Supporting Material

A Convergent Solution Phase Synthesis of the Macrocycle, Ac-Phe-[Orn-Pro-D-Cha-Trp-Arg], a Potent New Anti-Inflammatory Drug.

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General Considerations. All amino acid derivatives were purchased from Novabiochem and used as received. DMF and TFA were “peptide grade” obtained from Auspep Pty Ltd Australia. THF was HPLC grade (less than 0.01% H₂O). ¹H NMR spectra (500 MHz) were recorded at 298 K in DMSO-d₆ and were referenced to the residual solvent peak δ 2.50 ppm. Assignments and chemical shifts for overlapped signals were determined from TOCSY and DQ-COSY spectra in each case. Standard analytical HPLC conditions were used for all compounds: 30% B to 100% B linear gradient over 20 min followed by a further 10 min at 100% B where solvent B was 90% MeCN, 10% H₂O + 0.1% TFA. Solvent A was H₂O + 0.1% TFA. Column was Phenomenex Luna C18 5 μ 100Å 250x4.6 mm, flow rate 1 mL/min.

Contents:

¹ H NMR spectra of intermediates 6, 8, 10, 12, 2, 18, 20, 24, 1	Pages 2–10
¹ H NMR expansions and TOCSY spectra of 1	Pages 11–15
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6

500 MHz, DMSO-d6

9.636

9.257

18

500 MHz, DMSO-d6

H₂O

2.524

2.155

1.278

S7

DMSO

S9

500 MHz, DMSO-d₆

H₂O

1

500 MHz, DMSO-d₆

DMSO

2.500

1

500 MHz, DMSO-d₆

DMSO

2.500

1.739

1

500 MHz, DMSO-d₆

S12

1 TOCSY 500 MHz, DMSO-d6 298 K

1 TOCSY 500 MHz, DMSO-d₆ 298 K

Reported by User: System

Project Name: paper3d53

SAMPLE INFORMATION

Sample Name: Boc-Orn(Z)-Pro-OMe 6
Sample Type: Unknown
Vial: 80
Injection #: 1
Injection Volume: 5.00 ul
Run Time: 30.0 Minutes
Sample Set Name:

Acquired By: System
Date Acquired: 12/4/02 4:32:15 PM
Acq. Method Set: 30 % B to 100 % B in 20 min
Date Processed: 2/18/03 4:38:26 PM
Processing Method: Peptides 214nm
Channel Name: 214 nm
Proc. Chnl. Descr.: PDA 214.0 nm

Auto-Scaled Chromatogram

	Processed Channel	Retention Time (min)	Area	% Area	Height
1	PDA 214.0 nm	11.321	210868	0.98	28256
2	PDA 214.0 nm	18.837	21214187	99.02	2406811

Reported by User: System

Project Name: paper3d53

SAMPLE INFORMATION

Sample Name: Boc-Phe-Orn(Z)-Pro-OMe 8
Sample Type: Unknown
Vial: 86
Injection #: 1
Injection Volume: 5.00 ul
Run Time: 30.0 Minutes
Sample Set Name:

Acquired By: System
Date Acquired: 11/1/02 11:40:05 AM
Acq. Method Set: 30 % B to 100 % B in 20 min
Date Processed: 2/18/03 4:41:43 PM
Processing Method: Peptides 214nm
Channel Name: 214 nm
Proc. Chnl. Descr.: PDA 214.0 nm

Auto-Scaled Chromatogram

	Processed Channel	Retention Time (min)	Area	% Area	Height
1	PDA 214.0 nm	7.804	711925	2.51	80352
2	PDA 214.0 nm	10.599	283518	1.00	39683
3	PDA 214.0 nm	17.744	1309102	4.61	198382
4	PDA 214.0 nm	20.851	24688573	86.95	2414559
5	PDA 214.0 nm	22.089	545953	1.92	82368
6	PDA 214.0 nm	24.520	540652	1.90	36904
7	PDA 214.0 nm	26.221	313243	1.10	30714

