SYNTHESIS OF 2,3-O,O-DIBENZYL-L-ASCORBICACID

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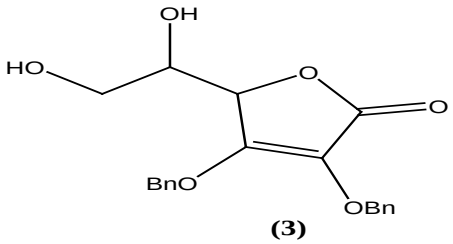
ABSTRACT

L-Ascorbic acid derivatives was synthesized on treatment with acetone and acetyl chloride afforded 5,6-acetal of L-ascorbic acid then benzylation of C-2&C-3 hydroxyl groups of the lactone ring was accomplished using K_2CO_3 and benzyl bromide in DMF. And then deblocking of the 5,6-O,O-protected derivative of L-Ascorbic acid with acetic acid and methanol gave 2,3-O,O-dibenzyl-L-Ascorbic acid. All the structures were characterized by ^1H NMR, ^{13}C NMR, D_2O exchange and Mass Spectroscopy.

Structure



(3)

(Where: Bn-Benzyl Group)

Fig 1. Structure of 2,3-O,O-dibenzyl-L-Ascorbic acid

Key word: L-Ascorbic acid, 5,6-acetal of L-Ascorbic acid, benzylation, hydrolysis.

INTRODUCTION

L-Ascorbic Acid is often called Vitamin C, abbreviated as LAA. L-Ascorbic Acid is one of the simplest vitamins and it's a vital nutrient for human beings. L-Ascorbic acid is the trivial name for the six-carbon sugar derivative L-thereo-hex-2-enono-1,4-lactone. Its derivatives are one of the important bio-molecules which act as anti-oxidant and radical scavenger is widely distributed in aerobic organisms. Thus it protects cellular compounds against oxidative damage by free radicals and oxidants[1]. Its derivatives have been found to possess antitumor and anti-viral activities [2]-[4]. It is also crucial for the bio-synthesis of collagen which is a main component of dentin, bone, connective tissue etc[5]. L-Ascorbic acid is present in all plants origin and is an essential micronutrient in man as a consequence of the absence of L-gulonolactone oxidase[6]. The well-known susceptibility of vitamin-C to thermal and oxidative degradation has led to interest in L-Ascorbic acid derivatives with increased stability. Numerous simple derivatives of L-Ascorbic acid have been synthesized and shown to possess important pharmacological properties. For example 5,6-di-O-modified ascorbic acid derivatives were clinically affective antitumor agents for various human cancer[7] and also induced apoptosis in tumor cells[8]. 2-C-alkylated derivatives have been shown to have immunostimulant activity[9], and 2-O and 3-O-alkylated lipid soluble derivatives are known to protect against the lipid peroxidation of the biomembrane[10]. Some pyrimidine and purine derivatives of 4,5-didehydro—5,6-dideoxy-L-ascorbic acid exerted pronounced cytostatic activities against malignant tumor cell lines[11],[12]. Furthermore, the biological importance of L-Ascorbic acid, known as putative palliative against the common cold, was initially associated with scurvy, the symptoms of vitamin deficiency [13]. The present study deals with the synthesis of 2,3-O,O-dibenzyl-L-Ascorbic acid.

MATERIALS AND METHODS

Materials

L-Ascorbic Acid was purchased from Sigma-Aldrich, USA, and other reagents such as Acetyl chloride, Acetone, benzyl bromide, Potassium carbonate, Dimethylformamide,

Acetic acid, Methanol etc. were purchased from Finar chemicals Ltd. India. Commercial solvents were used for work up note during synthesis. All the analyses were done in Sapala Organics pvt.Ltd, India. All experiments were done in Aevum Bio Labs pvt. Ltd, India.

