Available online at www.sciencedirect.com
SCIENCE DIRECT®
Tetrahedron
Tetrahedron 60 (2004) 1731–1748

Inhibitors of biotin biosynthesis as potential herbicides

Ayelet Nudelman,[a] Dana Marcovici-Mizrahi,[b] Abraham Nudelman,[c,*] Dennis Flint[d] and Vernon Wittenbach[e]

[a]*QBI Enterprises Ltd., Weizmann Science Park, Ness Ziona 70400, Israel*
[b]*Department of Organic Chemistry, Israel Institute for Biological Research, PO Box 19, Ness Ziona 74100, Israel*
[c]*Department of Chemistry, Bar Ilan University, Ramat Gan 52900, Israel*
[d]*E. I. Du Pont de Nemours and Company, Central Research and Development, Experimental Station, Wilmington, DE 19880-0328, USA*
[e]*E. I. Du Pont de Nemours and Company, Stine-Haskell Research Center, Newark, DE 19714, USA*

Received 2 December 2003; accepted 18 December 2003

Abstract—Isosteric derivatives and analogues of the 7-keto-8-aminopelargonic acid (KAPA), 7,8-diaminopelargonic acid (DAPA) and desthiobiotin (DTB) vitamer intermediates involved in the biosynthetic pathway of biotin were prepared and evaluated as potential herbicides. The most active compound was desmethyl-KAPA which displayed a GR_{50} (concentration of the active compound that causes a 50% growth inhibition) value of 8 ppm, where values <50 ppm are considered herbicidal. Other KAPA analogs where the terminal Me group was replaced by bulkier substituents such as Et, *i*-Pr and $HOCH_2$ showed moderate activity.

1. Introduction

Biotin, a water soluble vitamin, functions as a coenzyme in carboxylation and transcarboxylation reactions critical to microorganisms, plants and animals.[1] The importance of biotin to metabolism is derived from its participation in many vital metabolic processes, such as gluconeogenesis, biosynthesis of fatty acids and metabolism of amino acids. Perhaps its most important role is in the carboxylation of acetyl-CoA to give malonyl-CoA, which is the first step in

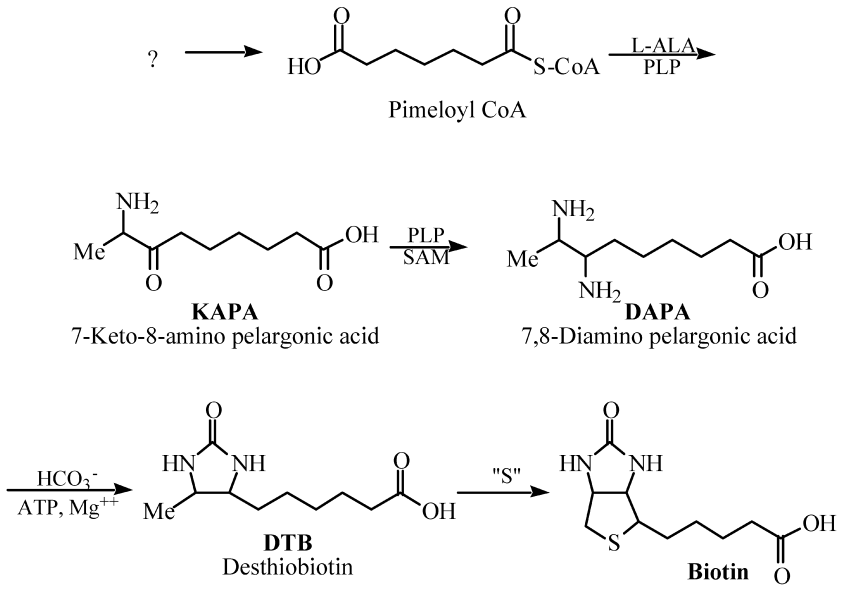
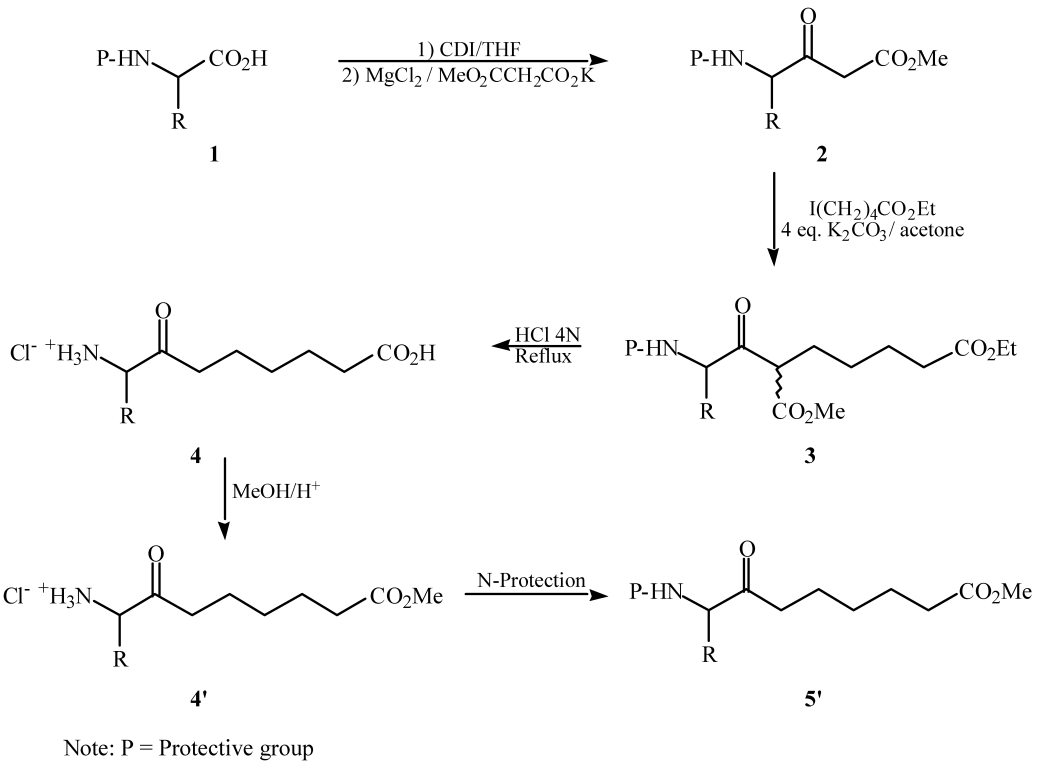


Figure 1. Biosynthetic pathway of biotin.

Keywords: Biotin; Biotin vitamers; 7-Keto-8-aminopelerginic acid (KAPA); 7,8-Diaminopelargonic acid (DAPA); Desthiobiotin (DTB); Herbicides; Enzyme inhibitors.
* Corresponding author. Tel.: +972-3-531-8314; fax: +972-3-535-1250; e-mail address: nudelman@mail.biu.ac.il

0040–4020/$ - see front matter © 2003 Elsevier Ltd. All rights reserved.
doi:10.1016/j.tet.2003.12.047

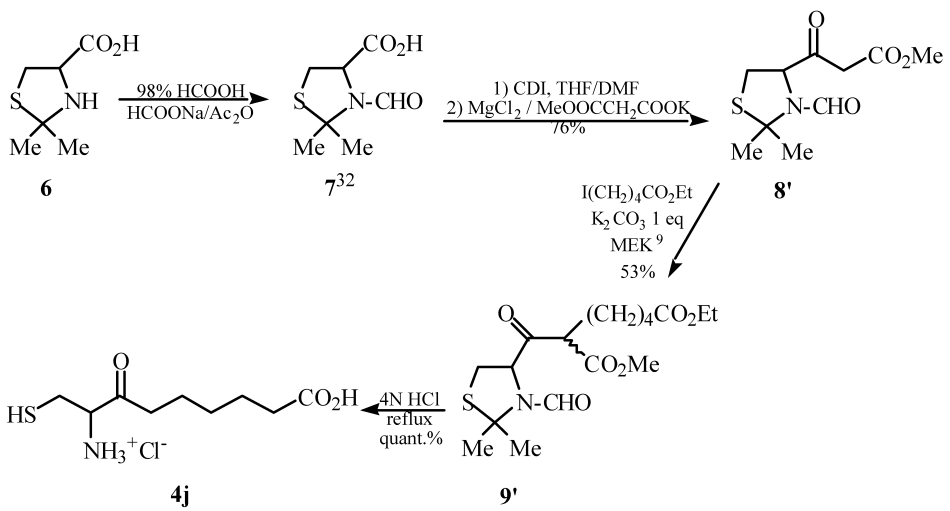


Scheme 1. General synthesis of KAPA analogs.

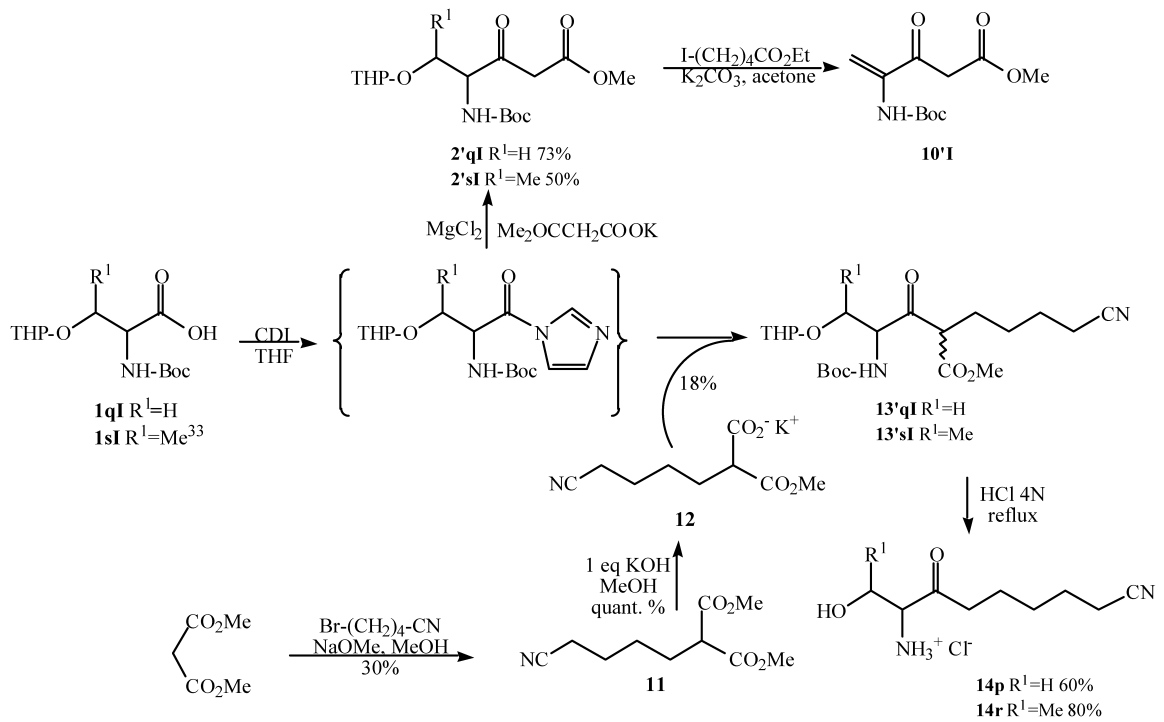
fatty acid biosynthesis. Since fatty-acid synthesis is essential for the growth and development of most organisms, biotin is thus an essential nutrient for plants and animals. Plants, microorganisms and some fungi biosynthesize their own biotin, while animals and man require trace amounts of the vitamin in their diet.

The biotin biosynthetic pathway[2] (Fig. 1), involving the KAPA, DAPA and DTB vitamers, is well understood in microorganisms[3] but has not been clearly elucidated in plants. However, there is increasing evidence that plants synthesize it by a similar route to that used by *E. coli*.[4] Recent studies indicate that the source of the sulfur atom, in the conversion of DTB to biotin, catalyzed by biotin synthase stems from an iron–sulfur cluster found in this enzyme.[5]

Inhibitors of enzymes involved in the biosynthesis of essential plant nutrients can cause irreparable damage to plants, and for this reason such enzymes can be useful targets for the rational design of inhibitors in the hopes of finding new herbicides. We describe the synthesis and biological evaluation of analogs of biotin and biotin vitamers (molecules, such as intermediates in the biotin biosynthetic pathway, that can be converted into biotin in vivo) as potential enzyme inhibitors with the objective of finding some that exhibit herbicidal activity. Moreover, since the enzymes involved in the biotin biosynthetic



Scheme 2. Synthesis of KAPA analogs derived from cysteine.



Scheme 3. Synthesis of KAPA analogs derived from THP-protected serine and threonine.

pathway are absent in higher organisms, it is expected that inhibitors of biotin biosynthesis will possess minimal toxicity in animals and man.

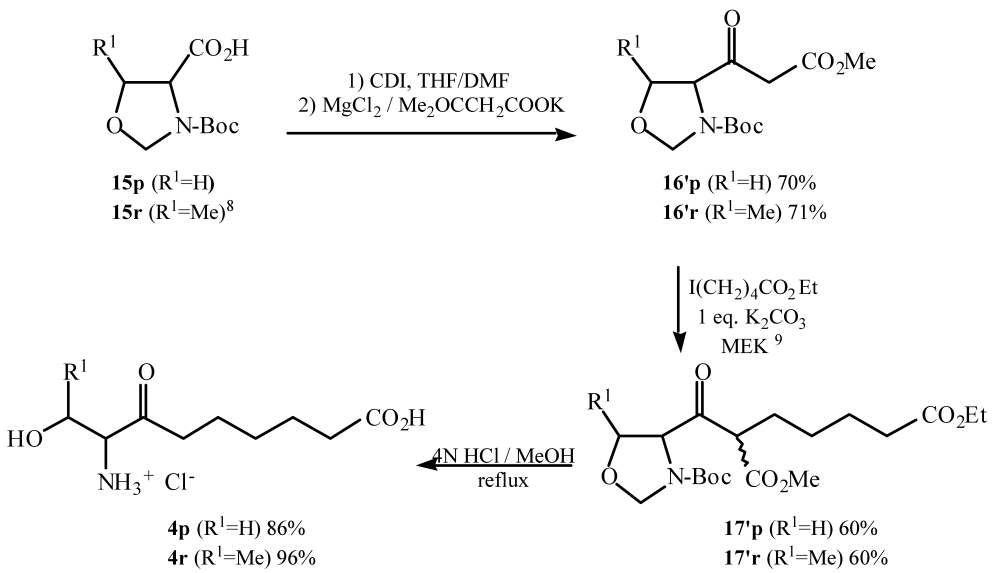
2. Results and discussion

Based on the structures of the vitamers of biotin, several families of analogs were developed, each featuring one or more isosteric modifications. The synthetic approaches to these derivatives were based to a large extent on our earlier synthesis of the vitamers.[6] The compounds described below are analog derivatives of KAPA, DAPA and DTB having chain lengths of nine carbon atoms, focusing primarily on compounds prepared from various amino acids, resulting in modifications of the terminal Me group.

