Résumé Non Technique (RNT)

Titre du projet de recherche (Doit être identique au titre dans la demande de projet)	**Studying the impact of colonic microbial mucus erosion by a dietary fiber-deprived gut microbiome in a mouse model of experimental autoimmune encephalomyelitis**	
But du projet de recherche (Cochez la case correspondante, des sélections multiples sont possibles)	Recherche fondamentale	☒
	Recherche translationnelle et appliquée	☒
	Application pour des fins régulatoires et la production de routine	☐
	Protection de l'environnement dans l'intérêt de la santé et du bien-être des hommes et des animaux	☐
	Préservation des espèces	☐
	Formation supérieure ou formation ayant le but d'obtenir, de préserver et de développer des capacités professionnelles	☐
	Examens forensiques / Requêtes légales	☐
	Conservation des colonies d'animaux génétiquement modifiés, qui ne sont pas utilisés dans d'autres projets de recherche	☐
Bénéfices prévus à travers ce projet de recherche (max. 1500 caractères) Quelles sont les avancées scientifiques qui pourront être tirées de ce projet (concernant l'homme et les animaux) ?	Dysbiosis of the intestinal microbiota can have severe consequences and it is becoming clear that a disturbance of the microbial community might play an important role in the pathogenesis of neurodegenerative diseases such as multiple sclerosis (MS). Little is known about the mechanisms of how a disturbance of the microbial community occurs and which possible impact this can have on the host immune responses. We have recently investigated the effects of a fiber-free (FF) diet in a gnotobiotic mouse model (wild-type Swiss Webster) containing a 14-member synthetic microbial community (SM) showing that mucus-degrading bacteria outcompete fiber-degrading bacteria causing a degradation of the mucus layer in the colon. Our follow-up unpublished research work with C57BL/6 mice harboring a complex microbial	

	community has shown that mucus-degrading bacteria cause an increase in certain immune cells, which are known to be involved in the pathogenesis of MS. Alterations in the human gut microbiota have recently been reported in MS patients. In detail, *Akkermansia muciniphila*, a member of our well-established 14SM model, is increased in different MS patient cohorts and is exacerbating paralysis symptoms when transferred to. The microbial dysbiosis, especially the increase of *A. muciniphila*, together with the changes in the immune cells caused by the FF-diet might contribute to an intensified symptom development in fiber-deprived mice. We have already performed preliminary experiments in collaboration with a research institute in Japan using our 14SM gnotobiotic mouse model and induced experimental autoimmune encephalomyelitis (EAE) in fiber-deprived mice. The promising results of this preliminary experiment demonstrate a higher EAE symptom development in FF mice compared to FR mice. Using the same EAE model, this project aims to investigate if a fiber-deprived diet exacerbates the symptom development in SPF mice with a complex microbial community.		
Dommages causés à travers ce projet de recherche (max. 500 caractères) Quels sont les éventuels effets néfastes attendus sur les animaux ? Qu'arrive-t-il aux animaux à la fin de l'expérience ? Nommez le niveau de sévérité selon le Chapitre III, article 15, 1. de la Directive européenne 2010/63/UE.	This protocol is classified as severe. The induction of EAE can cause weight loss and paralysis in the animals; therefore, frequent surveillance will take place. If the mice have signs of suffering, they will be evaluated and a veterinarian will be consulted. Food and water combined with hydrogel will be provided in a petri dish in order to prevent dehydration. A point system has been created in order to reduce any pain and suffering of the animals. Daily symptom scoring will be performed and actions will be taken in case animals suffer. If the animals present grade four of the scoring sheet, the humane endpoint will be applied and the mouse will be sacrificed.		
Espèce et nombre d'animaux utilisés Cochez la case correspondante et indiquez le nombre. Des sélections multiples sont possibles.	Souris Rats Poissons zébra Autres	☒ ❑ ❑ ❑	n=40 souris C57BL/6 (female) n= ..... n= ..... n= .....
Application du principe des 3 R de Russel et Burch (1959) (Laissez-vous guider par les questions)			

Replacement / Remplacement (max. 500 caractères) Pourquoi le but de ce projet ne peut-il être atteint par d'autres méthodes ? Pourquoi l'expérimentation animale ne peut-elle être remplacée ?	We aim to investigate the impact of dietary fiber deprivation in a complex microbial community on the symptom development in an EAE model. These experiments intend to assess complex physiologic interactions for which a functioning and complete organism is essential. Alternative approaches to animal use are thus not possible. The interplay of diet, microbial community abundance, and EAE symptom development will be evaluated. Our goal for this experiment is, to reproduce the thick and thin mucus layer phenotypes by feeding these mice a FF and a FR diet and investigate the severity of induced EAE in a complex microbial community. This experimental setup is only possible in a living organisms and will help to answer the pressing question if mucus degradation leads to an exacerbation of EAE symptoms.
Reduction / Réduction (max. 500 caractères) Expliquer comment le nombre d'animaux utilisé est réduit au minimum indispensable (biostatistiques) ?	A commonly used model in MS research is the EAE mouse model. We have chosen to use C57BL/6 mice for our experiments because this species is known to be a well-developed model for the investigation of the human immune system. This species is widely used in MS research in the context of EAE induction. In regards to the animal numbers, we have determined 20 animals per group to obtain statistically significant results (40 in total; with different concentration of Pertussis toxin (PTX); 200 ng and 400 ng PTX per animal). This number of animals is needed for the readouts. It is known that some mice could have to be euthanized with regard to the human endpoint due to the development of paralysis.
Refinement /Amélioration (max. 500 caractères) Expliquer les mesures générales mises en œuvre pour minimiser les répercussions négatives sur le bien-être animal? Comment les dommages sont-ils réduits ? Pourquoi utilisez-vous cette espèce précise ?	The procedure of the EAE induction in mice, as well as the symptom development, may cause pain or suffering in the animals. The suffering of the mice will, therefore, be reduced to a minimal level in order to maintain the well being. The occurring symptoms during the development of EAE are generally considered as severe. The mice will develop paralysis of the tail and/or hind and front legs. Due to these symptoms, it is important to assure the well-being of the animals. Food and water combined with hydrogel will be permanently offered to the animals (provided at the bottom of the cage). The animals will be evaluated daily and the

	symptoms will be scored according to an EAE scoring system which will help us to determine potential suffering of the mice. In case of severe suffering, the animal will immediately be euthanized.