Studies on Covalent Adducts of Dehydrouric Acid

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Received November 11, 1993

The structure of 5-chloro and 5-alkoxy-5,7-dihydro-3*H*-purine-2,4,6-triones (**6** and **7–9**) has been corroborated by their spectral properties and X-ray crystallography. The stereochemical model (*R*)-**10** of the key uricolytic intermediate was prepared using menthol as chiral auxiliary. In acidic solutions, depending on the N-substitution, the ring cleavage occurred either at the 4,9-bond **6b** → **13** (R^1 = Me) or at the 3,4-bond **6c** → **14** (R^1 = H). Opening of the 1,6-bond is the dominant process under alkaline conditions. Decarboxylative rearrangement into 1,3-dimethylallantoin (**15**) was specific for N(7)-unsubstituted derivatives. Evidence for intermediacy of the bicyclol tautomeric form was supplied by isolation of 1-menthoxy-2,4-dimethyl-3,7-dioxo-2,4,6,8-tetraazabicyclo[3.3.0]octane (**16**); fragmentation of **10** into 5-menthoxy-imidazolidin-2,4-dione (**17**) also takes place under forcing conditions. Conversely, no allantoin rearrangement was encountered in the $R^7 \neq$H cases; **7d** underwent the pyrimidine ring fragmentation to give 5-methoxy-1-methyl-4-methylimino-imidazolidine-2-one (**18**) or 5-carboxamido-5-methoxy-1-methyl-4-methylimino-imidazolidine-2-one (**19**). A possible mechanism for these ring transformation reactions is discussed.

INTRODUCTION

Quinonoid dehydrouric acid systems **2** and **3** and their derived addition compounds were notable by relative unavailability and transient character.[1,2] In many instances, the suggested structures were proven to be incorrect or at least questionable.[3–5] The

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elusive hydroperoxide **4**, presumably involved in the reaction of uric acid (**1**) with photochemically generated singlet oxygen,[6] mimics the incipient urate-dioxygen interaction catalyzed by uricase. Mechanistically, it is not yet clear what occurs in the crucial act of the uricase reaction, but early indications,[7,8] recently elaborated,[9,10] leave little doubt that the first intermediate observed after the enzymic oxidation of urate is (-)-(S)-5-hydroxy-5,7-dihydro-3H-purine (**5a**).[11] Circumstantial evidence for the transient formation of covalent hydrates **5** was provided by the ultraviolet spectra of the intermediates in electrochemical and peroxidase-mediated oxidations of **1a**[12] and its methylated analogues.[13] Adducts **6**, derived by chlorination of uric acids (**1**), have been employed in the synthesis of the unique example of an *ortho*-quinonoid structure, dehydro-1,3-dimethyluric acid (**2b**).[14] Given the assigned role that covalent adducts play in the interpretation of chemical and enzymic oxidative breakdown of purines, we describe in this communication the synthesis and characterization of related alkoxy derivatives, as suitable models for mechanistic and stereochemical studies.
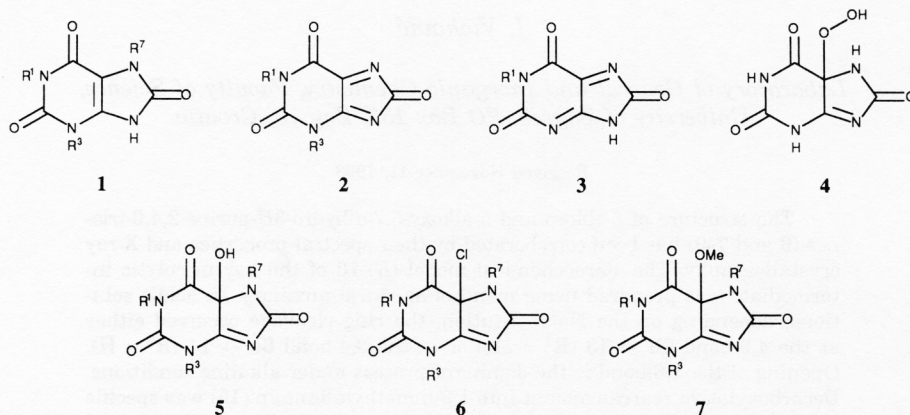
a: R^1= R^3= R^7= H; **b**: R^1= R^3= Me, R^7= H; **c**: R^1= H, R^3= R^7= Me; **d**: R^1= R^3= R^7= Me; **e**: R^1= R^3= Me, R^7= Ac

RESULTS AND DISCUSSION

It seemed desirable first to provide assured assignment of the basic dihydropurinetrione skeleton, which is usually called »isouric acid«.[2,15] The first tentative structural proposal, advanced by Biltz in 1910,[15c] was supported through NMR spectra of the three accessible methoxy homologues **7b–d**. Similarly, structure **6** of a fairly stable chloro derivative, 7-acetyl-5-chloro-1,3-dimethyl-5,7-dihydro-3H-purine-2,4,6-trione (**6e**),[16] was substantiated by NMR spectra, which clearly excluded the salt type structure as a possible alternative (see Experimental). In all other reported cases,[2] the high reactivity and the lack of spectral data or the complexity of chemical transformations make structural assignments of dubious value.

The X-ray crystal structure of 5-methoxy-1,3,7-trimethyl-5,7-dihydro-3H-purine-2,6,8-trione (**7d**) furnished the first direct information about the conformation of a model
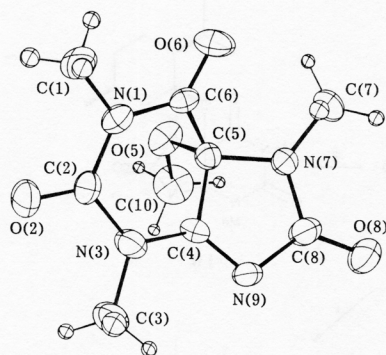
Figure 1. ORTEP view of **7d** (50% probability ellipsoids).

