L'assunzione di *Bifidobatteri* migliora la plasticità sinaptica nell'ippocampo e le performances cognitive: studio multidisciplinare su un modello animale

Dott. Giuseppe Talani
Consiglio Nazionale delle Ricerche

Microbiota and its many functions

What about brain functions?

- ✓ Neural pathways
- ✓ Enteroendocrine signaling
- ✓ Serotonin and tryptophan metabolism
- ✓ Immune signaling

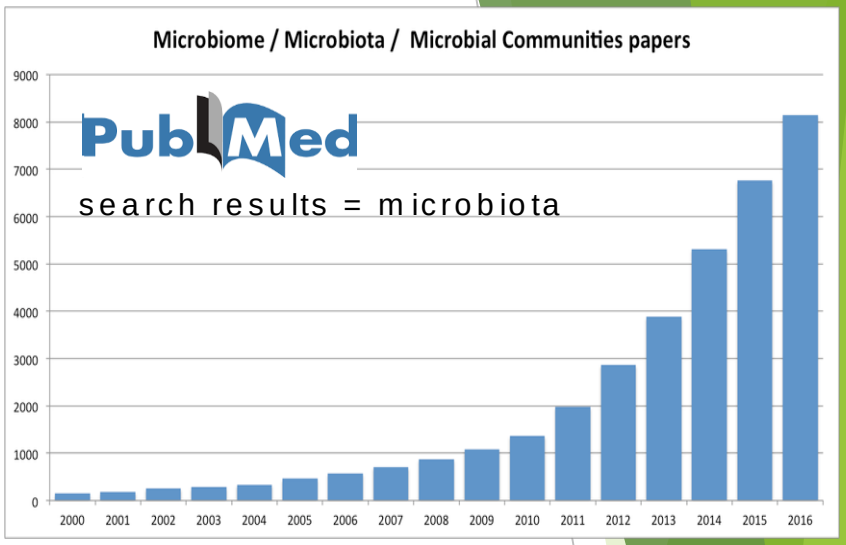
J Physiol 592.14 (2014) pp 2981–2988

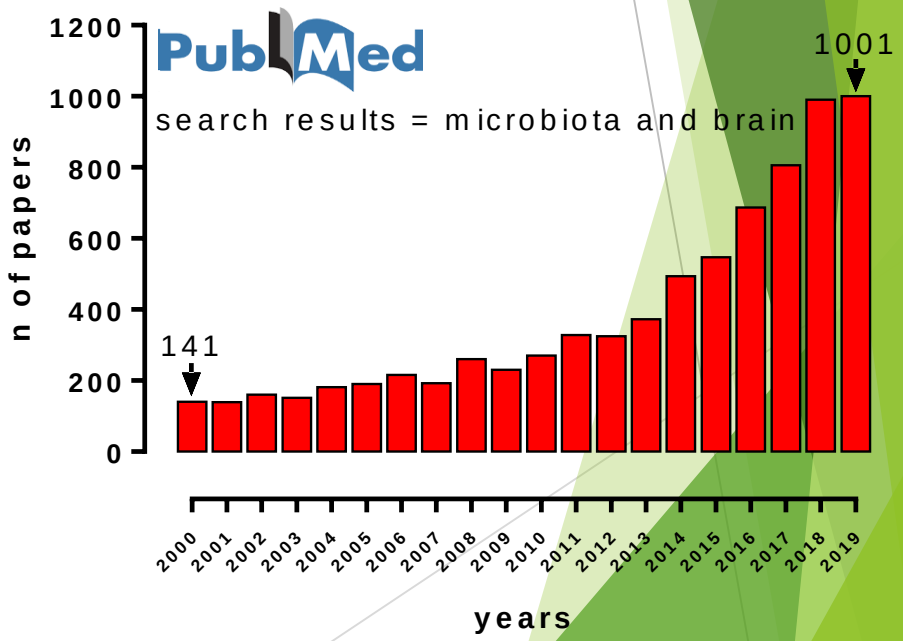
REVIEW ARTICLE

It's a gut feeling: How the gut microbiota affects the state of mind

Adam D. Farmer, Holly A. Randall and Qasim Aziz

Microbiota and brain: the great revoluzion





Dott. G. Talani: L'assunzione di Bifidobatteri migliora la plasticità sinaptica nell'ippocampo e le performances cognitive: studio multidisciplinare su un modello animale

Stress & gut-brain axis: regulation by the microbiome

Foster et al., 2017 Neurobiology of stress

	Behavioural outcomes	Physiological outcomes	References
Clinical evidence			
B-GOS	Increased cognitive processing of positive versus negative attentional vigilance	Reduced cortisol awakening response	(Schmidt et al., 2015)
Lactobacillus casei strain Shirota	Reduced anxiety scores in patients with chronic fatigue syndrome	Increased numbers of *Lactobacillus* and *Bifidobacterium* in faecal samples	(Rao et al., 2009)
	Improved mood in individuals with a low mood prior to taking the probiotic	NA	(Benton et al., 2007)
Probiotic formulation: *Lactobacillus helveticus* and *Bifidobacterium longum*	Reduced psychological distress as measured by the HADS	Reduced 24-h UFC levels	(Messaoudi et al., 2011a)
Multispecies probiotic formulation: *Lactobacillus* and *Bifidobacterium* species	Reduced cognitive processing of sad mood; decreased aggressive feelings and rumination	NA	(Steenbergen et al., 2015)
Preclinical evidence			
Prebiotic- FOS and GOS	Antidepressant and anxiolytic-like effects in adult mice. Reversed the behavioural effects of chronic psychosocial stress in mice.	Increased BDNF, NR1 and NR2A mRNA, and protein expression in the dentate gyrus and frontal cortex Reduced acute and chronic stress-induced corticosterone release. Modified specific gene expression in the hippocampus and hypothalamus. Reduced chronic stress-induced elevations in pro-inflammatory cytokines levels	(Savignac et al., 2013; Burokas et al., 2017)
Prebiotic- 3′Sialyllactose and 6'sialyllactose	Anxiolytic effect in mice exposed to SDR	Prevented SDR-mediated reduction in the number of immature neurons	(Tarr et al., 2015)
Prebiotic- GOS & polydextrose with lactoferrin (Lf) and milk fat globule membrane	Reduced immobility time of maternally separated rats in a forced swim test	Improves NREM Sleep, Enhance REM Sleep Rebound and Attenuate the Stress-Induced Decrease in Diurnal Temperature Attenuated exaggerated IL-6 response in maternally separated rats following concanavalin A stimulation	(Thompson et al., 2016) (Desbonnet et al., 2010)
Bifidobacterium infantis			
Bifidobacterium breve	Improved depressive and anxiety-related behaviours in mice	No effect upon circulating corticosterone	(Savignac et al., 2014)
Bifidobacterium longum	Anxiolytic effect in step-down inhibitory avoidance	Anxiolytic effect mediated via the vagus nerve	(Bercik et al., 2011b)
Lactobacillus plantarum PS128	Reduced immobility time and increased sucrose preference in ELS mice	Decreased basal and stress-induced circulating corticosterone levels; attenuated circulating TNF-α and IL-6 levels while increasing IL-10 levels in ELS mice	(Liu et al., 2016b)
Lactobacillus rhamnosus	Reduced immobility time in the forced swim test Decreased stress-induced anxiety-like behaviour	Decreased stress-induced circulating corticosterone secretion and altered central GABA receptor subunit expression Attenuated chronic stress-related activation of dendritic cells while increasing IL-10 + regulatory T cells	(Bravo et al., 2011) (Bharwani et al., 2017)
Lactobacillus fermentum NS9	Reduced ampicillin-induced anxiety behaviour	Decreased ampicillin-induced corticosterone secretion and increased hippocampal mineralocorticoid receptor and NMDA receptor levels	(Wang et al., 2015)
Butyric acid	Reduced immobility time in Flinders sensitive line rats exposed to a forced swim test	Increased BDNF expression within the prefrontal cortex	(Wei et al., 2014)

The gut-microbiota-brain axis

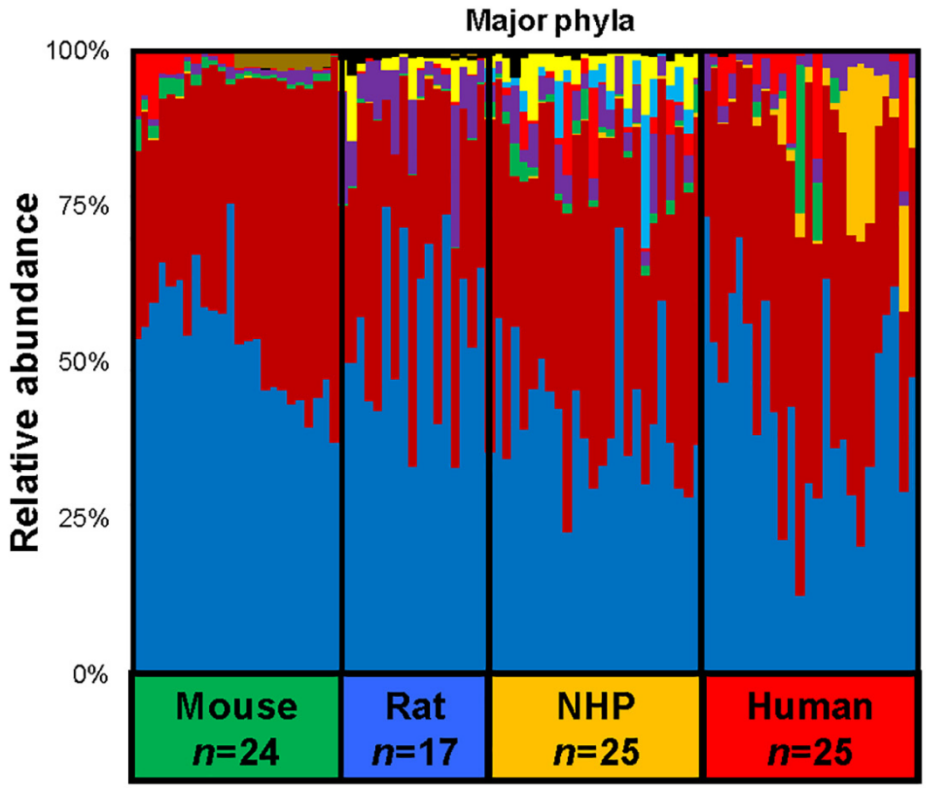
Studies in animal models:
gut microbiome involved:

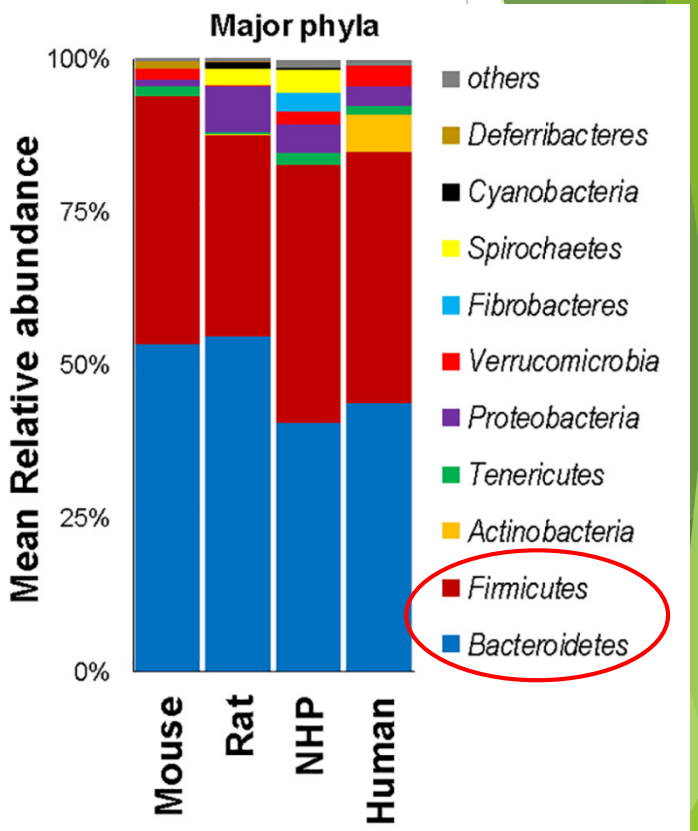
- brain development
- stress sensitivity
- Depression
- Anxiety
- Autism
- Stroke
- Parkinson
- Alzheimer desease

Crayan and Dinan, 2012

Comparative Microbiome Signatures and Short-Chain Fatty Acids in Mouse, Rat, Non-human Primate, and Human Feces

Nagpal et al., Frontier in Microbiology 2018





REVIEW SUMMARY

Science 2019 AAAS

MICROBIOLOGY

Microbiota and the social brain

Eoin Sherwin, Seth R. Bordenstein, John L. Quinn, Timothy G. Dinan, John F. Cryan*

Supplementation of live bacteria (*Bifidobacteria and Lactobacillus*) can lead to natable improvements in social behavior both in early life and adulthood

Microbiota signaling

- Immune activation
- Production microbial peptides
- Acivation vagus nerve
- Production various neurotransmitters

Microbiota and neurotransmitter

Probiotics function mechanistically as delivery vehicles for neuroactive compounds: Microbial endocrinology in the design and use of probiotics

Mark Lyte , 2011

Genus	Neurochemical
Lactobacillus, *Bifidobacterium*	GABA
Escherichia, *Bacillus*, *Saccharomyces*	Norepinephrine
Candida, *Streptococcus*, *Escherichia*, *Enterococcus*	Serotonin
Bacillus, *Serratia*	Dopamine
Lactobacillus	Acetylcholine

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Microbiota and GABA

PNAS

Ingestion of *Lactobacillus* strain regulates emotional behavior and central GABA receptor expression in a mouse via the vagus nerve

2011

Javier A. Bravo[a,1], Paul Forsythe[b,c,1], Marianne V. Chew[b], Emily Escaravage[b], Hélène M. Savignac[a,d], Timothy G. Dinan[a,e], John Bienenstock[b,f,2], and John F. Cryan[a,d,g,2]

The Journal of Physiology

Postnatal microbial colonization programs the hypothalamic–pituitary–adrenal system for stress response in mice

Sudo N. et al, 2004

Contents lists available at ScienceDirect

Behavioural Brain Research

journal homepage: www.elsevier.com/locate/bbr

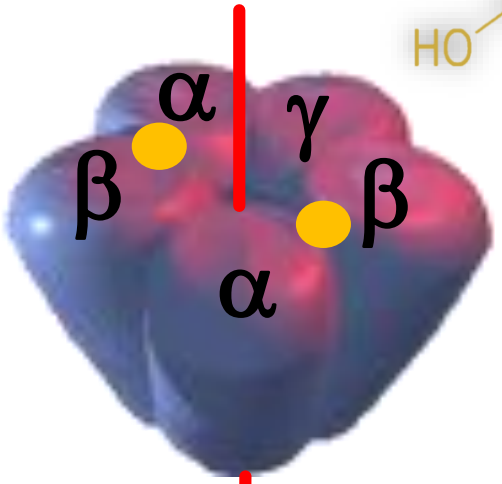
Research report

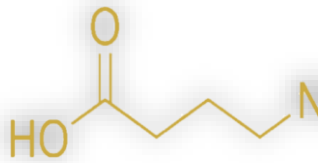
Bifidobacteria modulate cognitive processes in an anxious mouse strain

H.M. Savignac[a,b,1], M. Tramullas[a,2], B. Kiely[c], T.G. Dinan[a,d,**], J.F. Cryan[a,e,*]

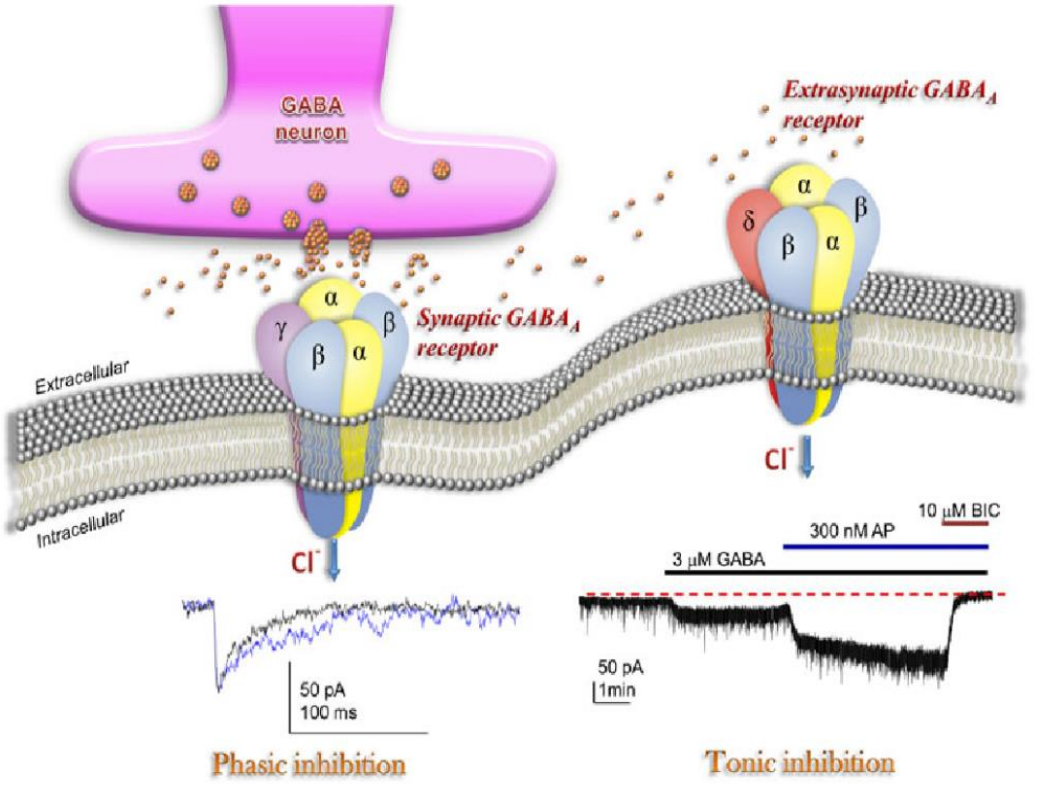
2015

The GABAergic system

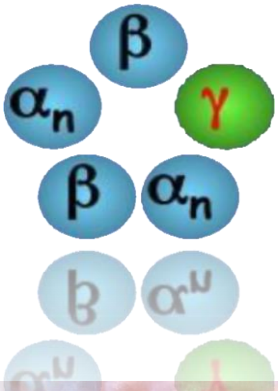


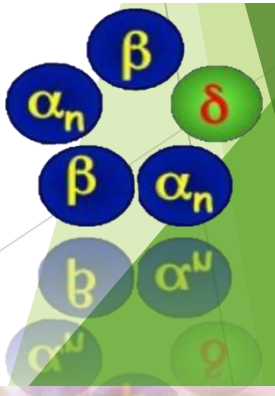


GABA



neuronal inhibition



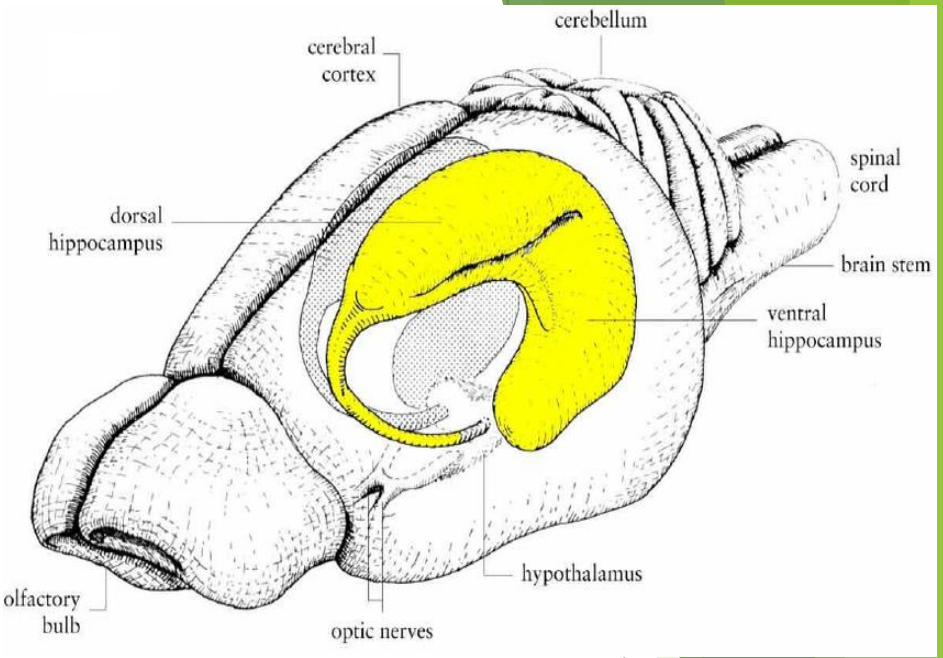


Major Aims

Is the treatment with Bifidobacteria capable to alter GABAARs subunit expression in Sprague Dawley Rats hippocampus

Is long term plasticity and its mechanisms altered by Bifidobacteria treatment?

