Chapter

Amaranth Seed Oil Composition

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Abstract

In this chapter, amaranth seed oil composition will be presented. The main component of this oil is triacylglycerols (TAGs). TAGs are composed of fatty acids, which have an important effect on oil stability, application, and nutritional properties. POL, PLL, POO, OLL, and LOO are the predominant TAGs in the amaranth seed oil. Linoleic acid (C18:2), oleic acid (C18:1), and palmitic acid (C16:0) are the predominant fatty acids present in the amaranth oil. Minor components of this oil are squalene, sterols, tocopherols, carotenoids, phospholipids, etc. Growth conditions of amaranth and extraction conditions can influence oil composition, which will be discussed in this chapter as well. Oil stability and quality parameters will be also discussed. The stability of this oil during different conditions of storage will be a part of this chapter.

Keywords: triacylglycerol, fatty acid, squalene, tocopherol, sterol

1. Introduction

Grain amaranth is considered as a gluten-free pseudocereal, which is a non-grass but cereal-like grain (true cereals are classified as grasses). It is suitable to be used as the celiac disease patient diet as it contains no gluten [1]. Among more than 60 species, the grain of *Amaranthus caudatus*, *Amaranthus hypochondriacus, Amaranthus cruentus, Amaranthus hybridus,* and *Amaranthus mantegazzianus* can be used as flour in some industries, such as bakery and confectionery. However, species of *Amaranthus retroflexus*, *Amaranthus viridis,* and *Amaranthus spinosus* are not safe to be consumed [2].

The amaranth grain is mainly composed of about 61.3–76.5% carbohydrate (mostly starch), 13.1–21.5% crude protein, 5.6–10.9% crude fat, 2.7–5% crude fiber, and 2.5–4.4% ash [3]. Proteins and lipids are two nutritiously important macromolecules of the amaranth grain. The content and even the quality of these two macronutrients are different from those with cereals. The amaranth grain has higher protein content in comparison to cereals. Lysine, which is the limiting amino acid in cereals, is found in higher amounts in amaranth grain. The high protein content of the amaranth grain is also evident from its high essential amino acid index (EAAI = 90.4%), which makes it comparable with egg protein [4].

In addition to protein content and special amino acid profile, amaranth grain usually contains 5–8% fat, which is important from the nutritional aspect [5]. However, *spinosus* and *tenuifolius* species can contain oil content as much as 17 and 19.3%, respectively. The fat content of the amaranth grain is dependent on the species, cultivars, and also accessions [6].

The fat content of amaranth grain is two to three times higher than cereals [7]. The oil is usually extracted from the grain by the solvent extraction method with the help of a non-polar organic solvent in a Soxhlet apparatus [8]. Supercritical carbon dioxide can be used as an alternative to traditional organic solvents for the extraction of the oil (supercritical fluid extraction method) [9, 10]. In the accelerated solvent extraction method, high pressure and temperature (even above the boiling point of the organic solvent) are used [6]. The oil yield with the Soxhlet method (62.1–75.7%) and accelerated solvent extraction method (65.1–78.1%) is almost similar; however, the latter is faster and uses lower organic solvent. The supercritical fluid extraction method has the lowest oil yield among the three methods (54.6–61.1%) [8].

Lipid fraction is mainly composed of triacylglycerols (TAGs) as the major component (around 80%) and other minor compounds, such as squalene, sterols, tocopherols, carotenoids, phospholipids, etc. [11]. Lipid fraction can also be divided into two groups: free lipids and bonded lipids. TAGs are the major free lipids, while phospholipids (up to 10.2% of total lipids) and glycolipids (6.4% of total lipid fraction) comprise the main part of the bounded lipids [11].

2. Triacylglycerol profile

TAGs are the major component of the amaranth oil, comprising 78–82% of the lipid fraction [11, 12]. Di- and monoacylglycerols comprise 5.1–6.5 and 3–3.5% of lipid fraction, respectively [11]. They are composed of fatty acids. Although the oxidative stability and the nutritional value of the oil are determined by the fatty acid profile, the functionality of oil is affected by the type and amount of TAGs [13]. The predominant structures in the amaranth oil are diunsaturated TAGs (UUS; 43.4–50.2%) and triunsaturated TAGs (UUU; 33–35.7%) [13].

