1 Fatty acids: structure, occurrence, nomenclature, biosynthesis and properties

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1.1 INTRODUCTION

Trans fatty acids have been present in the Western diet for as long as milk and butter have been staple commodities. However, in the last century with the discovery of catalytic hydrogenation by Sabatier and Senderens (Hastert, 1996; Hoffmann, 1989), food technologists came to recognise the improved physical characteristics which *trans* fatty acids could bestow on food products. The protection of the foodstuffs from the off flavours, which developed when highly unsaturated oils were incorporated into foods, was an added advantage which hydrogenation gave.

However, in the last 50 years, studies have been conducted into the effects of increased quantities of *trans* fatty acids on human health and nutrition. The result has been the requirement for food processors to be able to claim that they have low or no *trans* fatty acids in their products (Korver and Katan, 2006).

To appreciate the reason for this changed consideration, we first need to look at the constituents of oils and fats.

As far as the world production is concerned, the major vegetable oils and fats are soya, palm, rape (canola), sunflower, cotton, groundnut, coconut, palm kernel and corn. The major animal fats, by comparison, are butter, tallow, lard and fish. During the year 2005, the production split between the animal and vegetable groups of oils and fats was 78.5% vegetable oils and 21.5% animal fats. In Chapters 8 and 9 on applications, we will see how the two sources of oils and fats are utilised.

Oils and fats are made up of:

- lipids, *viz.* triacylglycerols (also called triglycerides), diacylglycerols (diglycerides), waxes, phosphoglycerols, sphingolipids, free fatty acids and hydrocarbons;
- certain vitamins;
- pigments and
- antioxidants.

These lipids cover a wide range of different chemical structures but there are two common features. Most lipids are water insoluble and they can all be biosynthetically related to fatty acids.

The triacylglycerols account for 90–95% by weight of oils and fats and in many senses are the most important part of these items of commerce. A generalised formula for a triacylglycerol is shown in Fig. 1.1.
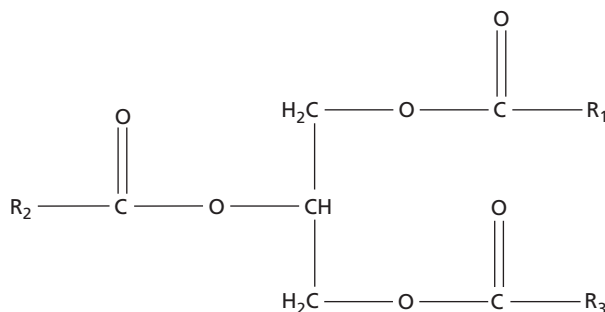
Fig. 1.1   General formula for a triacylglycerol.

If the fatty acids in this triacylglycerol, R_1COOH, R_2COOH and R_3COOH, are all identical, i.e. $R_1 = R_2 = R_3$, the triacylglycerol would be referred to as a monoacid triacylglycerol or a single-acid triacylglycerol. More usually, each triacylglycerol will have two or three different fatty acids.

Gunstone (1967) claimed that over 300 fatty acids were known in nature. By the time of a more recent book in 1996, he estimated that there were over 1000 fatty acids (Gunstone, 1996). Thus the diversity of these oils and fats (Gunstone, 2004) is considerable as will be manifested in Chapter 4 on analysis.

One simplifying feature is that the major fatty acids, in nature, have an even number of carbon atoms. In addition, there are usually only five to seven major fatty acids in most commercially important oils and fats.

1.2   FATTY ACID NOMENCLATURE

Fatty acid nomenclature is complicated by the fact that many acids were well known before any system of naming them had been determined. Thus the names of oleic, stearic and palmitic acids were well established before any rules were developed.

1.2.1   Saturated acids

Fatty acids are named according to the number of carbon atoms in the chain. In turn, the name of the fatty acids refers back to the name of the saturated hydrocarbon with the same number of carbon atoms. So stearic acid has 18 carbon atoms and is related to the alkane with 18 carbon atoms, i.e. octadecane. To obtain the name of the acid, the 'e' is removed from octadecane giving 'octadecan' and the ending 'oic' is added to indicate the carboxylic acid. Thus, octadecan(*e*) → octadecan(*oic*) acid → octadecanoic acid, which is the full and correct name for stearic acid.

Whilst it is convenient to use the trivial names, such as oleic and linoleic acid, many of the acids encountered later in our discussions have no simple trivial names. Even the use of formulae, as given in Tables 1.1 and 1.2, is not very quick and easy. An alternative shorthand method has been devised. This system reduces the acid to the minimum statement that is needed to define it.

Table 1.1 Structures of saturated fatty acids.

Shorthand notation	Chain length	Proper name	Common name	Structure
4:00	4	Butanoic	Butyric	
6:00	6	Hexanoic	Caproic	
8:00	8	Octanoic	Caprylic	
10:00	10	Decanoic	Capric	
12:00	12	Dodecanoic	Lauric	
14:00	14	Tetradecanoic	Myristic	
16:00	16	Hexadecanoic	Palmitic	
18:00	18	Octadecanoic	Stearic	
20:00	20	Eicosanoic	Arachidic	$CH_3(CH_2)_{18}COOH$
22:00	22	Docosanoic	Behenic	$CH_3(CH_2)_{20}COOH$

In the case of stearic acid, first the total number of carbon atoms in the chain is stated, i.e. 18, and then the number of double bonds is given which, in the case of stearic acid, is 0. The shorthand system then inserts a colon between the number of carbon atoms and the number of double bonds and so, for stearic acid, the shorthand is 18:0.

Stearic acid is shown in Table 1.1, where the long straight chain is given by the zigzag representation.

Some of the main straight-chain saturated fatty acids are also given in Table 1.1.

1.2.2   **Monounsaturated acids**

Oleic acid is an unsaturated fatty acid that can be represented by the formula shown in Fig. 1.2. Thus oleic acid has 18 carbon atoms, and it has one double bond at position 9 from the carboxyl end. Since oleic acid has 18 carbon atoms and one ethylenic double bond, the name is based on octadecene. In this instance the 'e' is removed and the ending for the carboxylic acid group octadecen(*e*) → octadecen(*oic*) acid. Thus is added 9-octadecenoic acid.

In the case of oleic acid, the double bond is in the *cis* configuration (also called the Z configuration from the German zusammen, meaning *together*). Thus to specify oleic acid precisely, the full name would be 9*c*-octadecenoic acid or 9*Z*-octadecenoic acid.

An isomer of oleic acid is elaidic acid, which has a *trans* double bond at the 9-position. The shorthand for this acid would therefore be 9*t*-octadecenoic acid. If the EZ system is to be used, the letter referring to the *trans* configuration is E, which stands for the German word entgegen, meaning *opposite*. These two acids are shown in Fig. 1.2.

From a chemist's point of view, the most important part of a fatty acid is the carboxylic acid group. The position of the double bond is therefore quoted with reference to the carboxylic acid group, i.e. 9 in the case of oleic acid. Using the shorthand method oleic acid is 18:1. Since the double bond is at the ninth carbon atom and the configuration of the double bond is *cis*, the name becomes 9c-18:1.

It is also possible to denote the position of the double bond by using the symbol Δ. Oleic acid is described as a Δ^9 acid, whilst petroselinic acid is a Δ^6 acid. Some of the main monounsaturated fatty acids are given in Table 1.2.
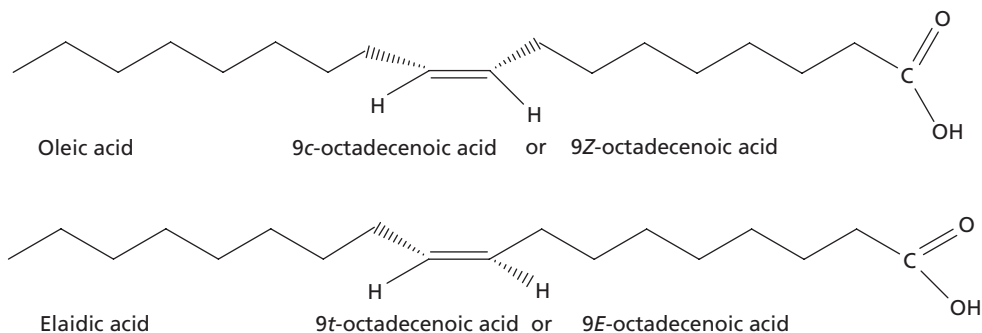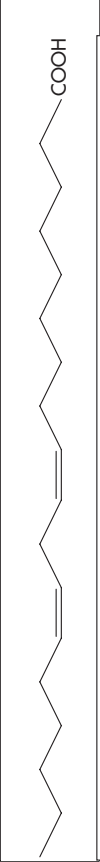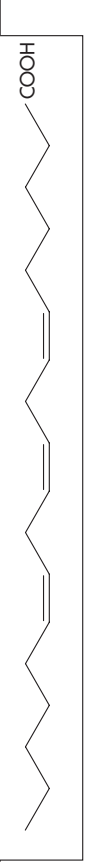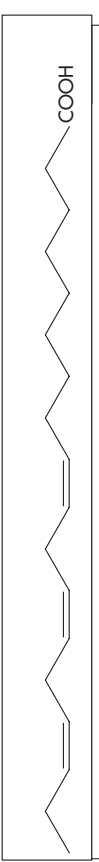

Fig. 1.2   Structures of oleic and elaidic acids.

