We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

6,300

Open access books available

172,000

International authors and editors

190M

Downloads

Our authors are among the

154

Countries delivered to

TOP 1%

most cited scientists

12.2%

Contributors from top 500 universities

WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index
in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.
For more information visit www.intechopen.com

Designing Novel Breeding Strategies for Producing High-Oil Crops Based on a Molecular Understanding of Triacylglycerol Metabolism

Masatake Kanai, Shoji Mano, Makoto Hayashi and Mikio Nishimura

Additional information is available at the end of the chapter

<http://dx.doi.org/10.5772/64465>

Abstract

Seeds are storage organ in plants and main resource of plant oils to human civilization and the demand of plant oils are increasing yearly and expansion of the production capacity is an urgent issue worldwide. Thus, it is necessary to improve oil yields per unit area and generation of crops with high-oil content is needed. *Arabidopsis thaliana* plays a vital role in advancement of genetics and molecular biology in plant sciences. The forward and reverse genetic approaches with *Arabidopsis* have provided an overview of triacylglycerol metabolism. The elucidation of the overview contributes to understanding of spatiotemporal regulation of a metabolic flow of triacylglycerol metabolism in plant cell. This understanding sheds light on bottlenecks in triacylglycerol biosynthesis and provides novel clues for increasing seed triacylglycerol content. Recent advance in metabolic engineering approaches demonstrate several evidences that triacylglycerol metabolism is coordinated with other metabolisms. Most notably, triacylglycerol biosynthesis competes with biosynthesis of starch or seed storage proteins. These studies indicate that alterations of the metabolic pathways to avoid the competitions could be a novel concept for increasing seed oil content.

Keywords: Seed, oil, Triacylglycerol, Metabolic engineering, *Arabidopsis thaliana*

1. Introduction

Seeds are storage organs in plants that accumulate massive quantities of carbohydrates, proteins, and oils, which are collectively referred to as seed storage reserves. Seed storage reserves are

utilized to supply energy and nutrients to the embryo during postgerminative growth until the plant acquires the ability to perform photosynthesis. Hence, seed storage reserves are an easily degradable source of organic matter for organisms.

Seeds are widely used as industrial materials. For example, plant oils are mainly derived from seeds. In 2014, the overall consumption of plant oils was 170,980 kt, which is nearly double that consumed in 2004. This increase is attributed to a greater demand for dietary oil, as well as industrial materials for use as carbon-neutral oils, representing an alternative to petroleum.

To improve oil productivity in plants, it is essential to increase seed yields in crops. However, the agricultural area worldwide has been flat for 40 years. Therefore, improving seed yield per plant has become increasingly important. Since increasing seed yield is one of the major issues in plant science, effective strategies for increasing yield have been investigated. Considering the many successes in the field of metabolic engineering of microorganisms, metabolic engineering represents a promising approach for increasing oil yields in seeds. Lipid metabolism has been extensively studied, and its metabolic pathways and regulatory systems have been elucidated. Additionally, in-depth analysis of crop genomes has been greatly expedited by recent advances in life science technologies (next-generation sequencing technology, genome editing, and so on). Therefore, numerous translational studies of model plant species have been performed, which have shed light on crop species. This chapter summarizes recent advances in our understanding of oil metabolism in seeds and introduces promising strategies for increasing oil production in crops.

2. Main components of seeds

Seeds accumulate carbohydrates, proteins, and oils as seed storage reserves. The ratios of these components dramatically differ among plant species, representing a major factor that determines the usage and application of seeds [1].

2.1. Cereal seeds

Starch consists of carbohydrates, which comprise polysaccharides composed of glucose [2–4]. Major cereal seeds, such as rice, wheat, and maize, contain large amounts of starch. Barley and rye seeds also accumulate starch as their main storage reserves. The starch contents in these cereal seeds are over 70%, whereas the contents of proteins and oils are less than 20% (**Table 1**). Therefore, cereal seeds represent an important energy source for animals.

2.2. Oil seeds

Oil seeds mainly accumulate lipids. Oil seeds are one of the major sources of plant oils. These seeds are compressed and heated to extract oils from their cells. Oil crops, which produce oil seeds, are cultivated worldwide. Oil palm, soybean, and rapeseed, the “big three”, represent three-quarters of total plant oil production (**Table 2**). Palm oil and olive oil are extracted from fruits, whereas many other oils are extracted from seeds (**Table 2**).

Grain	Carbohydrates	Proteins	Oils
wheat	72.2	10.6	3.1
rice	73.8	6.8	2.7
maize	70.6	8.6	5.0
barley	72.1	10.9	2.1
rye	70.7	12.7	2.7

Table 1. Carbohydrate, protein, and fat contents in cereal seeds. Values (%/w) in this table were extracted from the Food Composition Database of the Ministry of Education, Culture, Sports, Science, and Technology in Japan (<http://fooddb.mext.go.jp/index.pl>).

Plant species	Production (kilotons, 2013–2014)	Rate (% total plant oil production)	Storage organ
1 oil palm	59,360	34.09	fruit
2 soybean	44,439	25.52	seed
3 rapeseed	27,029	15.52	seed
4 sunflower	16,169	9.29	seed
5 cotton	4,859	2.79	seed
6 peanut	3,548	2.04	seed
7 olive	3,416	1.96	fruit
8 corn	3,148	1.80	seed

Table 2. Major oil crops and worldwide oil production in 2013–2014. The rates of total production and % total plant oil production are summarized for major oil crops; the values were obtained from FAOSTAT (<http://faostat.fao.org/>).

