RESEARCH FRONT

Full Paper

CSIRO PUBLISHING

Aust. J. Chem. **2011**, *64*, 540–544

www.publish.csiro.au/journals/ajc

Combinatorial Solid-Phase Synthesis of 6-Aryl-1,3,5-triazines via Suzuki Coupling

Jae Wook Lee,[A] *Hyung-Ho Ha,*[B,C] *Marc Vendrell,*[D] *Jacqueline T. Bork,*[A] *and Young-Tae Chang*[B,D,E]

[A]Department of Chemistry, New York University, New York, NY 10003, USA.
[B]Department of Chemistry and Medchem Program of Life Sciences Institute, National University of Singapore, 117543, Singapore.
[C]College of Pharmacy, Sunchon National University, Suncheon, 570-742, Korea.
[D]Singapore Bioimaging Consortium, Agency for Science, Technology and Research (A*STAR), Singapore, 138667, Singapore.
[E]Corresponding author. Email: chmcyt@nus.edu.sg

A synthetic methodology to prepare collections of trisubstituted aryl 1,3,5-triazines with broad structural diversity via Suzuki coupling has been developed. We first optimized the combinatorial derivatization of the triazine core using Suzuki cross-coupling. Second, in order to further expand the methodology for the preparation of negatively charged triazines, we adapted this approach to polymer-supported amino acids and prepared aryl triazines with different charge distribution. With a collection of 160 aryl triazine derivatives in good purities and without any purification step, we proved the viability of this orthogonal scheme for the preparation of triazine libraries using amine/amino acid-captured solid supports and Suzuki cross-coupling.

Manuscript received: 20 January 2011.
Manuscript accepted: 17 February 2011.

Introduction

Combinatorial chemistry is a powerful method for the synthesis of large and structurally distinct libraries with broad molecular diversity.[1–4] In conjugation with high-throughput screenings, combinatorial chemistry has demonstrated its value for the generation and optimization of lead compounds in drug discovery.[5] One of the most popular techniques for the rapid production of combinatorial libraries is based on the use of solid-phase organic synthesis (SPOS). Major advantages of SOPS include the ease of product isolation by filtration and the use of an excess of reagents to force reactions to completion, leading to an overall reduction of the total time required for the synthesis. The 1,3,5-triazine scaffold is of particular interest in combinatorial chemistry approaches due to its synthetic accessibility, high chemical stability, and drug-like hydrogen bonding properties. Moreover, a broad range of biological activities have been reported for these structures, such as anticancer,[6–9] antimicrobial,[10] and antiretroviral compounds,[11] inhibitors of cyclin-dependent kinases (CDKs) and p38 mitogen-activated protein kinases (MAPKs),[12,13] oestrogen receptor modulators,[14] or inosine monophosphate dehydrogenase,[15] and tubulin polymerization inhibitors.[16,17] Such a wide range of biological activities has been the main driving force to put a considerable effort into the synthesis of large libraries of triazine derivatives.[18,19] Our research group has reported the synthesis of combinatorial triazine libraries and proved their potent biological activity for several applications, such as microtubule destabilization[17] or inhibition of β-amyloid plaques aggregation.[20] Whereas the activities of these molecules depend on the aromatic and aliphatic substituents of the triazine core, fine-tuning their interaction with specific proteins may require the incorporation of wider chemical diversification. In order to broaden the molecular diversity of triazine compounds, we envisioned that the combinatorial adaptation of Suzuki coupling for the derivatization of the triazine core would allow the construction of libraries using infrequent boronic acids as building blocks for the main diversity source. Furthermore, we considered the introduction of negative charges in the substituents of the triazines to increase the number of possible interactions with their target proteins. For this purpose, amino acids are excellent building blocks, mainly due to their bioavailability, structural and charge diversity, and good accessibility. Since the implementation of boronic acids within the aryl triazine core required the optimization of an orthogonal synthetic scheme, we designed a solid-phase methodology for the preparation of polyethyleneglycol (PEG)-tagged aryl triazines via Suzuki coupling that was compatible with the incorporation of charge distribution variability. The preparation of a collection of 160 PEG-tagged aryl triazines (Chart 1) in high purities and without any purification demonstrates the adequacy of this approach for combinatorial chemistry syntheses.

