Vedolizumab treatment in antiretroviral drug treated chronic HIV infection

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HAVARTI

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Protocol Signature Page

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Investigator's Agreement:

My signature below confirms that I have reviewed and approved this protocol and agree to carry out its terms in accordance with applicable regulations and to follow ICH GCP guidelines for good clinical practice.

Investigator name (please print): _____

Investigator Signature: _____

Date: _____

The HAVARTI Trial: Vedolizumab treatment in antiretroviral drug treated chronic HIV infection, followed by analytical treatment interruption (HIV-ART-vedolizumab-ATI).

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1. Background

HIV/AIDS originally emerged as a zoonotic infection, and its introduction resulted in sustained human transmission over the last century. The source of the zoonosis was crossover of simian immunodeficiency virus (SIV_{cpz}) from chimpanzees to humans in central west Africa, with sustained epidemic amplification by iatrogenic parenteral and person-to-person sexual transmission first within the African continent, and ultimately around the world. Pandemic dissemination has included several HIV clades or types as well as recombinants (Pepin, 2011). Cumulative human infections are increasing at two million annually, with 38 million people estimated to be living with HIV as of 2015 (UNAIDS, 2016). In the first 35 years of the pandemic there have been approximately 35 million deaths due to AIDS. Present mortality is estimated at one million per year (HIV/AIDS, 2016; Pepin, 2011).

HIV is a chronic active infection of lymphocytes resulting in a progressive and profound depletion of this cell subset and ultimately in the Acquired Immune Deficiency Syndrome (AIDS). Chronic immune activation in the presence of sustained plasma viremia results in CD4 T cell deficiency. Opportunistic infections and malignancies accelerate the progressive immunodeficiency of HIV infection, which without treatment leads to symptomatic illness and death over a period of about 10 years from initial infection. Generally, there appears to be a simple direct relationship between the density of plasma viremia and the development of immunodeficiency after primary infection (Arnaout et al., 1999). There are a few individuals with HLA- or co-receptor variations, or unknown factors, who have low or absent viremia and who are slow- or non-progressors.

Research in immunopathogenesis has not precisely elucidated the mechanism(s) by which HIV causes AIDS. The infection of susceptible lymphocytes and macrophages begins with the binding of a free or cell-associated virion to a cell surface receptor (CD4). This complex interacts with a cell surface co-receptor (CXCR4, or CCR5) resulting in subsequent viral entry to the cell. After intracellular reverse transcription of HIV RNA, pro-viral DNA is integrated into the host cell's chromosomal DNA. Newly infected cells may undergo lytic productive infection upon activation (lymphocytes), or undergo chronic productive infection (macrophages). Additionally, the infected lymphocyte may remain dormant with latent HIV infection and as it undergoes replication, so too does the integrated pro-viral DNA, expanding the pool of latently infected cells, which, if activated will undergo lytic infection. Primary infection disproportionately affects and depletes gut-associated lymphoid tissue (GALT) but all tissues, including some relatively sequestered compartments such as the central nervous system, experience monocytoid and lymphoid infection. Immune escape, viral latency, persistent active infection, and ultimate viremic escape of partial immune suppression are the natural course of HIV. The initial immune response to primary infection with HIV leads to a significant reduction in viremia with an incompletely neutralizing specific immune response. An unstable equilibrium between active infection and specific immunity emerges following this. Lymphocyte activation against emerging specific immune responses (to HIV or other agents) is targeted and deleted or

attenuated by lymphocytolytic HIV infection. In the late stages of progressive HIV immunodeficiency there are markers of immunological exhaustion or immunosenescence, and there is generalized lymphocyte depletion.

Continuous anti-retroviral drug treatment (ART) suppresses viremia and allows some degree of immune healing so as to permit near-normal health and life expectancy. However, interruption of ART is uniformly followed by rebound of viremia and progressive immunodeficiency. This effect has been established previously in clinical and research practice. ART interruption is a common clinical event in long-term ART for many patients. 'Structured treatment interruption' (STI) has been a tactic in past clinical trials of HIV therapy and has been adopted in immunotherapeutic trials as an evaluation of immunologically mediated treatment strategies. Previously, immunotherapies including therapeutic vaccination have been followed by planned analytical treatment interruption (ATI) in order to assess the impact of this treatment strategy on infection. These treatments have shown some biological effects, including changes in the timing and extent of recurrent plasma viremia, without clinically significant immune-mediated remission. ATI after experimental interventions added to ART with monitoring for rebound and recurrent plasma viremia (PVL) remains the gold-standard in investigation of novel therapies, and has been shown to be a safe test for assessment of functional or sterilizing 'cure' of HIV infection (Graziani & Angel, 2015) (Routy, Boulassel, Nicolette, & Jacobson, 2012).

