


**Xerostomia and
hyposalivation in HIV
positive patients with and
without HAART**



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WESTERN CAPE

DECLARATION

I, Anney Parakulath Cherian, declare that “*Xerostomia and hyposalivation in HIV positive patients with and without HAART*”, is my own work and that all the sources I have quoted have been acknowledged by references. This thesis has not been submitted for any other degree.

Signed: _____

ANNEY P CHERIAN

Date: _____



DEDICATION

To my late father, Oommen Cherian.

You will always be my inspiration.



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I wish to express my gratitude all those who have played a vital role in the completion of my work.

- To God Almighty, who has made this possible.
- Dr A Jeftha, thank you for your supervision of this work. Your continued support and encouragement has been invaluable.
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ABSTRACT

Introduction: Xerostomia and reduced salivary flow have been reported often enough among HIV positive patients. Strong associations have also been established between HIV infection and oral effects of reduced salivary flow like xerostomia, high DMFT, increased candidial infection etc. Besides the direct effect of HIV infection, xerostomia and reduced salivary flow have also been reported as a side effect of Highly Active Anti Retroviral Treatment (HAART). Studies have shown that xerostomia has a negative effect on the quality of life of people living with HIV & AIDS. Although reduced salivary flow is a main cause for xerostomia, complaints of xerostomia is also found in the absence of salivary flow deficiency. An exact correlation between the two is not always found.

Aim: The aim of this study is to compare the prevalence of xerostomia and hyposalivation, in HIV positive patients on HAART, HIV positive patients not on HAART and HIV negative patients, attending Empilweni Gompo community health centre (EGCHC) in East London.

Study Methods: This is a cross sectional analytical study. Xerostomia and resting & chewing-stimulated salivary flow rates were measured for 150 patients who were from three groups: group 1- HIV negative, group 2- HIV positive not on HAART and group 3- HIV positive on HAART for more than two years. Each group had 50 patients. Xerostomia was measured using a questionnaire and salivary flow rates were calculated after saliva collection over a three minute period.

Results: There was significant difference in the prevalences for xerostomia ($p=0.006$) and less than normal chewing-stimulated flow rate ($p=0.041$) among the three groups with the HIV positive group not on HAART showing the greatest deficiency. HAART was not found to have a negative effect on salivary function. A statistical significance was also observed while comparing mean resting ($p=0.010$) and chewing-stimulated ($p=0.034$) salivary flow rates among the three groups. The mean salivary flow rate of those complaining of xerostomia was found to be significantly lower than that of those who did not have xerostomia ($p=0.005$).

Conclusion: HIV positive patients not on HAART are more vulnerable to salivary gland dysfunction. HAART in itself does not to adversely affect xerostomic perceptions or salivary flow rates. The xerostomia questionnaire is a useful tool in indicating those with possible low salivary flow rates.

Key words: HIV, Salivary gland hypofunction, Xerostomia, HAART, Salivary flow rate, Saliva collection.



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LIST OF ABBREVIATIONS

3TC- Lamivudine

AIDS- Acquired Immunodeficiency Syndrome

ART- Antiretroviral Therapy

ARV- Antiretroviral

AZT- Zidovudine

BKV- BK Virus (named after the initials of the renal transplant patient from whom it was first cultured)

BLEC- Benign Lymphoepithelial Cyst

CD4- Cluster of Differentiation Antigen 4

CD8- Cluster of Differentiation Antigen 8

CMV- Cytomegalovirus

DILS- Diffuse Infiltrative Lymphocytosis Syndrome

DMFT- Decayed, Missing and Filled Teeth

EBV- Epstein Barr Virus

EGCHC- Empilweni Gompo Community Health Centre

FDC- Fixed Drug Combination

FNA- Fine Needle Aspiration

HAART- Highly Active Antiretroviral Therapy

HIV- Human Immunodeficiency Virus

HIV-SGD- HIV-associated Salivary Gland Disease

HSRCA- Human Sciences Research Council

II- Integrase Inhibitor

IRIS- Immune Reconstitution Inflammatory Syndrome

NNRTI- Non-Nucleoside Reverse Transcriptase Inhibitor

NRTI- Nucleoside Reverse Transcriptase Inhibitor

PGE- Parotid Gland Enlargement

PI- Protease Inhibitor

QOL- Quality of Life

SLIP- Secretary Leukocyte Protease Inhibitor

SS- Sjogren's Syndrome



CHAPTER - 1

INTRODUCTION

According to the Human Sciences Research Council's (HSRC) National HIV Prevalence, Incidence and Behaviour Survey that was released in April 2014, South Africa ranks first in HIV incidence in the world (Shisana et al., 2014). The number of South Africans infected with HIV now stands at 6.4-million; 12.2% of the population. This is an increase of 1.2 million more than in 2008. The HSRC attributes this increase in prevalence “largely due to the combined effects of new infections and a successfully expanded ART programme. The latter has increased survival among HIV-infected individuals” (Shisana et al., 2014).

Since the early days of its emergence, HIV has been associated with a variety of oral lesions. Klein et al. (1984) were among the first to observe and associate the presence of oral candidial infections to the subsequent development of AIDS. There has been a general decrease in oral manifestations related to HIV in patients on HAART (Greenspan et al., 2004; Schmidt-Westhausen et al., 2000; Masiwa and Naidoo, 2011; Ortega et al., 2009). Oral changes seen in HIV positive patients on HAART were the reduction of candidiasis, hairy leukoplakia and Kaposi's sarcoma. (Nittayananta et al., 2010a; Schmidt-Westhausen et al., 2000; Hamza et al., 2006).

HIV-associated Salivary Gland Disease (HIV-SGD) in general seems to be slowly increasing in prevalence during the post HAART era (Greenspan et al., 2001; Patton et al., 2000). A higher prevalence of 50% and 47% for HIV-SGD has been reported from Africa (Naidoo and Chikte, 2004; Matee et al., 2000). An increase in salivary gland disease was also reported in a cohort with established use of HAART (Patton et al., 2000; Freeman et al., 2012). Reduced salivary flow or xerostomia was also reported with the use of HAART (Freeman et al., 2012; Navazesh et al., 2003; Nittayananta 2010a).

The most common salivary gland changes relate to their function of saliva production and manifest as xerostomia and hyposalivation. Xerostomia is the perceived feeling of dry mouth which may or may not be associated with salivary gland hypofunction. It is subjective and can be measured by means of questionnaires (Sreebny & Valdini, 1988)

or visual analogue scales (Pai et al., 2001). On the other hand, hyposalivation is a demonstrable reduction in salivary flow rate that can be measured objectively by collecting saliva over a specified period of time (Navazesh et al., 1992a). Often these terms have been used interchangeably but studies have demonstrated that a report of xerostomia does not necessarily indicate an actual measurable reduction in salivary flow rate. The reverse is also true, as a few of the patients that did show reduced flow rates did not complain of dry mouth symptoms (Sreebny & Valdini, 1988).

A reduction in salivary flow rates would adversely affect its functions which include lubrication, buffering capacity, tooth remineralisation, antimicrobial and antifungal protection. This would result in an increase in dental caries, certain oral infections and a general oral discomfort. Difficulty in denture retention is often experienced by those with low salivary flow rates. Salivary gland hypofunction also has a high predictive value for recurrent candidial infection (McCarthy et al., 1991). Busato et al. (2013), in their study, concluded that xerostomia reduces the quality of life of people living with HIV & AIDS

With HIV infection being progressively managed successfully by the country's health care services, more and more HIV infected people are living longer and healthier lives while on HAART. Therefore, it is imperative that the general dental practitioner be aware of the salivary gland effects and resultant oral consequences seen in HIV infection and subsequent HAART.

These factors, along with the fact that little data is available from South Africa on the long term effects of HAART on the oral health of this increasing population subset, has been a motivation for this study.

The present study evaluates and compares the prevalence of self reported xerostomia through a questionnaire (Sreebny & Valdini, 1988) and the prevalence of hyposalivation through measuring salivary flow rates as described by Navazesh et al., (1992a, 2003) among 3 sub groups: HIV positive on long term HAART, HIV positive not on HAART and HIV negative individuals.

CHAPTER-2

LITERATURE REVIEW

The basis of knowledge required for this review will begin by defining the normal physiological responses of the salivary glands and the saliva they produce. This will enable a concise evaluation and understanding of the loss of this function and the sequelae thereof. The literature review begins with a summary of studies and review articles that have accrued the knowledge that is currently available. Having understood the physiological characteristics and functions of normal salivary mechanisms, subjective and objective ways to assess its health is also reviewed.

The effect of systemic disease, especially HIV infection, and HAART, on the salivary mechanism will be explored followed by ways to alleviate salivary gland dysfunction to complete a holistic review on this topic.

2.1. SALIVARY GLANDS AND SALIVA

Saliva is an exocrine secretion that is produced by 3 major and numerous minor salivary glands. The paired major salivary glands - parotid, submandibular and sublingual, secrete about 90% of the total volume of saliva (Napeñas et al., 2009). The sublingual and submandibular glands are located in the floor of the mouth. Minor salivary glands are found all over the mouth: palate, lower lip, tongue, cheek and pharynx. While the parotid glands produce mostly serous secretions, the sublingual and the minor salivary glands produce purely mucous secretions and the submandibular produces a mixed sero-mucous secretion (Cassolato & Turnbull, 2003).

Saliva is a slightly acidic, hypotonic complex mix of oral secretions composed of 90% water. Some of the important constituents of saliva include electrolytes, immunoglobulins, nitrogenous products, enzymes and other proteins.

Each of the many components in saliva plays an important function in maintaining the oral homeostasis. These constituents impose an arsenal of protective functions that are depicted in greater detail in Table-2.1.

Table 2.1- Functions and components of saliva

Functions	Component in saliva
Protection of teeth and oral, pharyngeal and oesophageal mucosa	
Mechanical cleansing of teeth and mucosa	Water
Lubrication of teeth and mucosa	Water, mucins
Keep oral mucosa intact, soft and moistened	Water, mucins, salts, epidermal growth factor, fibroblast growth factor, nerve growth factor
Prevent tooth demineralisation	Proline-rich proteins, statherins, cystatins, histatins, calcium and phosphate
Buffer capacity	Bicarbonate, phosphate and protein
Antimicrobial activities	
Antibacterial functions	Amylases, cystatins, histatins, mucins, peroxidase, lysozyme, lactoferrin, calprotectin, immunoglobulins, chromogranin A.
Fungicidal functions	Histatins, immunoglobulins, chromogranin A
Antiviral functions	Cystatins, mucins, immunoglobulins
Digestive properties	
Formation of food bolus	Water, mucins
Facilitation of mastication and swallowing	Water and, mucins
Initial digestion	-amylases, lipases, ribonucleases, proteases, water, mucins
Dissolution of taste compounds	Gustin (carbonanhydrase), zinc (Zn ²⁺), water
Facilitation of speech	Water, mucins

Adapted from- AML Pedersen. 2007, "Saliva", Zendium, available from

<<http://www.zendium.dk/Files/zendium.dk/material/publikationer/saliva.pdf>>

Mucins are vital in providing lubrication for speech, mastication and deglutination (Mandel, 1989). Antibacterial, antiviral and antifungal components like lactoferrins, lysozymes, histatins, salivary protease inhibitors, immunoglobulins etc. are important in providing host defence in the oral cavity (Mandel, 1989, Humphrey & Williamson,

2001). Electrolytes maintain pH, buffering and re-mineralization of enamel. Lavarge or mechanical cleansing action is brought about by water, which provides for the quantitative bulk of saliva.

In a healthy adult, an average of 800ml to 1.5 litres of saliva is produced daily (Hall, 2010). During the resting phase, 65% unstimulated saliva is produced by the submandibular gland, 20% by the parotid, 7-8% from the sub-lingual and the remaining from minor salivary glands. More than 50% of the saliva produced under stimulation is from the parotid gland (De Almeida et al., 2008). Ericsson & Hardwick (1978) classified whole saliva secretory rates as very low, low and normal. A normal resting flow is between 0.25ml and 0.35 ml per minute and a normal stimulated salivary flow rate is between 1ml to 3ml per minute. Salivary flow rates can vary among persons. A community based study by Yeh et al. (1998) showed that both resting and stimulated salivary flow reduced with age. This finding was also reported by Toida et al. (2010) in a large study population of 1188 adults. Although there is histological evidence of glandular parenchymal changes with increasing age, a number of studies have reported a decline only in the unstimulated flow rate and no significant reduction in the stimulated flow rate (Yeh et al., 1998; Percival et. al., 1994; Navazesh et. al., 1992b; Osterberg et. al., 1984). It appears that the submandibular salivary glands are more affected by age and or the parotid gland has a good response capacity. Most of the above studies show a decrease in salivary flow among females. This could possibly be due to the fact that males have submandibular glands that are up to 50% larger than those in females (Scott, 1975).

Nagler and Hershkovich (2005), in their age related study of salivary flow rates, found a significant increase in the concentration of some salivary components like protein, amylase, lactate dehydrogenase, IgA, IgG, potassium, chloride, phosphorus, calcium, magnesium and uric acid in the older age group (70 – 86 yrs). Although the concentrations were high, when the reduced salivary flow rate was taken into account, there was a significant reduction in the actual output of proteins, lactate dehydrogenase, IgA, IgG, sodium, calcium and magnesium. In addition to a decrease in the volume of saliva, the reduction in the total amount of available salivary components further compromises the protective functions of saliva and may account for an increase in oral diseases in the elderly (Nagler and Hershkovich, 2005). Age

related reduction in salivary mucin concentration in individuals above the age of 65 has also been recorded (Denny et al., 1991; Navazesh et al., 1992b).

Ship et al. in 1995 did not see any age or gender related changes in salivary flow in healthy, non-medicated individuals. In 1993, Wu & Ship proposed that aging does not affect salivary gland flow, per say, but a reduction in salivary gland function, especially in the submandibular gland is due to increasing systemic problems and associated poly-medication. “The submandibular gland may be more sensitive to physiologic permutations than the parotid gland.” (Wu & Ship, 1993). A possible reason why the complaints of xerostomia increase with age, even though there is no actual reduction in salivary flow, is due to reduced concentration of mucins. Mucin production by minor salivary glands is important in lubrication of the oral cavity, and reduced concentrations of it can be perceived as dry mouth (Navazesh et. al., 1992b). A Swedish study on 1427 people, by Flink et al., (2008) has shown that the prevalence of hyposalivation among the different age groups did not vary much until the age of 50 for women and 60 for men. There is a consensus that advanced age cause changes to salivary gland physiology.

Apart from inter-individual differences, salivary flow rates show up to 45% intra-individual variations. (Ghezzi et. al., 2000) This is due to circadian (Dawes, 1972) changes. Hydration can also influence flow rate in a healthy individual (Ship and Fisher, 1997). All saliva collection in this study was done between 9am and 3pm and all patients were requested not to drink or eat for 90 minutes before saliva collection.

2.2. SALIVARY GLAND HYPOFUNCTION

The term “salivary gland hypofunction” may be used to cover xerostomia, hyposalivation and salivary compositional changes (Nederfors, 2000) A more updated definition should also include other oral sensory changes like burning mouth and taste dysgeusia. Each of these inter-related entities can exist together or independent of each other; making a co-relation difficult to establish.

2.2.1 Xerostomia

Xerostomia is the perceived feeling of dry mouth. The causes encompass those of salivary gland origin, and those that are not of salivary gland origin. Although a strong co-relation exists between xerostomia and reduced salivary flow rate (Osterberg et al., 1984; Farsi, 2007), it may not always be the cause of xerostomia (Sreebny & Valdini, 1988; Fox et al., 1987; Toida et. al., 2010).

Alterations in salivary composition, in particular, glycoproteins like mucins, can also cause patients to report xerostomia. Mucins provide a moistening and lubricating function, and a reduction in mucin can increase the symptoms of mucosal dryness (Mandel, 1989, Navazesh et al., 1992b). All the etiological causes for reduced salivary flow (listed in table 2.2) can also be considered as salivary causes for xerostomia.

Non-salivary causes for xerostomia include dehydration, mouth breathing, cognitive alteration and psychological disorders (Fox, 1996). Atkinson et al. (2005) suggested that xerostomia complaints may not necessarily be due to hyposalivation, but could reflect dehydration or other systemic conditions. Narhi (1994) and Fox et al., (1987) have proposed that, since perception of oral dryness is related to mucosal dehydration, non-uniform wetting of the mouth or localised areas of dryness can increase xerostomic perception.

Table 2.2 – Causes of Salivary Gland Hypofunction and Xerostomia

1. Water/Electrolyte loss
Dehydration Reduced water intake Loss of water through the skin (fever, burns, excessive sweating) Blood loss Vomiting Diarrhoea Renal water loss: Polyuria (Diabetes Insipidus) Osmotic diuresis (Diabetes Mellitus) Malnutrition Bulimia Anorexia
2. Changes to the salivary glands
Therapeutic irradiation to the head and neck region Autoimmune diseases Sjogren’s syndrome, graft-versus-host-disease, systemic lupus erythematosis rheumatoid arthritis, etc. HIV-1 infection Infections Hepatitis C HIV Ageing Local Sialadenitis Sialolithiasis
3. Interference with neural transmission
Medications/drugs Autonomic dysfunction Cerebrovascular disease, brain tumors etc. Conditions affecting the CNS Alzheimer's disease Parkinson’s disease Psychogenic disorders Depression Anxiety Stress Trauma to the nerves involved in salivary secretion Decrease in mastication

Adapted from- Sreebny, L.M. & Schwartz, S.S. 1997, "A reference guide to drugs and dry mouth–2nd edition", Gerodontology, vol. 14, no. 1, pp. 33-47.

According to a review by Orellana et al., (2006), the prevalence of xerostomia, in most of the studies until then, ranged from 0.9% to 64.8%. This population review once again highlights the wide range of prevalences for xerostomia that has been reported. Osterberg et al. (1984) conducted one of the first large-scale studies on the prevalence of xerostomia. 1148, seventy year old residents from Goteborg, Sweden, partook in it. In this study 25% of women and 16% of men reported xerostomia. In 1992, Narhi et al. did a study on 368 subjects and found a prevalence of 46% for xerostomia among 75-85 year olds. The older age group has also shown prevalences as high as 52% (Putten et. al., 2011). Studies that reflect the population at large include those by Sreebny and Valdini (1988), Nederfors et al. (1997) and Flink et al. (2008). A prevalence of 29% was seen in the study by Sreebny and Valdini (1988). 3311 individuals of all ages were included in the study by Nederfors et al. (1997) and it showed a xerostomic prevalence of 28% for females and 23% for males. In all of the studies, females were more prone to xerostomic complaints. Flink et al. (2008) have shown in their study that the prevalence of xerostomia and hyposalivation did not vary much until the age of 50 years for women and 60 years for men.

Interestingly, xerostomia or dry mouth is rarely a primary complaint among patients. Being a subjective symptom, like pain, it has to be taken at face value. Diagnosis of xerostomia can be made either from a positive response to just the single question “Does your mouth feel dry often?”, (Osterberg et. al., 1984; Flink et al., 2008; Toida et. al. 2010) or from a response to a multi-item inventory of up to 16 xerostomia related questions (Narhi, 1994).

Thomson et al. (1999a) made an 11 item inventory that reflects both the individual’s awareness of xerostomia and actions taken as a consequence of the symptoms. This inventory is considered as the standard for xerostomia assessment in the Australasian region. Most authors from the North American region use a set of four questions developed by Fox et al. in 1987 (Navazesh et. al. 2009; Napeñas et al., 2009; Ship, 2002). In 1988 Sreebny and Valdini selected four questions from their inventory and found that these questions had a high predictive value and specificity for detection of reduced salivary flow rate. These four questions have been used in this study to assess xerostomia. Apart from “yes or no” answers, a graded response of “never”, “hardly

ever”, “occasionally”, “frequently” or “always” is also used (Thomson et. al., 1999a).
Table-2.3 gives a list of questions used in some of these inventories.

Table 2.3 – Examples of Xerostomia Inventories

Fox PC, Busch KA, Baum BJ. 1987, “Subjective reports of xerostomia and objective measures of salivary gland performance”, *J Am Dent Assoc*, Vol. 115, no.4, pp. 581-584.

1. Does the amount of saliva in your mouth seem too little?
2. Does your mouth feel dry when eating a meal?
3. Do you have difficulty swallowing any food?
4. Do you sip liquids to aid in swallowing dry food?

Response options: Yes/ No

Sreebny L M, Valdin A. Xerostomia. 1988, “Part I: Relationship to other symptoms and salivary gland hypofunction”, *OralSurg OralMed Oral Pathol*, vol. 66, no. 4, pp.451-458.

1. Does your mouth usually feel dry?
2. Do you regularly do things to keep your mouth moist?
3. Do you get out of bed at night to drink fluids?
4. Does your mouth usually become dry when you speak?

Response options: Yes/ No

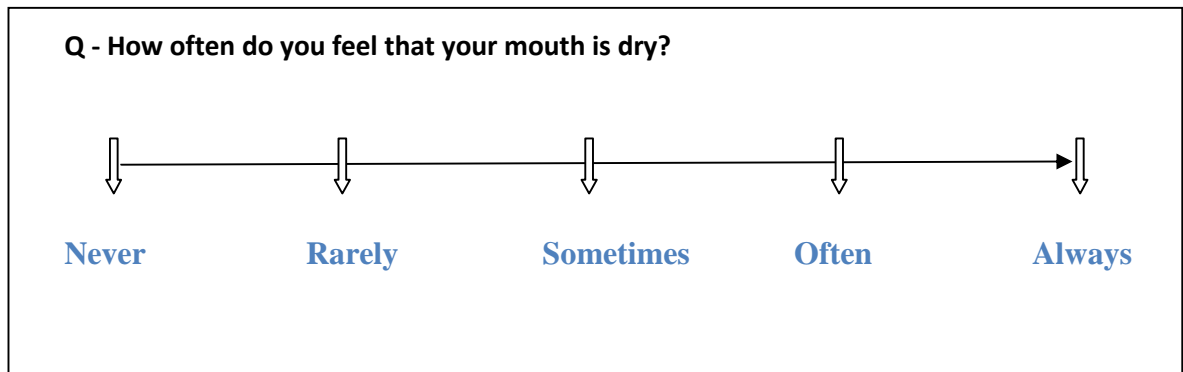
Thomson WM, Chalmers JM, Spencer AJ, Williams SM. 1999, “The Xerostomia Inventory: a multi-item approach to measuring dry mouth”, *Community Dent Health*, vol. 16, no. 1, pp. 12-7.

1. I sip liquids to aid in swallowing food
2. My mouth feels dry when eating a meal
3. I get up at night to drink
4. My mouth feels dry
5. I have difficulty in eating dry foods
6. I suck sweets or cough lollies to relieve dry mouth
7. I have difficulties swallowing certain foods
8. The skin of my face feels dry
9. My eyes feel dry
10. My lips feel dry
11. The inside of my nose feels dry

Response options: Never (scoring 1), Hardly ever (2), Occasionally (3), Fairly often (4), Very often (5)

A visual analogue scale, similar to the pain scale, can also be used on a colorimetric or graduating scale of 1 to 10 (Pai et. al., 2001; Lopez-Verdin et al., 2013). Fig -2.1 shows an example of a visual analogue scale that can be used in evaluating xerostomia.

Figure 2.1 - Visual Analogue Scale for a xerostomia question.



2.2.2 Hyposalivation

Hyposalivation is a demonstrable reduction in salivary flow rate that can be measured objectively by collecting saliva over a period of time (Navazesh et al., 1992a; Navazesh, 1993). While 58% of the xerostomic subjects in one study showed hyposalivation (Torres et. al., 2002) and 54% in another (Sreebny & Valdini, 1988), only 22.1% was observed by Thomson et al (1999b). A positive correlation of xerostomia and reduced salivary flow is not always found.

Some of the causes for reduction in salivary flow include: medication, radiotherapy, systemic diseases, localised salivary gland disease and dehydration. These causes may be classified according to the way it affects the salivary glands – damage to gland, neurotransmission alterations and water loss in the body (Sreebny & Schwartz, 1997). Table 2.2 gives a list of some of these causes. There are over 400 pharmaceutical drugs that are implicated in causing xerostomia (Sreebny & Schwartz, 1997). Evidence of reduced salivary flow is documented for some of them (Navazesh et al., 1996; Sreebny & Valdini, 1989; Wu and Ship, 1993; Osterberg et. al., 1984; Putten, 2011). Anti-hypertensives, anti-retrovirals, anti-depressants, anti-cholinergics, anti-psychotics, anti-cancer drugs etc are some of the common culprits. A summarised list of those medications with the potential to reduce salivary flow is given in Table-2.4.

