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PAPER

Transformations of the natural cytokinin N6-isopentenyladenine in aqueous acidic media: structural aspects†

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N6-Isopentenyladenine (L1) was subjected to variously acidic media in 0.1 M, 1 M and 2 M HCl. In dependence on the acidity of the medium, the formation of three main acid hydrolysis products, involving the N6-isopentenyladeninium (HL1) (**1**), 7,8,9,10-tetrahydro-7,7-dimethyl-3*H*-pyrimido[2,1-*i*]purin-6-ium (HL2) (**2**) or 5-amino-4-(4,4-dimethyl-3,4,5,6-tetrahydropyrimidin-2-yl)-imidazolium (H_2L3) (**3–5**) cations, were determined and characterized by multinuclear solution-state NMR spectroscopy and in the solid state by single crystal X-ray analysis. The coordination abilities of these transformation products have been also investigated. The compounds of the compositions $[Zn(HL1)Cl_3]\cdot H_2O$ (**1**), $[Zn_3(HL2)_2Cl_8]$ (**2**), $(H_2L3)[CuCl_4]$ (**4**) and $(H_2L3)[ZnCl_4]$ (**5**) have been prepared in dependence on the acidity of the medium used by the reactions of L1 with $ZnCl_2\cdot 1.5H_2O$ or $CuCl_2\cdot 2H_2O$. Based on the NMR spectroscopic and X-ray crystallographic results, the mechanism of transformation of L1 in the acidic medium, involving the protonation, cyclization and ring fission, has been suggested.

Introduction

N6-isopentenyladenine (L1) belongs to a group of plant growth regulators (hormones) known as cytokinins whose main physiological role is to induce plant cell division.[1] This compound was among the first substances identified as nonspecific *cyclin-dependent kinase inhibitors* (CDKI).[2] Cyclin-dependent kinases (CDK) represent a group of serine-threonine kinase enzymes, which control the progression of cell cycle in proliferating eukaryotic cells.[3,4] CDK become active after association with their regulatory partners, *i.e.* coenzymes called cyclins. Inappropriate expression and/or mutations of CDK, cyclins, and natural cyclin-dependent kinase inhibitors (CDKI) have been found in various cancers.[5] For these reasons, CDK activity has been targeted for drug development and a number of small molecules have now been identified as pharmaceutical CDK inhibitors, which can be used for the reduction of cell division in various tumours.[6] However, N6-isopentenyladenine as CDKI was of limited interest because of its poor selectivity.[2] Later, it was found that the compounds of similar composition and structure (generally C2, N6, N9 trisubstituted derivatives of adenine) proved to act as more potent and selective CDKI. The most promising representative of the adenine-based CDKI, *roscovitine*, 2-(1-ethyl-2-hydroxyethylamino)-N6-benzyl-9-isopropyladenine, has successfully passed *in vivo* and preclinical testing and it is currently being tested (named as Seliciclib or CYC202) in the 2b-phase of clinical trials on patients with non-small cell lung cancer.[7,8]

We have focused on the investigation of the reaction course and reaction products between cytokinins, CDKI or derived compounds and simple inorganic salts of selected transition metals in dependence on different pH and molar ratio in our systematic study of these systems. The findings of our laboratory have proved that the resulting biological activity (cytotoxicity, SOD-like activity) can be substantially increased by coordination of the organic substance to a suitable transition metal centre.[9,10] The question of stability of adenine-based cytokinins in low pH has to be considered, because acidic treatments are usually performed during their isolation and purification from plant tissues. From the literature sources, we have found that the acid hydrolysis of N6-isopentenyladenine has been investigated and two products have been isolated: the hydrated derivative N6-(3-hydroxy-3-methylbut-2-enyl)adenine and, under more vigorously acidic conditions, tricyclic 7,8,9-trihydro-7,7-dimethyl-3*H*-pyrimido[2,1-*i*]purine. The products were characterised by elemental analysis, UV and ^{1}H NMR spectroscopy and mass spectrometry.[11] Ring-fission was not observed under the used conditions. The study of stability of a structurally similar compound, zeatin (2-methyl-4-(7*H*-purin-6-ylamino)but-2-en-1-ol), another natural cytokinin, has been

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† Electronic supplementary information (ESI) available: Additional data regarding the hydrogen bonding for compounds **1–5** (Table S1–S5) and crystal structures (Fig. S1–S5) of the presented compounds. CCDC reference numbers 809163–809167. For ESI and crystallographic data in CIF or other electronic format see DOI: 10.1039/c1ob05649b

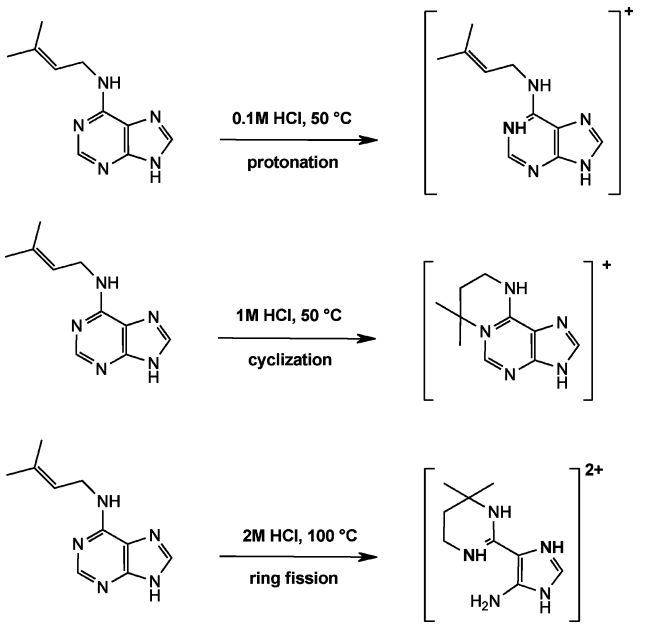
performed.[12] The transformation of zeatin in 1 M HCl at 100 °C was studied; a mixture of three products was acquired and characterized by elemental analysis, IR, UV and ^{1}H NMR spectroscopy, EI MS spectrometry and HPLC analysis. These compounds were identified as a diol resulting from the hydration of the double bond of the aliphatic chain, and as two cyclized products, a hydroxypyrrolidine and dihydropyrrole. The cyclization of the aliphatic chain to purine forming a tricyclic compound was not proved and no ring-fission was detected under the used conditions.

In general, ring-opening of different adenine derivatives has been extensively studied in various pH and temperatures, by ^{15}N isotope marking or with different influence of substituents.[13–19] Also, the ring-opening mechanism occurs in varied synthetic procedures.[20–23] For example, recently, the ring-opening reactions of 3-β-D-ribofuranosyl-3,7,8,9-tetrahydropyrimido[1,2-*i*]purin-8-ol (prepared by cyclization from adenosine and epichlorohydrin) in basic pH and of its aglycon in acidic conditions, have been reported. Both reactions afforded 2-(5-amino-1-R-imidazol-4-yl)-1,4,5,6-tetrahydropyrimidin-5-ol (R = β-D-ribofuranosyl or H).[24] This compound was reacted with CS_2 or HNO_2 leading to ring-closure which resulted in a thione, and triazine product, respectively. The physical study of the compounds was performed by HPLC, MS and NMR methods.

Although various adenine derivatives (as mentioned above, Ref. 13–24) have been studied for their behaviour in acidic conditions, their hydrolysis products have not been characterized by single crystal X-ray analysis yet. Moreover, to our best knowledge, no ring-opening products have been confirmed and/or described for adenine-type cytokinins or cytokinin-derived compounds so far. Herein, we describe the first concurrent NMR spectroscopic and X-ray crystallographic study of the transformation products of N6-isopentenyladenine (L1), in aqueous HCl in the presence of Zn(II) and Cu(II). The aims of this study included determination of the reaction conditions (especially, pH and reaction time) leading to L1 acid hydrolysis to the point of ring-opening and unambiguous elucidation of the structures of the hydrolysis products, and moreover, we also studied the coordination abilities of the L1 acid hydrolysis products to the metal ions [Cu(II) and Zn(II)].