Counter to past expectations for cell-mediated immunity, broadly neutralizing monoclonal antibodies (nmAb) have been developed which show anti-HIV activity *in vivo* in reducing viremia (Caskey, Klein, & Nussenzweig, 2016), and in slightly reducing rebound and recurrent viremia following ATI (Bar et al., 2016). Monoclonal antibody (mAb) therapies against host cellular markers such as HIV co-receptors have been put forward as potential strategy in immunological treatment of HIV infection (Pace & Markowitz, 2015). If activated lymphocytes could be protected from cytolytic HIV infection in the presence of concurrent active HIV infection of other cells and tissues, then potent specific immunity might emerge and confer lasting immunological remission.

Immunological remission or functional cure of HIV infection must address the problems of latency in lymphoid cells, chronic infection of monocytoid cells, and tissue compartmentalization of infection from systemic immunity and sequestration from anti-HIV drug penetration. Reactivation after brief (therapeutic vaccine trials) or long (case report, Mississippi baby) remission is observed, which might be due to weak or waning systemic immunity and emergence form latency, or emergence from reservoir compartments like the central nervous system as observed in macaques (Gama et al., 2017). Past work on the emergence of disparate CNS virological breakthrough from ART identified the CNS as a virological compartment and/or pharmacological sequestrum (Khaliq, Gallicano, Venance, Kravcik, & Cameron, 2000). Thus, those with a systemic remission of viremia should be carefully evaluated for CNS remission (Spector & Rappaport, 2017)in anticipation of incompleteness or transience of remission of PVL or progressive disease.

This year, an unexpected finding of sustained remission of plasma viremia level (PVL) to below quantifiable assay limits in a rhesus macaque SIV infection model was reported (Byrareddy et al., 2016). The envelope of SIV (and HIV) appear to bind alpha4-beta7 integrin, a lymphocyte homing receptor for trafficking to gut mucosal-associated lymphoid tissue that was the target of monoclonal antibody therapy in this study. This anti-α4β7 mAb was administered serially every three weeks at 50 mg/kg for 8 doses to a series of acutely infected animals on established ART and was followed by ATI and subsequent discontinuation of mAb administration. This was followed by a diminished rebound of PVL and then by sustained remission of PVL and GALT associated viral loads for a variable period lasting up to 24 months with persistence of SIV latency as demonstrated by pro-viral DNA recovery from GALT tissue. The mechanism of this unexpected post-treatment effect is unknown. In human HIV infection, this effect sustained over time with continued remission of progressive immunodeficiency would meet definitional criteria of "functional cure".

Two human therapeutic monoclonal antibodies against the α4β7 integrin are currently available: Vedolizumab and Ertolizumab. Humanized monoclonal antibody against the β7 subunit of the heterodimeric integrins α4β7 and αEβ7 (ertolizumab, RhuMAb β7) is the newest such agent (Anand et al., 2014; Armuzzi & Felice). Humanized anti-α4β7 integrin mAb (vedolizumab, Entyvio) is a licensed treatment for inflammatory bowel disease with a safety record of 4811 person-years observation, in which 2830 patients received vedolizumab from one to 1977 (median 328) days during which there was no increased risk of any infections (<1% overall for TB or other serious infection, and no reported case of progressive multifocal leukoencephalopathy), compared to placebo and standard non-biological therapy (Colombel et al., 2016). In the macaque study above, serial mAb dosing was 50 mg/kg. In humans, dosing of vedolizumab at 0.2 mg/kg (~14 mg/dose) to 10 mg/kg (~700 mg/dose) resulted in saturation of the α4β7 receptor in the target peripheral blood lymphocyte subset, and showed a plasma elimination half-life of 25 days (Entyvio Product Monograph, Appendix 1). Approved serial dosing of vedolizumab in IBD is 300mg, about 4 mg/kg.

