δ 1.98 (dd, 1 H, OH), 3.32–3.72 (4 m, 5 H); MS (FD) *m/e* 413 (M + 1)$^+$, 381 (M − CH_2OH)$^+$. Anal. ($C_{27}H_{56}O_2$) C, H.

***rac*-(*Z*,*Z*)-(2-Ethoxy-10,13-nonadecadien-1-yl)phosphocholine (22).** The above alcohol **19** (3.92 g, 12 mmol) was transformed to the desired phospholipid **22** (1.80 g, 30.5%) by the previously described procedure. As in the previous instance, this material was stable in an O_2-free/anhydrous environment: NMR ($CDCl_3$) δ 2.0 (m, 4 H), 2.75 (m, 2 H), 3.35 (s, 9 H), 4.3 (m, 2 H), 5.3 (m, 4, vinyl H); MS (FAB) *m/e* 490 (M + 1)$^+$, 184. M_r for $C_{26}H_{53}NO_5P$: calcd, 490.3662; found, 490.3660.

***rac*-(2-Ethoxypentacos-1-yl)phosphocholine (23).** By the above procedure, alcohol **20** (0.206 g, 0.5 mmol) was converted to the desired product **23**: 0.175 g (60%); MS (FAB) *m/e* 578 (M)$^+$. Anal. ($C_{32}H_{68}NO_5P$) C, H, N.

Acknowledgment. We thank John Shain for preparation of the precursor to **23**, Sharon Rinzel for the in vitro cytotoxicity results, and Dr. Ken Kurz and his colleagues for blood pressure studies. We are also grateful to Gerry Wells for typing this paper.

Registry No. **5**, 104532-20-9; **6**, 104532-21-0; **7**, 104548-62-1; **8**, 104532-22-1; **9**, 104532-23-2; **10**, 104532-24-3; **11**, 104532-25-4; **12**, 104548-80-3; **13**, 104532-26-5; **14**, 104532-27-6; **15**, 104532-28-7; **15** (ethyl ether), 104532-34-5; **16**, 104532-29-8; **17**, 104532-30-1; **18**, 104532-31-2; **18** (acetate), 104532-35-6; **19**, 104532-32-3; **20**, 104532-33-4; **21**, 104548-81-4; **22**, 104548-82-5; choline tosylate, 55357-38-5; oleyl aldehyde, 2423-10-1; linoleyl aldehyde, 2541-61-9; 1-tetracosanal, 57866-08-7.

Chemically Stable Homocinnamyl Analogues of the Leukotrienes: Synthesis and Preliminary Biological Evaluation[1]

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The synthesis and biological characterization of a series of stable leukotriene analogues (**2**) are reported. They are derivatives of (5*S*,6*R*,7*Z*)-6-peptidyl-5-hydroxy-9-phenyl-7-nonenoic acid, in which the phenyl group is variously substituted with a heptanyl, 2-heptenyl, or hexanyloxy chain (R^1) and the peptide is either glutathionyl, cysteinylglycinyl, or cysteinyl. The most potent agonist is (5*S*,6*R*,7*Z*)-6-*S*-glutathionyl-5-hydroxy-9-(4-heptanylphenyl)-7-nonenoic acid. This analogue has an EC_{50} value of 74.5 nM, in the presence of 1-serine borate (45 mM), on guinea pig tracheal spirals. The agonist activity of the cysteinylglycinyl- and the cysteinyl-substituted analogues was inhibited by FPL-55712. Three of the analogues were weak leukotriene antagonists in vitro on guinea pig tracheal spirals. The most potent of these was (5*S*,6*R*,7*Z*)-6-*S*-cysteinyl-5-hydroxy-9-(2-heptanylphenyl)-7-nonenoic acid. At 10 μM, this analogue inhibited by 28% the contraction induced by 8 nM LTE_4.

Reported herein is a series of chemically stable, biologically active analogues of the peptidoleukotrienes **1**.[1,2] The leukotrienes are a class of potent spasmogenic agents,[3] which make up the active components of the "Slow Reacting Substance of Anaphylaxis" (SRS-A) first discovered by Kellaway over 40 years ago.[4] The early research on SRS-A was hampered because its structure was unknown. Key factors delaying its structural elucidation were the limited availability of crude SRS-A (leukotrienes) from biological sources and its intrinsic instability.[5] The elucidation of the leukotriene structures by Corey and Samuelsson prompted a resurgence of interest in these mediators,[6] which are believed to play a significant role in the mediation of asthma and other diseases. Recently, reports of clinical studies on the effects of pure leukotrienes in man have shown them to be bronchospastic and cause an asthmatic-like response in normal individuals.[7]

Since our team's primary goal is the development of clinically useful antiasthma drugs, we decided to explore the possible utility of leukotriene antagonists as such drugs. A classical approach to the synthesis of antagonists is via structural modification of the corresponding agonist.[8] In the case of the leukotrienes, we felt that their inherent chemical instability would preclude the development of drugs that retained those structural elements responsible for instability. As a first step, it was necessary to develop relatively stable analogues that interacted with the leukotriene receptors. Ideally these would be antagonists. If, however, they were potent leukotriene receptor agonists, we were confident that they would serve as a new starting

[†] Department of Medicinal Chemistry.
[‡] Department of Pharmacology.

(1) A preliminary account of this work was presented at the 1984 International Chemical Congress of Pacific Basin Societies, Honolulu, HI; Bernstein, P. R.; Adams, E. J.; Maduskuie, T. P.; Snyder, D. W.; Vacek, E. P.; Willard, A. K.; Abstract 10E0098.
(2) For recent reports of other examples of leukotriene antagonists based upon the structure of the leukotrienes, see the following: (a) Perchonock, C. D.; McCarthy, M. E.; Erhard, K. F.; Gleason, J. G.; Wasserman, M. A.; Muccitelli, R. M.; DeVan, J. F.; Tucker, S. S.; Vickery, L. M.; Kirchner, T.; Weichman, B. M.; Mong, S.; Crooke, S. T.; Newton, J. F. *J. Med. Chem.* **1985**, *28*, 1145. (b) Perchonock, C. D.; Uzinskas, I.; Ku, T. N.; McCarthy, M. E.; Bondinell, W. E.; Volpe, B. W.; Gleason, J. G.; Weichman, B. M.; Muccitelli, R. M.; DeVan, J. F.; Tucker, S. S.; Vickery, L. M.; Wasserman, M. A. *Prostaglandins* **1985**, *29*, 75. (c) Saksena, A. K.; Green, M. J.; Mangiaracina, P.; Wong, J. K.; Kreutner, W.; Gulbenkian, A. R. *Tetrahedron Lett.* **1985**, *26*, 6427.
(3) For a review on the leukotrienes, see the following: *The Leukotrienes, Chemistry and Biology*; Chakrin, L. W., Bailey, D. M., Eds.; Academic: New York, 1984.
(4) Kellaway, C. H.; Trethewie, W. R. *Q. J. Exp. Physiol. Cogn. Med. Sci.* **1940**, *30*, 121.
(5) *SRS-A and the Leukotrienes*; Piper, P. J., Ed.; Wiley: New York, 1981.
(6) Corey, E. J.; Clark, D. A.; Goto, G.; Marfat, A.; Mioskowski, C.; Samuelsson, B.; Hammarstrom, S. *J. Am. Chem. Soc.* **1980**, *102*, 1436.
(7) Smith, L. J.; Greenberger, P. A.; Patterson, R.; Krell, R. D.; Bernstein, P. R. *Am. Rev. Respir. Dis.* **1985**, *131*, 368.
(8) Craig, P. N. In *Burger Medicinal Chemistry*, 4th ed.; Wolff, M. E., Ed.; Wiley: New York, 1980; Part 1, pp 331–348.

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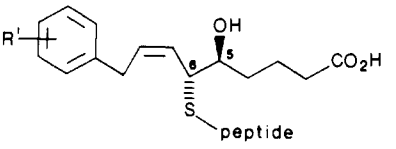
Table I. Leukotriene Analogues[a]

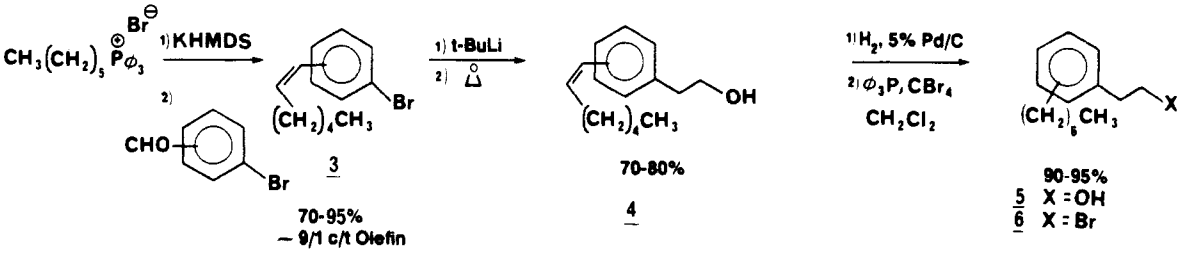


2

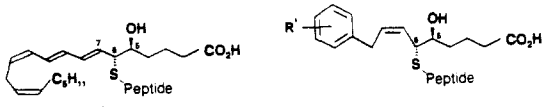
2	R^1	position of R^1	S-peptide	% yield[b]	mp, °C	formula[c]
a	C_7H_{15}	para	glutathionyl	95	193–200 dec	$C_{32}H_{46}N_3O_9SK_3 \cdot 3H_2O$
b		para	cysteinylglycyl	50	168–183 dec	$C_{27}H_{40}N_2O_6SK_2 \cdot H_2CO_3$
c		para	cysteinyl	46	>225	$C_{25}H_{37}NO_5SK_2 \cdot H_2O$
d		meta	glutathionyl	68	197–199 dec	$C_{32}H_{46}N_3O_9SK_3$
e		meta	cysteinylglycyl	63	183–186 dec	$C_{27}H_{40}N_2O_6SK_2 \cdot H_2CO_3$
f		meta	cysteinyl	40	125–130	$C_{25}H_{37}NO_5SK_2$
g		ortho	glutathionyl	54	180 dec	$C_{32}H_{46}N_3O_9SK_3 \cdot 2H_2O$
h		ortho	cysteinylglycyl	83	110 dec	$C_{27}H_{40}N_2O_6SK_2 \cdot H_2CO_3 \cdot H_2O$
i		ortho	cysteinyl	63	198–200	$C_{25}H_{37}NO_5SK_2$
j	$C_6H_{13}O$	para	glutathionyl	88	205 dec	$C_{31}H_{44}N_3O_{10}SK_3 \cdot H_2O$
k		para	cysteinylglycyl	78	198–204 dec	$C_{26}H_{38}N_2O_7SK_2 \cdot H_2CO_3 \cdot H_2O$
l		meta	cysteinylglycyl	70	190–194	$C_{26}H_{38}N_2O_7SK_2 \cdot H_2CO_3 \cdot 2H_2O$
m		ortho	cysteinylglycyl	60	149–153	$C_{26}H_{38}N_2O_7SK_2 \cdot H_2CO_3 \cdot 4H_2O$
n	$C_5H_{11}CH{=}CH$	para	glutathionyl	68	195–201	$C_{32}H_{44}N_3O_9SK_3 \cdot H_2O$
o		meta	cysteinylglycyl	78	183–188	$C_{27}H_{38}N_2O_6SK_2 \cdot H_2CO_3$

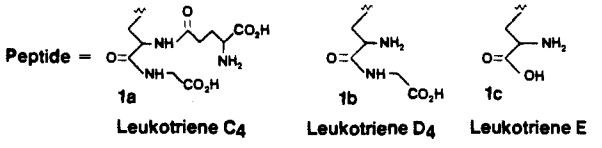
[a] The detailed experimentals for compounds 2a–c are given in the Experimental Section. Partial spectra for 2d–n and the intermediates leading to them are given in the microfilm edition of this journal. [b] Final hydrolysis step. [c] Satisfactory C, H, N analyses were obtained for all compounds. All of the cysteinylglycyl-substituted analogues analyzed for bicarbonate salt formation.

Scheme I



point for further modifications that would lead to antagonists.[9]

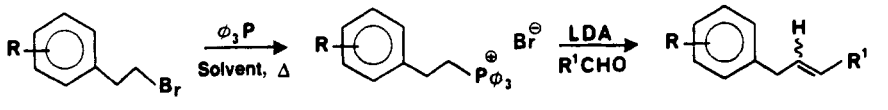




The analogues 2 (see Table I) are homocinnamyl derivatives. Their design was based on a number of observations made by us and others concerning the structure–activity relationships of the leukotrienes.[3] First, we had shown that the "unnatural" 5,6 diastereomers of leukotriene D_4 (LTD_4) were weak agonists, having $^1/_{100}$ to $^1/_{1000}$ the potency of LTD_4.[10] Since this had demonstrated the critical nature of configuration at the 5-hydroxyl and 6-peptide positions for agonist activity, we felt that the 5*S*,6*R* stereochemistry was important. More recently, however, it has been demonstrated in a series of 2-nor leukotriene analogues that 4*R*,5*S* stereochemistry affords antagonist activity.[11] Second, our evaluation of the ^1H NMR of LTD_4 revealed a preferred solution conformation, in which the triene region is planar.[12] As a mimic of this planar, electron-rich region, we chose to utilize a benzene ring. Preliminary studies in our laboratories have shown that leukotriene D_4 analogues, using either a nonsubstituted phenyl or cinnamyl group as the triene replacement, were devoid of all biological activity.[13] These compounds were tested for both agonist and antagonist (against 8 nM LTE_4) activity at 10^{-5} M. Furthermore, these studies also demonstrated that whereas substitution with an alkyl chain had no effect on the lack of activity of the directly linked phenyl series, very weak agonist activity was seen at 10^{-5} M with an alkyl-substituted cinnamyl analogue.[14] However, the cinnamyl analogue is also relatively unstable, presumably since it is a styrene derivative. At about this time, Drazen et al. reported that in a series of LTD_4

(9) Our progress is exemplified by the three antagonists reported in this paper. A future paper by us (manuscript in preparation) will report on a series of non-peptidyl-containing analogues with enhanced antagonist activity.
(10) Tsai, B. S.; Bernstein, P.; Macia, R. A.; Conaty, J.; Krell, R. D. *Prostaglandins* **1982**, *23*, 489.
(11) Weichman, B. M.; Wasserman, M. A.; Holden, D. A.; Osborn, R. R.; Woodward, D. F.; Ku, T. W.; Gleason, J. G. *J. Pharmacol. Exp. Ther.* **1983**, *227*, 700.
(12) Loftus, P.; Bernstein, P. R. *J. Org. Chem.* **1983**, *48*, 40.
(13) Unpublished results of Drs. A. K. Willard and B. S. Tsai, these laboratories.
(14) The specific compound referred to is (5*SR*,6*RS*,7*E*)-6-(*S*-cysteinylglycinyl)-5-hydroxy-8-[4-[1(*Z*),4(*Z*)-decadien-1-yl]-phenyl]-7-octenoic acid. This was tested as an unstable mixture of diastereomers, and it showed significant agonist activity (increase in basal tension) at 10^{-5} M on guinea pig tracheal spirals (see ref 13).