of the key uricolytic intermediate (Figure 1). Unsaturation in the five-membered ring removes flexibility from the bicyclic system, stabilizing a particular conformation with the torsion angles of junction N(3)-C(4)-C(5)-C(6) 44.2° and N(7)-C(5)-C(4)-N(9) −12.1°. A structure nearest 5-sofa conformation of pyrimidine accommodates these demands at the *quasi-trans* fusion.[17] The geometric features of closely related alloxan-like structures constitute a good basis for the analysis of conformational transmissions in the isouric system (5,7-dihydro-3*H*-purine-3,6,8-trione). An X-ray analysis of its alloxan prototype[18] has shown a nearly perfect 5-sofa conformation with torsion angles N(3)-C(4)-C(5)-C(6) 18.7° and O(4)-C(4)-C(5)-O(51) −43.0°, which correspond to the strain-free *quasi-trans* fusion. Thus, the isouric system can be schematically characterized by a large opening of the torsion angle of junction in the pyrimidine ring. The angular substituent [O(5)-C(5)-C(4)-N(9) 109.9(2)° and N(3)-C(4)-C(5)-O(5) −68.8(2)°] causes crowding, which is relieved by adopting a *gauche* orientation N(7)-C(5)-O(5)-C(10) 52.9(2)° and C(4)-C(5)-O(5)-C(10) −59.6(2)°. This geometrical relationship of the sistem must be favourable for bond-breakage in the pyrimidine moiety. Relief of the strain enforced by the geometry is also a powerful driving force in assisting ring modifications and openings in quinonoid purines, which set them apart from the related pterins and flavines.

We have explored the generality of Biltz's synthesis as a source of stereochemical models. The chloro adducts **6** are unstable in solution and react with added nucleophiles or protic solvent in an apparent substitution reaction. Isopropanol causes **6b** to give ether **8**, but only an unchanged compound or its decomposition products are obtained when the same treatment is applied to a tertiary alcohol. The differences for the isopropyl and *tert*-butyl groups reflect the effect of an additional C-substituent; the angular *tert*-butoxyl group experiences strong repulsive interactions that cannot be relieved by rotation around the conjoining bonds. Reactions with secondary alcohols are considerably slower than those in methanol and the use of pyridine as the solvent is dictated by the relative insolubility in alternate media. Reaction of **6b** with optically active alcohols allowed access to diastereomeric ethers. (+)-(*S*)-2-Butanol afforded an unresolvable mixture of (*S,S*)-and (*R,S*)-ethers **9** (<3% de). The incorporation of menthol as a chiral auxiliary showed a distincly higher stereoselectivity

i, *i*-PrOH/Py; ii, (*S*)-2-BuOH/Py; iii, *l*-menthol/Py; iv, EtOH, H⁺
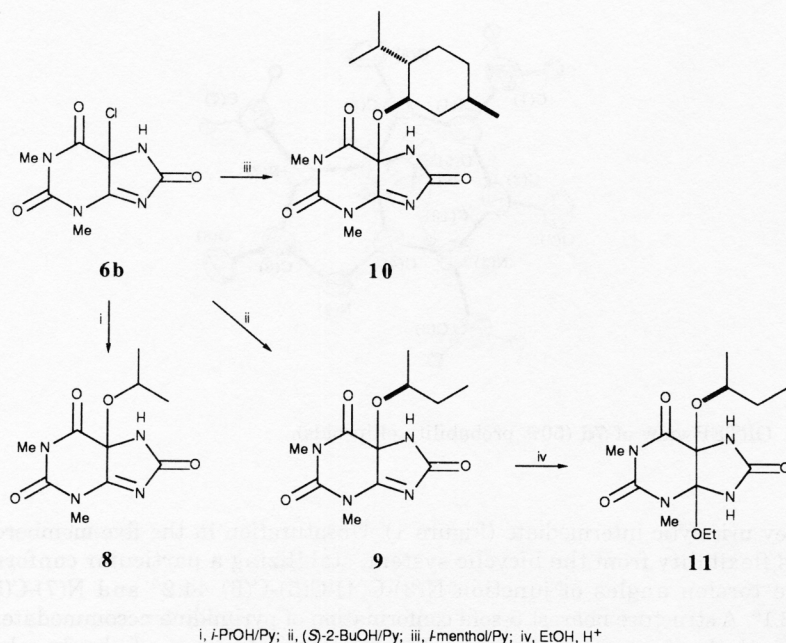
Scheme 1.

(**10**, 20% de), and the menthyl ether (+)-**10**, $[\alpha]_D^{25}$ 63°, was obtained in the pure form by fractional crystallization (Scheme 1). Proof of the stereochemistry of the 5,7-dihydro-3*H*-purine-2,6,8-trione ring system in (+)-**10** rests primarily on the correlation network for the uricase reaction used recently to establish the absolute (*S*)-configuration of the key uricolytic intermediate (-)-**5a**.[11] Comparing the circular dichroism (CD) spectrum of (+)-**10** (Figure 2) with the set of ORD data obtained for the short-lived intermediate in the reaction with uricase (two negative Cotton effects near 275 and 225 nm and one positive Cotton effect near 245 nm, in 0.1 M phosphate, pH 8.0),[8] we propose the absolute (*R*)-configuration of the former. Independent evidence for the absolute configuration of (+)-**10** is currently being explored.

Compounds **8**–**10** cannot be converted into the corresponding dicovalent *sec*-alcoholates. Differences in behaviour between the primary and secondary alcohols towards the formation of 4,5-adducts probably reflect the steric crowding. With ethanol, **9** is observed to undergo conversion into **11**, whereas no such mixed adducts could be detected with **10**, even under apparently favourable circumstances (Scheme 1).

