The Effectiveness of Combinatorial Antiretroviral Treatments on Treating Patients with HIV-1

Kyle Lee (kslee at sdsu.edu)

Abstract

A double-blind, placebo-controlled clinical trial was conducted to compare the use of a three-drug program that is comprised of Indinavir, Lamivudine, and Zidovudine or Stavudine with the two-drug program that is comprised of just Lamivudine and Zidovudine or Stavudine. Researchers are interested in identifying the set of predictors that influence treatment failure by developing Cox Proportional Hazard models in order to develop new treatment regimens for future studies. What makes HIV so difficult to destroy is the complexity and speed at which HIV replicates and so new combinatorial antiretroviral treatment regimens have been proposed since then.

Though rates of HIV are different across different races and sexes, it is unclear whether race and sex influence the development of AIDS or death in patients. We also consider the level of CD4 cells at the time of screening, IV drug use history, and as well as other factors as part of this analysis. Studying different sets of covariates would allow medical professionals and researchers to determine the most appropriate course of treatments as combinatorial antiretroviral treatment (CART) become more complex [1].

I. INTRODUCTION

According to UNAIDS, 36.7 million people around the world were diagnosed with human immunodeficiency virus (HIV) in 2016[2]. As of June 2017, 20.9 million people had access to antiretroviral treatment (ART) programs. However, there is currently no cure to completely eradicate it due to the presence of a latent reservoir of viral cells that remain undetectable when treatment options suppress the effects of HIV-1 until they become reactivated by a stop in treatment. Therefore, patients with HIV-1 can live full lives if they continue combinatorial antiretroviral treatment. Much research done today is not so much about complete eradication of the presence of HIV-1 in patients. Rather, it is about suppression of the viral replication process by analyzing the CD4 cell counts. Researchers are also interested in employing the best CART-based treatment regimes for patients with HIV-1 and how these drugs affect CD4 cell counts. However, CD4 cells are not the only covariate of interest. We are interested in considering how other factors such as age, race, and gender play a role in the development of AIDS, if there is any effect at all. We would like to consider what is the appropriate subset of variables that can explain most accurately the differences between the two treatment regimens.

II. METHODS

The study was a double-blind, placebo-controlled clinical trial that consisted of 1156 patients who were collected from 33 AIDS Clinical Trials Units and 7 National Hemophilia Foundations sites from January 29, 1996 to January 27, 1997 [3]. Patients were stratified by their CD4 cell counts which were collected at the time of screening. The two stratum were either that the patient had a CD4 concentration of 50 or fewer cells per cubic millimeter or 51 to 200 cells per cubic millimeter. Censoring for the purposes of our analysis is defined by the event that the patient developed acquired immune deficiency syndrome (AIDS) or death. A separate column in the data represents the event of death only which we do not analyze.

The statistical analysis performed here consists of Kaplan-Meier curve estimates, log-rank tests, Cox proportional hazards modeling, and diagnostic analysis. R was used to conduct the analysis along with several core packages such as $surv, stargazer, survminer$. Note that the dataset consists of only 1151 observations which the 5 missing observations are unaccounted for. Also, the study was terminated early due to "demonstration ... of clinical superiority"[4] of the three-drug regimen over the two-drug regimen.

III. RESULTS

A. Summary Statistics

There are 1151 total patients which $577(50.1\%)$ took the control regime and the other $574(49.9\%)$ took the treatment regime which includes Indinavir (IDV). Refer to **Table I** for the summary statistics of the continuous variables from the dataset.

TABLE I: Summary Statistics of Continuous Variables

Statistic	N	Mean	St. Dev.	Min	Max
time	1,151	230.183	89.877	1	364
karnof	1,151	91.303	7.726	70	100
cd4	1,151	86.460	70.054	0.000	392.000
priorzdv	1,151	30.419	29.218	3.000	312.000
age	1,151	38.647	8.811	15	73

We note that there were $96(8.3\%)$ censoring events and $1055(91.7\%)$ non-censoring events.

When CD4 cell counts were collected, $439(38.1\%)$ patients had 50 or fewer CD4 cells per cubic millimeter and $712(61.9\%)$ patients had 51 to 200 CD4 cells per cubic millimeter.

$951(82.6\%)$ of patients were male and $200(17.4\%)$ were female.