Results and discussion

Five compounds $[Zn(HL1)Cl_3]\cdot H_2O$ (**1**), $[Zn_3(HL2)_2Cl_8]$ (**2**), $(H_2L3)Cl_2\cdot 2H_2O$ (**3**), $(H_2L3)[CuCl_4]$ (**4**) and $(H_2L3)[ZnCl_4]$ (**5**) have been obtained employing N6-isopentenyladenine (L1) and an appropriate zinc(II) or copper(II) salt as the starting compounds subjected to 0.1 M HCl (in the case of **1**), 1 M HCl (in the case of **2**) and 2 M HCl (in the cases of **3**–**5**). These variously acidic reaction conditions were used in this study because, in general, the knowledge of the stability of cytokinins and cytokinin-derived compounds is essential for determining the suitable conditions for their isolation and purification from plant tissues[12,25,26] where wide ranges of pH are commonly used (*e.g.* pH 2.9 in the process of extraction of zeatin from barley[27]). Moreover, acidic conditions are also often used during the preparation of their transition metal complexes.[28] Aqueous hydrochloric acid was used for the presented syntheses based on the information from the literature research and mainly from the studies[11,12] focused on structurally similar compounds where the authors used variously concentrated HCl for acid hydrolysis. Additionally, in our study centred on the influence of the presence of the metal in the reaction mixture on the reaction course, we used simple inorganic salts $ZnCl_2\cdot 1.5H_2O$ or $CuCl_2\cdot 2H_2O$. Therefore, using HCl in these syntheses was rational in order not to introduce a different anion (*i.e.* from another acid) into the reaction system. The course of acid hydrolysis of L1 can be observed on the prepared compounds **1**, **2** and **3**–**5**. Simple protonation at the N1 atom of the purine moiety occurs in 0.1 M HCl. When the concentration is raised to 1 M, the cyclization of the isopentenyl chain to the purine skeleton is observed, which affords the formation of the 7,8,9,10-tetrahydro-7,7-dimethyl-3*H*-pyrimido[2,1-*i*]purin-6-ium cation (HL2). Finally, when L1 is held in 2 M HCl and 100 °C, the ring-opening occurs within the pyrimidine moiety which is accompanied by the elimination of the C2 atom, thus giving the 5-amino-4-(4,4-dimethyl-3,4,5,6-tetrahydropyrimidin-2-yl)-imidazolium dication (H_2L3) (Scheme 1).



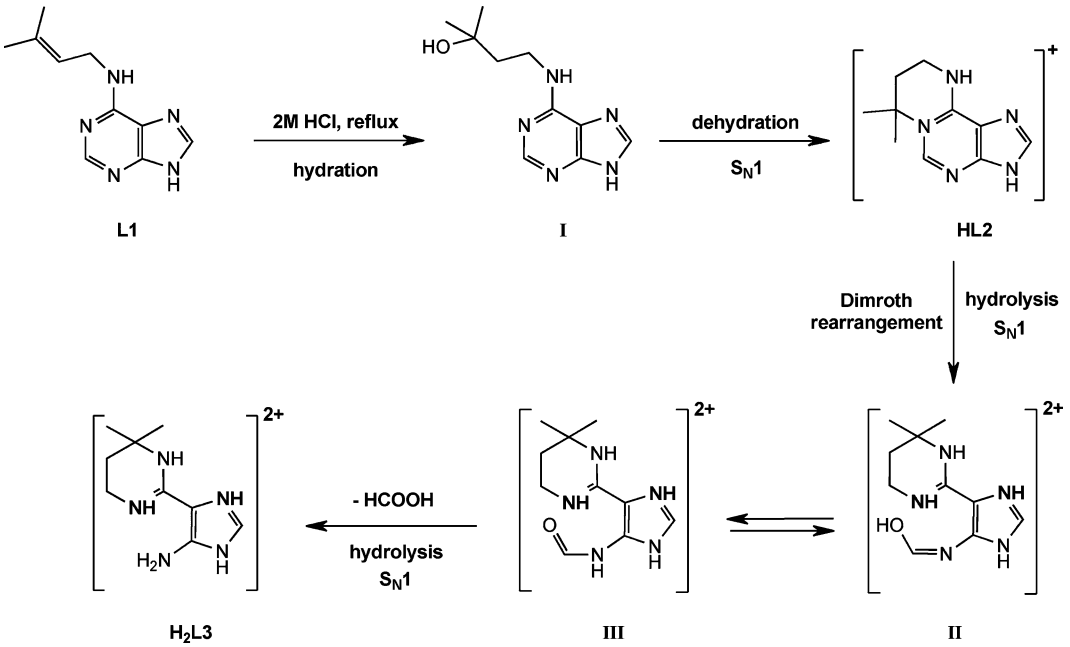
Scheme 1 A schematic representation showing the reaction conditions leading to individual acid hydrolysis transformation products of N6-isopentenyladenine.

The previously reported studies on acid hydrolysis of the title compound, cytokinin L1,[11] and the structurally related zeatin,[12] also a cytokinin, attracted our attention to the problem of acid hydrolysis in cytokinins. The authors Martin and Reese[11] described as early as in 1968 the course of acid hydrolysis of L1 up to the tricyclic compound HL2 and they suggested the ring-opening in the system, however, no direct proof of the assumption about the formation of any product resulting from pyrimidine ring fission was presented. The isolated compounds were all characterized by means of physical-chemical methods analyzing the samples in solution. Our motivation was to supply the most unambiguous evidence about the hydrolysis product structures in the solid state, which can be unequivocally accomplished only by obtaining single crystals of the compounds and determining their molecular and crystal structures by X-ray analysis. Additionally, we wanted to go one step further in the acid hydrolysis of L1 to find the

reaction conditions for successful isolation of the ring-fission hydrolysis product and confirm the structural assumptions from the literature. Metal ions Zn(II) and Cu(II) were introduced in the system because no similar study focused on the metal presence influence on acid hydrolysis has been performed up to now. We wanted to find out if there would be any differences in the reaction courses with and without the metal in the system. In our efforts to elucidate the X-ray structures of the acid hydrolysis products, we were unable to prepare single crystals of most of the organic compounds, which is why we tried adding a metal in the system and see if the resulting coordination compounds might more easily form single crystals. We wanted to analyze if any and what type of coordination compounds would form in the process of acid hydrolyses. We were confident that L1 would coordinate to the transition metals based on our previous rich experience in coordination chemistry of cytokinins and cytokinin-derived compounds.[9,10,28] No data have been found in the literature about the formation of metal complexes with the compounds formed during acid hydrolyses. The isolation of the coordination compounds of the hydrolysis products might, moreover, be interesting for future studies of biological activity (*e.g.* cytotoxicity) of these compounds. The starting title compound L1 exhibits intrinsic biological activity (see Introduction). Our experience from previous years confirms that coordination to a suitable transition metal ion can lead to significant increase in biological activity.[9,10,28] It would therefore be interesting to look at the differences in biological activity with the change of structural parameters caused by acid hydrolysis.

In determining the reaction times for isolating the hydrolysis products, the aliquots were checked by tlc. The desired termination of reactions was, when no mixture of compounds was found by tlc, so that we could identify the final hydrolysis product under the used conditions. Moreover, in our efforts to obtain single crystals of the hydrolysis products, the probability of single crystal formation is higher, if a single chemical individuum is present in the system. Slow evaporation of the solution solvent then might lead to the successful formation of single crystals suitable for X-ray analysis. This approach was, however, less successful in the case of the acid hydrolysis performed without the presence of the metals. When L1 was subjected to 0.1 M HCl for 30 min, slow evaporation of the solvent, after the termination of the reaction, afforded a white powder, whose composition was after NMR spectroscopic analysis suggested to be a mixture of three protonated L1 molecules with the proton attached to the nitrogens N7, N3 and N1 in the approximate ratio of 6 : 3 : 1. The compounds were inseparable and have almost the same R_f on tlc. On the other hand, when the same reaction was performed in the presence of $ZnCl_2 \cdot 1.5H_2O$, single crystals were obtained, and NMR in the solution and X-ray analysis in the solid state proved the formation of $[Zn(HL1)Cl_3] \cdot H_2O$ (**1**). The coordination of the organic molecule to Zn(II) *via* N7 caused the redistribution of electron density and the preferred site for protonation was therefore N1. A similar situation occurred in the case of acid hydrolysis in 1 M HCl. We were not able to find the reaction time that would lead to the formation of a single product in the system; NMR showed the tricyclic compound HL2 to be the prevailing component but no single crystals were obtained. This is in accordance with the findings of Martin and Reese,[11] who concluded that a quantitative conversion of L1 to the tricyclic HL2 occurred only under more vigorously acidic conditions, *e.g.* in aqueous alcoholic fluoroboric acid at 80° (the product isolated as a crystalline fluoroborate) or in trifluoroacetic acid at 60°. On the other hand, when $ZnCl_2 \cdot 1.5H_2O$ was present, single crystals of $[Zn_3(HL2)_2Cl_8]$ (**2**) involving the 7,8,9,10-tetrahydro-7,7-dimethyl-3*H*-pyrimido[2,1-*i*]purin-6-ium cation coordinated also *via* N7, were obtained and the molecular and crystal structures were determined. The formation of the trimetallic structure bridged by the chlorido ligands might be possibly connected to the higher concentration of the chloride anions in the reaction mixture and their high donor ability. The ring-opening did not occur under these conditions no matter what reaction time was used. More vigorously acidic conditions were needed. The only case, where we were able to isolate single crystals of the purely organic acid hydrolysis product, was from a 2-day reaction at 100 °C in 2 M HCl. The compound was identified to be the in previous studies assumed product of ring fission $(H_2L3)Cl_2 \cdot 2H_2O$ (**3**), involving the 5-amino-4-(4,4-dimethyl-3,4,5,6-tetrahydropyrimidin-2-yl)-imidazolium dication. Still, as compared to the reaction involving $ZnCl_2 \cdot 1.5H_2O$ or $CuCl_2 \cdot 2H_2O$, the acid hydrolysis without the presence of the metal took twice as long reaction time (24 h with, and 2 days without the metal ion). When we analysed the purely organic product after the reaction time of 24 h, the NMR study identified a mixture of two predominant compounds, HL2 and H_2L3. Concerning the coordination ability, the H_2L3 organic cation was not shown to coordinate to the metal atom, even if copper, with a greater variability in the coordination number than zinc, was used as the metal atom. Only the formation of ionic compounds with the complex anion $[MCl_4]^{2-}$ was observed (M = Zn, Cu). This fact might be again connected with the even higher concentration of the Cl^- anions than in the case of **2** and their high donor ability.

