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Horticultural Research in Japan. Production of vegetables and ornamentals in hydroponics, constraints and control measures

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Abstract: Japan has a very mountainous topography, steep landforms, and varied climate making it less suitable than other countries for growing horticultural crops. However, good crop production technologies make it possible to grow high quality produce in Japan. The major horticultural crops are mainly produced in greenhouses and high tunnels however, a vast majority of greenhouse systems are outdated. Horticultural production practices could be more sustainable through the use of hydroponics in the greenhouse. Commercial hydroponic systems have proved more productive (20-25% higher) than conventional systems of agriculture. A number of research projects have been conducted to better understand production constraints in hydroponics, such as autotoxicity, and to identify suitable cultivars and supplementation techniques. This review discusses greenhouse production techniques for vegetables and ornamentals in hydroponics, as well as constraints and means for sustainable production.

1. Introduction

Japan is located between 24° and 46° north latitude and has four distinct seasons, spring, summer, autumn, and winter. It has a very mountainous topography and steep landforms that are poorly suitable for cultivation of horticultural crops. However, despite the rough mountainous terrain and poor soils, the Japanese have still managed to be successful in growing many crops. Japan enjoys mostly a temperate zone climate which varies considerably in different parts because the country stretches from north to south and is surrounded by the sea. The main horticultural crops include apples, grape, cherries, melon, onions, sweet corn, carrots, tomatoes, Japanese radish, Chinese yams and beans. Several types or floricultural crops are also produced and exported to other countries (Araki, 2002). Tomato, cucumber, welsh onion, strawberry, watermelon and spinach are mostly produced in greenhouses and high tunnels. Strawberries are usually grown on high beds for comfortable work and lower labor costs whereas, tomatoes are produced in glasshouses and high tunnels in order to prevent crop damage by rain (Araki, 2002).

Japan uses 69% of its total greenhouse area for vegetable production, only 17% for flowers, and 13% for fruit tree production. Only 3% of greenhouses use the modern facilities suitable for crop production through hydroponics (Nichols, 2008) although a gradual change has been taking place in greenhouse production. More sustainable horticulture practices in greenhouses are desirable and possible through the use of hydroponic methods that allow only small amounts of water to drain off and usually offer a higher yield. Commercial hydroponic systems have proved to be more productive than conventional systems of agriculture. Hydroponics has averaged around 20 to 25% higher yields than conventional soil cultivation. This could truly benefit Japan's current GDP in agriculture while feeding more people with sustainable and fresh horticultural crops. Conventional hydroponic techniques use culture solution with bare root systems and do not use growing media. On the other hand, soilless cultivation is a very sustainable crop production practice because many of the components in modern media such as perlite and vermiculite take many years to decay and go back into the environment. The improved and consistent vegetable quality is second to none because of the elimination of pesticides and herbicides. This keeps toxic chemicals away from the culture system and it will in turn help keep Japan's groundwater and oceans free from contamination. Hydroponics is currently a managed culture technique

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that is largely used for the production of vegetables as it is capable of being sustainable with low amounts of water loss and use of soilless media. Therefore, advanced management of this technique has been practiced throughout Japan with an aim toward yield maximization and higher quality assurance. However, constraints have been found and several researchers have tested strategies to overcome them. This review discusses greenhouse production techniques of vegetables and ornamentals in hydroponics, and the constraints and means for sustainable production.

2. Hydroponics for higher yield and production of specialty horticultural crops

Hydroponics is a method for growing plants using mineral nutrient solutions, in water, without soil. In this system plants can be grown with their roots in the mineral nutrient solution only orin an inert medium. It is possibly the most intensive method of crop production providing efficient use of water and mineral nutrients with a minimal use of space. It has been used successfully by commercial growers for fast-growing horticultural crops such as lettuce, strawberries, tomatoes, cucumbers and ornamentals in Japan. This technology enables a more precise control of growth conditions, making it easier to study the variable factors or parameters. One specialty of this technique is the vigorous development of root systems and the efficient uptake of the essential nutrients from culture solution resulting in a better crop yield. Therefore, this technology has gained popularity for producing high value crops with high quality and specialty production providing human health benefits. However, there are limits due to self-toxicity from root exudates in recycled nutrient solutions. Finding suitable control measures to overcome these constraints could result in sustainable horticultural crop production in greenhouses. In the following sections, hydroponic production techniques for root crops (carrot), production of functional foods for kidney dialysis patients (melon), and enhanced crop quality (turnip) will be discussed.

