# Eleven Novel Diarylheptanoids and Two Unusual Diarylheptanoid Derivatives from the Seeds of *Alpinia blepharocalyx*

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An EtOH extract of the seeds of *Alpinia blepharocalyx* afforded 11 novel diarylheptanoids, named deoxycalyxin A (**1**), epicalyxin F (**2**), calyxin K (**3**), epicalyxin K (**4**), calyxin I (**5**), epicalyxin I (**6**), calyxin J (**7**), epicalyxin J (**8**), and calyxin L (**9**), an epimeric mixture of calyxin M (**10**) and epicalyxin M (**11**), and two unusual diarylheptanoid derivatives, named neocalyxins A (**12**) and B (**13**), together with four known calyxins, calyxins A (**14**), F (**15**), E (**16**), and G (**17**). Structures were elucidated by spectroscopic techniques including 2D NMR spectroscopy. All compounds were examined for cytotoxicity toward murine colon 26-L5 carcinoma and human HT-1080 fibrosarcoma cells. Diarylheptanoids **2**, **3**, and **5** were cytotoxic against both cell lines, while **4** and **6**−**8** were cytotoxic against human fibrosarcoma cells.

*Alpinia blepharocalyx* K. Schum. (Zingiberaceae) is widely distributed in the southwest of China including Yunnan and Shichuan provinces and Tibet. The seeds are used in Chinese traditional medicine for the treatment of stomach disorders.[1] Previously we isolated novel diarylheptanoids bearing a chalcone or a flavanone moiety, calyxins A−H and epicalyxins B−D, G, and H, from an ether-soluble fraction of an EtOH extract of the seeds of *A. blepharocalyx*.[2] To further investigate the bioactive natural products from this species, the residual fraction of the EtOH extract was subjected to a series of chromatographic separations to afford 26 new and seven known diarylheptanoids. The new diarylheptanoids are classified into five groups: acyclic diarylheptanoids, cyclic diarylheptanoids, dimeric diarylheptanoids, novel diarylheptanoids having either a chalcone or a flavanone moiety, and other unusual diarylheptanoid derivatives. In the two preceding papers,[3] we reported the structures of the new diarylheptanoids belonging to the first three classes. In this paper, we report the structures of diarylheptanoids of the other two classes and their cytotoxicity toward murine colon 26-L5 carcinoma[4] and human HT-1080 fibrosarcoma[5] cells.[6]

## Results and Discussion

The IR spectrum of deoxycalyxin A (**1**) showed absorptions at 3400 (OH) and 1610 $cm^{-1}$ (C═O). The molecular formula was determined by HRFABMS to be $C_{35}H_{34}O_8$, one oxygen atom less than that of calyxin A[2b] (**14**), also isolated from the same extract. The $^1H$ and $^{13}C$ NMR spectra of **1** were identical to those of **14**, except for differences in the chemical shifts for CH-3 and $CH_2$-4, suggesting that **1** should have a hydroxyl group at C-3 instead of a hydroperoxyl group in **14**. This was also supported by the similarity of the spectral data with those of a permethylate of deoxycalyxin A, previously obtained by methylation of **14**.[2b]

Epicalyxin F (**2**) was obtained as a mixture with calyxin F (**15**) and separated by HPLC with a chiral column. The molecular formula of **2** was determined by HRFABMS to be $C_{35}H_{34}O_8$, the same as that of **15**. The IR spectrum of **2** showed absorption bands at 3400 (OH) and 1610 $cm^{-1}$ (C═O). The $^1H$ and $^{13}C$ NMR data of **2** were very similar to those of **15** (Tables 1, 2) and showed signals for six sets of *ortho*-coupled aromatic protons, a singlet aromatic proton, two *trans*-olefin protons, a methoxyl group, two oxymethine protons, and four methylenes. However, H-7 of **2** appeared at slightly higher field ($\delta$ 5.05) than that of **15** ($\delta$ 5.13), suggesting that **2** was an epimer of **15** at C-7. This was confirmed by the ROESY experiment. In the ROESY spectrum of **2**, significant correlations were observed between H-$3_{ax}$ and H-$6_{ax}$ and between H-$4_{ax}$ and H-$7_{ax}$. These correlations indicated that H-3, H-7, H-$6_{ax}$, and H-$4_{ax}$ should all be on the same side and that the pyran ring should have a boat conformation. Considering the absolute configuration of calyxin F (**15**) as 3*S*,5*R*,7*R* reported,[2b] the absolute configuration of epicalyxin F (**2**) was concluded to be 3*S*,5*R*,7*S*.

The molecular formulas of calyxin K (**3**) and epicalyxin K (**4**) were determined to be the same as **2** ($C_{35}H_{34}O_8$) by negative ion HRFABMS. Their $^1H$ and $^{13}C$ NMR spectra were similar to each other and to those of **2** (Tables 1, 2) and showed the presence of the same diarylheptanoid moiety as in **2**. However, the signals for *trans*-olefin disappeared, and those for one methylene and one oxygenated methine appeared. Moreover, H-5″ and C-5″ of **3** and **4** appeared at lower field (**3**, $\delta_H$ 6.15, $\delta_C$ 95.1; **4**, $\delta_H$ 6.16, $\delta_C$ 95.4) than those of **2** ($\delta_H$ 5.95, $\delta_C$ 93.4). The former chemical shifts indicate a flavanone moiety as in calyxins C−E and epicalyxins C and D, while those of **2** indicate a chalcone moiety as in calyxins A and B and epicalyxin B.[2a,b] Thus, calyxin K (**3**) and epicalyxin K (**4**) were considered to be diarylheptanoids bearing a flavanone moiety instead of the chalcone moiety in **2**. The stereochemistry at the chiral centers on the tetrahydropyran ring in **3** and **4** was determined by ROESY experiments. The ROESY correlations H-3/H-5 and H-5/H-7 indicated the protons H-3, H-5, and H-7 to be *cis*. Because the diarylheptanoids obtained from the EtOH extract of *A. blepharocalyx* were assumed to have 3*S* configurations from biogenetic considerations,[2a−c] the stereochemistry in the heptanoid part of **3** and **4** should be 3*S*,5*R*,7*S*; **3** and **4** should be epimers at C-9″.

