

**SYNTHESIS OF A NEW ENANTIOMERICALLY ENRICHED
 α -AMINO ACID USING THE GLASER REACTION**

L. A. HAYRIYAN^{a,b}

^aScientific and Production Center “Armbiotechnology” of NAS RA
14, Gyurjyan Str., 0056, Yerevan, Armenia
Fax: (+37410) 654180

^bInstitute of Pharmacy, Yerevan State University
1, Alex Manoukyan Str., 0025, Yerevan, Armenia
Fax: (374-60)710410
E-mail: lianahayriyan@mail.ru

A method for the synthesis of a new enantiomerically enriched (S)-2-amino-8-hydroxy-octa-4,6-dienoic acid using the Glaser reaction has been developed. Ni(II) Schiff base complex of propargylglycine and a chiral auxiliary agent (S)-2-N-[N'-(benzylpropyl)-amino]benzophenone was used as the starting synthon. The enantiomeric excess (ee) of the isolated target amino acid exceeds 98%.

Table 1, references 10.

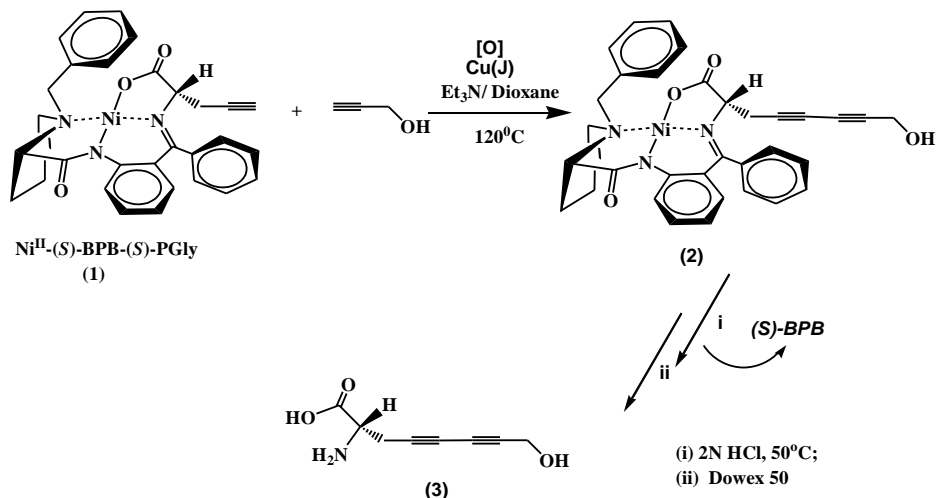
Non-protein amino acids have completely different application areas [1]. In humans, they play an important role as intermediate metabolic products, for example, in the biosynthesis of the neurotransmitter of gamma-aminobutyric acid (GABA) [2]. Many amino acids are used to synthesize other molecules, for example: tryptophan is a precursor of the neurotransmitter serotonin [3]. L-Tyrosine and its predecessor, phenylalanine, are precursors of the neurotransmitters dopamine, adrenaline and norepinephrine of catecholamines [4]. Glycine is the precursor of porphyrins [5]. Some non-protein amino acids are used by plants to protect against herbivores. For example, canavanine is an analogue of arginine, which is found in many legumes and in particularly large quantities in *Canavalia gladiata* (xiphoid canal) [6]. This amino acid protects plants from predators, for example insects, and when some untreated legumes are used

they can cause diseases in humans and poisoning in animals grazing in the areas of these plants growing [7].

Non-protein amino acids, which are derivatives of protein amino acids, contain radicals of different nature in the side group, as for example, acetylene, olefin, aromatic, heterocyclic substituents. Among these amino acids there is a large class of unsaturated amino acids that are considered to be valuable synthetic compounds in the field of organic chemistry. For example, studies of the biological activity of propargylglycine have shown that it strongly inhibits the growth of *Saccharomyces cerevisiae* and to a lesser extent, *E. coli*; it also acts as an amylase suppressor and activates pyridoxal phosphate-dependent enzymes [8].

This paper reports on the synthesis of a new unsaturated amino acid containing two acetylenic groups linked by a C–C bond (see Scheme). Ni^{II} complex of the Schiff base of (*S*)-propargylglycine and chiral auxiliary (*S*)-BPB (**1**) was used as the initial amino acid synthon, and propargyl alcohol was used as the second terminal agent. Complex **1** was synthesized according to the previously developed method [9].