What kind of behavioral aspects are subjected to modification after the treatment?



Hippocampus

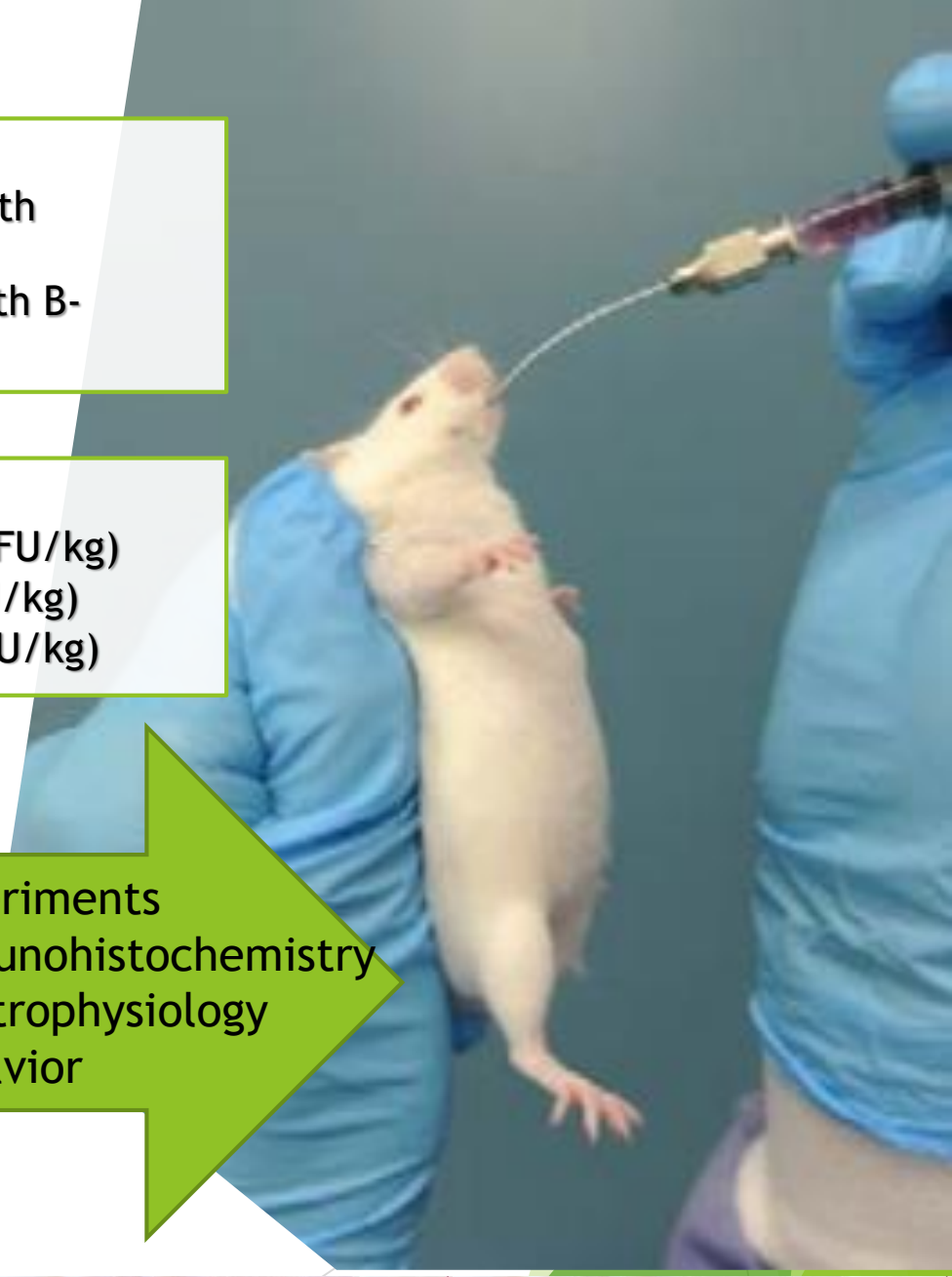
- ✓ Learning
- ✓ Memory
- ✓ Cognition
- ✓ Stress related responses (HPA)

Experimental groups

- Controls: animals treated for 2 moths with vehicle
- Treated: animals treated for 2 month with B-MIX

B-MIX

- Bifidobacterium longum (BB536, $3x10^9$ CFU/kg)
- Bifidobacterium breve (M-16V, $1x10^9$ CFU/kg)
- Bifidobacterium infantis (M-63, $1x10^9$ CFU/kg)



Time table

- Gavage

From PND 45
(daily from 12:00 to 13:00)

- Experiments
- Immunohistochemistry
- Electrophysiology
- Behavior

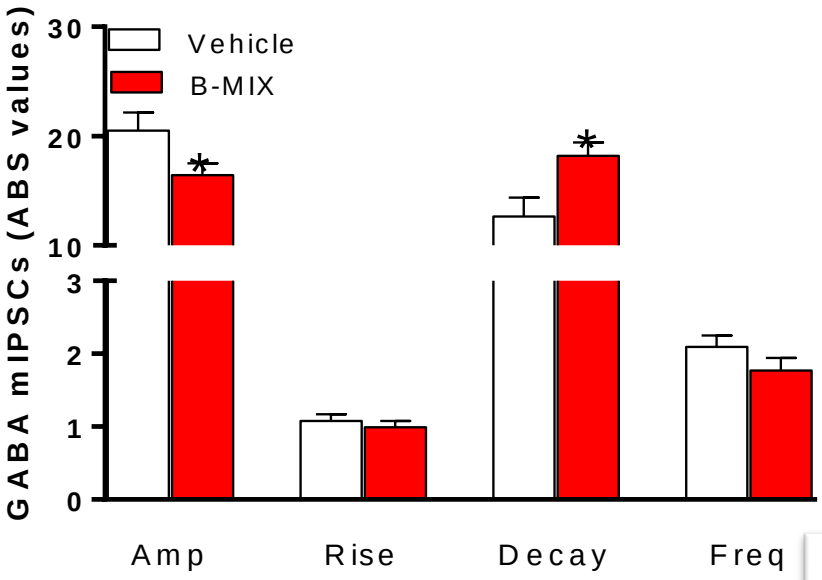
Bifidobacterium inhabiting the human and animal intestinal tract is known for its health-promoting effect.

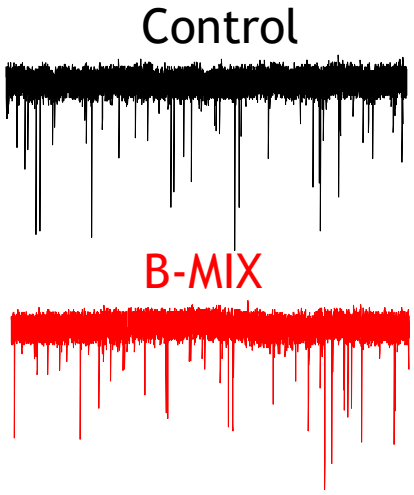
Probiotic Bifidobacterium longum NCC3001 Reduces Depression Scores and Alters Brain Activity: a Pilot Study in Patients With Irritable Bowel Syndrome
Pinto-Sanchez et al., 2017, Gastroenterology

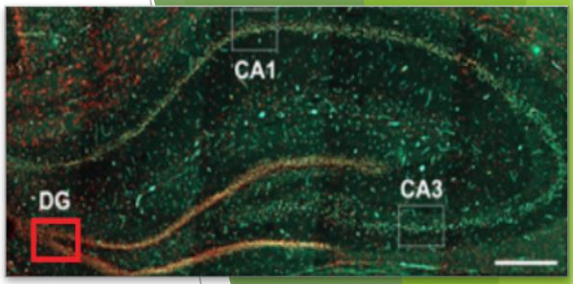
BDNF expression in the hippocampus of maternally separated rats: does Bifidobacterium breve 6330 alter BDNF levels?
O'Sullivan et al., 2011, Benef Microbes

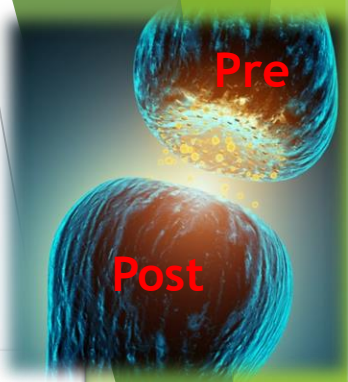
Effects of the probiotic Bifidobacterium infantis in the maternal separation model of depression.
Desbonnet et al., 2010, J. Neurosci

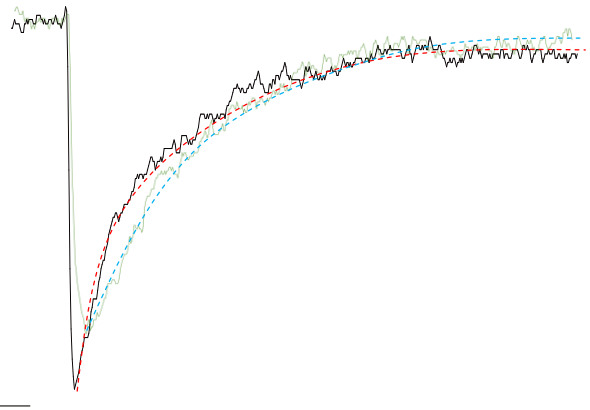
B-MIX treatment alters mIPSC GABAergic currents…



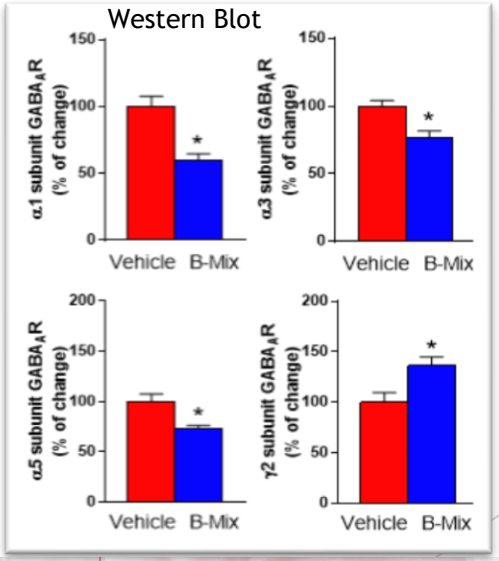


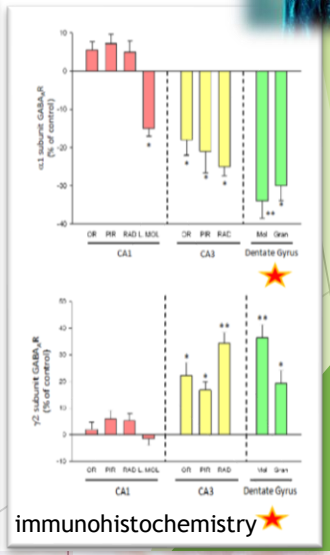






Talani et al. (In preparation)





