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# HIGH-THROUGHPUT SYNTHESIS OF SUBSTITUTED HYDRAZINE DERIVATIVES

**Michio Kurosu,* Prabagaran Narayanasamy, and Dean C. Crick**

*This paper is dedicated to the memory of Professor Ivar Ugi, an inspirational scientist.*

Department of Microbiology, Immunology, and Pathology, College of Veterinary Medicine and Biomedical Sciences, Colorado State University, 1682 Campus Delivery, Fort Collins, CO 80523-1682, USA.

E-mail: michio.kurosu@colostate.edu

**Abstract** – Regioselective alkylations of the hydrazine derivatives are achieved by using the (2,6-dichloro-4-methoxyphenyl)(2,4-dichlorophenyl)methoxycarboxyl resin. High-throughput synthesis of monosubstituted and 1,1-disubstituted hydrazine building blocks is described.

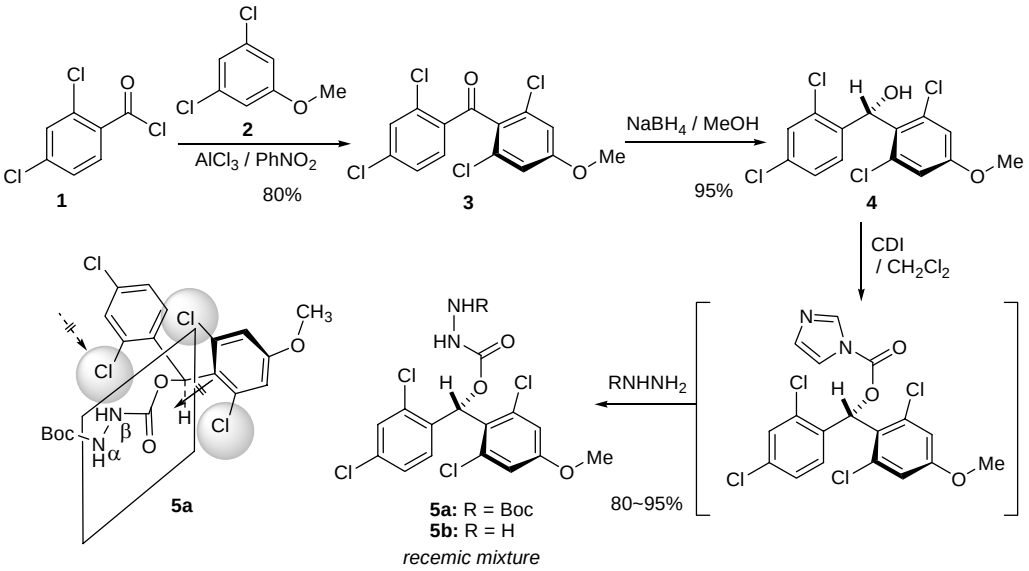
We recently validated that MenA (1,4-dihydroxy-2-naphthoate prenyltransferase),[1] the sixth enzyme in menaquinone biosynthesis, is a novel target for the development of new drug leads for MDR Gram-positive pathogens.[2] In the discovery of MenA inhibitors, we have generated a library of molecules based on the 4-alkoxydiphenylmethanones which contain *tertiary*, *secondary* amines and hydrazines and the library of molecules was evaluated in an enzymatic assay *in vitro* ($IC_{50}$) against MenA, and bacterial growth assays (MIC).[3] From these synthesis-assay processes it was found that the structure of hydrazine significantly influences MenA and bacterial growth inhibitory activities. Thus, in an attempt to deliver target-specific library for the development of MenA inhibitors, it is indispensable to diversify the library structure with a variety of hydrazine building blocks. However, generation of such a library has been limited by the lack of availability of diverse structures of hydrazine molecules[4] from commercial sources.

