The interaction of uracil or uridine or uracil analogs with target biomolecules. Here, we targeted the characterization of certain physicochemical properties of CR, BR, and AU, as compared with uridine. These modifications of the uracil ring involved mostly substitution of various groups at the uracil C5 or C6 positions.8

Other pyrimidine-based antiviral and anticancer drugs, e.g., 6-azauracil, have been developed.8 In this type of uracil analog, the atoms constituting the ring have been replaced by others, e.g., a carbon atom was replaced by a nitrogen atom in 6-azauracil. Such changes affect chemical properties of the uracil ring, such as H-bonds, degree of aromaticity, and acid–base character. Another kind of uracil modification involves varying the size of the ring, e.g., a seven-membered ring analog of the uracil ring, azepine ring.9

The significance of the interaction of uracil or uridine or related nucleotides with target proteins in health and disease encouraged us to study binding patterns of uridine.10 Our data mining analysis of complexes of protein with uracil nucleosides/nucleotides provided insights regarding molecular recognition patterns. Thus, molecular recognition of the uracil moiety in uridine involves three types of interactions: π–π interactions between the uracil moiety and aromatic residues in the proteins were found in 59% of the inspected structures. Hydrogen bonds were the most dominant interaction between the uracil moiety and its binding proteins. The most abundant hydrogen bond interaction, present in 98% of the structures, was found between the uracil N3-H and a protein H-bond acceptor. The uracil O2 and O4 atoms act as H-bond acceptors in 80 and 93% of the cases, respectively (Figure 1).7

Based on these findings, the potential number of H-bonds formed by uracil is expected to increase by replacing the C5 and/or C6 carbon atoms of the uracil ring by NH/carbonyl moieties.

Indeed, such uridine mimetics, e.g., cyanuryl ribose (CR, 1), barbituryl ribose (BR, 2), and 6-aza-uridine (AU, 3), have been synthesized before.8,10 Uridine mimetics, cyanuryl-2'-deoxyriboside and 5-thiophene-6-aza-uridine, have been incorporated in nucleic acids, and their effect on various features of nucleic acids was studied.11–13

Here, we targeted the characterization of certain physicochemical properties of CR, BR, and AU, as compared with...
uridine, with a view to identify the usefulness of the former as agents for biochemical and pharmacological applications. Specifically, we studied features of compounds 1−3 that are indicative of potential binding interactions of these analogs, such as acid−base equilibria, base pairing, base stacking, and nucleoside conformation, as compared to each other and to uridine.

RESULTS AND DISCUSSION

Synthesis. For the improvement of the synthesis of CR\(^1\) (1), we employed the Vorbrüggen method, which is commonly used for the synthesis of nucleosides.\(^{15,16}\) However, the Vorbrüggen reaction suffers from unpredictable regioselectivity in some cases when heterocycles containing multiple basic sites are used, such as cyanuric acid.\(^{17,18}\)

Indeed, when we treated silylated cyanuric acid with 1-O-acetyl-2,3,5-tri-O-benzoyl-β-D-ribofuranose at RT for 2 h, CR was obtained in only 2% yield together with disubstituted triazine 6, obtained in 19% yield due to reaction of protected ribose 5, with two nitrogen atoms in silylated cyanuric acid (Scheme 1).

Therefore, an improved synthesis was developed based on the Vorbrüggen reaction to prevent the formation of disubstituted triazine 6.\(^{19}\) Specifically, we performed a one-pot reaction, where TMSCl (1 equiv) and SnCl\(_4\) (2 equiv) were added to an excess of cyanuric acid 4 (3 equiv) and 1-O-acetyl-2,3,5-tri-O-benzoyl-β-D-ribofuranose 5 (1 equiv) in HMDS and acetonitrile at RT. The turbid solution turned clear after about 1 h. Compound 7 was obtained in 82% yield. Finally, CR (1) was obtained quantitatively upon treatment of 7 with NaOH in MeOH at RT for 1 h.

Analogs 2 and 3 were commercially available.

