Final Project Report

Synthesis and Characterization of New Oxazole Containing Pharmacophores and Their Biological Evaluation as Antidiabetics

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BAHADUR SHAH ZAFAR MARG
NEW DELHI – 110 002.

Principal Investigator
Professor Shailesh R. Shah

Department Of Chemistry
Faculty of Science
The M S University of Baroda
Vadodara 390002
India

2017

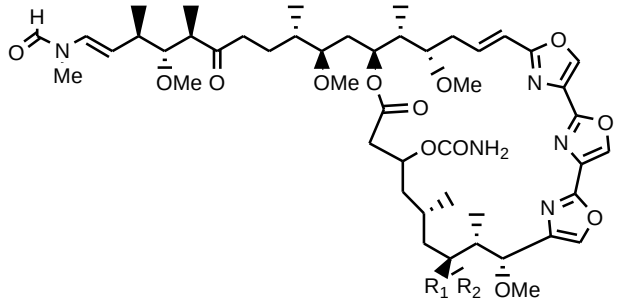
Summary of the project

Title : Synthesis and Characterization of New, Oxazole Containing Pharmacophores and Their Biological Evaluation as Antidiabetics.

Metabolic disorders and diabetes:

The medical disorders such as, hyperinsulinemia, hyperglycemia, dyslipidemia, high blood pressure, insulin resistance and obesity are considered to be causing due to metabolic disorders. They increase the risk of developing cardiovascular diseases and diabetes. In India the number of people affected by diabetes is increasing at an alarming rate and to an unbelievable number.

There are several approaches which are being persuaded for treatment of the disease. There are a number of heterocyclic compounds which are found effective in treatment of diabetes. Out of such new chemical compounds which are being persuaded for the exploration, the oxazole derivatives are of recent interest due to their presence in several natural products (e.g.**1**). This made us interested in exploring and undertaking synthesis and study of some new oxazole compounds mainly with chemistry point of view.



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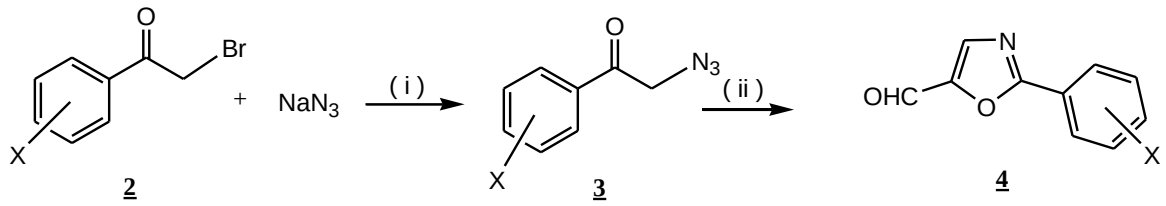
Oxazole with 2.5-di-substitution were prepared by using acyl azides in the presence of the Vilsmeier reagent. The formation of the said oxazoles under the conditions was unusual and investigated further for its mechanistic pathway. The mechanism of the reaction was proposed based on the evidences obtained in form of the intermediates isolated.

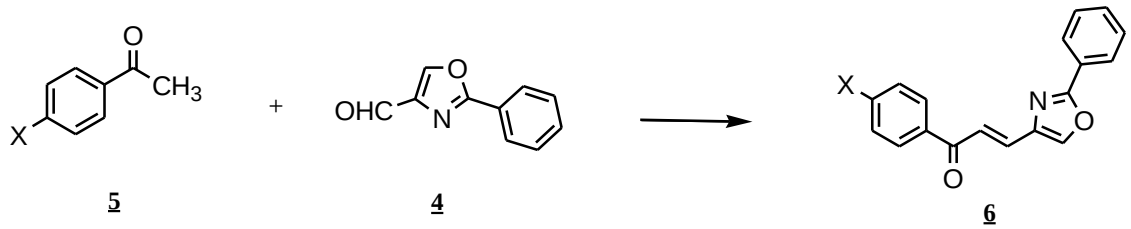


2 3 4

(i) DMF, 0-10 ºC (ii) $POCl_3$, 80-90 ºC

Scheme-1

Several new formylated oxazoles were thus prepared as shown in **Scheme 1** and were employed for the synthesis of several new chalcones as outlined in **Scheme 2.**



5 4 6

Scheme-2

The newly prepared oxazole containing chalcone compounds were subjected to their sugar lowering capacity in vivo. The results were showing that these compounds have moderate effect on the sugar levels. The most effective compound was found to e when X= -Br. The results are included in the following table.

Table-1: Compounds' (Compounds-6) Effect on Blood Glucose level:

Experimental Groups	Initial (mg/dl)	Final (mg/dl)	% difference
Normal Control	90.88 ± 6.52	88.01 ± 4.06	02.22
Diabetic Control	310.10 ± 5.16	300.25 ± 7.74	03.66
Compound a X= -H	420.70 ± 2.02	314.63 ± 2.76	26.00
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Compound d X= $-CH_3$	250.12 ± 4.83	210.14 ± 4.59	16.00
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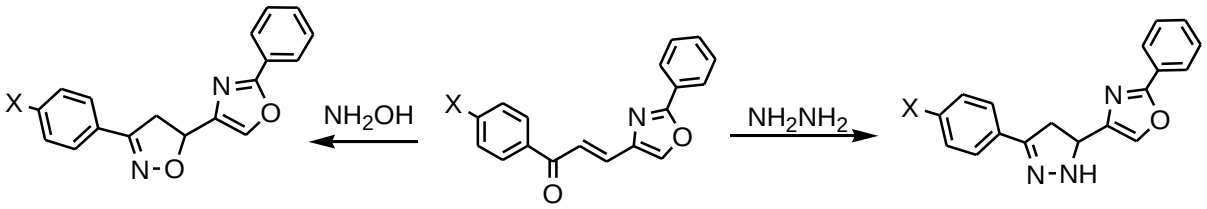


7 **6** **8**

Scheme-3

The final compounds were also subjected to antidiabetic activity, anti-inflammatory activity and anticancer activity studies. Anticancer activity was done *in vitro* at the National Cancer Institute USA while antifungal activity was kindly carried out by Zydus Research Centre Ahmedabad in a collaborative work. The compounds showed good anti-inflammatory activity. All the bio activity results including the anticancer activity results for different cell lines are included in the final detailed report.

Final Project Report

Title of the Project:

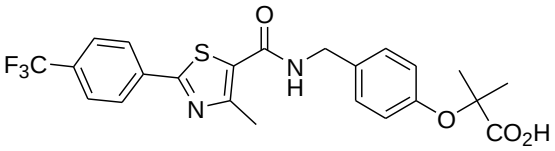
Synthesis and Characterization of New, Oxazole Containing Pharmacophores and Their Biological Evaluation as Antidiabetics.

Introduction:

Metabolic disorders and diabetes:

Metabolic syndrome causes the medical disorders such as, hyperinsulinemia, hyperglycemia, dyslipidemia, high blood pressure, insulin resistance and obesity. These disorders increase the risk of developing cardiovascular diseases and diabetes. In India the number of people affected by diabetes is increasing at an alarming rate and to an unbelievable number.

