INTRODUCTION:

1.1 Indane-1,3-dione:

Indane-1,3-dione is the compound having benzene fused with cyclopentane-1,3-dione.

The last two decades have witnessed profound changes in indane-1,3-dione chemistry in both quality and quantity. Synthesis of compounds in a number up to now unexplored fields has been developed. Some old problems have been reconsidered. Physiochemical methods and quantum-chemical calculations have been extensively used.

Indane-1,3-dione constitutes a unique group of compounds due to the simultaneous presence of three characteristic features.1

- Enormous synthetic possibilities offered by the presence of β-dicarbonyl derivatives often serve as the starting material for more complex chemical structures.

- Specific physiochemical properties, which offer a wide scope for studies in the problems of theoretical organic chemistry, particularly based on indane-1,3-dione tautomerism, dual reactivity etc.

- A wide range of biological activity, covering analgesic, anti-inflammatory,1-7 anticoagulant,8-10 anticancer,11,12 antipyretic,13-15
antimicrobial,\textsuperscript{10,16} antihistaminic activity,\textsuperscript{17-19} herbicidal activity,\textsuperscript{20} antihyperlipidemetic activity.\textsuperscript{21} Binding studies\textsuperscript{22-26} were also carried out for the derivatives of indane-1,3-dione. Several scientists were working to synthesize various derivatives of indane-1,3-dione\textsuperscript{27-57} indicating the importance of the nucleus.

1.2 Heterocyclic compounds:

Heterocycles occur in array of natural products and are of great importance in wide variety of applications. They play a vital functional role in organic chemistry, which has valuable synthetic capabilities. In particular, it can necessitate new potentials for drug design and medicinal chemistry. The remarkable ability of the heterocyclic nuclei to serve as active pharmacophores has largely contributed to their unique value as traditional key elements of numerous drugs. Among the heterocycles, five, six and seven member nitrogen-containing heterocycles are especially important, since they have a variety of bioactivities for the drug synthesis.

1.2.1 Benzodiazepine:

Benzodiazepine nucleus is a pharmacophoric scaffold\textsuperscript{58} and many benzodiazepines have recently received great attention because of wide range of therapeutic and pharmacological properties. Many members of the benzodiazepine family are nowadays widely used as analgesic and anti-inflammatory,\textsuperscript{58-63} anticancer,\textsuperscript{64-69} anticonvulsant activities.\textsuperscript{70-72} Because of wide range of biological applications, the development of mild efficient and environmentally friendly protocols continues to be a challenging endeavor in synthetic organic chemistry.
As a result, considerable attention has been drawn recently to new improved methods for the preparation of 1,5-benzodiazepines.

**1.2.2 Pyrimidine and thiopyrimidine:**

Pyrimidine derivatives in medicinal chemistry have been well known for their therapeutic application. One possible reason for their activity is the presence of a pyrimidine base in thymine, cytosine and uracil, which are essential building blocks of nucleic acids, DNA, and RNA. One important class of pyrimidine is 2-thiopyrimidine and its derivatives, which are also called as 2-mercaptopyrimidine compounds. The sulfur in 2-thiopyrimidine act as an interesting replacement for the existing oxygen atom bonded to C-2 in uridine base. Considering this assumption 2-thiopyrimidine has attracted substantial interest of synthetic biochemists. The literature indicated that compounds having pyrimidine nucleus possess broad range of biological activities like analgesic and anti-inflammatory, anticancer, antimicrobial activities, etc. Thiopyrimidine also possess biological activities like antiparkinsonism, analgesic and anti-inflammatory, antioxidant, antimicrobial activities, anticancer, and anti-HIV activity.

**1.2.3 Oxazole:**

Oxazole is a five-member heterocyclic ring containing azole and oxygen at 1 and 3 positions. *Ab initio*, quantum mechanical studies indicate that the heterocyclic lone pair plays a decisive role in directing binding reaction with receptor. Oxazole derivatives possess good analgesic, antioxidant, and anti-inflammatory activity.
antimicrobial,\textsuperscript{121-124} anticonvulsant,\textsuperscript{125} cardiovascular activities,\textsuperscript{126} and anticancer activities\textsuperscript{127,128}.

