Food Chemistry 141 (2013) 3317–3322

Contents lists available at SciVerse ScienceDirect

Food Chemistry

journal homepage: www.elsevier.com/locate/foodchem

# Enzymatic synthesis of feruloylated lysophospholipid in a selected organic solvent medium

Hongli Yang [a], Ying Mu [a], Hongtao Chen [a], Zhilong Xiu [a,*], Tiankui Yang [a,b,*]

[a] School of Life Science and Biotechnology, Dalian University of Technology, Dalian 116024, People's Republic of China
[b] Wilmar (Shanghai) Biotechnology Research & Development Center Co., Ltd., Shanghai 200137, People's Republic of China

ARTICLE INFO

*Article history:*
Received 23 January 2013
Received in revised form 19 April 2013
Accepted 4 June 2013
Available online 13 June 2013

*Keywords:*
Feruloylated lysophospholipids
Ethyl ferulate
Lipase
Phosphatidylcholine
Enzymatic modification

ABSTRACT

Feruloylated lysophospholipids (FLPs) were firstly synthesized from phosphatidylcholine (PC) and ethyl ferulate (EF) using lipase-catalysed interesterification in selected solvents at controlled water content. Kinds of lipases and single solvents were screened. Novozym 435 and toluene were found to be the suitable biocatalyst and solvent, respectively. Then *tert*-butanol, *n*-butanol, chloroform, isopropanol, acetone and DMSO were respectively added into toluene in order to increase conversion of products. The results showed that toluene/chloroform could significantly increase the conversion and the optimal combination of toluene and chloroform was 90:10 (v/v). The optimal conditions generated for FLPs production were a substrate molar ratio of 5:1 (PC/EF), a PC's hydrolytic time of 1.5 h, an enzyme load of 60 mg/ml, a solvent dosage of 5 ml and a molecular sieves concentration (4 Å) of 100 mg/ml. Under these conditions, 40.51% of EF can be converted to FLPs, which were identified by TLC and HPLC–MS.

© 2013 Elsevier Ltd. All rights reserved.

## 1. Introduction

Ferulic acid (4-hydroxy-3-methoxy cinnamic acid, FA) is a phenolic acid that is ubiquitous in grains, vegetables and fruits. It has shown antioxidant activity, antimicrobial, anticarcinogenic and anti-inflammatory properties (Rice-Evans, Miller, & Paganga, 1997; Tung, Wu, Kuo, & Chang, 2007). Many studies confirmed the derivatives of FA could suppress inflammatory responses and skin tumour promotion (Murakami et al., 2002; Srinivasan, Sudheer, & Menon, 2007). However, practical application of FA and its derivative was actually limited due to their poor solubility in both lipophilic and hydrophilic systems and low bioavailability. To overcome this limits, the modification of FA with acyl groups could expand potential applications due to adjustment of the physicchemical properties of the product while maintaining desirable antioxidative properties. For example, the solubility of FA in lipophilic and/or hydrophilic systems could be increased by esterification with sugar and aliphatic alcohol, respectively (Compton, Laszlo, & Berhow, 2000; Nicks et al., 2012; Yang, Guo, & Xu, 2012). In addition, modification of FA through its incorporation into triglycerides was carried out (Laszlo & Compton, 2006; Yang, Glasius, & Xu, 2012). Nevertheless, to our best knowledge, there are none reports on modification of phospholipids to produce feruloylated phospholipids.

Phospholipids are good emulsifiers, stabilizers and antioxidants, and widely used in food, pharmaceutical and cosmetic industries. In addition, phospholipids are major constituents of cell membranes and play essential roles in biochemistry and physiology of cell functions (Guo, Vikbjerg, & Xu, 2005). Some researchers have attempted to prepare phospholipids complex using phospholipids as materials in order to improve the bioavailability of bioactive compounds by using emulsification technique, for example, puerarin-, curcumin- and silybin-phospholipids complex (Kuntal, Kakali, Arunava, Bishnu, & Pulok, 2007; Li, Yang, Chen, Chen, & Chan, 2008; Xiao, Song, Chen, & Ping, 2006). However, the emulsions were not welcome in practical uses because of their poor stability and unsatisfactory bioavailability. Thus, we reasoned that structuring phospholipids with FA could produce novel bifunctional feruloylated phospholipids, because the natural antioxidative properties of FA could be maintained, while permeability of FA across cell membrances could be improved. Meanwhile, changes in product partitioning as well as improved emulsification properties also expanded the applications of FA.

