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# Evaluation of the Role of Imidazolidinone Motif of Antineoplastic 4-Phenyl-1-arylsulfonylimidazolidinones Using 4-Phenyl-2-arylsulfonyloxazolines

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To evaluate the role of imidazolidinone moiety of potential anticancer 4-phenyl-1-arylsulfonylimidazolidinones **1** for their cytotoxicity, conformationally similar 4-phenyl-2-arylsulfonylaminooxazolines **2** were synthesized and compared their cytotoxicities with those of the corresponding **1**. Compounds **2** showed much reduced activity compared to N-arylsulfonylimidazolidinones **1**. This result might indicate that the imidazolidinone ring of **1** have the other roles for the activity as an essential structural motif in addition to conformational contribution.

**Key words:** Anticancer, 4-phenyl-1-arylsulfonylimidazolidinone, 4-phenyl-2-arylsulfonylaminooxazolines, Cytotoxicity

## INTRODUCTION

Arylsulfonylimidazolidinones **1** were reported (Jung, *et al.*, 1996; 1996; 1997; 1997) as analogs possessing broad spectrum of potent activity against the various human cancer cell lines. Previous structure activity relationship study of **1** indicated that two phenyl moieties at 4-position and sulfonyl group are essential for the activity (Jung, *et al.*, 1997) and the optimum distance between two phenyl groups as about 8.7Å between the centers of two phenyl rings (Jung, *et al.*, 2000; Park, *et al.*, 2000). Phenyl moiety itself at 4-position is considered to be optimum size for binding to its putative receptor (Jung and Kwak, 1997; Jung, *et al.*, 1996). However their activity were varied on the kind of substitutent on phenyl group on sulfonyl function of **1** (Jung, *et al.*, 1996). Quantitative structure activity relationship study revealed that the enlargement of the substituents at 4-position of this phenyl enhance the activity regardless of their electronic or hydrophobic parameters (Lee, *et al.*, 2000). Conformational relationship between two phenyl functions has been considered as major important factor for their activity. However, the roles of imidazolidinone motif for the activity have not been investigated, although the importance of imidazolidinone motif of **1** for their conformation is obvious. Replacement of imidazolidinone with conformationally similar five member rings might provide the opportunity to find out these roles. Coformationally similar oxazoline moiety was initially considered by model study. Accordingly, oxazolines **2** with overall conformational similarity were synthesized and compared their cytotoxicity to those of the corresponding analogs of **1**.

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## MATERIALS AND METHODS

Melting points (m.p.) were determined on Electrothermal 1A 9100 MK2 apparatus and are uncorrected. All commercial chemicals were used as obtained and all solvents

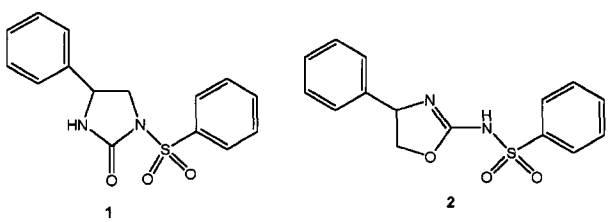



**Fig. 1.** Comparison of the optimized structure 1(thin line) overlaid on 2(thick line).

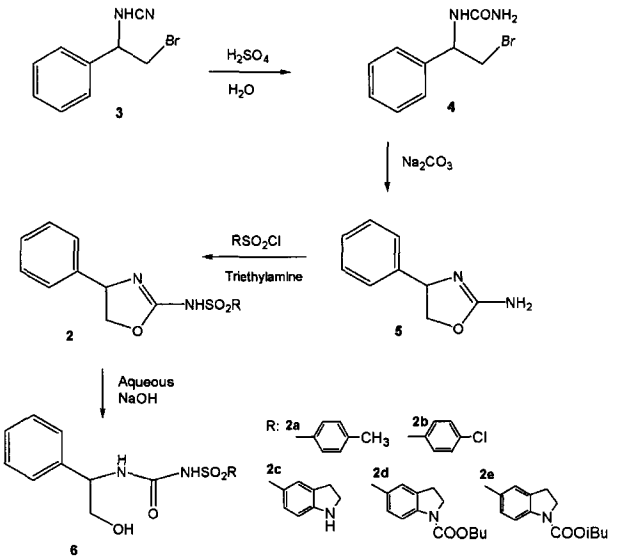
were purified by the standard procedures prior to use (Perrin, 1982). Thin-layer chromatography was performed on E Merck silica gel GF-254 precoated plates and the identification was done with UV light and colorization with spray 10% phosphomolybdic acid followed by heating. Flash column chromatography was performed with E. Merck silica gel (230-400mesh). IR spectra were recorded with Jasco IR-Report-100 IR spectrometer in $cm^{-1}$ and corrected against peak at 1601 $cm^{-1}$ of polystyrene. NMR spectra were measured in δ against the peak of tetramethylsilane by JEOL JNM-EX90 NMR (89.45 MHz) spectrometers. Elemental analysis was performed with EA1110 elemental analyzer (CE Instrument).

