Synthetic Development of New 3-(4-Arylmethylamino)butyl-5-arylidene-rhodanines under Microwave Irradiation and Their Effects on Tumor Cell Lines and against Protein Kinases

Camille Déliko Dago, Christelle Ambeu, Wacothon-Karime Coulibaly, Yves-Alain Békro, Janat Mamyrbékova, Audrey Defontaine, Blandine Baratte, Stéphane Bach, Sandrine Ruchaud, Remy Le Guével, et al.

To cite this version:
Camille Déliko Dago, Christelle Ambeu, Wacothon-Karime Coulibaly, Yves-Alain Békro, Janat Mamyrbékova, et al.. Synthetic Development of New 3-(4-Arylmethylamino)butyl-5-arylidene-rhodanines under Microwave Irradiation and Their Effects on Tumor Cell Lines and against Protein Kinases. Molecules, MDPI, 2015, 20 (7), pp.12412-12435. <10.3390/molecules200712412>. <hal-01187427>
Synthetic Development of New 3-(4-Arylmethylamino)butyl-5-arylidene-rhodanines under Microwave Irradiation and Their Effects on Tumor Cell Lines and against Protein Kinases

Camille Déliko Dago 1,2, Christelle N’ta Ambeu 1,2, Wacothon-Karime Coulibaly 3, Yves-Alain Békro 2, Janat Mamyrbéko 2, Audrey Defontaine 4, Blandine Baratte 4, Stéphane Bach 4, Sandrine Ruchaud 4, Rémy Le Guével 5, Myriam Ravache 5, Anne Corlu 5 and Jean-Pierre Bazureau 1,*

1 Université de Rennes 1, Institut des Sciences Chimiques de Rennes (ISCR), UMR CNRS 6226, Groupe ICMV, Bât. 10A, Campus de Beaulieu, 263 Avenue du Général Leclerc, CS 74205, 35042 Rennes Cedex, France; E-Mails: deliko.dago@univ-rennes1.fr (C.D.D.); christelle.ambeu@univ-rennes1.fr (C.N.A.)

2 Laboratoire de Chimie Bio-Organique et de Substances Naturelles (LCBOSN), Université Nangui Abrogoua, Abidjan 02, BP 802, Cote d'Ivoire; E-Mails: bekro2001@yahoo.fr (Y.-A.B.); kojanova1926@hotmail.fr (J.M.)

3 UFR des Sciences Biologiques, Université de Péléforo Gon Coulibaly, Korhogo, BP 1328, Cote d'Ivoire; E-Mail: wacothon@yahoo.fr

4 Station Biologique de Roscoff, USR 3151, CNRS-UPMC, Kinase Inhibitory Specialized Screening facility, KISSf, Place George Teissier, BP 74, 29682 Roscoff, France; E-Mails: audrey.defontaine@sb-roscoff.fr (A.D.); blandine.baratte@sb-roscoff.fr (B.B.); stephane.bach@sb-roscoff.fr (S.B.); sandrine.ruchaud@sb-roscoff.fr (S.R.)

5 Université de Rennes 1, ImPACcell, SFR Biosit, Bât. 8, 2 Avenue du Professeur Léon Bernard, CS 34317, 35043 Rennes Cedex, France; E-Mails: remy.leguevel@univ-rennes1.fr (R.L.G.); myriam.ravache@gmail.com (M.R.); anne.corlu@univ-rennes1.fr (A.C.)

* Author to whom correspondence should be addressed; E-Mail: jean-pierre.bazureau@univ-rennes1.fr; Tel.: +33-223-236-603; Fax: +33-223-236-374.

Academic Editor: Marilena Radoiu

Received: 27 May 2015 / Accepted: 30 June 2015 / Published: 8 July 2015

Abstract: A new route to 3-(4-arylmethylamino)butyl-5-arylidene-2-thioxo-1,3-thiazolidine-4-one 9 was developed in six steps from commercial 1,4-diaminobutane 1 as starting material. The key step of this multi-step synthesis involved a solution phase “one-pot
two-steps" approach assisted by microwave dielectric from \(N\)-(arylmethyl)butane-1,4-diamine hydrochloride \(6a–f\) (as source of the first point diversity) and commercial bis-(carboxymethyl)-trithiocarbonate reagent \(7\) for construction of the rhodanine platform. This platform was immediately functionalized by Knoevenagel condensation under microwave irradiation with a series of aromatic aldehydes \(3\) as second point of diversity. These new compounds were prepared in moderate to good yields and the fourteen synthetic products \(9a–n\) have been obtained with a Z-geometry about their exocyclic double bond. These new 5-arylidene rhodanines derivatives \(9a–n\) were tested for their kinase inhibitory potencies against four protein kinases: Human cyclin-dependent kinase 5-p25, \(Hs\)CDK5-p25; porcine Glycogen Synthase Kinase-3, GSK-3\(α/β\); porcine Casein Kinase 1, \(Ss\)CK1 and human \(Hs\)Haspin. They have also been evaluated for their \textit{in vitro} inhibition of cell proliferation (HuH7 D12, Caco 2, MDA-MB 231, HCT 116, PC3, NCI-H727, HaCat and fibroblasts). Among of all these compounds, \(9j\) presented selective micromolar inhibition activity on \(Ss\)CK1 and \(9i\) exhibited antitumor activities in the HuH7 D12, MDA-MBD231 cell lines.

**Keywords:** one-pot two-steps; Knoevenagel condensation; 5-arylidene rhodanine; protein kinase; inhibitor; \(Ss\)CK1; \(Hs\)CDK5-p25; cell lines; Alzheimer’s disease; cancer

1. Introduction

During the last decades, the five-membered heterocycle rings (FMHRs) are considered “privileged scaffolds” in medicinal chemistry [1]. Among the five-membered heterocycle rings, the 5-arylidene-2-thioxo-1,3-thiazolidine-4-ones or 5-arylidene rhodanine derivatives represented particularly privileged moieties in drug discovery because they have an inherent tendency for biological activity [2], such as DDX3 inhibitor for HIV replication [3], as potent and selective inhibitors of the “atypical” dual-specificity phosphatase (DSP) family member-JNK-stimulating phosphatase-1 (JSP-1) [4], as pancreatic cholesterol esterase (CEase) inhibitor [5] with IC\(_{50}\) values ranging from 1.44 to 85 \(\mu\)M. To discover chemical probes to further understand the function of human DNA polymerase \(λ\) in cancer, an inhibitor high-throughput screening (HTS) using SYBR\textsuperscript{®} Green-base assay [6] revealed that three 5-arylidene-2-thioxo-1,3-thiazolidine-4-ones were identified as strong inhibitors. A new series of D-glutamic acid-base \textit{Escherichia coli} MurD inhibitors incorporating the 5-arylidene rhodanine scaffold have been designed, synthesized and evaluated [7]. Substituted 5-arylidene-2-thioxo-1,3-thiazolidine-4-ones were able to inhibit HIV replication in MT-4 cells at low micromolar concentration with an appreciable selectivity index and are good scaffolds for the development of novel HIV-1 integrase inhibitors [8]. Recently, a family of 5-arylalkylidene rhodanine derivatives presented antiviral activity against chikungunya virus (LR2006\_OPY1) in Vero cell culture by cytopathic effect CPE reduction assay [9]. For Alzheimer’s disease, the FMHRs derived from 5-arylidene-2-thioxo-1,3-thiazolidine-4-ones have been described for amyloid polypeptide fibril formation [10], regulation of Cathepsin D immuno-reactivity in the senile plaques [11] and inhibition of tau aggregation [12].

Protein kinases represent an important class of enzymes that play an important role in the regulation of various processes. These enzymes catalyze protein-phosphorylation on serine, threonine and tyrosine
residues, which are frequently deregulated in human diseases. Only the 518 human kinases have been investigated as potential therapeutic targets [13]. Consequently, the search of protein-kinase inhibitors represented interesting targets in the pharmaceutical industry for new therapeutic agents. Over the past decade, our research group have investigated the chemical development of five-membered heterocycle rings derived from marine alkaloid as low-molecular weight-inhibitors of dual specificity, tyrosine phosphorylation-regulated kinases (DYRKs) and CLKs (edc2-like kinases) [14–16], two families of kinases involved in various diseases including Alzheimer’s disease (AD) [17], and also cancer [18–20].

Continuing in the effort to identify new DYRK inhibitors, particularly DYRK1A, we continued to explore successively the synthesis of \(N,N'\)-bis-(5-arylidene-4-oxo-3,5-dihydro-4H-imidazol-2-yl) diamines [21], \(N,N'\)-bis-(5-arylidene-4-oxo-4,5-dihydrothiazolidine-2-yl)aminopropylpiperazines [22] and finally unsymmetrical linked bis-5-arylidene-rhodanine derivatives linked in \(N\)-3 position [23] as potential kinases inhibitors (Figure 1). Among the three series of symmetrical or unsymmetrical \(N,N'\)-diamines bearing various platforms (imidazolidine-4-one or 2-thioxo-1,3-thiazolidine-4-one moieties), only one of these compounds has shown nanomolar inhibition potency (IC\(_{50}\) 40 nM) towards DYRK1A. Owing to the fact that none of the unsymmetrical linked bis-5-arylidene rhodanines presented a significant activity against representative tumoral cell lines, we decided to change one of the two heterocyclic platforms by various (arylmethyl)aminobutyl moieties grafted on \(N\)-3 position of 5-arylidene-rhodanine derivatives in order to obtain significant potential biological activities on tumoral cell lines and effects on some protein kinases (\(Hs\)CDK5-p25, \(Ss\)GSK3α/β, \(Ss\)CK1 and \(Hs\)Haspin). Herein, we present the building of this 3-(4-arylmethylamino)butyl-5-arylidene-rhodanine library in which the 5-arylidene-2-thioxo-1,3-thiazolidine-4-one moiety was mostly carried out under microwave irradiation [24] and the biological activities of these compounds.

![Diagram](image.png)

**Figure 1.** Symmetrical linked bis-5-arylidene imidazoline-4-ones, symmetrical linked bis-5-arylidene-1,3-thiazoline-4-ones and unsymmetrical linked bis-5-arylidene rhodanines derivatives (as inhibitors of the protein kinase DYRK1A or against cell proliferation on cell tumor lines) identified and developed in our group.

## 2. Results and Discussion

### 2.1. Chemistry

Access to the planned 3-(4-arylpheynlamino)butyl-5-arylidene-2-thioxo-1,3-thiazolidine-4-ones \(9\) is outlined in Scheme 1. For this study, we selected a diamino linker \(1\) with a butyl chain in order to obtain a good molecular flexibility between the 4-arylmethyl moiety and the 2-thioxo-1,3-thiazolidine-4-one
platform. A molecule with the lowest number of carbons for the diamino linker is more conformationally restrained [25]. The 1,4-diamino-butane linker 1 was treated with di-tert-butylcarboxylate (Boc₂O) in 1,4-dioxane at room temperature to afford mainly the mono-N-Boc protected amine 2 in good yield (84%) [26]. To obtain a sufficient number of compounds suitable for a preliminary biological screening, we privileged the transformation of N-Boc-1,4-diamino-butane 2 into mono protected N-arylmethyl diamine 5 by reductive amination in two steps. The (4-arylmethylamino)butyl chain appended on the N-3 position of the 2-thioxo-1,3-thiazolidine-4-one platform represent the first point of diversity for the desired target compounds 9. Preparation of 4 was easily realized by reaction between of appropriate arylaldehyde 3a–f with N-Boc diamine 2 in the presence of molecular sieves 3Å and, the condensation was conducted in a solution of diethyl ether Et₂O at room temperature during 24 h. Then, transformation of arylaldimines 4 into mono protected N-arylmethyl diamines 5 could be readily accomplished in good yields (85% to 98%) using NaBH₄ (5 equiv.) in MeOH at 50 °C during 24 h. For the cleavage of N-Boc group, the use of trifluoroacetic acid in dichloromethane is often efficient, but in our case, we observed the formation of impurities resulting from uncontrolled degradation of 5 in this strong acidic media. We thus preferred to use a more classical and practical approach by using a solution of 6M HCl. The deprotection was conducted in 1,4-dioxane at room temperature after 4 h of reaction time. 

As can be seen from the data presented in Table 1, the N-(arylmethyl)butane-1,4-diamine hydrochloride 6a–f were efficiently prepared with electron-rich and electron-poor aldehydes 3a–f in yields ranging from 72% to 98%.

Scheme 1. Route used for the preparation of 3-(4-arylphenylamino)butyl-5-arylidene-2-thioxo-1,3-thiazolidine-4-ones 9 via the “one-pot two-steps” reaction under microwave. 

