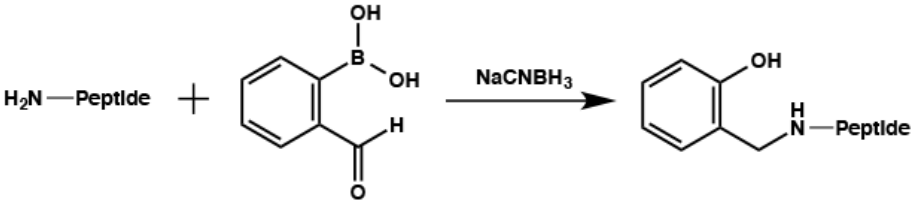
SAND2015-7544J

Isomer-sensitive deboronation in reductive aminations of aryl boronic acids

Leave this area blank for abstract info.

Brad H. Jones, David R. Wheeler, Jill S. Wheeler, Lance L. Miller, Todd M. Alam, and Erik D. Spoerke



Tetrahedron Letters
journal homepage: www.elsevier.com

Isomer-sensitive deboronation in reductive aminations of aryl boronic acids

Brad H. Jones[a,*], David R. Wheeler[b], Jill S. Wheeler[c], Lance L. Miller[d], Todd M. Alam[a], and Erik D. Spoerke[c]

[a]*Organic Materials Science, Sandia National Laboratories, Albuquerque, NM 87185 USA*
[b]*Special Technologies, Sandia National Laboratories, Albuquerque, NM 87185, USA*
[c]*Electronic, Optical, and Nano Materials, Sandia National Laboratories, Albuquerque, NM 87185, USA*
[d]*Materials Reliability, Sandia National Laboratories, Albuquerque, NM 87185, USA*

ARTICLE INFO

Article history:
Received
Received in revised form
Accepted
Available online

Keywords:
Boronic acid
Reductive amination
Peptide modification
Deboronation
Formylphenylboronic acid

ABSTRACT

Deboronation is observed during the reductive amination of formylphenylboronic acid (FPBA) to the amine termini and side chains of peptides. This deboronation is sensitive to the isomerism of the boronic acid (BA), with *ortho*-FPBA yielding complete deboronation in the preparation of an *N*-terminally-modified dipeptide. The observed behavior is also clearly mediated by the chemical identity of the amine substrate. These results reveal a previously undocumented subtlety of BA functionalization and highlight the importance of thorough spectroscopic characterization in the preparation of peptide and small molecule BAs.

© 2015 Elsevier Ltd. All rights reserved.

Boronic acids (BAs) are useful functionalities for programmable stimuli-responsive chemistry, due to their Lewis acid character with pK_a values tunable across physiological pH and their reversible binding to saccharides, catechols, and other compounds bearing 1,2-diol moieties.[1] In addition, BAs have emerged as valuable reagents and intermediates in important synthetic pathways, such as Suzuki cross-coupling reactions,[1] and as selective enzyme inhibitors, exemplified by the commercial success of the anti-cancer drug Bortezomib.[2] Over the last decade, researchers have studied a variety of BA-modified peptides, focusing on the inhibitory properties of the BA or the use of the BA as a functional element in peptide self-assembly.[3] For example, our group has recently reported the reversible self-assembly behavior of a BA dipeptide, controllable by pH, ionic strength, and the addition of polyols.[3s]

BAs are frequently attached to peptides, and more generally a variety of small molecules, via conventional acid-amine coupling chemistry or reductive aminations. More specifically, formylphenylboronic acid (FPBA) has been used in solid phase peptide synthesis to graft the phenylboronic acid functionality through lysine side chains of the peptide via reductive amination.[3g,3k,4] In addition, a variety of small molecule BAs have been derived from reductive aminations involving FPBA.[5] Although several pathways are known by which deboronation of BA compounds can occur, most notably by strong oxidants such as peroxides,[1] deboronation has not been reported in reductive aminations. However, some groups have noted instability of *ortho*-FPBAs to the borohydride reducing agents typical of such aminations, resulting in the formation of benzoxaborole species.[5j,5l,5m,5o,6]

Based on the past literature, then, we were surprised to discover that reductive amination of *ortho*-FPBA to the *N*-terminus of a simple dipeptide resulted in a product **(2)** completely devoid of boron. Briefly, the dipeptide of phenylalanine (Phe-Phe), resin-bound at its *C*-terminus and bearing a deprotected *N*-terminus was shaken with an excess of *ortho*-FPBA and sodium cyanoborohydride, $NaCNBH_3$, predissolved in dimethylformamide (DMF). Additional synthetic details are provided in the Supplementary Information. Extensive deboronation was also similarly observed in the reductive amination of *ortho*-FPBA to the lysine side chain of a linear nonapeptide (**3**, Figures S1-S3). In contrast, the reductive amination of *para*-FPBA to the same dipeptide gives a product (**1**) that is more consistent with the expected outcome of amination, *i.e.*, there was little apparent deboronation.

Figure 1 shows the result of electrospray ionization-mass spectrometry (ESI-MS) performed on the *N*-terminally modified peptides obtained using the *para* and *ortho* isomers of FPBA. The spectrum for the *para* product **1** (Figure 1a) shows the expected $[M-H]^-$ ion indicating the successful attachment of FPBA, as well as a small signature of the unmodified dipeptide, $[M-H-FPBA]^-$. Furthermore, the $[M-H]^-$ ion exhibits isotopic splitting characteristic of boron; namely, the principal peak is preceded by a peak less one Da, reflecting the natural occurrence

* Corresponding author. Tel.: +1-505-284-6364; fax: +1-505-844-9781; e-mail: bhjones@sandia.gov

of ^{10}B. The spectrum for the *ortho* product **2** (Figure 1b) instead shows a product that is 18 Da less than the expected $[M-H]^-$ ion. Indeed, this product no longer exhibits the isotopic splitting characteristic of boron, suggesting the elimination of B(OH) from the product. ^{1}H, ^{11}B, and ^{13}C nuclear magnetic resonance (NMR) spectroscopy (Figures S4, S5, and S7) confirms these ESI-MS results. Specifically, the *para* product shows a single ^{11}B resonance with a chemical shift consistent with typical BAs, whereas the *ortho* product shows no ^{11}B signal whatsoever. In addition, the *ortho* product exhibits a distinct ^{13}C resonance at 156 ppm that is not found in the *para* product, consistent with an aromatic carbon adjacent to a hydroxyl substituent.[7] On the basis of these results, we postulate the reactions drawn in Scheme 1.

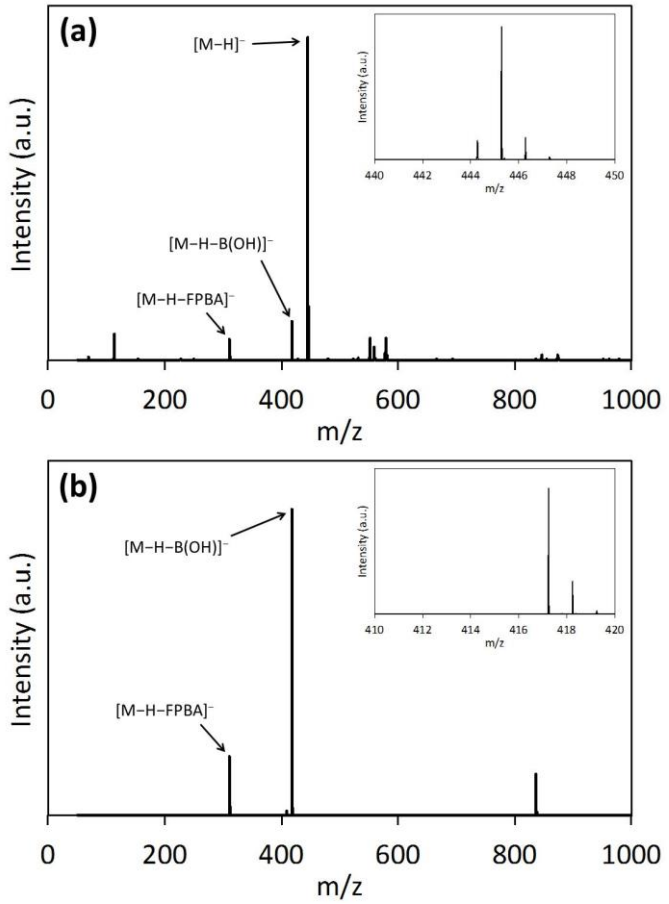


Figure 1. Negative mode ESI-MS of Phe-Phe *N*-terminally modified by reductive amination of **(a)** *para*-FPBA and **(b)** *ortho*-FPBA. M indicates 446.2 Da, the mass of the desired, BA-modified dipeptide. The insets show the isotopic splitting of the highest intensity peak in each spectrum.

We subsequently found that FPBA further exhibits an isomer-sensitive stability of the aldehyde functionality when exposed to the reducing agent $NaCNBH_3$. When *para*-FPBA is exposed to $NaCNBH_3$ in d_7-DMF, ^{1}H NMR spectroscopy shows no apparent change in the identity of the FPBA (Figure 2a). In contrast, when *ortho*-FPBA is exposed to $NaCNBH_3$, the aldehyde functionality is clearly lost, with the appearance of a new proton signal well upfield of the original aldehyde proton (Figure 2b). In addition, the corresponding ^{11}B NMR spectra clearly show that the BA functionality is unstable only in the case of the *ortho* isomer (Figure S9). These data collectively suggest the reduction of *ortho*-FPBA to the corresponding benzoxaborole **4** by $NaCNBH_3$, in agreement with previously reported data.[5j,5l,5m,5o,6] Indeed, the NMR spectra of a commercially available sample of the hemiester of 2-(hydroxymethyl)phenylboronic acid (Figure S10) were consistent with the NMR spectra of the reduced *ortho*-FPBA. Similar results were obtained when CD_3OD was used as the solvent (Figure S11 and Figure S12), although reduction of *para*-FPBA was also evident over longer time scales (Figure S13).**Scheme 1.** Isomer-sensitive deboronation in *N*-terminal modification of Phe-Phe by reductive amination of FPBA.

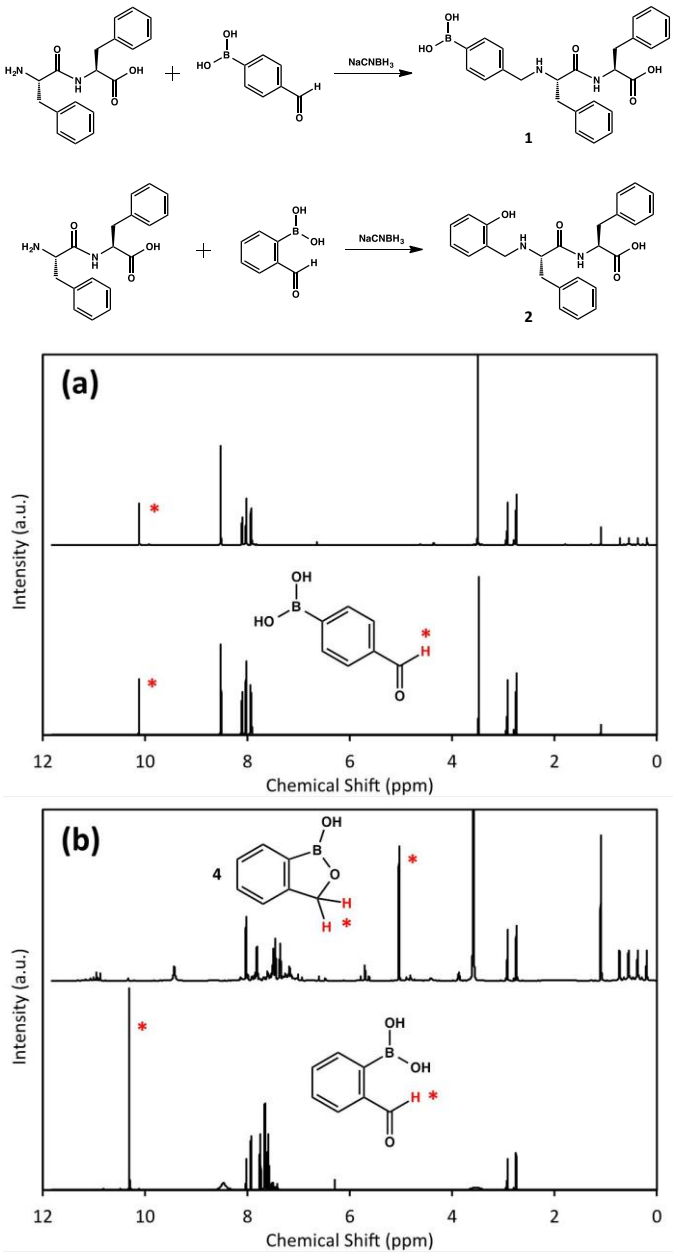
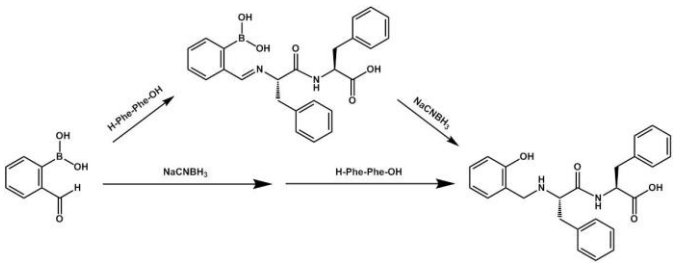


Figure 2. ^{1}H NMR spectra showing isomer-dependent sensitivity of **(a)** *para*-FPBA and **(b)** *ortho*-FPBA to reducing agent $NaCNBH_3$. In both panels, the bottom spectrum shows the neat FPBA in d_7-DMF, while the top spectrum shows the FPBA in d_7-DMF after addition of $NaCNBH_3$. The proton signals indicating the stability of *para*-FPBA to $NaCNBH_3$ and the reduction of *ortho*-FPBA by $NaCNBH_3$ to the corresponding benzoxaborole **4** are denoted by the red asterisks.

These data led us to consider the possibility that isomer-dependent deboronation was associated in some manner with the instability of *ortho*-FPBA to reducing agents. Therefore, we repeated the reductive amination as a two-step process, first reacting the *N*-terminus of the peptide with *ortho*-FPBA to give the usual imine species, and then reducing the imine via $NaCNBH_3$. However, in this case, the identical deboronated

product **2** was obtained (Figure S14). Thus, both the reduction of *ortho*-FPBA to the benzoxaborole followed by exposure to an amine, and the formation of an imine between *ortho*-FPBA and an amine followed by exposure to $NaCNBH_3$, leads to installation of the phenyl ring as a deboronated substituent (Scheme 2).



Scheme 2. A deboronated product is observed independent of the order of amination and reduction.

To provide further insight into the origin of deboronation in these reactions, as well as to explore its generality, we conducted NMR experiments tracking a solution-based reductive amination of FPBA to different primary amines. Figure 3 shows the result of ^{1}H and ^{11}B NMR spectroscopy performed on a mixture of *ortho*-FPBA and H-Phe-Phe-NH_2 in d_7-DMF, to which $NaCNBH_3$ is subsequently added (3:3:1 [FPBA]:[$NaCNBH_3$]:[H-Phe-Phe-NH_2]). H-Phe-Phe-NH_2 was used in this experiment instead of H-Phe-Phe-OH (Scheme 2) because of the poor solubility of the latter in DMF and MeOH. As expected, the ^{1}H spectrum in Figure 3a indicates attachment of a substituent to the *N*-terminus of the peptide, along with the reduction of excess *ortho*-FPBA to benzoxaborole. In addition, a prominent resonance is found at 6.8 ppm, this resonance being absent in the spectra of the benzoxaborole (Figure 2b and Figure S10), H-Phe-Phe-NH_2 (Figure S15), and a mixture of *ortho*-FPBA and H-Phe-Phe-NH_2 (Figure S17). In oxidative deboronation reactions, a distinct upfield shift of aromatic resonances has been previously shown to accompany the conversion of phenylboronic acids to phenols.[8] The ^{11}B spectrum in Figure 3b, as expected, indicates reduction of excess *ortho*-FPBA to the benzoxaborole. However, there is also a distinct resonance at 20 ppm, consistent with the formation of boric acid as the byproduct of deboronation (see Figure S18 for the NMR spectra of neat boric acid).

Interestingly, none of the aforementioned features are observed in the corresponding NMR spectra when a simple aliphatic amine is substituted for the dipeptide in these reductive aminations. Instead, both the ^{1}H and ^{11}B spectra (Figure S19) suggest successful installation of the phenylboronic acid with no conspicuous loss of the boronic acid functionality, *i.e.*, liberation of boric acid. This observation is consistent with the relatively widespread and apparently successful use of *ortho*-FPBA in reductive aminations for the preparation of aminomethylphenylboronic acids.[3g,3k,4-5,9]

Based on this collective information, it is therefore apparent that the deboronation phenomenon observed is influenced by not only the isomerism of the boronic acid, but also the chemical identity of the amine substrate. Related phenomena have been reported recently for several chemical reactions involving boronic acids.[8,10] For example, Fu, *et al.*, reported that secondary amines catalyze air oxidation of aryl boronic acids and boronate esters to the corresponding phenols, while primary and tertiary amines do not, due to the relative basicity of amines: 2° > 1° > 3°.[8b] Their work could implicate the secondary amine resulting from imine reduction as having a critical role in deboronation; however, *ortho*-substituted products, unlike *para*-substituted ones, can form a B-N dative bond accompanied by decreased basicity of the amine,[5a] which is contrary to the poorer stability of the *ortho*-substituted product we have observed. Other work has shown oxidation of aryl boronic acids to be dramatically accelerated by the presence of electron-withdrawing substituents on the phenyl ring, yet the aminomethyl substituents at present are electron-donating in character.[10b] Thus, the mechanism of deboronation at present is decidedly more subtle. Cammidge and coworkers,[8a] and Zhu and coworkers,[10a] have found that catalytic amounts of certain quinones and *N*-oxides, respectively, can effect deboronation of various aryl boronic acids. We suspect a similar process is occurring here, wherein the peptide catalyzes deboronation to which the *ortho*-substituted boronic acid is particularly susceptible.

