Chapter 2

Literature Reviews

1. Tangerine

The genus *Citrus* is native to Southeast Asia, occurring from northern India to China and south through Malaysia, the East Indies and the Philippines. There are about 16 species and they are all small evergreen trees and shrubs, usually with spines on trunks and branches. The ecology of wild species is now hard to establish because of drastic habitat modifications in the region as well as extensive hybridization between wild and domesticated plants. The history of domestication has also been hard to establish because archaeological evidence is lacking and it has been difficult to link names and descriptions in ancient accounts with the actual species we know today. Records of domestication go back to about 500 BC. (Reed, 1942; Wacer, 1943; Wikipedia, 2007a and 2007b)

The Mandarin was probably domesticated in tropical Southeast Asia. By 500 BC it was known in China and by 300 BC it was being grown commercially in central China. By 400 AD, grafting methods were being used to clone favourable varieties. They were also using tree-nesting Taylor Ants (*Oecophylla smaragdina*) to defend trees. It appears that the Mandarin has and still is the most important citrus species in China, both commercially and in people's gardens. It was introduced to Japan at an early stage, where it also became the most favoured of the citrus species, and it was here that the Satsuma variety was developed. Despite its popularity in Asia, it was only in the 1800's that *Citrus reticulata* was established in Europe, North Africa, West Indies, North and South America, and Australia. (Sauer, 1993)

Tangerine was cultured local in South of Thailand. Fruits should like but large sized of tangerine. Mature fruit in North of Thailand was peel yellow way mixes red but in South peel yellow way. The peel was thin, peel easy, peel off, fragile and fragrant. Carpel membrane was tight, soft and orange colored. Juice sac was dark orange and a lot of water. Many people like tangerine because sourwish sweet of taste. Quality of tangerine was high of titrate acid and total solution solids. (Ramingwong, 2003; Department of Agriculture, 2006b)

1.1 Fruit structure

The fruit of citrus is a special type of berry termed “hesperidium”. It is a true fruit arising through growth and development of the ovary, consisting of a variable number of united, radically arranged carpels. Phylogenetically, carpels are considered by most authors to be modified leaves oriented vertically with their margins curved to join the central axis, thereby forming locules (segments) in which seeds and juice sacs develop (Ting and Attaway, 1971; Nagy *et al*., 1977; Spiegel-Roy and Goldsehmidt, 1996; Lack and Evans, 2005).

A small, secondary fruit (navel) is sometimes present at the stylar end of the main fruit. Whereas in certain types of mandarins (tangerine) the navel appears as a tiny, undeveloped fruit, in Washington navel orange the secondary fruit attains a diameter of 2-3 cm, slightly protruding, although still enclosed by the peel of the main fruit. The ontogeny of the navel has been described in detail by Lima and Davies (1984).

Citrus fruits are composed of two major, morphologically distinct regions- the pericarp, commonly known as the 'peel' or 'rind' and the endocarp, which is the edible portion of fruit, often called the 'pulp'. A further distinction is made within the peel; the external, colored portion is the epicarp, mostly known as the flavedo, whereas the internal, white layer of the peel is the mesocarp, generally known as the albedo. The 'flavedo' is composed of the cuticle-covered epidermis and a few compactly arranged parenchyma cell layers adjacent to it. Embedded in the flavedo are multicellular, schyzolysogenic oil glands containing essential oils. During early stages of fruit development the flavedo is a dark green, photosynthetically active tissue, with a relatively small number of stomata (20 to 40). As the fruit approaches maturation, chlorophyll is gradually lost and chloroplasts are transformed into carotenoid rich chromoplasts (Goldschmidt, 1988). The deeper layers of the flavedo merge into the white, spongy 'albedo'. In the mature fruit the albedo consists of large, deeply lobed cells with very large intercellular spaces and scattered vascular elements. During the early phase of fruit development, when peel growth predominates, the albedo may occupy 60 to 90 % of fruit volume. Later, when pulp growth takes over, the albedo becomes thinner and the portion of the albedo declines. In numerous mandarin and orange cultivars the albedo gradually degenerates and disappears, leaving only a net of vascular elements between the flavedo and the pulp - this is the 'reticulum' for which mandarins have been named *C. reticulata*. Physiological diseases such as 'creasing' and 'puffing' are evidently related to lysis and disintegration of the albedo, but the underlying biochemical processes are still poorly understood. Renewed growth of the albedo and thickening of the peel occur in certain cultivars during fruit maturation and senescence (Goldschmidt, 1988).

The pulp, which is the edible portion of the fruit, consists of segments, the ovarian locules, enclosed in a locular membrane and filled with juice sacs (sometimes called juice vesicles). Development of the juice sacs has been followed in detail by Schneider (1968). Juice sacs are initiated at about bloom, appearing at first as dome shaped protrusions from the locular membrane into the locules. Development of the domes into juice sacs occurs through apical meristem activity and subsequently by an obese mass of meristematic tissue giving rise to the body of the sac. In the mature fruit, juice sacs appear as elongated, mostly spindle-shaped multicellular structures, projecting from a stalk implanted in the periphery of the segment toward the central axis, where the seeds are found.

Juice sacs are the ultimate 'sink' organ of citrus and their development presents intriguing physiological problems. The fact that the juice sacs are not connected to the vascular system which provides water and assimilates to the fruit has long been recognized by students of fruit morphology (Schneider, 1968). Recent studies by Koch and coworkers (Koch *et al*., 1986; Koch and Avigne, 1990) have confirmed and extended these observations. Neither juice sacs nor their stalks nor the segment epidermis to which the stalks are attached show any differentiation of transfer organs. Thus, juice sacs must obtain their supply over long distances (up to 3 cm) of post phloem, through nonvascular cell-to-cell transport. Labeling experiments indicate that the entry of water and assimilates into juice sacs is extremely slow. It is still not clear whether the cell-to-cell transfer of assimilates takes place via symplastic (i.e. plasmodesmata) or apoplectic (i.e. cell wall) routes. The biophysical mechanisms of water and solute uptake by the growing fruit as well as the biochemical reactions involved need further elucidation.

