



SDI Review Form 1.6

Journal Name:	American Journal of Experimental Agriculture
Manuscript Number:	2013_AJEA_7285
Title of the Manuscript:	A Tomato Inducer of CBF Expression 1 (SlICE1) is Involved in Cold Stress Signaling
Type of the Article	

General guideline for Peer Review process:

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PART 1: Review Comments

	Reviewer's comment	Author's comment (if agreed with reviewer, correct the manuscript and highlight that part in the manuscript. It is mandatory that authors should write his/her feedback here)
Compulsory REVISION comments	(1) Stockinger E. J., Gilmour S. J., & Thomashow M. F. (1997). can be cited in this work (2) Two abiotic stresses (cold and salinity) were studied but the title reflects only cold stress. Why? Even if much was not done on salt stress I think the title should heed of it. (3) Where did you grow your tomato plants?	We deeply appreciated detailed comments and informative suggestions raised by the reviewer #1 on our manuscript. We have carefully revised the manuscript according to suggestions made by reviewers, as follows. 1) We agree with a comment pointed by reviewer#3 about an additional reference. Thus, the sentence was revised as following. “Molecular genetic studies focusing on cold-stress-induced genes unveiled that dehydration-responsive-element-binding protein (DREB) / C-repeat-binding factor (CBF) genes are key transcription factors which are responsible for gene regulation network under cold and drought stresses (Stockinger et al. 1997; Shinozaki and Yamaguchi-Shinozaki 2000; Zhang et al. 2004).” 2) 1) We agree with a comment pointed by reviewer#3 about the title. Thus, it was changed as following. “A Tomato Inducer of CBF Expression 1 (SlICE1) is Involved in Cold and Salt Stress Signaling” 3) Growth condition was revised as following. “, tomato plants were grown for 4 week in phytotron (25oC) at glass house in Kyushu University as previously described (Yuasa et al. 2012).”
Minor REVISION comments	(1) In material and methods (line 92) the authors should	We fully agree with the indicated matter by



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	write sowing instead of sawing. (2) In results and discussions Wan et al. (2007), Martin and Busconi (2001)(line 183) were cited but not written in the references. (3) In Line 210, Chinnusamy et al. (2010) was cited but not written in the reference section. (4) Tomikubo et al. (2007) [line 213] was cited but not in the references. (5) Increase of line 220 should not be in capital letter. (6) In figure 2 (B) it should be in response to cold stress instead of chold stress. (7) It is 25 KDa not K252a (8) In line 413 it should be "Introduced restriction sites are showed (instead of show) by underlines (9) From line 179 to line 181 the results obtained and analyzed were from figure 2B and Fig 2C respectively not from Figure 2A and 2B as written by the authors. (10)In fig 2A you wrote IICE1, do you mean SLICE1 to be checked over? ICE1 should not be confounded to SLICE1	reviewer #3. We thoroughly corrected words and sentences including errors pointed by reviewer#2 in revised manuscript as follows. (1) The part of "After sawing tomato seeds" was corrected to "After sowing tomato seeds". (2) The reference of "Wan B, Lin Y, Mou T (2007) Expression of rice Ca²⁺-dependent protein kinases (CDPKs) genes under different environmental stresses. FEBS Lett. 581: 1179-1189" was added to references. (3) The part of "Chinnusamy et al (2010)" was corrected to ""Chinnusamy et al. (2003) as cited in References. (4) The reference of "Tomikubo Y, Yuasa T, Iwaya-Inoue M (2007) Analysis of chilling-induced trehalose-6-phosphate synthase (TPS) in tomato plants. Cryobiol Cryotechnol 53: 95-100" was added to references. (5) The part was corrected to " The part was corrected to "RT-PCR, increase of SLICE1 protein". (6) The part in Fig. 2B was corrected to "of tomato ICE1 protein levels in response to cold stress." (7) "Kinase inhibitor" was changed to "Ka252a" and 52kDa at left of panel was left. (8) The part was corrected to "Introduced restriction sites are showed by underlines". (9) The part was revised as following, "When tomato leaves were subjected to cold stress, induction of ICE1 protein levels was not observed in the presence of EGTA (Ca²⁺ chelater) and weak signal was induced in the presence of LaCl3 (a Ca²⁺ channel blocker).
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		On the other hand, cold stress significantly enhanced the tomato ICE1 protein level in the presence of Ca²⁺ as expected (Fig. 2B). In addition, protein kinase inhibitor (K252a) treatment suppressed the cold stress-induced upregulation of the tomato ICE1 proteins (Fig. 2C, left).". (10) The signal at open arrowhead was mentioned as "ICE1-related polypeptide" in the Figure legend.
<u>Optional/General</u> comments	It Is OK with the introduction Methodologies described in this manuscript are sufficient The authors need to work on Results and discussion to correct some mistakes. The conclusion is supported and based on the data obtained	