Growing carrots hydroponically using perlite substrate

Root vegetables are often discouraged to grow by hydroponics possibly because of poor root development. Carrot [*Daucus carota* L.] is a root vegetable which forms numerous hairy roots with a reduced tap root in the nutrient solution. Moreover, inside nutrient solution this storage organ form hypertrophy due to the ample supply of water and nutrient resulting decrease in length and weight compared to non-submerged condition (Terabayashi *et al.*, 2008). Therefore, appropriate moisture content of the growth medium is crucial for optimum growth and development of the storage root (Eguchi *et al.*, 2008). It has been found that the use of suitable soilless media can increase both marketable yield and quality of root crops by many folds (Hanna, 2009). Perlite is widely preferred as soilless media, as it encourages faster root development, reduces risk of damping off, avoids water logging and provides an optimum balance of air and water. Its strong attraction for water automatically draws up solution from the reservoir at the same rate that the plants remove water, leaving excess solution in the reservoir. Therefore, an optimum moisture level can be maintained around the root, and this is a significant advantage over rockwool with less capillary action. Recently perlite has emerged as an excellent medium with versatile use. It has been widely used to grow many horticultural crops including tomatoes, cucumber, melon, peppers, lettuce and rose (Szmidt *et al.*, 1988; Cantliffe *et al.*, 2003; Hochmuth and Hochmuth, 2003; Fascella and Zizzo, 2005; Frezza *et al.*, 2005; Rodriguez *et al.*, 2006).

Suitable perlite size, along with optimal concentration of culture solution, has been suggested for maximizing carrot yield in hydroponics (Asaduzzaman *et al.*, 2013 a). It was found that carrot plants grown in 0.6 mm perlite supplied with 100% nutrient solution produced significantly higher root yield compared to larger perlite particles and higher concentrations of nutrient solution. Interestingly it was observed that when carrots are grown in 0.3 mm perlites, shorter roots are produced which are wider near the proximal end and whitish in the distal end due to excessive water content causing oxygen deficiency. This ultra fine perlite can hold excessive water causing oxygen deficiency in the substrate air zone and as a result roots become whitish with a reduced amount of carotenoids. In the study, the feasibility of growing carrot in used perlites from previous culture was also evaluated. The residual nutrients available in the reused perlite lowers in turn the demand of nutrients in the second culture where carrot plants can uptake about 50% residual nitrogen. Therefore, it was recommended to use 0.6 mm perlite and 100% (for first culture) or 75% (for second culture) 'Enshi' nutrient solution for growing carrots hydroponically for maximum yield and higher quality.

Production of low potassium content melon fruits in hydroponics through managing nutrient solution

Potassium is crucial for the normal functioning of muscles, heart, and nerves in the human body. It is one of the main electrolytes and is concentrated within the body cell. About 90% of body K is normally excreted by the kidneys but patients with kidney dysfunction suffering from chronic kidney disease (CKD) cannot completely excrete it and thus it accumulates. Abnormally elevated levels of K in the blood (hyperkalemia) can cause adverse effects (Kes, 2001). This author also reported that a normal kidney has the capacity to excrete in excess of 400 m mol K day^{-1}, and it is unlikely that an individual will become chronically hyperkalemic without some degree of chronic renal impairment. Sometimes hyperkalemia causes arrhythmias, muscle weakness, disturbed consciousness, heart failure, and can even lead to sudden death (Spital and Stems, 1988; Putcha and Allon, 2007). The CKD patient population is increasing year by year and it is expected that the total number of patients will continue to increase progressively. Therefore, preventive measures have been stressed to divert the prevalence of this disease in a decreasing trend. Restricted diets are mainly used as a means of treatment for CKD patients. As a primary control measure, foods with high K content are restricted but a normal daily diet including fruits such as melon, fresh vegetables, seaweed, beans and potatoes are rich in K (Weiner and Wingo, 1998). K is a major nutrient, essential for normal growth and development of plants (Schachtman and Liu, 1999). Plants absorb more K than any other mineral element with the exception of N (Tisdale and Nelson, 1975; Mäser *et al*., 2002; Britto and Kronzucker, 2008; Szczerba *et al*., 2009). It is the only monovalent cation that is essential for all higher plants and it is involved in three major functions: enzyme activation, charge balance, and osmoregulation (Mengel, 2007; Szczerba et *al.*, 2009). It is inevitable that reduced K supply will inhibit plant growth and yield. Therefore, investigations on minimal requirements of K in plants to maintain their normal growth and development are important. In the above context, investigations have been conducted for the production of melon fruits with low K content to provide supplementary diet to dialysis patients (Asao *et al*., 2013).

In Japan greenhouse cultured raw melon generally has higher K content: 340 mg 100 g^{-1} fresh weight (MEXT, 2011). This amount of K was decreased considerably, improving the diet of dialysis patients (Asao *et al*., 2013). The researchers applied a hydroponic method which enabled a more precise control of growth conditions and made it easier to study the variable factors or parameters. Regular nutritional testing can be conducted in hydroponics, making it possible to determine if the desired amount of nutritional content is present in the plants or not. Precise control over the concentration and composition of culture solution allows the production of either mineral enriched or deficient fruits or vegetables.