The HRFABMS of calyxin I (**5**) showed a quasi-molecular ion at *m/z* 678.2598 ($C_{42}H_{38}O_9$). Its IR spectrum showed a broad absorption band at 3400 $cm^{-1}$ (OH) and a sharp absorption band at 1700 $cm^{-1}$ (C═O). The $^1H$ NMR spectrum displayed signals corresponding to six sets of *ortho*-coupled aromatic protons, a singlet aromatic proton,

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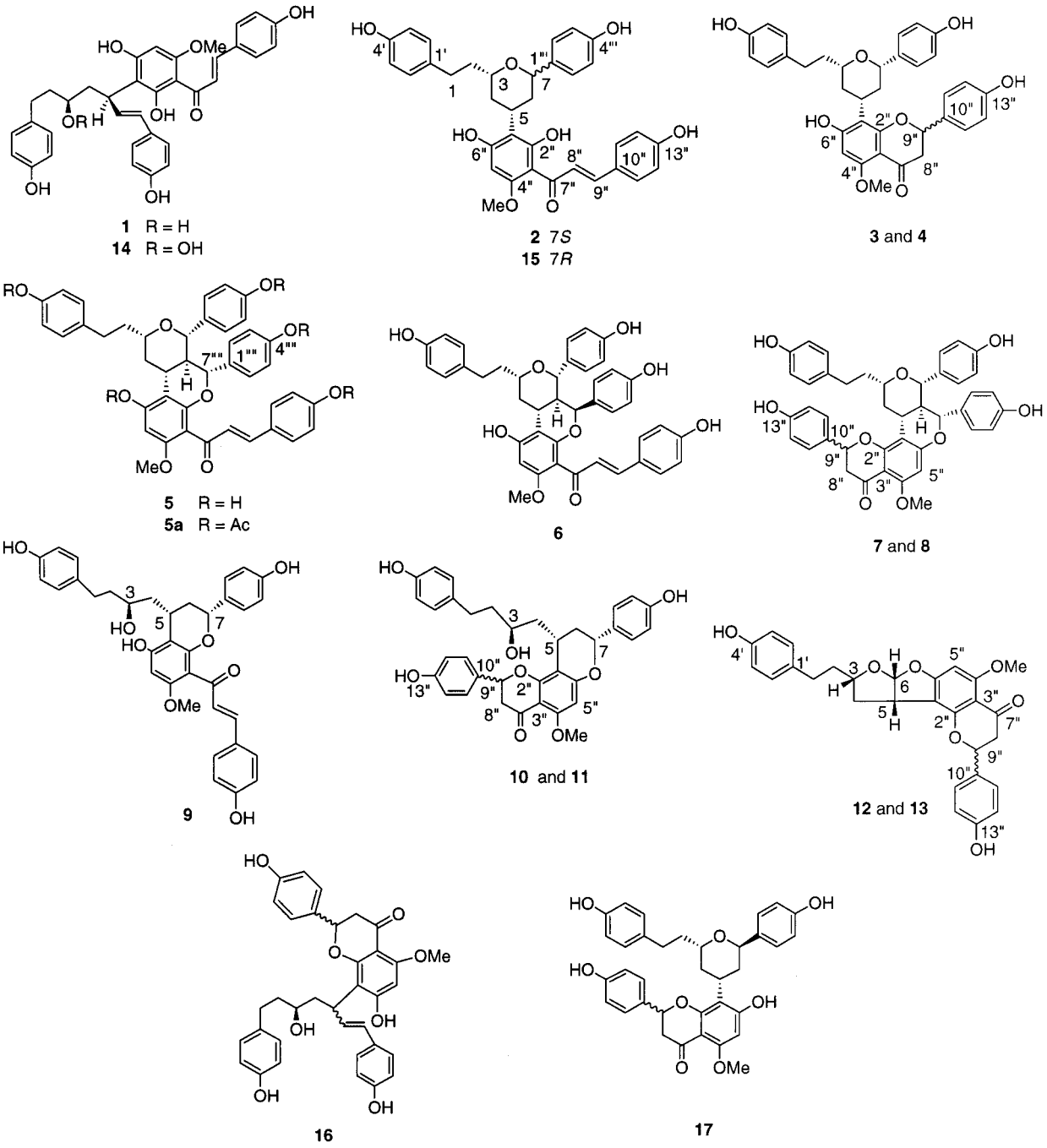
**Chart 1**




a methoxyl group, two *trans*-olefin protons, two oxymethine protons, and two methylene groups (Table 1). These signals were almost identical to those of **2**. In addition, the extra signals of two sets of *ortho*-coupled aromatic protons ($\delta$ 7.65, 6.33, both d, $J = 8.5$ Hz) and an oxymethine proton ($\delta$ 4.64, d, $J = 10$ Hz) were observed, suggesting **5** to have an extra *para*-substituted benzene group and an oxymethine group. This was further supported by corresponding signals in the $^{13}$C NMR spectrum (Table 2). Based on detailed analyses of the COSY and TOCSY spectra, C-6 of the tetrahydropyran ring was connected to the additional oxymethine carbon. The significant correlations observed between H-7′′′′ and C-5 and C-6 in the HMBC spectrum also supported this conclusion (Figure 1a in Supporting Information). Correlation between H-7′′′′ and C-1′′′′ and C-2′′′′ in the HMBC spectrum revealed the attachment of a *p*-hydroxybenzyl group to C-6 of the tetrahydropyran ring. The slight low-field shift of C-2′′ of **5** ($\delta$ 167.4) as compared to the corresponding signal of **2** ($\delta$ 166.7) indicated an ether linkage between C-7′′′′ and C-2′′, an observation confirmed by acetylation of **5**, which gave only a pentaacetyl derivative (**5a**). The relative stereochemistry in **5** was elucidated on the basis of coupling pattern of the protons in the rings A and B and by the ROESY data (Figure 1b in Supporting Information). Both H-7 and H-7′′′′ appeared as doublets with coupling constants of 10.0 Hz, suggesting they should have diaxial interaction with H-$6_{ax}$ (q, $J = 10.0$ Hz), while the triple doublet of H-5 was considered to be due to diaxial coupling with H-$4_{ax}$ (8.0 Hz) and H-$6_{ax}$ (10.0 Hz) and axial−equatorial coupling with H-$4_{eq}$ (4.0 Hz) in a chair conformation. On the other hand, ring B was concluded to have distorted chair conformation based on the coupling constant (10.0 Hz) between H-7′′′′ and H-$6_{ax}$. This was further supported by intense cross-

