Biofortification: A New Tool to Reduce Micronutrient Malnutrition

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Abstract

The density of minerals and vitamins in food staples eaten widely by the poor may be increased either through conventional plant breeding or through use of transgenic techniques, a process known as ***biofortification.*** HarvestPlus seeks to develop and distribute varieties of food staples (rice, wheat, maize, cassava, pearl millet, beans, sweetpotato) which are high in iron, zinc, and provitamin A through an interdisciplinary, global alliance of scientific institutions and implementing agencies in developing and developed countries.

Biofortified crops offer a ***rural-based*** intervention that, by design, initially reaches these more remote populations, which comprise a majority of the undernourished in many countries, and then penetrates to urban populations as production surpluses are marketed. In this way, biofortification complements fortification and supplementation programs, which work best in centralized urban areas and then reach into rural areas with good infrastructure. Initial investments in agricultural research at a central location can generate high recurrent benefits at low cost as adapted biofortified varieties become available in country after country across time at low recurrent costs.

In broad terms, three things must happen for biofortification to be successful. First, the breeding must be successful – high nutrient density must be combined with high yields and high profitability. Second, efficacy must be demonstrated – the micronutrient status of human subjects must be shown to improve when consuming the biofortified varieties as normally eaten. Thus, sufficient nutrients must be retained in processing and cooking and these nutrients must be sufficiently bioavailable. Third, the biofortified crops must be adopted by farmers and consumed by those suffering from micronutrient malnutrition in significant numbers.

Keywords:biofortification, nutrition, micronutrient deficiency, iron, zinc, provitamin A, plant breeding, benefit-cost ratio, bioavailabilty, efficacy, consumer acceptance, farm extension, low income countries

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1. Rationale for Biofortification

Modern agriculture has been largely successful in meeting the energy needs of poor populations in developing countries. In the past 40 years, agricultural research in developing countries has met Malthus's challenge by placing increased cereal production at its center. However, agriculture must now focus on a new paradigm that will not only produce more food, but deliver better quality food as well[1].

Through plant breeding, biofortification can improve the nutritional content of the staple foods poor people already eat, providing a comparatively inexpensive, cost-effective, sustainable, long-term means of delivering more micronutrients to the poor. This approach will not only lower the number of severely malnourished people who require treatment by complementary interventions, but will also help them maintain improved nutritional status. Moreover, biofortification provides a feasible means of reaching malnourished ***rural*** populations who may have limited access to commercially marketed fortified foods and supplements.

Unlike the continual financial outlays required for traditional supplementation and fortification programs, a one-time investment in plant breeding can yield micronutrient-rich plants for farmers to grow around the world for years to come. It is this multiplier aspect of biofortification across time and distance that makes it so cost-effective.

1.1. Comparative Advantages of Biofortification

Reaching the Malnourished in Rural Areas

Poor farmers grow modern varieties of crops developed by agricultural research centers supported by the Consultative Group on International Agricultural Research (CGIAR) and by national public and private agricultural research systems (NARS), and disseminated by non-governmental organizations (NGOs) and government extension agencies. The biofortification strategy seeks to put the micronutrient-dense trait in the most profitable, highest-yielding varieties targeted to farmers and to place these traits in as many released varieties as is feasible. Moreover, as marketed surpluses of these crops make their way into retail outlets, reaching consumers in both rural and urban areas. The direction of the flow, as it were, is from rural to urban in contrast to complementary interventions that begin in urban centers.

Cost-Effectiveness and Low Cost

Biofortified staple foods cannot deliver as high a level of minerals and vitamins per day as supplements or industrially fortified foods, but they can help to bring millions over the threshold from malnourishment to micronutrient sufficiency. Figure 1 shows this potential schematically when a high percentage of the iron-deficient population is relatively mildly deficient. For those who are severely deficient, supplements (the highest cost intervention) are required.

In an analysis of commercial fortification, Horton and Ross (2003) estimate that the present value of each annual case of iron deficiency averted in South Asia is approximately US$20.[2] Consider the value of 1 billion cases of iron deficiency averted in years 16–25 after a biofortification research and development project was initiated (100 million cases averted per year in South Asia). The nominal value of US$20 billion (1 billion cases times a value of US$20 per case) must be discounted because of the lags involved between the time that investments are made in biofortification and when benefits are realized. At a three percent discount rate, the present value would be approximately US$10 billion, and at a 12 percent discount rate, the present value would be approximately US$2 billion. This benefit is far higher than cost of breeding, testing, and disseminating high iron and high zinc varieties of rice and wheat for South Asia (< $100 million in nominal costs).

Sustainability of Biofortification

Once in place, the system described in the previous section is highly sustainable. The major, fixed costs of

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[1] An important part of the overall solution is to improve the productivity of a long list of non-staple food crops. Because of the large number of foods involved, achieving this goal requires a very large investment, the dimensions of which are not addressed here.

[2] A World Bank study (1994) assigns a present value benefit of US$45 to each annual case of iron deficiency averted through fortification (a mix of age-gender groups). The same study gives a present value of US$96 for each annual case of vitamin A deficiency averted for pre-schoolers.

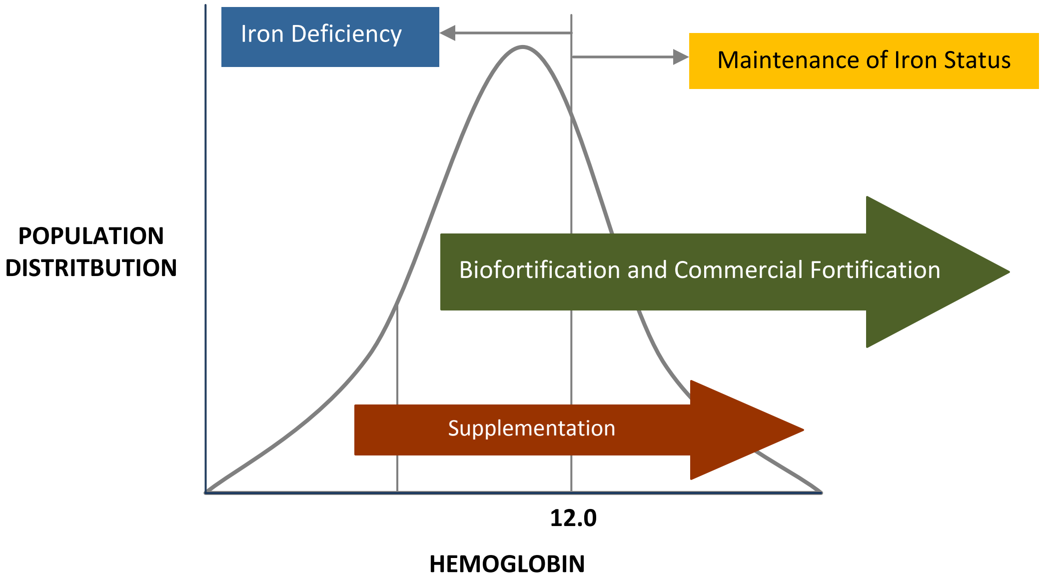


Figure 1. Biofortification improves status for those less deficient and maintains status for all at low cost.

developing the varieties and convincing the nutrition and plant science communities of their importance and effectiveness are being covered by programs such as HarvestPlus (www.harvestplus.org). However, the nutritionally improved varieties will continue to be grown and consumed year after year. To be sure, recurrent expenditures are required for monitoring and maintaining these traits in crops, but these recurrent costs are low compared with the cost of the initial development of the nutritionally improved crops and the establishment, institutionally speaking, of nutrient content as a legitimate breeding objective.

