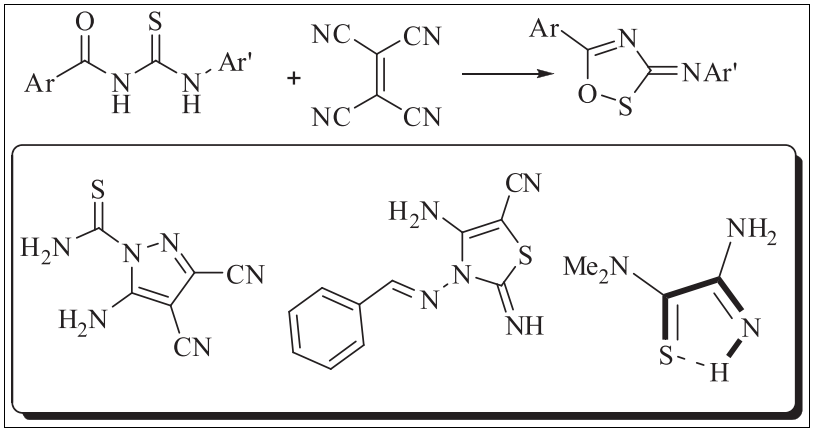
Ashraf A Aly,[a]* Tamer El Malah,[b] Esam A. Ishak,[c] Alan B. Brown,[d] and Wael M. Elayat[e]

[a]Chemistry Department, Faculty of Science, Minia University, 61519 El-Minia, Egypt
[b]National Research Centre, El Buhouth St., Dokki, Cairo, Egypt
[c]Chemistry Department, Faculty of Science, Al-Azhar University, Assiut, Egypt
[d]Chemistry Department, Florida Institute of Technology, Melbourne, FL 32901, USA
[e]Chemistry Department, Faculty of Science, Aljouf University, Sakaka, Aljouf, Saudi Arabia
*E-mail: ashrafaly63@yahoo.com
Received December 15, 2013
DOI 10.1002/jhet.2236
Published online 7 March 2016 in Wiley Online Library (wileyonlinelibrary.com).



Reaction of 1,1,2,2-ethenetetracarbonitrile (TCNE) with aroyl thioureas in dioxane catalyzed by few drops of piperidine, led to the corresponding 1,2,4-oxathiazoles. Under the same reaction condition, thiosemicarbazide or thiosemicarbazone reacted with either TCNE or 1-amino-1,2,2-ethenetricarbonitrile. The structures of the products were elucidated via NMR, IR, mass spectra, and elemental analyses. The mechanism of formation was discussed. Biological (against Gram-positive and Gram-negative bacteria) and antioxidant activities were tested. Some of these heterocyclic compounds showed high activity as antioxidant and antibacterial compounds. The structure–activity relationship was investigated.

J. Heterocyclic Chem., **53**, 963 (2016).

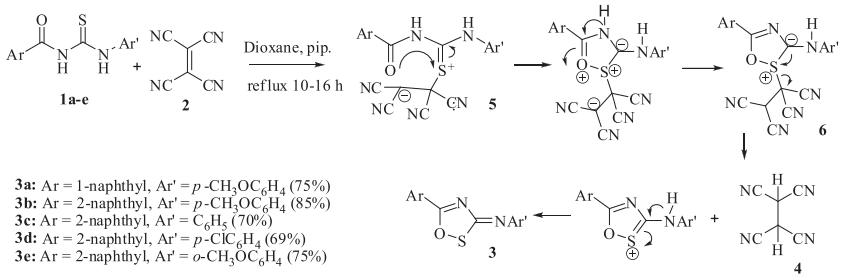
INTRODUCTION

Thiourea moieties are important chemical building blocks that have numerous chemical and pharmaceutical applications. For example, recent reports describe thiourea derivatives as efficient guanylating agents both in solution and on solid support [1]. Hyrazinothioureas are another class of thioureas such as 1-acylthiosemicarbazides, which reacted with phenyl propiolate in acetic acid under reflux to afford triazolothiazines [2]. Aly *et al.* [3] demonstrated that the reaction between *N*-aroyl-*N′*-aryl-thioureas and 2-(1,3-dioxoindan-2-ylidene)-malononitrile furnished indeno[1,2-*d*]-[1,3]thiazepines, some of which showed antitumor and antioxidant activities. One indenothiazepine derivative showed a high inhibition of the cell growth of Hep-G2 cells compared with the growth of untreated control cells, as concluded from their low IC_{50} value of 21.73 μM [3]. It has been reported that 1,3-thiazines were obtained from the reaction of *N*-aroyl-*N′*-substituted thioureas with ethyl propiolate, dimethyl but-2-ynedioate, and (*E*)-1,4-diphenyl-but-2-ene-1,4-dione [4]. Additionally, diethyl maleate reacted with *N*-substituted-hydrazino-carbothioamides to form ethyl [1,2,4]triazolo[3,4-*b*][1,3]thiazine-5-carboxylates [5]. Thus, it appears that the reaction pathways of substituted thioureas vary from one reagent to another. Herein, we report the synthesis of various novel heterocycles during the reaction of various *N*-naphthoyl-*N′*-aryl-thioureas (**1a–e**) with 1,1,2,2-ethenetetracarbonitrile (TCNE, **2**). The naphthalene moiety was chosen because it is reported to exhibit antifungal, antibacterial, and antitumor activities [6,7]. We also report on one-pot reactions of other hydrazino-carbothioamides with π-acceptors such as TCNE and 1-aminoethenetricarbonitrile (AETC, **13**). The antioxidant and antibacterial biological activities of the obtained products were investigated, and structure–activity relationships (SAR) were discussed.

