Study on Preparation of Conjugated Linoleic Acids with Alkali Catalyst from Natural Unsaturated Fatty Acid Methyl Esters

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Abstract: Conjugated linoleic acids (CLAs) are a group of positional and geometrical isomers of conjugated octadecadienoic acids, which are naturally unsaturated fatty acid with conjugated double bonds. CLAs have been widely applied in food, pharmaceutical, health care, cosmetics and many other fields, due to its anticarcinogenesis and anti-atherosclerosis, participating in fat metabolism and enhancing immune function. Chemical modification methods, therefore, to prepare CLAs from natural unsaturated fatty acids have attracted more and more attention nowadays, which mainly include dehydration of ricinoleic acid in castor oil, alkali- or metal-catalyzed isomerization of linoleic acid, enzyme-catalyzed transformation and microorganism fermentation. In this paper, breaking traditional synthetic methods, synthesis of CLAs with methyl linoleate as starting material has been investigated. The synthesized CLAs were characterized by GC and UV-Vis. The content of CLAs reached 90% with sodium hydroxide as catalyst, which concentration was 20% compared with methyl linoleate, at 170 °C for 4 h.

Key words: catalysis, conjugated linoleic acid, synthesis, fatty acid methyl esters

Introduction

In 1978, the researchers from Wisconsin found some substance in fried ground beef, which could suppress cells mutation and shown to inhibit carcinogenesis [1]. In 1987, the group of Pariza notarized this substance as conjugated linoleic acids (CLAs) [2]. In recent years, with the depth understanding of CLAs various healthy physiological functions and the increasing demand, how to prepare low-cost and high-purity CLAs has become the focus in food, pharmaceutical, health care, chemicals and other related areas. Chemical modification methods, therefore, to prepare CLAs from natural unsaturated fatty acids have become the mainstream, which mainly include dehydration of ricinoleic acid in castor oil [3], alkali- or metal-catalyzed isomerization of linoleic acid (LA) [4-5] and biosynthesis [6-7].

But the shortcomings of the aforementioned methods are obvious. The reaction of dehydration of ricinoleic acid in castor oil is not only difficulty to complete, but provides low CLAs yields. Although alkali-catalyzed isomerization of LA is simple, the reaction requires high temperature. It is also difficulty to purify and separate the isomers of CLAs in the products. Because of hydrogenation, the method of metal-catalyzed isomerization of LA is infected in different degrees on the fractional conversion and selectivity of isomerization. Biosynthesis is more flexible and convenient, besides, the most important feature is that the product of CLAs is single isomer. But
the disadvantage is the low-yield which makes the industrial product impossible. Therefore a simple and economical method to obtain CLAs has become the focus of current research.

From current study, the method of alkali-catalyzed isomerization is the most promising approach. If improved, it would significantly reduce costs, which made prepare CLAs from natural fatty acid tend to be practical. In this paper, methyl linoleate was used as the precursor to obtain CLAs, which could avoid saponification in conventional alkali-catalyzed process, as well enhance the mobility of the reaction system. Specifically, at certain reaction time and temperature, with methyl linoleate as starting material, sodium hydroxide as catalyst, the synthesis of CLAs was investigated in details by changing the quantity of catalyst. The synthesized of CLAs were characterized and compared with each other by UV-Vis.

Experimental Section

**Materials.** LA (98 %) was purified from safflower oil. The other chemicals and reagents (analytical grade) were all purchased from Sinopharm Chemical Reagent Co., Ltd. (SCRC). Ultrapure water, with the resistivity of 18.2 MΩ·cm, was twice distilled and filtered.

**Methyl Linoleate.** 50 g of purified LA was esterified by refluxing for 5h in a solution of 1 % H₂SO₄ in methanol (30 g) under nitrogen atmosphere. After cooling the mixture to room temperature, the resulting methyl esters were washed with ultrapure water, dried over anhydrous sodium sulfate, filtered, and concentrated.

**Conjugated Linoleic Acids.** Typically, methyl linoleate (5 g) was added to 7.5 mL of ethylene glycol containing 2.5 g of sodium hydroxide. The mixture was heated at 170 °C for 4 h while a slow stream of nitrogen was passed through the reaction mixture. The reaction mixture was acidified with 10 % HCl to pH 3. The isomerized fatty acids were extracted with anhydrous ether. The combined ether layers were washed with ultrapure water, dried over anhydrous sodium sulfate, ether-evaporated, filtered, and concentrated.

**UV-Vis Analysis.** When the concentration of CLAs was 8×10⁻⁵ mol/L, the solutions of synthesized CLAs was analysis by UV-Vis spectrophotometer (Beijing Purkinje General Instrument Co., Ltd.).

**GC Analysis.** Samples (0.1 g) were added to the solution of boron trifluoride/ methanol (1 mL). The mixture was heated in boiling water for 5 min and then cooled to room temperature. 1 mL of anhydrous ether was added to extract the organic phase. GC was performed by using FULI 9790 instrument under the following conditions: SrAdv chromatographic workstation, PEG 20000 capillary column (30 m×0.32 mm), helium as carrier gas, column temperature for 220 °C, injector temperature for 270 °C, FID detector temperature for 270 °C.

Results and Discussion

As conjugated double bonds had obvious ultraviolet absorption peak at 234 nm in ultraviolet spectrogram, the peak would be increased after synthesis (Fig. 1). At certain reaction time and temperature, with sodium
hydroxide as catalyst, the synthesis of CLAs was investigated by changing the quantity of catalyst. The results were characterized and compared with each other by UV-Vis.

Under the same conditions, with the quantity of catalyst decreasing, the ultraviolet absorption peak became lower, which meant CLAs in products became less. With the detection of GC, while catalyst: methyl linoleate = 1:5 (w/w), sodium hydroxide as catalyst at 170 °C for 4 h, the content of CLAs reached 90 %, which still achieved good catalytic effect. Compared with conditional alkali-catalyzed process (alkali: fatty acid=1: 2, w/w, the content of CLAs was 92 %), as the content of CLAs was almost the same, the process with methyl linoleate as starting material could reduce the amount of the base, which reduced system saponification and increased the mobility.

Conclusions

At certain reaction time and temperature, with methyl linoleate as initial material and sodium hydroxide as catalyst, the synthesis of CLA was investigated in details by changing the quantity of catalyst. The synthesized CLAs were characterized and compared with each other by UV-Vis. The conclusions were as follows:

(1) The optimum quantity of catalyst was catalyst: methyl linoleate = 1:5 (w/w), with sodium hydroxide as catalyst at 170 °C for 4 h. And the content of CLAs was 90.74 % by detection of gas chromatography.

(2) Compared with conditional alkali-catalyzed process, as the content of CLAs was almost the same, the process with methyl linoleate as starting material could reduce the amount of the base and increase the system mobility, which achieved the expected affect.
References:


