

University of Jaffna

**Professor Sivapathasuntharam Mageswaran
Memorial Lecture - 2017**

**Heat induced Isomerization and
auto oxidation reactions of
C18 unsaturated fatty acids
in edible oils**

by

Professor Alfred A. Christy
Department of Science Faculty of Engineering
and Science, University of Agder, Serviceboks
422, 4604 Kristiansand, Norway.

on

**Friday 17th February 2017
at 3.00 p.m**

at

**Kailasapathy Auditorium
University of Jaffna**



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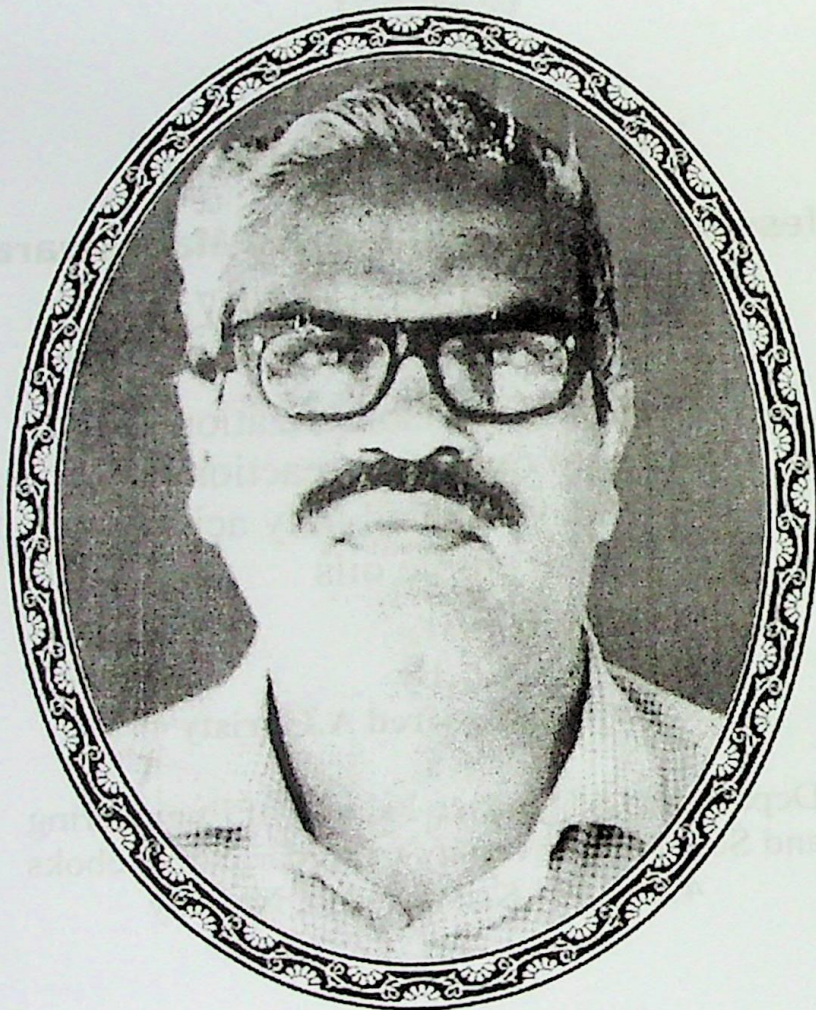
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Professor Sivapathasuntharam Mageswaran

MESSAGE FROM THE VICE-CHANCELLOR



It is my great pleasure and honour to welcome all of you for Late Professor Sivapathasuntharam Mageswaran Memorial Lecture – 2017 this afternoon. We were indeed fortunate to had Late Professor Sivapathasuntharam Mageswaran, one of the most important pioneers of the University of Jaffna.

He has taken immense interest and devoted his life for the improvement and up grading the standard of the faculty of Science especially of the Department of Chemistry.

He was virtually single handed took up the challenges that confronted him with fortitude and courage. He always takes appropriate action to maintain the student discipline and had been the role model to many Academic Staff.

I am happy that Professor Alfred A. Christy is delivering the Memorial Lecture. He will speak to us on the topic. "Heat induced Isomerization and auto oxidation reactions of C18 unsaturated fatty acids in edible oils".

I thank Professor Alfred A. Christy for accepting our invitation and delivering this lecture.

Prof. (Ms.) V. Arasaratnam
Vice-Chancellor
University of Jaffna, Jaffna
17.02.2017

Edible oils and Fatty acids

Frying and deep frying in the cooking process of food is universe and has become one of the important methods in preparing food in all cultures. The process is carried out usually in edible oils such as olive, coconut, soya, sunflower, peanut oil, palm oil etc. An overview of the world consumption of different edible oils is shown in Table 1.