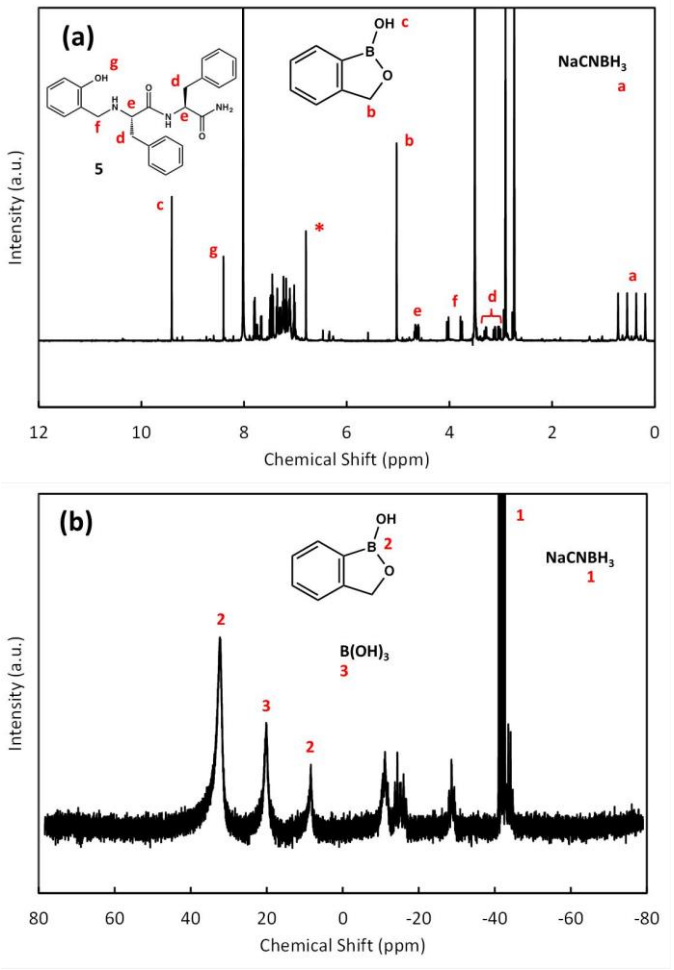


Figure 3. (a) ^{1}H and **(b)** ^{11}B NMR spectra of reductive amination of *ortho*-FPBA to H-Phe-Phe-NH_2 with $NaCNBH_3$ as reducing agent. The solvent is d_7-DMF. Deboronation is suggested by the appearance of a new aromatic resonance in the ^{1}H spectrum, marked by the red asterisk, as well as the appearance of a new resonance in the ^{11}B spectrum corresponding to liberation of boric acid.

Although additional work is required to elucidate further the mechanism and details of deboronation that we have observed, we feel the present results serve as an important cautionary account, particularly in light of increasing interest in peptide and small molecule BAs. ESI-MS is regularly used to confirm the outcome of solid phase peptide synthesis, and deboronation has been reported as an artifact of in-source fragmentation of aryl BAs.[11] However, our combined MS and NMR characterizations have clearly shown that extensive deboronation can occur during

the reductive amination of *ortho*-FPBA to peptide substrates. Therefore, the signatures of deboronation in MS must be given careful consideration when determining the outcome of peptide BA syntheses. If isomerism of the boronic acid is irrelevant, the improved stability of *para*-FPBA to $NaCNBH_3$ appears to advocate its use in reductive amination schemes. Alternatively, we recommend the use of carboxyphenylboronic acids, which can be used to install BAs onto peptides via standard acid-amine coupling chemistry (exemplified by compound **6**, Figures S20-S22) with no apparent deboronation.

Acknowledgments

We gratefully acknowledge James Hochrein for use of MS equipment and Alina Martinez for performing HPLC. This research was supported by the US Department of Energy, Office of Basic Energy Sciences, Division of Materials Science and Engineering, Project KC0203010. Sandia National Laboratories is a multi-program laboratory operated by Sandia Corporation, a wholly owned subsidiary of Lockheed Martin Corporation, for the US Department of Energy's National Nuclear Security Administration under contract DE-AC04-94AL85000.

References

1. Hall, D. G. In *Boronic Acids: Preparation and Applications in Organic Synthesis and Medicine;* Hall, D. G., Ed.; Wiley-VCH: Weinheim, Germany, 2005; pp 1-100.
2. Fu, H.; Fang, H.; Sun, J.; Wang, H.; Liu, A.; Sun, J.; Wu, Z. *Curr. Med. Chem.* **2014**, *21*, 3271-3280.
3. (a) Kettner, C.; Mersinger, L.; Knabb, R. *J. Biol. Chem.* **1990**, *265*, 18289-18297; (b) Elgendy, S.; Deadman, J.; Patel, G.; Green, D.; Chino, N.; Goodwin, C. A.; Scully, M. F.; Kakkar, V. V.; Claeson, G. *Tetrahedron Lett.* **1992**, *33*, 4209-4212; (c) Kimura, T.; Takeuchi, M.; Nagasaki, T.; Shinkai, S. *Tetrahedron Lett.* **1995**, *36*, 559-562; (d) McCormack, T.; Baumeister, W.; Grenier, L.; Moomaw, C.; Plamondon, L.; Pramanik, B.; Slaughter, C.; Soucy, F.; Stein, R.; Zühl, F.; Dick, L. *J. Biol. Chem.* **1997**, *272*, 26103-26109; (e) Kobayashi, H.; Koumoto, K.; Hwa Jung, J.; Shinkai, S. *J. Chem. Soc. Perk. T. 2* **2002**, 1930-1936; (f) Duggan, P. J.; Offermann, D. A. *Aust. J. Chem.* **2007**, *60*, 829-834; (g) Edwards, N. Y.; Sager, T. W.; McDevitt, J. T.; Anslyn, E. V. *J. Am. Chem. Soc.* **2007**, *129*, 13575-13583; (h) LeBeau, A. M.; Singh, P.; Isaacs, J. T.; Denmeade, S. R. *Chem. Biol.* **2008**, *15*, 665-674; (i) Chen, C. S.; Ji, T. J.; Xu, X. D.; Zhang, X. Z.; Zhuo, R. X. *Macromol. Rapid Commun.* **2010**, *31*, 1903-1908; (j) Knott, K.; Fishovitz, J.; Thorpe, S. B.; Lee, I.; Santos, W. L. *Org. Biomol. Chem.* **2010**, *8*, 3451-3456; (k) Bicker, K. L.; Sun, J.; Lavigne, J. J.; Thompson, P. R. *ACS Comb. Sci.* **2011**, *13*, 232-243; (l) Ikeda, M.; Tanida, T.; Yoshii, T.; Hamachi, I. *Adv. Mater.* **2011**, *23*, 2819-2822; (m) Xu, X. D.; Chu, Y. F.; Chen, C. S.; Chen, J. X.; Cheng, S. X.; Zhang, X. Z.; Zhuo, R. X. *Small* **2011**, *7*, 2201-2209; (n) Chen, C. S.; Xu, X. D.; Wang, Y.; Yang, J.; Jia, H. Z.; Cheng, H.; Chu, C. C.; Zhuo, R. X.; Zhang, X. Z. *Small* **2013**, *9*, 920-926; (o) Melicher, M. S.; Chu, J.; Walker, A. S.; Miller, S. J.; Baxter, R. H. G.; Schepartz, A. *Org. Lett.* **2013**, *15*, 5048-5051; (p) Ren, J.; Zhang, Y.; Zhang, J.; Gao, H.; Liu, G.; Ma, R.; An, Y.; Kong, D.; Shi, L. *Biomacromolecules* **2013**, *14*, 3434-3443; (q) Ikeda, M.; Tanida, T.; Yoshii, T.; Kurotani, K.; Onogi, S.; Urayama, K.; Hamachi, I. *Nat. Chem.* **2014**, *6*, 511-518; (r) Neupane, L. N.; Han, S. Y.; Lee, K. H. *Chem. Commun.* **2014**, *50*, 5854-5857; (s) Jones, B. H.; Martinez, A. M.; Wheeler, J. S.; McKenzie, B. B.; Miller, L. L.; Wheeler, D. R.; Spoerke, E. D. *Chem. Commun.* **2015**, DOI: 10.1039/C5CC05207F.
4. Heinrichs, G.; Schellenträger, M.; Kubik, S. *Eur. J. Org. Chem.* **2006**, *2006*, 4177-4186.
5. (a) Wiskur, S. L.; Lavigne, J. J.; Ait-Haddou, H.; Lynch, V.; Chiu, Y. H.; Canary, J. W.; Anslyn, E. V. *Org. Lett.* **2001**, *3*, 1311-1314; (b) Gray, C. W.; Houston, T. A. *J. Org. Chem.* **2002**, *67*, 5426-5428; (c) Johnson, L. L.; Houston, T. A. *Tetrahedron Lett.* **2002**, *43*, 8905-8908; (d) Asano, T.; Nakamura, H.; Uehara, Y.; Yamamoto, Y. *ChemBioChem* **2004**, *5*, 483-490; (e) Bosch, L. I.; Fyles, T. M.; James, T. D. *Tetrahedron* **2004**, *60*, 11175-11190; (f) Luvino, D.; Smietana, M.; Vasseur, J.-J. *Tetrahedron Lett.* **2006**, *47*, 9253-9256; (g) Tan, W.; Zhang, D.; Zhu, D. *Bioorg. Med. Chem. Lett.* **2007**, *17*, 2629-2633; (h) Adamczyk-Woźniak, A.; Brzózka, Z.; Cyrański, M. K.; Filipowicz-Szymańska, A.; Klimentowska, P.; Żubrowska, A.; Żukowski, K.; Sporzyński, A. *Appl. Organomet. Chem.* **2008**, *22*, 427-432; (i) Adamczyk-Woźniak, A.; Cyrański, M. K.; Żubrowska, A.; Sporzyński, A. *J. Organomet. Chem.* **2009**, *694*, 3533-3541; (j) Adamczyk-Woźniak, A.; Cyrański, M. K.; Jakubczyk, M.; Klimentowska, P.; Koll, A.; Kołodziejczak, J.; Pojmaj, G.; Żubrowska, A.; Żukowska, G. Z.; Sporzyński, A. *J. Phys. Chem. A* **2010**, *114*, 2324-2330; (k) Adamczyk-Woźniak, A.; Madura, I.; Velders, A. H.; Sporzyński, A. *Tetrahedron Lett.* **2010**, *51*, 6181-6185; (l) Adamczyk-Woźniak, A.; Fratila, R. M.; Madura, I. D.; Pawełko, A.; Sporzyński, A.; Tumanowicz, M.; Velders, A. H.; Żyła, J. *Tetrahedron Lett.* **2011**, *52*, 6639-6642; (m) Adamczyk-Woźniak, A.; Madura, I.; Pawełko, A.; Sporzyński, A.; Żubrowska, A.; Żyła, J. *Cent. Eur. J. Chem.* **2011**, *9*, 199-205; (n) Adamczyk-Woźniak, A.; Cyrański, M. K.; Frączak, B. T.; Lewandowska, A.; Madura, I. D.; Sporzyński, A. *Tetrahedron* **2012**, *68*, 3761-3767; (o) Adamczyk-Woźniak, A.; Borys, K. M.; Sporzyński, A. *Chem. Rev.* **2015**, *115*, 5224-5247.
6. Akama, T.; Virtucio, C.; Dong, C.; Kimura, R.; Zhang, Y. K.; Nieman, J. A.; Sharma, R.; Lu, X.; Sales, M.; Singh, R.; Wu, A.; Fan, X. Q.; Liu, L.; Plattner, J. J.; Jarnagin, K.; Freund, Y. R. *Bioorg. Med. Chem. Lett.* **2013**, *23*, 1680-1683.
7. Bose, A. K.; Srinivasan, P. R. *Tetrahedron* **1975**, *31*, 3025-3029.
8. (a) Cammidge, A. N.; Goddard, V. H. M.; Schubert, C. P. J.; Gopee, H.; Hughes, D. L.; Gonzalez-Lucas, D. *Org. Lett.* **2011**, *13*, 6034-6037; (b) Fu, Y.; He, Q.; Zhu, D.; Wang, Y.; Gao, Y.; Cao, H.; Cheng, J. *Chem. Commun.* **2013**, *49*, 11266-11268.
9. Hargrove, A. E.; Ellington, A. D.; Anslyn, E. V.; Sessler, J. L. *Bioconjugate Chem.* **2011**, *22*, 388-396.
10. (a) Zhu, C.; Wang, R.; Falck, J. R. *Org. Lett.* **2012**, *14*, 3493-3497; (b) Lozada, J.; Liu, Z.; Perrin, D. M. *J. Org. Chem.* **2014**, *79*, 5365-5368.
11. Wang, L.; Dai, C.; Burroughs, S. K.; Wang, S. L.; Wang, B. *Chemistry* **2013**, *19*, 7587-7594.

Supplementary Material

Supplementary material (details of peptide syntheses and characterization techniques, additional NMR, HPLC, and MS data) associated with this article can be found, in the online version, at.

Isomer-sensitive deboronation in reductive aminations of aryl boronic acids

Brad H. Jones,[a] David R. Wheeler,[b] Jill S. Wheeler,[c] Lance L. Miller,[d] Todd M. Alam,[a] and Erik D. Spoerke[c]

[a] Organic Materials Science, Sandia National Laboratories, Albuquerque, NM 87185 USA
[b] Special Technologies, Sandia National Laboratories, Albuquerque, NM 87185, USA
[c] Electronic, Optical, and Nano Materials, Sandia National Laboratories, Albuquerque, NM 87185, USA
[d] Materials Reliability, Sandia National Laboratories, Albuquerque, NM 87185, USA

Materials

The deuterated solvents d_7-DMF and D_2O were purchased from Sigma-Aldrich. CD_3OD and ND_4OD were purchased from Cambridge Isotopes Laboratories.

The boronic acids *ortho*-formylphenylboronic acid (*ortho*-FPBA), *para*-formylphenylboronic acid (*para*-FPBA), and *para*-carboxyphenylboronic acid (*para*-CPBA) were purchased from Alfa Aesar.

For peptide synthesis, the fluorenylmethyloxycarbonyl (Fmoc) protected amino acids Fmoc-Phe-OH, Fmoc-Ala-OH, Fmoc-Lys(Boc)-OH, Fmoc-Lys(Mtt)-OH, Fmoc-Asp(OtBu)-OH, and Fmoc-Glu(OtBu)-OH were purchased from EMD Millipore. All amino acids were L enantiomers of purity ≥ 99%. Fmoc-Phe-Wang resin, Rink Amide AM resin, and Fmoc-Asp(OtBu)-Wang resin, all 100-200 mesh, were purchased from EMD Millipore. Activating reagents 2-(1H-Benzotriazole-1-yl)-1,1,3,3-tetramethyluronium hexafluorophosphate (HBTU) and (Benzotriazol-1-yloxy)tripyrrolidinophosphonium hexafluorophosphate (PyBOP) were purchased from EMD Millipore. Acetic acid (AcOH, ≥ 98%), 1-hydroxybenzotriazole hydrate (HOBt, 97%), *N*-ethyldiisopropylamine (DIEA, ≥ 98%), hexylamine (99%), piperidine (99%), sodium borohydride ($NaBH_4$, ≥ 98.5%), sodium cyanoborohydride ($NaCNBH_3$, 95%), trifluoroacetic acid (TFA, 98%), and triisopropylsilane (TIS, 99%) were purchased from Sigma-Aldrich. *N*,*N*-

Dimethylformamide (DMF, sequencing grade), dichloromethane (DCM, HPLC grade), and *N*-methyl-2-pyrrolidone (NMP) were purchased from Fisher Scientific.

All materials were used as received without further purification.

Peptide Synthesis

Peptides were synthesized using standard, Fmoc-based, solid-phase protocols on a Liberty Microwave Peptide Synthesizer (CEM Corporation). The peptides Fmoc-Phe-Phe-Wang resin, Fmoc-Phe-Phe-Rink Amide AM resin, and Fmoc-Asp(OtBu)-Glu(OtBu)-$(Ala)_2$-Lys(Mtt)-$(Ala)_2$-Lys(Boc)-Asp(OtBu)-Wang resin were assembled from *C*-terminus to *N*-terminus on 0.1 mmol or 0.25 mmol scales using Fmoc-Phe-Wang resin, Rink Amide AM resin, and Fmoc-Asp(OtBu)-Wang resin, respectively. Fmoc deprotection was accomplished using a 0.1 M solution of HOBt in 20:80 (vol:vol) piperidine:DMF. The resin was treated with two cycles of deprotect solution at 75 °C for 180 s. Fmoc-protected amino acids were coupled to the deprotected amine of the resin-bound peptide using 5 eq. amino acid, 4.5 eq. HBTU, and 10 eq. DIEA relative to the resin loading. Each coupling reaction was conducted at 75 °C for 180 s. The amino acids, HBTU, and DIEA were utilized as 0.2 M solutions in DMF, a 0.5 M solution in HBTU, and a 2 M solution in NMP, respectively.

For Fmoc-Phe-Phe-Wang resin, following the completion of peptide assembly, the Fmoc-protected, resin-bound peptide was washed with DMF (3 x 15 mL) and DCM (3 x 15 mL). The final Fmoc deprotection was conducted manually by shaking the resin in 30:70 (vol:vol) piperidine:NMP for 30 min, followed by additional washing with DMF (3 x 15 mL) and DCM (3 x 15 mL). At this stage, a number of different procedures were attempted to introduce a phenylboronic acid moiety to the *N*-terminus of the peptide. For reductive aminations, a 10-fold excess (relative to the resin loading) of the boronic acid aldehydes *ortho*-FPBA or *para*-FPBA and a 10-fold excess of reducing agents $NaCNBH_3$ or $NaBH_4$ were used. DMF, NMP, and 9:1 DMF:MeOH (vol:vol), with or without 1 vol. % added AcOH, were tested as solvents.