The major TAG composition of *Amaranthus cruentus* is presented in **Table 1**. POL, PLL, POO, OLL, and LOO are dominant TAGs in the amaranth oil with carbon number ranging between 50 and 54 [7, 11, 13]. According to the TAG profile,

	Reference no. [7]	**Reference no.** [13]	**Reference no.** [11]
LLL	4	5.94	Not reported
OLL	12.1	10.97[a]	2.4
PLL	13.8	14.48[b]	16.7[b]
LOO	11.8	10.95[c]	2.6
POL	20[d]	16.69	25.4
PPL	7.5	7.01	22.6
OOO	7.9	4.82[e]	3.6
POO	12.5[f]	11.8[g]	16.7

M, myristic acid; P, palmitic acid; Po, palmitoleic acid; S, stearic acid; O, oleic acid; L, linoleic acid; Ln, linolenic acid.

[a] *OLL + OOLn*

[b] *PLL + PLnO*

[c] *LOO+PoOO*

[d] *POL + SLL*

[e] *OOO + MSO*

[f] *POO+SOL*

[g] *POO+PSL.*

Table 1.
Major triacylglycerol composition of the oil from Amaranthus cruentus*.*

amaranth oil is similar to corn and cottonseed oils [7, 14]. Like other vegetable oils, unsaturated fatty acids generally occupy the sn-2 position in the TAG structure of the amaranth grain oil. Linoleic acid and oleic acid are the two predominant fatty acids occupying the sn-2 position in the TAG structure of the amaranth grain oil, with percentages of 61.3 and 35.5, respectively, resembling cereals and also cottonseed and sesame seed oils [7]. Germination of the grain causes a decrease in TAG content as a result of increasing the lipase activity. Heat treatment of the grain, such as popping and cooking, decreases the TAG content [11].

3. Fatty acid composition

The fatty acid composition of the oil gives information about oxidative stability and nutritional quality. **Table 2** presents the fatty acid profile of some species of *Amaranthus* grain. Investigation on 104 genotypes from 30 species of *Amaranthus* grain revealed that palmitic acid, oleic acid, and linoleic acid were predominant in the oil with average percentages of 21.3, 28.2, and 46.5, respectively. Other fatty acids such as stearic and linolenic are also present in the oil, but in minor amounts [15]. The oil is highly unsaturated, containing more than 70% unsaturated fatty acids. The ratio of saturated to unsaturated fatty acids ranges between 0.26 and 0.32 [16]. The fatty acid profile of the amaranth oil is similar to that of cottonseed, buckwheat, and corn oils [13, 14].

	C16	C18:0	C18:1	C18:2	Source
A. cruentus	15.8–27	Tr-4.2	20.3–38.9	33.6–47	[7, 13, 15–19]
A. caudatus	12.3–20.5	2.2–4.7	23.8–32.9	35.6–49.8	[11, 18, 20]
A. hypochondriacus	17.9–24	0.9–3.7	16.3–33.7	38.9–52.5	[13, 15, 16, 18]
A. hybridus	18.6–22	1.3–4.4	18.7–26.3	47.4–55.9	[12, 15, 16, 18]
A. tricolor	19.5–24.3	1–3.6	25.9–27.5	46.4–51.5	[15, 16, 21]
A. dubius	15.7–25.9	0.7–4.1	14.8–30.5	46.9–53.5	[15, 18, 21]

Tr, trace.

Table 2.
Fatty acid composition of Amaranthus *species grain oil.*

4. Squalene

Squalene is a triterpene ($C_{30}H_{50}$) with six double bonds at carbon numbers 2, 6, 10, 14, 18, and 22, which is present in the unsaponifiable fraction of the oil (**Figure 1**). It is an intermediate molecule for the biosynthesis of phytosterols and cholesterol [22]. The main sources of squalene are whale and shark liver oil (40–86%). However, due to the concerns about the extinction of these marine animals, attempts are made to replace the animal source of squalene with a plant one [23].

