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###### Study title: A series of N-of-1 trials to assess therapeutic interchangeability of two Enalapril formulations in the treatment of hypertension, Addis Ababa, Ethiopia

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# Synopsis of protocol

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| **Objective(s)** | This N-of-1 clinical study is designed to determine the therapeutic interchangeability of two Enalapril tablet formulations, Envas (Cadila pharmaceutical, Ethiopia) and Ena-Denk (Denk Pharma, German) in individual patients  |
| **Methodology** | Study design: Randomised, partially blinded, two-treatment, two-period, three-cycle crossover trial in a single patient. |
| **Subjects** | Number: 30 individual patients with controlled hypertension |
| **Inclusion criteria** | Male and female patients with primary hypertension controlled on Enalapril/Enalapril containing regimen- those who have achieved a blood pressure target of 140/90 mmHg or less in at least the last 2 months (clinic readings) |
| Subjects who are between 18 – 80 years of age |
| Serum electrolytes and creatinine within the normal range (or the clinical investigator considers the deviation to be irrelevant for the purpose of the study) |
| Normal ECG or stable abnormalities which the clinical investigator does not consider a disqualification for participation in the study |
| Willingness to undergo a pre-study physical examination and laboratory investigations  |
| Ability to comprehend and willingness to sign statement of informed consent |
| Women of child bearing potential having effective contraception in place. If this is oral contraception, then they should have been on it at least 2 months. |
| **Primary outcome** | Mean home Systolic BP difference among individual patients  |
| **Criteria to establish therapeutic equivalence** | Therapeutical equivalence will be defined using Minimal Clinical Important Difference (MCID). An average SBP difference of 5 mm Hg is accepted as MCID and equivalence will be established if the following two criteria are fulfilled; 1. Absence of clinically important difference - when the predetermined MCID is greater than the upper limit of the 95% CI of difference of the two treatments.
2. Equivalence should be evident in at least in two cycles
 |
| **Secondary outcomes** | 1. Change in mean home DBP for both evening and morning diastolic BP values
2. Change in mean clinic DBP for both evening and morning diastolic BP values
3. Change in mean clinic DBP for both evening and morning diastolic BP values
4. Change in mean home DBP measured in the morning, 24 hours after drug intake.
5. Change in mean home DBP measured in the evening, 12 hours after drug intake.
6. Change in mean home SBP measured in the evening, 12 hours after drug intake.
7. Change in mean home SBP measured in the morning, 24 hours after drug intake.
8. Study feasibility outcome and
9. Safety outcome (number and severity)
 |
| **Statistical analysis** | Change in average mean BP is calculated for each period95% CI for the difference of two means Standard statistical procedures will conducted using the R-package |

**INTRODUCTION**

* 1. **Background**

The burden of major non-communicable diseases in Ethiopia is increasing. In 2011, the World Health Organization (WHO) estimated that 34% of the Ethiopian population will die from non-communicable diseases, with a national cardiovascular disease prevalence of 15%, cancer and chronic obstructive pulmonary disease prevalence of 4% each, and diabetes mellitus prevalence of 2%[1](#_ENREF_1" \o "Alwan, 2011 #106). In particular, hypertension is a major public health challenge because of its high frequency and associated risks. Ethiopian studies have found a significantly high prevalence of hypertension at the work place (27.3%)[2](#_ENREF_2" \o "Angaw, 2015 #107) and in an urban community (30%)[3](#_ENREF_3" \o "Tesfaye, 2009 #108). In Ethiopia, hypertension is becoming one of the biggest risk factors for death. In 2008, the Federal Ministry of Health (FMOH) identified hypertension as the seventh leading cause of mortality[4](#_ENREF_4" \o "Ethiopian, 2008 #98). This makes it the single most important cause of mortality among non-communicable diseases.

Drug therapy for hypertension involves the use of a series of drug classes and often requires taking multiple drugs, which makes the treatment expensive in economic terms. With the purpose of providing better access to affordable drugs, the Ethiopian government legalized production and commercialization of preparations with the pharmacological name of the drug with the same dose, presentation and administration route as the reference drug. Because of their lower cost, use of generic drugs is supported by healthcare systems, which recommend physicians to prescribe them, rather than brand-name drugs[5-7](#_ENREF_5" \o "Håkonsen, 2009 #109). Therefore, treatment of hypertension with generics is an ideal option to reduce health care costs in Ethiopia.

In most places in the world, an application for marketing approval of a new generic product must reference a corresponding product, which was approved on the basis of clinical trials to support claims of safety and efficacy. This means generics must show bioequivalence (BE) to a reference product and this is accepted by the European Union (EU)[8](#_ENREF_8" \o "Fontaine, 2001 #112), the United States of America (USA)[9](#_ENREF_9" \o "Chen, 2001 #113) and WHO[10](#_ENREF_10" \o "Organization, 1999 #114). However, whether locally produced drugs are therapeutically equivalent and thus interchangeable with their earlier innovative products is highly questionable in Ethiopia.Though there are strong movements to have one, there is no accredited bioequivalence centre in Ethiopia. Therefore, currently, the Ethiopian medicine regulation authorities do not require bioequivalence reports for locally manufactured drugs.

