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Solvent Free Synthesis and 1,3-Dipolar Cycloaddition Reactions of *N*-Methyl-4-(Trimethylsilyl)-C-Phenyl Nitrone and Potential Anticancer Activities of Cycloadducts

Bhaskar Chakraborty[1*], Sushma Tamang Pradhana[2], Saurav Kafley[1] and Archana Sharma[1]

[1]*Organic Chemistry Laboratory, Sikkim Government College (NBBDC), Gangtok 737102, Sikkim, India.*

[2]*Academic Visitor, School of Chemistry, Cardiff University, Park Place, Cardiff CF10 3AT, Wales, UK.*

***Corresponding author**

Bhaskar Chakraborty,
Organic Chemistry Laboratory,
Sikkim Government College (NBBDC),
Gangtok 737102, Sikkim,
India.

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Abstract

Synthesis and cycloaddition reactions of N-Methyl-4-(Trimethylsilyl)-C-phenyl nitrone using mechanochemical procedure has been reported. Trimethyl substituent is chosen as this group is known to enhance nitrone geration and anticarcinogenic activities in cycloadducts. Trimethylsilyl group at 4-position of the phenyl ring activates aldehyde group and thereby the development of N-Methyl-4-(trimethylsilyl)-C-phenyl nitrone is much faster than other nitrones A notable change in reaction rate as well as yields of the trimethylsilylnitrone and trimethylsilyl cycloadducts has been observed comparing to microwave and coventional cycloaddition procedures. Solid phase mechanochemical procedure thus found to be more suitable than other greener techniques. The present study reports synthesis of N-Methyl-4-(Trimethylsilyl)-C-phenyl nitrone and their cycloaddition reactions with activated alkenes and electron deficient alkynes. Few synthesized new trimethylsilyl cycloadducts found to exhibit significant anticancer activities too.

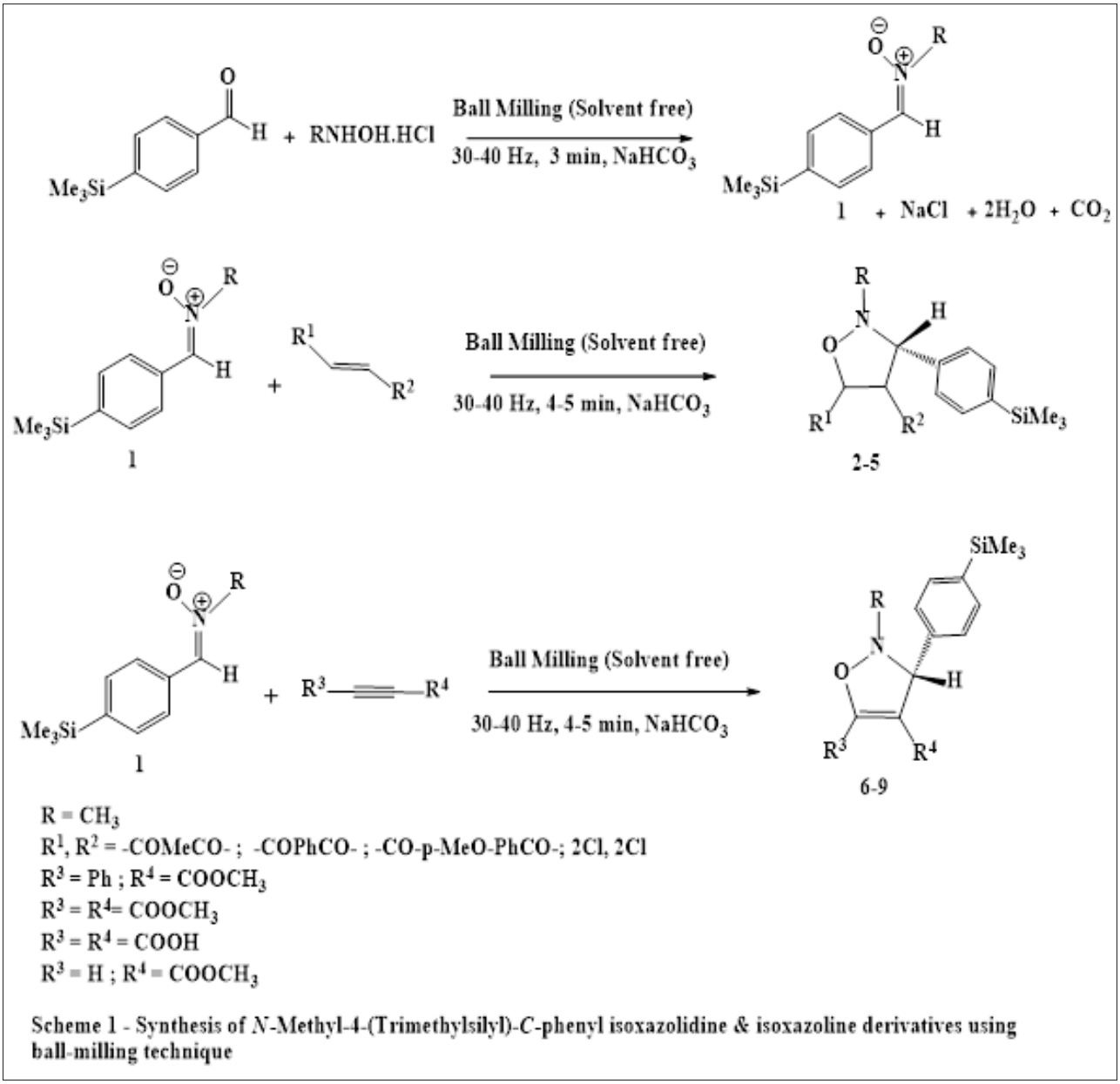
Keywords: Mechanochemistry, trimethylsilyl nitrone, intermolecular cycloaddition reaction, trimethylsilyl isoxazolidine & isoxazolines, anticancer activity.