With $NaBH_4$ as reducing agent, a two-step procedure was used. The resin was first shaken over FPBA solution with activated 3 Å molecular sieves for 18 hr. Then, the requisite amount of $NaBH_4$ was added and shaking was continued for an additional 4 hr. The reaction solution was filtered off and the resin then washed sequentially with DMF, MeOH, and DMF (3 x 15 mL each). Finally, the resin was separated from the sieves by floating off in DCM. With $NaCNBH_3$ as reducing agent, both single-step and two-step procedures were used. The single-step procedure involved predissolving the requisite amount of $NaCNBH_3$ and FPBA in solvent, shaking the resin over this solution for 18 hr, filtering off the reaction solution, and finally washing the resin with DMF and DCM. For the two-step procedure, the resin was first shaken over FPBA solution for 4 hr, washed with DMF and DCM, shaken over $NaCNBH_3$ solution for 18 hr, and washed once again with DMF and DCM. Alternatively, the boronic acid functionality was introduced by coupling *para*-CPBA to the *N*-terminus of the peptide. This coupling was achieved by shaking the resin in a 3:3:6-fold excess of *para*-CPBA:PyBOP:DIEA dissolved in 10 mL DMF for 2 hr, followed by washes with DMF and DCM. After each procedure attempted for the attachment of boronic acid, the peptide was cleaved from the resin by shaking in 95:2.5:2.5 (vol:vol:vol) TFA:TIS:H_2O for 3 hr. The peptide solution in TFA was then separated from the resin by filtration, and the peptide was recovered by drying under vacuum. Finally, the peptide was further purified by RP-HPLC. The above-described procedures in which *para*-FPBA was used afforded the product compound **1** (Scheme 1). Yield (0.1 mmol scale): 31 mg (70%). Purity (by RP-HPLC): 99%. ^{1}H-NMR (500 MHz, in D_2O + 0.25 vol. % ND_4OD): δ 7.34 (d, *J* = 7.7 Hz, 2H), 7.04-7.13 (m, 8H), 6.92 (d, *J* = 6.9 Hz, 2H), 6.85 (d, *J* = 7.8 Hz, 2H), 4.29 (q, *J* = 4.7 Hz, 1H), 3.21 (t, *J* = 7.1 Hz, 1H), 3.08 (q, *J* = 12.6 Hz, 2H), 3.01 (dd, *J* = 4.6, 13.9 Hz, 1H), 2.65 (m, 3H). ^{11}B-NMR (160 MHz, in D_2O + 0.25 vol. % ND_4OD): δ 10.9. ^{13}C-NMR (125 MHz, in D_2O + 0.25 vol. % ND_4OD): δ 177.5, 174.4, 137.6, 136.8, 131.8, 129.3, 129.1, 128.6, 127.5, 126.9, 62.8, 56.1, 50.6, 38.7, 38.1. HR-MS (ESI): calculated for $C_{25}H_{27}BN_2O_5$ $[M-H]^-$: 445.1934; found: 445.1961. The above-described procedures in which *ortho*-FPBA was used afforded the deboronated compound **2** (Scheme 1). Yield (0.25 mmol scale): 51 mg (49%). Purity (by RP-HPLC): 96%. ^{1}H-NMR (500

MHz, in D_2O + 0.25 vol. % ND_4OD): δ 7.02-7.17 (m, 11 H), 6.75 (d, *J* = 7.4 Hz, 1H), 6.68 (t, *J* = 7.4 Hz, 1 H), 6.58 (d, *J* = 8.1 Hz, 1 H), 4.38 (q, *J* = 4.7 Hz, 1H), 3.25 (m, 2H), 3.15 (d, *J* = 13.7 Hz, 1H), 3.09 (dd, *J* = 5.0, 14.4 Hz, 1H), 2.76 (m, 2H), 2.59 (dd, *J* = 8.3, 13.7 Hz, 1H). ^{11}B-NMR (160 MHz, in D_2O + 0.25 vol. % ND_4OD): no signal above noise. ^{13}C-NMR (125 MHz, in D_2O + 0.25 vol. % ND_4OD): δ 177.7, 174.3, 156.3, 137.6, 137.1, 129.8, 129.3, 129.1, 128.9, 128.7, 128.6, 126.9, 126.8, 124.3, 119.3, 116.0, 62.6, 56.1, 47.8, 38.6, 38.0. HR-MS (ESI): calculated for $C_{25}H_{26}N_2O_4$ $[M-H]^-$: 417.1814; found: 417.1822. The above-described procedures in which *para*-CPBA was used afforded the product compound **6** (see Figure S20). Yield (0.25 mmol scale): 67 mg (58%). Purity (by RP-HPLC): 98%. ^{1}H-NMR (500 MHz, in D_2O + 0.25 vol. % ND_4OD): δ 7.63 (d, *J* = 8.1 Hz, 2H), 7.39 (d, *J* = 8.1 Hz, 2H), 7.04-7.18 (m, 10H), 4.66 (dd, *J* = 5.4, 10 Hz, 1H), 4.34 (dd, *J* = 4.8, 8.2 Hz, 1H), 3.08 (m, 2H), 2.82 (m, 2H). ^{11}B-NMR (160 MHz, in D_2O + 0.25 vol. % ND_4OD): δ 6.3. ^{13}C-NMR (125 MHz, in D_2O + 0.25 vol. % ND_4OD): δ 177.5, 171.9, 170.4, 137.4, 136.9, 134.1, 133.3, 129.3, 129.1, 128.6, 128.4, 126.9, 126.7, 126.4, 56.1, 55.2, 37.8, 36.6. HR-MS (ESI): calculated for $C_{25}H_{25}BN_2O_6$ $[M-H]^-$: 459.1727; found: 459.1725.

For Fmoc-Phe-Phe-Rink Amide AM resin, following the completion of peptide assembly, final Fmoc deprotection and cleavage from the resin was accomplished as described above. This affords the peptide H-Phe-Phe-NH_2. The peptide was also recovered as described above and purified by RP-HPLC The ^{1}H NMR spectrum was found to be consistent with a previously reported spectrum for this peptide.[1] Yield (0.25 mmol scale): 55 mg (71%). Purity (by RP-HPLC): 98%. ^{1}H-NMR (500 MHz, in d_7-DMF): δ 8.94 (d, *J* = 8.0 Hz, 1H), 8.53 (br, 2H), 7.63 (s, 1H), 7.20-7.40 (m, 10H), 7.18 (s, 1H), 4.70 (m, 1H), 4.43 (q, *J* = 4.6 Hz, 1H), 3.37 (dd, *J* = 4.6, 14.0 Hz, 1H), 3.14 (m, 2H), 3.00 (dd, *J* = 8.9, 13.8 Hz, 1H). HR-MS (ESI): calculated for $C_{18}H_{21}N_3O_2$ $[M+H]^+$: 312.1713; found: 312.1724. Alternatively, with Fmoc-Phe-Phe-Rink Amide AM resin, following the completion of peptide assembly, the final Fmoc deprotection was conducted manually by shaking the resin in 30:70 (vol:vol) piperidine:NMP for 30 min, followed by additional washing with DMF (3 x 15 mL) and DCM (3 x 15 mL). A 10-fold excess (relative to the resin loading) of *ortho*-FPBA and a 10-fold excess of

$NaCNBH_3$ were dissolved in 10 mL NMP with 1 vol. % AcOH. The resin was shaken over this solution for 18 hr, after which the resin was filtered off and washed with DMF (3 x 15 mL) and DCM (3 x 15 mL). The peptide was cleaved from the resin and recovered as described above and further purified by RP-HPLC. This affords the deboronated compound **5**. Yield (0.25 mmol scale): 70 mg (67%). Purity (by RP-HPLC): 98%. ^{1}H-NMR (500 MHz, in CD_3OD): δ 7.04-7.17 (m, 11 H), 6.92 (d, *J* = 7.5 Hz, 1H), 6.83 (t, *J* = 7.4 Hz, 1 H), 6.68 (d, *J* = 7.9 Hz, 1 H), 4.80 (q, *J* = 5.2 Hz, 1H), 3.92 (t, *J* = 7.3 Hz, 1H), 3.19 (m, 2H), 3.13 (d, *J* = 13.0 Hz, 1H), 3.06 (dd, *J* = 5.0, 13.5 Hz, 1H), 2.76 (m, 2H). ^{11}B-NMR (160 MHz, in CD_3OD): no signal above noise. ^{13}C-NMR (125 MHz, in CD_3OD): δ 173.7, 156.0, 137.6, 137.0, 129.1, 128.8, 128.5, 128.2, 127.5, 126.7, 119.5, 118.0, 115.7, 114.8, 68.3, 54.0, 51.5, 37.2, 36.0. HR-MS (ESI): calculated for $C_{25}H_{26}N_2O_4$ $[M+H]^+$: 418.2131; found: 418.2181.

For Fmoc-Asp(OtBu)-Glu(OtBu)-$(Ala)_2$-Lys(Mtt)-$(Ala)_2$-Lys(Boc)-Asp(OtBu)-Wang resin, following the completion of peptide assembly, the Fmoc-protected, resin-bound peptide was washed with DMF (3 x 15 mL) and DCM (3 x 15 mL). The Mtt protection of the central Lys residue was then removed by shaking the resin over 10 mL of DCM with 1% TFA and 5% TIS by vol. for 2 min. This procedure was repeated three additional times, and the resin was then washed with DCM (3 x 15 mL). *ortho*-FPBA and $NaBH_4$ were used to attach phenylboronic acid to the deprotected side-chain of the Lys residue via the reductive amination procedure described above, with 9:1 DMF:MeOH (vol:vol) as solvent. Thereafter, final Fmoc deprotection was accomplished as described above. Cleavage from the resin and deprotection of the additional Boc and OtBu side-chain protections were accomplished in a single step using the same cleavage procedure described above. The peptide was further purified by RP-HPLC. This affords the peptide product H-Asp-Glu-$(Ala)_2$-Lys(PBA)-$(Ala)_2$-Lys-Asp-OH (**3**). Here, PBA indicates phenylboronic acid or the corresponding deboronated substituent attached to the side-chain of the Lys residue. RP-HPLC purification was unable to resolve the boronated and deboronated versions of this nonapeptide and, as such, the signatures of both species are found in the NMR and mass spectra. This is shown in greater detail by Figures S1-S3. Yield (0.1 mmol

scale): 51.8 mg (49%). ^{1}H-NMR (500 MHz, in D_2O, aromatic signals indicative of boronated and deboronated components demarcated by * and †, respectively): δ 7.64* (d, *J* = 6.7, 1H), 7.36* (quint, *J* = 8.8 Hz, 2H), 7.29* (d, *J* = 7.4 Hz, 1H), 7.17† (q, *J* = 8.4 Hz, 2H), 6.81† (t, *J* = 7.4 Hz, 2H), 4.09-4.21 (m, 10H), 2.80-2.93 (m, 8H), 2.33 (t, *J* = 6.9 Hz, 2H), 1.94 (quint, *J* = 7.6 Hz, 1H), 1.83 (quint, *J* = 7.6 Hz, 1H), 1.47-1.73 (m, 8H), 1.29 (m, 4H), 1.21 (d, *J* = 7.0 Hz, 12H). ^{11}B-NMR (160 MHz, in D_2O): δ 19.1. ^{13}C-NMR (125 MHz, in D_2O, aromatic signals indicative of boronated and deboronated components demarcated by * and †, respectively): δ 174.2, 173.7, 173.4, 173.3, 172.6, 172.5, 168.6, 155.0†, 135.4*, 135.2*, 131.7*, 131.4*, 131.3*, 131.1*, 129.1†, 120.5†, 117.5†, 115.5†, 115.1†, 53.3, 53.1, 51.9, 49.4, 49.0, 46.8, 39.1, 35.4, 34.7, 30.4, 29.7, 26.2, 24.9, 21.9, 16.4. HR-MS (ESI): calculated for boronated component $C_{44}H_{70}BN_{11}O_{18}$ $[M+H-H_2O]^+$: 1034.4920; found: 1034.4970; calculated for deboronated component 1024.4952; found: 1024.4950.

Mass Spectrometry (MS)

MS was performed using a Waters Corporation (Manchester, UK) Synapt G2-HDMS™ time-of-flight (TOF), high-definition spectrometer with direct infusion in negative or positive ionization mode (electrospray ionization, ESI). Mass analysis was performed in sensitivity mode with the following TOF-MS settings: source temperature of 125°C, sampling cone at 40.0 V, extraction cone at 5.0 V, acquisition mass range of 50-2000 Da, scan time of 0.5 seconds, detector at 2000 V, and data format set to continuum. For negative and positive ionization mode, peptide samples were dissolved in deionized H_2O with added ammonium hydroxide or formic acid, respectively.

Nuclear Magnetic Resonance (NMR) Spectroscopy

Solution NMR spectroscopy was performed using a Bruker Avance-III 500 MHz spectrometer. Quartz sample tubes were used to limit background signal from boron present in standard borosilicate glass tubes. ^{1}H NMR spectra were acquired

over 32 scans using a 5 s delay time. ^{1}H chemical shifts were referenced to residual partially hydrogenated solvent as an internal standard, DMF: δ 2.75 ppm (quint), δ 2.92 ppm (quint), δ 8.03 ppm (s); MeOH: δ 3.31 ppm (quint), δ 4.78 ppm (s); H_2O δ 4.8 ppm (s). ^{11}B NMR spectra were acquired over 512-1024 scans using a 5 s delay time with the chemical shifts referenced to $BF_3{\cdot}Et_2O$ as an external standard (δ 0 ppm). ^{13}C NMR spectra were acquired over a minimum of 8000 scans using a 5 s delay time with the chemical shifts referenced to tetramethylsilane as an external standard (δ 0 ppm).

The NMR spectra of reaction mixtures were monitored over time until no further changes are apparent. The maximum time scale required for equilibrium to be reached was almost exclusively on the order of several hr and the spectra provided were collected after equilibrium had been achieved. Instances where longer times were required to reach equilibrium are noted accordingly.

Reverse Phase-High Performance Liquid Chromatography (RP-HPLC)

RP-HPLC was conducted using an Agilent 1200 series preparative-scale system equipped with two model G1361A pumps, a model G1315D diode array detector, a model G1315D fraction collector, a Rheodyne model 3725i-038 manual injector, and a Zorbax 300SB C18 column. A mobile phase gradient between H_2O with 0.05 vol. % TFA (A) and MeCN with 0.045 vol. % TFA (B) was employed, starting from 99:1 (vol:vol) A:B with a linear increase of 1 %B/min. Elution was monitored at 206 nm (10 nm bandwidth) from which a 450 nm reference (80 nm bandwidth) was subtracted.

Additional Characterization Data

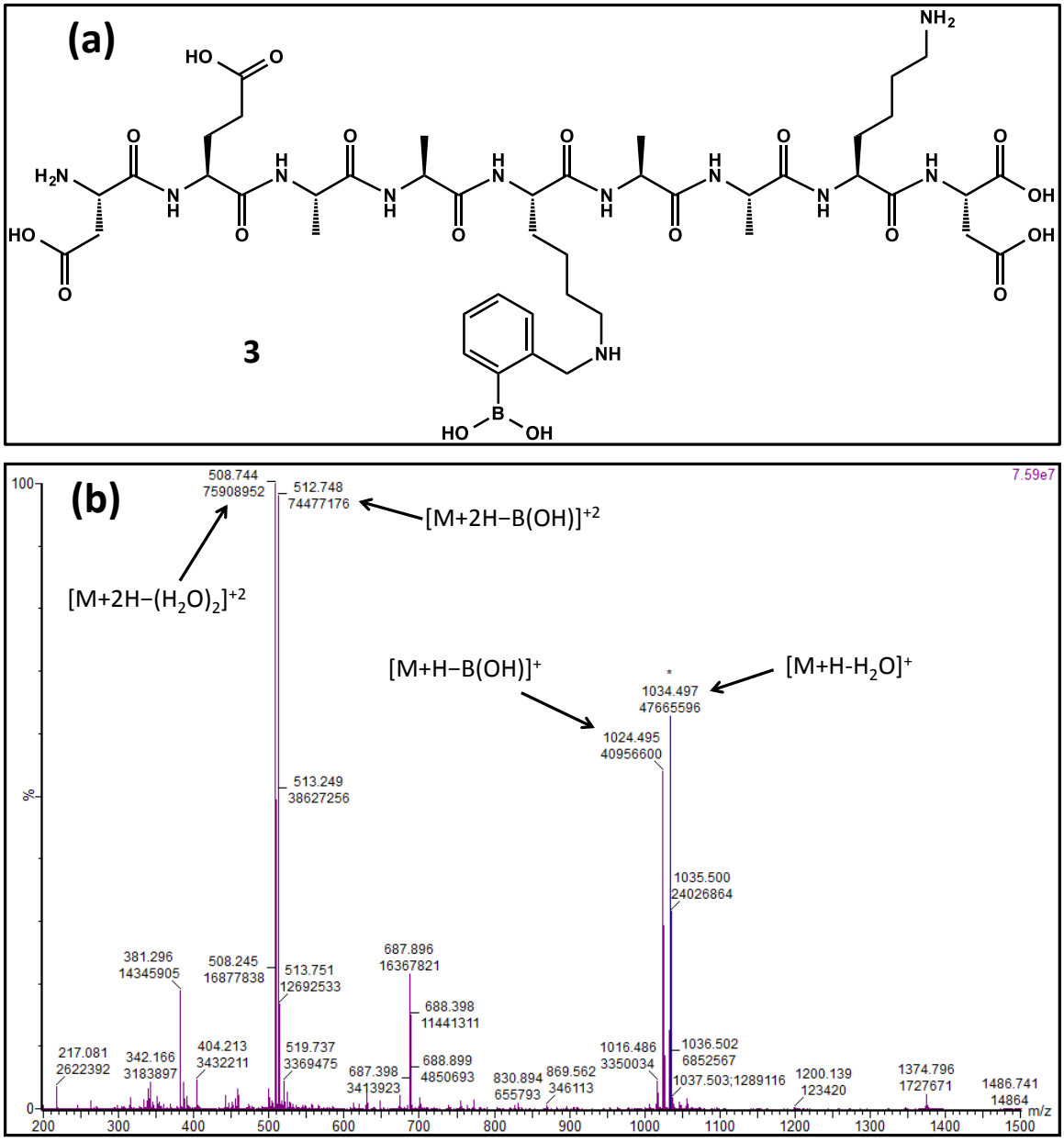


Figure S1. (a) Chemical structure of boronic acid-bearing linear nonapeptide H-Asp-Glu-(Ala)$_2$-Lys(PBA)-(Ala)$_2$-Lys-Asp-OH (compound **3**) intended by reductive amination of *ortho*-FPBA to the side chain of the central lysine residue. The mass of the intended product is M = 1051.5. **(b)** ESI-MS positive mode spectrum of nonapeptide product. The peaks at m/z = 1034.5 and m/z = 508.7 are consistent with dehydrated forms of the intended product, *i.e.*, successful incorporation of the boronic acid functionality into the peptide. However, the peaks at m/z = 1024.5 and m/z = 512.7 indicate substantial deboronation of the product, similar to the dipeptide example described in the main text.

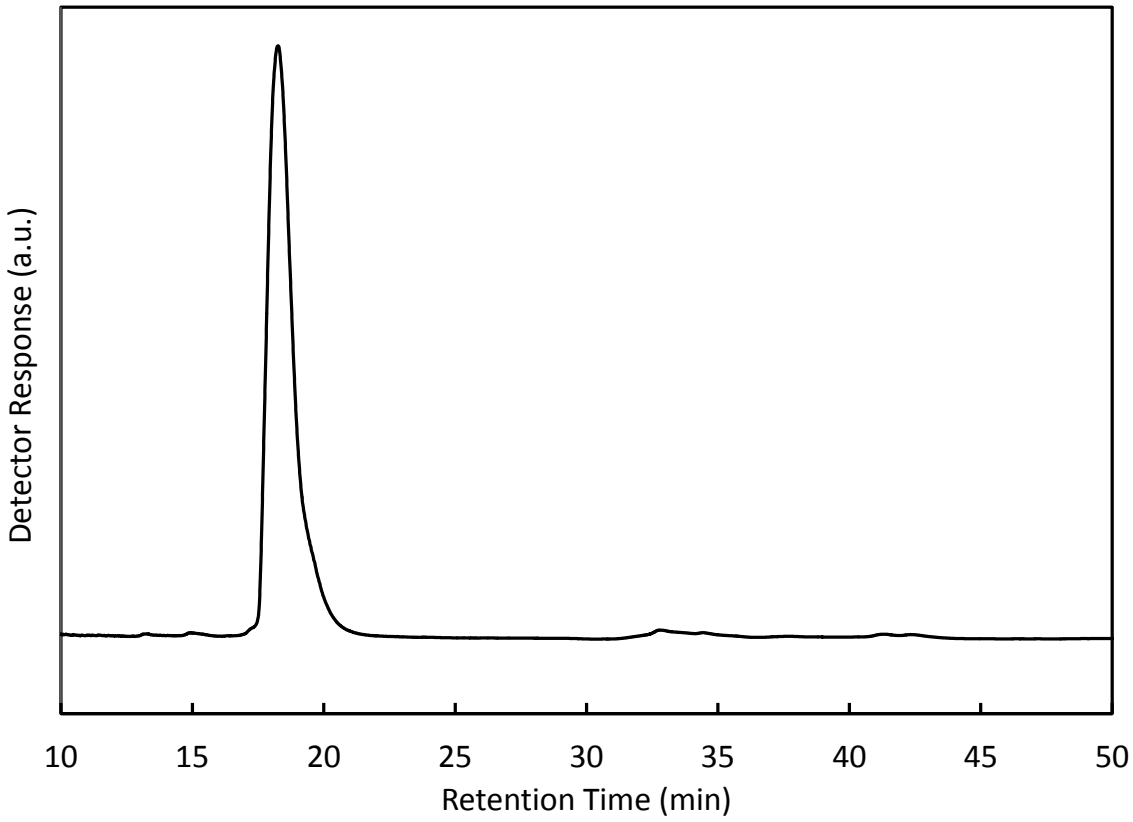
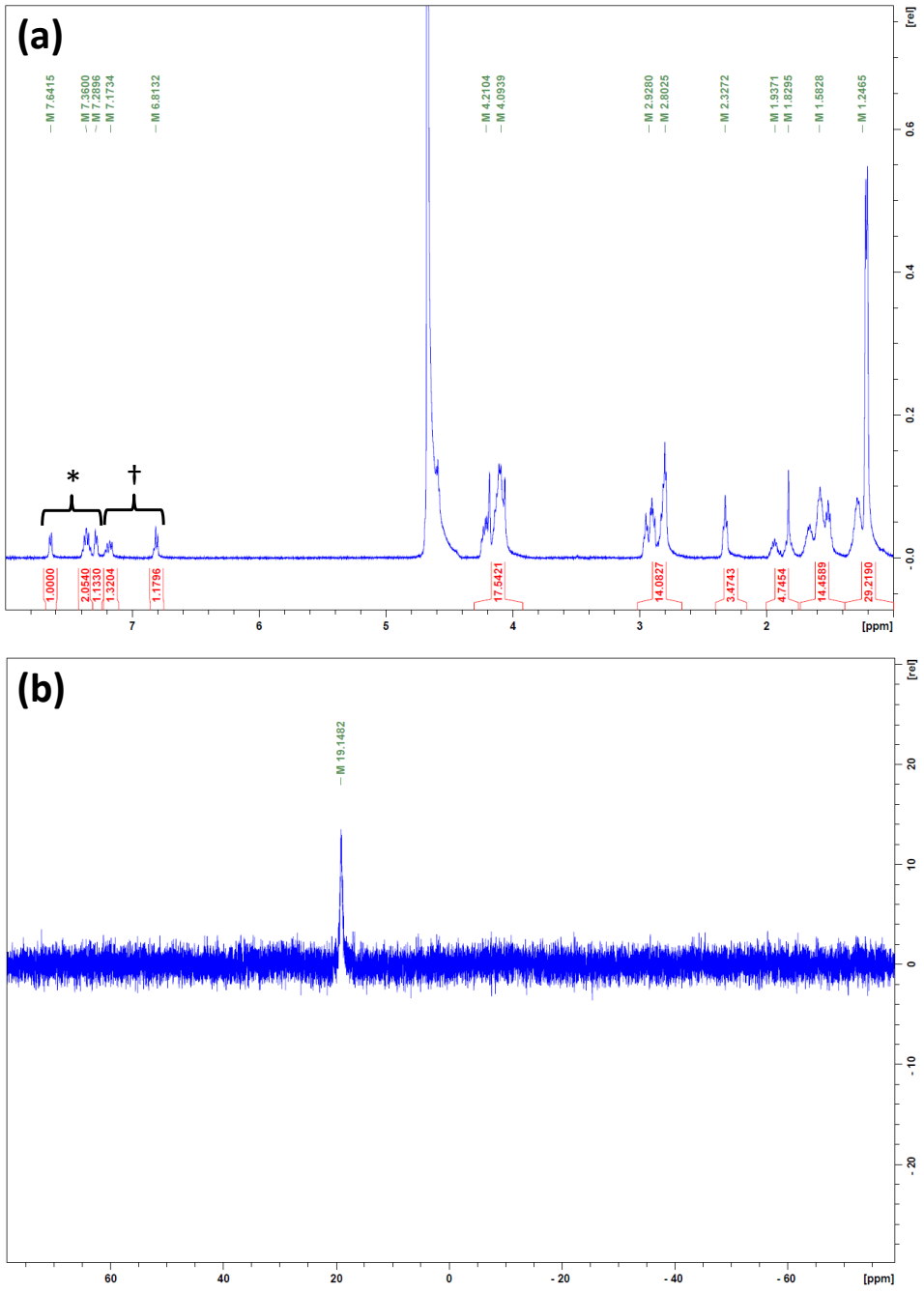


Figure S2. RP-HPLC chromatograph of compound **3**. The chromatograph exhibits a single peak, suggestive of a highly pure, single species. However, the corresponding mass spectrum (Figure S1) and NMR spectra (Figure S3) clearly indicate the presence of both the boronated and deboronated versions of **3**. Thus, the RP-HPLC was unable to effectively resolve these two distinct species.


(a)
M 7.6415
M 7.3600
M 7.2896
M 7.1734
M 6.8132
M 4.2104
M 4.0939
M 2.9280
M 2.8025
M 2.3272
M 1.9371
M 1.8295
M 1.5828
M 1.2465
*
†
1.0000
2.0540
1.1330
1.3204
1.1796
17.5421
14.0827
3.4743
4.7454
14.4589
29.2190
[ppm]
[rel]
(b)
M 19.1482
[ppm]
[rel]

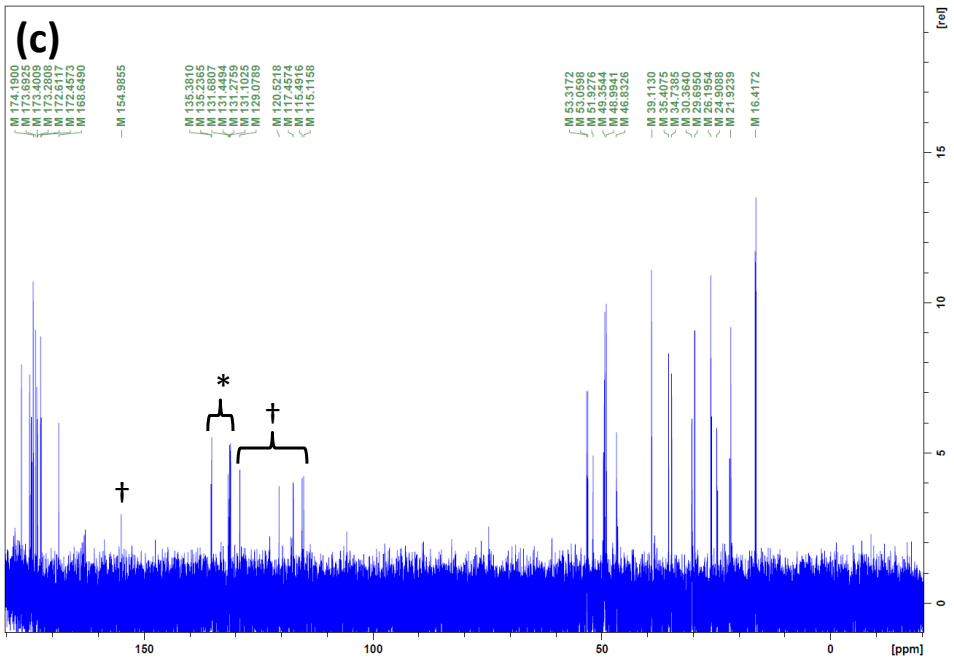


Figure S3. (a) ^{1}H, **(b)** ^{11}B, and **(c)** ^{13}C NMR spectra of compound **3** in D_2O. The aromatic ^{1}H and ^{13}C resonances corresponding to boronated and deboronated species are demarcated by * and †, respectively.

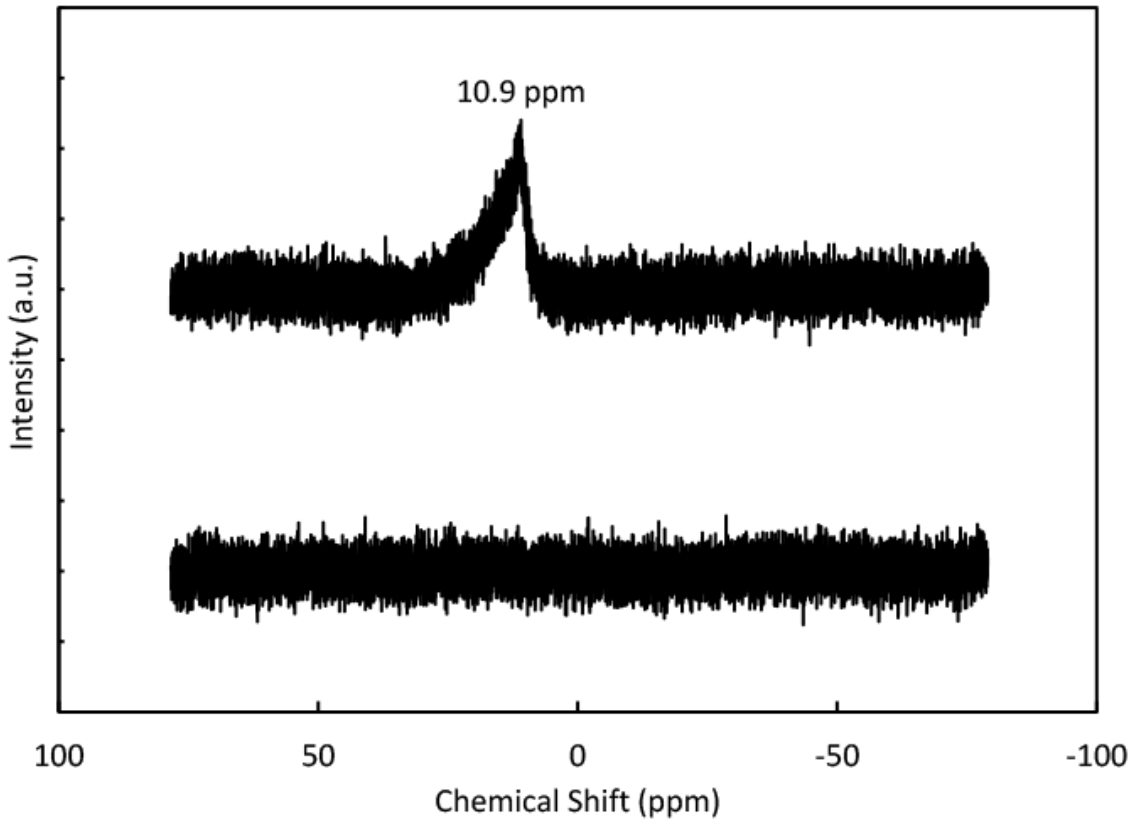


Figure S4. ^{11}B NMR spectra of dipeptide of phenylalanine (Phe-Phe) *N*-terminally modified by reductive amination of **(top)** *para*-FPBA (**1**) and **(bottom)** *ortho*-FPBA (**2**). The solvent is pH 10 D_2O. The lack of a ^{11}B signal above noise only in the case of *ortho*-FPBA supports the deboronation proposed in Scheme 1 of the main text.

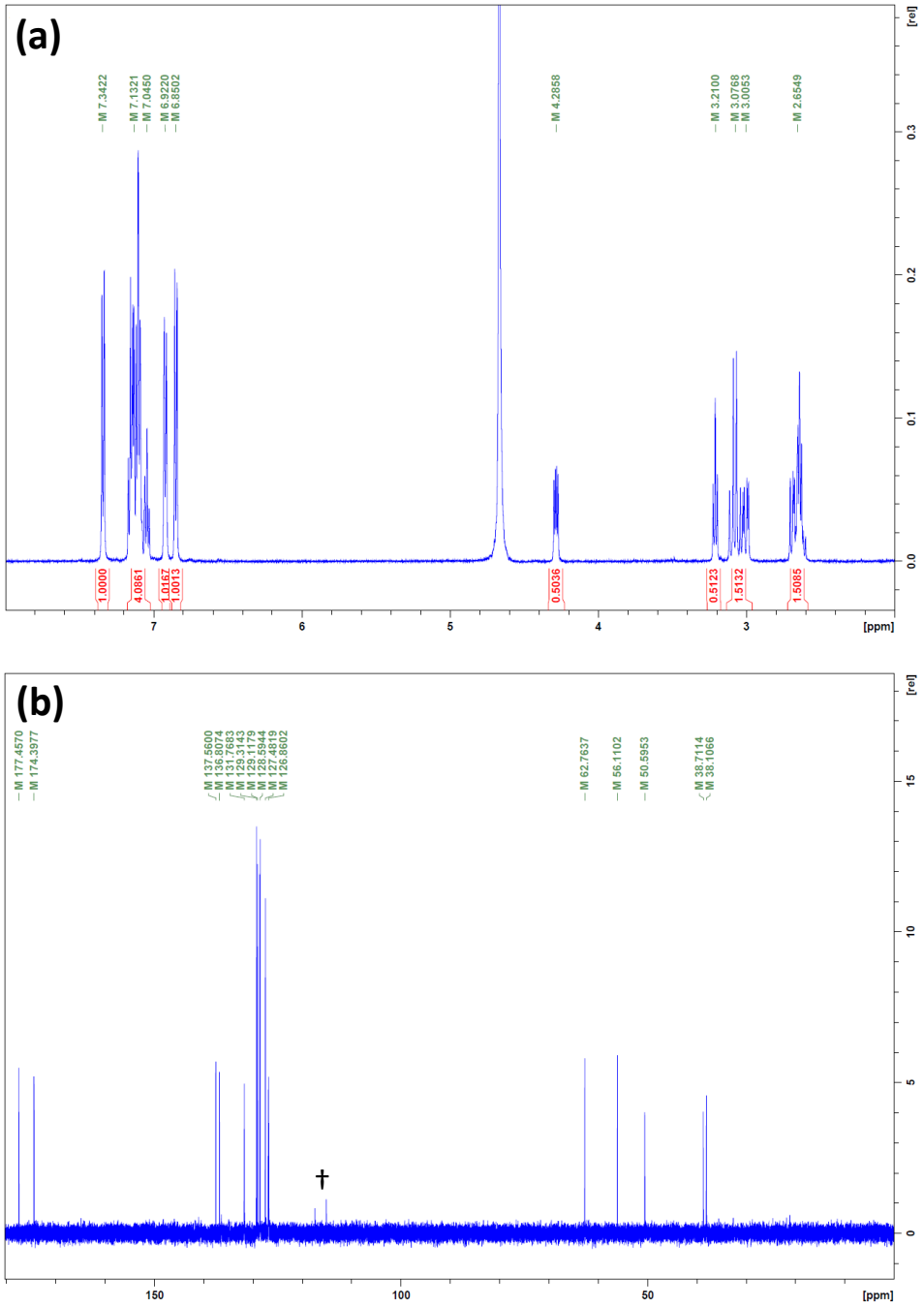


Figure S5. (a) ^{1}H and **(b)** ^{13}C NMR spectra of compound **1** in D_2O with 0.25 vol. % ND_4OD. A slight deboronated impurity, demarcated by †, is evident in the ^{13}C spectrum.