Results and Discussion

We first designed the synthesis of PEG-tagged triazines with aryl diversity via Suzuki coupling according to Scheme 1. A set of solid supports was prepared by coupling 10 amine building

10.1071/CH11034 0004-9425/11/050540

blocks to a Peptide Amide Linker polystyrene (PAL-PS) resin via reductive amination in the presence of $NaBH(OAc)_3$. PAL-PS resins are compatible with palladium-catalyzed cross-coupling conditions, and have been reported for the solid-phase preparation of triazine libraries.[21–23]

After derivatization of cyanuric chloride with *N*-Boc-2,2′-(ethylenedioxy)diethylamine in solution phase, the resulting *N*-Boc-2-(ethylenedioxy)diethylamine-4,6-dichloro-1,3,5-triazine was coupled to 10 different amine resins by nucleophilic substitution in THF at 60°C using *N*,*N*-Diisopropylethylamine (DIEA) as a base. Afterwards, resin-bound chlorotriazines were derivatized under Suzuki cross-coupling conditions with a set of aryl boronic acids (Chart 2) to yield the trisubstituted aryl 1,3,5-triazines.

Finally, the treatment of the resins with a TFA-DCM (1:9) solution released 80 different 2-(ethylenedioxy)diethylamine-4-alkylamino-6-aryl-1,3,5-triazines. While we observed that the purity of some products showed dependency on the nature of the substituents within the building blocks (e.g. boronic acids A, F, and M, Chart 2) exhibited a relatively lower purity than the rest of the library) most of the compounds could be isolated in very good purities without any purification step (average purity: 88%, Table 1).

In view of the successful adaptation of the Suzuki cross-coupling derivatization to a combinatorial triazine library approach, we aimed to extend the structural and charge diversity with the use of amino acids as building blocks. We prepared eight different resins by loading amino acids with aliphatic (alanine, valine, leucine, isoleucine) and aromatic (phenylalanine, tyrosine) side chains, as well as polar ones (serine, asparagine). After attaching 2-(ethylenedioxy)diethylamine-4,6-dichloro-1,3,5-triazine to the resins by nucleophilic substitution and carrying out Suzuki coupling derivatization as above mentioned, we obtained 80 negatively charged trisubstituted triazines in very high purities without any purification (average purity: 92%, Table 2). Remarkably, none of these synthetic steps required the protection of the carboxylic acid groups of the amino acids, which facilitated the optimization of a general protocol and improved the overall synthetic yields.

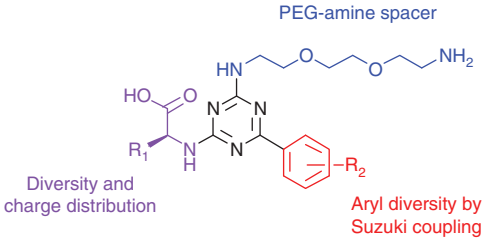
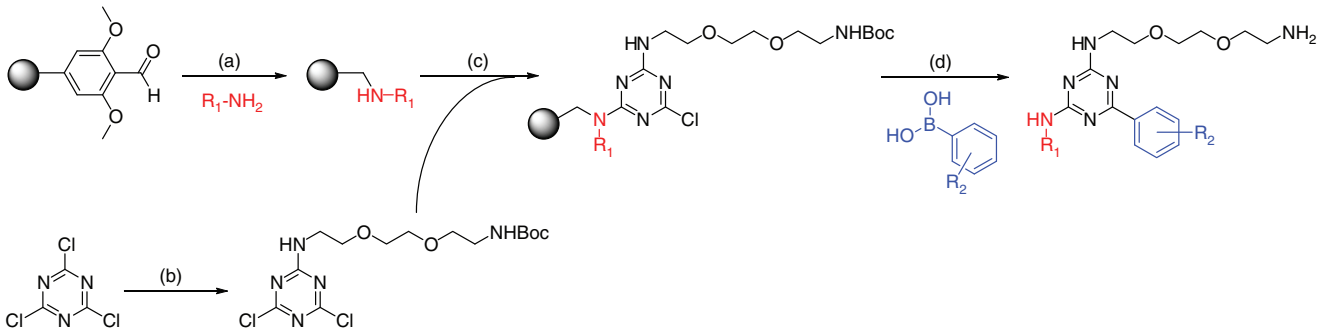


Chart 1. General structure for the library of differently charged PEG-tagged aryl 1,3,5-triazines.