Scheme



To determine the optimal conditions for the Glaser reaction of complex **1**, various systems (solvent and base) CH₃CN/K₂CO₃, CH₃CN/NaOH, 1,4-dioxane/K₂CO₃, 1,4-dioxane/NaOH, 1,4-dioxane/triethylamine were investigated. The reaction was carried out both at room temperature and when heated to 120°C and with periodic air supply. The reaction was monitored by TLC [SiO₂, CH₃COOC₂H₅/CH₃COCH₃=5/1] by the disappearance of traces of the starting complex **1** and the formation of products **2**. The results are shown in Table.

Table

Cross-coupling reaction of complex 1 with propargyl alcohol

Solvent	Basis, (eq.)	Duration, hour	T, °C	Chemical yield, % *
CH ₃ CN	K ₂ CO ₃ (3)	4	20	<5
CH ₃ CN	K ₂ CO ₃ (5)	4	100	15
1,4-dioxane	K ₂ CO ₃ (3)	6	45	10
1,4-dioxane	NaOH (3)	6	60	5
1,4-dioxane	NaOH (5)	8	120	7
1,4-dioxane	Triethylamine ^{**}	7	60	50
1,4-dioxane	Triethylamine ^{**}	10	120	60-65

The ratio of the original complex / CuJ/H $\text{C}\equiv\text{C}-\text{CH}_2-\text{OH}$ = 1/1/3

* – Chemical yield was determined according to TLC, according to the ratio of the original and the resulting complex

** – for 1 g of the initial complex 10 ml of 1,4-dioxane and 10 ml of triethylamine

As follows from the data of Table, the best results on the degree of conversion of the starting complex **1** were recorded in dioxane with heating and an excess of triethylamine base.

Isolation and purification of the target amino acid from the reaction mixture of the alkylated complex **2**, was carried out according to the standard procedure. The enantiomeric excess (ee) of the isolated amino acid, according to chiral HPLC, exceeded 99%.

Thus, as a result of the research, a new enantiomerically enriched (S)-2-amino-8-hydroxy-octa-4,6-dienoic acid (**3**) containing bis-acetylene bonds was obtained.

Experimental part

The ¹H and ¹³C NMR spectra were recorded on a Mercury 300 Varian instrument (300 MHz). Optical rotation was measured on a Perkin Elmer polarimeter, and the enantiomeric purity of the amino acid was determined by HPLC analysis using a chiral phase of the “Diaspher-110-Chirasel-E-PA” type 6.0 μm , column 4.0 \times 250 mm. The melting point was measured on a “Stuart SMP3” instrument. We used amino acid and other reagents from Aldrich and Reahim.

The technique of cross-coupling reaction. To 1 g (0.002 mol) of complex with 0.384 g (0.002 mol) of copper iodide in 10 ml of 1,4-dioxane, 10 ml of triethyl amine and 0.345 ml (0.006 mol) of propargyl alcohol were added. The mixture was stirred while heating to 110-120°C. The reaction was

monitored by TLC [SiO_2 , $\text{CH}_3\text{COOC}_2\text{H}_5/\text{CHCl}_3$ (3/1)] following the disappearance of traces of the initial complex **1**. After completion of the reaction, the mixture was extracted with CHCl_3 , concentrated, the complex was purified by column chromatography [SiO_2 , 3×20 cm, $\text{CH}_3\text{COOC}_2\text{H}_5/\text{CHCl}_3$ (3/1)].

