A RESEARCH ON SYNTHESIS AND FORMULATION OF HETEROCYCLIC ANALOGS TOWARDS POTENTIAL INHIBITORS IN NITROGEN AND SULPHUR COMPOUNDS

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ABSTRACT

The beta-amylase enzyme is the most effective treatment target for type 2 diabetes mellitus. In order to find new therapeutic agents that can act as -amylase inhibitors, the research that is presented in this dissertation focuses on the synthesis of nitrogen- and sulfur-containing heterocyclic compounds using an approach called molecular hybridization. These compounds have a variety of different moieties, such as thiazolidin-4-one, 2,4-thiazolidinediones, 1,3-thiazoles, and benzothiazoles clubbed pyrazole or ox All of the molecular hybrids were tested at a range of different concentrations against the -amylase produced by Aspergillus oryzae. The whole study is broken up into five parts, with the first one focusing on the structure and mechanism of action of beta-amylase.

INTRODUCTION

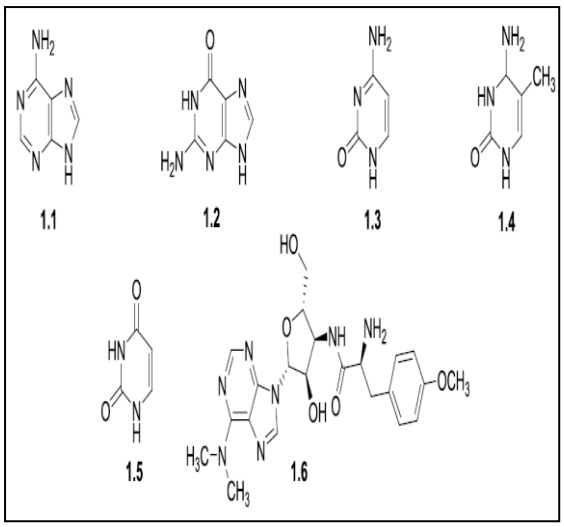
Heterocyclic compounds are a subset of carbocyclic chemicals. They vary from carbocyclic compounds in that they have a ring structure that incorporates at least one heteroatom, frequently nitrogen, oxygen, or sulfur, in place of carbon. Heterocyclic substances may be either aliphatic or aromatic.

The study of the chemical events that take place within heterocyclic compounds is among the most significant elements of organic chemistry. Researchers in the domains of medicinal chemistry, pharmacology, and biology are interested in researching them because of the crucial therapeutic and biological activities they are related with.

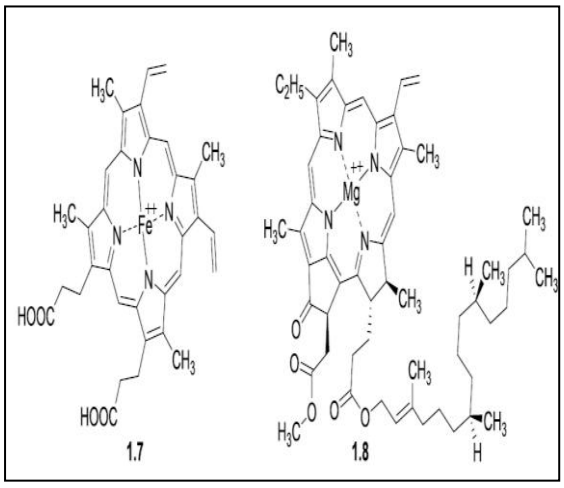
Because the bulk of medications are formed around heterocyclic nucleus, heterocyclic compounds are like undiscovered gold mines of physiologically relevant substances. The bulk of the drugs are heterocyclic compounds with five or six members and one to three heteroatoms in their nucleus. These compounds are termed 5- and 6-membered heterocycles.

To acquire the needed activity for the welfare of mankind, the structure of heterocycles may be easily adjusted to create the appropriate outcomes. 1,2 As a consequence, heterocycles have developed to become the most important scaffolds for research in the area of organic chemistry.

The chemical components that make up hereditary materials like DNA and RNA are known as purines, which comprise adenine (1.1) and guanine (1.2), and pyrimidines, which include cytosine (1.3), thymine (1.4), and uracil (1.5). (1.5). 3,4 Some of the purine and pyrimidine derivatives, such as puromycin, are nucleoside antibiotics that hinder the formation of protein. This is the situation with purines and pyrimidines (1.6). (1.6).

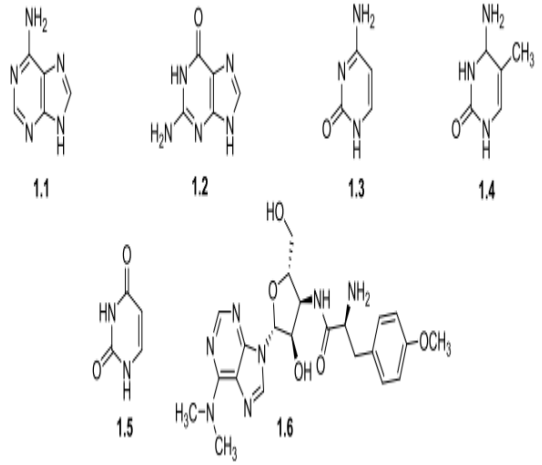


In a similar line, naturally occurring heterocyclic moieties include porphyrin ring derivatives such as the oxygen-transporting pigment heme (1.7) and the photosynthesizing pigment chlorophyll. Both of these pigments play an important role in photosynthesis (1.8).