RESULTS AND DISCUSSION

Chemistry. On adding one equivalent of TCNE (**2**) to a solution of **1a–e** in dioxane containing a few drops of piperidine, the reaction proceeded under reflux for 10–16 h to give the corresponding products **3a–e** and 1,1,2,2-tetracyanoethane (**4**, 20%; Scheme 1). The

Scheme 1. Proposed mechanism of formation of oxathiazoles **3a–e**.

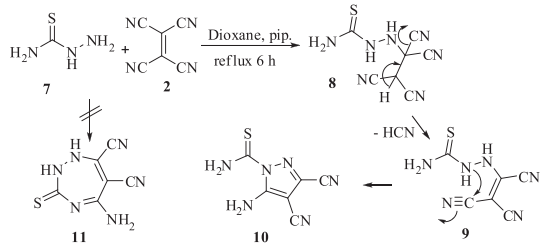


structures of **3a–e** were elucidated by ^{1}H NMR, ^{13}C NMR, IR in addition to elemental analyses. The IR and ^{1}H NMR spectral data of **3a** proved the absence of any NH groups or any protons in heterocyclic ring. Additionally, the IR and ^{13}C NMR spectral data indicated the absence of C═S and C═O groups. The two C═N carbon signals appeared in the ^{13}C NMR at $\delta = 158.6$ and 156.5, and the carbon signal of Ar–*C*–OCH_3 appeared at $\delta = 152.5$. These observations indicate that TCNE (**2**) behaved as an oxidizing agent. The mechanism is proposed to involve conjugate addition of the sulfur atom of compound **1** to the strong electrophilic C═C bond of **2**, followed by cyclization and elimination of 1,1,2,2-tetracyanoethane (**4**, Scheme 1). Tetracyanoethane (**4**), proposed as a by-product, was in fact isolated.

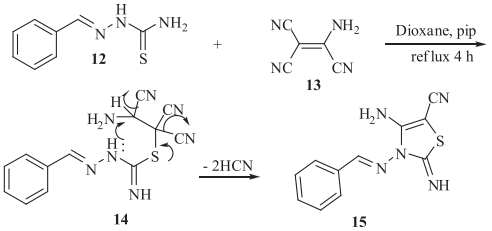
To show the reactivity of the thioamide group thiosemicarbazide (**7**) (hydrazine-carbothioamide), we reacted compound **7** with TCNE (**2**) under the same reaction conditions mentioned earlier. The reaction gave the corresponding pyrazole **10** *via* tricyanovinylation product of **9** (82%; Scheme 2). The mechanism is proposed to involve nucleophilic attack of the hydrazine-NH on the electrophilic C═C of **7**. Elimination of a molecule of HCN from **8** followed by cyclization would give the pyrazole **10** (Scheme 2). Cyclization via attack of the thioamido-NH_2 on a nitrile carbon, leading to the triazepine **11**, was excluded on the bases of proton integration with the relation (1:1). The four protons in compound **10** appeared as two broad singlets at $\delta = 10.4$ and 6.6, assigned to the thiourea-NH_2 and the other free amino protons.

Surprisingly, reaction of 2-(1-phenyl-ethylidene) hydrazinecarbothio-amide (**12**) [8] with AETC (**13**) afforded the corresponding 2-iminothiazole **15** (Scheme 3). The structure of **15** is supported by NMR, IR, and mass spectra in addition to elemental analysis. The ^{1}H NMR spectrum revealed a singlet at $\delta = 10.50$, assigned to the imino-NH; the other NH protons resonated in the region of phenyl protons. In the ^{13}C NMR spectrum, C-2 resonated at $\delta = 156.0$. No C═S carbon signal appeared, but two carbon signals at $\delta = 70.0$ and 178.0 were assigned to C-4 and C-5, respectively. The mechanism was based upon nucleophilic addition of the lone pair of thione-*S* in

Scheme 2. Proposed mechanism for formation of pyrazole **10**.



Scheme 3. Reaction of thiosemicarbazone **12** with acceptor **13**.

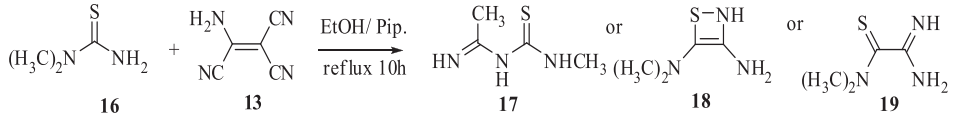


12 to the C-2 in **13**, followed by cyclization and the elimination of two HCN molecules to produce **15**.

Under similar condition, the reaction of equimolar quantities of *N*,*N*-dimethylthiourea (**16**) and **13** gave one product assigned as 2-amino-2-imino-*N*,*N*-dimethylethane-thioamide (**19**, Fig. 1). Mass spectra give a molecular weight of $m/z = 131$, consistent with the formula $C_4H_9N_3S$.