Results indicated that a general trend of decreasing K content in fruit with a decrease of KNO_3 concentration in the nutrient solution. In spring 2009, the K content of fruits harvested from plants grown in the nutrient solution with 1/4th KNO_3 was about 39% lower compared to those harvested from plants grown in standard nutrient solution, while it was about 35% and 43% lower in fruits from plants grown with 1/16th and without KNO_3 in the spring 2010 and 2011, respectively. The content of Na in fruits however increased progressively with the decrease of K in the nutrient solution. A consistent antagonistic relation with fruit K concentration was found due to the reduced levels of KNO_3. Compared to control plants, about 83% (spring 2010) and 51% (spring 2011) increased Na were found in fruit harvested from plants grown in nutrient solution with 1/6th or without K, respectively. Low K content melon fruit can provide human health benefits to dialysis patients but the higher Na content would likely cause hyperpiesia and edema. Therefore, it is necessary to evaluate the benefits of the reduction of K against the risks of the increased Na intake by the dialysis patients, whose K intake should be restricted to 1500-2000 mg day^{-1} (Agondi *et al*., 2011) and NaCl intake (equivalent to 2000-3200 mg Na) should be restricted to 5000-8000 mg day^{-1}. Na intake must be limited to 1.3-1.6 times of K intake. However, the benefits of reducing the intake of K are greater than the risks of increasing the intake of Na.

Nutrient level changes in anthocyanin and nitrate-N contents in hydroponically grown turnip

"Tsuda-kabu" (*Brassica rapa* L. rapifera group) is a local turnip cultivar popularly grown in Shimane prefecture in Japan. The cultivar is red with a comma-shaped root. The crop is normally sown in September and harvested in December. Harvesting turnip in early December is profitable mainly since many people in Japan send it as a year-end gift. However, sowing turnip in September in field conditions is often delayed due to incessant rainfall. Moreover, turnip in soil culture requires extra labor for harvesting and washing. In hydroponics, turnip cultivation seems to be less influenced by environmental changes and harvesting, and processing is also convenient. Another problem with soil cultivation is that most of this turnip root remains above the ground and only a few centimeters of the root remains in the soil. This red turnip has large amounts of anthocyanin in the root, adding quality for processing and delicious foods. Previous research reported that nitrate inhibited the sucrose-induced accumulation of anthocyarine in grapevine (Faust, 1965; Pirie and Mullins, 1976). Thus, an optimum level of nutrients in the culture solution is a precondition for harvesting quality products with maximum yield through hydroponic culture.

In this context, Asao *et al*. (2005) investigated the influence of different nutrient solution concentrations on the growth of turnip in hydroponics. It was found that NO_3^--N contents in the shoots and roots of turnip were decreased with decreasing NO_3^--N levels in the nutrient solution. On the other hand, anthocyanin concentration in the root was inversely proportional to the content of NO_3^--N in the same. However, when NO_3^--N content at 50% nutrient solution concentration was changed from full (8 m mol^{-1}) to half (4 m mol^{-1}), NO_3^--N content and anthocyanin contents in the turnip root were not affected. These results indicate that the effects of other nutrients except NO_3^--N might be associated with the formation of anthocyanin in turnip. Therefore, they suggested that the medium range (50%) nutrient solution with 4 m mol^{-1} NO_3^--N may be treated as optimum concentration for production of quality turnip by hydroponic culture.

3. Production constraints in recycled hydroponic culture

Autotoxicity in cucumber (Cucumis sativus L.) under closed hydroponics

Autotoxicity in cucumber plants was reported in previous research as due to accumulation of phytotoxic phenolic compounds in the substrates used for long-term cultivation (Politycka *et al*., 1984; Yu and Matsui, 1994). Root exudates from the cucumber cv. Shougoin-Aonaga-Fushinari gave maximum inhibitory effect on seedling growth of its own cultivar, as well as other test cultivars of cucumber

(Asao *et al*., 1998 a). The growth of tomato (Yu and Matsui, 1993 a, b) and cucumber plants (Yu and Matsui, 1994) increased upon addition of activated charcoal (AC) to the nutrient solutions. Yu and Matsui (1993 c, 1994) collected the root exudates of cucumber cv. Tokiwa from hydroponic culture through a continuous trapping technique (Tang, 1986) and identified a number of autotoxic chemicals.

