Communications to the Editor

Chem. Pharm. Bull. 31(6)2164—2167(1983)

SYNTHESIS AND BIOLOGICAL ACTIVITIES OF 5-SUBSTITUTED 6-PHENYLTHIO AND 6-IODOURIDINES, A NEW CLASS OF ANTILEUKEMIC NUCLEOSIDES

Hiromichi Tanaka, Akira Matsuda, Shuji Iijima, Hiroyuki Hayakawa, and Tadashi Miyasaka*
School of Pharmaceutical Sciences, Showa University, Hatanodai 1-5-8, Shinagawa-ku, Tokyo 142, Japan

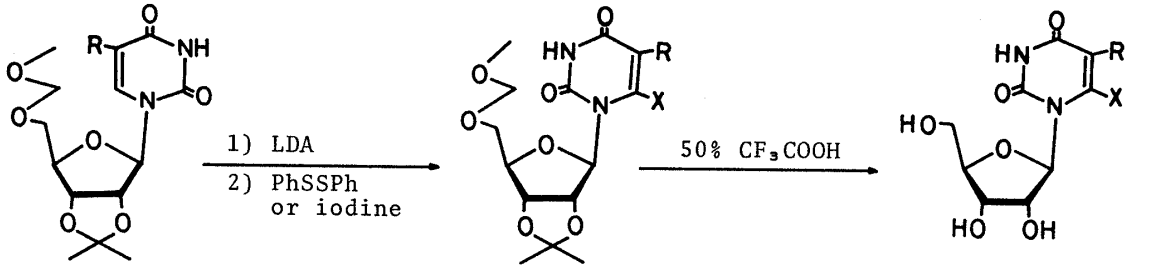
A new class of 5,6-disubstituted uridines, in which the C-6 position was occupied by phenylthio group or iodine, were synthesized *via* lithiation of the corresponding 5-substituted 2',3'-*O*-isopropylidene-5'-*O*-methoxymethyluridines and subsequent electrophilic reactions. These newly-synthesized uridine derivatives exhibited antileukemic activities against mouse leukemia L5178Y cells in culture.

KEYWORDS—— lithiation; LDA; 6-phenylthiouridine derivative; 6-iodouridine derivative; 5,6-disubstituted uridine derivative; mouse leukemia L5178Y; antileukemic activity

Recently, we reported the synthesis of various types of 6-substituted uridines based on the regiospecific lithiation of 2',3'-*O*-isopropylidene-5'-*O*-methoxymethyluridine (**1**).[1] Among these 6-substituted uridines, 6-phenylthiouridine (**2**) exhibited weak activity against mouse leukemia L5178Y cells in culture (ED_{50} 70 μg/ml). On the other hand, we found that **2** and its 5'-phosphate were highly susceptible to nucleophilic addition-elimination reactions with sulfur nucleophiles even under very mild conditions.[2] Given **2**'s chemical and biological behaviors, one would predict that uridine derivatives bearing a leaving group at the C-6 position would react irreversibly with biological nucleophiles, and thus would manifest cytotoxic activity. In fact, the result of assay of 6-iodouridine[3] (**3**: ED_{50} 8 μg/ml) was promising for further investigations. In this communication, as an extension of our preliminary study, synthesis and biological evaluation of some 5-substituted 6-phenylthio and 6-iodouridines are described.

Examination of the literature revealed that the classical condensation method was unsuccessful in synthesizing such a type of nucleoside, presumably due to the steric hindrance imposed by the C-6 substituent.[4] The only exception is the preparation of 6-fluorothymine glycosides.[5] Thus, in the synthesis reported here, we utilized our recent methodology[1] to overcome this problem.

5-Substituted 2',3'-*O*-isopropylidene-5'-*O*-methoxymethyluridine (**4a~e**)[6] were prepared in high yields by the acetal exchange of dimethoxymethane with the corresponding 2',3'-*O*-isopropylidene derivatives in the presence of acetone and a catalytic amount of methanesulfonic acid. Lithiation of **4** was carried out with 2.5 eq of LDA in THF below -70°C to give a clear solution of the dilithio derivative.



1 (R= H)
4a∿e

5a∿e
5a'∿e'

2 (R= H, X= SPh)
3 (R= H, X= I)
6a∿e
6a'∿e'

a: R= F (X= SPh) a': R= F, X= I
b: R= Cl (X= SPh) b': R= Cl, X= I
c: R= Br (X= SPh) c': R= Br, X= I
d: R= I (X= SPh) d': R= I, X= I
e: R= Me (X= SPh) e': R= Me, X= I

Table I

	yield(%) of 5		yield(%) of 6
5a	100	6a	85
5a'	92	6a'	86
5b	91	6b	79
5b'	94	6b'	81
5c	97	6c	83
5c'	87	6c'	51
5d	85	6d	76
5d'	88	6d'	46
5e	38	6e	99
5e'	28	6e'	100

Yields were not optimized, and those of 6c' and 6d' can certainly be improved.