$596(51.8\%)$ of patients were white non-hispanic, $327(28.4\%)$ were black non-hispanic, $203(17.6\%)$ were hispanic, $14(1.2\%)$ were Asian or Pacific Islander, and $11(1.0\%)$ were American Indian or Alaskan Native.

Considering a patient's IV drug use history, $968(84.1\%)$ of patients never used IV drugs, $4(0.3\%)$ were using IV drugs at the time of the clinical trial, and $179(15.6\%)$ have used IV drugs at some point before the clinical trial.

Out of 1151 patients, $1116(97.0\%)$ do not have hemophilia and $35(3.0\%)$ do have hemophilia.

The distribution of the number of months prior to ZDV use is as follows: $929(80.8\%)$ between 2.69 and 50, $202(17.6\%)$ between 50 and 100, and $20(1.7\%)$ between 100 and 300.

The distribution of age before treatment began is as follows: 15.7% were between 15 and 30, 65.1% were between 31 and 45, 17.3% were between 46 and 60, and 1.8% were between 61 and 75.

The distribution of the baseline CD4 cells is as follows: $439(38.1\%)$ were between 0 and 50, $247(21.5\%)$ were between 50 and 100, $363(31.5\%)$ were between 100 and 200, $82(7.1\%)$ were between 200 and 400.

The Karnofsky Performance Scale was used to evaluate the likelihood of survival across different treatment regimes and to assess the patient's normal functioning capabilities. A Karnofsky score of 100 indicated that AIDS were not present while a score of 70 indicated severe impairment. So, out of 1151 patients, $32(2.8\%)$ scored a 70, $182(15.8\%)$ scored an 80, $541(47.0\%)$ scored a 90, and $396(34.4\%)$ scored a 100

Plots of the distributions of the continuous variables are omitted due to space constraints.

B. Kaplan Meier Model

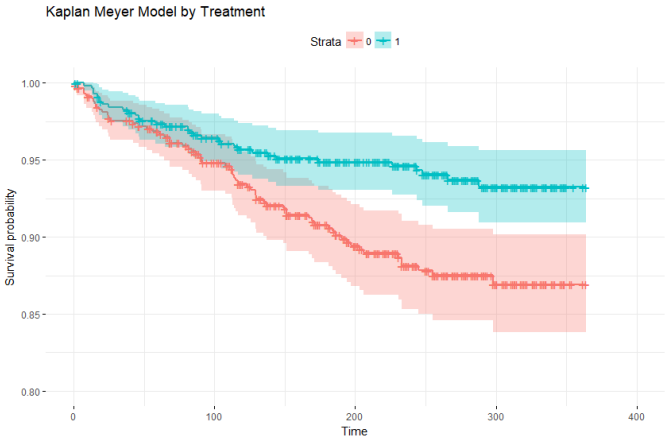

Fig. 1: Kaplan Meier Curve

The Kaplan Meier curves were used to estimate the survival likelihood functions of patients between the two treatment groups. In **Figure 1**, the 95% confidence intervals between the two curves do not overlap starting from 200 days which suggest that the treatment regimen with IDV decreases the likelihood AIDS diagnosis or death after six to seven months of treatment.

C. Variable Selection

Backward selection was used in order to select the best set of predictors for a Cox proportional hazards model. This is done by starting with all the predictors in the dataset. Then, the predictor with the highest p-value is removed while checking that the predictor is moderately significant. Afterwards, a new model is created and the process is repeated until the last model consists mainly of statistically significant predictors.

In **Table II**, the p-values are listed for each variable and the empty cell for a predictor means that predictor was removed from the model. Because race and the use of iv drugs in the past are categorical variables, they have to be factored accordingly and weighted against the set of non-factored covariates. Thus, $hemophil$, $strat2$, $priorzdv$,sex, and $race$ were removed in that order. Race had to be considered as an average for removal. Also, the stepwise variable selection feature from the $surv$ package was used to confirm this set of predictors. There was reason to believe that sex and $race$ had an interaction since some diseases vary across sex and $race$ but it was deemed statistically insignificant.

