Reviewers' comments and our responses

The result of evaluations of three reviewers and responses of program leader to each reviewer's comments is as follows:

1) Action classjudgment of the research program

[B]

2) Comments and suggestions for the research program

In general, the research program has been carried out using appropriate approaches and methods and it appears of fair scientific quality. The program consists of 8 research projects, in general focusing on highly relevant topics. a)However, the coordination among projects seemed to be lacking. The cooperation between the projects and the contribution of each of the projects to the overall program could be made more clear and in some cases improved. The link of the projects with the real problems of today's greenhouse horticulture in Japan, being increased costs, reduced profitability, an ageing population of growers and the public demand for reduction of environmental load, could be made stronger.

The first five projects had to do with greenhouse production of vegetables and ornamentals. They developed some innovative approaches to decreasing energy/labor use but they did not seem to coordinate among the projects but rather worked independently of each other. A better inter-communication among the projects facing very similar topics (i.e. 141a0, 141b0 and 141c0) is advisable for trying to reach a synergistic interaction avoiding large overlapping of the research activities. Similarly, efforts should be put also to improve transversal inter-communication between projects sharing common methodologies and approaches (e.g. genetic improvement through molecular and genomic tools). The projects on molecular breeding in vegetables seem to be completely separated from breeding in ornamentals, whereas the molecular tools and techniques are expected to be the same. Although there was excellent work being done in breeding, molecular genetics and molecular breeding, it was not clear that the researchers were working closely with their breeding colleagues at NARO or otherwise.

The research program deals with important topics regarding greenhouse cultivation in Japan and it will surely help to improve the yield and reduce the environmental load and inputs necessary by this sector. b)However it seems that there is a very huge gap between research about a high-tech automation and the actual majority situation of low-

tech plastic-house cultivation. As the researchers developed the innovative approaches for greenhouse production, it was not clear how these new approaches were being communicated to the growers. What is the extension effort to get these innovations to the growers and used? Are there programs available to help the smaller growers with financial and/or marketing aspects of these innovations in greenhouse production? We suspect that some growers will need both financial help as well as help with marketing their products/new crops. c)Therefore the more applied research needs collaboration with and input from greenhouse growers. When 'research-minded' growers would be invited to participate in 'project advisory groups' this gap could be made smaller or even disappear.

(Response to the underlined comments)

a) The explanation provided on review days was insufficient because of time limitations, but in reality, there was strong cooperation between the different research projects. For example, the technology for carbon dioxide application shown in project 141c0 was developed in cooperation with project 141a0. Studies for the trial of genomic selection of tomatoes; breeding using gene marker selection of an eggplant cultivar that was parthenocarpic, thorn-less and resistant to soil diseases; and the development of a DNA marker in strawberries showing tolerance to anthracnose were carried out by researchers belonging to both project 141 g0 and project 141f0. We would like to carry out the coming projects with closer and more intense cooperation for more effectiveness and efficiency.

b) It is a fact that a large gap exists between the cultivation methods used by small farmers and the advanced model production systems that are the subject of our projects. However, many techniques developed by the projects are considered to be applicable to smaller scale greenhouse production. The number of greenhouse growers who are keen to adopt the latest techniques and the amount of larger scale production is increasing in advanced production districts. We have shared information about the new techniques over a wide area and made coaching available to the research demonstration growers. We are making active efforts to reduce the gap. For example, some advanced growers are selected as reviewers to evaluate the effectiveness of the techniques, such as cost and benefit for the growers.

c) There are many growers who are very keen to use the best techniques in the advanced production districts and research demonstration farms. By providing these growers with

technical guidance and exchanging opinions with them on-site, we would like to promote technology development studies using a new approach that includes advisory groups made up of growers as part of the project.

3) Comments and suggestions for the 8 research projects

(1) System development of greenhouse vegetable production for high productivity and environmental load reduction (RP141a0)

This project focuses in general on highly relevant questions: how to improve yield in greenhouses, how to reduce production costs. The project deals with strategic aspects for obtaining reduced production costs coupled with an increase in the yield. The approaches and scientific quality is generally good. In some cases focus could be more on the physiological background of the findings.