Table 1.2 Structures of monoenoic acids.

Shorthand notation	Chain length	Proper name	Common name	Structure
14:1 5c	14	5c-Tetradecenoic		$CH_3 (CH_2)_7 CH=CH (CH_2)_3 COOH$
14:1 9c	14	9c-Tetradecenoic	Myristoleic	$CH_3 (CH_2)_3 CH=CH (CH_2)_7 COOH$
16:1 9c	16	9c-Hexadecenoic	Palmitoleic	$CH_3 (CH_2)_5 CH=CH (CH_2)_7 COOH$
18:1 6c	18	6c-Octadecenoic	Petroselenic	$CH_3 (CH_2)_{10} CH=CH (CH_2)_4 COOH$
18:1 9c	18	9c-Octadecenoic	Oleic	$CH_3 (CH_2)_7 CH=CH (CH_2)_7 COOH$
18:1 9t	18	9t-Octadecenoic	Erucic	$CH_3 (CH_2)_7 CH=CH (CH_2)_7 COOH$
18:1 11c	18	11c-Octadecenoic	Vaccenic acid	$CH_3 (CH_2)_5 CH=CH (CH_2)_9 COOH$
22:1 13c	22	13c-Docosenoic	Erucic	$CH_3 (CH_2)_7 CH=CH (CH_2)_{11} COOH$
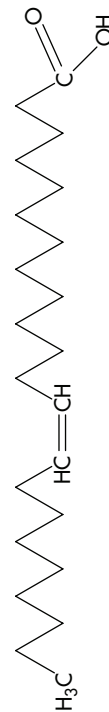
Table 1.3 Structures of polyunsaturated acids.

Shorthand notation	Name	Structure
9c,12c-18:2	Linoleic acid	
6c,9c,12c-18:3	γ-Linolenic acid	
9c,12c,15c-18:3	α-Linolenic acid	
9c,11t-18:2	Rumenic acid	

1.2.3 Diunsaturated acids

Linoleic acid is a diunsaturated acid with two double bonds and 18 carbon atoms and is named from the diunsaturated hydrocarbon octadecadiene (Table 1.3).

Octadecadien(*e*) → octadecadien(*oic*) acid → 9, 12-octadecadienoic acid

Again, the stereochemistry of the double bonds is known to be *cis* and so the correct name for linoleic acid is 9*c*,12*c*-octadecadienoic acid, with its shorthand name 9c,12c-18:2.

There is another system of numbering the position of the double bond, which came into operation because of the way in which the fatty acid is built up during biosynthesis. In Section 1.4, it will be seen that the starting point for biosynthesis is a two-carbon unit that becomes the methyl end of the final fatty acid. Each time that another two-carbon unit is added to the chain, the name of the new fatty acid would alter and the position of any double bond would also alter with respect to the chemist's fixed point, i.e. numbering from the carboxyl group.

It was recognised that it might be advisable to use a system of nomenclature, which started at the methyl end of the acid chain.

This is called the n-*x* system or the ω system. Thus linoleic acid is 9c,12c-18:2, where the carboxyl group is the starting point for the numbering. The alternative name for linoleic acid starts the numbering at the methyl end. In this case the double bond is now of six carbon atoms from the methyl group, and the position of the double bond is represented as n-6 or ω6. The ω tells us that we start counting from the methyl end. Linoleic acid would be described as ω6,9-18:2 in this alternative system. The ω notation for monounsaturated acids is given in Table 1.2.

Rumenic acid is a conjugated diene fatty acid, 9c,11t-18:2, which is dealt with in Chapter 3.

1.2.4 Triunsaturated acids

The structures of two of the major triunsaturated acids γ-linolenic acid and α-linolenic acid are given in Table 1.3. Their full names are 6*c*,9*c*,12*c*-octadecatrienoic acid and 9*c*,12*c*,15*c*-octadecatrienoic acid respectively. The name derived as above from octadeca with the trienoic added shows that there are three ethylenic double bonds.

Octadeca(ne) → octadecatrienoic acid → 9*c*,12*c*,15*c*-octadecatrienoic acid

1.3 OCCURRENCE

Of the saturated fatty acids, palmitic acid is the most widely occurring in both animal fats and vegetable oils, whilst stearic acid is found in lesser quantities in vegetable oils. Stearic acid is present in large quantities only in animal tallows and in vegetable fats, such as cacao butter and Borneo tallow. Butyric acid is found in butterfat (also referred to as anhydrous milk fat) produced from cow's milk. Caprylic, capric and myristic acids are present in coconut and palm kernel oil.

Oleic acid is the most widely distributed monounsaturated fatty acid. In some oils it is found in high proportions, ranging from 50 to 80%, e.g. olive, cashew and pistachio.

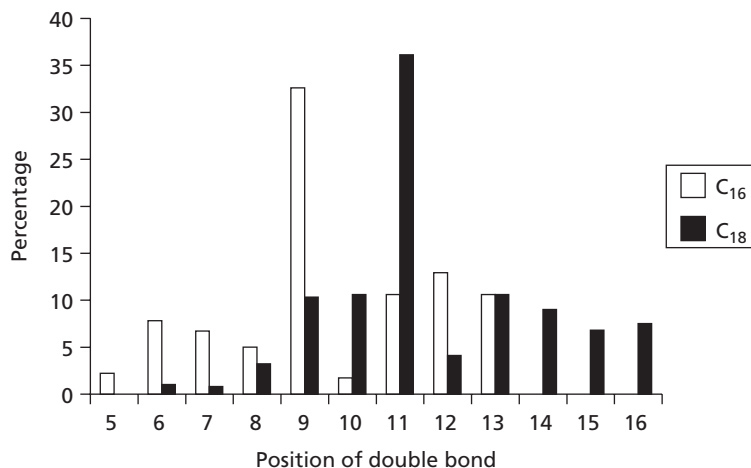**Fig. 1.3** *Trans* isomeric monoene C_{16} and C_{18} fatty acids in butter.

Whereas most of the unsaturated fatty acids in nature have a *cis* double bond, there are some acids that have the *trans* configuration. We can concern ourselves mainly with *trans* fatty acids from now on. There are three main sources of *trans* fatty acids in the human diet; *viz.*, they can be derived from animals or from the plant kingdom, or produced in the processing of oils and fats.

In animals, *trans* fatty acids are derived from dietary lipids. It is believed that biohydrogenation by bacteria in the rumen of the dietary lipids results in a mixture of *trans* fatty acids. Such fatty acids are found in all ruminant milk fats. Rumenic acid (9c,11t-18:2) is the major conjugated fatty acid in ruminant fats. Rossell (2001) reported the *trans* content of subcutaneous adipose tissue in beef, sheep and pig to be 1.3–6.6%, 11.0–14.6% and 1.1–1.4% by weight respectively. In the case of farm animals, where the feed may contain *trans* fatty acids, the animal will metabolise some of the *trans* fatty acids and place some *trans* fatty acids in the adipose tissue.

Hay and Morrison (1970) showed that amongst the *trans* isomers in butterfat, the monenoic C_{16} and monoenoic C_{18} are the major components (Fig. 1.3). The major isomer for C_{16} is palmitelaidic acid Δ^9 (32%) and for C_{18} *trans* vaccenic acid Δ^{11} (36.1%).

Trans fatty acids in most vegetable oils are present, if at all, in very minor proportions and in some oils, at the trace level.

In the vegetable kingdom, *trans* fatty acids do occur naturally and sometimes in significant quantities; i.e. there is 6–12% of eleostearic acid 9c,11t,13t-18:3 in cherry oils, which have now been accepted as safe for food oils (Comes *et al.*, 1992).

Petroselaidic acid, 6t-18:1, is found along with petroselinic acid in *Heracleum nipponicum, Conium maculatum, Phelopterus litoralis, Ligusticum acutifolium, Bupleurum falcatum, Osmorhiza aristata, Conioselinum univittatum, Hedera japonica, Panax schinseng* and *Aralia elata* (Placek, 1963).