Human resources and regulatory and equipment capacity sufficient to monitor the quality of locally produced and imported drugs may not always be available in resource-poor settings such as Ethiopia. A regulatory system assessment of sub-Saharan African countries (including Ethiopia) by WHO concluded that these countries did not have the capacity to control the quality, safety and efficacy of the medicines circulating in their markets or passing through their territories[11](#_ENREF_11" \o "Organization, 2010 #115).One practical example for this is that the Ethiopian medicine authority posted that production of one locally manufactured drug was banned upon receiving claims of ineffectiveness of the drug from various health professionals[12](#_ENREF_12" \o "FMHACA, 2010 #79).

Several studies have reported high levels of negative perceptions regarding generic drugs among health professionals and patients in Ethiopia[13](#_ENREF_13),[14](#_ENREF_14). Proving the interchangeability of local generics with brand name drugs will maximize patient benefit and enhance trust of locally manufactured drugs. Therefore, testing the feasibility of a clinical care tool to assess the clinical equivalence of local generics with the reference product is imperative.

**N-of-1 trials**

N-of-1 tests are indicated whenever there is substantial uncertainty regarding the comparative effectiveness of different treatments being considered for an individual patient. Guidelines commissioned by the US Department of Health and Human Services documented the pragmatic use of N-of-1 tests as a means of formally assessing the bioequivalence of generic drugs[15](#_ENREF_15" \o "Kravitz, 2014 #118). A recently published book on N-of-1 trials also reported that N-of-1 trials can be used to compare different brands of the same medicine[16](#_ENREF_16" \o "Nikles, 2015 #119). Researchers have used N-of-1 tests to prove therapeutic interchangeability of generic warfarin[17](#_ENREF_17" \o "Pereira, 2005 #120) and nifedipine[18](#_ENREF_18" \o "Pollak, 2010 #121). A feasibility N-of-1 trial also concluded that follow up of hypertension using home blood pressure measurement is possible[19](#_ENREF_19).

Ethiopian hypertension treatment guidelines recommend an [angiotensin-converting-enzyme](https://en.wikipedia.org/wiki/ACE_inhibitor) (ACE) inhibitor as one of the first line treatment options in the routine management of patients with hypertension, either as first line monotherapy or as a suitable combination treatment for all other types of antihypertensive drugs[20](#_ENREF_20" \o "FMHACA, 2010 #123).

N-of-1 trials are suitable in the following condtions: stable or chronic conditions; treatments that have a rapid onset and offset of effect (short half-life) resulting in minimal washout periods (i.e. time needed for one treatment to wear off and the next to initiate and stabilize); and when there are validated measures for treatment effects[21](#_ENREF_21),[22](#_ENREF_22). Enalapril meets all the conditions of a treatment amenable to N-of-1 trials[19](#_ENREF_19)

**Justification, research question and study objective**

Enalapril is a commonly prescribed drug for the treatment of hypertension in Ethiopia. CADILA Pharmaceuticals Ltd (Ethiopia) produce Enalapril (Envas) in Ethiopia. However, it lacks bioequivalence data. The **question** is therefore “does locally produced Enalapril used in the management of individual patients with hypertension work as well as the counterpart brand products?” Therapeutic equivalence tests (N-of-1 tests) may prove interchangeability, thus adressing the negative perception of generics held by patients and health professionals, and being able to provide assurance that cheaper local drugs can be taken confidently. Enalapril has been selected for this N-of-1 feasibility study to assess intechangeability of generic drugs in the treatment of hypertension. ACE inhibitors are one of the first line drugs for the management of hypertension in Ethiopia [20], and currently (according to physicians‘ views), many patients in ALERT Hospital have been taking this drug.

Enalapril is an angiotensin-converting-enzyme (ACE) inhibitor used in the treat hypertension, symptomatic heart failure, and asymptomatic left ventricular dysfunction[23](#_ENREF_23). ACE converts the peptide hormone angiotensin I to angiotensin II. One of the actions of angiotensin II is the vasoconstriction of blood vessels, resulting in an increase in blood pressure. After absorption, effective inhibition occurs in 2-4 hours after oral administration. Onset of antihypertensive activity is at 1 hour, with peak reduction of blood pressure achieved by 4-6 hours after administration. The extent of absorption of enalapril from oral enalapril tablet is approximately 60%. The effective half-life for accumulation of enalparil following multiple doses of enalapril is 11 hours. It is metabolized in in the liver and excretion is primarily renal[24](#_ENREF_24). One study demonstrated that the first dose of enalapril was effective and produced effects similar to those measured after 7 days and 1 month of treatment[25](#_ENREF_25).

Apart from improving the quality of clinical care for individual patients, conducting the pilot study is crucial, in that in the future running N-of-1 tests on every patient is not practical for many reasons. Therefore, the result of this validation study will be used to inform the design and the implementation of a further study - aggregated N-of-1 trials. Compared to parallel RCTs, aggregated N-of-1 trials require a smaller sample size to produce generalizable data[22](#_ENREF_22" \o "Nikles, 2011 #125), [26](#_ENREF_26). Therefore, as a proxy measure to a bioequivalence study, data from a modest number of patients who are involved in N-of-1 tests (aggregated ones) could be used to determine overall therapeutic equivalence in the population in the future.

