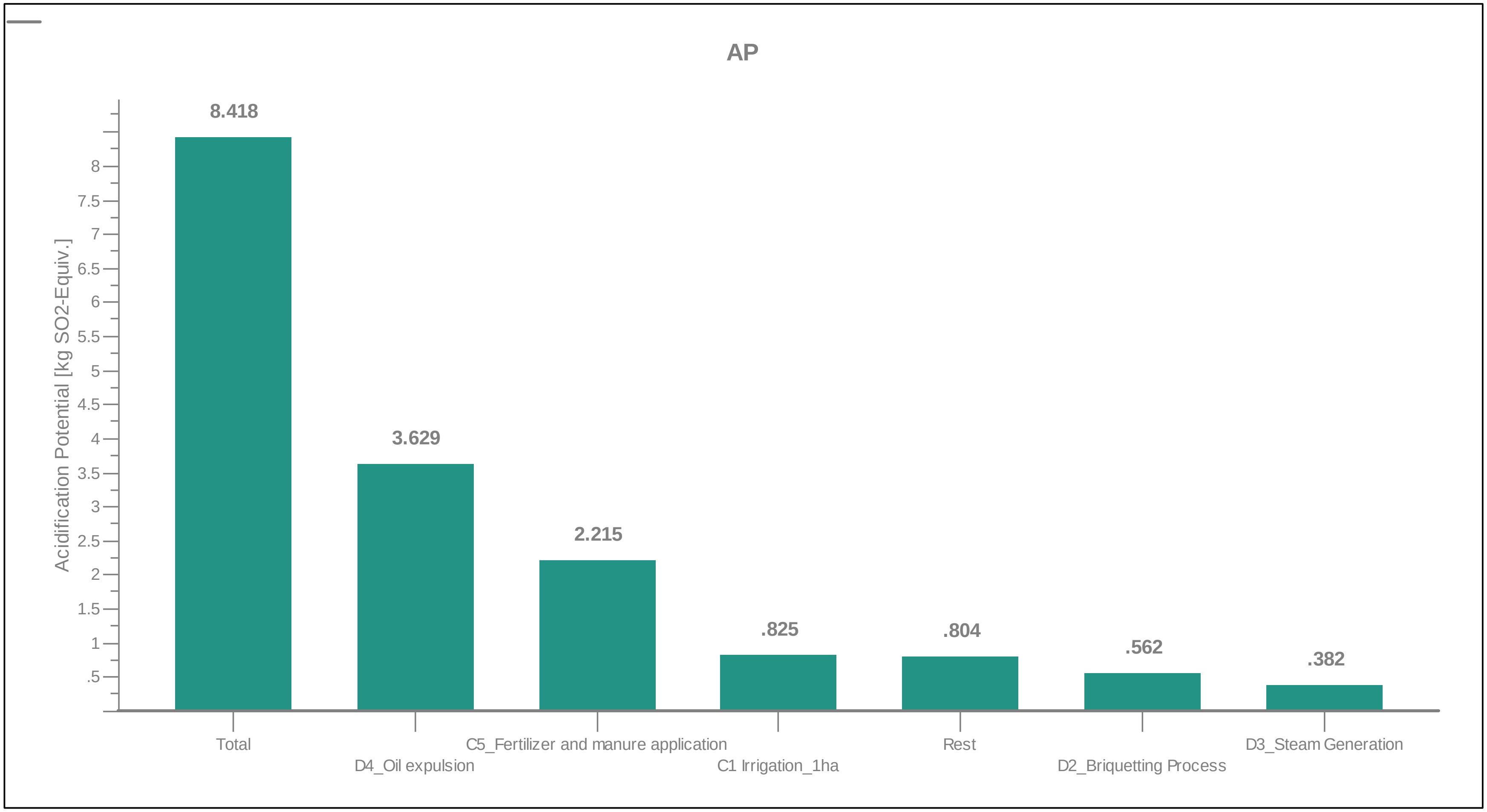


Figure 63b. Life cycle impact of cultivation of *Jatropha* and generation of co-products (*Jatropha* oil, *Jatropha* shell briquettes, *Jatropha* cake) from 1 ha area (1 year): AP (Acidification potential).

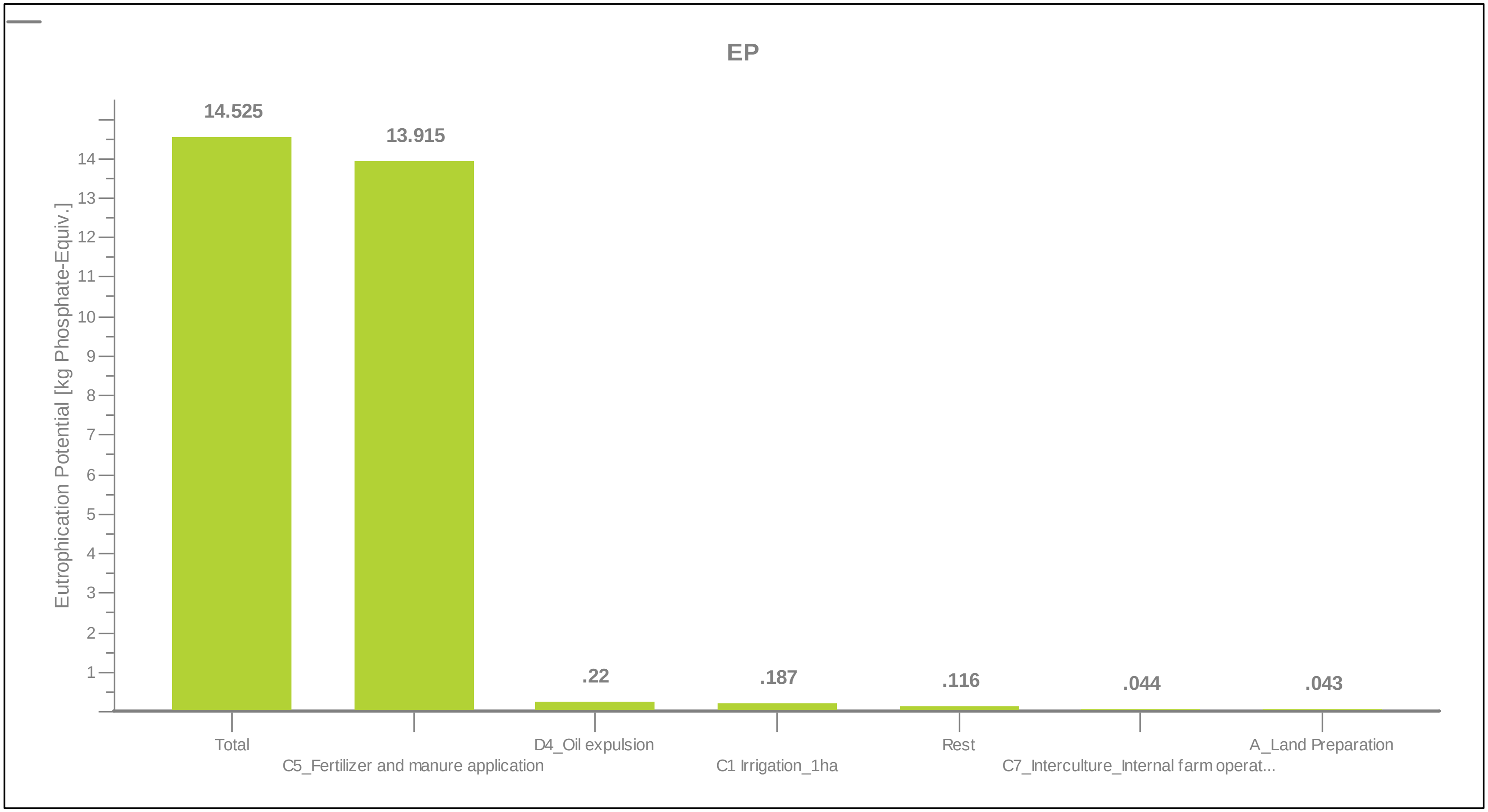


Figure 63c. Life cycle impact of cultivation of *Jatropha* and generation of co-products (*Jatropha* oil, *Jatropha* shell briquettes, *Jatropha* cake) from 1 ha area (1 year): EP (Eutrophication potential).

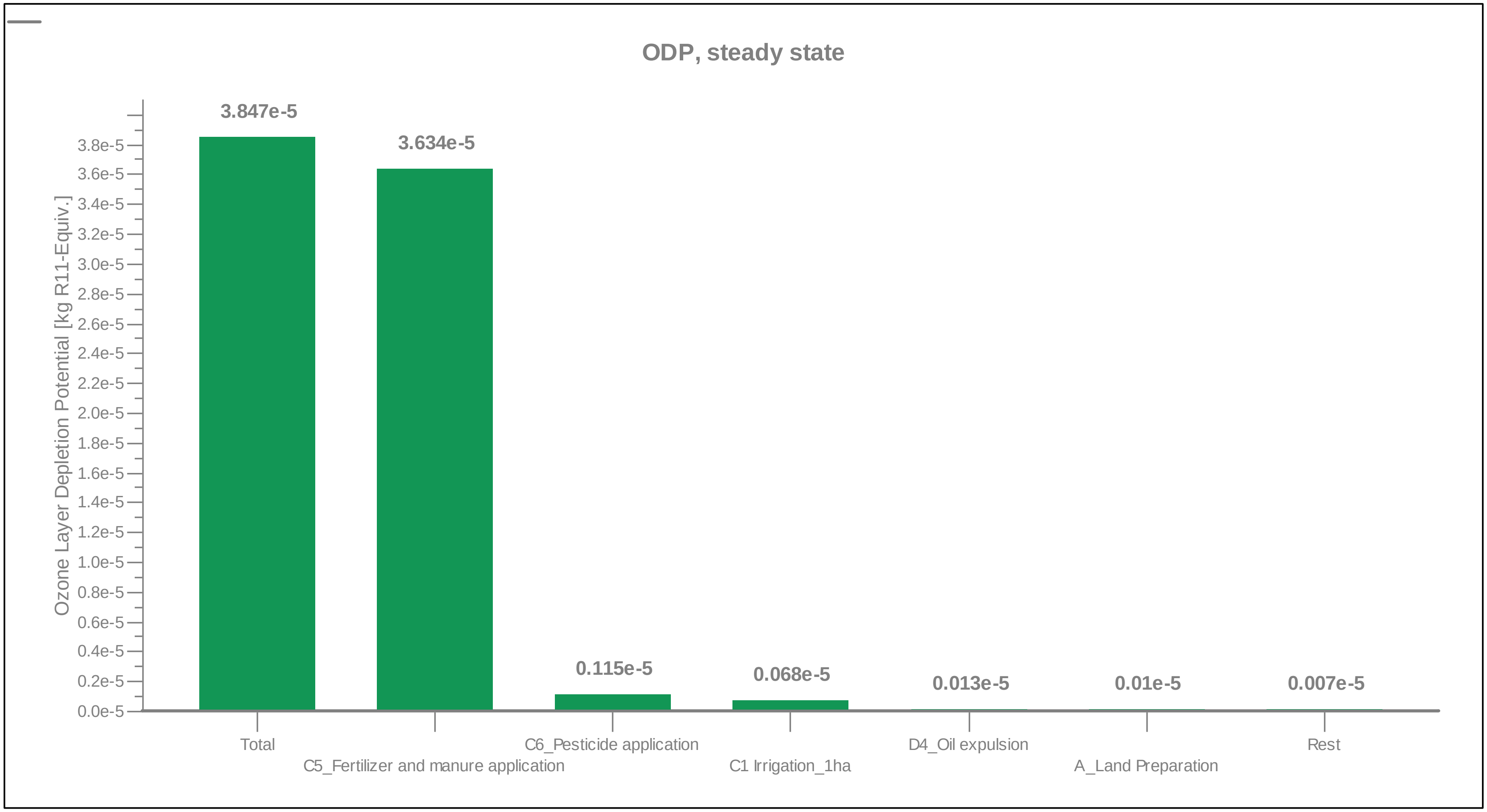


Figure 63d. Life cycle impact of cultivation of *Jatropha* and generation of co-products (*Jatropha* oil, *Jatropha* shell briquettes, *Jatropha* cake) from 1 ha area (1 year): ODP (Ozone depletion potential).

The overall Life Cycle Impact Assessment for the production of *Jatropha* oil, cake and briquette from 1 ha (1 year) reveals that the upstream processes involved in the production of raw materials (fruit) account for 64.9% of GWP, while the downstream processes involving the de-shelling, extraction of oil and production of briquettes account for 35% of GWP. The overall impact towards GWP is negative (-768.87 Kg CO_2 eq) when CO_2 sequestered in biomass of the plant (average 2.27 t CO_2/ha/yr based on 1.215 tons average biomass built up/ha/yr) is considered **(Figure F)**. This reflects a favorable impact of Jatropha cultivation on wastelands in reducing Greenhouse Warming Potential even after investing initially on extensive land amendment as a result of which the carbon lost by removing the native shrubs is more than compensated by the luxuriant growth of *Jatropha* enabling greater carbon sequestration in standing biomass.

Among the upstream processes, one can clearly see that fertilizer production and its application in the field accounts for 49.6% of the total GWP, thereby serving as one of the major contributors of GWP of the entire life cycle up to oil, cake and briquette production **(Figure F)**. Fertilization and the manuring process accounted for 95.8% (13.915 Kg Phosphate eq) impact in Eutrophication Potential and 26.31% impact in Acidification Potential (Kg SO_2 eq) and 94.46% impact in Ozone Depleting Potential **(Figure 63b, c, d)**.

Figure F shows that irrigation accounts for 7.7% (117.06 Kg CO_2 eq) which is mainly due to the PVC materials (38.9% of total irrigation GWP) employed for drip irrigation and the emissions to run the pumps (48.5% of total irrigation GWP), which has been detailed in **Figure C1**. However, there is a scope for further reduction in greenhouse gas emissions if one considers irrigation using other techniques. In the land preparation module, burning of the biomass resulted in major contributions to all the impact categories evaluated, followed by combustion of diesel in the excavator.

Land preparation which involved environmental engineering i.e., trenching by excavator and burning of the cleared biomass, however, accounted for only 4.9% of the entire life cycle GWP. Within this also trenching contributed only 16% to overall land preparation process. This engineering of the root zone in soil is one of the main determinants towards the growing of *Jatropha* under shallow and rocky wastelands and may result in the uprooting of well grown-up plantations by wind as we had experienced **(Figure 14)** if not properly taken care of. Hence the model of cultivation involving deep trenching of the soil for better establishment of the plantation is expected to be an important investment whose outcome in the future **(Figure 64)** would more than offset the modest carbon input made.

Figure 64. Shallow and rocky wastelands amended for cultivation of *Jatropha.*

Among the upstream processes, oil expulsion accounted for a GWP of 26.7% (407.28 Kg CO_2 eq) and was the second major contributor of GWP **(Figure 63a)**. The oil expulsion process accounted for 1.5% and 43.11% impact in Eutrophication and Acidification Potential, respectively **(Figure 63b, c)**. The use of electricity constituted the primary source of most of the environmental impacts in the downstream processes such as oil expulsion, briquetting, decortication and steam generation accounting for more than 97% of overall impacts.

Inference

We emphasized harvesting dry fruits and maximizing the value addition at the farmer's end, including the use of *Jatropha* cake in the field **(Figure 65)**. It may be highlighted that two fuels are obtained from *Jatropha*, the first being liquid biodiesel on conversion from oil and second, the solid fuel in the form of briquettes. For dry fruit cost scenarios of Rs. 8,000 per ton and 11,200 per ton, and considering that all by-products are marketed, B100 Jatropha biodiesel production cost works out to Rs. 45 per liter and Rs. 65 per liter, respectively, assuming commercial scale production. Considering that two thirds of the fruit weight is seed weight, these cost scenarios translate to a seed price of Rs. 12,000 per ton and Rs. 17,000 per ton, respectively.

Figure 65. *Jatropha* value chain.

NB: The integrated process is covered by US patent *No. 7,666,234* granted in 2010. A still more comprehensive integration, with co-production of biodegradable plastic, is covered through a second international patent application (WO/2011/027353) which has been cleared for patentability.

Though LCA up to the production of biodiesel was not attempted due to the shortened duration of the project, it is expected that the overall impact would still remain favorable because of very novel process developed (US Patent No. 7,666,234) by the Institute) with a net energy balance calculated ca. 5.57. Nevertheless, the project vehicle (Chevrolet Tavera) successfully completed plying 100,000 km using neat *Jatropha* biodiesel during the project period **(Figure 66)**.

Figure 66. Dr. Candace Wheeler (PI from GM) pouring neat *Jatropha* biodiesel in GM Tavera car (Background: *Jatropha* plantation on wastelands of Bhadrawal, Gujarat).

This Tavera vehicle provided for the project achieved an additional feat, when this vehicle was flagged off by former Honorable Minister of Science and Technology, Government of India, Late Shri Vilas Rao Deshmukh for a first test-run in India using B-100 biodiesel from marine micro-algae, produced by the Institute **(Figure 67)**.

Figure 67. Hon. Late Shri Vilasrao Deshmukh filling B100 microalgal bio-diesel at CSIR headquarters in New Delhi on 30 March 2012 before flagging off the vehicle on a 200 km test run using General Motors Chevrolet Tavera car.

At the end of the project, the Institute has generated over 400,000 elite true-to-type cuttings which can, at present, be given to interested stakeholders for raising productive plantations. Innovative ways of making shallow wastelands and water-logged lands amenable to Jatropha cultivation was demonstrated on a large scale, while effective water harvesting and efficient water use techniques were developed. The use of Jatropha cake as an organic manure not only was able to offset mineral fertilizer requirements but also improved the physiological and bio-chemical parameters in Jatropha, simultaneously raising the yields of the plants. Jatropha cake reduced the load of pathogenic fungus in soil. Further, the major output from this collaborative

Jatropha project was the development of a sustainable strategy for cultivation of Jatropha on wastelands, thereby achieving an attractive carbon balance computed up to production of oil and briquette from whole dry fruits. It provided good impetus to re-affirm that *Jatropha* has enormous potential to turn barren wastelands green and productive and at the same time also produce feedstock for bio-energy in a carbon-efficient manner. We emphasize the need for further research to boost *Jatropha* productivity by finding practical solutions to overcome challenges that have been identified, primarily among which are: making available in large numbers true-to-type planting material of proven accession for productive large scale plantations (targeting a few million hectares), making provision of water for irrigation in these parched lands while improving water use efficiency, imparting tolerance to diseases and abiotic stresses.

In a very recent development, CSIR-CSMCRI's Jatropha biodiesel has been cleared for field trials in their locomotives by the Indian Railways **(Figure 68)**.

Figure 68.

THE ECONOMIC TIMES

By PTI | 9 Sep, 2013, 05.00PM IST

Railways clear jatropha bio-diesel for field trials

AHMEDABAD: Research Design Standard Organisation (RDSO), the research wing of Indian Railways, has cleared Jatropha bio-diesel developed by Central Salt Marine & Chemical Research Institute for field trials in locomotives.

"RDSO has cleared Jatropha bio-diesel of CSMCRI for field trials in locomotives. A few more approvals are required and possibly in next 3-4 months we shall commence the trials on two locomotives in Jetalsar," an official said.

"RDSO has cleared Jatropha bio-diesel of CSMCRI for field trials in locomotives. A few more approvals are required and possibly in next 3-4 months we shall commence the trials on two locomotives in Jetalsar," a Western Railway official said.

"During initial trials it would be 10 per cent bio-diesel and 90 per cent conventional diesel, but as we progress the bio-diesel content will be scaled up," he said.

Railways use diesel-run locomotives on several routes where electric lines are yet to be laid. Jetalsar (Rajkot district) to Dhasa (Amreli district) is one such line.

"Laboratory tests have been conducted to test locomotives on B-100 bio-diesel (neat bio-diesel), but there are some issues with it. Hopefully once resolved, we aim to run the locomotives on hundred per cent bio-diesel," the official said.

CSMCRI Director Dr Pushpito Ghosh said, "Yes, our bio-diesel has been cleared for field trials by the RDSO.

"Jatropha biodiesel can be produced for between Rs 45-65 per litre assuming all-inclusive dry fruit cost of Rs 8,000 per tonne," an institute official said.

Earlier, the institute's US patented technology to extract bio-diesel from Jatropha was successfully adopted by Vehicle Research Development Establishment, a defence laboratory in Ahmednagar to run a few vehicles on this environment-friendly fuel.

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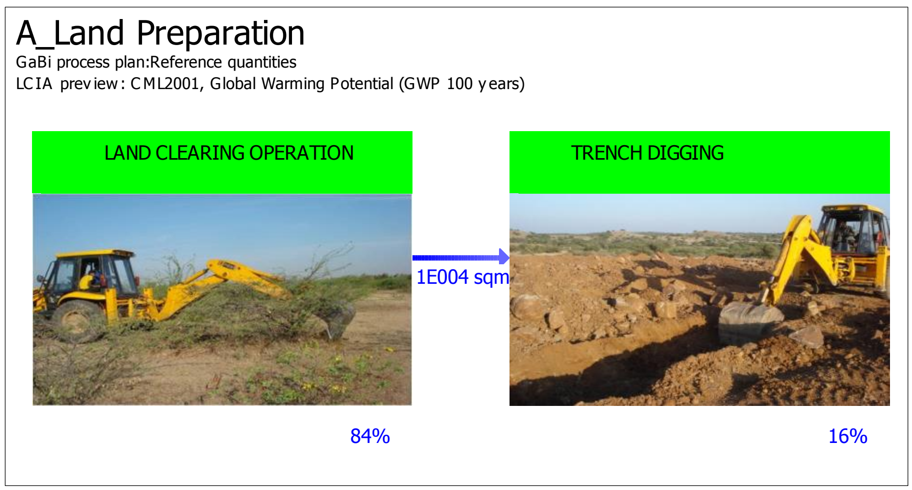
APPENDIX-1

Individual LCA modules, Data Input and Their Life Cycle Impact Assessments

Land preparation

The preliminary step in developing plantation in wasteland involves land preparation. As described in the earlier sections a new methodology was adopted for growing *J. curcas* in rocky wastelands. This module describes the land use change and its impacts and has two sub processes viz.

