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Syntheses and radical scavenging activities of resveratrol derivatives

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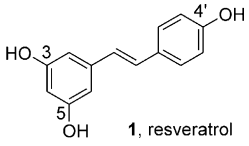
Abstract—Nine new resveratrol derivatives, having bromo, iodo, and fluoroethyl groups, were designed and synthesized. All compounds having free phenol groups showed good free radical scavenging activity. Among them, 2-bromoresveratrol **19** has a similar free radical scavenging activity to (+)-catechin.

1. Introduction

Resveratrol (**1**), 3,4′,5-trihydroxy-*trans*-stilbene found in grapes and a variety of medicinal plants,[1] is a naturally occurring phytoalexin that protects against fungal infections. Its biological properties[2] include antifungal,[3] antibacterial,[4] anticancer,[5] antiviral,[6] estrogenic,[7] platelet antiaggregating,[8] and heart protecting activities.[9] Despite high fat diet and heavy smoking habits, the Southern French people have very low incidence of coronary heart disease (CHD). This so-called *French paradox* has strongly been related with wine consumption.[1f] Frankel et al. have shown that total phenolic compounds extracted from red wine inhibit the oxidation of human low-density lipoproteins (LDL).[10] As the oxidized LDL may be responsible for promoting atherogenesis, the phenolic compounds in red wine could be the cause of the French paradox.[11]



1, resveratrol

Although resveratrol has numerous biological activities in vitro, there is little data about its bioavailability and tissue distribution in vivo. Recently, Mérillon et al. reported the absorption and tissue distribution of [^{14}C]-*trans*-resveratrol following oral administration to mice.[12] The results demonstrated that *trans*-resveratrol is bioavailable following oral admistration and remains mostly in intact form. There are also not many studies on the structure–activity relationship (SAR) of resveratrol.[11,13] Fauconneau et al. published the comparative study of radical scavenger and antioxidant properties of natural phenolic compounds isolated from *vitis vinifera* cell cultures.[11] Ho et al. evaluated the hydroxylated derivatives of resveratrol as potential antioxidants.[13a] Several glucopyranoside derivatives of resveratrol were isolated, synthesized, and evaluated by Orsini et al.[13a]

Positron emission tomography (PET) and single photon emission computed tomography (SPECT) are widely used for medical imaging. Bromine-76 ($t_{1/2}=16$ h), iodine-123 ($t_{1/2}=13.1$ h), and fluorine-18 ($t_{1/2}=110$ min) are very attractive radionuclides for the labeling of organic molecules as PET and SPECT radiotracers, because of their many favorable characteristics such as stable bonding to carbon, convenient half-life, and ease of production.[14] Labeled resveratrols with either positron or single photon emitted radionuclides are important for studying biological behaviors and activities in vivo as well as in vitro. In this report, we have designed and synthesized several bromo, iodo, and fluoro resveratrol derivatives. Through their free radical scavenging activities and the amount of total polyphenol, we obtained information on the SAR of resveratrol.

Keywords: Resveratrol; Radical scavenger; Stilbene; Antioxidant; Phenol.
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2. Chemistry

Resveratrol is one of unsymmetrical and (*E*)-geometrical stilbene. Among the various methods to make unsymmetrical stilbenes,[15] we have used the general methodology for the synthesis of the derivatives of resveratrol as shown in Scheme 1. Wittig condensation of appropriate aldehydes with phosphonium salts which were generated from *O*-protected 3,5-dihydroxybenzyl bromides **2**–**6** afforded stilbenes **7**–**15** in 74–93% yields. The stilbenes were a mixture of (*E*) and (*Z*)-geometrical isomers with the ratio of 2:1. These *E*/*Z* mixtures efficiently converted to (*E*)-geometrical isomers through heating with the catalytic amount of I_2 in refluxing heptane for 12 h.[16]

