**Breadfruit**

**Definition:** Breadfruit from *The Hutchinson Unabridged Encyclopedia with Atlas and Weather Guide*

Fruit of two tropical trees belonging to the mulberry family. It is highly nutritious and when baked is said to taste like bread. It is native to many South Pacific islands. (*Artocarpus communis* and *A. altilis*, family Moraceae.)

**Summary Article:** Breadfruit from *Comprehensive Biotechnology*

Breadfruit, *Artocarpus altilis* (Parkinson) Fosberg, has been a staple food and traditional crop in the Pacific for more than 3000 years and is widely cultivated in the Caribbean and other tropical regions. Distribution of breadfruit began in the late 1790s under the leadership of Captain Bligh in his voyages beginning with the *Bounty* and, eventually, successfully transporting 678 plants to Vincent and Jamaica. Recently, methods have been developed for large-scale propagation and mass production of breadfruit plants for worldwide distribution. The regenerated breadfruit plants have been found to be free of bacterial, fungal, or virus contamination, and are vigorous and rapidly growing. The distribution of these plants provides the first opportunity for large-scale development of the crop for food security and commercial products.

**Keywords**

- *Artocarpus altilis*
- Breadfruit
- Food security
- Micropropagation
- Phenolics
- Tropical plant

**4.18.1 Introduction: Breadfruit (*Artocarpus altilis*)**

Breadfruit (*Artocarpus altilis*, Moraceae) has been used for more than 3000 years by Pacific islanders as a staple food and traditional crop and has recently been identified as one of 35 priority crops listed in the International Treaty on Plant Genetic Resources for Food and Agriculture (http://www.planttreaty.org/). Furthermore, The Global Crop Diversity Trust (http://www.croptrust.org) has initiated a conservation strategy that includes breadfruit as one of the plants of importance for food security worldwide. There is a long history of the use of breadfruit for food security. In 1769, Sir Joseph Banks, traveling with Captain Cook to Tahiti, recognized the potential of breadfruit as a food crop for other tropical regions. His observation that “... if a man plant ten [breadfruit] trees in his life, which he can do in about an hour, he would completely fulfill his duty to his own as well as future generations,” prompted the infamous voyage by the HMS *Bounty* to collect breadfruit plants for introduction as a food source for the British colonies in the Caribbean [1]. Unfortunately, the mutineers tossed the original collection of breadfruit plants from Tahiti into the ocean; however, Bligh made a
subsequent voyage that was successful and breadfruit has been cultivated in the Caribbean and other tropical regions since the late 1700s [15].

Breadfruit is a member of the Moraceae (mulberry/fig) family and is related to jackfruit (*Artocarpus heterophyllus*). The tree can grow to a height of over 20 m, but is often pruned to maintain a shorter stature in cultivation. It is well adapted to the wet tropical regions of the world, doing best in climates that receive more than 1500 mm of rain per year and temperatures that remain above 15 °C. Nutritionally, breadfruit is a good source of protein (Jones et al., 2010a) [5], iron, calcium, and other minerals (Jones et al., 2010) [6], and several important vitamins [15]. The versatile and nutritious fruit can be cooked and eaten at all stages of maturity; the fruits can be roasted, baked, boiled, dried, pickled, fermented, and made into flour ([15]; Jones et al., 2010c) [7]. Prepared breadfruit has a moderate glycemic index and there are multiple nutritional benefits to including breadfruit as a dietary staple [16]. Breadfruit trees also provide medicine, insecticides, adhesives, timber, and shelter for their growers, and are a primary component of traditional agroforestry systems in Oceania [15, 18].

Dramatic cultural and environmental changes in the Pacific Islands following World War II have had serious impacts on breadfruit germplasm. Although it is still a widely used staple crop, the cultivation and use of breadfruit is declining in many areas, and a number of cultivars have already disappeared or are becoming rare [15]. Hurricanes and other devastating tropical storms and droughts have contributed to the demise of breadfruit trees and diversity. Since 2000, countless breadfruit trees have been damaged or destroyed by hurricanes in Chuuk and Yap in the Federated States of Micronesia, Guam, the Northern Mariana Islands, American Samoa, Niue, Samoa, Vanuatu, the Cook Islands, as well as on islands throughout the Caribbean.

In spite of the potential of breadfruit as a reliable food source for many parts of the world, distribution of the crop has been limited by difficulties in propagating and transporting trees. Recently, our lab in collaboration with the National Tropical Botanical Garden (NTBG, Hawaii, the United States) and Global Breadfruit (www.globalbreadfruit.com) developed processes for plant tissue culture, mass propagation, and distribution of the trees that solved these problems [12, 13, 19]. We now anticipate that between 2 and 5 million trees will be planted in the next 5 years in not-for-profit projects to produce local food for food security and for-profit plantations to produce foodstuffs for world markets. Several memoranda of understanding have been signed between the program and the governments of the islands of origin of the breadfruit germplasm for equitable benefit sharing, conservation, and sustainable use of the crop (http://ntbg.org/breadfruit/).

