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ORIGINAL PAPER

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# Differential accumulation of dehydrin-like proteins by abiotic stresses in *Deschampsia antarctica* Desv.

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**Abstract** Dehydrins are proteins that accumulate during environmental stresses leading to cell dehydration. *Deschampsia antarctica* is one of the two vascular plants that have colonized the Maritime Antarctic. This plant is usually exposed to cold, salt and desiccating winds in the field. We proposed that among the factors that allow *D. antarctica* to survive the harsh environmental conditions is the presence of dehydrins. We studied the accumulation of dehydrins by abscisic acid (ABA), dehydration, NaCl and low osmotic potential. Western blots using an anti-dehydrin antibody revealed a complex pattern of dehydrin-like proteins (DLPs) accumulation in the different treatments. DLPs with apparent molecular weight of 58, 57, 55, 53, 48, 42, 32, 30, 28 and 25 kDa were detected in the different treatments. DLPs accumulation was associated with a decrease in the relative water content (RWC) of the plants. These results suggest that DLPs accumulation could contribute to explain how *D. antarctica* can survive under adverse Antarctic conditions.

## Introduction

Drought, high salinity, and low temperature are the most common environmental stress factors that influence the plant growth and development and place major limits on plant productivity in cultivated areas worldwide (Boyer 1982; Thomashow 1994). Plants react to such environmental stresses by initiating a number of physiological and developmental changes, which in turn require altered gene expression. Different stresses may induce similar mechanisms of resistance. For example, low temperature and drought result in mechanical constraints, changes in activities of macromolecules, and reduced osmotic potential in the cellular milieu. High salinity includes ionic (chemical) and osmotic (physical) components (Thomashow 1998; Xiong et al. 2002). Therefore, it is not surprising that plants respond similarly to low temperature, drought, and salinity at the molecular level (Shinozaki and Yamaguchi-Shinozaki 2000).

The expression of numerous genes is altered in response to low temperature, osmotic stress and high salt in plants (Skriver and Mundy 1990; Zhu et al. 1997; Hsieh et al. 2002; Xiong et al. 2002; Zarka et al. 2003). Both osmotic and cold stresses increase the level of the phytohormone abscisic acid (ABA) (Ishitani et al. 1997). The expression of many osmotic and cold stress-responsive genes can be induced by application of ABA (Skriver and Mundy 1990; Zhu et al. 1997).

Dehydrins (LEA D-11) constitute one of the typical families of proteins that occur in plants in response to dehydration, low temperature, osmotic stress, salt stress, seed drying and exposure to abscisic acid (Close and Lammers 1993; Close 1997; Richard et al. 2000; Parmetier-Line et al. 2002). The most distinctive motif of dehydrins is a highly conserved lysine-rich 15-amino acid sequence (consensus sequence EKKGIMDKIKEKPLG), referred to as the K segment, which is often repeated several times within the polypeptide (Close 1996). Although there is no clear understanding of the entire sequence of events, dehydrins are thought to play a major role in protecting plants from dehydration. This is based on their presence in many plants species, hydrophilic character, possible compatible solute-like properties, and accumulation correlated with cold and drought tolerances (Close 1996; Bravo et al. 1999). Cold-induced dehydrins have been shown to

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cryoprotect freezing-labile macromolecules in vitro (Bravo et al. 2003). A dehydrin induced by low temperature exposure in the bark of peach trees exhibits antifreeze activity (Wisniewski et al. 1999). Indeed, there is an emerging evidence for the participation of dehydrins in the stabilization of cell membranes against dehydration-induced or freezing-induced injury (Thomashow 1999; Koag et al. 2003).