Several synthetic methods for monoalkylations of the hydrazines using urethane or phthaloyl protecting group were independently reported by Jamart-Grégoire and Ragnarsson.[5] Recently Mäeorg extensively studied on alkylations of PhNHNHBoc with $^{n}$BuLi.[6] Although these methods are useful for alkylations of the specific substrates, their procedures involve multiple steps for protection and deprotection or require

carefully controlled amount of reagents and temperatures. To date, no high-throughput synthesis of substituted hydrazine derivatives have been reported.[7]

To efficiently perform selective alkylations of functionalized hydrazines on polymer-support, we devised a protecting group for hydrazine nitrogen that 1) can be cleaved by a volatile and mild acid such as TFA, 2) has stability to relatively strong bases, and 3) create a sterically encumbered environment which prevents overalkylations of the protected $N^{\beta}$ atom of hydrazine unit. In addition, such a protecting group should be easily grafted onto a polymer-support by a reliable chemical transformation.

We have identified that the (2,6-dichloro-4-methoxyphenyl)(2,4-dichlorophenyl)methyl hydrazinecarboxylate derivative **5b** (Scheme 1), conveniently synthesized by Friedel-Crafts reactions followed by $NaBH_4$ reduction, and CDI-mediated urethane formation reactions, showed an unusual stability to acids such as 10~15% TFA, 30% HF, 2N HCl, HBr/AcOH, $TiCl_4$, $ZnCl_2$, $AlCl_3$, $B(C_6F_5)_3$, $BCl_3$, $BBr_3$, TMSOTf, and $La(OTf)_3$ at rt, bases such as 40% $NH_4OH$, 10% LiOH, 6N NaOH, TlOEt, and DBU at rt, and nucleophiles such as *primary* and *secondary* amines at 80 °C, and $NH_2NH_2$ at rt. However, The urethane **5b** could conveniently be deprotected by using 20% TFA in $CH_2Cl_2$ to afford hydrazine•TFA salt and (2,6-dichloro-4-methoxyphenyl)(2,4-dichlorophenyl)methyl 2,2,2-trifluoroacetate in quantitative yields.

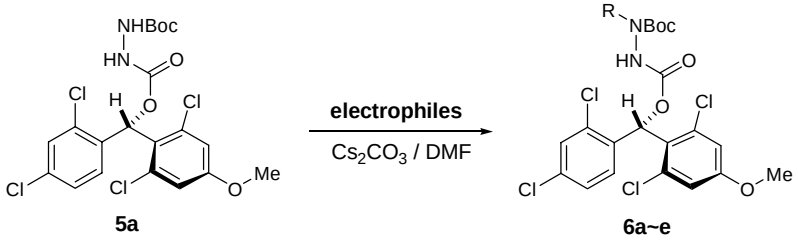



**Scheme 1.** Synthesis of (2,6-dichloro-4-methoxyphenyl)(2,4-dichlorophenyl)methylhydrazine carboxylate derivatives, **5a** and **5b**.

We first investigated the reactivity of **5a**[8] against representative electrophiles such as benzyl bromide, allyl bromide, alkyl iodide, and methyl 2-chloroacetate in the presence of $Cs_2CO_3$ in DMF. In order to determine the degree of overalkylation at the $N^{\beta}$ position of **5a**, a large excess of electrophiles was utilized in all reactions. Gratifyingly no overalkylations of **5a** were observed in the reactions with electrophiles listed in

Table 1 even at 50 °C (for the stable electrophiles). On the other hand, the same reactions with BocHNNHBoc gave rise to significant amounts of $N^{\alpha}$,$N^{\beta}$-dialkylsubstituted products.

