

ADVANCES IN ISATIN DERIVATIVES FOR MULTIFACETED BIOLOGICAL ACTIVITY APPLICATIONS

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Abstract: Isatin is a heterocyclic compound which is widely used in drug discovery. It is widely recognized as a privileged scaffold in medicinal chemistry due to its unique structural features, broad spectrum of biological activities and synthetic versatility. Isatin shows remarkable pharmacological properties, including anticancer, antimicrobial, anticonvulsant, antidepressant and for Alzheimer's disease. It consists of two cyclic rings, one with five membered and other with six membered. The 5 membered ring has anti-aromatic property and 6- membered which is aromatic. The indole ring consist two carbonyl groups make it useful for the development of new therapeutic agents. Synthesis of isatin can be done by various methods and its reactivity shows that various different derivatives can be produced from isatin which can bind with various biological targets. The growing relevance of isatin in future drug discovery is largely attributed to its structural advantages and synthetic flexibility. The presence of multiple reactive sites enables easy functionalization at different positions of the scaffold, allowing researchers to generate diverse chemical libraries and optimize biological activity. Its planar aromatic structure promotes favorable π - π interactions with biological targets, while the carbonyl groups contribute to strong hydrogen-bonding interactions that enhance target binding. In addition, isatin can be readily incorporated into hybrid molecules containing other pharmacophores, facilitating the development of multitarget-directed therapeutics. Owing to its favorable drug-like characteristics, broad biological potential, and adaptability to modern medicinal chemistry strategies, isatin continues to serve as a valuable molecular platform for the discovery of innovative drugs aimed at addressing current and emerging healthcare challenges. In this review article, various synthetic methods of isatin, its reactivity, structure-activity relationships and various pharmacological activity profiles are established. This would provide a reference to newer researchers discovering new drugs.

Keywords: Isatin, 1H-indole-2,3-dione, Sandmeyer synthesis, Antiepileptics, Anticancer, Antimicrobial

INTRODUCTION

Isatin (1H-indole-2,3-dione) is an important heterocyclic compound belonging to the indole family and is widely recognized as a valuable pharmacophore in medicinal chemistry. Isatin was first identified in 1840 by Otto Linne Erdmann and Auguste Laurent during the oxidation of indigo dye^{[1][2]}. Although initially considered only a synthetic product, later investigations revealed its natural occurrence in several plants such as *Couroupita guianensis*, *Isatis tinctoria*, and *Calanthe discolor*, also in animals and humans. In the human body, dietary tryptophan is metabolized into indole by intestinal bacteria, which is subsequently oxidized in liver by the cytochrome P450 enzyme system to produce isatin, establishing it as an endogenous biomolecule^[3].

Structurally, isatin contains an indole nucleus having highly reactive carbonyl groups at the C-2 and C-3 positions, allowing extensive chemical modification and the synthesis of numerous derivatives. Because of this synthetic versatility, isatin is regarded as privileged scaffold in modern drug discovery. Functionalization at the N-1, C-3, C-5, and C-7 positions has enabled the development of compounds with diverse pharmacological profiles^[4]. Researchers have synthesized Schiff bases, spiro compounds, hydrazones, and fused heterocycles derived from isatin, many of which exhibit significant biological activities^{[5][6]}. The ease of structural modification also makes isatin suitable for multicomponent reactions, hybrid molecule synthesis, and environmentally friendly synthetic approaches^[6].

Research on biology has shown that isatin derivatives possess broad spectrum of therapeutic activities, including anticancer, antimicrobial, antiviral, anti-inflammatory, anticonvulsant, antioxidant, and antitubercular effects. These derivatives act on several important biological targets like topoisomerases, epidermal growth factor receptor, histone deacetylases (HDAC), carbonic

anhydrase, tyrosine kinases, cyclin-dependent kinases (CDKs), adenylate cyclase, and protein tyrosine phosphatase (Shp2)^{[7][8]}. Owing to these properties, numerous isatin-based molecules have investigated as a potential drug candidates for the cancer chemotherapy and infectious diseases^{[9][10]}. Different studies shown that suitable substitutions on the isatin framework can significantly enhance potency, selectivity, and pharmacokinetic behaviour^{[11][12]}.

Apart from its pharmaceutical importance, isatin also have a major role in biochemical and neurological processes. It has said to have an impact on neurotransmission, cellular signalling pathways, enzyme regulation, and oxidative stress mechanisms. In the nervous system, isatin exhibits neuromodulatory properties and interacts with dopamine, serotonin, and monoamine-related pathways, resulting in research into its potential connection to neurodegenerative disorders, anxiety, epilepsy, and Parkinson-like conditions^{[13][14][15]}. The molecule is also associated with redox biology because its electron-rich indole ring and diketone structure can participate in oxidation–reduction reactions^[16]. Interestingly, some isatin derivatives act as antioxidants, whereas others selectively induce oxidative stress on tumor cells, highlighting their importance for the development of targeted anticancer therapies^{[17][18]}.

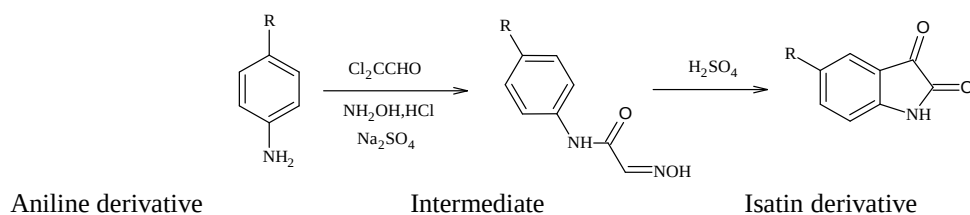
SYNTHESIS OF ISATIN

Common method of synthesis involves

1) Sandmeyer isatin synthesis

The Sandmeyer isatin synthesis is classical method to prepare isatin derivatives from aromatic amines. Aniline derivatives react with chloral hydrate and hydroxylamine hydrochloride to form an intermediate, which cyclizes in concentrated sulfuric acid to produce isatin.

