A. APPLICANT PROFILE

I. Personal Information:

Applicant Full Name	Ilyas Bendahmane
Applicant Role	Trainee
Canadian Residency Status	Work Permit
Country of Citizenship	Morocco
Primary Institution	Laval University
Position Title	Postdoctoral Researcher
Laboratory website (if applicable)	www.crchudequebec.ulaval.ca/chercheur/manu-rangachari/
Area(s) of Expertise	Neuroimmunology
Research Specialization Keywords	Progressive Multiple Sclerosis (**MS**), Peripheral Helper T Cells (**Tph**), Disease-Associated Microglia (**DAM**), Tertiary Lymphoid organs (**TLO**), single-cell RNA-seq.

II. Education / Training:

Degree / Qualification	Start and End Date (MM/YYYY – MM/YYYY)	Institution and Supervisor	Area of Study
Doctorate, PhD	04/2022 – 09/2025	Université de Toulouse III | Supervisors: Hanna Mila, Annabelle Meynadier	Infectiology, Pathophysiology, Toxicology, Genetics, and Nutrition
Master's degree	09/2019 – 07/2021	Université de Toulouse III | Supervisor: Bruno Guiard	Pharmacological Innovation, Pharmacology
Bachelor's degree	09/2017 – 07/2019	Université de Toulouse III | Supervisor: Isabelle Castan-Laurell	Cellular Biology and Physiology

III. Positions / Scientific Appointments:

Position / Scientific Appointment	Institution/ Department	Start and End Date (MM/YYYY – MM/YYYY)
Postdoctoral Researcher	Research Centre of CHU de Québec, Neuroscience Axis - Université Laval - Québec	04/2026 – 04/2030 (Expected)
Guest Lecturer	École d'ingénieurs de Purpan, Agronomy Department - Toulouse	03/2022 – 03/2025

B. PERSONAL STATEMENT

I am a multidisciplinary biomedical researcher interested in understanding how interactions between biological systems shape health and disease. During my Master's training, I developed my first expertise

in **microglial biology, cellular metabolism, and chronic inflammation**. In the first year, I investigated how hyperglycemia affects microglial morphology and function, providing an early foundation in CNS-resident immune cells. During the second year, I extended this interest to **metabolic stress and chronic vascular inflammation**, examining how hyperglycemia influences cell fate during atherosclerosis. Together, these projects introduced me to the relationship between metabolism, inflammation, and tissue-resident cells. During my Ph.D, I broadened this perspective by studying the **gut-brain axis during early development**. My research integrated microbiome, metabolome, behavioral, and physiological data to investigate how nutrition and the gut microbiota interact with host development. This work allowed me to develop expertise in 16S rRNA sequencing, multi-omics data analysis, advanced statistical modelling, and identification of candidate predictive biomarker. It also strengthened my ability to integrate complex biological datasets and to link biological changes with clinically relevant phenotypes.

My postdoctoral training is now expanding my experimental expertise through T-cell–microglia co-culture systems, high-speed cell sorting, spectral flow cytometry, and Seahorse metabolic flux analysis. This combination of established and newly acquired expertise allows me to move from identifying associations in complex biological datasets toward experimentally testing cellular, molecular, and metabolic mechanisms of neuroinflammation.

Overall, my trajectory reflects a progressive transition from studying metabolic stress and tissue-resident immune cells, to investigating microbiome-gut-brain interactions, and ultimately to studying immune-CNS interactions in progressive MS. This multidisciplinary foundation now positions me to develop an independent research program in translational neuroimmunology, combining mechanistic studies in experimental models with patient-derived data to identify biomarkers and therapeutic targets relevant to progressive neuroinflammatory disease.

C. MOST SIGNIFICANT CONTRIBUTIONS

1- <u>Maternal nutrition, microbiome and early-life metabolic programming</u>. Low birth weight and impaired early growth are associated with increased neonatal mortality and may contribute to long-term metabolic disturbances. During my PhD, I investigated whether maternal nutritional intervention could influence early-life growth, microbial and metabolic development in newborn puppies. We showed that maternal supplementation with *Saccharomyces boulardii* I-1079 during gestation and lactation was associated with changes in early growth microbial and metabolic trajectories, including modulation of pathways involved in proline biosynthesis, an amino acid important for musculoskeletal development. These findings provided evidence that maternal nutrition can influence early-life metabolic programming and highlighted the potential of targeted interventions to support neonatal health. **Relevance to MS research:** This work established my expertise in integrating microbiome and metabolomic data to investigate interactions between microbial and metabolic processes. These skills are directly transferable to MS research, where metabolic and immune dysregulation contribute to disease heterogeneity and progression. **Role and contribution (~90%):** I played a leading role throughout the study, from data generation to interpretation and dissemination. I analyzed Illumina 16S rRNA sequencing data to characterize early microbial dynamics and integrated these data with serum and urine metabolomic profiles. I developed the statistical models, contributed to the biological interpretation of the multi-omics results. As first author, I led manuscript preparation, generated the figures, organized the underlying datasets, and presented the findings at three international conferences (**ESAR**, Nantes; **EVSSAR**, Barcelona; **ESVCN**, Belfast). This work was published in ***Frontiers in Nutrition*** (**2025**).