1.2. Limitations of Biofortification

Varying impact throughout the lifecycle

Biofortified staple foods can contribute to body stores of micronutrients such as iron, zinc, and vitamin A (the three target nutrients under HarvestPlus) throughout the lifecycle, including those of children, adolescents, adult women, men, and the elderly. The potential benefits from biofortification are, however, not equivalent

across all of these groups and depend on the amount of staple food consumed, the prevalence of existing micronutrient deficiencies, and the micronutrient requirement as affected by daily losses of micronutrient from the body, and special needs for processes such as growth, pregnancy, and lactation. Some special considerations are noted in the following points.

- During pregnancy, maternal micronutrient needs are substantially increased. For iron, in particular the requirement is so great the additional contribution from biofortification will be low. Additional means of meeting iron requirements during pregnancy will be required. A potentially more significant contribution of biofortification to women's iron status is through improving her iron intake and status before entering pregnancy.

For iron and zinc, improved maternal nutritional status during pregnancy may also lead to increased transfer of iron and zinc to the fetus in late gestation and during birthing. Infants rely on these stores for their iron and zinc requirements during the first 4-6 months of life.

- Breast milk vitamin A concentration decreases as a result of maternal vitamin A deficiency, and maternal consumption of pro-vitamin A biofortified staple foods may help to maintain normal breast milk vitamin A concentrations. Therefore, all breastfed children (up to 6 months of age), particularly those for whom breast milk provides a major source of total energy, may benefit indirectly from biofortification with pro-vitamin A due to increased intake of vitamin A from breast milk.

Maternal iron or zinc status does not affect breast milk content, so maternal consumption of iron- and zinc-biofortified foods is not expected to provide indirect benefits to the breastfed child.

- Children between 6 and 23 months of age are particularly vulnerable to micronutrient deficiencies and are the most gravely affected by their consequences. Children under 24 months of age consume relatively smaller amounts of staple foods, and have relatively higher micronutrient requirements, compared with other age groups. The contribution of industrial fortification or biofortification (which both use food as a vehicle for augmenting nutrition) to the micronutrient adequacy in this vulnerable group will be relatively low in comparison with requirements.

- Due to the particularly high pro-vitamin A content of several orange sweet potato varieties, regular consumption of these varieties can contribute substantially to vitamin A requirements of breastfed children 6-23 months of age.

__Time dimension to deliver biofortified crops and to build up and maintain body stores__

It will take a decade before a first wave of biofortified crops is widely adopted in several developing countries. It is only when this happens and

attributable impact is confirmed as measured by significant reductions in the prevalence of iron, zinc, and vitamin A deficiencies that biofortification will take its place beside supplementation, fortification, and nutrition education as an effective strategy for reducing micronutrient malnutrition.

2. Implementing Biofortification

For biofortification to be successful, three broad questions must be addressed:

- Can breeding increase the micronutrient density in food staples to reach target levels that will make a measurable and significant impact on nutritional status?
- When consumed under controlled conditions, will the extra nutrients bred into the food staples be bioavailable and absorbed at sufficient levels to improve micronutrient status?
- Will farmers grow the biofortified varieties and will consumers buy/eat them in sufficient quantities?

Much of the evidence available to address three questions has been generated under the HarvestPlus Challenge Program. HarvestPlus is an interdisciplinary alliance of research institutions and implementing agencies which is developing biofortified varieties of rice, wheat, maize, cassava, pearl millet, beans, and sweetpotato as shown in Table 1 below.

Table 1. Schedule of product release.

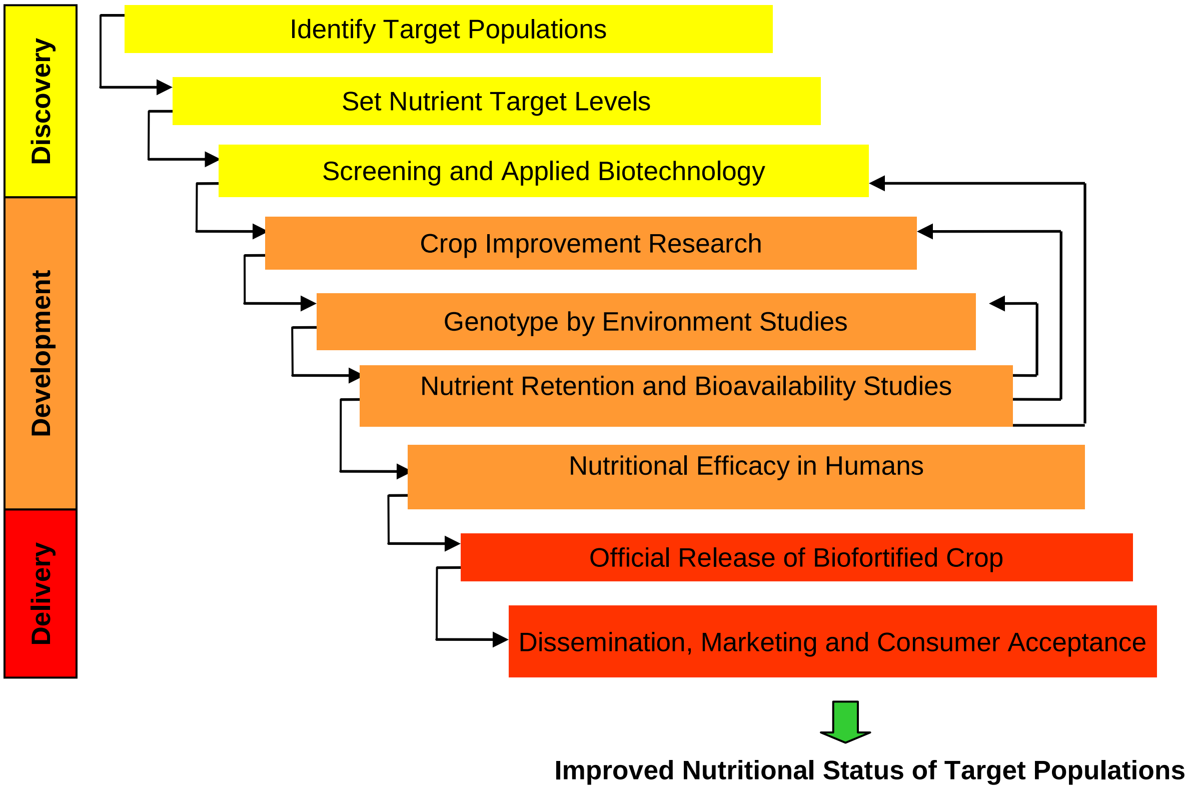
Crop	Nutrient	Countries of first release	Agronomic trait	Release year[a]
Sweetpotato	Pro-vitamin A	Uganda, Mozambique	Disease resistance, Drought tolerance, acid soil tolerance	2007
Bean	Iron, Zinc	Rwanda, DR Congo	Virus resistance, Heat and drought tolerance	2010
Pearl Millet	Iron, Zinc	India	Mildew resistance, Drought tolerance	2011
Cassava	Pro-vitamin A	Nigeria, DR Congo	Disease resistance	2011-12
Maize	Pro-vitamin A	Zambia	Disease resistance, Drought tolerance	2011-12
Rice	Zinc, Iron	Bangladesh, India	Disease and pest resistance, cold and submergence tolerance	2012-13
Wheat	Zinc, Iron	India, Pakistan	Disease resistance, Lodging	2012-2013