There are several approaches which are being persuaded for treatment of the disease. They target various enzymes such as Peroxisome Proliferator Activated Receptors (PPAR), Dipeptidyl Peptidase IV (DPP-IV), Glycogen synthase kinase-3 (GSK-3), Protein Tyrosine Phosphatises 1B (PTP-1B) or G protein-coupled receptor 119 (GPR 119) which are involved in regulating blood sugar. There are a number of heterocyclic compounds which are found effective in reducing of glucose level in diabetic animals. Some of such compounds are projected in **Figure-1.**



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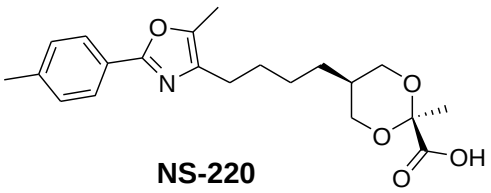

NS-220

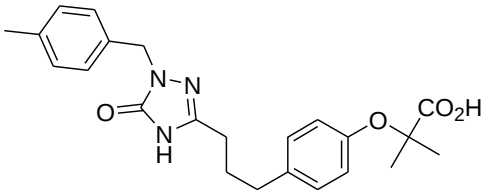

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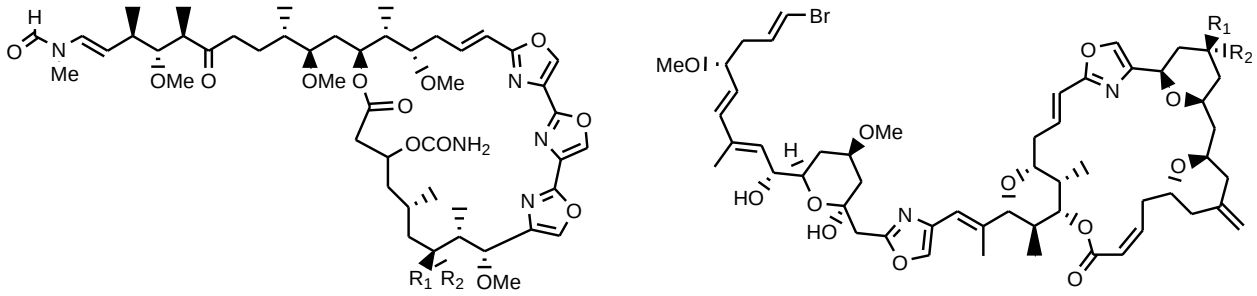
Figure-1 Some compounds with heterocycles having promising antidiabetic activity.

In addition to antidiabetic activity, oxazole heterocycle is associated with various other biological activities such as anticancer[1], antidiabetic[2], antiviral[3], antiinflammatory[4] antibiotic[5] activities.

Apart from their medicinal value investigation, the oxazole derivatives are of recent interest due to their presence in several natural products isolated from marine sources (**Figure-2**)[6]



1 **2**

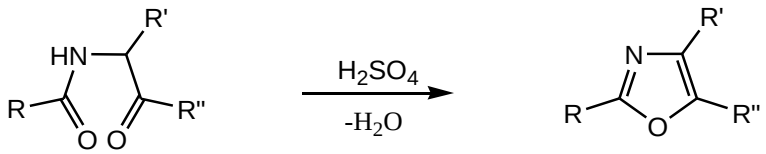
Figure-2. Some naturally occurring oxazole compounds.

In the pursuit of the exploration of new oxazoles for their investigation for their biological activity, the oxazole containing compounds required to be prepared for which the presently reported methods were studied.

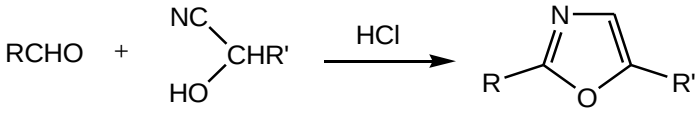
Methods of Synthesis of Oxazoles:

There are several methods for the synthesis of oxazoles as summarized below.

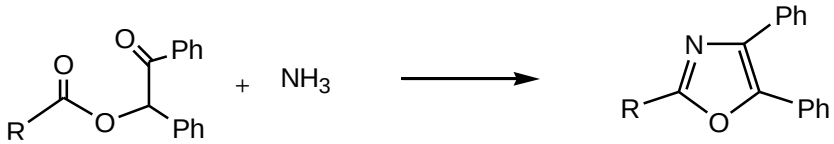
1) α-Acyl-amino ketones can be cyclised to oxazoles using sulphuric acid.[7]



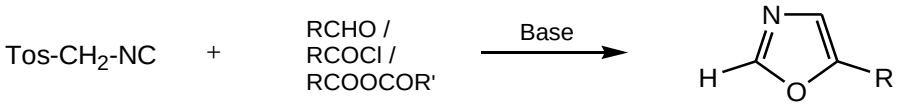
2) The reaction of cyanohydrins with aldehydes in the presence of acid leads to 2,5-di substituted oxazoles.[8]



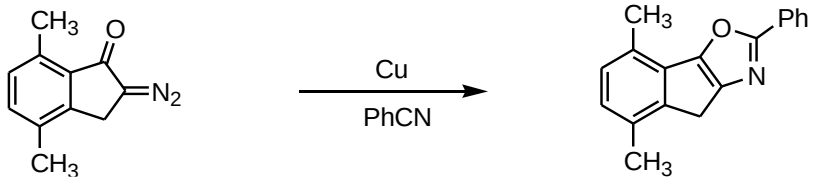
3) The reaction of esters from benzoin on reaction with ammonia also gives aryl substituted oxazoles.[9]



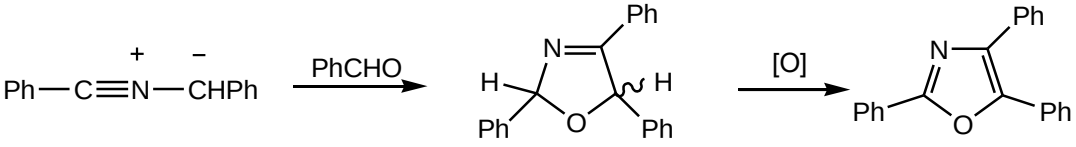
4) Tosylmethylisocyanide (TosMIC) is conveniently employed for the synthesis of 4,5-substituted oxazoles by reacting them with aldehydes, acid chlorides or anhydrides after treating them with base.[10]



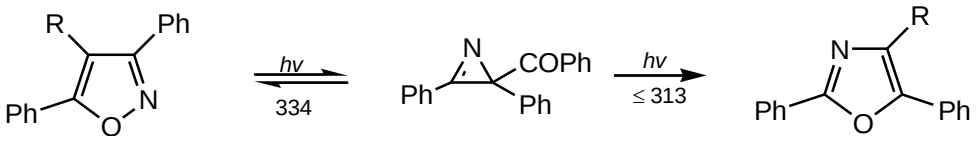
5) Carbonyl carbenes generated from α-diazoketones undergo 1,3-dipolar cycloaddition reaction with nitriles giving moderate yields of oxazoles.[11]



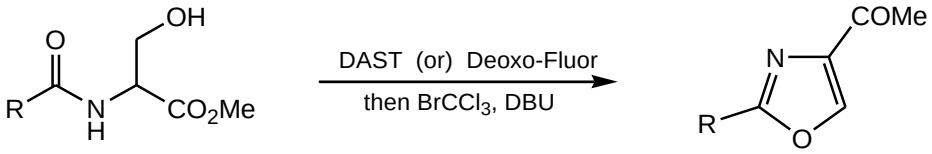
6) Nitrile ylide also can undergo dipolar cycloadditions to aldehydes give oxazolines which can be oxidized to oxazoles[12], as for example:



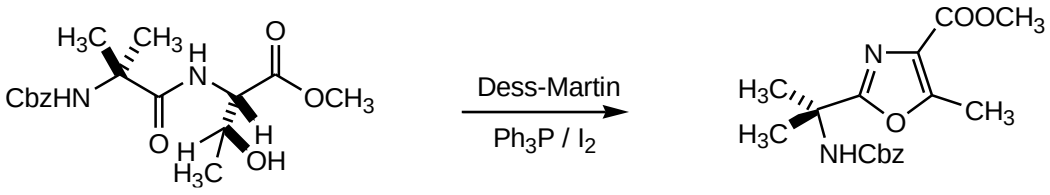
7) Isoxazoles or azirines on photolysis at less than 313 nm are converted to oxazoles, at higher wave length (≥ 334 nm) azirine is in equilibrium with isoxazole.[13]