Scheme II



6 R = $CH_3(CH_2)_6$–
19 R = H

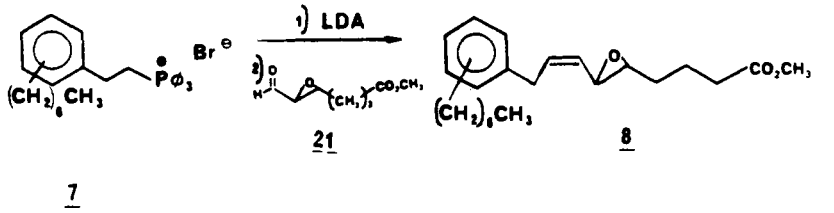
7 R = $CH_3(CH_2)_6$–
20 R = H

22 R = H

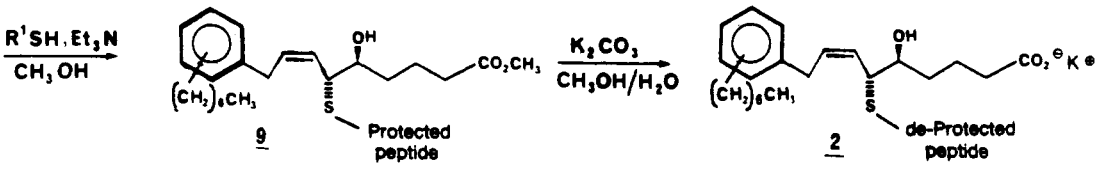
Solvent	% Yield 20
Toluene	0% - decomposition
THF	0% - decomposition
EtOH (18 hr.)	>90% (60% recrystallized)
MeOH (60 hr.)	80%

R'CHO	% Yield 22
Φ CHO	70% (c/t ratio 56/44)
$C_6H_{13}CHO$	82% (c/t ratio 80/20)

Scheme III

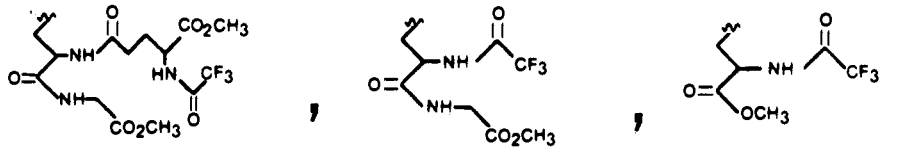


7 21 8



9 2

R^1 =

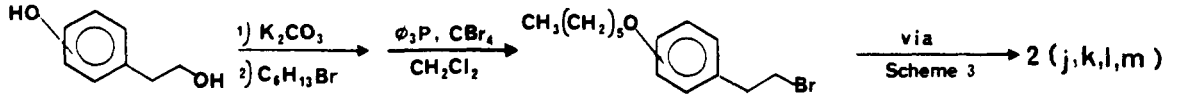


10c Protected C peptide

10d Protected D peptide

10e Protected E peptide

Scheme IV



7 (j,k,l,m)

analogues that varied in their degree of saturation of the double bonds, the 7,8 double bond was important for biological activity.[15] Combining these findings, we decided to retain the 7,8 double bond and the idea of a benzene group as a planar-region mimic but separate the two groups by a methylene linkage.

Chemistry

The analogues **2** were prepared as outlined in Schemes I, III, and IV. Wittig[16] condensation of a bromobenzaldehyde with *n*-hexyltriphenylphosphonium bromide afforded the heptenyl bromides **3** as an approximately 9/1 mixture of the *Z* and *E* olefins in 70–95% yield.[17] Treatment of these bromides with *t*-BuLi in THF at −78 °C, followed by trapping of the intermediate aryllithium species with ethylene oxide, produced the phenethyl al-

(15) Drazen, J. M.; Lewis, R. A.; Austen, K. F.; Toda, M.; Brion, F.; Marfat, A.; Corey, E. J. *Proc. Natl. Acad. Sci. U.S.A.* 1981, *78*, 3195.

(16) Johnson, A. W. *Ylid Chemistry*; Academic: New York, 1966; pp 132–192.

(17) All new compounds gave satisfactory ^1H NMR and IR analyses. Where possible, compounds were also analyzed by mass spectroscopy. All compounds up through the epoxides **8a–o** gave satisfactory mass spectral results. The high molecular weight protected-peptide adducts **9a–o** did not give consistent results on mass spectroscopy. The ionic nature of the isolated salts **2a–o** precluded mass spectral analysis on our instrumentation. Selected intermediates and the final analogues **2a–o** were analyzed by combustion analysis.

Table II. Relative Potency of Leukotrienes and Leukotriene Analogues Alone and in the Presence of L-Serine Borate, L-Cysteine, and FPL-55712

agonist	peptide type	control EC_{50}	L-serine borate (45 mM) EC_{50}	L-cysteine (3 mM) EC_{50}	FPL-55712 sensitive[a]
LTC_4	C	$3.37 \pm 1.03 \times 10^{-9}$ M	$5.20 \pm 1.31 \times 10^{-10}$ M		yes
LTD_4	D	$3.92 \pm 0.47 \times 10^{-9}$ M		$5.13 \pm 1.40 \times 10^{-10}$ M	yes
LTE_4	E	$4.16 \pm 0.93 \times 10^{-9}$ M			yes
2a	C	$5.74 \pm 0.78 \times 10^{-7}$ M	$7.45 \pm 0.87 \times 10^{-8}$ M		no
2d	C	$1.36 \pm 0.22 \times 10^{-6}$ M	$4.47 \pm 0.39 \times 10^{-7}$ M		no
2g	C	$1.86 \pm 0.36 \times 10^{-5}$ M	$1.84 \pm 0.05 \times 10^{-5}$ M		no
2b	D	$2.68 \pm 0.78 \times 10^{-6}$ M		$2.51 \pm 0.54 \times 10^{-7}$ M	yes
2e	D	weakly active[b]		$4.77 \pm 0.90 \times 10^{-6}$ M	yes
2h	D	weakly active[b]		$1.87 \pm 0.55 \times 10^{-6}$ M	yes
2c	E	$1.70 \pm 0.37 \times 10^{-6}$ M			yes
2f	E	$4.83 \pm 1.76 \times 10^{-6}$ M			yes
2i	E	not active			
2j	C	$1.28 \pm 0.18 \times 10^{-6}$ M	$2.56 \pm 0.37 \times 10^{-7}$ M		no
2k	D	$9.10 \pm 4.1 \times 10^{-6}$ M		no data	yes
2l	D	not active			
2m	D	not active			
2n	C	$3.82 \pm 0.49 \times 10^{-6}$ M	$4.51 \pm 0.75 \times 10^{-7}$ M		no
2o	D	$1.30 \pm 0.5 \times 10^{-5}$ M		no data	yes

[a] Sensitivity measured in the absence of metabolic inhibitors. [b] Onset of contraction at $\geq 10^{-5}$ M.

cohols **4** in 70–80% yield. If *n*-BuLi was utilized in this reaction, the yield was reduced due to formation of a *n*-butyl-coupled side product.[18] For preparation of the unsaturated analogues **2** (R^1 = (*Z*)-$CH{=}CHC_5H_{11}$), the pure *Z* olefins were isolated at this stage by careful silica gel chromatography of the hydroxy olefin. For preparation of the saturated analogues **2** ($R^1 = C_7H_{15}$), it was more efficient to hydrogenate the mixture of olefins over 5% Pd/C. In either case, the alcohols were converted into the corresponding bromides **6** in 90–95% yield via treatment with triphenylphosphine and carbon tetrabromide.