Reagents and reaction conditions: (i) (t-BuO₂C)₂O, 1,4-dioxane, 25 °C, 24 h. (ii) 3 1 equiv., molecular sieve 3Å, Et₂O, 25 °C, 16 h. (iii) NaBH₄ 5 equiv., MeOH, 50 °C, 24 h. (iv) HCl 6 M, 1,4-dioxane, 25 °C, 4 h. (v) 6 1 equiv., 7 1 equiv., Et₃N 2 or 3 equiv., DME, MWI, 90 °C, 15 or 30 min. (vi) 3 1 equiv., MWI, 110 °C, 15 or 30 min.

With the desired salts 6 in hand, we wished to examine the construction of the 2-thioxo-1,3-thiazolidine-4-one platform followed by Knoevenagel condensation using arylaldehyde 3 under microwave irradiation for installation of the 5-arylidene moiety. The choice of arylaldehydes 3 in Knoevenagel condensation represent, in fact, the second point of diversity in the final structure of the targeted compounds 9. In a previous work issued from our laboratory [23], we have developed a “one-pot two-steps” method under microwave irradiation, which was applied for the synthesis of unsymmetrical linked bis-arylidene rhodanines from symmetric diamines.
Table 1. Results for the preparation of compounds 2, 4, 5, 6 and 9.

<table>
<thead>
<tr>
<th>Compound</th>
<th>Starting Product</th>
<th>Reaction Conditions under MWI ( ^a )</th>
<th>Yield ( ^b ) (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>-</td>
<td>-</td>
<td>84</td>
</tr>
<tr>
<td>4a</td>
<td>3a</td>
<td>-</td>
<td>97</td>
</tr>
<tr>
<td>4b</td>
<td>3b</td>
<td>-</td>
<td>99</td>
</tr>
<tr>
<td>4c</td>
<td>3c</td>
<td>-</td>
<td>98</td>
</tr>
<tr>
<td>4d</td>
<td>3d</td>
<td>-</td>
<td>98</td>
</tr>
<tr>
<td>4e</td>
<td>3e</td>
<td>-</td>
<td>98</td>
</tr>
<tr>
<td>4f</td>
<td>3f</td>
<td>-</td>
<td>99</td>
</tr>
<tr>
<td>5a</td>
<td>4a</td>
<td>-</td>
<td>93</td>
</tr>
<tr>
<td>5b</td>
<td>4b</td>
<td>-</td>
<td>99</td>
</tr>
<tr>
<td>5c</td>
<td>4c</td>
<td>-</td>
<td>85</td>
</tr>
<tr>
<td>5d</td>
<td>4c</td>
<td>-</td>
<td>90</td>
</tr>
<tr>
<td>5e</td>
<td>4e</td>
<td>-</td>
<td>97</td>
</tr>
<tr>
<td>5f</td>
<td>4f</td>
<td>-</td>
<td>98</td>
</tr>
<tr>
<td>6a</td>
<td>5a</td>
<td>-</td>
<td>92</td>
</tr>
<tr>
<td>6b</td>
<td>5b</td>
<td>-</td>
<td>72</td>
</tr>
<tr>
<td>6c</td>
<td>5c</td>
<td>-</td>
<td>92</td>
</tr>
<tr>
<td>6d</td>
<td>5d</td>
<td>-</td>
<td>92</td>
</tr>
<tr>
<td>6e</td>
<td>5e</td>
<td>-</td>
<td>92</td>
</tr>
<tr>
<td>6f</td>
<td>5f</td>
<td>-</td>
<td>92</td>
</tr>
</tbody>
</table>

Notes: \( ^a \) Reaction realized in a tube (sealed with a snap cap) under microwave irradiation (μω) with the Monowave\( ^{30} \) 300 Anton-Paar reactor. \( ^b \) Isolated yield. \( ^c \) Reaction temperature: 90 °C for the preparation of 8 (1st period of microwave irradiation). \( ^d \) Reaction temperature: 110 °C for condensation reaction after addition of 3 (2nd period of microwave irradiation).

Application of this methodology, based on a “modified Holmberg method”, first involved, in our case, the reaction of the commercial bis-(carboxymethyl)-trithiocarbonate reagent 7 with the synthesized...
N-(arylmethyl)butane-1,4-diamine hydrochloride 6 to afford the intermediate 8 and secondly, Knoevenagel condensation to produce the desired 5-arylidene rhodanines 9. This “one-pot two-steps” methodology was realized under microwave dielectric heating because the use of commercial scientific laboratory microwave apparatus favoured higher product yields and significant rate enhancements compared to reactions which run with conventional heating (i.e., in oil bath) [27]. After several experiments, optimal reaction conditions for this “one-pot two-steps” synthesis involved reaction of a stoichiometric mixture of N-(arylmethyl)butane-1,4-diamine hydrochloride 6 and commercial reagent 7 solubilized in dimethoxyethane with one or two equivalents of triethylamine. This starting reaction mixture was placed in a commercial glass tube closed with a snap cap and was irradiated with appropriate reaction time (15 or 30 min.) at 90 °C. After this first period of microwave irradiation followed by a cooling down to room temperature, aromatic aldehyde 3 was directly added to the crude suspension and was submitted again to microwave dielectric heating at 110 °C during 15 or 30 min for condensation.

The desired compound 9 was obtained as precipitate after addition of methanol in the solventless crude reaction mixture (after elimination of the volatile compounds in vacuo) and triturated, followed by successive washings with deionized water, ethanol, ether and finally was recrystallized from absolute EtOH to increase the quality of the precipitated product 9. A set of 14 pure compounds 9a–n was prepared in 5%–59% yield (Table 1) and all the products 9 were characterized by 1H-, 13C-NMR and HRMS before entering the biological tests. For the exocyclic double bond (CH=C) in C-5 position of all the 3-(4-arylmethylamino)butyl-2-thioxo-1,3-thiazolidine-4-one 9a–n, it’s possible in theory to observe E- and/or Z-geometrical isomers. Examination of their 1H-NMR spectra in DMSO-d6 showed only one signal for the methylene proton (CH=) in the range 7.54–7.74 ppm at lower field values than those expected for the E-isomers, which indicates that all the compounds 9a–n have the Z-configuration due to the high degree of thermodynamic stability of this isomer [28,29]. In 13C-NMR, the signal of C-5 (C=) appears in the range 120.0–127.5 ppm, and we also observed only one signal for the exocyclic methylene proton (134.6 < δCH= < 135.9 ppm) that confirmed the presence of only the Z-geometrical isomer for the targeted compounds 9a–n.

2.2. Biology

As an initial effort to investigate their in vitro bioactivity, the new synthesized 3-(4-arylmethylamino)butyl-2-thioxo-1,3-thiazolidine-4-one 9a–n and also their precursors 6a–f were evaluated for their in vitro inhibition of cell proliferation. For this study, we used a panel of seven representative tumoral cell lines, namely HuH7 D12 (differential hepato cellular carcinoma), Caco 2 (differentiating colorectal adenocarcinoma), MDA-MBD231 (prostate carcinoma), HCT 116 (actively proliferating colorectal adenocarcinoma), PC3 (prostate carcinoma), NCI-H727 (lung carcinoma), HaCat keratinocyte and, diploid skin fibroblasts as normal cell lines for control. Roscovitine, Doxorubicine and Taxol were also used as positive controls and their IC50 values are compared with those obtained for compounds 6 and 9. Results of the in vitro antiproliferative data activity are reported in Table 2. None of the N-(arylmethyl)butane-1,4-diamines hydrochloride 6a–f presented a significant activity against the seven representative tumoral cell lines. For the other compounds 9, the most active compound was clearly 9i (Figure 2) and exhibited antitumor activities in the HuH7 D12, MDA-MBD231 and HaCat cell lines with IC50 values lower than 10 µM (HuH7 D12 and MDA-MBD231: IC50 9 µM) and did not inhibit the growth of normal
fibroblasts (IC\textsubscript{50} > 25 \mu M). In addition, compounds 9d, 9h, 9j and 9n presented moderate antitumor activities (IC\textsubscript{50} 10–17 \mu M) in all cell lines of the panel but without selectivity.

**Table 2.** Antiproliferative activity of compounds 6(a–e) and 9(a–n) on six representative tumor cell lines.

<table>
<thead>
<tr>
<th>Compound</th>
<th>% of Survival \textsuperscript{a} and IC\textsubscript{50} (\mu M) of Selected Compounds \textsuperscript{b}</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Huh7 D12</td>
</tr>
<tr>
<td>6a</td>
<td>102</td>
</tr>
<tr>
<td>6b</td>
<td>107</td>
</tr>
<tr>
<td>6c</td>
<td>82</td>
</tr>
<tr>
<td>6d</td>
<td>107</td>
</tr>
<tr>
<td>6e</td>
<td>91</td>
</tr>
<tr>
<td>6f</td>
<td>88</td>
</tr>
<tr>
<td>9a</td>
<td>85</td>
</tr>
<tr>
<td>9b</td>
<td>105</td>
</tr>
<tr>
<td>9c</td>
<td>88</td>
</tr>
<tr>
<td>9d</td>
<td>7 (10)</td>
</tr>
<tr>
<td>9e</td>
<td>74</td>
</tr>
<tr>
<td>9f</td>
<td>102</td>
</tr>
<tr>
<td>9g</td>
<td>111</td>
</tr>
<tr>
<td>9h</td>
<td>7 (10)</td>
</tr>
<tr>
<td>9i</td>
<td>5 (9)</td>
</tr>
<tr>
<td>9j</td>
<td>22 (11)</td>
</tr>
<tr>
<td>9k</td>
<td>72</td>
</tr>
<tr>
<td>9l</td>
<td>90</td>
</tr>
<tr>
<td>9m</td>
<td>97</td>
</tr>
<tr>
<td>9n</td>
<td>41 (15)</td>
</tr>
<tr>
<td>Roscovitine</td>
<td>21 (15)</td>
</tr>
<tr>
<td>Doxorubicine</td>
<td>63 (0.03)</td>
</tr>
<tr>
<td>Taxol</td>
<td>36 (0.003)</td>
</tr>
<tr>
<td>DMSO</td>
<td>100 (&gt;25)</td>
</tr>
</tbody>
</table>

Notes: \textsuperscript{a} Percentage of survival measured at 25 \mu M (after 48 h using a single dose, triplicate). \textsuperscript{b} IC\textsubscript{50} values in brackets are expressed in \mu M and are the average of three assays, standard error ± 0.5 \mu M.

**Figure 2.** Structure of compounds 9d, 9(h–j) and 9n, which are active against protein kinases and/or tumor cell lines.
Then, we evaluated the intermediates salts 6a–f and fourteen final compounds 9a–n on four protein kinases: HsCDK5-p25 (cyclin-dependant kinase 5-p25), SsGSK-3α/β (glycogen synthase kinase-3α/β) SsCK1δ/ε (Caseine kinase 1) and HsHaspin as an essential mitotic kinase [30]. All assays were run in the presence of 15 μM of ATP and appropriate substrates. IC50 values were determined from dose-response curves. Concerning the library of the fourteen (5Z) 3-(4-arylmethylamino)butyl-5-arylidene-2-thioxo-1,3-thiazolidine-4-ones 9a–n and also the six diamines salts 6a–f, the results are given in Table 3. Among all the products, the main part of their inhibition activity was focused on two proteins kinases (SsCK1 and HsCDK5-p25) for four products, which exhibited inhibitory activity in the micromolar range (SsCK1: 1.4 μM < IC50 < 6.6 μM; HsCDK5-p25: IC50 1.2 μM). For the compound 9j and 9n, we observed micromolar inhibition activity on SsCK1 (IC50 values for 9j: 1.4 μM and 9n: 2 μM) with good selectivity. It is interesting to note that these two compounds are respectively substituted with an hydroxyl function on para-position of the 5-arylidene moiety and also with a similar bulky group (1,3-benzodioxol-5-yl for 9j and 2,3-dihydro-benzo[1,4]dioxin-6-yl for 9n) on the arylmethylamino moiety grafted on the N-3 butyl chain. On the contrary, the presence of 1,3-benzodioxol-5-yl (for 9h) or 2,3-dihydro-benzo[1,4]dioxin-6-yl moiety (for 9i) in the 5-arylidene group induced a moderate loss of inhibition activity for SsCK1 (IC50 for 9h: 6.6 μM and 9i: 5.4 μM) associated with micromolar affinity for HsCDK5-p25 (IC50 for 9h: 1.1 μM and 9i: 1.3 μM). Compound 9j presented also inhibition proliferation activity on Huh7 D12, MDA-MBD231 cell lines and is inactive against normal fibroblasts.

Table 3. Effects of the compounds 6a–f and 9a–n on the catalytic activity of four purified protein kinases a.

