Electronic Supplementary Material (ESI) for Organic & Biomolecular Chemistry

Concise and Efficient Syntheses of $preQ_1$ base, Q base, and *(ent)*-Q Base

Supporting Information

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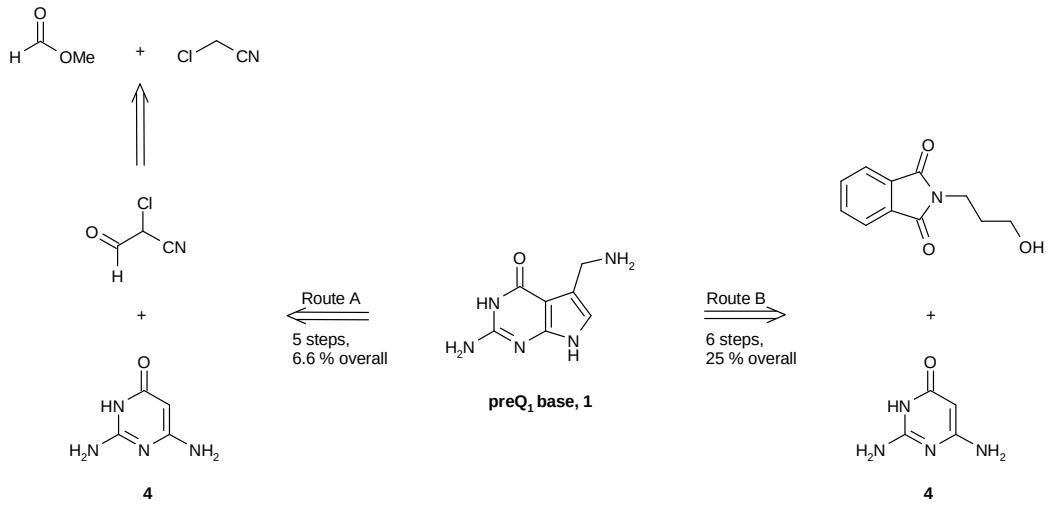
Overview and brief discussion of the most recently published procedures for preQ$_1$ by Carell et al.: [1]

The two most recent procedures suggested to access the modified guanine analogue preQ$_1$ base were reported by *Carell et al.* in 2005, both starting from pyrimidine **4** (Scheme S1).[1] The first one (Route A), being a short and seemingly straightforward five-step synthesis, involves intermediate formation of preQ$_0$ base from pyrimidine **4** and 2-chloro-3-oxopropanenitrile, followed by a subsequent hydrogenation step to yield preQ$_1$ base. However, this approach only yields 6.6 % of the desired nucleobase. The second route represents a higher-yielding, yet synthetically slightly more elaborate six-step sequence rendering preQ$_1$ base in 25 % overall yield. After an intermediate phthalimide deprotection step with hydrazine, the resulting product mixture is reprotected by treatment with $(Boc)_2O$ in DMF to enable separation of the product from the simultaneously formed side-products (mainly Phthalhydrazide) by flash chromatography yielding 31 % of Boc-protected preQ$_1$ base. Final deprotection gives rise to the desired nucleobase **1**.

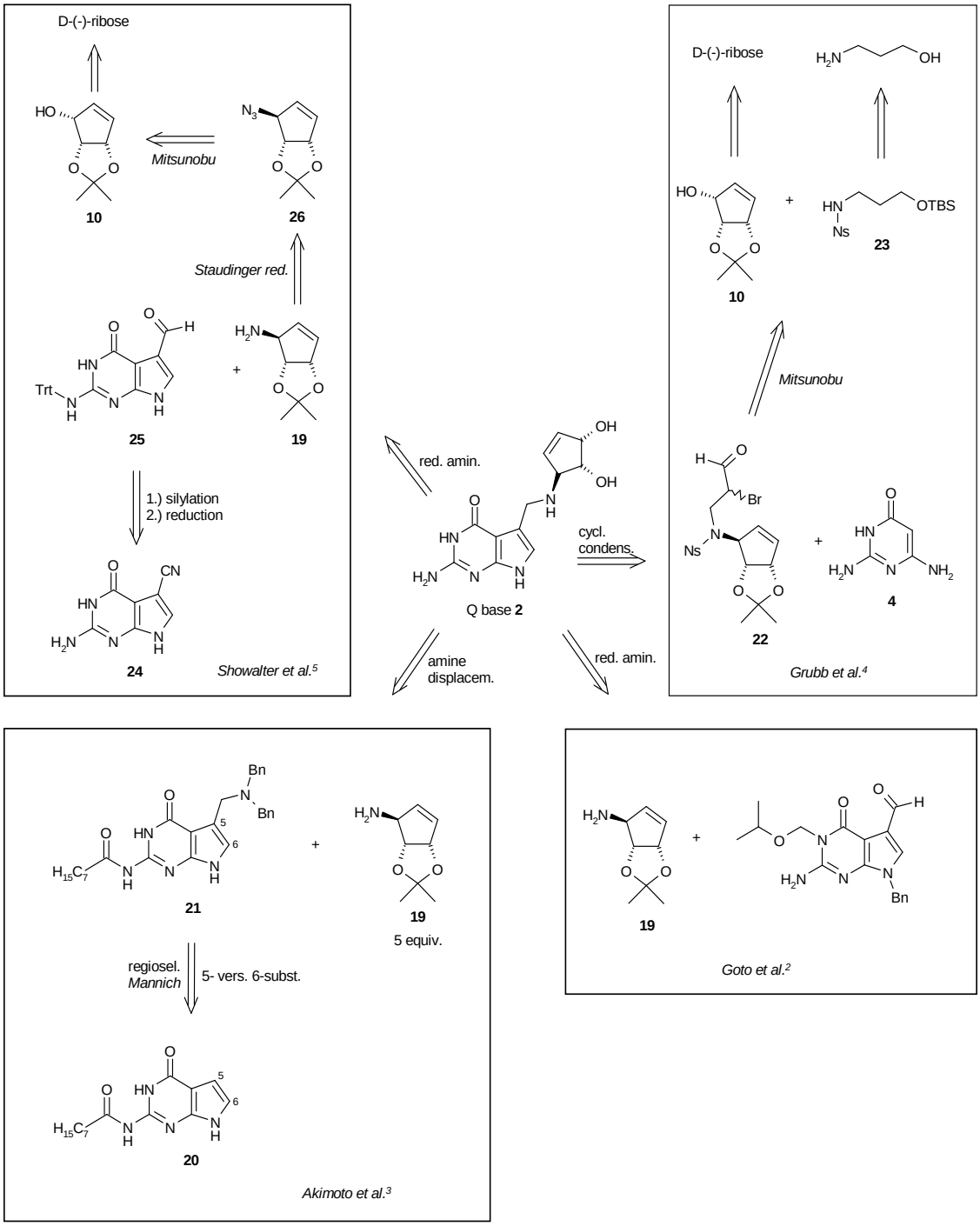


Scheme S1: Retrosynthetic approaches of most recent preQ$_1$ syntheses by *Carell et al.*[1]

Overview and brief discussion of all hitherto published synthetic approaches for Q base:

The first synthesis was reported by *Goto et al.*[2] in 1983. This rather lengthy and time-consuming route finally rendered Q base in 19 steps applying a reductive amination with cyclopentenylamin **19** as key step. In 1988, *Akimoto et al.*[3] published a shorter, apparently more straightforward synthetic approach. The crucial step of their strategy is based on a regioselective *Mannich reaction* at the pyrrolo moiety of the previously prepared octanoyl-protected heterocyclic core **20** to introduce a dibenzylated aminomethyl side chain at position 5 of the heterocycle. The resulting intermediate **21** finally allows implementation of the required side chain moiety of queuine by an amine exchange reaction utilizing excessive amounts of the above-mentioned amine **19**. Although the regioselectivity of the *Mannich reaction* (ratio of 5- versus 6-position substitution = 14.2 : 1) is quite high, the obtained product still might contain low amounts of the unwanted 6-substituted isomer. In addition, the synthesis of the required substrate **19** for the following amine exchange is rather challenging and, moreover, has to be applied in large excess (five-fold) in order to achieve a reasonable yield. In 2000, *Grubb et al.*[4] described an alternative route utilizing a different disconnection approach. Herein, the key reaction is the ring closure via a cyclic condensation to install the pyrrolopyrimidine core of the nucleobase

employing 2,6-diaminopyrimidine-4-one (**4**) and α-bromoaldehyde intermediate **22**, prior to this derived via cyclopentenol **10** starting from D-(-)-ribose, and the fully protected 3-aminopropanol precursor **23**.



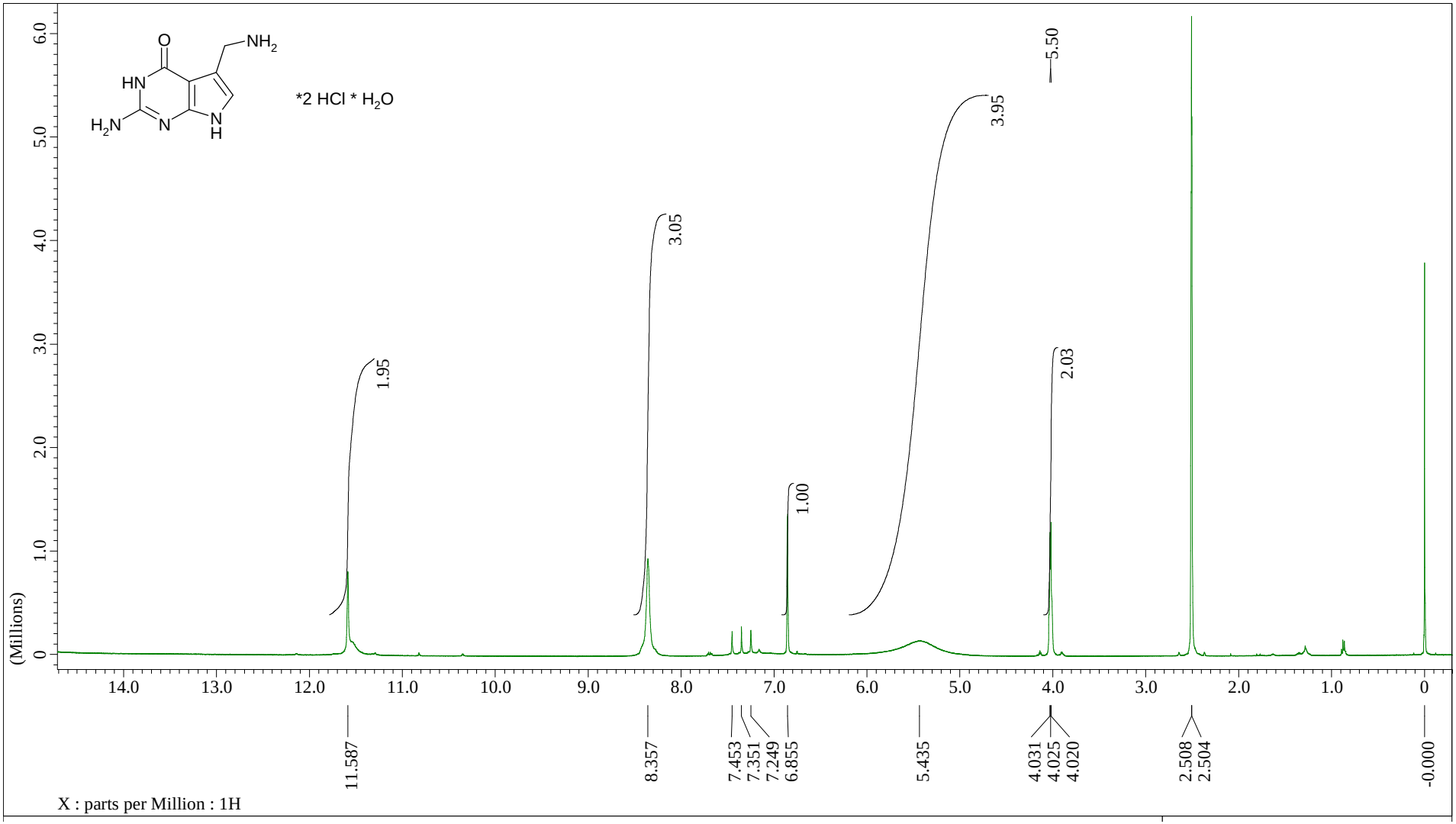
Scheme S2: Retrosynthetic approach and key steps of previously reported syntheses of Q base

In total, this sequence requires 14 steps in a convergent approach applying several protection strategies to finally yield Q base (**2**) in an overall yield of 1.6 % from D-(-)-ribose. In 2010, *Showalter et al.*[5] reported a significantly shorter strategy using the biochemical $preQ_1$ precursor $preQ_0$ base (**24**), available in two preceding steps from heterocycle **4**,[6] as starting material. Reductive amination of the trityl-protected formyl deazaguanine derivative **25**, obtained from $preQ_0$ base **24**, with cyclopentenylamine **19**, furnishes, after a

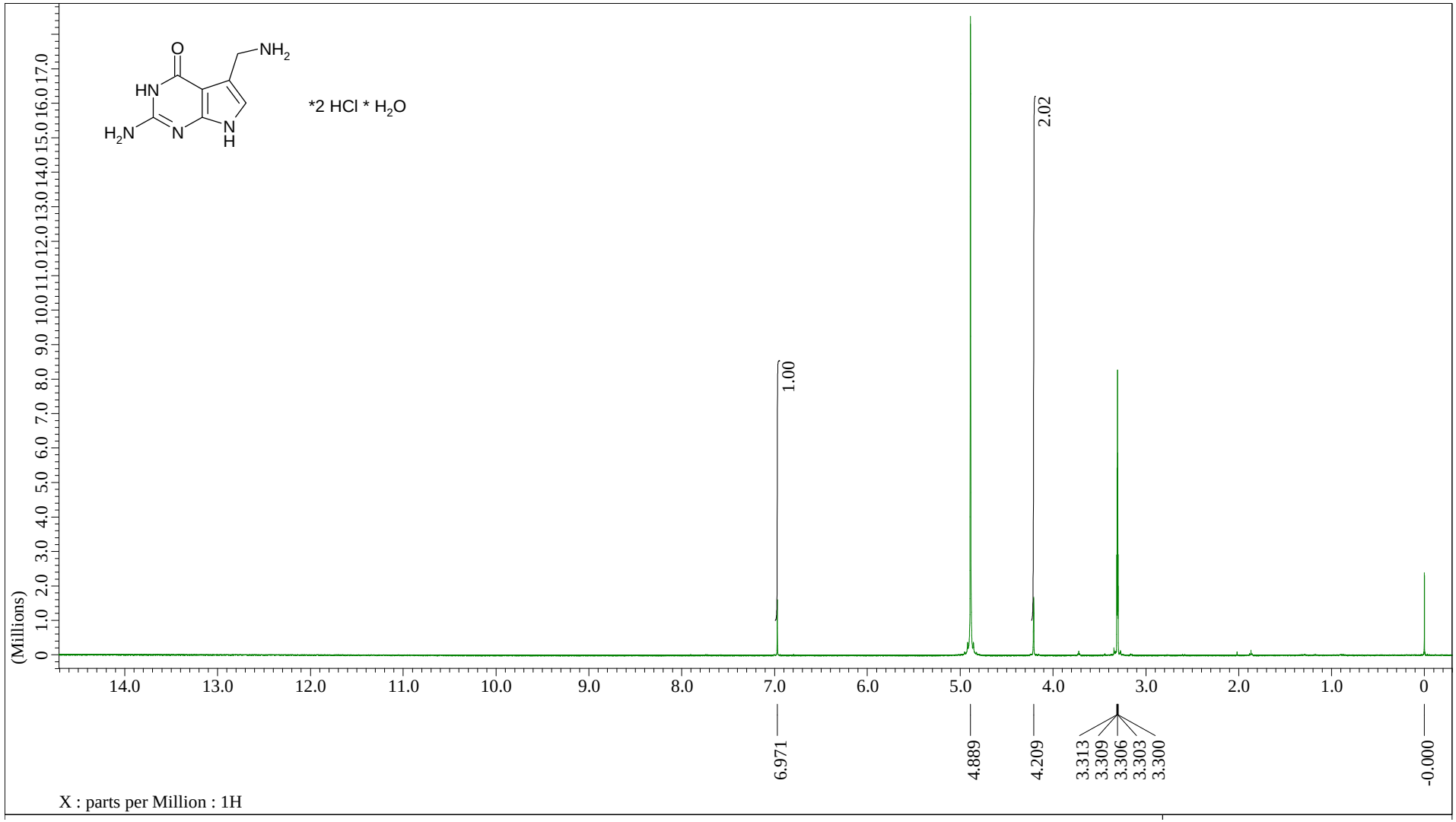
final deprotection step, queuine (**2**) as monohydrochloride in 36 % over four steps from **24**. Cyclopentenylamine **19** is accessible via the corresponding cyclopentenylazide **26** by a modified four-step protocol of *Carell et al.*[7] in 14.5 % overall yield. However, a necessary requirement of this synthesis is the implementation of an intermediate silylation step with **24** thus gaining sufficient solubility of the heterocyclic intermediate for the successive nitrile reduction with DIBAL-H to form the trityl-protected derivative **25** at lower temperatures.

1. F. Klepper, K. Polborn and T. Carell, *Helv. Chim. Acta*, 2005, **88**, 2610-2616.
2. T. Kondo, T. Ohgi and T. Goto, *Chem. Lett.*, 1983, **12**, 419-422.
3. H. Akimoto, E. Imamiya, T. Hitaka, H. Nomura and S. Nishimura, *J. Chem. Soc., Perkin Trans. 1*, 1988, 1637-1644.
4. C. J. Barnett and L. M. Grubb, *Tetrahedron*, 2000, **56**, 9221-9225.
5. A. F. Brooks, G. A. Garcia and H. D. H. Showalter, *Tetrahedron Lett.*, 2010, **51**, 4163-4165.
6. M. T. Migawa, J. M. Hinkley, G. C. Hoops and L. B. Townsend, *Synth. Commun.*, 1996, **26**, 3317-3322.
7. F. Klepper, E.-M. Jahn, V. Hickmann and T. Carell, *Angew. Chem. Int. Ed.*, 2007, **46**, 2325-2327.

2-Amino-5-(aminomethyl)-3,7-dihydro-4*H*-pyrrolo[2,3-*d*]pyrimidin-4-one dihydrochloride hydrate (1, DMSO-d_6)



2-Amino-5-(aminomethyl)-3,7-dihydro-4*H*-pyrrolo[2,3-*d*]pyrimidin-4-one dihydrochloride hydrate (1, CD_3OD)