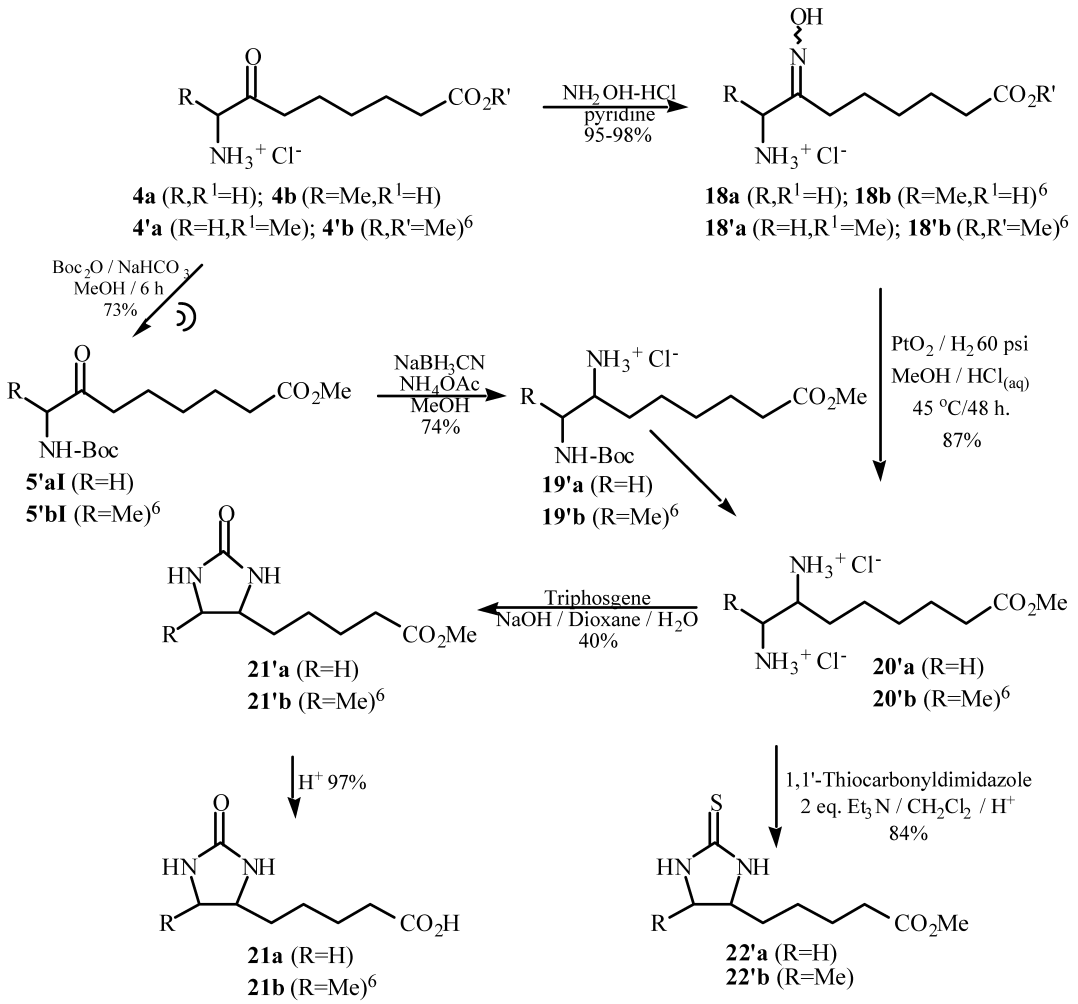
2.1. KAPA analogs

Substrates **1** (Scheme 1, Table 1) were derivatives of natural L-amino acids with the exclusion of the racemic *N*-Boc-α-aminobutyric acid (**1c** R=Et), *S*-benzyl-cysteine (**1k** R=$BnSCH_2$) and *S*-Boc-cysteine (**1k** R=$BocSCH_2$).

The syntheses of KAPA analogs derived from cysteine **4j**, serine **4p** and threonine **4r**, that required additional protection of the *S* and *O* atoms, are shown in Schemes 2–4. An initial attempt to prepare **4j** involved reaction of



Scheme 4. Synthesis of KAPA analogs derived from serine and threonine.



Scheme 5. Synthesis of DTB and thio-DTB derivatives.

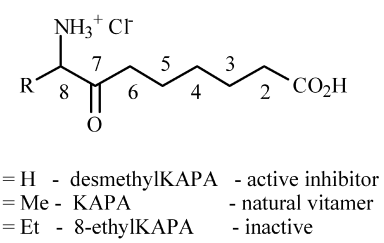
Bn-*S*-cysteine (Bn) to give the desired β-ketoester analog **2m**. However, due to the high nucleophilic character of the *S* atom, alkylation of **2m** with I–$(CH_2)_5$–COOEt took place preferentially at *S* rather than at *C* (Scheme 1). Other cysteine derivatives protected both at the *S* and the *N* with Boc (**1kI**), CBZ (**1lII**) or Ac (**1nIV**)[7] groups, gave poor yields of the respective derivatives **2**. The desired **4j** was finally obtained from thiazolidine **7** that possessed a sterically hindered sulfide group (Scheme 2).

Attempted alkylation on the respective β-ketoester derivatives of *N*-Boc-*O*-THP-protected serine **2′qI** or threonine **2′sI** led to elimination of THP-OH to give compound **10′I**, which was not isolated and was identified only by its characteristic vinylic peaks in the NMR spectrum. To prevent this base catalyzed elimination an alternative approach to **4p** and **4q** involved the coupling of the diprotected *N*-Boc-*O*-THP-serine and threonine with the nitrile derivative **12**. Although the desired cyano esters **13′qI** and **13′sI** were obtained, subsequent hydrolysis and decarboxylation gave KAPA-analogs **14q** and **14s** where the CN group remained intact (Scheme 3).

The desired **4p** and **4r** were finally obtained from the amino acids protected as their respective *N*-Boc oxazolidines **15p** and **15r**,[8] via the intermediate diesters **16′p** and **16′r** (Scheme 4). In contrast to compounds of structure **2**, which were alkylated in acetone in the presence of four equivalents of K_2CO_3, the alkylation of **16′p** and **16′r** readily proceeded with one equivalent of base in 2-butanone (MEK).

2.2. DAPA and DTB analogs

The synthesis of the neighboring diamino functionality found in the DAPA skeleton was approached via oximation of the α-amino-keto group of the KAPA analogs **4**.[6] Although the α-amino-oximes **18a** and **18b** were obtained as mixtures of *syn* and *anti*-isomers, one isomer predominated. This encouraged us to believe that the chiral center at C-8 could induce stereoselectivity upon oximation and would do so in the subsequent reductive step leading to



R = H - desmethylKAPA - active inhibitor
R = Me - KAPA - natural vitamer
R = Et - 8-ethylKAPA - inactive

Figure 2. SAR of KAPA derivatives.

Table 1. P and R substituent assignments

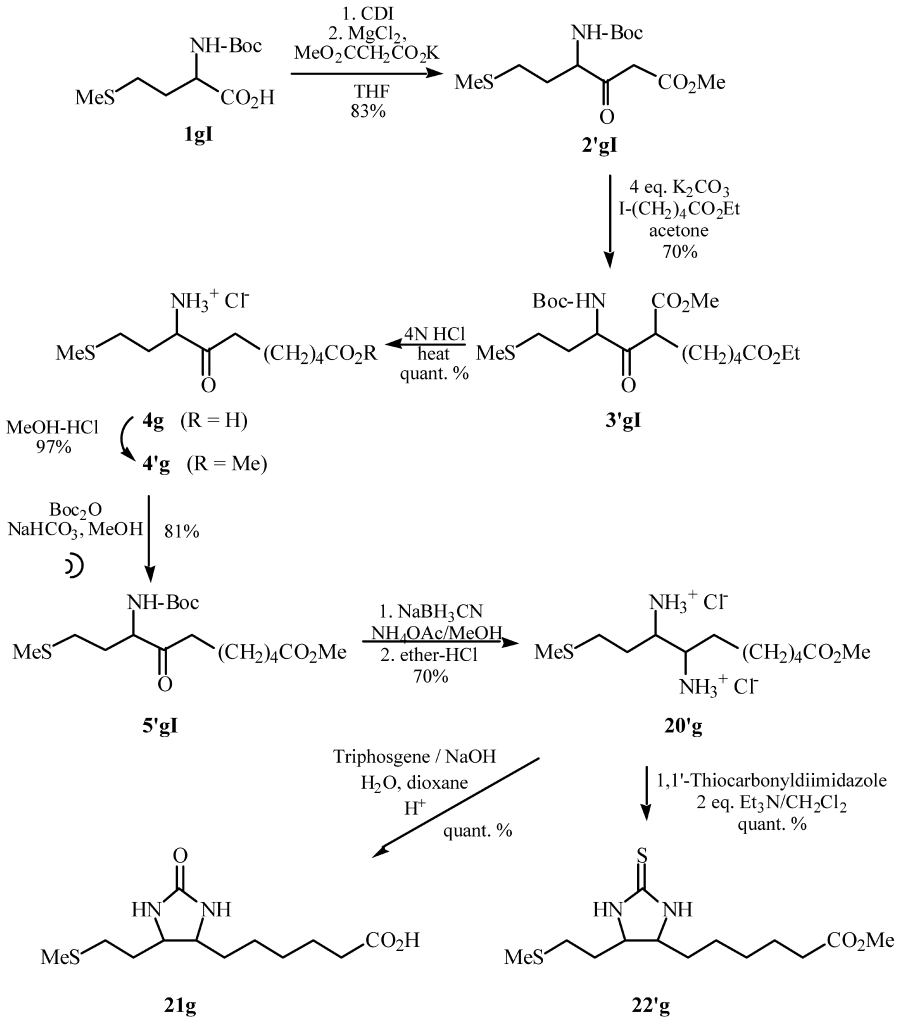
P *N*-Protective group	Code
Boc	I
CBZ	II
MeOCO	III
Ac	IV
R-substituent	Code
H-	a
Me–	b
Et–	c
i-Pr–	d
i-Bu–	e
sec-Bu–	f
$MeSCH_2CH_2$–	g
$MeS(O)CH_2CH_2$–	h
$MeSCH_2$–	i
$HSCH_2$–	j
Boc-SCH_2–	k
CBZ-SCH_2–	l
Bn-SCH_2–	m
Ac-SCH_2–	n
CH_2=CH–	o
$HOCH_2$–	p
THP-OCH_2–	q[28]
HOCH(Me)–	R
THP-OCH(Me)–	s[28]

Note: all Me esters are labeled as #′.

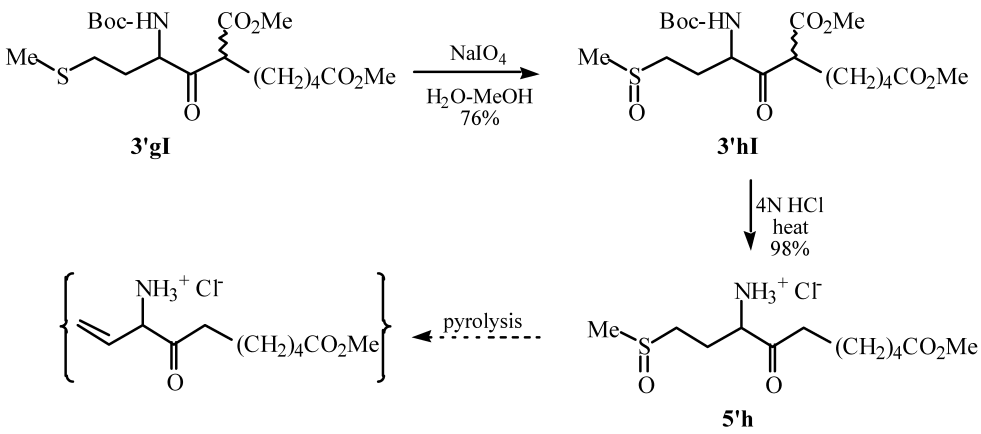
desired *cis*-diamino analogs. Alas, the diamines **19′a** and **19′b** were obtained as equimolar mixtures of two diastereomers. A similar loss of chiral discrimination was described earlier.[6] Based on biological data (see below), derivatives **20′a** and **20′b** were cyclized to the respective DTB analogs **21′a** and **21′b** (Scheme 5). Since *S*-containing KAPA-analogs **5′g–n** were unsuitable to catalytic hydrogenation due to catalyst poisoning, for these compounds an alternative route to the DAPA structure was developed involving reductive amination with $NaBH_3CN$. This procedure was also convenient for the threonine analog **4rI**. Since standard *N*-Boc protection of amines involves basic conditions, to avoid possible decomposition of free amino ketones obtained upon acid neutralization of derivatives **4′**, mild basic conditions coupled with sonnication were found to be satisfactory for the preparation of the *N*-Boc derivatives **5′aI** and **5′bI**. These compounds were amenable to subsequent $NaBH_3CN$ reduction in organic media. The DAPA analogs **21a** and **21b** were found to be racemic at the C-7 centers (Scheme 5).[6]

2.3. Vinyl derivatives

The natural vitamer, KAPA **4b**, possesses a terminal R=Me group. In these studies, the first analog found to possess



Scheme 6. DTB and thio-DTB analogs derived from methionine.



Scheme 7. Attempted synthesis of vinyl-KAPA.

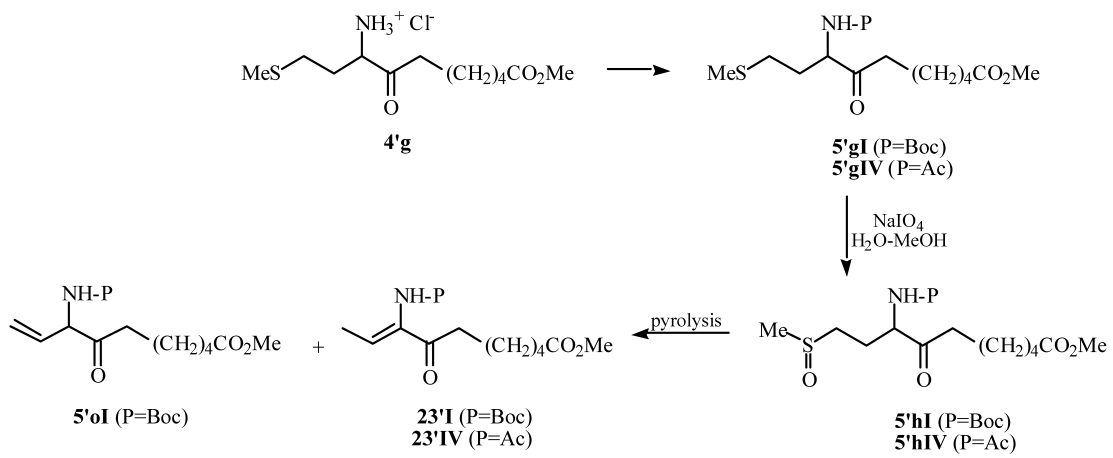
inhibitory effects on plant growth was the analog **4a** where R=H. However when an additional carbon was added, **4c** R=Et, no inhibition was detected. Being that a vinyl group is larger than a Me group, yet smaller than an Et group, introduction of a vinyl substituent at position 8 was expected to provide additional information on the steric demands for molecular fit at this part of the molecule (Fig. 2).

Given that L-alanine was the substrate for KAPA, and glycine was used in the preparation of desmethyl-KAPA,[9] L-vinylglycine,[10] **1o**, was the preferred starting material for the synthesis of the 9-vinyl derivative **4o**. In addition to its enzyme-inhibitory and antibiotic properties L-vinylglycine has become an important chiral starting material for a variety of other amino acid syntheses and optically active products. Since the attempted direct formation of the β-keto ester of vinylglycine, analog of compound **2′o**, failed, apparently due to the acidity of the methine proton of vinylglycine, the β-keto ester of methionine **2′gI** was prepared from *N*-Boc-methionine **1gI** by Mansour's method.[11] By analogous methodology to that described in Scheme 1, **2′gI** was further converted into KAPA, DAPA, DTB and thio-DTB analogs **4g**, **20′g**, **21′g** and **22′g**, respectively (Scheme 6). The lack of herbicidal activity of these derivatives may be attributed to the presence of the $MeSCH_2$-substituent on C-8, being probably too big to fit into the enzymatic 'cavity' suitable for a Me group.

