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Participatory crop improvement and formal release of *Jethobudho* rice landrace in Nepal

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Abstract *Jethobudho* is an aromatic rice landrace of the Pokhara valley in middle hills of Nepal. Although local consumers are willing to pay a high price for its purchase, the landrace has a problem with quality variation. Decentralized participatory population improvement for specific market-identified traits was conducted on "*Jethobudho*" populations collected from farmers' fields in seven geographic regions of the valley in Nepal. The preferred post harvest quality traits, field tolerance to blast and lodging, and superior post harvest quality traits of *Jethobudho* were established by a consumer market survey. These traits were used for screening the materials. 338 sub-populations of *Jethobudho* were evaluated for yield, disease, lodging resistance, and post harvest quality traits. Significant variation was found for culm strength, neck blast tolerance, awn characteristics, panicle length, number of grains per panicle, test grain weight and post harvest quality traits, whereas no significant variation was found in grain yield, plant height, tiller number, maturity period and leaf blast. Based on these identified traits and micro-milling evaluations, 183 populations were screened in on-farm and on-station nurseries, and in succeeding years populations were further screened by plant breeders and expert farmers in research trials, resulting in the selection of 46 populations for post harvest quality traits. Six accessions with similar agronomic traits, field tolerance to blast and lodging, and superior post harvest quality traits, were bulked and evaluated on-farm using participatory variety selection (PVS). The enhanced *Jethobudho* accessions were also evaluated for aroma using simple sequence repeat (SSR) and found to have unique aromatic genetic constitution. Community based seed production groups were formed, linked to the Nepal District Self Seed Sufficiency Programme (DISSPRO), and trained to produce basic seeds (truthfully labeled) of *Jethobudho*. The National Seed Board of Nepal released the enhanced landrace in the name of "*Pokhareli Jethobudho*" in 2006, as the first bulk variety of traditional high quality aromatic rice improved through participatory plant breeding to be formally released in Nepal for general cultivation under the national seed certification scheme. Landrace improvement is shown as an important option for supporting programmes for in situ conservation of landraces on-farm.

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Introduction

Traditional crop varieties or landraces are an important element of crop genetic resources and are valued by plant breeders and farmers because of diversity (a heterogeneous population), rarity (embodying unique traits) and adaptability (exhibiting wide ecological and socio-cultural adaptation) (Brush and Meng 1998; FAO 1998; Smale 2006; Gauchan et al. 2006). Farmers throughout the world continue to maintain and manage these traditional varieties within their production systems (Frankel and Eennett 1970; Hawkes 1971; Duvick 1984; Brush et al. 1995; Brush 2004; Jarvis et al. 2008; FAO 2010). Yet the value they contain for the farming communities that maintain them has not been fully capitalised on.

Not all landraces are equally valued by farmers. Some landraces are adapted to marginal ecosystems (Vandermeer 1995, Bezancon et al. 2009; Barry et al. 2007; Rana et al. 2008). Others have cultural, religious, or nutritional value (Rana et al. 2007b; Sthapit et al. 2008; Johns and Sthapit 2004). Some landraces maybe highly valued but their use is constrained by poor access to quality and quantity of seeds or planting materials (Tripp 2001; Almekinders et al. 2007; Sperling et al. 2008; Hodgkin et al. 2007). Landrace populations may, themselves, not be uniform in their adaptive or quality traits, having significant variation both within and among populations (Teshome et al. 2001; Harlan 1975; Mariac et al. 2006; Barry et al. 2007). One way of distinguishing those varieties that provide high public value, i.e., varieties that are high in diversity of crop genetic resources from which people residing elsewhere and in the future may benefit (Smale 2006), is to classify varieties in terms of their immediate and future plant breeding benefits (Smale et al. 2004). This required consultation with farmers and breeders but also other concerned actors including consumers, millers and retailers (Sthapit et al. 2001; Sperling et al. 2001; Bellon et al. 2003; Witcombe et al. 2006a, b).

The recent movement in participatory and decentralized plant breeding over the last decade has shown that improving varietal performance in low input systems can help improve local livelihoods (Ceccarelli et al. 2000; Smith et al. 2001; Almekinders and Elings 2001; Zeven 2000; Dawson et al. 2008). Sthapit (1992), Sthapit et al. (1996) and Witcombe et al. (1996) have demonstrated that the value of local cold tolerant rice varieties can be improved by selection of preferred traits from the heterogeneous populations, collected locally before any crop improvement programme is initiated. Genebank curators and plant breeders continue to collect traditional varieties for future use in plant breeding, and a significant amount of this conserved material is used in academic research (Dudnik et al. 2001). However, insufficient attention has been given to the potential use of the existing landrace variability in production systems to provide direct benefits to local communities (Sthapit and Rao 2009).

Jethobudho is an aromatic rice landrace of the Pokhara valley in middle hills of Nepal. *Jethobudho* is valued for its superior cooking qualities such as softness, taste, aroma and volume expansion ability and for its superior milling recovery (Rijal et al. 1998; Sthapit et al. 2001). Variability in quality is the main concern of consumers and market entrepreneurs in marketing of this popular variety. Consumers are willing to pay a high price (Poudel and Johnsen 2009) for its purchase, but the landrace has a problem with quality variation. This significant quality variation, coupled with poor access to quality seed and inadequate policy support, including variety release policies and quality seed production for landraces, has limited the ability of *Jethobudho* rice to be competitive in relation to other high quality rice varieties.

This study documents an eight year process of decentralized participatory population improvement for specific market identified traits of the *Jethobudho* rice landrace from the initial setting of breeding goals to the registration and release by the National Seed Board of Nepal for general cultivation under the national seed certification scheme. The enhanced landrace in "*Pokhareli Jethobudho*" is the first Nepalese nationally released bulk variety of traditional high quality aromatic rice improved through participatory plant breeding.

Materials and methods

Site description

The study sites are located in the Pokhara valley, central Nepal (28°11′35.10″ N and 83°58′6.62″ E). The valley was formed from an ancient river terrace composed of calcareous, gravely and fluvial deposits and occupies an area of 625 km^2, with an annual rainfall of 3500–4500 mm. Mean air temperature from April to October is above 20°C. Summers are warm with ample rainfall and winters are drier and colder. Rice is grown in the flat valley plains from <600 m asl to terraced rice fields in the surrounding hills up to 2200 m asl. Diverse types of rice ecosystems are found, which include: upland (unbunded[1] and not irrigated), rainfed bunded, irrigated and marshy land types (Rana 2004; Rijal 2007). Earlier studies found that farming communities in this region maintain a substantial amount of diversity within and between rice landraces at the community level (Rijal et al. 1998). In Begnas, Kaski district, 73% of the rice farm area in the community is devoted to traditional varieties (landraces sensu Harlan 1975), made up of 51 traditional rice varieties (Rana et al. 2007a; Jarvis et al. 2008). Communities from Begnas Village Development Committee (VDC) identified *Jethodbudho* as most preferred landrace for germplasm enhancement using participatory four-cell analysis (Sthapit et al. 2001). *Jethobudho* falls into the category of traditional varieties that are commonly grown by many households on a larger area of their rice plots (Rana et al. 2007a). Habitats for plant selection (target production environment) were located through consultations with expert farmers, consumers and local agricultural development officers. Table 1 lists the locations and different water regimes where the plant material for this study was collected.