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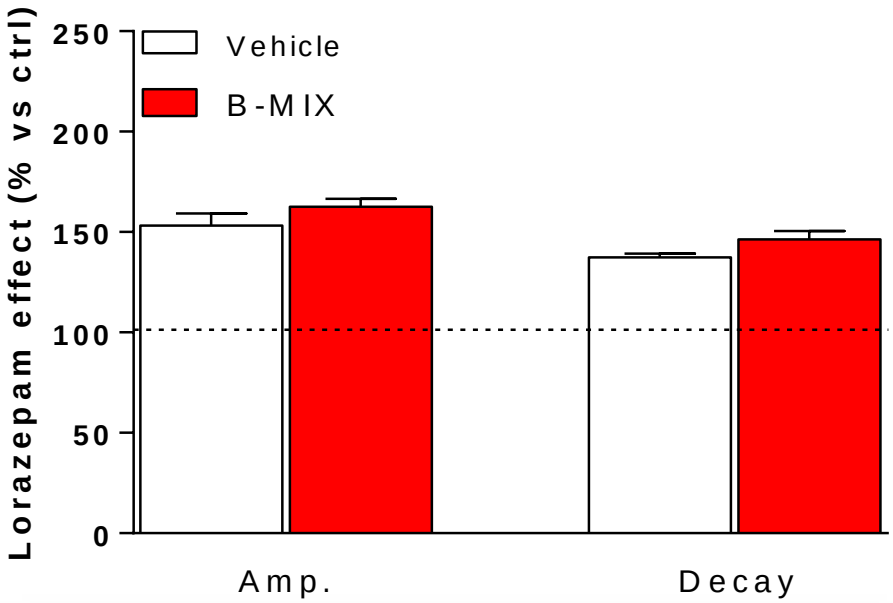
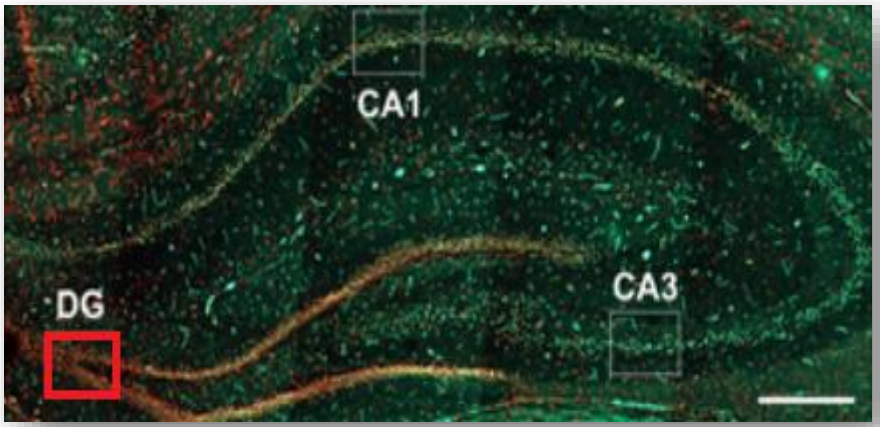
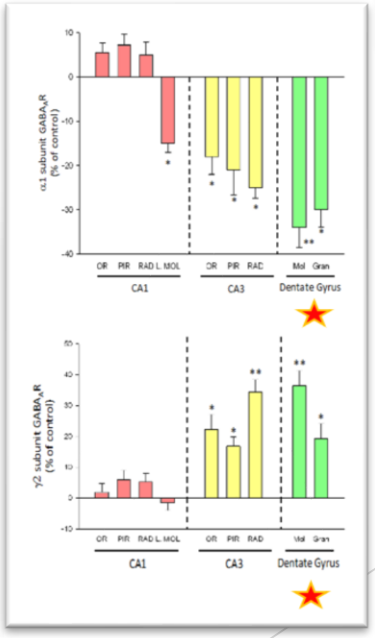
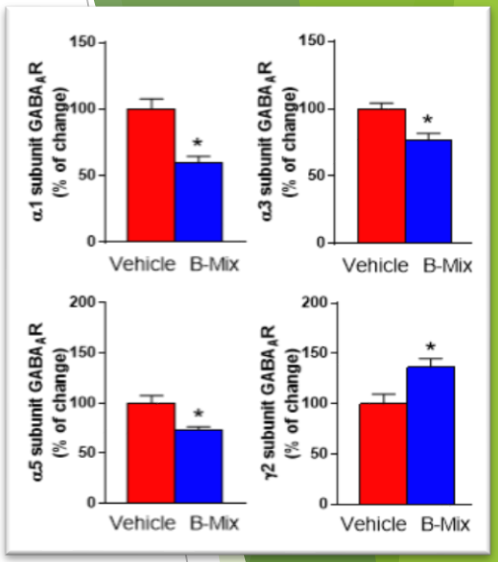
...with no change in benzodiazepine response





immunohistochemistry

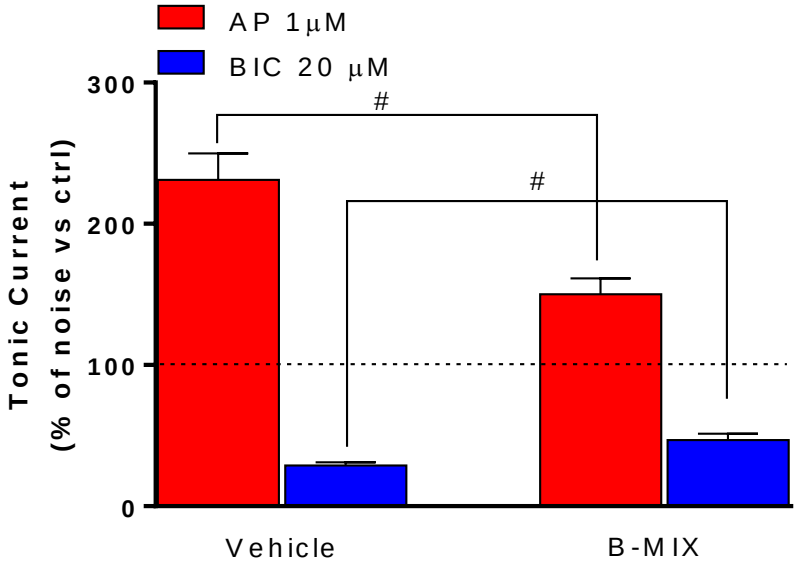




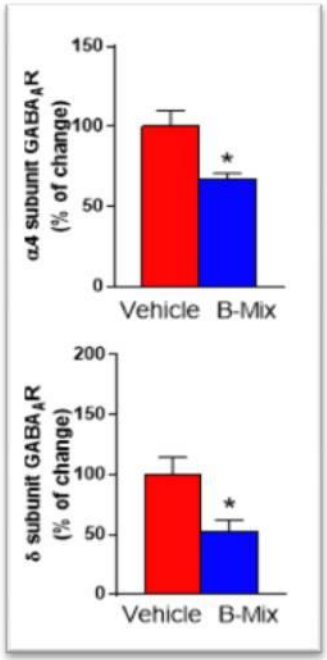
Western Blot

Talani et al. (In preparation)

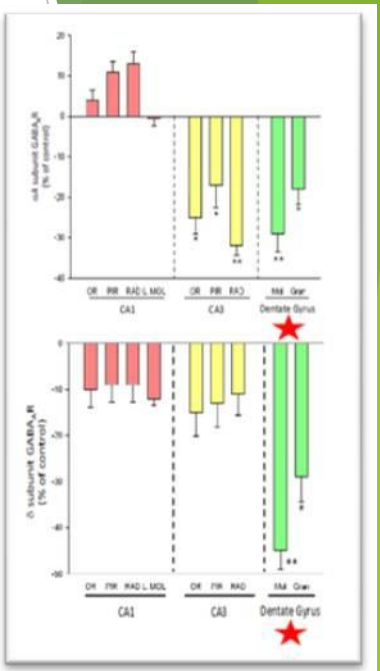
B-MIX treatment decreases the tonic component of GABAergic currents