**Table 1.** $^1$H NMR Data for Diarylheptanoids **1**−**13** in $CD_3OD$[f]

| no. | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
|---|---|---|---|---|---|---|---|
| 1 | 2.52 m | 2.65 m (2H) | 2.46 ddd (14.5, 9.2, 4.7) | 2.40 m | 2.57 m (2H) | 2.57 m (2H) | 2.41 m |
| | 2.57 m | | 2.27 m | 2.16 m | | | 2.30 m |
| 2 | 1.65 m; 1.71 m | 1.76 m (2H) | 1.57 m; 1.41 m | 1.47 m; 1.34 m | 1.78 m (2H) | 1.69 m (2H) | 1.70 m; 1.53 m |
| 3 | 3.42 m | 3.78 m | 3.53 m | 3.53 m | 3.62 m | 3.47 tdd (12.5, 11.4, 5.2) | 3.42 m |
| 4 | 2.25 m; 1.85 m | 2.02 m; 1.78 m | 2.28 m; 1.27 m | 2.13 m; 1.26 m | 3.32 m; 1.32 m | 3.32 m[d], 1.27 m | 3.15 m; 1.27 m |
| 5 | 4.21 q (8.0) | 3.38 m | 3.01 m | 3.02 m | 3.08 ddd (10.0, 8.0, 4.0) | 2.93 td (10.0, 3.5) | 3.05 m |
| 6 | 6.58 dd (15.5, 8.0) | 2.25 m | 2.38 m | 2.40 m | 2.26 q (10.0) | 2.53 td (10.0, 4.3) | 2.38 m |
| | | 1.92 m | 1.90 q (11.5) | 1.86 q (11.5) | | | |
| 7 | 6.26 d (15.5) | 5.05 dd (12.0, 2.0) | 4.78 m[a] | 4.78 m[b] | 4.16 d (10.0) | 3.82 d (10.0) | 4.15 d (9.7) |
| 2′,6′ | 6.94 d (8.5) | 7.02 d (8.5) | 6.84 d (8.5) | 6.82 d (8.4) | 6.94 d (8.5) | 6.67 d (8.5) | 6.74 d (8.5) |
| 3′,5′ | 6.60 d (8.5) | 6.67 d (8.5) | 6.64 d (8.5) | 6.64 d (8.4) | 6.64 d (8.5) | 6.62 d (8.5) | 6.62 d (8.5) |
| 5″ | 6.03 s | 5.95 s | 6.15 s | 6.16 s | 5.85 s | 6.00 s | 6.03 s |
| 8″ | 7.79 d (15.5) | 7.80 d (16.0) | 3.08 dd (17.0, 14.0) | 3.02 dd (17.0, 13.5) | 7.78 d (16.0) | 7.81 d (15.5) | 3.08 m |
| | | | 2.65 dd (17.0, 2.5) | 2.65 dd (17.0, 3.0) | | | 2.67 dd (17.0, 3.2) |
| 9″ | 7.68 d (15.5) | 7.70 d (16.0) | 5.29 dd (14.0, 2.5) | 5.29 dd (13.5, 3.0) | 7.70 d (16.0) | 7.71 d (15.5) | 5.32 dd (13.5, 3.2) |
| 11″,15″ | 7.49 d (8.7) | 7.58 d (8.5) | 7.33 d (8.5) | 7.32 d (8.4) | 7.48 d (8.5) | 7.51 d (8.5) | 7.39 d (8.5) |
| 12″,14″ | 6.82 d (8.7) | 6.82 d (8.5) | 6.80 d (8.5) | 6.79 d (8.4) | 6.84 d (8.5) | 6.82 d (8.5) | 6.83 d (8.5) |
| OMe | 3.91 s | 3.90 s | 3.81 s | 3.81 s | 3.90 s | 3.89 s | 3.71 s |
| 2‴,6‴ | 7.14 d (8.5) | 7.58 d (8.5) | 7.18 d (8.5) | 7.19d (8.4) | 6.62 d (8.5)[c] | 6.94 d (8.5) | 6.64 d (8.5) |
| 3‴,5‴ | 6.66 d (8.5) | 6.79 d (8.5) | 6.77 d (8.5) | 6.77 d (8.4) | 6.33 d (8.5) | 6.65 d (8.5) | 6.33 d (8.5) |
| 2⁗,6⁗ | | | | | 6.65 d (8.5)[c] | 7.22 d (8.5) | 6.63 d (8.5) |
| 3⁗,5⁗ | | | | | 6.33 d (8.5) | 6.88 d (8.5) | 6.31 d (8.5) |
| 7⁗ | | | | | 4.64 d (10.0) | 4.82 m[e] | 4.70 d (9.7) |

| no. | 8 | 9 | 10 | 11 | 12 | 13 |
|---|---|---|---|---|---|---|
| 1 | 2.40 m | 2.64 m | 2.58 m (2H) | 2.58 m (2H) | 2.56 ddd (13.0, 9.5, 4.7) | 2.56 ddd (13.3, 8.4, 4.7) |
| | 2.31 m | 2.58 m | | | 2.40 ddd (13.0, 7.2, 5.3) | 2.40 m |
| 2 | 1.69 m; 1.54 m | 1.88 m; 1.72 m | 1.76 m; 1.68 m | 1.76 m; 1.68 m | 1.52 m; 1.38 m | 1.50 m; 1.44 m |
| 3 | 3.41 m | 3.52 m | 3.88 m | 3.88 m | 4.26 m | 4.26 m |
| 4 | 3.12 m | 2.25 q (12.0) | 2.14 q (12.0) | 2.06 q (12.0) | 2.33 ddd (13.0, 9.3, 7.8) | 2.33 ddd (13.0, 9.3, 7.8) |
| | 1.25 m | 1.52 dt (12.0, 2.0) | 1.52 m | 1.52 m | 1.96 dt (13.0, 2.3) | 1.96 dt (13.0, 2.3) |
| 5 | 2.96 m | 3.58 m | 3.90 m | 3.90 m | 3.94 ddd (9.3, 6.1, 2.3) | 3.94 ddd (9.3, 6.3, 2.3) |
| 6 | 2.23 m | 2.45 q (12.0) | 2.38 q (12.0) | 2.46 q (12.0) | 6.37 d (6.1) | 6.37 d (6.3) |
| | | 1.63 dt (12.0, 2.0) | 1.56 dt (12.0, 2.0) | 1.56 dt (12.0, 2.0) | | |
| 7 | 4.08 d (9.5) | 4.35 dd (12.0, 2.0) | 4.26 dd (12.0, 2.0) | 4.26 dd (12.0, 2.0) | | |
| 2′,6′ | 6.77 d (8.5) | 6.98 d (8.5) | 6.96 d (8.3) | 6.94 d (8.3) | 6.84 d (8.5) | 6.86 d (8.5) |
| 3′,5′ | 6.63 d (8.5) | 6.68 d (8.5) | 6.68 d (8.3) | 6.66 d (8.3) | 6.62 d (8.5) | 6.62 d (8.5) |
| 5″ | 5.96 s | 5.98 s | 6.11 s | 6.11 s | 6.21 s | 6.22 s |
| 8″ | 3.05 m | 7.75 d (15.5) | 2.96 dd (16.5, 12.0) | 2.98 dd (16.5, 13.0) | 3.03 dd (16.8, 12.0) | 3.06 dd (17.0, 13.0) |
| | 2.63 dd (17.0, 2.0) | | 2.65 br d (16.5) | 2.68 br d (16.5) | 2.72 dd (16.8, 3.0) | 2.70 dd (17.0, 3.0) |
| 9″ | 5.30 dd (13.5, 2.0) | 7.62 d (15.5) | 5.26 dd (12.0, 3.5) | 5.30 dd (13.0, 2.5) | 5.41 dd (12.0, 3.0) | 5.36 dd (13.0, 3.0) |
| 11″,15″ | 7.75 d (8.5) | 7.48 d (8.5) | 7.35 d (8.5) | 7.35 d (8.5) | 7.28 d (8.5) | 7.30 d (8.5) |
| 12″,14″ | 6.84 d (8.5) | 6.80 d (8.5) | 6.86 d (8.5) | 6.86 d (8.5) | 6.79 d (8.5) | 6.81 d (8.5) |
| OMe | 3.62 s | 3.90 s | 3.78 s | 3.78 s | 3.84 s | 3.84 s |
| 2‴,6‴ | 6.63 d (8.5) | 7.22 d (8.5) | 7.04 d (8.5) | 7.04 d (8.3) | | |
| 3‴,5‴ | 6.33 d (8.5) | 6.72 d (8.5) | 6.73 d (8.5) | 6.66 d (8.3) | | |
| 2⁗,6⁗ | 6.63 d (8.5) | | | | | |
| 3⁗,5⁗ | 6.31 d (8.5) | | | | | |
| 7⁗ | 4.62 d (9.5) | | | | | |