General methods

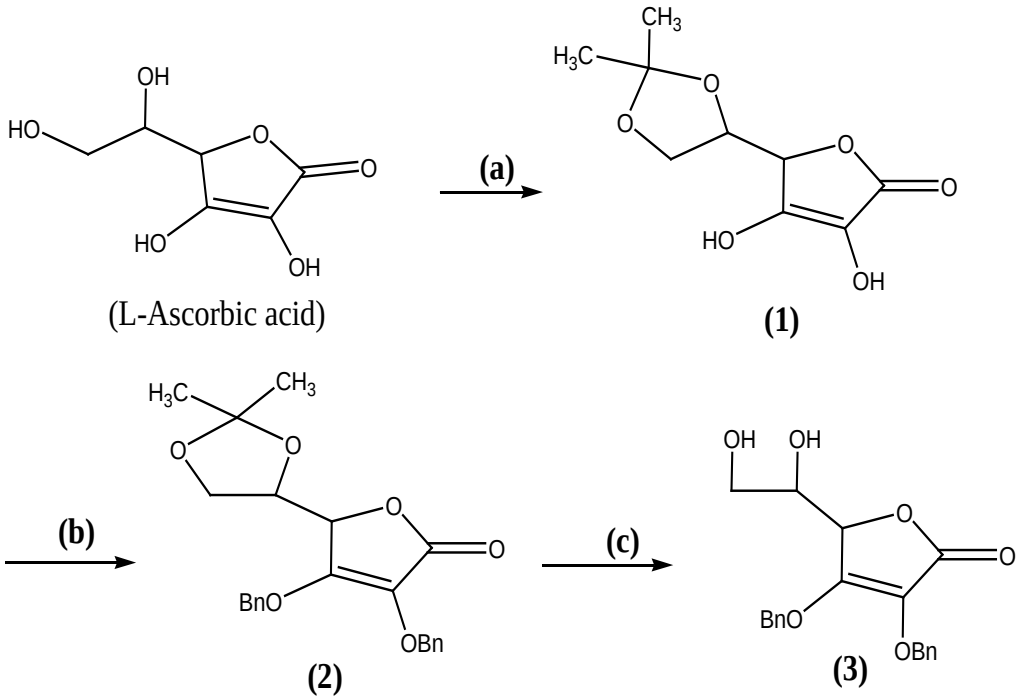
All the Ascorbic acid derivatives were characterized by ^{1}H and ^{13}C NMR and electron impact mass spectra. Melting points of compounds were determined with a Kofler micro hot-stage (Reichert, Wein) are uncorrected. recoated Merck silica gel 60F-254 plates were used for thin layer chromatography (TLC) and spots were detected under UV light (254 nm).the electron impact mass spectra were recorded with an EXTREL FT MS 2001 instrument with ionizing energy 70 eV. The ^{1}H and ^{13}C NMR spectra were recorded on a Varian Gemini 300 spectrometer, operating at 75.46 MHz for the ^{13}C resonance. The samples were dissolved in DMSO-d_6 or $CDCl_3$ chemical shift values are in ppm, referred to TMS. Acetone was dried over calcium chloride and followed by potassium carbonate in reflux condition. And DMF was dried over calcium hydride for above 12 Hrs in reflux condition.

RESULT AND DISCUSSION

Chemistry

The 5,6-acetal of L-Ascorbic acid (1), benzylation compound of the C-2 and C-3 hydroxyl groups (2) and deblocking compound of the 5,6-O,O-protected derivative (3) were synthesized as described previously by Von[14]. But in this paper it was synthesized with convenient method by the reaction with Acetic acid and methanol in aqueous media. It was modified by without distillation of solvent and column chromotography with good yield (97%) was outlined in scheme-1.

Scheme-1



Reagents and Conditions:(a)acetylchloride/acetone/rt/24hrs
(b)Benzylbromide/K_2CO_3/dimethylformamide/rt(c)50% aqueous acetic acid/methanol/85^{0}C/2hrs

Fig 2. Root of synthesis of 2,3-O,O-dibenzyl-L-Ascorbic acid.