The **aim** of this pilot study is to assess the feasibility of N-of-1 tests to fill the knowledge gap on *bioequivalence* through generating objective evidence on *therapeutic equivalence* in individual patients.

The **specific objective** of the project is to compare the therapeutic equivalence of locally produced Enalapril- Envas (CADILA Pharmaceuticals, Ethiopia) with Ena-Denk (Denk Pharma, Germany) by conducting a series of N-of-1 trials.

**Bioequivalence and therapeutic equivalence trials**

Before the 1970s, efficacy and safety of generic drugs was established using clinical trials[27](#_ENREF_27). In the late 1970s, because running a trial was a very expensive, complex and time consuming process and because of the discovery of a reliable bioequivalence test technology, the concept and practice of assessing equivalence has changed. Currently, therapeutic equivalence trials are often conducted to identify drugs of comparable efficacy but with less side effects and/or less cost.

**Comparison of bioequivalence and therapeutic equivalence trials**

|  |  |  |
| --- | --- | --- |
| **Variable** | **Bioequivalence** | **Therapeutic equivalence trials** |
| **Endpoints/target parameter** | Bioequivalence studies do not have a clinical Endpoint. Concentration-time profiles are used as an adequate surrogate for proof of similar therapeutic efficacy and safety.  | Instead of a unique outcome variable (concentration level), often there are a variety of clinical endpoints as target parameters, eg BP change, time to sleep, change in quality of life |
| **Choice of comparator drug** | Contain the same pharmacologically active substance as the drug under test and its selection is based on the existence of a full clinical file in which its efficacy and safety have been documented. | The aim is to ensure that equivalence is established with a reference standard medicine with proven efficacy and safety.If drugs which contain the same pharmacologically active substance are compared, like the bioequivalence studies, use of innovator drugs as a comparator is an ideal option  |
| **Acceptance limit/range** | Commonly, acceptance limit of +/- 80% from the reference drug | A tolerable distance (D) between a test drug and the comparator is defined as the acceptance range. Acceptance limit of 0.90 - as opposed to 0.80 in bioequivalence assessment - can be used. Also, a minimal clinically important difference (MCID) for the management of the condition can be defined at the beginning of the trial  |

Our method of comparator drug selection needs to be a compromise. Because the innovator drug is not marketed in Ethiopia, the choices of comparator drug for this study need to be other criteria. Many physicians believe that drugs imported from the Western countries have a superior quality than those from the Eastern. During market analysis, one such drug which is imported from a country with strong medicine regulation is Ena-Denk (Pharma-Denk, German).

1. METHODS
	1. **Trial design**

This is a therapeutic eqiuvalence study using N-of-1 trials or tests. Each N-of-1 test will be a randomized, partially-blind, multiple crossover study designed to simulate the routine clinical practice of switching a patient between generic and brand forms of Enalapril. We believe this study constitutes a trial of a clinical decision making method. ***It is NOT a trial testing the efficacy of Enalapril.***

Each N-of-1 trial will consist of three successive 14-day treatment pairs, each pair comprising 7 days of Envas and 7 days of Ena-Denk taken once daily in the morning. BP measurements taken in the first two days of each period will be discarded during analysis stage to allow for washout. The order of treatments in each period will be determined by block randomization. BP will be assessed during and at the end of each period. A period will be a 7-day interval (six periods for each patient, during which either Envas or Ena-Denk) will be taken; a day will be the 24-hour interval during which BP will be measured morning and evening. See Fig. 1 below). Description about the number of cycles and duration is given in section 2.11.2.

**Cycle 3**

**Cycle 2**

**Cycle 1**

Randomization Ena-Denk   Envas

EnaDenk

 for 7d

Envas for 7d

Envas for 7d

Ena-Denk for 7d

Envas for 7d

Ena-Denk for 7d

**Figure 1.** Diagram of N-of-1 test for treatment of hypertension using two Enalapril formulations

* 1. **Study setting**

This is a single site institute-based study that will be conducted in AHRI/ALERT centre, one of the government owned institutes located in the capital city of Ethiopia (Addis Ababa). The complex comprises ALERT specialized hospital, and the Armauer Hanson Research Institute (AHRI). In collaboration, the hospital and the research institute have conducted a number of clinical trials. AHRI is one of the leading biomedical and clinical trial centres in Ethiopia. The hospital facilities also provide an excellent opportunity for clinical research. ALERT specialized hospital provides both inpatient and outpatient services. It also provides chronic care services for patients with chronic conditions including hypertension. Therefore, AHRI/ALERT provides a good venue for piloting N-of-1 studies in Ethiopia.

The complex is also the home of a local ethics review committee (ERC), AHRI/ALERT Ethics Review Committee (AAERC). AAERC is a vibrant ERC with good attributes in aspects such as composition and experience.

**Inclusion and exclusion criteria for participants**

A total of 30 subjects, who fulfil the inclusion criteria, do not meet any of the exclusion criteria, and who have given written informed consent, will be enrolled into the study.