### Synthesis of 4 and 5

The solution of compound **3** (1 g, 4.44 mmol) in ether (1 mL) was cooled to 5°C and the 50% aqueous sulfuric acid (1 mL) was slowly added. The resulting mixture was stirred at room temperature for one hour. After dilution with water (30 mL) previously cooled at 5°C, the mixture was extracted with dichloromethane (30 mL) three times. The organic layers combined was washed with water two times, dehydrated with anhydrous sodium sulfate, and then evaporated under vacuum to give pure **4** (0.65 g, 50% yield). NMR ($CDCl_3$) δ 3.70 (d, J=7.2 Hz, 2H), 4.64 (br s, 2H), 5.14 (t, J=7.2 Hz, 1H), 5.53 (br s, 1H), 7.30 (m, 5H). Urea **4** was converted to oxazoline **5** during the storage within two days at room temperature. Therefore oxazoline **5** was obtained by one pot procedure from cyanamide **3**. Compound **3** (1.56 g, 6.93 mmol) was dissolved in ether (20 mL) and then 20% aqueouse sulfuric acid (20 mL) was slowly added on the ice-bath. The resulting mixture was stirred at room temperature for one hour and then basified with sodium carbonate. The mixture was stirred for one hour and then extracted with dichloromethane three times. The organic layers combined were dehyrated with anhydrous sodium sulfate and evaporated under vacuum to give pure **5** (0.67 g, 60% yield). m.p. 109-110°C; IR (KBr) 3415, 3215, 1680 $cm^{-1}$; NMR ($CDCl_3$) δ 4.08 (dd, J=7.2, 7.8 Hz, 1H), 4.64 (dd, J=7.8, 9.0 Hz, 1H), 5.12 (dd, J=7.2, 9.0 Hz, 1H), 5.48 (s, 2H, exchangeable with $D_2O$), 7.26 (m, 5H); Anal. Calcd for $C_9H_{10}N_2O$: C 66.65, H 6.21, N 17.27; Found C 66.58, H 6.17, N 17.12.

### General procedure for the preparation of 2

The corresponding arylsulfonyl chloride (1 equivalent) was added to the mixture of oxazoline **5**, two equivalents of triethylamine, and one equivalent of sodium carbonate in tetrahydrofuran-water (1:1). The resulting mixture was stirred at room temperature for two hours. Then the mixture was diluted with water and extracted with dichloromethane three times. The organic layers combined was dehydrated with anhydrous sodium sulfate and evaporated under vacuum. The residue was then separated by flash column chromatography to give compound **2**.




**Scheme 1.** Synthesis of 4-phenyl-2-arylsulfonyloxazolines

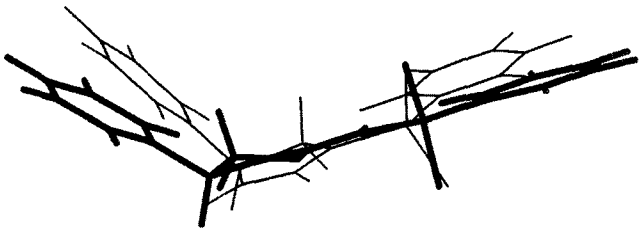


**Fig. 1.** Comparison of the optimized structure **1** (thin line) overlaid on 2 (thick line)

### 2-(4-methylbenzenesulfonylamino)-4-phenyloxazoline (2a)

#### *p*-Toluenesulfonyl chloride was used as arylsulfonyl chloride

Rf 0.45 (Hexane:EA=2:1) ; yield 30.2% ; m.p. 141.5 - 143.0°C; white solid ; IR (KBr) 1760, 1360, 1160 $cm^{-1}$ ; NMR($CDCl_3$) δ 2.37 (s, 3H), 4.27 (dd, J=3.3, 8.9 Hz, 1H), 4.72 (dd, J=8.6, 8.9 Hz, 1H), 5.44 (dd, J=3.3, 8.6 Hz, 1H), 7.06-7.48 (m, 9H) ; Anal. Calcd for $C_{16}H_{16}N_2O_3S$: C 60.74, H, 5.10, N, 8.85, S 10.14 ; Found C 60.58, H 5.04, N 8.54, S 10.21.