TABLE II: P values of Model Selection

	Dependent variable: time					
	(1)	(2)	(3)	(4)	(5)	(6)
tx	$p = 0.003^{***}$	$p = 0.003^{***}$	$p = 0.003^{***}$	$p = 0.003^{***}$	$p = 0.003^{***}$	$p = 0.002^{***}$
strat2	$p = 0.857$	$p = 0.858$				
sex	$p = 0.490$	$p = 0.495$	$p = 0.494$	$p = 0.498$		
factor(raceth)2	$p = 0.190$	$p = 0.189$	$p = 0.191$	$p = 0.198$	$p = 0.231$	
factor(raceth)3	$p = 0.721$	$p = 0.722$	$p = 0.715$	$p = 0.705$	$p = 0.673$	
factor(raceth)4	$p = 0.173$	$p = 0.174$	$p = 0.173$	$p = 0.174$	$p = 0.169$	
factor(raceth)5	$p = 0.810$	$p = 0.789$	$p = 0.790$	$p = 0.789$	$p = 0.804$	
factor(ivdrug)2	$p = 0.463$	$p = 0.464$	$p = 0.476$	$p = 0.487$	$p = 0.489$	$p = 0.465$
factor(ivdrug)3	$p = 0.081^{*}$	$p = 0.080^{*}$	$p = 0.081^{*}$	$p = 0.081^{*}$	$p = 0.083^{*}$	$p = 0.068^{*}$
hemophil	$p = 0.873$					
karnof	$p = 0.00001^{***}$	$p = 0.00001^{***}$	$p = 0.00001^{***}$	$p = 0.00001^{***}$	$p = 0.00001^{***}$	$p = 0.00001^{***}$
cd4	$p = 0.0003^{***}$	$p = 0.0002^{***}$	$p = 0.000^{***}$	$p = 0.000^{***}$	$p = 0.000^{***}$	$p = 0.000^{***}$
priorzdv	$p = 0.774$	$p = 0.789$	$p = 0.797$			
age	$p = 0.040^{**}$	$p = 0.041^{**}$	$p = 0.042^{**}$	$p = 0.043^{**}$	$p = 0.050^{**}$	$p = 0.060^{*}$

D. Testing for Functional Forms

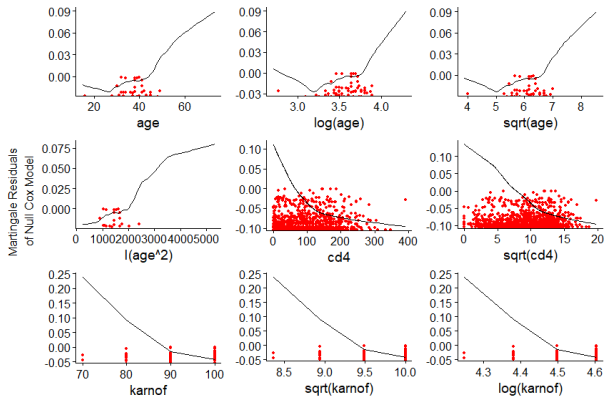

Fig. 2: Martingale Residuals

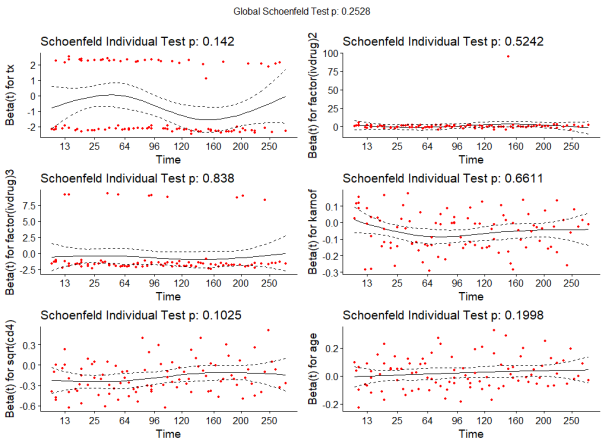

Fig. 3: Schoenfeld residuals

Then, in order to find the appropriate functional forms for the terms age, $cd4$, and $karnof$, the Martingale residuals were plotted in **Figure 2**. Note that the Martingale residual corresponding to age is linear so this suggests that the functional form for age should remain as it is. However, age^2 seems to be linear as well so we consider $age + age^2$ as a functional form. By comparing the models after stratifying for treatment with age and $age + age^2$ using a log likelihood test, we find that age is sufficient ($p = .623$). Therefore, age^2 was dropped from the final model.

However, for cd, the Martingale plot suggests that the functional form is $\sqrt{cd4}$ since the residual LOESS curve is mostly linear.