1.2 Fruit development

In a classical study, Bain (1958), Charles and Coggins (1986), Monselise (1986) and Kale and Adsule (1995) divided the development of Valencia orange fruit into three major stages: cell division (I), cell enlargement (II) and fruit maturation (III). Bain's division seems appropriate for most citrus fruits, although time and duration of developmental stages may vary, according to cultivar, climate, etc.. Tangerine are growing simple sigmoid curve. (Krezdorm, 1986)

The cell-division period (stage I) may be assumed to commence at fruit set, immediately following anthesis. However, differences in fruit size between leafy and leafless inflorescences are already evident in the ovaries prior to anthesis (Guardiola and Lazaro, 1987; Hofman, 1988). The size of the ovary also varies as a function of the number of flowers per tree (Guardiola *et al*., 1984). These differences in size probably reflect differences in cell division during floral development, indicating that fruit development actually begins before anthesis, a view expressed long ago by Nitsch (1953).

Cell division appears to terminate in all fruit tissues, except the outermost flavedo layers and the tips of juice sacs, within five to ten weeks after bloom. The increase in fruit size during stage I is mainly due to growth of the peel, consisting of cell division, but there is already a component of cell enlargement. The peel reaches its maximum width soon after the end of stages I; this has been shown repeatedly for oranges (Bain, 1958; Goren and Monselise, 1964; Holtzhausen, 1982) and mandarins (Kuraoka and Kikuki, 1961; Kale and Absule, 1995). Peel volume increases nevertheless somewhat further during stage II.

Stage II, the cell-enlargement phase, may also be envisaged as the pulp growth stage. Juice sacs enlarge and fill the locules (segments) quite early, with their juice and sugar content increasing mainly towards the end of this stage. The rapidly expanding pulp exerts considerable pressure outwards on the peel, which stretches, getting increasingly thinner. The shape of the oil glands also changes as the season progresses, due to stretching of the peel (Holtzhausen, 1982; Kale and Absule, 1995). Fruit splitting, which is quite widespread among thin-peeled mandarin and orange cultivars, is believed to result from excessive pressure of the developing pulp on the thin, over-stretched peel (Goldschmidt *et al*., 1994).

Stage III is known as the fruit maturation and ripening phase (Bain, 1958). In fact, fruit growth continues during this stage as well, the growth rate depending to a large extent on climatic conditions. As mentioned, renewed growth and thickening of the peel may be observed during stage III, particularly under warm, growth-favoring conditions (Kuraoka, 1962; Herzog and Monselise, 1968; Reuther, 1973). Pulp growth, on the other hand, almost stops at this stage in certain cultivars, leading to formation of cracks between the peel and the pulp. When, combined with lysis and degradation of the albedo these cracks develop into large, hollow air spaces, a condition known as 'puffing' (Kuraoka, 1962).

In subtropical climates, chlorophyll pigments disappear from the flavedo and carotenoid pigments are revealed. In many cultivars, carotenoids increase substantially and/or are converted into highly colored pigments during and after the loss of chlorophyll from the flavedo. Depending on the type of citrus fruit being considered, regreening occurs during stage III (Charles, 1986).

Sugars and water continue to accumulate in juice sacs and the concentration of organic acids decreases (Charles, 1986; Kale and Adsule, 1995).

Studies of citrus fruit development have generally focused on a specific cultivar. However, when the whole spectrum of citrus fruits is considered attention has to be paid to the diversity of peel/pulp ratios in mature fruit. On one extreme are certain types of citron (*Citrus medica*), which lack a fleshy pulp altogether. The segments are thick-walled, empty locules containing only the seed. All the rest of the fruit cross-sectional area is occupied by the albedo and by the massive central axis. At the other end are thin-peeled mandarins. Here, the pulp predominates while the albedo had degenerated and almost disappeared. Between these extremes are all other kinds of citrus-pummeloes, grapefruits and oranges represent, in a decreasing order, different peel/pulp ratios. This is undoubtedly a genetically controlled trait which might, to some extent, be mediated by plant hormones.

1.3 Maturation, ripening and senescence of citrus fruit

Ripening of citrus fruit is quite different from that of most other fruits. Ripening of fruits like apple, avocado, tomato and banana involves rather abrupt changes in fruit texture and composition. In citrus fruit such changes are rather limited and take place in a slow and gradual manner. Citrus fruit approaching maturation does not contain starch and must, therefore, achieve internal maturity on the tree, prior to harvest. The biochemical changes occurring in avocado, tomato, etc. appear to be intimately related to the climacteric rise in respiration and ethylene evolution (Theologis *et al*., 1992). Citrus fruits, on the other hand, are 'non-climacteric'. Respiration declines slowly throughout the later stages of fruit development and ethylene evolution of the mature fruit is extremely low (Aharoni, 1968; Eaks, 1970; Goldschmidt *et al*., 1993). Use of the term 'ripening' with regard to citrus is often refuted on these grounds and the term 'maturation' is preferred.

Structural and physiological differences between peel and pulp of citrus fruit have already been pointed out in the foregoing discussion of fruit development. During maturation peel and pulp behave in most respects as separate organs, although some coordination does exist. Describe some of the major changes occurring in peel and pulp of Shamouti orange during maturation. The changes which take place during maturation of the peel are comparable to the senescence of other chlorophyllous tissues, as revealed mainly in transformation of the chloroplasts into chromoplasts (Goldschmidt, 1988). The decline in rind chlorophyll takes several months and the onset of carotenoid accumulation almost coincides with the disappearance of chlorophyll. Just prior to their build-up the carotenoids go through a 'trough' which marks the transition from carotenoids of the photosynthetic chloroplast to the intensely colored carotenoids of the chromoplast (Eilati *et al*., 1969; Gross, 1987).

Maturation of the pulp is characterized by gradual changes in juice content and in some of its constituents. On one hand, there is a decline in titratable acidity (TA) brought about by decomposition of citric acid, which is the principal organic acid of citrus juice (Monselise, 1986). On the other hand, there is an increase in sugars, usually expressed as total soluble solids (TSS). With acidity declining and sugars increasing towards maturation, the TSS/TA ratio is extremely sensitive and is commonly used, therefore, as a 'maturity index'. It’s indicates a TSS/TA ratio of 8 which has been reached, in this case, at about the same time that chlorophyll disappeared. This is not the rule, and peel coloration of different cultivars may often precede or lag behind internal maturity. Degreening with ethylene is practiced in the latter case, mainly with early cultivars (Grierson *et al*., 1986).

The increase in the percentage of expressible juice (juice content) is at least partly due to release of water within the pulp tissue, also occurring postharvest. Maturity standards are legally enforced in several different countries, varying for different species and cultivars. For oranges, grapefruits and mandarins they usually include an accepted range of TSS/TA ratios, minimum juice content and an acceptable peel color. Juice content is the only maturity parameter used for lemons and limes, which are commonly used as a non-sweetened acid ingredient (Soule and Grierson, 1986).