Uridine Mimetics 1 and 2 Prefer Ribose N-Conformer and gg/gt-Rotamers. In the 70s, the conformation of various nucleotides and nucleosides was studied by NMR spectroscopy.\(^9,20,21\) NMR studies indicated that the ribose ring interconverts rapidly between N and S conformations with approximately equal residence. For example, uridine prefers N puckering (56%) over the S conformation (Figure 2). In addition, slight preference for the gauche−gauche rotamer of the exocyclic CH\(_2\)OH group was indicated (Figure 3).\(^{22}\)

Conformational analysis of BR was not reported, while sugar puckering of CR and the conformation of its exocyclic CH\(_2\)OH group have been studied only by low-field NMR.\(^{21,22}\) The ribose conformation of AU is known.\(^{10}\)

Here, we employed detailed high-field \(^1\)H NMR and \(^13\)C NMR spectroscopy to analyze the solution conformation and to

uridine, with a view to identify the usefulness of the former as agents for biochemical and pharmacological applications. Specifically, we studied features of compounds 1−3 that are indicative of potential binding interactions of these analogs, such as acid−base equilibria, base pairing, base stacking, and nucleoside conformation, as compared to each other and to uridine.

Scheme 1. Synthesis of CR\(^{2a}\)

Reagents and conditions: (a) i: cyanuric acid, dry HMDS, reflux, 6 h. ii: dry CH\(_3\)CN, protected ribose (5), SnCl\(_4\), 1 h at RT.\(^{18}\) (b) cyanuric acid, protected ribose (5), CH\(_3\)CN, HMDS, TMSCl, SnCl\(_4\), 1 h at RT. (c) 1 M NaOH in MeOH.

Figure 1. Recognition patterns in uracil-nucleos(t)ide binding proteins we previously reported.\(^7\) Numerical values in brackets represent percentages of occurrences of H-bonding, π−π interactions, π−cation interactions in the set of protein-uracil-nucleos(t)ide complexes studied.

Figure 2. NMR analysis of the conformation of uridine (1). NMR studies indicated that the ribose ring interconverts rapidly between N and S conformations with approximately equal residence. For example, uridine prefers N puckering (56%) over the S conformation (Figure 2). In addition, slight preference for the gauche−gauche rotamer of the exocyclic CH\(_2\)OH group was indicated (Figure 3).\(^{22}\)
characterize physicochemical properties, such as base pairing, base stacking, and acid–base equilibrium of CR, BR, and AU.

Sugar Puckering. The chemical shifts and coupling constants for the sugar protons of CR, BR, and AU vs those of uridine are shown in Table S1 in the Supporting Information and were used for the determination of ribose puckering.

The chemical shifts for H-1′,2′, and 3′ in CR, BR, and AU show a small degree of de-shielding relative to uridine, while the other three protons (H-4′, S′, and S″) are slightly shielded. This suggests that carbonyls C2/C6 of the cyanurly and barbituryl moieties are above the ribose ring and close to H-4′, S′, and S″ (in the case of AU, an N atom replaces a C6 carbon atom) (Figure 4). This is consistent with the data shown below for the conformation of the exocyclic ribose methylol. The anti-glycosidic angle (−1 to 44° for the N conformer and 39−66° for the S conformer) observed for uridine is needed for removal of steric hindrance of the exocyclic ribose methylol with C2 carbonyl. Here, both the cyanurly and barbituryl moieties possess two carbonyls (C2/C6), and therefore, steric hindrance is removed by rotation of C2/C6 carbonyl above the ribose ring and away from the 5′-hydroxymethylene group. In addition, this conformation is stabilized by an intramolecular H-bond between C2/C6 carbonyl and 5′-OH (Figure 4).

To analyze the population of N and S conformers of uridine mimetics, we have extracted J1′,2′ and J3′,4′ coupling constants from their 1H NMR spectrum at 600 MHz at 300 K (Supporting Information, Table S1). The corresponding data for uridine are included for comparison. Peak assignments were confirmed by obtaining 1H × 1H correlations in COSY spectra.
Since the equilibration rate of the two conformers is fast in the NMR timescale, the observed vicinal couplings are weighted averages of their values in the two conformers. Thus, the mole fractions of conformers S and N were calculated directly from the observed values of $J_{1'3'}$ and $J_{1'5'}$ (see the Supporting Information, Si) and are shown in Table 1.

### Table 1. Conformational Parameters of CR, BR, and AU in D$_2$O Solution

| compound | sugar puckering %N | conformer population around C4′−C5′ bond |
|----------|--------------------|------------------------------------------|
|          |                    | %gg | %tg | %gt |
| CR       | 70                 | 46  | 10  | 44  |
| BR       | 73                 | 46  | 10  | 44  |
| AU       | 60                 | 48  | 14  | 38  |
| U$^{35}$ | 56                 | 58  | 20  | 22  |

Our NMR data are consistent with literature$^{10,25}$ showing that the ribose ring is preferentially in the C3′-endo (N), which constitutes 70% of the total population for CR, 73% for BR, and 60% for 6-azauridine, whereas for the uridines, the value is 56% (Table 1).