The chronic active infection of lymphoid and monocytoid cells is targeted in this strategy. It remains to be seen whether latently infected cells and compartments could escape immune control spontaneously, or following new immune-activation triggered by intercurrent illness or iatrogenic event (ie. with vaccination). In planning novel HIV treatment strategies, many experts agree that a combination of strategies will be necessary in order to address all aspects of chronic HIV infection including the active, latent, and concealed reservoirs. One of the most commonly reviewed strategies is the "kick and kill" model, which involves re-activation of latently infected cells with latency reversal agents (LRA), followed by a therapy aimed at eliminating or neutralizing the virus from the host (Deeks et al., 2016). One promising LRA is romidepsin, a Histone Deacetylase (HDAC) inhibitor, which has been shown to effectively induce HIV expression in latently infected CD4 T cells (Wei et al., 2014). Recently, a study for a novel therapeutic HIV-1 vaccine conducted in Spain employed such a strategy by combining romidepsin and vaccination in an effort to induce a neutralizing immune response to HIV-1 with promising results (Mothe, 2017).This study supports the role of latency reversal agents in combination with immune-based therapies as a means of decreasing the viral reservoir in order

to establish and maintain remission of HIV, as 5/20 participants demonstrated remission of viremia after ATI.

We propose a phase II dose- and exposure-finding pilot trial in healthy adults with ART-treated chronic HIV infection to assess vedolizumab safety and sustained anti-HIV effect after ATI. This trial proposal is of a short duration (planned pilot under one year) and simple design to assess short-term safety and tolerance of vedolizumab according to dose, and to assess whether plasma viremia rebound and remission occur after ATI. This trial design accommodates the collection and cryopreservation of biological specimens (sera, plasma, peripheral blood lymphocytes, rectal/sigmoid biopsy samples, CSF), and stool) that will permit ongoing mechanistic immunological and virological studies to be done in the event that treatment effects are observed.

2. Hypothesis and Objectives

We plan to test the following Hypotheses:

1. That vedolizumab is safe in short-term use (6 months) in ART-treated chronic HIV infection (observed on protocol to 12 months), as has been observed in inflammatory bowel disease (Colombel et al., 2016).

2. That ATI (of ART) is safe during and after vedolizumab treatment in HIV-ART, and if followed by rebound and lasting recurrent PVL, that HIV infection remains treatable without adverse outcome as in prior immunotherapeutic trials utilizing ATI (Graziani & Angel, 2015; Routy et al., 2012)

3. That long-lasting remission of PVL may follow short-term PVL rebound, after ATI and after subsequent discontinuation of vedolizumab dosing as observed in the animal model of SIV infection (Byrareddy et al., 2016).

Objectives

1. To identify safety and tolerance of serial dosage of vedolizumab in healthy adults treated with HIV antiretroviral therapy (HIV- ART).

2. To identify whether healthy adults with HIV-ART treated with adjunctive vedolizumab will show a sustained post-treatment remission of PVL/gut viral loads rebound and recurrence after ATI.

3. To identify the clinical, therapeutic, and laboratory immunovirological measures that characterize HIV-ART cases treated with vedolizumab before, during and after adjunctive vedolizumab treatment, ATI, and possible remission of PVL.

3. Study design

This trial is a Phase II single-arm 5 dose-group prospective intentional cohort study in healthy adult ART-treated HIV infected volunteers, administering sequential vedolizumab dosages (600, 300, and 150mg) in a regimen of up to 12 infusions overlapping an ATI with observation for PVL/gut viral load rebound and sustained recurrence or remission.

4. Outcomes

4.1 Primary outcomes

1. Safety and tolerability of vedolizumab treatments of up to 7 doses.
2. The incidence of lasting remission of PVL after vedolizumab treatment and ATI.

4.2 Secondary outcomes

1. For recurrent sustained PVL rebound after ATI, suppression of PVL after resuming ART
2. For recurrent sustained PVL rebound after ATI, recovery of CD4 cell count after resuming ART.
3. Kinetics and trajectory of PVL and CD4 T lymphocyte count during the study year.

In addition to the secondary outcomes noted above, the collection of specimens for storage for:

4. Pharmacokinetic measures of vedolizumab in plasma over the course of the trial.
5. Cerebrospinal fluid plasma virus load (CSF-PVL) in cases with post-ATI suppression of blood PVL.
6. Rectal/sigmoid mucosal biopsy for immunology, immunohistochemistry, and HIV DNA PCR before vedolizumab treatment, and during vedolizumab treatment before and after ATI.

5. Sample size justification and analytical plan

This is an exploratory pilot study of 24 adult volunteers/which will permit observation of the incidence, degree and duration of virological rebound and remission in a series of adult volunteers receiving vedolizumab treatment across ATI. Historically, this does not occur. If it occurs in more than a very few persons, the outcome would be remarkable and clinically meaningful. For this study, only a small sample size required in order to observe a categorical effect on virological rebound and sustained recurrence or remission resulting from vedolizumab treatment.