Conclusions

In the present work, we optimized an orthogonal approach for the solid-phase synthesis of PEG-tagged triazines with aryl diversity via Suzuki coupling. A 160-member aryl triazine



Scheme 1. Synthesis of differently charged trisubstituted 1,3,5-triazines. Reagents and conditions: (a) (i) amino acid or amine building block, AcOH-THF (2:98), rt, 1 h, (ii) $NaBH(OAc)_3$, AcOH-THF (2:98), rt, 8 h; (b) *N*-Boc-2,2′-(ethylenedioxy)diethylamine, DIEA, THF, 0°C, 2 h; (c) DIEA, THF, 60°C, 3 h; (d) (i) boronic acid building blocks, $Pd(PPh_3)_4$, Cs_2CO_3, dioxane, 90°C, 15 h, (ii) TFA-DCM (1:9), rt, 0.5 h.

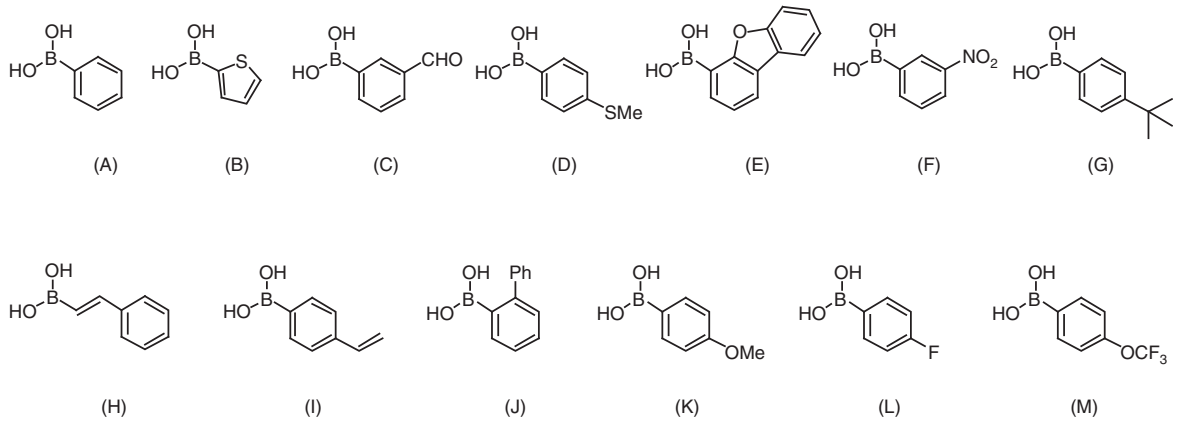


Chart 2. Boronic acids used for the library synthesis.

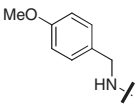
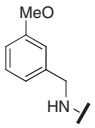
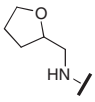
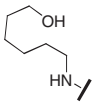
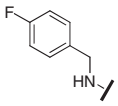
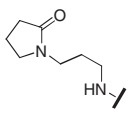
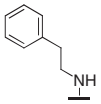
library was designed in order to incorporate broad structural diversity and different charge distribution by using different amine and amino acid-captured solid supports. The optimized synthetic strategy enabled the incorporation of several boronic acids as building blocks and different charge distribution via diverse amino acids and amines, and proved to be compatible with a wide range of chemical functionalities, asserting its application in combinatorial chemistry syntheses. In addition to employing non-protected building blocks, with this approach we isolated the final triazine compounds in very good purities, without any purification steps. The screening of the biological properties of this library is currently ongoing.