**Conformation of 5′-Hydroxymethylene Group (CH$_2$OH).** In general, the nucleoside coupling constants $J_{1'5'}$ and $J_{1'3'}$ can be interpreted in terms of three classical staggered rotamers (gg, tg, and gt) with a preferred gauche–gauche (gg) conformation.$^{26}$ The mole fractions of each staggered rotamer of C4′−C5′ were calculated as described in the Supporting Information (Sii).

The resulting percentages of populations of conformers gg, tg, and gt (Figure 3) calculated for uridine mimetics 1–3 are presented in Table 1. In contrast to uridine, 1–3 do not show preference for the gauche–gauche rotamer. However, major rotamers for 1–3 are both gauche–trans and gauche–gauche rotamers. This is apparently due to H-bonding between the S′-hydroxyl and one of the carbonyls groups of the cyanuryl \(\beta\)-barbituryl moiety and the N6 atom of 3, respectively, possible in those two rotamers. The conformations of the ribose ring of \(\beta\)-barbituryl ribose and \(\beta\)-cyanuryl ribose are similar, 70−73% N due to the steric hindrance between the carbonyl at position 6 and C2′. The percentage of N conformer of AU is lower than those of BR and CR but similar to that of uridine since there is no significant steric hindrance with C2′ position. The major rotamers of both CR and BR exo-cyclic methylol are gg and gt due to H-bonding between the S′-hydroxyl and one of the carbonyls groups of the barbituryl and cyanuryl moiety. AU shows higher preference for gg and gt than uridine. This is apparently due to H-bonding between the S′-hydroxyl and N6 nitrogen atom, possible in those two rotamers (Figure 4).

**Uridine Mimetics 1–3 Preserve Uridine’s H-Bonding Pattern with Adenosine.** The natural base pairing pattern of modified nucleic acids is useful for hybridization-based diagnostics, therapeutics, research, etc.$^{27}$ Therefore, we investigated here the mode of base pairing between adenosine and CR, BR, and AU by $^1$H NMR-monitored titration (Figures 5 and 6). Specifically, 0.1 M CR, BR, and AU in DMSO-$d_6$ were added to 0.1 M adenosine in DMSO-$d_6$ and the NH-3/5 signal of the uridine mimetics, which may be involved in H-bond interactions with adenosine, was inspected (Figure 5).

We observed a decrease in the signals of NH protons of the uracil-mimetic moiety, until they disappeared. This indicates the formation of H-bonding between the uracil-mimetic moiety and adenine moiety. Unexpectedly, de-shielding of the protons of the adenosine’s exocyclic amine was not significant. This is presumably due to the averaging of both signals of the N9-amine protons.

Furthermore, we tested the selectivity of molecular recognition of adenosine vs guanosine by CR, BR, and AU. For this purpose, we have added gradually 0.1 M CR, BR, and AU in DMSO-$d_6$ to 0.1 M guanosine in DMSO-$d_6$ (Figure 5, CR titrations shown in Supporting Information, Figure S5). No change in the base NH proton signal was observed.
Base-Stacking Was Observed for CR but Not for BR and AU. Next, we investigated by NMR the possibility of self-base stacking interactions of the cyanuryl, barbituryl, and 6-azauracil moiety of compounds 1−3 at high concentrations. We monitored the shift of chemical shifts of H5 and H1′ of the ribose proton in AU and BR. In CR, we monitored just the ribose H1′ proton, which indicates stacking interactions between the base moieties, due to the lack of CH group in the cyanuryl moiety.

Table 2 shows the 1H chemical shifts of protons of the sugar moiety of CR and of uridine in dilute solution (0.005 M). These chemical shifts represent the monomeric forms of 1−3 and uridine (δ0). In addition, we determined the asymptotic chemical shifts (δ∞) of the stacked heterocyclic moieties obtained at high concentration (0.4 M).

