

Puroindoline: Antimicrobial wheat endosperm specific protein

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During the hydrated part of seed life, i.e. maturation, germination, that is very sensitive to virus, fungi and bacteria. Many proteins involved in the microbial defense mechanism of plants. Plant seeds contain high concentration of many antimicrobial proteins. Puroindolines (13 kDa) are the most abundant proteins isolated from wheat endosperm by phase partitioning with the nonionic detergent Triton X-114. Puroindoline is the main component of a new family of proteins that has been suggested to exert an antimicrobial activity in plant seeds. Antimicrobial peptides play a role in the immune systems of animals and plants by limiting pathogen infection and growth. Puroindoline may also be a membranotoxin that might play a role in the defense mechanism of plants against microbial pathogens. In this review, properties and importance of Puroindoline protein is proposed. Thus, deserve further studies aimed at establishing their possible future application in the field of food and health care.

Key words: Wheat, Puroindoline, Triton X-1 14, Antimicrobial activity

Introduction

A new basic protein has been isolated from wheat endosperm by Triton X-1 14 phase partitioning. This protein isolated from wheat endosperm has been named puroindoline. Puroindoline is protein associated to starch, either by synthesis or because their location in wheat grain endosperm. The puroindoline, endosperm-specific proteins involved in wheat seed hardness, are small proteins reported to have in vitro antimicrobial properties. Puroindoline may also be a membranotoxin that might play a role in the defense mechanism of plants against microbial pathogens. Plants and animals produce antimicrobial peptides as part of their natural defense systems to control disease-causing microorganisms.

These peptides may act against bacteria, fungi and plants contain numerous antimicrobial peptides that may be involved in protecting the plant from pathogens (Krishnamurthy, *et al.*, 2001) during maturation and

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germination, seeds are very sensitive to viruses, fungi and bacteria (Blochet *et al.*, 1993). Many proteins involved in the microbial defense mechanism of plants have been identified and some of them display *in vitro*, an effective antimicrobial activity (Collinge and Slusarenko, 1987 and Dixon and Harrison, 1990). Thionins were the first antimicrobial proteins to be isolated from wheat endosperm half a century ago and they have been subjected to several studies. (Garcia-Olmedo, *et al.*, 1989). Puroindolines are effective *in vivo* in antimicrobial proteins and could be valuable new tools in the control of a wide range of microbial pathogens of crop plants.

Antibacterial and hemolytic activities

Like many other antimicrobial peptides, puroA could potentially be developed into a new antibiotic. Furthermore, the antimicrobial activity of puroindoline may be improved by mutating the corresponding gene sequences encoding puroA. *Escherichia coli* and *Staphylococcus aureus* were used to determine the antimicrobial activities of puroA and puroB. The MICs of puroA for *E. coli* and *S. aureus* were 7 and 16 μM , respectively, indicated that puroA exhibited activity against both gram-positive and gram-negative bacteria. The MICs of puroB for *E. coli* and *S. aureus* were over 200 μM . The hemolytic activities of the puroA and puroB peptides were shown extremely low concentration of 50% hemolysis, >1,000 $\mu\text{g/ml}$ (Weiguo Jing, *et al.*, 2003).

Grain Hardness (Kernel texture)

Grain hardness (kernel texture) is one of the most important end use quality characteristics of cultivated common wheat. As it has a profound effect on milling, processing and utilization (Hiroyuki Tanaka, *et al.*, 2008) Based on the texture of the mature kernel, common wheat varieties are typically classified as hard or soft. Soft wheat varieties are used for cakes, cookies, pastries and some type of noodles, whereas hard wheat varieties are used for breads and other yeast-leavened foods (Morris and Rose, 1996). To investigate a direct causal relationship between puroindolines and grain hardness, rice, which has no homologues to the puroindolines (Gautier *et al.*, 1994) that was transformed with wild – type puroindolines genes (Krishnamurty and Giroux, 2001). Textural analysis of the transgenic rice seeds indicated that expression of wild-type Puroindolines genes reduced rice grain hardness. These data supported the hypothesis that puroindolines play an important role in conterling kernel hardness.

Genetic Diversity

The SDS-PAGE Profile of endosperm protein of eight varieties was performed. Analysis of puroindoline protein from eight varieties suggested that endosperm protein is a determining factor of finding difference and similarities between these varieties. Significant correlations between these varieties were detected due to similar band patterns, concentration of puroindoline protein, with the help of puroindoline protein and the diversity among wheat varieties (Vinod Kumar Dhatwalia, *et al.*, 2008).

Transgenic Plants

It is interesting that a protein might be involved in both the protection of wheat seeds from fungal pathogens and a seed quality property like seed hardness (Krishnamurthy *et al.*, 2001). Transformation of rice with the *pin* genes, which normally have no puroindolines, concomitantly increased seed softness (Krishnamurthy and Giroux 2001) and resistance against two important fungal pathogens. Presumably, the presence of puroindolines in wheat evolved to protect seeds from fungal pathogens but it was selected to against by breeders to produce softer or harder seeds as a result of this secondary property of the puroindoline proteins. Wheat puroindolines were effective *in vivo* antifungal proteins in rice, which normally does not produce these proteins. The two most devastating fungal diseases of rice were controlled at significant levels. There is no reason to suspect that similar results could not be achieved with other fungal pathogens of other important crops.

Conclusion

Recent investigations have demonstrated that puroA have good antimicrobial activity and puroindoline protein has direct causal relationship with grain hardness. The puroindolines are normally found in wheat endosperm and thus are routinely ingested by humans and animals alike, their use in transgenic crops may be less objectionable than gene products, normally not in plants or in the edible portion of plants. This work may help to improve the efficiency of wheat and other crops breeding programs in cultivar development. In this regard, puroindoline protein and humankind enjoy an interesting interspecific relationship.

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