Table 2.4 – Medications that have potential to reduce salivary flow rate

Anticholinergic agents Antiemetics/drugs for vertigo Anti-parkinsonian drugs Antispasmodics: Gastrointestinal Mydriatics/cycloplegics
Medications with anticholinergic effects Antiarrhythmics: Sodium channel antagonists Antihypertensive agents: -adrenergic blockers -blockers calcium antagonists Antihistamines Antidepressants Antipsychotics Monoamine oxidase inhibitors
Psychotropic agents Anxiolytic agents: Benzodiazepines
Medications causing changes in fluid and electrolyte balance Diuretics: Thiazides Loop diuretics Potassium sparing diuretics
Others Antineoplastic agents: Interleukin-2a

Adapted from- Närhi, T.O., Meurman, J.H. & Ainamo, A. 1999, "Xerostomia and hyposalivation", Drugs & aging, vol. 15, no. 2, pp. 103-116.

Some drugs, although causing xerostomia, do not actually cause a reduction in salivary flow (Atkinson et. al., 1989a). The different ways that therapeutic drugs can affect salivary gland function is through disruption of neural salivatory function, vasoconstriction within the gland or electrolyte loss affecting salivary gland function and xerostomic perception. Most of the salivary dysfunction due to medication is reversible on stopping the drug. There have been a number of studies that show a negative co-relation between the salivary flow rate and the number of medication and systemic diseases. (Wu & Ship, 1993; Osterberg et al., 1984; Nederfors et al., 1997).

Radiotherapy as part of treatment for head and neck cancer causes irreversible destruction of the salivary gland parenchyma. The parotid glands are most sensitive to ionising radiation, showing up to a 50% decrease in salivary flow within 24 hours of radiation (Shannon et. al., 1978).

Several systemic conditions can cause salivary dysfunction. The most drastic of these is Sjogren's syndrome (SS). SS is an autoimmune disease, mostly seen in women, involving salivary and lacrimal glands. It is characterised by lymphocyte infiltration of salivary and lacrimal glands, causing inflammation, acinar destruction and decreased secretion (Fox, 2005). Xerostomia and hyposalivation are major concerns for sufferers of this condition. Some of the other systemic conditions associated with salivary gland dysfunction are HIV/AIDS (Mandel et al., 1992, Yeh et al., 1988), diabetes mellitus (Sreebny et al., 1992) cystic fibrosis, hepatitis C, sarcoidosis, rheumatoid arthritis, systemic lupus erythematosus and scleroderma (Von Bultzingslowen et al., 2007). Compositional changes in the saliva are also seen in systemic conditions like HIV and diabetes. Hyposalivation is also seen in transplant patients who develop Graft Vs Host disease (Mandel, 1990).

Dehydration has been known to cause reduced salivary flow (Ship & Fisher, 1997; Napeñas et al., 2009). Dehydration can be due to reduced water intake, haemorrhage, vomiting, diarrhoea or renal water loss. Excess alcohol intake and anorexia can also cause reduced salivary flow. The mechanical action of chewing stimulates salivary flow. Therefore, where there is reduced mastication, due to loss of teeth, there is reduced salivary flow (Osterberg et al, 1984). Localised gland diseases like bacterial

sialadenitis, sialolithiasis and salivary gland tumors and cysts can also cause hypofunction.

It may be noted at this point, that hyposalivation may be a symptom of a range of underlying conditions, some of which are physiological, some systemic and more sinisterly, some pathological. Therefore, when present, diagnosing hyposalivation and exploring its reasons can be critical not only for the oral health but also for the general wellbeing of the patient.

An accurate presentation of the prevalence of hyposalivation is difficult since, in some studies, hyposalivation is described as a flow rate below a certain value (Ericsson & Hardwick, 1978; Napeñas et al., 2009; Navazesh et al., 2003; Flink et al., 2008; Osterberg et al., 1984; Sreebny & Valdini, 1988; Navazesh et al., 1992a), and in some studies, hyposalivation was determined as values below the 10th percentile (Farsi, 2007; Wu & Ship, 1993). Furthermore, different values were used as cut-offs for abnormal flow rate by different authors. Nevertheless, literature reviews of some of the studies give a prevalence of 20.2% for hyposalivation (Flink et. al., 2008), in a study of different age groups and 30-33% (Osterberg et al., 1984), in an older age group.

Subjective measures of salivary flow rates can be made by collecting resting/unstimulated saliva and stimulated saliva from the whole mouth or individually from the different salivary glands (Navazesh, 1993). The saliva is collected in a tube over a period of time, usually 3 to 5 minutes, and the flow rate is then calculated. Resting whole mouth saliva is collected by drooling or spitting into a tube fitted with a funnel. Mechanical (paraffin wax, gum, rubber band), or gustatory (1-10% citric acid solution, candy), stimulation may be used to collect stimulated saliva. Parotid gland saliva is collected using a lashley cup or a modified Carlsson-Crittendan device. Submandibular and sublingual saliva can be isolated and suctioned gently into a tube. Minor salivary gland secretions can be collected with a micropipette of filter paper and quantity can be measured using a Periotron (Navazesh, 1993).

Although slight variations exist in the cut-off values for determining abnormal flow rate, most authors agree on a cut off of 0.1 ml/min for resting and 0.7 ml/min for

stimulated whole-mouth saliva flow rate. (Navazesh et. al., 2003; Flink et. al., 2008; Ericsson & Hardwick, 1978; Sreebny & Valdini, 1988)

Clinical signs and symptoms that may help with diagnosing hyposalivation include dry mucosa (tongue depressor sticks to dry buccal mucosa), dry lip (Navazesh et al., 1992a; Farsi, 2007), dry fissured tongue, lack of salivary pooling in the floor of the mouth, increased dental caries (especially cervical and incisal caries) candidial infection (Torres et. al., 2002; McCarthy et al., 1991), difficulty in denture retention and enlarged (sometimes tender), salivary glands (Navazesh et al., 1992a). Salivary flow can also be measured by Scintigraphy. Uptake and secretion of Technicium per Technatate can be measured to determine accinar function (Napeñas et al., 2009).

2.2.3 Compositional Changes

Salivary compositional changes, due to gland dysfunction, can be observed in the presence of systemic diseases like pancreatitis, diabetes mellitus (Mandel, 1990), depression (De Almeida et. al., 2008) and Graft Vs Host disease (Mandel, 1990). Other conditions include nutritional deficiencies (Humphrey & Williamson, 2001), increased alcohol consumption (Mandel, 1990), and increased physical exercise (De Almeida et. al., 2008). Salivary compositional changes in HIV positive patients were also noted by Mandel et al. (1992), Atkinson et al., (1990), Jainkittivong et al.,(2009) and Lin et al. (2001 & 2003).

2.3. HIV AND HAART

In the 33 years since the first case of AIDS was recognized in 1981, HIV infection has evolved into a condition affecting 34 million people worldwide (Joint UN program on HIV/AIDS, 2012). Meanwhile, a large amount of resources and manpower has been poured globally into research bodies towards an increase in knowledge, exploring treatment modalities and prevention strategies for this pandemic. Infection by HIV is from transfer through blood, blood products and bodily fluids. Although, initially it was discovered in the homosexual community, today, most of the transmission globally is heterosexual. Intravenous drug users and people receiving blood or blood products are also at risk. Most of the HIV positive children have been infected by vertical transmission from mother to child.

The average person with HIV infection produces 10 billion virions per day (Ho et. al., 1995). Anti retroviral treatment (ARVs) is the key to HIV/AIDS management. Before 1987, treatment for HIV/AIDS consisted of the symptomatic treatment of immunodeficiency complications. Zidovudine (AZT) was the first US govt approved ART introduced for HIV treatment. Initially it was used in monotherapy and later in a two drug regimen along with Lamivudine (3TC)

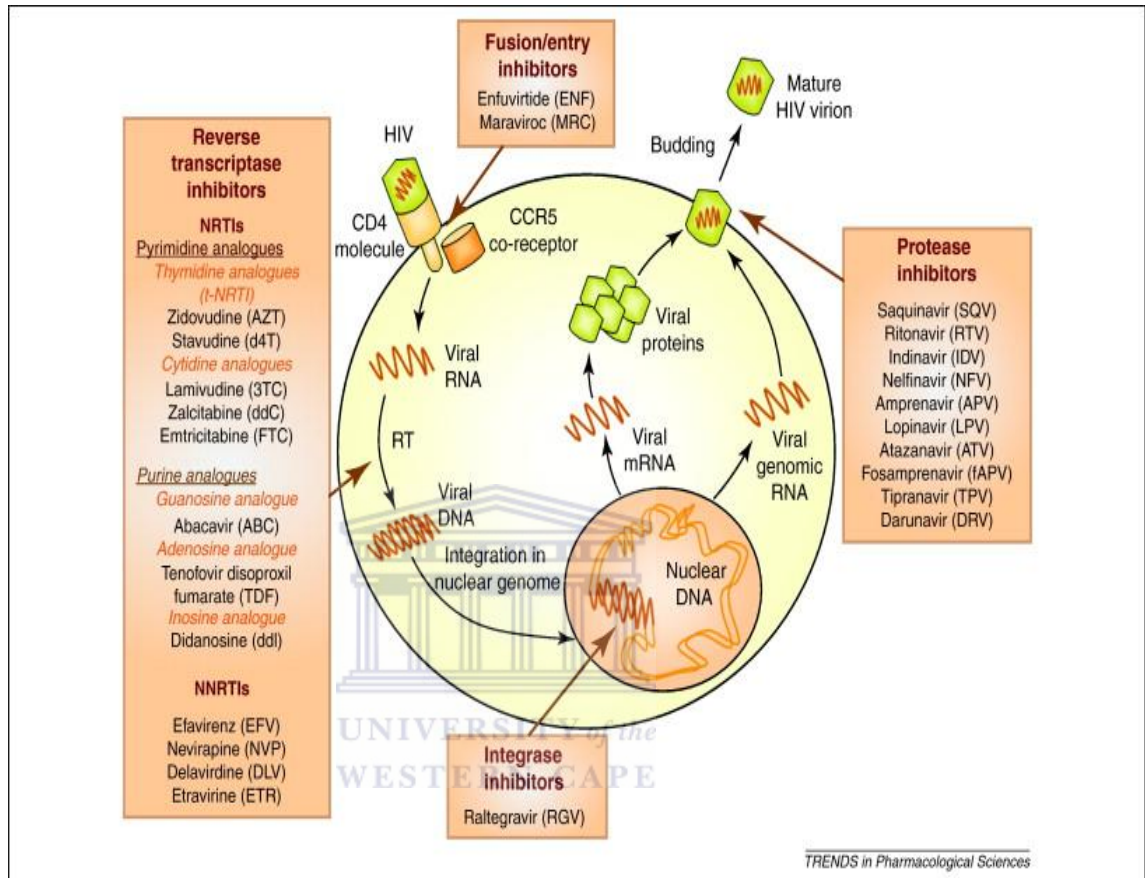
2.3.1 HAART

In the mid 1990s, David Ho and other researchers proposed the idea that a three drug regimen was the answer to hitting the virus hard and reducing its chance of developing resistance. This approach was called Highly Active Anti Retroviral Therapy (HAART). HAART has effectively turned the tide of HIV/AIDS mortalities and made it a chronic manageable disease rather than a death sentence (ART Cohort Collaboration, 2008).

There are different types of ARVs and they are mainly classified into 5 groups depending on their action in the life cycle of the HIV – Nucleoside Reverse Transcriptase Inhibitor (NRTIs), Non-nucleoside Reverse Transcriptase inhibitors (NNRTIs), Protease Inhibitors (PIs), Integrase Inhibitors (IIs), and entry or fusion inhibitors. Figure-2.2 illustrates the sites in the life cycle of the HIV upon which the

antiretro-viral drugs may act. Table-2.5 lists the classifications of the various ARVs that are able to act on the HIV at the different sites of its life cycle.

Figure 2.2- Blocking of HIV replication by ARVs



Taken from: Apostolova, N., Blas-García, A., & Esplugues, J. V. 2011, "Mitochondrial interference by anti-HIV drugs: mechanisms beyond Pol-inhibition", Trends in pharmacological sciences, Vol.32, no.12, pp.715-725.

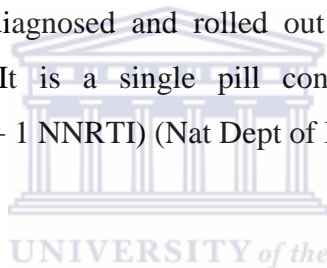
Table 2.5 - Classification of Anti Retrovirals

Class	Site and Mechanism of Action	Example
NRTI-Nucleoside/ Nucleotide Reverse Transcriptase Inhibitor (NtRTI)	When the Viral RNA is converted to DNA- Competitive inhibitor of reverse transcriptase	NRTI- e.g.- Zidovudine (AZT), Lamivudine (3TC), Didanosine (ddI), Stavudine (d4T), Emtricitabine (FTC), and Abacavir (ABC) NtRTI- e.g.- Tenofovir (TDF)
NNRTI- Non Nucleoside Reverse Transcriptase Inhibitor	When the Viral RNA converts to DNA- Non competitive inhibitor of Reverse Transcriptase	Efavirence (EFV), Nevirapine (NVP)
PI- Protease Inhibitors	Prevent cleavage of the viral proteins	Lopinavir, Ritonavir (RTV), Indinavir (IDV)
II- Integrase Inhibitors	Inhibits the integration of Viral DNA into the DNA of the cell	Raltegravir (RAL), Elvitegravir
Entry Inhibitors or Fusion Inhibitors	Interfere with binding, fusion and entry into the host cell	Maraviroc (MVC), Enfuvirtide

HAART regimens usually consist of 3 or more anti retrovirals from at least 2 different classes ensuring antiviral action at different stages of HIV replication. HAART regimens may consist of any of the combinations below:

- 2 NRTIs + 1 NNRT
- 2 NRTIs + 1 PI
- 2 NRTIs + 2 PIs
- 2 NRTIs + 1 II.

In South Africa the HAART regimens currently in use are 2 NRTIs + 1 NNRTI for first line of treatment and 2 NRTIs + 1 PI for second line of treatment. In April 2013, the National Department of Health launched a Fixed Drug Combination (FDC), to be initiated in the newly diagnosed and rolled out in phases to the different groups according to priority. It is a single pill containing Efavirenz, Tenofovir and Emtricitabine (2 NRTIs + 1 NNRTI) (Nat Dept of Health, 2013).

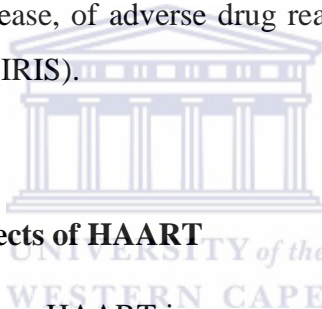


2.3.2 Oral manifestations of HIV in the HAART era

Since the early days of its emergence, HIV has been associated with a variety of oral lesions. A good comparison of the studies on oral manifestations in HIV was done by Patton et al. in 2002 and more recently, in 2013. The pre HAART prevalence for oral manifestations was 92% (Klein et. al., 1991), 82% (Nittayananta & Chungpanich, 1997) and 60% (Arendorf et. al., 1998). A clear decline in oral manifestations was seen with the advent of HAART (Greenspan et. al., 2004; Schmidt-Westhausen et. al., 2000; Ortega et. al., 2009; Masiwa & Naidoo, 2011). The most significant change was seen in the reduction of oral candidiasis, oral hairy leukoplakia and Kaposi's sarcoma (Greenspan et. al., 2004; Nittayananta et. al., 2010a; Schmidt-Westhausen et. al., 2000; Hamza et. al., 2006; Ortega et. al., 2009). Apart from improving immune status, PIs have been shown to reduce candidial infection. This could possibly be due to the direct protease inhibitory action by the PIs on the candidial aspartyle proteases (Cassone et al., 2002; Munro & Hube, 2002). In Zimbabwe, Masiwa & Naidoo (2011) conducted a prospective study to determine the prevalence of oral lesions before and after

HAART. Manifestations of oral lesions reduced from 86% at baseline to 57% in 6 months. Although a decrease in most oral lesions is seen with HAART initiation, an increase in prevalence was noted for some other lesions like oral pigmentation (Umadevi et. al., 2007), oral warts (Masiwa & Naidoo, 2011; Patton et. al., 2000; Greenspan et. al., 2001), and HIV associated salivary gland disease (HIV-SGD) (Greenspan et. al., 2001; Patton et. al., 2000; Naidoo & Chikte, 2004; Matee et. al., 2000).

Oral manifestations are commonly useful in indicating HIV infection, predicting immune status and in staging and classification of the disease (Coogan et. al., 2005). In the HAART era, oral manifestations can still be useful as indicators of reduced immunity due to treatment failure from non-adherence or development of drug resistance (Greenspan & Greenspan, 2002). Distinctions should still be made between manifestations of the disease, of adverse drug reaction and of immune reconstitution inflammatory syndrome (IRIS).



2.3.3 Orofacial Side Effects of HAART

As the number of people on HAART increases, and the drugs used in therapy become more complex, the incidences of undesirable or adverse effects will only increase. They may vary from mild oral pigmentations to a serious systemic reaction like erythema multiforme (Dios & Scully, 2014). Reports of clinical symptoms and laboratory adverse effects can be up to 78% (Fellay et. al., 2001), during HAART.

With NRTIs, especially Zidovudine, bone marrow suppression may be observed. This can manifest as oral ulcerations secondary to neutropenia (Dios & Scully, 2013). Other oral side effects include xerostomia, erythema multiforme and lichenoid reactions (Dios & Scully, 2014). Abnormal liver functions may be seen with Lamivudine and Stavudine. Hyperpigmentation is listed as a side effect for Emtricitabine. Xerostomia is associated with didanosine (Allan et. al., 1993), Emtricitabine and Lamivudine (Dios & Scully, 2014). Facial lipoatrophy is seen in treatment with Stavudine and Tenofovir. NNRTIs generally are tolerated well and have less oral side effects. Erythema Multiforme and ulcers are the most common expression of hypersensitivity

on treatment with NNRTIs (Dios & Scully, 2014). PIs have been associated with parotid lipomatosis (Olive et. al., 1998; Ceballos-Salobrena et al., 2000), xerostomia, reduced salivary flow (Navazesh et. al., 2003), taste disturbances (Murri et. al., 2000) and circumoral parasthesia (Porter & Scully, 1998). The exact mechanism of ARVs causing reduced salivary flow is unknown. It may be a result of the direct action of the drug leading to acinar changes or lipotrophic tissue changes within the gland causing an anti secretory effect (Navazesh et al., 2003; Olive et al., 1998). A concise list of the orofacial effects of the various classes of ARVs can be seen in table- 2.6

Table 2.6 – Orofacial adverse effects of ARVs

Drug	Possible Orofacial adverse effects
Abacavir (ABC)	Erythema multiforme
Didanosine (DDI)	Dry mouth Facial lipoatrophy Erythema multiforme
Emtricitabine (FTC)	Hyperpigmentation Dry mouth
Lamivudine (3TC)	Dry mouth
Stavudine (D4T)	Facial lipoatrophy
Tenofovir disoproxil (TDF)	Facial lipoatrophy,
Zidovudine(azidothymidin, AZT, ZDV)	Erythema multiforme Hyperpigmentation Lichenoid lesions Facial lipoatrophy Ulcers
Efavirenz (EFV)	Erythema multiforme Swelling Burning mouth Clefts
Nevirapine (NVP)	Erythema multiforme Swelling Ulcers Taste disturbance Lichenoid lesions Dry mouth Burning mouth

Indinavir (IDV)	Cheilitis Parotid lipomatosis Dry mouth Taste disturbance
Ritonavir (RTV)	Perioral paraesthesia Parotid lipomatosis Dry mouth Taste disturbance Swelling

Adapted from- Diz Dios, P. & Scully, C. 2014, "Antiretroviral therapy: effects on orofacial health and health care", Oral diseases, vol. 20, no. 2, pp. 136-145.

2.3.4 Immune Reconstitution Inflammatory Syndrome (IRIS)

IRIS is the worsening of a pre-existing condition or an unmasking of an occult infection in an immunocompromised patient on HAART. It is not due to drug toxicity and occurs within weeks or a few months of treatment initiation. This is seen in conjunction with a quantitative increase in CD4 count and a decrease in viral load (Zolopa et al., 2009). Disease relapse, drug resistance and non-compliance have to be excluded before making this diagnosis (Shelburne et. al., 2006). Approximately 25%-32% of all patients on HAART develop IRIS (Feller et. al., 2007). Ramirez-Amador et al. (2009) suggested an inclusion of oral candidiasis, oral hairy leukoplakia and recurrent oral ulcers as possible manifestations of IRIS. Feller et al. (2007, 2008) reported Herpes Zoster and Kaposi's sarcoma as possible oral manifestations of IRIS. HPV associated oral lesions that were seen to be on the increase in patients on HAART could also be a part of IRIS (Greenspan et. al., 2001; Patton et. al., 2000; Flint et. al., 2006)

A retrospective study by Ortega et al. (2009) reported salivary gland enlargement as the most common oral manifestation in a group presenting with IRIS. An increase in salivary gland disease in patients on HAART was also seen by Greenspan et al. (2001). This could be due to infiltration of the salivary gland by CD8 lymphocytes as part of Diffuse Infiltrative Lymphocytosis Syndrome (DILS). IRIS may be a manifestation of a recovering immune system that is functionally incomplete and so its

effectiveness may vary in response to the presence of different pathogens and antigens (Greenspan et. al., 2001).



2.4. HIV ASSOCIATED SALIVARY GLAND DISEASE (HIV-SGD)

In 1989, Schiodt et al. (1989) first used the term HIV associated Salivary Gland Disease (HIV-SGD) to describe a group of salivary gland disorders that affected the HIV positive population. Non-neoplastic changes like Benign Lymphoepithelial Cysts (BLEC), diffuse infiltration of salivary glands by CD8 lymphocytes, Sjogren's syndrome-like condition and Sicca complex were all seen in HIV positive patients. How much of it was overlap, was not clear. Hence, he coined the term HIV-SGD to designate "Xerostomia, enlargement of one or more of the major salivary glands, or both" (Schiodt et al., 1989). This was essentially what was clinically observed in the above conditions – salivary gland enlargement and/or xerostomia in the HIV positive individuals. Since then, compositional changes in the saliva by HIV infection have also been reported (Mandel et. al., 1992; Atkinson et. al., 1990; Lin et. al., 2001, 2003, 2004; Jainkittivong et al., 2009; Johar et. al., 2011).

HIV-SGD is often pathognomic of HIV in the paediatric population and is not observed as much in the adult population. Naidoo and Chikte (2004) reported a prevalence of 50% for Parotid gland enlargement in 71 institutionalised HIV + children in South Africa. None of them were on Anti Retroviral Treatment. In 2012, Sales-Peres et al. observed 23% Parotid Gland Enlargement (PGE), and 76% hyposalivation among HIV/AIDS patients in a paediatric hospital in Mozambique. Some of these children were on ART. In a Tanzanian study, Hamza et al. (2006), reported PGE in 18.2% of the children who were pre-HAART and this rose to 20% in those who were on HAART. They also remarked on the lack of significant decrease in other oral manifestations among children who were on HAART.