For uridine, as for other nucleosides, the protons of the sugar moiety are deshielded as the concentration is increased due to base stacking interactions, e.g., positive Δδ values for H5, H6, and H1′.28 This observation is not surprising because in solution, nucleosides assume the anti-conformation with respect to the glycosidic bond.29

Unexpectedly, the data for CR were different. The chemical shifts of the protons in the ribose moiety of CR were shielded as concentration increased, resulting in a negative Δδ with an absolute value similar to that of uridine. This phenomenon may be due to the different stacking interactions of β-cyanuryl vs uracil moieties. Presumably, the cyanuryl moieties form H-bonds (similar to H-bonds in β-sheets of proteins) rather than stacking like aromatic purine and pyrimidines bases. The latter tend to have smaller upfield shifts because their ring currents are smaller than those in purines.30

In a previous report, uridine showed an upfield shift at an elevated concentration28 (Table 2); the same was observed for AU, for the H5 proton, but to a minute degree. There was no change in the chemical shift of AU’s H1′ proton. In contrast to CR, the chemical shifts of the protons in the ribose moiety of BR ribose did not shift at a high concentration. These findings imply that there is no base-stacking in BR and AU. It is noteworthy that
Pyrimidine derivatives are expected to produce smaller upfield shifts than purines because their ring currents are smaller. CR/AU and BR are 100- and 10,000-fold more acidic than U, respectively. We studied the effect of replacement of C5−C6 double bond in uridine by an amide moiety or additional N atom at position C6 in CR, BR, and AU on the uridine’s acid-base equilibria. Specifically, we established the pKₐ values of these analogs by pH titration monitored by ¹³C NMR.

A drastic downfield shift of the signals of carbonyls C2, C6, and C4 of the cyanuryl ring was observed (Figure 7). While the ribose signals only slightly shifted, the C4 chemical shift was highly affected by increasing pH and shifted downfield up to 22 ppm. C2 and C6 signals were shifted by 12 ppm, as expected.

At high pH, downfield shifts of the signals of carbonyls C2, C4, and C6 of BR ring and C2 and C4 of AU were observed, while at low pH, the downfield shifts were moderated (Figure 7).

Figure 7. pH dependence of ¹³C chemical shifts of CR (1), BR (2), and AU (3). (1) 0.9 M CR solution was titrated with diluted HCl and NaOH, and the changes in ¹³C NMR spectra (at 600 MHz) were monitored as a function of pH. (A) Chemical shifts of C2 and C6 in a pH range of 3−9.5. (B) Chemical shifts of C2 and C6 in a pH range of 10−13.1. (C) Chemical shifts of C4 in a pH range of 3−9.5. (D) Chemical shifts of C4 in a pH range of 10−13.1. (2) 0.9 M BR solution was titrated with diluted HCl and NaOH, and the changes in ¹³C NMR spectra (at 600 MHz) were monitored as a function of pH. (A) Chemical shifts of C2 and C6 in a pH range of 2−7. (B) Chemical shifts of C2 and C6 in a pH range of 8−13.26. (C) Chemical shifts of C4 in a pH range of 2−7. (D) Chemical shifts of C4 in a pH range of 8−13.26. (3) 0.9 M AU solution was titrated with diluted HCl and NaOH, and the changes in ¹³C NMR spectra (at 600 MHz) were monitored as a function of pH. (A) Chemical shifts of C2 in a pH range of 2−12.7. (B) Chemical shifts of C4 in a pH range of 2−12.7.
In contrast to CR where the shift of C4 was up to 22 ppm, the signals of C2, C4, and C6 in both BR and AU were affected by increasing pH and shifted downfield 10 ppm on average.

These shifts reflect the electron-rich nature of the deprotonated ring systems and indicate the existence of singly and doubly deprotonated N3 and N5 NH groups in CR, BR, and AU.

In contrast to uridine, CR and BR have two acidic protons. Two inflection points were observed in the sigmoid graph (Figure 7). Calculated pK$_{a1}$ and pK$_{a2}$ values for CR and for BR were 7.24 and 12.73 and 5.37 and 11.95, respectively. One inflection point was observed in the sigmoid graph for AU (Figure 7), and the calculated pK$_a$ was 7.29 vs 9.30 for uridine.$^{28}$

The 100-fold higher acidity of CR vs uridine is due to resonance stabilization of the anion by the oxygen atom on C4 (Supporting Information, Figure S6). Therefore, the signal of C2, C4, and C6 in both BR and AU were a averaged of two peaks of NH obtained as one peak.

Due to its high acidity, BR significantly absorbs light ($\varepsilon$ 14,900 M$^{-1}$ cm$^{-1}$) at pH 5.5. Under pH 12, the BR di-anion strongly absorbs ($\varepsilon$ 46,400 M$^{-1}$ cm$^{-1}$).