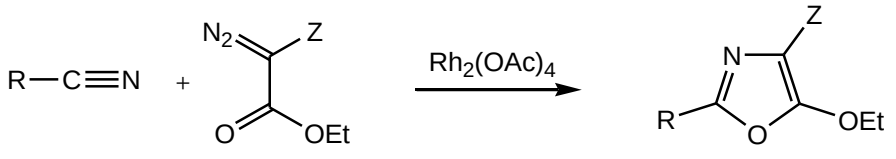
8) A mild and highly efficient cyclization of highly functionalized β-hydroxy amides to oxazolines and their one pot oxidation leads to oxazoles.[14]



9) A synthesis of functionalized oxazoles and bis oxazoles has been reported by side chain oxidation of β-hydroxy amides with the Dess-Martin periodinane followed by cyclodehydration with triphenyl phosphene / iodine in the presence of triethyl amine.[15]

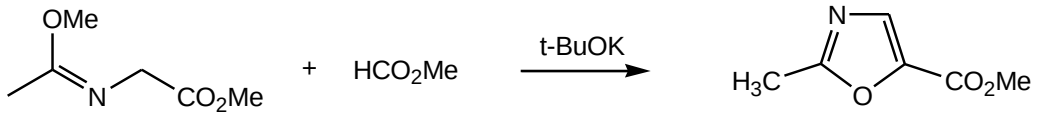


10) Diazo dicarbonyl compounds in a rhodium catalyzed reaction with nitriles, provides useful, functionalized oxazoles in a direct one step synthesis.[16]

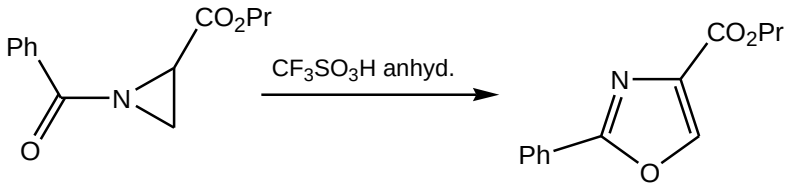


Z=SO_2Ph, $PO(OEt)_2$, CN or substituted oxazole

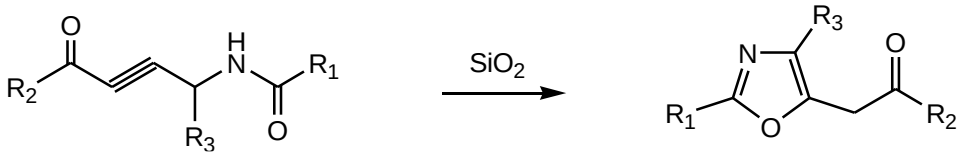
11) 2-Substituted-1,3-oxazole-4-carboxylates can be prepared using alkylidene amino acetate and methyl formate.[17]



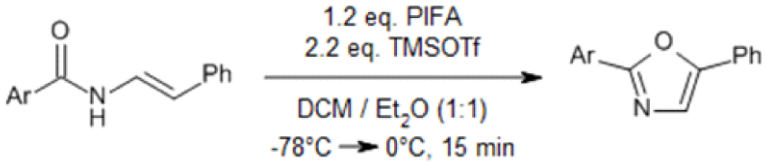
12) N-Acyl aziridines have been ring expanded to disubstituted oxazolines using triflouro methane sulphonic acid.[18]



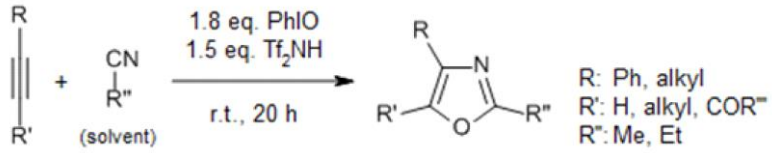
13) A mild silicagel mediated cycloisomerization of propargyl amides has been reported giving 2,5-disubstituted and 2,4,5-trisubstituted oxazol-5-yl carbonyl compounds.[19]



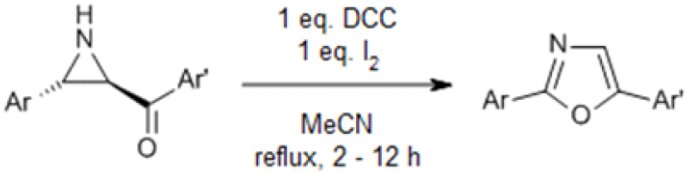
14) N-Styryl bezamides on cyclization with hypervalent iodine reagent leads to 2,5-disubsituted oxazoles.[20]



15) 2,4-Disubstituted or 2,4,5-trisubstituted oxazoles were prepared by reacting alkynes with nitriles in presence of an oxygen source.[21]



16) Acyl aziridines can be converted to 2,5-disubstituted oxazoles in the presence of DCC and iodine.[22]



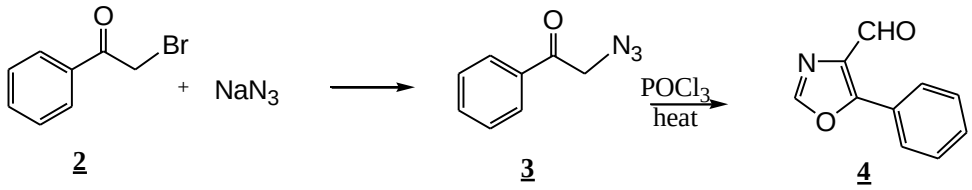
Advancements in the chemistry through the Present Project.

With the aim to design new bioactive compounds with the help of incorporation of various bioactive heterocycles one of which include 1,3-oxazole which has been reported to have a good activity against various microorganisms and also having possibility of anticancer activity. The various approaches were studied for the construction of functionalized oxazoles as starting point for the construction of the new molecules with a good biological activity.

Synthesis of New Oxazole Derivatives.

Out of the known methods as reported above, we looked for functionalized oxazoles which further can be reacted to incorporate additional substitutions including induction of new heterocylic moieties. One of the methods which attracted our attention was the construction of oxazoles and imidazoles under the Vilsmeier condition.[23] This method involves the synthesis of the heterocycles starting from aryl acyl azides which may be prepared in two steps. The synthesis results in aryl-oxazoles with the reactive formyl functionality on the heterocycle.

On exploring the reaction as reported in the literature, the oxazole compounds formed should have structure as shown in **Scheme-1.**[23]



2 **3** **4**

Scheme-1

The X-ray structure **(Figure-3)** observed for one of the oxazole aldehyde showed the oxazoles formed in fact had different structure than the reported one and that the reaction was leading to the 2-aryl-5-formyl oxazoles rather than the reported earlier.

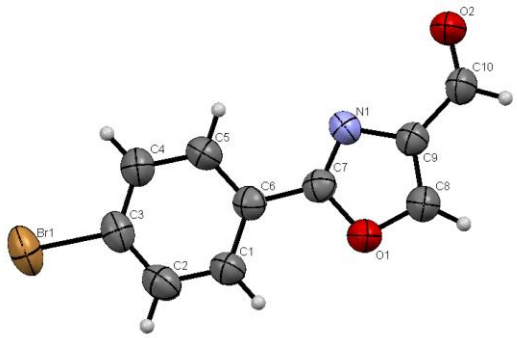
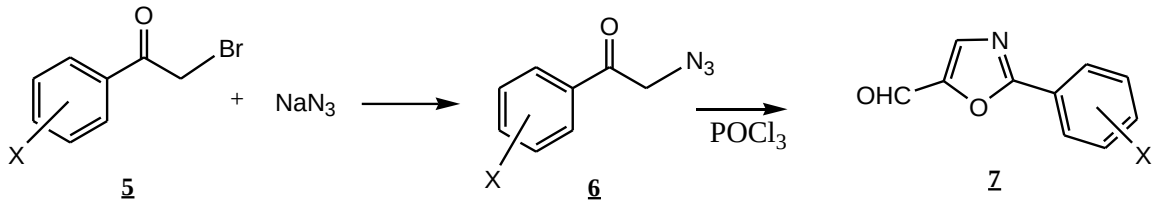