EXPERIMENTAL SECTION

Synthesis of 5,6-O-isopropylidene-L-Ascorbic acid (1) was described previously by Von[14]. MP 198-202^{0}C, MS m/z 215 (MH^-). ^{13}C NMR (DMSO-d_6) δ: C-1(170.329), C-2(152.527), C-3(118.312), C-4(74.390), C-5(73.575), C-6(64.997), C-7(109.149), CH_3(25.941-25.537). ^{1}H NMR (DMSO-d_6) δ: H-4(4.714-4.708,d,1H), H-5(4.282-4.241,dt,1H), H-6(4.118-4.079,t,1H), H-6(3.901-3.864,t,1H), CH_3(1.255,s,6H), 2-OH(11.302,s,1H), 3-OH(8.489,s,1H).

D_2O Exchange: The peaks at 11.302 and 8.489 were exchanged by D_2O.

Synthesis of 5,6-O-isopropylidene-2,3-O,O-dibenzyl-L-Ascorbic Acid (2) was synthesized previously by Von[14]. MP 127-130^{0}C, MS m/z 397 (MH^+).^{13}C NMR ($CDCl_3$) δ: 168.976(C-1), 121.045(C-2), 156.487(C-3), 74.530(C-4), 73.658(C-5), 65.153(C-6), 110.120(C-7), 25.802 &25.562(CH_3), 73.806&73.658(CH_2), 135.839-127.664(C_6H_5). ^{1}H NMR($CDCl_3$)δ: 1.409 & 1.366(s,6H,CH_3), 4.026-3.988(dd,2H,H-6), 4.272-4.231(dt,1H,H-5), 4.537-4.530(d,1H,H-4), 5.205-5.063(m,4H,CH_2Ph), 7.401-7.195(m,10H,C_6H_5).

Synthesis of 2,3-O,O-dibenzyl-L-Ascorbic acid (3): To a solution of 2 (3.96g, 0.1 mol) in methanol (35.6 ml) and 50% Acetic Acid (22 ml+22ml of H_2O) was refluxed at 80-85^{0}C for 2 Hours (Note: At the time of addition reaction mass should be homogeneous solution). Then take Reaction mass, adjust P^H with Na_2CO_3 to neutral P^H and extract with Ethyl acetate (50 ml) with 20 minutes stirring. Separate out organic layer and wash with excess of water. Distill out organic layer afforded oily substance and then crystallize by N-Hexane and di-isopropyl ether (1:1).Reaction was monitored by TLC (R_f value; 0.3 & mobile phase; 5:5 of n-hexane and ethyl acetate) Product weight 3.45g (97%) MP 80.2-83^{0}C, MS m/z 356.9(MH^+). ^{13}C NMR (DMSO-d_6) δ: 169.437(C-1),158.166(C-2),120.691(C-3),74.670(C-4),68.792(C-5),61.687(C-6),136.292-127.738(C_6H_5),73.633,72.678(CH_2Ph).^{1}HNMR (DMSO-d_6) δ:4.902(1H,s,H-4), 3.733-3.681(1H,q,H-5),3.493-3.386(2H,m,H-6),5.270-5.192(2H,q,OCH_2),4.991-4.927(2H,q,OCH_2),5.165-5.150(1H,d,5-OH),4.887-4.873(1H,d,6-OH),7.429-7.314(10H,m,C_6H_5). D2O Exchange: At the region of 5.165-5.150(1H,d,5-OH), 4.887- 4.873(1H,d,6-OH) were exchanged by the action of D_2O.

CONCLUSION

The present work describes the synthesis of hydrolysis of 5,6-acetal of the lactone ring of **2** by using strategy of selective methods. Spectral analysis of 2,3-O,O-Dibenzyl-L-Ascorbic Acid (3) and the structures (1-2) were elucidated by ^{1}H NMR,^{13}C NMR, D_2O Exchange and Mass spectroscopy. The aim of research is the synthesis and biological screening of new nucleoside analogues of L-Ascorbic acid by using the L-Ascorbic derivatives and pyrimidine derivatives with new selective methods, which will be performed shortly.

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