Instructions before you submit your thesis to supervisor

Before you submit your thesis to me for revision, take note of the followings:

(1) ALL references are correct – refer to my Endnote guidelines (a summarized version is below as a quick kick)
(2) You know how to use Punctuations – see punctuation guidelines below
(3) The structure of your thesis is well organized – see template below
(4) The first chapter (Chapter 1) and last chapter (Conclusion) are crucial – see templates below
(5) Make sure you double check your thesis for grammar & typo mistakes
(6) The Plagiarism report created by "**turnitin**" is enclosed with your thesis (read instructions on the lab website on how to use **turnitin**)

Important: If you have published or accepted paper(s) DO NOT submit it to turnitin

Quick guide on how to use Endnote with your thesis

You normally write each chapter separately with its own references (its own endnote library). When you finish all chapters, you will need to merge all chapters in a single word document (combined thesis), however, this is not an easy task and some tricks are there. To make sure that each chapter will have its own references starting from ref no. 1, you need to follow these simple steps that I summarized for you to make the task easy:

1- At the end of your first chapter (after the references), put the mouse cursor in an empty space and then click on "insert" menu --> "break" --> "section break (continuous)

2- Then you can copy your second chapter and paste it after the first chapter (after the break you just inserted)

3- Do step 1 again at the end of chapter 2 to paste chapter 3 and so on.

4- In Endnote, go to "Edit" menu, "output style", then select your style (I prefer Biochemistry style, so we all should stick to this), "Edit" , go to "Sections" and tick the second option "separate sections"

This should produce a single Word document containing separate chapters, each with its own bibliography

Punctuations guidelines

A Rough Guide to Punctuation

The Learning Centre • http://www.lc.unsw.edu.au

Good punctuation is crucial for successful academic writing. Many students' essays use little punctuation beyond commas and full stops. But to be restricted to just two forms of punctuation mark, when writing your essay, is like building a house using only a hammer and a saw: you can do it; but not very well. By learning to use more, or all, of the available forms of punctuation you will be able to communicate and express your ideas, and arguments, more clearly.

Full Stop (.) Full stops have three distinct uses: 1. To mark the end of a sentence 2. To indicate abbreviated words (unless first and last letters of the word are shown). 3. To punctuate numbers and dates	**Examples:** *The cat is completely black.* *The teacher will be John Smith (B. Sci.).* *All assignments should be submitted by 6. 6. 03.*
Colon (:) A colon can be used to indicate that a list, quotation or summary is about to follow. A colon can also be used to separate an initial sentence/clause from a second clause, list, phrase or quotation that supports the first in a particular way.	**Examples:** *Buy these things: a packet of peanuts, two loaves of bread and a kilogram of steak.* *Writing an assignment is not easy: to begin with you have to do a lot of research.* *The Television set, as the icon of the information age, represents the realisation of a dream for humankind: that knowledge and experience can be transmitted and shared across the boundaries of time and space.*
Semi-Colon (;) Separates two complete sentences that are, however, closely linked. The semi-colon can be replaced by a full stop, but the direct link between the two parts is lost. A semi-colon also serves as a second level of punctuation in a series of words or phrases which already have commas, making some internal divisions.	**Examples:** *To err is human; to forgive, divine.* *Don't go near the lions; they could bite you.* *She came out of the house, which had a long drive, and saw him at the end of the path; but instead of continuing towards him, she hid until she left.*
Comma (,) Commas are used in longer sentences to separate information into readable units. A single comma ensures correct reading of a sentence which starts with a long introductory element. Pairs of commas help in the middle of a sentence to set off any string of words which is either a parenthesis, or in contrast, to whatever went before. Sets of comma act as a means of separating items in a list	**Example:** *When Australia celebrated its sesquicentenary in 1938, there was a little of the confidence or enthusiasm of the centennial celebrations of 1888.* *Yet in representing ourselves to ourselves, as film and television and television do, these media are constantly introducing and reinforcing the assumptions.* *Ward traced the origins of the type through the common man's response to the bush, through convicts, outback workers, gold diggers, trade unions, and the* Bulletin.
Question Mark (?) A question mark is used at the end of a sentence which is a question.	**Example:** *Have the students completed the exam?*

