



High oleic peanuts for Asia and Africa to meet the needs of the food processing industries

Janila P.¹, Radhakrishnan T.², Murali TV., Bera SK.³, Dobariya KL.⁴, Nigam SN., Varshney RK.⁵, Pandey MK.⁶, Manohar SS.⁷, Manivannan N.⁸, Vasanthi RP⁹

1. Abstract

High oleic peanuts offer longer shelf-life benefits to food processing industry, health benefits to consumers and increases profitability to farmers through premium price compared to normal peanuts. International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) in collaboration with national partners of India has developed high oleic peanuts in Spanish and Virginia Bunch growth habit suitable for cultivation in Asia and Africa. The oleic acid concentration in high oleic peanuts is 80+2% as against 45-50% in normal peanuts. The high oleic lines were developed using SunOleic 95R as donor parent from the USA employing marker-assisted selection (MAS) and marker-assisted backcrossing (MABC) approaches. Process innovation in breeding and testing pipeline that include, high through phenotyping using Near Infrared Reflectance Spectroscopy (NIRS), genotyping, rapid generation advancement under controlled conditions, target site testing to fix the best allele combinations and multi-location testing resulted in enhanced rate of genetic gain for high oleic trait. ICRISAT has shared high oleic peanut lines with national partners in India, Uganda, Tanzania, Mali, Malawi, Ethiopia, Bangladesh, Myanmar and Australia.

High oleic lines are in national trials under All India Co-ordinated Research Project on Peanut (AICRP-G) during rainy 2017 and 2018 and this is the first such speciality trial being conducted in India and it is expected that India will release its first high oleic peanut variety in 2019. Fast-track development and commercialization of high oleic varieties in India was enabled through partnerships.

¹ International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Hyderabad, India - p.janila@cgiar.org

² Directorate of Groundnut Research (DGR) of Indian Council of Agricultural Research (ICAR), Junagadh, India

³ International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Hyderabad, India

⁴ Junagadh Agricultural University (JAU), Junagadh, India

⁵ International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Hyderabad, India

⁶ International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Hyderabad, India

⁷ International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Hyderabad, India

⁸ Tamil Nadu Agricultural University (TNAU), Coimbatore, India

⁹ Acharya NG Ranga Agricultural University (ANGRAU), Tirupati, India

2. Context and challenge, including key interactions (range and nature) the case study addresses

Peanut oil and peanut based food products with the high-oleic content have five to 10 times longer shelf life than that of normal peanut. A diet with high oleic acid and low palmitic acid, is an exceptional way to reduce the risk of cardiovascular diseases, promotes a healthier ratio of high density lipoprotein (HDL) to low density lipoprotein (LDL), and reduces triacylglycerol and blood glucose levels. Peanut oil contains eight fatty acids, the two main fatty acids *i.e.* oleic and linoleic acids which are mono- and poly-unsaturated fatty acids, respectively are present in ~2:1 ratio in normal peanuts and together these two fatty acids contribute about 80-85% of the total fatty acids (Moore and Knauff, 1989). Genetically, peanuts with the high-oleic trait lack functional fatty acid desaturase (FAD) genes that encode the enzyme for conversion of oleic to linoleic acid. Because peanut is an allotetraploid species, there are two such homeologous gene sequences (*FAD2A* and *FAD2B*) in the two progenitor species genomes. Mutations in both genes are required for the high oleic accumulation in peanut kernels.

In 1987, Norden and co-workers naturally identified the first high-oleate peanut mutant line, F435 with about 80% oleic acid and 2% linoleic acid. The first high oleate peanut variety, SunOleic 95R was bred in USA through conventional breeding (Gorbet and Knauff, 1997). So far, high-oleic peanut cultivars are registered and are under cultivation in the USA, Australia, Argentina and Brazil, with Australia having 100% high oleic peanuts.

A sound and fast track breeding strategy that involved genotyping at early generation using molecular markers, phenotyping at later generations for recurrent parent phenotype and high oleic trait, rapid generation advancement in glasshouse to increase the number of cycles per year resulted in development of agronomically superior high oleic peanut lines within a short span of five years. The non-destructive and robust phenotyping using Near-Infrared reflectance spectroscopy (NIRS) (Figure 1) for assessing oil chemistry was quite helpful as it enabled screening a large number of populations within a relatively short time. For using NIRS, calibration equation was developed for palmitic, oleic and linoleic acid and the RSO and 1-VR values for the equation is depicted in Table 1. The absorption spectrum of NIRS for two samples differing in their oleic acid contents is depicted in Figure 2. For developing the calibration equation for oil quality- the oleic, linoleic and palmitic acid content in 208 F2:3 population of the cross ICGV 06420 × SunOleic95R was estimated using gas chromatography (Figure 3). The use of NIRS for screening samples for oil and fatty acid content in whole peanut kernels has greatly bolstered the selection efficiency at ICRISAT. It enabled considerable saving of time and resources, as we are able to analyse 150-200 samples in a day and select samples based on trait preference.

