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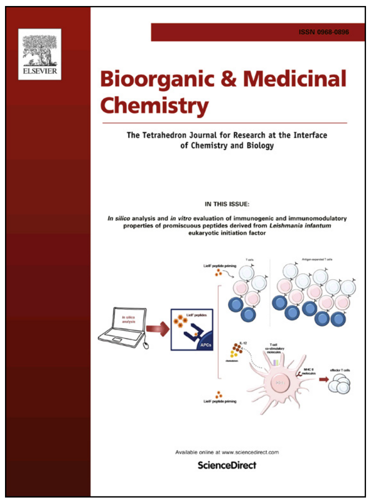

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Selenium-containing analogues of WC-9 are extremely potent inhibitors of *Trypanosoma cruzi* proliferation

María N. Chao,[a] Melissa Storey,[b] Catherine Li,[b] Maricel G. Rodríguez,[c] Florencia Di Salvo,[c] Sergio H. Szajnman,[a,*] Silvia N. J. Moreno,[b] Roberto Docampo,[b] and Juan B. Rodriguez[a,*]

[a]*Departamento de Química Orgánica and UMYMFOR (CONICET–FCEyN), Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires, Pabellón 2, Ciudad Universitaria, C1428EHA, Buenos Aires, Argentina,* [b]*Center for Tropical and Emerging Global Diseases and Department of Cellular Biology, University of Georgia, Athens, Georgia, 30602, USA,* [c]*Departamento de Química Inorgánica, Analítica y Química Física/INQUIMAE-CONICET, Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires, Pabellón 2, Buenos Aires, C1428EGA, Argentina*

Abstract– The obligate intracellular parasite, *Trypanosoma cruzi* is the etiologic agent of Chagas disease or American trypanosomiasis, which is the most prevalent parasitic disease in the Americas. The present chemotherapy to control this illness is still deficient particularly in the chronic stage of the disease. The ergosterol biosynthesis pathway has received much attention as a molecular target for the development of new drugs for Chagas disease. Especially, inhibitors of the enzymatic activity of squalene synthase were shown to be effective compounds on *T. cruzi* proliferation in *in vitro* assays. In the present study we designed, synthesized and evaluated the effect of a number of isosteric analogues of **WC-9** (4-phenoxyphenoxyethyl thiocyanate), a known squalene synthase inhibitor, on *T. cruzi* growth in tissue culture cells. The selenium-containing derivatives turned out to be extremely potent inhibitors of *T. cruzi* growth. Certainly, 3-phenoxyphenoxyethyl, 4-phenoxyphenoxyethyl, 4-(3-fluorophenoxy)phenoxyethyl, 3-(3-fluorophenoxy)phenoxyethyl selenocyanates and (±)-5-phenoxy-2-(selenocyanatomethyl)-2,3-dihydrobenzofuran arose as relevant members of this family of compounds, which exhibited effective ED_{50} values of 0.084 μM, 0.11 μM, 0.083, μM, 0.085, and 0.075 μM, respectively. The results indicate that compounds bearing the selenocyanate moiety are at least two orders of magnitude more potent than the corresponding skeleton counterpart bearing the thiocyanate

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group. Surprisingly, these compounds exhibited excellent selectively index values ranging from 900 to 1,800 making these molecules promising candidates as antiparasitic agents.

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Introduction

Trypanosoma cruzi is the etiologic agent of a chronic zoonotic disease namely American trypanosomiasis or Chagas disease, which is one of the important parasitic diseases worldwide. As a result of public policies for the control of vectors the number of infected people with *T. cruzi* was reduced from 18 million in 1991 to 6 million in 2010.[1–3] However, even now, it is the most prevalent parasitic disease in the Americas.[4] Like other trypanosomatids, *T. cruzi* has a complex life cycle that involves Reduviid bugs with blood-sucking activity and mammalian hosts.[5] The parasite multiplies in the insect gut as an epimastigote form and is released as a non-dividing metacyclic trypomastigote in the insect excrements and invades the host through the intact mucosa or wounds produced by the blood-sucking activity of the vector. In the mammalian host, *T. cruzi* proliferates intracellularly as an amastigote form, which is further released into the blood stream as a non-dividing highly infective trypomastigote.[5] Infection of *T. cruzi* can also take place *via* the placenta or through blood transfusion or organ transplantation, which is the way of transmission in areas where of Chagas disease is not endemic due to increasing migration of infected people.[6]

The present chemotherapy is still deficient and based on two empirically-discovered drugs: nifurtimox (Lampit®, Bayer - El Salvador, **1**) and benznidazole (Abarax®, Elea - Argentina, **2**), which are not FDA-approved in the United States where they are available only from CDC under investigational protocols (Figure 1).[1] These compounds are able to cure at least 50% of recent infections, but they are not effective against the chronic stage of the disease.[7,8] Certainly, therapy with benzonidazole of chronic patients is not satisfactory at all, but a beneficial effect of treatment in such individuals has been well demonstrated.[9,10] Their major drawback is long-term-treatment associated to severe side effects such as vomiting, anorexia, peripheral neuropathy and allergic dermopathy.[7] So far, there are no vaccines available to prevent infection of *T. cruzi*.[8]

nifurtimox benznidazole

Figure 1. Chemical structures of the antiparasitic agents nifurtimox and benznidazole.

Squalene synthase (SQS) is essential in ergosterol biosynthesis. This enzyme catalyzes the first committed step consisting in a two-step reductive dimerization reaction between two molecules

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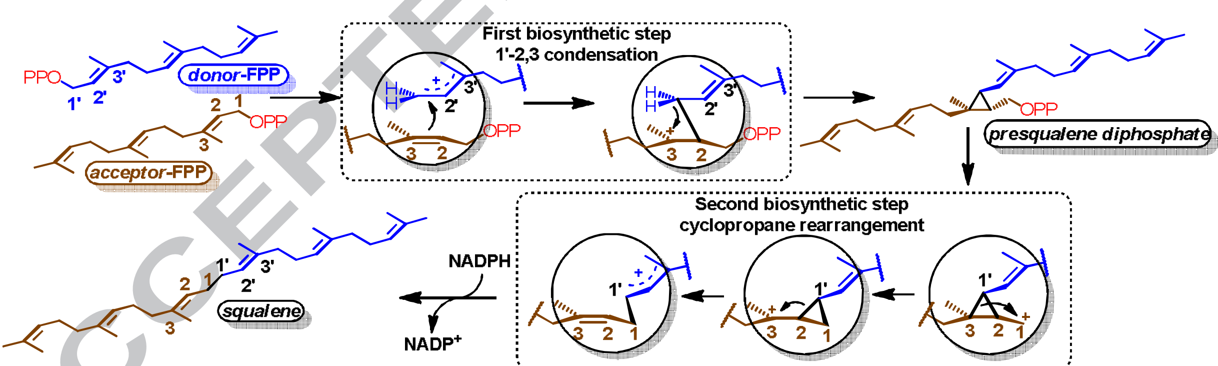
of farnesyl diphosphate (FPP) to give rise to one molecule of squalene,[11] which is an obligated metabolite for the biosynthesis of the required endogenous sterols. There is strong evidence to indicate that the reaction occurs in two well-defined biosynthetic steps.[12,13] As shown in Scheme 1 one molecule of farnesyl diphosphate losses its diphosphate moiety to produce a carbocation species that attacks another molecule of farnesyl diphosphate *via* an 1′-2,3-condensation to give rise to the corresponding intermediate cyclopropylpresqualene diphosphate, which experiences, after leaving the residual diphosphate, a rearrangement reaction of the resulting cyclopropylcarbocation. This carbocation experiences a cyclopropyl ring-opening followed by hydride attack employing NADPH as hydride source.[12,13] Human SQS is a highly conserved membrane-bound protein,[14] and it was crystallized as truncated and active form.[15] A soluble recombinant truncated enzyme from *T. cruzi* (*Tc*SQS), expressed in *Escherichia coli* was also crystallyzed.[16] *Tc*SQS has a dual subcellular localization being equally distributed between glycosomes and mitochondria.[17] SQS has no homologous pyridine dinucleotide-binding motifs suggesting that this enzyme would adapt a distinctive NADPH binding mode from the Rossmann fold in the active site.[18] In this sense, SQS might have one or two overlapping catalytic sites to perform both consecutive reactions.[18] Structural determination of a membrane protein is not a trivial task. [19]



Scheme 1. Reductive dimerization of two molecules of farnesyl diphosphate to produce one molecule of squalene.

There are a number of ergosterol biosynthesis inhibitors that can induce parasitological cure in both acute and chronic experimental models of Chagas disease.[17] It worth mentioning that quinuclidine derivatives E5700 and ER-119884, two potent orally active inhibitors of the enzymatic activity of SQS, arise as relevant members of inhibitors targeting SQS. The former

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one, E5700, has shown to have both in vitro and in vivo anti-*T. cruzi* activity.[20] 4-Phenoxyphenoxyethyl thiocyanate (compound **1**; **WC-9**) is a potent inhibitor of the intracellular amastigote forms of *T. cruzi*[21] **WC-9** acting as a non-competitive inhibitor of *Tc*SQS at a low nanomolar range.[22] Additional synthetic derivatives of **WC-9** such as **2–8** are shown in Figure 2.[21,23–28]

Figure 2. Chemical structure of **WC-9** and other closely related inhibitors of *T. cruzi* proliferation.

Rationale

A thiocyanate moiety covalently bonded to the main frame in the molecule of **WC-9** constitutes one of the few cases where this group behaves as a key functional group in a pharmacologically important lead structure.[29] In order to get further insights about the role of the sulfur atom present in the thiocyanate group, isosteric analogues of **WC-9** were envisioned by replacing this sulfur atom by a selenium atom or an oxygen atom leading to selenocyanate or cyanate derivatives respectively. In addition, to study the influence of the special orientation of the thiocyanate group, conformationally rigid analogues of **WC-9** were considered based on the concept that it should exist an optimal conformation for molecular recognition.[30,31] A variety of biological properties of the selenocyanate group have been described [32] and very recently molecules bearing this functionality have been reported as moderate inhibitors of *Leishmania infantum* and *L. braziliensis*[33,34]

Although the target of **WC-9** has been identified as *Tc*SQS, little is known about the precise mode of action of this allosteric inhibitor. The crystal structure of the complex **WC-9–**TcSQS is not yet available, but an X-ray crystal structure of **WC-9** with human SQS has been recently reported.[35] A high degree of similarity is observed between the *T. cruzi* and human protein structures. However, these crystallographic data did not provide enough information concerning several points: (a) there was no inhibition data, that is, IC_{50} values were not available; (b) it was not clear which was the role of the sulfur atom present in the thiocyanate moiety; (c) **WC-9** in the crystal **WC-9–***h*SQS was present in an unusual unfavorable conformation, which was more

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than 210 kJ/mol to that found in the crystal structure of **WC-9** alone. Coordinates of **WC-9** in the crystal **WC-9–*h*SQS** were taken from Protein Data Bank (PDB) (3WCD) while the coordinates of **WC-9** are provided in the present study either by single crystal X-ray diffraction determination or by electronic structure calculations by DFT (Density Functional theory). Single point energy calculations for comparison were done employing Gaussian 16 program at DFT/B3LYP using 6-311 ++G(d,p) basic set.[36] Moreover, the crystalline structure of **WC-9** with dehydrosqualene synthase from *Staphylococcus aureus*, an enzyme that catalyzes dehydrosqualene formation, did not provide further insight in the precise mode of action of this compound.[37]

Results and Discussion

In our experience, small structural variations on the **WC-9** chemical structure had a deep effect on the biological activity. The development of the Buchwald coupling reaction has proven to be very useful to access a variety of **WC-9** analogues, which might be putative growth inhibitors of either *T. cruzi* or *T. gondii* cells.[38–42] Since our finding that insect juvenile hormone analogues are cell growth inhibitors, particularly, for *T. cruzi* proliferation,[43] we were able to establish a rigorous structure activity relationship (SAR) on **WC-9** chemical structure.[21,23–28,44] The development of **WC-9** as a lead structure has been reviewed.[1,45,46]

A sulfur atom as a linker between the two aromatic rings was the first isosteric modification of **WC-9** and related analogues conceived. The introduction of this sulfur atom to form the corresponding asymmetric diarylthioether skeletons was accomplished *via* a Buchwald coupling reaction employing potassium orthophosphate as a base, thiophenol, and ethylenglycol as a co-solvent and as a ligand.[47] Therefore, the already described synthetic intermediates **11**[27] and **15**[27] under these particular Buchwald coupling reaction conditions gave the respective tetrahydropyranyl derivatives **12** and **16** in low but reproducible yields. Each tetrahydropyranyl protecting group present in these compounds was removed by treatment with pyridinium 4-toluenesulfonate producing the corresponding alcohols **13** and **17** in excellent yields, which were tosylated to give **14** and **18** in 72% and 82% yields, respectively. On treatment with potassium thiocyanate, in separate experiments, these compounds were converted into the target molecules **9** and **10**, respectively, as illustrated in Scheme 2.

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Scheme 2. Synthetic approach for the preparation of thiocyanate derivatives bearing a diarylthioether as a non polar skeleton.

Conformationally constrained analogues of **WC-9** were straightforwardly prepared starting from 4-phenoxyphenol (**21**). Then, this compound was reacted with allyl bromide *via* a Williamson etherification reaction to give the allyl ether **22** in 93% yield. Claisen rearrangement on **22** by treatment with *N*,*N*-dimethylaniline at 210 ºC produced the respective rearranged product **22**,[48,49] which was epoxidized by treatment with *m*-chloroperoxybenzoic acid in chloroform to produce the corresponding racemic epoxyphenol **24**. Nucleophilic ring opening of the epoxy ring under acidic conditions (diluted hydrochloric acid) gave the expected fused five-membered ring **25** according to the literature.[48,50,51] Certainly, nucleophilic attack took place at the most substituted carbon atom as the reaction *via* a carbocationic transition state.[48,50–52] Once the conformationally constrained **25** was at hand, tosylation of this compound followed by S_N2 nucleophilic substitution with potassium thiocyanate produced the title compound **19**. Aromatization of the ring on **19** was successfully carried out by treatment with 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) in refluxing 1,4-dioxane[53] to yield the target molecule **20** in 24% yield. The synthesis of these conformationally restricted analogues is shown in Scheme 3.

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Scheme 3. Synthetic strategy to access conformationally constrained analogues of **WC-9**.

The isosteric analogues of **WC-9** were easily synthesized. The selenocyate group was introduced by a S_N2 reaction between a suitable tosylate and the commercially available potassium selenocyanate in refluxing tetrahydrofuran in the presence of 18-crown-6.[54] Then, the known tosylates **31**[27] and **33**,[21] prepared from 3-phenoxyphenoxyphenol (**30**) and 4-phenoxyphenoxyphenol (**32**) were transformed, in independent experiments, into the selenocyanate derivatives **27** and **28**, by reaction with potassium selenocyanate in 64% and 71% yields, respectively. The cyanate **29** was synthesized starting from already depicted 4-phenoxyphenoxyethanol (**32**),[55,56] which on treatment with trichloroacetyl isocyanate at −15 ºC followed by digestion with potassium carbonate in methanol-water gave carbamate **34**. This compound was converted into the title compound **29** by treatment with trichloroacetyl isocyanate at −15 ºC in 98% yield[55,56] as shown in Scheme 4.

Scheme 4. Straightforward preparation of the first selenium-containing analogues of **WC-9** and the cyanate derivative **29**.