Materials

Samples were then collected at the time of maturity from 338 *Jethobudho* populations in seven different "*phants*" or productive flat agricultural areas in the Pokhara valley where rice is grown (Fig. 1; Table 1). Five panicles (neck blast free) were randomly selected from each of the 338 *Jethobudho* populations, and the seed bulked together and marked as an accessions. Each accession was divided into three packets and numbered. One packet of each of the bulked accession was used for blast screening and nursery observation in the Malepatan Agriculture Research Station (850 m asl). The remaining two parts were used to conduct on-farm diversity assessment trials in two major target environments: Malmul (600 m asl) and Fewa (900 m asl) Phants, which are traditional habitats of *Jethobudho*. Table 2 illustrates the detailed chronological steps of *Jethobudho* enhancement work carried out from 1998 to 2006.

Field screening

All 338 *Jethobudho* accessions from the seven geographic sites were grown on-station and on-farm conditions in 2000 in two locations of the Pokhara Valley: on-farm trials at Malmul, Begnas, and on-station at Malepatan. The experiment was conducted with a randomized complete block design (RCBD) with two replicates in each site. The plot size was 1.5 m × 1 m, with 20 × 10 cm spacing. Thirty plants from each accession (both on-farm and on-station) were measured for a set of qualitative (apiculus color, awn characteristics, culm strength, leaf and neck blast and lodging tolerance) and quantitative traits (plant height, tillers number per plant, panicle length, grain numbers per plant, test weight and grain yield per row). The mean, standard deviation, range and coefficient of variation were estimated for *Jethobudho* landrace accessions for plant height (≤150 cm), number of tillers $plant^{-1}$, panicle length, number of grains $panicle^{-1}$, test grain weight, grain yield, leaf and neck blast tolerance, culm strength, lodging tolerance, and awn characteristics (Gyawali et al. 2003).

In 2001, 183 *Jethobudho* accessions were screened for leaf and neck blast, lodging tolerance, and yield components. The accessions were exposed to the natural inoculums pressure of blast caused by *Magnaporthe grisea* (T. T. Hebert) M. E. Barr in the field. The blast infection was scored in 0–5 scale for leaf and neck blast using Standard Evaluation System for Rice (IRRI 1996). The accessions scoring more than two were rejected from further evaluation in each year.

[1] Rice in upland is dry seeded during pre-monsoon in Ghaiya khet whereas bunds (mud walls around terrace) are created to retain water for transplanting rainfed or irrigated rice (Rana et al. 2007a, b).

Table 1 Collecting sites of *Jethobudho* sub-populations in Pokhara valley in 1999

Phant (geographic area)	Altitude (m asl)	Water regime and source of water[a]	No of farmers' plots sampled for seed
Satmuhane and Malmul	600	Partially irrigated; Rupa lake water	50
Sisuwa and Maidi	675	Partially irrigated; Begnas lake water	50
Khaste	700	Irrigated; Khaste lake water	35
Arghun and Rithepani	700	Irrigated; snowmelt Seti river water	50
Kundhar Arba and Kamal Pokhari	775–800	Partially irrigated; natural spring	50
Biruwa	850	Irrigated; Fewa lake water	51
Fewa	900	Irrigated; cold Harpen river	52

[a] *Jethobudho* grown in organic fertilizer with cold water is perceived to have better aroma and quality

A lodging tolerance index was used to screen collected accessions similarly. The trials were conducted under farmers' management conditions and in lodging prone areas. Highly fertile rice fields with perennial source of water where farmers have experienced lodging of his/her source of *Jethobudho* every year were used to screen *Jethobudho* for lodging tolerance. The lodging tolerance was assessed in a scale of 0–5 (IRRI 1996). *Jethobudho* accessions scoring more than two were rated as lodging prone and rejected from future selection. Lodging tolerance was also verified by the culm strength measured qualitatively.

Laboratory screening

We developed a laboratory method of screening a large number of *Jethobudho* accessions using physical, micro-milling and organoleptic tests for post-harvest quality traits. We screened accessions based on the standard physio-chemical analysis (gel consistency, volume expansion, water absorption, and amylose contents).

Micro-milling (GRAINMAN micro-mill model no. 60-220-50-2AT) was then conducted on 143 accessions. The number was reduced to 143 accessions because 40 of the original 183 accessions had insufficient seeds to perform micro-milling. The 143 accessions were then further screened for blast, lodging, yield components using methods described above, and for organoleptic assessment (taste, aroma, softness, flakiness). To determine organoleptic traits screening, a consumer survey was conducted on 60 individuals made up of millers, hotel chefs, supermarket owners, and consumers (consumers consisted of predominantly female farmers) to determine important grain and cooking quality traits of *Jethobudho*. These traits were used for organoleptic traits screening by a panel of judges, representing hotels, millers and consumers. A total of 46 accessions were selected for further evaluation in 2002.

Field trials

In 2002, 46 accessions screened from the 143 accessions were grown in on-farm trials in Malmul and Pame as well as on on-station trials at the Agricultural Research Station (ARS), Malepatan, Pokhara. The plot size was of 2 m × 1 m arranged in randomized complete block design (RCBD) with three replicates in each location. Again plants were selected for shorter plant height, higher effective tillers plant^{-1}, longer panicle length, higher test weight and grain yield. The aroma and other grain characters were evaluated and best accessions were selected as farmers' preferred traits. At the later stage of the breeding program during 2004–2005 when there was sufficient seed, *Jethobudho* accessions were evaluated in 9 m^2 (3 m × 3 m) plots arranged in RCBD with three replicates. During the selection process in the field, farmers were invited to on-farm trials at the time of maturity to identify better accessions specially based on panicle types, neck blast, grain characters and general phenotypic acceptance. The selected best accessions were further evaluated for standard post harvest quality traits (IRRI 1996).