The formula requires two degrees of unsaturation (rings or π bonds). The ^{13}C NMR spectrum (Table 1) shows two sp^2 carbons, the chemical shifts of which is required belong to two C═Z bonds (Z═heteroatom) as in **17** or **19**, rather than to one C═C bond plus a ring as in **18**. There is no vinylic H. The two methyl carbons give HMBC correlation with each other's attached protons, requiring that these protons and carbons be near each other, most likely within three bonds; and both methyl proton signals give HMBC correlation with the same nitrogen, which does not bear a proton.

The simplest explanation is that the dimethylamino group remains intact and the methyl groups being non-equivalent



16 13 17 18 19

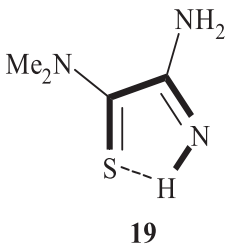
Figure 1. Various proposed structure of product of the formula $C_4H_9N_3S$.

Table 1

NMR spectral data of product **19**.

^{1}H NMR (DMSO-d_6)	H-H COSY	H-N HSQC	H–N HMBC	Assignment
9.91 (b; 1H)	8.00	64		=NH
8.00 (b; 1H)	9.91, 7.52	108		$–NH_2$
7.52 (b; 1H)	8.00	108		$–NH_2$
1.92 (s; 3H)			301	$–NMe_2$
1.91 (s; 3H)			301	$–NMe_2$
^{13}C NMR (DMSO-d_6)	H–C HSQC	H–C HMBC		Assignment
178.36		9.91		C=S
151.63		9.91, 1.92, 1.91		C=N
25.01	1.92	1.91		NMe_2
17.54	1.91	1.92		NMe_2

because of restricted rotation. Of the three N–H protons, two give HSQC correlation with the same nitrogen and presumably are attached to it, again being non-equivalent because of restricted rotation; the third is on a different nitrogen. So, the structure consists of the following parts: $–NMe_2$, $–NH_2$, –NH–, two =C, and –S–. If the sp^2 carbons are not doubly bonded to each other, their double bonds must be to the other divalent groups, that is, there must be C=S and C=NH double bonds, and the two sp^2 carbons must be singly bonded together as in structure **19**. Here, the NH undergoing slower exchange is assigned as being involved in the internal hydrogen bond (Fig. 2); and ChemDraw predicts $\delta = 186$ for C=S and 163.0 for C=N. Compound **19** is not known, but several precedents support the observed characteristics. Both thioamides and amidines are known to exhibit restricted rotation about C–N bonds [9]. The mass spectrum contains a fragment at $m/z = 88$, resulting from loss of H_2N–C=NH, confirming the regiochemistry; this fragment would be unlikely if the NH_2 and NMe_2 groups were interchanged. Therefore, the product was identified as **19**. Mechanistically, compound **13** is described as an oxidizing agent, hence, it enhances dimerization and oxidation of **16** to form adduct **20** (Scheme 4). Rearrangement of intermediate **20** accompanied by transformation of amino group to the adjacent carbon followed by elimination of $[(CH_3)_2N^+H]S^-$ would occur to give **19**.



19

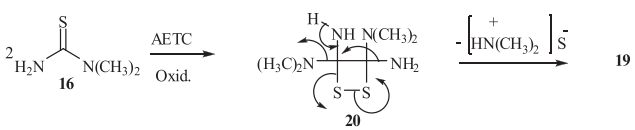
Figure 2. Proposed hydrogen bond in compound **19**.

Antioxidant and antibacterial results. ***Antioxidant activity and relation between structure and the antioxidant activity.*** The thione group in the starting materials acts as an active donating group (electron reducing agent), so that low antioxidant activity was noted of the products compared with *L*-ascorbic acid. In compounds **10** and **15**, the amino and the thione moieties increase the electron-donating property and so increase the antioxidant activity (Table 2). The same trend was observed for addition of serum mixture, although absorbance of compounds in serum (Table 3) is higher than in CUPRAC solution.

Antimicrobial activity and the SAR. Because of the broad spectrum of Ampiclox, we used it as an antibiotic reference in both cases of Gram-positive chosen bacteria, Gram-positive bacteria (*Staphylococcus aureus*), and Gram-negative bacteria (*Escherichia coli*.) [10].

Structure–activity relationship describes the relationship between the chemical or 3D structure of a molecule and its biological activity. The analysis of SAR enables the determination of the chemical groups responsible for evoking a target biological effect in the organism. This allows

Scheme 4. Rationale for the formation of **19**.



16 20 19

Table 2

Relation between concentration (mg/dl) and absorbance of compounds **3a–d**, **10**, **15** and *L*-ascorbic acid in CUPRAC solution.

Conc. (mg/dl)	Abs.	Ascorbic acid	3a	3b	3c	3d	10	15
100		0.4	0.41	0.62	0.49	0.46	0.65	0.79
10		0.3	0.14	0.29	0.31	0.31	0.26	0.48
1		0.06	0.091	0.15	0.21	0.24	0.19	0.41
0.1		0.03	0.054	0.06	0.06	0.09	0.08	0.09

Table 3

Relation between concentration (mg/dl) and absorbance of compounds **3a–d**, **11**, **15** and *L*-ascorbic acid in serum solution.

Conc. (mg/dl)	Abs.	Ascorbic acid	3a	3b	3c	3d	10	15
100		0.73	0.48	0.62	0.67	0.92	1.02	1.01
10		0.51	0.33	0.29	0.43	0.67	0.76	0.89
1		0.21	0.27	0.15	0.29	0.45	0.48	0.37

modification of the effect or the potency of a bioactive compound (typically a drug) by changing its chemical structure.

