[a030]

1-(2',3',4',6'-TETRA-O-ACETYL-β-D-GLUCOPYRANOSYL)-3-(4",6"-DIARYLPYRIMIDINE-2"-YL)-THIOUREAS

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Abstract

Some compounds of 1-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)-3-(4",6"-diarylpyrimidine-2"-yl)thioureas have been synthesized from corresponding isothiocyanates and 2-amino-4,6-diarylpyrimidines using two different synthetic methods. The hydrolysis of these acetated thioureas lead to form the corresponding deacetyled thioureas. Their spectroscopic properties have been studied.

INTRODUCTION

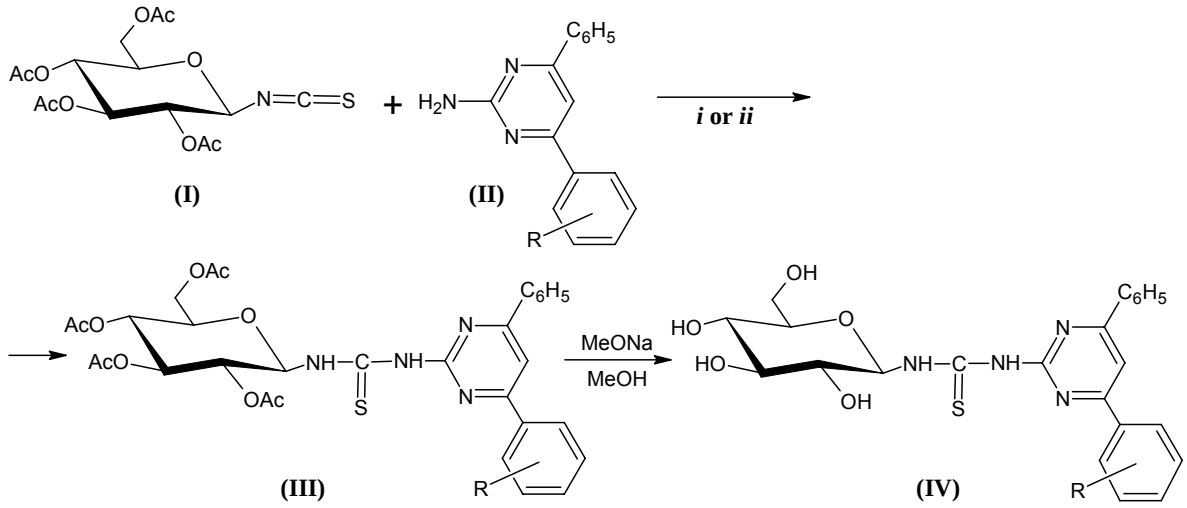
Carbohydrates can serve as structural components of natural products, energy sources or key elements in various biomolecular recognition phenomena. Carbohydrate-mediated signalling is especially important during bacterial and viral infections, cell-cell adhesion in inflammation and metastases implantation, tissue differentiation, development and the regulation of many other inter- and intracellular communication and signal transduction events [1]. The molecules involved are characterized by a wide complexity, which contributes to their diversity and biological activity. In other hand, sugar isothiocyanates are among the most versatile synthetic intermediates in carbohydrate chemistry. They play a pivotal role in the preparation of a broad series of functional groups such as amide, isonitrile, carbodiimide, and N-thiocarbonyl derivatives allowing, simultaneously, the covalent coupling of a quite unrestricted variety of structures to the saccharide part [2,3]. Moreover, isothiocyanates are important reagents in heterocyclic chemistry, which may be exploited in the synthesis of nucleosides and other N-glycosyl structures [4,5].

In the present study, we report the synthesis of various glucosyl thioureas containing pyrimidine nucleus, together with some of their spectroscopic properties.

