

A Comparison of Lycopene and Other Phytochemicals in Tomatoes Grown under Conventional and Organic Management Systems

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AVRDC

The World Vegetable Center

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Written by Heidi Lumpkin, AVRDC—The World Vegetable Center

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AVRDC—The World Vegetable Center is an international not-for-profit organization committed to alleviating poverty and malnutrition through research, development, and training.



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Contents

Chapters	i
Tables	i
Figure	ii
Acknowledgements	iii
Executive Summary	iv

Chapters

1	Introduction	1
2	Protocols for Site Selection of Farm Pairs for Comparative Studies	3
2.1	A review of previous comparative studies	3
2.2	Summary of general protocols for site selection and experimental design	7
2.3	Agronomic and environmental influences on secondary plant metabolites	9
2.4	Experimental design for statistical power and data interpretation	12
3	Materials and Methods	15
3.1	Study area and experimental design	15
3.2	Soil sampling and analyses	18
3.3	Variety selection and transplant procedures	18
3.4	Data collection and field observations	18
3.5	Tomato sampling and nutritional analyses	24
3.6	Statistical analyses	25
4	Results and Discussion	26
5	Summary and Conclusions	39
	References	41
	Photo Gallery	47

Tables

Table 1.	Soil series and types by farm pair	17
Table 2.	Weed management practices by farm pair	19
Table 3.	Fertility management practices by farm pair	20
Table 4.	Pest management practices by farm pair	21
Table 5.	Disease management practices by farm pair	22
Table 6.	Pesticide use information for conventional farms	23
Table 7.	Mean values for tomato quality evaluation by farm pair	26

Table 8.	Mean values for tomato antioxidant and nutritional evaluation by farm pair	27
Table 9.	Mean values for soil chemical characteristics by farm pair	28
Table 10.	Mean comparison of fruit quality parameters by farm type	28
Table 11.	Mean comparison of antioxidant components by farm type	29
Table 12.	Mean comparison of antioxidant components by farm type for Farm Pair B	30
Table 13.	Mean comparison of fruit quality parameters by farm type for Farm Pair B	30
Table 14.	Mean comparison of antioxidant components by farm type for Farm Pair C	30
Table 15.	Mean comparison of fruit quality parameters by farm type for Farm Pair C	31
Table 16.	Mean comparison of antioxidant components by farm type for Farm Pair D	31
Table 17.	Mean comparison of antioxidant components by farm type for Farm Pair E	32

Figure

Figure 1	Location of farm pairs	16
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Executive Summary

Lycopene and β -carotene have been shown to act as powerful antioxidants in humans. A diet containing moderate amounts of lycopene has been associated with the prevention of cardiovascular disease and cancers of the prostate and gastrointestinal tract. Many health experts have recommended increasing levels of dietary lycopene through the consumption of fresh tomatoes and tomato products.

In order to maximize the content of lycopene in tomato, numerous investigations have been conducted to evaluate the influences of genotype, agricultural practices and environment. Specific varieties, deficit irrigation practices, plant nutrient interactions, temperature and sunlight conditions have been identified that enhance lycopene production. In addition, there is growing debate as to whether growing vegetables under organic management systems will lead to higher concentrations of lycopene compared to conventional management systems.

An on-farm comparative trial was conducted on 10 matched pairs of organic and conventional farms in central and southern Taiwan. Management practices and environmental conditions were evaluated for influences on fruit quality and the development of lycopene and other antioxidant compounds in tomato.

Aggregation of farms by type resulted in no significant differences between organic and conventional farming systems for nutritional parameters investigated, which included β -carotene, lycopene, ascorbic acid, total phenolics and antioxidant activity. Organic systems produced tomatoes of higher pH, but there were no significant differences found for soluble solids, acidity or color values between the farming systems.

However, when matched pairs were evaluated as individual case studies, significant differences were found between two pairs of conventional and organic farms for ascorbic acid, total phenolics and lycopene concentrations as well as for pH, soluble solids, and color value. Notable differences, while not significant, were found on two other farm pairs. Results were inconsistent and uniformity in farming system effects on antioxidant compounds was not discovered. Nevertheless, environmental conditions and management practices were identified that were influential in determining outcomes.

This investigation showed that organic farmers with no experience in on-farm trials could successfully conduct horticultural research. It was also determined that high quality tomatoes with increased levels of antioxidant compounds can be grown on organic farms in humid, sub-tropical conditions.

1 Introduction

Tomato (*Lycopersicon esculentum*) is one of the world's major vegetables with 4.4 million ha under production and 115 million t produced worldwide in 2004 (FAOSTAT data, 2004). It is an excellent source of many nutrients and secondary metabolites that are important for human health: folate, potassium, vitamins C and E, flavonoids, chlorophyll, β -carotene and lycopene (Wilcox et al., 2003). Chlorophyll, β -carotene and lycopene are involved in photosynthetic reactions and are produced in plastids with the highest accumulation found in chloroplasts and especially chromoplasts. These color-producing compounds are synthesized by plants and microorganisms but not by animals; chlorophyll produces a green pigment and the carotenoids, lycopene and β -carotene, produce the familiar red and yellow colors associated with tomatoes. Carotenoids function as photoprotectants in that they have the ability to neutralize harmful byproducts of photooxidation (Bartley and Scolnik, 1995; Clinton, 1998).

Lycopene and β -carotene have been shown to act as powerful antioxidants in humans. A diet containing moderate amounts of lycopene has been associated with the prevention of cardiovascular disease and cancers of the prostate and gastrointestinal tract (Gann et al., 1999; Agarwal and Rao, 2000). Increasing levels of dietary lycopene through the consumption of fresh tomatoes and tomato products has been recommended by many health experts (Tonucci et al., 1995; Giovannucci, 1999).

In order to maximize the content of this important phytonutrient, the influences of genetics, agricultural practices and environment have been investigated. Studies have found consistent differences in lycopene concentrations between tomato varieties, which can be magnified by environmental conditions and agricultural practices, especially those affecting plant nutrient status (Abushita et al., 2000; Binoy et al., 2004). A relationship has been established associating temperature and light intensity with lycopene destruction and accumulation. Precursors of lycopene are inhibited and production of lycopene is stopped at temperatures below 12 °C and above 32 °C. Temperatures ranging from 22–25 °C give the most favorable rate of lycopene production, which is further enhanced by sunlight.

Specific agricultural management practices associated with lycopene development have been investigated but results are not always conclusive. Dumas et al. (2002) reviewed several studies that looked at agronomic and environmental factors that influenced lycopene concentrations in tomatoes. Moisture stress, for example, reduced lycopene content in some tomato varieties but increased it as well as β -carotene content in others. Nitrogen (N) and phosphorus (P) fertilizers have been shown to increase lycopene concentrations but results were inconsistent and dependent on seasonal variations in climate (Bruulsema et al., 2004). A controlled environment study in which potassium (K) fertilizer was applied to tomatoes grown in pots filled with quartzite-silica sand obtained more reliable results in terms of lycopene response (Trudell and Ozbun, 1971); however, it is unclear if the same results would be obtained in open fields where soils and environmental conditions are variable.

Dumas et al. (2002) concluded it would be very difficult to define optimal growing conditions for lycopene development and storage in tomato fruits given the information available. They recommended more investigations be made as to how the combination of cultural practices such as water management and soil fertility influence, for example, plant canopy development, which mediates heat and temperature regimes during fruit formation. Furthermore, biosynthesis of carotenoids depends on the presence of certain enzymes in the plant as well as essential precursors, phytoene and phytofluene (Bartley and Scolnik, 1995). Mineral elements in the soil act as co-factors in these metabolic reactions (Trudell and Ozbun, 1970) and the quantity and availability of these plant nutrients will influence the rate and amount of secondary plant metabolites synthesized (Grusak et al., 1999; Brandt and Molgaard, 2001).

There is growing debate as to whether growing vegetables and fruit crops under organic management systems will lead to higher concentrations of lycopene and other secondary metabolites compared to growing these crops under conventional systems (Woese et al., 1997; Brandt and Molgaard, 2001). A comparative study investigating antioxidant activity and phenolics in strawberries found no consistent differences between these two management types (Hakkinen and Torronen, 2000). A more recent study found significantly higher levels of lycopene and other microconstituents in tomatoes grown on an organic farm versus a conventional research station when tomatoes were evaluated on a fresh matter content basis (Caris-Veyrat et al., 2004). However, in both of these studies the authors stated that results needed to be verified over additional years of replicated research.

When comparing the effects of organic and conventional farming systems on the development of secondary plant metabolites, the comparison of fertilization and irrigation practices and their influences on nutrient transport are important areas for further examination. Organic and conventional farmers generally use different methods for managing soil fertility. Most organic farmers grow green manures and apply composts and animal manures for satisfying plant nutrition. These practices supply nutrients that are initially unavailable to plants until mineralized by soil microorganisms. By comparison, conventional farmers satisfy crop demands by applying inorganic mineral fertilizers that are readily soluble and immediately available to plants. Evaluating the effect of these different management practices and identifying the presence or absence of interaction among different variables on the development of secondary plant metabolites may prove to be as valuable as investigating the influences of pest and disease control practices (Stout et al., 1998; Hakkinen and Torronen, 2000; Asami et al., 2003; Stamp, 2003).

The objectives of this study were to 1) develop a protocol for the selection of organic and conventional farm pairs to be used in comparative trials for determining the effects of management practices on the development of the secondary plant metabolites; and 2) to compare lycopene, β -carotene and other antioxidant concentrations between tomatoes grown under conventional production practices with those grown under organic practices.

2 Protocols for Site Selection of Farm Pairs for Comparative Studies

2.1 A review of previous comparative studies

Comparative studies between organic and conventional farming systems have been conducted for many years. Various methodologies have been used to investigate differences in soil properties, plant yields, nutrients and secondary plant metabolites. Trials were either located at research institutions or on commercial farms. At the Delaware Agricultural Experiment Station, Svec et al. (1976) evaluated the mineral and ascorbic acid content of vegetables grown on garden plots in which ‘organic’ and ‘conventional’ treatments had been established in a field with known cropping history and single soil type. Experimental plots were separated by 1.83 m and the same varieties were used for comparison.

One of the earliest on-farm trials was conducted by Lockeretz et al. (1980). Maize yields and soil nutrient levels were compared on 26 pairs of neighboring commercial farms located in five states in the midwestern United States. Criteria for selection of organic farms were established for fertilizers used, farm enterprise type (mixed grain-livestock), minimum number of years under organic management (4 years), minimum field size (40 ha), soil characterization (USDA mapping), and proximity to a conventional neighbor with a maize field of matching soil type willing to cooperate in the trial. For this study, cropping history was the same for both organic and conventional farms for 13 of the 26 matched pairs. Within a farm pair, in addition to controlling for soil type and location, variety and planting date were also matched. A subset of the same farms was used in a comparative trial the following year assessing crude protein and amino acid composition in maize. Again fields were matched for location, soil type, variety and planting date (Wolfson and Shearer, 1981).

These early studies established methodologies for experiment station trials as well as protocols for site selection of commercial farms to be used in comparative studies. Paired farms and experimental plots needed to be in close proximity, have the same soil type, have a similar cropping history and farm enterprise classification, the same variety, and synchronized planting and harvesting dates, if crop constituents and yields were under consideration. Later, researchers added more stringent requirements for farm proximity and cropping history.

Reganold (1995), in a review summarizing biodynamic and conventional on-farm comparative trials, described additional desirable criteria for selection of farm pairs for comparing effects of agricultural systems on soils. Farms were required to satisfy three conditions: 1) be adjacent to one another but under different management systems currently while previously under the same management system; 2) have experimental fields located side-by-side so that all soil-forming factors as described by Jenny (1941) (climate, natural vegetation, relief, parent material, and time), except management practices,

are similar; and 3) have allowed sufficient time to pass for different management systems to impact soil properties. These criteria added to earlier protocols for site selection were used in paired soil surveys between different farming systems in many studies (Reganold, 1988; Reganold et al., 1993; Nguyen et al., 1995; Reganold and Palmer, 1995; Gerhardt, 1997); however, other comparative studies found it was not always possible to satisfy all of the site selection criteria and adjusted experimental designs to compensate.