### 4.18.2 Traditional Propagation of Breadfruit

The traditional methods of propagating breadfruit plants were passed through generations in the remote islands of Oceania. Traditionally bred cultivars produce few seeds, which have poor viability, are produced by outcrossing, and, therefore, do not retain the genetic character of the parental plants. The most common method of propagation used by early islanders was to make root cuttings or to excise adventitious shoots from the roots of mature trees (http://www.agroforestry.net/tti/A.altilis-breadfruit.pdf). However, using root cuttings poses several problems for large-scale international distribution such as:

1. the limited number of roots that are available from each tree means that there is always a limited number of plants;
2. plants produced from root cuttings generally have a poor survival rate and have difficulty becoming acclimatized and established in new environments;

3. bacterial and fungal infections can cause the young propagated plants to die and can spread diseases between distant regions; and

4. the worldwide distribution of the trees requires that propagated plants travel long distances, where root cuttings would have to be quarantined for extended periods, and may not pass agricultural inspections.

An alternate approach is the use of air-layering techniques ([http://www.agroforestry.net/tti/A.altiis-breadfruit.pdf](http://www.agroforestry.net/tti/A.altiis-breadfruit.pdf)), in which a branch of the tree is scored to remove the outer bark and packed with moistened sphagnum moss for formation of roots. Like the root cuttings, there are many limitations to this approach such as: (1) only very few air layers can be started without causing damage to the parent tree, (2) scoring weakens the branch which can then be snapped or broken during high winds, (3) about half of the air layers do not form roots but form a hardened callus instead, (4) if successful, it is a time-consuming and labor-intensive process, taking 2–4 months to form roots on each air layer, and (5) all of the import and transportation restrictions of agricultural inspections apply to whole plants generated through air layering. Another approach that has been attempted is the grafting of breadfruit scions onto rootstocks of the related species breadnut (*Artocarpus camansi*) which has been reported to have a better than 80% success rate for varieties in Sri Lanka [5].

### 4.18.3 Advantages of Micropropagation of Breadfruit

Plant tissue culture or micropropagation offers solutions to many of these problems and is a long-proven method to establish, propagate, and distribute a wide range of different plant types. Micropropagation is the process by which each individual plant cell can be induced to form a whole plant, a phenomenon known as totipotency [8]. Mass propagation and regeneration of any species relies on the fundamental principles of the regulation of plant growth and development [20]. In this fundamental hypothesis, the redirection of plant growth is primarily dependent on the relative concentrations of two classes of phytohormones, namely, auxin and cytokinins, with other growth substances essential for the process in some species. Over the last 50 years, this hypothesis has been validated with innumerable experimental systems and has formed the basis for modern plant breeding and genetics programs [14]. However, the implementation of micropropagation techniques for tropical species in general and breadfruit in particular has been difficult. Problems with micropropagation of breadfruit have included oxidative browning of the tissues, persistent fungal and microbial contamination, and relatively poor survival of plantlets during weaning and acclimatization [17, 19, 21]. Adaptation of standard micropropagation techniques for tropical species such as breadfruit is required for large-scale production and these techniques have now been optimized for bioreactor production of several varieties of breadfruit plants.

### 4.18.4 Micropropagation and Bioreactor Production of Breadfruit

Micropropagation protocols proceed through a series of stages first described by Murashige in 1974 [10] and later modified by Debergh and Maene (1981) [3]. The principal objective of any micropropagation system is to produce disease-free, true-to-type plants, beginning with a mature stock plant (stage 0) and proceeding through a process of cell de-differentiation and re-differentiation.
to produce an entirely new plant (stage 4) [2].

4.18.4.1 Stage 0: Selection of Stock Plants, Pathogen Indexing, and Maintenance under Quarantine Conditions

The major difficulty in establishing a micropropagation system for breadfruit has been stage 0, establishing the cultures. Tropical trees have high rates of bacterial and fungal contamination, tissue browning, and differential requirements to induce plant growth [12, 13, 17, 19, 21]. For successful establishment of de novo plants in culture, explants are collected in the field at Hana, Maui, surface-sterilized using a triage field sterilization kit containing isopropyl alcohol, soap, bleach, and sterile water, and shipped to the tissue culture laboratory in British Columbia (Figure 1(a) and 1(b) [12, 13, 19]).