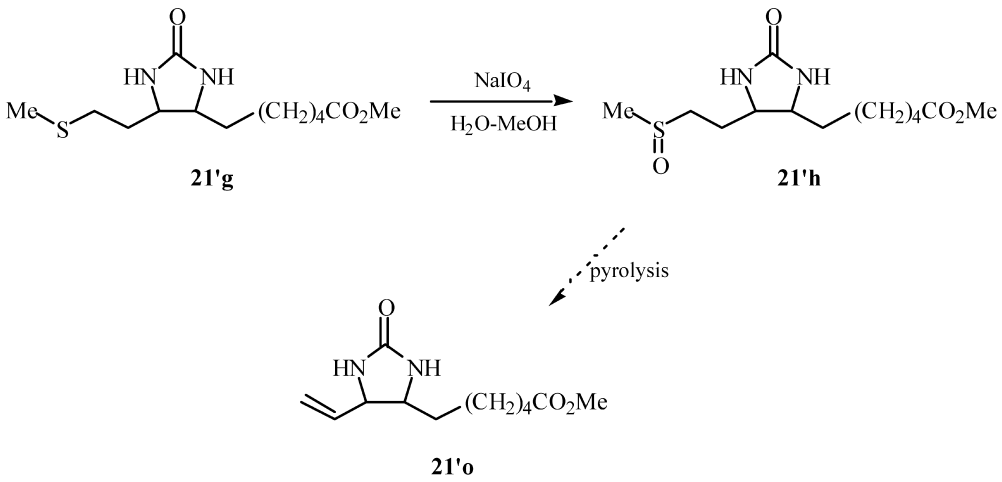
Periodate oxidation of **3′gI** to sulfoxide **3′hI**[12] followed by removal of the *N*-Boc protective group gave the KAPA analog **5′h**. Because the attempted pyrolysis of **5′h** to the corresponding vinylic KAPA derivative failed (Scheme 7), an alternative approach to the vinylic derivative was developed. Reaction of **4′g** with di-*tert*-butyl dicarbonate or acetylimidazole yielded sulfides **5′gI** and **5′gIV**, that were oxidized to sulfoxides **5′hI** and **5′hIV**, respectively. Whereas pyrolysis of **5′hI** afforded a mixture of **5′oI** and **23′I**, the *N*-acetylated **5′hIV** due to the higher acidity of the methine hydrogen α to the amido group, gave only the corresponding **23′IV** (Scheme 8). Although in the former case some of the desired **5′oI** was isolated, because of concomitant loss of the Boc group under the pyrolysis conditions, the overall yield of the mixture of olefins was very low.

In an attempt to avoid the isomerization of the double bond in **5′oI** leading to **23′oI**, attributed to the acidity of the proton α to the carbonyl group, sulfoxide **21′h**, obtained by periodate oxidation of **21′g**, was pyrolyzed, however the reaction was unsuccessful (Scheme 9).

To stay away from ketonic groups having acidic α-protons, DAPA derivatives protected with groups other than *N*-Boc, were examined. Sulfoxide **20′hII** was obtained in excellent yield when **20′gII** suspended in MeOH was dissolved in CH_2Cl_2, and was oxidized with $NaIO_4$ in a mixture of



Scheme 8. Formation of isomeric vinylic derivatives of KAPA.



Scheme 9. Attempted pyrolysis of a DTB-sulfoxide derivative.

MeOH and H_2O. In the course of pyrolysis of **20′hII** partial loss of the CBZ groups was observed, giving a mixture of benzyl alcohol and imidazolones **21′o** and **24′o**. The *N* on to which the CBZ was attached in **24′o** was not determined. In both products only the desired terminal vinyl substituent was found, without a trace of the undesired isomer (Scheme 10).

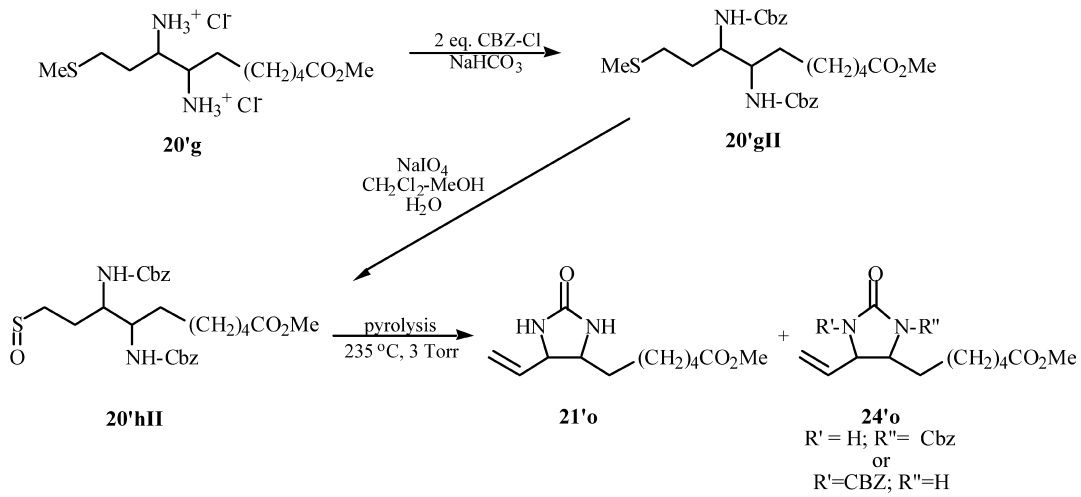
To avoid the cyclization observed in the course of the pyrolysis, the amino groups where protected as methyl carbamates. Although carbamates are usually prepared using methyl chloroformate and NaOH,[13] the bis-carbamate derivative **20′gIII** was prepared in excellent yield using dimethyl dicarbonate, by an analogous procedure to that used with di-*t*-butyl dicarbonate. Oxidation of **20′gIII** gave the sulfoxide **20′hIII**, which was pyrolyzed to give **20′oIII** as a mixture of two diastereomers. Whereas acidic hydrolysis of **20′oIII** gave a mixture of vinylic isomers **20o** and **25o** in a 9:1 ratio, basic hydrolysis led to the mono-carbamoylated cyclic urea isomers **21o** and **26o** where the location of the *N*-carbamate was not established (Scheme 11).

2.4. Biological results

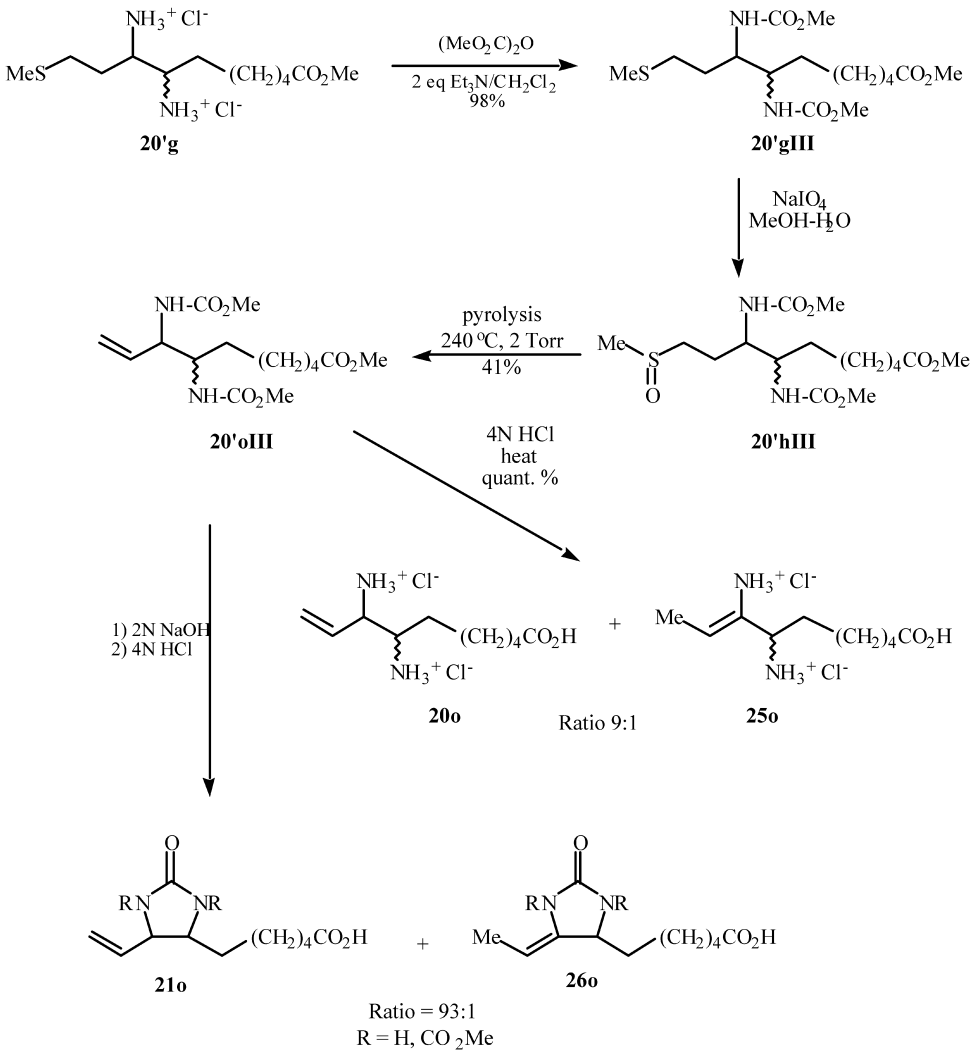
Enzyme assays on compounds described in this paper were not conducted. Instead, herbicidal activity was measured at difference concentrations of the compounds and the results were recorded as GR_{50} values. The term of GR_{50} refers to the concentration of the active compound that causes a 50% growth inhibition. In our tests commercial herbicides gave GR_{50} values of <50 ppm, so compounds with activity in this range were considered herbicidal. When compounds described in this paper exhibited herbicidal activity, reversal tests were carried out by treating the plants with biotin. If the herbicidal activity of a compound was reversed by biotin, it was deemed likely that the compound was active because it disrupted biotin biosynthesis.

Initial herbicidal activity tests were carried out on *Arabidopsis*. Compounds active against *Arabidopsis* where tested further tested on *Lemna*, Back Mexican Sweet Corn Cells (BMS), and Barnyard grass.

In conclusion, the growth inhibitory activities (GR_{50}) of a selected number of compounds are listed in Table 2. The most active compound found was desmethyl-KAPA **4a**. Other KAPA analogs where the terminal Me group was replaced by bulkier substituents such as Et, *i*-Pr and $HOCH_2$ showed moderate activity, whereas the thio-derivatives ($HSCH_2$, $MeSCH_2$ and $Me_2CH_2CH_2$) as well as that derived



Scheme 10. Pyrolysis of a di-Cbz-DAPA sulfoxide derivative.



Scheme 11. Attempted formation of 9-vinyl-DTB.

from threonine (MeCHOH) were considerably weaker inhibitors. DAPA and DTB analogs replacing the Me by H or Et also showed moderate activity. When comparing free acids to their respective more lipophilic Me esters, the latter where found to be more active. Subsequent reports will describe the inhibitory activity of isosters involving additional structural modifications based on the natural vitamers.

3. Experimental

3.1. Biology—methods and materials

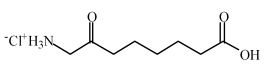
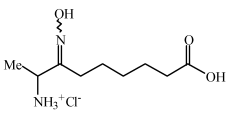
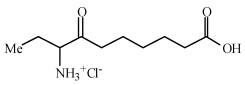
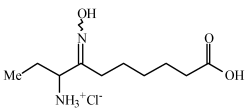
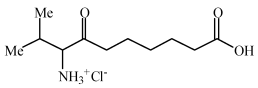
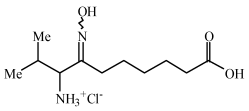
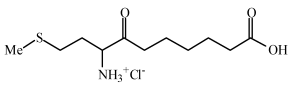
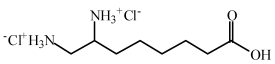
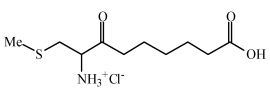
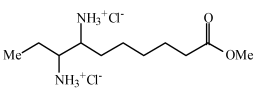
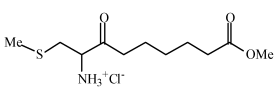
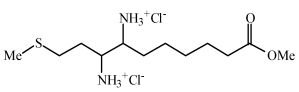
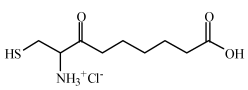
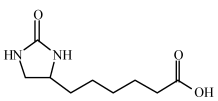
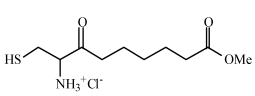
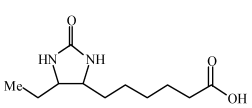
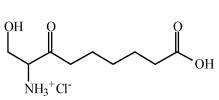
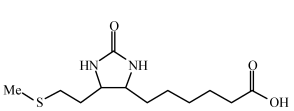
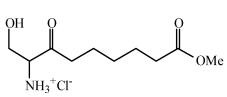
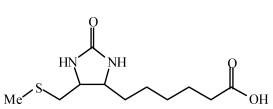
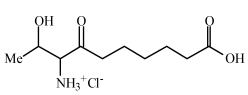
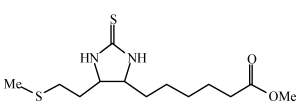
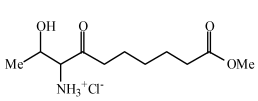
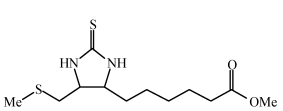
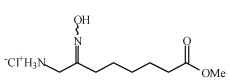
3.1.1. *Arabidopsis* test. *Arabidopsis thaliana* was grown in 12-well titer plates under sterile conditions on 0.8% agar medium containing nutrient solution as described by Somerville and Ogren.[14] Concentrated treatment solutions were filter sterilized and were added to the agar solution while it was still in a liquid state (45 °C). Three mL aliquots of the agar solution were pipetted into the wells. Concentrated treatment solutions were filter sterilized and were added to the agar solution while it was still in a liquid state (45 °C). When the agar had solidified and cooled, seeds were sown on the surface. The plates were placed in a 26 °C incubator under continuous fluorescent lighting with an intensity of 55 $\mu E\ m^{-2}\ s^{-1}$. The plants were rated after three weeks against the controls for growth inhibition due to the inhibitor treatment. These ratings were used to determine the GR_{50} value, which is the concentration that resulted in a 50% growth reduction.

To obtain more tissue for biotin determinations, seven-day old *Arabidopsis* seedlings grown on media without the inhibitor were watered over the foliage with different concentrations of the tested inhibitor in 8 mM phosphate buffer, at a pH 6.5 value. Plants were harvested at 0, 7 and 14 days after treatment and were weighed and extracted for total biotin determination.