Apostrophe (‘) There are two uses for the apostrophe: **1. Contractions** A contraction is a shortened version of a word. An apostrophe is used to show that something has been left out, and where it has been left out. 2. Possessives An apostrophe is used to indicate ownership/ possession with nouns. To show ownership by a single individual, insert the apostrophe between the noun and the ‘s’. To show ownership by more than one individual, use the apostrophe at the end of the word. **Be careful: It’s** is the contraction of ‘it is’. ***It’s*** is not a possessive (a possessive denotes ownership).	**Examples:** *don’t (do not) It’ll (It will) she’ll (she will)* *It’s too cold to go swimming today.* *I don’t think she’ll come to the party.* *the dog’s tail* (belonging to a single dog) *the women’s magazines* *boys’ football boots* (belonging to more than one boy) *Einstein’s theory of relativity* *The dog is chasing its own tail!*
Hyphen (-) When used correctly, a hyphen links two or more words, that normally would not be placed together, in order that they work as one idea and these are called compound nouns.	**Examples:** *Stonier’s post-industrial economy is a service economy.* *There are four types of information-related machines.*
Dashes (—) Hyphens are not the same as dashes. Dashes are like brackets; they enclose extra information. A colon and semi-colon would work just as well in the example opposite. Dashes are rarely used in academic writing. Although often used in pairs, dashes can also be used singularly.	**Examples:** *To the three divisions of the economy—agriculture, manufacturing, and service industries—Jones has added a fourth.* *Have an orange—or would you prefer a banana?* *While the importance of sport to Pay TV is clear, the opposite perspective is less certain—the importance of Pay TV to sport.*
Parentheses ( ) Parentheses are brackets used to include extra or nonessential material in sentences. Parentheses should be used sparingly and always appear in pairs. In citation systems like Harvard, parentheses are used to include in-text references.	**Example:** *It was unusual to see Paul awake so early (as he often studied late into the night) and Jane greeted him with amazement.* *Larsen and Greene (1989) studied the effects of pollution in three major cities.* *“Australia is a settler society” (Hudson & Bolton 1997, p. 9).*
Exclamation Mark (!) An exclamation mark is used at the end of a sentence and indicates surprise, anger, or alarm. Exclamation marks should be used very sparingly and are not often used in academic writing.	**Example** *The police stormed in and arrested her!* *How disgraceful!*
Ellipsis (…) An ellipsis consists or three full stops. It indicates that material have been left out of a quotation. When quoting, it is sometimes necessary to leave out words or lines for reasons of relevance or length. Using an ellipsis makes any omissions known to your reader.	**Example:** *“But to be restricted to just two forms of punctuation mark ... is like building a house using only a hammer and a saw: you can do it; but not very well.”*

An example of combined thesis (with the cover pages)

is below

UNIVERSITY OF KWAZULU-NATAL

TITLE OF YOUR THESIS HERE

2013

YOUR NAME HERE

TITLE HERE

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2012

A thesis submitted to the School of Pharmacy and Pharmacology, Faculty of Health Science, University of KwaZulu-Natal, Westville, for the degree of XXXXXX.

This is the thesis in which the chapters are written as a set of discrete research publications, with an overall introduction and final summary. Typically these chapters will have been published in internationally recognized, peer-reviewed journals.

This is to certify that the contents of this thesis are the original research work of Mr/Mr. YOUR NAME.

As the candidate's supervisor, I have approved this thesis for submission.

Supervisor:

Signed: ---------- Name: **Dr. Mahmoud Soliman** Date: ----------

Abstract is really a mini thesis (2 pages maximum) and has to answer the following specific question:
What was done? Why was it done? How was it done? What was found? What is the significance of the findings?

ABSTRACT

Very concise Introduction

In the African continent, the AIDS epidemic has reached soaring numbers. Of the 42 million individuals infected with HIV worldwide, 30 million are in Africa. It is believed that an important contributing reason to this soaring figure is that the HIV-1 subtypes prevalent in Africa have not been paid as adequate attention from researchers as those subtypes in North America and Western Europe. Out of the major two subtypes of HIV-1, subtype C accounts for about 95 % of HIV infections in South Africa.

The HIV -1 protease enzyme slices the *gag* and *pol* nonfunctional polypeptide into functional proteins essential for development of mature infectious HIV particles, therefore, it is considered a strategic target for designing of anti-AIDS inhibitors. Therefore, we believe that molecular insight into drug binding to this enzyme will enable us to understand the mechanisms by which the enzyme develops resistance. The current study will be focusing on the South African subtype C HIV PR (C-SA HIV PR) since the inhibitory profile of the current FDA-approved drugs has been lacking in the literature.

Methods used and results/findings

In this thesis, we report the first account of a comparative analysis of the binding affinities of nine FDA approved drugs against South African subtype B as well as subtype C HIV PR (C-SA) using molecular dynamics simulations (MD) and binding free energy calculations. The computed binding free energies showed a similar trend to the experimental binding free energies. Decomposition of the calculated binding free energy provided insight into the favourable modes of interactions of the inhibitors with the active site and the flap regions of the PR. Results confirmed that, by themselves, natural polymorphisms exist in the C-SA HIV-1 PR do not significantly interfere with the FDA-approved drug binding when compared to subtype B.