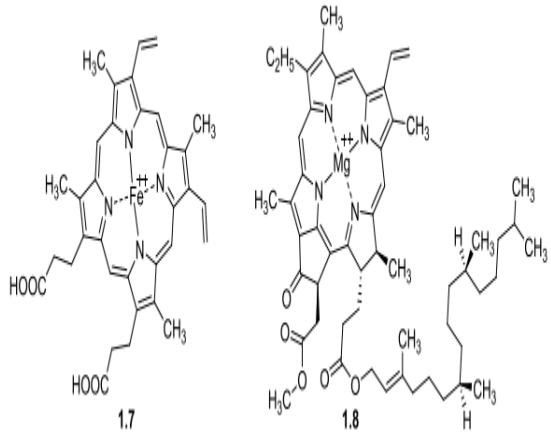


LITERATURE REVIEW

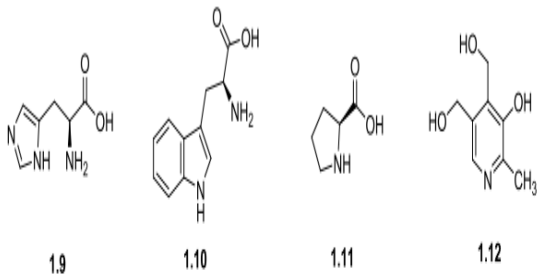
Heterocycles have become the most important scaffolds for research in organic chemistry. The purines [adenine (1.1) and guanine (1.2)] and pyrimidines [cytosine (1.3), thymine (1.4) and uracil (1.5)] are the chemical building blocks of hereditary materials such as DNA and RNA.3,4 Some of the derivatives of purines and pyrimidines act as nucleoside antibiotics that inhibit the synthesis of protein e.g., puromycin (1.6).



Similarly, porphyrin ring derivatives such as oxygen-transporting pigment heme (1.7) and photosynthesizing pigment i.e., chlorophyll (1.8) are the naturally occurring heterocyclic moieties.



Essential amino acids like histidine (1.9), tryptophan (1.10) and proline (1.11), the vitamins: pyridoxine (1.12) (vitamin B6), biotin (1.13) (vitamin B7), ascorbic acid (1.14) (vitamin C), α-tocopherol (1.15) (vitamin E), thiamine (1.16) (vitamin B1) and riboflavin (1.17) (vitamin B2) are also the common heterocyclic compounds involved in the biological processes. Besides this, some natural products also possess different heterocyclic systems.



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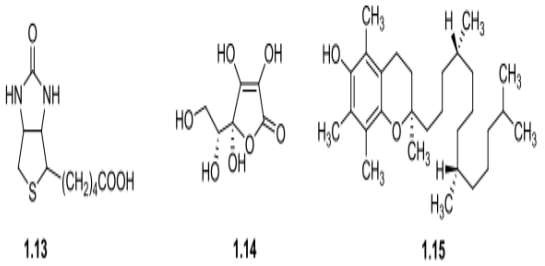

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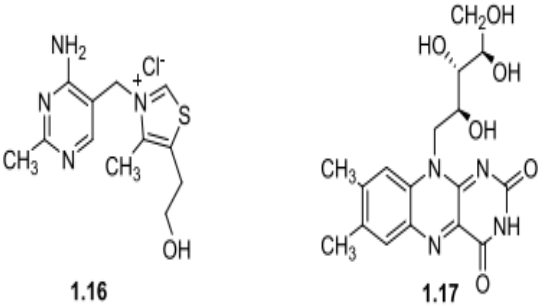
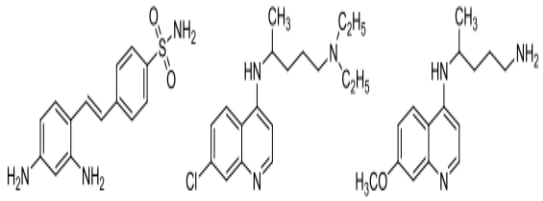
Human beings have been using natural products since antiquity for therapeutic purposes which include the antimalarial drugs (quinine), β-lactam antibiotics (penicillin), narcotic analgesic opiate alkaloids (codeine and morphine), anticholinergic drugs (atropine) etc.



1.16 1.17

In 1939, Gerhard Domagk, a renowned German pathologist synthesized first commercially available antibiotic, sulfonamidochrysoidine (1.18). Since then, a plethora of synthesized heterocyclic compounds8 showed their worth as antitumor, antiinflammatory, analgesic, antimalarial, antiHIV, antibiotics, antidepressant, antidiabetic agents etc. Synthetic drugs act as antimalarialswhich comprise 4- aminoquinoline i.e., chloroquine (1.19) and 8-aminoquinoline i.e., primaquine (1.20), nonsteroidal aromatase inhibitor (estrogen synthetase inhibitors) containing triazole moiety i.e., anastrozole (1.21), anthelmintic drug i.e., albendazole (1.22) having benzimidazole moiety and antidepressant i.e., paroxetine (1.23) having piperidine nucleus.



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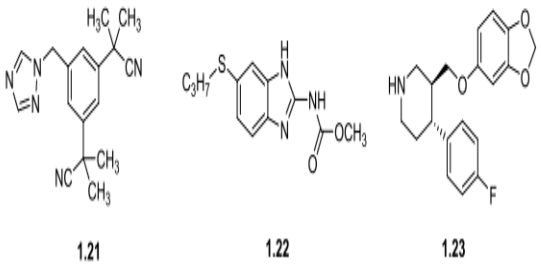
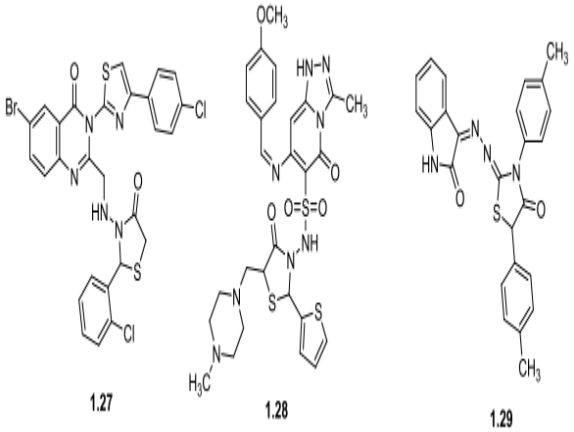