Fruit softening, which is a dominant feature of ripening in most climacteric fruits, does not play a significant role in the maturation of citrus fruits. Although both peel and pulp of citrus fruit are rich in polyuronides (pectins), their decomposition into smaller, soluble subunits is very slow, with the exception of certain mandarins, which undergo a more pronounced softening. The postharvest reduction in fruit firmness is largely due to loss of water, mainly from the pulp, leading to shrivelling and deterioration of fruit appearance and quality. Real 'softening' is brought about by several pathogenic postharvest diseases involving massive secretion of cell wall degrading enzymes (Eckert and Eaks, 1989).

Various systems interact in the regulation of fruit growth and numerous factors turn out to be limiting during the course of fruit development. An ample supply of water is a prerequisite for all stages of fruit development. Water stress can be particularly dangerous during fruit set, leading to a massive drop of fruitlets (Monselise, 1986). Increase in size and juice content are also largely dependent upon the availability of water (Marsh, 1973). Mineral nutrition, with its complex elemental interactions, is important as well. Potassium seems to play a special role in fruit development - potassium deficiency reduces fruit size (Barnette *et al*., 1931; Chapman, 1968; Embleton *et al*., 1973; Brown *et al*., 1987) and potassium sprays are used to strengthen the peel (Embleton *et al*., 1973) and increase fruit size. The supply of carbohydrates has also been suggested as a limiting factor for fruit set and enlargement. Girdling at full bloom increases fruit set (Monselise *et al*., 1972; Erner, 1988), presumably by provision of more photosynthate to the young fruitlets (Schaffer *et al*., 1985). Girdle during the fruit enlargement stage increases fruit size (Cohen, 1984; Fishler *et al*., 1983), probably by eliminating competition for photosynthate by the root system and other growing organs.

1.4 Fruit composition

The lists several classes of native constituents of citrus fruits, and also gives some estimates of their concentrations in peel and juice of orange and lemon. The significance of some of these compounds is related to citrus fruits' edible and nutritional qualities, while others are important by-products of the processing industry. The soluble sugar pool contains mainly glucose, fructose and sucrose. In orange, grapefruit and mandarin juice the amount of sucrose exceeds that of fructose and glucose (Braverman, 1949). Pulp sucrose levels increase markedly towards ripening, reaching in certain mandarins 15 to 18 % of fresh weight (Tzur, 1994). In the peel, the soluble sugar pool often contains more reducing sugars (glucose, fructose) than sucrose. In juices of lemon and lime the amounts of sucrose are minimal.

Ascorbic acid, better known nutritionally as vitamin C, is bio-chemically related to sugars although its precise biosynthetic route in citrus has not been elucidated. Citrus fruits have been known for a long time as a major dietary source of ascorbic acid. The early seventeenth century citrus fruits have been found to prevent the scurvy (scorbutus) disease. A teaspoonful of citrus juice was often included, therefore, in the daily ration of seamen (Sinclair, 1984). Citrus fruit peel has higher ascorbic acid content than the juice. Fruit exposed to sunlight have significantly higher ascorbic acid content, as well as higher total soluble solids content. The ascorbic acid content does not change much after harvest, even during several months' storage. Processing of fruit may involve considerable losses, unless special precautions are exercised (Eaks, 1964; Sinclair, 1984).

Pectins are major cell-wall components of fleshy fruits. Pectins are large, complex carbohydrate macromolecules, composed of partly methyliated polygalacturonic acid backbones and considerable amounts of other sugar residues. Various methods for extraction and separation of pectins from other cell-wall components have been developed. Extensive research has been conducted on the pectic materials of citrus fruits (Sinclair, 1984). Citrus pectins have a high galacturonic acid content. Citrus fruit, particularly the peel, serve as an important raw material for production of high-quality commercial pectin. Pectin is used in the food industry, mainly as a jellying agent.

Acidity is a major determinant of fruit taste and edibility. Organic acids play a central role in fruit metabolism (Ulrich, 1970). Malic acid in apple, tartaric acid in grape and citric acid in citrus, all show a distinct peak at the midst of the growth period, followed by a descent towards ripening (Monselise, 1986). The citric acid of citrus juice is probably produced by a side cycle, coexisting with the tricarboxylic acid cycle. Accumulation of citric acid has been proposed to be brought about by the high concentration of citramalate, which blocks aconitase activity (Wallace *et al*., 1977). Citrus fruits vary greatly in their acid content; even cultivars of the same species (e.g. orange, lime) show extreme differences. The genetics of the acidless trait has been studied in several Citrus species and found to be complex (Cameron and Soost, 1979). The biochemical background of these differences in acidity is as yet poorly understood (Wallace *et al*., 1977).

While citric acid is the main acid component of juice, with malic acid coming next, malic and malonic are the major acids in the flavedo and albedo (Sasson and Monselise, 1977). The total acid content of the peel is much lower, however, than that of the juice (5 %). Malonic acid appears to increase during rind senescence and off the tree (Monselise, 1986).

The flavonoids are an important class of plant secondary metabolites, belonging to the broader family of plant phenolics. The flavonone glucosides of citrus have been subject to numerous studies over the years (Kesterson and Hendrickson, 1953; Goren, 1965; Sinclair, 1984). Considerable progress has been achieved recently in their analysis (Castillo *et al*., 1992) and their biosynthetic pathway, including some of the key enzymes involved, has been elucidated (Hasegawa and Maier, 1982; Peled *et al*., 1993; Berhow *et al*., 2000). Hesperidin and naringin are known to be the major flavonone glucosides of orange and grapefruit, respectively. The overall distribution of flavonoids in Citrus species is more complex, however, and attempts have been made to use it as a chemotaxonomic tool (Tatum *et al*., 1978). Citrus plasmids accumulate in young, rapidly developing organs attaining up to 75 % of the dry weight of young fruits of approximately 1 cm diameter (Kesterson and Hendrickson, 1953). No specific role can be assigned so far to the flavonoids in the physiology of citrus. Citrus flavonoids have aroused much interest due to their organoleptic properties. Whereas hesperidin is tasteless, naringin is extremely bitter and as such responsible for the bitterness of grapefruit. Ncohesperidin dihydrochalcone is an intense artificial sweetener (Horowitz and Gentili, 1986) which can be produced from naringin or, even more easily, from neohesperidin, which is a natural flavanone glucoside of *Citrus aurantiura* (Castillo *et al*., 1992).