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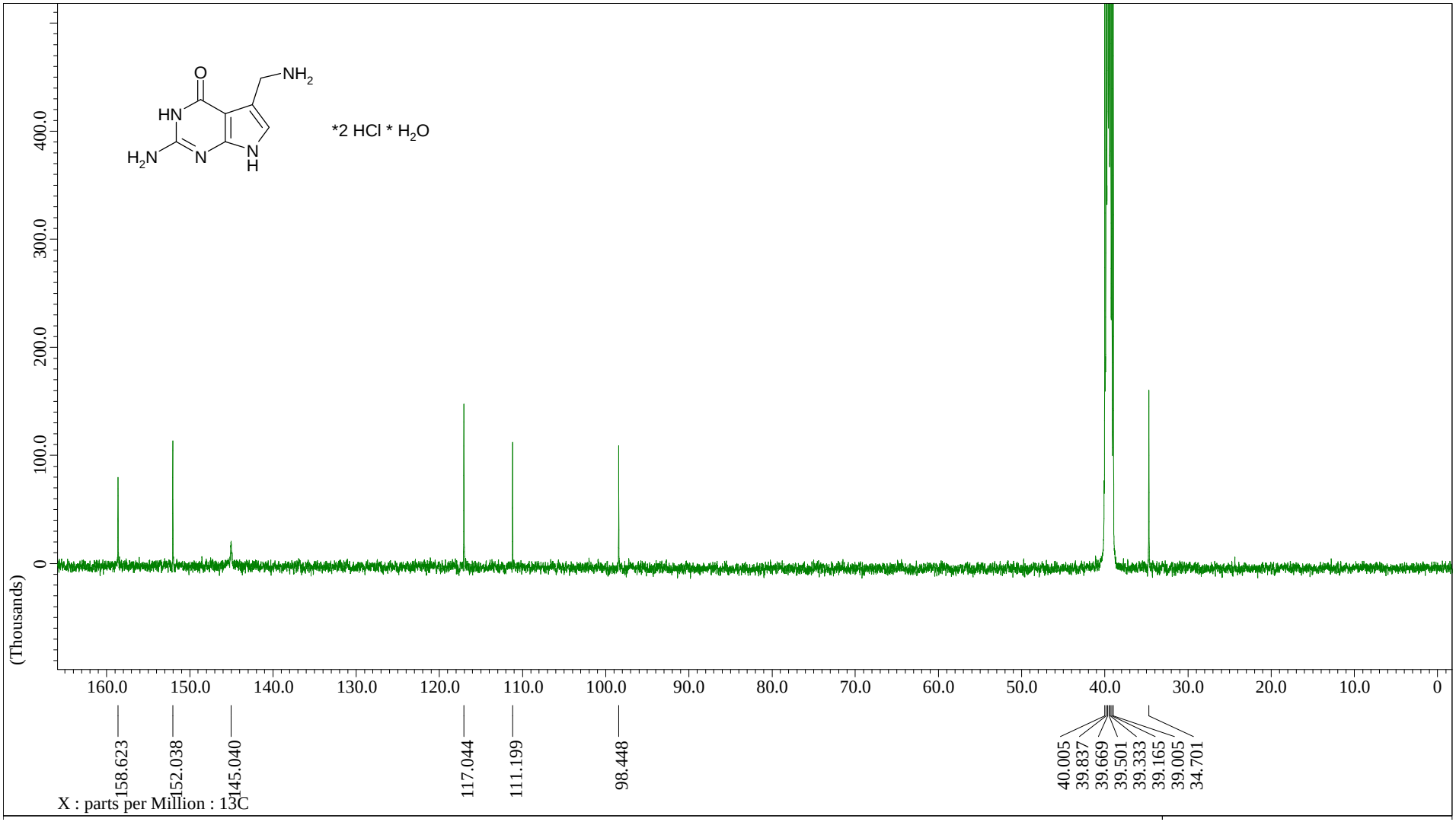
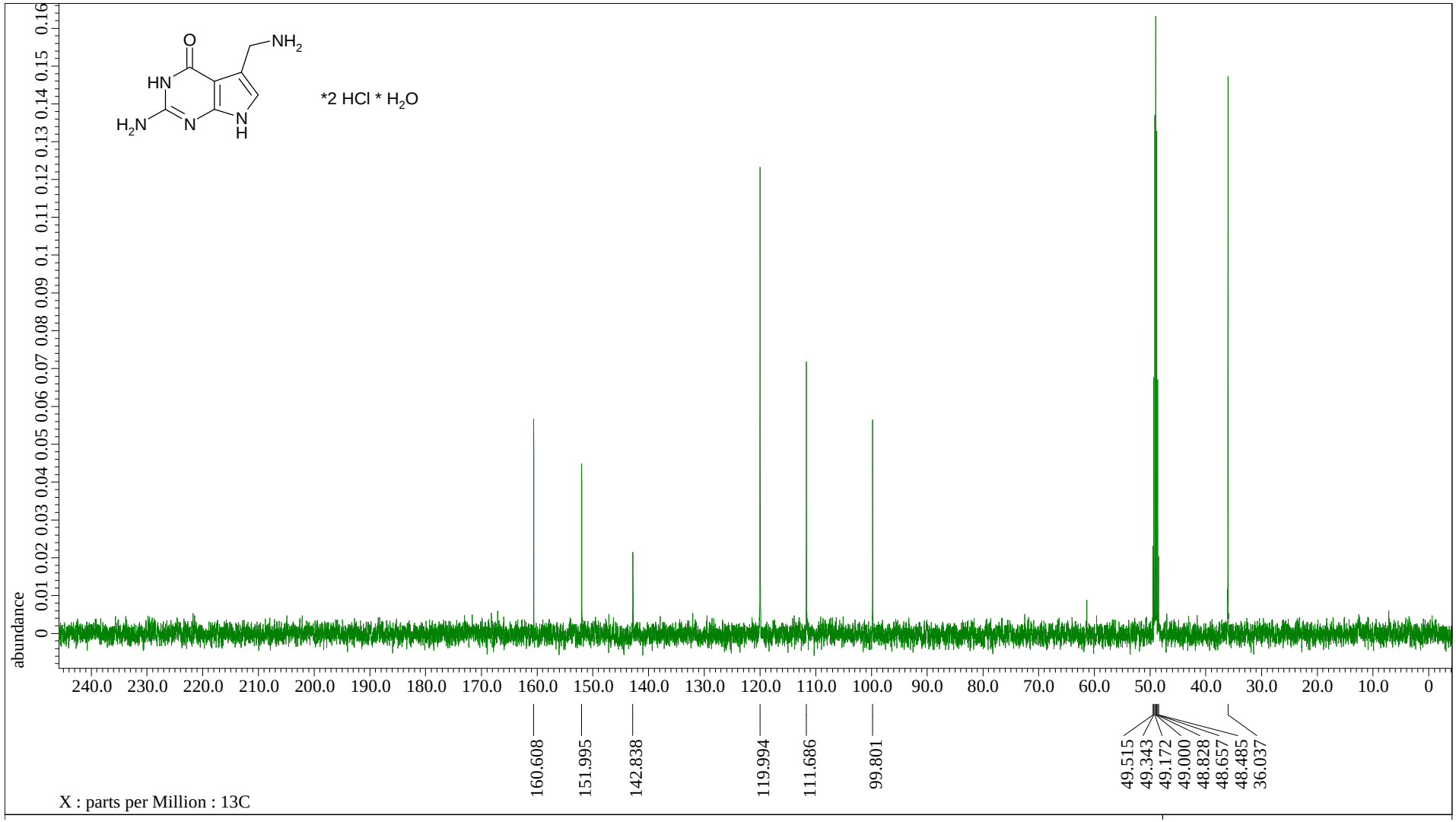
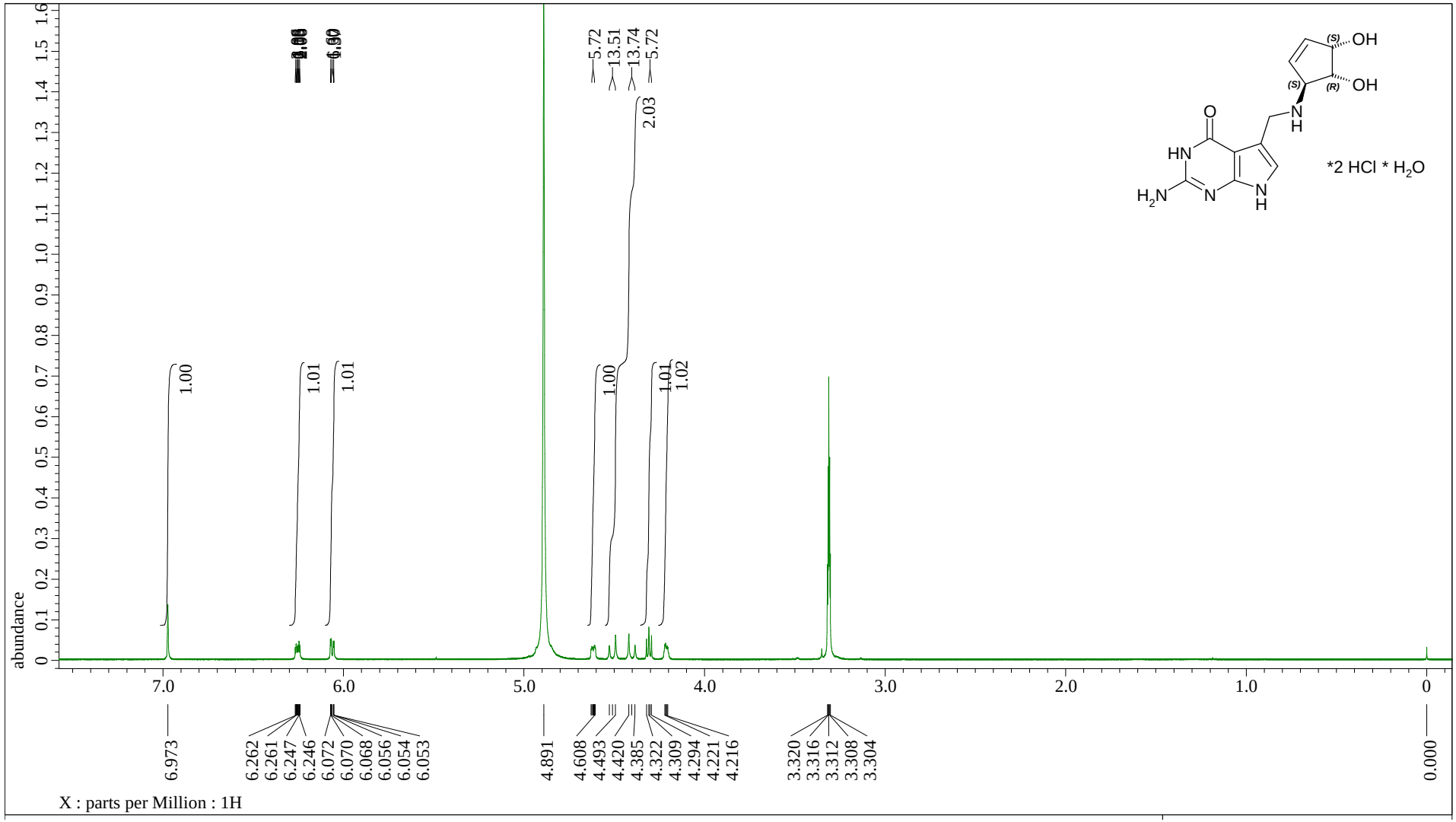


2-Amino-5-(aminomethyl)-3,7-dihydro-4*H*-pyrrolo[2,3-*d*]pyrimidin-4-one dihydrochloride hydrate (1, D_2O/CD_3OD)



2-Amino-5({[(1*S*,4*S*,5*R*)-4,5-dihydroxycyclopent-2-en-1-yl]amino}methyl)-3,7-dihydro-4*H*-pyrrolo[2,3-*d*]pyrimidin-4-one dihydrochloride hydrate (2, CD_3OD)



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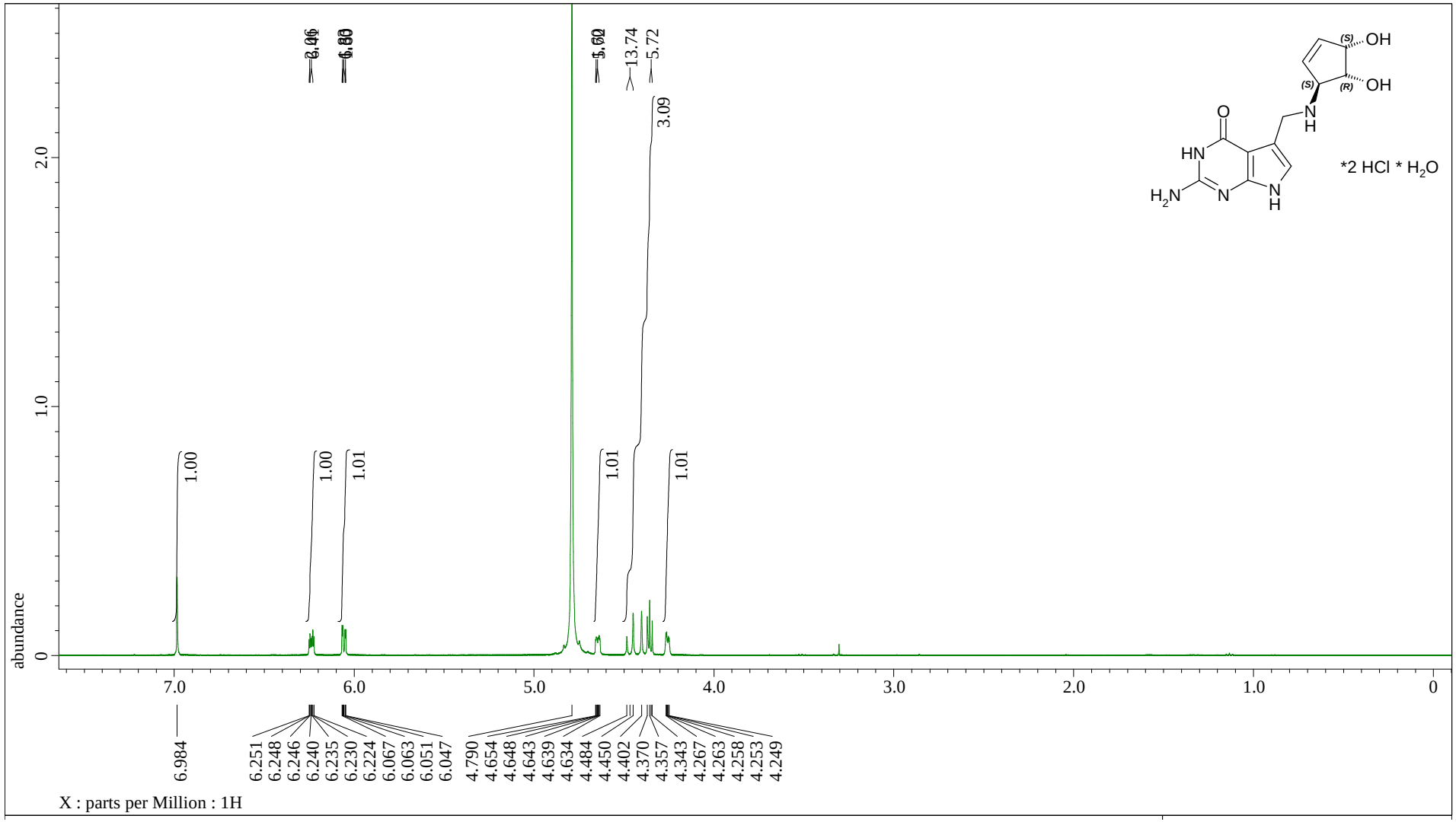

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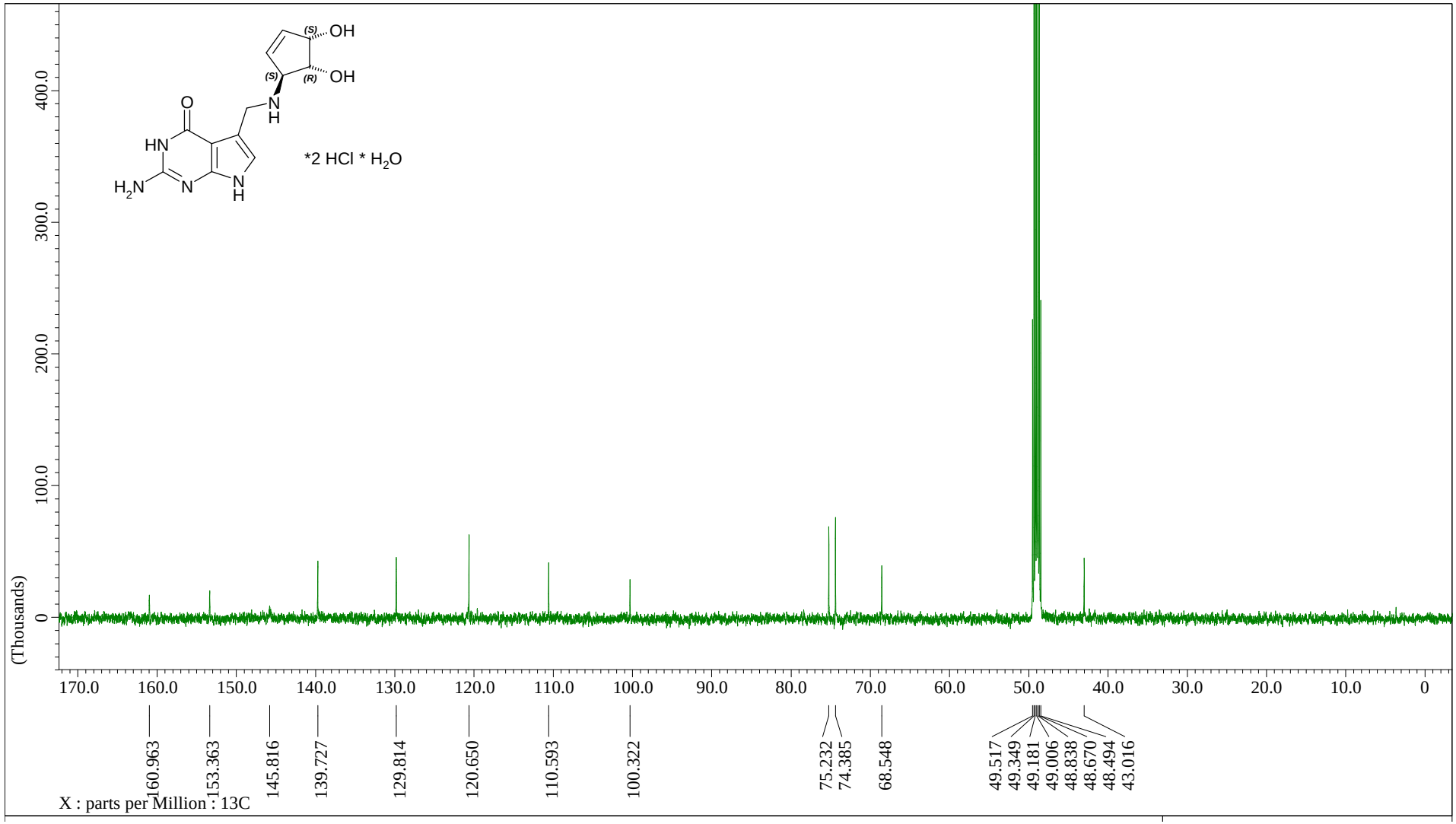

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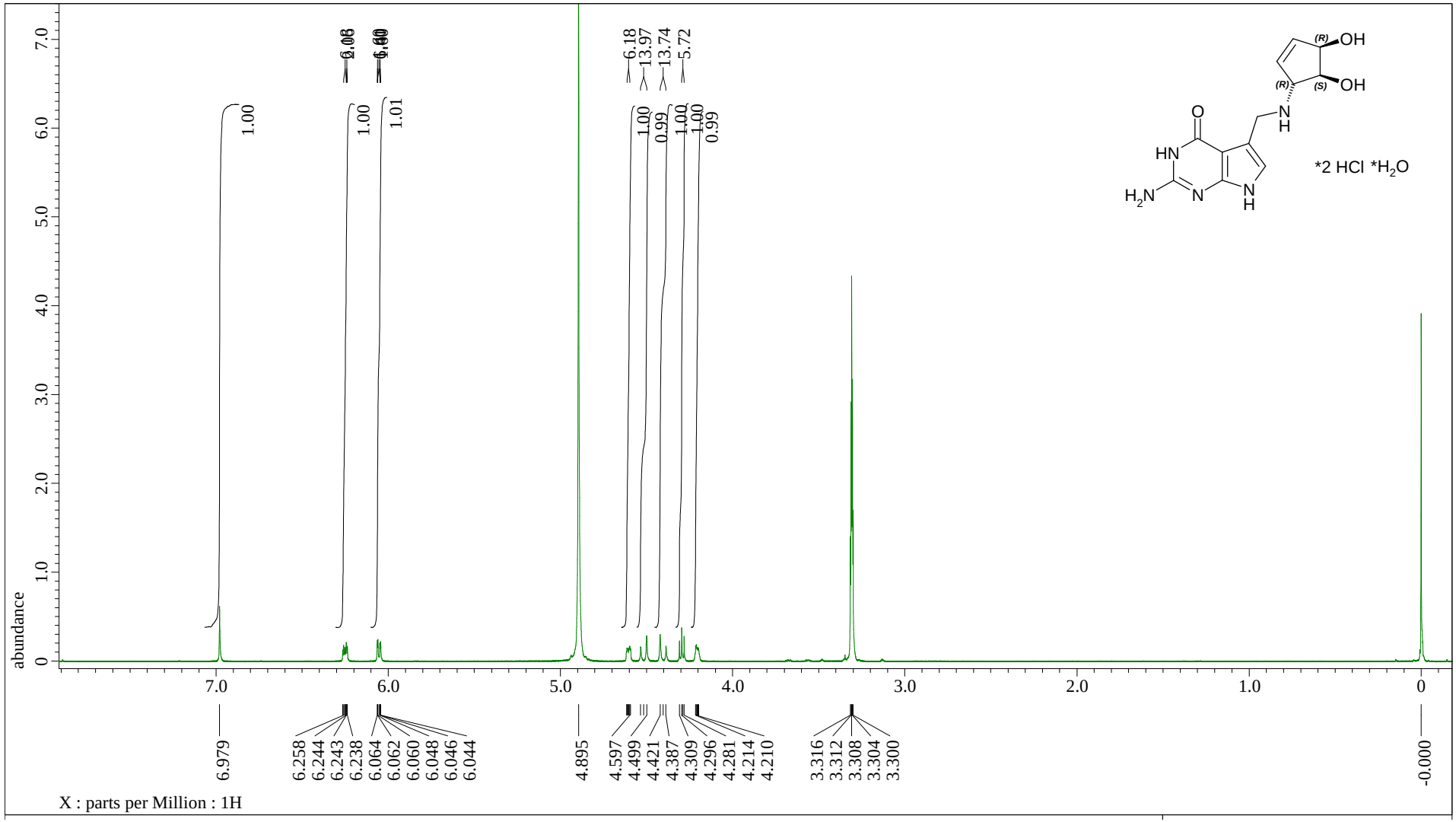

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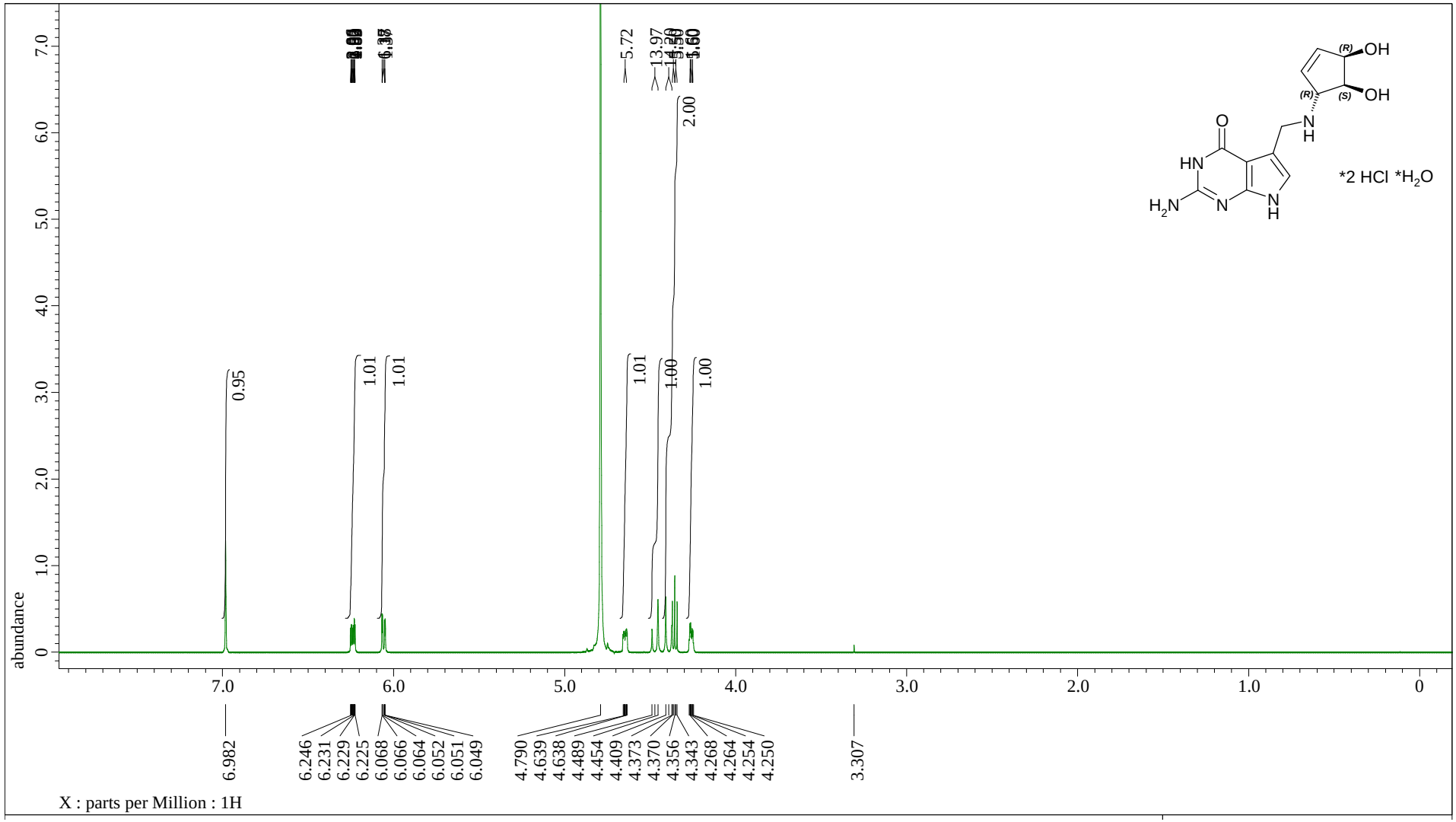