Most studies on dehydrins have been carried out in crop plants or *A. thaliana*. Only few studies have been made in wild species from dry or cold regions (Sauter et al. 1999; Ndima et al. 2001). *Deschampsia antarctica* Desv. (Poaceae) is one of the two vascular plants that have naturally colonized the Maritime Antarctic (Edward and Lewis-Smith 1988; Casaretto et al. 1994). *D. antarctica* is found in the South Orkney Islands and in most of the Maritime Antarctic down to approximately 68°S, but has not extended into the continental Antarctic (Alberdi et al. 2002). In the proximity of the coast (Maritime Antarctic) temperatures ameliorate. During summer, plants are subjected to frequent diurnal freeze-thaw cycles. The air temperatures in coastal Antarctic regions seldom exceed 5°C (mean temperature during summer is 1°C). Occasionally the air temperature may reach 10–15°C but only for brief periods. In winter (mean temperature during winter is −4°C), snow accumulation protects the vegetation for 6–7 months each year. Such stands are seldom exposed to very low temperatures, but plants in more windswept field sites may occasionally experience extreme winter temperatures of −25°C to −35°C (Lewis-Smith 2003). Antarctic soils, especially those of the coastal areas, are characterized by a high content of coarse mineral particles and low total organic carbon, a low C/N ratio, acidic pH, and are frequently enriched with nutrients due to the influence of sea spray and input of seabirds (Alberdi et al. 2002).

Therefore, it has been suggested that *D. antarctica* has developed metabolic adaptations to survive the harsh conditions of the Maritime Antarctic, especially during the growing season (Bravo et al. 2001). *D. antarctica* does not have unusual contents of total polar lipids or high degree of unsaturation of fatty acids compared with other Poaceae (Zúñiga et al. 1994). High amounts of sucrose and fructans are found in this plant mainly toward the end of summer under field conditions (Zúñiga et al. 1996). This is consistent with the findings that *D. antarctica* has relatively high net photosynthetic rates on cold days (Xiong et al. 1999). At 0°C, this plant maintains about 30% of the photosynthetic rate found at the optimum temperature (Edwards and Lewis-Smith 1988). *D. antarctica* is usually exposed to cold, salt spray from the sea and desiccating winds in the field. For this reason, it has been proposed that dehydrin-like proteins (DLPs) are part of *D. antarctica*'s responses to this complex interaction of stresses (Olave-Concha et al. 2004). The objective of this work was to further study changes in DLPs accumulation in response to dehydration, salt, osmotic stress and ABA application.

## Materials and methods

### Plant material

*Deschampsia antarctica* Desv. (Poaceae) was collected from the Coppermine Peninsula on Robert Island, Maritime Antarctic (62°22′S: 59°43′W). Plants were transported in plastic bags to the laboratory. *Deschampsia antarctica* was reproduced vegetatively in plastic pots using a soil: peat mixture (2:1) and maintained at 15 ± 1°C in a growth chamber with a photon flux density of 150 μmol $m^{-2}$ $s^{-1}$ and 16 h/8 h light/dark period. Plants were fertilized with Phostrogen (Solaris, Buckinghamshire, UK), using 0.12 g $l^{-1}$ once every 2 weeks.

### Dehydration, osmotic, salt and ABA treatment

For all treatments, plant roots were washed thoroughly to remove soil. Dehydration stress was simulated as observed in nature, where some rooted tillers are dislodged from the parental tussocks by the action of water streams and strong winds. Therefore, plants with naked roots remain on the surface of the soil. Under laboratory conditions dehydration was imposed by holding the plant with naked roots on a layer of humid vermiculite and exposing it to air. During dehydration stress, the water status was measured as relative water content (RWC). For other stresses, plants were placed in pots with vermiculite. For salt stress, NaCl was added to the vermiculite to final concentrations of 0.25, 0.5, 0.75, and 1 M. For osmotic stress, polyethylene glycol (PEG) 15,000–20,000 was added to the vermiculite to reach a final concentration of 13.5% w/w (−0.3 MPa), 19% w/w (−0.6 MPa), 23% w/w (−0.9 MPa), and 25% w/w (1.2 MPa). ABA treatment was carried out with 100 μM of (±) ABA isoform. The hormone solution was added to the vermiculite. Pots were covered with aluminum foil to avoid light-induced isomerization of ABA. All treatments were for 4 days, with the exception of ABA treatment, that was done only for 24 h. For Western blot analysis, two samples of 0.1 g fresh leaf tissues were collected in duplicates from two independently treated plants during a time course. For RWC analysis, 30 homogeneous plants were taken for each time point during the experiment.

### Protein extraction and immunoblot analysis

Leaf material from control and treated plants was frozen in liquid nitrogen, and powdered using a mortar and pestle. The frozen powder was transferred directly to the extraction buffer (60 mM Tris-HCl, pH 6.8; 100 mM NaCl) and boiled for 5 min, then chilled on ice and centrifuged. The supernatant was transferred to a fresh tube and stored at −20° C.