Conversion of these bromides to the required phosphonium salts was not trivial. Attempted coupling of triphenylphosphine with the bromides under standard conditions, e.g., in refluxing THF, acetonitrile, or toluene as solvent, led only to the destruction of the starting materials. This result was presumably due to styrene formation by β-elimination and subsequent polymerization.

At the time of these studies, the only reference on the parent analogue, phenethyltriphenylphosphonium bromide (**20**), stated that it is not possible to prepare these salts.[19] Combining our results with the literature report prompted us to backtrack to phenethyl bromide (**19**) as a model system and to develop conditions for synthesizing and using phosphonium bromide **20** (see Scheme II). We found that reaction of phenethyl bromide (**19**) or bromides **6** with triphenylphosphine in refluxing anhydrous ethanol affords the corresponding phosphonium salts **20** or **7** in 60–90% yield. We further found that **20** could be coupled under standard Wittig conditions with either benzaldehyde or heptanal to afford a mixture of the *Z*/*E*-olefins **22** in good yield (see also Scheme III).

Treatment of the phosphonium salts **7** with lithium diisopropylamide in THF at −78 °C, followed by the addition of optically pure epoxy aldehyde **21**,[20] afforded the pure epoxy *Z*-olefins **8** in 35–70% yields following chromatography (see Scheme III). In contrast to the model Wittig reactions reported above, only 2–5% of the *E*-olefin product (relative to the *Z* olefin) could be detected in the crude reaction mixtures. We believe that this difference is due to the electron-withdrawing effect of the epoxide on the carbonyl. Addition of the appropriate protected peptide synthon (**10c**, **10d**, or **10e**)[21] to the epoxide was then accomplished in methanol containing triethylamine, producing adducts **9** in 60–95% yields. These were hydrolyzed with potassium carbonate in methanol/water to afford the desired analogues **2** in 40–95% yield, following purification by reversed-phase chromatography and lyophilization to a powder.

A small series of aryl ether analogues **2j–m** was also prepared (see Scheme IV). The synthesis of these analogues began with the alkylation of the ortho, meta, and para isomers of hydroxyphenethyl alcohol with hexyl bromide and potassium carbonate in ethanol. The resultant phenethyl alcohols were converted to the final analogues **2j–m** via a sequence of reactions identical with that described above for the alkyl analogues **2a–i**.

As lyophilized salts, the analogues **2** are highly hygroscopic and are best handled in a drybox under an inert atmosphere. No decomposition could be detected by thin-layer chromatography after storage periods of up to 6 months at room temperature under nitrogen.

Even though all of the analogues were purified by using the same reversed-phase chromatography column conditions, the different peptide substituents did not all behave similarly. The LTD analogues gave combustion analyses indicative of formation of the bicarbonate salt whereas the LTC and LTE analogues gave analyses consistent with the free amino carboxylate. The reason for this difference is not clear.

Biological Results and Discussion

The activities of the test compounds were evaluated in vitro with use of isolated guinea pig tracheal spiral strips. Their pharmacological profiles were determined and compared against synthetic leukotrienes. In this system authentic leukotrienes (LT) C_4, D_4, and E_4 appear equipotent,

(18) This could occur either by coupling of *n*-BuLi with the starting aryl bromide or by coupling of the *n*-BuBr, formed during the metalation step, with the intermediate aryllithium.

(19) (a) Seyferth, D.; Fogel, J. S.; Heeren, J. K. *J. Org. Chem.* **1964**, *86*, 307. (b) More recently Hine has reported the preparation of **20** by reaction in xylene and "purification" by boiling a residual solid in ethanol. Hine, J.; Skoglund, M. J. *J. Org. Chem.* **1982**, *47*, 4758.

(20) The epoxy aldehyde **21** was synthesized from 2-deoxy-D-ribose by following the procedure of Rokach et al.: Rokach, J.; Zamboni, R.; Lau, C.-K.; Guindon, Y. *Tetrahedron Lett.* **1981**, *22*, 2759.

(21) The indicated protected peptide synthons for glutathione, cysteinylglycyl, and cysteinyl were prepared as outlined in ref. 5 and as in the following: Corey, E. J.; Clark, D. A.; Marfat, A.; Goto, G. *Tetrahedron Lett.* **1980**, *21*, 3143.

since they undergo metabolic conversion by the tracheal tissue.[22] LTC_4 is converted to LTD_4 by γ-glutamyl transpeptidase, and LTD_4 is converted to LTE_4 by an aminopeptidase. Selective blockade of these metabolic processes enhances the contractile activity of specific leukotrienes. Thus, L-serine borate (45 mM) blocks the activity of γ-glutamyl transpeptidase, thereby increasing the potency of LTC_4. Similarly, L-cysteine (3 mM) inhibits the activity of the aminopeptidase, increasing the potency of LTD_4. Thus, in the presence of metabolic inhibitors, the relative potencies of the leukotrienes are $LTC_4 = LTD_4 > LTE_4$.[22]

As illustrated in Table II, all but three of the stable analogues show agonist activity in isolated guinea pig tracheal strips. Unlike the authentic leukotrienes, the C-peptide analogues are more potent than the D-peptide analogues in each series. Except for the ortho-substituted C_7H_{15} analogues, the contractile activity of the C-peptide analogues is enhanced by L-serine borate, which suggests that, like LTC_4, these analogues are substrates for γ-glutamyl transpeptidase. In the D-peptide series, L-cysteine enhances the contractile activity of all the analogues tested. This finding is similar to that seen with authentic LTD_4.[22] The D-peptide analogues that show agonist activity are all antagonized by the leukotriene antagonist FPL-55712.[23] Likewise, the E-peptide analogues that possess contractile activity are sensitive to blockade by FPL-55712.

However, the C-peptide analogue, all of which are agonists, are not sensitive to blockade by FPL-55712 even in the absence of metabolic inhibitors. We believe that this may be explained by the following: First, we previously have shown that there is more than one receptor for the peptidoleukotrienes in guinea pig tracheal spirals.[23b] Of these receptors, the one that LTC_4 binds to is not blocked by FPL-55712. The blockade of LTC_4 by FPL-55712 in the absence of metabolic inhibitors is due to the rapid conversion of the LTC_4 to LTD_4 and LTE_4, which are equipotent and FPL-sensitive.[23b] Second, the potency at the C receptor of the C–peptide analogues appears to be much greater than the potency of the D analogues or E analogues at the D/E receptor (see Table II). Combining these two points implies that in order for us to see blockade by FPL-55712 of the activity of the C-peptide analogues, they would have to be fully converted to the weaker D/E-peptide analogues. Although the increase in potency in the presence of the metabolic inhibitors indicates metabolism, we believe that these analogues may be metabolized more slowly (relative to the amount of starting C-peptide analogue) than the leukotrienes. This is because the bath concentrations of the analogues required to produce an equivalent biological response are 100 times greater than the concentrations of the leukotrienes.