<table>
<thead>
<tr>
<th>Compound</th>
<th>HsCDK5/p25</th>
<th>SsGSK-3α/β</th>
<th>SsCK1δ/ε</th>
<th>HsHaspin</th>
</tr>
</thead>
<tbody>
<tr>
<td>6a–f</td>
<td>&gt;10</td>
<td>&gt;10</td>
<td>&gt;10</td>
<td>&gt;10</td>
</tr>
<tr>
<td>9a–g</td>
<td>&gt;10</td>
<td>&gt;10</td>
<td>&gt;10</td>
<td>&gt;10</td>
</tr>
<tr>
<td>9h</td>
<td>1.1</td>
<td>&gt;10</td>
<td>6.6</td>
<td>&gt;10</td>
</tr>
<tr>
<td>9i</td>
<td>1.3</td>
<td>&gt;10</td>
<td>5.1</td>
<td>&gt;10</td>
</tr>
<tr>
<td>9j</td>
<td>&gt;10</td>
<td>&gt;10</td>
<td>1.4</td>
<td>&gt;10</td>
</tr>
<tr>
<td>9k–m</td>
<td>&gt;10</td>
<td>&gt;10</td>
<td>&gt;10</td>
<td>&gt;10</td>
</tr>
<tr>
<td>9n</td>
<td>10</td>
<td>&gt;10</td>
<td>2</td>
<td>&gt;10</td>
</tr>
</tbody>
</table>

Note: a Compounds were tested at various concentrations on each kinase as described in Experimental Section. IC50 values, calculated from the dose-response curves, are reported in μM > 10, inhibitory but IC50 > 10 μM.

3. Experimental Section

3.1. Chemistry Section

3.1.1. General Section

Melting points were determined on a Kofler melting point apparatus and were uncorrected. Thin-layer chromatography (TLC) was accomplished on 0.2-mm precoated plates of silica gel 60 F-254 (Merck, Fontenay-sous-Bois, France). Visualization was made with ultraviolet light (254 and 365 nm) or with a fluorescence indicator. 1H-NMR spectra were recorded on BRUKER AC 300 P (300 MHz) spectrometer, 13C-NMR spectra on a BRUKER AC 300 P (75 MHz) spectrometer. Chemical shifts are expressed in parts per million downfield from tetramethyilsilane as an internal standard. Data are given in the
following order: $\delta$ value, multiplicity (s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet; br, broad), number of protons, coupling constants $J$ is given in Hertz. The mass spectra (HRMS) were taken respectively on a MS/MS ZABSpec Tof Micromass (EBE TOF geometry) at an ionizing potential of 8 eV and on a VARIAN MAT 311 at an ionizing potential of 70 eV in the Centre Régional de Mesures Physiques de l’Ouest (CRMPO, Rennes, France). Reactions under microwave irradiations were realized in the Anton Paar Monowave 300® microwave reactor (Anton-Paar, Courtaboeuf, France) using borosilicate glass vials of 10 mL equipped with snap caps (at the end of the irradiation, cooling reaction was realized by compressed air). The microwave instrument consists of a continuous focused microwave power output from 0 to 800 W for this Monowave 300® apparatus. All the experiments in the microwave reactor were performed using a stirring option. The target temperature was reached with a ramp of 5 min and the chosen microwave power stayed constant to hold the mixture at this temperature. The reaction temperature is monitored using calibrated infrared sensor and the reaction time included the ramp period. The microwave irradiation parameters (power and temperature) were monitored by the Monowave software package of the Monowave 300® microwave reactor. Solvents were evaporated with a BUCHI rotary evaporator. All reagents and solvents were purchased from Acros, Sigma-Aldrich Chimie (Saint-Quentin Fallavier, France), TCI France and Fluka France and were used without further purification.

tert-Butyl (4-aminobutyl)carbamate (2). In a 250 mL two-necked round-bottomed flask, provided with magnetic stirrer and condenser, commercial 1,4-diaminobutane 1 (14.3 mL, 12.5 g, 0.14 mol) was solubilized in 69 mL of 1,4-dioxane at room temperature. To this mixture was added dropwise a solution of commercial di-tert-butyldicarbonate (6.5 g, 30 mmol) in 1,4-dioxane (85 mL) over a period of 3 h. After vigorous stirring at 25 °C during a 12 h period, the volatile compounds of the reaction mixture were eliminated in vacuo and into the crude reaction mixture was poured 150 mL of deionized water. The mixture was extracted with methylene chloride (5 × 50 mL), organic phases were collected and dried over magnesium sulfate. The filtrate was concentrated in a rotary evaporator under reduced pressure and was dried under high vacuum (10⁻² Torr) at 25 °C for 10 min. The desired carbamate 2 (1.58 g) was obtained as a colourless mobile oil in 84% yield and was further used without purification. ¹H-NMR (DMSO-$d_6$) $\delta$: 1.34 (s, 9H, Me₂CO); 1.40 (m, 4H, CH₂); 2.61 (t, 2H, $J = 6.7$ Hz, CH₂); 3.02 (m, 2H, CH₂NH); 4.93 (br s, 2H, NH₂); 5.71 (br s, 1H, NH). ¹³C-NMR (DMSO-$d_6$) $\delta$: 27.2 (CH₂); 28.4 (OC(CH₃)₃); 29.5 (CH₂); 40.2 (CH₂); 41.1 (CH₂); 78.9 (OC(CH₃)₃); 156.1 (C=O). HRMS, $m/z$: 189.1600 found (calculated for C₉H₂₁N₂O₂ [M + H]$^+$ requires 189.1603).

3.1.2. Standard Procedure for the Preparation of Aldimines 4a–f from tert-Butyl (4-aminobutyl) Carbamate 2 and Aromatic Aldehyde 3a–f

In a 100 mL two-necked round-bottomed flask, provided with magnetic stirrer and condenser, containing a solution of tert-butyl (4-aminobutyl)carbamate 2 (1.97 g, 10.5 mmol) in anhydrous ether (50 mL) was added dropwise during 30 min, a suspension of commercial aldehyde (10 mmol) in anhydrous ether (30 mL) and molecular sieves (2 g, 3 Å, 8–12 mesh). The crude reaction mixture is stirred vigorously for 16 h at room temperature until the disappearance of aromatic aldehyde 3 controlled by thin-layer chromatography on 0.2-mm precoated plates of silica gel 60 F-254 (Merck). The crude reaction mixture
was filtered on filter paper and then concentrated in a rotary evaporator under reduced pressure. The crude residue was dried under high vacuum (10^{-2} Torr) at 25 °C for 10 min. The desired aldimine 4 was obtained as yellowish viscous oil and was further used without purification.

[4-(Benzyldiene-amino)-butyl]-carbamic acid tert-butyl ester (4a). Compound 4a was prepared in 97% yield (2.68 g) from benzaldehyde 3a (1.06 g, 10 mmol) according to the standard procedure as yellowish viscous oil. 1H-NMR (DMSO-d6) δ: 1.37 (s, 9H, Me 3C); 1.40–1.48 (m, 2H, CH2, H-2); 1.54–1.62 (m, 2H, CH2, H-3); 2.96 (q, 2H, J = 6.8 Hz, CH2NH, H-1); 3.55 (t, 2H, J = 6.8 Hz, CH2N=, H-4); 6.79–6.83 (br s, 1H, NH); 7.41–7.46 (m, 3H, H-3’, H-4’, H-5’, Ar); 7.71–7.75 (m, 2H, H-2’, H-6’, Ar); 8.32 (s, 1H, N=CH). 13C-NMR (DMSO-d6) δ: 27.3 (C-2); 27.8 (C-3); 28.2 (OC(CH3)3); 39.6 (C-1); 60.1 (C-4); 77.2 (OC(CH3)3); 127.7 (C-2’, C-6’, Ar); 128.5 (C-3’, C-5’, Ar); 130.4 (C-4’, Ar); 136.1 (C-1’, Ar); 155.5 (C=O); 160.5 (N=CH).

{4-[(4-Methoxy-benzylidene)-amino]-butyl}-carbamic acid tert-butyl ester (4b). Compound 4b was prepared in 99% yield (3.03 g) from 4-methoxybenzaldehyde 3b (1.36 g, 10 mmol) according to the standard procedure as yellowish viscous oil. 1H-NMR (DMSO-d6) δ: 1.37 (s, 9H, Me 3C); 1.32–1.46 (m, 2H, CH2, H-2); 1.52–1.61 (m, 2H, CH 2, H-3); 2.94 (q, 2H, J = 6.9 Hz, CH2NH, H-1); 3.50 (t, 2H, J = 6.8 Hz, CH2N=, H-4); 3.80 (s, 3H, OCH3); 6.77–6.82 (br s, 1H, NH); 6.98 (d, 2H, J = 8.8 Hz, H-2’, H-6’, Ar); 7.66 (d, 2H, J = 8.8 Hz, H-3’, H-5’, Ar); 8.24 (s, 1H, N=CH). 13C-NMR (DMSO-d6) δ: 27.4 (C-2); 28.0 (C-3); 28.2 (OC(CH3)3); 39.6 (C-1); 55.2 (OCH3); 60.1 (C-4); 77.3 (OC(CH3)3); 114.0 (C-2’, C-6’, Ar); 129.0 (C-1’, Ar); 129.3 (C-3’, C-5’, Ar); 155.5 (C=O); 159.7 (N=CH); 161.0 (C-4’, Ar).

{4-[(2-Chloro-benzylidene)-amino]-butyl}-carbamic acid tert-butyl ester (4c). Compound 4c was prepared in 98% yield (3.05 g) from 2-chlorobenzaldehyde 3c (1.41 g, 10 mmol) according to the standard procedure as yellowish viscous oil. 1H-NMR (DMSO-d6) δ: 1.37 (s, 9H, Me 3C); 1.34–1.48 (m, 2H, CH2, H-2); 1.56–1.63 (m, 2H, CH 2, H-3); 2.96 (q, 2H, J = 6.7 Hz, CH2NH, H-1); 3.61 (t, 2H, J = 6.8 Hz, CH2N=, H-4); 6.78–6.82 (br s, 1H, NH); 7.36–7.52 (m, 3H, H-4’, H-5’, H-6’, Ar); 7.94–7.97 (m, 1H, H-3’, Ar); 8.65 (s, 1H, N=CH). 13C-NMR (DMSO-d6) δ: 27.3 (C-2); 27.7 (C-3); 28.2 (OC(CH3)3); 39.6 (C-1); 127.4 (C-5’, Ar); 128.0 (C-4’, Ar); 129.8 (C-6’, Ar); 132.0 (C-3’, Ar); 132.7 (C-1’, C-2’, Ar); 133.8 (C-1’, C-2’, Ar); 155.5 (C=O); 156.6 (N=CH).

{4-[(3-Methoxy-benzylidene)-amino]-butyl}-carbamic acid tert-butyl ester (4d). Compound 4d was prepared in 98% yield (3.03 g) from 3-methoxybenzaldehyde 3d (1.41 g, 10 mmol) according to the standard procedure as yellowish viscous oil. 1H-NMR (DMSO-d6) δ: 1.37 (s, 9H, Me 3C); 1.34–1.48 (m, 2H, CH2, H-2); 1.56–1.63 (m, 2H, CH2, H-3); 2.96 (q, 2H, J = 6.7 Hz, CH2NH, H-1); 6.19 (s, 1H, N=CH). 13C-NMR (DMSO-d6) δ: 27.3 (C-2); 27.7 (C-3); 28.2 (OC(CH3)3); 39.6 (C-1); 55.0 (OCH3); 60.1 (C-4); 77.2 (OC(CH3)3); 111.9 (C-5’, Ar); 116.6 (C-6’, Ar); 120.6 (C-4’, Ar); 129.6 (C-2’, Ar); 137.6 (C-1’, Ar); 155.5 (C=O); 159.4 (C-3’, Ar); 160.4 (C=O).

{4-[(Benzo[1,3]dioxol-5-ylmethylene)-amino]-butyl}-carbamic acid tert-butyl ester (4e). Compound 4e was prepared in 98% yield (3.13 g) from 3,4-methylenedioxybenzaldehyde 3e (1.51 g, 10 mmol).
according to the standard procedure as yellowish viscous oil. \(^{1}\)H-NMR (DMSO-\(d_6\)) \(\delta\): 1.37 (s, 9H, Me\(\text{C}\)); 1.30–1.46 (m, 2H, CH\(\text{2}\), H-2); 1.51–1.61 (m, 2H, CH\(\text{2}\), H-3); 2.94 (q, 2H, \(J = 6.9\) Hz, CH\(\text{2}\)NH, H-1); 3.49 (t, 2H, \(J = 6.7\) Hz, CH\(\text{2}\)N=, H-4); 6.07 (s, 2H, H-2', Ar); 6.77–6.82 (br s, 1H, NH); 6.77–6.82 (m, 1H, H-4', Ar); 6.97 (d, 1H, \(J = 8\) Hz, H-6', H-7', Ar); 7.16–7.20 (m, 1H, H-6', H-7', Ar); 7.27–7.28 (m, 1H, H-4', Ar); 8.20 (s, 1H, N=CH). \(^{13}\)C-NMR (DMSO-\(d_6\)) \(\delta\): 27.3 (C-2); 27.9 (C-3); 28.2 (OC(CH\(\text{3}\))\(\text{3}\)); 39.6 (C-1); 59.8 (C-4); 77.4 (OC(CH\(\text{3}\))\(\text{3}\)); 101.4 (C-2', Ar); 105.8 (C-6', Ar); 108.1 (C-7', Ar); 124.0 (C-4', Ar); 130.9 (C-5', Ar); 147.8 (C-3a', C-7a', Ar); 149.3 (C-3a', C-7a', Ar); 155.5 (C=O); 160.0 (C=N).