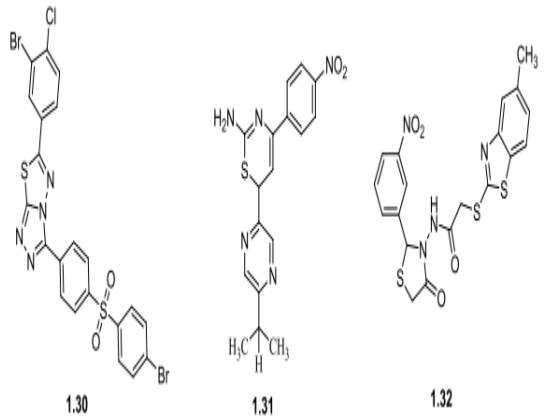
1.21 1.22 1.23

Dyes containing phenazine moiety i.e., mauveine, pesticides comprising pyrimidine moiety with thiophosphoric acid ester group i.e., diazinon, herbicides having bipyridine moiety i.e., paraquat are some of the examples of heterocycles present in nature.

Sulphur and nitrogen containing heterocyclic systems have attracted medicinal and organic chemist as far their biological relevance is concerned. These heterocyclic systems are associated with wide range of biological activities such as antineoplastic [i.e.,dasatinib (1.24)], antifungal i.e., ravuconazole (1.25), anticancer20 i.e., 2-(3-indolyl)-3-[4-(pyridin-2-ylamino)sulfonyl]phenylthiazolidin-4-one

(1.26), antiinflammatory21 i.e., 2-(4'-oxo-2'(o-chlorophenyl)-thiazolidin-3'-yl-aminomethyl)-3- [4"-(p-chlorophenyl)thiazol-2"-yl]-6-bromoquinazolin-4-one (1.27), antitumor22 i.e., (Z)- 7-((4-methoxybenzylidene) amino)-3-methyl-N-(5-((4-methylpiperazin-1-yl)methyl)-4-oxo-2-(thiophen-2-yl)thiazolid -in-3-yl)-5-oxo-1,5-dihydro-[1,2,4]triazolo[4,3-a]pyridine6-sulfonamide (1.28), CNS (central nervous system) depressant23 i.e., 2-(((Z)-2- oxoindolin-3-ylidene)hydrazono)-3, 5-di-p-tolylthiazolidin-4-one (1.29), antimicrobial24 i.e., 6-(3-bromo-4-chloro)phenyl-3-[4-(4-bromophenylsulfonyl)phenyl]-[1,2,4] triazolo [3,4-b][1,3,4]thiadiazole (1.30), antiHIV25 i.e., 6-(5-isopropylpyrazin-2-yl)-4-(4- nitrophenyl)-6H-1,3-thiazin-2-amine (1.31), antidiabetic26 i.e., 2-(5-methylbenzo[d] thiazol-2-ylthio)-N-(2-(3-nitrophenyl)-4-oxothiazolidin-3-yl)acetamide (1.32) etc.





NEED OF THE STUDY:

The aim of this study is to theheterocycles as α-amylase inhibitors for treatment of diabetes mellitus. This study presents information, gained through the glimpse of literature describing the structure and mechanism of action of α-amylase. It also gives a brief account of biological application of nitrogen and sulphur containing heterocycles and their role as α-amylase inhibitors.The experiment was performed according to the method given by Shetty et al. 68 with slight modifications. A stock solution of 1 mg/mL concentration was prepared by using the DMSO solvent. The activity of α-amylase was assayed at three different concentrations (25, 50, 100 µg/mL) and the reagent solution without a test sample was used as control. The α-amylase (Aspergillusoryzae) enzyme solution was prepared by mixing 5 mg in 100 mL (50 µg/mL) of 20 mM sodium-potassium buffer (pH 6.9). The starch solution was prepared by dissolving starch (500 mg) in 25 mL of 0.5 N NaOH and heat the solution for 5 min at 100oC. After cooling in ice H2O, the pH of the solution was adjusted to 7 with 2M HCl, and water was added to adjust the volume to 100 mL. Coloring reagent i.e., 96 mM 3,5-dinitrosalicyclic acid (DNSA) with sodium

potassium tartrate solution was used which was prepared by dissolving 2.18 g of DNSA (MW=228.1 g/mol) in 80 mL of 0.5 M NaOH by heating and stirring at 70oC. Then added 30 g of sodium potassium tartrate (MW=282.2 g/mol) and stirred until get dissolved.

Cooled the solution at room temperature and bring the volume to 100 mL with purified water. Acarbose was used as a standard at the concentration of 1 mg/mL.The experiment was performed by mixing 1 mL of enzyme solution and 1 mL of different concentration of synthesized compounds 2.59a-2.59u (25, 50, 100 µg/mL) in DMSO, and the solution was incubated for 30 min at 37oC. To this, add 1 mL of starch solution and the mixture was further incubated for 15 min at 37oC. Then 1 mL of 96 mM of DNSA coloring reagent was added to the above solution. After shaking the reaction mixture, the closed test tubes were placed in the water bath at 85oC for 15 min.

After then, the reaction mixture was removed from the water bath, cooled and absorbance value was determined at 650 nm. Individual blanks were prepared for correcting the background absorbance.In the last decades, 1-3 combinatorial chemistry has provided access to chemical libraries based on privileged structures, with heterocyclic moieties. The appearance of a large number of research articles for the nitrogen and sulphur-containing heterocyclic compounds, further shows the keen interest of synthetic as well as medicinal chemists in the synthesis of these compounds. There are numerous biologically active molecules with five-membered rings, containing two heteroatoms. Among them, the study of thiazolidin-4-one and pyrazole is of considerable interest for medicinal chemist.