[a,b,e] Overlapped with the $H_2O$ signal but appeared in acetone-$d_6$ at $\delta$ 4.71 (dd, $J$ = 11.0, 2.0 Hz), 4.70 (dd, $J$ = 10.6, 2.0 Hz), and 4.88 (d, $J$ = 4.3 Hz), respectively. [c] Values may be interchanged. [d] Overlapped with the solvent signal but appeared in acetone-$d_6$ at $\delta$ 3.32 (dt, $J$ = 12.5, 3.5). [f] The chemical shifts are in $\delta$ values and coupling constants (parentheses) are given in Hz.

peaks H-5/H-7, H-5/H-7⁗, H-5/H-3, H-4/H-6 in the ROESY spectrum. Since all the diarylheptanoids isolated from *A. blepharocalyx* possessed 3*S* configuration,[2a−c] the absolute configuration at the chiral centers of **5** was assumed to be 3*S*,5*R*,6*S*,7*S*,7⁗*R*.

The HRFABMS of epicalyxin I (**6**) indicated the same molecular formula as **5**. The $^1$H and $^{13}$C NMR spectra were similar to those of **5**, except for some differences in the chemical shifts and splitting patterns (Tables 1, 2), suggesting **6** to be a stereoisomer of **5**. Analysis of the $^1$H NMR spectrum and the COSY and HMQC spectra indicated that the coupling constant between H-6 and H-7⁗ in **6** (4.3 Hz) differed from that in **5** (10.0 Hz), while the coupling constants for the other protons were almost the same. Thus, **6** was considered to be the C-7⁗ epimer of **5**. From this and the ROESY correlations among H-3, H-5, and H-7 and between H-6 and H-$4_{ax}$ in methanol-$d_4$ and between H-6 and H-7⁗ in acetone-$d_6$, the absolute configuration of **6** was likely to be 3*S*,5*R*,6*S*,7*S*,7⁗*S*.

The molecular formula of calyxin J (**7**) was determined to be $C_{42}H_{38}O_9$, the same as **5** and **6**. The IR spectrum of **7** showed absorption bands at 3300 (OH) and 1635 cm$^{-1}$ (C=O). The $^1$H and $^{13}$C NMR spectra of **7** were similar to those of **5** (Tables 1, 2), but they showed the lack of the conjugated double bond in **7** and the presence of an oxygenated methine ($\delta$ 5.12) and a methylene ($\delta$ 3.05; $\delta$ 2.68) which were correlated with the $^{13}$C signals at $\delta$ 80.2 and 46.2, respectively, in the HMQC spectrum. The vicinal coupling constants for H-8″ were in close agreement with a typical value of flavanone,[7] and not of dihydrochalcone.[8] Thus, calyxin J (**7**) was concluded to have a flavanone moiety instead of a chalcone moiety.

Together with calyxin J (**7**), a mixture of **7** and its epimer, epicalyxin J (**8**), was also obtained, which could not be separated with a chiral column. The mixture showed a single spot on TLC using various solvent systems. However, the $^1$H and $^{13}$C NMR data of each compound