Complex 2. Yield 66.6%. Calcd for $\text{C}_{33}\text{H}_{29}\text{N}_3\text{NiO}_4$ (589.15): C 67.14; H 4.95; N 7.12. Found: C 67.06; H 4.82; N 7.08. Mp. 229 °C. $[\alpha]_{\text{D}}^{20} = +2'447.3^\circ$ (c 0.15, CH_3OH). ^1H NMR (CDCl_3 , δ , J/Hz.): δ = 2.04-2.17 (2H, m, γ, δ -H Pro); 2.33 (1H, w, dd, $J=17.8$, 7.9, CH_2CH); 2.41-2.55 (1H, m, β -H Pro); 2.75 (1H, wd, $J=17.8$, CH_2CH); 2.90-3.02 (1H, m, β -H Pro); 3.20 (1H, br. OH); 3.52 (1H, dd, $J=10.8$, 6.1, α -H Pro); 3.56-3.67 (2H, m, γ , δ -H Pro); 3.63 (1H, d, $J=12.5$, CH_2Ph); 4.02 (1H, dd, $J=7.9$, 1.9, CHCH_2); 4.42 (1H, d, $J=12.5$, CH_2Ph); 4.23 (2H, ws, OCH_2); 6.66 (1H, dd, $J=8.2$, 2.2, C_6H_4); 6.69 (1H, ddd, $J=8.2$, 6.4, 1.1 C_6H_4); 7.03 (1H, br.d, $J=7.4$, Ar); 7.17 (1H, ddd, $J=8.6$, 6.4, 2.2, C_6H_4); 7.19 (1H, br. t, $J=7.4$, Ph); 7.26-7.30 (1H, m, Ar); 7.32-7.39 (2H, m, Ph); 7.45-7.56 (3H, m, Ar); 8.08-8.12 (3H, m, Ar); 8.13 (1H, dd, $J=8.6$, 1.1, C_6H_4). ^{13}C NMR (75.5 m.d., Hz, CDCl_3): δ = 23.9 (γ - CH_2 Pro); 24.3 ($\text{CH}_2\text{C}\equiv\text{C}$); 30.7 (β - CH_2 Pro); 51.2 (CH_2OH); 57.7 (δ - CH_2 Pro); 63.9 (CH_2 Ph); 67.0 (CH); 70.2 ($\text{C}\equiv\text{C}$); 70.8 (CH) 71.4 ($\text{C}\equiv\text{C}$); 75.8 ($\text{C}\equiv\text{C}$); 77.4 ($\text{C}\equiv\text{C}$); 120.9 (4-CH C_6H_4); 124.0 (6-CH C_6H_4); 126.3; 126.8 (CH); 127.8 (CH); 129.0 (3,3- \square -CH Ph); 129.0 (CH); 129.2 (CH); 129.3 (CH); 130.1 (CH); 131.7 (2,2- \square -CH Ph); 132.6 (3-CH C_6H_4); 133.5 (5-CH C_6H_4); 133.6; 134.2; 142.9; 172.0; 178.6; 180.6.

Isolation of the target amino acid from the complex was carried out according to the standard procedure [10]. To do this, 5 ml of CH_3OH was added to the complex and 5 ml of an aqueous solution of 2N HCl was added to the mixture with stirring and heating to 50°C. After the red color, characteristic of these complexes, disappeared from the solution, the initial chiral auxiliary reagent (S)-BPB was extracted with chloroform. The aqueous fraction was subjected to ion exchange purification using cation exchange resin Ku-2 \times 8 in the H^+ form (eluent 8% NH_4OH), the ammonia eluate was concentrated under vacuum.

(S)-2-Amino-8-hydroxy-octa-4,6-diynoic acid (3) Yield 48%. Calcd. for $\text{C}_8\text{H}_9\text{NO}_3$ (167.07), C, 57.48; H, 5.43; N, 8.38. Found: C 57.42; H 5.42; N 8.51. Mp. 202 °C, $[\alpha]_{\text{D}}^{20} = +77.3^\circ$ (c 0.15, HCl). ^1H NMR (D_2O , δ , m.d., Hz): δ = δ 4.37 (2H, t, $J = 0.9$, $\text{CH}_2\text{-OH}$), 4.01 (1H, t, $J = 5.6$, CH), 3.06 (2H, d, $J = 5.6$, CH_2).