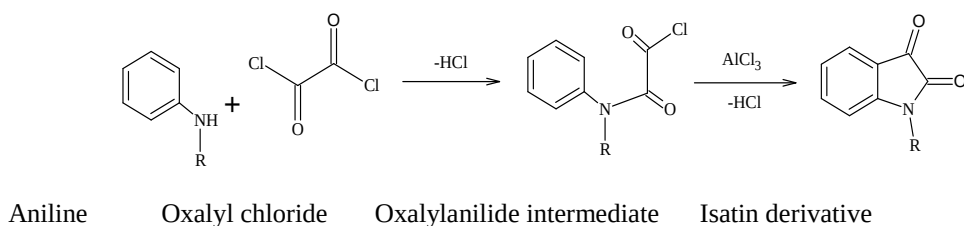
Aniline is first mixed with chloral hydrate and hydroxylamine hydrochloride in water. Sodium sulfate may be added to maintain reaction conditions. The mixture is heated gently, producing an intermediate called isonitrosoacetanilide through condensation and oxime formation. After cooling, slowly added concentrated sulfuric acid with stirring. The acidic medium promotes cyclization and rearrangement of the intermediate to form the heterocyclic isatin ring. Then the reaction mixture is poured onto ice, causing isatin crystals to precipitate. The product was filtered, rinsed with cold water, recrystallized from alcohol, and dried, resulting in pure isatin^[19].



2) Stolle Synthesis

The Stolle Synthesis is a method to prepare isatin derivatives from aniline and oxalyl chloride. The methodology reported by Stolle for the synthesis of isatin is an alternative to the Sandmeyer reaction.

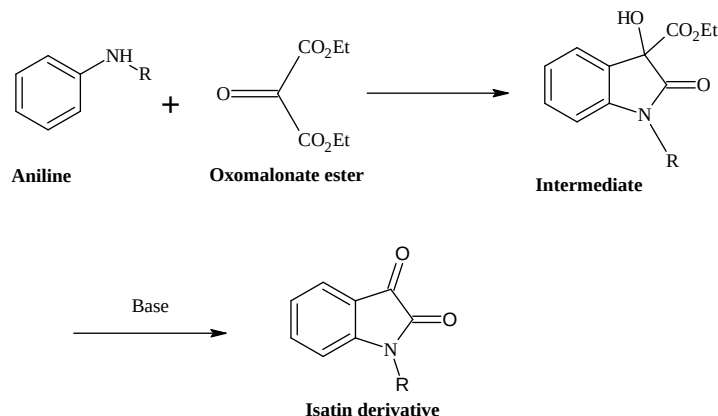
When a Lewis acid catalyst, like aluminium chloride, is present, aniline and oxalyl chloride react. Initially, oxalyl chloride hydrolyse the amino group to form an oxalylanilide intermediate. Under heating, intramolecular cyclization occurs, producing the isatin ring system. Then the reaction mixture is hydrolysed with water or dilute acid to complete isatin formation. Finally, product is isolated by filtration, purified through recrystallization, and dried to obtain pure crystalline isatin derivatives^[19].



3) Martinet isatin synthesis

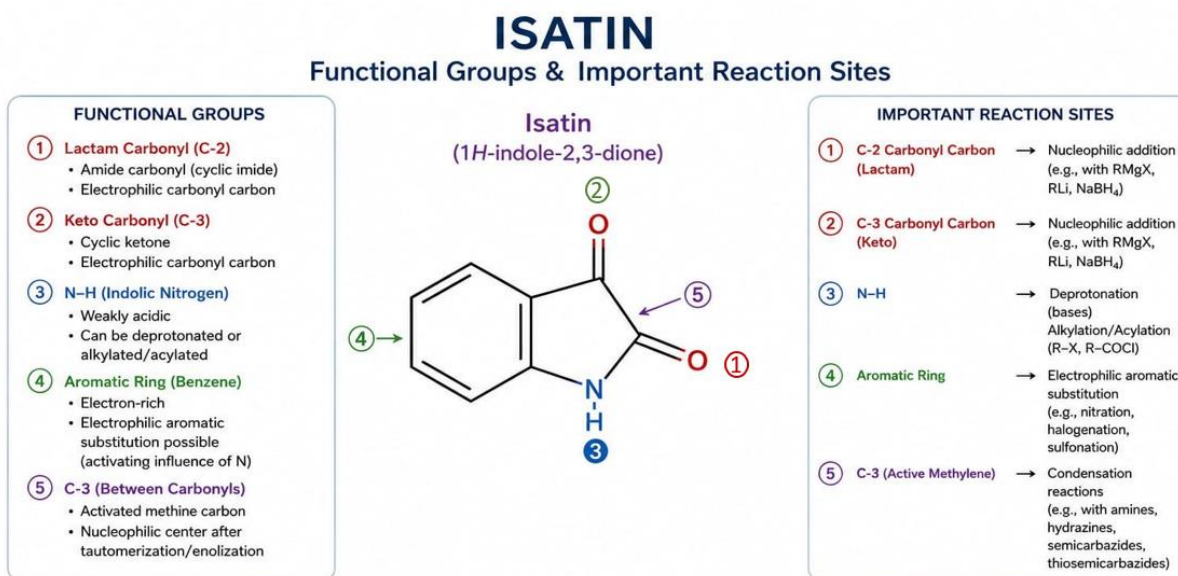
Martinet synthesis was reported by French chemist Henri Martinet. An aromatic amine reacts with an oxomalonate ester under acidic conditions to form isatin derivatives.

The Martinet Isatin Synthesis procedure involves heating an aromatic amine, such as aniline, with an oxomalonate ester in the presence of concentrated sulfuric acid. The amine first condenses with the ester to form an intermediate, which undergoes cyclization under acidic conditions. The cyclic product is then hydrolyzed and decarboxylated during continued heating to yield an Isatin derivative. The product is precipitated by cooling the reaction mixture and then pouring it into freezing water. The crude isatin is filtered, washed, and purified by recrystallization. This method is widely used for preparing substituted isatins in organic synthesis^[20].