In the same study, Hamza et al. (2006) also reported 2.2% prevalence for PGE among the adult population who were not on HAART and this rose to 4.9% among those who were on HAART. Although there was a significant decline in many of the other oral manifestations, the sharp increase in PGE could be attributed to IRIS. In their study, Ceballos-Salobrena et al. (2000) noted a prevalence of 15.5% for xerostomia and 4.5% for PGE among the HIV positive subjects. All of the study subjects were being treated with HAART which included PIs. It does, however, remain unclear whether the PGE and xerostomia could be attributed to HIV infection, HAART or a combination

thereof. As mentioned earlier, many ARVs are implicated in causing xerostomia and reduced salivary flow, especially the PIs. PGE due to lipomatous changes have also been observed during treatment with PIs (Olive et al., 1998; Ceballos-Salobrena et al., 2000)

Among the adult population, HIV-SGD does not seem to be limited to any particular risk group in terms of mode of transmission and can occur in any stage of the disease (Schiødt et al., 1992). HIV-SGD is prominently associated with DILS (Diffuse Infiltrative Lymphocytosis Syndrome) where CD8 lymphocytic infiltration is seen in other organs as well. According to Itescu et al. (1990), DILS appears to have a predilection to the male black population. McArthur et al. (2000) has estimated the prevalence of DILS in a Cameroonian study of AIDS patients to be 48%. Although, according to Itescu et al. (1993), and Rivera et al. (2003), DILS appears to have a predilection to the male population, a recent study by Kungoane (2010) showed that lymphoid proliferation among children had an almost equal gender distribution.

The exact mechanism of salivary gland dysfunction due to HIV infection is not clearly understood. While some investigators like Greenberg et al. (1997) have suggested a strong relationship between xerostomia and a co-infection with the Cytomegalovirus in HIV patients, most are agreeable on lymphocytic infiltrate of the glandular tissue as the reason for HIV-SGD (Schiødt et al., 1992).

2.4.1 Pathophysiology

During embryonic development, a few (5-10) lymph nodes are trapped in the Parotid gland tissue, and trapped within these lymph nodes, are pieces of salivary gland tissue. During the course of HIV infection, due to viral replication, there is an initial host response to lymphoid proliferation. This is characterised by the generalised lymphadenopathy seen in the disease. The lymphoproliferative activity within the intraglandular lymph nodes or by the lymphocytes normally present in the gland can cause gland enlargement (Mandel & Reich, 1992).

In the early stages, glandular tissue and lymph nodes show varying degrees of progressive CD8 lymphocyte infiltration, which can later lead to cystic changes

(Mandel et. al., 1998; Itescu et. al., 1990, 1993). Initially, cystic formation was thought to be due to the lymphoid proliferation causing ductal obstruction (Ioachim & Ryan, 1988). Microscopic studies have shown epithelial cells lining the cyst (Elliott & Oertal, 1990). These are thought to be derived from gland accini and ductal tissue that were trapped within the intraglandular lymph node during embryogenesis. Thus, a proliferation of this tissue within the lymph nodes is implicated in the formation of cysts filled with keratinaceous and proteinaceous debris (Mandel et. al., 1998).

Due to the differences during embryonic development, the lymph nodes associated with the submandibular gland stay extra glandular and hence the lymphadenopathies associated with these glands are extra glandular (Ioachim & Ryan, 1988; Mandel & Reich, 1992). Histologically, apart from diffuse CD8 lymphocyte infiltrates, the parenchymal tissues also show degeneration (Schiødt et. al., 1992). Periductal lymphocytic infiltration, accinar atrophy and ductal ectasia with varying amounts of fibrosis were noted by Rivera et al. (2003).

Itescu et al. (1990) reported a decrease in the CD4/CD8 ratio in patients with HIV related parotid gland enlargement (PGE) and stated that they have a more favourable progress. Katz et al. (1993) has also indicated that the presence of salivary gland enlargement is associated with a slower progression to death. This may be due to the protective effect of the increased CD8 T-Cells that are seen in these conditions. Salivary gland enlargement due to lymphocytic infiltration is a hallmark of DILS (Itescu et al., 1990); seen in some HIV positive patients and not in others. It could also mark a favourable ability by this subset of the HIV infected population to mount a defensive response to the HI Virus (Itescu et al., 1990; Jeffers & Webster-Cyriaque, 2011).

DILS is characterised by swelling of the salivary and lacrimal glands. There is an increased CD8 lymphocyte infiltration of these glands and the kidney, nerves, muscles, lungs, gastric mucosa and lymph nodes (Itescu et al., 1990; Rivera et al., 2003). DILS of the salivary gland mimics Sjogren's syndrome and so it is sometimes referred to as an SS-like condition. Although the clinical signs and symptoms are similar, the differences are unmistakable. While in DILS and related HIV-SGD, the lymphocyte infiltrate is predominantly CD8, in SS, there is an increase in the CD4

lymphocyte within the glandular tissues. There is also a lack of autoimmune antibodies in DILS (Itescu et al., 1990; Rivera et al., 2003). SS is almost always seen in women, but in reports of DILS, there seems to be a male predilection (Rivera et al., 2003). Viral etiologies have been proposed for both (Jeffers & Webster-Cyriaque, 2011).

According to Itescu et al. (1990), the CD8 infiltration appears to be a genetically determined (major histocompatibility) host immune response in the presence of HLA-DR5; possibly, to a viral antigen (Itescu et al., 1993; Mandel et al., 1998). Many virological etiologies have been investigated for HIV-SGD. HIV (Mandel et al., 1998; Rivera et al., 2003; Itescu, 1990), Cytomegalovirus (CMV) (Greenberg, 1997), Epstein Barr Virus (EBV) (Rivera et al., 2003), and recently the BK Virus (Jeffers & Webster-Cyriaque, 2011; Burger-Claredon, 2014), have all been proposed as causative pathogens. Over time, CMV has been ruled out by some investigators (Rivera et al., 2003; McArthur, 2000). The presence of the EBV antigen and HIV1-p24 antigen within the ductal epithelial cells has been demonstrated by Rivera et al. (2003). Lack of significant evidence to imply EBV as a cause has ruled it out (Chetty et al., 1999).

In 2009, Jeffers et al. were able to demonstrate tropism by BKV, a polyoma virus, for salivary gland cells. They detected BKV in the saliva of patients with HIV-SGD and proposed an oral route of transmission. Symptoms of BKV in generally healthy people are subclinical; followed by viral dissemination to sites like urinary tracts and kidney for life-long persistence (Jeffers & Webster-Cyriaque 2011). Like most opportunistic pathogens, clinical manifestation occurs during immunosuppression. Since HIV-SGD is seen to be more prevalent in children than adults, Jeffers & Webster-Cyriaque (2011) postulated that “this may indicate primary viral infection Vs residual immunity in adults”. Currently, a clinical trial has been undertaken by the University of North Carolina to learn more about the role of BK virus in Salivary gland diseases and to determine its relationship to HIV-SGD (Webster-Cyriaque, 2014).

Apart from the parotid glands, HIV-SGD also affects the submandibular, sublingual glands and minor salivary glands (Mc Arthur et al., 2000; Schiodt et al., 1989).

2.4.2 Clinical Presentations

Clinically, HIV-SGD can present as any of the following in an HIV positive patient.

- Enlargement
- Symptoms of Xerostomia
- Changes in salivary flow rate
- Changes in salivary composition.

The above may be present alone or along with the other signs and symptoms. Salivary gland enlargement is more readily noted in the parotid than in the submandibular or sublingual glands. Parotid gland enlargement is more prevalent in children (50%) than in adults (2.2%), (Naidoo & Chikte, 2004; Hamza et al., 2006). PGE in children is an important clinical sign as it has been linked to favourable prognosis (Katz et al., 1993; Itescu et al. 1990). The swelling may be firm mostly non-tender on palpation. Sometimes fluctuant swellings can be found depending on the size and stage of cystic formation. On initiation of ARVs in a treatment-naive individual, there is a regression of the enlargement in most of the patients (Mandel et al., 1998, Schiødt et al., 1992; Kungoane, 2010)

Xerostomia and reduced salivary flow have been associated with HIV since its early stages of discovery (Yeh et al., 1988; Mandel et al., 1992). In a longitudinal study Atkinson et al. (1989b) have observed that in HIV positive individuals, the function of the submandibular gland is affected earlier in the disease and the parotid is affected later. Navazesh et al.(2000,2003), in a longitudinal studies of a large cohort of HIV+ and “at risk” women, have associated being HIV positive , low CD4 counts , high viral loads and HAART to reduced salivary flows and increased xerostomic complaints. Lopez-Verdin et al. (2013) could not associate a low CD4 or increased viral load to an increase in xerostomia or to a reduction in salivary flow rate. Schiødt et al., (1992) proposed that the reduction in salivary flow is “likely to be a function of the degree of inflammatory infiltrate in the gland but not associated with degree of immune deficiency”. Nittayananta & Chungpanich (1997) reported a prevalence of 63% for xerostomia among a group of Thai people with AIDS. Australian and Brazilian studies

by Jeganathan et al., (2012) and Busato et al., (2013) have reported 38% & 40% xerostomia respectively among HIV positive people.

Reduced salivary flow in HIV positive patients has been documented in so many studies that its association to the disease is globally accepted (Yeh et al., 1988; Mandel et al., 1992; Navazesh et al., 2000, 2003, 2009; Nittayananta 2010a). Lopez-Verdin (2013) has reported taste disturbances in those patients that are not on ARVs; ruling out a drug side-effect as a reason. Salivary compositional changes are also present due to HIV infection. An increase in the concentration of lactoferrins, lysozymes, IgA, histatins, chlorides, sodium, mucins and peroxidases was observed (Mandel et al., 1992; Fox, 1992; Atkinson et al., 1990, Yeh et al., 1988; Jainkittivong et al., 2009; Lin et al., 2001 & 2003). Although there is an increase in the concentration of many protective components, due to the reduction in salivary flow rate, the secretory rate of these in the saliva is reduced, thereby, reducing the antimicrobial, antifungal and antioxidant properties in saliva (Lin et al., 2003). Lin et al. (2001) also reported an increase in candidial carriage in the saliva of HIV positive subjects. These compositional changes along with reduced salivary flow, which in itself is a risk for candidial infection (Torres et al., 2002), would account for the increased prevalence of oral fungal infections in these immune-compromised individuals.

2.4.3 Effect of HAART on HIV Associated Salivary Gland Disease

Xerostomia and Parotid Gland Enlargement have often been associated with HAART (Ceballos-Salobrena et al., 2000; Greenspan et al., 2001; Lopez-Verdin et al., 2013; Navazesh et al., 2003). In a longitudinal cohort study of 1000 HIV positive subjects by Ramirez-Amador et al. (2003), xerostomia was found to decrease in prevalence as they progressed from being treatment-naive to monotherapy to HAART. HIV-SGD (possibly indicating PGE) did not show a significant reduction.

In 2010, two Thai studies were published by Nittayananta et al. (2010a & 2010b); one on the oral health status of HIV+ individuals before the HAART era and the other on the long term effects of HAART. HIV positivity was a significant factor in increased xerostomia and reduced salivary flow. Long term (> 3 yrs) HAART was more

associated with increased hyposalivation than short term (<3 yrs) HAART. Xerostomia appears to reduce with HAART initiation but slowly increases with increased duration of HAART. Navazesh et al., (2003) and Lopez-Verdin et al. (2013) also reported reduced salivary flow, with increasing number of years on HAART.

Navazesh et al. (2009) and Silverberg et al. (2004) have reported from a large cohort study of HIV positive women that the occurrence of xerostomia and reduced salivary flow is least in the “stable HAART” group, higher in “HAART naive” participants and was highest among those who switched or discontinued HAART.

Most of the earlier studies on the effect of HAART on salivary glands were of HAART with Protease Inhibitor-regimens. Recently, Johar et al. (2011), and Pavithra et al. (2013), have shown no change and even a slight increase in salivary flow rates in those undergoing HAART. In the latter study, all the patients were on 2 NRTIs + 1 NNRTI. Individuals on HAART appear to show improvement in the taste disturbances associated with some HIV patients (Lopez-Verdin et al., 2013). In their study, Lin et al. (2006) reported that although there is a marked increase in candidial carriage in the saliva of HIV positive subjects, there is a slight reduction in candidial carriage among those on HAART; also noted by Navazesh et al. (2005). Uric acid, which is an antioxidant, appears to be greater in concentration in the HAART group when compared to those not on HAART (Lin et al., 2006).

Saliva contains secretory leukocyte protease inhibitors (SLPI) that have an anti-HIV activity accounting for its low infectivity through saliva (Lin et al., 2004). Lin et al. (2004) reported that although the secretory rate of SLPI is reduced in HIV, there is a slight increase in the rate of SLPI under HAART. This further reduces the chance of salivary transmission of HIV from those under treatment.

Although the rate of secretion of many salivary components is reduced in HIV positive individuals, HAART does not appear to make it worse (Lin et al., 2006; Jaikittivong et al., 2009).

Navazesh et al. (2005) have shown in their study that HAART is a significantly independent risk factor in the increased occurrence of various bacterial pathogens in

the saliva of HIV positive subjects, suggesting that “HAART promotes an increasing pathogenic salivary microbiota, at least temporarily”.

Since the symptoms of salivary gland dysfunction are similar, it is difficult to discern how much of it is the course of the disease, how much is due to adverse effects from HAART usage and how much is part of IRIS due to host response from a recovering immune system when under treatment.



2.5. IMPACT OF SALIVARY GLAND DISEASE

The old adage that “You don’t miss something until it is gone”, is true in the case of saliva. Normal salivary function, and the impact that it has on our quality of our life is mostly taken for granted until there is salivary gland dysfunction. As discussed earlier, the prevalence of xerostomia and hyposalivation in the general population varies but is more among the aged population. With the increasing spread of diseases like HIV and Hepatitis C, salivary gland dysfunction is going to be encountered more and more by the clinician.

Although a consistent relationship between xerostomia and reduced salivary flow has often been hard to establish, Rostron et al. (2002), have reported decreased health related quality of life (QOL), in xerostomic patients who did not show any reduction in salivary flow. Busato et al. (2013), and Jeganathan et al. (2012), have also reported on reduced QOL in HIV patients who complain of xerostomia. Xerostomia has also been associated with other oral sensory complaints like burning mouth and taste alteration (Lopez-Verdin et al., 2013).

In patients with reduced salivary flow, the saliva is often thick, stringy or of a foamy consistency. The oral mucosa loses its healthy glisten and can become dry and is more prone to trauma and infection (Atkinson & Fox, 1992; Torres et. al., 2002). The tongue may appear dry, cracked or fissured with loss of papillae (Navazesh et al., 1992a; Sreebny & Schwartz, 1997).

Recurrent candidial infections (Torres et. al., 2002; Mandel et al., 1992; Navazesh et. al., 2003), gingivitis and angular cheilitis are also a common suffering. These individuals often have problems with chewing, swallowing, tasting and speaking (Cassolato & Turnbull, 2003). Difficulties in speech and mastication can cause some people to avoid social engagements. Some people who suffer from chronic xerostomia may be at an increased risk of depression (Stevenson et. al., 2004). Due to difficulty encountered in eating foods that are dry, spicy or acidic, dietary habits are often changed. Oral health status influences food choices, and thus, the intake of key nutrients (Rhodus, 1988).

Patients who have “Dry mouth” often experience sleep disturbances due to the need to quench their thirst (Cassolato & Turnbull, 2003). Generally, salivary flow is lowest during the night (Dawes, 1972), and the discomfort is worsened in the presence of salivary gland dysfunction. This is compounded further in mouth breathers (Narhi, 1994). Denture wearers often have trouble with denture retention and recurrent candidial infections. Reduced salivary flow impairs salivary defence mechanisms (Lin et al. 2003), and reduces lavarge, pH and buffering action. All this makes these individuals prone to dental caries, especially at non-susceptible sites like cervical and incisal margins (Percival et. al., 1994; Sales-Perez et. al., 2012). Children who are HIV positive should be considered as high risk for caries due to reduced salivary flow and intake of chronic syrupy medication (Naidoo & Chikte, 2004). Navazesh et al., (2010) have associated low salivary flow to increased HIV-1 shedding in saliva, increasing the risk of salivary transmission. Facial asymmetry and disfiguration caused by varying degrees of PGE is also a cause for concern among patients affected by HIV-SGD.



2.6. TREATMENT OPTIONS

A good medical history and thorough oral examination is essential before making a diagnosis of salivary gland dysfunction. A step by step management (Table – 2.7) should include:

1. Alleviating symptoms
2. Treatment of oral conditions
3. Taking preventive measures
4. Improving salivary gland function
5. Managing underlying conditions

Table 2.7- Treatment of Xerostomia, Hyposalivation & Related Oral Complications

Management of symptoms	Diet and habit modifications Frequent and regular sips of water Avoidance of dry, hard, sticky, acidic foods Avoidance of excess caffeine and alcohol Salivary substitutes and lubricants Artificial saliva, Rinses, Gels, Sprays, Toothpastes Use of bedside humidifier during sleeping hours
Preventive measures	Increased frequency of oral/dental evaluation Topical fluoride application, Varnish (0.5% NaF) Daily use of fluoridated dentifrice, Topical: over-the-counter (0.05% NaF); prescription (1.0% NaF, 0.4% SnF)
Treatment of oral conditions	Dental caries Restorative therapy, topical fluoride Oral candidiasis <i>Chlorhexidine (CHX) 0.12%: rinse, swish, and spit 10 ml twice daily</i> <i>Nystatin/triamcinolone ointment</i> for angular cheilitis: apply topically 4 times daily <i>Clotrimazole troches: 10 mg dissolved orally 4–5 times daily for 10 days</i> <i>Systemic therapy</i> for immunocompromised patients <i>Denture antifungal treatment: soaking of denture for 30 min daily in CHX or 1% sodium hypochlorite</i> Bacterial infections

	Systemic antibiotics for 7–10 days Ill or poor fitting prostheses Denture adjustment; Hard and soft relines; Use of denture adhesives; Implant-borne prostheses
Increase salivary flow	Sugar-free, xylitol-containing mints, candies, and gum Sialogogues: Pilocarpine: 5–10 mg orally 3 times daily Cevimeline: 30 mg orally 3 times daily Acupuncture
Manage underlying systemic conditions	Multidisciplinary management with other health-care providers.

Adapted from- Napeñas, J.J., Brennan, M.T. & Fox, P.C. 2009, "Diagnosis and treatment of xerostomia (dry mouth)", Odontology, vol. 97, no. 2, pp. 76-83.

2.6.1 Alleviating symptoms

The discomfort of having a dry mouth can be alleviated to some extent by using salivary substitutes and lubricants that can provide mucosal wetting. Artificial saliva solutions, rinses, gels and sprays are available. Most of them contain carboxymethylcellulose or mucins or mucopolysaccharides; alone or in various combinations. In addition, sipping water/fluids throughout the day can help. Dietary adjustments can be recommended to avoid foods that cause irritation or that are difficult to chew or swallow. A balanced diet should be planned so that nutritional elements are not missed out. Consuming food accompanied by frequent sips of water can help. Avoidance of excess caffeine, alcohol and smoking can be advised. The use of an indoor humidifier, especially at night, can alleviate some of the discomfort from dry mouth, dry eye, dry nose and dry skin.

2.6.2 Treatment of oral conditions

Dental caries is a major problem in patients with reduced salivary flow. Apart from removal of caries and restoration of teeth, the clinician must ensure that the margins of restorations are accessible for cleaning and integrity checks. Fluoride-releasing materials like glass ionomers and compomers are the materials of choice. Oral thrush, angular cheilitis and gingivitis can be treated with antifungals and antibiotics. In the immunocompromised patient, systemic antifungals may be needed, depending on immune capacity. Topically, Nystatin cream, amphotericin B lozenges may be used.

Dentures should be treated by soaking in chlorhexidine or 1% Sodium hypochlorite solution. Antifungal cream can also be used on the denture. Chlorhexidine mouth wash can be used twice a day to treat gingival inflammations and ulcers. Use of triamcinolone based ointments for oral ulcers are useful.

In the case of Parotid gland enlargement (PGE), due to lymphocyte infiltrate, initiation of ART usually shows regression (Schiodt et. al., 1992; Hamza et. al., 2006; Mandel & Surattanont, 2002). Sometimes, swellings arise as part of IRIS while on HAART. These eventually regress. Syebele & Butow (2011) reported a 100% reduction in PGE due to Benign Lymphoepithelial Cysts (BLEC) with the initiation of HAART. No IRIS was reported in this study of 10 patients. Surgical treatment to restore facial symmetry has been recommended by some authors (Schiodt et. al., 1992). Due to the close association of the facial nerve to the parotid gland, nerve palsy is of concern during parotidectomy procedures.

Repeated fine-needle aspiration (FNA) has shown good results (Jacob et al., 2013). FNA is also useful for confirmation of cysts and ruling out malignant changes. Lymphoepithelial cysts have been known to develop non-Hodgkin's Lymphomas (B-cell type), making it a pre malignant lesion (Itescu et. al., 1992; Mandel et. al., 1998; Rivera et al., 2003). Hence, long-term monitoring is necessary. Low-dose radiotherapy has been used successfully by some researchers (Beitler et al., 1995). Due to the long term consequences, radiotherapy in benign lesions is to be questioned. Sclerotherapy by intraoral injection of BLEC with doxycycline or alcohol has also been done (Jacob et. al., 2013).

2.6.3 Taking preventive measures

Frequent oral examination is mandatory, especially until caries control is achieved. In-office application of acidulated phosphofluoride (APF) gel in a tray, 4 times a year, is recommended. Meticulous oral hygiene and a low sugar diet are advised. Neutral PH fluoride daily rinses should be part of routine oral hygiene. Use of alcohol-based mouth washes should be avoided as they can further dry out the mucosa. Increased use of topical and or systemic fluorides can increase fluoride levels in the saliva to therapeutic levels (Duckworth et al, 1987). This can assist in remineralisation of enamel.

If radiotherapy is planned for lymphoepithelial cysts or tumors, a pre-radiation oral treatment plan should include removal or minimising of all potential risks for dental abscess. A discussion with the radiologist can be made on the possibility of sparing one or more salivary glands. Radiation protection stents can be fabricated to protect the glands of the ipsilateral side. Surgical transfer of submandibular glands, to outside the radiation field, before radiotherapy, is an option being recommended (Jha et al., 2003).

2.6.4 Improving salivary gland function

Mastication is a strong stimulant for salivation and it also has a long term beneficial effect on salivary secretions. Jenkins & Edgar (1989) showed that chewing sugar-free gum 4 times daily for 8 weeks increased unstimulated whole saliva. Research by Steinberg et al. (1992) has shown that using gum with xylitol or sorbitol improves oral health by reducing plaque accumulation, gingival inflammation and assists in enamel remineralisation.

Drug therapy with Pilocarpine and Cevimeline, in patients with enough functioning exocrine tissue and in whom it is not medically contra-indicated, has shown appreciable increase in salivary flow. Pilocarpine taken orally as 5 mg, 4 times daily, half an hour before a meal, and at bedtime has been recommended (Napeñas et al., 2009 ; Ship, 2002). Cevimeline is taken as 30mg tablets orally, 3 times daily. Side effects of these drugs include: increased perspiration and increased bowel and bladder activity (Von Bultzingslowen et. al., 2007).

Some acupuncture techniques have also been shown to increase salivary flow (Napeñas et al., 2009).

2.6.5 Managing underlying conditions

If the salivary gland hypofunction is due to medication, the possibility of changing the type of medication to one with lesser anti-cholinergic effects or changing the time of administration can be discussed with the physician. In the case of hyposalivation caused due to the effect of HAART, changing the drug regimen is not always a feasible option. Fortunately, the PI based regimen, which has been the one repeatedly implicated in salivary gland hypofunction, is no longer included in the first- line

treatment of newly diagnosed HIV positive patients (Nat Dept of Health, 2013). In the current guidelines, the PI based regimen is reserved for those patients failing on the first line of treatment, thereby, reducing the number of total patients on protease inhibitors currently. In these patients, when they have been diagnosed with xerostomia or hyposalivation, other preventive, therapeutic and supportive measures, mentioned previously, may be used.



2.7 CONCLUSION

In summary, the presence of salivary gland sub-function in some HIV positive patients, is determined in part by the effect of the viral antigens and leukocytic infiltration present in the glandular tissue, in part by the CD4 count and extent of immune suppression and in part by the effect of HAART directly or indirectly by an inflammatory response from a recouping immune system.

Despite the cause, diagnosis of xerostomia and hyposalivation and its alleviation should play an important part in the overall management of the HIV positive patient.

Knowledge about the prevalence, co-relations and possible confounders of xerostomia and hyposalivation is the first step in making the clinician aware of the need for exploring the presence of these conditions while managing the oral health of HIV positive patients. This study aims to contribute towards this.



