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Antagonism between abscisic acid and gibberellins is partially mediated by ascorbic acid during seed germination in rice

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The antagonism between abscisic acid (ABA) and gibberellin (GA) plays a key role in controlling seed germination,[1,2] but the mechanism of antagonism during this process is not known. In the associated study,[3] we investigated the relationship among ABA, reactive oxygen species (ROS), ascorbic acid (ASC) and GA during rice seed germination. ROS production is reduced by ABA, which hence results in decreasing ASC accumulation during imbibition. GA accumulation was also suppressed by a reduced ROS and ASC level, whereas application of exogenous ASC can partially rescue seed germination from ABA treatment. Further results show that production of ASC, which acts as a substrate in GA biosynthesis, was significantly inhibited by lycorine which thus suppressed the accumulation of GA. Consequently, expression of GA biosynthesis genes was suppressed by the low levels of ROS and ASC in ABA-treated seeds. These studies reveal a new role for ASC in mediating the antagonism between ABA and GA during seed germination in rice.

Seed germination is a complicated process, which is controlled by many plant hormones.[4] Among them, the antagonism between ABA and GA plays a key role in controlling seed germination.[5,6] Little is known about the mechanism of this antagonism in rice seed during germination. Previous works have shown that ROS plays a key role in seed biology.[7-9] Different studies have proved ROS is a critical component in ABA signaling pathway in leaves and interacts with GA during seed germination,[10,11] indicating that ROS could be good candidates in mediating the ABA and GA antagonism.

ASC was discovered in the late 1920s. Although it is well recognized as a powerful antioxidant,[12] the question 'what is the function of ASC?' is still far from being answered.[13,14] In orthodox seeds, neither ASC norASC peroxidase activity exist at the quiescent stage, but they re-start after a few hours from the onset of imbibition, which is suggested to be crucial to ensure seed germinability.[15-17] Recently, ASC was found to act as a substrate for many 2-oxoacid-dependent dioxygenases (2-ODDs), which are involved in the synthesis of the plant hormones ethylene, GA, and ABA.[18,19] Both of these two properties indicate a possible function during seed germination except the antioxidant role.

Here in the associate study, we also showed that ROS production is reduced especially in the embryo region by a relative high level of ABA during germination in rice seed. As a result, the ASC production is reduced by the perturbed redox state in the imbibing seeds. The suppression is not only found in the treatments of ABA, but also in the treatments of DPI, a ROS scavenger, and lycorine which is of inhibitor of ASC biosynthesis (**Fig. 1A**), further suggesting that ABA suppresses ASC production is mediated by a reduced ROS production during imbibition in rice seed. Interestingly, when ASC was applied to imbibing seeds treated with ABA, the inhibition of seed germination by ABA is partially rescued by 1mM ASC application, indicating that ASC is involved in the process that ABA inhibits seed germination.

It is suggested that suppressing the ASC content during seed germination will lead to other consequences, because ASC is much more than just an antioxidant.[13,20] To answer this question, an inhibitor, lycorine, was used since this alkaloid has been proved to be a strong inhibitor of (l-galactono-c-lactone) GaL dehydrogenase (EC 1.3.2.3), the last enzyme of the ASC biosynthetic pathway, both in vivo[21-23] and in vitro,[24] was applied to the imbibing seed in rice. Consequently, seed germination was inhibited by lycorine, which is similar to the inhibition effect of ABA and DPI (**Fig. 1B**). This result further proves that ASC plays a role in ABA signaling pathway during seed germination. In agreement with our result, a recent work has revealed that low ASC triggers ABA- and jasmonate-dependent signaling pathways that together regulate growth through ABI4.[25]

Since the GA accumulation which is indicated by the expression of GA biosynthesis genes and amylase activities, similarly to ASC production, is reducedby ABA and DPI treatments during seed germination. It is quite possible that ASC plays a key role in mediating the antagonism between ABA and GA during seed

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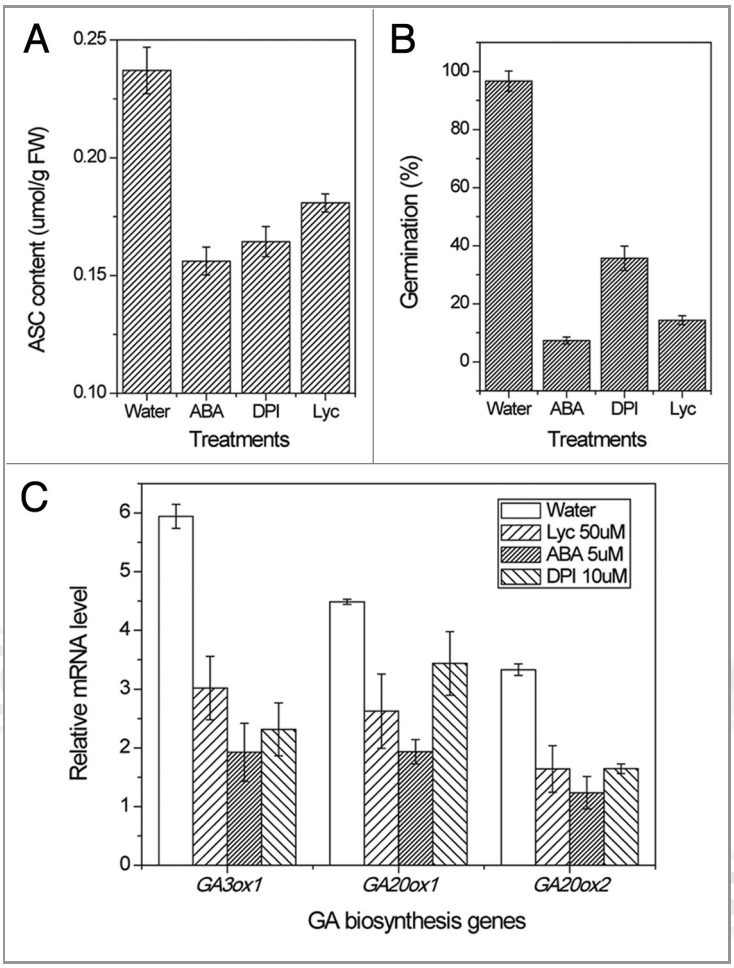

Figure 1. Inhibition of ASC production (A) and the consequent effect on seed germination (B) and GA accumulation (C). Rice seeds were imbibed at 28 °C in the presence of water, ABA, diniconazole, Tiron, DPI and lycorine solutions for 36h. Seed samples were collected and stored at –80 °C for ASC determination and QRT-PCR. Values are means ± SD (n = 5).

germination. Indeed, exogenous application of lycorine reduced the expression of key genes in the GA biosynthesis pathway in seeds treated with lycorine, which is similar to treatment with ABA and DPI (**Fig. 1C**). The similar effect of ABA and lycorine on GA accumulation has revealed that antagonism between abscisic acid and gibberellins is partially mediated by ascorbic acid during seed germination in rice.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

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