[a] Approved for release by National Governments after intensive multi-location testing for agronomic and micronutrient performance.

HarvestPlus activities are presented along a pathway of impact and are classified in three phases of discovery, development, and dissemination (see Figure 2). Research developments at any one stage may necessitate revisiting previous stages to refine and ensure high quality of biofortified products.

Discovery and development research include standardizing analytical methodologies, protocols, and proof of concept research in relation to crop improvement, testing, and nutritional efficacy. Dissemination activities are highly dependent on the success of the discovery and development phase, as well as establishing partnerships between HarvestPlus and country agencies, which will lead the delivery of biofortified seeds to farmers and introducing biofortified crops to consumers.

Figure 2. HarvestPlus Pathway to Impact



Stage 1: Identifying Target Populations and Staple food Consumption Profiles: Overlap of cropping patterns, consumption trends, and incidence of micronutrient malnutrition determine target populations. This in turn determines the selection and geographic targeting of focus crops.

For each of the seven staple food crops listed in Table 1, the following activities have been undertaken:

- Identified countries with high per capita consumption of the food staple (based on FAO databases);
- Established estimates of the prevalence of iron, zinc, and vitamin A deficiencies for these countries;
- Gathered information on existing and planned expansion and effectiveness of alternative micronutrient interventions in these countries;
- Information mentioned above were used to identify countries with highest potential impact;
- Preliminary evaluation of the feasibility of developing and delivering biofortified crops in the high-potential-impact countries.

This included an assessment of:

1. The scientific capability and institutional strength of the national agricultural research and extension programs (NARES);
2. Present levels of adoption of improved, modern varieties by poor farmers, the level of development of seed distribution systems, and the feasibility of realizing significant adoption of high-yielding/high-profit biofortified varieties combined with superior agronomic characteristics of newly-introduced varieties;
3. Political stability and strength of supporting government and non-government enabling institutions.

Formal *ex ante* impact and benefit-cost analyses were conducted to help refine the targeting exercise.[3]

Stage 2: Setting Nutrient Target Levels: Nutritionists work with breeders to establish nutritional breeding targets based on the food intake of target populations, nutrient losses during storage and processing, bioavailability of nutrients related to the presence or absence of complementary compounds.

One of the first questions asked by breeders and nutritionists in the development of the HarvestPlus program strategy was "By how much do we need to increase the micronutrient content of our crops to improve the micronutrient status of their consumers?" As a food-based strategy, the additional micronutrient intake resulting from biofortification would ideally be enough to fill the gap between current intakes and the amount that would result in the majority of the population having intakes above the theoretical mean dietary requirement level (the estimated average requirement, or EAR) for the respective micronutrient. Universal food fortification programs recommend this approach in their design (WHO, 2006). However, quantitative information on micronutrient intakes for most potential target populations does not exist, or exists in very limited form. There are also differences in staple food processing, storage, and cooking practices and inclusion of other foods that can result in large differences in the micronutrient content and bioavailability in the staple food across different populations.

HarvestPlus set preliminary "minimum" target levels for micronutrient content using gross assumptions about staple food intake (grams/day); bioavailability (percent nutrient absorbed) or, in the case of vitamin A, the retinol equivalency of pro-vitamin A carotenoids; losses of the target nutrient with milling, processing, storage, and cooking; and the proportion of the daily nutrient requirement that should be achieved from the additional amount of micronutrient in the staple food. Table 2 presents examples of the types of data used to estimate target levels for micronutrient contents of biofortified crops. As information of this type becomes available for specific populations, target levels for micronutrient contents in different staple food crops can be refined and adjusted. If preliminary target levels are determined inadequate, for a specific population, the breeding process will continue until breeders reach or surpass the necessary micronutrient content.

As with universal fortification of staple foods, biofortification will lead to some degree of increased micronutrient intakes among individuals in all life stages. A possible exception is exclusive breastfed children, but even in this case, increased intakes of pro-vitamin A by lactating women may result in increased content in the breast milk and hence transfer to the breastfed infant. Young children and women of reproductive age typically suffer the greatest consequences of micronutrient deficiencies because of their increased requirements for growth and for pregnancy and lactation, and hence they may be considered the primary targets for this strategy. Biofortification as the sole micronutrient strategy may not be sufficient to cover the deficit in micronutrient intakes by very young children (e.g., under 2 years of age), who have particularly high micronutrient needs and relatively low staple food intakes. Therefore, HarvestPlus estimated appropriate target levels for the micronutrient contents of biofortified foods taking into account the potential impact in children approximately 4 to 6 years of age and in non-pregnant, non-lactating, pre-menopausal women (Table 2). It is estimated that with the lower staple food intakes by younger children (i.e., 1 to 3 years of age) who may still be breastfed, the same target levels for biofortified foods may still cover approximately one-quarter to one-third of their micronutrient requirements. The potential biological impact of a lower increment in micronutrient impact would need to be determined.