**CONCLUSIONS**

Our goal here was to identify uridine mimetics that potentially form more binding interactions with a potential protein partner, or nucleic acid, than those formed with uridine.

Indeed, based on the above NMR studies, we found that CR and BR as well as AU retain specific uridine's H-bonds with adenosine but not guanosine. In particular, CR, BR, and AU keep uridine's natural H-bonding involving N3-H and C4-carbonyl.

Replacement of uridine's C5—C6 double bond by an amide moiety changes dramatically the acid—base character of the molecule. Thus, CR/AU and BR are 100- and 10,000-fold more acidic than uridine, respectively. Namely, under physiological pH, 54% of CR, 51% of AU, and 77% of BR molecules are ionized vs only 13% ionization for uridine, thus indicating potentially tighter binding of those uridine mimetics vs uridine to any protein partner via ionic interactions.

Furthermore, the electron-rich nature of CR and BR vs uridine under physiological pH was reflected by their 13C NMR chemical shifts and $\varepsilon$ values vs that of uridine (13,500 and 16,600 vs 9820 M$^{-1}$ cm$^{-1}$), respectively. Thus, π-interactions with target biomolecules are expected to be significant for CR and BR.

In addition, we studied the conformation of the ribose ring in compounds 1–3 vs uridine. Our findings indicate that all compounds prefer N conformation, up to 73% vs 56% for uridine. Unlike uridine that prefers gg conformation (48%), for compounds 1–3, both major rotamers are gg and gg. Preference for both N conformation and gg and gg rotamers is more significant for 1–3 vs uridine due to the greater probability of H-bonding between the 5'-hydroxyl and C2/C6-carbonyl groups of the heterocyclic moiety. This conformational change of 1–3 vs uridine may also affect the molecular recognition of the former structures by biopolymers.

In conclusion, replacement of uridine's C6 by N or carbonyl, or C5–C6 by an amide, results in significant changes in U's ionization, electron density, conformation, base-stacking, etc., leading to potentially tighter binding than U with a target protein or nucleic acid and potential use for various biochemical and pharmacological applications.

**EXPERIMENTAL SECTION**

Synthesis of $\beta$-Cyanuryl Ribose. Below is an improved procedure for the synthesis of $\beta$-cyanuryl ribose based on literature.$^{36}$ The original procedure involves several steps. We combined them to a one-pot reaction that affords the product in high yield. To cyanuric acid (4) (0.018 mol, 2.30 g, 3 equiv) and 1-O-acetyl-2,3,5-tri-O-benzoyl-$\beta$-D-ribofuranose ($\delta$) (6 mmol, 3
g. 1 equiv) in 50 mL acetonitrile hexamethyldisilazane (HMDS) (0.018 mmol, 3.76 mL, 3 equiv), tetramethylsilyl chloride (TMSCl) (6 mmol, 0.8 mL, 1 equiv) and tetrachlorotriode (10 mmol, 1.2 mL, 2 equiv) were added consecutively. After few minutes, the clear solution became turbid. The mixture was stirred at room temperature for another 1 h. The progress of the reaction was monitored by TLC (ethyl acetate/toluene, 1:4). At the end of the reaction, two spots were observed on the TLC plate (Rf 0.58, Rf 0.17). Then, MeOH was added and the mixture was centrifuged for 10 min. After evaporation, the residue was dissolved in toluene and the organic phase was concentrated under vacuum. The resulting mixture was dissolved in toluene and the organic phase was concentrated and purified by flash column chromatography (toluene/ethyl acetate, 4:1). The fractions containing the product were evaporated, affording CR (7) (2.8 g, 83% yield). 1H NMR (D2O, 400 MHz): δ 9.67 (s, 2H, NH) 8.04–7.87 (3d, 6H, Bz), 7.50–7.20 (m, 9H, Bz), 6.17 (dd, 1H, H-2), 6.08 (t, 1H, H-3), 4.80–4.65 (m, 3H, H-4, H-5) ppm. 13C NMR (100 MHz, CDCl3) 166.6, 165.9, 165.5 (CO), 148.4 (CO) 133.4, 129.6, 128.9, 128.8, 128.5 (C-Ar), 87.4 (C-1), 79.4 (C-4), 74.1 (C-2), 71.2 (C-3), 64.2 (C-5) ppm. HRMS [M + NH4]−: 591.1703 [C29H23N3O10]. 1H NMR spectra of 1 M CR, BR, and AU were determined in aqueous solutions at a pH range of 2–12. At 300 K. 1H NMR spectra were measured in D2O at 600 MHz. The data were collected at 300 K. 1H NMR spectra were measured in D2O at 700 MHz and 300 K on an Bruker Avance – III instrument (700.5 MHz).