Figure-3 ORTEP diagram of 2-(4-bromophenyl)-4-formyl oxazole

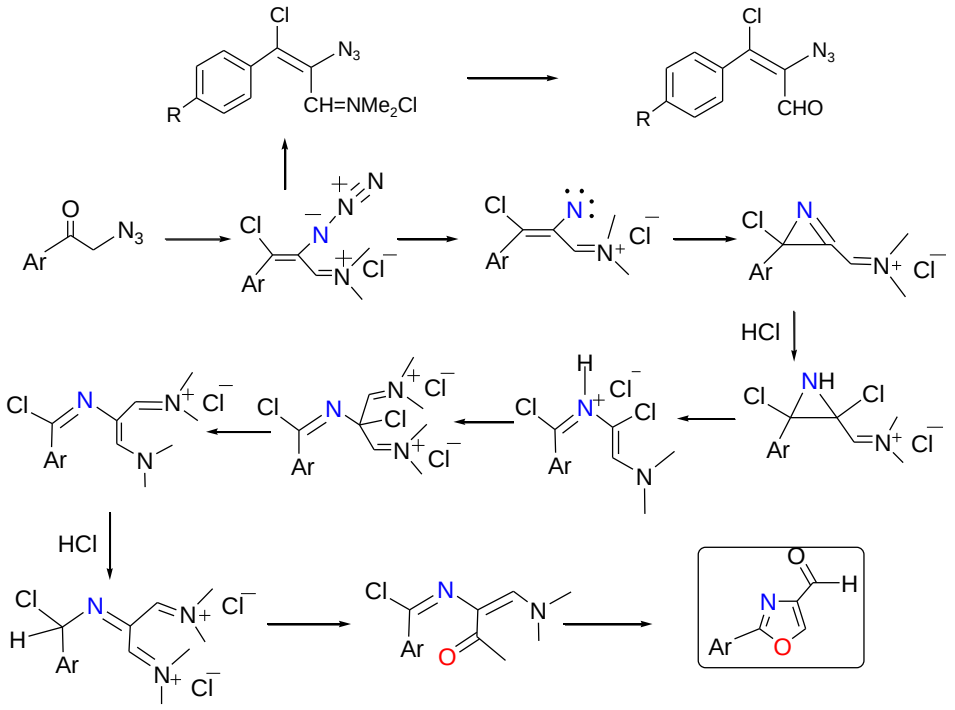
The formation of the said oxazoles under the conditions was unusual and investigated further for its mechanistic pathway.

The mechanism of the reaction was proposed based on the evidences obtained in form of the intermediates isolated. **Scheme-2**.



Scheme-2

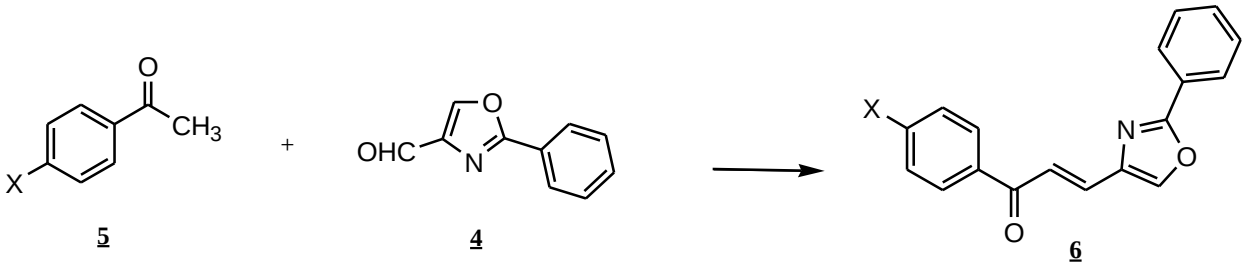
The mechanism of the unusual formation of 2-aryl-4-formyl oxazole was proposed in the following **Scheme -3.**[24]



Scheme-3 Proposed mechanism of the formation of the 4-formyl oxazole ring involving a rearrangement.

Several new formylated 2-aryl oxazoles were prepared as shown in **Scheme 2** and were employed for the synthesis of several new chalcones as outlined in **Scheme 4.** In the process of

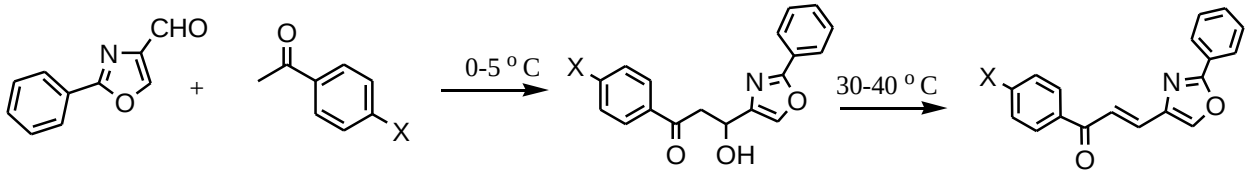
the synthesis of chalcones, the primary b-hydroxy ketones were also isolated by a careful experimentation as depicted in the following Scheme, **Scheme-5**.



5 **4** **6**

Scheme-4

Synthesis of β-hydroxy- ketones and oxazole chalcones.



4 **5** **6** **7**

a:- X= H, **b**:- Cl, **c**:- Br, **d**:- CH_3, **e**:- OCH_3

Scheme-5

The newly prepared oxazole containing β-hydroxy ketones **6** were subjected to their sugar lowering capacity *in vivo*. The results were showing that these compounds have moderate effect on the sugar levels. The most effective compound was found to be when X= -Br. The results are included in the following table.

Table-1: Compounds' (Compounds-6) Effect on Blood Glucose level:

Experimental Groups	Initial (mg/dl)	Final (mg/dl)	% difference
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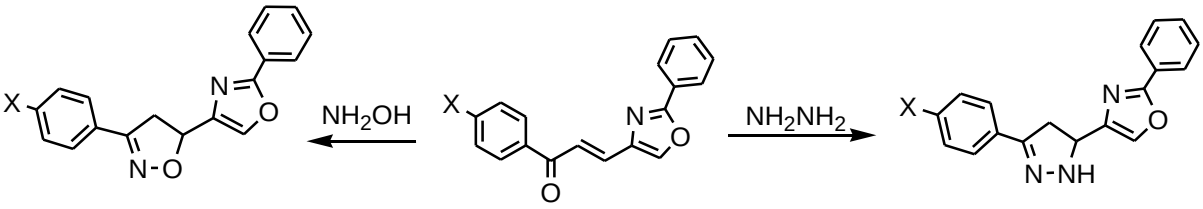
Compound e X= -OCH_3	246.71 ± 7.66	209.37 ± 6.31	15.01

The compounds were also subjected for their sugar lowering effect in blood serum.

Evolving of other heterocyclicic compounds from the chalcones from the oxazole aldehydes.

The chalcones thus prepared were employed for buiding of the other azole rings without affecting the 1,3-oxazole present in them. Thus, the chalcones were reacted with hydroxyl amine hydrochloride leading to the substituted 1,2-oxazoline heterocycle as shown in the following scheme, **Scheme-6**..



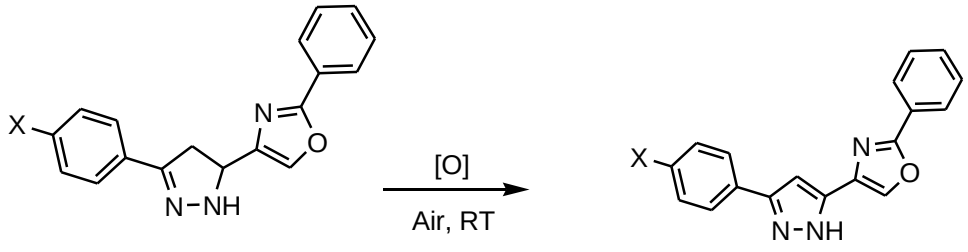
8 **7** **9**

Scheme-6

Series of the new compounds were prepared, purified and characterized using IR, NMR, Mass and elemental analysis.