**Table 1.** Alkylations of **5a** with representative electrophiles.



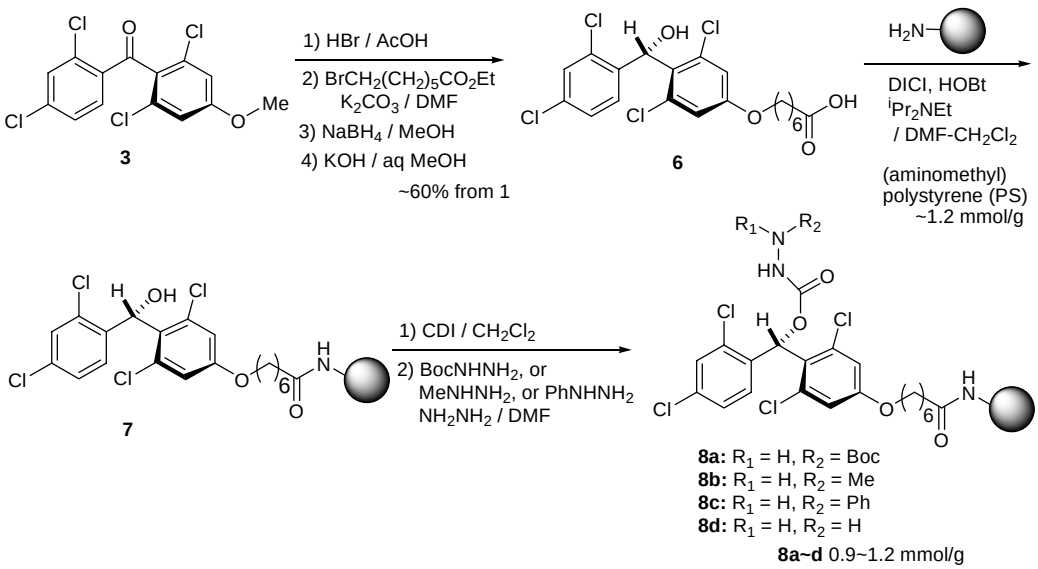

**5a** **6a~e**

| entry | electrophile[1] | temperature (°C) | time (h)[2] | product | yield (%)[3] |
|---|---|---|---|---|---|
| 1 | *p*-ClPhCH$_2$Br | 50 | 12 | **6a**[9] | 91 |
| 2 | allyl bromide | 25 | 12 | **6b** | 92 |
| 3 | $C_8H_{17}I$ | 50 | 12 | **6c** | 90 |
| 4 | $CH_3I$ | 5 | 12 | **6d** | 95 |
| 5 | $ClCH_2CO_2Me$ | 50 | 12 | **6e** | 60[4] |

[1] A large excess (10~15 equiv) of electrophile was used. [2] The reaction was completed within 6 h.
[3] Isolated yield. [4] The reaction requires a longer time to achieve complete conversion.

Through X-ray analysis of (2,6-dichloro-4-methoxyphenyl)(2,4-dichlorophenyl)methyl acetate and molecular modeling of **5b**,[10] the origin of the excellent regioselectivity of alkylations of **5a** and stability against bases, acids, and nucleophiles can be attributed to the following reasons. The dihedral angles formed by two planar chlorosubstituted-benzenes and by -CO-O- linkage and the ether methine proton are 83.5° and 159.8°, respectively. There must be a significant electronic repulsion between the *o*-chloro atoms in two benzene rings. The *o*-chloro atom in dichlorobenzene locates towards the carbonyl ester plane. Thus, the chloro atoms at *o*-positions in two benzenes hinder nucleophilic attacks at the ester carbonyl from both *re*- and *si*-faces. The 3,5-dichloro atoms in the anisole moiety attenuate an electron donating character of the methoxy group. Therefore, The urethane **5b** would exhibit stability against bases, nucleophiles, and acids. To date, the generation of dianion of the hydrazine dicarboxylates under $Cs_2CO_3$ in DMF at 50 °C has not been reported.[6] Thus, the observed $N^{\alpha}$,$N^{\beta}$-dialkylation of BocHNNHBoc could be explained by a stepwise process. Nevertheless, the generation of anion at the $N^{\beta}$ position or the approach of electrophiles is prohibited because of the stereoelectronic factors governed by two planar chlorosubstituted-benzenes.