The two E analogues that retained agonist activity (**2c** and **2f**) have lower intrinsic activity than the C/D analogues. The maximum response obtained with these E-peptide analogues is 60% of that obtained with the reference agonist, carbachol, whereas with the C/D-peptide analogues the maximum response is >100% of the initial response with carbachol. Because of this, it appears that **2f** is more active than **2e** (Table II). In reality **2e**, in the absence of L-cysteine, does not reach its maximum response at the concentration tested.

Table III. Inhibition of LTE_4-Induced Contraction of Isolated Guinea Pig Trachea by Leukotriene Analogues

analogue	concentration, mM	% inhibn of LTE_4 (8 nM) induced contraction
2i	10	28%
2l	10	24%
2m	10	18%

Three of the analogues (**2i**, **2l**, and **2m**) failed to contract guinea pig trachea at concentrations as high as 100 μM. These analogues were then tested to see if they could inhibit the contractile response produced by 8 nM LTE_4. As illustrated in Table III, these compounds at 10 μM produce nearly equal inhibition (28%, 24%, and 18%, respectively) of the LTE_4 response. Thus, very slight modifications of the structure of stable leukotriene analogue agonists transformed them to antagonists.

If we further examine the results, focusing on analogues **2a–i**, two more points become apparent. The first is that if we rank them by potency as agonists, then the para- > meta- > ortho-substituted isomer. The second is that the C/D-peptide analogues in the presence of metabolic inhibitors are more potent and more efficacious than the E-peptide analogues. The first point is also supported by the EC_{50} values we find for **2j** and **2k**.

Summary

The results demonstrate that a structurally novel series of chemically stable analogues of peptide leukotrienes can be developed that are pharmacologically active either as agonists or antagonists. The blockade of the effects of the D and E analogues and the lack of effect on the activity of the C analogues by FPL-55712 show specific action of these analogues at the leukotriene receptor sites. Although the most potent C-peptide analogue is approximately 100-fold less potent than natural LTC_4, it is approximately 100 times more potent than either histamine or carbachol.

Experimental Section

Synthetic Procedures. Melting points were taken on either a Fisher-Johns or a Thomas-Hoover melting point apparatus and are uncorrected. Analytical samples were homogeneous by TLC, IR, and ^1H NMR and afforded spectroscopic results consistent with the assigned structures. In addition, non-ionic intermediates were analyzed by mass spectroscopy. Analytical thin-layer chromatography (TLC) was conducted either on prelayered silica gel GHLF plates (Analtech, Newark, DE) or on Whatman MKC 18F reversed-phase TLC plates (RP-TLC). Visualization of the plates was accomplished by using UV light and/or phosphomolybdic acid–sulfuric acid charring. Infrared spectra (IR) were taken on Perkin-Elmer 727B or 781 spectrometers; band locations are reported in cm^{-1}. Proton nuclear magnetic resonance spectra (NMR) were obtained by using either a Bruker WM-250, an IBM NR-80, or a Varian EM-360 spectrometer. Peak positions are reported in parts per million (δ), with use of tetramethylsilane as an internal standard. Mass spectra (MS) were recorded on a Kratos MS-80 instrument operating either in the electron impact (EI) or chemical ionization (CI) mode as indicated. Elemental analyses were performed on a Perkin-Elmer 241 elemental analyzer and are within 0.4% of the theoretical values unless otherwise indicated. Flash chromatography[24] was conducted on Kieselgel 60, 230–400 mesh (E. Merck, Darmstadt, West Germany) or on J. T. Baker octadecylsilyl (ODS) packing material, 40 μm. High-pressure liquid chromatography (HPLC) was performed on a Beckmann Model 334 HPLC.

All organic starting materials and reagents were obtained from Aldrich Chemical Co. and were used without additional purifi-

(22) Snyder, D. W.; Aharony, D.; Dobson, P.; Tsai, B. S.; Krell, R. D. *J. Pharmacol. Exp. Ther.* **1984**, *231*, 222.

(23) The fact that our C, D, and E analogues show differential potency as agonists in the absence of metabolic inhibitors and also that they are not equally blocked by FPL-55712 acted as a spur to our previously reported work on multiple leukotriene receptors: (a) Krell, R. D.; Tsai, B. S.; Berdoulay, A.; Barone, M.; Giles, R. E. *Prostaglandins* **1983**, *25*, 171. (b) Snyder, D. W.; Krell, R. D. *J. Pharmacol. Exp. Ther.* **1984**, *231*, 616.

(24) Still, W. C.; Kahn, M.; Mitra, A. *J. Org. Chem.* **1978**, *43*, 2923.

cation, unless otherwise indicated. Solvents were either reagent or HPLC grade and were obtained from Fisher Scientific or J. T. Baker Chemical Co. Tetrahydrofuran (THF) was distilled under nitrogen from sodium benzophenone ketyl directly prior to use. Triethylamine (TEA) was distilled from CaH_2 and stored over KOH pellets under nitrogen. Lithium diisopropylamide (LDA) solutions were prepared in THF from *n*-BuLi and diisopropylamine directly prior to use.

Unless otherwise indicated, all reactions were carried out under an inert atmosphere of nitrogen with vigorous magnetic stirring. Evaporations were carried out in vacuo on a rotary evaporator.

4-(1-Hepten-1-yl)bromobenzene (3a). A 5-L flask was charged with 213.7 g (0.5 mol) of *n*-hexyltriphenylphosphonium bromide (Alfa) and 2.0 L of THF. The resultant suspension was cooled to −60 °C, and 580 mL of potassium hexamethyldisilazide (Callery Chemicals) solution (1.0 M in THF) was added slowly such that the temperature remained ≤−50 °C. The orange suspension was stirred for $^1/_2$ h at −50 °C and then allowed to warm slowly to ~10 °C over 3 h to afford a clear red-orange solution. This solution was cooled to −50 °C, and 92.5 g (0.5 mol) of *p*-bromobenzaldehyde in 200 mL of THF was added to it. After being kept for $^1/_2$ h at −50 °C, the orange/brown reaction mixture was allowed to warm slowly to 0 °C overnight and then poured into 6 L of water and extracted with ethyl acetate (2 × 1.0 L). The ethyl acetate extracts were washed with 1.5 L of brine, dried over anhydrous Na_2SO_4, and concentrated to afford 340 g of crude product. The crude product was suspended in 1.5 L of hexane and stirred vigorously for 1 h. Filtration removed most of the triphenylphosphine oxide. Concentration of the filtrate gave 153 g of a yellow oil, which was further purified by flash chromatography on a 2 in. × 16 in. silica gel column, with use of hexane as eluent. Combination of the appropriate fractions and concentration afforded 101.2 g (79% yield) of **3a** as a colorless oil, R_f 0.62 (hexane): IR (CCl_4) 2910, 2845, 1480, 1070 cm^{-1}; NMR ($CDCl_3$, 250 MHz) δ 0.89 (t, J = 7.5 Hz, 3 H), 1.30 (m, 4 H), 1.38–1.50 (m, 2 H), 2.22–2.32 (m, 2 H), 5.68 (dt, J = 5, 12.5 Hz, 1 H), 6.32 (brd, J = 12.5 Hz, 1 H), 7.14 (d, J = 8.5 Hz, 2 H), 7.43 (d, J = 8.5 Hz, 2 H); MS-EI, m/e 252 (M^{-1}, 60%), 254 (M^{+1}, 59%), 116 (100%). Anal. ($C_{13}H_{17}Br$) C, H, Br.

