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Synthesis of sterically-hindered 1,7-dicarba-closo-dodecarborane thiourea analogs

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ABSTRACT

We herein report the first successful synthesis of 1-thioisocyanato-1,7-dicarba-closo-dodecarborane, which afforded novel lipophilic thiourea analogs by reacting with weak and bulky nucleophilic amines including 1-adamantylamine, and m-carboranylamine, aniline, and 2-aminopyridine. Newly synthesized thiourea analogs were fully characterized by NMR, ESI-MS, and FTIR.

1. Introduction

Carboranes ($C_2B_{10}H_{12}$), polyhedral borane clusters, have been evaluated as effective boron-delivery moieties for boron-neutron capture therapy (BNCT) for more than 50 years due to the fact that they can efficiently achieve the necessary boron concentrations for BNCT due to the high number of boron atoms per molecule [1–6]. Recently, they have been exploited as surrogates of lipophilic pharmacophores to replace hydrophobic benzene or cyclohexane rings in the field of medicinal chemistry because they can increase the lipophilicity of the parent drugs, improve their metabolic stability, and enhance their binding affinities for target proteins *via* hydrophobic interactions [7–14]. Furthermore, the unique features of carboranes such as high thermal stability, neutron shielding and electron transmission have also been applied in the field of polymer and material chemistry [15–18].

Isothiocyanate (N=C=S) is an important functional group in drug research and development. It has been investigated as the reactive electrophilic group toward nucleophiles of proteins in bioconjugate chemistry, leading to produce stable thioureas and sulfur-containing heterocyclic analogs [19–21]. Phenylisothiocyanate (PITC, Fig. 1) was used as reactant with N-terminal amino group of peptides in Edman degradation for sequencing amino acids of peptides. Isothiocyanate-containing fluorescent dyes such as fluorescein isothiocyanate (FITC) and rhodamine B isothiocyanate (RBITC) have been extensively utilized as labeling agents of peptides and proteins in the research field of flow cytometry and molecular imaging [22,23].

Carboranyl isothiocyanates are attractive boron-containing molecules because they can be easily conjugated with pharmacologically-active compounds and, thus, can be potentially utilized as lipophilic pharmacophores in medicinal chemistry and as boron carriers for BNCT. To the best of our knowledge, however, the successful synthesis of carboranyl isothiocyanates has never been reported up to date. We herein report the first simple and facile synthesis of *m*-carboranyl isothiocyanate and investigate its chemical reactivity with less nucleophilic amines such as aromatic/heteroaromatic amines and sterically hindered amines.

2. Experimental

2.1. General

m-Carborane was purchased from Katchem s.r.o. (Prague, Czech Republic). The other chemicals and solvents were purchased from Aldrich and Acros. ^{1}H NMR, ^{11}B NMR and ^{13}C NMR spectra were recorded on a BRUKER Biospin AVANCE 600 MHz and 300 MHz spectrometer. Chemical shifts are reported as δ values downfield from internal TMS in appropriate organic solutions. Mass spectra were recorded on a Agilent 6530 Accurate Q-TOF LC/MS spectrometer. IR spectra were recorded on Thermo NICOLET 6700 FT-IR

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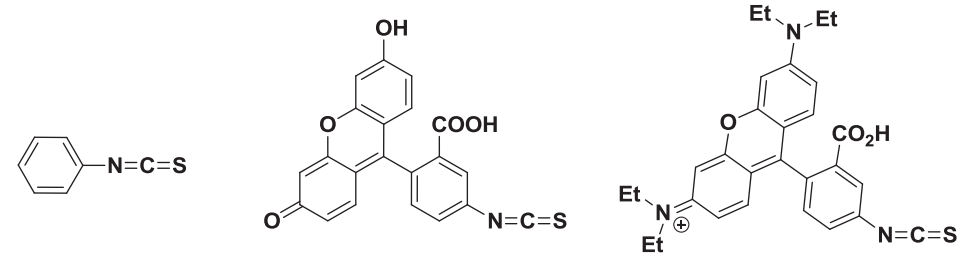


Phenylisothiocyanate (PITC) | Fluorescein isothiocyanate (FITC) | Rhodamine B isothiocyanate (RBITC)

Fig. 1. Isothiocyanate-containing molecules.

spectrometer. Column chromatography was performed using Merck Silica Gel F_{254}.