Consumer surveys

Consumer surveys consisted of two focus group discussions (FGD) with 8–12 farmers in each group

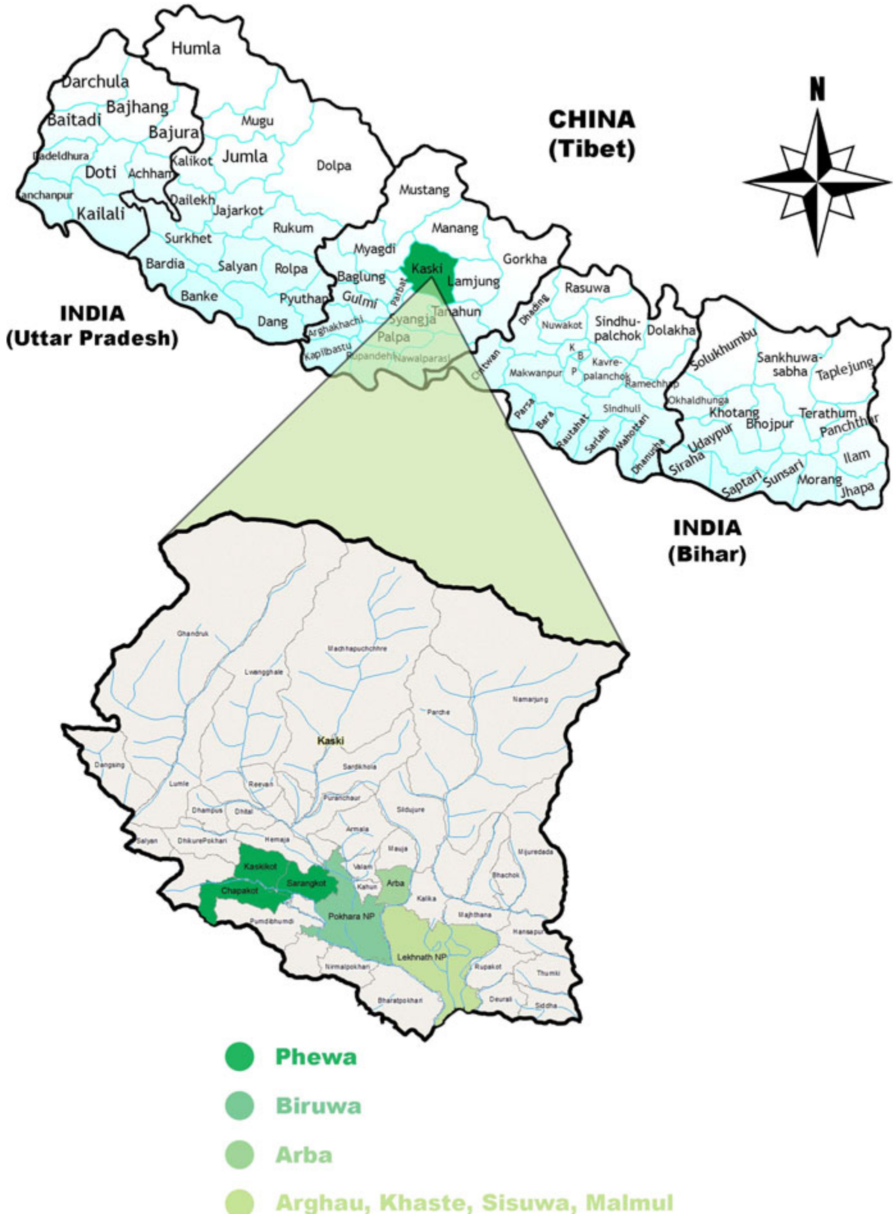

Fig. 1 Locations of 338 *Jethobudho* accession collection in Pokhara valley, Nepal

in 1999. In 2002, this was followed by 60 individual surveys that included housewives, consumers who consume *Jethobudho* year round, cooks from hotels within the valley, and farmers. Surveys contained questions on farmers' preferences for post-harvest quality traits, including physical appearances (grain type and grain color), milling traits (milling recovery, head rice recovery, broken rice) and organoleptic tests (aroma, cooking and eating qualities such as softness, taste and flakiness).

Participatory variety selection

Six agro-morphologically similar enhanced *Jethobudho* accessions were bulked and evaluated in 208 participatory variety selection (PVS) trials during 2003–2005 to compare overall performance of enhanced *Jethobudho* with farmers' checks (farmers own source of *Jethobudho*) and to multiply seed on-farm. Participatory variety selection (Joshi and Witcombe 1996) was employed in farmer's fields of

Table 2 Chronological steps of *Jethobudho* enhancement programme in Pokhara Valley, Nepal

Year	Activity and objectives	Location	Selected lines/cv	Selection criteria
1998	PRA, Baseline survey and FCA for understanding local crop diversity and setting breeding goal for landrace enhancement	Begnas ecosite	JB population	Landrace that has market incentive for selection
1999	Preferred trait analysis and sample collection at the harvesting time	Satmuhane and Malmul, Sisuwa and Maidi, Khaste, Arghun and Rithapan, Kundhar and Arba, Biruwa and Fewa	338	Visual selection in situ by researchers and farmers
2000	Diversity assessments for important traits: blast, lodging, height, seed characteristics, yield components	Malepatan, Pokhara (950 m) Malmul, Begnas (600 m) Pame, Phewaphant (900 m)	183	Awnless or with short awn; field tolerance to leaf and neck blast; strong culm; plants with 150 cm and productive plants
2001	(a) Market survey to determine quality traits of JB and screening for preferred traits (b) Evaluation of 183 JB lines under disease pressure	Pokhara valley market Malepatan Research Station (900 m)	143 46	Selection for post-harvest quality traits using micro-milling and organoleptic tests; 40 lines with insufficient seeds were rejected for further testing 143 JB accessions were further screened against leaf/neck blast, lodging, yield components and maturity
2002	Screening for post-harvest traits; PVS and SSR evaluation for aroma	Pokhara valley (900 m)	6	Milled rice type; rice colour, softness of cooked rice, flakiness, aroma, overall cooking quality and preference
2003	Agronomic and post harvest traits evaluation	A total of 7 sites in Malepatan, Pokhara; Malmulphant; Pame, Fewaphant	Bulk seed of six selected JB	Plant height; tillering ability, panicle length, grains per panicle, 1000 grain weight, grain yield, blast, lodging resistance and culm strength
2004	PVS; community based seed productions	Malmulphant, Khola-ko-Chew; Pame, Fewaphant	Bulk seed distributed to 12 farmers	Farmer's perception of target traits
2005	PVS; community based seed production; field visit by VRRC, National seed board	Begnas, Pame and Arghun	Enhanced JB	Performance evaluation by farmers and consumers
2006	Variety release; community based seed production	Pokhara valley	Pokhareli JB	Enhanced JB was released based upon its superior quality traits, milling recovery, blast and lodging tolerance

Pokhara Valley to compare the overall performance of enhanced *Jethobudho* with the farmer's own source of *Jethobudho* using the farmer's own evaluation criteria. Each farmer received about 2 kg of seed of the enhanced *Jethobudho* to compare with their own seed source of *Jethobudho*. In 2003, 15 farmers participated in the PVS trials because of limited availability of seed. Participating farmers were asked to keep the seed if other interested farmers were interested in enhanced *Jethobudho*. In 2004, 47 farmers participated in the on-farm PVS of enhanced materials. In 2005, 146 PVS trials were conducted in collaboration with the District Agriculture Development Office (DADO) and farmers' groups (Table 3). Evaluations conducted by researchers were validated by participatory variety selection technique as well. During 2003, 2004 and 2005, 90 farmers were surveyed with a formal household survey questionnaire 1 month after the harvest of the crop to obtain farmers' feedback on overall

Table 3 Distribution of participatory variety selection (PVS) trials in Pokhara valley, Nepal during 2003–2005

Year	Total amount of seed distributed (kg)	No of farmers who participated	No of farmers surveyed for HLQ	% of farmers who responded
2003	30	15	10	66.7
2004	94	47	33	70.2
2005	292	146	47	32.2

performance of enhanced *Jethobudho*. Selected accession for farmers' acceptance on post harvest quality traits in participatory variety selection (PVS) were analysed using household level questionnaires.

