



(51) International Patent Classification:

C07D 413/14 (2006.01) C07D 487/04 (2006.01)
C07D 413/12 (2006.01) C07D 263/48 (2006.01)
C07D 417/12 (2006.01) A61P 35/00 (2006.01)
C07D 417/14 (2006.01) A61K 31/422 (2006.01)

(21) International Application Number:

PCT/GB2017/053822

(22) International Filing Date:

19 December 2017 (19.12.2017)

(25) Filing Language:

English

(26) Publication Language:

English

(30) Priority Data:

1622365.3 29 December 2016 (29.12.2016) GB

(71) Applicant: LIFEARC [GB/GB]; 7th Floor Lynton House,
7-12 Tavistock Square, London Greater London WC1H
9LT (GB).

(72) Inventors: OSBORNE, Simon; LifeArc, 7th Floor Lyn-
ton House, 7-12 Tavistock Square, London Greater London
WC1H 9LT (GB). LARGE, Jonathan; LifeArc, 7th Floor
Lynton House, 7-12 Tavistock Square, London Greater
London WC1H 9LT (GB).

(74) Agent: D Young / Co LLP; CLYDE-WATSON, Zoe, 120
Holborn, London Greater London EC1N 2DY (GB).

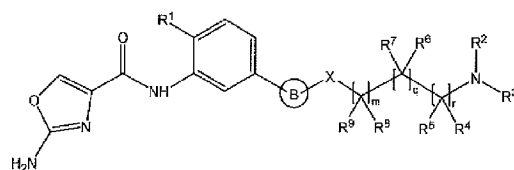
(81) Designated States (unless otherwise indicated, for every
kind of national protection available): AE, AG, AL, AM,
AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ,
CA, CH, CL, CN, CO, CR, CU, CZ, DE, DJ, DK, DM, DO,
DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN,
HR, HU, ID, IL, IN, IR, IS, JO, JP, KE, KG, KH, KN, KP,
KR, KW, KZ, LA, LC, LK, LR, LS, LU, LY, MA, MD, ME,
MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ,
OM, PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SA,
SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN,
TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) Designated States (unless otherwise indicated, for every
kind of regional protection available): ARIPO (BW, GH,
GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, ST, SZ, TZ,
UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU, TJ,
TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK,
EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV,
MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM,
TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW,
KM, ML, MR, NE, SN, TD, TG).

Published:

— with international search report (Art. 21(3))

(54) Title: OXAZOLE DERIVATIVES FOR USE IN THE TREATMENT OF CANCER



(I)

(57) Abstract: A first aspect of the invention relates to a compound of formula (I), or a pharmaceutically acceptable salt or ester thereof, wherein: • B is an aryl or heteroaryl group optionally substituted by one or more R¹⁰ groups; and * X is selected from O, (CR¹¹ R¹²) p and (CR¹¹ R¹²) p CO. Said compounds are capable of inhibiting PAICS and are useful in the treatment of proliferative disorders. Further aspects relate to pharmaceutical compositions, therapeutic uses and process for preparing compounds of formula (I).



OXAZOLE DERIVATIVES FOR USE IN THE TREATMENT OF CANCER

The present invention relates to substituted oxazole derivatives that are capable of inhibiting PAICS. The compounds find applications in the treatment of a variety of disorders, including proliferative disorders such as cancer.

BACKGROUND TO THE INVENTION

PAICS (phosphoribosylaminoimidazole carboxylase, phosphoribosylaminoimidazole succinocarboxamide synthetase) is a bifunctional 46kD enzyme catalysing the 6th and 7th steps of the *de novo* purine pathway (see Figure 1).

PAICS converts 5-aminoimidazole ribonucleotide (AIR) to 4-carboxy-5-aminoimidazole ribonucleotide (CAIR) in an ATP dependent reaction, before finally generating 4-(N-succinylcarboxamide)-5-aminoimidazole ribonucleotide (SAICAR) in a carboxylation reaction (see Figure 2). This reaction series feeds into the overall generation of inositol monophosphate (IMP), a nucleotide that forms the substrate for AMP and GMP production, from phosphoribosyl pyrophosphate (PRPP). The inhibition of folic acid, pyrimidine and purine biosynthetic pathways has proved an attractive drug target for cancer chemotherapy as rapidly dividing cancer cells have a high biosynthetic requirement in comparison to non-transformed cells.

Recent literature has highlighted PAICS as an emerging novel target for cancer therapeutics. PAICS was identified as an anti-apoptotic oncogene, with PAICS shRNA protein knock-down reducing the proliferation of a melanoma cell line *in vitro*. Furthermore, subcutaneous injection of PAICS knock-down cells in a xenograft model significantly reduced the rate of tumour growth (Eißmann *et al.*, PLoS One, 2013 May 22;8(5):e64873).

PAICS expression is significantly upregulated in lung cancer, and moreover expression levels were related to the prognosis of the patient population; increased expression of PAICS was coupled with tumours of a more aggressive nature. Xenograft models performed using lung cancer PAICS knock-down cells led to a significant reduction in tumour volume and weight after several weeks (Goswami *et al.*, Oncotarget, 2015 Sep

15;6(27):23445-61). PAICS over-expression has been also associated with a wide range of other tumour types.

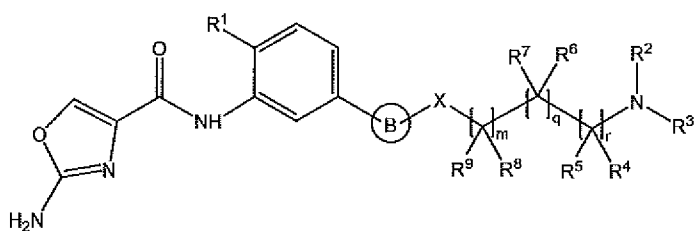
5 Further studies indicated that PAICS may be a useful biomarker too for poor prognosis prostate cancer, with heightened expression found in prostate cancer and the severe castration-resistant form, relative to benign prostate hyperplasia samples (Barfeld *et al.*, *Oncotarget*, 2015 May 20;6(14):12587-602).

10 During the recent emergence of PAICS as a potential target for cancer therapeutics, studies have demonstrated that the PAICS gene is overexpressed as part of a nine gene-expression signature that is strongly associated with poor-prognosis in triple negative breast cancer (TNBC) patients. Experimental knock-down of any one of these genes had a marked inhibitory effect on cancer cell growth and metastasis *in vitro* and *in vivo*. Specifically, shRNA inhibition of PAICS expression strongly impaired primary
15 tumour growth when breast cancer cells were injected orthotopically in the mammary fat pad of mice. Down regulation of PAICS expression in highly metastatic human breast cancer cells abolished the ability of these cells to form metastases to the lungs when injected intravenously into immunocompromised mice. Of note, this highly predictive gene-expression signature has a similar prognostic power in breast cancer patients as
20 the gene-expression signatures, MammaPrint® or OncotypeDX®, currently used in the clinic.

The present invention seeks to provide small molecule inhibitors of PAICS. In a preferred aspect, the invention seeks to provide small molecule inhibitors of PAICS that
25 target the SAICAR synthetase domain. Such small molecule inhibitors have potential therapeutic applications in the treatment of proliferative disorders such as cancer.

STATEMENT OF INVENTION

30 A first aspect of the invention relates to a compound of formula (I), or a pharmaceutically acceptable salt or ester thereof,



(I)

wherein:

- B is an aryl or heteroaryl group, each of which is optionally substituted by one or more
 5 R¹⁰ groups;
 X is selected from O, (CR¹¹R¹²)_p and (CR¹¹R¹²)_pCO;
 each R¹ is independently selected from Cl and SR¹³;
 R² is selected from H, alkyl, cycloalkyl, heterocycloalkyl and COR³³, wherein said alkyl,
 cycloalkyl or heterocycloalkyl group is optionally substituted by one or more R¹⁴
 10 substituents;
 R³ is selected from alkyl, cycloalkyl and heterocycloalkyl, each of which is optionally
 substituted with one or more R¹⁵ substituents; or
 R² and R³ are linked together with the nitrogen to which they are attached to form a
 saturated heterocyclic group optionally containing one or more additional heteroatoms
 15 selected from O, N and S and optionally further substituted by one or more R¹⁶ groups;
 each R⁴ and R⁵ is independently selected from H, alkyl, (CH₂)_tOR²⁴ and (CH₂)_uNR²⁵R²⁶;
 or
 one of R⁴ and R⁵ is H or alkyl and the other is linked to R³ to form a saturated
 heterocyclic group;
 20 each R⁶ and R⁷ is independently selected from H, alkyl, (CH₂)_vOR²⁷ and (CH₂)_wNR²⁸R²⁹;
 or
 one of R⁶ and R⁷ is H or alkyl and the other is linked to R³ to form a saturated
 heterocyclic group;
 each R⁸ and R⁹ is independently selected from H, alkyl, (CH₂)_xOR³⁰ and (CH₂)_yNR³¹R³²;
 25 or
 one of R⁸ and R⁹ is H or alkyl and the other is linked to R³ to form a saturated
 heterocyclic group; or one of R⁸ or R⁹ is linked to one of R⁴ or R⁵ to form a cyclic group;
 R¹⁰ is selected from alkyl, OH, halogen, alkoxy, CO₂-alkyl, COOH, CO-alkyl, NO₂ and
 CN;
 30 each R¹⁴, R¹⁵, R¹⁶ and R³³ is independently selected from (CH₂)_s-R¹⁷,

R¹⁷ is selected from alkyl, NR¹⁸R¹⁹, OR²⁰, SR²¹, COR²² and CO₂R²³;

R¹¹, R¹² and R¹⁸-R³² are each independently selected from H and alkyl;

R¹³ is alkyl;

m, q and r are each independently 0, 1 or 2;

5 p is 0 or 1;

such that the sum of m, p, q and r is 0, 1, 2, 3, 4 or 5, preferably 0, 1, 2, 3 or 4; and each of s, t, u, v, w, x, y is independently 0, 1, 2, 3 or 4.

10 A second aspect of the invention relates to a pharmaceutical composition comprising at least one compound as described above and a pharmaceutically acceptable carrier, diluent or excipient.

A third aspect of the invention relates to a compound as described above for use in medicine.

15

A fourth aspect of the invention relates to a compound as described above for use in treating a proliferative disorder.

20 A fifth aspect of the invention relates to the use of a compound as described above in the preparation of a medicament for treating or preventing a proliferative disorder.

A sixth aspect of the invention relates to a method of treating a proliferative disorder in a subject in need thereof, said method comprising administering to the subject a therapeutically effective amount of a compound as described above.

25

A seventh aspect of the invention relates to a method of treating a subject having a disease state alleviated by inhibition of PAICS, wherein the method comprises administering to the subject a therapeutically effective amount of a compound as described above.

30

An eighth aspect of the invention relates to the use of a compound as described above in an assay for identifying further candidate compounds capable of inhibiting PAICS.

A ninth aspect of the invention relates to a combination comprising a compound as described above and a second therapeutic agent.

5 A tenth aspect of the invention relates to a process for preparing compounds as described herein.

DETAILED DESCRIPTION

The present invention relates to substituted oxazole derivatives that are capable of inhibiting PAICS.

10

"Alkyl" is defined herein as a straight-chain or branched alkyl radical, for example, methyl, ethyl, propyl, isopropyl, butyl, isobutyl, tert-butyl, pentyl, hexyl. Preferably, the alkyl group is a C₁₋₁₂-alkyl group, more preferably, a C₁₋₆-alkyl group, even more preferably a C₁₋₄-alkyl group.

15

"Cycloalkyl" is defined herein as a monocyclic alkyl ring, such as, cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl or cycloheptyl, or a fused bicyclic ring system such as norbornane. Preferably, the cycloalkyl group is a C₃₋₈-cycloalkyl group, more preferably a C₃₋₆-cycloalkyl group.

20

"Halogen" is defined herein as chloro, fluoro, bromo or iodo.

As used herein, the term "saturated heterocyclic group" is defined herein as a saturated monocyclic or bicyclic group comprising one or more heteroatoms (that may be the same or different), such as oxygen, nitrogen or sulfur, which is optionally interrupted by one or more -(CO)- groups in the ring. Preferably, the saturated heterocyclic group is a C₃-C₇-heterocycloalkyl group, more preferably a C₃-C₆-heterocycloalkyl group. Alternatively, the heterocycloalkyl group is a C₄-C₇-heterocycloalkyl, more preferably a C₄-C₆-heterocycloalkyl. Preferred saturated heterocyclic groups include, but are not limited to, piperazinyl, piperidinyl, morpholinyl, thiomorpholinyl and pyrrolidinyl.

30

As used herein, the term "aryl" refers to an aromatic group. Preferably, the aryl group is phenyl.

"Heteroaryl" is defined herein as a monocyclic or bicyclic C₂₋₁₂ aromatic ring comprising one or more heteroatoms (that may be the same or different), such as oxygen, nitrogen or sulfur. Examples of suitable heteroaryl groups include thienyl, furanyl, pyrrolyl, pyridinyl, oxazolyl, thiazolyl, imidazolyl, pyrazolyl, isoxazolyl, isothiazolyl, oxadiazolyl, triazolyl, thiadiazolyl etc. and benzo derivatives thereof, such as benzofuranyl, benzothienyl, benzimidazolyl, indolyl, isoindolyl, indazolyl etc.; or pyridyl, pyrazinyl, pyrimidinyl, pyridazinyl, triazinyl etc. and benzo derivatives thereof, such as quinolinyl, isoquinolinyl, cinnolinyl, phthalazinyl, quinazolinyl, quinoxalinyl, naphthyridinyl etc.

10 More preferably, the heteroaryl group is a monocyclic 5- or 6-membered aromatic ring comprising one or more heteroatoms (that may be the same or different), such as oxygen, nitrogen or sulfur. Non-limiting examples of suitable heteroaryl groups include thienyl, furanyl, pyrrolyl, pyridinyl, oxazolyl, pyrazinyl, thiazolyl, imidazolyl, pyrazolyl, isoxazolyl, isothiazolyl, oxadiazolyl, triazolyl, thiadiazolyl, pyrimidinyl, pyridazinyl, triazinyl and the like.

In one preferred embodiment, B is a 5- or 6-membered monocyclic aryl or heteroaryl group, each of which is optionally substituted by one or more R¹⁰ groups.

20 In one preferred embodiment:
each R⁴ and R⁵ is independently selected from H and alkyl; or
one of R⁴ and R⁵ is H or alkyl and the other is linked to R³ to form a saturated heterocyclic group.

25 In one preferred embodiment:
each R⁶ and R⁷ is independently selected from H and alkyl; or
one of R⁶ and R⁷ is H or alkyl and the other is linked to R³ to form a saturated heterocyclic group.

30 In one preferred embodiment:
each R⁸ and R⁹ is independently selected from H and alkyl; or
one of R⁸ and R⁹ is H or alkyl and the other is linked to R³ to form a saturated heterocyclic group.

In one preferred embodiment:

each R⁴ and R⁵ is independently selected from H and alkyl; or
one of R⁴ and R⁵ is H or alkyl and the other is linked to R³ to form a saturated heterocyclic group;

5 each R⁶ and R⁷ is independently selected from H and alkyl; or
one of R⁶ and R⁷ is H or alkyl and the other is linked to R³ to form a saturated heterocyclic group;

each R⁸ and R⁹ is independently selected from H and alkyl; or
one of R⁸ and R⁹ is H or alkyl and the other is linked to R³ to form a saturated

10 heterocyclic group.

Preferably, where:

- one of R⁶ and R⁷ is H or alkyl (more preferably methyl) and the other is linked to R³ to form a saturated heterocyclic group, or
- 15 - one of R⁸ and R⁹ is H or alkyl (more preferably methyl) and the other is linked to R³ to form a saturated heterocyclic group, or
- one of R⁴ and R⁵ is H or alkyl (more preferably methyl) and the other is linked to R³ to form a saturated heterocyclic group;

the saturated heterocyclic group is a 4-, 5- or 6- membered heterocyclic group, more
20 preferably a 5- or 6- membered heterocyclic group, even more preferably, a pyrrolidinyl or piperidinyl group.

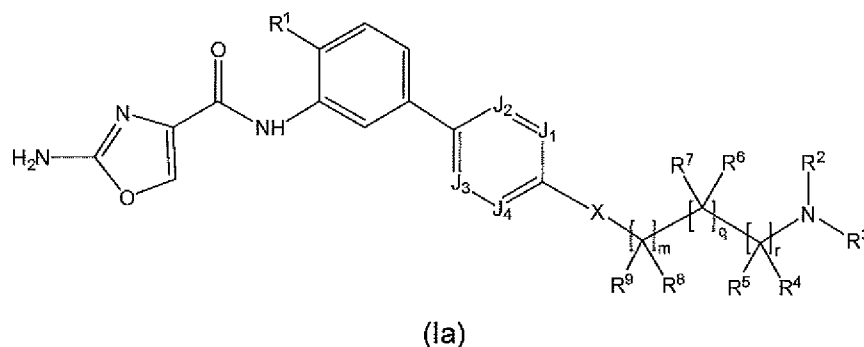
In one preferred embodiment, B is selected from phenyl, thienyl, furanyl, pyrrolyl, pyridinyl, oxazolyl, pyrazinyl, thiazolyl, imidazolyl, pyrazolyl, isoxazolyl, isothiazolyl,
25 oxadiazolyl, triazolyl, thiadiazolyl, pyrimidinyl, pyridazinyl and triazinyl.

In one preferred embodiment, B is selected from thienyl, furanyl, pyrrolyl, pyridinyl, oxazolyl, pyrazinyl, thiazolyl, imidazolyl, pyrazolyl, isoxazolyl, isothiazolyl, oxadiazolyl, triazolyl, thiadiazolyl, pyrimidinyl, pyridazinyl and triazinyl.

30

In a more preferred embodiment, B is selected from thiazolyl, pyridinyl, pyrazolyl, pyrimidinyl and phenyl.

In one preferred embodiment, the compound is of formula (Ia), or a pharmaceutically acceptable salt or ester thereof,



5

wherein J_1 , J_2 , J_3 and J_4 are each independently selected from =N, CH and CR^{10} and X, R^{1-10} , m, q and r are as defined above.

In one preferred embodiment, J_1 is CH, CR^{10} or =N, and J_2 , J_3 and J_4 are all CH or CR^{10} .

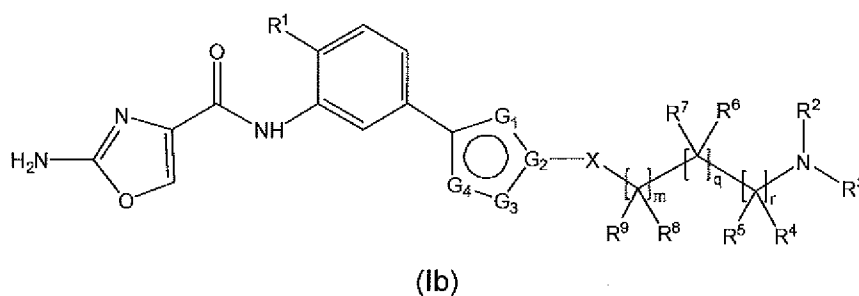
10

In one preferred embodiment, J_1 , J_2 , J_3 and J_4 are all CH.

In one preferred embodiment, J_1 is =N and J_2 , J_3 and J_4 are all CH.

15

In one preferred embodiment, the compound is of formula (Ib), or a pharmaceutically acceptable salt or ester thereof,



20

wherein:

- (i) G_1 is S, G_2 is =C, one of G_3 and G_4 is =N and the other is CH or CR^{10} ; or
 (ii) G_3 is S, G_1 is =N, G_2 is =C, and G_4 is CH or CR^{10} ; or
 (iii) G_1 and G_4 are each selected from CH and CR^{10} , G_2 is N and G_3 is =N; or

25

(iv) G_3 and G_4 are each selected from CH and CR^{10} , G_2 is N and G_1 is =N;
and X, R^{1-10} , m, q and r are as defined above.

5 In one preferred embodiment, G_1 is S, G_2 is =C, one of G_3 and G_4 is =N and the other is
CH or CR^{10} .

In one preferred embodiment, G_3 is S, G_1 is =N, G_2 is =C, and G_4 is CH or CR^{10} .

10 In one preferred embodiment, G_1 and G_4 are each selected from CH and CR^{10} , G_2 is N
and G_3 is =N.

In one preferred embodiment, G_3 and G_4 are each selected from CH and CR^{10} , G_2 is N
and G_1 is =N.

15 In one preferred embodiment, R^1 is selected from Cl and SMe.

In one particularly preferred embodiment, R^1 is Cl.

20 In another particularly preferred embodiment, R^1 is SMe.

In one preferred embodiment, X is O.

In another preferred embodiment, X is $(CR^{11}R^{12})_p$, where p is 0 or 1.

25 In one preferred embodiment, X is CH_2 .

In one preferred embodiment:

R^2 is selected from H, methyl, ethyl and isopropyl; and

30 R^3 is selected from methyl, ethyl, isopropyl and piperidinyl, wherein the piperidinyl group
is optionally substituted by one or more R^{15} substituents.

In one preferred embodiment, R^2 and R^3 are linked together with the nitrogen to which
they are attached to form a 5- or 6-membered saturated heterocyclic group optionally
substituted by one or more R^{16} groups.

In a more preferred embodiment, R² and R³ are linked together with the nitrogen to which they are attached to form a pyrrolidinyl, piperidinyl or piperazinyl group, each of which is optionally substituted by one or more substituents selected from alkyl and (CH₂)_sNR¹⁸R¹⁹.

5

In one preferred embodiment, R⁸ and R⁹ are both H.

In another preferred embodiment, R⁶ and R⁷ are both H.

10

In one preferred embodiment, R⁴ and R⁵ are both H.

In one preferred embodiment, m, q and r are each independently 0 or 1.

In one preferred embodiment:

15

m is 1;

q is 1;

r is 1;

one of R⁸ and R⁹ is H or alkyl and the other is linked to R³ to form a saturated heterocyclic group; and

20

R⁴, R⁵, R⁶ and R⁷ are each independently H or alkyl, more preferably H.

In one preferred embodiment, R⁸ and R⁹ is H or alkyl and the other is linked to R³ to form a piperidinyl group.

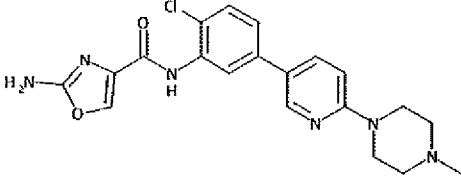
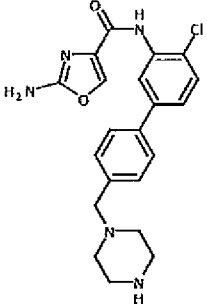
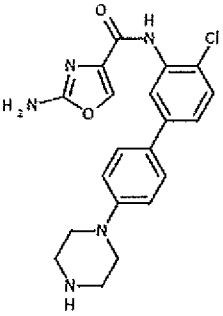
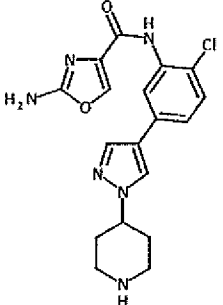
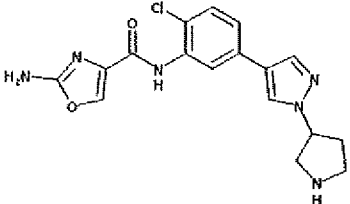
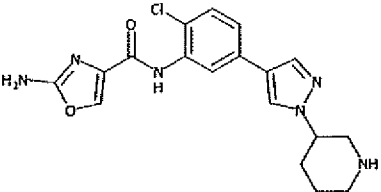
25

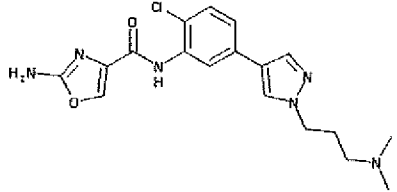
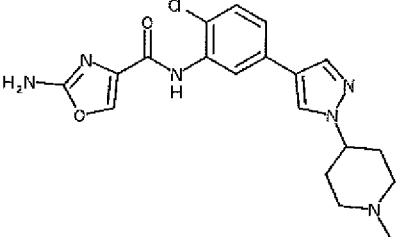
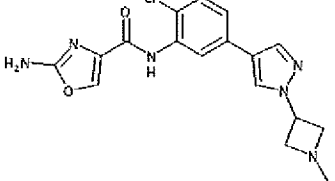
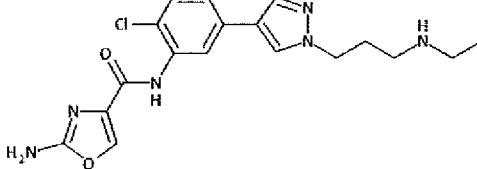
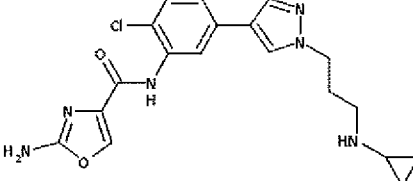
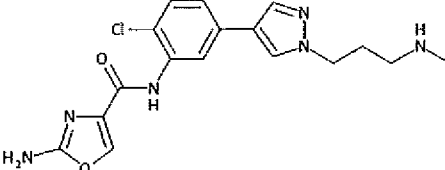
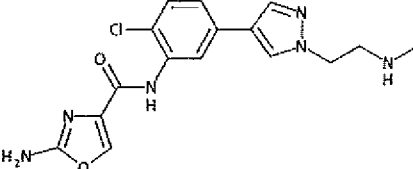
In one preferred embodiment, one of R⁸ or R⁹ is linked to one of R⁴ or R⁵ to form a cyclic group, preferably a 5- or 6-membered cyclic group, preferably, a cyclohexyl group.

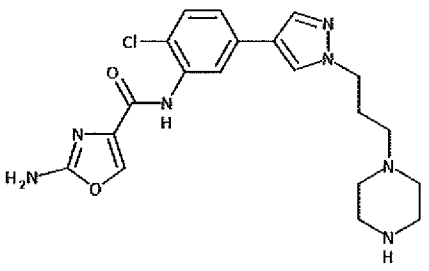
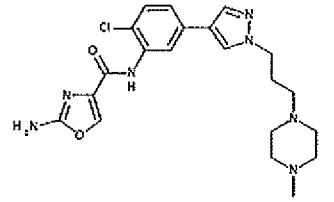
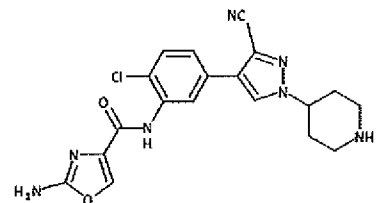
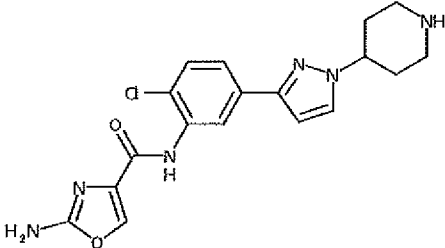
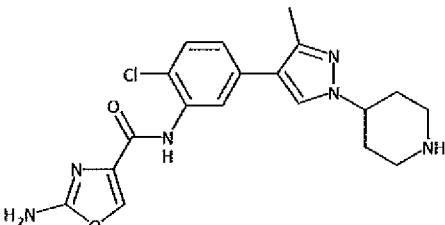
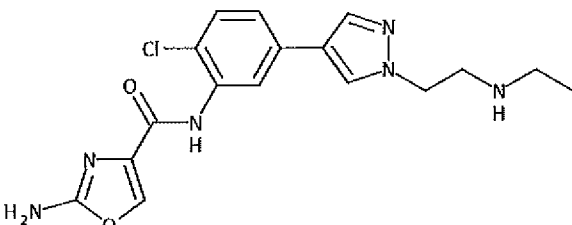
In one preferred embodiment, X is (CR¹¹R¹²)_p and p is 0.

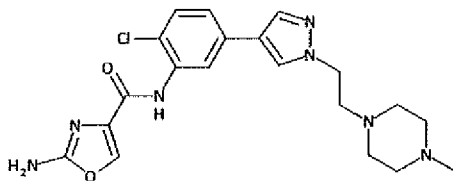
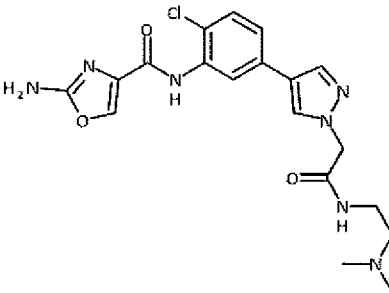
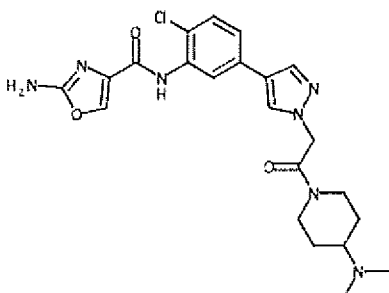
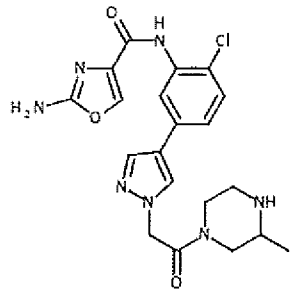
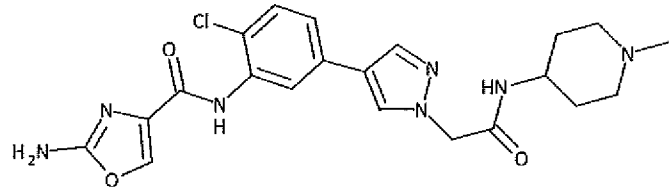
30

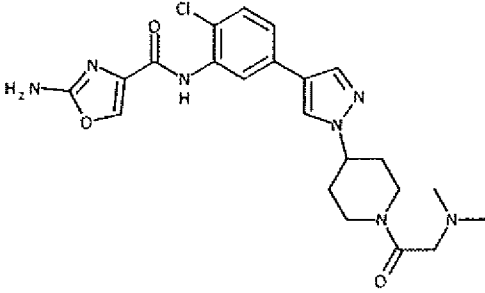
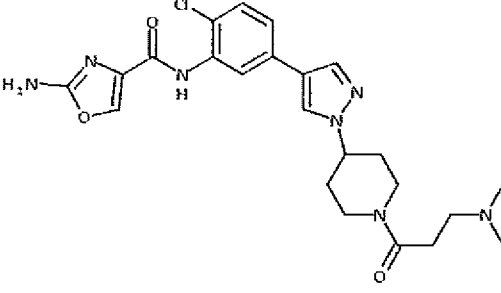
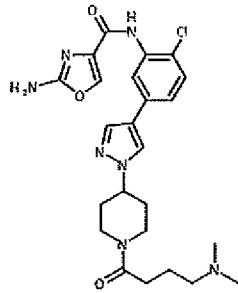
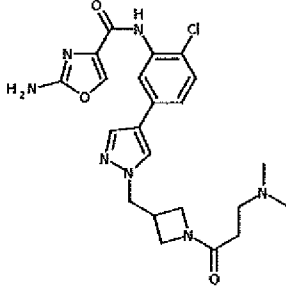
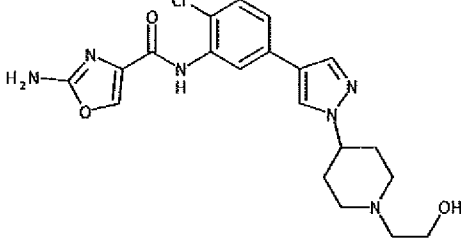
In one highly preferred embodiment, the compound is selected from the following:

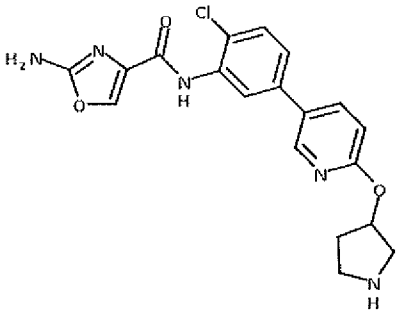
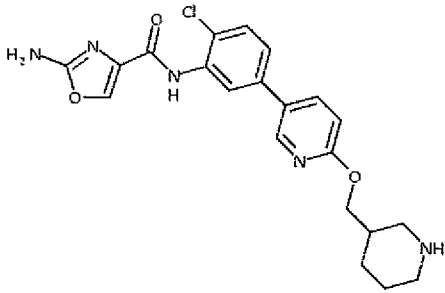
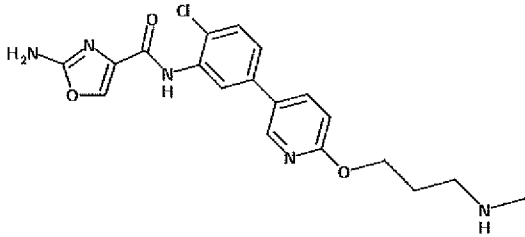
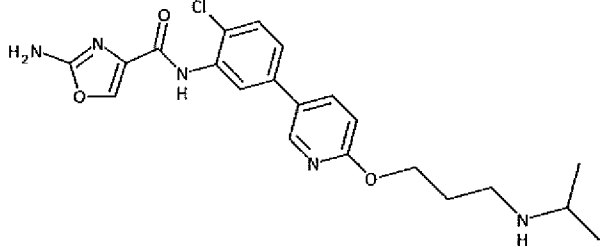
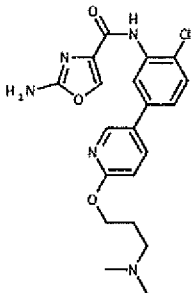
	37
	38
	39
	40
	41
	42