Humidity control and CO_2 enrichment caused a dramatic yield increase in grafted tomato. a)However you need to determine the trade off with increased disease pressure and/or quality of fogging. Results same in warm vs cool season?

b)The use of abundantly available biomass resources for greenhouse heating seems logic, however, the solution of applying both a gas fan heater and a biomass heater in a greenhouse might be a too high investment. For new greenhouses would biomass heater be sufficient? Where does biomass come from? What is long term supply perspective? Might there be biomass from waste in future? c)On the other hand, local heating versus whole greenhouse heating is a nice approach. Any explanation why it works?

The work on a 'fruit setting robot that sprays phytohormones' does not seem to match with the general program aims. d)The activity for automatic spraying of phytohormones is, to certain aspects, in contrast with the international tendency to reduce the chemical inputs. Automation is nice but is it appropriate for small growers? Instead of hormone sprays might there be a genetic approach...ie parthenocarpic tomatoes (link to 141g0, genomics work). e)Working towards parthenocarpic cultivars seems a better solution to reduce labor demand involved in improving fruit set.

The conclusion that in this project over a period of 5 years a 50% reduction in labor has been achieved compared to conventional cultivation is impressive. However, it seems that this has been achieved under high-tech conditions whereas conventional greenhouses are low-tech. Some very high-tech equipment appear very far from the Japanese average level of the greenhouse technology. f)The project could certainly benefit from so called 'grower groups' that would participate in defining the experiments such that these are more relevant for conventional growers in Japan.

Many times researchers do great work but do not market the application well. However,

praiseworthy is the demonstration initiative to exhibit the plant factory and the extension program for plant factory. The collaboration with private companies on greenhouse climate control (CO_2, fogging) is very good. The number of published papers is high, but could be more focused on higher impact (more physiological) journals.

(Response to the underlined comments)

a) At this stage we have focused only on measuring the compound positive effects of humidity control and CO_2 enrichment, under Japanese environmental conditions and with Japanese cultivars. We intend to consider the relationship between humidity control and plant diseases at a later stage. We plan to estimate the effects of supplementory lighting in winter and prolongation of CO_2 application time in the hot and humid Japanese summer season, in combination with humidity control and CO_2 enrichment.

b) We are trying to lower the total cost by reducing running costs using a hybrid heating system, even if the initial cost might be higher than that of a conventional system. The hybrid system of using a biomass heater (for main and base heating) and an oil heater (for critical and supplemental control) is considered a better solution than using biomass heating alone. (A biomass heater capable of producing sufficient heat to cope with the extremely low temperatures that occasionally occur would be very expensive; using a supplementary oil heater during extreme low temperature conditions is a more economical solution.)

c) We are now studying the physiological mechanisms involved in this project. We are paying particular attention to changes in water content. We are going to push forward with elucidating the mechanism with a nondestructive measurement device and stable isotopes.

d) This project was aimed exclusively at reducing labor requirements for spraying in the Japanese agricultural industry. Because of this, we may not have placed much emphasis on following the global trend towards reducing the use of chemicals. However, we did show that using a spraying robot can reduce the quantity of chemicals required. For this reason, we think that the technique is still environmentally sound.

e) We believe that the development of stable parthenocarpic cultivars still requires more time. We have developed the fruit setting robot as one component of a totally automated

robotic system that will be able to carry out a variety of tasks, including harvesting, transporting and gathering plant information. We don't intend to provide one-purpose robots for growers.

f) A study consortium system for plant factory production by private companies has been built up over the past five years. And field studies are now being carried out with farmers. We intend to cooperate more with producers to promote the introduction of advanced techniques in greenhouses.

(2) Development of a low-cost structure design and environment control for safe, energy-saving, and optimized greenhouse production (RP141b0)

This project aces important aspect to make plastic-house structures less susceptible when subjected to strong wind pressure, effects of earthquakes or weight of snow; similarly important is the cooling by exploiting natural ventilation to improve year-round crop production. It makes use of a unique wind tunnel for testing greenhouse constructions and positions. Some very nice structural analysis was done but we did not see a suggestion on how to better design a greenhouse for better wind stability. Co-operation with 141c0 should be stimulated (also working on greenhouse cooling and making use of CFD techniques). a)It is not clear what greenhouse designing advice follows from the analysis on structural analysis. Is the double arch system the solution to increase greenhouse strength?

b)We saw good suggestions about snow damage management. How often is this a problem? c)It was showed a differential temperature along length of greenhouse but no idea of implications on plant productivity or management challenges. A fogging system showed to decrease temperature by 2°C but increase humidity by 20%. Also decrease ventilation. d)Any suggestions on how the design greenhouse to improve ventilation with fogging?

