Discovery of Orally Active Nonpeptide Vitronectin Receptor Antagonists Based on a 2-Benzazepine Gly-Asp Mimetic

William H. Miller,*,[†] Doreen P. Alberts,[†] Pradip K. Bhatnagar,[†] William E. Bondinell,[†] James F. Callahan,[†] Raul R. Calvo,[†] Russell D. Cousins,[†] Karl F. Erhard,[‡] Dirk A. Heerding,[†] Richard M. Keenan,[†] Chet Kwon,[†] Peter J. Manley,[†] Kenneth A. Newlander,[†] Stephen T. Ross,[‡] James M. Samanen,[†] Irene N. Uzinskas,[†] Joseph W. Venslavsky,[†] Catherine C.-K. Yuan,[†] R. Curtis Haltiwanger,[‡] Maxine Gowen,[‡] Shing-Mei Hwang,[‡] Ian E. James,[‡] Michael W. Lark,[‡] David J. Rieman,[‡] George B. Stroup,[‡] Leonard M. Azzarano,[‡] Kevin L. Salyers,[‡] Brian R. Smith,[‡] Keith W. Ward,[‡] Kyung O. Johanson,[‡] and William F. Huffman[†]

Research & Development Division, SmithKline Beecham Pharmaceuticals, 1250 South Collegeville Road, P.O. Box 5089, Collegeville, Pennsylvania 19426-0989, and 709 Swedeland Road, P.O. Box 1539, King of Prussia, Pennsylvania 19406-0939

Received August 30, 1999

Introduction. The vitronectin receptor, also known as $\alpha v \beta 3$, is a member of the integrin family of heterodimeric transmembrane glycoprotein complexes that function in cellular adhesion events and signal transduction processes.[1] Integrin $\alpha v \beta 3$ is expressed on almost all cells originating from the mesenchyme and has been shown to mediate several biologically relevant processes, including adhesion of osteoclasts to the bone matrix,[2−5] migration of vascular smooth muscle cells,[6−8] and angiogenesis.[9] As a result, antagonists of integrin $\alpha v \beta 3$ are expected to have utility in the treatment of several human diseases, including osteoporosis,[10] restenosis following percutaneous transluminal coronary angioplasty (PTCA),[6,7] and diseases involving neovascularization, such as rheumatoid arthritis,[11,12] cancer,[13−16] and ocular diseases.[17,18] Like many, but not all, integrins $\alpha v \beta 3$ is known to recognize the arginine−glycine−aspartic acid (RGD) tripeptide sequence.[1] Therefore, we[19−23] and others[10,13,24−27] have investigated peptidomimetic approaches to identify $\alpha v \beta 3$ antagonists. In this communication, we describe a new series of small molecule RGD mimetics that are highly potent, orally active $\alpha v \beta 3$ antagonists. Selected members of this series are potent inhibitors of bone resorption in vitro and in vivo and have activity in an animal model of osteoporosis.

Chemistry. The benzazepine derivatives were prepared by the procedures outlined in Schemes 1−3. The syntheses of **5** and **7** illustrate the methods. Commercially available 4-bromo-3-methylanisole (**8**) was converted to benzazepine **14** using a slight modification of our general method for the synthesis of 2-benzazepine derivatives (Scheme 1).[28] The racemic benzazepine **14**

* To whom correspondence should be addressed. Tel: 610-917-7937. Fax: 610-917-4206. E-mail: william_h_miller@sbphrd.com.
† 1250 S. Collegeville Rd.
‡ 709 Swedeland Rd.

was resolved into its (*R*)- and (*S*)-enantiomers by chiral HPLC.[29] The absolute configurations were assigned following X-ray crystallographic analysis of the (*S*)-phenol **16**,[30] which was obtained by demethylation of **15** with BBr$_3$.

For the synthesis of **5**, the (*S*)-phenol **16** was reacted with the pyridine derivative **18** in a Mitsunobu reaction (Scheme 2).[31,32] The resulting ether derivative **19** was converted to **5** by a reduction/saponification sequence described previously for related compounds.[23,33]

Compound **7** was prepared from the (*S*)-phenol **16** and 6-(methylamino)-2-pyridylethanol (**26**), which was prepared straightforwardly from 2-amino-6-picoline (Scheme 3). Compound **26** was conveniently purified by crystallization of the corresponding formate salt.

Results and Discussion. In our preliminary studies,[19] we identified **1** (Table 1), a potent, 1,4-benzodiazepine-based $\alpha v \beta 3$ antagonist with a good in vitro profile but with low oral bioavailability in rats. Followup studies on **1**[20−22] indicated that neither biological activity nor oral bioavailability could be improved substantially in the 1,4-benzodiazepine series. However, **1** proved to be a suitable template for the design of second-generation $\alpha v \beta 3$ antagonists with improved properties. An initial success was the identification of **2**,[23] wherein the lactam amide, the linking amide, and N-1 of the 1,4-benzodiazepine system were replaced with more hydrophobic groups and the benzimidazole arginine (Arg) mimetic was replaced with an aminopyridine Arg mimetic. Compound **2** has improved biological activity relative to **1** and has high oral bioavailability in rats (approximately 100%).[34] We were concerned, however, about the level of biological activity of **2** in cell-based assays,[35] so we continued our investigations in search of orally bioavailable $\alpha v \beta 3$ antagonists with improved biological activity.