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[3] This involved developing a methodology for undertaking *ex ante* benefit-cost analysis. This is described in detail in Stein et al 2005. The following publications are based on this methodology: Stein et al, 2007, Stein et al 2008, Qaim, et al, 2007, J.V. Meenakshi et al, 2007.

Researchers will compile empirical data on staple food intakes for different age groups in a variety of populations in order to refine these estimates. How these levels of increased micronutrient intake translate into changes in nutrition and health status remains to be determined. As breeding for biofortification progresses, the achievable micronutrient content may exceed the current minimum target level, and thus make a greater contribution to the micronutrient needs among those groups with elevated requirements.

Table 2. Information and assumptions used to set target levels for micronutrient content of biofortified staple food crops.

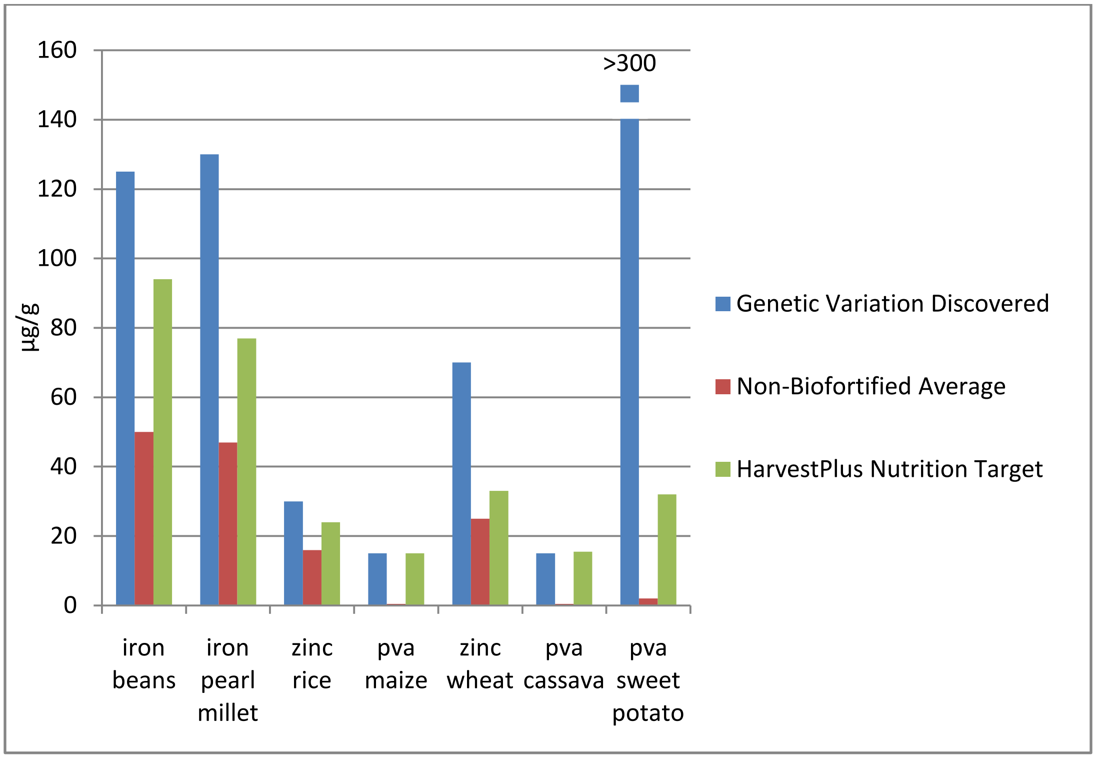
	Iron		Zinc		Pro-vitamin A	
Age/physiological status group	Non-pregnant non-lactating women	Children 4-6 yr of age	Non-pregnant, non-lactating women	Children 4-6 yr of age	Non-pregnant non-lactating women	Children 4-6 yr of age
% of daily micronutrient requirement to achieve[a]	~30%		~40%		~50%	
Estimated average requirement (μg/day)[a]	1,460	500	1,860	830	500	275
	Maize (whole) or Wheat (whole)					
Intake (g/day)[b]	400	200	400	200	400	200
Micronutrient retention after processing	90%		90%		50%	
Bioavailability[c]	5%		25%		12:1	
Baseline micronutrient content (μg/g)	30		25		0 - 0.5	
Additional content required (μg/g)	+ 22		+ 8		+ 15	
Total final content (μg/g)	52		33		15.5	
	Rice (polished) or Cassava (fresh weight)					
Intake (g/day)[b]	400	200	400	200	400	200
Micronutrient retention after processing	90%		90%		50%	
Bioavailability[c]	10%		25%		12:1	
Baseline micronutrient content (μg/g)	2 (rice) 4 (cassava)		16 (rice) 4 (cassava)		0 - 0.5 (cassava)	
Additional content required (μg/g)	11		8		15	
Total final content (μg/g)	13 (rice) 15 (cassava)		24 (rice) 12 (cassava)		15.5	
	Pearl Millet (whole)					
Intake (g/day)[b]	300	150	300	150	300	150
Micronutrient retention after processing	90%		90%		50%	
Bioavailability[c]	5%		25%		12:1	
Baseline micronutrient content (μg/g)	47		47		0	
Additional content required (μg/g)	30		11		20	
Total final content (μg/g)	77		58		20	
	Beans					
Intake (g/day)[b]	200	100	200	100	200	100
Micronutrient retention after processing	85%		90%		50%	
Bioavailability[c]	5%		25%		12:1	
Baseline micronutrient content (μg/g)	50		32		0	
Additional content required (μg/g)	44		17		30	
Total final content (μg/g)	94		49		30	

Stage 3: Screening and Applied Biotechnology: The global germplasm banks of the CGIAR institutes and the germplasm banks held in trust by national partners provide a reservoir of staple crops germplasm for screening by HarvestPlus. Genetic transformation provides an alternative strategy to incorporate specific genes that express nutritional density.

The first step in conventional breeding is to determine whether sufficient genetic variation exists to breed for a particular trait of interest —in the specific case of HarvestPlus whether breeding parents can be found with target levels (or higher) of iron, zinc, and pro-vitamin A. Researchers have analyzed approximately 300,000 samples for trace minerals or for pro-vitamin A carotenoids during screening.

The second step is to determine from the screening results whether, as shown in the chart below (Figure 3), sufficient genetic variation exists to breed for (1) high-zinc rice and wheat, (2) high-iron beans and pearl millet, and (3) high-pro-vitamin A cassava, maize, and sweetpotato.

Figure 3. Micronutrient content of staple crops, across varieties from screening activities.