3.2. Chemistry—general

^1H NMR spectra 200 and 300-MHz were obtained on Bruker AC-200 and AM-300 spectrometers, respectively. Chemical shifts are expressed in ppm downfield from Me_4Si used as internal standard. When D_2O was used as solvent, its own peak was used as internal standard for ^1H NMR, while additional MeOH was used as an internal standard for ^{13}C NMR. The values are given in δ scale. Mass spectra were obtained on a Varian Mat 731 spectrometer (CI=chemical

Table 2. Growth inhibitory activity (GR_{50}) of selected isosters of the biotin vitamers

Compounds	Structure	GR_{50} (ppm)	Compounds	Structure	GR_{50} (ppm)
4a[29]		8	**18b**[6]		40
4c[30]		43	**18c**		>780
4d		37	**18d**		>780
4g		>780	**20a**[31]		53
4i		>780	**20′c**		76
4′i		250	**20′g**		>780
4j		>780	**21a**[26]		35
4′j		120	**21c**[27]		35
4p		100	**21g**		>780
4′p		53	**21i**		46
4r[20]		250	**22′g**		>780
4′r[20]		100	**22′i**		44
18a		53			

ionization). HRMS were obtained on a VG AutoSpec E spectrometer. Progress of the reactions was monitored by TLC on silica gel (Merck, Art. 5554) or alumina (Riedel-de Haen, Art. 37349). Flash chromatography was carried out on silica gel (Merck, Art. 9385). Commercially available chemicals were used without further purification.

3.3. *N*-Boc protection of amino acids 1

Method A.[15]To a stirred solution of an amino acid (0.03 mol) and NaOH (0.03 mol) in water (4 mL) and *tert*-BuOH (6 mL) was added di-*tert*-butyl dicarbonate (0.03 mol). The mixture was stirred overnight at room

temperature, water (15 mL) was then added and the aqueous phase was washed with hexane (3×50 mL). The aqueous phase was acidified with $KHSO_4$ (to pH=2.5), extracted with EtOAc (4×50 mL). The combined organic layers were dried ($MgSO_4$), filtered and evaporated to afford the desired *N*-Boc protected amino acid.

Method B.[16] A suspension of a KAPA.HCl methyl ester derivative (1 mmol), $NaHCO_3$ (3 mmol), and di-*tert*-butyl dicarbonate (1 mmol) in dry MeOH (10 mL), was sonicated at room temperature for 6 h. The resulting mixture was filtered and evaporated to dryness. The residue was dissolved in ether (20 mL) and 1 N HCl (20 mL). The aqueous layer was washed with ether (3×15 mL). The combined organic layer was washed with 5% $NaHCO_3$ (2×20 mL), and brine (2×20 mL). The organic phase was dried ($MgSO_4$), filtered and evaporated to give the desired *N*-Boc product.

Method C.[17] To a solution of L-cystine (4 mmol), NaOH (8 mmol) in 15 mL water at 0 °C, was slowly added DMF (15 mL). The mixture was brought to room temperature and di-*tert*-butyl dicarbonate (8 mmol) was added in one portion. The pH was maintained at ~9 by adding NaOH for 3 h and stirring at room temperature was continued overnight. To the cloudy mixture, water (50 mL) was added and was washed with EtOAc (2×50 mL). To the aqueous solution obtained, EtOAc was added (50 mL). The mixture was cooled to 0 °C and HCl 1 N was added until pH 3 was obtained. The organic phase was separated and washed with water (6×20 mL). The organic phase was dried ($MgSO_4$), filtered and evaporated to give the desired product.

Method D. A mixture of the amino acid ester derivative (1 mmol) and Et_3N (1 mmol) in dry CH_2Cl_2 (10 mL) was stirred at room temperature for 10 min. Di-*tert*-butyl dicarbonate (1 mmol) was added and the reaction mixture was stirred at room temperature for 18 h. The resulting mixture was washed with water (3×25 mL). The organic layer was dried ($MgSO_4$), filtered and evaporated to dryness, to afford the desired product.

3.4. 2-(3-Formyl-2,2-dimethyl-thiazolidine-4-carbonyl)-heptanedioic acid 7-ethyl ester 1-methyl ester (9′)

C-Alkylation of **8′** with ethyl 5-iodovalerate (53% yield). Note: due to large amounts of O-alkylation products, only one equivalent of K_2CO_3 in 2-butanone (bp 73 °C) was used. ^{1}H NMR ($CDCl_3$) δ 1.23 (t, J=7 Hz, 3H, CH_2Me), 1.38 (m, 2H, $CH_2CH_2CH_2CO_2$), 1.63 (superimposed, 5H, *Me*C and $CH_2(CH_2)_3CO_2$), 1.81 (superimposed, 5H, *Me*C and $CH_2CH_2CO_2$), 2.30 (t, J=7.5 Hz, 2H, CH_2CO_2), 3.23 (ABq of d, J_{AB}=13.8 Hz, J_{AX}=5 Hz, J_{BX}=4.8 Hz, 2H, SCH_2), 3.74 and 3.75 (two s, 3H, OMe), 3.75 and 3.93 (two t, J=7 Hz, 1H, $COCHCO_2$), 4.12 (q, J=7 Hz, 2H, CH_2Me), 4.92 (td, J=7, 1 Hz, 1H, CHNH), 8.20 (d, J=1 Hz, 1H, COH).

3.5. General procedure for the preparation of methyl esters, using a diazomethane derivative, trimethylsilyl diazomethane[18]

A 2 M solution of TMS-diazomethane in hexane (0.65 mL, 1.3 mmol) was added dropwise to a solution of an *N*-protected amino acid (1 mmol) in hexane (5 mL) and anhydrous MeOH (2 mL). The mixture was stirred at room temperature overnight, and became cloudy. The solvent was evaporated and the residue was dissolved in $CHCl_3$ and was washed with 5% $NaHCO_3$. The aqueous layer was extracted with $CHCl_3$ (2×) and the combined organic layers were dried ($MgSO_4$), filtered and evaporated, to give the esters. Although it was not required to wash the product with 5% $NaHCO_3$, this was found convenient to obtain the product without a trace of the starting acid.

3.6. Conversion of N-protected amino acid derivatives to the corresponding β-keto esters

To a solution of an N-protected amino acid (1 mmol) in dry THF (10 mL) under N_2, CDI (1.2 mmol) was added portion wise. The mixture was stirred at room temperature for 1 h, then $MgCl_2$ (1 mmol) and monomethyl malonate K salt (1 mmol) were added at once. The mixture was stirred at 35 °C overnight. The resulting slurry was filtered and the filtrate was evaporated. The residue was dissolved in EtOAc (20 mL) and 1 N HCl (20 mL). The aqueous phase was extracted with EtOAc (3×20 mL). The organic layers were washed with 5% $NaHCO_3$ (2×20 mL), brine (20 mL). The organic layer was dried ($MgSO_4$) and evaporated to give the desired product.

3.6.1. 4-*tert*-Butoxycarbonylamino-3-oxo-hexanoic acid methyl ester (2′cI). From *N*-Boc-*dl*-2-aminobutyric acid, **1′cI** (60% yield). ^{1}H NMR ($CDCl_3$) δ 0.93 (d, J=7.5 Hz, 3H, CH_2Me), 1.45 (s, 9H, Me_3C), 1.61 (m, 1H, CH_2Me), 1.95 (m, 1H, CH_2Me), 3.57 (ABq, J=15 Hz, 2H, CH_2CO), 3.75 (s, 3H, OMe), 4.32 (m, 1H, CHNH), 5.16 (m, 1H, NH); ^{13}C NMR ($CDCl_3$) δ 9.5 (CH_2Me), 24.1 (CH_2Me), 28.3 (Me_3C), 46.1 (CH_2CO), 52.4 (OMe), 60.8 (CHNH), 80.1 (C), 155.4 ($HNCO_2$), 167.2 (CO_2Me), 201.9 (CO); MS (EI) *m/e* 260 (MH^+, 34), 204 ($MH^+-C_4H_8$, 100), 186 (MH^+-t-BuOH, 17), 160 (MH^+-Boc, 68); HRMS (DCI, CH_4) calcd for $C_{12}H_{22}NO_5$ (MH^+) 260.1497 found 260.1420.

3.6.2. 3-(3-Formyl-2,2-dimethyl-thiazolidin-4-yl)-3-oxo-propionic acid methyl ester (8′). From 2,2-dimethyl-thiazolidine-4-carboxylic acid hydrochloride, **7** (76% yield). Note: dry DMF (ca. 0.5 mL) was added to the mixture, to allow better solubility of the starting acid. The product was obtained as a single isomer, derived from the major rotamer of the starting material. ^{1}H NMR ($CDCl_3$) δ 1.84 (s, 6H, Me_2C), 3.30 (ABq of d, J_{AB}=12.2 Hz, J_{AX}=7 Hz, J_{BX}=6 Hz, 2H, CH_2S), 3.70 (ABq, J_{AB}=16.1 Hz, 2H, CH_2CO), 3.75 (s, 3H, OMe), 5.04 (ddd, J=7, 6, 1 Hz, 1H, CH_2CH), 8.29 (d, J=1 Hz, 1H, CHO). ^{13}C NMR ($CDCl_3$) δ 30.1 (CH_2S), 30.9 (*Me*C), 31.5 (*Me*C), 46.9 (CH_2CO), 52.3 (OMe), 68.1 ($CHCH_2$), 70.3 (Me_2C), 158.90 (CON), 167.3 (CO_2), 198.6 (COCH). MS (CI/NH_3) *m/e* 263 (MNH_4^+, 100), 246 (MH^+, 37).

3.7. C-Alkylation of β-keto esters using K_2CO_3[6]

To a solution of a β-keto ester derivative (1 mmol) in dry acetone (20 mL) and anhydrous K_2CO_3 (4 mmol) or dry 2-butanone (MEK) (20 mL) and K_2CO_3 (1 mmol), was added an alkyl iodide (1 mmol). The resulting suspension

was refluxed under nitrogen for 6–18 h, cooled and filtered. The salts were washed with acetone. The filtrate was evaporated and the residue was flash chromatographed on a silica gel column (hexane/EtOAc 2:1 or 3:1), to give the desired C-alkylated product, containing mixtures of diastereomers.

3.7.1. 2-*tert*-Butoxycarbonylaminoacetyl-heptanedioic acid 7-ethyl ester 1-methyl ester (3′aI). From **2′aI** (55% yield). ^{1}H NMR ($CDCl_3$) δ 1.25 (t, J=7 Hz, 3H, OCH_2Me), 1.45 (s, 9H, Me_3C), 1.50–1.78 (m, 4H, $CHCH_2CH_2CH_2$), 1.90 (m, 2H, $CHCH_2CH_2CH_2$), 2.30 (t, J=7 Hz, 2H, CH_2CO_2Et), 3.50 (t, J=7 Hz, 1H, $CHCO_2Me$), 3.73 (s, 3H, OMe), 4.15 (m, superimposed, 4H, CH_2NH, OCH_2Me), 5.20 (m, 1H, NH); ^{13}C NMR ($CDCl_3$) δ 14.2 (OCH_2Me), 24.4 ($CH_2CH_2CO_2$), 25.1 ($CHCH_2$), 26.8 ($CHCH_2CH_2$), 28.3 (Me_3C), 33.9 (CH_2CO_2), 50.1 (CH_2NH), 52.6 (OMe), 55.9 ($CHCO_2Me$), 60.3 (OCH_2Me), 80.0 (C), 155.5 ($HNCO_2$), 169.4 (CO_2Me), 173.2 (CO_2Et), 200.9 (CO); MS (CI, *i*-Bu) *m/e* 360 (MH^+, 1), 304 ($MH^+-C_4H_8$, 13), 256 (MH^+−Boc, 100); HRMS (DCI, CH_4) calcd for $C_{17}H_{30}NO_7$ (MH^+) 360.2022 found 360.1995.

3.8. Preparation of KAPA·HCl derivatives (hydrolysis and decarboxylation of compounds 3)[6]

A suspension of a C-alkylated ester **3** (1 mmol) in 4 N HCl (2 mL) was refluxed for 2 h. Gas evolution was observed. The resulting dark yellow solution was evaporated under high vacuum. If the color of the resulting product darkened, the residue was dissolved in distilled water (minimum amount) and was treated with charcoal, filtered and the filtrate was evaporated under high vacuum to give a solid residue. Recrystallization was carried out from EtOH–ether, or ether–HCl, to give the desired HCl salt.

3.8.1. 8-Amino-7-oxo-octanoic acid hydrochloride (4a).[19] Hydrolysis and decarboxylation of **3′aI**, isolated as a white solid mp 129–131 °C (84% yield). ^{1}H NMR (D_2O) δ 1.40 (m, 2H, $CH_2CH_2CH_2CO_2$), 1.70 (m, 4H, $CH_2CH_2CH_2CH_2CO_2$), 2.40 (t, J=7 Hz, 2H, CH_2CO_2), 2.70 (t, J=7 Hz, 2H, CH_2CO), 4.18 (s, 2H, CH_2N); ^{13}C NMR (D_2O) δ 22.9 ($CH_2CH_2CO_2$), 24.6 (CH_2CH_2COCH), 28.2 ($CH_2CH_2CH_2CO$), 34.4 (CH_2CO_2), 39.9 (CH_2COCH), 47.7 (CH_2N), 180.0 (CO_2), 206.9 (CO); MS (CI, NH_3) m/e 174 (MH+, 100); HRMS (DCI, CH_4) calcd for $C_8H_{16}NO_3$ (MH^+) 174.1130 found 174.1140.

3.8.2. 8-Amino-9-methyl-7-oxo-decanoic acid hydrochloride (4d). From **3′dI** (95% yield). ^{1}H NMR (D_2O) δ 0.86 (d, J=7 Hz, 3H, Me), 1.11 (d, J=7 Hz, 3H, Me), 1.33 (m, 2H, $CH_2(CH_2)_2CO_2$), 1.60 (m, 4H, $CH_2CH_2CH_2CH_2CO_2$), 2.37 (t, J=7.5 Hz, 2H, CH_2CO_2), 2.55 (m, 2H, $CHMe_2$), 2.68 (m, 2H, superimposed, CH_2CO), 4.24 (d, J=3.5 Hz, 1H, CHN); ^{13}C NMR (D_2O) δ 15.8 (Me), 19.0 (Me), 22.8 ($CH_2(CH_2)_3CO_2$), 24.6 ($CH_2CH_2CO_2$), 28.6 ($CHMe_2$), 34.4 (CH_2CO_2), 39.7 (CH_2CO), 64.7 ($CHCO$), 177.7 (CO_2), 209.6 (CO); MS (DCI, NH_3) *m/e* 216 (MH^+, 100); HRMS (DCI, CH_4) calcd for $C_{13}H_{26}NO_3$ (MH^+) 244.1912 found 244.1918.

3.8.3. 8-Amino-9-hydroxy-7-oxo-decanoic acid hydrochloride (4r). Hydrolysis and decarboxylation of **17′b** (96% yield). ^{1}H NMR (D_2O) δ 1.32 (m and d, superimposed, J=6.6 Hz, 5H, Me, $CH_2CH_2CH_2CO_2$), 1.49–1.66 (m, 4H, $CH_2CH_2CH_2CH_2CO_2$), 2.34 (t, J=7.3 Hz, 2H, CH_2CO_2), 2.70 (dt, J=2.5, 7.2 Hz, 2H, CH_2CO), 4.22 (d, J=2.5 Hz, 1H, CHN), 4.60 (dq, J=2.5, 6.6 Hz, 1H, $CHMe$); ^{13}C NMR (D_2O) δ 19.68 (Me), 22.79 ($CH_2CH_2CO_2$), 24.43 (CH_2CH_2CO), 28.06 ($CH_2(CH_2)_2CO_2$), 34.06 (CH_2CO_2), 39.33 (CH_2CO), 64.44 ($CHMe$), 64.89 (CHN), 179.18 (CO_2), 207.53 (CO); MS (CI, NH_3) *m/e* 218 (MH^+, 18), 200 (MH^+-H_2O, 12), 174 (MH^+-CO_2, 15); HRMS (DCI, CH_4) calcd for $C_{10}H_{20}NO_4$ (MH^+) 218.1392 found 218.1390.