Introduction

Perhaps nitrones are one of the most important reaction intermediates in synthetic organic chemistry as far as the construction of 5-membered oxygen-nitrogen heterocyclic molecules is concerned (Thakur et al., 2021; Tian et al., 2024). The synthesis, isolation and cycloaddition reactions of nitrones with activated olefins/ alkynes are always a point of great interest not only because of the synthetic potentials of cycloadducts (isoxazolidine & isoxazoline derivatives) but also for their significant biological activities (Patterson et al., 1992; Wagner et al., 2004; Confalone & Huie, 1988; Feuer, 2008; Padwa & Pearson, 2002). Many new approaches in this chemistry is widely known but greener synthesis (mechanochemical) and cycloaddition reactions of silyl nitrones and silyl cycloadducts has not reported too many (Kim & Lee, 1991; Ishikawa et al., 2000; Liu et al., 2019). Asymmetric silyl nitrone cycloadditions with N-acryloyl (2R)-bornane-10,2-sultam, and N-methacryloyl (2R)-bornane-10,2-sultam have been reported by J.Y Lee and coworkers in early 1991 followed by S. Saito and his group on silyl derivatives of nitrone cycloaddition reactions in early 2000. These silyl nitrone cycloaddition methodologies provide a general route for the regioselective and diastereoselective intramolecular asymmetric synthesis of 2-isoxazolines and isoxazolidine derivatives respectively. Due to versatile applications of silyl nitrone derived cycloadducts in antitumour activities they are of special interest in organic synthesis. The introduction of silyl atom as trimethylsilyl group in a particular position of an organic molecule can significantly change the bioactivity of the molecule (Gassman et al., 1992). Trimethylsilyl group at 4-position of the phenyl ring activates aldehyde group and thereby the development of N-Methyl-4-(trimethylsilyl)-C-phenyl nitrone (1) is much faster than other nitrones due the high electron donating effect of trimethylsilyl group. In anticancer studies, the silicon (Si) atom plays an important role with enhanced anti-tumor activity and reduced toxicity by strategically carbon-silicon bioisosteric replacement and thereby improving selectivity towards cancer cells. Silicon atom and silyl groups are capable of modifying the properties of potential anticancer drugs (Liu et al., 2019). Due to this property, we have chosen trimethyl silyl group for the strategic improvement of selectivity, physicochemical properties and drug-like characteristics of silyl derivatives of cycloadducts as anticancer agents. The silicon atom is capable of influencing many properties which includes solubility, permeability and stability and are crucial for drug efficacy. So choosing the type and position of silicon-containing groups, we can utilize these

properties to enhance the action of our synthesized drug (silyl cycloadducts). Researchers proved that silicon and silyl groups are capable of exhibiting enhanced selectivity for cancer cells compared to carbon-based functionalities (Liu et al., 2019). Trimethylsilyl groups in 1H NMR spectrum are found to be very close or even merged with reference TMS in maximum occasions at 0ppm. This chemical inertness makes it useful in many organic syntheses as well (Gassman et al., 1992).

Our group has reported many environment friendly (green chemistry) procedures already in nitrone cycloaddition reactions and further applications of cycloadducts including peptide synthesis (Chakraborty et al., 2017; Chakraborty & Luitel, 2015; Chakraborty, 2020; Chakraborty & Chhetri, 2018; Chakraborty et al., 2013; Chakraborty & Luitel, 2013; Chakraborty & Sharma, 2012; Chakraborty & Rai, 2018). SiMe3 as substituent plays vital role in synthesis and also, we have found its many applications of SiMe3 substituted isoxazolidine and isoxazoline derivatives (Liu et al., 2019). We report in this communication synthesis of some important new stable trimethylsilyl cycloadducts (isoxazolidine & isoxazoline derivatives) using mechanochemical procedure (Scheme 1; Table 1) (Ranu & Stolle, 2015; Huskic et al., 2012; Hernandez & Juaristi, 2011; Hernandez et al., 2012; Wang et al., 2012; Jorres et al., 2013; Trotzki et al., 2008; Mack & Shumba, 2007; Waddell& Mack, 2009; Schneider et al., 2009; Cravotto et al., 2012; Fulmer et al., 2009; Thorwirth et al., 2010; Thorwirth et al., 2011; Cook et al., 2013; Estevez et al., 2013; Su et al., 2011; Strukil et al., 2012; Hernandez & Juaristi, 2010; Tan et al., 2014; Fang et al., 2014; Crossey et al., 2015; IDokli & Gredicak, 2015; Oliveira et al., 2014; Metro et al., 2015; Achar et al., 2014) and also significant anticancer activities of some new trimethylsilyl cycloadducts. We have compared the synthesis of silyl cycloadducts in mechanochemical procedure with microwave and conventional methodologies and found significant acceleration in reaction rate and yield of the silyl cycloadducts in mechanochemical procedure. Mechanochemical procedures involving ball-milling technique are very popular in today's scenario for its many environmental friendly aspects and hence attracted the attention of mainly synthetic organic chemists (Ranu & Stolle, 2015; Huskic et al., 2012). Conducting organic synthesis under environment friendly conditions are always a challenge therefore, we have conducted ball-milling technique as a lucrative methodology in our synthesis. After utilizing almost all the greener procedures and reported the results from our laboratory we recommend "mechanochemical procedure" as one of the best procedures for future organic chemists (Hernandez & Juaristi, 2011; Hernandez et al., 2012; Wang et al., 2012; Jorres et al., 2013; Trotzki et al., 2008; Mack & Shumba, 2007; Waddell& Mack, 2009; Schneider et al., 2009; Cravotto et al., 2012; Fulmer et al., 2009; Thorwirth et al., 2010; Thorwirth et al., 2011; Cook et al., 2013; Estevez et al., 2013; Su et al., 2011; Strukil et al., 2012; Hernandez & Juaristi, 2010; Tan et al., 2014; Fang et al., 2014; Crossey et al., 2015; IDokli & Gredicak, 2015; Oliveira et al., 2014; Metro et al., 2015; Achar et al., 2014). Like majority of usual nitrones, trimethylsilyl nitrone synthesized in our laboratory are stable (melting point: 72^0C), therefore utilization of the nitrone in cycloadditions reaction becomes very easy. Moreover, newly synthesized silyl-cycloadducts found to exhibit significant anticancer activities. Hence, trimethylsilyl nitrone may be regarded as an important precursor for the synthesis of new anticancer drugs which could attract organic and medicinal chemists in research.



Scheme 1 - Synthesis of *N*-Methyl-4-(Trimethylsilyl)-*C*-phenyl isoxazolidine & isoxazoline derivatives using ball-milling technique

Results & Discussion

In our new endeavour of *N*-Methyl-4-(Trimethylsilyl)-C-phenyl nitrone (1) synthesis, we have conducted the reactions taking one equivalent each of 4-trimethylsilylbenzaldehyde and *N*-methylhydroxylaminehydrochloride along with one equivalent of sodium bicarbonate for the synthesis of trimethylsilyl nitrone 1 (Scheme 1; R = Me). The synthesized trimethylsilyl nitrone has been used for cycloaddition reactions without further purification. 4-Trimethylsilyl benzaldehyde derived silyl nitrone was stable (m.p: 72^{0}C) and cycloaddition reactions were performed with dipolarophiles in 1:1 ratio. In ball-milling procedure, usually heat and slight pressure is developed in the reaction vessel. In our study, best results have been obtained when 1:1 ratio of starting materials were used but we have also observed incomplete conversion to molecules when tried with different (1:2 and 2:1) ratios of starting materials.