Stage 4: Crop Improvement: Crop improvement along with nutritional bioavailability and efficacy (stage 7 below) make up the two largest stages of all research activities. Crop improvement includes all breeding activities falling within a product concept that produces varieties containing those traits that (in target populations, in target areas) improve nutrient content while giving high agronomic performance, and preferred consumer quality.

Biofortification Crop Improvement is divided into three phases:

- Early Stage Product Development and Parent Building (phase 1)
- Intermediate Product Development (phase 2)
- Final Product Development (phase 3)

Phase 1 and Phase 2 just above are undertaken at CGIAR Centers. Final product development (phase 3) for a particular growing “mega-environment” may take place at the CGIAR center or at the NARES. Once promising high-yielding, high-nutrient lines emerge from final product development, these promising lines are then tested by the NARES in multi-location trials throughout the target country (sometimes referred to as “genotype by environment, or G x E, testing”).

A subset of these promising lines that do well on average across these several in-country sites are then submitted formally to Varietal Release Committees for testing for official release. VRCs perform independent, multi-location trials before officially approving varieties for release. During these multi-location trials, in anticipation of a favorable decision by the VRCs and to save time, often NARES begin to multiply seed prior to "pre-launch".

This entire process may take up to 6-8 years to complete. Table 3 characterizes current progress throughout the five-phase breeding pipeline for seven crops. Progress is measured by the nutrient levels expressed as a percentage of the absolute target levels given in breeding lines in a specific phase of development. High-yielding, high-nutrient biofortified varieties ***currently*** emerging from the end of this breeding "pipeline" may have lower levels of nutrients and/or lower yields than prototype lines ***currently*** entering the front of the pipeline -- owing to new discoveries made, such as higher nutrient germplasm (breeding parents), discovered while lines about to be released have been making their way through the breeding pipeline.

For example, all orange sweetpotato varieties — whether currently being tested for official release (stage 5) or currently just under initial development (phase 1) — all have at least 100% of the target levels of 30 ug/g pro-vitamin A carotenoids.

By contrast, for cassava, much recent and rapid progress has been made in developing lines which meet the target of 15 ug/g pro-vitamin A carotenoids. Thus, only varieties currently in phase 1 of the breeding process have 100% of the target level for pro-vitamin A carotenoids. High-yielding, yellow cassava varieties with 30-50% of the target level are currently being tested for release (stage 5), reflecting a relative lack of progress in attaining high pro-vitamin A levels just 2-3 years ago. Even though varieties currently in stage 5 have pro-vitamin A levels well below target levels, they are high-yielding and thus can be approved for release.

Finally, "fast-track" options in Table 3 refer to lines, which due to their high yields and favorable agronomic characteristics are in the regular breeding program, but have been discovered (serendipitously during germplasm screening) to be relatively high in iron, zinc, or pro-vitamin A. Breeding for high nutrient levels is not necessary. They are coursed directly through multi-location testing, then (assuming favorable results) to the varietal release committees.

Stage 5: Genotype by Environment (GxE) Interactions on Nutrient Density: Germplasm is tested in target countries for their suitability for release. Genotype x environment interaction can greatly influence genotypic performance across different crop growing scenarios. HarvestPlus researchers are looking for high and stable expression of high micronutrient content across environments as well as alternative farming practices that enhance the uptake of nutrients in the edible portion of the crop.

By 2008, all crops have, or are about to enter GxE trials. Table 3 presents progress of the first and successive generations of biofortified crops. GxE analysis of sweet potato, for example, shows that varieties are demonstrating 100% of the target. GxE analysis of cassava currently finishing all stages of breeding have accomplished 50% of the target, but those improved varieties that are just entering the cassava breeding process, and have built on the successes of previous generations, are demonstrating their ability to reach 100% target.

Stage 6: Nutrient Retention and Bioavailability: HarvestPlus nutrition teams are measuring the effects of usual processing, storage and cooking methods on micronutrient retention for biofortified crops and evaluating practices that could be used by target populations to improve retention.

Recent research results are suggestive that retention of micronutrients may also be genetically determined which then adds retention heritability to the plant breeding portfolio. Nutritionists use various methods to study the degree to which the nutrients bred into crops are absorbed by using in vitro and animal models, and with the most promising varieties, by direct study in humans in controlled experiments. These studies guide plant breeders in refining their breeding objectives

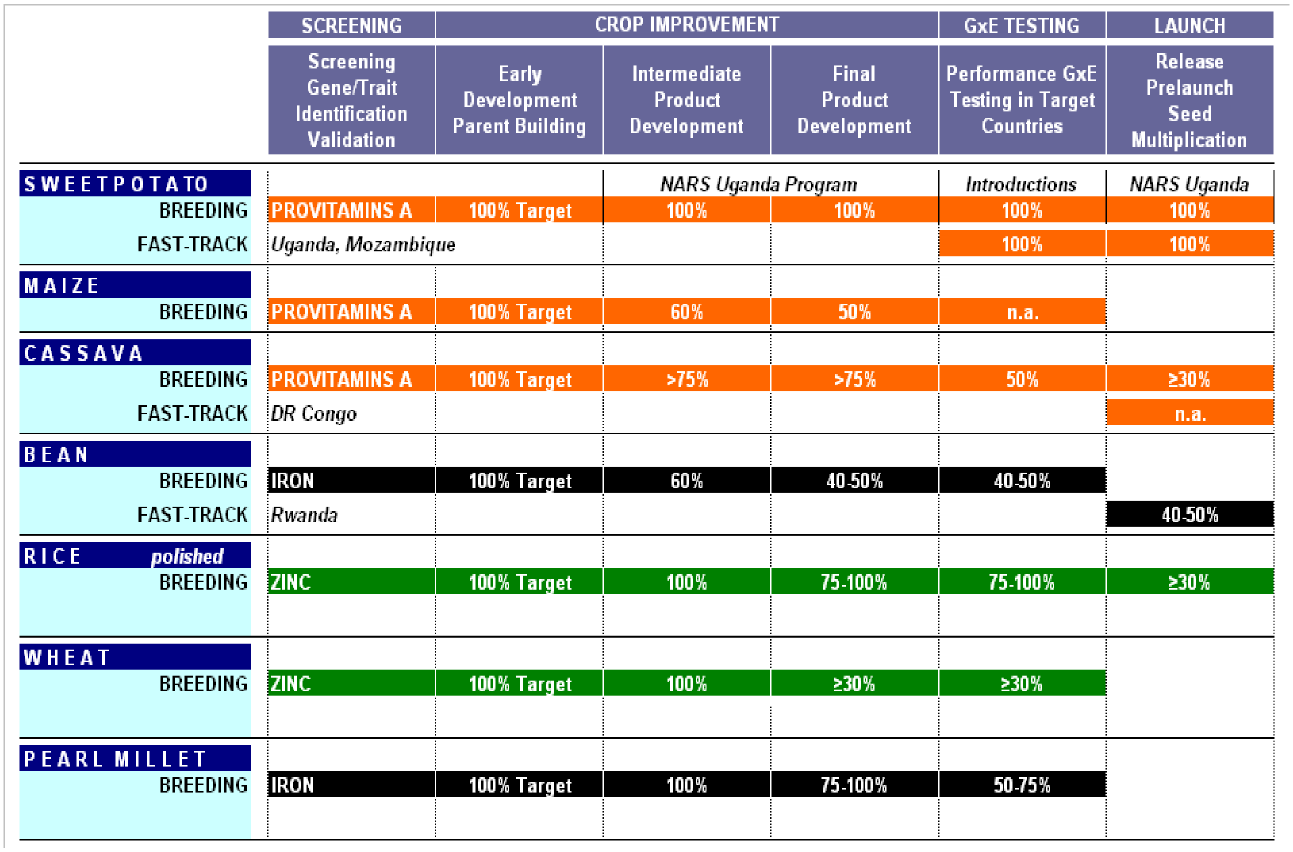
Stage 7: Nutritional Efficacy Studies on Human Subjects: Although nutrient absorption by the body is a prerequisite to preventing micronutrient deficiencies, ultimately the change in prevalence of micronutrient deficiencies with long term intake of biofortified staple foods needs to be measured directly. Thus, randomized controlled efficacy trials demonstrating the impact of biofortified crops on micronutrient status will be required to provide evidence to support the release of biofortified crops at the level of nutrient density thus far achieved (i.e., the minimum target level).