The type of the substitution on the naphthyl ring did not affect the antimicrobial activity. However, the substitution on the benzene ring is important. The presence of an electron-withdrawing group such as a chlorine atom at the *para* position of the benzene ring (Ar′) in **3d** increased the antimicrobial activity. However, electron-donating groups such as methoxy led to higher activity when in the *para* position of Ar′ (e.g., **3b**) than when in the *ortho* position (**3e**). When Ar′ was an unsubstituted phenyl group (**3c**), antimicrobial activity was higher than when electron-donating substituents were present (Table 4).

- The antimicrobial screening of **3a–e** led to the following assumptions about the SAR against Gram-positive bacteria represented by *S. aureus* (Table 4):
- Pyrazole-carbothiamido derivatives are known for their biological activities [11]. In our case, the biological activity of **10** is higher than in that of the reference antibiotic (Fig. 3).
- Thiazoles are also known to have high biological activity. In our case, the biological activity of **15** is higher than that of the reference antibiotic (Fig. 3).
- Thioureas are well known by their biological activities, especially as antimicrobial agents [8]. Compound **19** showed high biological activity against Gram-positive bacteria (*S. Aureus*), (Fig. 3).

The SAR of **3a–e** against Gram-negative bacteria represented by *E. coli* (Table 5) can be summarized as follows:

- As mentioned before, the type of the substitution on the naphthyl ring in Ar did not largely affect the antimicrobial activity, but the substitution on the benzene ring is important. The presence of an electron-withdrawing

Table 4

Antimicrobial activity of **3a–d**, **10**, **15** and **19** against Gram-positive bacteria (*Staphylococcus aureus*).

	Inhibition zone (mm)			
Conc (mg/dl)	100	10	1	0.1
Methanol	Zero			
Reference antibiotic (Ampiclox)	4.1			
Compounds				
3a	5.2	1.6	0.9	0.6
3b	5.4	2.0	1.1	0.7
3c	5.9	2.5	1.9	1.0
3d	6.1	2.0	1.0	0.9
3e	4.7	1.8	1.3	0.7
10	4.8	1.3	1.0	0.5
15	5.6	2.1	1.5	0.6
19	8.2	4.8	2.6	1.5

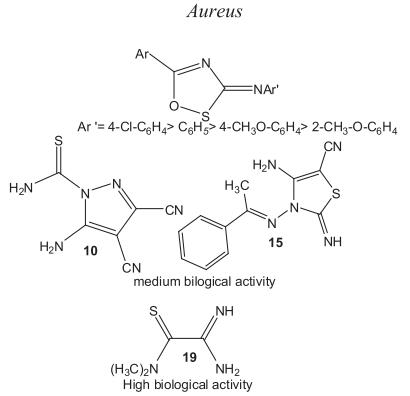


Figure 3. Order of antimicrobial activity of **3a–d**, **10**, **15** and **19** against Gram-positive *Staphylococcus Aureus.*

Table 5

Antimicrobial activity of selected compounds against Gram-negative bacteria (*E. coli*).

	Inhibition zone (mm)			
Conc (mg/dl)	100	10	1	0.1
Methanol	Zero			
Reference antibiotic (Ampiclox)	4.1			
Compounds				
3a	3.2	1.2	0.5	0.04
3b	3.4	1.3	0.6	0.06
3c	3.0	1.5	0.4	0.03
3d	4.6	2.0	0.8	0.08
3e	2.9	1.0	0.3	0.02
10	4.2	1.8	1.6	0.06
15	4.4	1.9	1.7	0.07
19	5.2	2.2	2.0	0.14

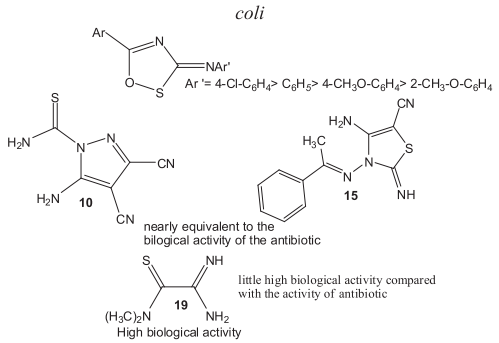


Figure 4. Order of antimicrobial activity of **3a–d**, **10**, **15** and **19** against Gram-negative *Escherichia coli*.

group such as a chlorine atom at the *para* position of the benzene ring (Ar′) in **3d** increased the antimicrobial activity. The same trend was observed in the cases of **3b**, **3c,** and **3d** (Fig. 4). The antibacterial activity of **3b** and **3c** is lower than that of the reference antibiotic, but in case of **3d**, the antibiotic activity against the assigned bacteria is higher than that of the reference antibiotic.

- As previously noted, the activity of compounds **19** against Gram-positive bacteria is high compared with the reference antibiotic. Compound **19** has higher activity against Gram-positive bacteria than against Gram-negative bacteria.

CONCLUSION

In conclusion, the activities of all products **3a–e**, **10**, **15,** and **19** against Gram-positive bacteria (*S. aureus*) are higher compared with their activities against Gram-negative bacteria (*E coli*). Much work should been carried out to discover more about the chemistry of AETC.