As preliminary results indicated that selenocyanates derivatives **27** and **28** behaved as very

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effective growth inhibitors of *T. cruzi* growth, it was decided to incorporate this group into the relevant skeletons of the thiocyanate series.[21,23–28] Scheme 5 illustrates the preparation of conformationally constrained selenium-containing analogues of **WC-9**. Therefore, compound **35** was straightforwardly prepared from tosylate **26** by treatment with potassium selenocyanate in refluxing tetrahydrofuran in the presence of 18-crown-6 in 86% yield.[54] The title compounds **36** and **37** were synthesized starting from 3-phenoxyphenol (**38**), which on reaction with *N*,*N*-dimethylaniline at 210 ºC for 10 hours[48,49] was converted into a non-separable mixture of the Claisen rearranged regioisomers **40** and **41**. Epoxidation of the mixture **40/41** by reaction with *m*-chloroperoxybenzoic acid in chloroform led to the respective epoxides, which experienced a spontaneous ring-closing to form alcohols **42** and **43**.[50–52] Each alcohol was treated with tosyl chloride, in independent experiments, and the resulting tosylates **44** and **45** yielded after treatment with potassium selenocyanate,[54] under the usual conditions, the target molecules **36** and **37** in 36% and 75% yields, respectively.

Scheme 5. Synthetic approach to access conformationally constrained selenocyanate derivatives.

The rest of the designed selenium-containing compounds were directly prepared from the appropriate tosylate, which had previously been used for the preparation of the thiocyanate counterpart. Therefore, the title compounds **46−55** were available by treatment of the respective already depicted tosylated (**56**,[26] **57**,[26] **58**,[25] **59**,[24] **60**,[24] **61**,[28] **62**,[28] **63**,[28] **64**,[27] and **65**[27]) with potassium selenocyanate as illustrated in Scheme 6.

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KSeCN, 18-crown-6, THF, reflux, 12 h.

Starting material	Product	Yield
56, $R^1 = R^2 = H; R^3 = I$	**46,** $R^1 = R^2 = H; R^3 = I$	64%
57, $R^1 = R^3 = H; R^2 = I$	**47,** $R^1 = R^3 = H; R^2 = I$	71%
58, $R^1 = R^3 = Cl; R^2 = H$	**48,** $R^1 = R^3 = Cl; R^2 = H$	84%
59, $R^1 = R^2 = H$; R^3 = 3-fluorophenoxy	**49,** $R^1 = R^2 = H$; R^3 = 3-fluorophenoxy	53%
60, $R^1 = R^2 = H$; R^3 = 4-fluorophenoxy	**50,** $R^1 = R^2 = H$; R^3 = 4-fluorophenoxy	76%
61, $R^1 = R^3 = H$; R^2 = 2-fluorophenoxy	**51,** $R^1 = R^3 = H$; R^2 = 2-fluorophenoxy	73%
62, $R^1 = R^3 = H$; R^2 = 3-fluorophenoxy	**52,** $R^1 = R^3 = H$; R^2 = 3-fluorophenoxy	69%
63, $R^1 = R^3 = H$; R^2 = 4-fluorophenoxy	**53,** $R^1 = R^3 = H$; R^2 = 4-fluorophenoxy	67%
64, $R^1 = R^3 = H$; R^2 = 4-chlorophenoxy	**54,** $R^1 = R^3 = H$; R^2 = 4-chlorophenoxy	72%
65, $R^1 = R^3 = H$; R^2 = 3-pyridyloxy	**55,** $R^1 = R^3 = H$; R^2 = 3-pyridyloxy	64%

Scheme 6. Preparation of selected selenocyanate derivatives based on privileged skeletons previously evaluated with the thiocyanate moiety.

Biological evaluation of these new isosteric analogues of **WC-9** was very encouraging. Title compounds **9** and **10** exhibited quite similar biological activity against intracellular *T. cruzi* and tachyzoites of *Toxoplasma gondii* than their isosteric counterparts, that is, **WC-9** and compound **4**, indicating that the replacement of the oxygen bridge between both phenyl groups by a sulfur atom had a weak effect on the activity against these parasites.

Conformationally constrained analogues of **WC-9** such as **19** and **20** were potent inhibitors of *T. cruzi* growth showing similar activity to our lead drug **WC-9** with ED_{50} values of 6.0 µM and 5.2 µM, respectively, whereas **WC-9** had an ED_{50} value of 5.0 µM.[27] Evidently, this fixed conformation was not still optimal for a better recognition. In fact, the potency of these compounds is maintained.

Biological data of isosteric derivatives of **WC-9** where the sulfur atom at the thiocyanate unit was replaced either by a selenium atom or an oxygen atom to give rise to **28** and **29** were very relevant. Definitely, the selenium-containing derivative **28** was an extremely potent inhibitor of *T. cruzi* proliferation showing an ED_{50} value of 0.11 µM, that is, almost 50 times more potent than our lead drug **WC-9** (ED_{50} = 5.0 µM).[27] In addition, this compound exhibited very low in vitro toxicity against Vero cells (ED_{50} > 100 µM) presenting a Selectivity Index (SI) close to 1,000. Interestingly, **28** exhibited practically the same degree of activity against *T. gondii* (ED_{50} = 3.7 µM) compared to **WC-9** (ED_{50} = 4.8 µM).[27] Cyanate derivative **29** was virtually devoid of antiparasitic activity against *T. cruzi* indicating that the atom bonded to the cyanide moiety had a strong influence on biological activity. In this case, the inhibitory action

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certainly increases as the number in the periodic table increases: OCN < SCN < SeCN. The obliged synthetic precursor **34** was inactive against *T. cruzi*. The effect of the selenium replacement was more noticeable in compound **27**, the isosteric analogue of **4**. **27** was a very potent growth inhibitor of *T. cruzi* (amastigotes) possessing an ED_{50} value as low as 0.085 μM being 130-fold more potent than the parent molecule **4** with a SI value of 1,765. **27** behaved similarly to **28** against *T. gondii* showing similar potency than its thiocyanate counterpart (compound **4**). Therefore, **28** exhibited an ED_{50} value of 2.9 μM against *T. gondii versus* 4.0 μM by **4**.[26]

Bearing in mind the above results, our efforts were focused on *T. cruzi* cells. Of the conformationally rigid selenium-containing analogues **35**, **36** and **37**, the first one emerged as the most potent molecule having EC_{50} values of 0.083 μM, 0.12 μM and 0.41 μM, respectively. These results offered further insights on the spatial distribution either of the rigid non-polar skeleton as well as the orientation of the selenocyanate group. In fact, superimposition of the chemical structures indicated that there was a marked difference among the disposition of the terminal phenyl ring that matched the differences of the inhibitory action observed as illustrated in Figure 3a. Moreover, because all of these regioisomers are racemic mixtures, it seems reasonable to assume, particularly for **35**, that only one enantiomer should have the proper orientation of the selenocyanate unit for better molecular recognition. Figure 3b shows clearly that it is reasonable to assume that one enantiomer should be accountable for biological action.

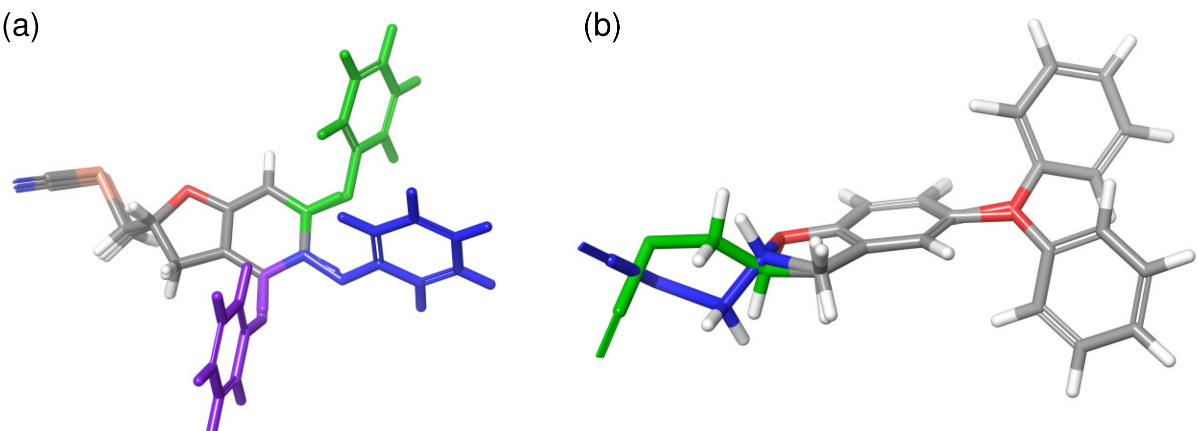

Figure 3. (a) Superimposition of regioisoimers **35–37** alienated on the selenocyanate group **35** blue, **36** green and **37** magenta; (b) different spatial orientations of both enantiomers of **35**: the *R* enantiomer blue and the *S* enantiomer green.

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The rest of the designed compounds also exhibited promising inhibitory action against amastigotes of *T. cruzi*. With the exception of the iodine-containing derivatives **46** and **47**, the rest of the target molecules consisting of a privileged skeleton bonded to an ethyl selenocyanate moiety exhibited a very efficient action as inhibitors of *T. cruzi* growth. Fluorine-containing molecules **49**, **52** and **53** turned out to be extremely potent growth inhibitors of *T. cruzi* proliferation. All of them were at least 50 times more potent than the thiocyanate counterpart being **52** the most potent member of this selenium family of compounds with an ED_{50} value as low as 0.075 μM. **49** and **53** showed ED_{50} values of 0.085 μM and 0.099 μM, respectively. These three compounds had low in vitro toxicity having SI values of more than 1,000. The chlorine-containing compound **54** was very potent as well with an ED_{50} value of 0.11 μM and a SI value greater than 1,000 (Table 1). At the present time, the precise mode of action is still unknown but it could be assumed that the selenium atom of the inhibitors might form a new selenium-sulfur bond with a cysteine residue at the binding site.[32]

The difference in the observed inhibitory action against *T. cruzi* and *T. gondii* may be attributed by the fact that *T. gondii* does not synthesize cholesterol and imports it from the host[57] suggesting that inhibitors of the host SQS could possibly slow *T. gondii* multiplication. As a matter of fact, mevalonate pathway inhibitors modulate proliferation of different Apicomplexan parasites including *Plasmodium falciparum*,[58–60] indicating that microorganisms where the mevalonate pathway is absent are reliant on host metabolites of isoprenoid biosynthesis. *T. gondii* lacks the mevalonate pathway and utilizes a prokaryotic-type 1-deoxy-D-xylulose-5-phosphate (DOXP) pathway instead to produce isopentenyl diphosphaste and dimethylallyl diphosphate. The DOXP pathway localizes to the apicoplast and is essential.[61,62]

X-Ray single crystal structure determination of WC-9 and 28

Molecular structure description. The molecular structures for **WC-9** (CCDC number: 1561895) and **28** (CCDC number: 1561896) have been unequivocally established by X-ray crystallography (Figure 4) and are in agreement with the spectroscopic data.[21] Crystal and data collection details are shown in Table 2 and Supplementary Data. Whereas **WC-9** crystallizes in the $P2_1/n$ space group, **28** does it in P-1. Both molecular structures show the diphenyl ether moiety with comparable bond distances and angles (Supplementary Data) but the C6O1C7 angle and

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C5C6O1C7 torsion angle show some differences: 117.60(14)º and 118.5(3)º for C6O1C7 and 4.28(1)º and −11.43(6)º for C5C6O1C7, respectively, but all of the values according to expected values (see CSD search analysis). The C13 and C14 next to the XCN group exhibit similar structural parameters and as expected, the identity of the X atom, S or Se, impacts in the XCN structural features (Table 2). The electronic effect exerted by the identity of the chalcogen in the XCN group could be influencing the structural differences observed in the diphenyl ether moiety.

Intramolecular parameters studies using Cambridge Structural Database (CSD). Using the Mogul program,[63] a knowledge base of molecular geometry derived from the CSD, the intramolecular parameters of **WC-9**, **28** and **WC-9** crystallized in SQS[35] (**WC-9**–*h*SQS) were compared with the corresponding ones from similar molecules deposited at the CSD. Bond lengths and angle of **WC-9** and **28** are in agreement with the CSD information but, **WC-9**–*h*SQS present some unusual structural values. The C10O2C13 (Figure 4) angle bond to the SCN and torsion angles C13C14S1(Se1)C15 and N1C15S1C14 related to the C–SCN lie far away from the Gaussian exhibited by the database information (Supplementary Data). These results would demonstrate unusual structural features observed for **WC-9**–*h*SQS through a comparison and statistical analysis based on experimental data of related compounds deposited at the CSD.

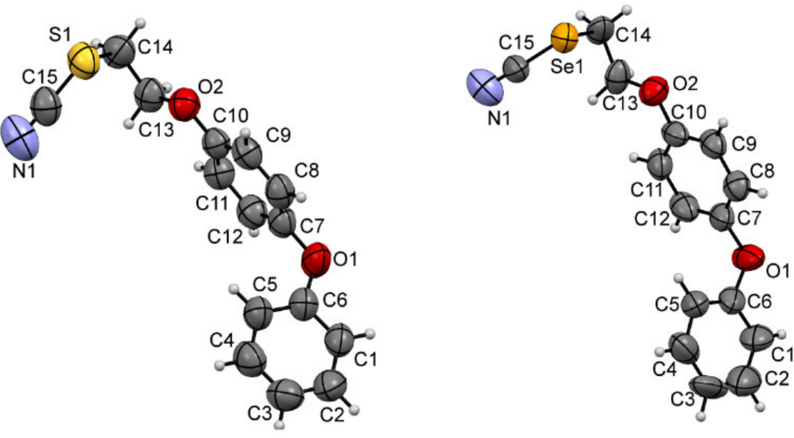

Figure 4. Molecular structure of **WC-9** and **28** with the atomic numbering scheme. H atoms labelling is omitted for clarity, ellipsoids are drown with 50% probability.

Supramolecular behaviour. The supramolecular packing of **WC-9** and **28** shows some similar features, but there is a distinct aspect related with the presence of the chalcogen. The selenocyanate derivative shows a weak H-bond interaction between the N and the C12–H which

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is not present in the thiocyanate analogue (Supplementary Data). The electronic effect exerted by the Se results in an increase of the electronic density in the N atom acting as H-bond acceptor which thus, gives place to the mentioned interaction. This result could be considered as an experimental demonstration of the supramolecular interactions that each derivative is able to experiment in different environments such as solvent and/or molecular targets. Electron density maps of both structures were created using the program Tonto implemented in CrystalExplorer[64] using DFT at the B3LYP/STO-3G[65] level to confirm the observed interactions in general and the different role of the chalcogen being the SeCN involved in stronger intermolecular interactions (Supplementary Data).

It can be concluded that most target molecules, that is, the selenium-containing analogues of **WC-9** designed and synthesized in the present work, exhibited potent inhibitory action against *T. cruzi* proliferation. These isoteric analogues of **WC-9** showed increased potency. In fact, each skeleton increased its efficacy when a thiocyanate group was replaced by a selenocyanate moiety by a factor ranging from 46 to 140. Figure 5 shows the increment in efficacy of the selenium-containing compounds versus the sulfur ones. Table 1, columns # 4 and # 5, indicates the precise increment values. This isosteric structural variation was not beneficial against *T. gondii* maintaining the same level of inhibitory action as it was the case for compounds **27** and **28**. Moreover, all of these title compounds can be considered as having drug-like characteristics[66] offering great potential for optimization of their chemical structure (For details parameters calculated by DataWarrior,[67] see Table S1 in Supplementary Data). Efforts to further optimize the structure of selenium-containing analogues of **WC-9** analogues are currently being pursued in our laboratory.