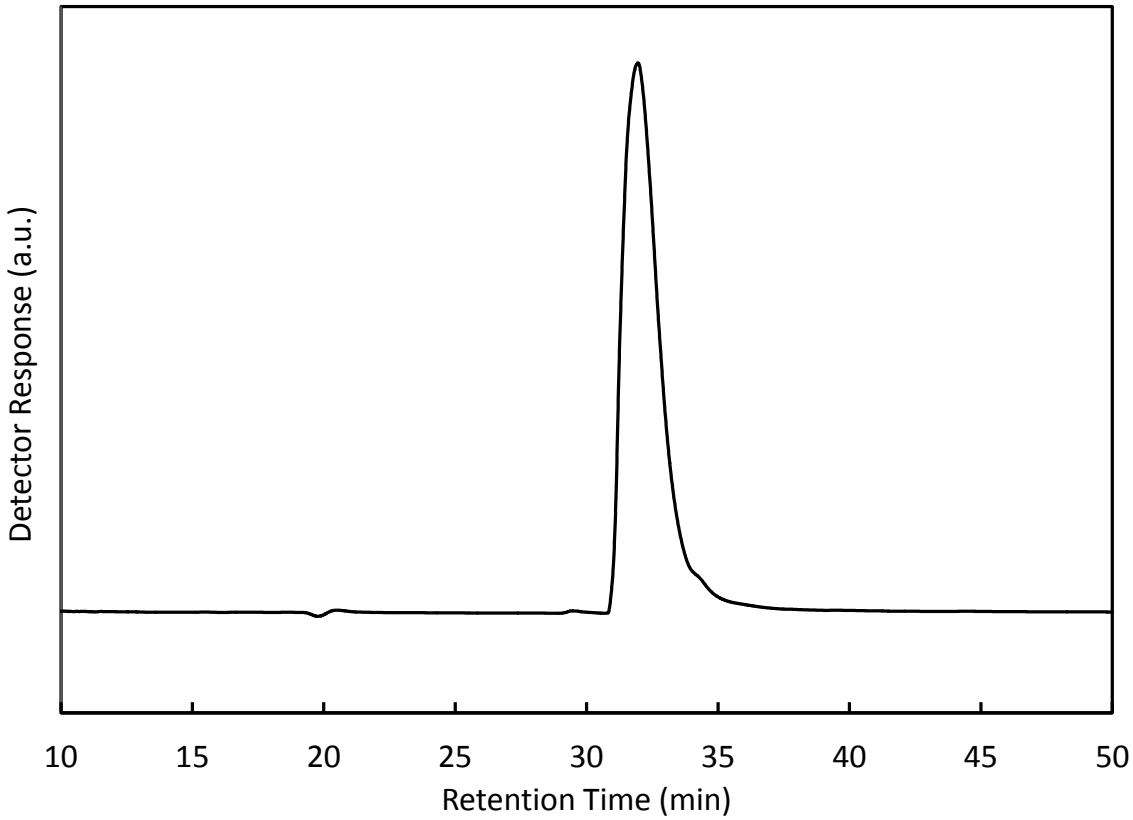


Figure S6. RP-HPLC chromatograph of compound **1**.

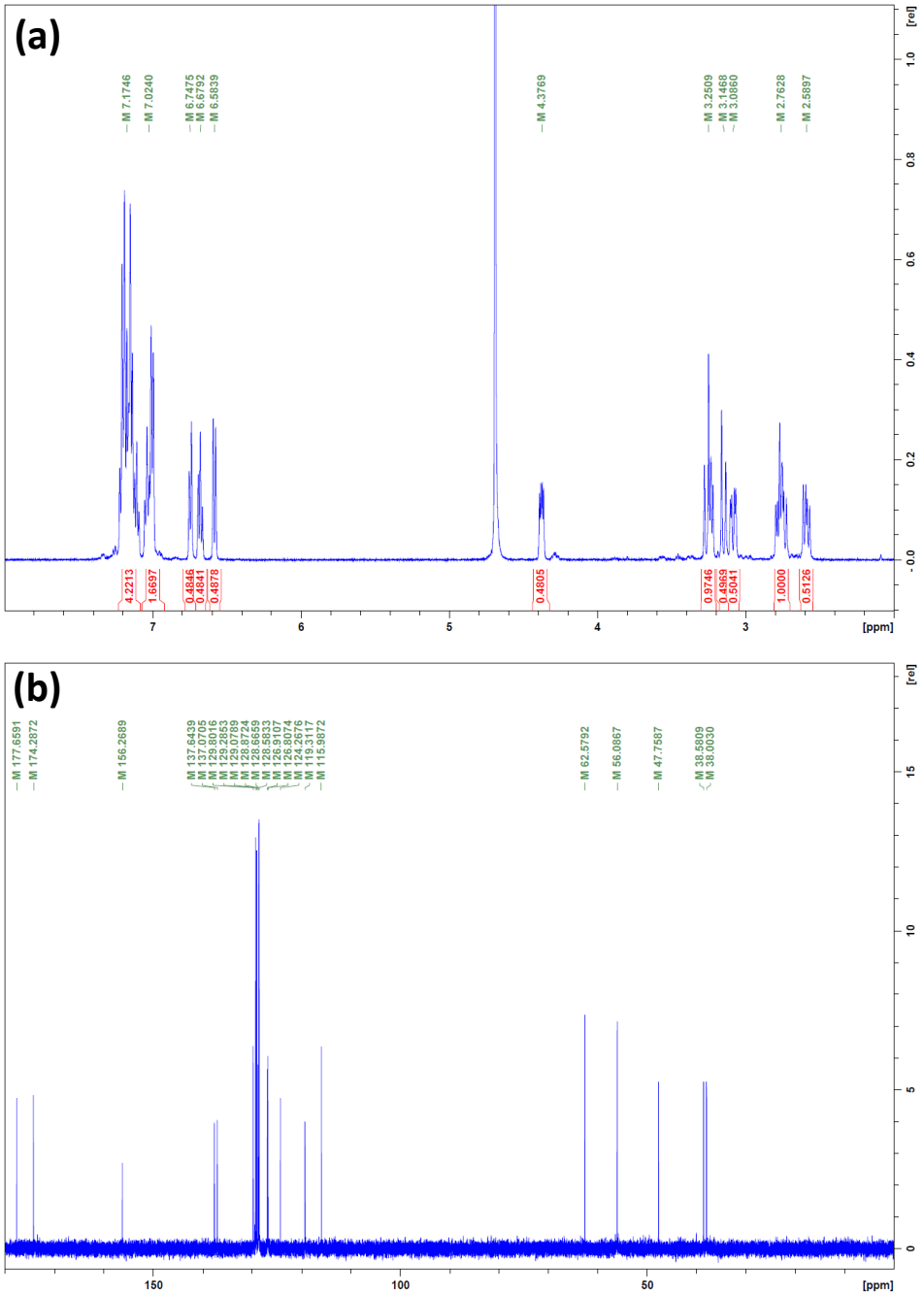


Figure S7. (a) ^{1}H and **(b)** ^{13}C NMR spectra of compound **2** in D_2O with 0.25 vol. % ND_4OD. Note the upfield shift of the aromatic protons in **(a)**, as well as the upfield and downfield shifts of the aromatic carbons in **(b)** relative to the boronated product **1** in Figure S5, further confirmation of the deboronated nature of **2**. In particular, the distinct resonance at 156 ppm in **(b)** is consistent with a phenolic product, arising specifically from the carbon adjacent to the hydroxyl substituent.[2]

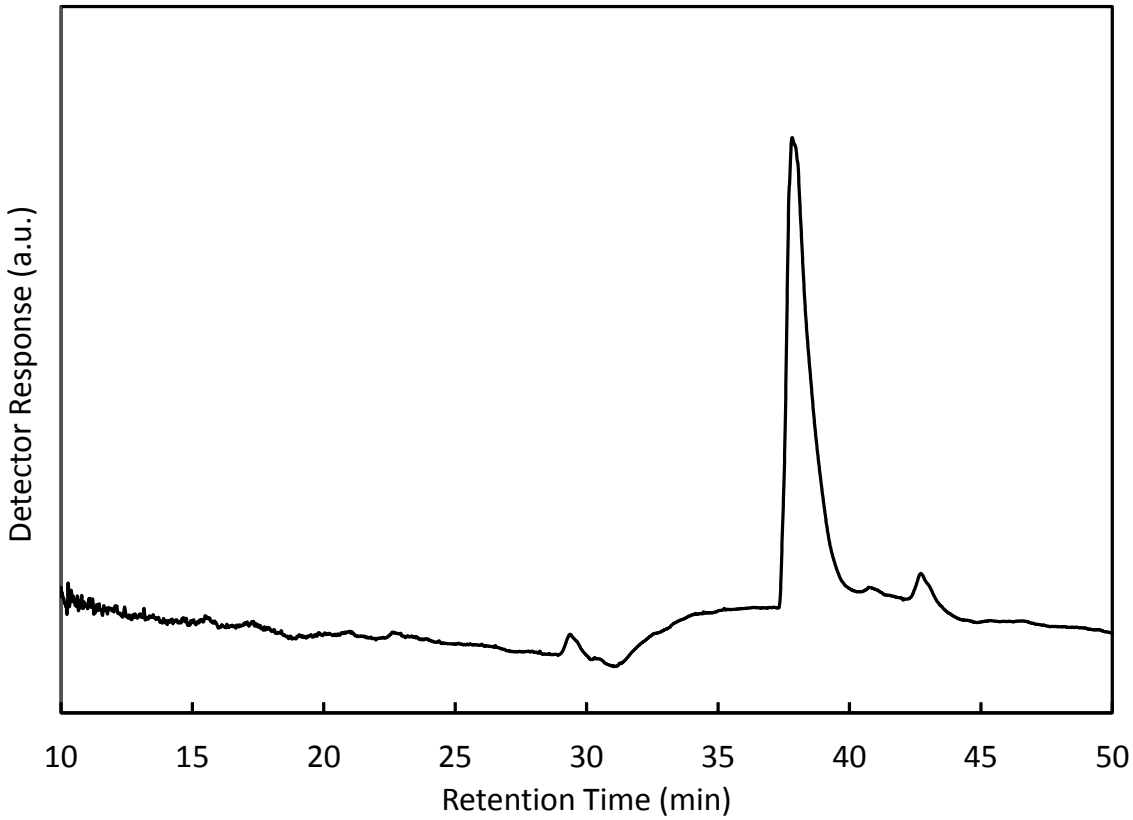


Figure S8. RP-HPLC chromatograph of compound **2**.

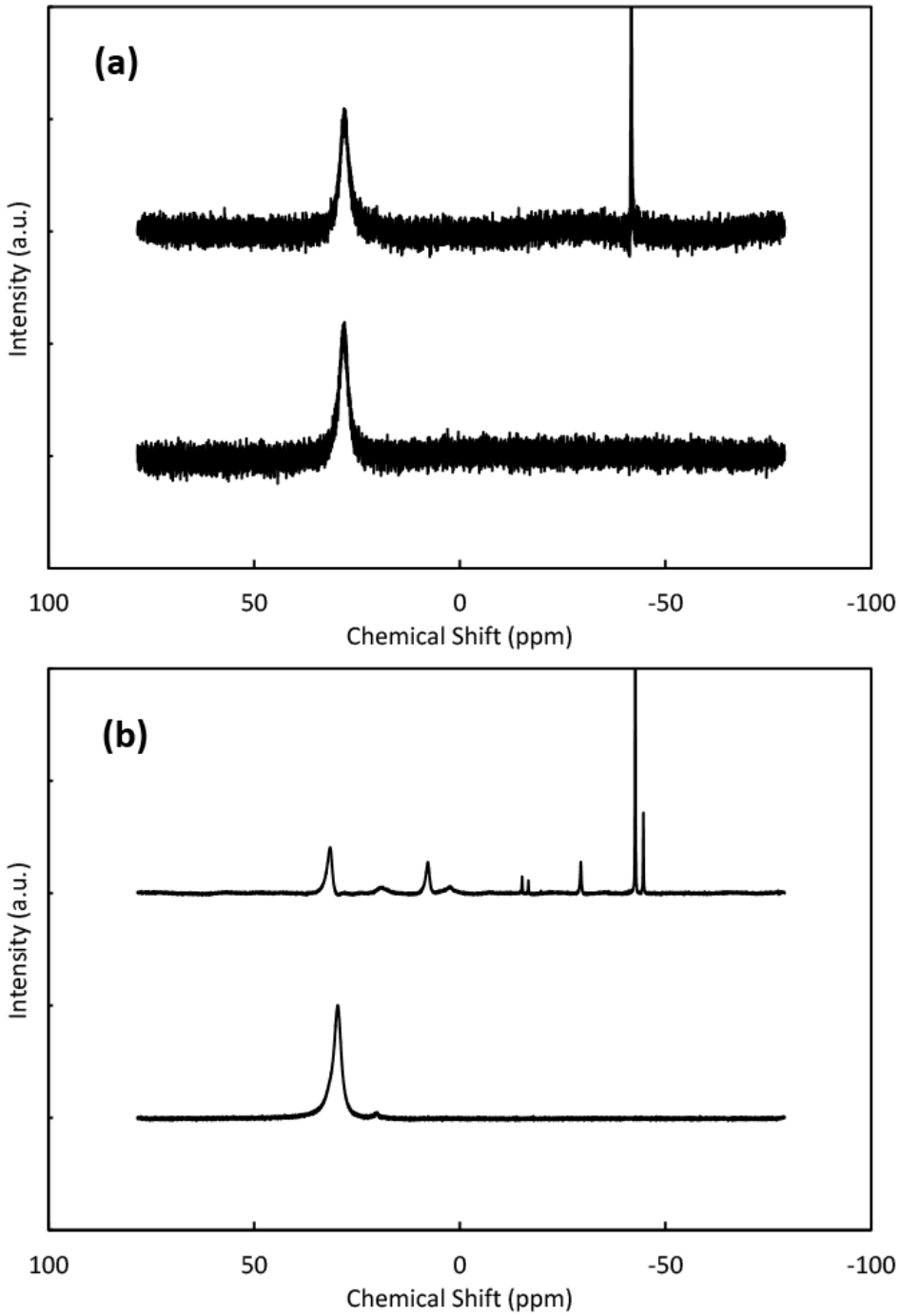


Figure S9. ^{11}B NMR spectra showing isomer-dependent sensitivity of **(a)** *para*-FPBA and **(b)** *ortho*-FPBA to reducing agent $NaCNBH_3$. In both panels, the bottom spectrum shows the neat FPBA in d_7-DMF, while the top spectrum shows the FPBA in d_7-DMF after addition of $NaCNBH_3$. The spectra clearly indicate that the boronic acid functionality of FPBA (δ 29.9 ppm) is stable to $NaCNBH_3$ (δ −41.5 ppm) in DMF only for the *para* isomer. Furthermore, the new resonances in the top spectrum of **(b)** at 31.6 ppm and 7.7 ppm are consistent with reduction of *ortho*-FPBA to the corresponding benzoxaborole **4**.[3]

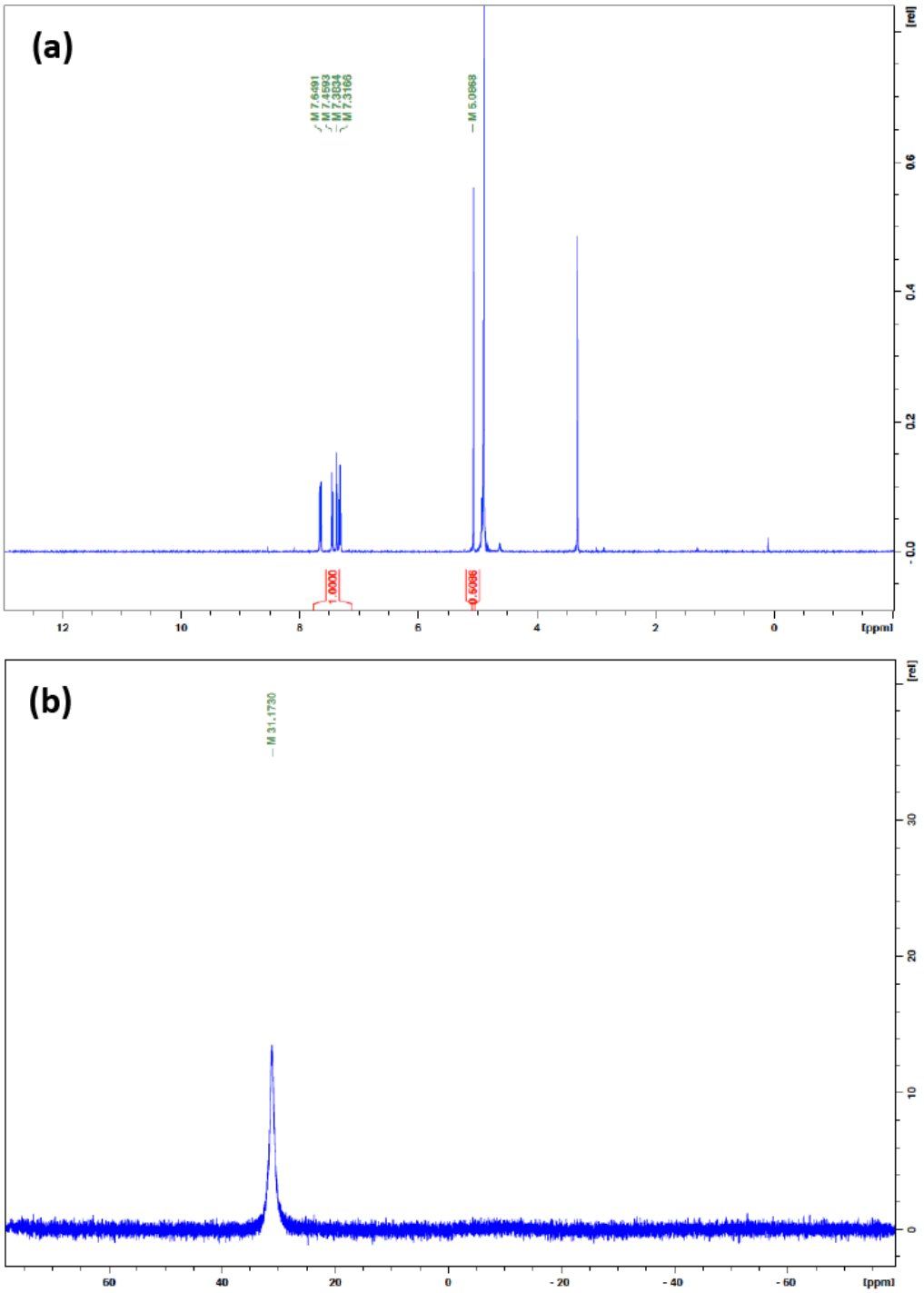


Figure S10. (a) ^{1}H and **(b)** ^{11}B NMR spectra of a commercially available sample of the benzoxaborole **4** proposed by reduction of *ortho*-FPBA – the hemiester of 2-(hydroxymethyl)phenylboronic acid. The solvent is CD_3OD. In **(a)**, the proton signal at 5.1 ppm arises from the aliphatic protons in the oxaborole ring, and is consistent with the proton signal of the reduced *ortho*-FPBA highlighted in Figure 2b of the main text. In **(b)**, the ^{11}B signal at 31.2 ppm is consistent with a resonance of the reduced *ortho*-FPBA found at an identical chemical shift (Figure S9b).