EXPERIMENTAL

Chemistry. TLC was performed on analytical Merck 9385 silica aluminum sheets (Kieselgel 60) with PF_{254} indicator. TLCs were viewed under $v = 254$ nm. Melting points were determined on a Stuart (Bibby Scientific Ltd., Stone, Staffordshire, UK) electrothermal melting point apparatus and are uncorrected. IR spectra were recorded as KBr disks on a Shimadzu-408 infrared spectrophotometer (Shimadzu Corp., Kyoto, Japan), Faculty of Science, El Minia University. NMR spectra were measured in DMSO-d_6, at 400 MHz for ^{1}H and 100 MHz for ^{13}C, using a Bruker AV-400 spectrometer (Bruker BioSpin Corp., Billerica, MA) at Florida Institute of Technology. Chemical shifts are expressed as δ (ppm). Coupling constants are stated in Hz. Electron impact (EI) mass spectra were recorded with a JEOL JMS-600 spectrometer (JEOL Ltd., Tokyo, Japan) at an ionization voltage of 70 eV at the Central lab, Assiut University and the Micro analytical center, Faculty of Science, Cairo University, Cairo, Egypt. Thiosemicarbazone **12** was prepared according to the literature [12]. All chemicals and solvents were bought from Aldrich.

General procedure. General procedure of preparing aroyl thioureas 1a-e. Under dry conditions, the aroyl chloride (30 mmoL) was added to a solution of (30 mmol, 2.28 g) NH_4SCN in dried acetone (15 mL) with magnetic stirring at room temperature over 10 min, after which a white precipitate of NH_4Cl appeared. Without filtration, 30 mmoL of the appropriate aromatic amine in acetone (20 mL) was slowly added at room temperature (30°C) with stirring. When the mixture reached room temperature, it was refluxed for 10 min. After cooling, the reaction mixture was poured slowly into 400-mL ice water with strong stirring. Aroyl thioureas **1a–e** were precipitated as solids. The crude product was filtered off, dried, and recrystallized from solvents then washed with cool diethyl ether.

Reaction of thioureas with 1,1,2,2-ethenetetracarbonitrile (2).

General procedure. To a stirred solution of 3 mmoL of thioureas in nearly 100 mL of 1,4-dioxane together with two drops of piperidine, a solution of **2** (0.384 g, 3 mmoL) was added dropwise in 10 min. The reaction was then refluxed for 10–16 h and the precipitates formed were filtered off and recrystallized. Then, 50 mL of *n*-hexane was added to the filtrate and the precipitated formed was identified as tetracyanoethane (**4**, mp 173°C, lit. [13] 170–175°C).

4-Methoxy-N-(5-naphthalen-1-yl)-3H-1,2,4-oxathiazol-3-ylidene)aniline (3a). Pale red crystals (EtOH), (0.75 g, 75%); mp 224–226°C. IR: $v = 3090$–3050 (w, Ar–CH), 2980–2875 (w, aliph.–CH), 1560 (s, C═C) cm^{-1}. ^{1}H-NMR: $\delta = 3.95$ (s, 3H, OCH_3), 6.94 (dd, 2H, $J = 8.0$, 1.0 Hz, Ar–H), 7.60–7.80 (m, 4H, naphth–H), 7.90–8.20 (m, 3H, naphth–H), 8.50–8.54 (dd, 2H, $J = 7.8$, 1.0 Hz, Ar–H). ^{13}C-NMR: $\delta = 158.6$, 156.5 (*C*═N), 152.5 (Ph–*C*–OCH_3), 142.6, 136.7 (naphth–*C*), 129.8 (naphth–2CH), 128.6 (Ar–2CH–*o*), 128.0, 127.8 (naphth–*C*H), 127.2 (naphth–*C*), 126.9 (naphth–2*C*H), 124.0 (Ar–*C*), 118.0 (naphth–*C*H–2), 116.8 (Ar–2CH–*m*), 57.8 (OCH_3). Calcd. for $C_{19}H_{14}N_2O_2S$ (334.39): C, 68.24; H, 4.22; N, 8.38; S, 9.59. Found: C, 68.00; H, 4.10; N, 8.50; S, 9.67%.

4-Methoxy-N-(5-naphthalen-2-yl)-3H-1,2,4-oxathiazol-3-ylidene)aniline (3b). Pale red crystals (EtOH), (0.85 g, 85%); m.p. 287–288°C. IR: $v = 3080$–3052 (w, Ar–CH), 2960–2870 (w, aliph.–CH), 1562 (s, C═C) cm^{-1}. ^{1}H-NMR: $\delta = 3.90$ (s, 3H, OCH_3), 6.90 (dd, 2H, $J = 8.0$, 1.0 Hz, Ar–H), 7.40–7.50 (m, 3H, naphtha–H), 7.62–7.72 (m, 3H, naphth–H), 7.80–7.84

(dd, 2H, $J = 8.0$, 1.0 Hz, Ar–H), 8.10–8.15 (d, 1H, $J = 1.2$ Hz, naphth–H). ^{13}C-NMR: $\delta = 156.8$, 156.0 (*C*═N), 153.0 (Ar–*C*–OCH_3), 145.0 (naphth–*C*–2), 135.6, 133.8 (naphth–*C*), 129.8 (Ar–2*C*H–*m*), 128.6 (naphth–2*C*H), 127.2 (naphth–*C*H), 126.9 (naphth–2*C*H), 124.0 (Ar–*C*H), 122.0 (naphth–*C*H), 119.0 (naphth–*C*H–1), 116.8 (Ar–2CH–*o*), 57.8 (OCH_3). Calcd. for $C_{19}H_{14}N_2O_2S$ (334.39): C, 68.24; H, 4.22; N, 8.38; S, 9.59. Found: C, 68.48; H, 4.10; N, 8.12; S, 9.70%.