CHAPTER - 3

RESEARCH DESIGN AND METHODOLOGY

3.1 AIM

To compare the prevalence of xerostomia and hyposalivation, in HIV positive patients on HAART, HIV positive patients not on HAART and HIV negative patients, attending Empilweni Gompo community health centre (EGCHC) in East London.

3.2 OBJECTIVES

1. To determine the prevalence of Xerostomia, using a questionnaire, in HIV positive patients on established HAART, in HIV patients not on HAART and in a HIV negative group.
2. To determine the prevalence of hyposalivation by measuring whole mouth salivary flow rates (resting and stimulated) in the above mentioned three groups.
3. To compare xerostomia responses and salivary flow rates across the 3 groups.
4. To co-relate reduced salivary flow with low CD4 counts in the HIV positive groups.

3.3 NULL HYPOTHESIS

“There is no difference in the prevalence of xerostomia and hyposalivation in HIV positive patients on HAART, HIV positive patients not on HAART and HIV negative patients”

3.4 METHODOLOGY

3.4.1 Study design

The study design was a cross sectional analytical study.

3.4.2 Site

The study was conducted at the Empilweni Gompo Community Health Centre, a primary health care facilities in the city of East London, Eastern Cape. East London is situated in the Eastern Cape. It has a population of about 267,000 people. Empilweni Gompo CHC is one of the two community health care centres in East London and falls under the Buffalo City Health district. It has a catchment population of about 57,900 from the surrounding areas.

About 12,000 patients are seen at the centre on a monthly basis as outpatients. There are about 1300 patients currently on the ARV programme here.

3.4.3 Sample/Population

Patients attending the HIV Counselling and Testing centre (HCT) and the ARV clinic at EGCHC in East London city of age group- 18 to 55 years were invited to partake in the study. Willing patients were included in the study on a daily basis until the set number required was fulfilled in each group. Patients were recruited from those attending the HCT (HIV Counselling and testing) section and the ARV clinic. Sample size was 150, with 50 individuals in each group.

3.4.4 Inclusion Criteria

- ❖ Patients 18-55 years of age.
- ❖ Subjects selected were divided into three sub groups.
 - Group 1- Patients attending the HIV counselling and testing centre (HCT) who had tested negative were invited.

- Group 2 – Included in this group were patients attending the HIV counselling and testing centre (HCT) who had tested positive and are HAART naive. HIV positive patients attending the centre, who had a CD4 count above 350 cells/mm³, and thus, not eligible for HAART, were also allocated in this group.
- Group 3- This group consisted of HIV positive patients, from the ARV clinic, who had been on HAART for 2 years or longer.

3.4.5 Exclusion Criteria

- ❖ Any patient who was on any other (other than HAART) acute or chronic meds that have a side effect of xerostomia.
- ❖ Any patient who had been diagnosed with any auto immune salivary gland disease.
- ❖ Any patient who had received any head and neck radiation.
- ❖ Completely edentulous patients who were not using any dentures.
- ❖ Female patients who were pregnant.
- ❖ Any patient who was diagnosed with diabetes mellitus.

3.4.6 Data Collection and Procedure

All saliva collection was done between 9:00 am and 3:00 pm and the subjects were asked to refrain from consuming food or fluids for 90 minutes before the procedure.

The volunteers were given a patient information sheet (Appendix 1-3) to take home and the whole process was explained to them in English, Afrikaans or Xhosa, with the help of an interpreter where necessary. A written consent (Appendix 4-6) in the language of their choice was then obtained. The subjects were asked to rinse out their mouth with tap water before saliva collection. A data collection form was used for recording details from each patient (Appendix -7).

Sterile, graduated plastic tubes, with a funnel placed on top, were used for collecting saliva (see picture 3.1& 3.2).

The subject was asked to hold the tube and sit forward with the head bent down (see picture- 3.3). A countdown timer was used to mark time elapsed.

Picture 3.1- Tube with funnel



Picture 3.2- Armamentarium for saliva collection

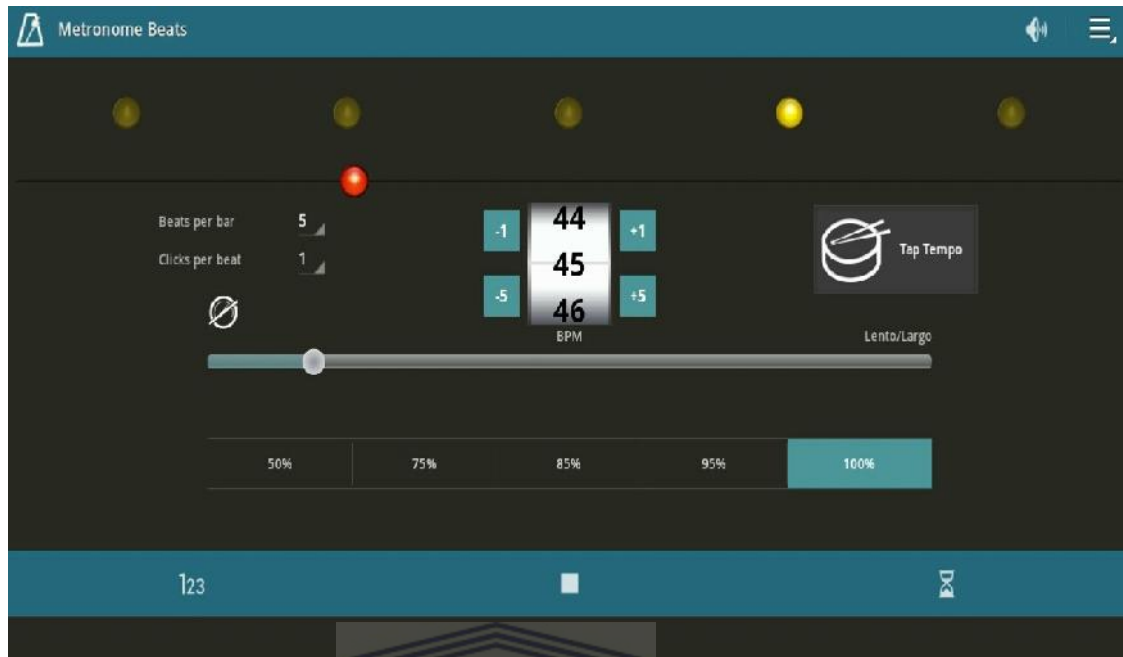


Picture 3.3- Patient positioning during saliva collection



Unstimulated whole-mouth saliva was collected by the “spitting method” into the tube for 3 mins. At the end of three minutes, the tubes were collected and new tubes used for collecting chewing stimulated saliva. A 2cm piece of sterile rubber (see picture -2) was used for chewing and saliva collected by the same method. The subjects were asked to chew for one minute and spit only the saliva into the tube. Keeping the rubber piece in the mouth, the process was repeated two more times and the rubber discarded separately. Chewing was regulated, to 45 strokes per minute, with the use of a metronome (see picture-3.4). The tubes were capped and marked with the subject number and a suffix “a” for resting and “b” for chewing stimulated samples.

Picture 3.4- Android Application for a Metronome, set at 45 beats per minute



A xerostomia questionnaire (Appendix-8) was filled with the help of Xhosa or Afrikaans- speaking translator when needed. This questionnaire is based on the four questions found by Sreebny & Valdini (1988) to have a high specificity and predictive value.

All saliva samples collected were weighed on a calibrated scale at the National Health Laboratory Services, East London (NHLS). Since the specific gravity of saliva is one, 1 gram is considered equivalent to 1 ml. The weight of the empty tube was predetermined and this was subtracted from the total weight. Each measurement was then divided by 3 to obtain the flow rate per minute. This was recorded on the subject's saliva record (Appendix-9).

An unstimulated salivary flow rate measure of 0.16 ml/min is considered as abnormal and a flow rate of 0.7 ml/min of stimulated saliva is considered as abnormal (Navazesh et al., 2003). A positive response to any of the xerostomia questions is considered as indicative of presence of xerostomia.

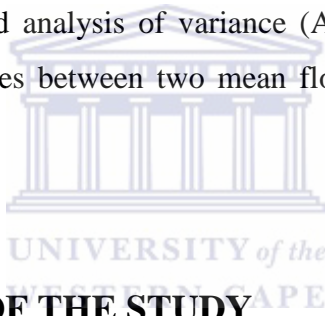
Since all the data was collected by a single examiner, inter-examiner variability was avoided.

3.5 DATA ANALYSIS

Data was captured on the forms and tabulated on Microsoft Excel. Data Analysis was done with the statistical software “R” version 2.15.0 (2012-03-30) (Copyright © 2012, The R Foundation for Statistical Computing). The data collected was subjected to descriptive analysis and prevalences were calculated. The significance of the differences of prevalences were calculated by Chi-squared test and, where applicable, Fischer’s exact test. Statistical significance was set at p- value <0.05.

The influence of independent variables such as sex, age etc. on xerostomia prevalence was examined using logistic regression. Low resting and chewing stimulated salivary flow rates were treated similarly.

The group differences in mean flow rates were examined by least squared linear regression and associated analysis of variance (ANOVA). The student’s t- test was used whenever differences between two mean flow rates needed to be analysed for significance.



3.6 LIMITATION OF THE STUDY

3.6.1 Data Collection

Due to departmental (Dept of Health) policy, stable patients on HAART have their CD4 counts taken only once a year. Hence, the latest available CD4 counts had to be used; some of these were older than a year and were not included. Since Viral Loads are routinely checked only at the time of HAART initiation and treatment follow-ups, viral load measurements were not available for any of the patients in group-2. Viral loads were available only for group-3 and 78% of them had a value lower than the detectable level and only 2% had a count less than 1000 copies/ml. This made it vapid to check the influence of viral loads on any of the outcome measures.

All saliva collection was done between 9:00 am and 3:00 pm. This wider time frame was selected to allow for maximum participation by the patients that attend the facility

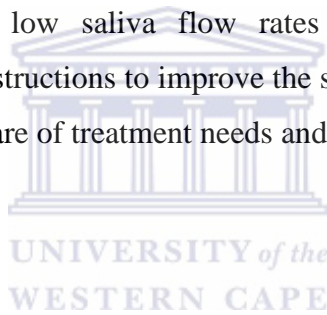
and to allow for the data collector to attend to regular patients in between, since it was not possible to set aside time only for data collection due to work commitments.

3.7 ETHICAL CONSIDERATION

Ethical approval was obtained from the Senate Research Committee, University of Western Cape (Appendix-10). Permission to conduct the study was obtained from the Department of Health, Eastern Cape (Appendix-11) and from the facility manager at EGCHC.

Patient confidentiality was maintained at all times. Subjects were allocated study numbers and names or personal details were not recorded anywhere on the data sheet.

Patients who recorded low saliva flow rates were given onsite oral hygiene instructions as well as instructions to improve the saliva flow. Patients requiring dental treatment were made aware of treatment needs and referred to the dental clinic.



CHAPTER - 4

RESULTS

4.1 DEMOGRAPHIC DETAILS

The 150 participants were allocated into three groups; Group-1 was HIV negative individuals; Group-2 HIV positive individuals who were HAART naive and Group-3 was HIV positive individuals who had been on HAART for more than 2 years. The demographic distribution according to Groups is illustrated in figures 4.1 & 4.2.

Figure 4.1- Age distribution by group

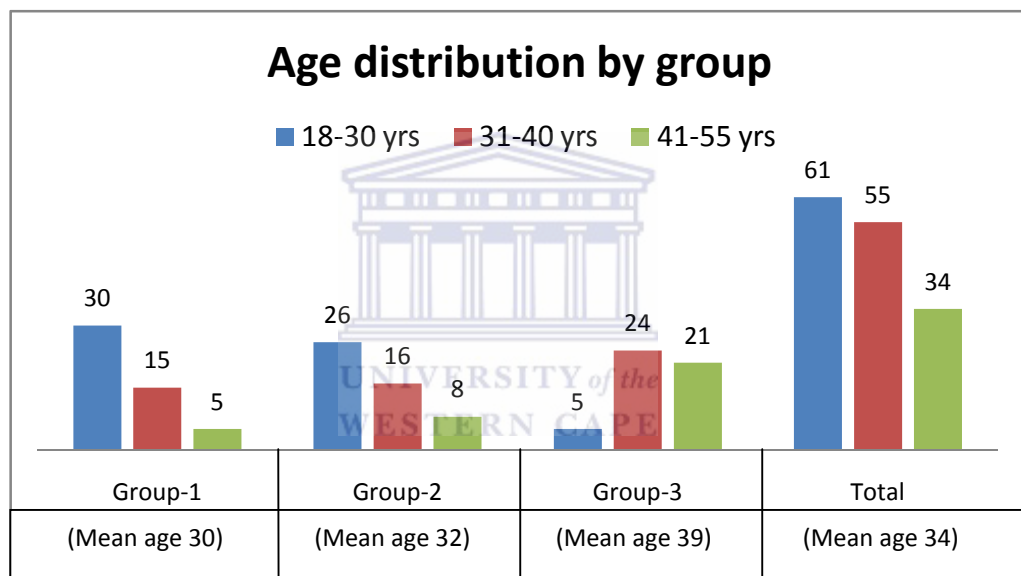
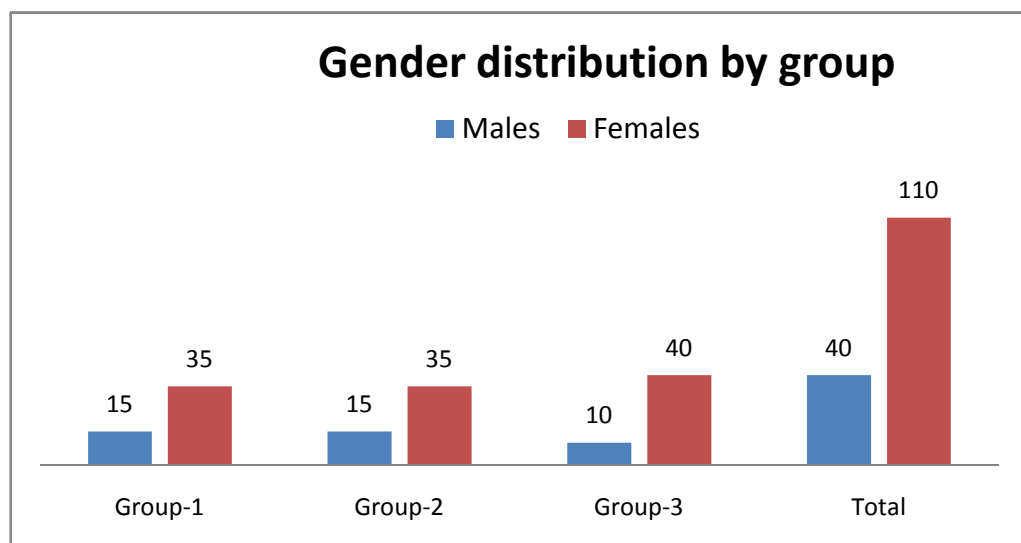


Figure 4.2- Gender distribution by group



Of the 150 patients, 40 (27%) were males and 110 (73%) were females. The average age of male participants was 36 years and of female participants it was 33 years. The average age of the whole study population was 34 years. The total number of smokers were 17 (11%) with 8, 7 & 2 in groups 1, 2 & 3 respectively. Of these only 2 (1%) patients, both in group-1, smoked more than 10 cigarettes per day (see table 4.1)

Table 4.1– Distribution by habit

Variables		GROUP-1 (N=50)	GROUP-2 (N=50)	GROUP-3 (N=50)	Total (N=150)
Smoking (cig/day)	<5	3 (6%)	3 (6%)	1 (2%)	7 (5%)
	6-10	3 (6%)	4 (8%)	1 (2%)	8 (5%)
	>10	2 (4%)	0	0	2 (1%)
	Total	8 (16%)	7 (14%)	2 (4%)	17 (11%)

CD4 counts were recorded for groups 2 and 3. The CD4 counts of five subjects in group-3 were missing or outdated for use. This was taken into consideration during analysis. Group 2 had significantly more patients with CD4 counts < 350 cells/mm³; 62% (n=31) of the patients in group-2 had a low CD4 count < 350 cells/mm³ when compared to 20% (n=9) in group-3. This is expected since the patients in group 3 had been on HAART for more than two years. The mean CD4 count in group-2 was 338.66 (SD 288.26) cells/mm³ and that of group-3 was 577.06 (SD 266.54) cells/mm³.

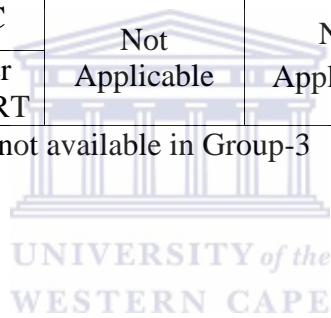
78% of the patients in group-3 had a viral load that was Lower than Detectable Limit (LTDL). Only one patient had a viral load greater than 1000 copies /ml. Viral load measurements were not available for any of the patients in group-2.

In group-2, although the HIV positive patients were not on HAART, 6 (12%) of them were on either Co-trimoxazole or Isoniacid (INH) and 2 of these were on both the drugs. In group -3, 12 (24%) HIV positive patients were on INH or Co-trimoxazole in addition to being on HAART. A complete tabulation of all the clinical data can be seen in table 4.2

Table 4.2- Distribution by clinical data

Variables		GROUP-1 (N=50)	GROUP-2 (N=50)	GROUP-3 (N=50)	Total (N=150)
CD4 Count(cells/mm ³)	350	Not Determined	31 (62%)	9* (20%)	40 (42%)
	>350		19 (38%)	36* (80%)	55 (58%)
Viral Load (copies /ml)	LTDL	Not Applicable	Not Determined	39 (78%)	N/A
	1000			10 (20%)	N/A
	>1000			1 (2%)	N/A
Taking Co-trimox &/ INH	Yes	Not Applicable	6 (12%)	12 (24%)	18 (18%)
	No		44 (88%)	38 (76%)	82 (82%)
HAART regimen	FDC	Not Applicable	Not Applicable	34 (68%)	N/A
	Other HAART			16 (32%)	N/A

* Five CD4 counts were not available in Group-3



4.2 PREVALENCE OF XEROSTOMIA

The overall prevalence of xerostomia was 50% in the total study population. Group -2 showed the highest prevalence with 33 (66%) out of 50 subjects responding affirmative to at least one of the xerostomia questions in the questionnaire. 25 (50%) subjects and 17 (34%) subjects, in groups 1 and 3 respectively, responded similarly.

The differences in prevalences between the groups were examined using the Chi-Squared test. The hypothesis of homogeneous prevalences was rejected at level 0.006. The difference between group-2 and group-3 was found to be significant at $p=0.002$. Although group-2 had a higher xerostomia prevalence than the HIV negative patients in group-1, the difference between the two prevalences was not significant at $p= 0.156$.

Table 4.3- Prevalence of xerostomia among the groups

Outcome	Group-1	Group- 2	Group-3	P value
Xerostomia	50%	66%	34%	0.006*

*Significant at $p<0.05$

Figure 4.3- Prevalence of xerostomia by group

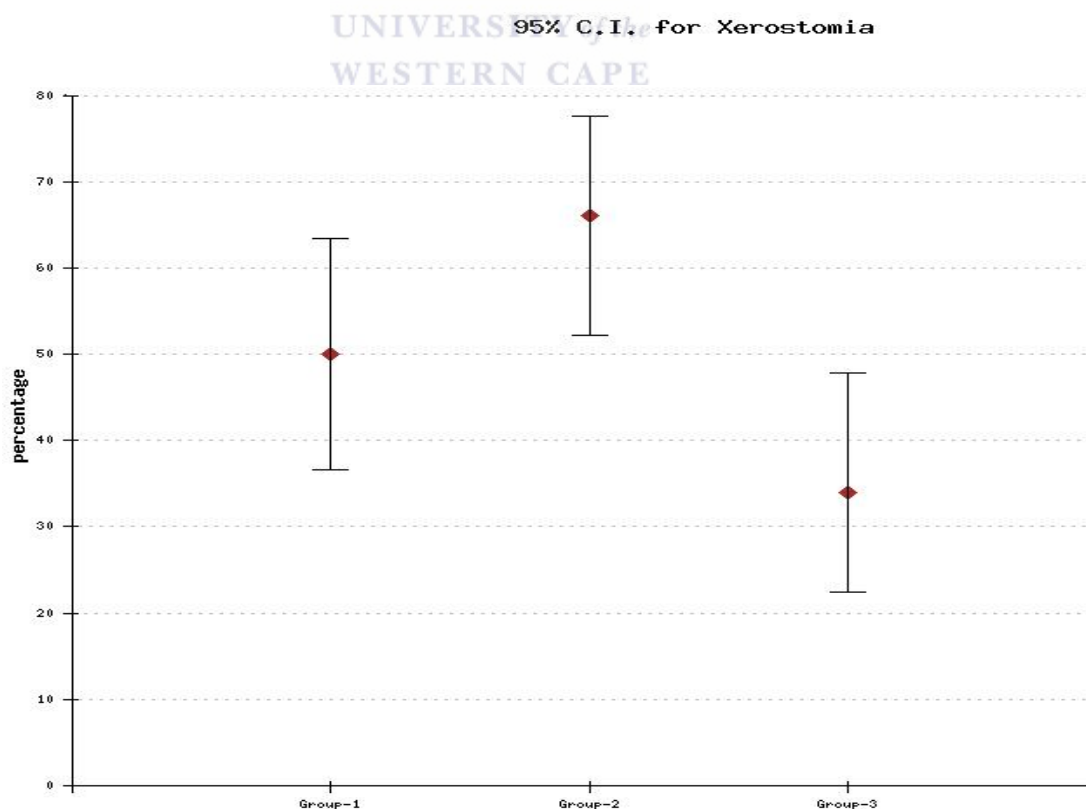


Table 4.4- Distribution of xerostomia scores according to number of questions

No: of Questions	GROUP-1 (N=50) (%)	GROUP-2 (N=50) (%)	GROUP-3 (N=50) (%)	TOTAL (N=150) (%)
Any Question	25 (50%)	33 (66%)	17 (34%)	75 (50%)
1 Question	11 (22%)	13 (26%)	7 (14%)	31 (21%)
2 Questions	7 (14%)	10 (20%)	5 (1%)	22 (15%)
3 Questions	5 (10%)	6 (12%)	3 (6%)	14 (9%)
4 Questions	2 (4%)	4 (8%)	2 (4%)	8 (5%)

Table 4.4 shows the frequency of positive responses to each of the four xerostomia questions. Question no-3 was the only one that showed a significant difference between the questions and only in group-2 with $p= 0.032$. Table 4.5 shows the distribution of the positive answers between the different questions.