Also via molecular dynamics simulations and binding free energy calculations, the impact of active site drug-resistant mutations of C-SA PR, V82A and V84F/I84V, on drug resistance was investigated. Unique per-residue interaction energy “footprints” were developed to map the individual amino acid contribution to the overall drug binding profile for the wild type and mutants. As evident from the MD analysis, these mutations

have modified the overall binding landscape of the amino acid residues not only in the active site region but in the flaps as well. Four FDA-approved drugs were investigated in this work - ritonavir, saquinavir, indinavir and nelfinavir. Computational results were compared against experimental findings and found to be complementary. Against the V82F I84V variant, saquinavir, indinavir, and nelfinavir lose remarkable entropic contributions relative to both wild type and V82A C-SA PRs. The per-residue energy "footprints" and binding forces analysis for the drug complexes with the wild type and mutants has also highlighted the nature of drug interactions.

The significance of your study in general in the field (will it assist medicinal chemist to discover new drugs, you come up with new techniques, …..etc) should be written at the end of your abstract

DECLARATION

I, **your name here**, declare that

1. The research reported in this thesis, except where otherwise indicated, is my original research.

2. This thesis has not been submitted for any degree or examination at any other university.

3. This thesis does not contain other persons' data, pictures, graphs or other information, unless specifically acknowledged as being sourced from other persons.

4. This thesis does not contain other persons' writing, unless specifically acknowledged as being sourced from other researchers. Where other written sources have been quoted, then:
   a. Their words have been re-written but the general information attributed to them has been referenced
   b. Where their exact words have been used, then their writing has been placed in italics and inside quotation marks, and referenced.

5. This thesis does not contain text, graphics or tables copied and pasted from the Internet, unless specifically acknowledged, and the source being detailed in the thesis and in the References sections.

A detail contribution to publications that form part and/or include research presented in this thesis is stated (include publications submitted, accepted, in *press* and published).

Signed ----------

LIST OF PUBLICATIONS

Put your "Published/accepted/submitted and under review" here (You need to mention the name of journal where it is published/accepted/ submitted)

ACKNOWLEDGEMENTS

Acknowledgments go here

sample of table of contents

TABLE OF CONTENTS

Chapter 1 is very crucial and it has to address all the points below

You start with a brief introduction about your study problem and the methods that you are going to use to investigate your problem

CHAPTER 1

1.1. Background and rational of this study

The work in this thesis is an effort towards understanding the challenges of HIV/AIDS drug treatment. In Africa, the AIDS epidemic has achieved dramatic figures. Of the 42 million individuals infected with HIV worldwide, 30 million are in Africa (*1*). It is believed that an important reason that might conceivably contribute to this soaring figure is that the HIV-1 subtypes prevalent in Africa have not received as adequate attention from researchers as those subtypes in North America and Western Europe (*2*). Out of the major ten subtypes of HIV-1, subtype C accounts for about 95 % of HIV infections in South Africa (*3*). Most of HIV-1 antiretroviral treatment has been developed and evaluated against subtype B, which is prevalent in North America, Western Europe and Australia. Besides, the limited data available reveals that these protease inhibitors exhibit reduced activities against the A and C subtypes, and this indicates that the therapy for non-subtype B viruses may be less effective. This apparent gap of information on the effectiveness of current HIV therapy against subtype C, including the South African variant, prompts us to conduct this current study.

The HIV -1 protease enzyme, which slices the *gag* and *pol* nonfunctional polypeptide into functional proteins essential for development of mature infectious HIV particles (*4*), is considered a crucial target for designing of anti- AIDS inhibitors (*5*). Therefore, It is believed that molecular understanding of this enzyme will enable us to design more potential inhibitors. The current study focuses on the South African subtype C HIV PR (C-SA HIV PR) (*3*) since the inhibitory profile of the current FDA-approved drugs has been lacking in the literature.

Sequence data from the National Institute for Communicable Diseases (NICD, South Africa) (*6*) have revealed the presence of the V82A mutation in PR inhibitor-treated patients. Also, the double mutation V82F/I84V is a widespread primary resistant mutation reported in clinical isolates and is known to confer cross-resistance to all the inhibitors currently in clinical use (*7-9*). Therefore, in this work, these mutations within

the frame of the C-SA HIV PR to obtain more insight into the nature of the resistance were investigated.