Being the smallest group (3 members), the number of published papers is impressive (22). However, the major publication list of 6 papers containing 2 papers in Acta Horticulturae (proceedings; not considered as refereed journal papers) and 2 papers "in press" seems an unexpected choice.

(Response to the underlined comments)

a) The wind pressure coefficient Cp had not previously been defined in the greenhouse standard for pipe-framed greenhouses. The Cp we obtained will be effective for the reinforcement of the structure of pipe-framed greenhouses. A further direction of this

study will be incorporating the improvement of the pipe-framed greenhouse into a new structure. The superiority of the double arch structure depends on the loading conditions and the greenhouse's structure. The double arch structure is just one of several options for reinforcement.

b) About eight percent of greenhouses are destroyed by heavy snows. The proposed measure is for greenhouses in regions with less snowfall and less frequent heavy falls. However, public officers and farmers in many areas regard the risk of heavy snow as important because of the huge amount of damage it can cause.

c) In this case, the main wind direction was along the longitudinal direction of the greenhouse, and the natural ventilation in the greenhouse produced a longitudinal distribution of internal air temperatures.

Internal air temperature distribution is an important factor for greenhouse crops. Our research will enable producers to predict air temperatures in different areas of the greenhouse, allowing them to pinpoint those areas where more careful treatment is needed. They will be able, for example, to employ spot fans in areas where higher air temperatures are predicted, thus preventing plant damage caused by exposure to overly warm air. It is believed this approach will lead to more effective methods of improving plant productivity and meeting management challenges.

d) While it is relatively easy to determine the optimum ventilation rate by checking temperature and humidity data and fogging amount, it is difficult to accurately realize the optimum ventilation rate by natural ventilation, which is performed by complex factors such as cooled down air flow, upward flow using buoyancy, and airflow resistance of vents affected by outside wind. Controlling each vent with consideration of real time data for those factors might be one approach. In the near future, the use of some kind of MEMS (Micro Electro Mechanical Systems), or MST (Microsystem Technology) device, especially, micro sensor will make it possible to detect those factors, and this idea might soon be realized.

(3) Development of a sustainable production system in greenhouse horticulture for the Western region (RP141c0)

This research team is not located in Tsukuba and focuses on a sustainable production system for the hilly/semi-mountainous areas in the western region of Japan. The research project appears to be mainly an applied technology adaptation project which

has developed analysis tools and greenhouse design for this region of country. Highly relevant, as hilly/semi-mountainous areas comprise 40% of the total farming area in Japan. a)The problems faced are similar to those of other Japanese regions (i.e. energy saving, reinforcement of plastic-house structure, yield); therefore a close co-operation with other projects such as 141a0 (also focusing on reducing energy costs and improving yields) and 141b0 should be stimulated (also on greenhouse cooling and CFD work). A tight collaboration among these research groups should have been advisable.

b)As this project is in mountainous area with multiple climatic conditions, should go beyond the initial crop and examine other potential crops. Some research were carried out with the involvement of private companies to exploit commercially and transfer the results to the growers, which are positive aspects to satisfy practical requirements.

A good number of publications, still in low-impact scientific journals. c)Considering the number of people working in this project, the number of papers could be expected to be higher. d)However, 9 patents obtained in 2011 is extremely high (although for the reviewers it is not clear what kind of patents these are and whether they can raise money for the organization).