The newly prepared compounds were subjected to antidiabetic activity and anticancer activity studies. Anticancer activity was performed *in vitro* at the National Cancer Institute USA.

The pyrazolines-**9** were oxidized to stable pyazoles **10** by aerial oxidation as shown in the following scheme **Scheme-7**.



9 **10**

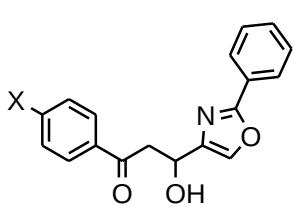
a:- X= H, **b**:- X=Cl, **c**:- X=Br, **d**:- X=CH_3, **e**:- X=OCH_3

Scheme-7

The compounds with two heterocyclic moieties compounds **8** and compounds**10** were evaluated for their **anti-inflammatory activity**. The results are included past anticancer activity study results.

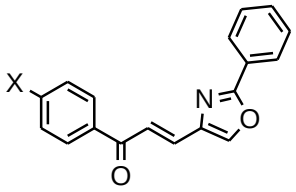
Anticancer activity:

Out of all the new compounds offered for the anticancer activity after signing an agreement with NCI five of the representative compounds **6 b**,**d** and **7 b**,**d**,**e** were selected for the preliminary anticancer activity study by the NCI, USA. The compounds have been screened at a single dose of 10 μM concentration against panel of all the 60 human cancer cell lines derived from nine different types of cancers: Leukemia, Non-Small cell lung cancer, Colon cancer, CNS cancer, Melanoma cancer, Ovarian cancer, Renal cancer, Prostate cancer and Breast cancer. The results which are made available to us have been discussed bellow.



6

b :- X=Cl
d :- X=Me



7

b :- X=Cl
d :- X=Me
e :- X=OMe

Figure-4

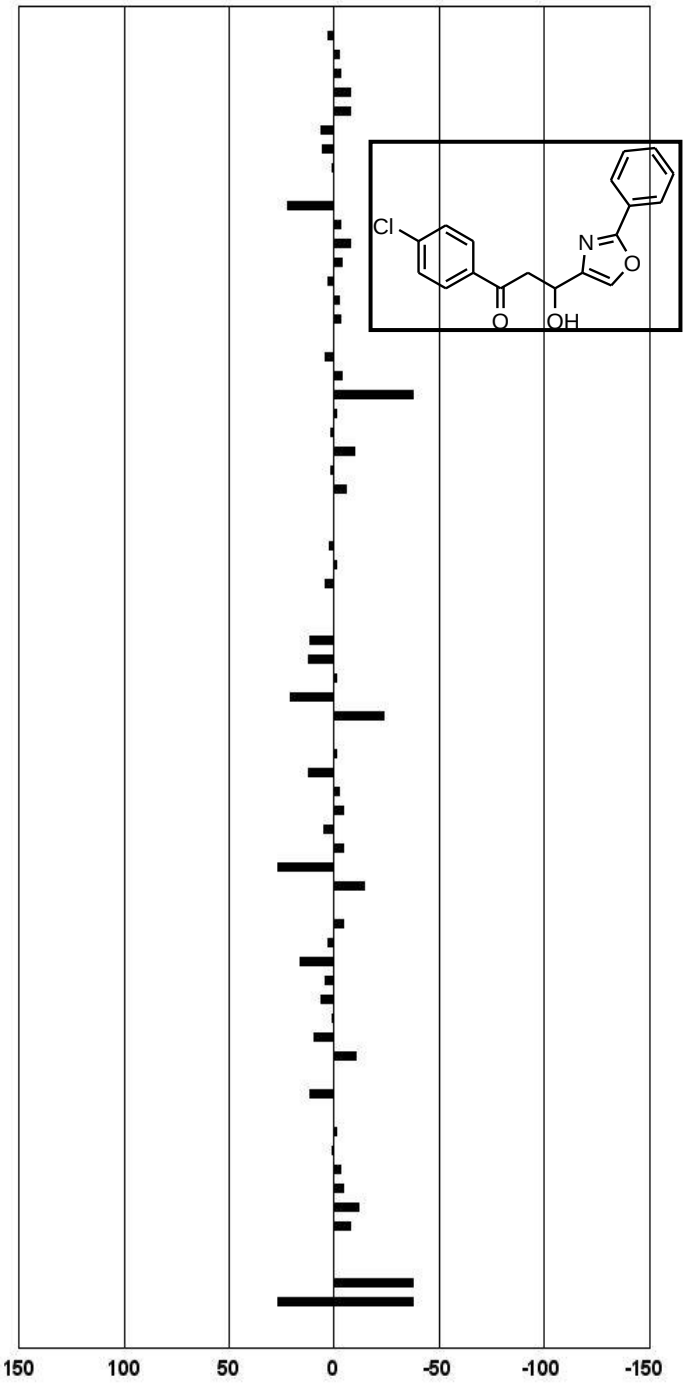
The β-hydroxy ketones **6b**,**d** were screened one having 4-methyl phenyl substitution **6d** and the other with 4-chloro phenyl substitution **6b** which representate the presence of electron reach phenyl ring and electron deficient phenyl ring on oxazole containing β-hydroxy ketones **6** respectively. Both of them are found to be ineffective in checking cancerous cell growth. The three α,β-unsaturated ketones **7b**,**d**,**e** (heterocyclic chalcones) have been screened for their anticancer activity. All the three compounds were found to have good activity against leukemia RPMI-8226 cell lines. The compound with 4-chloro phenyl substitution **7b** showed maximum growth inhibition of 86% for the cell line. All the three chalcones derivatives exhibit activity by showing more or less inhibition on different kinds of breast cancer cell lines, the maximum activity of 73% inhibition is observed for 4-chloro phenyl derivative **7b** against MCF7 breast cancer cell lines. All the three compounds are found to promote the growth of TK-10 renal cancer cell line, 4-methyl phenyl **7d** derivative is promoting the growth up to 290%.

% growth inhibition greater than 25% against multiple cell lines is shown in graphical form after the activity results included as received from NCI.

Developmental Therapeutics Program	NSC: 748002 / 1	Conc: 1.00E-5 Molar	Test Date: Aug 18, 2008
One Dose Mean Graph	Experiment ID: 0808OS98		Report Date: Sep 17, 2008

Panel/Cell Line	Growth Percent
Non-Small Cell Lung Cancer	
A549/ATCC	103.48
EKVX	98.31
HOP-62	97.49
NCI-H226	92.91
NCI-H23	92.66
NCI-H322M	106.55
NCI-H460	105.80
NCI-H522	101.08
Colon Cancer	
COLO 205	122.45
HCC-2998	97.35
HCT-116	92.37
HCT-15	96.41
HT29	103.26
KM12	97.77
SW-620	97.60
Breast Cancer	
BT-549	104.91
HS 578T	96.38
MCF7	63.13
MDA-MB-231/ATCC	99.35
MDA-MB-435	101.79
MDA-MB-468	90.90
NCI/ADR-RES	101.74
T-47D	94.48
Ovarian Cancer	
OVCAR-3	100.79
OVCAR-4	102.84
OVCAR-5	99.64
OVCAR-8	104.79
SK-OV-3	100.45
Leukemia	
CCRF-CEM	112.18
HL-60(TB)	112.32
K-562	99.55
MOLT-4	121.38
RPMI-8226	76.61
Renal Cancer	
786-0	99.64
A498	112.55
ACHN	98.14
CAKI-1	96.28
RXF 393	105.56
SN12C	96.29
TK-10	126.88
UO-31	85.89
Melanoma	
LOX IMVI	96.08
M14	102.99
MALME-3M	116.65
SK-MEL-2	104.49
SK-MEL-28	106.76
SK-MEL-5	101.12
UACC-257	109.86
UACC-62	90.14
Prostate Cancer	
DU-145	111.95
CNS Cancer	
SF-268	99.41
SF-295	101.16
SF-539	97.42
SNB-19	96.04
SNB-75	88.66
U251	92.95
Mean	100.46
Delta	37.33
Range	63.75