The carotenoids of citrus fruits have been extensively investigated, particularly by Gross and coworkers (Gross, 1987). The Citrus genus is remarkable for producing the largest number of carotenoids found in fruit. About 115 different pigments have been reported, including a large number of isomers, some of which might be formed during isolation.

Each species and hybrid has a characteristic carotenoid complex which is responsible for its typical color; peel (flavedo) and pulp pigments reveal certain differences. In yellow citrus fruits (pummelo, grapefruit, lemon, lime) the total amount of carotenoids is low and most of these belong to the colorless carotenoids. Thus, in white Marsh seedless grapefruit the colorless phytoene and phytofluene account for 74 % of the carotenoids of the flavedo. The accumulation of these colorless precursors is the result of a genetically determined metabolic block which hinders further dehydrogenation steps leading to the colored carotenoids. In pink and red grapefruit cultivars lycopene and β-carotene are the major carotenoids, particularly in the pulp, indicating that the genetic block had been removed. Lycopene is also responsible for the rapid formation of red color in citrus fruit treated with tertiary amine bioregulators (Yokoyama and Keithley, 1991).

Orange-colored citrus fruits (orange, sour orange, mandarin) contain relatively large amounts of a complex mixture of carotenoids. Some of these (e.g. cryptoxanthine, β-citraurin) appear in small amounts but have a high tinctorial value. The molecular regulation of this extreme biochemical diversity within the Citrus genus is still greatly unknown.

Blood oranges, on the other hand, owe their color to a different class of pigments the anthocyanins. Like most anthocyanin-containing fruits, blood oranges also develop the desired, intense coloration in cooler regions. This is presumably the reason for the success of blood oranges under the cool maritime climate of Sicily.

Essential oils are volatile, fatty plant constituents contained in the oil glands found in most citrus organs, particularly in the flavedo. The essential oils are chemically composed of terpenes, aldehydes, alcohols, acids and hydrocarbons and rarely exist as esters of the ordinary fatty acids. Lemon oil was found by Kesterson *et al*. (1971) to contain more than 100 chemical constituents, 68 of which have been definitely identified. The monoterpene, d-limonene, is the principal constituent (60-90 %) of fruit essential oils, whereas leaves have other monoterpenes as their major constituents. Analyses of leaf essential oils have been used for chemotaxonomy of Citrus species. Citrus essential oils are widely used for cosmetic and pharmaceutical purposes, as well as flavoring agents in the food and beverage industry. Records of commercial lemon oil production in Italy date back more than four centuries. The Italian lemon oil, produced by the old, hand-pressing method has been renowned for its high quality. In Sicily and other lemon-growing areas of southern Italy, a considerable portion of the crop, at times even most of it, was diverted to the essential oil industry. There are two types of commercial essential oils: cold pressed oil and distilled oil. The production method affects the yield and chemical composition of the oil. Commercial yields of lemon oil range between 0.54 and 0.78 % of fruit weight (Sinclair, 1984).

About 30 limonoid triterpene derivatives have been identified in Citrus species. Limonin is the major member of this group. Limonin is the principle bitter compound of citrus juices and as such has been subject to extensive research (Maier *et al*., 1980; Hasegawa and Maier, 1982; Berhow *et al*., 2000). In intact fruit, limonin is present in the monolactone form, which is the nonbitter precursor oflimonin (Maier and Beverly, 1968). This explains the delayed development of bitterness in citrus juices. Considerable efforts have been put into the development of enzymatic debittering techniques. Similar approaches have been undertaken also with regard to the naringin bitterness of grapefruit juice (Hasegawa and Maier, 1982).

2. Plant nutrients

A plant nutrient is chemical elements that are essential for plant growth and reproduction. Essential element is a term often used to identify a plant nutrient. The term nutrient implies essentiality, so it is redundant to call these element essential nutrients. Commonly, for an element to be a nutrient, it must fit certain criteria. The principal criterion is that the element must be required for a plant to complete its life cycle. The second criterion is that no other element substitutes fully for the element being considered as a nutrient. The third criterion is that all plants require the element. All the elements that have been identified as plant nutrients, however, do not fully meet these criteria, so, some debate occurs regarding the standards for classifying an element as a plant nutrient (Buasap, 2001; Barker and Pillbeam, 2007; Zekri and Obreza, 2008; Zekri *et al*., 2008).

For an element to be essential, it must be required for a plant to complete its life cycle, it must be required by all plants, and no other nutrient can replace this requirement fully. If an element does not meet all of these requirements, for example, being required by some plants or only enhancing the growth of plants, the element may be a beneficial element. Much interest in plant nutrition lies in the development and use of diagnostic techniques for assessment of the status of plants with respect to plant nutrients and beneficial elements (Buasap, 2001; Barker and Pillbeam, 2007; Zekri and Obreza, 2008; Zekri *et al*., 2008).

Seventeen elements are considered to have met the criteria for designation as plant nutrients. Carbon, hydrogen, and oxygen are derived from air or water. The other 14 are obtained from soil or nutrient solutions. It is difficult to assign a precise date or a specific researcher to the discovery of the essentiality of an element. For all the nutrients, their roles in agriculture were the subjects of careful investigations long before the elements were accepted as nutrients (Buasap, 2001; Barker and Pillbeam, 2007). Many individuals contributed to the discovery of the essentiality of elements in plant nutrition. Much of the early research focused on the beneficial effects or sometimes on the toxic effects of the elements. Generally, an element was accepted as a plant nutrient after the body of evidence suggested that the element was essential for plant growth and reproduction, leading to the assignment of certain times and individuals to the discovery of its essentiality (Zekri and Obreza, 2008; Zekri *et al*., 2008).

3. Nutrient balance

Continuous cropping without adequate restorative practices may endanger the sustainability of agriculture. Nutrient depletion is a major form of soil degradation. A quantitative knowledge on the depletion of plant nutrients from soils helps to understand the state of soil degradation and may be helpful in devising nutrient management strategies. Nutrient-balance exercises may serve as instruments to provide indicators for the sustainability of agricultural systems. Nutrient-budget and nutrient-balance approaches have been applied widely in recent years. Studies have been undertaken at a variety of levels: plot, farm, regional, national and continental. Widespread occurrence of nutrient mining and soil fertility decline has been reported (Roy *et al*., 2003).