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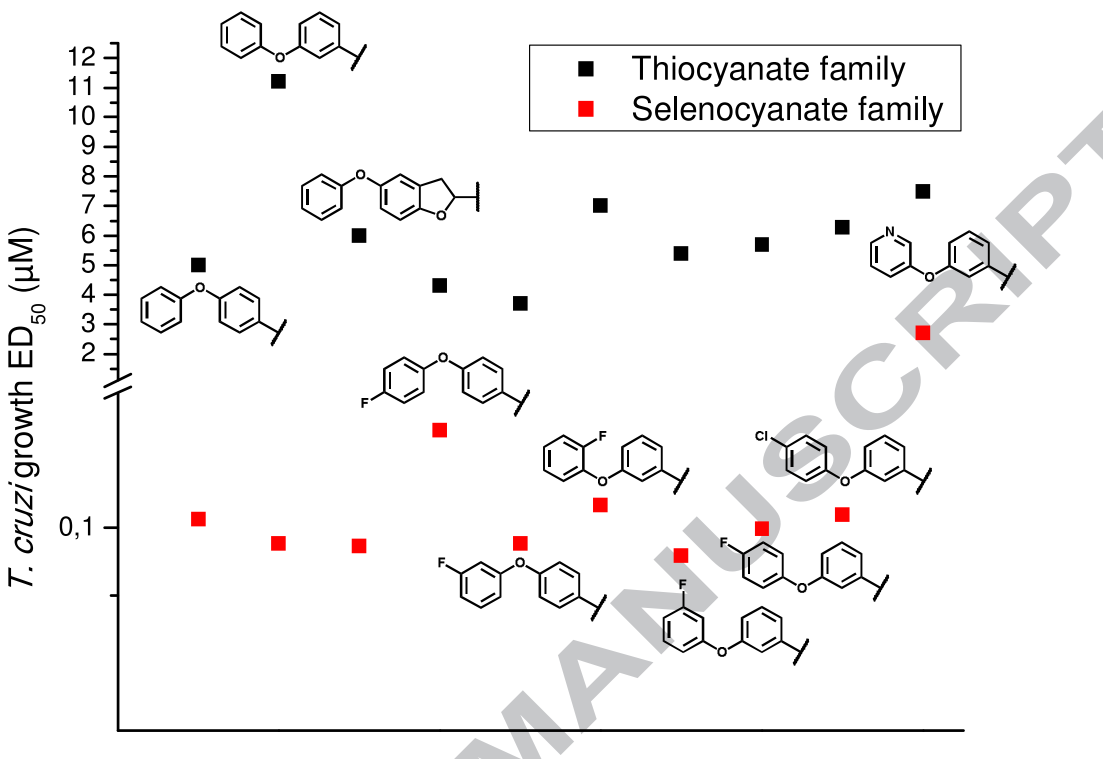

Figure 5. *T. cruzi* amastigote replication ratio between ED_{50} values (μM) of thiocyanate/selenocyanate derivatives.

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Table 1. Biological activity of isosteric analogues of **WC-9** against *T. cruzi* (amastigotes), *T. gondii* (tachyzoites) and Vero cells.

Compd	*T. cruzi* growth ED_{50} (μM)[a]	*T. gondii* growth ED_{50} (μM)[a]	*T. cruzi* growth SCN counterpart ED_{50} (μM)	Ratio *T. cruzi* growth SCN/SeCN	Cytotoxicity ED_{50} (μM)	Selectivity Index [b]
9	5.73 ± 0.41	4.583 ± 1.512	-	-	> 62.5	> 10.9
10	> 10.0	3.581 ± 0.944	-	-	> 50.0	5
19	5.99 ± 0.55	> 10.0	-	-	> 62.5	> 10.4
20	5.19 ± 0.32		-	-		
27	0.085 ± 0.004	2.867 ± 0.924	11.2[26]	131.8	> 150	> 1,765
28	0.109 ± 0.011	3.704 ± 0.463	5.0 ± 1.1[26]	45.9	> 100	> 917
29	> 10.0	Not tested	-	-		
34	> 10.0	Not tested	-	-		
35	0.083 ± 0.019	Not tested	5.99 ± 0.55	72.2	> 125	> 1,500
36	0.120 ± 0.016	Not tested	-	-	> 125	> 1,042
37	0.407 ± 0.051	Not tested	-	-	> 125	> 307
46	> 10.0	Not tested	-	-	> 50	5
47	5.44 ± 0.23	Not tested	-	-	> 62.5	> 11.5
48	1.38 ± 0.29	Not tested	>100[25]	-	> 62.5	> 45.3
49	0.085 ± 0.020	Not tested	4.3[24]	50.6	152.7 ± 63.8	1,796
50	0.271 ± 0.010	Not tested	3.7[24]	13.6	121.5 ± 20.4	1,227
51	0.126 ± 0.04	Not tested	7.01 ± 0.51[28]	55.6	140.4 ± 35.3	1,114
52	0.075 ± 0.005	Not tested	5.38 ± 0.82[28]	71.3	>> 62.5	>> 833
53	0.099 ± 0.020	Not tested	5.69 ± 0.47[28]	57.5	> 125	> 1,263
54	0.114 ± 0.020	Not tested	6.27 ± 0.75[27]	55.0	> 125	> 1,096
55	2.71 ± 0.31	Not tested	7.49 ± 1.39[27]	2.8	47.8 ± 27.5	17.6
WC-9	5.0 ± 1.1[27]	4.8 ± 0.41[27]	5.0 ± 1.1[27]	-	82.6 ± 7.3[27]	16.5
benznidazol	1.50 ± 0.08					

[a]Data are the mean_SD of one experiment carried out in triplicate. [b]Selectivity index: (ED_{50}cytotoxicity)/(ED_{50}parasite).

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Table 2. Crystal data and structure refinement details for **WC-9** and **28.**

Compound	**WC-9**	**28**
CCDC Number	1561895	1561896
Empirical formula	$C_{15}H_{13}NO_2S$	$C_{15}H_{13}NO_2Se$
Formula weight	271.32	318.22
Temperature/K	298.15	298.15
Crystal system	monoclinic	triclinic
Space group	$P2_1/n$	P-1
a/Å	8.5141(5)	8.7845(6)
b/Å	18.0328(6)	8.8130(6)
c/Å	9.7129(5)	10.0457(9)
α/°	90	111.320(8)
β/°	111.742(6)	92.726(7)
γ/°	90	100.516(6)
Volume/$Å^3$	1385.16(12)	706.79(10)
Z	4	2
$\rho_{calc}g/cm^3$	1.301	1.495
μ/mm^{-1}	0.230	2.653
F(000)	568.0	320.0
Crystal size/mm^3	0.5 × 0.2 × 0.1	0.5 × 0.3 × 0.1
Radiation	MoKα (λ = 0.71073)	MoKα (λ = 0.71073)
2Θ range for data collection/°	8.014 to 58.388	7.482 to 57.82
Index ranges	$-11 \leq h \leq 11$, $-24 \leq k \leq 24$, $-13 \leq l \leq 13$	$-11 \leq h \leq 11$, $-11 \leq k \leq 11$, $-13 \leq l \leq 13$
Reflections collected	17783	16553
Independent reflections	3430 [R_{int} = 0.0487, R_{sigma} = 0.0312]	3406 [R_{int} = 0.0705, R_{sigma} = 0.0532]

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Data/restraints/parameters	3430/0/172	3406/0/172
Goodness-of-fit on F^2	1.023	1.045
Final R indexes [I>=2σ (I)]	R_1 = 0.0474, wR_2 = 0.1116	R_1 = 0.0434, wR_2 = 0.0933
Final R indexes [all data]	R_1 = 0.0815, wR_2 = 0.1314	R_1 = 0.0759, wR_2 = 0.1137
Largest diff. peak/hole / e $Å^{-3}$	0.19/-0.22	0.35/-0.39

Supplementary Data. The Supplementary Data is available at . Physical data and spectral information of the target molecules and the corresponding intermediates as well as copies of the ^{1}H NMR, ^{13}C NMR, and ^{19}F NMR spectra are included as Supplementary Data. Moreover, additional Figures indicating intramolecular interaction, electronic density surface, CSD structural studies, X-ray diffraction studies tables and atomic coordinates for compounds **WC-9** and **28**.

Experimental Section

The glassware used in air- and/or moisture-sensitive reactions was flame-dried and reactions were carried out under argon. Unless otherwise noted, chemicals were commercially available and used without further purification. Solvents were distilled before use. Tetrahydrofuran was distilled from benzophenone ketyl. Dichloromethane was distilled from phosphorus pentoxide. Nuclear magnetic resonance spectra were recorded with a Bruker Avance II 500 MHz or a Bruker Fourier 300 spectrometers. The ^{1}H NMR spectra are referenced with respect to the residual $CHCl_3$ proton of the solvent $CDCl_3$ at δ = 7.26 ppm. Coupling constants are reported in Hz. ^{13}C NMR spectra were fully decoupled and are referenced to the middle peak of the solvent $CDCl_3$ at δ = 77.0 ppm. Splitting patterns are designated as s, singlet; d, doublet; t, triplet; q, quadruplet; dd, double doublet, etc. Melting points were determined with a Fisher–Johns apparatus and are uncorrected. IR spectra were recorded with a Nicolet Magna 550 spectrometer. Elemental analyses were performed with an Exeter CE-440 Elemental Analyzer. Analytical TLC was performed on commercial 0.2 mm aluminum-coated silica gel plates (F_{254}) and visualized by

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254 nm UV or immersion in an aqueous solution of $(NH_4)_6Mo_7O_{24}$•$4H_2O$ (0.04 M), $Ce(SO_4)_2$ (0.003 M) in concentrated H_2SO_4 (10%).

Synthesis of selenocyanates. General procedure

A solution of the corresponding tosylate (10 mmol), potassium selenocyanate (11 mmol), and 18-crown-6 (0.1 mmol) in anhydrous tetrahydrofuran (30 mL) was refluxed for 10 h. The solution was cooled to room temperature and the mixture was partitioned between brine (50 mL) and methylene chloride (30 mL). The aqueous phase was extracted with methylene chloride (3 × 25 mL). The combined organic layers were dried ($MgSO_4$) and the solvent was evaporated. The product was purified by column chromatography (silica gel) employing mixtures of hexane-EtOAc as eluent or by HPLC eluting with mixtures of MeOH−H_2O or MeCN−H_2O.

4-(Phenylthio)phenoxyethyl Tetrahydro-2*H*-pyran-2-yl Ether (12). A mixture of compound **11** (1.549 g, 4.5 mmol), thiophenol (490.2 mg, 4.5 mmol), copper(I) iodide (42.3 mg, 0.22 mmol), ethylene glycol (0.57 mL; 641.3 mg, 10.4 mmol), and potassium phosphate tribasic (1.1 g, 5.2 mmol) under anhydrous conditions was evacuated and back-filled with argon. This sequence was repeated twice. Then, 2-propanol (5.0 mL) was added and the reaction mixture was stirred at 80 ºC for 4 days. The mixture was cooled to room temperature and was partitioned between methylene chloride (20 mL) and water (20 mL). The aqueous layer was extracted with methane chloride (2 × 20 mL) and the combined organic phases were washed with brine (5 × 50 mL), dried ($MgSO_4$) and the solvent was evaporated. The product was purified by column chromatography (silica gel) employing hexane−EtOAc (19:1) as eluent to produce 312.3 mg (21% yield) of pure **12** as a colorless oil: R_f 0.51 (hexane−EtOAc (4:1); ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 1.51−1.77 (m, 6H, H-3‴, H-4‴, H-5‴), 3.53 (m, 1H, H-6‴$_a$), 3.82 (ddd, J = 11.3, 6.3, 4.2 Hz, 1H, H-6‴$_b$), 3.91 (ddd, J = 11.3, 8.3, 3.1 Hz, 1H, H-1$_a$), 4.06 (m, 1H, H-1$_b$), 4.17 (m, 2H, H-2), 4.71 (dd, J = 3.9, 3.3 Hz, 1H, H-2‴), 6.93 (d, J = 8.9 Hz, 2H, H-3′), 7.11-7.25 (m, 5H, aromatic protons), 7.40 (d, J = 8.9 Hz, 2H, H-2′); ^{13}C NMR (50 MHz, $CDCl_3$) δ 19.3 (C-4‴), 25.4 (C-5‴), 30.5 (C-3‴), 62.2 (C-6‴), 65.7 (C-1), 67.5 (C-2), 99.0 (C-2‴), 115.7 (C-2′), 124.4 (C-4″), 125.8 (C-4′), 128.2 (C-3′) 128.9 (C-3″), 135.3 (C-2″), 138.6 (C-1″), 159.1 (C-1′).

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4-(Phenylthio)phenoxyethanol (13). A solution of **12** (301.1 mg, 0.9 mmol) in methanol (10 mL) was treated with pyridinium 4-toluenesulfonate (30 mg). The reaction mixture was stirred at room temperature overnight. Then, water (50 mL) was added and the mixture was extracted with methylene chloride (3 × 50 mL). The combined organic layers were washed with brine (3 × 50 mL), dried ($MgSO_4$), and the solvent was evaporated. The product was purified by column chromatography (silica gel) eluting with hexane−EtOAc (17:3) to give 221.3 mg (100% yield) of **13** as a colorless oil: R_f 0.15 (hexane−EtOAc, 4:1); ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 4.00 (t, *J* = 4.6 Hz, 2H, H-1), 4.12 (t, *J* = 4.6 Hz, 2H, H-2), 6.94 (d, *J* = 8.8 Hz, 2H, H-3′), 7.17-7.30 (m, 5H, aromatic protons), 7.43 (d, *J* = 8.8 Hz, 2H, H-2′); ^{13}C NMR (125.77 MHz, CDCl3) δ 61.3 (C-1), 69.3 (C-2), 115.5 (C-2′), 116.9 (C-4″), 125.0 (C-4′), 125.9 (C-4′), 128.4 (C-3′) 128.9 (C-3″), 135.2 (C-2″), 138.3 (C-1″), 158.8 (C-1′).

4-(Phenylthio)phenoxyethyl 4-Toluenesulfonate (14). A solution of **13** (192 mg, 0.78 mmol) in pyridine (3 mL) was treated with *p*-toluenesulfonyl chloride (650 mg, 3.4 mmol) and the mixture was stirred at 0 ºC for 2 h. Then, 5% HCl (50 mL) was added and the reaction mixture was stirred for an additional hour. The mixture was extracted with methylene chloride (50 mL) and the organic layer was washed with 5% HCl (3 × 50 mL) and water (3 × 50 mL). The organic phase was dried ($MgSO_4$), and the solvent was evaporated. The product was purified by column chromatography (silica gel) employing a mixture of hexane−EtOAc (91:9) as eluent to give 243.5 mg of **14** (72% yield) as a colorless oil: R_f 0.35 (hexane−EtOAc, 4:1); ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 2.45 (s, 3H, C*H*3), 4.15 (m, 2 H, H-1), 4.37 (m, 2H, H-2), 6.77 (d, *J* = 8.8 Hz, 2H, H-3′), 7.13-7.24 (m, 5H, aromatic protons), 7.34 (d, *J* = 7.9 Hz, 2H, H-3‴), 7.36 (d, *J* = 8.8 Hz, 2H, H-2′), 7.82 (d, *J* = 8.4 Hz, 2H, H-2‴); ^{13}C NMR (125.77 MHz, CDCl3) δ 21.7 (CH3), 65.5 (C-1), 67.9 (C-2), 115.5 (C-2′), 126.0 (C-4″), 127.2 (C-4′), 128.0 (C-2‴), 128.6 (C-3′) 129.0 (C-3″), 129.9 (C-3‴), 132.8 (C-4‴), 135.0 (C-2″), 138.3 (C-1″), 145.0 (C-1‴), 158.1 (C-1′).