3.8.4. 8-Amino-9-hydroxy-7-oxo-nonanenitrile hydrochloride (14p). From **13′qI**. The reaction mixture was washed with EtOAc (2×10 mL) and then the aqueous solution was evaporated to afford the product (60% yield). ^{1}H NMR (D_2O) δ 1.40 (m, 2H, $CH_2(CH_2)_2CN$), 1.61 (m, 4H, $CH_2CH_2CH_2CH_2CN$), 2.44 (t, J=7 Hz, 2H, CH_2CN), 2.70 (t, J=7 Hz, 2H, CH_2CO), 4.05 (dd, J=3.4, 12.8 Hz, 1H, CH_2OH), 4.17 (dd, J=4, 12.8 Hz, 1H, CH_2OH), 4.35 (t, J=3.7 Hz, 1H, $CHCH_2$); ^{13}C NMR (D_2O) δ 16.8 (CH_2CN), 22.3 (CH_2CH_2CN), 24.7 (CH_2CH_2CO), 27.8 ($CH_2(CH_2)_2CN$), 38.9 (CH_2CO), 59.4 (CH_2OH), 61.3 (CHN), 122.7 (CN), 207.1 (CO).

3.9. Formation of methyl esters of amino acids 4

Method A. Concentrated HCl (1.12 mL) was added to a solution of a KAPA derivative (1 mmol) in 2,2-dimethoxypropane (15 mL). The mixture was stirred at room temperature overnight. The solvent was evaporated and the residue was dissolved in a small amount of MeOH. Addition of dry ether resulted in crystallization. The crystals were filtered and washed with ether to give hydrochloride salt.

Method B. A solution of a KAPA·HCl derivative (1 mmol) in MeOH·HCl (8 mL) was stirred at room temperature overnight. Evaporation under vacuum gave the hydrochloride salt.

Method C. To a solution of a KAPA derivative (1 mmol) in dry MeOH (10 mL) at 0 °C was dropwise added AcCl (3 mL).[20] The resulting mixture was stirred at room temperature overnight. Evaporation yielded the hydrochloride salt.

3.9.1. 8-Amino-7-oxo-octanoic acid methyl ester hydrochloride (5′a). Esterification of DMK·HCl **4a** (quantitative yield). ^{1}H NMR (D_2O) δ 1.30 (m, 2H, $CH_2CH_2CH_2CO_2$), 1.60 (m, 4H, $CH_2CH_2CH_2CH_2CO_2$), 2.40 (t, J=7.3 Hz, 2H, CH_2CO_2), 2.60 (t, J=7.4 Hz, 2H, CH_2CO), 3.70 (s, 3H, OMe), 4.05 (s, 2H, CH_2N); ^{13}C NMR (D_2O) δ 22.8 ($CH_2CH_2CO_2$), 24.5 (CH_2CH_2CO), 28.2 ($CH_2CH_2CH_2CO_2$), 34.0 (CH_2CO_2), 39.9 (CH_2CO), 52.7 (OMe), 58.1 (CH_2N), 178.0 (CO_2Me), 209.8 (CO); MS (CI, *i*-Bu) *m/e* 187 (MH^+−HCl); HRMS (DCI, CH_4) calcd for $C_9H_{18}NO_3$ (MH^+) 188.1286 found 188.1260.

3.9.2. 8-Acetylamino-10-methylsulfanyl-7-oxo-decanoic acid methyl ester (5′gIV). N-Acetylation of **4′g**—CDI (0.22 g, 1.3 mmol) was added to a solution of AcOH (0.08 g, 1.3 mmol) in dry THF (10 mL) under N_2. The

mixture was stirred at room temperature for 1 h and was transferred via a cannula to a flask containing **4′g** (0.35 g, 1.16 mmol). The mixture was stirred at room temperature under N_2 overnight and was evaporated. The residue was partitioned between 1 N HCl and EtOAc and the aqueous layer was extracted with EtOAc (4×). The combined organic layers were washed with 5% $NaHCO_3$ (2×), brine, dried ($MgSO_4$), filtered and evaporated to give **5′gIV** as an oil, 57% yield). ^{1}H NMR ($CDCl_3$) δ1.34 (m, 2H, $CH_2(CH_2)_2CO_2$), 1.64 (m, 4H, $CH_2(CH_2)_3CO_2$), 2.04 (s, 3H, MeCO), 2.10 (s, 3H, SMe), 2.27 (m, 2H, SCH_2CH_2), 2.32 (t, *J*=7.4 Hz, 2H, CH_2CO_2), 2.50 (superimposed, m, 4H, CH_2S and $CHCOCH_2$), 3.67 (s, 3H, OMe), 4.74 (td, *J*=7.5, 4.5 Hz, 1H, CH), 6.30 (brd, *J*=6.2 Hz, 1H, NH). MS (CI/*i*-Bu) m/e 304 (MH^+, 100), 272 (MH^+-MeOH, 51), 229 ($MH^+-MeSCH_2CH_2$, 71), 146 (91), 104 (67); HRMS (CI/CH_4) calcd for $C_{14}H_{26}NO_4S$ (MH^+), 304.06436 found 304.04163.

3.9.3. 2-(4-Cyanobutyl)-malonic acid dimethyl ester (11).[21] Dimethyl malonate (0.02 mol, 2.64 g) in MeOH (5 mL) was added to an ice-cold solution obtained by addition of NaH (60% in oil, 0.02 mol, 0.98 g) to dry MeOH (20 mL). The mixture was stirred for a few minuets at room temperature and then 5-bromovaleronitrile (0.02 mol, 3.25 g) dissolved in MeOH (5 mL) was added followed by addition of a catalytic amount of KI. The mixture was refluxed for 48 h, filtered and evaporated. The inorganic salts were dissolved in water (20 mL) and were washed with EtOAc (3×20 mL). The organic phase was dried ($MgSO_4$), filtered and evaporated and the residue was flash chromatographed (hexane/EtOAc 2:1), to give the *C*-alkylated product 30% yield. ^{1}H NMR ($CDCl_3$) δ 1.47 (quintet, *J*=7 Hz, 2H, $CH_2(CH_2)_3CN$), 1.70 (quintet, *J*=7 Hz, 2H, $CH_2(CH_2)_2CN$), 1.93 (q, *J*=7.5 Hz, 2H, CH_2CH_2CN), 2.37 (t, *J*=7 Hz, 2H, CH_2CN), 3.37 (t, *J*=7.4 Hz, 1H, CH), 3.74 (s, 6H, OMe); ^{13}C NMR ($CDCl_3$) δ16.7 (CH_2CN), 24.8 (CH_2CH_2CN), 26.1 ($CHCH_2$), 51.1 (OMe), 52.3 (*C*H), 119.1 (CN), 169.3 (CO_2Me); MS (CI, NH_3) *m/e* 231 (MNH_4^+, 100), 214 (MH^+, 6); HRMS (DCI, CH_4) calcd for $C_{10}H_{16}NO_4$ (MH^+) 214.1079 found 214.1074.

3.9.4. 2-(4-Cyanobutyl)-malonic acid monomethyl ester potassium salt (12). KOH (1 mmol) in MeOH (5 mL) was added to a stirred solution of **11** (1 mmol) in MeOH (2 mL). The mixture was stirred overnight, and was then evaporated to dryness to give the product in quantitative yield. ^{1}H NMR ($CDCl_3$) δ 1.45 (m, 2H, $CH_2(CH_2)_2CN$), 1.63 (m, 2H, $CH_2(CH_2)_3CN$), 1.88 (m, 2H, CH_2CH_2CN), 2.41 (t, *J*=6.7 Hz, 2H, CH_2CN), 3.18 (t, *J*=6.2 Hz, 1H, CH), 3.71 (s, 3H, OMe); ^{13}C NMR ($CDCl_3$) δ16.9 (CH_2CN), 25.0 (CH_2CH_2CN), 27.0 ($CHCH_2CH_2$), 29.1 ($CHCH_2$), 52.0 (OMe), 55.5 (*C*H), 120.6 (CN), 174.2 (CO_2^-), 174.7 (CO_2Me); MS (CI, NH_3) *m/e* 173 ($MNH_4^+-CO_2K$, 100), 214 (MH^+, 6).

3.10. Acylation of *N*-Boc-amino acids with 12

To a solution of an *N*-Boc-amino acid (0.84 mmol) in dry THF (5 mL), CDI (1 mmol) was added portion wise and the obtained mixture was stirred at room temperature for 1 h. $MgCl_2$ (0.84 mmol) and **12** (0.84 mmol) were then added and the resulting mixture was stirred under reflux overnight. The obtained suspension was filtered, evaporated, and the crude residue was purified by flash chromatography (hexane/EtOAc usually 2:1).

3.10.1. 2-(2-*tert*-Butoxycarbonylamino-propionyl)-6-cyano-hexanoic acid methyl ester (13′b). From **1bI** and **12** The product was obtained as a mixture of two diastereomers, 23% yield). ^{1}H NMR ($CDCl_3$) δ 1.34 (two d, *J*=7.2 Hz, 3H, Me), 1.44–1.45 (two s, 9H, *t*-Bu), 1.44 (m, 2H, $CH_2(CH_2)_2CN$), 1.67 (quintet, *J*=7.4 Hz, 2H, CH_2CH), 1.89 (quintet, *J*=7.4 Hz, 2H, CH_2CH_2CN), 2.36 (t, *J*=7 Hz, 2H, CH_2CN), 3.73 (s and m, superimposed, 4H, OMe, $CHCO_2$), 4.39 (m, 1H, *CH*Me), 5.11 (bt, *J*=7.6 Hz, 1H, NH); ^{13}C NMR ($CDCl_3$) δ16.7 and 16.8 (CH*Me*), 17.4 (CH_2CN), 25.0 and 25.1(CH_2CH_2CN), 26.3 and 26.4 ($CHCH_2CH_2$), 28.2 (Me_3C), 27.6 and 27.9 ($CHCH_2$), 52.4 ($CHCO_2$), 52.46 and 52.52 (OMe), 54.9 (*C*HMe), 80.01 (C), 119.2 (CN), 155.0 (CO_2NH), 169.4 (CO_2Me), 204.4 (CO); MS (CI, NH_3) *m/e* 344 (MNH_4^+, 100), 327 (MH^+, 10.9), 288 ($MNH_4^+-C_4H_8$, 74), 271 ($MH^+-C_4H_8$, 7), 227 (MH^+-Boc, 11); HRMS (DCI, CH_4) calcd for $C_{16}H_{27}N_2O_5$ (MH^+) 327.1920 found 327.1840.

3.10.2. 2-[2-*tert*-Butoxycarbonylamino-3-(tetrahydro-pyran-2-yloxy)-propyl]-6-cyano-hexanoic acid methyl ester (13′qI). From **1q** and **12**. The product was obtained as a mixture of two diastereomers, 18% yield). ^{1}H NMR ($CDCl_3$) δ 1.46 (s, 9H, Me_3C), 1.46–1.94 (m, 12H, $(CH_2)_3CH$ and $(CH_2)_3CH_2CN$), 2.35 (t *J*=7 Hz, 2H, CH_2CN), 3.60 (m, 2H, CH_2CHN), 3.70 and 3.71 and 3.73 and 3.74 (four s, 3H, OMe), 3.85 (m, 3H, CH_2OCH and *CH*COO), 4.59 (m, 2H, *CH*NH and OCHO), 5.63–5.38 (m, 1H, NH); ^{13}C NMR ($CDCl_3$) δ 16.9 (CH_2CN), 19.3 and 19.6 and 19.7 (CH_2CH_2CH), 25.2 (CH_2CH_2CN), 26.4 and 26.5 and 26.8 and 26.9 (CH_2CH_2O and $CH_2(CH_2)_2CN$), 27.2 and 27.5 ($CH_2(CH_2)_3CN$), 28.3 (Me_3C), 30.3 and 30.4 and 30.5 (CH_2CHO), 52.4 and 52.6 (OMe), 54.8 and 55.0 and 55.2 and 55.4 ($CHCO_2$), 59.0 and 59.4 and 59.7 and 60.4 (*C*HNH), 62.3 and 62.8 and 63.0 (CH_2OCH), 67.1 and 67.7 and 68.9 (CH_2O), 80.1 and 80.3 (C), 99.2 and 99.5 and 99.9 (O*C*HO), 119.3 (CN), 155.4 ($NHCO_2$), 169.2 and 169.3 (CO_2Me), 202.2 and 203.5 (CO); MS (CI, NH_3) *m/e* 444 (MNH_4^+, 100), 427 (MH^+, 6), 388 ($MNH_4^+-C_4H_8$, 21), 360 ($MNH_4^+-C_5H_8O$, 85), 343 (MNH_4^+-HBoc, 32).

3.10.3. 2-(2-*tert*-Butoxycarbonylamino-3-hydroxy-butyryl)-6-cyano-hexanoic acid methyl ester (13′r). In the course of the workup of the previous reaction, the THP protective group was removed. The product was obtained as a mixture of two diastereomers, 12% yield. ^{1}H NMR ($CDCl_3$) δ 1.25 (d, *J*=6.3 Hz, 3H, CH*Me*), 1.47 (s, 9H, Me_3C), 1.70 (m, 4H, $(CH_2)_2(CH_2)_2CN$), 1.92 (m, 2H, CH_2CH_2CN), 2.36 (t *J*=7 Hz, 2H, CH_2CN), 3.37 (t, *J*=7.4 Hz, 1H, *CH*Me), 3.73 (m, 1H *CH*CO_2), 3.75 (s, 3H, OMe), 4.31 (m, 1H, *CH*NH), 5.33 (m, 1H, NH); ^{13}C NMR ($CDCl_3$) δ 16.9 (CH_2CN), 19.9 (CH*Me*), 25.1 ($CH_2(CH_2)_3$-CN), 26.4 ($CH_2(CH_2)_2CN$), 28.0 (CH_2CH_2CN), 28.3 (Me_3C), 51.3 ($CHCO_2$), 52.6 (OMe), 68.2 (*C*HMe), 80.2 (C), 119.3 (CN), 169.5 ($NHCO_2$), 171.9 (CO_2OMe), 205.0 (CO).

3.11. Oxidation of KAPA·HCl analogs[22]

To a solution of NH_2OH·HCl (1.5 mmol) in dry pyridine

(0.75 mL) was added to an ice-cold solution of a KAPA·HCl derivative (1 mmol) in dry pyridine (0.75 mL). The mixture was stirred at room temperature for 24 h and was then evaporated to dryness. The residue was dissolved in distilled water (3 mL) and was basified with 0.5 N NaOH to pH=8. This solution was extracted with CH_2Cl_2 (6×5 mL). The aqueous phase was evaporated yielding the desired product.