Total protein content was determined by the Bradford assay (Bradford 1976). Ten micrograms of protein from each sample were separated by SDS-PAGE in 10% gel and stained with colloidal Coomassie brilliant blue G-250 (Neuhoff et al. 1988). The separated polypeptides were transferred onto a nitrocellulose membrane for 30 min at 450 mA. After transfer, the membrane was blocked with 10% (w/v) dry non-fat milk in Tween Tris-buffered saline (TTBS) (0.1% (w/v) Tween-20 in TBS; 20 mM Tris-HCl, pH 7.5; 500 mM NaCl) for 1 h. After blocking, the membrane was incubated overnight at 4°C with the first antibody, a polyclonal antiserum raised against the dehydrin consensus peptide, also called K segment (EKKGIMDKIKEKLPG), at a dilution of 1:1,000 in TBS (Close and Lammers 1993). After three consecutive washes of 20 min each in TTBS, the membrane was incubated for 1 h at 4° C with the secondary antibody, anti-rabbit IgG raised in goat and conjugated to alkaline phosphatase at a dilution of 1:1,000 TBS. After three washes in TTBS, the membrane was developed by incubating it for 5 min in 0.4 mM nitroblue tetrazolium chloride (NBT) and 0.4 mM 5-bromo-4-chloro-3-indolyl phosphate (BCIP) in developing buffer (100 mM Tris, pH 9.0; 100 mM NaCl; 5 mM $MgCl_2$).

### Physiological parameters

Relative water content (RWC) was determined in whole plants under dehydration. RWC was evaluated according to the method of Barrs and Weatherley (1962) and was based on the following calculation: RWC = (FW−DW)/(SW−DW)×100, where FW is plant fresh weight, DW is the weight of plants after drying at 85°C for 4 days, and SW is the turgid weight of plants after soaking in water for 4 h at room temperature. The excess water was removed with absorbent paper before the turgid weight was determined.

### Fluorescence measurements

After 4 days of treatment with different concentration of NaCl, plants were removed from the chamber for fluorescence measurements at room temperature. Fluorescence signals were generated by a saturated pulse by plant efficiency analyzer (PEA, Hansatech Instruments, Norfolk, UK). Fully developed attached leaves from treatment and control plants were adapted to dark for 30 min (to obtain open PSII centers) using the instrument leaf-clips to ensure maximum photochemical efficiency. The light intensity of saturating pulses was 2,250 μmol $m^{-2}$ $s^{-1}$. The recording was performed for 40 s. Data analyses were performed with the software provided with the instrument.

### Statistical analysis

Values reported correspond to the means of three independent assays. A one-way ANOVA, followed by a Tukey test, was used for Fv/Fm and RWC analyses ($P \leq 0.05$).

## Results

### Effect of dehydration treatment on dehydrins accumulation

Immunoblots indicated that DLPs with $M_r$ of 58, 53, 48, and 30 kDa were induced by dehydration treatment showing different accumulation kinetics. Only the 58 kDa polypeptide was detected after 10 h of dehydration; the 48 and 30 kDa polypeptides were both detected a day after the treatment. The 30 kDa polypeptide remained high even after 4 days of dehydration while 48 kDa band decreased. Finally, a 53 kDa band was detected only after 2 days of dehydration. Interestingly, all DLPs reached their maximum accumulation after 2 days of dehydration, decreasing afterwards (Fig. 1A). RWC decreased from 0.76 to 0.24 during water stress treatment (Fig. 1B). Statistical analysis showed significant difference ($P < 0.05$) in the RWC after 10 h of dehydration stress with respect to control. This significant decrease in RWC was concomitant with the appearance of the 58 kDa DLP (Fig. 1A, B).