*2.1.1. 1,7-Dicarba-closo-dodecarborane-1-carboxylic acid (**1a**)*

To a solution of *m*-carborane (2.0 g, 13.9 m mol) in diethylether (150 mL) was added *n*-BuLi (9.6 mL, 15.4 m mol, 1.6 M in hexane) slowly over 20 min. The reaction mixture was stirred for another 20 min at −78 °C. Dry ice (5.0 g) was crushed into small pieces and added immediately to the reaction mixture, which was stirred for 1 h at room temperature. The excessive solvent was removed under reduced pressure. Water (60 mL) was added to the residue and unreacted *m*-carborane was extracted with hexanes (30 mL × 2). The aqueous layer was acidified with 3 N HCl and extracted with hexanes (30 mL × 4). The combined organic layer was dried over $MgSO_4$, filtered and concentrated under reduced pressure to afford **1a** in 95% yield as a white solid.

^{1}H NMR (600 MHz, $CDCl_3$): δ 10.48 (s, 1H, C_{car}—COOH), 3.03 (s, 1H, C_{car}—H), 1.27–3.28 (m, 10H, B_{car}—H). ^{13}C NMR (150 MHz, $CDCl_3$): δ 167.21 (C̲OOH), 71.04 (C̲$_{car}$—C), 54.84 (C̲$_{car}$—H).

*2.1.2. tert-Butyl-N-(1,7-dicarba-closo-dodecaborane-1-yl) carbamate (**1b**)*

To a solution of **1a** (0.950 g, 5.01 m mol) in *tert*-butanol (150 mL) was added triethylamine (2 mL, 14.34 m mol) and a catalytic amount of 4-DMAP (60 mg, 0.3 m mol) at room temperature, followed by the slow addition of diphenylphosphoryl azide (1.376 g, 5.00 m mol) over 5 min. The reaction mixture was stirred under reflux for 18 h at 120 °C. The excessive solvent was removed under reduced pressure. The residue was dissolved in ethyl acetate (100 mL) and washed with 1 N HCl solution, followed by brine. The organic layer was dried over $MgSO_4$, filtered and concentrated under reduced pressure. The crude product was subjected to the silica-gel chromatography (hexane/ethyl acetate = 10:1, R_f = 0.42) to afford **1b** in 72% yield as a white solid.

^{1}H NMR (600 MHz, $CDCl_3$): δ 5.19 (s, 1H, N—H), 2.90 (s, 1H, C_{car}—H), 1.43 (s, 9H), 1.28–3.78 (m, 10H, B_{car}—H). ^{13}C NMR (150 MHz, $CDCl_3$): δ 151.89 (C̲=O), 81.71(C̲—O), 81.07 (C̲$_{car}$—N), 52.86 (C̲$_{car}$—H), 28.13 (C̲H$_3$). HRMS (ESI^-): $[M - H]^-$, calcd for $C_7H_{20}B_{10}NO_2$ (*m/z*): 258.2445, found: 258.2445.

*2.1.3. 1-Amino-1,7-dicarba-closo-dodecarborane (**1c**)*

To a solution of compound **1b** in dichloromethane (15 mL) was added trifluoroacetic acid (4 mL). The reaction mixture was stirred for 4 h at room temperature. After monitoring the reaction progress, the excessive solvent was removed under reduced pressure. The crude product was dissolved in ethyl acetate (30 mL) and washed with water (30 mL) and with brine (30 mL). The organic layer was dried over $MgSO_4$, filtered and concentrated under reduced pressure. The crude product was subjected to the silica-gel chromatography (hexane/ethyl acetate = 9:1, R_f = 0.23) to afford **1c** in 63% yield as a white solid.

^{1}H NMR (600 MHz, $CDCl_3$): δ 2.89 (s, 1H, C_{car}—H), 2.30 (s, 2H, N—H), 1.28–3.78 (m, 10H, B_{car}—H). ^{13}C NMR (150 MHz, $CDCl_3$): δ 87.61 (C̲$_{car}$—N), 53.18 (C̲$_{car}$—H). ^{11}B NMR (192 MHz, $CDCl_3$): δ −3.27, −10.12, −11.96, −11.91. HRMS (ESI^-): $[M - H]^-$, calcd for $C_9H_{17}B_{10}N_2S$ (*m/z*): 160.2127, found: 160.2127.