RESULTS AND DISCUSSION

1. Synthesis of acetated glucosyl thioureas

The derivatives of 1-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)-3-(4",6"-diarylpyrimidine-2"-yl)thioureas (III) could be easily synthesized by the addition of corresponding amino compounds (II) on isothiocyanate derivatives (I). We performed this reaction by using two methods, by refluxing in dried toluene in about 10 hrs [6] or by executing in microwave oven in several minutes [7]. Then obtained acetated glucopyranosyl thioureas have been undergone hydrolysis in the present of natrium methylate into corresponding β-D-glucopyranosyl-3-(4',6'-diarylpyrimidine-2'-yl)thioureas (IV). The synthetic processes could be represented in Figure 1.



where, II, III and IV: a R=H, b R=*p*-Cl, c R=*m*-Cl, d R=*p*-Br, d R=*p*-OMe. (**i**) Method A, by refluxing in dried toluene in 10 hrs.; (**ii**) Method B, by using microwave oven, 2-3 minutes.

Figure 1. Reaction transformations of tetra-O-acetyl-β-D-glucopyranosyl isothiocyanate.

We have found that nucleophile addition of 2-amino-4,6-diarylpyrimidine [8] to 2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl isothiocyanate has taken place fairly easily. Reaction yields were high in both the methods (refluxing and using MW oven), in 60-68% and 72-77%, respectively. All these obtained thioureas could be dissolved in common organic solvents (such as ethanol, methanol, toluene, benzene, DMF,…) and couldn't be dissolved in water. Their structure have been affirmed by spectroscopic data (such as IR-, NMR- and mass-spectra).

In the IR-spectra of these above glucopyranosylthioureas, stretching band of C=S bond in thiourea linkage have appeared in region of 1362-1364 cm^{-1}, furthermore, N–H bonds in thioureas have absorption band in region of 3622-3410 cm^{-1}, specified for stretching vibration of those bonds. These bands, maybe, have been superimposed each other, hence one absorption band was sometimes appeared in their IR-spectra. These bands were specified for N,N'-substituted thioureas [2,3]. The characteristic of tetraacetated glucopyranose ring was the present of absorption band in region of 1754-1748 cm^{-1} for stretching vibration of C=O bond in ester function (acetyl group in these cases).

In the ^{1}H-NMR spectra of these thioureas there are the resonance signals which are specifed for protons in thiourea-NH groups at δ=11.157-12.036 ppm. Some resonance signals are in region δ=7.625-8.350 ppm belong to some aromatic protons in amino component. Protons C–H in pyranose ring of monosaccharide have some resonance peaks with chemical shifts from 6.212 ppm to 4.208 ppm as observed in ^{1}H-NMR spectra of monosaccharide compounds [9]. Proton H_1 has chemical shift in region δ=6.188-6.212 ppm (in triplet) with couple constant J_{12}=9.0-9.5 Hz. Resonance signal of proton H_2 appears in triplet in region δ=5.020-5.064 ppm with J_{21}=9.0-9.5 Hz. The values of couple constant are correlative with *trans*-H–H couple interaction and indicate β-anomer configuration of NH-thiourea group [9]. Another protons such as H_3, H_4 have triplet resonance signals in regions δ=5.516-5.531 ppm (with couple constants $J_{3,4}$=9.5 Hz) and δ=5.020-5.036 ppm (with couple constants $J_{4,3}$=$J_{4,5}$=9.5 Hz), respectively. The ^{1}H-NMR of thiourea IIIe (R=*p*-OMe) is represented in Figure 1. In the COSY it's shown that proton H_1 have interacted with proton H_2 and proton of NH bond in thiourea group, and shown that these signals have been

degenerated into triplet.

Table 1. Some derivatives of 1-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)-3-(4'',6''-diarylpyrimidine-2''-yl)thioureas

N°	R	Melting Point (°C)		Yield (%)		IR spectra (cm^{-1})					
						A			B		
		A	B	A	B	ν_{N-H}	$\nu_{C=Oest}$ ν_{C-O-C}	$\nu_{C=S}$	ν_{N-H}	$\nu_{C=Oest}$ ν_{C-O-C}	$\nu_{C=S}$
IIIa	H	229-230	229-230	60	75	3622; 3531	1754; 1231	1362	3622; 3529	1754; 1231	1362
IIIb	p-Cl	218-219	218-219	68	76	3410	1750; 1222	1364	3410	1750; 1222	1364
IIIc	m-Cl	190-191	190-191	67	72	3410	1750; 1223	1364	3410	1750; 1223	1364
IIId	p-Br	223-224	223-224	66	76	3410	1748; 1223	1363	3410	1748; 1223	1363
IIIe	p-OMe	213-214	213-214	68	77	3434	1750; 1223	1364	3434	1750; 1223	1364

