Review Article

Chapter 1

SELECTION AND TESTING FIELD PERFORMANCE OF CANDIDATE TEA CLONES IN INDIA

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ABSTRACT

The early efforts by tea breeders in India for selecting candidate tea clones from natural variability existing in seed-grown plantations are reviewed. The procedures and criteria in selecting elite clones are described. The genetical and morphological differences between seed and clonal plantations and their complementarity as planting materials are highlighted. The need to conserve valuable tea germplasm present in existing old seed-grown plantations is suggested. The danger of genetic vulnerability of new clonal plantations using few popular clones which is resulting into narrowing of their genetic base, is highlighted.

Keywords: *Camellia, seed, clone, biodiversity, germplasm, selection procedure, criteria, cultivar, tolerance.*

I. INTRODUCTION

Robert Bruce (1823) discovered local type of tea plant in Assam, India, commonly known as Assam type *(Camellia assamica* (Masters) Wight). The possibilities of its commercial cultivation had to wait till 1834 when Lord William Bentinck appointed a Tea Committee to advice the Government on the subject. The early improvement of tea *(Camellia sinensis* (L). Kuntze) in India dates back to 1835 when tea seeds and plants were introduced from China by G.J. Gordon which were initially used to start tea gardens in Assam, the Himalayas and the Nilgiris. After a decade or so the Assam type became the major tea variety of commerce. Another cambod variety *(Camellia assamica* spp. *lasiocalyx* Plankton) was introduced in India from South East Asia in the beginning of 20th. century. These tea species growing side by side crossed freely among themselves and thus produced highly heterogeneous early populations. These diverse populations which were utilised by the earlier tea growers and later on by the tea scientists to develop seed populations and candidate clones (Wight 1948, 1956, 1958 a, 1961 a, b; Barua 1963; Barua 1963 b; 1989, Bezbaruah, 1967 a, b; 1968, 1969, 1974, 1982 and Singh, 1980).

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II. BIODIVERSITY IN INDIAN CAMELLIAS AND ITS EXPLOITATION

(i) Status

India can claim to be one of the pioneers in scientific approach to tea cultivation in the world. Organised efforts to collect tea germplasm in India began after the establishment of the Scientific Department of the Indian Tea Association and its experimental station at Tocklai, Jorhat, Assam in 1900 (Singh, 1984). Today, Tocklai Experimental Station, Jorhat can claim to have the largest single collection of Camellia germplasm anywhere in the world (Singh and Bera, 1994). The collection is represented by 14 species with 2507 accessions maintained at three main centres, i.e. Jorhat, Nagrakata and

Table 1. Camellia germplasm maintained by Tocklai Experimental Station in N.E. India

Species	Source of collection	No. of accessions
Camellia assamica	Assam, Manipur, Sri Lanka, S. India	2337
C. sinensis	China, Darjeeling Hills	35
C. assamica ssp *lasiocalyx*	Indo China, Myanmar, Assam	60
C. kissi (drupifera)	Meghalaya	50
C. caudata	Assam	–
Eurya japanica	N. E. India	7
E. acuminata	N. E. India	2
Gordonia excelsa	N. E. India	2
G. imbricata	Sri Lanka	2
C. japonica	U.S.A., Japan	–
C. sasanqua	U.S.A., Japan	–
C. irrawadiensis	Upper Myanmar	2
C. japanica var. Kyoniski	Japan	1
C. rosiflora	Sri Lanka	1
Total		2507

Source: Singh, I.D. & Bera, B. 1994: Indian Journal of Plant Genetic Resources 7(2): 125-131

Darjeeling (Table 1). Maximum diversity is shown in *Camellia assamica* followed by C. *assamica* ssp. *lasiocalyx,* mostly collected from the north east region of India.

(ii) Considerations

Discovery of a simple method of vegetative propagation of the tea plant paved the way for improvement of tea on scientific lines. The method was discovered in the early thirties (Tunstall, 1931) but its standardization for large-scale use in tea estates and for developing selection procedures for clones took nearly 15 years (Barua, 1989). This encouraged tea breeders to develop clonal varieties like in many fruit crops. This has been a major breakthrough responsible for the development of clonal varieties of tea in N.E. India.

In selection of clones objectives like yield, cup quality, tolerance to biotic (pests, diseases) and abiotic (drought, water logging, etc.) stresses prolonged harvesting season are the criteria depending upon the agro-climatic conditions.