A. Land Clearing, biomass burning and
B. Trenching



Land clearing and biomass burning module incorporates the impacts of various emissions that are caused by removal of existing plants and biomass thereof by excavator. The amount of diesel that would be required for land clearing was exactly determined in addition to other inputs such as lubricating oil which is required by the machine in its hydraulics. The module described here proposes the burning of biomass in order to obtain energy which can be used for various purposes which is an approximation to the use of logs by local as fuels. An Eco-invent module describing the impacts of burning of logs is used as an approximation for calculating the energy that can be obtained. Combustion of diesel in excavator was based on the already existing process of excavation by hydraulic digger, but the diesel used was measured actually while excavating in the field. It can be seen that for GHG potential, trenching constitutes only 16%, while the bulk is attributed to land clearing operation in which also 93.4% is attributed to combustion of the removed biomass for energy **(Figure A and A1).** The assumptions and input values used for modeling land clearing are given in **Table A1 and A2**.

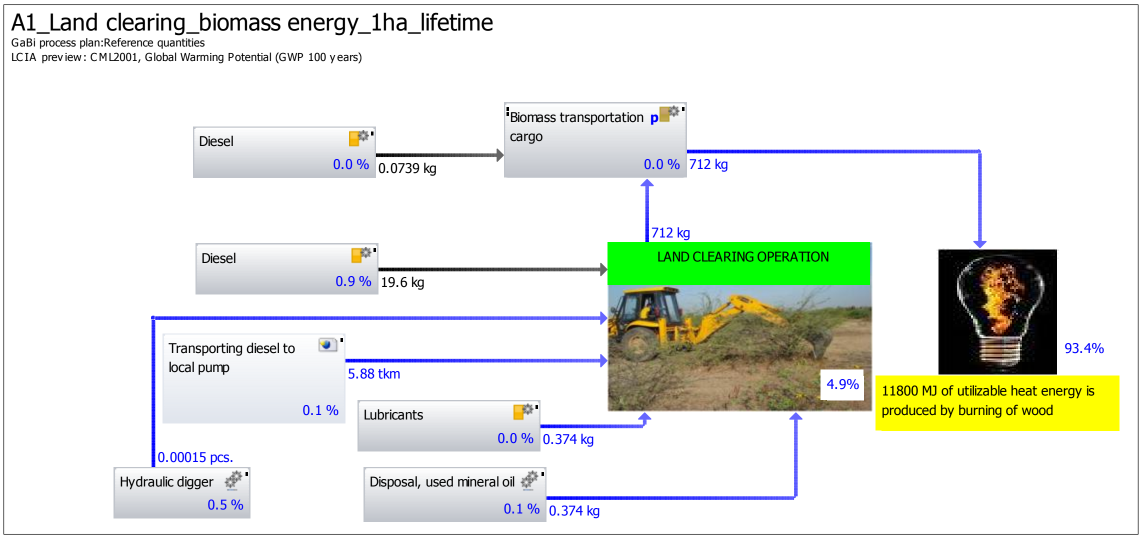


Table A1. GaBi module description for Land Clearing and Biomass energy.

Process name	Module purpose	Comments
Trenching by excavator	A custom module used to calculate the impacts of all the sub processes along with emissions from combustion of diesel	Emissions for combustion of diesel were calculated from Eco-invent 2.2 module of hydraulic digger.
Diesel	PE professional data set calculates the impacts of upstream crude oil production its transport and refining of diesel at Indian refinery	
Transport	This modified Eco-invent 2.2 process calculates the impacts of transport of diesel from refinery at Koyali near Vadodara to local gas station	The Eco-invent module of transport using 20-28 t fleet average was used for the purpose of determining the impact of emissions. This customized module uses Indian data set for diesel. This Eco-invent process is used as a proxy as specific data set for Indian conditions is not available
Hydraulic digger	This Eco-invent 2.2 process was used as a proxy for excavator and calculates the impacts during its production	
Lubricants	PE professional data set calculates impacts for lubricant production at Indian refinery	
Disposal of mineral oil	This Eco-invent 2.2 process calculates the impacts during its disposal of lubricating oil	This Eco-invent process is used as a proxy as specific data set for Indian conditions is not available
Cargo transport	PE professional data set calculates impacts of diesel combustion during transport of cleared biomass from the field to the site of utilization	The sulphur content in diesel was assumed to be 10 ppm which is standard set in Europe
Thermal energy	This modified Eco-invent 2.2 process calculates impacts of burning of biomass obtained from land clearing	Though native biomass after clearing of shrubs was not burnt, and instead left in field itself, it has been assumed that the biomass generated would be utilized as fuels by native people and impacts of such a burning was included.

Table A2. Inputs for Land clearing and biomass burning module

Parameter	Value	Unit	Documentation	Source
Diesel	19.6	Kg	Amount of diesel used in excavator/hydraulic digger	Own data (measured)
No. of pieces of excavator	0.000149725	No. of pieces	Assumption based on total life of the machine	
Transport	5.88	tkm	Transport of diesel from Koyali refinery to region storage for distance of 300 km	Estimated data
Lubricating oil	0.374	Kg	The amount of lubricating oil that would employed in hydraulics of the excavator	
Disposal of mineral oil	0.374	Kg	The amount of lubricating oil that is subjected to disposal	
Diesel	0.0739	Kg	Amount of diesel used for transporting cleared biomass (715kg) over a distance of 5 km (assumption) from plantation for burning in order to obtain energy	Own data (Data of Neswad site used here)

Trenching Module

In this module, we describe excavating soil by trenching to a depth of 70-80 cm and width of 75 cm by hydraulic excavator **(Figure A2)**. Diesel constituted the major input and it required ca. 5 hours for making trenches for one hectare of the field by excavator. The transport module describes the transport of diesel from refinery to local storage station. As can be seen, 79.9% of GWP (CML 2001) is being attributed to emission due to operation of excavator. The assumptions and input values used for modeling of trenching module are given in **Table A3 and A4**.

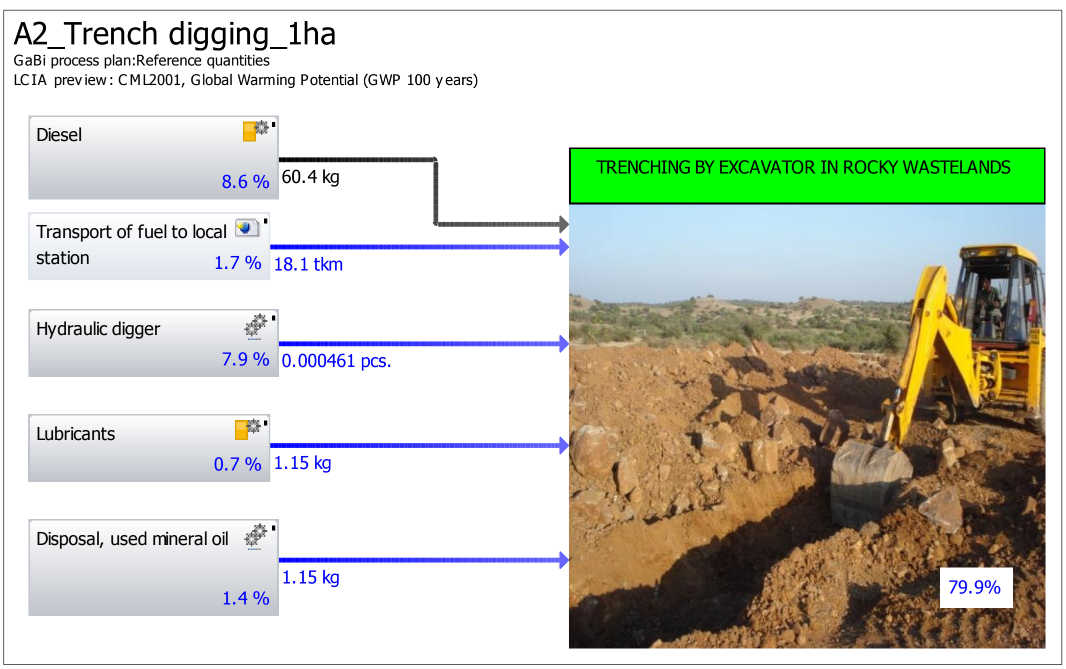


Table A3. GaBi module describing trenching

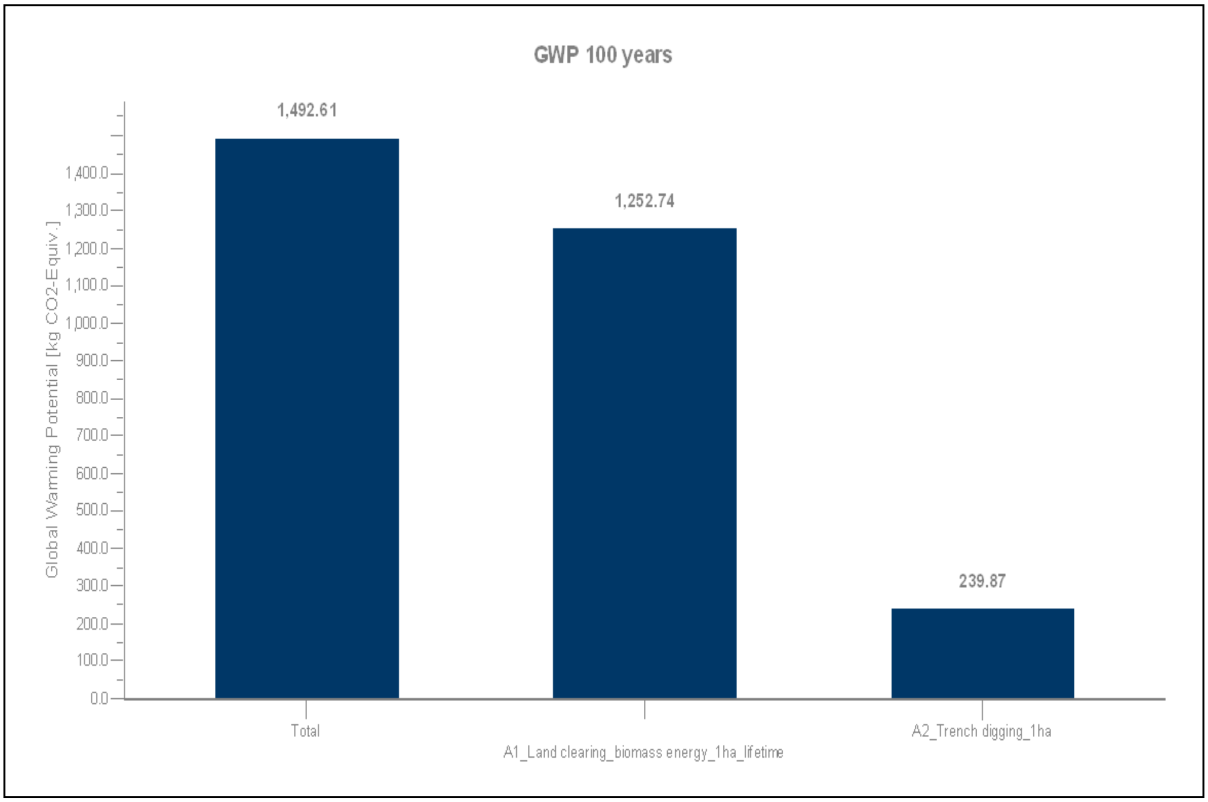
Process name	Module purpose	Comments
Trenching by excavator	A custom module used to calculate the impacts of all the sub processes along with emissions from combustion of diesel	Emissions for combustion of diesel were calculated from Eco-invent 2.2 module of hydraulic digger.
Diesel	PE professional data set calculates the impacts of upstream crude oil production its transport and refining of diesel at Indian refinery	
Transport	This modified Eco-invent 2.2 process calculates the impacts of transport of diesel from refinery at Koyali near Vadodara to local gas station	The Eco-invent module of transport using 20-28 t fleet average was used for the purpose of determining the impact of emissions. This customized module uses Indian data set for diesel. This Eco-invent process is used as a proxy as specific data set for Indian conditions is not available
Hydraulic digger	This Eco-invent 2.2 process was used as a proxy for excavator and calculates the impacts during its production	The number of pieces of the machinery in the database was customized by parameterizing diesel requirement with life of the equipment
Lubricants	PE professional data set calculates impacts for lubricant production at Indian refinery	
Disposal of mineral oil	This Eco-invent 2.2 process calculates the impacts during its disposal of lubricating oil	This European process is used as a proxy to Indian condition

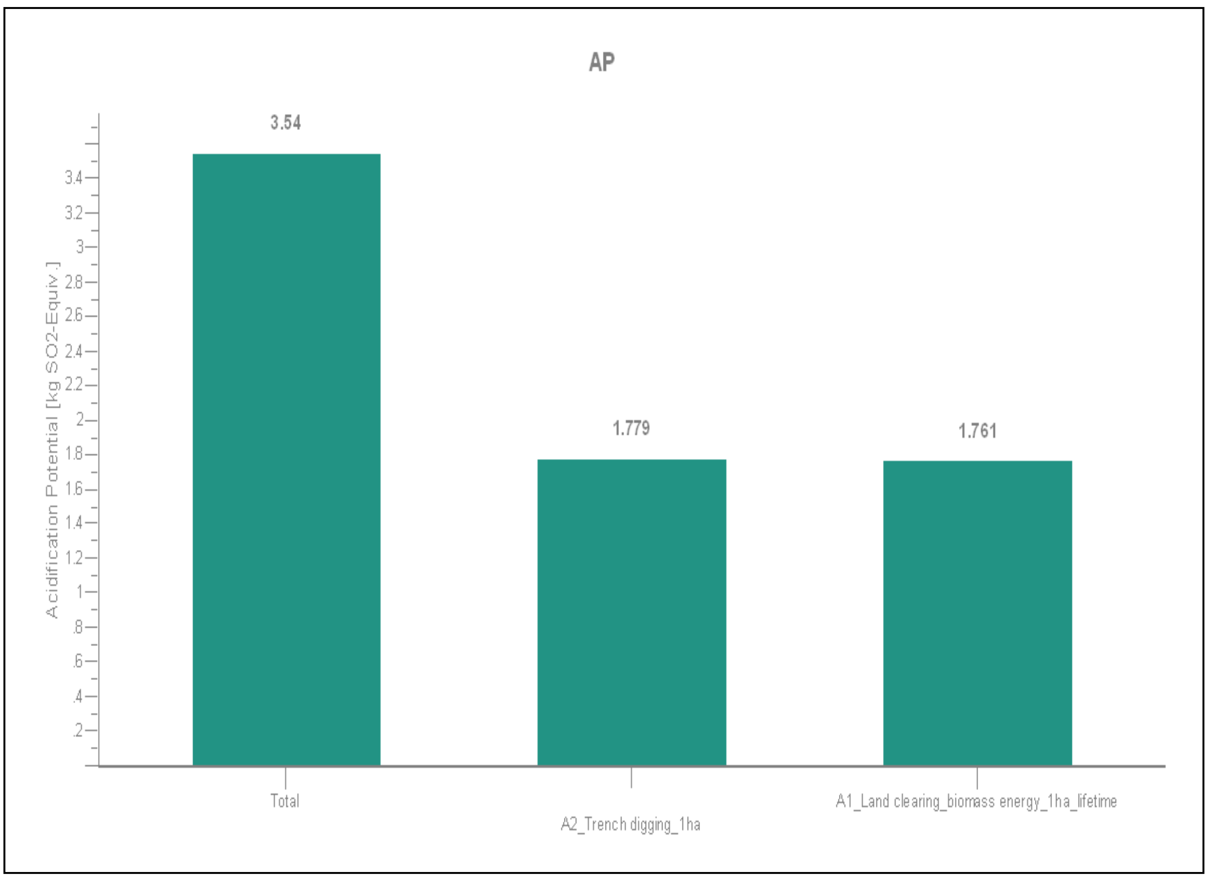
Table A4. Inputs for Trenching module

Parameter	Value	Unit	Documentation	Source
Diesel	60.4	Kg	For use in excavator/hydraulic digger	Own data (measured)
No. of pieces of excavator	0.000461153	No. of pieces	Assumption based on total life of the machine	Eco-invent database
Transport	18.1	tkm	Transport of diesel from Koyali refinery to region storage for distance of 300 km	Estimated
Lubricating oil	1.15	Kg	The amount of lubricating oil that would employed in hydraulics of the excavator	Eco-invent database
Disposal of mineral oil	1.15	Kg	Amount of lubricating oil that is subjected to disposal	Eco-invent database

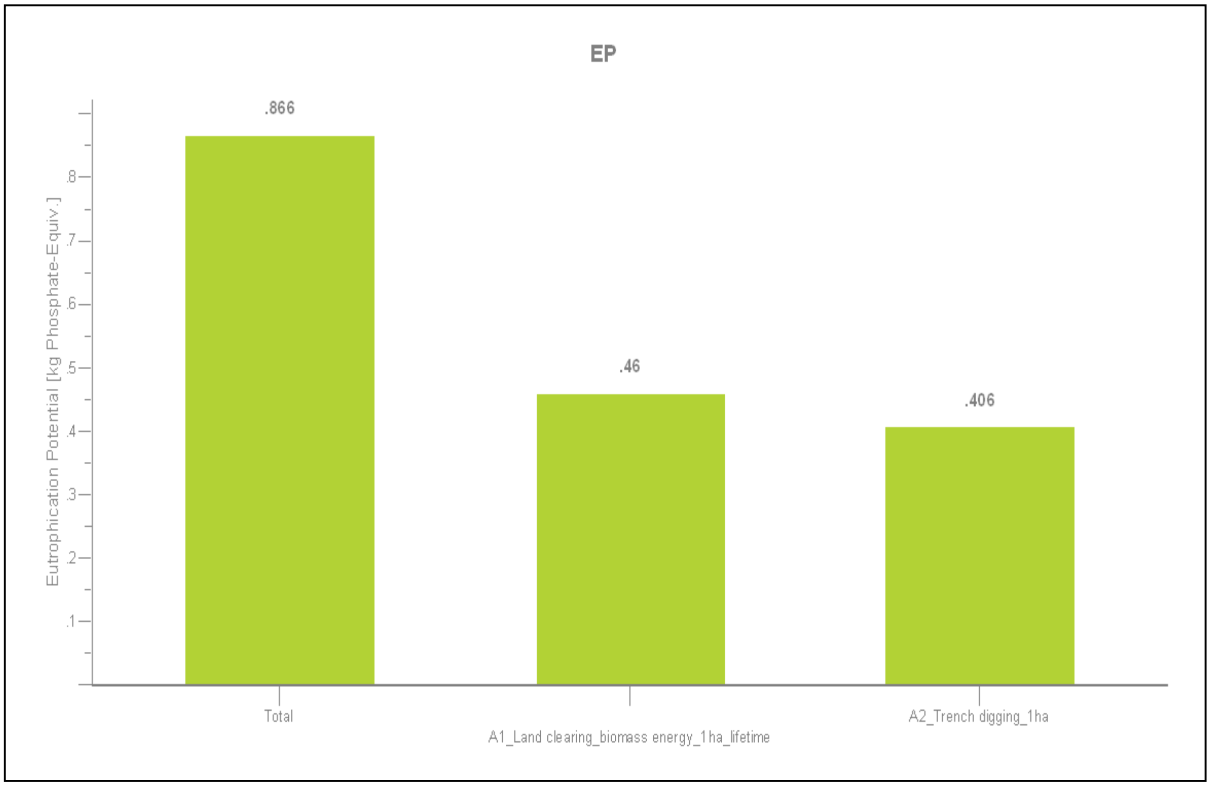
The life cycle impact across all the categories pertaining to overall land preparation is given in **Figure A3a-d.**

b


GWP 100 years
Global Warming Potential [kg CO2-Equiv.]
1,492.61
1,252.74
239.87
1,400.0
1,300.0
1,200.0
1,100.0
1,000.0
900.0
800.0
700.0
600.0
500.0
400.0
300.0
200.0
100.0
0.0
Total
A1_Land clearing_biomass energy_1ha_lifetime
A2_Trench digging_1ha