Mean Growth Percent - Growth Percent



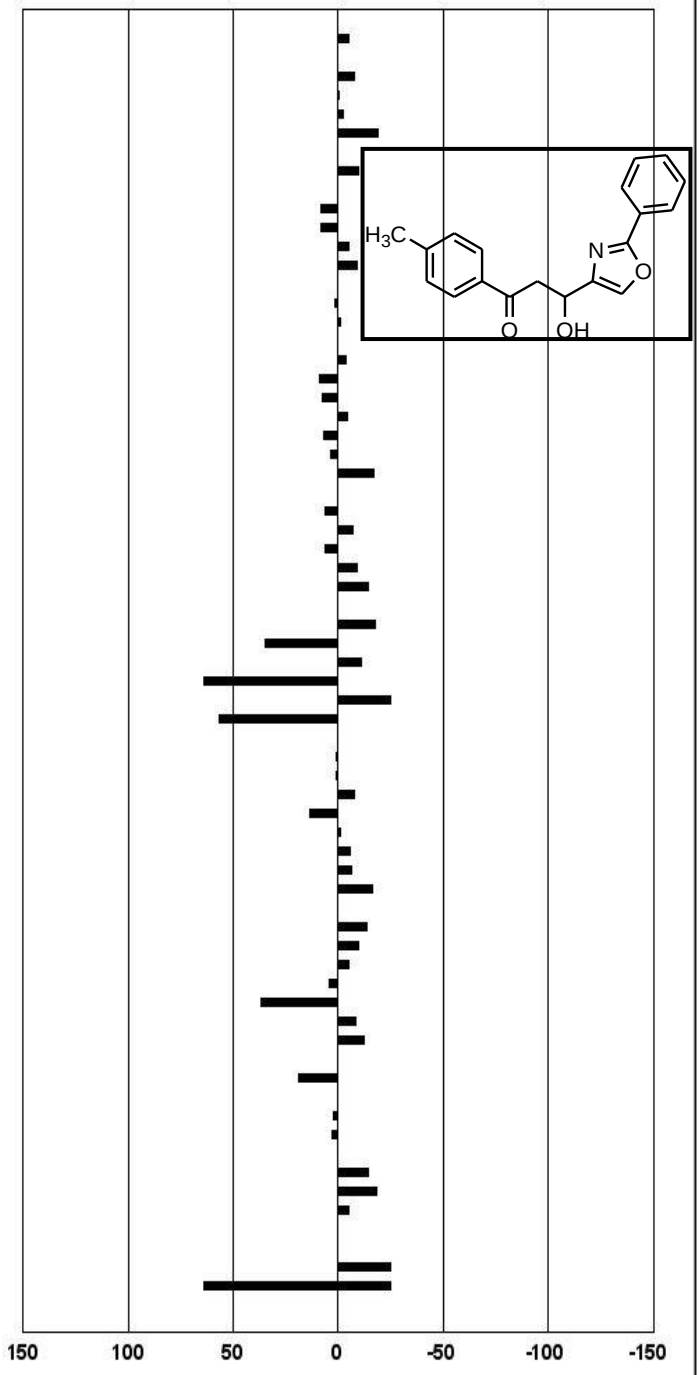
Developmental Therapeutics Program

One Dose Mean Graph

NSC: 748003 / 1	Conc: 1.00E-5 Molar	Test Date: Aug 18, 2008
Experiment ID: 0808OS98		Report Date: Sep 17, 2008

Panel/Cell Line	Growth Percent
Non-Small Cell Lung Cancer	
A549/ATCC	104.92
EKVX	110.56
HOP-62	102.21
NCI-H226	109.86
NCI-H23	107.48
NCI-H322M	91.32
NCI-H460	110.35
NCI-H522	100.44
Colon Cancer	
COLO 205	118.30
HCC-2998	118.30
HCT-116	104.88
HCT-15	100.80
HT29	110.12
KM12	111.93
SW-620	109.09
Breast Cancer	
HS 578T	106.57
MCF7	119.15
MDA-MB-231/ATCC	117.81
MDA-MB-435	105.45
MDA-MB-468	117.14
NCI/ADR-RES	113.39
T-47D	93.27
Ovarian Cancer	
OVCAR-3	116.49
OVCAR-4	102.81
OVCAR-5	116.53
OVCAR-8	100.80
SK-OV-3	95.84
Leukemia	
CCRF-CEM	92.57
HL-60(TB)	144.61
K-562	99.26
MOLT-4	173.79
RPMI-8226	84.87
SR	166.53
Renal Cancer	
786-0	110.80
A498	110.77
ACHN	102.51
CAKI-1	123.71
RXF 393	108.71
SN12C	104.54
TK-10	103.55
UO-31	93.93
Melanoma	
LOX IMVI	96.11
M14	100.40
SK-MEL-2	105.28
SK-MEL-28	114.38
SK-MEL-5	146.58
UACC-257	101.76
UACC-62	97.70
Prostate Cancer	
DU-145	128.92
CNS Cancer	
SF-268	112.48
SF-295	112.88
SF-539	110.20
SNB-19	95.89
SNB-75	92.04
U251	105.09
Mean	110.10
Delta	25.23
Range	88.92

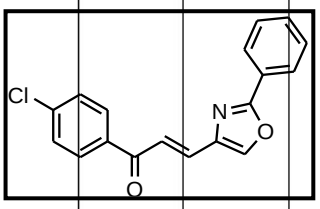
Mean Growth Percent - Growth Percent



Developmental Therapeutics Program	NSC: 748004 / 1	Conc: 1.00E-5 Molar	Test Date: Aug 18, 2008
One Dose Mean Graph	Experiment ID: 0808OS98		Report Date: Sep 17, 2008

Panel/Cell Line	Growth Percent
Non-Small Cell Lung Cancer	
A549/ATCC	96.05
EKVX	96.06
HOP-62	86.87
NCI-H226	90.91
NCI-H23	102.40
NCI-H322M	90.35
NCI-H460	98.01
NCI-H522	84.57
Colon Cancer	
COLO 205	109.82
HCC-2998	104.88
HCT-116	89.79
HCT-15	95.35
HT29	105.13
KM12	84.56
SW-620	90.40
Breast Cancer	
HS 578T	97.11
MCF7	26.49
MDA-MB-231/ATCC	95.73
MDA-MB-435	98.33
MDA-MB-468	48.49
NCI/ADR-RES	102.99
T-47D	45.32
Ovarian Cancer	
OVCAR-3	88.70
OVCAR-4	94.62
OVCAR-5	100.82
OVCAR-8	90.67
SK-OV-3	88.96
Leukemia	
CCRF-CEM	79.11
HL-60(TB)	89.58
K-562	99.12
MOLT-4	106.85
RPMI-8226	14.08
SR	47.10
Renal Cancer	
786-0	99.04
A498	115.16
ACHN	98.66
CAKI-1	89.89
RXF 393	95.43
SN12C	97.66
TK-10	149.49
UO-31	73.83
Melanoma	
LOX IMVI	81.84
M14	100.50
MALME-3M	73.51
SK-MEL-2	92.21
SK-MEL-28	112.75
SK-MEL-5	102.72
UACC-257	100.85
UACC-62	89.64
Prostate Cancer	
DU-145	100.28
CNS Cancer	
SF-268	100.30
SF-295	77.44
SF-539	93.63
SNB-19	81.69
SNB-75	90.14
U251	79.51
Mean	89.92
Delta	75.84
Range	135.41

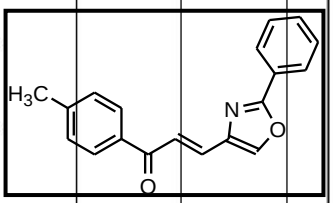
Mean Growth Percent - Growth Percent



Developmental Therapeutics Program	NSC: 748006 / 1	Conc: 1.00E-5 Molar	Test Date: Aug 18, 2008
One Dose Mean Graph	Experiment ID: 0808OS98		Report Date: Sep 17, 2008