Molecular characterization of aroma in *Jethobudho*

Seven enhanced *Jethobudho* populations were evaluated for aroma along with three known aromatic varieties, viz. Azucena (a japonica cultivar from IRRI), Pusa Basmati-1 (popular Indian aromatic variety), local Rato Basmati (aromatic landrace from Nepal), and IR36 (one non-aromatic modern variety). The seven enhanced *Jethobudho* were selected on the basis of superiority in agronomic as well as post harvest quality traits. Seven SSR primers, distributed in the rice genome, having association with aroma in rice, were tested to understand this trait in enhanced *Jethobudho* populations using procedures developed by McCouch et al. (1997). Markers examined were RM223, RM42, RM1, RM241, RM 348, RM 202, and RM229 distributed in chromosomes 8, 1, 4, and 11 (Chen et al. 1997; Nagaraju et al. 2002). A pairwise similarity matrix was calculated using Jaccard's coefficient and employed to construct a dendogram to show the relationships among cultivars for aroma (data not shown) (Bajracharya et al. 2005). Genetic relationships among six enhanced *Jethobudho*, one bulk *Jethobudho* populations and four different aromatic and non-aromatic check varieties for aroma were detected by the allelic data of two SSR loci (RM42 and RM223) mapped in the location of RFLP probe RG1–RG28 linked to aroma in chromosome 8. In addition, five other markers (RM1, RM202, RM229, RM241, and RM348) were also used to understand the allelic behavior of enhanced *Jethobudho* populations and compared the alleles with that of aromatic and non-aromatic check varieties under study.

Institutionalization of community based seed production and marketing

Farmers growing *Jethobudho* were invited to participate in a series of village level meetings involving custodian farmers having knowledge of *Jethobudho* seed selection, rice millers and merchants, and District of Agriculture Development Office (DADO) officials. Farmers were identified from these meetings for community level seed production groups by their interest to maintain and multiply *Jethobudho* seeds. Three farmers, Man Bd Sunar and Ganga Giri from Fewa and Kedar Pd Kafle of Biruwa Phants were instrumental in forming the seed production groups. The community level groups were formed into a seed network led by the Fewa Seed Production Group, which provided foundation seed to village level groups for seed multiplication. Official application for registration of Fewa Seed Production Group to the DADO was facilitated by a collaborative group of researchers from the Nepal Agricultural Research Council (NARC), Bioversity International, and Local Initiatives for Biodiversity Research and Development (LI-BIRD).

Varietal release

The Variety Approval, Release and Registration Committee (VARRC) of Nepal[2] carried out field assessments organized both in farmers' fields and on on-station trials in 2005. The assessment included discussions with farmers and mill owners for direct feedback on the produce acceptance.

Statistical analysis

SPSS for Windows version 10.1 was used to produce descriptive statistics of survey data. The ANOVA and Fischer's protected LSD were employed to compare variances and means of *Jethobudho* accessions using MINITAB Release 12.21 and MSTATC.

[2] A technical group of the National Seed Board of Nepal.

Results

The results of the eight years of collaborative work are divided into a five step process: (i) setting breeding goals; (ii) assessing populations for selected traits; (iii) enhancement of local populations for selected traits; (iv) evaluating improved populations; (v) testing improved population against farmers' checks; (vi) seed multiplication and enhancing seed dissemination systems, and (vii) variety release.

Setting breeding goals

Farmers and breeders jointly decided to improve populations of *Jethobudho* landraces because of farmers' expressed interest and availability of intra-varietal variations for useful selection. The 60 surveyed farmers ranked softness (1) and taste (2) before flakiness (3) and aroma (4) as the most important quality traits, for which they would be willing to pay a higher price for *Jethobudho* compared to Basmati rice (Table 4). In addition to these post harvest traits, the focus group discussions (FGDs) identified that more effective tillers, longer panicle length, dense grain setting, absence of leaf and neck blast, and non-lodging as the traits appreciated by the farmers. The presence of awns in *Jethobudho*, a common characteristic of many landraces, was not preferred by farmers and mill owners. In addition, millers specifically identified high milling recovery with a low percent of broken rice grains as an important trait.

Assessing and selecting populations for selected traits

Characterization of 338 accessions in 2000 showed high variation in panicle length, grain number per panicle, grain weight, awn characteristics, disease tolerance (especially neck blast) and culm strength (Gyawali et al. 2005). Of the 338 accessions, 183 accessions- ranked as neck blast tolerant under natural inoculum pressure, lodging tolerant in farmers' management conditions and with plant height shorter than 150 cm.

Organoletpic assessment of the 143 of the 183 accessions collected from seven sites[3] that had sufficient seeds for micro milling, are shown in (Table 5). High overall values, based predominantly on softness, aroma, and brightness of the grain, came from Biruwa and Fewa Phants (Table 5). Forty-six accessions had more than 71% milling recovery and higher than 400 organoleptic weighted score; these accessions were selected for landrace enhancement (Gyawali et al. 2005).

[3] Of the 183 accessions only 143 accessions had sufficient seeds for micro-milling.

Table 4 Setting breeding goals for post harvest quality traits for *Jethobudho*

Traits	FGD[a]	Consumer survey[b]
Aroma	1	4
Taste	4	2
Softness	2	1
Flakiness	5	3
Volume expansion	3	5

[a] Ranking of traits from Focus Group Discussion (FGD) meeting in Begnas village; [b] Mean ranking of 60 individual survey representing farmers, millers, hotel chefs, housewives, retailers and researchers

Enhancement of populations for selected traits

Assessment by plant breeders' along with expert farmers' of agronomic performance in 2002 in two sites (Malmul site -on-farm and Malepatan site -on-station) of the selected 46 accessions showed significant variation for panicle length, number of grains per panicle, and 100 test weights, awn characteristics, neck blast, and culm strength indicating possible genetic gain from selection (Table 6). Variation in plant height and yield were not significant at the 0.05 probability level (Table 6). The lodging tolerance of *Jethobudho* accession was not significantly different but we found significant differences in culm strength and those with strong culm were selected to improve lodging tolerance. The *Jethobudho* accessions were found to be highly variable for volume expansion, water absorption and kernel elongation. This helped us to select for enhanced *Jethobudho* for post harvest quality traits. Six *Jethobudho* accessions out of 46 accessions showed the most desirable post harvest quality traits and their overall performance for disease and lodging tolerance (Tables 7, 8). There were no significant differences among important agronomic traits between the selected six *Jethobudho* accessions (Table 7). Therefore six enhanced accessions were bulked and distributed for participatory