Since FA and phospholipids were heat-sensitive and oxidation-sensitive, chemical synthesis of feruloylated phospholipids was limited. On the contrary, enzymatic approach for incorporation of FA into phospholipids would be a preferable, in which there were

* Corresponding authors. Address: School of Life Science and Biotechnology, Dalian University of Technology, No. 2 Linggong Road, Dalian 116024, Liaoning Province, People's Republic of China. Tel.: +86 18621569666; fax: +86 21 58487667 (T. Yang), tel.: +86 411 84706369; fax: +86 411 84706369 (Z. Xiu).
*E-mail addresses:* zhlxiu@dlut.edu.cn (Z. Xiu), yangtiankui@wilmar-intl.com (T. Yang).

0308-8146/$ - see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.foodchem.2013.06.012

mild reaction conditions and no waste or by-product could be released into the environment (Sun et al., 2008; Zhang et al., 2000).

Organic solvents in enzymatic reactions provide numerous industrially attractive advantages, such as increased solubility of substrates, reversal of the thermodynamic equilibrium of hydrolysis reactions, suppression of water-dependent side reactions, alternation of substrate specificity and enantioselectivity, and elimination of microbial contamination (Doukyu & Ogino, 2010).

Therefore, in the paper, feruloylated lysophospholipids (FLPs) were enzymatically synthesized from phosphatidylcholine (PC) and ethyl ferulate (EF) as shown in Fig. 1A. The products FLPs included 1-FLP (1-feruloy-lysophosphatidylcholine) and/or 2-FLP (2-feruloyl-lysophosphatidylcholine). Two-steps reaction was applied to the synthesis of FLPs: Firstly, PC was hydrolyzed by lipase-catalysed to its lyso-form (lysophosphatidylcholine, LPC and glycerophosphatidylcholine, GPC); Secondly, EF was added in solution of LPC and GPC, and then the interesterification of LPC and GPC with EF was carried out under certain conditions. The parameters of enzyme sources and its load, single organic solvents, binary organic solvents and their dosage, substrate molar ratio, PC's hydrolytic time, molecular sieves type and concentration on the conversion of target products were investigated as well. The identification of produced FLPs was conducted by TLC and HPLC–MS.

## 2. Materials and methods

### 2.1. Enzymes and chemicals

Soybean phosphatidylcholine (PC content not less than 94%) was purchased from Aladdin, Inc. (Shanghai, China). FA (purity >99%) and EF (purity >99%) were purchased from Suzhou Chang Tong Chemical Co., Ltd. (Shanghai, China). The 3 Å 1/16 and 4 Å 1/16 molecular sieves were purchased from UOP Co., Ltd. (Shanghai, China). Novozym 435 (lipase B from *Candida antarctica*, immobilised on a macroporous resin), Lipozyme RM IM (lipase from *Rhizomucor miehei*, immobilised on an anionic exchange resin) and Lipozyme TL IM (lipase from *Thermomyces lanuginosus*, immobilised on silica granulation) were purchased from Novozymes (Shanghai, China). Free Phospholipase $A_1$ ($PLA_1$) was donated by Novozymes (Shanghai, China). All other solvents and reagents were of analytical or chromatographic grades.

