7 Summary

Water uptake through the fruit surface is an important factor in cracking of sweet cherry fruit (Prunus avium L.). A better understanding of the mechanisms, pathways and factors that affect water uptake is needed to develop strategies against rain cracking of sweet cherry fruit. The objectives of this study were to

1. provide evidence for polar pathways across the sweet cherry exocarp,
2. investigate the effect of selected salts on water uptake,
3. establish the mechanism of Fe-salts in reducing water uptake, and
4. determine whether Fe-salts are suitable for applications under field conditions.

1. Polar pathways across the sweet cherry exocarp. Water uptake through the exocarp of intact cherry fruit (pedicel removed, pedicel/fruit juncture sealed with silicone rubber) was determined gravimetrically using an immersion assay. The osmotic water permeability (Pf) of the exocarp was linearly related to the inverse of viscosity of the incubation solution (gum Arabic at 10 to 300 g l\(^{-1}\), equivalent to dynamic viscosities of 1.3 \(\times\) 10\(^{-3}\) to 115.9 \(\times\) 10\(^{-3}\) Pa s), indicating that viscous flow must have contributed to water uptake. Viscous flow as a mechanism of water uptake requires a liquid continuum across the exocarp that acts as a “polar pathway”. When fruit were incubated in isotonic solutions of selected osmotica, water uptake decreased and the apparent water potentials increased as molecular weight (MW) of the osmotica increased [MW = 58 to 6000 g mol\(^{-1}\) for NaCl to polyethylene glycol (PEG) 6000]. Since solutions were isotonic, the size dependent water uptake must be accounted for by a size dependent penetration of the osmotica. Since the osmotica used were polar, penetration must have occurred through a size selective, porous polar pathway. Reflection coefficients (\(\sigma\)) estimated from the relationship between MW and the apparent water potential were closely related to the hydrodynamic radii (\(r\)) of the osmotica \([\sigma = 1.0 \pm 0.0 - 10.9 \pm 0.9 \times 10^{-11} r^{-1} (m^{-1}), r^2 = 0.97^{***}, P = 0.0001]\). Sucrose was the largest penetrating molecule with \(r = 0.47\) nm and \(\sigma = 0.74\), PEG 1500 the smallest non-penetrating molecule with \(r = 1.15\) nm und \(\sigma = 1\). Thus, the maximum pore radius of the polar pathways in the cherry exocarp ranged from 0.47 to 1.15 nm.

The self diffusion permeability (\(P_d\)) of exocarp segments (ES) and isolated cuticular membranes (CM) for radio labeled substances was determined using an Infinite-Dose-Diffusion system. In this system diffusion is monitored from a dilute donor through an interfacing membrane (ES or CM) into a receiver solution under steady-state-conditions. The \(P_d\) of sweet cherry ES for 1-naphthyl[1-\(^{14}\)C]acetic acid (NAA, \(pK_a = 4.2\)) and the activation
energy (E<sub>a</sub>) of NAA penetration was significantly higher for the predominantly undissociated (lipophilic) NAA [pH 2.2, P<sub>d</sub> = 10.2 (± 0.8) × 10<sup>-8</sup> m s<sup>-1</sup>, E<sub>a</sub> = 67.0 ± 1.7 kJ mol<sup>-1</sup>] than for the predominantly dissociated (polar) NAA [pH 6.2, P<sub>d</sub> = 1.1 (± 0.2) × 10<sup>-8</sup> m s<sup>-1</sup>, E<sub>a</sub> = 51.8 ± 1.9 kJ mol<sup>-1</sup>]. Furthermore, the E<sub>a</sub> for penetration of the dissociated NAA (r<sup>2</sup> = 0.84***, P = 0.0001) was more closely related to stomatal density (d<sub>sto</sub>) than that of the non-dissociated NAA (r<sup>2</sup> = 0.30*, P = 0.03), indicating that polar pathways in sweet cherry fruit are associated with the stomatal apparatus.