4-(Phenylthio)phenoxyethyl Thiocyanate (9). A solution of **14** (198.3 mg, 0.50 mmol) in anhydrous *N*,*N*-dimethylformamide (3 mL) was treated with potassium thiocyanate (235 mg, 2.42 mmol). The reaction mixture was heated at 90 ºC for 5 h. The mixture was allowed to cool to room temperature and water (20 mL) was added. The aqueous phase was extracted with methylene chloride (2 × 30 mL) and the combined organic layers were washed with brine (5 × 30

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mL) and water (2 × 30 mL). The organic phase was dried ($MgSO_4$), and the solvent was evaporated. The residue was purified by column chromatography (silica gel) eluting with hexane−EtOAc (93:7) to give 89.1 mg (62% yield) of **9** as a colorless oil:: R_f 0.32 (hexane−EtOAc, 4:1); ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 3.69 (t, *J* = 5.8 Hz, 2 H, H-1), 4.35 (t, *J* = 5.8 Hz, 2H, H-2), 6.92 (d, *J* = 8.9 Hz, 2H, H-3′), 7.18-7.30 (m, 5H, aromatic protons), 7.44 (d, *J* = 8.9 Hz, 2H, H-2′); ^{13}C NMR (125.77 MHz, $CDCl_3$) δ 33.2 (C-1), 65.9 (C-2), 111.6 (S*C*N), 115.6 (C-2′), 126.1 (C-4″), 126.3 (C-4′), 128.8 (C-3′) 129.0 (C-3″), 134.9 (C-2″), 137.8 (C-1″), 157.8 (C-1′). HRMS (ESI) calcd. for $C_{15}H_{14}O_2NS_2$ $[M+H]^+$ 288.0517; found 288.0523.

3-(Phenylthio)phenoxyethyl Tetrahydro-2*H*-pyran-2-yl Ether (16). A mixture of compound **15** (1.00 g, 2.9 mmol), thiophenol (316 mg, 2.9 mmol), copper(I) iodide (27.3 mg, 0.14 mmol), ethylene glycol, (0.37 mL; 413.7 mg, 6.7 mmol), and potassium carbonate (793 mg, 5.7 mmol) was evacuated and back-filled with argon. This sequence was repeated twice. Then, 2-propanol (3.0 mL) was added and the reaction mixture was stirred vigorously at 80 ºC for 7 days. The mixture was cooled to room temperature and was partitioned between methylene chloride (20 mL) and water (20 mL). The aqueous layer was extracted with dichloromethane (2 × 20 mL). The combined organic phases were washed with brine (5 × 50 mL), dried ($MgSO_4$), and the solvent was evaporated. The product was purified by column chromatography (silica gel) employing hexane−EtOAc (97:3) as eluent to afford 419 mg (44% yield) of **16** as a colorless oil: R_f 0.48 (hexane−EtOAc, 4:1); ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 1.50−1.64 (m, 4H, H-4‴, H-5‴), 1.70−1.75 (m, 1H, H-3‴$_a$), 1.79−1.86 (m, 1H, H-3‴$_b$), 3.51 (m, 1H, H-6‴$_a$), 3.79 (ddd, *J* = 11.0, 6.5, 4.3 Hz, 1H, H-6‴$_b$), 3.88 (ddd, *J* = 11.3, 8.3, 3.0 Hz, 1H, H-1$_a$), 4.01 (m, 1H, H-1$_b$), 4.10 (m, 2H, H-2), 4.68 (t, *J* = 3.8 Hz, 1H, H-2‴), 6.81 (ddd, *J* = 8.3, 2.4, 1.0 Hz, 1H, H-6′), 6.92 (m, 2H, aromatic protons), 7.25 (m, 1H, aromatic proton), 7.30 (m, 2H, aromatic protons), 7.35 (m, 2H, aromatic protons); ^{13}C NMR (50 MHz, $CDCl_3$) δ 19.3 (C-4‴), 25.4 (C-5‴), 30.5 (C-3‴), 62.2 (C-6‴), 65.7 (C-1), 67.5 (C-2), 99.0 (C-2‴), 113.6 (C-6′), 116.9 (C-2′), 123.2 (C-4′), 127.2 (C-4″), 129.2 (C-3″) 129.9 (C-5′), 131.3 (C-2″), 135.4 (C-1″), 137.0 (C-3′), 159.4 (C-1′).

3-(Phenylthio)phenoxyethanol (17). A solution of **16** (399 mg, 1.2 mmol) in methanol (10 mL) was treated with pyridinium 4-toluenesulfonate (30 mg). The reaction mixture was stirred at room temperature overnight. Then, water (50 mL) was added and the mixture was extracted with

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methylene chloride (3 × 50 mL). The combined organic layers were washed with brine (3 × 50 mL), dried ($MgSO_4$), and the solvent was evaporated. The residue was purified by column chromatography (silica gel) eluting with hexane−EtOAc (17:3) to give 290 mg (97% yield) of pure alcohol **17** as a colorless oil: R_f 0.13 (hexane−EtOAc, 4:1); ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 2.01 (t, *J* = 6.2 Hz, 1H, O*H*), 3.93 (m, 2H, H-1), 4.02 (t, *J* = 6.2 Hz, 2H, H-2), 6.79 (ddd, *J* = 8.3, 2.5, 0.9 Hz, 1H, H-6′), 6.87 (dd, *J* = 2.4, 1.8 Hz, 1H, H-2′), 6.93 (ddd, *J* = 7.7, 1.7, 0.9 Hz, 1H, H-4′), 7.21 (t, *J* = 8.0 Hz, 1H, H-5′), 7.27 (m, 1H, H-4″), 7.32 (m, 2H, H-3″), 7.38 (m, 2H, H-2″); ^{13}C NMR (50 MHz, $CDCl_3$) δ 61.4 (C-1), 69.2 (C-2), 113.2 (C-6′), 116.4 (C-2′), 123.2 (C-4′), 127.4 (C-4″), 129.2 (C-3″) 130.0 (C-5′), 131.6 (C-2″), 135.0 (C-1″), 137.5 (C-3′), 159.1 (C-1′).

3-(Phenylthio)phenoxyethyl 4-Toluenesulfonate (18). A solution of **17** (280 mg, 1.1 mmol) in pyridine (3 mL) was treated with 4-toluenesulfonyl chloride (650 mg, 3.4 mmol) and the mixture was stirred at room temperature for 4 h. Then, 5% HCl (50 mL) was added and the reaction mixture was stirred for an additional hour. The mixture was extracted with methylene chloride (50 mL) and the organic layer was washed with 5% HCl (3 × 50 mL) and water (3 × 50 mL). The organic phase was dried ($MgSO_4$), and the solvent was evaporated. The product was purified by column chromatography (silica gel) employing a mixture of hexane−EtOAc (91:9) as eluent to give 376 mg of **18** (82% yield) as a colorless oil: R_f 0.37 (hexane−EtOAc, 4:1), ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 2.44 (s, 3H, PhC*H*$_3$), 4.08 (m, 2H, H-1), 4.33 (t, *J* = 5.8 Hz, 2H, H-2), 6.65 (ddd, *J* = 8.3, 2.6, 0.5 Hz, 1H, H-6′), 6.70 (t, *J* = 2.5 Hz, 1H, H-2′), 6.90 (ddd, *J* = 7.7, 1.4, 0.8 Hz, 1H, H-4′), 7.16 (t, *J* = 8.0 Hz, 1H, H-5′), 7.27 (m, 1H, H-4″), 7.31 (d, *J* = 8.6 Hz, 2H, H-3‴), 7.32 (m, 2H, H-3″), 7.35 (m, 2H, H-2″), 7.79 (d, *J* = 8.4 Hz, 2H, H-2‴); ^{13}C NMR (50 MHz, $CDCl_3$) δ 21.6 (Ph*C*H$_3$), 65.4 (C-1), 68.0 (C-2), 113.2 (C-6′), 116.5 (C-2′), 123.5 (C-4′), 127.4 (C-4″), 128.0 (C-2‴), 129.3 (C-3″), 129.8 (C-3‴), 130.0 (C-5′), 131.5 (C-2″), 132.8 (C-4‴), 135.0 (C-1″), 137.5 (C-3′), 145.0 (C-1‴), 158.4 (C-1′).

3-(Phenylthio)phenoxyethyl Thiocyanate (10). A solution of **18** (369 mg, 0.92 mmol) in anhydrous *N*,*N*-dimethylformamide (3 mL) was treated with potassium thiocyanate (447 mg, 4.6 mmol). The reaction mixture was heated at 80 ºC for 3 h. The mixture was allowed to cool to room temperature and water (20 mL) was added. The aqueous phase was extracted with

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methylene chloride (2 × 30 mL) and the combined organic layers were washed with brine (5 × 30 mL) and water (2 × 30 mL). The organic phase was dried ($MgSO_4$), and the solvent was evaporated. The residue was purified by column chromatography (silica gel) eluting with hexane−EtOAc (93:7) to give 175 mg (66% yield) of **10** as a colorless oil: R_f 0.37 (hexane−EtOAc, 4:1); ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 3.30 (t, *J* = 5.8 Hz, 2H, H-1), 4.25 (t, *J* = 5.8 Hz, 2H, H-2), 6.79 (ddd, *J* = 8.3, 2.5, 0.8 Hz, 1H, H-6′), 6.86 (dd, *J* = 2.4, 1.8 Hz, 1H, H-2′), 6.94 (ddd, *J* = 7.7, 1.7, 0.9 Hz, 1H, H-4′), 7.22 (t, *J* = 8.0 Hz, 1H, H-5′), 7.28 (m, 1H, H-4″), 7.33 (m, 2H, H-3″), 7.38 (m, 2H, H-2″); ^{13}C NMR (50 MHz, $CDCl_3$) δ 33.2 (C-1), 65.8 (C-2), 111.6 (S*C*N), 113.2 (C-6′), 116.3 (C-2′), 123.7 (C-4′), 127.5 (C-4″), 129.3 (C-3″) 130.1 (C-5′), 131.7 (C-2″), 134.8 (C-1″), 138.0 (C-3′), 158.2 (C-1′). HRMS (ESI) calcd. for $C_{15}H_{13}ONS_2Na$ $[M+Na]^+$ 310.0336; found: 310.0336.

4-Phenoxyphenyl Prop-2-en-1-yl Ether (22). A solution of 4-phenoxyphenol (**21**; 1.00 g, 5.4 mmol) in dimethyl sulfoxide (5.0 mL) was treated with potassium hydroxide (60.3 mg, 10.7 mmol) and the mixture was stirred for 5 min. Then, allyl chloride (41.1 mg, 0.44 ml, 5.4 mmol) was added dropwise. The reaction mixture was stirred at room temperature for 30 min. The reaction mixture was extracted with methylene chloride (2 × 25 mL) and the combined organic layers were washed with brine (5 × 50 mL), dried ($MgSO_4$), and the solvent was evaporated. The product was purified by column chromatography (silica gel) eluting with a mixture of hexane−AcOEt 9:1 to give 1.13 g (93% yield) of **22** as a yellow pale oil: R_f = 0.79 (hexane−EtOAc, 4:1); IR (film, cm^{-1}) 3050,2950, 1580, 1470, 1200, 1100, 900, 800; ^{1}H-NMR ($CDCl_3$, 300.18 MHz) δ 4.52 (dt, *J* = 5.3, 1.6 Hz, 2H, H-1), 5.30 (dq, *J* = 10.5, 1.4 Hz, 1H, H-$3_{cis\ \mathrm{to\ H\text{-}2}}$); 5.42 (dq, *J* = 17.2, 1.6 Hz, 1H, H-$3_{trans\ \mathrm{to\ H\text{-}2}}$), 6.06 (ddt, *J* = 17.3, 10.5, 5.3 Hz, 1H, H-2), 6.88-7.07 (m, 7H, aromatic protons), 7.27-7.32 (m, 2H, aromatic protons); ^{13}C NMR ($CDCl_3$, 75.48 MHz) δ 69.3 (C-1), 115.8 (C-2″); 117.65 (C-3), 117.68 (C-2′), 120.7 (C-3′), 122.5 (C-4), 129.6 (C-3″), 133.3 (C-2), 150.3 (C-4′), 154.9 (C-1′), 158.4 (C-1″).

2-(Prop-2-en-1-yl)-4-phenoxyphenol (23). A solution of **22** (1.2 g, 5.3 mmol) in *N*,*N*-dimethylaniline (1.0 mL) was stirred at 210 ºC for 10 h. The mixture was allowed to cool to room temperature. Then, methylene chloride (20 mL) was added and the mixture was extracted with a 10% aqueous solution of sodium hydroxide (2 × 15 mL). The aqueous phase was acidified

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with 10% hydrochloric acid, and then was extracted with methylene chloride (2 × 25 ml). The combined organic phases were washed with brine (5 × 50 mL), dried ($MgSO_4$), and the solvent was evaporated. The product was purified by column chromatography (silica gel) employing hexane−EtOAc (97:3) as eluent to afford 220 mg (18% yield) of **23** as a colorless oil: R_f = 0.39 (hexane-EtOAc, 4:1); ^{1}H-NMR ($CDCl_3$, 300.18 MHz) δ 3.38 (dt, *J* = 6.4, 1.4 Hz, 2H, H-1), 5.14 (m, 1H, H-3$_{cis\ \mathrm{to\ H\text{-}2}}$); 5.18 (m, H-3$_{trans\ \mathrm{to\ H\text{-}2}}$, 5.99 (ddt, *J* = 17.5, 9.7, 6.4 Hz, H-2), 6.79-6.84 (m, 3H, aromatic protons), 6.91-6.96 (m, 2H, aromatic protons), 7.03 (tt, *J* = 7.4, 1.0, 1H, H-4″), 7.29 (dd, *J* = 8.6, 7.4 Hz, 2H, H-3″); ^{13}C NMR (75.48 MHz, $CDCl_3$) δ 35.1 (C-1), 116.7 (C-5′), 116.8 (C-3), 117.5 (C-2″), 119.0 (C-6′), 121.7 (C-3′), 122.3 (C-4″), 124.5 (C-6′), 126.8 (C-2′), 129.6 (C-3″), 135.9 (C-2), 150.0 (C-1′), 150.3 (C-4′), 158.5 (C-1″).