### 2-(4-Chlorobenzenesulfonylamino)-4-phenyloxazoline (2b)

#### 4-chlorobenzenesulfonyl chloride was used as arylsulfonyl chloride

Rf 0.53 (Hexane:Ethyl acetate=2:1); yield 28.6%; m.p. 122-124°C; white solid; IR (KBr) 1775, 1370, 1165 $cm^{-1}$ ; NMR($CDCl_3$) δ 4.32 (dd, J=3.4, 8.8 Hz, 1H), 4.75(dd, J =8.3, 8.8 Hz, 1H), 5.44(dd, J=3.4, 8.3 Hz, 1H), 7.13-7.51

(m, 9H); Anal. Calcd for $C_{15}H_{13}ClN_2O_3S$: C 53.49, H 3.89, N 8.32, S 9.52 ; Found C 53.38, H 3.80, N 8.28, S 9.31.

**2-(Indoline-5-sulfonylamino)-4-phenyloxazoline (2c)**

**N-Trifluoroacetylindoline-5-sulfonyl chloride was used as arylsulfonyl chloride. Upon basic work-up, deprotection of trifluoroacetyl group ocurred**

Rf 0.32 (Hexane:Ethyl acetate=2:1); yield 34.3%; m.p. 159 -161°C; white solid; IR (KBr) 1760, 1355, 1170 $cm^{-1}$; NMR ($CDCl_3$) δ 2.9 (t, J=8.4 Hz, 2H), 3.63 (t, J=8.1 Hz, 2H), 4.23 (dd, J=3.4, 8.5 Hz, 1H), 4.68 (dd, J=8.5, 8.6 Hz, 1H), 5.38(dd, J=3.4, 8.6 Hz, 1H), 6.33(d, 1H, exchangeable with $D_2O$), 7.04-7.39(m, 8H); Anal. Calcd for $C_{17}H_{17}N_3O_3S$: C 59.46, H 4.99, N 12.24, S 9.34; Found C 59.38, H 4.88, N 12.18, S 9.27.

**2-(1-Butoxycarbonylindoline-5-sulfonylamino)-4-phenyloxazoline (2d)**

**1-Butoxycarbonylindoline-5-sulfonyl chloride was used as arylsulfonyl chloride**

Rf 0.37 (Hexane:Ethyl acetate=2:1); yield 31.4%; m.p. 158-160°C; white solid; IR (KBr) 1770, 1350, 1180 $cm^{-1}$; NMR($CDCl_3$) δ 0.98 (t, J=6.5 Hz, 3H), 1.32-1.72 (m, 4 H), 3.02 (t, J=8.6 Hz, 2H), 3.96-4.36 (m, 5H), 4.11 (dd, J=3.4, 8.8 Hz, 1H), 4.74 (dd, J=8.4, 8.8 Hz, 1H), 5.43 (dd, J= 3.4, 8.4 Hz, 1H), 7.26-7.64 (m, 8H); Anal. Calcd for $C_{22}H_{25}N_3O_5S$: C 59.58, H 5.68, N 9.47, S, 7.23; Found C 59.41, H 5.58, N 9.36, S 7.16.

**2-(1-Isobutoxycarbonylindoline-5-sulfonylamino)-4-phenyloxazoline (2e)**

**1-Isobutoxycarbonylindoline-5-sulfonyl chloride was used as arylsulfonyl chloride**

Rf 0.37 (Hexane:Ethyl acetate=2:1); yield 12.56%; m.p. 159-160°C; white solid; IR (KBr) 1765, 1350, 1180 $cm^{-1}$; NMR ($CDCl_3$) δ 0.99 (d, J=7.0 Hz, 6H), 2.01 (m, 1H), 3.03 (t, J=8.2 Hz, 2H), 3,99-4.17 (m, 4H), 4.29 (dd, J=3.3, 8.8 Hz, 1H), 4.73 (dd, J=8.4, 8.8 Hz, 1H), 5.43 (dd, J=3.3, 8.4 Hz, 1H), 7.23-7.74 (m, 8H); Anal. Calcd for $C_{22}H_{25}N_3O_5S$: C 59.58, H 5.68, N 9.47, S, 7.23; Found C 59.44, H 5.60, N 9.42, S 7.11.