Beginning with **1**, we envisioned replacing the linking amide with an ether linkage, similar to that in **2**, while retaining the seven-membered ring lactam (Figure 1). Previously, in our work leading to **2**, we found that replacing the lactam amide with a phenyl ring caused a 4-fold drop in affinity for $\alpha v \beta 3$,[23] which suggests that the lactam amide may interact favorably with the receptor. Since replacing the linking amide with an ether linkage in the 1,4-benzodiazepine series would generate a 4-alkoxyaniline derivative, we were con-
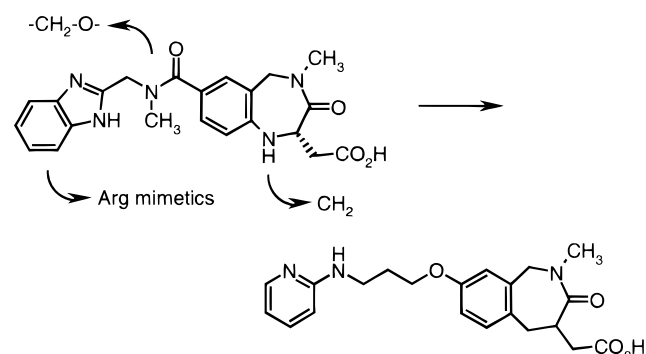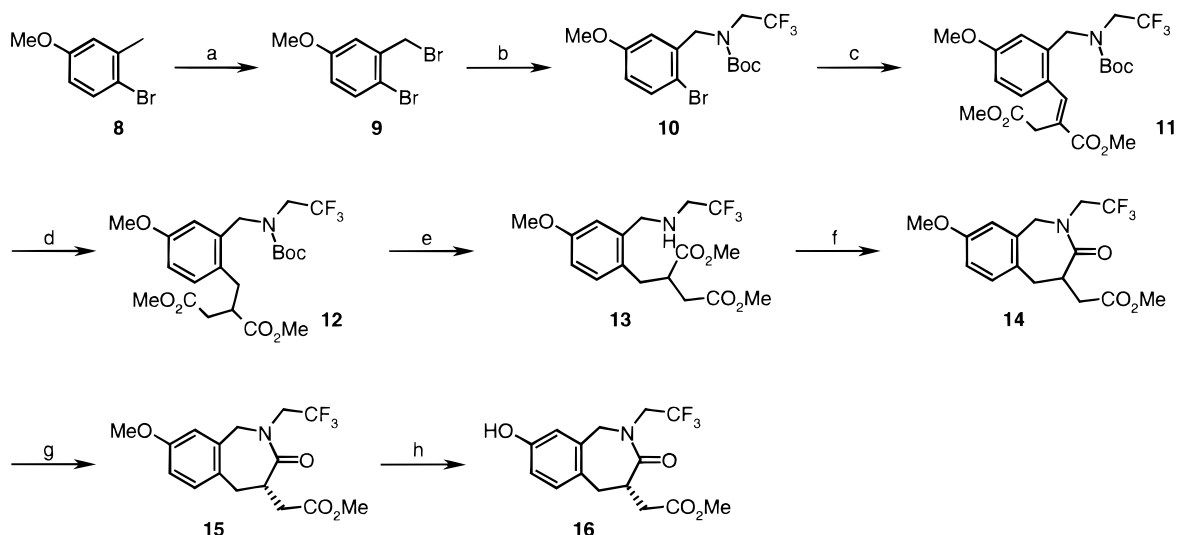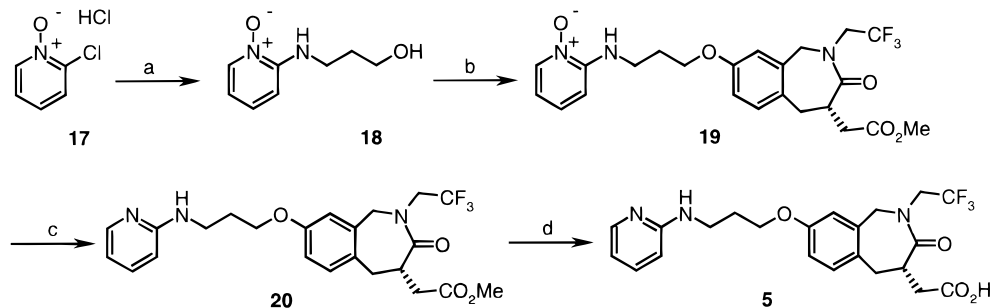

Figure 1. Strategy for modification of **1**.

Scheme 1[a]

[a] (a) NBS, (BzO)$_2$, $h\nu$, CH$_2$Cl$_2$ (81%); (b) NaN(Boc)CH$_2$CF$_3$, DMF (77%); (c) dimethyl itaconate, Pd(OAc)$_2$, P(o-tol)$_3$, (i-Pr)$_2$NEt, CH$_3$CH$_2$CN, reflux (92%); (d) H$_2$, Pd/C, EtOAc (90%); (e) TFA, anisole, CH$_2$Cl$_2$ (86%); (f) (n-Pr)$_3$N, TFA, xylenes, reflux (81%); (g) chiral HPLC (46%, 99+% ee); (h) BBr$_3$, CH$_2$Cl$_2$ (99%).