Table 1. World consumption of edible oils[1]

Edible oil	World consumption in million metric tons
Palam	41.31
Soybean	41.28
Rapeseed	18.24
Sunflower	9.91
Peanut	4.82
Cotton seed	4.99
Palam kernel	4.85
Coconut	3.48
Olive	2.84

Edible oils are triglycerides (Triacylglycerols), esters of glycerol and fatty acids (Fig. 1). The fatty acids in edible oils can contain 14-20 carbon atoms (Table 2). Edible oils contain saturated and

unsaturated fatty acids in varying quantities. Among unsaturated fatty acids, fatty acids containing 18 carbon atoms can reach up to 95% or more in certain edible oils (Table 2). The unsaturated fatty acids include mono-unsaturated oleic acid (18:1 9c), polyunsaturated linoleic (18:2 9c12c) and linolenic acids (18:3 9c12c15c). All these unsaturated fatty acids are found in *cis*-configurations in nature. During frying, these fatty acids undergo isomerization, oxidation, polymerization and decomposition and the degree of transformation through these processes depend on the stability of the unsaturated fatty acids in the triglyceride molecules [2-6]. The oxidation rates of linoleic acid and linolenic acid are 10 and 100 times faster than oleic acid respectively. The *trans* isomers produced during frying process are unhealthy and has been linked to the risk of coronary heart disease [7-12]. The chemical compounds produced during frying are toxic and unhealthy [10].

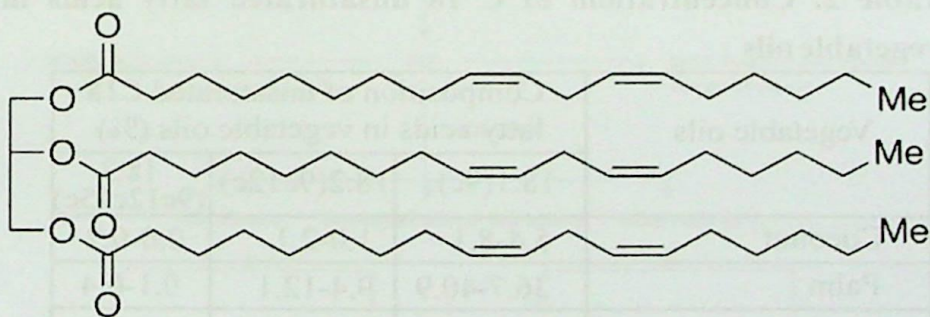


Fig 1. A triglyceride molecule containing three linoleic acid molecules

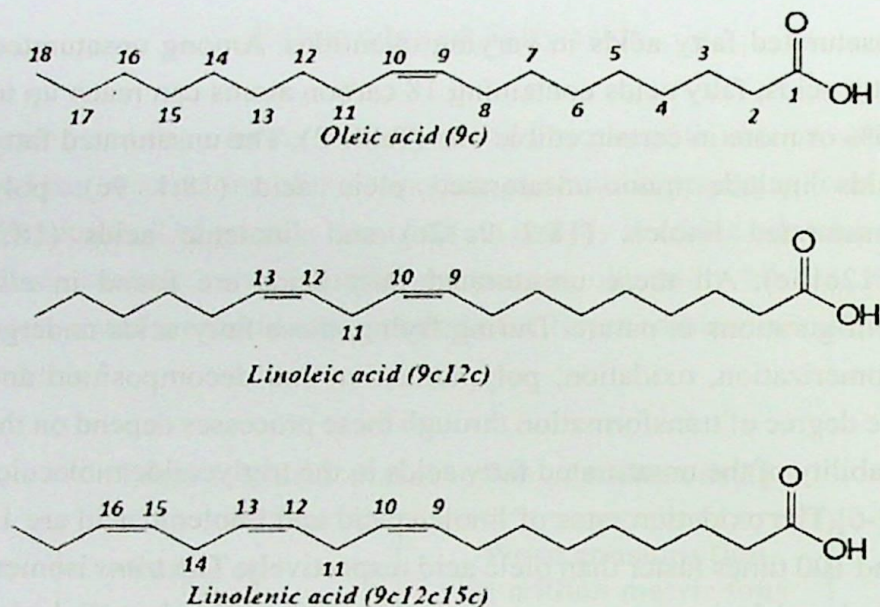


Fig. 2. Unsaturated fatty acids containing 18 carbon atoms

Table 2. Concentration of C 18 unsaturated fatty acids in vegetable oils

Vegetable oils	Composition of unsaturated C18 fatty acids in vegetable oils (%)		
	18:1(9c)	18:2(9c12c)	18:3 (9c12c15c)
Coconut	5.4-8.1	1.0-2.1	0.0-0.2
Palm	36.7-40.9	9.4-12.1	0.1-0.4
Corn	24.6-42.2	39.4-60.4	0.7-1.4
Peanut	36.4-67.1	14.0-43.0	0.0-0.1
Olive	55-83	3.5-21.0	0.0-1.5
Soybean	17.7-25.1	49.8-57.1	5.5-9.5
Safflower seed	8.4-21.3	67.8-83.2	0.0-0.1
Sunflower	13-40	40-74	0.0-0.3

This article looks at the chemical (auto oxidation) and isomerization reactions taking place in C18 unsaturated fatty acids in edible oils during heating.