E. Testing for Proportional Hazards Assumption

For each covariate, the Schoenfeld residuals are plotted in **Figure 3** in order to test the proportional hazards assumption. Therefore, for each covariate, a valid Cox model would demonstrate that the Schoenfeld residuals are horizontal across time and centered at 0. Note that tx shows a systematic departure from a horizontal line which suggests that the proportional hazards assumption is violated. Although it can be argued that $\sqrt{cd4}$ violates the proportional hazards assumption by not being centered at 0, tx is non-linear and not centered at 0.

We also confirm this by observing in **Table III** that the p-value of tx is 0.142. However, the global p-value is $.253 > .10$ which indicates that the proportional hazards assumption is satisfied. This seems contradictory so we proceed to stratify treatment.

F. Outliers

There are several large values for $\sqrt{cd4}$ (see **Figure 4**) and age (most of the residual plots are omitted due

TABLE III: P-values for testing PH-assumption

	rho	chisq	p
tx	-0.1507	2.1566	0.142
factor(ivdrug)2	0.0663	0.4057	0.524
factor(ivdrug)3	-0.0209	0.0418	0.838
karnof	-0.0458	0.1922	0.661
sqrt(cd4)	0.2013	2.6661	0.103
age	0.1290	1.6437	0.200
GLOBAL	NA	7.8042	0.253

TABLE V: Final Model

	coef	exp(coef)	Conf.level	p-value
factor(ivdrug)2	0.820	2.270278	(0.306, 16.831)	p = 0.423
factor(ivdrug)3	0.5436665	−0.609	(0.282, 1.050)	p = 0.070
karnof	0.9473023	−0.054	(0.925, 0.970)	p = 0.00001
sqrt(cd4)	0.8337657	−0.182	(0.787, 0.884)	p = 0.000
age	1.021049	0.021	(0.999, 1.044)	p = 0.064

Note: *p<0.1; **p<0.05; ***p<0.01

to space) but it is understandable that the counts of the baseline cd can be extremely large. Some patients appeared on several plots and are denoted in **Table IV**. What patients 495, 1043, and 641 have in common is that they are older and they also scored low on the Karnofsky scale. This is probably due to HIV-1 having already compromised their immune systems at such a later stage in life. Therefore, it would be unwise to disregard any patient data.

A remark has to be made that the labels on the residuals may have different patient identification numbers since 5 patients are missing.

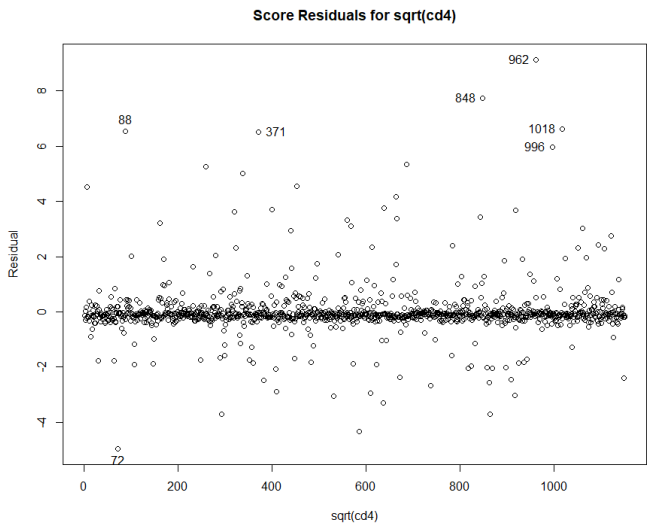

Fig. 4: Score Residuals

TABLE IV: Identified Outliers

id	time	censor	tx	txgrp	strat2	ivdrug	karnof	cd4	age
495	144	1	1	2	1	2	70	57	56
1,043	221	0	0	1	1	2	80	71.500	47
641	290	0	0	1	0	1	70	6	58

IV. Discussion

Table V lists the coefficients as well as 95% confidence intervals for the exponentiated coefficients. The p-value for $factor(ivdrug)2$ is not significant with a hazard ratio $HR = 2.27$ which indicates a weak relationship between patients who are currently taking IV drugs versus never taking IV drugs and increased risk of death while stratifying for treatment. However, the p-value for $factor(ivdrug)3$ is statistically significant with a hazard ratio of .543 which indicates a strong relationship between patients who previously have taken IV drugs and decreased risk of death while stratifying for treatment.

