

## SOME HYBRID LINKER MODE $C_2$ -SYMMETRICAL 1,3,5-TRIAZINE DERIVATIVES AND THEIR BIOLOGICAL EVALUATION

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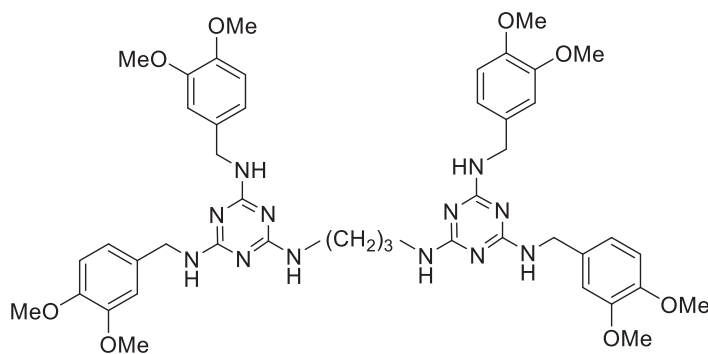
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**Abstract** – We report the preparation of some new  $C_2$ -symmetrical multivalent hybrid-type molecules having a methylene linker group and 1,3,5-triazine (TAZ) recognition moieties in the molecule and we also report the results of biological evaluation of their anti-herpes simplex virus type 1 (anti-HSV-1) activity and cytotoxic activity against Vero cells. All mid-size  $C_2$ -symmetrical multivalent hybrid-type molecules (**3a-2**, **3a-4**, **3b-2**, **3b-3**, **3b-4**) showed considerably high levels of anti-HSV-1 activity ( $EC_{50} = 7.6 \sim 95.6 \mu\text{M}$ ) with low levels of cytotoxicity ( $CC_{50} > 200 \mu\text{M}$ ) against Vero cells. Among the tested compounds, the hybrid-type  $C_2$ -symmetrical multivalent molecule (**3a-4**) seems to be an interesting new lead in the search for new hybrid-type multivalent mid-size antiviral compounds.

Carbohydrate-containing glycoproteins, proteoglycans and glycolipids on the cell surface interact with various cell growth factors to control cell activity for the processes of bacterial or viral infection and pathological phenomena such as metastasis of cancer cells.<sup>1-3</sup> Moreover, supramolecular interaction by macromolecules with two-fold ( $C_2$ ) or three-fold ( $C_3$ ) geometry is one of the common interactions in many (crucial) biological responses.<sup>4</sup> To develop new oligovalent symmetrical bioactive molecules or leads, we have recently designed and synthesized a few new compounds with such geometry and evaluated their bioactivities in order to find new types of bioactive leads.<sup>5-15</sup> In connection with our

projects, we have recently reported the preparation of a few new hybrid-type  $C_2$ -symmetrical trivalent 1,3,5-triazine (TAZ) derivatives and the results of biological evaluation of the synthesized mid-size symmetrical TAZ derivatives.<sup>15</sup> Among previously targeted hybrid-type mid-size  $C_3$ - and  $C_2$ -symmetrical molecules, we found that a hybrid-type  $C_2$ -symmetrical tri-substituted TAZ derivative (**A**) showed a considerably high level of anti-HSV-1 activity ( $EC_{50} = 19.1 \mu\text{M}$ ) and a low levels of cytotoxic activity ( $CC_{50} > 200 \mu\text{M}$ ) against Vero cells,<sup>15</sup> and this non-peptide synthetic mid-size TAZ derivative is considered to be a potential new lead in the search for multivalent antiviral active molecules (Figure 1). The carbohydrate recognition property of  $C_3$ -symmetrical TAZ derivatives<sup>13</sup> with high levels of anti-HSV-1 activity also prompted us to investigate this new unique symmetrical multivalent TAZ derivative **A** and related compounds to find a more promising new antiviral lead.

For further synthesis of oligovalent target hybrid-type TAZ derivatives, we adopted a linear methylene linker of symmetrical hybrid-type compound **A**, and we also referred to the benzodioxole substituent for the structure of the TAZ ring moiety that generally showed high levels of anti-HSV-1 activity in the  $C_3$  geometric TAZ derivatives.<sup>15</sup> We synthesized a few such multivalent hybrid-type  $C_2$ -symmetrical TAZ derivatives having methylene linker groups. In this paper, we also describe the results of evaluation of their biological activities and the structure-activity relationships (SARs) of these unique mid-size multivalent symmetrical TAZ-related derivatives.