Based on the literature research and also results in this work, the course of acid hydrolysis of L1 in 2 M HCl can be described in four steps. The first step involves hydration of the isopentenyl double bond of the aliphatic chain following the Markovnikov's rule, thus affording the intermediate **I** (Scheme 2). We were not able to isolate and identify this intermediate **I**, but it is described in Ref. 11, where the authors, after a reaction of N6-isopentenyladenosine in HCl at 95 °C for 35 min, obtained a mixture of unmodified L1 and its hydrated analogue (**I**). This compound was characterized by ^{1}H NMR, MS, UV-Vis and MS spectroscopy. This step of hydration is followed by protonation, and consequently by the dehydration and cyclization of the aliphatic chain to the purine skeleton, which results in the formation of HL2 with a stable six-membered ring. Up to this point, the acid hydrolysis products were described by Martin and Reese,[11] but still the structures of the transformation products were elucidated indirectly from a combination of solution-state physical-chemical methods (NMR, UV-Vis, MS). Subsequently, this cationic compound HL2 undergoes hydrolysis followed by the Dimroth rearrangement, when the C10 atom shifts from the position N1 to the exocyclic N6 atom. The N1–C2 bond undergoes cleavage, which results in ring-fission, thus forming the keto-enolic tautomeric dication (**II**, **III**). The formamine group at the former C4 position of purine is formed. The existence of the intermediate **III** was suggested by Reese *et al.*, however, in their work, it was supposedly formed during alkaline hydrolysis of L1. Neither in their, nor in our work, the intermediate was isolated, which might be explained by the fact that this intermediate is probably not very



Scheme 2 A schematic representation of the suggested hydrolysis pathway of L1 in 2 M HCl.

stable and easily transforms into H_2L3. The whole process of acid hydrolysis is finalized by the elimination of formic acid and the formation of H_2L3 (see Scheme 2). The detailed characterizations by NMR spectroscopy and X-ray analysis of the isolated products are given below.

NMR spectroscopic study

An extensive NMR spectroscopic study was performed in order to elucidate the course of N6-isopentenyladenine (L1) acid hydrolysis and the composition of the hydrolysis products in the solution phase. Therefore, the reference spectra of L1 were compared with those of the isolated products of L1 acid hydrolysis, namely $[Zn(HL1)Cl_3]\cdot H_2O$ (**1**), $[Zn_3(HL2)_2Cl_8]$ (**2**), $(H_2L3)Cl_2\cdot 2H_2O$ (**3**) and $(H_2L3)[ZnCl_4]$ (**5**). The chemical shifts of the individual proton and carbon signals were assigned by the one-dimensional ^1H and ^{13}C as well as two-dimensional correlation experiments ^1H–^1H *gs*-COSY, ^1H–^{13}C *gs*-HMQC and ^1H–^{13}C *gs*-HMBC.

The proton and carbon spectra of $[Zn(HL1)Cl_3]\cdot H_2O$ (**1**) were qualitatively similar to those of N6-isopentenyladenine (L1) because the organic part of complex **1** maintains the L1 structure. Only N1-protonation and N7-coordination to the zinc(II) atom influence the chemical shifts in the spectra of **1**. The changes in the chemical shifts of the resonances can be discussed as coordination shifts, $\Delta\delta = \delta_{complex} - \delta_{ligand}$. The analysis of coordination shifts then gives crucial information leading to elucidation of protonation and coordination sites in **1**. Therefore, in the ^1H NMR spectrum of **1**, the most significant coordination shifts were found for the signals belonging to C2H (0.42 ppm) and C8H (0.67 ppm) due to N1-protonation, and coordination *via* N7, respectively. The greatest coordination shift was, however, found for the signal N6H, which was shifted downfield by 2.47 ppm. This can be explained by the incorporation of the N6H group in the intramolecular N–H···Cl hydrogen bonds (as proved by X-ray analysis, see below). The results following from the ^{13}C NMR spectrum supported the conclusions from the ^1H NMR spectrum. Due to the ligand coordination to Zn(II) *via* the N7 atom, the signals of C5 ($\Delta\delta$ = –6.75 ppm) and C8 ($\Delta\delta$ = 4.87 ppm) were most significantly shifted. Moreover, because of N1 being the protonation site, the signal of C2 was shifted upfield by 5.69 ppm and C6 upfield by 3.37 ppm. Additionally, significant coordination shifts were detected for C9 (3.93 ppm), C10 (–3.47 ppm) and C11 (3.17 ppm), which can be caused by a different orientation of the aliphatic chain in the structure of **1** and also the presence of a network of non-covalent interactions (proved in the solid state by X-ray analysis, see below) also influencing the chemical shifts of the carbon resonances.

The organic part of $[Zn_3(HL2)_2Cl_8]$ (**2**) is represented by the 7,8,9,10-tetrahydro-7,7-dimethyl-3*H*-pyrimido[2,1-*i*]purin-6-ium cation (HL2), a tricyclic acid hydrolysis product of L1 in 1 M HCl. As expected, the proton and carbon spectra of **2** differed significantly from the spectra of L1. The signals exhibited significant changes in chemical shifts. The cyclization of the aliphatic chain results in hindrance to free rotation, therefore, the two C9-attached hydrogens appeared as two distinct signals mutually shifted by 0.15 ppm, contrary to the L1 ^1H NMR spectrum where the two hydrogens exhibited the equivalent chemical shifts. Moreover, one new signal of C-attached hydrogen was observed in the spectrum of **2** because there are two hydrogens bonded to the C10 atom (there is no C10═C11 double bond, as in L1). The change of C10 from a methynyl to methenyl carbon results in a shift of the hydrogen signals towards lower frequencies. The C10Ha and C10Hb resonances were detected to be mutually shifted by 0.19 ppm. This change of the C10 carbon environment was even more significant in the ^{13}C NMR spectrum. It was clearly distinguishable from the APT (attached proton test) experiment, where the peak assignable to C10 was positive (= CH_2 group) contrary to the C10 signal in L1, which was inverted (= CH group). Moreover, the signal in the spectrum of **2** was shifted upfield by 90.23 ppm. A similarly high change in chemical shift as compared

to L1 was observed for the C11 signal (in L1 at 133.76 ppm and in **2** at 60.08 ppm), as this carbon is a part of a cycle and bonded to nitrogen in the structure of **2**.

The compounds $(H_2L3)Cl_2{\cdot}2H_2O$ (**3**) and $(H_2L3)[ZnCl_4]$ (**5**) contain the 5-amino-4-(4,4-dimethyl-tetrahydropyrimidin-2-yl)-imidazolium dication (H_2L3) which formed after L1 hydrolysis in 2 M HCl. Therefore, the NMR spectra of the two compounds are analogous apart from several chemical shift differences caused most likely by diverse network of hydrogen-bonds in both structures (confirmed also in the solid state by X-ray analysis, see below). The ring-fission results in two single-bonded heterocyclic rings, the tetrahydropyrimidine and imidazole ones. The C-attached protons on the tetrahydropyrimidine ring became equivalent and their resonances appeared at the same values of chemical shift. Also the signals of all the six methyl hydrogens were in the ^{1}H NMR spectra of **3** and **5** observed as one singlet. Additionally, the C2H signal disappeared from the ^{1}H spectra as well as the C2 signal (in the original purine ring numbering) from the carbon spectra. This is due to deformylation in the final phase of L1 acid hydrolysis (see Scheme 2). This change in the number of carbon atoms is well observable on the comparison of the ^{1}H–^{13}C *gs*-HMQC spectra of **2** and **5** which are displayed in Fig. 1.

The values of chemical shifts from the NMR spectra of all the studied compounds are listed in the Experimental section. The atom numbering used in the description of NMR spectra is consistent with the numbering in the section X-ray structures (below, Fig. 2–6). The carbon atom numbering of the organic parts in **2** and **3** is also shown in Fig. 1.