AP
Acidification Potential [kg SO2-Equiv.]
3.54
1.779
1.761
3.4
3.2
3
2.8
2.6
2.4
2.2
2
1.8
1.6
1.4
1.2
1
.8
.6
.4
.2
Total
A2_Trench digging_1ha
A1_Land clearing_biomass energy_1ha_lifetime

d



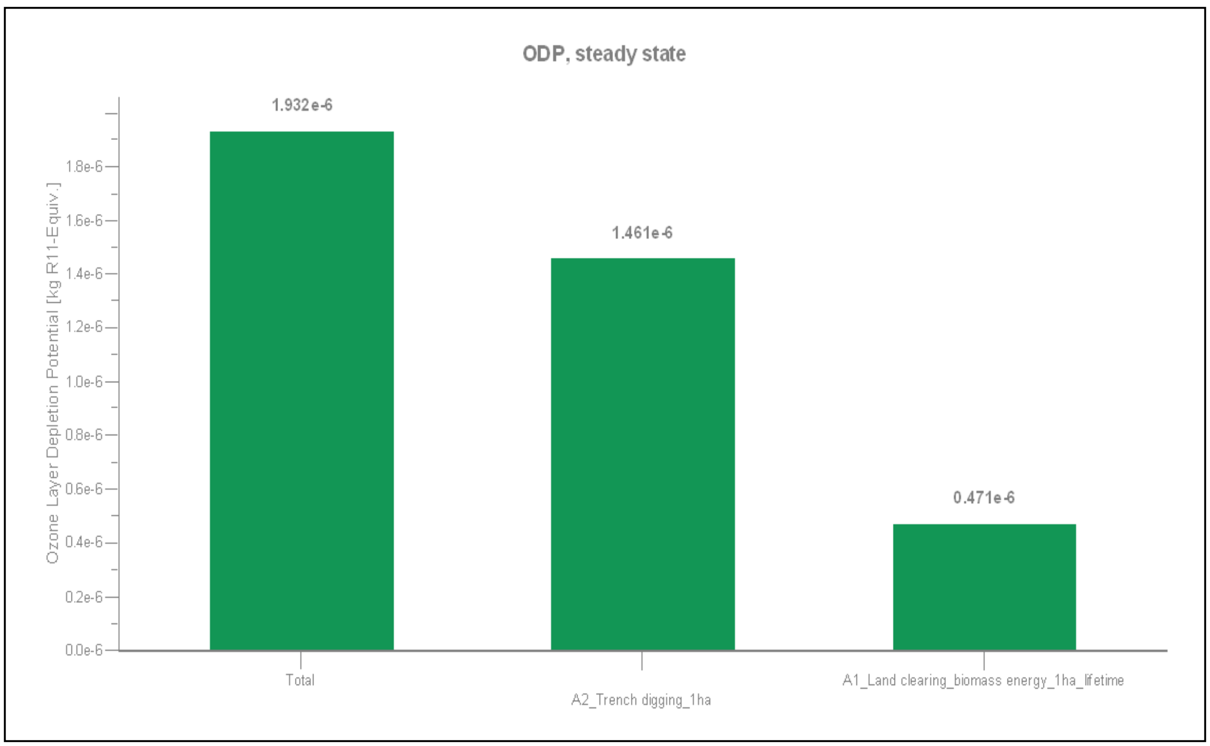
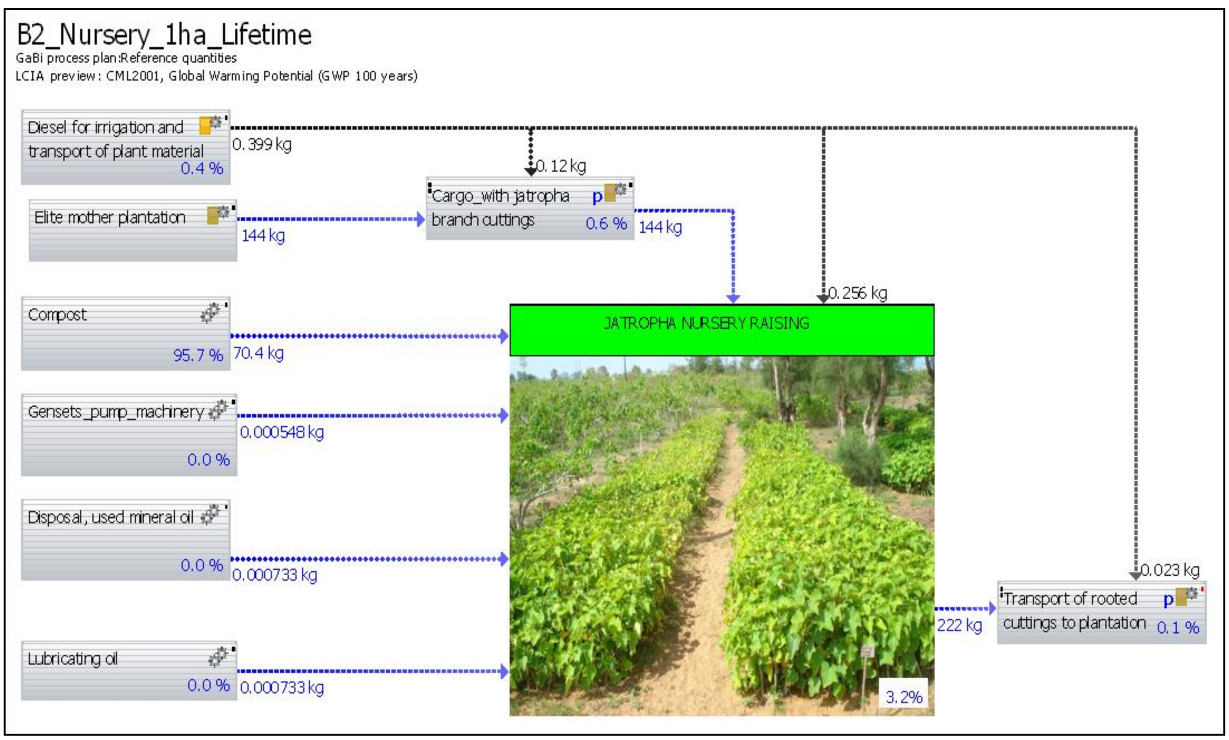


Figure A3. Life cycle impact of land preparation (1 ha area): a. GWP (Global warming potential); b. AP (Acidification Potential); c. EP (Eutrophication potential) d. ODP (Ozone depletion potential).

Nursery

This plan was generated to calculate the various environmental impacts during raising of nursery **(Figure B2)**. The nursery was raised through cuttings generated from elite germplasm. An innovative method of raising cuttings in sand beds was adopted here which has lower carbon foot print than raising nursery using conventional polythene bags. The duration of the nursery was seven months. All the inputs were apportioned based on the number of cuttings required for one hectare of plantation. A total of 1440 cuttings were raised in the nursery for every 1111 plants required for planting in hectare of plantation with 3m x 3 m spacing assuming a rooting percentage of 70. Water required for irrigation was met by pumping it from a well that was excavated manually. The nursery was irrigated manually. No inorganic fertilizer was used in the process and only compost was used for nutrient requirement. No pesticide application was done during the nursery period.



The assumptions and input values used for modeling land clearing is given in **Table A5 and A6**.

Table A5. GaBi plan describing various procesess in Nursery

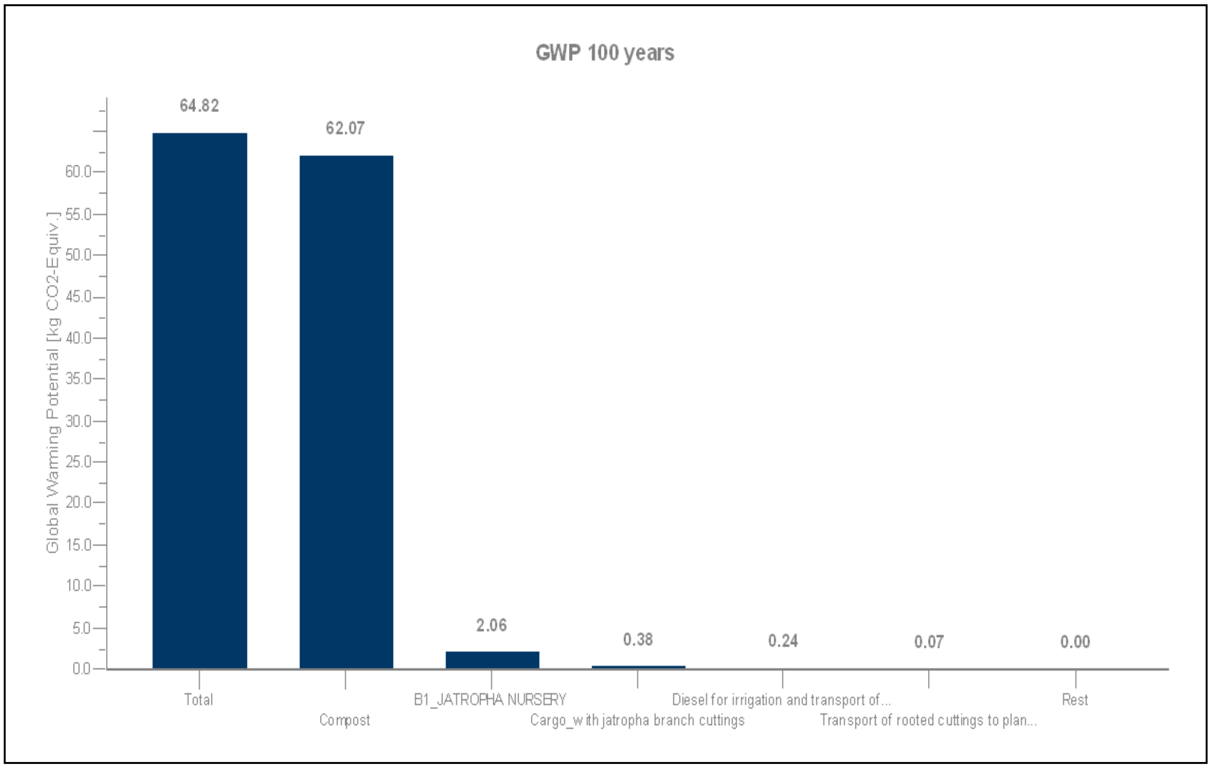
Process name	Module purpose	Comments
Nursery	Custom module calculating the impacts of all the sub processes given below along with emissions for combustion of diesel used in diesel gen-set cum pump	
Cargo with *Jatropha* cuttings	PE professional data set calculates impacts of diesel combustion for the transport of cargo with *Jatropha* cuttings	The sulphur content in diesel was assumed to be 10 ppm which is standard set in Europe.
Compost	This Eco-invent 2.2 process was used as a proxy for Indian compost preparation and calculates the impacts during its production	The impacts following the application of compost was calculated and also included. (Reference- IPCC 2006; Sharma et al., 2008; Garg et al., 2012).
Diesel for irrigation and transport of plant material	PE professional data set calculates the impacts of upstream crude oil production its transport and refining of diesel at Indian refinery	In order to determine the impacts of combustion of diesel in diesel run pump, emissions from Eco-invent database describing burning of diesel in gen-set was used.
Diesel genset and pump machinery	This Eco-invent 2.2 process was used as a proxy for calculating the impacts during production of the machinery	
Lubricants	PE professional data set calculates impacts for lubricant production at Indian refinery	
Disposal of mineral oil	This Eco-invent 2.2 process calculates the impacts during disposal of lubricating oil	This Eco-invent process is used as a proxy as specific data set for Indian conditions is not available
Elite mother plantation	A custom module required for connecting the transport of cargo module	This process does not calculate any impacts
Transport of rooted cuttings to plantation	PE professional data set calculates impacts of diesel combustion for the transport of rooted cuttings	The sulphur content in diesel was assumed to be 10 ppm which is standard set in Europe

Table A6. Inputs for Nursery module

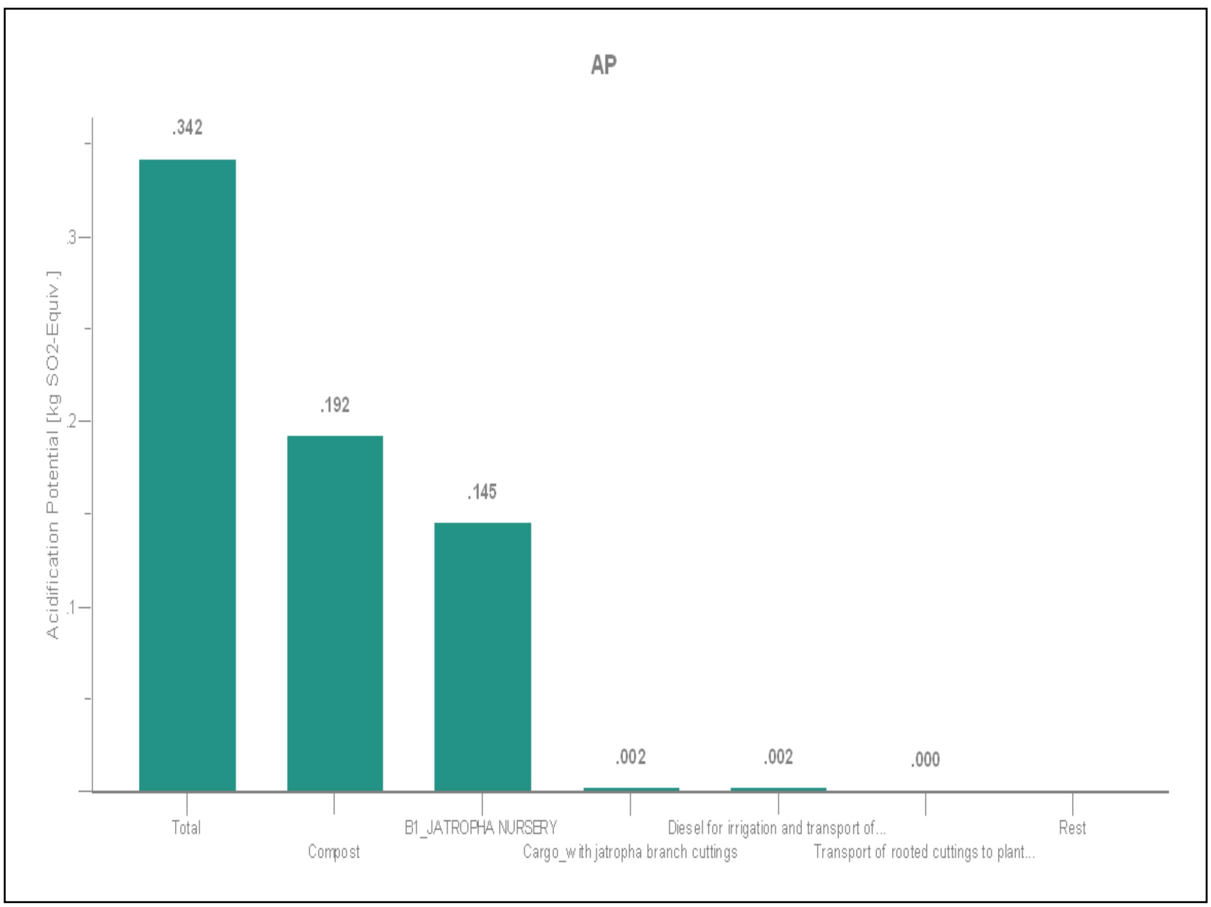
Parameter	Value	Unit	Documentation	Source
Cuttings	144	Kg	Average weight of cuttings was assumed to be 100g	Own data
No. of cuttings	1440	Kg	No of cuttings in nursery	Own data
Manure	70.4	Kg	Total amount of compost applied to 1440 plants.	Own data
Diesel	0.399	Kg	Total amount of diesel that is required for pumping water (0.000036192 kg of diesel was used for pumping 1 liter of water from well, assuming the density of diesel as 0.832Kg/L) and transport of elite cuttings in and out of nursery. The distance from mother plantation to field is assumed to be 5kms whereas, the distance between nursery and plantation is assumed to be 40kms.	Own data
Agricultural machinery	0.000548	Kg	Calculated as a product of pump life (146000 h - assumption), pump use per hectare (0.3078) and total wt. of the diesel pump (260 kg)	
Lubricants	0.0007334	Kg	Amount of lubricant required in diesel engine	Eco-invent database
Disposal of mineral oil	0.0007334	Kg	Amount of lubricant that is subjected to disposal in municipal incineration	Eco-invent database
Water	7080	L	Amount of water that is required for irrigating 1440 plants @ 70 ml per cutting every third day for a period of seven months	Own data

The life cycle impact across all the categories pertaining to overall land preparation is given in **Figure A4a-d.**

b



GWP 100 years
Global Warming Potential [kg CO2-Equiv.]
0.0
5.0
10.0
15.0
20.0
25.0
30.0
35.0
40.0
45.0
50.0
55.0
60.0
64.82
62.07
2.06
0.38
0.24
0.07
0.00
Total
Compost
B1_JATROPHA NURSERY
Cargo_with jatropha branch cuttings
Diesel for irrigation and transport of...
Transport of rooted cuttings to plan...
Rest



AP
Acidification Potential [kg SO2-Equiv.]
.1
.2
.3
.342
.192
.145
.002
.002
.000
Total
Compost
B1_JATROPHA NURSERY
Cargo_with jatropha branch cuttings
Diesel for irrigation and transport of...
Transport of rooted cuttings to plant...
Rest

d

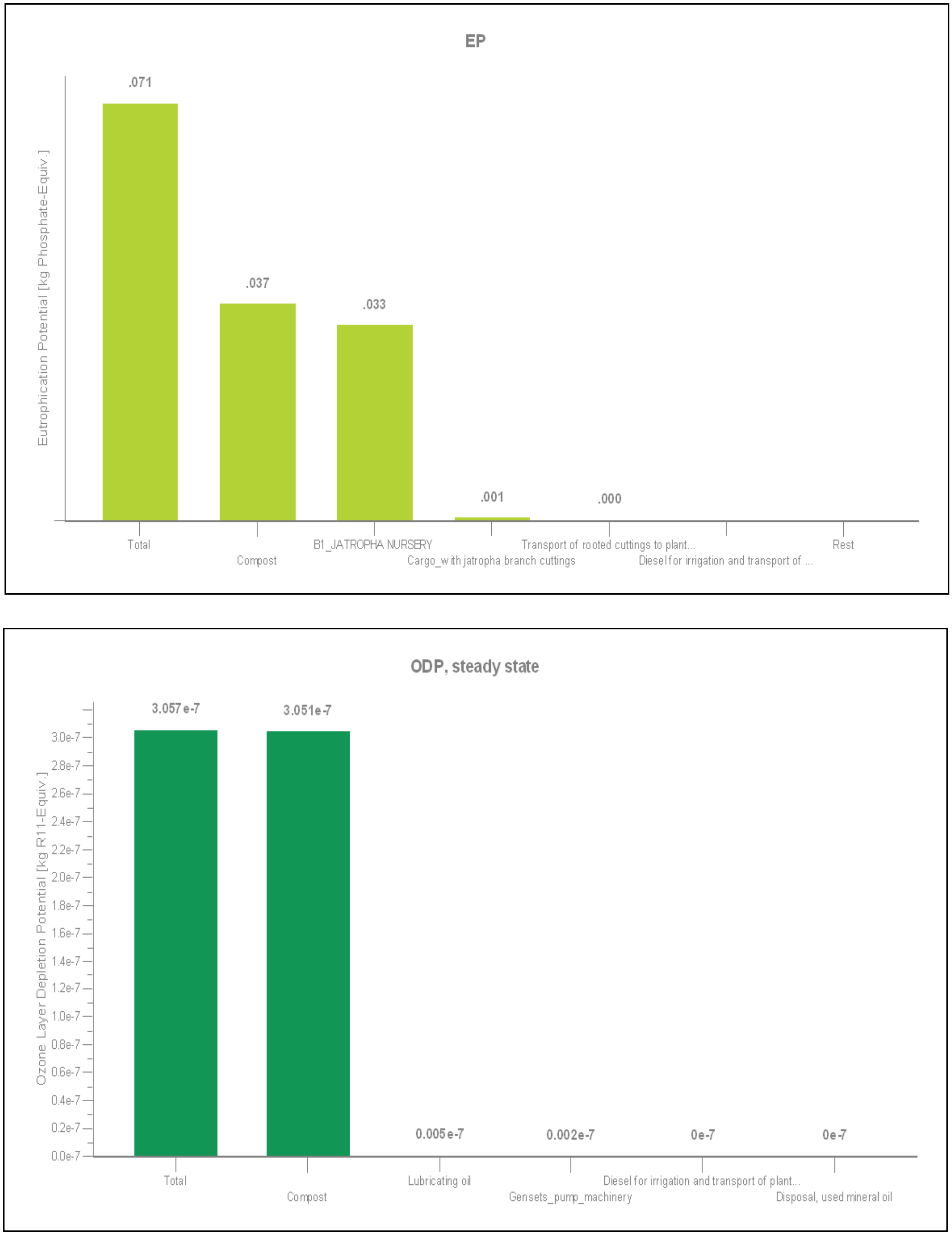
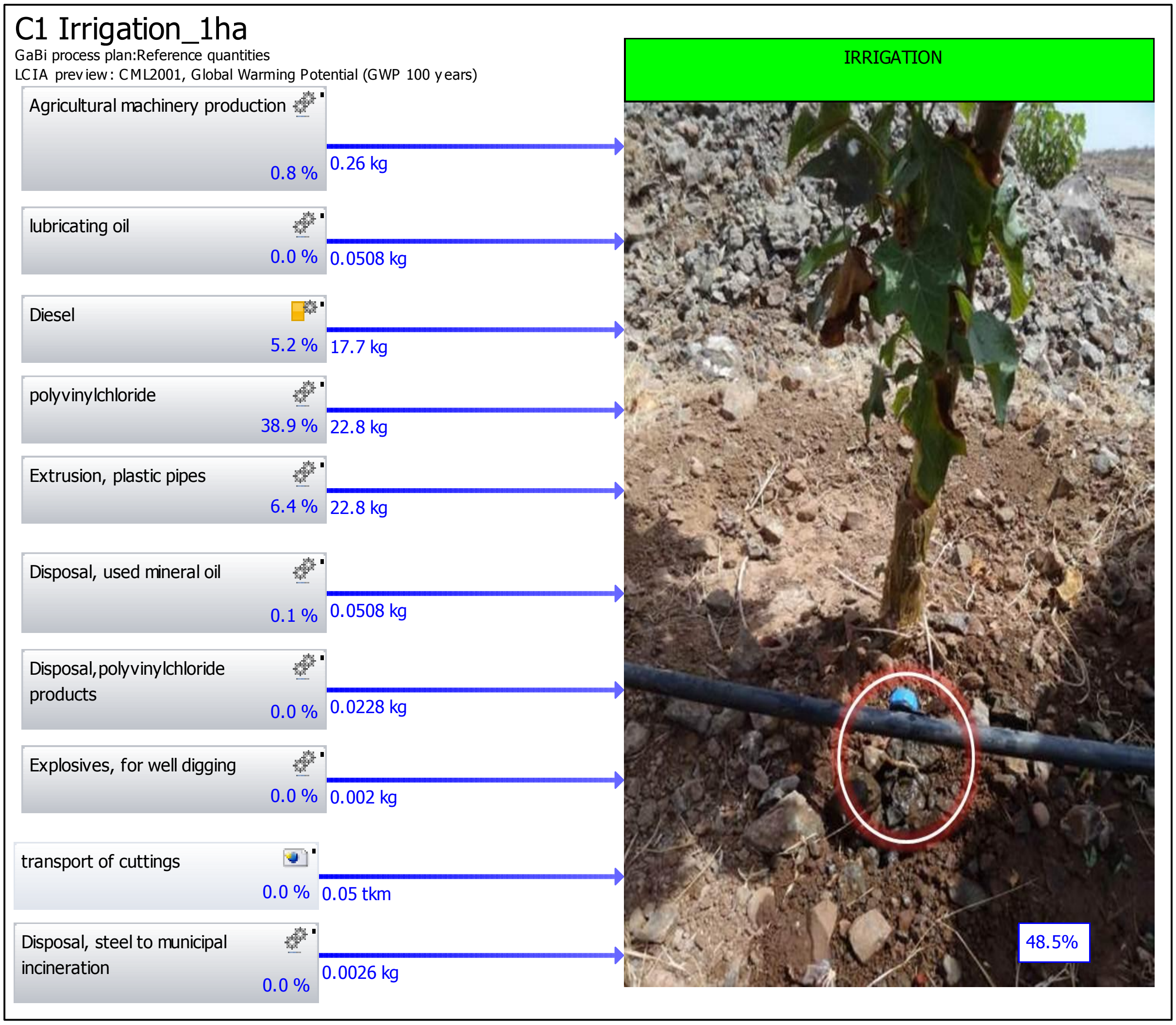


Figure A4. Life cycle impact of Nursery preparation (1 ha area): a. GWP (Global warming potential); b. AP (Acidification Potential); c. EP (Eutrophication potential) d. ODP (Ozone depletion potential).