Breadfruit explants, especially when taken from mature trees, exhibit a high rate of contamination even after surface sterilization, and opportunistic microbes that consume the tissue when the explants are stressed are present [17]. After testing numerous antibiotics and fungicides, we determined that chemical treatment has only limited efficacy and the most effective antimicrobials cause severe damage or death of the explants. In general, buds are surface-sterilized with 70% ethanol for 1 min and 10% commercial bleach (5.25% sodium hypochlorite) for about 15 min before washing several times with sterile distilled water [19].

Figure 1 Micropropagation of breadfruit, Artocarpus altilis: (a) young breadfruit meristems used as explants; (b) a young sanitized breadfruit explant in tissue culture; (c) a breadfruit explant after the successful induction of regeneration; (d) mass propagation of breadfruit plants in tissue culture; (e, f) breadfruit plants growing in tissue culture; (g) developed breadfruit plants transferred to rooting medium; (h) a rooted breadfruit plant; (i) breadfruit plants produced in a larger scale bioreactor system; (j) a breadfruit plantlet that has been acclimatized; and (k) rapidly growing established breadfruit plants produced through tissue culture.

Tissue browning and oxidative stresses, as well as a latex exudate from the tissue, have increased the difficulty in establishing breadfruit in tissue culture. This is especially problematic with apical meristems that are found under several layers of plant tissue. Upon excision, the meristems have not yet developed chlorophyll and are pale in color. Shortly after surface sterilization, these explants begin to discolor and release what are presumably phenolic compounds into the media. This phenomenon has
also been observed frequently in larger adventitious buds; thus, small buds that are in their early stages of development are preferred. The inclusion of such antioxidants into the culture media has been used to reduce tissue browning with some success [17]. In other instances, antioxidants have been found to damage the tissues and cause imminent death. In general, we have managed the problem of browning, latex effluent, and oxidative stress by serial subculture of the explants at 48-h intervals for the first 6–8 weeks after culture initiation.

4.18.4.2 Stage 1: Establishment of Aseptic Culture and Confirmation of Bacterial Indexing

Skoog and Miller [20] originally hypothesized that plant growth and regeneration were dependent on the relative ratios of two principal phytohormones, auxins and cytokinins. However, even their original manuscript indicated the potential need for other mediating growth regulators or nutrients [20]. More recently, Murch and Saxena (2004) described several systems in which the induction of regeneration is not dependent on the auxin-to-cytokinin ratio but is influenced by a range of other growth factors. One such growth factor can be the synthetic agrochemical thidiazuron in some systems [14]. Interestingly, thidiazuron seems to be toxic to breadfruit explants, even in very low levels, as do several other auxin-to-cytokinin combinations. As such, the protocol for establishing breadfruit in tissue culture must be optimized individually for each cultivar. In addition, it may be the chemical nature of individual cytokinins rather than a standardized amount of the chemical class that determines the capacity of the inductive signal [12].

The development of buds into shoots from field-harvested tissues requires 3 months to 2 years of continuous aseptic subculture onto a variety of different media and is made even more difficult by the different growth regulator requirements for induction of regeneration of individual cultivars [19]. During this period, the surviving buds appear healthy and green but remain dormant until they receive the appropriate inductive stimulus. Unfortunately, the necessary composition of the medium varies among breadfruit cultivars and requires that a series of media be evaluated for each genotype. For example, Artocarpus shooting medium (AS) comprised of 4.33 g l\(^{-1}\) Murashige and Skoog (MS) salts [11], 1 ml l\(^{-1}\) B5 vitamins [4], 30 g l\(^{-1}\) sucrose, 2.5 g l\(^{-1}\) gellan gum, 2 µM benzylaminopurine (BA), and 3 µM kinetin successfully induced shoot formation in the cultivars ‘Ma‘afala’, ‘Puou’, and ‘Puupuu’ [12], but the cultivars ‘Ulu fiti’ and ‘Puaa’ required the addition of 1 µM gibberellic acid to the medium (Artocarpus shooting with Giberellins; ASG) [9]. Due to the unique requirements of each cultivar, a single induction medium is not possible and a series of media need to be evaluated for the establishment of new cultivars.

4.18.4.3 Stage 2: Clonal Mass Propagation

Once the initial bud dormancy is overcome in stage 1, breadfruit grows readily and vigorously in vitro. To date, attempts to induce indirect regeneration of shoots via a callus phase have been unsuccessful, but plantlets can be rapidly propagated by direct regeneration using node cuttings. At this stage, the different media requirements exhibited among cultivars are much less pronounced and an ASG growth medium has proved to be adequate for all cultivars tested. Plantlets have been multiplied on solid medium in magenta boxes using ASG medium, as well as in liquid lab bioreactors (Figure 1) using liquid ASG medium with the gellan gum omitted. While both solid and liquid culture methods enable the mass propagation of breadfruit, liquid culture vessels can accommodate a larger number of plants and allow for the media composition to be changed without the need to subculture the plantlets. These factors may make liquid culture systems more practical for larger-scale propagation. It is estimated that starting with only four or five buds, over 100 000 breadfruit plants could be generated within 1 year.

https://search.credoreference.com/content/topic/breadfruit
using this method [12]. As such, the micropropagation of millions of plantlets is feasible and large-scale distribution will be a reality in the near future.

