COMBRETASTATINS: POTENTIAL TUBULIN INHIBITORS

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ABSTRACT

Combretastatin is a tubulin inhibitor that distinguishes from many other naturally occurring, synthetic, and semi-synthetic tubulin inhibitors discovered in the past. Several pro-drugs have undergone thorough clinical testing as vascular targeting medicines, such as combretastatin A4 phosphate derived from combretastatins (Vascular disrupting Agents). The clinical study of combretastatins as Vascular Disrupting Agents was prompted by their improved selectivity towards endothelial cells as well as their intrinsic capacity to swiftly cause vascular shutdown and inhibit tumour development. The molecular target of combretastatins and the huge majority of their tubulin, synthetic variants, is well known. Disruption of the microtubule by combretastatins that target it reduces cell growth and causes aberrant mitotic spindles in the G2-M phase of cell cycle. Combretastatin A-4 inhibits tubulin polymerization by engaging with the microtubule's colchicine binding site and has shown a significant lethal impact in a broad range of preclinical tumour types. By altering the dynamics of tubulin, such as depolymerization and polymerization combretastatin has an impact. Numerous analogues made from combretastatin have also been created due to their structural simplicity and have shown strong promise as inhibitors of tubulin polymerization. In this review, the pharmacological properties of combretastatin as a category of effective antimitotic anticancer agents are presented and discussed.

Keywords: Antitumour agents, Combretastatin, Microtubule, Tubulin, Tubulin inhibitor.
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Introduction

Tubulin, a biological protein, is essential for the reproduction of cells. When DNA is copied and chromosomes are packaged into two new cells, the cell cycle is complete and the process repeats. Microtubules, cylindrical organelles with a hollow core of around 5 nm and a diameter of 25 nm, are responsible for separating the daughter chromosomes during mitosis. Microtubules are made up of /tubulin heterodimers, with each monomer weighing in at 55 kDa in terms of mass. Tubulins polymerize into microtubules after binding to GTP and going through a series of processes. Eukaryotic cells include microtubules, dynamic structures that play a critical role in cell division.

Microtubule dynamics are tightly regulated during mitosis, causing cell death by apoptosis if microtubule polymerization or depolymerization is interrupted. Antimitotic antitumor medicines are based on ligands that disturb the system's intrinsic dynamic instability. These ligands have three unique small-molecule binding sites. Taxoid binding area on the polymerized microtubule, colchicine site, and vinca alkaloid domain on monomeric unpolymerized tubulin are all examples. Vinblastine, vincristine, and combretastatin A-4 are all ligands that interfere with tubulin polymerization, whereas ligands that interact with the taxoid binding site (like epothilone and taxol) stabilise microtubule polymerization. These ligands prevent cell division by interfering with the microtubule system's dynamic instability[1]. Antitumor agents are known as antitumor, anticancer, chemotherapeutic, or antimetastatic agents because they prevent or decrease the creation or growth of tumours. Cancer cells with a high rate of cell division are particularly vulnerable to antitumor drugs like Coumarin, Combretastatin, and their substituted derivatives.

Naturally occurring stilbenoid phenols such as Combretastatins are powerful and reversible tubulin polymerization inhibitors which are secluded from the African willow species Combretum caffrum[2]. Many cancer cell lines, even drug-resistant ones, are sensitive to CA-4 (combretastatin A-4). This is because it inhibits tubulin polymerization and is an effective cytotoxin[3]. CA-4 showed an IC50 value of 2-3 M in a tubulin polymerization experiment, indicating that it was effective. Inhibiting tubulin-colchicine binding, the compound had a Ki value of 0.14 M. The murine L1210 leukaemia cell line was also shown to be sensitive to CA-4, which had an ED50 value of 7 nM and was shown to be a powerful inhibitor of microtubule assembly. In contrast to what its activity against tubulin would seem to indicate, CA-4 is far more cytotoxic, and four theories have been put out to describe this paradox[4]: (i) CA-4 detects a pertinent target in addition to tubulin; (ii) The way tubulin and CA-4 interact is fundamentally different; (iii) In vivo, CA-4 targets a subset of tubulin; and (iv) Currently undiscovered mechanisms underlie CA-4's rapid and selective disruption of endothelial tubulin-dependent cellular activities in vivo. Combretastatin family molecules often have three structural characteristics in common: a trimethoxy A ring, a B ring with substituents frequently at C3' and C4', and an ethane bridge between the two rings that gives the requisite structural stiffness. Research conducted by Koji Ohsumi et al. found that one of the water-soluble amino acid prodrugs of amino-combretastatins (1), had increased toxicity, enhanced solubility, and ease of formulation making it suitable for administration in vivo[5]. CA-4 phosphate is a term for the cis-stilbene CA-4 water-soluble prodrug (2, CA-4P). Several preclinical studies have indicated that CA-4P reduces blood supply to tumour cells, which results in their mortality[6,7].