Experimental

Materials and Methods

Unless otherwise noted, materials and solvents were obtained from commercial suppliers (Acros and Aldrich) and were used without further purification. PAL-aldehyde (4-formyl-3,5-dimethoxyphenoxymethyl) resin from Midwest Bio-Tech (Cat #: 20840, Lot #: SY03470, loading: 1.10 mmol g^{-1}) was used for the generation of library compounds. The scaffold *N*-Boc-2-(ethylenedioxy)diethylamine-4,6-dichloro-1,3,5-triazine was prepared by solution-phase chemistry and purified by normal-phase flash column chromatography on Sorbent Technologies silica gel, 60 Å (63–200 mesh). TLC was carried out on SAI

Table 1. Purities of isolated 6-aryl-1,3,5-triazines with amine-captured solid supports
Purities were determined by integration of absorbance peaks at 254 nm

R_1	R_2							
	A	B	K	E	F	G	L	M
	85	88	90	97	80	87	90	83
	85	90	93	95	82	88	87	83
	85	90	92	95	89	95	90	85
	85	82	93	98	71	90	90	83
	85	82	94	97	90	92	91	86
	85	85	90	97	85	88	89	80
	85	90	95	96	82	90	92	83
	80	90	89	98	90	87	90	85
	85	90	90	95	83	94	95	82
	88	93	83	91	75	95	93	82

F254 pre-coated silica gel plates (250 μm layer thickness). All library products were characterized by HPLC-MS (Agilent Technology, HP1100) using a C_{18} column (20 × 4.0 mm^2) and a gradient of 5–95% CH_3CN-H_2O (containing 0.1% AcOH) over 4 min. Condensation reactions were performed in a standard heat block from VWR Scientific Products using 4-mL glass vials with paper-lined caps purchased from Fisher Scientific. Resin filtration procedures were carried out in 70-μm PE-frit cartridges from Applied Separations (Cat. #: 2449).

Loading of Amino Acids and Amines onto PAL Resin via Reductive Amination

To a suspension of 4-formyl-3,5-dimethoxyphenoxymethyl-functionalized polystyrene resin (PAL) (1.0 g, 1.1 mmol) in THF (40 mL) were added the building blocks R_1 (5.5 mmol), followed by the addition of AcOH (0.9 mL). After shaking at rt for 1 h, $NaBH(OAc)_3$ (1.63 g, 7.7 mmol) was added, and the reaction was further shaken at rt for 8 h. Using PE-frit cartridges, the solvents and excess reagents were filtered off and the resins were washed with DMF, DCM, and MeOH (20 mL × 3) and dried under nitrogen gas.

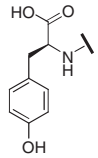
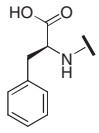
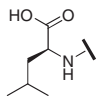
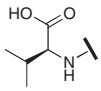
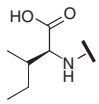
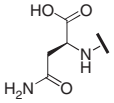
Resin Capture of Triazine Scaffold by Amine or Amino Acid Substitution

To a suspension of the resin-bound amines or amino acids (125 mg, 0.13 mmol) in THF (2.5 mL) was added *N*-Boc-2-(ethylenedioxy)diethylamine-4,6-dichloro-1,3,5-triazine (0.4 mmol, 3 equiv.), followed by addition of DIEA (0.8 mmol, 6 equiv.). The reaction vial was placed in a heating block at 60°C for 2.5 h, and afterwards the solvents and excess reagents were filtered off through a PE-frit cartridge and washed with DMF, DCM, and MeOH (3 mL × 3 for every solvent).