2-(Oxiran-2-ylmethyl)-4-phenoxyphenol (24). A solution of **23** (202 mg, 0.89 mmol) in methylene chloride (20 mL) was added *m*-chlorobenzoic acid (308 mg, 1.8 mmol). The solution was stirred at room temperature for 5 days. Then, the mixture was washed with a saturated solution of sodium bicarbonate (3 × 30 mL), and water (2 × 20 ml). The organic phase was dried ($MgSO_4$), and the solvent was evaporated to give 205 mg (95% yield) of **24** as a yellowish oil, which was used in the next step without further purification: R_f = 0.32 (hexane−EtOAc, 4:1); ^{1}H NMR ($CDCl_3$, 300.18 MHz) δ 2.68 (dd, *J* = 15.1, 7.4 Hz, 1H, H-1$_a$), 2.74 (dd, *J* = 4.3, 2.9 Hz, 1H, H-3$_a$), 2.94 (t, *J* = 4.1 Hz, 1H, H-3$_b$), 3.19 (dd, *J* = 15.1, 2.4 Hz, 1H, H-1$_b$), 3.30 (ddt, *J* = 5.0, 3.9, 2.5 Hz, 1H, H-2), 6.81-6.96 (m, 5H, aromatic protons), 7.01-7.07 (m, 1H, aromatic proton), 7.27-7.32 (m, 2H, aromatic protons); ^{13}C NMR (75.48 MHz, $CDCl_3$) δ 34.8 (C-1), 48.1 (C-3), 53.5 (C-2), 117.6 (C-6′), 118.2 (C-2″), 120.0 (C-3′), 122.1 (C-5′), 122.4 (C-4″), 124.5 (C-6′), 129.6 (C-3″), 151.8 (C-4′),149.8 (C-1′), 158.5 (C-1″).

(5-Phenoxy-2,3-dihydrobenzofuran-2-yl)methanol (25). Compound **24** (205 mg, 0.85 mmol) was dissolved in acidified $CHCl_3$ (prepared by shaking 180 ml of $CHCl_3$ with 7 drops of concentrated HCl). After 20 min the mixtures was extracted with 1% aqueous $NaHCO_3$. The organic phase was dried (Na_2SO_4) and the solvent was evaporated. The product was purified by column chromatography (silica gel) employing hexane−EtOAc (39:11) as eluent to afford 110 mg (54% yield) of **25** as a colorless oil: R_f = 0.36 (hexane−EtOAc, 3:2); ^{1}H-NMR ($CDCl_3$, 300.18 MHz) δ 3.04 (dd, *J* = 16.0, 7.6 Hz, 1H, H-3$_a$), 3.25 (dd, *J* = 15.8, 9.3 Hz, 1H, H-3$_b$), 3.77

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(dd, J = 12.0, 6.1 Hz, 1H, CH_aHOH), 3.89 (dd, J = 12.1, 3.2 Hz, 1H, CH_bHOH), 4.96 (dddd, J = 9.4, 7.6, 6.1, 3.3 Hz, 1H, H-2), 6.76 (d, J = 8.5 Hz, 1H, H-7), 6.81-6.97 (m, 4H, aromatic protons), 7.02-7.08 (m, 1H, aromatic proton), 7.31 (dd, J = 8.6, 7.4 Hz, 2H, aromatic protons); ^{13}C NMR (75.48 MHz, $CDCl_3$) δ 31.5 (C-3), 64.8 (CH$_2$OH), 83.5 (C-2), 109.7 (C-7), 117.1 (C-6), 117.5 (C-2′), 119.5 (C-4), 122.3 (C-4′), 128.0 (C-4_a), 129.6 (C-3′), 150.4 (C-5), 155.4 (C-7_a), 158.7 (C-1′).

(5-Phenoxy-2,3-dihydrobenzofuran-2-yl)methyl 4-Toluenenesulfonate (26). A solution of alcohol **25** (98.7 mg, 0.41 mmol) in pyridine (3 mL) was treated with 4-toluenesulfonyl chloride (233.0 mg, 1.2 mmol) and the mixture was stirred at room temperature for 4 h. The mixture was extracted with methylene chloride (50 mL) and the organic layer was washed water (3 × 50 mL). The organic phase was dried ($MgSO_4$), and the solvent was evaporated. The product was purified by column chromatography (silica gel) employing a mixture of hexane-EtOAc (91:9) as eluent to afford 140 mg of **26** (87% yield) as a colorless oil: R_f = 0.18 (hexane-EtOAc; 8:2); ^{1}H-NMR ($CDCl_3$, 300.18 MHz) δ 3.04 (dd, J = 16.0, 7.0 Hz, 1H, H-3_a), 3.27 (dd, J = 16.0, 9.6 Hz, 1H, H-3_b), 4.19 (d, J = 5.1 Hz, 2H, CH_2OH), 4.97 (ddt, J = 9.7, 7.1, 5.0 Hz, 1H, H-2), 6.64 (d, J = 8.5 Hz, 1H, H-7), 6.76-6.93 (m, 4H, aromatic protons), 7.01-7.07 (m, 1H, aromatic proton), 7.28 (d, J = 7.4 Hz, 1H, aromatic proton), 7.31 (d, J = 7.5 Hz, 1H, aromatic proton), 7.35 (d, J = 8.0 Hz, 2H, H-3″), 7.80 (d, J = 8.4 Hz, 2H, H-2″).

(±)-5-Phenoxy-2-(thiocyanatomethyl)-2,3-dihydrobenzofuran (19). A solution of **26** (163 mg, 0.34 mmol) in anhydrous *N,N*-dimethylformamide (3.0 mL) was treated with potassium thiocyanate (167 mg, 1.7 mmol). The reaction mixture was stirred at 100 ºC for 3 h. The mixture was allowed to cool to room temperature and water (20 mL) was added. The aqueous phase was extracted with methylene chloride (2 × 30 mL) and the combined organic layers were washed with brine (5 × 30 mL) and water (2 × 30 mL). The combined organic phases were dried ($MgSO_4$) and the solvent was evaporated. The residue was purified by column chromatography (silica gel) eluting with hexane–EtOAc (9:1) to give 80.1 mg (82% yield) of **19** as a colorless oil: R_f =0.33 (hexane–EtOAc, 4:1); ^{1}H-NMR ($CDCl_3$, 500.13 MHz) δ 3.06 (dd, J = 16.1, 6.4 Hz, 1H, H-3_a), 3.25 (dd, J = 13.6, 6.4 Hz, 1H, CH_aHSCN), 3.28 (dd, J = 13, 3.2 Hz, 1H, CH_bHOH), 3.46 (dd, J = 16.1, 9.2 Hz, 1H, H-3_b), 5.10 (ddt, J = 9.2, 6.4, 5.5 Hz, 1H, H-2), 6.77 (d, J = 8.5 Hz,

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1H, H-7), 6.84 (ddt, J = 8.6, 2.4, 0.7 Hz, 1H, aromatic proton), 6.89 (m, 1H, aromatic proton), 6.94 (m, 2H, aromatic protons), 7.05 (tt, J = 7.4, 1.0 Hz, 1H, aromatic proton), 7.29 (d, J = 7.4 Hz, 1H, aromatic proton), 7.31 (d, J = 7.4 Hz, 1H, aromatic proton); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 34.8 (C-3), 38.3 (CH$_2$SCN), 80.6 (C-2), 110.2 (C-7), 111.7 (SCN), 116.9 (C-6), 117.7 (C-2′), 119.9 (C-4), 122.6 (C-4′), 126.5 (C-4$_a$), 129.5 (C-3′), 151.0 (C-5), 154.7 (C-7$_a$), 158.4 (C-1′).

5-Phenoxy-2-(thiocyanatomethyl)benzofuran (20). To a solution of **19** (21.9 mg, 7.73×10^{-2} mmol) in anhydrous dioxane (1.5 mL) was added 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) dissolved in 1.5 ml of the same solvent. The solution was refluxed for 24 h and then was cooled. The solid was filtered off and the filtrate was evaporated under reduced pressure. The residue was purified by column chromatography (silica gel) eluting with hexane-EtOAc (49:1) to give 5.3 mg (24% yield) of **20** as a colorless oil: R_f = 0.30 (hexane−EtOAc, 4:1); ^{1}H-NMR ($CDCl_3$, 500.13 MHz) δ 4.29 (s, 2H, CH_aHSCN), 6.76 (s, 1H, H-3), 6.98 (m, 2H, aromatic protons), 7.07 (m, 2H, aromatic protons), 7.18 (d, J = 2.5 Hz, 1H, aromatic proton), 7.32 (d, J = 7.4 Hz, 1H, aromatic proton), 7.33 (d, J = 7.4 Hz, 1H, aromatic proton), 7.44 (d, J = 8.9 Hz, 1H, aromatic proton); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 31.3 (CH$_2$SCN), 107.4 (C-7), 111.2 (SCN), 111.4 (C-3), 112.2 (C-4), 118.0 (C-6), 118.1 (C-2′), 122.9 (C-4′), 128.7 (C-4$_a$), 129.7 (C-3′), 150.9 (C-7$_a$), 151.9 (C-5), 153.0 (C-2), 158.2 (C-1′).

3-Phenoxyphenoxyethyl 4-Toluenesulfonate (31). A solution of **30**[26] (288.4 mg, 1.2 mmol) in pyridine (3.0 mL) cooled at 0 °C was treated 4-toluenesulfonyl chloride (710.3 mg, 3.7 mmol) portion wise, and the mixture was stirred at 0 ºC for 3 h. Then, a 5% aqueous solution of hydrochloric acid (10 mL) was added and the reaction mixture was stirred for an additional hour. The mixture was extracted with methylene chloride (30 mL) and the organic layer was washed with 5% hydrochloric acid (3 × 25 mL) and water (3 × 30 mL). The organic phase was dried ($MgSO_4$) and the solvent was evaporated. The product was purified by column chromatography (silica gel) eluting with hexane−EtOAc (19:1) to give 312 mg (68 % yield) of **31** as a colorless oil: R_f = 0.47 (hexane−EtOAc, 1:1); ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 2.46 (s, 3H, CH_3), 4.10 (m, 2H, H-2), 4.34 (m, 2H, H-1), 6.41 (t, J = 2.2 Hz, 1H, H-2′), 6.52 (dd, J = 8.3, 2.2 Hz, 1H, H-6′), 6.62 (dd, J = 8.2, 2.0 Hz, 1H, H-4′), 7.00 (d, J = 8.4 Hz, 2H, H-2″), 7.12 (t, J = 7.4 Hz, 1H,

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H-5′), 7.18 (t, *J* = 8.2 Hz, 1H, H-4″), 7.32 (d, *J* = 8.3 Hz, 2H, H-3‴), 7.34 (t, *J* = 8.0 Hz, 2H, H-3″), 7.80 (d, *J* = 8.2 Hz, 2H, H-2‴); ^{13}C NMR (125.77 MHz, $CDCl_3$) δ 21.6 (CH_3), 65.5 (C-2), 68.0 (C-1), 105.5 (C-2′), 109.1 (C-6′), 111.6 (C-4′), 119.1 (C-2″), 123.5 (C-4″), 128.0 (C-2‴), 129.76 (C-3″), 129.82 (C-3‴), 130.2 (C-5′), 132.8 (C-4‴), 145.0 (C-1‴), 156.8 (C-1″), 158.5 (C1′), 159.3 (C-4′).

(4-Phenoxyphenoxyethyl) 4-Toluenesulfonate (33). Alcohol **32**[21] (436.4 mg, 1.9 mmol) in pyridine (3.0 mL) was treated with 4-toluenesulfonyl chloride (1,049 g, 5.1 mmol) as depiected for the preparation of **31**. The residue was purified by column chromatography (silica gel) employing hexane-EtOAc (9:1) as eluent to afford 499.3 mg of **33** (72% yield) as a white solid: mp 65 ºC; ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 2.45 (s, 3 H, C*H*$_3$), 4.14 (m, 2 H, H-2), 4.37 (m, 2 H, H-1), 6.77 (d, *J* = 9.1 Hz, 2H, H-2′), 6.92 (m, 2H, H-2″), 6.93 (d, *J* = 9.1 Hz, 2H, H-3′), 7.05 (tt, *J* = 7.4, 1.0 Hz, 1H, H-4″), 7.30 (dd, *J* = 8.4, 7.4, 2H, H-3″), 7.35 (d, *J* = 8.0 Hz, 2H, H-3‴), 7.85 (d, *J* = 8.3 Hz, 2H, H-2‴); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 21.6 (CH_3), 66.0 (C-1), 68.1 (C-2), 115.8 (C-2″), 117.8 (C-2′), 120.7 (C-3′), 122.6 (C-4″), 128.0 (C-2‴), 129.6 (C-3″), 129.8 (C-3‴), 132.9 (C-4‴), 145.0 (C-1‴), 150.8 (C-4′), 154.2 (C-1′), 158.2 (C-1″).

3-Phenoxyphenoxyethyl Selenocyanate (27). A solution of **31** (99 mg, 0.24 mmol) in anhydrous *N*,*N*-dimethylformamide (3.0 mL) was treated with potassium selenocyanate (125 mg, 1.29 mmol) according to the general procedure. The product was purified by column chromatography (silica gel) using a mixture of hexane-EtOAc (9:1) as eluent to give 45 mg (64% yield) of **27** as a colorless oil: R_f = 0.32 (hexane−EtOAc; 4:1); ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 3.41 (t, *J* = 6.0 Hz, 2H, H-1), 4.35 (t, *J* = 6.0 Hz, 2H, H-2), 6.57 (t, *J* = 2.3 Hz, 1H, aromatic proton), 6.64 (m, 2H, aromatic protons), 7.03 (dd, *J* = 8.7, 1.0 Hz, 2H, aromatic protons), 7.13 (t, *J* = 7.4 Hz, 1H, aromatic proton), 7.23 (t, *J* = 8.3 Hz, 1H, aromatic proton), 7.35 (dd, *J* = 8.3, 8.0 Hz, 2H, aromatic protons); ^{13}C NMR (125.77 MHz, $CDCl_3$) δ 28.0 (C-1), 66.5 (C-2), 101.6 (Se*C*N), 105.5 (C-2′), 109.2 (C-6′), 111.9 (C-4′), 119.2 (C-2″), 123.6 (C-4″), 129.8 (C-3″), 130.4 (C-5′), 156.7 (C-1″), 158.7 (C-1′), 159.1 (C-4′). HRMS (ESI) calcd. for $C_{15}H_{13}O_2NSeNa$ $[M+Na]^+$ 342.0009; found 342.0005.

4-Phenoxyphenoxyethyl Selenocyanate (28). Tosylate **33** (350.1 mg, 1.0 mmol) was treated

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according to the general procedure. The product was purified by column chromatography (silica gel) employing a mixture of hexane–EtOAc (9:1) as eluent to obtain 225.9 mg (71% yield) of **28** as a white solid: mp 54 ºC; R_f = 0.59 (hexane–EtOAc, 7:3); IR (film, cm^{-1}) 3041, 2868, 2152, 1588, 1501, 1486, 1212, 1008, 839, 755, 690; ^1H NMR (500.13 MHz, $CDCl_3$) δ 3.44 (t, *J* = 5.9 Hz, 2 H, H-1), 4.37 (t, *J* = 5.9 Hz, 2H, H-2), 6.90 (d, *J* = 9.2 Hz, 2H, H-2′), 6.95 (m, 2H, H-2″), 6.99 (d, *J* = 9.2 Hz, 2H, H-3′), 7.06 (tt, *J* = 7.4, 1.0 Hz, 1H, H-4″), 7.31 (dd, *J* = 8.7, 7.4, 2H, H-3″); ^{13}C NMR (125.77 MHz, $CDCl_3$) δ 28.1 (C-1), 67.1 (C-2), 101.1 (Se*C*N), 115.9 (C-2″), 117.9 (C-2′), 120.8 (C-3′), 122.7 (C-4″), 129.7 (C-3″), 151.2 (C-4′), 153.9 (C-1′), 158.1 (C-1″). HRMS (ESI) calcd. for $C_{15}H_{13}O_2NSeNa$ $[M+Na]^+$ 342.0009; found 341.9996. *Anal*. Calculated for ($C_{15}H_{13}O_2NSe$) C 56.61, H 4.12, N 4.40; found C 56.50, H 4.27, N 3.92.

