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A One-Pot Procedure for the Preparation of *N*-9-Fluorenylmethyloxycarbonyl-α-amino Diazoketones from α-Amino Acids

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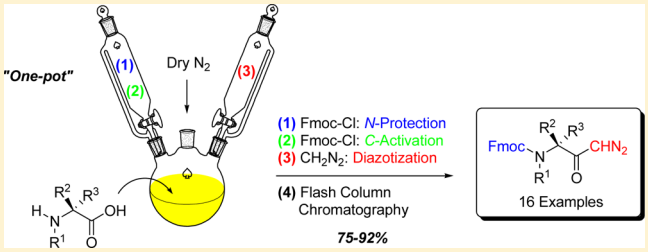
Supporting Information

ABSTRACT: The study describes a new "one-pot" route to the synthesis of *N*-9-fluorenylmethyloxycarbonyl (Fmoc) α-amino diazoketones. The procedure was tested on a series of commercially available free or side-chain protected α-amino acids employed as precursors. The conversion into the title compounds was achieved by masking and activating the α-amino acids with a single reagent, namely, 9-fluorenylmethyl chloroformate (Fmoc-Cl). The resulting *N*-protected mixed anhydrides were reacted with diazomethane to lead to the α-amino diazoketones, which were isolated by flash column chromatography in very good to excellent overall yields. The versatility of the procedure was verified on lipophilic α-amino acids and further demonstrated by the preparation of *N*-Fmoc-α-amino diazoketones also from α-amino acids containing side-chain masking groups, which are orthogonal to the Fmoc one. The results confirmed that *tert*-butyloxycarbonyl (Boc), *tert*-butyl (^{t}Bu), and 2,2,4,6,7-pentamethyldihydrobenzofuran-5-sulfonyl (Pbf), three acid-labile protecting groups mostly adopted in the solution and solid-phase peptide synthesis, are compatible to the adopted reaction conditions. In all cases, the formation of the corresponding *C*-methyl ester of the starting amino acid was not observed. Moreover, the proposed method respects the chirality of the starting α-amino acids. No racemization occurred when the procedure was applied to the synthesis of the respective *N*-Fmoc-protected α-amino diazoketones from L-isoleucine and L-threonine and to the preparation of a diastereomeric pair of *N*-Fmoc-protected dipeptidyl diazoketones.



INTRODUCTION

Amino acid-derived α-amino diazoketones have recently emerged as a new class of chiral group transfer agents.[1] Nowadays, it is possible to prepare a great number of terminal and internal chiral α-amino diazoketones by applying the available synthetic methods under mild and racemization-free conditions. These compounds are usefully employed to produce derivatives of certain synthetic and biological interest.[2,3] For example, they can be transformed into β-lactams having pharmaceutical applications.[4−6] Furthermore, chiral α-amino diazoketones obtained from natural α-amino acids have extensively been employed in classical Arndt−Eistert homologation protocols for the solution and solid-phase synthesis of enantiomerically pure β-amino acids[7−13] and β-peptide chains.[14−19] With respect to their wide synthetic applications, *N*-9-fluorenylmethyloxycarbonyl-protected (Fmoc) α-amino diazoketones cover a predominant position in the research fields discussed before. A high number of procedures suitable for the preparation of α-amino diazoketones from *N*-Fmoc-protected natural α-amino acids have already been published. The most common route is represented by the Arndt−Eistert step realized starting from differently carboxy-activated *N*-protected α-amino acids.[20,21] In this context, the acylation of diazoalkanes using acid chlorides and fluorides,[22,23] or less commonly acid anhydrides,[24,25] are familiar approaches for the synthesis of α-amino diazoketones. Nevertheless, some practical problems of safety due to the use of fluorinating reagents are associated with the preparation of protected *N*-Fmoc-α-amino acid fluorides,[26] while the corresponding chlorides cannot be prepared starting from α-amino acids containing acid-labile and the benzyloxycarbonyl (Z) groups. In fact, the application of these substrates is strongly hampered by their spontaneous decomposition into the corresponding Leuch's anhydrides. This limitation can be circumvented by the in situ treatment of the corresponding carboxylic acid with isobutyloxycarbonyl chloride or ethyl chloroformate to generate the expected mixed anhydride.[27,28] However, in these procedures the use of a tertiary base causes partial removal of the Fmoc protecting group and loss of chirality. Furthermore, it is to note that the reagents involved in the methods mentioned above must also be purified prior to use, in order to improve yields and avoid the formation of complex reaction mixtures containing unwanted side-products. Recently, the alternative use of the pentafluorophenyl ester method (OPfp) has been proposed[29] to overcome the problem of protecting group compatibility in the synthesis of *N*-Fmoc-α-amino diazoketones. The literature reports also on a number of

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different carboxy activating groups.[30−35] However, reactions are often characterized by low to unsatisfactory yields, and the stability and reactivity of the activated species have been reported as the main drawbacks of procedures based on the use of activated esters. The activation of carboxylic species in situ has recently been proposed.[36,37] Coupling agents as 2-(1*H*-benzotriazol-1-yl)-1,1,3,3-tetramethyl-uronium hexafluorophosphate (HBTU), and the 1-hydroxybenzotriazole/*N*,*N*′-dicyclohexylcarbodiimide (HOBt/DCC) system can be employed as possible routes to *N*-Fmoc-α-amino diazoketones.[38,39] In these cases, moisture can determine the formation of the corresponding *N*-Fmoc-α-amino acid methyl esters as side-products in a good extent. In addition, DCC is a potent skin irritant, and the insoluble DCU side-product needs to be completely removed, which is a cumbersome purification step. Finally, two improved Arndt−Eistert syntheses of α-diazoketones under milder conditions have been reported. The first method[40] implies the treatment of acid halides with an equimolar quantity of diazomethane in the presence of calcium oxide as acid trapping. The second one[41] proposes the activation of carboxylic acids by treatment in situ with the *N*-bromosuccinimide/triphenylphosphine (NBS/PPh_3) system. Nevertheless, the reaction of the acylphosphonium salt is extremely sensitive to moisture traces, temperature and purity of reagents. Besides, commercial triphenylphosphine must be purified by recrystallyzation prior the use in order to enhance its reactivity, and triphenylphosphine oxide formed as a reaction side-product cannot perfectly be removed also by column chromatography. More recently, other papers directed toward the synthesis and application of *N*-*Z*-protected α-amino diazoketones have been published.[42,43] The studies previously cited also reveal that all the known methods suffer from a delicate balance between both processes of diazotization and *C*-methyl ester formation, depending on the temperature, addition rate, and stability of the employed reagents. Hence, many approaches do not seem to be ideal for the efficient and versatile synthesis of chiral *N*-Fmoc-α-amino diazoketones. Diazomethane, despite its hazardous nature, remains the most important reagent employed in the diazotization of the activated carboxylic species. Trimethylsilyldiazomethane ($TMSCHN_2$) offers an alternative approach to the preparation of *N*-Fmoc-α-amino diazoketones,[44−46] but reactions require longer times upon temperature control, and the formation of the corresponding methyl ester cannot be avoided.