Locating farm pairs such that experimental fields are side-by-side and management histories are comparable proved difficult to achieve. Munro et al. (2002) only matched environmental conditions and soil types when comparing the sustainability of topsoils on 14 pairs of farms in England. Liebig and Doran (1999) only met criteria for proximity and soil type when investigating the impact of organic production practices on soil quality indicators on five paired farms in Nebraska and North Dakota. In this latter study, while agricultural production systems were considered the basis of comparison, they were not compared in aggregate due to differences in climate and topography between sites; comparisons were only made between farm pairs of the same soil type.

Others found that matching soil types on farm pairs was also problematic. Armstrong-Brown et al. (2000) surveyed topsoil characteristics in a comparative study of 30 farm pairs conducted in England. Fields with common boundaries were sampled wherever possible on farm pairs in order to increase the probability that identical soil types would be found. The inability to match soil types was overcome by using a large number of farm pairs so that farming systems were adequately represented and variability due to soil type was minimized. Matching farms according to farm enterprise type (horticultural, arable, permanent pasture) was an important criterion for making valid comparisons in this survey.

Mercadante and Rodriguez-Amaya (1991) only controlled for farm type ('natural' versus conventional) and proximity (neighboring) in order to achieve environmental parity when selecting two commercial farms for a comparison of carotenoid composition in kale in Brazil. Soil types were not intentionally matched but may have been inadvertently similar due to the proximity of the two farms.

When evaluating perennial crops, a different experimental methodology was taken by Hakkinen and Torronen (2000). A comparison was made in the content of flavonols and four different phenolic acids in three strawberry varieties grown organically versus conventionally in Finland. Commercial farmers were chosen from the same geographic area, varieties were matched, and berry samples from farms were pooled for analyses. No reference was made to matching farms based on soil type. Berries were collected from bushes that were either one or two years old.

Combining the experimental methodology used for simulated 'organic' and 'conventional' experiment station plots with on-farm research, Goh et al. (2000) conducted a comparison of farming system effects on orchard soil organic matter content and sustainability. In this study, an apple orchard with organic, conventional, and integrated

treatments was established at a university horticultural research area. Management practices for the different treatments were determined by following practices established by the appropriate industry guidelines. Results from the experimental plots were compared with nearby organic, conventional and integrated commercial farms. Soil texture was the same for all orchards but two different soil series were found in treatments located in the experimental plots. The apple variety was the same. Trees in commercial orchards were between 7–9 years old while experimental orchards were approximately 4 years old.

Instead of surveying representative organic and conventional paired farms, other comparative studies used single commercial farms that had both organic and conventional production systems in operation (Asami et al., 2003; Lombardi-Boccia et al., 2004). These studies investigated nutritional differences in crops, measuring concentrations of vitamins and secondary plant metabolites.

Using a commercial farm in which three farming systems were in operation, Asami et al. (2003) compared total phenolic and ascorbic acid contents in strawberries, marionberries and corn produced using organic, conventional and sustainable agricultural practices. Experimental plots were not adjacent to each other and soil types, as stated in the study, varied among treatments for corn and were not recorded for marionberry. Crops were evaluated over one season only. It should be noted that this study was subsequently criticized for having significant technical flaws in its methodology by Felsot and Rosen (2004). They regarded the experimental design as being unrepresentative of conventional and sustainable systems and specifically mentioned the use of fields that were not well matched with regard to soil type, location, and years under their respective management systems (conventional marionberries were 21–22 years, organic 4 years, and sustainable 2 years).

On the other hand, Lombardi-Boccia et al. (2004) applied a similar methodology as Asami et al. (2003) and chose a commercial fruit farming institute on which to conduct a comparison of nutrients and antioxidant compounds in organically and conventionally grown yellow plums. Organic fields were established 600 m away from conventional fields and isolated by a hedge. In this case, three different organic soil management practices represented organic treatments with one matched to conventional plot conditions; comparisons were made within organic treatments as well as between matched organic and conventional. The trial was carried out over three years; all treatments had the same amount of time to influence soil properties. No information was given on investigation of soil type; however, it was suggested that because experimental fields were located on the same farm, variability in plums due to soil effects could be excluded.

In a comparative study of mineral concentrations and yields in wheat, Ryan et al. (2004) met all the established protocols for on-farm trial site selection and matched farm proximity, environmental conditions (aspect), soil type, number of years in operation and farm enterprise classification. In the end, the results they obtained did not allow them to conclude that organic farms in general produced grains with higher mineral

contents. They noted that the increasing use of lime fertilizer to neutralize acidic soils by organic farmers was an important factor when interpreting their results; since this is also a common practice with conventional farmers they could not consider it a farming system effect. This study included a discussion of the lack of scientifically rigorous comparative studies between organic and conventional farming systems. Variability within and between organic and conventional farms and the differing environmental conditions in which they are located often contributed to inconclusive findings. Even with well-matched farm pairs, results are still difficult to interpret and sometimes inconclusive.

Other scientists expressed similar frustrations as Ryan et al. (2004). After reviewing contradictory and inconclusive results produced from on-farm comparative trials, and citing the interaction of often uncontrollable numbers of variables, many researchers have decided that growing conditions and management practices need to be standardized in order to obtain sound scientific data. Instead of conducting comparative studies of whole systems on commercial organic and conventional farms, scientists went back to establishing simulated 'organic' and 'conventional' conditions in plots at agricultural research centers as was done by Svec et al. (1976).

Warman and Havard (1997, 1998) used this approach in a two-year study of carrot, cabbage, potato, and sweet corn in which yields, vitamins, and minerals were compared. They converted a fallow field where no synthetic chemicals had been applied for three years into 'organic' and 'conventional' treatments.

The results of this trial were cited as representative of organic and conventional farming systems in another study in which the authors also expressed concern for more well-controlled and defined research for comparing management system effects on vegetable quality (Wszelaki et al., 2005). In this study, researchers established neighboring 'organic' and 'conventional' fields, although experimental plots were not side-by-side, at the Ohio Agricultural Research and Development Center. Potatoes were evaluated for sensory quality, mineral and glycoalkaloid concentrations with two organic treatments being compared to one conventional. Fließbach and Mäder (2000) also used this methodology in a long-term comparison of biodynamic, organic, and conventional farming systems conducted in Switzerland. It has been argued whether these simulated farms represent a true comparison of organic and conventional whole farm systems or are just an abstraction. Conditions and knowledge found at an agricultural research institute may not adequately represent site conditions and management practices found on commercial farms.

In a paper promoting the necessity for on-farm research, Andersen (1992) described three types of problems that can only be investigated on commercial farms because of the conditions they require: 1) problems that require a specific management history, physical conditions or natural phenomena; 2) problems that investigate farm management itself; and 3) problems that require the integration of whole farm dynamics such as crop rotations and compost production from available feedstocks. The biological environment of the farm has a significant influence and should be considered when investigating effects of farming systems. This is often not factored into results when

conducting comparisons using simulated organic and conventional cropping systems established at research institutes.

Finally in this review of comparative trials, a study evaluating tomatoes and derived purees grown ‘conventionally’ versus organically is included due to its unique experimental methodology. Caris-Veyrat et al. (2004) applied a combination of on-farm trial research with simulated farming conditions established at a research institute. A nearby commercial organic farm (within 1 km) was matched with a simulated ‘conventional’ plot established at the Balandran Applied Research Center of Centre Technique Interprofessionnel des Fruits et Légumes in France. Three tomato varieties were evaluated for contents of microconstituents (lycopene, β -carotene, vitamin C, chlorogenic acid, rutin and naringenin). The authors also measured human blood plasma levels of β -carotene and lycopene after supplementation with tomato puree derived from both production systems. Tomatoes were grown under plastic tunnels, and factors such as fertilizer applications, irrigation practices and mechanical weed control were standardized. Soil analysis was conducted after fertilization for comparison of soil properties, but no information regarding soil type for either location was provided. Environmental conditions at the organic farm were said to be similar to those at the research station’s conventional plot.

What is noteworthy about this study is that the experimental methodology combined an investigation of the effects of production systems on microconstituent contents of tomatoes with a test of how the human body metabolized products from the two systems. This approach has been suggested as being a more definitive comparison of farming systems. Advocates of this approach believe that it is not enough to provide information on food composition between organic and conventional farms; rather, researchers should go beyond the fields and conduct human and animal feeding trials to reveal how these foods are digested and metabolized to discover the full health effects (Worthington, 1998; Bourn and Prescott, 2002; Grinder-Pedersen et al., 2003).

After reviewing the previous comparative trials, whether using paired commercial farms or simulated ‘organic’ and ‘conventional’ plots established at experiment stations, general protocols for site selection and experimental design can be extracted and are described in the next section.

2.2 Summary of general protocols for site selection and experimental design

When conducting a comparative study between organic and conventional farming systems the following basic criteria for site selection and experimental methodology should be considered: 1) participating farms (or research plots) must be *representative* of organic and conventional farming systems; 2) paired farms must be in *close proximity*; 3) *soil type* should be the same; 4) different production systems should be in operation long enough for management practices to have had *time* to affect soil properties; 5) farm *enterprise classification* should be matched; 6) *cropping histories* should at least be

compared if not matched; 7) *planting and harvesting dates* should be synchronized; and 8) *varieties* should be matched.

For on-farm trials, care must be taken in the initial selection of participants such that farms are truly *representative* of farming systems to be compared and the resulting paired farms are considered characteristic of organic and conventional farms found in a particular region of interest. Most paired trials rely on accredited certifying agencies, local university agricultural extension units, or surveys for guidance in locating and selecting suitable candidates for participation. The limitations to this approach are that only farms located in areas where organic farming is well developed will be available for participation in comparative trials. Experiment station trials using simulated 'organic' and 'conventional' plots should apply production practices recommended by industry guidelines established for respective farming systems.

Farms should be in *close proximity* to one another, thereby assuring that uncontrollable environmental factors related to climate and topography are the same and parameters of interest (crops and soils) are exposed to similar temperature and rainfall patterns.

Soil type can be an important consideration due to influences of soil morphology and texture on soil and plant development, and attempts should be made to match *soil types* within farm pairs deliberately, if information is available (pretrial site sampling or review of published soil survey maps) or by selecting neighboring farms with the expectation that soil types will be similar. Not all comparative trials reviewed above were able to satisfy this condition and soil type effects can vary. Patil et al. (1995) concluded that influences of location and genotype overrode soil type effects when measuring quercetin concentrations in onions. On the other hand, Lester and Eischen (1996) found soil type differences to be significant when measuring quantities of carotenoids in melons grown on fine sandy loam versus silty clay loam. While matching for soil type, controlling for soil fertility status was not addressed. Studies reviewed did not undertake extensive testing of soil properties in order to match underlying nutrient status within paired farms.

After considering soil type, farm history should be established (e.g., number of years certified for organic farms) indicating sufficient *time* has passed for respective farming systems to have had an impact on soil properties and develop steady-state conditions.

In addition to the above criteria, farm *enterprise classification* is matched while *cropping histories* are compared but not usually matched due to the tendency for cropping sequences to be different in organic and conventional farming systems. Finally it is important to synchronize *planting and harvesting dates* and choose the same *varieties* when comparing crops so that differences related to crop maturity and genotype are avoided.

Selecting the appropriate venue for comparing organic and conventional production systems is primarily a matter of variability and control. Conducting comparative trials on commercial farms using farmer operators versus conducting comparative trials un-

der simulated ‘organic’ and ‘conventional’ conditions at research institutions managed by scientists obtains differing degrees of control over site selection parameters, experimental design and error, level of variability, data collection, overall project control and presumably quality of results. On-farm trials managed by farmers are entirely dependent upon the interest and abilities of the participants, and outcomes will reflect levels of commitment and expertise. Electing to conduct a comparative trial of different farming systems in simulated fields at research institutions produces far less variability and greater control, but possibly at the expense of authenticity such that results are not valid for the farming systems represented due to oversimplification of relevant factors.