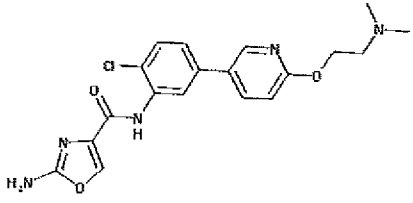
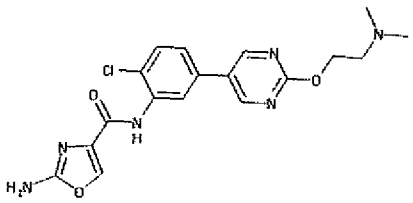
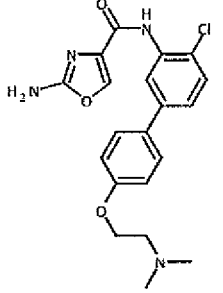
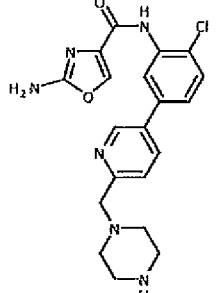
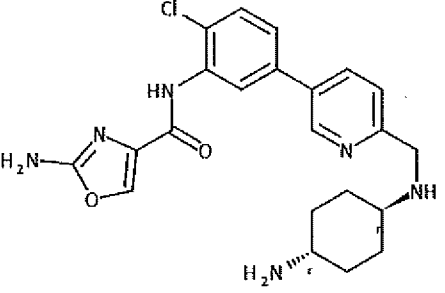
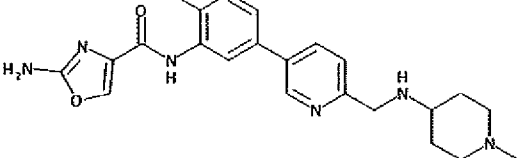
	<p>43</p>
	<p>44</p>
	<p>45</p>
	<p>47</p>
	<p>48</p>
	<p>49</p>
	<p>50</p>

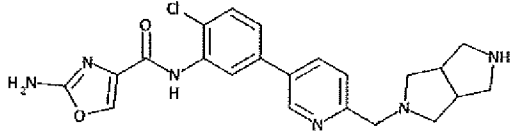
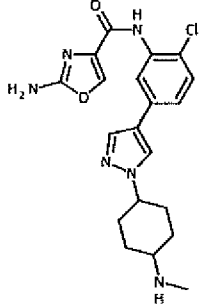
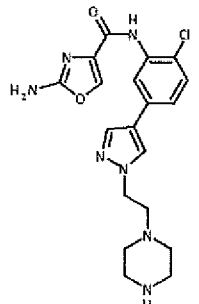
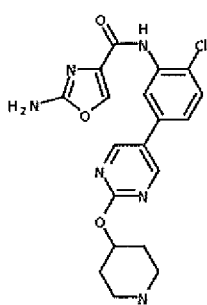
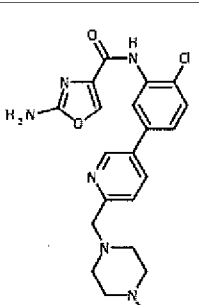
	<p>51</p>
	<p>52</p>
	<p>53</p>
	<p>54</p>
	<p>55</p>
	<p>56</p>

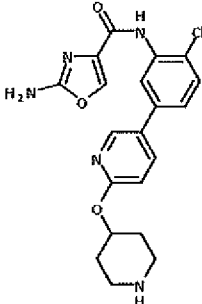
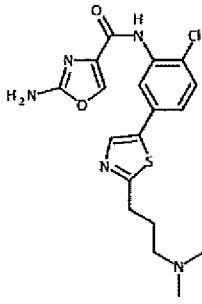
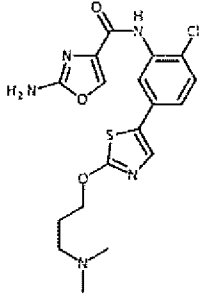
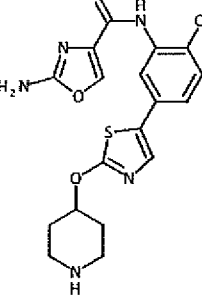
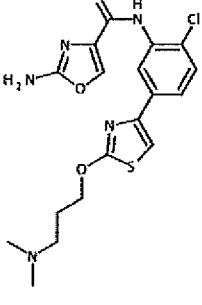
	<p>57</p>
	<p>58</p>
	<p>59</p>
	<p>60</p>
	<p>61</p>

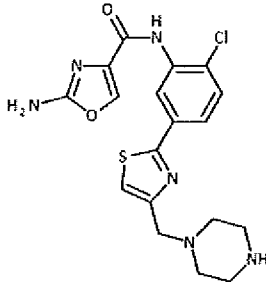
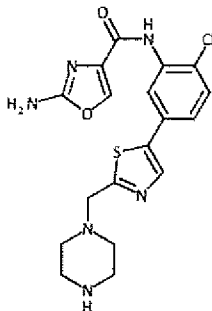
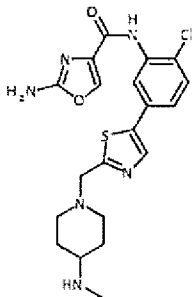
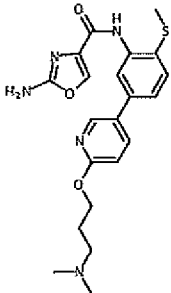
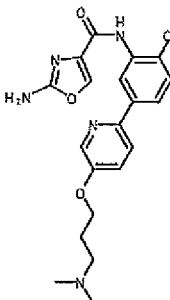
	<p>62</p>
	<p>63</p>
	<p>64</p>
	<p>65</p>
	<p>66</p>

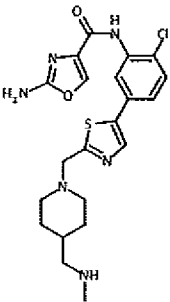
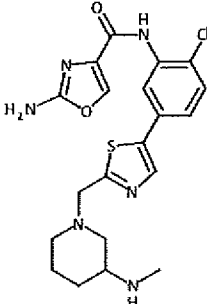
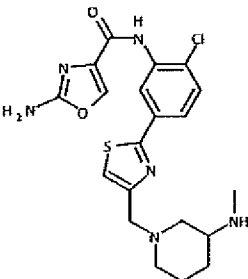
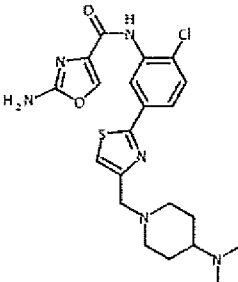
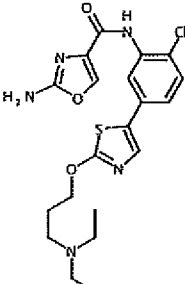
	<p>68</p>
	<p>69</p>
	<p>70</p>
	<p>71</p>
	<p>72</p>

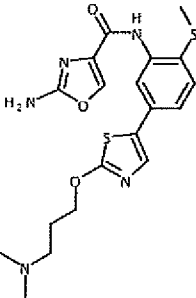
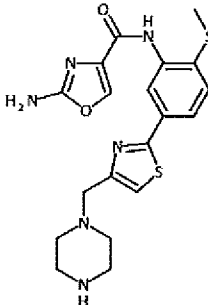
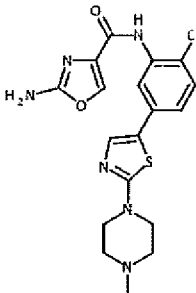
	<p>73</p>
	<p>74</p>
	<p>75</p>
	<p>76</p>
	<p>77</p>
	<p>78</p>

	79
	80
	81
	82
	83

	<p>84</p>
	<p>85</p>
	<p>86</p>
	<p>87</p>
	<p>88</p>

	<p>89</p>
	<p>90</p>
	<p>91</p>
	<p>92</p>
	<p>93</p>

	<p>94</p>
	<p>95</p>
	<p>96</p>
	<p>97</p>
	<p>98</p>

	99
	100
	101

and pharmaceutically acceptable salts and esters thereof.

In one even more highly preferred embodiment, the compound is selected from 47, 49,
5 80, 85, 86, 87, 89, 90, 91 and 94.

THERAPEUTIC APPLICATIONS

A further aspect of the invention relates to a compound as described above for use in
10 medicine.

Another aspect of the invention relates to a compound as described above for use in
treating a proliferative disorder.

The term "proliferative disorder" is used herein in a broad sense to include any disorder that requires control of the cell cycle, for example cardiovascular disorders such as restenosis and cardiomyopathy, auto-immune disorders such as glomerulonephritis and rheumatoid arthritis, dermatological disorders such as psoriasis, anti-inflammatory, anti-fungal, antiparasitic disorders such as malaria, emphysema and alopecia. In these disorders, the compounds of the present invention may induce apoptosis or maintain stasis within the desired cells as required.

In one preferred embodiment, the invention relates to a compound as described above for use in preventing or reducing metastasis. Thus, in one preferred embodiment, the compound is for use in preventing or alleviating or treating metastatic cancer, for example, secondary malignant growths at a distance from a primary site of cancer.

In another preferred embodiment, the invention relates to a compound as described above for use in blocking cell growth.

In one preferred embodiment, the proliferative disorder is cancer or leukemia. Preferably, the cancer is selected from solid cancers at any stage. In another preferred embodiment, the cancer is in a late-stage, with metastatic lesions.

Preferably, the cancer is selected from breast cancer, colon cancer, prostate melanoma, bladder, pancreatic, head and neck and ovarian cancer, with or without metastasis, and haematological cancers such as acute myeloid leukemia (AML), chronic lymphocytic leukemia (CLL), acute lymphocytic leukemia (ALL), multiple myeloma (MM) and non-Hodgkins lymphoma.

In one preferred embodiment, the proliferative disorder is selected from breast cancer, colon cancer, lung cancer, melanoma and prostate cancer. Studies by the applicant have demonstrated that PAICS mRNA is upregulated in these tumour types.

In one particularly preferred embodiment, the proliferative disorder is breast cancer. More preferably, the proliferative disorder is metastatic breast cancer or triple negative breast cancer (TNBC). Triple-negative breast cancer refers to any breast cancer that does not express the genes for estrogen receptor (ER), progesterone receptor (PR) or

Her2/neu. This makes it more difficult to treat since most chemotherapies target one of the three receptors, so triple-negative cancers often require combination therapies.

5 Another aspect relates to the use of a compound as described above in the preparation of a medicament for treating or preventing a proliferative disorder, for example, cancer or leukemia.

10 Another aspect relates to method of treating a proliferative disorder in a subject in need thereof, said method comprising administering to the subject a therapeutically effective amount of a compound as described above.

Preferably, the compound is administered in an amount sufficient to inhibit PAICS.

15 Another aspect relates to a compound of the invention for use in the prevention or treatment of a disorder caused by, associated with or accompanied by any abnormal activity against a biological target, wherein the target is PAICS.

20 Yet another aspect relates to the use of a compound of the invention in the preparation of a medicament for the prevention or treatment of a disorder caused by, associated with or accompanied by any abnormal activity against a biological target, wherein the target is PAICS.

25 Another aspect of the invention relates to a method of treating a PAICS related disease or disorder. The method according to this aspect of the present invention is effected by administering to a subject in need thereof a therapeutically effective amount of a compound of the present invention, as described hereinabove, either *per se*, or, more preferably, as a part of a pharmaceutical composition, mixed with, for example, a pharmaceutically acceptable carrier, as is detailed hereinafter.

30 Yet another aspect of the invention relates to a method of treating a mammal having a disease state alleviated by inhibition of PAICS, wherein the method comprises administering to a mammal a therapeutically effective amount of a compound according to the invention.

Preferably, the subject is a mammal, more preferably a human.

The term "method" refers to manners, means, techniques and procedures for accomplishing a given task including, but not limited to, those manners, means, 5 techniques and procedures either known to, or readily developed from known manners, means, techniques and procedures by practitioners of the chemical, pharmacological, biological, biochemical and medical arts.

The term "administering" as used herein refers to a method for bringing a compound of 10 the present invention and PAICS together in such a manner that the compound can affect the enzyme activity of the PAICS either directly; i.e., by interacting with the PAICS itself or indirectly; i.e., by interacting with another molecule on which the catalytic activity of the PAICS is dependent. As used herein, administration can be accomplished either *in vitro*, i.e. in a test tube, or *in vivo*, i.e., in cells or tissues of a 15 living organism.

Herein, the term "treating" includes abrogating, substantially inhibiting, slowing or reversing the progression of a disease or disorder, substantially ameliorating clinical 20 symptoms of a disease or disorder or substantially preventing the appearance of clinical symptoms of a disease or disorder.

Herein, the term "preventing" refers to a method for barring an organism from acquiring a disorder or disease in the first place.

25 The term "therapeutically effective amount" refers to that amount of the compound being administered which will relieve to some extent one or more of the symptoms of the disease or disorder being treated.

For any compound used in this invention, a therapeutically effective amount, also 30 referred to herein as a therapeutically effective dose, can be estimated initially from cell culture assays. For example, a dose can be formulated in animal models to achieve a circulating concentration range that includes the IC₅₀ or the IC₁₀₀ as determined in cell culture. Such information can be used to more accurately determine useful doses in humans. Initial dosages can also be estimated from *in vivo* data. Using these initial

guidelines one of ordinary skill in the art could determine an effective dosage in humans.

Moreover, toxicity and therapeutic efficacy of the compounds described herein can be
5 determined by standard pharmaceutical procedures in cell cultures or experimental
animals, e.g., by determining the LD₅₀ and the ED₅₀. The dose ratio between toxic and
therapeutic effect is the therapeutic index and can be expressed as the ratio between
LD₅₀ and ED₅₀. Compounds which exhibit high therapeutic indices are preferred. The
data obtained from these cell cultures assays and animal studies can be used in
10 formulating a dosage range that is not toxic for use in human. The dosage of such
compounds lies preferably within a range of circulating concentrations that include the
ED₅₀ with little or no toxicity. The dosage may vary within this range depending upon
the dosage form employed and the route of administration utilized. The exact
formulation, route of administration and dosage can be chosen by the individual
15 physician in view of the patient's condition. (see, e.g., Fingl *et al*, 1975, In: The
Pharmacological Basis of Therapeutics, chapter 1, page 1).

Dosage amount and interval may be adjusted individually to provide plasma levels of
the active compound which are sufficient to maintain therapeutic effect. Usual patient
20 dosages for oral administration range from about 50-2000 mg/kg/day, commonly from
about 100-1000 mg/kg/day, preferably from about 150-700 mg/kg/day and most
preferably from about 250-500 mg/kg/day. Preferably, therapeutically effective serum
levels will be achieved by administering multiple doses each day. In cases of local
administration or selective uptake, the effective local concentration of the drug may not
25 be related to plasma concentration. One skilled in the art will be able to optimize
therapeutically effective local dosages without undue experimentation.

As used herein, "PAICS related disease or disorder" refers to a disease or disorder
characterized by inappropriate or abnormal PAICS activity or over-activity.
30 Inappropriate or abnormal activity refers to either; (i) expression in cells which normally
do not express the protein; (ii) increased expression leading to unwanted cell
proliferation, differentiation and/or growth; or, (iii) decreased expression leading to
unwanted reductions in cell proliferation, differentiation and/or growth. Over-activity of
PAICS refers to either amplification of the gene encoding PAICS or production of a level

of PAICS activity, which can correlate with a cell proliferation, differentiation and/or growth disorder (that is, as the level of the PAICS increases, the severity of one or more of the symptoms of the cellular disorder increases). Over-activity can also be the result of ligand-independent or constitutive activation as a result of mutations such as
5 deletions of a fragment of the protein responsible for ligand binding.

Thus, the present invention further provides use of compounds as defined herein for the manufacture of medicaments for the treatment of diseases where it is desirable to inhibit PAICS. Such diseases include proliferative disorders such as cancer or
10 leukemia.

PHARMACEUTICAL COMPOSITIONS

For use according to the present invention, the compounds or physiologically acceptable salt, ester or other physiologically functional derivative thereof, described
15 herein, may be presented as a pharmaceutical formulation, comprising the compounds or physiologically acceptable salt, ester or other physiologically functional derivative thereof, together with one or more pharmaceutically acceptable carriers and optionally other therapeutic and/or prophylactic ingredients. The carrier(s) must be acceptable in the sense of being compatible with the other ingredients of the formulation and not
20 deleterious to the recipient thereof. The pharmaceutical compositions may be for human or animal usage in human and veterinary medicine.

Examples of such suitable excipients for the various different forms of pharmaceutical compositions described herein may be found in the Handbook of Pharmaceutical
25 Excipients, 2nd Edition, (1994), Edited by A Wade and PJ Weller.

Acceptable carriers or diluents for therapeutic use are well known in the pharmaceutical art, and are described, for example, in Remington's Pharmaceutical Sciences, Mack
Publishing Co. (A. R. Gennaro edit. 1985).

30

Examples of suitable carriers include lactose, starch, glucose, methyl cellulose, magnesium stearate, mannitol, sorbitol and the like. Examples of suitable diluents include ethanol, glycerol and water.

The choice of pharmaceutical carrier, excipient or diluent can be selected with regard to the intended route of administration and standard pharmaceutical practice. The pharmaceutical compositions may comprise as, or in addition to, the carrier, excipient or diluent any suitable binder(s), lubricant(s), suspending agent(s), coating agent(s),
5 solubilising agent(s), buffer(s), flavouring agent(s), surface active agent(s), thickener(s), preservative(s) (including anti-oxidants) and the like, and substances included for the purpose of rendering the formulation isotonic with the blood of the intended recipient.

Examples of suitable binders include starch, gelatin, natural sugars such as glucose,
10 anhydrous lactose, free-flow lactose, beta-lactose, corn sweeteners, natural and synthetic gums, such as acacia, tragacanth or sodium alginate, carboxymethyl cellulose and polyethylene glycol.

Examples of suitable lubricants include sodium oleate, sodium stearate, magnesium
15 stearate, sodium benzoate, sodium acetate, sodium chloride and the like.

Preservatives, stabilizers, dyes and even flavoring agents may be provided in the pharmaceutical composition. Examples of preservatives include sodium benzoate, sorbic acid and esters of p-hydroxybenzoic acid. Antioxidants and suspending agents may be also used.

20 Pharmaceutical formulations include those suitable for oral, topical (including dermal, buccal and sublingual), rectal or parenteral (including subcutaneous, intradermal, intramuscular and intravenous), nasal and pulmonary administration e.g., by inhalation. The formulation may, where appropriate, be conveniently presented in discrete dosage
25 units and may be prepared by any of the methods well known in the art of pharmacy. All methods include the step of bringing into association an active compound with liquid carriers or finely divided solid carriers or both and then, if necessary, shaping the product into the desired formulation.

30 Pharmaceutical formulations suitable for oral administration wherein the carrier is a solid are most preferably presented as unit dose formulations such as boluses, capsules or tablets each containing a predetermined amount of active compound. A tablet may be made by compression or moulding, optionally with one or more accessory ingredients. Compressed tablets may be prepared by compressing in a suitable

machine an active compound in a free-flowing form such as a powder or granules optionally mixed with a binder, lubricant, inert diluent, lubricating agent, surface-active agent or dispersing agent. Moulded tablets may be made by moulding an active compound with an inert liquid diluent. Tablets may be optionally coated and, if
5 uncoated, may optionally be scored. Capsules may be prepared by filling an active compound, either alone or in admixture with one or more accessory ingredients, into the capsule shells and then sealing them in the usual manner. Cachets are analogous to capsules wherein an active compound together with any accessory ingredient(s) is sealed in a rice paper envelope. An active compound may also be formulated as
10 dispersible granules, which may for example be suspended in water before administration, or sprinkled on food. The granules may be packaged, e.g., in a sachet. Formulations suitable for oral administration wherein the carrier is a liquid may be presented as a solution or a suspension in an aqueous or non-aqueous liquid, or as an oil-in-water liquid emulsion.

15 Formulations for oral administration include controlled release dosage forms, e.g., tablets wherein an active compound is formulated in an appropriate release-controlling matrix, or is coated with a suitable release-controlling film. Such formulations may be particularly convenient for prophylactic use.

20 Pharmaceutical formulations suitable for rectal administration wherein the carrier is a solid are most preferably presented as unit dose suppositories. Suitable carriers include cocoa butter and other materials commonly used in the art. The suppositories may be conveniently formed by admixture of an active compound with the softened or
25 melted carrier(s) followed by chilling and shaping in moulds. Pharmaceutical formulations suitable for parenteral administration include sterile solutions or suspensions of an active compound in aqueous or oleaginous vehicles.

Injectable preparations may be adapted for bolus injection or continuous infusion. Such
30 preparations are conveniently presented in unit dose or multi-dose containers which are sealed after introduction of the formulation until required for use. Alternatively, an active compound may be in powder form which is constituted with a suitable vehicle, such as sterile, pyrogen-free water, before use.

An active compound may also be formulated as long-acting depot preparations, which may be administered by intramuscular injection or by implantation, e.g., subcutaneously or intramuscularly. Depot preparations may include, for example, suitable polymeric or hydrophobic materials, or ion-exchange resins. Such long-acting formulations are particularly convenient for prophylactic use.

Formulations suitable for pulmonary administration via the buccal cavity are presented such that particles containing an active compound and desirably having a diameter in the range of 0.5 to 7 microns are delivered in the bronchial tree of the recipient.

As one possibility such formulations are in the form of finely comminuted powders which may conveniently be presented either in a pierceable capsule, suitably of, for example, gelatin, for use in an inhalation device, or alternatively as a self-propelling formulation comprising an active compound, a suitable liquid or gaseous propellant and optionally other ingredients such as a surfactant and/or a solid diluent. Suitable liquid propellants include propane and the chlorofluorocarbons, and suitable gaseous propellants include carbon dioxide. Self-propelling formulations may also be employed wherein an active compound is dispensed in the form of droplets of solution or suspension.

Such self-propelling formulations are analogous to those known in the art and may be prepared by established procedures. Suitably they are presented in a container provided with either a manually-operable or automatically functioning valve having the desired spray characteristics; advantageously the valve is of a metered type delivering a fixed volume, for example, 25 to 100 microlitres, upon each operation thereof.

As a further possibility an active compound may be in the form of a solution or suspension for use in an atomizer or nebuliser whereby an accelerated airstream or ultrasonic agitation is employed to produce a fine droplet mist for inhalation.

Formulations suitable for nasal administration include preparations generally similar to those described above for pulmonary administration. When dispensed such formulations should desirably have a particle diameter in the range 10 to 200 microns to enable retention in the nasal cavity; this may be achieved by, as appropriate, use of a powder of a suitable particle size or choice of an appropriate valve. Other suitable

formulations include coarse powders having a particle diameter in the range 20 to 500 microns, for administration by rapid inhalation through the nasal passage from a container held close up to the nose, and nasal drops comprising 0.2 to 5% w/v of an active compound in aqueous or oily solution or suspension.

5

Pharmaceutically acceptable carriers are well known to those skilled in the art and include, but are not limited to, 0.1 M and preferably 0.05 M phosphate buffer or 0.8% saline. Additionally, such pharmaceutically acceptable carriers may be aqueous or non-aqueous solutions, suspensions, and emulsions. Examples of non-aqueous solvents
10 are propylene glycol, polyethylene glycol, vegetable oils such as olive oil, and injectable organic esters such as ethyl oleate. Aqueous carriers include water, alcoholic/aqueous solutions, emulsions or suspensions, including saline and buffered media. Parenteral vehicles include sodium chloride solution, Ringer's dextrose, dextrose and sodium chloride, lactated Ringer's or fixed oils. Preservatives and other additives may also be
15 present, such as, for example, antimicrobials, antioxidants, chelating agents, inert gases and the like.

Formulations suitable for topical formulation may be provided for example as gels, creams or ointments. Such preparations may be applied e.g. to a wound or ulcer either
20 directly spread upon the surface of the wound or ulcer or carried on a suitable support such as a bandage, gauze, mesh or the like which may be applied to and over the area to be treated.

Liquid or powder formulations may also be provided which can be sprayed or sprinkled
25 directly onto the site to be treated, e.g. a wound or ulcer. Alternatively, a carrier such as a bandage, gauze, mesh or the like can be sprayed or sprinkled with the formulation and then applied to the site to be treated.

According to a further aspect of the invention, there is provided a process for the
30 preparation of a pharmaceutical or veterinary composition as described above, the process comprising bringing the active compound(s) into association with the carrier, for example by admixture.

In general, the formulations are prepared by uniformly and intimately bringing into association the active agent with liquid carriers or finely divided solid carriers or both, and then if necessary shaping the product. The invention extends to methods for preparing a pharmaceutical composition comprising bringing a compound of general formula (I) in conjunction or association with a pharmaceutically or veterinarily acceptable carrier or vehicle.

SALTS/ESTERS

The compounds of the invention can be present as salts or esters, in particular pharmaceutically and veterinarily acceptable salts or esters.

Pharmaceutically acceptable salts of the compounds of the invention include suitable acid addition or base salts thereof. A review of suitable pharmaceutical salts may be found in Berge *et al*, J Pharm Sci, 66, 1-19 (1977). Salts are formed, for example with strong inorganic acids such as mineral acids, e.g. hydrohalic acids such as hydrochloride, hydrobromide and hydroiodide, sulfuric acid, phosphoric acid sulfate, bisulfate, hemisulfate, thiocyanate, persulfate and sulfonic acids; with strong organic carboxylic acids, such as alkanecarboxylic acids of 1 to 4 carbon atoms which are unsubstituted or substituted (e.g., by halogen), such as acetic acid; with saturated or unsaturated dicarboxylic acids, for example oxalic, malonic, succinic, maleic, fumaric, phthalic or tetraphthalic; with hydroxycarboxylic acids, for example ascorbic, glycolic, lactic, malic, tartaric or citric acid; with amino acids, for example aspartic or glutamic acid; with benzoic acid; or with organic sulfonic acids, such as (C₁-C₄)-alkyl- or aryl-sulfonic acids which are unsubstituted or substituted (for example, by a halogen) such as methane- or p-toluene sulfonic acid. Salts which are not pharmaceutically or veterinarily acceptable may still be valuable as intermediates.

Preferred salts include, for example, acetate, trifluoroacetate, lactate, gluconate, citrate, tartrate, maleate, malate, pantothenate, adipate, alginate, aspartate, benzoate, butyrate, digluconate, cyclopentanate, glucoheptanate, glycerophosphate, oxalate, heptanoate, hexanoate, fumarate, nicotinate, palmoate, pectinate, 3-phenylpropionate, picrate, pivalate, proprionate, tartrate, lactobionate, pivolate, camphorate, undecanoate and succinate, organic sulfonic acids such as methanesulfonate, ethanesulfonate, 2-

hydroxyethane sulfonate, camphorsulfonate, 2-naphthalenesulfonate, benzenesulfonate, p-chlorobenzenesulfonate and p-toluenesulfonate; and inorganic acids such as hydrochloride, hydrobromide, hydroiodide, sulfate, bisulfate, hemisulfate, thiocyanate, persulfate, phosphoric and sulfonic acids.

5

Esters are formed either using organic acids or alcohols/hydroxides, depending on the functional group being esterified. Organic acids include carboxylic acids, such as alkanecarboxylic acids of 1 to 12 carbon atoms which are unsubstituted or substituted (e.g., by halogen), such as acetic acid; with saturated or unsaturated dicarboxylic acid, 10 for example oxalic, malonic, succinic, maleic, fumaric, phthalic or tetraphthalic; with hydroxycarboxylic acids, for example ascorbic, glycolic, lactic, malic, tartaric or citric acid; with amino acids, for example aspartic or glutamic acid; with benzoic acid; or with organic sulfonic acids, such as (C₁-C₄)-alkyl- or aryl-sulfonic acids which are unsubstituted or substituted (for example, by a halogen) such as methane- or p-toluene 15 sulfonic acid. Suitable hydroxides include inorganic hydroxides, such as sodium hydroxide, potassium hydroxide, calcium hydroxide, aluminium hydroxide. Alcohols include alkane alcohols of 1-12 carbon atoms which may be unsubstituted or substituted, (e.g. by a halogen).

20 ENANTIOMERS/TAUTOMERS

In all aspects of the present invention previously discussed, the invention includes, where appropriate all enantiomers, diastereoisomers and tautomers of the compounds of the invention. The person skilled in the art will recognise compounds that possess optical properties (one or more chiral carbon atoms) or tautomeric characteristics. The 25 corresponding enantiomers and/or tautomers may be isolated/prepared by methods known in the art.

Enantiomers are characterised by the absolute configuration of their chiral centres and described by the *R*- and *S*-sequencing rules of Cahn, Ingold and Prelog. Such 30 conventions are well known in the art (e.g. see 'Advanced Organic Chemistry', 3rd edition, ed. March, J., John Wiley and Sons, New York, 1985).

Compounds of the invention containing a chiral centre may be used as a racemic mixture, an enantiomerically enriched mixture, or the racemic mixture may be separated using well-known techniques and an individual enantiomer may be used alone.

5 STEREO AND GEOMETRIC ISOMERS

Some of the compounds of the invention may exist as stereoisomers and/or geometric isomers – e.g. they may possess one or more asymmetric and/or geometric centres and so may exist in two or more stereoisomeric and/or geometric forms. The present invention contemplates the use of all the individual stereoisomers and geometric
10 isomers of those inhibitor agents, and mixtures thereof. The terms used in the claims encompass these forms, provided said forms retain the appropriate functional activity (though not necessarily to the same degree).

ISOTOPIC VARIATIONS

15 The present invention also includes all suitable isotopic variations of the agent or a pharmaceutically acceptable salt thereof. An isotopic variation of an agent of the present invention or a pharmaceutically acceptable salt thereof is defined as one in which at least one atom is replaced by an atom having the same atomic number but an atomic mass different from the atomic mass usually found in nature. Examples of
20 isotopes that can be incorporated into the agent and pharmaceutically acceptable salts thereof include isotopes of hydrogen, carbon, nitrogen, oxygen, phosphorus, sulfur, fluorine and chlorine such as ^2H , ^3H , ^{13}C , ^{14}C , ^{15}N , ^{17}O , ^{18}O , ^{31}P , ^{32}P , ^{35}S , ^{18}F and ^{36}Cl , respectively. Certain isotopic variations of the agent and pharmaceutically acceptable salts thereof, for example, those in which a radioactive isotope such as ^3H or ^{14}C is
25 incorporated, are useful in drug and/or substrate tissue distribution studies. Tritiated, i.e., ^3H , and carbon-14, i.e., ^{14}C , isotopes are particularly preferred for their ease of preparation and detectability. Further, substitution with isotopes such as deuterium, i.e., ^2H , may afford certain therapeutic advantages resulting from greater metabolic stability, for example, increased *in vivo* half-life or reduced dosage requirements and hence may
30 be preferred in some circumstances. For example, the invention includes compounds of general formula (I) where any hydrogen atom has been replaced by a deuterium atom. Isotopic variations of the agent of the present invention and pharmaceutically acceptable salts thereof of this invention can generally be prepared by conventional procedures using appropriate isotopic variations of suitable reagents.

PRODRUGS

The invention further includes the compounds of the present invention in prodrug form, i.e. covalently bonded compounds which release the active parent drug according to general formula (I) *in vivo*. Such prodrugs are generally compounds of the invention
5 wherein one or more appropriate groups have been modified such that the modification may be reversed upon administration to a human or mammalian subject. Reversion is usually performed by an enzyme naturally present in such subject, though it is possible for a second agent to be administered together with such a prodrug in order to perform the reversion *in vivo*. Examples of such modifications include esters (for example, any
10 of those described above), wherein the reversion may be carried out by an esterase etc. Other such systems will be well known to those skilled in the art.

SOLVATES

The present invention also includes solvate forms of the compounds of the present
15 invention. The terms used in the claims encompass these forms.

POLYMORPHS

The invention further relates to the compounds of the present invention in their various crystalline forms, polymorphic forms and (an)hydrous forms. It is well established within
20 the pharmaceutical industry that chemical compounds may be isolated in any of such forms by slightly varying the method of purification and or isolation from the solvents used in the synthetic preparation of such compounds.

ADMINISTRATION

The pharmaceutical compositions of the present invention may be adapted for rectal, nasal, intrabronchial, topical (including buccal and sublingual), vaginal or parenteral (including subcutaneous, intramuscular, intravenous, intraarterial and intradermal), intraperitoneal or intrathecal administration. Preferably the formulation is an orally administered formulation. The formulations may conveniently be presented in unit
25 dosage form, i.e., in the form of discrete portions containing a unit dose, or a multiple or sub-unit of a unit dose. By way of example, the formulations may be in the form of tablets and sustained release capsules, and may be prepared by any method well
30 known in the art of pharmacy.

Formulations for oral administration in the present invention may be presented as: discrete units such as capsules, gellules, drops, cachets, pills or tablets each containing a predetermined amount of the active agent; as a powder or granules; as a solution, emulsion or a suspension of the active agent in an aqueous liquid or a non-aqueous liquid; or as an oil-in-water liquid emulsion or a water-in-oil liquid emulsion; or as a bolus etc. Preferably, these compositions contain from 1 to 250 mg and more preferably from 10-100 mg, of active ingredient per dose.

