(19) Europäisches Patentamt
European Patent Office
Office européen des brevets

(11) EP 0 988 311 B9

(12) **CORRECTED EUROPEAN PATENT SPECIFICATION**

Note: Bibliography reflects the latest situation

(15) Correction information:
Corrected version no 1 (W1 B1)
Corrections, see page(s) 6

(48) Corrigendum issued on:
15.12.2004 Bulletin 2004/51

(45) Date of publication and mention
of the grant of the patent:
04.08.2004 Bulletin 2004/32

(21) Application number: **98926258.9**

(22) Date of filing: **03.06.1998**

(51) Int Cl.^{7}: **C07J 1/00**, C07J 63/00,
A61K 31/11

(86) International application number:
PCT/US1998/011456

(87) International publication number:
WO 1998/055497 (10.12.1998 Gazette 1998/49)

(54) BETULINOL DERIVATIVES

BETULINOLDERIVATE

DERIVES DE BETULINOL

(84) Designated Contracting States:
DE FR GB IT

(30) Priority: **04.06.1997 US 48621 P**

(43) Date of publication of application:
29.03.2000 Bulletin 2000/13

(60) Divisional application:
04075511.8 / 1 473 301

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- **V. POUZAR ET AL.: "Triterpenes LII: Reduction and steric interaction of 12-lupanone derivatives" COLLECT. CZECH. CHEM. COMM., vol. 41, no. 11, 1976, pages 3452-3466, XP001097915**
- **J. SEJBAL ET AL.: "Triterpenes Part CVI: Functionalization of lupane with cromium(VI)oxide. A remark on the structure of clerodone" COLLECT. CZECH. CHEM. COMM., vol. 61, no. 9, 1996, pages 1371-1379, XP001097929**

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Printed by Jouve, 75001 PARIS (FR)

Description

[0001] The present invention relates generally to betulinol derivatives and, in particular, to methods for making these betulinol derivatives.

[0002] Betulinol is one of the more plentiful triterpenes, constituting up to 24 per cent of the outer bark of the white birch (Betula alba) and as much as 35 per cent of the outer bark and about 5 per cent of the inner bark of the Manchurian white birch (Betula platyphylla) (Hirota et al., J.S.C.I. Japan, 47:922 (1944)). It also occurs in the free state in the barks of the following trees: the yellow and black birch (Steiner, Mikrochemie, Molisch-Festschrift, p. 405 (1936)), Corylus avellana, Carpinus betulus (Feinberg et al., Monatsh, 44:261 (1924); Brunner et al., Monatsh, 63:368 (1934); and Brunner et al., Monatsh, 64:21 (1934)), and Lophopetalum toxicum (Dieterle et al., Arch. Pharm., 271:264 (1933)). The exudate from the bark of Trochodendron aralioides, which constitutes Japanese bird-lime, contains betulin palmitate (Shishido et al., J.S.C.I. Japan, 45:436 (1942)). Betulin has also been isolated from rosehips (Zimmermann, Helv. Chim. Acta, 27:332 (1944)) and from the seeds of Zizyphus vulgaris Lamarck var. spinosus Bunge (Rhamnaceae) (Kawaguti et al., J. Pharm. Soc. Japan, 60:343 (1940)). Ruhemann et al., Brennstoff-Ch., 13:341 (1932) discloses the presence of betulin, allobetulin, and an "oxyallobetulin" in the saponifiable portion of a benzene-alcohol extract of mid-German brown coal. In addition, the following group of lupon-row derivatives from the birch cortex extract have been identified: (a) betulinol, (b) betulinic acid, (c) betulin aldehyde, (d) betulonic acid, and (e) betulon aldehyde (Rimpler et al., Arch. Pharm. Und. Ber. Dtsh. Ppharmaz Jes, 299: 422-428 (1995); Lindgren et al., Acta Chem., 20:720 (1966); and Jaaskelainen, P. Papperi Ja Puu-Papper Och Tra., 63:599-603 (1989)).