UV Measurements. Absorption spectra of CR, BR, AU, and uridine were determined in aqueous solutions at a pH range of 2–12. Samples were measured at room temperature in a 10 mm quartz cell with a 1 cm path length. Absorption spectra were measured on a UV-2401PC UV–VIS recording spectrophotometer (Shimadzu, Kyoto, Japan). Extinction coefficients of those compounds were determined using samples at a concentration range of 10–100 μM.

Evaluation of Base-Pairing of CR, BR, and AU with Purine Nucleosides. CR (14 mg), BR (27 mg), AU (24.5 mg), adenosine (26.7 mg), and guanosine (28.3 mg) in volumetric flasks were stored under vacuum overnight to remove absorbed water. 1H NMR spectra were measured in dry DMSO-d6 at 600 MHz. The data were collected at 300 K. 1H NMR spectra were obtained at a range of concentrations of CR, BR, and AU (0.025, 0.05, 0.1, and 0.15 M) titrated with 0.1 M adenosine in dry DMSO-d6 solution. Adenosine solution (0.1 M, 400 μL) was titrated with 0.1 M compounds 1–3 at ratio 4:1 (adenosine/uridine mimic) until 2:3, respectively. The final volume of the solution in the tube was 1 mL. This protocol was repeated for guanosine.

Base-Stacking Experiments. CR (50 mg), AU (39 mg), and BR (43.5 mg) in volumetric flasks were dried under vacuum overnight. 1H NMR spectra were measured in D2O at 600 MHz. The data were collected at 300 K. 1H NMR spectra were obtained at a range of concentrations of compounds 1–3 (0.003, 0.025, 0.04, 0.05, 0.25, and 0.4 M). NaN3 was added to increase the ionic strength to 0.1 M.

pKα Measurements. Dilute DCl and NaOD solutions were added to 0.1 M CR, BR, and AU in D2O to reach the following pH values: for CR, 3.4, 4.1, 4.4, 4.8, 5.0, 5.3, 5.7, 6.1, 7.6, 8.9, 10.0, 11.0, 11.7, 12.1, 12.7, and 13.1; for BR, 2.32, 2.69, 3.22, 3.54, 4.55, 5.32, 5.51, 5.92, 6.39, 8.34, 9.45, 9.95, 11.49, 12.14, 12.34, 12.59, 12.72, and 12.85; and for AU, 2.32, 2.82, 4.32, 5.52, 6.15, 6.61, 8.37, 9.53, 10.03, 11.48, and 12.29. Apparent pD values were measured with a Hanna instruments pH meter equipped with an electrode. pH is estimated from the pH meter measurement (apparent reading from the pH meter) as 0.41. 13C NMR spectra were measured in D2O at 150 MHz. The data were collected at 300 K. 13C NMR chemical shifts of the bases’ carbonyls were monitored as a function of pH.

The chemical shifts of C2, C4, and C6 were plotted vs pD, and a sigmoid curve was obtained. The pKα values were obtained from the inflection points, which are determined by the second derivative of the fitted sigmoid function using Matlab. A five-parameter sigmoid function was fitted to the data using SigmaPlot.

### ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge at https://pubs.acs.org/doi/10.1021/acsomega.0c04788.

1H and 13C NMR spectra, UV–vis spectra, chemical shifts and coupling constants, equations of population, resonance, and tautomeric forms (PDF)

### AUTHOR INFORMATION

**Corresponding Author**

Bilha Fischer — Department of Chemistry, Bar-Ilan University, Ramat-Gan 52900, Israel; orcid.org/0000-0001-8837-0978; Phone: 972-3-5318303; Email: Bilha.fischer@biu.ac.il; Fax: 972-3-6354907

**Authors**

Helaneh Salameh — Department of Chemistry, Bar-Ilan University, Ramat-Gan 52900, Israel

Michal Afri — Department of Chemistry, Bar-Ilan University, Ramat-Gan 52900, Israel

Hugo E. Gottlieb — Department of Chemistry, Bar-Ilan University, Ramat-Gan 52900, Israel

Complete contact information is available at: https://pubs.acs.org/10.1021/acsomega.0c04788

**Notes**

The authors declare no competing financial interest.

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