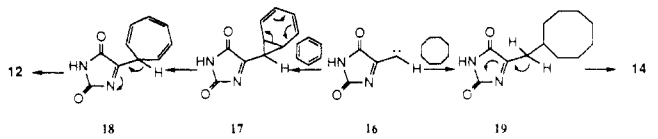
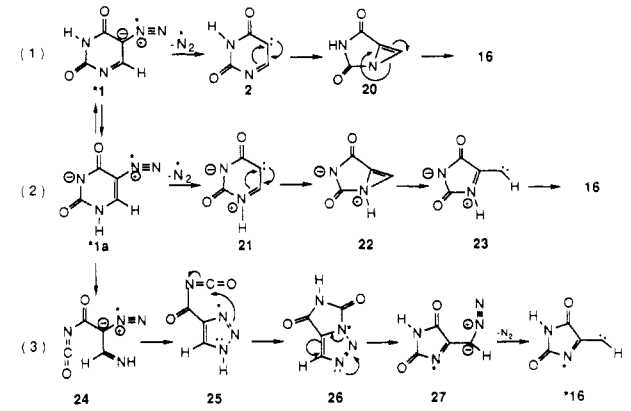


Three mechanistic routes to **16** from **1** are considered as illustrated below. Mechanism 1 involves loss of N_2 from **1** and rearrangement of carbene **2** via 1*H*-azirene **20**. This mechanism, however, fails to explain why a methyl group instead of hydrogen at N-3 changes the chemistry of **1** and **3** so spectacularly. Further, decomposition of **1** in benzene in the presence of triethylamine yields **13** instead of **12**. A more attractive possibility, mechanism 2, which is not available to **3**, results from tautomerism of **1** to **1a** and avoids **20**.[7] Expulsion of N_2 then leads to **16** via azirenium dipolar ion **22**. A much different and more complicated process, mechanism 3, is ring opening of **1a** to carbonyl isocyanate **24** which converts to 4-carbonyl-1*H*-1,2,3-triazoleisocyanate (**25**) and then to triazole **26** by cyclization and proton transfer. Collapse of **26** then gives 4-(diazomethyl)-1*H*-imidazole-2,4-dione (**27**), which loses N_2 to yield carbene **16**. An important difference in the three mechanisms is that, with *1 containing the (*) nitrogen labeled with ^{15}N, in mechanism 3 the labeled nitrogen is incorporated in carbene **16** and thus in products **12** and **14**, whereas in mechanisms 1 and 2 the products will *not* contain the isotopic nitrogen as it is lost earlier as $*N_2$.



(7) (a) 1*H*-azirenes are antiaromatic (4π electron) and usually thermally unstable. When substituted by electron-withdrawing substituents, their stabilities are increased.[7b] (b) Regitz, M.; Arnold, B.; Davidson, D.; Schubert, H.; Fusser, G. *Bull. Soc. Chim. Belg.* **1981**, *90*, 615.

Studies of the thermal reactions of *1 with benzene and cyclooctane are now summarized.

Synthesis of *1 was accomplished by: (1) nitration of uracil with 1:1 molar mixture of $H^{15}NO_3$ and fuming nitric acid in sulfuric acid at 100 °C, (2) reduction of the 5-[^{15}N]nitrouracil with ammoniacal sodium dithionite,[8a] and (3) diazotization of the resulting 5-[^{15}N]aminouracil with sodium nitrite and aqueous HCl.[8b] Mass spectral analysis (M^+ peak) reveals that the diazo group in *1 is 45 ± 2% enriched. Decompositions of the *1 in benzene and in cyclooctane at 150–160 °C yield products identical with **12** and **14**, respectively. Most importantly, the ^{15}N NMR spectrum of **12** exhibits a single resonance at 108.7 ppm, and the *E* and *Z* isomers of **14** display ^{15}N NMR peaks at 107.2 and 108.5 ppm in a 5:1 ratio. These resonances are typical of amide nitrogens[9] and indicate that the ***12** and ***14** contain ^{15}N at N-1 of their imidazolidinedione moieties. Further, mass spectral analyses of the ***12** and ***14** reveal that their ^{15}N contents are identical with that of initial *1. Formation of *12 and *14 is therefore consistent with mechanism 3 for isomerization of **1** via carbonyl isocyanates **24** and **25** to carbene ***16** and excludes any partial involvements of mechanisms 1 and 2. These labeling experiments also eliminate the possibility of formation of **12** from carbene **2** and rearrangement of spirocycloheptatriene **10**.