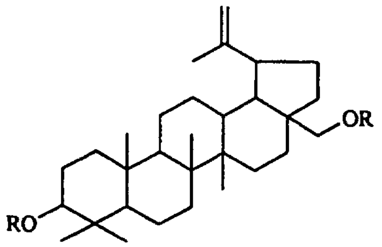
[0003] V. Pouzar et al., Collect. Czech. Chem. Comm., 41:3459 (1976) discloses a process for the preparation of a betulin diether involving reacting the corresponding dialcohol with methyl iodide.

[0004] Birch tree cortex-extracted betulinol was first mentioned as an antiseptic in 1899. Subsequently, compounds singled out from extracts of Hyptis emory and Alnus oregonu, identified as pentacyclic styrenes and their derivatives, were shown to inhibit carcinosarcoma growth (Sheth et al., J. Pharm. Sci., 61:1819 (1972) and Sheth et al., J. Pharm. Sci., 62:139-140 (1973)). It has been suggested that betulinic acid is the main anti-tumor agent in the mixture of terpenoids (Tomas et al., Planta Medicina, 54:266-267 (1988) and Ahmat et al., J. Indian Chem. Soc., 61:92-93 (1964)). In particular betulinic acid showed cytotoxic activity against carcinoma cell line CO-115 of the large intestine (LD 50 = 0.375 mg/ml) (Ukkonen et al., Birch bark extractive kemia kemi, 6:217 (1979)).

[0005] The use of chemotherapeutic agents in the treatment of a variety of cancers has become a well established part of cancer treatment regimens, especially where the disease has progressed to an advanced stage. However, these chemotherapeutic agents act not only on malignant cells but have adverse effects on non-target cells as well, particularly on the rapidly proliferating cells of the gastrointestinal tract and bone marrow. When employed in the high concentrations frequently required to be effective in killing cancer cells, these cytotoxic drugs give rise to undesirable and frequently severe side effects. Although the concept of site-directed chemotherapy is quite old, only a small number of antineoplastic drugs and toxins have been successfully coupled to monoclonal and polyclonal antibodies.

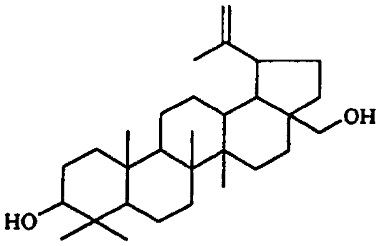
[0006] Therefore, a need continues to exist for chemotherapeutic agents and, in particular, for site-directed chemotherapeutic agents. The present invention is directed to meeting this need.

[0007] According to the invention, there is provided a method of synthesizing a diether having the formula:



wherein R is alkyl,
said method comprising:

providing a dialcohol having the formula:



and alkylating the dialcohol with a nitrile having the formula:

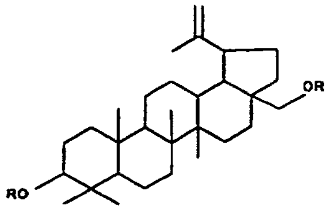
$$R\text{-}C\equiv N$$

under conditions effective to form the diether.

[0008] The diethers formed by the present invention can be used to treat patients suffering from cancer.

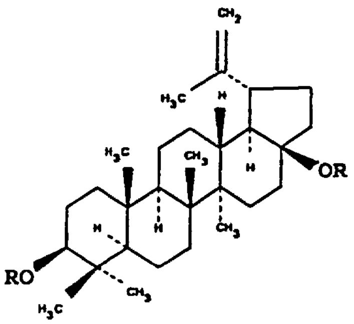
[0009] Figure 1 is a schematic flow diagram depicting a process for producing betulinol.

[0010] The present invention forms a diether having the formula:

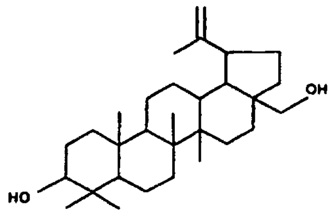


[0011] R is an alkyl group. R can be an unsubstituted alkyl, or it can be substituted with any number and combination of known substituents, such as sulfo, carboxy, cyano, halogen (e.g., fluoro, chloro), hydroxyl, alkenyl (e.g., allyl, 2-carboxy-allyl), alkoxy (e.g., methoxy, ethoxy), aryl (e.g., phenyl, p-sulfophenyl), aryloxy (e.g., phenyloxy), carboxylate (e.g., methoxycarbonyl, ethoxycarbonyl), acyloxy (e.g., acetyloxy), acyl (e.g., acetyl, propionyl), amino (including unsubstituted- monosubstituted-, and disubstituted-amino as well as cyclic amino groups (such as piperidino and morpholino) and the like. The alkyl group can be linear, branched, or cyclic. Illustrative examples of suitable alkyl groups include, methyl, ethyl, n-propyl, isopropyl, n-butyl, isobutyl, tert-butyl, n-pentyl, neopentyl, cyclopentyl, n-hexyl, and cyclohexyl. Preferably R is methyl, in which case the diether is betulinol dimethyl ether (also designated as "cornelon").

[0012] The diether formed by the present invention has a number of optically active carbon atoms. It is preferred that the diether be optically pure, and it is yet more preferred that each of the chiral centers in the diether have the conformation of that of naturally occurring betulinol as shown in the formula below:



[0013] The method according to the present invention includes alkylating a dialcohol having the formula:



with a nitrile having the formula:

$$R\text{-}C{\equiv}N$$

[0014] The dialcohol starting material for this reaction can be betulinol, such as betulinol isolated from natural products. Methods for isolating betulinol from a variety of sources are well known. For example, betulinol can be isolated from the outer layer of the bark of the white birch tree Betula alba by sublimation (Lowitz, Crell's Annalen, 1:312 (1788) and Mason, Silliman's Am. J., 20: 282 (1831), which are hereby incorporated by reference) or by extraction with an alcohol, such as ethanol (Hunefeld, J. Prakt. Chem., 7:53 (1836) and Hess, Poggendorff's Annalen, 46:319 (1839), which are hereby incorporated by reference). Other sources of betulinol and methods for its isolation and purification have been described in, for example, Sheth et al., J. Pharm. Sci., 61:1819 (1972) (raw vegetables and extracts of Hyptis emory) and Sheth et al., J. Pharm. Sci., 62: 139-140 (1973) (Alnus oregonu), which are hereby incorporated by reference.

[0015] In a preferred method, betulinol is isolated from the non-saponifiable substance of floral soap, such as, for example, by the method depicted in Figure 1. Briefly, the crushed initial leaf wood and components of a sulfate boiling procedure (NaOH, Na_2SO_4, $Na_2S_2O_3$, Na_2SO_3) are lodged to a boiling pot in a batch or continuous process. Under the temperature of 110°C to 120°C and, optionally, at increased pressure, lignin (the component of wood) dissolves. Crude cellulose is derived from the pulping liquor which is composed of lignin, cellulose, and black buck. Black buck is a composition of black buck with salts of tall acid and non-saponifiable substances. The crude cellulose is used in paper production, whereas the sulfate soap is separated from the black buck by centrifugation or by a settling process. Treatment of the sulfate soap with sulfuric acid produces tall oil. The non-saponifiable substances are separated as crude betulinol. Recrystallization of the crude betulinol, such as from acetone, ethyl acetate, isopropanol, butanol, ethanol, and the like, yields pure betulinol. The black buck residue present after centrifugation or settling can be advantageously recycled as shown in Figure 1.

[0016] Although the purity of the betulinol used as the starting material in the synthesis of the diether is not critical to the practice of the present invention, it is preferred that betulinol having a purity of at least 92-94% and a melting point of 241-243°C be used. Betulinol having these properties can be obtained using the preferred isolation and purification methods described above.