As outlined under Stage 2 above, nutrient targets have been set for breeders based on assumptions about retention of micronutrients in the staple food crop food following usual processing and cooking methods and how bioavailable these nutrients will be when consumed by micronutrient deficient populations. These assumptions need to be studied and tested empirically. Eventually, efficacy needs to be evaluated as well.

The results of research on retention, bioavailability, and efficacy are summarized in Tables 4, 5, and 6 below. In general, findings show that retention and bioavailability are higher than assumed. If these promising results are validated with further research to be undertaken under HarvestPlus II, this could eventually allow for a lowering of minimum target levels for breeders. If, however, breeders can attain the targets already set, the thus-far-promising results suggest that impacts could be higher than expected.

Stage 8: Release Biofortified Crops: Varietal release regulations differ by country and often by states within countries. Proof that the variety is new, distinguishable, and adds value must be established in order to register new varieties of crops. HarvestPlus works with NARES to gather the relevant information for registration and formal release of biofortified crops in target regions.

Table 3. Breeding progress as of 2007-08 (iron, zinc, pro-vitamin A expressed as percent of breeding target in lines at indicated stage of breeding).



	SCREENING	CROP IMPROVEMENT			GxE TESTING	LAUNCH
	Screening Gene/Trait Identification Validation	Early Development Parent Building	Intermediate Product Development	Final Product Development	Performance GxE Testing in Target Countries	Release Prelaunch Seed Multiplication
SWEETPOTATO			*NARS Uganda Program*		*Introductions*	*NARS Uganda*
BREEDING	PROVITAMINS A	100% Target	100%	100%	100%	100%
FAST-TRACK	*Uganda, Mozambique*				100%	100%
MAIZE						
BREEDING	PROVITAMINS A	100% Target	60%	50%	n.a.	
CASSAVA						
BREEDING	PROVITAMINS A	100% Target	>75%	>75%	50%	≥30%
FAST-TRACK	*DR Congo*					n.a.
BEAN						
BREEDING	IRON	100% Target	60%	40-50%	40-50%	
FAST-TRACK	*Rwanda*					40-50%
RICE *polished*						
BREEDING	ZINC	100% Target	100%	75-100%	75-100%	≥30%
WHEAT						
BREEDING	ZINC	100% Target	100%	≥30%	≥30%	
PEARL MILLET						
BREEDING	IRON	100% Target	100%	75-100%	50-75%	

Table 7 provides information related to projected years for GxE and varietal release activities for biofortified varieties currently in target country multi-location testing (phase 4). For example, ZARI is expected to submit high-pro-vitamin A open pollinated varieties OPVs and hybrids to the Zambian VRC sometime in

2009, and approval for their release is expected in 2011/12. ZARI completed its own multi-location testing of the first wave of products in 2009 (phase 4).

Stage 9: Facilitate Dissemination, Marketing and Consumer Acceptance: Market chain analysis, seed development and production capacity, consumer acceptance studies, and the cultivation of an enabling policy environment for the uptake and production of biofortified crops in country are essential corner stones for the development of a sustainable, independent, demand-driven, national biofortification research and implementation program.

The dissemination strategy for nutrients that are invisible (iron and zinc) is to "piggy-back" on superior agronomic characteristics that will drive adoption of the newly-introduced varieties and capture a large share of total supply and so consumption in a given country. For example, high-iron beans that are drought and heat tolerant are undergoing national release trials in Africa. High-zinc wheat varieties to be released in India and Pakistan will be resistant to newly-evolved yellow rust viruses, to which current popular varieties are not resistant.

For nutrients which are visible – high provitamin A sweetpotato, maize, and cassava are orange or yellow -- nutritional messages must be delivered simultaneously with release of high yielding, high profit biofortified varieties to effect a switch from production/consumption of white varieties (which is currently the norm) to production/consumption of orange or yellow varieties.

HarvestPlus' experience in the dissemination of biofortified crops is limited to orange sweetpotato, which is very high in pro-vitamin A. A published pilot study in Mozambique showed that (1) behavior can be changed among farmers by switching from production of white to orange varieties, and change in consumption to orange varieties by their families, and that (2) vitamin A deficiency can be improved. As a result, vitamin A deficiency among pre-school children in treatment villages declined from 60% to 38%, while vitamin A deficiency remained constant in control villages. HarvestPlus is now concentrating on identifying activities and messages that will effect this same behavior change at the lowest cost possible.

In 2006, HarvestPlus embarked upon its first dissemination activity of high-pro-vitamin A carotenoid (pVAC) sweetpotato in Uganda and Mozambique. Researchers and implementation specialists are gathering lessons learned in strengthening seed systems, developing markets, and generating consumer demand through behavior change for this nutrient-dense orange variety of sweetpotato. Best practices will be applied to, 1) expansion of sweetpotato to other regions of the world and, 2) to instruct dissemination strategies of other pVAC-dense (orange) biofortified staple crops.