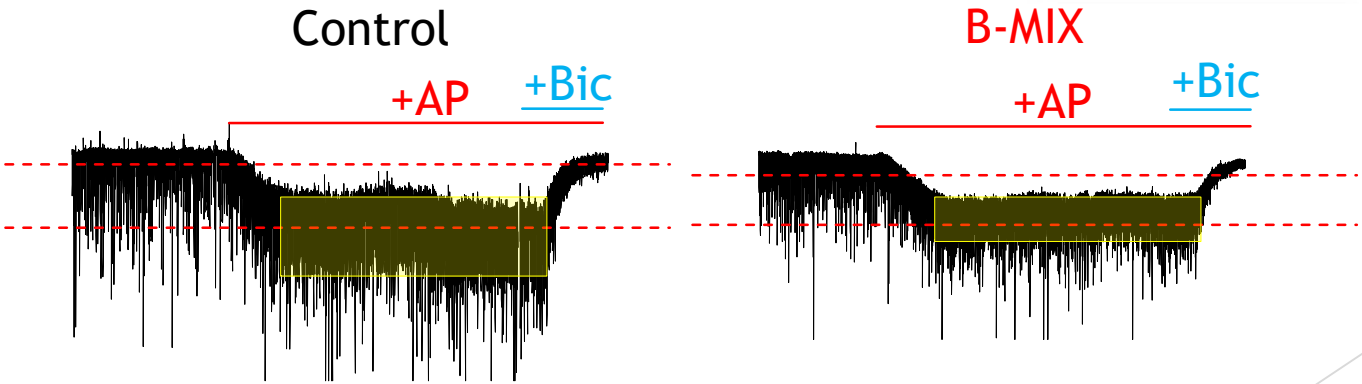


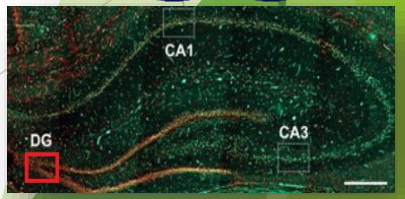
Western Blot



immunohistochemistry

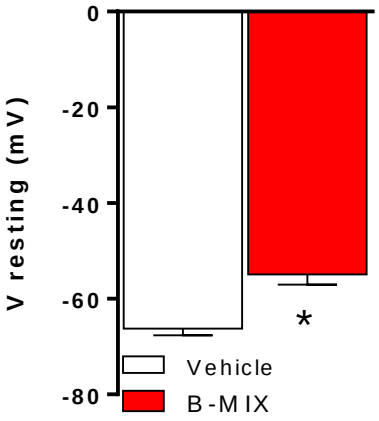


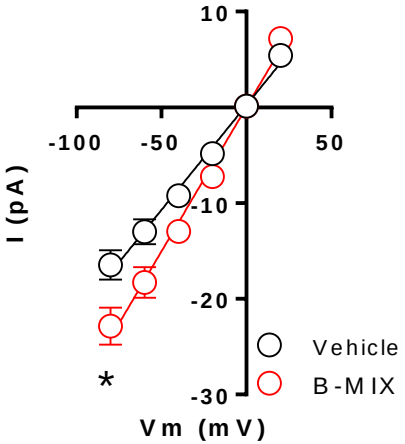


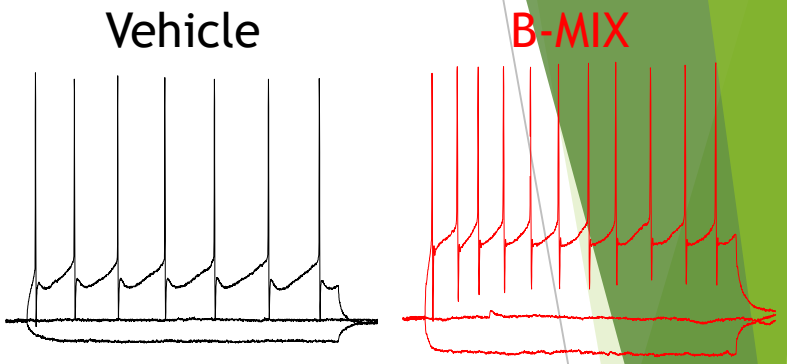


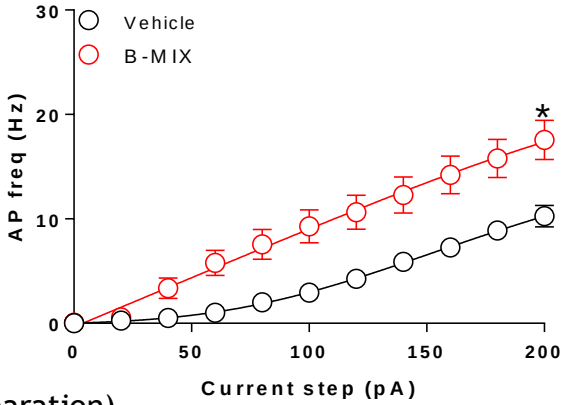
Talani et al. (In preparation)

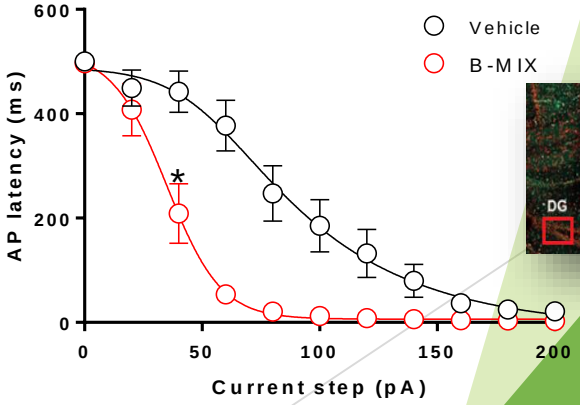
B-MIX treatment enhances the excitability of granule neurons in the hippocampus

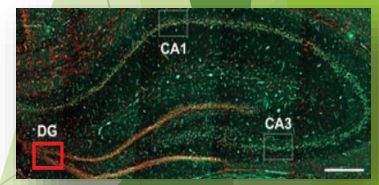






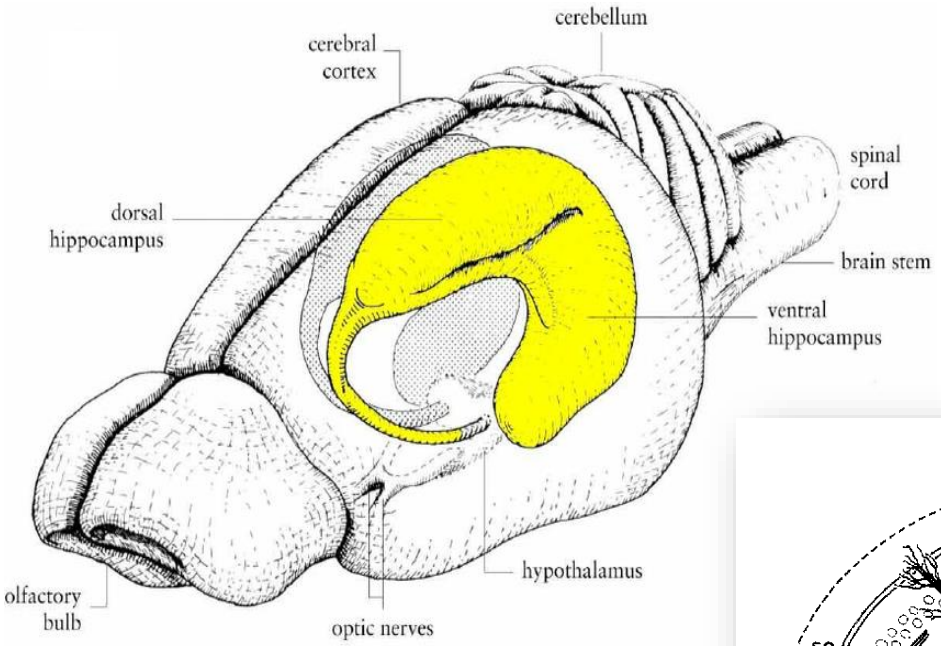


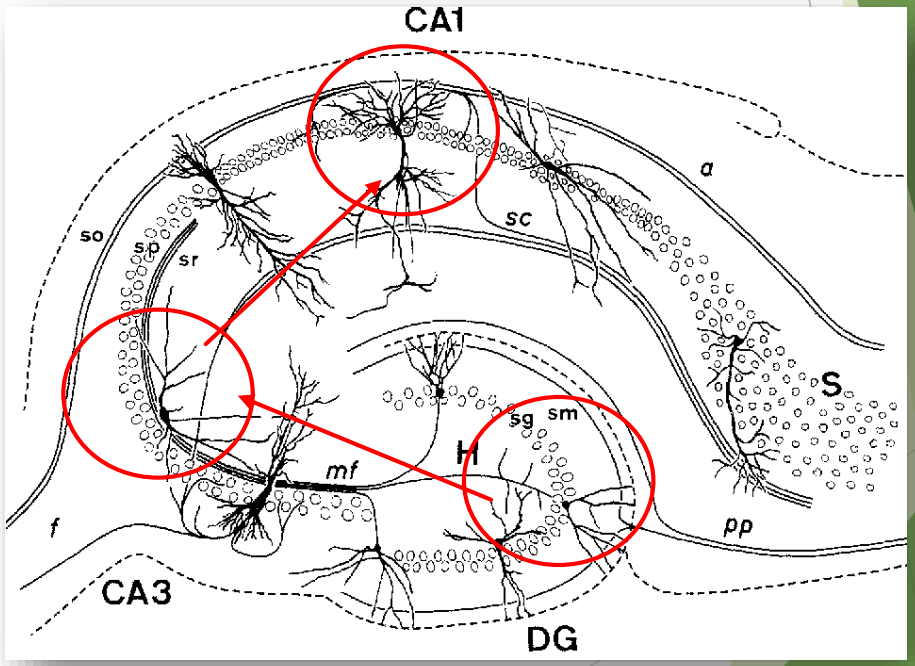




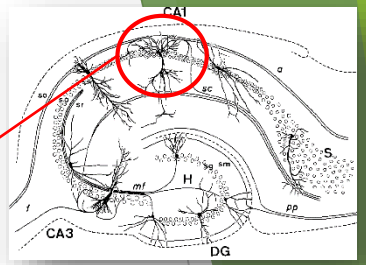
Talani et al. (In preparation)

The trisynaptic nature of the hippocampus



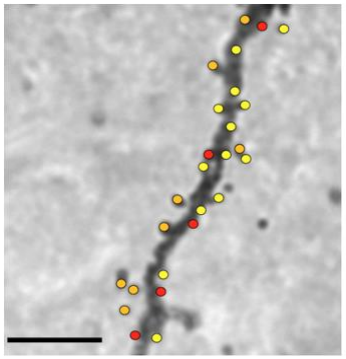


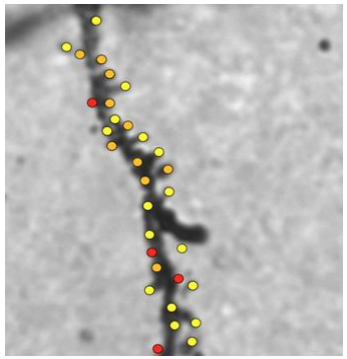
B-MIX treatment increases the density of dendritic spines in rat hippocampal CA1 pyramidal neurons



