Supplementary Appendix

This appendix has been provided by the authors to give readers additional information about their work.

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SUPPLEMENTARY APPENDIX

Timing of Antiretroviral Therapy after Diagnosis of Cryptococcal Meningitis.

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Authorship Contributions:

DRB, DBM, AK, ENJ, and PRB conceived of the trial. DRB wrote the clinical protocol with statistical assistance from TLB and input from DBM, YCM, and NIAID personnel. TLB and MAR generated the randomization sequence. KHH and MAR conducted statistical analysis, with the analysis being guaranteed by KHH. Site principal investigators were: DBM, CM, and GM. Clinical data were collected in Kampala by AM, HWN, DAW, RR, JR, MWL; in Mbarara by KT; in Cape Town by CS and FT. Microbiology data were collected by CS, KT, and verified by KN. DRB wrote the first draft of the manuscript and is overall study guarantor. All authors reviewed, revised, and approved the final version of the manuscript.

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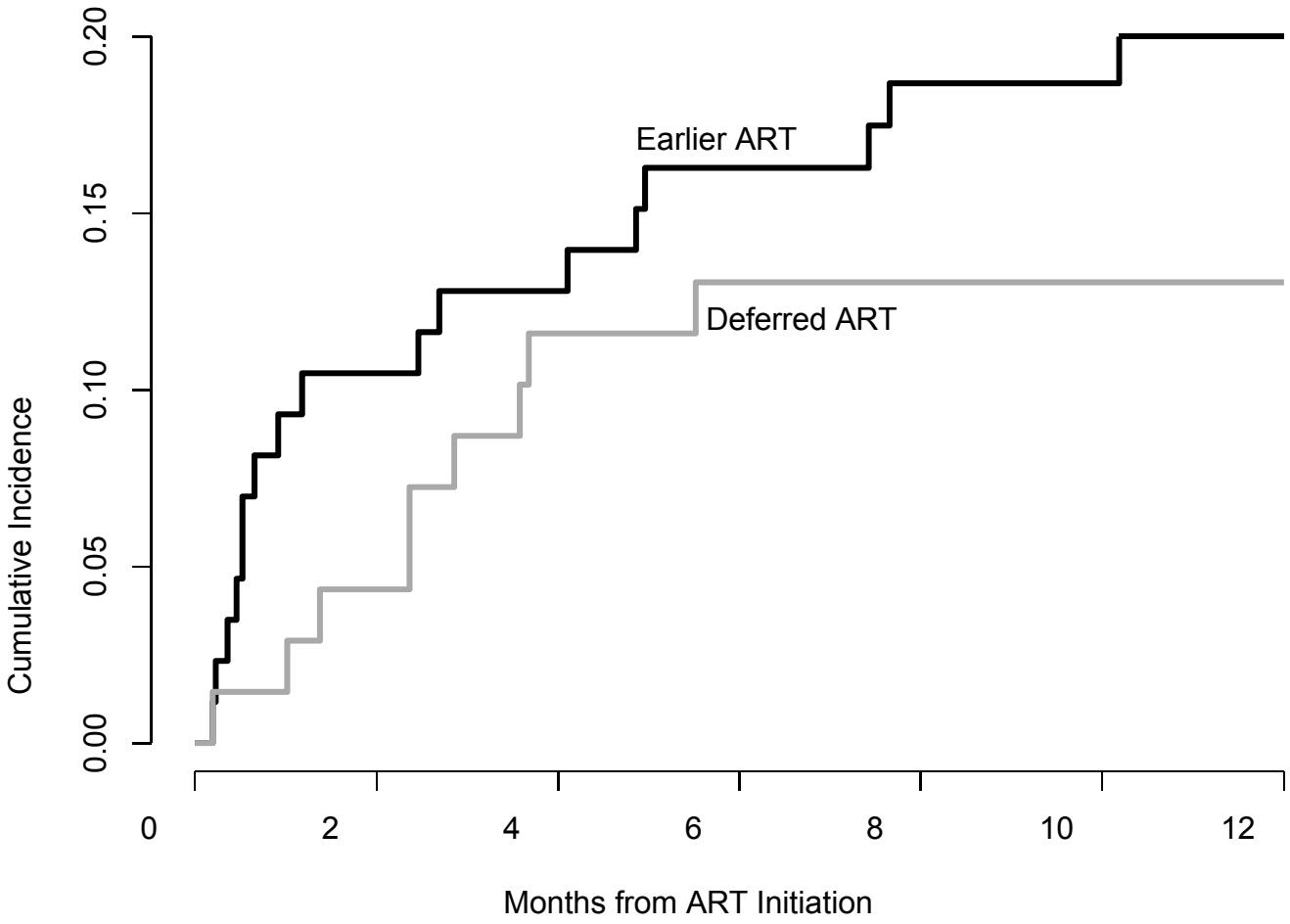
Methods:

External Adjudication Committee Procedures:

All deaths had a primary cause of death determined, at the time of death, by the COAT study physician(s) attending to the research participant. A panel of three independent clinicians, with expertise in HIV and cryptococcosis, retrospectively adjudicated deaths for agreement. Digital images of the original source documents were provided to the committee for their review. A consensus of 2 of 3 adjudicators was needed for the cause of death. There was 88% agreement between the study physicians and adjudicator committee for cryptococcal-related deaths, and 79% agreement with all causes of death. Disagreements with the original study physician diagnosis of cause of death were most often based on the availability to the committee of all final microbiology and postmortem data (specifically, final postmortem histopathology results); or often subtle differences in the differentiation between the proximal cause of death versus contributory causes of death.

Suspected immune reconstitution inflammatory syndrome (IRIS) events were also adjudicated per the International Network for the Study of HIV-associated IRIS (INSHI) published cryptococcal-IRIS case definition, requiring consensus of all three adjudicators. Any major clinical event or re-hospitalization was adjudicated by the committee. Among the earlier ART arm, 44 major clinical events occurred among 30 subjects. In the deferred ART arm, 45 major clinical events occurred among 24 subjects. IRIS was adjudicated as definite/probable IRIS (n=6 in both arms), possible IRIS (n=11 with earlier ART vs. n=3 with deferred ART), or not cryptococcal-IRIS.

Figure S1. Cumulative Incidence of Cryptococcal IRIS Events



Number at Risk:						
Earlier:	87	42	38	35	32	29
Deferred:	69	61	54	53	52	50

Figure S1 displays the cumulative incidence from the time of antiretroviral therapy (ART) initiation to time of first cryptococcal-related paradoxical immune reconstitution inflammatory syndrome (IRIS) as externally adjudicated by a three physician masked panel. Within the COAT trial randomization ART was initiated in the earlier ART group between 7-13 days after cryptococcal meningitis diagnosis and in the deferred ART group at 5 $\pm$1 week from diagnosis. In total, 17 (20%) IRIS events occurred in the earlier ART arm, and 9 (13%) IRIS events occurred in the deferred ART arm within 46 weeks from randomization. The number at risk indicates those persons alive on ART without an IRIS episode. Persons who never initiated ART are censored from the initial number at risk.