4-(1-Hepten-1-yl)phenethyl Alcohol (4a). A solution of 101 g of **3a** in 1.0 L of THF was placed in a dry 3-L, three-necked flask equipped with a mechanical stirrer, a 500-mL addition funnel, and a gas-outlet adapter connected to a sulfuric acid bubbler. The reaction mixture was cooled to −60 °C, and 440 mL of *t*-BuLi (2.15 M in hexanes) was slowly added. After the addition was complete, the mixture was stirred at −70 °C for ~1 h, and the addition funnel was replaced with a 50-mL jacketed additon funnel. The jacket was charged with a slurry of dry ice/acetone, and ~26 mL of ethylene oxide (WARNING: CANCER SUSPECT AGENT) was condensed directly into the funnel. The ethylene oxide was then added over ~2 min to the emerald-colored solution. The reaction mixture was allowed to warm to room temperature over 2 h, and then cooled in ice water (~5 °C) and adjusted carefully to pH 2 with 3.0 M HCl (~230 mL of HCl). After $^1/_2$ h at room temperature, the reaction mixture was diluted with 2.0 L of water and extracted with diethyl ether (3 × 500 mL). The diethyl ether extracts were combined, washed with 500 mL of saturated sodium bicarbonate solution, dried over Na_2SO_4, filtered, and concentrated to afford 103.4 g of crude product. This material was purified by flash chromatography on a 2 in. × 16 in. silica gel column, with first hexane and then 20% diethyl ether/hexane as eluents. Combination of the appropriate fractions and concentration gave 79.7 g (91.5% yield) of the pure phenethyl alcohol **4a** as a clear, colorless oil, R_f 0.15 (ether/hexanes, 1/3): IR (CCl_4) 3550–3200, 2950, 2850, 1050 cm^{-1}; NMR ($CDCl_3$, 60 MHz) δ 0.90 (t, 3 H), 1.21–1.66 (m, 4 H), 1.70–1.94 (m, 2 H), 2.33 (dt, J = 5 Hz, 2 H), 2.83 (t, J = 6 Hz, 2 H), 3.88 (brt, J = 5 Hz, 2 H), 5.65 (dt, J = 7,11 Hz, 1 H), 6.38 (d, J = 11 Hz, 1 H), 7.22 (s, 4 H); MS-EI, m/e 219 (M^{+1}, 4%), 218 (M^+, 27%), 131 (100%). Anal. ($C_{15}H_{22}O$) C, H.

4-Heptylphenethyl Alcohol (5a). A suspension of 69.5 g (0.318 mol) of unsaturated alcohol **4a** and 3.5 g of 10% Pd/C (Alfa-Ventron) in 600 mL of absolute ethanol was hydrogenated at atmospheric pressure until 7.9 L of hydrogen was taken up (~2 h, theory 7.7 L). The mixture was flushed with nitrogen and filtered through a pad of diatomaceous earth. The pad and reaction vessel were washed with ethanol, and the combined filtrates were concentrated to afford 71.9 g (quantitative yield) of the saturated alcohol **5a**: IR (CCl_4) 3500–3200, 2940, 2850, 1050 cm^{-1}; NMR ($CDCl_3$, 60 MHz) δ 0.86 (brt, 3 H), 1.15–1.50 (m, 8 H), 1.63–1.92 (m, 2 H), 2.56 (t, J = 7 Hz, 2 H), 2.79 (t, J = 6 Hz, 2 H), 3.80 (t, J = 6 Hz, 2 H), 7.10 (s, 4 H); MS-EI, m/e 221 (M^{+1}, 12%), 220 (M^+, 62%), 189 (100%).

4-Heptylphenethyl Bromide (6a). To an ice-chilled solution of 71.9 g (0.326 mol) of alcohol **5a** and 90.5 g (0.345 mol) of triphenylphosphine in 500 mL of dichloromethane was added a solution of 114.5 g (0.345 mol) of carbon tetrabromide in 300 mL of dichloromethane over 1 h. After the addition was completed, the reaction mixture was stirred at 5 °C for 1 h and then concentrated to a viscous yellow oil. The mixture was diluted with 1.2 L of hexane and stirred vigorously for 1 h and then filtered to remove $Ph_3P{=}O$. Concentration of the filtrate yielded 126 g of the crude bromide as a pale yellow oil, which was purified via flash chromatography on a $1^1/_2$ in. × 12 in. silica gel column with use of hexane as eluent. Concentration of the appropriate fractions gave 86.2 g (93% yield) of bromide **6a** as a clear oil, R_f 0.32 (hexanes): IR (CCl_4) 2920, 2850, 1120 cm^{-1}; NMR ($CDCl_3$, 60 MHz) δ 0.87 (t, J = 7 Hz, 3 H), 1.06–1.66 (m, 10 H), 2.55 (t, J = 7 Hz, 2 H), 3.10 (t, J = 7 Hz, 2 H), 3.3 (t, J = 6 Hz, 2 H), 7.1 (s, 4 H); MS-EI, m/e 284 (M^{+1}, 44%), 282 (M^{-1}, 45%), 117 (100%).

(4-Heptylphenethyl)triphenylphosphonium Bromide (7a). A mixture of 86.0 g (0.306 mol) of bromide **6a** and 87.6 g (0.334 mol) of triphenylphosphine in 1.5 L of absolute ethanol was heated at gentle reflux for 3 days. The mixture was concentrated, and the residue was azetroped (3×) with chloroform to give a straw-colored syrup, which was diluted with 1.0 L of ether, resulting in formation of a precipitate. Filtration, suspension of the solid in 500 mL of warm ethyl acetate for 2 h, refiltering, and drying of the white solid in a vacuum oven overnight afforded 133.3 g (80.5% yield) of phosphonium salt **7a**, mp 99–102 °C. This material could then be used directly in the next reaction; however, recrystallization of a small portion for analysis from hot ethyl acetate afforded fine white needles, mp 113–115 °C, R_f 0.25 (7% MeOH, $CHCl_3$): IR (KBr) 2910, 2840, 1438, 1105 cm^{-1}; NMR ($CDCl_3$, 60 MHz) δ 0.87 (t, J = 7 Hz, 3 H), 1.19–1.37 (m, 8 H), 1.48–1.68 (m, 2 H), 2.51 (t, J = 9 Hz, 2 H), 2.84–3.24 (m, 2 H), 3.97–4.34 (m, 2 H), 7.0 (d, J = 14 Hz, 2 H), 7.19 (d, J = 10 Hz, 2 H), 7.52–8.0 (m, 15 H). Anal. ($C_{33}H_{48}PBr$) C, H, P.