1.3 Pharmacological activity:

1.3.1 Anti-inflammatory and Analgesic activity:

Inflammation is a complex defensive mechanism of the body to any noxious stimulus. This process may vary localized to a generalized response characterized by accumulation of fluids and leukocytes leading to edema and pain. Different physiological and immunological mediators mediate the inflammatory response. These mediators play a major role in inflammation. Inflammation is of two types; one is acute inflammation and other is chronic inflammation.\textsuperscript{129} Acute inflammation occurs as the initial response to tissue injury, which is mediated by autacoids like prostaglandins, histamine, bradykinin, and leukotrienes. The chronic inflammation involves release of mediators as interleukins, interferon and tumor necrosis factor $\alpha$.

Prostaglandins are lipid mediators. These mediators are short lived. They exert biological activity by activating G-protein coupled receptor. They are formed from arachidonic acid. Arachidonic acid attain hairpin configuration to access to cyclooxygenase (COX) through a hydrophobic channel. Non-steroidal anti-inflammatory drugs are competitive inhibitors at the COX site. Two types of COXs have been identified as COX-1 and COX-2. COX-1 is involved in gastric cytoprotection and haemostatasis. COX-2 is regulated in its expression by cytokines and mitogens, and it is largely account for prostaglandin formation in inflammation and cancer. COX-1 derived
prostaglandins can contribute to inflammation. This served as a basis for the development of selective COX-2 inhibitors. Up regulation of COX-2 in healing ulcers and evidence from the use of COX-2 inhibitors might delay the healing of existing ulcers. Therefore, selective inhibitors of COX-2 have showed efficacy in the treatment of pain and inflammation when compared to Non-steroidal anti-inflammatory drugs.

Tumor Necrosis Factor is a key signaling protein in immune system. TNF is best known for immune defenses to protect a localized area from injury. Anti-TNF therapy can reduce the deceased associated with inflammation. TNF acts on two receptors TNFR1 and TNFR2. TNFR2 pathway serves as a therapeutic target for several autoimmune diseases. It has been demonstrated that TNF-α production in proinflammatory cells becomes attenuated by an elevation of intracellular cyclic adenosine 3′,5′-monophospate. The phosphodiesterase family regulates this secondary messenger. It was found that indane-1,3-dione is found to inhibit the calcium independent and cyclic adenosine 3′,5′-monophospate specific PDE4.

The antithrombotic drugs must have a rapid onset of effect with minimal effect on bleeding and should be compatible with thrombolytic agents. Such agent could have good anti-inflammatory properties and thereby reducing in neuronal damage associated with ischemia-riper-fusion injury.
1.3.2 Antimicrobial:

In the 1970s, it was believed that any microbial infection could be treated by wide antimicrobial agents (antibiotics) were available. The belief was short lived, when pathogens resistant to the conventional antibiotics used to treat microbial infections emerged. The use of antibiotics allowed many microbial strains to show ways to adapt or become resistant to the present available regimens. Resistance to antimicrobial agents by antimicrobial agents is rapidly becoming a major worldwide problem. The design of new compounds to deal with bacteria has become one of the most areas of antibacterial research today. Biochemical similarity of the human cell and fungi forms a handicap for selective activity. It also easily gained resistance which is the main problem encountered in developing safe and efficient antifungal agents which resulting in urgent need for new antimicrobial drugs. Many dyes are classified as antimicrobial functions and some of these have recently been shown to possess remarkable antimicrobial activity. Indane-1,3-dione derivatives possess dye character due to un-saturation. The dye containing heterocyclic moiety have been found to have good range of antimicrobial activity.

Considering this importance in mind, it was planned to evaluate the heterocyclic derivatives of indane-1,3-dione derivatives for antimicrobial activity.
1.4 In silico screening:

1.4.1 Docking:

The docking process involves prediction of ligand confirmation and orientation or posing with a targeted binding site. Two aims of docking studies are:

a. Accurate structural modeling
b. Correct prediction of activity.