X-ray structures

In an effort to independently validate the results following from the solution-state NMR spectra regarding the L1 acid hydrolysis products, we decided to prepare single crystals of all the above-discussed products for X-ray characterizations in the solid state. The crystals of the compounds **1–5** were obtained by slow evaporation of a solvent from the reaction solution at room temperature. The detailed description of molecular and crystal structures is given below.

The structure of $[Zn(HL1)Cl_3]{\cdot}H_2O$ (1)

The crystal structure of **1** consists of one molecule of $[Zn(HL1)Cl_3]$ and one crystal water molecule (Fig. 2). The Zn(II) atom is four-coordinated by three terminal chlorido ligands and single protonated L1 molecule (HL1) forming a $ZnNCl_3$ donor set. HL1 is represented by the N1-protonated N9-tautomer coordinated to Zn *via* the N7 atom of the purine skeleton. The geometry around the Zn(II) centre can be described as a distorted tetrahedron (see angles in the Fig. 2 legend). The distortion can be quantified using a four-coordinate geometry index developed by Houser *et al.*[29] $\tau_4 = [360°–(\alpha + \beta)]/141°$ (α and β being the largest angles in the four-coordinate species); for **1** the value equals 0.89. The selected bond lengths and angles are listed in the legend to Fig. 2 and the hydrogen bond parameters are summarized in Table S1 (ESI†).

The coordinated organic ligand contains two aromatic rings, *i.e.* pyrimidine and imidazole. These rings were found to be nearly coplanar, because they form the dihedral angle of 0.81(15)°. One methyl group (C13 atom) of the isopentenyl chain of HL1 has been found disordered over two positions with the occupancy factors of 0.819(8) and 0.181(8). The molecular structure of uncoordinated N6-isopentenyladenine (L1) was previously reported.[30] Significant differences have been found after comparing the interatomic parameters of free L1 and HL1 coordinated in **1**. The angle C8–N7–C5 is significantly enlarged in **1** [104.5(3)°] as compared to free L1 [101.83(5)°] due to coordination to Zn(II). The same

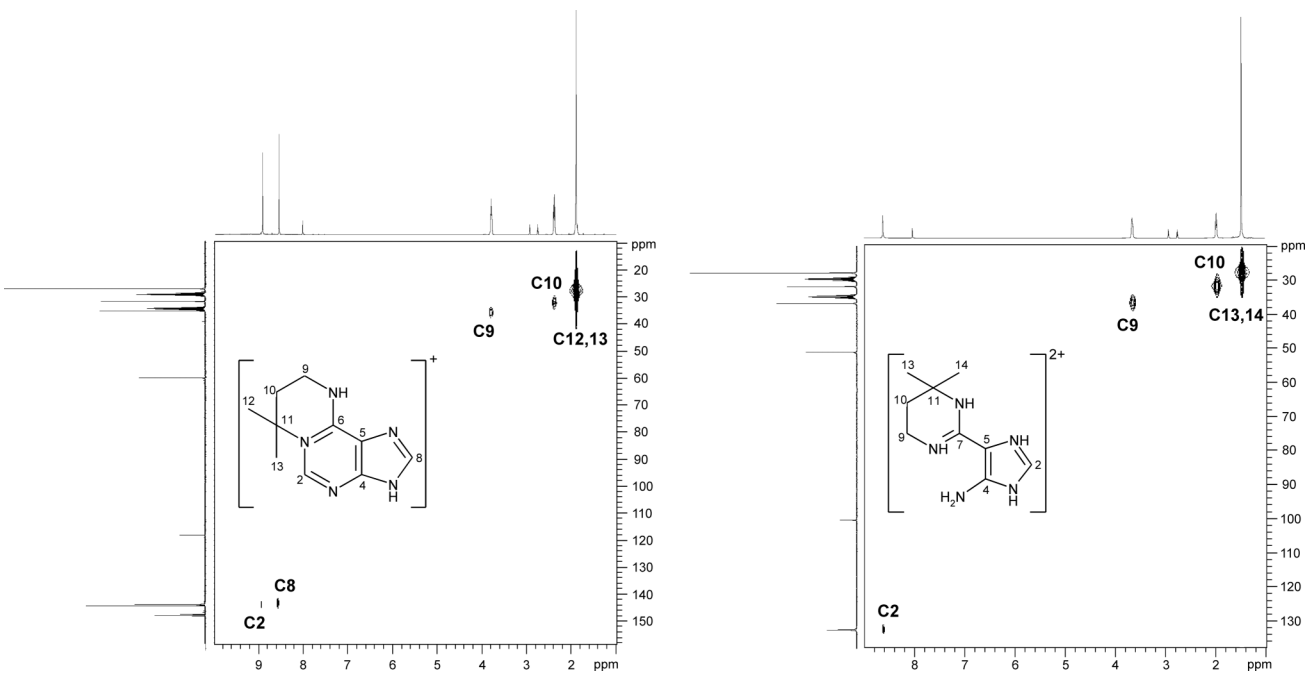


Fig. 1 The ^{1}H–^{13}C *gs*-HMQC NMR spectra of **2** (left) and **5** (right) demonstrating the change in the number of carbon atoms after ring fission (right); inset: the cationic parts of the compounds with the carbon atom numbering.

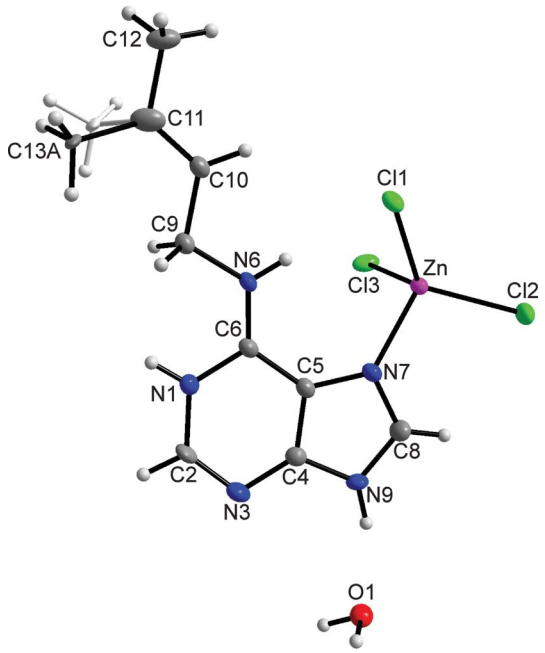


Fig. 2 The molecular structure of $[Zn(HL1)Cl_3]\cdot H_2O$ (**1**). One methyl group on the isopentenyl chain has been found disordered over two positions with the occupancy factors of 0.819(8) and 0.181(8). The atoms with the occupancy factor 0.181(8) are displayed in light grey colour for clarity reasons. The non-hydrogen atoms are displayed as displacement ellipsoids at the 50% probability level. Selected bond lengths (Å) and angles (°): Zn–N7, 2.062(3); Zn–Cl1, 2.2557(11); Zn–Cl2, 2.2778(11); Zn–Cl3, 2.2223(11); N1–C2, 1.368(5); N3–C4, 1.350(6); N7–C8, 1.327(5); N9–C8, 1.345(6); N7–Zn–Cl1, 108.28(10); N7–Zn–Cl2, 101.63(10); N7–Zn–Cl3, 104.85(10); Cl1–Zn–Cl2, 105.60(4); Cl1–Zn–Cl3, 118.35(4); Cl2–Zn–Cl3, 116.62(5); C8–N7–Zn, 120.7(3); C8–N7–C5, 104.5(3); C8–N9–C4, 107.4(4); C2–N1–C6, 124.1(4); C2–N3–C4, 112.0(4).

result arises from protonation at N1; the angle C2–N1–C6 equals 124.1(4)° in **1** and 119.05(6)° in L1. Moreover, due to hydrogen bonding, in which protons attached to N9 and N6 are involved, the angles at these sites decrease in comparison to free L1 [C8–N9–C4 equals 107.4(4)° in **1** and 103.69(5)° in L1; and C6–N6–C9 is 122.6(3)° in **1** and 123.6(5)° in L1]. Additionally, the aliphatic chain bonded at N6 has been found to be differently oriented in both discussed compounds, because the torsion angles C6–N6–C9–C10 in **1** and L1 differ significantly; 172.73(35)° as compared to 94.08(71)° in **1**, and L1, respectively.

12 molecular structures have been found in the Cambridge Structural Database (CSD),[31] which contain an N7-coordinated adenine derivative coordinated to Zn(II) in the $ZnCl_3N$ donor set. The average Zn–Cl and Zn–N7 bond lengths in the compounds were calculated to be 2.246(19) Å and 2.064(23) Å, respectively. These dimensions are quite comparable with those found in **1**, where the Zn–Cl bond lengths are equal to 2.2223(11)–2.2778(11) Å and the Zn–N7 bond length to 2.062(3) Å.