Investigation is being made of (1) detection and isolation of **25** and **26**,[10,11] (2) synthesis and the chemistry of **27**, and (3) the behavior of **1** and **3** in the presence of tertiary amines and various catalysts.

Acknowledgment. We thank the National Cancer Institute (Grant 5 R01 CA11185) for financial support of this research.

Supplementary Material Available: X-ray data for **12** (9 pages); structure factor listings (13 pages). Ordering information is given on any current masthead page.

(8) (a) Bogart, M. T.; Davidson, D. *J. Am. Chem. Soc.* **1933**, *55*, 1667. (b) Thurber, T. C.; Townsend, L. B. *J. Heterocycl. Chem.* **1972**, *9*, 629.

(9) Levy, G. C.; Lichter, R. L. *Nitrogen-15 Nuclear Magnetic Resonance Spectroscopy*; John Wiley & Sons: New York, 1979; p 58.

(10) (a) Hydrates of 5-diazouracils having hydrogen at N-3 hydrolyze to 1,2,3-triazole-4-carboxamides when heated. Such transformations are not observed with 5-diazo-3-methyluracils.[10b,c] (b) Thurber, T. C.; Townsend, L. B. *J. Heterocycl. Chem.* **1973**, *95*, 3081. (c) Thurber, T. C.; Townsend, L. B. *J. Org. Chem.* **1976**, *41*, 1041. (d) The different behavior of the above diazouracils might have their origins in that 5-diazo-3-methyluracils can not open to carbonyl isocyanates such as **24**.

(11) 1,2,3-Triazole-4-carboxamide, as presumably derived by hydrolysis of **25** and loss of carbon dioxide, has now been found upon thermolysis and workup of **1** in benzene.

Novel Modifications of Peptides: Simple Syntheses of Difunctionalized Enamines, Enol Ethers, and Thioenol Ethers from Carboxylic Acids via Acylimidazole and Enol Phosphate Intermediates[1]

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Summary: The condensation of carboxylic acids **5** (N-protected amino acids or dipeptides) and active methylene compounds to give difunctionalized enols **6** is accomplished by the use of 1,1′-carbonyldiimidazole as an activating reagent. Enamines **7a**, enol ethers **7b**, and thioenol ethers **7c** are obtained directly from the corresponding nucleophiles and the intermediates formed from enols **6** by the action of phenyl phosphorodichloridate. The integrity of peptide chiral centers is maintained during these transformations.

(1) Presented in part at the 72nd Canadian Chemical Conference (Organic Division) Victoria, Canada, June 1989.

(2) Fellow of the NSERC-Canada, 1985–.

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In an effort to gain better understanding of the mechanisms of enzyme–substrate interactions, several modifications[3] of the carboxylic function of protease substrates have been reported. Some of these modified analogues, including compounds **1b**, **1c**, and **1d**, have proved to be more effective substrates[4] than peptides with the usual amide function (**1a**). Others are known to be inhibitors[5] (e.g. **1e**). Recently, we reported convenient syntheses of novel backbone-modified peptides[6] in which mono- and difunctionalized enamines **2a** and enols **2b** (E, E′ = electron-withdrawing groups) have been introduced using thioamides as starting materials. This creates an electrophilic center that can interact with nucleophilic groups at the active site of enzymes such as serine and cysteine proteases. Moreover, the electron density of the electrophilic center can be modulated by varying the nature of the electron-withdrawing groups.