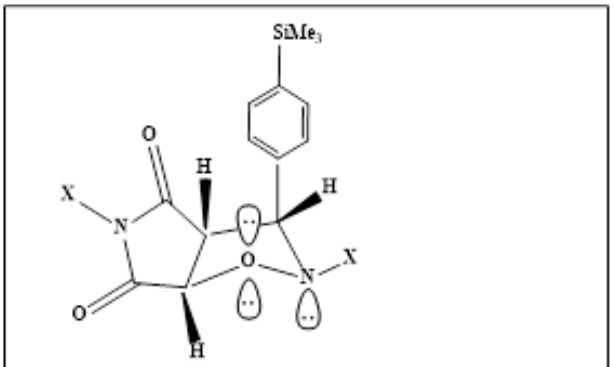
We added sodium bicarbonate in the reaction mixture in the synthesis of silyl nitrones because we had observed that it could activate the N-methylhydroxylamine. The probable reason could be due to the addition of sodium bicarbonate the reaction mixture becomes faintly alkaline and the liberated HCl is neutralized. We have also observed that if we conduct the ball-milling procedure without adding sodium bicarbonate, the reaction leads to develop a paste or a gum and ingredients do not mix well. Solid material also helps in free flow proceedings in ball-milling techniques. This was our purpose to use solid sodium bicarbonate for more efficiency in the process and to obtain best results.

^{1}H NMR spectrum of the crude products has been also studied also at a lower frequency (10-20Hz) but we didn't see any indications of developments of our target molecules. May be at a lower frequency the reactions rates were very slow and needed almost 45 minutes for the completion of the reaction and in few reactions, the ingredients 4-Trimethylsilylbenzalaldehyde and *N*-methylhydroxylamine was also present along with the crude products. It could be due to lesser amount of energy per impact is involved and ball-milling procedure with an interval

develops lower conversion rates from starting materials to products.

In mechanochemistry, the development of nitrone was fast and therefore it becomes easy for conducting in situ cycloaddition reactions if researchers so desire. But in our study, we wanted to study the stability and characterize the new trimethylsilyl nitrone, therefore we had conducted intermolecular cycloaddition reactions of the trimethylsilyl nitrone with various activated double bonded dipolarophiles (maleimides) as well as electron deficient dipolarophiles (alkynes). After successful study using various reaction conditions and trials, finally we decided to run the cycloaddition reactions in ball-milling process at a frequency of 30-40Hz and found excellent development of silyl cycloadducts in 4-5 minutes of ball-milling. We have also studied these reactions using acetonitrile (1 ml) as solvent (polar solvent) in ball-milling procedure and found slight less developments and lower yields of silyl cycloadducts. The yields of silyl cycloadducts using mechanochemical procedure were also compared (Table 1) with microwave technology (MWI) under solvent-free conditions (Loupy, 2006; Banerji et al., 2004). We found much lower yields of silyl nitrone and silyl cycloadducts in MWI. Microwave methodology, for the synthesis of silyl nitrone and silyl cycloadducts required high temperature (100-1100C) and average time required for the synthesis of silyl nitrone and silyl cycloadducts were found to be 30 – 40 minutes respectively. In addition, complete conversion of the starting materials to our target molecules was not satisfactory. Probably, it could be due to the degradation of N-methylhydroxylaminehydrochloride. Continuing our efforts, we have also studied DMF as solvent in MWI methodology for cycloaddition reactions but yields (silyl isoxazolidine & isoxazolines) were found to be 74-75% which was regarded as poor in comparison with ball-milling process.

1H NMR spectroscopy technique was mainly used for the confirmation of the structures of newly synthesized molecules (trimethylsilyl isoxazolidine & isoxazoline derivatives) and trimethylsilyl nitrone (Deshong et al., 1991; Gandolfi & Grunanger, 1999; Yu et al., 1993). In addition, ^{13}C NMR, MS & IR spectroscopic techniques also have been used successfully. 1H NMR spectrum of the silyl isoxazolidine and isoxazoline derivatives (2-5 & 6-9) reveals that the structures are expected to be symmetrical in nature. The silyl isoxazolidine derivatives exhibit enantioselectivities as well. Nitrone 1 exists exclusively in Z configuration and syn cycloadducts are believed to develop from Z nitrone via an exo transition state geometry. The configurations of C_3, C_4, C_5 protons of the silylcloadducts are expected to be syn in nature because the coupling constant (J ~ 2.00-3.00Hz, for C_4-C_5 & J ~ 2.00-3.00 Hz, for C_3-C_4) values are in good agreement with reported values in our published research articles in literature Deshong et al. (1991); Gandolfi & Grunanger (1999) and Yu et al. (1993) as well. Therefore, we may assume from these J values that the dipolarophiles with syn configuration produced syn silyl cycloadducts and the addition of silyl nitrone to electron rich alkenes (maleimides) were stereospecifically syn in nature. In maleimide cycloadducts, 3-H, 4-H protons are syn orientated and are diastereoselective in nature. The coupling constant (J) values of 3-H, 4-H protons ($J_{3,4}$ ~ 2.00 Hz) are also in good agreement (Deshong et al., 1991; Huisgen, 1976; Houk & Yamaguchi, 1984) in favour of cis oriented structure. Due to the absence of 4-H protons, in case of fluoro isoxazoline derivatives, exact geometry could not be determined. Trimethylsilyl groups in all the cycloadducts are found to have merged with reference TMS in ^1H NMR spectrum.



Probable conformational structure of trimethylsilyl isoxazolidines

The probable conformational structure indicates the substituents at the C-3, C-4 & C-5 positions tries to have equatorial positions while all the H-atoms at these positions are axial respectively. We have observed and obtained expected fragmentation peaks in the mass spectrum including molecular ion (M^+) and base peaks (BP) in all the trimethylsilyl isoxazolidine derivatives. In case of silyl isoxazoline derivatives in addition to M^+ other prominent peaks are also obtained. This is due the fragmentations of $COOCH_3$, Ph and COOH groups which subsequently develop of aziridine derivatives.