Table II

	ED_{50} (μg/ml) against L5178Y
6a	30
6a'	0.02
6b	8
6b'	55
6c	20
6c'	52
6d	40
6d'	55
6e	60
6e'	4
2	70
3	8
FUR	0.01
FU	0.1

Subsequent addition of diphenyl disulfide or iodine (2.0 eq) to the anion solution furnished the 6-phenylthio (5a∿e) or 6-iodo (5a'∿e') derivatives after quenching with AcOH, followed by chromatographic purification on a silica gel column. In the cases of 5e and 5e', where the electron-donating effect of the methyl group can participate in decreasing the acidity of H-6, the extent of metalation through an "acid-base mechanism"[7] seems to be reduced, as shown in Table I. The structures of 5a∿e[8] and 5a'∿e'[9] were confirmed by MS and PMR spectra. The by-products derived from aryne formation, nucleophilic attack by the lithiating agent, or halogen-lithium exchange were hardly detected during the lithiation and subsequent electrophilic reactions. As pointed out by Cushley and his co-workers,[10] the anomeric proton of the 5-fluorouridine derivative 4a appeared as a double doublet due to a long-range coupling between it and the fluorine atom. In contrast to 4a, 6-substituted 5-fluorouridine analogues 5a and 5a' were void of such an extra splitting in the anomeric signals. This observation could be attributed to the glycosidic "syn-conformation" of these compounds vs. "anti-conformation" of 4a. That is, while H-1' of 4a lies in a favorable geometrical arrangement (zig-zag pattern), those of 5a and 5a' do not.

Concurrent deprotection of the isopropylidene and methoxymethyl groups in 5a∿e 5a'∿e' was successfully accomplished with 50% aqueous trifluoroacetic acid at room temperature to give the requisite free nucleosides 6a∿e and 6a'∿e'.[11]

The newly-synthesized compounds were tested for their cytotoxic effectiveness against mouse leukemia L5178Y cells in culture, and the results are shown in Table II in terms of their ED_{50} (μg/ml). 5-Fluorouracil (FU) and 5-fluorouridine (FUR) are also included for comparison. As we intended, all the compounds are found to be more or less toxic to mouse leukemia cells. Above all, 5-fluoro-6-iodouridine (6a') is five times more potent than 5-fluorouracil and two times less potent than 5-fluorouridine. Since 6-iodouridine (3) itself showed significant effectiveness, it seems likely that the mechanism of action of 6a' is quite different from those of 5-fluorouracil and 5-fluorouridine. Thus, although no well-defined relationships between structures and activities have been obtained at present, it must be emphasized that our studies have disclosed a new class of antileukemic substances in the nucleoside field. Further work is in progress to elucidate the mechanism of action of the compounds involved in the present investigation.

ACKNOWLEDGEMENT We would like to express our gratitude to Dr. Kenjiro Kodama, Research Laboratories, Yamasa Shoyu Co. Ltd., for performing the cytotoxic studies.

REFERENCES AND NOTES

1) H. Tanaka, H. Hayakawa, and T. Miyasaka, Chem. Pharm. Bull., 29, 3565 (1981); idem, Nucleic Acids Symposium Series, 10, 1 (1981); idem, Tetrahedron, 38, 2635 (1982).

2) H. Tanaka, S. Iijima, A. Matsuda, H. Hayakawa, T. Miyasaka, and T. Ueda, Chem. Pharm. Bull., 31, 1222 (1983).

3) Compound 3 was obtained as crystals (mp 130∿132°C) from 1 by the same reaction sequences as the preparation of 2 and gave physical data consistent with its structure.

4) L. Pichat and G. Chatelain, Bull. Chim. Soc. Fr., 1970, 1833.

5) D. Barwolff, G. Kowollik, and P. Langen, Collect. Czech. Chem. Commun., 39, 1494 (1974); M. von Janta-Lipinski and P. Langen, Nucleic Acids Symposium Series, 9, 41 (1981).

6) Compounds 4a∿e were obtained as crystals and gave physical data consistent with their structures. Melting points of 4a∿e: 4a mp 126∿127°C; 4b mp 109∿111°C; 4c mp 134∿135°C; 4d mp 146∿148°C; 4e mp 109∿110°C.

7) H. W. Gschwend and H. R. Rodriguez, "Organic Reactions," Vol. 26, ed. by W. G. Dauben, John Wiley and Sons, Inc., New York, Chichester, Brisbane, and Toronto, 1979, pp. 1∿360.

8) Compounds 5a∿e were obtained as syrups and gave physical data consistent with their structures.

9) Compounds 5a'∿e' were obtained as crystals and gave physical data consistent with their structures. Melting points of 5a'∿e': 5a' mp 204∿205°C; 5b' mp 189∿190°C; 5c' mp 198∿199°C; 5d' mp 167.5∿169.5°C; 5e' mp 197.5∿198.5°C.

10) R. J. Cushley, I. Wempen, and J. J. Fox, J. Am. Chem. Soc., 90, 709 (1968).

11) Compounds 6a∿e and 6a'∿e' were obtained as crystals and gave physical data consistent with their structures. Melting points of 6a∿e and 6a'∿e': 6a mp 88∿95°C; 6b mp >300°C; 6c mp 255∿260°C (dec.); 6d mp 104∿107°C; 6e mp 162.5∿164.5°C; 6a' mp 163∿166°C (resolidified at 170°C, decomp. at 208°C); 6b' mp 160∿162°C (resolidified at 165°C, decomp. at 255°C); 6c' mp 164∿166°C (resolidified at 168°C, decomp. at 175°C); 6d' mp 144∿146°C (resolidified at 150°C, decomp. at 180°C); 6e' mp 133∿136°C.

(Received May 16, 1983)