INCORPORATION OF ISOMERIC OCTADECENOIC ACIDS INTO ALK-1-ENYL MOIETIES OF CARDIAC GLYCEROPHOSPHOLIPIDS OF THE RAT

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1. Introduction

The alkyl and alk-1-enyl moieties of alkoxylipids in mammalian tissues consist predominantly of saturated and monounsaturated chains [1] that are derived from the corresponding long-chain fatty acids *via* aldehydes and alcohols [2]. Ether moieties derived from oleic acid, *cis*-9-octadecenoic acid, are common constituents of mammalian lipids [3]; their formation from the immediate precursor, *cis*-9-octadecenol, is well documented [4,5].

Little is known on alkoxylipids that might be derived from positional and geometrical isomers of oleic acid. So far, alk-1-enyl moieties resulting from vaccenic acid, *cis*-11-octadecenoic acid, have been found to occur in the ethanolamine phosphoglycerides of rat brain [6] and human brain [7]. Alk-1-enyl moieties derived from *cis*-13-octadecenoic acid have been detected in human brain [7] and rat placenta [8].

It is not known, so far, whether isomeric octadecenoic acids that are common constituents of dietary fats prepared by catalytic hydrogenation [9], are utilized by the enzymes involved in the biosynthesis of alkoxylipids. To assess whether such positional and geometrical isomers of oleic acid are incorporated into alkoxylipids, we have characterized the alk-1-enyl moieties of the cardiac lipids of rats fed partially hydrogenated triacylglycerols, which were abundant in isomeric *cis*- and *trans*-octadecenoyl moieties. Cardiac tissue was chosen because extensive biosynthesis of alkoxylipids is known to occur in the heart [1,5]. Our data reveal that most of the 'unnatural' *cis*- and *trans*-octadecenoic acids are indeed utilized for the biosynthesis of alkoxylipids in the rat.

2. Materials and methods

Male albino rats were fed, for 3 months, a diet containing 20% by wt of a partially hydrogenated soybean oil and the cardiac lipids were isolated as in [10,11].

The glycerophospholipids were separated from the total cardiac lipids by chromatography on layers of silica gel H using hexane–diethyl ether (60:40, v/v) as solvent and eluted from the adsorbent [12]. The major constituents of the glycerophospholipids, analyzed by two-dimensional thin-layer chromatography [13], were found to be diradylglycerophosphocholines and diradylglycerophosphoethanolamines. The glycerophospholipids were applied on a layer of silica gel H, the adsorbent was exposed to fumes of hydrochloric acid [14], and the aldehydes, liberated from the alk-1-enyl moieties, were separated by chromatography using hexane–diethyl ether (8:2, v/v) as solvent. The aldehyde fraction was then eluted from the adsorbent with diethyl ether, saturated with water.

The aldehydes derived from the alk-1-enyl moieties of the glycerophospholipids were reduced to alcohols [13] which were subsequently converted to alkyl acetates. Argentation chromatography of the alkyl acetates on layers of silica gel G, containing 20% silver nitrate, which were developed twice with hexane–diethyl ether (8:2, v/v), afforded *cis*- and *trans*-octadecenyl acetates. Each of these octadecenyl acetate fractions was subjected to reductive ozonolysis, and the resulting products, i.e., aldehydes and aldacetates, were analyzed by gas chromatography essentially as in [10].

3. Results and discussion

Isomeric octadecenoic acids of dietary origin have been shown to be incorporated in a specific manner into the lipids of mammalian tissues [10,11]. The individual *cis*- and *trans*-octadecenoic acids or their derivatives have been found to be utilized, although to varying degrees, by various enzyme systems, such as

Table 1
Octadecenyl groups in alk-1-enyl moieties of cardiac glycerophospholipids in comparison to octadecenoyl moieties of dietary triacylglycerols

		Positional isomer (% of total *cis* or *trans*)										
		Δ5	Δ6	Δ7	Δ8	Δ9	Δ10	Δ11	Δ12	Δ13	Δ14	Others
Octadecenyl groups in alk-1-enyl moieties of cardiac glycero-phospholipids	*cis*	<1	1	1	12	66	2	11	4	2	<1	1
	trans	1	1	1	3	20	11	14	13	10	25	1
Octadecenoyl moieties of dietary triacylglycerols	*cis*	<1	<1	<1	1	76	4	10	5	2	1	1
	trans	<1	<1	1	3	8	20	27	16	12	7	6

acyl-CoA synthetase [16,17], acyl-CoA:phospholipid acyltransferase [18,19], acyl-CoA:cholesterol-*O*-acyltransferase [20], acyl-CoA desaturase [21] and acyl-CoA elongase [22]. It is certainly of interest to know, whether the isomeric octadecenoic acids and their derivatives also serve as substrates for acyl-CoA reductase, alkyldihydroxyacetonephosphate synthase and alkyl desaturase, which are the key enzymes in the biosynthesis of alkoxylipids [23].

Our data, summarized in table 1, show the isomer distribution of *cis*- and *trans*-octadecenyl groups in the alk-1-enyl moieties of cardiac glycerophospholipids of rats that were fed partially hydrogenated triacylglycerols. The composition of positional isomers of *cis*- and *trans*-octadecenoyl moieties in the dietary triacylglycerols is given for comparison. It is evident from these results that each of the *cis*-5- to *cis*-13-octadecenoic acids and *trans*-5- to *trans*-14-octadecenoic acids of dietary origin is incorporated into alk-1-enyl moieties of cardiac glycerophospholipids. Analysis of cardiac glycerophospholipids of rats fed a diet, which was almost devoid of positional and geometrical isomers of oleic acid, revealed that the octadecenyl groups of the alk-1-enyl moieties were composed of *cis*-9 (72%) and *cis*-11 (28%) isomers only.

It is well established that glycerophospholipids containing alk-1-enyl moieties are derived from fatty acids via aldehydes, alcohols and alkylglycerophospholipids [2]. Therefore, it is quite obvious from our data (table 1), that most of the 'unusual' positional and geometrical isomers of oleic acid, when supplied with the diet, are utilized by the enzymes involved in the biosynthesis of alkoxylipids.

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