Reported by User: System

Project Name: paper3d53

SAMPLE INFORMATION

Sample Name: Ac-Phe-Orn(Z)-Pro-OMe 10
Sample Type: Unknown
Vial: 87
Injection #: 1
Injection Volume: 5.00 ul
Run Time: 30.0 Minutes
Sample Set Name:

Acquired By: System
Date Acquired: 11/1/02 10:56:15 AM
Acq. Method Set: 30 % B to 100 % B in 20 min
Date Processed: 2/18/03 4:42:08 PM
Processing Method: Peptides 214nm
Channel Name: 214 nm
Proc. Chnl. Descr.: PDA 214.0 nm

Auto-Scaled Chromatogram

	Processed Channel	Retention Time (min)	Area	% Area	Height
1	PDA 214.0 nm	7.787	386860	1.84	46132
2	PDA 214.0 nm	13.213	341693	1.63	51282
3	PDA 214.0 nm	15.963	19210710	91.44	2211279
4	PDA 214.0 nm	17.745	701998	3.34	112151
5	PDA 214.0 nm	21.776	367922	1.75	58567

Reported by User: System

Project Name: paper3d53

SAMPLE INFORMATION

Sample Name: Ac-Phe-Orn(Boc)-Pro-OMe 12
Sample Type: Unknown
Vial: 15
Injection #: 1
Injection Volume: 5.00 ul
Run Time: 30.0 Minutes
Sample Set Name:

Acquired By: System
Date Acquired: 1/24/03 1:56:07 PM
Acq. Method Set: 30 % B to 100 % B in 20 min
Date Processed: 2/18/03 4:42:57 PM
Processing Method: Peptides 214nm
Channel Name: 214 nm
Proc. Chnl. Descr.: PDA 214.0 nm

Auto-Scaled Chromatogram

	Processed Channel	Retention Time (min)	Area	% Area	Height
1	PDA 214.0 nm	15.172	14227272	97.85	1851999
2	PDA 214.0 nm	17.116	312407	2.15	47257

Reported by User: System

Project Name: paper3d53

SAMPLE INFORMATION

Sample Name: Ac-Phe-Orn(Boc)-Pro-OH 2
Sample Type: Unknown
Vial: 84
Injection #: 1
Injection Volume: 5.00 ul
Run Time: 30.0 Minutes
Sample Set Name:

Acquired By: System
Date Acquired: 12/4/02 7:28:18 PM
Acq. Method Set: 30 % B to 100 % B in 20 min
Date Processed: 2/18/03 4:43:32 PM
Processing Method: Peptides 214nm
Channel Name: 214 nm
Proc. Chnl. Descr.: PDA 214.0 nm

Auto-Scaled Chromatogram

	Processed Channel	Retention Time (min)	Area	% Area	Height
1	PDA 214.0 nm	13.018	14028348	98.92	1787155
2	PDA 214.0 nm	13.455	153729	1.08	20695

Reported by User: System

Project Name: paper3d53

SAMPLE INFORMATION

Sample Name: Boc-Trp(For)-Arg-OEt 18
Sample Type: Unknown
Vial: 85
Injection #: 1
Injection Volume: 5.00 ul
Run Time: 30.0 Minutes
Sample Set Name:

Acquired By: System
Date Acquired: 12/4/02 8:12:11 PM
Acq. Method Set: 30 % B to 100 % B in 20 min
Date Processed: 2/18/03 4:25:36 PM
Processing Method: Peptides 280nm
Channel Name: 280 nm
Proc. Chnl. Descr.: PDA 280.0 nm

Auto-Scaled Chromatogram

	Processed Channel	Retention Time (min)	Area	% Area	Height
1	PDA 280.0 nm	10.669	129880	2.97	21572
2	PDA 280.0 nm	12.863	38417	0.88	6675
3	PDA 280.0 nm	13.153	4075640	93.17	557253
4	PDA 280.0 nm	16.341	73180	1.67	11017
5	PDA 280.0 nm	16.652	57388	1.31	8550