Species	Concentration (%)		Variety
	Oil	Protein	
soybean	20.1	41.4	Kariyutaka
rapeseed	41.0	25.6	Kizakinonatane
sunflower	33.8	30.4	Mammoth Russian
peanut	42.6	25.8	Omasari
sesame	49.9	21.6	–

Table 3. Oil and protein concentrations in major oil seed crops. The oil and protein concentrations were measured following the methods of Kanai et al. (2016). The variety of sesame used in this experiment was not identified.

2.2.1. Oil contents of oil seeds

Of the major oil crops, the oil content of oil palm fruit is over 50%, whereas that of soybean and rapeseed is 20.1% and 41%, respectively (**Table 3**) [5, 6]. The seed oil content in sunflower, peanut, and sesame is 33.8%, 42.6%, and 49.9%, respectively (**Table 3**). Thus, the seed oil contents of typical oil crops range from approximately 20–50%. These values are relatively low compared to the starch contents of rice, wheat, maize, and so on, suggesting that there is strong potential for increasing oil contents in seeds.

2.2.2. Protein contents of oil seeds

The protein content of major cereal seeds is less than 15% (**Table 1**), whereas the protein content of soybean and rapeseed, which are major suppliers of plant oils, is 41.4% and 25.6%, respectively (**Table 3**). Most oil seeds accumulate more proteins, i.e., seed storage proteins (hereafter referred to as SSPs), than cereal seeds.

3. Triacylglycerol metabolism in seeds

Lipids are essential components not only for animals, but also for plants. Various species of lipids are biosynthesized throughout plant organs. Some lipids, such as membrane lipids [7], cuticular waxes [8], and volatile oils, comprising fatty acids [9–11], alcohols, terpenes, and so on, are produced in plants in response to changes in the external environment. On the other hand, seed oils, the primary subject of this chapter, are composed of triacylglycerol (referred to hereafter as TAG) [12–14]. Except for palm oil, major seed oils are liquids at room temperature (**Figure 1A**). One molecule of TAG is composed of a glycerol backbone and three fatty acids (**Figure 1B**).

3.1. Triacylglycerol biosynthesis

TAG biosynthesis primarily involves two steps, i.e., fatty acid biosynthesis and the assembly of fatty acids with a glycerol backbone (**Figure 1C**).

Figure 1. Triacylglycerol in plant seeds. (A) Plant oil extracted from seeds. (B) Structure of TAG. TAG is produced by the assembly of one glycerol with three fatty acids. (C) Schematic diagram of TAG biosynthesis in plant cells.

3.2. Fatty acid biosynthesis

In plant cells, fatty acids are synthesized in plastids. Fatty acid biosynthesis begins with the carboxylation of acetyl-CoA to malonyl-CoA by acetyl-CoA carboxylase (reaction (1) in **Figure 2**). The malonyl group in malonyl-CoA is transferred to acyl carrier protein (hereafter referred to as ACP), producing malonyl-ACP (reaction (2) in **Figure 2**). Next, 3-keto-butryl-ACP is synthesized via the condensation reaction of malonyl-ACP with acetyl-CoA (reaction (3) in **Figure 2**). The 3-keto-butryl-ACP molecule is converted to butyryl-ACP via reduction and dehydration (reaction (4) in **Figure 2**). The provision of C2 units, reduction, and dehydration are repeated, leading to the elongation of the carbon chain of acyl-ACP (carbon chain elongation in **Figure 2**). Synthesized acyl-ACP is catalyzed by thioesterase to form free fatty acids, which are again converted to acyl-CoA and imported to the endoplasmic reticulum (hereafter referred to as ER).

Figure 2. Fatty acid biosynthesis in plastids: (1) acetyl-CoA carboxylase, (2) malonyl CoA:ACP transacylase, (3) 3-ketoacyl-ACP synthase, and (4) 3-ketoacyl-ACP reductase, 3-hydroxyacyl ACP dehydrase, and enoyl-ACP reductase.

3.3. Assembly of fatty acids with the glycerol backbone

Acyl-CoA is a donor of the acyl-group for TAG biosynthesis. *De novo* synthesis of TAG begins with the transfer of the acyl-group to the sn-1 position of glycerol 3-phosphate, leading to the production of lysophosphatidic acid (reaction (1) in **Figure 3**). Subsequently, phosphatidic acid is produced by the transfer of the acyl group to the sn-2 position (reaction (2) in **Figure 3**). Next, the sn-3 position of phosphatidic acid is dephosphorylated and converted to diacylglycerol (reaction (3) in **Figure 3**), and, finally, TAG is synthesized by the transfer of the acyl-group to the sn-3 position of diacylglycerol (hereafter referred to as DAG) (reaction (4) in **Figure 3**). In

addition to the *de novo* synthesis pathway, TAG is produced via an alternative pathway through phosphatidylcholine (hereafter referred to as PC) [15, 16]. PC, one of the main components of membrane lipids, is present in pools in the ER [16]. PC pools affect *de novo* TAG synthesis and the acyl-CoA pool in the cytosol to supply DAG as a precursor for TAG [16].