Table 4.5- Distribution of the positive responses to the different xerostomia questions

Xerostomia Questions	GROUP-1 (N=50)	GROUP-2 (N=50)	GROUP-3(N=50)
1) Does your mouth feel dry often?	12 (24%)	18 (36%)	9 (18%)
2) Do you regularly do things to keep your mouth moist?	17 (34%)	19 (38%)	12 (24%)
3) Do you get out of bed at night to drink fluids?	10 (20%)	20 (40%)	10 (20%)
4) Does your mouth usually become dry when you speak?	9 (18%)	10 (20%)	3 (6%)

The influence of covariates on the prevalence of xerostomia was examined by fitting generalized linear models with the dependent variable xerostomia and independent

variables Group, Age, Sex, Smoking, CD4 and use of C0-trimoxazole/ INH (using only Groups 2 and 3). Multiple logistic regression analysis revealed that the significant predictor besides group was age. Age was found to have a significant negative correlation at p=0.002. The distribution of the prevalence according to the covariate influence is tabulated in table 4.6

Table 4.6- Prevalence of xerostomia in the different groups and covariate categories

Co-variates	Group-1 (N=50)		Group-2 (N=50)		Group-3 (N=50)		Total (N=150)	
	(n)	%	(n)	%	(n)	%	(n)	%
AGE (yrs)								
18-30	17 (30)	56%	20 (26)	77%	3 (5)	60%	40 (61)	66%
31-40	6 (15)	40%	8 (16)	50%	10 (24)	42%	24 (55)	44%
41-55	2 (5)	40%	5 (8)	62%	4 (21)	19%	11 (34)	32%
Gender								
M	7 (15)	47%	8 (15)	53%	1 (10)	10%	16 (40)	40%
F	18 (35)	51%	25 (35)	71%	16 (40)	40%	59(110)	54%
CD4(cell/mm ³)								
350	Not Determined		21 (31)	68%	2 (9)*	22%	23(40)*	58%
>350		12 (19)	63%	14 (36)*	39%	26(55)*	47%	
Use of Co-trimox &/ INH								
Yes	Not Applicable		5 (6)	83%	7 (12)	58%	12 (18)	67%
No		28 (44)	64%	10 (38)	26%	38 (82)	46%	

*Five CD4 counts were not available in Group-3

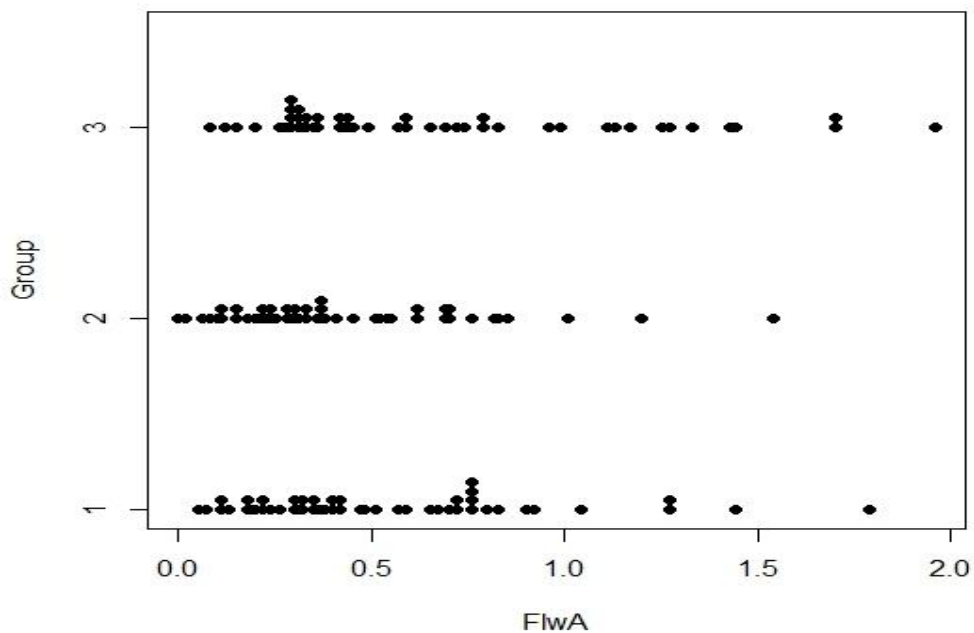
4.3 PREVALENCE OF LESS THAN NORMAL RESTING FLOW RATE

A resting flow rate (Flow Rate A) $0.1\text{ml}/\text{min}$ is considered as below normal (Navazesh et al., 2003; Sreebny & Valadini, 1988). Of the 150 patients only 8 (5%) had a flow rate $\leq 0.1\text{ml}/\text{min}$. Of these 2 were from group -1, 5 from group-2 and 1 from group-3. The prevalence of hyposalivation in resting flow rates among the groups is shown in table- 4.7. There was no statistical significance to the differences in prevalences as revealed by the Fisher's exact test. The hypothesis of equal prevalence for less than normal resting flow rate between groups could not be rejected at level 0.277

Table 4.7- Prevalence of less than normal ($0.1\text{ml}/\text{min}$) resting flow rate

Outcome	Group-1	Group- 2	Group-3	P value
Resting Flow Rate $0.1\text{ml}/\text{min}$ (Flow rate- A)	4%	10%	2%	0.277

Figure 4.4- Dot Plot for Resting Flow Rates (Flow Rate-A) by Group



The influence of independent variables on the prevalence of less than normal salivary resting flow rates were analysed by logistic regression, Chi- squared and Fisher's exact tests. None of them had statistical significance. The prevalence of less than normal resting flow rate as influenced by the different co-variates can be seen in table-4.8.

Table 4.8- Prevalence of less than normal resting flow rate (0.1 ml/min) in the different groups with co-variates.

Co-variates	Group-1 (N=50)		Group-2 (N=50)		Group-3 (N=50)		Total (N=150)	
	(n)	%	(n)	%	(n)	%	(n)	%
AGE (yrs) 18-30	1 (30)	3%	1 (26)	4%	0 (5)	0%	2 (61)	3%
31-40	1 (15)	6%	3 (16)	19%	0 (24)	0%	4 (55)	7%
41-55	0 (5)	0%	1 (8)	13%	1 (21)	5%	2 (34)	6%
UNIVERSITY OF WESTERN CAPE								
Gender M	0 (15)	0%	1 (15)	6%	0 (10)	0%	1 (40)	3%
F	2 (35)	6%	4 (35)	11%	1 (40)	3%	7(110)	6%
UNIVERSITY OF WESTERN CAPE								
CD4 (cell/mm ³) 350	Not Determined		3 (31)	10%	0 (9)*	0%	3 (40)*	8%
>350			2 (19)	11%	1 (36)*	3%	3 (55)*	6%
UNIVERSITY OF WESTERN CAPE								
Use of Co-trimox &/INH Yes	Not Applicable		2 (6)	33%	1 (12)	8%	3 (18)	17%
No			3 (44)	75%	0 (38)	0%	3 (82)	4%

*Five CD4 counts were not available in Group-3

4.4 PREVALENCE OF LESS THAN NORMAL CHEWING STIMULATED FLOW RATE

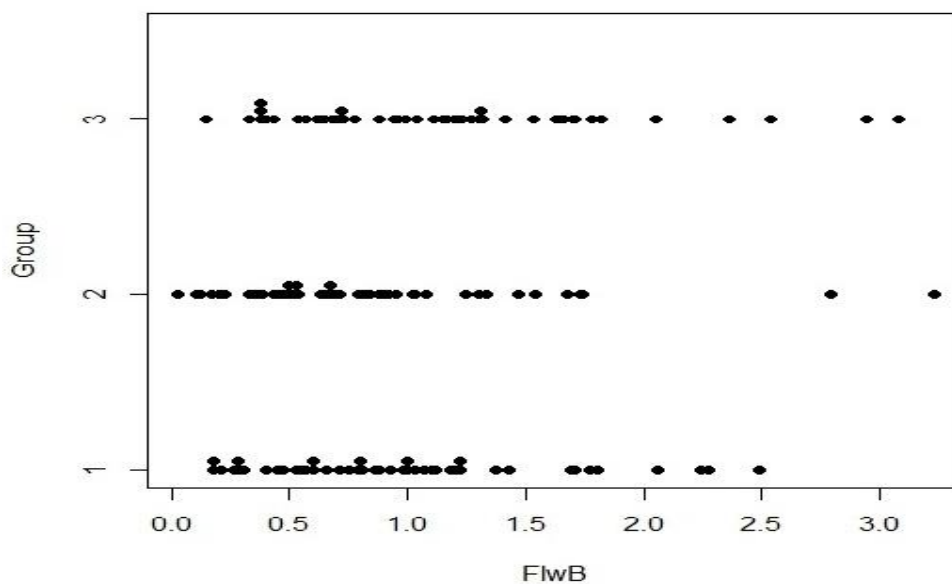
A chewing-stimulated flow rate (Flow Rate B) of 0.7ml/min is considered as below normal (Navazesh et al., 2003). Of the 150 patients 63 (42%) had a flow rate-B that was 0.7ml/min. As with the xerostomia and low resting flow rate trend, group-3, showed the least frequency with 16 (32%) patients having a less than normal chewing-stimulated flow rate. Group-1 had 19(38%) and group 2 had the most with 28 (56%). The prevalence of hyposalivation in chewing stimulated flow rates among the groups is shown in table- 4.9. There was a statistical significance in the differences in prevalence between groups (p=0.041). The significance was even greater when patients not on HAART (group-2) were compared to those receiving HAART (group-3), p=0.013. The hypothesis of equal prevalence for hyposalivation among the groups can be rejected here with regard to less than normal chewing stimulated flow rate.

Table 4.9- Prevalence of less than normal (0.7ml/min) chewing-stimulated flow rate

Outcome	Group-1	Group- 2	Group-3	P value
Chewing-Stimulated Flow rate 0.7ml/m (Flow Rate-B)	38%	56%	32%	0.041*

*Significant at $p < 0.05$

Figure 4.5- Dot Plot for Chewing-stimulated Flow Rate (Flow rate – B) by Group



Upon logistic regression analysis, apart from group, a low CD4 count was found to be of significant ($p=0.015$) influence on the prevalence of a less than normal chewing-stimulated flow rate. A low CD4 count is described here as one that is < 350 cells/mm³.

Table 4.10- Prevalence of less than normal chewing-stimulated flow rate (0.7 ml/min) in the different groups with co-variates

Co-variates	Group-1(N=50)		Group-2(N=50)		Group-3(N=50)		Total(N=150)	
	(n)	%	(n)	%	(n)	%	(n)	%
AGE (yrs)								
18-30	11 (30)	37%	14 (26)	54%	3 (5)	60%	28 (61)	46%
31-40	5 (15)	33%	10 (16)	63%	7 (24)	29%	22 (55)	40%
41-55	3 (5)	60%	4 (8)	50%	6 (21)	29%	13 (34)	38%
Gender								
M	4 (15)	27%	7 (15)	47%	2 (10)	20%	13 (40)	32%
F	15 (35)	43%	21 (35)	60%	14 (40)	35%	50(110)	45%
CD4(cell/mm ³)								
350	Not Determined		20 (31)	65%	4 (9)*	44%	24(40)*	60%
>350			8 (19)	42%	10(36)*	28%	18(55)*	33%
Use of Co-trimox &/INH								
Yes	Not Applicable		4 (6)	67%	5 (12)	42%	9 (18)	50%
No			24 (44)	55%	11 (38)	29%	35(82)	43%

*Five CD4 counts were not available in Group-3

4.5 DIFFERENCE IN MEAN FLOW RATES

The availability of individual flow rates had made it compulsive to consider if there was a significant difference in the mean flow rates between the groups. The mean resting flow rates (Flow Rate –A) for each group was calculated and subjected to a one way Analysis of Variance (ANOVA). Mean chewing-stimulated flow rates (Flow Rate-B) were also treated in the same way.

There was a significant difference in both resting and chewing-stimulated flow rates between the groups as seen in table-4.11. Figures 4.6 & 4.7 illustrate this difference graphically. The difference in the means between Group-2 and Group-3 was more significant with $p=0.003$ for resting flow rate and $p=0.012$ for chewing-stimulated flow rates.

Table 4.11- Outcome measures for salivary flow rates

Salivary Flow Rates		GROUP-1 (N=50)	GROUP-2 (N=50)	GROUP-3 (N=50)	p- value
Resting (Flow Rate A)	Mean (SD) (ml/min)	0.53 (0.37)	0.42 (0.31)	0.66 (0.47)	0.010*
	Median (ml/min)	0.41	0.33	0.45	N/A
	IQR	0.48	0.38	0.62	N/A
Chewing Stimulated (Flow Rate B)	Mean (SD) (ml/min)	0.96 (0.59)	0.81 (0.63)	1.14 (0.67)	0.034*
	Median (ml/min)	0.84	0.67	1.08	N/A
	IQR	0.68	0.57	0.84	N/A

*Significant at $p<0.05$

Figure 4.6- Mean resting flow rate by group

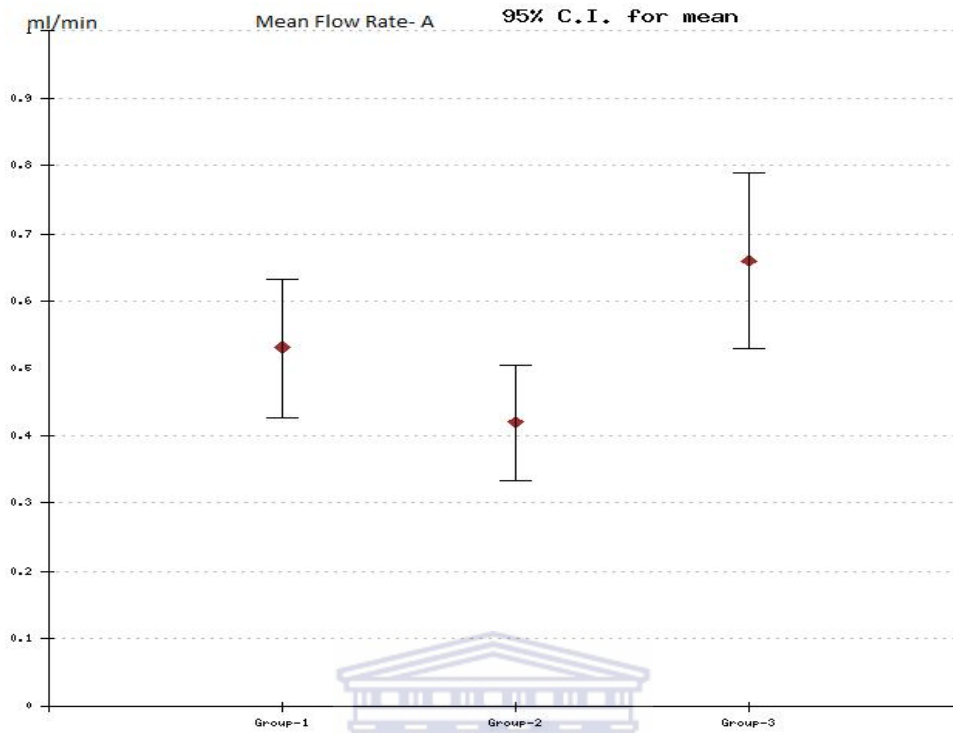
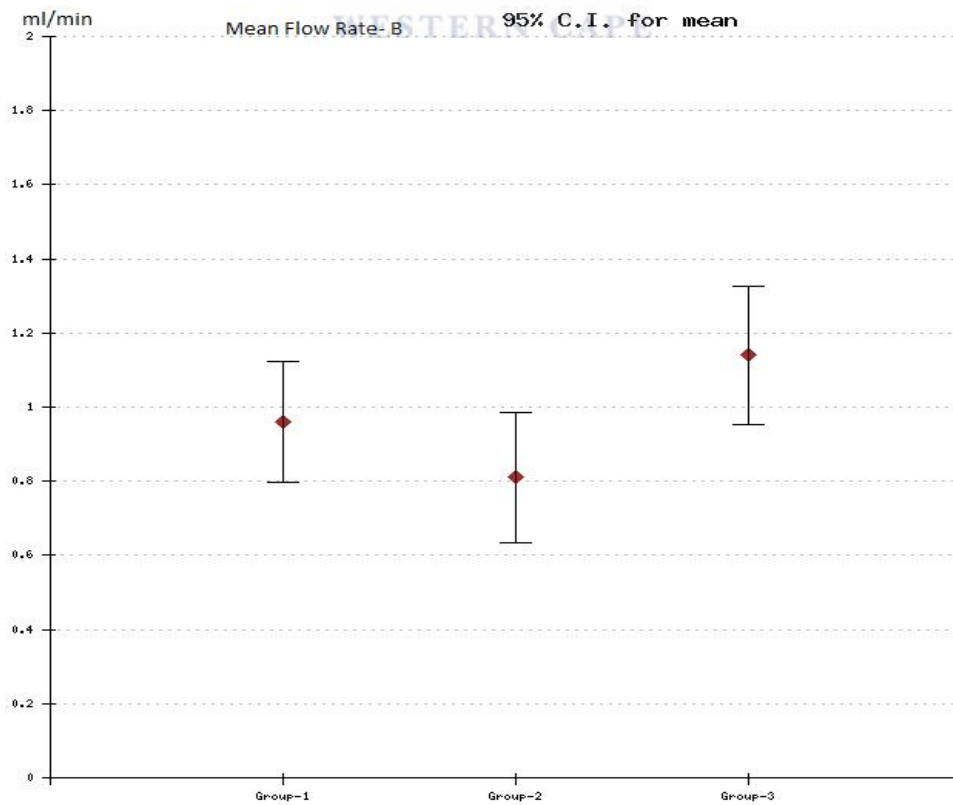


Figure 4.7- Mean chewing-stimulated flow rate by group



4.6 CORRELATING PREVALENCE OF XEROSTOMIA WITH MEAN FLOW RATES

Although, the prevalence of low chewing-stimulated flow rates did show a better association to the xerostomia scores than resting flow rates, there was a general lack of comprehensive association between the xerostomia scores and the prevalence of low salivary flow rates. However, when the mean flow rates of those with a xerostomia score of 1-4 was compared to the mean flow rates of those that had a xerostomia score of 0, both resting and chewing stimulated flow rates showed a statistical significance in their differences. See table 4.12.

Table 4.12- Correlating xerostomia with mean flow rates.

Mean Flow Rates	Xerostomia (n=75) ml/min (sd)	No xerostomia (n=75) ml/min (sd)	p- value (t-test)
Resting (Flow Rate –A)	0.45 (0.33)	0.63 (0.44)	0.005*
Chewing stimulated (Flow Rate –B)	0.80 (0.61)	1.14 (0.62)	0.0009*

*Significant at $p < 0.05$

A negative correlation was observed between the mean resting flow rates and xerostomia scores. The mean resting flow rates reduced as the xerostomia score increased (see table 4.13).

Table 4.13-Correlating xerostomia scores with mean resting flow rates

Xerostomia Scores	Mean Resting flow rate (Flow rate –A) ml/min (sd)
1	0.50 (0.28)
2	0.46 (0.37)
3	0.38 (0.42)
4	0.32 (0.28)

This correlation was not seen for mean chewing-stimulated flow rates.

CHAPTER- 5

DISCUSSION

The HIV prevalence in the Eastern Cape is estimated to be 11.6%. The Buffalo City Metro, under which Empilweni Gompo Community Health Centre falls, has a prevalence of 13.6% (Shisana et al., 2014). The oral health status of the 100 HIV positive patients included in this study is believed to be an appropriate reflection of the HIV infected population in this Metro.

Many studies have linked salivary gland hypofunction and xerostomia to HIV infection (Mandel et al., 1992, Yeh et al., 1988). Some studies have also linked anti retroviral medication to low salivary flow rates and xerostomia (Navazesh et al., 2000; Nittayananta 2010a)

In total, there were 150 patients who partook in this study. 73% of them were females with 70% in Group -2 and 80% in Group -3. This significant disproportion was also seen in the SA national HIV Prevalence, Incidence and Behaviour survey (Shisana et al., 2014) and was attributed possibly to the differences in the health seeking behaviour between the two sexes.

Saliva collection was done from 9:00 am to 3:00 pm so that circadian influences would be minimised (Navazesh et al., 1992a).

The age of 55 was selected as a cut off age in this study to minimize any age related influence on salivary flow rate (Flink et al., 2008). All patients were requested not to drink or eat for 90 minutes before saliva collection to minimise the influence of stimulus from mastication and food ingestion.

5.1 PREVALENCE OF XEROSTOMIA AND LESS THAN NORMAL SALIVARY FLOW RATES

The prevalence of xerostomia ranges widely. It is influenced by age, gender, systemic conditions, medication and even nutritional status. In a large population based study, Flink et al., (2008) reported the prevalence of xerostomia to range from 8% to 39%. In the present study the HIV negative group (Group-1) had a prevalence of 50%. This could be attributed to the fact that these subjects were patients seeking medical treatment for some sort of acute condition or the other. Most of them were suffering from the common cold, viral flu, etc. Although none of them were on any medication, general ill health could have negatively influenced their salivary function temporarily.

The HIV positive group that were not on HAART (Group-2), were a mixed group of those that were acutely ill leading to a recent diagnosis of immune deficiency and those that were HIV positive but healthy enough not to warrant HAART initiation. This group had the highest prevalence for xerostomia at 66%. The average CD4 count of this group was 339cell/mm³.

Group-3 was made up of HIV positive patients who had been on HAART for more than 2 years. Most of the individuals were generally otherwise healthy and attending the health centre for monthly follow up and medication. The mean CD4 count for this group was 577cell/mm³. They had the lowest prevalence for xerostomia at 34%. The overall prevalence of xerostomia was 50% in the total study population.

In a similar study from Thailand by Nittayananta et al., (2010a), they found 32% xerostomia among HIV negative individuals, 61% in HIV positive not on HAART and 39% in HIV positive individuals on HAART for more than 3years. In India, Pavithra et al., (2013) reported a positive response for the question “Often my mouth feels dry” as 36% among HIV positive patients not on HAART and almost 3% for these patients, after 6 months of being on ARVs. In Brazil, 40% xerostomia was seen by Busato et al., (2014) among HIV positive patients. A much lower prevalence was reported from the USA by Silverberg et al., (2004). 6.9% for HIV negative individuals, 16.2% for HIV positive not on HAART. It appears that this study also identifies with the higher prevalences seen in developing countries.

In this study, the difference in prevalences for xerostomia between the groups were found to be significant at $p= 0.006$. The difference between group-2 and group-3 was also found to be significant at $p=0.002$.

Low salivary flow rates among HIV positive patients have been reported in many studies (Yeh et al., 1988; Atkinson et al., 1989b; Lin et al., 2001; Navazesh et al., 2003). In this study 0.1 ml/min and 0.7 ml/min were used as the cut off for establishing less than normal resting and chewing-stimulated flow rates respectively among the subjects. Nittayananta et al., (2010b) reported a 35% prevalence for less than normal resting flow rate and 16% for less than normal chewing-stimulated flow rate among HIV positive individuals who were not on anti-retroviral treatment. A prevalence of 17% and 11% were also seen in the same study for less than normal resting and chewing-stimulated flow rate respectively among HIV negative individuals. Some of the other studies that evaluated prevalence of low flow rates reported the results as odds ratios (Navazesh et al., 2003; 2000). Hence, a comparison could not be made to this study. In this study the prevalence of less than normal resting flow rates were low in all three groups with group-2 showing the highest prevalence at 10%. The difference in prevalences of less than normal chewing-stimulated flow rate among the groups appear to be significant at $p= 0.041$. The HIV positive group not on HAART (group-2) showed the highest prevalence at 56%; followed by the HIV negative group (group-1) at 38% and the HIV positive group on HAART (group-3) showed the least at 32%.

It would appear that, in this study, there is a consistency in the extent to which the different groups are affected by xerostomia and hyposalivation, with group-2 showing the most salivary incapacitation and group-3 showing the least salivary gland dysfunction.

In many of the other studies related to evaluation of salivary flow rates, mean flow rates were used for comparison of salivary hypofunction between groups (Navazesh et al., 2009; Lin et al., 2006; Nittayananta et al., 2010a; Pavithra et al., 2013; Johar et al., 2011). Due to differences in methodology and some of the studies being on mostly PI based HAART regimen, a comparison to this study for mean salivary values was not conclusive.

5.2 OUTCOME MEASURES IN HIV POSITIVE INDIVIDUALS AS INFLUENCED BY HAART

It is clear from comparing the outcomes for the two groups, that, not being on anti-retroviral treatment puts HIV positive patients at risk for salivary gland dysfunction. Table-5.1 lays this picture out clearly. This is in contrast to studies that have shown HAART to be a risk for xerostomia and low salivary flow rates (Navazesh et al., 2003, 2009) and also to those studies that did not show any difference between HIV positive individuals on HAART and not on HAART (Lin et al., 2006; Jankittivong et al., 2009; Johar et al., 2011; Pavithra et al., 2013).