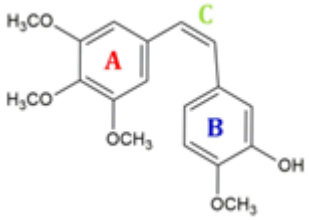


Fig no. 1: Structure of Combretastatin

Alterations to CA-4's A- and B-ring substituents have been documented, among other things. Even so, replacing the 30 -OH with an amino group boosts the bioactivity and enhances the water solubilization in contrast to the majority of B-ring modifications, which have been proven to diminish bioactivity. With this water-soluble amino acid prodrug of CA-4, clinical trials are undertaken. CA-4's bioactivity is improved by substituted pattern of 3,4,5-trimethoxy in ring A, which mimics the colchicine's trimethoxyaryl ring. A heterocyclic rigid ring scaffold structure is present in certain conformationally hindered analogues of CA-4 in place of the cis-double bond. It has been discovered that the potent colchicine binding inhibitor CA-4 also inhibits the growth and development of blood vessels[8]. CA-4's cytotoxicity may potentially be explained via mitogen-activated protein kinases. Antitumor effects of benzothiazoles are particularly noteworthy since they are an essential therapeutic scaffold with a variety of pharmacological actions. Some 6-substituted 2-phenyl benzotriazole derivatives have demonstrated promising anticancer potential, according to reports in the literature. ERK phosphorylation has also been demonstrated to be inhibited by several benzothiazole derivatives as a substructure. The cytotoxicity of CA-4 has been linked to its effects on the MAPK family of proteins and benzothiazole derivatives, which suppress ERK phosphorylation, according to one study. Hall JJ rt al. 2008 concluded that synthesised novel conjugates of combretastatin based on the promising anticancer profile of combretastatin and benzothiazole scaffolds and their influence on MAP kinases[9].

Combretastatin as VTA agent

Clinic investigation of combretastatins is mostly being done as vascular targeting/disrupting agents. Thorpe et al.2004 provided a detailed description of the clinical characteristics of a VTA[10]. Basically, a VTA destroys already-existing tumour blood arteries, which results in tumour ischemia and hemorrhagic necrosis, which kills tumour cells. Siemann et al., 2004 revealed that VTAs are an appealing anti-cancer drug because of their quick (within minutes) and tumour-type-independent actions. Because they target tubulin, which is crucial for providing the juvenile tumour vasculature with structural support, microtubule targeting medicines (MTAs) are excellent candidates for VTAs[11.] Tubulin targeting VTAs cannot destroy mature vasculature because of the developed basement membrane, actin filaments and vessel-associated pericytes give additional structural support. It is widely known that other MTAs, such as vinblastine and colchicine, may suppress the development of tumours via a vascular mechanism[12]. Vinca alkaloids and colchicine were not clinically studied as a VTA because of their toxicity and small therapeutic window[13.] Watts et al. 1997, originally discovered that CA-1 increased endothelial permeability, indicating that this family of drugs might eventually target tumour vasculature, resulting in tumour necrosis. Watts et al., A decade later, when it was shown that concentrations of CA-4P 10 times lower than the maximum tolerated dose (MTD) may reduce tumour blood flow, clinical studies of CA-4 as an antiangiogenic agent were initiated[14].