4.18.4.4 Stage 3: Preparation for Rooting
Shoot explants of 2 cm, with at least two nodes, were found to be the ideal size for root induction. Smaller shoots with only a single node often fail to develop, and, when they do, they are less vigorous. Initial studies comparing various types and concentrations of auxins found that media comprised of 4.33 g l\(^{-1}\) MS salts, 1 ml l\(^{-1}\) B5 vitamins, 30 g l\(^{-1}\) sucrose, 2.5 g l\(^{-1}\) gellan gum, and 3 µM indole acetic acid (IAA) was the most effective rooting media [13]. Using this rooting medium (AR), 63% of the shoots developed roots within 3 weeks of culture compared to only 20% of shoots grown on similar media without IAA. More consistent and prolific rooting has recently been achieved by dipping microcuttings into commercial IBA rooting powder (#3) and before subculturing them into basal medium lacking plant growth regulators supplemented with 1 g ml\(^{-1}\) activated charcoal. Using this method, rooting rates of about 80–100% have been achieved. Rooting efficiency in the liquid lab bioreactors is generally much more efficient than it is on solid media, and the roots are much more vigorous [12]. Using the liquid system, an average of 60% of shoots rooted without the addition of auxin, and 100% rooted with the addition of 5 µM IAA. As such, efficient root induction methods have been developed for both solid and liquid propagation systems.

4.18.4.5 Stage 4: Microplant Establishment
Rooted plantlets have been successfully acclimatized from the liquid lab culture system to in vivo conditions with a 100% success rate by separating the plants, rinsing the roots of culture media, and planting them in soilless potting mix (Pro-mix BX, Premier Horticulture Ltd., Quebec, Canada) in a humidity-controlled growth chamber programmed to maintain 95% room temperature (RH), with weekly decreases of 5% RH until 70% was achieved [12]. However, highly efficient acclimatization of plants from both solid and liquid systems can be achieved using much more rudimentary methods. Rooted plants can be removed from the growth media, rinsed under tap water, planted in soilless potting mix (Pro-mix BX, Premier Horticulture Ltd., Quebec, Canada), covered with a clear plastic dome to maintain high humidity, and placed in a greenhouse. As the plants begin to grow, the plastic cover can be gradually opened and eventually removed. Although this simple method provides less control than a humidity-controlled growth chamber, survival rates are typically 90% or greater and it requires little specialized equipment. Provided care is taken to ensure that the breadfruit plantlets are kept warm and moist, and the humidity is decreased slowly, they readily acclimatize to greenhouse and outdoor conditions and rapidly grow into healthy trees.

4.18.5 Conclusions
Breadfruit is an ancient crop that has played a major role in Oceania for thousands of years. One of the major limitations to modern large-scale distribution of the plants outside of Oceania has been the slow, inefficient methods of propagation in conjunction with the persistence of microbial contamination and the associated agricultural import restrictions that limit international distribution. The recent developments described in this article show how biotechnology has been applied to overcome these difficulties enabling rapid, large-scale propagation of disease-free breadfruit trees. This technique has recently made large numbers of trees commercially available for charitable organizations and private customers through Global Breadfruit (www.globalbreadfruit.com). Widespread availability of planting material using modern biotechnology will help realize the full potential of breadfruit to increase food
security and fight hunger in the tropics.

**Relevant Websites**

http://www.agroforestry.net - Agroforestry.net; Species Profiles for Pacific Island Agroforestry

http://www.globalbreadfruit.com - Global Breadfruit: Food security for a growing world

http://www.croptrust.org - Global Crop Diversity Trust

http://www.planttreaty.org - The International Treaty on Plant Genetic Resources for Food and Agriculture

http://www.breadfruit.org

**Glossary**

**de-differentiation**
The process in which a specialized cell type reverts back into an unspecialized cell.

**differentiation**
The process in which an unspecialized cell develops into a specific cell type.

**direct regeneration**
The regeneration of a plant, or plant organ, directly from differentiated cells with no intermediate callous phase.

**indirect regeneration**
The regeneration of a plant, or plant organ, from tissue through an intermediate callous phase.

**micropropagation**
The production of large numbers of plants from a small amount of starting material using tissue-culture techniques.

**plant growth regulator**
A substance that directs the growth and/or development of a plant cell at low concentrations.

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