Not unexpectedly, these model compounds were found to undergo facile decomposition and rearrangement processes. Attempts to form a hydroxy analogue **5** with water in various solvents, like the similar reaction with alcohols, resulted solely in formation of ring cleaved products. The first step is almost certainly the formation of a dihydrate **12**. The ring opened product **13**, originally thought to be a dicovalent hydrate **12b**,[15a] was formed from **6b** (R¹ = Me, R⁷ = H). The mode of hydrolytic ring
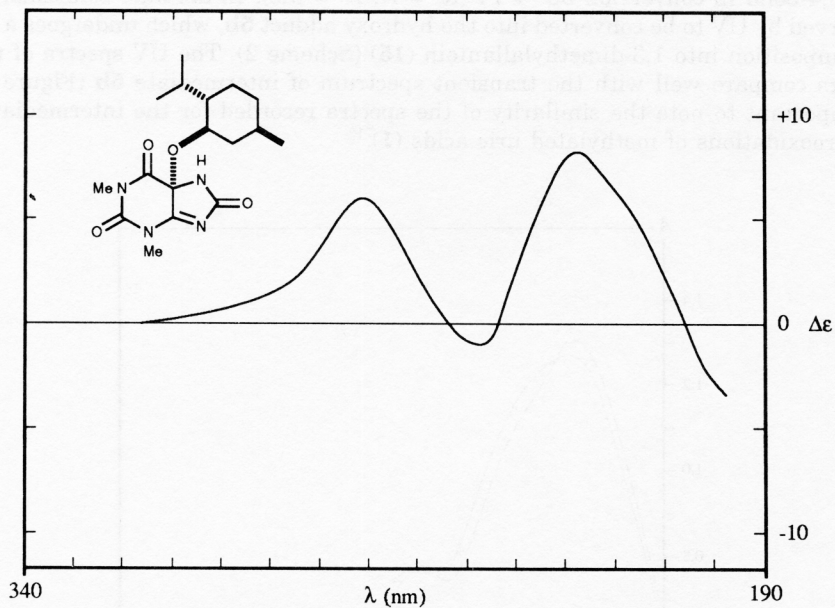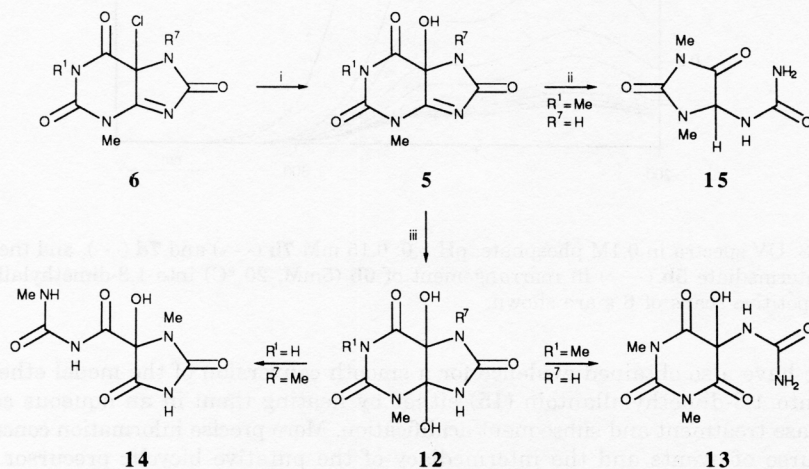
Figure 2. CD spectrum of (+)-**10** in MeOH (0.848 mg/ml); Cotton effects: $\Delta\varepsilon_{272} = +6.0$, $\Delta\varepsilon_{247.5} = -1.0$, and $\Delta\varepsilon_{229} = +8.2$.

Scheme 2.

opening at the amidine carbon, however, switched round completely to cleavage of the 3,4-bond in conversion **6c** → **14** (R^1 = H, R^7 = Me). In alkaline solutions, **6b** is observed by UV to be converted into the hydroxy adduct **5b**, which undergoes a rapid decomposition into 1,3-dimethylallantoin (**15**) (Scheme 2). The UV spectra of model ethers compare well with the transient spectrum of intermediate **5b** (Figure 3); it is important to note the similarity of the spectra recorded for the intermediates in electrooxidations of methylated uric acids (**1**).[13]
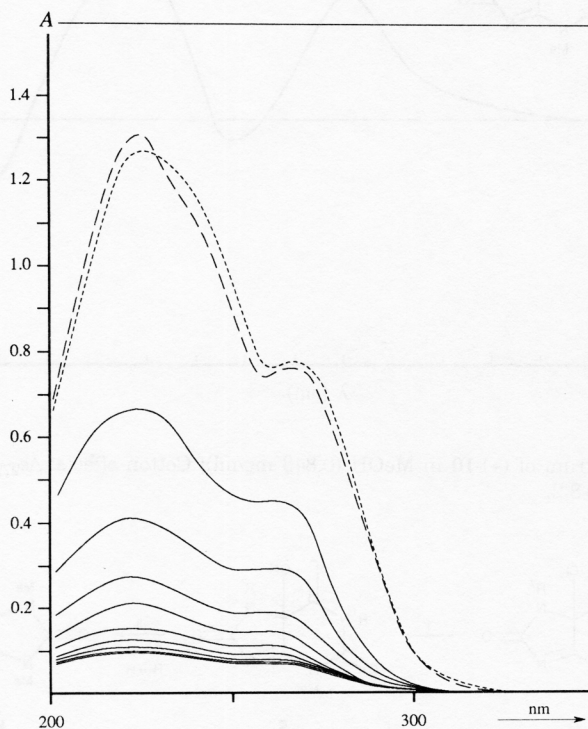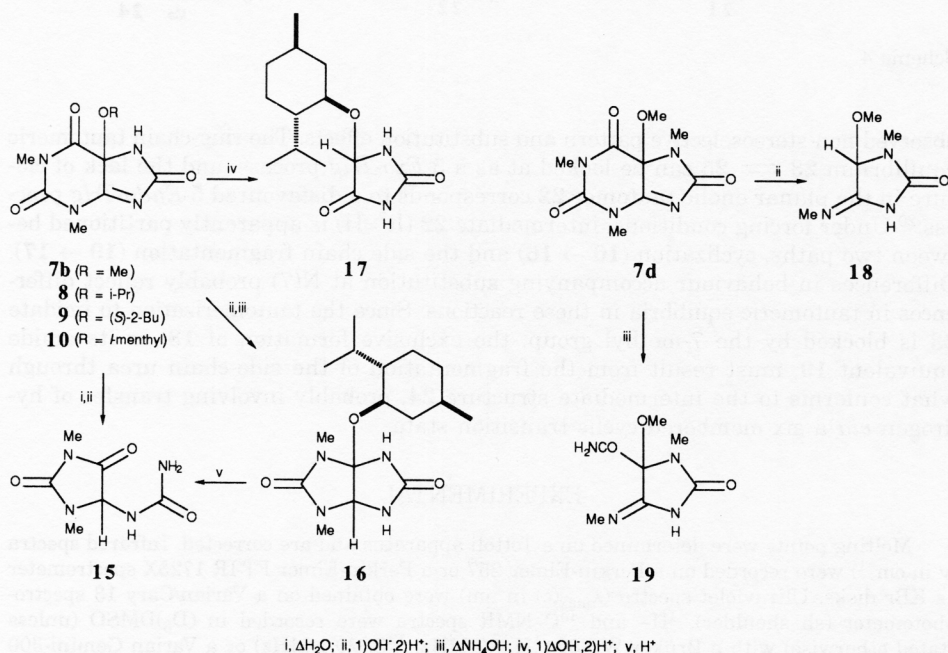

Figure 3. UV spectra in 0.1M phosphate, pH 8.0: 0.15 mM **7b** (- - -) and **7d** (· · ·), and the short-lived intermediate **5b** (——) in rearrangement of **6b** (5mM, 20 °C) into 1,3-dimethylallantoin (**15**); repetitive scans of 6 s are shown.

