Review Article

Research Advances on Cuticular Waxes Biosynthesis in Crops: A Review

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Abstract

Cuticular waxes are the hydrocarbon consisting of very long chain primary alcohols, aldehydes, fatty acids, alkane and esters. They are hydrophobic layer which protect aerial plant organs and help plant species for adaptation in different environments. Wax deposition and chemical composition vary considerably among crop species. Cuticular waxes play a significant role against major abiotic stresses in plants such as drought, high salinity and cold. So, it draws close attention to molecular processes of cuticular wax biosynthesis under stress factors. Here, we briefly summarized to the existing knowledge on the cuticular waxes properties, diversity, morphological changes in leaf surface wax crystals and amount and composition of cuticular waxes. We also provide information about wax biosynthesis genes in crops. Recently, due to progress of plant genome sequence, numerous genes involved in biosynthesis of cuticular waxes have been characterized both for model plant (Arabidopsis) and crops such as rapeseed (*Brassica napus*), *Camelina spp*, potato (*Solanum tuberosum*), eggplant (*S. macrocarpon*), tomato (*S. lycopersicum*), barley (*Hordeum vulgare*), rice (*Oryza sativa*), maize (*Zea mays*), wheat (*Triticum astivum*), broccoli (*B. olericea*), sesame (*Sesamum indicum*), tobacco (*Nicotiana tabacum*), cucumber (*Cucumis sativus*), cabbage (*B. oleracea*) etc. Basic compositions of cuticular wax are alcohols, branched alkanes, alkenes, aldehydes, fatty acids, esters, ketones, triterpenoids and sterols in crops. However, they vary from one crop species to the other. Cuticular wax biosynthesis is organ-specific and depends upon developmental stages of crops, and induced by environmental stimuli. The genetic factors also control wax biosynthesis and composition. However, cuticular wax also acts as a photoprotector layer during photosynthesis and protect from UV light radiation. It is also linked to gas exchange and plant development. In this review, we have summarized the cuticular wax amounts and contents in different organs, and genes to be involved in cuicular wax biosynthesis in several crops. This knowledge may be helpful in potential applications for selection of crop for agricultural sustainability. © 2019 Friends Science Publishers

Introduction

In growth and development stages, plants have to face enormous environmental stresses like drought, salinity, cold, heat, UV light, high radiation, insect or fungal and pathogens. The cuticular waxes are a surface layer of the plant which provide defense against pests and pathogens (Wink, 1988; Holmes and Keiller, 2002; Bargel *et al.*, 2004; Yeats and Rose, 2013). The cuticle is composed of two distinct layers which chemically separate compounds including a lipophilic cutin polymer matrix and waxes (Holloway, 1982; Jeffree, 1996; Kunst *et al.*, 2005). Cuticular waxes are most important elements which prevent uncontrolled evaporation of water at the leaves surfaces (Jetter and Riederer, 2000; Knoche *et al.*, 2000). The cuticular wax is composed of very long-chain fatty acid compounds (VLCFAs; C20 to C34). These VLCFAs compounds consist of Branched alkanes, primary alcohols, alkenes, aldehydes, secondary alcohols, β- and OH-β-diketones, esters and often triterpenoids and flavonoids (Jetter *et al.*, 2006; Samuels *et al.*, 2008). The genetic and environmental factors influence on the deposition and composition of cuticular waxes (Bianchi, 1995; Post-Beittenmiller, 1996). Cuticular wax biosynthetic pathways have been studied extensively in Arabidopsis (Hannoufa *et al.*, 1993; McNevin *et al.*, 1993; Jenks *et al.*, 1995; Suh *et al.*, 2005; Kunst and Samuels, 2009; Nawrath *et al.*, 2013). Several cuticular waxes genes from Arabidopsis were identified such as *FATB, LCAS1, LACS2, LCAS4, ACC1, KCS1, KCS2/DAISY, KCS6/CER6/CUT1, KCS9, KCS20, KCR1, HCD/PAS2, ECR/ECR10, CER2, CER2-LIKE1, CER2-LIKE2, CER1, RST1, CYTB5-B, CYTB5-C, CYTB-D, CYTB5-E, MAH1, FAR3/CER4* and *WSD1* (Lee and Suh, 2015). Number of cuticular wax genes has already been identified in crops (Table 1). Transcription, mRNA and post-translational modification are controlled by these genes in waxy and waxless plants (Von Wettstein-Knowles, 1995; Pu *et al.*, 2013; Lee *et al.*, 2015). However, genetic mechanism related to deposition of cuticular waxes in crops is still elusive and subject to further investigations.

Cuticular wax composition also depends on leaf color, insect-plant interaction and plant development. The quantity of plant cuticular wax largely depends upon environment conditions. Researchers have great interest to comprehend the detail genetic behavior of wax biosynthesis genes in crops. In this review, we summarized the amount and contents of cuticular waxes in the different crops and focused on recent progress about the molecular and biological function of genes engaged in biosynthesis of cuticular wax. We also briefly discussed the properties and diversities of cuticular waxes.

Properties of Cuticular Wax

Plant response and adaptation to abiotic and biotic stresses: Plant transpiration depends on two factors. Basically plant transpiration take place through stomata and a non-stomatal component is also there. Bernard and Joubès (2013) reported that there is significance correlation between the lipid cuticle layer and transpiration which was first proof about the role of cuticle for non-stomatal water loss (Stiles, 1994). Stomata remains close during water stress or night time, and provide space for significant cuticule transpiration. Several plant studies such as on tobacco and sesame also showed that wax biosynthesis was increased during water stress, and played an important role in preventing the cuticuler desiccation (Cameron *et al.*, 2006; Kim *et al.*, 2007). In Arabidopsis, water and osmotic stresses increased wax deposition that in turn were associated with a resistance to water stress, and cuticle formation could be a part of mechanism to acquire tolerance to water stress (Kosma *et al.*, 2009). Amount of wax and resistance to water flow depends on the cuticle biosynthesis enzymes (Aharoni *et al.*, 2004; Bourdenx *et al.*, 2011; Seo *et al.*, 2011). The regulation of cuticle permeability mainly depends on the wax deposition mechanisms during water stress. Several studies have reported that increase in cuticle permeability reduces wax load and vice versa (Chen *et al.*, 2003; Zhang *et al.*, 2005; Kosma *et al.*, 2009; Lü *et al.*, 2009, 2012; Bourdenx *et al.*, 2011; Seo *et al.*, 2011). Higher cuticle permeability depends on increased amount of cutin and waxes depositions.

Cuticular waxes need for plant development: Beside their major contribution to stress tolerance, cuticular waxes are active players in the growth and developmental processes in plant. Kurdyukov *et al.* (2006) reported that organ fusion phenotypes were frequently associated with severe imperfections in either cuticular wax or cutin biosynthesis, as noticed in, *bodyguard, Wddlehead, cer3/wax2* and numerous other mutants. Other studies on Arabidopsis also indicated that *lacs1lacs2* double-mutant plants demonstrated pleiotropic phenotypes such as organ fusion, unusual flower development and decreased seed set (Weng *et al.*, 2010). The wax-deficient *cer1* mutant of Arabidopsis had a conditional male-sterile phenotype that reduced pollen viability contributed to the low seed yield

(Aarts *et al.*, 1995). These studies indicated that a deficiency in cuticular waxes biosynthesis or cutin synthesis has a effects on cuticular barrier, water movement, defend against drought stress and protect organ fusion. Arabidopsis mutant's analyses help us to understand of changes in cuticular waxes amount and composition during different developmental stages and add conception to the action of cuticular waxes in plant physiology.

Diversity of Cuticular Wax

Despite our relatively advanced understanding of wax compound structure and biosynthesis in Arabidopsis, crucial questions remain unanswered about how chemical composition determines the physical properties of the cuticular wax mixture. Before addressing these questions, a thorough understanding of the major dimensions of cuticular wax diversity is needed, in particular, the diversity in the chemical structures and diversity in the waxes covering on different biological organs. It will move us closer to a fundamental understanding of the relationships between structure and function in the plant cuticular wax diversity.

Structural diversities of wax compound: Structural diversity may be evident from the aliphatic tail (e.g., number of unsaturations, aliphatic branches, TCN, etc) or functional groups (e.g., number of functional groups, positions on the aliphatic tail, oxidation state, etc) in the wax molecules. Some of those compounds were found in large amount in different plants and crops, indicating that they play a vital role in the properties of cuticular waxes and alter cuticular wax mixture. Moreover, their biosynthesis mechanisms of converting branched wax precursors into branched wax compounds are still not clear. It is needed to characterize branched wax compounds biosynthesis pathway to remove uncertainty of model species. Fatty acids, primary alcohols, alkenes, wax esters, aldehydes and branched alkanes are major cuticular wax components. However, wax profiles in different plant organs also revealed that some plants accumulate secondary functional groups with major cuticular wax compounds (Gunthardt-Goerg, 1986; Wen *et al.*, 2006). Ketones, ketoalcohols, and ketoaldehydes found on the surfaces of the fern *Osmunda re-alias* (Jetter and Riederer, 2000), while β-diketones are present on the surface of wheat and barley (Tulloch and Weenink, 1969; Jackson, 1971; Han-Avivi *et al.*, 2016; Schneider *et al.*, 2016; Huang *et al.*, 2017). It was proved that the true diversity of cuticular waxes presents in different plants. Thus, our knowledge about branched wax compound and biosynthesis is still nascent stage.

