AMENDMENT N°1
TO THE VFI-A24-031
MATERIAL TRANSFER AGREEMENT

WHEREAS,

a) Vaccine Formulation Institute CH Ltd. (VFI), a company organised and existing under the laws of Switzerland, having its principal place of business at Rue du Champ-Blanchod 4, 1228 Plan-Les-Ouates, Geneva, Switzerland, and its affiliates **("VFI")**,

and

b) Palacky University Olomouc, Faculty of Medicine and Dentistry, Department of Immunology, ID number 61989592, Hnevotinska 976/3, 779 00 Olomouc, Czech Republic (**"PUO"**).

VFI and PUO are individually referred to as "Party" or collectively as the "Parties".

AND WHEREAS,

The Parties signed a Material Transfer Agreement effective from Dec 6th, 2024 that records the terms under which the above Parties will make available adjuvants, antigens and assay technology as further described in Annex 1 of the Agreement (which may be amended from time to time to include further research projects and work schedules between the two Parties related to adjuvant development).

NOW THEREFORE :

In order to continue the collaboration, the Parties agree to extend the scope of the Agreement by this amendment (hereinafter, "MTA Amendment N°1").

Therefore, the Parties agree as follows:

1) All capitalized terms used in this Amendment N°1 shall have the same meaning as defined in the Agreement unless otherwise specifically indicated herein.
2) Except as expressly amended and modified in this Amendment N°1, all Articles of the Agreement remain unchanged, such that the terms and conditions of the Agreement shall also apply to the updated Statement of Work (see Annex 1 below).
3) The Work Plan of the Agreement is hereby modified to include an additional study plan, included here in Amendment N°1 as Annex 1.
4) The term of the Agreement shall be extended to accommodate the additional work, such that the term shall continue without interruption from the effective date of the Agreement until completion of the Statement of Work of Annex 1.

Rue du Champ-Blanchod 4 – 1228 Plan-Les-Ouates, Switzerland – www.vaccineformulationinstitute.org

-Signature page follows-

Accepted and Agreed on behalf of

Palacky University Olomouc

[redacted]

Signature:

Date: 26-06-2025

[redacted]

Accepted and Agreed on behalf of

VFI

[redacted]

Signature:

Date: [redacted] Date: 2025.06.17 12:35:35 +02'00'

Accepted and Agreed on behalf of

Palacky University Olomouc

[redacted]

Signature:

Date: [redacted]

Accepted and Agreed on behalf of

VFI

[redacted]

Signature: [redacted]

Date: Date: 2025.06.17 08:33:25 +02'00'

ANNEX 1: Further Work Plan relating to A24-031

STATEMENT OF WORK

Evaluation of VFI adjuvants with Palacky University HIV vaccine candidate

Author: [redacted] & [redacted] **Version:** 4.0 **Date:** 10.06.2025

1. AIM OF THE STUDY

The aim of this study is to evaluate the compatibility and short-term stability of recombinant HIV antigens (MLB-036 or MLD-108), both in soluble form as well as in a VLP-form, in formulation with VFI adjuvants.

2. BACKGROUND

The antigen MLB-036 and MLD-108 are based on myomedin. Myomedin-derived variants can mimic particular V3 glycan epitopes of prominent anti-HIV-1 bNAbs, ascertain the potential of particular glycans controlling neutralizing sensitivity of individual HIV-1 pseudoviruses, and represent promising prophylactic candidates for HIV-1 vaccine development. The MLB-036 and MLD-108 antigens were developed by [redacted] et al. at the Palacky University in Czech Republic. The antigens have been optimized to have high specificity for PGT121 and PGT126 HIV broadly neutralizing antibodies respectively. They have been evaluated pre-clinically in combination with a liposomal adjuvant containing MPLA using an intradermal route and they induce broadly neutralizing antibodies. Current investigations aim to evaluate one of these antigens with clinically relevant adjuvants, by intramuscular route.

3. ABBREVIATIONS

DOPC	dioleyl phosphatidyl choline
Q	QS21
HPLC	High pressure liquid chromatography
LC-MS	Liquid chromatography coupled to mass spectrometry
LMQ	liposome containing the TLR4 ligand 3D6AP and the saponin QS21
LQ	liposomes consisting of DOPC and cholesterol, containing the saponin QS21
PBS	GIBCO phosphate buffer saline, pH 7.2
SQ	Oil-in-water emulsion containing the saponin QS21
SWE	Squalene-in-water emulsion
VLP	Virus-like particle

4. MATERIALS

Rue du Champ-Blanchod 4 – 1228 Plan-Les-Ouates, Switzerland – www.vaccineformulationinstitute.org

4.1. Materials and reagents provided by the partner

- Recombinant HIV antigen
    - myomedin-based soluble protein antigen MLB-036 or MLD-108 (N-terminal His-tag, C-terminal V5 tag, 16 kDa) in TNI buffer (50mM Tris, 300mM NaCl, 250mM imidazole, PH 8.0)
    - myomedin-based MLB-036 or MLD-108-VLP (envelope particle, 55 kDa) in PBS

4.2. Materials and reagents provided by VFI

- Sepivac SWE™ (SWE) provided through Seppic
- SQ, LQ, LMQ provided by VFI

5. STUDY OUTLINE AND PROCEDURE

5.1. Formulation study

A formulation study will be completed at VFI to monitor adjuvant physicochemical characteristics and compatibility through antigen integrity.
The formulations are listed in Table 1.