4-Phenoxyphenoxyethyl carbamate (34). To a solution of 4-phenoxyphenyethanol (**32**; 500 mg, 2.2 mmol) in anhydrous methylene chloride (5.0 mL), cooled at −15 ºC, was added trichloroacetyl isocyanate (451 mg, 0.28 mL, 2.4 mmol) dropwise under an argon atmosphere. The reaction mixture was stirred at this temperature for 10 min. Then, methanol (1.0 mL) was added to get rid of the excess of trichloroacetyl isocyanate. Then, the mixture was concentrated under reduced pressure. The residue was dissolved in methanol (15 mL) and an aqueous 2.0 *M* solution of potassium carbonate (7.5 mL) was added. The resulting mixture was stirred at room temperature for additional 5 h. The reaction mixture was extracted with methylene chloride (2 × 25 mL) and the combined organic layers were washed with brine (5 × 50 mL), dried ($MgSO_4$), and the solvent was evaporated to give 584.0 mg (98% yield) of **34** as a white solid, which was used as such in the next step: mp 126 ºC; R_f = 0.27 (hexane–EtOAc, 3:2); IR (KBr, cm^{-1}) 1684, 1504, 1491, 1233, 1065, 839,754, 689; ^1H NMR (300.18 MHz, $CDCl_3$) δ 4.16 (m, 2H, H-1), 4.43 (m, 2H, H-2), 4.72 (br s, 2H, C(O)CH_2), 6.89 (d, *J* = 9.2 Hz, 2H, H-2′), 6.94 (m, 2H, H-2″), 6.98 (d, *J* = 9.2 Hz, 2H, H-3′), 7.05 (tt, *J* = 7.4, 1.0 Hz, 1H, H-4″), 7.30 (dd, *J* = 8.6, 7.4, 2H, H-3″); ^{13}C NMR (75.48 MHz, $CDCl_3$) δ 63.6 (C-1), 66.8 (C-2), 115.7 (C-2″), 117.7 (C-2′), 120.8 (C-3′), 122.5 (C-4″), 129.6 (C-3″), 1506.2 (C-4′), 156.7 (C-1′), 156.5 (O*C*(O)NH_2), 158.3 (C-1″).

4-Phenoxyphenoxyethyl Cyanate (29). To a solution of carbamate **34** (50 mg, 0.18 mmol) in anhydrous methylene chloride (16 mL), cooled at −78 ºC, was added anhydrous diisopropylethylamine (14.2 mg, 0.19 mL, 1.1 mmol) followed by addition of

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trifluoromethanesulfonic anhydride (77.0 mg, 45.8 μL, 0.27 mmol) under an argon atmosphere. The solution was stirred at −78 ºC for 1.5 h. Then, the reaction was quenched by addition of a 5% aqueous solution of sodium bicarbonate. The solution was allowed to warm to room temperature and the organic layer was separated, washed with a 5% aqueous solution of sodium bicarbonate, dried ($MgSO_4$), and the solvent was evaporated. The product was purified by column chromatography (silica gel) employing hexane−EtOAc (83:17) as eluent to afford 11.1 mg (24% yield) of **29** as a colorless oil: R_f = 0.62 (hexane−EtOAc, 3:2); IR (film, cm^{-1}) 2250, 1506, 1484, 1220, 871, 843, 688; ^{1}H NMR (300.18 MHz, $CDCl_3$) δ 4.33 (t, *J* = 5.9 Hz, 2 H, H-1), 4.74 (t, *J* = 5.9 Hz, 2H, H-2), 6.93 (d, *J* = 9.2 Hz, 2H, H-2′), 6.97 (m, 2H, H-2″), 7.02 (d, *J* = 9.2 Hz, 2H, H-3′), 7.08 (tt, *J* = 7.4, 0.9 Hz, 1H, H-4″), 7.33 (dd, *J* = 8.5, 7.4, 2H, H-3″); ^{13}C NMR (75.48 MHz, $CDCl_3$) δ 78.0 (C-1), 65.8 (C-2), 116.0 (C-2″), 117.9 (C-2′), 120.7 (C-3′), 122.8 (C-4″), 129.7 (C-3″), 151.4 (C-4′), 153.8 (C-1′), 158.1 (C-1″).

3-Phenoxyphenyl Prop-2-en-1-yl Ether (39). A solution of 3-phenoxyphenol (**38**; 1.5 g, 8.0 mmol) in DMSO (5.0 ml) was added KOH (904 mg, 16.1 mmol) and was stirred 5 min. Then was added allyl chloride (617 mg, 0.66 mL, 8.6 mmol) slowly. After stirred 10 min, the reaction mixture was extracted with methylene chloride (2 × 25 ml). The combined organic phases were washed with brine (5 × 50 mL), dried ($MgSO_4$), and the solvent was evaporated affording 1.61 g (90% yield) of **39** as colorless oil which was used as such in the next step: R_f = 0.71 (hexane−EtOAc, 4:1); ^{1}H-RMN (300.18 MHz, $CDCl_3$) δ 4.50 (dt, *J* = 5.3, 1.5 Hz, 2H, H-1), 5.28 (dq, *J* = 10.4, 1.4 Hz, 1H, H-$3_{cis\ \mathrm{to\ H\text{-}2}}$); 5.39 (dq, *J* = 17.3, 1.6 Hz, 1H, H-$3_{trans\ \mathrm{to\ H\text{-}2}}$), 6.03 (ddt, *J* = 17.3, 10.6, 5.3 Hz, 1H, H-2), 6.59 (m, 2H, aromatic protons), 6.66 (m, 1H, aromatic proton), 7.02 (m, 2H, aromatic protons), 7.11 (tt, *J* = 7.4, 1.1 Hz, 1H, H-4″), 7.21 (m, 1H, aromatic proton), 7.35 (dd, *J* = 8.5, 7.5 Hz, 2H, H-3″); ^{13}C NMR (75.48 MHz, $CDCl_3$) δ 68.9 (C-1), 105.5 (C-2′), 109.5 (C-6′), 111.1 (C-4′), 117.9 (C-3), 119.1 (C-2″), 123.4 (C-4″), 129.7 (C-3″), 130.1 (C-5′), 133.0 (C-2), 156.9 (C-1″), 158.4 (C-1′), 159.9 (C-4′).

2-Allyl-5-phenoxyphenol (40) and 2-Allyl-3-phenoxyphenol (41). A solution of **39** (1.80 g, 7.9 mmol) in *N,N*-dimethylaniline (6.0 mL) was stirred at 210 ºC for 10 h. The reaction mixture was allowed to cool to room temperature; then methylene chloride (20 mL) was added. The mixture was extracted with 10% aqueous solution of sodium hydroxide (2 × 15 mL). The aqueous phase

was acidified with a 10% aqueous solution of hydrochloric acid; then it was extracted with methylene chloride (2 × 25 mL). The combined organic phases were washed with brine (5 × 50 mL), dried ($MgSO_4$), and the solvent was evaporated. The product was purified by column chromatography (silica gel) employing hexane-EtOAc (47:3) as eluent to afford 718 mg (40% yield) of a non-separable mixture of compounds **40** and **41** in a 1.1:1:ratio. For spectroscopic characterization, an analytical sample of this mixture was purified by HPLC eluting with a mixture of acetonitrile-water (7:3) employing a semi-preparative column Beckmann Ultrasphere-ODS-2 (5 μM) as a yellow oils: Compound **40**: R_f = 0.58 (hexane−EtOAc, 4:1); ^{1}H RMN (500.13 MHz, $CDCl_3$) δ 3.38 (dt, J = 6.3, 1.5 Hz, 2H, H-1), 5.16 (m, 1H, H-$3_{trans\ \mathrm{to\ H\text{-}2}}$), 5.18 (dq, J = 9.4, 1.6 Hz, 1H, H-$3_{cis\ \mathrm{to\ H\text{-}2}}$), 6.01 (ddt, J = 16.8, 10.3, 6.4 Hz, 1H, H-2), 6.49 (d, J = 2.4 Hz, 1H, H-6′), 6.54 (dd, J = 8.2, 2.4 Hz, 1H, H-4′), 7.01 (dd, J = 8.7, 1.1 Hz, 1H, H-2″), 7.04 (d, J = 8.3 Hz, 1H, H-3′), 7.10 (tt, J = 7.4, 1.1 Hz, 1H, H-4″), 7.33 (dd, J = 8.6, 7.4 Hz, 2H, H-3″); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 34.6 (C-1), 106.6 (C-6′), 111.1 (C-4′), 116.5 (C-3), 119.0 (C-2″), 120.0 (C-2′), 123.3 (C-4″), 129.7 (C-3″), 131.1 (C-3′), 136.5 (C-2), 155.1 (C-5′), 157.0 (C-1′), 157.1 (C-1″). Compound **41**: R_f 0.52 (hexane−EtOAc, 4:1); ^{1}H-RMN (500.13 MHz, $CDCl_3$) δ 3.48 (dt, J = 6.2, 1.6 Hz, 2H, H-1), 5.10 (dq, J = 10.1, 1.6 Hz, 1H, H-$3_{cis\ \mathrm{to\ H\text{-}2}}$), 5.14 (dq, J = 17.1, 1.6 Hz, 1H, H-$3_{trans\ \mathrm{to\ H\text{-}2}}$), 5.96 (ddt, J = 17.2, 10.1, 6.2 Hz, 1H, H-2), 6.51 (dd, J = 8.2, 1.0 Hz, 1H, H-6′), 6.65 (dd, J = 8.1, 1.0 Hz, 1H, H-4′), 6.92-6.95 (m, 2H, aromatic protons), 7.04-7.09 (m, 2H, aromatic protons), 7.30 (dd, J = 8.5, 7.5 Hz, 2H, H-3″); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 28.1 (C-1), 111.5 (C-4′), 112.1 (C-6′), 117.5 (C-2′), 117.9 (C-2″), 122.7 (C-4″), 129.6 (C-3″), 127.8 (C-5′), 135.8 (C-2), 155.3 (C-1″), 155.8 (C-1′), 157.9 (C-3′).

(±)-6-Phenoxy-(2,3-dihydrobenzofuran-2-yl)methanol (42) and (±)-4-phenoxy-(2,3-dihydrobenzofuran-2-yl)methanol (43). A solution of a mixture of compounds **40** and **41** (375 mg, 1.7 mmol) in chlorofom (30 mL) was treated with 70% *m*-chloroperbenzoic acid (613 mg, 3.3 mmol). The solution was stirred at room temperature for 5 days. Then, the mixture was extracted with a 5% aqueous solution of sodium bicarbonate (3 × 30 mL) to remove the resulting *m*-chlorobenzoic acid. The organic phase was washed with water (2 × 20 ml), dried ($MgSO_4$), and the solvent was evaporated. The products were purified by column chromatography (silica gel) employing a mixture of hexane-EtOAc (87:13) as eluent to yield 53.1 mg of **42** (26% yield) and 49.3 mg of **43** (26% yield) as colorless oils: Compound **42**: R_f 0.12 (hexane−EtOAc, 4:1); ^{1}H

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RMN (500.13 MHz, $CDCl_3$) δ 2.99 (ddd, *J* = 15.3, 7.4, 1.0 Hz, 1H, H-3_a), 3.22 (dd, *J* = 15.5, 9.3 Hz, 1H, H-3_b), 3.75 (m, 1H, CH_aHOH), 3.86 (ddd, *J* = 12.0, 6.6, 3.2 Hz 1H, CH_bHOH), 4.96 (dddd, *J* = 9.5, 7.4, 6.3, 3.2 Hz 1H, H-2), 6.46 (d, *J* = 2.1 Hz, 1H, H-6), 6.50 (dd, *J* = 8.0, 2.2 Hz, 1H, H-5), 7.01 (m, 2H, aromatic protons), 7.09 (tt, *J* = 7.4, 1.0 Hz, 2H, H-2′), 7.32 (dd, *J* = 8.7, 7.4 Hz, 2H, H-3′); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 30.7 (C-3), 64.9 (CH_2OH), 84.2 (C-2), 101.2 (C-7), 111.1 (C-5), 118.8 (C-2″), 121.3 (C-4_a), 123.2 (C-4″), 125.2 (C-4), 129.7 (C-3′), 157.3 (C-1′), 157.6 (C-6), 160.4 (C-7_a). Compound **43**: R_f = 0.20 (hexane−EtOAc, 4:1); ^{1}H RMN (500.18 MHz, $CDCl_3$) δ 2.88 (dd, *J* = 15.9, 7.3 Hz, 1H, H-3_a), 3.12 (dd, *J* = 15.9, 9.5 Hz, 1H, H-3_b), 3.73 (dt, *J* = 11.4, 5.7 Hz, 1H, CH_aHOH), 3.83 (ddd, *J* = 12.0, 6.6, 3.2 Hz 1H, CH_bHOH), 4.93 (dddd, *J* = 9.5, 7.3, 6.4, 3.2 Hz 1H, H-2), 6.45 (dd, *J* = 8.2, 0.7 Hz, 1H, H-7), 6.59 (d, *J* = 7.6 Hz, 1H, H-5), 6.99 (dd, *J* = 8.7, 1.1 Hz, 2H, H-2′), 7.06-7.11 (m, 2H, aromatic protons), 7.33 (dd, *J* = 8.6, 7.4 Hz, 2H, H-3″); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 29.1 (C-3), 64.9 (CH_2OH), 83.7 (C-2), 105.0 (C-7), 111.1 (C-5), 117.1 (C-4_a), 118.3 (C-2′), 123.1 (C-4″), 129.3 (C-6), 129.7 (C-3′), 153.5 (C-4), 156.7 (C-1′), 161.6 (C-7a).

(±)-6-Phenoxy-(2,3-dihydrobenzofuran-2-yl)methyl 4-Toluenesulfonate (44). A solution of alcohol **42** (70.2 mg, 0.29 mmol) in pyridine (3 mL) was treated with 4-toluenesulfonyl chloride (166 mg, 0.87 mmol) as described for the preparation of **31**. The product was purified by column chromatography (silica gel) employing a mixture of hexane−EtOAc (9:1) as eluent to afford 59.8 mg of **44** (52% yield) as a colorless oil. R_f 0.27 (hexane−EtOAc, 4:1); ^{1}H RMN (300.18 MHz, $CDCl_3$) δ 2.43 (s, 3H, PhCH_3), 2.95 (dd, *J* = 15.6, 6.9 Hz, 1H, H-3_a), 3.25 (dd, *J* = 16.1, 9.7 Hz, 1H, H-3_b), 4.18 (d, *J* = 5.2 Hz, 2H, CH_2OTs), 4.98 (ddt, *J* = 9.9, 6.9, 5.0 Hz 1H, H-2), 6.33 (d, *J* = 2.2 Hz, 1H, H-7), 6.49 (dd, *J* = 8.1, 2.2 Hz, 1H, H-5), 6.98 (m, 2H, aromatic protons), 7.04 (dt, *J* = 8.1, 1.0 Hz, 1H, H-4), 7.09 (tt, *J* = 7.4, 0.9 Hz, 1H, H-4′), 7.31 (d, *J* = 8.6 Hz, 2H, H-2″) 7.32 (dd, *J* = 8.7, 7.4 Hz, 2H, H-3′), 7.78 (d, *J* = 8.4 Hz, 2H, H-2″); ^{13}C NMR (75.48 MHz, $CDCl_3$) δ 21.6 (PhCH_3), 29.7 (C-3), 70.4 (CH_2OTs), 80.1 (C-2), 101.2 (C-7), 111.3 (C-6), 118.8 (C-2′), 120.0 (C-4_a), 123.3 (C-4′), 125.2 (C-4), 128.0 (C-2″), 129.7 (C-3′), 129.9 (C-3″), 132.7 (C-4″), 145.0 (C-1″), 157.2 (C-1′), 157.8 (C-6), 160.1 (C-7_a).