Most nutrient-balance studies provide rapid findings, based on a short time-frame exercise, and necessarily depend on a number of assumptions relating to system dynamics. However, questions remain concerning the validity of such assumptions, their reliability, and their capability to provide insight into dynamic processes and lend support for extrapolation. Also pertinent is the issue as to which new approaches/directions, investigations and extra efforts are required and feasible in order to enhance the validity of the assumptions and findings. Questions have been raised as to whether nutrient budgets provide the information required for understanding the status and dynamics of soil fertility across farming systems and whether such analysis may provide reliable direction and support to policy formulation on soil fertility management (Scoones and Toulmin, 1998).

Nutrient-balance assessments are valuable tools for delineating the consequences of farming on soil fertility. Various approaches and methods for different situations have been used. This bulletin presents a state-of-the-art overview of nutrient-balance studies. It brings out the evolution of the approaches and methods, provides for comparisons among them, features the improvements made and highlights remaining issues. This analysis will be useful in further development of the assessment methodologies as reliable tools for devising time-scale soil fertility management interventions (Roy *et al*., 2003).

As nutrients are removed from a piece of land through the harvested product, it is important to assess if these nutrients are adequately replenished through internal processes and/or external inputs, such as fertilizers. Internal processes that make nutrients available to plants include the mineralization of soil organic matter, releases from the soil matrix, and the biological fixation of atmospheric nitrogen. At the same time, nutrients can be lost from the system through processes such as leaching, lateral flow, soil erosion and identification. It is not easy to measure many of these processes rapidly. Simplified nutrient balances are, therefore, often used as a first indication of the nutrient dynamics of a system. The working group limited its calculations to the three major plant macronutrients: nitrogen (N), phosphorus (P) and potassium (K) (Vosti *et al*., 2007; IPNI, 2007).

4. Citrus nutrition

Citrus trees are demanding feeders and are prone to many disorders related to mineral nutrition. Citrus species are also sensitive to an excess of certain elements in the soil or the irrigation water, especially to an excess of chloride, sodium, boron and manganese, which can injure the trees (Weir and Sarooshi, 2002; Chapman, 1966 and 1968). Mineral nutrients arc required for various physiological processes and structural components (Reuther *et al*., 1968; Tisdale and Nelson, 1975).

4.1 Nitrogen (N)

Nitrogen concentrations in citrus tissues are highest in the leaves and are next highest in immature fruits. Leaves contain enzymes for the PCR cycle, carbohydrate metabolism and nitrate reduction. Nitrogen is a structural component of chlorophyll and an important constituent of proteins. It is essential for cell division and expansion. Acute lack or shortage of this element arrests vegetative growth and results in bronzing or yellowing of foliage. New leaves of trees deficient in N are thin and fragile, and the angle between stem and leaf is rather narrow. Citrus orchards deficient in N may exhibit a decrease in flowering and fruiting even before striking leaf symptoms develop. This is largely due to a reduction in growth. Over-fertilization with N may cause excess growth, reduction of yield and decrease in fruit quality (Reuther *et al*., 1968; Tisdale and Nelson, 1975; Kool *et al*., 1984; Mooney *et al*., 1993; Obreza *et al*., 1999; Maurer and Truman, 2000; Buasap, 2001; Futch and Tucker, 2008).

Nitrogen is the element that has the greatest effect on citrus production, and citrus needs more nitrogen than any other nutrient (Weir and Sarooshi, 2002).

Nitrogen is a component of chlorophyll (the green pigment in leaves), and is associated with important tree functions such as growth, leaf production, flower initiation, fruit set, and fruit development and quality (Weir and Sarooshi, 2002).

A nitrogen shortage causes the loss of green colour from the leaves, resulting in an even paleness. A deficiency of nitrogen in the spring makes the leaves pale and small. Old leaves shed early in the season, causing the foliage cover to become thin, and twigs to die back. Tree growth is retarded, and cropping suffers through poor fruit set and smaller fruit (Weir and Sarooshi, 2002).

Pauline *et al*. (1993) were summary research that nitrogen must be maintained in balance with other elements in a citrus tree. Leaf analysis is critical for getting the balance right. Young trees need small; split applications of N to promote each growth flush and develop strong canopies. Bearing trees (5 years and older) need autumn applications only, to build up reserves for spring and promote fruit quality in the next season.

4.2 Phosphorus (P)

Phosphorus is a highly mobile element within the tree. Phosphorus is a component of nucleoproteins and phospholipids, and is involved in energy transfer. A deficiency in phosphorus results in excessive abscission of old leaves, bronze, lusterless leaves and sparse bloom. Some typical fruit disorders have also been described (Chapman, 1966 and 1968). Excess of P has been found to accentuate zinc deficiency

Phosphorus performs many vital functions in the plant in photosynthesis, in enzyme activity, and in the formation and movement of sugars. It is important in flowers and developing fruit (Weir and Sarooshi, 2002).

It is rare for tree growth or yield to be affected by low soil phosphorus in the orchard, or for deficiency symptoms to be seen in leaves. Where such extreme deficiency does occur, leaves are a dull bronzed green and they shed readily. Low phosphorus affects fruit quality, causing misshapen fruit with open centers and coarse, thickened rinds. The fruit is pulpy and has a low juice percentage, and the juice is acidic. The quantity of total soluble solids (sugar content) of the juice is usually not affected. The effect of phosphorus deficiency on fruit quality is worse when too much nitrogen fertilizer has been used. A balanced supply of nitrogen and phosphorus gives both a high yield and good fruit quality (Weir and Sarooshi, 2002).

4.3 Potassium (K)

Large amounts of potassium are required by citrus. Potassium is an important constituent of the fruit (40 % of the total mineral content). It is involved in the translocation of carbohydrates. It acts as an osmotic agent in the opening and closing of stomata. It plays an important role in controlling the acidity of the fruit juice. It functions in charge balancing and membrane transport. Reduction in yields due to K deficiency is attributed mainly to reduced vegetative growth. However, the effect of low K on decreasing average fruit size is striking (Tisdale and Nelson, 1975; Kool *et al*., 1984; Barber and Peterson, 1995; Obreza *et al*., 1999; Maurer and Truman, 2000; Buasap, 2001; Barker and Pillbeam, 2007; Zekri and Obreza, 2008).

Potassium is important in the formation and functioning of proteins, fats, carbohydrates and chlorophyll, and in maintaining the balance of salts and water in plant cells (Barber and Peterson, 1995; Obreza *et al*., 1999; Maurer and Truman, 2000; Zekri and Obreza, 2008).