We have also obtained evidence for a smooth conversion of the model ethers (R^7 = H) into 1,3-dimethylallantoin (**15**) either by heating them in an aqueous solvent or by base treatment and subsequent acidification. More precise information concerning the course of events and the intermediacy of the putative bicyclic precursor of allantoin was obtained from a study of **10**. Under the same reaction conditions, the diastereomeric mixture of bicyclol ether **16** (38% de) can be isolated and shown to be a real intermediate by subsequent acid hydrolysis into the normal product **15**.[19] If, however, the reaction is carried out at elevated temperature, then it is found to yield a mixture of **16** and **17**, a product of decarboxylation and simple hydrolytic deg-

radation of the pyrimidine ring. Exposure of **10** to forcing conditions with an excess of base furnished a diastereomeric mixture of **17** (30% de) in a 35% yield (Scheme 3). Identical results obtained for conversions of (+)-**10** into **16** and/or **17** indicate non-stereoselective reaction pathways.

In contrast to N(7) unsubstituted cases, substituted analogues (R^7 = Me) could not be caused to rearrange to the corresponding allantoins. An interesting transformation has been observed with **7d** under the conditions employed for rearrangement of **7b**. Not only is there no evidence for formation of an allantoin or its bicyclic precursor, but the decarboxylation product **18** obtained shows clearly that the fission of the pyrimidine ring has occurred at the 1,6- and 2,3-bonds. A rather similar situation arises with **7d** in boiling ammoniacal solution, where the corresponding amide **19** was produced (Scheme 3). Since the absolute configuration of closely related alloxanic acid amide is known,[11] we considered whether it would be feasible to cleave ammoniolytically the 1,6-bond of (+)-**10**. Much to our surprise, boiling in ammoniacal solution turned out to be the method of choice for the conversion **10 → 16**.

7b (R = Me)
8 (R = i-Pr)
9 (R = (S)-2-Bu)
10 (R = l-menthyl)

i, ΔH₂O; ii, 1)OH⁻,2)H⁺; iii, ΔNH₄OH; iv, 1)ΔOH⁻,2)H⁺; v, H⁺

Scheme 3.

Uricolysis is a complex reaction with considerable variations, depending on the reagent, substrate and reaction conditions employed. A feature of these model reactions is the ease with which 5,7-dihydro-3H-purine-2,6,8-trione system undergoes the base-catalyzed ring opening at the 1,6-position. This is relatively straightforward with a multi-step mechanism involving the formation and decomposition of a tetrahedral intermediate **20** and smooth decarboxylation of **21** under acidic conditions. The resultant tautomeric structures **22 ⇌ 23 ⇌ 24** (Scheme 4) nicely account for the
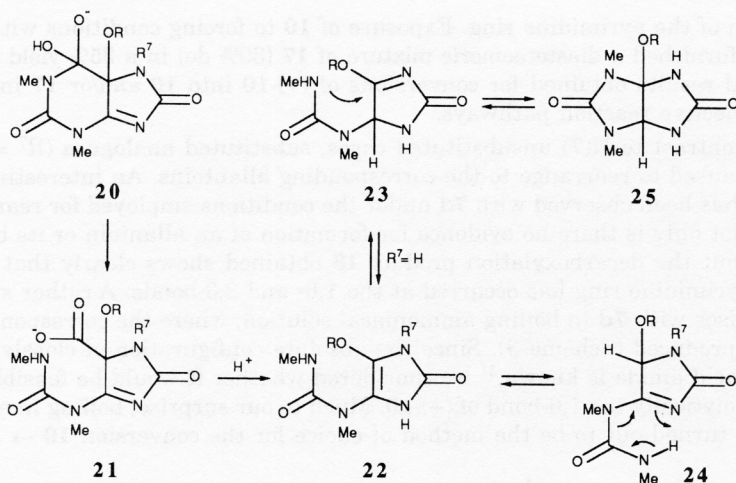
Scheme 4

observed non-stereoselective pattern and substitution effects. The ring-chain tautomeric equilibrium **23** ⇌ **25** can be looked at as a 5-*Exo-Trig* process, and the lack of closure in the planar enolic tautomer **22** corresponds to a disfavoured 5-*Endo-Trig* process.[20] Under forcing conditions, intermediate **22** (R[7]=H) is apparently partitioned between two paths, cyclization (**10 → 16**) and the side chain fragmentation (**10 → 17**). Differences in behaviour accompanying substitution at N(7) probably reflect differences in tautomeric equilibria in these reactions. Since the tautomerization to imidate **23** is blocked by the 7-methyl group, the exclusive formation of **18**, or its amide equivalent **19**, must result from the fragmentation of the side-chain urea through what conforms to the intermediate structure **24**, probably involving transfer of hydrogen *via* a six-membered cyclic transition state.

EXPERIMENTAL

Melting points were determined on a Tottoli apparatus and are corrected. Infrared spectra (v in cm^{-1}) were recorded on a Perkin-Elmer 257 or a Perkin-Elmer FT-IR 1725X spectrometer as KBr disks. Ultraviolet spectra ($\lambda_{max}(\varepsilon)$ in nm) were obtained on a Varian/Cary 13 spectrophotometer (sh shoulder). ^{1}H– and ^{13}C–NMR spectra were recorded in (D$_6$)DMSO (unless stated otherwise) with a Bruker WP-80 (^{1}H, 80 MHz; ^{13}C, 20.1 MHz) or a Varian Gemini-300 (^{1}H, 300 MHz; ^{13}C, 75 MHz) instrument. Chemical shifts are given in δ (ppm) from an internal TMS standard and coupling constants are expressed in Hz (br broad, s singlet, d doublet, t triplet, q quartet, m multiplet, umc unresolved multiplet centre); multiplicities in ^{13}C spectra, where listed, were determined by off-resonance decoupling. CD spectra were obtained with a Jasco J40CS spectropolarimeter (0.2 mm cells). TLC: silica gel 60F$_{254}$ plates (Merck); detection with UV light (unless stated otherwise). (*S*)-2-Butanol, b.p. 97–99°, $[\alpha]_D^{25}$ 13.4 (neat) was resolved according to literature procedure.[21]

5-Chloro-5,7-dihydro-3H-purine-2,6,8-triones (**6**).