General auto-oxidation mechanism

Fatty acid oxidation goes through three steps namely Initiation, propagation and termination

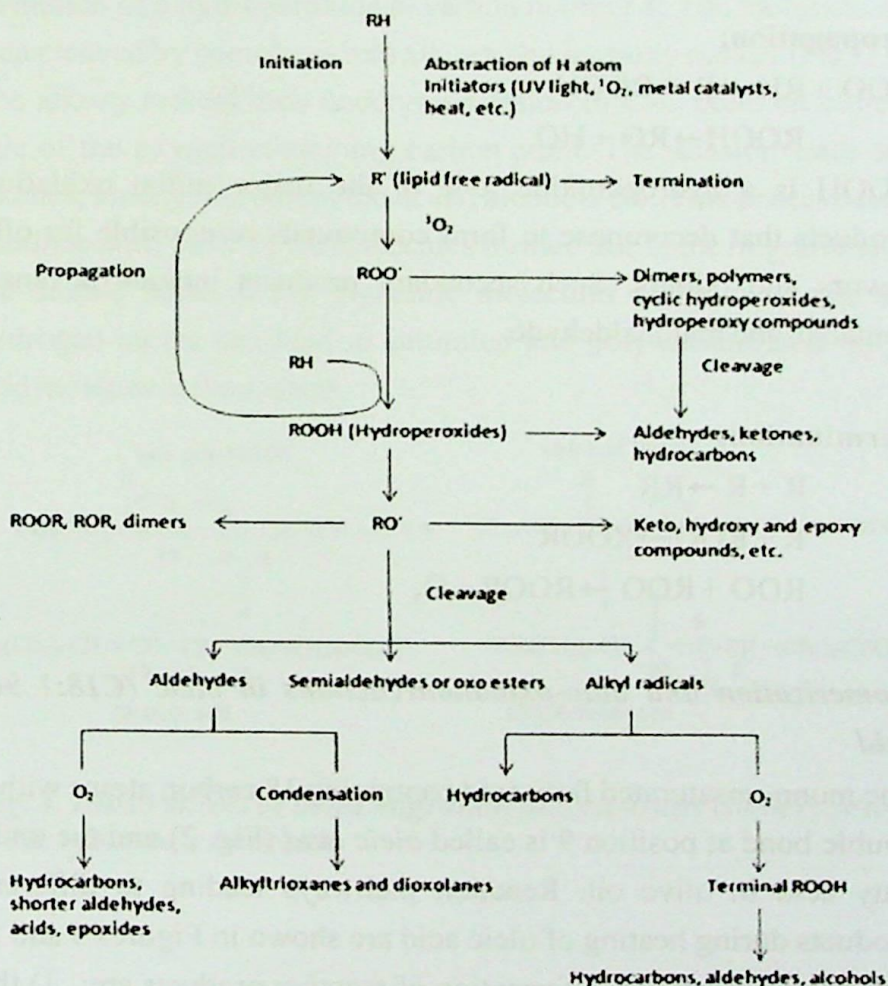
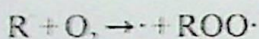
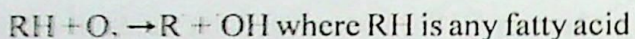


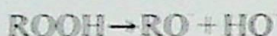
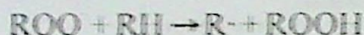
Fig. 3. Generalized scheme for auto-oxidation of lipids [13]

Initiation:



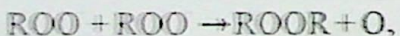
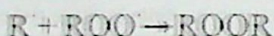
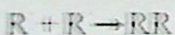
R· is a free radical formed by removing a labile hydrogen from a carbon atom adjacent to a double bond

Propagation:



ROOH is a hydroperoxide, one of the major initial oxidation products that decompose to form compounds responsible for off-flavors and odours. Such secondary products include hexanal, pentanal, and malonaldehyde.

Termination:



Isomerization and auto-oxidation reactions in oleic (C18:1 9c) acid

The mono-unsaturated fatty acid containing 18 carbon atoms with a double bond at position 9 is called *oleic acid* (Fig. 2) and the main fatty acid in olive oil. Reaction pathways leading to different products during heating of oleic acid are shown in Figures 4 and 5. The possibilities for the formation of reaction products are: 1) the *cis* double bond can simply open up and rotate freely along the 9,10

C-C bond and intraconvert to a *trans* double bond, 2) with the opening of the double bond and hydrogen transfer, the double bond can migrate in both directions from the 9-10 position and form several *cis* and *trans* isomers of the 18:1 fatty acids, or 3) the molecule can undergo oxidative fission at different positions of the carbon chain backbone. The first step in the fission reaction is the formation of a hydroperoxide at carbon number 8. The molecule is then cleaved by homolysis into alkoxy and hydroxy radical (Fig. 2). The alkoxy radical then undergoes β -scission of C-C bond on either side of the oxygen containing carbon atom. The scission leads to alkenes, aldehydes, aldehydic acids, alcohols etc. This process can continue with some of the molecules formed due to the migration of the double bond in the glyceride molecules or 4) exchange of hydrogen atoms can lead to saturated and poly-unsaturated fatty acid moieties in the system.