2- <u>Gut microbiota, behavior and the gut–brain axis</u>. The relationships between gut microbiota, behavior, and gastrointestinal health during early development had not previously been characterized in puppies. During my PhD, I investigated how the transition through weaning shapes the gut–brain axis

and identified distinct behavioral profiles in Golden Retriever puppies one week after weaning. We found that these profiles were associated with specific gut microbial signatures and fecal quality, and identified quantitative behavioral parameters, including exploration time and interactions with novel objects, that could predict later behavioral profiles. These findings revealed links between behavioral profiles, gut microbial composition, and gastrointestinal health during a critical developmental window, establishing a framework for investigating early-life gut-brain interactions and their potential consequences for later development. **Relevance to MS research:** This work strengthened my expertise in investigating how peripheral biological signals can influence brain function and behavior through the gut–brain axis, providing a complementary perspective to my current research on immune–CNS interactions in MS. **Role and contribution (~90%):** As the primary lead researcher, I conceptualized the experimental design, established the Illumina 16S rRNA sequencing and bioinformatics workflow, performed the behavioral and microbiome statistical analyses, and integrated microbial, behavioral, and gastrointestinal data. I supervised veterinary doctoral students, led the interpretation of the findings, and was responsible for drafting the manuscript and presenting the work internationally. This research led to the submission of a manuscript to ***Applied Animal Behaviour Science***. It was presented at **IPC 2025** (Athens), where it received the **Best Oral Presentation Award**, as well as at **ECBB** (Zurich) and **EVSSAR** (Barcelona). The work also received national media coverage in **France 3** (T.V.) and ***La Dépêche du Midi*** (text).

3- <u>Standardization of clinical tools for gastrointestinal health assessment</u>. Methodological rigor is the cornerstone of reliable biomedical science. In this project, I contributed to the validation of standardized visual fecal scoring systems, addressing an important methodological gap in veterinary gastroenterology and neonatal monitoring. By establishing validated tools for objective assessment of digestive health, this work improved consistency in the evaluation of gastrointestinal disorders and provided an accessible framework for research and clinical monitoring. Beyond its scientific value, this work has practical implications for young animals, in which gastrointestinal disturbances are a common reason for veterinary consultation and normal variations in fecal consistency may be mistaken for diarrhea. Our validated scoring framework may ultimately support more accurate monitoring of digestive health and improve veterinary triage. **Relevance to MS research:** This work strengthened my expertise in developing and validating reproducible phenotypic measures, an essential skill for translating complex biological findings into robust and clinically meaningful outcomes in MS research. **Role and contribution (~75%):** I co-designed the experimental approach, supervised students involved in data collection, contributed to the statistical analyses, and participated in manuscript preparation. I also contributed to the interpretation of the findings and refinement of the scoring framework. This work was published in *Preventive Veterinary Medicine* (**2026**), where I served as a co-author.

Across these contributions, I developed and led a multidisciplinary research program spanning microbiome, metabolism, nutrition, behavior, and gastrointestinal health. This work resulted in **three scientific publications, 4 oral and 3 poster presentations at international conferences, and the supervision of six students** at the Bachelor's, Master's, and Veterinary Doctorate degrees. I also established research collaborations with **Jean-Charles Martin** to study metabolome and **Mila Salonen** for the development of behavioural assessment tools.

D. RESEARCH FUNDING AND RECOGNITIONS

Best Oral Presentation Award (**400€**), International Scientific Conference on Probiotics, Prebiotics, Gut Microbiota and Health (**IPC2025**), Athens, Greece, 2025.

E. SUPERVISORY AND MENTORSHIP ACTIVITIES

Student Supervision :

Kenza Kezimana - Université Paul Sabatier | 2nd year Master | January - March 2023. Supervised a behavioral research project characterizing behavioral profiles in 2-month-old Golden Retriever puppies exposed to novel situations. Provided training in experimental design, standardized behavioral assessment, video analysis, and exploratory statistics.

Anaïs Deloste - Université Paul Sabatier | 2nd year Bachelor | March - June 2024. Supervised a clinical research project investigating chronic stress during puppy weaning and adoption using hair cortisol as a physiological marker. Provided training in sample preparation, ELISA-based cortisol quantification, statistical analysis, and scientific writing.

Milan Rotter - Université Paul Sabatier | 2nd year Bachelor | March - June 2024. Supervised a longitudinal study investigating the early establishment of the gut microbiota in newborn puppies using high-throughput Illumina sequencing. Guided the student in biological sample processing, microbiota data analysis and interpretation, and scientific reporting.