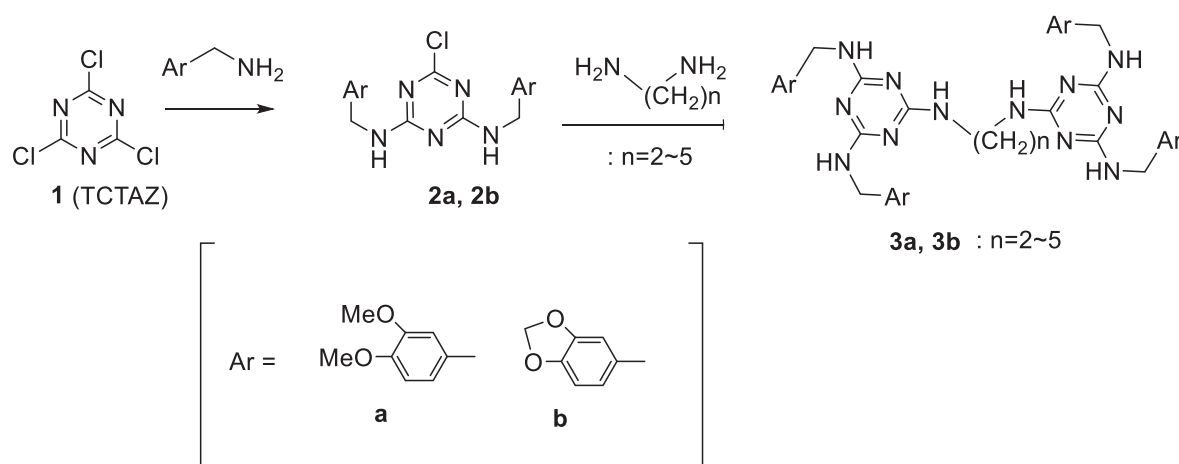
**Figure 1.** Anti-HSV-1 active hybrid-type  $C_2$ -symmetrical TAZ derivative **A**

## RESULTS AND DISCUSSION

For the preparation of various new target multivalent hybrid-type TAZ mid-size molecules (**3**), we used a conventional synthetic route consisting of stepwise coupling of intermediate TAZ derivatives (**2**) with various diaminoalkanes (**1**  $\rightarrow$  **2**  $\rightarrow$  **3**, see Scheme 1). The structures of the obtained new hybrid-type  $C_2$ -symmetrical multivalent TAZ derivatives (**3**) were established by spectroscopic methods and

elemental analysis. Correct molecular ion peaks were observed in high-resolution positive FAB-MS spectra of all  $C_2$ -symmetrical TAZ derivatives (**3**). The details of the stepwise preparation of these symmetrical TAZ derivatives (**3**) are given in EXPERIMENTAL. The structures of target hybrid-type  $C_2$ -symmetrical multivalent TAZ derivatives obtained in this study are shown in Table 1. As can be seen in the Table, the yields obtained by this procedure with the starting 2,4,6-trichloro-1,3,5-triazine (TCTAZ) (**1**) to target new hybrid-type  $C_2$ -symmetrical trivalent 1,3,5-triazine (TAZ) derivatives (**3a-2**, **3a-4**, **3b-2**, **3b-3**, **3b-4**) were good, and this method for synthesis of hybrid-type TAZ derivatives was reproducible and was reconfirmed to be useful as a general procedure for synthesis of hybrid-type  $C_2$ -symmetrical multivalent TAZ derivatives.<sup>12</sup>

The results of biological evaluation of these hybrid-type derivatives [anti-HSV-1 activity ( $EC_{50}$ ) and cytotoxicity ( $CC_{50}$ )] together with two previously reported compounds (**3a-3**:  $n=3$  and **3a-5**:  $n=5$ ) and the data for aciclovir<sup>17</sup> are also shown in Table 1. All of the new linear methylene linker mode multivalent  $C_2$ -symmetrical TAZ derivatives **3** (**3a-2**, **3a-4**, **3b-2**, **3b-3**, **3b-4**) showed varying degrees of anti-HSV-1 activity ( $EC_{50} = 7.6 \sim 95.6 \mu\text{M}$ ) with low levels of cytotoxicity ( $CC_{50} > 200 \mu\text{M}$ , see Table 1).