We are now interested in new vinylic derivatives **2c–e** in which the oxygen atom in **1a–d** is replaced by disubstituted carbon, and which includes substituents X possessing different degrees of lability. The impact of the introduced substituent, which may act as a leaving group or be involved in hydrogen bonding, could result in enhanced enzyme–analogue interactions. Although enols **2b** can be obtained from enamines **2a** by hydrolysis, in this communication we describe a general, short synthesis of difunctionalized enols[7] **6** directly from carboxylic acid **5**. The procedure involves the condensation of an appropriate active methylene compound with the activated acylimidazole[8] intermediate **3**. For example, treatment of the Boc-phenylalanine (**5** (R = Boc)) in THF with 1,1′-carbonyldiimidazole (1.1 equiv) at 0 °C, and then warming the mixture to room temperature for 1 h, provided the activated species **3** (R = Boc). The solution was then cooled to −78 °C prior to the addition of the appropriate carbanion (1.05 equiv; generated from sodium hydride and active methylene compound at 0 °C) to give enol **6** (R = Boc) in high yield (Table I, **6a–c**). In this way, several dipeptide enols[9] **6d–j** (R = N-Ac-Leu; chymotrypsin substrate analogues) were also successfully prepared from the corresponding carboxylic acid dipeptide **5** (R = N-Ac-Leu) in yields ranging from 47% to 96%. As described before,[6] analysis of the ^{1}H NMR spectrum of the dipeptidic enol **6g** did not show the presence of the other diastereoisomer (*N*-Ac-L-Leu-D-Phe synthesized for comparison) providing evidence that racemization does not occur under these experimental conditions.

Scheme I

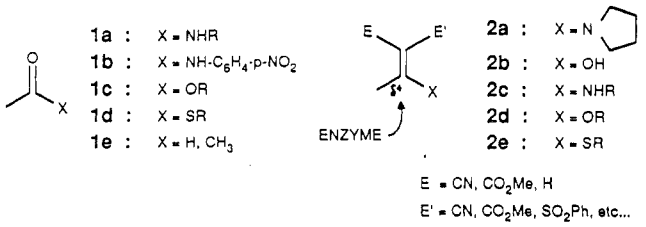


Table I. Preparation of Enols 6

structure	R	E	E′	yield, %
6a	Boc	CN	CN	97
6b	Boc	CN	CO_2Me	93
6c	Boc	CN	C_6H_4-*p*-NO_2	81
6d	*N*-Ac-Leu	CN	C_6H_4-*p*-NO_2	63
6e	*N*-Ac-Leu	CN	$P(O)(OEt)_2$	47
6f	*N*-Ac-Leu	CN	CN	96
6g	*N*-Ac-Leu	CN	CO_2Me	55
6h	*N*-Ac-Leu	CN	2-pyridyl	59
6i	*N*-Ac-Leu	CO_2Me	SO_2-C_6H_5	72
6j	*N*-Ac-Leu	CO_2Me	$P(O)(OMe)_2$	66

The enol function of **6** was further elaborated to give enamines **7a**, enol ethers **7b**, and thioenol ethers **7c**. This allows introduction of various substituents X (**7a–c**, Table II) in the vinyl function with different steric and electronic properties in the isosteric peptide bond. Phenyl phosphorodichloridate[10] was found to be an effective enol activating agent for these transformations, while other well-known reagents[11] proved to be unsuccessful under our conditions. Treatment of enol **6** (R = Boc; E, E′ = CN) in THF with triazole[12] (5 equiv) and phenyl phosphorodichloridate (1.2 equiv) at 0 °C, followed by warming to room temperature for 3 h, led to intermediate **4** (R = Boc; E, E′ = CN). Addition of monosubstituted amines then afforded enamines **7a** in fairly good yields (80% to 57%, Table II). Even disubstituted amines can be used as nucleophiles, although the yields are lower (35%, **7a**, entry 5).

Interestingly, enamine **7a** is obtained (79%; Table II, entry 6) when the methyl ester of alanine is employed as the nucleophile. This product may be viewed as a novel dipeptide structure in which an endo amide bond has been replaced by a difunctionalized enamine group. Similarly, enol and thioenol ether derivatives (**7b** and **7c**, Table II) were also prepared in high to moderate yields. Alcohols and thiols are added in anionic form to the activated enol using sodium hydride or potassium *tert*-butoxide as the base (2.5 equiv) in the presence of 18-crown-6[13] (1.5 equiv) in THF to generate the naked anion. Yields of thioenol ethers are generally higher than those of enol ethers, probably due to the higher nucleophilicity of thiols. The unsubstituted (N and S) enamine and thioenol (**7a**, **7c**, entry 1) were obtained by using ammonia and hydrogen sulfide as nucleophiles. Thioenol ether **7c** (Table II, entry

(3) Spatola, A. F. *Chemistry and Biochemistry of Amino Acids, Peptides and Proteins*; Weinstein B., Ed.; Dekker: New York 1983; Vol. 7, p 263.