RESEARCH TYPE

The type of analysis defines the essence of the data in the study. Given the nature of the data, the work currently under way will a qualitative cum quantitative aspect, but is mainly quantitative in aspect, as most findings of this analysis willfocus on quantify measures. They willassociate with significant therapeutic and biological activities which draw the attention of medicinal chemists, pharmacologists and biologists. Heterocyclic compounds will the gold mines of biologically relevant compounds as most of the drugs willbase on heterocyclic nucleus. Most of the drugs will five- and six member heterocyclic compounds containing one to three heteroatoms in their nuclei.

SAMPLE DESIGN

In certain cases of science, analyzing the entire universe is almost impossible; the only alternative will to use sampling. The current researches will the same character. The procedure for deciding the sample of the analysis is samplingthe chemistry of heterocyclic compounds is one of the most imperative extents of organic chemistry.The structure of heterocycles will be easily manipulated to achieve the desired activity for the benefit of mankind. Although variety of heteroatoms will be integrated in the heterocycles, but the heterocyclic systems which comprise of nitrogen and/or sulphur, are the centers of attraction in organic and medicinal chemistry, due to their vast biological significance such as antineoplastic, anticancer, antimicrobial, anti-HIV,

antimalarial, anti-diabetic etc.

DATA COLLECTION

Data collection is the systematic way to collect and measure data from sources to get complete and precise data for research activities. In all areas of study the facts collection component is not unusual with body and social sciences, the humanities and corporations. It allows scientists and analysts to collect key factors as the information they collect. In contrast with the approaches in terms of subject matter, the value of maintaining the right and truthful sequence remains the same. Current data collection is essential for preserving the credibility of research and for ensuring excellent outcomes and their findings. This study will be secondary research method.

SECONDARY DATA

Secondary data will the data collect by an individual rather than the user. A researcher who is not associated with the analysis / recherché study collects secondary information for a different purpose, and in the past at quite different times such data are readily accessible and cost effective in comparison to primary data.

Sources of secondary data collection will as follows:

- Government department's journals,
- Organizational records,
- Magazines,
- Journals, books,
- Newspapers and
- The information which is collected originally for other research purpose.

RESULT

Synthesis of Some Nitrogen and Sulphur Containing Heterocyclic Compounds as Potential α-Amylase Inhibitors

The study of the chemical events that take place within heterocyclic compounds is among the most significant elements of organic chemistry. Researchers in the domains of medicinal chemistry, pharmacology, and biology are interested in researching them because of the crucial therapeutic and biological activities they are related with. Because the bulk of medications are formed around heterocyclic nucleus, heterocyclic compounds are like undiscovered gold mines of physiologically relevant substances.

The bulk of the drugs are heterocyclic compounds with five or six members and one to three heteroatoms in their nucleus. These chemicals are termed cycloheterocycles. To acquire the needed activity for the welfare of mankind, the structure of heterocycles may be easily adjusted to create the appropriate outcomes. Although a broad range of heteroatoms have been combined into heterocycles, it is the heterocyclic systems that consist of nitrogen and/or sulphur that are the center of research in organic and medicinal chemistry. This is owing to the immense biological relevance of heterocyclic systems, which includes qualities such as antineoplasticity, anticancer, antimicrobial activity, anti-HIV activity, anti-malarial activity, and anti-diabetic activity, among other properties.

Diabetes mellitus is one of the most prevalent non-communicable metabolic illnesses. It is characterized by chronic hyperglycemia, commonly known as high blood glucose levels, as well as changes in carbohydrate, lipid, and protein metabolism that occur from an absolute or relative absence of insulin production.

Diabetes mellitus inhibits a person's ability to metabolize carbs, lipids, and proteins properly. Enzymes that govern glycogenolytic pathways are key biological targets for therapeutic interventions as a consequence.

Among the various enzymes that are known, beta-amylase is a crucial essential enzyme that is responsible for the digestion of carbohydrates. Inhibitors of -amylase are capable of effectively slowing the digestion and absorption of starch during its early phases. As a consequence, they are able to dramatically minimize the risk of postprandial hyperglycemia and have a beneficial influence on insulin resistance. Because of its capacity to catalyze the hydrolysis of -(1,4)-glycosidic bonds in starch, beta-amylase is regarded to be one of the best targets for the development of therapeutic medications for type 2 diabetes. Both acarbose and voglibose are acknowledged inhibitors of -amylase, and both are presently being utilized in clinical contexts. On the other side, they typically result in substantial gastrointestinal unpleasant effects such as cramping in the stomach area, burp, and diarrhea.

In recent years, beta-amylase has emerged as a viable target for the study and development of novel anti-obesity and anti-diabetic drugs. Numerous research have been carried out in the effort to identify innovative inhibitors of beta-amylase that do not result in any undesirable side effects. The current research effort on nitrogen and sulphur containing heterocyclic compounds has been undertaken with the following rationale in order to extend this pool of inhibitors in the present endeavor. The research study will be carried out in order to:

1. To carry out the synthesis of novel N- and S- containing hybrid heterocyclic molecules i.e.,

i. 5-[(3-aryl-1-phenyl-1H-pyrazol-4-yl)methylene]-2-(p-arylimino)thiazolidin-4-ones,

ii. (3'S, 4'S)-4'-(aryl)-1'-methyl-1-((1-(aryl)-1H-1, 2, 3-triazol-4-yl)methyl) dispiro[indoline-3,2'-pyrrolidine-3',5"-thiazolidine]-2,2",4"-triones,

iii. 1-((1-phenyl-3-aryl-1H-pyrazole-4-yl) methylene)-2-(4-arylthiazole-2-yl) hydrazines,

iv. (E)-N-(4-(4-(benzo[d]thiazol-2-yl) aryloxy) benzylidene)-(aryl)hydrazides and

v. 2-(4-(4-(benzo[d]thiazol-2-yl)aryloxy) phenyl)-5-(aryl)-1,3,4-oxadiazoles.