**Scheme 1.** Synthesis of  $C_2$ -symmetrical multivalent hybrid-type TAZ derivatives (**3a**, **3b**)

It is noteworthy that all of the  $C_2$ -type hybrid-type multivalent compounds (**3a** and **3b**) having a linear methylene linker in the molecule showed considerably high levels of anti-HSV-1 activity (see Table 1). Among the tested hybrid-type  $C_2$ -symmetrical multivalent TAZ derivatives (**3a-2~3a-5**), the  $C_2$ -symmetrical derivative (**3a-4**) having 3,4-dimethoxyphenyl groups showed the highest anti-HSV-1 activity ( $EC_{50} = 7.6 \mu\text{M}$ ) and low cytotoxicity ( $CC_{50} > 200 \mu\text{M}$ ). In contrast, all of the  $C_2$ -symmetrical multivalent TAZ derivatives (**3b-2~3b-4**) having a 2,4-methylenedioxy functionality in four aryl groups linked to the TAZ rings showed modest anti-HSV-1 activity ( $EC_{50} = 36.8 \sim 54.4 \mu\text{M}$ ); however, the most active one was a multivalent compound (**3b-2**,  $EC_{50} = 36.8 \mu\text{M}$ ) having a short linear  $C_2$ -methylene linker

in the molecule. From the above-described results, we consider that the methylene linker length in the molecule may be one of the important factors, in addition to the nature of functional groups on aryl rings, for the expression of anti-HSV-1 activity of these multivalent hybrid-type molecules. The obtained results suggest that the hybrid-type  $C_2$ -symmetrical multivalent molecule (**3a-4**) is a useful lead in the search for new multivalent mid-size antiviral TAZ derivatives.

On the basis of the information obtained by evaluation of the biological activities of the new  $C_2$ -symmetrical hybrid-type multivalent mid-size series together with recent information on the  $C_3$ -type TAZ series,<sup>12,13</sup> we are now investigating chemical properties of anti-HSV-1-active TAZ derivatives (**3a-4**) such as sugar recognition with the aim of developing new non-peptide multivalent mid-size lead compounds with anti-HSV-1 activity.

**Table 1.** Anti-HSV-1 activity ( $EC_{50}$ ) and cytotoxicity ( $CC_{50}$ ) against Vero cells of target hybrid-type TAZ derivatives (**3**)

Compound	n	Structure	$EC_{50}$ ( $\mu$ M)	$CC_{50}$ ( $\mu$ M)
<b>3a-2</b>	2	<p style="text-align: center;"><b>3a</b></p>	95.6	>200
<b>3a-3<sup>a)</sup></b>	3		19.1	>200
<b>3a-4</b>	4		7.6	>200
<b>3a-5<sup>a)</sup></b>	5		>100	>200
<b>3b-2</b>	2		<p style="text-align: center;"><b>3b</b></p>	36.8
<b>3b-3</b>	3	54.4		>200
<b>3b-4</b>	4	38.5		>200
aciclovir <sup>b)</sup>				1.1

a) Data were taken from reference 15. b) Data were taken from reference 17.