Fruit yield of cucumber was improved by the addition of AC to the nutrient solution (Asao *et al*., 1998 b) because the added charcoal adsorbed the phytotoxic root exudates from the nutrient solution and thus favored cucumber growth. It was also found that the fruit yield of cucumber plants decreased significantly at late reproductive stage (2 weeks ahead of final harvest) while this inhibition was recovered if nutrient solutions were renewed biweekly or supplemented with AC to the nutrient solution (Asao *et al*., 1998 a). Characteristic shrunken cucumber fruits were harvested from the plant grown in non-renewed culture solution. Autotoxicity of cucumber was also found to be different among cultivars (Asao *et al*., 1998 b). Fruit harvesting of a susceptible cucumber cultivar grown in a closed nutrient flow system was prolonged by grafting onto a non-autotoxic cultivar (Asao *et al*., 1999 b). Thus, cucumber root exudates from a closed hydroponic system were analyzed and 2,4-dichlorobenzoic acid (DCBA) was found to be the strongest inhibitor among a number of growth inhibitors detected (Asao *et al*., 1999 c; Pramanik *et al*., 2000).

Autotoxicity in strawberry (Fragaria × ananassa Duch.) under closed hydroponics

In Japan, closed hydroponics have been considered feasible for strawberry cultivation (Takeuchi, 2000; Oka, 2002; Koshikawa and Yasuda, 2003), however it was reported that a yield reduction, caused by unknown factors, occurred in this production system for strawberry (Oka, 2002). Many researchers showed that root exudates can be removed by adding AC to the nutrient solution (Koda *et al*., 1977; Asao *et al*., 1998 a, 1999 c; Sato, 2004). Subsequently, Kitazawa *et al*. (2005) identified potential chemicals in root exudates of strawberry and investigated the effects of non-renewed nutrient solution, and on vegetative and reproductive growth of strawberry with the addition of AC.

Non-renewed nutrient solution resulted in a significant decrease in the growth of strawberry plantlets compared to growth when the nutrient solution was renewed. The number of flower clusters, flowers and fruits harvested all decreased in non-renewed nutrient solution. This phenomenon was also evident in cucumber (Asao *et al.*, 1998 a), mitsuba (Koda *et al*., 1980) and rose (Sato, 2004). GC-MS analysis of strawberry root exudates revealed the presence of lactic, benzoic, succinic, adipic, and p-hydroxybenzoic acids. These acids showed phytotoxicity during bioassay and benzoic acid was found to be the strongest inhibitor of vegetative and reproductive growth in strawberry plantlets. Therefore, it was concluded that reduction in performance of strawberry grown in closed hydroponic systems occurred thorough autotoxic root exudates from the strawberry plant itself.

Autotoxicity in some leaffy vegetables when grown in recycled hydroponics

Yield reduction due to continuous culture of numerous crops has been demonstrated by researchers around the world. Allelopathic effects from crop residues and root exudates have been extensively studied in crops such as alfalfa (Miller, 1983; Nakahisa *et al.*, 1993, 1994; Chon *et al*., 2002, Chung *et al*., 2011), asparagus (Young, 1984; Young and Chou, 1985; Hartung *et al*., 1990), cucumber (Yu and Matsui, 1994, 1997), watermelon (Kushima *et al*., 1998; Hao *et al*., 2007), taro (Asao *et al*., 2003), strawberry (Kitazawa *et al.*, 2005), tomato (Yu and Matsui, 1993 b), lettuce (Lee *et al*., 2006) and so on. Thus, autotoxicity is considered to be one of the causes of growth retardation on the successive culture of vegetables.

It was found that many leaffy vegetables demonstrated autotoxicity when grown in recycled hydroponics (Asao *et al*., 2001 a). In another study autotoxic substances from eight leaffy vegetables were identified as lactic, benzoic, m-hydroxybenzoic, p-hydroxybenzoic, adipic, succinic, and vanillic acids (Asao *et al*., 2004 b). Benzoic acid was the strongest inhibitor overall for leaffy vegetables among the substances tested. These results confirm that some unidentified compounds inhibited growth of leaffy vegetables in hydroponic culture (Asao *et al*., 2001 b).

Autotoxicity in taro (Colocasia esculenta Schott.) under closed hydroponics

Yield of taro plants decreased when cultivated consecutively for several years on the same land (Takahashi, 1984). Rotation with other crops for at least three years (Miyoshi *et al*., 1971), in combination with organic matter and soil disinfectants (Murota *et al*., 1984), has been suggested to improve the yield of taro. However, even in a fixed crop rotation system, there was a great difference in the growth and yields of taro plants. Miyaji and Shirazawa (1979) found that taro residues in soils after harvest were inhibitory to its growth.

Asao *et al.* (2003) identified the chemicals exuded by taro roots and evaluated their phytotoxicity on growth and yield of taro using a hydroponic system. GC-MS analysis of taro root exudates detected methyl esters of lactic, benzoic, m-hydroxybenzoic, p-hydroxybenzoic, vanillic, succinic, and adipic acids. Among them benzoic and adipic acids significantly inhibited the growth of taro plantlets at concentrations ranging from 0 to 400 $\mu M\ L^{-1}$. Benzoic acid affected growth of taro plants even at 50 $\mu M\ L^{-1}$. Their conclusion was that the decline in yield in successive culture of taro appeared to be related to the allelochemicals exuded from taro plants.