The crystal structure of **1** has been found to be stabilized by hydrogen bonds of the types N–H···Cl, N–H···O and O–H···Cl (Fig. S1). Also, the non-covalent interactions C–H···Cl, C–H···O and C···Cl contribute to stabilization of the crystal structure [C9 ···Cl1^{ii} = 3.841(4) Å, C8 ···Cl3^{iii} = 3.515(5) Å, C13A ···O1^{iv} = 3.550(7) Å, C6···Cl1^{v} = 3.372(4) Å; symmetry codes: (ii) 1 – *x*, –0.5 + *y*, 0.5 – *z*; (iii) –*x*, 1 – *y*, –*z*; (iv) 1 + *x*, 0.5 – *y*, 0.5 + *z*; (v) 1 – *x*, 1 – *y*, –*z*]. The figure showing a part of the crystal structure of **1** (Fig. S1) is presented in ESI.†

The structure of $[Zn_3(HL2)_2Cl_8]$ (2)

The molecular structure of **2** (Fig. 3) is comprised of one trimetallic Zn(II) complex $[Zn_3(HL2)_2Cl_8]$ with two bridging and six terminal chlorido ligands, and two terminal N-donor organic ligands, *i.e.* the 7,8,9,10-tetrahydro-7,7-dimethyl-3*H*-pyrimido[2,1-*i*]purin-6-ium cations (HL2). All the three Zn(II) atoms are tetrahedrally coordinated, the central Zn(II) centre in a $ZnCl_4$ donor set, while the other two terminal Zn(II) atoms in $ZnNCl_3$ donor sets. The Zn(II) atoms are mutually separated by 3.780(4) Å [Zn1 ··· Zn2] and 3.885(4) Å [Zn2 ··· Zn3], while the terminal ones are distanced by 7.285(5) Å [Zn1 ··· Zn3]. The four-coordinate geometry indexes[29] are equal to 0.88, 0.93, and 0.91 for the geometry around Zn1, Zn2 and Zn3, respectively. The selected bond lengths and angles are given in the legend to Fig. 3 and the hydrogen bond parameters are summarized in Table S2 (ESI†).

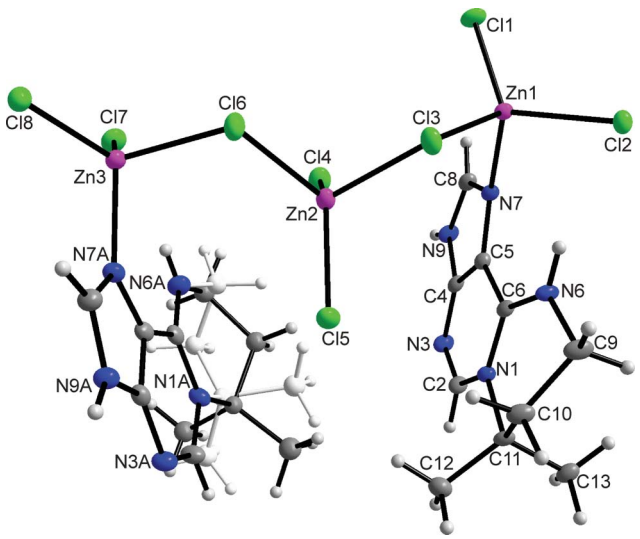


Fig. 3 The molecular structure of $[Zn_3(HL2)_2Cl_8]$ (**2**). A part of the 3,4,5,6-tetrahydropyrimidine ring in HL2A has been found disordered over two positions with the occupancy factors 0.823(5) and 0.177(5). The atoms with the occupancy factor 0.177(5) are displayed in light grey colour for clarity reasons. The non-hydrogen atoms are displayed as displacement ellipsoids at the 50% probability level. Selected bond lengths (Å) and angles (°): Zn1–N7, 2.040(2); Zn1–Cl1, 2.1975(6); Zn1–Cl2, 2.2660(6); Zn1–Cl3, 2.3059(6); Zn2–Cl4, 2.2018(7); Zn2–Cl5, 2.2278(7); Zn2–Cl6, 2.3399(7); Zn2–Cl3, 2.3576(6); Zn3–N7A, 2.044(2); Zn3–Cl7, 2.2137(7); Zn3–Cl8, 2.2316(7); Zn3–Cl6, 2.3227(7); N7–Zn1–Cl1, 108.60(6); N7–Zn1–Cl3, 104.90(6); Cl1–Zn1–Cl2, 120.95(2); Cl4–Zn2–Cl5, 113.49(3); Cl4–Zn2–Cl6, 111.02(3); Cl4–Zn2–Cl3, 114.13(3); Cl6–Zn2–Cl3, 94.48(2); N7A–Zn3–Cl7, 112.70(6); N7A–Zn3–Cl8, 105.75(6); Cl7–Zn3–Cl8, 118.86(3); Cl7–Zn3–Cl6, 111.38(3).

The two coordinated organic cations (further denoted as HL2 and HL2A) contain three heterocyclic rings, *i.e.* pyrimidine (A), imidazole (B) and 3,4,5,6-tetrahydropyrimidine (C). The rings A and B are almost coplanar forming dihedral angles of 2.59(8)° and 2.81(8)° in HL2, and HL2A, respectively. A part of the ring C in HL2A (C9, C10, C11, C12, C13 and the attached hydrogen atoms) has been found disordered over two positions with the occupancy factors of 0.823(5) and 0.177(5). The only molecular

structure deposited in CSD[31] containing the same heterocyclic rings, yet differently substituted and protonated on a different nitrogen, is 7,8,9,10-tetrahydro-7,8-dihydroxy-7,9-dimethyl-3*H*-pyrimido[2,1-*i*]purine-9-carboxylate dihydrate.[32]

The Zn(II) ions in the structure of **2** are connected by chlorido bridges. Chloride, and halide in general, anions often act as bridging ligands due to their high donor abilities connected to the presence of four lone pairs in the valence shell. For this reason, they can connect up to four metal centres forming polynuclear species. The $[Zn_2Cl_6]^{2-}$ anion can be named as a simple representative of a zinc compound involving chlorido bridges. There are two Cl^- anions bridging the two metal centres and four terminal chlorido ligands. The search in CSD[31] showed that 32 X-ray structures have already been deposited in the database containing this complex anion. Moreover, the CSD database also contains 2 X-ray structures[33,34] in which three Zn(II) centres are bridged by one chlorido ligand and 3 X-ray structures[35–37] where one Cl^- anion bridges four Zn(II) atoms. The specific search in CSD for a trimetallic moiety involving three Zn(II) centres one-dimensionally bridged by the chlorido ligands, as present in **2**, showed that no molecular structures of such trinuclear Zn(II) complexes have been determined up to now. Therefore, the molecular structure of **2** presented herein represents the first complex of this kind. Only 11 polymeric and polynuclear Zn(II) complexes involving at least three metal centres with chlorido bridges have been deposited in the database. The calculated average Zn–Cl(bridging) bond length was found to be 2.348(59) Å. The Zn–Cl(bridging) bond lengths in **2** equal 2.3059(6)–2.3576(6) Å and thus are quite comparable with the average bond length. In general, it can be said that the Zn–Cl(bridging) are significantly longer than the Zn–Cl(terminal) ones, which equal 2.1975(6)–2.2660(6) Å and are in good agreement with those in **1** [2.2218(12)–2.2777(12) Å].

N–H···Cl and N–H···N hydrogen bonds stabilize the crystal structure of **2** (Fig. S2). Furthermore, the non-covalent contacts of the types C–H···Cl and C···Cl are present within the crystal structure of **2** [C12A···Cl5^{i} = 3.730(4) Å, C10···Cl4^{iv} = 3.746(3) Å, C13···Cl4^{iv} = 3.720(3) Å, C8···Cl1^{v} = 3.543(3) Å, C13A···Cl6^{vi} = 3.682(4) Å; C8A···Cl7^{iii} = 3.416(3) Å, C5···Cl2^{vii} = 3.375(2) Å, C6···Cl2^{vii} = 3.255(2) Å; symmetry codes: (i) −1 + *x*, *y*, *z*; (iii) 1 − *x*, 1 − *y*, 1 − *z*; (iv) *x*, −1 + *y*, *z*; (v) 1 − *x*, 1 − *y*, −*z*; (vi) −1 + *x*, −1 + *y*, *z*; (vii) 1 − *x*, −*y*, −*z*]. The figure displaying a part of the crystal structure of **2** (Fig. S2) is presented in ESI.†

The structure of $(H_2L3)Cl_2\cdot 2H_2O$ (3)

The asymmetric unit of **3** consists of one doubly protonated 5-amino-4-(4,4-dimethyl-3,4,5,6-tetrahydropyrimidin-2-yl)-imidazolium cation (H_2L3), two chloride anions and two water molecules of crystallization (Fig. 4). Selected bond lengths and angles are summarized in the legend to Fig. 4 and the hydrogen bond parameters are given in Table S3 (ESI†).