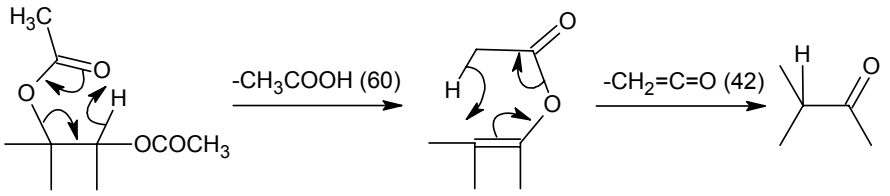
(A): by refluxing; (B): by using microwave oven.

In the ^{13}C-NMR spectra, it's could be noticed that number of carbon atoms in spectra and this one in molecular formulas of each thiourea were identical each other. It's could can be parted the spectra of these thioureas into four regions as follows: 169.297-169.930 ppm, 157.549-106.736 ppm, 81.730-61.718 ppm and 20.479-20.174 ppm. The magnetic resonance signals of the carbonyl bonds C=O in acetyl groups have appeared in the low-field region of δ=169.297-169.930 ppm. In addition, there were some resonance peaks in high-field region of 20.479-20.174 ppm that's indicated the present of methyl groups on acetyl functions. In HMBC spectra, these peaks had some long-range and short-range C–H interactions with the protons in methyl groups. This aspect was agreed with common resonance position of acetyl group. The aromatic and hetero-aromatic carbon atoms had chemical shifts in region of 157.549-106.736 ppm. Six carbon atoms in pyranose ring had clearly resonance signals in region of 81.730-61.718 ppm and these peaks also had several C–H interaction types with proton on these carbon atoms or adjacent ones.

In NMR spectra using HMBC and HSQC experiments the long-range and the short-range C–H interactions could be determined. For example, carbon atom C_1 had long-range interaction with proton H_2 and proton H_b; carbon atom C_2 interacted with protons H_1 and H_3, ect... Other long-range interactions in thiourea molecules could shown their 2D-NMR HMBC spectra.

The mass spectra of tetra-O-acetyl-β-D-glucopyranosylthioureas containing pyrimidine ring had some features that are similar with ones of typical hexopyranose pentaacetates. As expected of such a highly substituted molecule, the molecular ion $M^{+\bullet}$ was of very low intensity and could be hardly detected on the spectra. The only observable peaks in the higher mass range were due to loss of the substituents and fall, therefore, at position of fragment ions such as $[M\text{-AcOH}]^+$, $[M\text{-AcOH,-AcO}]^+$ and $[M\text{-2AcOH,-AcO}]^+$, and some peaks such as $[M\text{-2AcOH,-}C_2H_2O]^+$, $[M\text{-3AcOH,-AcO}]^+$ with intensity of 1-3% could be specified for glycosides having amino structure. A very important mode of fragmentation of these acetated compounds was the loss of acetic acid (*m/z* 60), a process well known for most esters of acetic acid, and the loss of ketene (*m/z* 42). This later process seemed to be greatly facilitated if preceded by loss of acetic acid; the resulting double bond seemed to play a significant role in the elimination of ketene, for which it could be suggest the

mechanics as following [10,11]:



This process was rather than a simple 1,2-elimination, producing a hydroxyl group. The elimination of 102 mass units in a given step was much more pronounced in compounds containing two acetoxy groups in a 1,2- or possibly a 1,3-relationship, but not if the acetoxy groups were farther apart. In addition to these small peaks at the high end of the spectra, there could be recognized mainly four series of fragments within which the individual peaks differed by 60 and 42 mass units.