When judging the relative merits between studies that compare organic and conventional farming systems using commercial farms versus those conducted using experiment station plots, one must take into consideration the importance of ecosystem effects. The biological environment that develops on commercial farms as farming systems establish themselves over time cannot be duplicated in simulated plots at research institutions. Findings from comparative studies would be more representative of farming system effects if the study is initially conducted on paired commercial farms with farmers responsible for all decisions regarding production. As was done by Lockeretz et al. (1980), researchers should determine field location, plot size and experimental design and then allow the farmer to produce the crop under consideration without further interference. Subsequent verification and further elucidation of effects could be done by employing factorial experiments at research institutes once specific farm management practices have been identified as being significant.

The above described criteria for site selection and experimental design are the first step in conducting a comparative study between organic and conventional farming systems. Once sites are selected, the following additional considerations need to be incorporated into the experimental design for evaluating farming system effects on secondary plant metabolites, especially β -carotene and lycopene.

2.3 Agronomic and environmental influences on secondary plant metabolites

Agronomic and environmental factors known to influence the development of tomato carotenoids, i.e., lycopene and β -carotene, were identified in previously described studies (Abushita et al., 2000; Dumas et al., 2002; and Binoy et al., 2004). Comparative trials investigating farming system effects on secondary metabolites should give additional consideration to the following: *variety* selection for maximizing lycopene production and minimizing genotype variation, *sunlight* availability and corresponding temperature/heat influences; *planting and harvesting dates*; *irrigation practices* and drainage conditions; and *soil fertility management practices* affecting plant nutrients found to influence carotenoid concentrations.

There are distinct differences between tomato genotypes and lycopene development. Selecting *varieties* that are bred to produce increased levels of lycopene is recommended so that measurable levels of compounds are more likely to be produced.

Once a suitable variety is chosen both farms must be matched for varieties and associated physical cultural practices. For example, indeterminate varieties that require staking need to be supported in the same manner on matched pairs. The decision as to whether tomatoes will be grown in open fields or under protective structures such as greenhouses or plastic covers should be made with the understanding that these conditions have to be standardized within farm pairs as they will influence both light intensity and temperature.

Carotenoid development is highly influenced by *sunlight*. Farms with sloped fields should be avoided as this creates uneven field conditions with regard to sunlight incidence on plants. Tomatoes growing in shaded areas of plants could develop lower lycopene concentrations; more exposed tomato fruits will experience higher temperatures than those less exposed to sunlight leading to further variation in conditions affecting lycopene development (McCollum, 1946). Therefore, experimental fields should have similar aspect and sun exposure so that plants experience the same degree of illumination and hours of daylight. Farm structures or natural features that might shade plots also need to be noted for their influences on microclimate conditions in the field. Degree of foliation and plant canopy characteristics will affect the developing tomato fruit's ability to conduct photosynthesis as well as protect from overexposure to heat and light. Observations need to be made as to the degree of difference between farming systems in overall development of tomato plant structure (Clark and Merrow, 1979).

Planting and harvesting dates need to be synchronized as stage of maturity will greatly influence lycopene concentrations. Sampling protocols should be standardized with tomatoes harvested at the fully red-ripe stage for maximum lycopene production.

Irrigation practices and other factors related to drainage conditions and water source contamination should be monitored for potential impacts on soil moisture content, plant moisture uptake and soil solution salinity. Mitchell et al. (1991) observed fruit quality in tomatoes improved, as measured by increased soluble solids and fruit acidity, when plants experienced drought-like conditions imposed by deficit irrigation. This was attributed to a concentration effect when fruit water content was reduced accompanying decreased water consumption by plants. Petersen et al. (1998) found tomato quality improved and β -carotene content increased as irrigation water increased in salinity. De Pascale et al. (2001) also found yield and size reductions and significant increases in fruit quality parameters and lycopene content when tomatoes were irrigated with moderately saline solutions combined with different N fertilizer treatments. Water quality and quantity may impact yields, soluble solids, and possibly carotenoid content in tomatoes. Therefore, irrigation practices and field conditions that may affect the movement of water through experimental plots should be determined so that these effects can be accounted for.

Detailed observations of *soil fertility* practices and plant nutrient status, especially for those minerals known to impact lycopene concentrations—N, P, K and magnesium (Mg)—should be carried out so that appropriate methodology for measuring their effects can be developed. Experimental designs need to be capable of capturing and

describing how nutrients are delivered to growing plants. Soil samples should be taken in order to determine baseline nutrient status at critical points during fruit development (e.g., fruit set, green fruit stage and harvest). Results from soil analyses can be correlated with nutrients measured in fruit and leaf tissue samples in order to determine whether soils are delivering adequate supplies of nutrients to growing plants. These results can be further correlated with results from antioxidant analyses.

With farm enterprise classification and soil types matched, a similar level of soil fertility might be assumed if both organic and conventional farmers had applied amendments with equal nutrient concentrations and maintained similar cropping sequences. However, soil fertility practices and crop rotations are two of the distinguishing differences between organic and conventional farming systems, subsequently producing different soil physical and chemical characteristics over time (Clark et al., 1998). In fact, significant differences have been found for measurable soil properties such as soil bulk density, available water-holding capacity, penetration resistance, topsoil thickness, cation exchange capacity, electrical conductivity, soil pH, nitrate-N, total N, available P, exchangeable K and sulfur (S), organic matter content and organic carbon (C) (Lockeretz et al., 1980; Reganold et al., 1993; Liebig and Doran, 1999; Armstrong-Brown et al., 2000; Condrón et al., 2000; Munro et al., 2002). Organic farms typically have lower levels of most available plant macronutrients, especially inorganic nitrogen. Organic carbon resources, on the other hand, are usually higher than those found on conventional farms.

A more fundamental understanding of the importance that plant nutrient status, especially C and N, has on the development of carotenoids can be found in ecological theories (e.g., Carbon/Nitrogen Balance and Growth/Differentiation Balance) predicting levels of secondary metabolites based on availability of plant nutrients and conditions that enhance disease resistance. According to these theories, plants will selectively produce compounds for growth or differentiation relative to the abundance of C and N resources (Stamp, 2003). Nitrogen-containing compounds will be favored over C-based secondary compounds when N is readily available and not limiting for growth. Carbon-based compounds, such as lycopene and β -carotene, will be produced in N-limiting conditions when photosynthetic activity is not simultaneously reduced (Stout et al., 1998). Decreased levels of nitrogen have been positively correlated with production of plant defense compounds such as phenolics and flavonols (Norbaek et al., 2003). Furthermore, N-limiting conditions have been found to promote general resistance to disease as well as prolong the effectiveness of disease-resistant varieties (Pedersen and Bertelsen, 1999).

According to the above ecological theories these conditions—higher C, lower N—are more favorable for the production of C-based secondary plant metabolites, such as lycopene (Brandt and Molgaard, 2001). Overall, organic farms may be expected to produce higher levels of these compounds due to farming practices and management decisions that reduce production intensity and create an environment that enhances disease and pest resistance in conjunction with increased use of resistant varieties.

However, C and N are not the only plant nutrients impacting the production of compounds such as lycopene, and the availability of these compounds should be evaluated relative to the availability of other important plant nutrients such as P and K, which have also been found to influence secondary metabolite production. These plant nutrients are not addressed in the Carbon/Nitrogen Balance and Growth/Differentiation Balance theories.

This section described environmental and agronomic influences that can be used to distinguish farming systems' effects on the production of secondary plant metabolites. Once these factors have been identified, designing the appropriate experimental methodology and applying advanced statistical tools can assist in separating the most relevant influences from those that have less significance or are less easily determined.

2.4 Experimental design for statistical power and data interpretation

On-farm trials often contain large variation in farm physical conditions and farmer management practices. Such heterogeneity can mask true treatment effects and consequently produce nonsignificant statistical test results. Since it is frequently not possible to limit variability and match farm pairs for all important nontreatment factors, this disadvantage can be addressed through increasing plot size and number of farm pairs making statistical tests more powerful in detecting actual treatment effects. However, the decision for determining number of matched pairs of farms must be made not only with consideration given to maximizing statistical sensitivity and minimizing variability, but also recognizing constraints of available resources and manageability.

Comparative on-farm trials that rely on few matched pairs for comparisons of technology performance are more manageable with less between-farm variability, but are only indicative of what is possible given particular site conditions. In this case, general conclusions regarding farming system effects cannot be made. On the other hand, if the decision is made to include numerous farm pairs, it should be done only if all pairs have reasonably similar environmental conditions when results from similar treatments (i.e., production systems) are to be aggregated and comparisons made between farm pairs.

To gain a broader perspective of how secondary plant metabolites are affected by differing locations, the experimental design could include farm pairs with similar soil types located across a variety of environmental landscapes. This approach would produce an assessment of ecoregion effects. For example, Gardner and Clancy (1996) evaluated the impact of farm management practices on soil quality indicators in North Dakota. Conventional, no-till and organic production systems were compared; farms representing each type were selected in three dominant eco-regions found in the state. Comparisons between management systems were made within regions.

Once matched pairs of farms are selected, attention shifts to field level considerations. Experimental plots can be set up using the same designs that research institutions use. Often a randomized complete block design (RCBD) is recommended; however,

other conditions in the field should be left alone, as if no trial were being conducted. Plots are embedded in farmers' fields and managed by farmers in the farmers' own way. This experimental design gives less control over non-treatment factors and produces a large amount of variability, for which on-farm trials have been criticized. It also makes data analysis and interpretation more difficult.

However, for the on-farm comparative trial to be truly representative of farming systems, some conditions of uniformity must be abandoned by the researcher. The physical conditions of the farm, including farmer management, should not be interfered with. By not interfering in a farmer's practices, one gains a more realistic assessment of parameters of interest under real farm conditions. To impose uniformity for soil fertility maintenance, for example, might be interfering in a farmer's usual practices to such an extent that the technology being tested would be under artificial conditions and effects would not be those from genuine farmer's management practices. In this situation, control over variables that are not related to the treatment is replaced with greater observations of farmers' actual practices. The risk is that variability due to differences in soil properties (e.g., fertility) becomes larger than treatment differences and real treatment effects cannot be detected. However, this variability should be analyzed and explained rather than artificially controlled.

One method for reducing variability is to increase the number of replications, or plot size, used with on-farm trials so that more information is gathered that characterizes site conditions and management effects. Data are collected and analyzed using standard analysis of variance (ANOVA) and comparison of means. More sophisticated statistical techniques are available that can further distinguish the effects from variable management practices between farmers.

Multivariate analysis of variance (MANOVA) can be performed for soil properties, for example, if characteristics are measured in the same way as other parameters on a per replication basis. In this way, MANOVA can test for differences between organic and conventional farms based on antioxidant/fruit quality and soil parameters, for example, simultaneously.

Applying multivariate analysis allows greater differentiation among factors such that true separation of farming systems can be achieved. Gundersen et al. (2000) applied principle component analysis (PCA) to the results obtained from measuring major and trace elements in onions and peas collected from organic and conventional farms. Onions were analyzed for 63 elements and 55 elements were quantified in peas. Farms were uniform with respect to varieties grown and geographic region. PCA was applied to the results in order to identify parameters that explained most of the variability among farms; organic and conventional farms were split into two groups according to farming method.

Once statistical analyses are performed, interpretation of results must be done with care so that conclusions made represent farming system effects. This was illustrated by Ryan et al. (2004) with results obtained from comparative trials on wheat farms as de-

scribed in Section 2.1. They argued that statements often made regarding the differences between organic and conventional farms were done without sufficient investigation of causative processes. Researchers must make distinctions between farming system effects and effects that are caused by the choices individual farmers make as illustrated by the application of particular amendments and their effects on crop constituents.

Ryan et al. (2004) cited the results of an organic versus conventional comparative vegetable trial conducted in Japan (Ren et al., 2001) as a case in which results may have been misinterpreted leading to false statements. In this study, organic greens were reported to have significantly higher levels of antioxidant activity as compared to conventional. Ren et al. (2001) concluded that organic farm management practices were responsible for the results, but Ryan et al. (2004), based on their experience with wheat farmers, concluded that the input of chitosan (deacetylated chitin from crustacean shells) to the organic crops was what induced the increase in antioxidant activity and not the organic farming system itself. The amendment chitosan is not exclusive to organic farmers and could also have been used by conventional farmers.