Figure 3. Triacylglycerol formation in the ER: (1) acyl-CoA:G3P acyltransferase, (2) acyl-CoA:LPA acyltransferase, (3) PA phosphatase, and (4) acyl-CoA:DAG acyltransferase.

3.4. Triacylglycerol accumulation

Synthesized TAGs accumulate in compartments in the ER (**Figure 1C**), which are converted to vesicles through budding. The vesicles then develop into oil-accumulating organelles, i.e., oil bodies [16–19]. Oil bodies are TAG storage organelles with a single layer membrane, whose major membrane protein is oleosin [18, 20, 21]. Oleosin blocks adhesion between neighboring oil bodies, which allows small oil bodies to be packed tightly together without adhesion in the cells of oil seeds [22].

4. Strategies for improving oil content by modifying triacylglycerol metabolism in oil seed crops

Since plant oils are commercially important, improving oil seed crops has long been a focus of breeders. Such breeding efforts, which began in ancient times, have led to the improvement of oil contents in several crops. In addition, the oil contents of the seeds of modern cultivars are significantly higher than those of wild species [23, 24]. Currently, many plant breeders have undertaken the challenge of further increasing seed oil contents. However, recent studies using quantitative trait loci analyses revealed that seed oil contents are controlled by many

genes with additive effects [25–28], suggesting that traditional breeding methods based on cross-fertilization may be inadequate for further increasing seed oil contents.

Figure 4. Seeds of *Arabidopsis thaliana*. (A) Microscopic image of Arabidopsis seeds. (B) Electron microscopic image of Arabidopsis seed cells. OB: oil body, an oil storage organelle in seeds. PB: protein body, an organelle that accumulates seed storage protein.

By contrast, genetic transformation may be an effective tool for increasing seed oil contents. The model plant *Arabidopsis thaliana*, a close relative of the major oil crop *Brassica napus*, produces typical oil seeds (Figure 4A and B). The use of Arabidopsis drastically simplifies both the processes of screening mutants with abnormal phenotypes and generating transformants, in which genes of interest are introduced. The use of Arabidopsis has accelerated the process of uncovering TAG metabolic pathways in plants. In fact, most genes encoding enzymes involved in TAG metabolism have been identified based on characterization of Arabidopsis mutants defective in TAG metabolism [29, 30]. Elucidating TAG metabolic pathways has revealed the limiting factors and key enzymes in this process and has led to the development of novel strategies for improving seed TAG contents. In this section, we review important strategies for improving TAG contents in seeds based on metabolic engineering approaches.

4.1. Enhancement of TAG biosynthesis

The TAG biosynthetic pathway, which extends across plastids, the cytosol, and the ER, involves many enzymes [30]. Additionally, DAG biosynthetic pathways include the *de novo* synthetic pathway and the phosphatidylcholine-derived pathway (hereafter referred to as PC-derived pathway), whose activities greatly differ among plant species [15, 16]. Thus, increasing the activity of enzymes in the *de novo* or PC-derived pathway does not always increase seed oil contents in every plant species. On the other hand, TAG production from DAG by acyl-CoA:DAG acyltransferase is a common pathway among plants. Overexpressing acyl-CoA:DAG acyltransferase significantly increases seed TAG contents in Arabidopsis [31] and in other plants species [32–37]. These findings indicate that the conversion of DAG to TAG is

one of the rate-limiting steps in the production of TAG in seeds, and they suggest that the acyl-CoA:DAG acyltransferase gene would be a promising target gene for increasing TAG contents.

4.2. Suppression of TAG degradation

Synthesized TAG is stored in oil bodies and degraded during germinative growth [17, 38, 39]. The TAG degradation pathway has also been uncovered, and most genes encoding enzymes in this pathway have been identified [17, 39]. The expression of these genes is upregulated after seed imbibition, and TAG degradation activity rapidly increases in imbibed seeds [40–42]. These genes are also expressed during seed development in several plants [43, 44]. In fact, TAG degradation occurs in developing seeds [45–47]. Therefore, the TAG degradation pathway is activated during seed development, and seeds lose some of the TAG synthesized during seed development. This finding suggests that suppressing TAG degradation would be a promising strategy for improving seed oil contents. Oil degradation begins with TAG hydrolysis via TAG lipase. TAG lipase was genetically identified as *SUGAR DEPENDENT 1* in Arabidopsis [48] and was subsequently identified in rapeseed and Jatropha [47, 49]. Suppressing *SUGAR DEPENDENT 1* expression significantly increases seed oil contents [47, 49]. These reports indicate that suppressing TAG degradation via suppressing *SUGAR DEPENDENT 1* expression may represent an effective strategy for increasing seed oil contents in oil seed crops.

5. Novel strategies focused on carbon flux in seeds

In production systems that utilize microorganisms and cultured cells for metabolic engineering, individual cells synthesize organic compounds, providing a limitless source of carbon in the medium. By contrast, the seed, representing only one of many plant organs, synthesizes storage reserves using a limited carbon source provided by photosynthesis. This limited carbon source is thought to represent another limiting factor to the use of seeds as production systems, in addition to the limited activities of metabolic enzymes. To utilize seeds for oil production, it is important to obtain a comprehensive understanding of the factors that regulate TAG production, including carbon flow, metabolite transport, TAG metabolism, compartmentation, competition between other reserves, and so on. Recent studies identifying other factors that limit oil production in seeds have opened up the possibility of developing novel strategies for improving seed oil contents.