Potassium deficiency symptoms vary in citrus. They are often not easy to recognize, and can be mistaken for other problems. Symptoms include slower tree growth, small leaves and a heavy leaf fall, often preceded by the leaves turning yellow or bronze. Dieback follows in the weakened twigs, and bloom decreases. In late summer the pattern of leaf yellowing is irregular and blotchy, with most yellowing starting near the apex half of the leaf. The yellowing later becomes bronzed and more irregular in pattern. Fruit are small; the skin is thin and smooth, tends to colour early, and splits easily. Creasing is more prevalent (Weir and Sarooshi, 2002).

Mild deficiency does not affect the yield, although the fruit may be smaller. Severe potassium deficiency reduces the yield by causing heavy flower and fruit drop (Zekri and Obreza, 2008).

4.4 Calcium (Ca)

Symptoms of calcium deficiency are rarely seen in citrus orchards, but magnesium deficiency occurs in most districts in New South Wales. Magnesium deficiency is common in the leached acid sandy soils of the Central Coast, but it can also be found inland (Weir and Sarooshi, 2002).

4.5 Magnesium (Mg)

Magnesium is part of the chlorophyll molecule, and an activator of photosynthesis and respiration. Deficiency causes a characteristic chlorosis of the foliage. It is a highly mobile element; deficiencies start on basal caves. They are especially noted in seeded citrus cultivars, because of translocation of Mg to the seeds. Magnesium is involved in the activation of several enzymes and maintenance of ribosome (Reuther *et al*., 1968; Tisdale and Nelson, 1975; Kool *et al*., 1984; Mooney *et al*., 1993; Barber and Peterson, 1995; Obreza *et al*., 1999; Maurer and Truman, 2000; Buasap, 2001; Barker and Pillbeam, 2007; Futch and Tucker, 2008; Zekri and Obreza, 2008).

4.6 Iron (Fe)

The plant needs iron to produce chlorophyll. Iron deficiency causes a distinct pattern in the leaf caused by a loss of chlorophyll; only the main veins stay green. Iron does not move easily within the plant, so young leaves are the worst affected, showing the symptoms, while older leaves may remain green (Weir and Sarooshi, 2002).

Iron deficiency is sometimes called ‘lime-induced chlorosis, as it is worse in calcareous (calcium rich) soils with a high pH. This makes iron unavailable. The problem is accentuated where free lime concretions are found near the surface of the soil. A high water table (which causes a low soil-oxygen supply to the roots) or a low soil temperature can further aggravate the problem (Weir and Sarooshi, 2002).

Foliar deficiency symptoms of both manganese (Mn) and iron (Fe) are chlorosis of leaves. Manganese deficiency symptoms occur both on young and old leaves. Iron deficiency starts on apical leaves. Iron deficiency starts on apical leaves. Chlorosis in Mn deficiency resembles Zn deficiency, but is usually less extreme. Severe Fe deficiency shows incomplete yellowing of leaves. Mn also activates several enzyme systems and is required in respiration and photosynthesis. Iron is involved in chlorophyll synthesis and is part of certain enzyme systems. It is involved in the reduction-oxidation process in photosynthesis and respiration (Reuther *et al*., 1968; Tisdale and Nelson, 1975; Kool *et al*., 1984; Mooney *et al*., 1993; Barber and Peterson, 1995; Obreza *et al*., 1999; Maurer and Truman, 2000; Buasap, 2001; Barker and Pillbeam, 2007; Futch and Tucker, 2008; Zekri and Obreza, 2008).

4.7 Manganese (Mn)

Manganese deficiency causes a diffuse pale green mottle between the veins in young and old leaves. The leaf size is normal. A narrow band remains green on each side of major veins. Symptoms are more noticeable on the southern side of the tree. Spring growth is affected, and both young and mature leaves can show the symptoms. Persistent severe deficiency reduces cropping and growth. Liming can accentuate the deficiency (Weir and Sarooshi, 2002).

4.8 Copper (Cu)

Copper deficiency symptoms are large, dark green leaves, gum pockets in woody tissue and between wood and bark, dieback of terminal shoots and multiple buds. Copper is part of the oxidation reduction systems, such as ascorbic acid oxidase, polyphenol and laccase oxidases. It is part of plastocyanin (chloroplast enzyme) (Obreza *et al*., 1999; Barker and Pillbeam, 2007; Futch and Tucker, 2008; Zekri and Obreza, 2008).

Copper deficiency (exanthema) was a common problem in coastal areas the Hills District around Sydney, the Hawkesbury Valley and Gosford. It is now rarely seen on the coast since the use of copper fungicidal sprays (Weir and Sarooshi, 2002).

Copper deficiency is occasionally found in the lower Murray and the MIA, where it causes a variety of symptoms. The leaves are often dark green and smaller than normal, except for a few giant leaves found on vigorous S-shaped branches which emerge from stunted bushy trees. Twig growth is weak and likely to die back. The most characteristic symptom is dark brown gum-pockets which develop on the flat sides of angular young shoots. They have a blister-like appearance. The rind of deficient fruit develops brown gum-stained areas, and the fruit splits (Weir and Sarooshi, 2002).

4.9 Zinc (Zn)

Zinc is a micronutrient. Next to nitrogen, zinc deficiency is perhaps the most widespread nutritional disorder in citrus, occurring under a wide range of soil conditions and environments. Deficiency symptoms are easily identified by characteristically mottled leaves, highly reduced leaf size, and often dieback of twigs and small misshapen fruit. Zinc is essential for the functioning of many enzymes, as well as the synthesis of tryptophan, a precursor of indoleacetic acid (IAA). Zinc deficiency causes a reduction in RNA synthesis and ribosome stability (Reuther *et al*., 1968; Tisdale and Nelson, 1975; Kool *et al*., 1984; Obreza *et al*., 1999; Maurer and Truman, 2000; Buasap, 2001; Barker and Pillbeam, 2007; Zekri and Obreza, 2008).

Zinc deficiency, described as ‘little leaf’, ‘mottle leaf’ and ‘resetting’, is one of the most damaging and widespread nutritional disorders of citrus. Leaf analyses conducted by NSW Agriculture have shown that over 60% of citrus orchards are low in zinc. The deficiency is most acute in alkaline soils. It also affects citrus growing on acid coastal soils (Weir and Sarooshi, 2002).