\{4-[(2,3-Dihydro-benzo[1,4]dioxin-6-ylmethylene)-amino]-butyl\}-carbamic acid tert-butylester (4f). Compound 4f was prepared in 99% yield (3.31 g) from 2,3-dihydro-benzo[1,4]dioxin-6-carbaldehyde 3f (1.50 g, 10 mmol) according to the standard procedure as yellowish viscous oil. \(^{1}\)H-NMR (DMSO-\(d_6\)) \(\delta\): 1.37 (s, 9H, Me\(\text{C}\)); 1.34–1.45 (m, 2H, CH\(\text{2}\), H-2); 1.51–1.60 (m, 2H, CH\(\text{2}\), H-3); 2.94 (q, 2H, \(J = 6.8\) Hz, CH\(\text{2}\)NH, H-1); 3.50 (t, 2H, \(J = 6.7\) Hz, CH\(\text{2}\)N=, H-4); 4.23–4.30 (m, 4H, H-2', H-3', Ar), 6.77–6.81 (br s, 1H, NH); 6.88–6.91 (m, 1H, H-7', Ar); 7.18–7.21 (m, 2H, H-5', H-8', Ar); 8.17 (s, 1H, N=CH).

\(^{13}\)C-NMR (DMSO-\(d_6\)) \(\delta\): 27.3 (C-2); 27.9 (C-3); 28.2 (OC(CH\(\text{3}\))\(\text{3}\)); 39.6 (C-1); 60.0 (C-4); 63.9 (C-2', Ar), 64.2 (C-3', Ar), 77.2 (OC(CH\(\text{3}\))\(\text{3}\)); 116.0 (C-7', Ar); 117.1 (C-8', Ar); 121.2 (C-5', Ar); 129.8 (C-6', Ar); 143.4 (C-4a', C-8a', Ar); 145.4 (C-4a', C-8a', Ar); 155.5 (C=O); 159.6 (C=N).

3.1.3. Standard Procedure for Reduction of Aldimines 4a–f into \(\text{N-Boc Monoprotected Diamines 5a–f}

In a 50 mL two-necked round-bottomed flask, provided with a magnetic stirrer and reflux condenser, compound 4 (5 mmol) was dissolved in methanol p.a. (30 mL) under vigorous stirring and cooled at 0 °C. To this solution was added by small portions commercial sodium borohydride NaBH\(_4\) (0.95 g, 25 mmol) over a period of 20 min. The resulting suspension was stirred at 50 °C for 24 h (monitored by TLC on 0.2-mm precoated plates of silica gel 60 F-254, Merck). After cooling down to room temperature, the volatile compounds were removed in a rotary evaporator under reduced pressure, then deionized water (40 mL) was added in one portion to the crude residue. The mixture was transferred to a separating funnel and was extracted with dichloromethane (3 × 50 mL). The combined organic phases were dried over magnesium sulphate MgSO\(_4\), filtered on filter paper and the solvent was eliminated in vacuo. The desired compound 5 was obtained as yellowish viscous oil and was further used without purification.

\(\text{(4-Phenylmethylamino-butyl)-carbamic acid tert-butylester (5a). Compound 5a was prepared in 93% yield (1.25 g) from [4-(benzylidene-amino)-butyl]-carbamic acid tert-butylester 4a (1.38 g, 5 mmol) according to the standard procedure that gave 5a as yellowish viscous oil. \(^{1}\)H-NMR (DMSO-\(d_6\)) \(\delta\): 1.37 (s, 9H, Me\(\text{C}\)); 1.34–1.42 (m, 4H, CH\(\text{2}\), H-2, H-3); 2.45 (t, 2H, \(J = 6.2\) Hz, CH\(\text{2}\)NH, H-4); 2.89–2.91 (m, 2H, CH\(\text{2}\)NH, H-1); 3.66 (s, 2H, ArCH\(\text{2}\)NH); 6.78–6.82 (br s, 1H, NHCO); 7.18–7.21 (m, 1H, H-4', Ar); 7.26–7.33 (m, 4H, H-2', H-3', H-5', H-6', Ar). \(^{13}\)C-NMR (DMSO-\(d_6\)) \(\delta\): 27.3 (C-2); 27.9 (C-3); 28.2 (OC(CH\(\text{3}\))\(\text{3}\)); 39.6 (C-1); 60.0 (C-4); 63.9 (C-2', Ar), 64.2 (C-3', Ar), 77.2 (OC(CH\(\text{3}\))\(\text{3}\)); 116.0 (C-7', Ar); 117.1 (C-8', Ar); 121.2 (C-5', Ar); 129.8 (C-6', Ar); 143.4 (C-4a', C-8a', Ar); 145.4 (C-4a', C-8a', Ar); 155.5 (C=O); 159.6 (C=N).} \)

\[\text{[4-(4-Methoxy-phenylmethylamino)-butyl]-carbamic acid tert-butylester (5b). Compound 5b was prepared in 99% yield (1.52 g) from} \]

\[\text{[4-[(4-methoxy-benzylidene)-amino]-butyl]-carbamic acid} \]
tert-butyl ester \( 4b \) (1.53 g, 5 mmol) according to the standard procedure that gave \( 5b \) as yellowish viscous oil. \(^1\)H-NMR (DMSO-\( d_6 \)) \( \delta \): 1.37 (s, 9H, MeC); 1.33–1.41 (m, 4H, H-2, H-3); 2.45 (t, 2H, \( J = 6.5 \) Hz, CH\(_2\)NH, H-4); 2.88–2.90 (m, 2H, CH\(_2\)NHCO, H-1); 3.60 (s, 2H, ArCH\(_2\)NH); 3.72 (s, 3H, OCH\(_3\)); 6.76–6.80 (br s, 1H, NHCO); 6.85 (d, 2H, \( J = 8.5 \) Hz, H-2', H-6', Ar); 7.22 (d, 2H, \( J = 8.6 \) Hz, H-3', H-5', Ar). \(^1\)C-NMR (DMSO-\( d_6 \)) \( \delta \): 26.6 (C-2); 27.4 (C-3); 28.2 (OC(CH\(_3\))\(_3\)); 39.8 (C-4); 48.2 (C-1); 52.3 (ArCH\(_2\)NH); 54.9 (OCH\(_3\)); 77.2 (OC(CH\(_3\))\(_3\)); 113.4 (C-2', C-6', Ar); 129.0 (C-3', C-5', Ar); 132.6 (C-1', Ar); 155.5 (C=O); 158.0 (C-4', Ar).

\[4-(2-Chloro-phenylmethylamino)-butyl\]-carbamic acid tert-butyl ester (\( 5c \)). Compound \( 5c \) was prepared in 85\% yield (1.33 g) from \( 4\)-[\(2\)-chloro-benzylidene]-amino]-butyl]-carbamic acid tert-butyl ester \( 4c \) (1.55 g, 5 mmol) according to the standard procedure that gave \( 5c \) as yellowish viscous oil.

\(^1\)H-NMR (DMSO-\( d_6 \)) \( \delta \): 1.37 (s, 9H, MeC); 1.34–1.43 (m, 4H, CH\(_2\), H-2, H-3, 4H); 2.47–2.52 (m, 2H, ArCH\(_2\)NH, H-4); 2.88–2.92 (m, 2H, CH\(_2\)NHCO, H-1); 3.75 (s, 2H, ArCH\(_2\)NH); 6.77–6.80 (br s, 1H, NHCO); 7.21–7.33 (m, 2H, H-4', H-5', Ar); 7.37–7.4 (m, 1H, H-6', Ar); 7.5–7.53 (m, 1H, H-3', Ar).

\(^1\)C-NMR (DMSO-\( d_6 \)) \( \delta \): 26.7 (C-2); 27.4 (C-3); 28.2 (OC(CH\(_3\))\(_3\)); 39.8 (C-4); 48.5 (C-1); 50.0 (ArCH\(_2\)NH); 77.2 (OC(CH\(_3\))\(_3\)); 126.9 (C-5', Ar); 128.0 (C-4', Ar); 128.9 (C-6', Ar); 129.6 (C-3', Ar); 132.5 (C-1', Ar); 138.2 (C-2', Ar); 155.5 (C=O).

\[4-(3-Methoxy-phenylmethylamino)-butyl\]-carbamic acid tert-butyl ester (\( 5d \)). Compound \( 5d \) was prepared in 90\% yield (1.39 g) from \( 4\)-[\(3\)-methoxy-benzylidene]-amino]-butyl]-carbamic acid tert-butyl ester \( 4d \) (1.53 g, 5 mmol) according to the standard procedure that gave \( 5d \) as pale pink viscous oil.

\(^1\)H-NMR (DMSO-\( d_6 \)) \( \delta \): 1.37 (s, 9H, MeC); 1.34–1.46 (m, 4H, H-2, H-3); 2.45 (t, 2H, \( J = 6.4 \) Hz, CH\(_2\)NH, H-4); 2.89–2.91 (m, 2H, CH\(_2\)NHCO, H-1); 3.64 (s, 2H, ArCH\(_2\)NH); 3.74 (s, 3H, OCH\(_3\)); 6.75–6.80 (br s, 1H, NHCO); 6.75–6.91 (m, 3H, H-4', H-5', H-6', Ar); 7.17–7.22 (m, 1H, H-2', Ar).

\(^1\)C-NMR (DMSO-\( d_6 \)) \( \delta \): 26.8 (C-2); 27.4 (C-3); 28.2 (OC(CH\(_3\))\(_3\)); 39.8 (C-4); 48.4 (C-1); 52.9 (ArCH\(_2\)NH); 54.8 (OCH\(_3\)); 77.2 (OC(CH\(_3\))\(_3\)); 111.8 (C-5', Ar); 113.2 (C-6', Ar); 120.0 (C-4', Ar); 128.9 (C-2', Ar); 142.8 (C-1', Ar); 155.5 (C=O); 159.2 (C-3', Ar).

\[4-[\text{Benzo[1,3]}\text{dioxol-5-ylmethyl}-amino]-butyl\]-carbamic acid tert-butyl ester (\( 5e \)). Compound \( 5e \) was prepared in 97\% yield (1.56 g) from \( 4\)-[\(\text{benzo[1,3]}\text{dioxol-5-ylmethylene}\)-amino]-butyl]-carbamic acid tert-butyl ester \( 4e \) (1.60 g, 5 mmol) according to the standard procedure that gave \( 5e \) as yellowish viscous oil.

\(^1\)H-NMR (DMSO-\( d_6 \)) \( \delta \): 1.37 (s, 9H, MeC); 1.23–1.40 (m, 4H, H-2, H-3); 2.43 (t, 2H, \( J = 6.5 \) Hz, CH\(_2\)NH, H-4); 2.89–2.91 (m, 2H, CH\(_2\)NHCO, H-1); 3.64 (s, 2H, ArCH\(_2\)NH); 5.96 (s, 2H, H-2', Ar); 6.73–6.82 (br s, 1H, NHCO); 6.73–6.90 (m, 3H, H-4', H-6', H-7', Ar).

\(^1\)C-NMR (DMSO-\( d_6 \)) \( \delta \): 26.8 (C-2); 27.4 (C-3); 28.2 (OC(CH\(_3\))\(_3\)); 39.8 (C-4); 48.1 (C-1); 52.7 (ArCH\(_2\)NH); 77.2 (OC(CH\(_3\))\(_3\)); 100.6 (C-2', Ar); 107.7 (C-6', Ar); 108.2 (C-7', Ar); 120.8 (C-4', Ar); 135.0 (C-5', Ar); 145.7 (C-3a', C-7a', Ar); 147.1 (C-3a', C-7a', Ar); 155.5 (C=O).

\[4-[\text{2,3-Dihydro-benzo[1,4]}\text{dioxin-6-ylmethyl}-amino]-butyl\]-carbamic acid tert-butyl ester (\( 5f \)). Compound \( 5f \) was prepared in 88\% yield (1.48 g) from \( 4\)-[\(2,3\)-dihydro-benzo[1,4]dioxin-6-ylmethylene]-amino]-butyl]-carbamic acid tert-butylerster \( 4f \) (1.67 g, 5 mmol) according to the standard procedure that gave \( 5f \) as yellowish viscous oil.