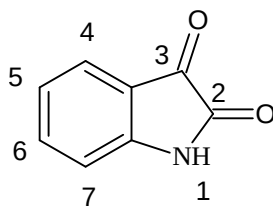


CHEMISTRY OF ISATIN

Isatin (1H-indole-2,3-dione) is an aromatic heterocyclic molecule having the chemical formula $C_8H_5NO_2$ and molecular weight is 147.13 g/mol. It contains benzene ring fused with a five-membered nitrogen containing ring with two adjacent carbonyl groups at positions 2 and 3^[21].



STRUCTURE-ACTIVITY RELATIONSHIP OF ISATIN



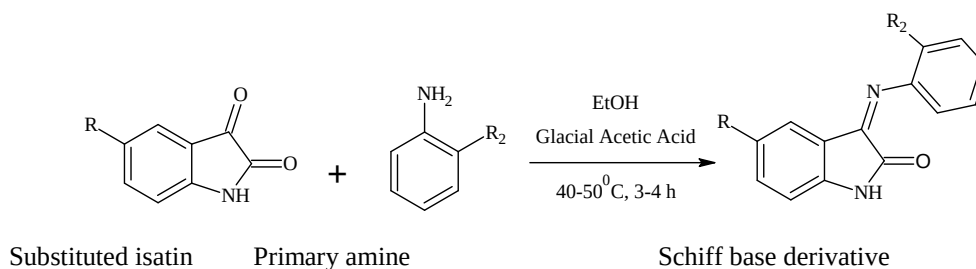
Isatin is an important heterocyclic scaffold containing an indole nucleus having a keto groups at 2- and 3- positions. Its biological activity is strongly influenced by substitutions at the nitrogen atom on the aromatic ring. SAR studies shows that modification at the N1 position significantly alters lipophilicity, membrane permeability and pharmacological activity. N-alkyl and N-benzyl substitutions generally enhance antimicrobial, anticancer and antiviral properties by increasing hydrophobic interactions with the biological targets. Substitution at C-3 carbonyl position through the formation of Schiff bases, hydrazones, oximes or spiro derivatives often improves antibacterial, anticonvulsant and anticancer activities. Electron withdrawing group such as halogens (Cl, Br, F) or nitro groups at the C-5 or C-6 positions usually enhance biological potency due to increased electron deficiency and better receptor binding^{[22][23]}. Electron-donating groups like methyl or methoxy may improve anti-inflammatory or antioxidant activity in some derivatives^[21]. Spiro-fused isatin derivatives frequently show enhanced anticancer and antimicrobial activity because of increased molecular rigidity and better interaction with enzymes or receptors^[24]. Maintaining biological activity requires the intact indole ring system, but extensive substitution can lower activity because of steric hindrance. Thus, the biological profile of isatin derivatives can be optimized by strategic substitution at the N-1, C-3, C-5, and C-7 positions^{[25][26]}.

REACTIONS OF ISATIN

Schiff base reaction

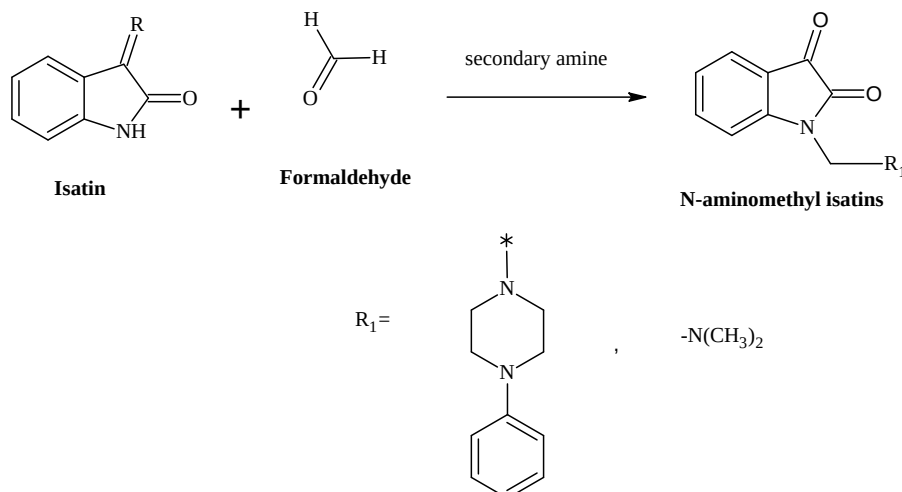
A Schiff base is an organic compound with an azomethane group which is formed by the condensation reaction between the C-3 carbonyl group of isatin with primary amines, hydrazines or thiosemicarbazides.

An equimolar amount of isatin and the appropriate primary amine dissolved in ethanol in a round-bottom flask. 2-3 drops of glacial acetic acid was then added as a catalyst, and mixture was refluxed under continuous stirring for 3–4 hours. Upon completion, mixture was allowed to cool to room temperature, leading to solid precipitate formation. The precipitated product was collected by filtration, washed using cold ethanol to remove residual impurities, and purified by recrystallization from ethanol or methanol to afford the pure Schiff base derivative^[27].