Scheme 2[a]

[a] (a) 3-Amino-1-propanol, NaHCO$_3$, *tert*-amyl alcohol, reflux (96%); (b) **16**, Ph$_3$P, DIAD, THF (75%); (c) cyclohexene, 10% Pd/C, i-PrOH, reflux (76%); (d) 1.0 N NaOH, dioxane, then 1.0 N HCl (86%).
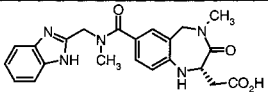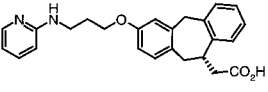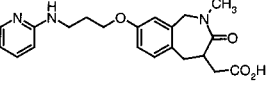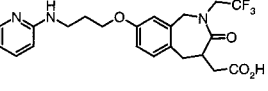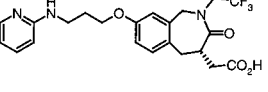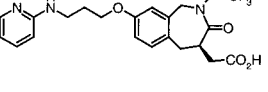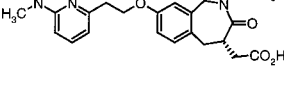
Scheme 3[a]
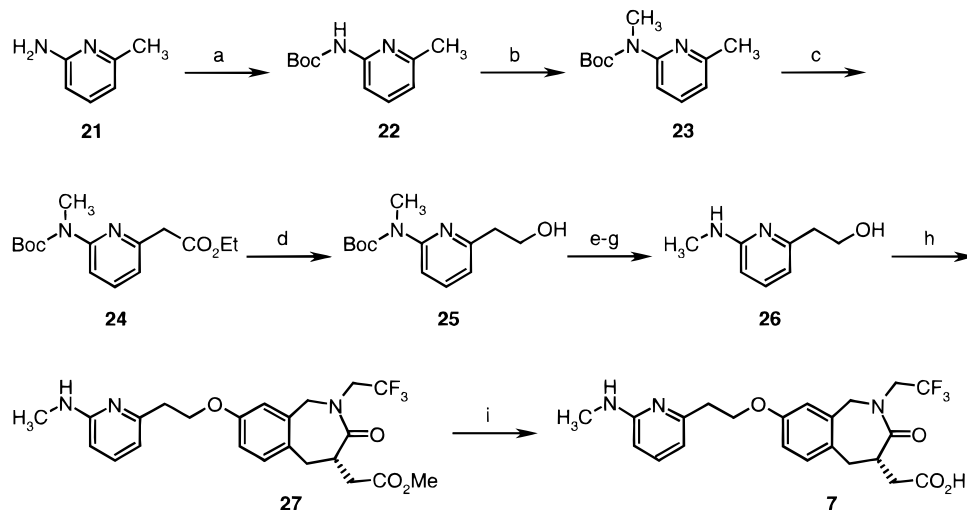
[a] (a) (Boc)$_2$O, neat, 50 °C (99%); (b) NaH, CH$_3$I, DMF (87%); (c) LDA, (EtO)$_2$C=O, THF, 0 °C (100%); (d) LiBH$_4$, THF, reflux (100%); (e) 4 N HCl/dioxane, anisole, then aq NaOH; (f) HCO$_2$H, EtOAc; (g) aq NH$_4$OH (52% from **25**); (h) **16**, Ph$_3$P, DIAD, THF (91%); (i) 1.0 N NaOH, MeOH, then acetic acid (82%).

cerned about potential oxidative instability. We therefore elected to replace the aniline nitrogen with a methylene group, as we expected these groups to be interchangeable based on the results of previous SAR studies. We also chose to incorporate aminopyridine Arg mimetics,[33] as our previous studies indicated that 1,4-benzodiazepine derivatives containing aminopyridine Arg mimetics had somewhat improved oral bioavailability relative to those with a benzimidazole Arg mimetic.[22] Our initial target was benzazepine **3**, which

Table 1. In Vitro Activity and Pharmacokinetic Profiles of Nonpeptide $\alpha v \beta 3$ Antagonists

Number	Structure	$\alpha v \beta 3$ Binding K_i (nM)	$\alpha IIb \beta 3$ Binding K_i (nM)	$\alpha v \beta 3$/HEK Cell Adhesion IC_{50} (nM)	$T_{1/2}$ (min)	Clp (mL/min/kg)	Oral F (%)
1		2.0 ± 0.1	$30{,}000 \pm 2000$	145	9-16	35 ± 2	3-7
2		4.0 ± 0.3	9000 ± 2000	60	192 ± 79	4 ± 2	≈ 100
3		1.9 ± 0.1	$30{,}000 \pm 3000$	75	16 ± 3	50 ± 4	9 ± 0.5
4		1.3 ± 0.2	9000 ± 1000	53	25 ± 3	63 ± 14	19 ± 13
5		0.9 ± 0.2	3300 ± 700	12	360 ± 32	16 ± 5	34 ± 10
6		20.0 ± 2.0	---	390	19 ± 5	60 ± 7	4 ± 2
7		1.2 ± 0.2	3800 ± 1000	3	53 ± 19	25 ± 1	72 ± 16

was prepared by the general methods described previously and above.