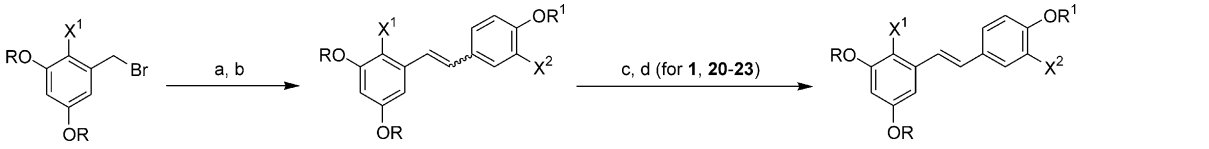
We tried using methyl group for the protective group of phenols. But with the exception of resveratrol, the other 3,4′,5-trimethylated resveratrol derivatives with bromo (**7**, **9**), iodo (**8**), or fluoroethyl groups decomposed during the demethylation step using BBr_3 in dichloromethane[13a] and other reagents. Thus we have used the MOM and TBS protecting groups of phenols for the syntheses of bromo, iodo, and fluoroethylated derivatives.

Scheme 2 shows the synthesis of benzyl bromides for the precursors of Wittig reagents, and Schemes 3 and 4 show the synthesis of aldehyde precursors of Wittig reagents. The synthetic route for the introduction of fluoroethyl group[17] included palladium coupling reaction, hydroboration, tosylation, and fluorination[18] as shown in Scheme 4. When we synthesized a fluoroethylated compound, classical displacement of sulfonate with fluoride ion gave mostly eliminated by-product with very low yield of desired fluoroethyl compound. We have recently reported a new fluorination method using ionic liquid [bmim][BF_4], providing fluoroethyl compound as a major product,[18] which allowed the synthesis of fluoroethyl compound in high yield. Nine new compounds were synthesized according to Scheme 1 and 4-iodoresveratrol was selectively prepared by direct iodination of unprotected resveratrol using I_2 in MeOH at rt for 2 h (Scheme 5).

3. Antioxidant studies

3.1. Free radical scavenging activity

The DPPH (1,1-diphenyl-2-picrylhydrazyl) radical scavenging effect was carried out according to the method first employed by Blois.[21] The 100 μL of sample solution was added to 900 μL of DPPH solution in ethanol (1.01×10^{-4} M). After incubation at rt for 30 min,

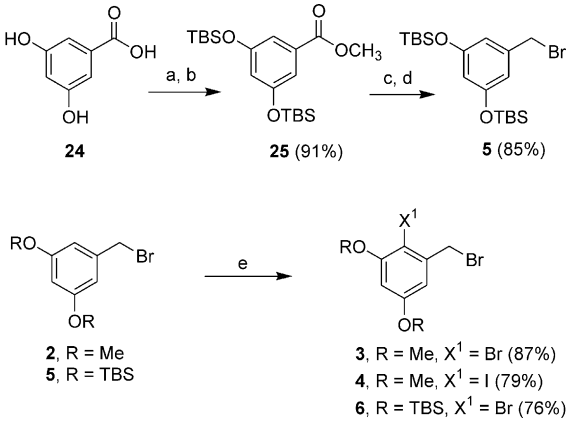


2, R = Me, X^1 = H
3, R = Me, X^1 = Br
4, R = Me, X^1 = I
5, R = TBS, X^1 = H
6, R = TBS, X^1 = Br

7, R = R^1 = Me, X^1 = Br, X^2 = H (93%)
8, R = R^1 = Me, X^1 = I, X^2 = H (92%)
9, R = R^1 = Me, X^1 = H, X^2 = Br (95%)
10, R = R^1 = TBS, X^1 = H, X^2 = H (83%)
11, R = R^1 = TBS, X^1 = Br, X^2 = H (74%)
12, R = R^1 = TBS, X^1 = H, X^2 = Br (81%)
13, R = R^1 = TBS, X^1 = H, X^2 = I (83%)
14, R = R^1 = TBS, X^1 = H, X^2 = CH_2CH_2F (85%)
15, R = Me, R^1 = TBS, X^1 = H, X^2 = CH_2CH_2F (80%)