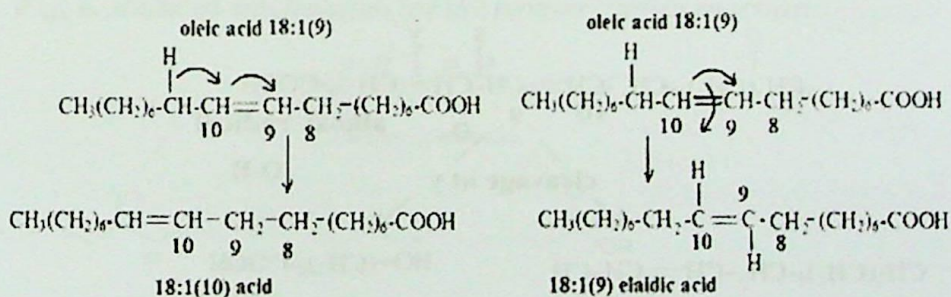
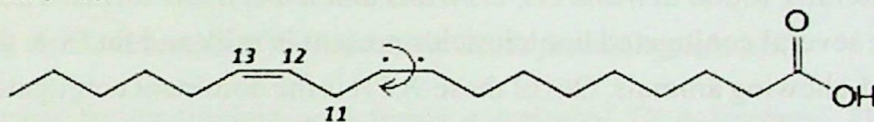


Fig. 4. Illustrations of bond migration and cis-trans isomerization in 18:1 fatty acid molecule

Isomerization and auto oxidation reactions in linoleic (C18:2 9c12c) fatty acid

Linoleic acid has two double bonds and Linoleic acid in triglycerides isomerizes into 9c12t, 9t12c and 9t12t fatty acids. The *cis-trans* isomerization can be explained by a radical mechanism shown in Fig. 6. The double bonds are relatively in the middle of the molecule and this gives equal probabilities in the formation of 9c12t and 9t12c isomers. However, heat induced isomerization slightly favours the formation of 9t12c. The chain length after the double bond 12-13 is smaller compared to the chain length after the double bond 9-10.



Linoleic acid (9c12c)

Fig. 6. Radical mechanism for the isomerization reaction

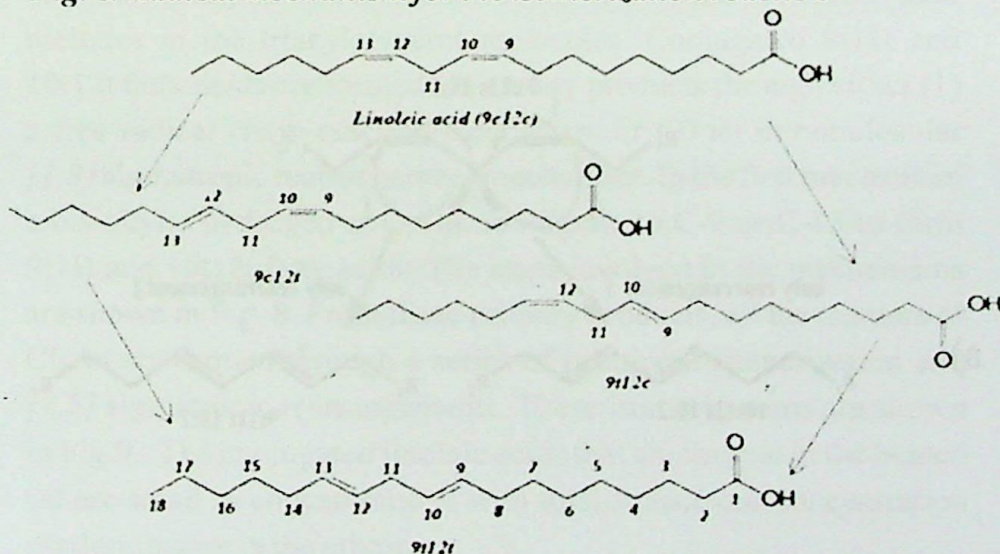
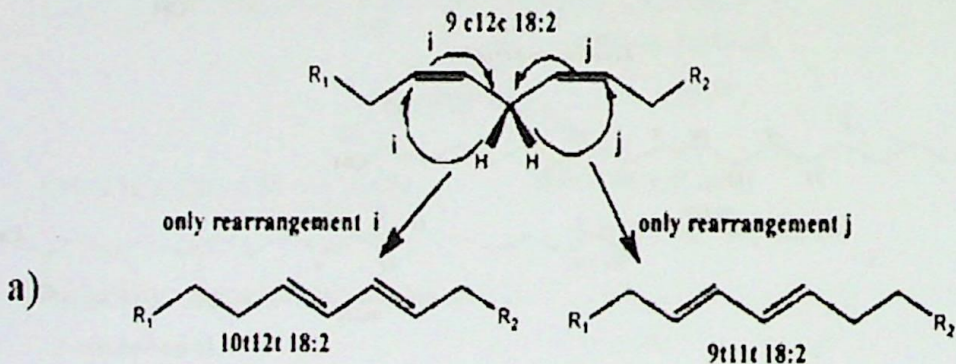


Fig. 7. Cis-trans Isomers of linoleic acid

Formation of Conjugated Linoleic Acids (CLAs)

Apart from the *cis-trans* isomers of linoleic acid, isomers of Conjugated Linoleic Acids (CLAs) and isomers due to the migration of double bonds in linoleic acids can also be formed during heating of vegetable oils [12,14].