**Inclusion criteria:** Subjects fulfilling the following criteria are eligible for participation in the study:

* Male and female patients with primary hypertension controlled on Enalapril/Enalapril containing regimen- those who have achieved a blood pressure target of 140/90 mmHg or less in at least the last 2 months (clinic readings)
* Subjects who are between 18 – 80 years of age
* Serum electrolyte and creatinine within the normal range (or the clinical investigator considers the deviation to be irrelevant for the purpose of the study)
* Normal ECG or stable abnormalities which the clinical investigator does not consider a disqualification for participation in the study
* Willingness to undergo a pre-study physical examination and laboratory investigations
* Ability to comprehend and willingness to sign statement of informed consent
* Women of child bearing potential having effective contraception in place. If this is oral contraception, then they should have been on it at least 2 months.

**Exclusion criteria:**

* Any evidence of clinically significant, poorly controlled hematologic, renal, hepatic, or gastrointestinal problems (current thrombocytopenia (platelet count < 100 ×109/L or hemoglobin < 10 g/dL); liver function test abnormalities (alanine aminotransferase or aspartate aminotransferase ≥ 2 × upper limit of normal [ULN]) or severe renal dysfunction (creatinine clearance < 25 mL/min)
* Any evidence within the last six months of clinically significant diseases involving the cerebrovascular, autoimmune, or cardiovascular systems, including poorly controlled angina pectoris, secondary hypertension, congestive heart failure, or myocardial infarction or stroke.
* Concomitant use of major psychotropic agents or antidepressant drugs or regular use of nonsteroidal anti-inflammatory agents, high-dose aspirin, or any agent that could raise or lower blood pressure within the last 2 months.
* A history of drug or alcohol abuse.
* sensitivity to angiotensin-converting enzyme (ACE) inhibitors,
* Unwillingness to follow the study protocol or study-related procedures (e.g. swallow tablets).
* Clinically significant abnormal ECG findings or vital signs during screening.
* Clinically significant illness or surgery within 4 weeks prior to study.
* Pregnant (positive pregnancy test) or breast-feeding women or women who are planning to be pregnant during the trial period.

## Interventions

* + 1. **Investigational products**

This N-of-1 therapeutic equivalence study will compare two formulations of Enalapril (Envas and Ena-Denk). See Table 1.

## Table1: Identity of investigational products

|  |  |  |
| --- | --- | --- |
| **Description** | **Test product** | **Reference product** |
| **Name** | Envas | Ena-Denk |
| **Drug substance** | Enalapril | Enalapril |
| **Administration**  | Peroral | Peroral |
| **Formulation** | Tablet | Tablet  |
| **Dose** | 5-20mg | 5-20mg |
| **Manufactured by** | **Cadila** Pharmaceuticals Ltd**, Ethiopia** | Denk Pharma, German |

##

## Trial drug management

For each patient, individualized study medication will be prescribed for the duration of the study (six weeks). Drugs will be bought from a wholesale pharmacy. The investigator will supply study medications to study pharmacist. Study medications will be packed and labelled by study pharmacist in six bottles containing one week’s supply of medicines each, and randomly ordered. The study medication will be stored at AHRI pharmacy store. Authorised persons will dispense medications for administration.

All medications are accounted for by the principal investigator or his delegate, and any discrepancies are explained and documented. This accounting of study medication consists of a dispensing record, including the identification of the person to whom the drug is dispensed (enrolment and randomization code), dose, quantity and the date of dispensing. Any unused drug will be returned to the investigator. This record is, in addition to any drug accountability, recorded in the electronic data capture system. The investigator will destroy the unused medication in accordance with the local regulation.

## Storage conditions

The investigational products will be stored at the clinical trial site under the close supervision of the principal investigator. The investigational products will be stored in a dry cool place at a temperature not exceeding 25o C and protected from light, according to the storage conditions of the products.

## Packaging, labelling and dispensing

The investigational medicinal products for each individual will be packed according to the randomization schedule for each period, at the clinical trial site. Packaging (including labeling) will be performed in accordance with Good Manufacturing Practice. Packaging and labeling of the products for the subjects will be documented in detail, including study ID and randomization code. This documentation will include all precautions taken to avoid and identify potential dosing mistakes. Each individual’s (weekly supply of investigational products will be packed in separate labelled containers indicating the study code, subject enrolment number, treatment period, name of the product, dose, contents (number of tablets), route of administration, name of the Principal Investigator, labelled “For Clinical Research Only”, batch number, expiry date, and storage conditions,. Individual subjects will be supplied their medication weekly according to the randomisation code. Seven tablets of either the test product or the reference product will be dispensed to individual subjects. Weekly packs of Enalapril tablets will be dispensed by the pharmacies and collected by individual subjects. The study site physician or nurse will verify that the tablets are actually dispensed according to the prescribed dose to all subjects.

## Selection of doses in the study

Participants in this study are those whose blood pressure is controlled with Enalapril. Therefore, participants will continue on the dose that they are currently taking for the duration of the trial. The dosage is within the recommended therapeutic dosage. Physicians have suggested that most patients who have active follow up in the ALERT hospital are currently on a daily dose of 10mg Enalpril. Other doses (5-20mg) will be considered depending on the study physician and patient decision.

## Mode of administration

Investigational medicines in this study will be taken every morning using per oral route. The tablets are to be swallowed whole unchewed with 250 ml of water with or without food.