Stage 10: Improved Nutritional Status of Target Population: Ultimately, biofortified crops are expected to improve the nutritional status of populations. Baselines and post-dissemination impact and effectiveness surveys are conducted in target regions with and without the intervention to determine whether biofortified crops can improve human health outside experimental conditions.

Some work is ongoing related to the dissemination of orange sweetpotato in Uganda and Mozambique (see above). Final results will be reported in mid-2010.

Table 4. Summary of progress on retention, bioavailability, and efficacy for high pro-vitamin A crops.

	Sweetpotato	Maize	Cassava
Assumed Retention in Calculating Breeding Target	50%	50%	50%
Assumed Conversion From pVAC to Retinol Equivalents	12:1	12:1	12:1
Summary of Research Findings and Research Plans			
Retention	Boiled: 50-130% Steamed: 69-84% Roasted: 40-110% Dried chips: 59-91% Dried flour: 62% Storage: 30-40% Storage of dried products: Study in progress	Wet milling, fermented and non-fermented porridges: 71-75%; Storage: Study in progress	Boiled: 66-100% Dried Flour: 27-66% Fermented: 92% Gari: 10-66% Fufu: 90% Drying: 40-60% High genotypic variation in retention. Range in advanced germplasm will be confirmed and causes to be determined*.
In Vitro and Animal Model – Gerbil	REq of beta-carotene being compared for sweetpotato with and without added vegetable oil.	3:1 REq for beta-carotene and beta-cryptoxanthin.	4:1 REq in Mongolian gerbil model. To determine REq of cis- and trans-beta-carotene isomers
Bioavailability (Retinol Equivalency = REq)	13:1 REq in previous study of OFSP (with oil). Currently determining REq without cooking oil, and effects of beta-carotene on zinc and iron absorption.	7:1 REq found in human model (plasma appearance method)	Study of REq underway in a human model.*
Background Nutrition Studies	Data on intakes of sweetpotato and total dietary vitamin A collected in Uganda and Mozambique; results are pending*.	Study of vitamin A status and intakes in pre-schoolers to commence in Zambia in 2008-2009*.	Vitamin A intakes and status in pre-schoolers to be determined in DR Congo. Existing data will be reanalyzed for Nigeria*.
Efficacy	Impact on vitamin A status established among Indonesian children, South African children, and Bangladeshi men (pre-HarvestPlus).	Study of efficacy among Zambian children planned for 2009-2010*.	Efficacy among pre-school children planned for Nigeria (2010 - 2011). An EU funded study planned for Kenyan school children in ~2011*.
Effectiveness	Small scale study reduced rate of vitamin A deficiency from 60% to 38% in Mozambiquan children. Assessing nutritional impact of large scale roll-out in Mozambique and Uganda 2008-2009*.	Pending	Pending

* Studies are currently being implemented.

Table 5. Summary of progress on retention, bioavailability, and efficacy for high zinc crops.

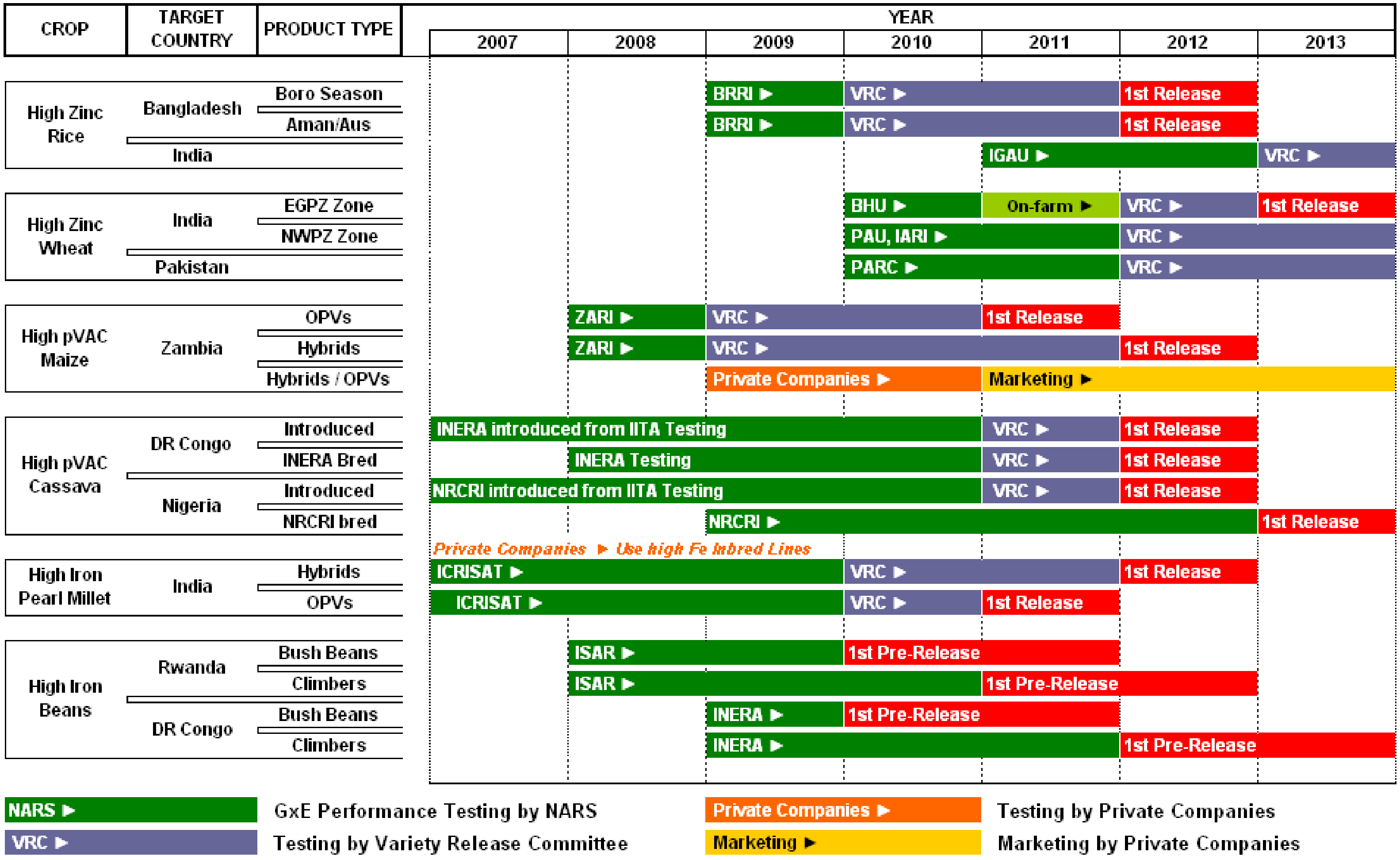
	Rice	Wheat
Assumed Retention in Calculating Breeding Target	90%	90%
Assumed Bioavailability in Calculating Breeding Target	25%	25%
Retention	Milled: 44-65% Effect of parboiling and discarding excess cooking water underway.	Chapati: 60-97% Parantha: 67-69% Zinc loss by degree of milling to be determined in biofortified wheat.
In Vitro and Animal Model (rat pup)	Caco-2 cell model and rat pup model being developed using Bangladeshi rice.	Assess usefulness of models with rice; if positive then use for zinc wheat from Pakistan/India
Background Nutrition Studies	Rice and zinc intakes & zinc status being determined in 2-4 yr old children and women 2007-2008	Measure wheat and zinc intakes and zinc status in pre-schoolers in Pakistan and India 2009-2010
Bioavailability – Stable Isotope	Zinc absorption being measured in 4 year old children 2008-2009. To repeat in India 2010	30% and 15% absorption in Mexican women for 80% and 95% extraction biofortified wheat. **To** determine effect of zinc & phytate in 1-2 yr old Pakistani children 2009-2010
Efficacy	Planned for Bangladeshi 2-4 yr old children in 2010-2011; to repeat in India 2011-2012	Planned for Punjabi (Pakistani or Indian) pre-school children 2012-2013
Effectiveness	Pending	Pending