For compositions for oral administration (e.g. tablets and capsules), the term “acceptable carrier” includes vehicles such as common excipients e.g. binding agents, for example syrup, acacia, gelatin, sorbitol, tragacanth, polyvinylpyrrolidone (Povidone), methylcellulose, ethylcellulose, sodium carboxymethylcellulose, hydroxypropyl-methylcellulose, sucrose and starch; fillers and carriers, for example corn starch, gelatin, lactose, sucrose, microcrystalline cellulose, kaolin, mannitol, dicalcium phosphate, sodium chloride and alginic acid; and lubricants such as magnesium stearate, sodium stearate and other metallic stearates, glycerol stearate stearic acid, silicone fluid, talc waxes, oils and colloidal silica. Flavouring agents such as peppermint, oil of wintergreen, cherry flavouring and the like can also be used. It may be desirable to add a colouring agent to make the dosage form readily identifiable. Tablets may also be coated by methods well known in the art.

A tablet may be made by compression or moulding, optionally with one or more accessory ingredients. Compressed tablets may be prepared by compressing in a suitable machine the active agent in a free flowing form such as a powder or granules, optionally mixed with a binder, lubricant, inert diluent, preservative, surface-active or dispersing agent. Moulded tablets may be made by moulding in a suitable machine a mixture of the powdered compound moistened with an inert liquid diluent. The tablets may be optionally coated or scored and may be formulated so as to provide slow or controlled release of the active agent.

Other formulations suitable for oral administration include lozenges comprising the active agent in a flavoured base, usually sucrose and acacia or tragacanth; pastilles comprising the active agent in an inert base such as gelatin and glycerin, or sucrose and acacia; and mouthwashes comprising the active agent in a suitable liquid carrier.

Other forms of administration comprise solutions or emulsions which may be injected intravenously, intraarterially, intrathecally, subcutaneously, intradermally, intraperitoneally or intramuscularly, and which are prepared from sterile or sterilisable solutions. Injectable forms typically contain between 10 - 1000 mg, preferably between 5 10 - 250 mg, of active ingredient per dose.

The pharmaceutical compositions of the present invention may also be in form of suppositories, pessaries, suspensions, emulsions, lotions, ointments, creams, gels, sprays, solutions or dusting powders.

10

An alternative means of transdermal administration is by use of a skin patch. For example, the active ingredient can be incorporated into a cream consisting of an aqueous emulsion of polyethylene glycols or liquid paraffin. The active ingredient can also be incorporated, at a concentration of between 1 and 10% by weight, into an ointment consisting of a white wax or white soft paraffin base together with such 15 stabilisers and preservatives as may be required.

DOSAGE

A person of ordinary skill in the art can easily determine an appropriate dose of one of the instant compositions to administer to a subject without undue experimentation. Typically, a physician will determine the actual dosage which will be most suitable for an individual patient and it will depend on a variety of factors including the activity of the specific compound employed, the metabolic stability and length of action of that compound, the age, body weight, general health, sex, diet, mode and time of 25 administration, rate of excretion, drug combination, the severity of the particular condition, and the individual undergoing therapy. The dosages disclosed herein are exemplary of the average case. There can of course be individual instances where higher or lower dosage ranges are merited, and such are within the scope of this invention.

30

In accordance with this invention, an effective amount of a compound of general formula (I) may be administered to inhibit PAICS. Of course, this dosage amount will further be modified according to the type of administration of the compound. For example, to achieve an "effective amount" for acute therapy, parenteral administration

of a compound of general formula (I) is preferred. An intravenous infusion of the compound in 5% dextrose in water or normal saline, or a similar formulation with suitable excipients, is most effective, although an intramuscular bolus injection is also useful. Typically, the parenteral dose will be about 0.01 to about 100 mg/kg; preferably
5 between 0.1 and 20 mg/kg, in a manner to maintain the concentration of drug in the plasma at a concentration effective to inhibit a kinase. The compounds may be administered one to four times daily at a level to achieve a total daily dose of about 0.4 to about 400 mg/kg/day. The precise amount of an inventive compound which is therapeutically effective, and the route by which such compound is best administered, is
10 readily determined by one of ordinary skill in the art by comparing the blood level of the agent to the concentration required to have a therapeutic effect.

The compounds of this invention may also be administered orally to the patient, in a manner such that the concentration of drug is sufficient to achieve one or more of the
15 therapeutic indications disclosed herein. Typically, a pharmaceutical composition containing the compound is administered at an oral dose of between about 0.1 to about 50 mg/kg in a manner consistent with the condition of the patient. Preferably the oral dose would be about 0.5 to about 20 mg/kg.

20 No unacceptable toxicological effects are expected when compounds of the present invention are administered in accordance with the present invention. The compounds of this invention, which may have good bioavailability, may be tested in one of several biological assays to determine the concentration of a compound which is required to have a given pharmacological effect.

25

COMBINATIONS

In a particularly preferred embodiment, the one or more compounds of the invention are administered in combination with one or more other active agents, for example, existing drugs available on the market. In such cases, the compounds of the invention may be
30 administered consecutively, simultaneously or sequentially with the one or more other active agents.

Drugs in general are more effective when used in combination. In particular, combination therapy is desirable in order to avoid an overlap of major toxicities,

mechanism of action and resistance mechanism(s). Furthermore, it is also desirable to administer most drugs at their maximum tolerated doses with minimum time intervals between such doses. The major advantages of combining chemotherapeutic drugs are that it may promote additive or possible synergistic effects through biochemical interactions and also may decrease or delay the emergence of resistance.

Beneficial combinations may be suggested by studying the inhibitory activity of the test compounds with agents known or suspected of being valuable in the treatment of a particular disorder. For example, the invention relates to the use of a compound as described above in an assay for identifying compounds that promote additive and synergistic activity upon anti-cancer activities when combined with the compound. Preferably the assay is a high-throughput cell based phenotypic screen. This procedure can also be used to determine the order of administration of the agents, i.e. before, simultaneously, or after delivery. Such scheduling may be a feature of all the active agents identified herein.

ASSAY

A further aspect of the invention relates to the use of a compound as described above in an assay for identifying further candidate compounds capable of inhibiting PAICS. Preferably, the candidate compound is capable of selectively inhibiting PAICS.

Preferably, the assay is a competitive binding assay.

More preferably, the competitive binding assay comprises contacting a compound of the invention with PAICS, and a candidate compound and detecting any change in the interaction between the compound according to the invention and the PAICS.

Preferably, the candidate compound is generated by conventional SAR modification of a compound of the invention.

As used herein, the term "conventional SAR modification" refers to standard methods known in the art for varying a given compound by way of chemical derivatisation.

Thus, in one aspect, the identified compound may act as a model (for example, a template) for the development of other compounds. The compounds employed in such

a test may be free in solution, affixed to a solid support, borne on a cell surface, or located intracellularly. The abolition of activity or the formation of binding complexes between the compound and the agent being tested may be measured.

- 5 The assay of the present invention may be a screen, whereby a number of agents are tested. In one aspect, the assay method of the present invention is a high throughput screen.

This invention also contemplates the use of competitive drug screening assays in which
10 neutralising antibodies capable of binding a compound specifically compete with a test compound for binding to a compound.

Another technique for screening provides for high throughput screening (HTS) of agents having suitable binding affinity to the substances and is based upon the method
15 described in detail in WO 84/03564.

It is expected that the assay methods of the present invention will be suitable for both small and large-scale screening of test compounds as well as in quantitative assays.

- 20 Preferably, the competitive binding assay comprises contacting a compound of the invention with a kinase in the presence of a known substrate of said kinase and detecting any change in the interaction between said kinase and said known substrate.

A further aspect of the invention provides a method of detecting the binding of a ligand
25 to PAICS, said method comprising the steps of:

- (i) contacting a ligand with PAICS in the presence of a known substrate of said kinase;
 - (ii) detecting any change in the interaction between the PAICS and said known substrate;
- 30 and wherein said ligand is a compound of the invention.

One aspect of the invention relates to a process comprising the steps of:

- (a) performing an assay method described hereinabove;
- (b) identifying one or more ligands capable of binding to a ligand binding domain;
and
- (c) preparing a quantity of said one or more ligands.

5

Another aspect of the invention provides a process comprising the steps of:

- (a) performing an assay method described hereinabove;
- (b) identifying one or more ligands capable of binding to a ligand binding domain;
and
- 10 (c) preparing a pharmaceutical composition comprising said one or more ligands.

Another aspect of the invention provides a process comprising the steps of:

- (a) performing an assay method described hereinabove;
- (b) identifying one or more ligands capable of binding to a ligand binding domain;
- 15 (c) modifying said one or more ligands capable of binding to a ligand binding domain;
- (d) performing the assay method described hereinabove;
- (e) optionally preparing a pharmaceutical composition comprising said one or more ligands.

20

The invention also relates to a ligand identified by the method described hereinabove.

Yet another aspect of the invention relates to a pharmaceutical composition comprising a ligand identified by the method described hereinabove.

25

Another aspect of the invention relates to the use of a ligand identified by the method described hereinabove in the preparation of a pharmaceutical composition for use in the treatment of one or more disorders as described hereinabove.

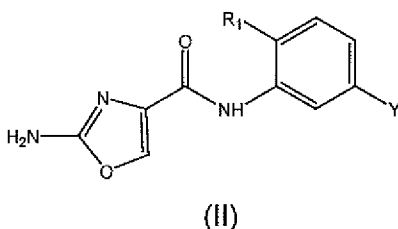
- 30 The above methods may be used to screen for a ligand useful as an inhibitor of one or more kinases.

Compounds of general formula (I) are useful both as laboratory tools and as therapeutic agents. In the laboratory certain compounds of the invention are useful in establishing whether a known or newly discovered protein contributes a critical or at least significant biochemical function during the establishment or progression of a disease state, a
5 process commonly referred to as 'target validation'.

SYNTHESIS

Another aspect of the invention relates to a process for preparing a compound of formula (I) as defined above, said process comprising the steps of:

10 (i) preparing an intermediate of formula (II):



15 wherein R¹ is defined in claim 1 and Y is selected from halogen (more preferably I), boronic acid and a boronate ester; and
(ii) converting said intermediate of formula (II) to a compound of formula (I).

The invention is further described by way of the following non-limiting examples, and
20 with reference to the following figures, wherein:

Figure 1 shows the domains of phosphoribosylaminoimidazole carboxylase, phosphoribosylaminoimidazole succinocarboxamide synthetase, and its role in *de novo* purine biosynthesis.
25

Figure 2 shows the reaction catalysed by human PAICS.

Figure 3 shows the Transcreener FI assay principle.
30

EXAMPLES

Materials and Methods

General procedures for synthesis of compounds

5 Chromatography

Preparative high pressure liquid chromatography was carried out using apparatus made by Agilent. The apparatus is constructed such that the chromatography is monitored by a multi-wavelength UV detector (G1365B manufactured by Agilent) and an MM-ES+APCI mass spectrometer (G-1956A, manufactured by Agilent) connected in series, and if the appropriate criteria are met the sample is collected by an automated fraction collector (G1364B manufactured by Agilent). Collection can be triggered by any combination of UV or mass spectrometry or can be based on time. Typical conditions for the separation process are as follows: Chromatography column was an Xbridge C-18 (19 x 100 mm); the gradient was run over a 7 minute period at a flow rate of 40 ml / min (gradient at start: 10% MeOH and 90% water, gradient at finish: 100% MeOH and 0% water; as buffer: either 0.1% formic acid, 0.1% ammonium hydroxide or 0.1% TFA was added to the water). It will be appreciated by those skilled in the art that it may be necessary or desirable to modify the conditions for each specific compound, for example by changing the solvent composition at the start or at the end, modifying the solvents or buffers, changing the run time, changing the flow rate and/or the chromatography column. Flash chromatography refers to silica gel chromatography and was carried out using an SP4 or an Isolera 4 MPLC system (manufactured by Biotage) and pre-packed silica gel cartridges (supplied by Biotage); or alternatively using conventional glass column chromatography.

25

Analytical Methods

¹H Nuclear Magnetic Resonance (NMR) spectra were typically recorded using an ECX400 spectrometer (manufactured by JEOL) in the stated solvent at around rt unless otherwise stated. In all cases, NMR data were consistent with the proposed structures. Characteristic chemical shifts (δ) are given in parts-per-million using conventional abbreviations for designation of major peaks: e.g. s, singlet; d, doublet; t, triplet; q, quartet; dd, doublet of doublets; br, broad.

30

Analytical LCMS was typically carried out using an Agilent HPLC instrument with C-18 Xbridge column (3.5 μm , 4.6 x 30 mm, gradient at start: 10% organic phase and 90% water, gradient at finish: organic and 0% water; as buffer: either 0.1% ammonium hydroxide or 0.1% TFA was added to the water). The organic solvent was either MeCN
5 or MeOH. A flow rate of 3 mL/min was used with UV detection at 254 and 210 nm. Mass spectra were recorded using a MM-ES+APCI mass spectrometer (G-1956A, manufactured by Agilent).

Compound Preparation

10 Where the preparation of starting materials is not described, these are commercially available, known in the literature, or readily obtainable by those skilled in the art using standard procedures. Where it is indicated that compounds were prepared analogously to earlier examples or intermediates, it will be appreciated by the skilled person that the reaction time, number of equivalents of reagents, solvent, concentration and
15 temperature can be modified for each specific reaction and that it may be necessary or desirable to employ different work-up or purification techniques.

Where reactions are carried out using microwave irradiation, the microwave used is an Initiator 60 supplied by Biotage. The actual power supplied varies during the course of
20 the reaction in order to maintain a constant temperature.

Some hydrogenations were carried out using an H-Cube® Continuous-flow Hydrogenation Reactor manufactured by ThalesNano. The catalysts are supplied by ThalesNano as "CatCarts" cartridges. The pressure, flow rate, temperature and
25 cartridge are indicated in the experimental section. The equipment was used in accordance with the manufacturer operating procedure. The person skilled in the art will appreciate that it may be necessary or desirable to run repeat cycles of the reaction mixture and in some instances, replace the cartridge between cycles to improve the yield of the reaction.

30

Abbreviations

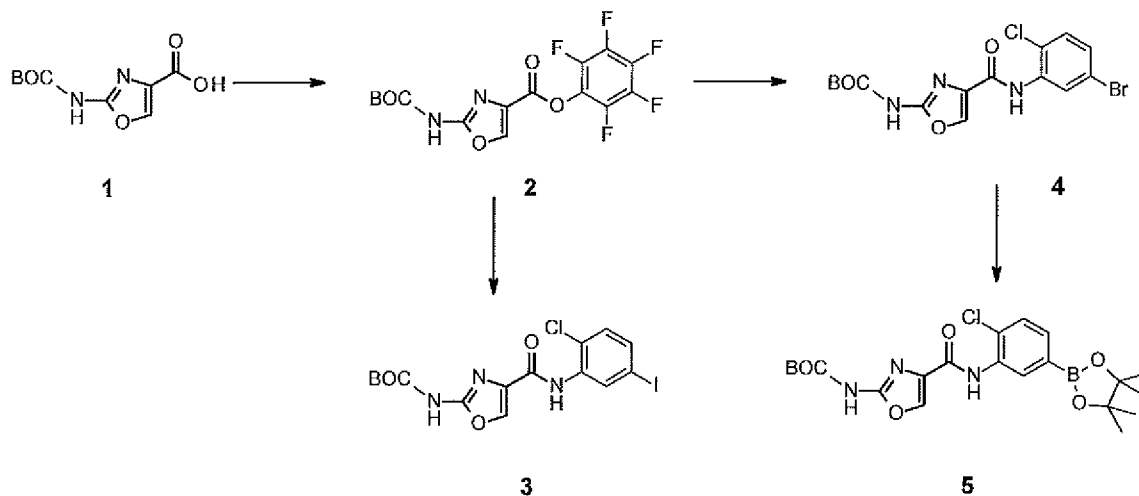
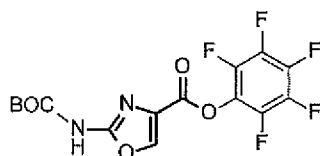
A list of some common abbreviations is shown below – where other abbreviations are used which are not listed, these will be understood by the person skilled in the art.

	AcOH	= Acetic acid
	BOC	= <i>tert</i> -Butyloxycarbonyl
	DCC	= 1,3-Dicyclohexylcarbodiimide
	DCM	= Dichloromethane
5	DIPEA	= N,N-diisopropylethylamine
	DMF	= N,N-Dimethylformamide
	DMSO	= Dimethylsulfoxide
	Et ₂ O	= Diethyl ether
	EtOAc	= Ethyl acetate
10	EtOH	= Ethanol
	HATU	= N,N,N',N'-Tetramethyl-O-(7-azabenzotriazol-1-yl)uranium-hexafluorophosphate
	LCMS	= Liquid Chromatography Mass Spectrometry
	MeCN	= Acetonitrile
15	MeOH	= Methanol
	MgSO ₄	= Magnesium sulfate
	Na ₂ SO ₄	= Sodium sulfate
	NH ₃	= Ammonia
	NH ₄ Cl	= Ammonium chloride
20	NaHCO ₃	= Sodium bicarbonate
	PdCl ₂ (dppf)	= [1,1'-bis(diphenylphosphino)ferrocene]dichloropalladium(II) dichloromethane adduct
	Pd(Ph ₃ P) ₄	= Tetrakis(triphenylphosphine)palladium(0)
	Pet ether	= 40/60 petroleum ether
25	rt	= Room temperature
	SCX	= Strong cation exchange
	TEA	= Triethylamine
	TFA	= Trifluoroacetic acid
	THF	= Tetrahydrofuran

30

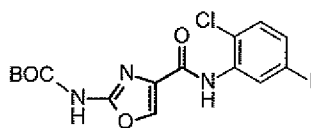
The synthesis of selected compounds of the invention is described below.

Scheme 1 – Preparation of intermediates 3-5

5 Pentafluorophenyl 2-(*tert*-butoxycarbonylamino) oxazole-4-carboxylate (2)

To a solution of 2-(*tert*-butoxycarbonylamino) oxazole-4-carboxylic acid **1** (1.5 g, 6.57
 10 mmol) in DMF (10 mL) was added 2,3,4,5,6-pentafluorophenol (0.96 g, 5.26 mmol),
 followed by DCC (2.03 g, 9.86 mmol) at 0°C and stirred the resulting mixture for 6h at rt.
 The reaction mixture was filtered to remove precipitated urea, charged ice cold water
 (50 mL) into filtrate and extracted with EtOAc (2 x 25 mL). The combined organic layer
 was washed with ice cold water (2 x 25 mL), the organic layer was dried over Na₂SO₄
 15 and concentrated *in vacuo*. The crude compound was purified by column
 chromatography (silica gel, 100-200 mesh, eluted with 20% EtOAc/pet ether) to obtain
 compound **2** (1.2 g, 46%) as a yellow solid; ¹H NMR (300 MHz, CDCl₃) δ ppm 8.21 (s,
 1H), 1.58 (s, 9H); LCMS (*m/z*): 395 [M+H]⁺.

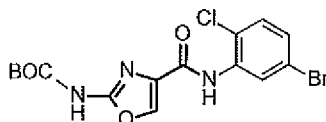
20 *tert*-Butyl 4-(2-chloro-5-iodophenylcarbamoyl) oxazol-2-ylcarbamate (3)



To a stirred solution of 2-chloro-5-iodoaniline (5 g, 19.8 mmol) in THF (50 mL) was added 60% sodium hydride (4.74 g, 98.9 mmol) at 0°C and stirred at rt for 1h. The reaction mixture was again cooled to 0°C, added pentafluorophenyl 2-(*tert*-butoxycarbonylamino) oxazole-4-carboxylate **2** (9.3 g, 23.7 mmol) lot-wise and stirred at
 5 rt for 3h. The reaction mixture was cooled to 0°C and quenched with ice-water. The reaction mixture was diluted with EtOAc (500 mL) and washed with water (200 mL), brine solution (200 mL), dried (over Na₂SO₄) and evaporated. The obtained crude compound was purified by column chromatography (silica gel, eluted with 20% EtOAc/pet ether) to obtain **3** (5.2 g, 59%) as an off-white solid; ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 11.06 (s, 1H), 9.40 (s, 1H), 8.55 (s, 1H), 8.50 (d, *J*=2 Hz, 1H), 7.55 (dd, *J*=8.4, 2.0 Hz, 1H), 7.36 (d, *J*=8.4 Hz, 1H), 1.47 (s, 9H); LCMS (*m/z*): 464/466 [M+H]⁺.

15

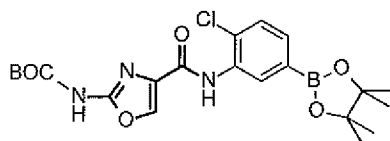
***tert*-Butyl (4-((5-bromo-2-chlorophenyl)carbamoyl)oxazol-2-yl)carbamate (4)**



To a solution of 5-bromo-2-chloroaniline (1.37 g, 6.34 mmol) in THF (20 mL) was added lithium hexamethyldisilazide (19 mL, 19 mmol) over 5 minutes at rt. The mixture was stirred for 1h, then a solution of pentafluorophenyl 2-((*tert*-butoxycarbonyl)amino)oxazole-4-carboxylate **2** (2.5 g, 6.34 mmol) in THF (20 mL) was added dropwise. The vessel contents were stirred at rt for 1h and then quenched with
 25 water (20 mL). The mixture was extracted with EtOAc (50 mL) and the organic layer washed with NH₄Cl solution (20 mL). Organic layer was concentrated *in vacuo* and the residue triturated in EtOAc (10 mL) to afford **4** (1.66 g, 63% yield); ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.43 (br s, 1H), 8.57 (s, 1H), 8.35 (d, *J*=2.3 Hz, 1H), 7.52 (d, *J*=8.6 Hz, 1H), 7.40 (dd, *J*=8.6, 2.4 Hz, 1H), 1.46 (s, 9H); LCMS (*m/z*): 440/442 [M+Na]⁺.

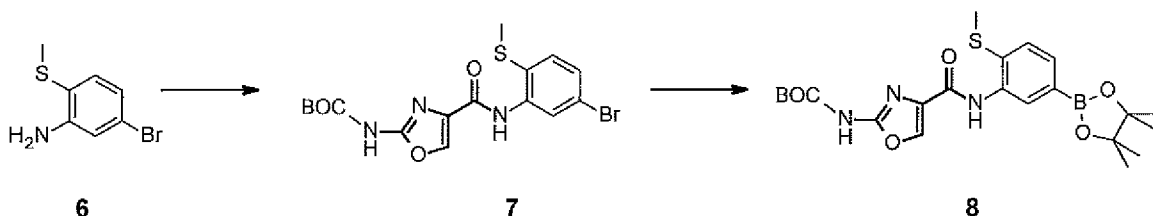
30

***tert*-Butyl (4-((2-chloro-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)carbamoyl)oxazol-2-yl)carbamate (5)**



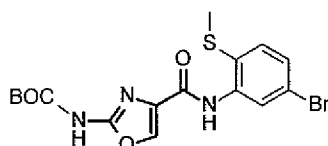
- 5 A mixture of *tert*-butyl (4-((5-bromo-2-chlorophenyl)carbamoyl)oxazol-2-yl)carbamate **4** (1.66 g, 3.98 mmol), potassium acetate (977 mg, 9.96 mmol) and bispinacolatodiboron (1.11 g, 4.38 mmol) in dioxane (40 mL) was degassed with nitrogen (sparge, 15 min). PdCl₂(dppf) (326 mg, 0.4 mmol) was added and the reaction heated to reflux for 1h. The reaction was cooled to rt and diluted with EtOAc (ca 100 mL). The reaction mixture
- 10 was washed with saturated NH₄Cl solution (100 mL). The organic phase was concentrated under reduced pressure and the crude material was filtered through a silica plug, eluting with EtOAc. The residue was dried azeotropically with DCM to afford **5** (1.96 g, quant yield); ¹H NMR (400 MHz, CDCl₃) δ ppm 9.11 (br s, 1H), 8.84 (d, *J*=1.4 Hz, 1H), 8.03 (s, 1H), 7.49 (dd, *J*=8.0, 1.5 Hz, 1H), 7.40 (d, *J*=7.9 Hz, 1H, 7.29 (s, 1H),
- 15 1.55 (s, 9H), 1.33 (s, 12H); LCMS (*m/z*): 464/466 [M+H]⁺.

Scheme 2 – Preparation of intermediates 7 and 8



20

***tert*-Butyl (4-((5-bromo-2-(methylthio)phenyl)carbamoyl)oxazol-2-yl)carbamate (7)**



- 25 To an ice cooled solution of 5-bromo-2-(methylthio)aniline **6** (4.62 g, 21.2 mmol in THF (50 mL) was added a solution of lithium hexamethyldisilazide (58 mL, 58 mmol, 1M).

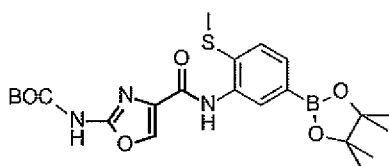
The reaction mixture was warmed to rt and stirred for 1h. A solution of pentafluorophenyl 2-((*tert*-butoxycarbonyl)amino)oxazole-4-carboxylate **2** (7.6 g, 19.3 mmol, 1 eq) in THF (50 mL) was added dropwise. The reaction mixture was stirred for an additional 3h at rt and was then quenched with saturated NH₄Cl solution (100 mL).

5 The reaction mixture was then extracted with EtOAc (3 x 100 mL) and the combined organics were washed with brine (100 mL). The organic phase was dried over MgSO₄, filtered and concentrated *in vacuo* to afford crude material. Purification by dry flash chromatography (50% heptanes in DCM) afforded 15 g of waxy solid which was further purified by trituration in *tert*-butyl methyl ether/heptanes to afford **7** (5.6 g, 66%) in 3

10 batches; ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 11.01 (s, 1H), 9.54 (s, 1H), 8.54 (s, 1H), 8.26 (d, *J*=2.2 Hz, 1H), 7.45 (d, *J*=8.5 Hz, 1H), 7.37 (dd, *J*=8.4, 2.2 Hz, 1H), 2.44 (s, 3H), 1.46 (s, 9H); LCMS (*m/z*): 428/430 [M+H]⁺.

***tert*-Butyl (4-((2-(methylthio)-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)carbamoyl)oxazol-2-yl)carbamate (8)**

15

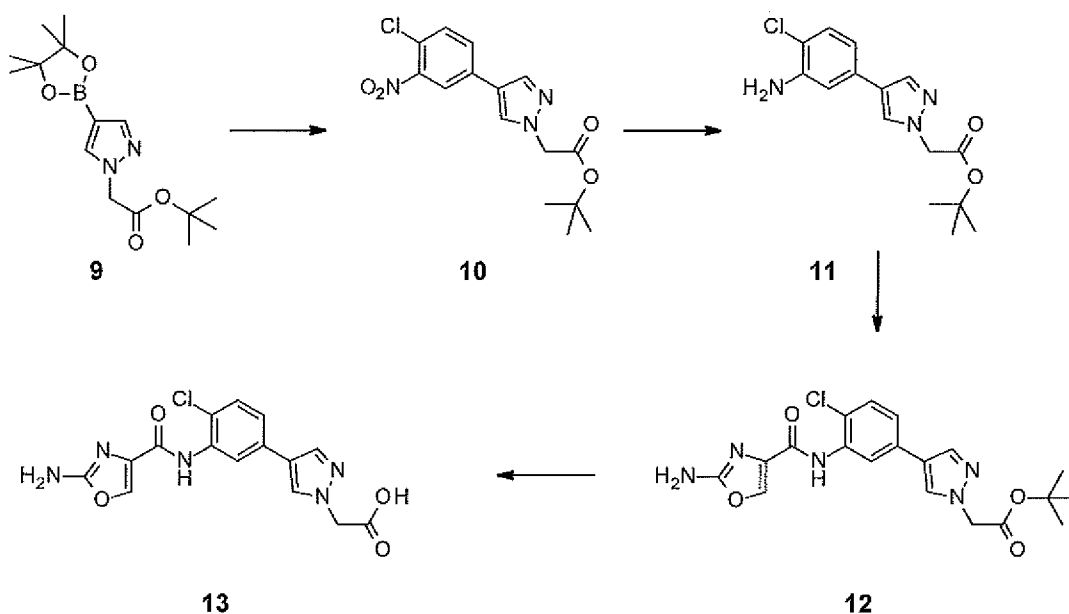
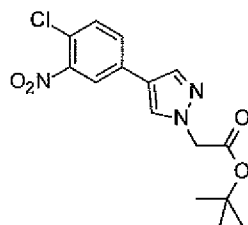


A solution of *tert*-butyl (4-((5-bromo-2-(methylthio)phenyl)carbamoyl)oxazol-2-yl)carbamate **7** (4.42 g, 9.89 mmol), potassium acetate (2.42 g, 24.7 mmol) and bispinacolatodiboron (2.76 g, 10.9 mmol) in dioxane (100 mL) was degassed with nitrogen (sparge, 15 min). PdCl₂(dppf) (3.6 g, 4.94 mmol) was added and the reaction was boiled under reflux for 1h. The mixture was cooled to rt and dicalite (15 g) was added. After stirring for 5 minutes the mixture was filtered and washed with EtOAc (ca

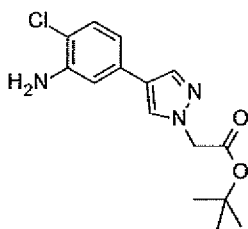
20 150 mL). The combined filtrates were washed with saturated NH₄Cl solution (100 mL) and brine (100 mL). The organic phase was dried over MgSO₄, filtered and solvent removed *in vacuo*. The crude material was purified by column chromatography (50% EtOAc in heptane) and then triturated in heptane with 1 drop of *tert*-butyl methyl ether to afford **8** (1.28 g, 26%) as an off white solid; ¹H NMR (400 MHz, CDCl₃) δ ppm 9.32 (s, 1H), 8.67 (s, 1H), 8.03 (d, *J*=2.5 Hz, 1H), 7.65 – 7.28 (m, 2H), 7.26 (d, *J*=2.4 Hz, 1H), 2.44 (s, 3H), 1.54 (s, 9H), 1.32 (s, 12H); LCMS (*m/z*): 476 [M+H]⁺.

30

Scheme 3 – Preparation of intermediate 13

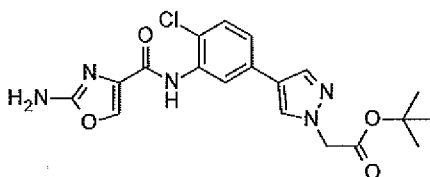
5 ***tert*-Butyl 2-(4-(4-chloro-3-nitrophenyl)-1H-pyrazol-1-yl)acetate (10)**

To a stirred solution of *tert*-butyl 2-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-1H-pyrazol-1-yl)acetate **9** (10 g, 32.5 mmol), 4-bromo-1-chloro-2-nitrobenzene (7.6 g, 32.5 mmol) in dioxane-water (2:1) (150 mL) was added potassium carbonate (6.7 g, 48.7 mmol) and PdCl₂(dppf) (2.3 g, 3.24 mmol) at rt and the reaction heated to 90°C for 16h. The reaction mixture was diluted with EtOAc (500 mL) and washed with water (2 x 100mL), brine solution (100 mL), dried (Na₂SO₄) and evaporated. The obtained crude compound was purified by column chromatography (100-200 mesh silica gel column, 25% EtOAc/pet ether) to obtain **10** (7.5 g, 68%) as a pale yellow solid; ¹H NMR (300 MHz, CDCl₃) δ ppm 7.96 (d, *J*=2.1 Hz, 1H), 7.83 (s, 1H), 7.80 (s, 1H), 7.62 (dd, *J*=2.1, 8.4 Hz, 1H), 7.53 (d, *J*=8.4 Hz, 1H), 4.86 (s, 2H), 1.49 (s, 9H); LCMS (*m/z*): 282/284 [M+H]⁺.

***tert*-Butyl 2-(4-(3-amino-4-chlorophenyl)-1H-pyrazol-1-yl)acetate (11)**

5 To a stirred solution of *tert*-butyl 2-(4-(4-chloro-3-nitrophenyl)-1H-pyrazol-1-yl) acetate **10** (2.5 g, 7.41 mmol) in EtOH (20 mL) and water (12 mL) was added iron powder (1.29 g, 22.2 mmol), NH₄Cl (2.35 g, 44.5 mmol) at rt and the reaction heated to 80°C for 5h. The reaction mixture was filtered through a bed of celite and the filtrate partitioned between EtOAc (300 mL) and water (100 mL). The organic layer was dried (over
 10 Na₂SO₄) and evaporated to dryness. The obtained crude compound was purified by column chromatography (100-200 mesh silica gel column, 50% EtOAc/pet ether) to obtain compound **11** (1.5 g, 65%) as a pale-yellow liquid; ¹H NMR (300 MHz, CDCl₃) δ ppm 7.75 (s, 1H), 7.65 (s, 1H), 7.21 (d, *J*=8.1 Hz, 1H), 6.89 (s, 1H), 6.80 (dd, *J*=8.4, 2.1 Hz, 1H), 4.02 (s, 2H), 4.05 (br s, 2H), 1.48 (s, 9H); LCMS (*m/z*): 308/310 [M+H]⁺.