As an aside, virological rebound will be compared with individual patients' pre-ART plasma virus load measures and with historical controls having undergone ATI in other reported trials. Mean and median levels of PVL rebound will be compared using standard statistical tests, analogous to pharmacokinetic parameters such as Tmax, Cmax, AUC and so on.

6. Eligibility Assessments

6.1 Population

The study population will be stable, otherwise healthy, chronically infected HIV positive adults receiving ART and followed at the Ottawa Hospital (TOH) HIV clinic or the University of Ottawa Outpatient Clinic upon referral to the TOH HIV clinic.

6.2 Inclusion criteria

1. Adult (≥ 18 years and <60 years) with HIV infection and documented pre-ART PVL measures.
2. Duration of ART 2 – 25 years.
3. Nadir CD4 T cell count ≥ 200 and current CD4 T cell count > 500 cells/mcL
4. Adherent on ART > 24 months with sustained, consistent PVL ≤ 50 copies/mL
5. Ability to comprehend and provided informed consent
6. Screening (within 3 months) routine safety laboratory (incl. liver enzymes, bilirubin, CBC, Creatinine) measures < grade 2 levels
7. Willingness for serial phlebotomy and storage and future viro-immunological assays

6.3 Exclusion criteria

1. Past AIDS-defining or AIDS-related immune deficiency diseases
2. Past drug-resistant HIV or treatment refractory PVL response (< 1.0 log10 reduction in PVL at 1 month ART, or > 50 copies/mL at 6 months).
3. Past or risk (in Investigator's assessment) of non-adherence to ART, to study treatment or protocol.
4. Documented past or current chronic HCV or HBV infection,
5. Untreated latent tuberculosis infection or inadequately treated prior TB infection
6. Past HIV-associated immune thrombocytopenia,
7. Past non-HIV associated autoimmune disease requiring immunomodulatory or immunosuppressive treatment,
8. Past non-HIV related immunosuppressive disease or treatment.
9. Pregnancy, lactation, or non-adherence with contraception if fertile.

7. Procedures

7.1 Patient Selection

Patients over the age of 18 with HIV infection and documented pre-ART PVL measures will be approached for the study by a member of their health care team (circle of care). If the patient is interested in participating in the trial the study coordinator will see the patient to further explain the trial and to carry out the informed consent process.

Once the participant has provided signed informed consent, screening for eligibility will occur. Upon confirmation of eligibility by the investigator (or co-investigator), baseline evaluations and administration of the study product will occur.

7.2 Treatment and Dosing

Study participants will be administered vedolizumab intravenously as per the product monograph. This will occur under the direct supervision of the Qualified, Principal and/or Co-Investigators. Doses will be given in intervals of 2-3 weeks during loading phase, and every 4 weeks thereafter as described for ulcerative colitis. The treatment schedule is as follows:

Group A (Current participants):

- Participants # 1 to # 4 will receive 300 mg intravenously at week 0, 2, 5, 8 and every 4 weeks thereafter up to 40 weeks, for up to 7 doses as tolerated and according to pVL response.
- Participants # 5 to # 8 will receive 150 mg intravenously at week 0, 2, 5, 8 and every 4 weeks thereafter up to 40 weeks, for up to 7 doses as tolerated and according to pVL response.

Group B (New participants):

- Participants # 9 to # 16 will receive 600 mg intravenously at week 0, 2, 5, 8 and every 4 weeks thereafter up to 40 weeks, for at least 7 and up to 12 doses as tolerated and according to pVL response.
- Participants # 17 to # 20 will receive 300 mg intravenously at week 0, 2, 5, 8 and every 4 weeks thereafter up to 40 weeks, for at least 7 and up to 12 doses as tolerated and according to pVL response.
- Participants # 21 to #24 will receive 150 mg intravenously at week 0, 2, 5, 8 and every 4 weeks thereafter up to 40 weeks, for at least 7 and up to 12 doses as tolerated and according to pVL response.

As in the appended SIV macaque model publication, ATI will follow the 3rd or 4th dose of vedolizumab. Patients will be observed every 3 to 4 weeks during vedolizumab and ATI up to one year. Monitoring for pVL rebound with serial testing will commence at the time of ATI and continue at regular intervals of 3 - 4 weeks as per the study schedule until week 52 or until sustained recurrence of viremia is documented. After completion of study protocol, patients

will have quarterly testing for CD4 cell count and safety labs and will be seen as needed according to PVL response and/or ART retreatment status and response. They will also have continued observation according to standard clinical practice.