Even in its earliest stages, zinc deficiency lowers yield, reduces tree vigor and makes fruit small and poor in quality. Leaf symptoms include small, narrow leaves (little leaf) and whitish-yellow areas between the veins (mottle leaf). Leaves also crowd along short stems (resetting), and smaller twigs die back. Symptoms are often more pronounced on the northern (sunny) side of the tree (Weir and Sarooshi, 2002).

4.10 Boron (B)

While boron deficiency has been a problem in some citrus orchards, excess B in the plant is even more common, especially in some irrigated regions and in soil areas high in boron. Boron deficiency is manifested by abnormal abortion of young fruits, peel discoloration of fruit and dieback of growth. Symptoms are, however, not always highly specific. The concentration range of B in plants between deficiency, normal concentration and toxicity is rather narrow. Brown pustules on dark green leaves are symptoms of toxicity. Boron, not readily translocated, appears to be required for sugar translocation. It is involved in pollen tube elongation and in cell division in root and shoots apices (Obreza *et al*., 1999; Maurer and Truman, 2000; Buasap, 2001; Barker and Pillbeam, 2007; Futch and Tucker, 2008; Zekri and Obreza, 2008).

Boron deficiency is uncommon in New South Wales citrus. The fruit develops a grey to brown discoloration, with pockets of gum in the white part of the rind. The gum is also found in the flesh, and where the flesh meets the rind. Some seeds are imperfectly developed, shriveled, and brown in colour and embedded in gum (Weir and Sarooshi, 2002).

Fruit which have been injured by spied citrus bug are often wrongly diagnosed as ‘boron-deficient’. Spied citrus bug damage is characterized by discrete dry fruit segments, whereas boron deficiency symptoms are not confined to one or two segments of the fruit. Rind marks, caused by the entry of the insect’s feeding tube, often with an exudation of gum, also distinguish the insect injury from boron-deficiency symptoms (Weir and Sarooshi, 2002).

Citrus is sensitive to an excess of boron and to its use where it is not needed. Only apply a boron fertilizer or spray if you are sure that boron is deficient (Zekri and Obreza, 2008).

5. Diagnosis of nutrient problems

Trained, experienced agronomists can often look at the leaves of crops and diagnose deficiency symptoms and other soil problems. Because of the large number of interactions, such as crop, variety, growth stage, weather conditions, tillage, land history, and other factors, this is often successful only if the agronomist can personally observe the crop. However, in some cases deficiency symptoms are difficult to diagnose in the field. The crop may not show sufficient symptoms to diagnose the problem, called "hidden hunger" in fertilizer promotions, or have multiple nutritional problems. Herbicide damage and diseases may make it difficult to clearly determine deficiencies and toxicities. Tissue sampling and analysis can be used to establish the deficiency or toxicity. The concentrations of nutrients in specific leaf tissues, response to nutrient addition, and crop yields have been shown to be highly related (Reuter and Robinson, 1986; Chang *et al*., 1992; Sivasin, 1994; Puranapong, 2005).

5.1 Plant tissue analysis

The analysis of tissues can be used to identify observed nutrient deficiencies, to survey geographical areas for possible problems, and to distinguish between nutrient problems and herbicide damage or other problems. Since plant symptoms are an anifestation of a lack or a buildup of particular compounds in plants, which may result from a variety of causes, the measurement of nutrient concentrations is a much clearer indication than the symptoms, and use of tissue analysis began as early as 1926 in the United States. Generally the youngest fully developed leaf of plants gives the largest range between sufficiency and deficiency. At early growth stages, whole plants can be used. Nutrient deficiencies may occur and then disappear at various stages of growth, but the most critical period to develop yield is the time of flowering and very early fruiting stage (Reuter and Robinson, 1986; Chang *et al*., 1992; Sivasin, 1994; Puranapong, 2005).

Scientists have developed indices of concentrations of nutrients in specific plant parts at certain growth stages. A system called the Critical Nutrient Range (CNR) establishes ranges of concentrations of nutrients for many crops, which delineate sufficiency, deficiency, and toxicities. This approach works quite well when one nutrient is mainly limiting. Occasionally, however, excesses of one nutrient, or deficiencies of several nutrients, cause the CNR to give misleading indications. This has prompted scientists to use nutrient concentration ratios to determine antagonisms between nutrients, such as N/S, K/Mg, K/Ca, N/P, and others. Comprehensive consideration of the many possible nutrient interactions is needed. In addition, it should be noted that nutrient concentrations may be high because nutrient deficiencies cause metabolic processes and growth to decrease. If one of the deficient nutrients is added, growth may then increase and other nutrients are shown to be deficient because their concentrations are essentially diluted in the tissue (Reuter and Robinson, 1986; Chang *et al*., 1992; Sivasin, 1994; Puranapong, 2005).

A system called DRIS (Diagnosis and Recommendation Integrated System) was developed by a South African researcher to overcome these limitations. The system compares ratios of the nutrient concentrations and can help determine when nutrients are in balance relative to each other. This gives a better indication of which nutrients are deficient (Reuter and Robinson, 1986; Chang *et al*., 1992; Sivasin, 1994; Puranapong, 2005).

The analysis of plants gives a reliable indication of what nutrients are actually available and recovered by the crop. However, in many cases, that knowledge comes too late to apply fertilizer to affect the growth of the current crop; it is likely useful for succeeding crops or for perennial crops. Tissue analyses also require fairly sophisticated instrumentation and a knowledge base for the interpretation. To maximize the benefits of money spent on fertilizers and to avoid crop nutritional problems and environmental problems, farmers need the techniques of soil testing and tissue analysis to determine which nutrients are needed and in what quantities (Reuter and Robinson, 1986; Chang *et al*., 1992; Sivasin, 1994; Puranapong, 2005).

5.2 Soil testing

Adjacent fields having the same soil but different fertilization and cropping histories develop different levels of soil fertility over time. This fertility level greatly affects the crop response to particular nutrients and obviously the most profitable fertilizer rate. If the farmer has a good estimate of the fertility status for each nutrient anticipated crop growth conditions, and the relation of nutrient addition and crop yield, he could fertilize at a rate that maximizes his profit given the prices for the fertilizer and the crop. This is difficult for most farmers to do, but soil testing can remove some of the guesswork Soil testing (often called somewhat inappropriately "analysis") is a process of taking a representative soil sample from a field and testing it for available nutrients. From this test, expected yield, and prices, a recommendation is made of how much fertilizer should be applied to maximize the profit for the farmer. Although not an exact science, the process has proved its worth in the past 50 years. Soil testing becomes more important after fertilizers are used for some years since large differences in fertility between fields in an area will develop, and P and K reveres increase in the soil. It is therefore very cost-effective to establish more accurately the necessary rates and monitor the soils for other potential problems (Reuter and Robinson, 1986; Chang *et al*., 1992; Sivasin, 1994; Puranapong, 2005).