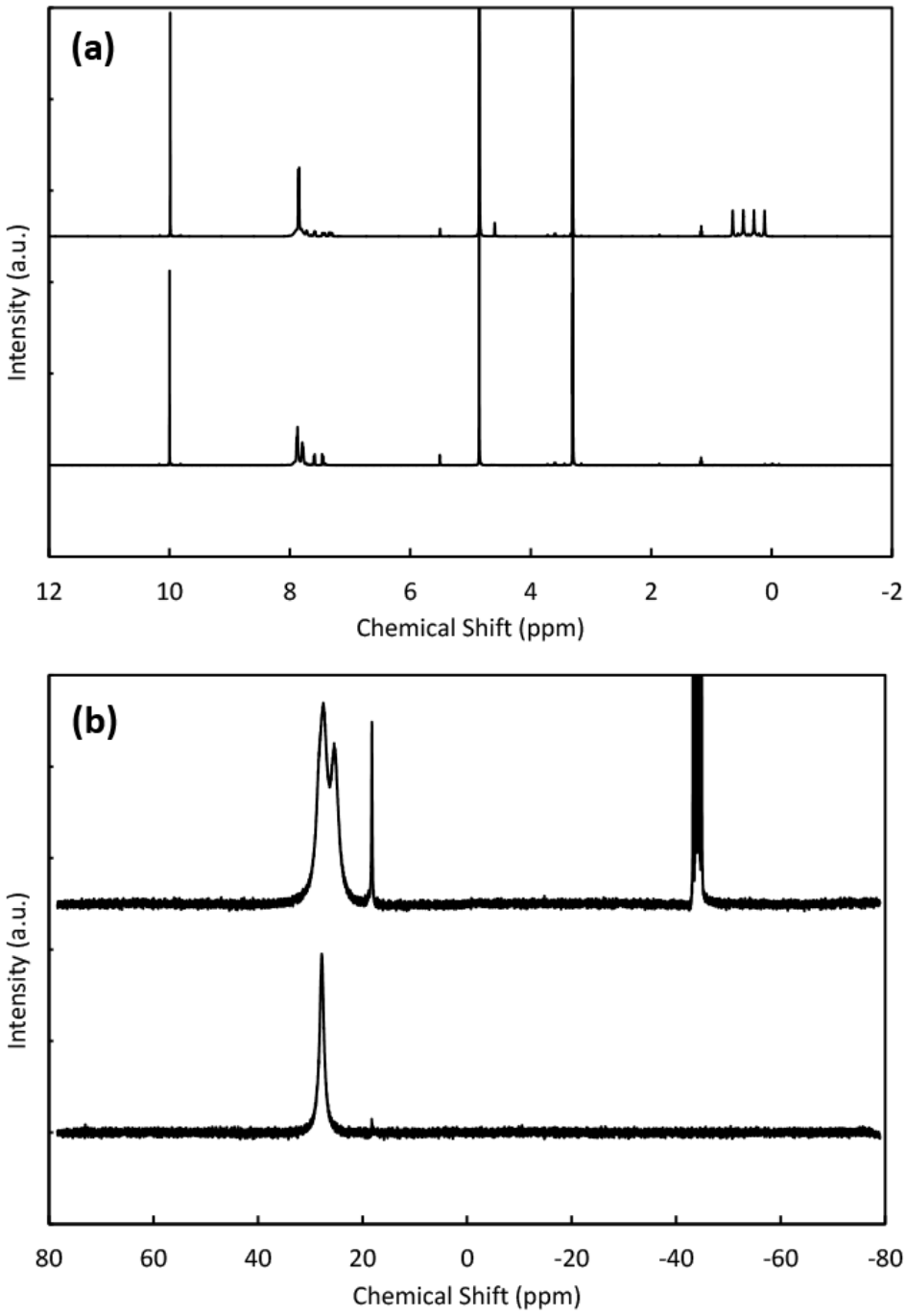


Figure S11. (a) ^{1}H and **(b)** ^{11}B NMR spectra of *para*-FPBA and $NaCNBH_3$ in CD_3OD. In both panels, the bottom spectrum shows the neat *para*-FPBA in CD_3OD, while the top spectrum shows the *para*-FPBA in CD_3OD after addition of $NaCNBH_3$. In comparison to Figure S12, the relative stability of *para*-FPBA to $NaCNBH_3$ in CD_3OD is apparent. However, the weak proton resonance at 4.6 ppm, as well as the new ^{11}B resonance at 25.3 ppm that appear after addition of $NaCNBH_3$, suggest a slow reduction of *para*-FPBA to *para*-(hydroxymethyl)phenylboronic acid (see Figure S13 for further detail).

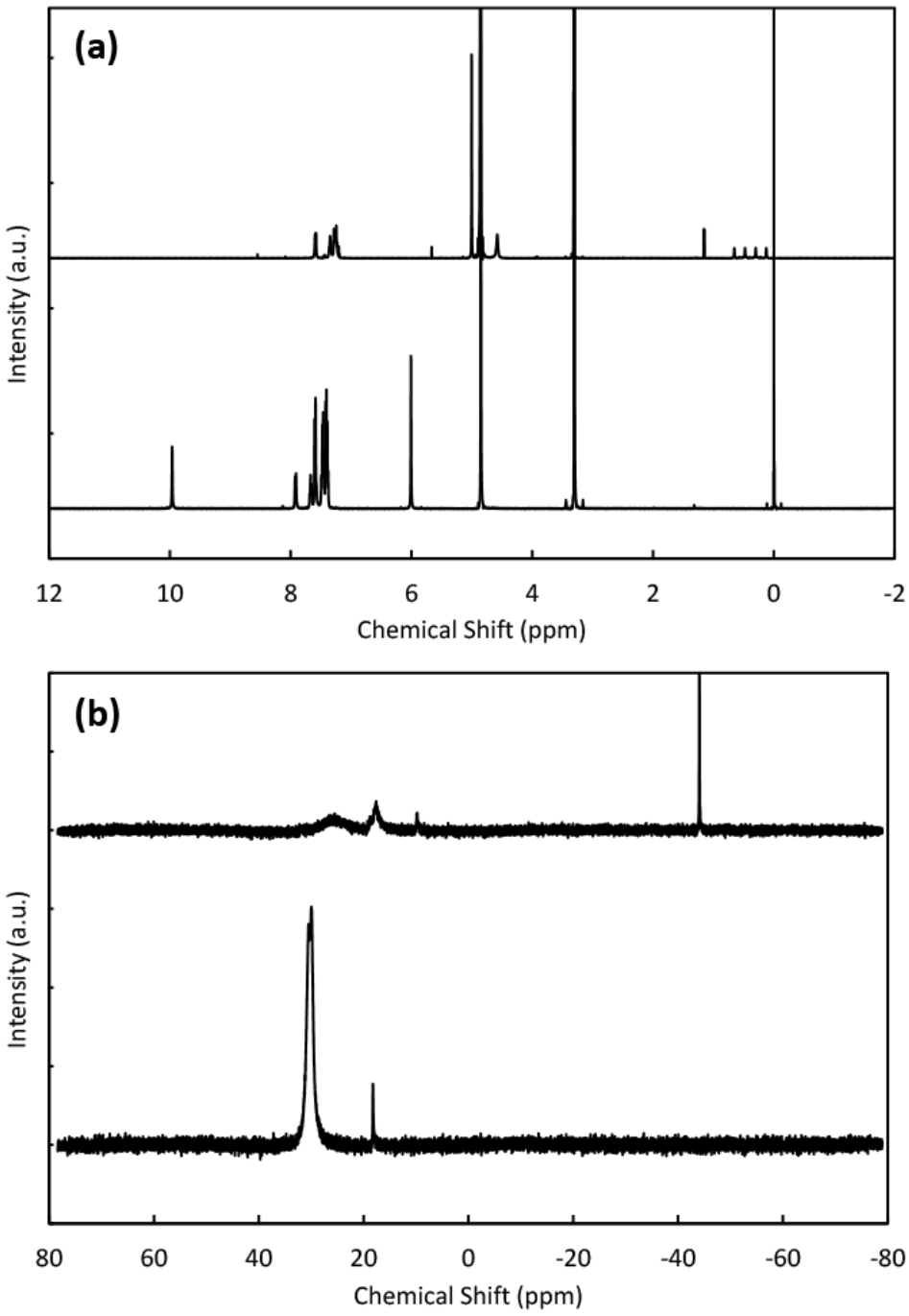


Figure S12. (a) ^{1}H and **(b)** ^{11}B NMR spectra of *ortho*-FPBA and $NaCNBH_3$ in CD_3OD. In both panels, the bottom spectrum shows the neat *ortho*-FPBA in CD_3OD, while the top spectrum shows the *ortho*-FPBA in CD_3OD after addition of $NaCNBH_3$. In comparison to Figure S11, the relative instability of *ortho*-FPBA to $NaCNBH_3$ in CD_3OD is apparent. Specifically, the disappearance of the aldehyde proton signal at 10.0 ppm and the appearance of a new proton signal at 5.0 ppm upon addition of $NaCNBH_3$ indicates complete reduction of *ortho*-FPBA to the corresponding benzaoxaborole **4**.

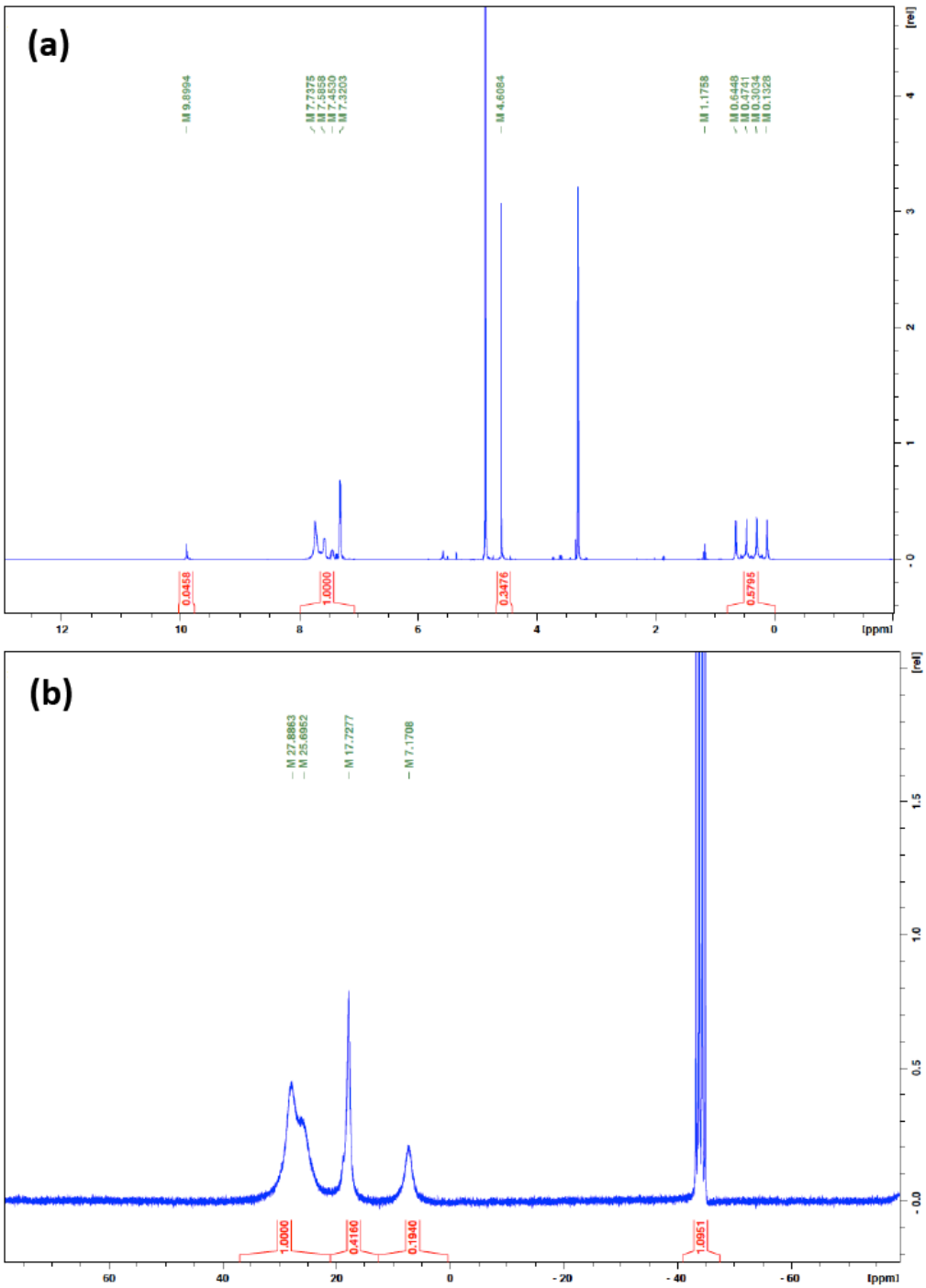


Figure S13. (a) ^{1}H and **(b)** ^{11}B NMR spectra of *para*-FPBA and $NaCNBH_3$ in CD_3OD after incubation for 3 days. These data suggest that *para*-FPBA is also reduced to *para*-(hydroxymethyl)phenylboronic acid by $NaCNBH_3$ in CD_3OD, albeit at a much slower rate than the corresponding reduction of *ortho*-FPBA. Specifically, the aldehyde proton signal at 9.9 ppm has almost completely disappeared, and has been replaced by a strong proton signal at 4.6 ppm, consistent with introduction of a hydroxymethyl group. Indeed, the ^{1}H spectrum agrees with a previously reported spectrum of *para*-(hydroxymethyl)phenylboronic acid.[4]

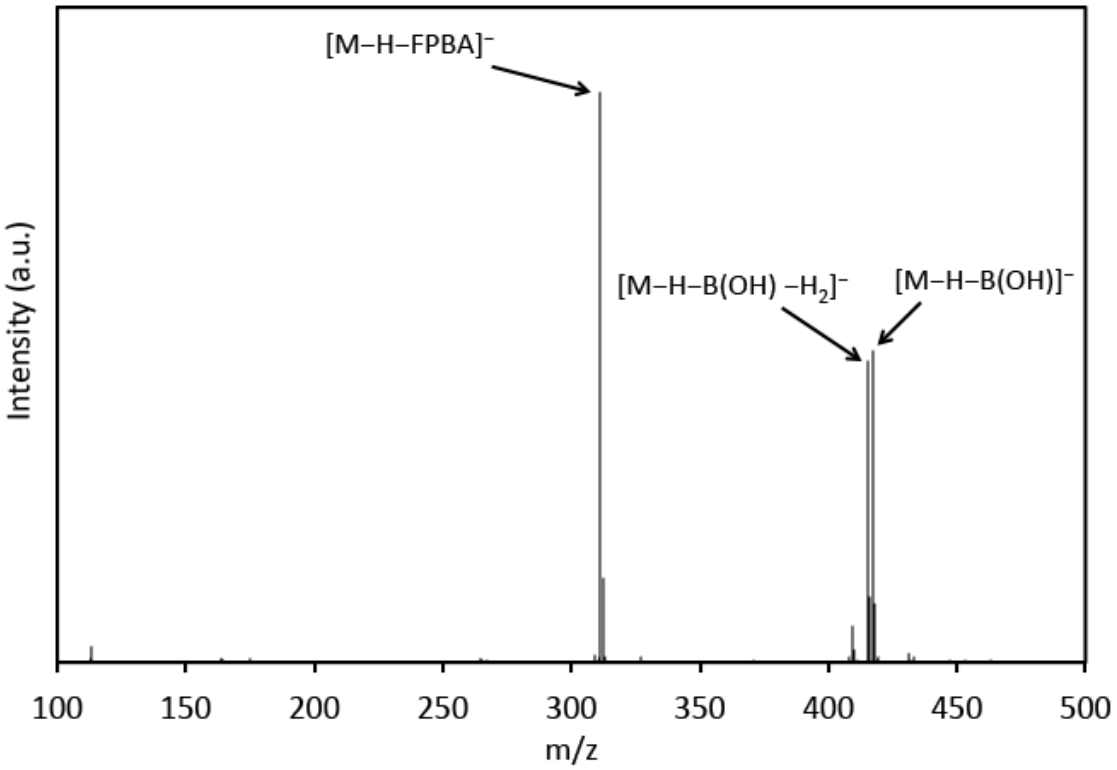


Figure S14. Negative mode ESI-MS of dipeptide of phenylalanine, Phe-Phe, *N*-terminally modified by two-step reductive amination of *ortho*-FPBA. The two-step process involves exposure of the dipeptide to *ortho*-FPBA to promote imine formation at the *N*-terminus, followed by exposure to $NaCNBH_3$ to promote reduction of the imine. M indicates 446.2 Da, the mass of the desired, BA-modified dipeptide. The spectrum shows that, as with a single-step reductive amination (Figure 1 of the main text), the product is devoid of boron. The primary species observed are: i) the dipeptide with nothing attached to the *N*-terminus ($[M-H-FPBA]^-$); ii) the dipeptide bearing a deboronated imine species at the *N*-terminus ($[M-H-B(OH)-H_2]^-$); and iii) the dipeptide bearing a deboronated secondary amine species at the *N*-terminus ($[M-H-B(OH)]^-$, *i.e.*, compound **2**.