In the plant kingdom, conjugated triene fatty acids often have one or more *trans* double bonds, e.g. jacaric acid 8c,10t,12c-18:3, calendic acid 8t,10t,12c-18:3, catalpic acid 9t,11t,13c-18:3, punicic acid 9c,11t,13c-18:3 and β-eleostearic acid 9t,11t,13t-18:3. There are also conjugated tetraenoic acids α- and β-parinaric acids 9c,11t,13t,15c-18:4 and 9t,11t,13t,15t-18:4 respectively. In addition the biosynthetic pathways given in Section 1.3

involve *trans* double bonds even when *cis* double bonds are being generated. So it can be seen that *trans* fatty acids occur naturally in both animals and plants.

In 1983, Sommerfeld stated that 'hardened oils do NOT contain *trans* fatty acids isomers other than those produced by the microflora of ruminants. Therefore claims that *trans* fatty acids isomers are "synthetic" "non-physiologic" or "unnatural" are unjustified if these words are used to imply "not produced by the living organism" '.

The presence in nature of conjugated linoleic acid double bonds contains *trans* which is further confirmation of Sommerfeld's statement. Conjugated linoleic acid is covered in full in Chapter 3.

The third source of *trans* fatty acids in foods is where they are produced in processing. Why was hydrogenation introduced into the oils and fats industry? Initially, it was to remedy a shortage of solid fats.

At its simplest, hydrogenation is the addition of two hydrogen atoms across the ethylenic double bond of the fatty acid. It was recognised that the more unsaturated the fatty acid, the more likely it was for the fatty acid to be oxidised, which leads to oxidative rancidity. By removing two double bonds from linolenic acid, a monoenoic acid would be formed, which would resist oxidation better.

$$\text{Triene} \rightarrow \text{diene} \rightarrow \text{monoene} \rightarrow \text{saturated acid} \tag{1.1}$$
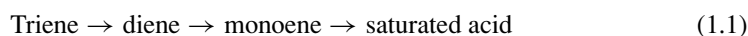
If the hydrogenation could proceed by the route suggested by Equation 1.1, the triene linolenic acid would yield the saturated acid, i.e. stearic acid. However, under industrial conditions, hydrogenation with a nickel catalyst is partial, giving rise to a mixture of products. From the above, it is still not obvious why there should be any *trans* fatty acid formed.

Dijkstra (2002) suggested an amendment to the Horiuti–Polanyi mechanism in which the monoene M forms a semihydrogenated intermediate MH (Eq. 1.2).

$$M + H \rightarrow MH, \quad \text{where M represents monoene} \tag{1.2}$$

Dijkstra explains that the hydrogen concentration is too low for these intermediates to go on to form stearic acid. In turn this allows dissociation to occur as in Eq. 1.3.

$$MH \rightarrow M + H \tag{1.3}$$
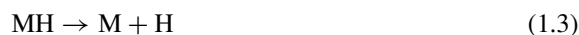
When an individual acid, e.g. oleic acid, is considered in these reactions, the changes can be represented as shown in Fig. 1.4.

When a fatty acid with a single *cis* double bond is partially hydrogenated, adsorption to (Step 1) and desorption from (Steps 3–5) the catalyst surface occurs, which produces a mixture of fatty acids. Some of the acids have a *trans* double bond. It is believed that the adsorption mechanism (Step 1) involves the formation of carbon─nickel bonds between the metal catalyst and the carbon atoms of the Δ^9 double bond C_9 and C_{10} to give a structure (a).

One hydrogen atom is then transferred (Step 2), probably from a Ni–H atom on the surface of the catalyst near the adsorbed fatty acid, to the carbon atom C_9 to give structure (b). If the addition goes further, another hydrogen atom is added, the C─Ni bond is broken and the hydrogen adds to C_{10}, with the formation of stearic acid as a desorption (Step 3).
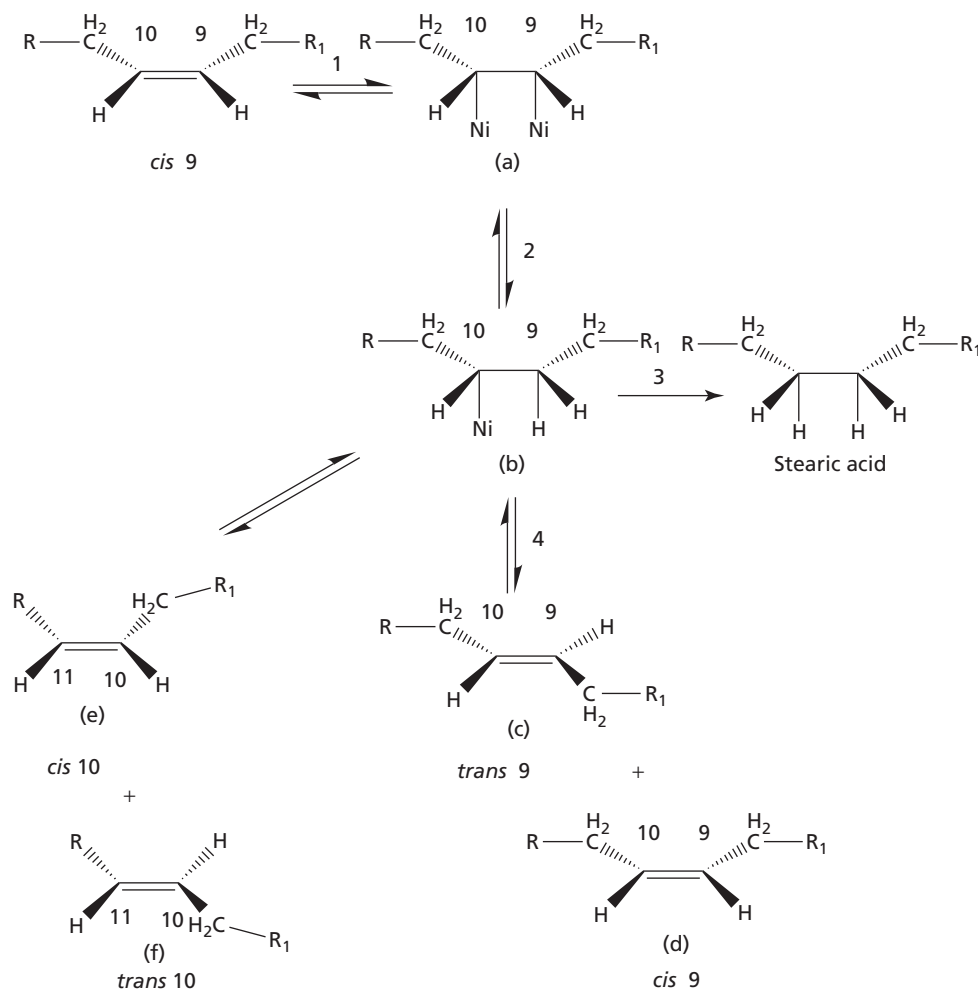
Fig. 1.4 Partial hydrogenation of oleic acid, where $R_1 = (CH_2)_7COOH$ and $R_2 = CH_3(CH_2)_7$.

The interactions between these species are reversible (Fig. 1.4), with the result that in structure (b) the hydrogen from C_9 and the C—Ni bond can be eliminated to reform a double bond between C_9 and C_{10} (Step 4). This results in a mixture of *cis* and *trans* isomers (c) and (d). It is also possible that when the C—Ni bond breaks, the hydrogen which is involved in the elimination comes from the C_{11}. This will produce a mixture of *cis* and *trans* Δ^{10} monoenes (e) and (f).

The production of the *trans* isomers can be seen more easily in Fig. 1.5.