(5*S*,6*S*,7*Z*)-Methyl 5,6-Oxido-9-(4-heptylphenyl)-7-nonenoate (8a). A suspension of 600 mg (1.1 mmol) of phosphonium salt **7a** in 10 mL of THF was cooled to −60 °C, and a solution of LDA (0.5 M in THF) was slowly added until a permanent yellow tint appeared (~0.5 mL). Then an additional 2.2 mL (1.1 mmol) of LDA was added (T < −55 °C). The orange solution was kept at this temperature for 10 min, warmed to −30 °C, and recooled to −60 °C, and then 234 mg (1.3 mmol) of epoxy aldehyde **21** as a solution in 1.0 mL of THF was added to it. The now-yellow reaction mixture was kept at this temperature for 45 min, slowly warmed to 0 °C, and diluted with 30 mL of hexanes and 4 mL of diethyl ether. The organic layer was washed with 1 × 7 mL of pH 7.5 phosphate buffer and 1 × 7 mL of brine, dried over Na_2SO_4, and concentrated in vacuo. The residue was stirred in hexanes/diethyl ether/TEA (95/5/2) and filtered to remove triphenylphosphine oxide. The filtrate was concentrated and the residue flash chromatographed on 10.3 g of silica gel with use of hexanes/diethyl ether/TEA (95/5/2) as eluent. Concentration of the appropriate fractions afforded 300 mg (76% yield) of epoxide **8a** as a clear colorless oil, R_f 0.38 (25/75 diethyl ether/hexane): IR ($CHCl_3$) 1735, 1430, 1150 cm^{-1}; NMR ($CDCl_3$) δ 0.88 (t, J = 7.6 Hz, 3 H), 1.3 (m, 9 H), 1.59 (m, 4 H), 1.85 (m, 2 H), 2.38 (t, J = 7.6 Hz, 2 H), 2.58 (t, J = 7.6 Hz, 2 H), 2.88 (dt, J = 2.5, 7.1 Hz, 1 H), 3.46 (brd, J = 7.5 Hz, 1 H), 3.54 (brd, J = 7.5 Hz, 2 H), 3.69 (s, 3 H), 5.15 (dd, J = 8.4, 10.1 Hz, 1 H), 5.86 (dt, J = 7.9, 10.1 Hz, 1 H), 7.11 (AA′, 4 H); MS-CI, m/e 359 (M^{+1}); exact mass (EI) ($C_{23}H_{34}O_3$) calcd 358.2508, found 358.2523.

(5*S*,6*R*,7*Z*)-6-[*N*-(Trifluoroacetyl)-*S*-glutathionyl dimethyl ester]-5-hydroxy-9-(4-heptylphenyl)-7-nonenoic Acid Methyl Ester (9a). To a solution of 56 mg (0.156 mmol) of epoxide **8a**, 1.0 mL of methanol, and 0.25 mL of TEA was added 113 mg (0.26 mmol) of protected-C-peptide **10c**. After the mixture was stirred overnight, it was diluted with 20 mL of ether, resulting

in precipitation of the excess C peptide (**10c**). Filtration followed by washing of the filtrate with water, drying of the ethereal phase over $MgSO_4$, filtration, and concentration afforded a viscous gum. Flash chromatography on 9.5 g of silica gel utilizing 10% ethyl acetate/diethyl ether as the eluent and concentration of the appropriate fractions afforded 99 mg (86% yield) of protected-C-peptide adduct **9a** as a waxy solid, R_f 0.31 (25/75 ethyl acetate/diethyl ether): IR ($CHCl_3$) 3400, 1750, 1724, 1650 cm^{-1}; NMR ($CDCl_3$) δ 0.88 (t, J = 7 Hz, 3 H), 1.3 (m, 6 H), 1.57 (m, 4 H), 1.75 (m, 1 H), 1.86 (m, 1 H), 2.21 (m, 1 H) 2.38 (t, J = 7.3 Hz, 1 H), 2.40 (t, J = 7 Hz, 2 H), 2.56 (t, J = 7.3 Hz, 2 H), 2.77 (s, 1 H), 2.79 (dd, J = 7.8, 14.6 Hz, 1 H), 3.0 (dd, J = 7.8, 14.6 Hz, 1 H), 3.47 (m, 2 H), 3.67 (s, 3 H), 3.73 (s, 3 H), 3.79 (s, 3 H), 3.96 (m, 3 H), 4.59 (m, 2 H), 5.56 (dd, J = 10.4 Hz, 1 H), 5.89 (dt, J = 7.5, 10.4 Hz, 1 H), 6.74 (m, 2 H), 7.11 (brs, 4 H), 8.39 (brd, 1 H); MS-CI, m/e 790 (M^+).

(5*S*,6*R*,7*Z*)-6-*S*-Glutathionyl-5-hydroxy-9-(4-heptylphenyl)-7-nonenoic Acid Tripotassium Salt (2a). To a suspension of 92 mg (0.116 mmol) of C-peptide adduct **9a** in 2.0 mL of methanol was added 462 mg (3.35 mmol) of potassium carbonate dissolved in 2.0 mL of water. The mixture was stirred at room temperature for 7 h, acidified to pH 7, and added to the top of a 1.2 cm × 15 cm ODS-packed flash column. Elution with 60/40 methanol/water, combination of the appropriate fractions, Millipore filtration to remove traces of ODS packing material, and concentration followed by lyophilization afforded 73 mg of **2a** as a white powder, mp 197–200 °C, R_f (RP-TLC) 0.64 (70/30 methanol/water): IR (KBr) 3200–3600, 1660, 1640, 1560, 1400 cm^{-1}; NMR (CD_3OD) δ 0.88 (t, J = 7 Hz, 3 H), 1.3 (m, 8 H), 1.56 (m, 4 H), 1.76 (m, 1 H), 2.18 (m, 4 H), 2.55 (t, J = 7.5 Hz, 4 H), 2.75 (dd, J = 10, 15 Hz, 1 H), 3.07 (dd, J = 5.5, 15 Hz, 1 H), 3.46 (brd, J = 7.5 Hz, 2 H), 3.8 (m, 3 H), 4.59 (dd, J = 4, 10 Hz, 1 H), 5.48 (dd, J = 8.4, 10.1 Hz, 1 H), 5.74 (dt, J = 7.9, 10.1 Hz, 1 H), 7.04 (d, J = 9.0 Hz, 2 H), 7.11 (d, J = 9.0 Hz, 2 H). Anal. ($C_{32}H_{46}N_3O_9SK_3 \cdot 3H_2O$) C, H, N.

(5*S*,6*R*,7*Z*)-6-[*N*-(Trifluoroacetyl)-*S*-cysteinylglycinyl methyl ester]-5-hydroxy-9-(4-heptylphenyl)-7-nonenoic Acid Methyl Ester (9b). Via a protocol essentially identical with that given in the procedure for **9a**, but with 45 mg (0.126 mmol) of epoxide **8a** and 70 mg (0.276 mmol) of protected-D-peptide **10d** as starting materials, there was obtained 48 mg (64% yield) of protected-D-peptide adduct **9b** following chromatography, with use of 30/1 diethyl ether/hexanes as eluent, R_f 0.5 (30/1 diethyl ether/hexanes): IR ($CHCl_3$) 1750, 1740, 1690 cm^{-1}; NMR ($CDCl_3$, 250 MHz) δ 0.88 (t, J = 7.5 Hz, 3 H), 1.30–1.38 (m, 8 H), 1.51–1.65 (m, 4 H), 1.65–1.94 (m, 2 H), 2.37 (t, J = 7.5 Hz, 2 H), 2.55 (t, J = 7.5 Hz, 2 H), 2.74–3.02 (m, 1 H), 2.63 (d, J = 5 Hz, 1 H), 3.46 (m, 2 H), 3.69 (s, 3 H), 3.77 (s, 3 H), 3.84–4.11 (m, 3 H), 4.52 (dd, J = 7.5, 7.5 Hz, 1 H), 5.59 (dd, J = 10 Hz, 1 H), 5.94 (dt, J = 5, 8.7 Hz, 1 H), 6.72 (brt, 1 H), 7.11 (s, 4 H), 7.5 (d, J = 9 Hz, 1 H).