The H_2L3 cation contains two heterocyclic ring systems, *i.e.* imidazole and 3,4,5,6-tetrahydropyrimidine. The imidazole ring only slightly deviates from planarity with the maximum deviation from the mean planes being 0.005(2) Å for the atom C2. The dihedral angle between the planes created through the two ring systems was found to equal 35.44(5)°. Based on the search in CSD,[31] no molecular structure containing the 4-(3,4,5,6-tetrahydropyrimidin-2-yl)-imidazole moiety has been deposited in this database.

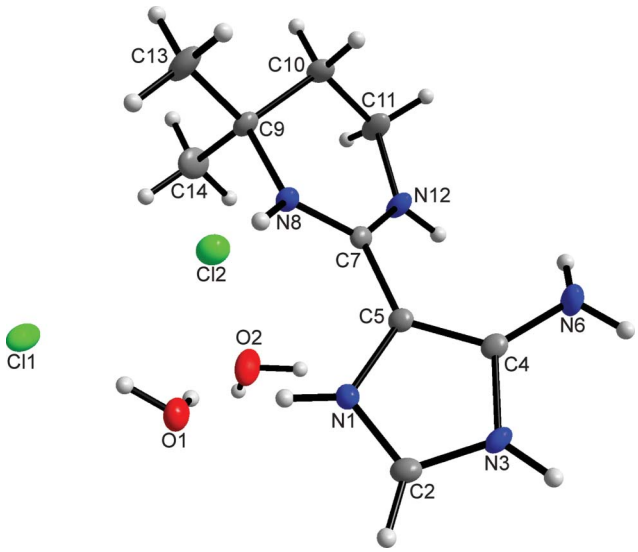


Fig. 4 The molecular structure of $(H_2L3)Cl_2\cdot 2H_2O$ (**3**). The non-hydrogen atoms are displayed as displacement ellipsoids at the 50% probability level. Selected bond lengths (Å) and angles (°): N1–C2, 1.3123(19); N3–C4, 1.3804(18); N8–C7, 1.3151(19); N12–C11, 1.4704(18); C2–N1–C5, 109.03(12); C2–N3–C4, 109.66(12); C7–N8–C9, 121.54(12); C7–N12–C11, 123.43(12).

An extensive network of hydrogen bonds was found to be present in the crystal structure of **3** (Fig. S3). Additionally, the non-covalent contacts also stabilize the crystal structure of **3**, *i.e.* C–H···Cl [C14···Cl2^{iv} = 3.704(2) Å], C–H···O [C2···O2^{v} = 3.440(2) Å] and C···Cl [C4···Cl2^{i} = 3.386(14) Å, C5···Cl2^{i} = 3.258(14) Å; symmetry codes: (i) 1 + *x*, *y*, *z*; (iv) 0.5 + *x*, 1.5 − *y*, −0.5 + *z*, (v) −0.5 + *x*, 1.5 − *y*, −0.5 + *z*]. The figure showing a part of the crystal structure of **3** (Fig. S3) is presented in ESI.†

The structure of $(H_2L3)[CuCl_4]$ (4)

The molecular structure (Fig. 5) of **4** is comprised of the doubly protonated 5-amino-4-(4,4-dimethyl-3,4,5,6-tetrahydropyrimidin-2-yl)-imidazolium cation and its positive charge is compensated by the presence of the $[CuCl_4]^{2-}$ complex anion. The Cu(II) atom is four coordinated by four terminal chlorido ligands. The geometry around Cu(II) is distorted tetrahedral. All the angles around the Cu(II) centre are very significantly different from the angle in "ideal tetrahedron", *i.e.* 109°47′ (see Fig. 5 legend). The compound **4** has a τ_4 value of 0.52. The hydrogen bond parameters are listed in Table S4 (ESI†).

The differences of the bond lengths and angles in H_2L3 in **3** and **4** are caused by diverse types of hydrogen bonds involved in the two structures. The greatest difference was found for the angle at N2 [N8 in **3**] which equals 124.9(2)° in **4** and 121.54(12)° in **3**. It is caused by the presence of the N8–H8···Cl2 hydrogen bond in the structure of **3**. The two heterocyclic rings, *i.e.* imidazole and 3,4,5,6-tetrahydropyrimidine, were found to be differently mutually oriented than in **3**. The dihedral angle between the planes through the ring systems was determined to equal 40.25(10)°. As for the $[CuCl_4]^{2-}$ anion, 580 compounds containing the $CuCl_4$ moiety deposited in CSD have been found. The average

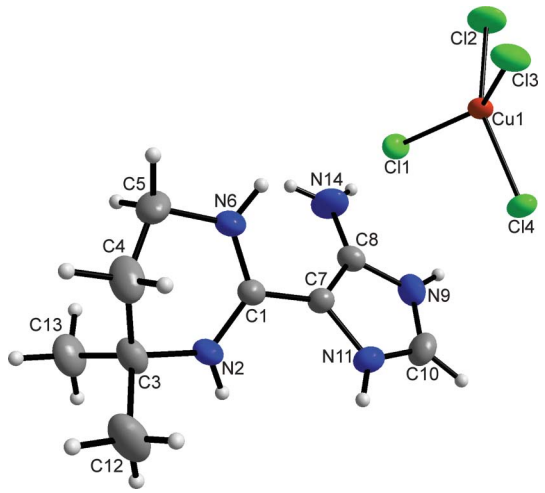


Fig. 5 The molecular structure of $(H_2L3)[CuCl_4]$ (**4**). The non-hydrogen atoms are displayed as displacement ellipsoids at the 50% probability level. Selected bond lengths (Å) and angles (°): Cu1–Cl1, 2.2733(8); Cu1–Cl2, 2.2121(8); Cu1–Cl3, 2.2741(8); Cu1–Cl4, 2.2382(7); N2–C3, 1.485(4); N6–C5, 1.455(4); N9–C8, 1.376(4); N11–C7, 1.398(3); N14–C8, 1.338(4); Cl1–Cu1–Cl3, 138.13(3); Cl1–Cu1–Cl2, 96.37(3); Cl2–Cu1–Cl4, 149.59(3); Cl3–Cu1–Cl4, 94.70(3); C1–N2–C3, 124.9(2); C1–N6–C5, 121.3(2); C8–N9–C10, 109.8(2); C7–N11–C10, 109.2(2).

Cu–Cl bond length was calculated to be 2.38(20) Å. This value is significantly longer than those found for **4** [2.2121(8)–2.2741(8) Å]. This could be caused by the fact, that greater variety of Cu–Cl bond types is involved in the average value, *i.e.* compounds containing the $CuCl_4$ moiety in other than tetrahedral geometry (octahedron *etc.*).

The crystal structure is stabilized by hydrogen bonds of the N–H···Cl type. Moreover, there are non-bonding interactions of the type C–H···Cl [C10···Cl2^{iii} = 3.552(3) Å, C10···Cl3^{iii} = 3.515(3) Å, C13···Cl2^{iv} = 3.876(4) Å; symmetry codes: (iii) x, $-1 + y$, z; (iv) $1 - x$, $2 - y$, $1 - z$]. The figure showing a part of the crystal structure of **4** (Fig. S4) is given in ESI.†

The structure of $(H_2L3)[ZnCl_4]$ (5)

The asymmetric unit of **5** consists of the 5-amino-4-(4,4-dimethyl-tetrahydropyrimidin-2-yl)-imidazolium dication and the $[ZnCl_4]^{2-}$ complex anion (Fig. 6). There are two crystallographically independent molecules of **5** in the unit cell. The geometry around the Zn(II) centre can be described as a distorted tetrahedron. The distortion from the regular tetrahedron is far smaller than that of the $[CuCl_4]^{2-}$ anion in the structure of **4**. The compound **5** has a τ_4 value of 0.95 for both crystallographically independent molecules. The selected interatomic parameters are listed in the legend to Fig. 6 and the hydrogen bond parameters are in Table S5 (ESI†).