***N*-(5-Naphthalen-1-yl)-3H-1,2,4-oxathiazol-3-ylidene)aniline (3c).** Brown crystals (EtOH), (0.64 g, 70%); mp 260–262°C. IR: $\nu = 3060$–3040 (w, Ar–CH), 1560 (s, C═C) cm^{-1}. ^{1}H-NMR: $\delta = 6.86$–6.90 (dd, 2H, $J = 8.0$, 1.0 Hz, Ph–H–*o*), 7.10–7.14 (m, 1H, Ph–*p*), 7.36–7.40 (dd, 2H, $J = 8.0$, 1.0 Hz, Ph–*m*), 7.60–7.64 (m, 2H, naphth–H), 8.10–8.30 (m, 4H, naphth–H), 8.25–8.27 (d, 1H, $J = 8.0$, 1.0 Hz, naphth–H–1). ^{13}C-NMR: $\delta = 160.0$, 158.2 (*C*═N), 145.0 (Ph–*C*), 135.4, 134.0 (naphth–*C*), 131.0 (Ph–2*C*H–*m*), 128.3 (naphth–*C*), 128.0 (naphth–2*C*H), 127.8 (naphth–*C*H), 126.6 (Ph–*C*H–*p*), 126.4 (naphth–2*C*H), 122.6 (naphth–*C*H), 119.2 (naphth–*C*H), 122.8 (Ph–2*C*H–*o*). Calcd. for $C_{18}H_{12}N_2OS$ (304.07): C, 71.03; H, 3.97; N, 9.20; S, 10.54. Found: C, 70.88; H, 4.10; N, 9.10; S, 10.50%.

***4*-Chloro-*N*-(5-naphthalen-1-yl)-3H-1,2,4-oxathiazol-3-ylidene) aniline (3d).** Pale red crystals ($CHCl_3$/MeOH), (0.70 g, 69%); mp 200–202°C. IR: $\nu = 3068$–3050 (w, Ar–CH), 1565 (s, C═C). ^{1}H-NMR: $\delta = 6.92$–6.95 (dd, 2H, $J = 7.8$, 1.0 Hz, Ph–*o*), 7.40–7.44 (dd, 2H, $J = 8.0$, 1.0 Hz, Ph–*m*), 7.58–7.62 (m, 2H, naphth–H), 8.08–8.22 (m, 4H, naphth–H), 8.40–8.43 (dd, 1H, $J = 7.8$, 1.0 Hz naphth–CH–1). ^{13}C-NMR: $\delta = 160.4$, 158.6 (*C*═N), 145.8 (Ph–*C*), 136.0, 134.8 (naphth–*C*), 132.0 (Ph–*C*–Cl), 130.4 (Ph–2*C*H–*m*), 128.2 (naphth–*C*), 128.0 (naphth–*C*H), 127.8 (naphth–2*C*H), 127.6 (naphth–*C*H), 126.6 (naphth–2*C*H), 122.4 (naphth–*C*H), 122.8 (Ph–2*C*H–*o*), 119.4 (naphth–*C*H–1). Calcd. for $C_{18}H_{11}ClN_2OS$ (338.81): C, 63.81; H, 3.27; N, 8.27; S, 9.46. Found: C, 63.70; H, 3.15; N, 8.20; S, 9.40%.

***2*-Methoxy-*N*-(5-naphthalen-1-yl)-3H-1,2,4-oxathiazol-3-ylidene) aniline (3e).** Pale red crystals (ethyl acetate), (0.75 g, 75%); mp 140–142°C. IR: $\nu = 3090$–3050 (w, Ar–CH), 2950–2840 (w, aliph.–CH), 1560 (s, C═C) cm^{-1}. ^{1}H-NMR: $\delta = 3.92$ (s, 3H, OCH_3), 6.70–6.73 (dd, 1H, $J = 7.8$, 1.0 Hzl, Ph–H), 7.00–7.30 (m, 3H, Ph–H), 7.40–7.60 (m, 4H, naphth–H), 8.10–8.22 (m, 2H, naphth–H), 8.40–8.43 (dd, 1H, $J = 7.8$, 1.0 Hz, naphth–H). ^{13}C-NMR: $\delta = 160.0$, 158.0 (C═N), 153.2 (Ph–*C*–OCH_3), 136.2, 135.0 (naphth–*C*), 134.2 (Ph–*C*), 130.2 (Ph–*C*H), 128.4 (naphth–2*C*H), 128.2 (Ph–*C*H), 128.0 (naphth–*C*), 127.0 (Ph–*C*H), 127.4 (naphth–2*C*H), 127.0, 125.8, 122.4 (naphth–*C*H), 120.2 (Ph–*C*H), 55.0 (O*C*H_3). Calcd. for $C_{19}H_{14}N_2O_2S$ (334.39): C, 68.24; H, 4.22; N, 8.38; S, 9.59. Found: C, 68.36; H, 4.30; N, 8.24; S, 9.40%.