Conjugated linoleic acids (CLAs) are natural components found in animal products such as meat and dairy products derived from ruminant animals. Conjugated linoleic acids have gained considerable attention in recent times because of their health benefits [15-31]. The conjugated isomers in milk and fats are generally found in *trans/cis*, *cis/trans* and *trans/trans* forms. There are several conjugated linoleic acids present in milk and fat from the cud-chewing animals. Out of these 9*c*11*t* is the dominant conjugated linoleic acid. The CLA isomers such as 9*c*11*t* and 10*t*12*c* can also be found in hydrogenated vegetable oils [32].



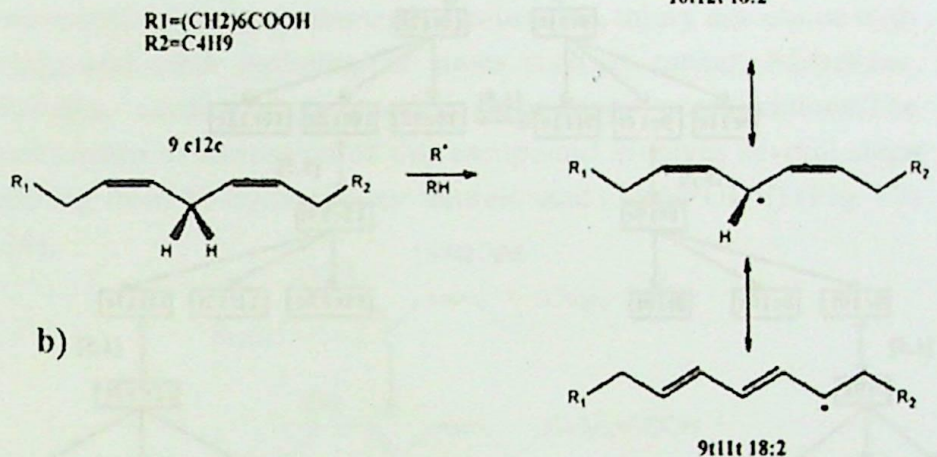


Fig. 8. Mechanism of formation of CLAs from Linoleic acid

During frying linoleic acid forms a series of CLA isomers. The mechanisms proposed by Destailats and Angers [33] can well be used here to explain the formation of CLAs from the 9c12c fatty acid moieties in the triacylglycerol molecules. Conjugated 9t11t and 10t12t fatty acids are formed as primary products through either (1) a free radical chain reaction mechanism or (2) an intramolecular [1,3] sigmatropic rearrangement mechanism. In the first mechanism a bis-allylic hydrogen at C-11 can migrate to C-9 or C-13 to form 9t11t and 10t12t fatty acids. The steps involved in the mechanisms are shown in Fig. 8. From these primary products, all the isomers of CLAs are formed through a series of positional isomerization and [1,5] sigmatropic rearrangements. These isomerizations are shown in Fig.9. The conjugated linoleic acids that are formed in the heated oil are small in concentrations with total *trans,trans* concentration predominating in the mixture.

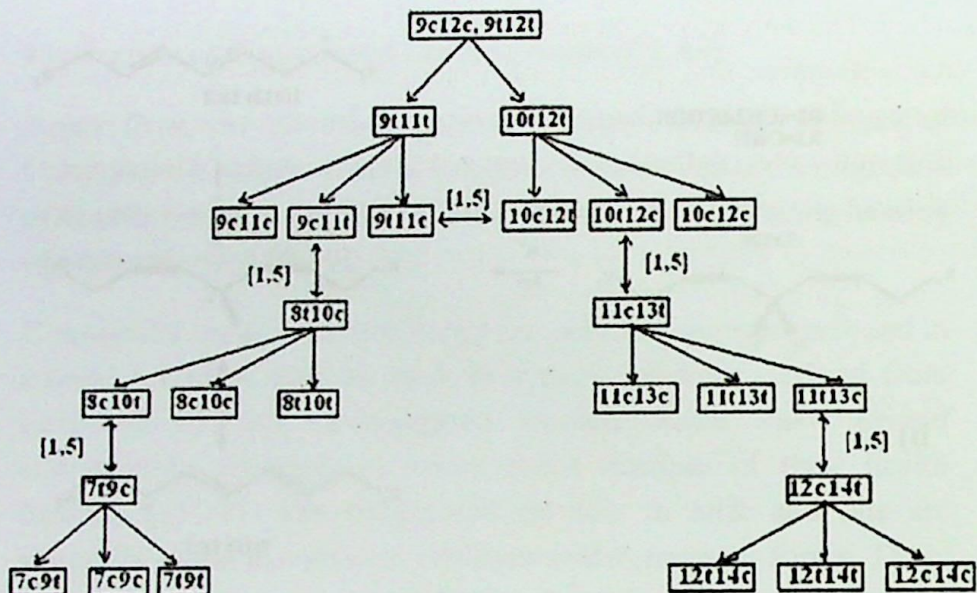


Fig. 9. An isomerization tree proposed by Christy [12] showing the formation of CLA isomers in the heated Linoleic acid