Combretastatin mechanism of action

Stabilizing or destabilising the microtubule system is one of the numerous anticancer medicines that target microtubule dynamics that have developed throughout time. These drugs have two different interactions with tubulin. Two examples are the colchicine binding domain and the vinca binding domain [15.] Drugs that bind to the colchicine domain include podophyllotoxin, combretastatin, and colchicine. Vincristine and vinblastine are, as

their names imply, vinca domain binding medicines. These first-generation medications have semi-synthetic derivatives as well. Paclitaxel, docetaxel, and other comparable molecules may attach to a taxane binding site that is not entirely separate from the colchicine binding site. There have been two taxane domains found, and they are not too far apart. Taxanes and medicines that bind to the colchicine domain function in the reverse direction of tubulin polymerization, which is interesting since they might be part of the colchicine domain [16].

The substances that bind to either the vinca or colchicine domains are not in direct competition with one another. It is necessary for tubulin polymerization that heterodimers interacting with each other be square. The pro-filaments' curvature is preserved when medicines are bound to them. As a result of the tubulin subunits' inability to return to a straight configuration after binding colchicine, microtubule assembly is inhibited. The colchicine binding sites are targeted by combretastatin, which impairs microtubule assembly. A significant role in tubulin polymerization is played by the colchicine binding pocket, and several medicines that bind to this pocket have been proposed as tubulin polymerization inhibitors. For combretastatin compounds, tubulin affinity is tightly linked with effectiveness. A tubulin-disrupting drug like combretastatin may prevent proliferative process of the cell during the G2/M phase of cell cycle because it depolymerizes tubulin assembly. Activation of apoptosis-inducing pathways occurs in conjunction with this process. A mitotic catastrophe may occur if tubulin dynamics are disrupted, causing an aberrant or premature entrance into mitosis[17].

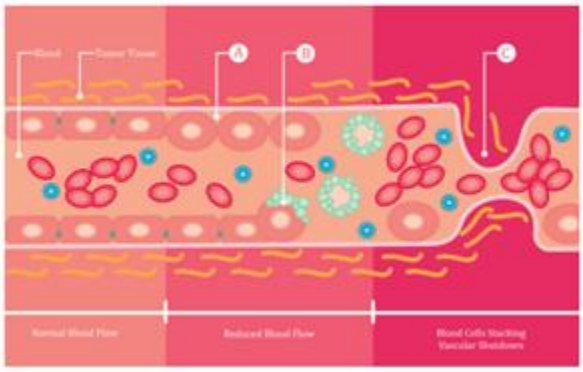


Fig no. 2: Mechanism of action of Combretastatin

Molecular insights of the induction of programmed cell death by combretastatins

Any multicellular creature that can degrade cells that have lost their physiological importance will operate properly. Apoptosis is the primary mechanism causing cell death, however, necrosis and mitotic catastrophe are alternative non-apoptotic types of cell death as well. In the following tumour cell lines, CA-4 promotes mitotic catastrophe, which results in cell death: H460, SC-M1 NUGC-3; MKN5; MKN1; KG-1a; THP-1; AGS, HL60; K562; U937, HELIUM; NB4; MKN74; and TSGH 8301; BFTC 905[18].

Apoptosis is activated and the cell cycle is halted in the G2/M phase, according to studies employing stomach cancer cell lines. CA-4 and its derivatives have been demonstrated to impede mitosis and activate the spindle checkpoint in non-small lung cancer cell lines. Researchers discovered that CA-4 increases apoptosis in cancer cell lines of lung using immunoblotting analysis with a PARP monoclonal antibody. Antibodies against active forms of caspases 9, 3 and 8 were employed in research on the cytotoxicity mechanism of CA-

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4. Caspases 9 and 3 are activated by the combretastatin, however, caspase 8 is unaffected. This suggests that combretastatin does not affect the activation of extrinsic apoptosis. A further study confirmed that Prolonged exposure to CA-4 has been shown to activate an intrinsic apoptotic pathway in NSCLC cells. Chronic lymphocytic leukaemia was used to test CA4P's ability to initiate apoptosis (CLL). Combretastatin phosphate induced an increase in the expression of caspase 9. CA4P has been shown to induce apoptosis in acute lymphoid leukaemia (ALL) cells by labelling cells with propidium iodide and annexin V and then using cytometers to analyse the results. Combretastatin A4 treatment of leukaemia cells led to the first defining sign of apoptosis, the externalisation of phosphatidylserine.