Elite clones and improved strains of seeds superior to the existing seed populations can be used for commercial cultivation. Clones can be selected from the commercial populations of mature hybrid tea or from progenies of artificially produced hybrids.

(iii) Seed Vs Clone

Barua (1963) had reported adaptability to be the basic difference between clone and seed progeny, "a seed population composed of a large number of genetically distinct units is elastic and can be fitted into a wide range of cultural and environmental conditions without much change in its overall performance. On the other hand, thousands of bushes of a clone widely separated in space and time behave, in most ways, as a single bush. Consequently, a clone lacks elasticity which makes it more selective of environment and cultural treatment" (Barua, 1989). Thus, a clone is expected to be more sensitive to any change in soil and climate than a seed population. In the light of above differences, many advantages and disadvantages of seed *vs* clone and vice-versa are described by Barua (1989). Their commercial cultivation will depend on many considerations. In general, they complement each other, hence the choice of one or the other is not the question. Wight (1956) had suggested the use of three to five clones for planting one fifth of an estate. Later on, as a long term strategy, it was advocated that clone and seed should be planted at 1:1 ratio in an estate. No single clone should occupy more than 10% of the estate area (Bezbaruah and Singh, 1980)

(iv) Complementarity between clone and seed

Elite clones are selected from existing natural genetic variability or created genetic variability. Their commercial multiplication is done by vegetative propagation (asexual reproduction) which is a dead end to evolution. Seed is the product of sexual reproduction, which is the natural process of evolution. It is this feature of the seed which provides variability for the selection of clones.

The choice of tea growers for clone or seed depends on many considerations as mentioned above but the race for development of clone and seed varieties will continue (Barua, 1963), one complementing the other.

Visser (1969) recognises the limitation of clonal selection from existing tea fields and stresses the importance of breeding for overall genetic improvement of tea. The time required to breed a seed variety or clonal variety varies in different tea growing countries.

It will depend on the state of the improvement programme, objectives, methodology of evaluation, selection criteria, growing conditions and the state of urgency. Considering the present state of tea plantation in the world and current tea improvements, total displacement of seed by vegetative clones can possibly be ruled out. It is more likely and desirable that development of seed varieties and selection of clones should proceed simultaneously. If present trend in India, the largest producer of tea in the world, is any guide, then given seed and clone varieties of similar merit, most growers will prefer seed varieties (Barua, 1989).

(v) Selection of clones

a) Genetic variability

The exploitation of wide genetic variability present in the early tea populations due to free hybridization among cultivated tea species was adopted as one of the early strategies for tea improvement (Wight, 1956; Singh and Bezbaruah, 1988 and Singh, 1994). In seed grown populations of N.E. India, about 10% of the bushes produced only 20% of the total crop and about 0.9% bushes produced as much as or more than 300% of the yield of an average bush possibly due to their inherent superiority in yield (Wight, 1939). Furthermore, Wight (1956, 1958a) pointed out that one out of every 40000 bushes from the old seed-grown populations may be selected as golden bush. In a later estimate Singh (1989) estimated it to be 1:80000.

Early improvement of tea in N.E. India was based primarily on mass selection, line breeding and clonal selections. Mass selection (Wight, 1956) failed to produce desired results. Hence, it was replaced later on by line-breeding (Wight, 1961a; P.K. Barua, 1963) where every selection was maintained separately to establish a new seed bari from which further selections were made. It was further extended to produce biclonal seed varieties. Based on the nature of genetic variability described here in the N.E. Indian tea plantations, the tea improvement scheme is commonly referred to as the "clone-seed cycle" (Figure 1).