Figure S2a. CD4 T cell response over 26 weeks

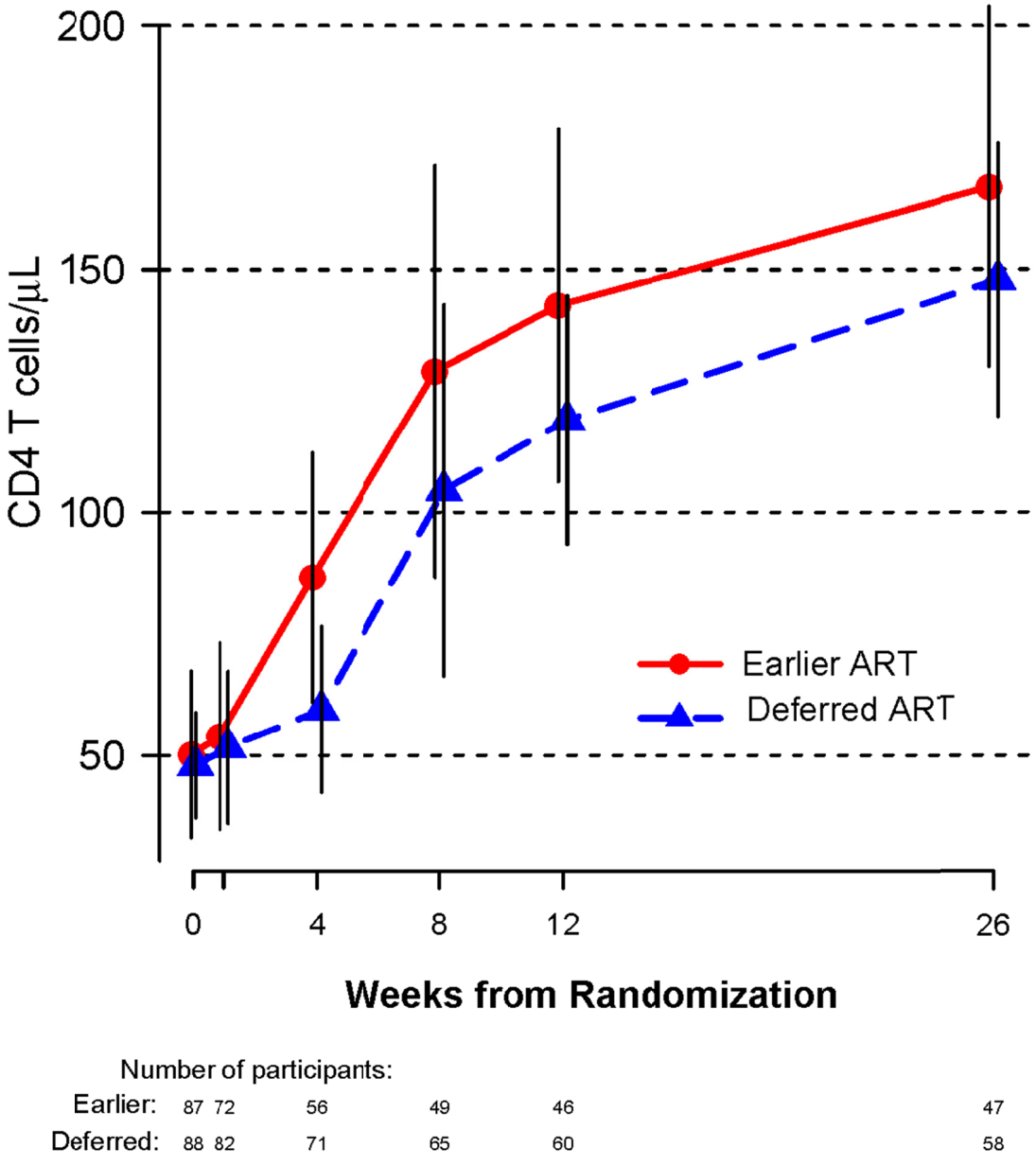


Figure S2a displays the mean (95% CI) response in CD4 T cell count/μL over 26 weeks from randomization by trial arm. Red solid line displays the earlier ART group; Blue dashed line displays the deferred ART group. At 26 weeks, there was no statistical difference in CD4 T cell counts (P=0.40).

Randomization occurred at a median of 8 days (IQR, 7-8) of antifungal therapy. Earlier ART was initiated a median of 1 day (IQR, 0-1) after randomization. Deferred ART was initiated a median of 28 days (IQR, 27-31) after randomization (i.e. approximately 5 weeks after cryptococcal meningitis diagnosis).