(2,6-Dichloro-4-hydroxyphenyl)(2,4-dichlorophenyl)methanone, which was obtained in ~70% overall yield by the treatment of the Friedel-Crafts products with sat. HBr in AcOH / water (2:1) followed by washing the crude solid with $CHCl_3$,[11] was converted to the hydroxy-carboxylic acid possessing C6-linker

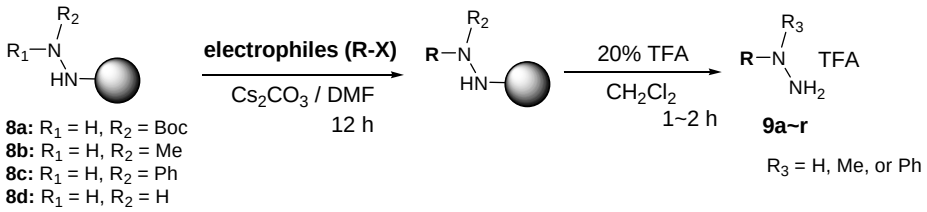



**Scheme 2.** The grafting of **6** on the PS resins and the syntheses of the PS-hydrazines.

**6** in three steps in 90% overall yield (1. $NaBH_4$ reduction, 2. alkylation with ethyl 7-bromoheptanoate, and 3. saponification reactions). The hydroxy-carboxylic acid **6** could be grafted onto the (aminomethyl)polystylene (~1.2 mmol/g)[12] using a reliable coupling condition [DICI, HOBt, $^{i}Pr_2NEt$ / DMF-$CH_2Cl_2$ (1/1)]. The resins **7** were then converted to the carbonylimidazole linker which exhibited excellent reactivities against hydrazine and monosubstituted hydrazines including $BocNHNH_2$, $MeNHNH_2$, and $PhNHNH_2$ to provide **8a~d** whose loading yields were determined to be 0.9~1.2 mmol/g by $^{1}$H-NMR analyses of the crude materials, after cleavage from the resins using 20% TFA in $CH_2Cl_2$ for 2 h.[13]

The alkylations observed with **5a** in solution were well transferred to the reactions with the resin **8a**. As summarized in Table 2[13], the reactions of **8a** with the substituted benzyl bromides (10~15 eqiv) in DMF in the presence of $Cs_2CO_3$ (5 equiv) at 50 $^{o}$C for 12 h gave the corresponding monobenzylated hydrazine•TFA salts **9a~f** in good yields, after cleavage from the resins with 20% TFA. Alkylation of **8a** with *n*-octyl iodide followed by cleavage from the resin afforded 1-octylhydrazine•TFA salt (**9g**) in excellent yield. Allylation of **8a** smoothly underwent at rt ~ 50 $^{o}$C to provide 1-allylhydrazine•TFA salt, after cleavage. In these reactions, no overalkylations at the $N^{\beta}$ position were observed. Similarly, alkylations of **8b** and **8c** gave the corresponding $N^{\alpha}$,$N^{\alpha}$-dialkylated hydrazine•TFA salts without detectable contamination of by-products in $^{1}$H-NMR spectra. Symmetrical $N^{\alpha}$,$N^{\alpha}$-dialkylated hydrazine•TFA could also be synthesized with the resin **8d** with excellent yields. It is worthwhile noting that the solvolytic displacement reactions of **9a~r** with TFA provide TFA ester of **7** in near quantitative yield and the alcohol resin **7** can be regenerated by the treatment with aq. $NH_4OH$ in THF-MeOH for 12 h. The regenerated resins could be reused for the synthesis of **9a** without noticeable decrease of yield.