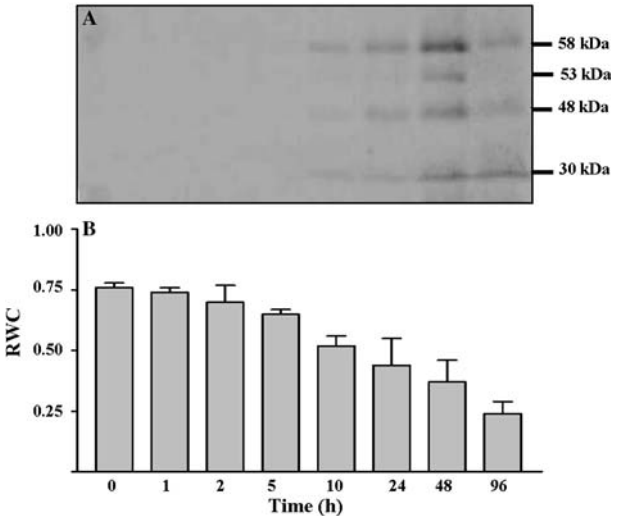



**Fig. 1** Effect of drought on dehydrins accumulation in *D. antarctica*. **A** Immunoblot of dehydrins from dehydration stress *D. antarctica*. Total protein was extracted at 0, 1, 2, 5, 10, 24, 48 and 96 h after dehydration stress. The *lines* indicate proteins (their size, in kDa) recognized by anti-dehydrin antiserum. **B** Effects of dehydration stress on relative water content (*RWC*) in whole plants of *D. antarctica*. Bars represent standard error of the mean ($n = 3$)

### Effect of PEG (osmotic stress) on dehydrins accumulation

The response of *D. antarctica* to PEG was studied by treating plants with different osmotic potentials, −0.3, −0.6, −0.9, and −1.2 MPa for 4 days. The anti-dehydrin antiserum detected seven DLPs with $M_r$ 58, 55, 53, 48, 32, 30, and 28 kDa. All osmotic potentials tested resulted in increased levels of DLPs (Fig. 2). The dehydrin band pattern varied with the intensity of osmotic stress. Higher apparent molecular mass polypeptides (58, 53, and 48 kDa) were present at −0.3 and −0.6 MPa treatments (Figs. 2 A, B, E, F). However, the accumulation of these proteins was higher at low osmotic potential treatments (Figs. 2 C, D, G, H). Lower apparent molecular mass polypeptides (32, 30, and 28 kDa) were only present at lower osmotic potentials, −0.9 and −1.2 MPa (Fig. 2C, D).

### Effect of salt (NaCl) treatment on dehydrin accumulation

The level of DLPs in *D. antarctica* growing under different concentration of NaCl (0.25, 0.5, 0.75, and 1 M) was also analyzed by immunoblotting. The anti-dehydrin antibody recognized eight polypeptide bands. Salt treatments led to different accumulation pattern of DLPs. The protein with Mr 58 kDa was the only observed band after a day of treatment at 0.25 M NaCl. Exposure of plants to higher concentrations of NaCl produced an early and increased accumulation level of this polypeptide (Fig. 3D, E, G, H). DLPs with $M_r$ 55, 53, 48, 30, and 28 kDa were detected during treatments with 0.5, 0.75, and 1 M NaCl (Fig. 3B–D). The proteins with smaller size (30 and 28 kDa) were detected after 2 days, 1 day and 10 h in plants treated with 0.5 M, 0.75, and 1 M NaCl, respectively (Fig. 3B–D). There was an additional inconspicuous band of 25 kDa that was observed only upon treatment with 1 M NaCl (Fig. 3D). To investigate the influence of salt stress on the physiological state of *D. antarctica*, fluorescence of PSII was used to determine Fv/Fm. The Fv/Fm values did not show significant differences ($P \leq 0.05$) between control (Fv/Fm = 0.81) and NaCl treated plants, where Fv/Fm was 0.82 (0.25 M), 0.76 (0.50 M), 0.77 (0.75 M), and 0.77 (1 M), respectively. These results suggest that even 1 M NaCl did not have a dramatic damaging effect on the status of the photosynthetic electron transport machinery of *D. antarctica*.