Table 6. Summary of progress on retention, bioavailability, & efficacy for high iron crops.

	Beans	Pearl Millet	Rice
Assumed Retention in Calculating Breeding Target	90%	90%	90%
Assumed Bioavailability in Calculating Breeding Target	5%	5%	10%
Summary of Research Findings and Research Plans			
Retention	Retention with Rwandan cooking practices to be determined (2009)	Retention with decortication and cooking methods in India (2009-2010)	Milled: 20-50% retained; retention with under-milling to be determined (2008-2009)
In Vitro	Polyphenol content has strong inhibitory effect on iron bioavailability	Caco-2 cell to determine genotypic variation in bioavailability (2009)	
Animal Model – Pig	High iron black bean (105 ppm) increased hemoglobin after 5 weeks compared with lower iron black bean (70 ppm)	No plans for animal model	
Background Nutrition Studies	Bean and iron intakes and iron status determined in Rwandan women and pre-school children (2009)	Pearl millet intakes, iron, intakes, and iron status in Indian women and pre-school children (2008-2009)	
Bioavailability – Stable Isotope	Iron absorption from beans with different polyphenol content in Rwandan women (2008)	Iron and zinc absorption in pre-school Indian children (2010-2011)	
Efficacy	Planned for Mexican school children (2008-2009) and adult Rwandan women (2009-2010)	Indian pre-school children (2012-2013)	Iron stores improved after nine months, +1.5 mg Fe/day; overall bioavailability 17%**
Effectiveness	Pending	Pending	

** Original study funded under pre-cursor CGIAR Micronutrients Project; additional analysis of data funded under HarvestPlus

Table 7. Schedule of first releases of biofortified crops by country by type of technology and by season.



CROP	TARGET COUNTRY	PRODUCT TYPE	2007	2008	2009	2010	2011	2012	2013
High Zinc Rice	Bangladesh	Boro Season			BRRI ►	VRC ►		1st Release	
		Aman/Aus			BRRI ►	VRC ►		1st Release	
	India						IGAU ►		VRC ►
High Zinc Wheat	India	EGPZ Zone				BHU ►	On-farm ►	VRC ►	1st Release
		NWPZ Zone				PAU, IARI ►		VRC ►	
	Pakistan					PARC ►		VRC ►	
High pVAC Maize	Zambia	OPVs		ZARI ►	VRC ►		1st Release		
		Hybrids		ZARI ►	VRC ►			1st Release	
		Hybrids / OPVs			Private Companies ►		Marketing ►		
High pVAC Cassava	DR Congo	Introduced	INERA introduced from IITA Testing				VRC ►	1st Release	
		INERA Bred		INERA Testing			VRC ►	1st Release	
	Nigeria	Introduced	NRCRI introduced from IITA Testing				VRC ►	1st Release	
		NRCRI bred			NRCRI ►				1st Release
			Private Companies ► Use high Fe Inbred Lines						
High Iron Pearl Millet	India	Hybrids	ICRISAT ►			VRC ►		1st Release	
		OPVs	ICRISAT ►			VRC ►	1st Release		
High Iron Beans	Rwanda	Bush Beans		ISAR ►		1st Pre-Release			
		Climbers		ISAR ►			1st Pre-Release		
	DR Congo	Bush Beans			INERA ►	1st Pre-Release			
		Climbers			INERA ►			1st Pre-Release	

NARS ►	GxE Performance Testing by NARS	Private Companies ►	Testing by Private Companies
VRC ►	Testing by Variety Release Committee	Marketing ►	Marketing by Private Companies

3. Conclusions

The biofortification strategy seeks to take advantage of the consistent daily consumption of large amounts of food staples by all family members, including women and children who are most at risk for micronutrient malnutrition. As a consequence of the predominance of food staples in the diets of the poor, this strategy implicitly targets low-income households.

After a one-time investment in developing seeds that fortify themselves, recurrent costs are low and germplasm may be shared internationally. It is this multiplier aspect of plant breeding across time and distance that makes it so cost-effective.

Once in place, production and consumption of nutritionally improved varieties is highly sustainable, even if government attention and international funding for micronutrient issues fade.

Biofortification provides a feasible means of reaching malnourished populations in relatively remote rural areas, delivering naturally fortified foods to people with limited access to commercially-marketed fortified foods, which are more readily available in urban areas. Biofortification and commercial fortification, therefore, are highly complementary.

Ultimately, good nutrition depends on adequate intakes of a range of nutrients and other compounds, in combinations and levels that are not yet completely understood. Thus, the best and final solution to eliminating undernutrition as a public health problem in developing countries is to provide increased consumption of a range of non-staple foods. However, this will require several decades to be realised, informed government policies, and a relatively large investment in agricultural research and other public and on-farm infrastructure.

In conceptualising solutions for a range of nutritional deficiencies, interdisciplinary communication between plant scientists and human nutrition scientists holds great potential. Human nutritionists need to be informed, for example, about the extent to which the vitamin and mineral density of specific foods, as well as compounds that promote and inhibit their bioavailability, can be modified through plant breeding. Plant breeders need to be aware of both the major influence that agricultural research may have had on nutrient utilisation in the past (e.g. the bioavailability of trace minerals in modern varieties versus bioavailability in traditional varieties), and the potential of plant breeding for future improvements in nutrition and health.

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