Thus, the preparation of *N*-Fmoc-protected α-amino diazoketones from natural, nonproteinogenic, and noncanonical α-amino acids deserves exploitation and represents a challenging synthetic target.

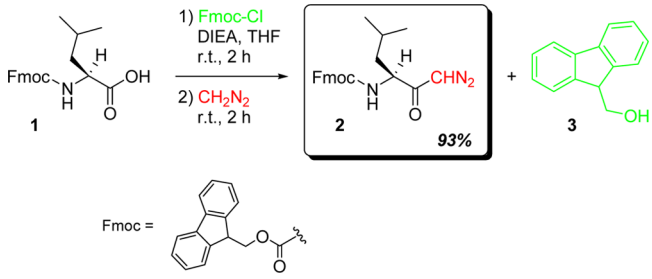
Our research group has previously been involved in studies concerning the protection/unblocking of α-amino acids by Fmoc-Cl,[47−50] and in the preparation of β-amino acids.[51−53] We thought that the use of a single reagent for the *N*-protection of α-amino acids and the coupling of the resulting species with diazomethane in situ could represent a valuable and short route to *N*-Fmoc-α-amino diazoketones. In fact, it is well-known that Fmoc-Cl is widely used for the *N*-protection of α-amino acids in the peptide chemistry. Moreover, this reagent has recently been proposed for the formation of mixed anhydrides not sensible to moisture, which may react smoothly in the presence of nucleophiles.[29,54] As consequence, we devised that Fmoc-Cl could be a suitable candidate in a "one-pot" synthesis of *N*-Fmoc-α-amino diazoketones, upon mild conditions, which may also allow the use of acid-labile orthogonal protections.

RESULTS AND DISCUSSION

In this work, we disclose a new rapid and clean method for the convenient synthesis of *N*-Fmoc-α-amino diazoketones starting from natural α-amino acids. In our procedure, α-amino acids are efficiently protected and activated with Fmoc-Cl in the presence of *N*,*N*-diisopropylethyl amine (DIEA) and then transformed into the corresponding *N*-Fmoc-α-amino diazoketones by diazotization in situ with a dichloromethane solution of diazomethane, prepared as previously reported.[55] Fmoc-Cl, proposed by Carpino[56−58] and Seela[59] for the introduction of Fmoc protection into α-amino acids, presents some evident and not discussable advantages: it is crystalline, stable, commercially available, and cheap. Moreover, it does not require any further purification step prior to use, and it is highly soluble in most of the solvents commonly adopted in the solution and solid-phase peptide chemistry. Finally, the mechanism of activation of the carboxylic function in an α-amino acid is well-known.[60]

The reactivity of Fmoc-activated α-amino acids to diazomethane was initially exploited, and the definition of the most appropriate reaction conditions for a possible one-pot strategy was further investigated. *N*-Fmoc-leucine (**1**, Scheme 1) was selected as the case of study.

Scheme 1. Preparation of *N*-Fmoc-α-amino Diazoketone 2 from *N*-Fmoc-leucine 1



In this preliminary test, 1 equiv of the *N*-Fmoc-protected α-amino acid was reacted with 1 equiv of Fmoc-Cl, in the presence of 1 equiv of DIEA in dry THF at room temperature. The total conversion of the starting *N*-Fmoc-α-amino acid required 2 h, as determined by TLC analysis of the reaction mixture. Diazomethane (3 equiv) was then added, and the stirring was continued for 2 h until the completion of the reaction. As well established by TLC, the spot corresponding to a commercial standard of *N*-Fmoc-leucine methyl ester was not detected, while the spot attributable to the *N*-Fmoc-protected α-amino diazoketone of leucine (**2**) was clearly visible together with that generated by the side-product 9-fluorenylmethanol (**3**). The formation of **3** was recognized by using a commercial sample as TLC standard. The ^1H NMR analysis of the crude material confirmed the absence of resonances attributable to the methyl ester group. Compound **2** was obtained pure by flash column chromatography in a 93% yield. The molecular structure of the expected *N*-Fmoc-α-amino diazoketone was confirmed by the complete monodimensional ^1H and ^{13}C NMR analysis. Spectral assignments were found to be in good agreement with those already reported for the same compound prepared by other methods.[9,51]

We thus exploited the feasibility of this protocol as a one-pot procedure in which Fmoc-Cl could be used for both the *N*-protection and the carboxy group activation. The designed

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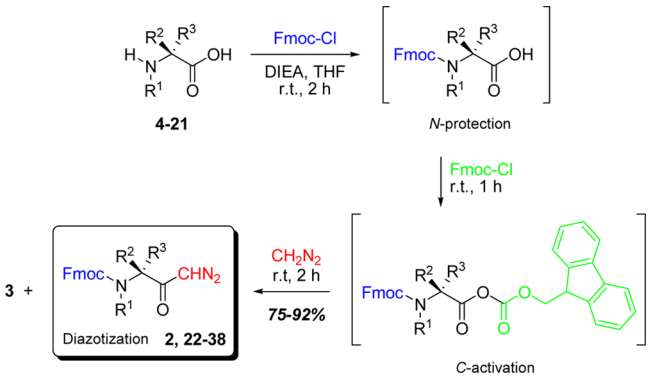
procedure was preliminary tested with the *N*-free α-amino acid leucine (**4**, Scheme 2). In this case, 1 equiv of the free α-amino

Scheme 2. One-Pot Synthesis of *N*-Fmoc-α-amino Diazoketones



acid hydrochloride dissolved in dry THF was initially treated with 1 equiv of Fmoc-Cl in the presence of 3 equiv of DIEA. The resulting suspension was stirred for 2 h at room temperature; after this time, TLC showed the complete consumption of the starting α-amino acid and the Fmoc-Cl. Another equivalent of Fmoc-Cl dissolved in dry THF was then added to the reaction mixture at room temperature, and stirring was maintained for an additional 1 h. Diazomethane was then added, and the mixture was stirred at room temperature until completion (2 h). The simple workup of the reaction mixture led to the isolation of a crude material containing the desired *N*-Fmoc-protected α-amino diazoketone and the side-product generated during the diazotization step, as established by TLC. The final product **2** was isolated pure by flash column chromatography in a 91% yield. Also in the case of this one-pot procedure, no traces of *N*-Fmoc-leucine methyl ester were detected by TLC as well as by ^{1}H NMR analysis performed on the crude material recovered from the reaction. It is worth noting that in this procedure only limited amounts of diazomethane and tertiary base are necessary for the formation of the expected products. Conversely, the most popular protocols involve the reaction of the carboxy activated derivatives with a larger excess of diazomethane.