*2.1.4. 1-Thioisocyanato-1,7-dicarba-closo-dodecarborane (**2**)*

To a solution of 1-amino-1,7-dicarba-closo-dodecarborane (230 mg, 1.44 m mol) in THF was added triethylamine (0.35 mL, 3.456 m mol) and thiophosgene (0.13 mL, 1.130 m mol) at −20 °C. The reaction mixture was stirred at −20 °C for additional 3 h. After monitoring the progress of the reaction by TLC, the excessive solvent was removed under reduced pressure. The residue was extracted with n-hexanes (30 mL) and washed with 1 N HCl solution (15 mL × 2). The combined organic layer was dried over $MgSO_4$, filtered and concentrated under reduced pressure. The crude product was subjected to the silica-gel chromatography (hexane/ethyl acetate = 500:3) to afford **2** in 88% yield as a white solid.

^{1}H NMR (600 MHz, $CDCl_3$): δ 2.94 (s, 1H, C_{car}—H), 1.73–3.43 (m, 10H, B_{car}—H). ^{13}C NMR (150 MHz, $CDCl_3$): δ 142.36 (N=C̲=S), 79.07 (C̲$_{car}$—N), 52.93 (C̲$_{car}$—H). ^{11}B NMR (192 MHz, $CDCl_3$): δ −3.59, −10.52, −11.80, −14.81, −15.28. HRMS (ESI^-): $[M - H]^-$, calcd for $C_3H_{10}B_{10}NS$ (*m/z*): 200.1537, found: 200.1542. FT-IR (cm^{-1}): 2611 (B̲$_{car}$—H), 2002 (N=C=S) cm^{-1}.

*2.1.5. 1-(1,7-Dicarba-closo-dodecaboran-1-yl)-3-phenyl thiourea (**3**)*

To a solution of **2** (80 mg, 0.40 m mol) in acetonitrile was added aniline (0.38 mL, 0.408 m mol) at −20 °C. The reaction mixture was stirred at −20 °C for 2 hrs. After monitoring the progress of the reaction by TLC, the excessive solvent was removed under reduced pressure. The residue was extracted with ethylacetate (10 mL) and washed with 1N HCl solution (10 mL × 2). The combined organic layer was dried over $MgSO_4$, filtered and concentrated under reduced pressure. The crude product was subjected to the silica-gel chromatography (hexane/ethylacetate = 5:1 R_f = 0.26) to afford **3** in 44% yield as a white solid.

^{1}H NMR (300 MHz, $CDCl_3$): δ 7.83 (s, 1H, NH), 7.29–7.48 (m, 5H, ArH), 6.45 (s, 1H, NH), 3.01 (s, 1H, C_{car}—H), 1.78–4.08 (m, 10H, B_{car}—H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 179.22 (C̲=S), 136.30 (Ar—C), 129.87 (Ar—C), 127.69 (Ar—C), 124.99 (Ar—C), 84.85 (C̲$_{car}$—N), 53.08 (C̲$_{car}$—H). ^{11}B NMR (192 MHz, CDCl3): δ −4.42, −10.90, −12.14, −14.81, −15.34. HRMS (ESI^-): $[M - H]^-$, calcd for $C_9H_{17}B_{10}N_2S$ (*m/z*):293.2116, found: 293.2122.

*2.1.6. 1-(1,7-Dicarba-closo-dodecaboran-1-yl)-3-(2-pyridyl) thiourea (**4**)*

Compound **4** was prepared according to the procedure described for compound **3** using 1-thioisocynato-1,7-dicarba-closo-dodecarborane (90 mg, 0.447 m mol) and 2-aminopyridine (42 mg, 0.446 m mol) at −20 °C for 2 h. Purification by silica-gel

chromatography (hexane/ethyl acetate = 4:1, R_f = 0.21) to afford **4** in 43% yield as a white solid.

^{1}H NMR (600 MHz, $CDCl_3$): δ 13.26 (s, 1H, NH), 8.15 (d, 1H, J = 5.2 Hz, 1.8 Hz), 8.12 (s, 1H, NH), 7.66 (dt, 1H, ArH, J = 7.2 Hz, 1.8 Hz), 7.01 (dd, 1H, ArH, J = 7.2 Hz, 5.1 Hz), 6.66 (d, 1H, ArH, J = 8.1 Hz), 2.95 (s, 1H, C_{car}—H), 1.78–4.10 (m, 10H, B_{car}—H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 178.66 (C═S), 152.42 (Ar—C), 145.39 (Ar—C), 118.52 (Ar—C), 118.90 (Ar—C), 82.95 (C_{car}—N), 52.68 (C_{car}—H). ^{11}B NMR (192 MHz, CDCl3): δ −3.19, −10.38, −12.49, −15.11, −15.42. HRMS (ESI^-): $[M - H]^-$, calcd for $C_8H_{16}B_{10}N_3S$ (*m/z*):294.2068, found: 294.2054.