The benzazepine ether derivative **3** has $K_i = 1.9$ nM in an $\alpha v \beta 3$ binding assay,[36] while affinity for the related RGD-binding integrin, $\alpha IIb \beta 3$,[37] is considerably lower ($K_i = 30\,000$ nM). Benzazepine **3** also has $IC_{50} = 75$ nM in an $\alpha v \beta 3$-mediated cell adhesion assay,[38] which measures the affinity of a compound for $\alpha v \beta 3$ in a cellular context. Since **3** is racemic, whereas both **1** and **2** are the single, biologically active (*S*)-enantiomers, these results clearly indicate that benzazepine ethers are potent $\alpha v \beta 3$ antagonists with improved activity relative to both the 1,4-benzodiazepine series (**1**) and the tricyclic series (**2**).

The pharmacokinetic profile[39] of benzazepine **3** in rats is not appreciably different from that of **1** (Table 1). The compounds have similar half-lives and oral bioavailabilities, while the plasma clearance is only marginally higher in **3**. Although **3** (ClogP = 2.6) is more lipophilic than **1** (ClogP = 1.4), the modest increase in lipophilicity was not sufficient to confer improved oral bioavailability. We therefore investigated methods for further increasing lipophilicity.

In the benzazepine series, lipophilicity can be conveniently increased by appropriate manipulation of the 2-position substituent. An investigation into 2-position substituents revealed a trend toward improved in vitro

biological activity and improved oral bioavailability as lipophilicity is increased. For example, the trifluoromethyl-substituted derivative **4**, with ClogP of 3.9, has $K_i = 1.3$ nM in the $\alpha v \beta 3$ binding assay and $IC_{50} = 53$ nM in the cell adhesion assay. In addition, **4** appears to have somewhat improved oral bioavailability in rats relative to **3** (19% for **4** vs 9% for **3**). Although individually none of these results are significantly different from those of **3**, taken together they seem to suggest that increasing lipophilicity has had a positive effect on both in vitro biological activity and oral bioavailability.

Since **4** has good biological activity, as well as good oral bioavailability, the individual enantiomers were prepared and evaluated. As expected, the (*S*)-enantiomer **5**, with $K_i = 0.9$ nM in the $\alpha v \beta 3$ binding assay and $IC_{50} = 12$ nM in the cell adhesion assay, was more active than the (*R*)-enantiomer **6**, which has $K_i = 20$ nM in the $\alpha v \beta 3$ binding assay and $IC_{50} = 390$ nM in the cell adhesion assay. Interestingly, however, the pharmacokinetic profiles in rats were very different. The (*S*)-enantiomer **5** has an outstanding pharmacokinetic profile, with a long half-life (360 min), low to moderate clearance (16 mL/min/kg), and good oral bioavailability (34%). In contrast, the pharmacokinetic profile of the (*R*)-enantiomer **6** is relatively poor, with a short half-life (19 min), high clearance (60 mL/min/kg), and low oral bioavailability (4%). The reasons for the differences

in the pharmacokinetic profiles of the enantiomers are not clear at this time but may be due to enantiospecific clearance or metabolism.

Interestingly, incorporation of the isomeric 6-(methylamino)pyridine Arg mimetic[22] has a significant impact on both biological activity and oral bioavailability. Compound **7** has $K_i = 1.2$ nM in the $\alpha v \beta 3$ binding assay, comparable to that of **5**, but has $IC_{50} = 3$ nM in the cell adhesion assay, which is a 4-fold improvement relative to **5**. Further, **7** has improved oral bioavailability in rats (72%).

The affinity of **5** and **7** for other RGD-binding integrins was evaluated, and both were found to bind to the closely related αv integrin, $\alpha v \beta 5$, with low nanomolar affinity (**5** $\alpha v \beta 5$ $K_i = 0.6 \pm 0.2$ nM; **7** $\alpha v \beta 5$ $K_i = 0.3 \pm 0.1$ nM). In contrast, the compounds both show minimal affinity for either $\alpha IIb \beta 3$ (**5** $\alpha IIb \beta 3$ $K_i = 3300 \pm 700$ nM; **7** $\alpha IIb \beta 3$ $K_i = 3800 \pm 1000$ nM) or $\alpha 5 \beta 1$ (**5** $\alpha 5 \beta 1$ $K_i = 1000 \pm 400$ nM; **7** $\alpha 5 \beta 1$ $K_i = 110 \pm 30$ nM). Consistent with their low affinity for $\alpha IIb \beta 3$, both compounds have $IC_{50} > 200$ μM at inhibiting human platelet aggregation.[40]

In biological studies,[41,42] both **5** and **7** were found to be active in models of bone resorption and osteoporosis. In an in vitro human osteoclast resorption assay,[43] which measures the ability of a compound to inhibit human osteoclast-mediated bone resorption, compound **5** has $IC_{50} = 29$ nM and compound **7** has $IC_{50} = 11$ nM. Both compounds were also active in the in vivo thyroidectomized−parathyroidectomized (TPTx) rat model of bone resorption, which measures the ability of a compound to inhibit the parathyroid hormone-stimulated calcemic response in hypocalcemic TPTx rats.[35] In this model, on continuous intravenous infusion, compound **5** has $EC_{50} = 35$ μM and compound **7** has $EC_{50} = 20$ μM. The compounds were also tested in the ovariectomized (Ovx) rat model of osteoporosis, a well-established model for evaluating the potential of a compound to prevent bone loss associated with estrogen deficiency. Significantly, on twice a day oral dosing in the Ovx rat, both **5** (at 5, 15, and 60 mg/kg) and **7** (at 3, 10, and 30 mg/kg) inhibited bone loss in a dose-dependent fashion. For both compounds, the level of inhibition was greater than 50% at the highest dose. The results of these biological studies will be described in greater detail elsewhere.