Table 5.1- Prevalences and mean flow rates as influenced by HAART

Outcome	Group -2 (Non HAART)	Group -3 (HAART)	P value	OR (CI)
Xerostomia	66%	34%	0.002*	0.269 (0.107,0.655)
Resting flow rate 0.1ml/m	10%	2%	0.204	0.186(0.004,1.757)
Chewing stimulated flow rate 0.7ml/m	56%	32%	0.026*	0.374(0.151,0.900)
Mean resting flow rate (Flow rate-A) ml/min	0.42 (0.31)	0.66 (0.47)	0.003*	N/A
Mean Chewing-stim Flow rate (Flow rate-B) ml/min	0.81 (0.63)	1.14 (0.67)	0.013*	N/A

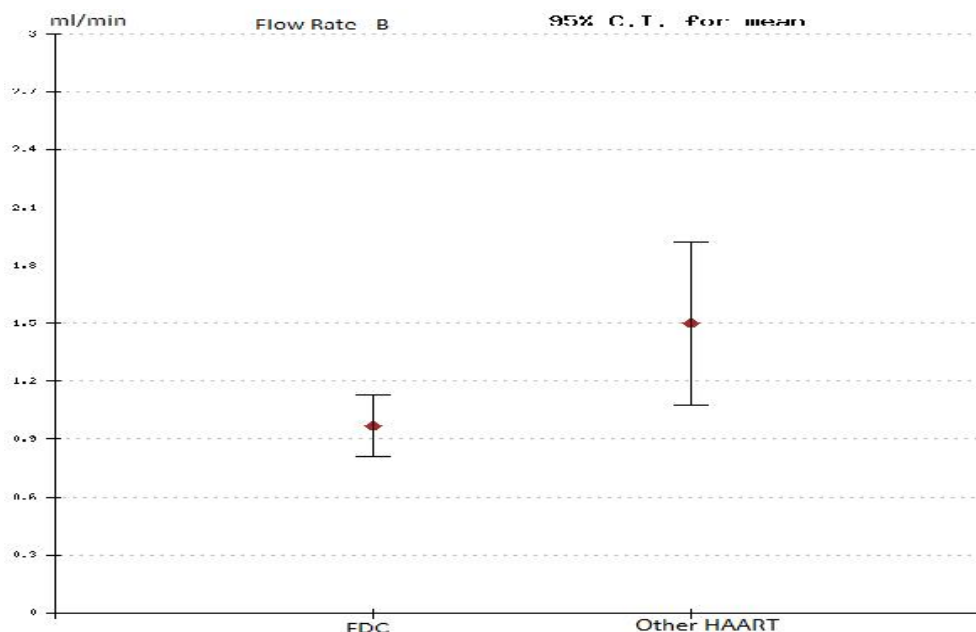
*Significant at $p < 0.05$

There was a decrease in salivary function and increase in xerostomic symptoms among those HIV positive patients that were not on HAART. The difference in mean flow rates between the groups was greater for resting flow rate than for chewing stimulated flow rate. This indicates that the submandibular gland, which is responsible for about 65% of the resting salivary flow, is affected more in this group. Atkinson et al. (1989b) have observed that in HIV positive individuals, the function of the submandibular gland is affected earlier in the disease and the parotid is affected over time.

Among those on long term HAART, the stimulated salivary flow, to which the parotid contributes more than 50%, appears to be more affected than resting salivary flow. This could be due to the changes that HAART brings about upon the parotid gland. Although the exact mechanism is unknown, accinar changes, lipomatous changes and IRIS have all been proposed (Navazesh et al., 2003; Olive et al., 1998; Greenspan et al., 2001)

In a study by Pavithra et al. (2013), the longitudinal effect of the drug regimen 2NRTI's + 1 NNRTI on salivary gland function was evaluated. No significant differences were noted in the flow rates between those on HAART and those not on HAART at baseline. However, there was an increase in flow rate and decrease in xerostomia complaints as the duration of HAART increased up to 6 months. In this study 68% (n=34) were on the Fixed Drug Combination pill (FDC - 2NRTI's + 1NNRTI). When comparing the outcomes of those on this regimen with those on other drug regimens, although not significant, there was an increase in the prevalences of xerostomia and hypofunction. A significant reduction was seen in the mean chewing stimulated flow rate (Flow Rate- B) for those on FDC; p= 0.034; when compared to those on other HAART regimens; illustrated in figure -5.1

Figure 5.1- Mean chewing-stimulated flow rates - FDC Vs Other HAART



This could be due to the fact that apart from 2 of the 34 subjects that were on FDC, the majority of them had changed their HAART regimen in the past 6 months. Silverberg et al., (2004) & Navazesh et al., (2009) had found in their study that patients on stable HAART usage had higher salivary flow rates and lesser xerostomia complaints than those that switched HAART or discontinued treatment in the previous 6 months. There was no significant difference in the xerostomia prevalence between these two groups.



5.3 Influence Of Other Variables

Imitating most of the other studies (Sreebny & Valdini, 1988; Narhi, 1994; Nederfors et al., 1997; Farsi, 2007; Flink et al., 2008), the prevalence of xerostomia was higher for females than males but not statistically significant at 54% Vs 40%. This trend was seen for the prevalence of less than normal resting and chewing stimulated flow rates. When mean flow rates were compared, males had a significantly more chewing stimulated flow rate than females, 1.18 ml/min Vs 0.91ml/min; $p=0.031$. Torres et al., (2002) also found a significant difference in the chewing stimulated flow rates between males and females. The reason for increased volumes of saliva produced by males could be explained by the findings by Scott (1975); males had up to 50% larger submandibular glands than females.

Unlike many other studies that associated increasing age with increasing xerostomia complaints (Flink et al., 2008; Nederfors et al., 1997; Sreebny & Valdini, 1988), there was a negative correlation between the two in this study; $p=0.002$. Although not significant, this negative correlation was also seen for mean chewing stimulated flow rate. This could be because the range of ages included here; 18-55 years; do not significantly affect xerostomia and the influence is coincidentally due to other systemic factors. According to Flink et al. (2008) the prevalence of xerostomia and hyposalivation did not vary much until the age of 50 years for women and 60 years for men.

While low CD4 counts ($<200 \text{ cell/mm}^3$) has been attributed as a significant risk factor for xerostomia and hyposalivation by many authors (Navazesh et al., 2000, 2003, 2009), some did not find this correlation significant (Nittayananta et al., 2010a; Pavithra et al., 2013; Lopez-Verdin et al., 2013). According to Schiødt et al., (1992) the reduction in salivary flow is “likely to be a function of the degree of inflammatory infiltrate in the gland but not associated with degree of immune deficiency”. In this study, CD4 counts ≥ 350 was used as the criteria for low CD4 count since this is the level at which HAART is initiated in the South African Public Health system. Xerostomia prevalence did not show a significant difference between the two CD4 groups. A statistical significance was found only in the prevalence of less than normal chewing stimulated flow rate with $p=0.015$ and not in the prevalence of less than

normal resting flow rate. When mean flow rates were compared, although, the flow rates were reduced in those with CD4 counts $< 350 \text{ cell/mm}^3$, there was no statistical significance. The difference in mean resting flow rates came close to significance at $p= 0.057$.

The mean resting flow rate for those with CD4 counts $< 350 \text{ cell/mm}^3$, when calculated in Group-2 alone, was found to be significant at $p= 0.036$. This, further points to the oral health vulnerability of these HIV positive patients with a low CD4 count in whom HAART is yet to be initiated and during the period while waiting for the therapeutic effect of HAART to improve the CD4 count.



5.4 CORRELATING THE XEROSTOMIA SCORES WITH THE FLOW RATES

Some of the previous studies (Fox et al., 1987; Thomson et al., 1999b; Narhi et al., 1994) did not find a significant correlation between xerostomia complaints and prevalence of low salivary flow rates. Dawes (1987) had reported that xerostomia complaints were seen only after there was a 40% -50% reduction in the actual salivary flow rate. On the other hand, xerostomia complaints can be made in the absence of actual reduction of flow rate due to the systemic and physiological factors affecting the perception of dry mouth. Xerostomia complaints were seen to be increased in subjects who were dehydrated (Ship & Fischer, 1997).

In this study, a significant association could not be made between the prevalence of xerostomia and that of low flow rates as assessed by pre-determined cut-off values;

0.1 ml/min for resting flow rate and 0.7 ml/min for chewing stimulated flow rates. However, using the student's t-test, when the mean flow rate of those that complained of xerostomia was compared to the mean flow rate of those that did not complain of xerostomia, a significance was found both in resting flow rates ($p=0.005$) and chewing stimulated flow rates ($p=0.0009$). See figures 5.2 & 5.3.

Figure 5.2- Mean resting flow rates - Xerostomia Vs No Xerostomia

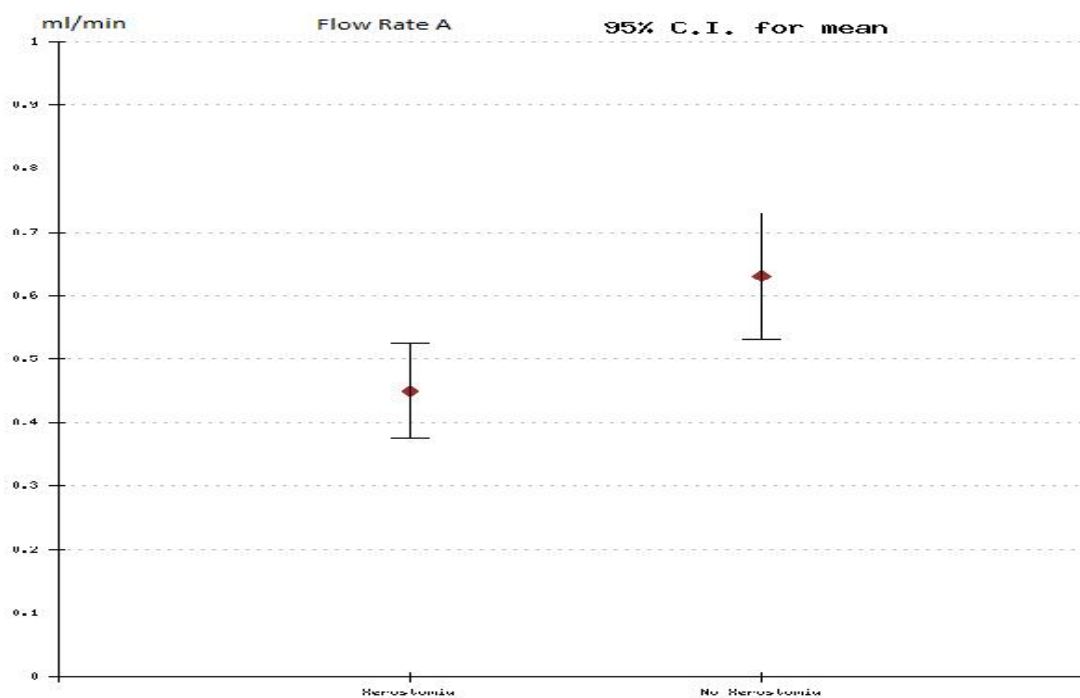
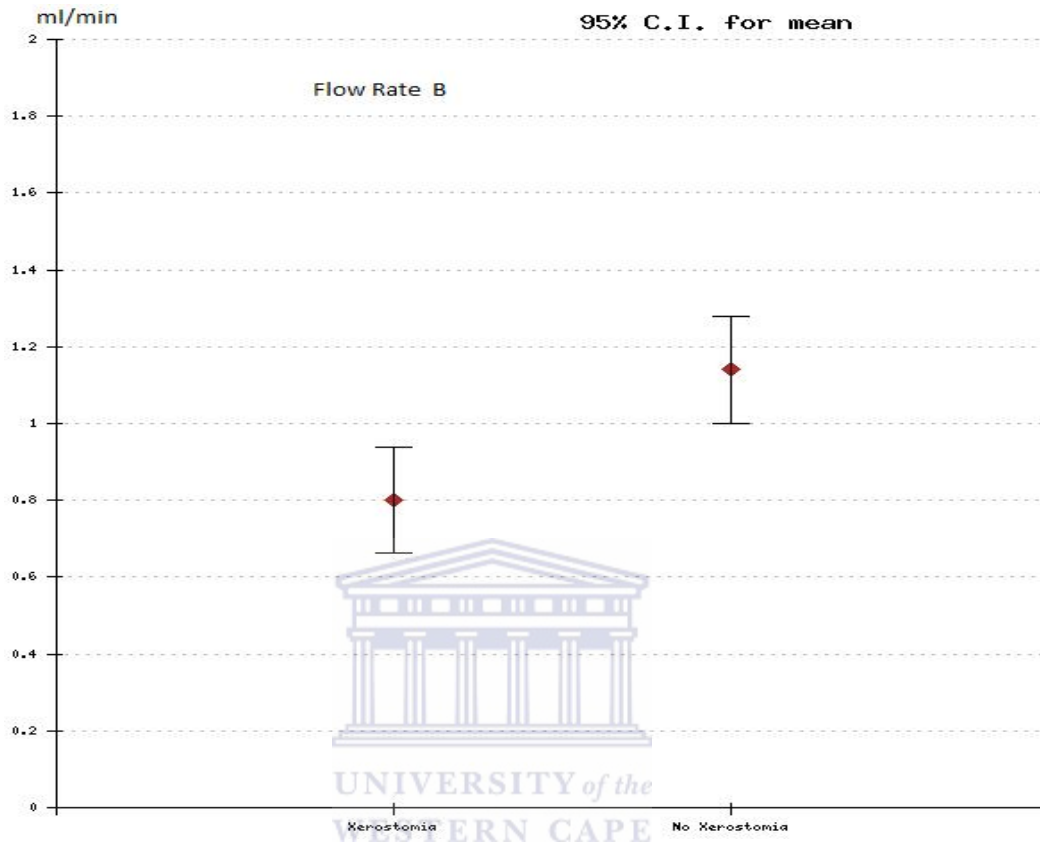


Figure 5.3- Mean chewing-stimulated flow rates - Xerostomia Vs No Xerostomia



This lack of consistency in the association of xerostomia to prevalence of low flow rates and xerostomia to mean flow rates could be due to the possibility that the cut –off values for calculating the prevalence of low flow rates were not compatible with the salivary attributes of this population. Further large scale studies would be necessary to establish criteria appropriate for the South African population.

The negative correlation seen between xerostomia scores and mean resting flow rates is expected since the questions used in this study are more reflective of salivary perceptions during rest rather than during function.

Chapter-6

CONCLUSION

Salivary gland dysfunction is observed more readily in those that are immunocompromised and not yet on HAART. Special attention should be paid in planning an intense prophylactic treatment regimen to prevent and manage the oral conditions that are associated with reduced salivary flow in these individuals.

HAART in itself does not appear to adversely affect xerostomic perceptions or salivary flow rate. The improved immunity that comes from being on Anti Retroviral treatment is beneficial to salivary gland function.

Duration of HAART, change in regimen, type of regimen, all seem to have an effect on salivary flow rate (Silverberg et al., 2004; Lopez- Verdin et al., 2013; Navazesh et al., 2009). Thus, studies on larger samples and of longitudinal design are necessary to explore the possibility of similar findings in the South African context. This would in turn further pin point those vulnerable to salivary hypofunction and its effects, enabling timeous prophylactic actions.

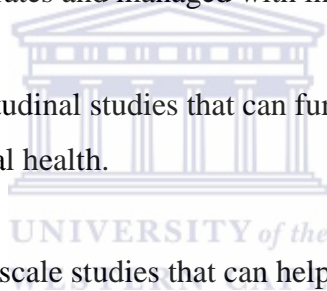
Salivary flow rate is influenced by inter and intra individual variations. Most of the large scale studies that were done to establish criteria for determining the presence of glandular hypofunction were conducted in first-world circumstances and might not be adequate for the South African population. There were instances in this study where a significance was found in mean flow rates but not in prevalences of less than normal flow rates (as dictated by cut-off values) when compared for the same situation. This indicated that mean flow rates are possibly better indicators of the salivary function of a population where the cut –off values were not established through bespoke studies for that population. Further large scale studies are needed to establish the cut –off values appropriate for this population.

The xerostomia questionnaire appears to be a useful tool in indicating those with low salivary flow rates, especially resting flow rates. Studies that evaluate its specificity and sensitivity are necessary before further endorsement.

CHAPTER-7

RECOMMENDATIONS

- All newly diagnosed HIV positive patients should be referred to the dentist for examination and establishing baseline oral conditions. Other health care workers involved in the diagnosis and management of the HIV positive patient should be made aware of the influence that the state of immune suppression and HAART status has on salivary gland function. Enough emphasis cannot be made on the effect that oral health status has on the general well-being and quality of life of an individual.
- Patients showing signs and symptoms of salivary dysfunction should be tested for salivary flow rates and managed with intense prophylactic emphasis.
- Support for longitudinal studies that can further evaluate the long term impact of HAART on oral health.
- Support for large scale studies that can help in evaluating salivary function norms for the South African population and thus help in formulating relevant xerostomia and salivary flow diagnostic measures. This would then pave the way to formalise guidelines for the management of salivary gland dysfunction. These management strategies should include cost effective regimens that will be accessible to all sectors of the South African population.



CHAPTER- 8

REFERENCES

1. Allan, J.D., Connolly, K.J., Fitch, H., Jackson-Pope, L., McLaren, C., Canetta, R. & Groopman, J.E. 1993, "Long-term follow-up of didanosine administered orally twice daily to patients with advanced human immunodeficiency virus infection and hematologic intolerance of zidovudine", *Clinical infectious diseases : an official publication of the Infectious Diseases Society of America*, vol. 16, Suppl 1, pp. S46-51.
2. Antiretroviral Therapy Cohort Collaboration 2008, "Life expectancy of individuals on combination antiretroviral therapy in high-income countries: a collaborative analysis of 14 cohort studies", *Lancet*, vol. 372, no. 9635, pp. 293-299.
3. Apostolova, N., Blas-García, A., & Esplugues, J. V. 2011, "Mitochondrial interference by anti-HIV drugs: mechanisms beyond Pol- inhibition", *Trends in pharmacological sciences*, Vol.32, no.12, pp.715-725.
4. Arendorf, T.M., Bredekamp, B., Cloete, C.A.C. & Sauer, G. 1998, "Oral manifestations of HIV infection in 600 South African patients", *Journal of oral pathology & medicine*, vol. 27, no. 4, pp. 176-179.
5. Atkinson, J.C., Shiroky, J.B., Macynski, A. & Fox, P.C. 1989a, "Effects of furosemide on the oral cavity", *Gerodontology*, vol. 8, no. 1, pp. 23-26.
6. Atkinson, J.C., Yeh, C., Bermudez, D., Fox, P.C. & Baum, B.J. 1989b, "Longitudinal evaluation of major salivary gland function in HIV-1 infected patients", *Journal of Oral Pathology & Medicine*, vol. 18, no. 8, pp. 469-470.
7. Atkinson, J.C., Yeh, C., Oppenheim, F.G., Bermudez, D., Baum, B.J. & Fox, P.C. 1990, "Elevation of salivary antimicrobial proteins following HIV-1 infection", *Journal of Acquired Immune Deficiency Syndromes*, vol. 3, no. 1, pp. 41-48.

8. Atkinson, J.C. & Fox, P.C. 1992, "Salivary gland dysfunction", *Clinics in geriatric medicine*, vol. 8, no. 3, pp. 499-511.
9. Atkinson, J.C., Grisius, M. & Massey, W. 2005, "Salivary hypofunction and xerostomia: diagnosis and treatment", *Dental clinics of North America*, vol. 49, no. 2, pp. 309-326.
10. Beitler, J.J., Vikram, B., Silver, C.E., Rubin, J.S., Bello, J.A., Mitnick, R.J., Gejerman, G. & Davis, L.W. 1995, "Low-dose radiotherapy for multicystic benign lymphoepithelial lesions of the parotid gland in HIV -positive patients: Long -term results", *Head & neck*, vol. 17, no. 1, pp. 31-35.
11. Burger-Calderon, R., Madden, V., Hallett, R.A., Gingerich, A.D., Nickeleit, V. & Webster-Cyriaque, J. 2014, "Replication of oral BK virus in human salivary gland cells", *Journal of virology*, vol. 88, no. 1, pp. 559-573.
12. Busato, I.M.S., Thomaz, M., Toda, A.I., Alanis, D.G.P., França, B.H.S., de Lima, A.A.S. & Azevedo-Alanis, L.R. 2013, "Prevalence and impact of xerostomia on the quality of life of people living with HIV/AIDS from Brazil", *Special Care in Dentistry*, vol. 33, no. 3, pp. 128-132.
13. Cassolato, S.F. & Turnbull, R.S. 2003, "Xerostomia: Clinical Aspects and Treatment", *Gerodontology*, vol. 20, no. 2, pp. 64-77.
14. Cassone, A., Tacconelli, E., De Bernardis, F., Tumbarello, M., Torosantucci, A., Chiani, P. & Cauda, R. 2002, "Antiretroviral therapy with protease inhibitors has an early, immune reconstitution-independent beneficial effect on Candida virulence and oral candidiasis in human immunodeficiency virus-infected subjects", *The Journal of infectious diseases*, vol. 185, no. 2, pp. 188-195.
15. Ceballos-Salobreña, A., Gaitán-Cepeda, L.A., Ceballos-Garcia, L. & Lezama-Del Valle, D. 2000, "Oral lesions in HIV/AIDS patients undergoing highly active antiretroviral treatment including protease inhibitors: a new face of oral AIDS?", *AIDS Patient Care and STDs*, vol. 14, no. 12, pp. 627-635.

16. Chetty, Manickavallie Vaithilingum, Rajendra Thejpal, Runjan 1999, "Epstein-Barr virus status and the histopathological changes of parotid gland lymphoid infiltrates in HIV-positive children", *Pathology*, vol. 31, no. 4, pp. 413-417.
17. Coogan, M.M., Greenspan, J. & Challacombe, S.J. 2005, "Oral lesions in infection with human immunodeficiency virus", *Bulletin of the World Health Organization*, vol. 83, no. 9, pp. 700-706.
18. Dawes, C. 1972, "Circadian rhythms in human salivary flow rate and composition", *The Journal of physiology*, vol. 220, no. 3, pp. 529-545.
19. Dawes, C. 1987, "Physiological factors affecting salivary flow rate, oral sugar clearance, and the sensation of dry mouth in man", *Journal of dental research*, vol. 66 Spec No, pp. 648-653.
20. De Almeida, P D V., Gregio, A., Machado, M., De Lima, A. & Azevedo, L.R. 2008, "Saliva composition and functions: a comprehensive review", *Journal of Contemporary Dental Practice*, vol. 9, no. 3, pp. 72-80.
21. Denny, P. C., Denny, P. A., Klauser, D. K., Hong, S. H., Navazesh, M., & Tabak, L. A. 1991. "Age-related changes in mucins from human whole saliva". *Journal of dental research*, vol.70, no. 10, pp. 1320-1327.
22. Diz Dios, P. & Scully, C. 2014, "Antiretroviral therapy: effects on orofacial health and health care", *Oral diseases*, vol. 20, no. 2, pp. 136-145.
23. Duckworth, R.M., Morgan, S.N. & Murray, A.M. 1987, "Fluoride in saliva and plaque following use of fluoride-containing mouthwashes", *Journal of dental research*, vol. 66, no. 12, pp. 1730-1734.
24. Elliott, J.N. & Oertel, Y.C. 1990, "Lymphoepithelial cysts of the salivary glands. Histologic and cytologic features", *American Journal of Clinical Pathology*, vol. 93, no. 1, pp. 39-43.

25. Ericsson, Y. & Hardwick, L. 1978, "Individual diagnosis, prognosis and counseling for caries prevention", *Caries research*, vol. 12, no. Suppl. 1, pp. 94-102.
26. Farsi, N.M. 2007, "Signs of oral dryness in relation to salivary flow rate, pH, buffering capacity and dry mouth complaints", *BMC oral health*, vol. 7, pp. 15.
27. Fellay, J., Ledergerber, B., Bernasconi, E., Furrer, H., Battegay, M., Hirschel, B., Vernazza, P., Francioli, P., Greub, G. & Flepp, M. 2001, "Prevalence of adverse events associated with potent antiretroviral treatment: Swiss HIV Cohort Study", *The Lancet*, vol. 358, no. 9290, pp. 1322-1327.
28. Feller, L., Anagnostopoulos, C., Wood, N.H., Bouckaert, M., Raubenheimer, E.J. & Lemmer, J. 2008, "Human immunodeficiency virus-associated Kaposi sarcoma as an immune reconstitution inflammatory syndrome: a literature review and case report", *Journal of periodontology*, vol. 79, no. 2, pp. 362-368.
29. Feller, L., Wood, N.H. & Lemmer, J. 2007, "Herpes zoster infection as an immune reconstitution inflammatory syndrome in HIV-seropositive subjects: a review", *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology*, vol. 104, no. 4, pp. 455-460.
30. Flink, H., Bergdahl, M., Tegelberg, Å., Rosenblad, A. & Lagerlöf, F. 2008, "Prevalence of hyposalivation in relation to general health, body mass index and remaining teeth in different age groups of adults", *Community Dentistry & Oral Epidemiology*, vol. 36, no. 6, pp. 523-531.
31. Flint, S.R., Tappuni, A., Leigh, J., Schmidt-Westhausen, A.M. & MacPhail, L. 2006, "(B3) Markers of immunodeficiency and mechanisms of HAART therapy on oral lesions", *Advances in Dental Research*, vol. 19, no. 1, pp. 146-151.
32. Fox, P.C. 1992, "Salivary gland involvement in HIV-1 infection", *Oral surgery, oral medicine, oral pathology*, vol. 73, no. 2, pp. 168-170.