Autotoxicity in some beans grown with non-renewed culture solution

Edible beans are grown as vegetables and intensively cultivated in the same farmland year after year. The production of these common bean plants and other perennial legumes declines in replanting conditions owing to autotoxicity, a form of intraspecific allelopathy that occurs when a plant species releases chemical substances that inhibit or delay germination and growth of the same plant species (Putnam, 1985; Miller, 1996; Singh *et al.*, 1999). Allelopathy has been investigated in some beans such as *Pisum sativum* (Kato-Noguchi, 2003), *Mucun pruriens* (Fujii *et al.*, 1991), *Glycine max* (Huber and Abney, 1986; Xiao *et al.*, 2006; Yan and Yang, 2008), and *Cicer arietinum* (Yasmin *et al.*, 1999). In field experiments, it has been reported that residues and extracts of pea plants suppressed the growth and population size of several plant species (Purvis, 1990; Schenk and Werner, 1991; Tsuchiya and Ohno, 1992; Akemo *et al.*, 2000). Phytotoxic substances in *Pisum sativum* root exudates have been reported by several researchers (Hatsuda *et al.*, 1963; Yu and Matsui, 1999) and, recently, pisatin has been identified as an inhibitory chemical from its shoots (Kato-Noguchi, 2003).

Removal of the inhibitory chemicals from soils or culture solution can permit continued crop cultivation on the same land for several years. Hydroponic culture technique has the ability to trap and isolate the chemicals released through plant roots. Elimination of these growth inhibitors from recycling culture solution is desirable from the viewpoint of conservation-oriented agriculture. Thus, identification of the allelochemicals from bean root exudates, evaluation of their phytotoxicity, and their removal would facilitate the maintenance of profitable crop production. Asaduzzaman and Asao, (2012) studied autotoxicity in *Pisum sativum*, *Phaseolus vulgaris*, and *Vicia faba*, and their responsible allelochemicals using hydroponics. They also evaluated phytotoxicity of the identified allelochemicals using seedling growth bioassay of the test plants. Results indicated that yield of these beans decreased greatly (over 50%) when grown in non-renewed culture solution, however addition of AC in the culture improved the yield significantly. The identified allelochemicals were benzoic, salicylic, and malonic acids in root exudates of *P. vulgaris* and lactic, benzoic, p-hydroxybenzoic, vanillic, adipic, succinic, malic, glycolic, and p-hydroxyphenylacetic acids in *V. faba*. Bioassay of the identified allelochemicals revealed that benzoic, salicylic, and malonic acids significantly reduced the growth of *P. vulgaris* even at low concentrations.

Autotoxicity in some ornamental plants evidenced in recycled hydroponics

Asao *et al.* (2007 a,b) investigated autotoxicity in some selected ornamentals along with a possible remedial measure to overcome growth inhibition. Among the 37 plants under study, growth of lily, prairie gentian, corn poppy, farewell-to-spring, rocket larkspur, and carnation was drastically reduced in the absence of AC, compared with those in the presence of AC in the nutrient solution. In this study the added AC adsorbed phytotoxic root exudated from nutrient solution leading to improved plant growth. Thus use of AC showed potentiality of overcoming autotoxicity under recycling hydroponics. Root exudates of some plants were analyzed and several organic compounds were detected. Strong growth inhibitors such as lactic acid in pot marigold, benzoic and p-hydroxybenzoic acids in lily, o-hydroxyphenylacetic acid in rocket larkspur, benzoic and p-hydroxybenzoic acids in sweet pea, and maleic and benzoic acids in prairie gentian were detected in the root exudates. The reduced growth of prairie gentian after prolonged cultivation in a field suggested that it could be avoided by amending the soil with AC at a rate of 60 kg 10 a^{-1}.

4. Causes of crop yield reduction in non-recycled hydroponics

Decrease of cucumber yield in non-renewed culture solution

Investigations were conducted to clarify the reasons for fruit yield reduction during a late growing period of cucumber cultured in hydroponic nutrient solution which was not renewed completely (Asao *et al.*, 1998 a). It was found that vegetative growth was unaffected by biweekly renewed or non-renewed nutrient solution whereas, fruit yield decreased when nutrient solutions were restored during culture as compared to total renewed or supplemented with AC.