Biological variability in wax coverage and composition: Aerial plant organs are covered with waxes throughout their developmental stages. The quantity and composition of wax in plants depend upon the stresses in an age-dependent manner. Indeed, previous studies had reported that wax amount and compositions vary between plant surfaces of

Table 1: Genes known to be involved in cuticular wax biosynthesis in crop species

Mechanism	Species	Organs	Protein family name	Abbreviation	Reference
		Seedling leaf	Arabidopsis *KCS6* homolog	*GL4*	Avato *et al.*, 1987; Liu *et al.*, 2009
		Seedling leaf	Arabidopsis *KCR* homolog	*GL8a*	Xu *et al.*, 1997; Dietrich *et al.*, 2005
	Maize	Seedling leaf	Arabidopsis *KCR* homolog	*GL8b*	Dietrich *et al.*, 2005
		Seedling leaf	Arabidopsis *CER3* homolog	*GL1*	Hansen *et al.*, 1997; Sturaro *et al.*, 2005
		Seedling leaf	Arabidopsis *CER2* homolog	*GL2*	Lemieux, 1996; Velasco *et al.*, 2002
		Leaf	KCS	*WSL1*	Yu *et al.*, 2008
		Shoot	KCS	*ONI1*	Ito *et al.*, 2011
		Anther	Arabidopsis *CER1* homolog	*WDA1*	Jung *et al.*, 2006
		Leaf	Arabidopsis *CER1* homolog	*OsGL1-6*	Zhou *et al.*, 2013
		Leaf	Arabidopsis *CER3*, maize *GL1* homolog	*OsGL1-2*	Islam *et al.*, 2009
		Leaf	Arabidopsis *CER3* homolog	*OsGL1-1/WSL2*	Qin *et al.*, 2011; Mao *et al.*, 2012
		Leaf	AMP-binding domin contained	*DWA1*	Zhu and Xiong, 2013
		Leaf	Arabidopsis *CER3* homolog/ Maize *GL1* homolog	*OsGL1-3*	Zhou *et al.*, 2015
		Leaf	A homolog of the *MBOAT* transferase family	*OsWS1*	Xia *et al.*, 2015
	Rice	Leaf	$NAD^+/NADP^+$-dependent sterol dehydrogenase	*OsHSD1*	Zhang *et al.*, 2016
		Leaf	*KCR*	*WSL3*	Gan *et al.*, 2016
		Leaf	*KCS*	*WSL3*	Hong-bing *et al.*, 2017
		Leaf	Arabidopsis *CER6* homolog	*WSL4*	Wang *et al.*, 2017
		Leaf	*CER6-like KCS*	*LeCER6*	Vogg *et al.*, 2004;
	Tomato	Fruit	B-Amyrin synthesis	*SlTTS1*	Wang *et al.*, 2011
Cuticular wax biosynthesis		Fruit	Oxidosqualene cyclase	*SlTTS2*	Wang *et al.*, 2011
		Leaf	Unknown	*BnaA.GL*	Pu *et al.*, 2013
	B. napus	Leaf	lipid transfer proteins	*BraLTP1*	Liu *et al.*, 2014
	Brassica rapa	Leaf	Arabidopsis *CER2* homolog	*BrWax1*	Zhang *et al.*, 2013a
		Leaf	Arabidopsis *KCS2* homolog	*CsKCS2*	
		Leaf	Arabidopsis *KCS6* homolog	*CsKCS6*	
		Leaf	Arabidopsis *KCR 1* homolog	*CsKCR1-1*	Lee *et al.*, 2014
	C. sativa	Leaf	Arabidopsis *KCR 1* homolog	*CsKCR1-2*	
		Leaf	Arabidopsis *ECR* homolog	*CsECR*	
		Leaf	Arabidopsis *KCS2* homolog	*CsMAH1*	
	Cucumber	Leaf	Arabidopsis *WAX2* homolog	*CsWAX2*	Wang *et al.*, 2015a
		Leaf	Arabidopsis *CER1* homolog	*CsCER1*	Wang *et al.*, 2015b
		leaf	lipid transfer proteins	*BoLTP2*	
		Leaf	Arabidopsis *CER3* homolog	*BoCER3*	
		Leaf	Arabidopsis *KCS1* homolog	*BoKCS1*	
	Cabbage	Leaf	Arabidopsis KCR1 homolog	*BoKCR1*	Laila *et al.*, 2017
		Leaf	Arabidopsis *LACS1* homolog	*BoLACS1*	
		Leaf	alkane hydroxylase *CYP96A15*	*BoMAH1*	
		Leaf	Arabidopsis *CER4* homolog	*BoFAR3*	
		Leaf	Arabidopsis *WSD1*-like family	*BoWSD1*	
		Leaf	Arabidopsis *CER4-6* homolog	*W1W2*	Zhang *et al.*, 2013b
		Leaf	*CER1* and *CER3* homologs	*W3*	Zhang *et al.*, 2015
		Leaf	Arabidopsis *CER4* homolog	*TaFAR1*	Wang *et al.*, 2015b
	Wheat	Leaf	Arabidopis *CER4* homolog	*TaFAR5*	Wang *et al.*, 2015c
		Leaf	Arabidopsis *CER4* homolog	*TaFAR2, TaFAR3, TaFAR4,*	Wang *et al.*, 2016
		Spike	miRNA (MIRNA)	*W1-COE and / or W2-COE*	Huang *et al.*, 2017
		Leaf	Arabidopsis *CER4* homolog	*Ae.tFAR1,Ae.tFAR2, Ae.tFAR3,Ae.tFAR4, Ae.tFAR6*	Wang *et al.*, 2017
		Leaf	Arabidopsis *CER4* homolog	*TaFAR6,TaFAR7, TaFAR8*	Chai *et al.*, 2018
	Barley	Spike	PKS (DMP), Hydrolase (DMH),CYP450 (DMC)	*Cer-cqu*	Hen-Avivi *et al.*, 2016; Schneider *et al.*, 2016
		Leaf	Protein phosphatase 2C family protein	*Cer-b*	Zhou *et al.*, 2017

different ages (Atkin and Hamilton, 1950; Gülz *et al.*, 1992; Viougeas *et al.*, 1995). Largely, the understanding of cuticular wax deposition on plant surfaces needs further studies (Suh *et al.*, 2005). Plant surface development depends on cuticular wax deposition at different environmental conditions. It is needed to investigate the dimensions of biological variability and its relationship with the developmental biology of the plants. Thus, Cuticular waxes display structural diversity in their aliphatic tails and functional group (s) and biological variability depending on surfaces of different species, surfaces of different plant organs, and organ surfaces at different ages.

Cuticular Wax Morphology

The wax morphology was examined on both the adaxial and abaxial sides of leaf by scanning electron microscopy (SEM) to achieve insight into epicuticular wax crystals (Fig. 1). Leaf blades were collected from D genome (*Aegilops tauschii*) at three plant development stages (seedling, heading and filling stages). Two forms of wax crystals: platelets and tubules (A–L Figs) were found in the wheat leaf. Barthlott (1998) classified the epicuticular waxes. Some platelets shape wax crystals were joined to their adjacent crystals making a dense network. The length of platelet shaped wax crystals was between 0.3 and 0.7 µm and height between 0.3 and 0.5 µm. The platelet shaped wax crystals had irregular margins, and were present at different angles with respect to each other (Fig. 1 A–L). So, the wax morphology changed during their development stages. It is concluded that as the plant ages, the cuticular wax morphology changes on the leaf surface (Wang *et al.*, 2015a).

Cuticular Waxes in Crops

Amounts and contents of cuticular waxes in crop: It is very well known that cuticular waxes deposition varies across crops and from organ to organ (Barthlott *et al.*, 1998; Kosma *et al.*, 2010; Buschhaus and Jetter, 2011; Bernard and Joubès, 2013). Cuticular wax contents were measured in different organs of Arabidopsis, rapeseed (*Brassica napus*), *Camelina* spp, potato (*S. tuberosum*), eggplant (*S. macrocarpon*), tomato (*S. lycopersicum*), barley (*Hordeum vulgare*), maize (*Zea mays*), rice (*Oryza sativa*), wheat (*Triticum astivum*), broccoli (*B. olericea*), sesame (*Sesamum indicum*), tobacco (*Nicotianatabacum*), cucumber (*Cucumis sativus*) and cabbage (*B. oleracea*) (Table 2). Previous studies have shown that in Arabidopsis ecotype Columbia-0, the wax was 0.7–1.5, 13–24, 13 µg/cm^2 in leaves, stems and siliques, while it was 23-82 and 36–170 µg/g in flowers and seed coat, respectively. Alkane contributed up to 50% of total wax loads and represented the most dominant wax compound in all organs of Arabidopsis. In stems, silique walls, flowers, and seed coats, secondary alcohols and ketones were present but their very low amounts was noticed on leaf (Jenks *et al.*, 1995; Bernard and Joubès, 2013; Lee and Shu, 2015).