Holding the other covariates constant and stratifying for treatment, every additional unit of the Karnofsky score reduces the hazard by a factor of .947 or by 5.3%. Also, note that there is a statistically strong relationship between the patient's CD4 cell count and decreased hazard risk. Holding other covariates constant, an additional unit of CD4 reduces the hazard by a factor of .833 or by 17.7%. Note that 1 is not in the 95% confidence interval for the CD4 cell count which also shows a positive relationship in decreasing hazard risk.

Since the 95% confidence interval for the hazard ratio of age includes 1, age makes a smaller contribution to the difference in HR compared to CD4 cell count while keeping other covariates constant. For every additional year of age, the hazard ratio in acquiring AIDS or death increases by a factor of 1.02 or by 2%.

Therefore, patients who have previously taken IV drugs, have higher Karnovsky scores, or have higher CD4 cell counts have a decreased risk of being diagnosed with AIDS or death. Older patients have an increased risk of censoring as well. **Figure 5** shows the final Cox Proportional Hazards model stratifying for treatment. Note that the treatment group that includes indinavir has a higher likelihood of survival compared to the treatment group without indinavir.

Several limitations to this study have to be considered. We stratified according to the treatment in order to analyze the effectiveness of the treatment regimes on the rate of survival across time. The reason it was stratified was because the treatment coefficient seemed to be a function of time. We also note that there are 5

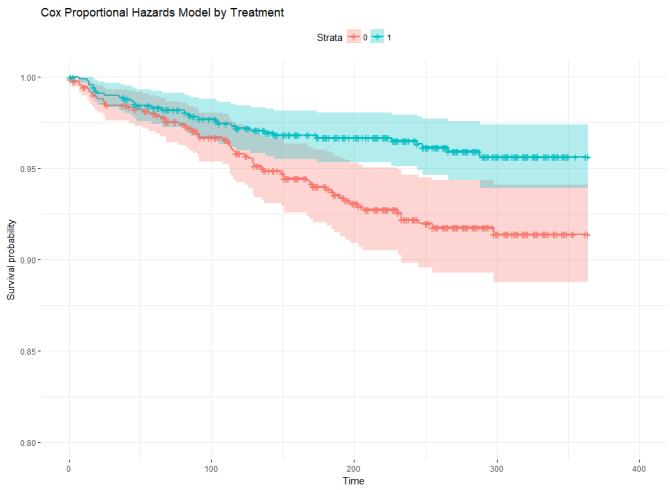

Fig. 5: Cox-PH Model

missing observations as the original data was supposed to contain 1156 patients.

My findings suggest that the treatment regime that consisted of indinavir, zidovudine, and laivudine statistically increased the likelihood of survival of patients with AIDS compared to the standard two-part treatment regime of zidovudine or stavudine with lamivudine. Hammer et. al stratified according to the baseline CD4 cell count and had similar conclusions regarding the effectiveness of the three-part treatment. Potential covariates such as the number of months prior to ZDV use and hemophilia are insignificant in determining the treatment difference. Therefore, we see that the use of combinatorial anti-retroviral treatment-based regimens will benefit patients contracted with HIV-1.

REFERENCES

[1] J. Maenza and C. Flexner, "Combination antiretroviral therapy for hiv infection." *Am Fam Physician*, vol. 57, no. 11, p. 27892798, Jun 1998.

[2] "Fact sheet - latest statistics on the status of the aids epidemic." [Online]. Available: http://www.unaids.org/en/resources/fact-sheet

[3] S. Hammer, K. Squires, M. Hughes, J. Grimes, L. Demeter, J. Currier, J. RR Eron, J. Feinberg, H. JR Balfour, L. Deyton, J. Chodakewitz, M. Fischl, J. Phair, L. Pedneault, B.-Y. Nguyen, and J. Cook, "A controlled trial of two nucleoside analogues plus indinavir in persons with human immunodeficiency virus infection and cd4 cell counts of 200 per cubic millimeter or less. aids clinical trials group 320 study team." vol. 337, 01 1997.

[4] J. Levin, "Actg 320, the controversial clinical endpoint study of crixivan, has been stopped due to convincing data demonstrating clinical superiority," Aug 1998. [Online]. Available: http://www.natap.org/1998/drug/320.html