These kinds of considerations should be incorporated into the experimental design for comparative studies such that effects not related to production systems are more easily identified. It is an important distinction to make as more and more conventional farmers are adopting practices that would have been considered typical organic production practices in the past.

In summary, this chapter described general protocols for site selection, experimental design, data analysis and interpretation of results for comparative studies of organic and conventional farming systems investigating secondary plant metabolites. These protocols are now applied to the on-farm comparative trial described as the second objective of this study—an investigation of the effects of farm management practices on tomato fruit quality and nutritional compounds produced on organic and conventional farms.

3 Materials and Methods

3.1 Study area and experimental design

The research was conducted on 10 farms in southern and central Taiwan during the winter of 2004–2005. In cooperation with representatives from the Taiwan District Agricultural Improvement Stations (DAIS) in Kaohsiung, Tainan and Taichung, the Taichung Farmer's Association, as well as sustainable farming associations involved in organic agriculture, five organic farms were identified and matched with five nearby conventional farms such that soil type, environmental conditions, and other relevant factors (except management practices), were similar for each pair of organic/conventional fields wherever possible.

Organic farms were selected based on two sources of recommendation: 1) approval by an accredited Taiwanese organic certifying agency based on each farm's record of compliance with certification requirements; and 2) approval by the local agricultural research station scientists conducting research into organic production. Conventional farm participants were selected based on a demonstrated ability to collaborate in on-farm research as well as proficiency in production of tomatoes. Farmers selected for participation in the project were found in Shanhua, Shinhua, Madou, Sihua, and Puli and are herein labeled farm pairs A, B, C, D, and E, respectively (Figure 1).

As described in Section 2.1, comparative studies often use soil type and environmental conditions as criteria for matching organic and conventional farm pairs. In this study, organic and conventional farms were selected that were in close proximity, thereby matching environmental conditions for temperature, precipitation and slope. Information on soil types was not readily available at the time farm pairs were chosen and this criterion could not be applied in the selection process. In order to identify soil type, a standard handheld global positioning system (GPS) unit was used to obtain longitude and latitude coordinates. Farm geographic locations were later identified on land use maps provided by the Taiwan Agriculture Research Institute produced from soil surveys conducted from 1963 to 1976 by the National Chung Hsing University Soils Department and other geologic institutions in Taiwan. Results were reviewed and soil series and soil types obtained for all farms (Table 1). This data showed that a criterion for selection of matching farm pairs, i.e., soil type, was satisfied for only Farm Pairs A and C.



Figure 1. Location of farm pairs

Table 1. Soil series and types by farm pair

Farm pair	Farm type	Longitude/ latitude	Soil series	Soil type	Soil characteristics
A	Organic	23°09.42N 120°18.76E	Shan-shang	Loam	Aged alluvial soil, fine texture, good drainage
	Conventional	23°09.43N 120°18.92E	Shan-shang	Loam	Aged alluvial soil, fine texture, good drainage
B	Organic	23°02.93N 120°17.85E	Tainan	Loam	Aged alluvial soil, medium texture, good drainage
	Conventional	23°01.47N 120°17.40E	Yen-chen	Extremely fine sand loam	Low terrace incalcareous alluvial soil, medium texture, good drainage
C	Organic	23°10.90N 120°12.78E	An-nei	Silt loam	Lowlands calcareous alluvial soil, medium texture, good drainage
	Conventional	23°11.15N 120°12.56E	An-nei	Silt loam	Lowlands calcareous alluvial soil, medium texture, good drainage
D	Organic	23°57.81N 120°29.38E	Erhlin	Loam	Lowlands calcareous alluvial soil, medium texture, incomplete drainage
	Conventional	23°57.43N 120°29.60E	Erhlin	Silt loam	Lowlands calcareous alluvial soil, medium texture, incomplete drainage
E	Organic	23°58.76N 120°59.22E	Shashuipu	Loam	Slate recent alluvial soils, natural drainage well to moderately well
	Conventional	24°00.03N 120°57.75E	Tingfanpo	Silt loam	Sandstone shale and slate alluvial soils, natural drainage well to moderately well

3.2 Soil sampling and analyses

Prior to transplanting, soil samples were collected from all 10 farms. Farmers were instructed not to apply any amendments to the experimental plots prior to sampling. Plots were divided into three equal segments. Using a soil auger, 10 subsamples were collected from a 0–20 cm depth using a randomized sampling pattern. Subsamples were mixed and a composite was made from each section so that baseline properties could be determined for all fields. The following parameters were analyzed: nitrate (NO_3); ammonium (NH_4); total inorganic N; total N; organic C; percent organic matter; available levels of P, K, Mg, calcium (Ca), sodium (Na), iron (Fe), manganese (Mn), copper (Cu), and zinc (Zn); electrical conductivity; pH; bulk density; and soil texture. Standard analytical methods were employed (Page et al., 1982).

3.3 Variety selection and transplant procedures

Two determinate processing tomato varieties with geminivirus resistance, PT 4769 (labeled V1) and PT 4762 (V2), were used. Both varieties had similar maturity and cultural requirements and were identified as superior to standard varieties for lycopene production. To enclose the experimental plot and provide separation between V1 and V2, a third tomato variety with similar geminivirus resistance and cultural habits, FMTT 848, was used.

Due to the difficulties with fungal diseases encountered growing the transplants under organic conditions, AVRDC produced tomato seedlings for both conventional and organic farms using conventional methods. Seedlings were grown in standard tomato transplanting media of 3 parts peat moss to 1 part vermiculite. Although no pesticides were used, a standard seedling pre-emergence and post-emergence synthetic fertilizer solution was applied. Seedlings were kept in plug trays for 6 weeks until ready for transplanting.

Seedlings were transplanted on matched farms on the same day: Shinhua and Madou on 18 October 2004; Puli and Sihua on 19 October; and Shanhua on 20 October. Raised beds had been prepared in advance by each cooperating farmer. The experimental design was RCBD with three replications. Plants were set in single rows spaced 40 cm apart with 50 cm between beds. Plot size was 21.6 m \times 7 m.

3.4 Data collection and field observations

From October until harvest, experimental plots were monitored weekly. In addition to field observations, farmers were provided with notebooks in which all management practices used in the experimental plot were recorded (Miller et al., 1992).

Typhoon season in Taiwan normally occurs from September through October and is over when winter season begins in November; however, in early December, Taiwan experienced a strong 100-year typhoon, Nanmadol. It impacted southern Taiwan more severely than other parts of the island. Farm Pair A and Organic Farm D suffered flood-

ing damage. Growing conditions remained normal for the remainder of the growing season until harvest.

Tables 2 through 6 summarize information collected from the participating farmers on cultural practices used within the experimental plots for control of weeds, soil fertility, pests, and diseases. In general, weeds were managed using synthetic mulches or hand weeding. Organic farmers applied purchased and homemade compost products made mainly from non-animal feedstocks due to concerns over heavy metal contamination (livestock in Taiwan, particularly pigs, are exposed to heavy metals from contaminated feed). Two conventional farmers also applied compost products, one made from pig and chicken manure, to experimental plots in addition to synthetic inorganic fertilizers. Some organic farmers applied mycorrhizal fungi to assist in the mobilization of soil nutrients. Some conventional farmers applied a root stimulating compound, indolebutyric acid, to enhance the establishment of transplants.

Organic farmers controlled insect pests with minimal spraying of botanical insecticides such as citronella oil and applications of beneficial bacteria such as *Bacillus thuringiensis* (*Bt*). Conventional farmers relied heavily on broad spectrum insecticides such as deltamethrin, a pyrethroid, and chlorpyrifos, an organophosphate. There were also cases of conventional farmers using pheromone traps for control of tomato fruitworm and hornworm and *Bt* for controlling whitefly. Organic farmers controlled diseases by repelling and trapping the insects that transmitted them, such as thrips, through the use of reflective silver mulches and blue and yellow sticky traps. They also employed various foliar sprays for prevention and control (see Tables 4 and 5 for details).

Table 2. Weed management practices by farm pair

Farm pair	Farm type	Weed management practice
A	Organic	Hand weeding
	Conventional	Hand weeding
B	Organic	Silver plastic film
	Conventional	Black plastic film, alachlor, glyphosate-ammonium, pendimethalin
C	Organic	Hand weeding
	Conventional	Hand weeding
D	Organic	Black plastic film
	Conventional	Rice straw mulch
E	Organic	Silver plastic film
	Conventional	Black plastic film

Table 3. Fertility management practices by farm pair

Farm pair	Farm type	Soil amendments
A	Organic	Commercial granular compost consisting of 4.5N–0.9P–1.1K and 75% organic matter (OM) derived from ricinus chaff, vegetable seed bran, peanut husk, sesame bran, rice chaff, soybean powder; mycorrhizal fungi; seaweed powder; chelated calcium powder; and liquid amino acid foliar spray
	Conventional	Synthetic granular fertilizer consisting of 20N–2.2P–8.3K
B	Organic	Commercial granular compost consisting of 5N–1P–1.2K and 81% OM derived from soybean powder, vegetable seed husk, fish powder, bone powder, alfalfa, rice husk, and miscellaneous soil amendments
	Conventional	Synthetic granular fertilizers consisting of 8N–3.4P–6.6K and 13N–6.9P–9.1K–1.8Mg; commercial compost consisting of 1N–0.4P–0.8K and 50% OM derived from bagasse, rice husk, chicken and pig manure, and tobacco leaves; indolebutyric acid; calcium chloride; potassium chloride; amino acid; and lime
C	Organic	Self-made compost consisting of 3N–1.3P–2.5K derived from soybean and bagasse
	Conventional	Synthetic granular fertilizer consisting of 15N–6.5P–12.5K; commercial organic liquid fertilizer consisting of 1N–0.1P–1.7K and 60% OM (dry base) derived from bagasse mixed with fermented molasses; lime; calcium phosphate
D	Organic	Commercial compost consisting of 4.2N–0.8P–2.1K derived from fish powder, bone powder, soybean powder, crab shell powder, rice bran, oil tea, camellia bran, and fermented acid; homemade liquid fertilizer derived from amino acid, milk powder, seaweed extract, hormone, potassium, and phosphate-mobilizing bacteria; calcium phosphate derived from bird guano
	Conventional	Synthetic granular fertilizers consisting of 15N–6.5P–12.5K–2.4Mg and 15N–6.5P–5.8K; indolebutyric acid; urea
E	Organic	Self-made liquid fertilizer derived from soybean powder, rice bran, molasses, shrimp/crab shell powder, seaweed powder, milk, <i>Bacillus subtilis</i> and <i>Streptomyces saraceticus</i>
	Conventional	Commercial liquid fertilizer consisting of 2.9N–0.1P–1.9K; synthetic granular fertilizer consisting of 10N–10.7Ca–1.2Mg

Table 4. Pest management practices by farm pair

Farm pair	Farm type	Pest control practices	
		Target	Control
A	Organic	Preventive Thrips (<i>Thrips palmi</i>) ¹ , whitefly (<i>Bemisia</i> spp.) ¹	Yellow sticky paper Blue sticky paper, lignite
		Preventative Tomato fruitworm (<i>Helicoverpa armigera</i>) Thrips and whitefly	Deltamethrin Chlorfenapyr Deltamethrin
	Conventional	Preventative Tomato fruitworm (<i>Helicoverpa armigera</i>) Thrips and whitefly	Deltamethrin Chlorfenapyr Deltamethrin
B	Organic	Diamondback moth (<i>Plutella xylostella</i>)	<i>Bacillus thuringiensis</i> (Bt)
	Conventional	Nematodes (<i>Meloidogyne</i> spp.) Soil-borne insects: Oriental mole cricket (<i>Gryllotalpa fossor</i>), scarab beetle (<i>Anomala expansa</i>), turnip moth (<i>Agrotis segetum</i>) and click beetle (<i>Melanotus tamsuyensis</i>)	Fenamiphos Chlorpyrifos
		Whitefly Aphids (<i>Myzus persicae</i> and <i>Aphis gossypii</i>) Leaf miners (<i>Liriomyza bryoniae</i>) Diamondback moth	Acetamiprid, mevinphos, Bt Imidacloprid Lambda-cyhalothrin, chlorpyrifos Pheromone trap
C	Organic	Preventative	Trap crop
	Conventional	Preventative	Deltamethrin, pymetrozine, abamectin, chlorpyrifos, acephate, carbaryl
D	Organic	Preventative Tomato hornworm (<i>Manduca quinquemaculata</i>) Leaf miners	Citronella oil Bt, hand picking None
		Tomato hornworm Leaf miners	Methomyl, emamectin benzoate Methomyl
	Conventional	Tomato hornworm Leaf miners	Methomyl, emamectin benzoate Methomyl
E	Organic	Preventative	Trap crop
	Conventional	Leaf miners Thrips	Cypermethrin Bifenthrin