Table 1. Calibration equations for predicting oil, protein and fatty acids (palmitic, oleic and linoleic acid) content in whole peanut kernels using NIRS

Constituent	N*	Mean	Range	Maths treatment	RSQ**	1-VR#
Palmitic acid (%)	208	11.42	6.77-16.06	2,4,4,1	0.88	0.80
Oleic acid (%)	208	52.12	23.44-80.79	2,4,4,1	0.97	0.95
Linoleic acid (%)	208	27.12	2.77-51.46	2,4,4,1	0.97	0.95

*N- Number of samples in calibration; **RSQ- coefficient of determination in calibrations; #1-VR- coefficient of determination in cross-validation



Figure 1. NIRS -low-cost, non-destructive, robust phenotyping technique for assessing oil chemistry.



Figure 2. The raw absorption spectra of peanut kernels representing two extreme oleic acid values

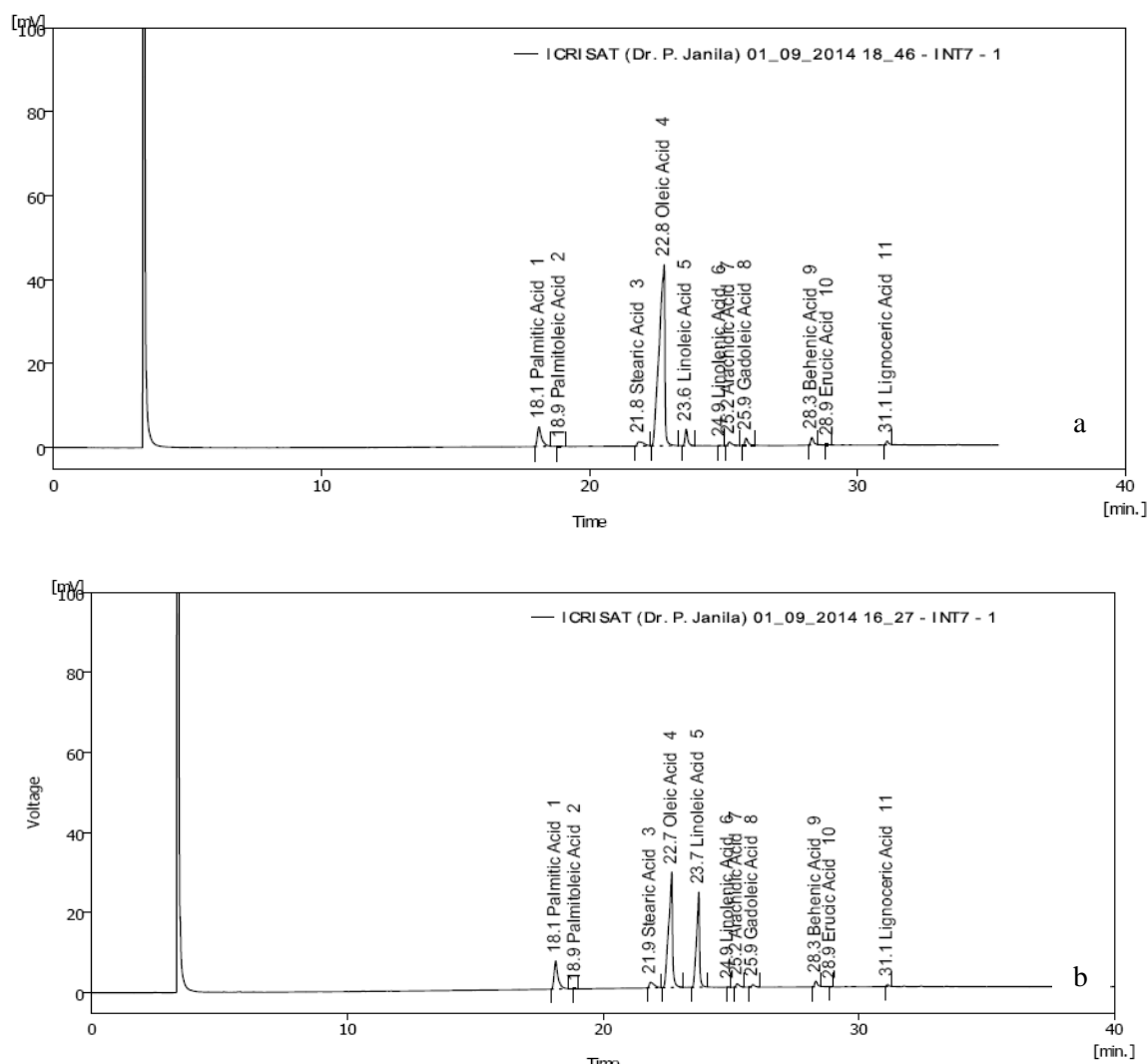


Figure 3. Gas chromatography peaks depicting the fatty acid profile of peanut derived from (a) SunOleic 95R and (b) Recurrent parent