(4) Harper, J. W.; Cook, R. R.; Roberts, C. J.; McLaughlin, B. J.; Powers, J. C. *Biochemistry* **1984**, *23*, 2995–3002.

(5) Imperiali, B.; Abeles, R. H. *Biochemistry* **1986**, *25*, 3760–3767.

(6) (a) Sauvé, G.; Mansour, T. S.; Lachance, P.; Belleau, B. *Tetrahedron Lett.* **1988**, *29*, 2295–2299. (b) Sauvé, G.; Le Berre N.; Zacharie, B. *Tetrahedron Lett.* **1988**, *29*, 2299–2302.

(7) Some conditions for the preparation of (a) difunctionalized enols: Nonhebel, D. C. *Tetrahedron* **1970**, *26*, 4443–4447. (b) Difunctionalized enamines: Dornow, A.; Schleese, E. *Chem. Ber.* **1958**, *91*, 1830–1840. (c) Difunctionalized enol ethers: Hayashi, T.; Hori, I.; Baba, H.; Midorikawa, H. *J. Org. Chem.* **1965**, *30*, 695–699. (d) Difunctionalized thioenols and thioenol ethers: Hartke, K.; Gölz, G. *Chem. Ber.* **1973**, *106*, 2353–2360. Rappoport, Z.; Gazit, A. *J. Am. Chem. Soc.* **1987**, *109*, 6698–6710.

(8) (a) Maibaum, J.; Rich, D. H. *J. Org. Chem.* **1988**, *53*, 869–873. (b) Harris, B. D.; Bhat, K. L.; Joullie, M. M. *Tetrahedron Lett.* **1987**, *28*, 2837–2840. (c) Ono, N.; Fujii, M.; Kaji, A. *Synthesis* **1987**, 532–535.

(9) The energy barriers for rotation around the carbon–carbon double bond are low for β,β′-difunctionalized enol groups. The interconversion between two isomeric forms of nonsymmetric (E and E′) enol function has been observed from variable-temperature NMR studies using different solvents. Except for the case of possible hydrogen-bonded enol hydroxyl group (e.g. **6b**, **6g**), which revealed collapse upon heating from room temperature to 100 °C in DMSO-d_6, we only observed one set of signals for both *E* and *Z* isomers that corresponds to rapid equilibriums at room temperature. For instance, enol **6c** exhibited two sets of relevant signals when cooled to −55 °C in acetone-d_6. The same conclusions are obtained for enamines **9** and **10** from identical studies.

(10) (a) Liu, H.-J.; Sabesan, S.-I. *Can. J. Chem.* **1980**, *58*, 2645–2648. (b) Ireland, R. E.; Norbeck, D. W.; Mandel, G. S.; Mandel, N. S. *J. Am. Chem. Soc.* **1985**, *107*, 3285–3294.

(11) For a review of activating reagents for the carboxylic acid function, see: Bodanszky M. *Principles of Peptides Syntheses*; Springer-Verlag: Berlin, New York, 1984; Vol. 1, pp 9–52. Other activating reagents surveyed were not effective. Among those tried were CDI, BOP, $ClCO_2Me$–pyridine, DCC–HOBt, C_6H_5COCl–pyridine, PPh_3–CCl_4, PPh_3–DEAD, $SOCl_2$–pyridine.

(12) The yields are profoundly influenced by the conditions used, including the nature and quantity of base, solvent, and activation time.

(13) Potassium *tert*-butoxide and 18-crown-6 are necessary for obtaining moderate yields using thiols; sodium hydride is used as the base in the case of alcohol introduction.