Anticancer Study

Majority of the newly synthesized heterocycles in recent years are screened for cancer studies (Sanjai et al., 2024; Campoccia et al., 2021) as their probability to act as anticancer drugs is very high. Cytotoxicity of the compounds was determined on the basis of measurement of in vitro growth inhibition of tumor cell lines (Sanjai et al., 2024; Campoccia et al., 2021), A549 derived from human alveolar adenocarcinoma epithelial cells (ATCC No.CCL-180), HeLa derived from human cervical cancer cells (ATCC No. CCL-14), MDA-MB-167 derived from human breast adenocarcinoma cells (ATCC No. HTB-35) and MCF7 derived from human breast adenocarcinoma cells (ATCC No. HTB-40) using the MTT assay. The IC50 values (50% inhibitory concentration in µM) are expressed as the average of two independent experiments. The effect of cycloadducts (2-9) on the growth of cancer cell lines were determined following the general procedure used by the National Cancer Institute for in vitro anticancer drug study. The procedure uses the protein-binding dye Sulphorhodamine B for the estimation of cell growth (Sanjai et al., 2024; Cai et al., 2019). In due course of time, the growth of the cells were counted (95 cells per well in 100 mL medium) in 90 microtitre plates. The study has been conducted keeping the cells for incubation for 40 hrs at 20^0C. The experimental set-up of three

different wells was conducted where the cells were kept for 36 hrs. This was followed by reacting the cells with 30% cold (5-10^{0}C) TCA. It was left for 2 hrs at 20^{0}C and then washed and dried in air. All the cells were stained with Sulphorhodamine B dye. The dye was dissolved in tris-buffer solution. The plates under study were taken in shaker and kept for 20-30 minutes. The cell growth was calculated using optical density (OD) study and the results were reported in terms of IC_{50} values (Chakraborty & Roy 2022). Doxorubicin was considered as standard reference.

From the study of IC50 values, it has been found that six (6) newly synthesized cycloadducts showed significant cytotoxicity against human alveolar adenocarcinoma epithelial cells, human cervical cancer cells, human breast adenocarcinoma cells and human breast adenocarcinoma cells respectively. Among all the tested cycloadducts (2-9), 2, 4, 5 & 7, 8 showed comparatively more potent IC50 value against (ATCC No.CCL-180), HeLa derived from human cervical cancer cells (ATCC No. CCL-14), MDA-MB-167 derived from human breast adenocarcinoma cells (ATCC No. HTB-35) and MCF7 derived from human breast adenocarcinoma cells (ATCC No. HTB-40) as compared to other silyl cycloadducts. Based upon the study, three most potent silyl isoxazolidine derivatives 2, 4 and 5 were taken for cell cycle analysis. Cell cycle analysis is going on at present.

Compound (µg mL^{-1})	HeLa (Cervical)	MDA-MB-231 (Breast)	MCF-7 (Breast)	A549 (Lung cancer)
2	96	88	95	95
3	55	93	82	56
4	77	94	64	84
5	75	86	70	74
7	74	69	50	20
8	66	60	48	40
9	57	62	50	69
Doxorubicin (Standard)	0.8	2.00	0.4	0.6

Table 1: IC50 values (µM) of various trimethylsilyl isoxazolidine & isoxazoline derivatives.

Experimental

Unless otherwise stated, all reagents were obtained from commercial sources and used without further purification. Progress of all the reactions were monitored by TLC using 0.25 mm silica gel plates (Merck 60F_{254}, UV indicator). Column chromatography was performed with silica gel (E. Merck, Germany) with 60–200 mesh. All other reagents and solvents were purified before starting reactions or column chromatography. ^{1}H NMR spectra were recorded on a Bruker DRX 300 (300 Hz) spectrometer at ambient temperature. 13C NMR spectra were recorded on a Bruker DRX 300 (75 Hz) spectrometer at ambient temperature. The coupling constants (*J*) are expressed in Hz. Infra-red spectra were recorded on a Perkin-Elmer RX 1-881 machine as a film or KBr pellets. Mass-spectrometry data was recorded using a Joel SX-102 (FAB). The ball mill used was a Retsch MM500 mixer mill digital GmbH,42781Haan, Germany. Reactions were carried out using stainless steel jars from Retsch. Milling balls were purchased from Germany.

General procedure 1–General procedure for the synthesis of *N*-Methyl-4-(Trimethylsilyl)-C-phenyl nitrone (1) in ball-milling procedure

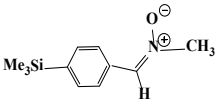
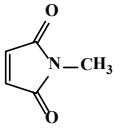
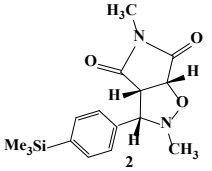
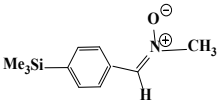
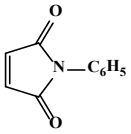
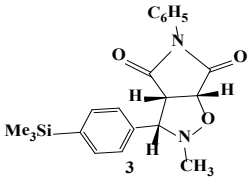
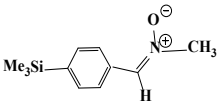
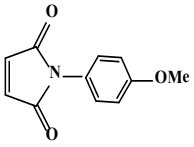
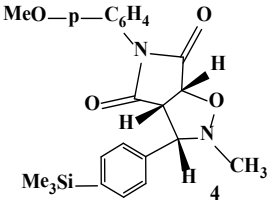
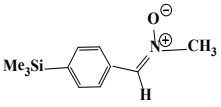
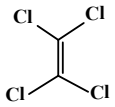
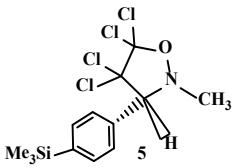
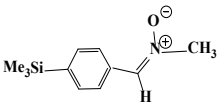
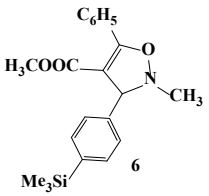
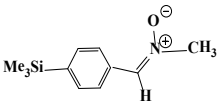
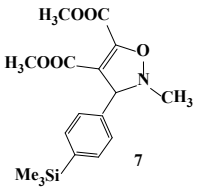
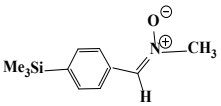
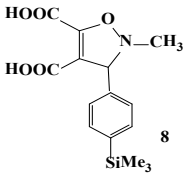
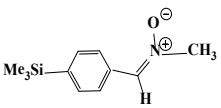
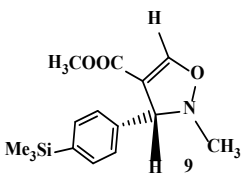
N-methylhydroxylamine hydrochloride (1 mmol), 4-(Trimethylsilyl)-benzaldehyde (1 equivalent), and $NaHCO_3$ (1.0 equivalent) was mixed together and ball-milled at 30 Hz for 3 min in a 25 mL steel vessel and 15 mm diameter balls. After the completion of reaction, the reaction mixture was taken in CH2Cl_2. It was filtered on cotton for the removal of NaCl. The filtrate was evaporated under vacuum to afford *N*-Methyl-4-(Trimethylsilyl)-C-phenyl nitrone 1 as white crystalline solid with high purity (92%, m.p; 72^{0}C).

Spectroscopic data for nitrone 1: UV λmax 235 nm; IR (KBr): υmax 3015 (m), 2230 (m), 1685 (m), 1620 (s), 1430 (m), 1155 (m), 786 (s) cm^{-1}. ^{1}H NMR ($CDCl_3$): δ 7.68-7.20 (m, 4H, phenyl protons), 3.38 (s, 3H, CH_3), 1.70 (s, 1H, -CH=N$^+$); ^{13}CNMR ($CDCl_3$): δ 142.12 (CH=N$^+$), 138.82, 135.30, 133.10, 130.00 (phenyl carbons), 30.55 (CH_3).