If we compare the interatomic parameters of H_2L3 in **3** and **5**, we find significant differences which are caused by different hydrogen bonding system. The biggest difference, like in the case of **4**, was found for the C7–N12–C11 angle [121.54(12)° and 123.6(3)° in **3** and **5**]. The heterocyclic rings, *i.e.* imidazole and 3,4,5,6-tetrahydropyrimidine, present in the two crystallographically independent molecules, have been found to be differently mutually oriented, they form dihedral angles of 28.69(10)° in H_2L3 and 39.70(10)° in H_2L3'. These values also differ significantly from the

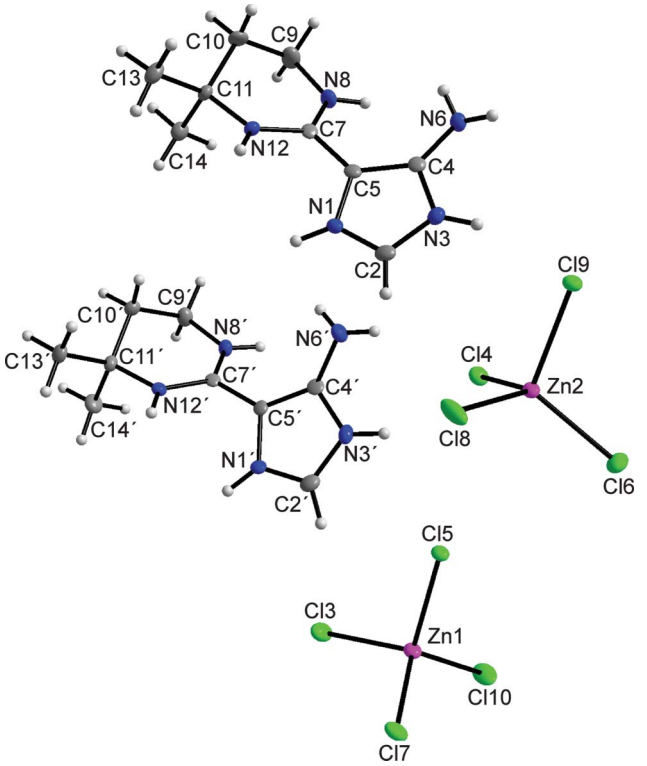


Fig. 6 The molecular structure of $(H_2L3)[ZnCl_4]$ (**5**), showing the two crystallographically independent molecules in the unit cell. The non-hydrogen atoms are displayed as displacement ellipsoids at the 50% probability level. Selected bond lengths (Å) and angles (°) for the molecule containing Zn2: Zn2–Cl4, 2.2881(8); Zn2–Cl6, 2.2270(7); Zn2–Cl8, 2.2700(8); Zn2–Cl9, 2.3349(7); N1–C2, 1.310(4); N6–C4, 1.354(4); N8–C7, 1.327(4); N12–C11, 1.488(4); Cl6–Zn2–Cl9, 109.93(3); Cl6–Zn2–Cl8, 114.86(3); Cl8–Zn2–Cl9, 103.38(3); Cl4–Zn2–Cl6, 112.91(3); C2–N1–C5, 109.7(2); C2–N3–C4, 109.9(2); C7–N8–C9, 122.7(3); C7–N12–C11, 123.6(3).

dihedral angle in **3** [35.44(5)°]. The search in CSD showed that 479 compounds have been deposited therein, which contain the $ZnCl_4$ moiety. The average calculated Zn–Cl bond length equals 2.271(32) Å. This value correlates well with the bond lengths found in **5**, 2.2360(8)–2.3202(7) Å.

A great number of hydrogen bonds of the type N–H···Cl have been found to contribute to stabilization of the crystal structure of **5** (Fig. S5). Furthermore, the non-covalent contacts of the types C–H···Cl [C9′···Cl6^{iii} = 3.699(4) Å, C9′···Cl10^{iii} = 3.611(4) Å, C14···Cl10^{iii} = 3.634(3) Å, C10′···Cl6^{v} = 3.814(3) Å, C13···Cl6^{vi} = 3.891(4) Å] and C···Cl [C7′···Cl7^{ii} = 3.427(3) Å; symmetry codes: (ii) $-x$, $1 - y$, $1 - z$; (iii) $1 - x$, $1 - y$, $1 - z$; (v) x, y, $-1 + z$; (vi) $-x$, $2 - y$, $1 - z$]. The figure showing a part of the crystal structure of **5** (Fig. S5) is presented in ESI.†

Experimental

Materials and general methods

$ZnCl_2 \cdot 1.5H_2O$, $CuCl_2 \cdot 2H_2O$, N6-isopentenyladenine and solvents were purchased from commercial sources (Sigma–Aldrich Co., Lachema Co.) and were used as received. Elemental analyses (C, H, N) were performed on a Fisons EA-1108 CHNS-O Elemental Analyzer (Thermo Scientific).

NMR spectroscopy

^{1}H and ^{13}C NMR spectra were recorded on a 400 MHz Varian spectrometer at 300 K in deuterated *N*,*N*′-dimethylformamide (DMF-d_7). The internal reference standard used for ^{1}H and ^{13}C NMR measurements was tetramethylsilane (TMS). *J* values are given in Hz. The individual ^{1}H and ^{13}C signals were assigned by 2D correlation experiments of ^{1}H–^{1}H *gs*-COSY, ^{1}H–^{13}C *gs*-HMQC and ^{1}H–^{13}C *gs*-HMBC. The samples for the NMR measurements were prepared by dissolution of the compounds L1 (obtained from commercial sources) and **1**, **2**, **3** and **5** (isolated from the reactions of L1, and L1 with $ZnCl_2 \cdot 1.5H_2O$ in variously acidic media) in DMF-d_7.

NMR data for N6-isopentenyladenine (L1): ^{1}H-NMR (DMF-d_7; TMS; ppm): δ_H 12.96 (1H, bs, N9H), 8.25 (1H, s, C2H), 8.17 (1H, s, C8H), 7.50 (1H, bs, N6H), 5.42 (1H, tt, *J* 6.9 and 1.4, C10H), 4.23 (2H, bs, C9H), 1.76 (3H, s, C12H), 1.71 (3H, s, C13]. ^{13}C-NMR (DMF-d_7, TMS, ppm): δ_C 154.96 (C6), 152.82 (C2), 150.21 (C4), 139.87 (C8), 133.76 (C11), 122.56 (C10), 119.52 (C5), 38.19 (C9), 25.21 (C13), 17.46 (C12).

X-ray crystallography

Single-crystal data were collected with an *Xcalibur2* (Oxford Diffraction) for **1**, **2** and **5**, and with four-circle kappa-axis KUMA KM-4 diffractometer (KUMA Diffraction, Wroclaw) for **3** and **4**, at 120 K using graphite-monochromated MoKa radiation and CCD detector. CrysAlis software package was used for data collection and reduction. The structures were solved by direct methods [SHELXS-97] and refined on F^2 using full-matrix least-squares procedure [SHELXL-97][38] All H-atoms were found from difference Fourier maps and were refined using a riding model, expect for those belonging to the crystal water molecule in **1** and **3**, whose positions were refined freely. Molecular graphics were made and additional structural parameters were interpreted by means of DIAMOND.[39]

Syntheses

[Zn(HL1)Cl$_3$]·H$_2$O (1). To a stirred warm (50 °C) solution of 0.10 g (0.5 mmol) of N6-isopentenyladenine in 35 ml of 0.1 M HCl, 2 mmol of $ZnCl_2 \cdot 1.5H_2O$ was added. The resulting mixture was kept under stirring at 50 °C for 30 min and then was allowed to stay at room temperature. After a week, crystals of **1** suitable for single crystal X-ray analysis formed by slow evaporation of the solvent. Found: C, 30.3; H, 4.0; N, 17.9. Calc. for $C_{10}H_{16}N_5OCl_3Zn$: C, 30.5; H, 4.1; N, 17.8%. Yield: 72%.

NMR data: ^{1}H-NMR (DMF-d_7, TMS, ppm): δ_H 1H, HN^9- not detected, 9.97 (1H, t, *J* 6.7, N6H), 8.84 (1H, s, C8H), 8.67 (1H, s, C2H), 5.43 (1H, t, *J* 6.7, C10H), 4.32 (2H, t, *J* 5.7, C9H), 1.77 (3H, s, C12H), 1.73 (3H, s, C13H). ^{13}C-NMR (DMF-d_7, TMS, ppm): δ_C 151.59 (C6], 147.35 (C4), 147.13 (C2), 143.74 (C8), 136.93 (C11), 119.09 (C10), 112.77 (C5), 42.12 (C9), 25.08 (C13), 17.54 (C12).

Crystal data and structure refinement for complex **1**: $C_{10}H_{16}Cl_3N_5OZn$, *M* = 394.00, monoclinic, *a* = 11.4767(3), *b* = 9.9382(3), *c* = 14.0372(5) Å, *V* = 1599.34(9) Å^3, *T* = 120(2)K, space group P 21/c, *Z* = 4, reflections collected/unique: 9458/2811 (R_{int} = 0.0144), final *R* indices: *R1* = 0.0413, *wR2* = 0.0904, *R* indices (all data): *R1* = 0.0418, *wR2* = 0.0905.

[Zn$_3$(HL2)$_2$Cl$_8$] (2). The preparation of **2** followed the same synthetic procedure as described for **1** employing N6-isopentenyladenine and $ZnCl_2 \cdot 1.5H_2O$ as the starting compounds. However, 1 M HCl was used as the medium and the reaction time was 5 h in this case. Colourless crystals of **2** were obtained by slow evaporation of the reaction solution. Found: C, 26.8; H, 3.2; N, 15.7. Calc. for $C_{20}H_{14}N_{10}Cl_8Zn_3$: C, 27.0; H, 3.2; N, 15.8%. Yield: 60%.