In conclusion, we have discovered a new class of potent and orally bioavailable $\alpha v \beta 3$ antagonists based on a 2-benzazepine Gly-Asp. Two representatives from this class, benzazepines **5** and **7**, inhibit bone resorption in vitro and in vivo and are orally active in an Ovx rat model of osteoporosis. These results suggest that $\alpha v \beta 3$ antagonists have the potential to be orally administered drugs for the treatment of osteoporosis in humans. Further evaluation of members of this benzazepine class of $\alpha v \beta 3$ antagonists for the treatment of other human diseases, including restenosis following PTCA and diseases involving neovascularization, is ongoing.

References

(1) For recent reviews of $\alpha v \beta 3$, see: (a) Samanen, J.; Jonak, Z.; Rieman, D.; Yue, T.-L. Vascular Indications for Integrin αv Antagonists. *Curr. Pharm. Des.* **1997**, *3*, 545−584. (b) Haubner, R.; Finsinger, D.; Kessler, H. Stereoisomeric Peptide Libraries and Peptidomimetics for Designing Selective Inhibitors of the $\alpha v \beta 3$ Integrin for a New Cancer Therapy. *Angew. Chem., Int. Ed. Engl.* **1997**, *36*, 1374−1389. (c) Horton, M. A. The $\alpha v \beta 3$ Integrin "Vitronectin Receptor". *Int. J. Biochem. Cell Biol.* **1997**, *29*, 721−725.

(2) Van der Pluijm, G.; Mouthaan, H.; Baas, C.; de Groot, H.; Papapoulos, S.; Löwik, C. Integrins and Osteoclastic Resorption in Three Bone Organ Cultures: Differential Sensitivity to Synthetic Arg-Gly-Asp Peptides During Osteoclast Formation. *J. Bone Miner. Res.* **1994**, *9*, 1021−1028.

(3) Helfrich, M. H.; Nesbitt, S. A.; Horton, M. A. Integrins on Rat Osteoclasts: Characterization of Two Monoclonal Antibodies (F4 and F11) to Rat $\beta 3$. *J. Bone Miner. Res.* **1992**, *7*, 345−351.

(4) Helfrich, M. H.; Nesbitt, S. A.; Dorey, E. L.; Horton, M. A. Rat Osteoclasts Adhere to a Wide Range of RGD (Arg-Gly-Asp) Peptide-Containing Proteins, Including the Bone Sialoproteins and Fibronectin, via a $\beta 3$ Integrin. *J. Bone Miner. Res.* **1992**, *7*, 335−343.

(5) Horton, M. A.; Taylor, M. L.; Arnett, T. R.; Helfrich, M. H. Arg-Gly-Asp (RGD) Peptides and the Anti-Vitronectin Receptor Antibody 23C6 Inhibit Dentine Resorption and Cell Spreading by Osteoclasts. *Exp. Cell Res.* **1991**, *195*, 368−375.

(6) Matsuno, H.; Stassen, J. M.; Vermylen, J.; Deckmyn, H. Inhibition of Integrin Function by a Cyclic RGD-Containing Peptide Prevents Neointima Formation. *Circulation* **1994**, *90*, 2203−2206.

(7) Choi, E. T.; Engel, L.; Callow, A. D.; Sun, S.; Trachtenberg, J.; Santoro, S.; Ryan, U. S. Inhibition of Neointimal Hyperplasia by Blocking $\alpha v \beta 3$ Integrin with a Small Peptide Antagonist G*penGRGDSPC*A. *J. Vasc. Surgery* **1994**, *19*, 125−134.

(8) Liaw, L.; Almeida, M.; Hart, C. E.; Schwartz, S. M.; Giachelli, C. M. Osteopontin Promotes Vascular Cell Adhesion and Spreading and is Chemotactic for Smooth Muscle Cells In Vitro. *Circ. Res.* **1994**, *74*, 214−224.

(9) Eliceiri, B. P.; Cheresh, D. A. The Role of αv Integrins During Angiogenesis: Insights into Potential Mechanisms of Action and Clinical Development. *J. Clin. Invest.* **1999**, *103*, 1227−1230.

(10) Engleman, V. W.; Nickols, G. A.; Ross, F. P.; Horton, M. A.; Griggs, D. W.; Settle, S. L.; Ruminski, P. G.; Teitelbaum, S. L. A Peptidomimetic Antagonist of the $\alpha v \beta 3$ Integrin Inhibits Bone Resorption In Vitro and Prevents Osteoporosis In Vivo. *J. Clin. Invest.* **1997**, *99*, 2284−2292.

(11) Storgard, C. M.; Stupack, D. G.; Jonczyk, A.; Goodman, S. L.; Fox, R. I.; Cheresh, D. A. Decreased Angiogenesis and Arthritic Disease in Rabbits Treated with an $\alpha v \beta 3$ Antagonist. *J. Clin. Invest.* **1999**, *103*, 47−54.