15

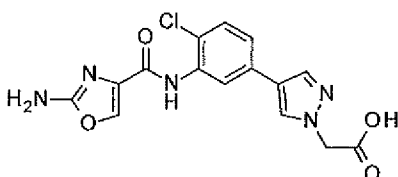
***tert*-Butyl 2-(4-(3-(2-aminooxazole-4-carboxamido)-4-chlorophenyl)-1H-pyrazol-1-yl)acetate (12)**

20

To a stirred solution of 2-aminooxazole-4-carboxylic acid (2.08 g, 16.3 mmol) in DMF (20 mL) was added *tert*-butyl 2-(4-(3-amino-4-chlorophenyl)-1H-pyrazol-1-yl)acetate **11** (5 g, 16.3 mmol), HATU (9.26 g, 26.5 mmol) and DIPEA (5.5 mL, 32.6 mmol) at 0°C and the mixture stirred at rt for 24h. The reaction mixture was partitioned between
 25 EtOAc (200 mL) and water (3 x 100mL). The organic layer was dried (over Na₂SO₄) and evaporated to dryness. Purification by column chromatography (100-200 mesh silica gel, 50% EtOAc/pet ether) gave **12** (2.5 g, 41%) as an off white solid; ¹H NMR (400

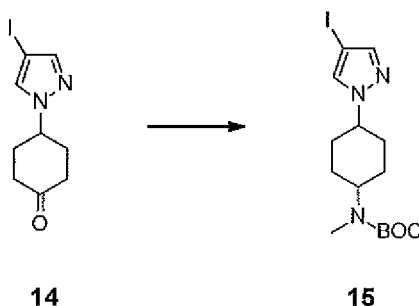
MHz, DMSO- d_6) δ ppm 9.24 (s, 1H), 8.47 (d, $J=2.4$ Hz, 1H), 8.18 (s, 1H), 8.09 (s, 1H), 7.89 (s, 1H), 7.53 (d, $J=8.0$ Hz, 1H), 7.40 (dd, $J=8.4, 2.0$ Hz, 1H), 7.14 (s, 2H), 4.97 (s, 2H), 1.43 (s, 9H); LCMS (m/z): 418/420 [$M+H$] $^+$.

5 **2-(4-(3-(2-Aminooxazole-4-carboxamido)-4-chlorophenyl)-1H-pyrazol-1-yl)acetic acid (13)**



10 To a stirred solution of *tert*-butyl 2-(4-(3-(2-aminooxazole-4-carboxamido)-4-chlorophenyl)-1H-pyrazol-1-yl)acetate **12** (2.8 g, 6.17 mmol) in DCM (28 mL) was added TFA (7 mL) at 0°C and stirred at rt for 16h. The reaction mixture was evaporated to dryness, washed with Et₂O and dried to obtain **13** (2.35, 73%) as an off white solid and as the TFA salt; ¹H NMR (400 MHz, DMSO- d_6) δ ppm 13.02 (s, 1H), 9.25 (s, 1H),
 15 8.46 (d, $J=2.4$ Hz, 1H), 8.18 (s, 1H), 8.09 (s, 1H), 7.88 (s, 1H), 7.53 (d, $J=8.0$ Hz, 1H), 7.40 (dd, $J=8.4, 2.0$ Hz, 1H), 7.16 (br s, 2H), 4.98 (s, 2H); LCMS (m/z): 362/364 [$M+H$] $^+$.

Scheme 4 – Preparation of intermediate 15



20

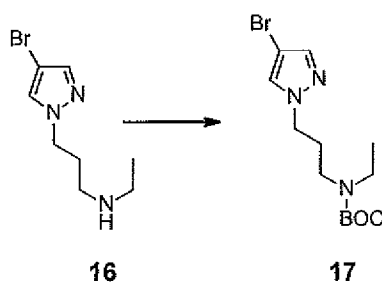
***tert*-Butyl [4-(4-iodo-1H-pyrazol-1-yl)cyclohexyl]methylcarbamate (15)**

A solution of 4-(4-iodo-1H-pyrazol-1-yl)cyclohexanone **14** (200 mg, 0.69 mmol) in DCM (2 mL) was treated with methylamine hydrochloride (56 mg, 0.83 mmol) and AcOH (0.09 mL, 1.52 mmol) and stirred at rt for 0.5h. Then sodium triacetoxyborohydride (321
 25

mg, 1.52 mmol) was added and the reaction stirred at rt for a further 18h. It was then basified with 1M sodium hydroxide solution (~ 7 mL) and extracted with DCM (4 x 10 mL). The combined organic extracts were dried (MgSO₄) and evaporated *in vacuo* to give the crude product. This was dissolved in dioxane (5 mL) and treated with TEA (0.38 mL, 2.76 mmol) and di-*tert*-butyldicarbonate (264 mg, 1.38 mmol) and stirred at rt for 18h. Concentration *in vacuo* and column chromatography (2 – 100% EtOAc / pet ether gradient) gave **15** (121 mg, 43% over 2 steps) as a colourless oil and an inseparable mixture of *cis*- and *trans*- isomers; ¹H NMR (400 MHz, CD₃OD) δ ppm 7.91 (s, 1H, isomer A), 7.81 (s, 1H, isomer B), 7.55 (s, 1H, isomer A), 7.51 (s, 1H, isomer B), 4.40 – 4.36 (m, 1H, isomer A), 4.24 – 4.18 (m, 1H, isomer B), 4.03 – 3.92 (m, 1H isomer A and 1H isomer B), 2.80 (s, 3H, isomer B), 2.69 (s, 3H, isomer A), 2.58 – 2.51 (m, 2H, isomer A), 2.19 – 2.14 (m, 2H, isomer B), 2.10 – 1.88 and 1.83 – 1.74 (total 4H isomer A and 4H isomer B), 1.59 – 1.50 (total 2H isomer A and 2H isomer B), 1.50 (s, 9H, isomer A), 1.47 (s, 9H, isomer B); LCMS (*m/z*): 406 [M+H]⁺.

15

Scheme 5 – Preparation of intermediate 17



20 ***tert*-Butyl (3-(4-bromo-1H-pyrazol-1-yl)propyl)(ethyl)carbamate (17)**

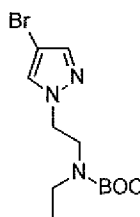
To a solution of 3-(4-bromo-1H-pyrazol-1-yl)-*N*-ethylpropan-1-amine **16** (310 mg, 1.34 mmol) in DCM (15 mL) was added TEA (162 mg, 1.60 mmol), di-*tert*-butyl-dicarbonate (321 mg, 1.47 mmol) and *N,N*-dimethylaminopyridine (cat). The reaction mixture was stirred for 18h at rt. The reaction mixture was concentrated and a solution of the residue in EtOAc (20 mL) was washed with NH₄Cl solution (20 mL). The solvent was removed *in vacuo* and purified by column chromatography (70% EtOAc in heptanes) to give **17** (280 mg, 63%) as a pale yellow oil; ¹H NMR (400 MHz, CDCl₃) δ ppm 7.46 (s, 2H), 4.12

(t, $J=7.0$ Hz, 2H), 3.31 – 3.05 (m, 4H), 2.15 – 1.97 (m, 2H), 1.45 (s, 9H), 1.07 (t, $J=7.0$ Hz, 3H); LCMS (m/z): 232/234 [(M-BOC)+H]⁺.

The following intermediates were prepared using an analogous procedure:

5

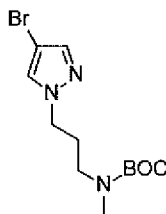
tert-Butyl [2-(4-bromo-1H-pyrazol-1-yl)ethyl]ethylcarbamate (18)



10 ¹H NMR (400 MHz, CDCl₃) δ ppm 7.47 (s, 1H), 7.36 (s, 1H), 4.31 – 4.16 (m, 2H), 3.55 (t, $J=6.1$ Hz, 2H), 3.10 – 2.85 (m, 2H), 1.45 (s, 9H), 1.08 – 0.87 (m, 3H); LCMS (m/z): 218/220 [(M-BOC)+H]⁺.

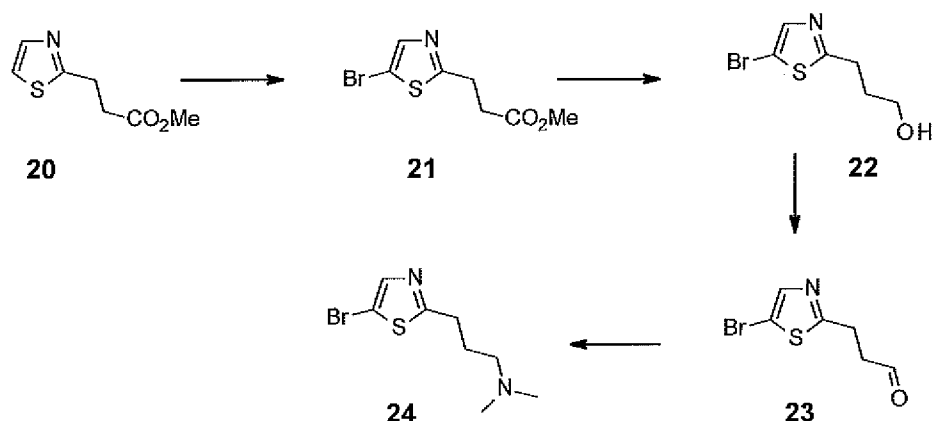
tert-Butyl [3-(4-bromo-1H-pyrazol-1-yl)propyl]methylcarbamate (19)

15

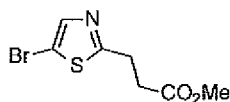


20 ¹H NMR (400 MHz, CDCl₃) δ ppm 7.46 (s, 2H), 4.11 (t, $J=7.0$ Hz, 2H), 3.25 (t, $J=6.8$ Hz, 2H), 2.82 (s, 3H), 2.14 – 2.01 (m, 2H), 1.45 (s, 9H); LCMS (m/z): 218/220 [(M-BOC)+H]⁺.

Scheme 6 – Preparation of intermediate 24



Methyl 3-(5-bromothiazol-2-yl)propanoate (21)

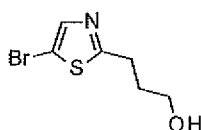


5

To a solution of methyl 3-(thiazol-2-yl)propanoate **20** (832 mg, 4.86 mmol) in DMF (15 mL) was added N-bromosuccinimide (1.03 g, 5.83 mmol). The mixture was stirred at rt. After 4h LCMS indicated that reaction was incomplete so a further 150 mg (0.84 mmol) of N-bromosuccinimide was added. After a further 1h at rt the reaction mixture was diluted with water (60 mL) and the product was extracted with *tert*-butyl methyl ether (2 x 30 mL). The combined organic extracts were washed with water (3 x 30 mL), dried over MgSO₄, filtered and the solvent removed *in vacuo* to afford a brown oil. The crude material was purified by flash column chromatography (0 to 20% EtOAc in DCM) to afford **21** (1.14 g, 94%) as a colourless oil; ¹H NMR (400 MHz, CDCl₃) δ ppm 7.54 (s, 1H), 3.70 (s, 3H), 3.27 (t, *J*=7.3 Hz, 2H), 2.82 (t, *J*=7.3 Hz, 2H); LCMS (*m/z*): 250/252 [M+H]⁺.

20

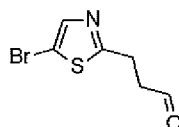
3-(5-Bromothiazol-2-yl)propan-1-ol (22)



To a solution of methyl 3-(5-bromothiazol-2-yl)propanoate **21** (1.14 g, 4.55 mmol) in THF (20 mL) was added lithium borohydride (198 mg, 9.11 mmol) followed by EtOH (1 mL) dropwise. The mixture was stirred vigorously at rt for 1.5h. Water (20 mL) was added carefully followed by EtOAc (20 mL). The mixture was stirred for 15 minutes.
5 NH₄Cl solution (10 mL) was added and the mixture was extracted with EtOAc (40 mL). The organic layer was washed with water (20 mL), dried over MgSO₄ and solvent was removed *in vacuo* to afford **22** (839 mg, 83%) as an oil which was used directly in the next step; ¹H NMR (400 MHz, CDCl₃) δ ppm 7.53 (s, 1H), 3.72 (br s, 2H), 3.14 – 3.01 (m, 2H), 2.07 – 1.89 (m, 2H); LCMS (*m/z*): 222/224 [M+H]⁺.

10

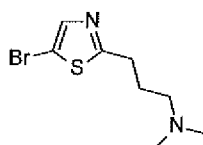
3-(5-Bromothiazol-2-yl)propanal (**23**)



15 To a stirred, cloudy solution of 3-(5-bromothiazol-2-yl)propan-1-ol **22** (839 mg, 3.78 mmol) in DCM (40 mL) was added Dess Martin periodinane (2.40 g, 5.67 mmol) over 20 minutes. The cloudy yellow mixture was stirred for 1h at rt. The reaction was quenched by the addition of water (20 mL) and 10% aq. sodium thiosulfate (20 mL). The product
20 was extracted with DCM (2 x 40 mL) and washed with NaHCO₃ solution (2 x 20 mL) and 10% aq. sodium thiosulfate (20 mL). The solvent was removed *in vacuo* and the crude was absorbed onto silica and purified by flash column chromatography (0-10% EtOAc in DCM) to afford **23** (871 mg, ~quant) as an oil which was used directly in the next step; ¹H NMR (400 MHz, CDCl₃) δ ppm 9.85 (s, 1H), 7.53 (s, 1H), 3.27 (t, *J*=6.9 Hz, 2H), 3.01 (t, *J*=7.0 Hz, 2H); LCMS (*m/z*): 220/222 [M+H]⁺ - product appears as 2 peaks.

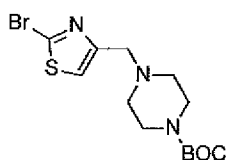
25

3-(5-Bromothiazol-2-yl)-*N,N*-dimethylpropan-1-amine (**24**)



To a solution of 3-(5-bromothiazol-2-yl)propanal **23** (831 mg, 3.78mmol) in DCM (30 mL) was added dimethylamine in EtOH (1.01 mL, 5.67 mmol, 33%). The reaction mixture was stirred at rt for 20 minutes. Sodium triacetoxyborohydride (801 mg, 3.78 mmol) was added and the mixture was stirred at rt overnight. Water (10 mL) and NaHCO₃ solution (20 mL) were added and the mixture was extracted with DCM (3 x 30 mL). The combined extracts were washed with water (2 x 20 mL) and the solvent removed to afford an orange oil (0.54 g). The oil was purified on an SCX-2 cartridge (MeOH, then 20% 7M NH₃ in MeOH) to afford **24** (467 mg, 50%) as a light, brown oil; ¹H NMR (400 MHz, CDCl₃) δ ppm 7.53 (s, 1H), 2.99 (t, *J*=7.5 Hz, 2H), 2.33 (t, *J*=7.2 Hz, 2H), 2.22 (s, 6H), 2.00 – 1.79 (m, 2H); LCMS (*m/z*): 249/251 [M+H]⁺.

tert-Butyl 4-[(2-bromo-1,3-thiazol-4-yl)methyl]piperazine-1-carboxylate (25)

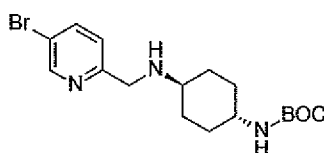


15

To a solution of *N*-BOC piperazine (267 mg, 1.43 mmol) in DCM (4 mL) was added 2-bromothiazole-4-carboxaldehyde (250 mg, 1.30 mmol) in DCM (1 mL) and the mixture stirred at rt for 30 minutes. Then sodium triacetoxyborohydride (414 mg, 1.95 mmol) was added portionwise and stirring continued at rt for 18h. Dilution with MeOH (5 mL), concentration *in vacuo* onto silica and column chromatography (5% to 100% EtOAc in pet ether gradient) gave **25** (291 mg, 62%) as a clear oil; ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 7.53 (s, 1H), 3.58 (s, 2H), 3.36 - 3.25 (m, 4H), 2.38 - 2.33 (m, 4H), 1.38 (s, 9H); LCMS: (*m/z*) = 362/364 [M+H]⁺.

25 The following intermediates were prepared using analogous procedures:

tert-Butyl (trans-4-[[[5-bromopyridin-2-yl)methyl]amino]cyclohexyl)carbamate (26)

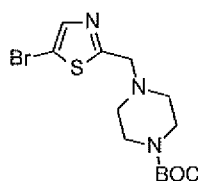


57

¹H NMR (400 MHz, CDCl₃) δ ppm 8.54 (d, *J*=1.8 Hz, 1H), 7.74 – 7.71 (m, 1H), 7.19 (d, *J*=8.2 Hz, 1H), 4.93 (br s, 2H), 4.50 (br d, *J*=5.5 Hz, 1H), 3.87 (s, 2H), 2.51 – 2.43 (m, 1H), 1.99 – 1.94 (m, 4H), 1.38 (s, 9H), 1.30 – 1.20 (m, 2H), 1.11 – 1.01 (m, 2H); LCMS: (*m/z*) = 384/386 [M+H]⁺.

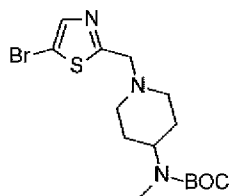
5

***tert*-Butyl 4-[(5-bromo-1,3-thiazol-2-yl)methyl]piperazine-1-carboxylate (27)**



10 ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 7.77 (s, 1H), 3.82 (s, 2H), 3.33 (t, *J*=4.3 Hz, 4H), 2.47 – 2.45 (m, 4H), 1.39 (s, 9H); LCMS: (*m/z*) = 362/364 [M+H]⁺.

***tert*-Butyl {1-[(5-bromo-1,3-thiazol-2-yl)methyl]piperidin-4-yl}methylcarbamate (28)**

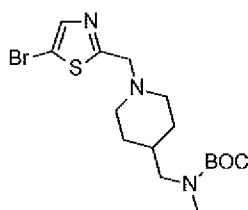


15

¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 7.75 (s, 1H), 4.69 (app d, *J*=5.5 Hz, 1H), 3.78 (s, 2H), 2.96 – 2.92 (m, 2H), 2.68 (s, 3H), 2.23 – 2.16 (m, 2H), 1.72 – 1.64 (m, 2H), 1.55 – 1.48 (m, 2H), 1.39 (s, 9H); LCMS: (*m/z*) = 390/392 [M+H]⁺.

20

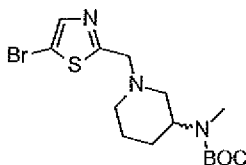
***tert*-Butyl ({1-[(5-bromo-1,3-thiazol-2-yl)methyl]piperidin-4-yl}methyl)methyl carbamate (29)**



58

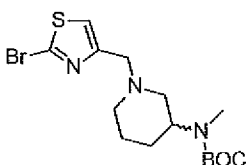
¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 7.74 (s, 1H), 3.75 (s, 2H), 3.05 (d, *J*=6.9 Hz, 2H), 2.87 (d, *J*=11.4 Hz, 2H), 2.75 (br s, 3H), 2.13 – 2.06 (m, 2H), 1.60 – 1.52 (m, 3H), 1.38 (s, 9H), 1.22 – 1.11 (m, 2H); LCMS: (*m/z*) = 404/406 [M+H]⁺.

5 ***tert*-Butyl {1-[(5-bromo-1,3-thiazol-2-yl)methyl]piperidin-3-yl}methylcarbamate (30)**



10 ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 7.76 (s, 1H), 3.80 (br s, 3H), 2.84 – 2.70 (m, 2H), 2.67 (s, 3H), 2.17 – 2.00 (m, 2H), 1.74 – 1.60 (m, 2H), 1.56 – 1.42 (m, 2H), 1.37 (s, 9H); LCMS: (*m/z*) = 390/392 [M+H]⁺.

***tert*-Butyl {1-[(2-bromo-1,3-thiazol-4-yl)methyl]piperidin-3-yl}methylcarbamate (31)**

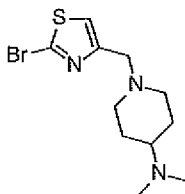


15

¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 7.52 (s, 1H), 3.92 – 3.72 (m, 1H), 3.58 (br s, 2H), 2.78 – 2.67 (m, 2H), 2.86 (s, 2H), 1.99 (br s, 1H), 1.90 – 1.84 (m, 1H), 1.86 – 1.40 (m, 5H), 1.35 (br s, 9H); LCMS: (*m/z*) = 390/392 [M+H]⁺.

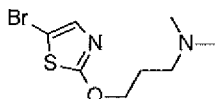
20

1-[(2-Bromo-1,3-thiazol-4-yl)methyl]-*N,N*-dimethylpiperidin-4-amine (32)



¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 7.50 (s, 1H), 3.53 (s, 2H), 2.84 (br d, *J*=11.9 Hz, 2H), 2.15 (s, 6H), 2.05 – 1.93 (m, 3H), 1.71 – 1.68 (m, 2H), 1.40 – 1.30 (m, 2H); LCMS: (*m/z*) = 304/306 [M+H]⁺.

5 **3-[(5-Bromo-1,3-thiazol-2-yl)oxy]-*N,N*-dimethylpropan-1-amine (33)**

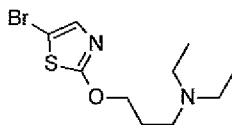


Sodium hydride (60% dispersion, 90 mg, 2.26 mmol) was added portionwise to a
 10 solution of 3-(dimethylamino)propan-1-ol (0.23 mL, 1.98 mmol) in THF (6 mL) at 0°C.
 The reaction mixture was stirred at 0°C for 1h. A solution of 2,5-dibromo-1,3-thiazole
 (500 mg, 2.08 mmol) in THF (2 mL) was added and the reaction mixture stirred at reflux
 for 18h. MeOH was added (10 mL) and then the mixture was concentrated *in vacuo*
 directly onto silica. Purified by Biotage Isolera, eluting with 1-22% MeOH/DCM to give
 15 **33** (0.26 g, 38%) as a yellow gum; ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 7.13 (s, 1H),
 4.45 (s, 2H), 2.54 - 2.47 (m, 2H), 2.30 (s, 6H), 2.13 – 1.97 (m, 2H); LCMS: (*m/z*) =
 265/267 [M+H]⁺.

The following intermediate was prepared using an analogous procedure:

20

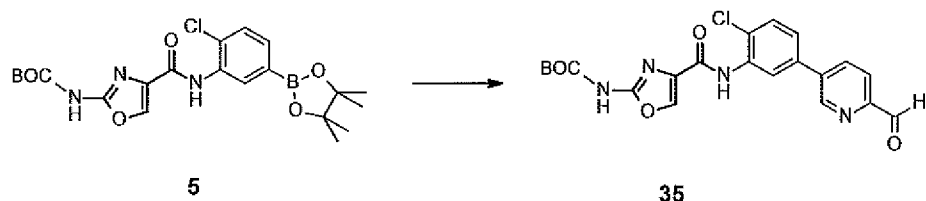
3-[(5-bromo-1,3-thiazol-2-yl)oxy]-*N,N*-diethylpropan-1-amine (34)



25 ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 7.31 (s, 1H), 4.38 (t, *J*=6.4 Hz, 2H), 2.47 – 2.40
 (m, 6H), 1.82 (quintet, *J*=6.8 Hz, 2H), 0.92 (t, *J*=7.1 Hz, 6H); LCMS: (*m/z*) = 293/295
 [M+H]⁺.

Scheme 7 – Preparation of intermediate 35

30



tert-Butyl (4-([2-chloro-5-(6-formylpyridin-3-yl)phenyl]carbamoyl)-1,3-oxazol-2-yl)carbamate (35)

5

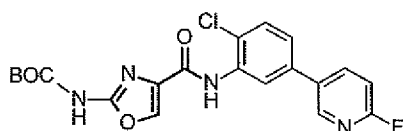
A solution of intermediate **5** (3.75 g, 8.05 mmol) and 5-bromopyridine-2-carboxaldehyde (1 g, 5.38 mmol) in dioxane (15 mL) and water (3 mL) was treated with PdCl₂(dppf) (439 mg, 0.54 mmol) and potassium phosphate (2.85 g, 13.4 mmol) was stirred at 110°C for 3h, cooled and filtered through a plug of silica. EtOAc (500 mL) and water (3 x 100 mL) were added, followed by lithium chloride solution (3 x 30 mL). The organic layer was separated, dried (MgSO₄) and concentrated *in vacuo*. Purification by column chromatography (10 to 20% EtOAc/pet ether gradient) gave the product as a yellow gum. Trituration with Et₂O gave pure **35** (187 mg, 8%) was a pale yellow powder; ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 11.07 (br s, 1H), 10.04 (s, 1H), 9.57 (s, 1H), 9.15 (d, *J*=2.3 Hz, 1H), 8.58 (s, 1H), 8.52 (d, *J*=1.8 Hz, 1H), 8.35 – 8.33 (m, 1H), 8.04 (dd, *J*=7.8, 1.8 Hz, 1H), 7.77 – 7.71 (m, 2H), 1.49 (s, 9H); LCMS: (*m/z*) = 442/444 [M+H]⁺.

15

The following intermediate was prepared using an analogous procedure:

tert-Butyl (4-([2-chloro-5-(6-fluoropyridin-3-yl)phenyl]carbamoyl)-1,3-oxazol-2-yl)carbamate (36)

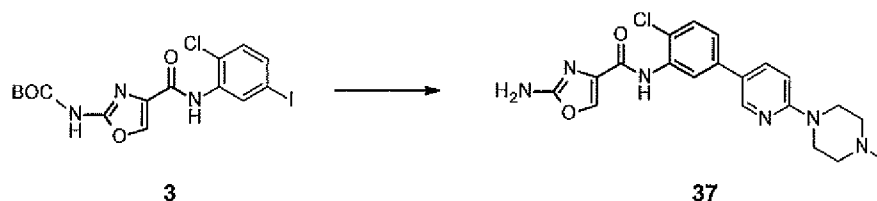
20



25

¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 11.05 (s, 1H), 9.53 (s, 1H), 8.55 (d, *J*=9.2 Hz, 2H), 8.41 (d, *J*=2.2 Hz, 1H), 8.28 (td, *J*=8.2, 2.7 Hz, 1H), 7.69 (d, *J*=8.4 Hz, 1H), 7.58 (dd, *J*=8.4, 2.3 Hz, 1H), 7.32 (dd, *J*=8.5, 2.8 Hz, 1H), 1.48 (s, 9H); LCMS (*m/z*): 433/435 [M+H]⁺.

Scheme 8 – Preparation of example 37



5 **2-Amino-N-(2-chloro-5-[6-(4-methylpiperazin-1-yl)pyridin-3-yl]phenyl)-1,3-oxazole-4-carboxamide (37)**

A microwave reaction vessel was charged with *tert*-butyl 4-(2-chloro-5-iodophenylcarbamoyl) oxazol-2-ylcarbamate **3** (111 mg, 0.24 mmol), Pd(Ph₃P)₄ (28 mg, 0.024 mmol), 1-methyl-4-[5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)pyridin-2-yl]piperazine (109 mg, 0.36 mmol) and potassium phosphate (76 mg, 0.36 mmol). Then DMF (1.6 mL) and water (0.4 mL) were added, the reaction mixture degassed with N₂ for 2 mins, the vessel capped and the reaction stirred at 120°C for 2h. The solvent was removed *in vacuo* and the crude reaction mixture passed through a 1g Isolute-NH₂ cartridge (eluting with MeOH-DCM, 2:1). Concentration of eluent gave the crude product, which was dissolved in DCM (0.7 mL), treated with TFA (0.3 mL) and stirred at rt for 18h. Concentration *in vacuo* and purification by preparative LC-MS gave the product **37** (9 mg, 9%) as a white solid; ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.29 (s, 1H), 8.52 (d, *J*=2.3 Hz, 1H), 8.43 (d, *J*=2.6 Hz, 1H), 8.11 (s, 1H), 7.83 (dd, *J*=8.9, 2.6 Hz, 1H), 7.59 (d, *J*=8.7 Hz, 1H), 7.43 (dd, *J*=8.7, 2.3 Hz, 1H), 7.17 (s, 2H), 6.97 (d, *J*=8.9 Hz, 1H), 3.73 - 3.46 (m, 4H), 3.33 (s, 3H), 2.64 - 2.53 (m, 2H), 2.44 - 2.22 (m, 2H); LCMS: (*m/z*) = 413/415 [M+H]⁺.

10

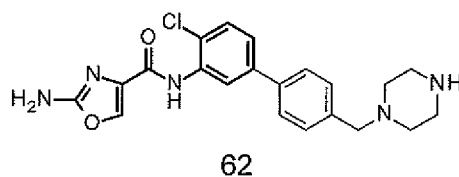
15

20

The following examples were prepared using an analogous procedure:

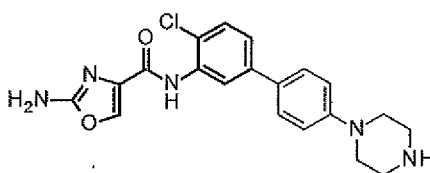
25

2-Amino-N-[4-chloro-4'-(piperazin-1-ylmethyl)biphenyl-3-yl]-1,3-oxazole-4-carboxamide (38)



¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.31 (s, 1H), 8.60 (d, *J*=2.3 Hz, 1H), 8.11 (s, 1H), 7.64 – 7.61 (m, 3H), 7.46 (dd, *J*=8.7, 2.3 Hz, 1H), 7.43 (d, *J*=8.2 Hz, 2H), 7.18 (s, 2H), 3.56 (s, 2H), 2.98 (br t, *J*=4.8 Hz, 4H), 4H not observed – under DMSO signal; LCMS: (m/z) = 412/414 [M+H]⁺.

2-Amino-N-[4-chloro-4'-(piperazin-1-yl)biphenyl-3-yl]-1,3-oxazole-4-carboxamide (39)

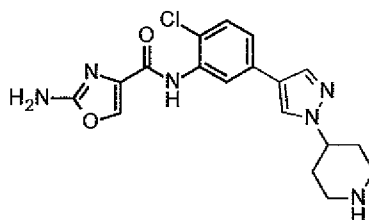


10

¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.27 (s, 1H), 8.55 (d, *J*=2.3 Hz, 1H), 8.10 (s, 1H), 7.56 (d, *J*=8.7 Hz, 1H), 7.50 (d, *J*=8.7 Hz, 2H), 7.40 (dd, *J*=8.5, 2.1 Hz, 1H), 7.17 (s, 2H), 7.02 (d, *J*=8.7 Hz, 2H), 3.12 – 3.09 (m, 4H), 2.86 – 2.82 (m, 4H); LCMS: (m/z) = 398/400 [M+H]⁺.

15

2-Amino-N-(2-chloro-5-[1-piperidin-4-yl]-1H-pyrazol-4-yl)phenyl)-1,3-oxazole-4-carboxamide (40)

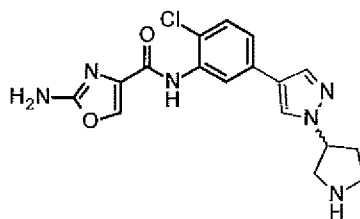


20

¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.24 (br s, 1H), 8.42 (d, *J*=2.3 Hz, 1H), 8.24 (br s, 1H), 8.10 (s, 1H), 7.84 (br s, 1H), 7.51 (d, *J*=8.2 Hz, 1H), 7.40 dd, *J*=8.1, 2.3 Hz, 1H), 7.16 br s, 2H), 4.24 – 4.18 (m, 1H), 3.05 (br d, *J*=12.4 Hz, 2H), 2.62 – 2.57 (m, 2H), 2.01 – 1.95 (m, 2H), 1.86 – 1.76 (m, 2H); LCMS: (m/z) = 387/389 [M+H]⁺.

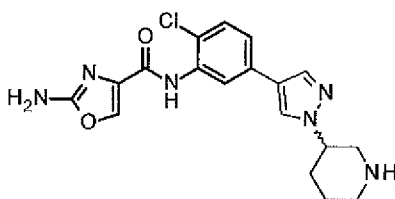
25

2-Amino-N-(2-chloro-5-[1-(pyrrolidin-3-yl)-1H-pyrazol-4-yl]phenyl)-1,3-oxazole-4-carboxamide (41)



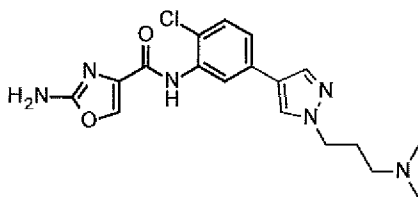
¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.24 (s, 1H), 8.42 (d, *J*=2.3 Hz, 1H), 8.26 (s, 1H), 8.10 (s, 1H), 7.84 (s, 1H), 7.52 (d, *J*=8.5 Hz, 1H), 7.40 (dd, *J*=8.5, 2.1 Hz, 1H), 7.16 (s, 2H), 4.85 – 4.79 (m, 1H), 3.13 (dd, *J*=11.7, 6.6 Hz, 1H), 3.07 – 3.00 (m, 1H), 2.96 (dd, *J*=11.7, 4.3 Hz, 1H), 2.87 – 2.81 (m, 1H), 2.23 – 2.14 (m, 1H), 2.07 – 1.99 (m, 1H); LCMS: (*m/z*) = 373/375 [M+H]⁺.