Identification of molecular features was responsible for specific biological activity or the prediction of compound modification that improve potency. Docking begins with application of algorithms that pose molecule, which are small to the active site. Sampling was performed with accuracy to identify the best confirmation that matches the receptor structure. Algorithms are complemented by SCORES, which are designed to predict the biological activity to the evaluation of interaction between compounds and the potential target. Simple scoring functions are used in the early stages of docking simulations. The ligand binding events are driven by the combination of enthalpic and entropic effect. Either enthalpy or entropy can modify interactions.

The three basic representation of receptor are atomic, surface, and grid. Among these three atomic representation is generally used in conjugation with potential energy function only during final RANKING procedures.

Treatment of ligand flexibility can be divided into three basic categories like systemic methods, random methods and simulation
Various approaches have been applied to be flexible for the target protein. Finally, the scoring evaluation and ranking of the predicted ligand confirmation is a crucial aspect of structure based virtual screening. Three types of scoring functions are currently applied. They are force field based, empirical based and knowledge-based scoring functions. Ligand flexibility has a greater effect on predicting structures correctly than size of polarity. The interplay between docking and scoring function is complex. It is often easier to produce reliable models of ligand, which makes it possible to distinguish between true ligand and false ligand.

**1.4.2 Physicochemical properties:**

Passive diffusion across biological membrane is governed by physicochemical parameters like lipophilicity (log P and log D), Polarity (charge, hydrogen bond) and molecular volume. Lipinski et al., proposed a series of rules imposing limitation on log P (octanol/water partition coefficient), molecular weight, number of hydrogen bond acceptors and donors’. This rule is known as rule of five. The rule states that most drug like molecules have

1. Log P ≤ 5
2. Molecular weight ≤ 500
3. Number of hydrogen bond acceptors ≤ 10
4. Number of hydrogen bond donors ≤ 5

Molecules violating more than one of these rules may have problems in bioavailability.
The Possible values for some descriptors to generate a drug-like collection are,

- Not more than one violation of Lipinski’s rule of 5
- Total polar surface area not more than 160
- Not more than 50 rigid bonds
- Not more than four ring system
- Not more than 35 heavy atoms
- Not more than 20 hetero atoms
- Formal charge within 0-3
- Sum of formal charge within -2 to 2
- Eliminate compounds having undesirable atom types and unacceptable functional groups.

1.5 Formulation:

It is a process in which different chemical substance, including active drug, are combined to form the final medicinal product. There are different types of formulation. One of the types of formulation is ointment.

Ointment:

It is one of the oldest types of medicament. The active medications can be applied and rubbed in the skin and the therapeutic agent present in it is absorbed systemically. It is a medicament intended to reach the epidermis, corium, subcutaneous fat or to be absorbed by the blood stream. All ointment consists of base, which act as a carrier for the therapeutic agent. The nature of the base also controls the
performance of ointment. The affinity between the drug and base
determine the release rate of the drug.

We are interested in the structural modification of indane-1,3-
dione ring system as a part of the development of good analgesic, anti-
inflammatory and antimicrobial drug. Since none of the drugs possess
these three activities in a single component, our aim is to find a
compound having analgesic, anti-inflammatory and antimicrobial
activities and formulate the best-hit molecule.

It is a well-known fact that the heterocyclic backbone could act on
various pharmacological targets and that display analgesic, anti-
inflammatory and antimicrobial activities. Combination of two or more
biologically active moieties increases or decreases the biological
activity. With this aspect in mind, we have worked on indane-1,3-
dione by substitution of various functionalized heterocyclic ring
system like benzodiazepine, pyrimidine, thiopyrimidine and oxazole
derivatives. These heterocycles are active pharmacophores in several
pharmacologically important molecules. In view of the importance of
benzodiazepine, pyrimidine, thiopyrimidine and oxazole moieties, it
has been considered of interest to incorporate this moiety in indane-
1,3-dione as a substituent in 2-position to form new hybrids of
indane-1,3-dione.