* 1. **Concomitant medications**

No new medications that could affect blood pressure will be introduced for the duration of this study. Subjects should not take any traditional medicine for at least 2 weeks prior to commencing the study, and throughout the conduct of the study. During the study, subjects are advised not to take any other anti-hypertensive medications other than medications(s) they were on at the start of the study. If concomitant medication is unavoidable in case of emergency, the use must be reported in the patient’s diary (comment log) (dose and time of administration) and possible effects on the study outcome must be addressed (see also sect 2.7). Patients will be permitted to take short courses of drugs that do not affect blood pressure control, such as antibiotics for intercurrent infections. All other medicines the patient takes will remain constant for the duration of the equivalence test.

* 1. **Education and instruction for subjects**

Subjects will be trained about the basic methods of BP self-measurement, the meaning of BP values, and the monitoring device to be used by the principal investigator and study physician. During follow up, subjects will be instructed to measure their BP three consecutive times in the sitting position in the morning before breakfast and in the evening after dinner. They will also be asked to visit the study site for follow up on a weekly basis. They will be asked to abstain from taking any medication (prescription and nonprescription drugs) without notifying the study staff. Patients who smoke and take alcohol will be asked to minimize their consumption. They will be asked to take the respective study medication in the sitting position together with 250 ml water at the same time each day. They will also be asked to bring pill boxes and all study forms to their weekly visits.

* 1. **Treatment compliance**

To avoid treatment non-compliance, every week, there will be a drug adherence counselling session by the study nurse before drug dispensing. Moreover, every week, the study nurse will check pill boxes and ask questions if there are missed medications.

* 1. **Study withdrawal or interruption**

If a concomitant medication has to be taken that might influence blood pressure that person will cease the trial until the new medicine has ceased or been stable for at least two weeks. If they wish, the participant can restart the study, substituting a new randomly packed cycle of medicines for the interrupted cycle. Unused medicines from the trial will be discarded. If the person has to continue on the new medicine, the trial will cease, and the data collected will be analysed, if at least one cycle has been completed.

Participants will also be withdrawn from the study under the following conditions:

* Patient request
* If, in the opinion of the treating clinician, the patient's interests are best served by withdrawing from the trial
* Patient non-compliance
* Development of an exclusion criterion.
	1. **Outcome measures**

The study physician will assess the patient before and after each treatment period (each week) and collect data.. Individual participants will be asked to record their blood pressure two times (morning and evening) daily using BP recording chart.

Below are the primary and secondary outcome measures.

**Primary Outcome:**

Therapeutic equivalence will be assessed by calculating change in mean seated SBP. The minimal clinically important difference (MCID) is the smallest treatment effect that would lead to a change in a patient's management[28](#_ENREF_28). Based on the literature[29](#_ENREF_29), an average SBP difference of 5 mm Hg is accepted as MCID. A difference of <5 mm Hg SBP between the two treatments is considered to be of no clinical importance. Clinically important differences can be defined using a predefined MCID and confidence interval of the difference of the two treatments[30](#_ENREF_30" \o "Man‐Son‐Hing, 2002 #133). (see figure 2)

**Clinical importance**

1. Definite
2. Probable
3. Possible
4. Definitely not

 minimal clinically important difference

* Point estimate and 95% confidence interval

**Figure 2**: Relationship between clinical importance and statistical significance

**Determination of clinical importance of results**

Clinical importance can take 4 forms, depending on the relationship of the MCID of the intervention to the point estimate (the best single value of the efficacy of the intervention that has been derived from the study results) and the 95% CI surrounding it[30](#_ENREF_30):

1. Definite – when the MCID is smaller than the lower limit of the 95% CI;
2. Probable – when the MCID is greater than the lower limit of the 95% CI, but smaller than the point estimate of the efficacy of the intervention;
3. Possible – when the MCID is less than the upper limit of the 95% CI, but greater than the point estimate of the efficacy of the intervention; and
4. Definitely not – when the MCID is greater than the upper limit of the 95% CI.

**Criteria to establish therapeutic equivalence:**

1. Absence of clinically important difference- for therapies to be considered equivalent, not only should the comparison of the efficacies of the two interventions not reach statistical significance, but also the upper limit of the confidence interval should be smaller than the predetermined MCID
2. Equivalence should be evident in at least in two cycles out of three.

**Secondary Outcomes:**

1. Change in mean home DBP for both evening and morning diastolic BP values
2. Change in mean clinic DBP
3. Change in mean clinic SBP
4. Change in mean home DBP measured in the morning, 24 hours after drug intake.
5. Change in mean home DBP measured in the evening, 12 hours after drug intake.
6. Change in mean home SBP measured in the evening, 12 hours after drug intake.
7. Change in mean home SBP measured in the morning, 24 hours after drug intake.
8. **Feasibility outcome**: Based on literature[31](#_ENREF_31), the following criteria will be used to measure success/acceptability of the pilot study:

 (a) Recruitment rate: at least 70% of all eligible patients can be recruited ,

 (b) Completion rate: at least 80% of all recruited subjects complete the study,

 (c) At least 90% of patients took every scheduled dose of the study drug, and

 (d) More than 80% of requested measurements obtained and valid. Measurements are considered invalid when either or both of systolic and diastolic BP readings are not compatible (less than 20 mm Hg difference).

(e) Patients and phycisians view on the trial

1. **Safety outcomes:** Adverse events (number, severity) identified by patients recording. AEs recorded by physician on AE CRF.