2. To study the inhibitory potential of all the newly synthesized compounds towards Aspergillus oryzae α-amylase (Taka-amylase A or TAA).

3. Studies of the most active molecule using in silico molecular docking against Aspergillus oryzae -amylase (PDB: 7TAA).

The whole body of work has been broken down into five parts, the first of which is a literature review that is supplied as Chapter 1 and is headed "heterocycles as -amylase inhibitors for treatment of diabetes mellitus." This chapter comprises information that has been obtained from a quick examination of the literature regarding the structure of beta-amylase and the way by which it acts. This material has been included here for your convenience. In addition to this, a description of the biological uses of heterocycles containing nitrogen and

sulfur, as well as their function as beta-amylase inhibitors, is presented in a condensed form.

In the second chapter, we will talk about the synthesis of molecular hybrids based on thiazolidin-4-one and pyrazolyl pharmacophores. More specifically, we will talk about 5-((3-(aryl)-1-phenyl-1H-pyrazol-4-yl)methylene)-2-(p-arylimino) thiazolidin-4-ones (5a-5u) as -amylase inhibitors. In this chapter, we also talk about the hybrids' possibilities from a biological standpoint. The synthesis of the target compounds, 5a-5u, was carried out in such a way as to adhere to the sequence of events that is specified in Scheme 1.

In the initial phase of the procedure, the p-arylthiourea (2a-2c) that was necessary for the synthesis of 2-(p-arylimino)thiazolidin-4-one (3a-3c) was prepared by the reaction of variously substituted amines (1a-1c) with KSCN in 6N HCl. This step was followed by the synthesis of 2-(p-arylimino)thiazolidin-4-one (3a-3c). This step was carried out in order to bring the synthesis of 2-(p-arylimino)thiazolidin-4-one to a successful conclusion (3a-3c). The chemicals 2a-2c that were produced as a consequence of this reaction were then reacted with ethyl bromoacetate in order to form the products 3a-3c, which are the key reactants for the synthesis of new hybrids.

In the subsequent step, a Knoevenagel condensation was performed on compound 3a-3c by combining it with 3-(aryl)-1-phenyl-1H-pyrazole-4-carbaldehyde (4a-4g) in the presence of the catalytic quantity of piperidine in ethanol that was heated to a simmer for 10-12 hours. This step took place in the presence of the catalytic quantity of piperidine. TLC was used in order to monitor the progress of the reaction as it proceeded. For this purpose, petroleum ether and ethyl acetate were combined in a volumetric ratio of 60:40 prior to the TLC analysis.

After the completion of the chemical reaction, the mixture produced by the reaction was allowed to cool down at room temperature. After the solid that had been generated had been filtered, a yield of 71-88% of the thiazolidin-4-one linked pyrazole-based molecular hybrids 5a-5u was obtained. In order to determine the structures of the compounds that were synthesized, the spectrum data of each individual chemical was used.

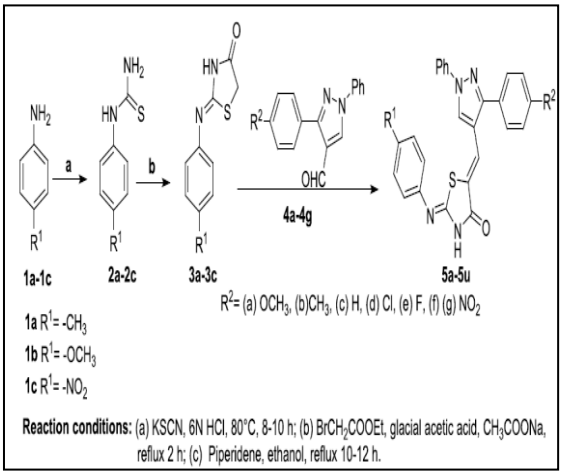


Figure: Synthesis of thiazolidine-4-one clubbed pyrazole based molecular hybrids (5a-5u)

A single isomeric product was obtained as a result of the electron-withdrawing group 5o-5u (R1= -NO2), which was formed as a result of the composition of the substituents, which had a key influence on the effectiveness of the technique involved (2Z,5Z-isomer). However, the electron-releasing group 5a-5n (R1= - CH3, -OCH3) led to the creation of two isomeric products, which are depicted in Figure 1 as the 2Z,5Z-isomer and the 2E,5Z-isomer, respectively. Both of these products are referred to as the 2Z,5Z-isomer.

In the proton NMR analysis, the ratio of the NMR integration ratio of

corresponding protons was utilized to calculate the ratio of the isomers 2E,5Z (37.1-42.0%) and 2Z,5Z (58.4-62.8%) for compounds 5a-5n. These ratios were in the range of 37.1-42.0% and 58.4-62.8%, respectively. It was projected that this ratio would be 58.4-62.8%. The findings of the studies conducted by DFT gave more evidence that supported the conclusions expressed before.

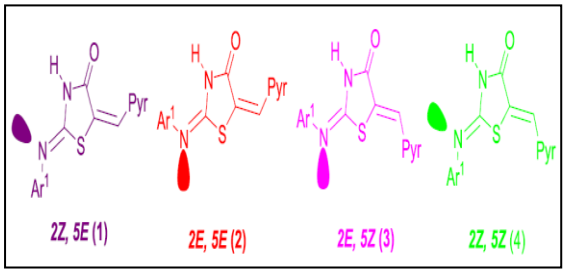


Figure: Four possible isomers for 5a-5n

After that, the two isomeric forms of 5a that were found by 1H-NMR were produced by applying comprehensive energy optimizations that were carried out using B3LYP/6-311G level of theory. This was done in order to construct the new forms. The isomer that has the configuration (2Z,5Z) is the one that has the least amount of energy and is the one that is the most stable.