Auto-oxidation of linoleic acid

Linoleic acid has two double bonds and therefore has several oxidation points in the molecule. The oxidation of linoleic acid produces additional mixture of aldehydes besides several shorter fatty acids, keto, hydroxy or epoxy compounds. Oxidation linoleic acid and other ω -6 fatty acids yield well defined compounds 2,4-decadienal and 3-nonenal from 9-hydroxy linoleate, hexanal and pentanal from 13-hydroxy linoleate and 2-heptanal from 13-hydroxy linoleate. 2-hexanal, 2-octenal, 2,4-nonadienal, 4,5-dihydroxydecenal and 4-hydroxy-2,3-trans-nonenal (4-HNE) are frequently found in heated vegetable oils. Among the aldehydes that are formed during the lipid oxidation 4-HNE is most extensively

studied. The 4-hydroxy 2-nonenal (HNE) is a highly toxic and most abundant stable end product of lipid peroxidation. It has been implicated in the tissue damage, dysfunction, injury associated with aging and other pathological states such as cancer, Alzheimer, diabetes, cardiovascular and inflammatory complications. The mechanism of formation of this compound involves several steps starting from 13-hydroperoxy-linoleic acid (13-HPODE) (Fig. 10) [34].

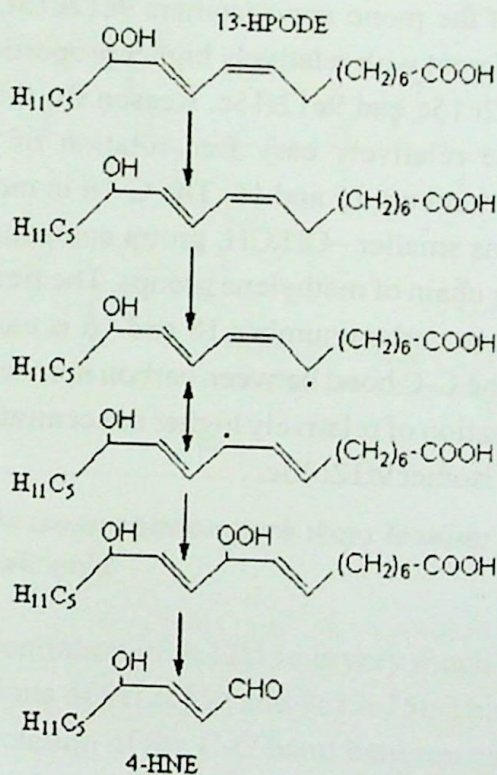


Fig. 10. Mechanism of formation of 4-HNE from 13-hydroperoxy-linoleic acid (13-HPODE) [34]

Isomerization and auto oxidation reactions in linolenic (C18:3 9c12c15c) acid

The linolenic acid molecule isomerizes into 7 different isomers during heating. The isomers are 9t12c15c, 9c12t15c, 9c12c15t, 9t12t15c, 9c12t15t, 9t12c15t and 9t12t15t. The concentrations of the isomers formed depend on their nature. The concentrations of the mono *trans* isomers 9c12c15t, 9t12c15c and 9c12t15c are different with relatively higher proportion of 9c12c15t compared to 9t12c15c and 9c12t15c. Reason for this difference in proportion is the relatively easy free rotation of the C-C bond between carbon numbers 15 and 16. The C-16 in methyl linolenate molecule contains smaller $-\text{CH}_2\text{CH}_3$ group compared to C-9 which contains a longer chain of methylene groups. The free rotation of the C-C bond between carbon number 15 and 16 is easy compared to free rotation of the C-C bond between carbon number 9 and 10. This leads to the formation of relatively higher concentration of 9c12c15t compared to the isomer 9t12c15c.