Lucy Montagnier - École Nationale Vétérinaire de Toulouse | Veterinary Doctorate | 2024–2026
Supervised a longitudinal study within the Toulouse GOLD cohort investigating behavioral development around adoption in 103 Golden Retriever puppies. Guided behavioral assessment, data analysis, and development of early behavioral profiles.

Emma Vellard - École Nationale Vétérinaire de Toulouse | Veterinary Doctorate | 2023–2025
Supervised a longitudinal study of digestive health during weaning in 88 Golden Retriever puppies. Guided fecal scoring, biological sample collection, longitudinal data management, and statistical analysis of gastrointestinal outcomes.

Alizée Amar - École Nationale Vétérinaire de Toulouse | Veterinary Doctorate | 2023–2025
Supervised a longitudinal study of behavioral development from puppyhood to one year using owner-reported questionnaires. Guided questionnaire design, longitudinal data collection, behavioral trait characterization, and statistical analysis.

Reviewing and Evaluation Activities:

June 2026: Served as an invited scientific evaluator for student research presentations at the Quebec Scientific Research Day (Université Laval), evaluating 15 research projects (oral presentations and posters).

F. RESEARCH PUBLICATIONS AND PRESENTATIONS

I. RESEARCH PUBLICATIONS:

1) **Bendahmane, I**; Garrigues, Q; Apper, E; Mugnier, A; Svilar, L; Martin, JC; Chastant, S; Meynadier, A; Mila, H. (**2025**). *Maternal oral supplementation with Saccharomyces boulardii I-1079 during gestation and early lactation impacts the early growth rate and metabolic profile of newborn puppies.* **Frontiers in Nutrition**, Volume 12-2025. (Published, First author). DOI: 10.3389/fnut.2025.1500600

2) Chauvel, C; **Bendahmane, I**; Vellard, E; Lyazrhi, F; Meynadier, A; Mila, H; Grellet, A. (**2026**). *Validation of photographic fecal scoring in puppies*. **Preventive Veterinary Medicine**, Volume 246. (Published, Co-first author). DOI: 10.1016/j.prevetmed.2025.106729
3) **Bendahmane, I**; Salonen, M; Richard, O; Montagnier, L; Kezimana, K; Lyazhri, F; Mila, H; Meynadier, A. (**2026**). *Two distinct temperamental profiles in Golden Retriever puppies one week post weaning: Association with stress, nutrition, impulsivity and fecal quality***. Journal of Applied Animal Behaviour**. (In press, First Author) DOI.org/10.2139/ssrn.5608815

II. RESEARCH PRESENTATIONS:

1) **Bendahmane, I.** (**2023**). *Oral* : *Effect of maternal supplementation with live yeast S. boulardii 1-1079 on the gut microbiota of newborn puppies*. **European Symposium on Animal Reproduction (ESAR)**, Nantes, France.
2) **Bendahmane, I.** (**2024**). *Oral* : *Maternal Oral Supplementation with S. boulardii I-1079 during gestation impacts the newborn puppies' metabolome and growth*. **European Society of Veterinary and Comparative Nutrition (ESVCN)**, Belfast, Ireland.
3) **Bendahmane, I.** (**2024**). *Poster : Evolution of puppy behavior around adoption period: Study on the cohort of 44 Golden Retriever puppies*. **European Conference on Behavioural Biology (ECBB)**, Zurich, Switzerland.
4) **Bendahmane, I**. (**2024**). *Oral* : *Maternal Oral Supplementation with S. boulardii I-1079 during gestation impacts the newborn puppies' metabolome and growth*. **European Veterinary Society For Small Animal Reproduction**, Barcelona, Spain.
5) **Bendahmane, I.** (**2025**). *Oral* : *Association Between Temperament and Fecal Microbiota in Golden Retriever Puppies at One Week Post Weaning*. **International Scientific Conference on Probiotics, Prebiotics, Gut Microbiota and Health (IPC2025)**, Athens, Greece.
6) **Bendahmane, I.** (**2025**). *Poster : Early dynamics of the gut microbiota in Golden Retriever puppies during the first 12 hours of life: A preliminary study*. **International Scientific Conference on Probiotics, Prebiotics, Gut Microbiota and Health (IPC2025)**, Athens, Greece.
7) **Bendahmane, I.** (**2025**). *Poster : Early dynamics of the gut microbiota in Golden Retriever puppies during the first 12 hours of life: A preliminary study*. **European Veterinary Society for Small Animal Reproduction (EVSSAR)**, Porto, Portugal.

G. OTHER RELEVANT INFORMATION

Current MS-focused research development: Manuscript in preparation as co-first author based on my current postdoctoral research in progressive multiple sclerosis, focusing on T-cell–microglia interactions and cellular metabolism. Submission is anticipated for spring 2027.

PhD Thesis published : Le sevrage du chiot, un stress alimentaire et social : rôle du microbiote digestif et programmation nutritionnelle. (**2025**). Université Paul Sabatier.