33. Fox, P.C., Busch, K.A. & Baum, B.J. 1987, "Subjective reports of xerostomia and objective measures of salivary gland performance", *Journal Of The American Dental Association* , vol. 115, no. 4, pp. 581-584.
34. Fox, P.C. 1996, "Differentiation of dry mouth etiology", *Advances in Dental Research*, vol. 10, no. 1, pp. 13-16.
35. Fox, R. I. (2005), "Sjögren's syndrome", *The Lancet*, vol. 366, no. 9482, pp. 321-331.
36. Freeman AD, Liberali SA, Coates EA, Logan RM. 2012, "Oral health in Australian HIV patients since the advent of combination antiretroviral therapy", *Australian Dental Journal*, Vol. 57, pp. 470–476.
37. Ghezzi, E.M., Lange, L.A. & Ship, J.A. 2000, "Determination of variation of stimulated salivary flow rates", *Journal of dental research*, vol. 79, no. 11, pp. 1874-1878.
38. Greenberg, M.S., Glick, M., Nghiem, L., Stewart, J.C., Hodinka, R. & Dubin, G. 1997, "Relationship of cytomegalovirus to salivary gland dysfunction in HIV-infected patients", *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology*, vol. 83, no. 3, pp. 334-339.
39. Greenspan, D., Canchola, A.J., MacPhail, L.A., Cheikh, B. & Greenspan, J.S. 2001, "Effect of highly active antiretroviral therapy on frequency of oral warts", *The Lancet*, vol. 357, no. 9266, pp. 1411-1412.
40. Greenspan, J. & Greenspan, D. 2002, "The epidemiology of the oral lesions of HIV infection in the developed world", *Oral diseases*, vol. 8, no. s2, pp. 34-39.
41. Greenspan, D., Gange, S.J., Phelan, J.A., Navazesh, M., Alves, M.E., MacPhail, L.A., Mulligan, R. & Greenspan, J.S. 2004, "Incidence of oral lesions in HIV-1-infected women: reduction with HAART", *Journal of dental research*, vol. 83, no. 2, pp. 145-150.

42. Hall, J.E. 2010, *Guyton and Hall Textbook of Medical Physiology*: Enhanced E-book, Elsevier Health Sciences.
43. Hamza, O.J., Matee, M.I., Simon, E.N., Kikwilu, E., Moshi, M.J., Mugusi, F., Mikx, F.H., Verweij, P.E. & van der Ven, André JAM 2006, "Oral manifestations of HIV infection in children and adults receiving highly active anti-retroviral therapy [HAART] in Dar es Salaam, Tanzania", *BMC oral health*, vol. 6, no. 1, pp. 12.
44. Ho, D.D., Neumann, A.U., Perelson, A.S., Chen, W., Leonard, J.M. & Markowitz, M. 1995, "Rapid turnover of plasma virions and CD4 lymphocytes in HIV-1 infection", *Nature*, vol. 373, no. 6510, pp. 123-126.
45. Humphrey, S.P. & Williamson, R.T. 2001, "A review of saliva: normal composition, flow, and function", *The Journal of prosthetic dentistry*, vol. 85, no. 2, pp. 162-169.
46. Ioachim, H.L. & Ryan, J.R. 1988, "Salivary gland lymphadenopathies associated with AIDS", *Human pathology*, vol. 19, no. 5, pp. 616-617.
47. Itescu, S., Brancato, L.J., Buxbaum, J., Gregersen, P.K., Rizk, C.C., Crosson, T.S., Solomon, G.E. & Winchester, R. 1990, "A diffuse infiltrative CD8 lymphocytosis syndrome in human immunodeficiency virus (HIV) infection: a host immune response associated with HLA-DR5", *Annals of Internal Medicine*, vol. 112, no. 1, pp. 3-10.
48. Itescu, S., & Winchester, R. 1992, "Diffuse infiltrative lymphocytosis syndrome: a disorder occurring in human immunodeficiency virus-1 infection that may present as a sicca syndrome", *Rheumatic diseases clinics of North America*, vol. 18, no. 3, pp. 683-697.
49. Itescu, S., Dalton, J., Zhang, H.Z. & Winchester, R. 1993, "Tissue infiltration in a CD8 lymphocytosis syndrome associated with human immunodeficiency virus-1 infection has the phenotypic appearance of an antigenically driven response", *The Journal of clinical investigation*, vol. 91, no. 5, pp. 2216-2225.

50. Jacob, L.E., Malavika, R., Vasudevan, V., Kumar, M.S. & Kailasam, S. 2013, "Lymphoepithelial Cyst of the Parotid Gland in an HIV-Positive Patient", *Journal of Indian Academy of Oral Medicine and Radiology*, vol. 25, no. 1, pp. 73-76.
51. Jainkittivong, A., Lin, A., Johnson, D., Langlais, R. & Yeh, C. 2009, "Salivary secretion, mucin concentrations and candida carriage in HIV-infected patients", *Oral diseases*, vol. 15, no. 3, pp. 229-234.
52. Jeffers, L.K., Madden, V. & Webster-Cyriaque, J. 2009, "BK virus has tropism for human salivary gland cells in vitro: implications for transmission", *Virology*, vol. 394, no. 2, pp. 183-193.
53. Jeffers, L. & Webster-Cyriaque, J.Y. 2011, "Viruses and salivary gland disease (SGD): lessons from HIV SGD", *Advances in Dental Research*, vol. 23, no. 1, pp. 79-83.
54. Jeganathan, S., Carey, H. & Purnomo, J. 2012, "Impact of xerostomia on oral health and quality of life among adults infected with HIV-1", *Special Care in Dentistry*, vol. 32, no. 4, pp. 130-135.
55. Jenkins, G.N. & Edgar, W.M. 1989, "The effect of daily gum-chewing on salivary flow rates in man", *Journal of dental research*, vol. 68, no. 5, pp. 786-790.
56. Jha, N., Seikaly, H., Harris, J., Williams, D., Liu, R., McGaw, T., & Barnaby, P. 2003. "Prevention of radiation induced xerostomia by surgical transfer of submandibular salivary gland into the submental space". *Radiotherapy and oncology*, vol. 66, no. 3, pp. 283-289.
57. Johar, N., Proctor, G.B. & Kumar, R. 2011, "A Comparative Study of Salivary Composition of HIV Seropositive Patients on HAART and Not on HAART", *Journal of Indian Academy of Oral Medicine and Radiology*, vol. 23, no. 1, pp. 29-32.

58. Joint United Nations Program on HIV/AIDS 2012, "Global report: UNAIDS report on the global AIDS epidemic: 2012", UNAIDS.
59. Katz, M.H., Mastrucci, M.T., Leggott, P.J., Westenhouse, J., Greenspan, J.S. & Scott, G.B. 1993, "Prognostic significance of oral lesions in children with perinatally acquired human immunodeficiency virus infection", *American Journal of Diseases of Children*, vol. 147, no. 1, pp. 45-48.
60. Klein, R.S., Harris, C.A., Small, C.B., Moll, B., Lesser, M. & Friedland, G.H. 1984, "Oral candidiasis in high-risk patients as the initial manifestation of the acquired immunodeficiency syndrome", *New England Journal of Medicine*, vol. 311, no. 6, pp. 354-358.
61. Klein, R. S., Quart, A. M., & Small, C. B. 1991, "Periodontal disease in heterosexuals with acquired immunodeficiency syndrome" *Journal of periodontology*, vol. 62, no.8, pp. 535-540.
62. Kungoane, T. 2011, "Lymphoid proliferation of the parotid gland in paediatric patients with HIV infection" (Thesis), Faculty of health sciences, University of Witwatersrand, Jhb, SA.
63. Lin, A.L., Johnson, D.A., Patterson, T.F., Wu, Y., Lu, D.L., Shi, Q. & Yeh, C. 2001, "Salivary anticandidal activity and saliva composition in an HIV-infected cohort", *Oral Microbiology & Immunology*, vol. 16, no. 5, pp. 270-278.
64. Lin, A.L., Johnson, D.A., Stephan, K.T. & Yeh, C.K. 2003, "Alteration in salivary function in early HIV infection", *Journal of dental research*, vol. 82, no. 9, pp. 719-724.
65. Lin, A.L., Johnson, D.A., Stephan, K.T. & Yeh, C. 2004, "Salivary secretory leukocyte protease inhibitor increases in HIV infection", *Journal of Oral Pathology & Medicine*, vol. 33, no. 7, pp. 410-416.
66. Lin, A.L., Johnson, D.A., Sims, C.A., Stephan, K.T. & Yeh, C. 2006, "Salivary gland function in HIV-infected patients treated with highly active

- antiretroviral therapy (HAART)", *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology*, vol. 102, no. 3, pp. 318-324.
67. López-Verdín, S., Andrade-Villanueva, J., Zamora-Perez, A.L., Bologna-Molina, R., Cervantes-Cabrera, J.J. & Molina-Frechero, N. 2013, "Differences in Salivary Flow Level, Xerostomia, and Flavor Alteration in Mexican HIV Patients Who Did or Did Not Receive Antiretroviral Therapy", *AIDS research and treatment*, vol. 2013.
68. Mandel, I. D. 1989, "The role of saliva in maintaining oral homeostasis", *Journal of the American Dental Association*, vol. 119, no.2, pp.298-304.
69. Mandel, I.D., Barr, C.E. & Turgeon, L. 1992, "Longitudinal study of parotid saliva in HIV-1 infection", *Journal of oral pathology & medicine*, vol. 21, no. 5, pp. 209-213.
70. Mandel, L. & Reich, R. 1992, "HIV parotid gland lymphoepithelial cysts: Review and case reports", *Oral surgery, oral medicine, oral pathology*, vol. 74, no. 3, pp. 273-278.
71. Mandel, L., Kim, D. & Uy, C. 1998, "Parotid gland swelling in HIV diffuse infiltrative CD8 lymphocytosis syndrome", *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology*, vol. 85, no. 5, pp. 565-568.
72. Mandel, L. & Surattanont, F. 2002, "Regression of HIV parotid swellings after antiviral therapy: case reports with computed tomographic scan evidence", *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology*, vol. 94, no. 4, pp. 454-459.
73. Masiwa, A. & Naidoo, S. 2011, "Oral lesions in HIV-infected patients, before and after antiretroviral treatment: original research", *Southern African Journal of Epidemiology and Infection*, vol. 26, no. 4, pp. 271-273.

74. Matee, M., Scheutz, F. & Moshy, J. 2000, "Occurrence of oral lesions in relation to clinical and immunological status among HIV-infected adult Tanzanians", *Oral diseases*, vol. 6, no. 2, pp. 106-111.
75. McArthur, C.P., Subtil-DeOliveira, A., Palmer, D., Fiorella, R.M., Gustafson, S., Tira, D. & Miranda, R.N. 2000, "Characteristics of salivary diffuse infiltrative lymphocytosis syndrome in West Africa: Human immunodeficiency virus-associated salivary ductal epithelial atypia?", *Archives of Pathology & Laboratory Medicine*, vol. 124, no. 12, pp. 1773-1779.
76. McCarthy, G.M., Mackle, I.D., Koval, J., Sandhu, H.S. & Daley, T.D. 1991, "Factors associated with increased frequency of HIV-related oral candidiasis", *Journal of Oral Pathology & Medicine*, vol. 20, no. 7, pp. 332-336.
77. Munro, C.A. & Hube, B. 2002, "Anti-fungal therapy at the HAART of viral therapy", *Trends in microbiology*, vol. 10, no. 4, pp. 173-177.
78. Murri, R., Ammassari, A., Gallicano, K., De Luca, A., Cingolani, A., Jacobson, D., Wu, A.W. & Antinori, A. 2000, "Patient-reported nonadherence to HAART is related to protease inhibitor levels", *Journal of Acquired Immune Deficiency Syndromes*, vol. 24, no. 2, pp. 123-128.
79. Nagler, R.M. & Hershkovich, O. 2005, "Relationships between age, drugs, oral sensorial complaints and salivary profile", *Archives of Oral Biology*, vol. 50, no. 1, pp. 7-16.
80. Naidoo, S. & Chikte, U. 2004, "HIV Disease Oro-facial manifestations in paediatric HIV: a comparative study of institutionalized and hospital outpatients", *Oral diseases*, vol. 10, no. 1, pp. 13-18.
81. Napeñas, J.J., Brennan, M.T. & Fox, P.C. 2009, "Diagnosis and treatment of xerostomia (dry mouth)", *Odontology*, vol. 97, no. 2, pp. 76-83.

82. Narhi, T.O., Meurman, J.H., Ainamo, A., Nevalainen, J.M., Schmidt-Kaunisaho, K., Siukosaari, P., Valvanne, J., Erkinjuntti, T., Tilvis, R. & Malika, E. 1992, "Association Between Salivary Flow Rate and the Use of Systemic Medication Among 76-, 81-, and 86-year-old Inhabitants in Helsinki, Finland", *Journal of dental research*, vol. 71, no. 12, pp. 1875-1880.
83. Narhi, T.O. 1994, "Prevalence of subjective feelings of dry mouth in the elderly", *Journal of dental research*, vol. 73, no. 1, pp. 20-25.
84. Närhi, T.O., Meurman, J.H. & Ainamo, A. 1999, "Xerostomia and hyposalivation", *Drugs & aging*, vol. 15, no. 2, pp. 103-116.
85. National Department of Health, South Africa; "The South African Antiretroviral Treatment Guidelines 2013". In: Sahivsoc.org (Internet). (updated 2013 March 21; cited 2014 July 17); Available from <http://www.sahivsoc.org/upload/documents/2013%20ART%20Guidelines-Short%20Combined%20FINAL%20draft%20guidelines%2014%20March%202013.pdf>
86. Navazesh, M., Christensen, C. & Brightman, V. 1992a, "Clinical criteria for the diagnosis of salivary gland hypofunction", *Journal of dental research*, vol. 71, no. 7, pp. 1363-1369.
87. Navazesh, M., Mulligan, R.A., Kipnis, V., Denny, P.A. & Denny, P.C. 1992b, "Comparison of whole saliva flow rates and mucin concentrations in healthy Caucasian young and aged adults", *Journal of dental research*, vol. 71, no. 6, pp. 1275-1278.
88. Navazesh, M. 1993, "Methods for collecting saliva", *Annals of the New York Academy of Sciences*, vol. 694, no. 1, pp. 72-77.
89. Navazesh, M., Brightman, V.J. & Pogoda, J.M. 1996, "Relationship of medical status, medications, and salivary flow rates in adults of different ages", *Oral surgery, oral medicine, oral pathology, oral radiology, and endodontics*, vol. 81, no. 2, pp. 172-176.

90. Navazesh, M., Mulligan, R., Komaroff, E., Redford, M., Greenspan, D. & Pkelan, J. 2000, "The Prevalence of Xerostomia and Salivary Gland Hypofunction in a Cohort of HIV-positive and At-risk Women", *Journal of dental research*, vol. 79, no. 7, pp. 1502-1507.
91. Navazesh, M., Mulligan, R., Barrón, Y., Redford, M., Greenspan, D., Alves, M. & Phelan, J. 2003, "A 4-year longitudinal evaluation of xerostomia and salivary gland hypofunction in the Women's Interagency HIV Study participants", *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology*, vol. 95, no. 6, pp. 693-698
92. Navazesh, M., Mulligan, R., Pogoda, J., Greenspan, D., Alves, M., Phelan, J., Greenspan, J. & Slots, J. 2005, "The effect of HAART on salivary microbiota in the Women's Interagency HIV Study (WIHS)", *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology*, vol. 100, no. 6, pp. 701-708.
93. Navazesh, M., Mulligan, R., Karim, R., Mack, W.J., Ram, S., Seirawan, H., Greenspan, J., Greenspan, D., Phelan, J. & Alves, M. 2009, "Effect of HAART on salivary gland function in the Women's Interagency HIV Study (WIHS)", *Oral diseases*, vol. 15, no. 1, pp. 52-60.
94. Navazesh, M., Mulligan, R., Kono, N., Kumar, S.K., Nowicki, M., Alves, M. & Mack, W.J. 2010, "Oral and systemic health correlates of HIV-1 shedding in saliva", *Journal of dental research*, vol. 89, no. 10, pp. 1074-1079.
95. Nederfors, T., Isaksson, R., Mörnstad, H. & Dahlöf, C. 1997, "Prevalence of perceived symptoms of dry mouth in an adult Swedish population-relation to age, sex and pharmacotherapy", *Community dentistry and oral epidemiology*, vol. 25, no. 3, pp. 211-216.
96. Nederfors, T. 2000, "Xerostomia and hyposalivation", *Advances in Dental Research*, vol. 14, pp. 48-56.
97. Nittayananta, W. & Chungpanich, S. 1997, "Oral lesions in a group of Thai people with AIDS", *Oral diseases*, vol. 3, no. S1, pp. S41-S45.

98. Nittayananta, W., Talungchit, S., Jaruratanasirikul, S., Silpapojakul, K., Chayakul, P., Nilmanat, A. & Pruphetkaew, N. 2010a, "Effects of long-term use of HAART on oral health status of HIV-infected subjects", *Journal of Oral Pathology & Medicine*, vol. 39, no. 5, pp. 397-406.
99. Nittayananta, W., Chanowanna, N., Jealae, S., Nauntofte, B. & Stoltze, K. 2010b, "Hyposalivation, xerostomia and oral health status of HIV-infected subjects in Thailand before HAART era", *Journal of Oral Pathology & Medicine*, vol. 39, no. 1, pp. 28-34.
100. Olive, A., Salavert, A., Manriquez, M., Clotet, B. & Moragas, A. 1998, "Parotid lipomatosis in HIV positive patients: a new clinical disorder associated with protease inhibitors", *Annals of the Rheumatic Diseases*, vol. 57, no. 12, pp. 749.
101. Orellana, M.F., Lagravère, M.O., Boychuk, D.G.J., Major, P.W. & Flores-Mir, C. 2006, "Prevalence of Xerostomia in Population-based Samples: A Systematic Review", *Journal of public health dentistry*, vol. 66, no. 2, pp. 152-158.
102. Ortega, K.L., Vale, D.A. & Magalhães, M.H. 2009, "Impact of PI and NNRTI HAART-based therapy on oral lesions of Brazilian HIV-infected patients", *Journal of oral pathology & medicine*, vol. 38, no. 6, pp. 489-494.
103. Österberg, T., Landahl, S. & Hedegård, B. 1984, "Salivary flow, saliva, pH and buffering capacity in 70-year-old men and women", *Journal of oral rehabilitation*, vol. 11, no. 2, pp. 157-170.
104. Pai, S., Ghezzi, E.M. & Ship, J.A. 2001, "Development of a Visual Analogue Scale questionnaire for subjective assessment of salivary dysfunction", *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology*, vol. 91, no. 3, pp. 311-316.
105. Patton, L.L., McKaig, R., Strauss, R., Rogers, D. & Eron Jr, J.J. 2000, "Changing prevalence of oral manifestations of human immuno-deficiency virus in the era of protease inhibitor therapy", *Oral Surgery, Oral Medicine,*

Oral Pathology, Oral Radiology, and Endodontology, vol. 89, no. 3, pp. 299-304.

106. Patton LL, Phelan JA, Ramos-Gomez FJ, Nittayananta W, Shiboski CH, Mbuguye T. 2002, Prevalence and classification of HIV-associated oral lesions. *Oral Diseases*; vol. 8, no.2, pp. 98–109.
107. Patton L, Ramirez-Amador V, Anaya-Saavedra G, Nittayananta W, Carrozzo M, Ranganathan K. 2013, Urban legends series: oral manifestations of HIV infection. *Oral Disease*, vol 19, no.6, pp. 533-550
108. Pavithra, S., Ranganathan, K., Rao, U.K., Joshua, E., Rooban, T. & Kumarasamy, N. 2013, "Impact of highly active antiretroviral therapy on salivary flow in patients with human-immuno deficiency virus disease in Southern India", *Journal of oral and maxillofacial pathology*, vol. 17, no. 1, pp. 17.
109. Pedersen AML. 2007, "Saliva", Zendium, available from (cited 29 June 2014)<<http://www.zendium.dk/Files/zendium.dk/material/publikationer/saliva.pdf>>
110. Percival, R.S., Challacombe, S.J. & Marsh, P.D. 1994, "Flow rates of resting whole and stimulated parotid saliva in relation to age and gender", *Journal of dental research*, vol. 73, no. 8, pp. 1416-1420.
111. Porter, S. & Scully, C. 1998, "HIV topic update: protease inhibitor therapy and oral health care", *Oral diseases*, vol. 4, no. 3, pp. 159-163.
112. Ramírez-Amador, V., Esquivel-Pedraza, L., Sierra-Madero, J., Anaya-Saavedra, G., González-Ramírez, I. & Ponce-de-León, S. 2003, "The changing clinical spectrum of human immunodeficiency virus (HIV)-related oral lesions in 1,000 consecutive patients: a 12-year study in a referral center in Mexico", *Medicine*, vol. 82, no. 1, pp. 39-50.
113. Ramirez-Amador, V.A., Espinosa, E., Gonzalez-Ramirez, I., Anaya-Saavedra, G., Ormsby, C.E. & Reyes-Teran, G. 2009, "Identification of oral

candidosis, hairy leukoplakia and recurrent oral ulcers as distinct cases of immune reconstitution inflammatory syndrome", *International Journal of STD & AIDS*, vol. 20, no. 4, pp. 259-261.

114. Rhodus, N.L. 1988, "Qualitative nutritional intake analysis of older adults with sjogren's syndrome", *Gerodontology*, vol. 7, no. 2, pp. 61-69.
115. Rivera, H., Nikitakis, N.G., Castillo, S., Siavash, H., Papadimitriou, J.C. & Sauk, J.J. 2003, "Histopathological analysis and demonstration of EBV and HIV p-24 antigen but not CMV expression in labial minor salivary glands of HIV patients affected by diffuse infiltrative lymphocytosis syndrome", *Journal of oral pathology & medicine*, vol. 32, no. 7, pp. 431-437.
116. Rostron, J., Rogers, S., Longman, L., Kancy, S. & Field, E.A. 2002, "Health-related quality of life in patients with Primary Sjögren's Syndrome and Xerostomia: a comparative study", *Gerodontology*, vol. 19, no. 1, pp. 53-59.
117. Sales-Peres, Sílvia Helena de Carvalho, Mapengo, M.A.A., Moura-Grec, P.G.d., Marsicano, J.A., Sales-Peres, A.d.C. & Sales-Peres, A. 2012, "Oral manifestations in HIV children in Mozambique", *Ciência & Saúde Coletiva*, vol. 17, no. 1, pp. 55-60.
118. Schiødt, M., Greenspan, D., Daniels, T.E., Nelson, J., Leggott, P.J., Wara, D.W. & Greenspan, J.S. 1989, "Parotid gland enlargement and xerostomia associated with labial sialadenitis in HIV-infected patients", *Journal of Autoimmunity*, vol. 2, no. 4, pp. 415-425.
119. Schiødt, M., Dodd, C.L., Greenspan, D., Daniels, T.E., Chernoff, D., Hollander, H., Wara, D. & Greenspan, J.S. 1992, "Natural history of HIV-associated salivary gland disease", *Oral surgery, oral medicine, oral pathology*, vol. 74, no. 3, pp. 326-331.
120. Schmidt-Westhausen, A.M., Priepeke, F., Bergmann, F.J. & Reichart, P.A. 2000, "Decline in the rate of oral opportunistic infections following

introduction of highly active antiretroviral therapy", *Journal of oral pathology & medicine*, vol. 29, no. 7, pp. 336-341.