Literature data are generally referred to the formation of a limited number of simple *N*-Fmoc-α-amino diazoketones. Therefore, we undertook the investigation of a larger number of free α-amino acids (**5**−**21**, Table 1). We also considered any possible different reactivity observable for unmasked α-amino acids, e.g., serine (**12**) and lysine (**13**), and the same starting materials with their side-chains containing acid-labile protections, namely the ^{t}Bu and Boc groups (**15** and **21**, respectively). The effectiveness and versatility of the method were definitively evaluated by exploiting the reactivity of tryptophan (**20**), without protection for the indolic NH group. The effects of diazotization in Arg containing the acid-labile Pbf group on the side-chain (**21**) were also considered. The results obtained are reported in Table 1.

Starting from α-amino acids **5**−**11** and **14**−**21**, the corresponding *N*-Fmoc-α-amino diazoketones **22**−**28** and **31**−**38** were uniquely formed. The process is very simple and can be distinct in three different moments. The *N*-protection is first effectuated by an equivalent of Fmoc-Cl and determines the total consumption of the starting material, as demonstrated by TLC. The addition of a second equivalent of the same reagent is used to activate the carboxy function by forming the corresponding mixed anhydride, which in turn reacts with diazomethane to generate the desired diazo compound together with the side-product **3**. The overall yields in final products recovered by flash column chromatography were good to excellent (75−92%), and the amounts of **3** isolated by chromatography were assumed as a further proof for the complete conversion of the starting materials observed in all cases. The general trend of higher yields for lipophilic α-amino acids was observed. The lowest value (75%) in this series was obtained for the treatment of glycine (**5**). In this case, the *N*-protection step did not proceed to completeness, most likely because of the limited solubility of **5** and of the corresponding

Table 1. *N*-Fmoc-α-amino Diazoketones from Commercial α-Amino Acids

R^1	R^2	R^3		α-amino acid	product	yield[a] (%)
H	$CH_2CH(CH_3)_2$	H	**4**	Leu	**2**	91
H	H	H	**5**	Gly	**22**	75
H	CH_3	H	**6**	Ala	**23**	87
H	$CH(CH_3)_2$	H	**7**	Val	**24**	92
H	$CH(CH_3)CH_2CH_3$	H	**8**	Ile	**25**	89
H	$CH_2C_6H_5$	H	**9**	Phe	**26**	90
H	CH_3	CH_3	**10**	Aib[b]	**27**	89
	R^1−R^2 = $(CH_2)_3$	H	**11**	Pro	**28**	88
H	CH_2OH	H	**12**	Ser	**29**	–
H	$(CH_2)_4NH_2$	H	**13**	Lys	**30**	–
H	$(CH_2)_2CONH_2$	H	**14**	Gln	**31**	83
H	$CH_2OC(CH_3)_3$	H	**15**	Ser(^{t}Bu)	**32**	90
H	$CH(CH_3)OC(CH_3)_3$	H	**16**	Thr(^{t}Bu)	**33**	91
H	$(CH_2)_4NHCO_2C(CH_3)_3$	H	**17**	Lys(Boc)	**34**	84
H	$CH_2CO_2C(CH_3)_3$	H	**18**	Asp(O^{t}Bu)	**35**	85
H	$CH_2C_6H_4OC(CH_3)_3$	H	**19**	Tyr(^{t}Bu)	**36**	90
H	CH_2Indolyl	H	**20**	Trp	**37**	86
H	$(CH_2)_3NHC(NH)NHPbf$[c]	H	**21**	Arg(Pbf)[c]	**38**	79

[a]Based on recovered products after flash-column chromatography. [b]Aib = 2-aminoisobutyric acid. [c]Pbf = 2,2,4,6,7-pentamethyldihydrobenzofuran-5-sulfonyl group.

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ammonium salt, which is initially formed through the reaction with DIEA. When the free α-amino acids **12** and **13** were subjected to the procedure, the initially formed white cloudy suspensions remained unaltered until the end. The course of reactions was checked by TLC as usual; in both cases, no spots other than those of the respective starting material and Fmoc-Cl were observed, indicating that the expected *N*-Fmoc-α-amino diazoketones **29** and **30** were not formed.

The results obtained with **5** and, successively, with **12** and **13**, quickly prompted us to realize that an evaluation of the reaction conditions employed was necessary, at least in terms of the solvent. Acetonitrile or dioxane were used in substitution of THF in the reactions performed with **5**, **12** and **13** at room temperature. The solubility of the starting material seemed to be not influenced by the change of the solvent, also when acetonitrile or dioxane were used as cosolvents jointly THF. In all cases, the formation of suspensions cannot completely be avoided. Among the three α-amino acids tested, only **5** furnished the expected product, which was isolated in overall yields not exceeding 65% also by prolonging the reaction time of each step of protection, activation, and diazotization until four hours at room temperature. As checked by TLC the formation of fluorene demonstrated that partial removal of the Fmoc group occurred. Consequently, THF was reputed to be the best solvent for the procedure, while the reaction times established by the experiment described for **4** were adopted in next experiments. Moreover, the reactivity of free serine (**12**) and lysine (**13**) suggested to us that the presence of masking groups on the α-amino acid side-chain could be desirable in order (i) to improve the solubility of α-amino acids bearing side-chain polar functionalities, and (ii) to obtain also the complete consumption of the starting materials. In all other cases reported in Table 1, the treatment successfully led to the formation of the desired *N*-Fmoc-α-amino diazoketones. None of the TLC and NMR data indicated the presence of the *C*-methyl ester of the corresponding starting α-amino acid. The ^{1}H NMR analysis of each crude reaction product also revealed the same, as well as the absence of signals attributable to side-products formed through the α-amino acid homocoupling. Moreover, the observation that all the activated species obtained from the α-amino acids used rapidly reacted with diazomethane suggested that steric hindrance due to bulkier side-chains seemed do not play a relevant role in the process. The structures of all final products were unambiguously established by ^{1}H and ^{13}C NMR spectroscopy. *N*-Fmoc-α-amino diazoketones **4**−**9**, **19**, **21**, **22**, **31** and **34** obtained by this procedure showed spectroscopic data in good agreement with those elsewhere reported for the identical compounds synthesized by applying different preparative methods.[9,27,28,39,51,61] Among the α-amino acids not containing protective groups in the side-chain, glutamine (**14**) constituted an interesting exception. In fact, in this case there was no need for protection of the reactive function in the side-chain, and solubility of the starting material was reputed appreciable. Analytical TLC showed complete consumption of **14** and the expected *N*-Fmoc-α-amino diazoketone **31** was recovered pure after flash column chromatography in a good overall yield (83%). The treatment of proline (**11**) led to the respective diazo compound **28** in a very high yield (88%). In this case mixtures of conformers were recognized. In fact, the ^{1}H and ^{13}C NMR spectra of pure **25** displayed two discrete and partially overlapped set of signals attributable to a 1:1 mixture of *cis* and *trans* conformers. These data strongly agreed with those previously reported by Ellmerer-Müller et al.[28] In the cases of side-chain masked α-amino acids, we considered the most commonly used acid-labile protective groups in the solution and solid-phase peptide chemistry. The α-amino acids **15**−**19** were smoothly transformed into the corresponding fully protected α-amino diazoketones **32**−**36**, which were isolated after chromatography in very high overall yields (84−91%). In none of these cases was the acid-labile masking group incorporated in the structure of the respective starting material cleaved.