Table 1: Formulation table

Antigen	Antigen dose	Adjuvant
MLB-036 or MLD-108-VLP	0.7 µg / 50 µL	-
MLB-036 or MLD-108-VLP	0.7 µg / 50 µL	SWE
MLB-036 or MLD-108-VLP	0.7 µg / 50 µL	SQ
MLB-036 or MLD-108-VLP	0.7 µg / 50 µL	LQ
MLB-036 or MLD-108-VLP	0.7 µg / 50 µL	LMQ
MLB-036 or MLD-108	10 µg / 50 µL	-
MLB-036 or MLD-108	10 µg / 50 µL	SWE
MLB-036 or MLD-108	10 µg / 50 µL	SQ
MLB-036 or MLD-108	10 µg / 50 µL	LQ
MLB-036 or MLD-108	10 µg / 50 µL	LMQ

5.1.1. Adjuvant physicochemical characterisation

Physicochemical characterization will be performed at 24 h (day 1) at 5°C and include visual inspection, particle size, polydispersity, zeta potential, pH, osmolality. Furthermore, squalene content (HPLC), lipid content (HPLC) and TLR4 ligand and QS21 content (UPLC-MS) will be tested.

5.1.2. Antigen integrity

Antigen integrity in the presence of VFI adjuvants will be evaluated by SDS-PAGE.

5.1. Immunogenicity study at Palacky University

In two experiments we will evaluate the HIV-1 antibody-neutralizing potency and breadth of either MLB-036 or MLD-108 adjuvanted with SWE, LQ, SQ, LMQ compared to liposomal-HIS-tagged MLB-036 or MLD-108.

(1) Pilot experiment in mice for antigen dose finding with 2 adjuvants.

- Mouse strain: 6–8-Week-old female BALBc
- Route: Intramuscular
- Immunization Schedule: Prime-2xBoost (Week 0, 3, 6)
- Bleeding Schedule: Week 0 (naïve), 3, 6, 9(terminal)
- Total number of mice: 40

***Table 2*: Immunogenicity study in mice: dose range**

Group	Antigen	Antigen dose	Adjuvant	No. of mice
1	MLB-036 or MLD-108	10µg/50µL	SWE	5
2	MLB-036 or MLD-108	10µg/50µL	LMQ	5
3	MLB-036 or MLD-108	5µg/50µL	SWE	5
4	MLB-036 or MLD-108	5µg/50µL	LMQ	5
5	MLB-036 or MLD-108	2µg/50µL	SWE	5
6	MLB-036 or MLD-108	2µg/50µL	LMQ	5
7	MLB-036 or MLD-108 (control)	10µg/50µL	Liposome-HIS tag	5
8	MLB-036 or MLD-108 (control)	10µg/50µL	-	5

(2) **Mice experiment with full adjuvant panel with selected dose to establish ranking in best inducer of neutralizing activity.** *Possibly look at persistence of response in mice (important data for NHP)*

- Mouse strain: 6–8-Week-old female BALBc
- Route: Intramuscular
- Immunization Schedule: Prime-2xBoost (Week 0, 3, 6)
- Bleeding schedule: Week 0 (naïve), 3, 6, 8, 10 (terminal)
- Total number of mice: 60

***Table 3*: Immunogenicity study in mice: adjuvant selection**

Group	Antigen	Antigen dose	Adjuvant	No. of mice
1	MLB-036 or MLD-108	Selected from the pilot study / 50 µL	SWE	10
2	MLB-036 or MLD-108	" "	SQ	10
3	MLB-036 or MLD-108	" "	LQ	10
4	MLB-036 or MLD-108	" "	LMQ	10

5	MLB-036 or MLD-108 (control)	" "	Liposome-HIS tag	10
6	MLB-036 or MLD-108 (control)	" "	-	10

6. REPORTING

A report documenting all data generated in the formulation and immunogenicity study will be drafted when the full set of data is available.

7. BUDGET

The budget for this study is 7600,- EUR. The full amount will be invoiced upon signature of the MTA.

Shipping of sera to Palacky University will be charged to Palacky University as invoiced by the shipping company.