2.1.7. 1-(1,7-dicarba-closo-dodecaboran-1-yl)-3-(tricyclo[3.3.1.1] decan-1-yl) thiourea (**5**)

Compound **5** was prepared according to the procedure described for compound **3** using 1-thioisocynato-1,7-dicarba-closo-dodecarborane (80 mg, 0.40 m mol) and 1-adamantylamine (52.5 mg, 0.40 m mol) at −20 °C for 2 h. Purification by silica-gel chromatography (hexane/ethyl acetate = 7:1, R_f = 0.32) to afford **5** in 48% yield as a white solid.

^{1}H NMR (300 MHz, $CDCl_3$): δ 6.41 (s, 1H, NH), 6.15 (s, 1H, NH), 2.98 (s, 1H, C_{car}—H), 2.23 (d, 6H, J = 2.4 Hz), 2.14 (d, 3H, J = 2.4 Hz), 1.71 (d, 6H, J = 2.4 Hz), 1.28–3.42 (m, 10H, B_{car}—H). ^{13}C NMR (150 MHz, $CDCl_3$): δ 177.66 (C═S), 82.66 (C_{car}—N), 55.95 (C—N), 52.81 (C_{car}—H), 40.90 (CH_2), 36.10 (CH_2), 29.41(CH). ^{11}B NMR (192 MHz, $CDCl_3$): δ −5.22, −11.33, −11.80, −14.73, −15.15. HRMS (ESI^-): $[M - H]^-$, calcd for $C_{13}H_{27}B_{10}N_2S$ (*m/z*):351.2898, found: 351.2904.

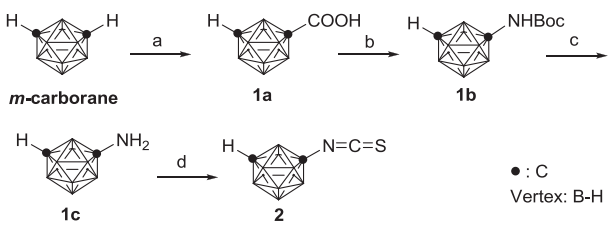
2.1.8. 1,3-Di(1,7-dicarba-closo-dodecaboran-1-yl) thiourea (**6**)

Compound **6** was prepared according to the procedure described for compound **3** using 1-thioisocynato-1,7-dicarba-closo-dodecarborane (80 mg, 0.40 m mol) and 1-amino-1,7-dicarba-closo-dodecarborane (63 mg, 0.395 m mol) at room temperature for 6 h. Purification by silica-gel chromatography (hexane/ethyl acetate = 8:1, R_f = 0.34) to afford **6** in 21% yield as a white solid.

^{1}H NMR (300 MHz, $CDCl_3$): δ 6.61 (s, 2H, NH), 2.99 (s, 2H, C_{car}—H), 1.28–3.42 (m, 10H, B_{car}—H). ^{13}C NMR (75 MHz, $CDCl_3$): δ 177.96 (C═S), 81.91 (C_{car}—N), 53.05 (C_{car}—H). ^{11}B NMR (192 MHz, $CDCl_3$): δ −4.50, −10.45, −11.10, −11.95, −14.77, −15.44. HRMS (ESI^-): $[M - H]^-$, calcd for $C_5H_{23}B_{20}N_2S$ (*m/z*): 359.3588, found: 359.3619. FT-IR (cm^{-1}): 2604 (B_{car}—H), 1548, 1370, 1266.

3. Results and discussion

The synthesis of 1-thioisocyanato-1,7-dicarba-closo-dodecarborane (*m*-carboranyl isothiocyanate) was achieved from commercial *m*-carborane in 4 steps as shown in Scheme 1. Compound **1c** (1-amino-1,7-dicarba-closo-dodecarborane), the key precursor for the synthesis of **2**, was synthesized by modifying the published synthetic procedures [24–26]. During the 2nd step for the preparation of **1c**, Curtius rearrangement was carried out using diphenylphosphoryl azide (DPPA) and a catalytic amount of 4-(dimethylamino)pyridine (DMAP) in *tert*-butanol (t-BuOH) at 120 °C for 18 h. The overall yield from *m*-carborane to **1c** in 3 steps was 33%. In order to convert **1** into the *m*-carboranyl isothiocyanate **2**, we first tried the mild reagents such as carbon disulfide/di-*tert*-butyl dicarbonate (CS_2/Boc_2O) and carbon disulfide/dicyclohexylcarbodiimide (CS_2-DCC), but the desired product was not obtained due to the low eletrophilicity of carbon disulfide and the moderate nucleophilicity of the carboranylamine group of **1c** [27,28]. Therefore, we employed thiophosgene, a highly reactive electrophile for the transformation of amines into the corresponding isothiocyanates, and could obtain the desired *m*-carboranyl isothiocyanate **2** by reacting it with **1c**.