**Treatment of 2a and 2b with aqueous sodium hydro-xide**

Compounds **2a** (or **2b**) was treated in 10% sodium hydroxide in methanol-water (1:1) for two hours at room temperature. After dilution with water, the mixture was extracted with dichloromethane. The organic layer was dehydrated with anhydrous sodium sulfate and evaporated under vacuum to give **6a** (or **6b**) in 50% yield with starting material.

**N-(2-Hydroxy-1-phenyl)ethyl-N'-(4-methylphenylsulfonyl)urea (6a)**

NMR (acetone-$d_6$ + $D_2O$) δ 2.36 (s, 3H), 3.70 (d, J=5.5 Hz, 2H), 4.40 (m, 1H), 7.10-7.20 (m, 7H), 7.70 (d, J= 8.4 Hz), 2H).

**N-(2-Hydroxy-1-phenyl)ethyl-N'-(4-chlorophenylsulfonyl)urea (6b)**

NMR (acetone-$d_6$ + $D_2O$) δ 3.80 (d=6.7 Hz, 2H), 4.50 (m, 1H), 7.00-7.30 (m, 7H), 7.63 (d, J=8.0 Hz, 2H).

**Biological assay**

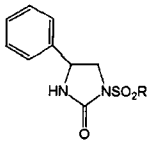
Cytotoxicities of compounds **1** (Jung, *et al.*, 1996, Lee, *et al.*, 2000) and **2** were measured against human ovary (SK-OV-3), leukemia (K 562), colon (Colo 205) cancer cell lines *in vitro* using MTT assay (Everitt *et al.*, 1987, Skehon *et al.*, 1990). The results from these tests are shown as $IC_{50}$ values in Table I.

## RESULTS AND DISCUSSION

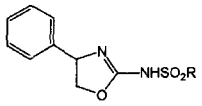
Oxazolines **2** were synthesized from bromocyanamide **3** (Jung and Kohn, 1985) as shown in scheme 1. Treatment of **3** with sulfuric acid at 5°C initially produced to urea **4**, which is subsequently undergo cyclization to hydrobromide salt of **5** during the storage at room temperature for two days as indicated in the literature (Jung and Kohn, 1985). Therefore cyanamide **3** was successfully converted to **5** in one pot procedure by sequential treatments with sulfuric acid for one hour and then aqueous sodium carbonate for two hours at room temperature with 60% yield. Oxazoline **5** was treated with the corresponding arylsulfonyl chloride in the presence of triethylamine and sodium carbonate to form compounds **2**. In this step, relatively low yields (about 30%) were observed. These results might be originated from the instability of **2** in the presence of acid or base. To confirm this property of **2**, compounds **2a** and **2b** were subjected to hydrolysis condition. Treatment of these compounds with aqueous sodium hydroxide at room temperature generated **6a** and **6b**, respectively. The high susceptibility of sulfonylisourea upon hydrolysis condition has been noticed in the hydrolysis of 4-phenyl-1-arylsulfonyl-2-alkoxyimidazolines to imidazolones **1** (Jung, *et al.*, 1996).

The cytotoxicities of compound **2** and the corresponding **1** were measured against human ovary (SK-OV-3), leukemia (K 562), and colon (Colo 205) cancer cell lines using MTT assay as shown in Table I. The activities of **2** are much weaker than those of **1** against all cell lines. The structural difference between **1** and **2** mainly comes from insertion of NH group between sulfonyl and oxazoline group in **2**. The conformational detail of the energy minimized structures of **1** and **2** using MM2 force field implemented in HyperChem (Hypercube, 1996) is illustrated in Fig. 1. Despite compound **2** shows about 2Å