(5*S*,6*R*,7*Z*)-6-(*S*-Cysteinylglycinyl)-5-hydroxy-9-(4-heptylphenyl)-7-nonenoic Acid Dipotassium Monobicarbonate Salt (2b). Via a protocol essentially identical with that given in the procedure for **2a**, but with 44 mg (0.068 mmol) of protected-D-peptide adduct **9b** as starting material, and with use of 50/50 methanol/water as the eluent for the chromatography, there was obtained 20 mg (50% yield of D-peptide analogue **2b** as a white power, mp 168–183 °C dec, R_f (RP-TLC) 0.47 (65/35 methanol/water): NMR [CD_3OD, 250 MHz] δ 0.88 (t, J = 7.5 Hz, 3 H), 1.24–1.38 (m, 8 H), 1.52–1.64 (m, 4 H), 1.70–1.89 (m, 2 H), 2.18 (t, J = 7.5 Hz, 2 H), 2.54 (t, J = 7.5 Hz, 2 H), 2.69 (dd, J = 9.5, 13.5 Hz, 1 H), 2.97 (dd, J = 9.5, 13.5 Hz, 1 H), 5.51 (dd, J = 10 Hz, 1 H), 5.74 (dt, J = 10 Hz, 1 H), 7.15 (dd, J = 8.5, 15 Hz, 1 H). Anal. ($C_{27}H_{40}O_6N_2SK_2 \cdot H_2CO_3$) C, H, N.

(5*S*,6*R*,7*Z*)-6-[*N*-(Trifluoroacetyl)-*S*-cysteinyl methyl ester]-5-hydroxy-9-(4-heptylphenyl)-7-nonenoic Acid Methyl Ester (9c). Via a protocol essentially identical with that given in the procedure for **9a**, but with 74 mg (0.21 mmol) of epoxide **8a** and 73.2 mg (0.32 mmol) of protected-E-peptide **10e** as starting materials, there was obtained 58 mg (47% yield) of protected-E-peptide adduct **9a** following preparative HPLC chromatography on Zorbax cyanopropyl packing eluting with 90/10/0.5 hexane/dichloromethane/methanol, Rv = 9.6 column volumes: IR ($CHCl_3$) 3400, 1730 cm^{-1}; NMR ($CDCl_3$, 250 MHz) δ 0.86 (t, J = 6 Hz, 3 H), 1.24–1.32 (m, 8 H), 1.48–1.61 (m, 4 H), 1.63–1.87 (m, 2 H), 2.32 (t, J = 7.5 Hz, 2 H), 2.35 (d, J = 5 Hz, 1 H), 2.55 (t, J = 8 Hz, 2 H), 3.02 (dd, J = 7.5 Hz, 2 H), 3.39 (brd, J = 7.5 Hz, 2 H), 3.62–3.75 (m, 1 H), 3.66 (s, 3 H), 3.76 (s, 3 H), 3.78–3.86 (m, 1 H), 4.79 (dd, 1 H), 5.45 (dd, J = 10, 13.5 Hz, 1 H), 5.86 (dt, J = 7.5, 10 Hz, 1 H), 7.08 (s, 4 H); MS-CI, m/e 572 (M^{+1} - [H_2O], 34%), 341 (100%).

(5*S*,6*R*,7*Z*)-6-*S*-Cysteinyl-5-hydroxy-9-(4-heptylphenyl)-7-nonenoic Acid Dipotassium Salt (2c). Via a protocol essentially identical with that given in the procedure for **2a**, but with 58 mg (0.1 mmol) of protected-E-peptide adduct **9c** as starting material and with the reaction proceeding at room temperature for 84 h, there was obtained 25 mg (47% yield) of E-peptide analogue **2c** as a white powder (mp >225 °C). Chromatography was accomplished with a gradient elution technique, starting with 50/50 methanol/water and going up to 100% methanol, R_f (RP-TLC) 0.31 (65/35 methanol/water): NMR (CD_3OD, 250 MHz) δ 0.88 (t, J = 7.5 Hz, 3 H), 1.22–1.35 (m, 8 H), 1.50–1.70 (m, 4 H), 1.70–1.80 (m, 2 H), 2.16 (brd, J = 7.5 Hz, 2 H), 2.54 (t, J = 7 Hz, 2 H), 3.47 (brd, J = 7.5 Hz, 2 H), 5.53 (dd, J = 10, 10 Hz, 1 H), 5.76 (m, 1 H), 7.1 (dd, J = 5, 7.5 Hz, 4 H). Anal. ($C_{25}H_{37}O_5NSK_2 \cdot H_2O$) C, H, N.

Biological Evaluation Procedures. Guinea pigs were killed by a sharp blow to the head and the tracheas removed and cut into spiral strips. Each trachea was divided into two sections for paired experiments. Each section was placed in a jacketed 10-mL tissue bath maintained at 37 °C and bathed with modified Kreb's buffer that was bubbled with 95% O_2 and 5% CO_2. The Kreb's buffer consisted of the following composition (mM): NaCl (119), KCl (4.6), $CaCl_2$ (1.8), $MgCl_2$ (0.5), $NaHCO_3$ (24.9), NaH_2PO_4 (1.0), and glucose (11.1). The bath fluid also contained indomethacin (5 μM). Isometric tension was monitored via a Grass Force Displacement Transducer and displayed on a Beckman Dynograph (Model R 611). Resting tension was set at 2 g, and the tissues were allowed to stabilize for 60 min as the bath fluid was changed every 15 min.

Cumulative concentration–response curves were obtained by successive increases in the bath concentration of the agonist. Contractile responses are expressed as a percentage of the response obtainable to a maximally effective concentration of carbachol (30 μM), which was added to the bath after the stabilization period. Following the carbachol challenge, the tissues were washed and allowed to restabilize for 60 min before the concentration–response curves were begun. EC_{50} values, the concentration of agonist necessary to produce a contraction equal to 50% of the maximal response, were derived by linear regression.[25] The SRS-A antagonist FPL-55712 (10 μM) and the metabolic inhibitors L-serine borate (45 mM), which blocks γ-glutamyl transpeptidase activity, and L-cysteine (3 mM), which blocks aminopeptidase activity, were incubated for 30 min prior to the starting of the concentration–response curves. EC_{50} values were determined in the absence and presence of antagonist or metabolic inhibitors, and the significance ($p < 0.05$) was determined with use of the Student's paired t test.[25] K_b values were determined according to the method of Furchgott.[26]

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Supplementary Material Available: Analytical data for compounds **2g–i,k–m,o**, **5l,m**, **6l,m**, **7d,g,j,l–o**, **8d,g,l,m**, and **9g–i** (4 pages). Ordering information is given on any current masthead page.

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