### Effect of ABA on dehydrins accumulation

Abscisic acid treatment resulted in significant accumulation of DLPs in *D. antarctica*. ABA treatment led to accumulation of eight polypeptide bands with different levels of accumulation and kinetics. Four polypeptides, 58, 53, 48, and 28 kDa were the most intense bands

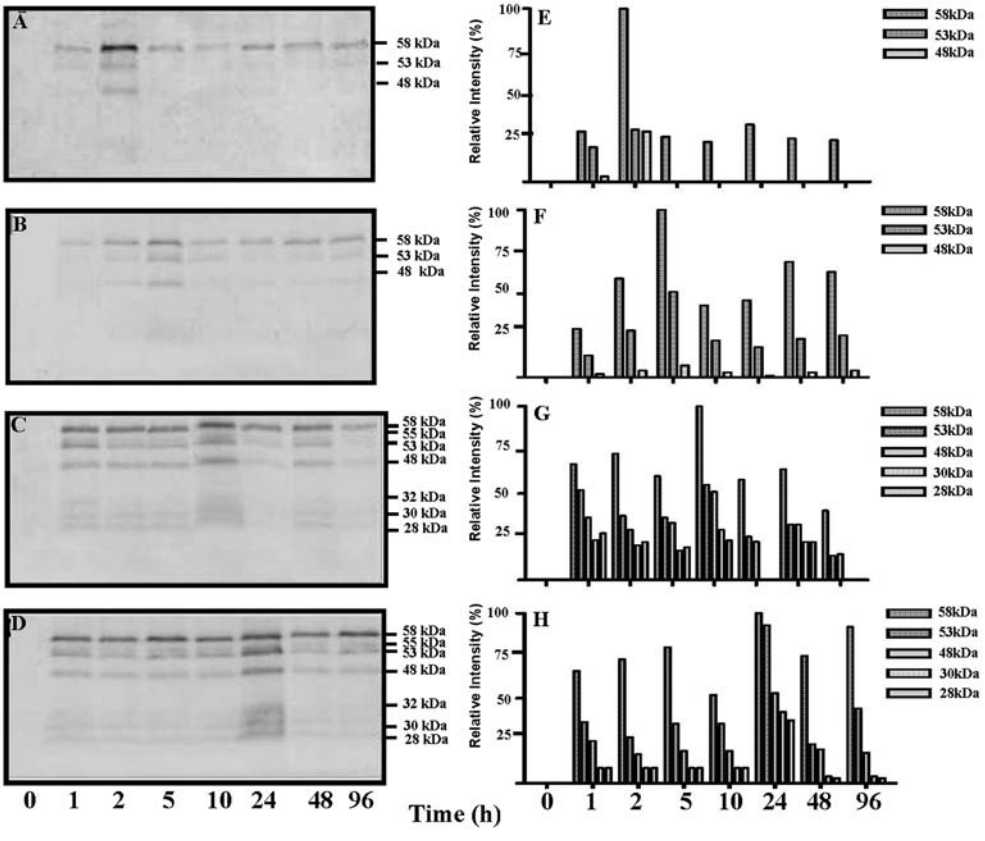



**Fig. 2** Effect of osmotic potential on dehydrin accumulation in *D. antarctica*. Immunoblot of dehydrins from PEG-treated plants were done by collecting leaves of *D. antarctica* at 0, 1, 2, 5, 10, 24, 48 and 96 h of treatment with different osmotic potential: **A** −0.3 MPa, **B** −0.6 MPa, **C** −0.9 MPa and **D** −1.2 MPa. The *lines* indicate the apparent molecular mass in kDa for each polypeptide recognized by the antibody. **E–H** Relative intensities of the various DLPs obtained by densitometric analyses of the corresponding western blot