The title compounds were prepared by chlorination of the corresponding xanthines or uric acids in acetic acid or chloroform according to Biltz's procedures:[15,16]

*5-Chloro-1,3-dimethyl-5.7-dihydro-*3H-*purine-2,6,8-trione (6b):* plates (81%), m.p. 226 °C dec (Ref.15a 225 °C dec); IR 3380 (NH), 1760, 1690 (CO), 1625–1605 (C=N).

*5-Chloro-3,7-dimethyl-5,7-dihydro-*3H-*purine-2,6,8-trione (6c):* very hygroscopic plates, containing HOAc of crystallization (84%), m.p. 168 °C dec (Ref. 15b. 168–9 °C dec).

*5-Chloro-1,3,7-trimethyl-5,7-dihydro-*3H-*purine-2,6,8-trione (6d)*: needles (88%), m.p. 158–159 °C dec (Ref. 15c 158 °C dec.). IR 1745, 1700 (CO), 1620–1605 (C=N).

*7-Acetyl-5-chloro-1,3-dimethyl-5,7-dihydro-*3H-*purine-2,6,8-trione (6e):* microscopic prisms (83%), m.p. 162–163 °C dec (Ref. 16 162 °C dec). IR 1781, 1775, 1767, 1740, 1720 (CO), 1630–1620 (C=N). ^{1}H–NMR, δ 3.32 (s, NMe), 3.11 (s, NMe), 2.45 (s, Me); ^{13}C–NMR, δ 176.4 (s, C-4), 168.0 (s, CO), 163.3 (s, C-6), 161.5 (s, C-8), 150.6 (s, C-2), 83.0 (s, C-5), 31.5 (q, NMe), 29.3 (q,NMe), 25.1 (q, Me).

5-Methoxy-5,7-dihydro-3H-purine-2,6,8-triones (7).

Title compounds were prepared according to Biltz's procedure[15] by reaction of the 5-chloro adducts **6** with dry methanol in the presence of pyridine (10%).

*5-Methoxy-1,3-dimethyl-5,7-dihydro-*3H-*purine-2,6,8-trione (7b):* plates from dry MeOH (71%), m.p. 206–207 °C dec (Ref. 15a 207 °C dec). UV (MeOH) 222 (8150), 270sh (4650). IR 3250, 3210, 3120 (NH), 1755, 1710 (C=O), 1625 (C=N). ^{1}H–NMR, δ 9.49 (s, 1H, NH), 3.41 (s, 3H, OMe), 3.29 (s, 3H, NMe), 3.11 (s, 3H, NMe). ^{13}C–NMR, δ 175.0 (s, C-4), 165.3 (s, C-6), 164.2 (s, C-8), 150.7 (s, C-2), 84.1 (s, C-5), 51.6 (q, OMe), 31.0 (q, NMe), 28.5 (q, NMe).

*5-Methoxy-3,7-dimethyl-5,7-dihydro-*3H-*purine-2,6,8-trione (7c):* prisms from dry MeOH (64%), m.p. 205–206 °C dec (Ref. 15b 205–206 °C dec). UV (MeOH) 220 (8250), 270sh (4500). IR 3240, 3135 (NH), 1763, 1735, 1713 (CO), 1620 (C=N). ^{1}H–NMR, δ 11.8 (s, 1H, NH), 3.39 (s, 3H, OMe), 3.36 (s, 3H, NMe), 3.08 (s, 3H, NMe). ^{13}C–NMR, δ 174.3 (s, C-4), 164.6 (s, C-6), 163.9 (s, C-8), 149.9 (s, C-2) 84.9 (s, C-5), 51.3 (q, OMe), 30.3 (q, NMe), 26.1 (q, NMe).

*5-Methoxy-1,3,7-trimethyl-5,7-dihydro-*3H-*purine-2,6,8-trione (7d)*: prisms from MeOH (75%), m.p. 204–205 °C dec (Ref. 15c 205 °C dec). UV (MeOH) 221 (8100), 270sh (4600); IR 1745, 1702, 1694 (CO), 1615 (C=N). ^{1}H–NMR, δ 3.37 (s, 3H, OMe), 3.30 (s, 3H, NMe), 3.10 (s, 3H, NMe), 3.06 (s, 3H, NMe). ^{13}C–NMR, δ 172.9 (s, C-4), 164.1 (s, C-6), 163.6 (s, C-8), 150.1 (s, C-2), 84.4 (s, C-5), 51.1 (q, OMe), 30.9 (q, NMe) 28.3 (q, NMe), 25.9 (q, NMe).

5-Isopropoxy-1,3-dimethyl-5,7-dihydro-3H-purine-2,6,8-trione (8).

To a stirred mixture of dry isopropanol (6 ml) and pyridine (1.5 ml), **6b** (1.5 g, 0.0065 mol) was added gradually. After being stirred for 30 min, the crystalline product was filtered off, washed with dry ether and recrystallized twice from dioxane/light petroleum to yield **8** (1.1 g, 66%) as prismatic crystals, m.p. 199–200 °C dec. UV (MeOH) 222 (8400), 270sh (4300). IR 3450, 3210, 3110 (NH), 1751, 1708, 1635 (C=O), 1612 (C=N). ^{1}H–NMR, δ 9.49 (s, 1H, NH), 3.75 (septet, 1H, CH, J = 6.1), 3.30 (s, 3H, NMe), 3.11 (s, 3H, NMe), 1.05 (d, 3H, Me, J = 6.1), 1.01 (d, 3H, Me, J = 6.1). ^{13}C–NMR, δ 174.9 (s, C-4), 165.5 (s, C-6), 164.2 (s, C-8), 150.7 (s, C-2), 84.0 (s, C-5), 67.7 (d, OCH). 31.2 (q, NMe), 28.5 (q, NMe), 23.3 (q, Me), 23.2 (q, Me). *Anal.* Calcd. for $C_{10}H_{14}N_4O_4$ (M_r=254.25): C 47.24, H 5.55, N 22.04; found: C 46.97, H 5.82, N 21.88%.

5-(S)-2-Butoxy-1,3-dimethyl-5,7-dihydro-3H-purine-2.6.8-trione (9).