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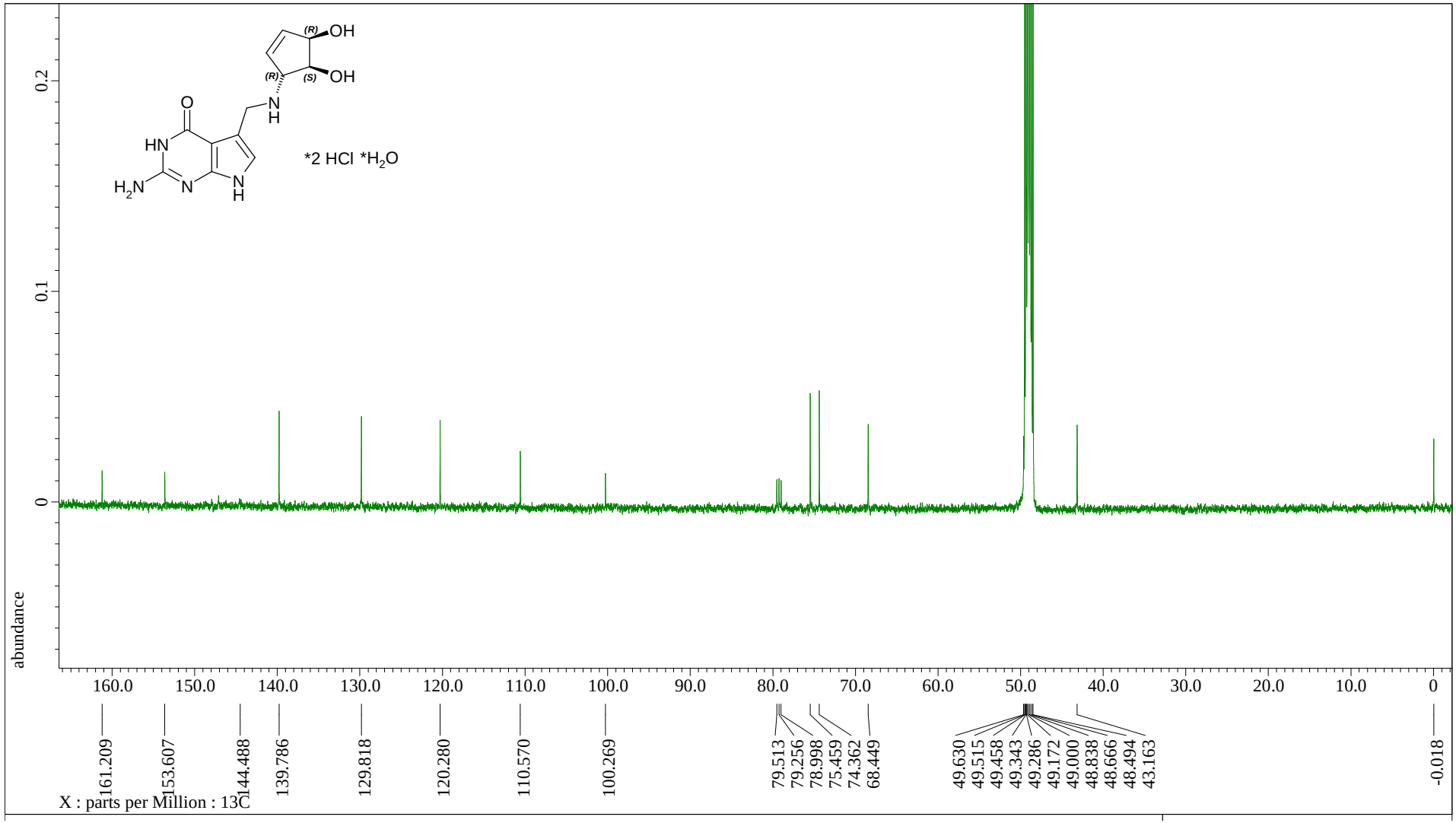

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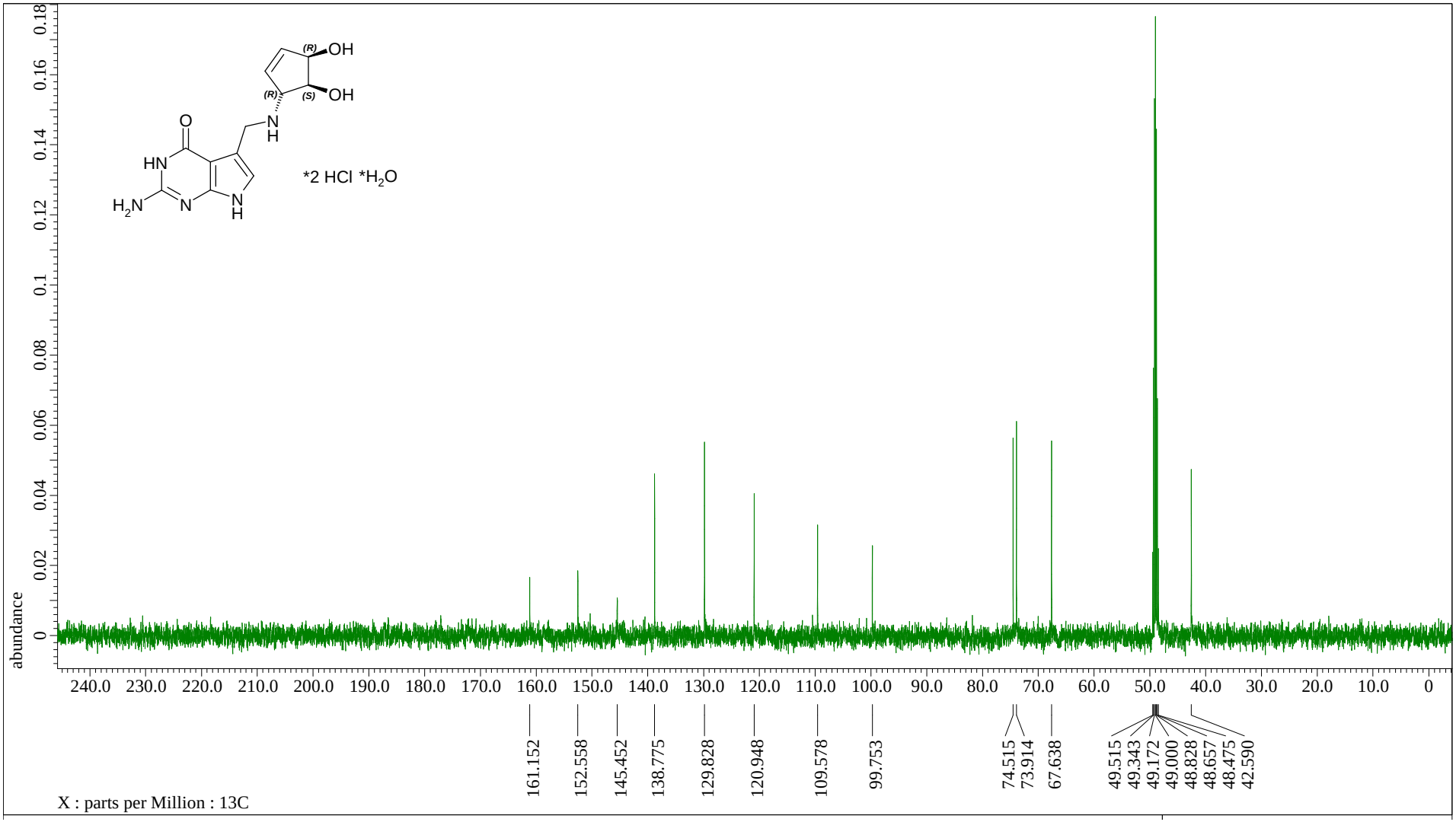
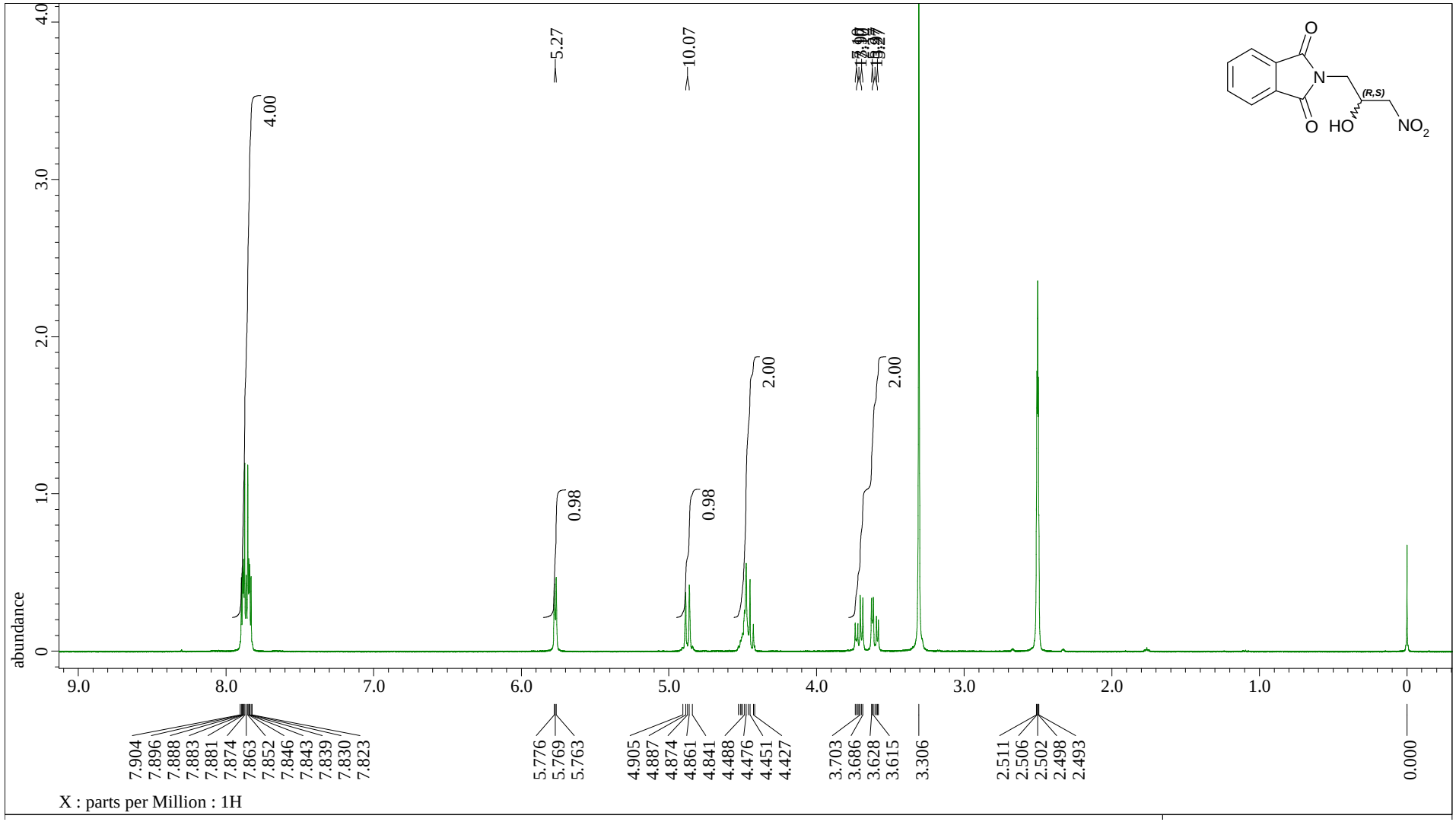
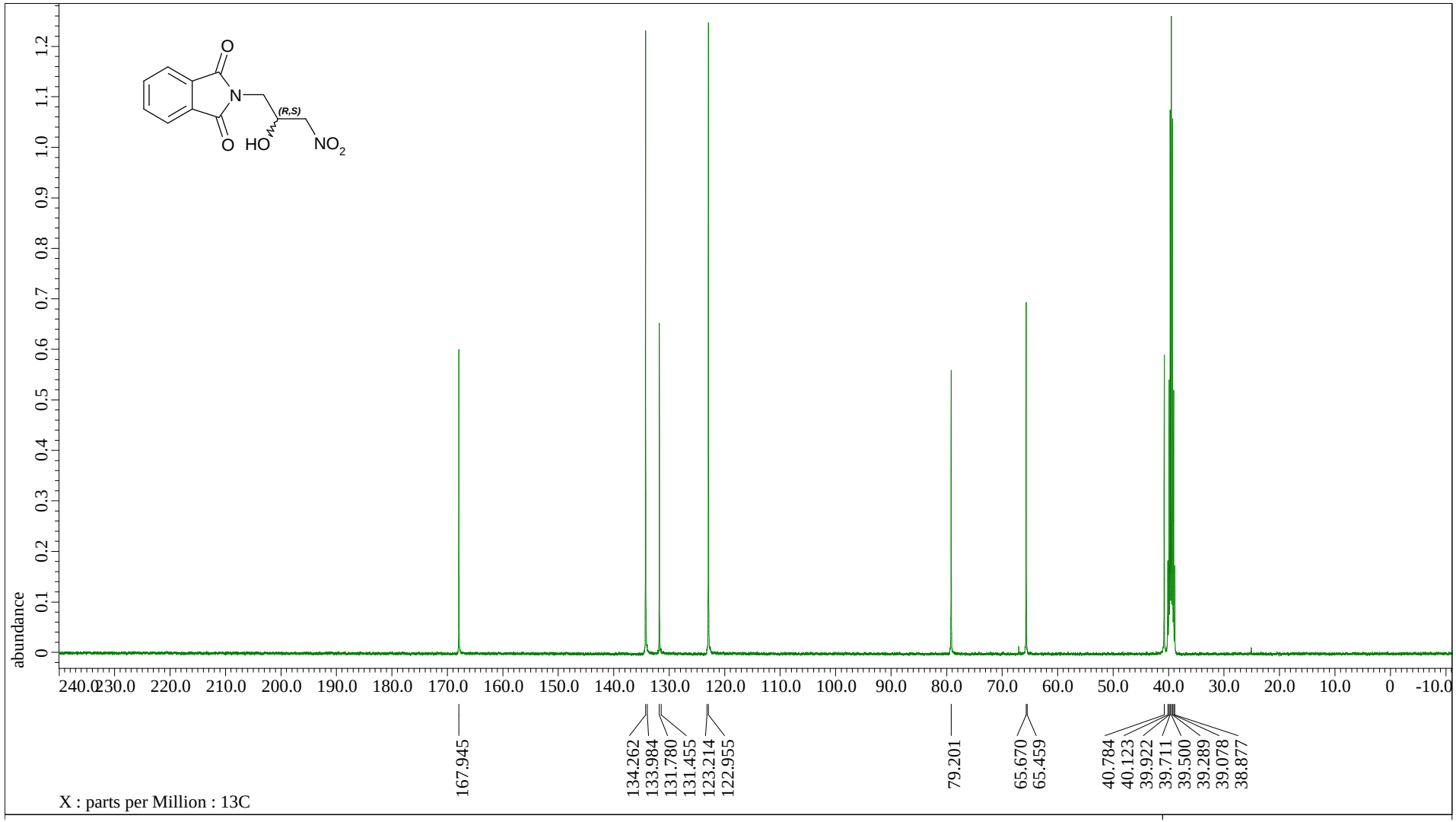


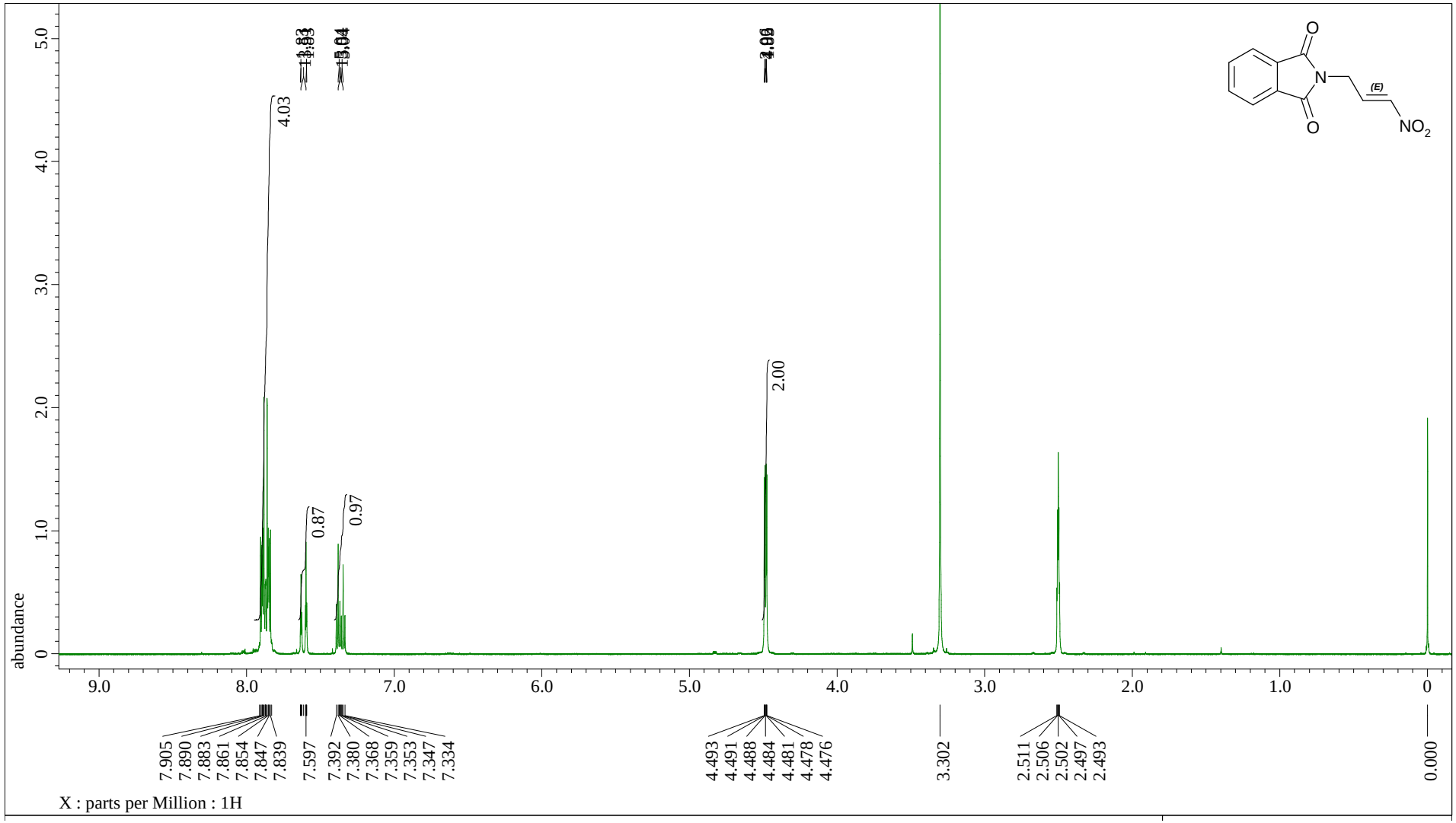
(*R*,*S*)-2-(2-Hydroxy-3-nitropropyl)-1*H*-isoindole-1,3(2*H*)-dione (6, DMSO-d$_6$)