Reaction of thiosemicarbazide 7 with **2**. To a stirred solution of thiosemicarbazide (**7**, 0.273 g, 3 mmol) in 50 mL of 1,4-dioxane together with two drops of piperidine, a solution of **2** (0.384 g, 3 mmoL) was added dropwise in 10 min. The reaction was then refluxed for 6 h and the precipitate formed was filtered off and recrystallized from EtOH.

Dicyano-5-imino-2,5-dihydro-1H-pyrazole-1-carbothioamide (10). Red crystals (0.48 g, 82%); mp 134–136°C. IR: $\nu = 3360$–3300 (m, NH_2, NH), 2220 (s, CN), 1560 (s, C═C) cm^{-1}. ^{1}H-NMR: $\delta = 10.4$ (s, 2H, NH–thioamido), 6.6 (s, 2H, NH_2). ^{13}C-NMR: $\delta = 178.6$ (C═S), 156.2 (C═N, C-5), 134.8 (C-3), 111.2, 113.6 (CN), 78.6 (C-4). Calcd. for $C_6H_4N_6S$: C, 37.49; H, 2.10; N, 43.73; S, 16.68. Found: C, 37.30; H, 2.08; N, 43.90; S, 16.60%.

Reaction of thiosemicarbazone 12 with AETC, **13**. To a stirred solution of thiosemicarbazone (**12**,0.179 g, 1 mmol) in nearly 30 mL of 1,4-dioxane together with two drops of piperidine, a solution of **13** (0.118 g, 1 mmoL) was added dropwise in 30 min. The reaction was then refluxed for 4 h and the precipitate formed was filtered off and recrystallized from EtOH.

***4*-Amino-3-(benzylidene-amino)-2-imino-2,3-dihydrothiazole-5-carbonitrile (15).** Yellow crystals (methanol), yield (0.206 g, 85%), mp 182–184°C. IR: $\nu = 3340$–3325 (NH_2–NH, m), 2220 (CN, s), 1600 (C═N, s), 1560 (C═C, m) cm^{-1}. ^{1}H-NMR: $\delta = 10.50$ (br, s, 1H, NH), 8.20 (s, 1H, CH═N), 7.10–6.80 (m, 7H, Ph–H, NH_2). ^{13}C-NMR: $\delta = 178.0$ (C-5), 156.0 (C═NH), 154.0 (CH═N), 132.0 (Ph–C), 128.0 (Ph–2CH–*o*), 127.2 (Ph–2CH–*m*), 126.0 (Ph–CH–*p*), 116.0 (CN), 78.0 (C-4). Calcd. for $C_{11}H_9N_5S$: C, 54.31; H, 3.73; N, 28.79. Found: C, 54.44; H, 3.78; N, 28.66%.

Reaction of 1,1-dimethylthiourea (16) with AETC, **13**. To a stirred solution of **16** (0.104 g, 1 mmol) in nearly 20 mL of 1,4-dioxane together with two drops of piperidine, a solution of **13** (0.118 g, 1 mmoL) was added dropwise in 30 min. The reaction was then refluxed for 10 h. The reaction was cooled and the precipitate was extracted with $CHCl_3$. The solvent was evaporated in *vacuum* and formed; the precipitate was filtered off and recrystallized from $CHCl_3$/EtOH.

***2*-Amino-2-imino-*N*,*N*-dimethylethanethioamide (19).** Brown crystals ($CHCl_3$/EtOH), yield (0.112 g, 85%), mp 260–262°C. IR: $\nu = 3320$–3250 (NH_2–NH, m), 1600 (C═N, s) cm^{-1}. ^{1}H-NMR: Table 1. ^{13}C-NMR: Table 1. MS (EI) 131 (100), 116 (67), 97 (12), 89 (10). Calcd. for $C_4H_9N_3S$: C, 36.62; H, 6.91; N, 32.03. Found: C, 54.44; H, 6.80; N, 31.88%.

Material and methods for antioxidant activity [14]. The antioxidant activity of each organic compound was measured alone.

Preparation of CUPRAC assay solution. $CuCl_2$ solution, 1.0×10^{-2} M was prepared by dissolving 0.426 g $CuCl_2.2H_2O$ in water and diluting to 0.250 mL. Ammonium acetate buffer at pH 7, 1.0 M, was prepared by dissolving 19.27 g NH_4AC in water and diluting to 250 mL. Neocuproine (Nc) solution, 7.5×10^{-3} M, was prepared by dissolving 0.039 g Nc in 96% ethanol and diluting to 25 mL with ethanol.

Preparation of standard solution of ascorbic acid antioxidant. The standard solution of ascorbic acid antioxidant was prepared at 1.0×10^{-3} M concentration by dissolving ascorbic acid in water.

Serum extraction and preparation for total antioxidant CUPRAC assay. Freshly collected serum samples were kept at +4°C until just prior to analysis. About 1 mL of serum was transferred to centrifuge tube, 2 mL of 96% ethanol, and 1 mL of distilled water were added and mixed well. About 4 mL of *n*-hexane were added to the mixture and mixed again, and the final mixture was allowed to stand for a few minutes for separation of the phases. The solution was separated using centrifuging technique at 1500 g (5000 rpm) for 5 min. The upper organic phase was separated and transferred to a dark tube. The hexane extraction procedure was repeated and the second hexane extracted was separated and then transferred to the original dark tube so as to combine with the first extract. The combined hexane extracts were dried under N_2 flow, and the residue was taken up in 1 mL of dichloromethane for assay of trichloroacetic acid (TCA)-Protein method.