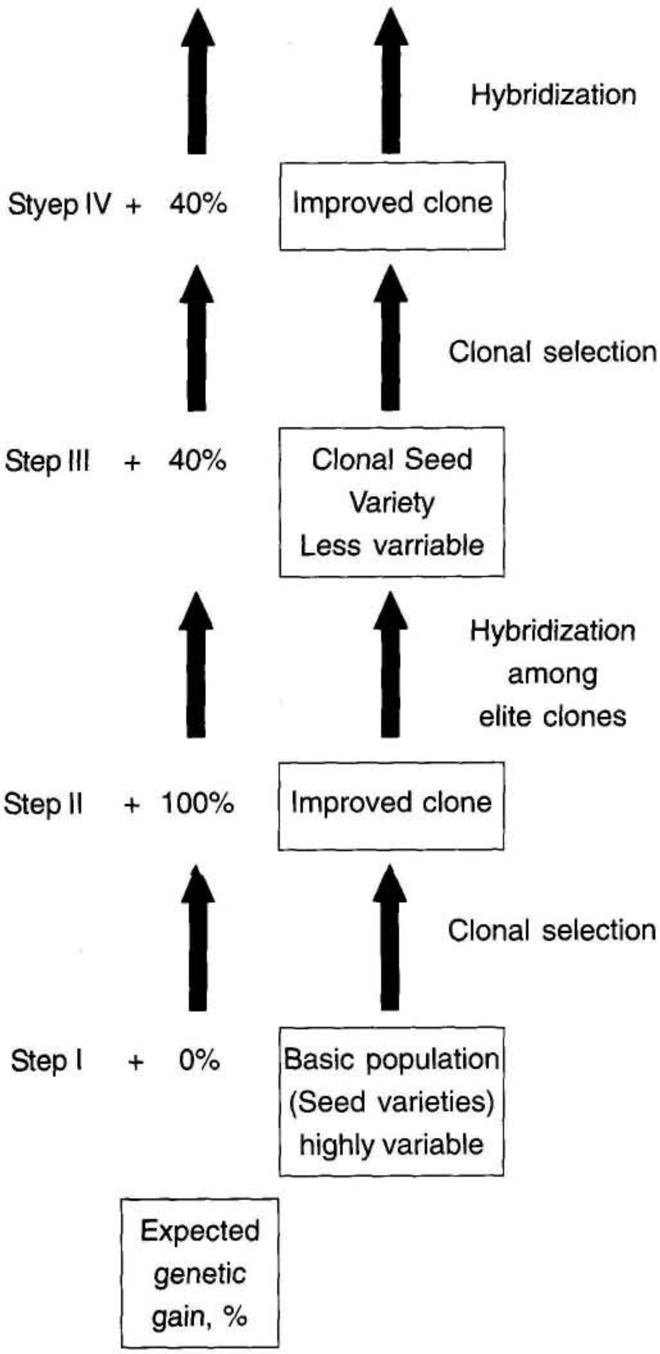
b) Selection criteria for yield

1. Mature bush yield

Based on various studies, many selection criteria have been established and have been used widely in the selection of elite clones. Some of these are described below:

(i) *Size or surface area:* Visser (1961) recommended size or surface area of the bush could be used for estimating yield. A positive correlation was observed between pruned or plucked surface area and yield of individual bushes (Cohen Stuart 1929, 1930, Visser 1969; Satyanarayana and Sharma, 1982)

Fig. 1: A schematic diagram of the "clone-seed cycle" of tea improvement.



(ii) *Leafiness:* Leafiness or total leaf area on a bush could be used as a criterion of yielding capacity. According to Barua (1961 a) a positive correlation between yield and area of mature leaf on a bush can be expected since young shoots harvested as crop develop mainly at the expense of food manufactured by the maintenance foliage and its reduction causes a proportional loss of crop. Visser (1969) obtained a significant positive correlation between total leaf area per bush and yield of shoots plucked from two groups of clones.

(iii) *Leaf size:* Leaf size which is related to size and weight of shoot, has been used as an yield criterion. Positive correlations have been reported between size of leaf and yield of tea in U.S.S.R. (Mamedov, 1961) in South India (Satyanarayana & Sharma, 1982); leaf size, leaf number and yield in Japan (Toyao, 1966) and length of the growing shoot and yield (Amma, 1975). However, these correlations were found only in bushes with relatively small leaf China & China – hybrid population. No such correlations were observed in large leafed bushes (Bezbaruah, 1969b; Visser, 1969). Since a bush with large leaf and big shoot has another advantage apart from yield, i.e. less plucking cost, weightage may be given to large sized leaf bushes in clonal selection provided other parameters permit.

(iv) *Plucking point density:* Plucking point density is one of the most important criterion for quick screening of mother bushes by eye judgement. It gives an estimate of the number of shoots actually plucked from a bush.

Barua and Dutta (1971) had reported that maximum density of plucking points occurs in the central zone of a tea bush and number of shoots get progressively thinner towards the periphery. It was found to be true in all bushes irrespective of their vigour. Barua, (1965) had also found good contribution towards high density of plucking points from the following morphological characters of a tea bush:

a) Thickness and distribution of pruned sticks

b) Bud break per pruning stick

c) Evenness of flush

(v) *Pruning weight:* Pruning weight among various growth parameters like number of plucking points, number of pruning sticks, weight of tipping and number of primaries tipped was found to be a fairly reliable criterion for bush yield (Anon., 1961 and Visser, 1969; Satyanarayana and Sharma, 1982).