**Participant time line**

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| ***Data Collection Form***  | ***Screening phase: Day -1*** | ***Screening phase: Day -0*** | ***Trial phase: Day 1-7*** | ***Trial phase: Day 8-14*** | ***Trial phase: Day 15-21*** | ***Trial phase: Day 22-28*** | ***Trial phase: Day 29-35*** | ***Trial phase: Day 36-42*** | ***Trial phase: Day 8*** | ***Trial phase: Day 15*** | ***Trial phase: Day 22*** | ***Trial phase: Day 29*** | ***Trial phase: Day 36*** | ***Trial phase: Day 43*** |
| Form 1-Screening and Registration Form | X |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Form 2-Final Eligibility Assessment Form |  | X |   |  |  |  |  |  |  |  |  |  |  |  |
| Form 3-Home BP and Side Effect Recording Forms |  |  |   X  |  X  |  X  |  X  |  X  |  X  |  |  |  |  |  |  |
| Form 4-Trial Phase Clinic Assessment Form |  |  |  |  |  |  |  |  |  X  |  X  |  X  |  X  |   X  |   X  |
| Form 5- dverse Events Form |  |  | X | X | X | X | X | X | X | X | X | X | X | X |
| Form 6- Comment and other Unscheduled Visit Forms |  |  | X  | X | X | X | X | X |  |  |  |  |  |  |

* 1. **Sample size estimation**

This is an individual N-of-1 trial to assess feasibility of N-of-1 tests to prove therapeutic equivalence of Enalapril (Envas, Ethiopia). An audit of sample sizes for pilot and feasibility trials undertaken in the United Kingdom reported that the median sample size for pilot trials were 30 participants per arm[32](#_ENREF_32). We assume that a sample size of 30 patients is adequate to assess feasibility.

* 1. **Participant recruitment**

ALERT hospital provides chronic care through its chronic care outpatient department. Subjects will be recruited among hypertensive patients who have follow up in the hospital. The principal investigator together with study physicians and nurses will recruit participants.

* 1. **Assignment of interventions**
		1. **Randomization**

We will use block randomization and with a block size of 2. The order in which patients receive drugs will be randomized by computer for each single case, such as BA-AB-BA or AB-BA-BA. This will be organised by study statistician.

* + 1. **Selection of number of treatment cycles and duration of period**

This N-of-1 trial consists of three cycles, each consisting of two treatment periods of seven days each, total length 6 weeks. To account for washout from the previous period, the first two days of BP readings in each treatment period will be discarded. Selection of five days period of usable data in our study was based on the study conducted by [Gilles Chatellier](http://hyper.ahajournals.org/search?author1=Gilles+Chatellier&sortspec=date&submit=Submit) et al[19](#_ENREF_19" \o "Chatellier, 1995 #136). One of that study‘s aims was to determine whether N-of-1 trials of two cycles, 10-day treatment pairs each, using BP self-measurement at home is methodologically appropriate. The study reported a period of 5 consecutive days of BP measurement is sufficient to accurately detect a drug-induced fall in BP in a single patient, provided that there are at least 30 readings in each 5-day trial period. However, the study did not consider washout period and the individual agreement between the two cycles was only moderate at best. Therefore, this study recommended a trial of longer periods with three cycles. By asking the participant to measure BP every day, we expect compliance will be improved compared to asking them to not record BPs for two days per week.

* + 1. **Blinding**

Masking the identity of the medicines (blinding) is ideal. We have evaluated three ways of blinding to determine which option is most suitable in this study.

**Encapsulation**: This is a relatively simple, effective and commonly used method to make trial drugs appear and taste identical. However, compounding pharmacies that can prepare medications in matching capsules are lacking in Ethiopia. We also could not find an encapsulation machine in the local pharmaceutical companies for this purpose. Beyond safety issues, manual encapsulation could raise conflict of interest with the local pharmaceutical company that produces Enalapril.

**Double dummy design**: Preparation of identical placebos for both Enalapril formulations and study drugs administered by a double dummy design is considered as a second choice. However, Ethiopian pharmaceutical companies are not experienced in placebo preparation at present.

**Partial blinding**: Patients will not be blinded; however, trial staff and the outcome assessor will be blinded. Following enrolment of patients for the individual N-of-1 test, doctors will prescribe both formulations of Enalapril. Both prescriptions together with the randomized medication order will be given to a pharmacist who is independent of the trial. The pharmacist will then dispense the appropriate treatment to the patient following the randomized medication order.

The third option, partial blinding, is the most practical in the situation. Therefore, this N-of-1 study is planned to be a partially-blinded trial. Therefore, when patients swallow the drugs, they may identify and appreciate the physical differences between the two drugs. However, blinding/unblinding of patients is a compromise between feasibility and minimal potential effects on reported outcomes. A review of relevant literature found thatthree N-of-1 papers, including one protocol publication, on hypertension were not fully blinded[18](#_ENREF_18),[19](#_ENREF_19),[33](#_ENREF_33) . Keeping the design less complex would make the trial resemble the usual way of switching drugs except that it is pre-planned, randomised and closely monitored[33](#_ENREF_33" \o "Samuel, 2016 #138).