Both the total energy (Etotal) of the (2Z,5Z) isomer of 5a and the total energy (Etotal) of the (2E,5Z) isomer were calculated, and the results were reported as the relative energy (Erel) in comparison to the total energy of the (2Z,5Z) isomer of 5a. The isomer that has the configuration (2Z,5Z) is more stable than the isomer that has the configuration (2E,5Z) by a difference of 3.41 kcal/mol according to the B3LYP/6-311G level of theory.

The in vitro -amylase inhibitory activity of all of the newly synthesized compounds were tested at three different concentrations (100, 50, and 25 g/mL) in order to evaluate their potential for usage in biological applications. 5-((3-(4-methoxyphenyl)-1-phenyl-1H-pyrazol-4-yl)methylene)-2-(p-tolylimino)thiazolidi-4-one (5a), 2-(4-methoxyphenyl)-1-phenyl-1H-pyrazol-4-yl)methylene, and 2-(4-methoxyphenyl)-1-phenyl-1H- (4-methoxyphenylimino) -5-((3-(4-nitrophenyl) Docking studies of compound 5a, 5n, and 5o against the active site of Aspergillus oryzae -amylase (often referred to as Taka-amylase A or TAA) gave more support for the in vitro -amylase inhibition (PDB ID: 7TAA). Molecular docking is a technology that may be used to study the possible binding modalities of a drug by assessing the active site of a target protein. This is done via the process of evaluating the active site of a target protein.

In order to make an informed assumption as to the most probable method of binding for the three most potent compounds, 5a, 5n, and 5o, the molecular docking study was carried out with the help of the Auto Dock Vina software. According to the findings of the docking studies, the binding interactions that were found to exist between beta-amylase and residues 5a (Asp206 and His210), 5n (Asp206 and His210), and 5o (Gln35, Trp83, Lys209 and His210) are comparable to the interactions that are responsible for the inhibition of beta-amylase by acarbose.

In Chapter 3, a study of eight distinct books is offered for the reader's perusal. (3'S,4'S) -4'-(aryl) -1'-methyl -1-((1-(aryl)-1H-1,2,3-triazol-4-yl)methyl)dispiro [indoline-3,2'-pyrrolidine-3',5''-thiazolidine] -2,2'',4''-trione (10a-10h) by the use of a one-pot, four-component cycloaddition technique using Z-(5)-(arylidene) thiazolidine-2,4-dione (6a-6d), 1-(prop-2-yn-1-yl)indoline-2,3-dione (7),

substituted aryl azides (8a-8c), and sarcosine (9) in PEG-400 in the presence of a catalyst consisting of an aqueous solution of CuSO4.5H2O (10 mol%) and sodium ascorbate (20 mol%) at a temperature of 100 degrees (Scheme 2). The structures of all of the newly synthesized compounds have been verified using elemental, spectroscopic (IR, 1H-NMR, MS), and single crystal X-ray analysis (Figure 2).

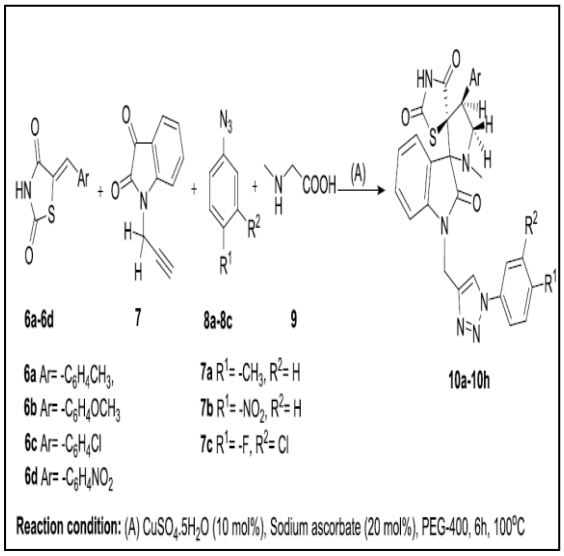


Figure: Synthesis of (3'S,4'S)-4'-(aryl)-1'-methyl-1-((1-(aryl)-1H-1,2,3-triazol-4-yl)methyl) dispiro[indoline-3,2'-pyrrolidine-3',5''-thiazolidine]-2,2'',4''-trione (10a-10h)

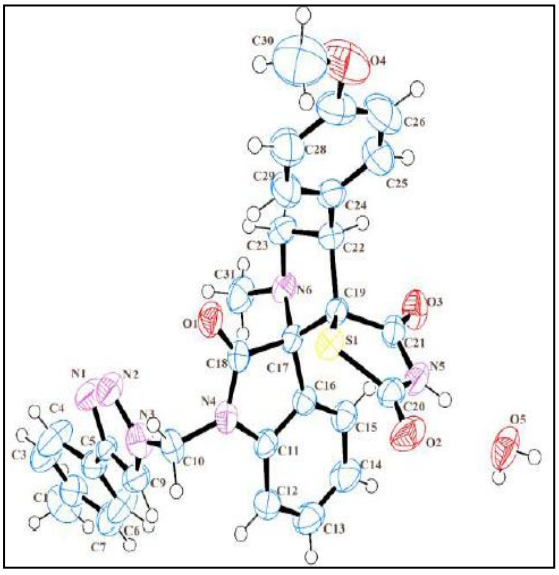


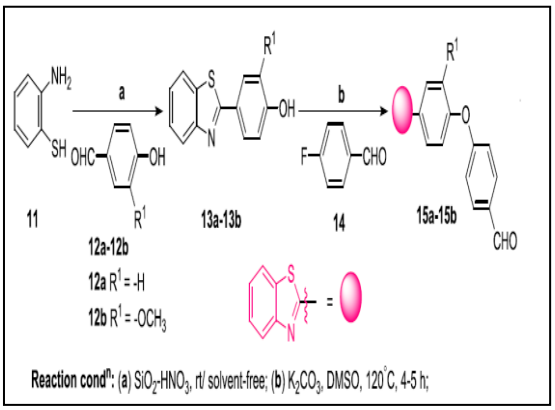
Figure: Crystal structure of compound 10c by single crystal X-ray diffraction study

