Industrial Crops and Products 41 (2013) 312–318

Contents lists available at SciVerse ScienceDirect

Industrial Crops and Products

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

Essential oils and fatty acids composition of Tunisian, German and Egyptian caraway (*Carum carvi* L.) seed ecotypes: A comparative study

Bochra Laribi[a,*], Karima Kouki[a], Taoufik Bettaieb[a], Abdelaziz Mougou[a], Brahim Marzouk[b]

[a] *Institut National Agronomique de Tunisie, 43 Av. Charles Nicolle, 1082 Tunis, Tunisia*
[b] *Laboratoire des Substances Bioactives, Centre de Biotechnologie à la technopole de Borj-Cédria (CBC), BP 901, 2050 Hammam-Lif, Tunisia*

ARTICLE INFO

Article history:
Received 18 February 2012
Received in revised form 15 April 2012
Accepted 30 April 2012

Keywords:
Carum carvi L.
Seeds
Essential oil
Fatty acids
Petroselinic acid
Carvone

ABSTRACT

The present study aims to compare for the first time a Tunisian caraway seed ecotype with German and Egyptian ones regarding their fatty acid and essential oil compositions by using gas chromatography (GC) and gas chromatography–mass spectrometry (GC–MS) analyses. Results showed that total fatty acid (TFA) content varied from 2.90 to 7.30%, based on dry matter weight (DMW). The Tunisian ecotype exhibited the higher TFA proportion (7.30% DMW) than the two other ones. Petroselinic acid (C18:1n-12) was the major fatty acid in the three ecotypes, with the following proportions: 31.12% in TCE, 30.88 and 29.46% in GCE and ECE, respectively. Moreover, TCE contained a higher unsaturated fatty acid (UFA) proportion (87.86% TFA) than GCE (82.94% TFA) and ECE (80.76% TFA). The essential oil yield (based on dry matter weight) presented significant differences between the three caraway seed ecotypes studied. It is interesting to point out that the highest essential oil yield was observed in TCE (1.41%), followed by ECE (1.21%). In addition, 41 volatile compounds were identified in the seed essential oils of the three caraway ecotypes, the main ones being carvone (61.58–77.35%) and limonene (16.15–29.11%). This study showed that caraway seeds are rich in an unusual fatty acid, the petroselinic one. Additionally, the prevalence of unsaturated over saturated fatty acids is considered to be positive from the nutritional point of view. Consequently, the use of this oil in human dietary and in the alimentary industry seems to be a promising alternative. Besides, caraway seed essential oil obtained from the three caraway ecotypes displayed the same chemotype, namely carvone. This bioactive compound has various applications, as fragrance and flavour, potato sprouting inhibitor, antimicrobial agent and also in the medical field.

1. Introduction

Apiaceae represent one of the best-known plant families, widely distributed in temperate climate regions where they are often used as spices, vegetables or drugs owing to the presence of useful secondary metabolites such as essential oils (Olle and Bender, 2010). These lasts are produced within the various organs of medicinal and aromatic plants and widely used as food flavours (Burt, 2004). They are complex mixtures of volatile compounds, such as terpenes (mostly monoterpenes and sesquiterpenes), phenolics and alcohols (Lucchesi et al., 2004). Essential oils and their components are gaining increasing interest because of their relatively safe status, their wide acceptance by consumers, and their use in many applications including in flavours and fragrances as well as in medicine (Burt, 2004; Di Pasqua et al., 2005; Angienda et al., 2010).

Recently, researchers have stated that seeds of some Apiaceae genera should be regarded as a useful source for the extraction of petroselinic acid, which represents an important oleochemical raw material (Avato et al., 2001). For example, this acid can be used as a precursor of both lauric acid, which is a component of detergents and surfactants, and adipic acid, which is the monomeric component of nylon 66 (Murphy et al., 1994; Murphy, 1996).