Table 5 Mean scores of organoleptic assessment traits for 143 *Jethobudho* accessions of seven different *Phant* from the Pokhara valley in 2002

Name of *phant*	Type of rice	Colour	Softness	Separateness	Taste	Aroma	Brightness	Overall ranking	Avg. score
Biruwa	2.57[a]	1.88	2.14	2.86	2.00	1.43	1.71	2.14	2.09
Check JB	3.71	3.29	3.14	2.29	4.43	4.29	3.86	4.14	3.64
Arghun	3.14	1.86	1.00	3.00	2.43	3.00	2.00	2.71	2.39
Malmul	3.00	2.29	2.43	3.00	3.29	3.71	1.86	2.71	2.79
Sisuwa	3.14	2.14	2.71	3.14	3.29	3.29	1.71	2.86	2.79
Sugandha-1	4.00	2.57	1.29	4.57	3.71	4.43	1.71	4.00	3.29
Kundahar	3.29	2.14	2.00	3.43	2.57	2.86	2.14	2.71	2.64
Fewa	2.43	2.29	2.29	2.29	2.29	2.29	2.57	2.43	2.36
Charade	2.86	2.14	2.00	2.29	2.14	1.57	2.43	2.57	2.25
Mean ± SD[a]	3.01	2.25	2.21	2.78	2.80	2.8	2.28	2.78	2.61
	±0.40	±0.44	±0.62	±0.44	±0.81	±0.99	±0.70	±0.59	±0.48

Scoring was done on a 1–5 scale, with lower scores representing better quality and higher scores representing poorer quality

[a] Mean and SD does not include the value of Sugandha-1 which is not a *Jethobudho* (JB) landrace. Check JB (JB = *Jethobudho* collected from the market) and Sungadha-1 (a variety developed by PPB project of LI-BIRD) were included as check

varietal selection (PVS; Joshi and Witcombe 1996) as an improved *Jethobudho* variety.

Evaluation of enhanced materials

Tables 7, 8 show the comparative evaluation of six enhanced *Jethobudho* accessions. Significant difference between enhanced *Jethobudho* and farmers' check were found for neck blast and lodging tolerance (Table 7). The micro-milling of selected *Jethobudho* accessions revealed that the length–width ratio ranged from 2 to 2.5 and none of the grains were found to have chalkiness; all were ghee color (translucent; Table 8). Enhanced *Jethobudho* populations were found to be superior in milling recovery (72%), excellent physical appearance and better organoleptic weightage[4] as compared to farmers' own source of *Jethobudho* (Table 8). The selected *Jethobudho* accessions recorded excellent volume expansion (>300%), higher water absorption (>200%), 100% kernel elongation and 23–24% amylase content, good indicator for the consumer preferred trait of flakiness. The predicted response to selection for selected traits was difficult to estimate due to lack of heritability estimate for each trait, however, selection differentials are shown at 4% selection intensity (Table 9).

[4] Cumulative weightage indicates superior organoleptic properties of the accessions.

Farmer's perception of the enhanced *Jethobudho* assessed by participatory variety selection

A total of 260 randomly selected *Jethobudho* growing farmers from Pokhara Valley took part in a participatory variety evaluation of improved *Jethobudho* with their own source *Jethobudho* during 2003 and 2005. Table 10 shows farmers' perceptions on overall performance of enhanced *Jethobudho* rice variety over three years compared to local *Jethobudho*. In 2003, farmers perceived that enhanced *Jethobudho* was better for traits such as blast disease tolerance, lodging tolerance, straw and grain yields, tillering ability, threshability and seedling establishment. However, the enhanced *Jethobudho* variety matured later than the farmers' own *Jethobudho*. Similar trends were found in PVS trials in 2004 and 2005 for most key traits.

Data of 47 PVS trials conducted in Kaski Valley in 2005 showed that the enhanced materials was highly preferred for post harvest grain quality traits, especially for higher milling recovery, aroma, softness, flakiness and other cooking qualities by the farmers (Table 10; Fig. 2).

Molecular evaluation

All bulked and individual samples of enhanced accessions of *Jethobudho* were found similar at the two SSR loci (RM42 and RM223) with the same

Table 6 Mean squares of agronomic traits combined over locations of *Jethobudho* enhancement in Malmul and Agricultural Research Station at Malepatan, Pokhara, 2002

Source of variation	df	PH	TL	PL	GPP	TW	GY	Awn	Leaf blast	Neck blast	Lodging tolerance	Culm strength
Location (L)	1	34434**	28.4**	7.9	148	107**	57.2	110	4.96	0.02	2.0	0.18
Error (a)	4	77.6	0.07	4.9	925	2.7	11.6	28	1.60	4.41	6.2	1.90
Jethobudho (JB)	45	67.9	0.70	3.1**	308**	3.3**	0.44	13**	0.12	0.85*	1.9	0.78**
JB × L	45	56.5	1.14	0.9	288**	1.4	0.57	13**	0.12	0.80*	0.8	0.35
Error (b)	180	53.9	0.83	0.8	160	1.4	0.46	7	0.14	0.55	0.55	0.25

**,* Significant at 0.01 and 0.05 probability level respectively

PH Plant height (cm), *TL* Tiller per hill (Number), *PL* Panicle length (cm), *GPP* Grain panicle^{-1} (number), *TW* Test weight (g), *GY* Grain yield (t ha^{-1}), Awn (0–5 scale), Leaf and neck blast (0–5 scale), Lodging (0–5 scale) and *CS* Culm strength (0–5 scale as indicator for lodging resistance)

Table 7 Means of various agronomic traits[a] of *Jethobudho* measured in Fewa Phant in 2005

Jethobudho accessions	PH (cm)	Till (n)	PL (cm)	GPP (n)	TW (g)	GY (t h^{-1})	Neck blast (0–5 scale)	Lodging (0–5 scale)
JB-T-010-025/5	167	4.7	27.7cd	156.8a	23.58a	2.71	1.66c	1.00b
JB-T-023-030/25	168	4.5	27.3cd	154.0ab	22.75ab	2.98	1.33c	1.00b
JB-T-103-237/12	166	4.8	27.5cd	154.7bcd	23.50a	2.78	1.66b	1.00b
JB-T-105-238/5	170	4.9	27.6ab	151.5ab	23.33ab	2.61	1.00c	1.00b
JB-T-147-296/6	172	4.8	28.2bc	150.6bcd	23.58ab	2.65	1.00c	1.00b
JB-T-168-316/3	170	4.9	27.5a	144.7b	22.58ab	2.69	1.00c	1.00b
Farmers localφ	167	4.8	26.8de	140.4bc	22.33ab	2.64	3.00b	2.00b
Farmers localφ	167	4.3	27.0cd	144.6cd	22.42b	2.54	3.33b	2.67b
Farmers localφ	169	4.5	27.2bc	143.7d	23.33a	2.66	3.33b	2.67b
Farmers localφ	165	4.3	26.4e	140.0e	21.08b	2.58	4.00a	4.00a
Mean	168	4.6	27.3	148.1	22.85	2.68	2.13	1.73
LSD	5.04	0.57	0.87	10.48	0.924	0.36	0.83	1.001
CV %	2.58	10.5	2.7	6.08	3.48	11.6	33.4	49.65