(Response to the underlined comments)

a) Some studies have been conducted in co-operation with 141a0 and 141b0. Some results have been published. Efficiency of carbon dioxide enrichment in a greenhouse was studied in collaboration with 141a0. The new pipe-framed greenhouse was developed and the 'Circulation fan use manual' was published in collaboration with 141b0.

b) The main aim of this study was to improve the productivity of summer-autumn tomatoes, and a high yield production system for tomatoes was developed. We have also examined other crops that can be gown using a year-round production system with greenhouses in mountainous area. These crops include strawberry, melon, sweet pepper, lettuces, edible wild plants etc. Unfortunately, good results with these crops have not yet been obtained.

c) Several manuscripts have been submitted to journals and are awaiting acceptance. Our studies have been conducted through substantive empirical research when collaborating with farmers and public research institutes in farmers' fields. In these experimental sites, it is very difficult to set up a control plot. This is one of the reasons we have published few papers.

d) We regard that some patents can raise money. For example, a monorail type semi-automatic sprayer, which can be carried by hand, is actually sold by Nishizawa Co.,Ltd. And the drip-irrigation system operated and controlled by solar radiation is also sold by PUTIO Ltd. Some other patents are kinds of defense patents.

(4) Development of a year-round high production system for strawberries and vegetables in greenhouse (RP141d0)

Research are finalized to the improvement of agronomic techniques to increase the yield of asparagus, lettuce and, mainly, strawberry. It is impressive that strawberry yields could more than double when combining a dense planting (movable beds) and environmental control (e.g. LED supplementary lighting, CO_2 enrichment). a)However, it seems that the main reason for this high yield is the very high intensity supplementary light (> 400 µmol m^{-2} s^{-1}) and one might wonder whether this is cost-effective (high investment costs and high electricity costs). Estimation of economic and energetic costs would be necessary to have a more complete picture of the proposed techniques. In such kinds of experiments, it would be important use different cultivars/genotypes of the studied species as that should give a better evaluation of the suggested techniques. For instance, in strawberry and asparagus exist genotypes with different physiological properties which could better take advantage of the agronomic practices proposes (e.g. day-neutral strawberry, early/late asparagus sprouting with different cold requirement).

Vapor heat system is a nice approach and directed heating (crown-temperature control) is an innovative approach to save energy costs. The work on developing cultural technologies for year-round stability and yield increase in strawberry and other vegetables seems to be practical (crown- temperature control system; FR light) rather than scientific (no hypotheses on working mechanisms). We note in the picture that the greenhouse roof is high - might it be possible to reduce energy cost by decreasing height of greenhouse structure as well?

Non-everbearing strawberries appear to be traditional in Japan but I wonder if a day neutral type would be better in a protected environment of the plant factory. You may need to breed with Japan quality standards in mind. b)Excellent work on exploring other crop alternatives for these small farmers but will probably need to give them marketing support to sell the crops.

c)The number of papers published (11) by this large group (on average 12 members during 5 years) is very low although some of them are published in international peer reviewed journal with IF.

(Response to the underlined comments)

a) We have estimated the economic cost of supplemental lighting and the data indicated the profitability was improved. But this technique would not be cost-effective because the investment cost of the LEDs is too high at the present time. We expect a big reduction in the price of LEDs in the near future. Additionally, we have obtained results that show that by combining environmental controls without supplementary light, yields until February were increased 1.8-fold. Also, we have already carried out studies using several cultivars.

b) We think that marketing support for small farmers is needed, and the studies on economic evaluation and marketing support will be carried out in the next research stage.

c) The number of papers shown does not include several papers concerned with studies carried out in the previous research stages. We have been in charge of strawberry and asparagus cultivation research. To study such vegetables takes a relatively long period. However, we have steadily published the obtained results as papers, and we intend to publish many more papers hereafter.

(5) Development of an efficient production system for ornamental plants (RP141e0)

The research about the molecular regulation of flowering in chrysanthemums is of high quality together with the differential effect of different wavelength light and have resulted in publications in high impact journals. This knowledge has to be exploited for genetic manipulation of the process in a contest of practical application (e.g. allelic variation, selective sensing). a)However, it will still be a very long way to use this knowledge in breeding programs or new cultivation strategies to grow chrysanthemums for certain peak demands and maintaining a general, stable supply system. Closer connection with breeding and projects like 141a0 (cultivation techniques) should be stimulated. Although day length sensitive plants are managed in greenhouse systems, a day neutral plant might be easier to manage. b)It would be suggested that the next target be converting photoperiodic plants into day neutral plants.