Vehicle B-MIX

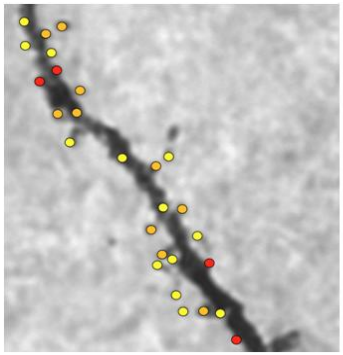
Apical

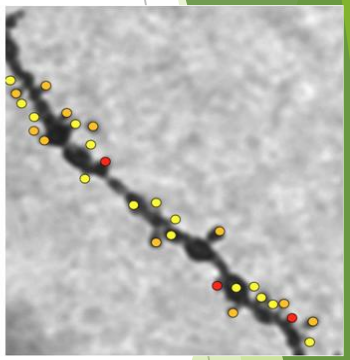


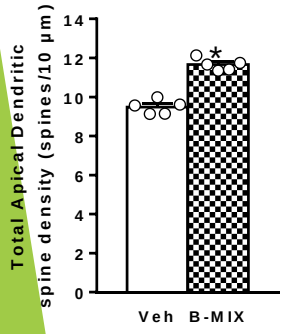


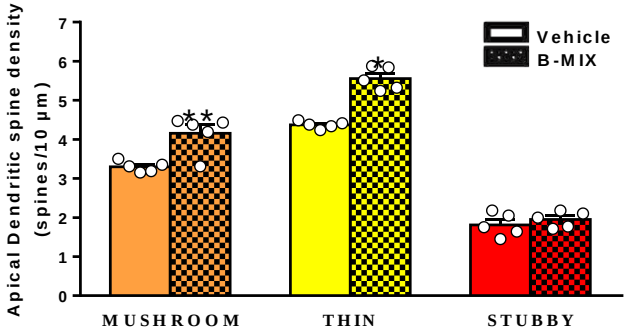
Vehicle B-MIX

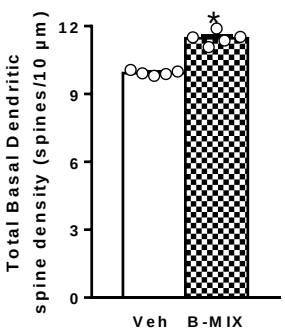
Basal

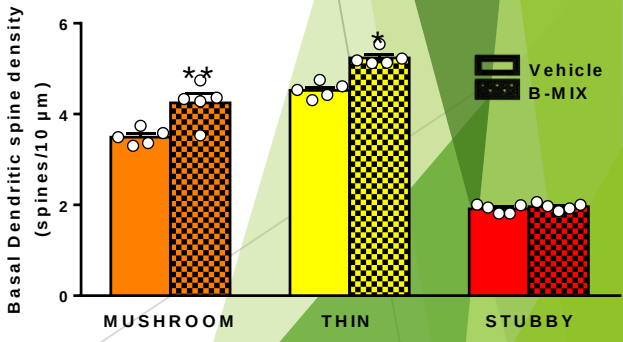






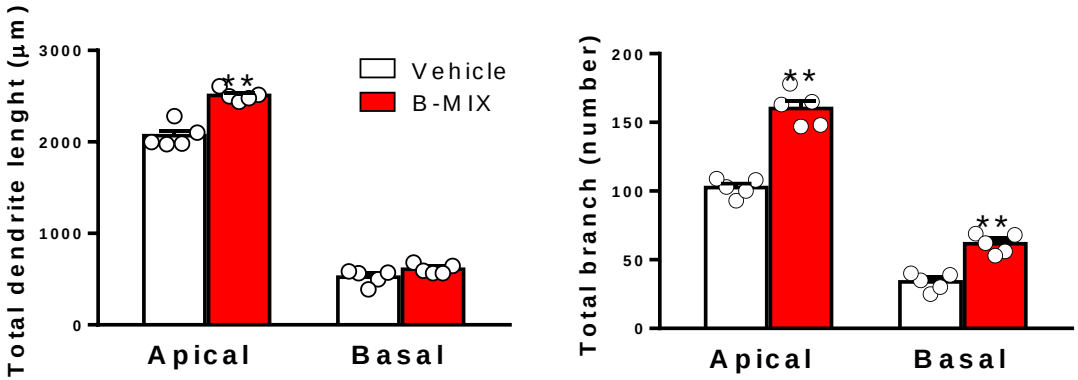


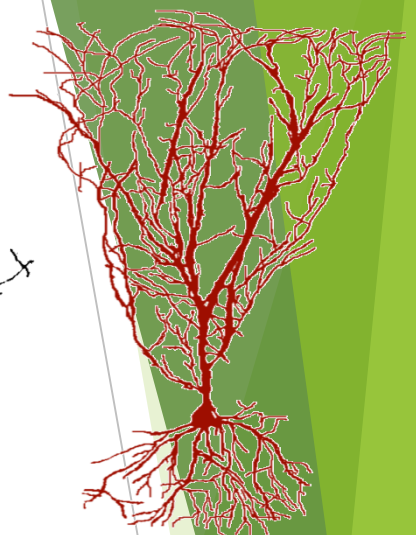




Talani et al. (submitted)

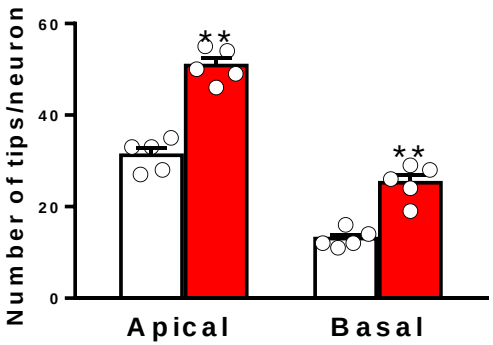
B-MIX treatment increases the dendritic arborization in rat hippocampal CA1 pyramidal neurons

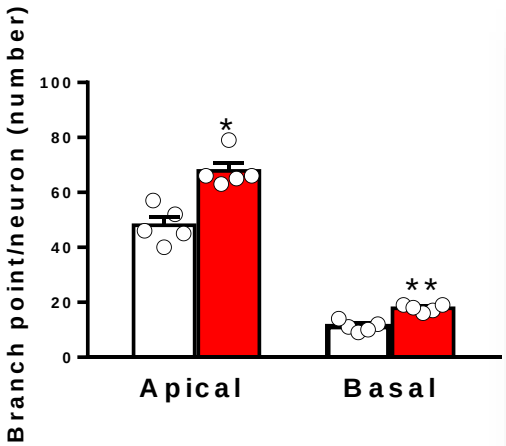


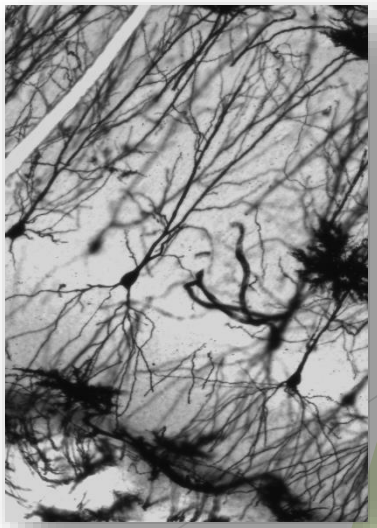


CTRL

B-MIX

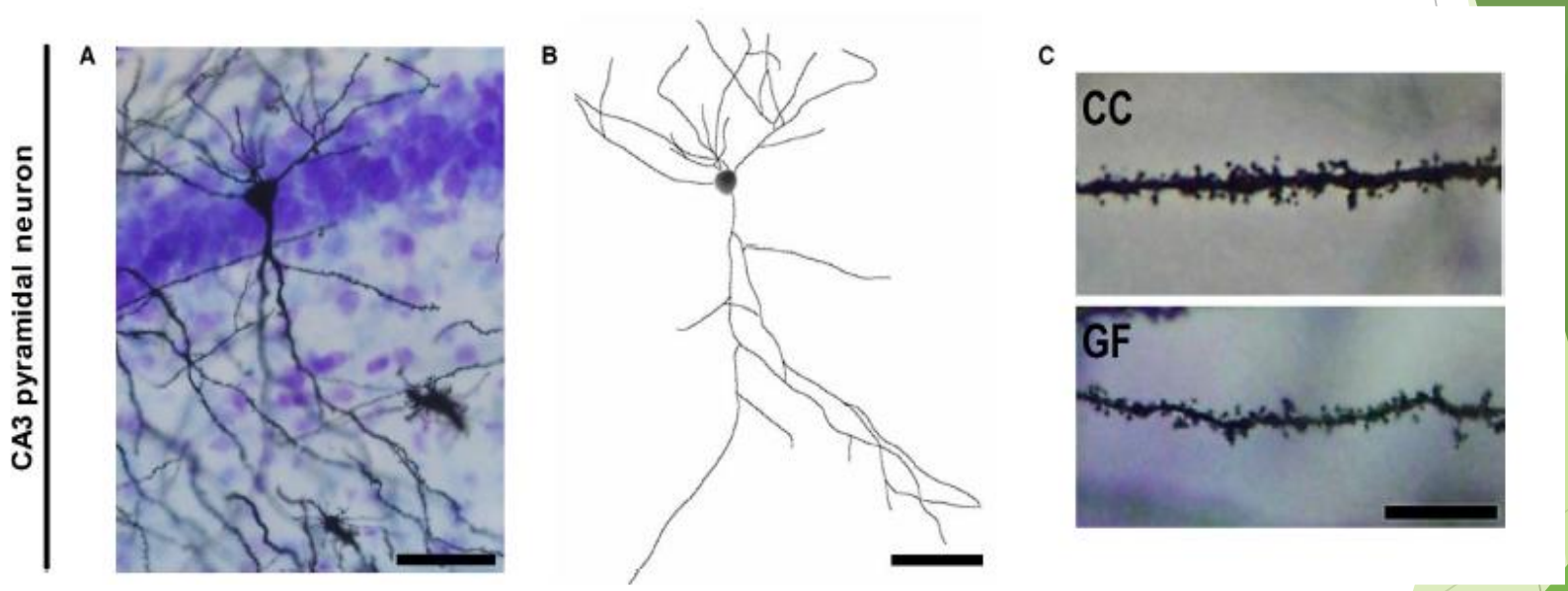




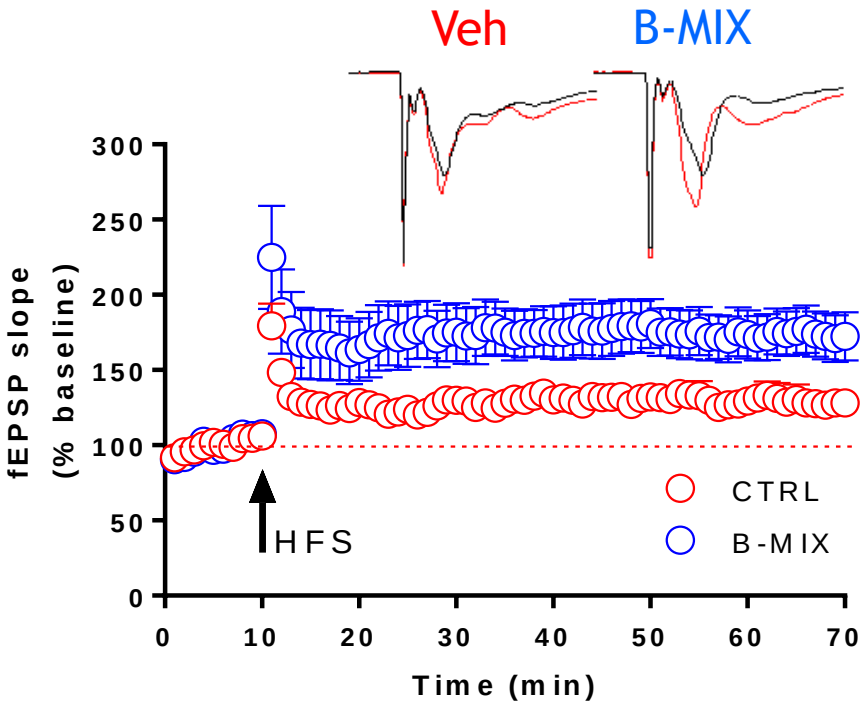


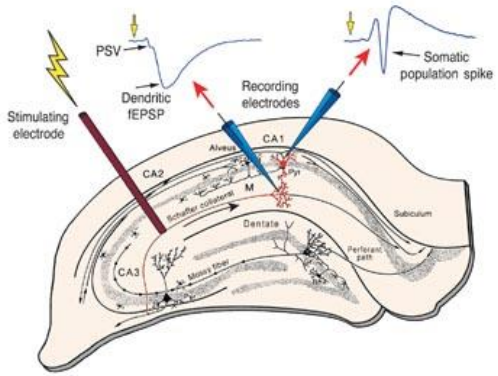
Talani et al. (submitted)