In rapeseed, cuticular wax amount in leaves is considerably higher than Arabidopsis leaves, while other wax compounds were similar (Pu *et al.*, 2013). In rapeseed breeding line 6-3476, the amount of wax in leaves (687–2255 µg/g) was similar to flower of Arabidopsis (2382 µg/g) (Tassone *et al.,* 2016). In *camelina sativum* var Celina, the amount of wax in leaves, stem, flower and seed coat was 6.24, 164, 264, 0.24 µg/cm^2, respectively. Wax esters (74%) in leaf and triterpenoids/sterols (53%) in stems were observed, but the primary alcohols (65%) were detected in seed coats (Razeq *et al.,* 2014).
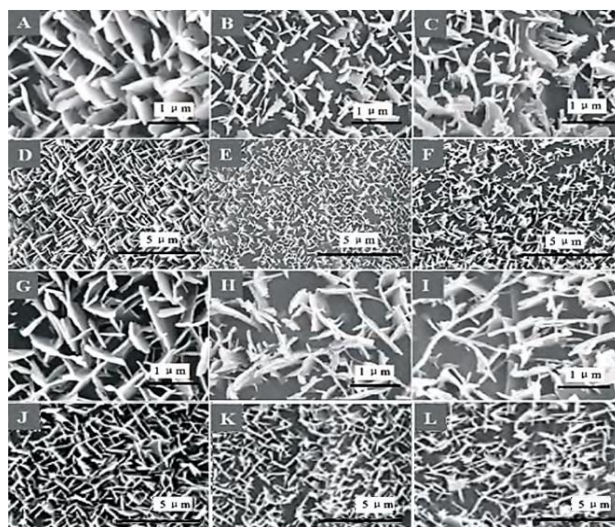
Fig. 1: Epicuticular wax crystals patterns on the adaxial and abaxial leaf surfaces of the wheat D genome (*A. tauschii*) detected by SEM at three stages of plant development. A, D are the adaxial surface of leaves during seedling stage; B, E are the adaxial surface of leaves during heading stage; C, F are the adaxial surface of leaves during filling stage; G, J are the abaxial of leaves during seedling stage; H, K are the abaxial surface of leaves during seedling stage; I, L are the abaxial surface of leaves during Filling stage. The micrographs are at a resolution of 10,000× and 30,000×, and the bars indicate 1 µm and 0.5 µm, respectively

Furthermore, in leaf of *Camelina MYB96* transgenic line, the amount of wax was 2.9 µg/cm^2 while in the leaf of Robinson variety it was 0.72 µg/cm^2 (Tomasi *et al.*, 2017). In another report, the wax compositions of cultivar *C. sativum* var Celina was similar to that reported by Razeq *et al.* (2014). Furthermore, the amount and composition in leaves of *C. sativa* 1.37 µg/cm^2, *C. rumelica* 2.01 µg/cm^2, *C. hispida* 0.85 µg/cm^2 and *C. microcarpa* 0.84 µg/cm^2 were reported but total wax loads were lower than younger leaves. In all Camelina species, the primary alcohols and alkanes were dominant components followed by wax esters, fatty acids and aldehydes. Interestingly, *C. sativa* var MYB96 synthesized higher levels of primary alcohols. It indicates that MYB96 might be an effective gene involved in the primary alcohols biosynthesis (Tomasi *et al.*, 2017). In potato, the amount of wax in the leaves was 5 µg/cm^2 in Perkoz, 6 µg/cm^2 in Aster and Maryna, and 7 µg/cm^2 in Ibis. Alkanes were dominant compound and primary alcohols were second major class in all potato varieties (Szafranek and Synak, 2006). In Gboma eggplant plant, the wax in cultivar UVPP was 2.7 µg/cm^2 and in Urafiki it was 2.3 µg/cm^2 were observed in leaves. Alkanes, primary alcohols, fatty acids, sterols, and triterpenols were also found. Alkanes (47–56%) were the dominant component in cuticular wax in Gboma Eggplant. Sterols content were observed much higher than triterpenes, consisting of 19 and 32% of the total waxes in Urafiki and UVPP cultivars, respectively (Halinski *et al.*, 2012).

Table 2: Cuticular wax amounts and contents in different organs in crops

Species	Organs	Total Loads	Fatty acids	Aldehydes	Alkanes	Primary alcohols	Sec. alcohols	Ketons	Wax esters	Iso-alkanes	Anteiso-alkanes	Alkenols	Alkenes	Triterpenoids & sterols	Ferulate Esters/Phenol	Reference
	Leaf	0.7–1.5[m]	1	14	73	8	0	0	4	0	0	0	0	0	0	Bernard and Joubès, 2013; Li-Beisson et al., 2013
Arabidopsis	Stem[a]	13–24[m]	1	7	44	12	9	22	5	0	0	0	0	0	0	Bernard and Joubès, 2013; Li-Beisson et al., 2013
	Silique[a]	13[m]		1	4	50	14	9	22	0	0	0	0	0	0	Kim et al., 2012
	Flower[a]	2382[n]	0	2	62	5	15	14	2	0	0	0	0	0	0	Bernard and Joubès, 2013
	Seed coat[a]	36–170[n]	0	9	51	13	18	9	0	0	0	0	0	0	0	Li et al. 2007; Bernard and Joubès, 2013
	Leaf	29.4[m]	1	4	55	2	10	27	2	0	0	0	0	0	0	Pu et al., 2013
Rapeseed	Leaf[b]	687–2255[n]	0	0	0	0	0		0	0	0	0	0	0	0	Tassone et al., 2016
	Leaf[c]	6.2[m]	3	0	3	20	0	0	74	0	0	0	0	0	0	
C. sativa	Steam[c]	16[m]	6	0	28	13	0	0	0	0	0	0	0	53	0	Razeq et al., 2014
	Flower[c]	264[n]	14	0	64	1	0	0	0	0	0	0	0	21	0	
	Seed coat[c]	0.2[m]	6	0	29	65	0	0	0	0	0	0	0	0	0	
C. sativa	Leaf[d]	2.9[m]	16	2	35	42	0	0	3	0	0	0	0	0	0	
C. sativa	Leaf[e]	0.72[m]	9	0	9	49	0	0	17	0	0	0	0	0	0	
C. sativa	Leaf[c]	0.83m	7	0	17	50	0	0	25	0	0	0	0	0		Tomasi et al., 2017
C sativum	leaf[c]	1.37m	12	1	22	43	0	0	14	0	0	0	0	0	0	
C. rumelica	Leaf	2.01[m]	6	1	35	40	0	0	17	0	0	0	0	0	0	
C. hispida	Leaf	0.85[m]	7	0	20	27	0	0	40	0	0	0	0	0	0	
C. microcarpa	Leaf	0.84[m]	4	0	45	44	0	0	0	0	0	0	0	0	0	
Potato	Leaf[f]	6[m]	9	0	61	12	1	1	3	0	0	0	0	2	0	
	Leaf[g]	7[m]	9	0	65	10	0	1	5	0	0	0	0	1	0	Szafranek and Synak, 2006
	Leaf[h]	6[m]	10	0	61	11	1	2	2	0	0	0	0	1	0	
	Leaf[i]	5[m]	5	0	61	7	1	2	2	0	0	0	0	1	0	
Egg Plant	Leaf[j]	2.7[m]	3	0	47	16	0	0	0	0	0	0	5	19	0	Halinski et al., 2012
	Leaf[k]	2.3[m]	5	0	56	18	0	0	0	0	0	0	5	32	0	
	Leaf[l]	6.7[m]	0	0	74	3	0	0	0	18	0	0	0	5	0	Wang et al., 2011
	Anther[l]	2155[n]	21	0	33	0	0	0	0	33	6	0	0	7	0	Smirnova et al., 2013
	Fruit[m]	8.4[m]	7	4	37	6	0	0	0	5	1	4	16	21	0	Isaacson et al., 2009
	Fruit[n]	10.2[m]	3	2	49	6	0	0	0	2	0	5	9	24	0	Kosma et al., 2010
Tomato	Leaf[L1]	2094[n]	2	0	72	1	0	0	0	0	0	0	0	10	0	
	Leaf[L2]	2118[n]	1	0	75	1	0	0	0	0	0	0	0	7	0	
	Leaf[L3]	1686[n]	1	0	73	1	0	0	0	0	0	0	0	12	0	
	Leaf[L4]	1131[n]	4	0	73	2	0	0	0	0	0	0	0	7	0	
	Leaf[L5]	1019[n]	3	0	77	1	0	0	0	0	0	0	0	7	0	
	Leaf[Pen1]	2351[n]	3	0	38	1	0	0	0	0	0	0	0	2	0	
	Leaf[Pen2]	3933[n]	4	0	53	2	0	0	0	0	0	0	0	7	0	Halinski et al., 2015
	Leaf[Pen3]	3738[n]	11	0	35	3	0	0	0	0	0	0	0	19	0	
	Leaf[Pen4]	3971[n]	1	0	71	1	0	0	0	0	0	0	0	1	0	
	Leaf[Pen5]	3116[n]	45	0	34	0	0	0	0	0	0	0	0	11	0	
	Leaf[LxPen]	2657[n]	1	0	62	1	0	0	0	0	0	0	0	4	0	
	Leaf[Pim1]	1342[n]	1	0	71	2	0	0	0	0	0	0	0	5	0	
	Leaf[LxPim]	1069[n]	2	0	71	1	0	0	0	0	0	0	0	5	0	
	Leaf[o]	14.7[m]	9	4	1	75	0	0	11	0	0	0	0	0	0	Von Wettstein-Knowles, 1971; Avato et al., 1982
Barley	Leaf[p]	14.6m	2	1	1	6	0	59	0	0	0	0	0	0	0	Yu et al., 2008