General procedure II–Mechanochemical synthesis of Trimethylsilyl isoxazolidine & isoxazolidine derivatives from *N*-Methyl-4-(Trimethylsilyl)-C-phenyl nitrone (Table 2; entry 1)

N-Methyl-4-(Trimethylsilyl)-C-phenyl nitrone (1 equivalent), *N*-methyl maleimide (1 equivalent) and $NaHCO_3$ (1 equivalent) were mixed together and ball-milled at 30 Hz for 4-5 min in a 25 mL steel vessel and 15 mm diameter balls. After the completion of reaction, the reaction mixture was taken in CH_2Cl_2. It was filtered on cotton for the removal of NaCl. The filtrate was evaporated under vacuum to afford crude isoxazolidine derivative as white crystals (95%). The crude product was directly charged on silica gel column and eluted with a mixture of ethyl acetate: n-hexane (1:8) resulting pure isoxazolidine 2 (entry 1, Table 2, Scheme 1). Same methodology was adopted for the synthesis of other trimethylsilyl isoxazolidine & isoxazoline derivatives (entry 2-9).

Table 2: Synthesis of new *N*-Methyl-4-(Trimethylsilyl)-C-phenyl-isoxazolidine derivatives

Entry	Nitrone (1)	Dipolarophile[a]	Time(min)	Cycloadduct[b](2-7	ee	Yield[c](%)
1			4 (40)		90	95 (82)
2			4 (45)		88	94 (80)
3			5 (45)		83	93 (76)
4			5 (40)		83	92 (76)
5			5 (45)		84	91(70)
6			5 (50)		84	90 (68)
7			5 (50)		88	90 (70)
8			5 (55)		88	90 (70)

[a]Reaction conditions: nitrone (1 mmol), dipolarophiles (1 equivalent), ball-milling, (30-40Hz)
[b]All products were characterized by IR,1H NMR, 13C NMR and MS spectral data.
[c]Isolated yield after purification. Figures in parentheses indicate yields obtained under MWI.

(3S)-2-methyl-3-(4-trimethylsilyl)-dihydro-5-methyl-2H-pyrrolo[3,4-d]isoxazole-4,6(5H,6a-H)-dione, 2

White crystals. M.P 1060C; Yield 93%; Rf= 0.66; IR (KBr): υmax 3300 (m), 2960 (m), 2840 (m), 1760 (s), 1665 (s), 1480 (m), 1320 (m), 785 (s) cm^{-1}; ^1H NMR (CDCl3): δ 7.76 – 7.28 (m, 4H, aromatic protons), 6.50 (d, 1H, *J* = 2Hz, C5H), 6.40 (d, 1H, *J* = 2 Hz, C3H), 3.83 (br, s, 6H, 2XCH3), 2.30 (dd, 1H, *J* = 2.00, 2.00 Hz, C4H); ^{13}C NMR ($CDCl_3$): δ 169.00, 168.oo (carbonyl carbons), 138.46, 138.10, 137.20, 131.42, 130.80. 128.15 (aromatic carbons carbons), 66.40 (C_5), 52.82 (C_4), 32.20 (C_3), 26.20, 25.00 (CH_3 carbons); FAB-MS: m/z 318 (M^+), 288, 169, 154 (BP), 149.

(3S)-2-methyl-3-(4-trimethylsilyl)-dihydro-5-phenyl-2H-pyrrolo[3,4-d]isoxazole-4,6(5H,6a-H)-dione, 3

White solid. M.P 115^{0}C; Yield 92%; R_f= 0.64; IR (KBr): υmax 3230 (m), 2980 (m), 2790 (m), 1760 (s), 1680 (s), 1440 (s), 1260 (m), 782 (s) cm^{-1}; ^1H NMR ($CDCl_3$): δ 7.70 – 7.68 (m, 4H, aromatic protons), 7. 60-7.20 (m, 5H, aromatic protons). 6.50 (d, 1H, *J* = 2.10 Hz, C_5H), 6.36 (d, 1H, *J* = 2.10 Hz, C_3H), 3.80 (dd, 1H, *J* = 2.00, 2.00 Hz, C_4H), 1.80 (s, 3H, N-CH_3); 13C NMR ($CDCl_3$): δ 172.27, 166.90 (carbonyl carbons), 136.40, 135.24, 133.50, 132.46, 131.00, 130.30, 130. 72, 128.20, 128.00, 127.68 (phenyl carbons), 72.30 (C_5), 55.50 (C_3), 50.00 (C_4), 28.60 (N-CH_3), 24.50; FAB-MS: m/z 380 (M^+), 365, 303, 231, 216 (BP), 149, 77.

(3S)-2-phenyl-3-(4-trimethylsilyl)-dihydro-5-methyl-2H-pyrrolo[3,4-d]isoxazole-4,6(5H,6a-H)-dione, 4

White solid. M.P 96^{0}C; Yield 92%; R_f= 0.60; IR (KBr): υmax 3240 (m), 2982 (m), 2785 (m), 1760 (s), 1682 (s), 1440 (s), 1250 (m), 788 (s) cm^{-1}; ^1H NMR ($CDCl_3$): δ 7.64 – 7.18 (m, 2x4H, aromatic protons), 6.52 (d, 1H, *J* = 2.10 Hz, C_5H), 6.40 (d, 1H, $_J$ = 2.00 Hz, C_3H), 3.60 (s, 3H, OMe), 2.30 (dd, 1H, *J* = 2.00, 2.00 Hz, C_4H), 1.25 (s, 3H, N-CH_3); ^{13}C NMR ($CDCl_3$): δ 172.27, 166.90 (carbonyl carbons), 136.40, 135.24, 133.50, 132.46, 130. 72, 128.20, 127.30, 127.15 (phenyl carbons), 72.30 (C_5), 55.50 (C_3), 50.00 (C_4), 28.60 (N-CH_3), 24.50 (O-Me); FAB-MS: m/z 410 (M+), 303, 246 (BP), 149, 107.

(3S)-2-methyl-3-(4-trimethylsilyl)-dihydro-4,5-tetrachloro-2H-pyrrolo[3,4-d]isoxazolidine 5

White solid. M.P 123^{0}C; Yield 91%; R_f= 0.66; IR (KBr): υmax 3250 (m), 2960 (m), 1760 (s), 1680 (s), 1365 (m), 780 (s) cm^{-1};^1H NMR (CDCl3): δ 7.80 – 7.25 (m, 4H), 3.80 (s. 3H. N-CH_3), 1.72 (s, 1H,C_3H); 13C NMR ($CDCl_3$): δ 135.90, 135.70, 135.30, 135.00 (phenyl ring carbons), 76.30 (C_5), 60.18 (C_3), 54.45 (C_4), 24.00 (N-CH_3); FAB-MS: m/z 373 (M+), 302, 224, 209 (BP), 149.