ԷՆԱՆԹԵՐԻՈՄԵՐԱՊԵՍ ՆԱՐՍՏԱՅՎԱԾ ՆՈՐ α -ԱՄԻՆԱԹՈՎԻ ՍԻՆԹԵԶ ԳԼԱԶԵՐԻ ՌԵԱԿՑԻԱՅԻ ԿԻՐԱՌՄԱՐԲ

Լ. Ա. ՆԱՅՐԻԱՆ

Մշակվել է սինթեզի մեթոդ, հիմնված պրոպարգիլլիցինի և (S)-2-N-[(N'-բենզիլպրոլիլ)ամինա]բենզոֆենոն (BPB) քիրալային օժանդակ ռեագենտի Շիֆի հիմքի հետ Ni(II) իոնի առաջացրած հարթ-քառակուսային կոմպլեքսի ամինաթիվային մնացորդի վրա: Որպես ռեագենտ օգտագործվել են պրոպարգիլլայիրո. Գլազերի ռեակցիան իրականացվել է 1,4-դիօքսանի միջավայրում եռթիլամինի ներկայությամբ և տաքացման պայմաններում, օդի հոսքի պարբերաբար մղմամբ: Արդյունքում սինթեզվել է երկակի եռակի կապ պարունակող էնանթիոմերապես հարստացված նոր ամինաթիվ (S)-2-ամինո-8-հիդրոքսիօկտա-4,6-դիինաթիվ:

Սինթեզի ստերեոսեփականությունը կազմում է 90-92%, իսկ սինթեզված նպատակային ամինաթիվի էնանթիոմերապես մաքրության աստիճանը գերազանցում է 95%: Սինթեզված ամինաթիվի և միջանկյալ կոմպլեքսի կառուցվածքն ու բացարձակ կոնֆիգուրացիան որոշվել են ֆրիկաքիմիական անալիզի ժամանակակից մեթոդներով:

СИНТЕЗ НОВОЙ ЭНАНТИОМЕРНО ОБОГАЩЕННОЙ α -АМИНОКИСЛОТЫ С ПРИМЕНЕНИЕМ РЕАКЦИИ ГЛАЗЕРА

Л. А. АЙРИЯН^{а,б}

^аНаучно-производственный центр “Армбиотехнология”

НАН Республики Армения

Армения, 0056, Ереван, ул. Гюрджяна, 14

Факс: (+37410) 654180

^бЕреванский государственный университет, Институт фармации,

Армения, 0025 Ереван, ул. А. Манукяна, 1

Факс: (374-60)710410

E-mail: lianahayriyan@mail.ru

Разработан метод синтеза новой энантиомерно обогащенной (S)-2-амино-8-гидроксиокта-4,6-дииннокислоты с помощью реакции Глазера. В качестве исходного синтона был использован Ni(II) комплекс основания Шиффа пропаргилглицина и хирального вспомогательного реагента (S)-2-N-[N'-(бензилпролил)-амино]бензофенона.

В настоящей работе сообщается о синтезе новой ненасыщенной аминокислоты, содержащей две ацетиленовые группы, соединенные С-С связью. Наилучшие результаты по степени конверсии исходного комплекса были зафиксированы в среде диоксана при нагревании и избытке основания триэтиламина.

Стереоселективность синтеза составляет 90-92%, а степень чистоты синтезированной целевой аминокислоты превышает 95%. Структура и абсолютная конфигурация синтезированной аминокислоты и промежуточного комплекса определены современными методами физико-химического анализа.

REFERENCES

- [1] *Saghyan A.S.* // Enantiomerically pure non-proteinaminoacids:methods of production. Monograph, Science, Moscow, 2010, p. 341.
- [2] *Yehezkel Ben-Ari* // Nature Reviews Neuroscience, 2002, v. 3, №9, p. 728
- [3] *Murray R.K., Granner D.K., Mayers P.A., Rodwell V.W.* Harper's biochemistry, 1988, p. 700.
- [4] *Alenikov T.L., Rubtsova G.V.* // Guide to practical classes in biological chemistry, 1988, p. 178.
- [5] *Leninger A.* Fundamentals of biochemistry: T. 2. Per.: World. 1985, p. 662.
- [6] *Brown D.H., Fowder L.* //Phytochemistry, 1966, v. 5, p. 887.
- [7] *Ellington E.V., Hassall C.H., Plimmer J.R.* // J. Chem. Soc., 1959, p. 80.
- [8] *Gershon H, Meek JS, Dittmer K.* // J. Am. Chem. Soc., 1949, v. 71, p. 3573.
- [9] *Saghyan A.S., Petrosyan S.Gh., Mkrtchyan A.F., Mkrtchyan G.M., Langer P.* // Chem. Journal of Armenia, 2013, v. 66, p. 588.
- [10] *Belokon 'Yu.N., Tararov V.I., Maleev V.I.* // Tetrahedron: Asymmetry, 1998, v. 9, p. 4249.