**Table I.** Comparison of the cytotoxicity of compund 1 and 2



1



2

| Entry No. | comp. 1[a] | IC50 (μg/mL)[b] SK-OV-3 | K562 | Colo2 05 | comp. 2[a] | IC50 (μg/mL)[b] SK-OV-3 | K562 | Colo2 05 |
|---|---|---|---|---|---|---|---|---|
| 1 | a | 15.0 | 18.0 | 17.0 | a | >20 | >20 | >20 |
| 2 | b | 14.0 | 17.0 | 16.2 | b | >20 | >20 | >20 |
| 3 | c | 10.2 | 6.0 | 11.5 | c | >20 | >20 | >20 |
| 4 | d | 3.4 | 2.4 | 0.1 | d | 18.3 | 11.3 | >20 |
| 5 | e | 2.0 | 5.3 | 0.3 | e | 12.2 | 8.9 | >20 |

[a]Groups R correspond to those as shown in Scheme 1.
[b]IC50 values are the mean value of three times measurement. Cell lines used for the test are human ovary (SK-OV-3), leukemia (K562), colon (Colo205) cancer cell lines.

bigger in the overall length, compounds **1** and **2** reveal the overall similarity in their conformation as shown in Fig. 1. The phenyl ring on sulfonyl of both compounds are exactly coplanar with five member ring and the angle of 4-phenyl ring plane with respect to five member ring is 124° in **1** and 137° in **2**, respectively. This similarity might render the moderate activity to **2d** and **2e** against SK-OV-3 and K 562 cell lines. This might imply that two aromatic groups of **2** are major contributor to the activity. Therefore the the reduced activity of **2** may be an indicative that imidazolidinone ring of **1** should have the other roles for the activity as an essential structural motif in addition to conformational contribution.

## ACKNOWLEDGEMENTS

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## REFERENCES

Everitt, E., Wohlfart, C., Spectrometric quantitation of anchorage-dependent cell numbers extraction of naphthol blue-black-stained cellular protein. *Anal. Biochem.*, 162, 122-129 (1987).

Hypercube, HyperChem version 5.0, Hypercube, Inc. (1996).

Jung, S. -H., Song, J. -S., Lee, H. -S., Choi, S. -U., and Lee, C. -O., Synthesis and evaluation of cytotoxicity of novel arylsulfonylimidazolidinones containing sulfonylurea pharmacophore. *Arch. Pharm. Res.*, **19**, 570-580 (1996).

Jung, S. H. and Kohn, H., Stereoselective synthesis of vicinal diamines from alkenes and cyanamide. *J. Amer. Chem. Soc.* **107**, 2931-2943 (1985).

Jung, S. -H., Song, J. -S., Lee, H. -S., Choi, S. -U., and Lee, C. -O., Synthesis and evaluation of cytotoxic activity of novel arylsulfonylimidazolidinones. *Bioorg. & Med. Chem. Letters*, **6**, 2553-2558 (1996).

Jung, S. -H. and Kwak, S. -J., Planar structural requirement at 4-position of 1-arylsulfonyl-4-phenyl-4,5-dihydro-2-imidazolones for their cytotoxicity. *Arch. Pharm. Res.*, 20, 283-287 (1997).

Jung, S. -H., Lee, H. -S., Song, J. -S., Kim, H. -M., Han, S. -B., Lee, C. -W., Lee, M., Choi, D. -R., Lee, J. -A., Chung, Y. -H., Yoon, S. -J., Moon, E. -Y., Hwang, H.-S., Seong, S. -K., and Lee, D. -K., Synthesis and antitumor activity of 4-phenyl-1-arylsulfonylimidazolidinones. *Bioorg. & Med. Chem. Letters*, 8, 2553-2558 (1997).

Lee, H. -S., Park, K. -L., Choi, S. -U., Lee, C. -O., and Jung, S. -H., Effect of substituents on benzenesulfonyl motif of 4-Phenyl-1-arylsulfonylimidazolidinones for their cytotoxicity. *Arch. Pharm. Res.*, 23, 579-584 (2000).

Park, K. -L., Moon, B. -G., Jung, S. -H, Kim, J. -G., and Suh, I. -W., Tautomeric evidence in an arylsulfonylimidazolone hydrochloride. *Acta Cryst.*, C56, 1247-1250 (2000).

Skehan, P., Storeng, R., Scudiero, D. A., Monks, A., MacMahon, J., Vista, D. T., Kenny, S., and Boyd, M. R. New colorimetric cytotoxicity assay for anticancer drug screening. *J. Natl. Cancer Inst.*, 82, 1107-1112 (1990).