The first series (series A, *see Fig.* 2) began with the peak F_1 at *m/z* 331, which formed from the molecular ion by loss of a Het-NHC(=S)NH-group (where, Het=4,6-diarylpyrimidine2-yl). The resulting carbonium ion would be resonance-stabilized by the free electron pairs of the ether oxygen. Further loss of two molecules of acetic acid gave rise to the admittedly very small peak at *m/z* 211, a "pyronium" ion, and the next step (elimination of ketene) lead to fragment $[F_4–2AcOH–CH_2=C=O]^+$. The process leading to this ion could be somewhat more complex. Further elimination of a molecule of acetic acid from C6 could lead to the fragment ion of *m/z* 109 (*see Fig.3*). In the spectra of these compounds, a second group of peaks began at *m/z* 242 (or *m/z* 241) and was followed by peaks at *m/z* 200, 140, and 98. This fragment must contain C5 including its substituents. It seemed to be formed by elimination of C1 plus the ether oxygen in simultaneous elimination of one molecule of acetic acid (*see Fig. 4*). It's seemed to be formed by elimination of C1 plus the ether oxygen in simultaneous elimination of one molecule of acetic acid, most probably by the path outlined in Figure 4 [11,12].

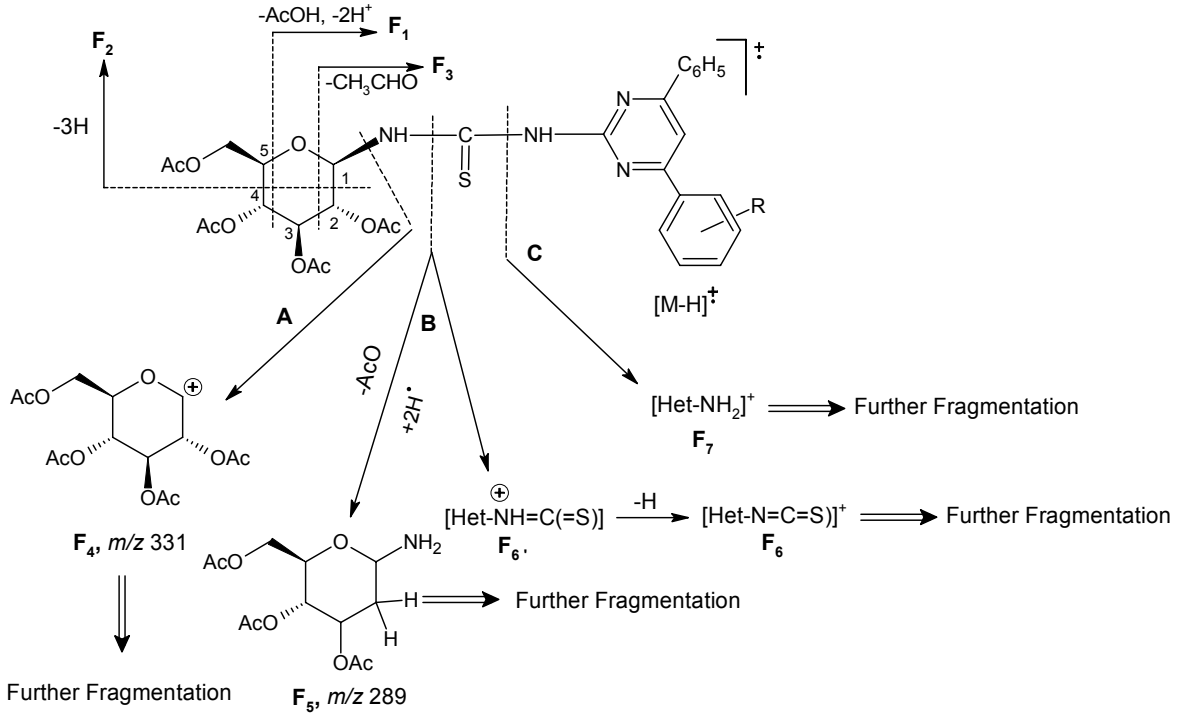


Figure 2. Some general fragmentations in MS spectra of some 1-(2',3',4',6'-tetra-O-acetyl-β-D-glucopyranosyl)-3-(4",6"-diarylpyrimidine-2"-yl)thioureas.

It could be indicated that the fragmentation of peracetate derivatives of glucopyranosylthioureas can be divided some tendencies as follows [3-5]:

1. Fragmentation enclosing cleavage of bond between pyrimidine ring and thiourea group.

2. Cleavage of substituents and cleavage of pyranose ring starting directly from $M^{+\bullet}$.

3. Cleavage of NH–Glc and formation of fragment ion F_4 (*m/z* 331) which is disintegrated in the next steps specifying for the corresponsive acetated monosaccharides.