¹Thrips and whiteflies were mainly controlled to prevent the spread of viruses

Table 5. Disease management practices by farm pair

Farm pair	Farm type	Disease control practices	
		Target	Control
A	Organic	Powdery mildew (<i>Leveillula taurica</i>) Tomato spotted wilt virus	<i>Bacillus subtilis</i> Lignite, blue sticky paper to trap vectors, extra compost and amino acids
	Conventional	Prevention Anthracnose (<i>Colletotrichum coccodes</i>)	Curzate Benomyl
B	Organic	None	
	Conventional	Bacterial wilt (<i>Ralstonia solanacearum</i>)	Kasugamycin + copper oxychloride
C	Organic	Soil-borne diseases	<i>Bacillus subtilis</i>
	Conventional	Southern blight (<i>Sclerotium rolfsii</i>) Phytophthora blight (<i>Phytophthora capsici</i>) Bacterial spot (<i>Xanthomonas campestris</i> pv. <i>vesicatoria</i>) Late blight (<i>Phytophthora infestans</i>) Powdery mildew	Flutolanil Etridiazole Kasugamycin + copper oxychloride Mancozeb Triadimefon
D	Organic	Prevention Early blight (<i>Alternaria solani</i>)	Soapberry oil <i>Streptomyces</i> sp.
	Conventional	Early blight, late blight Bacterial spot	Chlorothalonil Kasugamycin + copper oxychloride
E	Organic	Prevention	<i>Bacillus subtilis</i> , <i>Streptomyces saraceticus</i>
	Conventional	Phytophthora blight, late blight Bacterial spot	Etridiazole, mancozeb Copper hydroxide

Table 6 displays pesticide use data for conventional farms only. It is intended to provide an overview of the expected residues that might be found on the tomatoes produced on these farms. Originally this study was to have conducted pesticide residue analysis and a comparison made between the residues found on conventional and organic farms. Unfortunately, due to reduced yields on some farms and sample size requirements for residue testing, this was no longer possible.

Table 6. Pesticide use information for conventional farms

Farm	Chemical	Rates / Frequency
A	[Transplanted Oct. 20, harvested Feb. 1] Curzate, benomyl, deltamethrin, chlorfenapyr	20–25 cc each per 16 L water; applied Nov. 6 and 27
B	[Transplanted Oct. 18, harvested Jan. 21] Fenamiphos Chlorpyrifos Acetamiprid, kasugamycin + copper oxychloride, imidacloprid, lambda- cyhalothrin Chlorpyrifos, kasugamycin + copper oxychloride, acetamiprid <i>Bt</i> Mevinphos, <i>Bt</i>	1.5 kg, 10%, applied Sept. 27 when preparing beds 1.5 kg, 5%, applied Sept. 27 when preparing beds Applied Oct. 20 Applied Nov. 3, 10 Applied Nov. 18, two days after fruit set Applied Dec. 6
C	[Transplanted Oct. 18, harvested Feb. 1] Etridiazole, flutolanil Kasugamycin + copper oxychloride, mancozeb, triadimefon, deltamethrin, abamectin, chlorpyrifos, acephate, carbaryl, pymetrozine Kasugamycin + copper oxychloride, mancozeb, triadimefon, deltamethrin, carbaryl, pymetrozine, abamectin	Applied days before transplanting Applied Nov. 29 Applied Dec. 4, 11
D	[Transplanted Oct. 19, harvested Feb. 2] Methomyl Kasugamycin + copper oxychloride Chlorothalonil Emamectin benzoate, kasugamycin + copper oxychloride Emamectin benzoate	Applied Oct. 24, Nov. 13 Applied Nov. 22 Applied Dec. 5 Applied Dec. 15 Applied Dec. 19
E	[Transplanted Oct. 19, harvested Feb. 2] Copper hydroxide Etridiazole Etridiazole, bifenthrin, mancozeb Cypermethrin, bifenthrin, mancozeb	Applied Oct. 30 Applied Nov. 8 Applied Nov. 10 Applied Nov. 26, Dec. 4

Information on pesticide regulation in Taiwan can be found in Li et al. (2002).

3.5 Tomato sampling and nutritional analyses

Tomatoes were sampled once at the fully red-ripe stage, approximately 104 days after transplanting, although sampling dates varied for each farm. Six plants were randomly selected from each replication and a minimum of six fruits were harvested per plant depending on fruit size.

Sample preparation. Each sample consisted of >600 g fully ripened fruit harvested from a single plot. Fruit were cut, blended with a homogenizer, and filtered through gauze to remove seeds, skin, and membranes. From each sample, six plastic bags were prepared, each containing 10–20 g of tomato slurry. The bags were sealed and immediately stored at -70°C for subsequent analyses of antioxidant activity (AOA), carotenoids, ascorbic acid and total phenolics. Supernatants obtained after centrifugation at 6000 g_n for 10 min were used the same day to measure color and soluble solids concentration.

Chemicals. ABTS (2,2-azino-bis (3-ethylbenz-thiazoline-6-sulfonic acid)), HRP (horseradish peroxidase, type VI-A, 1000 unit/mg solid), linoleic acid, β -carotene and lycopene were purchased from Sigma Chemical Co., St. Louis, Missouri. Trolox (6-hydroxy-2,5,7,8-tetramethyl-chroman-2-carboxylic acid) was purchased from Aldrich Co., St. Louis, Missouri. Other reagents used in this study were all analytical reagent grade.

Antioxidant activity. Entries were analyzed for AOA by TEAC (Trolox equivalent antioxidant capacity) methods as described in Arnao et al. (2001) with some modifications. This method measures the capacity of different components to scavenge the ABTS radical cation as compared to the standard antioxidant Trolox (0–4 mM) in a dose response curve. Trolox is a vitamin E analog. The 10X reaction mixture contained 20 mM ABTS, 0.41 mM H_2O_2 and 50 units of HRP in 50 mM sodium phosphate buffer (pH 7.5) in a total volume of 10 mL and a stable absorbance at 730 nm was obtained in 2 min. The mixture was then diluted to 1X with ethanol. Twenty μL of antioxidant sample with appropriate dilution in water or methanol was added to the 1 mL of 1X reaction medium. The decrease in absorbance, which is proportional to the ABTS quenched, was determined after 5 min by spectrophotometer. The AOA of a sample for the ARP assay was measured within the linear relationship of concentration vs. optical density decrease, and presented as Trolox equivalent (TE) in $\mu\text{mol/g}$ tomato fruit (fresh weight basis).

Carotenoid contents. Ten g of frozen tomato slurry were blended with 100 mL hexane:acetone (6:4, v/v), and 0.1 g MgCO_3 in a homogenizer. Acetone was then washed out five times with salt-saturated water. The hexane extract was filtered with a $0.45\text{ }\mu\text{m}$ filter. Analyses were performed using high-performance liquid chromatography (HPLC, Waters, Mass.) equipped with a 717 plus autosampler, 600 controller, 2487 detector (read at 436 nm) with a $125 \times 4\text{ mm}$ LiChrospher[®] 100 RP-18e column, $5\text{ }\mu\text{m}$ (Merck, Darmstadt, Germany) under isocratic conditions at ambient temperatures. The mobile phase was acetonitrile: methanol (75:25, v/v) at a flow rate of 1.5 mL/min. Commercial β -carotene and lycopene were used as standards.

Ascorbic acid. The determination of total ascorbic acid was on the basis of coupling 2,4-dinitrophenylhydrazine (DNPH) with the ketonic groups of dehydroascorbic acid through the oxidation of ascorbic acid by 2,6-dichlorophenolindophenol (DCPIP) to form a yellow-orange color in acidic conditions (Pelletier, 1985). Twenty g of frozen slurry was blended with 80 mL, 5% meta-phosphoric acid in a homogenizer and centrifuged. After centrifuging, 2 mL of the supernatant was poured into a 20 mL test tube containing 0.1 mL of 0.2% 2,6-DCPIP sodium salt in water, 2 mL of 2% thiourea in 5% meta-phosphoric acid and 1 mL of 4% 2, 4-DNPH in 9N sulfuric acid. The mixtures were kept in a water bath at 37 °C for 3 h followed by an ice bath for 10 min. Five mL of 85% sulfuric acid was added and the mixtures were kept at room temperature for 30 min before reading at OD 520 nm. 2,4-DNPH was added during the ice bath as a blank for a control. Commercial L-(+)-ascorbic acid was used for calibration.

Total phenolics. Total soluble phenolics were extracted from frozen tomato slurry with methanol and determined using Folin-Ciocalteu reagent (Singleton and Rossi, 1965). The reaction mixtures included 0.2 mL methanol extract, 3.2 mL distilled water, 0.2 mL 1 N Folin-Ciocalteu reagent, and 0.4 mL 35% sodium carbonate in water. The absorbance was read at 760 nm after 30 min incubation at room temperature. Chlorogenic acid was used for quantification.

Quality. Color was measured by a colorimeter (Nippon Denshoku Kogyo Co., Ltd. Osaka, Japan) on three scales represented as a, b and L. Color values of fresh tomato slurry were calculated as a/b. Soluble solids concentration was measured with a digital refractometer (PR-101, Atago, Tokyo, Japan). The pH value was measured using a pH meter for the supernatant. Acidity was determined by titration with 0.05 N NaOH to reach pH 8.1 of supernatants and represented as citric acid equivalent (% w/v).

3.6 Statistical analyses

A combined analysis of variance across farms was performed on the fruit quality (pH, soluble solids, acidity, and color) and antioxidant (β -carotene, lycopene, ascorbic acid, total phenolics, and antioxidant activity) data collected from 10 farms using the PROC GLM procedure of the Statistical Analysis System (SAS). The mean differences between the organic and conventional farms were compared using the least significant difference (LSD) test. Simple correlation analysis based on Pearson's product-moment correlation coefficients was carried out as a first step to determine the relationships between the variables measured, using the PROC CORR procedure in SAS. Principal component analysis was performed to summarize the multivariate set of data (fruit quality, antioxidant concentrations and soil properties) into a few components to better understand the structures of the measured responses. It was aimed to derive a few representations (reduced dimensionality) of the data set. Scatter plots of loadings extracted for the first (PRIN1) and second (PRIN2) principal components were constructed to examine, graphically, the interrelationships among the various response variables and the patterns of variations (similarities or dissimilarities) inherent in the sample sets. The PROC PRINCOMP procedure in SAS was used for this analysis.

4 Results and Discussion

Due to severe flooding from Typhoon Nanmadol in December the conventional farm in Farm Pair A lost 80% of its plants. While the flooding was not as severe on the organic farm, it suffered from an infection of tomato spotted wilt virus transmitted by thrips from a neighboring field, which resulted in severely stunted plants throughout the experimental plot. Because of these adverse conditions, the results for Farm Pair A were excluded. The following results and discussion represent the remaining farm pairs.

Mean values for tomato quality analyses measuring pH, soluble solids, acidity, and color value are summarized in Table 7. Mean values for the analyses of tomato nutritional compounds, i.e., carotenoids (β -carotene and lycopene), ascorbic acid, total phenolics, and AOA, are summarized in Table 8.