Irrigation module:

In order to increase the water use efficiency a drip irrigation system was used. All the inputs were apportioned based on the number of cuttings required for one hectare of plantation. The irrigation module consisted of PVC pipes, laterals, filters and cocks. Water for irrigation is pumped from well by a diesel run pump into storage tank from where it is distributed.



The assumptions and input values used for modeling land clearing is given in **Table A7 and A8**.

Table A7. GaBi plan describing the irrigation system

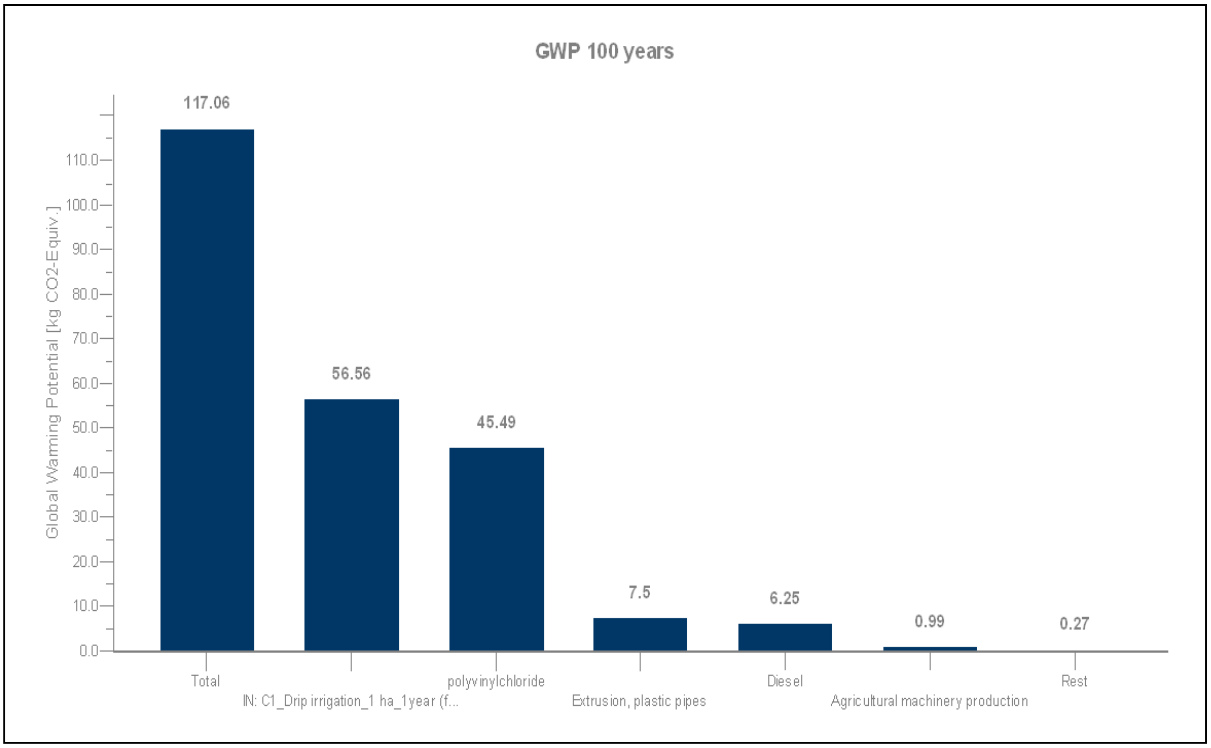
Process name	Module purpose	Comments
Irrigation	Custom module calculating the impacts of all the sub processes given below along with emissions for combustion of diesel used in diesel gen-set cum pump	In order to determine the impacts of combustion of diesel in diesel run pump, emissions from Eco-invent database describing burning of diesel in gen-set was used.
Agricultural machinery	This Eco-invent 2.2 process was used as a proxy for calculating the impacts during production of the machinery	
Lubricating oil	PE professional data set calculates impacts for lubricant production at Indian refinery	
Diesel	PE professional data set calculates the impacts of upstream crude oil production its transport and refining of diesel at Indian refinery	
Polyvinyl chloride	This Eco-invent 2.2 process was used as a proxy for calculating the impacts during production of the PVC granules	As no LCA data on PVC production in India is available Eco-invent database was used for the purpose
Extrusion, plastic pipes	This Eco-invent 2.2 process calculates the impacts during conversion of PVC granules into pipes	As no LCA data on PVC extrusion in pipes is available in India Eco-invent database was used as a proxy for the purpose
Disposal of mineral oil	This Eco-invent 2.2 process calculates the impacts during its disposal of lubricating oil	This Eco-invent process is used as a proxy as specific data set for Indian conditions is not available
Disposal Polyvinylchloride products	This Eco-invent 2.2 process calculates the impacts during disposal of polyvinyl products	This Eco-invent process is used as a proxy as specific data set for Indian conditions is not available
Explosives	This Eco-invent 2.2 process calculates the impacts during production of explosives.	This Eco-invent process is used as a proxy as specific data set for Indian conditions is not available. Explosives were used during digging of well.
Transport of cuttings	This customized module calculates the impacts of transporting PVC and other drip accessories to the field	The Eco-invent module of transport using 20-28 t fleet average was used for the purpose of determining the impact of emissions. This customized module uses Indian data set for diesel
Disposal steel municipal incineration	This Eco-invent 2.2 process calculates the impacts during disposal of steel used in pump	This Eco-invent process is used as a proxy as specific data set for Indian conditions is not available

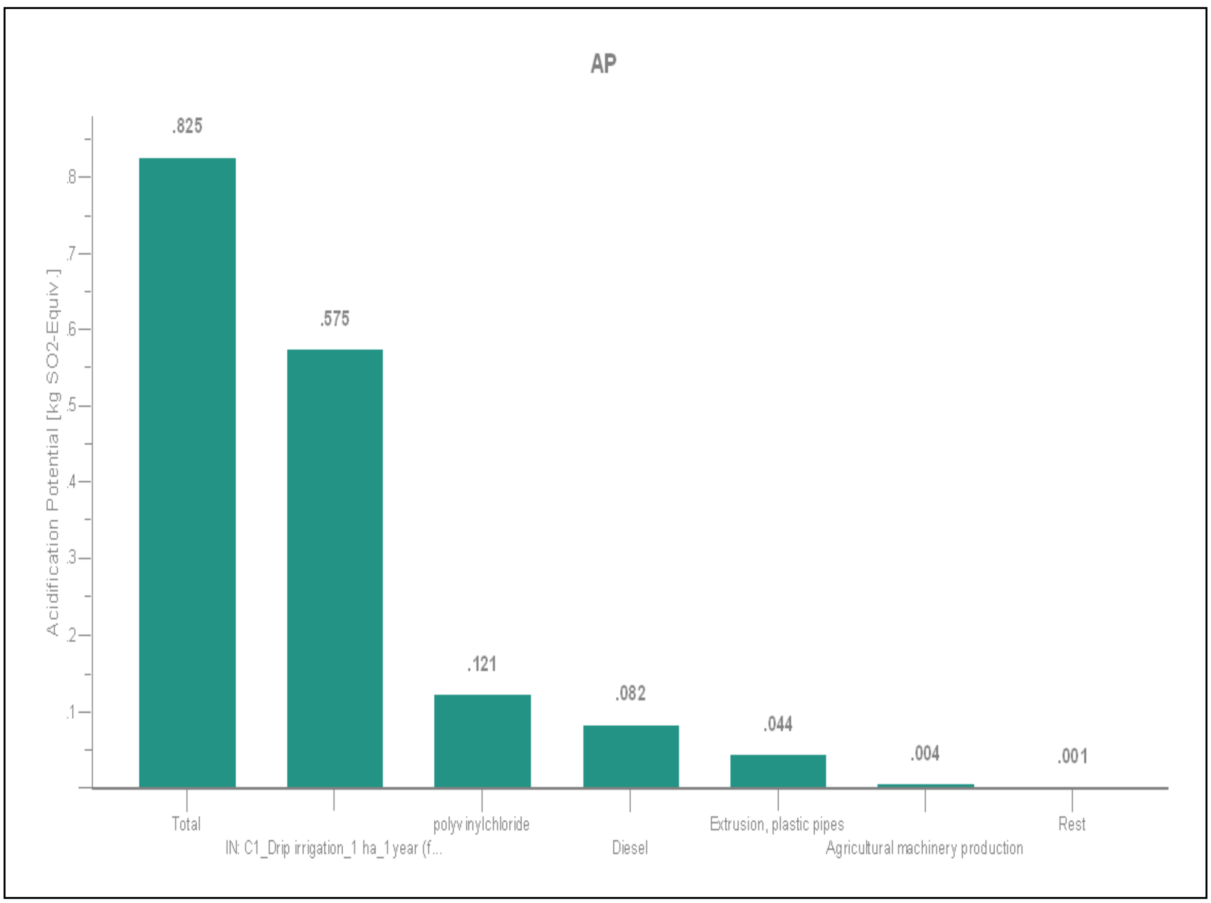
Table A8. Inputs for Irrigation module

Parameter	Value	Unit	Documentation	Source
Agricultural machinery	0.26	Kg	Apportioning of 260 kg pump and diesel gen set with a life of 100 years (assumption) for one year as one pump can suffice for irrigating 10 ha.	Own data
Transport of cuttings	0.05	tkm	Apportioning of the transport of 500 kg material over a distance of 25 kms (assumption) for one year.	Own data
Diesel	17.7	Kg	Amount of diesel that is used for pumping 278000 liters of water from well over a period of one year for one hectare	Own data
Lubricants	0.0508	Kg	Amount of lubricant used in diesel unit of the pump	Own data
Disposal of mineral oil	0.0508	Kg	Amount of lubricating oil that is subjected to disposal	Eco-invent database
Polyvinyl chloride	22.8	Kg	Amount of PVC granules that is apportioned for one year out of a total of 456.3 kg with an assumed life span of 20 yrs	
Extrusion, plastic pipes	22.8	Kg	Amount required for extrusion of granules into pipes	Estimation based on own data
Disposal of Polyvinyl chloride products	0.0228	Kg	0.1% of the total weight of PVC is subjected to disposal (assumption)	
Explosives	0.002	Kg	Apportioned amount of explosive that is used in the well which is assumed to last for 100 yrs and sufficient for 25 ha irrigation	Own data
Disposal of steel	0.0026	Kg	0.1% of the total wt of agricultural machinery	
Water	278000	L	Product of water requirement per irrigation per plant (50 L), number of irrigations per year and number of plants per ha (1111). Parameterized in the process for calculating the amount of diesel requirement in diesel pump	Own data

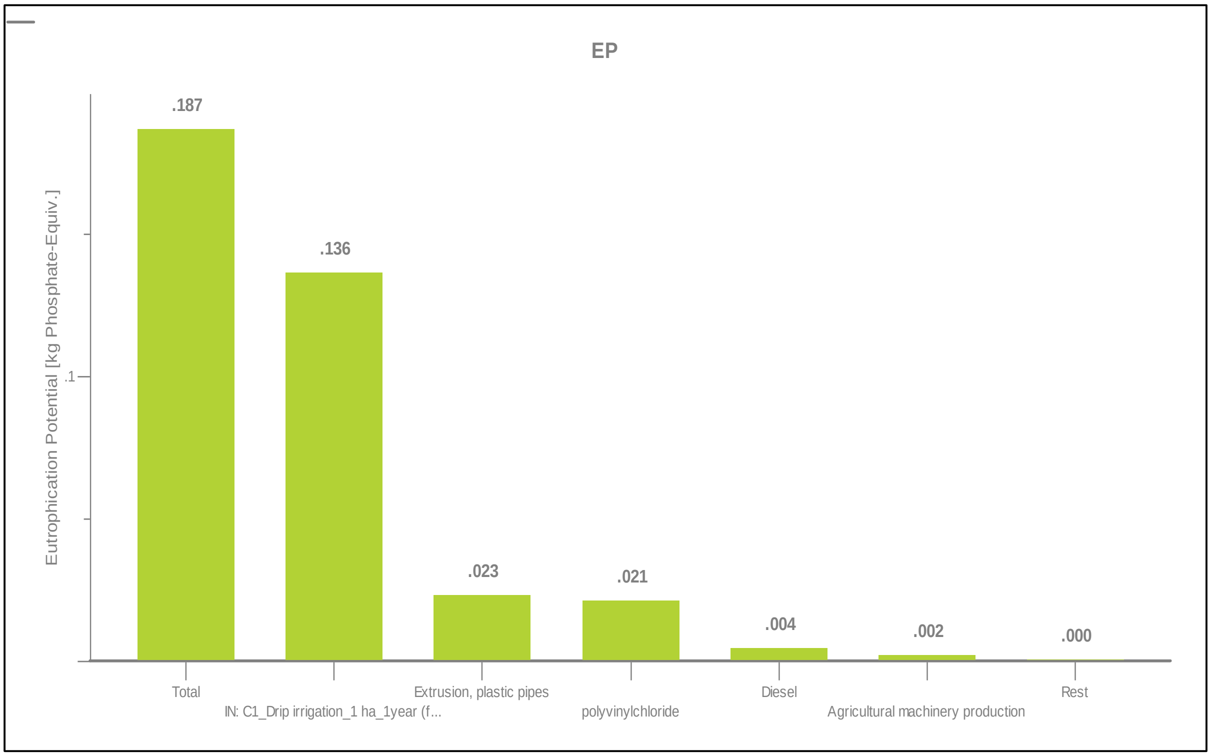
The life cycle impact across all the categories pertaining to overall land preparation is given in **Figure A5a-d.**

b


GWP 100 years
Global Warming Potential [kg CO2-Equiv.]
117.06
56.56
45.49
7.5
6.25
0.99
0.27
Total
IN: C1_Drip irrigation_1 ha_1year (f...
polyvinylchloride
Extrusion, plastic pipes
Diesel
Agricultural machinery production
Rest


AP
Acidification Potential [kg SO2-Equiv.]
.825
.575
.121
.082
.044
.004
.001
Total
IN: C1_Drip irrigation_1 ha_1year (f...
polyvinylchloride
Diesel
Extrusion, plastic pipes
Agricultural machinery production
Rest

d



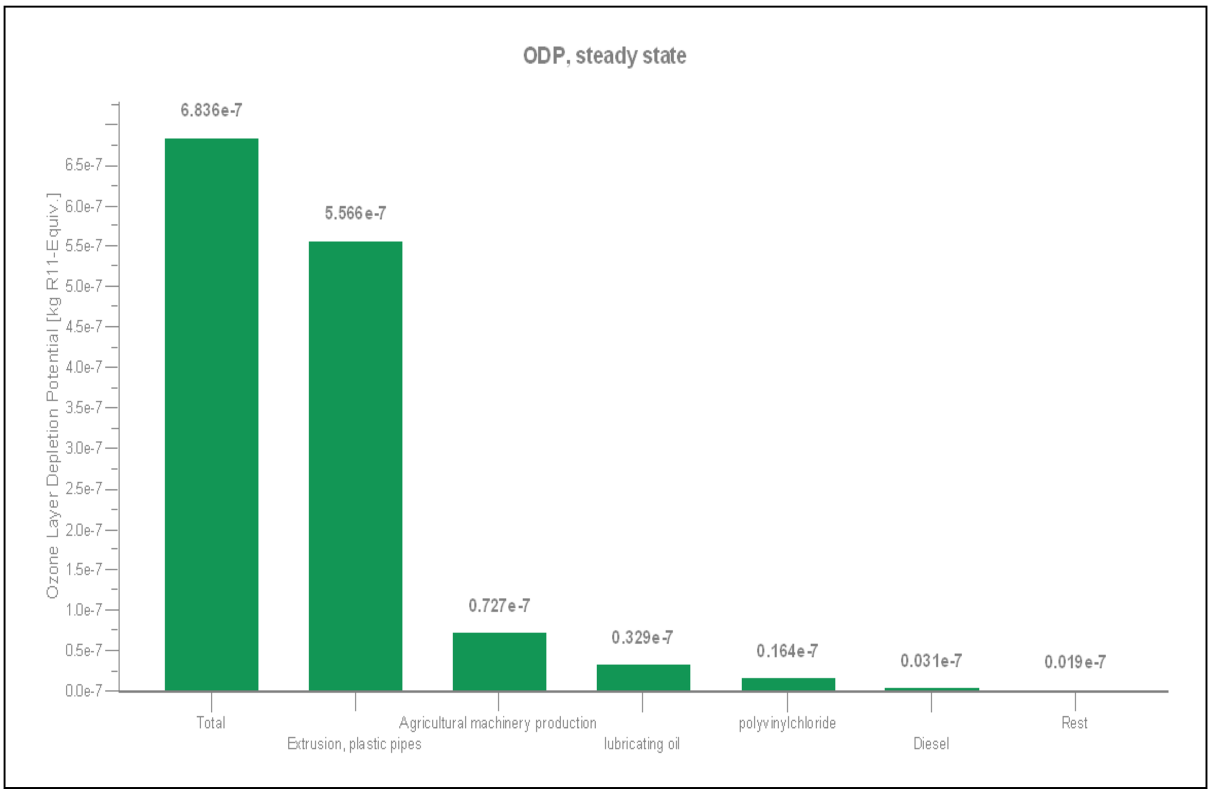
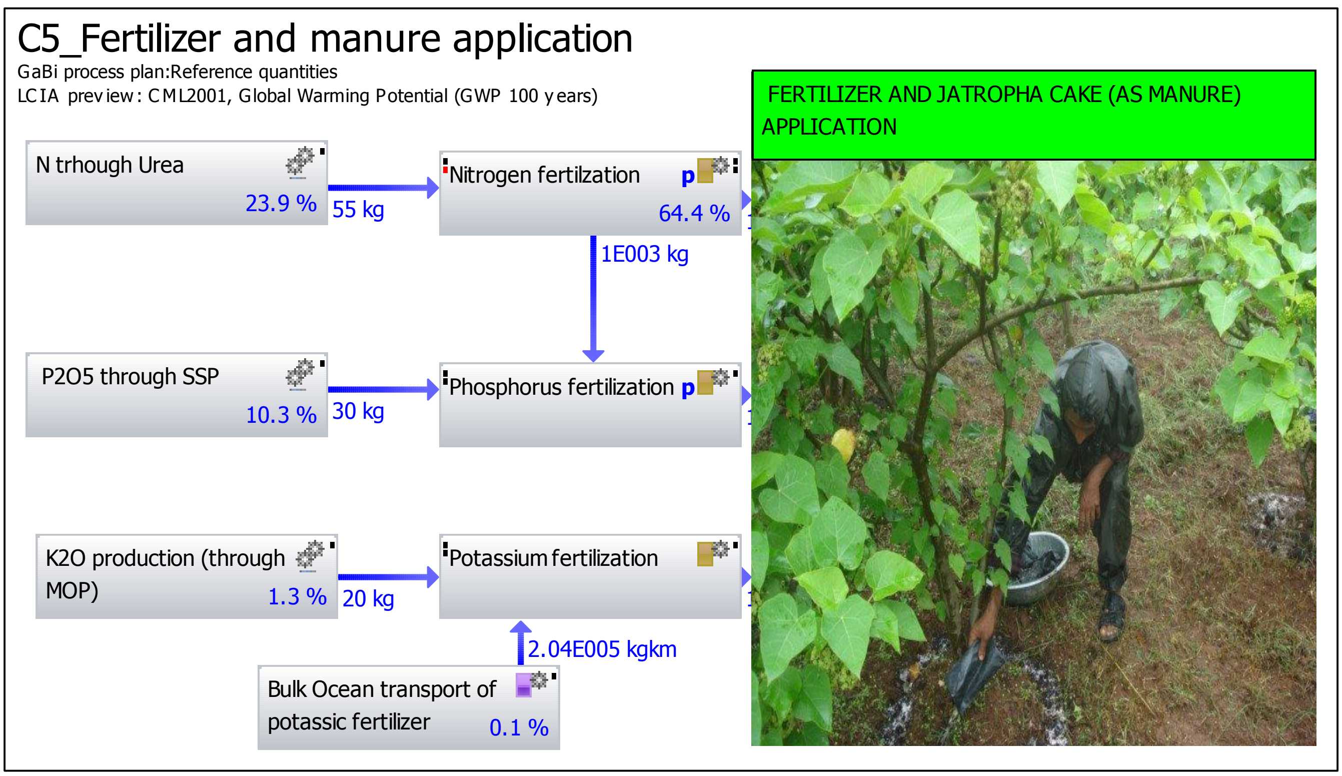


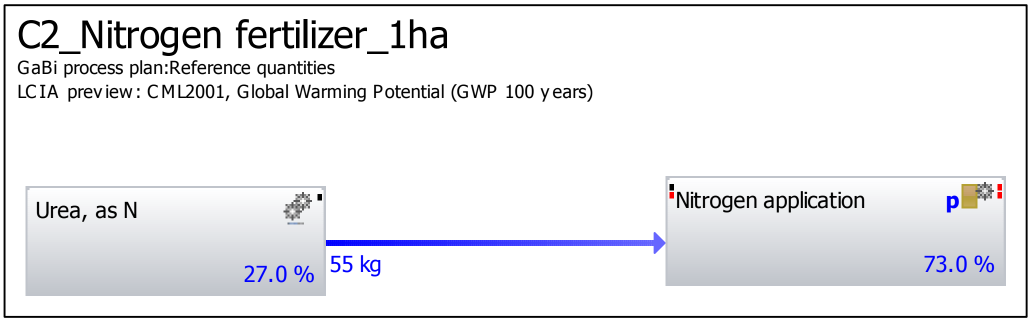
Figure A5. Life cycle impact of Irrigation (1 ha area): a. GWP (Global warming potential); b. AP (Acidification Potential); c. EP (Eutrophication potential) d. ODP (Ozone depletion potential).