NMR data: ^{1}H-NMR (DMF-d_7, TMS, ppm): δ_H 10.37 (1H, s, N6H), 9.16 (1H, bs, N9H), 8.94 (1H, s, C2H), 8.56 (1H, s, C8H), 3.79 (2H, t, *J* 6.0, C9H), 2.37 (2H, t, *J* 6.0, C10H), 1.88 (6H, s, C12,C13H). ^{13}C-NMR (DMF-d_7, TMS, ppm): δ_C 147.44 (C6), 146.92 (C4), 143.81 (C2), 143.32 (C8), 117.84 (C5), 60.16 (C11), 35.66 (C9), 32.13 (C10), 27.51 (C12,C13).

Crystal data and structure refinement for complex **2**: $C_{20}H_{28}Cl_8N_{10}Zn_3$, *M* = 888.23, triclinic, *a* = 8.3518(3), *b* = 10.0642(5), *c* = 21.5006(10) Å, *V* = 1646.72(13) Å^3, *T* = 120(2)K, space group P$\bar{1}$, *Z* = 2, reflections collected/unique: 11581/5752 (R_{int} = 0.0082), final *R* indices: *R1* = 0.0214, *wR2* = 0.0695, *R* indices (all data): *R1* = 0.0223, *wR2* = 0.0702.

(H$_2$L3)Cl$_2$·2H$_2$O (3). A solution containing N6-isopentenyladenine (0.20 g, 1 mmol) in 2 M HCl (20 ml) was refluxed at 100 °C for 2 days. The reaction solution was left standing at room temperature and crystals of **3** suitable for single crystal X-ray analysis formed after a few days. Found: C, 35.5; H, 7.0; N, 22.9. Calc. for $C_9H_{21}N_5O_2Cl_2$: C, 35.8; H, 7.0; N, 23.2%. Yield: 45%.

NMR data: ^{1}H-NMR (DMF-d_7, TMS, ppm): δ_H N1,N8H – not detected, 10.36 (1H, s, N12H), 10.06 (1H, s, N3H), 9.54 (2H, bs, N6H), 8.91 (1H, s, C2H), 3.61 (2H, m, C11H), 1.93 (2H, t, *J* 6.1, C10H), 1.49 (6H, s, C13,C14). ^{13}C-NMR (DMF-d_7, TMS, ppm): δ_C 148.42 (C7), 143.48 (C4), 131.80 (C2), 99.97 (C5), 51.35 (C9), 36.46 (C11), 31.76 (C10), 27.90 (C13,14).

Crystal data and structure refinement for **3**: $C_9H_{21}Cl_2N_5O_2$, *M* = 302.21, monoclinic, *a* = 7.0591(4), *b* = 15.6552(7), *c* = 13.2731(6) Å, *V* = 1451.69(12) Å^3, *T* = 120(2)K, space group P 21/n, *Z* = 4, reflections collected/unique: 7984/2562 (R_{int} = 0.0354), final *R* indices: *R1* = 0.0261, *wR2* = 0.0674, *R* indices (all data): *R1* = 0.0278, *wR2* = 0.0684.

(H$_2$L3)[CuCl$_4$] (4). N6-isopentenyladenine (0.20 g, 1 mmol) was dissolved in 10 ml of 2 M HCl. Then, a solution (10 ml) of $CuCl_2 \cdot 1.5H_2O$ (0.16 g, 1 mmol) in the same solvent was added. The resulting mixture was refluxed at 100 °C for 24 h. By slow evaporation of the solvent at room temperature, single crystals formed after a period of 5–6 weeks. Found: C, 27.1; H, 4.2; N, 17.4. Calc. for $C_9H_{17}N_5Cl_4Cu$: C, 27.0; H, 4.3; N, 17.5%. Yield: 58%.

Crystal data and structure refinement for **4**: $C_9H_{17}N_5Cl_4Cu$, *M* = 400.62, triclinic, *a* = 7.6673(4), *b* = 7.8804(5), *c* = 13.4349(7) Å, *V* = 794.67(8) Å^3, *T* = 120(2)K, space group P$\bar{1}$, *Z* = 2, reflections collected/unique: 4809/2778 (R_{int} = 0.0448), final *R* indices: *R1* = 0.0352, *wR2* = 0.0944, *R* indices (all data): *R1* = 0.0366, *wR2* = 0.0955.

(H$_2$L3)[ZnCl$_4$] (5). The same synthetic procedure as described above for **4** was applied, only $ZnCl_2 \cdot 1.5H_2O$ was used as the inorganic salt. During the reaction time, the colour of the reaction solution changed from colourless to pale yellow. Pale yellow single

crystals formed by slow evaporation of the solvent. Found: C, 27.0; H, 4.1; N, 17.1. Calc. for $C_{18}H_{34}N_{10}Cl_8Zn_2$: C, 26.9; H, 4.3; N, 17.4%. Yield: 65%.

NMR data: ^{1}H-NMR (DMF-d_7, TMS, ppm): δ_H N1,N2H – not detected, 10.20 (2H, bs, N6H), 9.72 (1H, bs, N8H), 9.60 (1H, bs, NH), 8.62 (1H, s, C2H), 3.64 (2H, m, C9H), 1.96 (2H, t, *J* 5.9, C10H), 1.46 (6H, s, C13,C14). ^{13}C-NMR (DMF-d_7, TMS, ppm): δ_C 149.89 (C7), 143.33 (C4), 132.59 (C2), 100.37 (C5), 51.11 (C11), 36.75 (C9), 31.85 (C10), 27.79 (C13,C14).

Crystal data and structure refinement for **5**: $C_9H_{17}N_5Cl_4Zn$, M = 402.45, triclinic, a = 7.8356(4), b = 13.1475(6), c = 15.8482(9) Å, V = 1568.96(14) Å^3, T = 120(2)K, space group $P\bar{1}$, Z = 4, reflections collected/unique: 9645/5487 (R_{int} = 0.0639), final R indices: $R1$ = 0.0403, $wR2$ = 0.1079, R indices (all data): $R1$ = 0.0432, $wR2$ = 0.1100.

Conclusions

The behaviour of N6-isopentenyladenine (L1) in variously acidic media and in the presence of Zn(II) or Cu(II) has been studied by solution-state NMR spectroscopy and single crystal X-ray analysis. In general, three transformation products have been observed, N1-protonated N6-isopentenyladenine (HL1), the tricyclic cation 7,8,9,10-tetrahydro-7,7-dimethyl-3*H*-pyrimido[2,1-*i*]purin-6-ium (HL2) originating from cyclization of the aliphatic chain to purine, and the 5-amino-4-(4,4-dimethyl-tetrahydropyrimidin-2-yl)-imidazolium dication (H_2L3), which is formed after ring-fission. In most cases, we were unable to obtain single crystals of the organic transformation products, which was one of the aims of this study. We therefore added the metal ions into the system and this study showed that the presence of a metal ion [in our case Zn(II) or Cu(II)] in the reaction mixture had significant influence on the course of the whole reaction process. The reaction speed of acid hydrolysis with the inorganic chlorides present was significantly increased and pure chemical species in the forms of single crystals suitable for X-ray analysis could be obtained. The results following from this approach involve structural description of the organic hydrolysis products and at the same time, elucidation of their interactions with the metal centres. Therefore, apart from the L1 acid hydrolysis product formation, the coordination abilities of the corresponding intermediates to the mentioned metal ions have been investigated. As can be seen from the composition of the $[Zn(HL1)Cl_3]\cdot H_2O$ (**1**), $[Zn_3(HL2)_2Cl_8]$ (**2**), $(H_2L3)[CuCl_4]$ (**4**) and $(H_2L3)[ZnCl_4]$ (**5**) complexes, the HL1 and HL2 cations directly coordinate to the metal ion through the N7 atoms, while H_2L3 does not coordinate and remains in its cationic form compensating the charge of the complex anion $[MCl_4]^{2-}$. This observation might be explained by a strong donor ability of chloride anions and their increasing concentration across the row **1**, **2**, **4** and **5**. Therefore, generally, the proton and Cl^- concentration are most likely indirectly responsible for coordination or non-coordination of the organic hydrolysis products to the metal sites. The proton concentration influences the acid transformation of the organic compounds and the chloride concentration affects coordination to the metal or the formation of the complex anion $[MCl_4]^{2-}$.

The first step of the ring opening of L1 follows the Dimroth rearrangement. As was previously reported,[13,18] in the non-substituted purine skeleton of adenine, the amidine group N7–C8–N9 is the hydrolysis target. However, if either N1 or N3 is alkylated {the case of 7,8,9,10-tetrahydro-7,7-dimethyl-3*H*-pyrimido[2,1-*i*]purin-6-ium cation}, the electrophilic N1–C2–N3 is the preferred amidine-group for the hydrolysis and the opening of the pyrimidine ring is therefore rendered possible. The composition of the presented compounds **1–5** in solution and solid state was elucidated by heteronuclear NMR spectroscopy, and X-ray analysis, respectively. The molecular and crystal structures of the acidic hydrolysis transformation products of an adenine derivative in general are herein reported for the first time.

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