**Table 2.** High-throughput syntheses of monosubstituted and 1,1-disubstituted hydrazine derivatives.[14]




| entry | starting material | electrophile (R) | $R_3$ | product | yield (%)[3] |
|---|---|---|---|---|---|
| 1 | **8a** | *p*-$ClC_6H_4CH_2Br$ | H | **9a** | 95 |
| 2 | **8a** | *p*-$CF_3OC_6H_4CH_2Br$ | H | **9b** | 93 |
| 3 | **8a** | *m*-$FC_6H_4CH_2Br$ | H | **9c** | 95 |
| 4 | **8a** | *o*-$FC_6H_4CH_2Br$ | H | **9d** | 92 |
| 5 | **8a** | *p*-$MeSC_6H_4CH_2Br$ | H | **9e** | 91 |
| 6 | **8a** | allyl bromide | H | **9f** | 98 |
| 7 | **8a** | *n*$C_8H_{17}I$ | H | **9g** | 92 |
| 8 | **8b** | *p*-$ClC_6H_4CH_2Br$ | Me | **9h** | 95 |
| 9 | **8b** | *p*-$CF_3OC_6H_4CH_2Br$ | Me | **9i** | 93 |
| 10 | **8b** | *n*$C_8H_{17}I$ | Me | **9j** | 92 |
| 11 | **8b** | $CH_3I$ | Me | **9k** | 98 |
| 12 | **8b** | allyl bromide | Me | **9l** | 97 |
| 13 | **8c** | *n*$C_8H_{17}I$ | Ph | **9m** | 92 |
| 14 | **8c** | allyl bromide | Ph | **9n** | 95 |
| 15 | **8c** | $CH_3I$ | Ph | **9o** | 91 |
| 16 | **8d** | allyl bromide | allyl | **9p** | 94 |
| 17 | **8d** | $C_6H_5CH_2Br$ | $C_6H_5CH_2$ | **9q** | 94 |
| 18 | **8d** | *n*$C_8H_{17}I$ | $C_8H_{17}$ | **9r** | 92 |

[1] A large excess (10~15 equiv) of electrophile and $Cs_2CO_3$ (5 equiv) were used. [2] The reaction was completed within 6 h. [3] Isolated yield. [4] The reaction requires a longer time to achieve complete conversion.

In conclusion, we have developed a high-throughput synthesis of mono and disubstituted hydrazine derivative using the tetrachlorodiphenylmethoxycarbonyl linker. The (2,6-dichloro-4-methoxyphenyl)(2,4-dichlorophenyl)methyl moiety in the hydrazino-urethanes completely blocks the alkylation at the $N^{\beta}$ position. In addition, the tetrachlorodiphenylmethoxycarbonyl linker developed here is stable to a wide variety of bases and can be cleaved under a mild condition (20% TFA). Thus, monosubstituted and 1,1-disubstituted hydrazines can be reliably synthesized with high yield and purity. Because monosubstituted and 1,1-disubstituted hydrazines are important building blocks for the generation

of active MenA inhibitors, we have demonstrated the syntheses of specific structure of hydrazine derivatives as summarized in Table 2. However, *tert*-butyl 2-alkylhydrazinecarboxylates (i.e. BocHNNHMe) were loaded onto the tetrachlorodiphenylmethoxycarbonyl linker at 50 $^{o}$C and the Boc group could be removed selectively by using 15% TsOH•$H_2O$ in THF-$CH_2Cl_2$ (1/1).[13] Thus, the method described here may be applicable to the synthesis of 1,2-disubstituted and 1,1,2-trisubstituted hydrazine derivatives in a high-throughput manner. Generation of library molecules using the hydrazine building blocks synthesized by the described method and evaluation of these molecules against MenA and MDR Gram-positive bacteria will be reported elsewhere.