Suzuki Cross-coupling Reactions

$Pd(PPh_3)_4$ (7 mg, 6.06 μmol) and Cs_2CO_3 (35 mg, 0.1 mmol) were dispensed to the different resin-bound triazines (10 mg, 11 μmol) in a glove box. Dioxane (1 mL) and boronic acids were distributed to each reaction vial, and the vials were shaken 90°C

Table 2. Purities of isolated 6-aryl-1,3,5-triazines with amino acid-captured solid supports
Purities were determined by integration of absorbance peaks at 254 nm

R_1	R_2									
	A	B	C	D	E	F	G	H	I	J
	98	95	95	92	91	80	99	80	93	98
	99	97	95	95	99	80	95	90	95	90
	95	98	96	92	95	99	99	90	99	95
	99	99	99	88	99	92	99	70	95	96
	99	99	95	85	99	95	99	94	95	97
	94	98	95	89	97	94	99	80	90	80
	95	93	94	80	96	96	94	80	90	99
	99	96	92	90	91	99	98	99	65	65

for 15 h under inert atmosphere. Excess reagents were filtered off, and the resins were washed with a solution of 0.05 M sodium diethyldithiocarbamate in DMF (1 mL × 5), and DMF, DCM, and MeOH (3 mL × 3 for every solvent).

Resin Cleavage

A solution of TFA-DCM (1:9) was added to the resins, which were shaken at rt for 30 min. The final compounds were filtered from the resin, washed with DCM, and evaporated under vacuum. An average recovery yield around 80% was estimated for the final compounds on the basis of the intensities of their HPLC absorbance peaks.

Accessory Publication

A list of amino acids and amine building blocks used for the library synthesis, representative HPLC-MS data for the library of PEG-tagged trisubstituted triazines are available on the Journal's website.

Acknowledgements

The authors gratefully acknowledge the financial support from the National Institute of Health (CA-96912) and the National University of Singapore (NUS) (Young Investigator Award: R-143-000-353-123). The authors thank Francis B. Peters for his critical reading of the manuscript.

References

[1] A. Nefzi, J. M. Ostresh, R. A. Houghten, *Chem. Rev.* **1997**, *97*, 449. doi:10.1021/CR960010B

[2] R. G. Franzén, *J. Comb. Chem.* **2000**, *2*, 195. doi:10.1021/CC000002F

[3] V. Krchnák, M. W. Holladay, *Chem. Rev.* **2002**, *102*, 61. doi:10.1021/CR010123H

[4] R. E. Ziegert, J. Torang, K. Knepper, S. Brase, *J. Comb. Chem.* **2005**, *7*, 147. doi:10.1021/CC049879V

[5] D. Falb, S. Jindal, *Curr. Opin. Drug Discov. Dev.* **2002**, *5*, 532.

[6] A. Dhainaut, G. Regnier, A. Tizot, A. Pierre, S. Leonce, N. Guilbaud, L. Kraus-Berthier, G. Atassi, *J. Med. Chem.* **1996**, *39*, 4099. doi:10.1021/JM960361I

[7] N. Baindur, N. Chadha, B. M. Brandt, D. Asgari, R. J. Patch, C. Schalk-Hihi, T. E. Carver, I. P. Petrounia, C. A. Baumann, H. Ott, C. Manthey, B. A. Springer, M. R. Player, *J. Med. Chem.* **2005**, *48*, 1717. doi:10.1021/JM049372Z

[8] M. Zheng, C. Xu, J. Ma, Y. Sun, F. Du, H. Liu, L. Lin, C. Li, J. Ding, K. Chen, H. Jiang, *Bioorg. Med. Chem.* **2007**, *15*, 1815. doi:10.1016/J.BMC.2006.11.028

[9] F. Saczewski, A. Bulakowska, *Eur. J. Med. Chem.* **2006**, *41*, 611. doi:10.1016/J.EJMECH.2005.12.012

[10] K. Srinivas, U. Srinivas, K. Bhanuprakash, K. Harakishore, U. S. Murthy, V. J. Rao, *Eur. J. Med. Chem.* **2006**, *41*, 1240. doi:10.1016/J.EJMECH.2006.05.013

[11] D. W. Ludovici, R. W. Kavash, M. J. Kukla, C. Y. Ho, H. Ye, B. L. De Corte, K. Andries, M. P. de Bethune, H. Azijn, R. Pauwels, H. E. Moereels, J. Heeres, L. M. Koymans, M. R. de Jonge, K. J. Van Aken, F. F. Daeyaert, P. J. Lewi, K. Das, E. Arnold, P. A. Janssen, *Bioorg. Med. Chem. Lett.* **2001**, *11*, 2229. doi:10.1016/S0960-894X(01)00411-5