The *cis* monoene, in an addition reaction (Step 1), gives the intermediate with two Ni atoms (a). Structure (a) can now react with a hydrogen on a neighbouring nickel atom, as explained above, to form the semihydrogenated intermediate (b) (Step 2); this is the structure MH in Equation 1.2. The elimination of the Ni–H atoms from (b) with the breaking of a C—H bond and a Ni—H bond results in the reformation of the *cis* double bond between C_9 and C_{10} (Step 3). This step is a desorption from the catalyst surface.
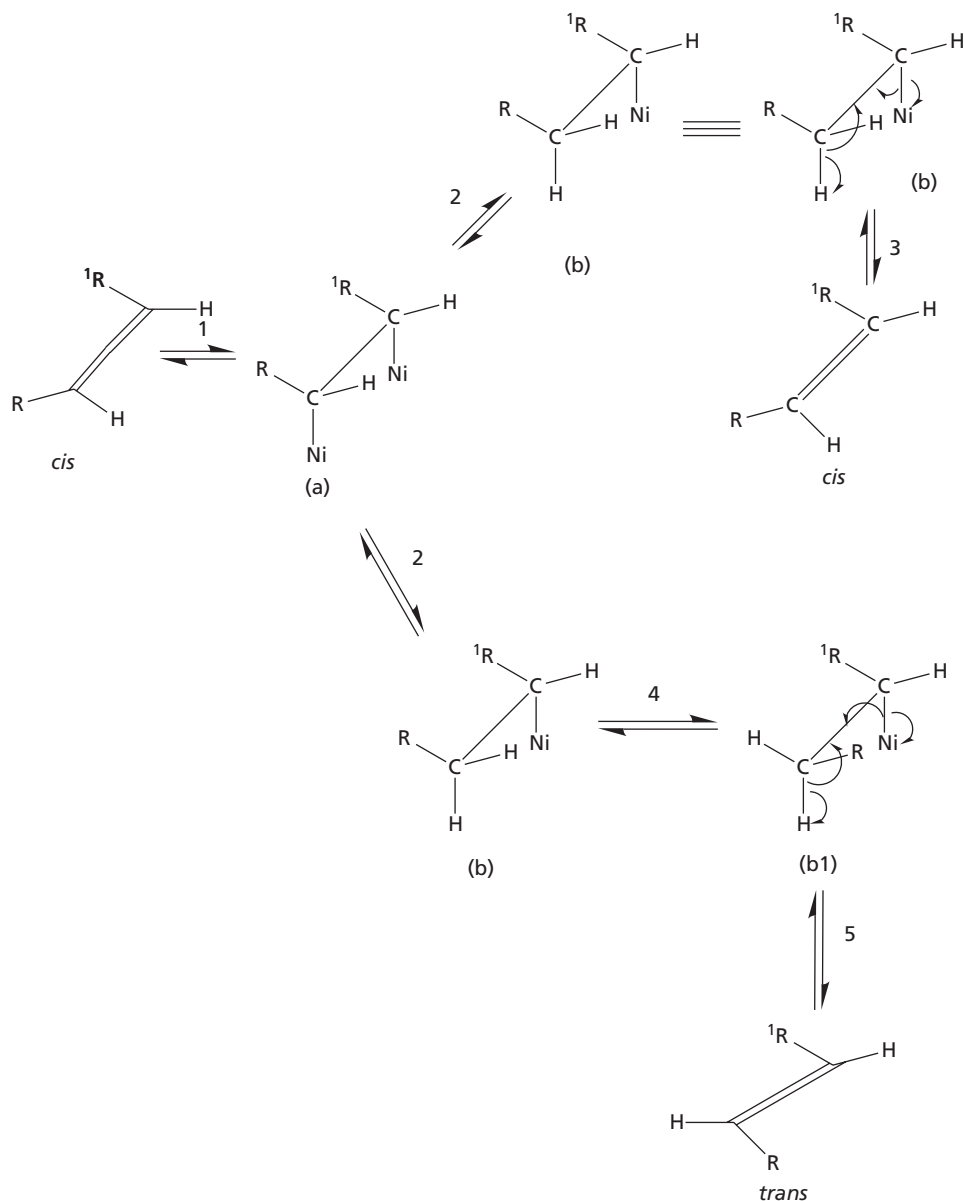
Fig. 1.5 Partial hydrogenation of oleic acid, where $R_1 = (CH_2)COOH$ and $R_2 = CH_3(CH_2)$, showing free rotation at Step 4.

However the semihydrogenated intermediate (b) can undergo free rotation about the C_9 to C_{10} bond (Step 4) when the fatty acid is attached to the metal atom at just one point to give the conformation (b1). When the Ni–H atoms from (b1) are eliminated in a desorption (Step 5), it is a *trans* isomer which is formed.

The oils and fats industry and catalyst manufacturers are working to permit hydrogenation without the production of *trans* fatty acids (see Chapter 6).

As will be explained later in Chapters 5, 7 and 9, industrial processors have reduced the level of *trans* fatty acid in foodstuffs from this source.

In processing, thermally induced geometrical isomerisation can occur as described in Chapter 5.

1.4 FATTY ACID BIOSYNTHESIS

1.4.1 Saturated fatty acids

When the pathways for fatty acid synthesis were being elucidated, it was realised that fatty acids were built up from acetic acid units. This finding made it easy to understand why so many naturally occurring acids had an even number of carbon atoms. It was subsequently found that only two carbon atoms of palmitic acid came directly from acetic acid, the carbons at positions 15 and 16 from the carboxyl end. The remainder of the carbon atoms came from malonyl coenzyme (CoA), as in Equation 1.4 (Gurr and James, 1980).

$$CH_3COOH + 7CH_2(COOH)_2 \rightarrow CH_3(CH_2)_{14}COOH \tag{1.4}$$

The synthesis of palmitic acid is carried out by fatty acid synthetase (FAS). In the case of FAS in *Escherichia coli*, the enzymes involved in individual steps are shown in Fig. 1.6. Malonyl CoA:ACP transacylase (a) activates the malonyl unit, whilst acetyl CoA:ACP transacylase (b) activates the acetic acid unit. The joining of these two activated forms to form a C_4 unit is catalysed by 3-ketoacyl-ACP synthetase (c). The ketone group in this C_4 unit is then reduced to a hydroxyl group in the presence of 3-ketoacyl-ACP reductase (d). It is in the next step that we see the formation of a *trans* fatty acid derivative as the hydroxyl group, and a hydrogen is eliminated in the presence of 3-hydroxyacyl-ACP dehydrase (e). The *trans* fatty acid is not released as such but the double bond is reduced to give a new C_4 fatty acid still in the activated form of ACP under the influence of enoyl-ACP reductase (f). These same reactions are performed to convert the C_4 up to the normal C_{16} palmitic acid, with the addition of further six malonyl units. This is the natural end point of this series of reactions, and longer chain length fatty acids depend on elongation reactions using FAS III.

From the viewpoint of *trans* fatty acids, it is important to recognise that the system accepts and utilises *trans* acids; i.e. *trans* fatty acids are not unnatural.

There are in fact three fatty acid synthetases FAS I, II and III. Type I consists of large molecular mass multifunctional proteins containing covalently bonded acyl carrier proteins (ACP) and is found in animals. Type II consists of individual enzymes that normally act as one complex and are found in bacteria and plants (Gunstone *et al.*, 1994). Type III FAS can elongate already formed fatty acids. The differences in these three synthetases relate to the subunits and the sequence of domains.

