

Summary of Doctoral Thesis

Enrollment year: 2018/03

UGAS Specialty: Plant Production

Name : Wambrauw Daniel Zadrak

Title	Effect of Supplemental Light on the Quality of Green Asparagus Spears in Winter 'Fusekomi' Forcing Culture
<p>Introduction and purpose</p> <p>Fusekomi forcing culture system has been conducted to produce asparagus in the winter. However, in these methods, spears are mostly growing under small amount of light because of much of snow and low sunlight in the production area causes some problems: the color of the spear is pale, and rutin content is lower compare to those harvested in spring. So the aim of this study was to obtain basic knowledge to improve the quality (color and rutin contents) of asparagus spear in order to produce high value of the spear during winter season by using different irradiation time (2012) and different quantity of fluorescent light (2013) on the winter 'fusekomi' forcing culture.</p> <p>Materials and methods</p> <p>UC157 was used as the plant material. In 2012, 3 trial plots with 2 repetitions was prepared as follows: Control; with no supplemental lighting, FL1-16; 1 supplemental lighting for 16 h a day and FL1-24; continuous (24 hours a day) lighting with 1 white fluorescence lamp. In 2013, 3 trial plots with 2 repetitions were prepared as follows: Control; with no supplemental light, FL1- 24; continuous (24 h a day) lighting with 1 white fluorescence lamps and FL2-24; continuous (24 h a day) lighting with 2 white fluorescence lamps. Spears harvested were measure to evaluated yields, also were used for spear color observation. Sugar content and rutin content analysis were conducted by using HPLC method.</p> <p>Results</p> <p>Spear color was getting better and rutin contents were getting higher according to the numbers of lamps. No negative impact on yields and sugar content by supplemental lighting. Although, the spear weight seems to be lighter in FL2-24 treatment.</p> <p>Conclusion and consideration</p> <p>Supplemental lighting was effective to improve the quality of asparagus spear cultivated in fusekomi forcing culture. According to the number of lamps, rutin content were getting higher and spears color also were getting better.</p>	

Title	Effects of different Quality and Quantity of Supplemental Lighting on the Quality of Green Asparagus Cultivated by Winter 'Fusekomi' Forcing Culture
<p>Introduction and purpose</p> <p>We found that different number of supplemental lighting, light intensity (light quantity) and different irradiation time affects the amount of rutin. In this study, in order to confirm the effects of light on the quality of asparagus spear and also to find the best conditions to produce high quality asparagus during winter season. Tests by using different quantity of fluorescent light and different quality of light on the winter 'fusekomi' forcing culture were conducted.</p> <p>Materials and methods</p> <p>UC157 were used as plant materials. In 2014, 3 trial plots with 2 repetitions was prepared as follows: Control; with no supplemental lighting, FL1-24; continuous (24 hours a day) lighting with 1 white fluorescence lamps and FL3-24; continuous (24 hours a day) lighting with 3 white fluorescence lamps. In 2015, 3 trial plots with 2 repetitions were prepared as follows: Control; with no supplemental light, FL1- 24; continuous (24 h a day) lighting with 1 white fluorescence lamp and FL4-24; continuous (24 h a day) lighting with 4 white fluorescence lamps. In 2016, 3 trial plots with 2 repetitions were prepared as follows: Control; with no supplemental light, Blue; continuous (24 h a day) lighting with 1 blue lamp and UV-B 24; continuous (24 h a day) lighting with 1 UV-B lamp. In 2017, 3 trial plots with 2 repetitions were prepared as follows: Control; with no supplemental light, FL4-24; continuous (24 h a day) lighting with 4 white fluorescence lamps and UV-B12; continuous (12 h a day) lighting with 1 UV-B lamp. Spears were harvested during harvest period (6 week) and used for rutin content analysis and spear color observation. Rutin analysis were conducted by using HPLC method.</p> <p>Results</p> <p>Rutin content of the spears grown under FL1, FL3, and FL4-24 treatment was significantly higher than in control. We found that according to the number of lamps and irradiation time, rutin contents were getting higher and spear color also getting better. Blue light shows significant role to enhance rutin whereas UV-B seems to cause severe damage to the spears, which leads to low rutin content. Interestingly, even blue light enhance rutin synthesis and have almost the same amount as FL1 but spears harvested under blue treatment are paler.</p> <p>Conclusion and consideration</p> <p>FL1, FL3, and FL4-24 treatment was effective to increased rutin content and also better color. Blue light treatment was also found effective to enhance rutin whereas UV-B caused severe damage to spears. UV-B for 24 hours seems to have more stresses to the plant. Blue light only enhance rutin but not the color of the spears.</p>	