10 **2-Amino-*N*-(2-chloro-5-[1-(piperidin-3-yl)-1H-pyrazol-4-yl]phenyl)-1,3-oxazole-4-carboxamide (42)**



15 ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.24 (s, 1H), 8.42 (d, *J*=2.3 Hz, 1H), 8.27 (s, 1H), 8.10 (s, 1H), 7.83 (s, 1H), 7.51 (d, *J*=8.2 Hz, 1H), 7.39 (dd, *J*=8.5, 2.1 Hz, 1H), 7.16 (s, 2H), 4.19 – 4.10 (m, 1H), 3.18 – 3.14 (m, 1H), 2.88 – 2.84 (m, 1H), 2.76 (dd, *J*=11.7, 10.3 Hz, 1H), 2.46 – 2.42 (m, 1H), 2.14 – 2.09 (m, 1H), 1.95 – 1.85 (m, 1H), 1.73 – 1.68 (m, 1H), 1.55 – 1.45 (m, 1H); LCMS: (*m/z*) = 387/389 [M+H]⁺.

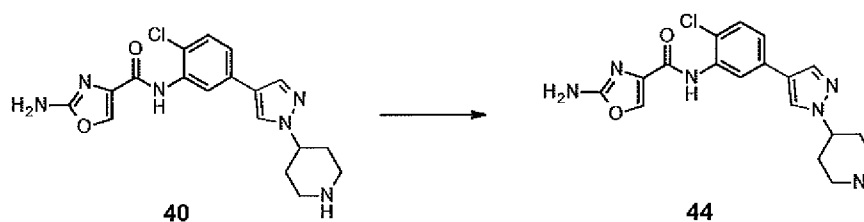
20 **2-Amino-*N*-(2-chloro-5-[1-[3-(dimethylamino)propyl]-1H-pyrazol-4-yl]phenyl)-1,3-oxazole-4-carboxamide (43)**



¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.24 (s, 1H), 8.43 (d, *J*=1.8 Hz, 1H), 8.20 (s, 1H), 8.10 (s, 1H), 7.85 (s, 1H), 7.52 (d, *J*=8.2 Hz, 1H), 7.39 (dd, *J*=8.5, 2.1 Hz, 1H), 7.17 (s, 2H), 4.14 (t, *J*=7.1 Hz, 2H), 2.22 (t, *J*=6.4 Hz, 2H), 2.16 (s, 6H), 1.94 (quintet, *J*=7.0 Hz, 2H); LCMS: (*m/z*) = 389/391 [M+H]⁺.

5

Scheme 9 – Preparation of example 44



10 **2-Amino-N-(2-chloro-5-[1-(1-methylpiperidin-4-yl)-1H-pyrazol-4-yl]phenyl)-1,3-oxazole-4-carboxamide (44)**

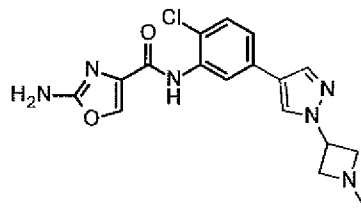
A solution of 2-amino-N-(2-chloro-5-[1-piperidin-4-yl]-1H-pyrazol-4-yl]phenyl)-1,3-oxazole-4-carboxamide **40** (50 mg, 0.13 mmol) in THF (0.8 mL) was treated with formaldehyde (37% in water, 0.014 mL, 0.16 mmol) and AcOH (0.017 mL, 0.29 mmol) and stirred for 10 minutes. Then sodium triacetoxyborohydride (62 mg, 0.29 mmol) was added and the mixture stirred at rt for 18h. Dilution with DMSO (0.1 mL) and MeOH (0.2 mL) and purification by preparative LC-MS gave **44** (4 mg, 8%) as a white solid; ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.24 (s, 1H), 8.42 (d, *J*=2.3 Hz, 1H), 8.27 (s, 1H), 8.10 (s, 1H), 7.84 (d, *J*=0.9 Hz, 1H), 7.51 (d, *J*=8.5 Hz, 1H), 7.39 (dd, *J*=8.5, 2.3 Hz, 1H), 7.16 (s, 2H), 4.17 - 4.08 (m, 1H), 2.88 - 2.83 (m, 2H), 2.20 (s, 3H), 2.07 - 1.94 (m, 6H); LCMS: (*m/z*) = 401/403 [M+H]⁺.

20

The following example was prepared using an analogous procedure:

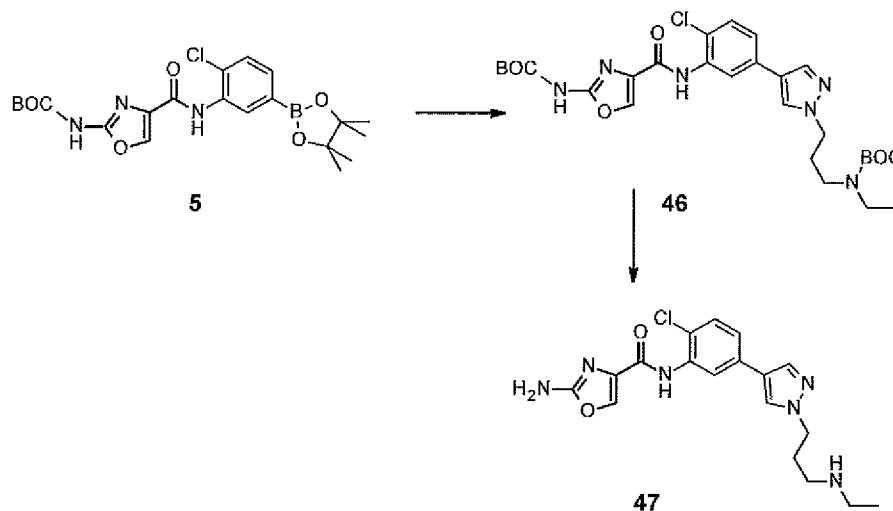
25

2-Amino-N-(2-chloro-5-[1-(1-methylazetidin-3-yl)-1H-pyrazol-4-yl]phenyl)-1,3-oxazole-4-carboxamide (45)

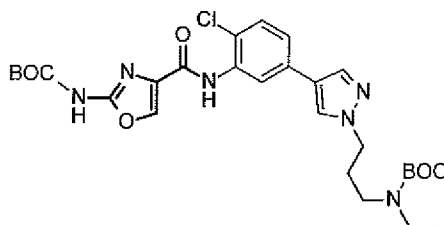


¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.25 (s, 1H), 8.44 (d, *J*=2.3 Hz, 1H), 8.36 (s, 1H), 8.10 (s, 1H), 7.92 (s, 1H), 7.53 (d, *J*=8.3 Hz, 1H), 7.42 (dd, *J*=8.5, 2.1 Hz, 1H), 7.16 (s, 2H), 4.96 (quintet, *J*=6.9 Hz, 1H), 3.71 – 3.68 (m, 2H), 3.40 – 3.37 (m, 2H), 2.33 (s, 3H); LCMS: (*m/z*) = 373/375 [M+H]⁺.

Scheme 10 – Preparation of example 47



10 **tert-Butyl (3-(4-(3-(2-((tert-butoxycarbonyl)amino)oxazole-4-carboxamido)-4-chlorophenyl)-1H-pyrazol-1-yl)propyl)(ethyl)carbamate (46)**

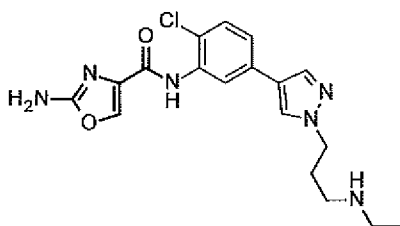


15 A mixture of *tert*-butyl (3-(4-bromo-1H-pyrazol-1-yl)propyl)(ethyl)carbamate **17** (196 mg, 0.59 mmol), *tert*-butyl (4-((2-chloro-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)carbamoyl)oxazol-2-yl)carbamate **5** (300 mg, 0.65 mmol) and potassium

carbonate (163 mg, 118 mmol) in dioxane (5 mL) and water (1 mL) was degassed with nitrogen (sparge, 5 min). PdCl₂(dppf) (52 mg, 0.006 mmol) was added and the reaction mixture was heated to 110°C for 1h. The mixture was diluted with EtOAc (25 mL), washed with NH₄Cl solution (2 x 15 mL) and concentrated *in vacuo*. The crude material
5 was purified by column chromatography (70% EtOAc in heptanes) to afford **46** (128 mg, 33%) as a colourless solid; ¹H NMR (400 MHz, CDCl₃) δ ppm 8.23 (d, *J*=2.1 Hz, 1H), 8.21 (s, 1H), 8.07 (s, 1H), 7.86 (s, 1H), 7.47 (d, *J*=8.4 Hz, 1H), 7.41 (dd, *J*=8.4, 2.1 Hz, 1H), 4.19 (t, *J*=6.8 Hz, 2H), 3.27 – 3.16 (m, 4H), 2.11 (quintet, *J*=7.0 Hz, 2H), 1.56 (s, 9H), 1.42 (s, 9H), 1.09 (t, *J*=7.1 Hz, 3H); LCMS (*m/z*): 589/591 [M+H]⁺.

10

2-Amino-N-(2-chloro-5-(1-(3-(ethylamino)propyl)-1H-pyrazol-4-yl)phenyl)oxazole-4-carboxamide (47)

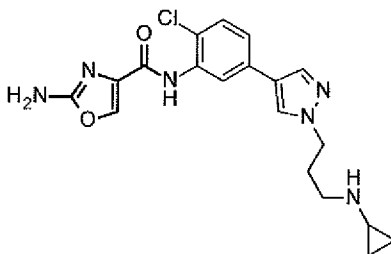


15

To a solution of *tert*-butyl (3-(4-(3-(2-((*tert*-butoxycarbonyl)amino)oxazole-4-carboxamido)-4-chlorophenyl)-1H-pyrazol-1-yl)propyl)(ethyl)carbamate **46** (128 mg, 0.212 mmol) in DCM (5 mL) was added TFA (0.5 mL). The reaction was stirred at rt for 1h and then concentrated *in vacuo*. The residue was treated with 7N NH₃ in MeOH
20 solution and evaporated again. The crude was purified by column chromatography (DCM/MeOH 9:1, 1% NH₃ solution) and then further purified with SCX-2 (DCM (10 mL), MeOH (10 mL), released with 7N NH₃ in MeOH) to give **47** (45 mg, 57%) as a colourless solid; ¹H NMR (400 MHz, CD₃OD) δ ppm 8.50 (d, *J*=2.1 Hz, 1H), 8.05 (d, *J*=0.9 Hz, 1H), 7.89 (s, 1H), 7.84 (s, 1H), 7.44 (d, *J*=8.4 Hz, 1H), 7.34 (dd, *J*=8.3, 2.1 Hz, 1H), 4.24 (t, *J*=6.8 Hz, 2H), 2.61 (quintet, *J*=7.5 Hz, 4H), 2.07 (quintet, *J*=7.0 Hz,
25 2H), 1.10 (t, *J*=7.2 Hz, 3H); LCMS (*m/z*): 389/391 [M+H]⁺.

The following examples were prepared using analogous procedures:

2-Amino-N-(2-chloro-5-(1-(3-(cyclopropylamino)propyl)-1H-pyrazol-4-yl)phenyl)oxazole-4-carboxamide (48)

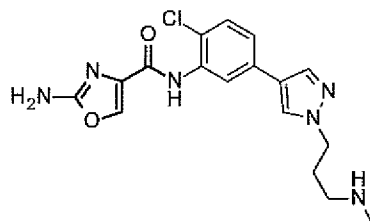


5

^1H NMR (400 MHz, CD_3OD) δ ppm 8.52 (d, $J=2.0$ Hz, 1H), 8.06 (s, 1H), 7.91 (s, 1H), 7.87 (d, $J=0.8$ Hz, 1H), 7.46 (d, $J=8.4$ Hz, 1H), 7.36 (dd, $J=8.4, 2.1$ Hz, 1H), 4.27 (t, $J=6.8$ Hz, 2H), 2.74 (t, $J=7.3$ Hz, 2H), 2.22 (tt, $J=7.1, 3.7$ Hz, 1H), 2.12 (quintet, $J=7.0$ Hz, 2H), 0.54 (td, $J=6.8, 4.7$ Hz, 2H), 0.42 (dt, $J=6.4, 4.4$ Hz, 2H); LCMS (m/z): 401/403

10 $[\text{M}+\text{H}]^+$.

2-Amino-N-(2-chloro-5-(1-(3-(methylamino)propyl)-1H-pyrazol-4-yl)phenyl)oxazole-4-carboxamide (49)

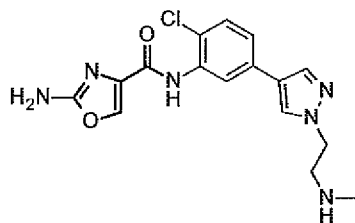


15

^1H NMR (400 MHz, CD_3OD) δ ppm 8.52 (d, $J=2.1$ Hz, 1H), 8.07 (d, $J=0.8$ Hz, 1H), 7.92 (s, 1H), 7.87 (d, $J=0.8$ Hz, 1H), 7.46 (d, $J=8.4$ Hz, 1H), 7.36 (dd, $J=8.4, 2.1$ Hz, 1H), 4.26 (t, $J=6.8$ Hz, 2H), 2.59 (dd, $J=7.8, 6.7$ Hz, 2H), 2.39 (s, 3H), 2.14 – 2.03 (m, 2H);

20 LCMS (m/z): 375/377 $[\text{M}+\text{H}]^+$.

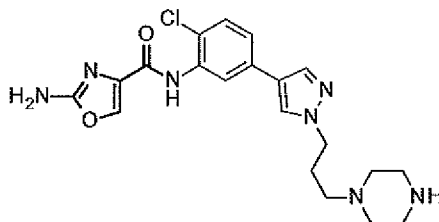
2-Amino-N-(2-chloro-5-(1-(2-(methylamino)ethyl)-1H-pyrazol-4-yl)phenyl)oxazole-4-carboxamide (50)



¹H NMR (400 MHz, CD₃OD) δ ppm 8.53 (d, *J*=2.1 Hz, 1H), 8.06 (d, *J*=0.8 Hz, 1H), 7.91 (s, 1H), 7.88 (d, *J*=0.8 Hz, 1H), 7.46 (d, *J*=8.3 Hz, 1H), 7.37 (dd, *J*=8.4, 2.1 Hz, 1H), 4.32 (t, *J*=6.2 Hz, 2H), 3.05 (t, *J*=6.2 Hz, 2H), 2.42 (s, 3H); LCMS (*m/z*): 361/363 [M+H]⁺.

2-Amino-N-(2-chloro-5-(1-(3-(piperazin-1-yl)propyl)-1H-pyrazol-4-yl)phenyl)oxazole-4-carboxamide (51)

10

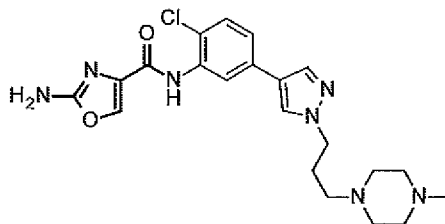


¹H NMR (400 MHz, CD₃OD) δ ppm 8.53 (d, *J*=2.1 Hz, 1H), 8.07 (s, 1H), 7.92 (s, 1H), 7.86 (d, *J*=0.8 Hz, 1H), 7.47 (d, *J*=8.3 Hz, 1H), 7.37 (dd, *J*=8.3, 2.1 Hz, 1H), 4.26 (t, *J*=6.7 Hz, 2H), 2.92 – 2.81 (m, 4H), 2.46 (s, 4H), 2.36 (t, *J*=7.3 Hz, 2H), 2.10 (quintet, *J*=6.9 Hz, 2H); LCMS (*m/z*): 430/432 [M+H]⁺.

15

2-Amino-N-(2-chloro-5-(1-(3-(4-methylpiperazin-1-yl)propyl)-1H-pyrazol-4-yl)phenyl)oxazole-4-carboxamide (52)

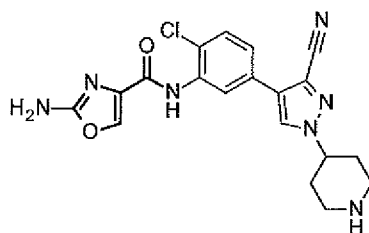
20



¹H NMR (400 MHz, CDCl₃) δ ppm 9.20 (s, 1H), 8.72 (d, *J*=2.1 Hz, 1H), 7.81 (s, 1H), 7.78 (s, 1H), 7.70 (s, 1H), 7.36 (d, *J*=8.3 Hz, 1H), 7.17 (dd, *J*=8.3, 2.1 Hz, 1H), 4.74 (s, 2H), 4.20 (t, *J*=6.9 Hz, 2H), 2.66 – 2.39 (m, 8H), 2.35 (t, *J*=7.0 Hz, 2H), 2.29 (s, 3H), 2.06 (quintet, *J*=7.0 Hz, 2H); LCMS (*m/z*): 444/446 [M+H]⁺.

5

2-Amino-*N*-(2-chloro-5-(3-cyano-1-(piperidin-4-yl)-1H-pyrazol-4-yl)phenyl)oxazole-4-carboxamide (53)

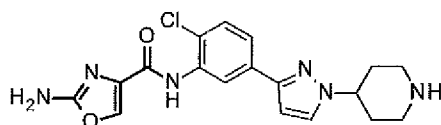


10

¹H NMR (400 MHz, CD₃OD) δ ppm 8.68 (d, *J*=2.2 Hz, 1H), 8.27 (s, 1H), 7.92 (s, 1H), 7.59 (d, *J*=8.4 Hz, 1H), 7.50 (dd, *J*=8.4, 2.2 Hz, 1H), 4.58 – 4.47 (m, 1H), 3.36 – 3.24 (m, 2H), 2.95 – 2.83 (m, 2H), 2.23 (d, *J*=11.7 Hz, 2H), 2.15 – 2.05 (m, 2H); LCMS (*m/z*): 412/414 [M+H]⁺.

15

2-amino-*N*-(2-chloro-5-[1-(piperidin-4-yl)-1H-pyrazol-3-yl]phenyl)-1,3-oxazole-4-carboxamide (54)

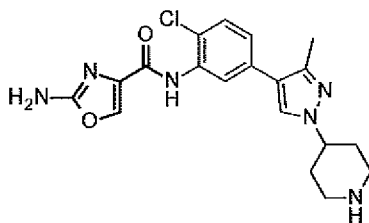


20

¹H NMR (400 MHz, CD₃OD) δ ppm 8.75 (d, *J*=2.0 Hz, 1H), 7.91 (s, 1H), 7.72 (d, *J*=2.4 Hz, 1H), 7.61 (dd, *J*=8.4, 2.1 Hz, 1H), 7.50 (d, *J*=8.5 Hz, 1H), 6.66 (d, *J*=2.4 Hz, 1H), 4.35 (tt, *J*=11.6, 4.1 Hz, 1H), 3.22 (dt, *J*=13.3, 3.3 Hz, 2H), 2.79 (td, *J*=12.7, 2.7 Hz, 2H), 2.20 – 2.12 (m, 2H), 2.01 (qd, *J*=12.4, 4.1 Hz, 2H); LCMS (*m/z*): 389/389 [M+H]⁺.

25

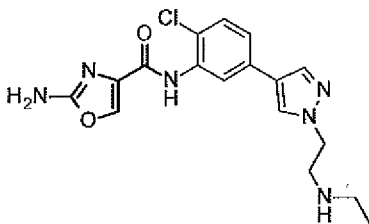
2-Amino-*N*-(2-chloro-5-(3-methyl-1-(piperidin-4-yl)-1H-pyrazol-4-yl)phenyl)oxazole-4-carboxamide (55)



¹H NMR (400 MHz, CD₃OD) δ ppm 8.48 (d, *J*=2.1 Hz, 1H), 7.90 (s, 1H), 7.89 (s, 1H), 7.49 (d, *J*=8.4 Hz, 1H), 7.25 (dd, *J*=8.4, 2.1 Hz, 1H), 4.25 (tt, *J*=11.8, 4.1 Hz, 1H), 3.25 – 3.17 (m, 2H), 2.78 (td, *J*=12.8, 2.6 Hz, 2H), 2.42 (s, 3H), 2.17 – 2.09 (m, 2H), 1.95 (qd, *J*=12.3, 4.2 Hz, 2H); LCMS (*m/z*): 401/403 [M+H]⁺.

2-Amino-N-(2-chloro-5-(1-(2-(ethylamino)ethyl)-1H-pyrazol-4-yl)phenyl)oxazole-4-carboxamide (56)

10

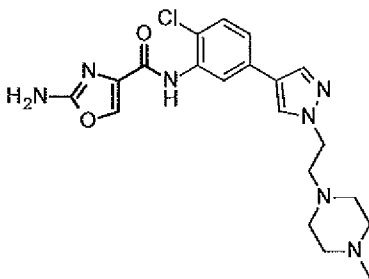


¹H NMR (400 MHz, CD₃OD) δ ppm 8.53 (d, *J*=2.1 Hz, 1H), 8.06 (d, *J*=0.8 Hz, 1H), 7.91 (s, 1H), 7.88 (d, *J*=0.8 Hz, 1H), 7.46 (d, *J*=8.4 Hz, 1H), 7.36 (dd, *J*=8.3, 2.1 Hz, 1H), 4.32 (t, *J*=6.3 Hz, 2H), 3.08 (t, *J*=6.3 Hz, 2H), 2.67 (q, *J*=7.2 Hz, 2H), 1.13 (t, *J*=7.2 Hz, 3H); LCMS (*m/z*): 375/377 [M+H]⁺.

15

2-Amino-N-(2-chloro-5-(1-(2-(4-methylpiperazin-1-yl)ethyl)-1H-pyrazol-4-yl)phenyl)oxazole-4-carboxamide (57)

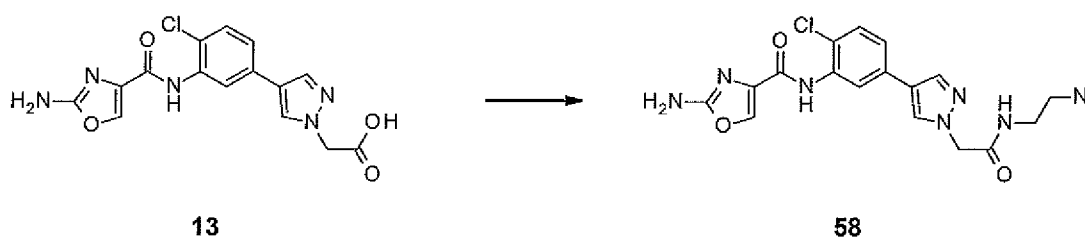
20



¹H NMR (400 MHz, CD₃OD) δ ppm 8.51 (d, *J*=2.1 Hz, 1H), 8.06 (d, *J*=0.8 Hz, 1H), 7.90 (s, 1H), 7.84 (d, *J*=0.8 Hz, 1H), 7.45 (d, *J*=8.4 Hz, 1H), 7.34 (dd, *J*=8.3, 2.1 Hz, 1H), 4.30 (t, *J*=6.5 Hz, 2H), 2.85 (t, *J*=6.5 Hz, 2H), 2.66 – 2.38 (m, 8H), 2.27 (s, 3H); LCMS (*m/z*): 430/432 [M+H]⁺.

5

Scheme 11 – Preparation of example 58

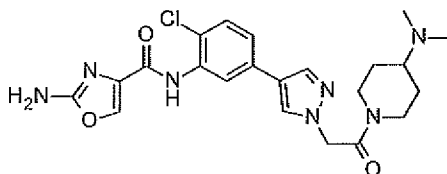


10 **2-Amino-N-[2-chloro-5-[1-(2-{[2-(dimethylamino)ethyl]amino}-2-oxoethyl)-1H-pyrazol-4-yl]phenyl]-1,3-oxazole-4-carboxamide (58)**

A solution of **13** (TFA salt, 100 mg, 0.21 mmol) in DMF (1 mL) at rt was treated with HATU (120 mg, 0.32 mmol), *N,N*-dimethylethylenediamine (0.03 mL, 0.25 mmol) and
 15 DIPEA (0.22 mL, 1.26 mmol) and stirred at rt for 18h. It was then concentrated *in vacuo* and the residue passed through a 1g NH₂ cartridge (eluting with DCM-MeOH, 1:1). The eluent was concentrated *in vacuo* and the residue purified by preparative LC-MS to give
58 (8 mg, 9%) as a white solid; ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.25 (s, 1H), 8.46 (d, *J*=1.8 Hz, 1H), 8.16 (s, 1H), 8.10 (s, 1H), 8.08 (t, *J*=5.5 Hz, 1H), 7.87 (d, *J*=0.9 Hz,
 20 1H), 7.53 (d, *J*=8.2 Hz, 1H), 7.40 (dd, *J*=8.5, 2.0 Hz, 1H), 7.17 (s, 2H), 4.82 (s, 2H), 3.21 – 3.16 (m, 2H), 2.29 (t, *J*=6.6 Hz, 2H), 2.14 (s, 6H); LCMS: (*m/z*) = 432/434 [M+H]⁺.

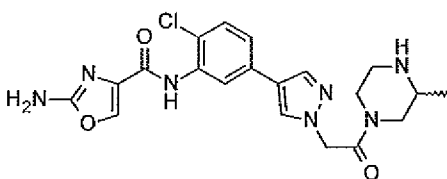
The following examples were prepared using an analogous procedure:

25 **2-Amino-N-[2-chloro-5-(1-{2-[4-(dimethylamino)piperidin-1-yl]-2-oxoethyl}-1H-pyrazol-4-yl)phenyl]-1,3-oxazole-4-carboxamide (59)**



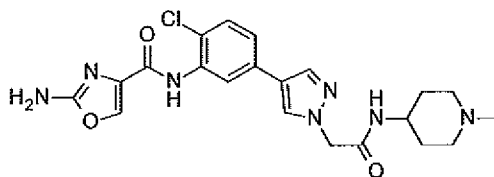
¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.25 (s, 1H), 8.46 (d, *J*=2.3 Hz, 1H), 8.11 (s, 1H), 8.10 (s, 1H), 7.85 (d, *J*=0.9 Hz, 1H), 7.52 (d, *J*=8.7 Hz, 1H), 7.40 (dd, *J*=8.5, 2.1 Hz, 1H), 7.17 (s, 2H), 5.21 – 5.11 (m, 2H), 4.30 – 4.27 (m, 1H), 3.93 – 3.90 (m, 1H), 3.08 – 3.02 (m, 1H), 2.68 – 2.65 (m, 1H), 2.36 – 2.30 (m, 1H), 2.17 (s, 6H), 1.79 – 1.73 (m, 2H), 1.42 – 1.32 (m, 1H), 1.27 – 1.18 (m, 1H); LCMS: (*m/z*) = 472/474 [M+H]⁺.

10 **2-Amino-N-(2-chloro-5-{1-[2-(3-methylpiperazin-1-yl)-2-oxoethyl]-1H-pyrazol-4-yl}phenyl)-1,3-oxazole-4-carboxamide (60)**



¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.25 (s, 1H), 8.46 (d, *J*=1.8 Hz, 1H), 8.11 (s, 1H), 8.10 (s, 1H), 7.52 (d, *J*=8.2 Hz, 1H), 7.39 (dd, *J*=8.5, 2.1 Hz, 1H), 7.17 (s, 2H), 5.24 – 5.17 (m, 1H), 5.11 – 5.05 (m, 1H), 4.13 (d, *J*=11.4 Hz, 1H), 3.76 (d, *J*=10.8 Hz, 1H), 3.03 – 2.96 (m, 1H), 2.67 – 2.50 (m, 4H), 3.24 (br s, 1H), 0.99 – 0.95 (m, 3H); LCMS: (*m/z*) = 444/446 [M+H]⁺.

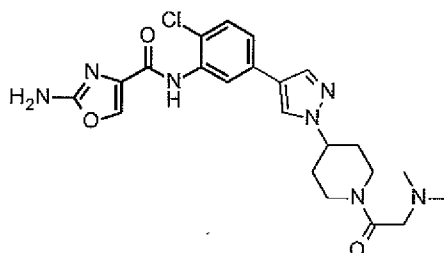
20 **2-Amino-N-[2-chloro-5-{1-{2-[(1-methylpiperidin-4-yl)amino]-2-oxoethyl]-1H-pyrazol-4-yl}phenyl]-1,3-oxazole-4-carboxamide (61)**



¹H NMR (400 MHz, CDCl₃) δ ppm 9.23 (s, 1H), 8.76 (d, *J*=1.8 Hz, 1H), 7.92 (s, 1H), 7.82 (s, 1H), 7.77 (s, 1H), 7.41 (d, *J*=8.2 Hz, 1H), 7.21 – 7.18 (m, 1H), 6.36 (br s, 1H), 4.81 (s, 2H), 4.72 (br s, 2H), 3.84 (br s, 1H), 2.83 (br s, 2H), 2.35 (s, 3H), 2.24 (br s, 2H), 1.96 – 1.93 (m, 2H); LCMS: (*m/z*) = 458/460 [M+H]⁺.

5

2-Amino-*N*-(2-chloro-5-{1-[1-(*N,N*-dimethylglycyl)piperidin-4-yl]-1H-pyrazol-4-yl}phenyl)-1,3-oxazole-4-carboxamide (62)

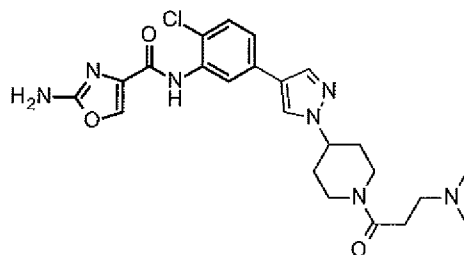


10

¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.24 (s, 1H), 8.42 (d, *J*=1.8 Hz, 1H), 8.29 (s, 1H), 8.10 (s, 1H), 7.86 (d, *J*=0.9 Hz, 1H), 7.52 (d, *J*=8.2 Hz, 1H), 7.40 (dd, *J*=8.2, 2.3 Hz, 1H), 7.16 (s, 2H), 4.49 – 4.42 (m, 2H), 4.19 – 4.15 (m, 1H), 3.19 – 3.12 (m, 2H), 3.05 – 3.02 (m, 1H), 2.78 – 2.71 (m, 1H), 2.19 (s, 6H), 2.09 – 2.04 (m, 2H), 1.94 – 1.72 (m, 2H); LCMS: (*m/z*) = 472/474 [M+H]⁺.

15

2-Amino-*N*-(2-chloro-5-{1-[1-(*N,N*-dimethyl-beta-alanyl)piperidin-4-yl]-1H-pyrazol-4-yl}phenyl)-1,3-oxazole-4-carboxamide (63)

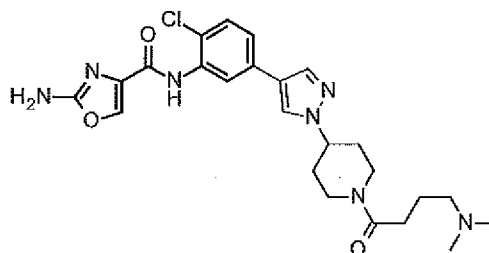


20

¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.22 (br s, 1H), 8.42 (d, *J*=1.8 Hz, 1H), 8.29 (s, 1H), 8.10 (s, 1H), 7.86 (s, 1H), 7.52 (d, *J*=8.2 Hz, 1H), 7.40 (dd, *J*=8.5, 2.1 Hz, 1H), 7.16 (s, 2H), 4.49 – 4.41 (m, 2H), 4.01 (br d, *J*=13.7 Hz, 1H), 3.22 – 3.15 (m, 1H), 2.75 –

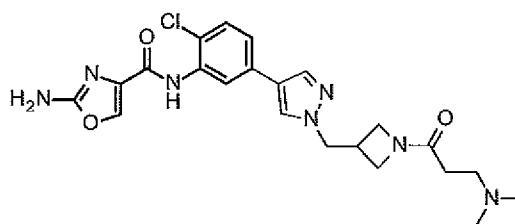
2.68 (m, 1H), 2.51 – 2.46 (m, 4H), 2.14 (s, 6H), 2.10 – 2.03 (m, 2H), 1.95 – 1.71 (m, 2H); LCMS: (m/z) = 486/488 [M+H]⁺.

5 **2-Amino-N-[2-chloro-5-(1-{1-[4-(dimethylamino)butanoyl]piperidin-4-yl}-1H-pyrazol-4-yl)phenyl]-1,3-oxazole-4-carboxamide (64)**



10 ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.24 (s, 1H), 8.42 (d, *J*=2.3 Hz, 1H), 8.29 (s, 1H), 8.10 (s, 1H), 7.86 (s, 1H), 7.52 (d, *J*=8.2 Hz, 1H), 7.39 (dd, *J*=8.5, 2.1 Hz, 1H), 7.16 (s, 2H), 4.50 – 4.41 (m, 2H), 3.98 (br d, *J*=14.2 Hz, 1H), 3.18 (br t, *J*=13.3 Hz, 1H), 2.76 – 2.69 (m, 1H), 2.35 (t, *J*=7.6 Hz, 2H), 2.20 (t, *J*=7.1 Hz, 2H), 2.11 (s, 6H), 2.09 – 2.02 (m, 2H), 1.94 – 1.71 (m, 2H), 1.63 (quintet, *J*=7.3 Hz, 2H); LCMS: (m/z) = 500/502 [M+H]⁺.