Influence of isolated phenolics on fruit yield in cucumber

Phenolics isolated from the nutrient solution growing cucumber plant had significant influence on fruit yield (Asao *et al.*, 1999 a, 1999 c). The researchers isolated and identified growth inhibiting substances of unknown origin in the nutrient solution culturing cucumber plants. The growth inhibitors were adsorbed on the AC and extracted by an organic solvent. The active substances were analyzed by GC-MS as benzoic acid, p-hydroxybenzoic acid, 2, 4-dichlorobenzoic acid (DCLBA) and phthalic aicd. Among these substances, DCLBA exhibited the strongest inhibitory activity toward cucumber seedlings and thus it was considered the most effective allelochemical of cucumber. It was also found to cause growth inhibition of cucumber seedlings in a seedling growth bioassay. Therefore, it was assumed that this phenol was one of the compounds responsible for the inhibition of cucumber growth in non-renewed hydroponic culture solution. Other researchers have also shown that combinations of certain phenolics can have interaction effects on the germination of various crops and weed species (Williams and Hoagland, 1982). Combination of DCLBA (20 µmol $liter^{-1}$) with benzoic acid, p-hydroxybenzoic acid, and phthalic acid resulted in growth suppression of cucumber (Asao *et al.*, 1999 a).

The effects of DCLBA on the number of harvested cucumbers were evaluated by split-root system hydroponic culture (Asao *et al.*, 2001 c). DCLBA applied in the nutrient solution at 10 µmol $liter^{-1}$ severely damaged the roots by disrupting the integrity of epidermal cells, and remarkably inhibited the uptake of NO_3^-, $H_2PO_4^-$, and K^+ ions leading to a decreased number of harvested cucumber fruits. DCLBA is a herbicide and also a synthetic auxin, and it stunts cucumber plants at low concentration but kills them at higher levels. This allelochemical was applied through nutrient solution to cucumber seedlings grown hydroponically and exposed to two microorganisms (TS-22 and TS-29) and one Rhizoplane ACI, isolated from soil and cucumber roots (Asao *et al.*, 2001 b). It was found that cucumber seedlings not exposed to DCLBA and different microorganisms grew vigorously, whereas those exposed to this chemical were stunted.

Effect of temperature and photoperiod on phytotoxins exudation in cucumber

Light and temperature have profound effects on the quality and quantity of exudates because they affect the processess of photosynthesis, translocation, and respiration in plants (Hale *et al.*, 1971; Hale and Moore, 1979). An increase in exudation at high temperature has been reported for many crops (Rovira, 1959, Vancura, 1967; Rovira, 1969; Hale *et al.*, 1978). The effect of temperature and photoperiod has been assessed on the quality and quantity of growth inhibitors exuded from the root of cucumber (Pramanik *et al.*, 2000). The researchers found that exudation rate varied greatly with the kind of acids, temperatures, and photoperiods, ranging from 0.2 to 4.17 μg day^{-1} $plant^{-1}$. Exudation tended to increased with plant growth and maximum exudation rate was recorded with high temperature and long photoperiod, and minimum with low temperature and short photoperiod.

5. Possible measures to control autotoxicity under closed hydroponics

Selection of suitable crop cultivars

Screening for differential autotoxic potential of cucumber cultivars has been conducted in a closed hydroponic system using seedling growth bioassay (Asao *et al.*, 1998 b). In this study, some commercial cucumber cultivars were classified into four groups, such as PI 169391, Encore I, Hokushin and Aodai. The cultivars showed intermediate, high, intermediate and low sensitivity to growth retarding substances in culture solutions once used for different cultivars, while growth reduction was not found, except in PI 169391. These findings indicate that there are intraspecific variations in the autotoxic potential of cucumber. Thus, cucumber cultivars with less growth reduction of seedlings in culture solution once used for the same cultivars would be most suitable for culturing in closed hydroponics.

Species differences in autotoxicity susceptibility

In another study species differences in susceptibility to autotoxicity among sixteen leaffy vegetables were studied in hydroponics (Asao *et al.*, 2001 a). Among the species, parsley inhibited most severely in the absence of AC followed by celery, edible burdock, garland chrysanthemum, kale, curled lettuce, pak-choi, head lettuce and mitsuba, whereas komatsuna, Chinese cabbage, radish, takana, welsh onion, perilla and spinach were found to be unaffected.

Use of autotoxin-tolerant plant as rootstock

Fruit yield of cucumber in a closed nutrient flow system has been increased by grafting “Shogoin-aonaga-fushinari” on “Hokushin” or “Aodai” seedlings (Asao *et al.*, 1999 b). The number of harvested fruit of “Shogoin-aonaga-fushinari” in the summer crop was increased by grafting on the rootstock of “Hokushin” or “Aodai”. The decreased weekly number of harvested fruits in the late harvest period on ungrafted plants was not observed on plants grafted on “Hokushin” or “Aodai” seedlings, thereby extending the harvest season. There was no evidence found that rootstocks influence vegetative growth.

Use of bloomless rootstocks to increase number of harvested cucumber fruits

The number of harvested fruits of cucumber cultivar “Shogoin-aonaga-fushinari” grafted on bloomless rootstocks such as “Hikari-power” and “Kitora” was increased slightly when AC was not added, but increased greatly when AC was added in the culture solution (Asao *et al.*, 2000). These results indicate that root exudates influence the harvested fruit number, which can be limited in bloomless rootstocks.