Reagents and conditions. (a) i; *n*-BuLi, CO_2 (dry ice), ether, -78 °C --> rt, 2 hr, 95% (b) DPPA, TEA, 4-DMAP, *t*-BuOH, 120 °C, 18 hr, 72% (c) TFA/CH_2Cl_2, rt, 4 hr, 63% (d) Thiophosgene, TEA, THF, -20 °C, 2 hr, 88%

Scheme 1. Synthesis of *m*-carboranyl isothiocyanate **2**.

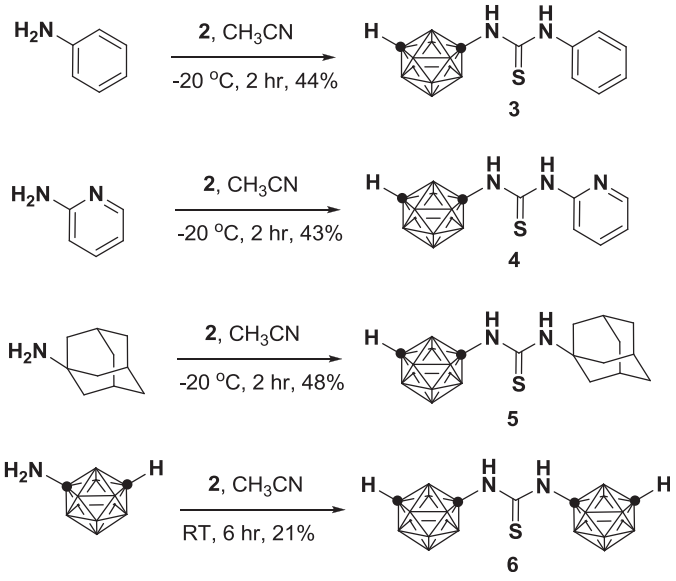
Among several different conditions, reaction of **1c** with thiophosgene in anhydrous THF at −20 °C for 3 h afforded **2** with the highest yield of 88% (see Table 1). Compound **2** was purified by silica gel chromatography using a mixture of hexane and ethyl acetate (500/3). TLC analysis studies showed that the R_f value of *m*-carboranyl isothiocyanate **2** (R_f: 0.73 in hexane only) is higher than those of phenylisothiocyanate (R_f: 0.51 in hexane only) and cyclohexylisothiocyanate (R_f: 0.32 in hexane only). The chemical structure of **2** was confirmed by ^{1}H NMR, ^{13}C NMR, ^{11}B NMR, FT-IR and ESI-HRMS (see all spectra in the Supporting information). The isothiocyanate carbon (N═C═S) of **2** has a chemical shift value of 142.26 ppm at the ^{13}C NMR spectrum. We also observed the diagnostic N═C═S stretching peak at 2002 cm^{-1} at the IR spectrum of **2**. Furthermore, ESI-HRMS in negative mode clearly showed an isotope pattern of carborane with a highest $[M - H]^-$ peak of **2** at 200.1538. This is the first report on the successful synthesis of carboranyl isothiocyanate. It is noted that the isothiocyanate group of **2** is directly linked to the carbon atom of *m*-carborane.

To prepare novel lipophilic *m*-carboranyl thioureas, we selected aromatic or bulky amines as nucleophiles. The tested amines were aromatic aniline, heteroaromatic 2-aminopyridine, sterically hindered 1-adamantylamine, and *m*-carboranylamine (Scheme 2). Our initial attempt to react **2** with an excess of aniline at room temperature produced the desired thiourea **3** in a very low yield (<3%). The corresponding *nido* form of **3** (molecular formula:$C_9H_{18}B_9N_2S_1$, the mass spectrum in the Supporting information) turned out to be a major product due to the fact that the excess aniline degraded the closo carborane into the *nido* one at room temperature. To minimize the production of by-products, **2** was treated with 1 equivalent of aniline in anhydrous acetonitrile (CH_3CN) at low temperature. We obtained 1-(1,7-dicarba-closo-dodecaboran-1-yl)-3-phenyl thiourea **3** in 44% yield at −20 °C with a trace amount of the *nido* compound (Scheme 2). By employing the same synthetic procedure to prepare **3**, compounds **4** and **5** were obtained by reacting **2** with 2-aminopyridine and 1-adamantylamine at −20 °C in 43% and 48% yields, respectively. Interestingly, compound **5**, 1-(1,7-dicarba-closo-dodecaboran-1-yl)-3-(tricyclo [3.3.1.1]decan-1-yl) thiourea, was precipitated during the reaction

Table 1
Reaction conditions for the conversion of **1c** into **2**.