The genus *Carum* is an important genus of the Apiaceae family, and contains about 20–30 species from Europe, North Africa and Asia. The best-known species of this genus is *Carum carvi* L. (caraway) which is one of the oldest aromatic and medicinal plants known (Nemeth, 1998; Papini et al., 2007). It has been used traditionally as a condiment or a spice due to its pleasant flavour (Johri, 2011). *C. carvi* seeds have also been used for the extraction of carvone, the main component of caraway essential oil, which inhibits sprouting in stored potatoes and onions (Toxopeus and Bouwmeester, 1993; Hartmans et al., 1995; Oosterhaven et al., 1995). Moreover, *C. carvi* essential oil has been shown to possess diuretic (Lahlou et al., 2007), antihyperglycaemic (Eddouks et al., 2004; Ene et al., 2007; Tahraoui et al., 2007), anti-hyperchlolesterol (Lemhadri et al., 2006) and anti-cancerous (Naderi-Kalali et al., 2005; Kamaleeswari et al., 2006) properties.

* Corresponding author. Tel.: +216 71 28 71 10; fax: +216 71 79 93 91.
E-mail address: bochra_laribi@yahoo.fr (B. Laribi).

0926-6690/$ – see front matter © 2012 Elsevier B.V. All rights reserved.
http://dx.doi.org/10.1016/j.indcrop.2012.04.060

In previous work dealing with the essential oil and fatty acid composition of Tunisian caraway seed ecotypes, we have demonstrated that their essential oil is characterized by the prevalence of carvone as the main volatile component and their seed oil is rich in petroselinic acid (Laribi et al., 2010). Thus, in order to demonstrate that Tunisian caraway ecotype can be considered as an important source of bioactive compounds, we compared it for the first time with the German and Egyptian ones by investigating their seed essential oil and fatty acid compositions.

2. Materials and methods

2.1. Plant material and growth conditions

Three caraway (*C. carvi* L.) seed ecotypes of cultivated origin were used in this study. The first one (Tunisian caraway, TCE) was collected from the field in the region of Souassi (South of Tunisia; latitude 35°34′12″N; longitude 10°16′6″E; altitude 41 m) while the two others were imported, one (GCE) from Germany (Munich, latitude 48°08′N; longitude 11°34′E; altitude 519 m) and the other (ECE) from Egypt (Luxor, latitude 32°39′N; longitude 25°41′E; altitude 82 m). The experiment was carried out in a greenhouse at day light (photoperiod varying from 13 to 16 h) and at a temperature varying from 18 to 20 °C during the day and from 10 to 12 °C during the night at the National Agronomic Institute of Tunisia (36°55′N, 10°11′E, 10 m above sea level). The three ecotypes were cultivated under the same environmental conditions. The experimental design was the complete random blocks with three replications. Each ecotype sown area was of 15 m^2 (10 m × 1.5 m). Seeds were sown on November 28, 2005 with row spacing of 0.40 m and by respecting a density of 125 plants m^{-2}. Fertilization consisted of 250, 200 and 100 kg ha^{-1} of P_2O_5, K_2O and N, respectively, incorporated uniformly to the soil before sowing, and supplemented by 100 kg ha^{-1} of N brought twice during the crop cycle. Pre-irrigation was done immediately after sowing for uniform emergence and establishment of seedlings. Irrigation was done by submersion one to twice frequencies per week. In addition, weeds were controlled by hand when needed. Harvest was on May 27, 2006. Harvested seeds were air-dried and stored at 4 °C until use for further analysis.

2.2. Total lipids extraction

Total lipids from seeds were extracted by a modified method of Bligh and Dyer (1959). Thus, 1 g air-dried seed was fixed in boiling water for 5 min and then ground manually using a mortar and pestle. A chloroform/methanol/hexane (LabScan Ltd.) mixture (4:3:2, v/v/v) was used for total lipid extraction (Marzouk and Cherif, 1981). After washing with water of fixation and decantation during 24 h at +4 °C, the organic phase containing total lipids was recovered and dried under a stream of nitrogen. The residue was dissolved in a known volume of toluene–ethanol (4:1, v/v) and stored at −20 °C for further analyses. Total lipid extraction was made in triplicate.

