# BROMINE, *N*-BROMOSUCCINIMIDE AND SULPHUR INDUCED ISOMERIZATIONS IN THE HOPANE SERIES

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Abstract. When reacted with bromine, *N*-bromosuccinimide or molten sulphur, triterpenoids of the 17β(H),21β(H)-hopane series were converted into their 17α(H),21β(H) and 17β(H),21α(H) isomers of geochemical significance.

Saturated hydrocarbons from the hopane series frequently possess in sediments the "geohopane" skeletons **1a** and to a lesser extent **2a** of respective 17α(H),21β(H) and 17β(H),21α(H) configurations[1] differing thus from the thermodynamically less stable 17β(H),21β(H) "biohopane" framework **3a** which characterizes hopanoids from living organisms.[2] To account for these diagenetically-induced isomerizations, laboratory simulations by heating of **3a** alone or in hopanoid mixtures with clays, shales or kerogen, as originally performed on steroids,[3] proved disappointing as they gave only traces of **1a** of doubtful origin.[4,5] Heating experiments with sulphur, a widespread constituent of sediments, appeared to us a new promising approach as sulphur-induced isomerizations of acyclic isoprenoid hydrocarbons as well as steranes into some of their geochemical analogues have been reported.[6] More generally motivated by the action of chemicals, even not present in sediments, on the biohopane skeleton **3**,[7] we describe here experiments with bromine and *N*-bromosuccinimide (NBS), both reportedly capable of oxidizing and/or isomerizing saturated triterpenic hydrocarbons,[8] as well as with sulphur.

a: R=H; b: R=CH₂CH₂OAc
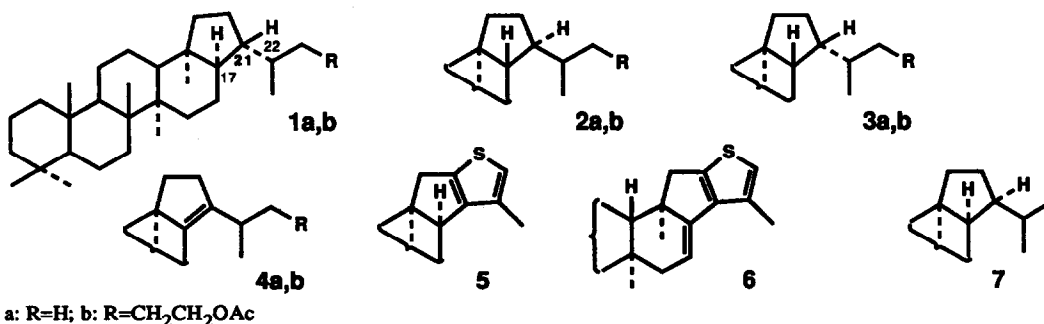
When a CCl_4 solution of hopanoid **3b** was heated to reflux for 5 h in the presence of NBS, in addition to more polar major (80 %) products, the following mixture with the same R_f as the starting material was characterized by GC-MS : **1b** (32%), **2b** (5%), **3b** (12%), **4b** (40%) and unknowns (11%). A similar series but containing less unsaturated compound was obtained in 30% yield after reflux for 30 min with bromine: **1b** (67%), **2b** (14%), **3b** (1%), **4b** (15%) and unknowns (3%). Although, as already reported, both reactions involve very probably a free radical mechanism,[8] the details of the epimerization pathway yielding to **1b** and **2b** are not known. Only in the case of the formation of the 17(21) unsaturation can one reasonably postulate the existence of 17- or/and 21-bromo intermediates as exemplified previously in the case of friedelane.[8a] Prompted by these encouraging results, we switched over with sulphur as a source of other possibly homolytic reactions. Having checked that refluxing in sulphur-saturated CCl_4 did not affect hopanoid **3b**, we eliminated the solvent in favour of heating experiments at a higher temperature in liquid sulphur (Table 1).

Table 1. Action of sulphur on hopanoids.[a]

Entry	Compound	Hopanoid/ Sulphur mass(mg)	T (°C)	Time(h)	Apolar compound yield(%)	Composition (%)
1	**3a**	12/50	240	3	50	1a(74.6), 2a(17.5), 3a(5.7) 4a(1.0), 5(0.4), 6(0.3), unknowns(0.5)
2	**3a**	12/50	175	96	95	1a(0.2), 2a(1.5), 3a(98.3)
3	**3a**	2/20	215	14	60	1a(71.0), 2a(15.0), 3a(12.5), 4a(0.3), 5(0.5), 6(0.2), unknowns(0.5)
4	**3b**	9/45	240	3	50	1b(72.0)[b], 2b(23.0)[b], 3a(4.0), unknowns (1.0)
5	**3b**	9/45	250	4.4	20	1b(71.0)[b], 2b(24.0)[b], 3b(3.0),unknowns (2.0)
6	**1a**	2/20	240	3	50	1a(98.0), 4a(1.0), 7(1.0)
7	**2a**	2/20	240	3	50	2a('98.5), 3a(1.5)

[a] A typical procedure is as follows: the hopanoid (purity over 98%, GC) was mixed with a large excess of powdered sulphur and transferred into a Pyrex glass vial. After purging with argon, the system was closed and heated as indicated. After cooling and opening, $CHCl_3$ was added to the dark brown reaction mixture that smelled H_2S. Sulphur was removed from the chloroform solution by passage through a small column of activated copper,[9] and the apolar colourless compounds were separated from more polar brown material by column chromatography on silica gel using cyclohexane as eluent. [b] Hopanoids **1b** and **2b** are here isomeric mixtures at C-22, the 22(R)-isomer being the major one (Entry 4: 96%, Entry 5: 85%)