(S)-methyl-2-methyl-3-(4-trimethylsilyl)-2,3-dihydro-5-phenylisoxazole-4-carboxylate, 6

Red thick liquid. Yield 91%; R_f= 0.70; IR (KBr): υmax3380 (m), 2250 (m), 1760 (s), 1700 (s), 1680 (s),1482 (s),1210 (s), 780 (s) cm^{-1}; ^1H NMR (CDCl3): δ 7.80 – 7.20 (m, merged signals of 5H & 4H aromatic protons), 3.80 (s, 3H, -$COOCH_3$), 3.34 (s, 3H, CH_3), 1.26 (s, 1H, C_3H)); 13C NMR ($CDCl_3$):δ 172.00 (-$COOCH_3$), 138.40, 138.00, 137.40, 137.10, 136.40, 132.80, 130.50, 129.66, 129.40 (phenyl carbons), 82.00 (C_5), 57.40 (C_3), 50.00 (C_4), 29.00 (-$COOCH_3$); FAB - MS (m/z):367 (M+), 308, 290, 218 (BP), 203, 77, 59.

(S)-dimethyl-2-methyl-3-(4-trimethylsilyl)-2,3-dihydroisoxazole-4,5-dicarboxylate, 7

Red liquid. Yield 90%; R_f= 0.60; IR (KBr): υmax3290 (m), 2180 (m), 1740 (s), 1670 (s), 1620 (s), 1260 (s), 870 (m), 776 (s) cm^{-1}; ^1H NMR ($CDCl_3$): δ 7.76 – 7.28 (m, 4H, phenyl protons), 3.88 (s, 6H, 2X -$COOCH_3$), 1.70 (s, 1H, C_3H), 1.25 (s,3H, N-CH_3); 13C NMR ($CDCl_3$):δ 172.00, 170.00 (-$COOCH_3$, carbonyl carbons of the ester group), 137.00, 133.40, 131.50, 130.00, 72.00 (C_5H), 57.50 (C_3H), 43.50 (C_4H), 27.00 (*N*-CH_3); FAB - MS (m/z): 349 (M+), 334, 200, 185 (BP), 149.

(S)-2-methyl-3-(4-trimethylsilyl)-2,3-dihydroisoxazole-4,5-dicarboxylic acid, 8

Colourless thick liquid. Yield 90%; R_f= 0.64; IR (KBr): υmax3180 (m), 2990 (br), 1760 (s), 1485 (s), 1360 (s), 1210 (s), 790 (s) cm^{-1}; 1H NMR ($CDCl_3$): δ 10.70 (s, 2H, 2XCOOH), 7.67 – 7.48 (m, 4H, phenyl protons), 3.36 (s, 3H, CH_3), 1.30 (s,1H, C_3H); 13C NMR ($CDCl_3$):δ 173.20, 172.50 (carboxyl carbons), 138.50, 137.00, 135.00, 134.50, 130.00 (phenyl carbons), 68.00 (C_5), 58.00 (C_3), 36.00 (C_4), 28.00 (*N*-CH_3 carbon); FAB - MS (m/z):321 (M+), 306, 172, 157 (BP), 45.

(S)-2-methyl-3-(4-trimethylsilyl)-2,3-dihydroisoxazole-4-dicarboxylic acid, 9

Red thick liquid. Yield 88%; R_f = 0.66; IR (KBr): υmax3230 (m), 2240 (m), 1740 (s), 1715 (s), 1680 (s), 1320 (s), 780 (s) cm^{-1}; ^1H NMR ($CDCl_3$): δ 7.76 – 7.30 (m, 4H, phenyl protons), 3.80 (s, 3H, -$COOCH_3$), 3.18 (s,1H, C_5H), 1.74 (s,1H, C_3H), 1.24 (s,3H, N-CH_3); 13C NMR ($CDCl_3$):δ 172.50 (-$COOCH_3$), 137.00, 136.00, 132.00, 129.30 (phenyl carbons), 66.50 (C_5), 56.00 (C_3), 46.00 (C_4), 24.00 (N-CH_3); FAB - MS (m/z): 291 (M+), 276, 149, 127 (BP), 59.

Conclusion

We have adopted one of the most favourable green chemistry methodology (mechanochemistry: ball-milling) in a solid phase for the synthesis of new *N*-Methyl-4-(Trimethylsilyl)-C-phenyl nitrone and trimethylsilyl cycloadducts with excellent yields in a minimum time frame. We believe this simple, cost efficient and time saving methodology will be adopted by many more researchers in synthetic organic chemistry. Finally, we have found promising anticancer activities in few newly synthesized trimethylsilyl cycloadducts where Si atom could have played a vital role and we are hopeful in near future that we shall be able to establish these molecules as “anti-cancer drugs” after proper applications of cancer screening.