[12] K. Leftheris, G. Ahmed, R. Chan, A. J. Dyckman, Z. Hussain, K. Ho, J. Hynes, Jr, J. Letourneau, W. Li, S. Lin, A. Metzger, K. J. Moriarty, C. Riviello, Y. Shimshock, J. Wen, J. Wityak, S. T. Wrobleski, H. Wu, J. Wu, M. Desai, K. M. Gillooly, T. H. Lin, D. Loo, K. W. McIntyre, S. Pitt, D. R. Shen, D. J. Shuster, R. Zhang, D. Diller, A. Doweyko, J. Sack, J. Baldwin, J. Barrish, J. Dodd, I. Henderson, S. Kanner, G. L. Schieven, M. Webb, *J. Med. Chem.* **2004**, *47*, 6283. doi:10.1021/JM049521D

[13] G. H. Kuo, A. Deangelis, S. Emanuel, A. Wang, Y. Zhang, P. J. Connolly, X. Chen, R. H. Gruninger, C. Rugg, A. Fuentes-Pesquera, S. A. Middleton, L. Jolliffe, W. V. Murray, *J. Med. Chem.* **2005**, *48*, 4535. doi:10.1021/JM040214H

[14] B. R. Henke, T. G. Consler, N. Go, R. L. Hale, D. R. Hohman, S. A. Jones, A. T. Lu, L. B. Moore, J. T. Moore, L. A. Orband-Miller, R. G. Robinett, J. Shearin, P. K. Spearing, E. L. Stewart, P. S. Turnbull, S. L. Weaver, S. P. Williams, G. B. Wisely, M. H. Lambert, *J. Med. Chem.* **2002**, *45*, 5492. doi:10.1021/JM020291H

[15] W. J. Pitts, J. Guo, T. G. Dhar, Z. Shen, H. H. Gu, S. H. Watterson, M. S. Bednarz, B. C. Chen, J. C. Barrish, D. Bassolino, D. Cheney, C. A. Fleener, K. A. Rouleau, D. L. Hollenbaugh, E. J. Iwanowicz, *Bioorg. Med. Chem. Lett.* **2002**, *12*, 2137. doi:10.1016/S0960-894X(02)00351-7

[16] E. El Fahime, M. Bouchentouf, B. F. Benabdallah, D. Skuk, J. F. Lafreniere, Y. T. Chang, J. P. Tremblay, *Biochem. Cell Biol.* **2003**, *81*, 81. doi:10.1139/O03-054

[17] H. S. Moon, E. M. Jacobson, S. M. Khersonsky, M. R. Luzung, D. P. Walsh, W. Xiong, J. W. Lee, P. B. Parikh, J. C. Lam, T. W. Kang, G. R. Rosania, A. F. Schier, Y. T. Chang, *J. Am. Chem. Soc.* **2002**, *124*, 11 608. doi:10.1021/JA026720I

[18] J. T. Bork, J. W. Lee, S. M. Khersonsky, H. S. Moon, Y. T. Chang, *Org. Lett.* **2003**, *5*, 117. doi:10.1021/OL027195V

[19] S. M. Khersonsky, Y. T. Chang, *J. Comb. Chem.* **2004**, *6*, 474. doi:10.1021/CC049965V

[20] W. Kim, Y. Kim, J. Min, D. J. Kim, Y. T. Chang, M. H. Hecht, *ACS Chem. Biol.* **2006**, *1*, 461. doi:10.1021/CB600135W

[21] J. V. Wade, C. A. Krueger, *J. Comb. Chem.* **2003**, *5*, 267. doi:10.1021/CC020061O

[22] Z. Hu, T. Ma, Z. Chen, Z. Ye, G. Zhang, Y. Lou, Y. Yu, *J. Comb. Chem.* **2009**, *11*, 267. doi:10.1021/CC800157K

[23] J. W. Lee, J. T. Bork, H. H. Ha, A. Samanta, Y. T. Chang, *Aust. J. Chem.* **2009**, *62*, 1000. doi:10.1071/CH09153