## ACKNOWLEDGEMENTS

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## REFERENCES AND NOTES

1. K. D. Suvana, R. Stevenson, R. Meganathan, and M. E. S. Hudspeth, *J. Bacteriol*., 1998, **180**, 2782.
2. M. Kurosu and D. C. Crick, Compositions and Methods of Use of Electron Transport System Inhibitors Serial Number: PCT / US2007 / 63122.
3. M. Kurosu, P. Narayanasamy, K. Biswas, R. Dhiman, and D. C. Crick, *J. Med. Chem.,* 2007 in press.
4. For a recent report on the alkylation of protected hydrazines, see: L. K. Rasmussen, *J. Org. Chem*., 2007, **71**, 3627; J. Barluenga, P. Moriel, F. Aznar, and C. Valdés, *Org. Lett.,* 2007, **9**, 275 and references therein. For a review, see: A. Zergra, *Curr. Med. Chem.,* 2005, **12**, 589.
5. N. Brosse, M. F. Pinto, J. Bodiguel, and B. Jamart-Grégorire, *J. Org. Chem*., 2001, **66**, 2869; O. Tśubrik and U. Mäeorg, *Org. Lett.,* 2001, **3**, 2297; N. Brosse, M. F. Pinto, and B. Jamart-Grégorire, *J. Org. Chem*., 2000, **65**, 4370; U. Mäeorg and U. Ragnarsson, *Tetrahedron Lett*., 1998, **39**, 681.
6. A. Bredihhin, U. M. Groth, and U. Mäeorg, *Org. Lett.,* 2007, **9**, 1097.
7. *N*-Amination reactions have been widely demonstrated in the synthesis of 1,1-disubstituded hydrazines, see: U. Ragnarson, *Chem. Soc. Rev.*, 2001, **3**0, 205.
8. Data for **5a**: $^{1}$H-NMR ($CDCl_3$, 300 MHz): δ 7.55 (s, 1H), 7.37 (m, 1H), 7.26 (m, 2H), 6.88 (s, 2H), 6.34 (bs, 1H ), 6.31 (bs, 1H), 3.81 (s, 3H), 1.46 (s, 9H).; $^{13}$C-NMR ($CDCl_3$, 100 MHz): 159.9, 155.3, 136.9, 130.5, 129.7, 126.5, 124.3, 115.3, 81.3, 72.4, 55.8, 28.1.; IR (neat, cm$^{-1}$): 3583, 2919, 2362, 1652, 1558. LRMS (ESI) $C_{20}H_{21}N_2O_5Cl_4$ ($M+H^+$) 509.0.
9. Data for **6a**: $^{1}$H-NMR ($CDCl_3$, 300 MHz): δ 7.56 (s, 1H), 7.25 (m, 7H), 6.88 (s, 2H), 6.36 (bs, 1H ),

4.72 (m, 2H), 3.81 (s, 3H), 1.42 (s, 9H).; $^{13}$C-NMR ($CDCl_3$, 100 MHz): 159.9, 154.9, 154.2, 137.0, 134.7, 134.2, 133.7, 130.6, 130.1, 129.5, 128.7, 126.4, 124.3, 115.2, 81.7, 72.8, 55.7, 52.9, 28.0.; IR (neat, $cm^{-1}$): 3583, 2919, 2367, 1652, 1558, 1540.; LRMS (ESI) $C_{27}H_{26}N_2O_5Cl_5$ ($M+H^+$) 633.4.

10. A SPARTAN conformational search by semiempirical calculation using AM1 basis set indicate that lowest energy conformation of (2,6-dichloro-4-methoxyphenyl)(2,4-dichlorophenyl)methoxy carbonyl moiety of **5b** was well in agreement with that of X-ray structure of (2,6-dichloro-4-methoxyphenyl)(2,4-dichlorophenyl)methyl acetate.
11. (2,6-Dichloro-4-hydroxyphenyl)(2,4-dichlorophenyl)methanone showed very poor solubility against $CHCl_3$, hexanes, and $Et_2O$, but dissolved in MeOH. Thus, this compound could be isolated without using chromatographic purification.
12. Purchased from Aldrich.
13. M. Kurosu, K. Biswas, and D. C. Crick, *Org. Lett.*, 2007, **9**, 1141.
14. The following example represents typical experimental procedure: The resins **8a** (100 mg, 0.1 mmol) in DMF (1 mL) was added $Cs_2CO_3$ (162.5 mg, 0.5 mmol) and *p*-$ClC_6H_4CH_2Br$ (204 mg, 1.0 mmol). The reaction mixture was gently stirred at 50 $^oC$ for 12 h. The resins were washed with DMF, THF-water (3/1), THF, and EtOAc, and dried under high vacuum. The resins were suspended in $CH_2Cl_2$ (1 mL) and TFA (~0.2 mL) was added. After 1 h the resins were filtered and washed with $CH_2Cl_2$. The combined $CH_2Cl_2$ solution was evaporated at 30~40 $^oC$ and dried under high vacuum. $^1$H-NMR spectroscopy was applied to the analysis of the crude product using 1-bromo-4-methoxybenzene as an internal standard.