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References

1. Thakur, S., Das, A., & Das, T. (2021). 1,3-Dipolar cycloaddition of nitrones: synthesis of multisubstituted, diverse range of heterocyclic compounds. *New J Chem, 45*(26), 11420-11456. DOI: https://doi.org/10.1039/D1NJ02023D
2. Tian, X., Xuan T., Gao, J., Zhang, X., Liu, T., Luo, F., Pang, R., Shao, P., Yang, Y-F., Wang, Y. (2024). Catalytic enantioselective nitrone cycloadditions enabling collective syntheses of indole alkaloids. *Nature Commun, 15*(1), 6429. https://www.nature.com/articles/s41467-024-50509-4
3. Patterson, W., Cheung, P. S., & Ernest, M. J. (1992). *J Med Chem*, 35, 507.
4. Wagner, E., Becan, L., & Nowakowska, E. (2004). Synthesis and pharmacological assessment of derivatives of isoxazolo[4,5-d]pyrimidine. *Bio Org Med Chem, 12*(1), 265-272. DOI: https://doi.org/10.1016/j.bmc.2003.10.004
5. Confalone, P. N., & Huie, E. M. (1988). *Org. React,* 36, 1–173, Chapter 1.
6. Feuer, H. (2008). Nitrile Oxides, Nitrones and Nitronates in Organic Synthesis. Nitrones: Novel Strategies in Synthesis. Hoboken, *N. J: Wiley-Interscience*. https://archive.org/details/nitrileoxidesnit002edunse
7. Padwa, A. & Pearson, W. H. (2002). Synthetic Applications of 1,3-Dipolar Cycloaddition Chemistry Toward Heterocycles and Natural Products. *John Wiley & Sons, Inc, NY*. https://www.scirp.org/%28S%28351jmbntv-nsjt1aadkposzje%29%29/reference/referencespapers?referenceid=419494
8. Kim, H. B., & Lee, Y. J. (1991). Tetrahedron Assemetry, 2, 1359.
9. Ishikawa, T., Kudo, T., Shigemon, K., & Saito, S. (2000). *J Am Chem Soc*, 122, 7633.
10. Liu, Q., Lin, S., Liao, Q., & Zhaohua, Y. (2019). *J Chem Sci*, 73, 131.
11. Gassman, P. G., Deck, P. A., Winter, C. H., Dobbs, D. A., & Cao, D. H. (1992). Understanding electronic effects in organometallic complexes. Electron donation by the trimethylsilyl group. *Organomettallics, 11*(2), 959-960. DOI: https://doi.org/10.1021/om00038a069
12. Chakraborty, B., Chettri, M. S., & Luitel, G. P. (2017). Synthesis of Some Novel Class of Bis(isoxazoline) and Bis(aziridine) Derivatives. *J Heterocyclic Chem, 54*(2), 1611-1618. DOI: https://doi.org/10.1002/jhet.2657
13. Chakraborty, B., & Luitel, G. P. (2015). Synthesis of Some Novel Bisisoxazolidine Derivatives from Glyoxal-derived Bisnitrones via Simultaneous Double Cycloaddition Reactions in Water. *J Heterocyclic Chem, 52*(3), 726-731. DOI: https://doi.org/10.1002/jhet.2181
14. Chakraborty, B. (2020). Solvent-free synthesis and 1,3-dipolar cycloaddition reactions of N-methyl-C-(2-furyl) nitrone in a ball mill and anticancer activities of the new cycloadducts. *J Heterocyclic Chem, 57*(1), 477-485. DOI: https://doi.org/10.1002/jhet.3804
15. Chakraborty, B. & Chhetri, E. (2018). Synthesis of Some Novel Class of Regioselective Spiro Isoxazolidine Derivatives via 1,3-Dipolar Cycloaddition Reaction of N-Benzyl-C-fluoro-substituted Phenyl Nitrones in Ionic Liquid. *J Heterocyclic Chem, 55*(5), 1157-1165. DOI: https://doi.org/10.1002/jhet.3148
16. Chakraborty, B., Sharma, P. K., & Kafley, S. (2013). Microwave-assisted synthesis of novel spiro isoxazolidine derivatives. *Green Chem Lett Rev, 6*(2), 141. DOI: https://doi.org/10.1080/17518253.2012.720719
17. Chakraborty, B., & Luitel, G. P. (2013). An efficient ecofriendly protocol for the synthesis of novel fluoro isoxazoline and isoxazolidines using N-benzyl fluoro nitrone via cycloaddition reactions. *Tetrahedron Lett, 54*(8), 765-770. DOI: https://doi.org/10.1016/j.tetlet.2012.11.111
18. Chakraborty, B., & Sharma, P. K. (2012). Synthesis of Ketone from Alkyl Halide Using α-Chloro Nitrone as Oxidizing Reagent: A New Approach. *Synth Commun, 42*(12), 1804-1812. DOI: https://doi.org/10.1080/00397911.2010.544833
19. Chakraborty, B., & Rai, N. (2018). Synthesis of Some New Scaffolds of Amino Isoxazolidines and Their Further Functionalization into New Class of Peptides: A New Approach. *J Heterocyclic Chem, 55*(4), 1053-1060. DOI: https://doi.org/10.1002/jhet.3137
20. Ranu, B., Stolle, A. (2015). Ball Milling Towards Green Synthesis: Applications, Projects and Challenges. *Royal Society of Chemistry*, UK. https://api.pageplace.de/preview/DT0400.9781782621980_A26556819/preview-9781782621980_A26556819.pdf
21. Huskic, I.; Halasz, I.; Friscic, T.; Vancik, H. (2012). Mechanosynthesis of nitrosobenzenes: a proof-of-principle study in combining solvent-free synthesis with solvent-free separations. *Green Chem, 14*(6), 1597-1600. DOI: https://doi.org/10.1039/C2GC35410A
22. For selected recent examples of organic synthesis in ball mills, see:
23. Hernandez, J. G., & Juaristi, E. (2011). Asymmetric Aldol Reaction Organocatalyzed by (S)-Proline-Containing Dipeptides: Improved Stereoinduction under Solvent-Free Conditions. *J Org Chem, 76*(5), 1464–1467. DOI: https://doi.org/10.1021/jo1022469
24. Hernandez, J. G., Garcıa-Lopez, V., & Juaristi, E. (2012). Solvent-free asymmetric aldol reaction organocatalyzed by (S)-proline-containing thiodipeptides under ball-milling conditions. *Tetrahedron, 68*(1), 92-97. DOI: https://doi.org/10.1016/j.tet.2011.10.093
25. Wang, Y. F., Chen, R. X., Wang, K., Zhang, B. B., Lib, B. Z., & Xu, D.Q. (2012). Green Chem, 14, 893
26. Jorres, M., Mersmann, S., Raabe, G. & Bolm, C. (2013). Organocatalytic solvent-free hydrogen bonding-mediated asymmetric Michael additions under ball milling conditions. *Green Chem, 15*(3), 612-616. DOI: https://doi.org/10.1039/C2GC36906K
27. Trotzki, R., Hoffmann, O., & Ondruschka, B. (2008). The Knoevenagel condensation at room temperature. *Green Chem, 10*(8), 873-878. DOI: https://doi.org/10.1039/B808265K