**Table 2.** $^{13}C$ NMR Data for Diarylheptanoids **1**−**13** in $CD_3OD$

| no. | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 32.0 | 33.0 | 31.8 | 31.8 | 31.4 | 31.8 | 31.4 | 31.6 | 31.7 | 31.7 | 31.7 | 32.6 | 32.7 |
| 2 | 40.6 | 41.5 | 39.6 | 40.0 | 39.0 | 39.4 | 39.1 | 39.2 | 39.3 | 39.5 | 39.4 | 39.9 | 39.9 |
| 3 | 70.6 | 69.6 | 71.5 | 71.9 | 77.9 | 78.5 | 78.2 | 78.1 | 79.0 | 79.1 | 79.1 | 82.7 | 82.8 |
| 4 | 41.8 | 42.8 | 40.1 | 40.4 | 37.3 | 36.9 | 36.6 | 37.0 | 35.3 | 37.6 | 37.9 | 36.2 | 36.3 |
| 5 | 36.7 | 27.3 | 30.9 | 31.0 | 40.0 | 32.1 | 40.1 | 40.1 | 32.9 | 33.5 | 33.5 | 45.3 | 45.2 |
| 6 | 131.4 | 34.8 | 44.5 | 45.0 | 49.8 | 47.6 | 49.3 | 50.3 | 37.0 | 35.8 | 35.6 | 115.6 | 115.6 |
| 7 | 129.7 | 76.2 | 79.5 | 79.6 | 83.0 | 82.5 | 83.6 | 83.3[a] | 81.2 | 81.6 | 81.6 | | |
| 1′ | 134.5 | 134.8 | 134.6 | 134.6 | 134.0 | 134.4 | 134.1 | 134.1 | 134.1 | 134.6 | 134.6 | 133.7 | 133.8 |
| 2′,6′ | 130.2 | 129.2 | 130.3 | 130.3 | 130.4 | 130.4 | 130.3 | 130.3 | 130.0 | 130.3 | 130.3 | 130.2 | 130.2 |
| 3′,5′ | 116.0 | 117.6 | 116.0 | 116.0 | 115.7 | 116.1 | 116.1 | 116.6 | 115.8 | 116.1 | 116.1 | 116.1 | 116.1 |
| 4′ | 156.0 | 156.8 | 156.1 | 156.1 | 155.7 | 156.2 | 156.2 | 156.1 | 155.8 | 156.2 | 156.2 | 156.4 | 156.4 |
| 1″ | 111.1 | 109.0 | 109.7 | 109.6 | 106.9 | 107.0 | 107.3 | 107.4 | 111.6 | 112.8 | 112.7 | 110.3 | 110.3 |
| 2″ | 163.9 | 166.7 | 164.5[a] | 164.4[a] | 167.4 | 167.8 | 164.8 | 165.0 | 166.5 | 164.4 | 164.4 | 160.5 | 160.8 |
| 3″ | 106.7 | 109.0 | 107.0 | 107.3 | 106.2 | 106.2 | 107.3 | 106.8 | 106.3 | 106.4 | 106.3 | 106.9 | 106.8 |
| 4″ | 161.0 | 162.8 | 161.6 | 161.7 | 162.0 | 162.8 | 162.2 | 161.9 | 162.0 | 162.0 | 162.0 | 165.2 | 165.2 |
| 5″ | 92.2 | 93.4 | 95.1 | 95.4 | 92.4 | 92.9 | 95.3 | 95.0 | 91.7 | 94.0 | 93.9 | 89.2 | 89.2 |
| 6″ | 162.6 | 161.7 | 164.7[a] | 165.1[a] | 163.5 | 163.2 | 164.5 | 163.9 | 163.9 | 164.7 | 164.7 | 167.0 | 166.9 |
| 7″ | 194.2 | 194.7 | 192.9 | 192. 8 | 193.9 | 194.4 | 193.0 | 192.8 | 193.7 | 192.9 | 193.0 | 192.4 | 191.9 |
| 8″ | 125.9 | 126.2 | 46.1 | 45.6 | 125.4 | 125.7 | 46.1 | 46.5 | 125.6 | 46.2 | 46.2 | 46.3 | 46.0 |
| 9″ | 143.4 | 144.7 | 80.8 | 80.9 | 143.5 | 143.9 | 81.2 | 80.6 | 143.0 | 80.2 | 80.0 | 80.4 | 80.3 |
| 10″ | 128.4 | 128.9 | 130.9 | 131.1 | 128.0 | 128.4 | 130.9 | 131.0 | 128.1 | 131.3 | 131.4 | 131.1 | 130.9 |
| 11″,15″ | 131.2 | 130.9 | 129.4 | 129.1 | 131.0 | 131.4 | 129.4 | 129.4 | 130.9 | 129.0 | 129.0 | 128.9 | 128.9 |
| 12″,14″ | 116.8 | 116.9 | 116.5 | 116.2 | 116.6 | 116.9 | 116.6 | 115.4 | 116.6 | 116.4 | 116.4 | 116.4 | 116.4 |
| 13″ | 158.3 | 161.7 | 159.2 | 159.1 | 160.7 | 161.2 | 159.3 | 159.1 | 160.6 | 158.9 | 158.9 | 159.0 | 159.0 |
| OMe | 56.2 | 57.1 | 56.2 | 56.2 | 56.0 | 56.4 | 56.2 | 56.2 | 55.9 | 56.0 | 56.0 | 56.7 | 56.7 |
| 1‴ | 131.3 | 133.8 | 133.0 | 133.0 | 130.7 | 130.1 | 131.1 | 131.1 | 135.3 | 135.7 | 135.7 | | |
| 2‴,6‴ | 128.2 | 128.9 | 128.4 | 128.5 | 130.2 | 129.8 | 130.6 | 130.6 | 128.4 | 128.9 | 128.8 | | |
| 3‴,5‴ | 116.1 | 116.8 | 116.2 | 116.5 | 115.1 | 115.9 | 115.5 | 116.1 | 115.5 | 115.9 | 115.9 | | |
| 4‴ | 157.3 | 158.3 | 158.3 | 158.4 | 156.7 | 158.6 | 157.3 | 157.2 | 157.2 | 157.7 | 157.7 | | |
| 1⁗ | | | | | 133.0 | 131.1 | 133.3 | 133.2 | | | | | |
| 2⁗,6⁗ | | | | | 130.2 | 131.2 | 130.6 | 130.7 | | | | | |
| 3⁗,5⁗ | | | | | 115.0 | 116.5 | 115.4 | 116.1 | | | | | |
| 4⁗ | | | | | 157.4 | 159.0 | 157.9 | 157.9 | | | | | |
| 7⁗ | | | | | 83.3 | 78.8 | 83.6 | 83.5[a] | | | | | |

[a,b] Values may be interchanged in each column.

could be assigned by the analyses of the COSY, HMQC, and HMBC spectra of the mixture. The stereochemistry at the chiral centers (C-3, C-5, C-6, C-7, C-7⁗) in **7** and **8** was determined to be the same as that in **5** on the basis of their coupling constants and the ROESY data. Thus, **7** and **8** were determined to be epimers at C-9″. However, the stereochemistry at C-9″ could not be determined due to the paucity of **7** and **8** available.

The IR spectrum of calyxin L (**9**) indicated hydroxyl and carbonyl groups, and its molecular formula was determined to be $C_{35}H_{34}O_8$, the same as **2** and **15**. The $^1H$ and $^{13}C$ NMR spectra of **9** displayed signals corresponding to three *para*-substituted benzene rings, one *trans*-double bond, one singlet aromatic proton, one methoxyl, one ketone carbonyl, three (including two oxygenated) methines, and four methylenes. These data suggested **9** to be a diarylheptanoid with a chalcone moiety. Analysis of the COSY and HMQC spectra indicated the same connectivities as those in **2** and **15**, but differences were observed in the $^1H$ and $^{13}C$ NMR data for C-3 to C-7. Moreover, the coupling patterns of H-5, $H_2$-6, and H-7 suggested these protons to be part of a six-membered ring; that is, C-7 should be bonded with either C-2″ or C-6″ through an oxygen atom. Since in the $^{13}C$ NMR spectrum C-6″ in **9** appeared at a position similar to that in **1** (**9**, $\delta_C$ 163.9; **1**, $\delta_C$ 162.6), while C-2″ appeared at lower field (**9**, $\delta_C$ 166.5; **1**, $\delta_C$ 163.9) as in the cases of **5** and **6**, C-7 should be connected with C-2″ through an oxygen atom. The large coupling constant of H-$6_{ax}$ with H-5 and H-7 (12.0 Hz) and the ROESY correlations between H-5 and H-7 indicated H-5 and H-7 to be *cis*. C-3 and C-5 were considered to have *S* and *R* absolute configuration, respectively, since all the diarylheptanoids with a chalcone moiety at C-5 (calyxins A, F, and I and epicalyxins F and I) had the 3*S*,5*R* configuration.[2b] Thus, the stereochemistry of **9** was determined to be 3*S*,5*R*,7*R*.

Calyxin M (**10**) and epicalyxin M (**11**) were obtained as an epimeric mixture, showing a single spot on TLC with various solvent systems. The mixture could not be separated with a chiral column, and HRFABMS revealed the molecular formula $C_{35}H_{34}O_8$, the same as **9**. The $^1H$ and $^{13}C$ NMR spectra of the mixture displayed two sets of signals in 3:2 ratio of intensity, and analysis of the COSY, HMQC, and HMBC spectra resulted in the assignments of signals to each epimer (Tables 1, 2). The assigned data resembled those of **9** but showed the lack of a conjugated double bond and the presence of an oxygenated methine and a methylene. This and the low-field shifts of H-5″ and C-5″, compared to those of **9** (H-5″), suggested the presence of a flavanone moiety instead of a chalcone moiety in **9**. This assignment was confirmed by the HMBC correlations. The $^1H$ and $^{13}C$ NMR data for **10** and **11** differed at C-7″ and C-9″, and the ROESY correlations of H-5 with H-7 and H-$6_{eq}$, of H-7 with H-$6_{eq}$, and of H-3 with H-4 indicated that **10** and **11** were epimers at C-9″.