A relevant result is also the energy saving due to the modification of day and night temperature, and CO_2 enrichment for Eustoma cultivation. This work is nice as a way to reduce energy use in winter production cycle of Eustoma. c)The future idea of developing a CO_2 supply system certainly should be undertaken in collaboration with project 141a0.

This project with on average 10.5 members during 5 years has an output of 33 papers

which is good. The scientific production was of high level with a significant number of papers on high IF journals.

(Response to the underlined comments)

a) We will cooperate with the vegetable researchers of project 141a0 to prompt practical application of these fundamental research results, even though, as the reviewers comment, this will not be easy to achieve. We are planning to utilize the molecular biological knowledge of the flowering process for chrysanthemum breeding.

b) Although the comment is true when applied to some plants, the short day length property is an important property of chrysanthemums that can be used to control flowering by controlling lighting. Growers prefer the summer-autumn flowering cultivars, which have a flowering time that can be strictly regulated by day length control.

c) We will develop more efficient CO_2 application techniques through cooperation with project 141a0.

(6) Breeding of fruit vegetables for high quality and productivity (RP141f0)

Excellent breeding work has been carried out which brought both to the development of innovative parental lines, new F1 cultivars or rootstocks in the interested species. An improved parthenocarpic eggplant F1 was released, an MYSV virus resistant cucumber line obtained and linked QTL associated to this trait identified and potentially usable for MAS, a new strawberry cultivar obtained as well as three pepper rootstocks and, finally, new tomato hybrid suitable for hydroponic cultivation.

a)In the objectives good processing quality is mentioned, but shelf life (fresh non-processed products) seems to be missing. b)What makes a genotype 'having good adaptability to hydroponics'? We would like to see a better definition of the traits needed for good adaptability for a hydroponic system. As the breeding continues, it might be useful to select for specific production cycles (spring, summer, fall, winter) as the light and temperatures are different. It would be interesting to know the underlying physiological reasons why Tomato TK8 and TK9 show both a high yield and a high soluble solid content.

The work in developing strawberry cultivars for specific regions and production cycles is excellent. c)Although still working with photoperiodic types, why not work towards developing day neutral types and you are protecting them so this would eliminate the need to consider day length as a factor in fruit initiation and fruit yield.

The initiated collaboration with several Asian countries on characterization, evaluation and utilization of plant genetic resources for food and agriculture is a very good way to go. What is available for strawberries?

On average 13 members working in this project during 5 years, therefore, the number of papers (22) seems somewhat low. Although the practical breeding activity, generally, cannot bring to a large number of publications on high IF journals, the members of this research groups published also in peer-reviewed journal. [d]More focus on publishing in international refereed journals with high impact factors should be stimulated.

(Response to the underlined comments)

a) Our objectives were to improve taste, and processing adaptability. Increasing shelf life was not a target in this project. We recognize that shelf life of vegetables is a very important trait for distribution on the market, especially for export. We will incorporate the shelf life trait as a breeding target in the next project.

b) We selected plants that showed high performance in fruit-setting and fruit-growth abilities, even if the plants were extremely vigorous. Consequently, selected lines with good adaptability to hydroponics have long internodes and big leaves. It is unclear whether there is a relationship between these traits and adaptability to hydroponics; we plan to clarify this in future studies.

c) We cannot definitively distinguish between day-neutral types and ever-bearing types under Japanese climate conditions. Therefore, both are regarded as ever-bearing types which initiate flower buds under long-day conditions in Japan. Because long-day treatment for ever-bearing types is easier than short-day treatment for June-bearing types, in order to promote flower bud initiation from late summer until late autumn, an attempt to use ever-bearing type cultivars for forcing culture has begun, as you suggested.

d) We have published new knowledge acquired through our practical breeding study. We intend to make further efforts to submit articles to international refereed high impact journals.

(7) Development of genome resources and molecular technologies for advanced breeding and cultivation (RP141g0)

This is the cutting-edge research project of the genetic improvement activities and focuses on molecular technologies for advanced breeding and cultivation of vegetables.

