MaaT034, a new co-cultured microbiome ecosystem therapy candidate, safely colonizes the gastro-intestinal tract of germ-free mice, restores a healthy gut physiology and stimulates immunity

Bastien Laperrousaz, Julie Reygner, Charlotte Petitjean, Aurore Duquenoy, Cyrielle Gasc, Diane Plouchart, Sophie Declomesnil, Carole Schwintner, Kathy McCoy, Nathalie Corvaia

MaaT Pharma, 70 avenue Tony Garnier, 69007 Lyon, France

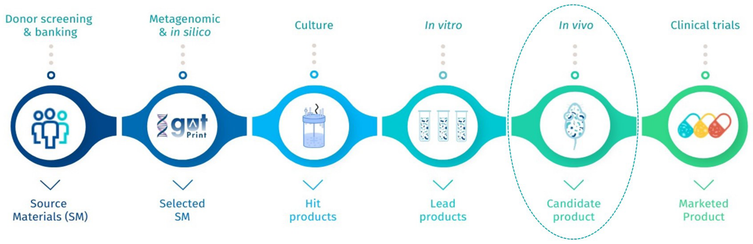
Abstract #2209

INTRODUCTION

Increasing evidence suggests that **gut microbiome composition modulates tumor response to therapies**, including immune checkpoint inhibitors (ICI). Clinical proofs of concept were obtained using ICI-responder fecal microbiota transplants to modulate the gut microbiome of non-responding cancer patients and improve their response to ICI [1-4]. These results support the development of microbiotherapies replicating the effects of ICI-responders as adjunctive therapies. MaaT Pharma is a late clinical-stage biotech leader in developing **Microbiome Ecosystem Therapies (MET) in oncology.** Its native, donor-derived, pooled MaaT013 product demonstrated positive Phase 3 results in acute Graft-versus-Host Disease and is currently being evaluated in a phase 2a randomized multicenter clinical trial in metastatic melanoma. In parallel, MaaT Pharma has developed a unique, ground-breaking, patented co-culture process allowing to replicate and leverage, at large industrial scale, the richness and diversity of native-based microbiome ecosystems while tuning the resulting product according to indication-specific compositions.

The objective of this study is to assess the impact of the co-cultured MaaT034 candidate on gut homeostasis and immune activation.

METHODS



We assessed the impact of MaaT034 on gut homeostasis and immune cell activation using a combination of methods:

- Metagenomic analysis (16S and shotgun)
- Germ-free mice bearing MC38 tumors
- Germ-free mice
- Histology
- Metabolite quantification (LC-MS and GC-MS)
- Mixed Lymphocyte Reaction
- PBMC killing assay

RESULTS

MaaT034 reproduces MaaT013 microbial functions

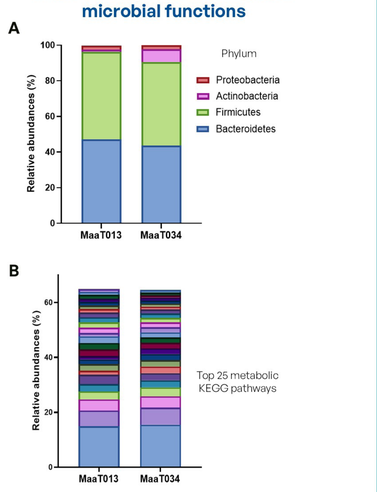


Figure 1. Metagenomic analysis of native MaaT013 and co-cultured MaaT034 ecosystem. 16S rDNA sequencing allowed to assess taxonomic classification. **A)** Stacked bar plot of phylum relative abundances (top 99% abundant phyla). **B)** Shotgun sequencing was performed to assess relative abundance of top 25 metabolic KEGG pathways.

MaaT034 and MaaT013 potentiate anti-PD1 mediated activity in germ-free mice bearing MC38 tumors

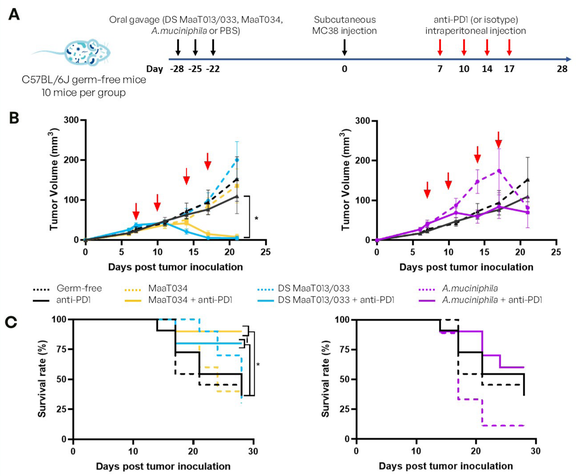


Figure 2. Anti-tumoral activity of MaaT013 and MaaT034 in combination with anti-PD1 in germ-free mice bearing MC38 tumors. A) Experimental design. **B)** Mean tumor volume (Left: MaaT013 and MaaT034; right: *Akkermansia muciniphila*). **C)** Survival rate (Left: MaaT013 and MaaT034; right: *Akkermansia muciniphila*). Results are expressed as mean ± SEM of 2 experiments (10 mice total). Chi-2 test for survival rate and mixed-effects analysis with Tukey's multiple comparisons test. * p<0.05, i p=0.06