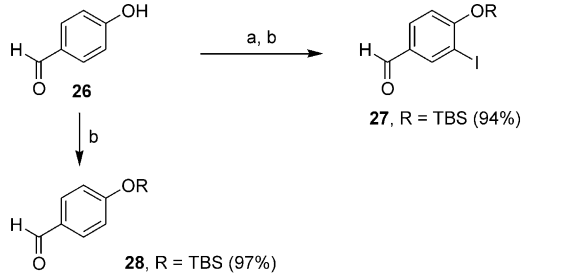
1, R = R^1 = H, X^1 = H, X^2 = H (85%)
16, R = R^1 = Me, X^1 = Br, X^2 = H (95%)
17, R = R^1 = Me, X^1 = I, X^2 = H (93%)
18, R = R^1 = Me, X^1 = H, X^2 = Br (95%)
19, R = R^1 = H, X^1 = Br, X^2 = H (64%)
20, R = R^1 = H, X^1 = H, X^2 = Br (94%)
21, R = R^1 = H, X^1 = H, X^2 = I (75%)
22, R = R^1 = H, X^1 = H, X^2 = CH_2CH_2F (79%)
23, R = Me, R^1 = H, X^1 = H, X^2 = CH_2CH_2F (64%)

Scheme 1. (a) PPh_3, THF, reflux, 10–20 h; (b) LDA, appropriate aldehyde derivatives, THF, 0 °C–rt, 30–60 min, 74–93%; (c) I_2, heptane, 12 h, reflux; (d) 1% HCl in MeOH, rt, 36 h, 64–95%.



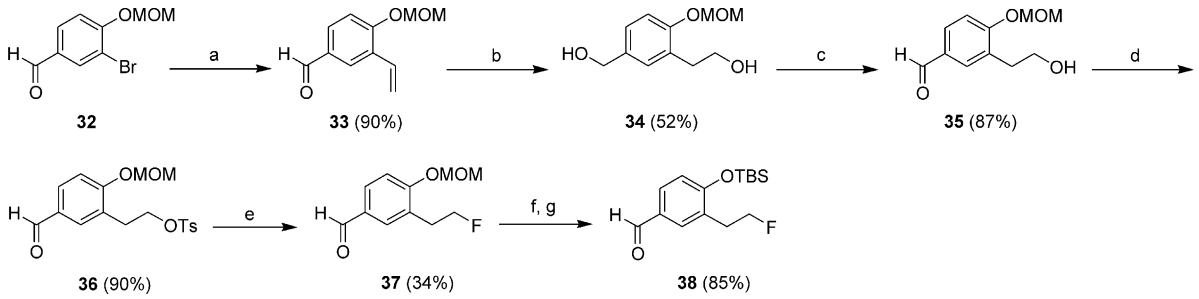
24
25 (91%)
5 (85%)
2, R = Me
5, R = TBS
3, R = Me, X^1 = Br (87%)
4, R = Me, X^1 = I (79%)
6, R = TBS, X^1 = Br (76%)

Scheme 2. (a) H_2SO_4, MeOH, 70 °C, 20 h; (b) TBS-Cl, imidazole, CH_2Cl_2, rt, 2 h; (c) $LiAlH_4$, THF, rt, 5 min; (d) PPh_3, CBr_4, THF, 0 °C, 9 h; (e) NBS[19] or NIS, CH_2Cl_2, rt, 1 h.



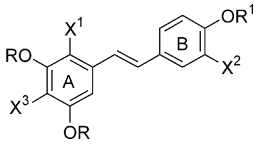
26
27, R = TBS (94%)
28, R = TBS (97%)
29
30
31, R = TBS (93%)
32, R = MOM (74%)

Scheme 3. (a) NIS,[20] CH_2Cl_2, 70 °C, 15 h; (b) TBS-Cl, imidazole, rt, 3 h; (c) Br_2, CH_2Cl_2, 60 °C, 12 h; (d) BBr_3, CH_2Cl_2, 0 °C–rt, 5 h; (e) MOM-Cl, K_2CO_3, acetone, 70 °C, 2 h.



Scheme 4. (a) Tributyl(vinyl)tin, $Pd(PPh_3)_4$, CuI, toluene, reflux, 16 h; (b) BH_3·THF then 4 N NaOH, H_2O_2, THF, 0 °C–rt, 18 h; (c) MnO_2, $CHCl_3$, 80 °C, 3 h; (d) NEt_3, TsCl, CH_2Cl_2, 0 °C, 8 h; (e) KF, [bmim][BF_4], 100 °C; (f) 50% AcOH, cat. H_2SO_4, 70 °C, 45 min; (g) TBS-Cl, imidazole, CH_2Cl_2, rt, 2 h.