Table 2: Continued

Table 2: Continued

Plant	Tissue	Total														Reference
	Leaf Blade[q]	7.57[m]	18	37	2	32	0	0	11	0	0	0	0	0	0	0	Mao *et al.*, 2012
	Leaf Sheath[q]	5.8[m]	14	32	3	36	0	0	16	0	0	0	0	0	0	0	
Rice	Leaf Blade[r]	8.4[m]	34	31	7	24	0	0	4	0	0	0	0	0	0	0	
	Leaf Sheath[r]	4.5[m]	43	24	9	23	0	0	0	0	0	0	0	0	0	0	
	Leaf Blade[s]	6.2[m]	58	0	19	22	0	0	0	0	0	0	0	0	0	0	Wang *et al.*, 2017
Maize[t]	Leaf Blade	8.2[m]	0	25	4	69	0	0	2	0	0	0	0	0	0	0	Javelle *et al.*, 2010
	Leaf Sheath	4.5[m]	0	42	14	39	0	5	0	0	0	0	0	0	0	0	
	Leaf[1]	3.4[m]	6	3	14	71	0	3	2	0	0	0	0	0	0	0	
	Leaf[2]	3.6 m	4	3	10	79	0	1	2	0	0	0	0	0	0	0	
	Leaf[3]	3.9[m]	5	4	15	68	0	6	3	0	0	0	0	0	0	0	
	Leaf[4]	3.6[m]	5	3	11	68	0	12	2	0	0	0	0	0	0	0	
Wheat	Spike[1]	4.5[m]	13	1	40	18	0	37	0	0	0	0	0	0	0	0	Wang *et al.*, 2015a
	Spike[2]	5.8[m]	11	1	31	30	0	27	0	0	0	0	0	0	0	0	
	Spike[3]	7.7[m]	10	1	19	7	0	63	0	0	0	0	0	0	0	0	
	Spike[4]	6.6[m]	10	1	31	11	0	47	0	0	0	0	0	0	0	0	
	Leaf[5]		1	0	34	1	0	63	1	0	0	0	0	0	0	0	Zhang *et al.*, 2015
	Leaf[6]	16[m]	1	3	9	0	0	14	9	0	0	55	0	0	0		Racovita *et al.*, 2016
	Peduncle[7]	4.9[m]	1	1	7	0	0	81	2	0	0	2	0	0	0		
	Seedling leaf[7]	5.4m	1	5	2	84	0	0	1	0	0	0	0	0	0		
	Flag leaf[7]	8.4m	2	9	6	77	0	1	1	0	0	0	0	0	0		Wang *et al.*, 2017
	Leaf sheaths[7]	3.5m	2	4	15	57	0	15	3	0	0	0	0	0	0		
	Peduncles[7]	2.9m	3	13	37	23	0	36	2	0	0	0	0	0	0		
	Glumes[7]	1.0m	2	7	10	30	0	10	2	0	0	0	0	0	0		
	Anthers[7]	0.3m	54	1	27	3	0	27	1	0	0	0	0	0	0		
Broccoli	Leaf18	1929 n	13	20	39	3	5	19	0	0	0	0	0	0	0		Lee *et al.*, 2015
	Leaf19	3733 n	15	13	40	3	7	22	0	0	0	0	0	0	0		
Sesame	Leaf10	7.69 m	0	11	68	0	0	0	0	0	0	0	0	0	0		Kim *et al.*, 2007
Tobacco	Leaf[11]	13.9m	7.8	0	74	7	0	0	0	0	0	0	0	0	0		Cameron *et al.*, 2006
Cucumber	Friut12	1.3m	21	22	46	0	3	0	0	2	0	0	2	0	3		Wang *et al.*, 2015a
	Stem12	1.6m	6	8	82	0	10	0	0	1	0	0	1	0	3		
	Leaf12	1.8m	8	15	62	0	8	0	0	5	0	0	0	0	2		
Cabbage	Leaf13	0	0	6	34	6	14	31	0	0	0	0	0	0	0		Laila *et al.*, 2017

Note: number 0 indicates that trace or undetectable amounts were audited. [a]ecotype col-0, [b]breeding line 6-3476, [c]cultivar *Cemelina sativum* var celina, [d] *C. sativum* var *MYB96*, [e]*C. sativum* var robinson, [f]potato cultivar aster, [g]potato cultivar ibis, [h]potato cultivar maryna, [i]potato cultivar uvpp, [k]Gboma egg plant cultivar urafiki, [l]cultivar micro tom, [m]cultivar tomato m82, [n]cultivar tomato ailsa craig, [11] tomato cultivar vf-36, [12]tomato cultivar wild (ecuador), [13]tomato cultivar wild (ecuador), [14]tomato cultivar nagcarlang, [15] tomato cultivar wild (usa),[pen1]tomato cultivar wild (peru), [pen2]tomato cultivar wild (peru), [pen3] tomato cultivar wild (peru), [pen4]tomato cultivar wild (peru), [pen5]tomato cultivar wild (peru), [lxpen]tomato cultivar hybrid, [pim1]tomato cultivar wild (ecuador), [lxpim]tomato cultivar hybrid, [o]barley cultivar bonus, [p]barley cultivar bowman, [q]rice cultivar nipponbare, [r]rice cultivar japonica, [s]maize inbred line a188, [1]wheat cultivar a14, [2]wheat cultivar jing 2001, [3]wheat cultivar fanmai 5,[4]wheat cultivar shanken 99, [5]wheat cultiver bob white,[6]wheat cultivar bethlehem,[7]wild grass *Ae. tauschii*. [8]broccoli cultivar mc 117,[9]broccoli cultivar mc91, [10]seasame cultivar various, [11]tobacco cultivar graha, [12]cucumber cultivar wild type, [13]cabbage cultivar *b. oleracea* , [m] unit = µg/cm^2, [n]unit = µg/g

In tomato cultivar Micro-Tom, amount and composition of cuticular waxes in leaf and anther were 6.7 µg/cm^2 and 2155 µg/g, respectively, and the most abundant components were alkanes (74%) in leaves (Wang *et al.*, 2011). Tomato cultivar M82 and cultivar Ailsa Craig in fruits contained 8.4 µg/cm^2 and 10.2 µg/cm^2 wax total load, respectively while alkanes and alkenes were the dominant wax component (Isaacson *et al.*, 2009; Kosma *et al.*, 2010). However, the most dominant waxes components of these varieties were alkanes ranged from 34 to 71% (Halinski *et al.*, 2015). In tomato leaves, fruits and anthers, branched alkanes, alkenes, and cyclic compounds were detected (Isaacson *et al.*, 2009; Wang *et al.*, 2011; Smirnova *et al.*, 2013; Halinski *et al.*, 2015). This finding was in the agreement with the other researchers that cuticular waxes in tomato leaf were consisted by hydrocarbons (Zygadlo *et al.*, 1994; Smith *et al.*, 1996; Vogg *et al.*, 2004). Primary alcohols, aldehydes and fatty acid were found to be principal components of cuticular wax, where alkanes occupied less than 15% of the total wax loads in leaves of barley, rice and maize (Von Wettstein-Knowles, 1971; Avato *et al.*, 1982; Javelle *et al.*, 2010; 2011; Mao *et al.*,

2012) whereas ketones were the most abundant component in leaves of barley cultivar Bowman (Yu *et al.*, 2008).

In wheat, primary alcohols were the major components in leaves cuticular wax followed by alkanes, esters, aldehydes and fatty acids. Ketones deposition variations were observed in wheat leaves. In wheat leaves, ketones were found in trace amount the cuticular wax. However, in spike, a traceable amount of ketones was identified (Wang *et al.*, 2015a, 2017). In addition, there were large differences in the amounts of ketones among different wheat varieties (Wang *et al.*, 2015a; Zhang *et al.*, 2015). Wheat cultivar Bethlehem revealed wax coverage of 16 µg/ cm^2 in leaf and 49 µg/cm^2 in peduncles (Zhang *et al.*, 2015). Furthermore, in wheat cultivar Bethlehem, alkanols were 55% of total wax loads in leaf and β-diketone and hydroxy-β-diketones collectively comprised 81% of the total wax loads in peduncle. This happened due to discrepancy in the regulation of the acyl-reduction and β-diketone biosynthetic pathways in the two examined organs (Racovita *et al.*, 2016).

In broccoli cultivars MC117 and MC91, fatty acids (13 and 15%, respectively), aldehydes (20 and 13%, respectively), alkanes (38 and 39%, respectively) and ketones (19 and 21%, respectively) were found in the total wax loads in leaf (Lee *et al.*, 2015). In sesame, major components of waxes in leaves were alkanes (68% of total wax) and aldehydes (11% of total wax) (Kim *et al.*, 2007). In tobacco, the primary component of cuticular wax was alkanes, which constituted 75% of the total wax load fully expanded leaves while fatty acids and alcohols occupied smaller proportion of total wax loads (Cameron *et al.*, 2006). In cucumber, alkenes, primary alcohols, branched alkanes, phenols, esters, and aldehydes, phenols were major compounds in the cuticular waxes (Wang *et al.*, 2015a). In cabbage leaf, alkane contributed 34% followed by ketones 31% of total wax loads (Laila *et al.*, 2017).

In spite of a vast variation in cuticular wax loads and contents depending upon crops and organs, the prime reasons that responsible for these variations are not identified. Nevertheless, the present information may be used as a genetic source for determination of new genes associated with cuticular wax biosynthsis.

Biosynthesis of cuticular wax genes in crops: In crops, the genes responsible for biosynthesis of cuticular wax are not well characterized such as in Arabidopsis, though, recently notable advances have been made in this respect (Table 1). Identification and functional expression of the glossy (*gl*) mutants provided an opportunity to understand about wax biosynthesis in maize. Mutants of *GL4* and *GL8* are homolog of *AtKCS6* and *AtKCR*, respectively. They displayed a spectacular decrease in the amount of alkanes, alcohols and aldehydes in the leaves of wild type seedling (Avato *et al.*, 1987; Dietrich *et al.*, 2005; Liu *et al.*, 2009). In another study on maize, it was found that *gl1* and *gl2* mutants which were homolog to *AtCER3* and *AtCER2*, respectively, declined wax deposition in seedling leaves,

especially the aldehydes levels significantly decreased or could not be identified. Contrarily, the *gl1gl2* double mutant enhanced the levels of wax esters (Bianchi *et al.*, 1979; Lemieux, 1996; Hansen *et al.*, 1997; Velasco *et al.*, 2002; Sturaro *et al.*, 2005). It was also found that *GL13*, an ABC transporter, was associated with cuticular wax deposition (Li *et al.*, 2013).