TSGH 8301 and BFTC 905 bladder cancer cell lines were observed to be inhibited by CA-4 in M phase cell cycle development, according to Jaroch K et al. 2016[19]. The sub-G1 cell population was similarly higher in BFTC 905 cells. CA-4 is more efficient at causing apoptosis in BFTC cells. BFTC cells have significant levels of the anti-apoptotic protein BubR1, which is required for M phase arrest. In BFTC 905 cells treated with caspase inhibitor Z-VAD-FMK, CA-4-induced PARP (poly (ADP-ribose) polymerase) and caspase-3 synthesis were absent. CA-4, on the other hand, could not entirely halt cell death, which suggests a different method of action. Mitotic catastrophe's phenotypic, large, multinucleated cells were discovered. Combretastatin derivative ST2151 led to the production of big cells with numerous nuclei, which is typical of mitotic catastrophe in NSCLC studies[20-22].

Tubulin Binding Activity

Anticancer drugs may target malignant tumours in various stages and places. Anticancer drugs are classified based on cell growth kinetics. First-category medicines impact DNA structure and function by disrupting transcription and synthesis. Second-class drugs inhibit proteins, while third-class drugs affect the endocrine system. The second category of anticancer drugs is most commonly used in clinical practise because to its great pharmacological efficiency and minimal toxicity. These drugs target tubulin. These drugs can bind tubulin and change microtubule dynamics [23].

Disruption of the microtubule by medications that target it slows cell growth and causes aberrant mitotic spindles in the G2-M phase. They are also known as anti-mitotic medicines because of their potential to affect microtubule stability and dynamic characteristics. For example, depending on the dosage, these drugs may either inhibit or accelerate stabilising microtubules, microtubule polymerization and increasing polymer mass. Two examples of microtubule-destabilizing drugs are colchicine and vinca alkaloids; taxanes are the other. The capacity to stop mitotic progression and trigger apoptosis is retained even at low doses of both microtubules stabilising and microtubule destabilising medicines because they interfere with microtubule dynamics rather than microtubule polymer mass. Paclitaxel, vincristine, docetaxel, vinorelbine, vinblastine, and vindesine are all extremely potent anticancer medicines that target microtubules. vincristine, vinblastine, and Colchicine depolymerize paclitaxel, docetaxel, and the polyisoprenyl benzophenones[24.]

Natural compounds such as combretastatins are also being tested in clinical studies for cancer treatment, in addition to those already mentioned. The vinca domain, the colchicine binding site, the taxane binding site are all well-known microtubule binding sites. b-tubulin binds to a GTP vinca domain site at the plus-end interface. The microtubule lumen has a taxane-binding site. It is located close to the lateral contact between consecutive protofilaments in

a deep hydrophobic pocket. Even though a- and b-tubulin and the colchicine site are both located in the intra-dimer interface. Laulimalide also takes up an additional binding site on b-tubulin. Because this binding site has yet to be identified, however, microtubule targeting agents cause cancer cells to die. When microtubule targeting agents are utilised, the G2/M phase of the cell cycle is the most probable to be arrested. There may be a link between apoptosis and MTA-induced mitotic arrest through CDC2- or JNK-induced Bcl-2 hyperphosphorylation or Bcl-xL hyperphosphorylation, CDK1 phosphorylation of BAD on serine 128 and Bim activation by JNK, according to Li et al. Tubulin is depolymerized by carbamazepine combretastatins at the vinca-independent binding site of colchicine on the b-tubulin subunit of tubulin [25-28].