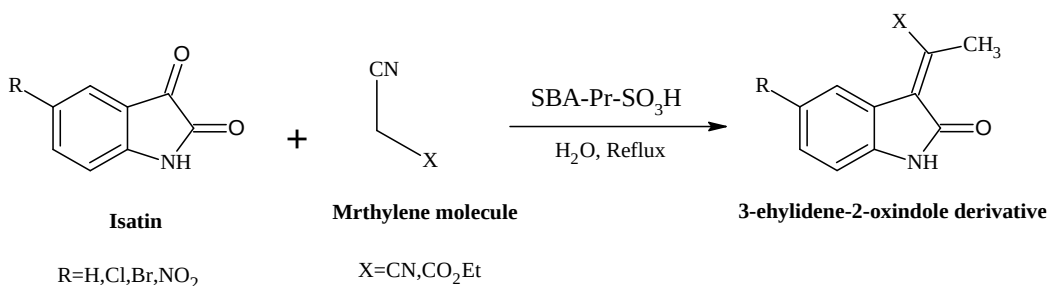
Mannich base reaction

The Mannich reaction can be conveniently carried on with Isatin and its derivatives. This reaction commonly leads to formation of N-aminomethyl isatins, which are generally referred to as Mannich bases. These compounds can also be synthesized through reactions involving amines or acetyl chloride to form N-chloromethylisatin as an intermediate. The intermediate subsequently react with potassium phthalimide or alcohols to yield N-phthalimidomethyl and N-alkoxymethyl isatin derivatives, respectively. In addition, several isatin derivatives, including isatin-3-hydrazones and isatin-3-thiosemicarbazones, are also capable of undergoing Mannich reactions successfully^{[28][29]}.



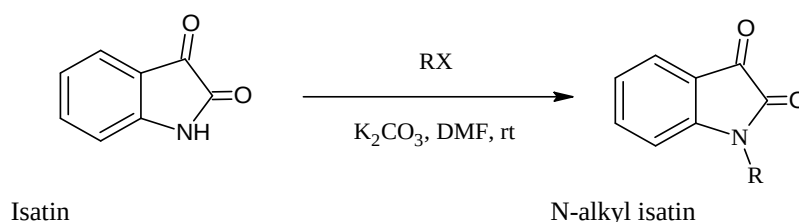
Knoevenagel condensation

Knoevenagel condensation of isatin involves dissolving isatin and an methylene molecule that is active such as malononitrile or ethyl cyanoacetate in presence of SBA-Pr-SO₃H in aqueous medium. Until the reaction is finished, the mixture is refluxed while constantly stirred for several hours^{[30][31]}.



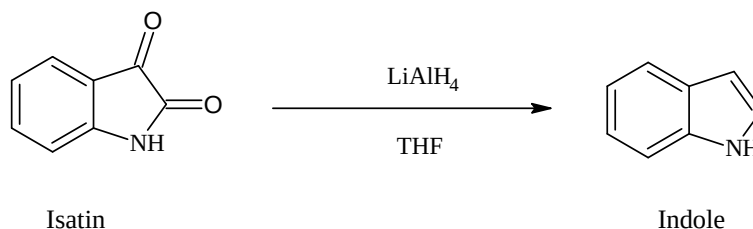
N-alkylation

Isatin undergoes N-alkylation when the hydrogen atom attached to its nitrogen atom is replaced by an alkyl group such as methyl, ethyl, or benzyl. Isatin react with alkylhalides in the presence of K₂CO₃ and DMF then stirred for 24 hours at the room temperature. DCM was then utilized to extract reaction mixture after it had been diluted with water. The combined organic layer were dried, concentrated under low pressure, and cleaned with brine^[32].



Reduction of isatin

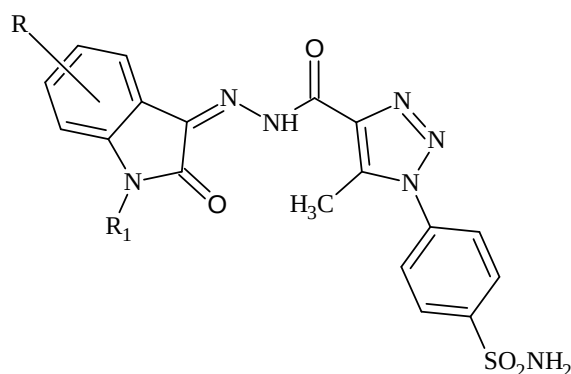
Isatin undergoes reduction in presence of Lithium aluminium hydride in dry tetrahydrofuran (THF) under an inert atmosphere of nitrogen or argon to yield indole. A round-bottom flask with reflux condenser and nitrogen inlet is charged with anhydrous THF and LiAlH₄. Then reaction mixture is cooled in ice bath, and solution of isatin in dry THF is added slowly by continuous stirring because reaction is exothermic. Following full addition, mixture is refluxed for few hours, during which LiAlH₄ lowers isatin's carbonyl groups to create intermediates that go through aromatization and deoxygenation to produce indole. After completion, the reaction mixture is cooled and carefully quenched with water or saturated ammonium chloride solution to destroy excess reagent. Aluminium salts are filtered off, and organic layer is extracted, dried, and concentrated to obtain indole, which is purified by recrystallization or chromatography^[33].



PHARMACOLOGICAL ACTIVITIES OF ISATIN

Antiepileptic activity

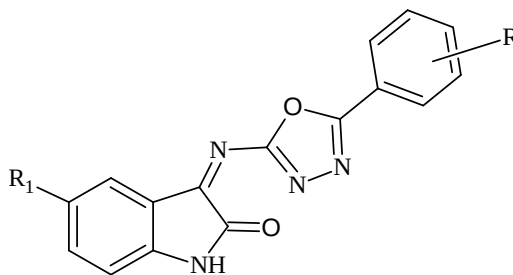
Wagdy M. Eldehna *et al*; synthesised isatin-based benzenesulfonamide derivatives (5a-f and 7a-e) as a dual inhibitors of human carbonic anhydrase II and VII for seizure treatment. The compounds were designed using a structure-guided one-tail approach that incorporated a sulfonamide zinc-binding group, a triazole linker derived from the antiepileptic scaffold of Rufinamide, and an isatin moiety. With compounds 5c, 5e, 5f, 7a, and 7d exhibiting nanomolar inhibitory action against hCA II/VII, this approach yielded extremely powerful molecules. Their anticonvulsant effects were assessed using Pentylentetrazole-induced and Pilocarpine-induced seizure models^[34]. Among the tested compounds, 5e and 7a demonstrated the strongest seizure protection, providing complete protection in the PTZ model and significantly delaying seizure onset while reducing seizure severity and mortality in the pilocarpine model, with better efficacy than Valproic acid. Biochemical studies showed restoration of KCC2 and mTOR levels, indicating stabilization of neuronal excitability and ionic balance. Toxicity studies confirmed the absence of major neurotoxic, hepatic, renal, or cardiac adverse effects, supporting further long-term investigation of compounds 5e and 7a for epilepsy treatment^{[35][36]}.