3.11.1. 8-Amino-7-hydroxyimino-octanoic acid hydrochloride (18a). Oximation of **4a** (98% yield). ^{1}H NMR (D_2O) δ 1.32–1.36 (m, 2H, $CH_2CH_2CH_2CO_2$), 1.55 (m, 4H, $CH_2CH_2CH_2CH_2CO_2$), 2.20 (t, J=7.3 Hz, 2H, CH_2CO_2), 2.41 (t, J=7.5 Hz, 2H, CH_2CNOH), 3.80 (s, 2H, CH_2N); ^{13}C NMR (D_2O) δ 24.8 ($CH_2CH_2CO_2$), 25.8 (CH_2CH_2CNOH), 26.2 (CH_2CNOH), 29.0 ($CH_2CH_2CH_2CO_2$), 37.4 (CH_2CO_2), 41.2 (CH_2N), 158.0 (CNOH), 183.6 (CO_2); MS (CI, NH_3) *m/e* 206 (MNH_4^+, 2), 189 (MH^+, 48).

3.11.2. 8-Amino-7-hydroxyimino-10-methyl-undecanoic acid hydrochloride (18e). Hydrolysis and decarboxylation of **3e** followed by in situ oximation (34% yield). ^{1}H NMR (D_2O) δ 0.98 (m, 7H, $CHMe_2$), 1.40 (m, 2H, CH_2CHMe_2), 1.72 (m, 8H, $(CH_2)_4CO_2$), 2.22 (t, J=7.3 Hz, 2H, CH_2CO_2), 4.05 (m, 1H, $CHNH_3^+$); MS (CI, NH_3) *m/e* 245 (MH^+, 67), 159 ($MH^+-C_5H_{11}N$, 100).

3.12. Preparation of DAPA derivatives 20 via catalytic reduction of KAPA oximes analogs 18[6]

A solution of the oxime analog (1 mmol) and 4 N HCl (2 mL) in absolute MeOH (10 mL) was hydrogenated in a Parr apparatus at 65 psi/40 °C/48 h over 10% PtO_2 (30 mg). The mixture was filtered through celite and washed with MeOH. The solvent was evaporated yielding the diamine product.

3.12.1. 7,8-Diamino-octanoic acid methyl ester dihydrochloride (20′a). Catalytic reduction of **18′a** (87% yield). ^{1}H NMR (D_2O) δ 1.43 (m, 4H, $CH_2CH_2CH_2CH_2CO_2$), 1.62 (m, 2H, CH_2CH_2CH), 1.75 (m, 2H, $CH_2CH_2CO_2$), 2.41 (t, J=7.3 Hz, 2H, CH_2CO_2), 3.33 (d, J=6.2 Hz, 2H, CH_2N), 3.63 (m, 1H, CH_2N), 3.68 (s, 3H, OMe); ^{13}C NMR (D_2O) δ 24.3 and 24.4 ($CH_2CH_2CH_2CH_2CO_2$), 28.3 ($CH_2CH_2CH_2CO_2$), 30.4 (CH_2CH), 34.3 (CH_2CO_2), 41.3 (CH_2N), 50.0 (CHN), 52.7 (OMe), 178.1 (CO_2Me); MS (DCI, NH_3) *m/e* 189 (MH^+, 100).

3.12.2. 7,8-Diamino-decanoic acid methyl ester dihydrochloride (20′c). Catalytic reduction of **18′c** (90% yield). ^{1}H NMR (D_2O) δ 1.08 (t, J=7.5 Hz, 3H, Me), 1.42 (m, 4H, $(CH_2)(CH_2)_2CO_2$), 1.77 (m, 6H, CH_2Me, $CH_2(CH_2)_2CH_2CH_2CO_2$), 2.42 (t, J=7.3 Hz, 2H, CH_2CO_2), 3.70 (m and s, superimposed, 5H, two CHN and OMe); ^{13}C NMR (D_2O) δ 9.7 (Me), 20.6 (CH_2Me), 24.3 ($CH_2CH_2CO_2$), 24.8 ($NCHCH_2CH_2$), 28.5 ($CH_2(CH_2)_3CO_2$), 34.1 (CH_2CO_2), 52.9 ($NHCH(CH_2)_2$), 179.2 (CO_2Me); MS (DCI, NH_3) *m/e* 217 (MH^+, 100), 203 (MH^+, 87); HRMS (DCI, CH_4) calcd for $C_{11}H_{25}N_2O_2$ (MH^+) 217.1916 found 217.1910.

3.12.3. 7,8-Diamino-9-methyl-octanoic acid methyl ester dihydrochloride (20′d). Catalytic reduction of **18′d** (92% yield). ^{1}H NMR (D_2O) δ 1.10 (m, 3H, $CHMe_2$), 1.46 (m, 4H, $CH_2(CH_2)_2CH_2CH_2CO_2$), 2.17 (m, 1H, $CHMe_2$), 2.43 (t, J=7 Hz, 2H, CH_2CO_2), 3.42 (m, 1H, $CHCHMe_2$), 3.70 (s, 3H, OMe), 3.82 (m, 1H, CHN); ^{13}C NMR (D_2O) δ 17.0 (Me), 17.5 (Me), 19.6 (CH_2Me_2), 24.4 ($CH_2(CH_2)_3CO_2$), 24.6 ($CH_2CH_2CO_2$), 27.8 ($CH_2(CH_2)_2CO_2$), 28.2 ($CH_2(CH_2)_4CO_2Me$), 34.1 (CH_2CO_2), 51.6 ($CH(CH_2)_5$), 52.3 ($CHCHMe_2$), 58.9 (OMe), 179.2 (CO_2Me); MS (DCI, NH_3) *m/e* 231 (MH^+, 100); HRMS (DCI, CH_4) calcd for $C_{12}H_{27}N_2O_2$ (MH^+) 231.2072 found 231.2110.

3.13. Preparation of DAPA derivatives 20 via sodium cyanoborohydride reductive amination of ketones 5[23]

To a solution of an *N*-Boc-KAPA derivative methyl ester (1 mmol) in dry MeOH (10 mL), NH_4OAc (10 mmol) was added. The solution obtained was stirred at room temperature for 10 min, then $NaBH_3CN$ (3.7 mmol) was added in one portion and the resulting mixture was stirred at room temperature for 48 h. The reaction was quenched with 1 N HCl (20 mL). The solvent was evaporated and the crude residue was dissolved in water. KOH was added till a basic solution was obtained. The aqueous phase was extracted with CH_2Cl_2 (3×20 mL). The combined organic layer was washed with brine (2×10 mL), dried ($MgSO_4$), filtered and evaporated to give the compound.

3.13.1. 7-Amino-8-*tert*-butoxycarbonylamino-octanoic acid methyl ester (19′aI). Reductive amination of **5′aI** (74% yield). ^{1}H NMR ($CDCl_3$) δ 1.33 (m, 4H, $(CH_2)_2(CH_2)_2CO_2$), 1.44 (s, 9H, Me_3C), 1.62 (m, 4H, $CH_2(CH_2)_2CH_2CH_2CO$), 1.78 (m, 2H, NH_2), 2.31 (t, J=7.5 Hz, 2H, CH_2CO_2), 2.86 (m, 2H, CH_2NH), 3.26 (m, 1H, CH), 3.67 (s, 3H, OMe), 4.98 (m, 1H, NH); ^{13}C NMR ($CDCl_3$) δ 24.8 ($CH_2(CH_2)_3CO_2$), 25.7 ($CH_2CH_2CO_2$), 28.4 (Me_3C), 29.2 ($CH_2(CH_2)_2CO_2$), 34.0 (CH_2CO_2), 35.5 ($CHCH_2CH_2$), 47.0 (CH_2NH), 51.3 (OMe), 51.5 (CH), 79.3 (C), 156.3 (CO_2NH), 174.2 (CO_2Me); MS (CI, NH_3) *m/e* 289 (MH^+, 100), 233 ($MH^+-C_4H_8$, 18), 189 (MH^+-Boc, 4); HRMS (DCI, CH_4) calcd for $C_{14}H_{29}N_2O_4$ (MH^+) 289.2127 found 289.2140.

3.14. Acid cleavage the *N*-Boc protecting group

To a suspension of an *N*-Boc-amino compound (1 mmol) in ether (5 mL), a solution of 1 N HCl–ether (3 mL) was added. The mixture was stirred at room temperature overnight. The solvent was evaporated to give the diamine dihydrochloride product. In some cases, a solution of HCl in EtOAc (prepared from a mixture of EtOAc, EtOH and AcCl was used.[16]

3.14.1. 7,8-Diamino-9-methylsulfanyl-nonanoic acid methyl ester dihydrochloride (20′i). From **5′iI** by reductive amination and hydrolysis (50% yield). ^{1}H NMR (D_2O) δ 1.40 (m, 4H, $(CH_2)_2(CH_2)_2CO_2$), 1.60 (m, 4H, $CH_2(CH_2)_4CO_2$), 2.23 (s, 3H, SMe), 2.42 (t, J=7.4 Hz, 2H, CH_2CO_2), 2.93 (ABq of d, J_{AB}=16 Hz, J_{AX}=9.5 Hz, J_{BX}=5.5 Hz, 2H, CH_2S), 3.70 (s, 3H, OMe), 3.80 (m, 2H, two CH's). ^{13}C NMR (D_2O) δ 15.4 (SMe), 24.4 ($CH_2CH_2CO_2$), 24.8 and 24.9 (two CH_2CH_2CHN), 27.4 ($CH_2(CH_2)_2CO_2$), 28.3 and 28.5 (two CH_2CHN), 31.6 and 32.7 (two CH_2S), 34.0 (CH_2CO_2), 51.4 (OMe), 51.9 and 52.6 (two $CHCH_2S$), 52.7 and 52.8 (two $CHCHCH_2S$), 175.0 (CO_2). MS (CI/NH_3) *m/e* 249 (MH^+-2HCl, 100).

3.15. DTB and derivatives—imidazolone formation

Method A.[24] A mixture of a DAPA·2HCl Me ester derivative (1 mmol) and Et_3N (0.2 g, 2 mmol) in dry CH_2Cl_2 (10 mL) was stirred in an ice bath for 30 min. A solution of CDI (0.2 g, 1.2 mmol) in dry CH_2Cl_2 (5 mL) was added and the mixture was stirred at room temperature overnight. The mixture was evaporated and the residue was partitioned between 1 N HCl and EtOAc. The aqueous layer was extracted with EtOAc (4×) and the combined organic layers were washed with 5% $NaHCO_3$ (2×), brine, dried ($MgSO_4$), filtered and evaporated to give the product, in the form of two diastereomers.

Method B.[25] An aqueous solution (10 mL) of NaOH (0.4 g, 10 mmol) and a DAPA·2HCl derivative (1 mmol) was stirred at room temperature for 10 min. A solution of triphosgene (0.3 g, 1 mmol) in dioxane (10 mL) was added. The mixture became hot and the pH dropped from ~10 to ~8. The resulting mixture was stirred at room temperature for 2 days and was then evaporated. Trituration of the residue with MeOH, filtration and evaporation afforded the product. Note: both the free acid and the ester could be used as substrates. The product was either the pure acid or a mixture of acid and ester. In the case of a mixture, the product was dissolved in 4 N HCl and stirred at room temperature overnight, to afford, after evaporation, the free acid.

3.15.1. 6-(2-Oxo-imidazolidin-4-yl)-hexanoic acid (21a).[26] From **20′a**, method A (97% yield). ^{1}H NMR (CD_3OD) δ 1.4 (m, 4H, (CH_2)$_2$(CH_2)$_2$$CO_2$), 1.64 (m, 4H, CH_2(CH_2)$_2$$CH_2$$CH_2CO_2$), 2.32 (m, 2H, CH_2CO_2), 3.07 (m, 1H, CHN), 3.80 (m, 2H, CH_2N); ^{13}C NMR (D_2O) δ 24.6 and 26.2 (CH_2(CH_2)$_2$$CO_2$ and CH_2(CH_2)$_3$$CO_2$), 29.0 ($CH_2CH_2$-$CO_2$), 35.1 ($CH_2CO_2$), 38.0 ($CH_2$($CH_2$)$_4$$CO_2$), 46.6 ($CH_2$N), 53.3 (CHN), 163.7 (NCON), 184.1 (CO_2); MS (CI, *i*-Bu) *m/e* 201 (MH^+, 100); HRMS (DCI, CH_4) calcd for $C_9H_{17}N_2O_2$ (MH^+) 201.1239 found 201.1200.

3.15.2. 6-(5-Ethyl-2-oxo-imidazolidin-4-yl)-hexanoic acid (21c).[27] Cyclization of **20′c**, method A (97% yield). ^{1}H NMR (D_2O) δ 0.89 (t, *J*=7 Hz, 4H, Me), 1.33 (m, 4H, CH_2CH_2(CH_2)$_2$$CO_2$), 1.52 (m, 6H, CH_2(CH_2)$_2$$CH_2$$CH_2CO_2$ and CH_2Me), 2.16 (t, *J*=7 Hz, 2H, CH_2CO_2), 3.41 (m, 1H, $CHCH_2Me$), 3.73 (m, 2H, CH(CH_2)$_5$); ^{13}C NMR (D_2O) δ 9.1 (Me), 24.6 (CH_2(CH_2)$_3$$CO_2$), 26.3 ($CH_2CH_2CO_2$), 28.4 ($CH_2$($CH_2$)$_2$$CO_2$), 29.1 ($CH_2CO_2$), 35.4 ($CH_2$Me), 38.1 ($CH_2$($CH_2$)$_4$$CO_2$), 58.3 ($CH_2CHN$), 60.1 (CHN), 165.5 (NCON), 175.2 (CO_2); MS (CI, NH_3) *m/e* 243 (MH^+, 42), 211 (MH^+-MeOH, 31); HRMS (DCI, CH_4) calcd for $C_{11}H_{21}N_2O_2$ (MH^+) 229.1552 found 229.1570.

3.16. General procedure for the preparation of imadizolidinethiones

A mixture of a DAPA·2HCl methyl ester derivative **20′** (1 mmol) and Et_3N (0.2 g, 2 mmol) in dry CH_2Cl_2 (10 mL) was stirred at ca. 0 °C for 30 min. A solution of *N*,*N*′-thiocarbonyldiimidazole (0.21 g, 1.2 mmol) in dry CH_2Cl_2 (10 mL) was added and the mixture was stirred at room temperature overnight. The resulting mixture was evaporated to dryness and the residue partitioned between 1 N HCl and EtOAc. The aqueous layer was extracted with EtOAc (4 times) and the combined organic layers were washed with 5% $NaHCO_3$ (twice) and brine (once), dried ($MgSO_4$), filtered and evaporated to give the desired cyclized product, as a mixture of two diastereomers.

3.17. 5-(5-Methyl-2-thioxo-imidazolidin-4-yl)-pentanoic acid methyl ester (22′b)

From **20′b**, as a viscous yellow oil (84% yield). MS (CI/NH_3) *m/e* 245 (MH^+, 100), 213 (MH^+-MeOH, 4); HRMS (CI/CH_4) calcd for $C_{11}H_{21}N_2O_2S$ (MH^+), 245.13238 found 245.06617.