\(^1\)H-NMR (DMSO-\( d_6 \)) \( \delta \): 1.37 (s, 9H, MeC); 1.33–1.43 (m, 4H, H-2, H-3); 2.42 (t, 2H, \( J = 6.4 \) Hz, CH\(_2\)NH, H-4); 2.88–2.90 (m, 2H, CH\(_2\)NHCO, H-1); 3.53 (s, 2H,
3.1.4. Standard Procedure for the Preparation of Hydrochloride Salts 6a–f after Deprotection of N-Boc Monoprotected Diamines 5a–f

In a 100 mL two-necked round-bottomed flask provided with a magnetic stirrer and condenser, N-Boc monoprotected diamine 5 (12.1 mmol, 1 equiv.) was solubilized in 1,4-dioxane (80 mL) at room temperature under vigorous stirring for 10 min. To this homogeneous solution was added dropwise for 1 h a solution of 6 M HCl (20 mL). The reaction mixture was stirred during 4 h at 25 °C and was concentrated in a rotary evaporator under reduced pressure for elimination of volatile compounds. To the crystallized crude reaction mixture was added 60 mL of anhydrous Et2O and after triturating, the insoluble salt 6 was collected by filtration, on a Büchner funnel (porosity N°4) then washed with 6 × 10 mL of Et2O. The desired salt 6 was dried under high vacuum (10⁻² Torr) at 25 °C for 1 h that gave a yellowish or white powder and was further used without purification.

N-1-Phenylmethyl-butane-1,4-diamine hydrochloride (6a). Compound 6a was prepared in 84% yield (2.18 g) from (4-phenylmethylamino-butyl)-carbamic acid tert-butyl ester 5a (3.37 g, 12.1 mmol) according to the standard procedure as yellowish powder. Mp > 260 °C. ¹H-NMR (DMSO-d₆) δ: 1.57–1.67 (m, 2H, CH₂, H-3); 1.70–1.81 (m, 2H, CH₂, H-2); 2.79 (t, 2H, J = 7.3 Hz, CH₂NH, H-4); 2.89 (t, 2H, J = 7.5 Hz, CH₂NH₂, H-1); 4.10 (s, 2H, CH₂NH, H-4); 7.4–7.45 (m, 3H, H-3ʹ, H-4ʹ, H-5ʹ, Ar); 7.58–7.62 (m, 2H, H-2ʹ, H-6ʹ, Ar); 8.19 (br s, 2H, NH₂); 9.57 (br s, 1H, NH). ¹³C-NMR (DMSO-d₆) δ: 22.3 (C-3); 24.0 (C-2); 38.0 (C-4); 45.5 (C-1); 49.7 (ArCH₂NH); 128.5 (C-3ʹ, C-5ʹ, Ar); 128.8 (C-4ʹ, Ar); 130.1 (C-2ʹ, C-6ʹ, Ar); 132.0 (C-1ʹ, Ar).

N-1-(4-Methoxy-phenylmethyl)-butane1,4-diamine hydrochloride (6b). Compound 6b was prepared in 97% yield (2.87 g) from (4-methoxyphenylmethylamino-butyl)-carbamic acid tert-butyl ester 5b (3.37 g, 12.1 mmol) according to the standard procedure as pale brownish powder. Mp > 260 °C. ¹H-NMR (DMSO-d₆) δ: 1.57–1.67 (m, 2H, CH₂, H-3); 1.69–1.84 (m, 2H, CH₂, H-2); 2.79 (t, 2H, J = 8.7 Hz, CH₂NH, H-4); 3.76 (s, 3H, OCH₃); 4.03 (s, 2H, ArCH₂NH); 6.97 (d, 2H, J = 8.7 Hz, H-2ʹ, H-6ʹ, Ar); 7.51 (d, 2H, J = 8.7 Hz, H-3ʹ, H-5ʹ, Ar); 8.13 (br s, 2H, NH₂); 9.41 (br s, 1H, NH). ¹³C-NMR (DMSO-d₆) δ: 22.3 (C-3); 24.0 (C-2); 38.0 (C-4); 45.3 (C-1); 49.7 (ArCH₂NH); 55.2 (OCH₃); 113.9 (C-2ʹ, C-6ʹ, Ar); 123.7 (C-1ʹ, Ar); 131.6 (C-3ʹ, C-5ʹ, Ar); 159.6 (C-4ʹ, Ar).

N-1-(2-Chloro-phenylmethyl)-butane1,4-diamine hydrochloride (6c). Compound 6c was prepared in 98% yield (2.95 g) from (2-chlorophenylmethylamino-butyl)-carbamic acid tert-butyl ester 5c (3.37 g, 12.1 mmol) according to the standard procedure as pale brownish powder. Mp = 206–208 °C. ¹H-NMR (DMSO-d₆) δ: 1.57–1.66 (m, 2H, H-3); 1.69–1.79 (m, 2H, H-2); 2.77–2.84 (m, 4H, H-1, H-4); 3.76 (s, 3H, OCH₃); 4.03 (s, 2H, ArCH₂NH); 6.97 (d, 2H, J = 8.7 Hz, H-2ʹ, H-6ʹ, Ar); 7.51 (d, 2H, J = 8.7 Hz, H-3ʹ, H-5ʹ, Ar); 8.15 (br s, 2H, NH₂); 9.41 (br s, 1H, NH). ¹³C-NMR (DMSO-d₆) δ: 22.3 (C-3); 24.0 (C-2); 38.0 (C-4); 45.3 (C-1); 49.2 (ArCH₂NH); 55.2 (OCH₃); 113.9 (C-2ʹ, C-6ʹ, Ar); 123.7 (C-1ʹ, Ar); 131.6 (C-3ʹ, C-5ʹ, Ar); 159.6 (C-4ʹ, Ar).
(br s, 1H, NH). $^{13}$C-NMR (DMSO-$d_6$) $\delta$: 22.3 (C-3); 24.0 (C-2); 38.0 (C-4); 46.1 (C-1); 46.7 (ArCH$_2$NH); 127.5 (C-5', Ar); 129.5 (C-4', Ar); 129.9 (C-1', Ar); 130.7 (C-6', Ar); 131.9 (C-3', Ar); 133.5 (C-2', Ar).

$N$-1-(3-Methoxy-phenylmethyl)-butane-1,4-diamine hydrochloride (6d). Compound 6d was prepared in 98% yield (2.98 g) from [4-(3-methoxy-phenylamino)-butyl]-carbamic acid tert-butyl ester 5d (3.73 g, 12.1 mmol) according to the standard procedure as pale brownish powder. Mp = 190–192 °C. $^1$H-NMR (DMSO-$d_6$) $\delta$: 1.58–1.67 (m, 2H, H-3); 1.71–1.81 (m, 2H, H-2); 2.78 (t, 2H, $J = 7.3$ Hz, CH$_2$NH, H-4); 2.87 (t, 2H, $J = 7.6$ Hz, CH$_2$NH$_2$, H-1); 3.77 (s, 3H, OCH$_3$); 4.08 (t, 2H, $J = 5.6$ Hz, ArCH$_2$NH); 6.94–6.97 (m, 1H, H-5', Ar); 7.12–7.14 (m, 1H, H-6', Ar); 7.29–7.35 (m, 2H, H-2', H-4', Ar); 8.18 (br s, 2H, NH$_2$); 9.58 (br s, 1H, NH). $^{13}$C-NMR (DMSO-$d_6$) $\delta$: 22.3 (C-3); 24.0 (C-2); 38.0 (C-4); 45.5 (C-1); 49.7 (ArCH$_2$NH); 55.2 (OCH$_3$); 114.5 (C-5', Ar); 115.5 (C-6', Ar); 122.0 (C-4', Ar); 129.6 (C-2', Ar); 133.4 (C-1', Ar); 159.2 (C-3', Ar).

$N$-1-Benzo[1,3]dioxol-5-ylmethyl-butane-1,4-diamine hydrochloride (6e). Compound 6e was prepared in 72% yield (2.25 g) from {4-[(benzo[1,3]dioxol-5-ylmethyl)-amino]-butyl}-carbamic acid tert-butyl ester 5e (3.90 g, 12.1 mmol) according to the standard procedure as pale brownish powder. Mp > 260 °C. $^1$H-NMR (DMSO-$d_6$) $\delta$: 1.58–1.67 (m, 2H, H-3); 1.71–1.81 (m, 2H, H-2); 2.73–2.82 (m, 4H, H-1, H-4); 3.97 (t, 2H, $J = 5.4$ Hz, ArCH$_2$NH); 4.24 (s, 4H, OCH$_2$CH$_2$O); 6.87 (d, 1H, $J = 8.2$ Hz, H-7', Ar); 7.01–7.04 (m, 1H, H-8', Ar); 7.15 (m, 1H, H-5', Ar); 8.18 (br s, 2H, NH$_2$); 9.43 (br s, 1H, NH). $^{13}$C-NMR (DMSO-$d_6$) $\delta$: 22.3 (C-3); 24.0 (C-2); 38.0 (C-4); 45.2 (C-1); 49.4 (ArCH$_2$NH); 101.2 (C-2', Ar); 108.2 (C-6', Ar); 110.4 (C-7', Ar); 124.1 (C-4', Ar); 125.4 (C-5', Ar); 147.2 (C-3a', C-7a', Ar); 147.6 (C-3a', C-7a', Ar).

$N$-1-(2,3-Dihydro-benzo[1,4]dioxin-6-ylmethyl)-butane-1,4-diamine hydrochloride (6f). Compound 6f was prepared in 92% yield (3.04 g) from {4-[(2,3-dihydro-benzo[1,4]dioxin-6-ylmethyl)-amino]-butyl}-carbamic acid tert-butyl ester 5f (4.07 g, 12.1 mmol) according to the standard procedure as pale brownish powder. Mp = 239–241 °C. $^1$H-NMR (DMSO-$d_6$) $\delta$: 1.57–1.66 (m, 2H, H-3); 1.68–1.78 (m, 2H, H-2); 2.73–2.82 (m, 4H, H-1, H-4); 3.97 (t, 2H, $J = 5.4$ Hz, ArCH$_2$NH); 4.24 (s, 4H, OCH$_2$CH$_2$O); 6.87 (d, 1H, $J = 8.2$ Hz, H-7', Ar); 7.01–7.04 (m, 1H, H-8', Ar); 7.15 (m, 1H, H-5', Ar); 8.18 (br s, 2H, NH$_2$); 9.43 (br s, 1H, NH). $^{13}$C-NMR (DMSO-$d_6$) $\delta$: 22.3 (C-3); 24.0 (C-2); 38.0 (C-4); 45.2 (C-1); 49.4 (ArCH$_2$NH); 101.2 (C-2', Ar); 108.2 (C-6', Ar); 110.4 (C-7', Ar); 124.1 (C-4', Ar); 125.4 (C-5', Ar); 147.2 (C-3a', C-7a', Ar); 147.6 (C-3a', C-7a', Ar).

3.1.5. Standard Procedure for the Preparation of N-Substituted 5-Arylidene Rhodanine Derivatives 9a–f under Microwave Irradiation Using “One-Pot Two-steps” Protocol

In a 10 mL glass tube were placed successively bis-(carboxymethyl)trithiocarbonate 7 (0.11 g, 0.50 mol, 1 equiv.), dimethoxylethane (2 mL), triethylamine (135 mL, 101 mg, 1 mmol, 2 equiv. or 202 mL, 152 mg, 1.5 mmol, 3 equiv.) and hydrochloride salt 6 (0.5 mmol, 1 equiv.). The glass tube was sealed with a snap cap and placed in the Monowave® 300 Anton Paar microwave cavity ($p = 800$ Watt). The reaction mixture was irradiated at 90 °C for 15–60 min. under vigorous magnetic stirring. After microwave dielectric heating, the crude reaction mixture was allowed to cool down at room temperature, aldehyde 3 (0.5 mol, 1 equiv.) was added to the cooled reaction mixture which was immediately
submitted to microwave irradiation at 110 °C for 5–30 min. After cooling down to room temperature, the volatile compounds of the reaction mixture were removed in a rotary evaporator under reduced pressure. To the crude reaction mixture was added 4 mL of MeOH and after triturating, the insoluble product 9 was collected by filtration on a Büchner funnel (porosity N°4), washed with deionized water (5 mL), triturated and mixing with ethanol (3 × 5 mL) during 3 h, washed successively with ethanol (5 mL) and anhydrus ether (5 × 2 mL), then dried under high vacuum (10⁻² Torr) at 25 °C for 1 h. The desired compound 9 was eventually purified by recrystallization in EtOH after control by ¹H-NMR in solution of CDCl₃/TFA (98:2).