[a] *PH* Plant height, *Till* Tiller per hill, *GPP* Grain panicle^{-1}, *TW* Test weight, *GY* Grain yield, letter followed by the same letter do not differ significantly at 0.05 probability level (DMRT)

φ, Farmers local *Jethobudho* as check variety

alleles as that of the check aromatic varieties: Azucena, Pusa Basmati-1, Rato Basmati local, but different from that of IR36 at these loci (Fig. 3). The IR36 allele was different from those in Azucena, Pusa Basmati-1, Rato Basmati and *Jethobudho* populations for RM223 except an individual DNA sample with big sized allele (shown by arrow in Fig. 3a). The reported size of PCR products for RM223 in Azucena was 150 bp. But RM42 was observed monomorphic with same product size across the *Jethobudho* and check varieties included in the study. *Jethobudho* had 2 alleles for RM223; alleles similar to aromatic checks: Azucena, Pusa Basmati-1, Rato Basmati and other one different from that of Azucena and IR36 (Fig. 3). A dendrogram constructed on the basis of allelic polymorphism using Jaccard's similarity correspond the genetic relationships among the populations for aroma (Fig. 3). All the enhanced *Jethobudho* populations except JB3 (JB-T3-103-237/12) correspond to a high level of similarity coefficient and are grouped into a cluster and show close association with Azucena for aroma traits. *Jethobudho* populations were found with unique bands for most of the markers studied. However the JB3 behaved similarly to that of Pusa Basmati-1, Rato Basmati local and IR36.

Table 8 Disease, micro-milling, and organoleptic quality traits of selected *Jethobudho* accessions in Kaski, 2003

Jethobudho Accessions	Source of materials	Blast (scale 0–5)	Head rice %	TWT of milled rice (g)	Length (mm)	Width (mm)	Water absorption (%)	Volume expansion (%)	Amylose (%)	Organoleptic weightage
JB-T-010-025/5	D.B Karki[a]	1	71	15.3	5.5	2.5	185	300	23.0	427.5
JB-T-023-030/25	M. Subedi[a]	1	71	14.8	5.4	2.4	199	305	24.1	465.0
JB-T-103-237/12	G. Giri[a]	1	68	15.3	5.3	2.4	209	333	24.3	427.5
JB-T-105-238/5	M.B. Sunar[a]	1	72	15.4	–	–	213	313	23.8	427.5
JB-T-147-296/6	K.P Kafle[a]	1	71	15.5	–	–	229	375	23.9	420.0
JB-T-168-316/3	B.P. Baral[a]	1	77	15.7	–	–	252	350	23.7	450.0
Jethobudho under two cycles of selection	143 accessions	1 ± 0.0	71 ± 5	15 ± 0.7	5.2 ± 0.5	2.4 ± 0.1	208 ± 56	302 ± 85	23.6 ± 1.4	431 ± 17
Farmers—non selected *Jethobudho*	10 samples	3 ± 0.94	69 ± 1.8	–	–	–	–	–	–	407 ± 23

Source: Gyawali et al. 2005

[a] They are defined as custodian farmers from whom the original seeds were collected for germplasm enhancement in 1998

"–" Information not available

Seed multiplication and enhancing seed supply systems

Custodian farmers now have enhanced skills for panicle selection, purity, and germination testing. Six of these custodian farmers from where the materials were collected were involved in the initial seed production. A network of three community based seed production groups was set up, made up of six village level seed producer groups. One of the three seed production groups, the Fewa Seed Producer Group, has been registered and formally linked to the District Seed Self-sufficiency Program (DISSPRO) of District Agricultural Development Office, Kaski (Table 11). Fewa Seed Producer Group initiated its seed production of enhanced *Jethobudho* with 80 kg of basic seed on-farm and 1.7 tonnes of truthfully labeled seed produced in 2004. Seeds were supplied to "farmer breeders", those farmers from whom the original seed was collected. The Fewa Group is involved in the marketing of truthfully labeled seed of enhanced *Jethobudho* in Kaski. In 2005, seed producer groups were willing to pay NPR 1600 per muri (70 kg husked paddy) compared to the NPR 1300 for the farmer's variety. At that time, farmers produced 1000 kg of truthfully labeled seed of enhanced *Jethobudho* prior to release of the variety. In 2006 the community seed producer groups collected 3300 kg seed for marketing.

Official release of farmer's variety

Initially, the Variety Approval, Release and Registration Committee (VARRC) of Nepal was concerned on releasing a mixed bulk population of enhanced *Jethobudho* accessions. However, after assessment of the enhanced *Jethobudho* bulk population in the field, and discussion with farmers, mill owners and rice merchants, the VARRC established that the bulk population was phenotypically uniform for agronomic, post harvest quality traits and market preferences and recommended the variety for release in 2005. The enhanced rice variety was formally registered, approved and released by the name of "*Pokhareli Jethobudho*" through the Variety Approval, Release and Registration Committee (VARRC) of Nepal, 20 June, 2006.

Table 9 Estimates of selection differential for important traits in the original population of *Jethobudho* rice evaluated in Nepal

Traits	N[a]	Mean of unselected population (X)	n[φ]	Mean of selected population (x)	Selection differential (s[d])
Blast (0–5 scale)	143	2.43 ± 1.28	6	1.00 ± 0.00	1.43
Awn (0–5 scale)	143	3.72 ± 2.99	6	2.00 ± 2.45	1.72
TGW (g 100^{-1} grains)	143	2.07 ± 0.09	6	2.15 ± 0.05	0.08
Milling recovery (g)	143	113 ± 8.17	6	116 ± 6.65	3.00
Softness (0–5 scale)	143	1.38 ± 0.48	6	1.00 ± 0.00	0.38
Flakiness (0–5 scale)	143	1.30 ± 0.45	6	1.17 ± 0.41	0.13
Aroma (0–5 scale)	143	1.41 ± 0.49	6	1.00 ± 0.00	0.41
Overall cooking quality (0–5 scale)	143	1.41 ± 0.49	6	1.00 ± 0.00	0.42

[a] The number of *Jethobudho* accessions screened for major traits; φ mean of selected six accessions that bulked finally as "Pokhareli *Jethobudho*"

Discussion

Setting breeding goals for participatory plant breeding requires both an understanding of needs and preferences of the producer, miller, and consumer, together with a determination of the potential and limitations of the available breeding materials. The quick targetted survey of farmers and millers, revealed a willingness to pay more for a "set" of post-harvest and organoleptic quality traits (softness, flakiness, taste, and high milling recovery) than a specific single trait such as taste or aroma. The perspective gained from this study showed that farmers did make a trade-off for "a set" of quality traits rather than a single quality trait such as aroma. One of the reasons for employing participatory plant breeding in rice is to capitalize on farmers' skills and knowledge on such trade-off traits that support maintenance of genetic diversity (Joshi et al. 2002; Witcombe et al. 2006a, b). Such qualitative information is difficult to obtain using the current international quality assessment developed by IRRI (1996). Such trade-offs have also been shown by Ceccarelli et al. (2003) for barley, where selection among breeders and farmers were not significantly different for grain yield but were for other traits such as superior height under severe drought, and for maize in Mexico and Honduras where farmers select for a set of post harvest traits as trade offs for yield (Smith et al. 2001).