5.1. Effective utilization of “the window” during seed development

Fertilized embryos grow into mature seeds through sequential development. Seed storage oils are produced only during a short period of seed development; high TAG biosynthesis activity only occurs for a short period of time. The master transcription factor of TAG biosynthesis, WRINKLED1 (hereafter referred to as WRI1), is expressed during this short period and induces the expression of genes related to fatty acid biosynthesis [50–54]. Although WRI1 positively regulates TAG biosynthesis, the overexpression of *WRI1* fails to increase TAG contents in seeds

[51, 53]. These findings suggest that, in order to increase seed TAG contents, it is necessary to reconsider the timing and duration of *WRI1* expression. Arabidopsis seeds initially accumulate TAG, followed by proteins [1]. A detailed analysis of seed development revealed that there is a time lag between the termination of TAG biosynthesis and the initiation of protein biosynthesis, i.e., there is a “window” period in the middle phase of seed development during which the activities of the TAG and protein synthesis pathways are low (**Figure 5**) [54]. Additionally, strong expression of *WRI1* during this window extends the duration of TAG biosynthesis and increases TAG contents in seeds [54], indicating that *WRI1* expression during this window effectively induces the TAG biosynthesis pathway. This finding has important implications for developing novel strategies for increasing seed TAG contents. The timing and duration of TAG and seed storage protein synthesis differ greatly among plant species. Therefore, identifying the window phase and fine tuning of *WRI1* expression are essential for generating oil crops with increased TAG contents in seeds.

Figure 5. Oil and protein biosynthesis during seed development. (A) Phases of oil and protein biosynthesis during seed development in *Arabidopsis thaliana*. (B) Schematic diagram of oil and protein biosynthesis during seed development in *Arabidopsis thaliana*.

5.2. Concentrating the carbon source into triacylglycerol biosynthesis by reducing the levels of other organic materials

Seeds accumulate various organic materials, including carbohydrates and proteins, in addition to TAG. Therefore, reducing carbohydrate and protein levels would help direct the carbon source into TAG biosynthesis.

5.2.1. Reducing polysaccharide biosynthesis

Seeds are generally covered with seed coats. Members of the Brassica family, including *Arabidopsis*, rapeseed, and so on, accumulate large amounts of mucilage consisting of pectin [55, 56]. Therefore, a considerable volume of the carbon source is consumed by mucilage production. Shen et al. reported that knockout of *GLABRA2*, which encodes a WRKY transcription factor controlling epidermal development, increases seed oil content in *Arabidopsis* [57]. Subsequent research demonstrated that increasing oil contents by suppressing *GLABRA2* expression reduces mucilage biosynthesis [58]. These findings indicate that reducing mucilage biosynthesis causes the carbon source to be directed into TAG biosynthesis. This transfer of the carbon source via reduction of polysaccharide levels has been verified in leaves. Starch comprises one of the main storage reserves in leaves. On the other hand, TAG biosynthesis activity is quite low in leaves. Therefore, to accumulate TAG in leaves, it is essential to both suppress starch biosynthesis and activate TAG biosynthesis. Sanjaya et al. found that overexpressing *WRI1* while suppressing the production of a small subunit of adenosine diphospho (ADP)-glucose pyrophosphorylase, a key enzyme for starch biosynthesis, significantly increases TAG contents in leaves [59]. These results indicate that reducing polysaccharide biosynthesis leads to the funneling of the carbon source into TAG biosynthesis, representing a possible novel strategy for increasing seed TAG contents in oil crops.

5.2.2. Reducing seed storage protein biosynthesis

As described above, oil seeds accumulate large amounts of seed storage proteins in addition to TAG. Crop breeders have reported a negative correlation between TAG and protein contents, especially in soybean [60, 61], suggesting that reducing the levels of seed storage proteins increases TAG contents in seeds. However, in *Arabidopsis*, knockout of genes encoding major seed storage proteins has little effect on TAG contents in seeds, although the protein content is reduced [54, 62]. These results may be due to the time lag between TAG and protein biosynthesis (**Figure 5**): little of the surplus carbon source derived from the suppression of protein biosynthesis is utilized for TAG biosynthesis because the TAG biosynthesis activity is quite low when protein synthesis is active in the late phase of seed development [54]. Therefore, simultaneously overexpressing *WRI1* and reducing protein synthesis greatly increases seed TAG contents due to effective utilization of the surplus carbon source [54]. This finding indicates that reducing protein synthesis indeed provides the surplus carbon source required for TAG production which, when combined with the simultaneous activation of TAG biosynthesis, leads to increased TAG production. This finding also demonstrates that the combination of these functional strategies has an additive effect on seed TAG content (**Figure 6**) [54, 63]. Breeding lines with reduced SSP content have been established in several crops [64, 65]. Introduction of *WRI1* into these breeding lines represents a potentially important strategy for high-oil seed production.

Figure 6. During the enlargement of seeds, the activation of TAG biosynthesis and the reduction of protein synthesis occur simultaneously. *FUS3pro:WRI1*; seeds from transgenic plants harboring *WRI1* under the control of the *FUS3* promoter in wild-type plants (Col-0): *WRI1* is expressed during the window phase. *FUS3pro:WRI1/12s1.4*; seeds from transgenic plants harboring *WRI1* under the control of the *FUS3* promoter in a double knockout mutant of *12S1* and *12S4*, encoding major seed storage proteins in *Arabidopsis thaliana*.