28. Mack, J., & Shumba, M. (2007). Rate enhancement of the Morita–Baylis–Hillman reaction through mechanochemistry. *Green Chem, 9*(4), 328-330. DOI: https://doi.org/10.1039/B612983H
29. Waddell, D. C. & Mack, J. (2009). An environmentally benign solvent-free Tishchenko reaction. *Green Chem, 11*(11), 79-82. DOI: https://doi.org/10.1039/B810714A
30. Schneider, F., Szuppa, T., Stolle, A., Ondruschka, B., & Hopf, H. (2009). Energetic assessment of the Suzuki–Miyaura reaction: a curtate life cycle assessment as an easily understandable and applicable tool for reaction optimization. *Green Chem, 11*(11), 1894-1899. DOI: https://doi.org/10.1039/B915744C
31. Cravotto, G., Garella, D., Tagliapietra, D., Stolle, A., Schusler, S., Leonhardt, S. E. S., & Ondruschka, B. (2012). New J Chem, 36, 1304.
32. Fulmer, D. A., Shearouse, W. C., Medonza, S. T., & Mack, J. (2009). Solvent-free Sonogashira coupling reaction viahigh speed ball milling. *Green Chem, 11*(11), 1821-1825. DOI: https://doi.org/10.1039/B915669K
33. Thorwirth, R. Stolle, A., & Ondruschka, B. (2010). Fast copper-, ligand- and solvent-free Sonogashira coupling in a ball mill. *Green Chem, 12*(6), 985-991. DOI: https://doi.org/10.1039/C000674B
34. Thorwirth, R., Stolle, A., Ondruschka, B., Schubert, U. S. & Wild, A. (2011). Fast, ligand- and solvent-free copper-catalyzed click reactions in a ball mill. *Chem Commun, 47*(15), 4370-4372. DOI: https://doi.org/10.1039/C0CC05657J
35. Cook, T. L., Walker, L., & Mack, J. A. (2013). Scratching the catalytic surface of mechanochemistry: a multi-component CuAAC reaction using a copper reaction vial. *Green Chem, 15*(3), 617-619. DOI: https://doi.org/10.1039/C3GC36720G
36. Estevez, V., Villacampa, M., & Menendez, C. J. (2013). Three-component access to pyrroles promoted by the CAN–silver nitrate system under high-speed vibration milling conditions: a generalization of the Hantzsch pyrrole synthesis. *Chem Commun*, 49, 591-593. DOI: https://doi.org/10.1039/C2CC38099D
37. Su, W., Yu, J., Li, Z., & Jiang, Z. (2011). Solvent-Free Cross-Dehydrogenative Coupling Reactions under High Speed Ball-Milling Conditions Applied to the Synthesis of Functionalized Tetrahydroisoquinolines. *J Org Chem, 76*(21), 9144-9150. DOI: https://doi.org/10.1021/jo2015533
38. Strukil, V.;. Bartolec, B.; Portada, T.; Đilovic, I.; Halasz, I. & Margetić, D. (2012). One-pot mechanosynthesis of aromatic amides and dipeptides from carboxylic acids and amines. *Chem Commun, 48*(99), 12100-12102. DOI: https://doi.org/10.1039/C2CC36613D
39. Hernandez, G. J. & Juaristi, E. (2010). Green Synthesis of α,β- and β,β-Dipeptides under Solvent-Free Conditions. *J Org Chem, 75*(21), 7107–7111. DOI: https://doi.org/10.1021/jo101159a
40. Tan, D., Strukil, V., Mottillo, C., & Friscic, C. T. (2014). Mechanosynthesis of pharmaceutically relevant sulfonyl-(thio)ureas. *Chem Commun, 50*(40), 5248-5250. DOI: https://doi.org/10.1039/C3CC47905F
41. Fang, Y., Salame, S., Woo, B. D. S., Friscic, T., & Cuccia, C. (2014). *Cryst Eng Comm*, 16, 7180.
42. Crossey, K, Cunningham, R. N., Redpath, P., & Migaud, M. E. (2015). Atom efficient synthesis of pyrimidine and purine nucleosides by ball milling. *RSC Adv, 5*(72), 58116-58119. DOI: https://doi.org/10.1039/C5RA12239B
43. IDokli, I.; Gredicak, M. Eur J Org Chem, 2015, 2727
44. Oliveira, P.F.M.; Baron, M.; Chamayou, A.; Andre-Barres, C.; Guidetti, B.; Baltas, M. (2014). Solvent-free mechanochemical route for green synthesis of pharmaceutically attractive phenol-hydrazones. *RSC Adv, 4*(100), 56736-56742. DOI: https://doi.org/10.1039/C4RA10489G
45. Metro, T.X., Bonnamour, J., Reidon, J., Duprez, A., Sarpoulet, J., Martinez, J., Lamaty, F. (2015). Comprehensive study of the organic-solvent-free CDI-mediated acylation of various nucleophiles by mechanochemistry. *Chem Eur–J, 21*(36), 12787-96. DOI: https://doi.org/10.1002/chem.201501325
46. Achar, T. K., Maiti, S., & Mal, P. (2014). IBX works efficiently under solvent free conditions in ball milling. *RSC Adv, 4*(25), 12834-12839. DOI: https://doi.org/10.1039/C4RA00415A
47. Loupy, A. (2006). Microwaves in Organic Synthesis. *Wiley*. https://onlinelibrary.wiley.com/doi/book/10.1002/9783527619559
48. Banerji, A., Biswas, P. K., Sengupta, P., Dasgupta, S., & Gupta, M. (2004). 1,3-Dipolar cycloadditions: Part VIII1 - Microwave irradiation assisted synthesis of N-methyl-C-aryl nitrones. *Indian J Chem, 43B*(4), 880-881. https://nopr.niscpr.res.in/handle/123456789/18762
49. Deshong, P., Li, W., Kennington, J. W., Ammon, H. L. & Leginvs, J. M. (1991). A nitrone-based cycloaddition approach to the synthesis of the glycosyl system of nogalomycin, menogaril, and their congeners. *J Org. Chem*, 56, 1364–1373. DOI: https://doi.org/10.1021/jo00004a008
50. Gandolfi, R., & Grunanger, P. (1999). The chemistry of heterocyclic compounds (Wiley Interscience), 49(2), 774. https://ir.nbu.ac.in/server/api/core/bitstreams/c3ced9cc-0cb6-4e96-a62e-d7b3ec8c80ce/content
51. Yu, Y., Ohno, M., & Eguchi, S. (1993). Tetrahedron, 49, 824.
52. Huisgen, R. (1976). 1,3-Dipolar cycloadditions. 76. Concerted nature of 1,3-dipolar cycloadditions and the question of diradical intermediates. *J Org. Chem, 41*(3), 403-419. DOI: https://doi.org/10.1021/jo00865a001
53. Houk, K. N. & Yamaguchi, K. (1984). 1,3-Dipolar Cycloaddition Chemistry. Wiley, New York, 2, 407. https://link.springer.com/article/10.1007/BF00472551
54. Sanjai, C., Hakkimane, S. S., Guru, B. R., & Gaonkar, S. L. (2024). A comprehensive review on anticancer evaluation techniques. *Bioorg Chem*, 142, 106973. DOI: https://doi.org/10.1016/j.bioorg.2023.106973

55. Campoccia, D., Ravaioli, S., Santi, S., Mariani, V., Santarcangelo, C., De Filippis, A., Montanaro, L., Arciola, C. R., & Daglia, M. (2021). Exploring the anticancer effects of standardized extracts of poplar-type propolis: In vitro cytotoxicity toward cancer and normal cell lines. Biomedicine & Pharmacotherapy, 141, 111895. DOI: https://doi.org/10.1016/j.biopha.2021.111895
56. Cai, L., Qin, X., Xu, Z., Song, Y., Jiang, H., Wu, Y., Chen, J., & Ruan, H. (2019). Comparison of Cytotoxicity Evaluation of Anticancer Drugs between Real-Time Cell Analysis and CCK-8 Method. *ACS-Omega, 4*(7), 12036 –12042. DOI: https://doi.org/10.1021/acsomega.9b01142
57. Chakraborty, B. & Roy, M. N. (2022). ACS-Omega, 7, 4457.