All of the newly synthesized compounds were tested for their ability to inhibit in vitro -amylase activity at three different concentrations: 50, 25, and 12.5 g/mL. This was done so that the compounds' potential biological applications could be evaluated. The research discovered that a number of the synthesized substances had significant inhibitory effects. Combination of the numbers three and four -1'-methyl -4'-(p-tolyl) -1-((1-(p-tolyl) - 1H-1,2,3-triazol-4-yl)methyl)dispiro [indoline-3,2'-pyrrolidine-3',5''-thiazolidine] -2,2'',4''-trione (10a) demonstrated exceptional inhibition at concentrations of 50 g/mL, 25 g/mL, and 12.5 g/mL, with percentages of inhibition of 88.05%, 76.65%, and 68.57%, respectively. This is in comparison to the reference medication acarbose, which demonstrated inhibition at 77.96%, 71.17%, and 67.25%. Molecular docking is a method that may be used to study the possible binding modes of a drug in order to find the location of the active site of a target protein. This can be accomplished by determining where the active site of the target protein is situated. In order to produce an informed assumption as to the most probable manner of binding for the most effective chemical 10a, the molecular docking study was carried out using the Auto Dock Vina tool. This allowed for an accurate prediction.

Experiments using docking were done with compound 10a against the active site of the amylase enzyme discovered in Aspergillus oryzae (PDB ID: 7TAA). According to the findings of the docking experiments, the binding interactions that

were discovered between 10a (Arg344 and Asp297) and -amylase are very comparable to the interactions that are responsible for acarbose's ability to inhibit -amylase. Specifically, Arg344 and Asp297 play the same roles as Arg344 and Asp297 do in acarbose's ability to inhibit -amylase

In Chapter 4, the results of the synthesis of novel molecular hybrids of benzothiazole clubbed aroylhydrazones or 1,3,4-oxadiazoles are presented. These hybrids have benzothiazole groups attached to aroylhydrazones. (E)-N'-(4-(4-(benzo[d]thiazol-2-yl) aryloxy) benzylidene)-(aryl)hydrazides (17a-17v) and 2-(4-(4-(benzo[d]thiazol-2-yl) aryloxy) benzylidene)-(aryl)hydrazides (17a-17v) are examples of these one-of-a-kind mo Starting with commercially available 2-aminothiophenol (11), p-hydroxybenzaldehyde (12a) or vanillin (12b), and 4-fluorobenzaldehyde, an essential intermediate called 4-(4-(benzo[d]thiazol-2-yl)aryloxy)benzaldehyde (15a-15b) was produced through a two-step process. This essential intermediate is known as 4-(4-(benzo[d]thiazol In the first stage, p-hydroxybenzaldehyde (12a) was involved, while in the second phase, vanill was engaged (14). Synthesis of 2-arylbenzothiazole was achieved in a solvent-free environment at room temperature by condensing 2-aminothiophenol (11) with p-hydroxybenzaldehyde (12a) or vanillin (12b) using SiO2-HNO3 as a solid supported catalyst. This reaction was carried out with 2-aminothiophenol (11), p-hydroxybenzaldehyde (12a), or vanillin (12b) (13a-13b). To obtain 4-(4-(benzo[d]thiazol-2-yl)aryloxy)benzaldehyde (15a-15b), 2-arylbenzothiazole (13a-13b) was arylated with p-fluorobenzaldehyde (14) in the presence of potassium carbonate (K2CO3) in dimethyl sulfoxide (DMSO) at a temperature of 120 degrees Celsius for 4-5 hours. The reaction was carried out at the This procedure was carried out in the presence of (eq.1).



(1)

Following that, a condensation reaction was performed using 4-(4-(benzo[d]thiazol-2-yl)aryloxy)benzaldehyde (15a-15b) with corresponding aryl/heteroaryl hydrazide (16a-16k) in refluxing EtOH: THF (80:20, v/v) in the presence of a catalytic amount of acetic acid. This reaction produced N'-(4-(4- (benz The whole reaction proceeded smoothly, and as a result, between 77 and 90% of 2-(4-(4-(benzo[d]thiazol-2-yl)aryloxy)phenyl-5-(aryl)-1,3,4-oxadiazole (18a-18v) was generated.

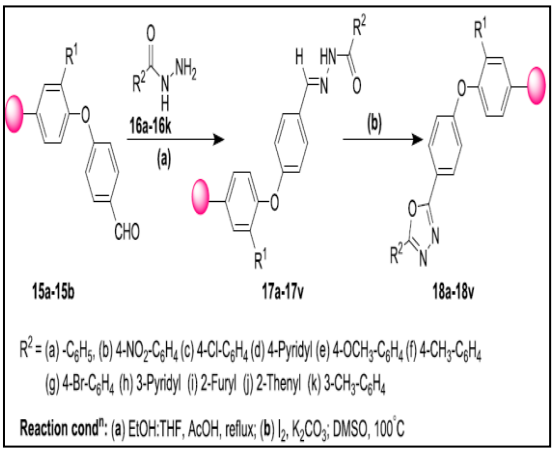


Figure: Synthesis of benzothiazole clubbed 1,3,4-oxadiazole based molecular hybrids (18a-18v).

All of the newly synthesized compounds were tested for their ability to inhibit in vitro -amylase activity at three different concentrations: 50, 25, and 12.5 g/mL. This was done so that the compounds' potential biological applications could be evaluated. The substances (E)-N'-(4-(benzo[d]thiazol-2-yl)-2-methoxy phenoxy)benzylidene)-4-chlorobenzo hydrazide (17n) and 2-(4-(benzo[d]thiazol-2-yl)phenoxy)phenyl) were present at a concentration of 50 g/mL. Both of these compounds were shown to have antitumor activity. -5-(p-tolyl) -1,3,4-oxadiazol The active site of the -amylase enzyme discovered in Aspergillus oryzae was used as a target in docking studies using compounds 17n and 18f (PDB ID: 7TAA). According to the results of the docking tests, the binding interactions that were identified between 17n (Tyr82, Arg204, and Glu230), and 18f (Lys209) and -amylase are highly analogous to the interactions that are responsible for acarbose's capacity to inhibit -amylase.