Vegetable oils can be used as dietary sources of squalene. There is about 0.5% squalene in olive oil; around 0.03% in corn, hazelnut, and peanut oils; and 0.01% in grape seed and soybean oils [24]. The deodorizer distillates of oils such as olive oil, soybean oil, and palm fatty acids have higher amounts of squalene, containing 10–30, 1.8–3.5, and 0.2–1.3%, respectively [25].

Amaranth grain is another natural plant source of squalene. Although amaranth grain has lower oil content compared to the other oil-containing seeds, its oil fraction is a rich source of squalene [26] (**Table 3**). The high content of squalene in the amaranth grain oil makes it a unique component, which can be used to recover squalene. Although the direct derivation of squalene from amaranth seed is not economically affordable, the recovery of squalene from amaranth oil as a coproduct of starch production is advantageous [26]. An extensive study on 104 genotypes from 30 species of *Amaranthus* grain revealed the squalene concentration in the oil fraction was trace, 7.3% with an average of 4.2% [15]. The total content of squalene is dependent on the method of oil extraction. It has been demonstrated that the oil extracted with supercritical CO_2 had the highest squalene concentration (about 7%), followed by oil extracted by chloroform: methanol (2: 1 v/v; 6%) and cold-pressed oil (5.7%) [27]. However, in another investigation, it has been shown that squalene yield is the highest by accelerated solvent extraction method (4.4–4.7%), followed by Soxhlet (3.8–4.2%) and supercritical fluid extraction (3.3–3.8%) methods, respectively [8]. It should be mentioned that heat treatments such as cooking and popping the seeds cause an increase in the squalene concentration in the lipid fraction [11].

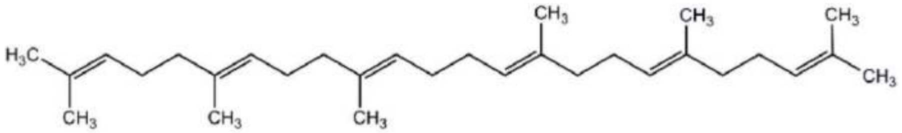


Figure 1.
Structure of squalene.

Amaranthus species	% Squalene	Reference
A. cruentus	6.56	[7]
	4.9	[11]
	5.74–6.95	[27]
	2.26–5.94	[17]
	4.2–5.44	[16]
	3.32–4.93	[15]
	9.16	[13]
	6.96	[14]
	5.29–6.25	[28]
A. hypochondriacus	4.74–6.98	[15]
	3.62–5.01	[16]
	9.96	[13]
	6.05–7.12	[28]
A. hybridus	5.23	[16]
	2.26–7.3	[15]
A. caudatus	0.67–8.19	[20]
	4.8	[11]
A. tricolor	4.73–5.75	[15]
	6.14	[16]
A. dubius	2.72–5.63	[15]

Table 3.
Squalene content of different species of Amaranthus *grain oil.*

5. Phytosterols

Plant sterols (phytosterols) are minor components of the vegetable oils, which comprise a large proportion of unsaponifiable fraction. They contribute to oxidative stability and extended shelf-life and have serum cholesterol-lowering properties [29, 30]. Phytosterols are found as 4-desmethysterols, 4-monomethylsterols, and 4, 4′-dimethylsterols. They can also be classified as free and esterified forms [31]. It has been reported that a large proportion of the phytosterols in amaranth oil are in esterified form and only low amounts are present in the free form (about 20%) [7]. However, in most of the vegetable oils, such as soybean, sesame, olive, cottonseed, safflower, palm and coconut oils, free sterols comprise the predominant form (54–85%) [32].

Total phytosterol content of the amaranth oil is between 1931 and 2762 mg/100 g oil [7, 21, 27, 33]. This level of phytosterol in amaranth oil is much higher than values established by Codex Alimentarius for most of common vegetable oils, such as coconut oil (40–120 mg/100 g), cottonseed oil (270–640 mg/100 g), flaxseed oil (230–690 mg/100 g), palm oil (30–70 mg/100 g), low-erucic acid