In the mass spectra of most studied compounds, fragment ion F_5 (*m/z* 288 or *m/z* 230) appeared. This ion maintained sugar chain and a part of thiourea bond. There were ion peaks $[M-219]^+$ (F_1 and F_2) with varying intensities and its element composition would allow to expect about two fragmentations of pyranose ring as indicating in Fig. 2.

Peak F_3 had different intensity in mass spectra, this ion was formed by cleaving C1–O and C2–C_3 bonds in pyranose ring. In addition, the cleavage took place through C1–C2 and C5–O bonds leading to form Het-NHC(=S)NHC≡O^+ ions specifying for some N-glycosides.

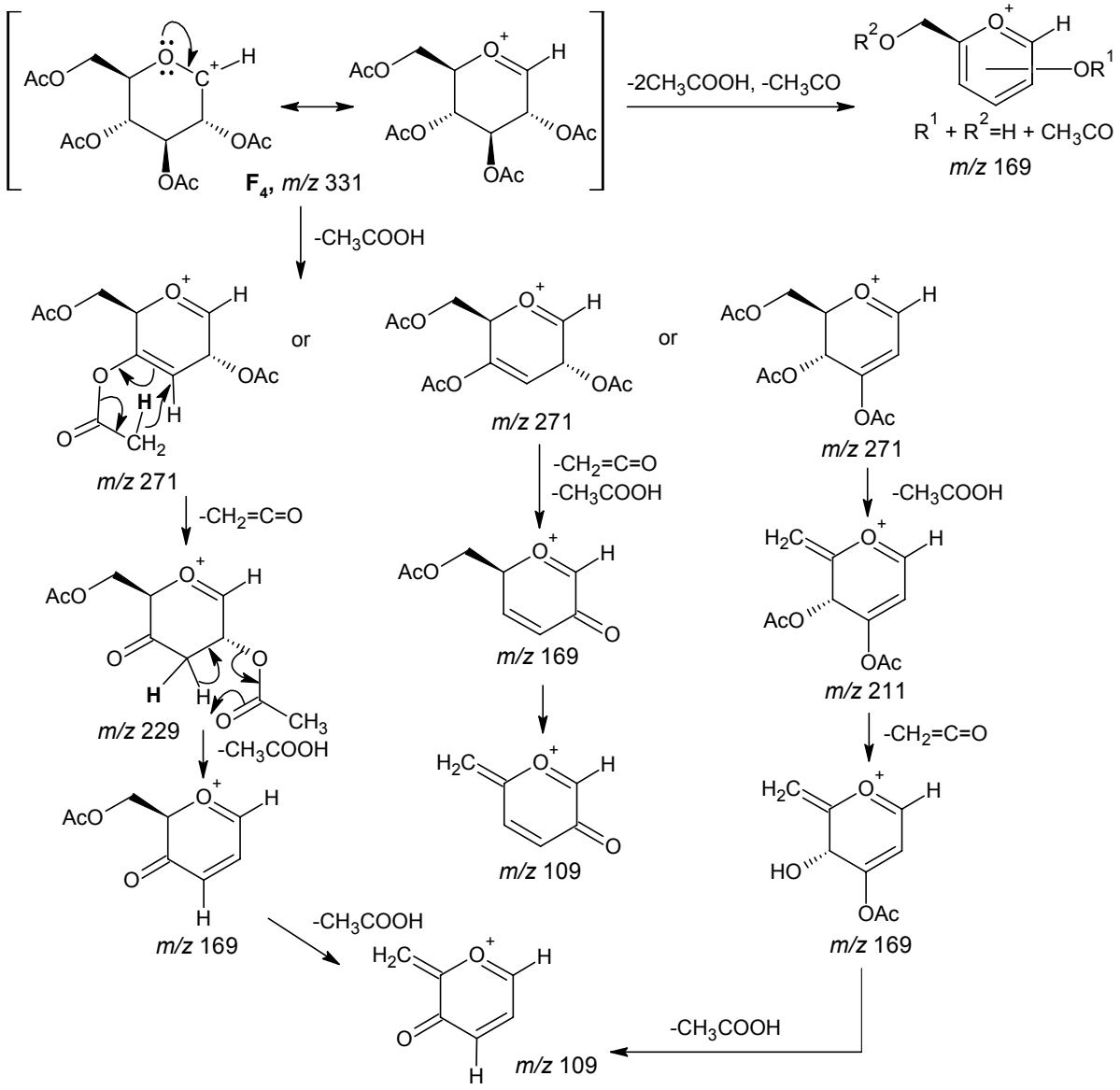


Figure 3. Fragmentation of fragment ion of *m/z* 331 (F_4).