(12) Stupack, D. G.; Storgard, C. M.; Cheresh, D. A. A Role for Angiogenesis in Rheumatoid Arthritis. *Braz. J. Med. Biol. Res.* **1999**, *32*, 573−581.

(13) Carron, C. P.; Meyer, D. M.; Pegg, J. A.; Engleman, V. W.; Nickols, M. A.; Settle, S. L.; Westlin, W. F.; Ruminski, P. G.; Nickols, G. A. A Peptidomimetic Antagonist of the Integrin $\alpha v \beta 3$ Inhibits Leydig Cell Tumor Growth and the Development of Hypercalcemia of Malignancy. *Cancer Res.* **1998**, *58*, 1930−1935.

(14) Yun, Z.; Menter, D. G.; Nicolson, G. L. Involvement of in $\alpha v \beta 3$ Cell Adhesion, Motility, and Liver Metastasis of Murine RAW117 Large Cell Lymphoma. *Cancer Res.* **1996**, *56*, 3103−3111.

(15) Brooks, P. C.; Strömblad, S.; Klemke, R.; Visscher, D.; Sarkar, F. H.; Cheresh, D. A. Antiintegrin $\alpha v \beta 3$ Blocks Human Breast Cancer Growth and Angiogenesis in Human Skin. *J. Clin. Invest.* **1995**, *96*, 1815−1822.

(16) Brooks, P. C.; Montgomery, A. M. P.; Rosenfeld, M.; Reisfeld, R. A.; Hu, T.; Klier, G.; Cheresh, D. A. Integrin $\alpha v \beta 3$ Antagonists Promote Tumor Regression by Inducing Apoptosis of Angiogenic Blood Vessels. *Cell* **1994**, *79*, 1157−1164.

(17) Friedlander, M.; Theesfeld, C. L.; Sugita, M.; Fruttiger, M.; Thomas, M. A.; Chang, S.; Cheresh, D. A. Involvement of Integrins $\alpha v \beta 3$ and $\alpha v \beta 5$ in Ocular Neovascular Diseases. *Proc. Natl. Acad. Sci. U.S.A.* **1996**, *93*, 9764−9769.

(18) Hammes, H.-P.; Brownlee, M.; Jonczyk, A.; Sutter, A.; Preissner, K. T. Subcutaneous Injection of a Cyclic Peptide Antagonist of Vitronectin Receptor-Type Integrins Inhibits Retinal Neovascularization. *Nature Medicine* **1996**, *2*, 529−533.

(19) Keenan, R. M.; Miller, W. H.; Kwon, C.; Ali, F. E.; Callahan, J. F.; Calvo, R. R.; Hwang, S. M.; Kopple, K. D.; Peishoff, C. E.; Samanen, J. M.; Wong, A. S.; Yuan, C. K.; Huffman, W. F. Discovery of Potent Nonpeptide Vitronectin Receptor ($\alpha v \beta 3$) Antagonists. *J. Med. Chem.* **1997**, *40*, 2289−2292.

(20) Keenan, R. M.; Miller, W. H.; Lago, M. A.; Ali, F. E.; Bondinell, W. E.; Callahan, J. F.; Calvo, R. R.; Cousins, R. D.; Hwang, S.-M.; Jakas, D. R.; Ku, T. W.; Kwon, C.; Nguyen, T. T.; Reader, V. A.; Rieman, D. J.; Ross, S. T.; Takata, D. T.; Uzinskas, I. N.; Yuan, C. C. K.; Smith, B. R. Benzimidazole Derivatives as Arginine Mimetics in 1,4-Benzodiazepine Nonpeptide Vitronectin Receptor ($\alpha v \beta 3$) Antagonists. *Bioorg. Med. Chem. Lett.* **1998**, *8*, 3165−3170.

(21) Keenan, R. M.; Lago, M. A.; Miller, W. H.; Ali, F. E.; Cousins, R. D.; Hall, L. B.; Hwang, S.-M.; Jakas, D. R.; Kwon, C.; Louden, C.; Nguyen, T. T.; Ohlstein, E. H.; Rieman, D. J.; Ross, S. T.;

Samanen, J. M.; Smith, B. R.; Stadel, J.; Takata, D. T.; Vickery, L.; Yuan, C. C. K.; Yue, T.-L. Discovery of an Imidazopyridine Containing 1,4-Benzodiazepine Nonpeptide Vitronectin Receptor Antagonist with Efficacy in a Restenosis Model. *Bioorg. Med. Chem. Lett.* **1998**, *8*, 3171−3176.

(22) Keenan, R. M.; Miller, W. H.; Barton, L. S.; Bondinell, W. E.; Cousins, R. D.; Eppley, D. F.; Hwang, S.-M.; Kwon, C.; Lago, M. A.; Nguyen, T. T.; Smith, B. R.; Uzinskas, I. N.; Yuan, C. C. K. Nonpeptide Vitronectin Receptor Antagonists Containing 2-Aminopyridine Arginine Mimetics. *Bioorg. Med. Chem. Lett.* **1999**, *9*, 1801−1806.