In rice, *crystal-spares leaf1* (*wsl1*) mutant, is a *KCS* gene which has a lesion of *WSL1* gene, catalyzed the creation of C20–C24 VLCFA precursors of leaf waxes. It reduced growth, fertility, leaf fusion and increased drought sensitivity due to wax-deficiency. This is indicated that *WSL1* might be involved in the deposition of other lipids associated with growth and development of the plant (Yu *et al.*, 2008). *ONION1 (ONI1)* is another homolog to rice *KCS* protein. It is accountable for synthesis of C20 and C22 saturated VLCFAs, which are important for development of shoot (Ito *et al.*, 2011). From functional expression of *wax-deficient anther1 (wda1)* mutant, it was observed that a *WDA1* protein, which is homolog to *AtCER1*, was responsible for synthesis of VLC alkenes and alkanes in pollen and anthers (Jung *et al.*, 2006). Functional characterization of *OsGL1-6*, which is also homolog to *AtCER1,* reported that it is needed for wax biosynthesis on leaf blades. It was also found that reduced expression of the *OsGL1-6* gene was linked with notable decreases in total wax loads and enhanced drought sensitivity (Zhou *et al.*, 2013). Characterization of rice mutants, *gl1-2* and *gl1-1/wsl2,* revealed that these mutants reduced overall cuticular wax loads and increased sensitivity to drought stress (Islam *et al.*, 2009; Qin *et al.*, 2011; Mao *et al.*, 2012). Over expression of *Drought-Induced Wax Accumulation 1* (*DWA1*) held an AMP-binding domain, which increased VLCFA synthesis and enhanced drought resistance (Zhu and Xiong, 2013). *OsGL1-3* is homologous to maize *GL1* and *Arabidopsis WAX2/YRE/CER3/FLP,* which significantly increased biosynthesis of cuticular wax and enhanced tolerance to water stress (Zhou *et al.*, 2015). *OsWS1* belongs to the membrane-bound O-acyl transferase gene family, and is associated with wax biosynthesis in rice (Xia *et al.*, 2015). *OsHSD1* belongs to the short-chain dehydrogenase reductase family, which enhanced VLCFAs biosynthesis and soluble fatty acids in the leaves of the *oshsd1* mutant (Zhang *et al.*, 2016). *WSL3* encodes *KCR* in rice, which contributes to VLCFA biosynthesis and wax depositions in leaf. On the other hand in rice mutant, *wax crystal-sparse leaf 3* (*wsl3*) gene reduced epicuticular wax crystals and wax composition on the leaf surface (Gan *et al.*, 2016). *ORF4* is homologous to the *KCS6* family of *KCS,* which is similar to *WSL3* gene of rice and regulates cuticular wax formation (Hong-bing *et al.*, 2017). *WSL4* encodes a *KCS*, a homolog of *AtCER6*, which increased the cuticular wax load in rice leaves (Wang *et al.*, 2017).

In tomato fruit cuticle, the *lecer6* mutant belongs to *CER6-like KCS* (*LeCER6*), decreased accumulation of alkanes and aldehydes but on the other hand, amounts of

triterpenoid increased in the total wax loads (Vogg *et al.*, 2004). In tomato fruits, the over expression of *SlTTS1* and *SlTTS2* enhanced biosynthesis of terpenoid (Wang *et al.*, 2011). However, alkane is the dominant wax component in the tomato wax but genes/proteins have not yet been identified for the biosynthesis of alkanes. In *B. napus*, glossy mutant *BnaA.GL* was characterized, which reduced wax biosynthesis and increased sensitivity to drought stress (Pu *et al.*, 2013). *BraLTP1* belongs to non-specific lipid transfer proteins (*nsLTPs*), which decreased wax deposition in leaves (Liu *et al.*, 2014). In *B. rapa*, the *BrWax1* gene is found on linkage group A01, which is involved in cuticular wax biosynthesis in leaves (Zhang *et al.*, 2013a). In *Camelina sativa,* few wax biosynthesis genes have been detected recently but their characterization is not complete yet (Lee *et al.*, 2014). In Cucumber, *CsWAX2* is homolog of *AtWAX2*, which performs fundamental functions in wax biosynthesis (Wang *et al.*, 2015a) and *CsCER1*, a homolog of *AtCER1*, played an important role in wax VLC alkanes biosynthesis (Wang *et al.*, 2015b). In Cabbage, *BoLTP2* gene is a non-specific *lipid-transfer protein1*, involved in the transformation of ketones to lipid which reduced wax deposition. *BoCER3* gene is homologous to *AtCER3* protein, which is related to wax deposition through converting aldehydes to alkanes. *BoKCS1* and *BoKCR1* are homologs to *AtKCS* and *AtKCR1* protein, respectively, which decreased acyl-CoAs synthesis and eventually influence total wax loads. Likewise, expressions of *BoLACS1* influenced wax depositions. Besides, the *BoMAH1* gene engaged in synthesis of secondary alcohols and ketones (Laila *et al.*, 2017).

In wheat, *W1W2* is a homolog of Arabidopsis *CER4-6* proteins. It produced hydroxyl-β-diketones, which enhanced drought tolerance through reducing cuticle permeability (Zhang *et al.*, 2013b). *W3*, a homolog of Arabidopsis *CER1* and *CER3,* is essential for β-diketone biosynthesis but suppresses its hydroxylation (Zhang *et al.*, 2015). *TaFAR1,* a homolog of Arabidopsis *CER4,* is an active acyl-CoA reductase. It produced primary alcohols, and as a result augmented total wax loads on wheat leaf blades (Wang *et al.*, 2015b). *TaFAR5,* a homolog to Arabidopsis *CER4,* is an alcohol-forming fatty acyl-coenzyme A reductase (*FAR*), which contributes significantly to produce primary alcohols in wheat leaf blade (Wang *et al.,* 2015c). *TaFAR2, TaFAR3,* and *TaFAR4* genes are a homolog of Arabidopsis *CER4* protein, which produced primary alcohols in cuticular wax (Wang *et al.*, 2016). From characterization of the *Iw* genes, it was found that *Iw* genes regulatory mechanism control *W-COE* expression and β-diketone formation (Huang *et al.*, 2017). In, Dgenome (*Ae. Tauschii*), *Ae.tFAR1*, *Ae.tFAR2*, *Ae.tFAR3*, *Ae.tFAR4*, and *Ae.tFAR6* are homolog to *AtCER4*, which principally accountable for deposition of primary alcohols (Wang *et al.*, 2017). In barley, the *Cer-cqu* gene cluster is involved in β-diketons biosynthesis which consists of several proteins families including type-III polyketide synthases, hydrolases, and cytochrome P450s

(Hen-Avivi *et al.*, 2016; Schneider*et al.*, 2016). The barley *eceriferum-b.2* (*cer-b.2*) mutant produces β-diketons, which makes glossy leaf sheaths and deficient in the cuticular wax component, 14, 16-hentriacontanedione (Zhou *et al.*, 2017).

Conclusion

This review has revealed that wax compositional differences exist among different crops even organ to organ. High levels of structural diversities associates with cuticular wax deposition in crops are largely influenced by different genes. Several genes have been already identified, which are related to cuticular wax biosynthesis. Cuticular wax components are produced by two different complex pathways due to the influence of biotic and abiotic stress, which allow adaptive mechanism at the time of crop-environment interactions. Moreover, specific single wax components yet remain unknown during development and growth stages of different crops. Very few studies have focused on primary alcohols and ketons in wheat and barley, but most of the factors are still unidentified. Additionally, genome sequencing technologies have been progressed enormously, allowed identifying new race of cuticular waxes biosynthesis genes in crops. Knowledge on the wax biosynthesis mechanisms in different crops will be helpful to breed new crops cultivars better tolerant to environmental stresses.

References

Aarts, M.G., C.J. Keijzer, W.J. Stiekema and A. Pereira, 1995. Molecular characterization of the *CER1* gene of arabidopsis involved in epicuticular wax biosynthesis and pollen fertility. *Plant Cell,* 7: 2115–2127

Aharoni, A., S. Dixit, R. Jetter, E. Thoenes, G.V. Arkel and A. Pereira, 2004. The *SHINE* clade of AP2 domain transcription factors activates wax biosynthesis, alters cuticle properties, and confers drought tolerance when overexpressed in *Arabidopsis. Plant Cell*, 16: 2463–2480

Atkin, D.S.J. and R.J. Hamilton, 1950. The changes with age in the epicuticular wax of sorghum bicolor. *J. Nat. Prod.*, 45: 697–703

Avato, P., G. Bianchi, A. Nayak, F. Salamini and E. Gentinetta, 1987. Epicuticular waxes of maize as affected by the interaction of mutant *gl8* with *gl3*, *gl4* and *gl15*. *Lipids*, 22: 11–16

Avato, P., J.D. Mikkelsen and P. von Wettstein-Knowles, 1982. Synthesis of epicuticular primary alcohols and intracellular fatty acids by tissue slices from cer-j59 barley leaves. *Carlsberg Res. Commun.,* 47: 377–390

Bargel, H., W. Barthlott, K. Koch, L. Schreiber and C. Neinhuis, 2004. 10-Plant cuticles: multifunctional interfaces between plant and environment. *In: Evolution Plant Physiology*, pp: 171–194. A.R. Hemsley and I. Poole (eds.). Academic Press, London

Barthlott, W., C. Neinhuis, D. Cutler, F. Ditsch, I. Muesel, I. Theisen and H. Wilhelm, 1998. Classification and terminology of plant epicuticular waxes. *Bot. J. Linn. Soc.*, 126: 237–260

Bernard, A. and J. Joubès, 2013. Arabidopsis cuticular waxes: advances in synthesis, export and regulation. *Prog. Lipid Res.*, 52: 110–129