	A. cruentus						***A. dubius***	***A. tricolor***
	I	**II**	**III**	**IV**	**V**	**VI**		
Cholesterol	Tr	0.01	0.01	—	—	—	—	—
24-Methylene cholesterol	0.3	0.42	0.25	1.64	1.54	1.41	—	—
Campesterol	1.6	0.76	1	1.83	1.96	1.96	2.61	1.57
Stigmasterol	0.9	0.77	0.44	1.28	1.08	1.49	20.09	13.7
Δ^7-Ergostenol	—	23.8	25.3	—	—	—	—	—
α-Spinasterol	—	34.2[a]	26.3[a]	44.94[b]	53.24[b]	56.31[b]	—	—
Sitostanol	Tr	0.25	0.18	1.18	1.35	1.09	—	—
Δ^7-Campesterol	24.8	—	—	—	—	—	31.19	24.35
Clerosterol	42	—	—	—	—	—	1.58	3.71
Sitosterol	1.3	—	—	—	—	—	2	1.74
Δ^5-Avenasterol	2	1.68	2.34	0.79	0.74	0.35	24.27	30.76
$\Delta^{5,24}$-Stigmastadienol	Tr	1.89	2.26	1.92	2.04	1.45	13.66	10.73
Δ^7-Stigmastenol	15.2	22.2	24.4	15.02	14.48	11.74	0.69	1.52
Δ^7-Avenasterol	11.9	13.4	14.9	8.56	7.27	8.09	0.15	6.11
Δ^7-Ergosterol	—	—	—	17.29	16.32	16.12	—	—
Cycloartenol	1.63	—	—	2.26	0	0	—	—
Citrostadienol	1.3	—	—	3.3	0	0	—	—
Total sterol (mg/100 g)	2460	2730	2590	2490	1931	2140	2488.7	2762
Reference	[7]	[33]	[33]	[27]	[27]	[27]	[21]	[21]

I, hexane extracted oil; II, crude oil extracted by hexane at 50–55°C under atmospheric pressure; III, refined amaranth oil; IV, oil extracted by supercritical CO_2 under 306 atm and 50°C; V, cold press oil; VI, solvent extracted oil by chloroform: methanol (2: 1 v/v).

[a] *α-Spinasterol + sitosterol + chondrillasterol.*

[b] *α-Spinasterol + sitosterol.*

Table 4.
Phytosterol composition of different Amaranthus *species.*

rapeseed oil (450–1130 mg/100 g), safflower oil (210–460 mg/100 g), sesame oil (450–1900 mg/100 g), soybean oil (180–450 mg/100 g), and sunflower oil (240–500 mg/100 g) [34, 35]. However, wheat germ oil (4240 mg/100 g) and rice bran oil (1050–3100 mg/100 g) have total phytosterol content higher than amaranth oil [34, 36].

The phytosterol composition of the different *Amaranthus* species is presented in **Table 4**. The predominant phytosterol in the *Amaranthus cruentus* seed oil is the mixture of α-spinasterol and sitosterol [19, 21, 27]. Δ^7-Sterols, that is, Δ^7-stigmastenol and Δ^7-avenasterol and in some cases Δ^7-ergosterol and Δ^7-ergostenol, are also present in considerable amounts in *Amaranthus cruentus* seed oil [7, 27, 33]. However, Δ^7-campesterol and Δ^5-avenasterol are the major phytosterols of *Amaranthus dubius* and *Amaranthus tricolor* species. They also contain stigmasterol and $\Delta^{5,24}$-stigmastadienol in considerable concentrations [21].