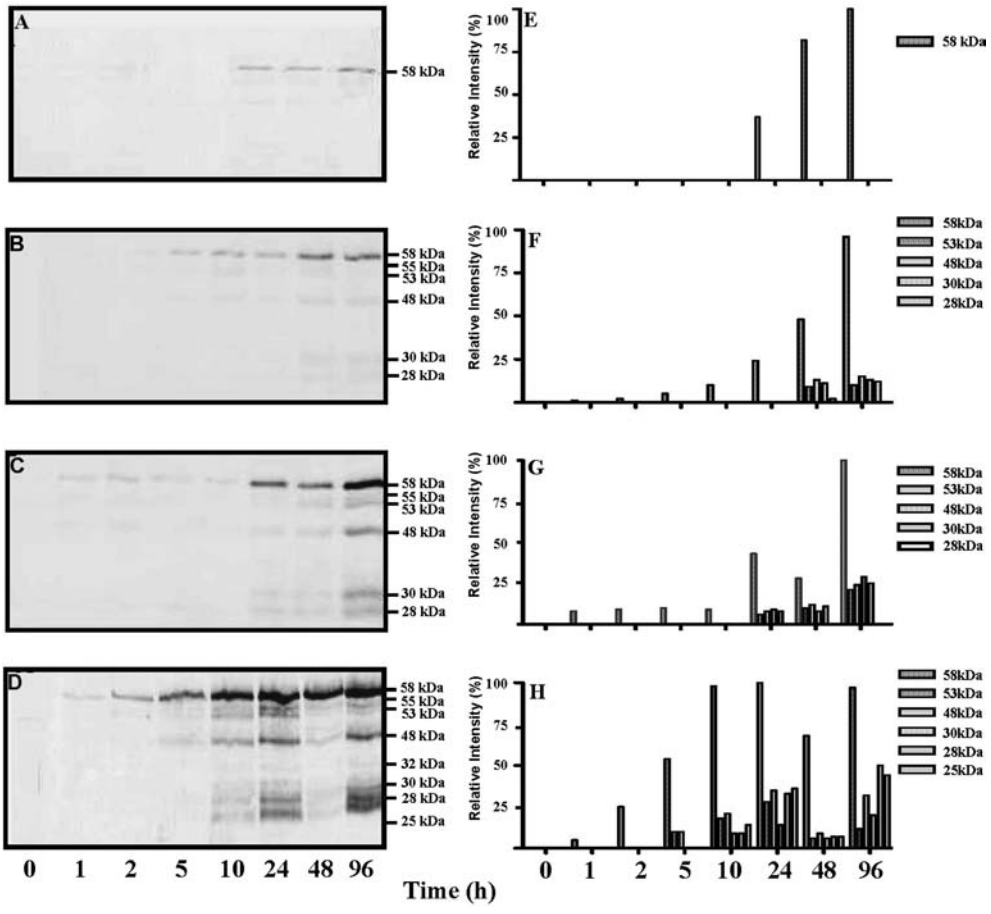


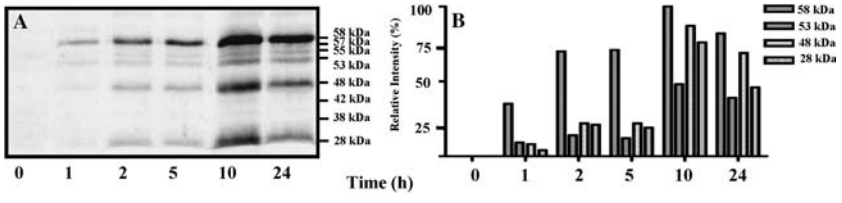

**Fig. 3** Effect of salt stress on dehydrins accumulation in *D. antarctica*. Immunoblot of dehydrins from NaCl-treated plants were done by collecting leaves of *D. antarctica* at 0, 1, 2, 5, 10, 24, 48, and 96 h of treatment with different concentration of NaCl: **A** 0.25 M, **B** 0.5 M, **C** 0.75 M, and **D** 1 M. The *lines* indicate the apparent molecular mass in kDa for each polypeptide recognized by the antibody. **E–H**) Relative intensities of the various DLPs obtained by densitometric analyses of the corresponding western blot

detected during ABA treatment (Fig. 4A, B). Three polypeptides (58, 53, and 48 kDa) were accumulated early, being detected after 1 h of treatment. Their accumulation reached a maximum after 10 h and slightly decreased after 24 h of treatment. Two polypeptides, 55 and 28 kDa were detected after 2 h, which also reached maximum after 10 h of ABA treatment. Three protein bands with Mr 57, 42 and 38 kDa were weakly detected after 10 h and remained at a similar level after 24 h of ABA treatment (Fig. 4A, B).