6. Conclusion

Molecular genetic analysis of *Arabidopsis* has led to the incredibly rapid elucidation of the mechanisms underlying the metabolism and regulation of seed storage reserves. The results from these basic studies provide powerful clues to help solve important issues in crop breeding. Optimizing the strategies developed based on the results of basic studies for use in crops will lead to crucial innovations for improving crop yields.

Author details

Masatake Kanai^{1,2}, Shoji Mano^{1,2,3}, Makoto Hayashi⁴ and Mikio Nishimura^{1*}

*Address all correspondence to: mikosome@nibb.ac.jp

1 Department of Cell Biology, National Institute for Basic Biology, Okazaki, Japan

2 Laboratory of Biological Diversity, Department of Evolutionary Biology and Biodiversity, National Institute for Basic Biology, Okazaki, Japan

3 Department of Basic Biology, SOKENDAI (The Graduate University for Advanced Studies), Okazaki, Japan

4 Department of Bioscience, Nagahama Institute of Bio-Science and Technology, Nagahama, Japan

References

- [1] Hills MJ: Control of storage-product synthesis in seeds. *Current Opinion in Plant Biology*. 2004;7:302–308. DOI: 10.1016/j.pbi.2004.03.003.
- [2] Santos-Mendoza M, Dubreucq B, Baud S, Parcy F, Caboche M, Lepiniec L: Deciphering gene regulatory networks that control seed development and maturation in *Arabidopsis*. *The Plant Journal*. 2008;54:608–620. DOI: 10.1111/j.1365-313X.2008.03461.x.
- [3] Smith AM, Zeeman SC, Smith SM: Starch degradation. *Annual Review of Plant Biology*. 2005;56:73–98. DOI: 10.1146/annurev.arplant.56.032604.144257.
- [4] Zeeman SC, Kossmann J, Smith AM: Starch: Its metabolism, evolution, and biotechnological modification in plants. *Annual Review of Plant Biology*, Vol 61. 2010;61:209–234. DOI: 10.1146/annurev-arplant-042809-112301.
- [5] Bourgis F, Kilaru A, Cao X, Ngando-Ebongue GF, Drira N, Ohlrogge JB, Arondel V: Comparative transcriptome and metabolite analysis of oil palm and date palm mesocarp that differ dramatically in carbon partitioning. *Proceedings of the National Academy of Sciences of the United States of America*. 2011;108:18186–18186. DOI: 10.1073/pnas.1115243108.
- [6] Dussert S, Guerin C, Andersson M, Joet T, Tranbarger TJ, Pizot M, Sarah G, Omore A, Durand-Gassel T, Morcillo F: Comparative transcriptome analysis of three oil palm fruit and seed tissues that differ in oil content and fatty acid composition. *Plant Physiology*. 2013;162:1337–1358. DOI: 10.1104/pp.113.220525.
- [7] Wang XM: Plant phospholipases. *Annual Review of Plant Physiology and Plant Molecular Biology*. 2001;52:211–231. DOI: 10.1146/annurev.arplant.52.1.211.
- [8] Samuels L, Kunst L, Jetter R: Sealing plant surfaces: Cuticular wax formation by epidermal cells. *Annual Review of Plant Biology*. 2008;59:683–707. DOI: 10.1146/annurev.arplant.59.103006.093219.
- [9] Dudareva N, Negre F, Nagegowda DA, Orlova I: Plant volatiles: Recent advances and future perspectives. *Critical Reviews in Plant Sciences*. 2006;25:417–440. DOI: 10.1080/07352680600899973.
- [10] Schwab W, Davidovich-Rikanati R, Lewinsohn E: Biosynthesis of plant-derived flavor compounds. *The Plant Journal*. 2008;54:712–732. DOI: 10.1111/j.1365-313X.2008.03446.x.
- [11] Weber H: Fatty acid-derived signals in plants. *Trends in Plant Science*. 2002;7:217–224. DOI:10.1016/S1360-1385(02)02250-1.
- [12] Baud S, Lepiniec L: Physiological and developmental regulation of seed oil production. *Progress in Lipid Research*. 2010;49:235–249. DOI: 10.1016/j.plipres.2010.01.001.