Tubulin polymerization inhibition

Antimitotic Combretastatin A4 suppresses microtubule activity. CA-4 binds tubulin's colchicine-binding site. It interacts to tubulin heterodimer, causing disruption and preventing microtubule polymerization. This generates aberrant mitotic spindles and interrupts metaphase mitosis. Colchicine is harmful and has few recognised therapeutic applications, although CA-4 binding to the tubulin colchicine site exhibits high cytotoxicity against cancer cells. New scaffolds reflect CA-4's structure. Compound 5ab's influence on tubulin polymerization in a cell-free in vitro experiment was used to examine the link between tubulin and cytotoxicity [29].

Recent advancements in tubulin polymerization inhibitors

Recent research has shown that when employed as vascular disruption agents, combretastatins and other colchicine site-binding medications prevent the growth of new blood vessels in cancer tissue[30]. CA-4, which has substantial antiproliferative and antiangiogenic characteristics, is one of the most efficient combretastatins. The prodrug CA-4P has proven effective in treating pathological myopia, advanced anaplastic thyroid carcinoma, and polypoidal choroidal vasculopathy in phase II clinical studies. There are also ongoing phase III clinical studies with carboplatin/paclitaxel using CA-4P to treat anaplastic thyroid cancer. Due to its poor solubility and unstable cis structure, CA-4 has a substantial cytotoxic impact on a variety of human cancer cell lines in a lab, which include MDR cancer cells, but not on living organisms. Combretastatin's phosphate prodrugs had to be developed because of the drug's low water solubility. Because of its antiproliferative action on the interior portion of the tumour tissue, CA-4P was able to leave a few viable tumours on the periphery in patients with solid tumours. To avoid this issue, The use of CA-4P in conjunction with other drugs is suggested. CA-4 has a significant amount of therapeutic value because of its short biological half-life, poor water solubility and cis-trans isomerization. CA-4 has the ability to undergo modifications for producing new, stronger molecules with enhanced therapeutic potential[25]. According to the previous studies, CA-4's cytotoxicity and anti-tubulin action is dependent on the existence of the C-2 bridge double bond as well as a trimethoxybenzene moiety and 4-methoxy-substituted B ring and a 3,4,5-trimethoxysubstituted A ring. The cis-configuration of the structure may be preserved by reducing the olefinic bridge and substituting it with carbocyclic rings or heterocyclic rings that include bridging functional groups. Additionally, it makes substances more soluble in water and have other effects similar to those of drugs. This may be done by substituting the stilbene moiety's ethenyl bridge (phenstatin) with a stable keto group (sulphide) or ether. By altering the substitutes at the meta location of the CA-4 B ring, its derivatives CA-4NH2Hcl, CA-

4P, and CA-4NH-Ser have shown significant antimitotic action and improved bioavailability[31].

Carbonyl oxygen increased the antiproliferative effect of diaryl ketone chalcone and aroylindole. Lung cancer cells responded better to 2-aminoimidazole-carbonyl than CA-4. Compared to CA-4, it destroys breast cancer cells more effectively. Indole or naphthalene analogues of CA-4's 3-hydroxy-4-methoxyphenyl (B-ring) have potent anticancer capabilities but are less water-soluble than CA-4 (1d). Polar groups and intramolecular hydrogen bonding promote solubility. This strategy may be difficult if polar groups are in hydrophobic places like the colchicine site. Gonzalez and coworkers used MPGI bonds low-polar areas of the target and enhances water solubility. Adding polar groups requires vicinal bulk. phenyl rings hide nitrogen atoms in isocombretastatin and dimethylaminocombretastatin analogues, forming pyridines. Many cancer cell lines were resistant to the antiproliferative agents [32].

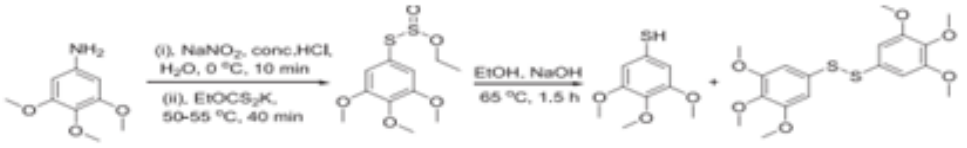
Table 1: Some recent studies on the antitumour agent (Combretastatin)