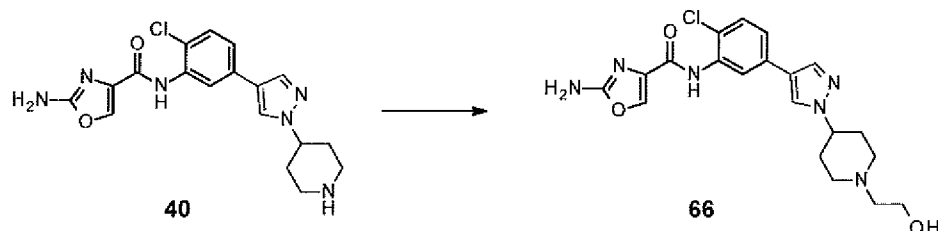
15 **2-Amino-N-[2-chloro-5-(1-{1-(*N,N*-dimethyl-beta-alanyl)azetididin-3-yl}methyl)-1H-pyrazol-4-yl)phenyl]-1,3-oxazole-4-carboxamide (65)**



20 ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.24 (br s, 1H), 8.44 (d, *J*=2.3 Hz, 1H), 8.26 (s, 1H), 8.10 (s, 1H), 7.87 (d, *J*=0.9 Hz, 1H), 7.52 (d, *J*=8.2 Hz, 1H), 7.39 (dd, *J*=8.5, 2.1 Hz, 1H), 7.15 (s, 2H), 4.37 (d, *J*=7.3 Hz, 2H), 4.19 (t, *J*=8.5 Hz, 1H), 3.97 (dd, *J*=8.7, 5.5 Hz, 1H), 3.88 (t, *J*=8.9 Hz, 1H), 3.67 (dd, *J*=9.6, 5.5 Hz, 1H), 3.10 – 3.01 (m, 1H), 2.42 – 2.38 (m, 2H), 2.15 – 2.11 (m, 2H), 2.09 (s, 6H); LCMS: (m/z) = 472/474 [M+H]⁺.

25

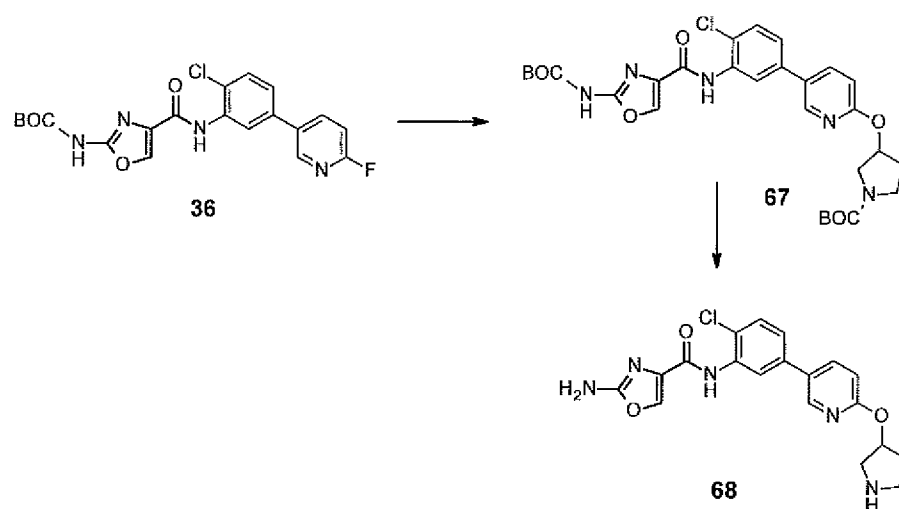
Scheme 12 – Preparation of example 66



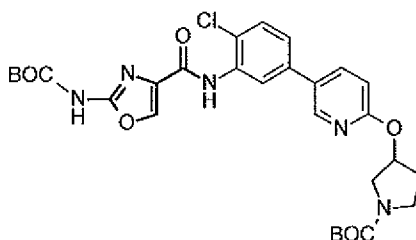
5 **2-Amino-N-(2-chloro-5-{1-[1-(2-hydroxyethyl)piperidin-4-yl]-1H-pyrazol-4-yl}phenyl)-1,3-oxazole-4-carboxamide (66)**

A suspension of **40** (100 mg, 0.26 mmol) in DMF (1 mL) containing potassium carbonate (143 mg, 1.03 mmol) was treated dropwise with 2-bromoethanol (0.03 mL, 0.39 mmol) and stirred at rt for 18h. Concentration *in vacuo* and direct purification by preparative LC-MS gave **66** (8 mg, 7%) as a white solid; ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.24 (s, 1H), 8.42 (d, *J*=1.8 Hz, 1H), 8.26 (s, 1H), 8.10 (s, 1H), 7.84 (s, 1H), 7.51 (d, *J*=8.7 Hz, 1H), 7.39 (dd, *J*=8.4, 2.3 Hz, 1H), 7.16 (s, 2H), 4.42 (t, *J*=5.3 Hz, 1H), 4.14 (tt, *J*=10.5, 5.1 Hz, 1H), 3.53 – 3.49 (m, 2H), 2.97 (br d, *J*=11.9 Hz, 2H), 2.42 (t, *J*=6.4 Hz, 2H), 2.13 (td, *J*=11.3, 3.0 Hz, 2H), 2.02 – 1.91 (m, 4H); LCMS: (*m/z*) = 431/433 [M+H]⁺.

Scheme 13 – Preparation of example 68



***tert*-Butyl 3-((5-(3-(2-((*tert*-butoxycarbonyl)amino)oxazole-4-carboxamido)-4-chlorophenyl)pyridin-2-yl)oxy)pyrrolidine-1-carboxylate (67)**

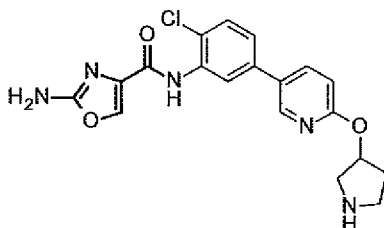


5

A mixture of *tert*-butyl (4-((2-chloro-5-(6-fluoropyridin-3-yl)phenyl)carbamoyl)oxazol-2-yl)carbamate **36** (150 mg, 0.35 mmol), sodium hydride (60% w/w in mineral oil, 21 mg, 0.52 mmol) and *tert*-butyl 3-hydroxypyrrolidine-1-carboxylate (97 mg, 0.52 mmol) in dioxane (3 mL) was heated at 80°C for 24h. A further 1 eq of alcohol and sodium hydride was added and the reaction mixture heated at 80°C for a further 4h. The reaction mixture was then partitioned between saturated NH₄Cl solution (40 mL) and EtOAc (40 mL) and the organic layer washed with water (40 mL), dried (MgSO₄) and concentrated *in vacuo*. The resulting residue was purified by column chromatography (70% EtOAc in heptane) to afford the **67** (220 mg, 100%) as a pale yellow viscous oil;

¹H NMR (400 MHz, CDCl₃) δ ppm 9.24 (s, 1H), 8.74 (d, J=2.8 Hz, 1H), 8.38 (s, 1H), 8.05 (s, 1H), 7.86 – 7.78 (m, 1H), 7.46 (d, J=8.3 Hz, 2H), 7.23 (dd, J=8.4, 2.2 Hz, 1H), 6.79 (d, J=8.6 Hz, 1H), 5.59 (d, J=4.5 Hz, 1H), 3.74 – 3.28 (m, 4H), 2.18 (s, 2H), 1.55 (s, 9H), 1.46 (s, 9H); LCMS (*m/z*): 600/602 [M+H]⁺.

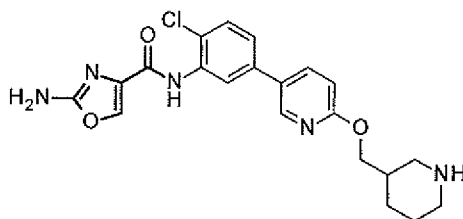
2-Amino-N-(2-chloro-5-(6-(pyrrolidin-3-yloxy)pyridin-3-yl)phenyl)oxazole-4-carboxamide (68)



To a solution of *tert*-butyl 3-((5-(3-(2-((*tert*-butoxycarbonyl)amino)oxazole-4-carboxamido)-4-chlorophenyl)pyridin-2-yl)oxy)pyrrolidine-1-carboxylate **67** (220 mg, 0.366 mmol) in DCM (10 mL) was added TFA (1 mL). The reaction was stirred at rt for 1h and then concentrated *in vacuo*. The residue was dissolved in MeCN and purified with SCX-2 cartridge and then purified by column chromatography (DCM/MeOH/7N NH₃ in MeOH, 80/20/2) to give **68** (79 mg, 54%) as a colourless solid; ¹H NMR (400 MHz, CD₃OD) δ ppm 8.60 (d, *J*=2.2 Hz, 1H), 8.43 (dd, *J*=2.6, 0.8 Hz, 1H), 7.97 (dd, *J*=8.6, 2.6 Hz, 1H), 7.92 (s, 1H), 7.57 (d, *J*=8.4 Hz, 1H), 7.40 (dd, *J*=8.4, 2.2 Hz, 1H), 6.90 (dd, *J*=8.6, 0.8 Hz, 1H), 5.57 (ddd, *J*=7.6, 4.7, 2.1 Hz, 1H), 3.29 – 3.12 (m, 3H), 3.04 (ddd, *J*=11.3, 8.4, 4.8 Hz, 1H), 2.28 – 2.02 (m, 2H); LCMS (*m/z*): 400/402 [M+H]⁺.

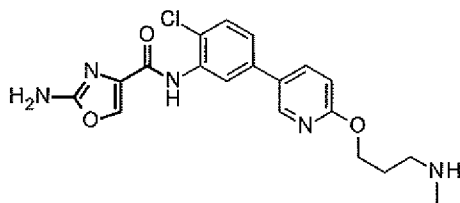
The following examples were prepared using analogous procedures:

2-Amino-N-(2-chloro-5-(6-(piperidin-3-ylmethoxy)pyridin-3-yl)phenyl)oxazole-4-carboxamide (69)



¹H NMR (400 MHz, CD₃OD) δ ppm 8.59 (d, *J*=2.2 Hz, 1H), 8.39 (dd, *J*=2.7, 0.7 Hz, 1H), 7.96 (dd, *J*=8.6, 2.6 Hz, 1H), 7.91 (s, 1H), 7.56 (d, *J*=8.4 Hz, 1H), 7.38 (dd, *J*=8.4, 2.2 Hz, 1H), 6.90 (dd, *J*=8.6, 0.8 Hz, 1H), 4.26 (dd, *J*=10.6, 5.4 Hz, 1H), 4.16 (dd, *J*=10.6, 7.4 Hz, 1H), 3.40 – 3.06 (m, 2H), 2.68 (td, *J*=12.3, 3.1 Hz, 1H), 2.57 (dd, *J*=12.3, 10.9 Hz, 1H), 2.20 – 2.05 (m, 1H), 1.96 (ddd, *J*=13.1, 3.8, 2.1 Hz, 1H), 1.82 (d quintets, *J*=13.6, 3.3 Hz, 1H), 1.63 (dddd, *J*=16.1, 13.8, 7.9, 4.0 Hz, 1H), 1.43 – 1.27 (m, 1H); LCMS (*m/z*): 428/430 [M+H]⁺.

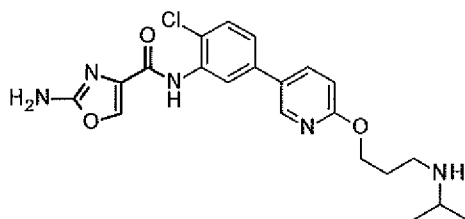
2-Amino-N-(2-chloro-5-(6-(3-(methylamino)propoxy)pyridin-3-yl)phenyl)oxazole-4-carboxamide (70)



¹H NMR (400 MHz, CD₃OD) δ ppm 8.61 (d, *J*=2.2 Hz, 1H), 8.42 (dd, *J*=2.7, 0.8 Hz, 1H), 7.99 (dd, *J*=8.6, 2.6 Hz, 1H), 7.92 (s, 1H), 7.58 (d, *J*=8.4 Hz, 1H), 7.41 (dd, *J*=8.4, 2.2 Hz, 1H), 6.92 (dd, *J*=8.7, 0.8 Hz, 1H), 4.43 (t, *J*=6.2 Hz, 2H), 2.87 – 2.77 (m, 2H), 2.47 (s, 3H), 2.05 (quintet, *J*=6.6 Hz, 2H); LCMS (*m/z*): 402/404 [M+H]⁺.

2-Amino-N-(2-chloro-5-(6-(3-(isopropylamino)propoxy)pyridin-3-yl)phenyl)oxazole-4-carboxamide (71)

10

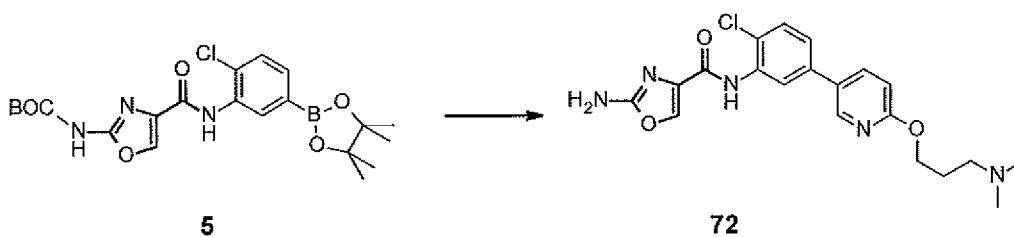


¹H NMR (400 MHz, CD₃OD) δ ppm 8.57 (d, *J*=2.2 Hz, 1H), 8.40 – 8.35 (m, 1H), 7.95 (dd, *J*=8.7, 2.6 Hz, 1H), 7.89 (s, 1H), 7.54 (d, *J*=8.4 Hz, 1H), 7.37 (dd, *J*=8.4, 2.2 Hz, 1H), 6.88 (dd, *J*=8.6, 0.8 Hz, 1H), 4.40 (t, *J*=6.2 Hz, 2H), 2.88 (hept, *J*=6.4 Hz, 1H), 2.80 (t, *J*=7.4 Hz, 2H), 2.07 – 1.94 (m, 2H), 1.11 (d, *J*=6.3 Hz, 6H); LCMS (*m/z*): 430/432 [M+H]⁺.

15

Scheme 14 – Preparation of example 72

20

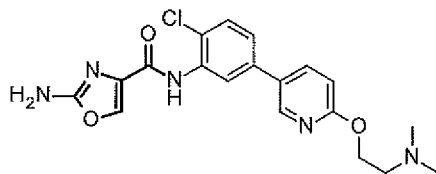


2-Amino-N-(2-chloro-5-{6-[3-(dimethylamino)propoxy]pyridin-3-yl}phenyl)-1,3-oxazole-4-carboxamide (72)

A microwave reaction vessel was charged with intermediate **5** (100 mg, 0.22 mmol),
5 PdCl₂(dppf) (12 mg, 0.014 mmol), potassium carbonate (50 mg, 0.36 mmol) and 3-[(5-bromopyridin-2-yl)oxy]-N,N-dimethylpropan-1-amine (37 mg, 0.14 mmol). Dioxane (1.2 mL) and water (0.3 mL) were added, the mixture degassed with nitrogen for 5 minutes and then the vessel sealed and stirred at 120°C for 18h. The solvents were removed *in vacuo* and the residue passed through a 1g Isolute NH₂ cartridge (eluting with 3 column
10 volumes of MeOH). The combined eluents were removed *in vacuo* to give the crude product. This was dissolved in DCM (0.7 mL), treated with TFA (0.3 mL) and stirred at rt for 18h. Concentration *in vacuo* and purification by preparative LC-MS gave **72** (8 mg, 13%) as an off white solid; ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.32 (s, 1H), 8.55 (d, *J*=2.3 Hz, 1H), 8.44 (d, *J*=2.7 Hz, 1H), 8.12 (s, 1H), 7.98 (dd, *J*=8.9, 2.7 Hz, 1H), 7.63
15 (d, *J*=8.7 Hz, 1H), 7.47 (dd, *J*=8.7, 2.3 Hz, 1H), 7.17 (s, 2H), 6.92 (d, *J*=8.9 Hz, 1H), 4.35 - 4.31 (m, 2H), 3.33 (s, 3H), 2.48 - 2.42 (m, 2H), 2.23 (s, 3H), 1.93 - 1.86 (m, 2H); LCMS: (*m/z*) = 416/418 [M+H]⁺.

The following examples were prepared using analogous procedures:

20

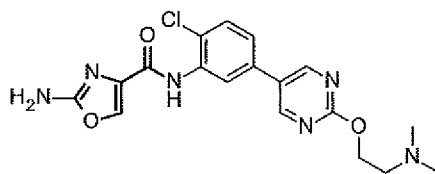
2-Amino-N-(2-chloro-5-(6-(2-(dimethylamino)ethoxy)pyridin-3-yl)phenyl)oxazole-4-carboxamide (73)

25

¹H NMR (400 MHz, CD₃OD) δ ppm 8.60 (d, *J*=2.2 Hz, 1H), 8.41 (dd, *J*=2.6, 0.7 Hz, 1H), 7.97 (dd, *J*=8.6, 2.6 Hz, 1H), 7.92 (s, 1H), 7.57 (d, *J*=8.4 Hz, 1H), 7.40 (dd, *J*=8.4, 2.2 Hz, 1H), 6.93 (dd, *J*=8.6, 0.7 Hz, 1H), 4.50 (t, *J*=5.6 Hz, 2H), 2.83 (t, *J*=5.6 Hz, 2H), 2.38 (s, 6H); LCMS: (*m/z*) = 402/404 [M+H]⁺.

30

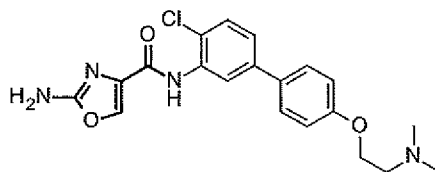
2-Amino-N-(2-chloro-5-(2-(2-(dimethylamino)ethoxy)pyrimidin-5-yl)phenyl)oxazole-4-carboxamide (74)



5

^1H NMR (400 MHz, $\text{DMSO-}d_6$) δ ppm 9.35 (s, 1H), 8.90 (s, 2H), 8.56 (d, $J=2.2$ Hz, 1H), 8.13 (s, 1H), 7.69 (d, $J=8.4$ Hz, 1H), 7.55 (dd, $J=8.4, 2.3$ Hz, 1H), 7.17 (s, 2H), 4.46 (t, $J=5.8$ Hz, 2H), 2.67 (t, $J=5.8$ Hz, 2H), 2.24 (s, 6H); LCMS: (m/z) = 403/405 [$\text{M}+\text{H}$] $^+$.

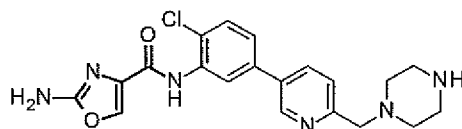
2-Amino-N-{4-chloro-4'-[2-(dimethylamino)ethoxy]biphenyl-3-yl}-1,3-oxazole-4-carboxamide (75)



15 ^1H NMR (400 MHz, $\text{DMSO-}d_6$) δ ppm 9.29 (s, 1H), 8.56 (d, $J=2.3$ Hz, 1H), 8.11 (s, 1H), 7.60 - 7.55 (m, 3H), 7.42 (dd, $J=8.2, 2.3$ Hz, 1H), 7.17 (s, 2H), 7.08 - 7.04 (m, 2H), 4.10 (t, $J=5.7$ Hz, 2H), 2.65 (t, $J=5.7$ Hz, 2H), 2.23 (s, 6H); LCMS: (m/z) = 401/403 [$\text{M}+\text{H}$] $^+$.

2-Amino-N-{2-chloro-5-[6-(piperazin-1-ylmethyl)pyridin-3-yl]phenyl}-1,3-oxazole-4-carboxamide (76)

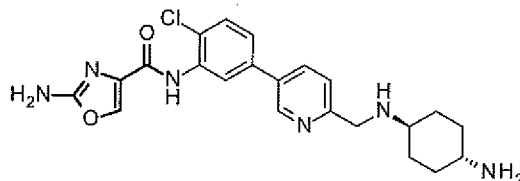
20



25 ^1H NMR (400 MHz, CD_3OD) δ ppm 8.75 (d, $J=2.3$ Hz, 1H), 8.66 (d, $J=2.3$ Hz, 1H), 8.07 (dd, $J=8.2, 2.3$ Hz, 1H), 7.90 (s, 1H), 7.63 (d, $J=8.2$ Hz, 1H), 7.59 (d, $J=8.7$ Hz, 1H),

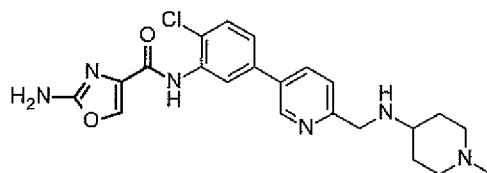
7.44 (dd, $J=8.7, 2.3$ Hz, 1H), 3.71 (s, 2H), 2.93 - 2.88 (m, 4H), 2.61 - 2.48 (m, 4H); LCMS: (m/z) = 413/415 $[M+H]^+$.

5 **2-Amino-*N*-[5-(6-[[*(trans*-4-aminocyclohexyl)amino]methyl)pyridin-3-yl)-2-chlorophenyl]-1,3-oxazole-4-carboxamide (77)**



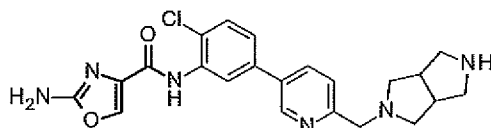
^1H NMR (400 MHz, $\text{DMSO-}d_6$) δ ppm 9.33 (br s, 1H), 8.76 (d, $J=2.3$ Hz, 1H), 8.60 (d, $J=2.3$ Hz, 1H), 8.13 (s, 1H), 8.01 (dd, $J=8.2, 2.3$ Hz, 1H), 7.67 (d, $J=8.2$ Hz, 1H), 7.56 - 7.51 (m, 2H), 7.18 (s, 2H), 3.86 (s, 2H), 2.54 - 2.45 (m, 1H), 2.36 - 2.29 (m, 1H), 1.87 (br d, $J=11.4$ Hz, 2H), 1.73 - 1.70 (m, 2H), 1.11 - 0.93 (m, 4H); LCMS: (m/z) = 441/443 $[M+H]^+$.

15 **2-Amino-*N*-[2-chloro-5-(6-[[1-methylpiperidin-4-yl]amino]methyl)pyridin-3-yl]phenyl]-1,3-oxazole-4-carboxamide (78)**



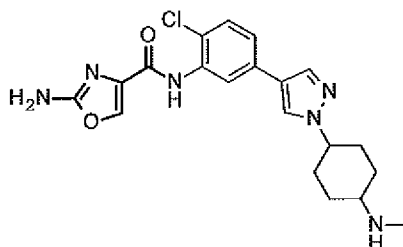
20 ^1H NMR (400 MHz, $\text{DMSO-}d_6$) δ ppm 9.34 (s, 1H), 8.76 (d, $J=1.8$ Hz, 1H), 8.60 (d, $J=2.3$ Hz, 1H), 8.13 (s, 1H), 8.01 (dd, $J=8.0, 2.5$ Hz, 1H), 7.67 (d, $J=8.2$ Hz, 1H), 7.57-7.51 (m, 2H), 7.18 (s, 2H), 3.86 (s, 2H), 2.68 (br d, $J=11.9$ Hz, 2H), 2.39 - 2.33 (m, 1H), 2.11 (s, 3H), 1.87 - 1.78 (m, 4H), 1.33 - 1.23 (m, 2H); LCMS: (m/z) = 441/443 $[M+H]^+$.

25 **2-Amino-*N*-{2-chloro-5-[6-(hexahydropyrrolo[3,4-*c*]pyrrol-2(1H)-yl)methyl]pyridin-3-yl]phenyl}-1,3-oxazole-4-carboxamide (79)**



¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.34 (br s, 1H), 8.75 (d, *J*=2.3 Hz, 1H), 8.60 (d, *J*=2.3 Hz, 1H), 8.13 (s, 1H), 8.03 (dd, *J*=8.2, 2.3 Hz, 1H), 7.67 (d, *J*=8.2 Hz, 1H), 7.54 – 7.51 (m, 2H), 7.18 (s, 2H), 3.68 (s, 2H), 2.79 – 2.75 (m, 2H), 2.62 – 2.52 (m, 6H), 2.32 – 2.29 (m, 2H); LCMS: (*m/z*) = 439/441 [M+H]⁺.

2-Amino-N-(2-chloro-5-{1-[4-(methylamino)cyclohexyl]-1H-pyrazol-4-yl}phenyl)-1,3-oxazole-4-carboxamide (80)



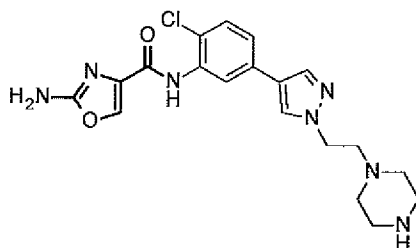
10

Inseparable mixture of *cis*- and *trans*- isomers; ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.23 (br s, total 1H isomer A and 1H isomer B), 8.43 (d, *J*=2.3 Hz, 1H isomer A), 8.42 (d, *J*=2.3 Hz, isomer B), 8.24 (s, 1H, isomer A), 8.23 (s, 1H, isomer B), 8.09 (br s, total 1H isomer A and 1H isomer B), 7.82 (br s, 1H isomer A and 1H isomer B), 7.51 (d, *J*=8.7 Hz, 1H isomer B), 7.51 (d, *J*=8.2 Hz, 1H, isomer A), 7.41 (dd, *J*=8.2, 2.3 Hz, 1H isomer A), 7.38 (dd, *J*=8.7, 2.3 Hz, 1H isomer B), 7.15 (br s, total 2H isomer A and 2H isomer B), 4.20 – 4.11 (m, total 1H isomer A and 1H isomer B), 2.60 (t, *J*=3.4 Hz, 1H, isomer A), 2.34 – 2.30 (m, 1H, isomer B), 2.28 (s, 3H, isomer B), 2.26 (s, 3H, isomer A), 2.17 – 1.98 (m, total 2H isomer A and 2H isomer B), 1.84 – 1.72 (m, total 4H isomer A and 4H isomer B), 1.61 – 1.52 (m, total 2H isomer A and 2H isomer B); LCMS: (*m/z*) = 415/417 [M+H]⁺.

20

2-Amino-N-(2-chloro-5-{1-[2-(piperazin-1-yl)ethyl]-1H-pyrazol-4-yl}phenyl)-1,3-oxazole-4-carboxamide (81)

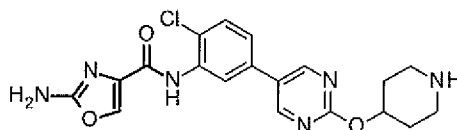
25



¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.24 (s, 1H), 8.44 (d, *J*=2.3 Hz, 1H), 8.19 (s, 1H), 8.10 (s, 1H), 7.83 (s, 1H), 7.52 (d, *J*=8.2 Hz, 1H), 7.37 (dd, *J*=8.2, 2.3 Hz, 1H), 7.15 (s, 2H), 4.23 (t, *J*=6.7 Hz, 2H), 2.70 – 2.67 (m, 2H), 2.65 (t, *J*=4.8 Hz, 4H), 2.35 – 2.32 (m, 4H); LCMS: (*m/z*) = 416/418 [M+H]⁺.

2-Amino-N-(2-chloro-5-[2-(piperidin-4-yl)oxy]pyrimidin-5-yl)phenyl)-1,3-oxazole-4-carboxamide (82)

10

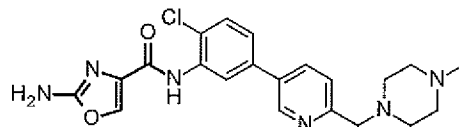


¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.34 (s, 1H), 8.87 (s, 2H), 8.54 (d, *J*=2.3 Hz, 1H), 8.12 (s, 1H), 7.68 (d, *J*=8.2 Hz, 1H), 7.52 (dd, *J*=8.3, 2.3 Hz, 1H), 7.16 (s, 2H), 5.10 – 5.03 (m, 1H), 2.99 (dt, *J*=12.6, 4.0 Hz, 2H), 2.64 – 2.57 (m, 2H), 2.01 – 1.97 (m, 2H), 1.60 – 1.56 (m, 2H); LCMS: (*m/z*) = 415/417 [M+H]⁺.

15

2-Amino-N-(2-chloro-5-{6-[(4-methylpiperazin-1-yl)methyl]pyridin-3-yl}phenyl)-1,3-oxazole-4-carboxamide (83)

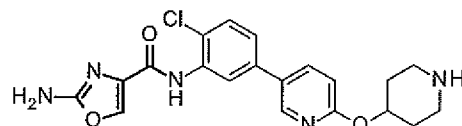
20



¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.34 (s, 1H), 8.77 (d, *J*=1.8 Hz, 1H), 8.61 (d, *J*=2.3 Hz, 1H), 8.13 (s, 1H), 8.03 (dd, *J*=8.2, 2.3 Hz, 1H), 7.68 (d, *J*=8.2 Hz, 1H), 7.54 –

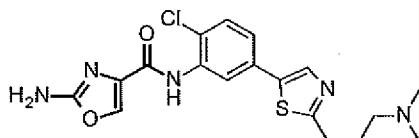
7.51 (m, 2H), 7.17 (s, 2H), 3.63 (s, 2H), 2.45 (br s, 4H), 2.33 (br s, 4H), 2.16 (s, 3H);
LCMS: (*m/z*) = 427/429 [M+H]⁺.

5 **2-Amino-N-(2-chloro-5-[6-(piperidin-4-yloxy)pyridin-3-yl]phenyl)-1,3-oxazole-4-carboxamide (84)**



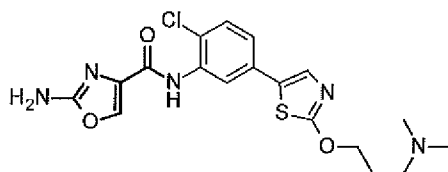
10 ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.31 (s, 1H), 8.53 (d, *J*=1.8 Hz, 1H), 8.43 (d, *J*=2.8 Hz, 1H), 8.12 (s, 1H), 7.95 (dd, *J*=8.7, 2.8 Hz, 1H), 7.63 (d, *J*=8.2 Hz, 1H), 7.45 (dd, *J*=8.5, 2.1 Hz, 1H), 7.17 (s, 2H), 6.88 (d, *J*=8.7 Hz, 1H), 5.12 – 5.05 (m, 1H), 2.96 (dt, *J*=12.7, 4.0 Hz, 2H), 2.61 – 2.55 (m, 2H), 1.99 – 1.94 (m, 2H), 1.54 – 1.45 (m, 2H);
LCMS: (*m/z*) = 414/416 [M+H]⁺.

15 **2-Amino-N-(2-chloro-5-{2-[3-(dimethylamino)propyl]-1,3-thiazol-5-yl}phenyl)-1,3-oxazole-4-carboxamide (85)**



20 ¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.30 (s, 1H), 8.52 (d, 8.54 (d, *J*=1.7 Hz, 1H), 8.12 (s, 1H), 8.07 (s, 1H), 7.62 – 7.60 (m, 1H), 7.49 (d, *J*=8.0, 2.3 Hz, 1H), 7.17 (br s, 2H), 3.00 (t, *J*=7.7 Hz, 2H), 2.28 (t, *J*=7.8 Hz, 2H), 2.13 (s, 6H), 1.86 (quintet, *J*=7.3 Hz, 2H);
LCMS: (*m/z*) = 406/408 [M+H]⁺.

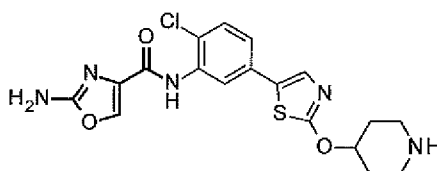
25 **2-Amino-N-(2-chloro-5-{2-[3-(dimethylamino)propoxy]-1,3-thiazol-5-yl}phenyl)-1,3-oxazole-4-carboxamide (86)**



¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.27 (s, 1H), 8.33 (d, *J*=2.3 Hz, 1H), 8.11 (s, 1H), 7.71 (s, 1H), 7.58 (d, *J*=8.2 Hz, 1H), 7.29 (dd, *J*=8.7, 2.3 Hz, 1H), 7.17 (s, 2H), 3.74 (t, *J*=7.3 Hz, 2H), 2.22 (t, *J*=6.9 Hz, 2H), 2.13 (s, 6H), 1.83 – 1.76 (m, 2H); LCMS: (*m/z*) = 422/424 [M+H]⁺.

2-Amino-*N*-(2-chloro-5-[2-(piperidin-4-yloxy)-1,3-thiazol-5-yl]phenyl)-1,3-oxazole-4-carboxamide (87)

10

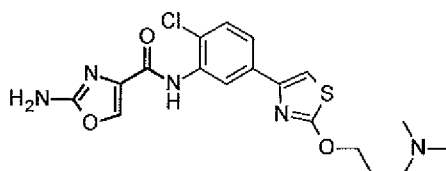


¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.28 (s, 1H), 8.42 (d, *J*=2.3 Hz, 1H), 8.12 (s, 1H), 7.63 (s, 1H), 7.58 (d, *J*=8.2 Hz, 1H), 7.40 (dd, *J*=8.2, 2.3 Hz, 1H), 7.17 (s, 2H), 5.04 – 4.97 (m, 1H), 3.00 (dt, *J*=12.7, 4.4 Hz, 2H), 2.68 – 2.62 (m, 2H), 2.07 – 2.03 (m, 2H), 1.67 – 1.58 (m, 2H); LCMS: (*m/z*) = 420/422 [M+H]⁺.