## EXPERIENTIAL

Melting points were determined using a micro melting point apparatus (Yanaco MP-S3) without correction. IR spectra were measured by a Shimadzu FTIR-8100 IR spectrophotometer. Low- and high-resolution mass spectra (LR-MS and HR-MS) were obtained by a JEOL JMS HX-110 double-focusing model equipped with an FAB ion source interfaced with a JEOL JMA-DA 7000 data system. <sup>1</sup>H- and <sup>13</sup>C-NMR spectra were obtained by ECG600R. Chemical shifts were expressed in δ ppm downfield from an internal TMS signal for <sup>1</sup>H-NMR and the carbon signal of the corresponding solvent [CDCl<sub>3</sub> (77.00 ppm), DMSO-*d*<sub>6</sub> (39.50 ppm)] for <sup>13</sup>C-NMR. The signal assignments were confirmed by two-dimensional (2D)-NMR analyses: <sup>1</sup>H-<sup>1</sup>H 2D correlation spectroscopy (COSY), <sup>1</sup>H-<sup>13</sup>C heteronuclear multiple-quantum coherence (HMQC), and <sup>1</sup>H-<sup>13</sup>C heteronuclear multiple-bond connectivity (HMBC). Microanalyses were performed with a Yanaco MT-6 CHN corder. Routine monitoring of reactions was carried out using precoated Kieselgel 60F<sub>254</sub> plates (E. Merck). Detection of products was accomplished with UV light and iodine. Microwave irradiation experiments were carried out in a CEM Discover Focused Microwave System. Flash column and centrifugal chromatography separations of the reaction products were performed on silica gel (Kanto 60N or Able-Biott) with a UV detector. Commercially available starting materials were used without further purification, and dry solvents were used in all reactions. Intermediate di-benzylamino-substituted TAZ derivatives (**2a** and **2b**) were prepared from TCTAZ (**1**) and the corresponding benzyamines by a previously reported procedure.<sup>12</sup>

### Procedure for C<sub>2</sub>-Symmetrical Multivalent Hybrid-type TAZ Derivatives (Table 1)

#### Synthesis of *N*<sup>2</sup>,*N*<sup>2'</sup>-(Ethane-1,2-diyl)bis[(*N*<sup>4</sup>,*N*<sup>6</sup>-bis(3,4-dimethoxybenzyl)-1,3,5-triazine-2,4,6-triamine) (**3a-2**)

To a suspension of intermediate **2a** (1.47 g, 3.30 mmol) in anhydrous dioxane (10 mL) were added ethylenediamine (94.4 mg, 1.57 mmol) and K<sub>2</sub>CO<sub>3</sub> (1.30 g, 9.43 mmol) at room temperature with stirring. The reaction mixture was irradiated with a 100 W microwave (MW) at 100 °C for 5 h with stirring. After addition of 1M HCl (10 mL), the resulting mixture was extracted with CH<sub>2</sub>Cl<sub>2</sub> (20 mL×3). The combined organic layer was washed with brine and dried over MgSO<sub>4</sub>. After evaporation of the solvent, the residual yellow oil was purified by centrifugal chromatography (CH<sub>2</sub>Cl<sub>2</sub> : EtOH = 98 : 2 → 93 : 7) to give compound **3a-2** (383 mg, 0.436 mmol, 28%) as a white solid. Mp 89-92 °C. IR (KBr) cm<sup>-1</sup>: 3404 (NH), 1574, 1514 (C=N), 1263, 1234, 1138, 1027 (C-N and/or C-O). <sup>1</sup>H-NMR (CDCl<sub>3</sub>) δ: 3.49 (4H, br s, H1''), 2''), 3.80, 3.82 (24H, OCH<sub>3</sub>), 4.46 (8H, br s, Hα), 5.37, 5.78 (6H br s, NH), 6.7-6.9 (12H, m, ArH). <sup>13</sup>C-NMR (CDCl<sub>3</sub>) δ: 41.29 (C1'' 2''), 44.36, 44.46\* (Cα), 55.76, 55.83 (OCH<sub>3</sub>), 110.85 (C5'), 111.02 (C2'), 119.69 (C6'), 131.78 (C1'), 148.11 (C4'), 148.94 (C3'), 166.01 (C2, 4), 166.42 (C6). (The observed <sup>13</sup>C-signals assignable to the predominant tautomer are asterisked.) Positive-ion FAB-MS *m/z*:

879 (M+H)<sup>+</sup>. HR-FAB-MS *m/z*: 879.4260 (Calcd for C<sub>44</sub>H<sub>55</sub>N<sub>12</sub>O<sub>8</sub>: 879.4266). Anal. Calcd for C<sub>44</sub>H<sub>54</sub>N<sub>12</sub>O<sub>8</sub>·0.8H<sub>2</sub>O: C, 59.15; H, 6.27; N, 18.81. Found: C, 59.19; H, 6.46; N, 18.57.