**Fig. 4** Effect of ABA application on dehydrin accumulation in *D. antarctica*. **A** Immunoblots of dehydrins from plants treated with 100 μM ABA . Extracts from leaves collected at 0, 1, 2, 5, 10, and 24 h of treatment were used. The *lines* indicate proteins (their size, in kDa) recognized by the antibody. **B** Relative intensities of the various DLPs obtained by densitometric analyses of the above western blot

## Discussion

The harsh environmental conditions experienced by plants in the Maritime Antarctic, such as constant low temperature, strong desiccating winds and sea spray, expose *D. antarctica* to natural combination of water, osmotic and salt stresses. In order to cope with these stresses *D. antarctica* has a variety of adaptive mechanisms (Alberdi et al. 2002). It is well established that dehydrins are a family of proteins that accumulate in a wide range of plant species under dehydration, salt and cold adaptation. Dehydrins are hydrophilic, and heat stable proteins, which may protect other macromolecules or cellular structures, and help in maintaining the physiological integrity of cells (Close and Lammers 1993; Bray 1997). In a previous work, we have found a family of DLPs induced by cold, salt and osmotic stresses and also by exogenous application of ABA (Olave-Concha et al. 2004).



In this work, we have detected 11 DLPs. Five of these polypeptides (58, 53, 48, 30, and 28 kDa) were found to accumulate under salinity, dehydration and osmotic stress. Interestingly, these five polypeptides were also accumulated in ABA treated plants, exhibiting an earlier and more intense accumulation. This suggests that they represent a common response to dehydrating forces probably associated with an ABA dependent signal transduction pathway. Dehydration-induced polypeptides have been observed in many studies (Bewley et al. 1993; Perez-Malphe-Balch et al. 1996; Arora et al. 1998; Riccardi et al. 1998). In this study, DLPs with polypeptides ranging from 30 kDa to 58 kDa, especially 58 and 48 kDa, were present, and their intensities increased with progressive water deficit when plant RWC dropped from 52% to about 37% (Fig. 1). These results indicated that DLP accumulation was induced strongly by dehydration stress. Nonetheless, severe dehydration caused a decrease in the DLPs accumulation probably due to damage. Jiang and Huang (2002) also found that the accumulation of 23 and 27 kDa DLPs in tall fescue depends on the water status of the plant. Accumulation of dehydrins proteins could protect cells from further dehydration during drought stress (Han and Kermode 1996; Cellier et al. 1998). However, the extent of protection could be limited under severe dehydration, where damage to the protein synthesis machinery and cell structure may be generalized and irreversible (Jiang and Huang 2002).

Furthermore, there were three DLPs (57, 42, and 38 kDa) that were only slightly detected by exogenous treatment of ABA (Fig. 4). A possible explanation for this result may be the high levels of ABA used for the exogenous treatment (100 μM), which may have stimulated some specific tissues and genes that respond to higher levels of ABA than those reached under salinity, osmotic or dehydration stresses. Similar results have been observed in winter wheat which exhibited a 52 kDa dehydrin expressed only under exogenous ABA treatment (Borovskii et al. 2002). These authors also found that drought, freezing and exogenous ABA treatment result in accumulation of DLPs in rye, wheat and maize. These results corroborate an important role of ABA in plant responses to stress-related dehydration (Cellier et al. 1998; Giordani et al. 1999; Xiong and Zhu 2001; Jiang and Huang 2002).

Salt stress may induce osmotic adjustment balancing the lowered water potential of the cytoplasm caused by sodium sequestration inside the vacuole and concomitant water migration to it. Sodium can accumulate in the vacuole up to concentrations that may have marked effect on the osmotic balance of the plant cell (Adams et al. 1998). Therefore, it is not surprising that salt and osmotic stress have similar signal transduction pathways (Shinozaki and Yamaguchi Shinozaki 1997). This agrees with the fact that seven out of eight DLPs that accumulated upon salt treatment were also detected under PEG treatment in *D. antarctica* (Table 1). However, the 25 kDa polypeptide observed only under high salt concentration (0.75–1 M NaCl) and not under PEG treatment may be a specific response to the ionic effect of NaCl. Unexpectedly, two DLPs with Mr 55 and 32 kDa accumulated in *D. antarctica* under salt and osmotic stresses but not under dehydration treatment. This supports the idea that salt and osmolytes may induce some dehydrin accumulation response independent of their intrinsic dehydration effect in *D. antarctica*. Alternatively, it is likely that cell dehydration was more severe under high salt and PEG than under the dehydration treatment. This would be consistent with the high control capacity of water loss attributed to this species by several authors (Montiel et al. 1999; Mantovani and Vieira 2000; Romero et al. 1999; Alberdi et al. 2002). Furthermore, severe osmotic stress (−1.2 MPa) and high salt concentration (1 M NaCl) are artificial conditions to which the plant has not been allowed to adapt. Dehydration, on the other hand, is naturally occurring in the field. It is common to observe rooted tillers dislodged from the parental tussocks, by the action of water streams and strong winds, which remain on the soil surface with naked roots for several days. The importance of this process is that some plants tolerate this drought stress and survive unrooted periods and are able to recolonize in soil. Vegetative reproduction by detached parts of the parent plants is probably the most reliable and frequent mean of dispersal of *D. antarctica* (Lewis-Smith 2003).