[0017] Once the dialcohol is provided, it is alkylated with a nitrile having the formula R-C≡N. The identity of the nitrile used depends on the identity of the R groups desired in the diether. For example, where betulinol dimethyl ether is desired, the nitrile is acetonitrile. Other nitriles suitable for use in preparing other diethers include propionyl nitrile, butyryl nitrile, pentanoyl nitrile, hexanoyl nitrile, benzylacetonitrile, and the like. Preferably the dialcohol and nitrile are present in at least a 1:2 molar ratio, more preferably, in a molar ratio of from about 1:20 to 1:60, and, most preferably, in a molar ratio of about 1:40. The reaction can be carried out without the use of a solvent in the case where the nitrile is a liquid in which the dialcohol is soluble, such as is the case where the nitrile is acetonitrile. In the case where the nitrile is a solid or a liquid in which the dialcohol fails to dissolve, the reaction can be carried out in a reaction solvent, preferably one in which both the dialcohol and the nitrile are appreciably soluble and with which neither reacts. Suitable solvents include, for example, ketone solvents, such as acetone, and chlorinated hydrocarbon solvents, such as methylene chloride and chloroform. The reaction can be carried out at a temperature from about room temperature to about the reflux temperature of the nitrile or the reaction solvent, preferably, from about 30°C to about 70°C, and, more preferably, at about 50°C. The duration of the reaction depends, in large measure, on the reactivity of the nitrile, the concentration of the reactants, and other factors. Typically, the reaction is carried out for a period of time from about 5 minutes to about 12 hours, preferably, from about 5 minutes to about 1 hour, and, more preferably, about 20 minutes.

[0018] Following the reaction, the diether is isolated. In cases where the diether is insoluble in the reaction medium (i.e., in the nitrile or in the reaction solvent), isolation is best carried out by filtering the precipitated diether, preferably after cooling the reaction mixture. In other cases, the diether can be separated, as an oil or as a precipitate, by addition of a solvent to the reaction mixture of a solvent in which the diether lacks appreciable solubility, typically an alkane, such as petroleum ether, or an ether, such as diethyl ether.

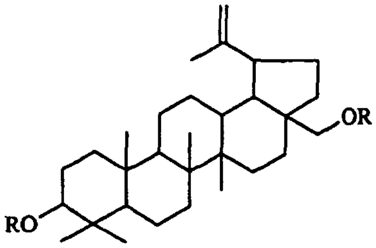
[0019] Once the diether is isolated from the reaction mixture, it can be purified, for example, by washing with a solvent, such as acetone, acetonitrile, methanol, and the like. Further purification can be carried out by standard techniques, such as recrystallization or chromatography.

Example -- Preparation of Cornelon

[0020] Betulinol was dissolved in acetonitrile in a betulinol-to-acetonitrile mole ratio of 1:40. The solution was heated to 50°C and stirred for 20 minutes. The crystalline residue, designated as "Cornelon", was washed with acetonitrile, filtered, and dried at 60°C. Cornelon was obtained in a 80-95% yield and was analyzed by HPLC.

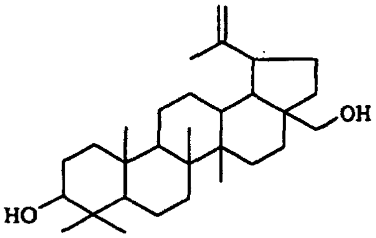
Claims

1. A method of synthesizing a diether having the formula:



wherein R is alkyl,
said method comprising:

providing a dialcohol having the formula:



and

alkylating the dialcohol with a nitrile having the formula:

R-C≡N

under conditions effective to form the diether.

2. A method according to claim 1, wherein R is methyl.

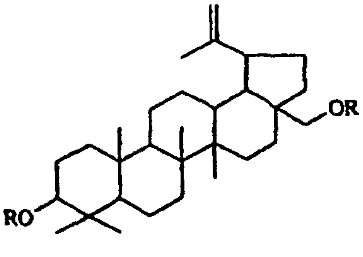
3. A method according to claim 1, wherein the dialcohol and the nitrile, respectively, are present in a mole ratio of from about 1:20 to about 1:60.

4. A method according to claim 1, wherein said alkylating is carried out at a temperature of from about

30°C to about 70°C.

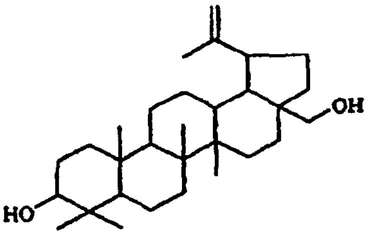
Patentansprüche

1. Ein Verfahren zum Synthetisieren eines Diethers der Formel:



in welcher R Alkyl ist,
wobei das genannte Verfahren folgendes umfasst:

Bereitstellung eines Dialkohols der Formel:



und
Alkylieren des Dialkohols mit einem Nitril der Formel:

R-C≡N

unter wirksamen Bedingungen zur Bildung des Diethers.