Hunig's base and diazomethane could racemize the chiral center of the α-amino acid. In a first test, isoleucine (**8**) was used to evaluate the stability of the configuration of the asymmetric α-carbon atom upon the conditions adopted for the one-pot procedure. The retention of chirality in the *N*-Fmoc-protected diazo derivative **25** obtained from **8** was strongly supported by the NMR spectra of the pure compound. In fact, the proton spectrum of **25** exhibited a single set of data consistent with the supposed structure. These results, at least under the sensitivity limits of the spectroscopic technique used, suggested that no change of the configuration at the α-carbon atom of the amino acid scaffold occurred during the process. The ^{13}C spectrum of the same compound confirmed the absence of racemization, showing a unique series of signals. Similar conclusions were made on the basis of the data collected from the proton NMR analysis effectuated on a sample of the crude reaction product as obtained by applying the usual workup at the end of the process. In this spectrum no traces of other diastereomers of **25** were detected. Furthermore, the spectroscopic analysis of the pure diazo compound **33** obtained from the threonine derivative **16** showed that the integrity of the configuration of the chiral carbon atoms was retained as well as in the case of **8**.

The absence of racemization was definitively assessed by reversed-phase ultrafast HPLC-mass spectrometric analysis (RP U-HPLC/MS) of dipeptidyl diazoketones *N*-Fmoc-L-Ala-L-Leu-CHN_2 and *N*-Fmoc-L-Ala-D-Leu-CHN_2. These diastereomers were synthesized by a classical solution-phase Fmoc-chemistry strategy.[62] In this process, the enantiomeric *N*-Fmoc-α-amino diazoketones synthesized as usual from L- and D-leucine were deprotected with *N*,*N*-diethylamine (DEA) and coupled to *N*-Fmoc-L-Ala-OH, which was activated with the 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride/1-hydroxybenzotriazole (EDCI/HOBt) system in the presence of DIEA. The RP U-HPLC/MS analysis of each crude diastereomeric dipeptidyl diazoketone and of a mixture of them showed no detectable changes of the chirality. MALDI-MS and MS/MS analysis was also carried out for the determination of exact masses of the diastereomeric dipeptidyl diazoketones.

Tryptophan (**20**) allowed us to further explore all the possible limitations of the method and its applicability to heterocyclic α-amino acids with NH moiety sensitive to methylation. As expected, the treatment of **20** by the usual protocol provided the *N*-Fmoc-α-amino diazoketone **37** in very satisfactory overall yields (86%) after chromatography. No methylation of the NH function of the side-chain indolyl ring occurred, as assessed by ^{1}H NMR analysis performed on the crude material obtained from the reaction. This respect of the heterocyclic structure confirmed the high versatility of the method. Some other natural α-amino acids contain polar functionalities in the side-chain, which can be susceptible to alkylation upon a series of experimental conditions. Therefore,

we investigated the behavior of the arginine derivative **21** containing the acid-labile Pbf group on the side-chain guanidine moiety. In fact, it is well-known that the nature of the sulfonyl protecting groups can affect the acidity of the NH proton of the corresponding *N*-terminal and/or side-chain sulfonamide group. For example, the enhancement of the nucleophilic character of the sulfonamide nitrogen atom of differently protected *N*-sulfonyl α-amino acids allows the easy methylation and ethylation of the NH group.[63] The *N*-Fmoc α-amino diazoketone **38** was efficiently prepared from **21** in a high yield (79%). The ^{1}H NMR analysis of **38** did not show methylation of the guanidine nitrogen atoms. Also the proton NMR analysis of the crude reaction product did not reveal the presence of any other possible *N*-methylated compound. Thus, Pbf could be used as an *N*-protecting group orthogonal to the Fmoc one, not affecting the backbone of the sulfonyl amide residue.

CONCLUSIONS

In summary, Fmoc-Cl and diazomethane are proposed as the components of a reagent system for the advantageous synthesis of *N*-Fmoc-α-amino diazoketones. These compounds can be obtained from a wide variety of cheap, commercially available α-amino acids upon activation of the *C*-terminal carboxylic group. The procedure is based on a "one-step" preparative route of the desired diazo compounds. Fmoc-Cl is used either to protect the α-amino function or to activate the carboxylic moiety of the α-amino acids. The resulting *N*-Fmoc-protected α-amino diazoketones are then obtained by acylating diazomethane in situ. This new method works at room temperature, under very mild conditions. It presents excellent masking group tolerance and displays some evident advantages in comparison with other methods already appearing in the literature. Unwanted side-reactions are completely avoided by the high level of *C*-activation assured by Fmoc-Cl. No side-products other than Fmoc-CH_2OH, arising from the attack of diazomethane on the more electrophilic carbon of the respective activated anhydride, are formed during the process. The protocol is simple, highly efficient, rapid, and clean and uses moderate amounts of diazomethane and tertiary base. Furthermore, the data obtained for isoleucine, threonine, and the pair of diastereomeric *N*-Fmoc-L-Ala-L-Leu and *N*-Fmoc-L-Ala-D-Leu diazoketones suggest that the chirality of the α-amino acid scaffolds is preserved. Limitations of the method were reached with polar α-amino acids not containing protections on their side-chains due to the low solubility of the starting materials. All these peculiarities indicate that the here disclosed one-pot protection−activation−diazotization protocol appears to be an appealing and highly versatile tool for the laboratory preparation of *N*-Fmoc-α-amino diazoketones without racemization. It may be proposed for a widespread application to systems other than α-amino acids for a broad variety of scopes.