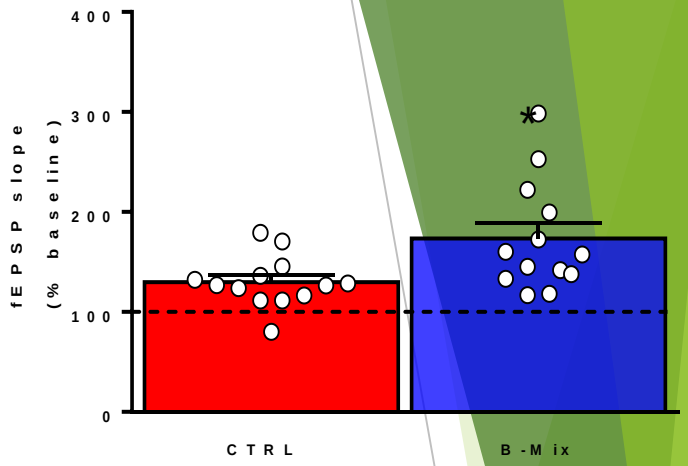
Adult microbiota-deficient mice have distinct dendritic morphological changes: differential effects in the amygdala and hippocampus
Luczynski et al., EJN, 2016

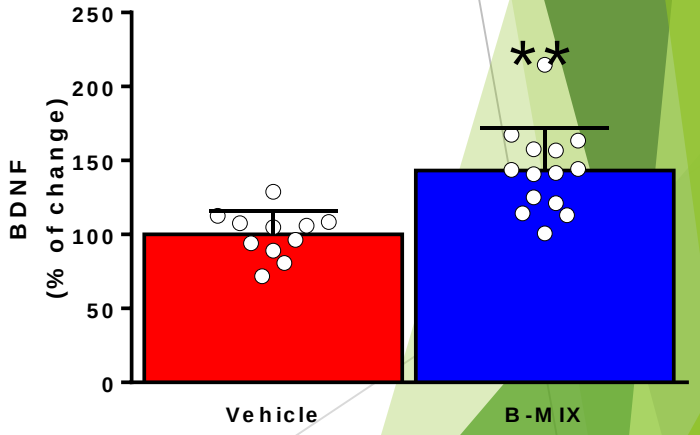


B-MIX improves long-term synaptic plasticity in CA1 pyramidal neurons









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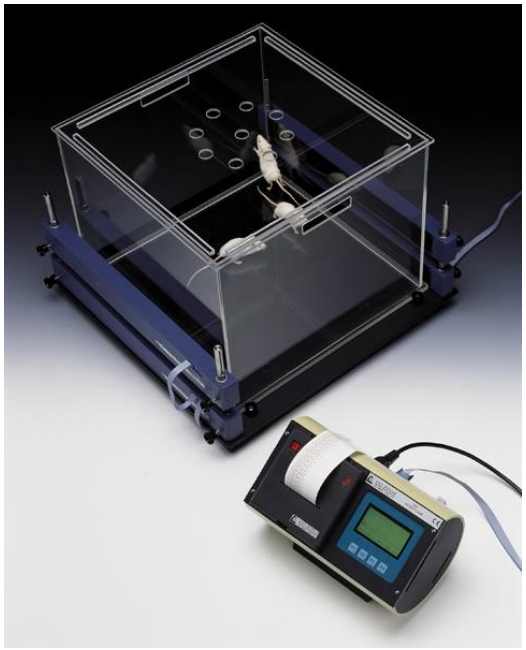
What about behavior?

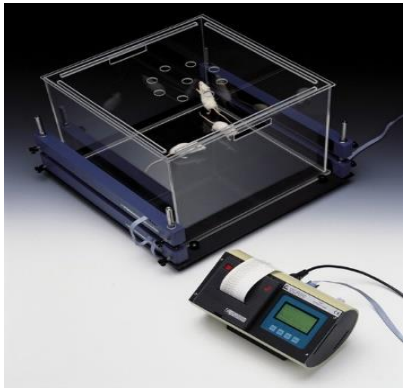


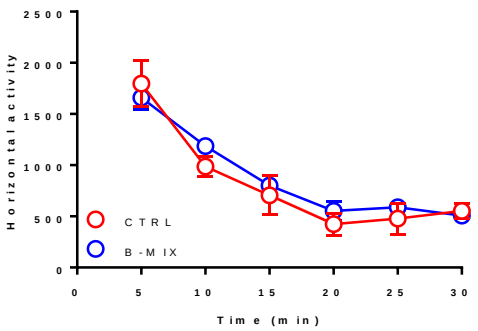
Motility meter
✓ Motor impairments

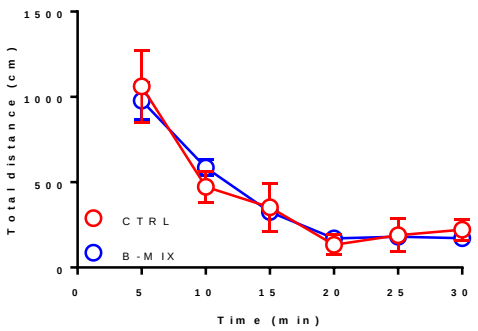
Barnes maze
✓ Memory and cognition impairments

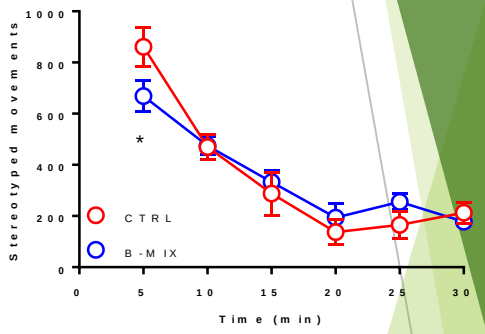
Novel Object Recognition
✓ Memory and cognition impairments

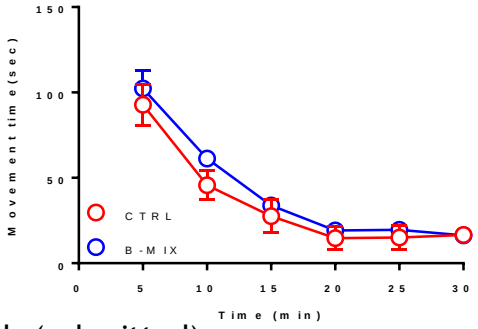
B-MIX treatment fails to alter motor functions

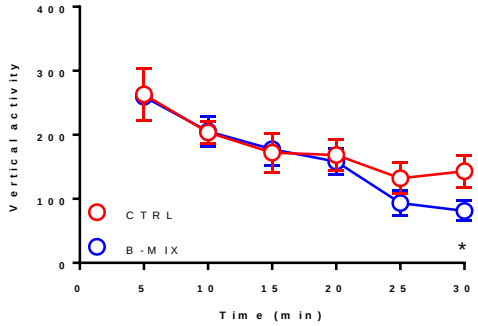


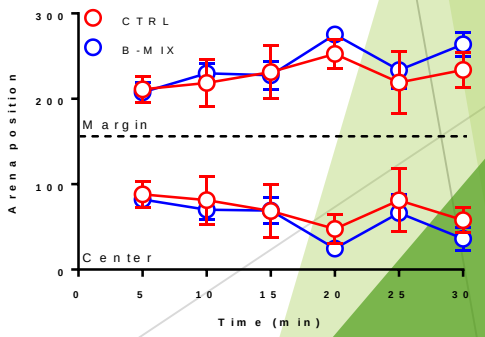












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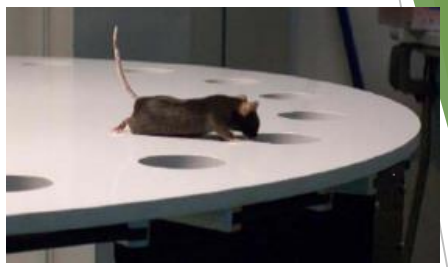
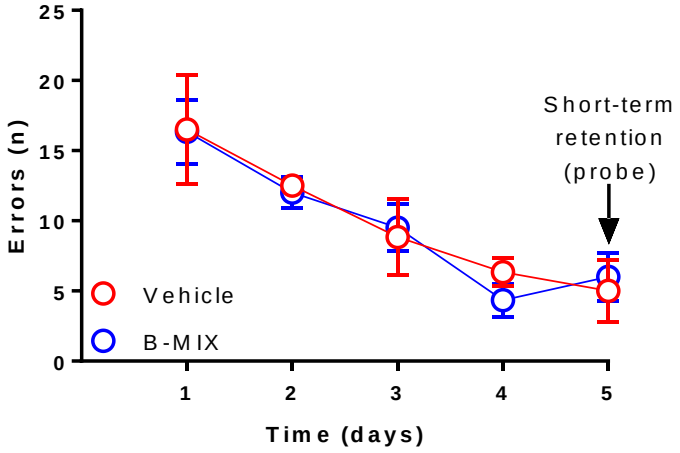
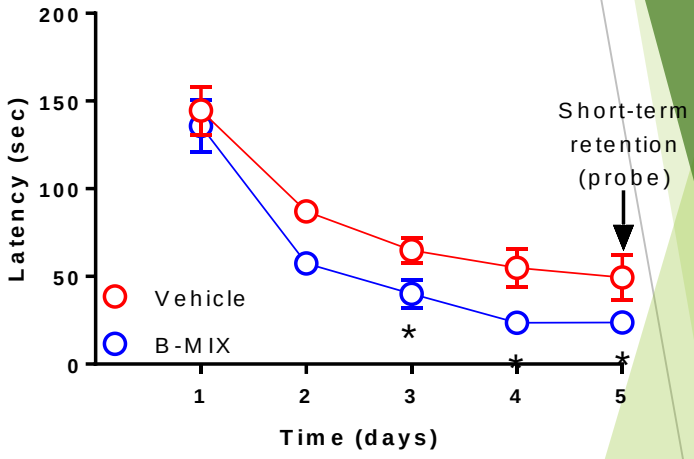
B-MIX treatment improves cognitive behavior