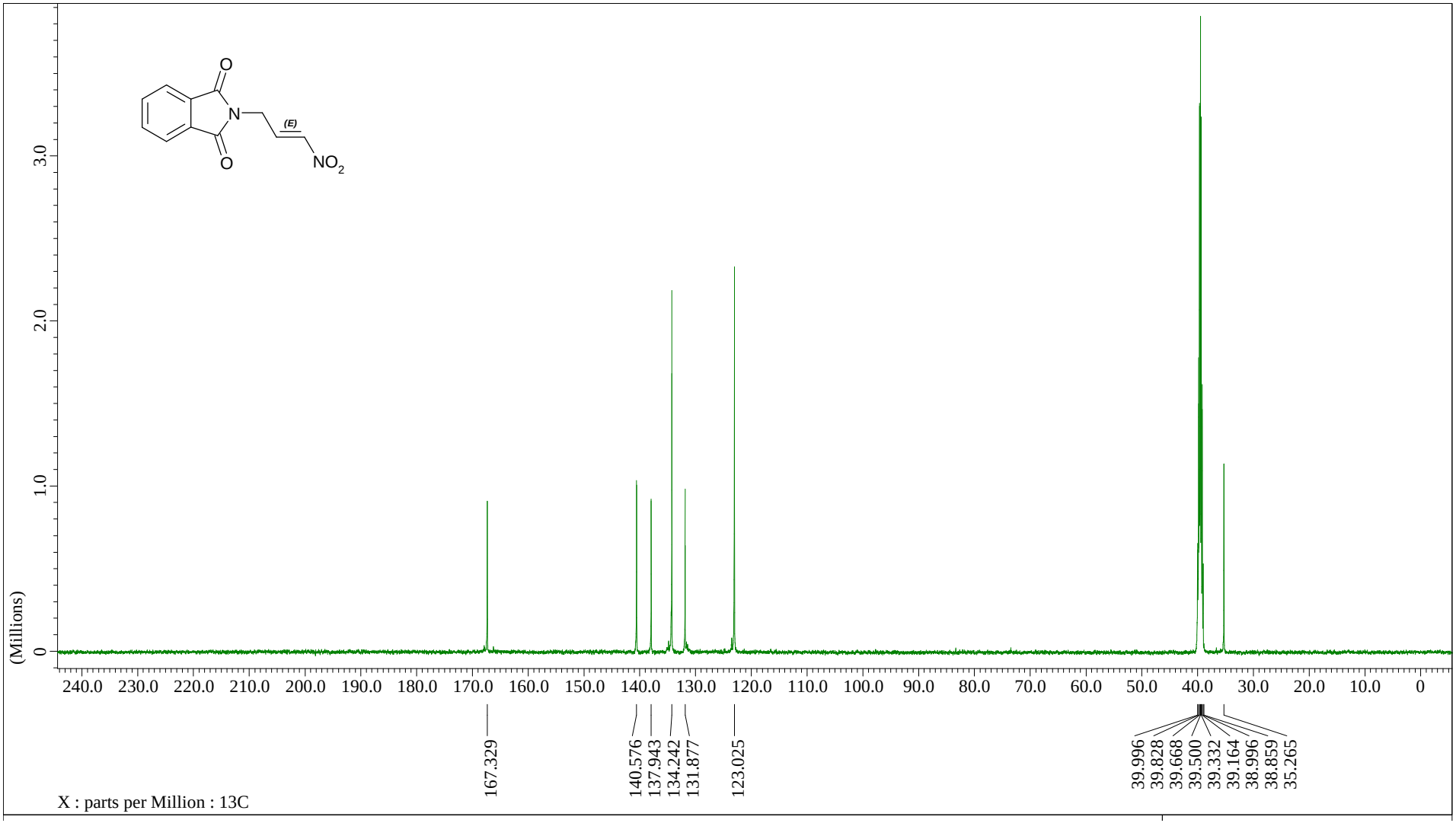
(*R*,*S*)-2-(2-Hydroxy-3-nitropropyl)-1*H*-isoindole-1,3(2*H*)-dione (6, DMSO-d_6)



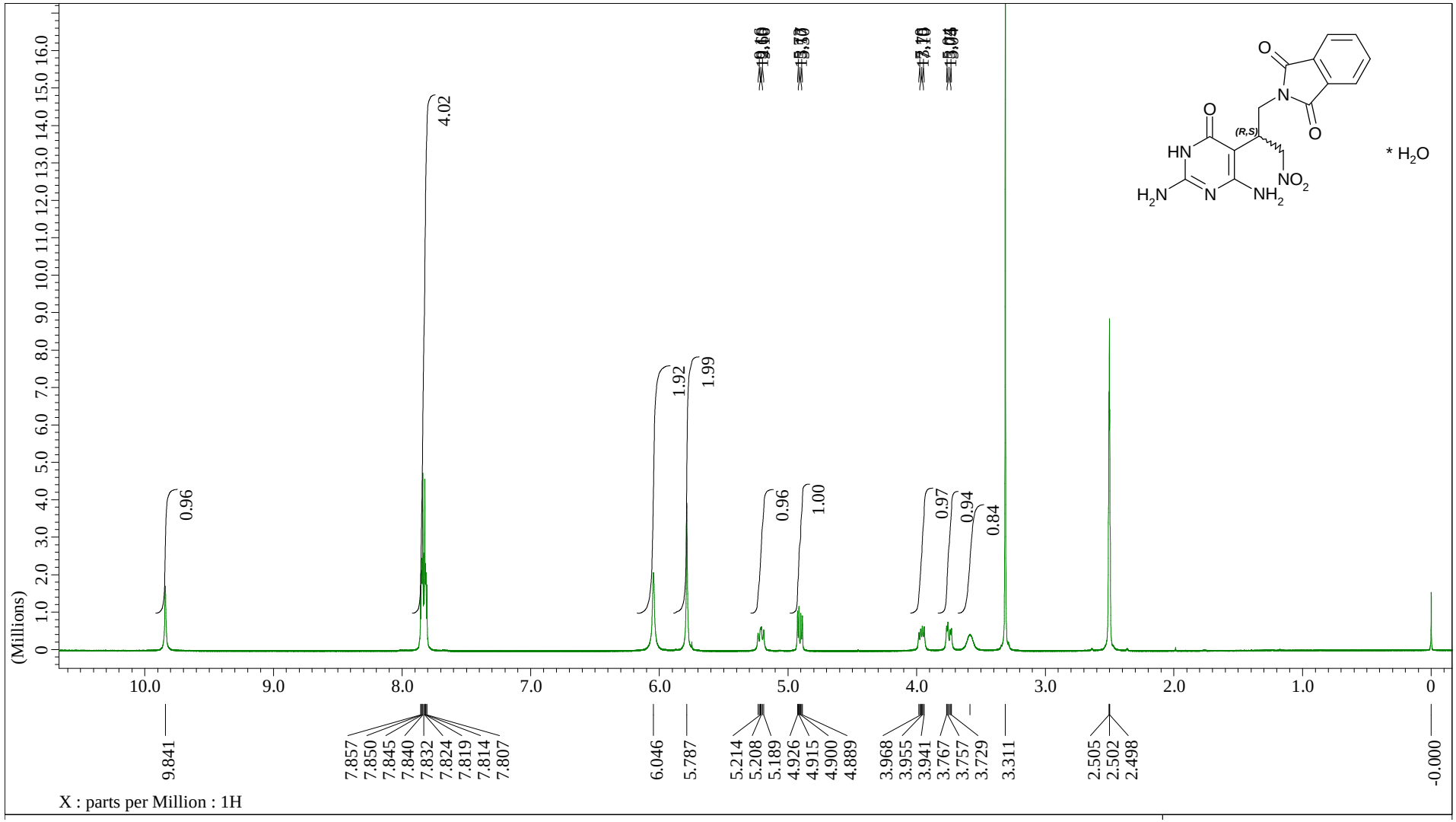
2-[(2*E*)-3-nitroprop-2-en-1-yl]-1*H*-isoindole-1,3(2*H*)-dione (7, DMSO-d_6)



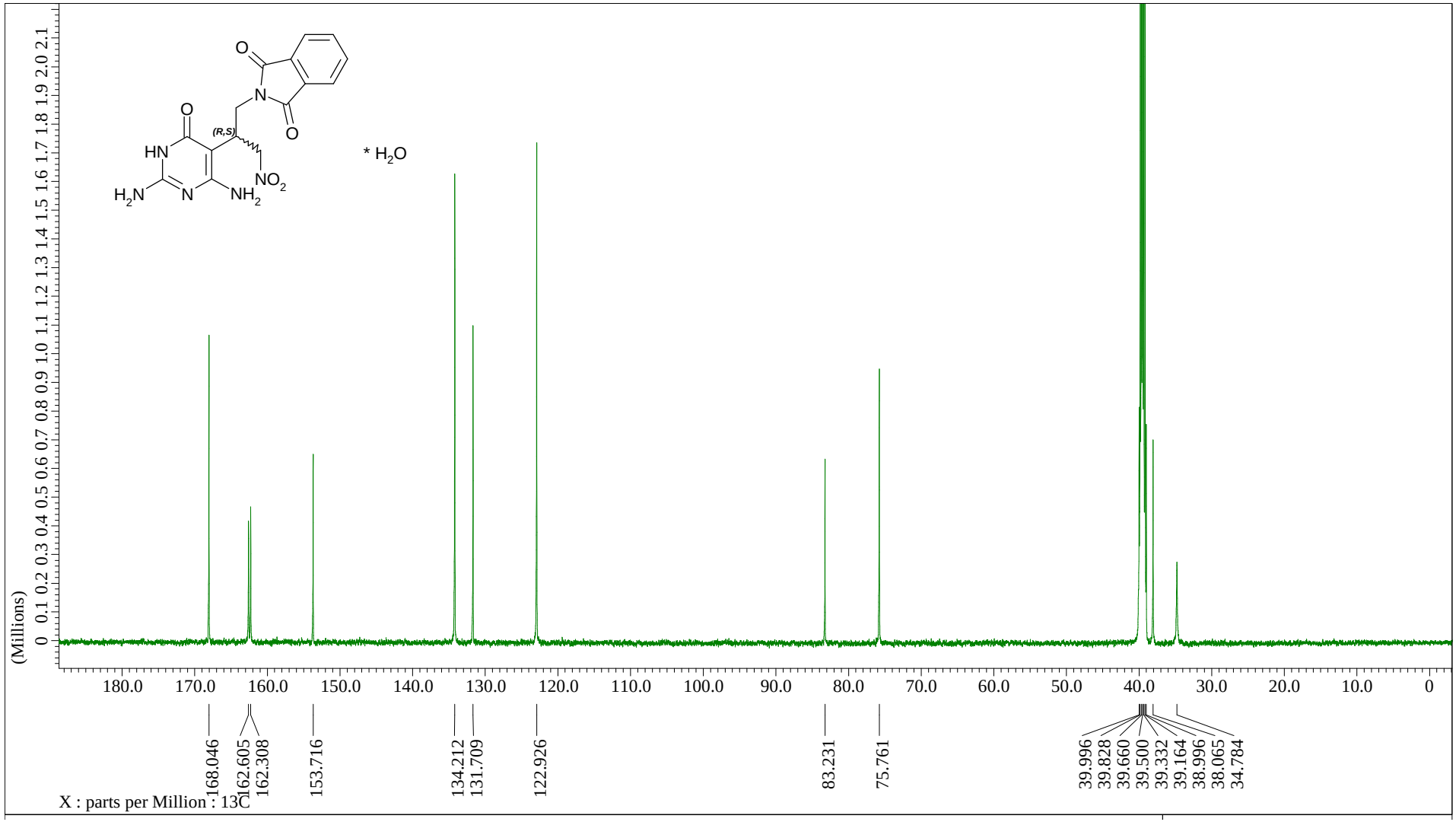
2-[(2*E*)-3-nitroprop-2-en-1-yl]-1*H*-isoindole-1,3(2*H*)-dione (7, DMSO-d_6)



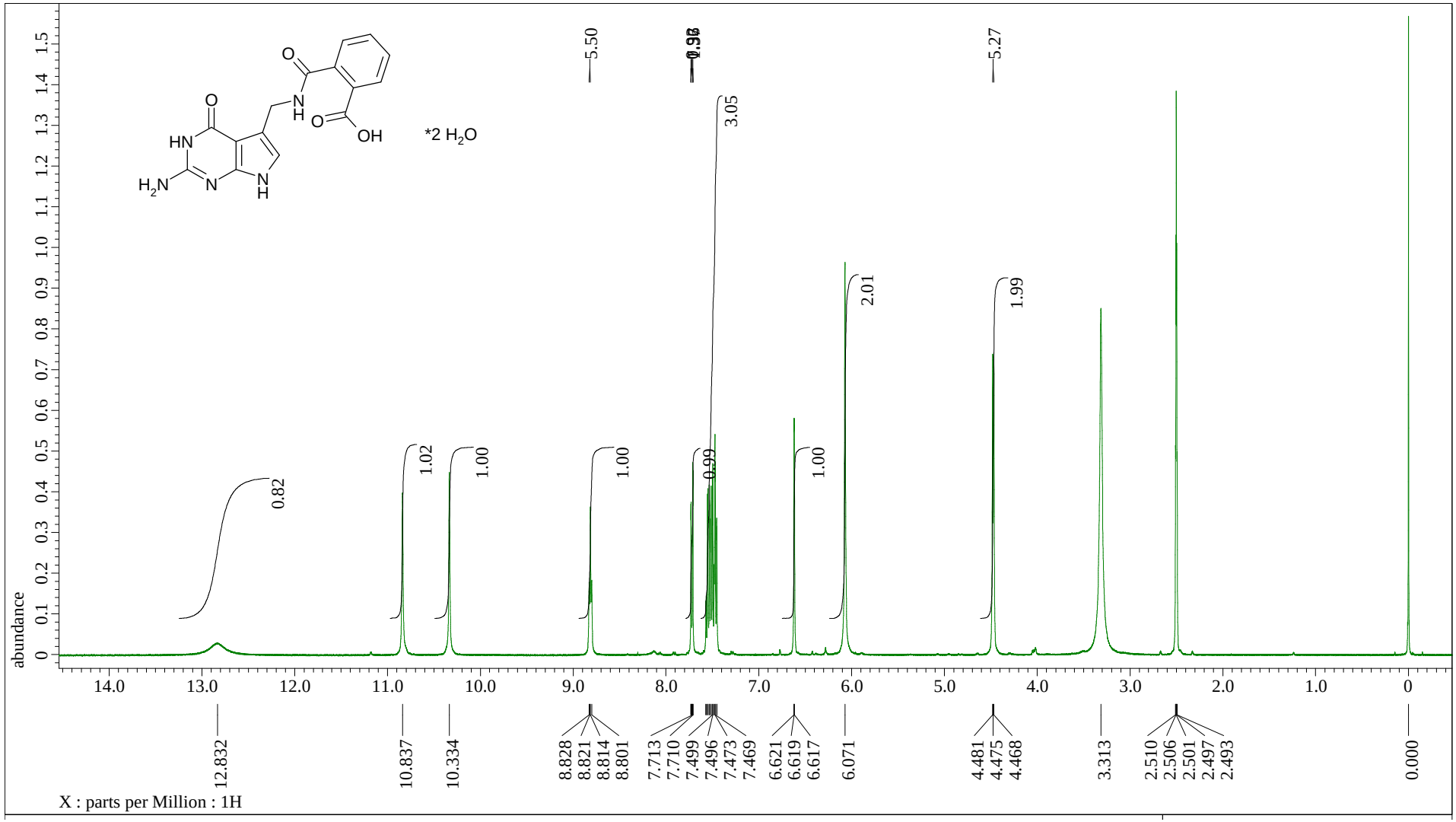
(*R*,*S*)-2-[2-(2,4-Diamino-6-oxo-1,6-dihydropyrimidin-5-yl)-3-nitropropyl]-1*H*-isoindole-1,3(2*H*)-dione hydrate (8, DMSO-d_6)



(*R*,*S*)-2-[2-(2,4-Diamino-6-oxo-1,6-dihydropyrimidin-5-yl)-3-nitropropyl]-1*H*-isoindole-1,3(2*H*)-dione hydrate (8, DMSO-d_6)



2-({[(2-Amino-4-oxo-4,7-dihydro-3*H*-pyrrolo[2,3-*d*]pyrimidin-5-yl)methyl]amino}carbonyl)benzoic acid dihydrate (9, DMSO-d_6)



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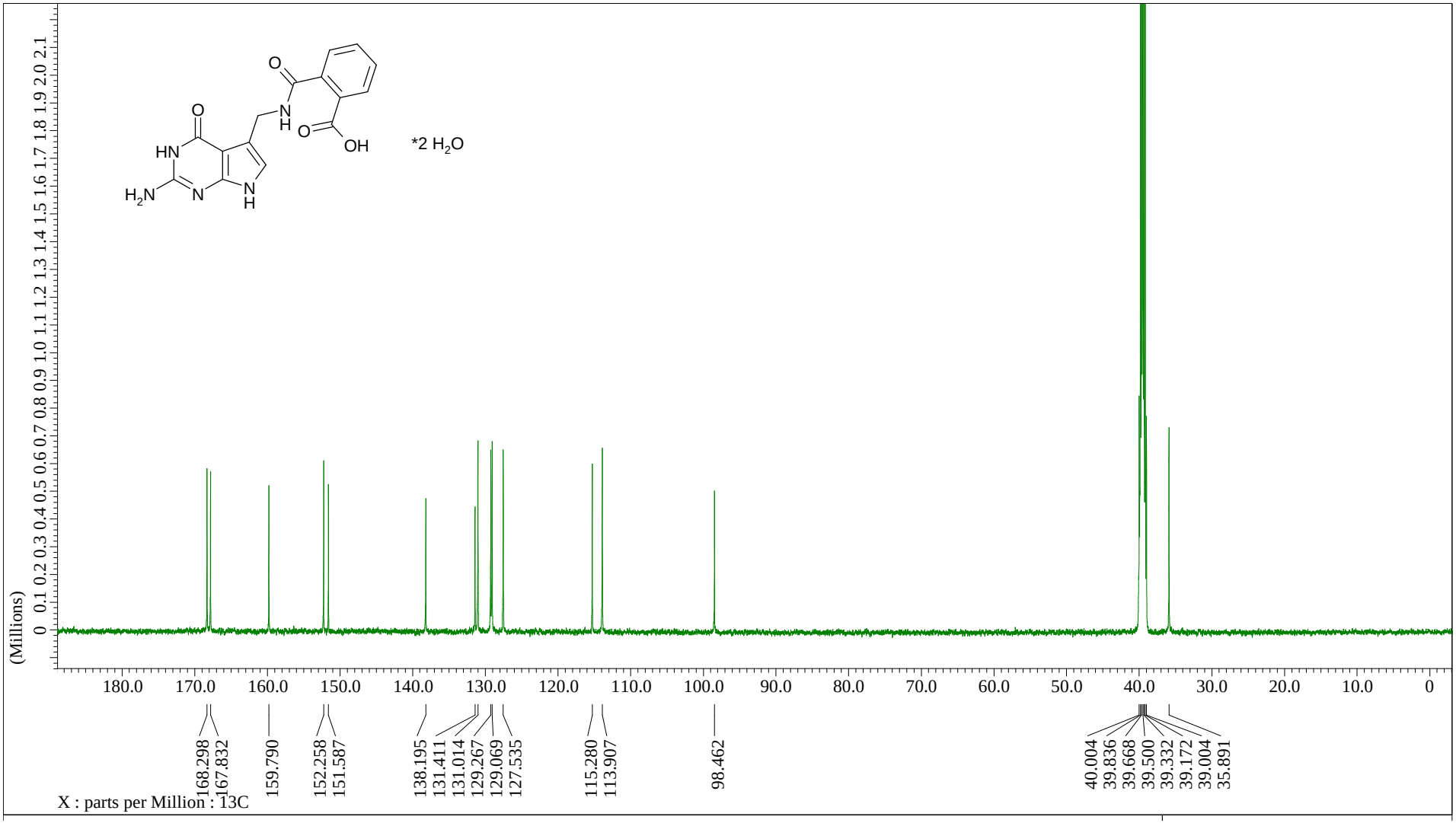
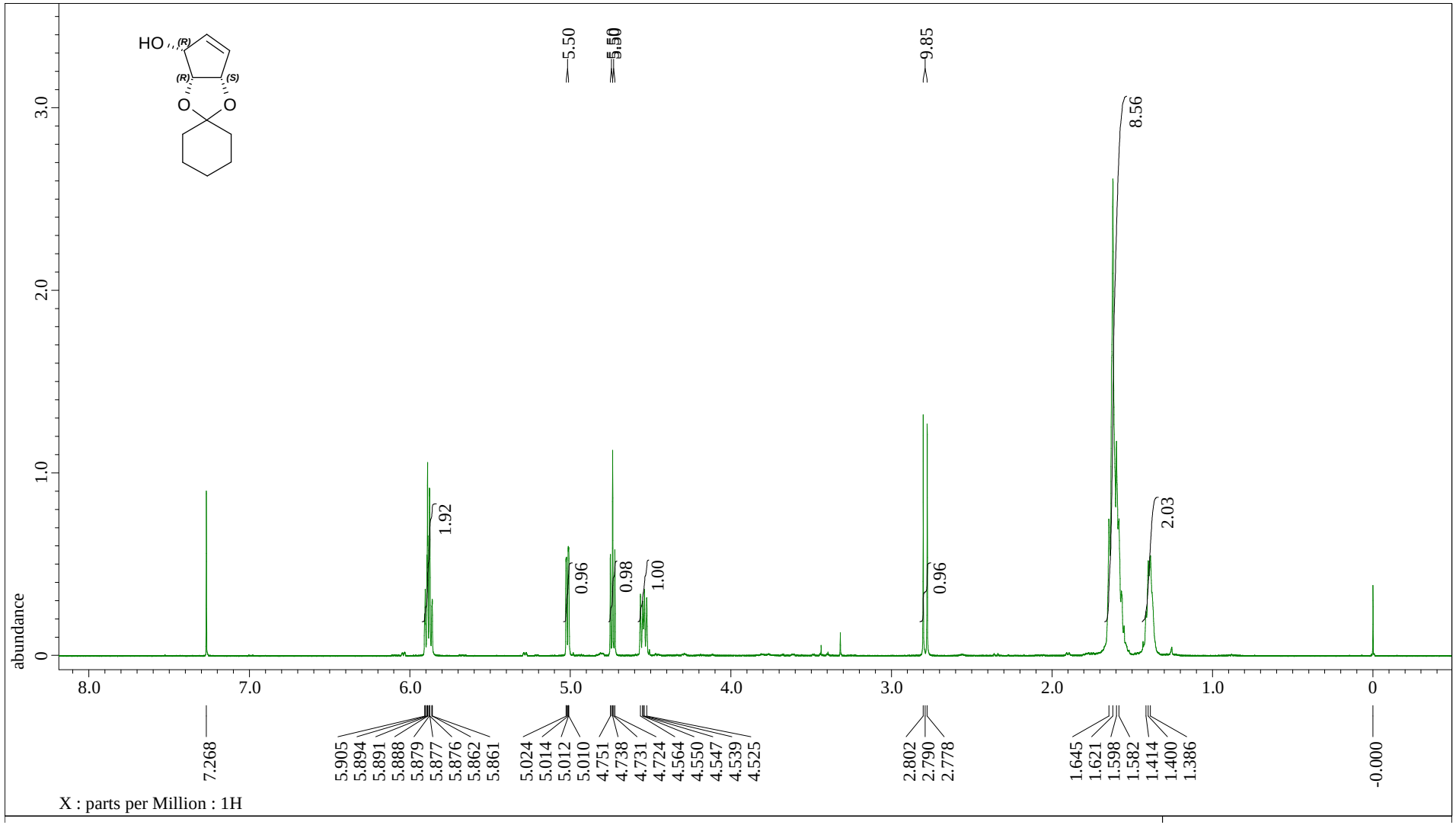
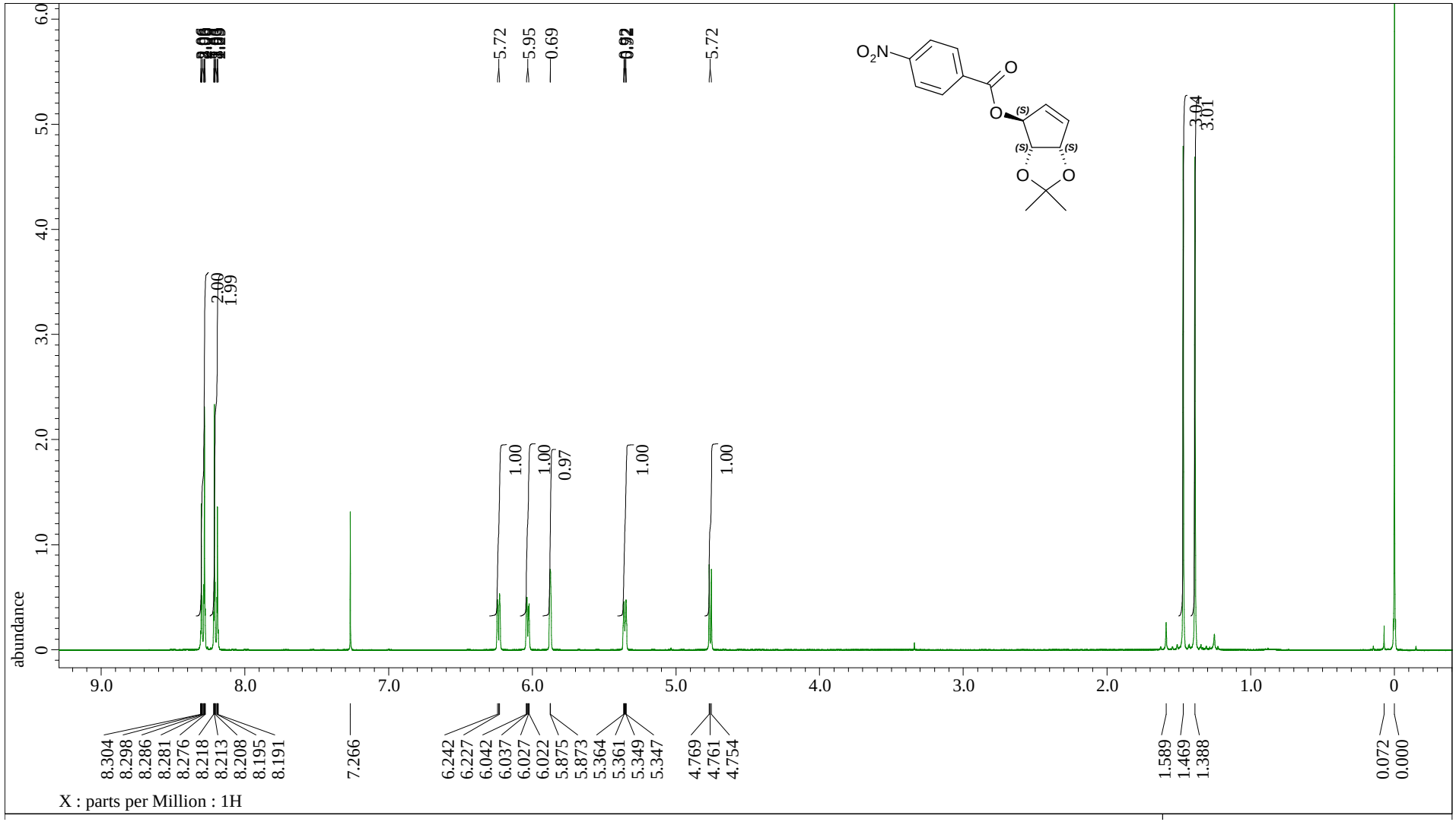