Panel/Cell Line	Growth Percent
Non-Small Cell Lung Cancer	
A549/ATCC	97.82
EKVX	92.21
HOP-62	89.39
NCI-H226	86.13
NCI-H23	88.17
NCI-H322M	103.62
NCI-H460	101.16
NCI-H522	86.00
Colon Cancer	
COLO 205	102.74
HCC-2998	106.45
HCT-116	75.49
HCT-15	96.16
HT29	105.01
KM12	99.49
SW-620	96.49
Breast Cancer	
HS 578T	112.60
MCF7	38.63
MDA-MB-231/ATCC	98.64
MDA-MB-435	107.35
MDA-MB-468	61.18
NCI/ADR-RES	103.22
T-47D	42.57
Ovarian Cancer	
OVCAR-3	100.61
OVCAR-4	82.39
OVCAR-5	105.79
OVCAR-8	96.39
SK-OV-3	79.50
Leukemia	
CCRF-CEM	94.70
HL-60(TB)	98.04
K-562	110.60
MOLT-4	102.50
RPMI-8226	42.38
SR	57.21
Renal Cancer	
786-0	103.91
A498	135.43
ACHN	96.47
CAKI-1	90.94
RXF 393	100.64
SN12C	99.49
TK-10	289.48
UO-31	70.39
Melanoma	
LOX IMVI	88.37
M14	98.87
MALME-3M	142.95
SK-MEL-2	94.62
SK-MEL-28	114.77
SK-MEL-5	102.04
UACC-257	103.96
UACC-62	86.14
Prostate Cancer	
DU-145	107.88
CNS Cancer	
SF-268	112.77
SF-295	103.18
SF-539	88.37
SNB-19	87.28
SNB-75	77.36
U251	95.19
Mean	97.34
Delta	58.71
Range	250.85

Mean Growth Percent - Growth Percent



150 100 50 0 -50 -100 -150

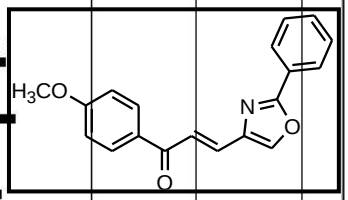
Developmental Therapeutics Program

One Dose Mean Graph

NSC: 748007 / 1	Conc: 1.00E-5 Molar	Test Date: Aug 18, 2008
Experiment ID: 0808OS98		Report Date: Sep 17, 2008

Panel/Cell Line	Growth Percent
Non-Small Cell Lung Cancer	
A549/ATCC	98.24
EKVX	103.62
HOP-62	104.38
NCI-H226	95.34
NCI-H23	92.03
NCI-H322M	110.68
NCI-H460	101.07
NCI-H522	87.32
Colon Cancer	
COLO 205	112.92
HCC-2998	98.31
HCT-116	93.89
HCT-15	100.16
HT29	113.74
KM12	98.96
SW-620	108.13
Breast Cancer	
BT-549	89.73
HS 578T	91.71
MCF7	72.32
MDA-MB-231/ATCC	94.60
MDA-MB-435	108.40
MDA-MB-468	83.65
NCI/ADR-RES	97.47
T-47D	64.25
Ovarian Cancer	
OVCAR-3	103.11
OVCAR-4	98.15
OVCAR-5	89.79
OVCAR-8	95.83
SK-OV-3	97.57
Leukemia	
CCRF-CEM	99.01
HL-60(TB)	104.41
K-562	112.29
MOLT-4	160.28
RPMI-8226	55.31
Renal Cancer	
786-0	106.69
A498	133.76
ACHN	103.09
CAKI-1	102.95
RXF 393	111.67
SN12C	103.36
TK-10	181.50
UO-31	78.80
Melanoma	
LOX IMVI	93.17
M14	101.55
MALME-3M	96.49
SK-MEL-2	97.50
SK-MEL-28	115.38
SK-MEL-5	99.09
UACC-257	101.86
UACC-62	92.01
Prostate Cancer	
DU-145	115.22
CNS Cancer	
SF-268	103.77
SF-295	109.96
SF-539	98.71
SNB-19	93.14
SNB-75	77.69
U251	95.99
Mean	100.89
Delta	45.58
Range	126.19

Mean Growth Percent - Growth Percent



150 100 50 0 -50 -100 -150

Projection of the major anticancer activity results:

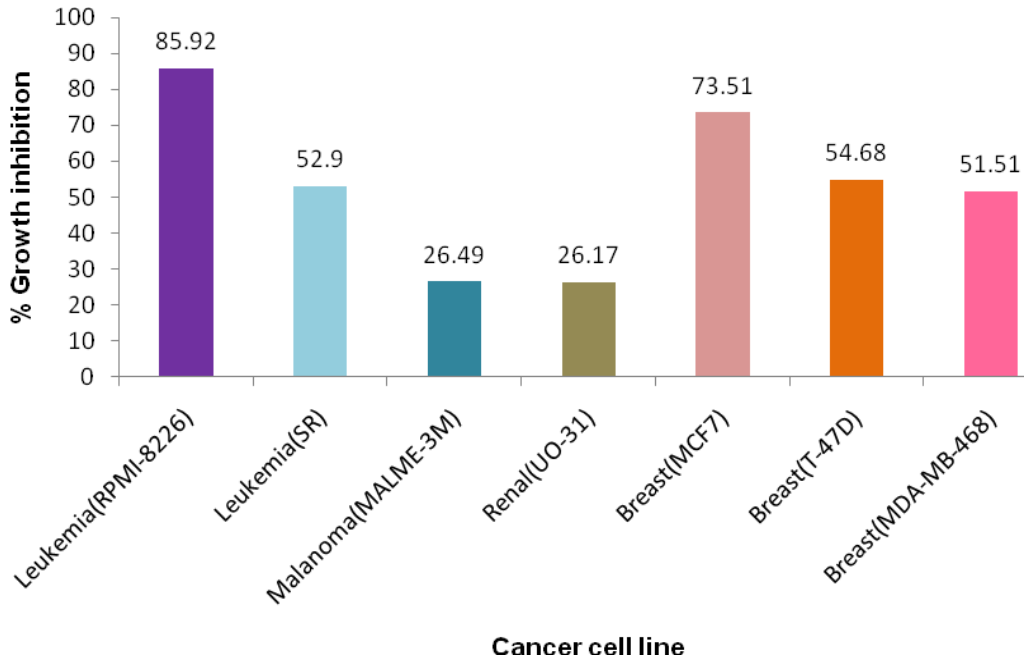


Figure-5 Results of preliminary anticancer activity of 7b at 10 μm concentration

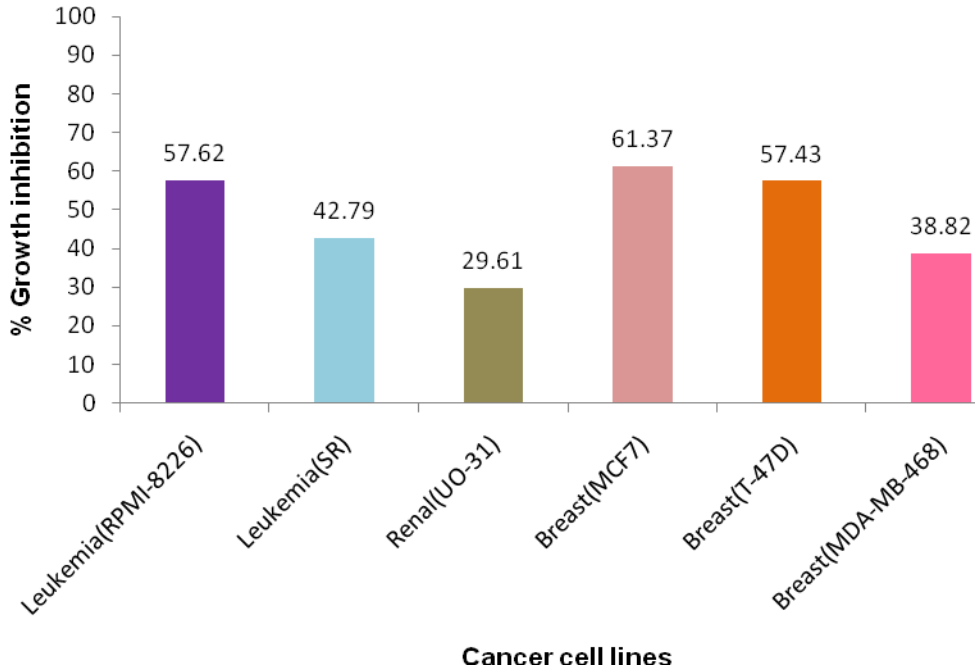


Figure-6 Results of preliminary anticancer activity of 7d at 10 □m concentration

Anti-inflammatory study of Pyrazoles and isoxazoles:

Anti-inflammatory activity of many pyrazole derivatives has been studied by various groups. Pyrano-pyrazoles and related analogues were synthesized and tested for their anti-inflammatory activity.[25] Several 5-substituted-3-(aminonephthyl)-pyrazoles were prepared and their anti-inflammatory and ulcerogenic *in-vivo* activity evaluated.[26] Anti-inflammatory, ulcerogenic and molluscicidal properties of novel bis-(acyl pyrazollines) were evaluated *in-vivo*.[27] A study on COX-1 and COX-2 inhibitory activities, ulcerogenic effects and acute toxicity for some newly designed pyrazole derivatives to find their value as anti-inflammatory agents was carried out.[28] Some indolyl-pyrazoline derivatives have been synthesized and evaluated as COX-2 and lipoxygenase (LOX) inhibitors.[29] Pyrazole and isoxazole derivatives from curcumine compounds were synthesized and evaluated for their antioxidant, COX-1, COX-2 and anti-inflammatory activities.[30] Some diaryl acetyl pyrazolines were synthesized and their anti-inflammatory and analgesic activities evaluated by paw oedema test on rats.[31]

Similarly there are many reports found for anti-inflammatory ativity of isoxazoles. Synthesis of imidazolyl substituted isoxazolines showed comparable anti-inflammatory acitivity.[32] Design and synthesis of 4,5-diphenyl-4-isoxazolines as inhibitors of COX-2 with analgesic and anti-inflammatory activities has been studied.[33] Isoxicam and related 4-hydroxy-N-isoxazolyl-benzothiazine-carboxamides were evaluated as anti-inflammatory agents in the carrageenin-induced rats.[34] Synthesis and anti-inflammatory activity of benzopyran substituted isoxazolines[35] and isoxazolinyl derivatives of anthranilic acid has been reported.[36]

After finding and studying a number of reports on the study of anti-inflammatory activity of pyrazoles and isoxazoles, the newly synthesized 3-aryl-5-(phenyl-oxazolyl)-pyrazoles and their isoxazole analogues were subjected to the study for their preliminary *in-vivo* anti-inflammatory activity using a single dose of 30mg/Kg body weight as suggested by our collaborators at Zydus Research Centre, Ahmedabad.

8 **10**

a:- X= H, **b**:- X=Cl, **c**:- X=Br, **d**:- X=CH_3, **e**:- X=OCH_3

Figure-7

Five 3-aryl-5-(2-phenyl-oxazole-4-yl)-4,5-dihydro-isoxazole **8** and five 3-aryl-5-(2-phenyl-oxazole-4-yl)-*1H*-pyrazoles **10** and were evaluated for anti-inflammatory activity using carrageenin-induced paw oedema standard method in rats.[37] Each compound was fed to a group of six male wistar rats (150-180 gm each) paw volumes were measured at 0, 3, and 5 hrs of carrageenin injection and the inhibition of paw volumes at different time interval was compared with the control group.

Results of this study as % inhibition in paw volume show that some compounds show weak inhibiting effect even at such a high concentration of 5-6 mg of test compounds in 300-400 μl methyl cellulose solution (0.5%) as vehicle per animal. The measurement using of plathysmometer after 3 hrs show that out of the compounds tested for *in-vivo* anti-inflammatory activity 4-methoxy phenyl **8e** and 4-chloro phenyl **8b** isoxazolines and 4-mehyl phenyl **10d** pyrazole derivatives show noteworthy anti-inflammatory activity compared to the others. While 4-methoxyphenyl isoxazoline **8e** and 4-chloro phenyl pyrazole **10b** have weaker anti-inflammatory effects compare to them.

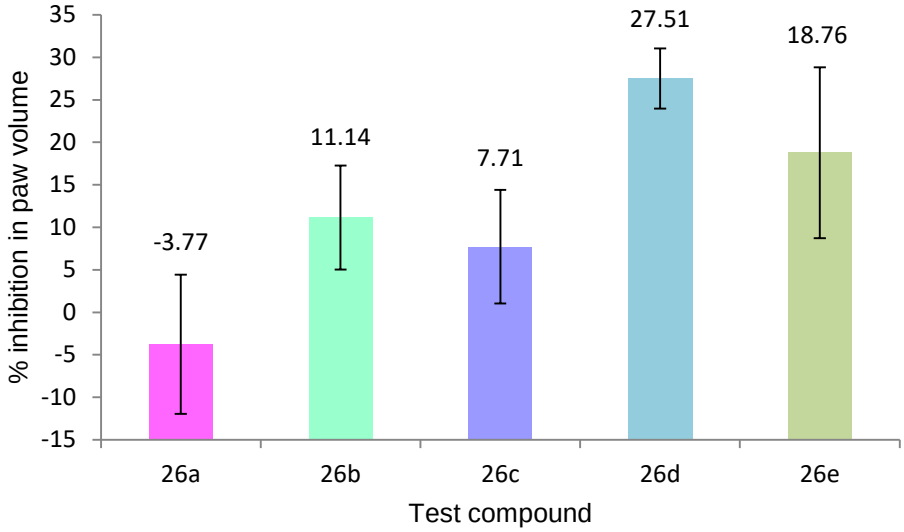


Figure-8 Results of % inhibition in paw volume at 3.0 hrs for Pyrazoles 10

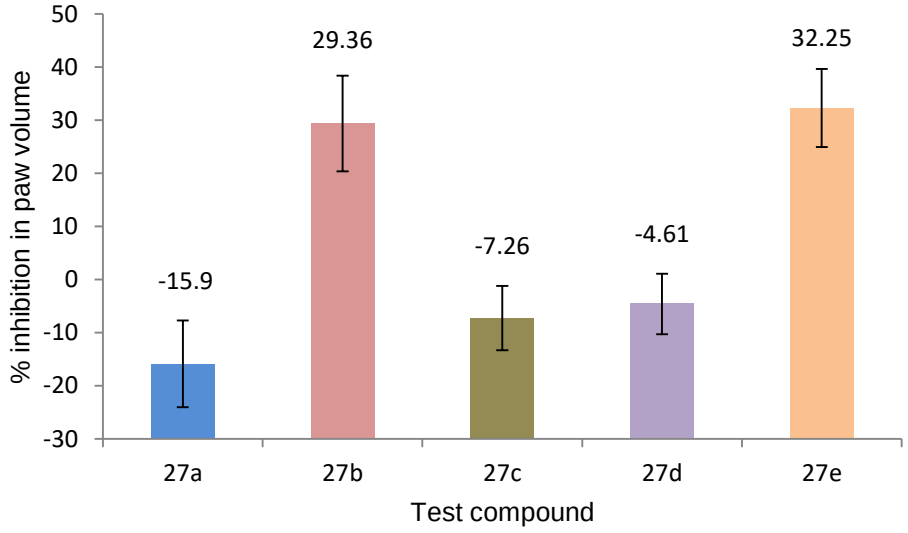


Figure-9 Results of % inhibition in paw volume at 3.0 hrs for Isoxazolines 8

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