Table 7. Mean values for tomato quality evaluation by farm pair

Farm pair	Farm type	Variety ¹	pH	Soluble solids (°Brix)	Acidity (% citric acid)	Color (a/b)
B	Organic	V1	4.31 \pm 0.02	3.9 \pm 0.25	0.36 \pm 0.02	2.09 \pm 0.08
		V2	4.31 \pm 0.05	3.8 \pm 0.62	0.38 \pm 0.04	2.00 \pm 0.19
	Conventional	V1	4.24 \pm 0.05	4.8 \pm 0.12	0.39 \pm 0.05	2.24 \pm 0.02
		V2	4.25 \pm 0.05	5.2 \pm 0.46	0.35 \pm 0.04	2.14 \pm 0.09
C	Organic	V1	4.14 \pm 0.06	4.9 \pm 0.68	0.50 \pm 0.06	2.04 \pm 0.07
		V2	4.23 \pm 0.05	4.8 \pm 0.32	0.40 \pm 0.10	2.09 \pm 0.08
	Conventional	V1	4.39 \pm 0.01	3.6 \pm 0.12	0.39 \pm 0.04	2.07 \pm 0.05
		V2	4.36 \pm 0.04	3.7 \pm 0.32	0.41 \pm 0.03	1.95 \pm 0.06
D	Organic	V1	4.26 \pm 0.02	3.4 \pm 0.10	0.36 \pm 0.03	1.93 \pm 0.03
		V2	4.33 \pm 0.02	3.4 \pm 0.10	0.34 \pm 0.01	1.94 \pm 0.04
	Conventional	V1	4.34 \pm 0.05	3.3 \pm 0.12	0.36 \pm 0.04	1.90 \pm 0.11
		V2	4.37 \pm 0.02	3.2 \pm 0.15	0.35 \pm 0.03	1.80 \pm 0.09
E	Organic	V1	4.16 \pm 0.02	3.5 \pm 0.21	0.34 \pm 0.03	1.58 \pm 0.21
		V2	4.22 \pm 0.02	3.4 \pm 0.15	0.34 \pm 0.03	1.50 \pm 0.15
	Conventional	V1	4.16 \pm 0.02	4.0 \pm 0.29	0.41 \pm 0.02	1.64 \pm 0.17
		V2	4.21 \pm 0.04	3.5 \pm 0.15	0.36 \pm 0.01	1.50 \pm 0.15

¹V1 = PT 4769, V2 = PT 4762.

Table 8. Mean values for tomato antioxidant and nutritional evaluation by farm pair

Farm type/ pair			β -carotene	Lycopene	Ascorbic acid	Total phenolics	Antioxidant activity	
				(mg/100 g)			(μ mol/g)	
B	Org.	V1	0.39 \pm 0.07	9.40 \pm 0.61	18.26 \pm 2.07	74.76 \pm 7.45	195.21 \pm 16.82	
		V2	0.35 \pm 0.09	9.17 \pm 0.97	27.89 \pm 3.72	89.17 \pm 5.47	259.84 \pm 51.76	
	Conv.	V1	0.36 \pm 0.16	9.29 \pm 2.44	21.02 \pm 3.55	112.31 \pm 9.23	248.53 \pm 20.20	
		V2	0.41 \pm 0.22	9.58 \pm 2.10	37.91 \pm 8.91	104.20 \pm 21.48	300.76 \pm 66.30	
	C	Org.	V1	0.60 \pm 0.09	7.58 \pm 0.20	20.58 \pm 1.85	100.37 \pm 25.83	263.07 \pm 56.67
			V2	0.64 \pm 0.17	6.12 \pm 1.06	33.77 \pm 1.62	122.21 \pm 18.87	312.61 \pm 99.34
Conv.		V1	0.50 \pm 0.04	10.94 \pm 0.56	18.67 \pm 2.20	65.20 \pm 6.95	260.91 \pm 35.65	
		V2	0.54 \pm 0.03	9.10 \pm 0.63	23.62 \pm 0.90	73.49 \pm 3.52	239.37 \pm 30.92	
D		Org.	V1	0.54 \pm 0.03	8.99 \pm 0.50	19.32 \pm 1.01	70.17 \pm 6.50	263.07 \pm 33.12
			V2	0.56 \pm 0.04	8.94 \pm 0.48	28.33 \pm 2.02	74.22 \pm 4.75	250.68 \pm 6.12
	Conv.	V1	0.43 \pm 0.02	8.05 \pm 0.98	15.24 \pm 1.37	54.25 \pm 5.60	260.38 \pm 23.54	
		V2	0.42 \pm 0.03	7.56 \pm 0.91	25.13 \pm 3.03	76.34 \pm 11.64	263.07 \pm 45.39	
	E	Org.	V1	0.54 \pm 0.08	5.75 \pm 1.67	20.26 \pm 2.50	69.88 \pm 3.53	307.23 \pm 26.56
			V2	0.60 \pm 0.09	5.60 \pm 1.11	28.15 \pm 4.17	80.11 \pm 12.84	366.46 \pm 34.89
Conv.		V1	0.70 \pm 0.04	6.82 \pm 1.28	26.56 \pm 0.99	79.54 \pm 12.81	351.92 \pm 26.27	
		V2	0.73 \pm 0.04	5.40 \pm 1.09	30.76 \pm 1.55	86.44 \pm 14.61	378.31 \pm 10.51	

¹V1 = PT 4769, V2 = PT 4762.

Mean values for soil properties, displayed by farm pairs, are given in Table 9. Soil samples were collected before tomatoes were transplanted and do not reflect changes that may have occurred due to subsequent fertilizer applications.

In general, macro- and micronutrients appear to be at levels adequate for tomato fruit growth. Phosphorus and K levels that could be considered very low and hence a limiting condition for lycopene development were found on Organic Farm D. Micronutrient deficiencies for Fe, Cu and Zn may exist on Conventional Farm D; however, detection of these minerals was problematic for this soil type and low levels may be a result of analytical capabilities.

Table 9. Mean values for soil chemical characteristics by farm pair

Farm pair/ type		pH	E.C. (dS/m)	Total N (%)	Org. C (%)	Ca (ppm)	P (ppm)
B	Organic	6.0	0.10	0.100	0.75	944.0	41.2
	Conventional	7.8	0.32	0.067	0.45	1,572.9	57.9
C	Organic	7.8	0.24	0.150	1.47	5,317.7	37.9
	Conventional	7.6	0.21	0.116	0.73	1,989.8	75.6
D	Organic	7.9	0.86	0.193	1.29	17,865.5	13.8
	Conventional	7.4	0.43	0.233	1.84	44,381.9	59.0
E	Organic	7.4	0.11	0.190	1.18	2,224.3	155.0
	Conventional	4.5	0.18	0.165	0.99	317.5	116.6
		K (ppm)	Mg (ppm)	Fe (ppm)	Mn (ppm)	Cu (ppm)	Zn (ppm)
B	Organic	145.9	142.4	727.7	24.6	5.3	4.1
	Conventional	124.5	193.8	652.9	78.6	2.1	4.8
C	Organic	232.0	587.5	797.5	203.5	5.1	9.3
	Conventional	195.5	354.5	573.2	123.6	5.7	9.3
D	Organic	17.9	566.5	236.0	331.3	2.5	6.8
	Conventional	181.9	1,039.0	0.0	196.9	0.0	0.0
E	Organic	57.8	175.4	1,032.7	120.6	8.7	14.4
	Conventional	146.8	35.0	1,297.9	27.7	6.7	4.7

Mean comparison of tomato fruit quality parameters by farm type (organic versus conventional) produced significant differences for pH only (Table 10) with conventional farms, in aggregate, being higher ($P \leq 0.01$). Quality parameters that are considered more important, i.e., soluble solids, acidity and color value, showed no significant differences between treatments.

Table 10. Mean comparison of fruit quality parameters by farm type

Farm type	pH	Soluble solids (°Brix)	Acidity (% citric acid)	Color (a/b) ¹
Organic	4.25	3.89	0.38	1.90
Conventional	4.29	3.89	0.38	1.91
Difference (O–C)	–0.04**	0.00 ^{NS}	0.00 ^{NS}	–0.01 ^{NS}

^{NS}, *, ** Nonsignificant or significant at $P \leq 0.05$ or 0.01, respectively.

¹Values for a and b were measured with a chromometer using a red standard surface. Immature green tomatoes have an a/b ratio less than zero. The a/b ratio increases to zero and above as fruit color becomes a dark red.

Mean comparisons of nutritional quality parameters by farm type (organic versus conventional) are presented in Table 11. No significant differences were found between farms for β -carotene, lycopene, ascorbic acid, total phenolics and AOA when farm types were aggregated by treatment. There was no evidence of varietal interaction, by farm, except for ascorbic acid where farm type effects differed by variety.

Table 11. Mean comparison of antioxidant components by farm type

Farm type	β -carotene	Lycopene (mg/100 g)	Ascorbic acid	Total phenolics	Anti-oxidant activity (μ mol/g)
Organic	0.53	7.69	24	85	277
Conventional	0.51	8.34	25	81	288
Difference (O–C)	0.02 ^{NS}	–0.65 ^{NS}	–1 ^{NS}	4 ^{NS}	–11 ^{NS}

^{NS} Nonsignificant at $P \leq 0.05$.

The finding of nonsignificance among treatments presented in Tables 10 and 11 does not indicate there were no differences among farms. Most likely it represents a situation in which extraneous factors more strongly influenced the statistical analyses such that treatment effects on the measured parameters were obscured and not detected when farms were aggregated by farm type. There was considerable variability within treatments such that a valid comparison of differences between organic and conventional farms as groups did not seem appropriate. Within conventional farms there were dissimilar environmental conditions, soil types and agronomic practices. Large variation was also present within organic farms.

As described in the Protocols for Site Selection (Section 2.1), Ryan et al. (2004) also cited variability within and between organic and conventional farms and the differing environmental conditions in which they are located as factors that often lead to inconclusive results.

To be able to draw more meaningful conclusions and offer better interpretation of results from this study, mean comparisons of individual farm pairs are interpreted; significant differences for antioxidant components between two of the four organic and conventional farm pairs were found.

Table 12 gives the results of the mean comparison of antioxidant components for Farm Pair B. Values for ascorbic acid and total phenolics were significantly different ($P \leq 0.01$) with the conventional farm obtaining higher concentrations. There were no significant differences in β -carotene, lycopene, or AOA between farms.

Table 12. Mean comparison of antioxidant components by farm type for Farm Pair B

Farm type	β -carotene	Lycopene (mg/100 g)	Ascorbic acid	Total phenolics	Anti-oxidant activity (μ mol/g)
Organic	0.36	9.29	23	82	228
Conventional	0.38	9.44	30	108	275
Difference (O–C)	–0.02 ^{NS}	–0.15 ^{NS}	–7 ^{**}	–26 ^{**}	–47 ^{NS}

^{NS}, *, ** Nonsignificant or significant at $P \leq 0.05$ or 0.01, respectively.

Table 13 gives results of the mean comparison of tomato fruit quality parameters for Farm Pair B. Differences were highly significant for soluble solids ($P \leq 0.01$) and color value ($P \leq 0.05$) in favor of the conventional farm, while the organic farm produced tomatoes with significantly higher pH ($P \leq 0.05$).

Table 13. Mean comparison of fruit quality parameters by farm type for Farm Pair B

Farm type	pH	Soluble solids (°Brix)	Acidity (% citric acid)	Color (a/b) ¹
Organic	4.31	3.83	0.37	2.05
Conventional	4.25	4.98	0.38	2.20
Difference (O–C)	0.06*	–1.15 ^{**}	–0.01 ^{NS}	–0.15*

^{NS}, *, ** Nonsignificant or significant at $P \leq 0.05$ or 0.01, respectively.

¹Values for a and b were measured with a chromometer using a red standard surface. Immature green tomatoes have an a/b ratio less than zero. The a/b ratio increases to zero and above as fruit color becomes a dark red.