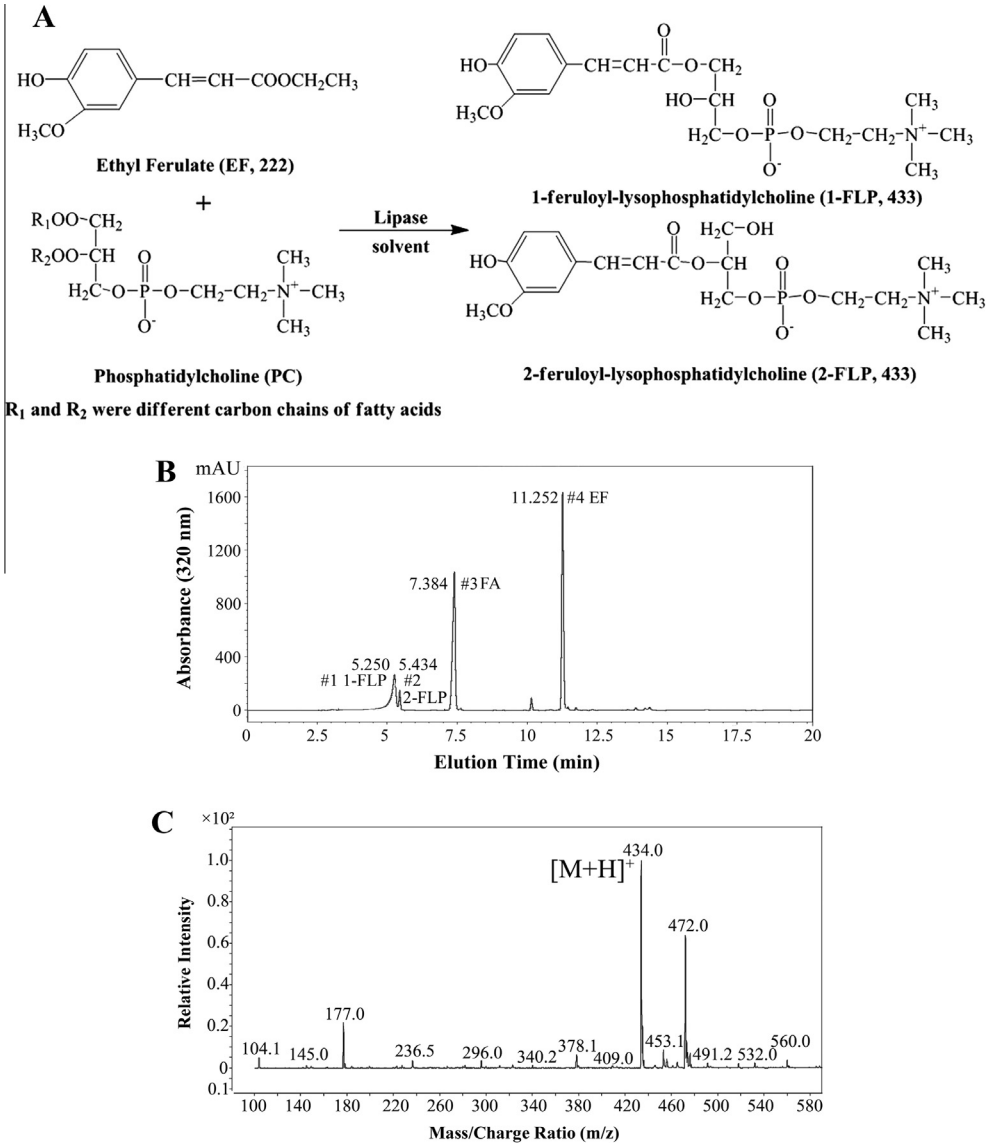


**Fig. 1.** (A) Reaction scheme of lipase-catalysed interesterification of PC and EF. (B) HPLC chromatogram analysis of components of interesterification reaction monitored by DAD/UV detection at 320 nm. Peak numbers were identified as followed: #1, 1-FLP; #2, 2-FLP; #3, FA and #4, EF. (C) Spectrum of electrospray ionisation mass spectrometry (ESI-MS) analysis of #1 and #2 (1-FLP and 2-FLP, respectively).

Download English Version:

https://daneshyari.com/en/article/10539790

Download Persian Version:

https://daneshyari.com/article/10539790

Daneshyari.com