(3'a*R*,4'*R*,6'a*S*)-4',6'a-dihydro-3'a*H*-spiro[cyclohexane-1,2'-cyclopenta[*d*][1,3]dioxole]-4'-ol (10b, $CDCl_3$)



(3a*S*,4*S*,6a*S*)-2,2-dimethyl-2*H*,3a*H*,4*H*,6a*H*-cyclopenta[*d*][1,3]dioxol-4-yl 4-nitrobenzoate (11a, $CDCl_3$)



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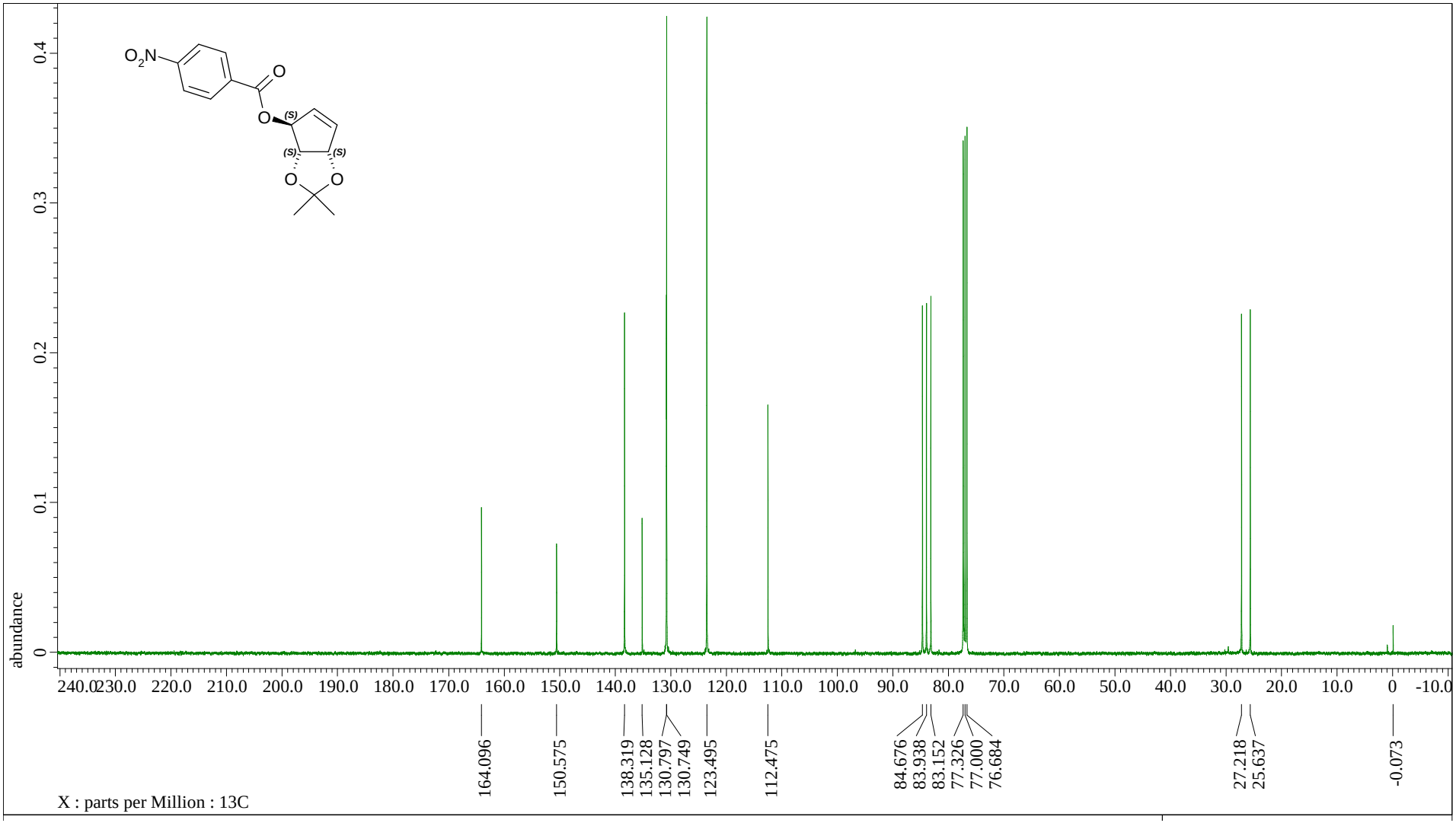

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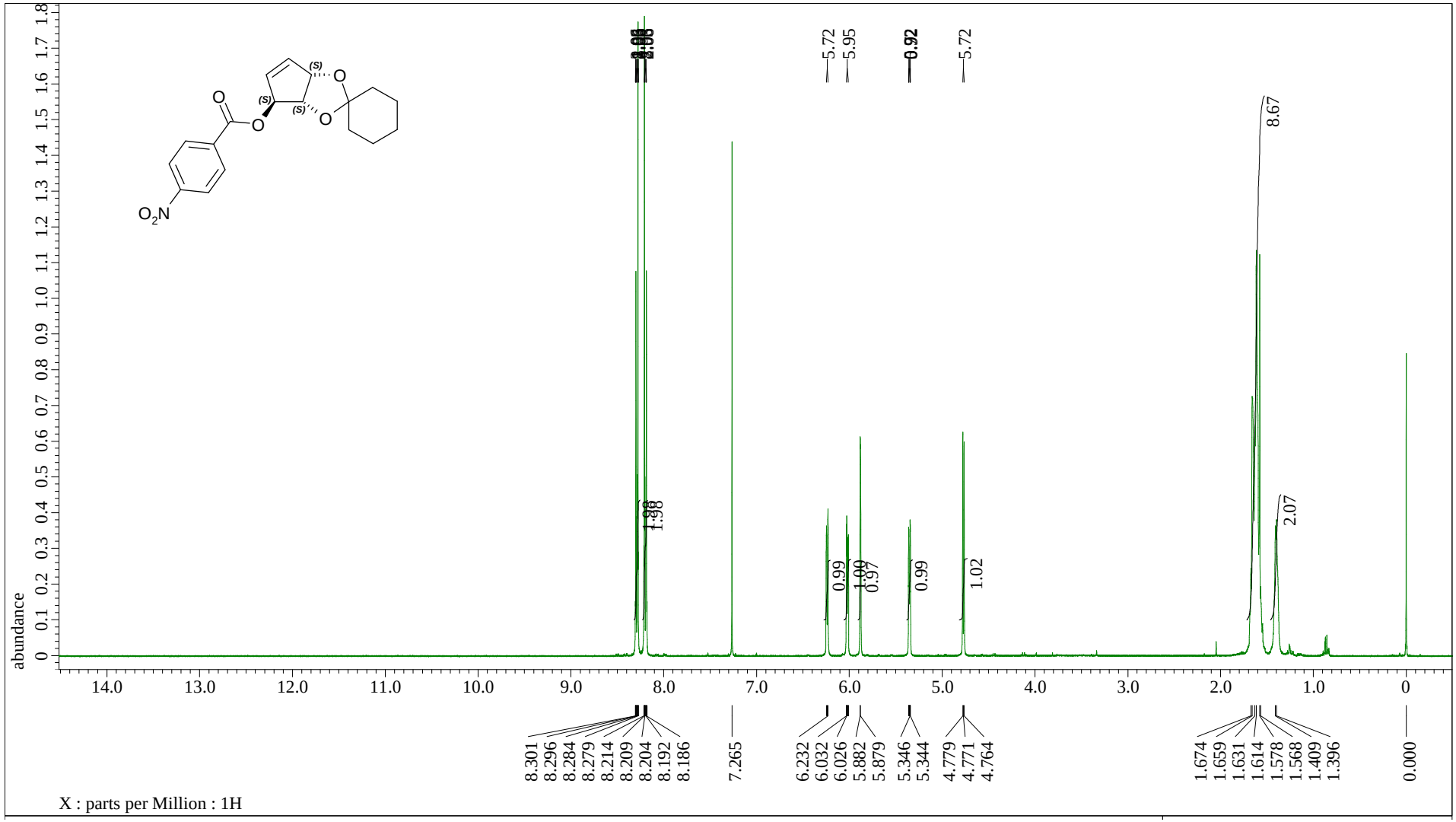

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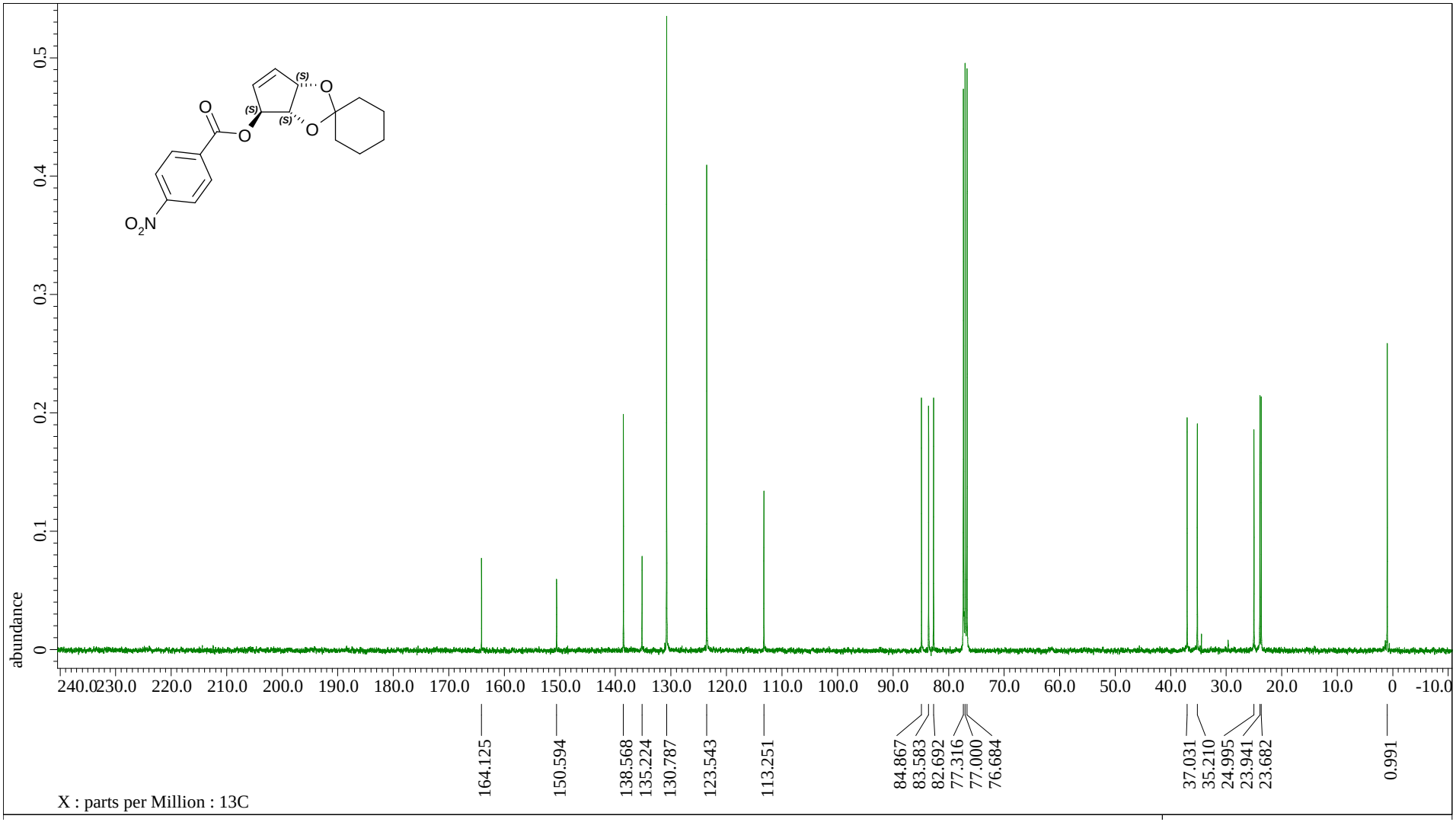

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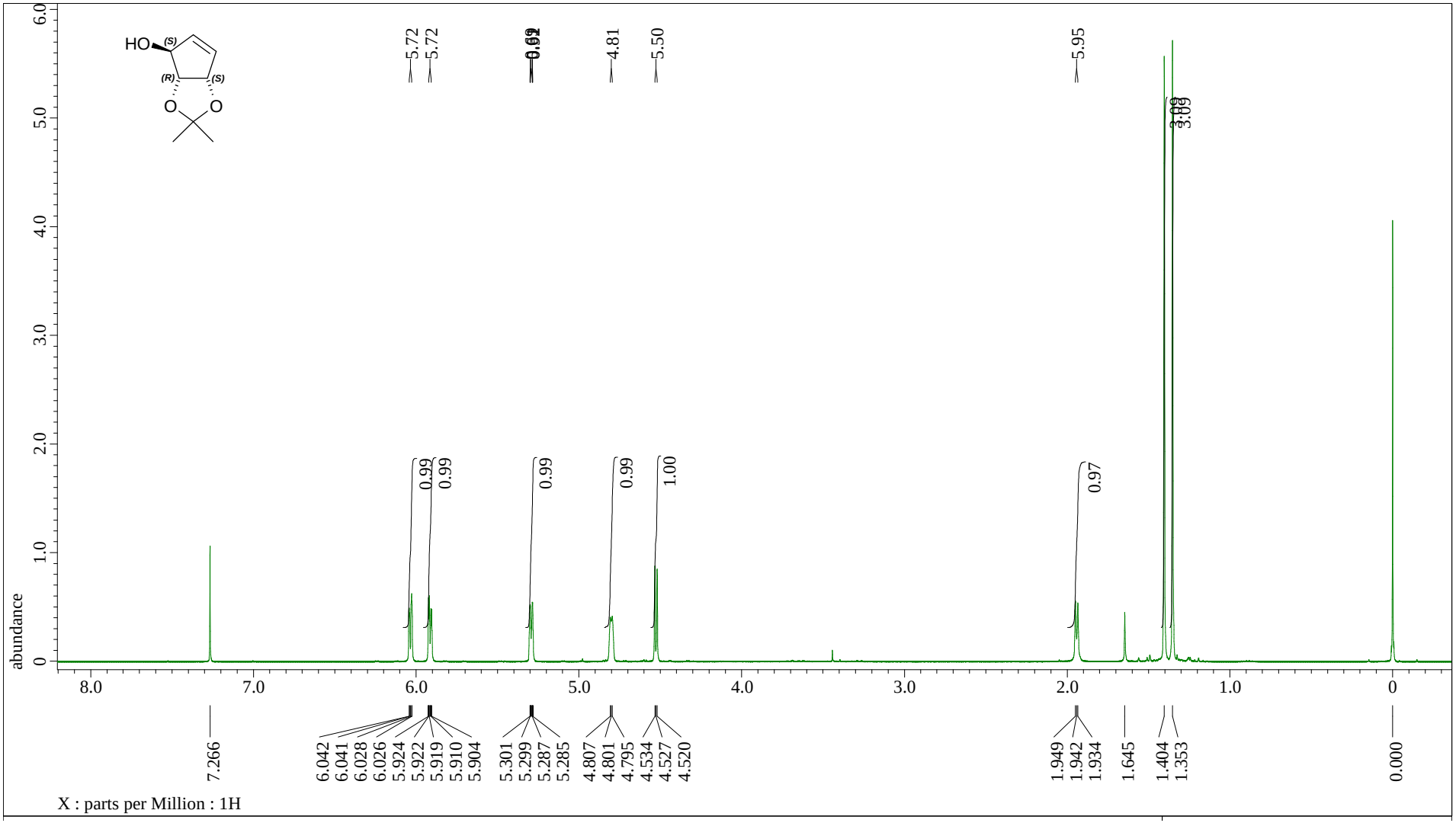

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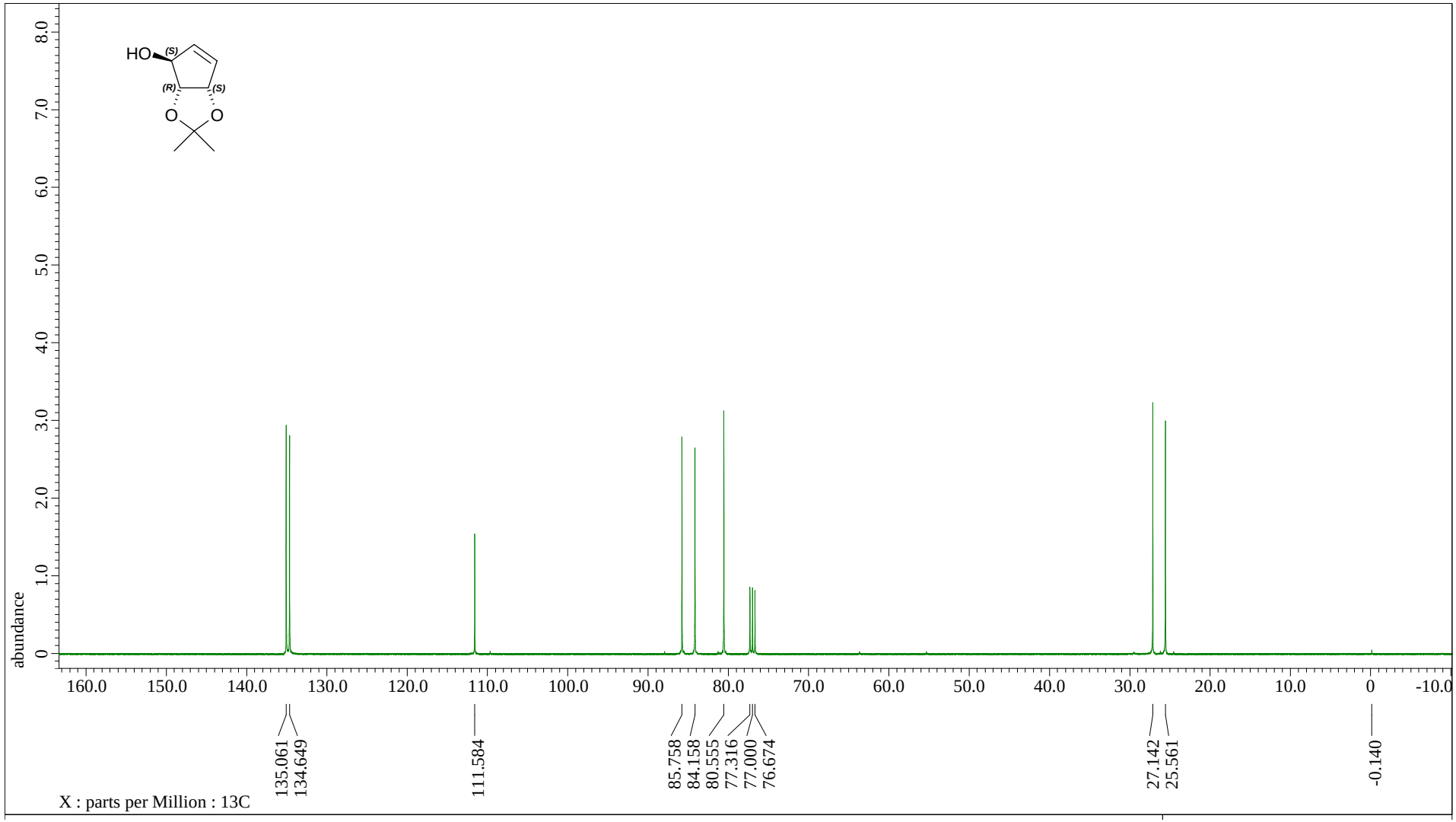