Table 14 gives the results for mean comparisons of antioxidant components for Farm Pair C. In this case, tomatoes grown on the organic farm had significantly higher concentrations of ascorbic acid ($P \leq 0.05$) and total phenolics ($P \leq 0.01$). The conventionally grown tomatoes, however, had significantly higher concentrations of lycopene ($P \leq 0.01$). There were no significant differences between farms in levels of β -carotene and AOA.

Table 14. Mean comparison of antioxidant components by farm type for Farm Pair C

Farm type	β -carotene	Lycopene (mg/100 g)	Ascorbic acid	Total phenolics	Anti-oxidant activity (μ mol/g)
Organic	0.62	6.85	27	112	288
Conventional	0.52	10.02	21	70	250
Difference (O–C)	0.10 ^{NS}	–3.17 ^{**}	6*	42 ^{**}	–38 ^{NS}

^{NS}, *, ** Nonsignificant or significant at $P \leq 0.05$ or 0.01, respectively.

Table 15 gives the results for the mean comparison of tomato fruit quality parameters for Farm Pair C. In this case, the conventional farm produced tomatoes with significantly higher pH ($P \leq 0.01$) while tomatoes on the organic farm developed significantly higher levels of soluble solids ($P \leq 0.01$). Acidity levels and color value were not significantly different between farms.

Table 15. Mean comparison of fruit quality parameters by farm type for Farm Pair C

Farm type	pH	Soluble solids (°Brix)	Acidity (% citric acid)	Color (a/b) ¹
Organic	4.19	4.88	0.45	2.07
Conventional	4.38	3.62	0.40	2.01
Difference (O–C)	–0.19**	1.26**	0.05 ^{NS}	0.06 ^{NS}

^{NS}, *, ** Nonsignificant or significant at $P \leq 0.05$ or 0.01, respectively.

¹Values for a and b were measured with a chromometer using a red standard surface. Immature green tomatoes have an a/b ratio less than zero. The a/b ratio increases to zero and above as fruit color becomes a dark red.

Table 16 gives the results for the mean comparison of antioxidant components for Farm Pair D. Although differences were not significant, the organic farm had higher values for all individual antioxidant compounds except AOA. Mean comparison of tomato fruit quality parameters between farms in Farm Pair D showed significant differences in pH only ($P \leq 0.05$) with the conventional farm being higher (results not shown).

Table 16. Mean comparison of antioxidant components by farm type for Farm Pair D

Farm type	β-carotene	Lycopene (mg/100 g)	Ascorbic acid	Total phenolics	Anti-oxidant activity (μmol/g)
Organic	0.54	8.97	24	72	257
Conventional	0.42	7.81	20	65	262
Difference (O–C)	0.12 ^{NS}	1.16 ^{NS}	4 ^{NS}	7**	–5 ^{NS}

^{NS}, *, ** Nonsignificant or significant at $P \leq 0.05$ or 0.01, respectively.

Table 17 gives the results for mean comparison of antioxidant components for Farm Pair E. While differences were not significant, tomatoes grown on the conventional farm had higher concentrations of all nutritional compounds. Mean comparison of tomato fruit quality between farms showed no significant differences in any of the four categories measured (results not shown).

Table 17. Mean comparison of antioxidant components by farm type for Farm Pair E

Farm type	β -carotene	Lycopene	Ascorbic acid	Total phenolics	Anti-oxidant activity
	(mg/100 g)				(μ mol/g)
Organic	0.57	5.68	24	75	337
Conventional	0.72	6.11	28	83	365
Difference (O–C)	–0.15 ^{NS}	–0.43 ^{NS}	–4 ^{NS}	–8 ^{NS}	–28 ^{NS}

^{NS} Nonsignificant at $P \leq 0.05$.

It is useful to examine farm pairs as single case studies in order to determine whether there are unique underlying causative processes or management practices responsible for the observed effects. Results are considered representative of specific site conditions and management practices. Each pair is treated as a single test of the hypothesis that farm management practices, specifically organic and conventional, influence the development of antioxidant compounds in significantly different ways.

A close inspection of Farm Pair B shows that the tomatoes grown under conventional management had significantly higher levels of ascorbic acid and total phenolics than tomatoes grown under organic management ($P \leq 0.01$) (Table 12). Possible explanations for this are varied. Even though farms in this pair were in close proximity (within 2 km), matching soil types was not achieved. The conventional farm had an extremely fine sandy loam soil while the organic farm had a loam soil according to soil survey maps from the Taiwan Agricultural Research Institute (TARI). However, laboratory analysis of soil samples conducted at AVRDC found both soils had similar textures, silty loam, as defined by the USDA textural triangle classification scheme based on the percentage of sand, silt, and clay. Therefore, it is possible that these two farms had soils that were more similar than indicated by information provided by TARI due to the resolution used by soil survey maps, which sometimes are not able to account for micro-variation at the field level.

Whether this apparent difference in soil types alone influenced the development of antioxidant compounds, specifically ascorbic acid and total phenolics, is difficult to determine. Lester and Eischen (1996) found soil type differences to have a significant effect when measuring quantities of carotenoids in melons grown on fine sandy loam versus silty clay loam soil. These two particular soil types are dissimilar in properties such as plant nutrient retention, moisture holding capacity and root development. The two soil types on Farm Pair B, extremely fine sandy loam and loam as identified by TARI, are not as dissimilar with respect to these specific properties as the two soil types in the melon study. Therefore, it is possible that soil type differences may not have been a significant factor in moderating the production of antioxidant compounds on Farm Pair B.

Aside from soil type effects, a reasonable explanation for significant differences in antioxidant compounds found within Farm Pair B is the effect of irrigation practices.

Both farmers used furrow irrigation but the conventional farmer applied deficit irrigation (described in Section 2.2) more deliberately than the organic farmer in order to enhance tomato quality due to his intention to market the tomatoes after the experimental trial was over. While not specifically measured, field observations for size and yield indicated the conventional tomatoes were much smaller than those produced on the organic farm and yields were reduced—both consequences of deficit irrigation. Similar results were obtained by Mitchell et al. (1991) and De Pascale et al. (2001) when measuring the effects of withholding water on tomato fruit quality, size and yield. Increased levels of carotenoids were also observed in this study. Quality analyses results for conventional Farm B are in agreement with these other studies showing significantly higher soluble solids ($P \leq 0.01$) and color value ($P \leq 0.05$).

The significant difference in soluble solids between the organic and conventional farm can be further explained by what is referred to as a ‘concentration effect’. In a meta-analysis of the nutritional differences in organic and conventionally grown foods over the past 50 years, Worthington (1998) concluded that trends could be identified supporting the conclusion that organic food was higher in ascorbic acid and lower in nitrates. However, no general trends could be established for differences in mineral content, vitamins and protein. The author believed that, in general, when nutritional differences were found between organic and conventional foods it was most likely attributed to differences in water content since organically grown foods consistently measured higher in dry matter content.

Worthington (1998) speculated that foods with higher solids contained more nutrients primarily as a result of lower water content, i.e., the nutrients became concentrated in a less dilute matrix. The concentration effect theory offers an explanation for differences observed in antioxidants on farms in Farm Pair B as well as Farm Pair C, to be discussed next. The higher soluble solids in the conventional tomatoes on Farm Pair B could be indicative of higher concentrations of ascorbic acid and total phenolics, but the correlation between soluble solids and antioxidants is not well established. Further investigation of the role that water content plays in eventual concentrations of nutritional compounds would be useful in establishing causative relationships.

The soil fertility practices used by the farmers of Farm Pair B were notably different. Plant nutrients did not appear to be limited, as indicated by results from initial soil analyses, on either farm. The organic farmer applied a vegetable-based compost amendment prior to transplanting. This application was the only contribution made for providing plant nutrients during the entire trial. The organic farmer was confident that his soil had enough residual fertility from previous green manure crops to satisfy tomato nutrient needs. No signs of nutrient deficiency were seen throughout the trial.

By comparison, the conventional farmer applied soluble inorganic fertilizers regularly as recommended by local farmer’s associations and agricultural improvement stations. An animal manure-based compost product as well as a root-stimulating compound, indolebutyric acid, was used at transplanting. Field observations of tomato plants

on the conventional farm indicated signs of nutrient deficiencies near the end of the trial (just prior to sampling), especially with regard to nitrogen.

Exhaustion of N supplies, or N stress, has been shown to increase concentrations of phenolic compounds in buckwheat (*Fagopyrum esculentum*) (Krause and Reznik, 1976 as cited in Patil et al., 1995). Therefore, it is possible that this apparent condition on the conventional farm, while not directly measured by soil samples or leaf tissue analyses, made a difference in antioxidant production at least with respect to concentrations of total phenolics.

Pest control practices between Farm Pair B differed substantially. The conventional farm applied mainly organophosphate broad-spectrum insecticides for control of soil-borne pests as well as other insects that were observed in the experimental plot, i.e., whiteflies, leafminers, and aphids. Two of the pesticides, fenamiphos and mevinphos, are rated Toxicity Class I and are considered highly toxic to humans. This farmer also used pheromone traps as a lure for diamondback moth (DBM).

The organic farmer, on the other hand, applied reflective silver synthetic film over beds for weed and insect control. Thrips, aphids and several other insect pests avoid crops planted in this type of synthetic mulch. Subsequently the organic farm did not suffer from similar outbreaks of pests observed on the conventional farm except for DBM, which was controlled by handpicking and the eventual application of *Bt*.

It has been hypothesized that pest pressures stimulate the production of antioxidant compounds, especially phenolics. In the case of Farm Pair B, the organic farm was more exposed to biotic stress in that a minimal level of pest pressure was tolerated before relatively non-toxic controls were applied; however, pest pressures overall were less than those observed on the conventional farm.

Conversely, the effect of synthetic pesticides has been studied and shown to both stimulate and inhibit the production of antioxidant compounds. The herbicide glyphosate was reported to have inhibited the production of β -carotene in nutsedge and speculated to act in the same manner in kale (Abu-Ismaileh and Jordan, 1978 as cited in Mercadante and Rodriguez-Amaya, 1991). Rouchard et al. (1983) reported the same results for the herbicide metoxuron which reduced the content of β -carotene in carrots when soil was treated one day after sowing. In the same study, however, organophosphate insecticides were reported to have increased the content of β -carotene (they did not evaluate other antioxidants). Significant differences were found in carrots grown in soil treated with the organophosphates fonofos, chlorfenvinphos and bromophos one day before sowing when compared to control plots where no insecticides had been sprayed. Conventional Farm B applied two organophosphates, fenamiphos and chlorpyrifos, when preparing beds prior to tomato transplanting and periodically throughout the trial. It must be considered that this practice also contributed to increased levels of antioxidants.

The results for Farm Pair C are much different than those of Farm Pair B. In this case, the organic farm produced significantly higher levels of ascorbic acid and total phenolics, but significantly lower levels of lycopene. For this farm pair, abiotic stress

had a strong impact on developing antioxidant compounds in addition to management practices.

The organic farm experienced temporary flooding conditions after Typhoon Nanmadol struck in December. According to local weather stations the area received 105.5 mm of precipitation from December 1–7. It has been shown that antioxidant compounds are sensitive to soil water content. Higher levels are produced when plants experience drought-like conditions imposed by deficit irrigation practices (Mitchell et al., 1991). After the typhoon, drainage was less efficient on the organic farm as compared to the conventional farm site and water remained in the experimental plot for a longer period of time. The abiotic stress imposed by anaerobic conditions from flooding on the organic farm impacted root development and plant growth. This interrupted the transport of nutrients and reduced oxygen levels in the soil atmosphere.

Hubbell et al. (1979) measured a 50% reduction in photosynthesis in tomato when soil oxygen was replaced with a N₂ gas mixture. It was also reported that heavy rainfall in Taiwan (200 mm) reduced oxygen concentration in the soil atmosphere to 4%. (A concentration of 2% causes severe wilting and reduced yields in tomatoes.) Thus conditions on the organic farm may have inhibited the ability of surviving tomato plants to produce antioxidant compounds such as lycopene, which rely heavily on photosynthetic activity.