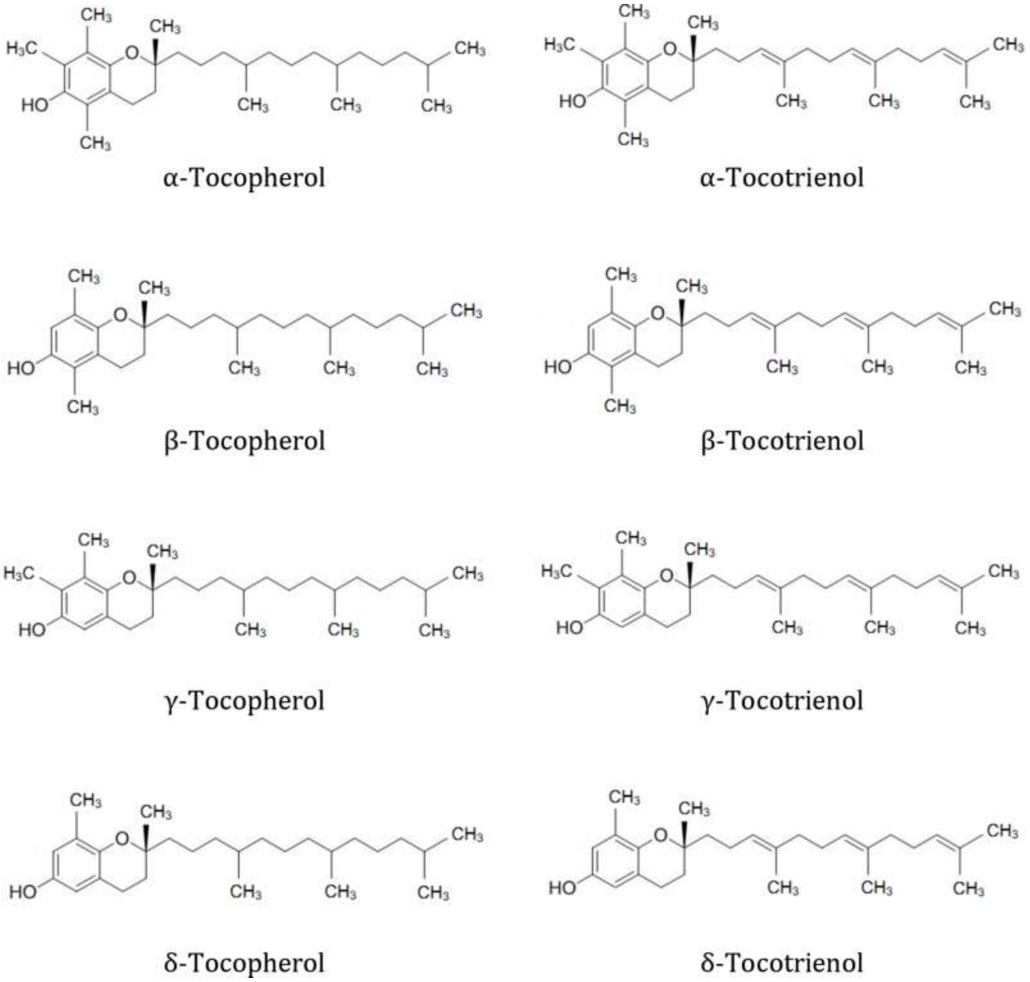
6. Tocopherols and tocotrienols

Tocopherols and tocotrienols (i.e., tocols) are a part of unsaponifiable fraction, which are forms of vitamin E and act as natural antioxidants in the vegetable oils. Tocotrienols are structurally similar to the tocopherols, except that tocotrienols have three double bonds within their phytol chains [37]. They have a chromanol ring attached to a phytol chain. Each of tocopherols and tocotrienols is divided into four subclasses, α-, β-, γ-, and δ- forms, which differ from each other as to the number of methyl groups on the chromanol ring [38]. The structure of eight homologs of tocopherols and tocotrienols is presented in **Figure 2**.

Tocopherols comprise the majority of the tocols in most of the common oils. However, tocotrienols are predominant in palm, rice bran, grape seed, and barely oils [39, 40]. It has been reported that amaranth seed has small or negligible amounts of tocotrienols [7, 18]. However, there are also reports that amaranth seed oil has tocotrienol content higher than some vegetable oils, such as soybean oil, peanut oil, and olive oil [21, 41].

γ-Tocopherol is the dominant tocol in most edible oils such as corn, soybean, rapeseed, sesame seed, and flaxseed oils. While α-tocopherol is the most abundant tocol in some vegetable oils such as safflower, sunflower, and olive oils [40]. Total and individual content of tocol homologs depends on the amaranth species, varieties, variation in analytical and extraction methods, and also growing location and cultivation conditions [18, 42]. The total tocol content of 21 amaranth accessions has been reported to be 31.5–78.3 mg/kg seed (wet basis), with an average of 49.4 mg/kg seed (wet basis) [18].