Use of AC in addition to dissolved oxygen levels to increase cucumber fruit number

Investigation on the effects of dissolved oxygen levels and addition of AC has revealed a significant influence on fruit yield of cucumber in closed hydroponics (Asao *et al.*, 1999 d). Addition of AC had no influence on the dissolved oxygen concentration (1.7-3.1 ppm and 5.0-7.5 ppm after 6 and 24 h aeration, respectively) while fruit yield per plant increased significantly. In this study, aeration hours or dissolved oxygen did not show any effect on yield of cucumber.

Addition of AC to adsorb allelochemicals from recycled culture solution and/or replanting soil

Activated charcoal, with its large surface area, pore volume and polarity, has tremendous adsorption capacity for many organic compounds. In cucumber, tomato, and asparagus, increased productivity has been observed after using AC in non-renewed culture solution or replanting soil (Yu *et al.*, 1993; Yu and Matsui, 1994; Asao *et al.*, 2003; Motoki *et al.*, 2006). Mat-rush does not grow well if it is cultivated consecutively for years in the same land. The continuous cultivation of mat-rush accumulates toxic allelochemicals from roots in soil and inhibits subsequent plant growth. Fujitomi *et al.* (1999) reported an approximate 45% yield reduction in second year cultivation of mat-rush. Asao *et al.* (2007 b) reported that mat-rush seedlings grown in soils collected from fields consecutively cropped with mat-rush for three years had lower shoot dry weight compared to plants grown in soils amended with AC (138 and 165% lower dry weight compared to coarse and fine AC, respectively).

Mitigation of autotoxicity using microbial strain

Microorganisms can degrade chemical substances in soil and water (Sundin and Waechter-Kristensen, 1994) and phytotoxins, both autotoxins and microbial toxins (Caspersen *et al.*, 2000; Asao *et al.*, 2003, 2004 a; Chen *et al.*, 2011). Similarly, many isolates from suppressive soils or others can degrade autotoxins in the rhizosphere of continuously cropped plants (Asao *et al*., 2004 a; Chen *et al*., 2011). Growth and yield of cucumber plants significantly decreased with addition of DCLBA (the strongest growth inhibitor released during reproductive stage) to the recycled nutrient solution but growth recovered upon addition of microbial strains. It is suggested that microorganisms, if added to nutrient solution at the reproductive stage of cucumber, can catabolize autotoxic compounds from root exudates into non-toxic compounds and thus increase fruit yield.

Mitigation of autotoxicity by supplementation of 2,4-dichlorophenoxyacetic acid (2,4-D) and 1-naphthaleneacetic acid

Phenolic compounds disrupt the endogenous hormonal balance in plants (Rice, 1984; Asao *et al*., 2001 c). Benzoic acid or similar structural compound substitution has been considered as anti-auxinous (Keitt and Barker, 1966; Karabaghli-Degron *et al*., 1998). Callis (2005) reported that fruit enlargement and maturation of strawberry depends on auxin. Thus, the effects of foliar applications of 2,4-D and NAA on growth of strawberry were investigated to mitigate the autotoxicity in growing plants in closed hydroponic systems (Kitazawa *et al*., 2007). Supplementation of 5.4 µM NAA was found to be the most effective treatment for alleviating autotoxicity of strawberry and increasing fruit yield.

Mitigation of autotoxicity by supplementation of amino acids

Amino acids are the nitrogenous compounds which form the basic component of all living cells (Furuya and Umemiya, 2002). Therefore, they have great potential for use in culture techniques: recently they were applied as foliar spray to improve the growth, yield and quality of several crops (Mazher *et al*., 2011; Takeuchi *et al*., 2008). Similarly, supplementation of amino acids has been investigated to recover growth and yield of strawberry plants with autotoxicity in closed hydroponics. In greenhouse experiments, 22 water soluble amino acids were sprayed on strawberry plants at 2 ml per plant and among them glutamic acid and hydroxy-proline spray produced 50% greater fruit yield compared to control water spray in plants grown under non-renewed culture (Mondal *et al*., 2013).

Mitigation of autotoxicity through electrodegradation of root exudates

In recycled hydroponic culture, significant growth inhibition of strawberry has been observed due to accumulation of toxic root exudates in the nutrient solution; benzoic acid was found to be the most potent growth inhibitor in root exudates (Kitazawa *et al*., 2005). Electrochemical methods have also been applied for degradation/oxidation of phenols and their derivatives from organic waste or pollutants by several researchers. Phenolic compounds, including phenols, catecol and hydroquinone in aqueous solution and even benzene were found to decompose when treated by electro-degradation (Fleszar and Ploszynka, 1985; Comninellis and Pulgarin, 1991; Feng and Li, 2003). These compounds are oxidized rapidly at the anode and decompose to CO_2. Thus, electrodegradation may cause the decomposition of allelochemicals, including benzoic acid exuded in the nutrient solution from plants, offering a possibly useful tool to mitigate autotoxicity in strawberry. Based on these results, electrodegradation has been attempted for the decomposition of benzoic acid in the culture solution (Asao *et al.*, 2008). It was found that benzoic acid exogenously added to nutrient solution was almost completely decomposed within 24 h. Electrodegradation of nutrient solution could mitigate autotoxicity in strawberry plants grown in closed hydroponic culture and it recovered fruit yield up to 71% of control. The appropriate timing and intensity of electrodegradation of nutrient solution has also been investigated and it was recommended that application of electrodegradation to non-renewed culture solution for 2 h at four-week intervals can recover fruit yield completely (99%) compared to non-renewed culture solution without electrodegradation (Asaduzzaman *et al*., 2012).