Bianchi, G., 1995. Plant waxes. *In: Waxes: Chemistry, Molecular Biology and Functions*. pp: 175–222. Hamilton, R.J. (ed.). The Oily Press, Dundee, Scotland, UK

Bianchi, G., P. Avato and F. Salamini, 1979. The absence of fatty acid aldehydes in *gl1 gl2* waxes. *Maize Gen Coop Newslett.*, 53: 103

Bourdenx, B., A. Bernard, F. Domergue, S. Pascal, A. Léger, D. Roby, M. Pervent, D. Vile, R.P. Haslam, J.A. Napier, R. Lessire and J. Joubès, 2011.Overexpression of Arabidopsis *ECERIFERUM1* promotes wax very-long-chain alkane biosynthesis and influences plant response to biotic and abiotic stresses. *Plant Physiol.*, 156: 29–45

Buschhaus, C. and R. Jetter, 2011. Composition differences between epicuticular and intracuticular wax substructures: how do plants seal their epidermal surfaces. *J. Exp. Bot.*, 62: 841–853

Cameron, M.A., Teece and L.B. Smart, 2006. Increased accumulation of cuticular wax and expression of lipid transfer protein in response to periodic drying events in leaves of tree tobacco. *Plant Physiol.*, 140: 176–183

Chai, G., C. Li, F. Xu, Y. Li, X. Shi, Y. Wang and Z. Wang, 2018. Three endoplasmic reticulum-associated fatty acyl-coenzyme a reductases were involved in the production of primaryalcohols in hexaploid wheat (*Triticum aestivum* L.). *BMC Plant Biol.*, 18: 41

Chen, X., S.M. Goodwin, V.L. Boroff, X. Liu and M.A. Jenks, 2003. Cloning and characterization of the *WAX2* gene of Arabidopsis involved in cuticle membrane and wax production. *Plant Cell*, 15: 1170–1185

Dietrich, C.R., M.A.D.N. Perera, D.M. Yandeau-Nelson, R.B. Meeley and B.J. Nikolau, 2005. Characterization of two *GL8* paralogs reveals that the 3-ketoacyl reductase component of fatty acid elongase is essential for maize (*Zea mays* L.) development. *Plant J.*, 42: 844–861

Gan, L., X. Wang, Z. Cheng, L. Liu, J. Wang, Z. Zhang, Y. Ren, C. Lei, Z. Zhao, S. Zhu, Q. Lin, F. Wu, X. Gao, J. Wang, X. Zhang and J. Wan, 2016. *Wax crystal-sparse leaf 3* encoding a β-ketoacyl-CoA reductase is involved in cuticular wax biosynthesis in rice. *Plant Cell Rep.*, 35: 1687–1698

Gülz, P.G., E. Müller, K. Schmitz, F.J. Marner and S. Güth. 1992. Chemical composition and surface structures of epicuticular leaf waxes of *Ginkgo biloba*, *Magnolia grandiflora* and *Liriodendron tulipif-era*. *Z Naturforsch C.*, 47: 516–526

Gunthardt-Goerg, M.S., 1986. Epicuticular wax of needles of *Pinus cembra*, *Pinus sylvestris* and *Picea abies*. *Eur. J. For. Pathol.*, 16: 400–408

Halinski, L.P., M. Kalkowska, M. Kalkowski, J. Piorunowska, A. Topolewska, P. Stepnowski, 2015. Cuticular wax variation in the tomato (*Solanum lycopersicum* L.) related wild species and their interspecific hybrids. *Biochem. Syst. Ecol.*, 60: 215–224

Halinski, L.P., M. Paszkiewicz, M.G. Biowski and P. Stepnowski, 2012. The chemical composition of cuticular waxes from leaves of the gboma eggplant (*Solanummacrocarpon* L.). *J. Food Comp. Anal.*, 25: 74–78

Hannoufa, A., J. McNevin and B. Lemieux, 1993. Epicuticular waxes of eceriferum mutants of *Arabidopsis thaliana*. *Phytochemistry*, 33: 851–855

Hansen, J.D., J. Pyee, Y. Xia, T.J. Wen, D.S. Robertson, P.E. Kolattukudy, B.J. Nikolau and P.S. Schnable, 1997. The *glossy1* locus of maize and an epidermis-specific cDNA from *Klenia odora* define a class of receptor-like proteins required for the normal accumulation of cuticular waxes. *Plant Physiol.*, 113: 1091–1100

Hen-Avivi, S., O. Savin, R.C. Racovita, W.S. Lee, N.M. Adamski, S. Malitsky, E. Almekias-Siegl, M. Levy, S. Vautrin, H. Bergès, G. Friedlander, E. Kartvelishvily, G. Ben-Zvi, N. Alkan, C. Uauy, K. Kanyuka, R. Jetter, A. Distelfeld and A. Aharoni, 2016. A metabolic gene cluster in the wheat *W1* and the barley *Cer-cqu* loci determines β-diketone biosynthesis and glaucousness. *Plant Cell*, 28: 1440–1460

Holloway, P.J., 1982. Structure and histochemistry of plant cuticular membranes: An overview. *In: The Plant Cuticle*, pp: 1–32. D.F. Cutler, K.L. Alvin and C.E. Price (eds.). Academic Press, London

Holmes, M.G. and D.R. Keiller, 2002. Effects of pubescence and waxes on the reflectance of leaves in the ultraviolet and photosynthetic wavebands: A comparison of a range of species. *Plant Cell Environ.*, 25: 85–93

Hong-bing, G., Z. Sheng-yuan, X. Xiang, Z. Ya-fang, C. Zong-xiang, Z. Shi-min, L. Chuang, L. Tian-zi, J. De-dao, Y. Bo, Q. Hua-fei, P. Xue-biao and S. Sheng-lan, 2017. Fine mapping of a novel *wax crystal-sparse leaf3* gene in rice. *J. Integr. Agric.*, 16: 497–502

Huang, D., J.A. Feurtadoa, M.A. Smitha, L.K. Flatmana, C. Koha and A.J. Cutlera, 2017. Long noncoding miRNA gene represses wheat β-diketone waxes. *Proc. Natl. Acad. Sci. USA*, 114: E3149-E3158

Isaacson, T., D.K. Kosma, A.J. Matas, G.J. Buda, Y. He, B. Yu, A. Pravitasari, J.D. Batteas, R.E. Stark, M.A. Jenks and J.K.C. Rose, 2009. Cutin deficiency in the tomato fruit cuticle consistently affects resistance to microbial infection and biomechanical properties, but not transpirational water loss. *Plant J.*, 60: 363–377

Islam, M.A., H. Du, J. Ning, H. Ye and L. Xiong, 2009. Characterization of *Glossy1* homologous genes in rice involved in leaf wax accumulation and drought resistance. *Plant Mol. Biol.*, 70: 443–456

Ito, Y., F. Kimura, K. Hirakata, K. Tsuda and T. Takasugi, 2011. Fatty acid elongase is required for shoot development in rice. *Plant J.*, 66: 680–688

Jackson, L.L., 1971. Beta-diketone, alcohol and hydrocarbons of barley surface lipids. *Phytochemistry*, 10: 487–490

Javelle, M., V. Vernoud, N. Depège-Fargeix, C. Arnould, D. Oursel, F. Domergue, X. Sarda and P.M. Rogowsky, 2010. Overexpression of the epidermis-specific homeodomain-leucine zipper IV transcription factor Outer Cell Layer1 in maize identifies target genes involved in lipid metabolism and cuticle biosynthesis. *Plant Physiol.*, 154: 273–286

Javelle, M., V. Vernoud, P.M. Rogowsky and G.C. Ingram, 2011. Epidermis: the formation and functions of a fundamental plant tissue. *New Phytol.*, 189: 17–39

Jeffree, C.E., 1996. Structure and ontogeny of plant cuticles. *In: Plant Cuticles: An Integrated Functional Approach*, pp: 33–82. Kersteins, G. (ed.). BIOS Scientific Publishers Ltd., Oxford, UK

Jenks, M.A., H.A. Tuttle, S.D. Eigenbrode and K.A. Feldmann, 1995. Leaf epicuticular waxes of the eceriferum mutants in *Arabidopsis*. *Plant Physiol.*, 108: 369–377

Jetter, R. and M. Riederer, 2000. Composition of cuticular waxes on *Osmunda regalis* fronds. *J. Chem. Ecol.*, 26: 399–412

Jetter, R., L. Kunst and A.L. Samuels, 2006. Composition of plant cuticular waxes. *In: Biology of the Plant Cuticle*, Vol. 23, pp: 145–181. M. Riederer and C. Müller (eds.). Blackwell, Oxford, UK

Jung, K.H., M.J. Han, D.y. Lee, Y.S. Lee, L. Schreiber, R. Franke, A. Faust, A. Yephremov, H. Saedler, Y.W. Kim, I. Hwang and G. An, 2006. *Wax-deficient anther1* is involved in cuticle and wax production in rice anther walls and is required for pollen development. *Plant Cell*, 18: 3015–3032

Kim, H., S.B. Lee, H.J. Kim, M.K. Min, I. Hwang and M.C. Suh, 2012. Characterization of Glycosyl phosphatidylinositol-Anchored *Lipid Transfer Protein2* (*LTPG2*) and overlapping function between *LTPG/LTPG1* and *LTPG2* in cuticular wax export or accumulation in *Arabidopsis thaliana*. *Plant Cell Physiol.*, 53: 1391–1403

Kim, K.S., S.H. Park and M.A. Jenks, 2007. Changes in leaf cuticular waxes of sesame (*Sesamum indicum* L.) plants exposed to water deficit. *J. Plant Physiol.*, 164: 1134–1143