(23) Miller, W. H.; Bondinell, W. E.; Cousins, R. D.; Erhard, K. F.; Jakas, D. R.; Keenan, R. M.; Ku, T. W.; Newlander, K. A.; Ross, S. T.; Haltiwanger, R. C.; Bradbeer, J.; Drake, F. H.; Gowen, M.; Hoffman, S. J.; Hwang, S.-M.; James, I. E.; Lark, M. W.; Lechowska, B.; Rieman, D. J.; Stroup, G. B.; Vasko-Moser, J. A.; Zembryki, D. L.; Azzarano, L. M.; Adams, P. C.; Salyers, K. L.; Smith, B. R.; Ward, K. W.; Johanson, K. O.; Huffman, W. F. Highly Orally Bioavailable Nonpeptide Vitronectin Receptor Antagonists with Efficacy in an Osteoporosis Model. *Bioorg. Med. Chem. Lett.* **1999**, *9*, 1807−1812.

(24) Mousa, S. A.; Lorelli, W.; Mohamed, S.; Batt, D. G.; Jadhav, P. K.; Reilly, T. M. $\alpha v\beta 3$ Integrin Binding Affinity and Specificity of SM256 in Various Species. *J. Cardiovasc. Pharmacol.* **1999**, *33*, 641−646.

(25) Rockwell, A. L.; Rafalski, M.; Pitts, W. J.; Batt, D. G.; Petraitis, J. J.; DeGrado, W. F.; Mousa, S.; Jadhav, P. K. Rapid Synthesis of RGD Mimetics with Isoxazoline Scaffolds on Solid Phase: Identification of $\alpha v\beta 3$ Antagonists Lead Compounds. *Bioorg. Med. Chem. Lett.* **1999**, *9*, 937−942.

(26) Corbett, J. W.; Graciani, N. R.; Mousa, S. A.; DeGrado, W. F. Solid-Phase Synthesis of a Selective $\alpha v\beta 3$ Integrin Antagonist Library. *Bioorg. Med. Chem. Lett.* **1997**, *7*, 1371−1376.

(27) Nicolau, K. C.; Trujillo, J. I.; Jandeleit, B.; Chibale, K.; Rosenfeld, M.; Diefenbach, B.; Cheresh, D. A.; Goodman, S. L. Design, Synthesis and Biological Evaluation of Nonpeptide Integrin Antagonists. *Bioorg. Med. Chem.* **1998**, *6*, 1185−1208.

(28) Miller, W. H.; Newlander, K. A.; Eggleston, D. S.; Haltiwanger, R. C. Synthesis of a 2-Benzazepine Analogue of a Potent, Nonpeptide GPIIb/IIIa Antagonist. *Tetrahedron Lett.* **1995**, *36*, 373−376.

(29) Methyl (±)-8-methoxy-3-oxo-2-(2,2,2-trifluoroethyl)-2,3,4,5-tetrahydro-2-benzazepine-4-acetate was resolved into its enantiomers by chiral HPLC using the following conditions: Daicel Chiralcel OJ column (21.2 × 250 mm), methanol mobile phase, 15 mL/min flowrate, UV detection at 295 nm, 400 mg injection; t_R for methyl (*R*)-(+)-8-methoxy-3-oxo-2-(2,2,2-trifluoroethyl)-2,3,4,5-tetrahydro-2-benzazepine-4-acetate = 4.9 min; t_R for methyl (*S*)-(−)-8-methoxy-3-oxo-2-(2,2,2-trifluoroethyl)-2,3,4,5-tetrahydro-2-benzazepine-4-acetate = 6.6 min.

(30) Tables of crystal data, fractional atomic coordinates, anisotropic thermal parameters for non-hydrogen atoms, and bond distances and angles have been deposited with the Cambridge Crystallographic Data Centre, Lensfield Road, Cambridge CB2 1EW, United Kingdom. Listings of structure factors are available from the authors upon request.

(31) Hughes, D. L. The Mitsunobu Reaction. *Org. React.* **1992**, *42*, 335−656.

(32) We have found through studies in a related series that the *N*-oxide derivative **18** gave much better yields in the Mitsunobu reaction that the corresponding free aminopyridine (R. D. Cousins, unpublished results).

(33) 2-Aminopyridine derivatives have been reported as Arg mimetics in the discovery of novel thrombin inhibitors; see: Misra, R. N.; Kelly, Y. F.; Brown, B. R.; Roberts, D. G. M.; Chong, S.; Seiler, S. M. Argatroban Analogues: Synthesis, Thrombin Inhibitory Activity and Cell Permeability of Aminoheterocyclic Guanidine Surrogates. *Bioorg. Med. Chem. Lett.* **1994**, *4*, 2165−2170.

(34) Ward, K. W.; Azzarano, L. M.; Bondinell, W. E.; Cousins, R. D.; Huffman, W. F.; Jakas, D. R.; Keenan, R. M.; Ku, T. W.; Lundberg, D.; Miller, W. H.; Mumaw, J. A.; Newlander, K. A.; Pirhalla, J. L.; Roethke, T. J.; Salyers, K. L.; Souder, P. R.; Stelman, G. J.; Smith, B. R. Preclinical Pharmacokinetics and Interspecies Scaling of a Novel Vitronectin Receptor Antagonist. *Drug Metab. Dispos.* **1999**, *27*, 1232−1241.