1.4.2 Monoenoic fatty acids

In the case of monoenoic acids in *E. coli* formed by type II FAS, there is a branch point when the chain length reaches ten carbon atoms, in an anaerobic pathway. In Fig. 1.7, it can be seen that when the dehydrase enzyme has worked on β-hydroxydecenoyl ACP, the resulting *trans*-2-decenoyl ACP can be elongated as normal and finish as palmitic acid. Alternatively, the dehydration step can lead to *cis*-3-decanoyl ACP which is then elongated to 9-palmitoleoyl
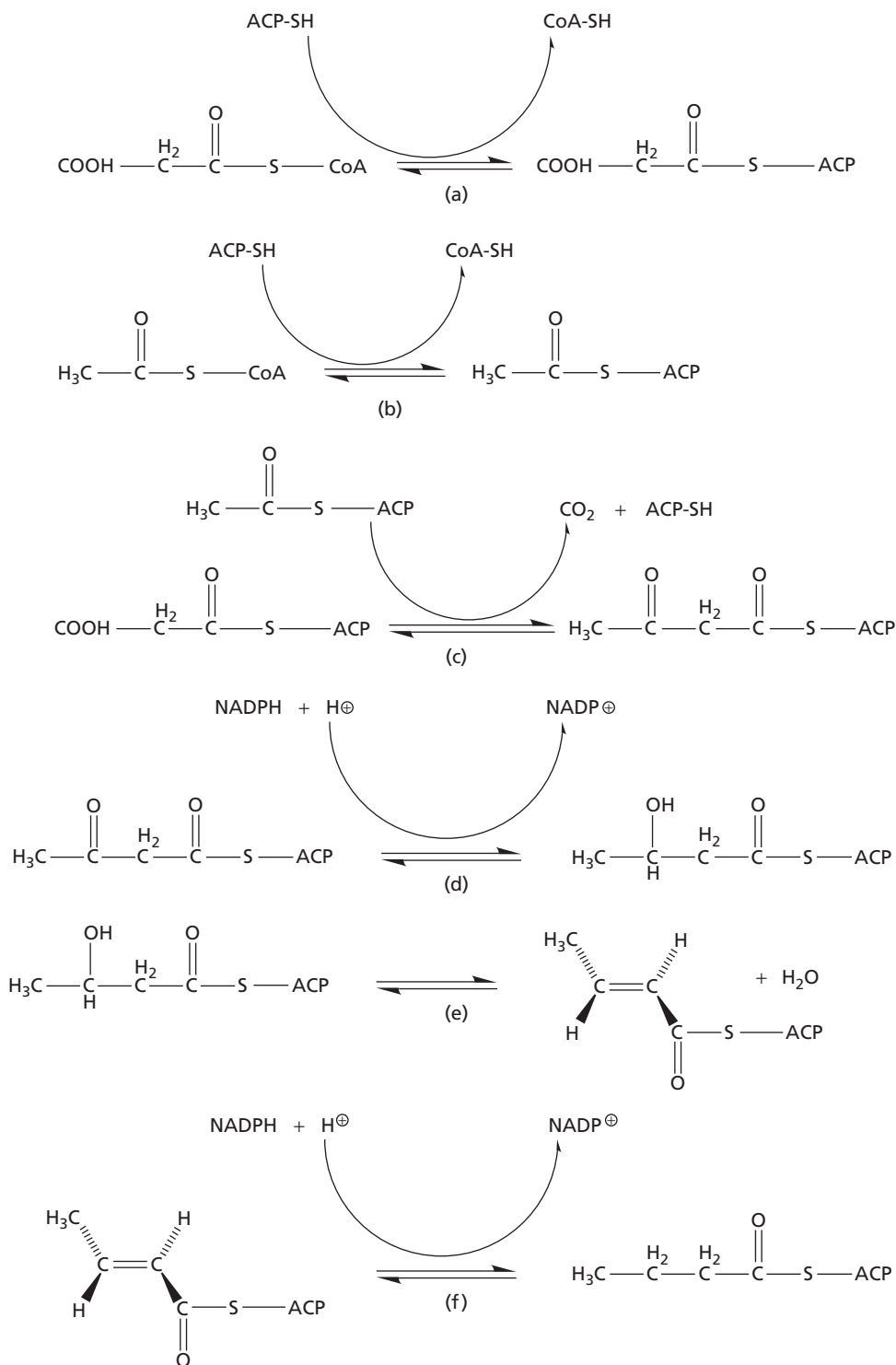
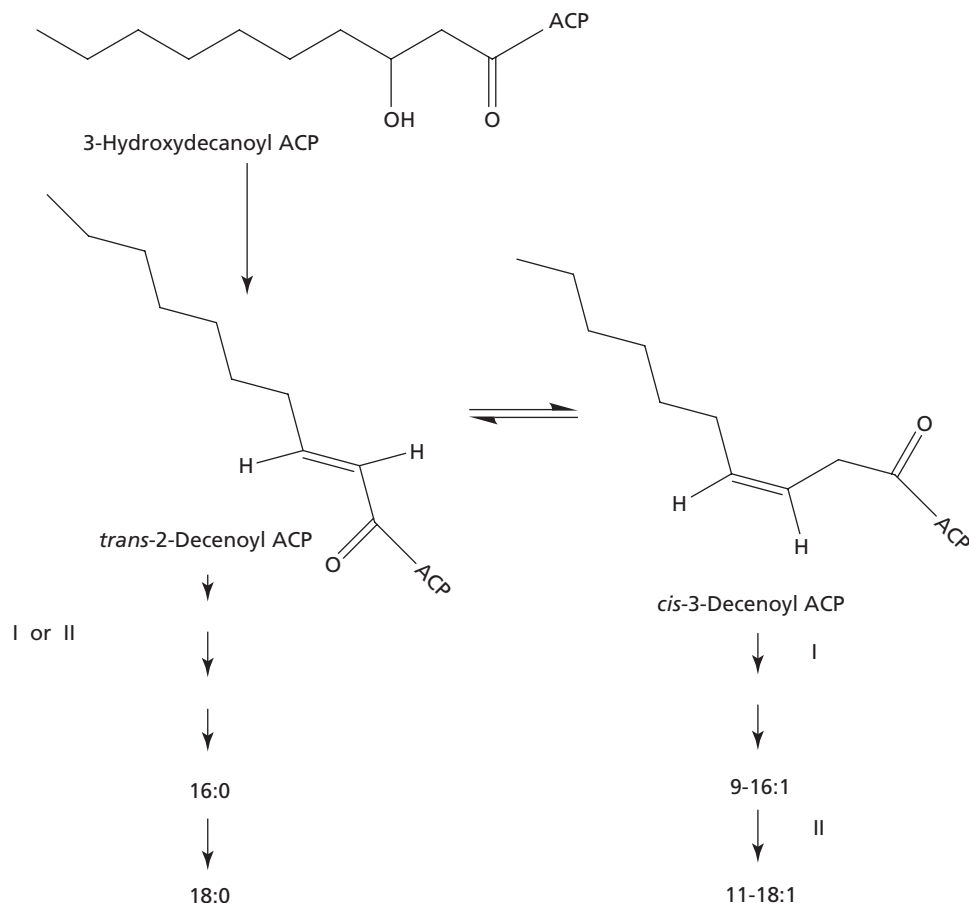**Fig. 1.6**   Partial biosynthetic reactions of FAS.

Fig. 1.7 Anaerobic pathway of fatty acid biosynthesis in bacteria showing the branching point in the formation of *cis* vaccenic acid, where I is 3-ketoacyl-ACP synthetase I, and II is 3-ketoacyl-ACP synthetase II.

ACP and then to 11-*cis*-vaccenyl ACP (the Lipid Library, www.lipidlibrary.co.uk). This results in the n-7 double bond being retained.

There are aerobic desaturases that remove two hydrogens from a saturated acyl chain stereospecifically. This is the system which is common in all organisms, where oxygen and a reducing cofactor are needed. The first double bond is usually produced at the 9 carbon atom catalysed by stearoyl–CoA Δ^9 desaturase in plants and algae with the production of oleic acid. Palmitoleic acid is derived in a similar way from palmitic acid.

Bacteria, uniquely, are able to produce Δ^{10} monoenoic acids. In addition some bacteria can remove a second pair of hydrogen atoms, giving rise to a diunsaturated double bond system, which is not a methylene interrupted one as is found in plants.

1.4.3   **Polyunsaturated fatty acids**

Mammals can introduce a second double bond to a monounsaturated fatty acid chain, but usually this new double bond cannot be inserted towards the methyl end of the chain.
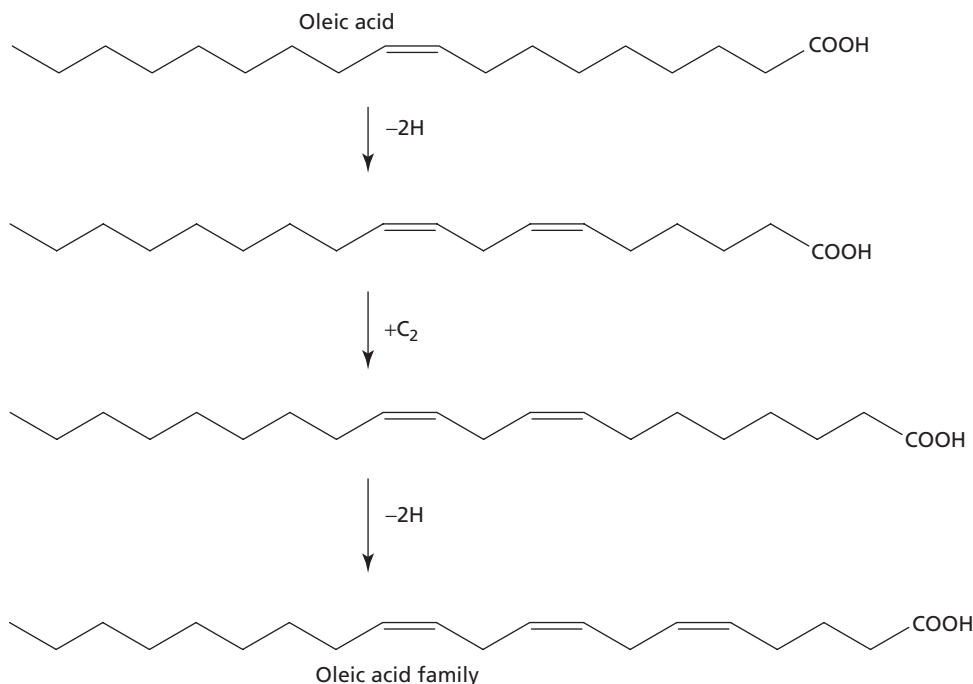
Oleic acid

—— COOH

↓ −2H

—— COOH

↓ +C$_2$

——COOH

↓ −2H

——COOH

Oleic acid family

Fig. 1.8   Biosynthetic relationships in the oleic acid family of fatty acids.

A second double bond can be introduced into oleic acid 9c-18:1, with the formation of 6c,9c-18:2 (Fig. 1.8). Addition of a further two carbon atoms under the influence of an elongase converts the C$_{18}$ chain length to 8c,11c-20:2. If this acid is desaturated, it produces 5c,8c,11c-20:3. Further elongation and desaturation steps will convert this acid to other acids in the oleic acid family which all have the first double bond from the methyl end at the 9-position, i.e. n-9 or ω9.