Knoche, M., S. Peschel, M. Hinz and M.J. Bukovac, 2000. Studies on water transport through the sweet cherry fruit surface: characterizing conductance of the cuticular membrane using pericarp segments. *Planta*, 212: 127–135

Kosma, D.K., B. Bourdenx, A. Bernard, E.P. Parsons, S. Lu, J. Joubès and M.A. Jenks, 2009. The impact of water deficiency on leaf cuticle lipids of Arabidopsis. *Plant Physiol.*, 151: 1918–1929

Kosma, D.K., E.P. Parsons, T. Isaacson, S. Lü, J.K. Rose, M.A. Jenks, 2010. Fruit cuticle lipid composition during development in tomato ripening mutants. *Physiol. Plantarum*, 139: 107–117

Kunst, L. and L. Samuels, 2009. Plant cuticles shine: advances in wax biosynthesis and export. *Curr. Opin. Plant Biol.*, 12: 721–727

Kunst, L., A.L. Samuels and R. Jetter, 2005. The plant cuticle: formation and structure of epidermal surfaces. *In: Plant Lipids—Biology, Utilisation and Manipulation*, pp: 270–302. D. Murphy (ed.). Blackwell, Oxford, UK

Kurdyukov, S, A. Faust, C. Nawrath, S. Bär, D. Voisin, N. Efremova, R. Franke, L. Schreiber, H. Saedler, J.P. Métraux and A. Yephremov, 2006. The epidermis-specific extracellular *BODYGUARD* controls cuticle development and morphogenesis in Arabidopsis. *Plant Cell*, 18: 321–339

Laila, R., A.H. Robin, K. Yang, J.I. Park, M.C. Suh, J. Kim and I.S. Nou, 2017. Developmental and genotypic variation in leaf wax content and composition, and in expression of wax biosynthetic genes in *Brassica oleracea* var**.** *capitata. Front. Plant Sci.*, 7: 1972

Lee, J., K. Yang, M. Lee, S. Kim, J. Kim, S. Lim, G.H. Kang, S.R. Min, S.J. Kim, S.U. Park, Y.S. Jang, S.S. Lim and H.R. Kim, 2015. Differentiated cuticular wax content and expression patterns of cuticular wax biosynthetic genes in bloomed and bloomless broccoli (*Brassica oleracea* var. italica). *Process Biochem.*, 50: 456–462

Lee, S.B. and M.C. Suh, 2015. Advances in the understanding of cuticular waxes in *Arabidopsis thaliana* and crop species. *Plant Cell Rep.*, 34: 557–572

Lee, S.B., H. Kim, R.J. Kim and M.C. Suh, 2014. Overexpression of Arabidopsis MYB96 confers drought resistance in *Camelina sativa* via cuticular wax accumulation. *Plant Cell Rep.*, 33: 1535–1546

Lemieux, B., 1996. Molecular genetics of epicuticular waxes biosynthesis. *Trends Plant Sci.*, 1: 312–318

Li, L., D. Li, S. Liu, X. Ma, C.R. Dietrich, H.C. Hu, G. Zhang, Z. Liu, J. Zheng, G. Wang and P.S. Schnable, 2013. The maize *glossy13* gene, cloned via BSR-Seq and Seq-Walking encodes a putative ABC transporter required for the normal accumulation of epicuticular waxes. *PloS One*, 8: e82333

Li-Beisson, Y., B. Shorrosh, F. Beisson, F.X. Andersson, V. Arondel, P.D. Bates, S. Baud, D. Bird, A. DeBono, T.P. Durrett, R.B. Franke, I.A. Graham, K. Katayama, A.A. Kelly, T. Larson, J.E. Markham, M. Miquel, I. Molina, I. Nishida, O. Rowland, L. Samuels, K.M. Schmid, H. Wada, R. Welti, C. Xu, R. Zallot and J. Ohlrogge, 2013. Acyl-lipid metabolism. *Arabidopsis Book*, 11: e0161

Liu, F., X. Xiong, L. Wu, D. Fu, A. Hayward, X. Zeng, Y. Cao, Y. Wu, Y. Li and G. Wu, 2014. *BraLTP1*, a lipid transfer protein gene involved in epicuticular wax deposition, cell proliferation and flower development in *Brassica napus. PloS One*, 9: e110272

Liu, S., C.R. Dietrich and P.S. Schnable, 2009. DLA-based strategies for cloning insertion mutants: cloning the *gl4* locus of maize using Mu transposon tagged alleles. *Genetics*, 183: 1215–1225

Lü, S., H.Y. Zhao, E.P. Parsons, C.C. Xu, D.K. Kosma, X. Xu, D. Chao, G. Lohrey, D.K. Bangarusamy, G. Wang, R.A. Bressan and M.A. Jenks, 2012. The glossyhead1 allele of *ACC1* reveals a principal role for multidomain acetyl-coenzyme A carboxylase in the biosynthesis of cuticular waxes by Arabidopsis. *Plant Physiol.*, 157: 1079–1092

Lü, S., T. Song, D.K. Kosma, E.P. Parsons, O. Rowland and M.A. Jenks, 2009. Arabidopsis *CER8* encodes long-chain acyl CoA synthetase 1 (*LACS1*) that has overlapping functions with *LACS2* in plant wax and cutin synthesis. *Plant J.*, 59: 553–564

Mao, B., Z. Cheng, C. Lei, F. Xu, S. Gao, Y. Ren, J. Wang, X. Zhang, J. Wang, F. Wu, X. Guo, X. Liu, C. Wu, H. Wang and J. Wan, 2012. *Wax crystal-sparse leaf2*, a rice homologue of *WAX2/GL1*, is involved in synthesis of leaf cuticular wax. *Planta*, 235: 39–52

McNevin, J.P., W. Woodward, A. Hannoufa, K.A. Feldmann and B. Lemieux, 1993. Isolation and characterization of *eceriferum* (*cer*) mutants induced by T-DNA insertions in *Arabidopsis thaliana. Genome*, 36: 610–618

Nawrath, C., L. Schreiber, R.B. Franke, N. Geldner, J.J. Reina-Pinto and L. Kunst, 2013. Apoplastic diffusion barriers in arabidopsis. *Arabidopsis Book*, 11: e0167

Post-Beittenmiller, D., 1996. Biochemistry and molecular biology of wax production in plants. *Annu. Rev. Plant Biol.*, 47: 405–430

Pu, Y., J. Gao, Y. Guo, T. Liu, L. Zhu, P. Xu, B. Yi, J. Wen, J. Tu, C. Ma, T. Fu, J. Zou and J. Shen, 2013. A novel dominant glossy mutation causes suppression of wax biosynthesis pathway and deficiency of cuticular wax in *Brassica napus. BMC Plant Biol.*, 13: 1

Qin, B., D. Tang, J. Huang, M. Li, X. Wu and L. Lu, 2011. Rice *OsGL1-1* is involved in leaf cuticular wax and cuticle membrane. *Mol. Plant*, 4: 985–995

Racovita, R.C., S. Hen-Avivi, J.P. Fernandez-Moreno, A. Granel, A. Aharoni, R. Jetter, 2016. Composition of cuticular waxes coating flag leaf blades and peduncles of *Triticum aestivum* cv. Bethlehem. *Phytochemistry*, 130: 182–192

Razeq, F.M., D.K. Kosma, O. Rowland and I. Molina, 2014. Extracellular lipids of *Camelina sativa*: characterization of chloroform-extractable waxes from aerial and subterranean surfaces. *Phytochemistry*, 106: 188–196

Samuels, L., L. Kunst and R. Jetter, 2008. Sealing plant surfaces: cuticular wax formation by epidermal cells. *Plant Biol.*, 59: 683–707

Schneider, L.M, N.M. Adamski, C.E. Christensen, D.B. Stuart, S. Vautrin, M. Hansson, C. Uauy and P. von Wettstein-Knowles, 2016. The *Cer-cqu* gene cluster determines three key players in a β-diketone synthase polyketide pathway synthesizing aliphatics in epicuticular waxes. *J. Exp. Bot.*, 67: 2715–2730

Seo, P.J., S.B. Lee, M.C. Suh, M.J. Park, Y.S. Go and C.M. Park, 2011. The *MYB96* transcription factor regulates cuticular wax biosynthesis under drought conditions in Arabidopsis. *Plant Cell*, 23: 1138–1152

Smirnova, A., J. Leide and M. Riederer, 2013. Deficiency in a very-long-chain fatty acid *β*-ketoacyl-CoA synthase (*SlCER6*) of tomato impairs micro gametogenesis and causes floral organ fusion. *Plant Physiol.*, 161: 196–209

Smith, R.M., J.A. Marshall, M.R. Davey, K.C. Lowe, J.B. Power, 1996. Comparison of volatiles and waxes in leaves of genetically engineered tomatoes. *Photochemistry*, 43: 753–758

Stiles, W., 1994. *Principles of Plant Physiology*. Discovery Publishing House, New Delhi, India

Sturaro, M., H. Hartings, E. Schmelzer, R. Velasco, F. Salamini and M. Motto, 2005. Cloning and characterization of *GLOSSY1*, a maize gene involved in cuticle membrane and wax production. *Plant Physiol.*, 138: 478–489

Suh, M.C., A.L. Samuels, R. Jetter, L. Kunst, M. Pollard, J. Ohlrogge and F. Beisson, 2005. Cuticular lipid composition, surface structure, and gene expression in Arabidopsis stem epidermis. *Plant Physiol.*, 139: 1649–1665

Szafranek, B.M. and E.E. Synak, 2006. Cuticular waxes from potato (*Solanum tuberosum*) leaves. *Phytochemistry*, 67: 80–90