Figure S2b. HIV-1 Plasma Viral Load response over 26 weeks

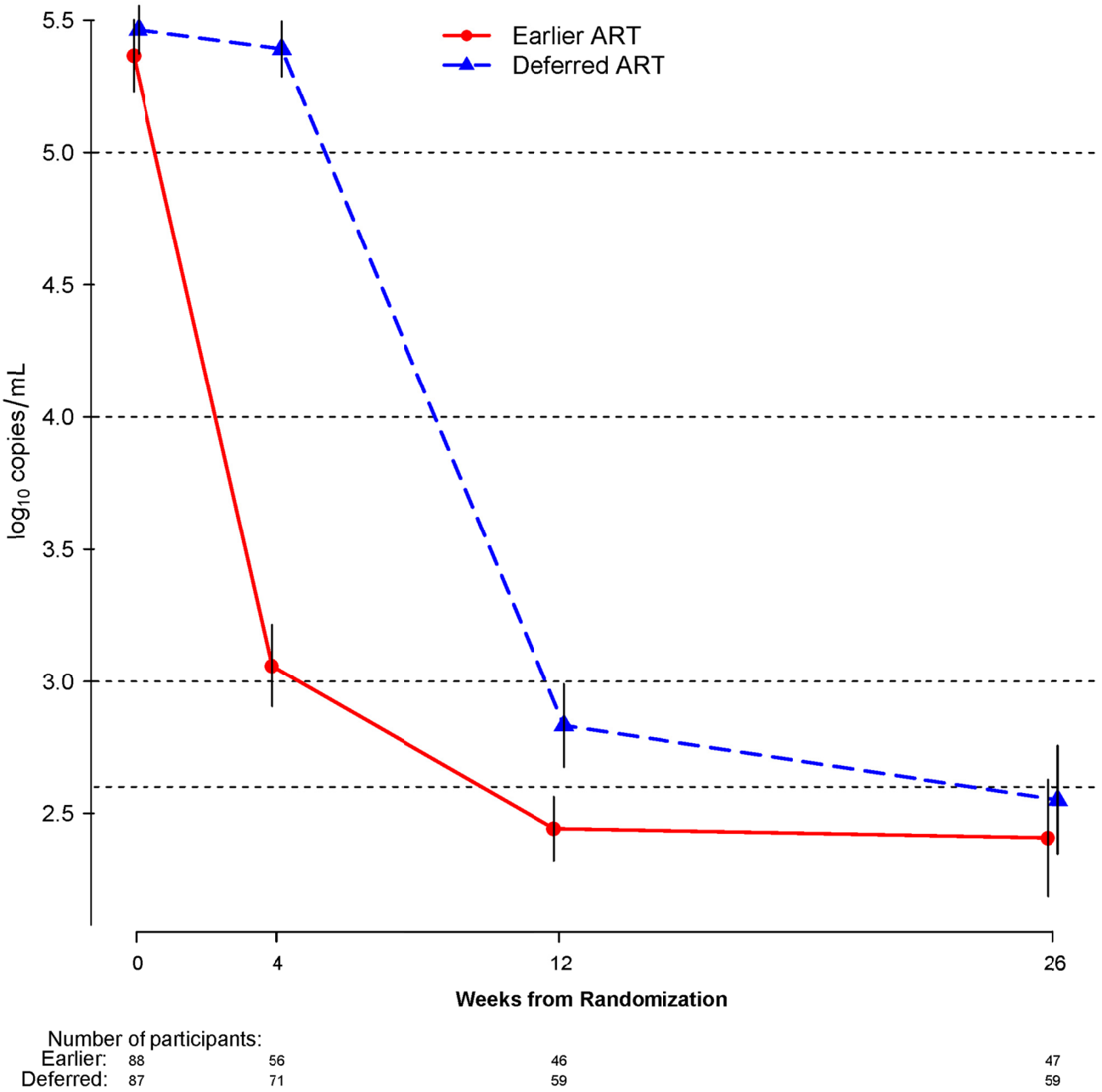
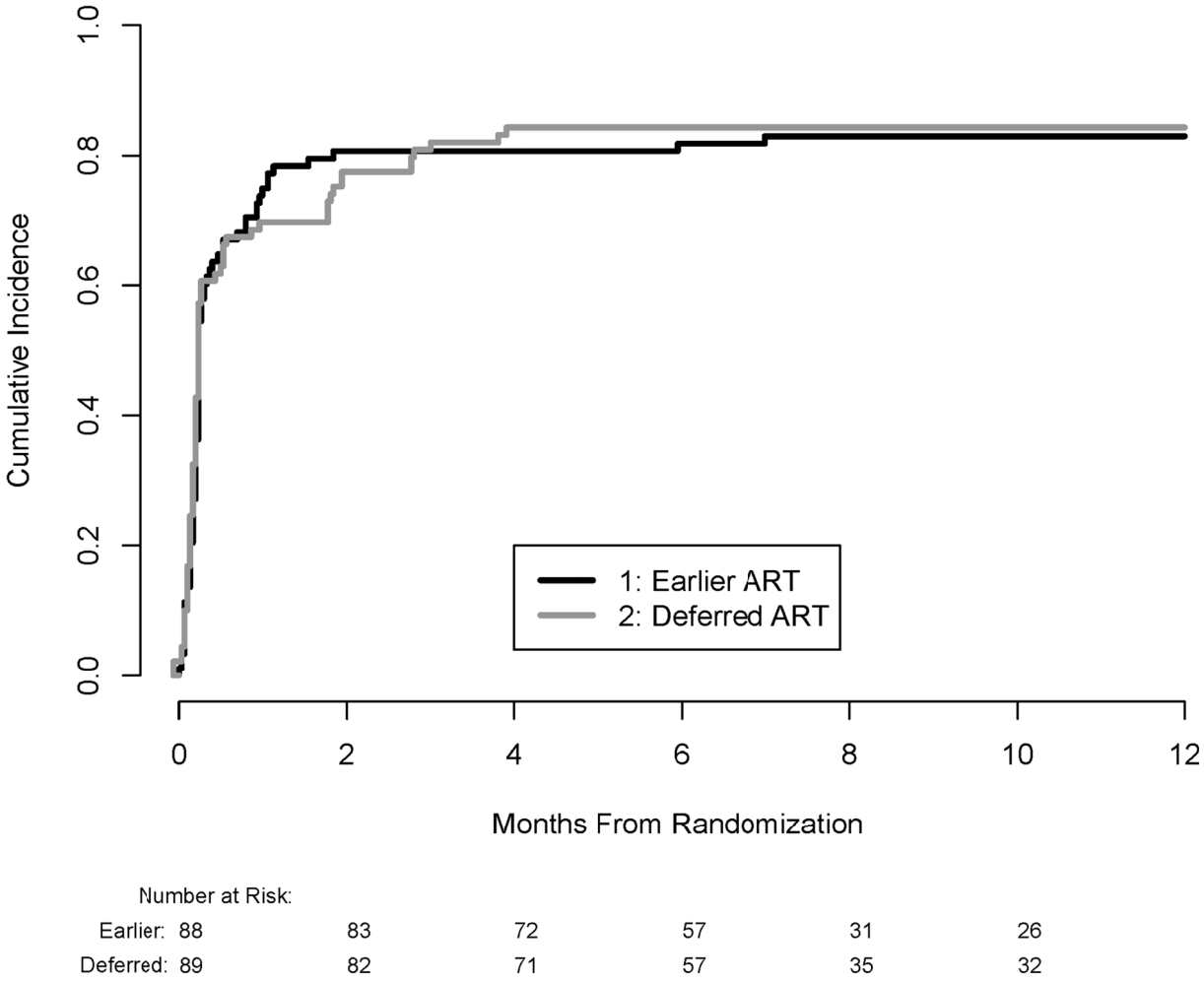