EXPERIMENTAL SECTION

Materials and Methods. Commercially available reagents were used as supplied unless stated otherwise. Solvents were purified and dried by the standard procedures and distilled prior to use. DIEA was distilled on ninhydrin and CaH_2. All the chiral α-amino acids were of the L-series. In the case of leucine, both the L- and D-enantiomers were used. ^{1}H NMR spectra were recorded at 300 MHz, while ^{13}C NMR spectra were measured at 75 MHz or at 125 MHz. Spectral analysis was performed at 293 K on diluted solutions of each compound by using $CDCl_3$ or DMSO-d_6 as the solvents. Chemical shifts (δ) are reported in ppm and referenced to $CDCl_3$ (7.25 ppm for ^{1}H and 77.0 ppm for ^{13}C spectra) and to DMSO-d_6 (2.51 ppm for ^{1}H and 40.0 ppm for ^{13}C spectra). Coupling costants (*J*) are reported in Hertz (Hz). Reaction mixtures were monitored by thin-layer chromatography (TLC) using silica gel 60-F_{254} precoated glass plates and UV light (254 nm) or 0.2% ninhydrin in ethanol and charring as visualizing agent. Kieselgel 60H without gypsum was used for flash column chromatography (FCC). LC−MS analysis was carried out using a U-HPLC instrument coupled to a triple-stage quadrupole mass spectrometer fitted with a heated electrospray ionization source (HESI II) operating in positive ion mode.[64] Chromatographic separation was achieved using a C_{18} RP analytical column (Hypersyl Gold, 2.1 × 50 mm, 1.9 μm particle size). The elution gradient consisted of H_2O (0.1% TFA) and CH_3OH (55:45). Flow rate was 0.4 mL/min. MALDI-MS analyses were performed using a 5800 TOF/TOF system, equipped with a Nd:YAG 1000 Hz laser.[65] A 1 μL aliquot of a premixed solution in CH_3CN (0.3% TFA) of each crude sample and α-cyano-4-hydroxycinnamic acid (α-CHCA; 5 mg/mL) was spotted on the matrix target, dried at room temperature, and directly analyzed. The MS spectra were acquired in reflectron mode averaging 2500 laser shots with a mass accuracy of 5 ppm. MALDI-MS/MS (CID) analysis was performed at a collision energy of 1 kV. Melting points were determined on a Kofler hot-stage apparatus and are uncorrected. All reactions were carried out using flame-dried glassware and under an inert atmosphere (dry N_2). The methylene chloride solution of diazomethane was prepared from *N*-methyl-*N*-nitrosourea using a classical procedure.[55] The concentration of the diazomethane solution (0.66 M) was determined by a back-titration performed with a standard benzoic acid solution. ***Caution!*** *Diazomethane is highly toxic.* Hence, this reagent must carefully be handled. Methylene chloride solutions of diazomethane are stable for long periods if stored on KOH pellets at −20 °C.

Preparation of the *N*-Fmoc-α-amino Diazoketone 2. A solution of Fmoc-Cl (1 mmol) in THF (10 mL) was slowly added dropwise to a solution of **1** (1 mmol) and DIEA (1 mmol). The solution was magnetically stirred for 2 h at room temperature, until the complete conversion of **1**, as monitored by TLC. Diazomethane (3 mmol) was then added, and the stirring was further maintained for 2 h at room temperature. Evaporation of the solvent under reduced pressure conditions afforded a residue, which was suspended in a 5% aqueous solution of $NaHSO_4$ (10 mL) and carefully extracted with ethyl acetate (3 × 10 mL). The organic layers were collected, washed with a 5% aqueous solution of $NaHCO_3$ (3 × 10 mL) and once with brine (10 mL), dried over Na_2SO_4 and filtered. Evaporation of the solvent to dryness under reduced pressure conditions afforded a crude product, which was purified by flash column chromatography to give **2** (351.0 mg) in a 93% overall yield.

Synthesis of of *N*-Fmoc-α-amino Diazoketones 2, 22−28 and 31−38. General Procedure. A solution of Fmoc-Cl (1 mmol) in THF (10 mL) was slowly added dropwise to a suspension of the appropriate α-amino acid hydrochloride **4**−**21** (1 mmol) and DIEA (3 mmol) in dry THF (10 mL). After the addition, the cloudy mixture was maintained under magnetic stirring for 2 h at room temperature, monitoring the complete consumption of the starting materials by TLC. A solution of Fmoc-Cl (1 mmol) was added dropwise, and the resulting mixture was stirred for 1 h at room temperature. Diazomethane (3 mmol) was then added, and the yellow mixture was maintained under stirring at room temperature for further 2 h. (Note that it was not possible to follow the disappearing of the starting color of the resulting mixture during the reaction; in fact, the addition of Fmoc-Cl made yellow the solution, and this color remained unaltered until the end of the process.) The organic solvent was removed under reduced pressure conditions to give a residue, which was dissolved in a 5% aqueous solution of $NaHSO_4$ (10 mL) and carefully extracted with ethyl acetate (3 × 10 mL). The organic layers were collected, washed with a 5% aqueous solution of $NaHCO_3$ (3 × 10 mL) and once with brine (10 mL), dried over Na_2SO_4 and filtered. Evaporation of the solvent to dryness under reduced pressure conditions afforded a crude product, which was purified by flash column chromatography. Compounds **2**, **22**−**28** and **31**−**38** were

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isolated in 75−92% overall yields. Compounds **2**, **4**−**9**, **19**, **21** and **22** showed NMR spectral characteristics similar to those elsewhere published for the same compounds obtained by other methods.[9,27,28,39,51,61] The enantiomer D of **2** was prepared by the same protocol starting from D-leucine.

2. Obtained as a pale yellow solid (343.5 mg, 91% yield): TLC R_f 0.40 (eluant: diethyl ether/petroleum ether 50:50); mp 92−94 °C (lit.[9] 90−91 °C); ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.91 (d, 6H, J = 6.2 Hz), 1.32−1.78 (m, 3H), 4.12−4.28 (m, 2H), 4.43 (m, 2H), 5.31 (s broad, 1H), 5.46 (d, 1H, J = 8.1 Hz), 7.28 (t, 2H, J = 7.4 Hz), 7.37 (t, 2H, J = 7.4 Hz), 7.57 (t, 2H, J = 7.4 Hz), 7.73 (d, 2H, J = 7.4 Hz); ^{13}C NMR (75 MHz, $CDCl_3$) δ 21.6, 23.0, 24.6, 41.1, 47.2, 53.7, 56.2, 66.5, 119.8, 124.9, 125.0, 127.0, 127.6, 141.2, 143.6, 155.9, 194.3. Anal. Calcd for $C_{22}H_{23}N_3O_3$: C, 70.01; H, 6.14; N, 11.13. Found: C, 70.26; H, 6.13; N, 11.09.