(±)-4-phenoxy-(2,3-dihydrobenzofuran-2-yl)methyl 4-Toluenesulfonate (45). To a solution of **43** (55.3 mg, 0.23 mmol) in pyridine (3.0 mL) was added 4-toluenesulfonyl chloride (166 mg,

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0.87 mmol) as described for the preparation of **31**. The product was purified by column chromatography (silica gel) employing a mixture of hexane−EtOAc (9:1) as eluent to afford 54.4 mg of **45** (60% yield) as a colorless oil. R_f = 0.35 (hexane−EtOAc, 4:1); ^{1}H-RMN (300.18 MHz, $CDCl_3$) δ 2.44 (s, 3H, PhC*H*$_3$), 2.83 (dd, *J* = 16.1, 6.7 Hz, 1H, H-3$_a$), 3.14 (dd, *J* = 16.1, 9.7 Hz, 1H, H-3$_b$), 4.16 (m, 2H, C*H*$_2$OTs), 4.95 (ddt, *J* = 9.9, 6.7, 5.0 Hz 1H, H-2), 6.43 (d, *J* = 8.2 Hz, 1H, H-7), 6.49 (d, *J* = 8.0 Hz, 1H, H-5), 6.97 (m, 2H, H-2′), 7.07 (m, 2H, aromatic protons), 7.32 (d, *J* = 8.6 Hz, 2H, H-3″), 7.32 (m, 2H, H-3′), 7.77 (d, *J* = 8.3 Hz, 2H, H-2″); ^{13}C NMR (75.48 MHz, $CDCl_3$) δ 21.7 (Ph*C*H$_3$), 29.6 (C-3), 70.4 (*C*H$_2$OTs), 79.7 (C-2), 105.0 (C-7), 111.3 (C-5), 115.8 (C-4$_a$), 118.3 (C-2′), 123.3 (C-4′), 128.0 (C-2″), 129.4 (C-6), 129.7 (C-3′), 129.9 (C-3″), 132.7 (C-4″), 145.0 (C-1″), 153.5 (C-4), 156.5 (C-1′), 161.8 (C-7a).

(±)-5-Phenoxy-2-(selenocyanatomethyl)-2,3-dihydrobenzofuran (35). Tosylate **26** (57.3 mg, 0.14 mmol) was treated with potassium selenocyanate (22.8 mg, 0.16 mmol) and 18-crown-6 (0.3 mg) following the general procedure. The product was purified by HPLC eluting with acetonitrile−water (4:1) employing a semi-preparative column Beckmann Ultrasphere-ODS-2 (5 μM) to yield 41.1 mg (86% yield) of 35 as a colorless oil: R_f = 0.32 (hexane−EtOAc, 4:1); ^{1}H-NMR ($CDCl_3$, 300.18 MHz) δ 3.06 (dd, *J* = 16.1, 6.6 Hz, 1H, H-3$_a$), 3.39 (m*AB*, 2H, C*H*$_2$SeCN), 3.45 (dd, *J* = 16.3, 9.3 Hz, 1H, H-3$_b$), 5.14 (ddt, *J* = 12.2, 9.0, 6.2 Hz, 1H, H-2), 6.75 (d, *J* = 8.6 Hz, 1H, H-7), 6.83 (m, 1H, aromatic proton), 6.88 (m, 1H, aromatic proton), 6.94 (m, 2H, aromatic protons), 7.05 (tt, *J* = 7.4, 1.0 Hz, 1H, H-4′), 7.30 (dd, *J* = 8.6, 7.4 Hz, 2H, H-3′); ^{13}C NMR (75.48 MHz, $CDCl_3$) δ 33.7 (C-3), 35.5 (*C*H$_2$SCN), 81.0 (C-2), 101.1 (Se*C*N), 110.1 (C-7), 116.9 (C-6), 117.7 (C-2′), 119.9 (C-4), 122.6 (C-4′), 126.8 (C-4$_a$), 129.6 (C-3′), 151.0 (C-5), 154.7 (C-7$_a$), 158.4 (C-1′). HRMS (ESI) calcd. for $C_{16}H_{13}O_2NSeNa$ $[M+Na]^+$ 354.0009; found 354.0012.

(±)-6-Phenoxy-2-(selenocyanatomethyl)-2,3-dihydrobenzofuran (36). A mixture of **46** (40.2 mg, 0.1 mmol), potassium thiocyanate (16.4 mg, 0.11 mmol) and 18-crown-6 (0.3 mg) in tetrahydrofuran (3.0 mL) was treated according to the general procedure. The product was purified by HPLC eluting with acetonitrile−water (4:1) employing a semi-preparative column Beckmann Ultrasphere-ODS-2 (5 μM) to yield 12.8 mg (38% yield) of **37** as a colorless oil: R_f = 0.24 (hexane−EtOAc, 4:1); ^{1}H-RMN (500.13 MHz, $CDCl_3$) δ 3.04 (ddd, *J* = 15.7, 6.6, 0.7 Hz,

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1H, H-3_a), 3.37 (dd, J = 12.5, 6.5 Hz, 1H, CH_aHSeCN), 3.40 (dd, J = 12.5, 5.4 Hz, 1H, CH_bHSeCN), 3.44 (ddd, J = 15.6, 9.2, 0.6 Hz, 1H, H-3_b), 5.14 (ddt, J = 9.1, 6.5, 5.4 Hz 1H, H-2), 6.46 (d, J = 2.2 Hz, 1H, H-7), 6.54 (dd, J = 8.1, 2.2 Hz, 1H, H-5), 7.01 (m, 2H, H-2′), 7.10 (m, 2H, H-4, H-4′), 7.33 (dd, J = 8.7, 7.4 Hz, 2H, H-3′); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 33.7 (CH_2SeCN), 34.8 (C-3), 81.6 (C-2), 101.0 (Se*C*N), 101.2 (C-7), 111.6 (C-6), 119.0 (C-2′), 119.9 (C-4_a), 123.4 (C-4′), 125.3 (C-4), 129.7 (C-3′), 157.0 (C-1′), 158.2 (C-6), 159.7 (C-7_a). HRMS (ESI) calcd. for $C_{16}H_{13}O_2NSeNa$ $[M+Na]^+$ 354.0009; found 353.9989.

(±)-4-Phenoxy-2-(selenocyanatomethyl)-2,3-dihydrobenzofuran (37). A mixture of compound **45** (30.8 mg, 0.078 mmol), potassium thiocyanate (12.4 mg, 0.086 mmol) and 18-crown-6 (0.3 mg) in tetrahydrofuran (3.0 mL) was treated following the general procedure. The product was purified by HPLC eluting with acetonitrile−water (4:1) employing a semi-preparative column Beckmann Ultrasphere-ODS-2 (5 μM) to give 14.4 mg (75% yield) of **37** as a colorless oil: R_f = 0.37 (hexane-EtOAc, 4:1); ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 2.93 (dd, J = 16.2, 6.4 Hz, 1H, H-3_a), 3.34 (dd, J = 16.2, 9.0 Hz, 1H, H-3_b), 3.35 (dd, J = 12.1, 6.8 Hz, 1H, CH_aHSeCN), 3.38 (dd, J = 12.5, 5.2 Hz, 1H, CH_bHSeCN), 5.13 (ddt, J = 9.2, 6.4, 5.2 Hz 1H, H-2), 6.48 (dd, J = 8.3, 0.6 Hz, 1H, H-7), 6.59 (dd, J = 8.0, 0.5 Hz, 1H, H-5), 6.99 (m, 2H, H-2′), 7.11 (m, 2H, aromatic protons), 7.34 (dd, J = 8.6, 7.4 Hz, 2H, H-3′); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 33.2 (CH_2SeCN), 33.8 (C-3), 81.1 (C-2), 101.0 (Se*C*N), 105.1 (C-7), 111.4 (C-5), 115.7 (C-4_a), 118.5 (C-2′), 123.4 (C-4′), 129.7 (C-6), 129.8 (C-3′), 153.7 (C-4), 156.5 (C-1′), 160.4 (C-7a). HRMS (ESI) calcd. for $C_{16}H_{13}O_2NSeNa$ $[M+Na]^+$ 354.0009; found 353.9996.

4-Iodophenoxyethyl Selenocyanate (46). Tosylate **56** was treated according to the general procedure to produce **46** in 64% yield as a white solid: mp 106 ºC; R_f = 0.27 (hexane−EtOAc, 4:1); ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 3.42 (t, J = 5.9 Hz, 2H, H-1), 4.35 (t, J = 6.0 Hz, 2H, H-2), 6.70 (d, J = 8.9 Hz, 2H, H-2′), 7.58 (d, J = 9.0 Hz, 2H, H-3′); ^{13}C NMR (125.77 MHz, $CDCl_3$) δ 27.8 (C-1), 66.6 (C-2), 84.1 (C-4′), 100.9 (Se*C*N), 117.0 (C-2′), 138.5 (C-3′), 157.7 (C-1′). HRMS (ESI) calcd. for $C_9H_8ONISeNa$ $[M+Na]^+$ 375.8713; found 375.8716.

3-Iodophenoxyethyl Selenocyanate (47). Tosylate **57** was treated following the general method to give **47** in 69% yield as a colorless oil: R_f = 0.30 (hexane−EtOAc, 4:1); IR (film, cm^{-1}) 3058,

2926, 2869, 2151, 1581, 1575, 1472, 1459, 1222, 766, 679; ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 3.41 (t, *J* = 5.9 Hz, 2H, H-1), 4.36 (t, *J* = 6.0 Hz, 2H, H-2), 6.88 (ddd, *J* = 8.3, 2.5, 0.8 Hz, 1H, H-6′), 7.02 (t, *J* = 8.1 Hz, 1H, H-5′), 7.27 (dd, *J* = 2.5, 1.6 Hz, 1H, H-4′), 7.34 (ddd, *J* = 7.8, 1.5, 0.9 Hz, 1H, H-2′); ^{13}C NMR (125.77 MHz, $CDCl_3$) δ 27.8 (C-1), 66.6 (C-2), 94.4 (C-3′), 100.9 (Se*C*N), 114.2 (C-6′), 123.9 (C-2′), 131.0 (C-4′, C-5′), 158.3 (C-1′). HRMS (ESI) calcd. for $C_9H_8ONISeNa$ $[M+Na]^+$ 375.8713; found 375.9569. *Anal*. Calculated for (C_9H_8ONISe) C 30.71, H 2.29, N 3.98; found C 31.25, H 2.78, N 3.29.

2,4-Dichlorophenoxyethyl Selenocyanate (48). Tosylate **58** (572.2 mg, 1.6 mmol) was treated with potassium selenocyanate (251.5 mg, 1.7 mmol) and 18-crown-6 (4.2 mg) following the general procedure. The product was purified by column chromatography (silica gel) eluting with hexane−EtOAc (97:3) to give 373.8 mg (84% yield) of **48** as a white solid: mp 70 ºC; R_f = 0.26 (hexane−EtOAc, 4:1); ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 3.48 (t, *J* = 6.0 Hz, 2H, H-1), 4.42 (t, *J* = 6.1 Hz, 2H, H-2), 6.88 (d, *J* = 8.8 Hz, 2H, H-6′), 7.21 (dd, *J* = 8.8, 2.6 Hz, 2H, H-5′), 7.39 (d, *J* = 2.5 Hz, 2H, H-3′); ^{13}C NMR (75.48 MHz, $CDCl_3$) δ 27.7 (C-1), 68.0 (C-2), 101.0 (Se*C*N), 114.9 (C-6′), 124.3 (C-2′), 127.2 (C-4′), 127.7 (C-5′), 130.3 (C-3′), 152.0 (C-1′). HRMS (ESI) calcd. for $C_9H_8ONISeNa$ $[M+Na]^+$ 317.8968; found 317.8971. *Anal*. Calculated for ($C_9H_7ONCl_2Se$) C 36.64, H 2.39, N 4.75; found C 36.57, H 2.34, N 4.88.

4-(3-Fluorophenoxy)phenoxyethyl Selenocyanate (49). Compound **59** (452.1 mg, 1.1 mmol); was treated with potassium selenocyanate (216.2 mg, 1.5 mmol) and 18-crown-6 (3.6 mg) following the general procedure. The residue was purified by column chromatography (silica gel) eluting with hexane−EtOAc (93:7) to yield 244.6 mg (53% yield) of **49** as a colorless oil: R_f = 0.27 (hexane−EtOAc, 4:1); ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 3.44 (t, *J* = 6.0 Hz, 2H, H-1), 4.39 (t, *J* = 6.0 Hz, 2H, H-2), 6.63 (dt, *J* = 10.4, 2.4 Hz, 1 H, H-2′), 6.72 (m, 1H, H-5″), 6.76 (ddd, *J* = 8.3, 2.4, 0.8 Hz, 1H, H-6″), 6.92 (d, *J* = 9.1 Hz, 2H, H-2′), 7.01 (d, *J* = 9.1 Hz, 2H, H-3′), 7.24 (dt, *J* = 8.3, 6.7 Hz, 1 H, H-4″); ^{13}C NMR (125.76 MHz, $CDCl_3$) δ 28.1 (C-1), 67.0 (C-2), 101.1 (Se*C*N), 105.1 (d, *J* = 24.9 Hz, C-2″), 109.3 (d, *J* = 21.3 Hz, C-4″), 113.0 (d, *J* = 3.0 Hz, C-6″), 116.0 (C-2′), 121.3 (C-3′), 130.4 (d, *J* = 9.8 Hz, C-5″), 150.2 (C-4′), 154.5 (C-1′), 159.7 (d, *J* = 10.6 Hz, C-1″), 163.5 (d, *J* = 246.5 Hz, C-3″); ^{19}F NMR (470.59 MHz, $CDCl_3$) δ −111.03 ppm. HRMS (ESI) calcd. for $C_{15}H_{12}O_2NFSeNa$ $[M+Na]^+$ 359.9915; found 359.9898.

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4-(4-Fluorophenoxy)phenoxyethyl Selenocyanate (50). Tosylate **60** (475.2 mg, 1.2 mmol) was treated with potassium selenocyanate (226.7 mg, 1.6 mmol) and 18-crown-6 (4.2 mg) following the general procedure. The product was purified by column chromatography (silica gel) eluting with hexane–EtOAc (22:3) to yield 367.9 mg (76% yield) of **50** as a colorless oil: R_f = 0.24 (hexane–EtOAc, 4:1); IR (film, cm^{-1}) 3073, 2869, 2152, 1490, 1198, 1006, 827, 764, 510; ^1H NMR (500.13 MHz, $CDCl_3$) δ 3.43 (t, *J* = 6.0 Hz, 2H, H-1), 4.37 (t, *J* = 6.0 Hz, 2H, H-2), 6.89 (d, *J* = 9.3 Hz, 2H, H-3′), 6.92 (m, 2H, H-3″), 6.95 (d, *J* = 9.3 Hz, 2H, H-2′), 7.00 (dd, *J* = 9.3, 8.1 Hz, 2H, H-2"); ^{13}C NMR (125.76, $CDCl_3$) δ 28.1 (C-1), 67.1 (C-2), 101.1 (Se*C*N), 115.9 (C-2′), 116.2 (d, *J* = 23.2 Hz, C-3″), 119.4 (d, *J* = 8.3 Hz, C-2″), 120.2 (C-3′), 151.7 (C-4′), 153.8 (d, *J* = 2.4 Hz, C-1″), 153.9 (C-1′), 158.5 (d, *J* = 241.1 Hz, C-4″); ^{19}F NMR (470.59 MHz, $CDCl_3$) δ −120.84 ppm. HRMS (ESI) calcd. for $C_{15}H_{12}O_2NFSeNa$ $[M+Na]^+$ 359.9915; found 359.9897. *Anal*. Calculated for ($C_{15}H_{12}O_2NFSe$) C 53.58, H 3.60, N 4.17; found C 53.56, H 3.74, N 3.81.