Scheme II

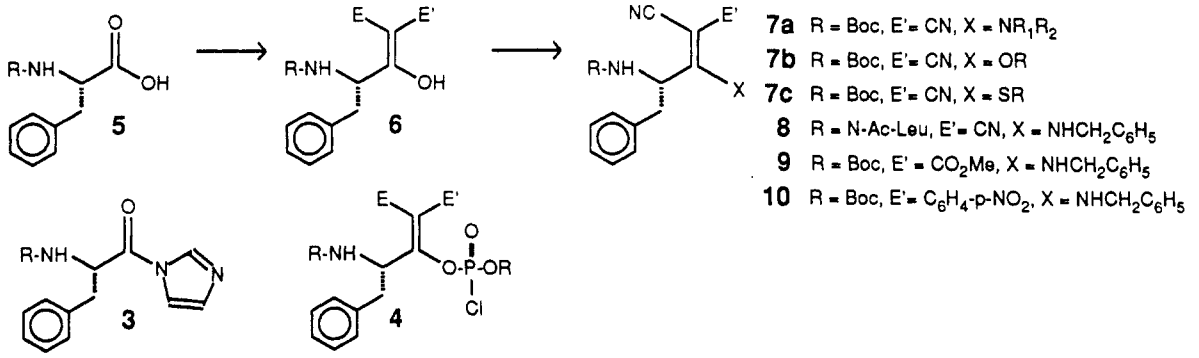
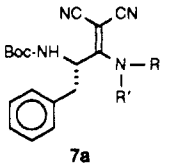
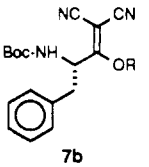


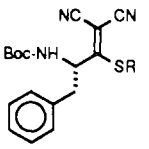
Table II. Preparation of Enamines 7a, Enol Ethers 7b, and Thioenol Ethers 7c



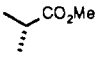
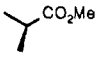
7a



7b



7c

entry	R'	R	yield, %	R	yield, %	R	yield, %
1	H	H	73			H	34
2	H	Ph	80	Ph	45	Ph	70
3	H	Bn	62	Me	41	Et	82
4	H	*i*-Pr	57	*i*-Pr	15	*i*-Pr	49
5		$-(CH_2)_4-$	35	*t*-Bu	0	*t*-Bu	15
6	H		79				45

6) is obtained with methyl mercaptopropionate[14] as the nucleophile. This methodology has been applied on *N*-Ac-leucylphenylalanine dicyano enol **6f**, a chymotrypsin substrate analogue, and to other Boc-phenylalanine functionalized enols. For instance, **6f**, **6b**, and **6c** afforded the corresponding enamines **8**, **9**, and **10** in 25%, 35%, and 40% yields, respectively, under similar reaction conditions with benzylamine as the nucleophile. The ^1H NMR spectrum of dipeptide enamine **7a** (Table II, entry 6) did not show the presence of the other diastereomer formed by racemization, indicating that optical integrity is preserved during the reaction.

This is the first time that difunctionalized enamines, enol ethers, and thioenol ethers have been reported as modifications for the peptide amide function, both at endo and C-terminal positions. As a result, effective electron-withdrawing functionalization has now been performed on α-chymotrypsin dipeptide substrates. Furthermore, the enol hydroxyl function has been elaborated through the use of phenyl phosphorodichloridate as an activating reagent. Importantly, the chiral integrity of peptides is maintained in these reactions. This methodology allows easy introduction of functionalities at the three positions of the vinyl isosteric peptide bond with several electron-withdrawing groups and different leaving groups. We are currently applying these transformations to peptidic enzyme substrates that may serve as useful probes for studying analogue–enzyme interactions. Full experimental details and the results of enzymatic studies using these analogues will be reported elsewhere.

(14) Preparation of methyl (*R*)-2-mercaptopropionate: Strijtveen, B.; Kellogg, R. M. *J. Org. Chem.* **1986**, *51*, 3664-3671.

Acknowledgment. This work was supported by the Natural Sciences and Engineering Research Council of Canada and the Fonds FCAR (Québec). N.L.B. is the recipient of a Fonds FCAR graduate fellowship and B. Z. is the recipient of a J.-L. Lévesque (I.A.F.) postdoctoral fellowship.

Supplementary Material Available: Experimental procedure and characterization data (^1H NMR spectra) for all products; ^{13}C NMR, IR, optical rotations, elemental analysis, and mass spectra for several products (15 pages). Ordering information is given on any current masthead page.

Total Synthesis of (±)-Isovelleral, a Mutagenic Sesquiterpene Dialdehyde from *Lactarius vellereus*

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Received March 7, 1990

Summary: The mutagenic sesquiterpene dialdehyde (±)-isovelleral, a component in the chemical defense mechanism of many basidiomycetes, has been prepared by the total synthesis outlined in the scheme.

Basidiomycetes of several genera, including *Lactarius* and *Russula*, have a complicated chemical defense mechanism in which fatty acid esters of the unstable sesquiterpene hemiacetal velutinal (1) appear to function as the

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