15

2-Amino-*N*-(2-chloro-5-{2-[3-(dimethylamino)propoxy]-1,3-thiazol-4-yl}phenyl)-1,3-oxazole-4-carboxamide (88)

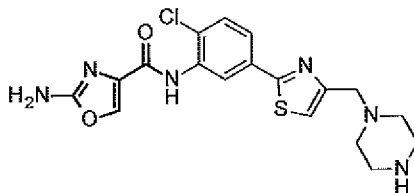
20



¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.29 (s, 1H), 8.75 (d, *J*=2.3 Hz, 1H), 8.11 (s, 1H), 7.67 – 7.64 (m, 1H), 7.59 (d, *J*=8.3 Hz, 1H), 7.51 (s, 1H), 7.16 (s, 2H), 4.47 (t, *J*=6.4 Hz,

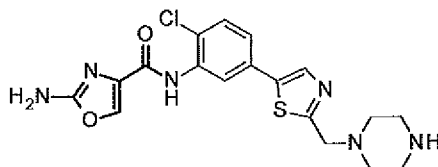
2H), 2.35 (t, $J=7.1$ Hz, 2H), 2.14 (s, 6H), 1.92 (quintet, $J=6.8$ Hz, 2H); LCMS: (m/z) = 422/424 $[M+H]^+$.

5 **2-Amino-*N*-{2-chloro-5-[4-(piperazin-1-ylmethyl)-1,3-thiazol-2-yl]phenyl}-1,3-oxazole-4-carboxamide (89)**



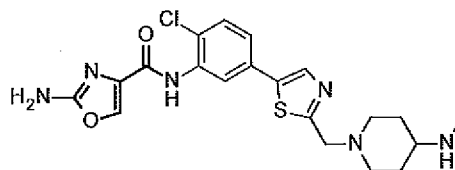
10 $^1\text{H NMR}$ (400 MHz, CD_3OD) δ ppm 8.97 (d, $J=1.8$ Hz, 1H), 7.91 (s, 1H), 7.75 (dd, $J=8.7$, 2.3 Hz, 1H), 7.57 (d, $J=8.2$ Hz, 1H), 7.46 (s, 1H), 3.74 (s, 2H), 2.91 (t, $J=4.8$ Hz, 4H), 2.61 (br s, 4H); LCMS: (m/z) = 419/421 $[M+H]^+$.

15 **2-Amino-*N*-{2-chloro-5-[2-(piperazin-1-ylmethyl)-1,3-thiazol-5-yl]phenyl}-1,3-oxazole-4-carboxamide (90)**



20 $^1\text{H NMR}$ (400 MHz, CD_3OD) δ ppm 8.64 (d, $J=2.3$ Hz, 1H), 7.98 (s, 1H), 7.90 (s, 1H), 7.52 (d, $J=8.7$ Hz, 1H), 7.42 (dd, $J=8.2$, 2.3 Hz, 1H), 3.86 (s, 2H), 2.93 (t, $J=5.0$ Hz, 4H), 2.63 (br s, 4H); LCMS: (m/z) = 419/421 $[M+H]^+$.

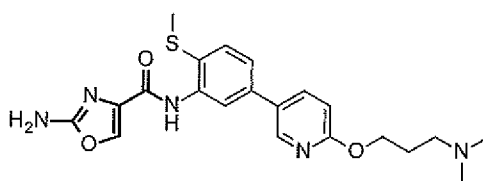
2-Amino-*N*-[2-chloro-5-(2-[4-(methylamino)piperidin-1-yl]methyl)-1,3-thiazol-5-yl]phenyl]-1,3-oxazole-4-carboxamide (91)



25

¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.30 (s, 1H), 8.56 (d, *J*=2.3 Hz, 1H), 8.12 (s, 1H), 8.09 (s, 1H), 7.61 (d, *J*=9.6 Hz, 1H), 7.51 (dd, *J*=7.8, 2.3 Hz, 1H), 7.19 (s, 2H), 3.79 (s, 2H), 2.88 (br d, *J*=11.9 Hz, 2H), 2.34 – 2.26 (m, 1H), 2.26 (s, 3H), 2.22 – 2.15 (m, 2H),
 5 1.82 – 1.78 (m, 2H), 1.33 – 1.23 (m, 2H); LCMS: (*m/z*) = 447/449 [M+H]⁺.

2-Amino-*N*-[5-{6-[3-(dimethylamino)propoxy]pyridin-3-yl}-2-(methylsulfanyl)phenyl]-1,3-oxazole-4-carboxamide (92)

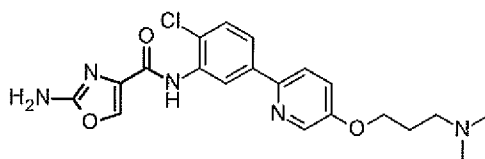


10

¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.48 (s, 1H), 8.47 (d, *J*=1.8 Hz, 1H), 8.43 (d, *J*=3.2 Hz, 1H), 8.08 (s, 1H), 7.96 (dd, *J*=8.7, 2.8 Hz, 1H), 7.59 (d, *J*=8.2 Hz, 1H), 7.45 (dd, *J*=8.0, 2.1 Hz, 1H), 7.15 (s, 2H), 6.91 (dd, *J*=8.7, 0.9 Hz, 1H), 4.32 (t, *J*=6.6 Hz,
 15 2H), 2.48 (s, 3H), 2.34 (t, *J*=7.1 Hz, 2H), 2.14 (s, 6H), 1.86 (quintet, *J*=6.9 Hz, 2H); LCMS: (*m/z*) = 428 [M+H]⁺.

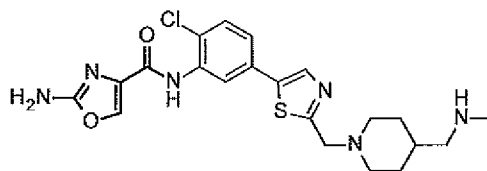
2-Amino-*N*-(2-chloro-5-{5-[3-(dimethylamino)propoxy]pyridin-2-yl}phenyl)-1,3-oxazole-4-carboxamide (93)

20



¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.30 (s, 1H), 8.96 (d, *J*=2.3 Hz, 1H), 8.39 (d, *J*=2.8 Hz, 1H), 8.11 (s, 1H), 7.88 (d, *J*=9.2 Hz, 1H), 7.79 (dd, *J*=8.7, 2.3 Hz, 1H), 7.61
 25 (d, *J*=8.7 Hz, 1H), 7.51 – 7.48 (m, 1H), 7.17 (s, 2H), 4.13 (t, *J*=6.4 Hz, 2H), 2.36 (t, *J*=7.1 Hz, 2H), 2.15 (s, 6H), 1.88 (quintet, *J*=6.6 Hz, 2H); LCMS: (*m/z*) = 416/418 [M+H]⁺.

2-Amino-N-[2-chloro-5-[2-({4-[(methylamino)methyl]piperidin-1-yl)methyl]-1,3-thiazol-5-yl]phenyl]-1,3-oxazole-4-carboxamide (94)



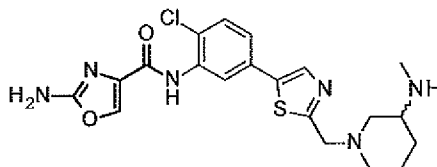
5

^1H NMR (400 MHz, DMSO- d_6) δ ppm 9.30 (s, 1H), 8.56 (d, $J=2.3$ Hz, 1H), 8.13 – 8.12 (m, 1H), 8.08 (s, 1H), 7.62 – 7.59 (m, 1H), 7.51 (dd, $J=8.7, 2.3$ Hz, 1H), 7.19 (s, 2H), 3.79 – 3.78 (m, 2H), 2.92 (br d, $J=11.0$ Hz, 2H), 2.35 (d, $J=6.4$ Hz, 1H), 2.25 (s, 3H), 2.14 – 2.08 (m, 2H), 1.68 (br d, $J=11.4$ Hz, 2H), 1.57 (br d, $J=11.0$ Hz, 1H), 1.44 – 1.36

10

(m, 1H), 1.23 – 1.13 (m, 2H); LCMS: (m/z) = 461/463 [$M+H$] $^+$.

2-Amino-N-[2-chloro-5-(2-{{3-(methylamino)piperidin-1-yl}methyl})-1,3-thiazol-5-yl]phenyl]-1,3-oxazole-4-carboxamide (95)



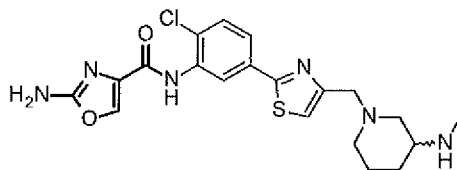
15

^1H NMR (400 MHz, DMSO- d_6) δ ppm 9.31 (s, 1H), 8.55 (d, $J=2.3$ Hz, 1H), 8.12 (s, 1H), 8.09 (s, 1H), 7.61 (d, $J=7.8$ Hz, 1H), 7.52 – 7.50 (m, 1H), 7.19 (s, 2H), 3.80 (s, 2H), 2.95 (br d, $J=8.2$ Hz, 1H), 2.76 (br d, $J=11.0$ Hz, 1H), 2.47 – 2.40 (m, 1H), 2.24 (s, 3H), 2.17 – 2.08 (m, 1H), 1.90 (t, $J=9.8$ Hz, 1H), 1.84 – 1.80 (m, 1H), 1.66 – 1.61 (m, 1H), 1.54 – 1.43 (m, 1H), 1.03 – 0.94 (m, 1H); LCMS: (m/z) = 447/449 [$M+H$] $^+$.

20

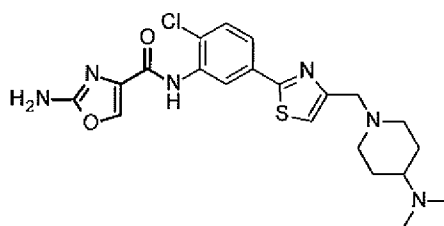
2-Amino-N-[2-chloro-5-(4-{{3-(methylamino)piperidin-1-yl}methyl})-1,3-thiazol-2-yl]phenyl]-1,3-oxazole-4-carboxamide (96)

25



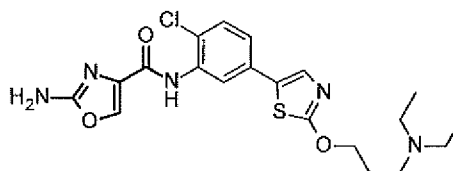
¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.34 (s, 1H), 8.93 (d, *J*=2.3 Hz, 1H), 8.14 (s, 1H), 7.72 – 7.69 (m, 1H), 7.67 (d, *J*=8.2 Hz, 1H), 7.54 (s, 1H), 7.20 (s, 2H), 3.64 (s, 2H), 2.91
5 (br d, *J*=8.2 Hz, 1H), 2.74 – 2.68 (m, 1H), 2.43 – 2.35 (m, 1H), 2.24 (s, 3H), 2.07 – 2.00 (m, 1H), 1.82 – 1.77 (m, 2H), 1.61 (dt, *J*=13.2, 3.5 Hz, 1H), 1.49 – 1.38 (m, 1H), 1.01 – 0.91 (m, 1H); LCMS: (*m/z*) = 447/449 [M+H]⁺.

2-Amino-*N*-[2-chloro-5-(4-[[4-(dimethylamino)piperidin-1-yl]methyl]-1,3-thiazol-2-yl)phenyl]-1,3-oxazole-4-carboxamide (97)
10



¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.34 (s, 1H), 8.92 (d, *J*=1.8 Hz, 1H), 8.14 (s, 1H),
15 7.92 – 7.69 (m, 1H), 7.68 – 7.65 (m, 1H), 7.54 (s, 1H), 7.20 (s, 2H), 3.62 (s, 2H), 2.92 (br d, *J*=11.9 Hz, 2H), 2.14 (s, 6H), 2.01 (br t, *J*=10.8 Hz, 3H), 1.70 (br d, *J*=12.4 Hz, 2H), 1.38 (qd, *J*=11.9, 3.7 Hz, 2H); LCMS: (*m/z*) = 461/463 [M+H]⁺.

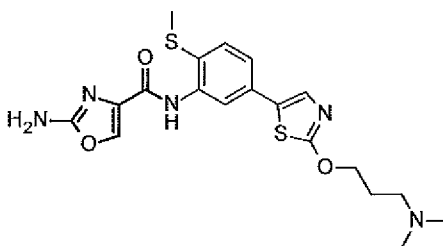
2-Amino-*N*-(2-chloro-5-(2-[3-(diethylamino)propoxy]-1,3-thiazol-5-yl)phenyl)-1,3-oxazole-4-carboxamide (98)
20



¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.27 (s, 1H), 8.32 (d, *J*=2.3 Hz, 1H), 8.12 (s, 1H), 7.72 (s, 1H), 7.58 (d, *J*=8.2 Hz, 1H), 7.29 (dd, *J*=8.2, 2.3 Hz, 1H), 7.18 (s, 2H), 3.73 (t, *J*=7.1 Hz, 2H), 2.43 (q, *J*=7.3 Hz, 4H), 2.38 (t, *J*=6.9 Hz, 2H), 1.79 (quintet, *J*=7.1 Hz, 2H), 0.92 (t, *J*=7.1 Hz, 6H); LCMS: (*m/z*) = 450/452 [M+H]⁺.

5

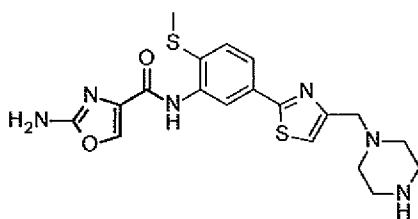
2-Amino-*N*-[5-{2-[3-(dimethylamino)propoxy]-1,3-thiazol-5-yl}-2-(methylsulfanyl)phenyl]-1,3-oxazole-4-carboxamide (99)



10

¹H NMR (400 MHz, CD₃OD) δ ppm 8.34 (d, *J*=1.8 Hz, 1H), 7.86 (s, 1H), 7.52 (d, *J*=8.2 Hz, 1H), 7.39 (s, 1H), 7.18 (dd, *J*=8.2, 2.3 Hz, 1H), 3.89 (t, *J*=6.6 Hz, 2H), 3.12 – 3.08 (m, 2H), 2.82 (s, 6H), 2.41 (s, 3H), 2.16 – 2.09 (m, 2H); LCMS: (*m/z*) = 434 [M+H]⁺.

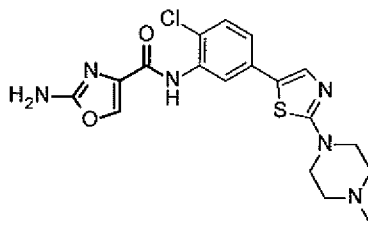
2-Amino-*N*-{2-(methylsulfanyl)-5-[4-(piperazin-1-ylmethyl)-1,3-thiazol-2-yl]phenyl}-1,3-oxazole-4-carboxamide (100)



¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.40 (s, 1H), 8.75 (d, *J*=1.7 Hz, 1H), 8.09 (s, 1H), 7.71 – 7.69 (m, 1H), 7.58 (d, *J*=8.6 Hz, 1H), 7.50 (s, 1H), 7.15 (s, 2H), 3.60 (s, 2H), 2.69 (t, *J*=4.6 Hz, 4H), 2.53 (s, 3H), 2.40 – 2.35 (m, 4H); LCMS: (*m/z*) = 430 [M+H]⁺.

2-Amino-*N*-{2-chloro-5-[2-(4-methylpiperazin-1-yl)-1,3-thiazol-5-yl]phenyl}-1,3-oxazole-4-carboxamide (101)

25



¹H NMR (400 MHz, DMSO-*d*₆) δ ppm 9.24 (s, 1H), 8.40 (d, *J*=2.3 Hz, 1H), 8.10 (s, 1H), 7.62 (s, 1H), 7.52 (d, *J*=8.6 Hz, 1H), 7.34 (dd, *J*=8.0, 2.3 Hz, 1H), 7.16 (s, 2H), 3.47 – 3.45 (m, 4H), 2.44 – 2.42 (m, 4H), 2.23 (s, 3H); LCMS: (*m/z*) = 419/421 [M+H]⁺.

PAICS 2D Cell Proliferation Assay

Cell Culture

10 Frozen cryovial stocks of LM2 and MDA-MB-231 breast cancer cell lines were thawed and cultured in 4.5 g/L glucose DMEM (31966-047, Invitrogen) supplemented with 10% Fetal Bovine Serum (10500-064, Invitrogen) until confluent in a T175 flask. Cells were incubated at 37°C with 5% CO₂ in a humidified incubator.

15 Day 1- Cell Plating

Cells were collected from T175 flasks by washing with PBS (10010-056, Invitrogen) and dissociated using Accutase (A6964, Sigma Aldrich). This was followed by centrifuging (1200rpm, 5min) and resuspending the cells in 10ml of media containing 1% penicillin/streptomycin (15140-122, Invitrogen). Cells were counted by trypan blue
20 exclusion method using an automated cell counter (Cellometer) and diluted to a concentration of 40,000/ml. 1000 cells were then seeded into 384-well white-walled plates (Corning, 3707) in a volume of 25µl. Plates were incubated overnight at 37°C with 5% CO₂.

25 Day 2- Compound addition

After 24 hours, PAICS compounds were added onto the assay plates using an HP D300 Digital Dispenser (Hewlett Packard). 10mM stocks were used and dispensed as a 10 point concentration response curve (CRC) ½ log dilution series with a final top concentration of 100µM. Controls were added onto each plate: positive controls
30 included Staurosporine CRC (100µM stock dispensed as a 10 point CRC ½ log dilution

series with a final top concentration of 1µM) and PAICS compound MRT00211919 (10mM stock dispensed as a 10 point CRC ½ log dilution series with a final top concentration of 100µM); a high control of Staurosporine (0.1µM (LM2), 0.316µM (MDA-MB-231) FAC) and a low control of media containing 1% DMSO. A final concentration of 1% DMSO was then added across the plates for normalisation. Following compound addition, assay plates were incubated for 72 hours at 37°C with 5% CO₂.

Day 5- Cytotoxicity and Cell Viability Assays

After 72h incubation, dead and viable cells were measured in each assay plate.

10 Cytotoxicity was assessed through fluorescent DNA staining using the CellTox™ Green Cytotoxicity Assay kit (G8743, Promega) following the manufacturer's multiplexing protocol. Impaired cell membrane integrity results in the access of dye to stain DNA allowing quantification of dead cells. Cell viability was determined following ATP quantification using the CellTiter-Glo® Luminescent Cell Viability Assay kit (G7572, 15 Promega) following the manufacturer's protocol. ATP released from lysed cells allowed for the generation of a signal proportional to the number of cells present in the well. All endpoint reads were performed using the PheraSTAR Plus (BMG Labtech). Data is expressed as % viable cells and % dead cells against controls (mean ± SEM of duplicate curves).

20

Biochemical assay to measure PAICS activity

Assay Principle:

25 The biochemical assay was used to measure PAICS SAICAR synthetase-mediated conversion of CAIR, aspartic acid and ATP to SAICAR, ADP and inorganic phosphate (P_i), post-AIR/CAIR equilibration (see Figure 3). This was achieved by detecting the ADP generated during the reaction, using Bellbrook Lab's Transcreener ADP² FI assay. An increase in the fluorescence intensity is directly proportional to the amount of PAICS activity.

30

The Transcreener ADP² FI assay is a homogeneous competitive displacement fluorescence intensity assay which uses direct immunodetection of ADP. Displacement of the tracer by ADP causes an increase in the fluorescence at excitation 590nm and emission 617nm (Figure 3).

Method:

5 $5\mu\text{l}$ WT full length PAICS (final assay concentration, *fac*, 2.5nM) in basic buffer, added to black, non-binding, 384-well plates (Corning #3575), columns 1 to 22. $5\mu\text{l}$ basic buffer was added to columns 23+24 (negative control). PAICS stock at $31.9\mu\text{M}$ from the PI's lab (Steve Firestine). Basic buffer contained 50mM Tris-HCl pH8 and 0.5mM EDTA, *fac*.

10 $1\mu\text{l}$ compound in 100% DMSO added per well, or $1\mu\text{l}$ 100% DMSO to positive controls (columns 1+2) and negative controls (columns 23+24). Final assay top concentration of compound either 1mM or $30\mu\text{M}$, serially diluted with half-log dilutions across the plate in duplicate (one 10-point concentration response curve across wells 3 to 12, another across 13 to 22). Compounds pre-incubated with PAICS enzyme for 30mins at RT.

15 $2\mu\text{l}$ CAIR added (*fac* $10\mu\text{M}$) in basic buffer plus 25mM MgCl_2 and 50mM KHCO_3 *fac* to all wells. CAIR stock 50mM from Steve Firestine's lab. 1hr RT incubation for AIR-CAIR equilibration. *NB: ~50% CAIR is decarboxylated during equilibration therefore $5\mu\text{M}$ remains for the synthetase reaction.*

20 $2\mu\text{l}$ ATP/aspartic acid added (*fac* $30\mu\text{M}/180\mu\text{M}$) in reaction buffer to all wells. 30mins RT incubation for appropriate level of ATP turnover. Reaction buffer contained basic buffer plus 10mM DTT, 0.01% BSA and 0.01% Brij 35, *fac*.

25 $10\mu\text{l}$ ADP detection reagent added to all wells (as per instructions, Transcreener ADP² FI kit, BellBrook Labs #3013-10K). Incubation for 1h at RT to allow antibody equilibration. Fluorescence intensity determined using a Tecan Safire2 (excitation at 590nm, emission at 617nm).

30 The results of the above assays for selected compounds of the invention are shown in Table 1.

Various modifications and variations of the described aspects of the invention will be apparent to those skilled in the art without departing from the scope and spirit of the invention. Although the invention has been described in connection with specific preferred embodiments, it should be understood that the invention as claimed should

not be unduly limited to such specific embodiments. Indeed, various modifications of the described modes of carrying out the invention which are obvious to those skilled in the relevant fields are intended to be within the scope of the following claims.

5

PAICS Biochemical activity IC₅₀

A = < 10 nM; B = 10 – 25 nM; C = 25 – 100 nM; D = > 100 nM; nt = not tested

PAICS 2D cell proliferation Activity EC₅₀

5 A = < 100 nM; B = 100 – 250 nM; C = 250 – 500 nM; D = 500 – 1000 nM

Table 1:

Example number	PAICS Biochemical IC₅₀	PAICS 2D Cell Proliferation EC₅₀
37	D	B
38	B	C
39	D	C
40	C	C
41	C	B
42	D	B
43	C	B
44	D	B
45	D	B
47	B	A
48	B	B
49	A	A
50	B	B
51	A	B
52	B	B
53	B	B
54	B	C
55	B	B

56	B	B
57	C	C
58	C	C
59	C	C
60	D	C
61	C	C
62	D	B
63	D	C
64	D	C
65	C	C
66	D	B
68	B	C
69	B	C
70	B	B
71	B	B
72	B	B
73	C	C
74	C	C
75	C	B
76	C	C
77	B	C
78	B	B
79	B	C
80	A	A

81	C	C
82	B	C
83	C	C
84	B	C
85	nt	A
86	A	A
87	A	A
88	C	C
89	A	A
90	B	B
91	A	A
92	C	C
93	C	C
94	B	A
95	A	B
96	nt	A
97	nt	B
98	nt	A
99	nt	B
100	nt	A
101	nt	B

CRISPR editing

Cas9 nuclease-mediated gene editing (Sander and Joung, 2014) was performed using a CRISPR RNA guide sequence (TACGAATTGTTAGACAGTCC, PAM: AGG) targeting exon 3 in the human PAICS gene (accession number: NM_006452).

Stable cell clones were isolated and gene disruption confirmed by DNA sequencing and the absence of PAICS cellular protein expression confirmed by Western blotting.

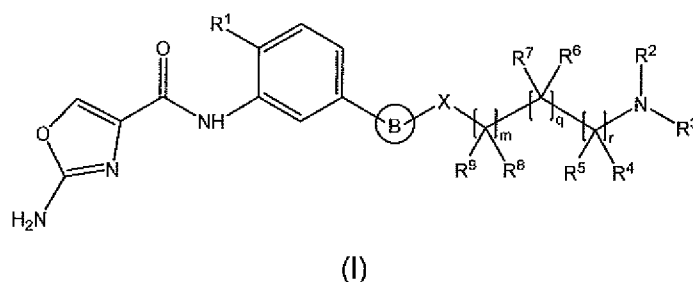
5 A small set of compounds were tested against wild type LM2 and CRISPR edited LM2 to see to what extent the cellular potency was driven by PAICS mediated effects. The data is shown in Table 2.

Table 2

Example number	Potency shift: CRISPR Cell
	EC_{50}/WT Cell EC_{50} 10
37	>10
47	>80
86	>30

CLAIMS

1. A compound of formula (I), or a pharmaceutically acceptable salt or ester thereof,



wherein:

B is an aryl or heteroaryl group, each of which is optionally substituted by one or more R^{10} groups;

X is selected from O, $(CR^{11}R^{12})_p$ and $(CR^{11}R^{12})_pCO$;

each R^1 is independently selected from Cl and SR^{13} ;

R^2 is selected from H, alkyl, cycloalkyl, heterocycloalkyl and COR^{33} , wherein said alkyl, cycloalkyl or heterocycloalkyl group is optionally substituted by one or more R^{14} substituents;

R^3 is selected from alkyl, cycloalkyl and heterocycloalkyl, each of which is optionally substituted with one or more R^{15} substituents; or

R^2 and R^3 are linked together with the nitrogen to which they are attached to form a saturated heterocyclic group optionally containing one or more additional heteroatoms selected from O, N and S and optionally further substituted by one or more R^{16} groups;

each R^4 and R^5 is independently selected from H, alkyl, $(CH_2)_tOR^{24}$ and $(CH_2)_uNR^{25}R^{26}$;

or

one of R^4 and R^5 is H or alkyl and the other is linked to R^3 to form a saturated heterocyclic group;

each R^6 and R^7 is independently selected from H, alkyl, $(CH_2)_vOR^{27}$ and $(CH_2)_wNR^{28}R^{29}$;

or

one of R^6 and R^7 is H or alkyl and the other is linked to R^3 to form a saturated heterocyclic group;

each R^8 and R^9 is independently selected from H, alkyl, $(CH_2)_xOR^{30}$ and $(CH_2)_yNR^{31}R^{32}$;

or

one of R⁸ and R⁹ is H or alkyl and the other is linked to R³ to form a saturated heterocyclic group; or one of R⁸ or R⁹ is linked to one of R⁴ or R⁵ to form a cyclic group; R¹⁰ is selected from alkyl, OH, halogen, alkoxy, CO₂-alkyl, COOH, CO-alkyl, NO₂ and CN;

each R¹⁴, R¹⁵, R¹⁶ and R³³ is independently selected from (CH₂)_s-R¹⁷,

R¹⁷ is selected from alkyl, NR¹⁸R¹⁹, OR²⁰, SR²¹, COR²² and CO₂R²³;

R¹¹, R¹² and R¹⁸-R³² are each independently selected from H and alkyl;

R¹³ is alkyl;

m, q and r are each independently 0, 1 or 2;

p is 0 or 1;

such that the sum of m, p, q and r is 0, 1, 2, 3, 4 or 5, preferably 0, 1, 2, 3 or 4; and

each of s, t, u, v, w, x, y is independently 0, 1, 2, 3 or 4.

2. A compound according to claim 1 wherein B is a 5- or 6-membered monocyclic aryl or heteroaryl group, each of which is optionally substituted by one or more R¹⁰ groups.

3. A compound according to claim 1 or claim 2 wherein:

each R⁴ and R⁵ is independently selected from H and alkyl; or

one of R⁴ and R⁵ is H or alkyl and the other is linked to R³ to form a saturated heterocyclic group;

each R⁶ and R⁷ is independently selected from H and alkyl; or

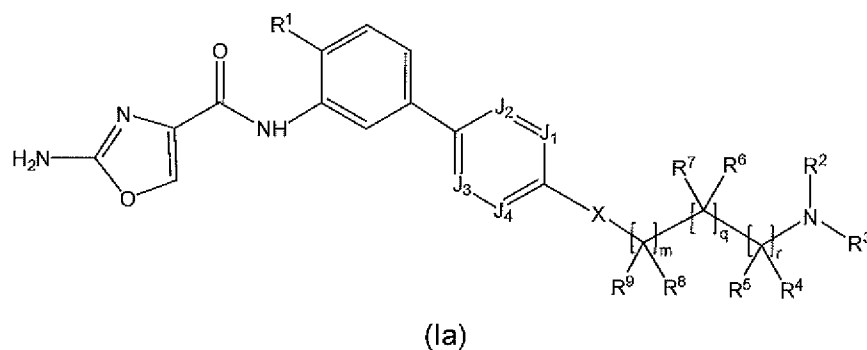
one of R⁶ and R⁷ is H or alkyl and the other is linked to R³ to form a saturated heterocyclic group;

each R⁸ and R⁹ is independently selected from H and alkyl; or

one of R⁸ and R⁹ is H or alkyl and the other is linked to R³ to form a saturated heterocyclic group.

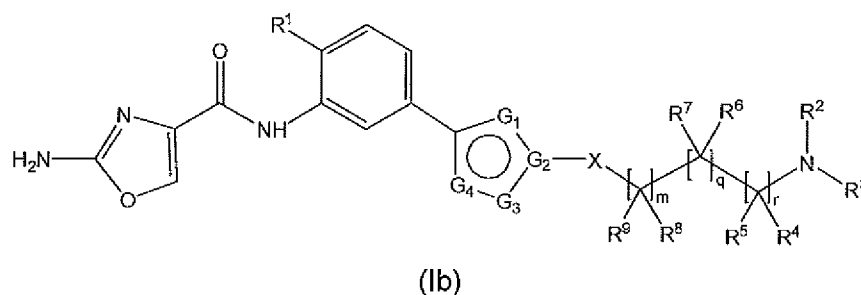
4. A compound according to any preceding claim wherein B is selected from thienyl, furanyl, pyrrolyl, pyridinyl, oxazolyl, pyrazinyl, thiazolyl, imidazolyl, pyrazolyl, isoxazolyl, isothiazolyl, oxadiazolyl, triazolyl, thiadiazolyl, pyrimidinyl, pyridazinyl and triazinyl.

5. A compound according to any preceding claim wherein B is selected from thiazolyl, pyridinyl, pyrazolyl, pyrimidinyl and phenyl.
6. A compound according to any one of claims 1 to 5 which is of formula (Ia), or a pharmaceutically acceptable salt or ester thereof,



wherein J_1 , J_2 , J_3 and J_4 are each independently selected from =N, CH and CR^{10} and X, R^{1-10} , m, q and r are as defined in claim 1.

7. A compound according to claim 6 wherein J_1 is CH, CR^{10} or =N, and J_2 , J_3 and J_4 are all CH or CR^{10} .
8. A compound according to claim 6 wherein J_1 , J_2 , J_3 and J_4 are all CH.
9. A compound according to claim 6 wherein J_1 is =N and J_2 , J_3 and J_4 are all CH.
10. A compound according to any one of claims 1 to 5 which is of formula (Ib), or a pharmaceutically acceptable salt or ester thereof,



wherein:

- (i) G_1 is S, G_2 is =C, one of G_3 and G_4 is =N and the other is CH or CR^{10} ; or
 - (ii) G_3 is S, G_1 is =N, G_2 is =C, and G_4 is CH or CR^{10} ;
 - (iii) G_1 and G_4 are each selected from CH and CR^{10} , G_2 is N and G_3 is =N; or
 - (iv) G_3 and G_4 are each selected from CH and CR^{10} , G_2 is N and G_1 is =N;
- and X, R^{1-10} , m, q and r are as defined above.

11. A compound according to any preceding claim wherein R^1 is selected from Cl and SMe.

12. A compound according to any preceding claim wherein X is O.

13. A compound according to any one of claims 1 to 10 wherein X is $(CR^{11}R^{12})_p$, more preferably CH_2 .

14. A compound according to any preceding claim wherein:

R^2 is selected from H, methyl, ethyl and isopropyl; and

R^3 is selected from methyl, ethyl, isopropyl and piperidinyl, wherein the piperidinyl group is optionally substituted by one or more R^{15} substituents.

15. A compound according to any one of claims 1 to 3 wherein R^2 and R^3 are linked together with the nitrogen to which they are attached to form a 5- or 6-membered saturated heterocyclic group optionally substituted by one or more R^{16} groups.

16. A compound according to claim 15 wherein R^2 and R^3 are linked together with the nitrogen to which they are attached to form a pyrrolidinyl, piperidinyl or piperazinyl group, each of which is optionally substituted by one or more substituents selected from alkyl and $(CH_2)_sNR^{18}R^{19}$.