3.17.1. 5-(5-Methyl-2-thioxo-imidazolidin-4-yl)-pentanoic acid (22b). Hydrolysis of **22′b**, yellow solid (quantitative yield). ^{1}H NMR (D_2O) δ cis diastereomer 1.33 (m, 4H, $CH_2CH_2CH_2CH_2CO_2$), 1.41 (d, *J*=6.5 Hz, 3H, Me), 1.57 (m, 4H, $CH_2CH_2CH_2CH_2CH_2CO_2$), 2.35 (m, 2H, CH_2CO_2), 4.12 (m, 1H, $CHCH_2$), 4.08 (q, *J*=6.5 Hz, 1H, CHMe). trans diastereomer 1.19 (d, *J*=6.6 Hz, 3H, Me), 1.33 (m, 4H, $CH_2CH_2CH_2CH_2CO_2$), 1.57 (m, 4H, CH_2-$CH_2CH_2CH_2CH_2CO_2$), 2.35 (m, 2H, CH_2CO_2), 3.72 (m, 1H, $CHCH_2$), 4.40 (q, *J*=6.8 Hz, 1H, CHMe). MS (CI/NH_3) *m/e* 231 (MH^+, 100); HRMS (CI/CH_4) calcd for $C_{10}H_{19}N_2O_2S$ (MH^+), 231.11672 found 231.09542; (M^+), 230.10890 found 230.09407.

3.17.2. 5-(5-Methylsulfanylmethyl-2-thioxo-imidazolin-4-yl)-pentanoic acid (22i). From **20′I**, tan crystals (82% yield). ^{1}H NMR ($CDCl_3$) δ 1.38 (m, 4H, $CH_2CH_2CH_2CH_2$-CO_2), 1.64 (m, 4H, $CH_2CH_2CH_2CH_2CH_2CO_2$), 2.14 and 2.15 (two s, 3H, SMe), 2.20 (m, 1H, CH_2S), 2.33 (t, *J*=7.3 Hz, 2H, CH_2CO_2), 2.65 (m, 1H, CH_2S), 3.68 (s, 3H, OMe), 3.70 (m, 1H, CH), 4.03 (m, 1H, CH), 6.60–6.90 (several m, 2H, two NH's). ^{13}C NMR ($CDCl_3$) δ 15.7 and 15.9 (two SMe), 24.63 and 24.9 (two $CH_2CH_2CO_2$), 25.8 and 26.2 (two $CH_2CH_2CH_2CH_2CO_2$), 28.7 and 28.8 (two $CH_2CH_2CH_2CO_2$), 29.7 ($CH_2CH_2CH_2CH_2CH_2CO_2$), 33.8 and 34.0 (two CH_2CO_2), 35.1 and 38.9 (two CH_2S), 51.5 (OMe), 58.4 and 59.9 (two $CHCH_2S$), 61.7 and 62.6 (two $CHCHCH_2S$), 173.9 (CO_2), 182.3 (CS). MS (CI/NH_3) *m/e* 291 (MH^+, 100), 259 (MH^+-MeOH, 4); HRMS (CI/CH_4) calcd for $C_{12}H_{23}N_2O_2S_2$ (MH^+), 291.12010 found 291.08042.

3.17.3. 5-[5-(2-Methylsulfanyl-ethyl)-2-thioxo-imidazolidin-4-yl]-pentanoic acid (22g). From **20′g**, yellow powder (quantitative yield). ^{1}H NMR ($CDCl_3$) δ 1.36 (m, 4H, $CH_2CH_2CH_2CH_2CO_2$), 1.64 (m, 4H, $CH_2CH_2CH_2CH_2CH_2$-CO_2), 1.88 (m, 1H, CH_2CH_2S), 2.11 (s, 3H, SMe), 2.32 (t, *J*=7.3 Hz, 2H, CH_2CO_2), 2.59 (t, *J*=7 Hz, 2H, CH_2S), 2.65 (m, 1H, CH_2CH_2S), 3.67 (s, 3H, OMe), 3.75 (m, 1H, CH), 4.03 (m, 1H, CH), 7.00–7.40 (several m, 2H, two NH's). ^{13}C NMR ($CDCl_3$) δ 15.5 (SMe), 24.7 ($CH_2CH_2CO_2$), 25.0 ($CH_2CH_2CH_2CH_2CO_2$), 28.8 ($CH_2CH_2CH_2CO_2$), 28.9 ($CH_2CH_2CH_2CH_2CH_2CO_2$), 30.3 (CH_2CH_2S), 31.2 (CH_2S), 33.9 (CH_2CO_2), 51.5 (OMe), 59.3 and 60.3 (two $CHCH_2CH_2S$), 62.1 and 63.0 (two $CHCHCH_2CH_2S$), 174.0 (CO_2), 181.8 (CS). MS (CI/NH_3) *m/e* 305 (MH^+, 100), 273 (MH^+-MeOH, 11); HRMS (CI/CH_4) calcd for $C_{13}H_{25}N_2O_2S_2$ (MH^+), 305.13575 found 305.07004.

3.18. General procedure for the preparation of *N*-Cbz amino acid methyl ester derivatives

Benzyl chloroformate (0.16 mL, 1.1 mmol) was added dropwise, during 30 min to an ice-cooled turbid mixture of the amino acid hydrochloride Me ester (1 mmol) and $NaHCO_3$ (0.4 g, 5 mmol) in EtOAc (3 mL)/H_2O (2 mL). The mixture was stirred at room temperature for 3 h and was then decanted. The organic layer was washed with 1 N HCl (2×), H_2O (2×), dried ($MgSO_4$), filtered and evaporated to give the product.

3.18.1. 7,8-Bis-benzyloxycarbonylamino-10-methylsulfanyl-decanoic acid methyl ester (20′gII). Compound **20′gII** was obtained as a viscous oil from **20′g** (63% yield). Note: two equivalents of benzyl chloroformate were used. ^{1}H NMR ($CDCl_3$) δ 1.33 (m, 4H, C*H*$_2$C*H*$_2$CH$_2$CH$_2$CO$_2$), 1.60 (m, 4H, C*H*$_2$(CH$_2$)$_2$CH$_2$CO), 1.75 (m, 2H, C*H*$_2$CH$_2$S), 2.07 (s, 3H, SMe), 2.28 (t, J=7.2 Hz, 2H, CH$_2$CO$_2$), 2.54 (m, 2H, CH$_2$S), 3.66 (s, 3H, OMe), 3.78 (m, 2H, two CH's), 4.70–5.00 (several m, 2H, two NH's), 5.08 (two s, 4H, C*H*$_2$Ar), 7.32 (two m, 10H, Ar). MS (CI/NH_3) *m/e* 548 (MNH_4^+, 67), 531 (MH^+, 100), 456 ($MNH_4^+-CH_2Ph$, 5), 440 (MH^+-CH_2Ph, 4), 397 ($MH^+-CO_2CH_2Ph$, 12); HRMS (CI/CH_4) calcd for $C_{28}H_{39}N_2O_6S$ (MH^+), 531.25228 found 531.25697.

3.18.2. 7,8-Bis-methoxycarbonylamino-10-methylsulfanyl-decanoic acid methyl ester (20′gIII). From **20′g**. Et_3N (0.2 g, 2 mmol) was added to a suspension of **20′g** (0.26 g, 1 mmol) in dry CH_2Cl_2 (5 mL). The mixture was stirred vigorously until it became homogenous and was further stirred at room temperature for 10 min. Dimethyl dicarbonate (0.26 g, 2 mmol) was added and the mixture was stirred at room temperature for 3.5 h. The resulting mixture was evaporated to dryness and the residue was partitioned between 1 N HCl and EtOAc. The aqueous layer was extracted with EtOAc (4×) and the combined organic layers were dried ($MgSO_4$), filtered and evaporated to give **20′gIII** (98% yield). ^{1}H NMR ($CDCl_3$) δ 1.35 (m, 4H, CH(C*H*$_2$)$_2$(CH$_2$)$_2$CO$_2$), 1.63 (m, 4H, CHC*H*$_2$(CH$_2$)$_2$C*H*$_2$), 2.11 (m, 1H, C*H*$_2$CH$_2$S), 2.10–2.12 (several s, 3H, SMe), 2.29 (m, 1H, C*H*$_2$CH$_2$S), 2.27 and 2.30 (two t, J=7.5, 7 Hz, 2H, CH$_2$CO$_2$), 2.55 (m, 1H, CH$_2$S), 2.68 (t, J=6.9 Hz, 1H, CH$_2$S), 3.65–3.76 (several s, 9H, CH$_2$CO$_2$*Me* and two NCO$_2$*Me*), 3.75–4.02 (several m, 2H, two CH's), 4.70–5.25 (several m, 2H, two NH's). MS (CI/NH_3) *m/e* 396 (MNH_4^+, 86), 379 (MH^+, 100), 364 (MH^+-MeOH, 10), 347 (MH^+-two MeOH, 45).

3.18.3. 7,8-Bis-benzyloxycarbonylamino-10-methanesulfinyl-decanoic acid methyl ester (20′hII). Periodate oxidation[10e] of **20′gII** (93% yield). Notes: (a) due to the insolubility of **20′gII** in MeOH and H_2O, the starting material was suspended in MeOH, while CH_2Cl_2 was added until complete dissolution and only then was the aqueous solution of $NaIO_4$ added (b) partial NMR spectrum of **20′hII** compared to that of **20′gII**. ^{1}H NMR ($CDCl_3$) δ 2.49 (s, 3H, SMe) compared to 2.07 (s, 3H, SMe), and 2.71 (m, 2H, CH$_2$S) compared to 2.54 (m, 2H, CH$_2$S). MS (DCI/NH_3) *m/e* 564 (MNH_4^+, 29), 547 (MH^+, 100), 456 (MH^+-CH_2Ph, 12), 439 (MH^+-OCH_2Ph, 61), 412 ($MH^+-CO_2CH_2Ph$, 13), 395 ($MH^+-CO_2CH_2Ph-O$, 44), 321 ($MH^+-CO_2-CH_2Ph-CH_2Ph$, 2), 305 ($MH^+-CO_2CH_2Ph-OCH_2Ph$, 96); HRMS (CI/CH_4) calcd for $C_{28}H_{39}N_2O_7S$ (MH^+), 547.24780 found 547.23947.

3.18.4. 10-Methanesulfinyl-7,8-bis-methoxycarbonylamino-decanoic acid methyl ester (20′hIII). Periodate oxidation[10e] of **20′gIII** (93% yield). Note: partial NMR spectrum of **20′hIII** compared to that of **20′gIII**. ^{1}H NMR ($CDCl_3$) δ 2.58–2.67 (s, 3H, SMe) compared to 2.10–2.13 (s, 3H, SMe) and 2.79 (m, 2H, CH$_2$S), as compared to 2.55 and 2.68 (two m, 2H, CH$_2$S. MS (DCI/NH_3) *m/e* 412 (MNH_4^+, 33), 395 (MH^+, 100), 363 (MH^+-MeOH, 17); HRMS (CI/CH_4) calcd for $C_{16}H_{31}N_2O_7S$ (MH^+), 395.18520 found 395.17896.

3.19. Pyrolysis of sulfoxides to vinyl derivatives

Method A.[10a] A mixture of the sulfoxide (1 mmol) and $CaCO_3$ (0.4 g, 4 mmol) in *o*-dichlorobenzene (5 mL) was stirred at room temperature for 1 h. The mixture was transferred to a pre-heated oil bath (ca. 174 °C). The pyrolysis was monitored by tlc (hexane/EtOAc 4:1, spraying with phosphomolybdic acid). The reaction was complete after 2 h and the product was obtained by in very poor yield flash chromatography.

Method B.[10b] The sulfoxide were distilled in a Kugelrohr apparatus at elevated temperatures (>200 °C) and high vacuum, and were usually obtained as oils that solidified at room temperature. Substrates containing labile Boc groups, or acidic hydrogens (e.g. β-keto esters) could not be pyrolyzed, due to decomposition of the substrate and product. Method B was usually followed, due to better yield.

3.19.1. 8-Acetylamino-7-oxo-dec-8-enoic acid methyl ester (23′IV). Undesired isomer obtained as from **5′hIV** when pyrolyzed at 220 °C and 1 Torr (70% yield). ^{1}H NMR ($CDCl_3$) δ1.34 (m, 2H, C*H*$_2$CH$_2$CH$_2$CO$_2$), 1.64 (m, 4H, C*H*$_2$CH$_2$C*H*$_2$CH$_2$CO$_2$), 1.85 (d, J=7.1 Hz, 3H, *Me*CH), 2.14 (s, 3H, MeCO), 2.32 (t, J=7.4 Hz, 2H, CH$_2$CO$_2$), 2.70 (dd, J=8, 6.7 Hz, 2H, CHCOC*H*$_2$), 3.67 (s, 3H, OMe), 6.67 (q, J=7.1 Hz, 1H, CH), 7.25 (brs, 1H, NH). ^{13}C NMR ($CDCl_3$) δ15.6 (*Me*CH), 24.2 (*Me*CO), 24.7 (*C*H$_2$CH$_2$CO$_2$), 25.0 (*C*H$_2$(CH$_2$)$_2$CO$_2$), 28.7 (*C*H$_2$CH$_2$CH$_2$CO$_2$), 33.8 (*C*H$_2$CO$_2$), 36.3 (*C*H$_2$COCH), 132.6 (CH), 134.5 (CNH), 173.7 (CON), 174.0 (CO$_2$), 197.4 (CH$_2$*C*OCH). MS (EI) m/e 255 (M^+, 18), 224 (M^+-MeOH, 2), 213 (M^+-MeCO$_2$, 100), 196 ($M^+-C_2H_4O_2$, 4), 182 (M^+-MeOH–MeCO$_2$, 48), 153 (M^+-MeCO$_2-C_2H_4O_2$, 20).

3.19.2. 7,8-Bis-methoxycarbonylamino-dec-9-enoic acid methyl ester (20′oIII). From **20′hIII**, at 240 °C and 2 Torr (41% yield). ^{1}H NMR ($CDCl_3$) δ 1.35 (m, 4H, (C*H*$_2$)$_2$-(CH$_2$)$_2$CO$_2$), 1.62 (m, 4H, C*H*$_2$(CH$_2$)$_2$C*H*$_2$CH$_2$CO), 2.32 (m, 2H, CH$_2$CO$_2$), 3.41 and 3.68 (two m, 1H, NC*H*CH$_2$), 3.67 (s, 9H, three OMe's), 3.68 and 4.21 (two m, 1H, CHC*H*CHCH$_2$CH$_2$), 4.50–5.00 (several m, 2H, two NH's), 5.22 and 5.30 (two m, 2H, C*H*$_2$=CH), 5.80 (m, 1H, CH$_2$=C*H*). ^{13}C NMR ($CDCl_3$) δ 23.0–35.0 (chain CH$_2$'s), 51.5 (CH$_2$CO$_2$*Me*), 52.3 and 53.4 (two NCO$_2$*Me*), 56.3 and 57.2 (two=CH*C*H), 59.0 and 61.7 (two N*C*HCH$_2$), 117.4 and 118.4 (two=*C*H), 134.1 and 137.3 (two *C*H$_2$CHN), 155.0 (CON), 174.0 (CO$_2$). MS (CI/NH_3) *m/e* 348 (MNH_4^+,

100), 331 (MH^+, 96), 316 (MNH_4^+-MeOH, 16), 299 (MH^+-MeOH, 41).

Note: experimental procedures for the following compounds may be obtained directly from the corresponding authors.

4-[(*tert*-Butyloxycarbonyl)amino]-5-(methylthio)-3-oxopentanoic acid methyl ester (**2′iI**).

4-[(*tert*-Butyloxycarbonyl)amino]-5-(*tert*-butyloxycarbonyl)thio]-3-oxopentanoic acid methyl ester (**2′kI**).

4-Benzyloxycarbonylamino-5-benzyloxycarbonylsulfanyl-3-oxo-pentanoic acid methyl ester (**2′lII**).