The enantiomer D of **2** was obtained as a pale yellow oil (301.9 mg, 80%). Anal. Calcd for $C_{22}H_{23}N_3O_3$: C, 70.01; H, 6.14; N, 11.13. Found: C, 70.18; H, 6.16; N, 11.16.

22. Obtained as a pale yellow solid (241.0 mg, 75% yield): TLC R_f 0.50 (eluant: chloroform/methanol 98:2); mp 109−111 °C (lit.[28] 110−112 °C); ^{1}H NMR (300 MHz, $CDCl_3$) δ 4.22 (t, 1H, J = 6.6 Hz), 4.30 (m, 1H), 4.45 (d, 2H, J = 6.6 Hz), 5.27 (s broad, 1H), 5.49 (m, 1H), 7.28−7.43 (m, 4H), 7.60 (m, 2H), 7.77 (d, 2H, J = 7.5 Hz); ^{13}C NMR (75 MHz, $CDCl_3$) δ 47.2, 50.3, 53.7, 67.0, 120.0, 125.1, 127.1, 127.7, 141.3, 143.7, 156.2, 190.0. Anal. Calcd for $C_{18}H_{15}N_3O_3$: C, 67.28; H, 4.71; N, 13.08. Found: C, 67,31; H, 4.70; N, 13.12.

23. Obtained as a pale yellow solid (291.8 mg, 87% yield): TLC R_f 0.62 (eluant: diethyl ether/petroleum ether 80:20); mp 116−118 °C (lit.[9,49] 110−112 °C and 118−119 °C); ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.37 (d, 3H, J = 7.1 Hz), 4.18−4.32 (m, 2H), 4.45 (m, 2H), 5.31 (s broad, 1H), 5.45 (d, 1H, J = 7.5 Hz), 7.29−7.45 (m, 4H), 7.60 (m, 2H), 7.78 (d, 2H, J = 7.2 Hz); ^{13}C NMR (75 MHz, $CDCl_3$) δ 18.5, 47.2, 53.5, 53.6, 66.7, 120.0, 125.0, 125.1, 127.0, 127.7, 141.3, 143.7, 155.6, 193.8. Anal. Calcd for $C_{19}H_{17}N_3O_3$: C, 68.05; H, 5.11; N, 12.53. Found: C, 68.28; H, 5.12; N, 12.50.

24. Obtained as a pale yellow solid (334.3 mg, 92% yield): TLC R_f 0.53 (eluant: diethyl ether/petroleum ether 70:30); mp 123−125 °C (lit.[9,49] 123−124 °C, 125−127 °C and 123−125 °C); ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.89 (d, 3H, J = 6.7 Hz), 0.98 (d, 3H, J = 6.7 Hz), 2.11 (m, 1H), 4.12 (m, 1H), 4.23 (t, 1H, J = 6.8 Hz), 4.44 (m, 2H), 5.34 (s broad, 1H), 5.46 (d, 1H, J = 7.1), 7.33 (t, 2H, J = 7.2 Hz), 7.41 (t, 2H, J = 7.2 Hz), 7.61 (m, 2H), 7.78 (d, 2H, J = 7.2 Hz); ^{13}C NMR (75 MHz, $CDCl_3$) δ 15.3, 17.4, 29.7, 31.0, 47.2, 54.8, 62.8, 66.9, 119.9, 125.0, 125.1, 127.1, 127.7, 141.3, 143.7, 156.3, 193.3. Anal. Calcd for $C_{21}H_{21}N_3O_3$: C, 69.41; H, 5.82; N, 11.56. Found: C, 69.58; H, 5.83; N, 11.58.

25. Obtained as a pale yellow solid (335.9 mg, 89% yield): TLC TLC R_f 0.42 (eluant: diethyl ether/petroleum ether 60:40); mp 141−143 °C (lit.[39,49] 143−144 °C); ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.97 (m, 6H), 1.18 (m, 1H), 1.49 (m, 1H), 1.80 (m, 1H), 4.21 (m, 1H), 4.26 (t, 1H, J = 6.6 Hz), 4.46 (m, 2H), 5.38 (s broad, 1H), 5.53 (d, 1H, J = 9.0 Hz), 7.34 (t, 2H, J = 7.2 Hz), 7.45 (t, 2H, J = 7.2 Hz), 7.64 (m, 2H), 7.82 (d, 2H, J = 7.2 Hz); ^{13}C NMR (75 MHz, $CDCl_3$) δ 11.4, 15.6, 24.5, 37.5, 47.2, 54.9, 62.2, 66.7, 119.9, 124.9, 125.0, 127.0, 127.6, 141.3, 143.7, 156.2, 193.3. Anal. Calcd for $C_{22}H_{23}N_3O_3$: C, 70.01; H, 6.14; N, 11.13. Found: C, 70.18; H, 6.14; N, 11.09.

26. Obtained as a pale yellow solid (370.3 mg, 90% yield): TLC R_f 0.44 (eluant: diethyl ether/petroleum ether 70:30); mp 134−136 °C (lit.[9,49] 133−135 °C, 136−137 °C); ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.05 (2 dd, 2H, J = 13.7 and 6.9 Hz), 4.19 (t, 1H, J = 6.8 Hz), 4.43 (d, 2H, J = 6.8 Hz), 4.49 (q, 1H, J = 6.9 Hz), 5.18 (s broad, 1H), 5.51 (d, 1H, J = 7.8 Hz), 7.15−7.36 (m, 7H), 7.42 (t, 2H, J = 7.5 Hz), 7.56 (t, 2H, J = 7.5 Hz), 7.78 (d, 2H, J = 7.5 Hz); ^{13}C NMR (75 MHz, $CDCl_3$) δ 38.3, 47.1, 54.8, 58.7, 66.8, 120.0, 125.0, 127.0, 127.7, 128.6, 129.5, 135.9, 141.3, 143.7, 155.7, 192.9. Anal. Calcd for $C_{25}H_{21}N_3O_3$: C, 72.98; H, 5.14; N, 10.21. Found: C, 73.08; H, 5.15; N, 10.24.