17. A compound according to any one of claims 1 to 13 wherein:

m is 1;

q is 1;

r is 1;

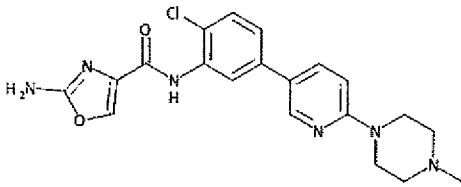
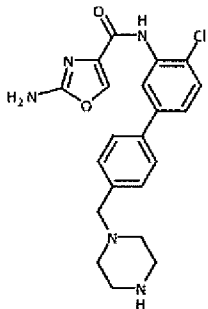
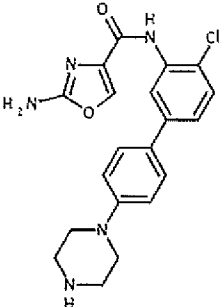
one of R^8 and R^9 is H or alkyl and the other is linked to R^3 to form a saturated heterocyclic group; and

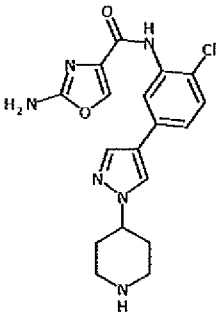
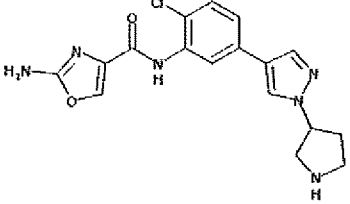
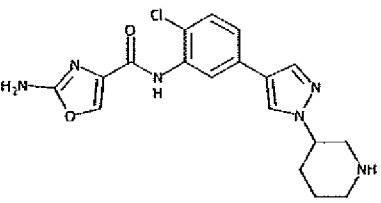
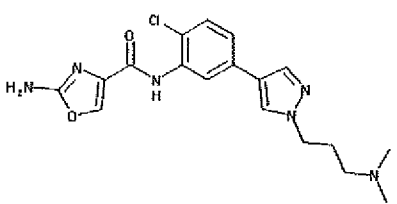
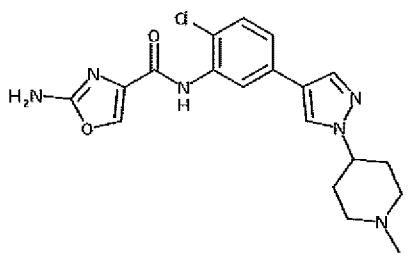
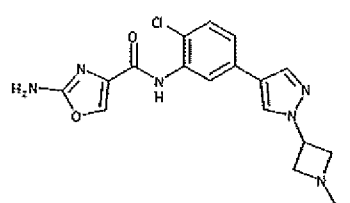
R^4 , R^5 , R^6 and R^7 are each independently H or alkyl, more preferably H.

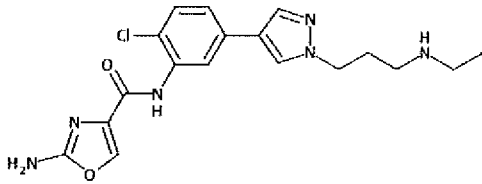
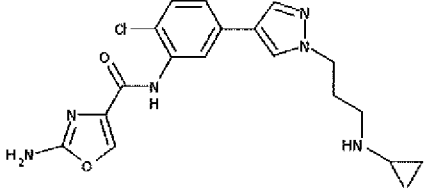
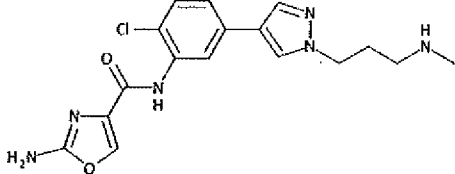
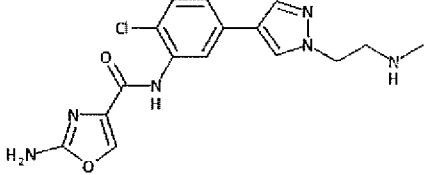
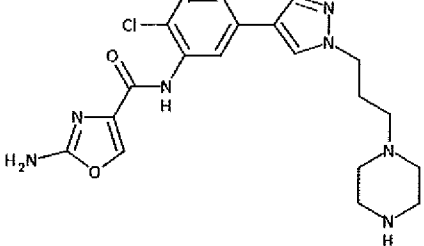
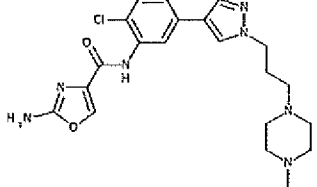
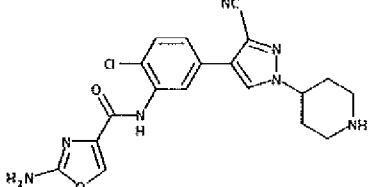
18. A compound according to claim 17 wherein one of R^8 and R^9 is H or alkyl and the other is linked to R^3 to form a piperidinyl group.

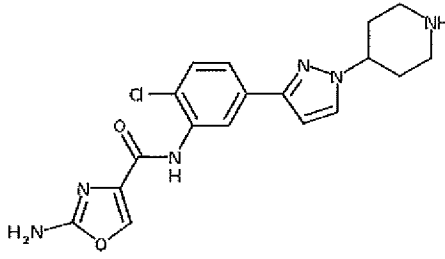
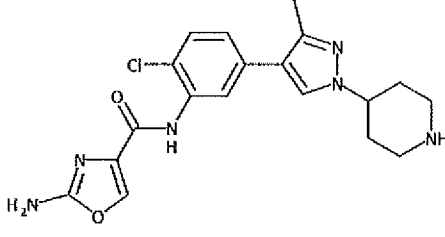
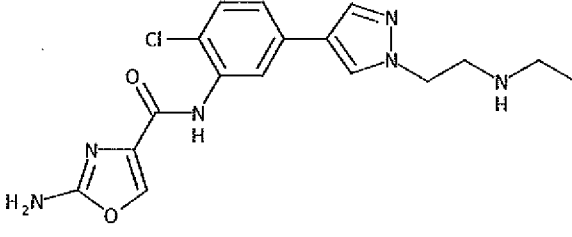
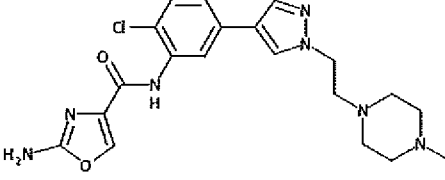
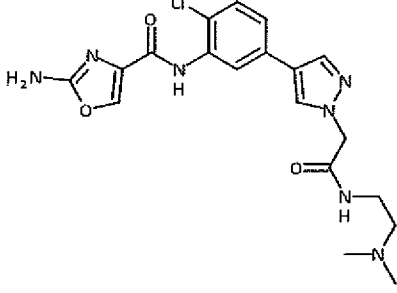
19. A compound according to claim 17 or claim 18 wherein X is $(CR^{11}R^{12})_p$ and p is 0.

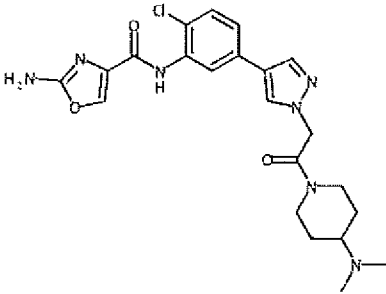
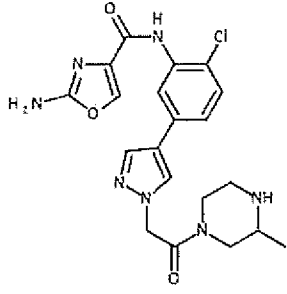
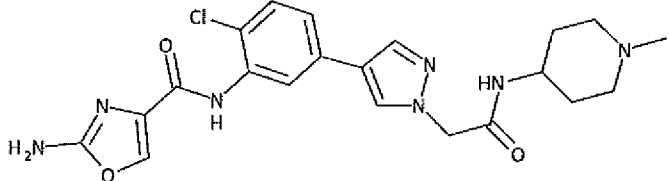
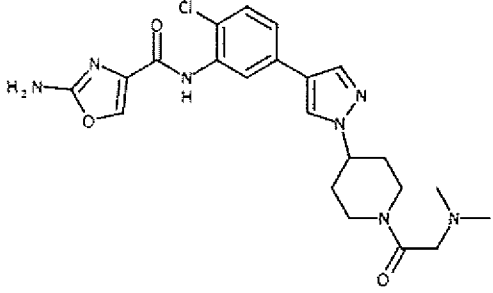
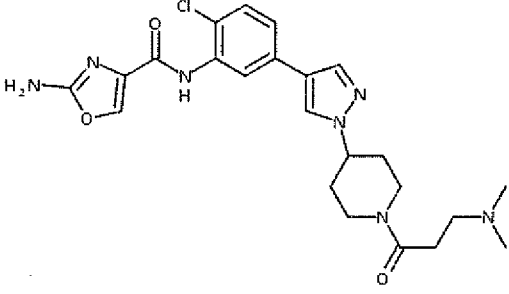
20. A compound according to any preceding claim which is selected from the following:

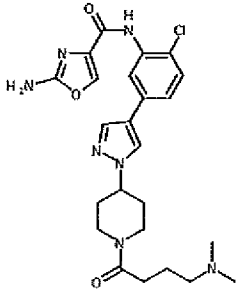
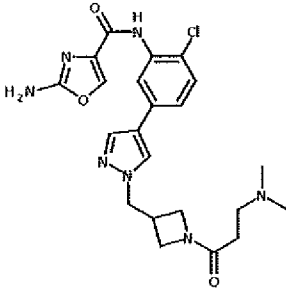
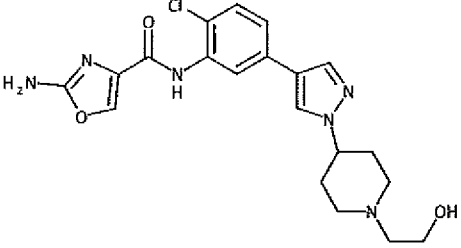
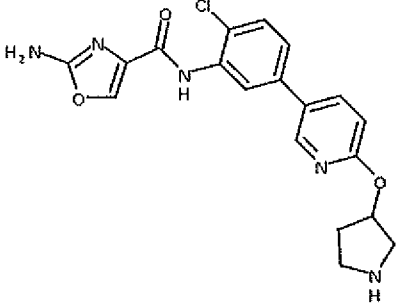
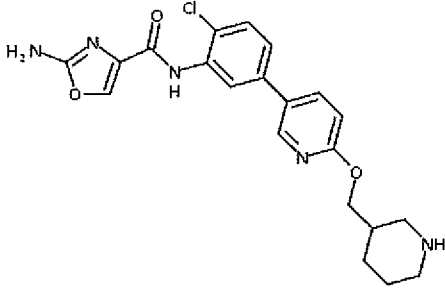
	37
	38
	39

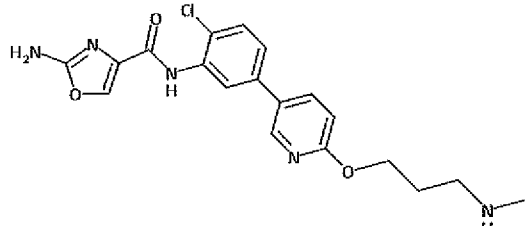
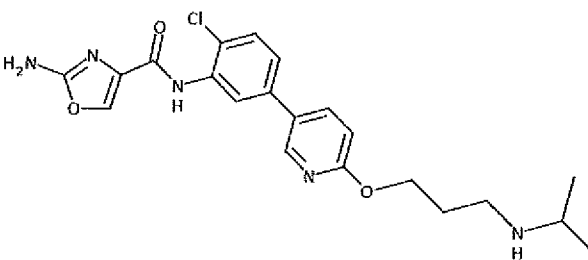
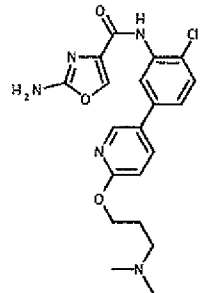
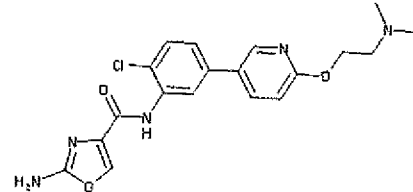
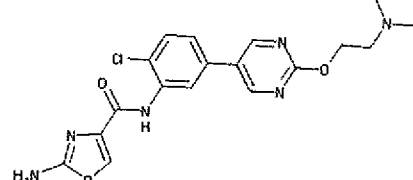
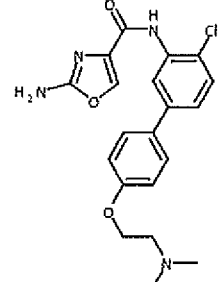
	<p>40</p>
	<p>41</p>
	<p>42</p>
	<p>43</p>
	<p>44</p>
	<p>45</p>

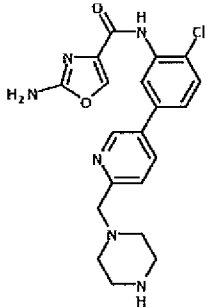
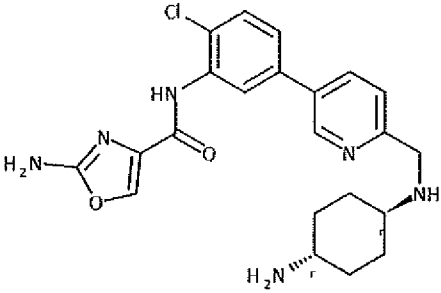
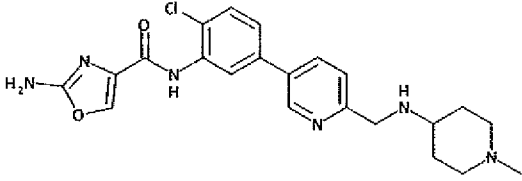
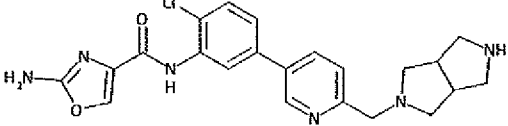
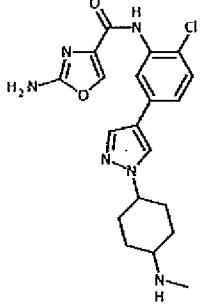
	<p>47</p>
	<p>48</p>
	<p>49</p>
	<p>50</p>
	<p>51</p>
	<p>52</p>
	<p>53</p>

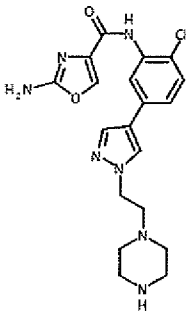
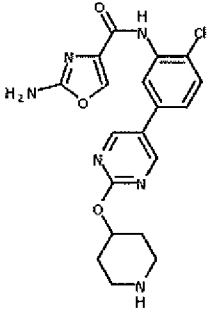
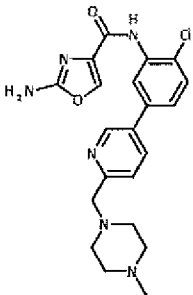
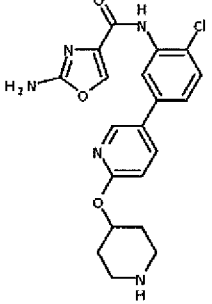
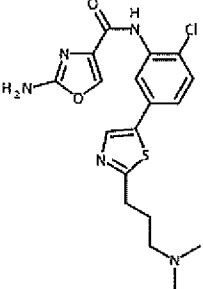
 <chem>Nc1nc(O)c(C(=O)Nc2ccc(Cl)cc2c3c[nH]1)nn3</chem>	54
 <chem>Cc1c[nH]1c2c[nH]1c3c[nH]2c3C(=O)Nc4ccc(Cl)cc4c5c[nH]1)nn5</chem>	55
 <chem>CCCNc1c[nH]1c2c[nH]1c3c[nH]2c3C(=O)Nc4ccc(Cl)cc4c5c[nH]1)nn5</chem>	56
 <chem>CN1CCNCC1c2c[nH]2c3c[nH]1c3C(=O)Nc4ccc(Cl)cc4c5c[nH]1)nn5</chem>	57
 <chem>CN(C)CC(=O)Nc1c[nH]1c2c[nH]1c3c[nH]2c3C(=O)Nc4ccc(Cl)cc4c5c[nH]1)nn5</chem>	58

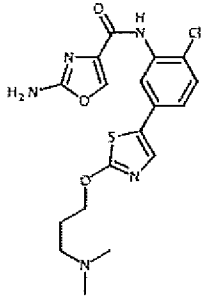
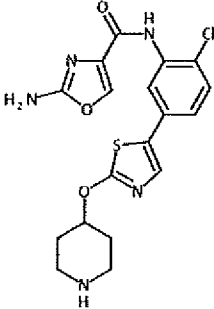
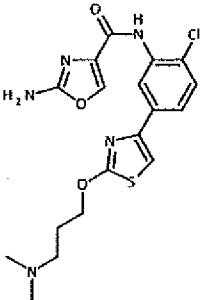
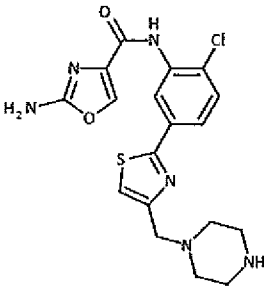
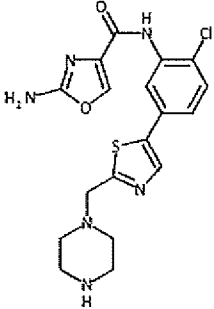
	<p>59</p>
	<p>60</p>
	<p>61</p>
	<p>62</p>
	<p>63</p>

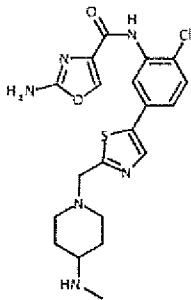
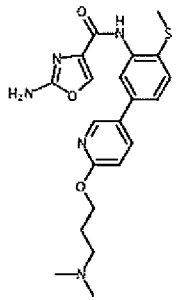
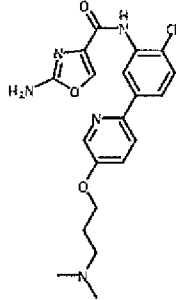
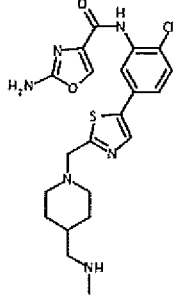
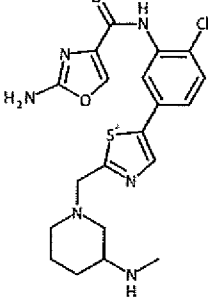
	<p>64</p>
	<p>65</p>
	<p>66</p>
	<p>68</p>
	<p>69</p>

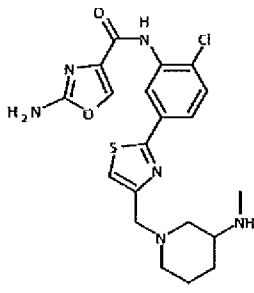
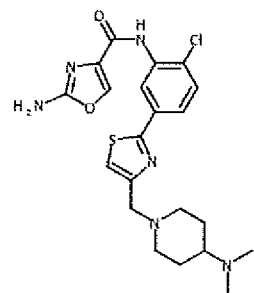
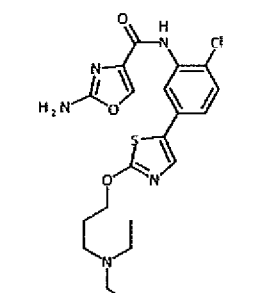
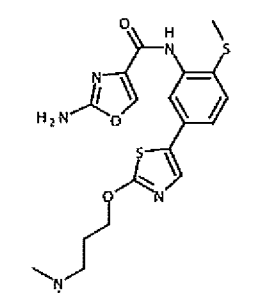
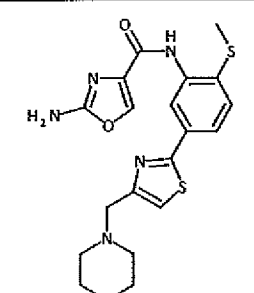
	<p>70</p>
	<p>71</p>
	<p>72</p>
	<p>73</p>
	<p>74</p>
	<p>75</p>

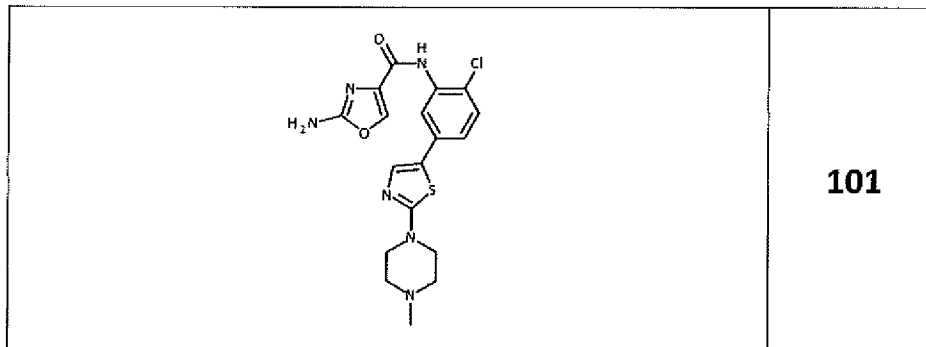
	<p>76</p>
	<p>77</p>
	<p>78</p>
	<p>79</p>
	<p>80</p>

	<p>81</p>
	<p>82</p>
	<p>83</p>
	<p>84</p>
	<p>85</p>

	<p>86</p>
	<p>87</p>
	<p>88</p>
	<p>89</p>
	<p>90</p>

	<p>91</p>
	<p>92</p>
	<p>93</p>
	<p>94</p>
	<p>95</p>

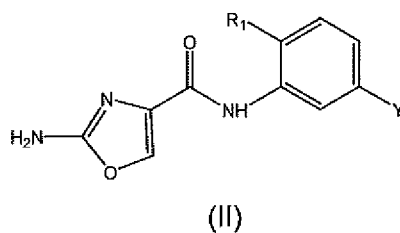
	96
	97
	98
	99
	100



and pharmaceutically acceptable salts and esters thereof.

21. A pharmaceutical composition comprising a compound according to any preceding claim and a pharmaceutically acceptable carrier, diluent or excipient.
22. A pharmaceutical composition according to claim 21 which further comprises a second therapeutic agent.
23. A compound according to any one of claims 1 to 20 for use in medicine.
24. A compound according to any one of claims 1 to 20 for use in treating a proliferative disorder, preferably cancer or leukemia.
25. Use of a compound according to any one of claims 1 to 20 in the preparation of a medicament for treating or preventing a proliferative disorder.
26. A method of treating a proliferative disorder in a subject in need thereof, said method comprising administering to the subject a therapeutically effective amount of a compound according to any one of claims 1 to 20.
27. Use of a compound according to any one of claims 1 to 20 in the preparation of a medicament for treating or preventing metastasis.
28. A method of treating or preventing metastasis in a subject in need thereof, said method comprising administering to the subject a therapeutically effective amount of a compound according to any one of claims 1 to 20.

29. A compound according to any one of claims 1 to 20 for use in preventing or treating a disorder caused by, associated with or accompanied by abnormal PAICS activity and/or abnormal PAICS expression..
30. A method of treating a subject having a disease state alleviated by the inhibition of PAICS, wherein the method comprises administering to the subject a therapeutically effective amount of a compound according to any one of claims 1 to 20.
31. Use of a compound according to any one of claims 1 to 20 in an assay for identifying further candidate compounds capable of inhibiting PAICS.
32. A combination comprising a compound according to any one of claims 1 to 20 and a further therapeutic agent.
33. A process for preparing a compound of formula (I) as defined in claim 1, said process comprising the steps of:
- (i) preparing an intermediate of formula (II):



- wherein R¹ is defined in claim 1 and Y is selected from halogen (more preferably I), boronic acid and a boronate ester; and
- (ii) converting said intermediate of formula (II) to a compound of formula (I).

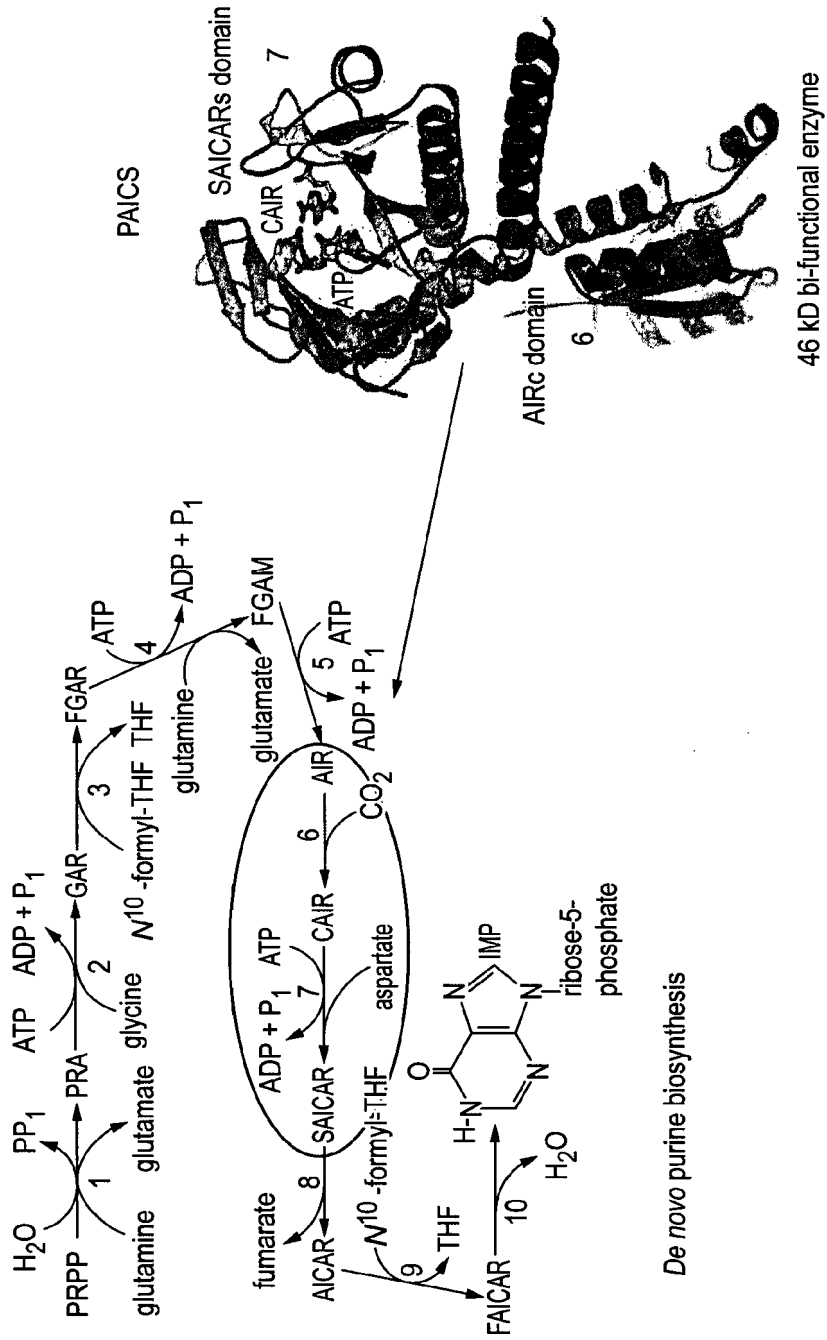


FIG. 1

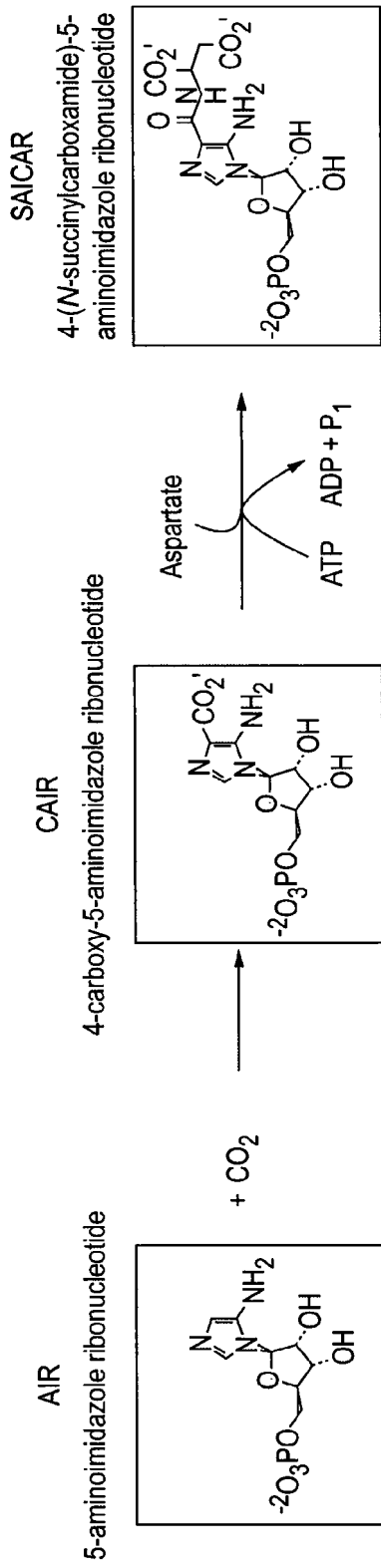


FIG. 2

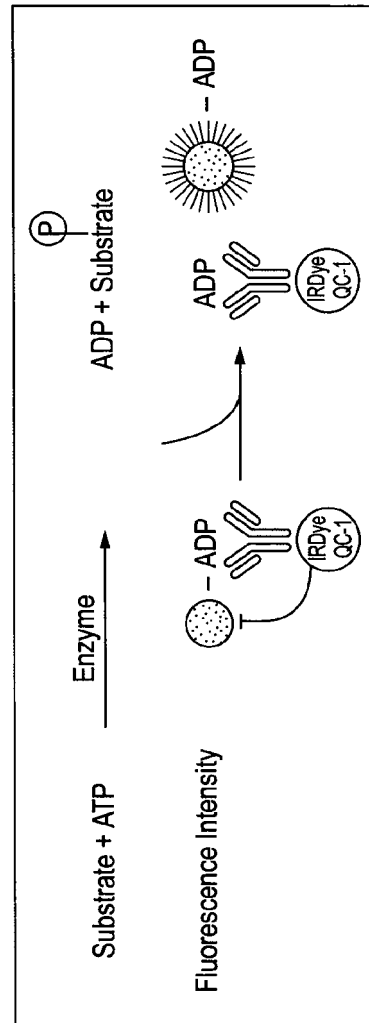


FIG. 3

INTERNATIONAL SEARCH REPORT

International application No
PCT/GB2017/053822

A. CLASSIFICATION OF SUBJECT MATTER		
INV. C07D413/14	C07D413/12	C07D417/12
C07D263/48	A61P35/00	A61K31/422
C07D417/14 C07D487/04		
ADD.		
According to International Patent Classification (IPC) or to both national classification and IPC		
B. FIELDS SEARCHED		
Minimum documentation searched (classification system followed by classification symbols) C07D A61P A61K		
Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched		
Electronic data base consulted during the international search (name of data base and, where practicable, search terms used) EPO-Internal, CHEM ABS Data, WPI Data		
C. DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	EP 2 009 005 A1 (ASTELLAS PHARMA INC [JP]) 31 December 2008 (2008-12-31) claims 1,10,11 -----	1-33
A	BALABHADRAPATRUNI V. S. K. CHAKRAVARTHI ET AL: "Expression and Role of PAICS, a De Novo Purine Biosynthetic Gene in Prostate Cancer : Purine Biosynthetic Gene in Prostate Cancer", PROSTATE., vol. 77, no. 1, 22 August 2016 (2016-08-22), pages 10-21, XP055448095, US ISSN: 0270-4137, DOI: 10.1002/pros.23243 page 14, right-hand column, line 1 - line 4 ----- -/--	1-33
<input checked="" type="checkbox"/> Further documents are listed in the continuation of Box C. <input checked="" type="checkbox"/> See patent family annex.		
* Special categories of cited documents :		
"A" document defining the general state of the art which is not considered to be of particular relevance "E" earlier application or patent but published on or after the international filing date "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified) "O" document referring to an oral disclosure, use, exhibition or other means "P" document published prior to the international filing date but later than the priority date claimed	"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art "&" document member of the same patent family	
Date of the actual completion of the international search	Date of mailing of the international search report	
7 February 2018	15/03/2018	
Name and mailing address of the ISA/ European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Authorized officer Johnson, Claire	

INTERNATIONAL SEARCH REPORT

International application No
PCT/GB2017/053822

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A,P	<p>Simon Osborne: "Fragment optimisation without co-crystal X-ray structures: from hits to nanomolar inhibitors of human PAICS", Fragments 2017 - 6th RSC-BMCS Fragmentbased Drug Discovery Meeting, 7 March 2017 (2017-03-07), XP055448256, Retrieved from the Internet: URL:http://www.maggichurchosevents.co.uk/bmcs/Downloads/Archive/Fragments%20-%20sborne%20Simon.pdf [retrieved on 2018-02-06] Slides 19, 32-35</p> <p style="text-align: center;">-----</p>	1-33
A,P	<p>"Sunday-Tuesday, 5th-7th March 2017 Second announcement and call for posters 6th RSC-BMCS Fragment-based Drug Discovery meeting Fragments 2017", 1 February 2017 (2017-02-01), XP055448262, Retrieved from the Internet: URL:http://www.rsc.org/events/detail/23352/fragments-2017-6th-rsc-bmcs-fragment-based-drug-discovery-meeting [retrieved on 2018-02-06] the whole document</p> <p style="text-align: center;">-----</p>	1-33

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No

PCT/GB2017/053822

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
EP 2009005	A1	31-12-2008	
		CA 2649043 A1	01-11-2007
		CN 101426774 A	06-05-2009
		EP 2009005 A1	31-12-2008
		JP 5182088 B2	10-04-2013
		JP W02007123269 A1	10-09-2009
		KR 20090004976 A	12-01-2009
		TW 200808788 A	16-02-2008
		US 2009286766 A1	19-11-2009
		WO 2007123269 A1	01-11-2007