Figure S2b displays the mean (95% CI) plasma HIV-1 viral load (log_{10} copies/mL) response over 26 weeks from COAT trial randomization. Red solid line displays the earlier ART group; Blue dashed line displays the deferred ART group. At 26 weeks, there was no statistical difference in HIV viral loads (P=0.34). At 26 weeks, 91% (43/47) of the earlier ART group had viral loads <400 copies/mL compared with 84% (49/59) of the deferred ART grroup (P=0.26). Detectable viral loads between 400-1000 copies/mL occurred in 4.3% (2/47) of the earlier ART group and 10% (6/59) of the deferred ART group. Potential virologic failure with viral loads >1000 copies/mL were present in 4.3% (2/47) of the earlier ART group and 6.8% (4/59) of the deferred ART group. Reported episodes of ART treatment interruption for >3 days did not differ between earlier ART initiation (5.7% (5/87)) and deferred ART initiation (1.4% (1/69); P=0.23).

Figure S3a. Cumulative Incidence of Grade 3−5 Adverse Events



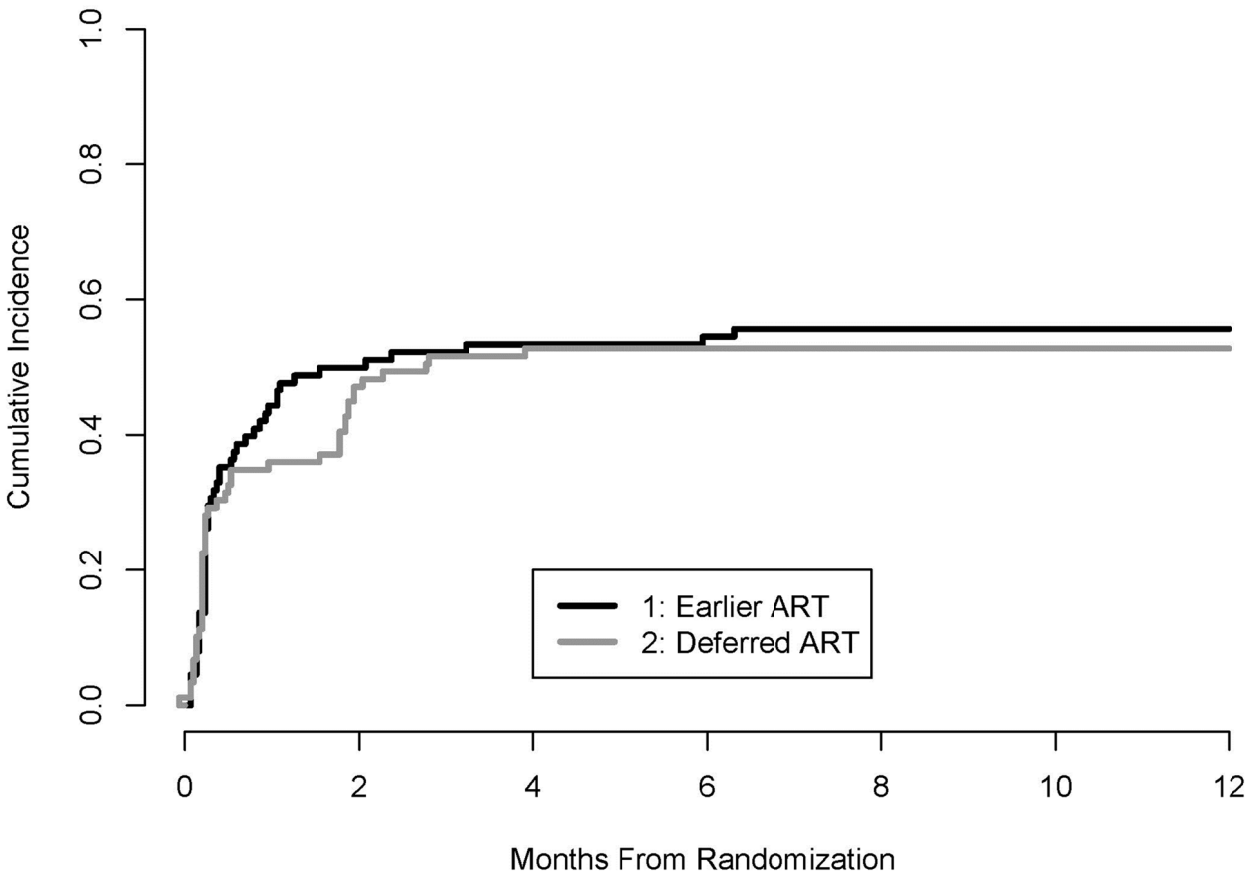
Number at Risk:						
Earlier:	88	83	72	57	31	26
Deferred:	89	82	71	57	35	32

Figure S3a displays the time from randomization to a first Grade 3-5 adverse event by trial arm as defined by the National Institute of Allergy and Infectious Diseases (NIAID) Division of AIDS (DAIDS) Toxicity Grading Scale, version 2009. The distribution of adverse events by body system was:

Distribution by Body System	Earlier ART	Deferred ART
Hematology	99 (47.8%)	128 (55.9%)
Chemistries	65 (31.4%)	53 (23.1%)
Infection	13 (6.3%)	26 (11.4%)
Neurological	11 (5.3%)	5 (2.2%)
Gastrointestinal	6 (2.9%)	8 (3.5%)
Cardiovascular	6 (2.9%)	5 (2.2%)
Systemic	4 (1.9%)	2 (0.9%)
Skin, dermatological	2 (1.0%)	1 (0.4%)
Respiratory	1 (0.5%)	1 (0.4%)
Overall (Number of events)	207 (100%)	229 (100%)

The most commons adverse events included anemia (n=88), neutropenia (n=54), leukopenia (n=24), hyponatremia (n=21), hypokalemia (n=23), and elevated creatinine (n=18).

Figure S3b. Cumulative Incidence of Grade 4 or 5 Adverse Events



Number at Risk:						
Earlier:	88	86	80	69	54	49
Deferred:	89	85	80	76	61	59

Figure S3b displays the time from randomization to a Grade 4 (life threatening) or Grade 5 (death) adverse event by trial arm as defined by the National Institute of Allergy and Infectious Diseases (NIAID) Division of AIDS (DAIDS) Toxicity Grading Scale, version 2009. Grade 5 events occurred in 14 (15.9%) participants randomized to earlier ART and 11 (12.3%) participants randomized to deferred ART. Grade 5 events do not include deaths due to cryptococcal meningitis.

Table S1: Adverse Event Listing		Grade 3-5 Adverse Events			Grade 4 and 5 Adverse Events	
Body System	**MedDRA Term**	**Earlier ART**	**Deferred ART**		**Earlier ART**	**Deferred ART**
Cardiovascular	Cardiac arrhythmias	1	0		1	0
	DVT	4	1		1	0
	Pulmonary embolus	0	1		0	1
	Tachycardia	0	1		0	0
Chemistries	Alanine aminotransferase increased	2	1		0	0
	Aspartate aminotransferase increased	5	4		2	1
	Blood alkaline phosphatase increased	2	0		1	0
	Blood bicarbonate decreased	6	5		1	2
	Blood bilirubin increased	1	0		0	0
	Blood sodium decreased	14	7		8	3
	Blood sodium increased	0	2		0	1
	Hyperkalemia	2	0		2	0
	Potassium serum decreased	15	8		4	1
	Serum creatinine increased	8	10		2	2
Gastrointestinal	Decreased appetite	1	1		0	0
	Diarrhea	3	2		1	1
	Nausea and vomiting	2	2		1	1
Hematology	Anemia	41	47		23	31
	Leukopenia	11	13		6	3
	Neutropenia	23	31		12	12
	Thrombocytopenia	4	13		1	3
Infection	Abdominal pain localized	0	1		0	0
	Acinetobacter bacteremia	0	1		0	1
	Acute pneumonia	1	0		0	0
	Bronchopneumonia	1	0		0	0
	Burkholderia cepacia complex	0	1		0	0
	Cystitis, Klebsiella	1	0		1	0
	Disseminated tuberculosis	0	2		0	2
	Klebsiella sepsis	1	1		1	1
	Pulmonary tuberculosis	0	1		0	0
	Salmonella bacteremia	0	1		0	0
	Salmonella sepsis	0	1		0	0
	Sepsis	1	4		1	3
	Septicemia due to pseudomonas	1	1		1	0
	Staphylococcus aureus septicemia	1	0		1	0
	Tuberculosis	0	1		0	1
Neurological	Acute mental status changes	5	3		4	0
	Intracranial pressure increased	3	0		0	0
	Muscle weakness right-sided	2	0		2	0
	Sensory neuropathy	1	0		0	0
Respiratory	Dyspnea	1	1		1	0
Dermatological	Erythroderma	1	0		0	0
	Rash - morbilliform	0	1		0	1
Systemic	HIV wasting syndrome	0	1		0	0
	Pyrexia	1	0		0	0