

Mutation of a barley KCS (3-ketoacyl-CoA-synthase) compromises *Blumeria graminis* infection

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The barley mutant *emr1* (*enhanced Magnaporthe resistance 1*) was selected in a mutational screen based on its quantitative resistance against cereal blast caused by *Magnaporthe oryzae* (Jansen *et al.*, 2006). As a pleiotropic phenotype, we observed that coverage of *emr1* leaves with crystal waxes was strongly reduced. Mapping analysis using the iSELECT Veracode barley chip with 92 F2 plants of the cross '*emr1* x GZ *mlo11*' and subsequent linkage analysis with JoinMap 4.0 revealed that resistance towards blast and reduced wax segregated as two independent recessive. Therefore, mutants exhibiting the latter phenotype are referred to as *lowal* (*low wax 1*). Co-segregation of *lowal* with a SNP-marker enabled us to identify a candidate gene (*HvKCS1*) in which we found a point mutation by sequencing. *HvKCS1* was already described as a β -ketoacyl-CoA-synthase, which is part of the fatty acid elongase enzyme complex involved in the stepwise elongation of fatty-acyl-CoAs to very-long chain fatty acids (VLCFAs) (Richardson *et al.*, 2007). GC-analysis of epicuticular waxes of *lowal*-plants revealed a strong reduction of the C₂₆-alcohol hexacosanol and the C₂₆-aldehyde hexacosanal, accompanied by an accumulation of the C₂₄-alcohol, indicating that the elongation step from C₂₄- to C₂₆-VLCFAs is impaired in the mutant. Recent reports using *in vitro* assays or nonhost plants such as maize already described the importance of hexacosanal for germination and appressorium formation of the barley powdery mildew fungus *Blumeria graminis* f. sp. *hordei* (*Bgh*) (Hansjakob *et al.*, 2011). Using the *lowal* mutant, we could show for the first time for barley that reduced hexacosanal on the leaf surface results not only in less germination and hampered appressorium formation of *Bgh* but finally also leads to reduced disease severity. A reduction of prepenetration processes was also found after inoculation of *lowal* mutants with different species of powdery mildew fungi, indicating a general requirement of hexacosanal for powdery mildews in sensing suitable surfaces for germination. The closest Arabidopsis homologue to barley *KCS1*, namely *AtCut1* (*required for cuticular wax production*) (Millar *et al.*, 1999) was shown to be essential for elongation of C₂₄-VLCFAs. Inoculation of the corresponding knock-out mutant with the compatible Arabidopsis powdery mildew *Golovinomyces orontii* results in a similar reduction in infection structures as known from the *lowal*-

mutant, highlighting the adaptation of powdery mildews to this clade before specialization to monocot or dicot hosts.