Negative ion HRFABMS of neocalyxin A (**12**) indicated its molecular formula to be $C_{28}H_{26}O_7$, and the IR spectrum showed an absorption band at 1615 $cm^{-1}$ (C═O). Its $^1H$ and $^{13}C$ NMR spectra revealed the presence of two *para*-substituted benzene rings, a pentasubstituted benzene ring, a methoxyl group, a ketone group, four (including two oxygen-substituted and a ketal) methines, and four methylenes (Tables 1, 2). COSY and HMQC data indicated the presence of the partial structures depicted (Figure 2 in Supporting Information) by bold lines, while the HMBC correlations suggested the presence of a flavanone moiety and the connectivity of the partial structures. As C-6 was

**Table 3.** Cytotoxic Activity (in vitro) of Diarylheptanoids **1**−**17**[a]

| compound | colon 26-L5 | HT-1080 |
|---|---|---|
| **1** | 27.4 | 26.5 |
| **2** | 0.89 | 1.71 |
| **3** | 7.73 | 5.09 |
| **4** | 33.0 | 4.75 |
| **5** | 8.39 | 9.08 |
| **6** | 12.1 | 5.88 |
| **7** | 23.2 | 8.19 |
| mixture of **7** and **8** (1:1) | 13.7 | 0.32 |
| **9** | 28.2 | 44.3 |
| mixture of **10** and **11** (3:2) | 42.1 | 10.1 |
| **12** | >100 | 10.7 |
| **13** | 78.0 | 20.2 |
| **14** | 13.1 | 10.7 |
| **15** | 10.4 | 10.4 |
| **16** | 98.1 | 21.7 |
| **17** | 42.2 | 25.9 |
| curcumin | 23.2 | 23.4 |
| 5-fluorouracil | 0.53 | 8.00 |

[a] $ED_{50}$ values (in $\mu$M) were calculated from the mean of six determinations.

ascribed to be a ketal from the chemical shifts (H-6, $\delta$ 6.37; C-6, $\delta$ 115.6) and the HMBC correlations of H-6 with C-3 and C-6″, the planar structure was deduced to be as indicated. In the ROESY spectrum, **12** showed the correlations H-3/H-$4_{ax}$, H-$4_{ax}$/H-5, and H-5/H-6, indicating H-3, H-5, and H-6 to be *cis*. From this and an assumption of 3*S* configuration at C-3, the stereochemistry of neocalyxin A (**12**) was deduced to be 3*S*,5*S*,6*R*, but that at C-9″ could not be determined.

HRFABMS of neocalyxin B (**13**) indicated the same molecular formula, $C_{28}H_{26}O_7$, as that of **12**. The $^1$H and $^{13}$C NMR spectra of **13** were very similar to those of **12** (Tables 1, 2), suggesting **13** to be an isomer of **12**. The coupling constants for H-3/H-$4_{ax}$, H-$4_{ax}$/H-5, and H-5/H-6 (6.3 Hz) were almost the same as those of **12**, and the ROESY spectrum showed the same H-3/H-$4_{ax}$, H-$4_{ax}$/H-5, and H-5/H-6 correlations as observed in **12**. Thus, neocalyxin B (**13**) was proposed to have the same configuration, 3*S*,5*S*,6*R*, as **12**; that is, **13** is a C-9″ epimer of **12**.

All the diarylheptanoids reported in this paper have a chalcone (**1**, **2**, **5**, **6**, **9**) or a flavanone (**3**, **4**, **7**, **8**, **10**−**13**) moiety at C-5 in the tetrahydropyran or tetrahydrofuran ring. Diarylheptanoids **5**−**8** possess an oxygenated *p*-hydroxybenzyl group at C-6 to present a novel carbon framework. Compounds **12** and **13** also possess a novel carbon framework which is assumed to be derived by the oxidative degradation of a diarylheptanoid with a flavanone moiety at C-5. Though the biogenetic pathways leading to the novel diarylheptanoids are not clear, they are assumed to be formed from deoxycalyxin A (**1**) or calyxin E (**16**), which would be derived from 1,2-dihydro-1,7-bis(de-*O*-methyl)curcumin and helichrysetin or 5-*O*-methylnaringenin, respectively (Scheme 1 in Supporting Information).[2b]

Cytotoxicity of all the isolated compounds was tested against murine colon 26-L5 carcinoma and human HT-1080 fibrosarcoma cells, using the standard MTT assay[9] (Table 3). Compounds other than **2**, **5**, and **9** showed stronger activity toward human HT-1080 fibrosarcoma cells than toward colon 26-L5 carcinoma cells. Epicalyxin F (**2**) showed more potent activity against colon carcinoma cells with an $ED_{50}$ value of 0.89 $\mu$M, while a 1:1 mixture of **7** and **8** exhibited more potent activity against human fibrosarcoma cells with an $ED_{50}$ value of 0.32 $\mu$M, which is more active than 5-fluorouracil (5-FU), a clinically used drug for the treatment of human cancers.[10] Interestingly, the activity of **7** was identical with 5-FU, and thus **8** should have a much smaller $ED_{50}$ value than 0.32 $\mu$M. Compounds **2**, **3**, and **5** exhibited potent activity against both cell lines, while compounds **4**, **6**, and **7** and a mixture of **7** and **8** (1:1) showed moderate to potent cytotoxicity against human fibrosarcoma cells with $ED_{50}$ values less than 10 $\mu$M. The $ED_{50}$ values for **2**−**4** and **6** and a mixture of **7** and **8** (1:1) toward human fibrosarcoma cells fall within the range of active cytotoxic agents ($ED_{50}$ < 4 $\mu$g/mL) recognized by Geran et al.[11]

## Experimental Section

**General Experimental Procedures.** Optical rotations were determined in MeOH solutions on a JASCO DIP-140 digital polarimeter at 25 °C. IR spectra were recorded on a Shimadzu IR-408 spectrophotometer in KBr disks or in $CHCl_3$ solutions. $^1$H and $^{13}$C NMR spectra were measured in $CD_3OD$ on a JEOL JNM-GX400 spectrometer with tetramethylsilane as an internal standard, and chemical shifts were recorded in $\delta$ values. FABMS were measured with a JEOL JMS-700T spectrometer with glycerol as a matrix. HPLC analyses were conducted with a Shimadzu LC-5A system using a Sumichiral OA-4700 column (4.6 mm i.d. × 25 cm; Sumika Chemical Analysis Service Ltd., Japan). The mobile phase was hexane−1,2-dichloroethane−EtOH (80:12:8 for the separation of **1** and **13**, 70:20:10 for **2** and **3** and for **11** and **12**), and UV (254 nm) was used for detection. Analytical and preparative TLC were conducted on precoated Merck Kieselgel $60F_{254}$ (0.25 and 0.50 mm) and RP-$18F_{254}$ (0.25 mm) plates.