Table 1. Radical scavenging activity of the resveratrol derivatives using DPPH assay and determination of total polyphenol



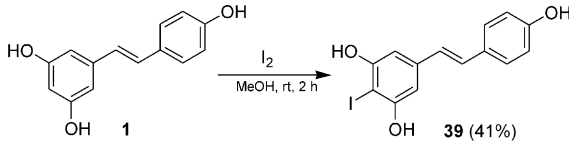
Compound	R	R^1	X^1	X^2	X^3	DPPH IC_{50} (μM)	DPPH relative IC_{50}[a]	Polyphenol (%)
1	H	H	H	H	H	123.3 (74.0)[b]	1	100
16	Me	Me	Br	H	H	—	—	26.6
17	Me	Me	I	H	H	—	—	—
18	Me	Me	H	Br	H	—	—	—
19	H	H	Br	H	H	35.9	3.43	85.8
20	H	H	H	Br	H	51.2	2.41	98.0
21	H	H	H	I	H	73.4	1.68	88.0
22	H	H	H	CH_2CH_2F	H	47.4	2.60	95.6
23	Me	H	H	CH_2CH_2F	H	69.5	1.77	50.4
39	H	H	H	H	I	70.6	1.75	72.0
Trans-Piceid	H, glycoside	H	H	H	H	(200)	0.37	
Astringin	H, glycoside	H	H	OH	H	(30.6)	2.42	
Astringinin	H	H	H	OH	H	(29.0)	2.55	
(+)-Catechin						(20.2)	3.66	
(−)-Epicatechin						(15.7)	4.71	

[a] DPPH assay: relative IC_{50} was expressed by IC_{50} of resveratrol **1**/IC_{50} of compound **19**–**23** and **39**.
[b] The values in parenthesis were from reference 11.

the absorbance of this solution was determined at 518 nm using a spectrophotometer and the remaining DPPH was calculated. All experiments were carried out in triplicate and repeated at least three times. Results are expressed as percentage decrease with respect to control values. Each fraction was evaluated at the final concentration of 100 μg/mL in the assay mixture.

3.2. Determination of total polyphenol

The reaction conditions used as the basis of comparison for most of these studies are the same as previous standard methods.[22] Samples of 50 μL were added to each 250 μL of diluted FCR (Folin–Ciocalteu reagent)[23] and preincubated at 37 °C for 1 min. Sodium bicarbonate (0.5 N, 750 μL) was added. The reaction solution was incubated at 37 °C for 24 h. The absorbance was measured at 725 nm. The control was prepared as above, with either H_2O or EtOH of the same volume of samples used instead of the sample solutions. Table 1 illustrates the free radical scavenging activity with a stable free radical, DPPH (1,1-diphenyl-2-picrylhydrazyl) as well as the amount of total polyphenol.



Scheme 5.

4. Discussion

Nine new resveratrol derivatives were designed and synthesized. The DPPH IC_{50} value of resveratrol we measured was 123.3 μM (lit. 74.0 μM[11]). Three methylated resveratrol derivatives **16**–**18** showed no free radical scavenging activity. However, when bromo, iodo and fluoroethyl groups were introduced to various positions, the DPPH IC_{50} of all compounds **19**–**23** and **39** in free phenol forms were improved as can be seen in the DPPH relative IC_{50} ranging from 1.75 to 3.43. The compound **19** with a bromo group at C2 position has the lowest IC_{50}, which corresponds to the highest free radical scavenging activity among the six free phenols,

while the compound **20** with a bromo group at C3′ position shows a slightly higher IC_{50} value but still has 2.41 times higher activity than resveratrol itself.