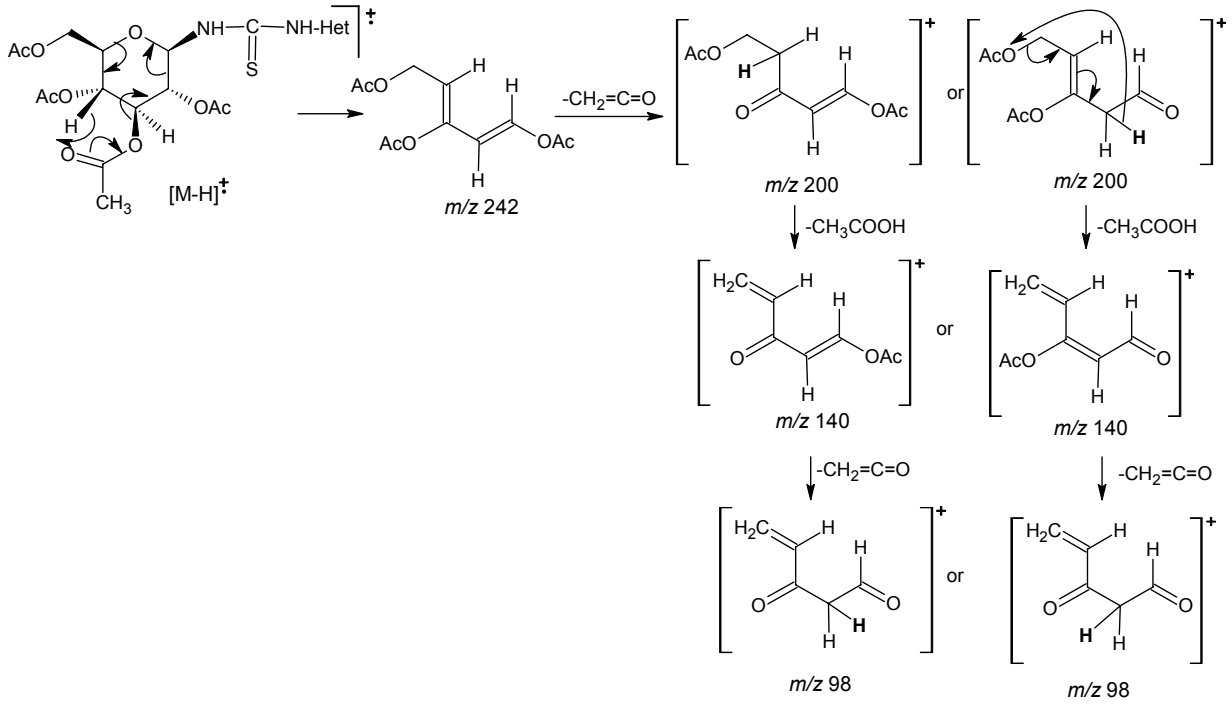


Figure 4. Formation and fragmentation of fragment ion of *m/z* 242/241.