The multicomponent synthesis of molecular hybrids including pyrazole and thiazole moieties and using hydrazone as a linker is the subject of study in Chapter 5. This chapter is included in the book. The synthesis of 1-((1-phenyl-3-aryl-1H-pyrazole-4-yl) methylene)-2-(4-arylthiazole-2-yl)hydrazine (23a-23r), which was done by refluxing, is of particular interest (method-A). After five minutes of refluxing, a solid was formed; after that, it was separated from the ethanol by filtration and recrystallization. TLC using petroleum ether and ethyl acetate at a ratio of 70:30 (v/v) was carried out in order to track how far along the reaction had progressed while it was still active. Another strategy (method-B) was used in order to synthesize the chemical 23a. This method included the reaction of the thiosemicarbazone of pyrazole carbaldehyde (21a) that had been produced before with 2-bromo-1- (4-bromophenyl)ethan-1-one. During this procedure, we made use of a compound known as 2-bromo-1-(4-bromophenyl)ethan-1-one (22a). The steps involved in the reaction have been dissected, and Scheme 4 provides a summary of them.

Method B requires a great deal more effort, and the quantity of product 23a that it generates in the end is negligible. While method A has an overall yield of 85% for the chemical 23a, technique B only has a total yield of 65% for the substance. In order to investigate the applicability of multicomponent synthesis (method A, Scheme 4), several substituted carbaldehydes (19a-19f) are reacted with thiosemicarbazide (20) and -bromoacetophenones (22a-22c).

This is done so that the yield and the length of time necessary for the reaction may be determined. The various 1-phenyl-3-(aryl)-1H-pyrazole-4-carbaldehyde (19a-19f) underwent a smooth conversion to the respective products 23a-23r in outstanding yields, which led to the discovery that the multicomponent synthesis is more possible than had been previously anticipated. All of the structures of the compounds were verified after being analyzed using elemental and spectroscopic techniques, and they were applied to the newly created compounds (IR, 1H-NMR, MS).

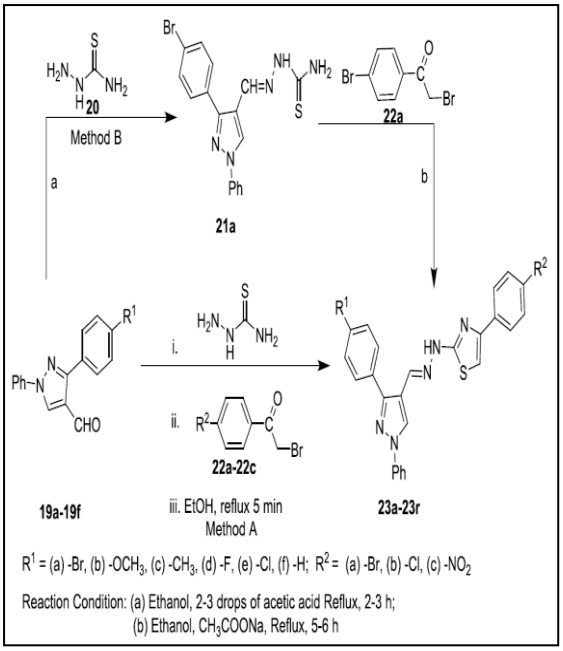


Figure. Synthesis of 1-((1-phenyl-3-aryl-1H-pyrazole-4-yl)methylene)-2-(4-aryl thiazole-2-yl) hydrazine (23a-23r)

The biological potential of all of the newly synthesized compounds was evaluated by measuring their ability to inhibit -amylase activity in vitro at three different concentrations (50, 25, and 12.5 g/mL). According to the results of the study, a handful of the compounds that were manufactured have detectable inhibitory characteristics. 1-((3-(4-methylphenyl)-1-phenyl-1H-pyrazole-4-yl))methylene is a chemical. 1-((3-(4-methylphenyl)-1-phenyl-1H-pyrazole-4-yl) methylene and 2-(4-(4-bromophenyl) thiazol-2-yl)hydrazine (23g) It was established that a dosage of 50 g/mL of 2-(4-(4-chlorophenyl)thiazol-2-yl)hydrazine (23h) was the most effective dose, with an inhibition rate of 89.15% and 88.42% respectively. The compounds 23g and 23h were evaluated for their potential to dock with the amylase active site in Aspergillus oryzae (PDB ID: 7TAA). The findings of the docking experiments indicate that the binding interactions between -amylase and residues 23g (Tyr82) and 23h (His210 and Tyr82) are comparable to the interactions that are responsible for acarbose's ability to inhibit -amylase.

Using the approach of molecular hybridization, a total of 91 heterocyclic compounds combining nitrogen and sulfur have been developed. This brings the total number of such compounds to 91. Thiazolidine-4-one, 2,4-thiazolidinediones, thiazole, and benzothiazoles coupled with pyrazole or oxadiazole are examples of the chemicals that fall under this category. These heterocyclic compounds are formed by combining a variety of pyrazole or oxadiazole groups in a clubbed configuration. At a range of different doses, each of the molecular hybrids was evaluated to see how well it performed against the -amylase produced by Aspergillus oryzae. It was shown that 5-((3-(4-methoxyphenyl)-1-phenyl-1H-pyrazol-4-yl)methylene)-2-(p-tolylimino) thiazolidin -4-one (5a, Scheme 1) had the highest rate of inhibition, which was 90%. This chemical was the most effective of these substances.

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