**Plant Material.** Seeds of *A. blepharocalyx* were procured from Mengha (1800 m from sea level), Yunnan Province, Peoples Republic of China, in August 1991. The plant sample was identified by Prof. Wu Te-Lin, South China-Institute of Botany, Academica Sinica, and a voucher specimen (CPU-9008037) is preserved in the herbarium of the China Pharmaceutical University.

**Extraction and Isolation.** The seeds (10 kg) of *A. blepharocalyx* were extracted with 95% EtOH by percolation at room temperature, and the extract was concentrated under reduced presure to give an EtOH extract (800 g), which was suspended in 10% $H_2O$−MeOH and partitioned into hexane, ether, and residual fractions. The residual fraction (60 g) was subjected to Sephadex LH-20 column chromatography (CC) with a $H_2O$−MeOH gradient system to provide 14 fractions.

Fraction 7 (13.9 g) was further subjected to CC on silica gel (700 g) with a $CHCl_3$−MeOH (99:1→70:30) gradient system to give 24 subfractions. Subfraction 9 ($CHCl_3$−MeOH = 92:8 eluate, 1.5 g) was separated by ODS column chromatography (MeOH−$H_2O$−MeCN, 5:3:2), followed by normal-phase PTLC ($C_6H_6$−$CHCl_3$−MeOH = 10:78:12), providing blepharocalyxin D (5.5 mg), calyxin I (**5**, 14.0 mg), epicalyxin I (**6**, 4.0 mg), calyxin L (**9**, 4.7 mg), and an epimeric mixture (18.2 mg) of calyxin J (**7**) and epicalyxin J (**8**). Subfraction 10 ($CHCl_3$−MeOH = 92:8 eluate, 1.47 g) was separated by reversed-phase PTLC (MeOH−$H_2O$−MeCN, 6:3:1) to afford (5*S*,6*S*)-5,6-dihydroxy-4″-de-*O*-methylcentrolobine (25.2 mg) and **9** (6.2 mg). Subfractions 11 and 12 ($CHCl_3$−MeOH = 91:9 eluate, 1.3 g) were combined and separated by ODS CC (MeOH−$H_2O$−MeCN, 5:3:2), followed by normal- ($C_6H_6$−$CHCl_3$−MeOH, 3:14:3) and reversed-phase (MeOH−$H_2O$−MeCN, 6:3:1) PTLC, to afford **5** (4.6 mg) and **7** (18.3 mg), together with two mixtures. The mixture of $R_f$ 0.47 ($CHCl_3$−MeOH, 9:1) was purified by HPLC to provide neocalyxins A (**12**, 2.2 mg, $t_R$ 29.3 min) and B (**13**, 2.2 mg, $t_R$ 30.9 min), while the other mixture of $R_f$ 0.28 ($CHCl_3$−MeOH, 9:1) was identified as a mixture (5.5 mg) of calyxin M (**10**) and epicalyxin M (**11**).

Fraction 9 (12.5 g) was chromatographed over silica gel with a $CHCl_3$−MeOH (99:1→50:50) gradient system to provide 14 subfractions. Normal-phase PTLC ($CHCl_3$−MeOH, 9:1), followed by reversed-phase PTLC (MeOH−$H_2O$−MeCN, 5:3:2), of subfractions 7 and 11 gave calyxin G (**17**, 1.2 mg) and calyxin E (**16**, 19.0 mg), respectively, while that of subfraction 9 provided an epimeric mixture ($R_f$ 0.30; $CHCl_3$−MeOH, 9:1),

which was separated by HPLC to afford calyxin K (**3**, 1.2 mg, $t_R$ 24.8 min) and epicalyxin K (**4**, 1.7 mg, $t_R$ 28.7 min).

Fraction 13 (12.0 g) was subjected to silica gel CC using a $CHCl_3$−MeOH (99:1→50:50) gradient system to give 15 subfractions. Subfraction 8 was separated by normal-phase PTLC ($CHCl_3$−MeOH, 9:1), followed by reversed-phase PTLC (MeOH−$H_2O$−MeCN, 5:3:2) and HPLC, to give epicalyxin F (**2**, 4.3 mg, $t_R$ 15.0 min) and calyxin F (**15**, 2.4 mg, $t_R$ 16.0 min). Subfraction 9, on normal-phase PTLC ($CHCl_3$−MeOH, 9:1) followed by reversed-phase PTLC (MeOH−$H_2O$−MeCN, 5:3:2), afforded **5** (8.3 mg) and **8** (5.6 mg), while that of subfractions 12 and 14 afforded deoxycalyxin A (**1**, 5.6 mg) and calyxin A (**14**, 33.0 mg), respectively.

**Deoxycalyxin A (1)**: light yellow amorphous solid; $[\alpha]_D^{25}$ +147.9° (*c* 0.035, MeOH); IR (KBr) $\nu_{max}$ 3400, 1610, 1510, 1440, 1340, 1140 $cm^{-1}$; $^1H$ and $^{13}C$ NMR, see Tables 1 and 2; HRFABMS *m/z* 583.2339 (calcd for $C_{35}H_{35}O_8$ $[M + H]^+$, 583.2332).

**Epicalyxin F (2)**: light yellow amorphous solid; $[\alpha]_D^{25}$ +103.1° (*c* 0.05, MeOH); IR (KBr) $\nu_{max}$ 3400, 1610, 1510, 1440, 1340, 1140 $cm^{-1}$; $^1H$ and $^{13}C$ NMR, see Tables 1 and 2; HRFABMS *m/z* 583.2332 (calcd for $C_{35}H_{35}O_8$ $[M + H]^+$, 583.2333).

**Calyxin K (3)**: pale yellow amorphous solid; $[\alpha]_D^{25}$ +35.5° (*c* 0.06, MeOH); $^1H$ and $^{13}C$ NMR, see Tables 1 and 2; HRFABMS *m/z* 581.2214 (calcd for $C_{35}H_{33}O_8$ $[M - H]^-$, 581.2176).

**Epicalyxin K (4)**: pale yellow amorphous solid; $[\alpha]_D^{25}$ −17.0° (*c* 0.085, MeOH); $^1H$ and $^{13}C$ NMR, see Tables 1 and 2; HRFABMS *m/z* 581.2240 (calcd for $C_{35}H_{33}O_8$ $[M - H]^-$, 581.2176).