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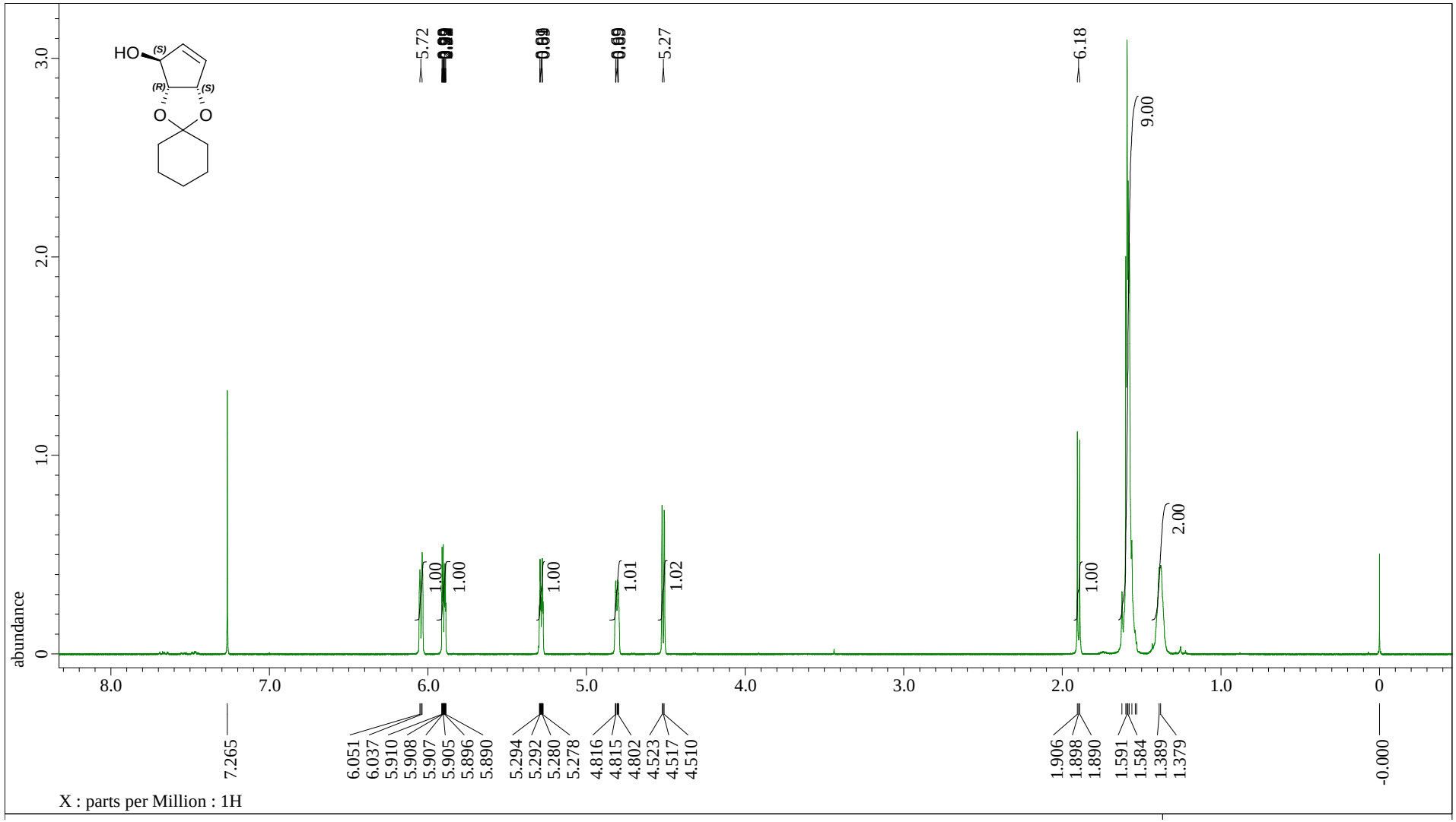

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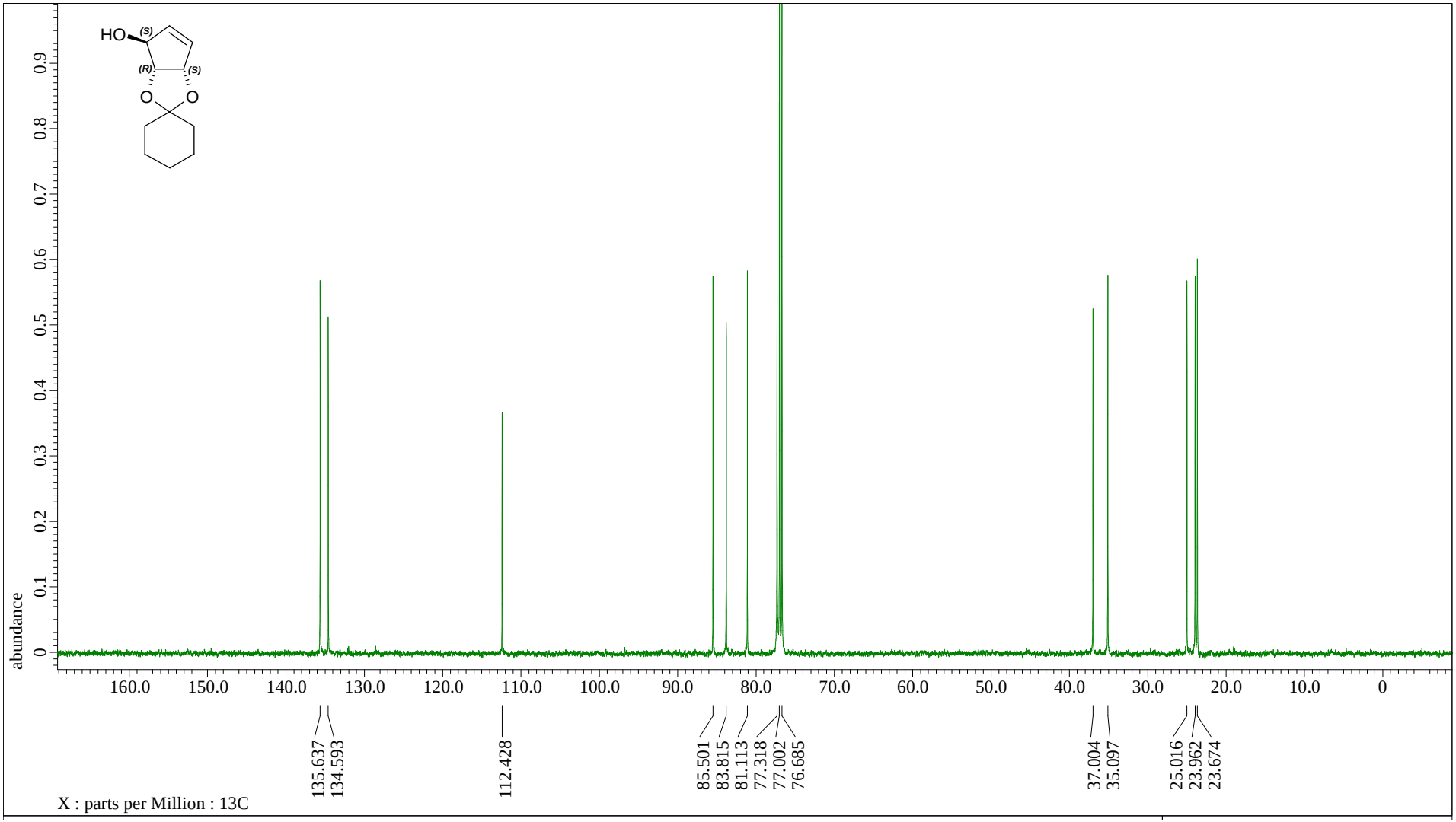
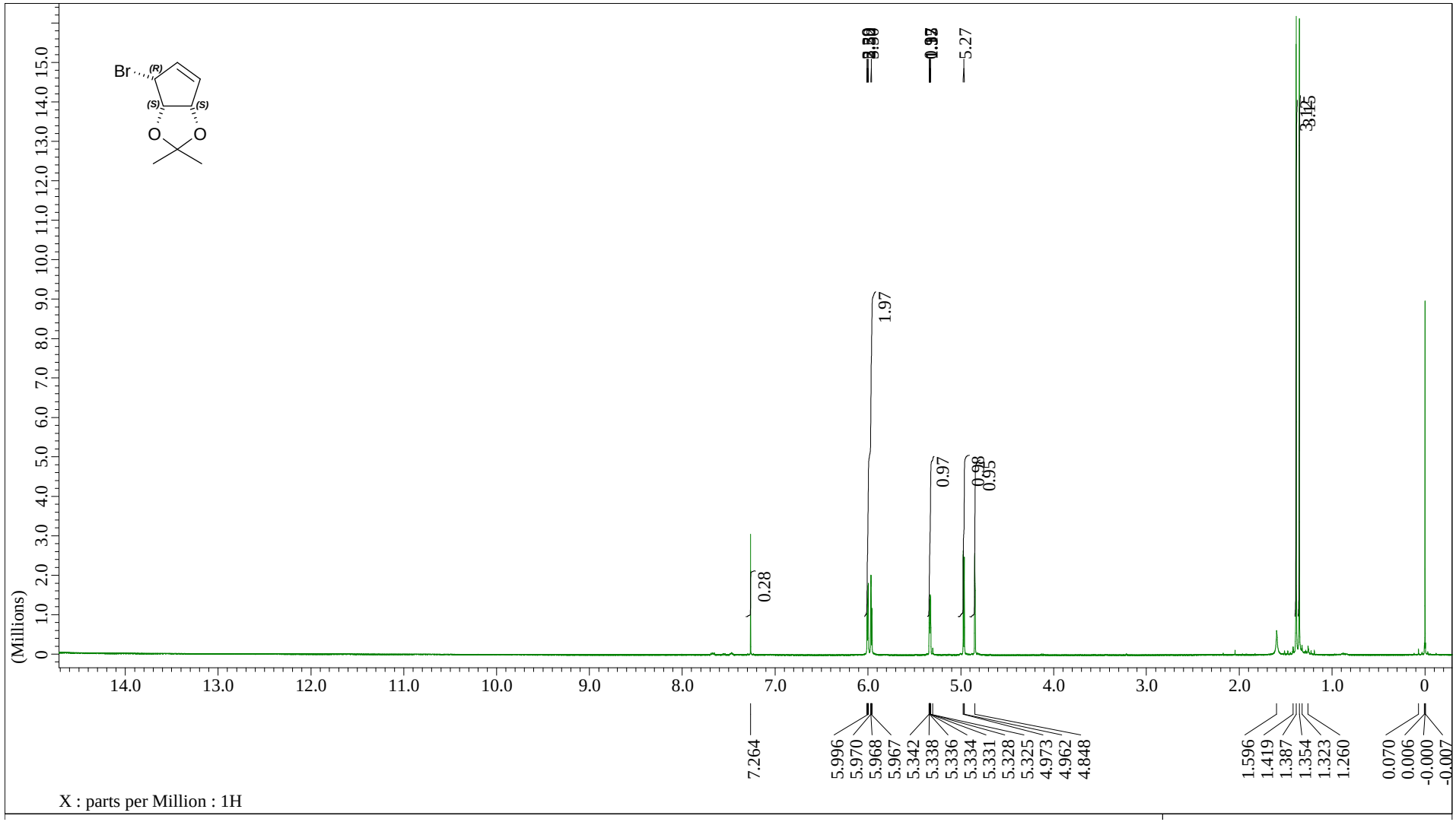
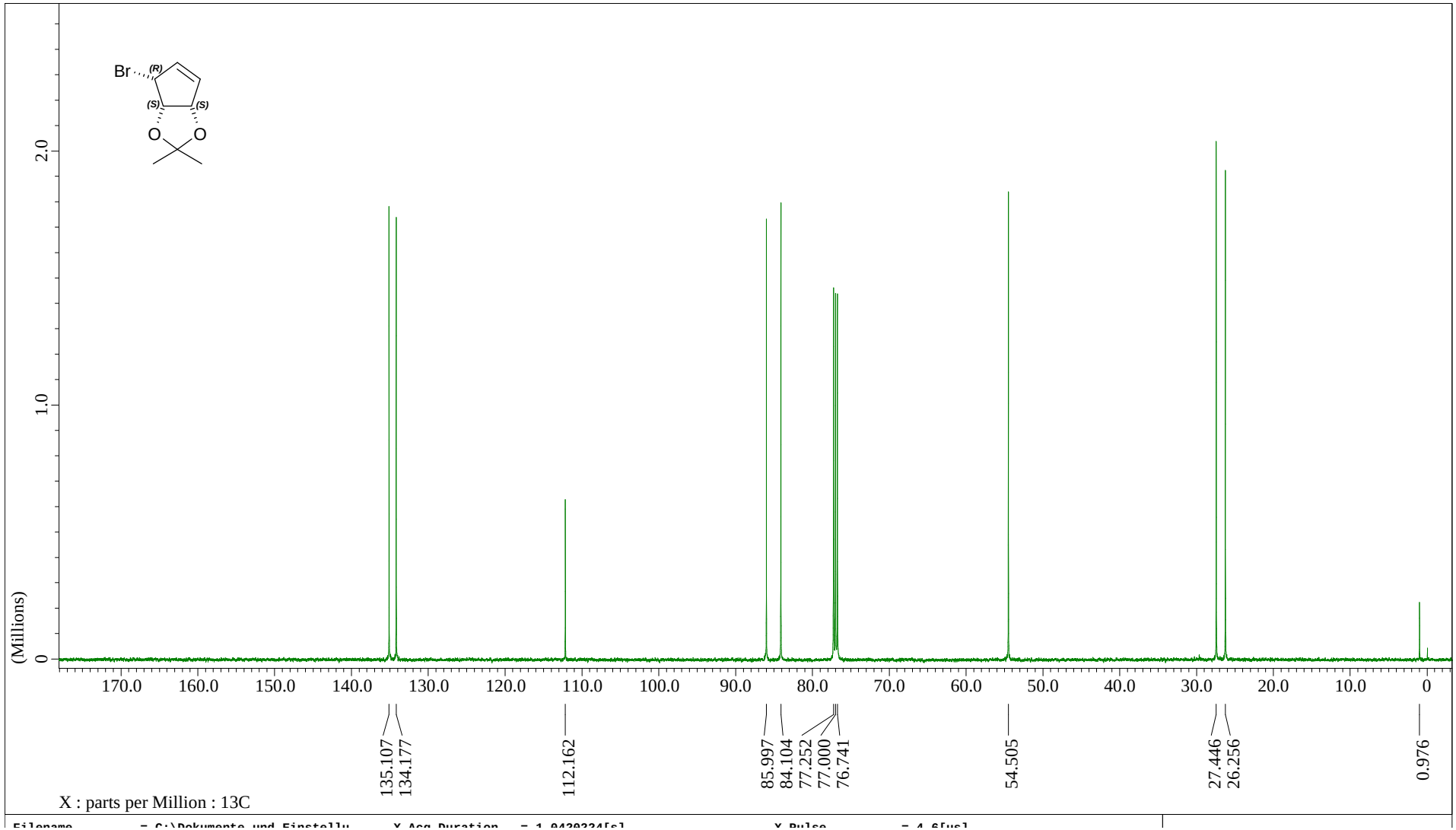


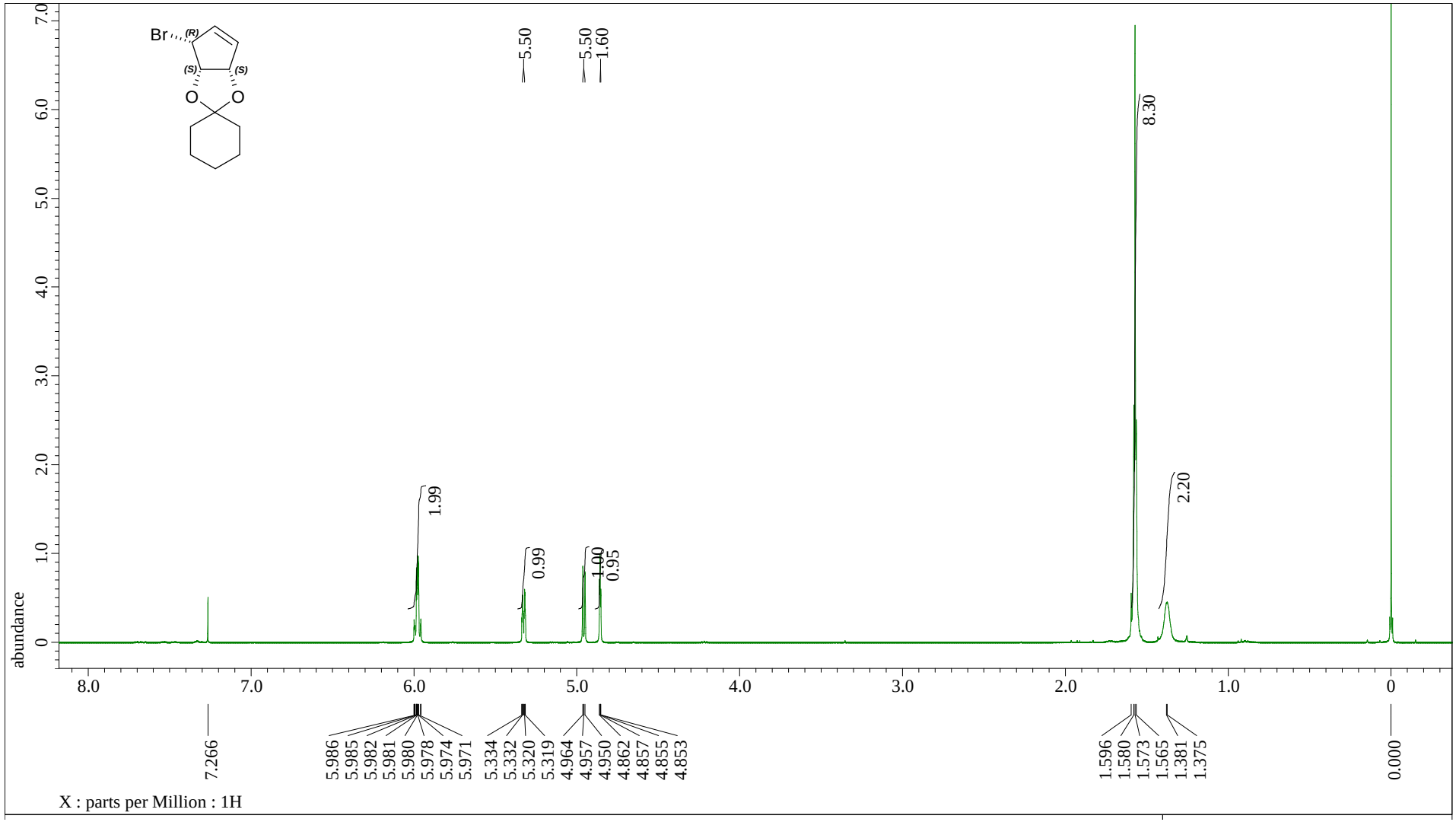
(3a*S*,4*R*,6a*S*)-4-bromo-2,2-dimethyl-2*H*,3a*H*,4*H*,6a*H*-cyclopenta[*d*][1,3]dioxole (13a, $CDCl_3$)