Entry	Reagents	Temperature	Yield
1	$CSCl_2$	RT	22%
2	$CSCl_2$	0 °C	62%
3	$CSCl_2$	−20 °C	88%
4	$CSCl_2$	−78 °C	53%
5	CS_2/Boc_2O[a]	RT	Trace
6	CS_2/DCC[b]	RT	Trace

[a] CS_2 (10 eq), triethylamine (1 eq), EtOH, 0 °C, 30 min → Boc_2O (1 eq), DMAP (0.1 eq), rt, 2 h.
[b] CS_2 (10 eq), DCC (1 eq), THF, −10 °C, 10 min → rt, 2 h.



Scheme 2. Synthesis of *m*-carboranyl thiourea analogs **3**–**6**.

Table 2
Featured spectroscopic properties of the *m*-carboranyl isothiocyanate **2** and *m*-carboranyl thiourea analogs **3**–**6**.

Compound	^{1}H NMR	^{13}C NMR	HRMS $[M-H]^-$	Yield
2	2.94 (C_{car}–H)	142.36 (N=C=S) 79.07 (C_{car}–N) 52.93 (C_{car}–H)	200.1542	88%
3	7.83 (NH) 6.45 (NH)3.01 (C_{car}–H)	179.22 (C=S) 84.85 (C_{car}–N) 53.08 (C_{car}–H)	293.2122	44%
4	13.26 (NH) 8.12 (NH) 2.95 (C_{car}–H)	178.66 (C=S) 82.95 (C_{car}–N) 52.68 (C_{car}),	294.2054	43%
5	6.41 (NH) 6.15 (NH) 2.98 (C_{car}–H)	177.66 (C=S) 82.66 (C_{car}–N) 52.81 (C_{car}–H)	351.2904	48%
6	6.61 (NH) 2.99 (C_{car}–H)	177.96 (C=S) 81.91 (C_{car}–N) 53.05 (C_{car}–H)	359.3588	21%

and purified by simple filtration. However, *m*-carboranylamine when treated with **2** at −20 °C afforded the corresponding thiourea **6** in a yield lower than 5%. The starting materials **2** and *m*-carboranylamine remained unreacted, suggesting that the weaker nucleophilicity of the *m*-carboranylamine than the other amines may lower the conjugation yield. After a longer reaction time of 6 h at room temperature, 21% conversion of **2** into 1,3-di(1,7-dicarba-closo-dodecaboran-1-yl) thiourea **6** occurred.

The chemical structures of the synthesized compounds **3**–**6** were fully analyzed and confirmed by ^{1}H NMR, ^{13}C NMR, ^{11}B NMR, and ESI-HRMS. The diagnostic signal for the carbon of thiourea (NHC(=S)NH) appeared at around 179 ppm from the ^{13}C NMR spectra. We also observed the broad singlet peaks for two NHs from the ^{1}H NMR spectra and carborane isotope patterns with a highest $[M-H]^-$ peak from ESI-HRMS spectra of **3**–**6** (see all the spectra in Supporting information). Featured spectroscopic properties of **3**–**6** and the *m*-carboranyl isothiocyanate **2** are summarized in Table 2.

4. Conclusions

We have successfully achieved the first synthesis of the *m*-carboranyl isothiocyanate in high yield. Reaction of the *m*-carboranyl isothiocyanate with the bulky and weak nucleophilic amines afforded four novel sterically-hindered *m*-carboranyl thiourea analogs in moderate yield. The newly synthesized compounds were fully characterized by means of ^{1}H NMR, ^{13}C NMR, ^{11}B NMR, and ESI-HRMS. The unique chemical and physiochemical properties of the *m*-carboranyl isothiocyanate have a potential to be utilized as boron-delivery moiety for BNCT and lipophilic pharmacophore in medicinal chemistry.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.jorganchem.2013.03.003.

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