Soil testing uses quite rudimentary analytical procedures; the problem is to select the correct extractants and measure parameters that are indices of the availability of each nutrient. This requires a chemical extraction or other method that gives numbers for the fertility status of each nutrient or other characteristics that relate well to expected crop response. Normally categories of (1) very deficient, (2) deficient, (3) sufficient, and (4) high are satisfactory. The test is analogous to a blood test. Knowing that your red blood cell count is 1,252/cc is not particularly useful unless you know if that number is considered normal, high, or low. Similarly, is an extractable P level of 15 ppm. P high or low is 150 kg K per hectare sufficient or deficient? Some agronomists spend their careers conducting experiments that allow them to relate the numbers of the tests to the most profitable use of fertilizers, and a great amount of information exists on soil tests and expected fertilizer response for different crops (Reuter and Robinson, 1986; Chang *et al*., 1992; Sivasin, 1994; Puranapong, 2005).

Soil testing procedures are used to measure soil pH and in places where they might be problematic, soil salinity and alkalinity (in soil science terms this refers to the Na content, not pH). This can be used to suggest management procedures, selection of crop, and liming recommendations and providing information on irrigation and drainage problems. The soil test recommendations give farmers guidelines for not only rates of each nutrient but also advice on the most efficient methods of application and timings of fertilizers. A major problem that occurs when soil testing is not used with fertilizer use is that soils become too fertile in certain nutrients. This not only causes inefficiencies but also can cause new nutrient problems to occur, excess acidity, contamination of water resources by nutrients, and excess leaching losses of other nutrients. Testing for N is problematic because of the rapid fluctuations in available N that can occur. However, if the soils, climate, and crops are well known, agronomists can make good recommendations based on past cropping and yield potential. In some cases they may additionally base it on a soil organic matter test. In recent years, more accurate N recommendation procedures have been developed by testing the soil for nitrate before N fertilizer topdressings to small grains or sidedressings on row crops. Chlorophyll meters, which measure the intensity of the green color of leaves, have also been used successfully (Reuter and Robinson, 1986; Chang *et al*., 1992; Sivasin, 1994; Puranapong, 2005).

The measurement of P availability in soils requires very good application of the art of soil testing because the soil extractant must remove P from many minerals of different solubilities in amounts proportional to the amount of P that will become available to the crop during growth. The P extractants are therefore chosen depending on prevalent soil characteristics of the area to give the best possible relationship between extractable P and crop response. The P test is one of the more valuable tests since it is a relatively expensive nutrient that can build to excess levels and cause deficiencies of other nutrients, such as Zn. Information on the most profitable application rate and the best method of applying P at particular soil test levels and with particular crops is valuable information for farmers (Reuter and Robinson, 1986; Chang *et al*., 1992; Sivasin, 1994; Puranapong, 2005).

The test for K is more straightforward and universal than that for the other nutrients. This test measures the amount of K^+ on cation exchange sites by displacing the K^+ with an excess of another cation. However, knowledge of K fixation and release of exchangeable K^+ on specific soils is very important to the interpretation. On a sandy soil, 150 kg K ha^{-1} may be quite sufficient, whereas on a clayey soil, profitable responses to additional applied K up to tests of 350 kg K ha^{-1} may be found. Similar tests for Ca and Mg may be conducted on soils suspected of having these deficiencies. The availability of other nutrients, such as S, Zn, B, Cu, Cl, Mo, Mn, and Fe, can be measured in soils, but these tests are far more difficult to conduct. Plant tissue testing is often used more successfully to diagnose these deficiencies (or toxicities) (Reuter and Robinson, 1986; Chang *et al*., 1992; Sivasin, 1994; Puranapong, 2005).

Despite its limitations, soil testing is a very valuable tool to improve the efficiency of fertilizer use and to avoid many soil management problems. In addition, it gives a scientific basis for fertilizers to be sold to farmers. Most agronomists believe fertilizers should not be used over a long period without a small investment in soil testing. The possibilities for creating new problems and the inefficiency of use require that soil testing be used (Reuter and Robinson, 1986; Chang *et al.*, 1992; Sivasin, 1994; Puranapong, 2005).

6. Achievement research in Thailand

Nowadays, some researchers start performing the plant and soil analysis to cope with the fertilizer management. Supakamnerd *et al*. (2005) studied the determination of nutritional requirement of citrus by means of plant analyses. The results showed that the concentrations of each element varied as leaf aged without any specific pattern and decreased when new shoots emerged. Moreover, it was indicated that fruit developed quite slowly at the first three months and the growth rate increased until one month before the harvest and slowed down until the harvest. The other of them Supakamnerd *et al*. (2002) studied the determination of nutrient requirement and nutritional status of lychee by means of plant analyses. It was found that concentrations of essential nutrients were higher in the leaves than the other plant parts except for the concentrations of iron, copper and zinc which were higher in the small roots

While Kumlung *et al*. (2003) studied the leaf analysis as a diagnostic tool and guide to fertilization of Lychee in the central plain of Thailand. The results showed that the differences among nutrient contents in the first, second and third pairs of newly flush leaves on a single shoot were not statistically significant in N, K, Fe, Cu and Zn concentration but significant differences in P, Mg, Ca and Mn concentration.

Kaosumain *et al*. (2002) studied the longan declined disease solution conducted in commercial longan orchards in Chiangmai and Lamphun provinces to analyze the major and minor elements. It was shown that the high phosphorous in soil was affected on the absorption of some elements such as zinc and copper. Definitely the protection and direction of declined disease as follows; first, soil element management by using soil analysis results for applying the fertilizer and apply trace element to nourish the plant.

Lastly, Poovarodom *et al*. (1998) studied the plant analysis as a means of diagnosing nutritional status and fertilizer recommendations for durain. The first part was to study the effects of fertilizer N (1,000, 1,500 and 2,000 gN/tree) and K (2,000 and 3,000 gK_2O/tree) on nutrient concentrations in leaf and yield of Mon Thong durian. It was found that the concentrations of N and Mg with durian yield were not significantly influenced by N and K application whereas K and Ca were slightly affected. The change in Mn, Zn and B concentrations were very slight and would not have been of practical importance in diagnosis by leaf analysis.