Motivational drive
Multimodal association cortex
Prefrontal cortex

Vs

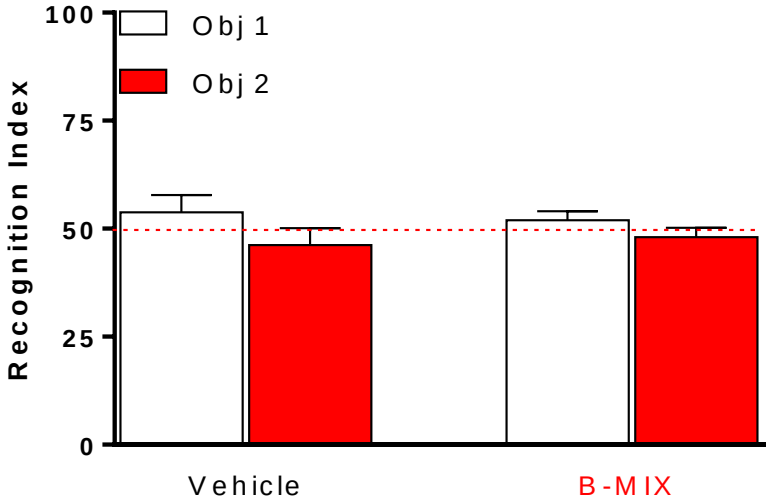
Decision making
Limbic circuits
hippocampus

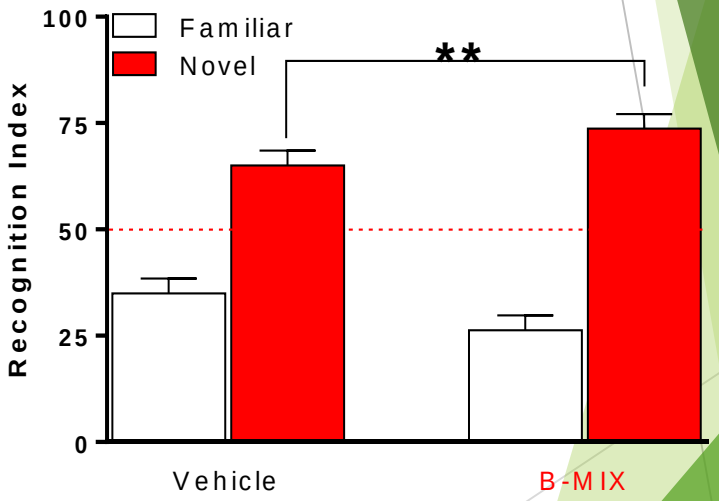
Talani et al. (submitted)

B-MIX treatment improves cognitive behavior

familiarization

test





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Probiotics and Prebiotics as a Therapeutic Strategy to Improve Memory in a Model of Middle-Aged Rats

frontiers in Aging Neuroscience

ORIGINAL RESEARCH
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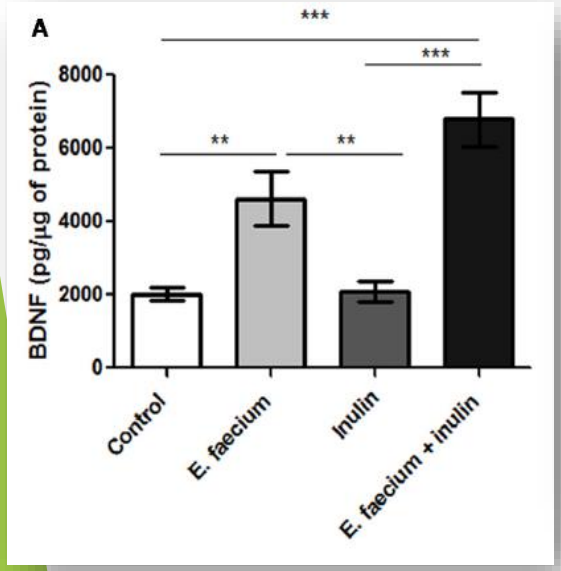
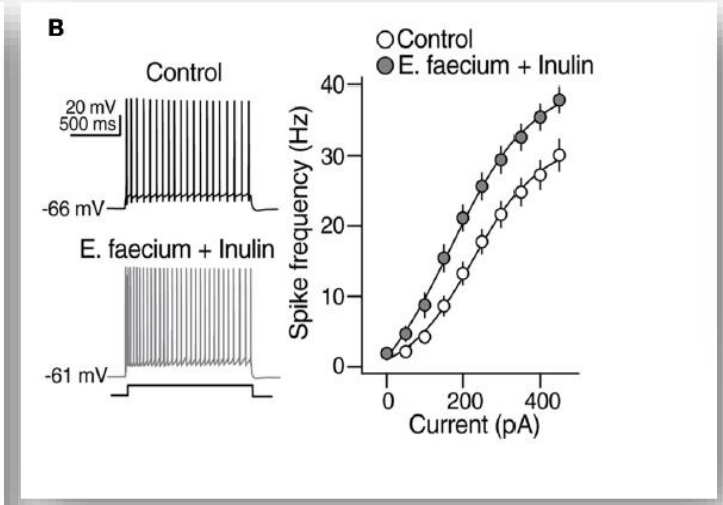
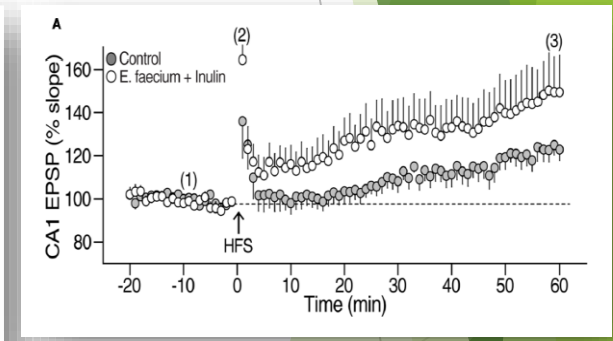
*Alejandra Romo-Araiza[1], Gabriela Gutiérrez-Salmeán[1], Emilio J. Galván[2], Melissa Hernández-Frausto[2], Gabriel Herrera-López[2], Hector Romo-Parra[1], Valentina García-Contreras[1], Ana María Fernández-Presas[3], Ricardo Jasso-Chávez[4], Cesar V. Borlongan[5] and Antonio Ibarra[1]**







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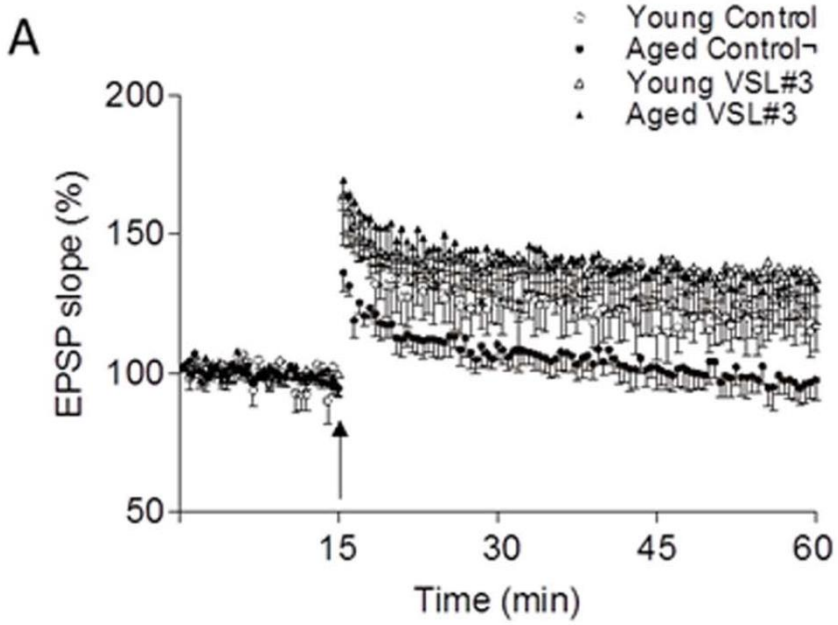
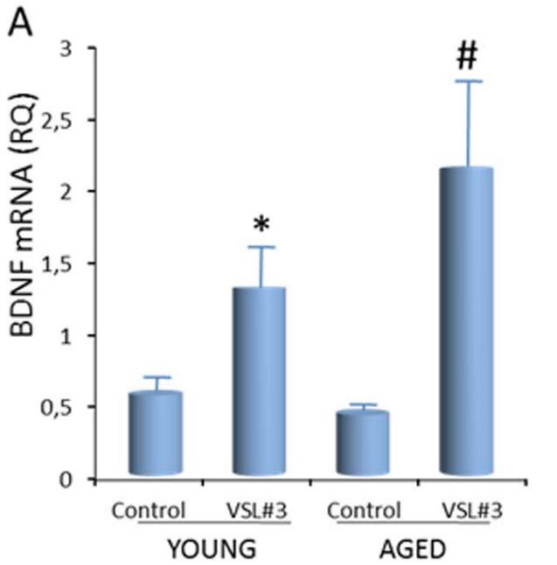
Modulation of Intestinal Microbiota by the Probiotic VSL#3 Resets Brain Gene Expression and Ameliorates the Age-Related Deficit in LTP

Eleonora Distrutti[1]*, Julie-Ann O'Reilly[2], Claire McDonald[2], Sabrina Cipriani[3], Barbara Renga[4], Marina A. Lynch[2], Stefano Fiorucci[4]





Conclusions and.....

Our results, conducted in healthy animals, suggest that the treatment with tree different strain of Bifidobacteria, alters the expression of different subunit of GABAA receptors, enhancing the excitability of granule neurons of the hippocampal formation, the principal output of this important brain area.

The decrease of GABAergic inhibition causes, in turn, an enancement of long term plasticity at glutamatergic synapses in CA1 neurons that represent the principal output of the hippocampal formation.

The increased plasticity observed at hippocampal level is accompained enanced levels of BDNF and dendritic structures in pyramidal CA1 neurons.

All these effects may explain the results obtained in behavioral experiments, in which animals perform better in cognitive tests without any significant modification in motor functions

…….future directions

The mechanisms of action of probiotics at the brain level appear still to be very complex and many efforts are yet to be made to evaluate the properties of different strains of gut bacteria.

Our findings suggest the potential utility of these three Bifidobacteria strains to ameliorate, even in humans, emotional and affective diseases as well as age- and stress associated cognitive decline.

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