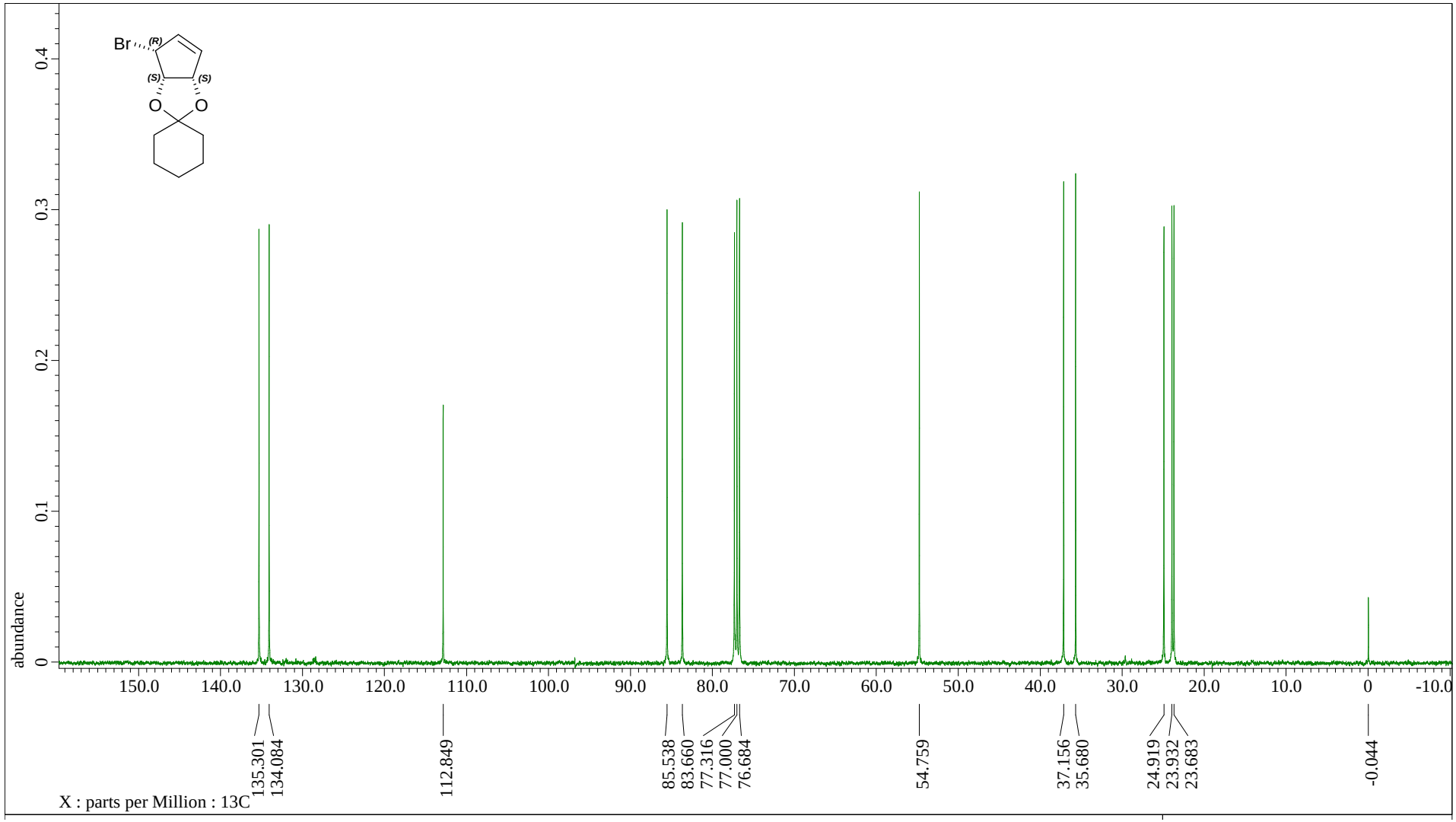
(3a*S*,4*R*,6a*S*)-4-bromo-2,2-dimethyl-2*H*,3a*H*,4*H*,6a*H*-cyclopenta[*d*][1,3]dioxole (13a, $CDCl_3$)



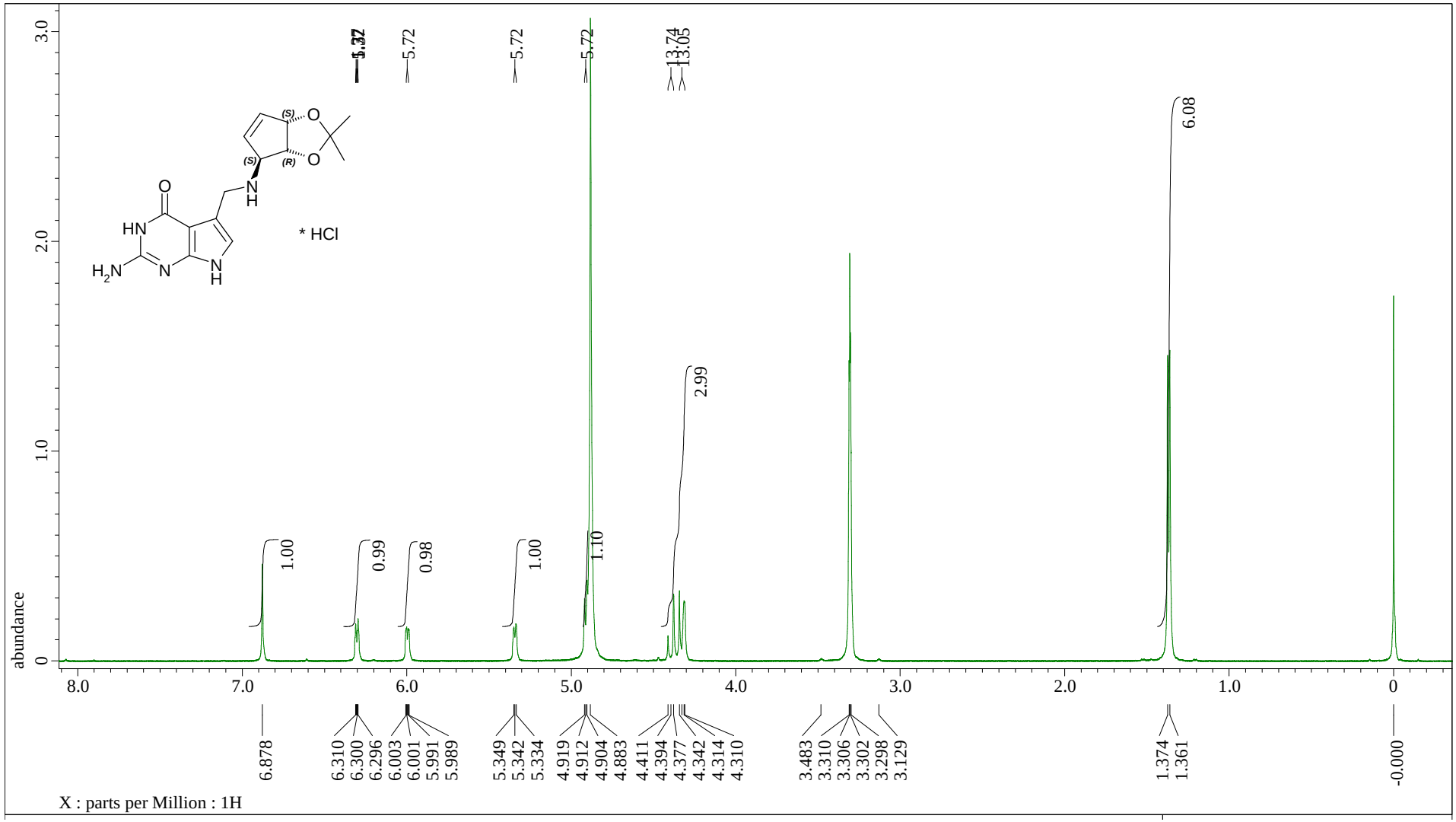
(3a*S*,4*R*,6a*S*)-4-bromo-4,6a-dihydro-3a*H*-spiro[cyclohexane-1,2-cyclopenta[*d*][1,3]dioxole (13b, $CDCl_3$)



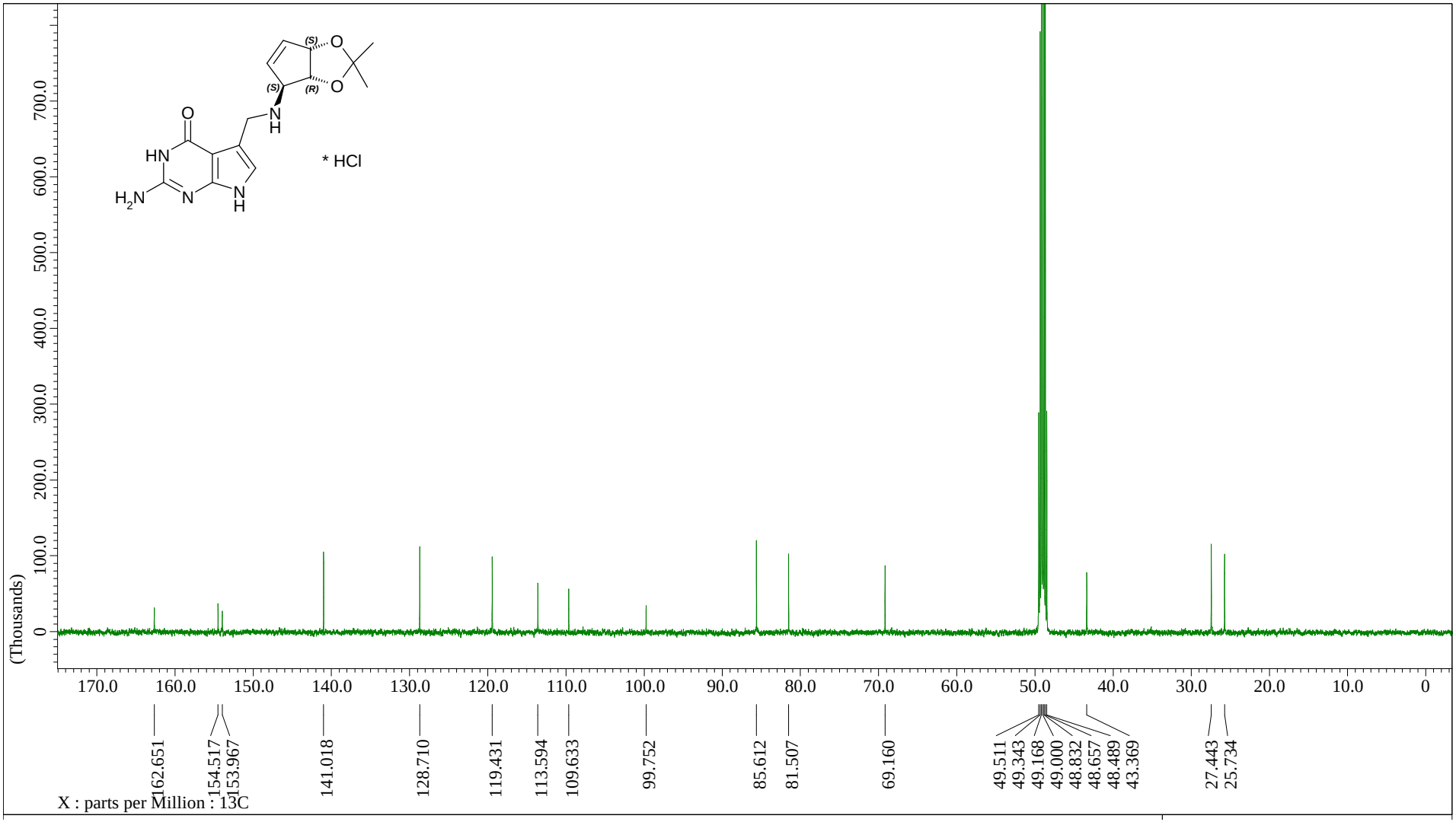
(3a*S*,4*R*,6a*S*)-4-bromo-4,6a-dihydro-3a*H*-spiro[cyclohexane-1,2-cyclopenta[*d*][1,3]dioxole (13b, $CDCl_3$)



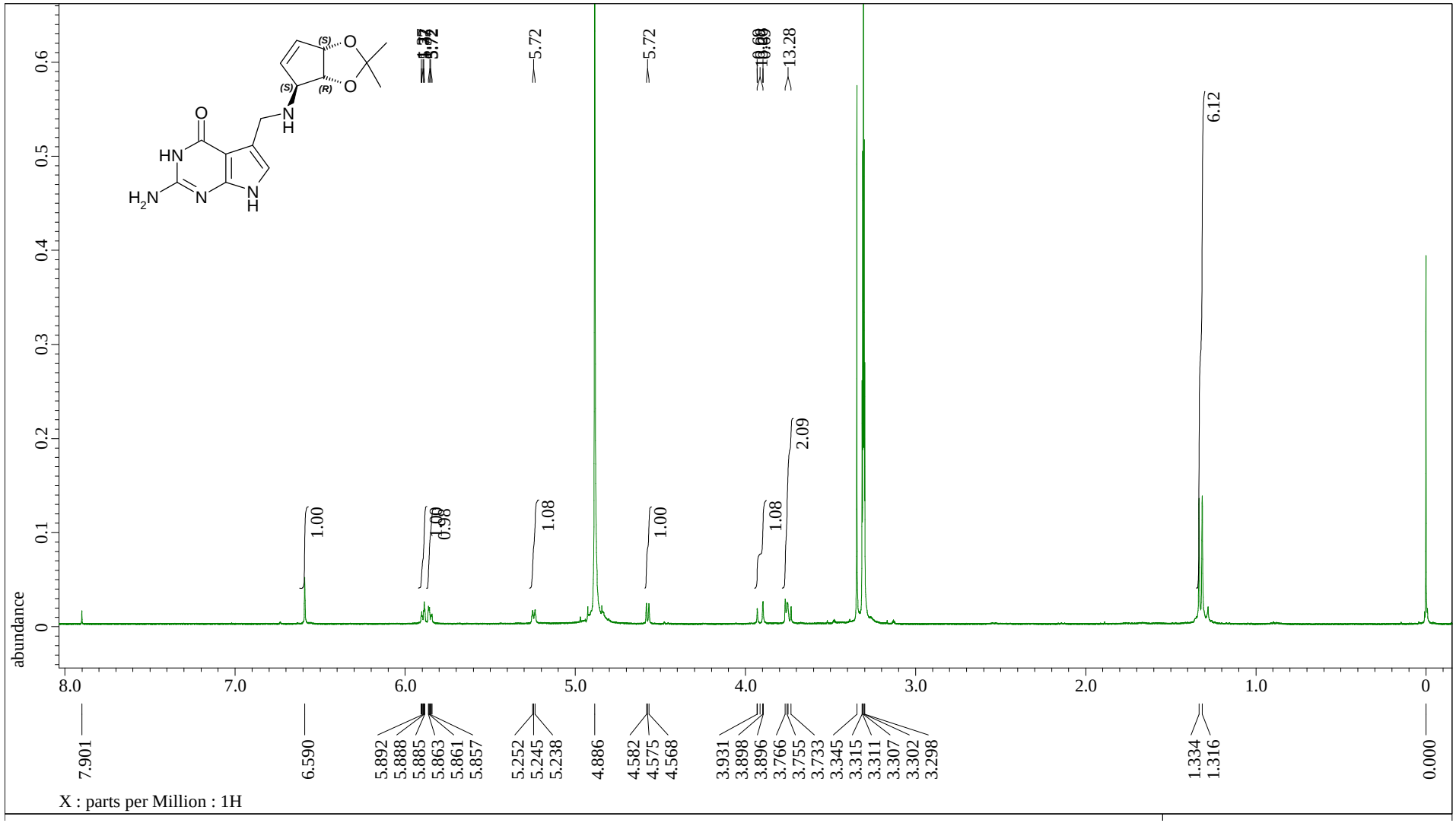
2-Amino-5({[(3a'*R*,4'*S*,6a'*S*)-2,2-dimethyl-4,6a'-dihydro-3a'*H*-cyclopenta[*d*][1,3]dioxol-4-yl]amino}methyl)-3,7-dihydro-4*H*-pyrrolo[2,3-*d*]pyrimidin-4-one hydrochloride (14a, CD_3OD)



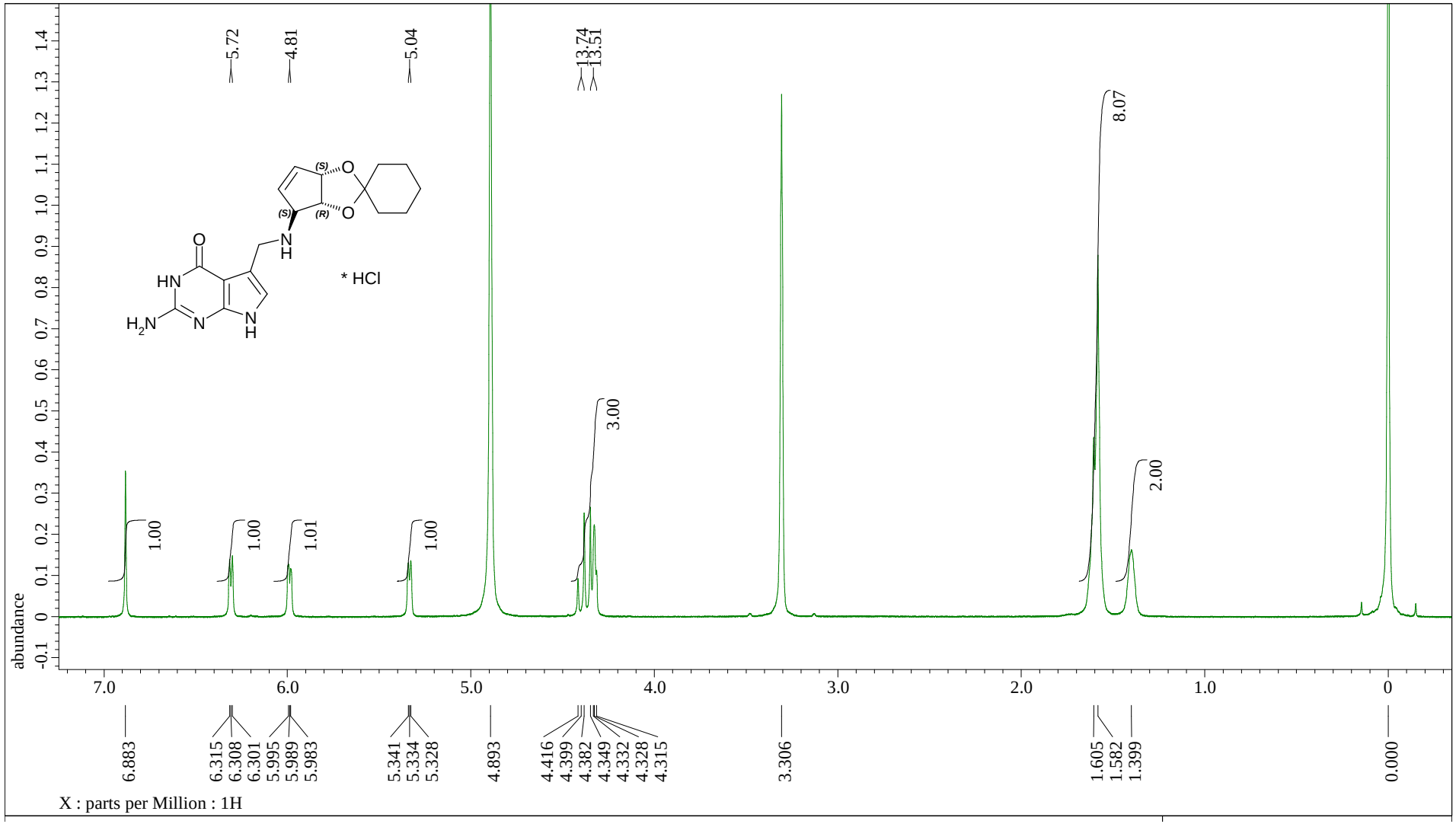
2-Amino-5({[(3a'*R*,4'*S*,6a'*S*)-2,2-dimethyl-4,6a'-dihydro-3a'*H*-cyclopenta[*d*][1,3]dioxol-4-yl]amino}methyl)-3,7-dihydro-4*H*-pyrrolo[2,3-*d*]pyrimidin-4-one hydrochloride (14a, CD_3OD)



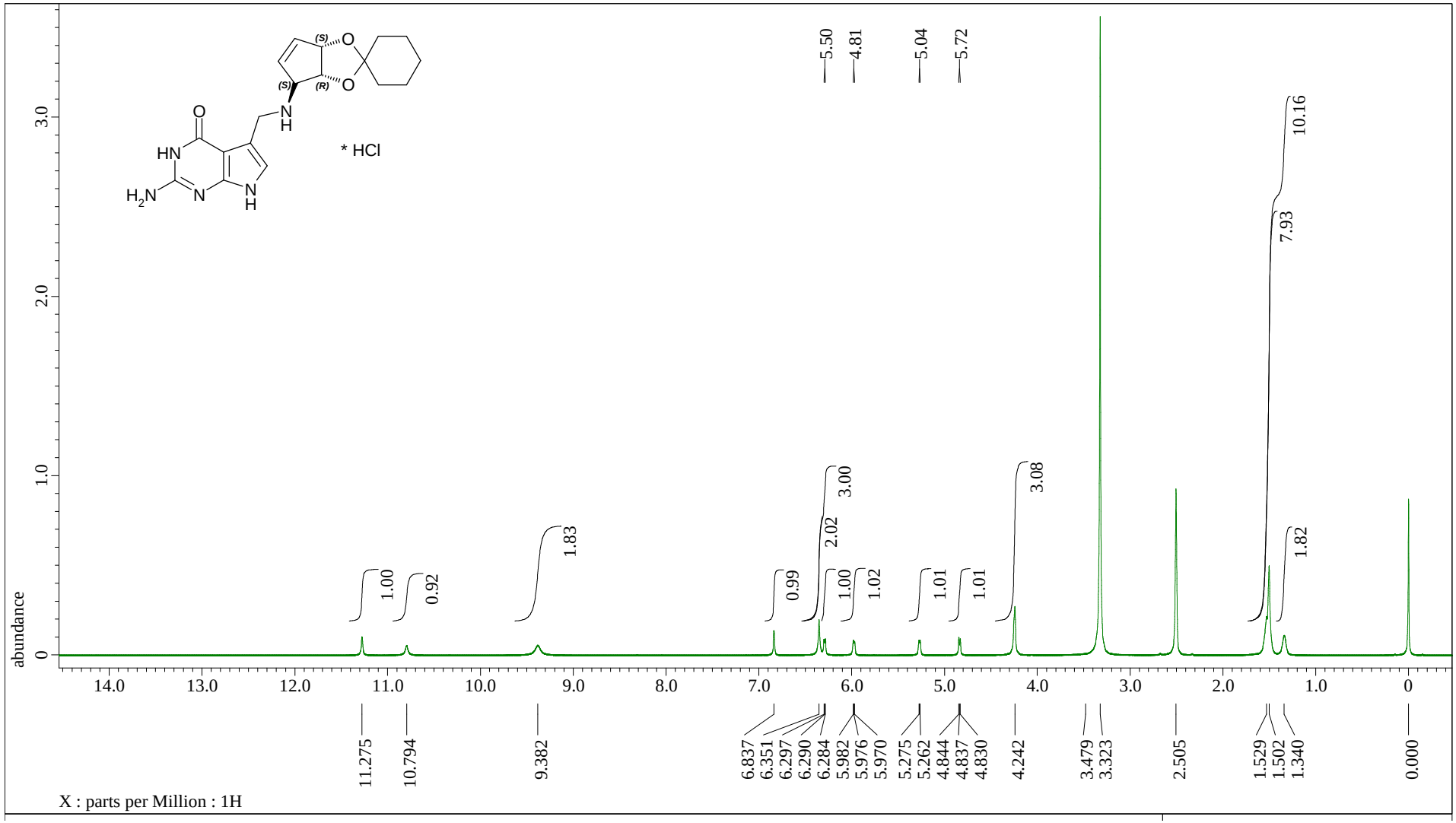
2-Amino-5({[(3a'*R*,4'*S*,6a'*S*)-2,2-dimethyl-4,6a'-dihydro-3a'*H*-cyclopenta[*d*][1,3]dioxol-4-yl]amino}methyl)-3,7-dihydro-4*H*-pyrrolo[2,3-*d*]pyrimidin-4-one (14a, CD_3OD)