7.3 Study Visit Schedules

Table 1: Study Visit Schedule for 7 doses of vedolizumab – Group A (Current Participants) (Participants enrolled before January 1 2018)

Study Visit[2] → / Study Activity ↓	Screening	Baseline[3]	Week 0[3]	Week 2	Week 5	Week 6, 7^	Week 8	Week 12	Week 16	Week 20	Week 24	Weeks 28, 32, 36, 40, 44, 48	Week 52	Q3 mos after week 52 x3
Informed consent	X													
Eligibility confirmation	X													
Concomitant Meds	X	X	X	X	X		X	X	X	X	X	X		
Pregnancy test if appl.+	X													
Safety Labs (CBC, diff WBC, biochemistry, ESR, CRP)	X[1]	X	X	X			X	X	X	X	X	X	X	X
Serum (10ml)		X	X	X	X		X	X	X	X	X	X		
Plasma (10ml)		X	X	X	X	X	X	X	X	X	X	X		
PBL**		X			X						X			
PVL	X	X					X	X	X	X	X	X	X	X
CD4	X	X					X	X	X	X	X	X	X	X
Vedolizumab IV infusion			X	X	X		X	X	X	X				
History & Physical	X	X	X	X	X		X	X	X	X	X	X	X	
Optional CSF Specimen											X			
Optional Rectal/Sigmoid Mucosal Biopsy		X									X		X	
Optional Stool Sample		X									X		X	
AE Assessment		X	X	X	X	X	X	X	X	X	x	X	X	

[1] If safety labs have been drawn within 1 year of the screening visit these results may be used for screening purposes.
[2] Study visits may occur ± 1 week from the time points outlined in the table.
[3] Baseline and Week 0 visits may occur at the same time
^ ATI (analytical treatment interruption) will occur at week 6-7
** 50 -100 ml phlebotomy for separation & cryopreservation
+ Pregnancy testing with urine dip for βHCG may be repeated at any study visit where it is felt to be indicated by the participant or the study investigator.

Table 2: Study Visit Schedule for up to 12 doses of vedolizumab – Group B (New Participants)

Study Visit[2] → / Study Activity ↓	Screening	Baseline[3]	Week 0[3]	Week 2	Week 5	Week 8	Week 10^	Week 12	Week 16	Week 20	Week 24*	Week 28*	Week 32*	Week 36*	Week 40*	Weeks 44, 48	Week 52	Q3 mos after week 52 x 3
Informed consent	X																	
Eligibility confirmation	X																	
Concomitant Meds	X	X	X	X	X	X		X	X	X	X	X	X	X	X	X		
Pregnancy test if appl.+	X																	
Safety Labs (CBC, diff WBC, biochemistry, ESR, CRP)	X[1]	X	X	X		X		X	X	X	X	X	X	X	X	X	X	X
Serum (10ml)		X	X	X	X	X		X	X	X	X	X	X	X	X	X		
Plasma (10ml)		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X		
PBL**		X			X						X							
PVL	X	X				X		X	X	X	X	X	X	X	X	X	X	X
CD4	X	X	X			X	X	X	X	X	X	X	X	X	X	X	X	X
Vedolizumab IV infusion			X	X	X	X		X	X	X	X*	X*	X*	X*	X*			
History & Physical	X	X	X	X	X	X		X	X	X	X	X	X	X	X	X	X	
Optional CSF Specimen											X[4]							
Optional Rectal/Sigmoid Mucosal Biopsy[5]		X				X					X[4]							
Optional Stool Sample		X									X[4]						X	
AE Assessment		X	X	X	X	X	X	X	X	X	x	X	X	X	X	X	X	

[1] If safety labs have been drawn within 1 year of the screening visit these results may be used for screening purposes.

[2] Study visits may occur ± 1 week from the time points outlined in the table.

[3] Baseline and Week 0 visits may occur at the same time

[4] Optional testing may occur within 2 weeks of study week 24 and will not necessarily occur during Week 24 study visit.

[5] Rectal/Sigmoid mucosal biopsies may occur within 4 weeks of the scheduled study visit for week 0, 8 or 24.

^ ATI (analytical treatment interruption) will occur at week 10

* Vedolizumab infusion to be given depending on tolerance and pVL response

** 50 -100 ml phlebotomy for separation & cryopreservation

+ Pregnancy testing with urine dip for βHCG may be repeated at any study visit where it is felt to be indicated by the participant or the study investigator.

