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IDENTIFICATION AND CHARACTERIZATION OF AN ERAF17 cDNA THAT EXPRESSION AT APICES WAS CORRELATED WITH THE SEX EXPRESSION IN CUCUMBER PLANTS

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Cucumber (*Cucumis sativus* L.) plants produce unisexual flowers with only stamens or pistils on the same plants (monoecious). Ethylene are known to induce the formation of female flowers. Although there are many evidences concerning the effect of ethylene on sex determination in cucumber plants, the action mechanisms of ethylene in the regulation of sex expression are remained to be explored. We tried to screen ethylene-responsive genes that were related to the induction of female flower formation by ethylene using the differential display technique and cloned an ERAF17 cDNA of which expression was induced by ethylene. The Pattern of expression of ERAF17 was correlated with that of sex expression in cucumber plants. Furthermore, the transcript was detected only in female flower buds at the apices of cucumber plants. These results suggest that the ERAF17 may be involved in the induction of female flower formation in cucumber plants. The isolated full length clone of ERAF17 was revealed that ERAF17 encoded both a MADS box domain and a K box domain.

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EFFECTS OF GIBBERELLIN ON SEX EXPRESSION AND ON EXPRESSION OF ACC SYNTHASE GENES (*CS-ACS1G* AND *CS-ACS2*) IN *CUCUMIS SATIVUS* L.

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Sex of flowers in cucumber plants is primarily regulated by level of ethylene at the apex. It is suggested that the level of ethylene at the apex is regulated by levels of transcripts of two ACC synthase genes (*CS-ACS1G* and *CS-ACS2*). We previously showed that the *CS-ACS2* expressed at apices of monoecious cucumber line and that both the *CS-ACS1G* and the *CS-ACS2* expressed at apices of isogenic gynoecious line. On the other hand, gibberellin induces the development of male flowers. It is suggested that gibberellin acts more upstream than ethylene on the regulation of sex expression. Therefore, we examined effects of gibberellin on the expression of *CS-ACS1G* and *CS-ACS2* at apices of monoecious and gynoecious lines. The treatment of monoecious line with gibberellin (GA₄) increased in a proportion of male flowers. The dose-dependent decrease in the expression of *CS-ACS2* transcript was observed at the apices of monoecious line that were treated with gibberellin. These results suggest that gibberellin acts on the regulation of sex expression by suppressing the expression of *CS-ACS2* transcript. Although the expression of *CS-ACS2* transcript was decreased by the treatment of apices of gynoecious line with gibberellin, the level of *CS-ACS1G* at the apices of gynoecious line was not affected by the treatment with gibberellin.

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CLONING AND CHARACTERIZATION OF AUXIN-RESPONSIVE ACC SYNTHASE GENES FROM MELON

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We have isolated two auxin-responsive ACC synthase genes (*CMe-ACS2* and *3*) from melon, and determined their nucleotide sequences. The 5'-flanking region of *CMe-ACS2* contained putative auxin responsive *cis*-elements resembling TGTCTC element of *GH3* gene from soybean and an auxin responsive *cis*-element (AuxRD B) of *PS-IAA4/5* from pea. We constructed chimeric gene consisting of a GUS gene directed by a fragment of *CMe-ACS2* promoter, and introduced into *Arabidopsis*. A 500bp fragment of *CMe-ACS2* promoter (containing AuxRE) was enough to increase GUS activity by NAA (50 μ M) treatment in transgenic *Arabidopsis*.

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ACC SYNTHASE, LE-ACS2, IS ACTIVATED BY PHOSPHORYLATION OF A SERINE RESIDUE

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ACC synthase (ACS) is a key enzyme in the pathway of ethylene biosynthesis. Some recent reports have pointed out the possibility that ACS is regulated not only transcriptionally but also posttranslationally. To elucidate how ethylene biosynthesis is regulated by ACS at the posttranslational level, we analyzed the modification of the ACS proteins in tomato (*Lycopersicon esculentum*) fruits using an anti-LE-ACS2 antibody.

We detected a phosphorylated LE-ACS2 by immunoprecipitation from the extract of wounded tomato fruits to which were fed ³²P-phosphorous. Analysis of phosphoamino acids showed that a serine residue was phosphorylated. Dephosphorylation of ACS by phosphatase reduced its enzymatic activity by 30%. These results indicated that LE-ACS2 was activated by phosphorylation of a serine residue.