In colder climates, tea remains dormant and bud break is determined by temperature and day length. Early shooting and flushing in spring was observed to be a good selection criterion as such bushes yield higher than the control (Toyao, 1964). For frost hardiness, late sprouting which is associated with resistance to cold, was found to be a good selection criterion (Harada, 1956). Generally, such bushes grow weaker. It was suggested by Visser (1969) to pre-select the best ones on the basis of the growth of their cuttings in the nursery.

2. Selection of seedlings for yield

Among the various correlation studied to predict the yield of a mature bush based on seedling growth of 1 to 4 years of age, the shoot: root ratio of 4 year old plants was found to be the best and consistent (Othieno, 1982). Less than two years old seedling plants are unlikely to provide reliable information about the predictability of potential clones.

3. Correlation between yield of a bush and its clonal progeny

No consistent correlations were found between the yield of selected mother bushes and their clonal progenies (Visser, 1969; Green, 1971). Since morphological criteria have failed to give reliable indication of yield, both of mature bushes and young seedlings, it is highly important to *test various* anatomical, physiological, and chemical parameters for the assessment of yield. The best way to assess the yield of selected clones is to conduct field trials for 2-3 pruning cycles.

4. Selection criteria for quality

Single criteria for a rapid identification of tea bushes having superior quality are scarce.

Some morphological characters showing positive correlation with quality of various degrees have been used in selecting elite clones.

a) *Pubescence:* Hairs on the under surface of the leaf are called pubescence, which were found to be positively correlated with quality (Wight and Barua,1954, Wight and Gilchrist, 1961, Wight *et al.*, 1963, Venkataramani *et al.*, 1964). Hairs themselves do not have a causal connection with quality (Wight and Gilchrist, 1961). In orthodox manufacturing of tea, they produce tips. However, in the tea manufacture, pubescence had no correlation with quality (Anu. Rep., Tocklai 1964).

b) *Colour of leaf:* Light green leaf of Assam type produced better tea quality than the green leaved types (Wellensiek, 1947; Venkataramani, 1964). However, Wight *et al.* (1963) had ascertained an optimum greenness for high quality. The darkest green and particularly palest green leaves produced poorer quality than those of medium green ones. Results reported herein clearly show that leaf colour alone can not be taken as an indicator of quality.

In search of criteria for quality and strength, Wight (1954) had reported pubescence, phloem index and vascular index to be positively associated with the quality. However, these factors are subjected to large phenotipic or environmental variations (Visser, 1963: Barua, 1989).

c) *Chloroform Test:* This test was developed to measure the fermentation of black tea and is widely used in many countries to screen the bushes for their fermentability (Sanderson, 1963; Toyao *et al.*, 1971; Ellis,1964; Richards, 1967; Toyao & Katsua, 1972). In this test, the colour of the leaf changes from green to brown. The rapidity of colour change indicates faster fermentability.

Selection procedure

The selection of clones is a laborious process and usually takes 7-10 years before a clone could be released for commercial planting (Barua, 1963a; Bezbaruah, 1968, 1974; Singh,1996). Details of steps involved in selection are given in Figure 2 (Singh, 1996). The initial selection is done mainly by eye estimation of the bush size, frame, and plucking point density and subsequent selection is based on rooting response, yield and quality (Barua, 1965; Singh, 1996). Later on, a simplified method of clone selection based on the vigour of the mother bush as well as their offspring in the nursery was developed and adopted (Bezbaruah, 1984). In addition to yield and cup quality, clones are also assessed for their tolerance to pests, diseases, water stresses (drought, water logging) and rooting behaviour.

It is not possible to outline a rigid selection procedure for all regions because of differences in the kind of tea made, the type of bushes grown and soil-climatic environments. For example, the requirements for the production of orthodox tea will be somewhat different from those of cut leaf manufacture. Selection for given tea quality will be entirely of a different nature. Similarly, in some regions flavour is the main selection criteria whereas in some region tolerance to disease and pest is the main criteria. Therefore, selection procedures suggested below for clonal selection in mature fields and seedling progenies be taken as guidelines which may need modification to suit local requirements.

a) Selection of clones in mature fields

Select old seed-grown, hybrid *jat* populations for selection of elite clones. The chances of getting outstanding clones are more in highly heterogeneous populations than those sections having morphologically more uniform bushes. Ideally, the section should neither be two young nor too old.