7.4 Clinical and Laboratory Evaluations

7.4.1 Clinical Evaluations

Medical history and AE screening will be performed at all study visits. Physical exams will be performed at screening and baseline visit, and when indicated based on medical history or AE screening assessment. Clinical history includes, but is not limited to, assessment for new or evolving neurological symptoms. Participant's concomitant medications will be reviewed at screening, baseline and all study visits thereafter.

7.4.2 Laboratory Evaluations and Specimen Collection

Pregnancy test if appl.

- Participants capable of becoming pregnant during the course of the study will undertake a urine pregnancy test at screening, and at any subsequent visit where it is clinically indicated on the basis of history, screening labs, or physical exam.

Safety Labs:

- Phlebotomy will be performed for whole blood to monitor laboratory markers of safety of the study intervention and analytical treatment interruption. These tests include a complete blood count with white blood cell differential, serum electrolytes, serum creatinine, serum liver enzyme tests, serum lipids, and serum markers of inflammation will be performed at each visit as indicated by the study schedule.

Serum, Plasma, and PBL:

- Phlebotomy will be performed for serum, plasma, and for PBL isolation will be collected for further testing at several visits as indicated in the study schedule. This testing may include antibody titres and avidity assays, and other very standard cellular immunity measures such as interferon gamma expression in response to HIV antigenic stimulation, and cellular proliferative responses. The techniques include PBMC cell culture, elispot and viral outgrowth assays, virus tropism and envelope phenotypes, viral genotypes or polymorphisms, all with conventional and standard tests.

PVL

- Phlebotomy will be performed for whole blood for plasma viral load testing to be performed as per standard of care test in the province of Ontario, every 4 weeks after treatment interruption at each visit indicated on the schedule.

CD4 cell count

- Phlebotomy will be performed for whole blood for flow cytometry to determine CD4 count at each visit indicated in the schedule.

Optional CSF testing:

- Lumbar puncture will be performed according to standard procedure by the study investigator or interventional radiologist for participants who achieve viral suppression or who have evidence of biological effect of vedolizumab on post-ATI viral load at or after week 24. CSF will be analyzed for cell count and white blood cell count differential, protein level, glucose, presence of JC virus by PCR testing, and additional testing for detection and quantification of HIV, and further testing of immunology and virology including HIV and provirus quantitation.
- CSF testing requires a lumbar puncture and may be obtained on a separately scheduled or dedicated visit within 2 weeks of study week 24.

Optional Rectal/Sigmoid Mucosal Biopsy:

- Biopsies of GI mucosa from the rectum and distal sigmoid will be performed by a Gastroenterologist at weeks 0, 8, and 24 weeks for all consenting participants. Rectal/Sigmoid mucosa will be processed for isolation of immune cells and immune cell subsets. Samples will also be collected for additional testing for characterization of HIV infection and HIV-specific immunity. The tests done will be immunohistochemical staining, standard histopathological microscopic examination, and HIV and provirus quantitation.
- Rectal/Sigmoid mucosal biopsy requires sigmoidoscopy performed by a gastroenterologist in an endoscopy suite, sigmoidoscopy and biopsy will be performed as per standard of care. Biopsies may be obtained on a separately scheduled or dedicated visit within 4 weeks of the relevant study week.

Optional Stool Samples

- Stool will be collected at weeks 0, 24, and 52 for all consenting participants. Stool will be processed and analyzed with nucleic acid probe testing for fecal microbiome, additional testing for identification of inflammatory markers will also be performed.

- Stool will be collected by participants at home according to stool collection protocol used for microbiologic stool samples at TOH and brought with participants to the study visits at week 0, 24, and 52. Stool may be collected and provided to the study team within 2 weeks of the scheduled visits.

Both Drs Byrareddy and Kumar offer intellectual contribution to the trial. Their laboratories will be performing standard research test of immunology and virology for HIV infection and HIV-specific immunity. More specifically, Dr. Ashok Kumar from CHEO (the Children's Hospital of Eastern Ontario) will be testing HIV specific immunity using peripheral blood (serum, plasma, PBMCs) and rectal/sigmoid mucosal tissue. The serum, plasma and cryopreserved PBMCs will be processed, preserved and stored at the OHRI, in Dr Jonathan Angel's laboratory. Dr. Siddappa Byrareddy from the University of Nebraska will be testing the cerebrospinal fluid (CSF) specimen, the biopsy and stool samples. Some of the work planned for Dr Kumar's laboratory may also be performed by Dr Byrareddy's laboratory, and by Dr Jonathan Angel's laboratory.