Two elite high oil lines, ICGV 06420 and ICGV 06142 and two low oil confectionary type, ICGV 06110 and ICGV 07368 were used as recurrent parents in the MABC and MAS program and SunOleic 95R was the donor parent for the high oleic trait. A cost effective three-step strategy was developed and successfully used to identify heterozygotes and homozygotes for *fad2A* and *fad2B* mutant alleles in different generations using allele specific and CAPS (Cleaved Amplified Polymorphic Sequence) markers. In later generations, NIRS was used to estimate fatty acid profile. Selected lines with high oleic acid content (>70% by NIRS) were consequently evaluated at ICRISAT, following which 65 lines were identified and proposed for multi-location evaluation (Fig 4) (Janila et al 2016). Of which, 16 lines were proposed for AICRP-G (All India Co-ordinated Research Project on Groundnut) testing. Four lines- ICGVs 15073, 15074, 15080 and 15083 showed stable performance across multiple locations and are probable candidates for release.

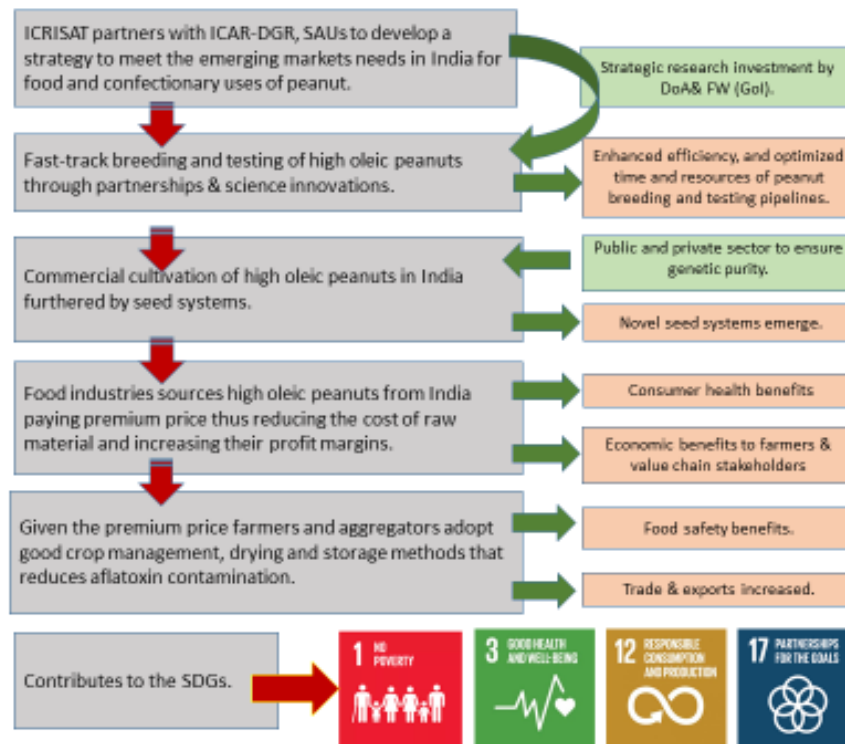


Figure 4. Evaluation trial of high oleic lines at ICRISAT during 2015 rainy season

3. How did research efforts deal with the synergies and trade-offs?

a) in the development of the TOC and impact pathways

High oleic peanuts have high demand in both domestic and international markets for enhanced shelf-life and consumer health benefits. As high oleic peanuts are not cultivated in India, food industries source high oleic peanuts from Australia which increases the cost of raw material and consequently lowers the profit margins to the industries and increases cost of the product to the consumers. Food industries such as, MARS and Mondelez International use high oleic peanuts in their food products and are looking at the opportunities of sourcing high oleic peanut within India for their processing plants based in India. They are willing to source high oleic peanuts at a premium price thus contributing to enhanced profitability to the farmers and all the stake holders in the high oleic peanut value chain. Premium price to the high oleic encourages the farmers and aggregators to adopt good crop production, drying and storage practices that contributes to reduced aflatoxin contamination, which is a food safety concern and trade barrier. The food industries sourcing high oleic peanut expect 95-98% purity of high oleic peanuts in the commodity and this is expected to promote seed systems to ensure supply of seed of high oleic varieties with highest genetic purity to ensure high standards of the commodity. Thus, a more organized sustainable seed supply chain with public and private seed agencies is expected to emerge for high oleic peanuts in India. A notable interest for high oleic peanut varieties has already been perceived from the seed agencies and food processing industries in India and they are keen to work with ICRISAT and our partners to bring high oleic into supply chain in a fast-track mode. Besides significant economic benefits to high oleic peanut producers and value chain actors, at least two other outcomes are evident from the synergies, one is development of seed systems to ensure genetic purity, and the other is production of food safe peanut produce. Thus, the TOC is increased farm incomes of groundnut producers in India and reduced poverty with an enhanced access to safety food.