Isatin-based benzenesulfonamide derivatives

Code	R	R ₁
5e	5,7-(CH ₃) ₂	H
7a	5,7-(CH ₃) ₂	CH ₂ C ₆ H ₅

Rajarshi Nath *et al*; has designed and synthesized series of N-(substituted benzothiazole-2-yl)-2-(2,3-dioxindolin-1-yl)acetamide derivatives (4a-i) and substituted-[3-((5-phenyl-1,3,4-oxadiazole-2-yl)imino)indolene-2-one] compounds (5a-f) rationally based on pharmacophoric requirements. These compounds were evaluated for anticonvulsant efficacy through the maximal electroshock seizure (MES) and subcutaneous pentylentetrazole (scPTZ) models. Their neurotoxicity was also examined in mice using a motor coordination impairment test^[37]. Among all the synthesized compounds, N-(5-chlorobenzo[d]thiazol-2-yl)-2-(2,3-dioxindolin-1-yl)acetamide (4a) demonstrated the highest anticonvulsant efficacy in both MES and scPTZ models. The compound exhibited ED50 values of 35.7 mg/kg in MES test and 88.15 mg/kg in the scPTZ test, while its TD50 value exceeded 500 mg/kg, indicating low toxicity. Furthermore, *in silico* analysis, including molecular docking studies, which performed to investigate the interaction of compound 4a with Na⁺ channels and GABA_A receptors^[38].

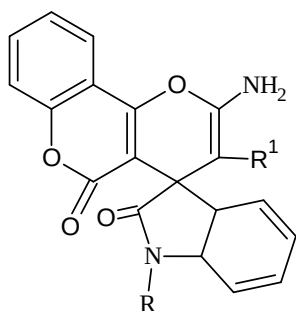


N-(substituted benzothiazole-2-yl)-2-(2,3-dioxindolin-1-yl)acetamide derivatives

R= H, 2-NO₂, 4-NO₂, 4-OCH₃

R₁= H, Br, Cl

Leila Emami *et al*; designed and synthesized novel heterogeneous catalyst based on graphene oxide which demonstrated excellent efficiency in producing spirooxindole-4H-pyran derivatives in ethanol, providing high product yields. This green synthetic approach gives several benefits, including the use of ethanol as a safe solvent, simple workup procedures, low production cost, catalyst recyclability, and product purification without chromatographic techniques. Nitrogen-containing groups having lone electron pairs enabled the catalyst (GO-A.P.) to function as a Lewis base, while EDS and FT-IR analysis confirmed the incorporation of nitrogen atoms within its structure. Fifteen synthesized spirooxindole-4H-pyran derivatives tested for anticonvulsant activity utilizing a PTZ-induced seizure model in mice. Most compounds significantly increased seizure thresholds, particularly A₃ and A₁₂, which showed the strongest activity along with favourable physicochemical and drug-likeness properties. Molecular docking studies targeting the GABA_A benzodiazepine receptor complex revealed good agreement between docking scores and anticonvulsant activity^[39].



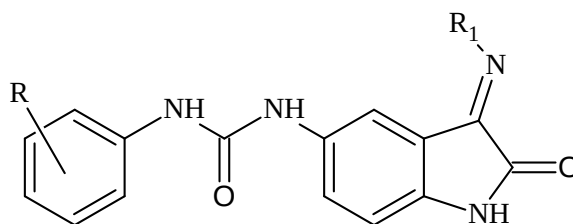
A₃: R=3-methylbut2-en, R¹=CN

A₁₂: R= sec-butyl, R¹= COOC₂H₅

Spirooxindole-4H-pyran derivatives

Anticancer activity

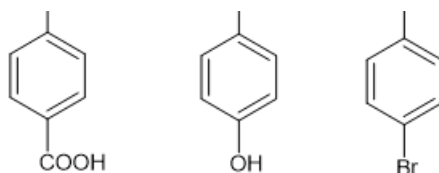
Noor Waleed Ibrahim *et al*; synthesized two series of compounds derived from 5-aminoisatin Schiff bases (IIa–c), namely IIIa–c containing a single methyl (CH₃) group and IVa–c containing two methyl groups. The effect of compounds IIIa–c and IVa–c on the human A549 cancer cell line, evaluated using the MTT colorimetric assay. Among the tested derivatives, compounds IIIa and IVa shown significant inhibitory efficacy against EGFR tyrosine kinase, with compound IIIa exhibiting an inhibition percentage comparable to the reference drug Erlotinib. These biological discoveries aligned with the GOLD program's molecular docking results. In addition, ADME evaluation showed that synthesized compounds (IIIa–c and IVa–c) complied to Lipinski's rule of five and showed favourable gastrointestinal absorption^{[40][41]}.



Isatin based phenylurea hybrid

III a-c & IV a-c

R₁=

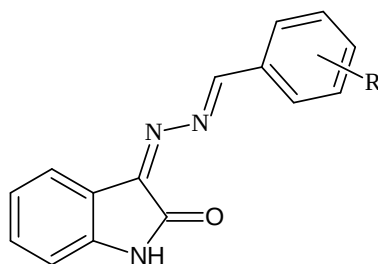


R =

3-CH₃(III), 2&6-diCH₃(IV)

Huda S. Al-Salem *et al*; has designed and synthesized series of novel isatin-hydrazone derivatives (4a–b and 4d–k), along with the previously reported compound 4c to investigate their cytotoxic potential as anticancer agents. From the synthesized compounds, derivative 4j demonstrated approximately two-fold greater cytotoxic activity than the reference anticancer drug Doxorubicin, while compound 4k exhibited comparable activity. The IC₅₀ values against human MCF7 cell lines was 1.51 µM for compound 4j and 3.56 µM for compound 4k, compared with 3.1 µM for doxorubicin^[42].