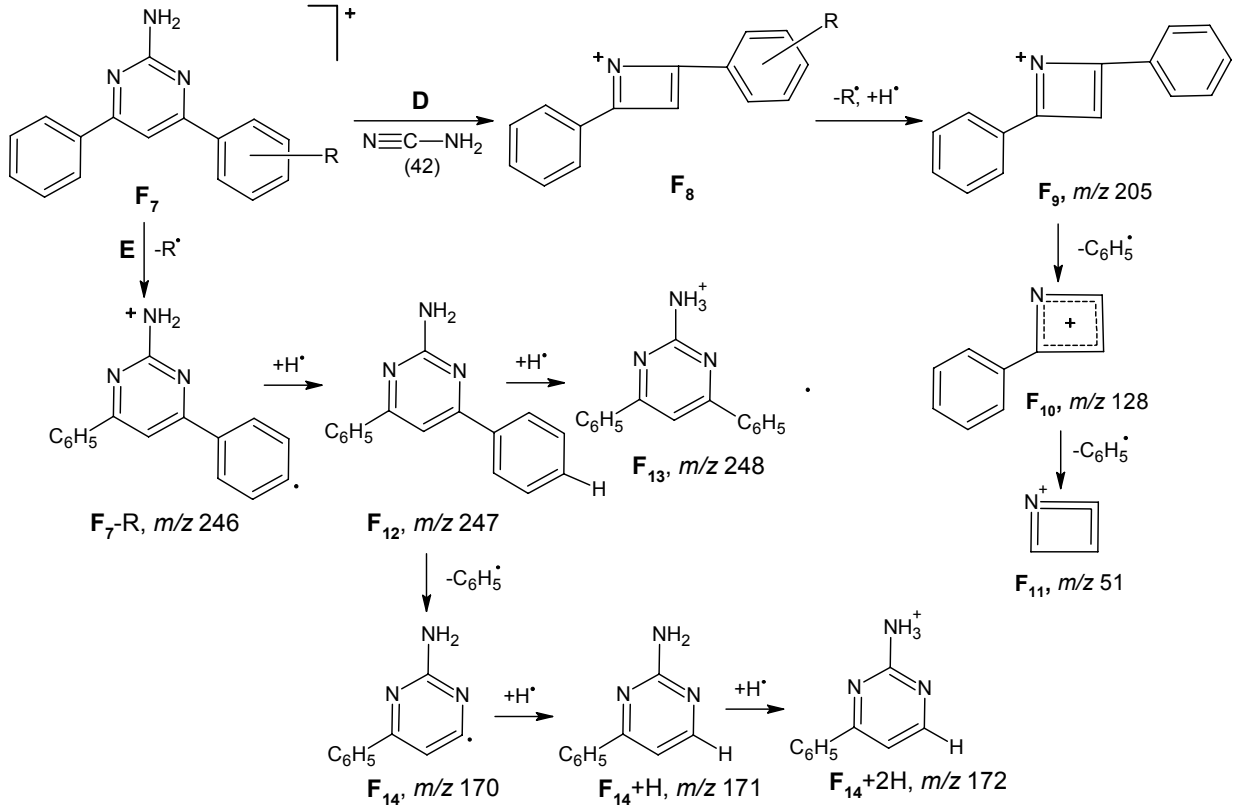


Figure 5. Fragmentation of fragment ion $[Het\text{-}NH_2]^+$ (F_7).

The third fragmentation (**C**) took place with cleavage of thiourea bond accompanying with formation of fragment ion F_7 $[Het–NH_2]^+$ which mass equals molecular weight of corresponding amines. Its intensity was usually the most high (from 90-99%) because the aromatic and heteroaromatic nuclei contributed to stabilize this fragment ion. The next fragmentation carried out in two directions, as follows [10-12] *(see Fig. 5, 6)*:

1. Cleavage of pyrimidine accompanying with elimination of $N{\equiv}C{-}NH_2$ radical and formation of azete ring.

2. Cleavage of phenyl out of pyrimidine ring.

EXPRIMENTAL PART

Melting point of the synthesized compound was measured by using Thiele's apparatus in capillary and uncorrected. The FTIR-spectra were recorded on Magna 760 FT-IR Spectrometer (NICOLET, USA) in form of mixing with KBr and using reflex-measure method. The ^{1}H-NMR was recorded on an AVANCE Spectrometer (BRUKER, German) at 500 MHz, using DMSO-d_6 as solvent and TMS as an internal reference. The high-resolution mass (HR-MS) spectra were recorded in instrument AutoSpec Premier (WATERS, USA). 2,3,4,6-Tetra-O-acetyl-β-D-glucopiranosyl isothiocyanate was synthesized by known method [2,3].