To a mixture of (+)-*S*-2-butanol[21] (0.5 ml) and dry pyridine (1.6 ml, 50–60 °C) **6b** (1.15 g, 0.005 mol) was added gradually. After stirring for 10 min at 60 °C, the mixture was filtered, the filtrate diluted with dry benzene (5 ml), and the tarry product removed by filtration. The clear solution was concentrated *in vacuo* and precipitated with light petroleum to yield a pale yellow product. Two recrystallizations from dioxane/ether/light petroleum afforded a mixture of diastereomers **9** (0.63–0.69 g, 55–60%) as colourless tiny needles, m.p. 141–144 °C. R_f 0.40 (benzene/acetone 3:1). All attempts to separate diastereomers were unsuccessful. UV(MeOH) 222 (8350), 270sh (4400). IR 3374 (NH), 1768, 1749, 1693, 1688 (C=O), 1622 (C=N). ^{1}H–NMR

(CDCl$_3$), δ 6.69, 6.60 (s, 1H, NH),. 3.58, 3.57 (umc, 1H, CH), 3.52, 3.51 (s, 3H, NMe), 3.32 (s, 3H, NMe), 1.44, 1.41 (umc, 2H, CH$_2$), 1.13, 1.09 (d, 3H, Me, *J* = 7.0), 0.84, 0.81 (t, 3H, Me, *J* = 7.0). ^{13}C–NMR (CDCl$_3$) δ 175.3, 174.9 (C-4), 165.1 (C-8), 163.7, 163.5 (C-6), 150.2 (C-2), 84.1, 83.9 (C-5), 74.4, 73.5 (OCH), 31.7 (NMe), 30.1 (CH$_2$), 29.2 (NMe), 21.6, 20.9 (Me), 9.6 (Me).

Anal. Calcd. for C$_{11}$H$_{16}$N$_4$O$_4$ (*M*$_r$ = 268.28): C 49.25, H 6.01, N 20.89; found C 48.91, H 6.33, N 20.56%.

5-Menthoxy-1,3-dimethyl-5,7-dihydro-3H-purine-2,6,8-trione (10).

Powdered **6b** (10 g, 0.043 mol) was gradually added to a stirred solution of *l*-menthol (7.0 g, 0.045 mol) in dry pyridine (25 ml, 50–60 °C). After 30 min, the insoluble residue was filtered off and washed with dry benzene (100 ml). A yellowish tarry product that separates from the combined filtrates was removed by cautions addition of light petroleum and filtration. The crude product was recrystallized twice from dioxane/ether (1:1)/light petroleum to give a diastereomeric mixture of **10** (20% de), as fine hairlike needles in an overall 60% yield; m.p. 179–182°C; TLC *R*$_f$ 0.25 (chloroform/acetone 9:1). UV (EtOH) 222 (10100), 245 sh (4800), 268 (3000). IR 3354 (NH), 1769, 1753, 1693 (C=O), 1620 (C=N). ^{1}H–NMR (CDCl$_3$) δ 6.88, 6.78 (s, 1H, NH), 3.50 (s, 3H, NMe), 3.40 (umc, 1H, CH), 3.31 (s, 3H, NMe),2.0–1.2 (m, 5H, CH, CH$_2$), 1.10 (umc, 4H, CH$_2$), 0.87 (d, 6H, CHMe$_2$, *J* = 6.5), 0.72, 0.68 (d, 3H, Me, *J* = 7.0). ^{13}C–NMR (CDCl$_3$) δ 175,6, 174.5 (C-4) 164.5, 164.7 (C-8), 163.3, 164.0 (C-6), 150.0, 150.1 (C-2), 83.7, 83.9 (C-5), 75.4, 77.0 (C-1'), 47.8 (C-2'), 42.4, 43.4 (C-6'), 33.6 (C-4'), 31.8, 32.0 (C-5'), 31.5 (NMe), 29.2 (NMe), 25.1, 25.3 (CH), 22.9 (C-3'), 22.1 (Me), 21.2 (Me), 15.8 (Me).

Fractional crystallization from dioxane/ether/light petroleum afforded the pure (*R*)-diastereomer **10** as needles, m.p. 211–212 °C, [α]$_D^{25}$ 63.0 (c 2.2, MeOH). IR 3362 (NH), 1765, 1750 sh, 1686 (C=O), 1617 (C=N). ^{1}H–NMR (CDCl$_3$) δ 6.80 (s, 1H, NH), 3.50 (s, 3H, NMe), 3.40 (umc, 1H, CH), 3.31 (s, 3H, NMe), 2.0–1.2 (m, 5H, CH, CH$_2$), 1.10 (umc, 4H, CH$_2$), 0.87 (d, 6H, CHMe$_2$, *J* = 6.5), 0.70 (d, 3H, Me, *J* = 7.0). ^{13}C–NMR (CDCl$_3$), δ 175.6 (C-4), 164.5 (C-8), 163.3 (C-6), 150.0 (C-2), 83.7 (C-5), 75.4 (C-1'), 47.8 (C-2'), 42.4 (C-6'), 33.6 (C-4'), 31.8 (C-5'), 31.5 (NMe), 29.2 (NMe), 25.1 (CH), 22.9 (C-3'), 22.1 (Me), 21.2 (Me), 15.8 (Me).

Anal. Calcd. for C$_{17}$H$_{26}$N$_4$O$_4$ (*M*$_r$ = 350.43): C 58.27, H 7.48, N 15.99; found: C 58.02, H 7.70, N 15.74%.

5-(S)-2-Butoxy-4-ethoxy-1,3-dimethyl-tetrahydro-purine-2,6,8-trione (11).

The title compound was obtained on attempted crystallization of **9** from ethanolic solutions containing traces of mineral acids. Recrystallization from ethanol gave a diastereomeric mixture of **11** (60–70%), as fine needles; m.p. 165–167 °C. IR 3215, 3150 (NH), 1737, 1720, 1675 (CO). ^{1}H–NMR, δ 8.73 (s, 1H, NH), 8.70 (s, 1H, NH), 4.23, 4.21 (s, 1H, CH), 3.40, 3.38 (q, 2H, CH$_2$, *J* = 7.1), 3.07 (s, 3H, NMe), 2.85 (s, 3H, NMe), 1.50 (umc, 2H, CH$_2$), 1.11 (umc, 6H, Me), 0.84 (t, 3H, Me, *J* = 7.0). ^{13}C–NMR, δ 165.5 (C-6), 156.7 (C-8), 150.1 (C-2), 97.5 (C-4), 83.0, 82.9 (C-5), 71.4 (OCH), 60.1 (OCH$_2$), 30.2, 29.7 (CH$_2$), 28.6 (NMe), 27.8 (NMe), 20.9, 20.2 (Me), 14.6 (Me), 9.0 (Me).

Anal. Calcd. for C$_{13}$H$_{22}$N$_4$O$_5$ (*M*$_r$ = 314.35): C 49.67, H 7.05, N 17.82; found: C 49.40, H 7.30, N 17.69%.