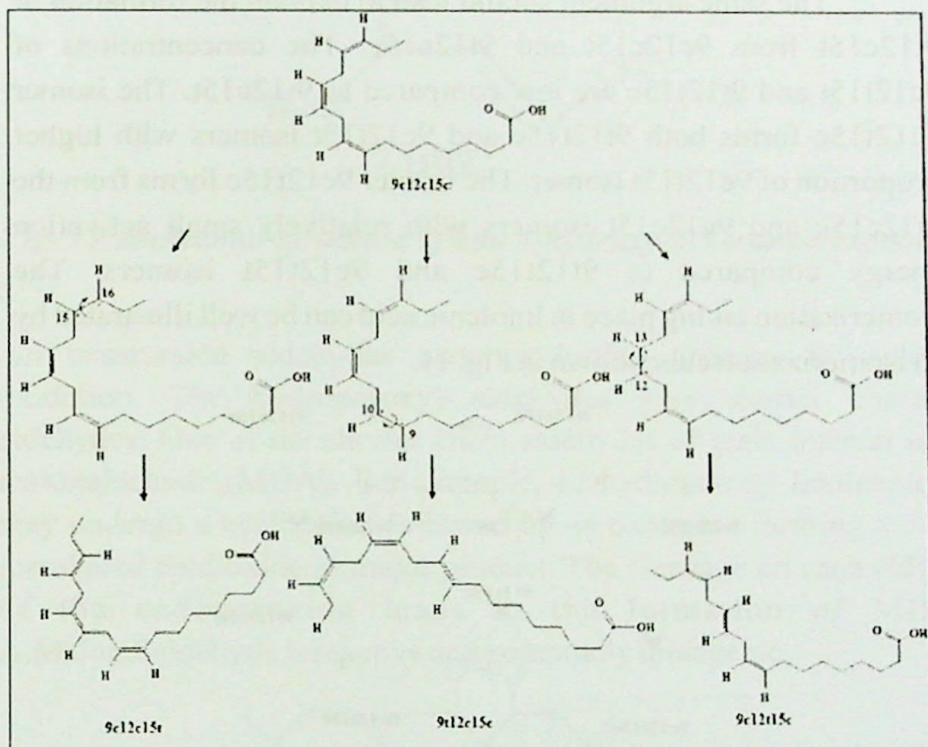


Fig. 11. Cis-trans isomerization path ways leading to mono trans isomers of linolenic acid

The concentration of 9c12t15c is very much low compared to the concentrations of 9t12c15c and 9c12c15t. This is because of the difficulty in rotation of the C-C bond between carbon numbers 12 and 13. Both carbons have larger groups and free rotation about the bond is difficult. The energy needed for the rotation of the bond between carbon numbers 12 and 13 is higher than the energy needed for rotation of the bonds between carbon numbers 9 and 10, and 15 and 16. Therefore, the proportion order is as follows $9c12c15t > 9t12c15c > 9c12t15c$.

The same argument should lead to explain the formation of 9t12c15t from 9c12c15t and 9t12c15c. The concentrations of 9c12t15t and 9t12t15c are low compared to 9t12c15t. The isomer 9c12t15c forms both 9t12t15c and 9c12t15t isomers with higher proportion of 9c12t15t isomer. The isomer 9c12t15c forms from the 9t12c15c and 9c12c15t isomers with relatively small activation energy compared to 9t12t15c and 9c12t15t isomers. The isomerization taking place in linolenic acid can be well illustrated by an isomerization cube shown in Fig. 11.

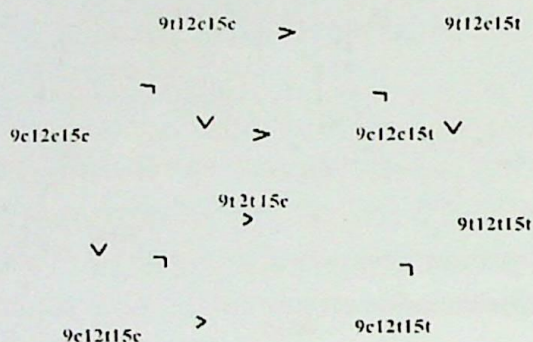


Fig. 12. An isomerization cube showing the isomerization pathways of linolenic acid

The oxidation of linolenic acid produces various compounds depending on the location of the hydroperoxyl group. Thus, 9-OOH linolenate gives 2,4,7-decatrinal and 3,6-nonadinal, 12-OOH linolenate gives 2,4-heptadienal and 3-hexenal, 13-OOH linolenate gives 3-hexenal and 2-pentenal and, finally, 16-OOH linolenate gives propanal [34].

Several other compounds are formed via less known mechanisms such as butanal, 4,5-epoxy hepta-2-cnal, 4-hydroperoxy hexenal, 4,5-dihydroxydeccenal and **4-hydroxy-2,3-trans-hexenal (4-IIIE)**.

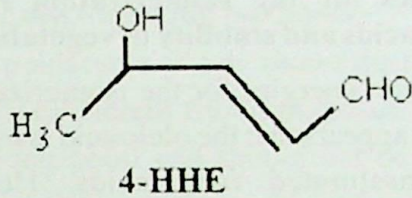


Fig- 13. Molecular structure of and 4-hydroxy-2,3-trans-hexenal [34]

All unsaturated aldehydes undergo further changes by auto oxidation. The hydroperoxyl aldehydes give shorter chain aldehydes. One of the shorter chain aldehydes of great interest is malonaldehyde (MDA). For example, 12-hydroperoxy linolenate may undergo a cyclization followed by an oxidation forming a 5-membered epidioxide as major product. The cleavage on each side of the endoperoxide leads to the formation of MDA. Malondialdehyde is reactive and potentially mutagenic.