Computational tools, especially molecular dynamics (MD) simulations, are considered very useful in estimating ligand binding affinities and investigating the origin of drug resistance. Several methods can be used for this purpose (*10*). These include, thermodynamic integration (TI), linear response (LR), free energy perturbation (FEP) (*11*), fluctuation–dissipation theorem (FDT) (*12*), and molecular mechanics generalized Born surface area (MM-GBSA) (*13, 14*). In this thesis, MD combined with MM-GBSA is used in order to obtain insight into drug binding and the origin of resistance.

1.2. Aims and objectives of this study

This is the first aim of your study and you split this aim into objectives. Aims & objectives should be specific. Do not mention any technical steps or software etc rather put it as an outcome

This study has two major aims:

aim #1

1. To investigate the effectiveness of the FDA-approved drugs against the subtype C HIV PR.

To accomplish this, the following objectives were outlined:

objectives

1.1. To estimate the drug binding free energies against subtype B and C-SA HIV PR
1.2. To obtain insight into the impact of natural polymorphisms exist in the C-SA HIV PR enzyme on drug binding

aim #2

2. To provide a molecular understanding on the origin of drug resistance against the clinically reported mutations, single mutation V82A and double mutation V82F/I84V.

To accomplish this, the following objectives were outlined:

objectives

1.1. To estimate the drug binding affinities and entropic contribution in case of the wild type C-SA HIV PR, single mutation V82A and double mutation V82F/I84V.
1.2. To estimate the contribution of each amino acid towards the overall binding to the drugs.

highlight why your study is important, did it fill up a gap in literature, did you come up with new approaches ,etc

1.3. Novelty and significance of this study

Only two previous studies have reported the experimental binding free energy of four of the FDA-approved anti HIV drugs against subtype C (*3, 15*). However, no computational investigations have been performed to date to support these experimental findings. The current study, until now, is the first and only study that reported the relative binding affinities of the nine FDA-approved anti-HIV drugs against both subtypes C and B using computational approaches.

Another important aspect, which highlights the novelty of this work, is that the lack of the X-ray crystal structure of the C-SA PR enzyme. Until late 2012, the X-ray structure of this enzyme was not available, therefore we built a "validated" computational model based on the protein amino acid sequence. Although the X-ray crystal structures was later revealed, we opted to still use the validated computational enzyme model for several reasons (see Chapter 4 and 5 for details).

To this end, the work presented in this thesis is considered a key cornerstone towards further understanding of HIV therapy, in general, and the South African version, in particular. Also, this study could serve as a road map for further HIV drug design and development

1.4. Overview of this thesis

This explain the overall structure of your thesis

This thesis is divided into six chapters, including this one:

Chapter 1: this chapter addresses the background, aim and objectives, significance as well as the general outline and structure of the thesis.

Chapter 2: it provides a general overview on the HIV/AIDS epidemic and therapy. The chapter starts with a historic background on HIV/AIDS epidemic then some updated statistics on the number of HIV infected peoples worldwide and in Africa follows. The chapter also highlights many aspects such as HIV virus structure, life cycle and the essential enzymes required for virus maturations, HIV drug targets, drug resistances and side effects. The HIV PR enzyme, as a crucial drug target and the main focus of the work, is then addressed in details including its structure, different mechanisms of actions,

inhibitor design strategies and the currently approved FDA drugs. A brief overview on the C-SA PR enzyme variant ends the chapter.

Chapter 3: this chapter provides a general introduction to computational chemistry and different molecular modeling and simulations techniques and their applications. Whenever possible, some theoretical descriptions of the computational methods have been explained. This is followed by a highlight on the various computational tools used in HIV research with main focus on molecular dynamics simulations, hybrid quantum mechanics and molecular mechanics (QM/MM), molecular docking and binding free energy calculations. Some significant previously reported computational studies on HIV PR were also included at the end of this chapter.

Chapter 4: (Published work)

This is a research paper from this study. The paper is entitled "Comparison of the molecular dynamics and calculated binding free energies for nine FDA-approved HIV-1 PR drugs against subtype B and C-SA HIV PR" and published in the journal of Chemical Biology and Drug Design (an ISI journal, IF = 2.4). It addresses the objectives 1.1 and 1.2.

Chapter 5: (Manuscript submitted and under review)

This is a research paper from this study. The paper is entitled "The impact of active site mutations of South African HIV PR on drug resistance: insight from molecular dynamics simulations, binding free energy and per-residue footprints" and submitted to the journal of Chemical Biology and Drug Design (an ISI journal, IF = 2.4). It addresses the objectives 2.1 and 2.2.

Chapter 6: This expounds the concluding remarks and future plans and recommendations.