Once plants recovered, increased weed incidence was observed on Organic Farm C. Giannopolitis et al. (1989) reported on the effect of weed interference on nitrate and β -carotene concentrations in lettuce. Significantly lower values for nitrate accumulation as well as β -carotene content were found in lettuce that was subjected to weed competition. β -carotene reductions between 51–83% were measured, which suggested that weeds had a strong impact on the development of these compounds. Species reported in that study included *Urtica urens*, annual grasses (*Avena sterilis* and *Phalaris brachystachys*) as well as annual broadleaves (*Raphanus raphanistrum*, *Sonchus oleraceus*, *Capsella bursa-pastoris*). Weeds observed on the organic farm were annual broadleaves, primarily *Amaranthus retroflexus* (allowed to grow to a size that was greater than neighboring tomato plants), *Solanum nigrum*, *Bidens bipinnata* and *Malvastrum coromandelianum*.

Lycopene is one of the biochemical precursors of β -carotene. It would be reasonable to conclude that weed interference would have a similar effect on antioxidants from which β -carotene is synthesized. Thus the lack of effective weed management on the organic farm combined with adverse soil drainage leading to compromised growing conditions may have been responsible for the significant differences measured in lycopene concentrations between Farm Pair C.

With regard to the other antioxidants, both farms were well matched with respected to soil type and series (Table 1) but differed in quantified soil properties (Table 9) for available Ca, P, Mg, Fe and Zn. The organic farm had ample supplies of Ca, Mg, Fe and Zn but was lower in P in comparison to the conventional farm. Soil fertility differences may have also contributed to differences measured in antioxidant compounds.

Soil fertility practices were routine for both farms with the organic farm applying a homemade compost while the conventional farm using synthetic NPK soluble fertilizers as well as calcium phosphate. Pest and disease control measures were minimal on the organic farm with only an adjacent trap crop used (effectively) for pest control and applications of beneficial microbes used for disease prevention. The conventional farmer regularly applied synthetic pyrethroids and organophosphate insecticides for prevention of pest outbreaks as well as synthetic fungicides for controlling fungal diseases.

The combined effects of management practices and site conditions contributed to the significant differences found for ascorbic acid and total phenolics on the two farms. Quality parameters for soluble solids were higher in the organically grown tomatoes. As with Farm Pair B, it is possible that this contributed to increased concentrations of ascorbic acid and total phenolics.

Farm Pairs D and E were well matched with respect to environmental influences (temperature, rainfall, and aspect) but differed in soil types. Farm Pair D had the same soil series but soil types were silt loam soil and loam soil, respectively. For Farm Pair E both the soil series and soil types were different, again silt loam versus loam. Since significant differences in the development of antioxidant compounds were not found for either farm pair, soil type may not have had a significant effect on results.

Examination of soil fertility practices for Farm Pairs D and E (Table 3) list compost mixtures, beneficial bacteria, bird guano and liquid fertilizers as amendments used by organic farmers for supplying plant nutrients. Conventional Farm D used inorganic granular fertilizers and indolebutyric acid, while Conventional Farm E used inorganic granular and liquid fertilizers.

Results of pre-trial soil analyses indicated available macronutrients were sufficient for all farms except Organic Farm D (Table 9). Levels of available P and K at this site were low prior to transplanting. These two nutrients have been shown to influence the development of carotenoids in tomatoes. Therefore, it might have been reasonable to expect Conventional Farm D would perform better and produce tomatoes higher in β -carotene and lycopene content due to its nutrient status. However, there were no significant differences for antioxidant concentrations between the organic and conventional farms in this matched pair, in fact, the organic farm had slightly higher concentrations of all individually measured antioxidants (excluding antioxidant activity).

The apparent P and K deficits on Organic Farm D appear to have been overcome through the use of phosphate-mobilizing bacteria added to liquid fertilizer to increase the availability of nutrients in the soil; seaweed extracts were also applied that contain chelated elements necessary for plant growth. Some nutrients in this form have been shown to be more effective in terms of crop response. Verkleij (1992) reported that seaweed extracts also act to increase the efficiency of available plant nutrients as well as enable the mineralization of otherwise unavailable nutrients for plant root uptake. These management practices as well as the application of bird guano containing calcium phosphate may have made up for deficits in available P and K indicated by initial soil analyses

such that the organic farm was able to produce tomatoes higher in antioxidant concentration than its conventional counterpart.

The farms of Farm Pair D experienced the same incidence of leaf miner and tomato hornworm infestations but strategies for control were quite different. The conventional farm applied methomyl, an acutely toxic broad spectrum insecticide, rated Toxicity Class I according to information available from the U.S. Environmental Protection Agency's Substance Registry System (<http://www.epa.gov/srs/>). The organic farm did not find it necessary to control for leaf miners and only applied *Bt* for control of tomato hornworm larvae when pest pressure was high enough to warrant. Prior to this, larvae were hand-picked from plants.

Pest control practices for Farm Pair E indicate the conventional farm applied synthetic pyrethroid insecticides (Toxicity Class II) to control leaf miner and thrips while its organic counterpart used preventative sprays of citronella oil and planted a trap crop adjacent to the experimental plot. The trap crop was used to lure insect pests away from the tomatoes into weed species that served as attractants. This appeared to be effective for replications further away from the trap crop but adjacent rows were not as well protected by this strategy.

Table 5 displays disease management practices and targets. Farm Pair D both had problems with early blight, but the farms used very different control measures. The organic farm used preventative applications of soapberry oil and beneficial bacteria while the conventional farm used a toxic broad spectrum organochlorine fungicide, chlorothalonil (Toxicity Class II). As for Farm Pair E, the organic farm used beneficial bacterial for preventing diseases, while the conventional farm relied on several toxic pesticides for control. Although these pest and disease management practices were quite different within Farm Pairs D and E, both organic and conventional practices proved effective at controlling outbreaks.

Plant stress from biotic and abiotic factors can induce the production of plant defense chemicals, i.e., secondary plant metabolites such as phenolic acids and carotenoids. While pests and diseases were present in plots at Farm Pairs D and E, their levels may not have been high enough to trigger such a protective response in plants.

Results for Farm Pairs D and E show small differences in the five antioxidants measured in tomatoes produced by the two farming systems. However, even though nonsignificant, there are trends in the differences measured. Organic Farm D produced tomatoes with higher antioxidant components than its conventional counterpart with yields on both farms comparable. For these two farms the most noticeable difference in management practices were in soil fertility amendments. Organic Farm D may have been able to increase levels of antioxidants in his tomatoes in comparison to his conventional counterpart through the use of beneficial bacteria and seaweed extracts that mobilized plant nutrients important for secondary plant metabolite production.

Results for Farm Pair E indicate the conventional farm performed better than its organic counterpart in all antioxidants measured. Field observations of yields indicated

the conventional farm also produced larger and more abundant fruit. Overall, the conventional farmer expressed more interest and had a higher level of horticultural knowledge than the organic farmer. The difference in antioxidant compounds found between these two farms could most likely be attributed to farmer commitment to the on-farm trial and better overall management of the experimental plot.

In order to determine which measured parameters were most influential in accounting for the differences among farm pairs in this study, a correlation analysis was used to compare values for measured soil properties with values for antioxidants and fruit quality in order to determine whether significant relationships existed.

Without distinguishing between organic or conventional soils, across all farms there was an increase in AOA associated with increased P ($r = 0.77$; $P < 0.02$) and Fe concentrations ($r = 0.72$; $P < 0.04$). For fruit quality parameters, a linear relationship was found between increasing K concentrations and acidity ($r = 0.77$; $P < 0.02$). However, there was a negative correlation between Fe and pH ($r = -0.76$; $P < 0.03$); this was also the case for P and color value ($r = -0.79$; $P < 0.02$). Correlation analysis also confirmed the relationship between soluble solids and total phenolics ($r = 0.98$; $P < 0.001$ and $r = 0.82$; $P < 0.05$ when outliers were removed), but not for ascorbic acid.

The results of this correlation analyses suggest that for farms participating in this study there was a positive correlation between P concentrations in the soil and AOA in tomatoes. The AOA value was obtained from methanol extract, which includes a mixture of phenolics, ascorbic acid and a lesser amount of β -carotene and non-antioxidants such as sugars and citric acid. Thus, AOA in this study mostly accounted for phenolic compounds and ascorbic acid and is not indicative of lycopene and β -carotene concentrations. Therefore, it appears that P concentrations in the soil were significant contributors to the development of total phenolics and ascorbic acid.

Iron was negatively correlated with fruit pH values and positively correlated with farms that had increased levels of P. Interestingly, farms that had significantly higher concentrations of ascorbic acid and total phenolics also had significantly lower values for fruit pH. Indirectly it can also be seen that increasing Fe concentration in the soil may have also influenced production of ascorbic acid and total phenolics.

5 Summary and Conclusions

In general, we could not identify significant differences between tomatoes grown under conventional or organic farming systems. With the exception of fruit pH, there were no significant differences identified for fruit quality parameters (soluble solids, acidity and color) or nutritional parameters (lycopene, β -carotene, ascorbic acid, total phenolics and antioxidant activity).

However, when comparisons were made between paired farms, inconsistent yet significant differences for ascorbic acid, total phenolics and lycopene were observed in two matched pairs; while notable but nonsignificant differences were found between two other pairs for ascorbic acid and total phenolics. Results were mixed as organic farms C and D and conventional farms B and E produced tomatoes with higher antioxidant compounds. With respect to lycopene, only Conventional Farm C produced significantly higher levels than its counterpart. This was most likely due to flooding conditions and weed interference on the Organic Farm C, which severely impacted lycopene production. With such contradictory results it was not possible to distinguish one farming system from another in terms of consistent effects on antioxidant compounds and fruit quality.

Various explanations have been put forward for consideration given the current knowledge of farm management practices and environmental influences on tomato fruit quality and nutritional constituents. Potential causative processes that may have been responsible for outcomes include unique soil amendments that enhanced nutrient availability, especially P; deficit irrigation practices that increased fruit quality and nutritional compounds by reducing water content; N-stress induced development of ascorbic acid and total phenolics; unknown effects of organophosphate pesticides on carotenoids; flooding that reduced plant growth and lycopene development; detrimental impact of weed interference on carotenoid concentrations; and soil P and Fe effects on increasing concentrations of ascorbic acid, total phenolics and fruit pH.

These findings may be useful for evaluating specific site conditions and their potential for affecting antioxidant development on future crops. Results from this study show that the development of secondary plant metabolites in tomatoes is influenced by many factors—some controllable, others not. Due to this lack of control it may not be possible to identify farming system effects that are consistently present on organic or conventional commercial farms wherever they are located. It may be more appropriate to take the lessons learned and devise methods for farmers, in general, to optimize management practices such that greater levels of these important nutritional compounds are produced in all agricultural production systems.

This study demonstrated that on-farm trials comparing agricultural production systems are possible in conditions where communication with farmers and farm associations is difficult due to language barriers and lack of experience in cooperating with research institutions. The organic farmers were initially unfamiliar with on-farm research but

were capable of understanding constraints required for experimental plot management as well as requirements for adequate record keeping. Some were very enthusiastic about participation and were very conscientious about ensuring experimental plots were well managed. Lastly, in this study we were able to demonstrate that high quality tomatoes with increased levels of antioxidant compounds can be produced on organic farms located in humid sub-tropical conditions, where pests and diseases are prolific.

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Photo Gallery



Tsai organic farm, Shinhua (Organic Farm B).



Research Assistant Shao-wen Chiang (left) and Lead Scientist Heidi Lumpkin observe plot.



Shao-wen Chiang observes ripening fruit.



Mr. Tsai, farmer of Organic Farm B in Shinhua.



Mr. Lin, farmer of Organic Farm C in Madou, discusses variety selection in his greenhouse.



Peter Yuo, farmer of Conventional Farm E in Puli.



Mr. Chen, Organic Farmer D (far left) and Mr. Druang, Conventional Farmer D (yellow hat), discuss the research project with Heidi Lumpkin and her research assistants at Mr. Druang's farm.



Harold Lin (center), farmer of Conventional Farm B in Shinhua, discusses his research contract with Shao-wen Chiang and Yu-chi Roan.



Farmers of Organic Farm E in Puli talk with AVRDC staff.

