SUMMARY AND CONCLUSIONS

The urgent need for the discovery highly effective antimalarial, with the importance of computational tools in designing of hybrid molecules, having amalgamation of two different chemical entities together in a single compound, prompted us to carry out this study. Besides that, the literature survey also revealed some important findings such as, the merging of heterocyclic moieties such as furanone, quinoline and pyrazole can leads to compounds with improved antimalarial activity, particularly against the resistant strains of the *plasmodium falciparum*.

In accordance to the above, the primary aim of this study was to design and synthesize some butenolide derivatives mainly based on furanone as new antimalarial agents. These derivatives were actually designed as hybrid compounds, having furanone as core moiety, which was further hybridized with pharmacophore mainly quinoline and pyrazole. The designed set of hypothetical compounds were then virtually screened and optimized on the basis of *in silico* approaches. After that the screened potent hits were synthesized and evaluated for the antimalarial activity using suitable *in vitro* assay methods. The process of achieving this aim involved the following steps:

1. Designing the library of hypothetical ligands having furanone fused with quinoline and pyrazole moieties.
2. Optimization of the ligands, using computational approaches such as Molecular docking and *in silico* ADME-T prediction, to screen out the best potent hits having “drug-likeliness” and nontoxic profile having good binding affinity with the receptor, better physiochemical (“drug like”) properties and less toxic profile.
3. Synthesis, purification, structural elucidation and confirmation of the new intermediate and final compounds using IR, $^1$H and $^{13}$C NMR, mass spectroscopy and elemental analyses.
4. Evaluation for *in vitro* antimalarial activity of the final compounds, in comparison to that of Chloroquine (CQ) as standard drug.

The whole work embodied this thesis has been represented in the form of five chapters. Each chapter is described in brief as below

**CHAPTER 1**

The first chapter presents a brief introduction of malaria, including general description like malaria burden till 2015, factors affecting malaria distribution, antimalarial drugs
and drug resistance. A brief discussion on causative parasite *Plasmodium*, its pathology especially for *P. falciparum* and pathogenesis behind sever complications like cerebral malaria is also given. This chapter also highlights the origin of the current study conducted, including the importance to explore the new pharmacophore and parasite specific drug targets.

**CHAPTER 2**

The second chapter covered the literature review based on the recent studies conducted to synthesize and various heterocyclic derivatives as antimalarials, specially based on furanone, quinoline and pyrazole moieties or their combination.

**CHAPTER 3**

The third chapter included the designing and virtual screening of the hypothetical butenolide (furanone) derivatives on the basis of *in silico* studies. The designed compounds were first subjected to the in *silico* ADME-T studies, to check whether these derivatives posses the “Drug-like” behavior or not. After that the molecular docking simulations were also carried out to pre-asses the binding mode of these derivatives against the 3D-crystal structure of the parasite specific receptor, enzyme *Plasmodium* Lactate dehydrogenase (PDB- 1LDG). The flow of virtual screening is shown below.

**LIBRARY GENERATION OF LIGANDS**

**ADME- TOXICITY FILTER**

**MOLECULAR DOCKING ANALYSIS**

**FINAL 29 HITS**

FLOW OF VIRTUAL SCREENING
CHAPTER 4
In this chapter it was described that from the library of total 64 hypothetical compounds, in silico ADME-T prediction and molecular docking analysis and the virtual screening process, left us with 31 best hits, which were synthesized in the laboratory. The synthetic route for the preparation of potent screened derivatives, 3-{(2-chloro-6-substituted-quinolin-3-yl)methylene}-5-(aryl-2-yl)-furan-2(3H)-one and 3-{(4-Substitutedphenyl-1-phenyl-1H-pyrazol-4-yl)methylene}-5-(aryl-2-yl)-furan-2(3H)-one has been illustrated in this chapter step wise. The synthesized derivatives were then purified and characterized on the basis of IR, \textsuperscript{1}H and \textsuperscript{13}C NMR, mass spectroscopy and elemental analyses.

CHAPTER 5
The evaluation of in vitro antimalarial activity of the synthesized derivatives was described in this chapter. The complete protocol has been discussed step wise like in vitro cultivation of \textit{P. falciparum}, maintenance of the grown culture and estimation of antimalarial activity.

Estimation of in vitro antimalarial activity was made against chroquine resistant strain (K1), using mainly two methods, Schizont maturation assay and Parasite lactate dehydrogenase assay. The test compounds were exhibited moderate to good antimalarial activity as compared to the standard drug Chloroquine

CONCLUSION
Overall study, wind up with the conclusions that the compounds showed good antimalarial activity with \textit{Pf}LDH inhibition. The results obtained suggest that compounds with activity in low micromolar range could be used as lead compound for development of more potent antimalarial agents. The \textit{in silico} analysis further confirm the drug like behavior of these final compounds, good binding affinity for target receptor pLDH. Binding mode analysis revealed that the docked ligands were exhibited binding with the same active site residues, as reported for well known inhibitor NADH and CQ. This observation suggested that the designed furanone hybrids may act as potential inhibitors against \textit{Pf}LDH, as new antimalarial potential agents. The best compounds can be explored further in future for designing the potent inhibitors of \textit{Pf}LDH as new potent antimalarial agents.
FUTURE PROSPECTIVE:

It is envisaged that on completion of these studies on each of the series of compounds, the following benefits will accrue for future use:

(a) New chemical entities or lead compounds could be added to the pool of bioactive compounds,

(b) New or improved synthetic pathways could be developed in the area of malaria chemotherapy and related fields.

(c) On the basis of molecular docking studies and biological evaluation data, these compounds may act as template for the development of new antimalarial drugs related to these compounds.