(5Z)3-[4-(4-Methoxyphenylmethylamino)butyl]-5-(1,3-benzodioxol-5-ylmethylene)-2-thioxo-1,3-thiazolidin-4-one (9a). According to the standard procedure, compound 9a was prepared in 10% yield (23 mg) from N-1-(4-methoxy-phenylmethyl)-butane-1,4-diamine hydrochloride 6b (122.3 mg, 0.5 mmol, 1 equiv.), triethylamine (202 µL, 152 mg, 1.5 mmol, 3 equiv.) and piperonaldehyde 3e (75.1 mg, 0.5 mmol, 1 equiv.) after 30 min. at 90 °C, followed by a second reaction time of 30 min. at 110 °C (for condensation step with 3e), which gave 9a as a yellowish powder. Mp = 170–229 °C (decomposition). ¹H-NMR (CDCl₃/TFA 98:2) δ: 1.61–1.69 (m, 4H, CH₂, H-2”, H-3”); 3.05–3.11 (m, 2H, ArCH₂NH); 3.73 (s, 3H, OCH₃); 4.00–4.08 (m, 4H, CH₂, H-1”); 5.97 (s, 2H, CH₂, H-2’); 6.80–7.16 (m, 7H, H-2””, H-3””, H-5””, H-6””, H-4’, H-6’, H-7’, Ar); 7.55 (br s, 1H, NH); 7.61 (s, 1H, HC=C). ¹³C-NMR (CDCl₃/TFA 98:2) δ: 23.1 (C-2”, 3”); 23.8 (C-3”, 2’”); 44.2 (C-4”, C-1’’’); 47.0 (ArCH₂NH); 52.1 (C-1’”’); 55.5 (O(CH₃)); 102.2 (C-2’’’); 109.4 (C-2’’’, C-6’’); 109.5 (C-3’”, C-5’’’); 115.0 (C-6’); 120.0 (C-1’”’); 127.5 (C=CH); 128.2 (C-7’); 131.2 (C-4’); 135.0 (C=CH); 148.8 (C-5’’); 148.9 (C-4’’’); 150.6 (C-3’a); 150.9 (C-7’a); 169.4 (C=O, C-4’); 192.9 (C=S, C-2). HRMS, m/z: 457.1256 found (calculated for C₂₃H₂₈N₂O₃S₂ [M + H]+ requires 457.1253).

(5Z)3-[4-(4-Methoxyphenylmethylamino)butyl]-5-(4-methoxybenzylidene)-2-thioxo-1,3-thiazolidin-4-one (9b). According to the standard procedure, compound 9b was prepared in 6% yield (13.3 mg) from N-1-(4-methoxy-phenylmethyl)-butane-1,4-diamine hydrochloride 6b (122.3 mg, 0.5 mmol, 1 equiv.), triethylamine (202 µL, 152 mg, 1.5 mmol, 3 equiv.) and 4-methoxybenzaldehyde 3b (68.1 mg, 0.5 mmol, 1 equiv.) after 30 min. at 90 °C, followed by a second reaction time of 30 min. at 110 °C (for condensation step with 3b), which gave 9b as a yellowish powder. Mp = 253–255 °C. ¹H-NMR (CDCl₃/TFA 98:2) δ: 1.67–1.73 (m, 4H, CH₂, H-2’, H-3’); 3.07–3.13 (m, 2H, ArCH₂NH); 3.81 (s, 6H, ArOCH₃); 4.04–4.11 (s, 4H, CH₂, H-1”, H-4”); 6.94 (dd, 4H, J = 8.8 Hz, H-2’, H-6’, H-2’”, H-6’”, Ar); 7.40 (d, 4H, J = 8.8 Hz, H-3’, H-5’, H-3’”, H-5’’”; Ar); 7.60 (br s, 1H, NH); 7.67 (s, 1H, CH=). ¹³C-NMR (CDCl₃/TFA 98:2) δ: 24.3 (C-2’’, C-3’’); 44.0 (C-1’, C-4’’); 46.8 (ArCH₂NH); 55.6 (C-4’, OCH₃, C-4’’’, OCH₃); 115.1 (C-2’, C-6’, C-2’”, C-6’”); 119.6 (C-1’, C-1’’’); 126.0 (C=); 133.1 (C-3’, C-5’, C-3’’, C-5’’’); 130.5 (C-3’, C-3’’’); 134.6 (CH=); 162.0 (C-4’, C-4’’’); 169.0 (C=O, C-4’); 193.2 (C=S, C-2). HRMS, m/z: 443.1463 found (calculated for C₂₃H₂₇N₂O₃S₂ [M + H]+ requires 443.1460).

(5Z)3-[4-(4-Methoxyphenylmethylamino)butyl]-5-(2,3-dihydro-1,4-benzodioxin-6-ylmethylene)-2-thioxo-1,3-thiazolidin-4-one (9c). According to the standard procedure, compound 9c was prepared in 5% yield (11.8 mg) from N-1-(4-methoxy-phenylmethyl)-butane-1,4-diamine hydrochloride 6b (122.3 mg, 0.5 mmol, 1 equiv.), triethylamine (202 µL, 152 mg, 1.5 mmol, 3 equiv.) and 2,3-dihydro-1,4-benzodioxin-
6-carboxaldehyde 3f (82.1 mg, 0.5 mmol, 1 equiv.) after 30 min. at 90 °C, followed by a second reaction time of 30 min. at 110 °C (for condensation step with 3f), which gave 9e as a yellowish powder. Mp = 255–261 °C. 1H-NMR (CDCl3/TFA 98:2) δ: 1.68 (s, 4H, CH2, H-2′, H-3′); 3.03–3.08 (m, 2H, ArCH2NH); 3.73 (s, 3H, OCH3); 4.07 (s, 4H, CH2, H-1″, H-4″); 4.20–4.25 (m, 2H, CH2, H-2′, H-3′); 6.84–6.97 (m, 7H, H-5′, H-7′, H-8′, H-2″′, H-3″′, H-5″′, H-6″′, Ar); 7.58 (s, 1H, CH=); 7.60 (br s, 1H, NH). 13C-NMR (CDCl3/TFA 98:2) δ: 24.2 (C-2″, C-3″); 44.1 (C-4″, C-1″); 47.1 (ArCH2NH); 55.0 (OCH3); 64.2 (C-2′); 64.8 (C-3′); 118.4 (C-5′, C-7′, C-8′); 119.7 (C-2″′, C-6″′); 120.3 (C=); 125.7 (C-3″′, C-5″′); 126.8 (C-6′); 135.0 (CH=); 143.9 (C-8′a); 146.7 (C-4′a); 169.5 (C=O, C-4); 193.2 (C=S, C-2). HRMS, m/z: 471.1412 found (calculated for C24H27N2O4S2 [M + H]+ requires 471.1415).

(5Z)-3-[4-(4-Methoxyphenylmethylamino)butyl]-5-(4-hydroxy-3-methoxybenzyliden)-2-thioxo-1,3-thiazolidin-4-one (9d). According to the standard procedure, compound 9d was prepared in 30% yield (68.8 mg) from N-1-(4-methoxy-phenylmethyl)-butane-1,4-diamine hydrochloride 6b (122.3 mg, 0.5 mmol, 1 equiv.), triethylamine (202 µL, 152 mg, 1.5 mmol, 3 equiv.) and 4-hydroxy-3-methoxybenzaldehyde 3g (76.1 mg, 0.5 mmol, 1 equiv.) after 30 min. at 90 °C, followed by a second reaction time of 30 min. at 110 °C (for condensation step with 3g), which gave 9d as a yellowish powder. Mp = 191–193 °C. 1H-NMR (CDCl3/TFA 98:2) δ: 1.69 (m, 4H, CH2, H-2′, H-3′); 3.06–3.07 (m, 2H, ArCH2NH); 3.72 (s, 3H, C-4″′, OCH3); 3.86 (s, 3H, C-3′, OCH3); 4.04–4.08 (m, 4H, CH2, H-1″, H-4″); 6.81–7.17 (m, 7H, H-2′, H-5′, H-6′, H-2″′, H-3″′, H-5″′, H-6″′, Ar); 7.55 (br s, 1H, NH); 7.60 (s, 1H, CH=). 13C-NMR (CDCl3/TFA 98:2) δ: 23.1 (C-2″′, C-3″′); 23.9 (C-2″′′, C-3″′); 43.1 (C-1″′, C-4″′); 47.0 (ArCH2NH); 55.5 (ArOCH3); 56.1 (ArOCH3); 112.6 (C-6′); 115.0 (C-2″′′, C-6″′); 115.6 (C-2′); 118.9 (C-1″′′); 121.2 (C=); 125.9 (C-1′); 126.8 (C-5′); 131.2 (C-3″′, C-5″′); 135.9 (CH=); 147.2 (C-4″′); 148.9 (C-3′); 160.8 (C-4′); 169.4 (C=O, C-4); 193.1 (C=S, C-2). HRMS, m/z: 459.1412 found (calculated for C23H22N2O4S2 [M + H]+ requires 459.1416).

(5Z)-3-[4-(3-Methoxyphenylmethylamino)butyl]-5-(1,3-benzodioxol-5-ylmethylene)-2-thioxo-1,3-thiazolidin-4-one (9e). According to the standard procedure, compound 9e was prepared in 21% yield (48.0 mg) from N-1-(3-methoxy-phenylmethyl)-butane-1,4-diamine hydrochloride 6d (122.3 mg, 0.5 mmol, 1 equiv.), triethylamine (202 µL, 152 mg, 1.5 mmol, 3 equiv.) and piperonaldehyde 3e (75.1 mg, 0.5 mmol, 1 equiv.) after 15 min. at 90 °C, followed by a second reaction time of 15 min. at 110 °C (for condensation step with 3e), which gave 9e as a yellowish powder. Mp = 178–246 °C (decomposition). 1H-NMR (CDCl3/TFA 98:2) δ: 1.82 (s, 4H, CH2, H-2″, H-3″); 3.19–3.22 (m, 2H, ArCH2NH); 3.83 (s, 3H, OCH3); 4.13–4.23 (m, 4H, CH2, H-1″, H-4″); 6.10 (s, 2H, CH2, H-2″); 6.92–7.28 (m, 6H, H-2″′, H-4″′, H-6″′, H-7″, H-8″, Ar); 7.33–7.38 (m, 1H, H-5″′, Ar); 7.55 (br s, 1H, NH); 7.70 (s, 1H, CH=). 13C-NMR (CDCl3/TFA 98:2) δ: 23.1 (C-2″′, C-3″′); 23.9 (C-3″′, C-2″′); 42.9 (C-4″′, C-1″′); 46.8 (ArCH2NH); 52.1 (C-1″′, C-4″′); 55.3 (OCH3); 102.1 (C-2″); 109.3 (C-6″′); 109.4 (C-5″′); 114.9 (C-4″′); 115.9 (C-2″′′); 119.8 (C-1″′′); 121.8 (C-6″); 127.4 (C=); 127.6 (C-5″); 127.9 (C-7″); 130.7 (C-4″′); 134.5 (CH=); 148.8 (C-7″a); 150.5 (C-3″a); 160.3 (C-3″′); 168.5 (C=O, C-4); 193.1 (C=S, C-2). HRMS, m/z: 457.1259 found (calculated for C23H25N2O4S2 [M + H]+ requires 457.1257).

(5Z)-3-[4-(3-Methoxyphenylmethylamino)butyl]-5-(2,3-dihydro-1,4-benzodioxin-6-ylmethylene)-2-thioxo-1,3-thiazolidin-4-one (9f). According to the standard procedure, compound 9e was prepared in 9%
yield (21.2 mg) from N-I-(3-methoxy-phenylmethyl)-butane-1,4-diamine hydrochloride 6d (122.3 mg, 0.5 mmol, 1 equiv.), triethylamine (202 μL, 152 mg, 1.5 mmol, 3 equiv.) and 2,3-dihydro-1,4-benzodioxin-6-carboxaldehyde 3f (82.1 mg, 0.5 mmol, 1 equiv.) after 15 min. at 90 °C, followed by a second reaction time of 15 min. at 110 °C (for condensation step with 3f), which gave 9f as a yellowish powder. Mp = 247–251 °C. 1H-NMR (CDCl3/TFA 98:2) δ: 1.84 (s, 4H, CH2, H-2″, H-3″); 3.04–3.12 (m, 2H, ArCH2NH); 3.95 (s, 3H, OCH3); 4.23 (s, 4H, CH2, H-1″, H-4″); 4.37–4.40 (m, 4H, H-2′, H-3′); 6.99–7.28 (m, 6H, H-2″”, H-4″”, H-5″”, H-6″”, H-7′, H-8′, Ar); 7.60 (br s, 1H, NH); 7.74 (s, 1H, CH=). 13C-NMR (CDCl3/TFA 98:2) δ: 24.1 (C-2″, C-3″); 44.1 (C-1″, C-4″); 47.3 (ArCH2NH); 64.2 (C-2″); 68.4 (C-3″); 118.5 (C-5′, C-7′, C-8″); 119.8 (C-4″”, C-6″”); 125.9 (C-2″”, C-5″”); 126.8 (C=); 135.3 (CH=); 143.8 (C-8′a); 146.6 (C-4′a); 168.5 (C=O, C-4); 193.3 (C=S, C-2). HRMS, m/z: 471.1412 found (calculated for C24H27N2O4S2 [M + H]+ requires 471.1415).