The current international standard for milled basmati rice is slender (length/width ratio should be 3.0), very long grain (7.51 mm) with high amylose (23–25). The basmati type rice has longer grain and rice remains long and separated when rice is cooked. In contrast, milled *Pokhareli Jethobudho* rice fell into the short grain category (length/width ratio 2.2) with similar amylose content (23–24%). The 23–24% amylase content of *Jethobudho* makes its highly preferable for non-sticky rice and a good indicator for the consumer preferred trait of flakiness. When the soaked rice is cooked, the volume of rice increases by over 300% and length of the kernel doubles. This ability to expand linearly gives a soft quality that retains the characteristics of flakiness (characteristic of cooked rice that does not stick during serving and remains soft while eating). This trait is very good for cooking Pulao or Biryani (soaked rice cooked with vegetables and meat together with ghee and spices), which is common in Arabian and Asian food culture (Rani et al. 2006). Singh et al. (2000) reported that the high export quality of Indian Basmati landraces should have more than 300% volume expansion, thus *Jethobudho* is highly suitable to meet these international standards of aromatic rice for export. The enhanced landrace also has a special aroma, which is distinct from basmati types (Bajracharya et al. 2005). Bajracharya et al. (2005) reported that the enhanced *Jethobudho* rice accessions carry the alleles of the site on the genome as detected in other aromatic varieties in her check, but that these alleles are different from IR36 for RM1 and RM223, establishing the uniqueness of the enhanced *Jethobudho*. Her study also showed that the post harvest grain quality traits were positively associated with aroma.

Table 10 Farmers' perceptions of enhanced *Jethobudho* on preferred traits compared to the local check in PVS[a]

Traits	Better than the local check (%)	Same as the local check (%)	Worse than the local check (%)
Plant height			
2003	62.5 (5)	25 (2)	12.5 (1)
2004	30 (10)	52 (17)	18 (6)
2005	57 (27)	17 (8)	26 (12)
Neck blast tolerance			
2003	89 (8)	11 (1)	0
2004	78.1 (25)	18.8 (6)	3.1 (1)
2005	85.1 (40)	14.9 (7)	0
Lodging resistance			
2003	100 (10)	0	0
2004	88 (28)	12 (4)	0
2005	91 (43)	2 (1)	3 (7)
Grain yield			
2003	70 (7)	0	30 (3)
2004	35 (9)	46 (12)	19 (5)
2005	60 (28)	26 (12)	14 (7)
Milling recovery			
2003	89 (8)	11 (1)	0
2004	33.3 (1)	66.7 (2)	0
2005	87.8 (36)	9.8 (4)	2.4 (1)
Softness			
2003	67 (4)	33 (2)	0
2004	50 (1)	50 (1)	0
2005	85 (34)	13 (5)	2 (1)
Aroma			
2003	83 (5)	17 (1)	0
2004	86 (6)	14 (1)	0
2005	95 (38)	2.5 (1)	2.5 (1)
Flakiness			
2003	50 (3)	50 (3)	0
2004	–	–	–
2005	67.5 (27)	30 (17)	2.5 (1)
Overall preference			
2003	86 (6)	14 (1)	0
2004	56.3 (9)	37.5 (6)	6.3 (1)
2005	88.5 (40)	6.7 (3)	4.4 (2)

Values in the parenthesis indicate number of participating farmers who responded to HLQs

[a] No. of participants that responded PVS feed back: 2003 = 10, 2004 = 33 and 2005 = 47

There is a growing global recognition of the importance of traditional varieties, both as components of sustainable production systems and as sources of genetic variation for modern plant breeding (Gepts 2006; Teklu and Hammer 2006; Jarvis and Hodgkin 2008; FAO 2010). However, even though considerable and unique local crop genetic diversity continues to be available in farmers' fields, the majority of breeding activities use genebank materials, rather than materials currently maintained in the farmers' production system (Fowler and Hodgkin 2004; Sthapit and Rao 2009). The work presented here emphasizes the importance of the direct use of materials collected on farm in plant improvement programs. The use of the rich intra-specific diversity maintained on farm allows for the maintenance of

Fig. 2 Typical grain and cooked rice characteristics of Pokhareli *Jethobudho* (photo credit; Sajal Sthapit)

diversity in some traits but without significant variation for the agronomic or quality traits selected for in the participatory improved materials (Ceccarelli 1994; Ceccarelli et al. 2001; Witcombe et al. 1996; Joshi et al. 2001; Gyawali et al. 2007). In the more developed economies of Europe, the use of on-farm materials is also emerging as method for farmer's union and associations, who question the conventional approaches to plant breeding, so use local materials (Chiffoleau and Desclaux 2006; Finckh 2008; Lammerts van Bueren et al. 2007).

Although selection of *Jethobudho* landrace populations was made against awns, a trait not preferred by farmers and millers, the significant G $\times$ E

