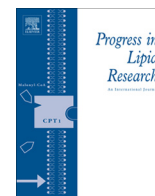




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Review

Biochemistry of high stearic sunflower, a new source of saturated fats

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ABSTRACT

Fats based on stearic acid could be a healthier alternative to existing oils especially hydrogenated fractions of oils or palm, but only a few non-tropical species produce oils with these characteristics. In this regard, newly developed high stearic oil seed crops could be a future source of fats and hard stocks rich in stearic and oleic fatty acids. These oil crops have been obtained either by breeding and mutagenesis or by suppression of desaturases using RNA interference. The present review depicts the molecular and biochemical bases for the accumulation of stearic acid in sunflower. Moreover, aspects limiting the accumulation of stearate in the seeds of this species are reviewed. This included data obtained from the characterization of genes and enzymes related to fatty acid biosynthesis and triacylglycerol assembly. Future improvements and uses of these oils are also discussed.

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1. Introduction

1.1. Generalities

Sunflower (*Helianthus annuus* L.) is a plant of the family of the Compositae (Asteraceae), belonging to the genus *Helianthus*. This plant is an annual crop, having a large yellow inflorescence containing small flowers that give rise to achenes containing a kernel rich in oil. This plant was first domesticated and cultivated by natives from Central and North America, and was introduced into Europe by Spanish explorers in the sixteenth century. The cultivation of sunflower was extended all over the world during the 19th and 20th centuries and is today the fourth largest vegetable oil product (after palm, soybean and rapeseed) and, thus, an important agricultural commodity. Today the main centers of production of sunflower are the USA, Ukraine and Argentina.

Sunflower is cultivated in temperate climates (temperature range between 20 and 25 °C). The plant grows better in dry climates with high solar irradiation and deep soils in which it is able to develop its long root system. The seeds of sunflower are produced within an achene and consist of a shell composed mainly of lignin and cellulose material and the kernel, which accounts for 80% of the total weight of the seeds and is rich in oil (up to 55% dry weight). The final oil content of sunflower seeds is usually around 50% w/w [1]. This oil is rich in linoleic acid, which accounts from 48% to 74% of the total fatty acids. It contains low levels of saturated fatty acids (mainly palmitic and stearic acids) and, unlike other seed oils such as soybean and rapeseed, negligible amounts of α -linolenic acid. The relative amount of linoleic compared to oleic acid is very variable and is related to temperature regulation of endogenous desaturases [2,3].

1.2. Sunflower biotechnology

As in the case of other oil crops, there has been increasing interest in manipulating the fatty acid composition of sunflower oil to produce oils with new properties and better performance for different applications. In this regard, sunflower has been differently affected by the biotechnology revolution that has taken place in the last 20 years. Thus, oil crops like rapeseed or soybean have been subject to many modifications and improvements through the application of genetic engineering. These manipulations involved the transfer of genes from other species or modification of the expression levels of endogenous genes by means of genetic transformation [4]. This has produced a variety of new lines with improved agronomical traits like resistance to herbicides and different oil compositions, varying from highly saturated oils to oils enriched in ω -3 very long chain fatty acids [5–8]. This revolution has taken place in a climate of some public controversy (especially in Europe) where there has been concern of the potential impact of genetically modified products (GMOs) on human health and natural ecosystems. Thus, in the labeling of food in Europe and in many places of Asia it is necessary to declare the presence of transgenic material in the corresponding formulations. In this regard, sunflower has not yet arrived into the GMO era. The reason is that sunflower is recalcitrant to transformation by means of *Agrobacterium* infection and is a plant very difficult to regenerate from cell cultures. Few advances have been made on transformation (either by floral dip or embryo infiltration) so there are no transgenic sunflower lines for commercial exploitation at the present time. However, these facts have not limited the improvement of sunflower lines using other techniques. Thus, sunflower is a plant easy to mutagenize by both physical and chemical methods. So, excellent results have been reported by using ethylmethane sulfonate or sodium azide to produce mutagenized populations of sunflower

seeds [9,10]. These chemical mutagens promote point mutations during DNA duplication. Furthermore, sunflower can also be mutagenized using ionizing radiation (X- or gamma-rays), which is responsible for the breakdown of large fragments of DNA, and thus cause deletions and mutations during the process of DNA repair. The population of mutagenized seeds can be later screened by TILLING [11] or, in the case of fatty acid compositional mutants, they can be found by direct GC analysis using half seeds (a non-destructive screening method in which the distal part of the seed is submitted to analysis and the apical part is used for propagation). Moreover, breeding of sunflower is eased by the simple diploid genetics of this plant. Another positive aspect of sunflower breeding is the relatively high genetic variability and the existence of several wild species of the genus *Helianthus* that could be a source of traits of interest to be transferred to commercial sunflower hybrids [12,13]. The breeding of sunflower, combined with the techniques of mutagenesis, have produced lines of sunflower with higher oil content, increased resistant to pests, dryness or salinity, as well as of mutants with altered fatty acid composition in their seeds. These improve agricultural, nutritional and technological characteristics of sunflower (and its oil). Moreover, since the crop is not a GMO it should, therefore, be more acceptable to a sceptical public (see above).

1.3. Sunflower mutant lines

In the last two decades many new lines of sunflower with modified fatty acid composition have been produced. The most successful development has been the production of high or ultra-high oleic mutants [14–16], which produce oils with fatty acid compositions similar to olive oil (Table 1, the basis of the healthy Mediterranean diet). Furthermore, the high levels of oleic acid make this oil more stable than most seed oils used for frying and storage. So, the cultivation and consumption of high oleic sunflower oil increases yearly all over the world with high acceptance both by industry and consumers. Moreover, more recent programs of sunflower improvement have produced a second generation of modified sunflower oils in which their fatty acid composition differed very significantly from that of regular sunflower (Table 1). These mutants display high levels of saturated fatty acids and therefore have oils with different physical properties, having a high potential for industrial applications [17,18]. The compositions of some of them are shown in Table 1, where typical compositions of high stearic and high palmitic sunflower mutants are shown. In a high linoleic background these oils can reach as much as 30% of either stearic or palmitic acids which, importantly, increases the solid fat content of such oils in a range of temperatures from 5 to 20 °C. In this regard, there is a deficiency of hard stock fats in the world because most currently-used vegetable oils are liquid. Solid fats are necessary for many industrial formulations including margarines, shortenings, fillings or confectionary [19]. These fats can be prepared from vegetable oils by hardening through hydrogenation. However, this process causes the production of unhealthy *trans*-fatty acids as a by-product [20]. The other alternative is based on the use of palm oil. Palm oil is rich in palmitate and has been extensively used for human nutrition. It is also a very productive crop [19,21]. However, the increased cultivation of palm trees in Indonesia is causing destruction of broad areas of tropical forest (accompanied by smoke pollution) giving ecological concerns to the production of these fats. There is some controversy as to whether high dietary palmitic acid can have an effect on plasma lipoproteins. However, no such potentially negative effects have been attributed to stearic acid. Thus, fats enriched in stearic acid, like high stearic sunflower, seem to be a promising alternative for a sustainable and 'healthy' source of solid fats. However, the original high stearic sunflower lines also contain high levels of linoleic acid, which diminishes

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