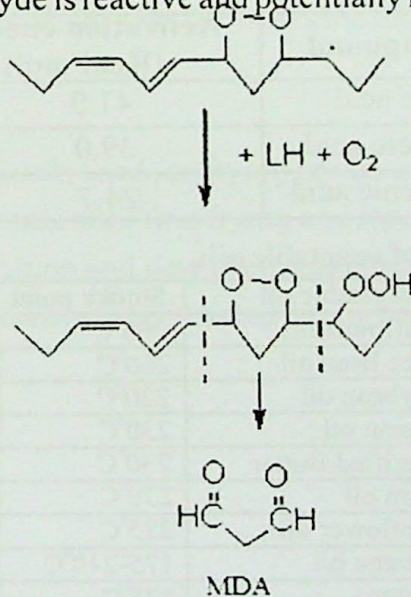


Fig. 14. Mechanism of formation of MDA from 12-hydroperoxy linolenate

Activation energies for the isomerization reactions in C18 unsaturated fatty acids and stability of vegetable oils

The activation energies for the isomerization reactions are shown in Table 3. It appears that the oleic acid is more stable than the other two polyunsaturated fatty acids. However, each of the glyceride molecules in vegetable oils usually contain three different fatty acid moieties and determining the stability of a vegetable oil based on its activation energy is difficult. Furthermore, when the oil is heated in the presence of oxygen the auto-oxidation process starts and the oil starts decomposing. Therefore, the stability consideration of vegetable oils must be based on their smoke points. The smoke points vary from oil to oil. Data with smoke points of different vegetable oils are given in Table 4.

Table 3. Activation energy for the isomerization reactions in 18:3 unsaturated fatty acids

Compound	Activation energy (Kcal/mol)
Oleic acid	41.0
Linoleic acid	39.0
Linolenic acid	24.7

Table 4. Smoke point of vegetable oils

Vegetable oil	Smoke point
Safflower oil	265°C
Rice Bran oil	260°C
Soybean oil	230°C
Peanut oil	230°C
Clarified Butter	230°C
Corn oil	230°C
Sunflower oil	225°C
Sesame oil	175-210°C
Butter	175°C
Coconut oil	175°C
Extra virgin olive oil	165-190°C

Choice of vegetable oils for frying purposes

The smoke point table clearly shows the type of cooking oil one should choose for different frying purposes. Extra virgin olive oil has a low smoking point and should not be used for medium temperature frying. Olive oil should only be used for low temperature frying and other cold food preparations. It is also clear from the table that most of the oils are only suitable for medium temperature frying.

Whatever oil is chosen for frying purpose, the oil is going to undergo several chemical reactions and going to accumulate a cocktail of chemical compounds that can be toxic. At present, it is difficult to determine the impact of all the chemical products formed during frying on our health. Our life style has changed and we rely on fast and convenient food. This facilitates the intake of toxic chemical compounds.

Vegetable oils used for frying produces several hundreds of chemical compounds after first frying and repeated frying produces further decomposition and new chemical compounds that are toxic. Reuse of frying oil should be avoided during cooking.

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Prof. Alfred A. Christy obtained his primary and secondary education from St. Patrick's College Jaffna. He entered then University of Ceylon (Peradeniya) and obtained a Bachelor degree in Chemistry (B.Sc., University of Peradeniya, 1976). The first position held by Professor Christy was as a Demonstrator/Assistant Lecturer in Chemistry at the University of Jaffna in Sri Lanka (1976-1977). He then



moved to Nigeria, where he worked first as a teacher and then as senior master in Chemistry at a local college (1977 - 1983) in Ondo State, Nigeria. He then moved to Bergen, Norway. In Bergen, he commenced furthering his education at the University of Bergen, while acting as a teaching assistant (1983-1987) and then as lecturer at the Department of Chemistry (1987-1991). He was awarded a master degree in Physical Chemistry in 1987 and Dr. Scient. Degree in 1990. He then worked first as a research Scientist (1991-1992) and later as a laboratory manager/research scientist in the FT-IR laboratory he built up at the department (1992-1998). He assumed a professorship in Chemistry at the Department of Chemistry, University of Agder in 1998.

Professor Christy has accumulated an impressive list of scientific achievements during his career. He has published over 100 papers in scientific journals and presented more than 90 lectures at conferences, institutions of higher learning and industrial establishments around the world.

Professor Christy's international standing earned him respect in his discipline and this fact is clearly reinforced by his appointment in the "Research Proposal Reviewer" for the National Science Foundation-Division of Earth Sciences, Instrumentation and facilities Program, USA. Furthermore, Professor Christy has been included in the "Marquis who's who in the world" and several other directories from 1991 onwards. He has also received several awards such as "Man of the Year 1994", "20th Century Achievement Award (1995)" and several other honours such "Five hundred leaders of influence in the world".

Prof. Christy has also been involved in International education as a Consultant & Co-ordinator for students in Norway, training Science teachers in Uganda on behalf of U.N.; serving in the scientific Committees for evaluating PhD theses from Universities in Pakistan and as an adviser and visiting professor at the University of Petroleum (Beijing) and Thammasat University in Thailand to name a few.