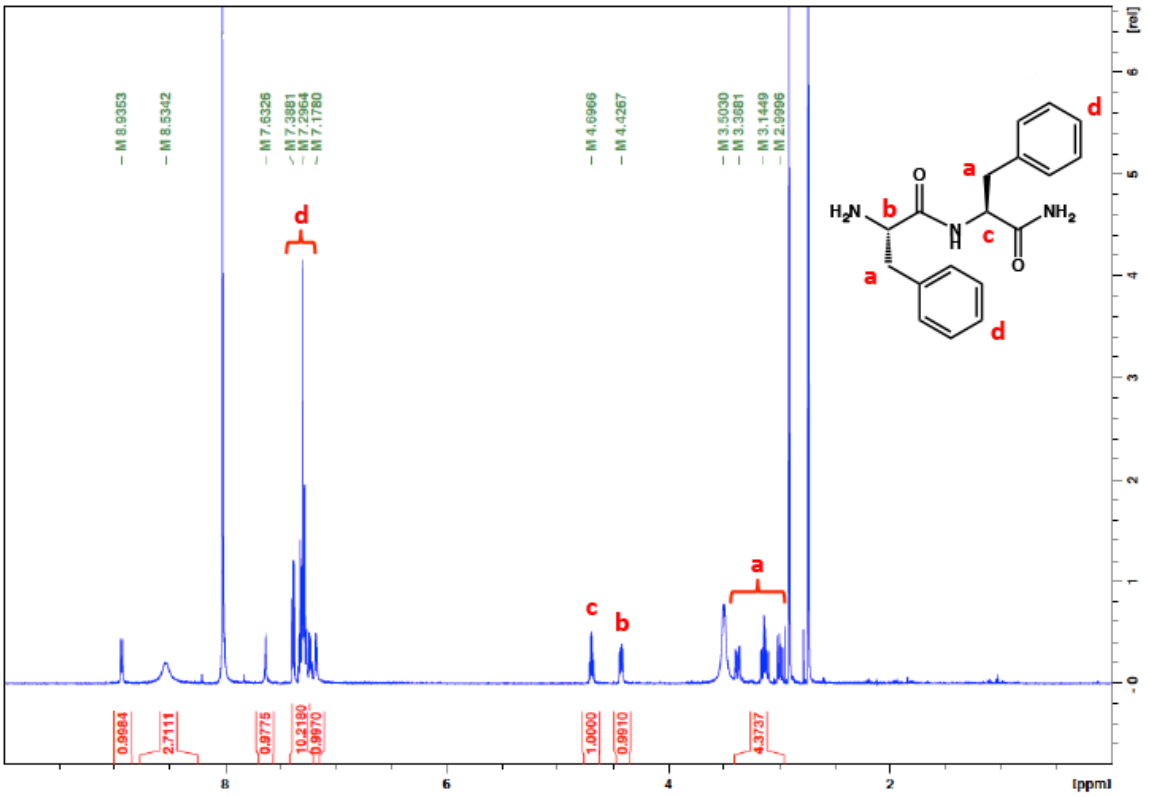


Figure S15. ^{1}H NMR spectra of H-Phe-Phe-NH_2 in d_7-DMF. Note the absence of any aromatic resonance in the range of 6-7 ppm, in contrast with the deboronated species **5** shown in Figure 3a of the main text.

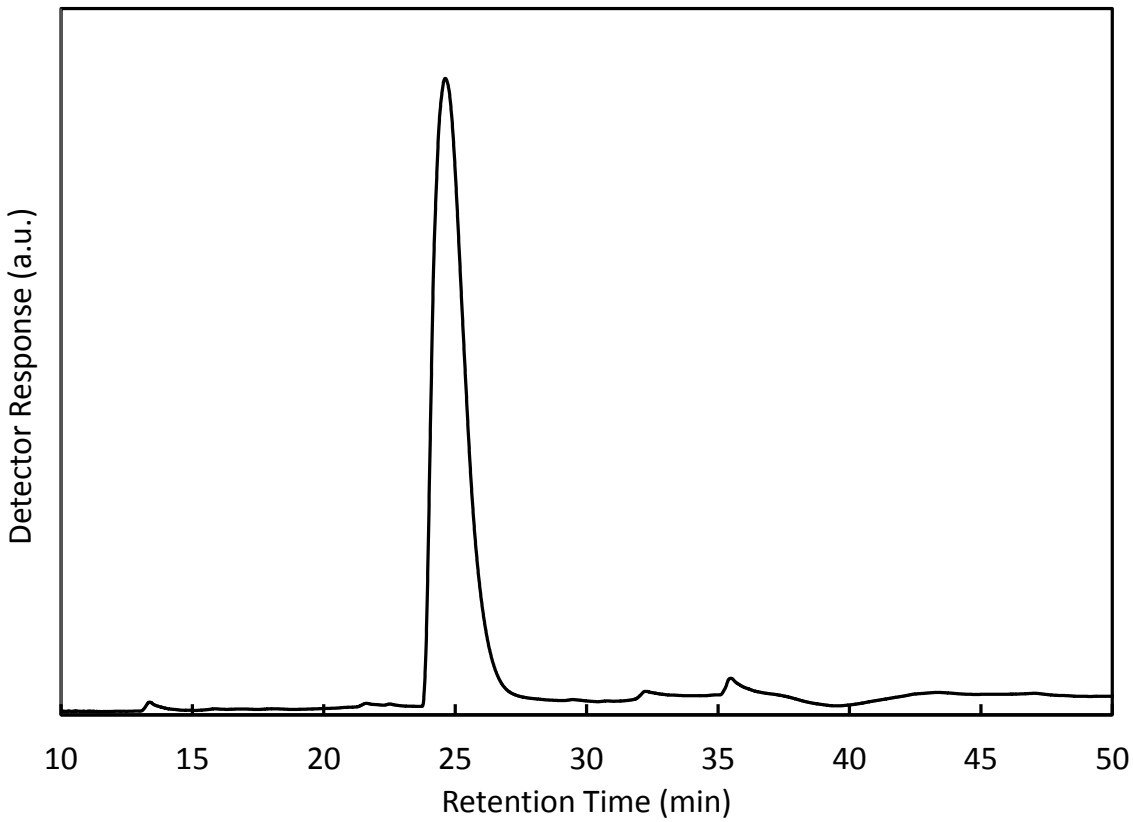


Figure S16. RP-HPLC chromatograph of H-Phe-Phe-NH_2.

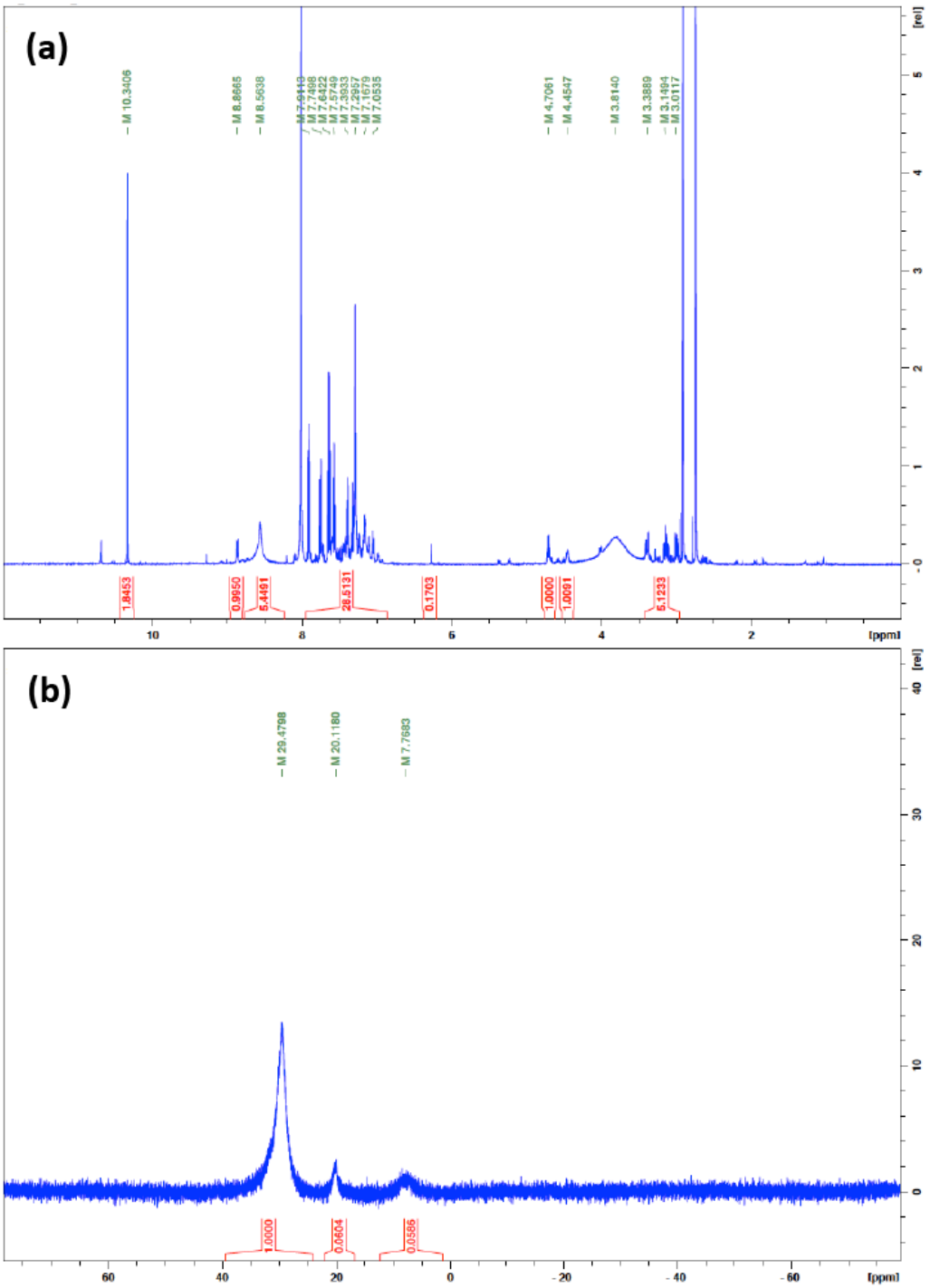


Figure S17. (a) ^{1}H and **(b)** ^{11}B NMR spectra of a mixture of *ortho*-FPBA and H-Phe-Phe-NH_2 in d_7-DMF. In **(a)**, note the absence of a significant aromatic resonance in the range of 6-7 ppm, in contrast with the deboronated species **5** shown in Figure 3a of the main text. Likewise, in **(b)** note the negligible production of boric acid (δ 20.1 ppm), in contrast with the same mixture after introduction of $NaCNBH_3$ (Figure 3b of the main text). Collectively, these observations indicate that deboronation occurs primarily upon introduction of $NaCNBH_3$.

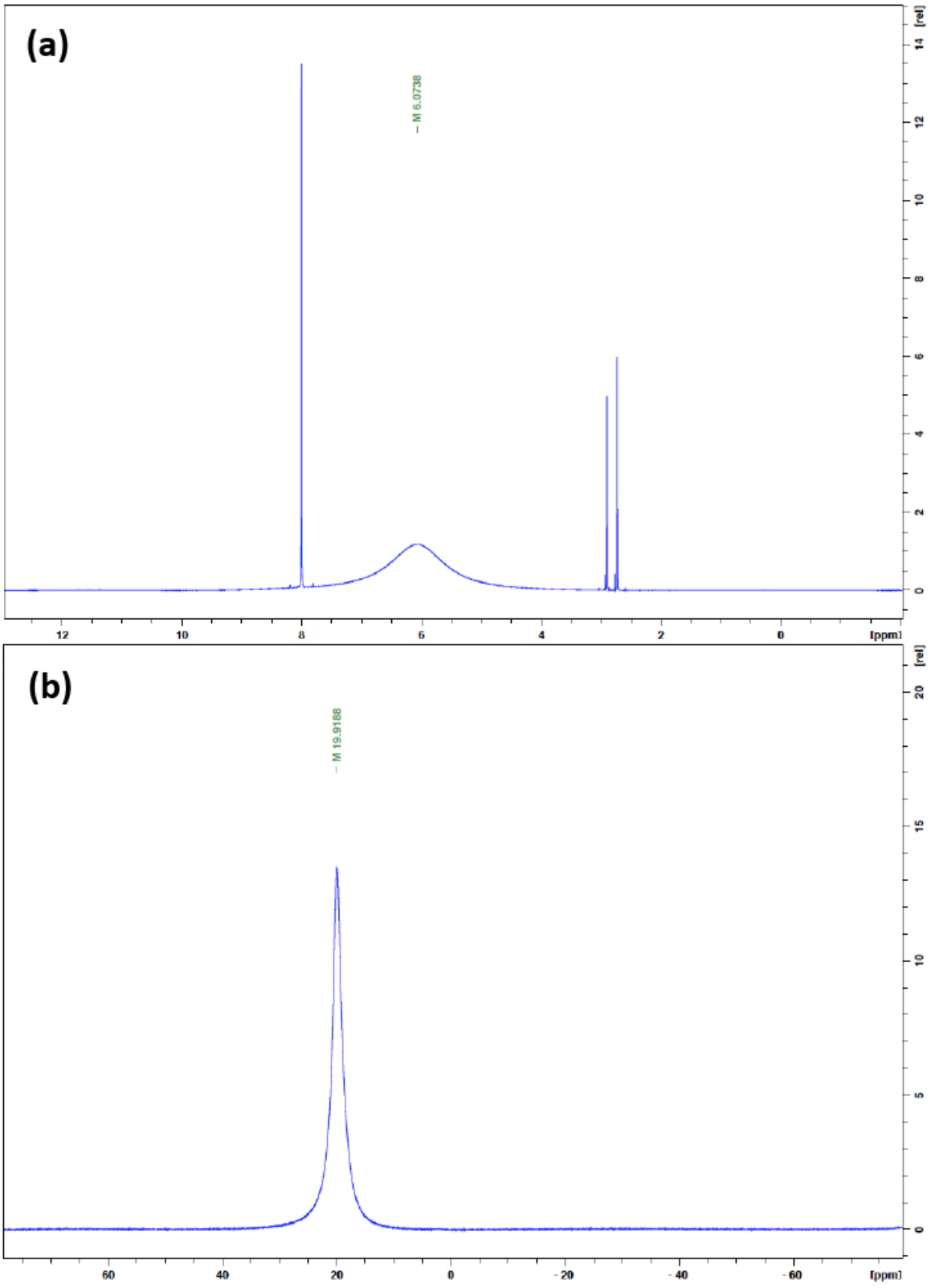


Figure S18. (a) ^{1}H and **(b)** ^{11}B NMR spectra of boric acid in d_7-DMF.

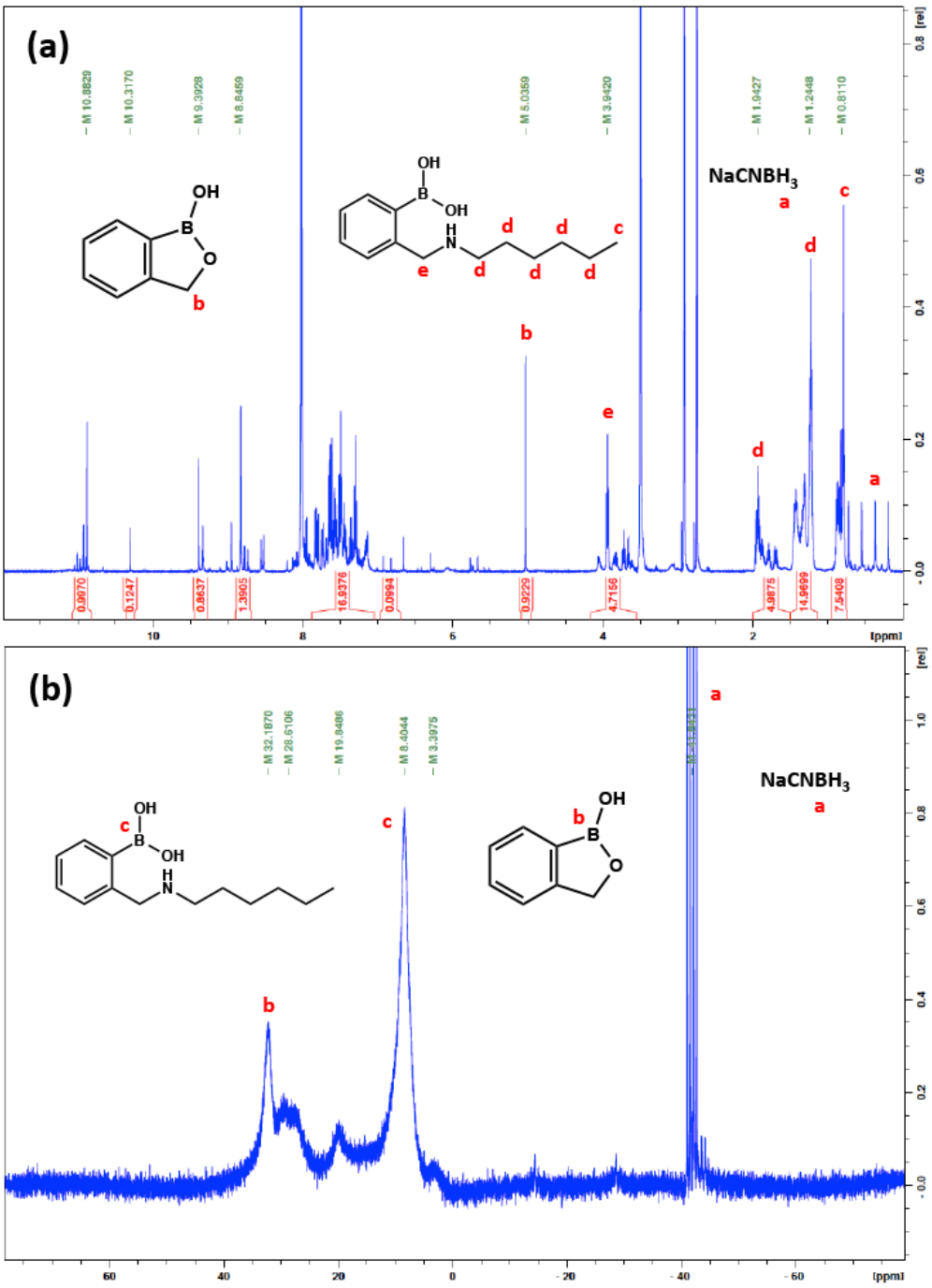


Figure S19. (a) ^{1}H and **(b)** ^{11}B NMR spectra of reductive amination of *ortho*-FPBA to hexylamine with $NaCNBH_3$ as reducing agent. The solvent is d_7-DMF. In **(a)**, note the absence of a significant aromatic resonance in the range of 6-7 ppm, in contrast with the deboronated species shown in Figure 3a of the main text. Likewise, in **(b)** note the negligible production of boric acid (δ 19.9 ppm), in contrast with the deboronated species shown in Figure 3b of the main text. Similarly, note the large resonance at 8.4 ppm, which is consistent with typical chemical shifts of aminomethylphenylboronic acids, indicating the absence of deboronation in this particular reductive amination.[5]