(35) Lark, M. W.; Stroup, G. B.; Hwang, S.-M.; James, I. E.; Rieman, D. J.; Drake, F. H.; Bradbeer, J.; Mathur, A.; Erhard, K. F.; Newlander, K. A.; Ross, S. T.; Salyers, K. L.; Smith, B. R.; Miller, W. H.; Huffman, W. F.; Gowen, M. Design and Characterization of an Orally Active Arg-Gly-Asp Peptidomimetic Vitronectin Receptor Antagonist, SB 265123, for the Prevention of Bone Loss in Osteoporosis. *J. Pharmacol. Exp. Ther.* **1999**, *291*, 612−617.

(36) Binding affinity for human $\alpha v\beta 3$ was determined in a competitive binding assay employing ^{3}H-SK&F 107260 as the displaced ligand. The K_i values represent the means of values determined in two to three separate experiments; see: Wong, A.; Hwang, S. M.; McDevitt, P.; McNulty, D.; Stadel, J. M.; Johanson, K. Studies on $\alpha v\beta 3$/Ligand Interactions Using a [^{3}H]SK&F-107260 Binding Assay. *Mol. Pharmacol.* **1996**, *50*, 529−537.

(37) Binding affinity for human $\alpha IIb\beta 3$ was determined in a competitive binding assay employing ^{3}H-SK&F 107260 as the displaced ligand. The K_i values represent the means of values determined in two to three separate experiments; see: Ali, F. E.; Bennett, D. B.; Calvo, R. R.; Elliott, J. D.; Hwang, S.-M.; Ku, T. W.; Lago, M. A.; Nichols, A. J.; Romoff, T. T.; Shah, D. H.; Vasko, J. A.; Wong, A. S.; Yellin, T. O.; Yuan, C.-K.; Samanen, J. M. Conformationally Constrained Peptides and Semipeptides Derived from RGD as Potent Inhibitors of the Platelet Fibrinogen Receptor and Platelet Aggregation. *J. Med. Chem.* **1994**, *37*, 769−780.

(38) Affinity for human $\alpha v\beta 3$ in a cellular context was determined by measuring the inhibition of the adhesion of purified human vitronectin to HEK 293 cells transfected with human $\alpha v\beta 3$. The IC$_{50}$ values represent the means of values determined in two experiments in triplicate with an internal standard; see: Kumar, C. S.; James, I. E.; Wong, A.; Mwang, V.; Field, J. A.; Nuthulaganti, P.; Conner, J. R.; Eichman, C.; Ali, F.; Hwang, S. M.; Rieman, D. J.; Drake, F. H.; Gowen, M. Cloning and Characterization of a Novel Integrin $\beta 3$ Subunit. *J. Biol. Chem.* **1997**, *272*, 16390−16397.

(39) Pharmacokinetic parameters were determined using noncompartmental analysis of plasma concentration of test compound versus time data determined in two to three separate animals. Oral bioavailability was calculated from the dose-normalized iv and po AUC values, where AUC is the area under the plasma concentration versus time curve; see: Gibaldi, M.; Perrier, D. Absorption Kinetics and Bioavailability. In *Pharmacokinetics*, 2nd ed.; Swarbrick, J., Ed.; Marcel Dekker: New York, 1982; Vol. 15, pp 145−198.

(40) Antiaggregatory activity was assessed by measuring inhibition of ADP-induced platelet aggregation in human platelet-rich plasma; see: Samanen, J.; Ali, F. E.; Romoff, T.; Calvo, R.; Sorenson, E.; Vasko, J.; Storer, B.; Berry, D.; Bennett, D.; Strohsacker, M.; Powers, D.; Stadel, J.; Nichols, A. Development of a Small RGD Peptide Fibrinogen Receptor Antagonist with Potent Antiaggregatory Activity In Vitro. *J. Med. Chem.* **1991**, *34*, 3114−3125.

(41) Lark, M. W.; Stroup, G. B.; Cousins, R. D.; James, I. E.; Hwang, S. M.; Bradbeer, J.; Hoffman, S.; Lechowska, B.; Vasko-Moser, J.; Zembryki, D.; Votta, B. J.; Lee-Rykaczewski, E.; Perng, D.; Salyers, K.; Smith, B. R.; Liang, X.; Dodds, R. A.; Newlander, K. A.; Ross, S. T.; Erhard, K. F.; Huffman, W. F.; Drake, F. H.; Miller, W. H.; Gowen, M. An Orally Active Vitronectin Receptor $\alpha v\beta 3$ Antagonist Prevents Bone Resorption In Vitro and In Vivo in the Ovariectomized Rat. *Bone* **1998**, *23*, S219.

(42) Lark, M. W.; Stroup, G. B.; Dodds, R. A.; Kapadia, R.; Hwang, S.; James, I. E.; Rieman, D. J.; Liang, X.; Ward, K.; Smith, B. R. The Orally Active Vitronectin Receptor Antagonist, SB 273005, Prevents Trabecular Bone Loss in the Ovariectomized (Ovx) Rat. *J. Bone Miner. Res.* **1999**, *14*, S482.

(43) James, I. E.; Lark, M. W.; Zembryki, D.; Lee-Rykaczewski, E. V.; Hwang, S. M.; Tomaszek, T. A.; Belfiore, P.; Gowen, M. Development and Characterization of a Human In Vitro Resorption Assay: Demonstration of Utility Using Novel Antiresorptive Agents. *J. Bone Miner. Res.* **1999**, *14*, 1562−1569.

JM990446U