Title	Gene Expression of Flavonoid Biosynthetic Genes in Green and White Asparagus Spears
<p>Introduction and purpose</p> <p>The amount of rutin was getting higher and the spear color are also getting better by introducing supplemental lighting and our previous study also showed that rutin is getting higher according to the number of lamps and light quality (blue light). However there is still a few studies on molecular regulation of flavonoid metabolism in asparagus; how light effects the accumulation of rutin and which genes in the flavonoid biosynthesis pathway are light related gene and/or the light response genes to enhance rutin production. Thus the aim of this study is to investigate the effect of light on rutin biosynthetic genes in asparagus spears by examined green spears; spear which grown in the natural sunlight and white spears; sunblocked asparagus.</p> <p>Materials and methods</p> <p>UC157 were used as plant materials. To produce white asparagus, white spears were covered with a sun blocking film, White Silver, TOKANKOSAN CO, LTD, Japan. RNA was extracted by using RNeasy Plant Mini Kit, reverse-transcribed and the cDNAs was used for gene analysis expression using real-time PCR methods. Rutin analysis were conducted by using HPLC.</p> <p>Results</p> <p>Rutin only detected on green spears but not in white spears. The expression analysis of the flavonoid upstream related genes was found no significant difference, however the downstream genes, F3'H and especially FLS were lower. FLS showed low expression level compare to other genes and even in green the expression was found low.</p> <p>Conclusion and consideration</p> <p>FLS expression level compare to other genes were lower and even in green the expression were found low. These results indicating that FLS needed light and FLS play an important role on rutin biosynthesis pathway.</p>	

Title	The Effect of Light Intensity on Asparagus Flavonoid Biosynthetic Gene Expression
<p>Introduction and purpose</p> <p>In the previous section, studies about gene expression of flavonoid related genes in light exposed green spear and the unexposed light white spear was conducted. We found that the downstream gene, <i>FLS</i> was a light related gene in flavonoid biosynthesis pathway. In this section, in order to clarify the mechanism of how light intensity effect flavonoid related genes expression; spears grown under different numbers of light were used in this study.</p> <p>Materials and methods</p> <p>Spears used in this experiment were as follows; White: unpigmented spears; Green: Green asparagus with no supplemental light; FL1: Spears grew under continuous (24 hours a day) lighting with 1 white fluorescence lamps; FL4-24; continuous (24 hours a day) lighting with 4 white fluorescence lamps. RNA was extracted by using RNeasy Plant Mini Kit, reverse-transcribed and the cDNAs was used for gene analysis expression using real-time PCR methods. Rutin analysis were conducted by using HPLC.</p> <p>Results</p> <p>The upstream genes were found no significant difference among the treatment. However, <i>FLS</i> was significantly lower in white compared to FL4 and FL4 has the highest expression level in <i>FLS</i> against other tested plots. There is no significant difference between green and FL1.</p> <p>Conclusion and consideration</p> <p>According to the light intensity (number of lamps), <i>FLS</i> were regulated and/or enhanced. This result indicated and supports our statement that, <i>FLS</i> is lightly related gene and plays an important role in flavonoid regulation in asparagus</p>	

Title	The Effect of Light Exposure Time on Asparagus Flavonoid Biosynthetic Gene Expression
<p>Introduction and purpose</p> <p><i>Flavonol Synthase (FLS)</i>, the downstream gene in flavonoid biosynthesis pathways showed as light related gene and/or the light response genes to enhance flavonoid regulation. The expression level was relatively lower in white spears compare to other genes because white spears do not contain rutin. In this section, to understand the mechanism of rutin regulation and its relationship with light, we made white asparagus first by using sunblocking film, and open the sunblock film in order to expose the spear to the natural sunlight until the white spear turned to be green. Samples were harvested in particular time.</p> <p>Materials and methods</p> <p>During the harvest period, after spears become white, the sunblock film was uncovered, and the spears were exposed to the natural sunlight. In particular of time the spears were harvested; 0 hours after open (white spears): 0 h, 4 h after open: 4 h, 8 hours after open : 8 h, 16 hours after open : 16 h, 24 hours after open : 24 h, 32 hours after open : 32 h, 40 hours after open : 40 h, 48 hours after open: 48 h, 72 hours after open: 72 h, 96 hours after open: 96 h, 144 hours after open: 144 h, and 1 week after open: 1 W. The same samples were used for both rutin analysis by using HPLC and gene expression analysis by using real time PCR.</p> <p>Results</p> <p>Rutin was not detected on spears harvested at 0 h to 16 h after open. However, rutin were increased sharply from 24 h and reach to highest at 1 W after open. The gene expressions of <i>flavonol synthase (FLS)</i> were gradually increased after light exposed; 4 h after open (light exposed) the expression were sharply increased and then decreased as it goes to 1W. The coloration in the spears was found at the begining on the tip of some spears at 8 h after open and continued until the spears become greener in 1 W after open.</p> <p>Conclusion and consideration</p> <p>Rutin content began to increase about 24 after and the coloration also began almost at the same time period. However, expression level of <i>FLS</i> was began at 4 h after open, reach to the max of its gene expression on 72 h after open and then decreased to 1W. These finding indicating that in the short time of light exposure rutin regulation was stimulated and the synthesis started even before the pigmentation occurs.</p>	