3-(2-Fluorophenoxy)phenoxyethyl Selenocyanate (51). Tosylate **61** (309.8 mg, 0.77 mmol) was treated with potassium selenocyanate (122.0 mg, 0.85 mmol) and 18-crown-6 (2.0 mg) following the general procedure. The product was purified by column chromatography (silica gel) eluting with hexane–EtOAc (97:3) to give 189.3 mg (73% yield) of **51** as a colorless oil: R_f = 0.39 (hexane–EtOAc, 4:1); IR (film, cm^{-1}) 3067, 2874, 2152, 1598, 1485, 1251, 1133, 757, 684; ^1H NMR (500.13 MHz, $CDCl_3$) δ 3.41 (t, *J* = 6.0 Hz, 2H, H-1), 4.35 (t, *J* = 6.0 Hz, 2H, H-2), 6.56 (t, *J* = 2.4 Hz, 1H, H-3′), 6.59 (dd, *J* = 8.3, 2.4 Hz, 1H, H-4′), 6.64 (ddd, *J* = 8.3, 2.4, 0.8 Hz, 1H, H-6′), 7.07-7.20 (m, 4H, aromatic protons), 7.22 (t, *J* = 8.3 Hz, 1H, H- H-5′); ^{13}C NMR (125.77 MHz, $CDCl_3$) δ 27.9 (C-1), 66.5 (C-2), 101.0 (Se*C*N), 104.1 (C-2′), 109.1 (C-6′), 110.3 (C-4′), 117.2 (d, *J* = 18.2 Hz, C-3″), 122.3 (d, *J* = 1.0 Hz, C-5″), 124.8 (d, *J* = 3.9 Hz, C-6″), 125.2 (d, *J* = 7.0 Hz, C-4″), 130.4 (C-5′), 143.2 (d, *J* = 11.5 Hz, C-1″), 154.4 (d, *J* = 249.2 Hz, C-2″), 158.7 (C-3′), 159.0 (C-1′); ^{19}F NMR (470.59 MHz, $CDCl_3$) δ −130.70 ppm. HRMS (ESI) calcd. for $C_{15}H_{12}O_2NFSeNa$ $[M+Na]^+$ 359.9915; found 359.9918. *Anal*. Calculated for ($C_{15}H_{12}O_2NFSe$) C 53.58, H 3.60, N 4.17; found C 53.23, H 3.79, N 3.71.

3-(3-Fluorophenoxy)phenoxyethyl Selenocyanate (52). Tosylate **62** (687.5 mg, 1.7 mmol); was treated with potassium selenocyanate (328.0 mg, 2.3 mmol) and 18-crown-6 (5.5 mg)

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following the general procedure. The product was purified by column chromatography (silica gel) eluting with hexane–EtOAc (91:9) to yield 379.2 mg (69% yield) of **52** as a colorless oil: R_f = 0.30 (hexane–EtOAc, 4:1); ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 3.42 (t, *J* = 6.0 Hz, 2H, H-1), 4.37 (t, *J* = 6.0 Hz, 2H, H-2), 6.60 (t, *J* = 2.3 Hz, 1H, H-3′), 6.67 (ddd, *J* = 8.1, 2.2, 0.6 Hz, 1H, H-4′), 6.67-6.73 (m, 2H, aromatic protons), 6.79-6.83 (m, 2H, aromatic protons), 7.27 (t, *J* = 8.4 Hz, 1H, H-5′), 7.28 (m, 1H, aromatic proton); ^{13}C NMR (125.77 MHz, $CDCl_3$) δ 27.9 (C-1), 66.6 (C-2), 101.0 (Se*C*N), 106.2 (C-2′), 106.4 (d, *J* = 24.7 Hz, C-2″), 110.0 (C-6′), 110.2 (d, *J* = 21.2 Hz, C-4″), 112.5 (C-4′), 114.3 (d, *J* = 3.2 Hz, C-6″), 130.5 (d, *J* = 9.5 Hz, C-5″), 130.6 (C-5′), 157.7 (C-3′), 158.3 (d, *J* = 10.6 Hz, C-1″), 159.1 C-1′), 163.5 (d, *J* = 246.9 Hz, C-3″); ^{19}F NMR (470.59 MHz, $CDCl_3$) δ −110.81 ppm. HRMS (ESI) calcd. for $C_{15}H_{12}O_2NFSeNa$ $[M+Na]^+$ 359.9915; found 359.9896.

3-(4-Fluorphenoxy)phenoxyethyl Selenocyanate (53). Tosylate **63** (559.0 mg, 1.4 mmol); was treated with potassium selenocyanate (267.7 mg, 1.9 mmol) and 18-crown-6 (4.4 mg) following the general procedure. The product was purified by column chromatography (silica gel) eluting with hexane–EtOAc (91:9) to yield 379.2 mg (67% yield) of **53** as a colorless oil: R_f = 0.25 (hexane–EtOAc, 4:1); IR (film, cm^{-1}) 3074, 2875, 2153, 1588, 1502, 1484, 1193, 1134, 833, 772, 685, 501; ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 3.41 (t, *J* = 6.0 Hz, 2H, H-1), 4.35 (t, *J* = 6.0 Hz, 2H, H-2), 6.52 (t, *J* = 2.2 Hz, 1H, H-3′), 6.59 (ddd, *J* = 8.2, 2.3, 0.9 Hz, 1H, H-4′), 6.64 (ddd, *J* = 8.3, 2.4, 0.8 Hz, 1H, H-6′), 6.99 (dd, *J* = 9.3, 4.7 Hz, 2H, aromatic protons), 7.04 (dd, *J* = 9.0, 8.4 Hz, 2H, aromatic protons), 7.23 (t, *J* = 8.2 Hz, 1H, H-5′); ^{13}C NMR (125.77 MHz, $CDCl_3$) δ 27.9 (C-1), 66.5 (C-2), 101.0 (Se*C*N), 105.0 (C-2′), 109.0 (C-6′), 111.2 (C-4′), 116.4 (d, *J* = 23.2 Hz, C-3″), 120.9 (d, *J* = 8.1 Hz, C-2″), 130.4 (C-5′), 152.3 (d, *J* = 2.5 Hz, C-1″), 159.0 (d, *J* = 242.2 Hz, C-4″), 159.08 (C-3′), 159.10 C-1′); ^{19}F NMR (470.59 MHz, $CDCl_3$) δ −119.50 ppm. HRMS (ESI) calcd. for $C_{15}H_{12}O_2NFSeNa$ $[M+Na]^+$ 359.9915; found 359.9903. *Anal*. Calculated for ($C_{15}H_{12}O_2NFSe$) C 53.58, H 3.60, N 4.17; found C 53.42, H 3.90, N 3.94.

3-(4-Chlorophenoxy)phenoxyethyl Selenocyanate (54). Tosylate **64** (441.9 mg, 1.06 mmol) was treated with potassium selenocyanate (167.0 mg, mmol) and 18-crown-6 (2.8 mg) following the general procedure. The product was purified by column chromatography (silica gel) eluting with hexane–EtOAc (93:7) followed by HPLC separation employing a semi-preparative column

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Beckmann Ultrasphere-ODS-2 (5 µM) as eluting with methanolwater (9:1) at a flow rate of 3.0 mL/min to give 269 mg (72% yield) of **54** as a colorless oil: R_f 0.42 (hexane−EtOAc, 4:1); ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 3.41 (t, *J* = 6.0 Hz, 2H, H-1), 4.36 (t, *J* = 5.9 Hz, 2H, H-2), 6.55 (t, *J* = 2.3 Hz, 1H, H-2′), 6.62 (ddd, *J* = 8.2, 2.3, 0.8 Hz, 1H, H-4′), 6.67 (dd, *J* = 8.3, 2.4,07 Hz, 1H, H-6′), 6.96 (d, *J* = 9.0 Hz, 2H, H-3″), 7.25 (t, *J* = 8.2 Hz, 1H, H-5′), 7.30 (d, *J* = 9.0 Hz, 2H, H-2″); ^{13}C NMR (125.77 MHz, $CDCl_3$) δ 27.9 (C-1), 66.6 (C-2), 101.0 (Se*C*N), 105.6 (C-2′), 109.5 (C-6′), 111.9 (C-4′), 120.4 (C-2″), 128.6 (C-4″), 129.8 (C-5′), 130.5 (C-3″), 155.4 (C-1″), 158.3 (C-1′), 159.1 (C-3′). HRMS (ESI) calcd. for $C_{15}H_{12}O_2NClSeNa$ $[M+Na]^+$ 375.9619; found 375.9594.

3-(3-Pyridyl)oxyphenoxyethyl Selenocyanate (55). Tosylate **65** (319.5 mg, 0.83mmol) was treated with potassium selenocyanate (131 mg, 0.91 mmol) and 18-crown-6 (2.2 mg) according to the general procedure. The product was purified by column chromatography (silica gel) eluting with hexane−EtOAc (93:7) to give 169 mg (64% yield) of **55** as a yellowish oil: R_f 0.36 (hexane−EtOAc, 1:1); IR (film, cm^{-1}) 3061, 2921, 2151, 1572, 1473, 1421, 1220, 1135, 708, 687; ^{1}H NMR (500.13 MHz, $CDCl_3$) δ 3.41 (t, *J* = 6.0 Hz, 2H, H-1), 4.63 (t, *J* = 6.0 Hz, 2H, H-2), 6.59 (t, *J* = 2.3 Hz, 1H, H-2′), 6.64 (ddd, *J* = 8.1, 2.3, 0.8 Hz, 1H, H-4′), 6.71 (ddd, *J* = 8.3, 2.4, 0.8 Hz, 1H, H-6′), 7.25-7.32 (m, 3H, aromatic protons), 8.38 (dd, *J* = 4.6, 1.6 Hz, 1H, H-4″), 8.41 (dd, *J* = 2.6, 0.7 Hz, 1H, H-2″); ^{13}C NMR (125.77 MHz, $CDCl_3$) δ 27.8 (C-1), 66.5 (C-2), 101.1 (Se*C*N), 105.7 (C-2′), 109.9 (C-6′), 111.8 (C-4′), 124.1 (C-6″), 125.8 (C-5″), 130.6 (C-5′), 141.5 (C-2″), 144.6 (C-4″), 153.4 (C-1″), 157.6 (C-1′), 159.1 (C-3′). HRMS (ESI) calcd. for $C_{14}H_{14}O_2N_2Se$ $[M+H]^+$ 321.0142; found 321.0138. *Anal.* Calculated for ($C_{14}H_{12}O_2N_2Se$) C 52.68, H 3.79, N 8.78; found C 52.35, H 3.79, N 8.63.

Drug Screening

***T. cruzi* amastigote assays**

These experiments were done as reported using tdTomato labeled trypomastigotes[68] with the modifications described by Recher et al., 2013.[69] Breifly, gamma-irradiated (2,000 Rads) Vero cells (3.4×10^4 cells/well) were seeded in 96 well plates (black, clear bottom plates from Greiner Bio-One) in 100 µL RPMI media (Sigma) with 10% FBS. Plates were incubated overnight at 35 ^{o}C and 7% CO_2. After overnight incubation, Vero cells were challenged with 3.4×10^5

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trypomastigotes/well (CL strain overexpressing a tdTomato red fluorescent protein) in 50 µL volume and incubated for 5 h at 35 °C and 7% CO_2. After infection, cells were washed once with Hanks solution (150 µL/well) to eliminate any extracellular parasites and compounds were added in serial dilutions in RPMI media in 150 µL volumes. Each dilution was tested in quadruplicate. Each plate also contained controls with host cells and no parasites (for background check), and controls with parasites and no drugs (positive control). Drugs were tested on *T. cruzi* at 1.56 µM, 3.125 µM, 6.25 µM, 12.5 µM, 25 µM. For each set of experiments, benznidazole was also used as a positive control 0.39 µM, 0.78 µM, 1.56 µM, 3.125 µM, and 6.25 µM. After drug addition, plates were incubated at 35 ºC and 7% CO_2. At day 3 post-infection, plates were assayed for fluorescence. IC_{50} values were determined by non-linear regression analysis using SigmaPlot. There was no evident cytotoxicity on the host cells (visual assay) with any of the drugs tested at concentrations as high as 25 µM. ED_{50} values were determined by non-linear regression analysis using SigmaPlot.

***T. gondii* tachyzoites assays**

Experiments on *T. gondii* tachyzoites were carried out as described previously[70] using *T. gondii* tachyzoites expressing red fluorescent protein[71] with the modifications described by Recher et al., 2013.[69] Plates were read with covered lids, and both excitation (544 nm) and emission (590 nm) were read from the bottom.

Cytotoxicity for Vero cells.

The cytotoxicity was tested using the Alamar Blue™ assay as described by Recher et al., 2013.[69]

Single-crystal XRD measurement, refinement and searching in the CSD

WC-9 single crystals were obtained through slow evaporation crystallization at RT from an ethyl acetate solution of the pure compound. **28** single crystals were obtained from a hexane–EtOAc (47:3) solution at low temperature (c.a. –20 ºC) in a closed vessel. For both compounds suitable crystals were selected and structurally characterized by single-crystal X-ray diffraction (sXRD) at RT using Mo radiation with an Oxford Gemini E diffractometer with a CCD detector. The measurements were collected with CrysAlis Pro computer program[72] and using Olex2 program[73]

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their structures were solved with ShelXD[74] structure solution program using Dual Space and refined with the ShelXL[75] refinement package using Least Squares minimization. Crystal data, data collection and structure refinement details for **WC-9** and **28** are summarized in Table 2. All H atoms were placed in idealized positions and refined in riding modes such that Uiso(H) = 0.06Ueq(parent). Mercury program was used to create sXRD graphics and analyse the molecular geometry, intermolecular interactions and packing. Statistics studies based on XRD results deposited on the Cambridge Structural Database (CSD, Version 5.34)[76] were performed through the Mogul Geometry Check menu.[63]

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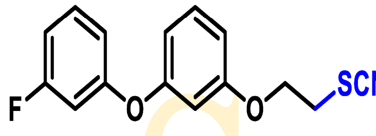
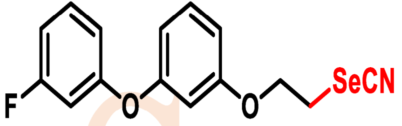


T. cruzi ED_{50} = 5.38 μM
Cytotoxicity ED_{50} > 50 μM
Selectivity Index > 10

Sulfur



Compound 52
T. cruzi ED_{50} = 0.075 μM
Cytotoxicity ED_{50} >> 62.5 μM
Selectivity Index >> 833

Selenium

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