The most active derivatives, 4j and 4k, further assessed for their capability to inhibit cyclin-dependent kinase 2 (CDK2). Both compounds exhibited inhibitory activity, having IC₅₀ values of 0.2456 µM and 0.3006 µM, respectively, which were close to that of Imatinib (0.1512 µM). In addition, ADME predictions indicated more favorable pharmacokinetic properties of these compounds relative to doxorubicin. Molecular docking analysis further shows that compounds 4j and 4k interacted effectively with residues within the ATP-binding pocket of CDK2, while lacking interactions with residues of the active-state DFG motif. These results shows both derivatives may act as ATP-competitive type II inhibitors of CDK2 protein kinase^[42].

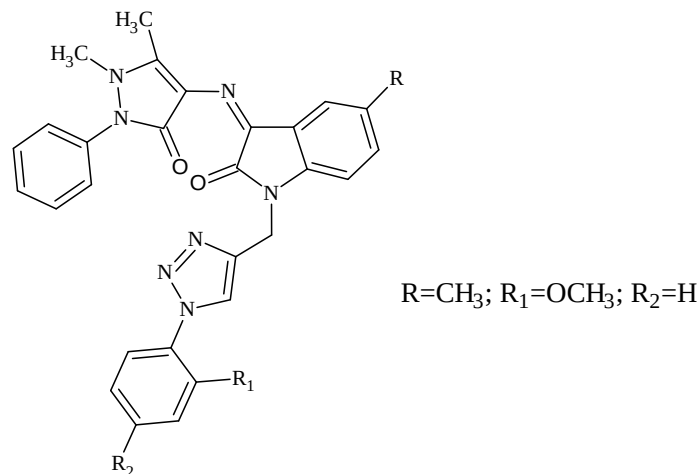


Isatin-hydrazone derivatives

4j = 2,6-di-Cl

4k = 2Cl, 6-F

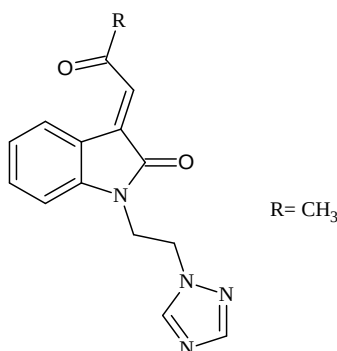
Israa A. Seliem *et al*; employed molecular hybridization approach to synthesize three series of isatin-based Schiff base derivatives, which then assessed for the anticancer activity against colon, breast, and cancerous pancreatic cell lines, including MCF7, HCT116, and PaCa-2. Several compounds showed moderate to strong antiproliferative effects when compared to standard drugs 5-fluorouracil and sunitinib. Among all the synthesized molecules, compound 17f emerged as most active, displaying 2.1-fold higher activity against the MCF7 breast cancer cell line than sunitinib. Additionally, some 5-methylisatin derivatives exhibited more better inhibitory activity against the HCT116 colon cancer cells than 5-fluorouracil. Compound 17f also showed pronounced anti-angiogenic properties by significantly suppressing blood vessel formation and inducing G1/S phase cell-cycle arrest, as confirmed by flow cytometry analysis. It further demonstrated strong inhibition of VEGFR-2, indicating its potential as a promising anti-breast cancer agent. These biological findings were strongly supported by detailed 2D-QSAR studies^[43].



Isatin-based Schiff base derivatives

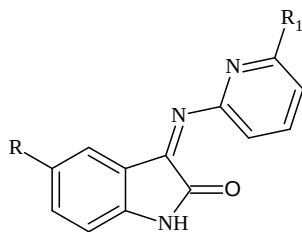
Antimicrobial activity

Vijai Kumar Reddy Tangadanchu *et al*; has developed isatin derived azoles molecules as new potential antimicrobial agents. Molecular hybridization has recently emerged as very promising strategy in drug discovery, attracting significant attention from medicinal chemistry for its potential to overcome the growing challenge of drug resistance. The isatin-azole hybrids were efficiently synthesized via multistep reactions starting from the commercially available isatin. Bioactivity assays demonstrated that the isatin–1,2,4-triazole hybrid compound 7a showed inhibitory activity to *Escherichia coli* ATCC 25922, with MIC of 1 µg/mL, making it 8 times more potent than standard drug Norfloxacin. Compound 7a was also capable of eliminating various fungal and bacterial strains while exhibiting a low tendency to promote resistance in *E. coli* ATCC 25922. According to preliminary mechanistic research, hybrid 7a may contribute to its antimicrobial efficacy by intercalating into DNA and perhaps interacting with the DNA polymerase III to prevent DNA replication^{[44][45]}.

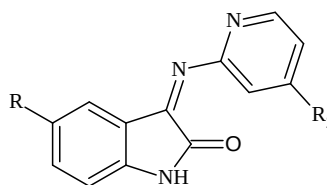


Isatin derived azoles molecules

Richa Mishra *et al*; has designed and synthesized novel series of isatin- based derivatives containing imine linkage and assessed for antimicrobial activity. These derivatives tested against several bacterial strains, including *Escherichia coli*, *Pseudomonas aeruginosa*, *Bacillus cereus*, and *Staphylococcus aureus*, as well as fungal pathogens such as *Fusarium oxysporum* and *Aspergillus flavus*. Various derivatives exhibited strong antibacterial activity, particularly against *E. coli*, outperforming standard antibiotics such as Chloramphenicol and Ciprofloxacin. Combining the compounds with these antibiotics showed significant synergistic effects, lowering MIC values by up to 32 times. QSAR analysis identified molecular descriptors such as ALOGP2, VP7, and chi descriptors as important contributors for biological activity. A potential mechanism of action was suggested by molecular docking studies that showed persistent contacts between the produced ligands and peptide deformylase enzyme. In antifungal studies, compounds 8 and 10 provide notable inhibitory activity against *F. oxysporum*. Furthermore, ADMET predictions indicated favourable intestinal absorption, oral bioavailability, and cell permeability^[46].