- [13] Durrett TP, Benning C, Ohlrogge J: Plant triacylglycerols as feedstocks for the production of biofuels. *The Plant Journal*. 2008;54:593–607. DOI: 10.1111/j.1365-313X.2008.03442.x.
- [14] Ohlrogge JB, Jaworski JG: Regulation of fatty acid synthesis. *Annual Review of Plant Physiology and Plant Molecular Biology*. 1997;48:109–136. DOI: DOI 10.1146/annurev.arplant.48.1.109.
- [15] Bates PD, Browse J: The significance of different diacylglycerol synthesis pathways on plant oil composition and bioengineering. *Frontiers in Plant Science*. 2012;3 DOI: 10.3389/fpls.2012.00147.
- [16] Bates PD, Stymne S, Ohlrogge J: Biochemical pathways in seed oil synthesis. *Current Opinion in Plant Biology*. 2013;16:358–364. DOI: 10.1016/j.pbi.2013.02.015.
- [17] Graham IA: Seed storage oil mobilization. *Annual Review of Plant Biology*. 2008;59:115–142. DOI: 10.1146/annurev.arplant.59.032607.092938.
- [18] Napier JA, Beaudoin F, Tatham AS, Alexander LG, Shewry PR: The seed oleosins: Structure, properties and biological role. *Advances in Botanical Research*, Vol 35. 2001;35:111–138. DOI: 10.1016/S0065-2296(01)35005-X.
- [19] Shimada TL, Hara-Nishimura I: Leaf oil bodies are subcellular factories producing antifungal oxylipins. *Current Opinion in Plant Biology*. 2015;25:145–150. DOI: 10.1016/j.pbi.2015.05.019.
- [20] Napier JA, Stobart AK, Shewry PR: The structure and biogenesis of plant oil bodies: The role of the er membrane and the oleosin class of proteins. *Plant Molecular Biology*. 1996;31:945–956. DOI: 10.1007/Bf00040714.
- [21] Tzen JTC, Cao YZ, Laurent P, Ratnayake C, Huang AHC: Lipids, proteins, and structure of seed oil bodies from diverse species. *Plant Physiology*. 1993;101:267–276.
- [22] Siloto RMP, Findlay K, Lopez-Villalobos A, Yeung EC, Nykiforuk CL, Moloney MM: The accumulation of oleosins determines the size of seed oilbodies in *Arabidopsis*. *The Plant Cell*. 2006;18:1961–1974. DOI: 10.1105/tpc.106.041269.
- [23] Skoric D: Achievements and future-directions of sunflower breeding. *Field Crops Research*. 1992;30:231–270. DOI: Doi 10.1016/0378-4290(92)90003-R.
- [24] Zheng P, Allen WB, Roesler K, Williams ME, Zhang S, Li J, Glassman K, Ranch J, Nubel D, Solawetz W, Bhatramakki D, Llaca V, Deschamps S, Zhong GY, Tarczynski MC, Shen B: A phenylalanine in dgat is a key determinant of oil content and composition in maize. *Nature Genetics*. 2008;40:367–372. DOI: 10.1038/ng.85.
- [25] Li HW, Zhao TJ, Wang YF, Yu DY, Chen SY, Zhou RB, Gai JY: Genetic structure composed of additive QTL, epistatic QTL pairs and collective unmapped minor QTL conferring oil content and fatty acid components of soybeans. *Euphytica*. 2011;182:117–132. DOI: 10.1007/s10681-011-0524-9.

- [26] Hwang EY, Song QJ, Jia GF, Specht JE, Hyten DL, Costa J, Cregan PB: A genome-wide association study of seed protein and oil content in soybean. *Bmc Genomics*. 2014;15 DOI: 10.1186/1471-2164-15-1.
- [27] Burns MJ, Barnes SR, Bowman JG, Clarke MHE, Werner CP, Kearsey MJ: QTL analysis of an intervarietal set of substitution lines in *Brassica napus*: seed oil content and fatty acid composition. *Heredity*. 2003;90:39–48. DOI: 10.1038/sj.hdy.6800176.
- [28] Jiang CC, Shi JQ, Li RY, Long Y, Wang H, Li DR, Zhao JY, Meng JL: Quantitative trait loci that control the oil content variation of rapeseed (*Brassica napus* L.). *Theoretical and Applied Genetics*. 2014;127:957–968. DOI: 10.1007/s00122-014-2271-5.
- [29] McGlew K, Shaw V, Zhang M, Kim RJ, Yang W, Shorrosh B, Suh MC, Ohlrogge J: An annotated database of Arabidopsis mutants of acyl lipid metabolism. *Plant Cell Reports*. 2015;34:519–32. DOI: 10.1007/s00299-014-1710-8.
- [30] Li-Beisson Y, Shorrosh B, Beisson F, Andersson MX, Arondel V, Bates PD, Baud S, Bird D, Debono A, Durrett TP, Franke RB, Graham IA, Katayama K, Kelly AA, Larson T, Markham JE, Miquel M, Molina I, Nishida I, Rowland O, Samuels L, Schmid KM, Wada H, Welti R, Xu C, Zallot R, Ohlrogge J: Acyl-lipid metabolism. *The Arabidopsis book / American Society of Plant Biologists*. 2013;11:e0161. DOI: 10.1199/tab.0161.
- [31] Jako C, Kumar A, Wei YD, Zou JT, Barton DL, Giblin EM, Covello PS, Taylor DC: Seed-specific over-expression of an Arabidopsis cDNA encoding a diacylglycerol acyltransferase enhances seed oil content and seed weight. *Plant Physiology*. 2001;126:861–874. DOI: 10.1104/Pp.126.2.861.
- [32] Vigeolas H, Waldeck P, Zank T, Geigenberger P: Increasing seed oil content in oil-seed rape (*Brassica napus* L.) by over-expression of a yeast glycerol-3-phosphate dehydrogenase under the control of a seed-specific promoter. *Plant Biotechnology Journal*. 2007;5:431–441. DOI: 10.1111/j.1467-7652.2007.00252.x.
- [33] Lardizabal K, Effertz R, Levering C, Mai J, Pedroso MC, Jury T, Aasen E, Gruys K, Bennett K: Expression of *Umbelopsis ramanniana* DGAT2A in seed increases oil in soybean. *Plant Physiology*. 2008;148:89–96. DOI: 10.1104/pp.108.123042.
- [34] Andrianov V, Borisjuk N, Pogrebnyak N, Brinker A, Dixon J, Spitsin S, Flynn J, Matyszczyk P, Andryszak K, Laurelli M, Golovkin M, Koprowski H: Tobacco as a production platform for biofuel: Overexpression of *Arabidopsis* DGAT and LEC2 genes increases accumulation and shifts the composition of lipids in green biomass. *Plant Biotechnology Journal*. 2010;8:277–287. DOI: 10.1111/j.1467-7652.2009.00458.x.
- [35] Wang ZK, Huang WJ, Chang JM, Sebastian A, Li YG, Li HY, Wu XX, Zhang BB, Meng FL, Li WB: Overexpression of SiDGAT1, a gene encoding acyl-CoA:diacylglycerol acyltransferase from *Sesamum indicum* L. Increases oil content in transgenic Arabidopsis and soybean. *Plant Cell Tissue and Organ Culture*. 2014;119:399–410. DOI: 10.1007/s11240-014-0543-z.