By contrast, when linoleic acid is subjected to a similar set of desaturation and elongation, the acids that are produced are shown in Fig. 1.9. Linoleic acid is converted to γ-linolenic acid, i.e. 6c,9c,12c-18:3, which is elongated to 8c,11c,14c-20:3. Further desaturation gives arachidonic acid, i.e. 5c,8c,11c,14c-20:4. Again, it is easy to see in Fig. 1.9 that all these acids in the linoleic acid family have the first double bond from the methyl end at the 6-position, i.e. n-6 or ω6.

Starting from α-linolenic acid, desaturation yields 6c,9c,12c,15c-18:4. Elongation of this acid gives 8c,11c,14c,17c-20:4, and a further desaturation leads to 5c,8c,11c,14c,17c-20:5. This is called eicosapentaenoic acid (67A) which gives the next member of the series, docosapentaenoic acid. This acid in turn loses 2 hydrogen atoms to give docosahexaenoic acid (DHA). Both EPA and DHA are found in fish oils. All of these acids in the linolenic acid family (Fig. 1.10) have the first double bond from the methyl end at the 3-position, i.e. n-3 or ω3. These pathways represent the formation of three different families of polyunsaturated fatty acids.

1.5   PROPERTIES OF *TRANS* FATTY ACIDS

The physical properties of *trans* fatty acids are different from the corresponding *cis* isomers.

Linoleic acid

COOH

−2H

CO$_2$H

+CH$_2$

CO$_2$H

−2H

CO$_2$H

Arachidonic acid
Linoleic acid family n-6
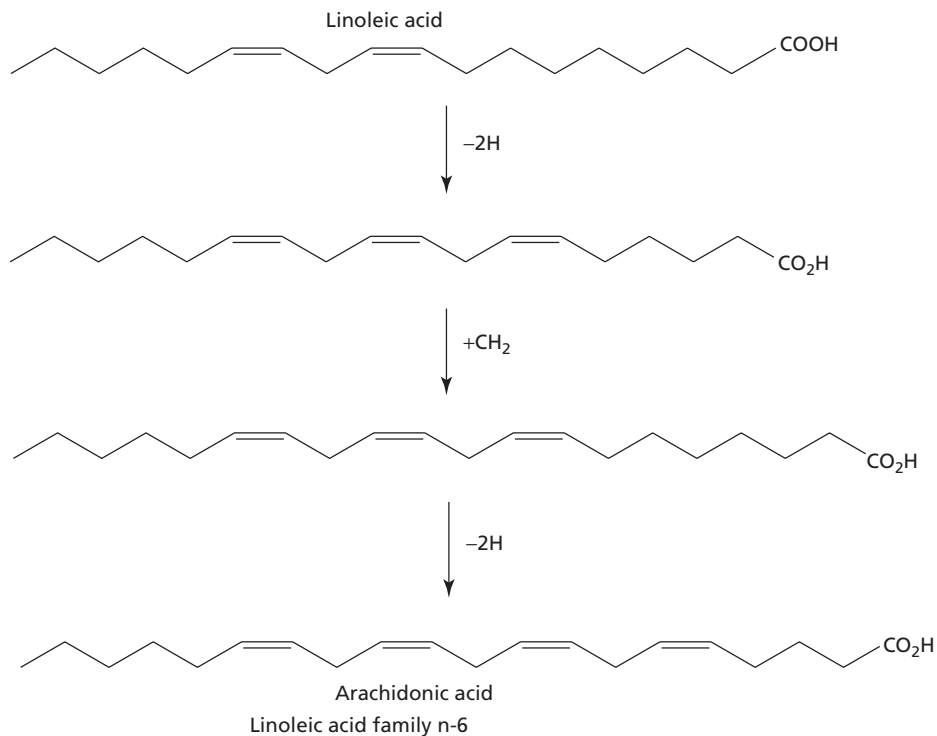
Fig. 1.9 Biosynthetic relationships in the linoleic acid (n-6) family of fatty acids.

Linolenic acid

CO$_2$H

−2H

CO$_2$H

+C$_2$

CO$_2$H

−2H

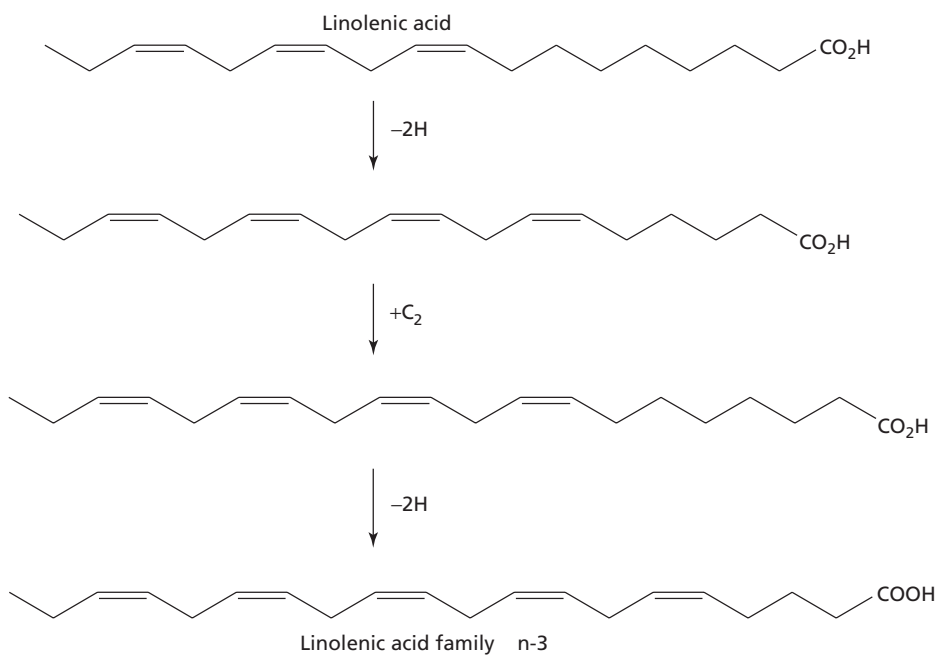COOH

Linolenic acid family   n-3

Fig. 1.10 Biosynthetic relationships in the linolenic acid (n-3) family of fatty acids.

Table 1.4 Melting points of monoacid triacylglycerols
with C_{18} monounsaturated acids.

Position of the double bond	*Cis* isomers	*Trans* isomers
4	34	53
5	14	41
6	28	52
7	7	39
8	24	49
9	5	41
10	27	49
11	10	43
12	32	51
13	26	44
14	44	58
15	43	56

1.5.1 Melting points

In Table 1.4 and Fig. 1.11 we can see that the melting points of the two series are very different with the *trans* isomers having the higher melting points (Hagemann *et al.*, 1975). It was this higher melting behaviour which made the *trans* fatty acids so valuable in commerce. These melting characteristics made it possible to produce the desirable properties for a plastic shortening by hydrogenation of cottonseed oil (and subsequently of other oils, e.g. soya bean oil).

The polymorphism of *trans* fatty acid triacylglycerols is indicated in Table 1.5 (Hagemann *et al.*, 1975).
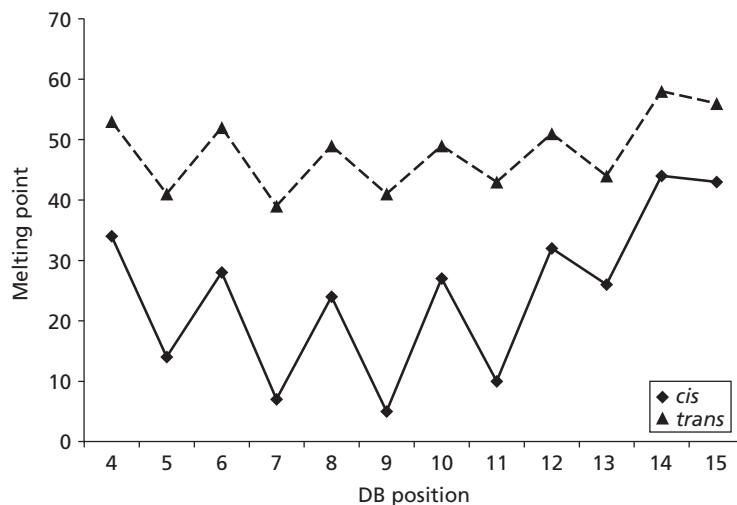

Fig. 1.11 Melting points of C_{18} triacylglycerols with differing positions of the double bond. DB, double bond; MP, melting point. (Adapted from Hagemann *et al.*, 1975.)

Table 1.5 Melting points (°C) of polymorphs of monoacid triacylglycerols with C_{18} monounsaturated acids.