27. Obtained as a pale yellow solid (310.9 mg, 89% yield): TLC R_f 0.38 (eluant: diethyl ether/petroleum ether 70:30); mp 127−129 °C; ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.45 (s, 6H), 4.19 (t, 1H, J = 6.3 Hz), 4.49 (d, 2H, J = 6.3 Hz), 5.29 (s broad, 1H), 5.38 (s broad, 1H), 7.32 (t, 2H, J = 7.2 Hz), 7.40 (t, 2H, J = 7.2 Hz), 7.59 (d, 2H, J = 7.2 Hz), 7.77 (d, 2H, J = 7.2 Hz); ^{13}C NMR (75 MHz, $CDCl_3$) δ 24.9, 47.4, 52.2, 59.5, 65.9, 120.0, 124.9, 125.0, 127.0, 127.7, 141.3, 143.8, 154.9, 196.4. Anal. Calcd for $C_{20}H_{19}N_3O_3$: C, 68.75; H, 5.48; N, 12.03. Found: C, 68.65; H, 5.46; N, 11.99.

28. Obtained as a pale yellow solid (318.0 mg, 88% yield): TLC R_f 0.45 (eluant: diethyl ether/petroleum ether 80:20); mp 135−137 °C. ^{1}H NMR (300 MHz, $CDCl_3$) indicated a 1:1 mixture of carbamate conformers δ 1.72−2.18 (m, 8H), 3.47 (m, 4H), 3.96 (m, 1H), 4.16 (t, 1H, J = 5.7 Hz), 4.23 (t, 1H, J = 6.7 Hz), 4.30 (m, 1H), 4.39 (dd, 1H, J = 10.5 and 6.7 Hz), 4.53 (m, 3H), 4.97 (s broad, 1H), 5.29 (s broad, 1H), 7.26−7.44 (m, 8H), 7.50−7.64 (m, 4H), 7.76 (m, 4H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 23.3, 24.2, 29.5, 30.9, 46.7, 47.2, 47.3, 52.6, 53.3, 63.5, 63.8, 66.9, 67.1, 119.8, 124.7, 124.8, 125.1, 127.0, 127.4, 127.6, 141.2, 143.8, 154.6, 155.0, 194.3, 164.7. Anal. Calcd for $C_{21}H_{19}N_3O_3$: C, 69.79; H, 5.30; N, 11.63. Found: C, 69.83; H, 5.29; N, 11.59.

31. Obtained as a pale yellow solid (325.7 mg, 83% yield): TLC R_f 0.30 (eluant: chloroform/methanol 95:5); mp 131−133 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 1.68 (m, 1H), 1.91 (m, 1H), 2.14 (t, 2H, J = 7.5 Hz), 3.97 (m, 1H), 4.18−4.40 (m, 3H), 5.96 (s broad, 1H), 6.77 (s broad, 1H), 7.27 (s broad, 1H), 7.34 (t, 2H, J = 7.2 Hz), 7.42 (t, 2H, J = 7.2 Hz), 7.74 (t. 2H, J = 7.2 Hz), 7.89 (d. 2H, J = 7.20 Hz); ^{13}C NMR (75 MHz, DMSO-d_6) δ 26.9, 31.6, 47.2, 53.3, 58.5, 66.1, 120.6, 125.7, 127.5, 128.1, 141.2, 144.2, 156.4, 173.9, 195.5. Anal. Calcd for $C_{21}H_{20}N_4O_4$: C, 64.28; H, 5.14; N, 14.28. Found: C, 64.47; H, 5.15; N, 14.31.

32. Obtained as a viscous pale yellow oil (366.7 mg, 90% yield): TLC R_f 0.43 (eluant: diethyl ether/petroleum ether 60:40); ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.16 (s, 9H), 3.46 (dd, 1H, J = 9.0 and 5.4 Hz), 3.78 (dd, 1H, J = 9.0 and 3.3 Hz), 4.23 (t, 1H, J = 6.9 Hz), 4.28 (m, 1H), 4.43 (dd, 1H, J = 10.7 and 6.9 Hz), 4.57 (dd, 1H, J = 10.7 and 6.9 Hz), 5.40 (s broad, 1H), 5.68 (d, 1H, J = 7.8 Hz), 7.28−7.46 (m, 4H), 7.62 (m, 2H), 7.78 (d, 2H, J = 7.5 Hz); ^{13}C NMR (75 MHz, $CDCl_3$) δ 27.3, 47.2, 54.2, 58.3, 61.6, 66.5, 73.7, 119.4, 120.0, 124.9, 125.1, 126.9, 127.0, 127.6, 127.7, 141.2, 141.3, 143.6, 143.7, 155.9, 192.8. Anal. Calcd for $C_{23}H_{25}N_3O_4$: C, 67.80; H, 6.18; N, 10.31. Found: C, 67.94; H, 6.20; N, 10.29.

33. Obtained as a viscous pale yellow oil (383.6 mg, 91% yield): TLC R_f 0.73 (eluant: diethyl ether/petroleum ether 60:40); ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.08 (d, 3H, J = 6.3 Hz), 1.21 (s, 9H), 4.14 (m, 2H), 4.23 (t, 1H, J = 6.7 Hz), 4.44 (dd, 1H, J = 10.7, and 6.7), 4.51 (dd, 1H, J = 10.7 and 6.7), 5.60 (s broad, 1H), 5.86 (d, 1H, J = 6.9 Hz), 7.32 (t, 2H, J = 7.5 Hz), 7.40 (t, 2H, J = 7.5 Hz), 7.61 (d, 2H, J = 7.5 Hz), 7.77 (d, 2H, J = 7.5 Hz); ^{13}C NMR (75 MHz, $CDCl_3$) δ 18.8, 28.3, 47.3, 54.8, 63.5, 66.7, 67.1, 74.7, 120.0, 125.0, 125.1, 127.0, 127.7, 141.3, 143.7, 156.0, 192.5. Anal. Calcd for $C_{24}H_{27}N_3O_4$: C, 68.39; H, 6.46; N, 9.97. Found: C, 68.57; H, 6.44; N, 9.98.