S.no.	Author	Recent Observations
1.	**Paidakula S et al. 2022**[33]	antitumor drug development employing CA-4 analogues as tubulin inhibitory agents, emphasising their structure-activity relationships (SAR) and resulting pharmacological efficacies are discussed in the most current trends and advancements.
2.	**Ahmadi R et al. 2022**[34]	Combretastatin CA-4 analogues were synthesised using the Suzuki-Miyaura coupling method, and their in vitro antiproliferative activity against MDA-MB 231, HELa, and A549 human cancer cell lines and the healthy cell line HEK-293 was evaluated.
3.	**Liu W et al. 2022**[35]	These novel imidazole-chalcone based anticancer and tubulin polymerization inhibitors were developed.
4.	**Haider K et al. 2019**[36]	It has been demonstrated that the novel class of quinoline derivatives of the tubulin polymerization inhibitors, combretastatin A-4, are effective. It was shown that these new chemicals have potent antiproliferative effects.
5.	**Reipsch F et al. 2021**[37]	N-methylimidazoles that are combretastatin analogs were shown to have similar vascular-disrupting effects to CA-4P.
6.	**Yang L et al. 2022**[38]	The anticancer potential of tubulin and MMP inhibitor conjugates is examined in this study. The highest antiproliferative action was seen in the conjugate 15g containing 2-(4-((diethoxyphosphono)(o-tolyl) methylamino)phenyl)acetic acid (19g).
7.	**Deng C et al. 2020**[39]	In clinical situations, CA4P as a potent anticancer drug candidate for fusion with CAR-T cells to treat solid tumours.

8.	**Atanasov G et al. 2020**[40]	To test whether or not they could create nanosized micelles and transport combretastatin-like anticancer drugs, they synthesised biodegradable and biocompatible block copolymers based on poly(lactide-co-glycolide) and poly (ethylene glycol) with linear (ABA-type) architecture.
9.	**An Y et al. 2020**[41]	The light-triggered conformation transition of Azo-azobenzene CA4's moiety reduces chemotherapy adverse effects.
10.	**Gu Y et al. 2019**[42]	Slow drug release from CA4 with a longer acyl chain could improve medication concentration in tumour tissue. These actions boost antitumor effectiveness.
11.	**Molloy JJ et al. 2018**[43]	This contra-thermodynamic EZ isomerization is validated in a series of stereoretentive transformations to produce Z-configured trisubstituted alkenes and in a succinct synthesis of Combretastatin A4.
12.	**Ngo QA et al. 2021**[44]	Newly synthesised aryl-substituted pyrazole compounds resembling celecoxib and combretastatin A-4's cis-diphenylethylene scaffold have strong antiproliferative and antiinflammatory properties.
13.	**Schmidt C et al. 2021**[45]	In vitro, the bioactive ligand combretastatin A4 (CA4) is 1000 times more toxic than cisplatin, making multi-action Pt(IV) derivatives of cisplatin with CA4 notable.
14.	**Jiang H et al. 2019**[46]	GA-modified LPs can transport anticancer medicines (combretastatin) to the liver for hepatocellular carcinoma.

Diarylsulfide, Diarylmethane, diarylsilane and diarylamine derivatives

Tubulin polymerization inhibition was explored by substituting isocombretastatin's B-ring with an indole systemTo increase the solubility characteristic, many changes have been made to the 3-position of phenstatins and 3-position of 1-methyl-5-indolyl-based isocombretastatins 1, 1-diarylethenes. CH2OH, through various processes, several substituents have been added to the indole system's third position, including COOH, CN, CONH2, NH2, and CH2NH2. All of the compounds that were generated showed significant cytotoxicity, but compounds 68, 72, and 71 distinguished out for their ability to completely block tubulin assembly at concentrations of 20 mM and there is a significant difference in activity between 3,4,5-trimethoxy-substituted phenyl rings and 2,3,4-trimethoxyphenyl rings[47, 48].