Position of double bond	Polymorph α	Polymorph β'
cis $\triangle$4	5	34
trans $\triangle$4	27	53
cis $\triangle$7	−36	5
trans $\triangle$7	16	39
cis $\triangle$9	−34	5
trans $\triangle$9	15	41
cis $\triangle$10	−20	27
trans $\triangle$10		49
cis $\triangle$12		32
trans $\triangle$12	21	51
cis $\triangle$13		26
trans $\triangle$13	23	44

Adapted from Hagemann *et al.*, 1975.

These differences in melting point are attributable to the different shapes of the *trans* fatty acids in comparison with the *cis* isomers. In Fig. 1.12a, we can see the straight chain of the *trans* isomer elaidic acid that hardly alters the overall shape compared with a saturated acid, stearic acid, as in Fig. 1.12b. By contrast the *cis* double bond in Fig. 1.12c inserts a bend in the chain with the result that the molecules do not pack together as well. The melting points of a selection of *trans* compounds are given in Tables 1.6 and 1.7 (Hagemann *et al.*, 1972; Jackson and Callen, 1951; Markley, 1947).

1.5.2 Ultraviolet spectra

Ultraviolet (UV) spectra are not used very often for the determination of the major fatty acids, because the UV λ-maximum for the *cis* unsaturated group is at 176 nm and for the *trans* double bond at 187 nm. The UV spectrum is much more informative when conjugated double bonds are present in the fatty acid (Hamilton and Cast, 1999). In cyclohexane, α-eleostearic acid, $9c,11t,13t$-octadecatrienoic acid, has λ_{max} 262, 272 and 283 nm, as shown in Fig. 1.13, whilst β-eleostearic acid, $9t,11t,13t$-octadecatrienoic acid, has very similar absorption maxima at λ_{max} 259, 270 and 281 nm (O'Connor *et al.*, 1947).

**Table 1.6** Melting points (°C) of the β polymorph of selected triacylglycerols.

Trielaidin	41
Triolein	5
Tripetroselenin	28
Tripetroselaidin	52
Trierucin	32
Tri-*trans*-13-docosenoin	58

Adapted from Hagemann *et al.*, 1972.

Table 1.7 Melting points of selected *cis* and *trans* lipids.

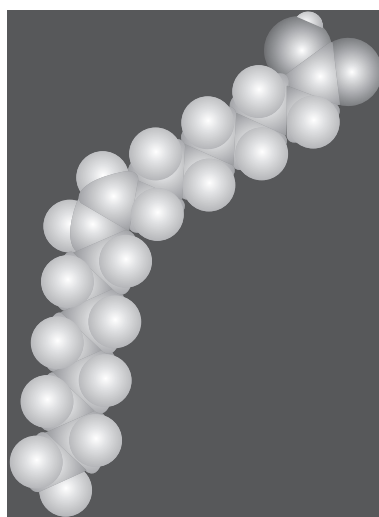| Lipids	Melting point (°C)
Elaidic acid	45.0–45.5
Oleic acid	16.3
Petroselaidic acid	53.2
Petroselenic acid	33
Vaccenic acid	43.5–44.1
Methyl elaidate	11.5
Methyl oleate	−5
Methyl petroselaidate	19.9
Triolein	5.1
Trielaidin	42.2

Adapted from Jackson and Callen, 1951; Markley, 1947.
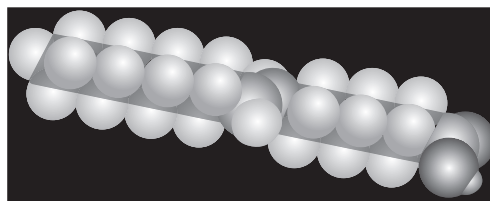
(a)
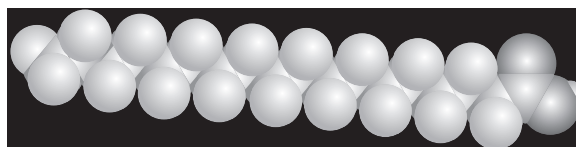
(b)

(c)

Fig. 1.12 Three-dimensional representations of (a) elaidic acid, (b) stearic acid and (c) oleic acid.
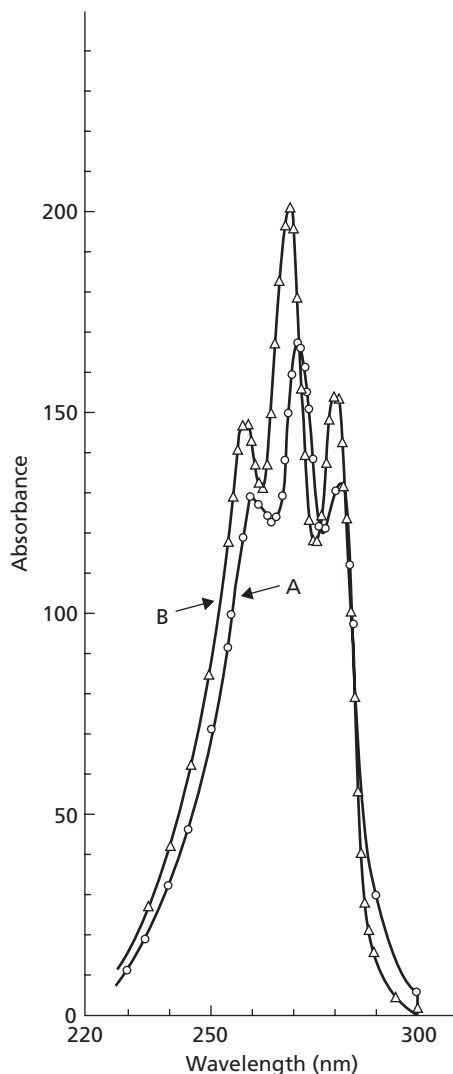
Fig. 1.13   UV absorption spectra of (A) α-eleostearic acid and (B) β-eleostearic acid. (Adapted from O'Connor *et al.*, 1947.)

1.5.3   Infrared spectra

The maxima for the *cis* double bonds are at 1660–1630 and 730–650/cm, and for the *trans* double bonds at 1680–1670 and 980–865/cm. When the double bonds are in conjugation, we get *cis*,*trans* conjugated bonds at 990–980 and 968–950/cm, whilst *trans*,*trans* double bonds are at 990–984/cm. For triunsaturated acids, we have a maximum at 989/cm for *cis*,*cis*,*trans* conjugated, at 991/cm for *cis*,*trans*,*trans* conjugated and at 994/cm for *trans*,*trans*,*trans* conjugated (Chapman, 1965).

A typical spectrum for trielaidin is shown in Fig. 1.14, with the characteristic band at 980/cm, in contrast with the spectrum for triolein (Feuge *et al.*, 1951).
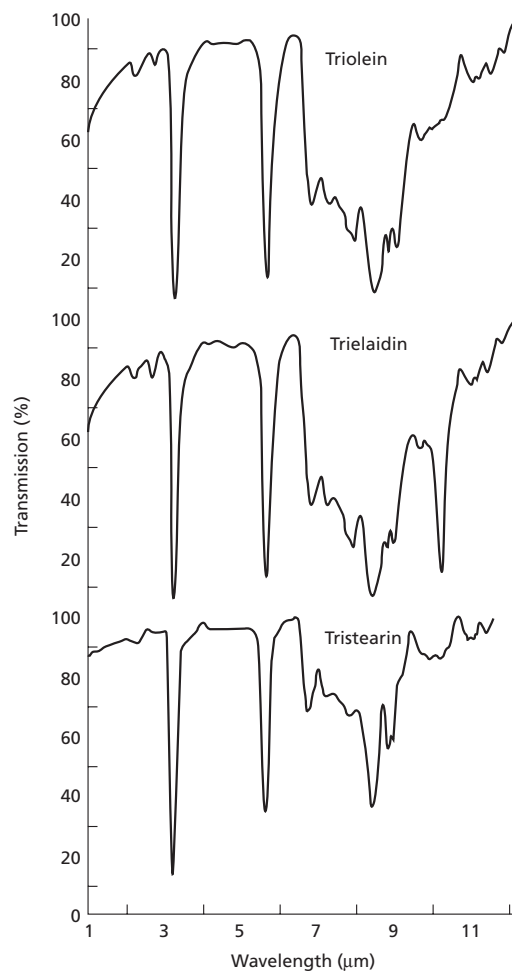
Fig. 1.14 Infrared absorption spectra of triolein, trielaidin and tristearin in chloroform. (Adapted from Feuge *et al.*, 1951.)

Table 1.8 Chemical shifts for the ethylenic carbon atoms in octadecenoic acid isomers by nuclear magnetic resonance.