The maximum photochemical efficiency of PSII, measured as the variable chlorophyll fluorescence and maximum fluorescence ratio (Fv/Fm) did not exhibit statistically significant differences in plants exposed to NaCl concentrations ranging from 0.25 M to 1 M NaCl. Therefore, potential PSII photochemical efficiency that normally decreases when PSII undergoes damage (Somersalo and Krause 1990) was unaffected even at high salt concentration, 1 M NaCl (sea water contains about 500 mM NaCl). Since photosystem II (PSII) is believed to play a key role in the response of photosynthesis to environmental perturbations (Baker 1991), the effects of salinity stress on PSII have been investigated extensively. However, the data collected on the effects of salinity stress

**Table 1** Detection of dehydrin-like proteins under different stresses and ABA treatment

| Treatment | 58 kDa | 57 kDa | 55 kDa | 53 kDa | 48 kDa | 42 kDa | 32 kDa | 30 kDa | 28 kDa | 25 kDa |
|---|---|---|---|---|---|---|---|---|---|---|
| ABA | + | + | + | + | + | + | – | + | + | – |
| SALT (NaCl) | + | – | + | + | + | – | + | + | + | + |
| DEHYDRATION | + | – | – | + | + | – | – | + | + | – |
| OSMOTIC (PEG) | + | – | + | + | + | – | + | + | + | – |

This summary is based on Figs. 1, 2, 3, and 4

on PSII photochemistry are conflicting. Some studies have shown that the salt stress inhibits PSII activity (Bongi and Lotero 1989; Mishra et al. 1991; Masojidek and Hall 1992; Belkhodja et al. 1994; Everard et al. 1994), whereas other studies have indicated that salt stress has no effect on PSII (Robinson et al. 1983; Brugnoli and Björkman 1992; Morales et al. 1992; Abadía et al. 1999). Most studies using PSII fluorescence in salt tolerant plants or halophytes have shown no effect on PSII photochemical efficiency (Lovelock et al. 1995). Furthermore, growth of *D. antarctica* plants exposed to 1 M NaCl for 4 days was completely restored after returning the plants to control conditions (data not shown). Therefore, *D. antarctica* may be considered a salt tolerant species. The mechanism that allows *D. antarctica* to survive high concentration of electrolytes in water is unknown. Studies in plants that inhabit under saline environments (halophytes) indicate that accumulated ions may be compartmentalized into cell vacuoles to protect the cytoplasm from toxic effects. To prevent dehydration of the cytosol, its osmotic potential must be adjusted to the level of the vacuole. This osmotic adjustment requires accumulation of compatible organic solutes (Simon-Sarkadi et al. 2002). For instance, free amino acids and polyamines accumulation has been shown to be an adaptive response in cereal crops (Simon-Sarkadi et al. 2002). It has been shown that *D. antarctica* exposed to low temperature under laboratory conditions at long photoperiod accumulates polysaccharides, sucrose and proline (Bravo et al. 2001). This plant also showed a high amount of non-structural carbohydrates specially in young leaves in the field (Zúñiga et al. 1996), where they are frequently exposed to freezing temperature, dehydrating winds and also sea-spray (Alberdi et al. 2002).

The induction of this family of dehydrins by environmental factors which may cause cell dehydration is consistent with the role of these proteins in dehydration injury prevention, and may be associated with the ability of this species to colonize and expand in the Antarctic territories. Therefore, the accumulation of DLPs in response to dehydrating factors may be associated to the high water use efficiency and capability to control water loss reported for *D. antarctica*, which may contribute to the enigmatic success of this species in the Antarctic.

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