(5Z)-3-[4-(3-Methoxyphenethylamino)butyl]-5-(3-methoxybenzylidene)-2-thioxo-1,3-thiazolidin-4-one (9g). According to the standard procedure, compound 9g was prepared in 5% yield (11.1 mg) from N-I-(3-methoxy-phenylmethyl)-butane-1,4-diamine hydrochloride 6d (122.3 mg, 0.5 mmol, 1 equiv.), triethylamine (202 μL, 152 mg, 1.5 mmol, 3 equiv.) and 3-methoxybenzaldehyde 3d (81.8 mg, 0.5 mmol, 1 equiv.) after 30 min. at 90 °C, followed by a second reaction time of 30 min at 110 °C (for condensation step with 3d), which gave 9g as a yellowish powder. Mp = 225–227 °C. 1H-NMR (CDCl3/TFA 98:2) δ: 1.73 (s, 4H, CH2, H-2″, H-3″); 3.06–3.10 (m, 2H, ArCH2NH); 3.80 (s, 6H, ArOCH3); 4.11 (s, 4H, CH2, H-1″, H-4″), 6.93–6.96 (m, 4H, H-2″”, H-6″”, H-7′, H-8′, Ar); 7.04 (d, 2H, J = 7.7 Hz, H-4′, H-4″”, Ar), 7.32 (m, 2H, H-5′, H-5″”, Ar), 7.55 (br s, 1H, NH); 7.66 (s, 1H, CH=). 13C-NMR (CDCl3/TFA 98:2) δ: 24.3 (C-2″”, C-3″); 44.0 (C-1″”, C-4″”); 47.1 (ArCH2NH); 55.5 (ArOCH3); 115.3 (C-6′”, C-6″”); 117.3 (C-2′, C-2″”); 123.1 (C=); 123.6 (C-4′, C-4″”); 130.5 (C-5′, C-5″”); 134.0 (CH=); 134.5 (C-1′, C-1″”); 160.0 (C-3′’, C-3″’); 168.5 (C=O, C-4); 193.3 (C=S, C-2). HRMS, m/z: 443.1463 found (calculated for C23H27N2O3S2 [M + H]+ requires 443.1465).

(5Z)-3-[4-(1,3-Benzodioxol-5-ylmethylamino)butyl]-5-(1,3-benzodioxol-5-ylmethylene)-2-thioxo-1,3-thiazolidin-4-one (9h). According to the standard procedure, compound 9h was prepared in 59% yield (139 mg) from N-I-benzol[1,3]dioxol-5-ylmethyl-butane-1,4-diamine hydrochloride 6e (129.4 mg, 0.5 mmol, 1 equiv.), triethylamine (135 μL, 101 mg, 1 mmol, 2 equiv.) and piperonaldehyde 3e (75.1 mg, 0.5 mmol, 1 equiv.) after 15 min. at 90 °C, followed by a second reaction time of 15 min. at 110 °C (for condensation step with 3e), which gave 9h as a yellowish powder. Mp = 245–256 °C. 1H-NMR (CDCl3/TFA 98:2) δ: 1.74 (s, 4H, CH2, H-2″, H-3″); 3.04–3.05 (m, 2H, ArCH2NH); 4.05–4.07 (m, 4H, H-2″”, H-3″”, H-4″”, H-6″”; Ar); 7.58 (s, 1H, CH=); 7.65 (br s, 1H, NH). 13C-NMR (CDCl3/TFA 98:2) δ: 23.1 (C-2″”, C-3″”); 24.0 (C-3″); 43.1 (C-1″”, C-4″”); 46.5 (ArCH2NH); 51.9 (C-4″”, C-1″”); 101.8 (C-2′, C-2″”); 102.2 (C-2″”, C-2″); 109.0 (C-6″”); 109.3 (C-6′”); 109.4 (C-4″”, C-7′”); 109.8 (C-7′); 119.8 (C-5″”); 122.6 (C=); 124.3 (C-4″”); 127.4 (C-4′); 128.1 (C-5′); 134.9 (CH=); 148.6 (C-7′a); 148.9 (C-3′a); 149.2 (C-7′′a); 150.6 (C-3″a); 168.9 (C=O, C-4); 193.0 (C=S, C-2). HRMS, m/z: 471.1048 found (calculated for C23H23N2O3S2 [M + H]+ requires 471.1049).
(5Z)-3-[4-(1,3-Benzodioxol-5-ylmethylamino)butylo]-5-(2,3-dihydro-1,4-benzodioxin-6-ylmethylen)-2-thioxo-1,3-thiazolidin-4-one (9i). According to the standard procedure, compound 9i was prepared in 59% yield (143 mg) from N-1-benzo[1,3]dioxol-5-ylmethyl-butane-1,4-diamine hydrochloride 6e (129.4 mg, 0.5 mmol, 1 equiv.), triethylamine (135 µL, 101 mg., 1 mmol, 2 equiv.) and 2,3-dihydro-1,4-benzodioxin-6-carboxaldehyde 3f (82.1 mg, 0.5 mmol, 1 equiv.) after 15 min. at 90 °C, followed by a second reaction time of 15 min. at 110 °C (for condensation step with 3f), which gave 9i as a yellowish powder. Mp = 253–257 °C. ¹H-NMR (CDCl₃/TFA 98:2) δ: 1.68 (s, 4H, CH₂, H-2”, H-3”); 3.05–3.06 (m, 2H, ArCH₂NH); 4.03–4.04 (m, 4H, CH₂, H-1”, H-4”); 4.20–4.25 (m, 4H, CH₂, H-2’, H-3’); 5.89 (s, 2H, CH₂, H-2’’); 6.70 (s, 3H, H-4’”, H-6’”, H-7’”, Ar); 6.85–6.96 (m, 3H, H-5’, H-7’, H-8’, Ar); 7.50 (br s, 1H, NH); 7.55 (s, 1H, CH=). ¹³C-NMR (CDCl₃/TFA 98:2) δ: 23.1 (C-2’, C-3’); 23.8 (C-3’, C-2’’); 43.1 (C-1’, C-4’); 47.0 (ArCH₂NH); 52.4 (C-4’’, C-1’’); 64.2 (C-2’, C-3’); 64.8 (C-3’, C-2’’); 101.8 (C-2’’’); 109.1 (C-6’’’); 109.5 (C-7’’’); 118.5 (C-7’); 119.7 (C-5’’); 119.8 (C-8’); 122.3 (C=); 124.1 (C-4’’’); 125.9 (C-5’); 126.6 (C-6’); 135.5 (CH=); 144.0 (C-7’’’a); 146.9 (C-3’’’a); 148.6 (C-8’a); 149.4 (C-4’a); 169.5 (C=O, C-4’); 193.2 (C=S, C-2). HRMS, m/z: 485.1205 found (calculated for C₂₄H₂₅N₂O₄S₂ [M + H]+ requires 485.1203).

(5Z)-3-[4-(1,3-Benzodioxol-5-ylmethylamino)butylo]-5-(4-hydroxy-benzylidene)-2-thioxo-1,3-thiazolidin-4-one (9j). According to the standard procedure, compound 9j was prepared in 34% yield (75.2 mg) from N-1-benzo[1,3]dioxol-5-ylmethyl-butane-1,4-diamine hydrochloride 6e (129.4 mg, 0.5 mmol, 1 equiv.), triethylamine (135 µL, 101 mg, 1 mmol, 2 equiv.) and 4-hydroxybenzaldehyde 3h (61.1 mg, 0.5 mmol, 1 equiv.) after 15 min. at 90 °C, followed by a second reaction time of 15 min. at 110 °C (for condensation step with 3h), which gave 9j as an orange powder. Mp = 93–124 °C (decomposition). ¹H-NMR (CDCl₃/TFA 98:2) δ: 1.59–1.73 (m, 4H, CH₂, H-2”, H-3”); 3.01–3.13 (m, 2H, ArCH₂NH); 4.00–4.05 (m, 4H, CH₂, H-1”, H-4”); 5.88 (s, 2H, CH₂, H-2’’’); 6.66–6.69 (m, H-4’’, H-6’’, H-7’’, Ar); 6.86 (dd, 2H, J = 8.7 Hz, H-2’, H-6’, Ar); 7.32 (dd, 2H, J = 8.7 Hz, H-3’, H-5’, Ar); 7.55 (br s, 1H, NH); 7.60 (s, 1H, CH=, 1H). ¹³C-NMR (CDCl₃/TFA 98:2) δ: 22.5 (C-2’, C-3’); 23.2 (C-3’, C-2’’); 42.5 (C-1”, C-4’’); 46.4 (ArCH₂NH); 51.9 (C-4’’, C-1’’); 101.2 (C-2’’’); 108.5 (C-6’’’); 108.8 (C-7’’’); 116.1 (C-2’, C-6’); 118.6 (C-5’’’); 121.7 (C=); 123.4 (C-4’’’); 125.6 (C-1’); 132.9 (C-3’, C-5’); 134.8 (CH=); 148.0 (C-7’’’a); 148.8 (C-3’’’a); 157.6 (C-4’); 169.0 (C=O, C-4’); 192.6 (C=S, C-2). HRMS, m/z: 443.1099 found (calculated for C₂₂H₂₃N₂O₄S₂ [M + H]+ requires 443.1095).

(5Z)-3-[4-(1,3-Benzodioxol-5-ylmethylamino)butylo]-5-(4-hydroxy-3-methoxybenzylidene)-2-thioxo-1,3-thiazolidin-4-one (9k). According to the standard procedure, compound 9k was prepared in 46% yield (109 mg) from N-1-benzo[1,3]dioxol-5-ylmethyl-butane-1,4-diamine hydrochloride 6e (129.4 mg, 0.5 mmol, 1 equiv.), triethylamine (202 µL, 152 mg, 1.5 mmol, 3 equiv.) and 4-hydroxy-3-methoxybenzaldehyde 3g (76.1 mg, 0.5 mmol, 1 equiv.) after 30 min. at 90 °C, followed by a second reaction time of 30 min. at 110 °C (for condensation step with 3g), which gave 9k as a red powder. Mp = 200–203 °C. ¹H-NMR (CDCl₃/TFA 98:2) δ: 1.71 (s, 4H, CH₂, H-2”, H-3”); 3.05–3.06 (m, 2H, ArCH₂NH); 3.88 (s, 3H, OCH₃); 4.04 (s, 4H, CH₂, H-1”, H-4”); 5.91 (s, 2H, CH₂, H-2’’’); 6.74 (s, 3H, H-4’’’, H-6’’’, H-7’’’, Ar); 6.87–7.02 (m, 3H, H-2’, H-5’, H-6’, Ar); 7.58 (s, 1H, CH=); 7.65 (br s, 1H, NH). ¹³C-NMR (CDCl₃/TFA 98:2) δ: 23.1 (C-2’, C-3’); 23.9 (C-3’, C-2’’); 42.9 (C-1”, C-4’’); 46.6 (ArCH₂NH); 52.1 (C-4’’, C-1’’); 56.1 (OCH₃); 101.8 (C-2’’’); 109.0 (C-6’’’); 109.6 (C-7’’’); 112.3
(C-6); 115.5 (C-2’); 119.1 (C-5’’); 122.7 (C=); 124.0 (C-5’); 125.8 (C-1’); 126.5 (C-4’’); 134.9 (C=H); 147.1 (C-3’); 148.6 (C-7’’a); 148.8 (C-4’); 149.2 (C-3’’a); 168.5 (C=O, C-4’’); 193.2 (C=S, C-2). HRMS, m/z: 473.1205 found (calculated for C22H25N2O5S2 [M + H]+ requires 473.1206).