In the case of 17β(H),21β(H)-hopane **3a**, the GC-MS analysis of the apolar fraction shows the disappearance of the starting material in favour of the two major isomers **1a** and **2a** and of three minor compounds **4a, 5** and **6** which were isolated by TLC on silver nitrate impregnated silica gel using cyclohexane-toluene (95:5, v/v) as eluent.[10]

When heated in liquid sulphur under the same conditions, acetate **3b** yielded the same proportion of isomeric compounds (Table 1), including in this case a partial isomerization at the asymmetric C-22 carbon atom, mimicking the known diagenetic isomerization at this center.[1a] Higher heating temperatures did not significantly change the relative amounts of apolar compounds, apart from an epimerization increase at C-22 (Table1, Entry 5) but did decrease the yield remarkably. When treated at 240°C (Table1, Entries 6 and 7), isomeric hopanes **1a** and **2a** appeared unchanged in the apolar fraction with the exception of minute amount of hop-17(21)-ene **4a** detected (GC) in the case of **2a**. The hopanoids were however recovered in 50% yield only, the rest belonging to a $CHCl_3$ soluble brown fraction.

These results match the recent thermal simulations performed with sulphur on long-chain hydrocarbons and on cholestane as both thiophene containing molecules and isomerized compounds were found.[6c,11] The proposed contribution of free radicals in these transformations supported by the detection of free radicals in sulphur heated over 172°C[12] seems relevant in our case. Indeed, the action of sulphur on hopane **3a** was effective but very slow at 175°C (Table 1, Entry 2): **3a** was recovered almost unchanged even after four days. Only a trace amount of hop-17(21)-ene **4a** and a slight odour of H_2S revealed the beginning of the reaction.

Although not representative of the diagenetic conditions, even in the case of sulphur,[13] all these reactions exhibit the remarkable regioselectivity of the hopane **3a** skeleton activation and allow a direct and probably radical initiated access to epimeric skeletons **1a** and **2a** of geochemical significance. Furthermore, in the experiments with sulphur, minor hopanoids **4-6** interestingly represent potential geochemical markers as both $\Delta^{17(21)}$ unsaturated[14,15] and ring E sulphur-functionalized[6c,16] hopanoids have been reported in sediments. Hop-17(21)-ene **4a** probably plays the role of an intermediate in the formation of the polar brown material (Table 1) and to a lesser extent of the sulphur containing derivatives **6** and **7** as in preliminary experiments, heating of this hopene with sulphur at 220°C (20min) yielded the two latter compounds as major sulphur derivatives, illustrating the greater reactivity of alkenes.[6c] Our attention is presently focused on further experiments with sulphur in order to find more geomimetic conditions and to study other hopanoids.

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10 Compounds **1a**, **1b**, **2a**, **4a** and **4b** were identified by direct comparison (^{1}H-NMR (400MHz) and/or GC, GC-MS) with reference compounds synthesized in the laboratory. **2b** was identified by interpretation of the characteristic moretane fragmentation pattern in mass spectrometry; A. Van Dorsselaer, *Dr. Sc. Thesis*, Université Louis Pasteur, Strasbourg, France. The structures of the thiophene derivatives **5** and **6** were tentatively assigned on the basis of the ^{1}H-NMR and MS spectra. Compound **5**, subjected to Raney nickel desulphurization followed by 10% carbon supported palladium catalytic hydrogenation,[14] gave a mixture of **1a** and **7** in the (4:1) ratio (GC). Comparison of the (250MHz) ^{1}H-NMR spectrum of the resulting mixture with published data supported 17α(H) assignment of configuration; R. E. Corbett, and C. K. Heng, *J. Chem. Soc. (C)*, 1971, 1885. *Selected spectroscopic data* with 400MHz ^{1}H NMR. spectra in CDCl$_3$ and MS measurements at 70eV. **Compound 5**: NMR, δ 0.784 (3H, s), 0.791 (3H, s), 0.799 (3H, s), 0.843 (3H, s), 1.069 (3H, s), 1.167 (3H, s) 2.163 (3H, s), 2.52 (1H, d, J=15Hz), 2.67 (1H, t, J=6Hz), 2.75 (1H, d, J=15Hz), 6.69 (1H, broad s); M.S., 438 (M$^+$, 100%), 423 (9%), 300 (4%), 246 (15%), 232 (32%), 217 (36%), 205 (8%), 203 (8%), 191 (71%), 177 (9%), 175 (10%), 163 (40%). **Compound 6**: NMR, δ 0.807 (3H, s), 0.831 (3H, s), 0.867 (3H, s), 0.933 (3H, s), 1.139 (3H, s), 1.170(3H, s), 1.79 (1H, dd, J=3Hz & 19Hz), 2.01 (1H, dd, J=3Hz & 13Hz), 2.27 (3H, d, J=1Hz), 2.32 (1H, dd, J=3Hz & 19Hz), 6.73 (1H, q, J=1Hz); M.S., 436 (M$^+$, 100%), 421 (11%), 298 (3%), 243 (10%), 230 (58%), 217 (20%), 215 (44%), 201 (10%), 191 (9%), 189 (4%), 185 (5%), 176 (20%), 162 (12%).

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