*S*ynthesis of 1-(2',3',4',6'-tetra-O-acetyl-(β-D-glucopiranosyl)-3-(4",6"-diarylpyrimidine-2"-yl)-thioureas

General Procedure 1 (Refluxing Method):

In a 50-ml. round-bottomed flask were placed 0.494 g (0.002 moles) of 2-amino-4,6-diphenylpyrimidine and 0.778 g (0.002 moles) of 2,3,4,6-tetra-O-acetyl-β-D-glucopiranosyl isothiocyanate. Added 20 ml. absolute dioxane in the mixture. Then the mixture was heated in refluxing about 10 hrs. Solvent was removed under reduced pressure to obtained ivory-white or white products. Recrystallized from a mixture of ethanol and toluene (1:1 in volume) obtained ivory-white crystals.

General Procedure 2 (using Microwave Oven):

Mixed 0.494 g (0.002 moles) of 2-amino-4,6-diphenylpyrimidine and 0.778 g (0.002 moles) of 2,3,4,6-tetra-O-acetyl-β-D-glucopiranosyl isothiocyanate. Then this mixture was irradiated about 2-3 min. at 750 Watts. The mixture had become dark-yellow. Cooled it to room temperature, recrystallized from a mixture of ethanol and toluene (1:1 in volume) obtained ivory-white crystals.

Results of these above syntheses were represented in Table 1.

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References

1. (a) Anu Agarwal et al, *Bioorg. Med. Chem. Lett*, (15) (2005), pp. 5218-5221. (b) Anu Agarwal, Ramesh, Ashutosh, Neena Goyal, Prem M. S. Chauhan, Suman Gupta, *Bioorg. Med.Chem,*(13) (2005), pp.6678-6684.
2. Z. J. Witczak, *Adv. Carbohydr. Chem. Biochem.*, **44**, (1986), pp. 91-145.
3. (a) J. M. García-Fernández, C. Ortiz-Mellet, *Sulfur Rep.*, **19** (1996), pp. 61-169. (b) J. M. García Fernández, C. Ortiz Mellet, *Adv. Carbohydr. Chem. Biochem*., **55** (1999), pp. 36-135.
4. I. Goodman, *Adv. Carbohydr. Chem.*, **13** (1958), pp. 215-236.
5. (a) Naito, T.; Sano, M., *Chem. Pharm. Bull.*, 9 (1961), pp. 709-714. (b) Ukita, T.; Hamada, A.; Yoshida, M., *Chem. Pharm. Bull.,* 12, 2(4) (1964), pp. 459-465. (c) Ogura,

H.; Takahashi, H., *Heterocycles* 8, (1977), pp. 125-146.
6. (a) Camarasa M. J., P. Fernandez-Resa, M. T. Garcia-Lopez, F. G. de las Heras, P. P. Mendez-Castrillon, and A. San Felix, *Synthesis,* (1984), pp. 509-510. (b) Carla Prata, Nathalie Mora, Jean-Michel Lacombe, Jean-Claude Maurizis, Bernard Pucci, *Carbohydrate Research.,* **321** (1999), pp. 4–14.
7. Jia Qiang DONG, Shu Jia ZHANG, Yan Guang WANG, *Chinese Chemical Letters*, **Vol. 14**, No. 9 (2003), pp. 904 – 906.
8. Nguyen Dinh Thanh, Nguyen Thi Thanh Mai, Dang Nhu Tai, Pham Hong Lan, *Proceedings of the Scientific Conference on the occasion of the 50th Aniversary of Faculty of Chemistry,* Hanoi University 1956-2006, Hanoi 10/2006, pp. 104-108.
9. E. Pretsch, P. Buhlmann, C. Affolter, *Structure Determination of Organic Compounds*, Springer-Verlag, Berlin, 2000.
10. Я.В. Рашкес, Ю.М. Мильгром, Р.Ф. Амбарцумова, *Масс-спектрометрическое исследование тиокарбамоилзамещенных 2-аминотиазолов и 2-иминотиазолинов. 2. N-Тиазолил- и N-тиазолинилиден-N'-перацетилгликозилтиомочевины*, Хим. Гетероцикл. Соед., N. 11 (1989), стр. 1539-1543.
11. K. Biemann, D.C. DeJongh and H.K. Schnoes, *Application of Mass Spectrometry to Structure Problems. XIII. Acetates of Pentoses and Hexoses*, J. Am. Chem. Soc., vol. 85 (1963), pp. 1763- 1771.
12. E. Haslam, *The Mass Spectra of Some Aryl Glucosides*, Carbohydr. Res., 5 (1967), 161-165.