Ring openings of 6 at the amidine carbon.

5-Hydroxy-1,3-dimethyl-5-ureido-pyrimidine-2,4,6-trione (**13**). The initial crystallization from a solution of **6b** (2.3 g, 0.01 mol) in water (5 ml) was induced by scratching the inside of the vessel with a glass rod. Recrystallization from water afforded **13** (1.6 g, 75%), as plates, m.p. 183–184 °C dec, identical with the compound (Ref. 15a 183 °C dec) originally formulated as *4,5-dihydroxy-1,3-dimethyl-tetrahydro-purine-2,6,8-trione* (**12b**); its traditional name *1,3-dimethyluric acid glycol* was devised by Biltz.[1] IR 3490 (OH), 3398, 3308 (NH$_2$), 3200 (NH), 1708, 1694, 1667 (CO). ^{1}H–NMR, δ 7.85 (s, 1H, NH), 7.11 (s, 1H, OH), 5.78 (br, 2H, NH$_2$), 3.12 (s, 6H, NMe). ^{13}C–NMR, δ 168.5 (s, C-4,6), 157.4 (s, CO), 150.3 (s, C-2), 76.4 (s, C-5), 28.5 (q, NMe).

1-(4-Hydroxy-3-methyl-2,5-dioxo-imidazolidine-4-carbonyl)-3-methyl-urea (**14**). Treatment of **6c** × HOAc (2.9 g) with water or NaOH solutions (5 ml, final pH 1–8) afforded a crystalline product, which was identical in all respects with an authentic sample of **14**.[5] Recrystallization from water gave **14**, as prisms, in 40–55% overall yields; m.p. 203 °C dec (Ref. 5 m.p. 203–204 °C dec).

1,3-Dimethylallantoin (15).

Method 1. A solution of ethers **7b, 8** or **9** (0.02 mol) in aqueous dioxane (10 ml, 50%) was heated under reflux for 30 min. The solvent was removed *in vacuo* and the product recrystallized from dry ethanol to yield **15** (2.2–2.6 g, 60–70%).

Method 2. A solution of either **6b** or **7b, 8** and **9** (0.02 mol) in NaOH (4 ml, 30%, 0 °C) was acidified with glacial HOAc (3 ml). After being left overnight at +4 °C, the precipitate was collected, and then in turns redissolved in hot EtOH (50 ml), filtered, and the bulk of the NaOAc removed by careful addition of ether (*ca.* 50 ml). An additional amount of ether was added to the clear filtrate and the precipitate was recrystallized twice from water. Recrystallization from ethanol gave colourless prisms of 1,3-dimethylallantoin (**15**, 0.92–1.49 g, 25–40%), m.p. 214 °C (Ref. 19 m.p. 214–215 °C).

An alternative involves reaction of **6b** (5 mg) in 0.1M phopsphate buffer (1 ml, pH 8); **15** was identified by TLC: R_f 0.66 (BuOH/HOAc/H$_2$O 2:1:1); it also gave a characteristic yellow spot by spraying with a mixture (9:1) of antipyrine (16 g/30 ml H$_2$SO$_4$/5 ml H$_3$PO$_4$/60 ml H$_2$O) and dimethylglyoxime (0.1 g/10 ml EtOH) and heating at 130 °C for 30 min.

1-Menthoxy-2,4-dimethyl-3,7-dioxo-2,4,6,8-tetraazabicyclo[3.3.0]octane (16).

Method 1. Ether **10** (0.35 g, 0.01 mol) was dissolved in NaOH (5 ml, 20%, 60 °C) and acidified with acetic acid. The mixture was left for 2 hrs at 4 °C and the crude product was collected, washed with water, and dried to yield **16** (0.2 g, 62%).

Method 2. Ether **10** (0.7 g, 0.02 mol) was heated (100 °C) with concentrated ammonia (2 ml) in a sealed tube for 30 min. The crystalline precipitate which separated on cooling was collected, washed with water, and dried to yield **16** (0.6 g, 92%).

Identical results were obtained for conversions of pure diastereomer (+)-**10**. Fractional crystallization from ethanol of both preparations (*ca.* 38% de, as determined by NMR analysis of crude products) gave pure diastereomers: (*R,R*)-**16**, m.p. 277–278 °C (Ref. 19 277–278 °C) and (*S,S*)-**16**, m.p. 213–214 °C ((Ref. 19 213–214 °C).

5-Menthoxy-imidazolidin-2,4-dione (17).

A solution of **10** (0.7 g, 0.01 mol) in hot NaOH (2 ml, 40%, 90 °C) was stirred for 40 min and acidified with HOAc. After standing at 4 °C for 24 hrs, the mixture was filtered and extracted with ether (5 x 10 ml). Dried extract (Na$_2$SO$_4$) was concentrated to 5 ml, and the product precipitated with light petroleum. Recrystallization from ether/light petroleum gave a diastereomeric mixture of **17** (0.18 g, 35%), as fine needles, m.p. 173–177 °C; the same diastereomeric mixture was obtained from (+)-**10**. R_f 0.21 (chloroform/acetone 9:1, 5% KMnO$_4$ spray. IR 3251, 3075 (NH), 2955, 2924, 2873 (CH), 1784, 1727 (CO). ^{1}H–NMR (30% de) δ 8.49 (br, 1H, NH), 6.53, 6.34 (s, 1H, NH), 5.22, 5.14 (d, 1H, CH, J = 1.5, 1.0), 3.73 (umc, 1H, OCH), 1.8–1.4 (m, 5H, CH, CH$_2$) 1.15 (umc, 4H, CH$_2$), 0.94, 0.91 (d, 6H, CHMe$_2$, J = 6.5, 7.0), 0.77, 0.81 (d, 3H, CHMe, J = 6.8). ^{13}C–NMR, δ 170.8 (C-4), 156.3 (C-2), 81.8, 80.4 (C-5), 75.8, 77.0 (OCH), 47.9, 48.1 (C-2'), 41.4, 41.2 (C-6'), 34.1 (C-4'), 31.5 (C-5') 25.3 (CH), 23.1 (C-3') 22.2 (Me), 20.9 (Me), 16.0 (Me).

Anal. Calcd. for C$_{13}$H$_{22}$N$_2$O$_3$ (M_r = 254.33): C 61.39, H 8.72, N 11.01; found C 61.10, H 8.97, N 10.86%

5-Methoxy-1-methyl-4-methylimino-imidazolidin-2-one (18).