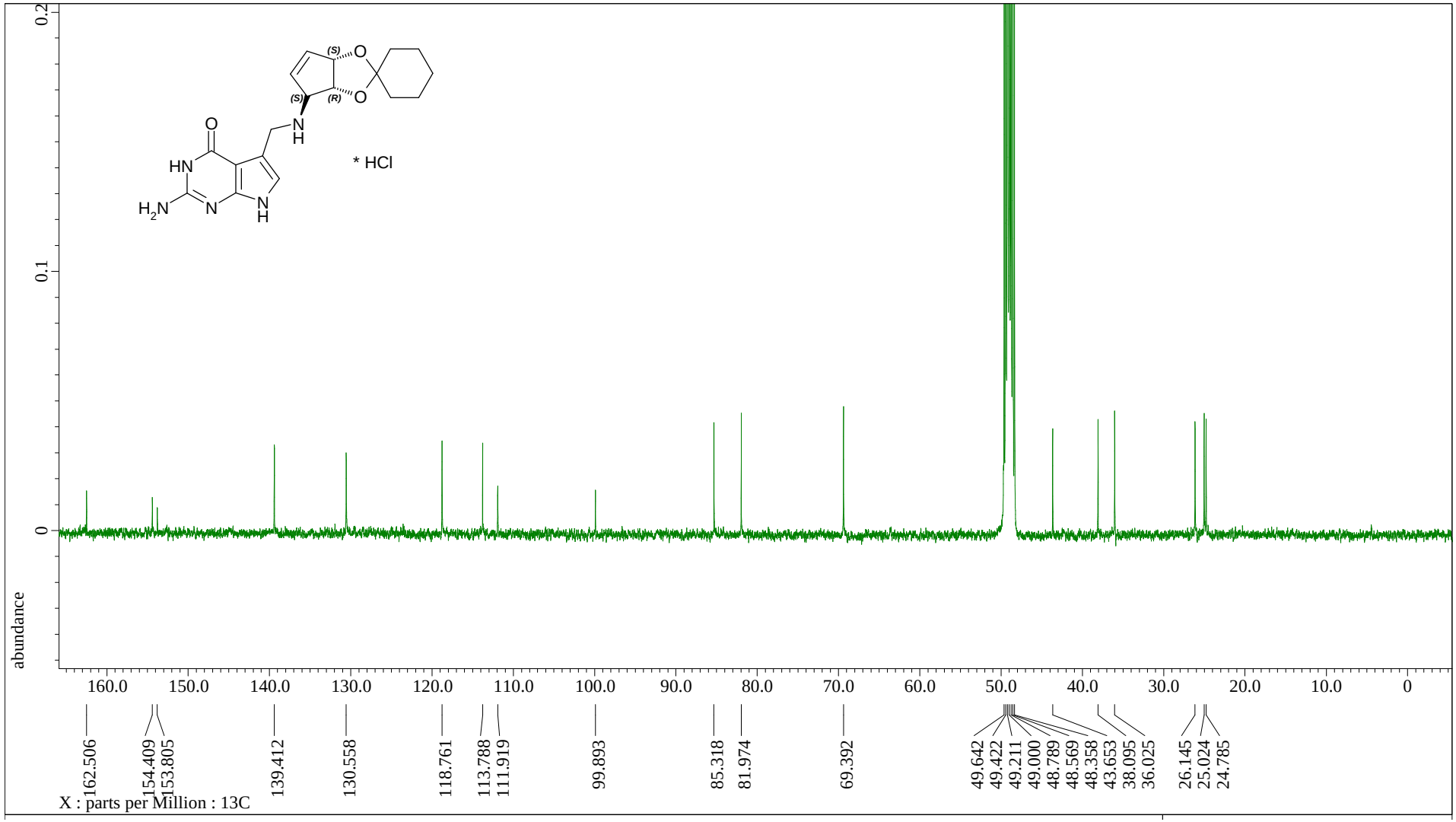
5-({[(3'a*R*, 4'*S*,6'a*S*)-4',6'a-dihydro-3'a*H*-spiro[cyclohexane-1,2'-cyclopenta[*d*][1,3]dioxole]-4'-yl]amino}methyl)-2-amino-3*H*,4*H*,7*H*-pyrrolo[2,3-*d*]pyrimidin-4-one hydrochloride (14b, CD_3OD)



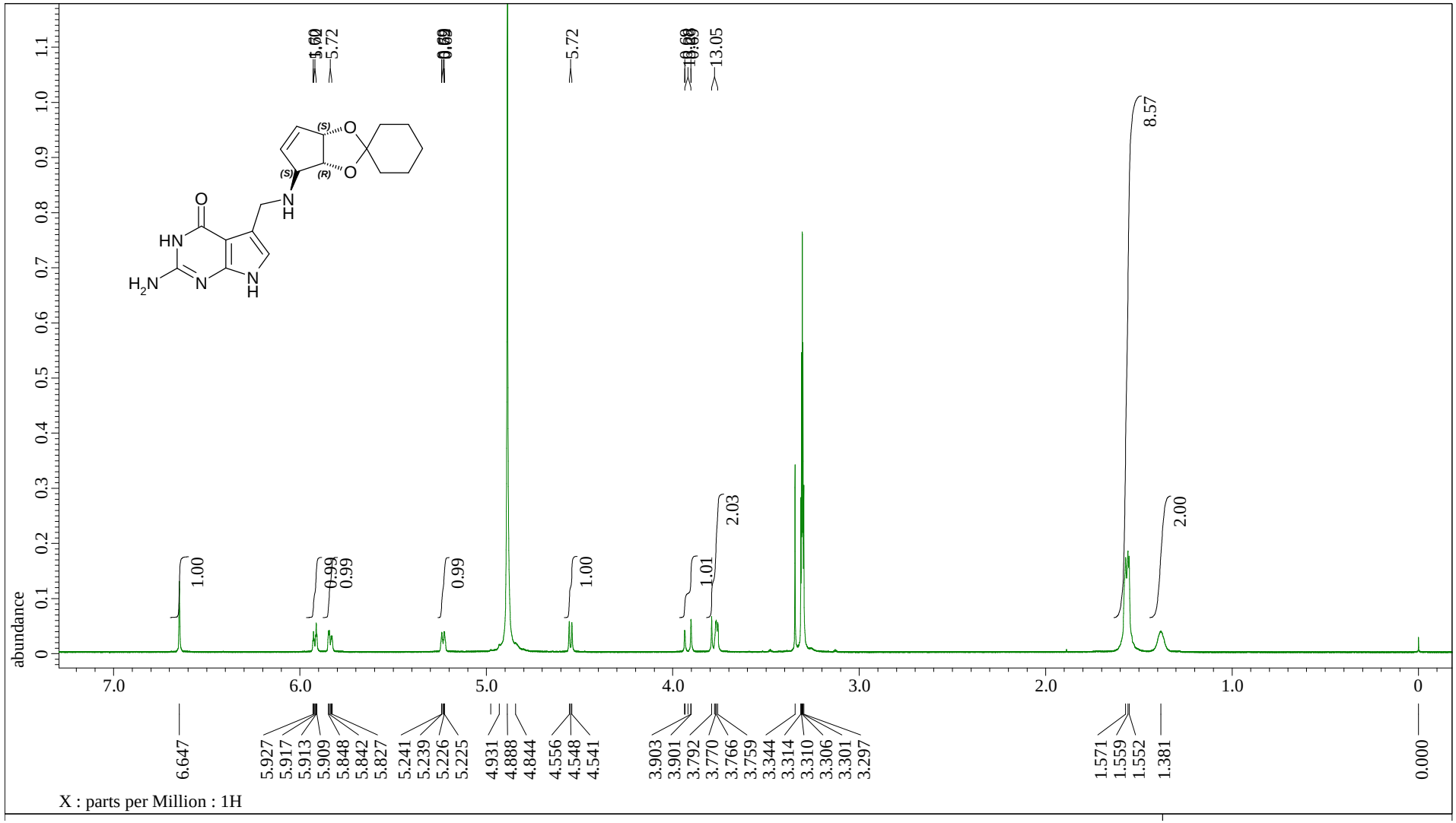
5-({[(3'a*R*, 4'*S*,6'a*S*)-4',6'a-dihydro-3'a*H*-spiro[cyclohexane-1,2'-cyclopenta[*d*][1,3]dioxole]-4'-yl]amino}methyl)-2-amino-3*H*,4*H*,7*H*-pyrrolo[2,3-*d*]pyrimidin-4-one hydrochloride (14b, DMSO-d_6)



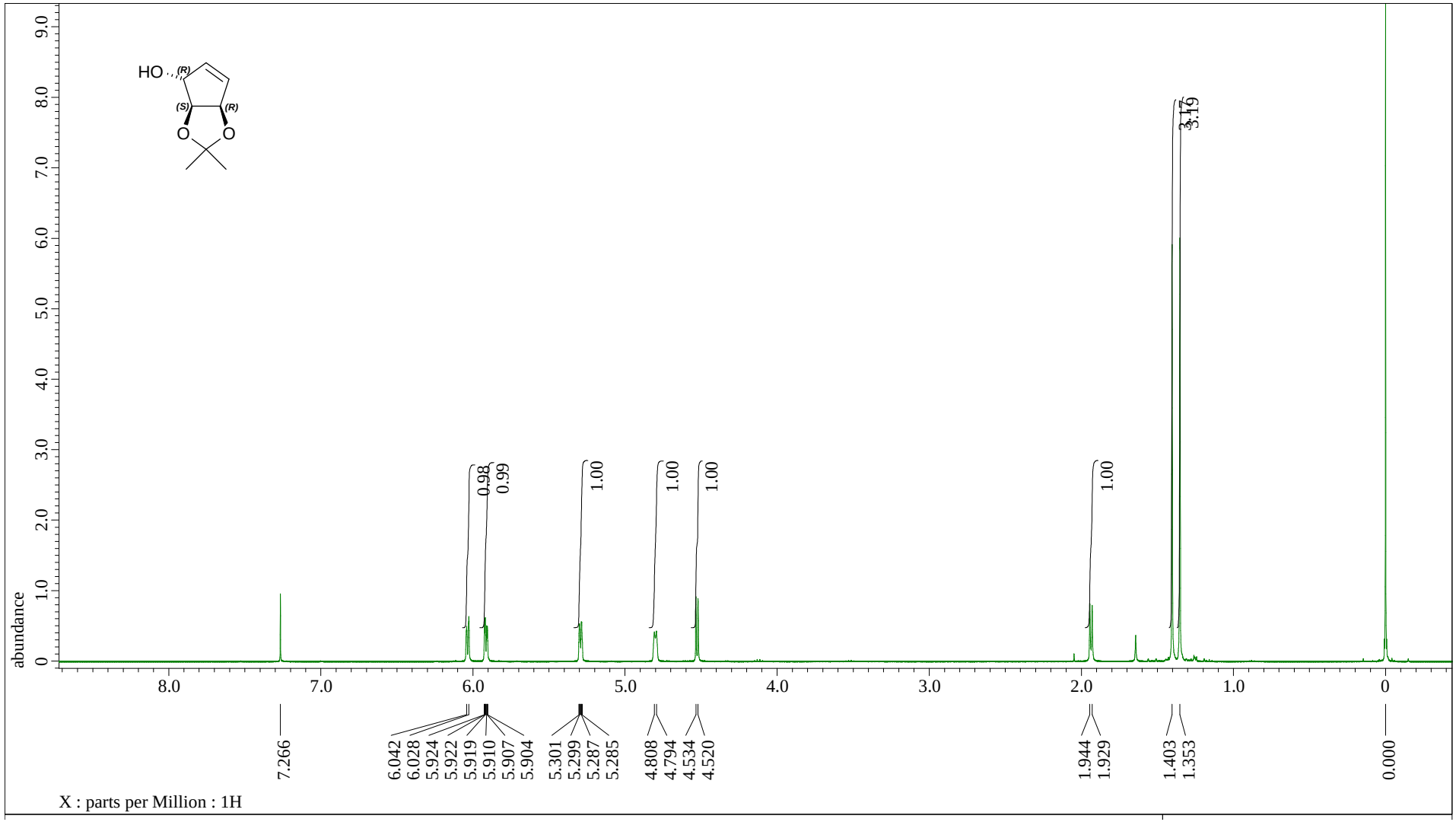
5-({[(3'a*R*, 4'*S*,6'a*S*)-4',6'a-dihydro-3'a*H*-spiro[cyclohexane-1,2'-cyclopenta[*d*][1,3]dioxole]-4'-yl]amino}methyl)-2-amino-3*H*,4*H*,7*H*-pyrrolo[2,3-*d*]pyrimidin-4-one hydrochloride (14b, CD_3OD)



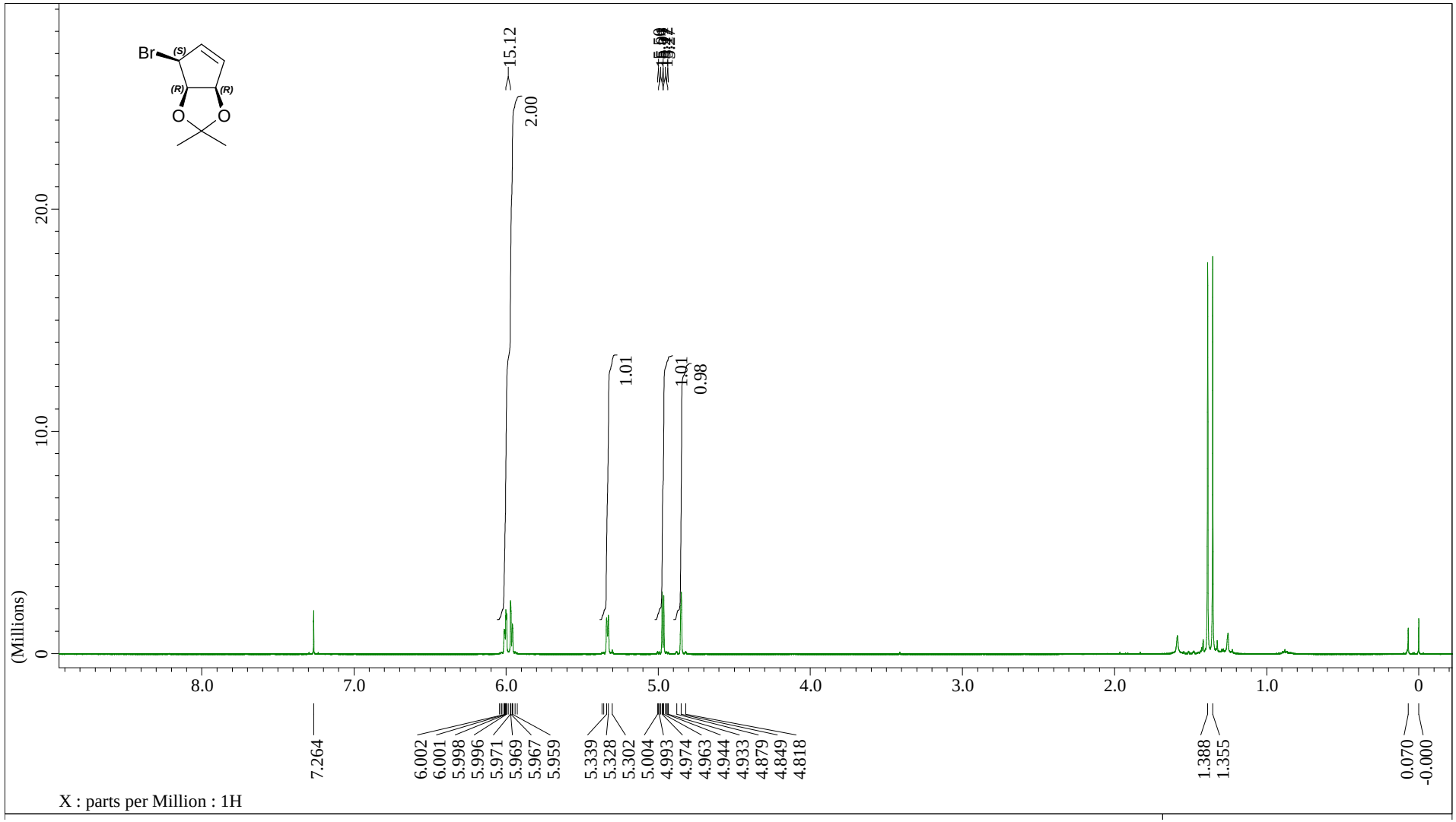
5-({[(3'a*R*, 4'*S*,6'a*S*)-4',6'a-dihydro-3'a*H*-spiro[cyclohexane-1,2'-cyclopenta[*d*][1,3]dioxole]-4'-yl]amino}methyl)-2-amino-3*H*,4*H*,7*H*-pyrrolo[2,3-*d*]pyrimidin-4-one (14b, CD_3OD)



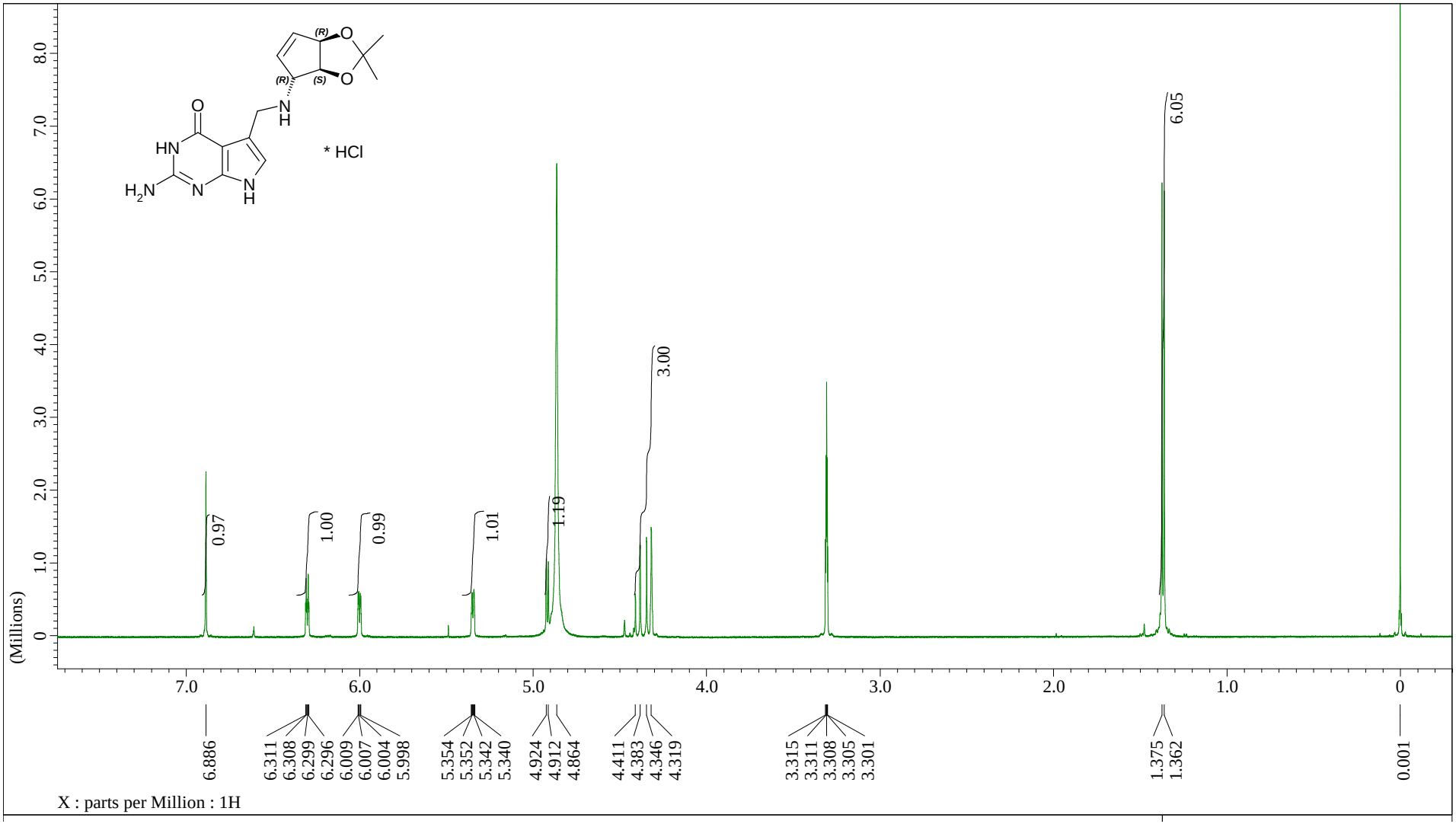
(3a*S*,4*R*,6a*R*)-2,2-dimethyl-2*H*,3a*H*,4*H*,6a*H*-cyclopenta[*d*][1,3]dioxol-4-ol (15, $CDCl_3$)



(3a*R*,4*S*,6a*R*)-4-bromo-2,2-dimethyl-2*H*,3a*H*,4*H*,6a*H*-cyclopenta[*d*][1,3]dioxole (16, $CDCl_3$)



2-Amino-5({[(3a'*S*,4'*R*,6a'*R*)-2,2-dimethyl-4,6a'-dihydro-3a'*H*-cyclopenta[*d*][1,3]dioxol-4-yl]amino}methyl)-3,7-dihydro-4*H*-pyrrolo[2,3-*d*]pyrimidin-4-one hydrochloride (17, CD_3OD)



2-Amino-5({[(3a'*S*,4'*R*,6a'*R*)-2,2-dimethyl-4,6a'-dihydro-3a'*H*-cyclopenta[*d*][1,3]dioxol-4-yl]amino}methyl)-3,7-dihydro-4*H*-pyrrolo[2,3-*d*]pyrimidin-4-one hydrochloride (17, CD_3OD)