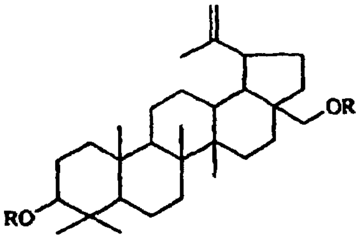
2. Ein Verfahren nach Anspruch 1, wobei R Methyl ist.

3. Ein Verfahren nach Anspruch 1, wobei der Dialkohol und das Nitril jeweils in einem Molverhältnis von ca. 1:20 bis ca. 1:60 vorliegen.

4. Ein Verfahren nach Anspruch 1, wobei die genannte Alkylierung bei einer Temperatur von ca. 30°C bis ca. 70°C durchgeführt wird.

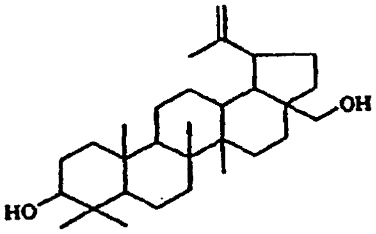
Revendications

1. Un procédé de synthèse d'un diéther possédant la formule :



dans laquelle R est un radical alkyle,
ledit procédé consistant en :

fournir un dialcool possédant la formule :



et
alkyler le dialcool à l'aide d'un nitrile possédant la formule :

R-C≡N

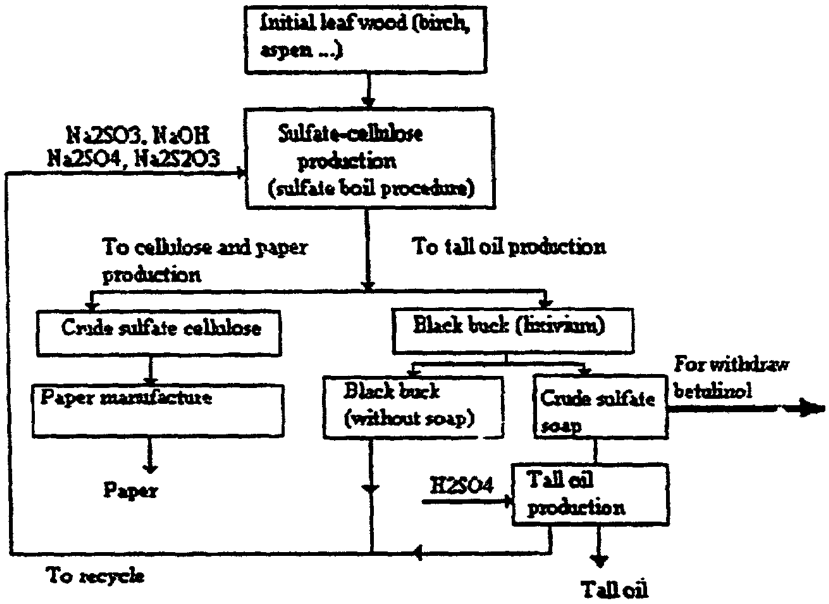
en des conditions efficaces pour former le diéther.

2. Un procédé selon la revendication 1, dans lequel R est un radical méthyle.

3. Un procédé selon la revendication 1, dans lequel le dialcool et le nitrile, respectivement, se trouvent présents dans une rapport molaire d'environ 1 :20 à environ 1 : 60.

4. Un procédé selon la revendication 1, dans lequel ladite alkylation est effectuée à une température d'environ 30 °C à environ 70 °C.

FIGURE 1


Initial leaf wood (birch, aspen ...)
Na2SO3, NaOH
Na2SO4, Na2S2O3
Sulfate-cellulose production (sulfate boil procedure)
To cellulose and paper production
To tall oil production
Crude sulfate cellulose
Black buck (lixivium)
Paper manufacture
Black buck (without soap)
Crude sulfate soap
For withdraw betulinol
Paper
H2SO4
Tall oil production
To recycle
Tall oil