2. Effect of selected salts on water uptake. The rate of water uptake (F) in cherry fruit was determined in immersion assays in the absence (F<sup>I</sup>, 0 to 0.75 h) and, subsequently, in the presence of selected mineral salts (10 mM; F<sup>II</sup>, 0.75 to 1.5 h; F<sup>III</sup>; 1.5 to 2.25 h). Effects of salts were indexed by the ratio of rates of uptake (F<sup>II</sup>/F<sup>I</sup>, F<sup>III</sup>/F<sup>I</sup>). The largest decrease in water uptake was achieved, when fruit were incubated in CuCl<sub>2</sub> (F<sup>II</sup>/F<sup>I</sup> = 0.42), HgCl<sub>2</sub> (F<sup>II</sup>/F<sup>I</sup> = 0.58), AlCl<sub>3</sub> (F<sup>II</sup>/F<sup>I</sup> = 0.50) and FeCl<sub>3</sub> (F<sup>II</sup>/F<sup>I</sup> = 0.49). The effect of FeCl<sub>3</sub> on water uptake was independent of the presence of CuCl<sub>2</sub> and AlCl<sub>3</sub>, indicating that there was no additive effect between those salts that decreased water uptake. These findings demonstrate that the mode of action of “effective” salts must have been the same.

3. Effect of Fe-salts. The mechanism of FeCl<sub>3</sub> in decreasing water uptake was investigated using the Infinite-Dose-Diffusion technique. Adding FeCl<sub>3</sub> (10 mM) to the donor decreased P<sub>d</sub> of cherry ES for <sup>3</sup>H<sub>2</sub>O by about 60%. P<sub>d</sub> remained at a decreased level when replacing the FeCl<sub>3</sub> donor again by water. FeCl<sub>3</sub> reduced the P<sub>d</sub> of <sup>3</sup>H<sub>2</sub>O, when the water receiver had a higher pH (pH 3, 4.5 or 6) than the FeCl<sub>3</sub> containing donor (pH 2.3). Similar data were obtained when FeCl<sub>3</sub> was added to the receiver instead. However, when FeCl<sub>3</sub> was added to donor and receiver, there was no effect on the P<sub>d</sub> of <sup>3</sup>H<sub>2</sub>O. The FeCl<sub>3</sub>-induced decrease in P<sub>d</sub> for <sup>3</sup>H<sub>2</sub>O was positively and linearly related to stomatal density of the ES. FeCl<sub>3</sub> had no effect on P<sub>d</sub> for <sup>3</sup>H<sub>2</sub>O in astomatous systems (ES and CM). FeCl<sub>3</sub> also decreased the P<sub>d</sub> for NAA and 2,4-dichloro[<sup>14</sup>C]phenoxyacetic acid (2,4-D, pK<sub>a</sub> = 2.6) through stomatous ES. The magnitude of the decrease depended on the degree of dissociation and was larger for the predominantly dissociated (polar, pH 6.2) than for the undissociated (lipophilic, pH 2.2) acids. FeCl<sub>3</sub> also reduced water uptake into sweet cherry fruit, when fruit were incubated in isotonic solutions of selected osmotica. The data suggest that FeCl<sub>3</sub> decreased transport of water and other polar substances by a pH-dependent precipitation reaction that occurs in the polar pathways across the sweet cherry fruit exocarp (see 1.).
4. Suitability of Fe-salts for practical use. Characteristics that are relevant for spray application of Fe-salts in the field were investigated using laboratory immersion assays and cracking tests. The Fe-salts FeCl$_3$ and Fe(III)-glucoheptonate decreased water uptake at concentrations as low as 0.1 to 1 mM. The minimum “contact” period necessary to reduce permeability of the exocarp was 1 min in cv. Sam and 5 min in cv. Summit. The FeCl$_3$-induced decrease in water uptake persisted after replacing the FeCl$_3$ donor by water. After exposure of FeCl$_3$ or Fe(III)-glucoheptonate treated fruit to atmospheric conditions for up to 36 h, water uptake remained at a decreased level. FeCl$_3$ also decreased water uptake through microcracks in the exocarp, but had no significant effect on uptake through holes in the exocarp or on uptake along the pedicel/fruit juncture. Percentage of cracked fruit after spray application of FeCl$_3$ in the field was lower than in the water control. However, the reduction in fruit cracking obtained after spray application was markedly smaller than that after incubating fruit in FeCl$_3$. Unfortunately, spray application of FeCl$_3$ or incubation of fruit in FeCl$_3$ resulted in severe black discoloration of fruit that is inacceptable from a marketing point of view. These discolorations most likely represented Fe-oxides and –hydroxides formed by the precipitation reaction in the exocarp. Since these precipitates are essential for reducing water uptake, the use of Fe-salts is of limited usefulness for horticultural practice. Nevertheless, the identified mechanism of decreasing water uptake by a precipitation reaction that plugs polar pathways across the exocarp is a promising strategy. It allows the search for alternative compounds that (1) avoid the disadvantages of the ferric salts and (2) have an acceptable ecotoxicological profile.