Fertilizer and manure application module:

This GaBi plan was developed to calculate the impacts of Fertilizer production as well as its application **(Figure C5)**. The module has sub plans describing Nitrogen, Phosphorus and Potassic fertilization and their use phase **(Figure C2, C3 and C4)** and contribution to their CML 2008 GWP potential for their manufacture and use phase is also. It can be seen that for nitrogen fertilization, production of *Jatropha* de-oiled seed cake generated as a by-product of oil expulsion was also used to supplement inorganic Nitrogen and Phosphorus fertilization however no allocation is claimed for the chemical fertilizer saved. All the inputs were calculated for basis of their requirement for one hectare per annum. Notably, for nitrogen fertilization, production of urea accounted for 27% of GWP out of 671.63 kg of CO2-Equiv/ha, while rest was emitted due to application of urea and manure **(Figure C2 & A6)**.



The assumptions and input values used for modeling land clearing is given in **Table A9 and A10**, while the life cycle impact across all the categories pertaining to overall land preparation is given in **Figure A7 a-d.**



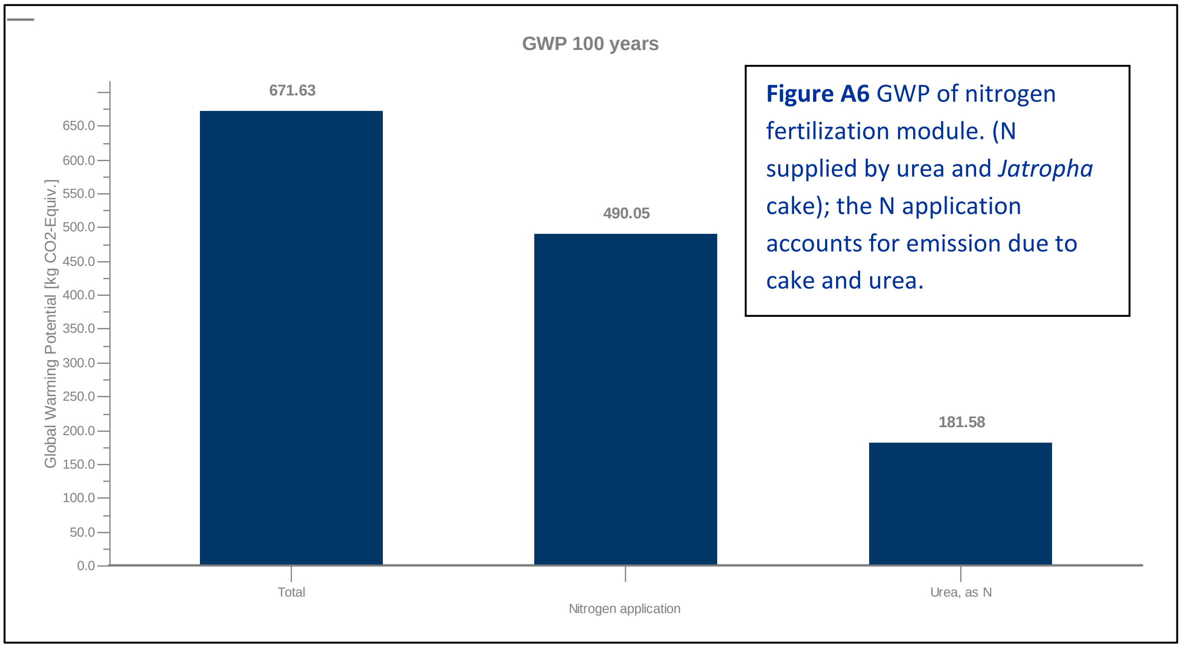
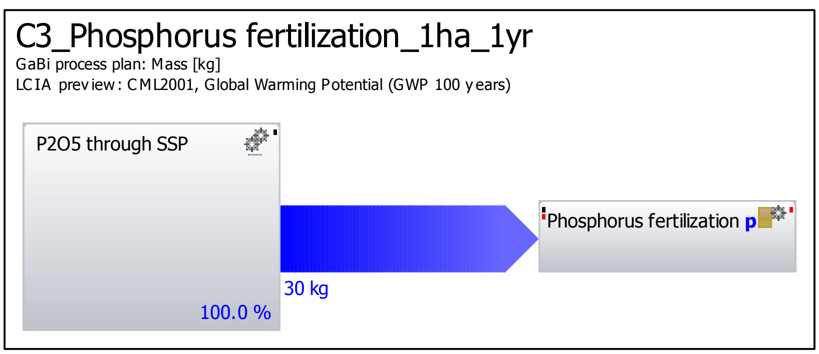


Figure A6 GWP of nitrogen fertilization module. (N supplied by urea and *Jatropha* cake); the N application accounts for emission due to cake and urea.



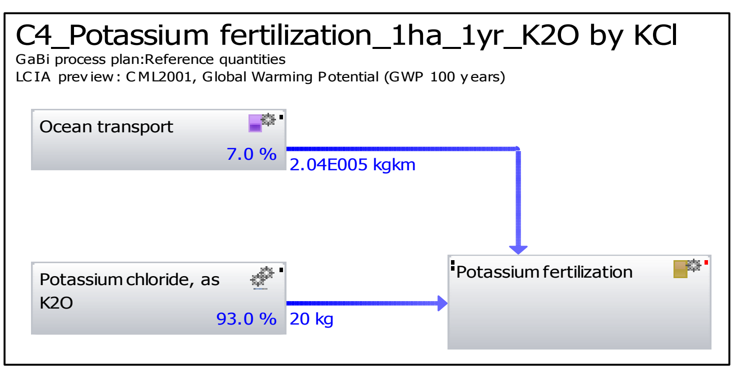
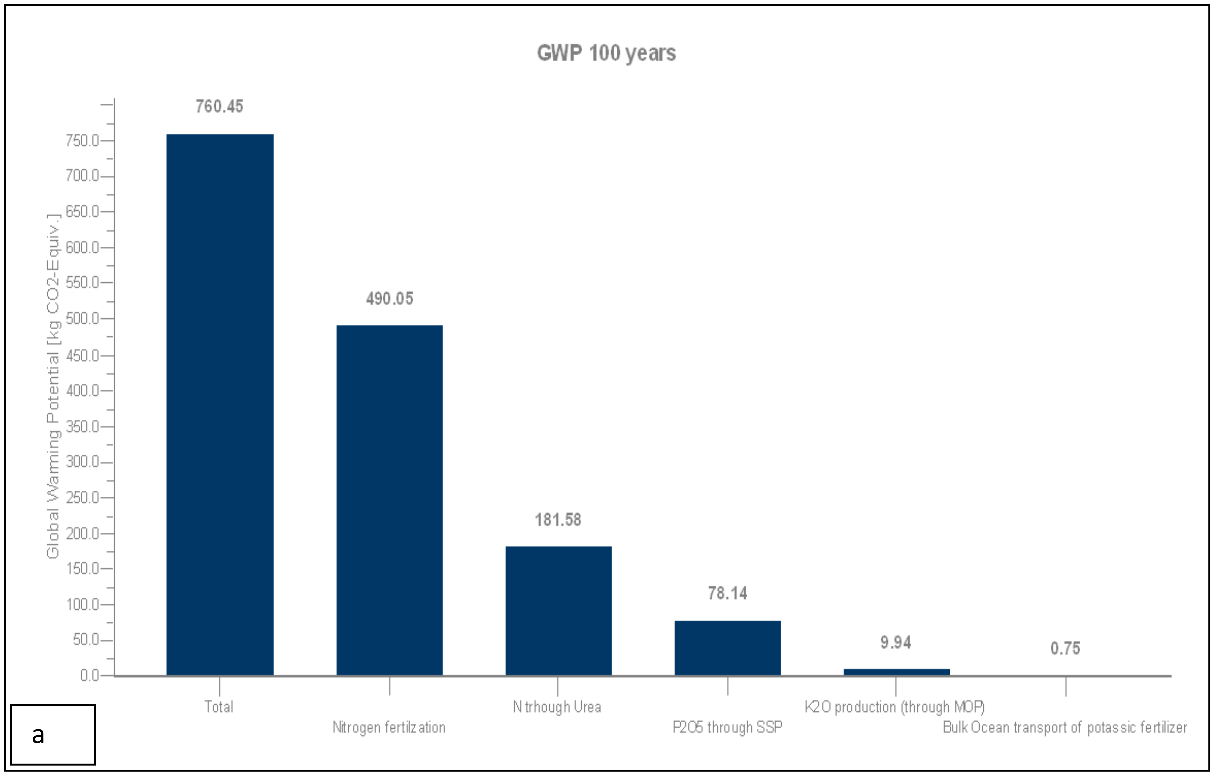


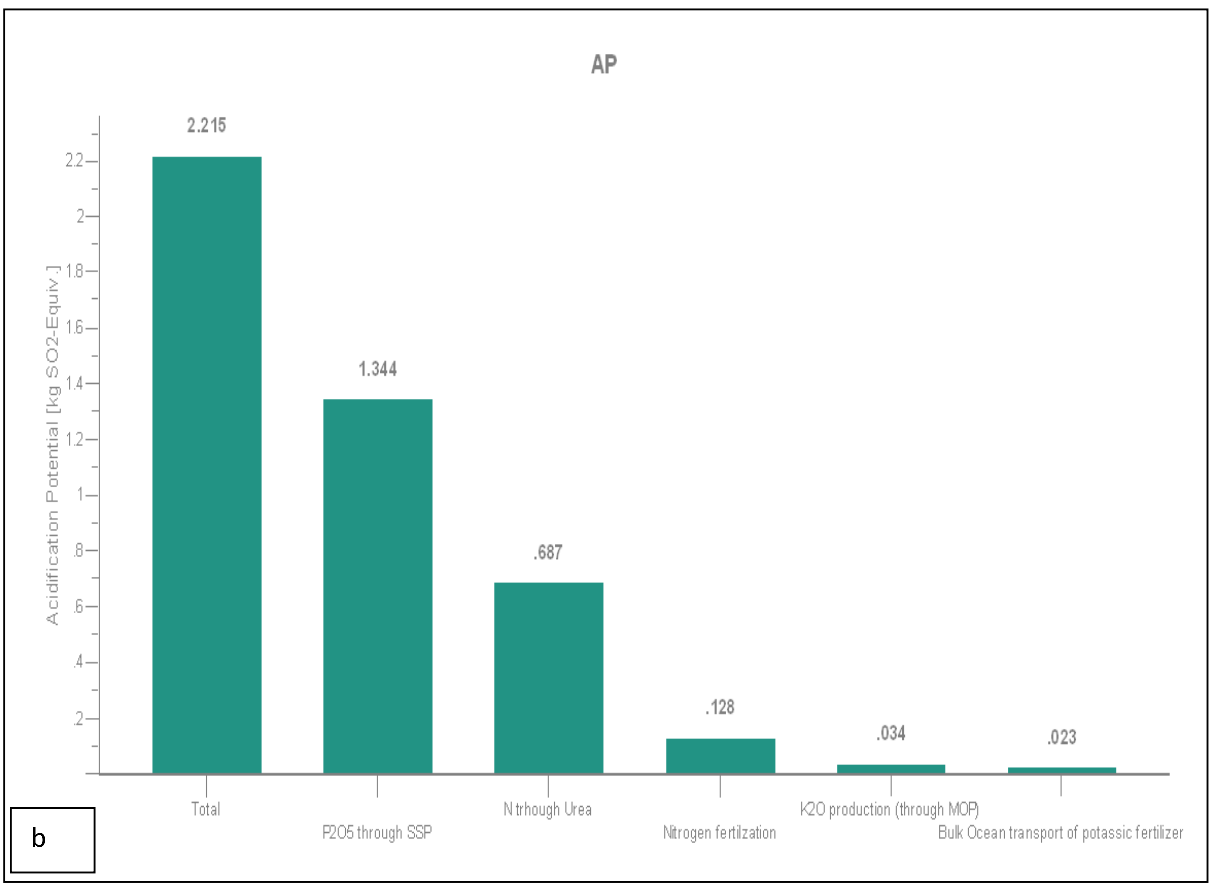
Table A9. GaBi plan for fertilization and manure

Process name	**Module purpose**	**Comments**
Fertilization and manure application	Custom GaBi module made by us for calculating the impacts of nitrogen, phosphorus as well as potassic fertilization	
Nitrogen application	This custom GaBi module calculates the emission impacts of both organic and inorganic nitrogen fertilizer production as well as their application in the field and nitrogen obtained during recycling of crop residue	Urea was used as an inorganic source of Nitrogen. This module was connected to Eco-invent 2.2 module Urea as N which calculates the emissions during Urea production. The latter Eco-invent process is used as a proxy, as specific data set for urea production in India is not available. *Jatropha* cake that is obtained as by product during oil expulsion was used as a source of organic fertilization. Nitrogen from excreta and urine of grazing animals is not considered as *Jatropha* is not grazed by herbivores.
Phosphorus fertilization	This custom GaBi module calculates the emission impacts of both organic and inorganic phosphorus fertilizer production as well as their application in the field	Single super phosphate was used as an inorganic source of Phosphorus. This module also includes Eco-invent 2.2 module SSP through P_2O_5 which calculates the emissions during SSP production. The latter Eco-invent process is used as a proxy, as specific data set for SSP production in India is not available. *Jatropha* cake that is obtained as by product during oil expulsion was used as a source of organic fertilization.
Potassic fertilization	This custom GaBi module calculates the emission impacts of transport of potassic fertilizer as well as their production and application in the field	Potassium chloride, as K_2O was used as a source of potassium fertilizer. The module also includes impacts resulting from transport of potassic fertilizer by oceanic bulk carrier from Belarus (assumed that bulk (>50%) of potassic fertilizer is imported into India from Belarus)

Table A10. Inputs for the module Fertilization and manure application

Parameter	Value	Unit	Documentation	Source
Urea as N	55	Kg	Amount of Urea N applied to one hectare in one year	Own data
Jatropha N cake	35	Kg	One tonne of cake was used with N content of 3.5%	Own data
Direct N_2O emission factor	0.0065		Emission factor for Indian conditions (N2O-N/kg N input)	Garg et al., 2012
N_2O leaching Factor	0		No leaching of N_2O is assumed due to use of drip and rocky soil	
Indirect N_2O volatilization emission factor	0.005		Emission factor for N volatilization and re-deposition, kg N2O–N (kg NH3–N + NOX–N Volatilized)	Garg et al., 2012
NO_x Fraction	0.21		Total NO_x Fraction	IPCC 2006
Leaf residue	1332.2	kg	Total amount of dry leaf residue per hectare @1.2 kg leaf residue/ plant	Own data
N content in leaf	0.02		2% N content in leaves	Own data
Wood/stem residue	555.5	Kg	Total amount of dead wood and stem residue per hectare per year @ 0.5 kg/ plant	Own data
N content in stem	0.004		0.4 % N content in stems	Own data
N crop residue	28.9	kg	Total N supplied per year through crop residues (kg/ha)	
NH_3 volatilization factor inorganic	0.08		8% lost as N- NH3 volatilization of total N supplied as inorganic fertilizer in wheat. Hence a similar value is assumed for *Jatropha*	Sharma et al., 2008
Carbon emission factor	0.2		Carbon emission factor from urea	IPCC 2006
NH_3 volatilization factor organic	0.2		Default Fraction for Volatilization from all organic N fertilizers applied	IPCC 2006
P_2O_5 as SSP	30	Kg	Amount of P_2O_5 supplied as SSP for one hectare	Own data
P Cake	12	Kg	Amount of P_2O_5 supplied through one tonne of cake with P_2O_5 content of 1.2%	
P leaching frac	0		Phosphorus leaching fraction is assumed as zero due combination of factors such as scanty rainfall, drip irrigation and presence of rock below	
P content factor	0.00095		The average P content for top soil is assumed to be 0.00095 kg/kg soil	Eco-invent report 15a
P run off fraction	0.175		Refers to the average quantity of P lost through run-off for arable land	Eco-invent report 15a
Correction factor for run off	1.14		Calculated according to formula in Eco-invent report	Eco-invent report 15a
Quantity Soil eroded	5000	t/ha/yr	Quantity of soil that is subjected to erosion in the state of Gujarat	Sharda et al., 2013
Enrichment factor for P	1.86		Correction factor that takes into account the higher content of P in eroded soils than average soil	Wilke and Schaub 1996
Fraction to river	0.2		This is the fraction of eroded soil that reaches river	Eco-invent report 15a
Potassium chloride as K_2O	20	Kg	Amount of MOP used in one hectare of plantation	
Oceanic transport	203720	Kgkm	Transport potassic fertilizer from Belarus to India in bulk oceanic transport carrier	


GWP 100 years
Global Warming Potential [kg CO2-Equiv.]
760.45
490.05
181.58
78.14
9.94
0.75
Total
Nitrogen fertilzation
N trhough Urea
P2O5 through SSP
K2O production (through MOP)
Bulk Ocean transport of potassic fertilizer
a


AP
Acidification Potential [kg SO2-Equiv.]
2.215
1.344
.687
.128
.034
.023
Total
P2O5 through SSP
N trhough Urea
Nitrogen fertilzation
K2O production (through MOP)
Bulk Ocean transport of potassic fertilizer
b

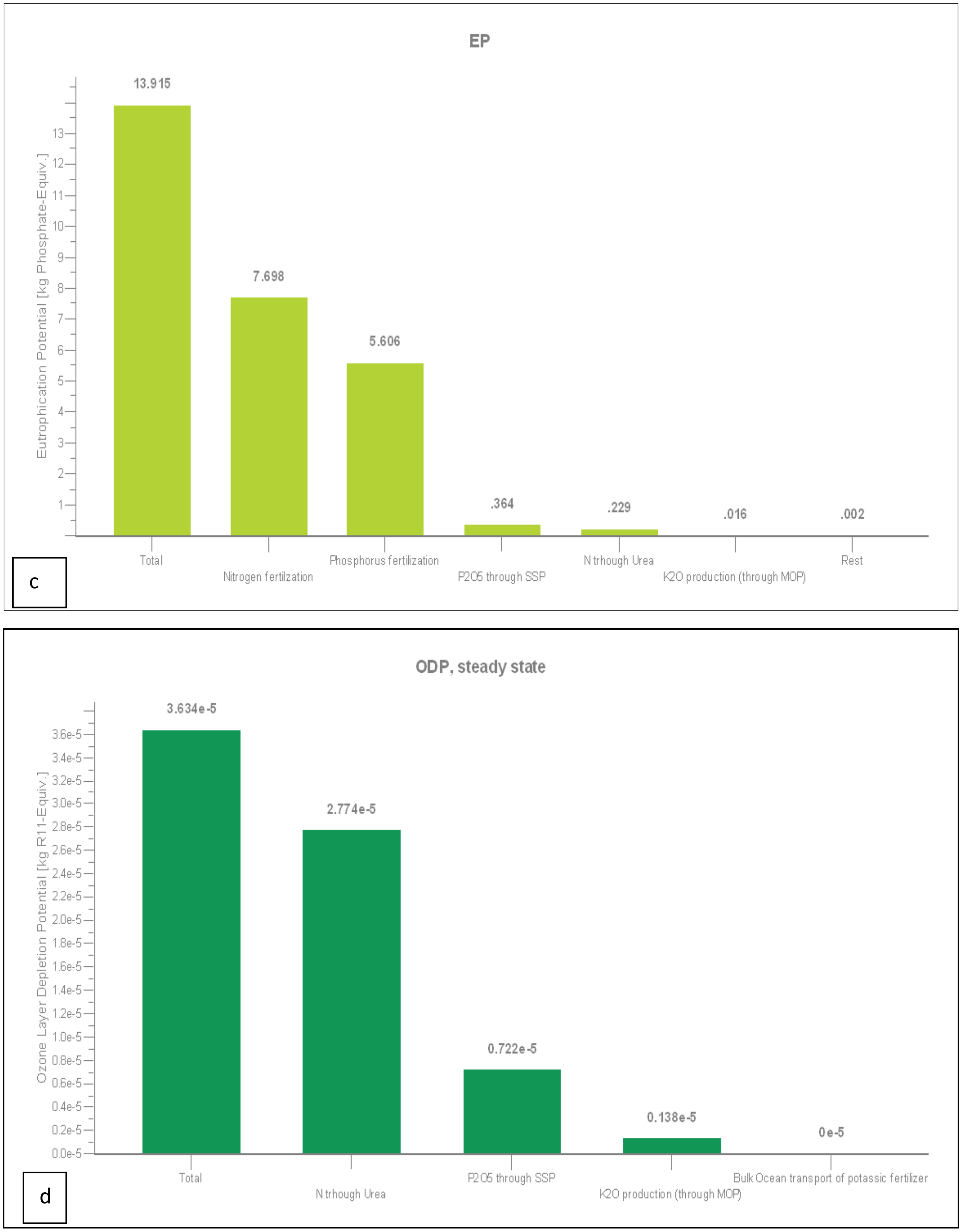


Figure A7. Life cycle impact due to fertilizer (1 ha area): a. GWP (Global warming potential); b. AP (Acidification Potential); c. EP (Eutrophication potential) d. ODP (Ozone depletion potential).