* 1. **Data collection, management, and analysis**

### Data collection

### Establishing controlled BP

In this study, those with an average clinic BP measurement controlled at 140/90 mmHg or less will be included. Only patients with hypertension controlled over the last one year will be included. This is because there is a considerable placebo controlled rate for hypertension, which increases after 1 year[34](#_ENREF_34). Hypertensive patients who have follow-up in ALERT hospital usually have a monthly visit schedule. To account for BP variations including seasonal ones, BP will be established by taking the average of three consecutive BP measurements taken in at least the last two months.

### Using home BP measurement

BP self-measurement at home has been shown to be a sensitive tool, ie, able to detect small changes, frequently resulting in reduced variability and greater reproducibility of measurements compared to office BP readings[35](#_ENREF_35),[36](#_ENREF_36). Measurement precision has an important role in quantification of the hypotensive effect and consequently on the physician’s treatment decisions. Home blood pressure measurement also eliminates the white-coat effect[37](#_ENREF_37). Multiple cross-sectional studies have reported that target organ damage is more strongly correlated with home BP measurements than with clinic BP measurements[38](#_ENREF_38),[39](#_ENREF_39). Several reports have pointed out that increasing the number of BP values by continuous monitoring allows reduction in the number of subjects necessary for meaningful results in drug trials[40](#_ENREF_40),[41](#_ENREF_41) The implication of these findings is that increasing the number of observations in 24 hour reduces the number of cycles and duration of each period required in N-of-1 trials.

The average systolic BP at home over five days is used to assess the effectiveness of treatment. The repeat measurement of BP over 5 days helps to narrow the variability of BP around the true mean BP value[42](#_ENREF_42),and generally provides the highest prognostic and diagnostic capacity in comparison with shorter monitoring periods[43](#_ENREF_43),[44](#_ENREF_44). Individual participants will be trained and asked to record their blood pressure two times (morning and evening) daily using an ambulatory blood pressure monitor. Morning measurements should taken before medication and breakfast. The measurements will be taken in a quiet room after five minutes of seated rest, with three readings taken one minute apart. BP should be recorded immediately in a study diary (Table 2).

**Table 3: Home BP recording chart (1 week example)**

|  |  |
| --- | --- |
| Intials:Subject No:  |  Start Date:   |
|   | **Day 1** | **Day 2** | **Day 3** | **Day 4** | **Day 6** | **Day 6** | **Day 7** |
| **M\*** |  **E\*** | **M\*** |  **E\*** | **M\*** |  **E\*** | **M\*** |  **E\*** | **M\*** |  **E\*** | **M\*** |  **E\*** | **M\*** |  **E\*** |
| **SBP 1** |   |   |   |   |   |   |   |   |   |   |   |   |   |   |
| **DBP 1** |   |   |   |   |   |   |   |   |   |   |   |   |   |   |
| **SBP 2** |   |   |   |   |   |   |   |   |   |   |   |   |   |   |
| **DBP 2** |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| **SBP 3** |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| **DBP 3** |   |   |   |   |   |   |   |   |   |   |   |   |   |   |
| **M\* Morning**  | E\* Evening |  |  |  |  |  |  |  |  |  |  |  |  |

### BP SELF-MONITORING DEVICE

A validated BP machine will be used for home blood measurement. Home BP measurement protocol is developed using standard methodology[45](#_ENREF_45). Home blood pressure measurement data will be collected at every clinic visit.

### Safety Evaluation

Because this is a study of medication that the patient has been taking, the risk of serious adverse events is minimal. All abnormal or unpleasant effects from medications will be considered as side effects. Individual patients will collect side effects using an open-ended questionnaire (Table 3) at home and bring it during their weekly visits. If patient develop series side effect, the study physician will take appropriate measure. The most common side effects of Enalapril include increased serum creatinine (20%), dizziness (2–8%), low blood pressure (1–7%), syncope (2%), and dry cough (1–2%). The most serious common adverse event is angioedema (swelling) (0.68%) which often affects the face and lips, endangering the patient’s airway.

**Table 3: Side effect questionnaire**

**Do you have any unpleasant side effects from the medication? Please fill in the following table**

|  |  |  |  |
| --- | --- | --- | --- |
| **Date** | **Side effect**  | **How much did this bother you? (put cross mark-X- when applicable)** | **Did this make you want stop the medication?** |
| Extremely | Somewhat | Very little | Not very much | Yes | No |
|  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |

### Trial procedure and data collection

### Data collection during the screening phase

During two days before the start of the study, screening and eligibility assessment will be performed including the following parameters:

* Personal data
* Medical history
* Physical examination (including: height, weight)
* 12-lead ECG
* Vital signs ((including: blood pressure (average of the last three month’s BP measurements), heart rate, temperature))
* Pregnancy test (female subjects)
* Laboratory tests (hgb, potassium, ALT and creatinine)

Data for screening will be collected using ***Form 1***-Screening and Registration Form and F***orm 2***-Final Eligibility Assessment Form.

### Data collection during the study

1. **Data collection at clinic**
2. The following are collected every week at each clinic visits:
* Questioning for subjective well-being
* Personal data (including smoking, exercise)
* Vital signs (including: three consecutive BP measurements taken in 1 minute interval, HR and temperature)
* Questioning for adverse events
* Compliance check (including pill count)

The results will be recorded in the ***Form 4***-Trial Phase Clinic Assessment Form and ***Form 5***-Adverse Events Form.