- [36] Taylor DC, Zhang Y, Kumar A, Francis T, Giblin EM, Barton DL, Ferrie JR, Laroche A, Shah S, Zhu W, Snyder CL, Hall L, Rakow G, Harwood JL, Weselake RJ: Molecular modification of triacylglycerol accumulation by over-expression of DGAT1 to produce canola with increased seed oil content under field conditions. *Botany-Botanique*. 2009;87:533–543. DOI: 10.1139/B08-101.
- [37] Weselake RJ, Shah S, Tang MG, Quant PA, Snyder CL, Furukawa-Stoffer TL, Zhu WM, Taylor DC, Zou JT, Kumar A, Hall L, Laroche A, Rakow G, Raney P, Moloney MM, Harwood JL: Metabolic control analysis is helpful for informed genetic manipulation of oilseed rape (*Brassica napus*) to increase seed oil content. *Journal of Experimental Botany*. 2008;59:3543–3549. DOI: 10.1093/jxb/ern206.
- [38] Mano S, Nishimura M: Plant peroxisomes. *Plant Hormones*. 2005;72:111–154. DOI: 10.1016/S0083-6729(05)72004-5.
- [39] Theodoulou FL, Eastmond PJ: Seed storage oil catabolism: A story of give and take. *Current Opinion in Plant Biology*. 2012;15:322–328. DOI: 10.1016/j.pbi.2012.03.017.
- [40] Carrera E, Holman T, Medhurst A, Peer W, Schmutz H, Footitt S, Theodoulou FL, Holdsworth MJ: Gene expression profiling reveals defined functions of the ATP-binding cassette transporter comatose late in phase ii of germination. *Plant Physiology*. 2007;143:1669–1679. DOI: 10.1104/pp.107.096057.
- [41] Kamada T, Nito K, Hayashi H, Mano S, Hayashi M, Nishimura M: Functional differentiation of peroxisomes revealed by expression profiles of peroxisomal genes in *Sesamum indicum* L. *Plant and Cell Physiology*. 2003;44:1275–1289. DOI: 10.1093/Pcp/Pcg173.
- [42] Kanai M, Nishimura M, Hayashi M: A peroxisomal ABC transporter promotes seed germination by inducing pectin degradation under the control of ABI5. *The Plant Journal*. 2010;62:936–947. DOI: 10.1111/j.1365-313X.2010.04205.x.
- [43] Turley RB, Trelease RN: Development and regulation of three glyoxysomal enzymes during cotton seed maturation and growth. *Plant Molecular Biology*. 1990;14:137-46.
- [44] Comai L, Dietrich RA, Maslyar DJ, Baden CS, Harada JJ: Coordinate expression of transcriptionally regulated isocitrate lyase and malate synthase genes in *Brassica-napus* L. *The Plant Cell*. 1989;1:293–300. DOI: 10.1105/Tpc.1.3.293.
- [45] Chia TYP, Pike MJ, Rawsthorne S: Storage oil breakdown during embryo development of *Brassica napus* (L.). *Journal of Experimental Botany*. 2005;56:1285–1296. DOI: 10.1093/jxb/eri129.
- [46] Chen H, Wang FW, Dong YY, Wang N, Sun YP, Li XY, Liu L, Fan XD, Yin HL, Jing YY, Zhang XY, Li YL, Chen G, Li HY: Sequence mining and transcript profiling to explore differentially expressed genes associated with lipid biosynthesis during soybean seed development. *Bmc Plant Biology*. 2012;12 DOI: 10.1186/1471-2229-12-122.
- [47] Kelly AA, Shaw E, Powers SJ, Kurup S, Eastmond PJ: Suppression of the *SUGAR-DEPENDENT1* triacylglycerol lipase family during seed development enhances oil