Tassone, E.E., A.E. Lipka, P. Tomasi, G.T. Lohrey, W. Qian, J.M. Dyer, M.A. Gore and M.A. Jenks, 2016. Chemical variation for leaf cuticular waxes and their levels revealed in a diverse panel of *Brassica napus* L. *Ind. Crop Prod.*, 79: 77–83

Tomasi, P., H. Wang, G.T. Lohrey, S. Park, J.M. Dyer, M.A. Jenks, H. Abdel-Haleem, 2017. Characterization of leaf cuticular waxes and cutin monomers of *Camelina sativa* and closely-related Camelina species. *Ind. Crops Prod.*, 98: 130–138

Tulloch, A.P. and R.O. Weenink, 1969. Composition of the leaf wax of Little Club wheat. *Can J. Chem.*, 47: 3119–3126

Velasco, R., C. Korfhage, A. Salamini, E. Tacke, J. Schmitz, M. Motto, F. Salamini, H.P. Doring, 2002. Expression of the *glossy2* gene of maize during plant development. *Maydica*, 47: 71–81

Viougeas, M.A., R. Rohr and A. Chamel, 1995. Structural changes and permeability of ivy (*Hedera helix* L.) leaf cuticles in relation to leaf development and after selective chemical treatments. *New Phytol.*, 130: 337–348

Vogg, G., S. Fischer, J. Leide, E. Emmanuel, R. Jetter, A.A. Levy and M. Riederer, 2004. Tomato fruit cuticular waxes and their effects on transpiration barrier properties: functional characterization of a mutant deficient in a very-long-chain fatty acid β-ketoacyl-CoA synthase. *J. Exp. Bot.*, 55: 1401–1410

von Wettstein-Knowles, P., 1971. The molecular phenotypes of the eceriferum mutants. *In: Barley genetics II*. pp: 146–193. Nilan, R.A. (ed.). Washington State University Press, Pullman, Washington, USA

von Wettstein-Knowles, P., 1995. Biosynthesis and genetics of waxes. *In: Waxes: Chemistry, Molecular Biology and Functions*, pp: 91–130. Hamilton, R.J. (ed.). Oily Press, Dundee, UK

Wang, M., H. Wu, J. Xu, C. Li, Y. Wang and Z. Wang, 2017. Five Fatty Acyl-Coenzyme A Reductases Are Involved in the Biosynthesis of Primary Alcohols in *Aegilops tauschii* leaves. *Front. Plant Sci.*, 8: 1012

Wang, W., X. Liu, X. Gai, J. Ren, X. Liu, Y. Cai, Q. Wang and H. Ren, 2015a. *Cucumis sativus* L. *WAX2* Plays a Pivotal Role in Wax Biosynthesis, Influencing Pollen Fertility and Plant Biotic and Abiotic Stress Responses. *Plant Cell Physiol.*, 56: 1339–1354

Wang, X., Y. Guan, D. Zhang, X. Dong, L. Tian and Q. LQ, 2017. A b-Ketoacyl-CoA Synthase Is Involved in Rice Leaf Cuticular Wax Synthesis and Requires a *CER2-LIKE* Protein as a Cofactor1. *Plant Physiol.*, 173: 944–955

Wang, M., Y. Wang, H. Wu, J. Xu, T. Li, D. Hegebarth, R. Jetter, L. Chen and Z. Wang, 2016. Three *TaFAR* genes function in the biosynthesis of primary alcohols and the response to abiotic stresses in *Triticum aestivum. Sci. Rep.*, 6: 25008

Wang, W., Y. Zhang, C. Xu, J. Ren, X. Liu, K. Black, X. Gai, Q. Wang and H. Ren, 2015b. Cucumber *ECERIFERUM1* (*CsCER1*), which influences the cuticle properties and drought tolerance of cucumber, plays a key role in VLC alkanes biosynthesis. *Plant Mol. Biol.*, 87: 219

Wang, Y., J. Wang, G. Chai, C. Li, Y. Hu, X. Chen and Z. Wang, 2015a. Developmental changes in composition and morphology of cuticular waxes on leaves and spikes of glossy and glaucous wheat (*Triticum aestivum* L.). *PloS One*, 10: e0141239

Wang, Y., M.L. Wang, Y.L. Sun, D. Hegebarth, T.T. Li, R. Jetter and Z. Wang, 2015b. Molecular characterization of *TaFAR1* involved in primary alcohol biosynthesis of cuticular wax in hexaploid wheat. *Plant Cell Physiol.*, 56: 1944–1961

Wang, Y., M.L. Wang, Y.L. Sun, Y.T. Wang, T.T. Li, G.Q. Chai, W. Jiang, L. Shan, C. Li, E. Xiao and Z. Wang, 2015c. *FAR5*, a fatty acyl-coenzyme A reductase, is involved in primary alcohol biosynthesis of the leaf blade cuticular wax in wheat (*Triticum aestivum* L.). *J. Exp. Bot.*, 66: 1165–1178

Wang, Z., O. Guhling, R. Yao, F. Li, T.H. Yeats, J.K.C. Rose and R. Jetter, 2011. Two oxidosqualene cyclases responsible for biosynthesis of tomato fruit cuticular triterpenoids. *Plant Physiol.*, 155: 540–552

Wen, M., C. Buschhaus and R. Jetter, 2006. Nanotubules on plant surfaces: Chemical composition of epicuticular wax crystals on needles of *Taxusbaccata* L. *Phytochemistry*, 67: 1808–1817

Weng, H., I. Molina, J. Shockey and J. Browse, 2010. Organ fusion and defective cuticle function in a *lacs1lacs2* double mutant of Arabidopsis. *Planta*, 231: 1089–1100

Wink, M., 1988. Plant breeding: importance of plant secondary metabolites for protection against pathogens and herbivores. *Theor. Appl. Genet.*, 75: 225–233

Xia, K., X. Ou, C. Gao, H. Tang, Y. Jia, R. Deng, X. Xu and M. Zhang, 2015. *OsWS1* involved in cuticular wax biosynthesis is regulated by *osa-miR1848. Plant Cell Environ.*, 38: 2662–2673

Xu, X., C.R. Dietrich, M. Delledonne, Y. Xia, T.J. Wen, D.S. Robertson, B.J. Nikolau and P.S. Schnable, 1997. Sequence analysis of the cloned *GLOSSY8* gene of maize suggests that it may code for a beta-ketoacyl reductase required for the biosynthesis of cuticular waxes. *Plant Physiol.*, 115: 501–510

Yeats, T.H. and J.K. Rose, 2013. The formation and function of plant cuticles. *Plant Physiol.*, 163: 5–20

Yu, D., K. Ranathunge, H. Huang, Z. Pei, R. Franke, L. Schreiber and C. He, 2008. *Wax Crystal-Sparse Leaf1* encodes a β-ketoacyl CoA synthase involved in biosynthesis of cuticular waxes on rice leaf. *Planta*, 228: 675–685

Zhang, H., O. Rowland and L. Kunst, 2005. Disruptions of the Arabidopsis enoyl-CoA reductase gene reveal an essential role for very-long-chain fatty acid synthesis in cell expansion during plant morphogenesis. *Plant Cell*, 17: 1467–1481

Zhang, X., Z. Liu, Q. Wang, S. Yang and H. Feng, 2013a. Fine mapping of *BrWax1*, a gene controlling cuticular wax biosynthesis in Chinese cabbage (*Brassica rapa* L. ssp. *pekinensis*). *Mol. Breed.*, 32: 867–874

Zhang, Z., W. Wang and W. Li, 2013b. Genetic interactions underlying the biosynthesis and inhibition of b-diketones in wheat and their impact on glaucousness and cuticle permeability. *PloS One*, 8: e54129

Zhang, Z., W. Wei, H. Zhu, G.S. Challa, C. Bi, H.N. Trick and W. Li, 2015. *W3* is a new wax locus that is essential for biosynthesis of β-diketone, development of glaucousness, and reduction of cuticle permeability in common wheat. *PloS One*, 10: e0140524

Zhang, Z., Z.J. Chen, L. Gan, H. Zhang, F.Q. Wu, Q.B. Lin, J.L. Wang, J. Wang, X.P. Guo, X. Zhang, Z.C. Zhao, C.L. Lei, S.S. Zhu, C.M. Wang and J.M. Wan, 2016. *OsHSD1*, a hydroxysteroid dehydrogenase, is involved in cuticle formation and lipid homeostasis in rice. *Plant Sci.*, 249: 35–45

Zhou, L.Y., E.D. Ni, J.W. Yang, H. Zhou, H. Liang, J. Li, D. Jiang, Z. Wang, Z. Liu and C. Zhuang, 2013. Rice *OsGL1-6* is involved in leaf cuticular wax accumulation and drought resistance. *PloS One*, 8: e65139

Zhou, Q., C. Li, K. Mishina, J. Zhao, J. Zhang, R. Duan, X. Ma, A. Wang, Q. Meng, T. Komatsuda and G. Chen, 2017. Characterization and genetic mapping of the β-diketone deficient eceriferum-b barley mutant. *Theor. Appl. Genet.*, 130: 1169

Zhou, X., L. Li, J. Xiang, G. Gao, F. Xu, A. Liu, X. Zhang, Y. Peng, X. Chen and X. Wan, 2015. *OsGL1-3* is involved in cuticular wax biosynthesis and tolerance to water deficit in rice. *PloS One*, 10: e116676

Zhu, X. and L. Xiong, 2013. Putative megaenzyme *DWA1* plays essential roles in drought resistance by regulating stress-induced wax deposition in rice. *Proc. Natl. Acad. Sci. USA,* 110: 17790–17795

Zygadlo, J.A., D.M. Maestri and N.R.Grosso, 1994. Alkane distribution in epicuticular wax of some Solanaceae species. *Biochem. Syst. Ecol.*, 22: 203–209