1 2 3 4

Fig no.3: Synthesis of disulfide comprising derivative of combretastatin

Heterocyclic analogues

Five membered heterocyclic ring : Containing two heteroatoms

Combretastatin's cis-olefin is a bioisostere, therefore a special sequence of thiazoles with a trimethoxyphenyl ring was developed by Romeo Romagnoli et al. 2008. that is similar to the a second aryl ring at the C-5 regions of the thiazole nucleus and combretastatin. The thiazole C-2 position has also undergone a number of changes. (o-ethyl-S-[2-oxo-2-(3,4,5-trimethoxyphenyl) ethyl]) or (2-bromo-1-(3,4,5-trimethoxyphenyl)ethanone 5) compounds are condensed which is a step in the production process. To provide suitable access, dithiocarbonate 6 is coupled with dimethyamino nitrile, N-methylthioure, thioacetamide, or piperidine. The Suzukie-Miyaura coupling process was then carried out on derivative 7 using PdCl2 and substituted boronic acid (dppf). The most effective inhibitor of tubulin polymerization was observed to be compound 5a (R1 14 NCH3, R2 14 4- Oc2H5eC6H4), while the others significantly inhibited tubulin[49].

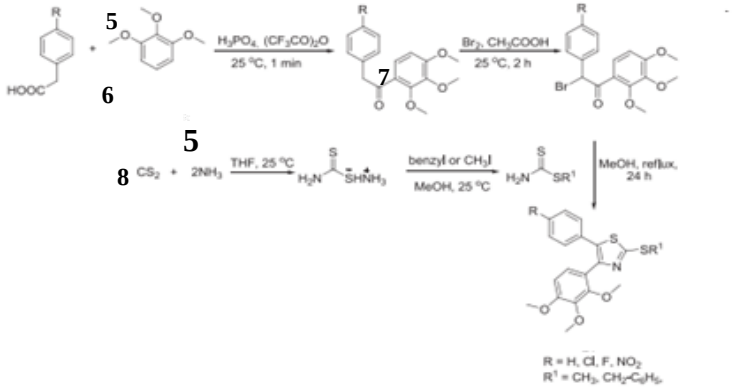


Fig no. 4: Synthesis of thiazole derivatives

Zhou and colleagues have designed and manufactured combretastatin analogues, which include the ethylene bridge being switched out for an oxazole linker as well as the B-ring being interchanged with 4-methoxy-1H-benzo[d]pyridine, rings of imidazole[50]. The tubulin polymerization inhibitory action of 2-alkylthio-4- (2,3,4-trimethoxyphenyl)-5-aryl-thiazoles has been studied by Assadieskandar A et al. 2013. By enabling substituted phenylacetic acid to react with trimethoxybenzene 10, 1-(2,3,4-trimethoxyphenyl)-2-(4-substituted phenyl)-1-ethanones 11 were generated, from which the a-bromoketone 12 was obtained. Bromoketones and benzyl carbamodithioate 13 combine to generate the end product is 2-alkylthio -4-(2,3,4-trimethoxyphenyl)-5-aryl-thiazole. CS2 and NH3 were combined to produce benzyl carba modithioate 14, and then either benzyl or methyl iodide was added. Compound 15a (R 14 Cl, R1 14 CH2eC6H5) (IC50 14 10 mM) was the most effective of the newly produced compounds, with an IC50 of 10 mM[51].

10 11 12 13 14 15

fig no. 5: Synthesis of 2-alkylthio-4-(2,3,4-trimethoxyphenyl)-5-aryl-thiazole

Containing three heteroatoms

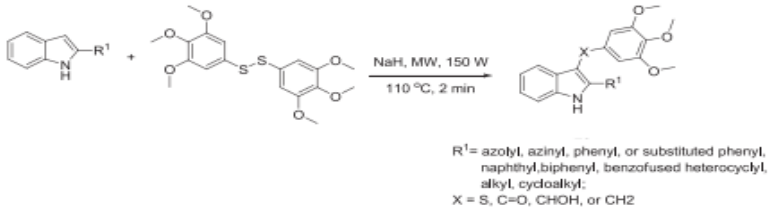
A small collection of 1,2,4-oxadiazole substituted 3,5-disubstituted combretastatin derivatives was produced by Akselsen ØW et al. 2012 to enhance the drug's solubility profile and antitubulin activity. To generate the starting material for the synthesis, substituted amidoxime and 3,4,5-trimethoxybenzoic acid undergo a coupling process in the presence of coupling agents (CDI and HOBt)[52.]