Finely powdered **7d** (2,4 g, 0.01 mol) was dissolved in NaOH (5 ml, 30%) at 0°C. After standing for 48 hrs the solution was carefully neutralized with cold 10% HCl and evaporated to dry-

ness *in vacuo* (<50 °C). The residue was repeatedly extracted with hot benzene (5 x 10 ml). Combined extracts were dried (Na_2SO_4), and concentrated until the crystals separated. Two recrystallizations from benzene gave **18** (1.1 g, 70%), as tiny prisms, m.p. 123 °C. IR 3203 (NH), 2940, 2875 (CH), 1716 (CO), 1629 (C = N). ^{1}H–NMR, δ 8.50 (br, 1H, NH), 5.43 (s, 1H, CH), 3.17 (s, 3H, OMe), 3.11 (br s, 3H, = NMe), 2.93 (s, 3H, NMe). ^{13}C–NMR, δ 173.7 (s, C-4) 166.8 (s, C-2), 86.9 (d, C-5), 50.0 (q, OMe), 28.4 (q, =NMe), 26.7 (q, NMe).

*Anal.*Calcd. for $C_6H_{11}N_3O_2$ (M_r = 157.18): C 45.85, H 7.05, N 26.74; found C 45.61, H 7.31, N 26.47%

*5-Carboxamido-5-methoxy-1-methyl-4-methylimino-imidazolidin-2-one (**19**).*

Finely powdered **7d** (2.4 g, 0.01 mol) and concentrated NH_4OH (5 ml) were heated in a sealed tube (110 °C, 40 min). The product which separated on cooling was recrystallized from water to give **19** (0.9, 45%), as flat prisms, m.p. 253–254 °C dec. IR 3470, 3400 (NH_2, 3200 (NH), 1726, 1702 (CO), 1630 (C = N). ^{1}H–NMR, δ 8.43 (br, 1H, NH), 7.88 (br, 1H, $CONH_2$) 7.53 (br, 1H, $CONH_2$), 3.08 (s, 3H, OMe), 2.88 (br s, 3H, =NMe), 2.63 (s, 3H, NMe).

Anal. Calcd. for $C_7H_{12}N_4O_3$ (M_r = 200.20): C 42.00, H 6.04, N 27.99; found C 41.75, H 6.27, N 27.73%.

*X-ray Crystal Structure Analysis of **7d**.*

A prism shaped crystal of **7d** (0.25 x 0.35 x 0.45 mm), prepared by slow crystallization from methanol, was used for single-crystal X-ray analysis.

Crystal data: $C_9H_{12}N_4O_4$, M_r = 240.22, monoclinic, space group $P2_1/n$, a = 14.792(5), b = 7.488(3), c = 9.947(4) Å, β = 98.67(4)° (from least squares fitting of setting angles for 16 reflections 5 ≤ θ ≤ 10°), V = 1089.2(7), Z = 4, D_x = 1.465 gcm^{-3}, μ(Mo $K\alpha$) = 1.10 cm^{-1}, $F(000)$ = 504, room temperature. Data were collected on a Philips PW 1100 diffractometer in θ−2θ scan mode, 6 < 2θ ≤ 60° (0 ≤ h ≤ 20, 0 ≤ k ≤ 10, −13 ≤ l ≤ 13); 2527 reflections were measured, of which 2315 were observed ($I ≥ 3\sigma I$). Data were corrected for Lorentz and polarization effects (locally written program) but not for absorption; 12 reflections (3 0-3. 4 0-2, 1 2-2, 2 2-2, 3 0-1, 2 1-1, 2 2-1, 4 0 0, 0 1 1, 0 2 1, 0 0 2, 4 0 2) were ignored because of secondary extinction effects. No significant variation in intensity of 3 check reflections was observed. The structure was solved by direct methods (MULTAN80).[22] Full matrix least-squares refinement of positional and anisotropic thermal parameters for all non-hydrogen atoms was continued until convergence, the hydrogen atom coordinates were located in a ΔF map (calculated for those attached to carbon and were as observed for those on oxygen and nitrogen). At convergence R = 0.041, R_w = 0.060 for 166 parameters, w = $1/\sigma^2(F_o)$, $\Sigma w(|F_o|-|F_c|)^2$ minimized, S = 2.0; $(\Delta/\sigma)_{max}$ = 0.58, $(\Delta/\sigma)_{av}$ = 0.16 for non-H atoms; maximum peak height in final ΔF map $\Delta\rho_{max}$ = 0.32eÅ^{-3}. Atomic scattering factors and anomalous dispersion coefficients were as defined by XRAY76.[23] The molecule (Figure 1) was drawn with ORTEPII.[24]

Supplementary material available:

Lists of atomic coordinates, anisotropic temperature factors, bond distances, angles, torsion angles, observed and calculated structure factors of **7d** (total 24 pages).

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SAŽETAK

Proučavanje kovalentnih adukata dehidromokraćne kiseline

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Struktura 5-kloro i 5-alkoksi-5,7-dihidro-3*H*-purin-2,4,6-triona (**6** i **7–9**) potvrđena je spektroskopskim i kristalografskim metodama. Stereokemijski model (*R*)-**10** ključnoga urikolitičkog intermedijara pripravljen je s pomoću mentola kao kiralnog reagensa. U kiselim otopinama, ovisno o N-supstituciji, prsten se otvara ili u 4,9-položaju **6b** → **13** (R¹ = Me), ili u 3,4-položaju **6c** → **14** (R¹ = H). Otvaranje u položaju 1,6 dominantan je proces u alkalnim okolnostima. Dekarboksilativni premještaj u 1,3-dimetilalantoin (**15**) svojstven je samo N(7)-nesubsitutiranim derivatima. Izolacija 1-mentoksi-2,4-dimetil-3,7-diokso-2,4,6,8-tetraazabiciklo[3.3.0]oktana (**16**) pokazuje da u premještaju sudjeluje biciklolski tautomerni oblik; fragmentacijom **10** u žešćim okolnostima nastaje i 5-mentoksi-imidazolidin-2,4-dion (**17**). Međutim, do alantoinskoga premje-

štaja ne dolazi u slučaju kad je $R^7 \neq H$; tako **7d** fragmentacijom pirimidinskoga prstena daje 5-metoksi-1-metil-4-metilimino-imidazolidin-2-on (**18**) ili 5-karboksamido-5-metoksi-1-metil-4-metilimino-imidazolidin-2-on (**19**). Razmotren je vjerojatni mehanizam tih reakcija pretvorbe prstena.