**Synthesis of N<sup>2</sup>,N<sup>2'</sup>-(Butane-1,4-diyl)bis[(N<sup>4</sup>,N<sup>6</sup>-bis(3,4-dimethoxybenzyl)-1,3,5-triazine-2,4,6-triamine) (3a-4)**

To a suspension of intermediate **2a** (892 mg, 2.00 mmol) in DMF (2.0 mL) were added 1,4-diaminobutane (53.4 mg, 0.606 mmol) and K<sub>2</sub>CO<sub>3</sub> (553 mg, 4.00 mmol) at room temperature with stirring. The reaction mixture was refluxed in a water bath for 10 h. After addition of water (20 mL), the resulting mixture was extracted with CH<sub>2</sub>Cl<sub>2</sub> (20 mL×3). The combined organic layer was washed with brine (20 mL) and dried over MgSO<sub>4</sub>. After evaporation of the solvent, the residual yellow oil was purified by centrifugal chromatography (CH<sub>2</sub>Cl<sub>2</sub> : EtOH = 96 : 4 → 93 : 7 ) to give compound **3a-4** (320 mg, 0.353 mmol, 58%) as a white solid. Mp 88-90 °C. IR (KBr) cm<sup>-1</sup>: 3406 (NH), 1566, 1513 (C=N), 1263, 1234, 1138, 1027 (C-N and/or C-O). <sup>1</sup>H-NMR (CDCl<sub>3</sub>) δ: 1.61 (4H, br s, H2'', 3''), 2.20 (1.3H, br s, NH), 3.42 (4H, br s, H1'', 4''), 3.80, 3.84, 3.85 (24H, br s, OCH<sub>3</sub>), 4.25, 4.39, 4.51 (8H, br s, Hα), 5.00, 5.28 (3.3H, br s, NH), 6.6-6.9 (12H, m, ArH), *ca.* 6.80(1.1H, br s, NH), 7.80 (0.3H, br s, NH). <sup>13</sup>C-NMR (CDCl<sub>3</sub>) δ: 23.58 (C2'', 3''), 40.65, 41.75 (C1'', 4''), 44.25 (Cα), 55.72, 55.82 (OCH<sub>3</sub>), 110.67 (C5'), 110.95 (C2'), 119.48 (C6'), 132.13 (C1'), 148.02 (C4'), 148.92 (C3'), 166.12 (C2, 4, 6). Positive-ion FAB-MS *m/z*: 907 (M+H)<sup>+</sup>. HR-FAB-MS *m/z*: 907.4577 (Calcd for C<sub>46</sub>H<sub>59</sub>N<sub>12</sub>O<sub>8</sub>: 907.4579). Anal. Calcd for C<sub>46</sub>H<sub>58</sub>N<sub>12</sub>O<sub>8</sub>·0.6H<sub>2</sub>O: C, 60.20; H, 6.50; N, 18.31. Found: C, 60.10; H, 6.49; N, 18.31.

**Synthesis of N<sup>2</sup>,N<sup>2'</sup>-(Ethane-1,2-diyl)bis[(N<sup>4</sup>,N<sup>6</sup>-bis(benzo[*d*][1,3]dioxol-5-ylmethyl)-1,3,5-triazine-2,4,6-triamine] (3b-2)**