5-Benzylsulfanyl-4-*tert*-butoxycarbonylamino-3-oxo-pentanoic acid methyl ester (**2′mI**).

4-Acetylamino-5-acetylsulfanyl-3-oxo-pentanoic acid methyl ester (**2′nIV**).

4-*tert*-Butoxycarbonylamino-3-oxo-5-(tetrahydro-pyran-2-yloxy)-pentanoic acid methyl ester (**2′qI**).

4-*tert*-Butoxycarbonylamino-3-oxo-5-(tetrahydro-pyran-2-yloxy)-hexanoic acid methyl ester (**2′sI**).

4-Methoxycarbonylacetyl-oxazolidine-3-carboxylic acid *tert*-butyl ester (**16′p**).

4-Methoxycarbonylacetyl-5-methyl-oxazolidine-3-carboxylic acid *tert*-butyl ester (**16′r**).

2-(2-*tert*-Butoxycarbonylamino-propionyl)-heptanedioic acid 7-ethyl ester 1-methyl ester (**3′bI**).

2-(2-*tert*-Butoxycarbonylamino-butyryl)-heptanedioic acid 7-ethyl ester 1-methyl ester (**3′cI**).

2-(2-*tert*-Butoxycarbonylamino-3-methyl-butyryl)-heptanedioic acid 7-ethyl ester 1 methyl ester (**3′dI**).

2-(2-*tert*-Butoxycarbonylamino-4-methyl-pentanoyl)-heptanedioic acid 7-ethyl ester 1-methyl ester (**3′eI**).

2-(2-*tert*-Butoxycarbonylamino-3-methyl-pentanoyl)-heptanedioic acid 7-ethyl ester 1-methyl ester (**3′fI**).

2-(2-*tert*-Butoxycarbonylamino-4-methylsulfanyl-butyryl)-heptanedioic acid 7-ethyl ester 1-methyl ester (**3′gI**).

2-(2-*tert*-Butoxycarbonylamino-4-methanesulfinyl-butyryl)-heptanedioic acid 7-ethyl ester 1-methyl ester (**3′hI**).

Ethyl 8-[(*tert*-butyloxycarbonyl)amino]-6-(methoxycarbonyl)-9-(methylthio)-7-oxononanoate (**3′iI**).

2-(3-*tert*-Butoxycarbonyl-oxazolidine-4-carbonyl)-heptanedioic acid 7-ethyl ester 1-methyl ester (**17′p**).

2-(3-*tert*-Butoxycarbonyl-5-methyl-oxazolidine-4-carbonyl)-heptanedioic acid 7-ethyl ester 1-methyl ester (**17′r**).

8-Amino-7-oxo-decanoic acid hydrochloride (**4c**).

8-Amino-10-methylsulfanyl-7-oxo-decanoic acid hydrochloride (**4g**).

8-Amino-10-methanesulfinyl-7-oxo-decanoic acid hydrochloride (**4h**).

8-Amino-9-methylsulfanyl-7-oxo-nonanoic acid hydrochloride (**4i**).

8-Amino-9-mercapto-7-oxo-nonanoic acid hydrochloride (**4j**).

8-Amino-9-hydroxy-7-oxo-nonanoic acid hydrochloride (**4p**).

8-Amino-9-hydroxy-7-oxo-decanenitrile hydrochloride (**14r**).

8-Amino-9-hydroxy-7-oxo-nonanoic acid methyl ester hydrochloride (**5′p**).

8-Amino-9-hydroxy-7-oxo-decanoic acid methyl ester hydrochloride (**5′r**).

8-Amino-10-methylsulfanyl-7-oxo-decanoic acid methyl ester hydrochloride (**5′g**).

8-Amino-9-methylsulfanyl-7-oxo-nonanoic acid methyl ester hydrochloride (**5′I**).

8-Amino-9-mercapto-7-oxo-nonanoic acid methyl ester hydrochloride (**5′j**).

8-*tert*-Butoxycarbonylamino-7-oxo-octanoic acid methyl ester (**5′aI**).

8-(*tert*-Butoxycarbonylamino)-9-hydroxy-7-oxo-decanoic acid methyl ester (**5′qI**).

8-*tert*-Butoxycarbonylamino-10-methylsulfanyl-7-oxo-decanoic acid methyl ester (**5′gI**).

8-*tert*-Butoxycarbonylamino-10-methanesulfinyl-7-oxo-decanoic acid methyl ester (**5′hI**).

8-*tert*-Butoxycarbonylamino-9-methylsulfanyl-7-oxo-nonanoic acid methyl ester (**5′iI**).

8-Amino-7-hydroxyimino-octanoic acid methyl ester hydrochloride (**18′a**).

8-Amino-7-hydroxyimino-octanoic acid hydrochloride (**18c**).

8-Amino-9-methyl-7-hydroxyimino-decanoic acid hydrochloride (**18d**).

8-Amino-7-hydroxyimino-9-methyl-undecanoic acid hydrochloride (**18f**).

8-Amino-7-hydroxyimino-9-mercapto-nonanoic acid hydrochloride (**18j**).

8-Amino-7-hydroxyimino-9-mercapto-nonanoic acid methyl ester hydrochloride (**18′j**).

7-Amino-8-*tert*-butyloxycarbonylamino-9-hydroxy-decanoic acid methyl ester (**19′rI**).

7,8-Diamino-9-hydroxy-decanoic acid methyl ester dihydrochloride (**20′r**).

7,8-Diamino-dec-9-enoic acid dihydrochloride (**20o**).

7,8-Diamino-10-methylsulfanyl-decanoic acid methyl ester dihydrochloride (**20′g**).

6-(2-Oxo-imidazolidin-4-yl)-hexanoic acid methyl ester (**21′a**).

6-[5-(2-Methylsulfanyl-ethyl)-2-oxo-imidazolidin-4-yl]-hexanoic acid (**21g**).

6-(5-Methylsulfanylmethyl-2-oxo-imidazolidin-4-yl)-hexanoic acid (**21i**).

6-[5-(2-Methanesulfinyl-ethyl)-2-oxo-imidazolidin-4-yl]-hexanoic acid (**21h**).

Acknowledgements

Generous support for this work by the 'Marcus Center for Pharmaceutical and Medicinal Chemistry' and the 'Bronia and Samuel Hacker Fund for Scientific Instrumentation' at Bar Ilan University, are gratefully acknowledged.

References and notes

1. (a) Izumi, Y.; Yamada, H. In *Biotechnology of vitamins, pigments and growth factors*; Vadamme, E. J., Ed.; Elsevier: London, 1989; pp 231–256. (b) In *Biotin. Ann. NY Acad. Sci.*, Dakshinamurti, K., Bhagavan, N. H., Eds.;, 1985; 447, pp 1–441. (c) Knowles, J. R. *Annu. Rev. Biochem.* **1989**, *58*, 195–221.
2. (a) Izumi, Y.; Yamada, H. In *Biotechnology of vitamins, pigments and growth factors*; Vadamme, E. J., Ed.; Elsevier: London, 1989; pp 231–256. (b) In *Biotin. Ann. NY Acad. Sci.*, Dakshinamurti, K., Bhagavan, N. H., Eds.;, 1985; 447, pp 1–441. (c) Knowles, J. R. *Annu. Rev. Biochem.* **1989**, *58*, 195–221.
3. Gibson, K. J.; Lorimer, G. H.; Rendina, A. R.; Taylor, W. S.; Cohen, G.; Gatenby, A. A.; Payne, W. G.; Roe, D. C.; Lockett, B. A.; Nudelman, A.; Marcovici, D.; Nachum, A.; Wexler, B. A.; Marsilii, E. L.; Turner, Sr. I. M.; Howel, D.; Kalbach, C. E.; Chi, H. *Biochemistry* **1995**, *34*, 10976–10984.
4. (a) Baldet, P.; Gerbling, H.; Axiotis, S.; Douce, R. *Eur. J. Biochem.* **1993**, *217*, 479–485. (b) Patton, D. A.; Johnson, M.; Ward, E. R. *Plant Physiol.* **1996**, *112*, 371–378. (c) Shellhammer, J.; Meinke, D. *Plant Physiol.* **1990**, *110*, 1021–1028. (d) Patton, D. A.; Volrath, S.; Ward, E. R. *Mol. Gen. Genet.* **1996**, *251*, 261–266. (e) Patton, D. A.; Schetter, A. L.; Franzmann, L. H.; Nelson, K.; Ward, E. R.; Meinke, D. W. *Plant Physiol.* **1998**, *116*, 935–946.
5. (a) Bui, B. T. S.; Florentin, D.; Fournier, F.; Ploux, O.; Méjean, A.; Marquet, A. *FEBS Lett.* **1998**, *440*, 226–230. (b) Marquet, A.; Florentin, D.; Ploux, O.; Bui, B. T. S. *J. Phys. Org. Chem.* **1998**, *11*, 529–535. (c) Méjean, A.; Bui, B. T. S.; Florentin, D.; Ploux, O.; Izumi, Y.; Marquet, A. *Biochem. Biophys. Res. Commun.* **1995**, *217*, 1231–1237.
6. Nudelman, A.; Nudelman, A.; Marcovici-Mizrahi, D.; Flint, D. *Bioorg. Chem.* **1998**, *26*, 157–168.
7. Zervas, L.; Photaki, I.; Ghelis, N. *J. Am. Chem. Soc.* **1963**, *85*, 1337–1341.
8. Conti, S.; Cossu, S.; Giacomelli, G.; Falorni, M. *Tetrahedron* **1994**, *50*, 13493–13500.
9. Nudelman, A. PhD Dissertation, Bar Ilan University, 1996, unpublished results..
10. (a) Hanessian, S.; Sahoo, S. P. *Tetrahedron Lett.* **1984**, *25*, 1425–1428. (b) Barton, D. H. R.; Crich, D.; Hervé, Y.; Potier, P.; Thierry, J. *Tetrahedron* **1985**, *25*, 4347–4357. (c) Afzali-Ardakani, A.; Rapoport, H. *J. Org. Chem.* **1980**, *45*, 4817–4820. (d) Pellicciari, R.; Natalini, B.; Marinozzi, M. L-Vinylglycine form L-homoserine; *Synth. Commun.* **1988**, *18*, 1715–1721. (e) Ardakani, A.; Rapoport, H. L-Vinylglycine. *J. Org. Chem.* **1980**, *45*, 4817–4820. (f) Pellicciari, R.; Natalini, B.; Marinozzi, M. *Synth. Commun.* **1988**, *18*, 1715–1721. (g) Carrasco, M.; Jones, R. J.; Kamel, S.; Rapoport, H.; Thien, T. *Org. Synth.* **1992**, *70*, 29–34.
11. Mansour, T. S.; Evans, C. A. *Synth. Commun.* **1990**, *20*, 773–781.
12. Meffre, P.; Vo-Quang, L.; Vo-Quang, Y.; Le-Goffic, F. *Synth. Commun.* **1989**, *19*, 3457–3468.
13. Buckley, T. F.; Rapoport, H. *J. Am. Chem. Soc.* **1981**, *103*, 6157–6163.
14. Somerville, C. R.; Ogren, W. L. *Isolation of photorespiration mutants in arabidopsis thaliana. In Methods in chloroplast molecular biology*; Elsevier: London, 1982.
15. Nudelman, A.; Patchornick, A.; Karoly-Hafely, E.; Braun, F.; Erickson, R. C. *J. Antibiot.* **1980**, *33*, 76–82.
16. Einhorn, J.; Einhorn, C.; Luche, J. L. *Synlett* **1991**, 37–38.
17. Ferraro, *J. Biochem. Prep.* **1971**, *13*, 39–41.
18. Hashimoto, M.; Aoyama, T.; Shioiri, T. *Chem. Pharm. Bull.* **1981**, *29*, 1475–1478.
19. Tauchi, M.; Matsumura, I. *Jpn. Kokai Tokkyo Koho*. JP07188133, JP07188203, **1995**..
20. Nudelman, A.; Bechor, Y.; Falb, E.; Fischer, B.; Wexler, B. A.; Nudelman, A. *Synth. Commun.* **1998**, *28*, 471–474.
21. Fonken, G. S.; Johnson, W. S. *J. Am. Chem. Soc.* **1952**, *74*, 831–833.
22. Harada, K.; Shiono, S. *Bull. Chem. Soc. Jpn* **1984**, *18*, 777–782.
23. Borch, R. F.; Bernstein, M. D.; Dupont, D. H. *J. Am. Chem. Soc.* **1971**, *93*, 2897–2904.
24. Wright, Jr. W. B. *J. Heterocyl. Chem.* **1965**, *2*, 42–43.
25. Falb, E.; Nudelman, A.; Hassner, A. *Synth. Commun.* **1993**, *23*, 2839–2844.
26. (a) Green, M. R. GB Patent GB2114886, **1983**.. (b) Christow, C. P. *Hildesheim, Ger. Ernst Rodenwalt Arch.* **1976**, *3*, 47–50. (c) Balikhina, V. N.; Zav'yalov, S. I.; Rodionova, I. Z.; Gracheva, E. P.; Filippov, V. V. *Uchenye Zapiski—Vladimirskii Gosudarstvennyi Pedagogicheskii Institut imeni P. I. Lebedeva-Polyanskogo, Seriya Fiziologiya Rastenii* **1971**, *31*, 49–55.
27. (a) Andreev, L. N. *Abh. Akad. Wiss. DDR, Abt. Math.; Naturwiss.; Tech* **1983**, 341–344. (b) Trunenkov, I. P.; Fabrichnyi, B. P.; Shalavina, I. F.; Kostrova, S. M.; Gol'dfarb,

Ya. L.; Filippov, V. V. *Uch. Zap.; Vladimirskii Gos. Pedagog. Inst.; Ser. Fiziol. Rast.* **1971**, *31*, 65–71. (c) Balkihina, V. N.; Fabrichnyi, B. P.; Shalavina, I. F.; Kostrova, S. M.; Gol'dfarb, Ya. L.; Filippov, V. V. *Uch. Zap.; Vladimirskii Gos. Pedagog. Inst.; Ser. Fiziol. Rast.* **1971**, *31*, 56–60. (d) Fabrichnyi, B. P.; Shalavina, I. F.; Kostrova, S. M.; Gol'dfarb, Ya. L.; Filippov, V. V. *Zh. Org. Khim.* **1970**, *65*, 1091–1100.

28. Miyashita, N.; Yoshikoshi, A.; Grieco, P. A. *J. Org. Chem.* **1977**, *42*, 3772–3774.
29. Tauchi, M.; Matsumura, I. *Jpn. Kokai Tokkyo Koho.* JP07188133, JP07188203, **1995**..
30. Balikhina, V. N.; Zav'yalov, S. I.; Rodionova, I. Z.; Gracheva, E. P.; Filippov, V. V. *Uch. Zap. Vladimirskii Gos. Pedagog. Inst.; Ser. Fiziol. Rast.* **1971**, *31*, 49–55.
31. (a) Rendina, A. R.; Taylor, W. S.; Gibson, K.; Lorimer, G.; Rayner, D.; Lockett, B.; Kranis, K.; Wexler, B.; Marcovici-Mizrahi, D.; Nudelman, A.; Nudelman, A.; Marsilii, E.; Chi, H.; Wawrzak, Z.; Calabrese, J.; Huang, W.; Jia, J.; Schneider, G.; Lindqvist, Y.; Yang, G. *Pesticide Sci.* **1999**, *55*, 236–247. (b) Warshawsky, A.; Wilchek, M.; Altma, J.; Shoef, N. US Patent US4652519, **1987**.