7.5 Investigational Product

The investigational product vedolizumab will be obtained through the Ottawa Hospital Pharmacy, who will also be responsible for drug accountability.

Vedolizumab is supplied in sterile 20 mL single-use glass vials containing 300 mg of vedolizumab. Each individual carton contains one single-use vial.

The vials will be stored in the outer carton to protect from light as outlined in the product monograph. These cartons will be stored in a refrigerator (2° to 8°C) and daily temperature monitoring will be recorded.

The product will be labelled in compliance with the Food and Drug Regulations Division 5, Part C. Once prepared, the IV bag will be labelled according to pharmacy procedure.

7.6 Participant Discontinuation

Participants may stop protocol treatment in the following instances:

- Adverse event which would in the judgment of the investigator require discontinuation of study product
- Request by the participant or their legally authorized representative (consent withdrawal)
- Non-compliance with the study protocol
- The sponsor or Health Canada may discontinue the study at any time.

7.7 Treatment Discontinuation

Vedolizumab may be discontinued and ART re-initiated in the following instances:

- pVL rebound after ATI to greater than 0.5log higher than the patient's pre-ART viral load
- Sustained viral rebound, ie. pVL duration of greater than 12 weeks based on at least 2 measurements at least 1 month apart.
- Confirmed CD4 count fall to <350 cells/mL or by >30% from the patient's average pre-enrollment CD4 level in the preceding 12 months, over at least 2 measurements at least 1 month apart.

7.8 Study Closure

The study may be closed if all the following criteria are met:

- All subject recruitment and enrollment is complete
- All subject specimens, records, data have been obtained
- No further contact with participants is necessary
- Analysis of subject identifiable data, records, specimens are complete

7.8.1 Premature Study Closure:

The study may be closed if at least one of the following criteria are met:

- The DSMB or Medical Monitor recommends study closure in the interest of protection of human participants.
- 25% or more of participants withdraw due to side effects of study intervention.
- A significant number of SAEs are encountered in participation of the study

In the event of premature study closure, the Sponsor and the Investigator will ensure that adequate consideration is given to the protection of the best interests of the participants including adequate follow-up. REB and Regulatory authorities will be informed.

7.9 Concomitant Medications

As per the product monograph, concomitant use of vedolizumab with TNFα antagonists and natalizumab have the potential for increased risk of infections and should therefore be avoided. Concomitant regular use of any other significantly immunosuppressive medication should likewise be avoided.

7.10 Risk Factors & Symptom Management

There is a potential for infusion related reactions (IRR) and hypersensitivity. These reactions can occur during the infusion, immediately after the infusion or several hours after the infusion.

In the event of a severe anaphylactic reaction or other severe reaction, administration of the study drug will be stopped immediately, and appropriate treatment will be provided.

Vedolizumab may increase the risk of upper and lower respiratory tract viral infections. Participants will be offered influenza vaccination during influenza season and monitored by clinical assessment at study visits for signs and symptoms of viral infections. Appropriate symptomatic or antiviral treatment will be offered as necessary.

Participants will be monitored for neurological signs and symptoms including progressive weakness on one side of the body, clumsiness of limbs, disturbance of vision, memory, confusion and personality changes. In participants from whom CSF is obtained, JC virus testing will be performed to assess markers of progressive multifocal leukencephalopathy.

8. Data Collection & Record Retention

Study information will be collected by the clinical research coordinator and reported on paper case report forms.

Data collected will be entered into a validated database developed by the OHRI Methods Centre. Case report forms and source documents will be kept in a secure location until study termination upon which time they will be stored in a secure facility for a period of 25 years as per Health Canada regulations. At that time they will be destroyed as per institutional policy.

9. Safety Reporting Requirements

Adverse events are defined as any unfavorable and unintended sign (including an abnormal laboratory finding); symptoms or disease temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product; pre-existing symptoms or conditions which worsen during a study.

Information about all adverse events whether volunteered by the subject, discovered by the investigator or designates questioning, laboratory test or other means, will be recorded and followed as appropriate.

Participants will be monitored for all AEs and SAEs and in addition, in this trial there will be particular vigilance for infusion related adverse events.

9.1 Causality

Relationship of the adverse event to the investigational product (causality) will be assessed by the investigator as follows:

- **Unrelated**: There is not a reasonable possibility that the adverse event may have been caused the drug
- **Possibly related**: The adverse event may have been caused by the study drug, however this is insufficient information to determine the likelihood of this possibility.