To a suspension of intermediate **2b** (892 mg, 2.00 mmol) in DMF (2.5 mL) were added ethylenediamine (146 mg, 0.700 mmol) and K<sub>2</sub>CO<sub>3</sub> (553 mg, 4.00 mmol) at room temperature with stirring. The reaction mixture was refluxed in a water bath for 10 h. After addition of water (20 mL), the resulting mixture was extracted with CH<sub>2</sub>Cl<sub>2</sub> (20 mL×3). The combined organic layer was washed with brine (20 mL) and dried over MgSO<sub>4</sub>. After evaporation of the solvent, the residual oil was purified by centrifugal chromatography (CH<sub>2</sub>Cl<sub>2</sub> : EtOH = 95 : 5 ) to give compound **3b-2** (486 mg, 0.596 mmol, 85%) as a white solid. Mp 187-191 °C (recrystallized from EtOH). IR (KBr) cm<sup>-1</sup>: 3458, 3424, 3273 (NH), 1584, 1525, 1498 (C=N), 1242, 1038 (C-N and/or C-O). <sup>1</sup>H-NMR (DMSO-*d*<sub>6</sub>) δ: 3.30 (4H, br s, H1'', 2''), 4.29 (8H, br s, Hα), 5.94 (8H, br s, H2'), 6.39, 6.43 (1.6H, br s, NH), 6.65-6.9 (12H, m, ArH), 6.73, 6.84 (1.1H, br s, NH), 7.00, 7.04, 7.17 (3.3H, br s, NH). <sup>13</sup>C-NMR (DMSO-*d*<sub>6</sub>) δ: 40.10 (C1'', 2''), 42.80\*, 43.07 (br s, Cα), 100.61 (C2'), 107.82 (br s, C4', 7'), 120.01, 120.23\* (br s, C6'), 134.84 (br s, C5'), 145.68 (C3a' or 7a'), 147.00 (C7a' or 3a'), 165.79, 165.67 (br s, C2, 4, 6). (The observed <sup>13</sup>C-signals assignable to the predominant tautomer are asterisked.) Positive-ion FAB-MS *m/z*: 815 (M+H)<sup>+</sup>. HR-FAB-MS *m/z*:



815.3017 (Calcd for C<sub>40</sub>H<sub>39</sub>N<sub>12</sub>O<sub>8</sub>: 815.3014). Anal. Calcd for C<sub>40</sub>H<sub>38</sub>N<sub>12</sub>O<sub>8</sub>: C, 58.96; H, 4.70; N, 20.63. Found: C, 58.77; H, 4.69; N, 20.56.

**Synthesis of N<sup>2</sup>,N<sup>2'</sup>-(Propane-1,3-diyl)bis[N<sup>4</sup>,N<sup>6</sup>-bis(benzo[d][1,3]dioxol-5-ylmethyl)-1,3,5-triazine-2,4,6-triamine] (3b-3)**

To a suspension of intermediate **2b** (724 mg, 1.75 mmol) in DMF (3.0 mL) were added 1,3-propanediamine (51.9 mg, 0.700 mmol) and K<sub>2</sub>CO<sub>3</sub> (484 mg, 3.50 mmol) at room temperature with stirring. The reaction mixture was irradiated with a 120 W microwave (MW) at 120 °C for 40 min with stirring. After addition of water (20 mL), the resulting mixture was extracted with CH<sub>2</sub>Cl<sub>2</sub> (20 mL×3). The combined organic layer was washed with brine (20 mL) and dried over MgSO<sub>4</sub>. After evaporation of the solvent, the residual oil was purified by centrifugal chromatography (CH<sub>2</sub>Cl<sub>2</sub> : 95% EtOH : 28% NH<sub>3</sub> = 95 : 4.7 : 0.3 ) to give compound **3b-3** (351 mg, 0.424 mmol, 61%) as a white solid. Mp 60-65 °C. IR (KBr) cm<sup>-1</sup>: 3411, 3264 (NH), 1565, 1501 (C=N), 1249, 1038 (C-N and/or C-O). <sup>1</sup>H-NMR (CDCl<sub>3</sub>) δ: 1.69 (2H, br s, H2''), 2.76 (0.5H, br s, NH), *ca.* 3.25, 3.35\* (4H, br s, H1'', 3''), 4.35, 4.45\* (8H, br s, Hα), 5.22, 5.41 (2.2H, br s, NH), 5.89 (8H, br s, H2'), 6.6-6.85 (12H, m, ArH), 6.95 (0.6H, br s, NH), 7.36, 7.61 (2.7H, br s, NH). (The observed <sup>1</sup>H-signals assignable to the predominant tautomer are asterisked.) <sup>13</sup>C-NMR (CDCl<sub>3</sub>) δ: 30.01 (C2''), 37.69 (br s, C1'', 3''), 44.22 (Cα), 100.86 (C2'), 107.99 (C4', 7'), 120.36 (C6'), 133.52 (br s, C5'), 146.49 (C3a' or 7a'), 147.62 (C7a' or 3a'), 165.97 (br s, C2, 4, 6). Positive-ion FAB-MS *m/z*: 829 (M+H)<sup>+</sup>. HR-FAB-MS *m/z*: 829.3176 (Calcd for C<sub>41</sub>H<sub>41</sub>N<sub>12</sub>O<sub>8</sub>: 829.3170). Anal. Calcd for C<sub>41</sub>H<sub>40</sub>N<sub>12</sub>O<sub>8</sub>·1.2H<sub>2</sub>O: C, 57.90; H, 5.03; N, 19.76. Found: C, 57.94; H, 5.17; N, 19.75.