**Calyxin I (5)**: light yellow amorphous solid; $[\alpha]_D^{25}$ −16.4° (*c* 0.05, MeOH); IR (KBr) $\nu_{max}$ 3400, 1610, 1510, 1440, 1340, 1200, 1140 $cm^{-1}$; $^1H$ and $^{13}C$ NMR, see Tables 1 and 2; HRFABMS *m/z* 687.2598 (calcd for $C_{42}H_{39}O_9$ $[M + H]^+$, 687.2594).

**Epicalyxin I (6)**: light yellow amorphous solid; $[\alpha]_D^{25}$ +28.3° (*c* 0.025, MeOH); IR (KBr) $\nu_{max}$ 3350, 1610, 1510, 1445, 1330 $cm^{-1}$; $^1H$ and $^{13}C$ NMR, see Tables 1 and 2; HRFABMS *m/z* 685.2433 (calcd for $C_{42}H_{37}O_9$ $[M - H]^-$, 685.2438).

**Calyxin J (7)**: light yellow amorphous solid; $[\alpha]_D^{25}$ +99.2° (*c* 0.185, MeOH); IR (KBr) $\nu_{max}$ 3300, 1635, 1505, 1440, 1130, 1090 $cm^{-1}$; $^1H$ and $^{13}C$ NMR, see Tables 1 and 2; HRFABMS *m/z* 685.2404 (calcd for $C_{42}H_{37}O_9$ $[M - H]^-$, 685.2438).

**Epicalyxin J (8)**: light yellow amorphous solid; IR (KBr) $\nu_{max}$ 3250, 1650, 1590, 1565, 1510, 1440, 1360, 1145, 1090 $cm^{-1}$; $^1H$ and $^{13}C$ NMR, see Tables 1 and 2; HRFABMS *m/z* 685.2471 (calcd for $C_{42}H_{37}O_9$ $[M - H]^-$, 685.2438).

**Calyxin L (9)**: light yellow amorphous solid; $[\alpha]_D^{25}$ +77.1° (*c* 0.05, MeOH); IR (KBr) $\nu_{max}$ 3350, 1600, 1510, 1440, 1220 $cm^{-1}$; $^1H$ and $^{13}C$ NMR, see Tables 1 and 2; HRFABMS *m/z* 581.2204 (calcd for $C_{35}H_{33}O_8$ $[M - H]^-$, 581.2175).

**An epimeric mixture of calyxin M (10) and epicalyxin M (11)**: yellow amorphous solid; IR (KBr) $\nu_{max}$ 3350, 1610, 1590, 1510, 1450 $cm^{-1}$; $^1H$ and $^{13}C$ NMR, see Tables 1 and 2; HRFABMS *m/z* 581.2171 (calcd for $C_{35}H_{33}O_8$ $[M - H]^-$, 581.2175).

**Neocalyxin A (12)**: pale yellow amorphous solid; $[\alpha]_D^{25}$ −21.5° (*c* 0.11, MeOH); IR ($CHCl_3$) $\nu_{max}$ 1615, 1590, 1510, 1465, 1100 $cm^{-1}$; $^1H$ and $^{13}C$ NMR, see Tables 1 and 2; HRFABMS *m/z* 473.1621 (calcd for $C_{28}H_{25}O_7$ $[M - H]^-$, 473.1600).

**Neocalyxin B (13)**: pale yellow amorphous solid; $[\alpha]_D^{25}$ −69.3° (*c* 0.11, MeOH); IR ($CHCl_3$) $\nu_{max}$ 1615, 1590, 1510, 1100 $cm^{-1}$; $^1H$ and $^{13}C$ NMR, see Tables 1 and 2; HRFABMS *m/z* 473.1616 (calcd for $C_{28}H_{25}O_7$ $[M - H]^-$, 473.1600).

**Acetylation of Calyxin I (5)**. A solution of **5** (1.0 mg) in dry pyridine (0.2 mL) and acetic anhydride (0.2 mL) was stirred overnight at room temperature. Then, the mixture was extracted with $CHCl_3$ (5 mL × 3), and the extract was washed with water, dried over anhydrous $MgSO_4$, and evaporated to yield calyxin I pentaacetate (**5a**, 0.5 mg): light yellow amorphous solid; $^1H$ NMR ($CDCl_3$, 400 MHz) $\delta$ 7.49 (2H, d, $J$ = 8.5 Hz, H-11″,15″), 7.34 (1H, d, $J$ = 16.0 Hz, H-9″), 7.05 (4H, d, $J$ = 8.5 Hz, H-12″,16″, H-2′,6′), 6.88 (2H, d, $J$ = 8.5 Hz, H-3′,5′), 6.86 (1H, d, $J$ = 16.0 Hz, H-8″), 6.77 (2H, d, $J$ = 8.5 Hz, H-3‴,5‴), 6.72 (2H, d, $J$ = 8.5 Hz, H-3⁗,5⁗), 6.64 (2H, d, $J$ = 8.5 Hz, H-2⁗,6⁗), 6.62 (2H, d, $J$ = 8.5 Hz, H-2‴,6″), 6.27 (1H, s, 5″), 4.73 (1H, d, $J$ = 10.0 Hz, H-7⁗), 4.20 (1H, d, $J$ = 10.0 Hz, H-7), 3.41 (1H, m, H-3), 5.05 (1H, ddd, $J$ = 10.0, 8.0, 4.0 Hz, H-5), 2.58 (2H, m, H-1), 2.52 (1H, m, H-4), 2.29 (1H, q-like, $J$ = 10.0 Hz, H-6), 2.24, 2.20, 2.18, 1.17, 1.12 (each 3H, s, OAc), 1.81, 1.70 (each 1H, m, $H_2$-2), 1.55 (1H, m, H-4); HRFABMS *m/z* 919.2917 (calcd for $C_{52}H_{48}O_{14}Na$ $[M + Na]^+$, 919.2942).

**Cytotoxicity.** Human HT-1080 fibrosarcoma and murine colon 26-L5 carcinoma cells were maintained in Eagle's minimum essential medium and RPMI-1640 medium (both Nissui Pharm. Co., Ltd., Tokyo, Japan), respectively. These media were supplemented with 10% fetal calf serum (Gibco BRL Products, Gaithersburg, MD), 0.1% $NaHCO_3$, and 2 mM glutamine (Wako Pure Chemicals Ind., Ltd., Kyoto, Japan). Cellular viability in the presence and absence of the experimental agents was determined using the standard MTT assay as described previously.[12] 5-Fluorouracil and curcumin were used as positive controls in the experiment.

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**Supporting Information Available:** Figure 1, showing significant HMBC and ROESY correlations of calyxin I (**5**) and bold line indicating the C−C bond deduced by the COSY and HMQC spectra; Figure 2, showing significant HMBC correlations of neocalyxin A (**12**) and bold line indicating the C−C bond deduced by the COSY and HMQC spectra; and Scheme 1, showing the possible biogenetic pathways for the novel diarylheptanoids. This material is available free of charge via the Internet at http://pubs.acs.org.

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