The selection unit should contain approximately 40000 bushes which can be sub divided into smaller units if necessary for the sake of convenience. The unit will have to be much bigger if, in addition to yield and quality, selection for disease and pest resistance is also envisaged (Barua, 1989).

Field selection is best carried out in stages using one rejection criteria at a time. It could be started at anytime of the year using the criteria appropriate for that state of growth. However, it is preferable to start selection with screening for density of plucking points where the plucking table is full. In case of longer pruning cycle, it should be delayed to 2nd or 3rd year from pruning to allow time for the filling of the whole plucking table. In annual prune bushes, selection should be done towards the end of the plucking season (Barua, 1989).

Bushes should be selected on the basis of various criteria as have been already described earlier. The step by step procedures involved upto the release

Fig. 2: Procedure of clonal selection in tea (After Singh, 1996)

Release of clone as new planting material

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Establishment of long term trial

(With selected clones and control(s) in R B D (randomized block design) with three repeats having equal number of plants (24) per repeat)

* Bring up the young plants per recommended practices viz. mulching, centering, re-centering etc.
* Guard against weeds, pests and diseases.
* Maintain crop records clonewise and repeatwise for 3 consecutive years starting from 2nd year and compare with control(s).
* Arrange manufacture and quality assessment of each clone separately and compare with control(s).
* Record weighment of pruning litters clonewise and repeatwise at the end of 3rd year and compare with control(s)
* Observe and record resistance to drought, diseases and pests.
* Record morphological characters of each clone.
* Ascertain the promising clones on the basis of above observations and trials.

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Retain mother bushes and reject others

* Maintain crop record of each bush individually in every plucking round and convert the total crop to yield per unit area of the bush.
* Prune and clean the bush frame properly.
* Compare growth records with control(s).
* Process green leaf in miniature manufacture and assess for quality.
* Allow the plant to grow un-disturbed.
* Plant cuttings bushwise and record number of cuttings per bush.
* Compare rooting ability with control(s).

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Select mother bushes with proper markings

Selection criteria

* Health, vigour, flushing behaviour and duration, free from diseases and pest, plucking point density, distribution of plucking points, leaf size-medium, leaf colour-intermediate, least presence of root suckers, preferably compact frame, erect or semi-erect leaf, position-medium, shoot size-intermediate, shoot colour, higher pubscenes, least floriferous.

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Old hybrid tea population

(Preferably under light prune or deep skiff) of a clone have been given briefly in Figure 2. The procedures adopted by plant breeders at Tocklai Experimental. Station, Jorhat, Assam in N.E. India (Barua, 1989) are described below.

i) Reject very small and branch canker affected bushes. Leave bushes bordering roads, paths and drains.

ii) Selection for density of plucking points. Retain about 5%, i.e. 2000 out of 40000 bushes of the selection unit at this stage.

iii) Select for pubescens, if planned. Depending on the type of tea, retain about 1000-500 bushes.

iv) Prune selected bushes and record the weight of prunings of each bush. Reject bushes giving less pruning weight than the average of all pruned bushed. Retain about 400-200 bushes.

v) Estimate pruned surface area and then weight of pruning per unit area of the bush. On this basis reject bushes having less than the average weight. Retain about 200-100 bushes.

vi) Allow majority of primary shoots on selected bushes to grow to tipping height and record bushwise weight of the tipping. Reject bushes having below average recovery. Retain about 100-50 bushes. If necessary, select for leaf size at this stage.

vii) Conduct replicated rooting trial by taking cuttings of selected bushes, preferably in nursery beds to judge the rooting style and success in rooting. Select 40-20 good rooters.

viii) Bring such selected mother bushes into bearing. Conduct miniature manufacturing trial from plucked shoots bushwise and taste them for cup characters atleast on eight occasions, spread over the growing season. Retain 20-10 bushes with above average quality.

ix) Prune selected mother bushes to take cuttings. Propagate and record rooting success again. Insure sufficient plants to establish a clonal field trial. If not, repeat propagation to produce sufficient plants.

x) Establish replicated long term field trial (minimum 3 repeat) along with 2-3 control in different soil-climatic regions.

Different selection criteria described earlier like chloroform test for fermentability, tolerance to pests, diseases and drought etc. may be undertaken, preferably at early stages of the clonal trial.

Adopt all standard field practices followed in the region in conducting the clonal trial.