**Synthesis of N<sup>2</sup>,N<sup>2'</sup>-(Butane-1,4-diyl)bis[N<sup>4</sup>,N<sup>6</sup>-bis(benzo[d][1,3]dioxol-5-ylmethyl)-1,3,5-triazine-2,4,6-triamine] (3b-4)**

To a suspension of intermediate **2b** (724 mg, 1.75 mmol) in DMF (2.0 mL) were added 1,4-diaminobutane (62 mg, 0.700 mmol) and K<sub>2</sub>CO<sub>3</sub> (484 mg, 3.50 mmol) at room temperature with stirring. The reaction mixture was refluxed in a water bath for 8 h with stirring. After addition of water (20 mL), the resulting mixture was extracted with CH<sub>2</sub>Cl<sub>2</sub> (20 mL×3). The combined organic layer was washed with brine (20 mL) and dried over MgSO<sub>4</sub>. After evaporation of the solvent, the residual oil was purified by centrifugal chromatography (CH<sub>2</sub>Cl<sub>2</sub> : EtOH = 99 : 1→97 : 3 ) to give compound **3b-4** (423 mg, 0.502 mmol, 72%) as a white solid. Mp 79-81 °C (recrystallized from EtOH). IR (KBr) cm<sup>-1</sup>: 3412, 3266 (NH), 1566, 1519 (C=N), 1250, 1038 (C-N and/or C-O). <sup>1</sup>H-NMR (CDCl<sub>3</sub>) δ: 1.62 (4H, br s, H2'', 3''), 1.79 (3H, br s, NH), 3.42 (4H, br s, H1'', 4''), 4.26, 4.36, 4.47 (8H, br s, Hα), *ca.* 4.5 (0.5H, br s, NH), 4.84, 5.08, 5.19 (1.6H, br s, NH), 5.90 (8H, br s, H2'), 6.5-6.9 (12H, m, ArH), 6.99 (0.3H, br s, NH), 7.56, 7.84, 8.18 (0.6H, br s, NH). <sup>13</sup>C-NMR (CDCl<sub>3</sub>) δ: 23.75, 26.78\*, 29.66 (C2'', 3''), 39.96, 40.77\* (C1'', 4''), 44.24 (br s, Cα), 100.90 (C2'), 108.05 (C4', 7'), 120.45 (br s, C6'), 133.71 (br s, C5'), 146.53 (C3a' or

7a'), 147.68 (C7a' or 3a'), 165.69 (br s, C6), 166.15 (br s, C2, 4). (The observed  $^{13}\text{C}$ -signals assignable to the predominant tautomer are asterisked.) Positive-ion FAB-MS  $m/z$ : 843 (M+H) $^+$ . HR-FAB-MS  $m/z$ : 843.3336 (Calcd for  $\text{C}_{42}\text{H}_{43}\text{N}_{12}\text{O}_8$ : 843.3327). Anal. Calcd for  $\text{C}_{42}\text{H}_{42}\text{N}_{12}\text{O}_8$ : C, 59.61; H, 5.04; N, 19.68. Found: C, 59.85; H, 5.02; N, 19.94.

### Antiviral Activity Assay and Cytotoxicity

The anti-HSV-1 activities ( $\text{EC}_{50}$ ) of the synthesized hybrid linker mode  $\text{C}_2$ -symmetrical TAZ derivatives (**3**) were measured by using a plaque reduction assay,<sup>16</sup> and their cytotoxicity against Vero cells ( $\text{CC}_{50}$ ) was also evaluated as we described previously. The results are summarized in Table together with data for aciclovir.<sup>17</sup> There were few distinct correlations between log  $P$  values and anti-HSV-1 activity ( $\text{EC}_{50}$ ).

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