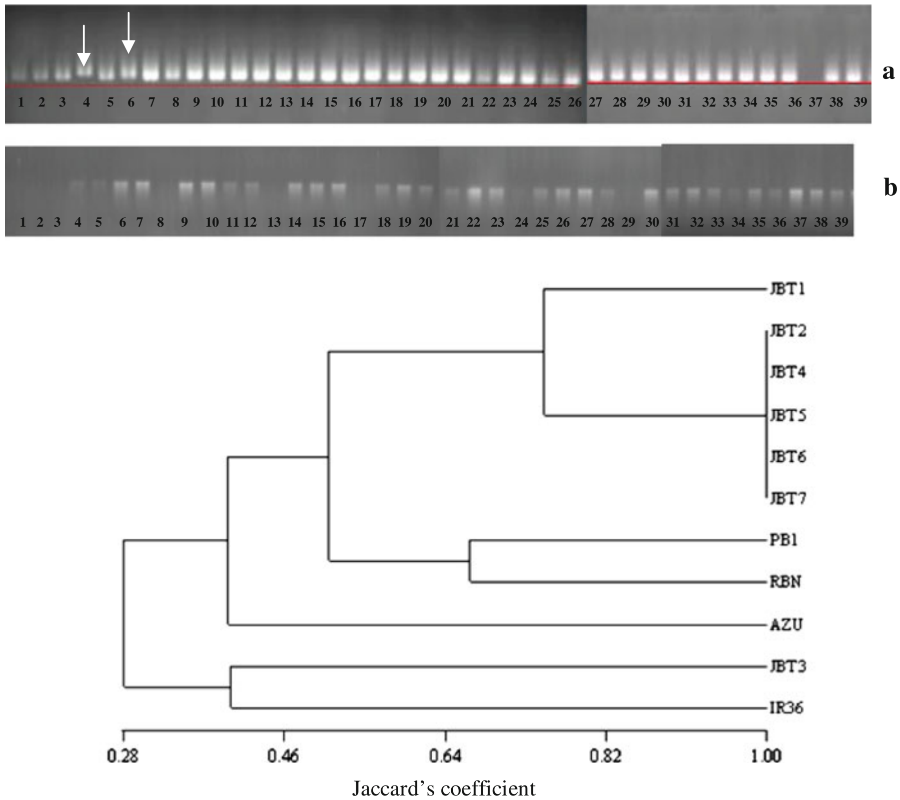

Fig. 3 Agarose gel electrophoresis of SSR products amplified using primers. **a** RM223 and **b** RM42 closely mapped to probe RG28 and linked to fragrance gene (fgr) for aroma in rice. The genetic structures of seven selected *Jethobudho* populations for aroma are compared with aromatic and non-aromatic rice varieties (lanes 1–4 were Azucena, Basmati, Pusa Basmati, IR36, and lanes 5–39 were individual DNAs and bulk DNA of 4 individuals of JBT1-JBT7 enhanced accessions). Dendrogram of seven *Jethobudho* enhanced accessions compared with aromatic and non-aromatic rice varieties based on SSRs associated with trait for aroma using Jaccard's similarity coefficients

interaction indicated a need for location-specific selection by farmers to minimize the presence of awns. Environmental heterogeneity of low-input environments makes it more difficult to apply consistent selection pressure in low-input systems (Haugerud and Collinson 1990; Dawson et al. 2008). Ceccarelli et al. (2003) have shown that environment can have a significant effect on specific agromorphological traits such as growth habit, plant height and kernel weight for barley varieties in Syria.

A limitation to mainstreaming the use of participatory plant breeding products has been the lack of uptake by the formal seed sector to distribute seeds (Ceccarelli and Grando 2007). The majority of participatory crop improvement programmes have relied on farmers' seed production to exchange and disseminate varieties (Almekinders and Elings 2001; Aw-Hassan et al. 2008). These programmes have concentrated on local social institutions (Eyzaguirre and Dennis 2007) and gaining an understanding the ways in which farmers produce, select, save and acquire healthy seeds (Hodgkin et al. 2007; Sperling et al. 2008; Weltzien and vom Brocke 2000; Cleveland and Soleri 2007). Mainstreaming of the PPB projects has been constrained by limited linkages among local or community based organizations and the national seed distribution system. A community based seed production and distribution system for the farmer-bred, state-released, enhanced materials is needed to both strengthen farmer-to-farmer seed systems and link community based seed production with the local seed entrepreneurs (Devkota et al. 2008). In Nepal, the formal seed sector contributes less than 3% of the total demand of rice seed (Baniya et al. 2003; Singh 2003), while the seeds disseminated through the informal seed system follow a diversity of seed exchange networks depending on differences in ecological and cultural differences (Subedi et al. 2003). The Community Based Seed Production (CBSP) program, instigated through this work in Nepal, was designed to integrate community based seed production with the government initiated District level Seed Self-sufficiency Programme (DISSPRO) to create a market for local seed producers. Through this programme the institutional capacity of the Fewa Seed Producer Group was

Table 11 Participatory landrace enhancement program of in situ conservation project in Kaski, Nepal

Product innovation stage	Stage in crop improvement program	Purpose of client participation lead to better orientation	Client participatory techniques that aid research efficiency	Optional and more collaborative techniques
Product design	Setting breeding goal (Specification of landrace and its traits to be improved)	Identify the target clients need (agronomic, post harvest and market traits)	Diversity fair, FCA involved farmers Micro milling, organoleptic assessment with farmers, housewives, rice merchants Physio-chemical properties with food technologists	
Product development	Using existing diversity Assessing diversity for various target population of traits (TPT) Selection for improved agronomic, post harvest quality and market	Used *Jethobudho* (high quality aromatic rice landraces) that meets clients/market need	Farmers participation in *on-farm* experiments on diversity assessments of *Jethobudho* Screening for blast and lodging tolerance in natural biotic and abiotic pressure under target population of environments (TPE) Use local knowledge of farmers, housewives and rice merchants on post harvest quality traits Used Physio-chemical knowledge base of researchers and extensionists	Farmers skills in selection enhanced through training
Product testing	Testing of enhanced *Jethobudho* in on-farm: TPE Testing *Jethobudho* for post harvest quality traits in the market with rice merchants and consumers	Getting feedback of farmers on various agronomic traits Testing *Jethobudho* for post harvest quality traits with the clients and incorporate them in selection process	During PVS, micromilling and Organoleptic tests clients collaborate with the researchers and extensionists Assessing quality aspect with house wives, cooks, rice millers and merchants	Market research with relevant clients add value to the selection process
Product Marketing	Seed and grain supply	To enhance the access of enhanced *Jethobudho* to the clients (Consumer chain)	Community based seed production Linking grain producer to rice millers and merchants	Linking seed initiatives of government and private sector
Consumer analysis	Harvesting benefits for farmers Outcome assessment	To create market incentives to farmers as an encouragement to in situ conservation if genetic resources	Involvement of policy makers in the process to make them aware of in situ conservation initiatives Developing *Jethobudho* case study for *sui generis* systems for Nepal	Linking policy initiatives to community level farmers organizations

strengthened by a joint non-government organization (LI-BIRD) and the local extension service, the District Agriculture Development Office (DADO) partnership that provided regular training, support, and services (Gyawali et al. 2007). The strengthening of the capacity of the local seed producers group was key to the success of the growing demand for the seed of enhanced *Jethobudho* in Kaski valley, as well as in neighboring districts with similar rice production domains.

With the approval of the proposal of enhanced *Jethobudho* release by the Variety Approval, Release and Registration Committee (VARRC) of Nepal in 2006 came the recognition of participatory data generated using PPB by VARRC and the release of the *Pokhareli Jethobudho*. The formal registration and release has both enhanced the access of quality seed of an enhanced bulked variety adapted to local conditions of the Nepali farmers, and improved the recognition of the importance of in situ conservation of crop biodiversity in farmers' fields at local and national levels. In addition, the on-farm evaluation of accessions and use of micro-milling and organoleptic quality traits in *Jethobudho* has not only broadened the breeders' understanding of landrace enhancement initiatives but also helped screening best accessions to identify the superior landraces accessions for quick scaling up through Participatory Varietal Selection (PVS), community based seed production, creating market incentives and developing mechanisms for equitable sharing of benefits from the use of local landraces. It is important to understand that not all landraces can be conserved on farms, and not all farmers can conserve them because of the costs involved (Smale and King 2005). The challenge is to create incentives for maintaining diversity that can benefit both current and future farmers and breeders. Participatory crop improvement, linked with national varietal release and supporting seed production units, can be one method that allows farmers to benefit from their local resources while ensuring the continued maintenance and evolution of crop genetic diversity within agricultural production systems.

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