Position of double bond	Trans			Cis		
5	131.98		128.72	131.44		128.21
6	131.13		129.49	130.62		129.01
7	130.81		129.89	130.31		129.45
8	130.67		130.13	130.17		129.65
9	130.54		130.23	130.09		129.78
10	130.47		130.21	130		129.83
11	130.43		130.34	129.96		129.89
12		130.14			129.94	
13		130.39			129.9	
14	130.63		130.13	130.16		129.66
15	131.93		129.45	131.54		129.39

Adapted from Gunstone, 1993.

Fourier transform infrared methods have been developed for the measurement of the *trans* content of oils, as noted by Ismail *et al*. in 1999.

1.5.4   Nuclear magnetic resonance spectroscopy

Gunstone (1993) has shown that the ^{13}C spectra of *cis* and *trans* fats can be used as a way of analysing partially hydrogenated fats. The values given in Table 1.8 show that there are differences between the *cis* and *trans* isomers in cases where the double bond is at C_5 through C_{15} of the octadecenoic acids.

Nutrition facts		
Serving size 1 cup (228 g)		
Serving per container 2		
Amount per serving		
Calories 260 Calories from fat 120		
		% Daily value*
Total fat	13 g	20%
Saturated fat	5 g	25%
Trans fat		
Cholesterol	30 mg	10%
Sodium	660 mg	28%
Total carbohydrate	31 g	10%
Dietary fiber	0 g	0%
Sugars	5 g	
Protein	5 g	
Vitamin A 4% Vitamin C 2%		
Calcium 15% Iron 4%		
*Percent daily values are based on a 2000 caloric diet. Your daily values may be higher or lower depending on your calorie needs:		

	Calories	2000	3000
Total fat	less than	65 g	80g
Sat fat	less than	20 g	25 g
Cholesterol	less than	200 mg	200 mg
Sodium	less than	2400 mg	2400 mg
Total carbohydrate		300 g	375 g
Dietary fiber		26 mg	30 mg

Calories per gram
Fat 9   Carbohydrate 4 Protein 4

Fig. 1.15   A United States of America nutrition facts label. (Adapted from Moss, 2006.)

1.6   LABELLING AND LEGISLATION

The Danish government has issued an order (Order no. 160), which came into operation on 31 March 2003. The order applies to oils and fats, including emulsions with fat as the continuous phase, which, either alone or as part of processed foodstuffs, are intended, or are likely, to be consumed by humans. The order does not apply to the naturally occurring content of *trans* fatty acids in animal fats or products governed under any other legislation. The order only applies to products sold to the final consumer. It states that it is prohibited to sell oils and fats covered by the order to consumers if they contain a higher level of *trans* fatty acids defined in the Annex than that quoted in Section 3. Section 3 states that, as from 1 June 2003, the content of *trans* fatty acids in the oils and fats covered by this order must not exceed 2 g/100 g of oil or fat. In products that are claimed to be 'free from fatty acids', the content of *trans* fatty acids in the finished product shall be less than 1 g/100 g of the individual oil or fat. Such has been the success of the Danish manufacturers/authorities that the level of intake of *trans* fatty acids from margarines and shortenings has fallen away completely from 4.5 g *trans* fatty acids per day in 1975, 2.2 g *trans* fatty acids per day in 1993 through 1.5 g *trans* fatty acids per day in 1995 to almost zero by 2005 (Leth *et al.*, 2006).

From 1 January 2006, the US government amended its regulations on nutrition labelling. This regulation is available on http://vm.cfsan.fda.gov/-Ird/fr991117.html and requires that all foodstuffs or products containing *trans* fatty acids, e.g. dietary supplements, should have the amount of *trans* fatty acids stated on the label. A typical example is shown in Fig. 1.15 (Moss, 2006), which is a listing of the grams of *trans* fat in a serving defined as the sum of all the unsaturated fatty acids that contain one or more isolated (non-conjugated) double bonds in the *trans* configuration. If the serving contains less than 0.5 g, it is possible to state the content as zero *trans* (Yurawecz, 2004). In October 2006, the Food navigator.com http://www.foodnavigator.com reported that the Australian government plans to work with industry to reduce *trans* fatty acids in Australian food.

REFERENCES

Chapman, D. (1965) Infrared spectroscopy of lipids. *J Am Oil Chem Soc* **42** (5), 353–371.

Comes, F., Farines, M., Aumelas, A. & Soulie, J. (1992) Fatty acids and triacylglycerols of cherry seed oil. *J Am Oil Chem Soc* **69**, 1224–1227.

Dijkstra, A.J. (2002) Hydrogenation and fractionation. In: *Fats in Food Technology* (ed. K.K. Rajah). Sheffield Food Technology, Sheffield, pp. 123–158.

Feuge, R.O., Pepper, M.B., O'Connor, R.T. & Field, E.T. (1951) Modification of vegetable oils. XI The formation of trans isomers during hydrogenation of methyl oleate and triolein. *J Am Oil Chem Soc* **28**, 420–426.

Gunstone, F.D. (1967) *An Introduction to the Chemistry and Biochemistry of Fatty Acids and Their Glycerides*. Chapman and Hall, London.

Gunstone, F.D. (1993) Composition of hydrogenated fats by high resolution ^{13}C nuclear magnetic resonance spectroscopy. *J Am Oil Chem Soc* **70** (10), 965–970.

Gunstone, F.D. (1996) *Fatty Acid and Lipid Chemistry*. Blackie Academic and Professional, Glasgow.

Gunstone, F.D. (2004) *The Chemistry of Oils and Fats, Sources, Composition, Properties and Uses*. Blackwell Publishing, Oxford.

Gunstone, F.D., Harwood, J.L. & Padley, F.B. (1994) *The Lipid Handbook*. Chapman and Hall, London.

Gurr, M.I. & James, A.T. (1980) *Lipid Biochemistry, an introduction*. Chapman and Hall, London.

Hagemann, J.H., Tallent, W.H., Barve, J.A., Ismail, I.A. & Gunstone, F.D. (1975) Polymorphism in single-acid triglycerides of positional and geometric isomers of octadecenoic acid. *J Am Oil Chem Soc* **52**, 204–207.

Hagemann, J.W., Tallent, W.H. & Kolb, K.E. (1972) Differential scanning calorimetry of single acid triglyc-erides: effect of chain length and unsaturation. *J Am Oil Chem Soc* **49**, 118–123.

Hamilton, R.J. & Cast, J. (1999) *Spectral Properties of Lipids*. Sheffield Academic Press, Sheffield.

Hastert, R.C. (1996) Hydrogenation. In: *Bailey's Industrial Oil and Fat Products*, Vol. 4 (ed. Y.H. Hui). J. Wiley and Sons, New York, pp. 213–300.

Hay, J.D. & Morrison, W.R. (1970) Isomeric monoenoic fatty acids in bovine milk fat. *Biochim Biophys Acta* **202**, 237–243.

Hoffmann, G. (1989) *Chemistry and Technology of Edible Oils and Fats and Their High Fat Products*. Academic Press, London.

Ismail, A.A., Nicodemo, A., Sedman, J., van de Voort, F.R. & Holzbaur, I.E. (1999) Infrared spectroscopy of lipids: principles and applications. In: *Spectral Properties of Lipids* (eds R.J. Hamilton & J. Cast). Sheffield Academic Press, Sheffield, pp. 235–269.

Jackson, F.E. & Callan, J.E. (1951) Evaluation of the Twitchell isooleic method: comparison with the infrared trans isooleic method. *J Am Oil Chem Soc* **28**, 61–65.

Korver, O. & Katan, M.B. (2006) The elimination of *trans* fats from spreads: how science helped to turn an industry around. *Nutr Rev* **64** (6), 275–279.

Leth, T., Jensen, H.G., Mikkelsen, A.A. & Bysted, A. (2006) The effect of the regulation on *trans* fatty acid content in Danish food. *Atheroscler Suppl* **7**, 53–56.

Markley, K.S. (1947) *Fatty Acids: Their Chemistry and Physical Properties*. Interscience Publishers, New York.

Moss, J. (2006) Labeling of trans fatty acid content in food, regulations and limits – the FDA view. *Atheroscler Suppl* **7**, 57–59.

O'Connor, R.T., Heinzelman, D.C., McKinney, R.S. & Pack, F.C. (1947) The spectrophotometric determination of alpha and beta isomers of eleostearic acid in tung oil. *J Am Oil Chem Soc* **24**, 212–216.

Placek, L.L. (1963) A review of petroselenic acid and its derivatives. *J Am Oil Chem Soc* **40**, 319–329.

Rossell, J.B. (2001) *Oils and Fats, Volume 2 Animal Carcass Fats*. Leatherhead Publishing, Leatherhead.

Sommerfeld, M. (1983) Trans unsaturated fatty acids in natural products and processed foods. *Prog Lipid Res* **22** (3), 221–233.

Yurawecz, M.P. (2004) FDA requires mandatory labeling of *trans* fat. *Inform* **15** (3), 184–185.