2.3. Fatty acid methylation and analysis

Total fatty acids of total lipids were converted into their methyl esters using 3% sodium methylate (Sigma Aldrich) in methanol according to the method described by Cecchi et al. (1985). Heptadecanoic acid methyl ester (C17:0) was used as an internal standard in order to quantify fatty acids. Fatty acid methyl esters (FAMEs) obtained were analysed by gas chromatography using a Hewlett-Packard HP 6890 gas chromatograph (Agilent Technologies, Palo Alto, CA, USA) equipped with a flame ionization detector (FID) and an electronic pressure control (EPC) injector. They were separated on a RT-2560 capillary column (100 m length × 0.25 mm i.d., 0.2 μm film thickness). The oven temperature was kept at 170 °C for 2 min, followed by a 3 °C min^{-1} ramp to 240 °C and finally held there for an additional 15 min period. Nitrogen (N_2, U) was used as carrier gas at a flow rate of 1.2 ml min^{-1}. The injector and detector temperatures were maintained at 225 °C. A comparison of the retention times of FAMEs with those of authentic standards (Labscan Ltd.) analysed in the same conditions was made to facilitate identification.

2.4. Essential oil extraction

Whole air-dried seeds (50 g) were subjected to hydrodistillation for 90 min (after a kinetic survey for 30, 60, 90 and 120 min). The hydrodistillation was performed by a simple laboratory Quik-fit apparatus which consisted of a 1000 ml steam generator flask, a distillation flask, a condenser and a receiving vessel. Essential oils were extracted from the distillate using diethyl-ether as solvent (v/v) dried over anhydrous sodium sulphate then concentrated at +35 °C using a Vigreux column and stored at −20 °C prior to analysis. All experiments were done in triplicates and results were expressed on the basis of dry matter weight (DMW).

2.5. Gas chromatography with flame ionization detection (GC-FID)

Essential oil analysis was performed using a Hewlett-Packard 6890 gas chromatograph (Agilent Palo Alto, CA, USA) equipped with a flame ionization detector (FID) and an electronic pressure control (EPC) injector. A polar HP Innowax (PEG) column and an apolar HP-5 one (30 m × 0.25 mm, 0.25 μm film thickness) were used. The carrier gas was nitrogen (N_2, U) at a flow rate of 1.6 ml min^{-1}. The split ratio was 60:1. The analyses were performed using the following temperature program: oven temperature isotherm at 35 °C for 10 min, from 35 to 205 °C at the rate of 3 °C min^{-1} and isotherm at 225 °C during 10 min. The injector and detector temperatures were held, respectively, at 250 and 300 °C.

2.6. Gas chromatography–mass spectrometry (GC/MS)

The GC/MS analyses were performed on a gas chromatograph HP 6890 (II) interfaced with a HP 5973 mass spectrometer (Agilent Technologies, Palo Alto, CA, USA) with electron impact ionization (70 eV). A HP-5MS capillary column (60 m × 0.25 mm, 0.25 μm film thickness) was used. The column temperature was programmed to rise from 40 to 280 °C at a rate of 5 °C min^{-1}. The carrier gas was helium with a flow rate of 1.2 ml min^{-1}. The scan time and mass range were 1 s and 50–550*m*/*z*, respectively. The injected volume was 1 μl.

2.7. Compounds identification

Essential oil components were identified by comparison of their retention index (RI) relative to (C_7–C_{20}) *n*-alkanes with those of the literature and/or with those of authentic compounds available in our laboratory, and by matching their recorded mass spectra with corresponding data (Wiley 275.L library) and other published mass spectra (Adams, 2001). FAMEs were identified by comparison of their retention times with those of pure reference standards. Quantitative data were obtained from the electronic integration of the FID peak areas.

2.8. Statistical analysis

Data were subjected to statistical analysis using the program package STATISTICA (Statsoft, 1998) and expressed as mean ± standard deviation (SD). The mean values were compared by using the one-way analysis of variance (ANOVA) followed by