The involvement in the tomato genome consortium and the production of a first draft of the eggplant genome make it possible to develop genomic tools to aid the breeding programs in tomato and eggplant. a)In this regard, a better connection with 141f0 project should be established or evidenced if it already exist. It is needed to use the greater information known about the genome in tomato to facilitate the work in eggplant.

The work on parthenocarpy in tomato and eggplant is highly relevant for the overall programme. Emphasis on parthenocarpy is excellent as these are traits that can improve fruit quality and yield consistency. It offers possibilities for yield improvement and reduced labor costs. The isolation of some key gene involved in parthenocarpy, fruit development and the construction of dense genetic maps are relevant results, which are useful means for MAS and precision breeding. b)The perspective of apply genomic selection in tomato and eggplant should be coupled with high throughput phenotyping procedures by using or by developing appropriate automatic field/movable apparatus.

c)With on average 8.5 project members during 5 years, a total output of only 13 papers seems low. The list of major publications (5 papers, with only one paper with a group member as first author) shows that this group has published in high impact journals.

(Response to the underlined comments)

a) We have been working through several studies in cooperation with 141f0, including genomic selection (GS) for simultaneous improvement of yield and soluble-solid content in tomato; marker-assisted breeding of parthenocarpic, prickleless, and soil-borne disease-resistant eggplant cultivars; DNA marker development for strawberry anthracnose resistance, and so on. Even though these studies were in progress and were therefore not picked up as main topics at the review, researchers in charge of each project (141f0 and 141g0) have been working in close coordination and understand the importance of such collaboration.

b) While high throughput phenotyping or 'phenomics' by using automatic field/movable apparatus is in fact a very pioneering and challenging approach, its potential in development of agricultural technology seems still unclear at present and we recognize that we have to keep abreast of global developments in this research area. For the next 5-year research period that will start from 2016, we plan to gather information in this research area and start preliminary study as needed.

c) Researchers in this project tend to focus on quality over quantity of papers and try to publish their papers in international higher-impact journals. The reviewers'

understanding in this regard would be greatly appreciated. However, we do note the reviewers' comments and we will increase our efforts to the maximum to achieve wider publication of our results. Also, we would like to ask reviewers to note that, of the five papers shown in the list, two papers have project members as the first author. In addition, two of the remaining three papers described the results of collaborative studies with other institutes outside of NARO, and for one of these two papers, one of our project members took the initiative and served as corresponding author.

(8) Molecular breeding of ornamental plants (RP141h0)

The scientific quality of this work goes beyond any doubt and is also shown by the high number of papers and patents. The major publications are all in high impact international journals. The genetic engineering of blue color petals and the elucidation of the molecular mechanism of the flavonoid accumulation in Chrysanthemums petals, as well as the development molecular map and genome draft in carnation and identification of QTL for key breeding traits are relevant results gathered by this research group. a)However, here a concern might be that the link with the overall programme and the current problems of Japanese greenhouse horticulture is not clear. Furthermore, no clear vision exists on how this molecular work can be used in actual breeding programmes. How can this be used by a breeder? It should be better defined the way to translate this research into breeding programs.

b)The work on control mechanisms of color is fascinating. Creating blue chrysanthemums via transformation appears to be a viable approach but need to determine how to prevent gene flow. Thus all blue cultivars need to be sterile. As only a few of the 100 cultivars they tested are readily transformed to create a useful blue cultivar, subsequent breeding with transformed materials to develop a blue mums with a wide range of flower characteristics will probably be needed. c)It is concluded that this work will lead to breeding for new ornamental cultivars, however, no co-operation with breeders exists until now. Such co-operation is considered necessary to have a real impact on practical horticulture.

(Response to the underlined comments)

a) We have already started studies for commercialization of transgenic blue-colored chrysanthemums. For carnation breeding, a selection marker of disease resistance has been used in breeding programs.

b) We are trying to develop a method for reducing risks of transgene flow such as a

technique for producing both male- and female-sterility simultaneously.

c) We have been co-operating with private breeding companies for commercialization of transgenic blue-colored chrysanthemums. We have also been co-operating on carnation breeding and a new carnation cultivar bred by collaboration with Aichi prefecture has been released. We will develop further co-operations with breeders.