Selection of ideal succeeding crops after asparagus, taro and beans

Successive culture of the same crop on the same land for years causes soil sickness or replanting injuries (Rice, 1984; Tsuchiya, 1990) resulting in reductions in both crop yield and quality. Among the possible reasons for this complex natural phenomenon self-allelopathy, or autotoxicity, has often been suggested (Asao *et al*., 2003; Asaduzzaman and Asao, 2012). This phenomenon has been evidenced in asparagus (Hartung and Stephens, 1983; Young and Chou, 1985; Lake *et al*., 1993), taro (Takahashi, 1984) and several beans (Putnam, 1985; Miller, 1996; Singh *et al*., 1999). Therefore, growth performances of 67 vegetable crop cultivars were evaluated through seedling growth bioassay using once used nutrient solution of asparagus and also replanting soil of asparagus, taro and three beans (Asaduzzaman *et al*., 2013 b). Strategies to overcome this problem in replanting soil or reuse of hydroponic culture solution have been suggested by many researchers. Bioassays using asparagus with used nutrient solution, with or without AC, suggest cucumber, garden pea, komatsuna, melon, pak-choi cv. 'Tyoukou', parsley, soybean (except cv. 'Tankurou'), cabbage cv. 'Early Ball' and lettuce cv. 'Shato' as possible succeeding crops. While, bioassays using replanting soil, with or without AC, have suggested that most of the cultivars tested can be planted after asparagus, taro, and three beans (*Vicia faba* L., *Pisum sativum* L. and *Phaseolus vulgaris* L.) with little adverse effects. Among the three methods of bioassay used (i.e. nutrient solution, direct seed sowing and seedling transplanting in replant soil) the nutrient solution bioassay proved more sensitive than replanting soil bioassay. However, results of nutrient solution bioassay may not be reproducible in field conditions. Therefore, the seedling transplanting method can be used as an easy and practical bioassay approach to select succeeding crops for fields with replanting problems.

6. Future challenges and endeavors in horticultural research in Japan

Currently Japan faces problems of an aging population, mainly in rural areas. In fact, many researchers are trying to breed new high quality cultivars to develop labor-saving and environmental-friendly technologies for horticultural cultivation. Tomato production in Japan is an especially up and coming market that needs to be looked at much more closely. In this regard, outdated hydroponic systems should be up-scaled with modern facilities. A number of favorable resources are found in Japan, such as rivers, the ocean, and a relatively temperate climate, and they can make greenhouse productions much more sustainable. Through the use of wind, solar and geothermal power these new greenhouses can be brought into sustainable horticultural production.

Recent advances in solar technology have created an outstanding 17% conversion efficiency in solar panels. These free-standing towers can be set up on the south end of greenhouses on an automatic swivel to rotate with the movement of the sun to ensure optimal light intensity at all times of the day. This solar energy collected during the day can be used to heat water tanks under the greenhouse for use during the night to heat the greenhouses as needed. Through the generation of electricity with wind and solar power, heating and electricity needs beyond these systems will be minimal, greatly reducing greenhouse electricity consumption from less sustainable sources. Therefore, development of Plant Factory supported facilities with this geothermal energy system should be expanded throughout Japan.

According to "Japanese Society of Nephrology" about 13.3 million people suffer from CKD (i.e. one in every eight adults) and it expected that the total number of patients will continue to increase progressively. Therefore, preventive measures should be stressed to divert the prevalence of this disease toward a decreasing trend. CKD patients are restricted from consuming K rich foods but our typical daily diet includes fruits, fresh vegetables, seaweed, beans and potatoes with high K content, making it difficult for sufferers to eat their usual diet with other family members. This restriction impacts on their quality of life greatly.In general, research efforts are directed at controlling the growth and yield of plants, however the complexity of plant responses to culture systems and environmental factors should be stressed in particular cases. Indeed, horticultural techniques can be a good way to improve fruit quality and increase the bioactive compounds in plants. Toward this aim, simple management of nutrient solutions, together with good knowledge about cultural practices of each particular crop and application of suitable crop technologies are key for controlling the nutritional composition of plants.

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