Preparation of different organic material. The different organic materials were dissolved in 96% ethanol, and serial concentrations were prepared.

Standard CUPRAC antioxidant capacity assay applied to serum extract with standard addition. To a test tube, 1 mL of copper(II) chloride solution, 1 mL of neocuproine solution, and 1 mL of NH_4Ac buffer solution were added in that order. An aliquot (1.5 mL) of serum extract was added to this tube. Ascorbic acid standard and the organic material to be tested were added to this extract. Absorbance readings were made against reagent blank at 450 nm.

Antimicrobial activity assay. The different organic materials were assessed for their antimicrobial activities by culturing *E. coli* on nutrient agar for 24 h and then subculture these Gram-negative bacteria with different concentration of the organic material and with 96% ethanol alone for 24 h. The zone of inhibition with each organic compound was measured and compared with the zone of inhibition caused by ethanol alone. By varying the concentration of the different organic compounds, we calculate minimal inhibitory concentration of such compounds.

CONCLUSION

In conclusion, we have synthesized a new series of *S*-heterocycles in good yields using a convenient set of synthetic conditions. The chemistry of 1-amino-1,2,2-ethenetricarbonitrile is under further investigation. The sulfur heterocycles described herein are promising prospective antioxidant and antibacterial drugs.

Acknowledgments. The authors express their gratitude to Aljouf University, KSA, for the research funds and its support of this investigation. The NMR spectrometer at Florida Institute of Technology was acquired with the assistance of the National Science Foundation (CHE 03–42251).

REFERENCES AND NOTES

[1] Katritzky, A. R.; Rogovoy, B. V. ARKIVOC 2005, iv, 49. http://www.arkat-usa.org/get-file/19522/

[2] Aly, A. A.; Brown, A. B.; Abdel-Aziz, M.; Abuo-Rahma, G. A. A.; Radwan, M. F.; Ramadan, M.; Gamal-Eldeen, A. M. J Heterocycl Chem 2012, 49, 726.

[3] Aly, A. A.; Brown, A. B.; Abdel-Aziz, M.; Abuo-Rahma, G. A. A.; Radwan, M. F.; Ramadan, M.; Gamal-Eldeen, A. M. J Heterocycl Chem 2010, 47, 503.

[4] Aly, A. A.; Ahmed, E. K.; El-Mokadam, K. M. J Heterocycl Chem 2007, 44, 1431.

[5] Aly, A. A.; Hassan, A. A.; Ibrahim, Y. R.; Abdel-Aziz, M. J Heterocycl Chem 2009, 46, 687.

[6] Upadhayaya, R. S.; Vandavasi, J. K.; Kardile, R. A. S.; Lahore, S. V.; Dixit, S. S.; Deokar, H. S.; Shinde, P. D.; Sarmah, M. P.; Chattopadhyaya, J. Eur J Med Chem 2010, 45, 1854.

[7] Milelli, A.; Tumiatti, V.; Micco, M.; Rosini, M.; Zuccari, G.; Raffaghello, L.; Bianchi, G.; Pistoia, V.; Fernando Diaz, J.; Pera, B.; Trigili, C.; Barasoain, I.; Musetti, C.; Toniolo, M.; Sissi, C.; Alcaro, S.; Moraca, F.; Zini, M.; Stefanelli, C.; Minarini, A. Eur J Med Chem 2012, 57, 417.

[8] (a) Chauhana, A.; Sharmaa, P. K.; Kaushika, N.; Kumara, N. Int J Pharm and Pharmaceutical Sci 2011, 3, 461; (b) Limban, C.; Missir, A. V.; Chirita, I. C. Rev Chim (Bucharest) 2011, 62, 685; (c) Semenov, V. E.; Mikhailov, A. S.; Voloshina, A. D.; Kulik, N. V.; Nikitashina, A. D.; Zobov, V. V.; Kharlamov, S. V.; Latypov, S. K.; Reznik,V. S. Eur J Med Chem 2011, 46, 4715.

[9] Jackman, L. M. Dyn Nucl Magn Reson Spectrosc 1975, 203. CAN 83:77850

[10] Abdullahi M.; Saleh M.; Ibrahim H. M.; Halima, I. M.; Safiyya A., Mani M. A. International J Sci and Tech 2013, 3, 300. ISSN 2224–357

[11] Desai, N. C.; Joshi, V. V.; Rajpara, K. M.; Vaghani, H. V.; Satodiya, H. M. J Fluorine Chem 2012, 142, 67.

[12] Tenorio, R. P.; Carvalho, C. S.; Pessanha, C. S.; de Lima, J. G.; de Faria, A. R.; Alves, A. J. de Melo, A. J. T.; Goès, A. J. S. Bioorg Med Chem Lett 2005, 15, 2575.

[13] Zolotoi, A. B.; Konovalikhin, S. V.; Lukin, P. M.; D'yachenko, O. A.; Zil'berg, S. P.; Nasakin, O. E. Chem Heterocycl Compds 1988, 24, 1166.

[14] Apak, R.; Gü lü, K.; Özyünek, M.; Bektaşoğlu, B.; Bener, M. Methods Mol Biol (Totowa, N J) 2010, 594, 215.