121. Scott, J. 1975, "Age, sex and contralateral differences in the volumes of human submandibular salivary glands", *Archives of Oral Biology*, vol. 20, no. 12, pp. 885-IN35.
122. Shannon, I.L., Trodahl, J.N. & Starcke, E.N. 1978, "Radiosensitivity of the human parotid gland", *Proceedings of the Society for Experimental Biology and Medicine*. Society for Experimental Biology and Medicine (New York, N.Y.), vol. 157, no. 1, pp. 50-53.
123. Shelburne, S.A., Montes, M. & Hamill, R.J. 2006, "Immune reconstitution inflammatory syndrome: more answers, more questions", *The Journal of antimicrobial chemotherapy*, vol. 57, no. 2, pp. 167-170.
124. Ship, J.A., Nolan, N.E. & Puckett, S.A. 1995, "Longitudinal analysis of parotid and submandibular salivary flow rates in healthy, different-aged adults", *The journals of gerontology. Series A, Biological sciences and medical sciences*, vol. 50, no. 5, pp. M285-9.
125. Ship, J.A. & Fischer, D.J. 1997, "The relationship between dehydration and parotid salivary gland function in young and older healthy adults", *The journals of gerontology. Series A, Biological sciences and medical sciences*, vol. 52, no. 5, pp. M310-9.
126. Ship, J. 2002, "Diagnosing, managing, and preventing salivary gland disorders", *Oral diseases*, vol. 8, no. 2, pp. 77-89.
127. Shisana, O, Rehle, T, Simbayi LC, Zuma, K, Jooste, S, Zungu N, Labadarios, D, Onoya, D et al. (2014), "South African National HIV Prevalence, Incidence and Behaviour Survey, 2012". Cape Town, Human Sciences Research Council Press.
128. Silverberg, M.J., Gore, M.E., French, A.L., Gandhi, M., Glesby, M.J., Kovacs, A., Wilson, T.E., Young, M.A. & Gange, S.J. 2004, "Prevalence of

- clinical symptoms associated with highly active antiretroviral therapy in the Women's Interagency HIV Study", *Clinical infectious diseases : an official publication of the Infectious Diseases Society of America*, vol. 39, no. 5, pp. 717-724.
129. Sreebny, L.M. & Valdini, A. 1988, "Xerostomia. Part I: Relationship to other oral symptoms and salivary gland hypofunction", *Oral Surgery, Oral Medicine, Oral Pathology*, vol. 66, no. 4, pp. 451-458.
130. Sreebny, L.M., Valdini, A. & Yu, A. 1989, "Xerostomia. Part II: Relationship to nonoral symptoms, drugs, and diseases", *Oral Surgery, Oral Medicine, Oral Pathology*, vol. 68, no. 4, pp. 419-427.
131. Sreebny, L.M., Yu, A., Green, A. & Valdini, A. 1992, "Xerostomia in diabetes mellitus", *Diabetes care*, vol. 15, no. 7, pp. 900-904.
132. Sreebny, L.M. & Schwartz, S.S. 1997, "A reference guide to drugs and dry mouth—2nd edition", *Gerodontology*, vol. 14, no. 1, pp. 33-47.
133. Steinberg, L.M., Odusola, F. & Mandel, I.D. 1992, "Remineralizing potential, antiplaque and antigingivitis effects of xylitol and sorbitol sweetened chewing gum", *Clinical preventive dentistry*, vol. 14, no. 5, pp. 31-34.
134. Stevenson, H., Jones, M., Rostron, J., Longman, L. & Field, E. 2004, "UK patients with primary Sjögren's syndrome are at increased risk from clinical depression", *Gerodontology*, vol. 21, no. 3, pp. 141-145.
135. Syebele, K. & Bütow, K. 2011, "Comparative study of the effect of antiretroviral therapy on benign lymphoepithelial cyst of parotid glands and ranulas in HIV-positive patients", *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology*, vol. 111, no. 2, pp. 205-210.
136. Thomson, W.M., Chalmers, J.M., Spencer, A.J. & Williams, S.M. 1999a, "The Xerostomia Inventory: a multi-item approach to measuring dry mouth", *Community dental health*, vol. 16, no. 1, pp. 12-17.

137. Thomson, W.M., Chalmers, J.M., Spencer, A.J. & Ketabi, M. 1999b, "The occurrence of xerostomia and salivary gland hypofunction in a population-based sample of older South Australians", *Special Care in Dentistry*, vol. 19, no. 1, pp. 20-23.
138. Toida, M., Nanya, Y., Takeda-Kawaguchi, T., Baba, S., Iida, K., Kato, K., Hatakeyama, D., Makita, H., Yamashita, T. & Shibata, T. 2010, "Oral complaints and stimulated salivary flow rate in 1188 adults", *Journal of oral pathology & medicine*, vol. 39, no. 5, pp. 407-419.
139. Torres, S.R., Peixoto, C.B., Caldas, D.M., Silva, E.B., Akiti, T., Nucci, M. & de Uzeda, M. 2002, "Relationship between salivary flow rates and Candida counts in subjects with xerostomia", *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology*, vol. 93, no. 2, pp. 149-154.
140. Umadevi, K., Ranganathan, K., Pavithra, S., Hemalatha, R., Saraswathi, T., Kumarasamy, N., Solomon, S. & Greenspan, J.S. 2007, "Oral lesions among persons with HIV disease with and without highly active antiretroviral therapy in southern India", *Journal of oral pathology & medicine*, vol. 36, no. 3, pp. 136-141.
141. von Bültzingslöwen, I., Sollecito, T.P., Fox, P.C., Daniels, T., Jonsson, R., Lockhart, P.B., Wray, D., Brennan, M.T., Carrozzo, M. & Gandera, B. 2007, "Salivary dysfunction associated with systemic diseases: systematic review and clinical management recommendations", *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology*, vol. 103, pp. S57. e1-S57. e15.
142. van der Putten, G., Brand, H., Schols, J. & Baat, C. 2011, "The diagnostic suitability of a xerostomia questionnaire and the association between xerostomia, hyposalivation and medication use in a group of nursing home residents", *Clinical oral investigations*, vol. 15, no. 2, pp. 185-192.
143. Webster-Cyriaque, J; Ass Prof, Univ N. Carolina, USA, "BK Virus in salivary gland disease". In: ClinicalTrials.gov [Internet]. Bethesda (MD):

National Library of Medicine (US). (Updated 2014 March 5; cited 2014 July 6). Available from <http://clinicaltrials.gov/show/NCT02068846>

144. Wu, A.J. & Ship, J.A. 1993, "A characterization of major salivary gland flow rates in the presence of medications and systemic diseases", *Oral surgery, oral medicine, oral pathology*, vol. 76, no. 3, pp. 301-306.
145. Yeh, C., Fox, P.C., Ship, J.A., Busch, K.A., Bermudez, D.K., Wilder, A., Katz, R.W., Wolff, A., Tylanda, C.A. & Atkinson, J.C. 1988, "Oral defense mechanisms are impaired early in HIV-1 infected patients", *JAIDS Journal of Acquired Immune Deficiency Syndromes*, vol. 1, no. 4, pp. 361-366.
146. Yeh, C., Johnson, D. & Dodds, M. 1998, "Impact of aging on human salivary gland function: a community-based study", *Aging Clinical and Experimental Research*, vol. 10, no. 5, pp. 421-428.
147. Zolopa, A.R., Andersen, J., Komarow, L., Sanne, I., Sanchez, A., Hogg, E., Suckow, C., Powderly, W. & ACTG A5164 study team 2009, "Early antiretroviral therapy reduces AIDS progression/death in individuals with acute opportunistic infections: a multicenter randomized strategy trial", *PloS one*, vol. 4, no. 5, pp. e5575.

CHAPTER-9
APPENDICES

Appendix-1

PATIENT INFORMATION

Dear Mr/Mrs/Ms _____

Thank you for agreeing to take part in the study.

Before we proceed, please ensure that your last meal, drink and or cigarette have been at least 90 mins or more ago. I will be asking you a few questions and will write down your answers. If you need further explanation at any point please do not hesitate to ask. After the questions we will begin the process of saliva collection. Saliva will be collected twice from you as explained below:

- Please sit and relax for 5 mins before we start.
- Please rinse your mouth with the water provided before we start.
- Saliva at rest: collection time is 3 mins.
You will be given a tube fitted with a funnel. Please allow saliva to collect in your mouth and gently spit into the funnel every minute. You will be notified of the time. At the end of 3 mins, spit all the saliva into the funnel to completely empty your mouth.
- Rest for 5 mins.
- Saliva under stimulation: collection time is 3 mins.
You will be given a new tube with a funnel. This time you will be given a piece of rubber to chew on and you will spit only the saliva gently into the tube every 1 min. Chewing will be at the rate of 45 stroke per minute keeping in time with a sound you will hear. You will be informed at the end of each minute to spit. At the end of the 3 mins, spit the saliva into the funnel to completely empty your mouth of saliva.

• You may discard the piece of rubber, rinse out your mouth and clean up
Thank you kindly for your participation in our effort to improve knowledge in the oral health field. If you have any questions or queries or would like more information, please contact Dr Cherian on 0437331289. Have a good day further.

Mr/Mrs/Ms _____

Ndiyabulela ngokuba uvumile ukuthabatha inkxhaxheba koluphando.

Uncede ungaryi, ungaseli kwaye ungatshayi iyure enesiqingatha phambi kokuba siqale. Ndizakubuza imibuzo endakuthi impendulo zakho ndizibhale phantsi, ukuba ufuna ingcaciso ethe vetshe ungalibazisi ukubuza. Emveni kwemibuzo ndizakuqokelela amathe akho. Amathe akho azakuqokelelwa kabini njengokuba kuchaziwe apha ngezantsi. Ndicela uhlale phantsi, uphumle imizuzu ibemihlanu phambi kokuba siqale.

- Xukuxa ngalo manzi alapho phambi ko kuba siqale.
- Sizakuthabatha imizuzu emithathu siqokele amathe akho ngokuziphumela kwawo emlonyeni wakho. Uzakunikwa umbhobho onefanele. Nceda uvumele amathe aqokelelane emlonyeni wakho emva koko uwathufe apha kwifanele qho emva komzuzu omnye. Sizakwazisa xa kuphela umzuzu.. emveni kwemizuzu emithathu, nceda uthufele kwifanele onke amathe asemfonyeni, kungashiyeki mathe.
- Phumla ke imizuzu emihlanu.
- Sizakuthabatha imizuzu emithathu siqokele amathe akho ngokuthi siwabizele ngokwethu. Uzakunikwa ifanele entsha kwakhona. siphinde sikunike intwana encinci ye rubber omawuyihlafune uze qho emveni komzuzu uthufe amathe odwa. Uzakuhlafuna qho xa usiva isandi esakuthi sisikhalise kanga mathuba amashumi amane anesihlanu emzuzwini. sizakwazisa xa ilixesha lokuthufa emveni komzuzu omnye. xa kuphele imizuzu emithathu thufela onke amathe asemfonyeni wakho.
- Sigqibile ngoku kwaye ungawuhlamba umlomo wakho uzicoce.

Ndiyabulela kakhulu ngokuba uyewathatha inkxhaxheba ekuphuhliseni iziko lwezempilo lwamazinyo. Ukuba unemibuzo okanye kukho izinto ongathanda ukuziqonda, nditsalele umnxeba kulenombolo 043 7331289.

Ulonwabele usuku lwakho.

Mnr. /Mev _____

Baie dankie dat u ingestem het om deel te neem aan die studie. Maak seker dat jy meer as n uur en n half laas iets geeet, gedrink of gerook het. Ek gaan u n paar vrae vra en u antwoorde neerskryf, vra asseblief as ek enige iets weer moet verduidelik. Nadat ek die vrae gevra het kan ek die proses begin om die speeksel te neem, soos ek hieronder verduidelik.

- Sit asseblief nou en ontspan vir 5 min voordat ons begin.
- Spoel asseblief jou mond uit met die water.
- Speeksel toets: tyd vir opname is 3 min.
- Jy gaan n buisie met n tregter kry. Laat die speeksel versamel in jou mond en versigtig spoeg die speeksel in die buisie na elke minuut. Jy sal gese word van die tyd. Na die einde van die 3min, spoeg al die speeksel in jou mond uit in die buisie.
- Rus nou vir 5 min.
- Speeksel onder stimulasie: tyd is 3 min. Jy gaan n nuwe buisie en tregter kry. Hierdie keer gaan ons vir jou n stukkie rubber gee om aan te kou en dan na elke 1 min versigtig in die tregter spoeg. Luister na die klank van die horlosie en kou elke keer as die horlosie tik. Jy sal gese word om na elke min versigtig die speeksel uittespoeg. Na 3min spoeg al die speeksel in jou mond uit in die buisie.
- U kan nou u mond uitspoel.

Baie dankie vir u deelname in hierdie studie en om kennis van mond gesondheid te verbeter. As u enige navrae of meer inligting wil he, kontak asseblief vir Dr. Cherian by 043 7331289.

Geniet die dag verder.

INFORMED CONSENT FORM

Appendix--4

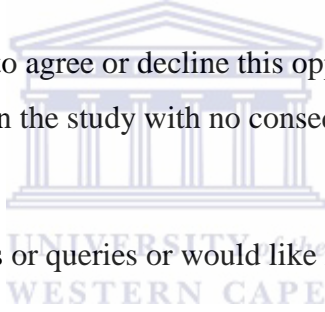
Patient No: _____

Good day Mr/Mrs/Ms _____

I am Dr Anney Cherian and I am a dentist at Empilweni Gompo Community Health Centre. I am also a Masters student at dental faculty of the University of Western Cape. I would like to invite you to take part in my research/ study. The study is to see if there is a relationship between dry mouth and HIV medication. It will involve answering a few questions and collecting your saliva (spit) in a tube. It will take about 20 -30 mins.

All information collected from you will be held confidentially and will be filed under a code number that has been allocated to you. Your name will not appear on any form.

You are completely free to agree or decline this opportunity to be involved in this study or not to take part in the study with no consequence to your treatment or care at this facility.



If you have any questions or queries or would like more information, please contact Dr Cherian on 0437331289.

If you would like to participate please sign the consent form below.

I acknowledge that this study has been explained to me and understand what the study is about and would like to take part in it. I also understand that the results will not disclose my identity and results used or published will be for the benefit in the medical/ dental field.

Name: _____ Signature: _____ Date: _____

Witness: _____ Signature: _____ Date: _____

IFORMU YEMVUMELVANO

Appendix- 5

Patient No: _____

Molo Mr/Mrs/Ms _____

Igama lam ngu Anney Cherian ugqira wamazinyo Empilweni Gompo. Ndikwa ngumfundi we zifundo eziphakamileyo kwicandelo lwezifundo zamazinyo kwi University yase Ntshona Koloni . Ndiyakucela undincedise ngokuthatha inxhaxheba kwizifundo nophando endilwenzayo. Sizama ukuphanda ukuba ingaba bukho na ubudlelwane phakathi kwamachiza okunyanga intsholongwana kaGawulayo no koma komlomo kwabo bawasebenzisayo.

Uzakundincedisa ngokuphendula imibuzwana nje embalwa nangokunikezela ngamathe akho. Sizakuthatha imizuzu engamashumi amabini uyokutsho kumashumi amathathu.

Yonke inkcazelo yakho neziphumo esizifumeneyo ayizokupapashwa nakubani, iyakugcinwa ikhuselwe phantsi kwe nombolo enikezwe wena kuphela. Igama lakho alizokubhalwa kulamaphepha emibuzo.

Ukhululekile ukuba uthathe inxhaxheba koluphando okanye wale, lilungelo lakho.

Ukuba unemibuzo okanye kukho izinto ongathanda ukuziqonda, nditsalele umnxeba kulenombolo 043 7331289.

Ndicela usayine apha ngezantsi ukuba uyavuma ukuthatha inxhaxheba.

Ndiyavuma ukuba ndicaciselwe ngoluphando kwaye ndiyaluqonda ukuba lungantoni. Ndiyavuma ukuthatha inxhaxheba. Ndiyayiqonda nento yokuba iziphumo zoluphando azizokupapashwa negama lam kwaye iziphumo zizakusetyenziselwa ukuphuhlisa imfundo kwicandelo lezempilo nelamazinyo.

Igama: _____ Sayina: _____ Umhla: _____

Igama leingqina: _____ Sayina: _____ Umhla: _____

INLIGTINGS VORM

Appendix- 6

Patient No: _____

Mnr/Mev _____

My naam is Dr. Anney en ek is n tandarts by Empilweni Gompo Gemeenskap Gesondheid kliniek. Ek is n Meestersgraad student aan die Tandheelkunde fakulteit van die Universiteit van die Wes Kaap. Ek wil u graag uitnoui om deel te neem aan my studie/navorsing. Die studie gaan oor die verwantskap tussen droe mond en HIV medikasie. Dit sal beteken dat u n paar vrae moet beantwoord en n monster van u speeksel moet geneem word. Dit sal omtrent 20 – 30 min van u tyd neem.

Die inligting wat ek neem sal as konfidentieel hanteer word en net as n nommer teen u naam verskyn. U naam sal nie op enige vorms verskyn nie.

U deelname aan die studie is vrywillig en sal nie negatiewe gevolge he op die gehalte diens en behandeling wat u ontvang by die kliniek nie.

As u enige navrae of meer inligting wil he, kontak asseblief vir Dr. Cherian by 043 7331289.

As u wil deelneem aan die studie, teken asseblief die aangehegte toestemmings vorm onderaan.

Ek erken dat die studie aan my verduidelik is, waaroor dit gaan en sal graag daaraan deelneem. Ek verstaan ook dat die resultate nie my identiteit sal weergee nie en dat dit net vir mediese navorsing gebruik sal word.

Naam: _____ Handtekening: _____ Datum: _____

Getuie: _____ Handtekening: _____ Datum: _____

DATA COLLECTION FORMS
GENERAL QUESTIONNAIRE

Appendix- 7

Pt No: _____

Patient Number: _____

Date: _____

Referred from: _____

1. Age _____ years

2. Gender: Male / Female

3. Last meal, drink, smoke _____

4. Do you smoke: (*Uyatshaya*) (Roker) : Yes / No

5. If yes, how many cigarettes do you smoke in a day? (*Ukuba uyatshayautshaya izigarethi ezingaphi ngosuku?*) (As ja, hoeveel sigarette rook u per dag?) < 5 / <10 / >10

6. HIV Status: _____ Since: _____

7. CD4 Count (nearest available) _____ Date Taken _____

8. Viral load (nearest available) _____ Date Taken _____

9. Are you currently on medication for HIV? Yes / No
(*Ingaba usebenzisa amachiza okunyanga ugawulayo?*) (Is U tans op medikasie vir HIV?)

If yes, dated started: (*Uqale nini?*) (As ja, watter datum?) _____

Pt No: _____

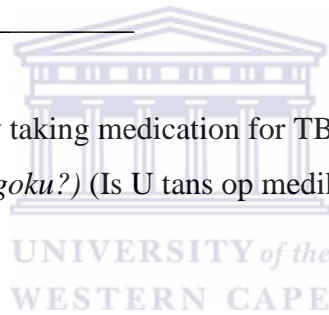
10. Details of HIV medication: Bactrim Yes/ No
IPT Yes/ No
HAART Yes/ No.
Any other Yes/ No

11. Details of HAART: _____

12. Have you tested Positive for TB?: (*Ingaba wakhe wafunyaniswa une TB*) (Het U getoets vir TB, is die toets positief) : Yes/ No

If yes, when (*Ufunyaniswe nini?*) (As ja, watter datum?)

13. Are you currently taking medication for TB: (*Ingaba usebenzisa amachiza okunyanga TB ngoku?*) (Is U tans op medikasie vir TB?): Yes/ No



If yes, date started: (*Ukuba kunjalo, uqale nini ukuwasebenzisa*) (As ja, wanneer begin) _____

14. Are you currently taking any other medication: (*ingaba akhona amanye amachiza ekungabe uyawasebenzisa ngoku angachazwanga apha*) (Neem U tans enige ander medikasie?) : Yes/ No

If yes, details. (*Khawusiphe inkcazelo*) (As ja, besonderhede) _____

Pt No: _____

15. Have you ever defaulted or stopped taking any medication on your own: Yes/
No

*(Wawukhe waziyekela ngokokwakho ukusebenzisa amachiza ungayalelwanga
ngu mongikazi okanye ugqira?) (Het u enige tyd self gestop om van die
medikasie te gebruik en later dan weer begin?)*

If yes, details *(Khawusiphe inkcazelo)*(As ja,
besonderhede)_____



XEROSTOMIA QUESTIONNAIRE

1. Does your mouth feel dry often? Yes / No
Uwuva umlomo wakho womile rhoqo?
Voel jou mond gereeld droog?
2. Do you regularly do things to keep your mouth moist? Yes / No
Ingaba uzifumana amaxesha amaninzi usenza amalinge okugcina umlomo wakho umanzi ukunqanda ukoma?
Doen jy gereeld iets om jou mond klam te hou?
3. Do you get out of bed at night to drink fluids? Yes / No
Ukhe uzifumane uvuka ebusuku ukuze ufumane into yokusela?
Staan jy op gedurende die nag om vloeistof te drink?
4. Does your mouth usually become dry when you speak? Yes / No
Ingaba umlomo wakho uyoma xa uthetha?
Droog jou mond uit wanneer jy praat?

XEROSTOMIA SCORE: _____

SALIVA RECORD

1. Unstimulated saliva (3 mins): Total weight: _____gms

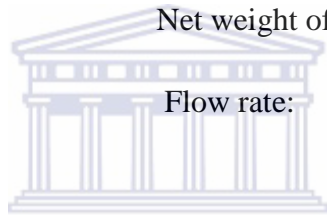
Net weight of saliva: _____gms

Flow rate: _____ml/min

2. Stimulated saliva (3 mins): Total weight: _____gms

Net weight of saliva: _____gms

Flow rate: _____ml/min



UNIVERSITY *of the*
WESTERN CAPE



Office of the Deputy Dean
Postgraduate Studies and Research
Faculty of Dentistry & WHO Collaborating Centre for Oral Health



UNIVERSITY OF THE WESTERN CAPE
Private Bag XI, Tygerberg 7505
Cape Town
SOUTH AFRICA

Date: 8th November 2013

For Attention: Dr AP Cherian
Oral Medicine & Periodontology

Dear Dr Cherian

STUDY PROJECT: Xerostomia and hyposalivation in HIV positive patients with and without HAART

PROJECT REGISTRATION NUMBER: 13/10/70

ETHICS: Approved

At a meeting of the Senate Research Committee held on Friday 8th November 2013 the above project was approved. This project is therefore now registered and you can proceed with the study. Please quote the above-mentioned project title and registration number in all further correspondence. Please carefully read the Standards and Guidance for Researchers below before carrying out your study.

Patients participating in a research project at the Tygerberg and Mitchells Plain Oral Health Centres will not be treated free of charge as the Provincial Administration of the Western Cape does not support research financially.

Due to the heavy workload auxiliary staff of the Oral Health Centres cannot offer assistance with research projects.

Yours sincerely

A handwritten signature in black ink, appearing to read 'S. Naidoo', written over a faint watermark of the University of the Western Cape building.

Professor Sureshni Naidoo



Eastern Cape Department of Health

Enquiries:	Zonwabele Merile	Tel No:	040 608 0830
Date:	09 th January 2014	Fax No:	043 542 1409
e-mail address:	zonwabele.merile@impilo.ecprov.gov.za		

Dear Dr AP Cherian

Re: Xerstomia and hyposalivation in HIV positive patients with and without HAART

The Department of Health would like to inform you that your application for conducting a research on the abovementioned topic has been approved based on the following conditions:

1. During your study, you will follow the submitted protocol with ethical approval and can only deviate from it after having a written approval from the Department of Health in writing.
2. You are advised to ensure, observe and respect the rights and culture of your research participants and maintain confidentiality of their identities and shall remove or not collect any information which can be used to link the participants.
3. The Department of Health expects you to provide a progress on your study every 3 months (from date you received this letter) in writing.
4. At the end of your study, you will be expected to send a full written report with your findings and implementable recommendations to the Epidemiological Research & Surveillance Management. You may be invited to the department to come and present your research findings with your implementable recommendations.
5. Your results on the Eastern Cape will not be presented anywhere unless you have shared them with the Department of Health as indicated above.

Your compliance in this regard will be highly appreciated.

DEPUTY DIRECTOR: EPIDEMIOLOGICAL RESEARCH & SURVEILLANCE MANAGEMENT



Ikamva etiqambileyo!