Pesticide application module:

This module calculates the impacts of application of fungicide and insecticide in *Jatropha* plantations. The insecticide was primarily used to control termites and white flies whereas, fungicide was applied to control the incidence of collar rot caused by *Macrophomina phaseolina*. The assumptions and input values used for modeling land clearing are given in **Table A11 and A12**. The life cycle impact across all the categories pertaining to pesticide application is given in **Figure A8a-d.**

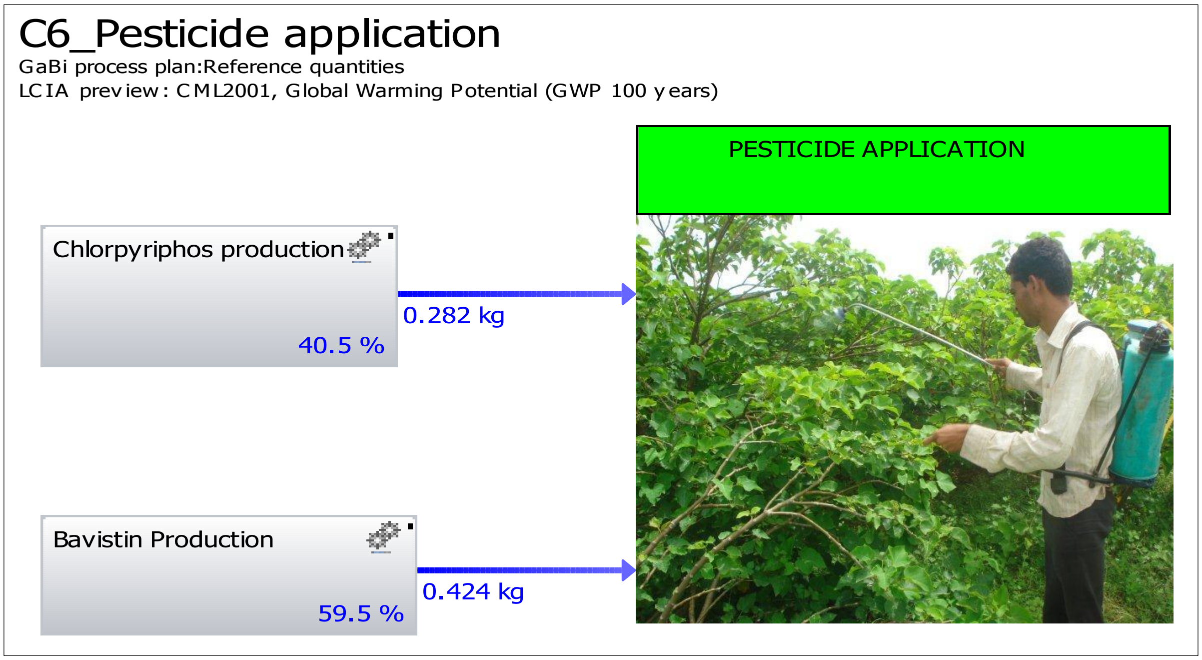
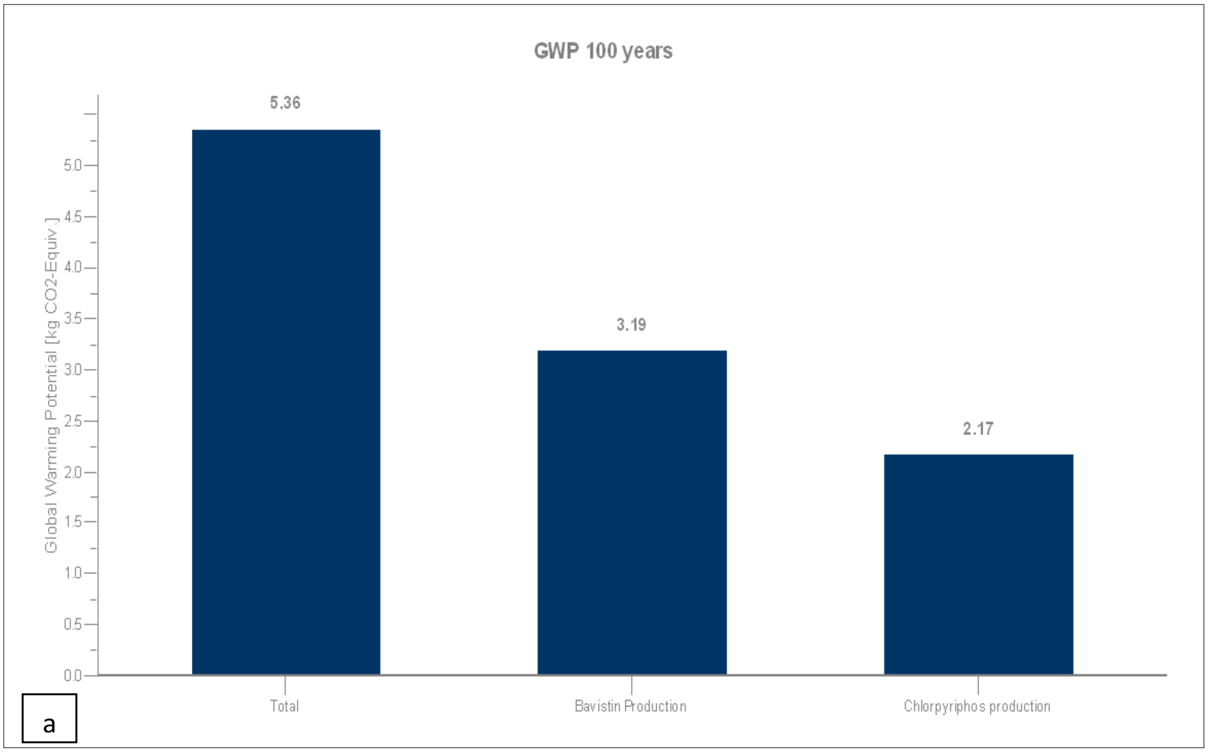


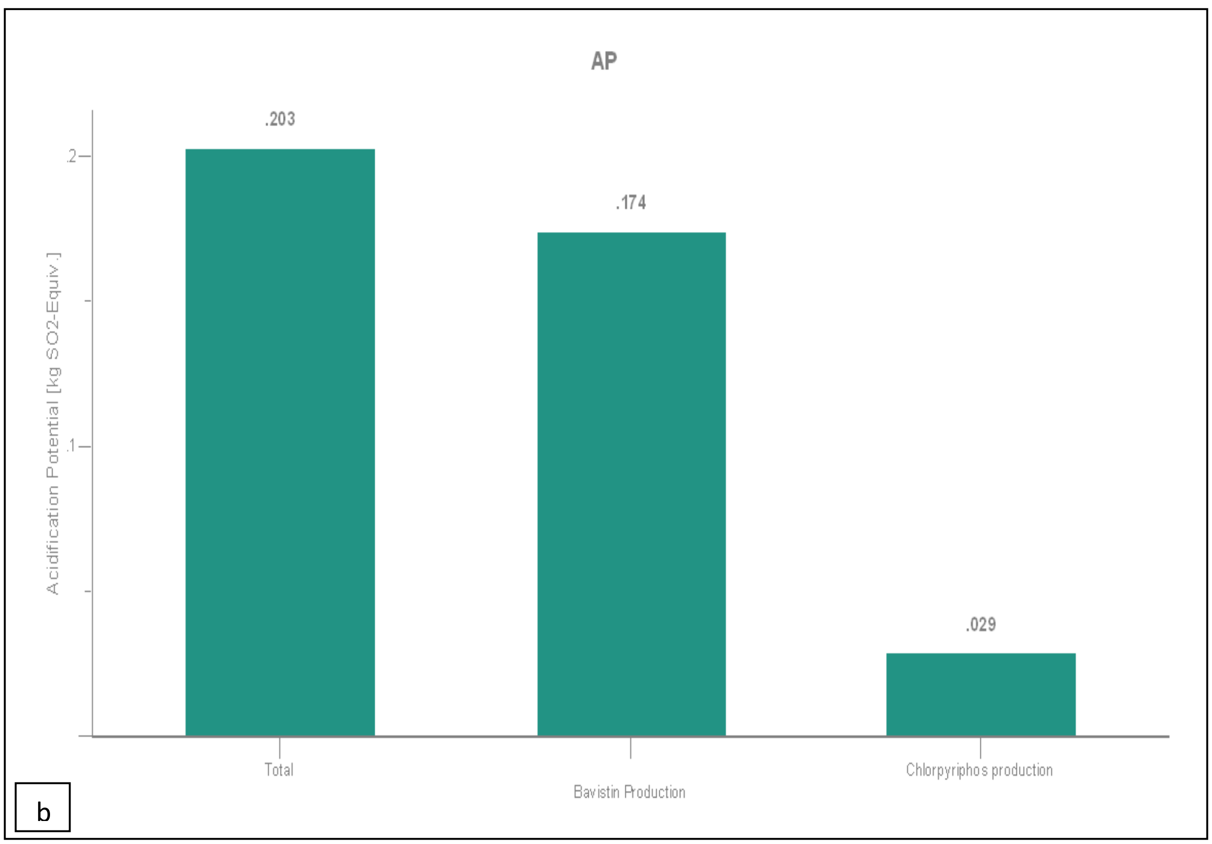
Table A11. GaBi plan to describe pesticide application

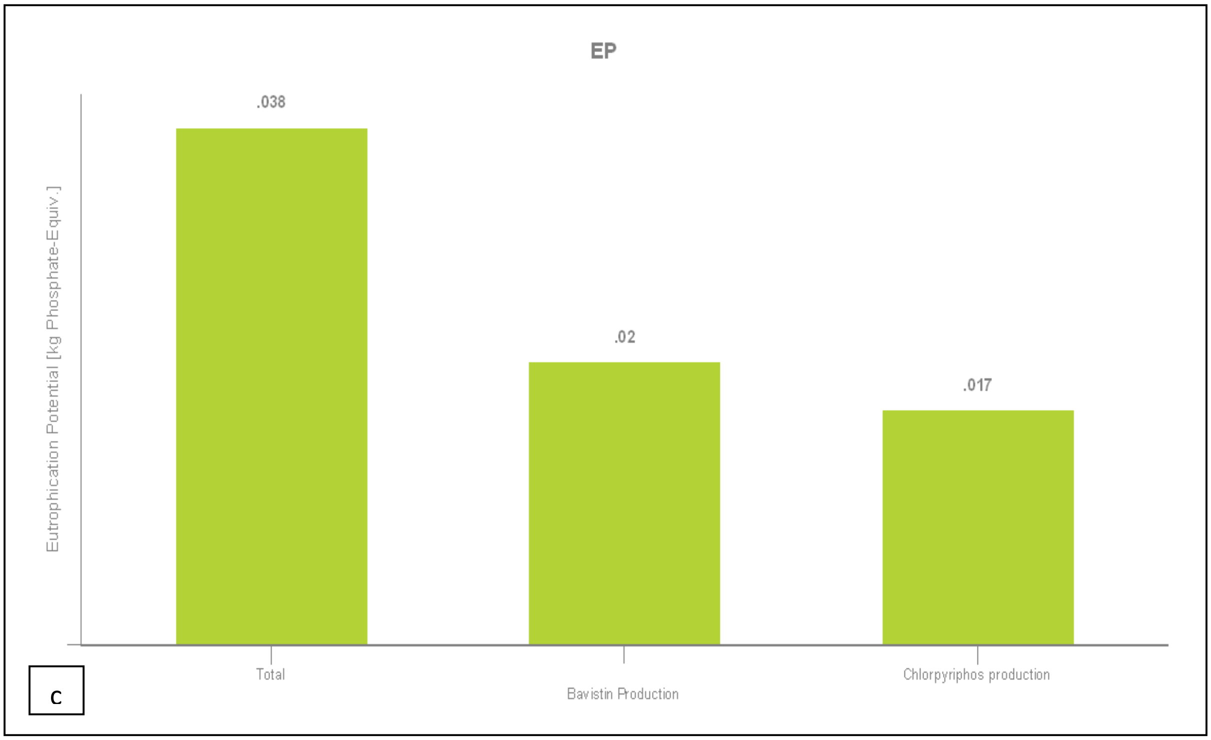
Process name	Module purpose	Comments
Pesticide application	Custom module that calculates the impacts of the sub-processes of the pesticides used both in terms of their production and application	
Chlorpyriphos production	This Eco-invent 2.2 module calculates the impacts of organophosphorus insecticide production	This Eco-invent process is used as a proxy as specific data set for Indian conditions is not available
Bavistin production	This Eco-invent 2.2 module calculates the impacts of benzimidazole production	This Eco-invent process is used as a proxy as specific data set for Indian conditions is not available

Table A12. Inputs of pesticide application module

Parameter	Value	Unit	Documentation	Source
Fungicide	0.424	Kg	Amount of fungicide that was used per hectare of plantation per annum (active ingredient at 75%)	Own measured data
Insecticide	0.282	Kg	Amount of insecticide that was used per hectare of plantation per annum active ingredient at 50%	Own measured data


GWP 100 years
Global Warming Potential [kg CO2-Equiv.]
5.36
3.19
2.17
Total
Bavistin Production
Chlorpyriphos production
a


AP
Acidification Potential [kg SO2-Equiv.]
.203
.174
.029
Total
Bavistin Production
Chlorpyriphos production
b



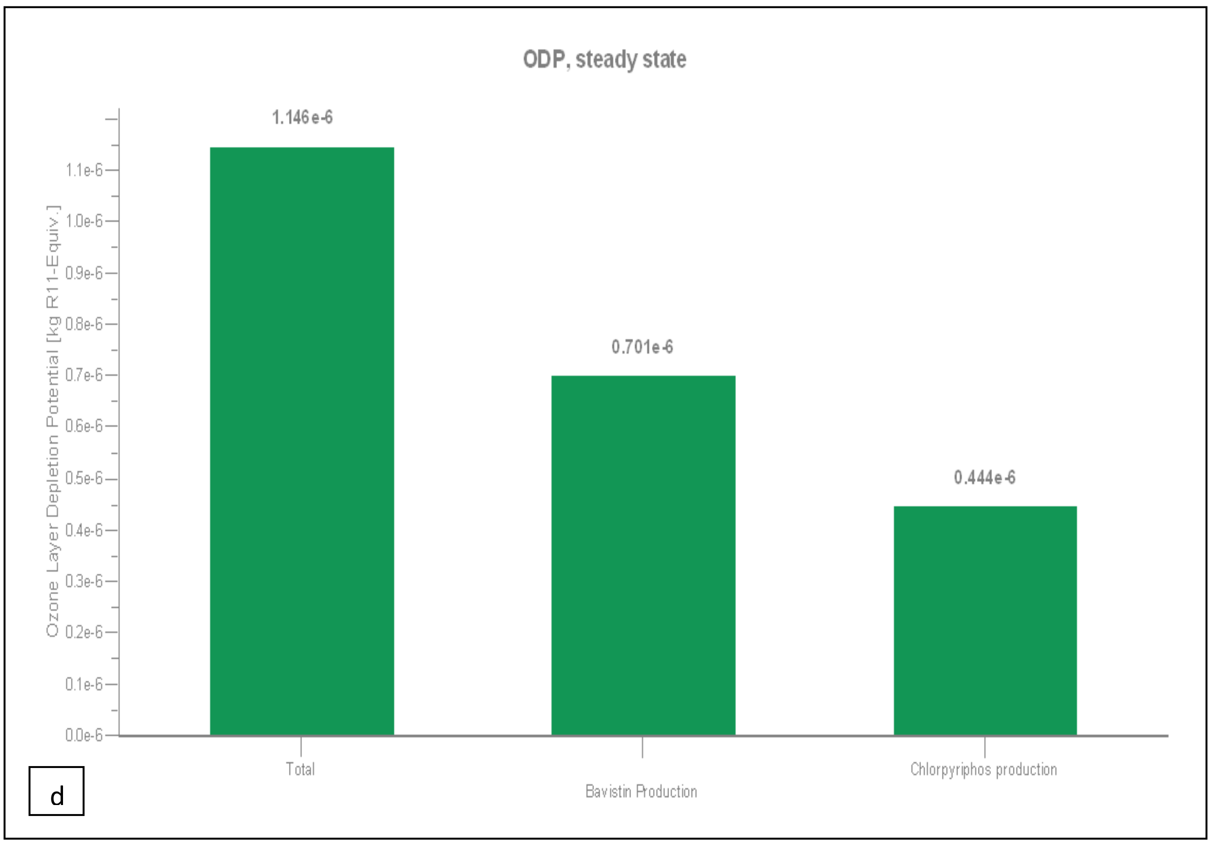


Figure A8. Life cycle impact of Pesticide and its application (1 ha area): a. GWP (Global warming potential); b. AP (Acidification Potential); c. EP (Eutrophication potential) d. ODP (Ozone depletion potential).

Internal farm operations:

This module was developed to calculate the impacts caused by weeding, transportation of fertilizers, produce, agricultural machinery and other miscellaneous items within the field as well as in and out of the field **(Figure C7)**. The assumptions and input values used for modeling land clearing are given in **Table A13 and A14**.