- **Related**: There is a reasonable possibility that the adverse event may have been caused by the study drug.

9.2 Expectedness

Adverse events will also be assessed according to the following categories:

- **Expected (anticipated):** the event is identified in nature, severity, or frequency in the applicable product information (investigators' brochure, current protocol, informed consent, and/or other current risk information).
- **Unexpected (unanticipated):** the event is not identified in nature, severity or frequency in the applicable product information (investigator's brochure, current protocol, informed consent, and/or other current risk information).
- **More Prevalent:** the event occurs more frequently than anticipated or at a higher prevalence than expected.

9.3 Grading

Grading of Adverse Events will be as per the Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events, attached to the current protocol as appendix 5.

9.4 Serious Adverse Event Reporting

A serious adverse event:

- Results in death
- Is life threatening
- Requires participant hospitalization or prolongation of existing hospitalization
- Results in persistent or significant disability/incapacity
- Is a congenital anomaly/birth defect

Some conditions may not be immediately life threatening or require hospitalization. Should the investigator feel that the event may jeopardize the participant or may require intervention to prevent more serious outcomes, then it should be treated as serious. Adverse events will be recorded once the study participant has completed the Baseline/Week 0 visit.

9.5 Reporting to the Research Ethics Board

Adverse events and serious adverse events will be reported to the REB as per their reporting guidelines and Institutional Standard Operating Procedures (SOPs).

9.6 Reporting to Health Canada

SAEs that are both unexpected and related or possibly related are subject to expedited reporting to Health Canada according to the regulations below. SAEs that are expected or that are unrelated to the study drug are not reportable. SAE reporting should occur as follows:

- When the SAE is neither fatal nor life-threatening, within 15 days after becoming aware of the information.
- Where the SAE is fatal or life threatening, immediately where possible and, in any event, within 7 days after becoming aware of the information.
- Within 8 days after having informed Health Canada of the SAE, the principal investigator will submit as complete as possible, a report which includes an assessment of the importance and implication of any findings.

Each SAE which is subject to expedited reporting should be reported individually using the CIOMS-I form. Any updated follow up information that becomes available regarding the SAE should be reported in a follow up report.

10. Quality Assurance

10.1 DSMB

To enhance patient safety, in addition to complying with the Health Canada Food and Drug regulations program, and GCP, an independent Data Safety Monitoring Board will be established for this trial. The DSMB will review safety data and efficacy and will raise any questions about protocol performance and outcomes. They will meet as per the approved DSMB terms of reference document.

10.2 Monitoring

The OHRI Internal Monitor will conduct a monitoring visit shortly after enrollment commences. This monitoring by OHRI does not replace the routine quality control to be performed by the investigator or designee. Routine quality assurance will be completed by an assigned monitor to ensure that the study is being conducted according to the protocol. The study will be monitored according to the approved monitoring plan.

To enhance patient safety, in addition to complying with the Health Canada Food and Drug regulations program, and GCP, and appointment of a DSMB, an independent Medical Monitor (MM) will be appointed. The MM will have the responsibility of providing oversight with respect to safety during the study. They will also be responsible for reviewing the protocol and any new information about the study product as it becomes available, including the Investigational Brochure, and reported safety events. The MM, in consultation with the DSMB, will provide safety review during the execution of the clinical trial. This oversight includes reviewing safety information and providing applicable recommendations, as appropriate, to members of the study team, including but not limited to: the investigators, DSMB, Sponsor, and Health Canada. The MM will be authorized to recommend re-treatment with ART and/or discontinuation of

vedolizumab and/or withdrawal from the study of any participant. The MM will also adjudicate eligibility with respect to inclusion and exclusion criteria, or treatment discontinuation.

10.3 Audits and Inspections

The investigator(s)/institution(s) will permit trial related monitoring, audits, REB review and regulatory inspections and will provide direct access to source data/documents as required.

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Appendices

Appendix 1 - Entyvio product monograph
Appendix 2 - Sustained virologic control in SIV+macaques after antiretroviral and α4β7 antibody therapy. Byrareddy, S. N., Arthos, J., Cicala, C., Villinger, F., Ortiz, K. T., Little, D., Ansari, A. A. (2016). Science, 354(6309), 197-202. doi:10.1126/science.aag1276
Appendix 3 - Division of AIDS (DAIDS) Table for Grading the Severity of Adult and Pediatric Adverse Events