The main polyphenolic compounds in red wine are of two major classes: flavonoids and non-flavonoids, particularly stilbenes. Among flavonoids, it is known that (+)-catechin and (−)-epicatechin have high free radical scavenging activities and their DPPH relative IC_{50} are 3.66 and 4.71, respectively. On the other hand, non-flavonoids such as *trans*-piceid, astringin, and astringinin which are the derivatives of resveratrol have lower DPPH relative IC_{50} than catechin derivatives, 0.37, 2.42, and 2.55, respectively. Thus, bromo compounds **19**, **20** and fluoroethyl derivative **22** have quite high free radical scavenging activities. In 3′-substituted derivatives **20**, **21**, **22**, and astringinin, all substituted groups such as bromo, iodo, fluoroethyl and hydroxy help increase free radical scavenging activity.

Compound **23** with bismethyl-protected phenyl groups on A ring has only one free phenol group at C4′ position and shows only half of total polyphenol amount. But the free radical scavenging activity of **23** did not drop much compared to compound **22** that has three free phenol groups. Based on these results, it is expected that the introduction of a fluoroethyl group to C2 position on A ring would increase the free radical scavenging activity of the compound.

In conclusion, we designed and synthesized nine resveratrol derivatives as potential free radical scavengers. All compounds having free phenol group(s) showed good free radical scavenging activities. Among them, 2-bromoresveratrol **19** has similar free radical scavenging activity to (+)-catechin. As bromine-76, iodine-123, and fluorine-18 are very attractive radionuclides for the labeling of organic molecules as PET and SPECT radiotracers, the resveratrol derivatives labeled with these radionuclides would be beneficial for in vivo detection of free radical. Syntheses of more resveratrol derivatives are currently pursued, and further biological evaluation of resveratrol derivatives with good biological property are in progress.