Reported by User: System

Project Name: paper3d53

SAMPLE INFORMATION

Sample Name: Boc-dCha-Trp(For)-Arg-OEt 20
Sample Type: Unknown
Vial: 86
Injection #: 1
Injection Volume: 5.00 ul
Run Time: 30.0 Minutes
Sample Set Name:

Acquired By: System
Date Acquired: 12/4/02 8:56:14 PM
Acq. Method Set: 30 % B to 100 % B in 20 min
Date Processed: 2/18/03 4:25:58 PM
Processing Method: Peptides 280nm
Channel Name: 280 nm
Proc. Chnl. Descr.: PDA 280.0 nm

Auto-Scaled Chromatogram

	Processed Channel	Retention Time (min)	Area	% Area	Height
1	PDA 280.0 nm	15.291	96395	6.12	6468
2	PDA 280.0 nm	16.811	59099	3.75	10051
3	PDA 280.0 nm	16.996	1419532	90.13	188484

Reported by User: System

Project Name: paper3d53

SAMPLE INFORMATION

Sample Name: Ac-F-Orn(Boc)-P-Cha-W-R-OH 22
Sample Type: Unknown
Vial: 87
Injection #: 1
Injection Volume: 5.00 ul
Run Time: 30.0 Minutes
Sample Set Name:

Acquired By: System
Date Acquired: 12/4/02 9:40:19 PM
Acq. Method Set: 30 % B to 100 % B in 20 min
Date Processed: 2/18/03 4:26:12 PM
Processing Method: Peptides 280nm
Channel Name: 280 nm
Proc. Chnl. Descr.: PDA 280.0 nm

Auto-Scaled Chromatogram

	Processed Channel	Retention Time (min)	Area	% Area	Height
1	PDA 280.0 nm	15.771	1609718	94.47	225078
2	PDA 280.0 nm	16.932	94261	5.53	10282

Reported by User: System

Project Name: paper3d53

SAMPLE INFORMATION

Sample Name: Ac-Phe-Orn-Pro-dCha-Trp-Arg-OH24
Sample Type: Unknown
Vial: 22
Injection #: 1
Injection Volume: 10.00 ul
Run Time: 30.0 Minutes
Sample Set Name: tripeptides

Acquired By: System
Date Acquired: 2/18/03 2:27:53 PM
Acq. Method Set: 30 % B to 100 % B in 20 min
Date Processed: 2/18/03 4:26:31 PM
Processing Method: Peptides 280nm
Channel Name: 280 nm
Proc. Chnl. Descr.: PDA 280.0 nm

Auto-Scaled Chromatogram

	Processed Channel	Retention Time (min)	Area	% Area	Height
1	PDA 280.0 nm	11.692	2222592	93.41	350634
2	PDA 280.0 nm	13.547	156717	6.59	16221

Reported by User: System

Project Name: paper3d53

SAMPLE INFORMATION

Sample Name: Ac-[Phe-Orn-Pro-dCha-Trp-Arg] 1
Sample Type: Unknown
Vial: 40
Injection #: 1
Injection Volume: 10.00 ul
Run Time: 30.0 Minutes
Sample Set Name:

Acquired By: System
Date Acquired: 12/6/02 11:28:31 AM
Acq. Method Set: 30 % B to 100 % B in 20 min
Date Processed: 2/18/03 4:43:44 PM
Processing Method: Peptides 214nm
Channel Name: 214 nm
Proc. Chnl. Descr.: PDA 214.0 nm

Auto-Scaled Chromatogram

	Processed Channel	Retention Time (min)	Area	% Area	Height
1	PDA 214.0 nm	14.045	211503	1.54	32255
2	PDA 214.0 nm	14.344	13506620	98.46	1987423