References: 

Follow this reference format (Use Biochemistry style in your Endnote) - some example below. Check consistency, journal abbreviations, etc as discussed in our Endnote workshop

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CHAPTER 2

2. INTRODUCTION TO HIV/AIDS & DRUG THERAPY

Your chapter 2 (about your disease, proteins, drugs, etc) here followed by references

CHAPTER 3

3. INTRODUCTION TO COMPUTATIONAL METHODS IN PROTEIN MODELING AND INTERACTIONS

Your computational/molecular modeling chapter here followed by references

(See printed templates in the lab)

CHAPTER 4

QSAR study on diketo acid and carboxamide derivatives as potent HIV-1 integrase inhibitor

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Abstract:

Herein, we present a validated predictive QSAR model to provide more insight into the relationship between the molecular properties of diketo acid and carboxamide derivatives and the HIV-I integrase inhibition. A set of 40 diketo acid and carboxamide derivatives possessing integrase inhibitory activity were subjected to 2D-QSAR using Discovery studio V3.5. The QSAR results presented herein were based on genetic function algorithm (GFA) approach. Logarithmic inverse values of IC_{50} were taken as dependent variable and physicochemical parameters were taken as independent variable. A suitable set of molecular descriptors was calculated using GFA (max 500 generations). Results showed that radius of gyration, zagreb index, weiner index and minimized energy are statistically significant with the correlation coefficient value of 0.8209 and play important role in HIV-1 integrase inhibition.

Key Words: Diketo acid, carboxamide, 2D-QSAR, GFA, Integrase Inhibitor

This is the last chapter of your thesis and also very crucial - follow this example below. You must use the PAST TENSE in section 1.6 of this chapter. However in section 6.2 you should use the future tense - Some template below here:

CHAPTER 6

6. GENERAL CONCLUSIONS AND RECOMMENDATIONS FOR FUTURE STUDIES

6.1. General Conclusions

The aim of this study was to investigate the effectiveness of nine FDA-approved anti-HIV drugs against the South African HIV PR variant as well as to provide insight into the molecular origin of drug resistance against clinically reported enzyme mutations, V821A and V82F/I84V. To a great extent, this work has accomplished the aims of the study. Results from this work confirmed the following conclusions:

1- Some members of first-generation drugs (such as RTV and APV) and second-generation drugs (such as LPV, DRV and ATV) showed better binding affinities towards C-SA HIV PR in comparison to the other inhibitors.

2- Most inhibitors exhibited slightly lower binding affinities towards subtype C relative to subtype B. This phenomenon is most likely due to the indirect influence of eight polymorphisms present in subtype C that appears to affect flap motion and increase the flexibility of C-SA PR.

3- The amino acid polymorphisms found in the proteases from the C HIV-1 subtype did not appear to be sufficient to induce drug resistance.

4- Drug-resistant mutations, especially V82F/I84V, have contributed to the weakened affinity of the PR towards the inhibitors by disturbing the overall binding landscape of the PR enzyme.

5- As evident from the consistency between the computational results and experimental data, the computational approaches adopted in this work proved to be efficient in providing further insight into drug-protein interactions and drug resistance mechanisms.

Start with the challenges you faced or uncompleted work and ways to improve the study either by using more advanced techniques, missing parts that would be done in the future ,… etc

6.2. Recommendation and Future Studies

One of the major challenges that we faced in this work was the lack of the X-ray crystal structures of the South African subtype C HIV PR (C-SA HIV PR). Another challenge was the lack of sufficient experimental data on the free binding energies for all FDA-approved drugs against C-SA HIV PR that we can compare our computational results against. Although these were challenges but these were the motivations of our work. Further experimental work such as determination of the X-ray crystal structure for ligand-C-SA HIV PR complexes would be a solid starting point for further computational investigations. Also determination of experimental binding energies for all FDA-approved HIV PR inhibitors would be a precise reference to further theoretical studies.

However, our results clearly contributed towards the molecular understanding of drug-enzyme interactions and resistance, further sophisticated computational approaches would be highly recommended to provide additional dimensions towards HIV drug resistance mystery. Recently developed computational approaches such as Principle Component Analysis (PCA) (ref), Residue Interaction Network (RIN) (ref), coarse grained molecular dynamics (ref) and Substrate Envelope Analysis (SEA) (ref) would certainly elaborate on the enzyme dynamics and conformational preferences as well as the drug-enzyme interaction networks. Comparative analysis using these approaches would also provide deeper insight into the mechanisms by which different mutations can develop drug resistance.

References

1.
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You need to put references for new methods/approaches that you recommend in your future work