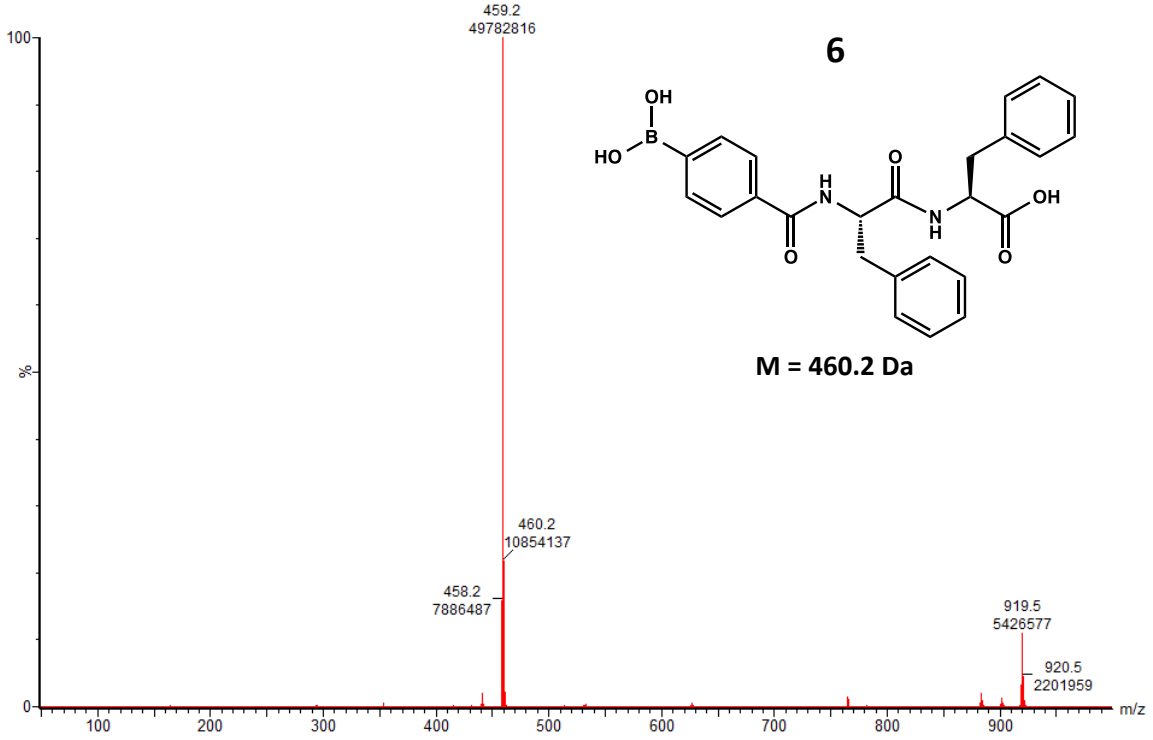


Figure S20. Negative mode ESI-MS of Phe-Phe *N*-terminally modified by coupling of *para*-CPBA (compound **6**). The principal peak at *m/z* = 459.2 is consistent with a singly charged ion of the desired product. Moreover, it exhibits the isotopic signature of a boron-containing compound; namely, the peak at *m/z* = 458.2 arises from the natural presence of ^{10}B. The peak at *m/z* = 919.5 is attributed to a dimerized species of **6**. Thus, in contrast to reductive amination of FPBA, *N*-terminal coupling of CPBA yields an exceptionally clean product with no apparent deboronation.

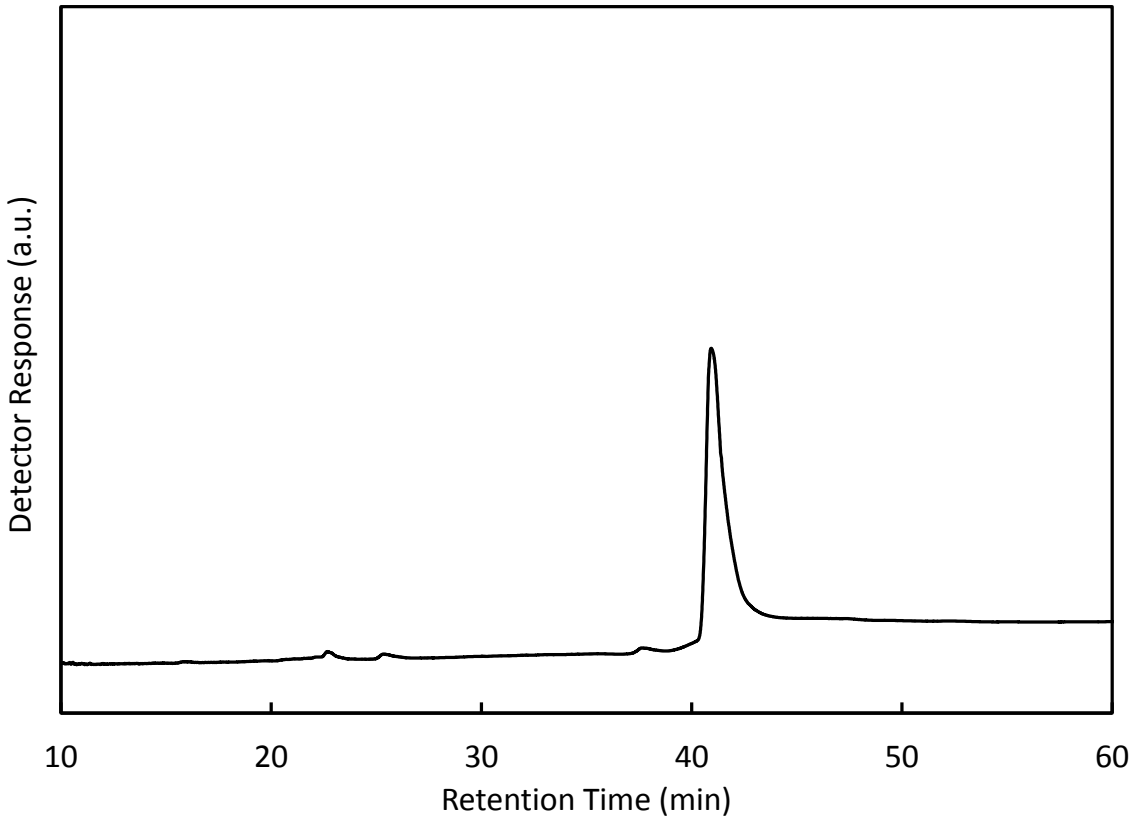
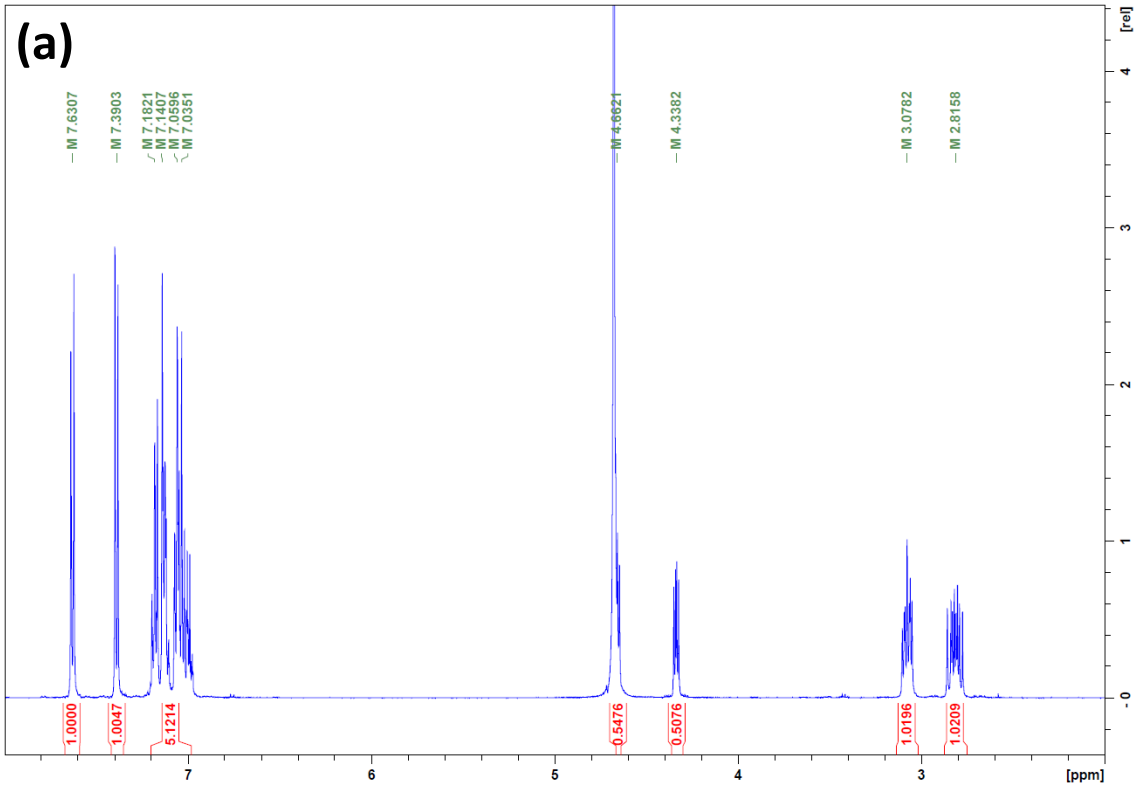
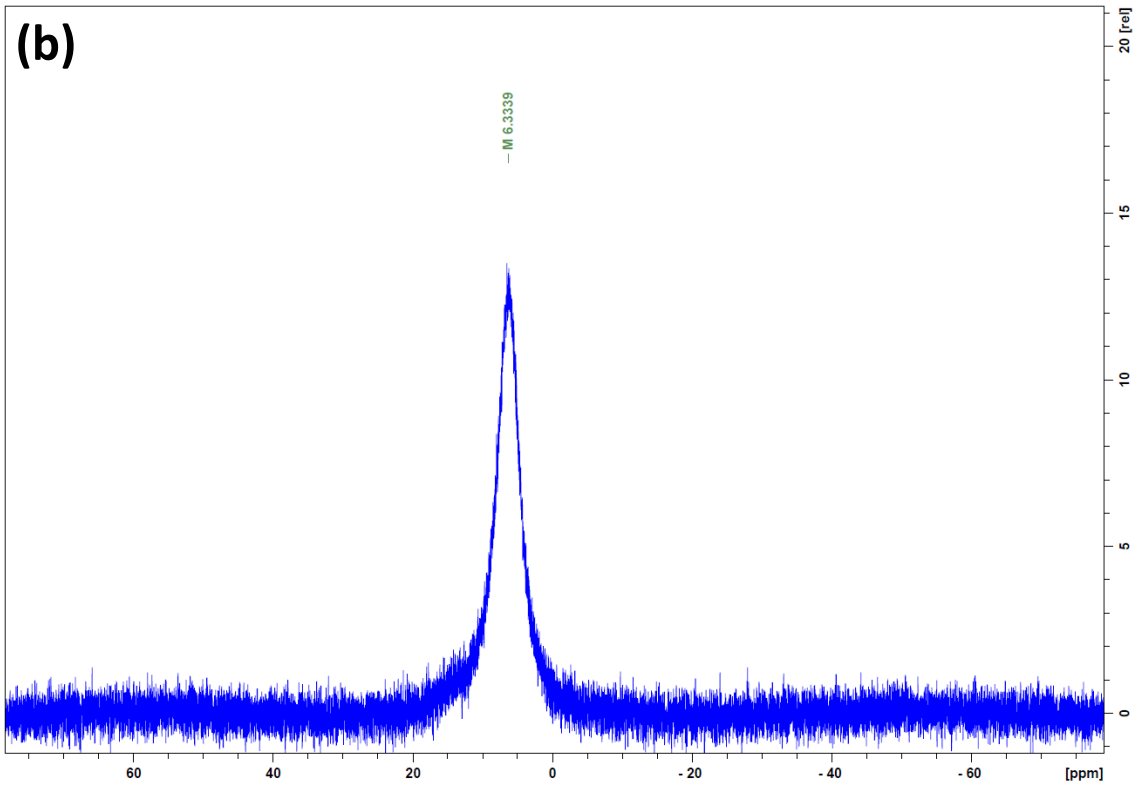


Figure S21. RP-HPLC chromatograph of compound **6**.


(a)
M 7.6307
M 7.3903
M 7.1821
M 7.1407
M 7.0596
M 7.0351
M 4.6621
M 4.3382
M 3.0782
M 2.8158
1.0000
1.0047
5.1214
0.5476
0.5076
1.0196
1.0209
[rel]
[ppm]


(b)
M 6.3339
[rel]
[ppm]

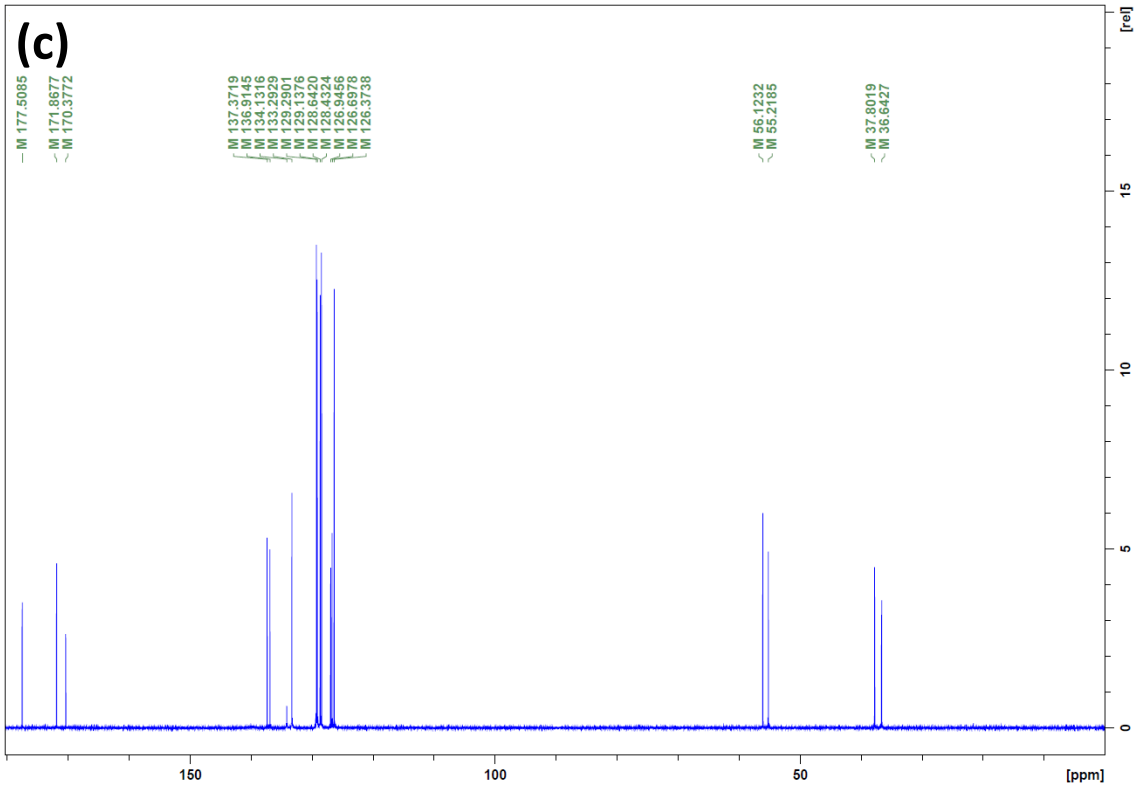


Figure S22. (a) ^{1}H, **(b)** ^{11}B, and **(c)** ^{13}C NMR spectra of compound **6** in D_2O with 0.25 vol. % ND_4OD.

References

1. Fransson, R.; Botros, M.; Sköld, C.; Nyberg, F.; Lindeberg, G.; Hallberg, M.; Sandström, A. *J. Med. Chem.* **2010**, *53*, 2383-2389.
2. Bose, A. K.; Srinivasan, P. R. *Tetrahedron* **1975**, *31*, 3025-3029.
3. Sene, S.; Bégu, S.; Gervais, C.; Renaudin, G.; Mesbah, A.; Smith, M. E.; Mutin, P. H.; van der Lee, A.; Nedelec, J.-M.; Bonhomme, C.; Laurencin, D. *Chem. Mater.* **2015**, *27*, 1242-1254.
4. Zhang, X.; Li, M.; Shi, Z.; Zhao, L.; Jin, R.; Yi, M.; Zhang, D.; Cui, Z. *Dyes Pigm.* **2012**, *92*, 982-987.
5. Wiskur, S. L.; Lavigne, J. J.; Ait-Haddou, H.; Lynch, V.; Chiu, Y. H.; Canary, J. W.; Anslyn, E. V. *Org. Lett.* **2001**, *3*, 1311-1314.