8; R=Br, R₁=F

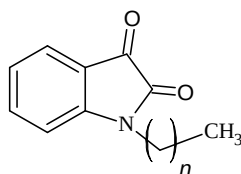


10; R=Br, R₂=OH

Isatin-pyridine schoff base

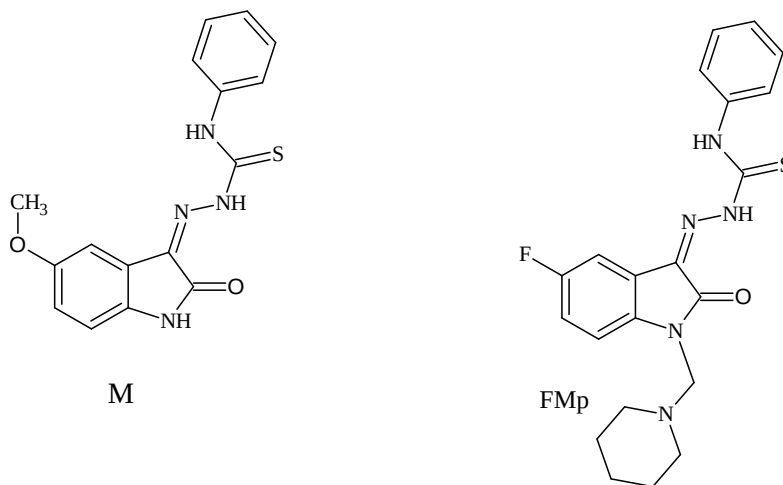
Alzheimer's disease

Kaitlyn N. Alcorn *et al*; has synthesized two series of N-alkyl substituted isatins and indoles with varying alkyl chain lengths and assessed for acetylcholinesterase and butyrylcholinesterase inhibitory activity. Among the N-alkyl isatin derivatives (4a–j), incorporation of the N-alkyl substituent enhanced inhibitory activity toward both enzymes when compared with the parent isatin compound. These derivatives displayed greater selectivity for BChE inhibition, and increasing the alkyl chain length further improved their potency. Compound 4i emerged as most effective BChE inhibitor, exhibiting IC₅₀ value of 3.77 μM and 22-fold selectivity over AChE. In contrast, the N-alkyl indole derivatives (5a–j) demonstrated moderate inhibition of AChE, with compound 5g showing the strongest activity, but they exhibited little to no activity against BChE. These findings points out the critical role of the 3-oxo group present in the isatin scaffold for effective BChE inhibition. Molecular docking studies of 4i compound with human BChE further revealed that this group forms essential hydrogen bond interactions with Ser198 and His438 residues within the enzyme's catalytic triad, contributing significantly to its inhibitory activity^[47].



N-alkyl substituted isatins

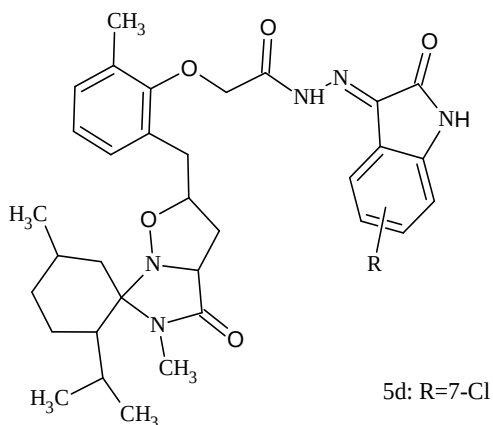
Barbara Mavroidi *et al*; investigated the multimodal therapeutic potential in two synthetic isatin thiosemicarbazones (ITSCs), previously identified as a inhibitors of Aβ aggregation. Alzheimer's disease is a complex neurodegenerative disorder characterized by multiple pathological processes, including formation of amyloid-β (Aβ) oligomers that aggregate into insoluble amyloid plaques, oxidative stress, neuroinflammation, and elevated acetylcholinesterase activity. Additionally, Aβ oligomers disrupt activity and expression of key signaling proteins such as glycogen synthase kinase-3 (GSK-3) and protein kinase B (PKB/Akt). Both compounds completely prevented Aβ-induced toxicity in SK-N-SH cells when administered alongside exogenous Aβ peptides at different pre-incubation intervals and at concentration of 1 μM. However, they failed to restore cell viability when applied after Aβ exposure. The compounds also showed no toxicity toward either wild-type or 5xFAD primary hippocampal cells. Furthermore, ITSC treatment reversed the Aβ-associated reduction in Akt and GSK-3β phosphorylation in 5xFAD cells. In addition, the compounds demonstrated strong antioxidant activity and moderate inhibitory effects against lipoxygenase and acetylcholinesterase. These findings indicate that isatin thiosemicarbazones represent promising scaffolds for the development of multifunctional therapeutic agents against AD^[48].



Isatin thiosemicarbazones

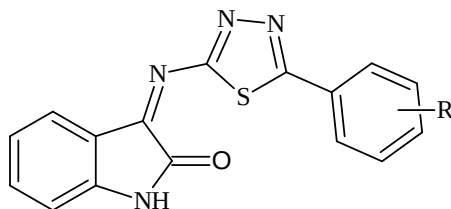
Antidiabetic activity

Siwar Ghannay *et al*; a new class of isoxazolidine–isatin hybrid compounds was successfully designed and synthesized, showing diverse and promising inhibitory effects against the enzymes α -amylase and α -glucosidase. From the synthesized derivatives, compound 5d demonstrated the strongest inhibitory activity, with IC_{50} values of $30.39 \pm 1.52 \mu M$ against α -amylase and $65.1 \pm 3.11 \mu M$ against α -glucosidase. Enzyme kinetic studies revealed that compound 5d shows similarity to standard drug acarbose, competitively interacted with active sites of both enzymes, effectively blocking substrate binding. Structure–activity relationship (SAR) highlighted the significant role of substituent groups attached to the isatin moiety in enhancing biological activity. Additionally, by validating favorable pharmacokinetic and safety characteristics, computer-assisted ADMET predictions bolstered compound 5d's prospects as a prospective therapeutic candidate. The stable binding of compound 5d within the active pockets of both target enzymes was demonstrated by molecular docking and molecular dynamics modeling studies, which also showed a strong correlation with the experimental results. The docking scores of -6.602 kcal/mol for the 5d–2QV4 complex and -5.414 kcal/mol for the 5d–3W37 complex further confirmed its strong affinity and identified it as a potential lead pharmacophore for future therapeutic development^[49].