(5Z)-3-[4-(2,3-Dihydro-benzo[1,4]dioxin-6-ylmethylamino)butyl]-5-(1,3-benzodioxol-5-ylmethylene)-2-thioxo-1,3-thiazolidin-4-one (9l). According to the standard procedure, compound 9l was prepared in 15% yield (36.3 mg) from N-1-(2,3-dihydro-benzo[1,4]dioxin-6-ylmethyl)-butane-1,4-diamine hydrochloride 6f (136.4 mg, 0.5 mmol, 1 equiv.), triethylamine (202 μL, 152 mg, 1.5 mmol, 3 equiv.) and piperonaldehyde 3e (75.1 mg, 0.5 mmol, 1 equiv.) after 15 min. at 90 °C, followed by a second reaction time of 15 min. at 110 °C (for condensation step with 3e), which gave 9l as a yellowish powder. Mp = 172–233 °C (decomposition). 1H-NMR (CDCl3/TFA 98:2) δ: 1.66–1.69 (m, 4H, CH2, H-2’’, H-3’’); 3.03–3.10 (m, 2H, ArCH2NH); 4.03–4.07 (m, 4H, CH2, H-1’, H-4’’); 4.16–4.19 (m, 4H, CH2, H-2’’, H-3’’); 5.97 (s, 2H, CH2, H-2’); 6.66–7.00 (m, 6H, H-4’, H-6’, H-7’, H-5’’, H-7’’, H-8’’, Ar); 7.55 (br s, 1H, NH); 7.59 (s, 1H, C=H). 13C-NMR (CDCl3/TFA 98:2) δ: 23.8 (C-2’’, C-3’’); 43.1 (C-1’’, C-4’’); 47.0 (ArCH2NH); 52.1 (C-4’’, C-1’’); 64.2 (C-2’’); 64.4 (C-3’’); 102.2 (C-2’); 109.4 (C-7’’); 109.5 (C-8’’); 118.5 (C-5’’’); 119.5 (C-6’’’); 122.0 (C=); 122.9 (C-6’); 127.3 (C-5’); 128.2 (C-7’); 135.0 (C-4’’); 135.5 (CH=); 143.9 (C-4’’’a); 145.1 (C-8’’’a); 150.6 (C-3’a); 150.8 (C-7’a); 168.5 (C=O, C-4’’); 193.0 (C=S, C-2). HRMS, m/z: 485.1205 found (calculated for C24H25N2O5S2 [M + H]+ requires 485.1206).

(5Z)-3-[4-(2,3-Dihydro-benzo[1,4]dioxin-6-ylmethylamino)butyl]-5-(2,3-dihydro-benzo[1,4]dioxin-6-ylmethylene)-2-thioxo-1,3-thiazolidin-4-one (9m). According to the standard procedure, compound 9m was prepared in 7% yield (17.5 mg) from N-1-(2,3-dihydro-benzo[1,4]dioxin-6-ylmethyl)-butane-1,4-diamine hydrochloride 6f (136.4 mg, 0.5 mmol, 1 equiv.), triethylamine (202 μL, 152 mg, 1.5 mmol, 3 equiv.) and 2,3-dihydro-1,4-benzo[dioxin-6-carboxaldehyde 3f (82.1 mg, 0.5 mmol, 1 equiv.) after 15 min. at 90 °C, followed by a second reaction time of 15 min. at 110 °C (for condensation step with 3f), which gave 9m as a yellowish powder. Mp = 170–243 °C (decomposition). 1H-NMR (CDCl3/TFA 98:2) δ: 1.64–1.69 (m, 4H, CH2, H-2’’, H-3’’); 3.04–3.10 (m, 2H, ArCH2NH); 4.03–4.08 (m, 4H, CH2, H-1’, H-4’’); 4.16–4.23 (m, 8H, CH2, H-2’’, H-3’’, H-4’’); 6.75–7.57 (m, 6H, H-5’, H-7’, H-8’, H-5’’, H-7’’, H-8’’, Ar); 7.60 (br s, 1H, NH); 8.77 (s, 1H, C=H). 13C-NMR (CDCl3/TFA 98:2) δ: 24.2 (C-2’’, C-3’’); 44.0 (C-1’’, C-4’’); 64.2 (C-2’’, C-2’’’); 64.8 (C-3’, C-3’’); 118.4 (C-7’, C-7’’’’’); 119.6 (C-8’, C-8’’’’’); 120.3 (C=); 125.6 (C-5’, C-5’’’’); 126.8 (C-6’, C-6’’’’); 134.5 (CH=); 144.0 (C-8’a, C-8’’’’a); 146.6 (C-4’a, C-4’’’’a); 166.6 (C=O, C-4’’); 182.5 (C=S, C-2). HRMS, m/z: 499.1364 found (calculated for C25H23N2O5S2 [M + H]+ requires 499.1366).

(5Z)-3-[4-(2,3-Dihydro-benzo[1,4]dioxin-6-ylmethylamino)butyl]-5-(4-hydroxy-3-methoxybenzylidene)-2-thioxo-1,3-thiazolidin-4-one (9n). According to the standard procedure, compound 9n was prepared in 34% yield (82.7 mg) from N-1-(2,3-dihydro-benzo[1,4]dioxin-6-ylmethyl)-butane-1,4-diamine hydrochloride 6f (136.4 mg, 0.5 mmol, 1 equiv.), triethylamine (202 μL, 152 mg, 1.5 mmol, 3 equiv.) and 4-hydroxy-3-methoxybenzaldehyde 3g (76.1 mg, 0.5 mmol, 1 equiv.) after 30 min. at 90 °C, followed by a second reaction time of 30 min. at 110 °C (for condensation step with 3g), which gave 9n as a red powder. Mp = 202–204 °C. 1H-NMR (CDCl3/TFA 98:2) δ: 1.69 (s, 4H, CH2, H-2’’, H-3’’); 3.06 (s, 2H, ArCH2NH);
3.86 (s, 3H, OCH₃); 4.03 (s, 4H, CH₂, H-1‴, H-4‴); 4.16 (s, 4H, CH₂, H-2‴‴, H-3‴‴); 6.67–7.03 (m, 6H, H-2‴, H-5‴, H-6‴, H-5‴‴, H-7‴‴, H-8‴‴, Ar); 7.60 (s, 1H, CH=). ¹³C-NMR (CDCl₃/TFA 98:2) δ: 23.1 (C-2‴ or 3‴); 23.8 (C-3‴ ou C-2‴); 43.1 (C-1‴ ou 4‴); 47.0 (ArCH₂NH); 52.1 (C-4‴, C-1‴); 56.1 (OCH₃); 64.4 (C-2‴‴, C-3‴‴); 64.4 (C-3‴‴, C-2‴‴); 112.6 (C-6‴); 115.6 (C-2‴); 118.4 (C-7‴‴); 118.6 (C-8‴‴); 119.0 (C-6‴‴); 122.0 (C=); 122.9 (C-5‴); 125.9 (C-1‴); 126.8 (C-5‴); 135.9 (CH=); 143.9 (C-8‴‴a); 145.1 (C-3’a); 147.2 (C-4‴‴a); 148.9 (C-4’);169.4 (C=O, C-4); 193.1 (C=S, C-2). HRMS, m/z: 487.1361 found (calculated for C₂₄H₂₇N₂O₅S₂ [M + H]+ requires 487.1359).

3.2. Biochemistry Section

3.2.1. Protein Kinase Assay Buffers

Buffer A: 10 mM MgCl₂, 1 mM EGTA, 1 mM DTT, 25 mM Tris-HCl pH 7.5, 50 µg heparin/mL.

Buffer B: 60 mM β-glycerophosphate, 15 mM p-nitrophenyl-phosphate, 25 mM Mops (pH 7.2), 5 mM EGTA, 15 mM MgCl₂, 1 mM DTT, 1 mM sodium vanadate, 1 mM phenylphosphate.

3.2.2. Kinase Preparations and Assays

Kinase activities for each enzyme were assayed in buffer A (25 mM Tris-HCl pH 7.5, 10 mM MgCl₂, 1 mM EGTA, 1 mM DTT, 50 µg/mL Heparin, BSA 0.15 mg/mL) or B (60 mM β-glycerophosphate, 30 mM p-Nitrophenylphosphate, 25 mM MOPS, 5 mM EGTA, 15 mM MgCl₂, 1 mM DTT, 0.1 mM Na vanadate), with their corresponding substrates, in the presence of 15 µM [γ-³²P] ATP (3000 Ci/mmol; 10 mCi/mL) in a final volume of 30 µL. After 30 min incubation at 30 °C, the reaction was stopped by harvesting, using a FilterMate harvester (Packard, Meriden, CT, USA), onto P81 phosphocellulose papers (GE Healthcare, Velizy-Villacoublay, France) which were washed in 1% phosphoric acid. Scintillation fluid was added and the radioactivity measured in a Packard counter. Blank values were subtracted and activities calculated as pmoles of phosphate incorporated during the 30 min incubation. The activities were expressed in a percentage of the maximal activity, i.e., in the absence of inhibitors. Controls were performed with appropriate dilutions of DMSO. CK1 and Haspin peptide substrates were obtained from Proteogenix (Oberhausbergen, France).

CDK5/p25 (human, recombinant) was prepared as previously described [31]. Its kinase activity was assayed in buffer B, with 1 mg histone H1/mL.

Casein kinase 1 (CK1δ/ε) (porcine brain, native) was assayed with 0.67 µg of CKS peptide (RRKHAAlIGpSAYsITA) [32].

GSK-3α/β (porcine brain, native) was assayed, as described for CDK5/p25 but in Buffer A and using a GSK-3 specific substrate (GS-1: YRRAAVPPSLSRHSSPHQSpEDEEE) (pS stands for phosphorylated serine) [33]. GS-1 was synthesized by Millegen (Labege, France).

Haspin kinase domain (HsHaspin-kd aa 470 to 798) encoding cDNA, obtain by RT-PCR, was cloned into pGex-6P-3. The fusion protein was expressed in Escherichia coli strain BL21-KRX (Promega, Madison, WI, USA) and purified by affinity chromatography on glutathione-agarose beads (Sigma). Haspin-kd activity was assayed with 3 µM Histone H3 (1-21) peptide, a specific Haspin substrate, (ARTKQTARKSTGGKAPRKQLA), in Buffer H (MOPS 25 mM pH 7.5; 10 mM MgCl₂).
3.3. Cell Culture and Survival Assays

Skin diploid fibroblastic cells were provided by BIOPREDIC International Company (Rennes, France). Caco2 (Ref ECACC: 86010202), Huh-7D12 (Ref ECACC: 01042712), MDA-MB-231 (Ref ECACC: 92020424), HCT-116 (Ref ECACC: 91091005), PC3 (Ref ECACC: 90112714), NCI-H727 (Ref ECACC: 94060303) cell lines were obtained from the ECACC collection and HaCaT (from Cell Lines Service, Eppelheim, Germany). Cells were grown according to ECACC recommendations [34]. The toxicity test of the compounds on these cells was as follows: 2 × 10³ cells for HCT-116 cells or 4 × 10³ for the other cells were seeded in 96 multiwell plates in triplicate and left for 24 h for attachment, spreading and growing. Then, cells were exposed for 48 h to increasing concentrations of the compounds, ranging from 0.1 to 25 μM in a final volume of 120 μL of culture medium. Cells were fixed in cooled solution of 90% ethanol/5% acetic acid, nuclei were stained with Hoechst 3342 (Sigma) and counted using automated imaging analysis (Cellomics Arrayscan VTI/HCS Reader, Thermo/Scientific, Waltham, MA, USA). The IC₅₀ were graphically determined.

4. Conclusions

In summary, we have developed, in this preliminary project, a new route to (5Z) 3-(4-arylmethylamino) butyl-5-arylidene-2-thioxo-1,3-thiazolidine-4-ones 9. Starting from commercial butane-1,4-diamine, the process involved six steps and the key step is a solution phase “one-pot two-steps” approach for the construction of the 2-thioxo-1,3-thiazolidine-4-one platform under microwave dielectric heating followed by Knoevenagel condensation for installation of the 5-arylidene moiety as second point of diversity. This methodology offered the possibility of preparing a library of fourteen new compounds in moderate to good yields and, the targeted compounds 9a–n have been built with a Z-geometry. The in vitro inhibition of cell proliferation was carried out on a panel of seven representative tumoral cell lines and the compounds 9a–n were also evaluated against four protein kinases. Among all of these compounds, the compound 9j turned out to be interesting because it presented selective micromolar inhibition activity on SsCK1 (IC₅₀ 1.4 μM). Molecules 9h and 9i were also bioactive on SsCK1 and HsCDK5-p25. The current results are the starting point of a new larger program within our group to investigate intensively the biological properties of these new inhibitors with potential application in Alzheimer’s disease or in cancer.

Acknowledgments

Two of us (C.D.D. and C.N.A.) wish to thank the “Benian International Fondation”, “ElecBTP” and the “Ministère de l’Enseignement Supérieur et de la Recherche de la Côte d’Ivoire” for their respective grants. Financial support of this program carried out under the French National Cancer Institute “Cancéropôle Grand Ouest” by contract “Ion Channel-Network CGO 2012”, is gratefully acknowledged.

Author Contributions

J.-P.B. was the manager of the project. J.-P.B., Y.-A.B. and J.M. conceived and designed the experiments. C.D.D., C.N.A. and W.-K.C. performed the chemical experiments. A.D. and B.B. performed the kinase assays. S.B. and S.R. analyzed the data of kinase assays. M.R. and R.L.G. performed the
antiproliferative activity assays. R.L.G. and A.C. analyzed the data of antiproliferative assays and J.-P.B. wrote the paper.

Conflicts of Interest

The authors declare no conflict of interest.

References


Sample Availability: Samples of the compounds 9a–n are available from the authors.

© 2015 by the authors; licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution license (http://creativecommons.org/licenses/by/4.0/).