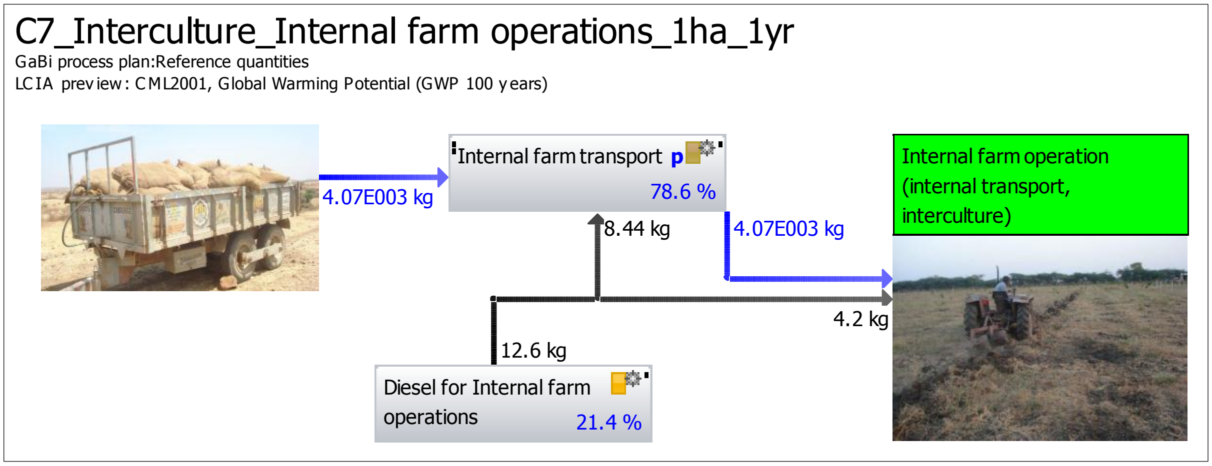


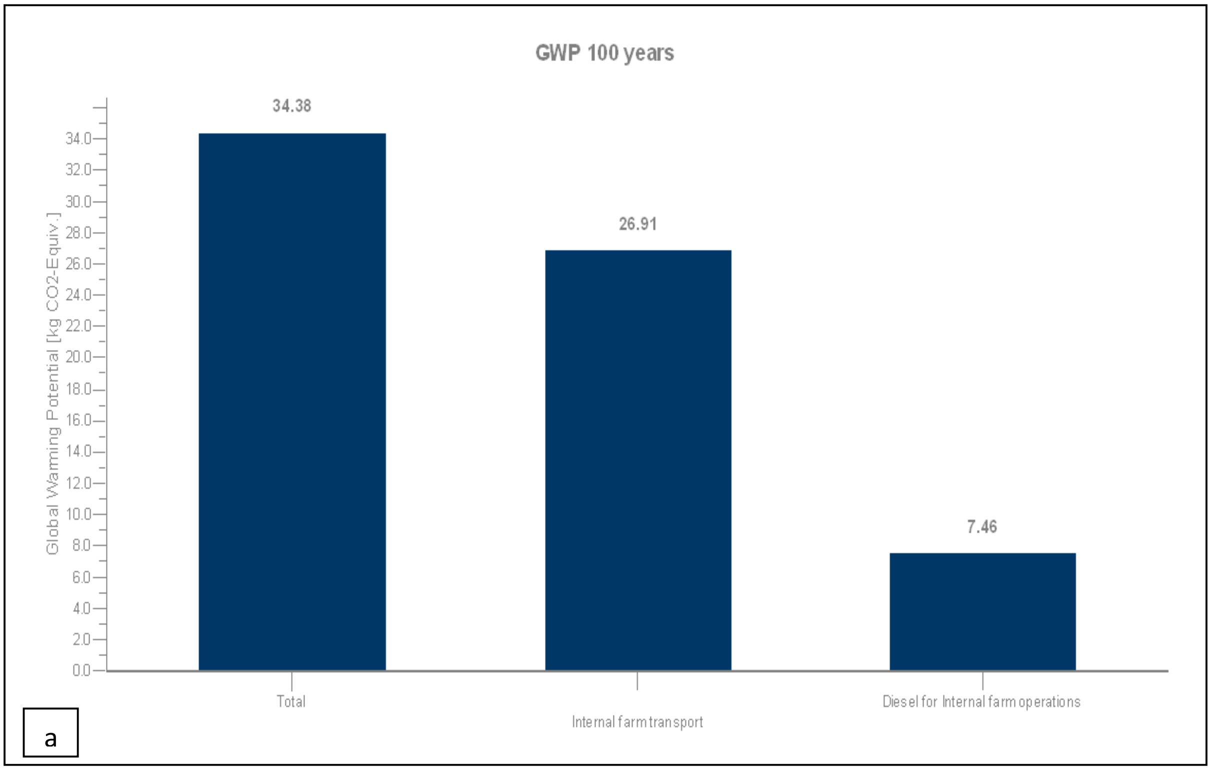
Table A13. GaBi plan for interculture and internal farm operations

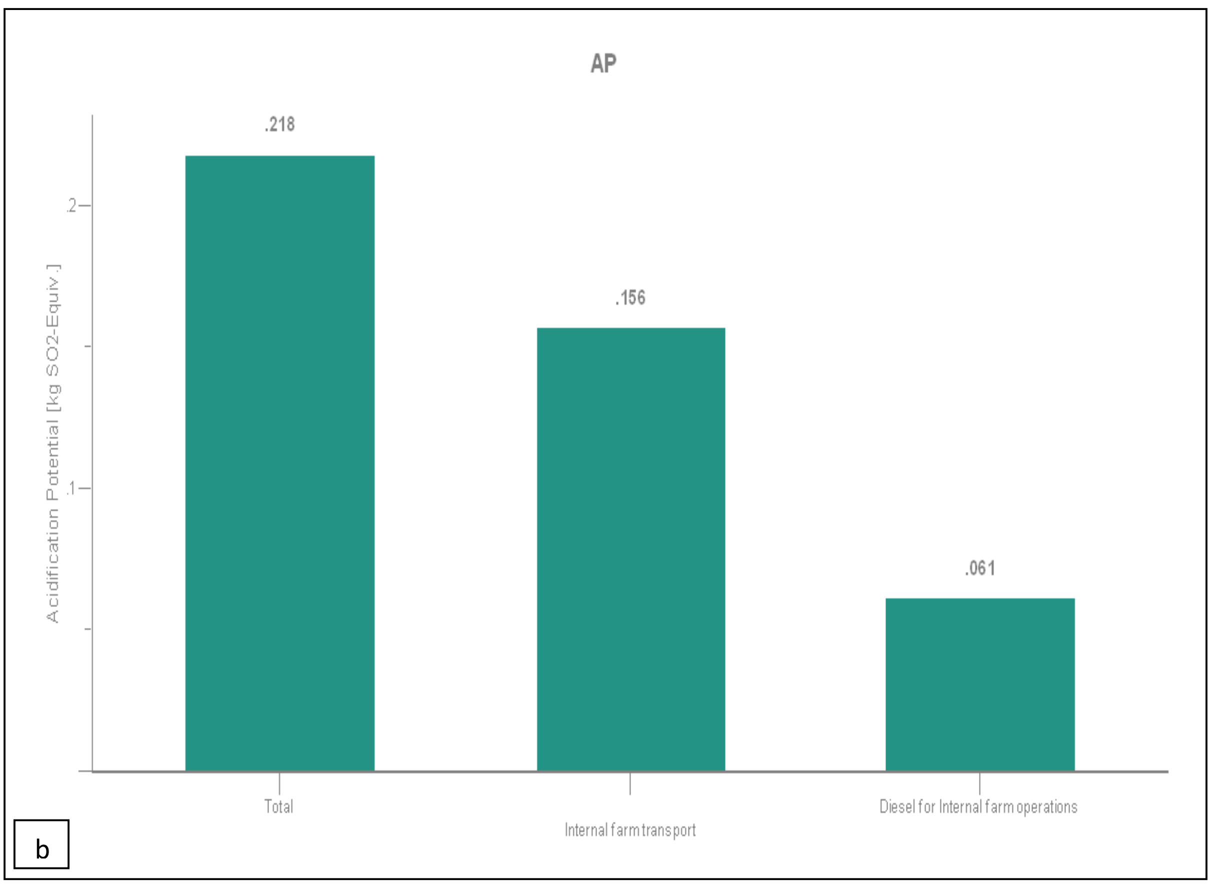
Process name	Module purpose	Comments
Internal farm operations	This custom module calculates the impacts for weeding operations along with transport of fertilizers and other miscellaneous items within and into field	
Transport	PE professional data set calculates impacts of diesel combustion during transport of cleared biomass from the field to the site of utilization	
Diesel	PE professional data set calculates the impacts of upstream crude oil production its transport and refining of diesel at Indian refinery	

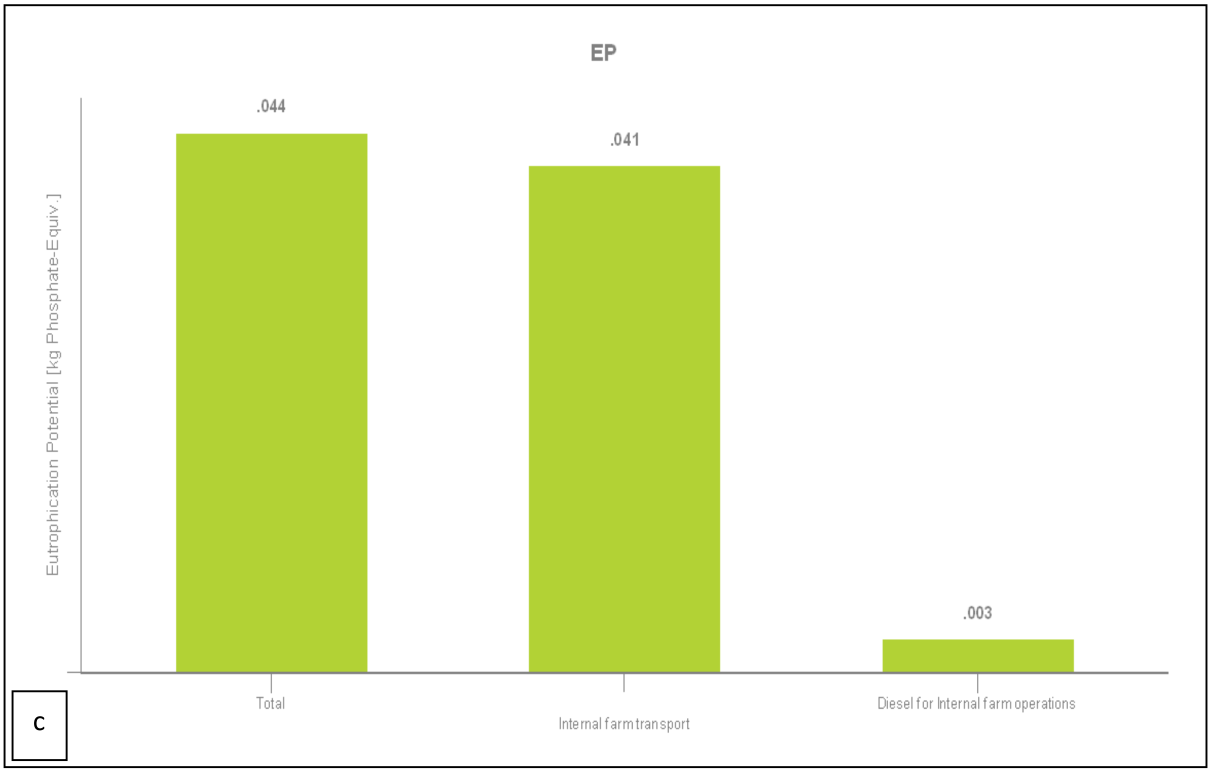
Table A14. Inputs of Interculture and Internal farm operation module

Parameter	Value	Unit	Documentation	Source
Cargo	4066.01	Kg	Total tonnage of fruits, fertilizers and cake transported (assumption)	Own data
Diesel	4.2	Kg	Amount of diesel used for tractor weeding @ 2 weedings per year for 1^{st} four years	Own data

The life cycle impacts across all the categories pertaining to overall land preparation are given in **Figure A9a-d.**


GWP 100 years
Global Warming Potential [kg CO2-Equiv.]
34.38
26.91
7.46
0.0
2.0
4.0
6.0
8.0
10.0
12.0
14.0
16.0
18.0
20.0
22.0
24.0
26.0
28.0
30.0
32.0
34.0
Total
Internal farm transport
Diesel for Internal farm operations
a


AP
Acidification Potential [kg SO2-Equiv.]
.218
.156
.061
.1
.2
Total
Internal farm transport
Diesel for Internal farm operations
b



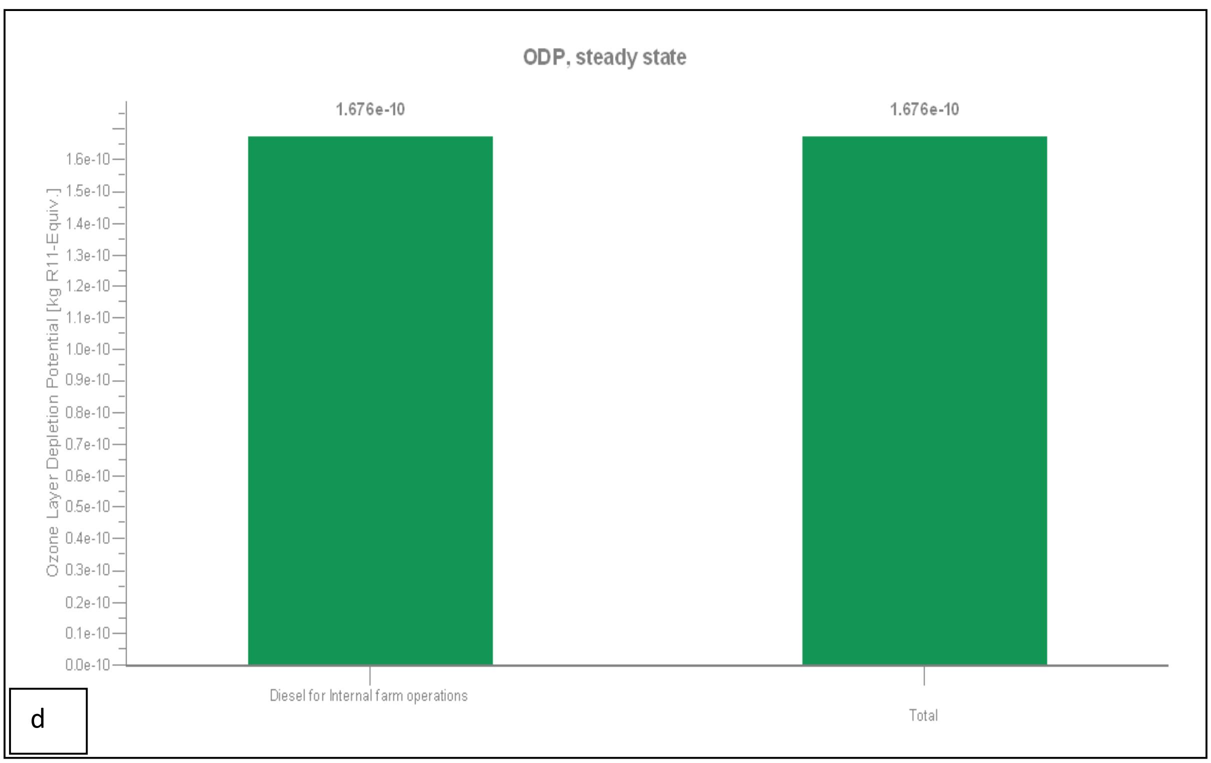
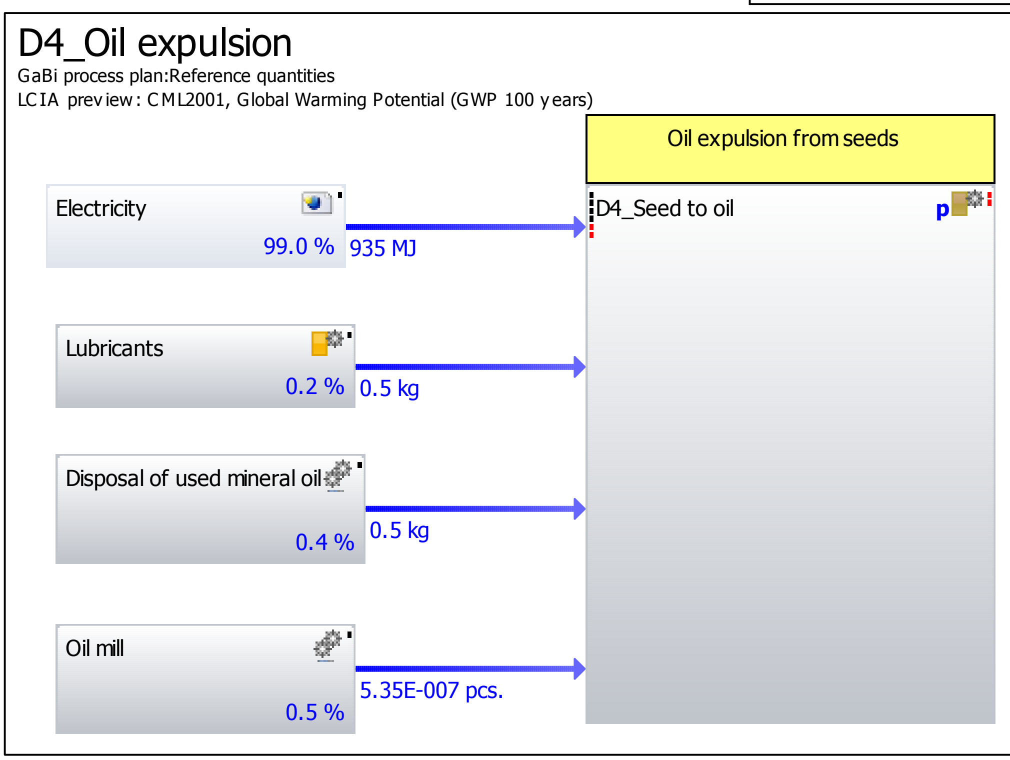


Figure A9. Life cycle impact of land preparation (1 ha area): a. GWP (Global warming potential); b. AP (Acidification Potential); c. EP (Eutrophication potential) d. ODP (Ozone depletion potential.

Jatropha Oil expulsion module

The plan describes the impacts resulting from expulsion of 510 kg of oil from 2.04 tonnes of *Jatropha* seeds which is obtained from 1 hectare of wastelands annually **(Figure D4)**. *Jatropha* de-oiled cake is obtained as a by-product, a part of which was put back for use in the plantation. Steam required for the process was also obtained by burning a portion of the shells (100 kg) obtained during dehusking step (data not separately shown but has been calculated in detail). The assumptions and input values used for modeling land clearing are given in **Table A15 & A16.**

Figure A10. Expulsion of oil from *Jatropha* seeds and generation of oil cake in a unit



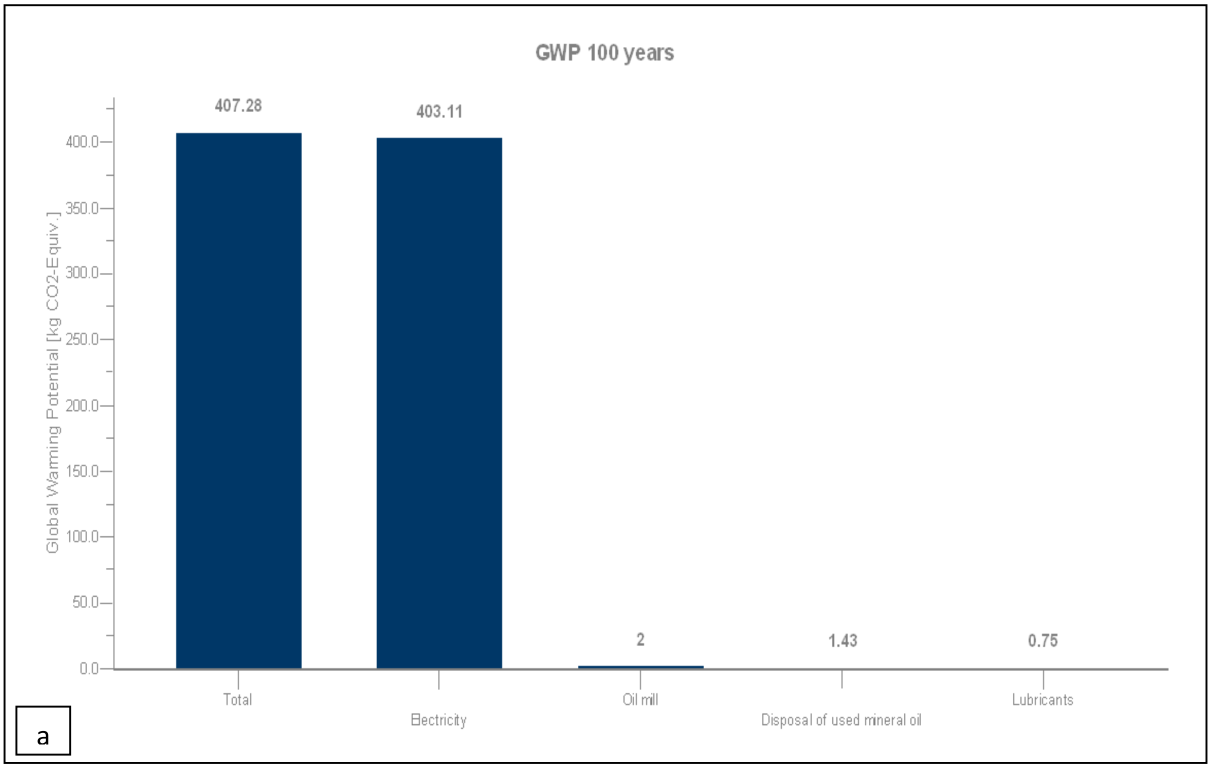
The life cycle impacts across all the categories pertaining to overall land preparation are given in **Figure A11a-d.**