Record plucking weight of each round for two pruning cycles. At pruning, record weight of the prunings.

Conduct CTC and Orthodox manufacturing and testing trials where sufficient leaf is available for 2 years.

Select clone(s) finally based on yield, cup characters, response to cultural treatments and adoptability to various growing conditions.

By adopting the procedure described above, in warm climates sufficient information can be obtained in 5-6 years after establishing the trial whereas in cold climate it may take 8-10 years. From the start of the selection programme to the final selection of the clones, it may take 10-15 years.

b) Selection of clones in the nursery

Grow seeds of hybridization programme at wider spacing in nursery beds to allow free growth of seedlings for 2 years before starting any selection. Firstly, screen seedlings by eye judgement for stem girth at collar region and height. Check for pubescence. Conduct Chloroform test at this stage.

Prune selected seedlings and weigh prunings. Retain selected seedlings which recover well from pruning. Conduct rooting trial of selected bushes by propagating their cuttings.

Screen plants on the basis of their rooting ability.

Propagate selected plants to establish clonal proving trial and observation plots. The remaining procedure from here onward is the same as described earlier under the selection of mature bushes (Barua, 1989).

c) Development of clonal cultivars

Tocklai Experimental Station, Jorhat, Assam in N.E. India can claim to be the pioneer in the development of clonal cultivars of tea. Following the clonal selection technique described herein, over 165 clones (Table 2) were released for commercial cultivation in N.E. India (Singh, 1984, 1992, 1999),40 in S.India (Sharma and Satyanarayana, 1987,1989) and two in Himachal Pradesh (Singh, R., 1989). While developing these clones natural as well as created genetic variability has been used. Bulk of these clones (197) were selected from natural variation found in the old plantations of seed jats.

Singh and Handique (1991) were successful in selecting clones (Table 3) tolerant to drought and water logging from old seed-grown sections.

From plantation area of over 7015 ha, 110 clones were selected, of which 105 were tolerant to drought and remaining 5 were tolerant to water logging.

Table 2. Release of clonal cultivars in India

Region	No. of clones selected from		Total
	Natural variability	Created variability	
N.E. India			
Plains	128	8	136
Darjeeling Hills	27	.2	29
Total	**155**	**10**	**165**
S. India	40	-	40
Himachal Pradesh	2	-	2
Grand Total	**197**	**10**	**207**

After Singh, 1994

Table 3. Development of clones tolerant to drought and water logging in N.E. India

Particulars	Selection against water-stress		Remark
	Drought	Water logging	
Plantation area surveyed, ha	6937	78	Assam, West Bengal & Tripura
No. of tolerant clones selected	105	5	Morphological characters and field Ratings

III. SUMMARY AND CONCLUSION

The exploitation of natural genetic variability in cultivated *Camellias* to select elite clones in India as well as elsewhere has paid handsomely. The procedure of simple selection by the process of elimination at various stages though laborious, has been quite effective in identifying elite clones. The techniques were found suitable during the early years of the tea industry. Since it takes 10-15 years to select a clone, which is quite long, it needs to be reduced by developing more efficient selection criteria. The modern techniques of biochemistry, physiology and biotechnology may be used for early screening of selected clones.

Limited efforts have been made so far by the tea breeders all over the world to create genetic variability through recombination, mutation, polyploidy and tissue culture. It should be explored as old seed-grown jats are being rapidly replaced with few high yielding and rarely good quality clones whose cultivation is a dead end to evolution.

Selection of seedlings for yield and other attributes holds a great potential and should be exploited for genetic improvement of tea. Genetic markers for identifying superior clones in the population may be introduced.

As in the past, selection procedures described herein, have been quite effective in selecting candidate clones, the same will also play a major role in evaluating clones developed by biotechnological tools as well. Thus, it needs further refinement for improving the efficiency in selection.

Field assessment of selected clones needs considerable improvement in the areas of field – plot techniques so as to reduce the long period (10-15 yrs) of assessment besides meeting the requirement of large quantity of planting materials.

Clonal plantings are gaining more popularity in major tea growing countries, where old seedling teas are being uprooted and replaced with a few popular clones. It is leading to narrowing of genetic base of tea plantations. It needs to be arrested by preserving *interesting bushes in* the old – seed grown sections besides planting diverse clones.

Since tea is an industrial crop, limited financial supports is being given by national as well as international funding agencies to take up conservation work. This scenario needs to be changed towards adequate funding by the industry so that our tea germplasm could be conserved before we lose them for ever.

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