MaaT034 engrafts in germ-free mice and produces key metabolites for gut homeostasis and immunity

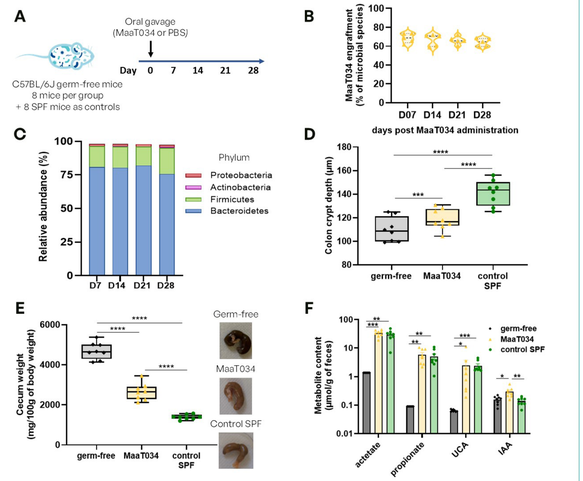


Figure 3. Impact of MaaT034 on germ-free mice physiology. A) Experimental design. **B)** 16S rDNA sequencing of mice feces allowed to assess MaaT034 microbial species engraftment in mice and **C)** gut microbiota taxonomic composition. **D)** Histological analysis of colon crypt depth. **E)** Cecum weight. ANOVA and Tukey's multiple comparisons tests. **F)** Key fecal metabolites were quantified by LC-MS and GC-MS. Kruskal-Wallis and Dunn's multiple comparisons tests. *p<0.05; **p<0.005; ***p<0.0005; ****P<0.0001.

MaaT034 promotes DC-mediated T cell activation and cancer cell killing *in vitro*

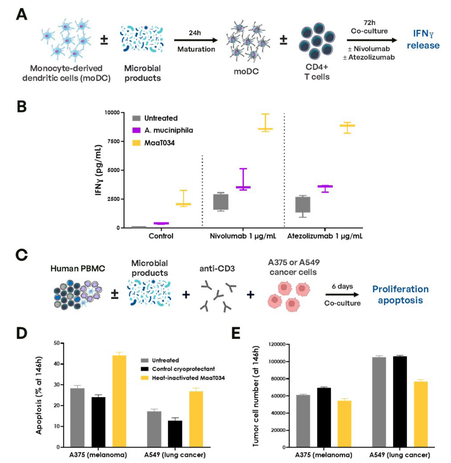
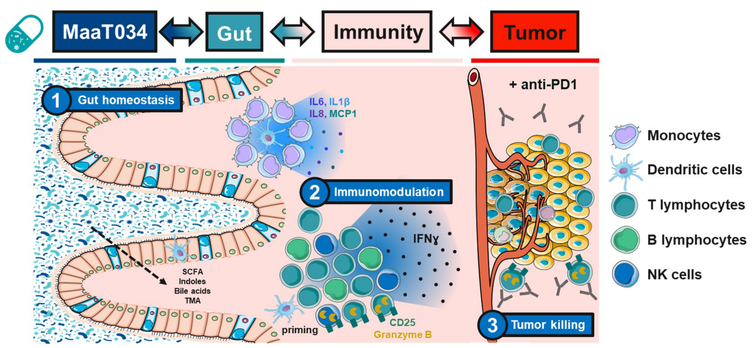


Figure 4. Immunomodulatory potential of MaaT034. A) Allogenic moDC/CD4 Mixed Lymphocyte Reaction. **B)** IFNγ release as a marker of T cell activation in response to MaaT034 or A. muciniphila conditioned medium ± Nivolumab or Atezolizumab. Example of one out of two HLA-mismatched donor pairs. **C)** PBMC killing assay. A375 melanoma and A549 lung cancer cell **D)** apoptosis and **E)** proliferation in response to anti-CD3 activated PBMCs ± MaaT034 via IncuCyte S3™ live cell imaging.

CONCLUSIONS



MaaT034:

- **replicates, at large industrial scale, the functions** of healthy native-based microbiome ecosystems
- **engrafts** durably in the gastrointestinal tract of germ-free mice
- **produces** key microbial-derived metabolites
- **improves** gastrointestinal physiology
- **potentiates anti-tumor effects** mediated by anti-PD-1 checkpoint blockade in germ-free mice bearing MC38 tumors

Altogether, these results highlight the potential of MaaT034 to improve gut physiology and to potentiate **ICI mediated anti-tumoral response**.

These outcomes paved the way for the identification of a promising frontrunner, **MaaT034,** slated for further advancements in clinical development.

REFERENCES

1. Gopalakrishnan V *et al.* Gut microbiome modulates response to anti–PD-1 immunotherapy in melanoma patients. Science. 2018
2. Davar D, *et al.* Fecal microbiota transplant overcomes resistance to anti-PD-1 therapy in melanoma patients. Science. 2021
3. Baruch EN, *et al.* Fecal microbiota transplant promotes response in immunotherapy-refractory melanoma patients. Science. 2021
4. Routy B, *et al.* Fecal microbiota transplantation plus anti-PD-1 immunotherapy in advanced melanoma: a phase I trial. Nature Medicine. 2023