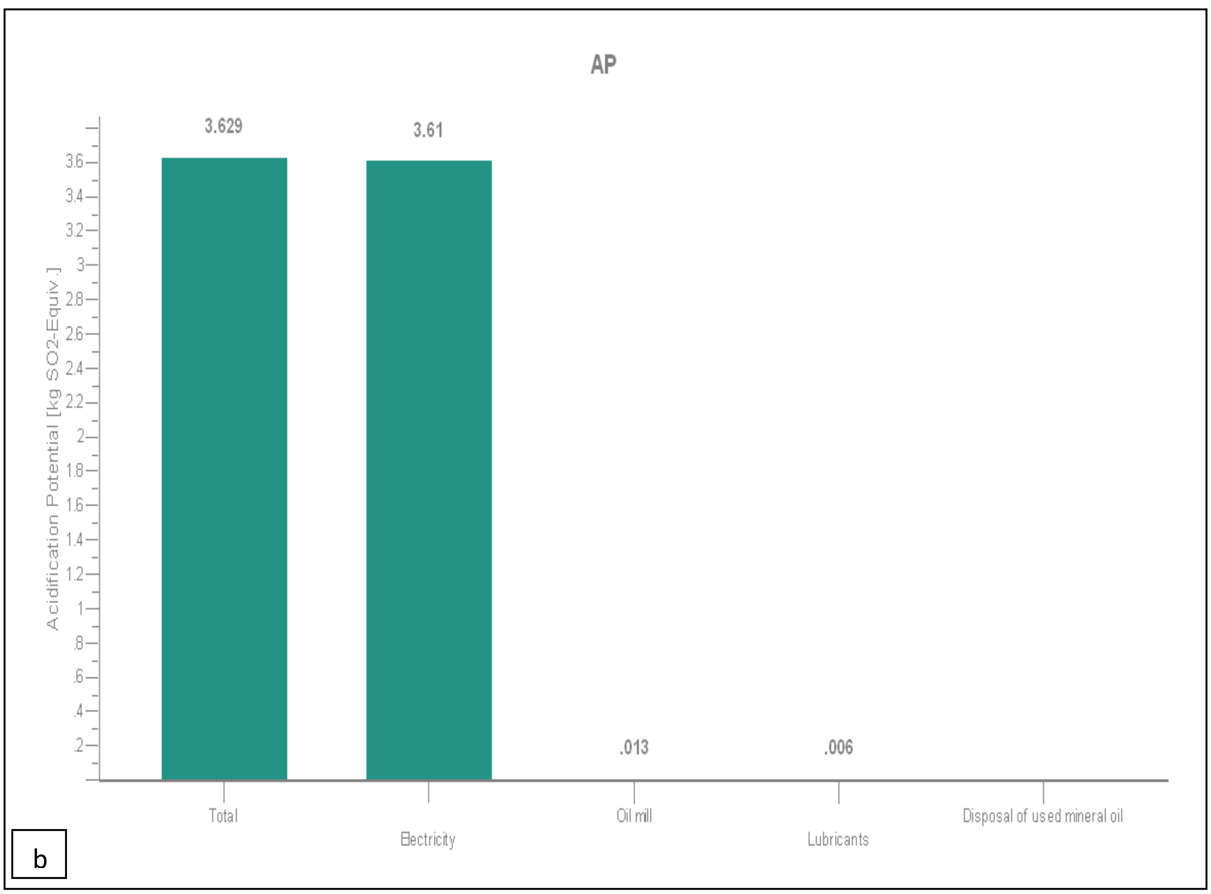
Table A15. GaBi plan for Oil expulsion

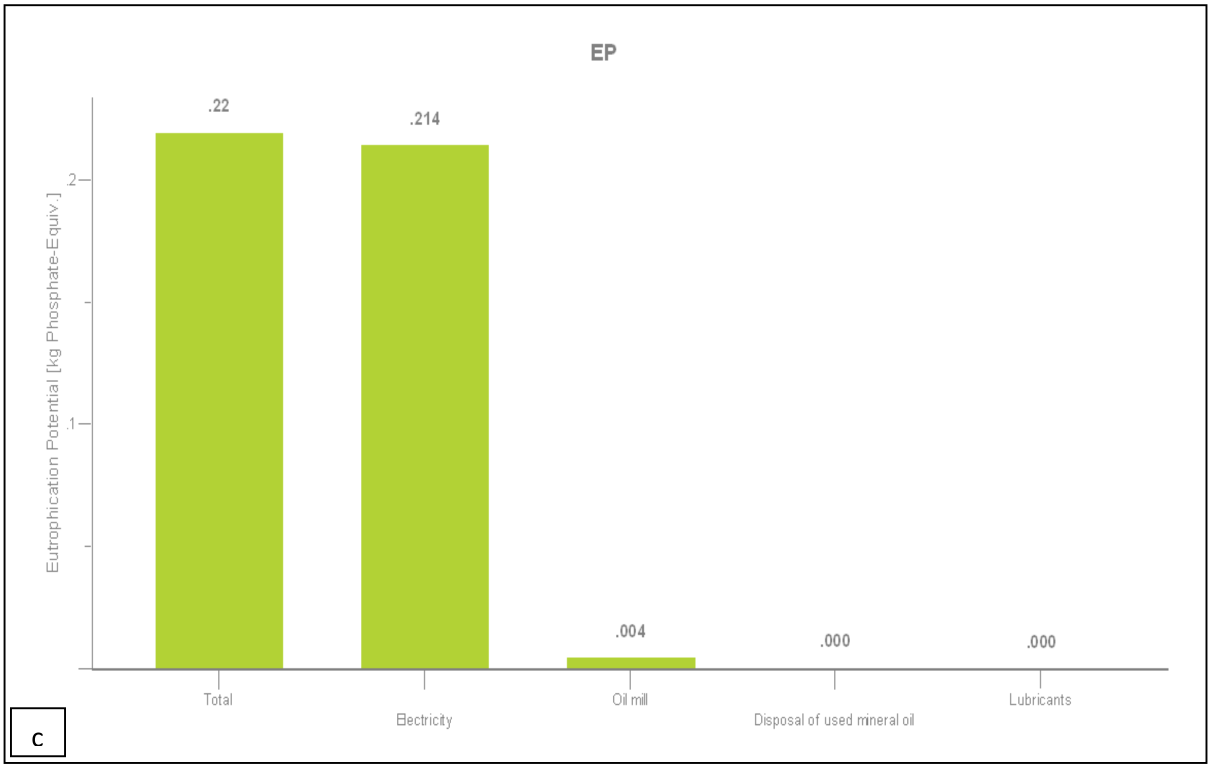
Process name	Module purpose	Comments
Seed to Oil	This custom module calculates the impacts resulting from expulsion of oil from *Jatropha* seeds obtained from one hectare	
Electricity	This PE international module calculates the impacts of generation of electricity under Indian conditions.	The module also includes transmission and distribution losses
Lubricants	PE professional data set calculates impacts for lubricant production at Indian refinery	
Disposal of mineral oil	This Eco-invent 2.2 process calculates the impacts during disposal of lubricating oil in municipal incinerator	This Eco-invent process is used as a proxy as specific data set for Indian conditions is not available
Transport	This customized module calculates the impacts of transporting seeds to oil expulsion unit	The Eco-invent module of transport using 20-28 t fleet average was used for the purpose of determining the impact of emissions. This customized module uses Indian data set for diesel
Oil mill	This Eco-invent 2.2 process calculates the impacts for establishment of oil mill	This Eco-invent process is used as a proxy as specific data set for Indian conditions is not available

Table A16. Inputs of Oil expulsion module

Parameter	Value	Unit	Documentation	Source
Jatropha seeds	2040	Kg	Amount of seeds that is produced from one hectare of *Jatropha* plantation	Own data
Electricity	934.58	MJ	Amount of electricity consumed by screw press expeller of 25HP, metering pump of 3HP and conveyer of 1 HP	Own data
Lubricants	0.5	Kg	Amount of lubricant required for 510 kg oil expulsion	Own data
Disposal of mineral oil to municipal incineration	0.5	Kg	Amount of lubricant that is subject to disposal in municipal incinerator	Own data
Oil mill	0.00000535	No. of pieces		Estimated
Steam	480	Kg	Amount of steam required for production of 510 kg of oil. The steam is generated by burning of *Jatropha* shells obtained after deshelling/decorticating *Jatropha* fruits.	Own data
Oil content	0.25		Assumption of 25% extractable oil from seeds	Own data


GWP 100 years
Global Warming Potential [kg CO2-Equiv.]
407.28
403.11
2
1.43
0.75
Total
Electricity
Oil mill
Disposal of used mineral oil
Lubricants
a


AP
Acidification Potential [kg SO2-Equiv.]
3.629
3.61
.013
.006
Total
Electricity
Oil mill
Lubricants
Disposal of used mineral oil
b



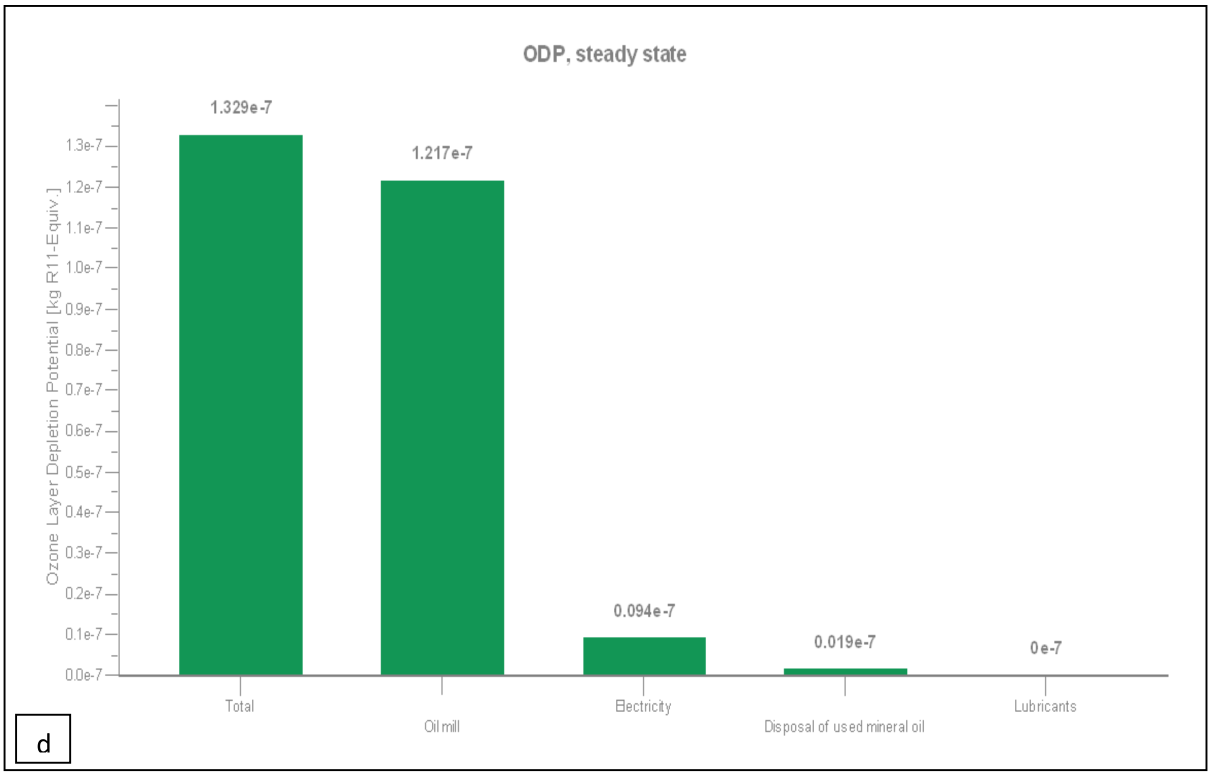


Figure A11. Life cycle impact of oil expulsion from seeds of 1 ha area: a. GWP (Global warming potential); b. AP (Acidification Potential); c. EP (Eutrophication potential) d. ODP (Ozone depletion potential.

Decorticating module

The plan describes the impacts resulting from de-shelling of *Jatropha* fruits (ca 3 t/ha/annum) to produce 2 t of seeds **(Figure D1)**. The processes machinery, lubricant and electricity have been described earlier and hence not tabulated in module description. The assumptions and input values used for modeling decortications of fruits are given in **Table A17**.

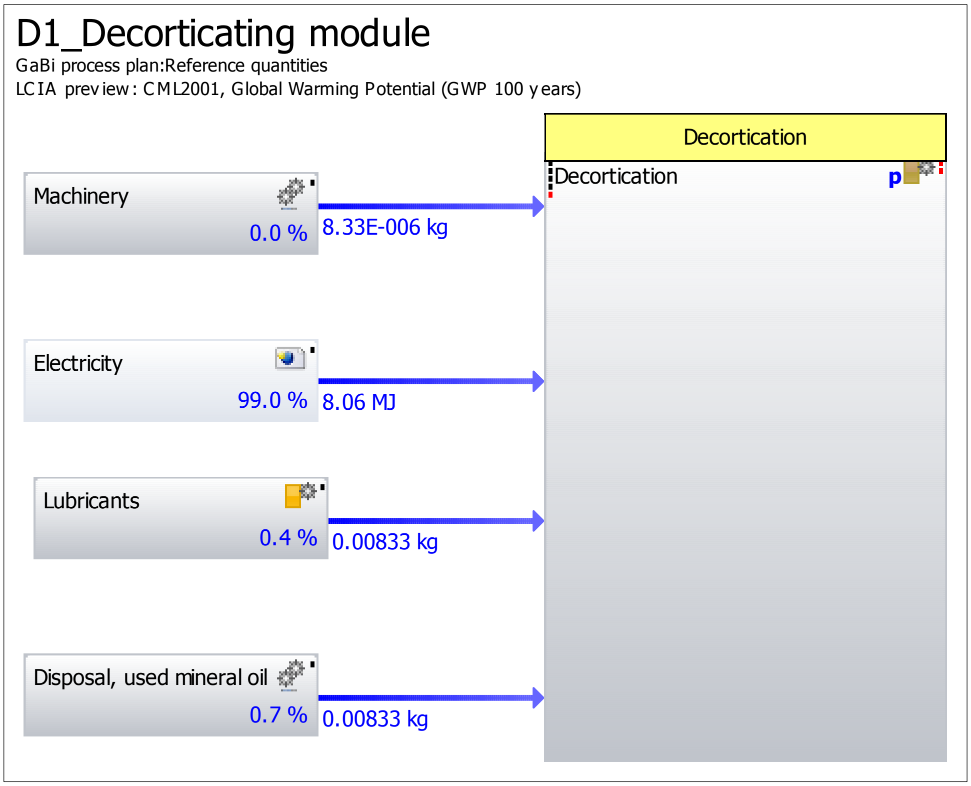


Table A17. Inputs of Decorticating module

Parameter	Value	Unit	Documentation	Source
Machinery	0.0000507	No. of pieces	The number of fruits processed by a 200 kg/unit machine is assumed 200Lakhs kg(Assuming 500 kg fruit per hour and 300 days/yr and 20 years life time)	Own data
Electricity	49.06191006	MJ	Amount of electricity required to run the deshelling machine	Own data
Lubricant	0.05072597	Kg	Amount of lubricant required for deshelling 3044.776 kg of fruit	Own data
Disposal of mineral oil	0.05072597	Kg	Amount of lubricant that is subjected to disposal after use	Own data

The electricity consumed accounted for 99% of GWP (3.52 kg-Equiv CO_2) and likewise, it contributed the maximum impact across all other LC impact categories (data not shown).

Briquetting module:

The plan describes the impacts resulting from briquetting operation of empty *Jatropha* capsules (or shells) after taking out the seeds from the whole fruits **(Figure D2)**. From ~3 t fruits, ~2t of seeds are obtained and remaining ~1t left out is *Jatropha* fruit shells that have calorific value comparable to coal (ca. 3900 Kcal/kg). A small part of *Jatropha* shells is directly used to generate steam, while remaining part is briquetted for further use as solid fuel. The processes machinery, electricity have been described earlier and hence not tabulated in module description. The assumptions and input values used for modeling land clearing are given in **Table A18**.

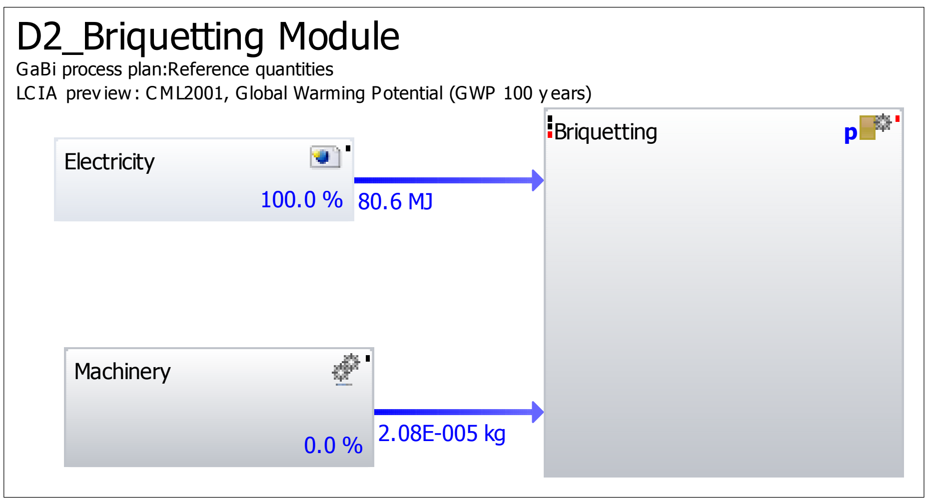


Table A18. Inputs of Briquetting module

Parameter	Value	Unit	Documentation	Source
Machinery	0.000038	No. of pieces	Ratio of processing 903 kg shell to the total shell processed by the machine during its entire life time which is assumed to be 240 Lakhs kg of shells processed.	Own data
Electricity	145.50	MJ	Amount of electricity consumed by the briquetting machine for briquetting 903 kg shells	Own data
Jatropha shells	903	Kg	Portion of shells obtained from one hectare of plantation for briquetting following partitioning for steam generation	Own data

Almost entire contribution to life cycle impact across all the categories studied here is contributed by electricity (data not separately shown). The relevant data is shown in aggregate product plan where contributions from all the component modules are depicted.

Steam generation module:

The module developed to calculate the various impacts resulting from steam generation from shells obtained from one hectare of plantation (102 kg) for use during oil extraction. The various sub processes and the inputs for the module are described in Table A19 and A20 respectively.

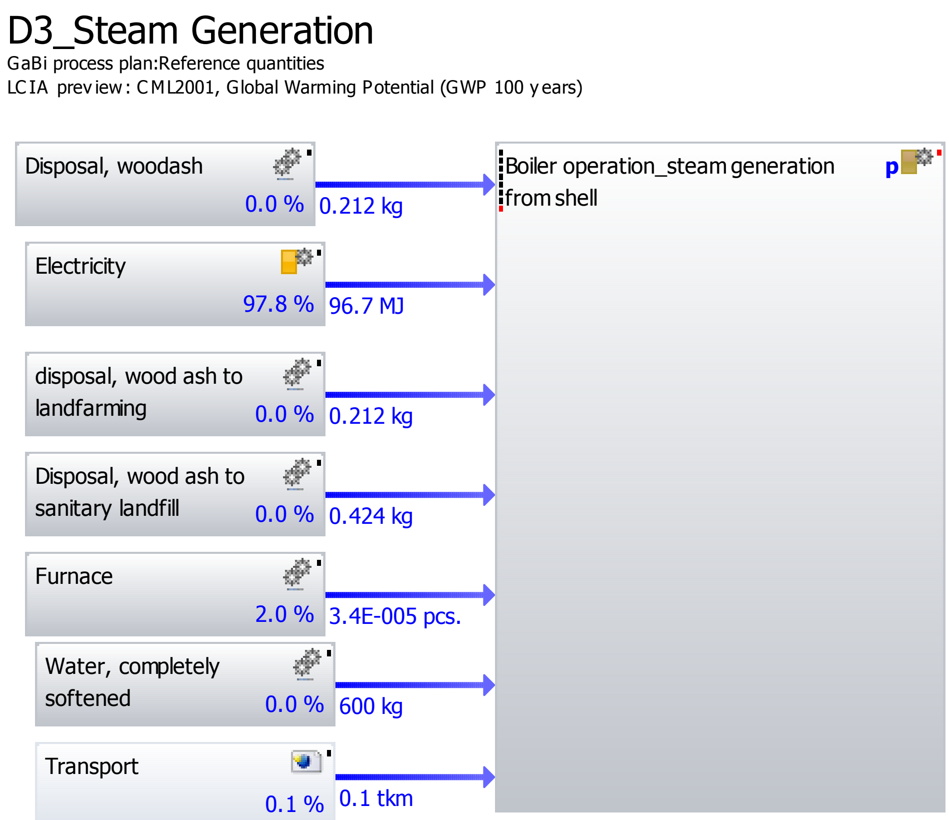


Table A19 Gabi plan for Steam generation module

Process name	Module purpose	Comments
Steam generation	This custom module aggregates and calculates the impacts of all the sub processes of the module	Based on a similar Eco-invent module for steam generation
Electricity	This PE international module calculates the impacts of generation of electricity under Indian conditions.	The module also includes transmission and distribution losses
Furnace	This Eco-invent 2.2 module calculates the impacts of production of the furnace for the requisite number of pieces	This Eco-invent process is used as a proxy as specific data set for Indian conditions is not available
Water, softened	This Eco-invent 2.2 module calculates the impacts of softening water at plant	This Eco-invent process is used as a proxy as specific data set for Indian conditions is not available
Transport	This customized module calculates the impacts of transporting shells from briquetting unit to steam generation unit	The Eco-invent module of transport using 20-28 t fleet average was used for the purpose of determining the impact of emissions. This customized module uses Indian data set for diesel
Disposal wood ash to municipal incineration	This Eco-invent 2.2 process calculates the impacts of the disposal of wood ash during municipal incineration	This Eco-invent process is used as a proxy as specific data set for Indian conditions is not available
Disposal wood ash to land farming	This Eco-invent 2.2 process calculates the impacts of the disposal of wood ash to land farming	This Eco-invent process is used as a proxy as specific data set for Indian conditions is not available
Disposal wood ash to sanitary land fill	This Eco-invent 2.2 process calculates the impacts of the disposal of wood ash to sanitary land fill	This Eco-invent process is used as a proxy as specific data set for Indian conditions is not available

Table A20 Inputs of Steam generation module

Parameter	Value	Unit	Documentation	Source
Jatropha shells	102	Kg	Amount of shells used as a source of energy for generating 490 kg steam	Own data
Furnace	0.0000347	No. of pieces	Number of pieces of a 300KW furnace (assumption) required for generating 490 kg of steam	
Electricity	98.615232	MJ	High pressure pump of rated power=3 HP operated for 12 hours to pump the water to softener and then to boiler against boiler pressure; 3x0.746x12	
Water, softened	612	Kg	Amount of water necessary to generate 490 kg of steam	Own data
Transport	0.102	tkm		Own data
Disposal wood ash to municipal incineration	0.216460812	Kg	Value represents disposal of 0.85% of ash generation from wood chips to municipal incineration	Eco-invent
Disposal wood ash to land farming	0.216460812	Kg	Value represents disposal of 0.85% of ash generation from wood chips to land farming	Eco-invent
Disposal wood ash to sanitary land fill	0.432816248	Kg	Value represents disposal of 1.7% of ash generation from wood chips to sanitary land fill	Eco-invent

Almost entire contribution to life cycle impact across all the categories studied here is contributed by electricity (data not separately shown).