Schematic representation of the process and anticipated outcomes.

b) in the development of partnerships/delivery approaches

ICRISAT engaged with ICAR-Directorate of Groundnut Research (ICAR-DGR) and State Agricultural Universities (SAUs) through the conception and development of the strategy to meet the market needs. With a clear delivery strategy, ICRISAT along with ICAR-DGR and SAUs, approached the Department of Agriculture, Cooperation and Farmer Development (DoAC&FD) of Government of India to make a strategic investment for the development and commercialization of high oleic peanuts in India. The two key elements that met the interests of the donor to extend financial assistance are (a) emerging domestic market demand for high oleic peanuts in India, and (b) increased use of peanuts for food and confectionary in India.

The partnership was built to leverage research and infrastructure capacities of partner institutes like NIRS phenotyping, genotyping and rapid generation advancement at ICRISAT, target site testing and multi-location testing of ICAR-DGR and SAU's, and national testing coordinated by ICAR-DGR. As fast-track delivery was envisaged the complementary capacities of the partnering teams was critical to bring high oleic peanut for commercialization in a span of 8 years. The partner centers did a good job of evaluating the lines and proposing the best bet entries were recommended for national testing under AICRP-G. Learning from the experience of USA, we began engaging with private and public seed producers for delivery of high oleic seed of high genetic purity once the high oleic varieties are commercialized. The engagement through field days and media helped to generate interest among seed agencies and some seed agencies like National Seeds Corporation (NSC) have approached us to become part of seed supply chain of high oleic peanuts in India.

c) in the development of metrics

As high oleic is a new trait and there is a perceived need to further the breeding and testing pipelines in terms of number of crosses attempted, and populations developed and tested. At ICRISAT itself ca. 100 crosses were attempted so far. Starting from 64 lines that were bred at ICRISAT and given for testing there are over 200 elite lines from ICRISAT, ICAR-DGR and SAUs are under different stages of testing. The complementary support for phenotyping and genotyping and collective multi-location testing resulted in selection of good number of lines from different breeding program for testing.

d) other

ICRISAT develops International Public Goods (IPGs). Higher rate of returns on the investments is possible through sharing of the IPGs, high oleic peanut lines in this case with partners. The networks of ICRISAT nurtured over several decades enabled the NARS from different countries to access the high oleic peanut lines from ICRISAT. A total of 208 lines were shared of which 93 lines were dispatched to partners in Asia (Bangladesh, Myanmar, and Vietnam) and 115 lines to Africa (Ethiopia, Nigeria, Mali, Malawi, Tanzania and Uganda) locations for evaluating at their respective locations and four lines were shared with Australia.

4. What kinds of partnerships were critical?

- Partnership with ICAR-DGR, SAUs in the design, testing and delivery has resulted in success in a short period as well as achieve the metrics.
- Partnership with industry helped us to get the sensory and product suitability feedback so that the right product is commercialized.
- Partnership with public and private seed producers is giving us confidence to scale-up seed chain when the lines are commercialized in 2019.
- Donor engagement was important to sustain the financial assistance from 2011 to 2018.
- Leveraging the complementarities of the partners rather than duplicating the efforts was important.

5. Lessons learnt, including knowledge gaps and good practices in employing these approaches at scale

- Identifying right partners with complimenting capacities to achieve the objective of developing high oleic lines in peanut.
- Engaging with partners from design, development, testing and commercialization.
- Partners involved at all stages of the development and giving them due credit for the successful implementation.
- Extensive coverage of activities and success stories in social and mainstream media.
- Identify the gaps in the research process and deploy key innovations to enhance the rate of genetic gain was critical for fast-track breeding, testing and release.
- Identifying potential bottlenecks in delivery, and engaging with partners to overcome these bottlenecks creates opportunities to new partners. For example engaging with public and private seed agencies to address genetic purity concerns.
- Lessons from the experiences in other countries can be useful after assessing it in the current situation. For example, lesson from USA on genetic purity issues were useful to the group to begin addressing it right from the starting point rather than fixing the problem later.

- Continuous dialogue with all the stakeholder including donors is critical to make progress and realize the research efforts into a commercial product.
- Identify available technologies for adoption that are complimentary to the commercial product will contribute to delivering a cassette of technologies for upscaling. Available production, drying and storage technologies to reduce aflatoxin contamination.

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