Benzofused five membered heterocyclic ring

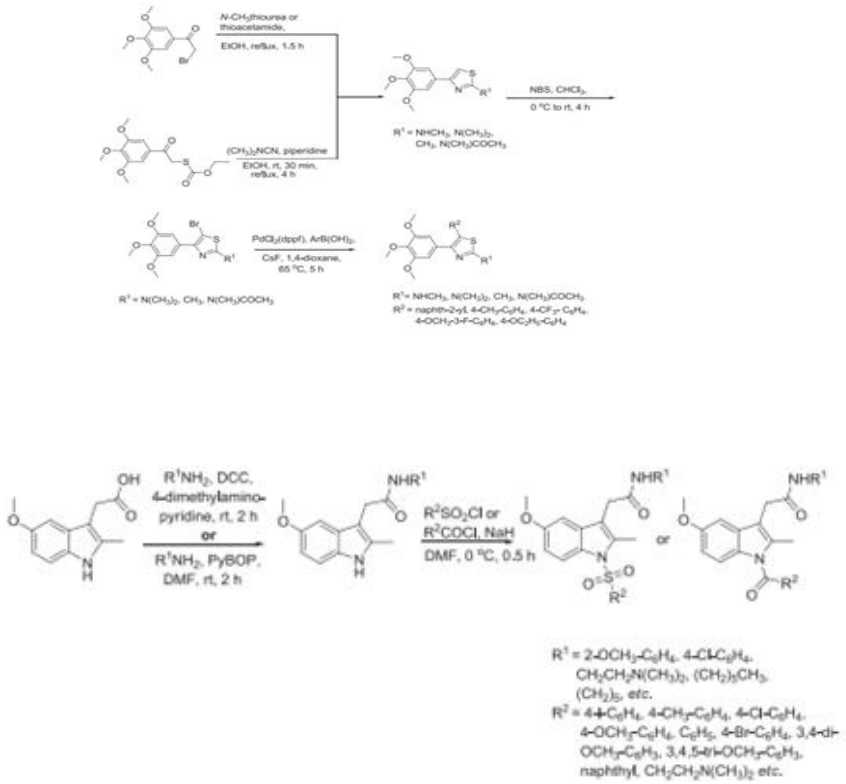
Tubulin polymerization inhibitors include ATIs, a significant tubulin polymerization inhibitor. A five-membered heterocycle nucleus has often substituted the 2-alkoxycarbonyl group of arylthioindoles in analogues with higher activity. Based on this, La Regina G et al. 2013 substituted several bioisosteric rings in the second position of the indole ring. In NaH under MW, the required counterpart, 18, was produced via the reaction between bis(3,4,5-trimethoxyphenyl) disulfide 17 and substituted indole 16[53].

16 17 18

Fig no.6: Synthesis of substituted arylthioindoles



Indomethacin and sulindac, two essential cyclooxygenase (COX) inhibitors, effectively inhibited cell proliferation and might trigger apoptosis. In an effort to find more effective and targeted anticancer medications, In order to create a new series of analogues, Chennamaneni S et al. 2012 modified the indomethacin and sulindac scaffold in a variety of ways[54].



19 **20** **21** **22**

Fig no. 7: Synthesis of indomethacin derivatives

Conclusion

A suitable strategy is the development of anticancer medications that particularly target the microtubule system. Despite its modest molecular weight and straightforward chemical composition, CA-4 is one of the most effective tubulin polymerization inhibitors via binding to the colchicine site. Due to their straightforward structure, strong binding strength at the colchicine binding pocket and incredible cytotoxicity, CA-4 attracted a lot of attention for the synthesis of many congeners. The biological activity profile of the molecule has been modified by the addition of several motifs, including cyclic and heterocyclic ones. In the near future, many analogues of the compound combretastatin may provide a potential lead for the production of therapeutically significant drug candidates.

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CONFLICT OF INTEREST: NIL

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