Acknowledgements

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References and notes

1. (a) Eddarir, S.; Abdelhadi, Z.; Rolando, C. *Tetrahedron Lett*. **2001**, *42*, 9127. (b) Adesanya, S. A.; Nia, R.; Martin, M.-T.; Boukamcha, N.; Montagnac, A.; Païs, M. *J. Nat. Prod*. **1999**, *62*, 1694. (c) Miller, N. J.; Rice-Evans, C. A. *Clin. Chem*. **1998**, *41*, 1789. (d) Soleas, G. J.; Diamandis, E. P.; Goldberg, D. M. *Clin. Biochem*. **1997**, *30*, 91. (e) Goldberg, D.; Yan, J.; Ng, E. *Clin. Chem*. **1995**, *46*, 159. (f) Renaud, S.; De Lorgeril, M. *Lancet* **1992**, *339*, 1523.
2. For recent reviews on resveratrol of biological activities: (a) Aziz, M. H.; Kumar, R.; Ahmad, N. *Int. J. Oncol* **2003**, *23*, 17. (b) Wolter, F.; Stein, J. *Drug Future* **2002**, *27*, 949. (c) Wu, J. M.; Wang, Z. R.; Hsieh, T. C.; Bruder, J. L.; Zou, J. G.; Huang, Y. Z. *Int. J. Mol. Med*. **2001**, *8*, 3. (d) Frémont, L. *Life Sci*. **2000**, *66*, 663.
3. Creasy, L.; Coffee, M. *J. Am. Soc. Hortic. Sci*. **1988**, *113*, 230.
4. Kubo, M.; Kimura, Y.; Shin, H.; Haneda, H.; Tani, T.; Namba, K. *Soyayugaku Zasshi* **1981**, *35*, 58.
5. (a) Jang, M.; Cai, L.; Udeani, G. O.; Slowing, K. V.; Thomas, C. F.; Beecher, C. W. W.; Fong, H. H. S.; Farnsworth, N. R.; Kinghorn, A. D.; Mehta, R. G.; Moon, R. C.; Pezzuto, J. M. *Science* **1997**, *275*, 218. (b) Schneider, Y.; Vincent, F.; Duranton, B.; Badolo, L.; Gossé, F.; Bergmann, C.; Seiler, N.; Raul, F. *Cancer Lett*. **2000**, *158*, 85.
6. Docherty, J. J.; Fu, M. M. H.; Stiffler, B. S.; Limperos, R. J.; Pokabla, C. M.; DeLucia, A. L. *Antiviral Res*. **1999**, *43*, 145.
7. (a) Bowers, J. L.; Tyulmenkov, W.; Jernigan, S. C.; Klinge, C. M. *Endocrinology* **2000**, *141*, 3657. (b) Gehm, B. D.; McAndrews, J. M.; Chien, P.-Y.; Jameson, J. L. *Proc. Natl. Acad. Sci. U.S.A*. **1997**, *94*, 14138.
8. Wang, Z. R.; Huang, Y. Z.; Zou, J. C.; Cao, K. J.; Xu, Y. N.; Wu, J. M. *Int. J. Mol. Med*. **2002**, *9*, 77.
9. Babich, H.; Reisbaum, A. G.; Zuckerbraun, H. L. *Toxicol. Lett*. **2000**, *114*, 143.
10. (a) Frankel, E. N.; Waterhouse, A. L.; Kinsella, J. E. *Lancet* **1993**, *341*, 1103. (b) Frankel, E. N.; Waterhouse, A. L.; Teissedre, P. L. *J. Agric. Food Chem*. **1995**, *43*, 890.
11. Fauconneau, B.; Waffo-Teguo, P.; Huguet, F.; Barrier, L.; Decendit, A.; Merillon, J.-M. *Life Sci*. **1997**, *61*, 2103.
12. Vitrac, X.; Desmoulière, A.; Brouillaud, B.; Krisa, S.; Deffieux, G.; Barthe, N.; Rosenbaum, J.; Mérillon, J.-M. *Life Sci*. **2003**, *72*, 2219.
13. (a) Pettit, G. R.; Grealish, M. P.; Jung, J. K.; Hamel, E.; Pettit, R. K.; Chapuis, J.-C.; Schmidt, J. M. *J. Med. Chem*. **2002**, *45*, 2534. (b) Orsini, F.; Pelizzoni, F.; Verotta, L.; Aburjai, T. *J. Nat. Prod*. **1997**, *60*, 1082.
14. (a) Bolton, R. *J. Labelled Compd. Radiopharm*. **2002**, *45*, 485. (b) Lasne, M. C.; Perrio, C.; Rouden, J.; Barre, L.; Roeda, D.; Dolle, F.; Crouzel, C. *Top. Curr. Chem*. **2002**, *222*, 201.
15. (a) Rao, V. P.; Jen, A. K.-Y.; Wong, K. Y.; Drost, K. J. *Tetrahedron Lett*. **1993**, *34*, 1747. (c) Meier, H.; Dullweber, U. *Tetrahedron Lett*. **1996**, *37*, 1191. (d) Kim, S.; Ko, H.; Park, J. E.; Jung, S.; Lee, S. K.; Chun, Y.-J. *J. Med. Chem*. **2002**, *45*, 160. (e) Guiso, M.; Marra, C.; Farina, A. *Tetrahedron Lett*. **2002**, *43*, 597.
16. Zhang, J.-T.; Dai, W.; Harvey, R. G. *J. Org. Chem*. **1998**, *63*, 8125.
17. Lee, K. C.; Moon, B. S.; Lee, J. H.; Chung, K.-H.; Katzenellenbogen, J. A.; Chi, D. Y. *Bioorg. Med. Chem*. **2003**, *11*, 3649.
18. (a) Kim, D. W.; Song, C. E.; Chi, D. Y. *J. Am. Chem. Soc*. **2002**, *124*, 10278. (b) Kim, D. W.; Choe, Y. S.; Chi, D. Y. *Nucl. Med. Biol*. **2003**, *30*, 345. (c) Kim, D. W.; Song, C. E.; Chi, D. Y. *J. Org. Chem*. **2003**, *68*, 4281.
19. Lan, A. J. Y.; Heuckeroth, R. O.; Mariano, P. S. *J. Am. Chem. Soc*. **1987**, *109*, 2738.
20. Carreno, M. C.; Ruano, J. L. G.; Sanz, G.; Toledo, M. A.; Urbano, A. *Tetrahedron Lett*. **1996**, *37*, 4081.
21. Blois, M. S. *Nature* **1958**, *26*, 1199.
22. Amerine, M. A. *Laboratory Procedures for Enologists*. Department of Viticulture and Enology, University of California, Davis, 1960, 130 pp.
23. Diluted FCR (Folin-Ciocalteu reagent) was prepared by dilution of 1 mL of FCR with 9 mL of water.