34. Obtained as a pale yellow solid (413.8 mg, 84% yield): TLC R_f 0.48 (eluant: ethyl acetate/petroleum ether 50:50); mp 114−116 °C (lit.[9] 117−118 °C); ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.70−1.89 (m, 1H), 1.30−1.67 (m, 5H), 1.43 (s, 9H), 3.09 (m, 2H), 4.13−4.32 (m, 1H), 4.20 (t, 1H, J = 6.6 Hz), 4.38 (dd, 1H, J = 10.8 and 6.6 Hz), 4.50 (dd, 1H, J = 10.8 and 6.6 Hz), 4.58 (s broad, 1H), 5.33 (s broad, 1H), 5.53 (d, 1H, J = 6.9 Hz), 7.30 (t, 1H, J = 7.5 Hz), 7.31 (t, 1H, J = 7.5 Hz), 7.39 (t, 2H, J = 7.5 Hz), 7.59 (m, 2H), 7.76 (d, 2H, J = 7.5 Hz); ^{13}C NMR (75 MHz, $CDCl_3$) δ 22.3, 28.4, 29.7, 31.8, 39.9, 47.3, 53.8, 60.4, 66.7, 79.20, 120.0, 125.0, 127.1, 127.7, 141.4, 143.7, 156.1, 192.9. Anal. Calcd for $C_{27}H_{32}N_4O_5$: C, 65.84; H, 6.55; N, 11.37. Found: C, 65.92; H, 6.56; N, 11.40.

35. Obtained as a pale yellow solid (370.1 mg, 85% yield): TLC R_f 0.46 (eluant: diethyl ether/petroleum ether 60:40); mp 72−74 °C (lit.[28,39] 71−72 °C); ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.43 (s, 9H), 2.58 (dd, 1H, J = 17.3 and 5.0 Hz), 2.92 (dd, 1H, J = 17.3 and 4.1), 4.22 (t, 1H, J = 6.2 Hz), 4.36−4.52 (m, 2H), 4.63 (m, 1H), 5.35 (s broad, 1H), 5 84 (d, 1H, J = 9.4 Hz), 7.31 (m, 2H), 7.39 (m. 2H), 7.58 (m, 2H), 7.78 (d, 2H, J = 7.2 Hz); ^{13}C NMR (75 MHz, $CDCl_3$) δ 28.0, 36.8, 47.4, 50.4, 53.8, 66.7, 81.9, 120.0, 124.9, 125.1, 127.1, 127.7, 141.5, 143.5, 155.9, 170.6, 192.6. Anal. Calcd for $C_{24}H_{25}N_3O_5$: C, 66.19; H, 5.79; N, 9.65. Found: C, 66.23; H, 5.80; N, 9.67.

36. Obtained as a viscous yellow oil (435.2 mg, 90% yield): TLC R_f 0.38 (eluant: diethyl ether/petroleum ether 60:40); ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.34 (s, 9H), 2.99 (d, 2H, J = 6.9 Hz), 4.18 (t, 1H, J = 6.6 Hz), 4.41 (m, 3H), 5.08 (s broad, 1H), 5.39 (d, 1H, J = 9.0 Hz), 6.91 (d, 2H, J = 8.1 Hz), 7.06 (d, 2H, J = 8.1 Hz), 7.30 (t, 1H, J = 7.2 Hz), 7.31 (t, 1H, J = 7.2 Hz), 7.40 (t, 2H, J = 7.2 Hz), 7.56 (m, 2H), 7.76 (d, 2H, J = 7.2 Hz); ^{13}C NMR (75 MHz, $CDCl_3$) δ 28.8, 37.9, 47.2, 54.3, 62.1, 66.8, 78.4, 120.0, 124.2, 125.0, 127.1, 127.7, 129.7, 130.7, 141.3, 143.7, 155.7, 154.5, 192.8. Anal. Calcd for $C_{29}H_{29}N_3O_4$: C, 72.03; H, 6.04; N, 8.69. Found: C, 72.12; H, 6.05; N, 8.66.

37. Obtained as a pale yellow solid (378.4 mg, 86% yield): TLC R_f 0.45 (eluant: chloroform/methanol 98:2); mp 126−128 °C; ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.22 (m, 2H), 4.17 (t, 1H, J = 7.1 Hz), 4.42 (d, 2H, J = 7.1 Hz), 4.56 (m, 1H), 5.05 (s broad, 1H), 5.47 (d, 1H, J = 8.7 Hz), 6.95 (s broad, 1H), 7.08−7.34 (m, 5H), 7.38 (t, 2H, J = 7.2 Hz), 7.57 (t, 2H, J = 7.2 Hz), 7.62 (m, 1H), 7.75 (d, 2H, J = 7.2 Hz), 8.16 (s broad, 1H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 28.3, 47.2, 54.5, 58.2, 66.8, 110.1, 111.3, 118.6, 119.9, 120.0, 122.3, 123.1, 125.0, 127.0, 127.4, 127.7, 136.1, 141.3, 143.7, 155.8, 193.6. Anal. Calcd for $C_{27}H_{22}N_4O_3$: C, 71.99; H, 4.92; N, 12.44. Found: C, 71.87; H, 4.94; N, 12.43.

38. Obtained as a viscous yellow oil (531.5 mg, 79% yield): TLC R_f 0.37 (eluant: ethyl acetate/petroleum ether 98:2); ^{1}H NMR (300 MHz, DMSO-d_6) δ 1.18−1.81 (m, 4H), 1.40 (s, 6H), 2.01 (s, 3H), 2.42 (s, 3H), 2.50 (s, 3H, DMSO signal partially overlapped), 2.96 (s, 2H), 3.03 (m, 2H), 4.22 (m, 1H), 4.30−4.47 (m, 3H), 4.50 (s, 1H), 5.98 (s broad, 1H), 6.42 (s broad, 1H), 6.75 (s broad, 1H), 7.34 (t, 2H, J = 7.2 Hz), 7.41 (t, 2H, J = 7.2 Hz), 7.60−7.81 (m, 3H), 7.88 (d, 2H, J = 7.2 Hz); ^{13}C NMR (125 MHz, DMSO-d_6) δ 12.6, 18.0, 19.2, 26.0, 28.6, 28.7, 42.9, 47.2, 53.1, 58.3, 65.7, 86.6, 116.6, 120.5, 124.7, 125.6, 127.4, 128.0, 131.8, 134.6, 137.6, 141.2, 141.8, 144.2, 156.5, 157.9, 195.4. Anal. Calcd for $C_{35}H_{40}N_6O_6S$: C, 62.48; H, 5.99; N, 12.49. Found: C, 62.62; H, 6.00; N, 12.52.

■ ASSOCIATED CONTENT

Supporting Information

^{1}H and ^{13}C NMR spectra for compounds **2**, **22**−**28** and **31**−**38**. Synthetic procedure, RP U-HPLC/MS, MALDI-MS and MS/MS analysis of *N*-Fmoc-L-Ala-L-Leu and *N*-Fmoc-L-Ala-D-Leu diazoketones. This material is available free of charge via the Internet at http://pubs.acs.org.

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Notes

The authors declare no competing financial interest.

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