Isoxazolidine–isatin hybrid compounds

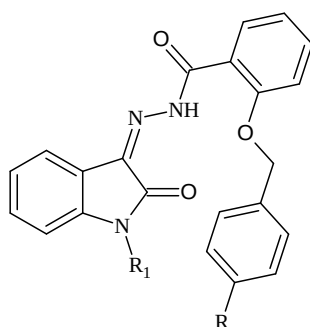
Monika *et al*; investigated the development and computational assessment of new thiadiazole–isatin hybrid derivatives as potential candidates for diabetes treatment. Building on the established antidiabetic properties of both thiadiazole and isatin moieties, six hybrid compounds (2a–2f) were designed, synthesized, and evaluated using molecular docking studies, ADME analysis, physicochemical characterization, and Lipinski's rule of five to determine their drug-likeness. The synthesis involved the formation of thiadiazole intermediates from thiosemicarbazide, which subsequently coupled with various isatin derivatives to generate the target molecules. Computational docking against important diabetes-associated targets, including human aldose reductase, monoglyceride lipase, glucagon-like peptide-1 (GLP-1), and α -amylase, identified compounds 2a and 2b as most promising derivatives. These compounds show strong binding affinities, with docking scores from -10.6 to -7.0 for 2a and -10.2 to -7.0 for 2b, suggesting effective interactions with multiple enzymes involved in glucose regulation and diabetic complications. In addition, both compounds met Lipinski's drug-likeness criteria and demonstrated favorable ADME profiles, indicating suitable pharmacokinetic characteristics. Among the evaluated derivatives, compound 2a showed slightly superior activity and overall drug-like behavior compared to compound 2b. Collectively, the results suggest that thiadiazole–isatin hybrid framework represents a promising approach for the discovery of multi-target antidiabetic agents^[50].



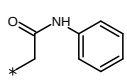
Thiadiazole–isatin hybrid

Antidepressant activity

Azadeh Mesripour *et al*; has designed and synthesized isatin derivatives with Schiff bases. The synthesis is started by N-alkylation and N-benzylation of isatin which is succeeded by the reaction with hydrazine hydrate which provide acid hydrazide derivatives. The final compound obtained by condensation of N-substituted isatin. Compounds 8b and 8e in both 25 mg/g and 50 mg/kg doses, and 8c and 8d only in 25 mg/kg dose had antidepressant activity. Compounds are evaluated for antidepressant activities in mice by locomotor activity, marble burying test, and forced swimming test. Monoamine oxidase–A (MAO–A) enzyme has used for molecular docking studies^[51].



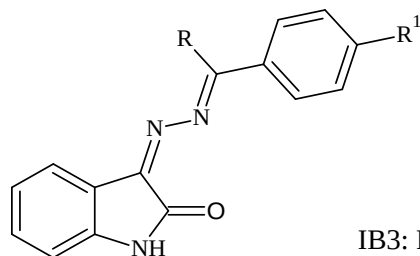
N- substituted isatin benzohydrazone

Compound	R ₁	R
8b		H
8c	CH ₂ -COOEt	H
8d		Cl
8e		Cl

Naseer Maliyakkal *et al*; This study focused on the synthesis and biological assessment of ten isatin-based hydrazone derivatives designed as the inhibitors of monoamine oxidase enzymes, namely MAO-A and MAO-B, which are important targets in neurological disorder treatment. Compounds were classified into two series: IA (isatin + acetophenone) and IB (isatin + benzaldehyde). In general, MAO-A was more strongly inhibited by all produced compounds than MAO-B, and the IB series exhibited better inhibitory efficacy than the IA series.

Among all compounds, IB4 exhibited most potent MAO-A inhibition with IC₅₀ value of 0.015 μ M, followed closely by IB3 with an IC₅₀ of 0.019 μ M. For MAO-B inhibition, IB3 was the most effective compound. Structural evaluation revealed that methyl substitution in the IA series reduced inhibitory activity, whereas halogen substitutions such as 4-chloro (IB3) and 4-bromo (IB4) significantly enhanced enzyme inhibition.

Kinetic studies confirmed that IB3 and IB4 acted as competitive and reversible inhibitors for both MAO isoforms. Molecular docking and dynamic simulations further demonstrated stable interactions between the compounds and enzyme active sites, particularly through hydrogen bonding with Asn181. Results demonstrate IB3 and IB4 are viable options for creating therapies for neurological conditions including depression^[52].



IB3: R=H, R¹=Cl

IB4: R=H, R¹=Br

Isatin-based hydrazone derivatives

CONCLUSION

Isatin is a heterogeneous molecule with a broad variety of biological activities. A unique scaffold for pharmacological and medical applications. Isatin is a useful scaffold in medical and pharmaceutical chemistry because it has antiepileptic, anticancer, antibacterial, antidiabetic, antidepressant, analgesic, and anti-tubercular properties. This study provides various methods for developing and synthesizing isatin and isatin derivatives. A lot of research has been done on isatin-based reactions to create new compounds having improved biological activities because of its distinct chemical structure and high reactivity. The creation of sustainable and environmentally friendly synthetic techniques for isatin derivatives has received more attention in an effort to get around the drawbacks of traditional synthesis. All things considered, isatin's many therapeutic uses and synthetic adaptability make it an essential foundation for further drug development and biological study.

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