The study on the effect of dosages of fertilization with macronutrients on the tocopherol profile of two varieties of *Amaranthus cruentus* seeds revealed that the total tocopherol content was 48.6–79.9 mg/kg (dry matter) [42]. Applying various extraction methods, the determined contents of tocopherol homologs of the commercial and wild *Amaranthus caudatus* seed were 12.5–47.84 (mg/kg seed) α-tocopherol, 19.55–61.56 (mg/kg seed) β-tocopherol, 0.6–4.99 (mg/kg seed) γ-tocopherol, and 2.1–48.79 (mg/kg seed) δ-tocopherol [20]. Depending on the supercritical CO_2 extraction parameters, the tocopherol homologs of amaranth seed have s been reported as follows: 2.37–9.79 (mg/kg seed) α-tocopherol, 82.42–211.8 (mg/kg seed) β-tocopherol, 12.36–57.07 (mg/kg seed) γ-tocopherol, and 14.89–38.59 (mg/kg seed) δ-tocopherol [43]. The tocopherol composition of n-hexane extracted amaranth grain oil is presented in **Table 5**. It has been reported that the total tocopherol content of n-hexane extracted amaranth oil is between 656.8 and 2588 mg/kg oil [7, 21, 33].



α-Tocopherol

α-Tocotrienol

β-Tocopherol

β-Tocotrienol

γ-Tocopherol

γ-Tocotrienol

δ-Tocopherol

δ-Tocotrienol

Figure 2.
Structure of different forms of tocopherols and tocotrienols.

	α-T	**β-T**	**γ-T**	**δ-T**	**Total tocopherols**	**Source**
A. tricolor	74.2	157.9	17.4	407.2	656.8	[21]
A. dubius	135	245.7	22.3	376.4	779.5	[21]
A. cruentus	248	546	—	8	802	[7]
A. cruentus (crude oil)	392	299	1187	710	2588	[33]
A. cruentus (refined oil)	232	225	728	603	1788	[33]

α-T, α-tocopherol; β-T, β-tocopherol; γ-T, γ-tocopherol; δ-T, δ-tocopherol.

Table 5.
Tocopherol concentration (mg/kg oil) of n-hexane extracted oils from different species of amaranth grain.

7. Carotenoids

Carotenoids are essential photosensitizers, which have an important role in plant photosynthesis. They are also considered as provitamin A and possess antioxidative properties [44]. The two carotenoids lutein (3.55–4.44 mg/kg seeds) and zeaxanthin (trace to 0.32 mg/kg seeds) have been detected in amaranth seeds, lutein being the predominant one. β-Carotene, the most known carotenoid, has not been detected in amaranth seeds [45].

8. Phospholipids

Phospholipids are essential polar lipid materials that have an important role in biological membranes. TAGs are the major components of the nonpolar fraction of the lipid. However, phospholipids are the main compounds of the polar fraction of the lipids, which are considered as bound lipids. The phospholipid content of the amaranth grain oil has been reported to be in the range of 9.1–10.2% of total lipids [11].

9. Oxidative stability

Concerning the high concentration of squalene and tocopherols, the amaranth oil is expected to have good oxidative stability. Oxidative stability of amaranth oil was determined by monitoring the peroxide value at 60°C for 30 days. It has been reported that amaranth oil had good oxidative stability, even better than the oxidative stability of sunflower oil [11]. However, direct investigation of the stability of crude amaranth oil obtained opposite results. It has been reported that although amaranth oil contains high concentrations of squalene and tocopherols, which are strong antioxidants, it did not have good oxidative stability [46].

10. Conclusion

Amaranth grain contains 5–8% oil, which is mainly comprised of triacylglycerols (78–82%). The oil also contains important minor phytochemicals, such as squalene (up to 10%), phytosterols (2–3%), tocopherols, carotenoids, and phospholipids (up to 10%). The high content of tocopherols and squalene, which act as antioxidants, provides high oxidative stability for amaranth oil. The unique composition of amaranth seed oil makes it a useful ingredient in the food, pharmaceutical, and cosmetic industries.

Conflict of interest

The authors declare no conflict of interest.

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