- yield in oilseed rape (*Brassica napus* L.). *Plant Biotechnology Journal*. 2013;11:355–361. DOI: 10.1111/pbi.12021.
- [48] Eastmond PJ: SUGAR-DEPENDENT1 encodes a patatin domain triacylglycerol lipase that initiates storage oil breakdown in germinating *Arabidopsis* seeds. *The Plant Cell*. 2006;18:665–675. DOI: 10.1105/tpc.105.040543.
- [49] Kim MJ, Yang SW, Mao HZ, Veena SP, Yin JL, Chua NH: Gene silencing of *Sugar-dependent 1* (*JcSDP1*), encoding a patatin-domain triacylglycerol lipase, enhances seed oil accumulation in *Jatropha curcas*. *Biotechnology for Biofuels*. 2014;7 DOI: 10.1186/1754-6834-7-36.
- [50] Maeo K, Tokuda T, Ayame A, Mitsui N, Kawai T, Tsukagoshi H, Ishiguro S, Nakamura K: An AP2-type transcription factor, *WRINKLED1*, of *Arabidopsis thaliana* binds to the AW-box sequence conserved among proximal upstream regions of genes involved in fatty acid synthesis. *The Plant Journal*. 2009;60:476–487. DOI: 10.1111/j.1365-313X.2009.03967.x.
- [51] Baud S, Wullemme S, To A, Rochat C, Lepiniec L: Role of *WRINKLED1* in the transcriptional regulation of glycolytic and fatty acid biosynthetic genes in *Arabidopsis*. *The Plant Journal*. 2009;60:933–947. DOI: 10.1111/j.1365-313X.2009.04011.x.
- [52] Baud S, Mendoza MS, To A, Harscoet E, Lepiniec L, Dubreucq B: *WRINKLED1* specifies the regulatory action of *LEAFY COTYLEDON2* towards fatty acid metabolism during seed maturation in *Arabidopsis*. *The Plant Journal*. 2007;50:825–838. DOI: 10.1111/j.1365-313X.2007.03092.x.
- [53] Cernac A, Benning C: *WRINKLED1* encodes an AP2/EREB domain protein involved in the control of storage compound biosynthesis in *Arabidopsis*. *The Plant Journal*. 2004;40:575–585. DOI: 10.1111/j.1365-313X.2004.02235.x.
- [54] Kanai M, Mano S, Kondo M, Hayashi M, Nishimura M: Extension of oil biosynthesis during the mid-phase of seed development enhances oil content in *Arabidopsis* seeds. *Plant Biotechnology Journal*. 2016;14:1241–1250. DOI: 10.1111/pbi.12489.
- [55] Western TL, Skinner DJ, Haughn GW: Differentiation of mucilage secretory cells of the *Arabidopsis* seed coat. *Plant Physiology*. 2000;122:345–355. DOI:10.1104/Pp.122.2.345.
- [56] Haughn GW, Western TL: *Arabidopsis* seed coat mucilage is a specialized cell wall that can be used as a model for genetic analysis of plant cell wall structure and function. *Frontiers in Plant Science*. 2012;3 DOI: 10.3389/Fpls.2012.00064.
- [57] Shen B, Sinkevicius KW, Selinger DA, Tarczynski MC: The Homeobox gene *GLABRA2* affects seed oil content in *Arabidopsis*. *Plant Molecular Biology*. 2006;60:377–387. DOI: 10.1007/s11103-005-4110-1.
- [58] Shi L, Katavic V, Yu YY, Kunst L, Haughn G: *Arabidopsis glabra2* mutant seeds deficient in mucilage biosynthesis produce more oil. *The Plant Journal*. 2012;69:37–46. DOI: 10.1111/j.1365-313X.2011.04768.x.

- [59] Sanjaya, Durrett TP, Weise SE, Benning C: Increasing the energy density of vegetative tissues by diverting carbon from starch to oil biosynthesis in transgenic *Arabidopsis*. *Plant Biotechnology Journal*. 2011;9:874–883. DOI: 10.1111/j.1467-7652.2011.00599.x.
- [60] Eskandari M, Cober ER, Rajcan I: Genetic control of soybean seed oil: II. QTL and genes that increase oil concentration without decreasing protein or with increased seed yield. *Theoretical and Applied Genetics*. 2013;126:1677–1687. DOI: 10.1007/s00122-013-2083-z.
- [61] Mao TT, Jiang ZF, Han YP, Teng WL, Zhao X, Li WB: Identification of quantitative trait loci underlying seed protein and oil contents of soybean across multi-genetic backgrounds and environments. *Plant Breeding*. 2013;132:630–641. DOI: 10.1111/pbr.12091.
- [62] Lin YM, Pajak A, Marsolais F, McCourt P, Riggs CD: Characterization of a cruciferin deficient mutant of *Arabidopsis* and its utility for overexpression of foreign proteins in plants. *Plos One*. 2013;8 DOI: 10.1371/journal.pone.0064980.
- [63] van Erp H, Kelly AA, Menard G, Eastmond PJ: Multigene engineering of boosts seed oil content in *Arabidopsis*. *Plant Physiology*. 2014;165:30-6. DOI: 10.1104/pp.114.236430.
- [64] Schmidt MA, Barbazuk WB, Sandford M, May G, Song ZH, Zhou WX, Nikolau BJ, Herman EM: Silencing of soybean seed storage proteins results in a rebalanced protein composition preserving seed protein content without major collateral changes in the metabolome and transcriptome. *Plant Physiology*. 2011;156:330–345. DOI: 10.1104/pp.111.173807.
- [65] Kawakatsu T, Hirose S, Yasuda H, Takaiwa F: Reducing rice seed storage protein accumulation leads to changes in nutrient quality and storage organelle formation. *Plant Physiology*. 2010;154:1842–1854. DOI: 10.1104/pp.110.164343.

IntechOpen