1. **Data collection at home**

 The following data are collected at home by participants:

* Daily home BP records using home BP recording chart - see Table 2.
* Undesired effect record using side effect questionnaire - see Table 3.

The results will be recorded in the ***Form 3***-Home BP and Side Effect Recording Forms

### Data management

Data that will be generated in this study will be appropriately documented and checked for validity and accuracy. Study data will be managed using an Excel database at The University of Queensland. Data from paper CRFs will be double entered into the database by two persons independently so that data will be matched and checked for validity and accuracy before being endorsed for analysis. As per ICH Good Clinical Practice (GCP) guidelines, the investigators will maintain information in the study subjects’ records which corroborates data collected and entered into the CRFs.

### Case report form

Medifacts International published the CRFs they used during a clinical trial that involved Ambulatory Blood Pressure Monitoring in patients with hypertension[46](#_ENREF_46). The CRFs for this study are adapted from the above study.

### Statistical methods

Analysis will be conducted based on BP measurements taken in the last 5 days of each of the six periods. The first two days’ BP measurements in each period will be discarded. Based on literature[29](#_ENREF_29), 6mmhg is accepted as MCID. Therefore a SBP difference of >5 is considered clinically significant. A mean change in evening SBP of < 5 mm Hg between the two test medicines in at least two treatment cycles will be considered as as therapeutically equivalent.

The following will be calculated for each patient:

Means (±1 SD) for the morning and evening systolic and diastolic BP values for each period.

Change in mean DBP measured in the evening, 12 hours after drug intake.

Change in mean DBP measured in the morning, 24 hours after drug intake.

Change in mean SBP for both evening and morning diastolic BP values

Change in mean SBP measured in the evening, 12 hours after drug intake.

Change in mean SBP measured in the morning, 24 hours after drug intake.

Change in mean DBP for both evening and morning diastolic BP values

Differences in means will be compared using a paired Student’s t test as appropriate. Significance will be set at P ≤ 0.05. Standard statistical procedures will conducted using the R-package.

Possible side effects of the study medication and any serious adverse events will be tabulated for individual patients.

* + 1. **Study result report**

**At** the end of the trial, BP measurements will be analysed and the result will be given to the treating physician. After looking at these results, the patient and the treating physician will be able to decide if the local drug works for treating the hypertension.

**What will be reported?**

Key elements of the report will include:

• Patient details

• Description of the trial – medications compared; order of medication periods; endpoints; date of report

• Conclusion/summary of overall response

• Summary of outcomes used to determine the overall response

• Use of other medications during the trial

• Detailed results of the individual outcome measures, including graphs and/or tables of relevant data points.

# ETHICS

## Independent ethics committee (IEC)

The study protocol and any accompanying materials will be submitted to UQ and AHRI Institutional Review Boards (IRB)/Independent Ethics Committee (IEC) for ethical approval. The accompanying material includes participant information sheets, informed consent form and terms of any compensation given to the participants as well as advertisements for the study. An approval letter (specifying the protocol number and title) from each IEC specifying the date on which the committee met and granted the approval must be obtained before study initiation by the investigator.

The purpose of this N-of-1 test is to systematically assess the therapeutic equivalence of the local Envas so that if equivalent, patients may take the less costly product with confidence. Such assessments, as long as conducted on already approved therapy and for the same indication, should be regarded as a clinical quality assurance project with no more risk than standard clinical care.

The trial will be registered with a clinical trials registry.

* 1. **Subject information and consent**

It is the responsibility of the principal investigator, or a person designated by the investigator, to obtain signed informed consent from each subject prior to participating in this study after adequate explanation of the aims, procedures and possible risks. In the case where both the subject and his/her legally acceptable representative are unable to read, an impartial witness will be present during the entire informed consent discussion. After the subject and representative have orally consented to participation in the trial, the witness’ signature on the form will attest that the information in the consent form was accurately explained and understood. The investigator or designee will also explain that the subjects are completely free to refuse to enter the study or to withdraw from it at any time, for any reason. The treatment offered by their treating physician will not be affected by a decision to withdraw.

## Confidentiality

The investigator will assure that subjects’ anonymity will be maintained and that their identities will be protected from unauthorized parties. The investigator will keep a participant enrollment log showing codes, names and addresses.

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**ACRONYMS/ABBREVIATIONS**

AE- Adverse Event; AAERC- AHRI/ALERT Ethics Review Committee; ALT- Alanine Aminotransferase; ACE- [Angiotensin-Converting-Enzyme](https://en.wikipedia.org/wiki/ACE_inhibitor%22%20%5Co%20%22ACE%20inhibitor); AHRI- Armauer Hanson Research Institute; BE- Bioequivalence; CI- Confidence Interval; CRF- Case Report Form; DBP- Diastolic Blood Pressure; ECG- Electrocardiography; ERC- Ethics review committee; EU- European Union; FMOH- Federal Ministry of Health; Hgb- Haemoglobin; ICH- International Conference on Harmonisation; IRB- Institutional Review Boards; MCID- Minimal Clinical Important Difference; RCTs- Randomized Clinical Trials; SBP- Systolic Blood Pressure; UQ- University of Queensland; USA- United States of America; WHO- World Health Organization

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